Assessing the Epidemiology of Hepatitis C to Inform Public Health Strategies towards Hepatitis C Elimination

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DECLARATION OF WORK

I, Dewi Nur Aisyah, declare that this thesis has been composed solely by myself and that it has not been submitted, in whole or in part, in any previous application for a degree. Except where states otherwise by reference or acknowledgment, the work presented is entirely my own.

ABSTRACT

Background: Major advances in hepatitis C virus (HCV) treatment suggest that HCV might be eliminated in the future. In this thesis, I have undertaken a series of studies (systematic review, secondary analysis of existing cohort study, modelling) to investigate factors that are likely to impact on the feasibility of achieving HCV elimination.

Overarching Aim: To improve our understanding of the epidemiology and natural history of hepatitis C in order to inform public health strategies working towards HCV elimination.

Methods: Systematic review and meta-analysis assessing HCV spontaneous clearance rate and its predictors (Chapters 2&3). Prevalence surveys to assess the burden of and risk factors for HCV in Guernsey (Chapter 4) and in vulnerable populations in London (Chapter 5). Development of a mathematical model to estimate the required scale-up of DAA treatment that would be required to eliminate HCV in PWID in London by 2030 (Chapter 6).

Findings:

- HCV prevalence was high in people screened in homeless centres, a prison and drug treatment centres. Increased case finding is needed in these settings.
- 35% of patients spontaneously clear HCV by 12 months - it may be appropriate to have a year observation before instigating treatment for recently infected patients in low and middle-income countries with low healthcare budgets.
- A wide range of risk factors predict spontaneous clearance. Notably drug users and those with HIV are less likely to spontaneously clear than other groups. Thus, early treatment for high risk groups is recommended for those who are less likely to achieve clearance and pose a higher risk of onward transmission.
- IL28B rs8103142, IL28B rs12979860, and IL28B rs8099917 are important host genetics predictors of clearance.
- Treatment prioritisation with “watch and wait” approach is probably more appropriate to be implemented for developing and less developed countries, where large number of HCV patients were infected by iatrogenic transmission and usually those settings have limited DAAs drugs supply. However, for developed countries such as UK, treatment prioritisation is probably less relevant

as the majority of HCV infection came from PWID and DAA's treatment are available.

- The modelling suggests that elimination of HCV in PWID in London by 2030 would require 46% annual treatment coverage of those infected – this represents a major scale up from current activity. Retreatment of treatment failures lowers the coverage needed to 29.5%. The model is highly sensitive to: SVR (Sustained Virologic Response - suggesting need to support adherence and prevent resistance) and injecting duration (suggesting the need for drug treatment services).

Conclusion:

Hepatitis C elimination would require substantial additional investment to raise treatment coverage and prevent transmission through injecting drug use. My work has identified a number of approaches would could support efforts to achieve this goal.

IMPACT STATEMENT

Enhancing Public Health Strategies towards Hepatitis C Elimination

Hepatitis C is one of major public health concerns. The virus has become recognize as the leading cause worldwide of chronic liver disease. WHO estimated there were around 71 million people who are chronically infected with hepatitis C. Not long ago, hepatitis C elimination seemed implausible, but major advances in HCV treatment bring hope that HCV elimination might be feasible in the near future. These new treatments have high effectiveness and tolerability, but they are expensive which limits their availability in developing and less developed countries. Supervised by Prof. Andrew Hayward and Dr. Laura Shallcross, Dewi Nur Aisyah has undertaken a series of studies to improve the understanding of the epidemiology of hepatitis C in order to inform public health strategies working towards HCV elimination during her PhD at UCL.

An unknown proportion of people who are infected with HCV will spontaneously clear the virus without treatment. Knowledge of which individuals are likely to achieve HCV spontaneous clearance could inform treatment and policy decisions about when and to whom DAA's should be given, enabling prioritization of treatment. Dewi found that 35% of HCV patients will continue to clear HCV until 12 months after infection, suggesting it may be appropriate to have a year observation before instigating treatment for recently infected patients. Furthermore, early treatment for high risk groups such as drug users and those with HIV is recommended for those who are less likely to achieve clearance and pose a higher risk of onward transmission.

The study suggests that treatment prioritization with a "watch and wait" approach is probably more appropriate to be implemented for developing and less developed countries, where large number of HCV patients were infected by iatrogenic transmission and usually those settings have limited DAAs drugs supply. However, for developed countries such as UK, treatment prioritisation is probably less relevant as the majority of HCV infections are in PWID and DAA's treatment costs are more affordable.

Modelling studies conducted provide evidence of the feasibility of future HCV elimination, but this will require strong political commitment and integrated public health intervention strategies at local and national level to achieve considerable scale up of screening and treatment services in vulnerable groups and to address transmission through addressing injecting behaviour. Harm minimisation strategies also play a pivotal role to support the HCV elimination program.

In high income countries, the burden of HCV rests in vulnerable populations and is largely driven by injecting drug use. Focusing efforts on tackling disease in these populations will be critical to reducing incidence. However, accessing these populations is challenging, particularly because many of them have multiple overlapping risk factors. Dewi underlines the needs to engage outreach teams and drug treatment services to support treatment adherence and prevent resistance or reinfection. The aspiration to eliminate HCV is unlikely to be achieved without such integrated services to maximise case detection, address underlying social issues, reduce high risk injecting and maximise adherence.

PUBLICATIONS AND PRESENTATIONS

The following publications arose from section of the thesis:

(1) **DN Aisyah**, L Shallcross, A O' Brien, AJ Hully, A Hayward. Assessing Hepatitis C Spontaneous Clearance and Understanding Associated Factors: A Systematic Review and Meta-Analysis. Journal of Viral Hepatitis. January 2018. DOI https://doi.org/10.1111/jvh.12866.

(2) **DN Aisyah**, L Shallcross, A Hayward, RW Aldridge, S Hemming, S Yates, G Ferenando, L Possas, E Garber, JM Watson, AM Geretti, TD McHugh, M Lipman, A Story. 2018. Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, People Who Inject Drugs and Prisoners in London. May 2018. DOI https://doi.org/10.1111/jvh.12936.

(3) **DN Aisyah**, L Shallcross, A O' Brien, AJ Hully, A Hayward. Host Genetic Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis. *Manuscript submitted for publication in BMC Infectious Disease*.

The following presentations arose from sections of the thesis:

(4) DN Aisyah, L Shallcross, A Hayward, RW Aldridge, S Hemming, S Yates, G Ferenando, L Possas, E Garber, JM Watson, AM Geretti, TD McHugh, M Lipman, A Story. 2017. Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, People Who Inject Drugs and Prisoners in London, UK [meeting abstract]. In: Proceedings of Lancet Public Health Science Conference; 2017 November 24; London, United Kingdom. The Lancet 390:S18. DOI 10.1016/S0140-6736(17)32953-7

(5) DN Aisyah, L Shallcross, A O' Brien, AJ Hully, A Hayward. 2017. Understanding Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis [meeting abstract]. In: Proceedings of International Liver Congress; 2017 April19-23; Amsterdam, Netherland. Journal of Hepatology THU-204. DOI https://doi.org/10.1016/S0168-8278(17)30871-1

(6) DN Aisyah, L Shallcross, A O' Brien, AJ Hully, A Hayward. 2017. Host Genetic Determinants Associated with Hepatitis C Spontaneous Viral Clearance: A Systematic Review and Meta-Analysis [meeting abstract]. In: Proceedings of the 26th Conference of the Asian Pacific Association for the Study of the Liver (APASL); 2017 February 15-19; Shanghai, China.

(7) DN Aisyah, L Shallcross, A O' Brien, AJ Hully, A Hayward. 2016. Understanding Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis [meeting abstract]. In: Proceeding of European Association for the Study of the Liver (EASL) Special Conference; 2016 September 23-24; Paris, France.

(8) DN Aisyah, L Shallcross, A Hayward, RW Aldridge, S Hemming, S Yates, G Ferenando, L Possas, E Garber, JM Watson, AM Geretti, TD McHugh, M Lipman, A Story. 2017. Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, People Who Inject Drugs and Prisoners in London [meeting abstract]. In: Proceedings of European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE); 2017 November 6-8; Stockholm, Sweden. Abstract no. C 20.5.

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Next, I would like to express my sincere gratitude to the UCL for letting me fulfill my dream of being a PhD student in here. I decided to become a PhD student at UCL Infectious Disease Informatics because I do believe I can learn a lot of things from UCL, learning the use of highest technology to solve health problems and gain deeper knowledge on infectious disease epidemiology and public health. I hope the knowledge and experience I got from UCL will enhance my skills in utilising the science in Indonesia. I am very proud to be part of UCL.

My appreciation also extends to Dr. Zisis Kozlakidis, a very supportive colleague at UCL Farr Institute of Health Informatics. With him, I learned how to bridge and develop international collaboration researches between UCL and Indonesian's institutions. I hope we can explore more of UK-Indonesian opportunities for health research collaboration in the future.

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To those who indirectly contributed in this research, your kindness means a lot to me. Thanks for all your encouragement!

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PREFACE

Major developments in hepatitis C drugs provide the opportunity to treat hepatitis C in populations which were previously difficult to treat, making elimination of hepatitis C virus (HCV) a possibility in the future. These new treatments have high effectiveness and tolerability but they are expensive which limits their availability in developing and less developed countries. In order to work towards eliminating HCV there is a need to improve diagnosis and screening for the infection, to ensure that infected individuals are treated effectively and to prevent new infections. Thus, research is required to develop evidence based public health strategies to address these issues. In my thesis, I conduct a series of studies which aim to address some of these questions. My goal is to **improve our understanding of the epidemiology of hepatitis C in order to inform public health strategies working towards HCV elimination**, with a particular focus on hard to reach groups. My thesis is divided into seven chapters.

In **Chapter 1**, I provide background information about HCV, including the recent estimates of hepatitis C prevalence worldwide, viral genotypes distribution, the burden of disease, disease transmission and risk factors for HCV. I also describe the development of hepatitis C new drugs, drug resistance, and challenges associated with eliminating hepatitis C in the future.

An unknown proportion of people who are infected with HCV will spontaneously clear the virus without treatment. Knowledge of which individuals are likely to achieve HCV spontaneous clearance could inform treatment and policy decisions about who to prioritise for HCV treatment. Therefore, in **Chapter 2**, I conduct an extensive systematic review and meta-analysis, estimating the rate of HCV spontaneous viral clearance and its predictors. Predictors examined in this chapter include variables from demographic, clinical, and behavioural factors. The results from this chapter provide insight into when and to whom DAA's should be given, enabling prioritisation of treatment which may be particularly relevant to low-income settings. In such settings, it may be reasonable to delay treatment access in order to allow patients time to spontaneously clear infection, as around one third of patients may spontaneously clear within a year of infection. The results are also relevant to parameterisation of models to inform control strategies.

Besides demographic, clinical, and behavioural factors, I also identified host genetic predictors of HCV spontaneous viral clearance in **Chapter 3**. I presented any host

genetic predictors found from literature review search, including interleukin, HLA-class I A, HLA class-I B, HLA class-I C, HLA-class II, KIR alleles, CCR5 genotype, and any other candidate genes. The results of this study also provide suggestion to which patients should be prioritized for DAA treatment, although widespread use of genetic testing is unlikely to be used in the near future. The results may also be valuable to scientists seeking insight into mechanisms of viral clearance to support treatment and vaccine design.

In **Chapter 4**, I describe the design and preparation of a seroprevalence and transmission study to estimate the true prevalence of hepatitis C in Guernsey and the relative importance of local transmission and importation of infection. The aim was to inform future work around treatment and elimination. As an independent island that is part of the UK, Guernsey provided a unique opportunity to investigate the epidemiology of HCV in a relatively closed population (62,229 people). However unfortunately we were unable to recruit to this study. In this chapter, I describe the design of this study and processes for ethical approvals.

In **Chapter 5**, I estimate the prevalence of hepatitis C among vulnerable populations such as People Who Inject Drugs (PWID), people who are homeless and prisoners who had been recruited to the TB Reach study (a cross-sectional study which enrolled individuals from homeless shelters, drug treatment services, and a prison). I also assessed the risk of HCV infection, risk of spontaneous clearance, risk of infection among non-injectors, and the overlapping risk factors between the three groups. This chapter underlined the importance of developing screening and treatment services tailored to the needs of these socially complex groups.

Finally, to consider the policy implications of working towards HCV elimination, in **Chapter 6**, I investigated the DAAs treatment scale-up that would be required among chronically infected PWID in London in order to achieve the WHO incidence elimination target of 90% reduction in incidence by 2030. To the best of my knowledge, this study is the first that assesses the treatment rate needed in order to achieve WHO incidence elimination target by 2030 in a UK setting. The model suggests the need for a step change in treatment coverage, close attention to ensuring high treatment effectiveness in socially complex groups, the importance of retreating treatment failures and the need to simultaneously invest in drug treatment services. Without these measures elimination targets are highly unlikely to be met.

In **Chapter 7**, I discuss the findings of my thesis in the context of the existing research literature and identify further research areas needed to support the development of HCV elimination strategies.

The **last section** contained supplementary information and appendices, and publications arising from this thesis.

1. Narrative Overview: Global Epidemiology of Hepatitis C

Chapter Summary:

The aim of this chapter is to summarise current knowledge about the global epidemiology of hepatitis C to contextualize my thesis. I provide information on recent estimates of hepatitis C prevalence worldwide, the distribution of viral genotypes, disease burden, transmission and risk factors for HCV. I also summarise recent developments regarding treatment for hepatitis C drugs resistance, and challenges associated with the aim to eliminate hepatitis C in the future.

1.1 Hepatitis C Virus

Hepatitis C Virus (HCV) is a small, hepatotropic virus with approximately 50nm diameter. It is enveloped, positive-sense, and single stranded RNA genome consisting of approximately 9.6 kb nucleotides. HCV is a member of the *Flaviviridae* family and is classified from the genus of *Hepacivirus*. It was firstly discovered from non-A non-B viral hepatitis virus derived from a cDNA clone genome sequences in 1989. Soon after its discovery, researchers learned that the virus had essential nucleotide sequence diversity leading to different genotypes and subtypes.(1) The HCV genotype and/or subtypes are determined particularly based on the 5'-UTR, core, E1, and NS5B regions. To date, at least 7 genotypes and 67 subtypes have been identified worldwide.(2)

1.1.1 HCV Genotypes Distribution

HCV types and subtypes display complex geographic distribution around the globe. A recent study (2015) estimated that genotype 1 is the most predominant HCV type worldwide, accounting for 46.2% of total cases; genotype 3 is the second commonest genotype, accounting for 30.1% of all cases; genotypes 2, 4, and 6 were found in 22.8% of total cases; and genotype 5 accounts for <1% cases.(3) The distribution of most common HCV genotypes worldwide including prevalence estimates are presented in Figure 1.1 and Figure 1.2 respectively.

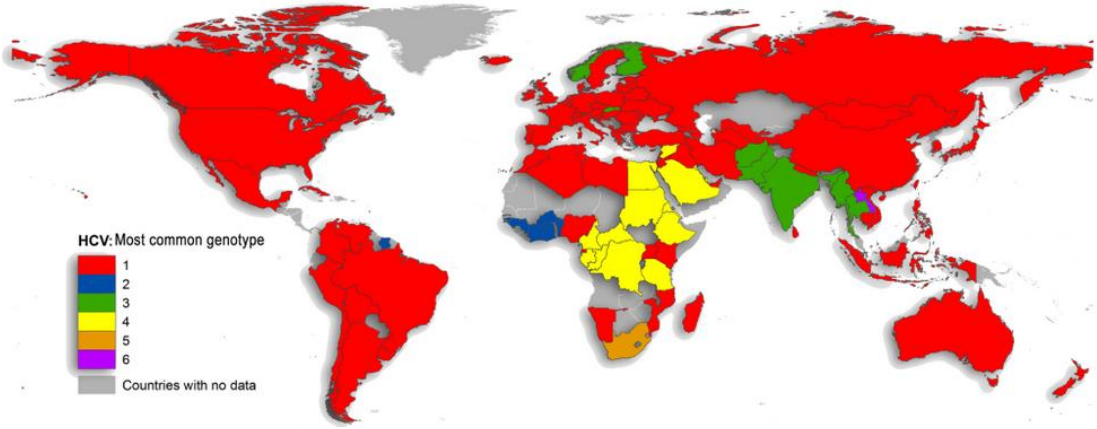


Figure 1.1 Map of Countries with the Majority HCV Genotype (3)

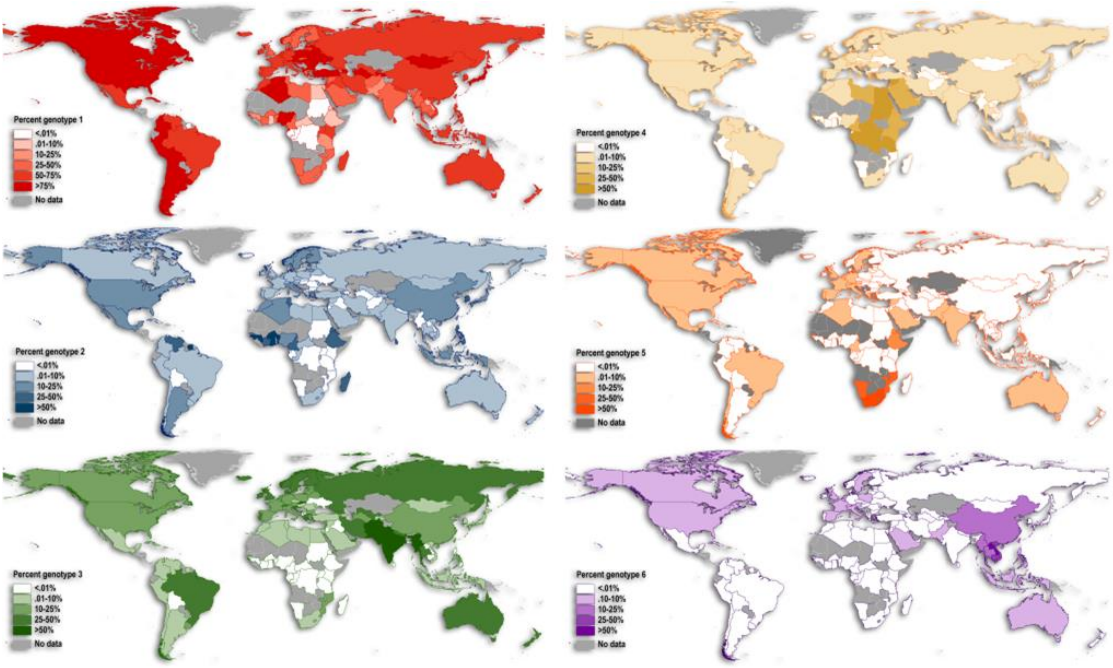


Figure 1.2 Estimate Prevalence of HCV Genotype Worldwide (3)

Messina et.al (3) extracted data from 1217 studies and found that genotype 1 was dominant in North America, genotype 2 was most common in West Africa, genotype 3 was predominated in South Asia and parts of Scandinavia, genotype 4 was prevalent in Central and North Africa, genotype 5 was most commonly found in South Africa, and genotype 6 was most common in Southeast Asia. This study also suggested that genotypes 1 and 3 are dominant in most countries regardless of the countries' economic status, yet the biggest proportion of genotype 4 and 5 infections were reported from low-income countries.

Although in Messina et.al the study had extracted data from numerous publications, there were some limitations of the study. It included publications from 1989 until

2013 which might introduce bias in HCV testing – older studies might have less accurate serological testing techniques. Most of the included studies used convenience sampling which might not representative be of the general population. Also the subtyping techniques were generally based on the HCV RNA 50' UTR region (the accuracy of HCV subtyping can be improved by additional sequencing of other viral regions such as core and NS5B).

Another study conducted in 2014 extracting a total of 2,320 studies also assessed the global distribution of HCV genotypes and found similar patterns of genotype distribution with regard to the dominance of genotype 1 worldwide, followed by genotypes 3, 2 and 4.(4) However, there were some differences in the geographic distribution of different genotypes. For example, genotype 1 was predominantly found in Australasia, Europe, Latin America and North America (53-71% of all cases), genotype 3 was most common in Asia accounting for 40% of all cases, and genotype 4 was prevalent (71%) in North Africa and the Middle East. In the United Kingdom, 90% of HCV infection was caused by genotype 1 and 3, with proportion of 45% and 45% respectively in 2014 (5), and 47% and 44% in 2015 (6).

Gower et.al applied strict criteria to exclude studies in non-representative populations, (e.g. PWIDs, haemophiliacs, minority ethnic groups, refugees, etc.) and only included studies with a sample size of more than 1000. However, it did not include non-English publications. Furthermore, although the study assessed the quality score for each publication included, only 16% studies received a quality score of 3 (good).

1.1.2 Challenges Assessing HCV Genotypes Distribution

The differences in prevalence estimates and geographic distributions reported in the two studies cited reflect the difficulties of obtaining accurate global estimates. Studies assessing HCV genotype distributions are usually derived from confirmed positive laboratory samples because those data are conveniently accessible and represent large sample sizes. However, those samples are very unlikely to be representative of the whole population of individuals with HCV, many of whom are undiagnosed in the community. In addition, individuals who are sampled and tested may be systematically different to individuals who are infected with HCV but have not been tested. For example, vulnerable populations such as intravenous drug users have a high prevalence of HCV but are often not in contact with healthcare services.

Many studies of HCV genotype and prevalence are conducted in specific populations in whom HCV is known to be common, such as PWID or haemophiliacs who were exposed to HCV through contaminated blood products. Those samples may not be representative of all HCV cases as this will depend very much on which groups are routinely tested within the country. For example, if testing is restricted to PWID, then the prevalence of genotype 3 compared to other genotypes (7-9) will appear high. These issues will bias estimates of both genotype distribution and prevalence.

A further challenge is that different studies are likely to employ different methodologies for HCV-testing and genotype screening. Studies may not use comparable diagnostic tests to assess the HCV genotype, especially between studies conducted in high and low-income settings. Furthermore, the quality of trained professional staff, procedures for sample shipment, and available local laboratory equipment might also affect the diagnostic results. Additionally, some countries may have limited access to specific strain typing and may only use genotyping for selected subgroups of patients.

Systematic reviews are often restricted to English-language publications tending to over-represent data from the North America, Oceania and Europe as some studies will have been excluded on the basis of language. This could be partially addressed by including non-English publications and grey literature such as government reports.

1.2 The Burden of Hepatitis C in Population

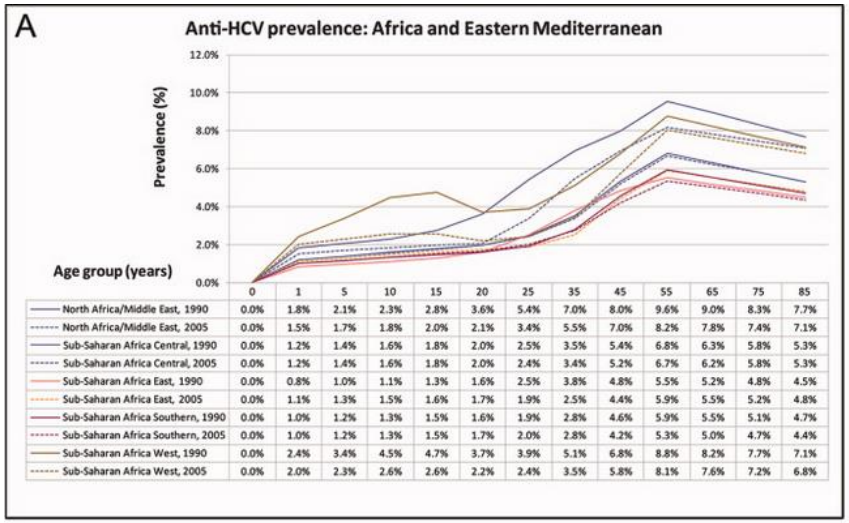
1.2.1 Assessing HCV Prevalence in different Population groups

Ascertaining the incidence of hepatitis C is difficult because most acute cases are asymptomatic. Thus, the available epidemiologic data are mainly based on cross sectional prevalence studies. These can either be general population samples or samples targeting known high-risk groups. HCV screening in “general population” samples (e.g. studies amongst blood donors) exclude high risk groups such as PWID and therefore underestimate risk. Studies based on high risk groups such as transfusion dependent patients, people who inject drugs, individuals with high-risk sexual behaviours can provide good estimates within these groups. However, knowledge of the size of these populations is needed to be able to then use this information to calculate national prevalence rates. Furthermore, the screening tests employed in different countries may have different levels of sensitivity and specificity

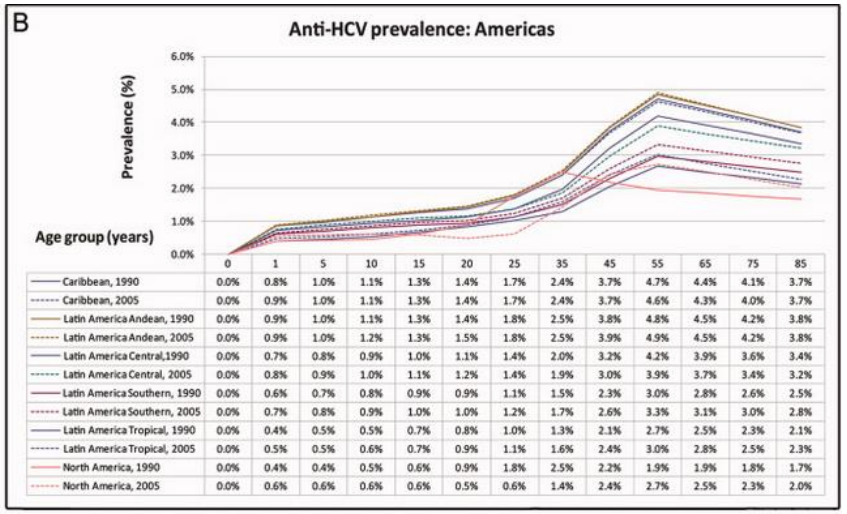
which may confound geographical differences of HCV prevalence. Results usually report HCV positivity based on antibodies rather than HCV RNA. This would lead to overestimation of positivity rates as a proportion will have cleared the virus.

Despite these limitations, WHO and others have attempted to compile studies to gain an overview of global burden of disease and how this varies in different parts of the world. This is important for informing global elimination plans. The most recent worldwide HCV prevalence estimate reported from WHO in 2015 was 1%, representing about 71 million people who are chronically infected with hepatitis C.(10) A recently published systematic review based on 232 studies indicates that this may substantially underestimate the global prevalence of HCV. Estimates from this review suggest that, the age-specific HCV seroprevalence between 1990 and 2005 was higher than that estimated by WHO (see Figure 1.3), indicating that the global prevalence of hepatitis C had increased from 2.3% (95% CI: 2.1%-2.5%) in 1990 to 2.8% (95% CI: 2.3-3.1%) in 2005. This represents an increase in the number of people infected from an estimated >122 million in 1990 to >185 million in 2005.(11) The study also showed an overall increase risk of infection for people age between 20-55 years old, with the peak at 55 years old.

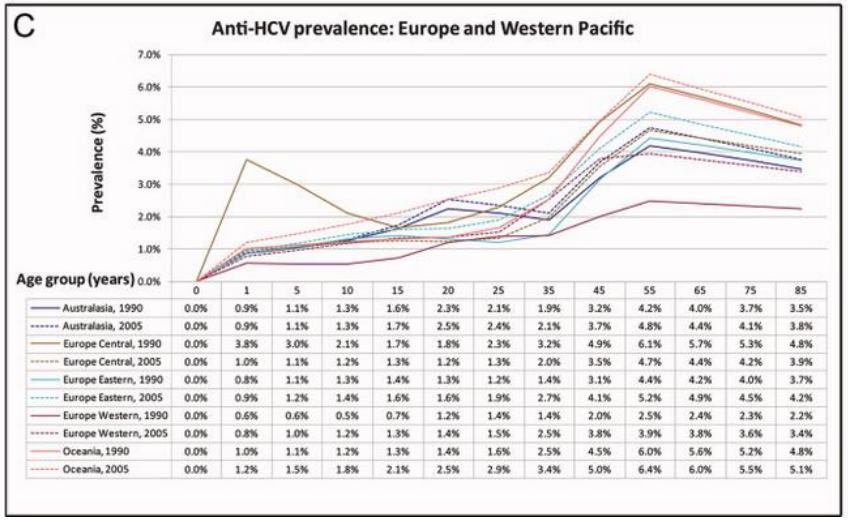
However, this study used anti-HCV prevalence which might introduce bias because it included those with past and current infections. Moreover, because the study excludes high-risk populations (e.g. PWID, blood donors, MSM), it tends to underestimate the overall prevalence. It also only includes English literature. Furthermore, the regional estimates presented reflect prevalence of countries with the most published data without necessarily reflecting the prevalence of countries with the largest population size. For example, in Asia Pacific most data were from Japan; in South Asia, the high prevalence of anti-HCV was driven primarily by data from Pakistan; whereas the Central sub-Saharan Africa region was represented only by the Central African Republic.



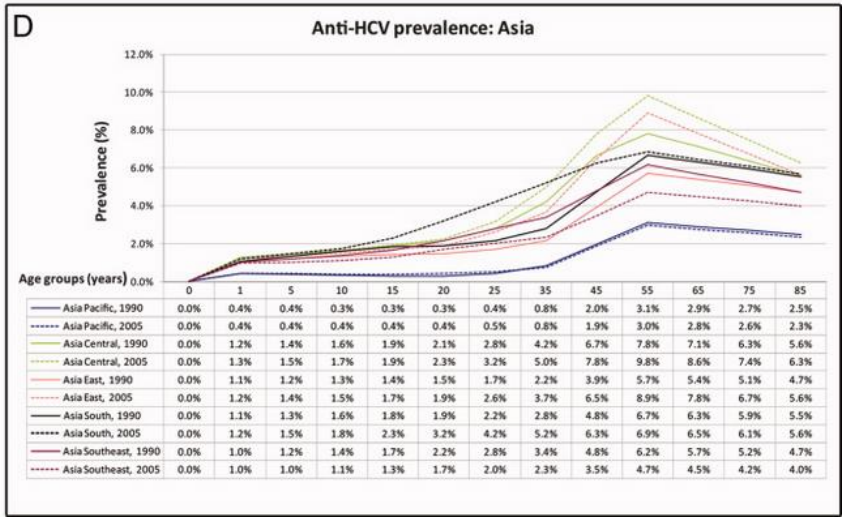
Age group (years)	0	1	5	10	15	20	25	35	45	55	65	75	85
North Africa/Middle East, 1990	0.0%	1.8%	2.1%	2.3%	2.8%	3.6%	5.4%	7.0%	8.0%	9.6%	9.0%	8.3%	7.7%
North Africa/Middle East, 2005	0.0%	1.5%	1.7%	1.8%	2.0%	2.1%	3.4%	5.5%	7.0%	8.2%	7.8%	7.4%	7.1%
Sub-Saharan Africa Central, 1990	0.0%	1.2%	1.4%	1.6%	1.8%	2.0%	2.5%	3.5%	5.4%	6.8%	6.3%	5.8%	5.3%
Sub-Saharan Africa Central, 2005	0.0%	1.2%	1.4%	1.6%	1.8%	2.0%	2.4%	3.4%	5.2%	6.7%	6.2%	5.8%	5.3%
Sub-Saharan Africa East, 1990	0.0%	0.8%	1.0%	1.1%	1.3%	1.6%	2.5%	3.8%	4.8%	5.5%	5.2%	4.8%	4.5%
Sub-Saharan Africa East, 2005	0.0%	1.1%	1.3%	1.5%	1.6%	1.7%	1.9%	2.5%	4.4%	5.9%	5.5%	5.2%	4.8%
Sub-Saharan Africa Southern, 1990	0.0%	1.0%	1.2%	1.3%	1.5%	1.6%	1.9%	2.8%	4.6%	5.9%	5.5%	5.1%	4.7%
Sub-Saharan Africa Southern, 2005	0.0%	1.0%	1.2%	1.3%	1.5%	1.7%	2.0%	2.8%	4.2%	5.3%	5.0%	4.7%	4.4%
Sub-Saharan Africa West, 1990	0.0%	2.4%	3.4%	4.5%	4.7%	3.7%	3.9%	5.1%	6.8%	8.8%	8.2%	7.7%	7.1%
Sub-Saharan Africa West, 2005	0.0%	2.0%	2.3%	2.6%	2.6%	2.2%	2.4%	3.5%	5.8%	8.1%	7.6%	7.2%	6.8%



Age group (years)	0	1	5	10	15	20	25	35	45	55	65	75	85
Caribbean, 1990	0.0%	0.8%	1.0%	1.1%	1.3%	1.4%	1.7%	2.4%	3.7%	4.7%	4.4%	4.1%	3.7%
Caribbean, 2005	0.0%	0.9%	1.0%	1.1%	1.3%	1.4%	1.7%	2.4%	3.7%	4.6%	4.3%	4.0%	3.7%
Latin America Andean, 1990	0.0%	0.9%	1.0%	1.1%	1.3%	1.4%	1.8%	2.5%	3.8%	4.8%	4.5%	4.2%	3.8%
Latin America Andean, 2005	0.0%	0.9%	1.0%	1.2%	1.3%	1.5%	1.8%	2.5%	3.9%	4.9%	4.5%	4.2%	3.8%
Latin America Central,1990	0.0%	0.7%	0.8%	0.9%	1.0%	1.1%	1.4%	2.0%	3.2%	4.2%	3.9%	3.6%	3.4%
Latin America Central, 2005	0.0%	0.8%	0.9%	1.0%	1.1%	1.2%	1.4%	1.9%	3.0%	3.9%	3.7%	3.4%	3.2%
Latin America Southern, 1990	0.0%	0.6%	0.7%	0.8%	0.9%	0.9%	1.1%	1.5%	2.3%	3.0%	2.8%	2.6%	2.5%
Latin America Southern, 2005	0.0%	0.7%	0.8%	0.9%	1.0%	1.0%	1.2%	1.7%	2.6%	3.3%	3.1%	3.0%	2.8%
Latin America Tropical, 1990	0.0%	0.4%	0.5%	0.5%	0.7%	0.8%	1.0%	1.3%	2.1%	2.7%	2.5%	2.3%	2.1%
Latin America Tropical, 2005	0.0%	0.5%	0.5%	0.6%	0.7%	0.9%	1.1%	1.6%	2.4%	3.0%	2.8%	2.5%	2.3%
North America, 1990	0.0%	0.4%	0.4%	0.5%	0.6%	0.9%	1.8%	2.5%	2.2%	1.9%	1.9%	1.8%	1.7%
North America, 2005	0.0%	0.6%	0.6%	0.6%	0.6%	0.5%	0.6%	1.4%	2.4%	2.7%	2.5%	2.3%	2.0%



Age group (years)	0	1	5	10	15	20	25	35	45	55	65	75	85
Australasia, 1990	0.0%	0.9%	1.1%	1.3%	1.6%	2.3%	2.1%	1.9%	3.2%	4.2%	4.0%	3.7%	3.5%
Australasia, 2005	0.0%	0.9%	1.1%	1.3%	1.7%	2.5%	2.4%	2.1%	3.7%	4.8%	4.4%	4.1%	3.8%
Europe Central, 1990	0.0%	3.8%	3.0%	2.1%	1.7%	1.8%	2.3%	3.2%	4.9%	6.1%	5.7%	5.3%	4.8%
Europe Central, 2005	0.0%	1.0%	1.1%	1.2%	1.3%	1.2%	1.3%	2.0%	3.5%	4.7%	4.4%	4.2%	3.9%
Europe Eastern, 1990	0.0%	0.8%	1.1%	1.3%	1.4%	1.3%	1.2%	1.4%	3.1%	4.4%	4.2%	4.0%	3.7%
Europe Eastern, 2005	0.0%	0.9%	1.2%	1.4%	1.6%	1.6%	1.9%	2.7%	4.1%	5.2%	4.9%	4.5%	4.2%
Europe Western, 1990	0.0%	0.6%	0.6%	0.5%	0.7%	1.2%	1.4%	1.4%	2.0%	2.5%	2.4%	2.3%	2.2%
Europe Western, 2005	0.0%	0.8%	1.0%	1.2%	1.3%	1.4%	1.5%	2.5%	3.8%	3.9%	3.8%	3.6%	3.4%
Oceania, 1990	0.0%	1.0%	1.1%	1.2%	1.3%	1.4%	1.6%	2.5%	4.5%	6.0%	5.6%	5.2%	4.8%
Oceania, 2005	0.0%	1.2%	1.5%	1.8%	2.1%	2.5%	2.9%	3.4%	5.0%	6.4%	6.0%	5.5%	5.1%



Age groups (years)	0	1	5	10	15	20	25	35	45	55	65	75	85
Asia Pacific, 1990	0.0%	0.4%	0.4%	0.3%	0.3%	0.3%	0.4%	0.8%	2.0%	3.1%	2.9%	2.7%	2.5%
Asia Pacific, 2005	0.0%	0.4%	0.4%	0.4%	0.4%	0.4%	0.5%	0.8%	1.9%	3.0%	2.8%	2.6%	2.3%
Asia Central, 1990	0.0%	1.2%	1.4%	1.6%	1.9%	2.1%	2.8%	4.2%	6.7%	7.8%	7.1%	6.3%	5.6%
Asia Central, 2005	0.0%	1.3%	1.5%	1.7%	1.9%	2.3%	3.2%	5.0%	7.8%	9.8%	8.6%	7.4%	6.3%
Asia East, 1990	0.0%	1.1%	1.2%	1.3%	1.4%	1.5%	1.7%	2.2%	3.9%	5.7%	5.4%	5.1%	4.7%
Asia East, 2005	0.0%	1.2%	1.4%	1.5%	1.7%	1.9%	2.6%	3.7%	6.5%	8.9%	7.8%	6.7%	5.6%
Asia South, 1990	0.0%	1.1%	1.3%	1.6%	1.8%	1.9%	2.2%	2.8%	4.8%	6.7%	6.3%	5.9%	5.5%
Asia South, 2005	0.0%	1.2%	1.5%	1.8%	2.3%	3.2%	4.2%	5.2%	6.3%	6.9%	6.5%	6.1%	5.6%
Asia Southeast, 1990	0.0%	1.0%	1.2%	1.4%	1.7%	2.2%	2.8%	3.4%	4.8%	6.2%	5.7%	5.2%	4.7%
Asia Southeast, 2005	0.0%	1.0%	1.0%	1.1%	1.3%	1.7%	2.0%	2.3%	3.5%	4.7%	4.5%	4.2%	4.0%

Figure 1.3 Estimated Anti-HCV Seroprevalence by Age and Global Burden of Disease (GBD) Region between 1990 and 2005 (11)
The figure above shows anti-HCV prevalence changes between 1990 and 2005 based on age group in 4 main regions (Africa and Eastern Mediterranean; Americas; Europe and Western Pacific; Asia). There was an overall increase risk of infection for people age between 20-55 years old, with the peak at 55 years old.

The most up to date HCV prevalence estimate was described by Gower et al. based on data from 87 countries who reported anti-HCV prevalence and 54 countries who reported rates of HCV viraemia (HCV-RNA positive). Overall this study reported lower estimates of global prevalence. Gower et.al argued that the previous estimates of HCV prevalence were likely to be overestimates due to the sampling frame (majority of country-level studies being conducted among adult population thus could not be interpreted for the entire population as infection rates are lower in the children) and selection bias (several studies included individuals who had achieved spontaneous clearance or been treated for HCV). The study estimated the global prevalence to be 2.0% (1.7–2.3%) in adults, representing 104 (87–124) million infections. Furthermore, 11 million children (<15 years) were estimated to be infected with HCV corresponding to a prevalence of total prevalence of 1.6% (1.3–2.1%).(4) The study also estimated viraemic prevalence of 1.1%, corresponding to 80 million of HCV infections. Accurate global estimates of the prevalence of hepatitis C are important for developing program intervention and strategies.

The same study (4) also reported the regional prevalence of HCV and found the highest anti-HCV prevalence in Central Asia followed by West and Central Sub-Saharan Africa, Eastern Europe, and Middle East countries (Figure 1.4). Based on laboratory diagnoses of viraemia (HCV-RNA positive cases excluding antibody patients who have spontaneously cleared), there are approximately 80 (95% CI: 64-103) million cases, 50% of which occur in individuals from China, Nigeria, Pakistan, India, Egypt, and Russia.(4)

Although the prevalence estimates from Gower et.al study tends to be more accurate because it presents the viraemic rate (HCV-antibody and HCV-RNA positive), only some countries had viraemic data available (54 out of 195 countries). Another limitation was the way national prevalence was reported (some studies reported the HCV prevalence based on the sampling in the study). Furthermore, not all countries provided age-specific prevalence rates.

Due to the nature of HCV transmission, the burden of disease in developed countries falls mainly on marginalised populations such as people who inject drug (PWID). Obtaining accurate estimates of the burden of HCV in marginalised populations is arguably even more challenging than deriving population estimates as they can be hard to access for epidemiological studies, especially if they are not in regular contact with drug treatment services. Better data are therefore urgently needed to inform strategies to reduce HCV infection in these groups. This issue is

discussed in detail in chapter 5 where I report findings from a study which estimates the prevalence of HCV among people who are homeless, drug users, and prisoners in London.

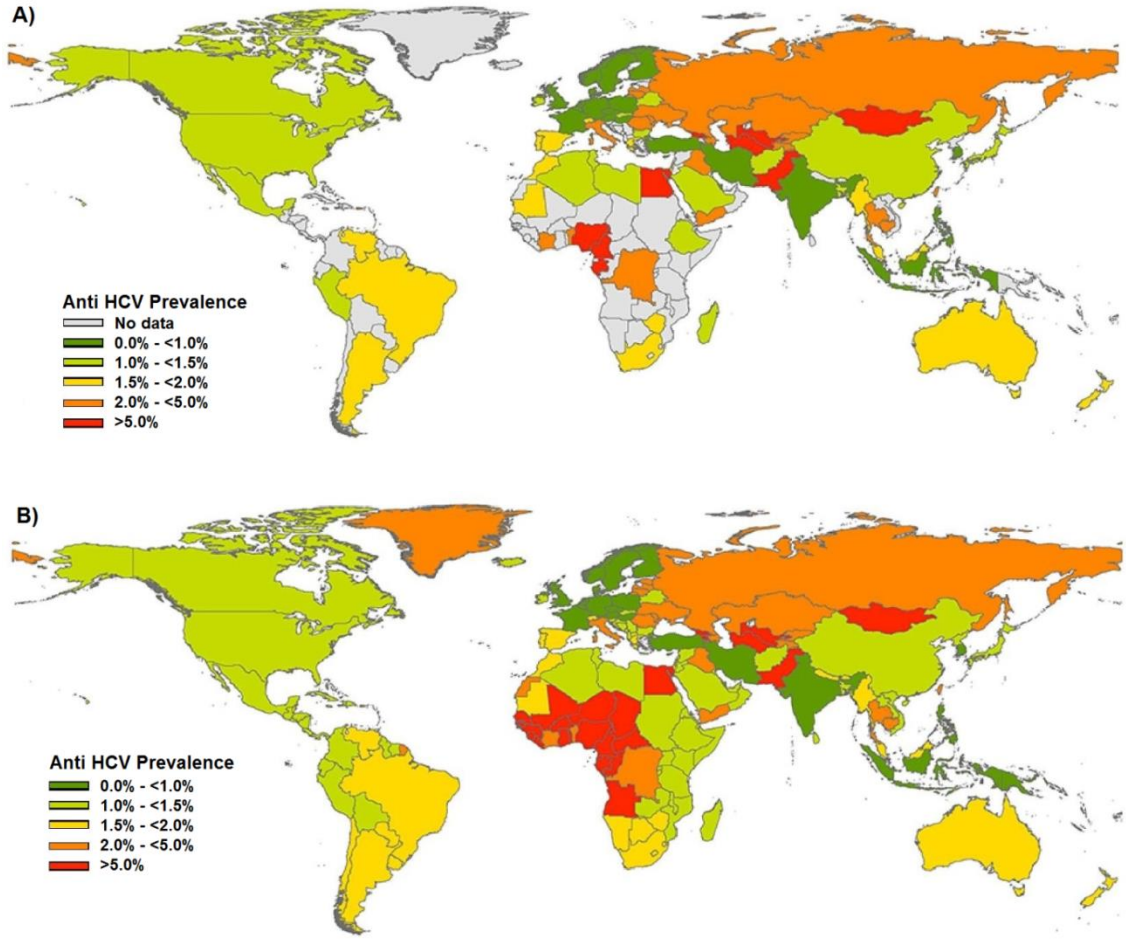


Figure 1.4 Global HCV Prevalence and Number of Infections among Adults (Gower et al.) (A) Anti-HCV Prevalence (Reported) (B) Anti-HCV prevalence (Reported and Estimated).(4)

1.2.2 Challenges Assessing HCV Prevalence

Assessing the accurate prevalence of hepatitis in general population is very challenging. The prevalence reported will depend on participants recruited and how they were selected, thus it is difficult to generate representative prevalence of the total population. Numerous prevalence studies have been conducted among high-risk populations, such as PWIDs, haemodialysis patients, men who have sex with men but these, of course overestimate the prevalence in the general population. In many studies, HCV prevalence among blood donors is widely used as an estimate as it represents an attractive data source due to the large sample size and the fact that it does not bias towards high risk groups. However, this population often corresponds to healthy screened adults who are not representative of the total

population and may explicitly exclude those with history of injecting drug use, thus potentially leading to artificially low population estimates.

The other challenge is because prevalence studies often derive data from convenient samples, such as confirmed laboratory reports rather than population screening. Again, confirmed laboratory report from specific setting or population (e.g. pregnant women, health care patients, screening participants, military recruits, blood donors, etc.) will affect the representativeness of the data. Next, the definition of HCV infection based on diagnostic test should be distinguished between anti-HCV prevalence and viraemic prevalence. Anti-HCV prevalence may include cured individuals due to spontaneous clearance or through treatment because using antibody-HCV test, whereas viraemic prevalence only represents individuals with current HCV infections (based on HCV-RNA positivity). Thus, using anti-HCV prevalence as an estimate might be over representative compared to viraemic prevalence.

On the other hand, assessing prevalence among high risk groups is also challenging. The estimated prevalence might be underrepresented because these populations are often hard to be screened. Studies usually recruit participants from specific settings such drug treatment services and sexual health clinics where they were only able to recruit individuals who were in contact with those services and did not include those who were not engage with the services. In order to monitor progress towards the WHO elimination targets in different countries it would be beneficial if WHO published standardised approaches to estimation of prevalence.

1.3 Epidemiology of Hepatitis C

1.3.1 Mode of Transmission

There are 3 main routes of HCV infections which have been well documented, including parenteral transmission (intravenous drug use, blood product transfusion, organ transplantation), permucosal transmission (usually by sex), and vertical transmission (mother to child around the time of birth). Based on many published papers, the majority of HCV infections worldwide are transmitted through blood transfusion from unscreened blood donors, injection drug use, unsafe therapeutic injections practices, and other health care related procedure.(12-14) Sexual transmission also poses a risk of transmission while a small proportion of HCV patients were infected by vertical transmission.

The main route of HCV transmission varies between countries, especially in developed and developing nations, which is likely to be explained at least in part by socioeconomic factors. In most developed countries, people who inject drugs (PWID) are the main source of transmission, accounting for 60-90% of HCV infections in several European countries, the United States, and Australia.(12) In developing and transitional economy countries, unsafe therapeutic injections became the major source of transmission due to the re-use of contaminated or poorly sterilized syringes and/or needles during health care procedures. In Egypt, the use of unsterilized syringes and needles during schistosomiasis mass treatment programs has led to hepatitis C prevalence of 21.9% (95% CI 21.0–22.8).(13) A systematic review conducted in 2011 tried to summarize the various routes of HCV infection in several countries worldwide, suggesting different main routes of infection geographically (see figure 1.5).(14, 15)

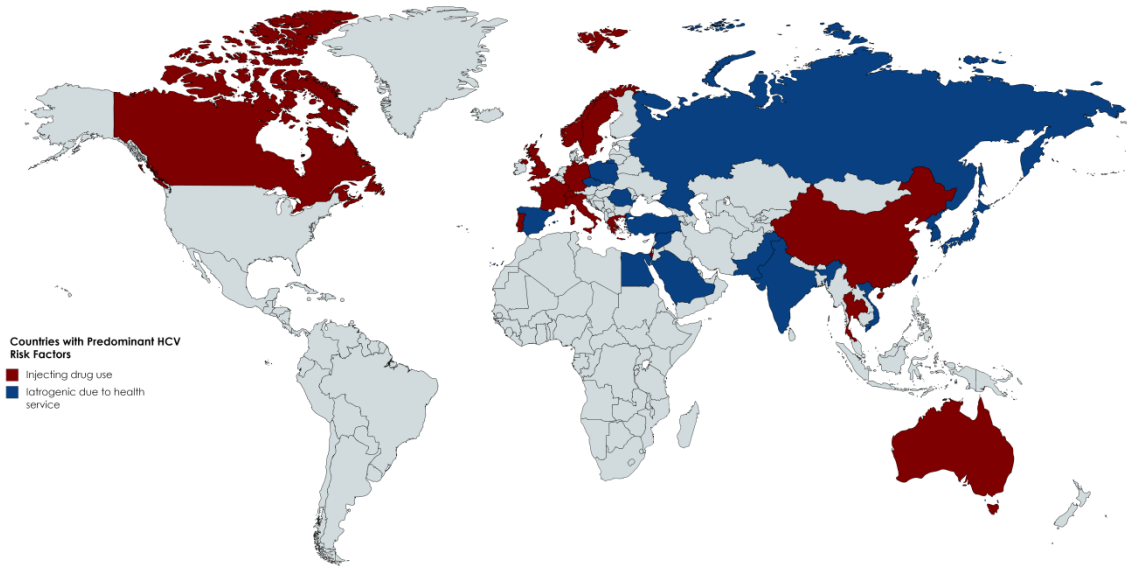


Figure 1.5 Countries with HCV predominant source of infection of (red) injecting drug use and (blue) iatrogenic due to health service[1]

Although both systematic reviews summarised the major route of transmission which are varied between countries, there are some limitations. Data were not available for many countries (only 30 out of 195 countries reported in the study). The studies did not distinguish between acute and chronic infections and there was considerable variability among the type and quality of prevalence studies among the countries assessed (some countries had undertaken population studies, others were limited to urban populations, some only included sub-populations e.g. blood

[1] The map was generated based on systematic review results from Cornberg et.al and Sievert et.al

donors, hospital workers). The sources of infections presented in some countries were based on summaries from several studies.

1.3.2 Risk Factors

Risk factors for acquiring HCV infection have been investigated from many prospective and retrospective cohorts, and case control studies. The majority of risk factors that have been identified relate to contamination with a source of HCV infected blood e.g. contaminated blood transfusion, injecting drug use, organ transplantation, unsafe therapeutic injections during medical procedure, birth to an infected mother, occupational exposure due to infected with contaminated needle sticks, or sex with infected or multiple sex partners.(16) It remains unclear whether behaviours such as tattooing, body piercing, acupuncture, dental procedure, intranasal cocaine use, and sharing personal items such as razor blades or toothbrushes play a role in transmission of HCV.

1.3.2.1 Blood Transfusion

Blood transfusion is a highly effective means of HCV transmission. Transfusion-associated HCV cases are mostly a result of inadequate screening of blood donors. Screening of donated blood for HCV only became possible after identification of the virus, was not widely adopted until after 1992 and is still not conducted in many low-income countries. People who received blood transfusions before 1992 are considered to have 2.6 times higher risk of HCV infection compared with individuals who did not receive transfusion.(17) Some studies in USA and Italy have investigated long term outcomes in individuals with transfusion-acquired HCV infection, demonstrating that 75% of patients were HCV positive within a mean of 15 years after transfusion.(18, 19) It has been estimated that approximately 10% of chronic hepatitis C among population was come from past blood transfusion.

Over the last five decades, the risk of HCV infection following blood transfusion has declined due to improved screening of donated blood, including appropriate laboratory tests for liver disease (anti-HCV test, and HCV nucleic acid test). In 2001, the risk of getting infected with HCV from a unit of transfused blood was less than one per million transfused units in developed countries. However, the risk of acquiring HCV through blood transfusion remains in developing nations. The underlying problems may be related to socioeconomic condition like poverty, lack of infrastructure, the shortage of trained professionals, and inadequate supplies and equipment.(20-22)

The current data from the World Health Organization (WHO) indicates that blood donations are not routinely tested for any blood borne diseases including hepatitis C, hepatitis B, HIV, and syphilis in 39 countries; whereas 47% of blood donations were tested without quality assurance in low-income countries.(23) A recent literature review assessing the safety of blood transfusion in Africa found that rates of blood transfusion-associated hepatitis C were high in West and North Africa but low in the Southern part of Africa.(24) A global review published in 2006 summarised the differences of blood transfusion systems among 178 countries grouped by Human Development Index (HDI) level (Table 1.2).(25) The study showed that the unit screened for anti-HCV in countries with low HDI is much lower compared to countries with medium and high HDI, which might explained the higher prevalence of anti-HCV among repeated donors and first time donors.

Table 1.1 Blood Transfusion System in 178 Countries by Human Development Index (25)

	Human Development Index (HDI)		
	Low	**Medium**	**High**
No. of countries (%)	36 (20)	88 (50)	54 (30)
Percentage of world population	11	71	18
No. of units donated per year, in millions (%)	2.3 (3)	29.4 (36)	49.4 (61)
Litres of plasma designated for fractionation, in millions	0.03	4.4	15.7
Estimated blood donation rates, per 1000 inhabitants	3.3	10.6	40
Blood donor system			
• **Voluntary donors (%)**	34	60	94
• **Paid donors (%)**	63	36	4
• **Replacement donors (%)**	3	4	2
Anti-HCV screening			
• **Units screened for anti-HCV (%)**	51.3	96.3	99.9
• **Prevalence of anti-HCV in repeat donors (%)**	1.54	0.21	0.007
• **Prevalence of anti-HCV in first time donors (%)**	3.89	1.49	0.21

The table shows lower anti-HCV screening in low HDI countries compared to medium and high HDI countries. The prevalence of anti-HCV in repeat donors and first-time donor was significantly higher in low HDI countries compared to medium and high HDI countries.

1.3.2.2 Injection Drug Use

Injection drug use has been identified as the primary mode of transmission for hepatitis C in developed nations. Injection drug use and unsafe health-care procedures were the leading causes of new HCV infections, accounting for most of the 1.75 million new infections in 2015 worldwide.(10) A recent systematic review including data from 25 countries estimated that 60–80% of PWIDs were anti-HCV

positive, equating to approximately 10 million newly infected cases in 2010 (range 6.0–15.2).(26) Furthermore, the study showed that China contributed approximately 1.6 million HCV cases among PWIDs, whereas USA and Russia contributing about 1.5 million and 1.3 million, respectively. The largest number of estimated HCV cases was in Eastern Europe with 2.3 million infected individuals (range 1.2–3.9), followed by China (1·6 million, range 1.1–2.2), Russia (1.3 million range 0.7–2.3), and USA (1.5 million, range 1.0-2.2). However, these estimates were based on number from HCV antibody positive participants, which might introduce bias because it includes those who might have cleared the virus. Convenience sampling was most often used in this study. Furthermore, other limitation was representativeness of PWID samples (some studies recruited participants who had ever injected drugs, some recruited those who had injected in the past year or were current users). Although the systematic review tried to maintain the quality of studies included using grading score, it also included older studies which potentially had less accurate serological testing techniques and included studies undertaken in countries where laboratory capacity is low which increases uncertainty about the validity of HCV reports. The prevalence map of hepatitis C infected individuals among PWID can be seen at Figure 1.6.

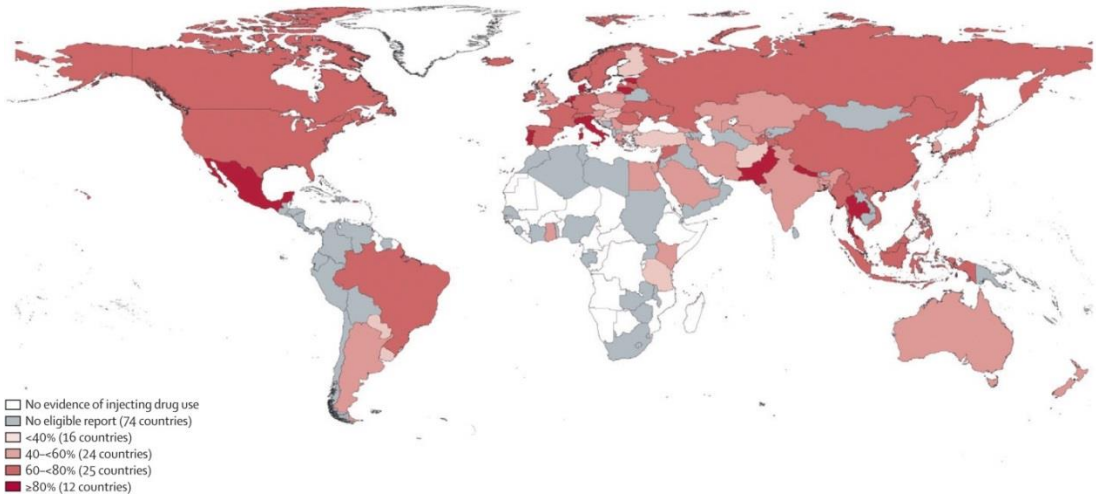


Figure 1.6 Prevalence of Hepatitis C Antibody Positive among IDU (26)

1.3.2.3 Unsafe Therapeutic Injections

It is difficult to quantify the relative contribution of health care related HCV transmission in developed countries. There have been a number of outbreaks due to negligence of aseptic techniques or medical practices. Most nosocomial transmission in hospital is related to haemodialysis. A Large number of studies have already assessed the prevalence of hepatitis C among transfusion dependent

patients, with a range of 4% up to 59% which varies between countries.(27) The Dialysis Outcomes and Practice Patterns (DOPPS) study estimated the median prevalence of HCV infection among dialysis patients to be 14% (range 2.6% - 22.9%) in developed countries between 1990 and 2007 (Table 1.3).

Table 1.2 Prevalence and Incidence of Hepatitis C in Haemodialysis Setting in Developed Countries, 1990-2007

Country	Prevalence (95% CI) (%)	Incidence (95% CI) (No. of cases per 100 person/year)
France	10.4 (9.7–11.2)	2.0 (1.4–2.8)
Germany	3.8 (3.3–4.4)	1.7 (1.2–2.5)
Italy	20.5 (19.4–21.7)	3.9 (2.9–5.2)
Japan	14.8 (14.0–15.6)	3.0 (2.3–3.9)
Spain	22.9 (21.7–24.1)	3.5 (2.5–4.8)
United Kingdom	2.6 (2.1–3.2)	1.2 (0.7–2.0)
United States	14.0 (13.6–14.5)	2.5 (2.1–1.9)

In developing nations, the risk of HCV infection is not only limited to occasional outbreaks, but can be associated with the supply of contaminated injection equipment and lack of adequately trained staff and use of injected medication.(28) In 2000, WHO estimated almost 40% of injections (6.7 billion) were given through re-used equipment, with the largest number of cases found in Middle East, Southeast Asia and Western Pacific.(29)

1.3.2.4 Sexual Transmission

The role of sexual intercourse in the transmission of HCV remains uncertain with inconsistencies between studies. Nevertheless, some well documented studies showed an increased risk of infection after a defined sexual exposure.(30-32) A study in United States found that 15-20% of HCV cases were transmitted through sex and the risk of infection increased with higher-risk sexual exposure, more sexual partners, and unprotected sex.(33, 34) Furthermore, sexual transmission has been confirmed in 15% of HCV individuals' sexual partners through phylogenetic analysis.(35) One possible explanation of these inconsistent results may relate to sexual activity during the acute phase of HCV infection when viral concentration is presumably higher and individuals are more likely to transmit/contract HCV. A study conducted in Italy also reported the association between acute HCV infection and multiple sexual partners (36) whereas a very low incidence of infection was found

among monogamous couples (37). A systematic review assessed the incidence of HCV among men who have sex with men (MSM) in HIV-positive and negative individuals. The study generated pooled incidence of 1.48/1000 person-years (95% CI: 0.75-2.21) for HIV-negative MSM subgroup and 6.08/1000 person-years (95% CI: 5.18-6.99) for HIV-positive MSM subgroup, indicating up to a 4-fold increase in the risk of HCV infection among HIV infected individuals.(38) However, this study did not consider injecting drug use as confounder which might introduce bias and affect the outcome, particularly if injection drug use is more prevalent in MSM who have multiple sexual partners.

1.3.2.5 Vertical Transmission

Studies investigating vertical transmission from HCV-infected mothers have been done through prospective cohort and retrospective studies. It was estimated that the risk of mother to child transmission ranges from 4% to 10% for HCV.(39) Mothers with higher levels of HCV RNA and HIV-positive mothers pose an increased risk of perinatal transmission.(40) The most updated meta-analysis conducted in 2014 revealed that the risk of vertical transmission was 5.8% (95% CI: 4.2%-7.8%) for children whose mothers were HIV-negative and 10.8% (95% CI: 7.6%–15.2%) for children of HIV-positive mothers.(41) The study suggested HIV coinfection as the most important risk factors for perinatal transmission (OR=2.56, 95% CI: 1.50–4.43).

1.3.2.6 Others

A range of studies have investigated the role of factors such as tattooing, body piercing, acupuncture, dental procedure, intranasal cocaine, use sharing razor blades or toothbrushes in HCV transmission.(42-44) A meta-analysis has been conducted to investigate the association of tattooing with HCV infection, extracting 124 studies for systematic review, 83 of which were included in meta-analysis. The study generated pooled odds ratio of 2.74 (95% CI: 2.38–3.15) with higher risk associated with tattooing in non-professional parlours or among friends (OR=2.8, 95% CI: 1.29-6.08).(45) The strongest association was found in Australia (OR=5.90, 95% CI: 2.62–13.30) comparing those who had history of tattooing and people with no history of tattoos, followed by Iran and Canada with odds ratio of 5.61 (95% CI: 2.31-13.62) and 5.15 (95% CI: 2.65-9.98) respectively.

A systematic review was conducted to investigate whether non-injection drug use (NIDU) was associated with an increased risk of HCV, for example through sniffing,

smoking or snorting drugs such as cocaine, heroin, crack or methamphetamine. A total of 28 studies included in an analysis generated the HCV median prevalence of 14% and broad range between 2.3% and 35.3%.(46) The broad range of prevalence might also be caused by people who fail to report their injecting use which leads to overestimation of risk. The study concluded that it was not clear whether NIDUs associated with HCV infection because much of the research did not aim to study HCV in users of non-injection drugs. Instead, NIDUs were typically included as a secondary research concern, with a principal focus on the problem of transmission of HCV in PWID populations. Another systematic review assessed the association between acupuncture and HCV infection. Fifteen studies were included in analysis, with the majority of studies having been conducted in Asia such as Taiwan and Japan. The study found a positive correlation with acupuncture only in HCV endemic areas and the association was modest ranging from OR=1.0 [95% CI: 0.9-11] to OR=3.3 [95% CI: 2.0-5.5]) .(43) Another study has demonstrated a considerable risk of HCV-RNA retrieved through toothbrushes among HCV patients, suggesting it is prudent for individuals with HCV to ensure that they do not share personal care items.(44)

1.3.2.7 Challenges Assessing HCV Risk Factors among Populations

Identifying risk factors for infection with HCV is challenging because patients rarely present during the acute phase of infection and an estimated 70%-80% proportion of cases are asymptomatic. This means the majority of studies take place in individuals who have severe or chronic HCV. Studies that take place in these populations may bias towards risk factors for severity/chronic infection as opposed to risk factors for HCV acute infection.

Second, studies also need to consider reverse causality in determining the risk factors of infection. Instead of an identified exposure causing HCV infection, it can be the other way around that infection HCV precedes the exposure. For example, a study conducted in China identified oesophageal dilatation as a risk factor for HCV; however it is also plausible that liver disease secondary to HCV condition induced the use of oesophageal balloon and endoscopy which may form part of the clinical management of liver disease.

Third, the sampling frame will have a major impact on the estimates of prevalence. For example, prevalence estimates are likely to vary depending on whether patients are recruited from secondary care, primary care, community or high-risk settings such as prisons. One method to try and minimise the impact of studies that have

focused on specific populations is to consider the role of confounding such as age, gender, ethnicity, study population, region, and healthcare setting. Utilising multivariate analysis will provide more robust results to measure the strength of relationships among various measurements compared to univariate analysis.

Finally, recall bias might also be a challenge in identifying risk factors of HCV infection. Some participants might not remember precisely the initial time of exposures. Moreover, it is also possible that no record of risk factors was available or some participants might conceal their answer (e.g. PWIDs did not want to tell that they were injecting).

1.4 Natural History of HCV

The Acute phase of HCV infection is usually mild, unrecognised, and rarely diagnosed. The majority of acute HCV patients are asymptomatic, and only 20-30% develop symptoms, such as jaundice, malaise, fatigue, nausea, abdominal pain, myalgia, or febrile illness, thus very few of infected individuals seek medical care during the early phase. These symptoms may persist from days up to 12 weeks after the initial infection. The average incubation period ranges between 2-12 weeks and HCV RNA becomes positive after 1-3 weeks of exposure (47) whereas the antibody becomes detected within 3-21 weeks (mean 7 weeks) (48).

About 15-35% of acute HCV patients will clear the virus spontaneously, yet the chance of clearance may be as high as 50% amongst symptomatic patients.(49) A number of studies have been undertaken to assess the relationship between spontaneous viral clearance (SVC) and host, viral and environmental factors, employing a range of study designs and populations. In chapters 2 and 3, I attempt to combine such studies through a systematic review and meta-analysis which estimates the prevalence of SVC and identifies risk factors associated with clearance.

Approximately 65%-85% of individuals will develop chronic HCV infection, defined as the persistence of HCV RNA for at least 6 months or longer in the serum. Disease progression is slow, with 4-20% of individuals with HCV developing cirrhosis over a 20 years period, depending on the study population. The rate of cirrhosis in healthy blood donors was 4% (95% CI: 1-7%) over 20 years, and 7% (95% CI: 4-10%), 22% (95% CI: 18-26%), 24% (95% CI: 11-37%) among community-based study, patients referred to liver clinics, and post-transfusion cohorts, respectively.(50, 51) A systematic review based on 111 studies estimated the prevalence of cirrhosis at 16% after 20 years of infection.(52)

Individuals infected with HCV may have no symptoms and live a normal, life, for decades, and remain unaware of their infection. Alternatively, chronically infected individuals may develop cirrhosis which is preceded by liver fibrosis (formation of scar tissue in the liver, categorised as stages 0-4). Fibrosis progression rates are extremely variable and can be influenced by host, viral and environmental factors. The rates of progression are not linear and may vary between fibrosis stages and accelerate with duration of infection or aging.(53, 54) Serial biopsies in patients with hepatitis C have shown annual rates of fibrosis progression to be between 0.1 and 0.2 stages per year.(55, 56) Several factors which increase an individual's risk of fibrosis progression have been identified, including age, male gender, alcohol consumption of more than 50 g/day, obesity, insulin resistance, type 2 diabetes, co-infection with hepatitis B or HIV, immunosuppressive therapy, and host genetic factors.(57, 58)

Individuals with cirrhosis are at increased risk of decompensated cirrhosis and hepatocellular carcinoma (HCC). Decompensated cirrhosis is a condition where the liver is not able to perform all its functions adequately. Without supportive care or procedure such as liver transplantation patients with decompensated cirrhosis will progress to liver failure and die from complications of liver disease.

A cohort study of individuals with HCV with more than 10 years follow-up found that once cirrhosis has developed, the risk of decompensated liver disease was about 4% per year, including development of ascites, variceal bleeding and encephalopathy.(59) The study estimated that 96% of individuals with compensated cirrhosis remained alive at 3 years, declining to 79% after 10 years. Individuals with HCV advanced cirrhosis and fibrosis are at higher risk of getting hepatocellular carcinoma (HCC), with estimated risk of development of HCC of 3% (range 1-7%) per year.(60, 61)

Several studies have investigated factors associated with hepatitis C disease progression. Male sex, older age, African-American race, obesity, HIV co-infection, diabetes and steatosis have been shown to be associated with an increased risk of HCC development.(62-64) Some modifiable factors include smoking, alcohol consumption, and obesity have also been linked to risk of HCC.(65, 66) Some studies suggested the risk of liver fibrosis progression increased with age.(67) and HCV acquisition after the age of 40 has been associated with accelerated progression of liver injury.(68) Co-infection with hepatitis B might induce rapid progression to cirrhosis.(69) A meta-analysis generated relative risk of cirrhosis

development at 2.33 (95% CI: 1.67-3.26) for people with heavy alcohol intake (>560 g/week) compared to people with 210-260 g/week alcohol intake.(70)

Another rare condition that may be found among hepatitis C infected patients is fulminant hepatitis. Fulminant hepatic failure is characterized by the development of severe liver injury with impaired synthetic capacity and encephalopathy in patients with previous normal liver or at least well compensated liver disease. The natural history of hepatitis C is summarised in figure 1.7 below.

1.4.1 Difficulties in Studying the Natural History of Hepatitis C Infection.

A number of difficulties complicate the measurement of disease progression rates in hepatitis C infection. Studies may not be able to accurately define the onset of infection. Furthermore, studies may not include representative patient groups. Since progression is slow follow up over many years in needed so patients are likely to be lost to follow up. This loss to follow up may affect vulnerable groups such as drug users more than other, more stable groups.

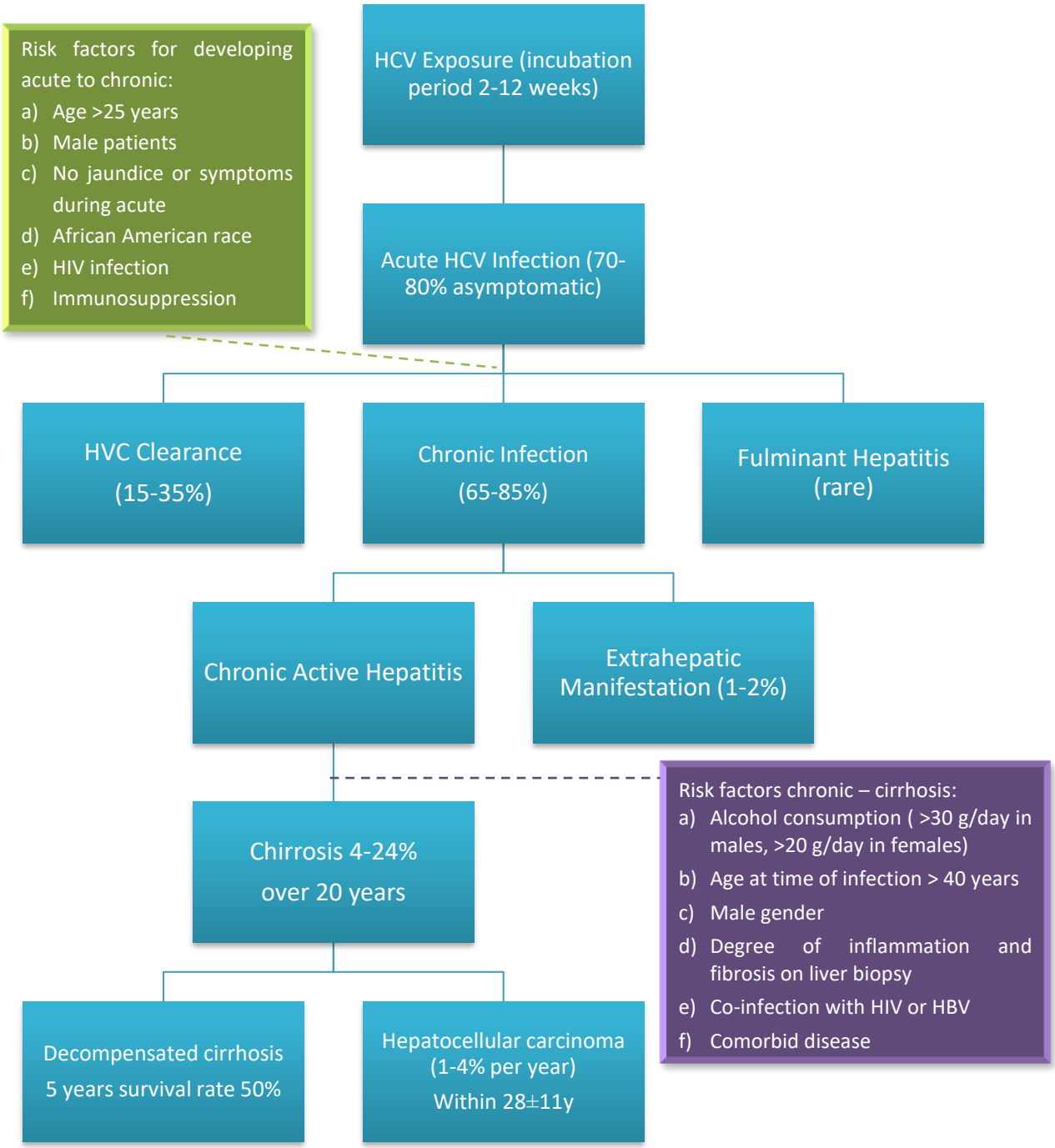


Figure 1.7 Natural History of Hepatitis C (71, 72)

1.5 Hepatitis C Treatment – Entering the Era of Direct Acting Antiviral Agents (DAA)

For more than a decade, the combination of PEGylated interferon-β (PEG-IFN) and ribavirin (RBV) administered for 24 or 48 weeks was the standard treatment for HCV chronic infection.(73) The length of treatment depended on HCV genotype, for example those who are infected with genotype 1 needed to have longer duration of treatment compared to patients with genotype 2 or 3. The efficacy of these treatments (defined as SVR – Sustained Virologic Response) ranges from 40% and 60% for patients with genotype 1 infection (74-76) and 80%-90% for patients with genotype 2 and 3 infection (77-79). However, a major limitation of PEG-IFN/RBV regimens is their side effects and tolerability. Reported side effects of interferon-based therapy include influenza-like symptoms, neuropsychiatric symptoms, hematologic abnormalities (75, 80, 81), and depression which can occur in up to 30% of patients (75). In phase 3 trials of 1530 patients, adverse events led to the need to reduce the dose of treatment in 32%-42% of patients and to discontinue therapy in 10% to 14%.

In order to address the limitations of PEG-IFN/RBV therapy, new direct-acting antivirals (DAAs) have been developed that target specific HCV functions. Several oral regimens combining DAAs from different nucleotide inhibitor families (NS5B nucleotide inhibitors, NS5B non-nucleoside inhibitors, NS5A replication complex inhibitors and NS3/4A protease inhibitors) have been developed.(82-84) These regimens achieve sustained virological response (SVR) rates in >90% of patients and have the added benefit of reducing the duration of treatment to 12 weeks or less.(85)

The first generation of DAAs treatment have been approved including bocepevir/BOC (86) and telapevir/TVR (87), for use in combination with the PEG-IFN/RBV. These new triple therapy regimens represented a dramatic advance in the efficacy of HCV treatment, ranging between 70% and 80% for patient with genotype 1 and significantly reduced treatment duration.(88-90) However, the first generation of DAA treatments were associated with a high pill burden, an inconvenient dosing frequency, as well as selection for DAA-resistant viral variants among patients who responded poorly to the PEG-IFN/RBV component of combination therapy.(91)

To overcome the limitations of the first generation of DAA's, newer DAAs have been developed with the goal of better efficacy, pan-genotypic treatment (works against

every genotype), increased convenience in terms of dosing and low propensity to develop viral resistance. Sofosbuvir (SOF) is the first-in-class which has been approved by the Federal Drug Authorities (FDA) and the European Medicine Agency (EMA) as a pan-genotypic treatment in IFN-free therapy combinations either with RBV or other DAAs.(92) The second wave of first generation DAA's approved by the FDA and EMA includes Simeprevir (SMV) with impressive antiviral activity in HCV genotypes 1 and 4. Daclatasvir is a potent first-in-class NS5A inhibitor that has been shown to have pan-genotypic activity.(93) Below is the summary of HCV treatment guideline recommended by EASL for HCV-monoinfected patients and HCV/HIV-coinfected patients with and without decompensated cirrhosis.(94)

Table 1.3 Treatment recommendations for HCV-monoinfected or HCV/HIV-coinfected patients with chronic hepatitis C without cirrhosis, includingtreatment-naïve patients (defined as patients who have never been treated for their HCV infection) and treatment-experienced patients (defined aspatients who were previously treated with pegylated IFN-a and ribavirin; pegylated IFN-a, ribavirin and sofosbuvir; or sofosbuvir and ribavirin).

Patients	Prior treatment experience	SOF/VEL	GLE/PIB	SOF/VEL/VOX	SOF/LDV	GZR/EBR	OBV/PTV/r + DSV
Genotype 1a	Treatment-naïve	12 wk	8 wk	No	8-12 wk	12 wk (HCV RNA ≤800,000 IU/ml)	No
	Treatment-experienced	12 wk	8 wk	No	No	12 wk (HCV RNA ≤800,000 IU/ml)	No
Genotype 1b	Treatment-naïve	12 wk	8 wk	No	8-12 wk	8 wk (F0-F2) 12 wk (F3)	8 wk (F0-F2) 12 wk (F3)
	Treatment-experienced	12 wk	8 wk	No	12 wk	12 wk	12 wk
Genotype 2	Treatment-naïve	12 wk	8 wk	No	No	No	No
	Treatment-experienced	12 wk	8 wk	No	No	No	No
Genotype 3	Treatment-naïve	12 wk	8 wk	No	No	No	No
	Treatment-experienced	12 wk	12 wk	No	No	No	No
Genotype 4	Treatment-naïve	12 wk	8 wk	No	12 wk	12 wk (HCV RNA ≤800,000 IU/ml)	No
	Treatment-experienced	12 wk	8 wk	No	No	No	No
Genotype 5	Treatment-naïve	12 wk	8 wk	No	12 wk	No	No
	Treatment-experienced	12 wk	8 wk	No	No	No	No
Genotype 6	Treatment-naïve	12 wk	8 wk	No	12 wk	No	No
	Treatment-experienced	12 wk	8 wk	No	No	No	No

)AA, direct-acting antiviral; DSV, dasabuvir; EBR, elbasvir; GLE, glecaprevir; GZR, grazoprevir; HCV, hepatitis C virus; HIV, human immunodeficiency virus; LDV, ledipasvir;)BV, ombitasvir; PIB, pibrentasvir; PTV, paritaprevir; r, ritonavir; SOF, sofosbuvir; VEL, velpatasvir; VOX: voxilaprevir.

Table 1.4 Treatment recommendations for HCV-monoinfected or HCV/HIV-coinfected patients with chronic hepatitis C with compensated (Child-Pugh A) cirrhosis, including treatment-naïve patients (defined as patients who have never been treated for their HCV infection) and treatment-experienced patients (defined as patients who were previously treated with pegylated IFN-a and ribavirin; pegylated IFN-a, ribavirin and sofosbuvir; or sofosbuvir and ribavirin).

Patients	Prior treatment experience	SOF/VEL	GLE/PIB	SOF/VEL/VOX	SOF/LDV	GZR/EBR	OBV/PTV/r + DSV
Genotype 1a	Treatment-naïve	12 wk	12 wk	No	12 wk	12 wk (HCV RNA ≤800,000 IU/ml)	No
	Treatment-experienced	12 wk	12 wk	No	No	12 wk (HCV RNA ≤800,000 IU/ml)	No
Genotype 1b	Treatment-naïve	12 wk	12 wk	No	12 wk	12 wk	12 wk
	Treatment-experienced	12 wk	12 wk	No	12 wk	12 wk	12 wk
Genotype 2	Treatment-naïve	12 wk	12 wk	No	No	No	No
	Treatment-experienced	12 wk	12 wk	No	No	No	No
Genotype 3	Treatment-naïve	No	12 wk	12 wk	No	No	No
	Treatment-experienced	No	16 wk	12 wk	No	No	No
Genotype 4	Treatment-naïve	12 wk	12 wk	No	12 wk	12 wk (HCV RNA ≤800,000 IU/ml)	No
	Treatment-experienced	12 wk	12 wk	No	No	No	No
Genotype 5	Treatment-naïve	12 wk	12 wk	No	12 wk	No	No
	Treatment-experienced	12 wk	12 wk	No	No	No	No
Genotype 6	Treatment-naïve	12 wk	12 wk	No	12 wk	No	No
	Treatment-experienced	12 wk	12 wk	No	No	No	No

DAA, direct-acting antiviral; DSV, dasabuvir; EBR, elbasvir; GLE, glecaprevir; GZR, grazoprevir; HCV, hepatitis C virus; HIV, human immunodeficiency virus; LDV, ledipasvir; OBV, ombitasvir; PIB, pibrentasvir; PTV, paritaprevir; r, ritonavir; SOF, sofosbuvir; VEL, velpatasvir; VOX: voxilaprevir.

Below is a summary of recent advances in the development of some DAA-based therapies, with particular emphasis on those compound classes/combinations that have shown the most encouraging antiviral activity for HCV patients.

Efficacy of Sofosbuvir/Ledipasvir

The efficacy of Sofosbuvir/Ledispavir was explored in the series of ION studies. Based on ION studies (consisting of ION-1, ION-2, ION-3), 97% (1886/1951) of genotype 1 patients who were treatment-naive and treatment-experienced with or without cirrhosis achieved SVR12 (Sustained Virologic Response at 12 weeks) across the ION studies by administering sofosbuvir (SOF) ledipasvir (LDV) fixed-dose combination.(95-97) In the ION-1 study, among treatment naive patients with genotype 1, 94% of cirrhotic patients and 99% of patients without cirrhosis achieved SVR for 12 weeks. In the ION-2 study, among treatment experienced patients with genotype 1, 86% of cirrhotic patients and 95% of patients without cirrhosis achieved SVR for 12 weeks. This study underlined that although the efficacy of DAA treatment is higher among cirrhotic patients compared to PEG-interferon therapy, patients with cirrhosis remained less effective than treatment of non-cirrhotic patients.

Paritaprevir, ombitasvir, dasabuvir (AbbVie)

Several phase 3 studies have already evaluated regimens of paritaprevir (PTV), ombitasvir (OBV), and dasabuvir (DSV) with or without ribavirin in diverse patients with HCV genotype 1: genotype 1a, genotype 1b, treatment-naive, treatment-

experienced, with or without compensated cirrhosis.(98-102) Based on the studies, the pooled efficacy for patients with genotype 1 without cirrhosis who achieved SVR12 was 96.0%, and were consistently high irrespective of prior treatment history (94.0%–100%). Among HCV genotype 1 patients with cirrhosis, overall the rate of SVR12 was 88.6% and 95.0% following treatment with OBV/PTV/r + DSV + RBV for 12 and 24 weeks respectively.

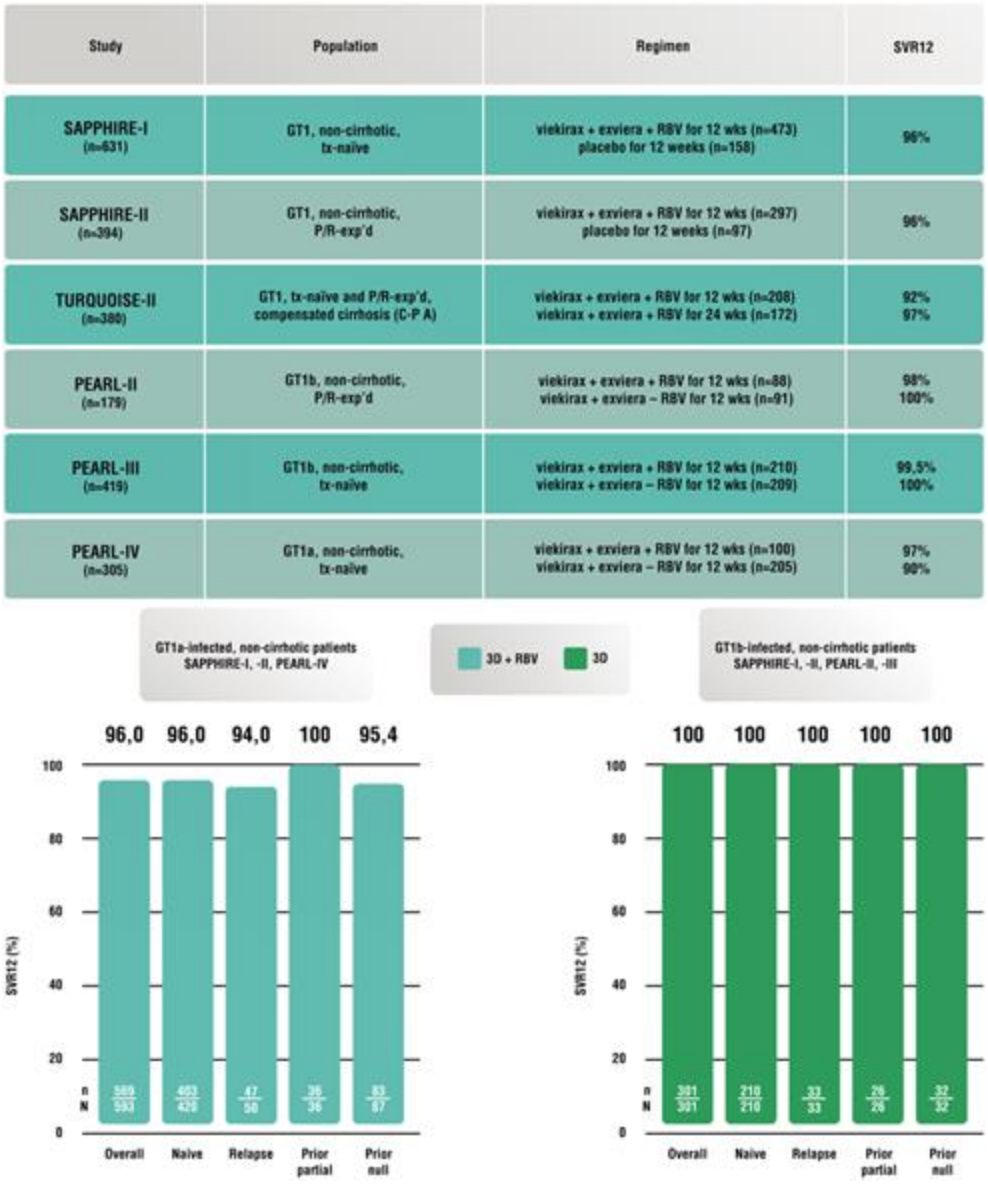
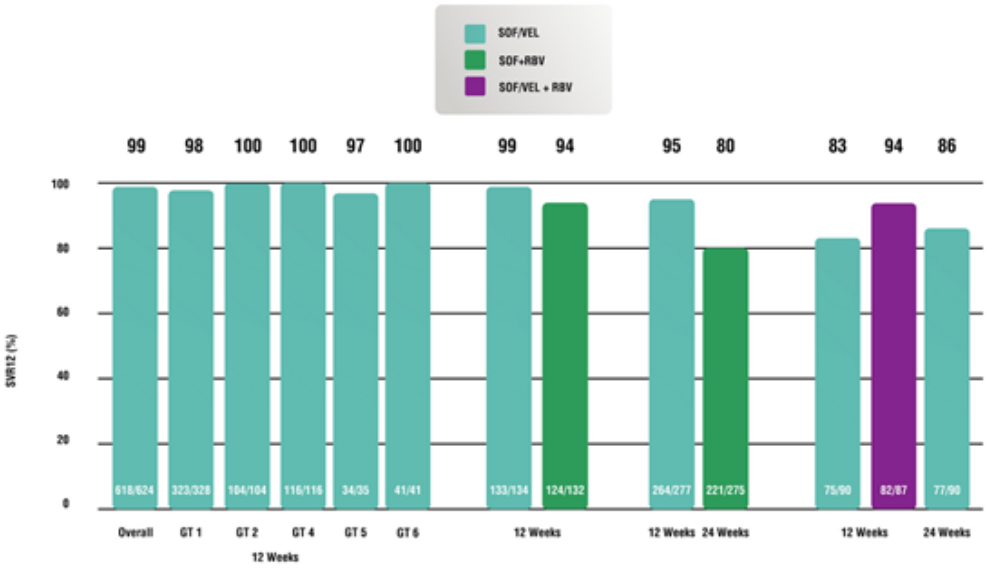


Figure 1.8 Studies results assessing PTV/OBV/DSV treatment among varied HCV genotype 1 patients (103)

The figure shows high efficacy of AbbVie from 6 studies, ranging from 90% to 100% among patients with genotype 1

Sofosbuvir/Velpatasvir

The ASTRAL study assessed the efficacy and safety of 12 weeks treatment with a fixed-dose combination of velpatasvir (VEL) and sofosbuvir (SOF) in a broad range of patients with hepatitis C. The ASTRAL-1, ASTRAL-2, ASTRAL-3, ASTRAL-4 studies recruited varied HCV patients including treatment-naive, treatment-experienced, and including those with compensated cirrhosis.(104, 105) Based on the study, 99% of patients with genotype 1, 2, 4, 5, 6 achieved SVR within 12 weeks (see figure 1.9). Among genotype 3 patients (30% of whom had cirrhosis), the efficacy was still high (95% for SOF/VEL and 80% for SOF +RBV 24 weeks treatment). As for decompensated cirrhosis patients, SOF/VEL regimens had 83% efficacy (SVR12), SOF/VEL + RBV had 94% (SVR12), and SOF/FEL had 86% (SVR24).



Study	Population	N	Treatment	Duration	SVR12 Rates
ASTRAL-1 116 subjects received PBO (0% SVR12)	GT 1, 2, 4, 5, 6 [19% (121/624) cirrhotic]	624	SOF/VEL	12 weeks	99% (618/624) Overall: GT 1: 98% (323/328) GT 2: 100% (104/104) GT 4: 100% (116/116) GT 5: 97% (34/35) GT 6: 100% (41/41)
ASTRAL-2	GT 2 [14% (38/266) cirrhotic]	134	SOF/VEL	12 weeks	99% (133/134)
		132	SOF+RBV	12 weeks	94% (124/132)
ASTRAL-3	GT 3 [30% (163/552) cirrhotic]	277	SOF/VEL	12 weeks	95% (264/277)
		275	SOF+RBV	24 weeks	80% (221/275)
ASTRAL-4	GT 1-6 Decompensated Cirrhosis (CTP B)	90	SOF/VEL	12 weeks	83% (75/90)
		87	SOF/VEL+RBV	12 weeks	94% (82/87)
		90	SOF/VEL	24 weeks	86% (77/90)

Figure 1.9 Studies results assessing SOF/VEL treatment among varied HCV patients (103)

Grazoprevir and Elbasvir (Merck)

Grazoprevir (GZR) phase 3 study (C-Surfer) has already completed and in the modified full analysis set found that 99% of HCV patients (115/116) achieved SVR12.(106) Efficacy was found to be consistently across different subgroups, genotype 1a and 1b, diabetes and haemodialysis. Another study assessed the efficacy of grazoprevir (GZR)/elbasvir (EBR) among 316 patients in the immediate-treatment group (C-EDGE study).(107) Among participants, (299) 95% achieved SVR12, which consists of 92% (144 of 157) in patients with genotype 1a, 99% (129 of 131) in those with genotype 1b, 100% (18 of 18) in those with genotype 4, and 80% (8 of 10) in those with genotype 6. The SVR12 was achieved in 97% (68 of 70) of cirrhotic patients and 94% (231 of 246) of non-cirrhotic patients.

Study	GT	Sample Size	Cirrhosis	Tx History	Comorbidity	Regimen (Weeks)	SVR
C-SURFER	1	237	± Cirrhosis	TN/PR-PTF	CKD 4-5	12, no RBV	94%
C-EDGE TN	1,4,6	421	± Cirrhosis	TN		12, no RBV	95%
C-EDGE TE	1,4,6	420	± Cirrhosis	PR-PTF	±HIV	12 or 16, ±RBV	92-97%
C-EDGE CO-INF-N	1,4,6	218	± Cirrhosis	TN	HIV	12, no RBV	95%
CO-STAR	1,4,6	300	± Cirrhosis	TN	OST, ±HIV	12, no RBV	96%*
C-EDGE H2H	1	250	± Cirrhosis	TN/PR-PTF	±HIV	12, no RBV	
C-EDGE InhBD	1,4,6	300	± Cirrhosis	TN/PR-PTF	InhBD	12, no RBV	

TN: Treatment Naïve
PR-PTF: Failed Prior Peg-IFN/RBV
InhBD = Inherited Blood Disorders
CKD 4-5: Chronic Kidney Disease Grades 4-5 (incl. Hemodialysis)
OST = Opiate Substitution Therapy
BOC = Boceprevir
RBV = Ribavirin

Figure 1.10 Studies results assessing GZR/EBR treatment among varied HCV patients (103)

Taken together, these trials demonstrate a step change in the efficacy of DAA's to treat all genotypes with a much improved side-effect profile in comparison to previous therapies. This creates a new opportunity to substantially reduce the global burden of HCV. However, it is important to recognise that the performance of drugs in trials may not reflect the real-world scenario when patients fail to adhere to treatment, and this is particularly likely among individuals with chaotic lifestyles. In addition, DAA's were only recently introduced so the importance of drug resistance remains to be seen.

1.6 RESISTANCE TO DAA TREATMENT

Despite the high efficacy of new DAA treatment in the context of trials, the rate of therapeutic failure with these DAAs regimens in everyday clinical practice is estimated between 10% and 15%.(108-112) Most of these cases are associated with the presence of drug resistant viral variants, resulting from mutations produced by amino acid substitutions in the target virus protein that reduce viral sensitivity to DAAs which reducing their efficacy (113-115). It is defined as resistance associated variants (RAVs). The existence of the RAVs before treatment does not appear to affect the SVR, thus testing for basal resistance before treatment is not recommended for naive patients, although testing is recommended in patients who have already experienced therapeutic failure with DAAs in order to determine the retreatment strategy.(114, 116)

1.6.1 THE MECHANISM OF DAA TREATMENT RESISTANCE

HCV replication is characterised by a high rate of virus production and high level of genetic diversity in circulating viruses.(117, 118) Therefore, HCV exists with quasispecies-like characteristics, as a complex mixture of genetically distinct but closely related viral populations that constitutes a reservoir for the emergence of resistant strains.(119)

Mutations that prevent antimicrobials working confer a selective advantage when organisms are exposed to that antimicrobial. Such mutations may also damage the ability of the organism to replicate such that they may be at a selective disadvantage in the absence of exposure to antimicrobials. People with hepatitis C may either have resistant strains because they were infected with a resistant strain (initial resistance) or because they acquired resistance through mutation following infection. It should also be noted that viruses can diversify within individuals such that resistant strains are present at low levels which may be below the threshold for detection. For patients with initial resistance, RAVs are present as natural polymorphisms, defined as a RAV present in at least 5% of isolates without drug pressure and may impact DAA-based treatment efficacy. The natural occurrence of resistance variants has been investigated quite a lot for genotype 1, but only very little data have been published for other genotypes. Characterisation of initial resistance is always related to detectability, as viruses can diversify within individuals such that resistant strains are present at low levels which may be below the threshold for detection.(113)

On the other hand, resistance may also be induced by the presence of drugs. When a treatment is given, resistant variants are selected from a heterogeneous quasispecies population. They can replicate more efficiently than others in the presence of a drug and this may have considerable implications for the treatment outcome. The first mechanism of resistance to DAA is related to development of amino acid mutations in the DAA target regions that reduce drug activity by means of conformational changes, lower binding affinity, or steric hindrance that blocks viral proteins in an inactive form.(120)

Unlike other chronic viral infections such as hepatitis B virus (HBV) and human immunodeficiency virus (HIV), where it required long life of drugs by controlling the level of viral load in the blood, the aim of HCV treatment is viral eradication by pressing the level of the virus very low (which is observed already spontaneously in a significant proportion of patients 20–40%).(121) While for some infected patients, weak antivirals and short-term treatments are sufficient, others require combination therapies with several highly active antivirals for longer durations. HCV genotype and stage of liver fibrosis have been found to be associated with virologic treatment response. Thus, the choice of DAA regimens, duration of therapy, and use of ribavirin depends on multiple viral and host factors, including HCV genotype, the detection of RAVs, prior treatment experience, and presence of cirrhosis.(113)

When patients are exposed to antimicrobials, strains that are resistant to that antimicrobial have a selective advantage and can become the dominant strain leading to reductions in treatment effectiveness or treatment failure. Antimicrobials that rapidly reduce pathogen load provide less opportunity for selection of resistance than antimicrobials with slower action. Adequate concentrations of antimicrobials need to be achieved and sustained to ensure rapid and sustained reductions in pathogen load, thus adherence and regular dosing can be key to preventing emergence of resistance.

Use of multiple antimicrobials together reduces the chances of resistance being selected since if the organism is resistant to one, it remains likely to be susceptible to the other agents used. Thus chronic infections (such as tuberculosis and HIV) are treated with multiple agents to reduce the development of resistance. Experiences in these infections have shown that despite, this, over time multidrug resistant strains can emerge particularly when treatment is erratic. Although the development of resistance has not been a major problem with the latest generation of hepatitis C antivirals, it is possible that as treatment is rolled out more widely to groups who

may have previously been considered to be too chaotic to treat, that resistance may start to become an increasing problem. There is therefore a need for surveillance of resistance to identify emerging problems, inform treatment regimens and potentially identify groups who need more intensive support to complete treatment.

There are several factors that are associated with the emergence of DAAs resistance, including (i) virus dependent factors, e.g. the quality or type of nucleotide alteration generating the resistance associated with the mutation (122, 123), HCV subtypes (122, 124), and the presence of variants associated with basal resistance to NS5A (only patients with cirrhosis), the presence of the Q80K polymorphism (genotype 1a patients receiving regimens with simeprevir) (114); (ii) pharmacologic factors, pharmacokinetics of the antiviral agent, number of mutations required to confer resistance to a particular drug, drug exposure, short-term regimens, poor adherence to treatment, and not giving ribavirin (114); (iii) host-dependent factors, including immune response, the degree of liver fibrosis at the start of treatment, male gender, the IL28B non CC genotype, the cirrhotic state, null responders to previous treatment with PEG-interferon/ribavirin (PEG/ RBV), and previous failure to multiple DAAs (114, 122).

The best way to prevent the appearance of resistant variants is to achieve rapid and deep viral suppression with the first treatment, using a combination of various drugs that have a potent antiviral effect with high genetic barrier to resistance.(114) It is recommended to give retreatment using regimens of two or three DAAs, including one with a high genetic barrier (sofosbuvir) and without cross-resistance, during 12 week accompanied by ribavirin, or 24 week if it is not possible to give ribavirin.(114, 116) Resistance to DAAs treatment can limit the efficacy of the therapy and affect the outcome of the treatments, thus optimising the treatment strategies is important to avoid treatment failures in the future.

1.7 Feasibility of Hepatitis C Elimination

Not long ago, hepatitis C elimination seemed implausible, but major advances in HCV treatment bring hope that HCV elimination might be feasible in the near future. Many studies have proven the high efficacy of DAAs ranging between 90% and 95%, their good tolerability, safety profile and applicability, creating the opportunity to cure individuals in many cases, reducing the risk of onwards transmission bringing a positive impact on the natural history of the infection and the associated costs.(116, 125, 126) To evaluate the impact of DAA treatment on a larger scale,

many modelling studies have demonstrated the potential effectiveness of HCV treatment among hepatitis C patients, including those in 'hard to reach' group such as PWID (127-132). In this context WHO has announced targets or elimination of hepatitis B and C, including a 65% reduction in mortality and 90% reduction in new chronic infections by 2030.(11)

Despite the advantages mentioned above, there are a range of barriers to achieving these aims. Two major barriers are the cost of the new treatments and the challenge of accessing high risk populations to instigate screening and treatment. If resistance becomes an increasing problem then this will also threaten the feasibility of elimination.

Cost of drugs

DAAs treatments were initially very expensive, ranging from $25,000 in Spain to $54,000 in the UK and $51,000-$84,000 in the USA for a 12 week course of treatment.(133) In England, for example, treatment with DAA was restricted to approximately 10,000 patients in 2016-2017 due to the large numbers of potential patients and the very high aggregate cost of the treatments involved.(134) However, the cost of the treatment is falling and is now less than £10,000 for a course of treatment in the UK.(135) Specific details regarding the pricing of DAA's are not publicly available due to commercially confidential pricing agreements. Nevertheless, it is important to optimise the use of these drugs and the timing of treatment in settings where cost represents a barrier to treatment. This is particularly pertinent in low-income countries where iatrogenic infection is the commonest source of HCV infection and infected individuals may pose a smaller risk of onward transmission in comparison to high-income countries where intravenous drug use is the major source of HCV infection. As a substantial proportion of individuals will clear HCV spontaneously, there may be an argument to delay treatment of low risk individuals in low-income countries, to maximise the effectiveness of available resources. In chapter 2, I consider this question in more detail by undertaking a systematic literature review and meta-analysis in which I estimate the true rate of HCV spontaneous clearance and the average time from infection to clearance. I also identify factors that predict clearance including demographic, clinical, and behavioural factors as well as the host genetic factors (chapter 2 and 3).

Who needs treatment? Estimating the burden of HCV in high risk populations

The burden of HCV is concentrated in high risk populations. Targeting DAAs treatment to these populations is pivotal to reduce the burden of hepatitis C in the population. However, these population groups are difficult to access and often don't engage with services, thus they often do not access treatment. Therefore, we need data to demonstrate the importance of these high-risk groups in disease transmission and provide better estimate of HCV prevalence in these vulnerable populations which could support the development of targeted strategies to reduce HCV infection and transmission in these groups. Using the data from Find & Treat study (a cross-sectional study which enrolled individuals from homeless shelters, drug treatment services, and a prison), I estimate the prevalence of HCV among vulnerable population such as PWID, homeless, and prisoners in London (chapter 5).

Scaling up treatment with DAA's to meet the targets for elimination

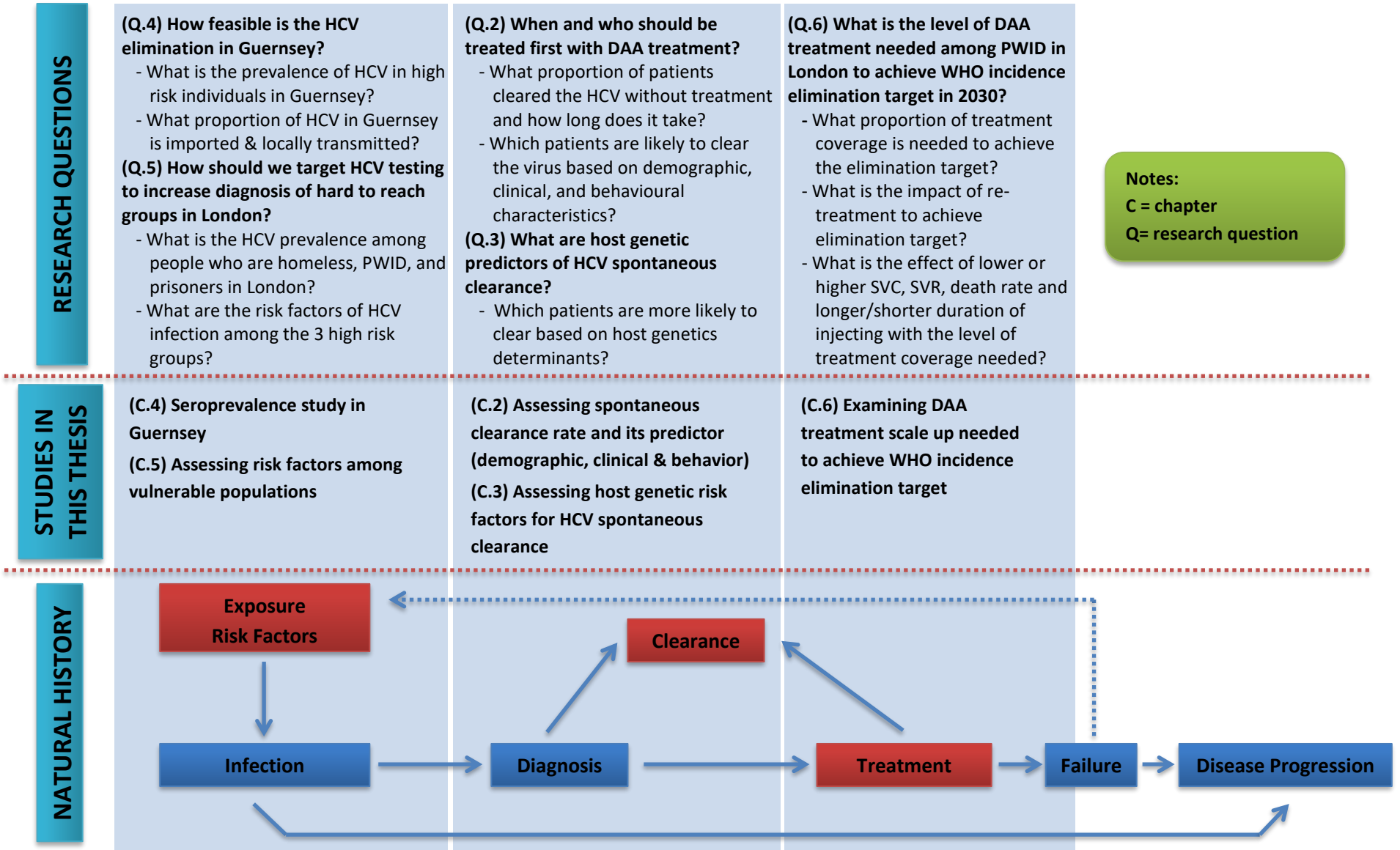
Finally, to consider the policy implications of working towards HCV elimination, I estimated what DAA treatment scale-up would be required among chronically infected PWID in London in order to achieve WHO incidence elimination goal by 2030. I have conducted modelling analysis to look at this issue and presented the results in chapter 6.

In chapter 4, I present the analysis plan for a seroprevalence study of HCV in Guernsey with linked genomic analysis (chapter 4) which I designed and obtained ethical approval for. The aim of this study was to assess the feasibility of hepatitis C elimination in the island by estimating the prevalence of HCV among high risk individuals in Guernsey and examining proportion of HCV in Guernsey which is imported and locally transmitted. However, I was unable to undertake the study because the local PI in Guernsey, who was initially highly supportive, decided not to continue to support the work due to competing priorities. Without her input, it was impossible to undertake recruitment or collection of samples in Guernsey. I have included this chapter to demonstrate the work that I undertook to develop a prospective study.

In my final chapter, I discuss the findings of my thesis and make recommendations for control and for future research. Overall, this thesis aims to provide insights into the feasibility of Hepatitis C elimination, identify strategies that might support this goal and improve the understanding of epidemiology of hepatitis C.

1.8 THE OVERALL OVERVIEW OF THE THESIS

Below is the overall framework of the thesis, describing the overview of several research questions addressed during my PhD.


RESEARCH QUESTIONS
(Q.4) How feasible is the HCV elimination in Guernsey?
- What is the prevalence of HCV in high risk individuals in Guernsey?
- What proportion of HCV in Guernsey is imported & locally transmitted?
(Q.5) How should we target HCV testing to increase diagnosis of hard to reach groups in London?
- What is the HCV prevalence among people who are homeless, PWID, and prisoners in London?
- What are the risk factors of HCV infection among the 3 high risk groups?
(Q.2) When and who should be treated first with DAA treatment?
- What proportion of patients cleared the HCV without treatment and how long does it take?
- Which patients are likely to clear the virus based on demographic, clinical, and behavioural characteristics?
(Q.3) What are host genetic predictors of HCV spontaneous clearance?
- Which patients are more likely to clear based on host genetics determinants?
(Q.6) What is the level of DAA treatment needed among PWID in London to achieve WHO incidence elimination target in 2030?
- What proportion of treatment coverage is needed to achieve the elimination target?
- What is the impact of re-treatment to achieve elimination target?
- What is the effect of lower or higher SVC, SVR, death rate and longer/shorter duration of injecting with the level of treatment coverage needed?
Notes:
C = chapter
Q= research question
STUDIES IN THIS THESIS
(C.4) Seroprevalence study in Guernsey
(C.5) Assessing risk factors among vulnerable populations
(C.2) Assessing spontaneous clearance rate and its predictor (demographic, clinical & behavior)
(C.3) Assessing host genetic risk factors for HCV spontaneous clearance
(C.6) Examining DAA treatment scale up needed to achieve WHO incidence elimination target
NATURAL HISTORY
Exposure Risk Factors
Infection
Diagnosis
Clearance
Treatment
Failure
Disease Progression

1.9 MY ROLE IN THIS THESIS

All work was conducted with advice from my supervisory team. My roles in this thesis included undertaking narrative reviews to inform the introduction and discussion sections. I developed the study protocols for the literature reviews of spontaneous viral clearance. I conducted the literature search, performed the initial screening and full text review, carried out data extraction, undertook meta-analysis and drafted research papers for publication. This work is presented in chapter 2 and 3. I also prepared the research protocols, data collection instruments (questionnaire, information sheet, clinical assessment form) and the submissions for data protection, sponsorship, and ethical approvals for the seroprevalence study in Guernsey presented in chapter 4. The London study of hepatitis C prevalence and risk factors was based on secondary analysis of a previously conducted study. This study had focussed on prevalence of latent tuberculosis infection but had also collected data on Hepatitis C. I designed the analysis plan, conducted the analysis and led on preparing a manuscript for publication (chapter 5). The primary data collection in chapter 5 was conducted by others but I worked alongside them to understand the methodologies involved and to write these up and interpret the findings. I designed the modelling study with support from my supervisors and a collaborator (Dr. Natasha Martin, an infectious disease modeller from San Diego University), presented in chapter 6. I developed the model structure, performed literature searches for parameters, ran the analysis conducted model calibration, and performed sensitivity analysis of the study. The work done by other colleagues is clearly acknowledged throughout the thesis.

Key Points:

- Hepatitis C is a major cause of chronic liver disease with an estimated viraemic (HCV-RNA positive) global prevalence of 1.1%, corresponding to 80 million of HCV infections.
- HCV genotype 1 is the most prevalent type of hepatitis C infection around the globe, accounting for almost half of total cases, followed by genotype 3, and 2, 4, and 6.
- Genotype 1 was mostly dominated in North America, whereas genotype 2 was most common in West Africa, genotype 3 was predominantly in South Asia and parts of Scandinavia, genotype 4 was prevalent in Central and North Africa, genotype 5 particularly found in South Africa, and genotype 6 mostly common in Southeast Asia.
- Hepatitis C has different main routes of transmission between countries where the majority risk of infection in developed nations was PWID while blood transfusion and contaminated injection adding the risk of transmission in developing nations.
- Ascertaining the HCV prevalence, the genotype distribution and risk factors is challenging. The varied results might be caused by different sampling frames, selection bias, different measurement of exposure and outcome, etc.
- Hepatitis C treatment had undergone major development which resulted on fewer side effects, shorter duration and can work against every genotype.
- In 2016, World Health Organization announced hepatitis B and C elimination as a public health targets, including a 65% reduction in mortality and 90% reduction in new chronic infections by 2030. However, there are some barriers to achieve this target, including cost of the drugs (need to think who should be treated first considering the possibility of spontaneous clearance) and who needs the treatment (need to estimate the burden of HCV among high risk groups)
- Therefore, the aim of this thesis is to improve our understanding of the epidemiology of hepatitis C in order to inform public health strategies working towards HCV elimination.

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2. Understanding Clinical, Demographic, and Behavioural Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Systematic Review and Meta-Analysis

Chapter's Aim:

The aim of this chapter is to estimate the rate of hepatitis C spontaneous viral clearance (SVC) and determine factors associated with clearance. I outline evidence about the relationship between HCV SVC and demographic factors (sex, age, ethnicity), clinical factors (symptomatic infection, viral co-infection and virus genotype), and behavioral factors (injection drug use and alcohol consumption).

2.1 Background

During the acute phase of infection, Hepatitis C virus (HCV) may completely resolve without treatment (spontaneous clearance) which is indicated by the disappearance of HCV RNA in the serum. The rate of HCV Spontaneous Viral Clearance (SVC) varies between studies, but it is believed to range from 20% to 30%. Factors that may predict clearance include female sex (1-4), ethnicity, immune responses (e.g. innate immune response – interferon stimulated genes, natural killer cells; cellular immune response – CD4+ T cells, CD8+ T cells; humoral immune response) (5-7), and host genetics (8, 9). Polymorphisms in interleukin-28 (IL28B) gene region are recognised as the strongest genetic factor associated with clearance.(8-10)

New advances in treatment for HCV which offer >90% effectiveness create the opportunity to eliminate hepatitis C in the future. However, the treatments are expensive, therefore estimating when and to whom treatment should be given is an important consideration for policy makers in the context of limited funding for treatment. In England, for example, treatment with DAA was restricted to approximately 10,000 patients in 2016-2017 who were in contact with treatment services due to the large numbers of potential patients and the very high aggregate cost of the treatment.(12) However, the major burden of HCV is in People Who Inject Drugs (PWID). Many of these individuals are not in contact with treatment services and consequently would have been ineligible for treatment. In this scenario (working on the assumption that universal access to DAA's is unlikely to become available in the short-term), knowledge of variation between population sub-groups

in terms of the natural history of infection and the prevalence of spontaneous clearance could inform policy decisions on which individuals should be prioritised for treatment with DAA's. This information would arguably be most relevant for low-income countries where cost represents a major barrier to treatment. In addition, iatrogenic transmission is the dominant route of HCV acquisition in low-income countries consequently delaying treatment to low-risk individuals with HCV may pose less risk of onward transmission in comparison with high income countries where most cases are infected through injection drug use (see figure 1.5 in chapter 1).

A systematic review published in 2006 indicated that approximately 26% of patients who were infected with HCV will spontaneously clear the virus.(1) However, there was significant heterogeneity in the study population which may have affected the clearance rate. Furthermore, the study was conducted over 10 years ago, so it is likely that a wide range of further studies have been published. In this chapter, I present a literature review which aims to obtain a more precise estimate of the proportion of patients who clear HCV spontaneously and to better characterise the factors which predict viral clearance. Better understanding of the time course and predictors of clearance will inform clinical decision making regarding the timing of antiviral treatment. It has also been hypothesised that patients with lower levels of spontaneous viral clearance may also be harder to treat and more likely to develop resistance.(11) Therefore knowledge of which patients have low spontaneous clearance rates may also inform strategies to identify and support such patients to be successfully treated.

2.2 METHODOLOGY

2.2.1 TYPE OF STUDIES

Any studies that reported the rate or proportion of spontaneous viral clearance in hepatitis C infected patients AND/OR investigated factors associated with clearance were eligible for inclusion in the analysis. To estimate the rate of HCV spontaneous clearance, I only included studies which recorded the initial time of infection with a minimum of two follow up HCV RNA tests after the initial diagnosis. SVC was defined as a minimum of two negative HCV RNA tests following diagnosis. Acute HCV infection was defined as one or more of the following criteria 1) HCV antibody negative and HCV RNA positive in the serum 2) HCV antibody and RNA were positive with a documented HCV RNA negative test in the past year 3) HCV antibody was positive with clinical evidence of symptomatic infection (jaundice, ALT

elevation >400U/L and/or history of high risk exposure within 3 months of clinical presentations).

The clearance of HCV infection was determined based on HCV RNA assessment where there are no detectable concentrations of HCV RNA in the blood. The factors associated with the clearance were divided into demographic factors (age, sex, race) clinical factors (symptomatic infection, genotype, viral co-infection), and behavioural factors (alcohol consumption and PWID). Alcohol consumption was divided into non-alcohol drinkers compare to alcohol drinkers or people with history of drinking excess alcohol or history of alcohol abuse.

2.2.2 INCLUSION AND EXCLUSION CRITERIA

1) *For studies reporting HCV viral clearance rate care was taken to only include studies with known timing of infection.*

Inclusion Criteria:

- Longitudinal cohort study with a minimum of one-year follow up which reported the initial time of infection (estimated as the midpoint between the last negative HCV antibody test result and the first evidence of HCV infection)
- HCV RNA was measured at baseline
- Had at least two follow up HCV RNA assessments after the initial diagnosis
- Individuals were untreated during follow up
- Duration of follow up for study population was clear

Exclusion Criteria:

- Case studies or reviews
- Studies were part of another study included in the analysis
- Studies that did not report HCV RNA
- Studies that reported clearance only among children
- Studies that were restricted to specific rare groups e.g. patients with lichen planus

2) *For studies that reported factors associated with HCV clearance the timing of infection was considered to be less important so I also included studies with unknown time of infection.*

Inclusion Criteria:

- HCV RNA measured at least once and reported for all study subjects
- Factors associated with HCV clearance were assessed

- Individual untreated for acute HCV infection

Exclusion Criteria:

- Case studies or reviews
- Studies were part of another study included in the analysis
- Study population was less than 40 participants
- Studies that were restricted to a specific rare group e.g. patients with lichen planus
- Studies that reported clearance only among children

In studies that include both treated and untreated individuals, only untreated individuals were included in the analysis.

2.2.3 SEARCH STRATEGY

I conducted systematic searches for any studies that reported at least one of the outcomes of interest. Only studies that were published after 1994 up to June 2015 and only English language publications were included in this review. I searched the following electronic databases from PubMed, Ovid Medline, and Ovid Embase, for published, unpublished or ongoing studies.

2.2.4 SEARCH TERMS

Concept 1: hepatitis C or HCV

Concept 2: natural history or clearance or vir* negativ*

Using relevant MeSH terms where appropriate

2.2.5 SELECTION OF STUDIES

The initial screening was conducted from publication titles and abstracts only. It was done by two independent reviewers to assess publications' relevance for the full-text review. I only included the most recent publication if hepatitis C reports were provided from the same place(s) and same participants in several years. If I identified articles where the same participants may have contributed data to multiple studies I contacted the study author where possible to clarify the study population. If typographical errors and/or calculation errors were observed in articles, reports were recalculated and clarified with authors when possible. Next, two independent researchers reviewed the full text of relevant articles against the pre-defined inclusion and exclusion criteria. Any discrepancies were resolved through discussion and/or with a third reviewer. The records of included and excluded publications were preserved for audit purposes, indicating reasons for any

exclusion. I performed and reported this systematic review using the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guideline. A protocol for this review can be accessed at http://www.crd.york.ac.uk/PROSPERO/ with registration number: CRD42015023499.

2.2.6 DATA EXTRACTION:

A pilot of the data extraction form was conducted using 20 papers from a range of study designs. The extraction form was amended following the pilot evaluation. The data extraction form was used to extract the relevant data from each included study, such as:

(1) study characteristics: setting, study design, method of data-analysis, duration of follow up, country, method of recruitment;
(2) participants: study population, number of participants, research participants characteristics such as age and gender, mode of HCV acquisition;
(3) outcomes: rate or proportion of HCV spontaneous clearance, factors associated with clearance

Given the scale of this review, I present results for the association between demographic, clinical and behavioural factors and spontaneous clearance in this chapter. The results of the host genetic factors review are available in the next chapter.

2.2.7 DATA ANALYSIS

For the subset of studies in which the time of infection was known precisely, I estimated the proportion of patients achieving HCV clearance at 3, 6, 12 and 24 months following the initial infection. For this analysis, I plotted the relationship between calendar time and the proportion of patients having SVC and estimated the HCV clearance rate and 95% confidence interval at 3, 6, 12, and 24 months after the initial infection.

To examine the relationship between demographic, clinical, and behavioural factors and HCV spontaneous clearance, I calculated odds ratios comparing the odds of clearance in patients with each risk factor to the odds of clearance in patients who lacked each risk factor. I included demographic factors (gender, age, and ethnicity); clinical factors (viral co-infection, HCV genotype, and symptomatic infection); and behavioural factors (alcohol consumption and PWID). I used meta-analysis to summarise the relationship between each risk factor and outcome. Odds Ratios were calculated and forest plots generated using Comprehensive Meta-Analysis

(CMA) version 3.0. I investigated heterogeneity using I^2 and assessed publication bias using a funnel plot of proportions of clearance against the study size. If there was evidence of heterogeneity (I^2 >50%), I used random effect models whereas fixed effect models were used when there was no evidence of heterogeneity. Sensitivity analysis was done by assessing the odds ratios restricted for studies with a minimum of 12 months follow up.

2.3 RESULTS

2.3.1 ARTICLE SCREENING

After excluding duplicates, I retrieved 9,357 publications from Ovid Embase, Pubmed, and Ovid Medline. By conducting an initial screening of titles and abstracts, there were 483 publications included for full-text review. A total of 183 publications were excluded due to: treatment after diagnosis (treatment-induced clearance), study design, overlap with a different study, insufficient information to estimate clearance. I identified 52 studies which assessed spontaneous clearance where six of those recorded the precise time of infection for the assessment of spontaneous clearance. Forty-three studies met the inclusion criteria for assessing demographic, clinical and behavioural risk factors and 86 assessed host genetic factors for clearance, representing a total of 53,185 individuals. The article screening process following PRISMA diagram can be seen Figure 2.1.

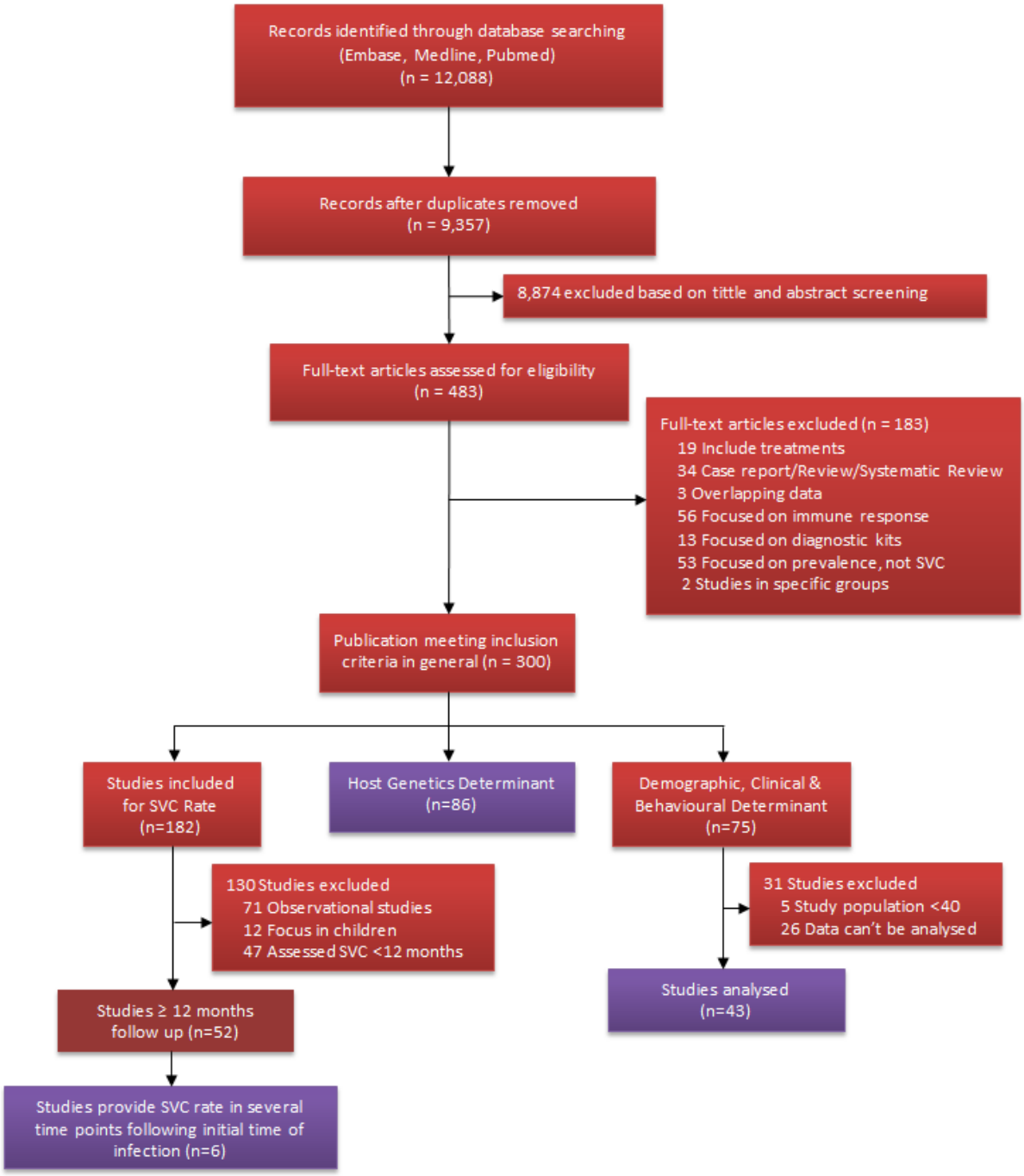


Figure 2.1 Article Screening Following PRISMA Diagram

2.3.2 CHARACTERISTIC OF STUDIES INCLUDED

Of the 43 studies included in the analysis, there were 27 prospective and 4 retrospective cohort studies, 6 case-control and 6 cross-sectional studies. Study participants were recruited from hospitals or related health centres (18 studies) and included patients who were transfusion dependent (4 studies), PWID (8 studies), patients in the community (2 studies), HIV positive patients (6 studies), and blood donors (5 studies). A total of 25 studies were published between 2010 and 2015. The majority of studies were conducted in North America (15), followed by European countries (9), Middle East (7), Asia (6), Australia (2), South America (1), and 3 multi-national studies (see figure 2.2). Characteristics of included publications are described in Table 2.1 and Table 2.2.

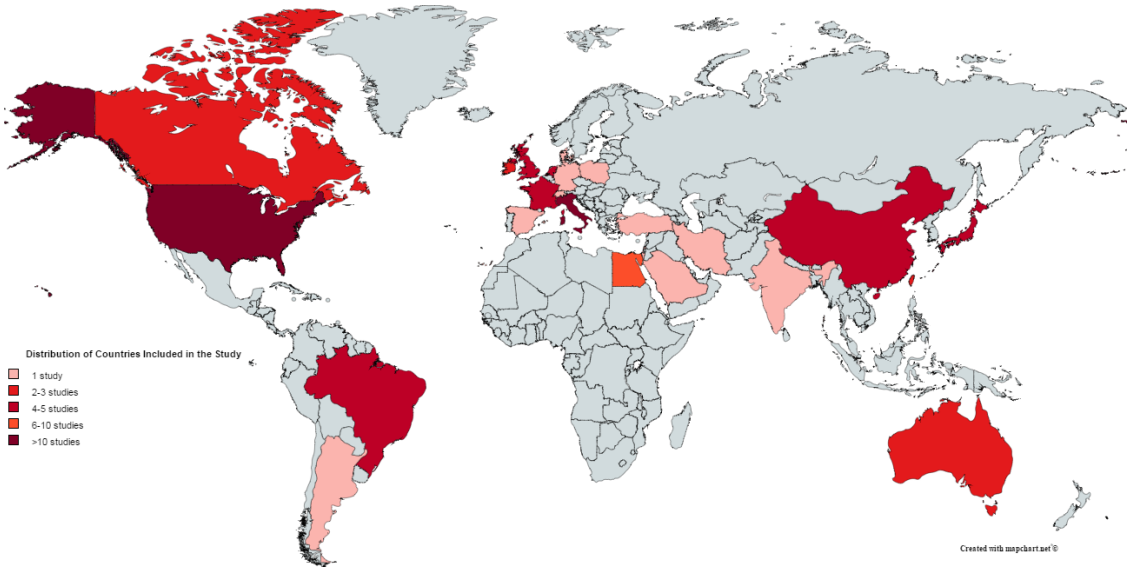


Figure 2.2 Distribution of Countries Included in the Analysis

Table 2.1 Characteristic of studies included in HCV Spontaneous Viral Clearance Rate (SVC) Analysis

First Author	Country	Year	Study Population	M/F	Age*	Σ HCV (+)	Σ Clearance	Proportion	95% CI
Noha Sharaf Eldin	Egypt	2008	HCV infected patients	69/48	n/a	117	51	43.59	34.55-53.06
Marianne Jauncey	Australia	2004	HCV infected PWID	27/30	n/a	57	24	42.11	29.4-55.88
Ximenez	Brazil	2010	HCV infected patients	25/40	45.7±12.4 (Range 20-77)	65	29	44.62	33.1-56.8
Jason Grebely	Australia, Canada, Netherland, USA	2014	HCV infected patients	404/228	n/a	632	173	27.4	24.0-31.0
Chia C. Wang	USA	2007	HCV infected patients	35/32	Median 31 (Range 17-82)	67	15	22.39	13.47-33.90
J. Tilman Gerlach	Germany	2003	HCV infected patients	25/35	n/a	60	24	44.4	31.90-57.80

*Mean of age, otherwise stated in the table

n/a = Not available

Table 2.2 Characteristic of Studies Included Assessing Demographic, Clinical and Behavioural Factors Associated with HCV Spontaneous Clearance

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	Proportion	95% CI
E.C. Seaberg	USA	2013	HCV infected PWID	528/0	Median 33.5 (Range 17-70)	528	118	22.35	18.91-26.20
Isabelle Morard	Switzerland	2014	HCV infected patients	886/564		1450	160	11.03	9.49-12.78
Chia C. Wang	USA	2007	HCV infected patients	35/32	Median 31	67	15	22.39	13.47-34.52

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	Proportion	95% CI
					(Range 17-82)				
Nathan J. Shores	USA, Spain, Italy	2008	HCV infected & HIV (+)	572/197	Median 41 (Range 37-45)	769	102	13.26	10.98-15.91
Vincent Soriano	Europe, Israel & Argentina	2008	HCV infected & HIV (+)	1348/592	Median 37.2	1940	444	22.89	21.95-24.84
Leslie H. Tobler	USA	2010	HCV infected Blood donors			302	100	33.11	27.89-38.77
Louise Nygaard Clausen	Denmark	2011	HIV (+)	215/112	Median 36 (Range 30-41)	327	76	23.24	18.85-28.27
K.J. Rolfe	UK	2011	HCV infected patients	202/119		321	102	31.78	26.78-37.22
Charlotte H.B.S. van der Berg	Netherland	2011	PWID	62/44	Median 28.5	106	35	33.02	24.37-42.91
Jason Grebely	Australia, Canada, USA, Netherland	2014	HCV infected patients	404/228		632	173	27.37	23.96-31.06
Edward L. Murphy	USA	2010	Blood donors	415/279		695	179	25.76	22.58-29.21
Kimberly Page	USA	2009	PWID	61/34		95	20	21.05	13.63-30.87
Jason Grebely	Canada	2007	HCV infected patients			762	179	23.49	20.56-26.70
Marianne Jauncey	Australia	2004	PWID	27/30		57	24	42.11	29.40-55.88
Nasheed Moqueet	Canada	2015	HIV (+)	367/174		541	79	14.60	11.79-17.92
Dimpy P. Shah	USA	2012	PWID	337/83		420	62	14.76	11.58-18.60

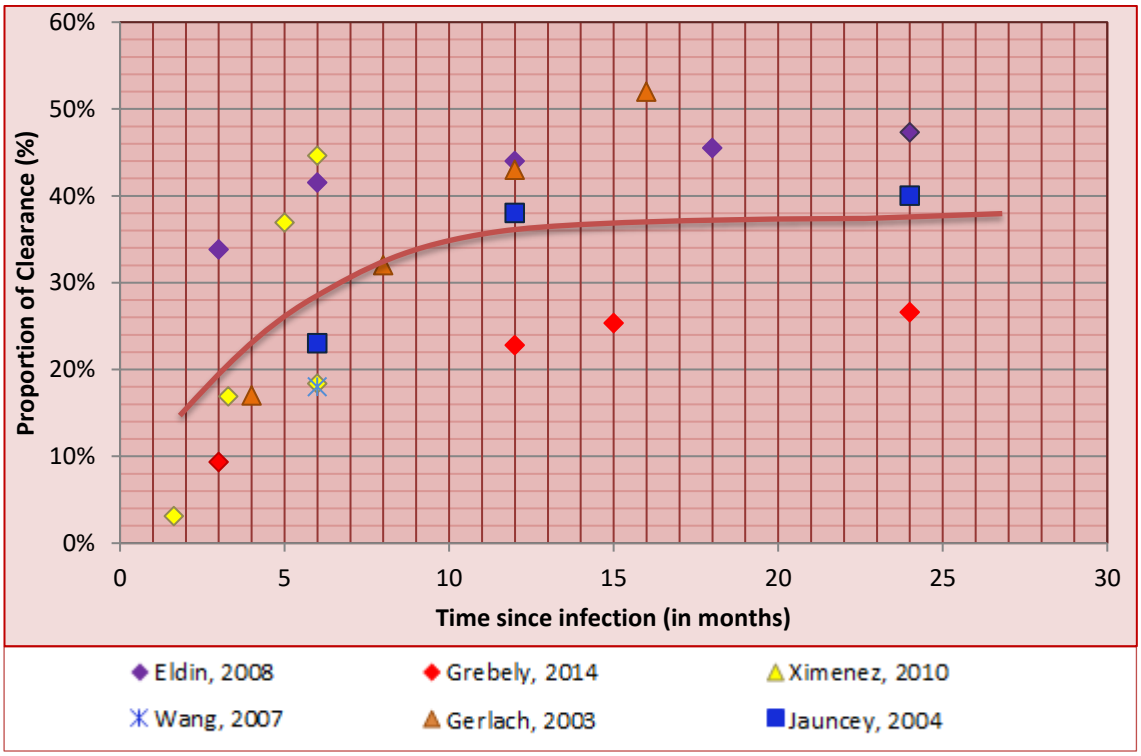
First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	Proportion	95% CI
Arthur Y. Kim	USA	2011	HCV infected patients	131/215		346	66	19.08	15.16-23.71
Monika Sarkar	USA	2013	HCV infected patients	0/897	39.5±6.5	897	168	18.73	21.00-27.49
David L. Thomas	USA	2000	PWID		Median 34 (Range 29.8-38.4)	919	90	9.79	7.98-11.94
G.H. Ibrahim	Egypt	2013	HCV infected patients			115	22	19.13	12.62-27.75
Yuan Dong	China	2015	HIV (+)		Median 34	432	97	22.45	18.66-26.74
Michael P. Busch	USA	2006	Blood donors	1261/794		2055	402	19.56	17.88-21.36
Lia L. Lewis-Ximenez	Brazil	2010	HCV infected patients	25/40	45.7±12.4 (Range 20-77)	65	29	44.62	32.47-57.41
Syczewska	Poland	2004	HCV infected patients	41/36		77	23	29.87	20.25-41.53
Patrick G. Quinn	USA	1999	HCV infected patients	155/103		258	44	17.05	12.78-22.33
Mingdong Zhang	USA	2006	Transfusion Dependent patients	671/41		712	192	26.97	23.77-30.42
Kohei Oda	Japan	2014	HCV infected patients	167/335	73 (Range 37-97)	502	149	29.68	25.76-33.92
T. Santantonio	Italy	2006	HCV infected patients	134/69	37.5 (Range 17-83)	203	73	35.96	29.44-43.02
S Keating	Ireland	2005	PWID	342/154	28.75 ±6.35	496	191	38.51	34.23-42.97
I Bakr	Egypt	2006	Sero incident cases	511/399		910	350	38.46	35.30-41.72
Hui-Ying Rao	China	2012	Blood donors	156/192	53.7±7.4	348	74	21.26	17.16-26.01
L. Alric	France	2000	HCV infected patients	171/174		345	63	18.26	14.41-22.83
Hossein Poustchi	Iran	2011	Sero incident cases	162/85		247	95	38.46	32.42-44.86
Madiha Mohamed El-Attar	Egypt	2010	HCV infected patients	115/85	46.5±13.6 (Range 12-75)	200	35	17.50	12.64-23.64
Sanaa M. Kamal	Egypt	2014	HCV infected patients	69/67		136	48	35.29	27.42-44.00

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	Proportion	95% CI
Gamal Esmat	Egypt	2011	HCV infected patients	53/43	5.9±2.4	96	31	32.29	23.31-42.71
J. Tilman Gerlach	Germany	2003	HCV infected patients	25/35	Range 17-63	60	24	40.00	27.83-53.45
Rebecca J Garten	China	2008	PWID	334/13	27.4±5.6	347	30	8.65	6.01-12.24
Enea Spada	Italy	2013	HCV infected patients	39/17	Median 31 (Range 19-78)	56	18	32.14	20.65-46.09
Noha Sharaf Eldin	Egypt	2008	HCV infected patients	69/48		117	51	43.59	34.55-33.06
H-Y Rao	China	2012	Blood donors	163/213	53.2±8	376	80	21.28	17.32-25.84
Barbara A. Piasecki	USA	2004	HCV infected patients	496/0		496	203	40.93	36.59-45.41
Ming-Lung Yu	Taiwan	2014	Transfusion Dependent patients	115/172	62±11.6	287	73	25.44	20.59-30.96

*Mean of age, otherwise stated in the table

2.3.3 HCV Spontaneous Clearance Rate

After assessing the inclusion and exclusion criteria, there were a total of 182 studies reporting spontaneous clearance rate. **To ensure less biased estimates of clearance, I restricted** the analysis to 6 studies (12-17) which provide information of **clearance at specific time points following** the initial infection, representing a total of 998 individuals. The restriction was very important since many previous studies did not consider the real time of initial infection, preventing the timing of clearance from being ascertained and potentially introducing bias. For example, inclusion of patients with a comparatively longer duration of follow-up will underestimate SVC because the rate of SVC declines over time. Meta-analysis revealed the proportion of spontaneous viral clearance to be 19.8% (95% CI: 2.6-47.5%), 27.9% (95% CI: 17.2-41.8%), 36.1% (95% CI: 23.5-50.9%), and 37.1% (95% CI: 23.7-52.8%) within 3, 6, 12, and 24 months after infection respectively (Figure 2.3). The forest plot of meta-analysis output can be seen in Appendix 1.



Months after infection	I^2	SVC Rate	Lower CI	Upper CI
3 months	96.966	19.8	2.6	47.5
6 months	91.261	27.9	17.2	41.8
12 months	90.616	36.1	23.5	50.9
24 months	90.708	37.1	23.7	52.8

Figure 2.3 Rate of Spontaneous Clearance within 3, 6, 12, and 24 Months after Infection

After fitting a non-linear regression line, **I found that those who had not spontaneously cleared by 12 months were unlikely to do so.** Therefore, I restricted my subsequent analysis of the rate of SVC to studies which included at least 12 months of follow up. This restriction was applied to any longitudinal studies who could not clarify when the time of initial infection happened. Finally, only 52 longitudinal studies were included in the analysis, representing a total of 11,807 study population. After extrapolating data on the clearance proportion, the overall rate of HCV spontaneous clearance in studies with at least 12 months of follow up was 28.2% (95% CI: 24.8-31.8%).

2.3.4 Demographic, Clinical, and Behavioural Factors Associated with Clearance

Forty-three studies (12-54) included in the analysis assessed demographic, clinical and behavioural factors associated with HCV clearance, representing a total of 20,110 individuals. The following groups were significantly less likely than others to spontaneously clear the hepatitis C virus: males (OR=0.68, 95% CI: 0.59-0.81), those with asymptomatic infection (OR=0.38, 95% CI: 0.27-0.55), black race (OR=0.38, 95% CI: 0.20-0.75), older adults (age ≥45 years compared to age < 45, OR=0.52, 95% CI: 0.64-0.97), those with HIV co-infection (OR=0.50, 95% CI: 0.37-0.67), those without hepatitis B co-infection (OR=0.24, 95% CI: 0.19-0.32), patients with non-genotype 1 infection (OR=0.63, 95% CI: 0.45-0.89), non-aboriginal groups (OR=0.47, 95% CI: 0.36-0.62), and those with excess alcohol use (OR=0.67, 95% CI: 0.47-0.95) and those with a history of injecting drug use (OR=0.59, 95% CI: 0.37-0.93). I show forest plots for these associations in Figures 2.4, 2.5, and 2.6. When I restricted the analysis of risk factors to patients with a minimum of 12 months follow-up I found similar associations (see Appendix 2).

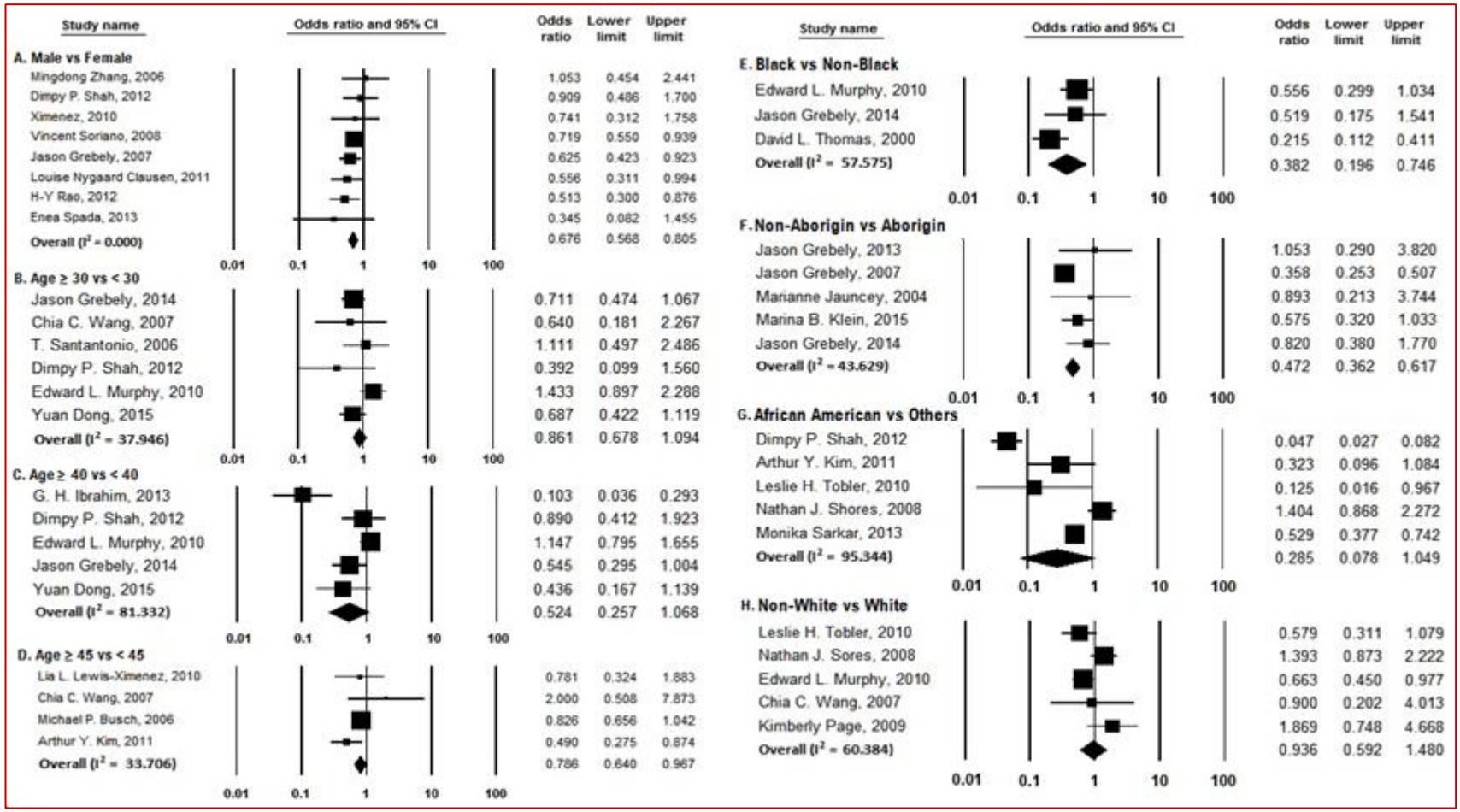


Figure 2.4 Forest Plot Assessing Demographic Factors Associated with HCV Clearance

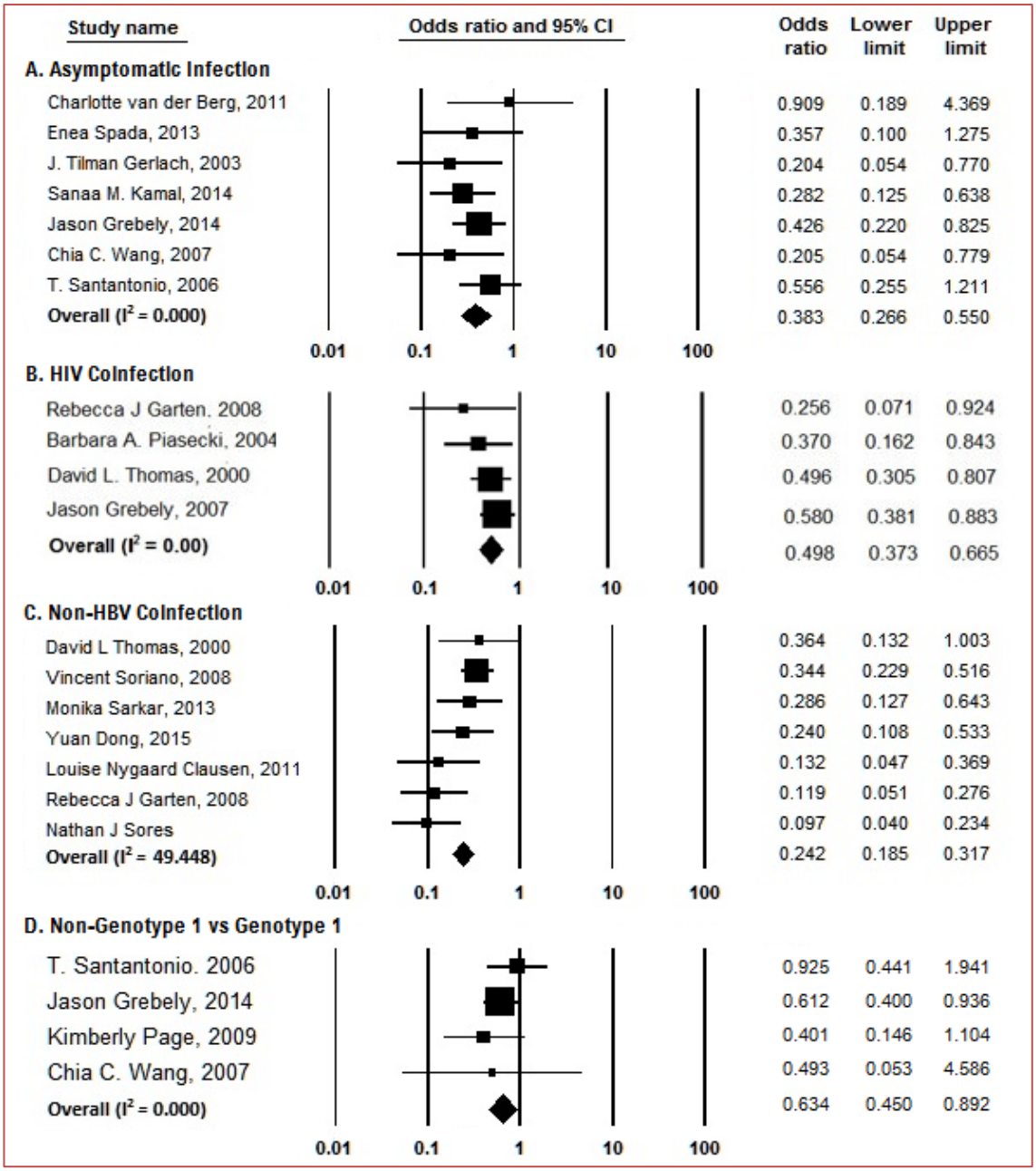


Figure 2.5 Forest Plot Assessing Clinical Factors Associated with HCV Clearance

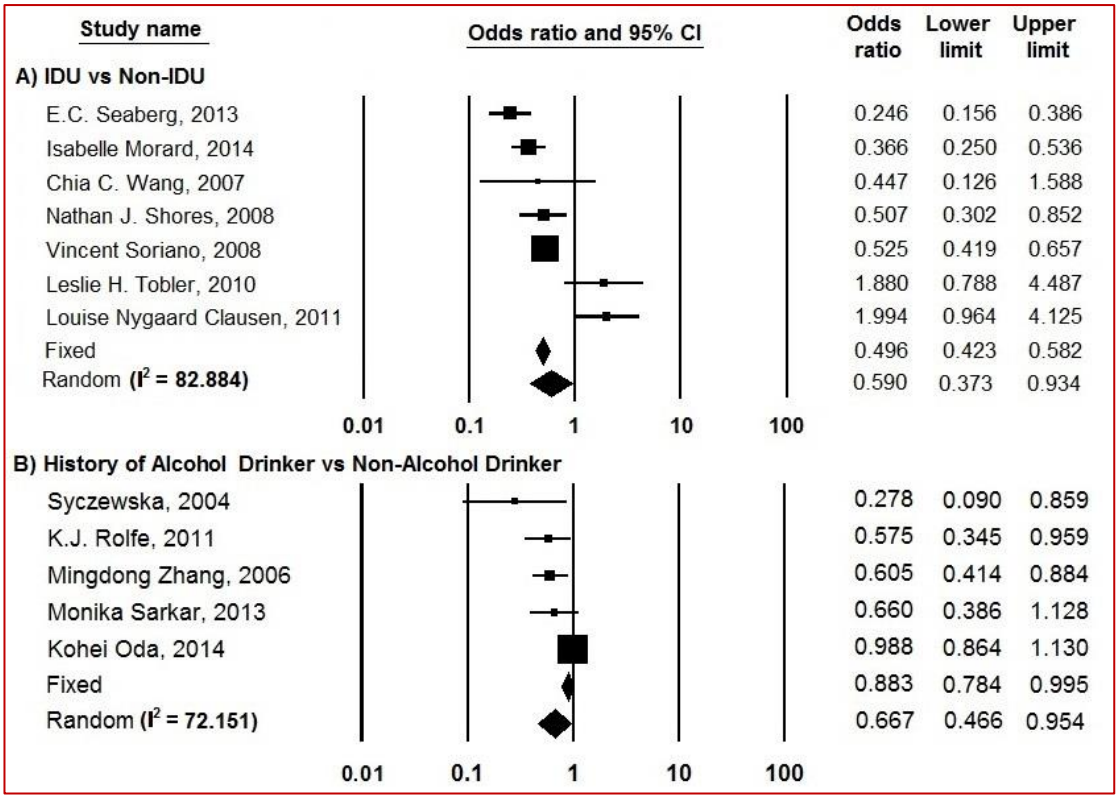


Figure 2.6 Forest Plot Assessing Behaviour Factors Associated with HCV Clearance

2.4 DISCUSSION

2.4.1 SUMMARY OF FINDINGS

In this systematic review and meta-analysis of hepatitis C spontaneous clearance, I included data from 43 studies, representing 20,110 individuals. I found that patients with HCV continue to spontaneously clear HCV for at least 12 months following initial infection but those who have not cleared by this point are unlikely to do so. Notably PWID, who represent the majority of HCV cases and pose a risk for ongoing HCV transmission were less likely to achieve SVC compared to individuals with no history of injection drug use. Other factors that reduce the risk of SVC included: HIV co-infection, non-genotype 1 infection, asymptomatic, black or indigenous race, and those with excess alcohol.

2.4.2 STUDY STRENGTHS AND LIMITATION

The 43 studies included in the analysis were of variable quality, but due to the wide range of study designs I did not conduct formal appraisal of study quality. However, I did use a range of inclusion and exclusion criteria to ensure studies were suitable for addressing the questions of the review. I discuss strengths and weaknesses of the review methodology and generic limitations of the component studies below.

In this study, I utilised a robust method to summarise the current knowledge of HCV clearance as well as to enhance the precision of odd ratios on several predictors of clearance. The strengths of my study are that I performed an extensive systematic search to identify any publications reporting spontaneous HCV clearance and factors associated with it. I used a robust and systematic approach based on the PRISMA guidelines to perform an extensive literature search. By excluding studies with unknown time of infection I was able to more accurately assess clearance rates at different time points following infection than in previous studies. This also suggested that clearance rates could be assessed in studies that had at least 12 months follow up, allowing inclusion of additional data points for estimation of clearance rates. This review followed an established and reproducible procedure for systematic review. To the best of my knowledge, this is the first meta-analysis examining how the proportion of SVC varies over time combined with an assessment of demographic, clinical, and behavioural determinants of HCV clearance. I also undertook a separate analysis of host-genetics factors associated with SVC (discussed in the next chapter).

My study has a range of important limitations related to study design, the heterogeneity of the different studies and the populations included and the quality of information available for case definitions and outcome.

Estimation of SVC: The studies included in this analysis will tend to underestimate SVC because patients who successfully clear infection, and especially those with asymptomatic infection, are less likely to present to the hospital and be included in research studies. Furthermore, most studies could not distinguish between continued infection and re-infection, potentially underestimating SVC in populations who are frequently re-exposed to HCV such as PWID (distinguishing between continuing infection and reinfection would require genomic characterisation of the infecting organism at different time points). I chose to base estimates of SVC on six studies due to the need for certainty about the timing of HCV RNA measurement relative to the time of infection. The drawback of this approach is that these studies represent only a small number of countries and settings which over-represent HCV infected patients in hospital. This may systematically exclude patients who are harder to engage in follow up, who may have different rates of viral clearance. Men are over-represented in these studies and whereas most of the studies are small, there is a single large multinational study which dominates the meta-analysis.(14) Consequently estimates based on these six studies may not be generalizable to the general population. The small number of individuals included in these estimates is

reflected by the broad confidence intervals around the estimate of SVC. A further limitation was the quality of data recorded on the frequency and timing of HCV-RNA re-testing which varied between studies.

Measurement of risk factors for SVC: Methods and definitions used to measure behaviours varied between studies, for example to define heavy alcohol intake. In addition, many studies omitted information on important variables such as age, alcohol consumption, or smoking behaviour. Such omissions are likely to have confounded the univariate associations between a given risk factor and SVC reported in my study. Only a small number of studies presented the results of multivariable analysis having attempted to control for potential confounders. For example, in meta-analysis assessing predictors of spontaneous clearance, only sex, HIV co-infection, and non-HBV co-infection variables were reported based on multivariate analysis. Meta-regression analyses which obtain individual level data from authors of multiple studies can allow the effect of confounding to be adjusted for but this would have been beyond the resources and time available to me. There was also an issue of the power of the study to assess risk factors for clearance as many studies were based on very small sample sizes. A key purpose of meta-analysis is to combine results across multiple small studies to provide sufficient power to assess associations. Nevertheless, there may have been insufficient power to assess the impact of risk factors with weaker associations or rare risk factors.

When hypothesised risk factors are associated with each other and with the outcome of interest then confounding is a potential reason for apparent associations between hypothesised risk factors and outcomes. For example, if injecting drug use and ethnicity are associated with each other and with SVC then each may confound the other leading to potentially spurious association. Multivariable analyses can adjust for the effect of confounding. Where possible I used odds ratios from multivariate analyses in the meta-analysis, however authors were inconsistent in their reporting of these. Options to address the issue of confounding are meta-regression and attempting to estimate the likely impact of confounding by assessing the individual quality of the included studies. I was unable to undertake meta-regression due to lack of individual-level data. The large number of included studies precluded detailed assessment of bias for each individual study so I have instead summarised the limitations of my approach and commented on how this might impact on my results.

The ideal study to estimate SVC would follow up patients at high risk of infection with regular blood samples to allow accurate measurement of the timing of infection onset. Initial samples would be subject to genetic analysis. Patients would not be treated. Regular samples would be taken throughout follow up with further genetic analysis to distinguish between continuance of infection and reinfection. The study would use validated instruments for measuring a wide range of risk factors. The study would be adequately powered to assess associations between these risk factors and clearance. Analysis would assess for interaction (differential effects of different risk factors in different groups) and control for confounding. Given the availability of highly effective treatments and the impetus to treat as many patients as possible, it is unlikely that such studies will now take place. Meta regression analyses of existing studies may be the most realistic option for improving understanding of SVC.

2.4.3 Discussion of the Results

In this systematic review and meta-analysis of hepatitis C spontaneous clearance, I included data from 43 studies, representing 20,110 individuals. I found that patients with HCV continue to spontaneously clear HCV for at least 12 months following initial infection but those who have not cleared by this point are unlikely to do so. Notably PWID, who represent most HCV cases and pose a risk for ongoing HCV transmission were less likely to achieve SVC compared to individuals with no history of injection drug use. Other factors that reduce the risk of SVC included: HIV co-infection, non-genotype 1 infection, asymptomatic infection, black or indigenous race, and those with excess alcohol.

My results suggest that 36% of those infected with HCV will spontaneously clear the virus by 12 months. This is higher than a previous estimate from a study conducted by Micallef et.al (1) which did not consider time since infection and only included studies with at least one follow-up assessment within 24 months of initial HCV infection. Although my results are based on just six studies, a strength of my approach is that I have certainty around the timing of infection – an important factor which may lead to underestimation of SVC. I also applied strict criteria to confirm SVC (at least 2 consecutive serum samples with undetectable HCV RNA). Interestingly, when I applied the 12 month cut-off to all studies, the estimate of SVC was comparable to that reported by Micallef et al.

I found a wide range of factors that affected viral clearance including HIV co-infection and intravenous drug use. Previous studies have suggested that HIV

associated immunodeficiency may weaken immune control, allowing substantial HCV virus replication following initial infection.(55, 56) which is supported by the observation that HCV-specific circulating CD4 and CD8 T cells are usually present in higher concentrations in individuals that go on to clear HCV.(57) There are considerable methodological challenges associated with assessing clearance rates among PWID in cohort studies. PWID have higher rates of loss to follow-up compared to individuals who do not inject drugs, potentially biasing estimates of SVC within these individuals. Alternative explanations for the reduced clearance HCV in PWID might reflect that these patients do clear the virus but are re-infected due to ongoing injecting before being re-tested. Reinfection rates in PWID have been found to vary between 1.8 to 46.8 per 100 person-years in PWID (58) which may increase the risk of new drug resistance (59-61).

I found that decreased clearance was associated with male sex, non-HBV co-infection, asymptomatic infection, non-genotype 1, and older age. Sex hormones have been demonstrated to influence immunity (62, 63), and females have been shown to produce more vigorous cellular and humoral immune reactions increasing their resistance to certain infections.(62) There is also evidence that oestrogens increase autoantibody production while testosterones decrease it. Other reports suggest females have greater immune and inflammatory responses.(64) However, knowledge of the mechanism and the data of sex-based differences in HCV clearance are still very limited. A study conducted by Tang et.al has discovered that the association of oestrogen receptor alpha, ESR2 rs4986938 AA genotype, was strongly associated with HCV clearance among the Chinese Han population.(65) Further studies are needed to examine the association between gender and HCV clearance as well as the underlying mechanisms.

The reasons why co-infection with hepatitis B co-infection increases the spontaneous HCV clearance also remain unclear. It is believed that there is a biological interaction between HBV and the HCV virus-specific T-cell response leading to production of interferons which may trigger a suppressive effect on HCV infection.(66)

People with asymptomatic infection seemed to have lower clearance compared to those that were symptomatic. It is speculated that persons with strong basal immune response are likely to produce jaundice or clinical manifestations but have a better likelihood of eradicating the HCV virus and controlling the infection.(67, 68) In addition, my results suggest individuals infected with HCV non-genotype 1 were

less likely to clear compared to genotype 1. Only a few studies have reported the association between HCV genotype and clearance due to the difficulties involved in recruitment and follow up of acutely HCV infected individuals. Many studies have reported that HCV interferon treatment is less effective for patients with genotype 1 infection.(69-71) However, patients with DAA treatment showed higher effectiveness for genotype 1 compared to genotype 3.(72, 73) Further studies are needed to explore the relationship between host viral mechanisms of genotype 1 infection and HCV clearance.

Many studies have investigated the association between age at time of infection and HCV clearance with conflicting findings. Based on the analysis, older age appeared to be associated with lower clearance. This might be due to younger people having a more vigorous immune response to viral infection.(74) However, since most HCV patients were asymptomatic, some studies could not clarify the true initial time of infection which might produce bias at estimating the age at time of infection.

Our analysis found that alcohol drinkers or people who had a history of drinking excess alcohol appeared to have a lower clearance. It has been recognised that alcohol consumption is associated with liver disease progression among chronic HCV patients and increases in progression of HCV to cirrhosis and hepatocellular carcinoma (HCC).(75, 76) Furthermore, high alcohol consumption has been demonstrated to have several immunosuppressive effects, for example, studies in mice have shown that alcohol ingestion was related with impaired immune response to HCV protein.(77, 78) Our study suggests that people with HCV infection and ongoing treatment should avoid alcohol consumption although more research is needed to define what level of alcohol consumption affects the risk of clearance or treatment outcomes.

The specific association between race and HCV clearance is not well understood and may be confounded by other factors such as prevalence of injecting drug use. Some studies have proposed differences in natural killer (NK) cell populations (79) and frequencies of HLA Class II alleles (80) may explain the dissimilarity of hepatitis C natural history, SVC rate, and response to antiviral treatment among racial groups. Ethnicity is also associated with IL28B polymorphism, which is believed as the strongest host genetic predictor of HCV clearance. (10, 81, 82) Again, more studies are needed to better explain the racial differences in HCV immunity.

2.4.4 IMPLICATION FOR POLICY AND PRACTICE

This study estimates the proportion of SVC following acute HCV infection at over 35% at 1-year post infection. This suggests that treatment may not be required for an estimated third of individuals who are infected with HCV. However, putting a "watch and wait" approach into practice is not straightforward for many reasons.

In my analysis, I restricted estimates of SVC to a small number of studies where the timing of clearance had been measured reliably. In reality, most patients with HCV are unaware of the date that they acquired infection and many remain asymptomatic for many years. Consequently, many patients who attend clinic will have been infected for at least 12 months and most of these individuals will require treatment. The work certainly suggests that there is little value in withholding treatment where patients are known to have been infected for at least 12 months as spontaneous clearance is unlikely after this. Delaying treatment by 12 months could make sense in settings where screening is being undertaken on a regular basis except that individuals who are screened regularly tend to be high-risk individuals such as PWID. There is a clear public health argument for early treatment/prioritisation for PWID because of the risk of onwards transmission of HCV. The importance of focusing on high-risk groups is borne out by my analysis of risk factors for HCV clearance which demonstrated significantly reduced clearance rates in individuals with HIV co-infection, active intravenous drug-use, and excessive alcohol intake. These data provide support for a strategy of early treatment for high-risk groups who are less likely to achieve SVC, may pose a higher risk of onward transmission and who may be more likely to be lost to follow up. Achieving this strategy will require outreach to these higher risk groups, active engagement with drug and alcohol liaison services to address addiction problems and reinforcement of harm minimisation approaches to reduce the risk of transmission and reinfection. The European Association for the Study of the Liver (EASL) have recently made similar recommendations.(83)

Perhaps the setting where these findings have greatest applicability is in low and middle-income countries, where the availability of DAA drugs is limited and the predominant HCV risk factor is iatrogenic transmission. In low risk individuals with a low-risk of onward transmission of HCV, the 'watch and wait' approach might maximise the use of available resources by giving sufficient time for infected individuals to achieve SVC and saving up to a third of DAA treatments.

2.4.5 MY ROLE IN THIS STUDY

I devised the idea for the study in collaboration with both of my supervisors. I developed the study protocol and registered it into PROSPERO. I undertook the literature search, reviewed all abstracts and performed full-text review of included publications with another independent reviewer. I also conducted the data extraction and undertook the meta-analysis. I wrote the first draft of the manuscript and submitted it into international peer-reviewed journal. This work has been published in the Journal of Viral Hepatitis (**DN Aisyah**, L Shallcross, A O' Brien, AJ Hully, A Hayward. Assessing Hepatitis C Spontaneous Clearance and Understanding Associated Factors: A Systematic Review and Meta-Analysis. Journal of Viral Hepatitis. January 2018. DOI https://doi.org/10.1111/jvh.12866).

Key Points:

- A total of 139 studies were included in the main analysis, comprising of 6 studies that calculated HCV clearance with information of initial time of infection; 43 studies that assessed demographic, clinical and behavioural factors related with HCV clearance; and 86 studies provided information of immunological determinants for clearance (presented in chapter 3)
- Based on 6 studies which reported timing of clearance accurately, acute HCV patients continued to spontaneously clear HCV up to at least 12 months following initial infection. The rates of spontaneous clearance were 19.8% (95% CI: 2.6-47.5%), 27.9% (95% CI: 17.2-41.8%), 36.1% (95% CI: 23.5-50.9%), and 37.1% (95% CI: 23.7-52.8%) within 3, 6, 12, and 24 months after infection respectively.
- Meta-analysis on factors associated with clearance found that female sex, hepatitis B co-infection, symptomatic infection, non-black or non-indigenous race, infection with genotype-1, younger age, those without alcohol or drug problems and HIV negative patients were predictors of spontaneous clearance.
- Some key limitations of my study related to study design, the heterogeneity of the different studies and the populations included and the quality of information available for case definitions and outcome. The 6 studies of SVC represent only a small number of countries, over-represent HCV infected patients in hospital, most of the studies are small (a single large multinational study which dominates the meta-analysis) thus reflected by the broad confidence intervals. For measurement of risk factors analysis, the limitations included the varied methods and definitions used to measure behaviours between studies, only a small number of studies presented the results of multivariable analysis, and the power of the study. Considering the lower chance of clearance and risk of transmission among high-risk groups, this study suggested early treatment for these populations.
- It is important to have active engagement with drug and alcohol liaison services identify patients early and to reduce the risk of transmission among high-risk individuals.
- The ‘watch and wait approach’ might be most relevant to be implemented in low and middle-income countries where the availability of DAA drugs is limited and the predominant HCV risk factor is iatrogenic transmission (thus has low-risk of onward transmission).

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3. Host Genetic Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis

Chapter's Aim:

The aim of this chapter is to identify host genetic factors associated with hepatitis C clearance through systematic review and meta-analysis. Host genetic factors investigated include: interleukin, HLA-class I A, HLA class-I B, HLA class-I C, HLA-class II, KIR alleles, CCR5 genotype, and other candidate genes.

3.1 Background

As previously described in chapter 2, an estimated 20% to 30% of hepatitis C infected patients will clear the virus without treatment, confirmed by the disappearance of HCV RNA in the serum. The result from chapter 2 shows higher spontaneous clearance of 36% after one year of initial infection. Whether HCV infection clears spontaneously or persists in the body depends upon a complex set of interactions between host and virus which are only partially understood. Other than demographic, clinical, and behaviour factors, several studies have demonstrated an association between polymorphisms in genes coding for interleukins, Human Leukocyte Antigen (HLA), cytokines and KIR (Killer Cell Immunoglobulin-like receptors) and spontaneous viral clearance. However, the findings of these studies are often conflicting, which may be driven by different study populations, differences in how HCV clearance is defined and/or sample size. A better understanding of host genetic determinants of the natural control of HCV infection could reveal novel therapeutic and preventive strategies.

In the age of DAA treatment where the effectiveness of the drugs is over 90%, optimising HCV treatment in terms of choice of drug and duration is a hot topic in HCV research. Given the high cost of treatment (1), it may make sense to prioritise early treatment for individuals who are unlikely to achieve spontaneous clearance. Host genetic predictors might help us to select the right candidates for early treatment, as several potential host genetic determinants have been shown to strongly predict clearance.(2, 3) Thus, genetic testing may help select individuals that would profit most from immediate treatment (high risk of progression and/or low risk of clearance). In addition it has been hypothesised that those who are least

likely to spontaneously clear virus may also be the hardest to treat and most likely to develop resistance.(4) Genetic testing may therefore also help to identify patients with favourable genotypes or alleles that might benefit from shorter duration of treatment or identify individuals who are likely to relapse or be slow responders. This review aims to assess the association between host genetics factors and hepatitis C spontaneous clearance.

3.2 METHODOLOGY

The methodology in this chapter is described in detail in chapter 2, but in brief, I selected studies that reported host genetic factors associated with SVC and performed a systematic search using the terms “hepatitis C” or “HCV” AND “natural history” or “clearance” or “vir* negativ*”. I searched Ovid Medline, Ovid Embase, and Pubmed for studies that were published in English between 1st January 1994 and 30th June 2015. The protocol for this review has been registered on the PROSPERO database (http://www.crd.york.ac.uk/PROSPERO/) reference: CRD42015023499.

I included all host genetic factors found from the systematic literature review and calculated odds ratios to analyse the relationship between host genetic factors and spontaneous HCV viral clearance. I performed meta-analysis to examine the relationship between each putative genetic determinant and clearance, where possible. I calculated Odd Ratios and generated forest plots generated using Comprehensive Meta-Analysis (CMA) version 3.0. I also performed sensitivity analysis by calculating the odd ratios for studies which have a minimum of 12 months follow up. The cut off of 12 months reflected the results from the previous chapter that HCV patients who had not spontaneously cleared by 12 months were unlikely to do so. Having identified host genetic determinants with the strongest association with HCV clearance, I performed meta-analysis to summarise the frequency of each predictor allele among HCV patients with spontaneous clearance and HCV patients with persistent infection and also comparing HCV patients with health controls to show which alleles are likely to be important at a population level. Finally, I tabulated the odds ratio and allele frequency to identify which genetic determinants were both common and had the highest impact on clearance. Heterogeneity was investigated utilizing I^2, and if there was evidence of heterogeneity (I^2 >50%), random effect models were used. I generated a funnel plot of proportions of clearance against the study size to assess for publication bias.

3.3 RESULTS

3.3.1 ARTICLE SCREENING

A total of 9,357 publications were identified from three databases after excluding duplicates. 483 publications met the inclusion criteria for full-text review (see Figure 3.1). I included 86 studies (5-90) which reported the association between host genetic factors and HCV spontaneous viral clearance, representing data from 38,605 participants. 47 studies reported polymorphisms in the genes coding for interleukins, 10 studies reported HLA-class I, 18 studies reported HLA class II, 3 studies reported CCR5, 3 studies reported KIR alleles and 17 studies reported other candidate genes.

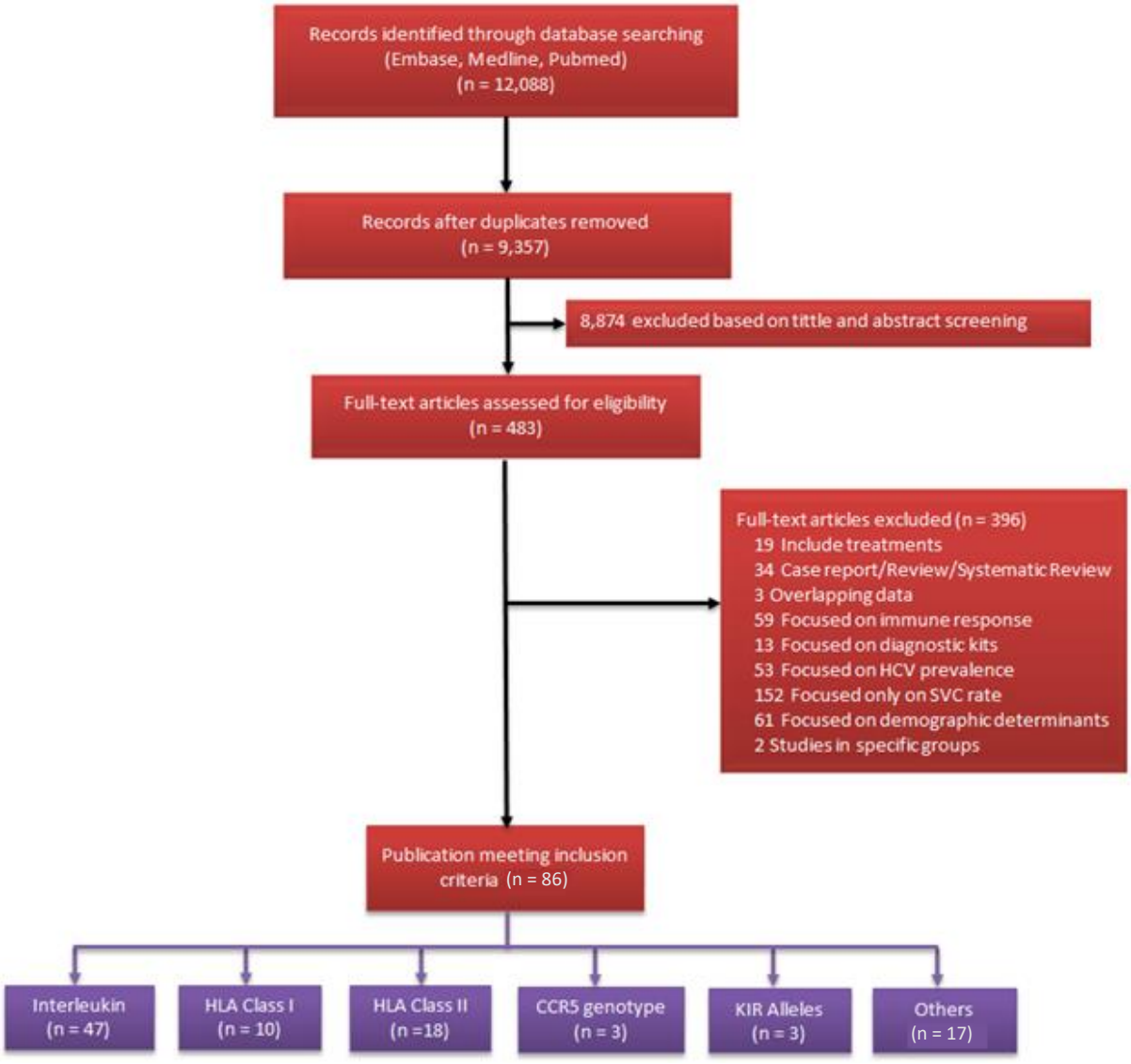


Figure 2.7 Article Screening Following PRISMA Diagram

3.3.2 CHARACTERISTIC OF STUDIES INCLUDED

Of the 86 studies I included in the review, there were 26 prospective and 2 retrospective cohort studies, 56 case-control and 2 cross-sectional studies. Study participants were recruited from hospitals or related health centres (66 studies) and included patients who were transfusion dependent (3 studies), people who inject drugs - PWID (5 studies), general population (3 studies), HIV positive patients (4 studies), and blood donors (5 studies). A total of 52 studies were published between 2010 and 2015. The majority of studies were conducted in European countries (36), followed by Asia (18), North America (17), Middle East (4), South America (4), Africa (4 studies), and 3 multi-national studies (see figure 3.2). Detailed characteristics of included publications are described in Table 3.1.

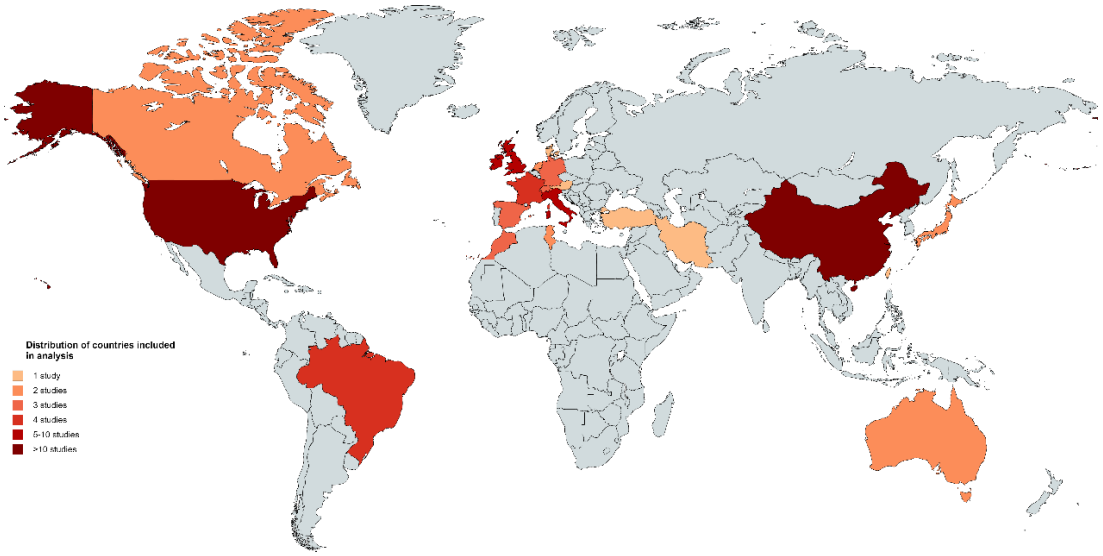


Figure 2.8 Distribution of Countries Included in the Analysis

Table 2.3 Characteristic of Studies Included Assessing Immunological Factors Associated with HCV Spontaneous Clearance

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
C. Goulding	Ireland	2005	Transfusion Dependent HCV patients	0/283		283	87	30.74	25.49-36.53
Elizabeth J. Minton	UK	2005	HCV infected patients	404/202		606	190	31.35	27.70-35.24
Ming-Lung Yu	Taiwan	2014	Transfusion Dependent patients	115/172	62±11.6	287	73	25.44	20.59-30.96
Matthew E. Cramp	UK	1998	HCV infected patients	61/43	SVC 15.5, Range (3-42) CHC 14.2, Range (2-40)	104	49	47.12	37.34-57.11
Qian Cui	China	2010	HCV infected patients	249/113	SVC 32.43±6.15; CHC 32.63±6.18	362	189	52.21	46.93-57.44
B.S. de Almeida	Brazil	2010	HCV infected patients	42/93	SVC 53 ± 12; CHC 51 ± 11	135	45	33.33	25.6-42.03
Julia di Iulio	Switzerland	2011	HIV (+)			460	227	49.35	44.7-54.01
Priya Duggal	International study	2013	HCV infected patients	1492/909		2401	919	38.28	36.34-40.26
Franziska S. Hoffmann	Germany	2015	HCV infected patients	355/439		794	285	35.89	32.57-39.35
Peng Huang	China	2014	Blood donors	159/566	SVC 57.32±7.93; CHC 57.73±8.05	725	193	26.62	23.47-30.03
Peng Huang	China	2015	HCV infected patients	312/152	SVC 39.10 ±12.17; CHC 40.47 ± 12.33	464	246	53.02	48.37-57.62

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
G.H. Ibrahim	Egypt	2013	HCV infected patients	59/34	Range 23-65	93	22	23.66	15.72-33.81
Leila Ksiaa Cheikrouhou	Tunisia	2007	HCV infected patients	48/51	Overall 56.7±12.4 SVC 55.5; CHC 58	99	24	24.24	16.43-34.08
Marco Antonio Montes-Cano	Spain	2005	HCV infected patients	109/87		196	65	33.16	26.71-40.28
Nasheed Moqueet	Canada	2015	HIV (+)	367/174	44±8.2	541	79	14.60	11.79-17.92
Isabelle Morard	Switzerland	2014	HCV infected patients	886/564	Median SVC 38; Median CHC 20	1450	160	11.03	9.49-12.78
Jacob Natterman	Germany	2011	HCV infected patients	0/396	24.7±4	396	119	30.05	25.62-34.87
Khadija Rebbani	Morocco	2014	HCV infected patients	85/88	SVC 60.2 ± 12.3; CHC 63.5 ± 10.5	173	54	31.21	24.51-38.76
Heidar Sharafi	Iran	2014	HCV infected patients	333/17	SVC 39.2±11.1; CHC 39.8±10.1	350	91	26.00	21.55-30.99
Haibo Sun	China	2015	General population	363/259	SVC 48.2±8.7; CHC 52±9.2	622	544	87.46	84.54-89.91
Shaidi Tang	China	2014	Blood donors	433/876	SVC 55.4±8.8; CHC 56.2±8.0	1309	429	32.77	30.24-35.40
Chloe L. Thio	USA	2002	HCV infected patients	581/94	SVC 25.1; CHC 27.3	675	231	34.22	30.67-37.95
David L. Thomas	USA	2009	HCV infected patients	802/206	SVC 33.9; CHC 32.0	1008	388	38.49	35.49-41.58
Hans L. Tillman	Germany	2010	HCV infected patients	0/190	24.6±4	190	67	35.26	28.57-42.55
Xing-xin Xue	China	2015	HCV infected patients	456/720		1176	444	37.76	34.99-40.61
Ming Yue	China	2013	General population	372/180	SVC 41.68±12.24; CHC 42.96±2.39	552	293	53.08	48.82-57.30
Valli De Re	Italy	2014	HCV infected patients			2931	397	13.54	12.33-14.84

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
S. Ezzikouri	Morocco	2013	HCV infected patients	78/135	SVC 59.81±12.81; CHC 63.09±12.06	213	63	29.58	23.64-36.27
Ming Yue	China	2014	HCV infected patients	382/353	SVC 44.99 ± 13.84; CHC 45.91 ± 13.76	735	317	43.13	39.53-46.81
Yu Liu	China	2013	Blood donors	42/58	SVC 30.0±10.7; CHC 31.8±10.3	100	24	24.00	16.27-33.77
Sayeh Ezzikouri	UK	2013	HCV infected patients	132/168	SVC 57.77±15.64; CHC 63.66±12.26	300	68	22.67	18.14-27.91
Juliene Antonio Ramos	Brazil	2012	HCV infected patients	88/91	SVC 44.4, range (21–73); CHC 52.4, range (24–74)	179	18	10.06	6.24-15.66
M. Bes	Spain	2012	Blood donors	43/26	SVC 46, range (27–61); CHC 43, range (23–63)	69	21	30.43	20.23-42.83
Xiaodong Shi	China	2012	General population	441/284	SVC 51.7±9.4; CHC 50.6±9.1	725	196	27.03	23.86-30.45
Fatma M. Shebl	USA	2011	PWID	825/384		1209	326	26.96	24.49-29.57
Qian Cui	China	2011	HCV infected patients	372/180	SVC 39.10±12.26; CHC 39.23±12.6	552	293	53.08	48.82-57.30
Fuad Kurbanov	Egypt	2011	HCV infected patients	126/153	Median 38	279	130	46.59	40.65-52.63
L. N. Clausen	Denmark	2011	HIV (+)	128/78	SVC 33, range (29–40); CHC 36, range (31–42)	206	47	22.82	17.40-29.28
Jane H Wang	USA	2009	HCV infected patients	62/43	SVC 26.0, range (19-33); CHC 25.6	105	49	46.67	36.96-56.63

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
					range (19-32)				
Ping An	USA	2008	HCV infected patients	536/95		658	241	36.63	32.96-40.46
Rebecca A. Harris	USA	2008	HCV infected patients	87/6	Median SVC 46; Median CHC 52	93	23	24.73	16.63-34.96
Viviana Romero	USA	2008	PWID	119/41	SVC 37.8; CHC 39.9	160	39	24.38	18.10-31.92
J. P. Pandey	Spain	2007	HCV infected patients	72/45		117	50	42.74	33.74-52.22
Branwen J. Hennig	UK	2007	HCV infected patients	345/282		631	112	17.75	14.89-21.01
Kazunori Kusumoto	Japan	2006	HCV infected patients	162/298	SVC 67.9 ± 11.3; CHC 63.4 ± 9.6	460	114	24.78	20.95-29.04
D A Price	UK	2006	HCV infected patients			420	108	25.71	21.65-30.22
TK Oleksyk	USA	2005	HCV infected patients			274	91	33.21	27.73-39.17
Liam J. Fanning	Ireland	2004	HCV infected patients	39/186		225	86	28.22	31.91-44.94
Janardan P. Pandey	USA	2004	HCV infected patients		Median SVC 36 (22-62); Median CHC 36 (23-54)	298	100	33.56	28.28-39.27
S. Barret	Ireland	2003	HCV infected patients	0/158	SVC 45.3±7.3; CHC 44.7±8.5	158	66	41.77	34.06-49.88
Jose Azocar	USA	2003	HCV infected patients	87/25	SVC 37.9; CHC 39.2	112	40	35.76	27.04-45.38
Chloe L. Thio	USA	2001	HCV infected patients	476/98	SVC 25.7; CHC 24.8	574	200	24.84	30.97-38.92
L. Alric	France	2000	HCV infected patients	171/174	SVC 42.1±15.4; CHC 46±12.3	345	63	18.26	14.41-22.83
Liam J. Fanning	Ireland	2000	HCV infected patients	0/156		156	84	53.85	45.71-61.80
Alessandra Mangia	Italy	1999	HCV infected patients			184	35	19.02	13.77-25.60
Laurent Alric	France	1997	HCV infected patients	67/61	SVC 40.6 ± 15.7;	128	25	19.53	13.26-27.67

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
					CHC 45.4 ± 12.4;				
Sandra Beinhardt	Austria	2012	HCV infected patients	64/56	37±16	120	59	49.17	39.99-58.41
Li Cai	China	2014	HCV infected patients	469/823	SVC 49.95±13.52; CHC 50.26±13.52	1292	479	37.07	34.44-39.78
Jeny R. Cursino-Santos	Brazil	2007	HCV infected patients	79/25	Median SVC 40 (27-56); Median CHC 42 (24-71)	104	29	27.88	19.75-37.67
Vito di Marco	Italy	2012	HCV infected patients	124/121	SVC 18.6±8; CHC 18.7±6.5	245	98	40.00	33.87-46.45
Karen Fitzmaurice	Ireland & Swiss	2014	HCV infected patients			780	332	42.56	39.07-46.12
Charlotte H.B.S. van der Berg	Netherland	2011	PWID	62/44	Median 28.5	106	35	33.02	24.37-42.91
Jason Grebely	Australia, Canada, Netherland, USA	2014	HCV infected patients	404/228		632	173	27.37	22.96-31.06
Arthur Y. Kim	USA	2011	HCV infected patients	131/215		346	66	19.08	15.16-23.71
Alessandra Mangia	Italy	2011	HCV infected patients	59/58	SVC 34.1±6.3; CHC 35.8±5.7	117	49	41.88	32.91-51.36
Susan M. McKiernan	Ireland	2000	HCV infected patients	0/243	SVC 48.75; CHC 47.86	243	95	39.09	32.97-45.56
E.J. Minton	UK	1998	HCV infected patients	106/67	SVC 37.9, range (17-70); CHC 37.2,	173	35	20.23	14.67-27.15

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
					range (20-77)				
Alessandra Mangia	Italy	2014	HCV infected patients	408/313	SVC 35.9±15; CHC 53.6±12.7	721	100	13.87	11.47-16.66
Serkan Ocal	Turkey	2014	HCV infected patients	130/59	Median 27, range (17-56)	189	57	30.16	23.82-37.32
Kohei Oda	Japan	2014	HCV infected patients	167/335	73, range (37-97)	502	149	29.68	25.76-33.92
H-Y Rao	China	2012	HCV infected patients	163/213	53.2±8	376	80	21.28	17.32-25.84
Maria Concetta Renda	Italy	2011	HCV infected patients	16/26	44.68±12.09	42	20	47.62	32.29-63.38
E.C. Seaberg	USA	2013	HCV infected patients	528/0	Median 33.5, range (17-70)	528	118	22.35	18.91-26.20
Enea Spada	Italy	2013	HCV infected patients	39/17	Median 31, range (19-78)	56	18	32.14	20.65-46.09
CL Thio	USA	2004	HCV infected patients	469/98		567	192	33.86	30.00-37.94
Sanaa M. Kamal	Egypt	2014	HCV infected patients	69/67	SVC 36.48 ± 7.64; CHC 37.17 ± 6.2	136	48	35.29	27.42-44.00
S Barret	Ireland	2001	HCV infected patients	0/155	SVC 45.8±4.9; CHC 45.7±6.0	155	68	43.87	35.99-52.06
Leila Ksiaa Cheikrouhou	Tunisia	2011	Transfusion Dependent patients	48/52	SVC 55.5±11.71; CHC 58±13.08	100	24	24.00	16.27-33.77
Maria Elisa Mancuso	Italy	2014	HCV infected patients	329/13	Median SVC 39.6 (34.8–55.3); Median CHC 47.2 (40.8–56.9)	342	59	17.25	13.48-21.77
Peter V. Aka	USA	2014	HCV infected patients			1075	185	17.21	15.03-19.63

First Author	Country	Year	Study Population	M/F	Age*	Σ of HCV (+)	∑ of Clearance	% of clearance	95% CI
Vagner Ricardo Lunge	Brazil	2011	HIV (+)	86/52	SVC 42.7±9.5; CHC 41.4±9.5	138	34	24.64	17.88-32.84
Melissa Laird	France	2014	HCV infected patients	22/11		33	19	57.58	39.39-7.05
Yin Huang	USA	2007	PWID			251	85	33.86	28.10-40.12
Wen Xiao	China	2015	HCV infected patients	231/588	SVC 57.1±7.9; CHC 56.2±7.6	819	219	26.74	23.76-29.94
Alessandra Mangia	Italy	2004	HCV infected patients	134/140	Range 20-79	220	50	22.73	17.48-28.95
Patricia K. Constantini	UK	2002	HCV infected patients	93/57	38.8, range (16-68)	150	57	38.00	30.31-46.31

*Mean of age, otherwise stated in the table

3.3.3 HOST GENETIC FACTORS ASSOCIATED WITH SVC

The systematic search identified 142 genetic factors, and through meta-analysis, I identified 24 factors which were significantly associated with SVC. Genetic factors found which were strongly associated with SVC included IL28B rs8103142 TT genotype (OR=4·06, 95% CI: 2·64-6·25), IL28B rs12979860 CC genotype (OR=3·27, 95% CI: 2·68-3·98), IL28B rs8099917 TT genotype (OR=2·83, 95% CI: 2·36-3·39), DRB1*01 (OR=2·50, 95% CI: 1·49-4·19), DQB1*0301 (OR=2·30, 95% CI: 1·49-3·57), HLA-B*27 (OR=2·29, 95% CI: 1·45-3·63), HLA-Cw*07 (OR=2·00, 95% CI: 1·36-2·92), DRB1*03011 (OR=0·47, 95% CI: 0·36-0·60), DQB1*02 (OR=0·36, 95% CI: 0·26-0·51), etc. Forest plots for each host genetic predictors are shown in Figure 3.3-3.9 below.

a) Interleukin 28B

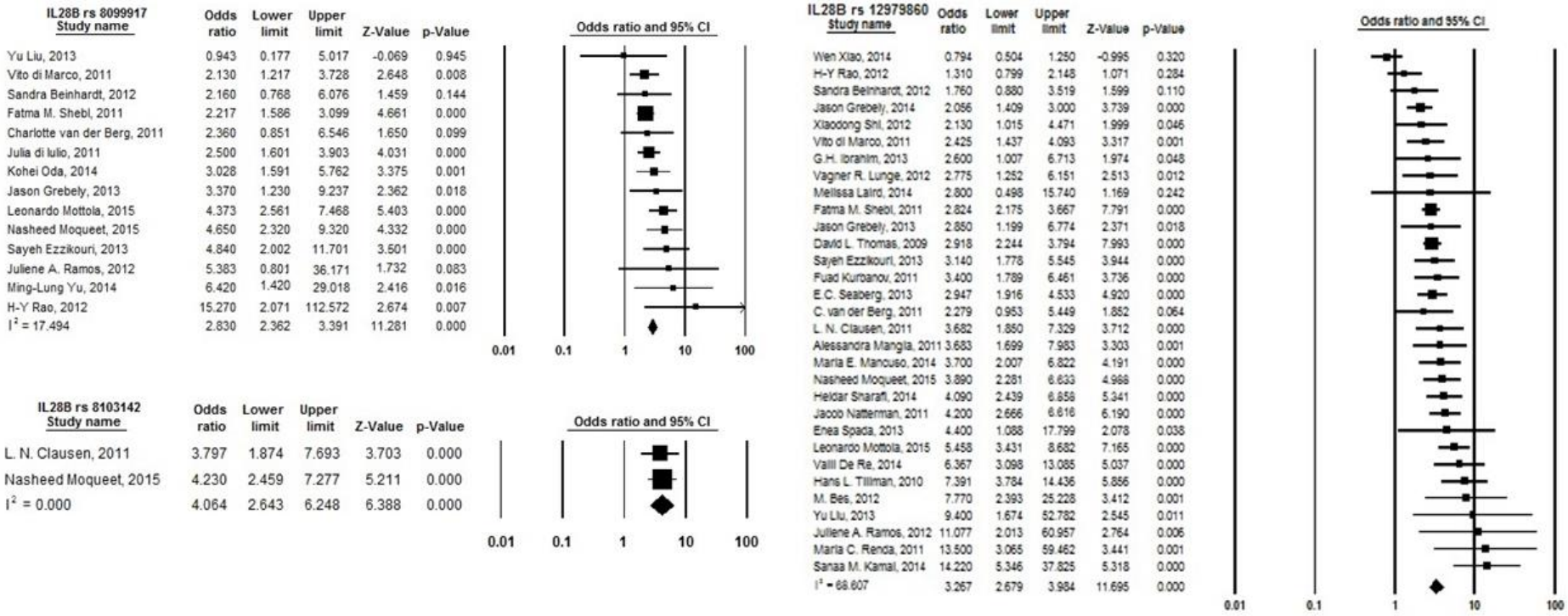
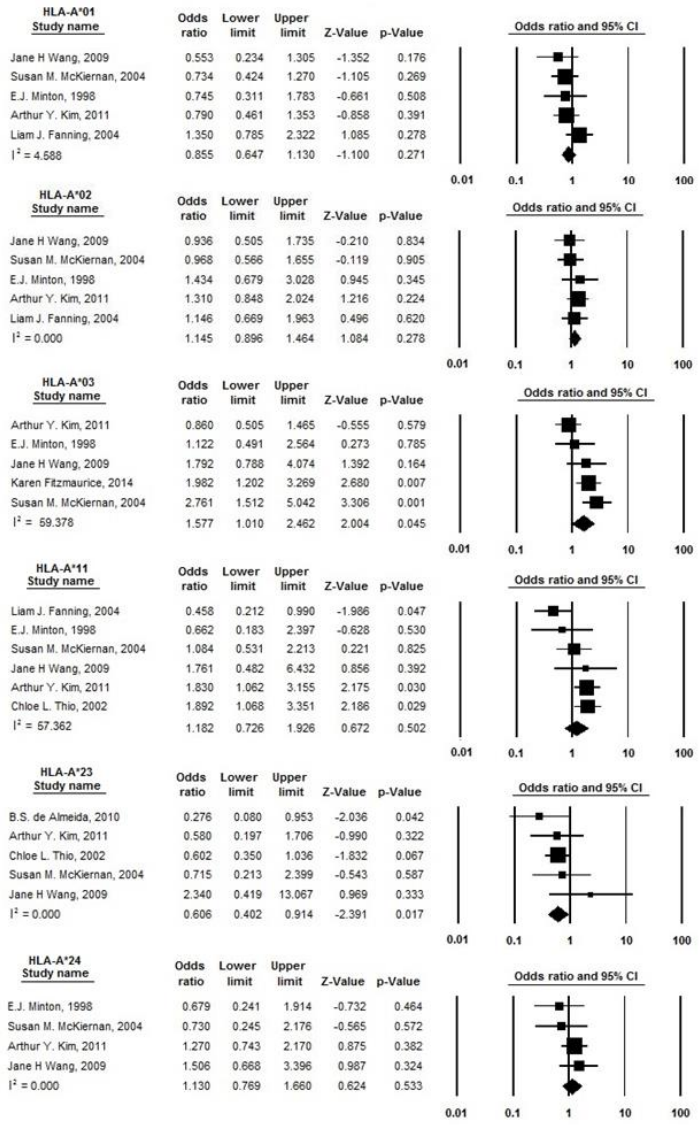


Figure 2.9 Meta-Analysis of IL28B rs 8099917, IL28B rs 8103142, and IL28B rs 12979860 to HCV Spontaneous Clearance

b) HLA Class-I A



HLA-A*01 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jane H Wang, 2009	0.553	0.234	1.305	-1.352	0.176
Susan M. McKiernan, 2004	0.734	0.424	1.270	-1.105	0.269
E.J. Minton, 1998	0.745	0.311	1.783	-0.661	0.508
Arthur Y. Kim, 2011	0.790	0.461	1.353	-0.858	0.391
Liam J. Fanning, 2004	1.350	0.785	2.322	1.085	0.278
I^2 = 4.588	0.855	0.647	1.130	-1.100	0.271

HLA-A*02 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jane H Wang, 2009	0.936	0.505	1.735	-0.210	0.834
Susan M. McKiernan, 2004	0.968	0.566	1.655	-0.119	0.905
E.J. Minton, 1998	1.434	0.679	3.028	0.945	0.345
Arthur Y. Kim, 2011	1.310	0.848	2.024	1.216	0.224
Liam J. Fanning, 2004	1.146	0.669	1.963	0.496	0.620
I^2 = 0.000	1.145	0.896	1.464	1.084	0.278

HLA-A*03 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Arthur Y. Kim, 2011	0.860	0.505	1.465	-0.555	0.579
E.J. Minton, 1998	1.122	0.491	2.564	0.273	0.785
Jane H Wang, 2009	1.792	0.788	4.074	1.392	0.164
Karen Fitzmaurice, 2014	1.982	1.202	3.269	2.680	0.007
Susan M. McKiernan, 2004	2.761	1.512	5.042	3.306	0.001
I^2 = 59.378	1.577	1.010	2.462	2.004	0.045

HLA-A*11 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Liam J. Fanning, 2004	0.458	0.212	0.990	-1.986	0.047
E.J. Minton, 1998	0.662	0.183	2.397	-0.628	0.530
Susan M. McKiernan, 2004	1.084	0.531	2.213	0.221	0.825
Jane H Wang, 2009	1.761	0.482	6.432	0.856	0.392
Arthur Y. Kim, 2011	1.830	1.062	3.155	2.175	0.030
Chloe L. Thio, 2002	1.892	1.068	3.351	2.186	0.029
I^2 = 57.362	1.182	0.726	1.926	0.672	0.502

HLA-A*23 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
B.S. de Almeida, 2010	0.276	0.080	0.953	-2.036	0.042
Arthur Y. Kim, 2011	0.580	0.197	1.706	-0.990	0.322
Chloe L. Thio, 2002	0.602	0.350	1.036	-1.832	0.067
Susan M. McKiernan, 2004	0.715	0.213	2.399	-0.543	0.587
Jane H Wang, 2009	2.340	0.419	13.067	0.969	0.333
I^2 = 0.000	0.606	0.402	0.914	-2.391	0.017

HLA-A*24 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
E.J. Minton, 1998	0.679	0.241	1.914	-0.732	0.464
Susan M. McKiernan, 2004	0.730	0.245	2.176	-0.565	0.572
Arthur Y. Kim, 2011	1.270	0.743	2.170	0.875	0.382
Jane H Wang, 2009	1.506	0.668	3.396	0.987	0.324
I^2 = 0.000	1.130	0.769	1.660	0.624	0.533

HLA-A*25 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2004	0.196	0.024	1.596	-1.523	0.128
Jane H Wang, 2009	0.567	0.051	6.327	-0.461	0.645
I^2 = 0.000	0.310	0.064	1.507	-1.452	0.147

HLA-A*29 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
E.J. Minton, 1998	0.694	0.147	3.281	-0.461	0.645
Susan M. McKiernan, 2004	1.118	0.461	2.711	0.247	0.805
Jane H Wang, 2009	0.182	0.022	1.523	-1.572	0.116
Arthur Y. Kim, 2011	0.340	0.089	1.302	-1.575	0.115
I^2 = 19.173	0.671	0.355	1.268	-1.230	0.219

HLA-A*30 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jane H Wang, 2009	0.411	0.106	1.593	-1.286	0.198
Arthur Y. Kim, 2011	0.890	0.408	1.940	-0.293	0.769
E.J. Minton, 1998	5.542	1.404	21.877	2.444	0.015
I^2 = 73.817	1.223	0.332	4.501	0.302	0.762

HLA-A*31 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2004	0.536	0.106	2.713	-0.754	0.451
Jane H Wang, 2009	1.452	0.379	5.564	0.544	0.586
I^2 = 0.000	0.968	0.344	2.724	-0.062	0.951

HLA-A*32 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2004	0.983	0.344	2.808	-0.032	0.974
Jane H Wang, 2009	2.340	0.419	13.067	0.969	0.333
E.J. Minton, 1998	2.370	0.741	7.583	1.454	0.146
I^2 = 0.000	1.581	0.778	3.216	1.266	0.206

HLA-A*68 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Arthur Y. Kim, 2011	0.950	0.443	2.036	-0.132	0.895
Susan M. McKiernan, 2004	1.247	0.417	3.727	0.395	0.693
Jane H Wang, 2009	1.546	0.337	7.089	0.561	0.575
I^2 = 0.000	1.100	0.617	1.961	0.322	0.748

c) HLA Class-I B

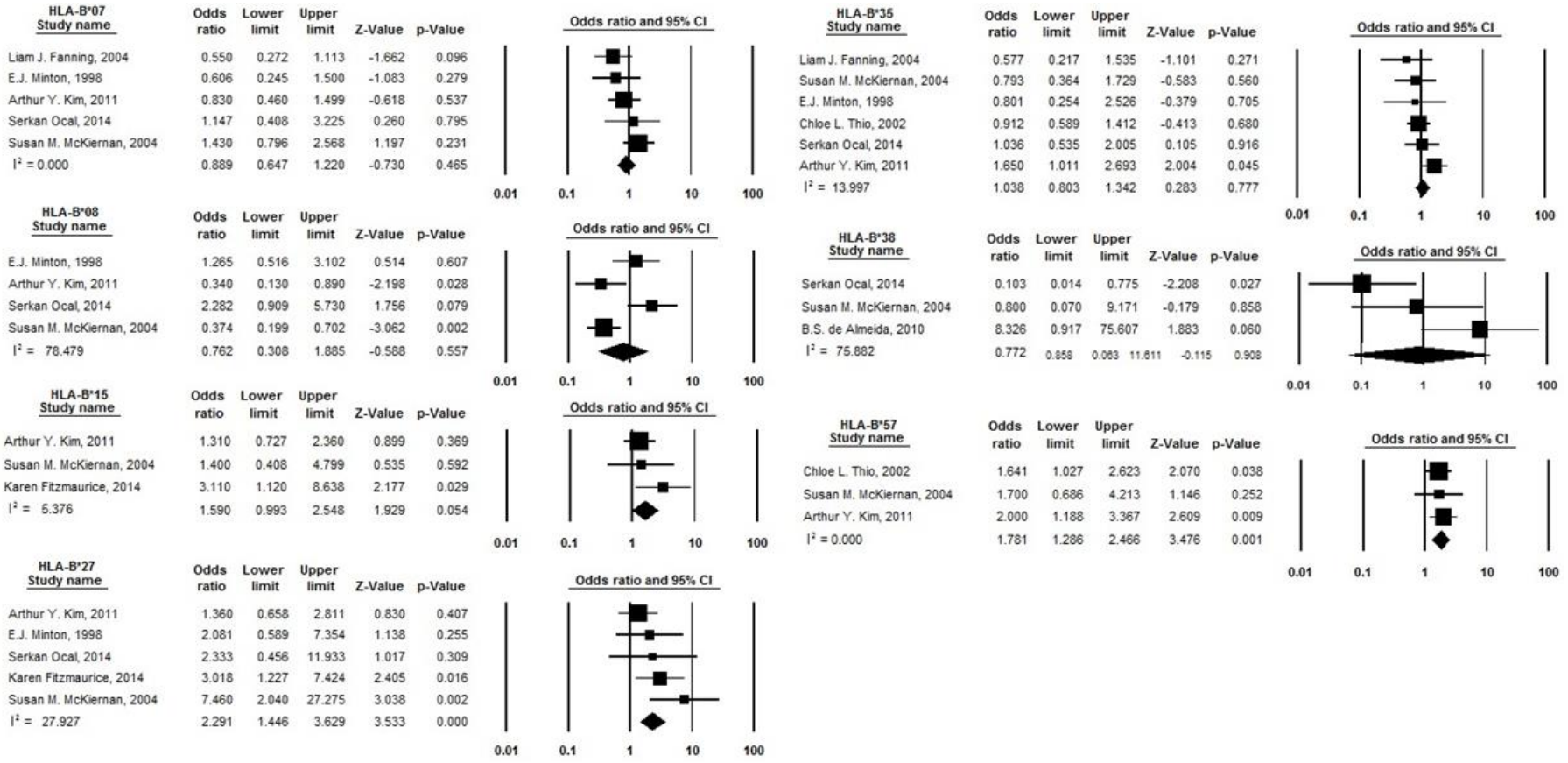


Figure 2.10 Meta-Analysis of HLA Class-I A and HLA Class-I B to HCV Spontaneous Clearance

d) HLA Class-I Cw

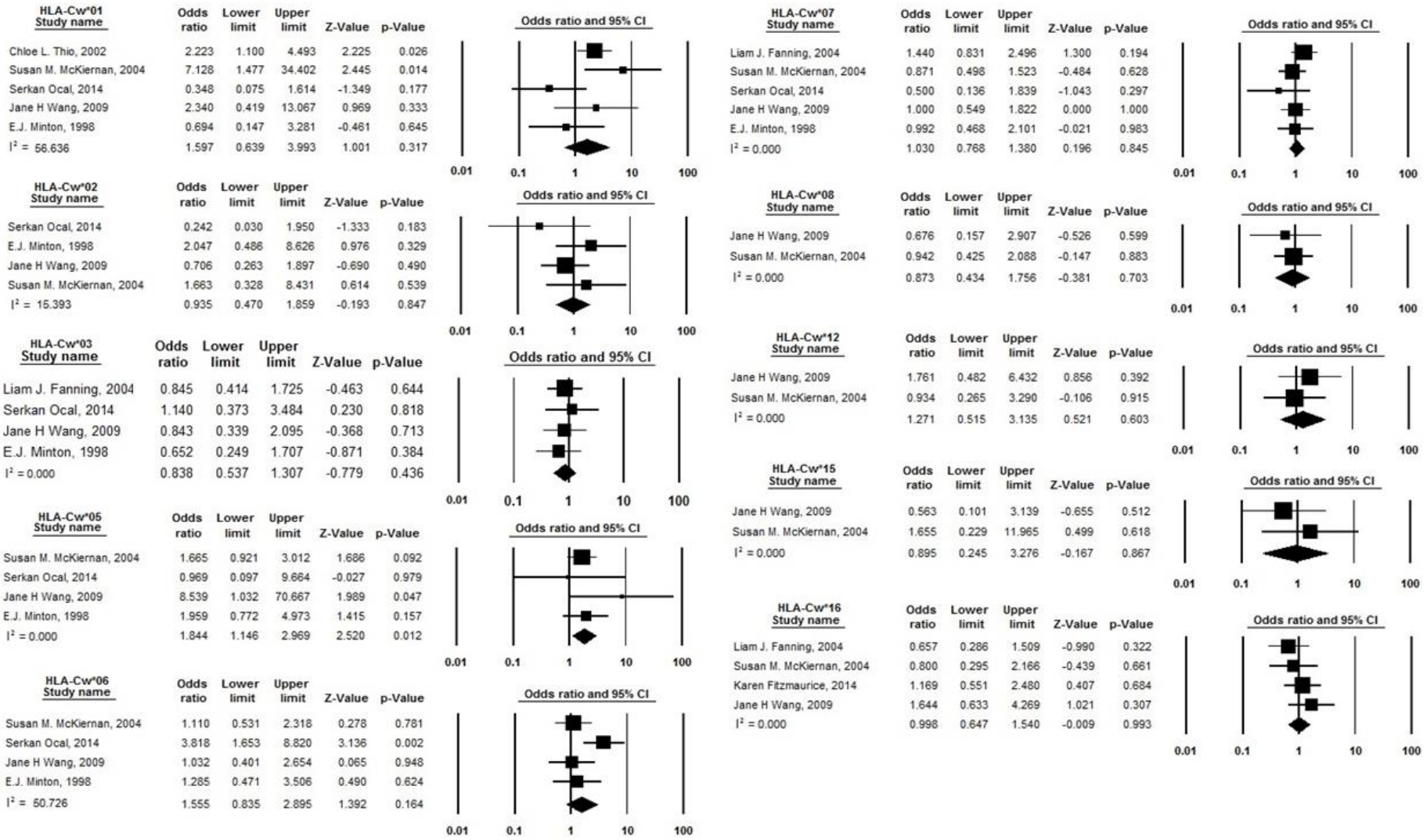
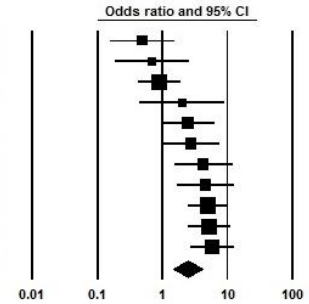


Figure 2.11 Meta-Analysis of HLA Class-I Cw to HCV Spontaneous Clearance

e) HLA Class II – DRB1

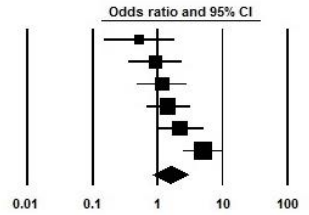
DRB1*01

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	0.489	0.155	1.540	-1.222	0.222
Laurent Alric, 1997	0.682	0.183	2.539	-0.571	0.568
L. Alric, 2000	0.884	0.416	1.878	-0.321	0.748
Leila K. Cheikrouhou, 2007	2.000	0.440	9.082	0.898	0.369
Jane H Wang, 2009	2.500	0.965	6.476	1.887	0.059
Liam J. Fanning, 2000	2.736	0.990	7.564	1.940	0.052
S Barret, 2001	4.257	1.520	11.924	2.757	0.006
Jeny R. C. Santos, 2007	4.590	1.699	12.401	3.005	0.003
Susan M. McKiernan, 2000	5.030	2.466	10.259	4.442	0.000
Susan M. McKiernan, 2004	5.190	2.468	10.913	4.343	0.000
Karen Fitzmaurice, 2014	5.793	2.665	12.539	4.434	0.000
I^2 = 69.465	2.500	1.492	4.190	3.479	0.001



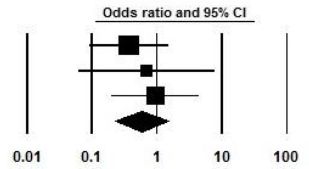
DRB1*0101

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Viviana Romero, 2008	0.520	0.148	1.821	-1.023	0.307
Alessandra Mangia, 2011	0.919	0.361	2.339	-0.177	0.859
Matthew E. Cramp, 1998	1.167	0.480	2.837	0.341	0.733
E.J. Minton, 1998	1.455	0.667	3.174	0.942	0.346
Chloe L. Thio, 2001	2.238	0.980	5.112	1.911	0.056
Susan M. McKiernan, 2000	5.030	2.466	10.259	4.442	0.000
I^2 = 67.115	1.563	0.838	2.915	1.404	0.160



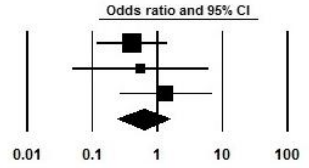
DRB1*0102

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Chloe L. Thio, 2001	0.373	0.092	1.512	-1.381	0.167
Alessandra Mangia, 2011	0.691	0.062	7.714	-0.300	0.764
Alessandra Mangia, 1999	0.944	0.199	4.475	-0.073	0.942
I^2 = 0.000	0.583	0.224	1.516	-1.107	0.268



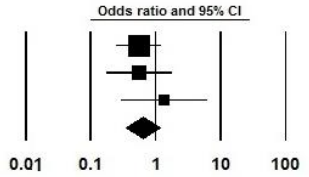
DRB1*0103

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Liam J. Fanning, 2000	0.406	0.116	1.423	-1.409	0.159
Matthew E. Cramp, 1998	0.552	0.049	6.251	-0.480	0.631
S Barret, 2001	1.347	0.261	6.956	0.356	0.722
I^2 = 0.000	0.619	0.246	1.557	-1.018	0.309



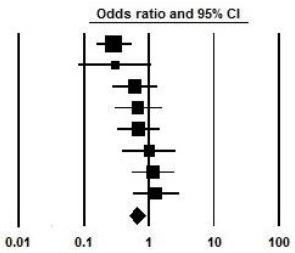
DRB1*02

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
L. Alric, 2000	0.549	0.249	1.210	-1.487	0.137
Laurent Alric, 1997	0.557	0.175	1.773	-0.991	0.322
Jose Azocar, 2003	1.364	0.298	6.247	0.400	0.689
I^2 = 0.000	0.635	0.348	1.157	-1.484	0.138



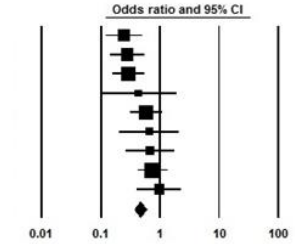
DRB1*03

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2000	0.289	0.154	0.542	-3.867	0.000
Jeny R. C. Santos, 2007	0.300	0.081	1.107	-1.807	0.071
L. Alric, 2000	0.605	0.273	1.339	-1.239	0.215
Jane H Wang, 2009	0.682	0.294	1.582	-0.891	0.373
S Barret, 2001	0.691	0.323	1.480	-0.951	0.341
Leila K. Cheikrouhou, 2007	1.007	0.390	2.602	0.014	0.989
Liam J. Fanning, 2000	1.152	0.538	2.467	0.364	0.716
Jose Azocar, 2003	1.275	0.561	2.899	0.580	0.562
I^2 = 47.614	0.652	0.489	0.868	-2.926	0.003



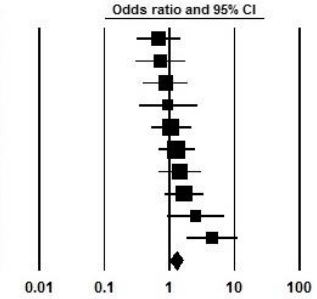
DRB1*03011

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Karen Fitzmaurice, 2014	0.246	0.119	0.508	-3.793	0.000
Susan M. McKiernan, 2004	0.280	0.143	0.550	-3.696	0.000
Susan M. McKiernan, 2000	0.289	0.154	0.542	-3.867	0.000
Alessandra Mangia, 1999	0.432	0.098	1.902	-1.110	0.267
Chloe L. Thio, 2001	0.580	0.308	1.093	-1.685	0.092
Laurent Alric, 1997	0.654	0.204	2.098	-0.714	0.475
Alessandra Mangia, 2011	0.670	0.260	1.727	-0.829	0.407
E.J. Minton, 1998	0.741	0.417	1.316	-1.022	0.307
Matthew E. Cramp, 1998	0.963	0.404	2.295	-0.085	0.932
I^2 = 42.577	0.466	0.362	0.599	-5.949	0.000



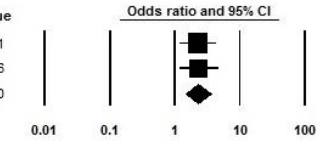
DRB1*04

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Liam J. Fanning, 2000	0.670	0.306	1.468	-1.000	0.317
E.J. Minton, 1998	0.729	0.305	1.744	-0.710	0.477
S Barret, 2001	0.866	0.395	1.899	-0.359	0.719
Jeny R. C. Santos, 2007	0.960	0.337	2.735	-0.076	0.939
L. Alric, 2000	1.052	0.519	2.134	0.140	0.888
Chloe L. Thio, 2001	1.284	0.670	2.459	0.754	0.451
Jane H Wang, 2009	1.455	0.690	3.069	0.985	0.325
Jose Azocar, 2003	1.701	0.845	3.424	1.489	0.137
Laurent Alric, 1997	2.529	0.935	6.843	1.827	0.068
Matthew E. Cramp, 1998	4.521	1.823	11.213	3.255	0.001
I^2 = 42.716	1.276	0.993	1.640	1.908	0.056



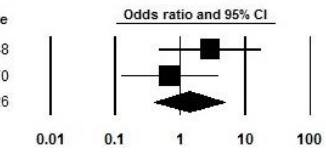
DRB1*0401

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2000	2.274	1.205	4.292	2.535	0.011
Karen Fitzmaurice, 2014	2.320	1.169	4.604	2.407	0.016
I^2 = 0.000	2.295	1.440	3.657	3.495	0.000



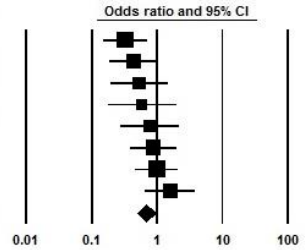
DRB1*0403

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 1999	2.902	0.476	17.699	1.155	0.248
Alessandra Mangia, 2011	0.688	0.123	3.839	-0.426	0.670
I^2 = 21.783	1.363	0.392	4.737	0.487	0.626



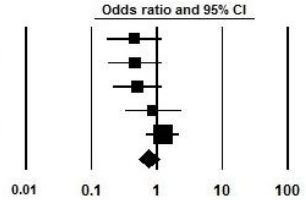
DRB1*07

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
L. Alric, 2000	0.322	0.148	0.700	-2.861	0.004
Jane H Wang, 2009	0.435	0.181	1.045	-1.862	0.063
Laurent Alric, 1997	0.532	0.195	1.450	-1.234	0.217
Leila K. Cheikrouhou, 2007	0.589	0.179	1.941	-0.870	0.384
Jeny R. C. Santos, 2007	0.770	0.272	2.183	-0.492	0.623
S Barret, 2001	0.877	0.386	1.992	-0.314	0.754
Liam J. Fanning, 2000	0.987	0.467	2.086	-0.034	0.973
Jose Azocar, 2003	1.571	0.647	3.813	0.998	0.318
I^2 = 27.450	0.684	0.499	0.936	-2.371	0.018



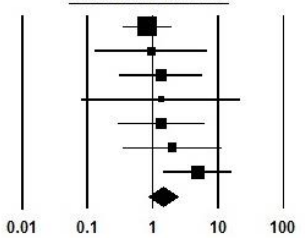
DRB1*0701

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 1999	0.456	0.174	1.197	-1.595	0.111
E.J. Minton, 1998	0.458	0.177	1.186	-1.608	0.108
Matthew E. Cramp, 1998	0.507	0.213	1.206	-1.536	0.125
Alessandra Mangia, 2011	0.874	0.326	2.343	-0.268	0.789
Chloe L. Thio, 2001	1.229	0.688	2.195	0.697	0.486
I^2 = 30.032	0.753	0.523	1.084	-1.527	0.127



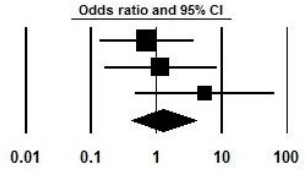
DRB1*08

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Chloe L. Thio, 2001	0.825	0.351	1.939	-0.441	0.659
Liam J. Fanning, 2000	0.939	0.128	6.882	-0.062	0.951
Jeny R. C. Santos, 2007	1.330	0.310	5.703	0.384	0.701
S Barret, 2001	1.333	0.081	21.883	0.201	0.840
Jose Azocar, 2003	1.364	0.298	6.247	0.400	0.689
E.J. Minton, 1998	1.985	0.348	11.314	0.772	0.440
B.S. de Almeida, 2010	4.889	1.462	16.346	2.577	0.010
I^2 = 0.000	1.441	0.855	2.430	1.373	0.170



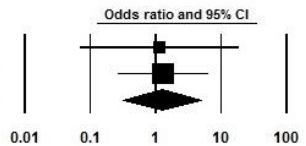
DRB1*09

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	0.713	0.135	3.763	-0.399	0.690
Jane H Wang, 2009	1.146	0.158	8.302	0.135	0.893
Jeny R. C. Santos, 2007	5.482	0.478	62.899	1.367	0.172
I^2 = 0.000	1.288	0.416	3.983	0.439	0.661



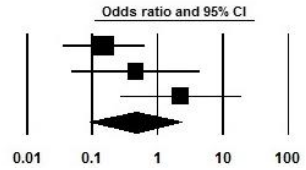
DRB1*0901

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Matthew E. Cramp, 1998	1.125	0.069	18.412	0.083	0.934
Chloe L. Thio, 2001	1.300	0.267	6.330	0.325	0.745
I^2 = 0.000	1.255	0.317	4.976	0.323	0.746



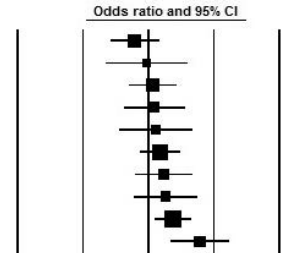
DRB1*1001

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 1999	0.150	0.035	0.636	-2.573	0.010
Alessandra Mangia, 2011	0.457	0.047	4.452	-0.674	0.500
Chloe L. Thio, 2001	2.239	0.267	18.764	0.743	0.457
I^2 = 53.346	0.464	0.091	2.355	-0.927	0.354



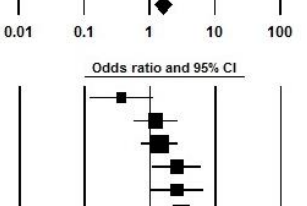
DRB1*11

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	0.616	0.261	1.455	-1.105	0.269
Liam J. Fanning, 2000	0.938	0.224	3.923	-0.088	0.930
Jane H Wang, 2009	1.163	0.497	2.722	0.348	0.728
Jeny R. C. Santos, 2007	1.240	0.420	3.660	0.389	0.697
S Barret, 2001	1.298	0.355	4.746	0.394	0.693
B.S. de Almeida, 2010	1.514	0.739	3.102	1.133	0.257
Rebecca A. Harris, 2008	1.700	0.614	4.707	1.021	0.307
Matthew E. Cramp, 1998	1.838	0.603	5.601	1.071	0.284
L. Alric, 2000	2.378	1.244	4.546	2.620	0.009
Laurent Alric, 1997	6.133	2.184	17.223	3.443	0.001
I^2 = 36.453	1.591	1.187	2.130	3.112	0.002



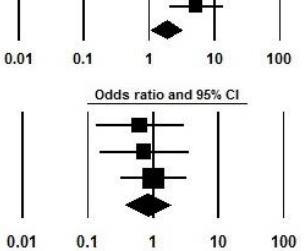
DRB1*1101

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Viviana Romero, 2008	0.363	0.119	1.107	-1.782	0.075
Chloe L. Thio, 2001	1.207	0.557	2.615	0.477	0.633
Alessandra Mangia, 1999	1.386	0.725	2.649	0.987	0.323
Alessandra Mangia, 2011	2.550	1.067	6.095	2.105	0.035
Laurent Alric, 1997	2.571	1.012	6.534	1.984	0.047
L. Alric, 2000	2.974	1.498	5.906	3.114	0.002
E.J. Minton, 1998	5.167	2.012	13.269	3.413	0.001
I^2 = 65.727	1.856	1.086	3.173	2.260	0.024



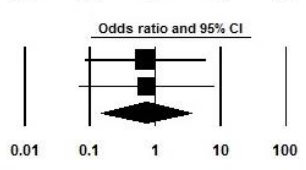
DRB1*1102

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
L. Alric, 2000	0.628	0.132	2.990	-0.584	0.559
Laurent Alric, 1997	0.719	0.149	3.472	-0.411	0.681
Chloe L. Thio, 2001	1.018	0.319	3.248	0.030	0.976
I^2 = 0.000	0.819	0.367	1.825	-0.488	0.625



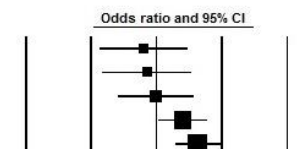
DRB1*1103

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 1999	0.705	0.084	5.935	-0.322	0.748
Chloe L. Thio, 2001	0.739	0.067	8.179	-0.247	0.805
I^2 = 0.000	0.720	0.146	3.545	-0.404	0.686



DRB1*1104

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
L. Alric, 2000	0.628	0.132	2.990	-0.584	0.559
Laurent Alric, 1997	0.719	0.149	3.472	-0.411	0.681
Chloe L. Thio, 2001	0.986	0.258	3.769	-0.021	0.984
Alessandra Mangia, 2011	2.550	1.067	6.095	2.105	0.035
Alessandra Mangia, 1999	4.303	1.933	9.578	3.575	0.000
I^2 = 53.371	1.683	0.788	3.593	1.345	0.179



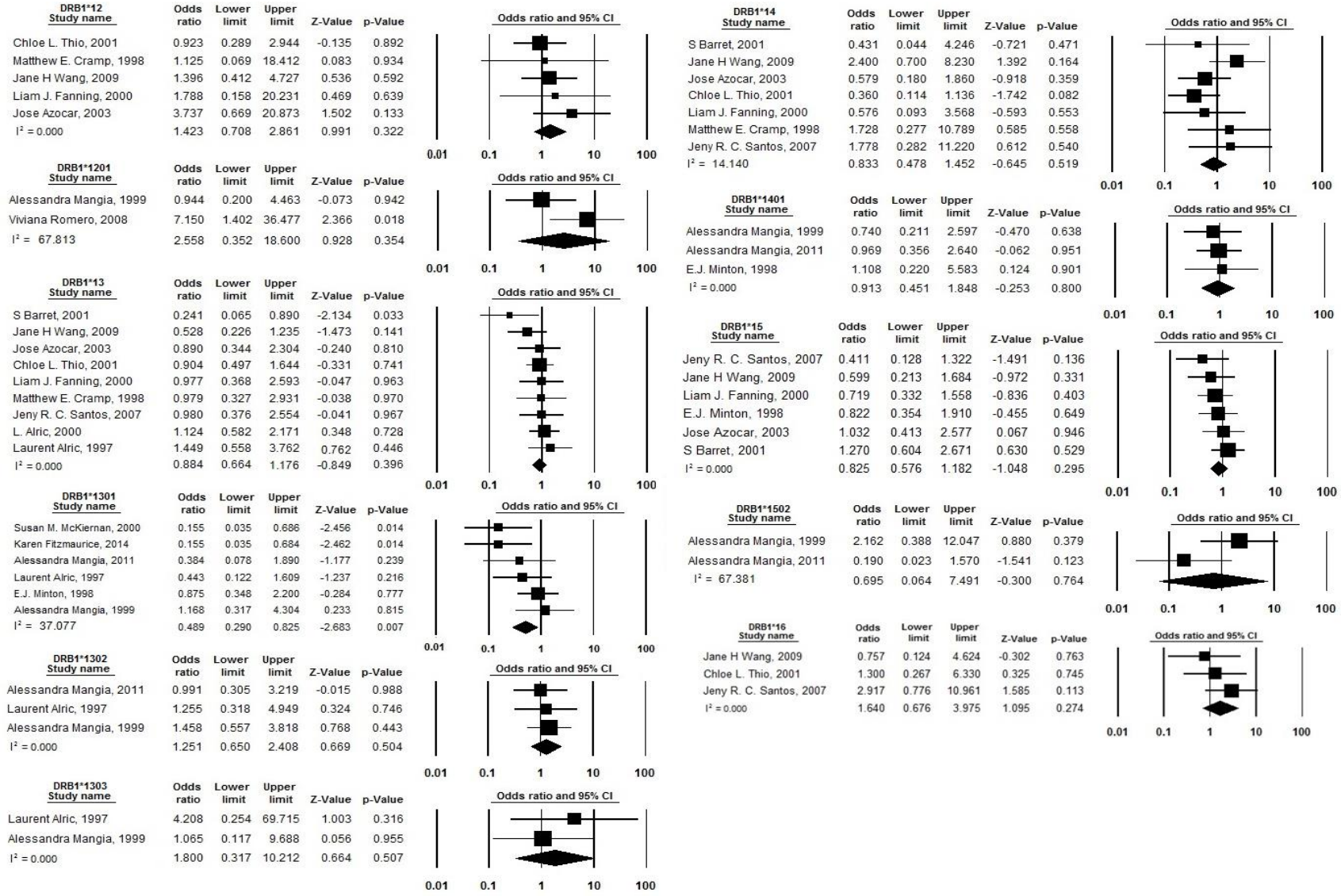


Figure 2.12 Meta-Analysis of HLA Class-II DRB1 to HCV Spontaneous Clearance

f) HLA Class II – DQA1

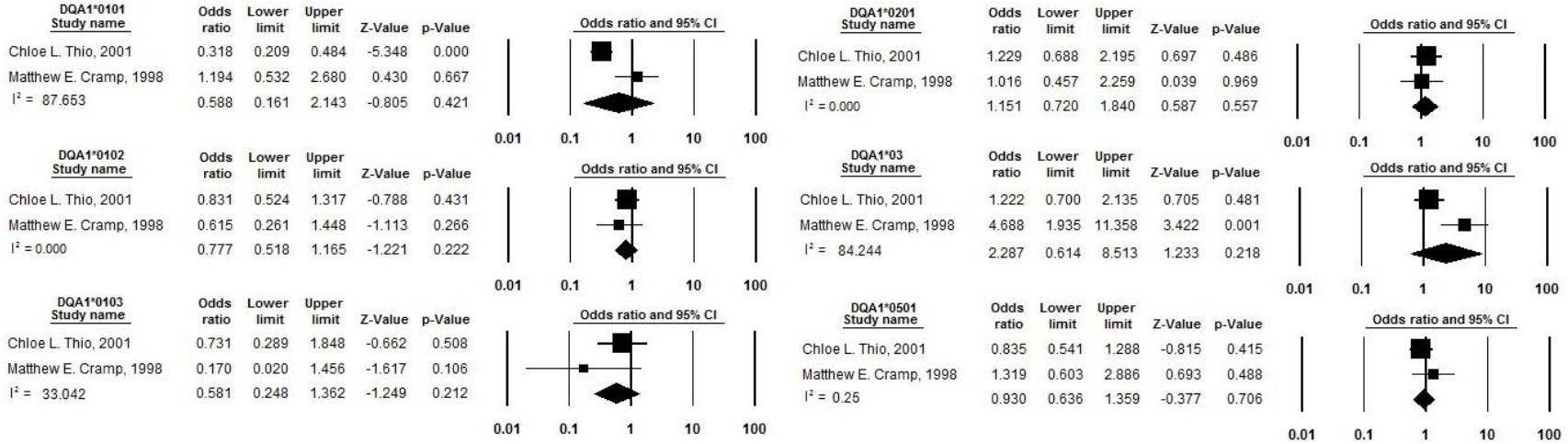
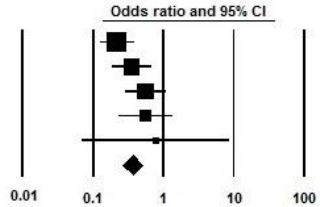


Figure 2.13 Meta-Analysis of HLA Class-II DQA1 to HCV Spontaneous Clearance

g) HLA Class II – DQB1

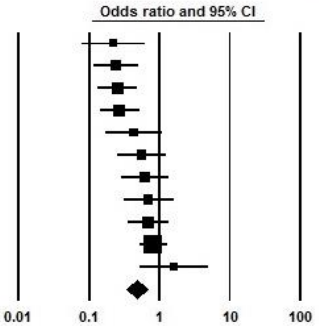
DQB1*02

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2000	0.218	0.123	0.386	-5.233	0.000
L. Alric, 2000	0.354	0.182	0.688	-3.061	0.002
Jane H Wang, 2009	0.559	0.282	1.107	-1.669	0.095
Jeny R. C. Santos, 2007	0.560	0.230	1.362	-1.279	0.201
Jose Azocar, 2003	0.772	0.069	8.648	-0.210	0.834
I^2 = 32.188	0.362	0.259	0.506	-5.943	0.000



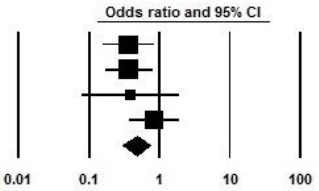
DQB1*0201

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 1999	0.220	0.077	0.629	-2.824	0.005
Karen Fitzmaurice, 2014	0.238	0.115	0.492	-3.877	0.000
Susan M. McKiernan, 2000	0.253	0.133	0.481	-4.198	0.000
Susan M. McKiernan, 2004	0.270	0.140	0.520	-3.911	0.000
Laurent Alric, 1997	0.435	0.172	1.099	-1.760	0.078
E.J. Minton, 1998	0.556	0.252	1.226	-1.455	0.146
Matthew E. Cramp, 1998	0.618	0.284	1.345	-1.212	0.225
Alessandra Mangia, 2011	0.699	0.310	1.578	-0.862	0.389
Liam J. Fanning, 2000	0.700	0.360	1.361	-1.052	0.293
Chloe L. Thio, 2001	0.810	0.520	1.261	-0.933	0.351
Jose Azocar, 2003	1.587	0.514	4.897	0.803	0.422
I^2 = 59.592	0.477	0.335	0.679	-4.117	0.000



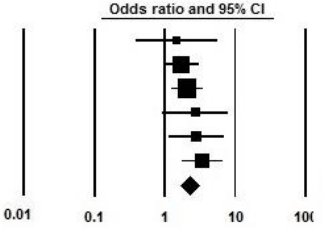
DQB1*0202

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Karen Fitzmaurice, 2014	0.362	0.158	0.830	-2.401	0.016
Susan M. McKiernan, 2000	0.366	0.170	0.789	-2.565	0.010
Alessandra Mangia, 2011	0.384	0.078	1.890	-1.177	0.239
Jose Azocar, 2003	0.837	0.373	1.878	-0.432	0.666
I^2 = 0.000	0.470	0.302	0.733	-3.333	0.001



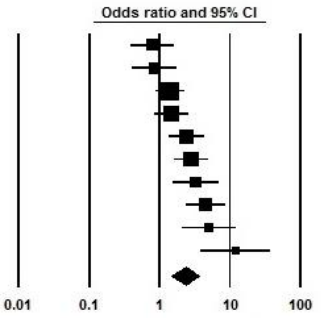
DQB1*03

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	1.463	0.381	5.614	0.555	0.579
Jane H Wang, 2009	1.724	0.970	3.064	1.857	0.063
Susan M. McKiernan, 2000	2.081	1.231	3.517	2.737	0.006
Rebecca A. Harris, 2008	2.700	0.929	7.846	1.825	0.068
Jeny R. C. Santos, 2007	2.830	1.140	7.023	2.243	0.025
L. Alric, 2000	3.426	1.742	6.738	3.568	0.000
I^2 = 0.000	2.256	1.679	3.030	5.403	0.000



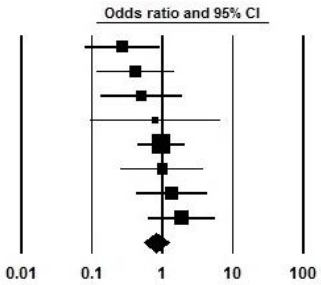
DQB1*0301

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Liam J. Fanning, 2000	0.780	0.380	1.601	-0.677	0.498
Jose Azocar, 2003	0.841	0.407	1.738	-0.467	0.640
Chloe L. Thio, 2001	1.391	0.869	2.226	1.375	0.169
Susan M. McKiernan, 2000	1.455	0.837	2.529	1.330	0.184
Alessandra Mangia, 2011	2.449	1.362	4.403	2.992	0.003
Alessandra Mangia, 1999	2.795	1.584	4.932	3.547	0.000
E.J. Minton, 1998	3.273	1.515	7.071	3.017	0.003
L. Alric, 2000	4.504	2.386	8.502	4.643	0.000
Matthew E. Cramp, 1998	5.087	2.098	12.334	3.600	0.000
Laurent Alric, 1997	12.024	3.809	37.956	4.240	0.000
I^2 = 76.779	2.303	1.487	3.567	3.739	0.000



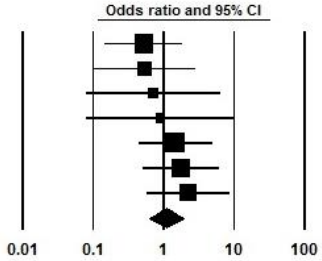
DQB1*0302

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
L. Alric, 2000	0.270	0.079	0.925	-2.083	0.037
E.J. Minton, 1998	0.413	0.117	1.456	-1.375	0.169
Alessandra Mangia, 2011	0.505	0.131	1.951	-0.991	0.322
Alessandra Mangia, 1999	0.791	0.094	6.675	-0.215	0.829
Chloe L. Thio, 2001	0.959	0.451	2.040	-0.109	0.913
Laurent Alric, 1997	0.992	0.254	3.871	-0.012	0.991
Matthew E. Cramp, 1998	1.361	0.424	4.368	0.518	0.604
Jose Azocar, 2003	1.877	0.634	5.557	1.137	0.256
I^2 = 12.051	0.822	0.545	1.240	-0.935	0.350



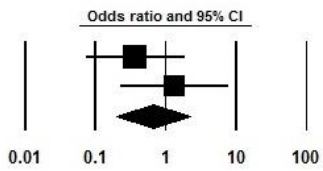
DQB1*0303

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Matthew E. Cramp, 1998	0.522	0.147	1.855	-1.005	0.315
Alessandra Mangia, 2011	0.546	0.104	2.870	-0.715	0.475
Laurent Alric, 1997	0.717	0.079	6.469	-0.296	0.767
Jose Azocar, 2003	0.899	0.080	10.086	-0.086	0.931
E.J. Minton, 1998	1.455	0.434	4.878	0.608	0.544
Chloe L. Thio, 2001	1.753	0.497	6.186	0.872	0.383
L. Alric, 2000	2.211	0.573	8.532	1.152	0.249
I^2 = 0.000	1.109	0.635	1.935	0.364	0.716

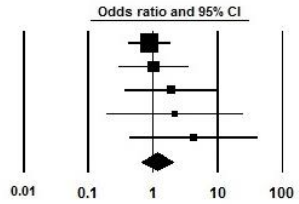


DQB1*04

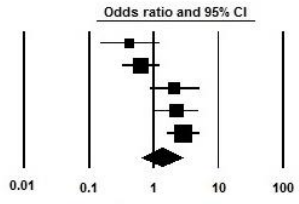
Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jane H Wang, 2009	0.368	0.073	1.861	-1.209	0.227
Jeny R. C. Santos, 2007	1.315	0.228	7.592	0.306	0.760
I^2 = 8.497	0.662	0.201	2.176	-0.680	0.497



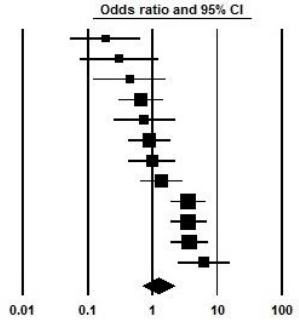
DQB1*0402 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Chloe L. Thio, 2001	0.881	0.411	1.889	-0.326	0.745
Jose Azocar, 2003	1.030	0.292	3.633	0.046	0.963
Alessandra Mangia, 1999	1.938	0.367	10.233	0.779	0.436
Liam J. Fanning, 2000	2.177	0.193	24.553	0.629	0.529
Alessandra Mangia, 2011	4.263	0.437	41.599	1.247	0.212
I^2 = 0.000	1.158	0.655	2.049	0.505	0.614



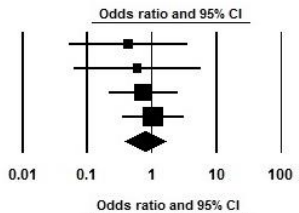
DQB1*05 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Laurent Alric, 1997	0.421	0.146	1.214	-1.601	0.109
L. Alric, 2000	0.635	0.329	1.227	-1.352	0.177
Jeny R. C. Santos, 2007	2.110	0.877	5.075	1.668	0.095
Jane H Wang, 2009	2.273	1.053	4.905	2.092	0.036
Susan M. McKiernan, 2000	2.859	1.590	5.140	3.510	0.000
I^2 = 78.281	1.348	0.648	2.807	0.799	0.424



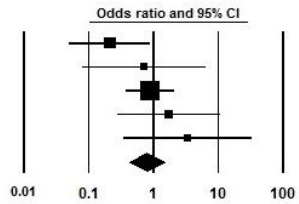
DQB1*0501 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	0.186	0.054	0.638	-2.675	0.007
Viviana Romero, 2008	0.300	0.074	1.216	-1.686	0.092
Laurent Alric, 1997	0.443	0.122	1.609	-1.237	0.216
Alessandra Mangia, 2011	0.659	0.294	1.478	-1.012	0.312
Alessandra Mangia, 1999	0.746	0.250	2.227	-0.525	0.599
Liam J. Fanning, 2000	0.910	0.428	1.933	-0.245	0.806
Matthew E. Cramp, 1998	0.986	0.428	2.272	-0.033	0.974
E.J. Minton, 1998	1.379	0.641	2.966	0.822	0.411
Susan M. McKiernan, 2000	3.528	1.894	6.572	3.972	0.000
Susan M. McKiernan, 2004	3.590	1.883	6.843	3.884	0.000
Karen Fitzmaurice, 2014	3.720	1.906	7.262	3.849	0.000
Chloe L. Thio, 2001	6.172	2.440	15.611	3.844	0.000
I^2 = 80.885	1.246	0.709	2.192	0.765	0.444



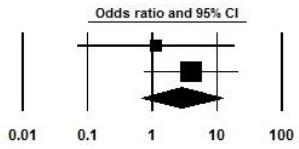
DQB1*0502 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Laurent Alric, 1997	0.432	0.052	3.609	-0.775	0.438
Jose Azocar, 2003	0.595	0.061	5.810	-0.447	0.655
Chloe L. Thio, 2001	0.735	0.218	2.478	-0.497	0.619
Alessandra Mangia, 2011	1.044	0.350	3.112	0.077	0.938
I^2 = 0.000	0.788	0.384	1.620	-0.647	0.518



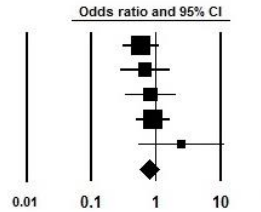
DQB1*0503 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Chloe L. Thio, 2001	0.210	0.050	0.886	-2.124	0.034
Laurent Alric, 1997	0.717	0.079	6.469	-0.296	0.767
Alessandra Mangia, 2011	0.881	0.365	2.126	-0.282	0.778
Matthew E. Cramp, 1998	1.728	0.277	10.789	0.585	0.558
Liam J. Fanning, 2000	3.313	0.337	32.599	1.027	0.305
I^2 = 27.655	0.787	0.416	1.487	-0.739	0.460



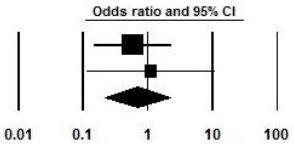
DQB1*0605 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Matthew E. Cramp, 1998	1.125	0.069	18.412	0.083	0.934
Laurent Alric, 1997	4.000	0.755	21.190	1.630	0.103
I^2 = 0.000	2.867	0.685	12.005	1.442	0.149



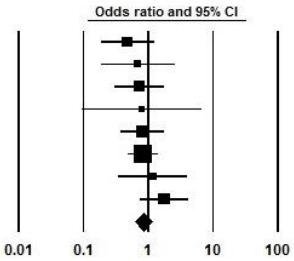
DQB1*06 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Jane H Wang, 2009	0.576	0.302	1.098	-1.676	0.094
Jeny R. C. Santos, 2007	0.670	0.275	1.633	-0.881	0.378
Laurent Alric, 1997	0.821	0.328	2.055	-0.421	0.674
L. Alric, 2000	0.887	0.479	1.643	-0.381	0.703
Jose Azocar, 2003	2.474	0.540	11.338	1.166	0.244
I^2 = 0.000	0.778	0.545	1.110	-1.386	0.166



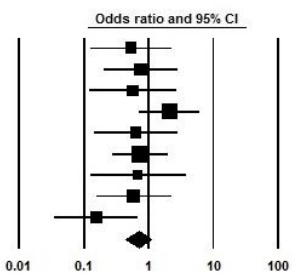
DQB1*0601 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 2011	0.582	0.147	2.307	-0.770	0.441
Chloe L. Thio, 2001	1.111	0.115	10.749	0.091	0.928
I^2 = 0.000	0.693	0.213	2.248	-0.611	0.541



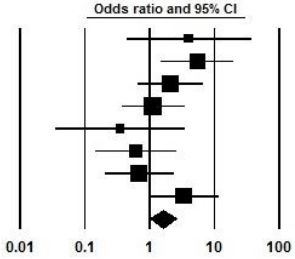
DQB1*0602 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Matthew E. Cramp, 1998	0.476	0.183	1.237	-1.523	0.128
Laurent Alric, 1997	0.691	0.183	2.607	-0.546	0.585
E.J. Minton, 1998	0.729	0.305	1.744	-0.710	0.477
Alessandra Mangia, 1999	0.791	0.094	6.675	-0.215	0.829
Liam J. Fanning, 2000	0.804	0.371	1.741	-0.553	0.580
Chloe L. Thio, 2001	0.818	0.476	1.406	-0.727	0.467
Alessandra Mangia, 2011	1.165	0.345	3.932	0.246	0.806
Jose Azocar, 2003	1.754	0.736	4.180	1.268	0.205
I^2 = 0.000	0.848	0.621	1.159	-1.034	0.301



DQB1*0603 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Matthew E. Cramp, 1998	0.533	0.126	2.255	-0.855	0.393
Laurent Alric, 1997	0.750	0.198	2.844	-0.423	0.672
E.J. Minton, 1998	0.569	0.122	2.650	-0.718	0.473
Alessandra Mangia, 1999	2.076	0.703	6.130	1.322	0.186
Liam J. Fanning, 2000	0.627	0.143	2.748	-0.619	0.536
Chloe L. Thio, 2001	0.732	0.270	1.986	-0.613	0.540
Alessandra Mangia, 2011	0.688	0.123	3.839	-0.426	0.670
Jose Azocar, 2003	0.584	0.154	2.219	-0.790	0.430
Susan M. McKiernan, 2000	0.155	0.035	0.686	-2.456	0.014
I^2 = 1.639	0.694	0.446	1.079	-1.624	0.104



DQB1*0604 Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
Matthew E. Cramp, 1998	4.000	0.433	36.956	1.222	0.222
Laurent Alric, 1997	5.432	1.500	19.669	2.578	0.010
E.J. Minton, 1998	2.083	0.663	6.546	1.256	0.209
Alessandra Mangia, 1999	1.130	0.366	3.488	0.213	0.832
Liam J. Fanning, 2000	0.348	0.035	3.443	-0.903	0.367
Chloe L. Thio, 2001	0.612	0.144	2.597	-0.666	0.506
Alessandra Mangia, 2011	0.681	0.199	2.329	-0.612	0.540
Jose Azocar, 2003	3.356	0.951	11.842	1.882	0.060
I^2 = 37.880	1.601	0.989	2.590	1.915	0.055



h) KIR Alleles

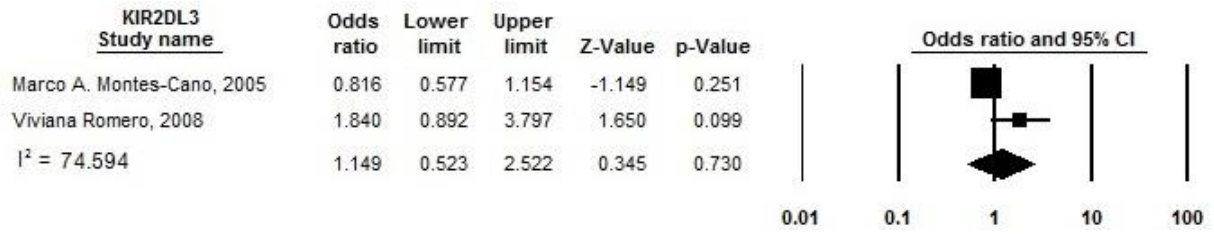


Figure 2.14 Meta-Analysis of KIR Alleles to HCV Spontaneous Clearance

i) CCR5 Genotype

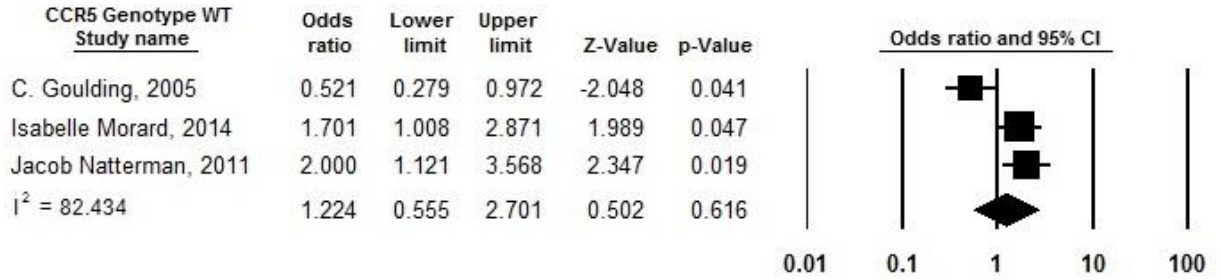


Figure 2.15 Meta-Analysis of CR5 Genotype to HCV Spontaneous Clearance

I found similar results when the analysis of risk factors was restricted to studies with a minimum of 12 months follow-up (table 3.2 below).

Table 2.4 Meta-Analysis of Host Genetic Determinants on SVC: All Studies vs Studies with Minimum 12 Months Follow Up

Host Genetic Variables	Σ of studies	I^2	All studies			Σ of studies	I^2	>=12 Months		
			OR	Lower CI	Upper CI			OR	Lower CI	Upper CI
IL28B rs12979860	31	68.607	3.267	2.679	3.984	12	59.801	3.043	2.358	3.926
IL28B rs 8099917	14	17.494	2.83	2.362	3.391	8	4.711	2.912	2.256	3.759
IL28B rs8103142	2	0	4.064	2.643	6.248	1	-	4.23	2.46	7.28
IL18-607A	2	57.406	2.45	0.953	6.297	1	-	1.385	0.488	3.929
IL18-137C	2	0	2.158	1.282	3.632	1	-	1.824	0.72	4.622
CCR5 Genotype WT vs WT/ 32	3	82.434	1.224	0.555	2.701	1	-	2	1.1	3.5
HLA-A*01	5	4.588	0.855	0.647	1.13	2	58.292	0.997	0.549	1.811
HLA-A*02	5	0	1.145	0.896	1.464	2	0	1.053	0.72	1.54
HLA-A*03	5	59.378	1.577	1.01	2.462	1	-	2.761	1.512	5.042
HLA-A*11	6	57.362	1.182	0.726	1.926	2	61.302	0.714	0.307	1.66
HLA-A*23	5	0	0.606	0.402	0.914	1	-	0.715	0.213	2.398
HLA-A*24	4	0	1.13	0.769	1.66	1	-	0.73	0.245	2.176
HLA-A*25	2	0	0.31	0.064	1.507	1	-	0.196	0.024	1.592
HLA-A*26	2	0	1.351	0.479	3.81	1	-	1.624	0.32	8.228
HLA-A*29	4	19.173	0.671	0.355	1.268	1	-	1.118	0.461	2.711
HLA-A*30	3	73.817	1.223	0.332	4.501					
HLA-A*31	2	0	0.968	0.344	2.724	1	-	0.536	0.106	2.716
HLA-A*32	3	0	1.581	0.778	3.216	1	-	0.983	0.344	2.807
HLA-A*33	1	-	0.375	0.038	3.661					
HLA-A*68	3	0	1.1	0.617	1.961	1	-	1.247	0.417	3.724
HLA-A*74	1	-	0.375	0.038	3.661					

Host Genetic Variables	Σ of studies	I^2	All studies			Σ of studies	I^2	>=12 Months		
			OR	Lower CI	Upper CI			OR	Lower CI	Upper CI
HLA-B*07	5	24.314	0.889	0.647	1.22	2	76.051	0.906	0.355	2.308
HLA-B*08	4	78.479	0.762	0.308	1.885	1	-	0.374	0.199	0.701
HLA-B*15	3	5.376	1.59	0.993	2.548	1	-	1.4	0.4	4.7
HLA-B*27	5	27.927	2.291	1.446	3.629	1	-	7.46	2.04	27.27
HLA-B*35	6	13.997	1.038	0.803	1.342	2	0	0.701	0.381	1.29
HLA-B*38	3	75.882	0.858	0.063	11.611	1	-	0.8	0.07	9.2
HLA-B*5301	1	-	0.577	0.326	1.022					
HLA-B*57	3	0	1.781	1.286	2.466	1	-	1.7	0.7	4.3
HLA-Cw*01	5	56.636	1.597	0.639	3.993					
HLA-Cw*02	4	15.393	0.935	0.47	1.859	1	-	1.663	0.328	8.43
HLA-Cw*03	4	0	0.838	0.537	1.307	1	-	0.845	0.414	1.725
HLA-Cw*04	6	0	0.601	0.463	0.78	2	56.971	0.425	0.157	1.149
HLA-Cw*05	4	0	1.844	1.146	2.969	1	-	1.665	0.921	3.013
HLA-Cw*06	4	50.726	1.555	0.835	2.895	1	-	1.11	0.532	2.321
HLA-Cw*07	7	41.623	1.995	1.362	2.924	1	-	7.128	1.477	34.405
HLA-Cw*08	2	0	0.873	0.434	1.756	1	-	0.942	0.425	2.088
HLA-Cw*12	2	0	1.271	0.515	3.135	1	-	0.934	0.265	3.288
HLA-Cw*15	2	0	0.895	0.245	3.276	1	-	1.655	0.229	11.969
HLA-Cw*16	4	0	0.998	0.647	1.54	2	0	0.712	0.376	1.349
DRB1*01	11	69.465	2.5	1.492	4.19	7	70.323	2.738	1.463	5.125
DRB1*0101	6	67.115	1.563	0.838	2.915	3	80.869	1.811	0.596	5.502
DRB1*0102	3	0	0.583	0.224	1.516	2	0	0.861	0.233	3.185
DRB1*0103	3	0	0.619	0.246	1.557	3	0	0.619	0.246	1.557
DRB1*02	3	0	0.635	0.348	1.157	2	0	0.552	0.287	1.059

Host Genetic Variables	Σ of studies	I^2	All studies			Σ of studies	I^2	>=12 Months		
			OR	Lower CI	Upper CI			OR	Lower CI	Upper CI
DRB1*03	8	47.614	0.652	0.489	0.868	5	54.831	0.548	0.32	0.939
DRB1*03011	9	42.577	0.466	0.362	0.599	6	35.333	0.426	0.302	0.603
DRB1*0302	1	-	0.607	0.217	1.7					
DRB1*04	10	42.716	1.276	0.993	1.64	6	62.563	1.332	0.751	2.361
DRB1*0401	2	0	2.295	1.44	3.657	1	-	2.274	1.205	4.293
DRB1*0403	2	21.783	1.363	0.392	4.737	2	21.783	1.363	0.392	4.737
DRB1*0406	1	-	1.065	0.117	9.681	1	-	1.065	0.117	9.681
DRB1*06	1	-	1.607	0.684	3.775					
DRB1*07	8	27.45	0.684	0.499	0.936	5	21.722	0.648	0.442	0.95
DRB1*0701	5	30.032	0.753	0.523	1.084	3	0	0.577	0.337	0.99
DRB1*08	7	0	1.441	0.855	2.43	3	0	1.2	0.406	3.547
DRB1*09	3	0	1.288	0.416	3.983	1	-	5.482	0.478	62.928
DRB1*0901	2	0	1.255	0.317	4.976	1	-	1.125	0.069	18.482
DRB1*1001	3	53.346	0.464	0.091	2.355	2	0	0.207	0.061	0.7
DRB1*11	10	36.453	1.591	1.187	2.13	6	28.468	2.122	1.414	3.186
DRB1*1101	7	65.727	1.856	1.086	3.173	4	0	2.173	1.488	3.173
DRB1*1102	3	0	0.819	0.367	1.825	2	0	0.672	0.222	2.034
DRB1*1103	2	0	0.72	0.146	3.545	1	-	0.705	0.084	5.954
DRB1*1104	5	53.371	1.683	0.788	3.593	4	58.034	1.858	0.78	4.429
DRB1*12	5	0	1.423	0.708	2.861	2	0	1.465	0.235	9.155
DRB1*1201	2	67.813	2.558	0.352	18.6	1	-	0.944	0.2	4.471
DRB1*13	9	0	0.884	0.664	1.176	6	4.492	0.969	0.663	1.415
DRB1*1301	6	37.077	0.489	0.29	0.825	4	26.192	0.452	0.224	0.91
DRB1*1302	3	0	1.251	0.65	2.408	3	0	1.251	0.65	2.408

Host Genetic Variables	Σ of studies	I^2	All studies			Σ of studies	I^2	>=12 Months		
			OR	Lower CI	Upper CI			OR	Lower CI	Upper CI
DRB1*1303	2	0	1.8	0.317	10.212	2	0	1.8	0.317	10.212
DRB1*14	7	14.14	0.833	0.478	1.452	4	0	1.005	0.385	2.627
DRB1*1401	3	0	0.913	0.451	1.848	2	0	0.872	0.399	1.909
DRB1*15	6	0	0.825	0.576	1.182	3	28.052	0.833	0.512	1.356
DRB1*1501	1	-	1.422	0.515	3.93	1	-	1.422	0.515	3.93
DRB1*1502	2	67.381	0.695	0.064	7.491	2	67.381	0.695	0.064	7.491
DRB1*16	3	0	1.64	0.676	3.975	1	-	2.917	0.776	10.956
DQA1*0101	2	87.653	0.588	0.161	2.143	1	-	1.194	0.532	2.681
DQA1*0102	2	0	0.777	0.518	1.165	1	-	0.615	0.261	1.447
DQA1*0103	2	33.042	0.581	0.248	1.362	1	-	0.17	0.02	1.467
DQA1*0201	2	0	1.151	0.72	1.84	1	-	1.016	0.457	2.26
DQA1*03	2	84.244	2.287	0.614	8.513	1	-	4.688	1.935	11.358
DQA1*0401	1	-	0.853	0.422	1.725					
DQA1*0501	2	0.25	0.93	0.636	1.359	1	-	1.319	0.603	2.886
DQB1*02	5	32.188	0.362	0.259	0.506	3	39.701	0.308	0.209	0.455
DQB1*0201	11	59.592	0.477	0.335	0.679	7	41.749	0.412	0.31	0.548
DQB1*0202	4	0	0.47	0.302	0.733	2	0	0.369	0.185	0.738
DQB1*03	6	0	2.256	1.679	3.03	3	0	2.562	1.757	3.736
DQB1*0301	10	76.779	2.303	1.487	3.567	7	77.659	2.773	1.601	4.805
DQB1*0302	8	12.051	0.822	0.545	1.24	5	0.509	0.674	0.367	1.238
DQB1*0303	7	0	1.109	0.635	1.935	4	0	0.862	0.404	1.84
DQB1*0304	1	-	1.205	0.197	7.367					
DQB1*04	2	8.497	0.662	0.201	2.176	1	-	1.315	0.228	7.599
DQB1*0402	5	0	1.158	0.655	2.049	3	0	2.457	0.759	7.955

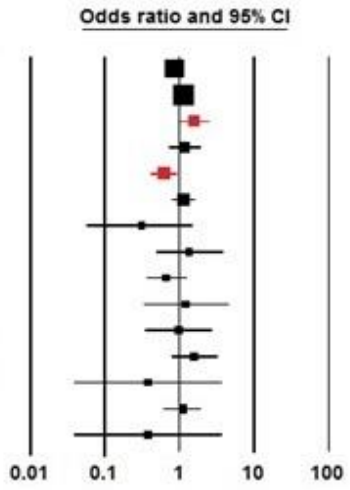
Host Genetic Variables	Σ of studies	I^2	All studies			Σ of studies	I^2	>=12 Months		
			OR	Lower CI	Upper CI			OR	Lower CI	Upper CI
DQB1*05	5	78.281	1.348	0.648	2.807	4	82.32	1.171	0.471	2.914
DQB1*0501	12	80.885	1.246	0.709	2.192	7	76.85	1.232	0.648	2.343
DQB1*0502	4	0	0.788	0.384	1.62	2	0	0.868	0.329	2.292
DQB1*0503	5	27.655	0.787	0.416	1.487	4	0	1.084	0.533	2.205
DQB1*06	5	0	0.778	0.545	1.11	3	0	0.813	0.521	1.266
DQB1*0601	2	0	0.693	0.213	2.248	1	-	0.582	0.147	2.309
DQB1*0602	8	0	0.848	0.621	1.159	5	0	0.729	0.449	1.186
DQB1*0603	9	1.639	0.694	0.446	1.079	6	37.305	0.722	0.411	1.269
DQB1*0604	8	37.88	1.601	0.989	2.59	5	49.837	1.471	0.776	2.788
DQB1*0605	2	0	2.867	0.685	12.005	2	0	2.867	0.685	12.005
KIR2DL2	1	-	1.405	0.947	2.083					
KIR2DL3	2	74.594	1.149	0.523	2.522					
KIR2DS3	1	-	0.23	0.085	0.617					

Red typing: statistically significant based on meta-analysis result ------ Blue typing: statistically significant based on 1 study

I was unable to conduct meta-analysis of some of the host genetic candidates because some genetic determinants were only included in single studies or different categories were used to calculate the OR, the summary of host genetic factors found which could not be assessed using meta-analysis is shown in Appendix 3. Forest plots of meta-analysis for HLA Class I, HLA Class II, and Interleukin can be found in figure 3.10 and 3.11.

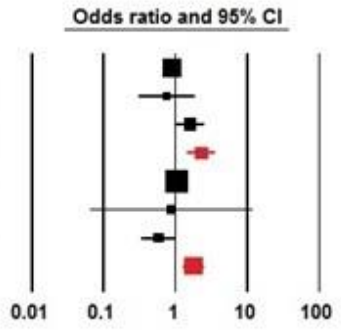
HLA Class I – A

	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
A*01	0.855	0.647	1.130	-1.101	0.271
A*02	1.145	0.896	1.464	1.081	0.280
A*03	1.577	1.010	2.462	2.004	0.045
A*11	1.182	0.726	1.925	0.672	0.502
A*23	0.606	0.402	0.914	-2.390	0.017
A*24	1.130	0.769	1.660	0.623	0.534
A*25	0.310	0.064	1.504	-1.453	0.146
A*26	1.351	0.479	3.810	0.569	0.570
A*29	0.671	0.355	1.268	-1.229	0.219
A*30	1.223	0.332	4.503	0.303	0.762
A*31	0.968	0.344	2.724	-0.062	0.951
A*32	1.581	0.778	3.214	1.265	0.206
A*33	0.375	0.038	3.681	-0.842	0.400
A*68	1.100	0.617	1.961	0.323	0.747
A*74	0.375	0.038	3.681	-0.842	0.400



HLA Class I – B

	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
B*07	0.889	0.647	1.221	-0.727	0.467
B*08	0.762	0.308	1.885	-0.588	0.556
B*15	1.590	0.993	2.547	1.929	0.054
B*27	2.291	1.446	3.629	3.532	0.000
B*35	1.038	0.803	1.342	0.285	0.776
B*38	0.858	0.063	11.648	-0.115	0.908
B*5301	0.577	0.326	1.022	-1.887	0.059
B*57	1.781	1.286	2.466	3.475	0.001



HLA Class I – C

	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
C*01	0.661	0.363	1.203	-1.355	0.175
C*02	0.951	0.676	1.338	-0.288	0.773
C*03	0.690	0.548	0.869	-3.147	0.002
C*04	1.468	1.002	2.151	1.969	0.049
C*05	1.460	1.043	2.044	2.203	0.028
C*06	1.030	0.768	1.381	0.198	0.843
C*07	0.952	0.745	1.217	-0.392	0.695
C*08	0.917	0.565	1.489	-0.351	0.726
C*12	1.318	0.791	2.196	1.060	0.289
C*16	0.918	0.628	1.341	-0.442	0.658

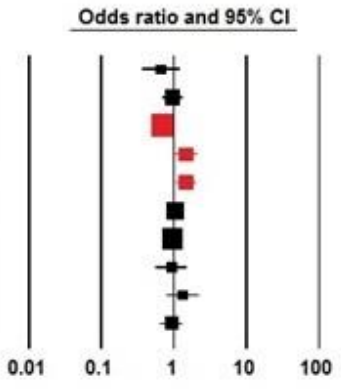
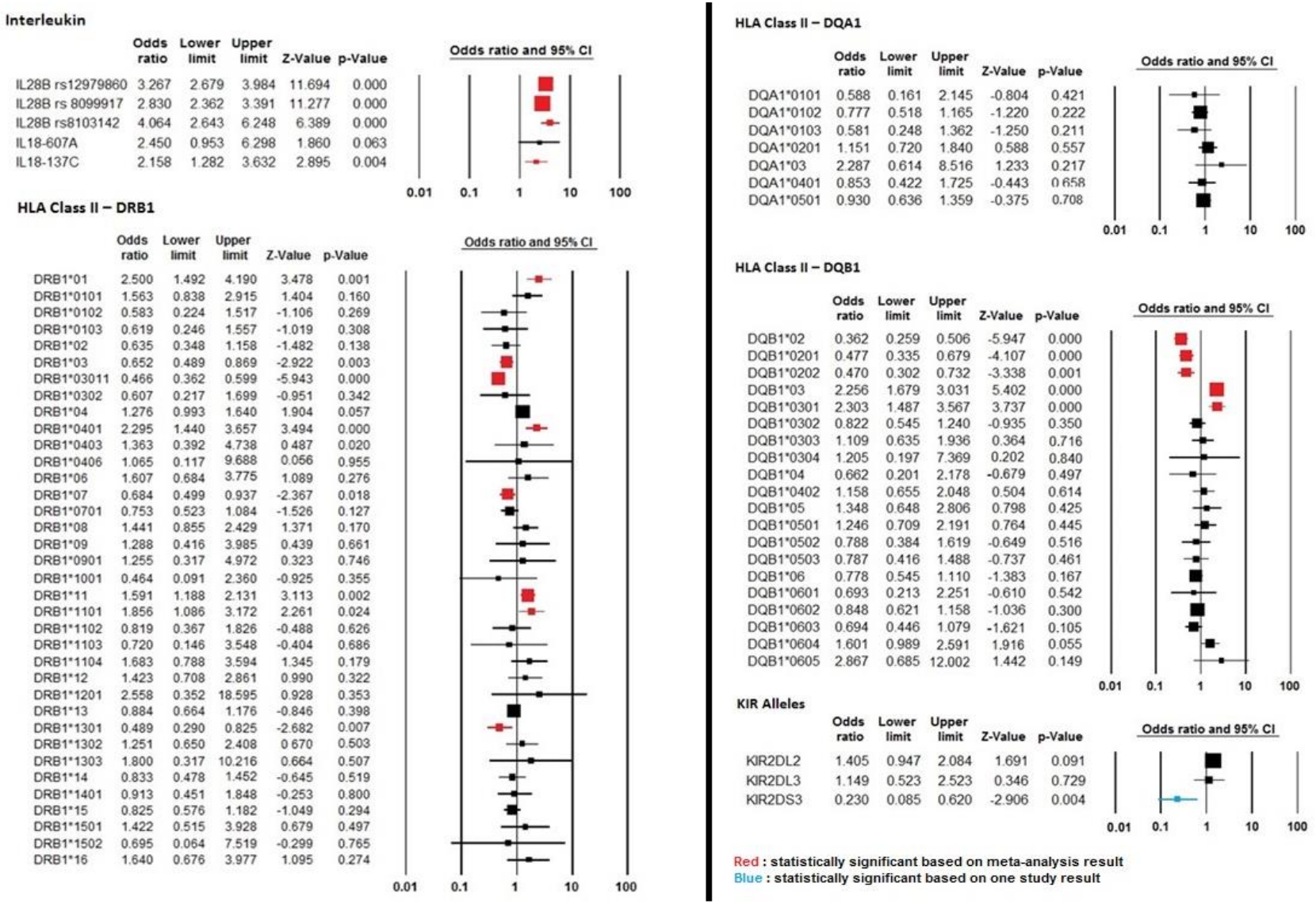


Figure 2.16 Forest Plot Assessing HLA Class I Associated with HCV Spontaneous Clearance



Interleukin	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
IL28B rs12979860	3.267	2.679	3.984	11.694	0.000
IL28B rs 8099917	2.830	2.362	3.391	11.277	0.000
IL28B rs8103142	4.064	2.643	6.248	6.389	0.000
IL18-607A	2.450	0.953	6.298	1.860	0.063
IL18-137C	2.158	1.282	3.632	2.895	0.004

HLA Class II – DRB1	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
DRB1*01	2.500	1.492	4.190	3.478	0.001
DRB1*0101	1.563	0.838	2.915	1.404	0.160
DRB1*0102	0.583	0.224	1.517	-1.106	0.269
DRB1*0103	0.619	0.246	1.557	-1.019	0.308
DRB1*02	0.635	0.348	1.158	-1.482	0.138
DRB1*03	0.652	0.489	0.869	-2.922	0.003
DRB1*03011	0.466	0.362	0.599	-5.943	0.000
DRB1*0302	0.607	0.217	1.699	-0.951	0.342
DRB1*04	1.276	0.993	1.640	1.904	0.057
DRB1*0401	2.295	1.440	3.657	3.494	0.000
DRB1*0403	1.363	0.392	4.738	0.487	0.020
DRB1*0406	1.065	0.117	9.688	0.056	0.955
DRB1*06	1.607	0.684	3.775	1.089	0.276
DRB1*07	0.684	0.499	0.937	-2.367	0.018
DRB1*0701	0.753	0.523	1.084	-1.526	0.127
DRB1*08	1.441	0.855	2.429	1.371	0.170
DRB1*09	1.288	0.416	3.985	0.439	0.661
DRB1*0901	1.255	0.317	4.972	0.323	0.746
DRB1*1001	0.464	0.091	2.360	-0.925	0.355
DRB1*11	1.591	1.188	2.131	3.113	0.002
DRB1*1101	1.856	1.086	3.172	2.261	0.024
DRB1*1102	0.819	0.367	1.826	-0.488	0.626
DRB1*1103	0.720	0.146	3.548	-0.404	0.686
DRB1*1104	1.683	0.788	3.594	1.345	0.179
DRB1*12	1.423	0.708	2.861	0.990	0.322
DRB1*1201	2.558	0.352	18.595	0.928	0.353
DRB1*13	0.884	0.664	1.176	-0.846	0.398
DRB1*1301	0.489	0.290	0.825	-2.682	0.007
DRB1*1302	1.251	0.650	2.408	0.670	0.503
DRB1*1303	1.800	0.317	10.216	0.664	0.507
DRB1*14	0.833	0.478	1.452	-0.645	0.519
DRB1*1401	0.913	0.451	1.848	-0.253	0.800
DRB1*15	0.825	0.576	1.182	-1.049	0.294
DRB1*1501	1.422	0.515	3.928	0.679	0.497
DRB1*1502	0.695	0.064	7.519	-0.299	0.765
DRB1*16	1.640	0.676	3.977	1.095	0.274

HLA Class II – DQA1	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
DQA1*0101	0.588	0.161	2.145	-0.804	0.421
DQA1*0102	0.777	0.518	1.165	-1.220	0.222
DQA1*0103	0.581	0.248	1.362	-1.250	0.211
DQA1*0201	1.151	0.720	1.840	0.588	0.557
DQA1*03	2.287	0.614	8.516	1.233	0.217
DQA1*0401	0.853	0.422	1.725	-0.443	0.658
DQA1*0501	0.930	0.636	1.359	-0.375	0.708

HLA Class II – DQB1	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
DQB1*02	0.362	0.259	0.506	-5.947	0.000
DQB1*0201	0.477	0.335	0.679	-4.107	0.000
DQB1*0202	0.470	0.302	0.732	-3.338	0.001
DQB1*03	2.256	1.679	3.031	5.402	0.000
DQB1*0301	2.303	1.487	3.567	3.737	0.000
DQB1*0302	0.822	0.545	1.240	-0.935	0.350
DQB1*0303	1.109	0.635	1.936	0.364	0.716
DQB1*0304	1.205	0.197	7.369	0.202	0.840
DQB1*04	0.662	0.201	2.178	-0.679	0.497
DQB1*0402	1.158	0.655	2.048	0.504	0.614
DQB1*05	1.348	0.648	2.806	0.798	0.425
DQB1*0501	1.246	0.709	2.191	0.764	0.445
DQB1*0502	0.788	0.384	1.619	-0.649	0.516
DQB1*0503	0.787	0.416	1.488	-0.737	0.461
DQB1*06	0.778	0.545	1.110	-1.383	0.167
DQB1*0601	0.693	0.213	2.251	-0.610	0.542
DQB1*0602	0.848	0.621	1.158	-1.036	0.300
DQB1*0603	0.694	0.446	1.079	-1.621	0.105
DQB1*0604	1.601	0.989	2.591	1.916	0.055
DQB1*0605	2.867	0.685	12.002	1.442	0.149

KIR Alleles	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value
KIR2DL2	1.405	0.947	2.084	1.691	0.091
KIR2DL3	1.149	0.523	2.523	0.346	0.729
KIR2DS3	0.230	0.085	0.620	-2.906	0.004

Figure 2.17 Forest Plot Assessing Interleukin, KIR Alleles and HLA Class II Associated with HCV Spontaneous Clearance

I also summarised alleles' frequencies for 24 host genetic predictors among 4 different groups: HCV patients with spontaneous clearance, HCV patients with persistent infection, healthy population, and patients with HCV infection (see Table 3.3). From table 3.3 below, five host genetic factors with highest proportion of alleles frequencies among patients with spontaneous clearance were IL28B rs8099917 (87.80%, 95% CI: 78.8%-93.3%), followed by IL28B rs8103142 (72.70%, 95% CI: 64.3%-79.8%), IL28B rs12979860 (65.10%, 95% CI: 60.2%-69.7%), DQB1*03 (57.40%, 95% CI: 35.9%-76.4%), and HLA-Cw*07 (42.40%, 95% CI: 24.6%-62.1%). On the other hand, five host genetic factors with lowest proportion of alleles frequencies among spontaneous clearance patients were HLA-A*23 (4.0%, 95% CI: 2.8%-5.6%), HLA-B*57 (8.1%, 95% CI: 6.1%-10.8%), DRB1*1301 (9.4%, 95% CI: 4.4%-18.7%), HLA-B*27 (11.4%, 95% CI: 7.4%-17.2%), and DQB1*0202 (12.1%, 95% CI: 4.0%-31.0%). The forest plot of each alleles' frequency predictor can be found in Appendix 4.

Table 2.5 Proportion of Alleles/Genes among Patients with Spontaneous Clearance, Persistence Infection, Healthy Control and HCV Patients

No	Variables	Spontaneous Clearance				Persistent Infection				Healthy Control				HCV Infection			
		Σ of studies	I^2	%	95% CI	Σ of studies	I^2	%	95% CI	Σ of studies	I^2	%	95% CI	Σ of studies	I^2	%	95% CI
1	IL28B rs12979860	26	79.30	65.10	60.2-69.7	21	96.61	37.30	30.7-44.5	7	97.36	57.90	38.9-74.8	8	88.15	41.90	36.5-47.4
2	IL28B rs8099917	11	90.22	87.80	78.8-93.3	10	96.26	66.90	57.4-75.2	3	89.61	84.30	67.2-93.4	3	29.02	84.20	81.6-86.4
3	IL28B rs8103142	2	0.00	72.70	64.3-79.8	2	0.00	39.60	35.8-43.5								
4	IL18-317C	2	70.72	18.00	5.5-45.2	2	0.00	18.90	15.5-22.8	1	-	12.00	6.9-20	1	-	14.00	8.5-22.3
5	HLA-A*03	3	79.86	26.50	15.0-42.5	3	94.00	24.60	10.7-47.0					1	-	49.10	35.6-48.4
6	HLA-A*23	4	0.00	4.00	2.8-5.6	4	67.65	6.70	4.2-10.5					1	-	5.70	3.4-9.6
7	HLA-B*27	3	21.80	11.40	7.4-17.2	3	39.92	3.80	2.3-6.4	1	-	5.00	2.7-9.1	2	58.24	4.90	2.4-9.7
8	HLA-B*57	2	43.09	8.10	6.1-10.8	2	35.80	5.00	3.8-6.5					1	-	8.80	5.8-13.3
9	HLA-Cw*04	5	86.67	13.80	7.3-24.7	5	93.82	22.50	13.2-35.6	1	-	33.70	27.4-40.5	2	97.37	31.50	9.3-67.2
10	HLA-Cw*05	4	86.78	15.30	5.5-35.7	4	87.51	8.30	3.1-20.2	1	-	4.50	2.4-8.5	2	95.38	10.30	1.0-56.2

No	Variables	Spontaneous Clearance				Persistent Infection				Healthy Control				HCV Infection			
		Σ of studies	I^2	%	95% CI	Σ of studies	I^2	%	95% CI	Σ of studies	I^2	%	95% CI	Σ of studies	I^2	%	95% CI
11	HLA-Cw*07	5	90.51	42.20	24.6-62.1	5	93.96	41.80	26.2-59.1	1	-	21.10	16.0-27.3	2	98.55	36.20	5.3-85.2
12	DRB1*01	9	77.70	18.00	11.6-26.8	9	68.43	12.20	8.8-16.5	2	87.48	19.20	15.1-24.2	1	-	18.10	13.8-23.5
13	DRB1*03	9	60.53	18.80	13.7-25.2	9	84.49	26.10	19.4-34.1	2	95.59	27.10	19.0-36.9	1	-	31.70	26.1-37.8
14	DRB1*03011	7	81.37	12.60	7.6-20.2	7	95.95	20.30	11.2-33.8	3	97.64	18.40	7.0-40.2	3	0.00	30.90	27.2-34.8
15	DRB1*0401	2	0.00	29.30	21.9-37.9	2	0.00	15.20	11.3-20.2	2	87.09	28.00	23.1-33.5	1	-	30.50	25.0-36.5
16	DRB1*07	8	56.12	18.20	13.0-24.8	8	82.32	24.70	18.1-32.8	1	-	33.40	30.2-36.7				
17	DRB1*11	11	78.79	20.20	13.6-28.9	11	66.32	13.90	10.7-17.9	2	0.00	24.90	22.2-27.7	1	-	13.50	8.1-21.5
18	DRB1*1101	6	89.17	18.70	10.0-32.3	6	89.68	11.90	7.4-18.7	2	76.61	17.40	13.2-22.5				
19	DRB1*1301	5	71.02	9.40	4.4-18.7	5	88.81	9.80	5.0-18.4	3	94.21	9.80	5.9-15.8	1	-	19.30	14.9-24.8
20	DQB1*02	6	71.10	22.60	14.7-33.2	6	92.34	37.40	24.0-53.1	1	-	50.10	46.7-53.6				
21	DQB1*0201	9	83.45	19.10	13.1-27.1	9	93.70	30.50	22.1-40.3	2	98.01	27.20	7.2-64.2	2	85.24	39.00	25.7-54.3
22	DQB1*0202	4	91.08	12.10	4.0-31.0	4	93.85	19.10	8.2-38.4								
23	DQB1*03	7	91.23	57.40	35.9-76.4	7	89.70	35.70	25.2-47.7	1	-	51.40	47.9-54.8				
24	DQB1*0301	11	88.85	42.50	32.2-53.5	11	71.90	25.50	21.8-29.6	3	79.32	36.80	30.6-43.5	1	-	34.60	26.1-44.2

Blue typing : data only available in one study

To identify genetic factors that were both common and have an important impact on clearance rates I tabulated the odds ratio for SVC against allele frequency in those who spontaneously cleared in figure 3.12. This highlights the importance of IL28B rs8103142, IL28B rs12979860, and IL28B rs8099917.

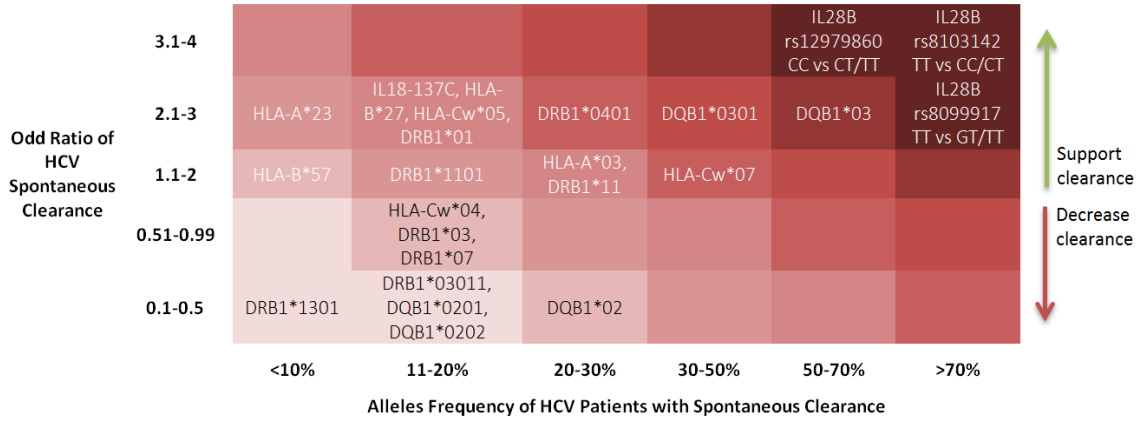


Figure 2.18 Odd Ratio of HCV Spontaneous Clearance in relation to Allele Frequency

3.4 Discussion

3.4.1 Summary of Findings

In this systematic review and meta-analysis of hepatitis C spontaneous clearance, I included data from 86 studies, representing 38,605 individuals. Notably, polymorphism in IL28B rs8103142 TT genotype, IL28B rs12979860 CC genotype, and IL28B rs8099917 TT genotype was found as the most important host genetic predictors for clearance, but a wide range of other genetic predictors with lower frequency also affected clearance.

3.4.2 Study Strength and Limitation

To the best of my knowledge, this is the first meta-analysis examining host genetic determinants of HCV clearance in one systematic review. It is also the first meta-analysis performed to investigate the association between HLA Class I and HCV spontaneous clearance. The strengths of my study are that I conducted an extensive systematic literature search and used robust methods to estimate the relationship between host genetic determinants and spontaneous HCV clearance. I included all host genetics determinants found during the systematic review and performed meta-analysis where possible. Unlike previous work which often focuses solely on odds ratios or relative risks, I also estimated the allele's frequency of

potential host genetics related to clearance using meta-analysis. This is important to help to understand population impact.

This study has a number of limitations due to the small number of studies available for some genetic determinants, different study designs, limited information on sample populations and different study populations. As can be seen from Figure 3.2, there are many countries which are not represented at all in this analysis. This will bias my results due to the strong association between genotype, ancestry and SVC. For example, IL28B rs12979860 CC genotype was found strongly associated with SVC among individuals of both European and African ancestry (44), and IL28B rs12979860 was found most associated with SVR in white patients, whereas in East Asians it seemed to be IL28B rs8099917 (91). Unfortunately in this study, I could not perform sub-analysis based on ethnic groups due to insufficient data, and the results might be different for each race. A meta-regression could not be conducted due to limited individual data.

Due to the small number of publications for some specific host genetic determinants, some meta-analysis was done by calculating the odds ratio from only two studies. Results from such analyses need to be interpreted with extreme caution given the small sample size and associated broad confidence intervals. Studies presented in this chapter are subject to the same issues with selection bias as outlined in Chapter 2. Individuals with asymptomatic infection and those who achieve SVC are less likely to present to hospital and be included in research studies. Whilst the 6 studies included in the estimates of SVC in chapter 2 were comparatively homogeneous in terms of their study population (mainly patients tested for HCV in hospital), studies of genetic determinants included in this chapter represent a much broader range of study designs and populations (HIV positive patients, transfusion dependent, PWID). This makes it challenging to address the wide range of potential confounding variables in these studies, most of which are not reported in individual studies. In case control studies the choice of control population can have a major impact on the estimate of effect that is being measured. Some studies included comparison with healthy controls whereas other studies compared HCV RNA-positive cases to HCV-RNA negative individuals as a control group

Only 35 out of 86 studies had more than 100 patients with HCV clearance representing relatively small sample sizes that might affect the power of the study, particularly in the context of multiple testing for a large number of genetic variants.

This emphasises the need for meta-analytic approaches. Different settings, as well as different populations recruited in the studies might also affect the results and explained heterogeneity in the results. The impact of confounding is likely to be less important for this analysis than in the analysis of demographic and behavioural risk factors as demographic and behavioural traits are unlikely to be strongly associated with genetic traits that influence spontaneous viral clearance. Ethnicity and genetic polymorphisms may however be strongly associated as discussed above.

3.4.3 DISCUSSION OF THE RESULTS

This study confirms that the strongest host genetic predictors of clearance were IL28B rs8103142 TT genotype, IL28B rs12979860 CC genotype, and IL28B rs8099917 TT genotype (39, 89, 91). It also confirmed a further 21 host genetics determinants associated with HCV spontaneous clearance (80, 84, 92). Polymorphisms in IL28B (IL28B rs12979860, IL28B rs8099917, IL28B rs8103142) can help to predict HCV clearance as well as treatment outcome. (93, 94)

Accumulating evidence shows that allele frequency of IL28B polymorphisms is significantly influenced by ethnicity (44, 91, 95), through studies undertaken in Chinese, Caucasians, Africans and Asian populations.(96) IL28B encodes endogenous antiviral cytokine interferon-λ3 (IFN-λ3) which constitutes the type III IFN-λ family together with IL-29 (IFN-λ1) and IL-28A (IFN-λ2). These genes are located on chromosome 19 (97, 98) and belong to the interleukin-10 (IL-10) superfamily (99). It is suggested that both IFN-α and IFN-λ are involved in host innate immune response to HCV infection, via signaling through the JAK-STAT (Janus kinase-signal transducer and activator of transcription) pathway.(100, 101) The JAK-STAT pathway induces a large number of interferon-stimulated genes (ISGs) which instigate antiviral cellular activity.(102, 103) Patients with favourable IL28B rs12979860 genotype CC were also found have better response to HCV treatment.(93, 94)

Our study also assessed the association between HLA class I alleles (including HLA -A, -B and -C antigens) and HCV spontaneous clearance, where no previous meta-analysis was found assessing this association before. The analysis showed that HLA-A*03, -B*27, -B*57, -Cw*05, -Cw*07, are associated with clearance whereas HLA-A*23, -Cw*04 and –Cw*03 are not associated. It is believed that HLA class I play an important role in adaptive cellular immune response.(104) HLA allele variations influence the course of HCV infection, probably by shaping the repertoire

of viral epitopes which can bind to relevant HLA molecules thus generating immune responses.(104, 105)

The most widely reported associations of HLA with HCV spontaneous clearance are with HLA class II alleles. (5, 90, 106) HLA genes are located on chromosome 6 and encode peptides involved in host immune response. In this study, we found that DRB1*01, DRB1*0401, DRB1*11, DRB1*1101, DQB1*03, and DQB1*0301 have strong association with HCV clearance whereas DRB1*03, DRB1*03011, DRB1*07, DRB1*1301, DQB1*02, DQB1*0201, and DQB1*0202 have the opposite effect. This result is supported by another meta-analysis conducted by Hong et al. which investigated the association between DQB1*0301 and DRB1*11 with HCV spontaneous clearance.(106) HLA class II polymorphisms may increase amino acid substitutions with different peptide-binding characteristics and determine antigenic specificities, thus influencing the immune response strength to HCV.(107) While there are consistent observations of some host genetic determinants for HCV spontaneous clearance, many studies produced conflicting results for others predictors. These inconsistencies may be due to ethnic differences, sample size, patient selection, and HCV-genotype/serotype. Many of the genetic factors are considered in isolation with no studies assessing all candidate markers to measure the combined influence of various genetic markers on risk of clearance. Large and well-designed studies and/or meta-regression approaches are required to investigate whether more accurate prediction of clearance can be achieved based on measurement of multiple markers rather than each marker in isolation. Predictive models may be further refined by also including behavioural and demographic factors.

3.4.4 Implication for Policy and Practices

In common with previous studies (39, 89, 91), I found several host genetic predictors of HCV spontaneous clearance. Polymorphism in the interleukin 28 (IL28B) gene regions, specifically from IL28B rs12979860 CC genotype, IL28B rs8099917 TT genotype and IL28B rs8103142 TT genotype were the strongest candidates based on both the size of the effect and their prevalence. Not only are these genotypes strongly associated with SVC, they are also highly prevalent in individuals who achieve clearance. This suggests that *IL28B* typing might be useful for treatment decision-making in acute HCV infection.(108) These results suggest that patients with unfavourable IL28B genotypes could be prioritised for having early treatment, given their low likelihood of spontaneous clearance.

Furthermore, patients with favourable IL28B rs12979860 genotype CC were found to have better response to PEGylated interferon and ribavirin treatment.(93, 94) Thus, in the DAA treatment era, *IL28B* typing might help to inform the decision about selection of therapy in patients with chronic genotype 1 HCV infection, particularly in the context of the limited availability of DAA therapy in many places. (109) However, with decreasing costs of treatment such prioritisation may not be needed. In resource poor settings it would be necessary to balance the costs of genetic testing with the potential savings from delaying treatment to assess whether such a strategy is cost saving or not. In patients with the good response *IL28B* genotype, peg-IFN and RBV therapy could still be considered as an alternative to DAAs because of the lower cost and with high rates of sustained virological response (SVR). However, the side effect profile is less favourable than direct acting agents.

In the future, the *IL28B* genotype might have a role in individualizing treatment regimens for patients with HCV depending on how DAA therapy evolves and whether drug resistance emerges as a genuine concern.(109) As more potent DAAs progress through the clinical drug development pathway, it might be anticipated that the contribution of host factors, such as the *IL28B* genotype, to treatment response will diminish.

From a population perspective, it is interesting to compare the effect size of genetic determinants of SVC versus clinical and behavioural determinants. Although IL28B is very strongly associated with clearance and has a potential role in terms of individualised treatment decisions, it seems unlikely that a genotypic test will be widely used in the short term, particularly in low-income countries. By contrast, although the effect sizes seen with epidemiological predictors in chapter 2 show a more modest association with SVC, all the predictors point to high-risk groups as the priority for HCV treatment – a finding that is highly relevant for treatment policy.

3.4.5 My Role in This Study

This study is part of an extensive systematic review presented in chapter 2. I developed the study protocol, conducted the literature search, and downloaded/stored all of the papers screened through the systematic search. I performed the initial screening (by reviewing papers' title and abstracts) and performed full-text review with another independent reviewer. I also conducted the data extraction and undertook the meta-analysis. I wrote the first draft of the manuscript which has been submitted for publication to BMC Infectious Disease.

Key Points:

- A total of 86 studies were included in the main analysis, representing 38,605 individuals
- Several genetic factors were identified which associate strongly with SVC including IL28B rs12979860 CC genotype (OR=3·27, 95% CI: 2·68-3·98), IL28B rs8099917 TT genotype (OR=2·83, 95% CI: 2·36-3·39), IL28B rs8103142 TT genotype (OR=4·06, 95% CI: 2·64-6·25), HLA-B*27 (OR=2·29, 95% CI: 1·45-3·63), HLA-Cw*07 (OR=2·00, 95% CI: 1·36-2·92), DRB1*01 (OR=2·50, 95% CI: 1·49-4·19), DRB1*03011 (OR=0·47, 95% CI: 0·36-0·60), DQB1*02 (OR=0·36, 95% CI: 0·26-0·51), DQB1*0301 (OR=2·30, 95% CI: 1·49-3·57), etc.
- After tabulating the odds ratio for SVC against allele frequency in those who spontaneously cleared, this study confirmed the importance of IL28B rs8103142 TT genotype, IL28B rs12979860 CC genotype, and IL28B rs8099917 TT genotype.
- These results suggest that patients with unfavourable IL28B genotypes could be prioritised for having early treatment, given their low likelihood of spontaneous clearance. In addition, *IL28B* typing might help to inform the decision to start therapy in patients with chronic genotype 1 HCV infection, particularly in the context of the impending availability of direct-acting antiviral (DAA) therapy.
- However, the benefits of genotyping would need to be considered in the context of the falling costs of DAA's treatment.

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4. Background of Hepatitis C Study in Guernsey

Chapter Summary:

In this chapter, I describe the protocol for a study which I planned to investigate the feasibility of hepatitis C elimination on the Island of Guernsey. The study planned to which intended to investigate the burden of hepatitis C in Guernsey and gain insight into the relative importance of local transmission versus importation of infection. The aim of this study was to describe the epidemiology of hepatitis C virus (HCV) infection and transmission in a closed population of high-risk individuals by combining clinical data with whole-genome sequencing. I designed the study and obtained ethical approvals and we were scheduled to start recruiting patients in January 2016. Unfortunately, the enthusiasm of our Guernsey collaborators waned and we were unable to recruit patients to the study. Therefore, in this chapter, I outline the methods that were proposed and my role in designing the study.

4.1 Introduction

As previously described in chapter 1, hepatitis C virus (HCV) infection is an important public health problem, and the development of new Direct Acting Antiviral (DAA's) creates the potential to eliminate HCV in the future. However, the major barrier to eliminating HCV through treatment is operational relating to the cost of treatment, limited access to treatment, availability of trained medical staff, identification of undiagnosed cases and treatment adherence. Before attempting to eliminate HCV in a large population, it makes sense to investigate the feasibility of eradicating HCV in a small, comparatively closed population. In Guernsey, critical questions to assess the feasibility of elimination are the prevalence of infection in different risk groups and the relative importance of local transmission and importation of infection.

Through clinical links between UCL and Guernsey healthcare service, the opportunity arose to undertake an epidemiological analysis of HCV on the island linked to whole genome sequencing of HCV isolates (in collaboration with an existing UCL-led study). Guernsey, as part of the Channel Island, provides an opportunity to assess the feasibility of eliminating or reducing HCV transmission through treatment in the context of a closed population. Guernsey has a population of 62,229 with small annual changes in population size ranging from a reduction of

0.5% reduction to 0.28% increase between 2013 and 2016.(1) Guernsey Health and Social Care is responsible for Guernsey's adult community services, hospital services, public health services, and children and family community services.(2) With its relatively closed population, well-established health care services, close inter-agency collaboration, well-characterised high-risk population, and funding opportunities to treat infected high-risk individuals using DAA's, Guernsey provided a unique opportunity to investigate the feasibility of HCV elimination.

In collaboration with a research team in Guernsey, I planned to recruit individuals through high-risk settings, such as drug treatment centres, drug and alcohol treatment services, prisons, and sexual health clinics to undertake a prevalence survey in order to understand the true burden of disease in these high-risk populations. This work would inform estimates of the number of undiagnosed HCV cases in Guernsey to support interventions to reduce HCV transmission in the community, in particular to understand the extent to which screening needed to be targeted and intensified.

We planned to recruit participants of known and unknown HCV status in 4 different settings (in the Orchard Clinic, Drug Concerns, Community Drug & Alcohol Team, and Guernsey prison). At each site, for individuals who were HCV negative, or for whom status was unknown, medical staff would perform the HCV oral screening test. If an individual screened positive on this test, a blood sample would be taken for confirmatory testing. For individuals with a positive blood test, residual viral material from the blood test would be submitted for whole genome sequencing by linking to Infection Response through Virus Genomics (ICONIC) study, a programme grant led by UCL which is funded by the Department of Health and the Wellcome Trust. Infection response through virus genomics (ICONIC) is a study assessing the viral pathogen using Whole Genome Sequencing (WGS) to support patient stratification and viral outbreak surveillance using five exemplar viruses: HCV, HIV, measles, norovirus and influenza. Clinical and epidemiological data for each case would be collected by the research nurse at each site and stored locally prior to pseudo-anonymization and transfer to UCL.

By analysing HCV sampled viruses collected during hepatitis C study in Guernsey linked to clinical and epidemiological data I planned to use whole genome sequencing (WGS) to investigate the HCV genotype distribution and

transmission in this specific population by linking to ICONIC study. Thus, these results would have helped us to understand the epidemiology of HCV in the Bailiwick of Guernsey and enabled the public health board to develop appropriate treatment and prevention plans for HCV on the island. In particular, if the sequencing indicating a high importance of regular introduction of new strains from outside Guernsey, then this would be anticipated to make elimination harder due to repeated “re-seeding” of the population.

4.2 Aim and Objectives

4.2.1 Aim

To estimate the prevalence of hepatitis C among high-risk individuals who access clinical services in Guernsey and use full-length viral genome sequencing to investigate HCV transmission in Bailiwick of Guernsey.

4.2.2 Objectives

- To assess the prevalence of HCV in high-risk individuals in Guernsey using a rapid salivary test for HCV
- To collect blood specimens from known and newly diagnosed patients with hepatitis C and to submit these samples for whole genome sequencing
- To collect demographic, clinical and epidemiological data from individuals who are at high risk of HCV and those with a confirmed HCV infection in order to optimally target screening.
- To perform whole genome sequencing on stored HCV positive samples and repeat samples from the same individual (where available) to investigate HCV transmission.
- To investigate the HCV genotype distribution across Guernsey, the initial time of HCV infection, and transmission networks among the infected population
- To distinguish between local transmission and infection acquired outside of Guernsey

4.3 Methods

4.3.1 Study Area

The Bailiwick of Guernsey is part of the Channel Islands with an area over 78 square km and a total population of 62,229 (July 2017). The median average age was 41.90 for males and 44.2 for females. The population is relatively stable with very small annual changes with the total population increased by 0.06% over the year ending 31st December 2016.(1) Guernsey attracts many visitors. The total number of visitors between January and September 2017 was 246,250, of which

14% travelled for business purposes, 17% were visiting friends or relatives, 54% were leisure visitors staying for at least one night and a further 15% visiting for a day trip.(3)

At the time of writing, a total of 108 HCV infected patients are currently registered at Guernsey Hepatitis Service, a service provided by the Orchard Clinic to identify and follow up hepatitis C patients across Guernsey. The clinic is part of Guernsey Sexual Health Service and led by Dr. Nicola Brink who is a consultant virologist and assistant director in medical public health, Guernsey Health and Social Services Department**.** Dr. Nicola was local PI for this study. I led on the development of the protocol with the support of Dr. Nicola Brink and my PhD supervisors. Presently there is limited island wide prevalence data on hepatitis C, but the number of hepatitis C cases suggests Guernsey is likely to have a similar HCV prevalence to the UK. An audit published in 2010 suggested that 84% of the Orchard Clinic service users acquired their infection through injecting drug use.(4) The examination of blood samples showed that more than 90% of those infected were infected with genotype 3a. This is in contrast to drug users on the mainland where genotype 1a is the most common genotype in injecting drug users. This is likely to reflect the importance of transmission within this relatively closed community.

The aim of this study was to recruit participants from four sites serving populations at high risk of HCV, namely the Orchard Clinic, prison, Drug Concern and CDAT (Community Drug and Alcohol Team).

a) The Orchard Clinic

The Orchard Clinic is an open access clinic for people who are concerned about their sexual health to attend and have an appointment without being referred by the doctor. Having professional and trained staff, the clinic offers services and consultation. The clinic opens 4 days a week, providing services on: Information & advice on Sexually Transmitted Infections; Screening for Sexually Transmitted Infections (STI's); HIV testing; Hepatitis A, B and C Testing; Cervical smear tests; Emergency contraception; Free condoms & lubricants; Pregnancy testing; and Management & Treatment of sexually transmitted infections and blood borne viruses. The Guernsey Viral Hepatitis Service is also located in the Orchard Clinic, and staff who are working in this service are responsible for the care of people living with hepatitis B and C infections locally. Staffs at the Orchard Clinic have successfully treated 41 cases to date. However, there are currently, about

40 patients infected with HCV registered in the clinic waiting for treatment with an interferon-free treatment regimen.

b) Drug Concern

Drug Concern is a local charity involved in addressing the needs of people whose lives are affected by substance misuse within the Bailiwick of Guernsey. This is achieved both in preventative ways, such as education or training, and by means of a variety of harm-reduction measures and treatments, including targeted psychosocial interventions, information, advice and support, and the provision of a needle exchange program.

Based on the 2013 Drug Concern Report, a total of 240 people have received structured work for substance misuse problems in Guernsey, of which 60% come from prison. Heroin and pharmaceutical opiates such as fentanyl and suboxone remain the primary abused substances, while alcohol is reported as the primary substance for which prisoners are seeking support (5).

c) Community Drug & Alcohol Team (CDAT)

CDAT provides confidential drug and alcohol services for residents of Guernsey. The drug service is primarily for people who are opiate dependent and the alcohol service is for chronic or physically dependent drinkers. Located in Castel Hospital, the CDAT team consists of a manager, a clinical nurse specialist, two senior staff nurses, a specialist social worker, a consultant psychiatrist, an associate specialist and a team secretary. Referrals are received from GPs and other professionals. CDAT drug and alcohol provides some services including:

- Community alcohol/opiate detox
- A supervised opiate substitution programme
- Group therapy
- Auricular Acupuncture.

d) Guernsey Prison

Guernsey Prison plays a vital role in the diagnosis and management of HCV-infected people in the Bailiwick. Working closely with Drug Concern, Guernsey Prison has established 4 years partnership to conduct Prison's Substances Misuse Service. The team concentrates on individual cases within the prison, specifically prisoners' needs whilst in custody and ensuring these needs are adequately met.

Throughout 2013, Drug Concern assessed 202 prisoners and provided structured interventions for 145 of these. Drug Concern has provided services to the prison, including group work for prisoners. The program targets offenders

who have reported problems related to drugs or alcohol, and aims to increase the participants' awareness of how substance use impacts on key areas of their lives, by using a variety of methods (6). Guernsey Prison Service has also provided active support and help for HCV-infected prisoners undergoing treatment.

4.3.2 Study Design and Timescale

Cross-sectional study with nested cohort of individuals who test positive for HCV on salivary test on the Island State of Guernsey. In this study, the participants were divided into:

a) High-risk individuals with unknown hepatitis C status who access services in Guernsey
b) People who have been previously diagnosed with HCV based on data from Guernsey Viral Hepatitis Service

This study planned to recruit participants between January and December 2016.

4.3.3 Sample Size Calculation

Previous studies have demonstrated a prevalence of HCV in the UK of approximately 0.67 (95% CI: 0.5-0.94).(7) With a population of 62,000 people in Guernsey, there will be an estimated 415 individuals with HCV. In the UK, the prevalence of HCV infection among PWIDs is about 50% (95% CI: 47-54%).(8) The sample numbers were primarily based on the number of high risk patients attending the settings. However, assuming we were able to screen 300 high risk individuals; we would be able to estimate a 50% prevalence with a 95% confidence interval from 44-56%.

4.3.4 Methodology

A. Group 1 – The Unknown Hepatitis C Status Participants (see diagram)

We aimed to recruit all high-risk individuals in Guernsey who accessed services in the 4 study sites. For all high-risk individuals, the member of staff would:

a) Obtain informed consent
a) Administer research questionnaire to participants
b) Take oral specimens for HCV testing
c) When the saliva test result is positive, obtain a blood sample for confirmatory testing
d) When the confirmatory blood test is positive, perform a clinical assessment

Where suitably qualified medical staffs were available at the study sites, the blood sample would be taken following a positive salivary test. If phlebotomy staff were unavailable, the participants were to have been referred to the Orchard Clinic to have the blood sample collection.

B. Group 2 – The Known Hepatitis C Participants (see diagram)

Some people in Guernsey have been previously diagnosed with Hepatitis C at the Orchard Clinic. This group of individuals was divided into three sub-groups:

a) Those who still routinely accessed the services in one of the study sites
b) Those who were not currently in contact with services at any of the 4 study sites
c) Those who have been diagnosed with hepatitis C as part of sexual health screening at the Orchard Clinic

For subgroup a) consisting of patients who have been previously diagnosed with HCV and are still in contact with services, the following procedures were to have been applied:

a) Obtain informed consent which included the use of the stored positive blood samples (if available)
b) Administer the research questionnaire
c) Collect blood samples if required
d) Perform clinical assessment for markers of liver disease

The known hepatitis C participants who visited CDAT and Drug Concern were to have been referred to the Orchard Clinic if phlebotomists were unavailable to do the blood sample collection. The clinical assessment would also have been performed at the Orchard Clinic conducted by a suitably qualified clinician. For participants in prison, if the medical staff were unavailable, the Orchard clinic staff would have visited the prison to collect a blood sample and perform the clinical assessment.

For the sub-group b) who were not in current contact with services, the procedure would be:

a) Invitation to participate in the research study sent by email, phone or postal address inviting the patient to attend to the Orchard Clinic
b) For participants, the research staff would:
    a. Obtain informed consent which will include the use of the stored positive blood samples (if available)
    b. Administer the research questionnaire
    c. Collect blood samples

d. Perform a clinical assessment to participants

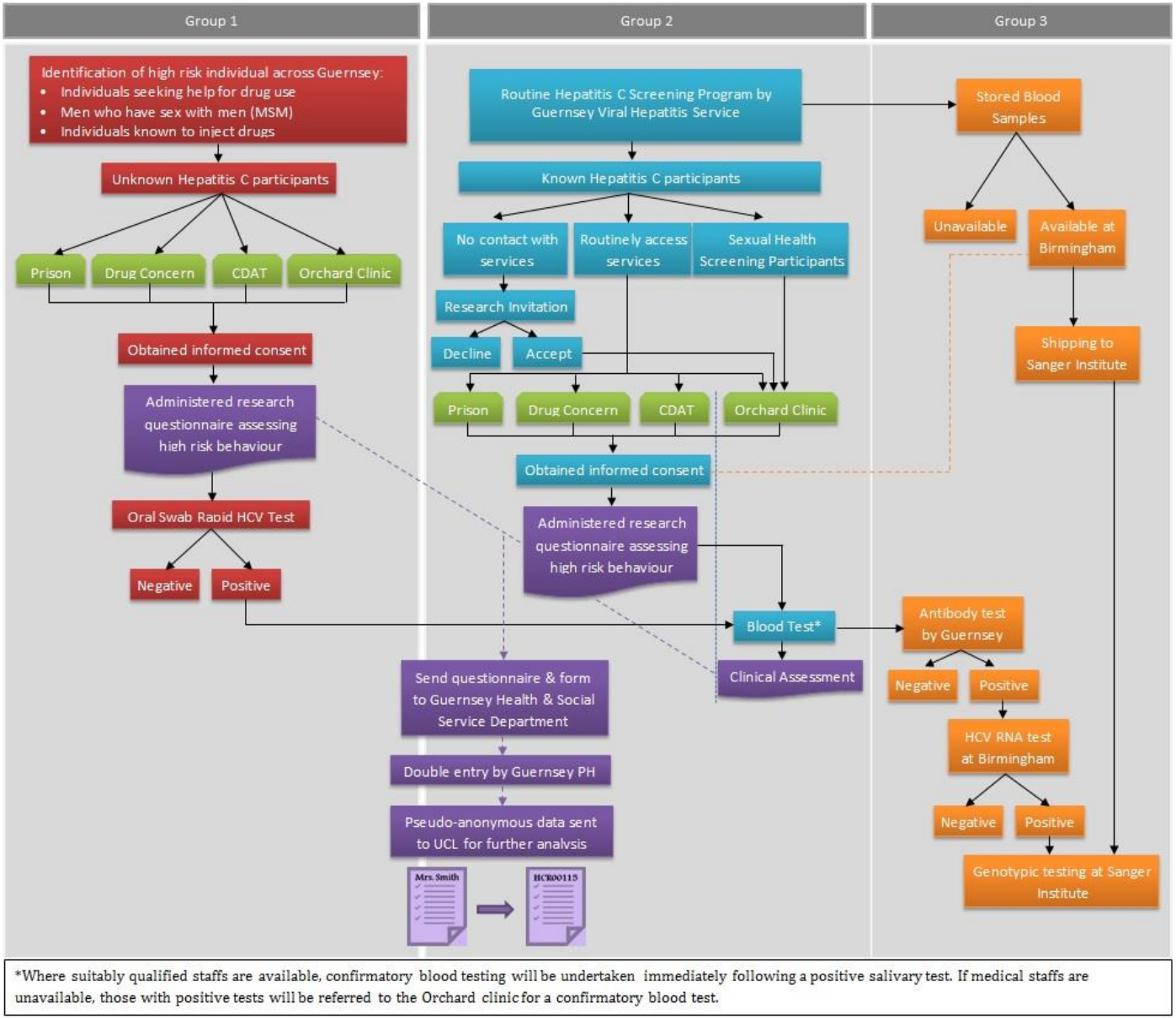
For subgroup c) individuals attending sexual health screening (which routinely includes the offer of hepatitis C testing) at the Orchard Clinic, the member of staff would:

a) Obtain informed consent which would include the use of the HCV antibody test result as part of sexual health screening
b) Administer the research questionnaire
c) Collect blood samples
d) Perform a clinical assessment to participants

C. **Stored HCV Positive Blood Samples** (see diagram in figure 4.1)

The Orchard Clinic routinely sent blood samples from patients to Birmingham for diagnostic testing. Some of the positive blood samples are stored in Birmingham and may be available for further analysis. The detail of blood samples diagnostic in Birmingham is explained in Blood Samples Collection section below. These samples were eligible for inclusion in the research study. The procedure for these samples was as follows:

a) Specimens were identified and labelled
b) Shipping pseudo-anonymised samples to Sanger Institute
c) Performed Whole Genome Sequencing Analysis



*Where suitably qualified staffs are available, confirmatory blood testing will be undertaken immediately following a positive salivary test. If medical staffs are unavailable, those with positive tests will be referred to the Orchard clinic for a confirmatory blood test.

Figure 2.19 Flow Chart Diagram of Hepatitis C Study in Guernsey

4.3.5 Inclusion / Exclusion criteria

All people who accessed services provided by the four project sites with identified risk factors for HCV infection were eligible for inclusion in the study.

This included:

- Individuals seeking help for drug use
- Men who have sex with men (MSM)
- Individuals known to inject drugs
- Patients with previously diagnosed hepatitis C infection

Exclusion criteria: individuals unable to give informed consent in English, age younger than 16, and those who lived temporarily in Guernsey (less than 24 weeks), and HCV infected individuals who had been treated.

4.4 INFORMED CONSENT

Participants recruited at the four project sites would be informed about the study by the designated nursing/medical staff at each site and given a patient information leaflet (Appendix 5). A different information sheet and informed consent were provided for the two groups (known and unknown hepatitis C participants). The project staffs were available on each research site to answer questions relating to the research. Individuals who are willing to participate would be asked to complete and sign a consent form (Appendix 6) administered by the project staff.

4.5 QUESTIONNAIRES

Each time a participant was successfully recruited and sampled, the designated staff at the four main sites would administer a brief research questionnaire (Appendix 7). Data were to be collected on the participant's demographic information (i.e. age, sex, ethnicity), occupation, date of recruitment, year of first entry to Guernsey, previous HIV/HCV/HBV test results, risk factors related with HCV infection, types of drugs used, potential risk-behaviour of HCV transmission, for example sharing needles, MSM, non-sterile drugs injection equipment, and the duration of those risk behaviours.

The completed questionnaires would be submitted to the site coordinator who would send them to the local principal investigator at the Orchard Clinic, Guernsey Health and Social Services. The questionnaire data would be double entered onto a password protected computer using EpiInfo software for data entry quality control.

The clean data collected would be sent to UCL in pseudo-anonymous format to protect participants' confidentiality.

4.6 Laboratory Procedure

4.6.1 Oral Specimens Collection

Oral specimens would be taken from participants with unknown hepatitis C status for rapid screening HCV test. The project nurse/doctor swabbed a testing device along the top and bottom gums of participants then placed it in a solution to process. A control line appeared if the test has run properly, and a result line became visible if the result was positive. The time between specimen collection and test interpretation was approximately 20 to 40 minutes.

Once the result had appeared, the research nurse would record the result on the participant's questionnaire. If the test was positive, a blood test would be performed to confirm this result. Where suitably qualified staff were available, confirmatory testing would be undertaken immediately following a positive salivary test. If phlebotomists were unavailable, those with positive tests would be asked to attend the Orchard clinic for a confirmatory blood test. For people in prison, if there were no qualified staffs available for drawing the blood samples, the medical staff from the Orchard Clinic would arrange blood samples collection on a later date. The local Principal Investigator or designated deputy was also available for immediate post-test counselling, if required.

4.6.2 Blood Samples Collection

Blood samples would have been taken from participants with positive salivary test and for those who were known to have been infected with hepatitis C. Blood specimens would be taken from HCV patients into anticoagulant-treated tubes by a nurse or medical staff. Specimens would be refrigerated to 4^{o}C immediately after collection. Samples collected from Guernsey Prison would be transported immediately to the laboratory at the Princess Elizabeth Hospital.

At the Princess Elizabeth Hospital Pathology Laboratory, all collected blood samples would be stored in -70^{o}C refrigerator. An HCV antibody test would be performed in the Pathology Laboratory at the Princess Elizabeth Hospital to detect the presence of antibodies to the virus, indicating exposure to HCV. Following positive result of HCV antibody test, the specimens would be batched and sent from local laboratory to Birmingham for the detection of HCV RNA. The shipping method

would follow the Guernsey laboratory procedure where the HCV RNA test would follow Birmingham laboratory procedure. When positive HCV RNA test result was found, the nucleic acid extraction of the samples would then be transported to Sanger institute for whole genome sequencing (WGS), using the pipeline that has been set up for the ICONIC study. The shipping method would follow the ICONIC shipment of RNA to the Sanger Institute and full-length sequencing procedure. A minimum of 400µl per viral sample is required for fully automated WGS. These specimens would be labelled linking with the unique study identifier in the questionnaire.

4.6.3 VIROLOGICAL TESTING

HCV positive blood samples stored at Birmingham are kept in RNA extracted form. The specimens would be batched and sent from laboratory directly to the Sanger institute for whole genome sequencing. The samples shipment process would follow Birmingham Standard Operational Procedure (SoP). These specimens would be labelled with unique ID (test under Orchard Clinic number), linking with the unique study identifier in the questionnaire. Once the specimens had been fully sequenced, the rest of the samples would be stored at Sanger Institute. The blood samples would be further analysed for genotypic testing. All of the oral specimens would be collected and disposed of in accordance with local infection, prevention and control guidelines.

4.6.4 CLINICAL ASSESSMENT

Known hepatitis C individuals recruited in the research would complete one page of a clinical assessment form with the designated doctor (Appendix 8). Data would be collected on any liver disease symptoms, diagnosis, drug allergies, responder status, previous antiviral treatment, and sexual health examination. For individuals of unknown hepatitis C status, a clinical assessment would only be conducted separately when there was evidence of an on-going hepatitis C infection in the form of detectable plasma HCV DNA. Participants would be invited to come to the Orchard Clinic to have clinical assessment as well as discuss further management of their hepatitis C infection. For people in prison, the medical staff from the Orchard Clinic would perform the clinical assessment in the prison or at the Orchard Clinic.

4.6.5 STAFFING AND TRAINING

In each research site, at least one member of staff would be recruited and trained for this study. They would be supported by a larger group of trained staff from the Orchard Clinic who would be able to provide on-site support, as required. The recruitment and the training would be coordinated by the Orchard Clinic (Guernsey Health and Social Services Department) and would be conducted at the beginning of the study. The training would consist of three aspects:

1. Oral specimen collection method
2. Interview and administering questionnaire
3. Oral specimens' disposal method

4.7 ETHICAL REVIEW

This study is divided into two ethic submissions for seroprevalence study and genomic study. The research project for seroprevalence study has already attained ethical approval from Guernsey Ethics Committee with registered number IJG/C5.4 (Appendix 9) and UCL Research Ethics Committee with reference number 6988/001 (Appendix 10). The ICONIC study covering whole genome sequencing for HCV has been approved by NRES (NHS) with REC reference number 13/LO/1303.

4.8 STUDY PROGRESS

This study has been registered for UCL Data Protection and UCL insurance and has been granted UCL sponsorship by UCL Joint Research Office (JRO) on 10th November 2015. I also had attained the ethics approval on 23rd of November 2015. Guernsey Health and Social Service Department (HSSD) has already attained ethics approval for the genomic study. Training for local research staff also has been conducted since the beginning of January 2016.

We planned to conduct data collection for the seroprevalence study in Guernsey at the beginning of the year 2016 and receive the first 20 subjects' data to evaluate the reliability of the designed questionnaire. Using the data collected, I planned to assess the prevalence of hepatitis C among high-risk individuals in Guernsey and investigate the HCV genotype distribution and understand the HCV transmission in the Bailiwick of Guernsey to inform future work around hepatitis C treatment and elimination.

Despite having worked closely with the research team in Guernsey to design the study and to obtain ethical approvals at UCL and locally in Guernsey, the study could not be executed according to the original plan due to circumstances beyond my control. The local PI who was initially eager to run the study using local staff became very difficult to contact. Since July 2016 I have been unable to reach the local PI either by telephone or email. I kept trying to contact her until January 2017. My supervisors have also tried to contact the PI, but unfortunately, there has been no response. Due to the limited time available for me to complete my PhD, my supervisors and I decided to drop the two studies in Guernsey.

4.9 Strength and Limitation of the Study Design

Had the study been conducted it would have provided a much better understanding of the prevalence of Hepatitis C and better information on risk factors for HCV. It would also have provided better information on the transmission dynamics of Hepatitis in Guernsey and the extent of imported infection through the use of whole genome sequencing. It would have remained difficult to estimate HCV prevalence in the general population as only high-risk groups were targeted. Also, the success of the genotypic transmission analyses would have depended on the proportion of those infected who were sampled and for whom sequencing was completed.

4.10 Lesson Learned

By preparing this study, there are at least three lessons that I have learned. First, I learned some of the difficulties in building research collaborations between institutions, in this case between UCL Farr Institute of Health Informatics and Guernsey Health and Social services. To ensure the research project will be conducted, I think it is essential to have a written formal letter of collaboration agreement signed by two parties. In this study, we did not include such formal collaboration agreements because most parts of the study were to have been conducted outside of the context of a funded research proposal and theirs was initially a high degree of enthusiasm from the local PI.

Second, I learned step by step the preparations needed in order to conduct the research in Guernsey, which is quite different from what I have done in my country. Previously, I have been involved in international collaboration research, but the studies I performed were only in Indonesia; thus I was familiar with the local procedures. In the preparation of this study, I learned the differences between health care system, the standard of patients screening, the procedure to conduct the research, etc. I also learned that I need to obtain UCL data protection, UCL

insurance & sponsorship, and ethical clearance both in UCL and Guernsey to perform the study. This experience taught me to be prepared, working fast, and quickly adapt to the procedure of the study location, wherever it is.

Third, I agree that communication is one of the most important things in collaboration. We need to maintain communication with the research collaborators and also to be open and communicate if there are any problems. During this preparation, I felt very difficult to reach the local PI, and this was exacerbated by the geographical distance between London and Guernsey. For the future, I believe a productive, and well-established formal relationship between individuals or institutions is important in initiating successful research collaborations. This needs to include some face to face meetings at the proposed study sites to establish relationships between collaborating groups.

4.11 My Role in This Study

I designed the study in collaboration with my supervisors and the Guernsey PI. I drafted the study protocol, and designed the information sheet, informed consent, questionnaire, and clinical assessment form. I was responsible for ensuring that UCL Data Protection, insurance, and sponsorship were in place. I also prepared paperwork for ethical approval in UCL and Guernsey. Initially, I was also responsible to conduct data analysis from data collected, perform monitoring and evaluations of the study, write and present the results of the research, and propose recommendation for local government related with appropriate intervention and prevention plans for HCV elimination program on the island. However, due to circumstances outlined above I was unable to perform these roles.

Key Points:

- Guernsey provides a unique opportunity to assess the feasibility of hepatitis C elimination because it has a closed population, well-established health care services, close inter-agency collaboration, well-characterized high risk populations, and funding opportunity to treat infected high-risk individuals with new DAA's.
- A cross sectional study with a nested cohort was planned to recruit individuals who are at high risk of HCV infection, including individuals seeking help for drug use, men who have sex with men (MSM), individuals known to inject drugs, and patients with previously diagnosed hepatitis C infection.
- I have already designed the study in collaboration with my supervisors, developed a collaboration to do this work obtained ethics approvals both in UCL and Guernsey, obtained approved UCL data protection and sponsorship, however, we could not conduct the study due to withdrawal of engagement by the local PI.
- The lesson learned by preparing this study are: 1) Building a research collaboration is necessary and we need to provide research collaboration agreement to ensure the implementation of study; 2) we have to learn and adapt the procedure of conducting research in the research country well; 3) Communication, including face to face meetings and visits to research sites is important in collaboration
- By doing the preparation of the study, I learned how to develop a good research proposal, designing appropriate study design, drafting questionnaire, how to get ethics approval, etc.

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5. Hepatitis C among Vulnerable Populations: A Seroprevalence Study of People who are Homeless, People Who Inject Drugs and Prisoners in London, UK

Chapter's Aim:
The aim of this chapter is to estimate the prevalence of and risk factors for hepatitis C infection among vulnerable populations such as people who are homeless, people who inject drugs, and prisoners. This study was conducted between May 2011 and June 2013 in London and recruited participants from hostels for the homeless, drug treatment services targeting homeless people and a prison through the Find & Treat Service, a specialist outreach team alongside with NHS and third sector front-line services.

5.1 INTRODUCTION

In high income countries, the burden of hepatitis C falls on People Who Inject Drugs (PWID), so prevalence is also likely to be high in marginalised populations where injecting drug use is common such as in homeless people and prisoners. Several studies have estimated the prevalence of hepatitis C among high risk groups such as PWID (1-8), but there have been fewer studies reporting the prevalence of hepatitis C virus (HCV) among prisoners (9-12), people who are homeless (13-16), and migrants from countries with high risk of HCV (17). More data are needed to better estimate the prevalence of HCV in these marginalised populations which could support the development of targeted strategies to reduce HCV infection and transmission in these groups.

Worldwide estimates of the prevalence of anti-HCV amongst drug-users vary widely between 9.8% in Paraguay to 97.4% in Mexico (18). In a recent UK survey across seven cities in England, the prevalence of HCV amongst people who inject drugs was estimated to vary between 27% in Middlesborough and 74% in Greater Manchester(19).

People who are homeless are also known to have high levels of exposure to injecting drug use such that services for homeless people may be good venues to screen for HCV and access populations of drug users who may otherwise be

difficult to reach. Homeless people have higher risk of blood borne infections, including hepatitis C (20) and they tend to have higher rates of morbidity and mortality (21). Data on prevalence of HCV among people who are homeless is quite limited because they are not usually included in studies. A meta-analysis performed in 2012 showed a random-effects pooled prevalence of HCV infection among people who are homeless as being 20.3% (95% CI 15·5–25·2).(22) Another study in Oxford reported 26.5% of people who are homeless infected with HCV.(23)

In addition, prisoners also have a high risk of infectious disease. A study from 2000 estimated HBV and HCV prevalence in 8 prisons across England and Wales found that 7% of participants were HCV-antibody positive.(24) Work conducted in 5 Scottish prisons in 1999 reported 20.3% prevalence with 95% CI: 18.3%–22.3%.(25) However, there may be some classification bias as both studies did not perform confirmatory HCV-RNA testing. A recent study performed in 2013 in Scottish prisons also found evidence of higher HCV prevalence among inmates in prison compared to the general population, estimating 19% prevalence. 53% of these reported a history of injecting.(26)

Considering the uncertainty and wide range of HCV prevalence among people who are homeless, drug users and prisoners, the aim of this study is to estimate the prevalence of and risk factors for HCV infection in unselected individuals who are susceptible to HCV by virtue of being homeless, in contact with drug-treatment services targeting homeless populations or in prison.

5.2 METHODS

5.2.1 STUDY POPULATION

I analysed data derived from a cross sectional study conducted between May 2011 and June 2013 in London. This was part of a previously published study where the primary purpose was to assess prevalence of latent tuberculosis infection (LTBI) and the risk of progression from latent infection to active disease, but the study also tested all those recruited for blood borne viruses (BBV) providing the opportunity for secondary analyses to measure prevalence of hepatitis C infection.(27) BBVs were measured in the original study as it was important to understand the levels of BBVs and LTBI co-infection due to their effects on eligibility and cost effectiveness of LTBI treatment. Patients were recruited from 39 homeless hostels and 20 drug treatment services that targeted homeless populations through the National Health Service Find and Treat (F&T) Service (28). The homeless hostels and drug treatment

services recruited in this study were venues across London that Find & Treat offers screening for active tuberculosis using mobile chest X-rays. The team consists of multidisciplinary staff such as social and outreach workers, TB nurse specialists, radiographers and expert technicians as well as including former TB patients who work as peer advocates. Mobile Tuberculosis (TB) X-Ray screening by F&T service is operating across London at a variety of sites to provide screening and testing for marginalised groups who have difficulty accessing mainstream healthcare. The service screens almost 10,000 high-risk people every year, covering every London borough.

As the study was originally designed to assess prevalence of latent tuberculosis infection and subsequent disease progression rates, participants were only eligible for the study if they had a recent chest X-ray through Find & Treat or another NHS provider that could help exclude active tuberculosis. In prison the requirement for a previous X-ray was met through screening alongside a prison X-ray screening service that was also being evaluated within the same programme grant. 511 inmates were recruited from a category B prison (a prison which does not require maximum security, but with inmates still recognised as being 'high risk' and requiring significant security measures to ensure they do not escape) in London by a separate team (TB Reach research staff) employed by the study. They were not able to recruit many high-risk prisoners in this study because the X-ray facilities for screening for active tuberculosis had been built in a separate wing to the drug detoxification wing. Since prior radiographic screening was an entry criterion for the study, this systematically reduced the chance of high-risk prisoners with substance use disorders being included in the study.

5.2.2 PRIMARY DATA COLLECTION

5.2.2.1 HOMELESS HOSTELS

A cross-sectional survey was conducted in homeless hostels in London with follow up of outcomes from onward referrals into NHS services. The primary outcome was prevalence of LTBI. Recruitment for the study took place between May 2011 and June 2013. The eligibility criteria included age 18 or over, resident at a homeless hostel on the day of F&T screening, tuberculosis screening chest radiograph by F&T within the last 6 months (to help rule out active tuberculosis), and ability to provide written informed consent. On the basis of screening results, individuals with current hepatitis B or hepatitis C infection or previously undiagnosed HIV infection were referred to 14 local health services and referral outcome data were collected 12

months after referral by the research team phoning and speaking to clinicians and nurses to whom the patients were referred.

The survey invited 804 individuals to participate following F&T mobile X-Ray screening. 542/804 (67.4%) consented to take part. Fifty-one (9.4%) individuals were subsequently excluded, mainly due to a lack of venous access for blood sampling (n=31). Therefore, 491 individuals were included in the analysis. The screening was conducted alongside the Mobile X-Ray Unit, which has a high turnover. It was not possible to approach, consent and gain venous access in a high proportion of eligible participants under this arrangement. Since completion of X-ray screening (to help rule out active disease in those with evidence of LTBI) was an eligibility criterion, this also limited the recruitment. It also often proved difficult to identify private spaces for taking blood.

5.2.2.2 Drug Treatment Services

The process for recruitment at drug treatment services screened by Find & Treat was the same as for homeless hostels (i.e. participants were recruited by research nurses working alongside with Find & Treat who approached those screened to recruit them for LTBI and blood borne virus screening according to capacity). Data on the proportion of those invited to participate who agreed were not available for this setting but were anecdotally reported to be similar to those within hostel settings.

5.2.2.3 Prison

A cross-sectional survey took place over a 6-month period between January 7th and June 28th 2013 at a male prison in London with a static digital X-ray facility for TB screening, and existing onward management of tuberculosis and blood borne viruses. Newly arrived prisoners were eligible for the TB X-ray screening, excluding those with a previous chest X-ray in the last 6 months. Existing prisoners were offered X-ray screening when possible. Any prisoner participating in the X-ray screening was eligible for the LTBI/BBV screening. No other specific exclusion criteria applied. The screening was conducted as 3 hourly sessions five days a week, either in the morning or afternoon. Prisoners with an active addiction problem are typically initially housed in a separate wing of the prison (the detoxification wing). As the X-ray facilities were built in a different wing, this meant that prisoners undergoing detoxification were systematically less likely to be screened than other prisoners as transfer between wings required additional security staff to be involved.

All prisoners who had received a chest X-ray with the TB screening programme were eligible to participate in the LTBI/BBV screening. Following the X-ray, interest to participate was noted on the X-ray list. Prisoners located on the admissions wing were consented and screened immediately when possible or encouraged to present themselves to the study clinic during free-flow (free time). The study team also knocked on cell doors on the admissions and substance misuse wings and requested officers to unlock interested prisoners to participate. Some participants were booked into a weekly phlebotomy clinic and escorted by prison officers.

A phlebotomy clinic was run weekly for consented prisoners where a blood sample could not be taken on the day. A link was set up with the prison IT system and latent TB and BBV results were fed into the prisoner's medical record. Results were also sent to the study team via email. Positive BBV results were phoned through directly to the study team by the virology department. Positive results were given in person to prisoners within 2 weeks by the study team for LTBI, or by the prison GP for BBV's, together with a letter explaining the results. Negative results were only fed back upon request. Participants with Hepatitis B, C and HIV infection were referred to local health services depending on existing local arrangements or asked to contact their GP to make a referral upon their release (HBV, HCV only).

For the radiographic screening (which was a prerequisite to the study of LTBI and BBV, a total number of 1491 chest X-rays were taken on 1484 individuals. There were 3032 new arrivals during the study, of which 618 (20%) were to the substance misuse wing. The overall screening coverage of new prisoners was 43% (1302/3032). The vast majority of X-rays were taken from new arrivals (87%), 1242 (84%) were from prisoners on the admission wing, 158 (11%) from the substance misuse wing, and 91 (6%) from other wings. The total number of active screening days was 112, with an average of 13 X-rays/day.

Of the 1484 of prisoners screened with a chest X-ray, 1444 were eligible to take part in the LTBI and BBV screening. 595 were approached by the research team, and 88% of these consented to partake in the study. The final analysis included 511 individuals. Despite prisoners being a "captive" population the team still experienced considerable challenges in recruitment due to operational restrictions within this setting. Prisoners were out of their cells for 3 ½ hours per day, when screening competed with paid prison work, attending courses that could lead to transfer to a lower category prison or earlier release on probation, exercise and social visits. Access outside of these times was very limited. During lockdown (security alert or

staff meetings) no movement among prisoners was allowed which further impacted on the screening uptake. This, and restricted operational capacity within the research team, meant that were unable to approach all eligible participants in the studies. Due to the study setting, a sampling framework was not possible for recruiting patients and therefore convenience sampling was used.

5.2.2 INFORMED CONSENT AND QUESTIONNAIRE

The research staff visited each study setting and provided information sheets for those eligible. Participants who agreed to join the study were required to complete and sign a consent form. A questionnaire (Appendix 11) was administered and completed by researchers employed by the study to collect demographic information (i.e. age, sex, ethnicity, country of birth), information on previous HCV test results, smoking status and risk behaviours and their duration including: history of imprisonment, homelessness, and drug taking (types of drugs, drug use duration, and needle sharing). The questionnaire was developed by the F&T service and TB Reach team and it has been piloted to all three targeted populations.

5.2.3 LABORATORY TESTING

Venous blood samples were taken from participants into anticoagulant-treated tubes by research nurses. Samples were refrigerated immediately after collection and transported to the laboratory to be tested at the Royal Free Hospital for hepatitis C. The collected specimens were labelled with unique ID linking with the study identifier in the questionnaire. Anti-HCV antibody was detected using Vitros chemiluminescence assay (Ortho Clinical Diagnostics). HCV-RNA was detected using PCR Assay or Abbott M2000 Real-Time hepatitis C assay. When samples were found to be reactive with anti-HCV but negative for HCV-RNA, further confirmation was done using Recombinant Line Immunoassay (INNO-LIA, Innogenetics) or Immuno Blot Assay (RIBA, Chiron). Patients were categorised as currently infected with HCV if they had a positive HCV-RNA and a positive antibody-HCV test. Patients with past infection were identified when they had a negative HCV-RNA and a positive antibody-HCV test. Ethical approval for this study was obtained from the East of England – Essex National Research Ethics Service Committee (reference number 10/H0302/5).

Samples were also tested for latent tuberculosis infection (LTBI), hepatitis B and HIV. Latent tuberculosis was measured using QuantiFERON-TB Gold gamma interferon release assay (Cellestis, Australia) and defined positive if the TB specific antigen response was >0.35 IU/ml and there was no evidence of active disease on

clinical assessment. HIV infection was assessed using the Architect combined HIV antibody/p24 antigen chemiluminiscence assay (Abbott Diagnostics). Hepatitis B was detected by the Architect immunoassay (Abbott Diagnostics, Germany). Hepatitis B current infection was defined as HBsAg positive, anti-HBc positive, anti-HBs negative. Hepatitis B past infection was defined as HBsAg negative, anti-HBc positive, anti-HBs positive or anti-HBs negative.

5.2.4 STATISTICAL ANALYSIS

From this study, I assessed the proportion of participants who had HCV current and past infection. A descriptive analysis was conducted to investigate the relationship between HCV status and the following variables: participants' age, sex, ethnicity, history of homelessness and imprisonment, alcohol, drug use, smoking and needle sharing behaviour. Problem alcohol use was defined as whether participants had ever been concerned about their drinking or had a health worker express concern about their alcohol consumption. I undertook univariate and multivariate logistic regression to identify factors associated with HCV infection and identified if there was any interaction between age group and duration of injecting variables. Forward stepwise method, with a likelihood ratio test threshold *p*-value of 0.05 was used for entry and 0.10 for removal of a variable was used to determine the best model for multivariate logistic regression. No imputation was done for missing data as I used pairwise deletion for treating missing data in the dataset.(29) Pairwise deletion, or available case analysis, using assumption of the MCAR (Missing Completely at Random), only removes the specific missing values from the analysis (not the entire case) thus it maximizes data availability increasing power in the analysis. A disadvantage with the use of pairwise deletion is that the standard errors computed by most software packages uses the average sample size across analyses thus for specific variables standard errors may be over or underestimated depending on the completeness of the data for that variable. Since all variables had a high level of completeness this is unlikely to have appreciably affected results. These analyses produced adjusted odds ratios and 95% confidence intervals for past or current HCV infection for the range of putative risk factors. Furthermore, I also assessed the odd ratios and 95% confidence interval for HCV infection among non-injectors and risk factors for HCV spontaneous clearance (based on those with antibody but with negative HCV PCR results). Venn diagrams were also created to show the proportion of individuals with overlapping risk factors for HCV relating to homelessness, drug use and imprisonment. I analysed data using Statistical

Package for Social Sciences SPSS version 20.0 (SPSS Inc., Chicago, USA) and generated the proportional Venn diagram using Venn Diagram Plotter (PPNL).

Power Calculations

Power calculations for this study were originally based on the ability to measure progression rates from latent tuberculosis to active disease. Subsequently, given challenges in recruitment these were revised to be based on the accuracy with which prevalence of LTBI could be measured. It is therefore possible that the study was underpowered to measure a wide range of risk factors such that some associations between risk factors and hepatitis C infection may have been missed.

5.3 RESULTS

Across the three settings 1207 participants were recruited during the study period, including 511/1207 from prison (42.34%), 491/1207 from homeless hostels (40.7%), and 205/1207 from drug treatment centres (17.0%) (Table 5.1). More than 90% (1093/1203) of participants were male and over half were aged 30-49 years (614/1204). 19% of participants (228/1204) were aged 50 years or older. 65.8% (794/1205) of participants were UK-born.

Almost three-quarters of participants (885/1207) reported they had been in a UK prison (including the 511 recruited at this venue) and sixty percent (693/1157) had been homeless at least once in their lives (including the 491 recruited at homeless hostels/shelters). Risk behaviours that were common among participants included: smoking (980/1207, 81.2%); problem alcohol use (408/1207, 33.8%) and drug use. Almost half of participants reported ever having either smoked heroin/crack and/or injected drugs in their life time (529/1205, 43.9%). The level of missing data in this analysis is less than 5% (Table 5.1).

Table 2.6 Characteristics of participants - HCV current infection is defined for patients with a positive HCV-RNA and a positive antibody-HCV test. HCV past infection is defined for patients with a negative HCV-RNA and a positive antibody-HCV test. The total infection shows the number for both current and past infections.

Characteristics		n	%	HCV-current infection	%	HCV-past infection	%	Total HCV infection	%
All		1207		98	8.1	38	3.2	136	11.3
Research Sites*	Homeless Residential Site	491	40.7	51	10.4	14	2.9	65	13.2
	Drug Treatment Service	205	17.0	31	15.1	20	9.8	51	24.9
	Prison	511	42.3	16	3.1	4	0.8	20	3.9
Sex	Male	1093	90.6	86	7.9	32	2.9	118	10.8
	Female	110	9.1	11	10.0	6	5.5	17	15.5
	Missing	4	0.3	1		-	-	1	
Age group	18-29 years	362	30.0	8	2.2	2	0.6	10	2.8
	30-49 years	614	50.9	66	10.8	26	4.2	92	15.0
	50+ years	228	18.9	23	10.1	10	4.4	33	14.5
	Missing	3	0.2	1		-		1	
Country of birth & Ethnicity	UK-white	542	44.9	60	11.1	26	4.8	86	15.9
	UK-others	231	19.1	11	4.8	1	0.4	12	5.2
	Non UK-white	187	15.5	22	11.8	7	3.8	29	15.5
	Non UK-others	199	16.5	4	2.0	4	2.0	8	4.0
	Missing	48	4.0	1		-	-	1	
Have been in UK prison*	Currently in prison	511	42.3	16	3.1	4	0.8	20	3.9
	Had been in prison before	374	31.0	58	15.5	25	6.7	83	22.2

Characteristics		n	%	HCV-current infection	%	HCV-past infection	%	Total HCV infection	%
	No	322	26.7	24	7.5	9	2.8	33	10.3
Have been in prison outside UK	Yes	83	6.9	12	14.5	4	4.8	16	19.3
	No	1069	88.6	70	6.6	31	2.9	101	9.5
	Missing	55	4.6	16		3		19	
Time spent homeless	Never	464	38.4	20	4.3	10	2.2	30	6.5
	< 1 year	350	30.0	29	8.3	10	2.9	39	11.1
	> 1 year	237	19.6	30	12.7	13	5.5	43	18.1
	Yes (unknown duration)	106	8.8	3	2.8	3	2.8	6	5.7
	Missing	50	4.1	16		2		18	
Illicit drug use	Neither	676	56.0	7	1.0	9	1.3	16	2.4
	Ever smoked heroin/crack only	317	26.3	8	2.5	2	0.6	10	3.2
	Inject drugs - no needle sharing	128	10.6	47	36.7	17	13.3	64	50.0
	Inject drugs with needle sharing	84	7.0	35	41.7	10	11.9	45	53.6
	Missing	2	0.2	1		-	-	1	
Duration of injecting	Non-injectors	993	82.3	15	1.5	11	1.1	26	2.6
	Injecting for <1 year	67	5.6	15	22.4	3	4.5	18	26.9
	Injecting for 2-9 years	61	5.1	23	37.7	11	18.0	34	55.7
	Injecting for ≥10 years	57	4.7	29	50.9	11	19.3	40	70.2

Characteristics		n	%	HCV-current infection	%	HCV-past infection	%	Total HCV infection	%
	Missing	29	2.4	16		4		20	
Smoker*	Yes	980	81.2	93	9.5	35	3.6	128	13.1
	No	227	18.8	5	2.2	3	1.3	8	3.5
Has alcohol problem*	Yes	408	33.8	45	11.0	19	4.7	64	15.7
	No	799	66.2	53	6.6	19	2.4	72	9.0

**Variable with non-missing data*

5.3.1 HCV INFECTION

Laboratory testing revealed that 98/1207 participants (8.1%, 95% CI: 6.7%-9.8%) were currently infected with hepatitis C and 38/1155 (3.2%, 95% CI: 2.4%-4.5%) had a history of hepatitis C infection. This suggests that (38/136) 27.9% (95% CI: 20.8%-36.4%) of HCV infected individuals had cleared the virus spontaneously (none had previously been treated for Hepatitis C). Co-infection was common among patients with active or previous HCV. For example, amongst the hepatitis C current infected patients 57/98 (58.2%, 95% CI: 47.8%-67.9%) were co-infected with hepatitis B virus, 21/98 (21.4%, 95% CI: 14.0%-31.1%) had LTBI and 3/98 (3.1%, 95% CI: 0.8%-9.3%) had HIV (Figure 5.1). Hepatitis C co-infection among past infection and total infection can be seen in figure 5.1. Overall, 15.1% (31/205) of participants recruited in drug treatment services, 10.4% (51/491) of individuals recruited through homeless hostels and 3.1% (16/511) recruited in prison in this study had current HCV infection (Table 5.1).

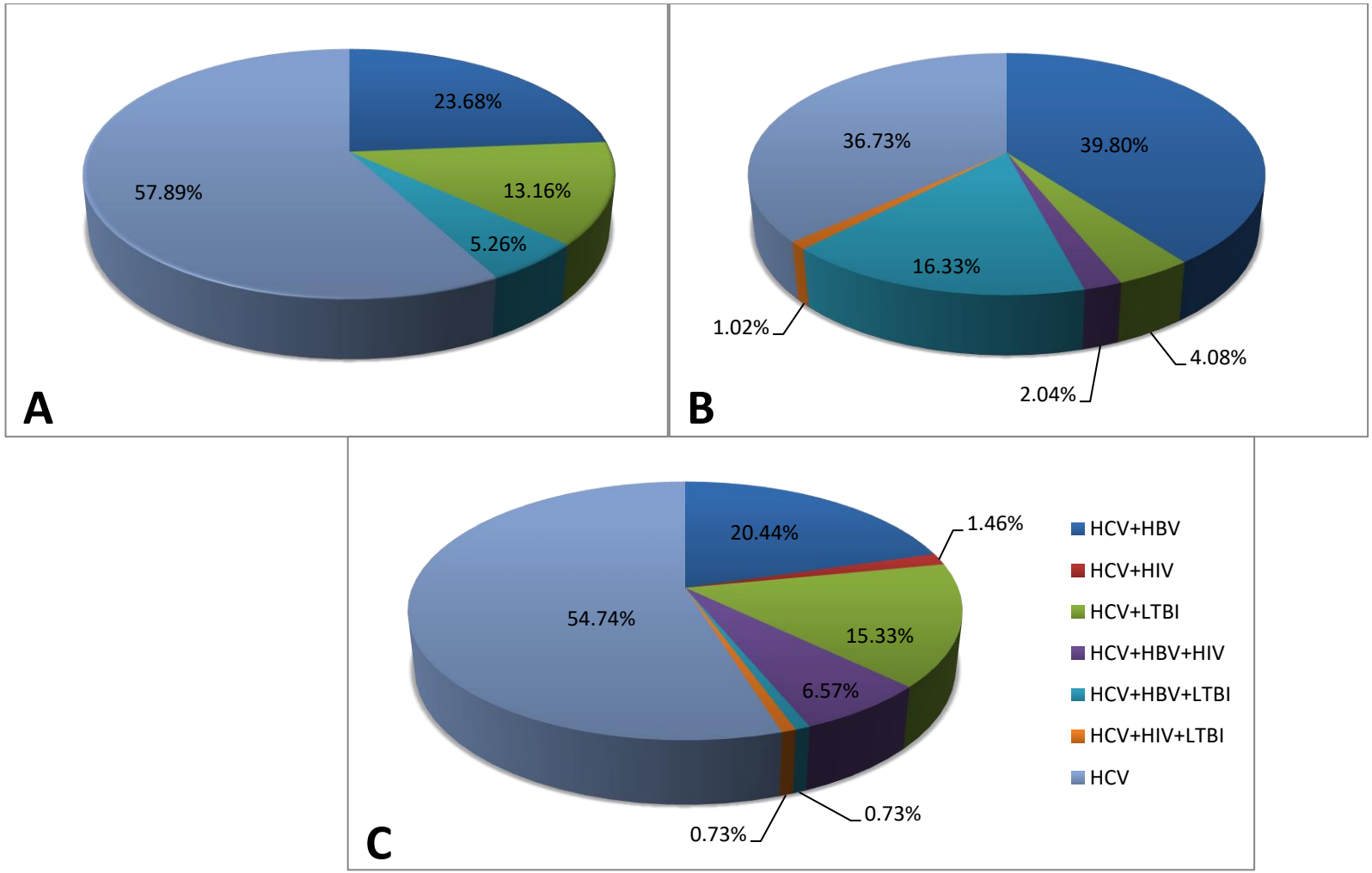

Figure 2.20 HCV Infection and Coinfection among Participants with (A) HCV Past (B) HCV Current Infection and (C) HCV among Current and Past Infection – Panel A shows the proportion of patients with past HCV infection (n=38) who had additional infections. Panel B shows equivalent data for the 98 participants with current HCV infection and panel C shows this for the 136 patients with past or current infection.

5.3.2 Overlapping Risk Factors

Overlapping risk factors were common among all the three groups. For example, among participants recruited in the prison, 173/511 (33.9%) also had a history of drug-use and 136/511 (26.6%) also had a history of homelessness (Figure 5.2). Similarly, among those recruited in homeless hostels, 194/491 (39.5%) also had a history of drug use and 263/491 (53. 6%) also had a history of imprisonment. 110/205 (53.7%) of participants recruited in the drug treatment centres also had a history of homelessness and 122/205 (59.5%) had been in prison.

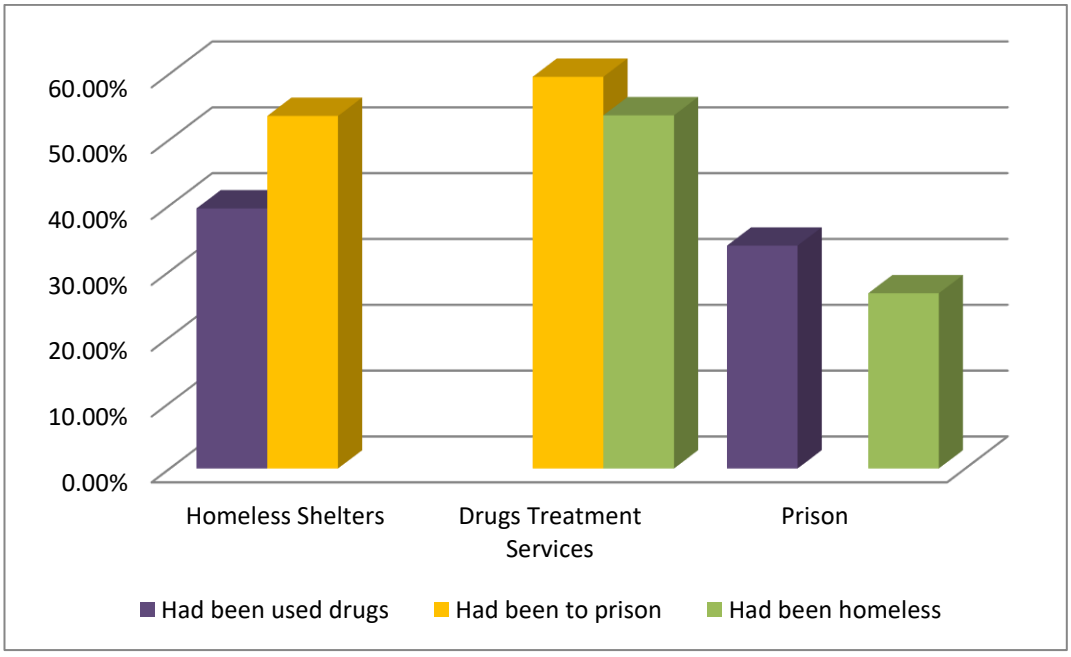

Figure 2.21 Overlapping Risk Factors among People who are Homeless, PWID and Prisoners. The figure, for participants recruited in each setting (homeless hostels n=591), drug treatment services (n=205) and prison (n=511).

Further analysis of overlapping risk factors found that 56.6% (77/136) of HCV infected participants had a history of all three of homelessness, drug use and imprisonment (Figure 5.3), 27.3% (37/136) of HCV infected individuals had 2 of these risk factors and 15.4% (22/136) of them had only one risk factor.

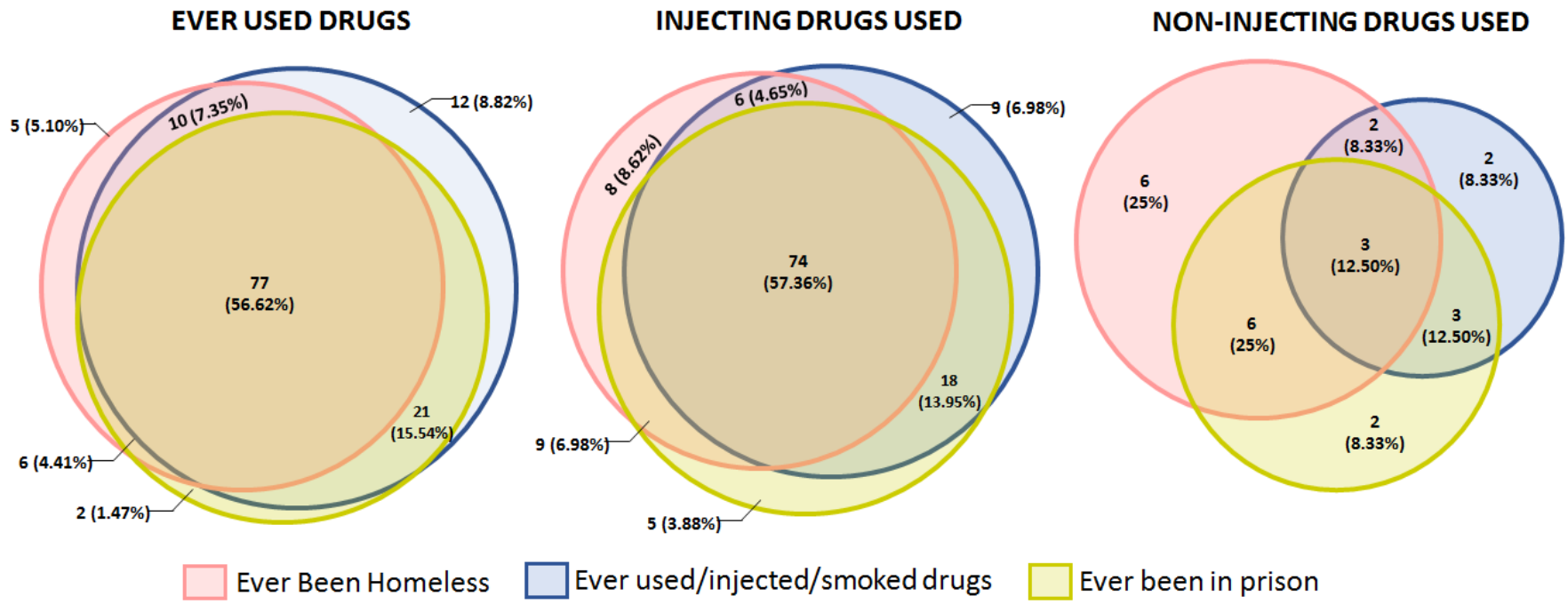

Figure 2.22 Overlapping Characteristics of Being Homeless, IDUs and Have Been in Prison among HCV infected individuals who were (A) Ever used drugs population (B) Injecting Drugs Users population (C) Non-injectors

When I analysed drug use behaviour among three recruitment settings (homeless residential sites, drug treatment services, and prison), we can see in figure 5.4 that in the drug treatment services had the highest number of people injecting drugs and sharing needles (14.6%, 29/199), compared to people in homeless shelters (6.0%, 27/448), and prison (3.7%, 19/511).

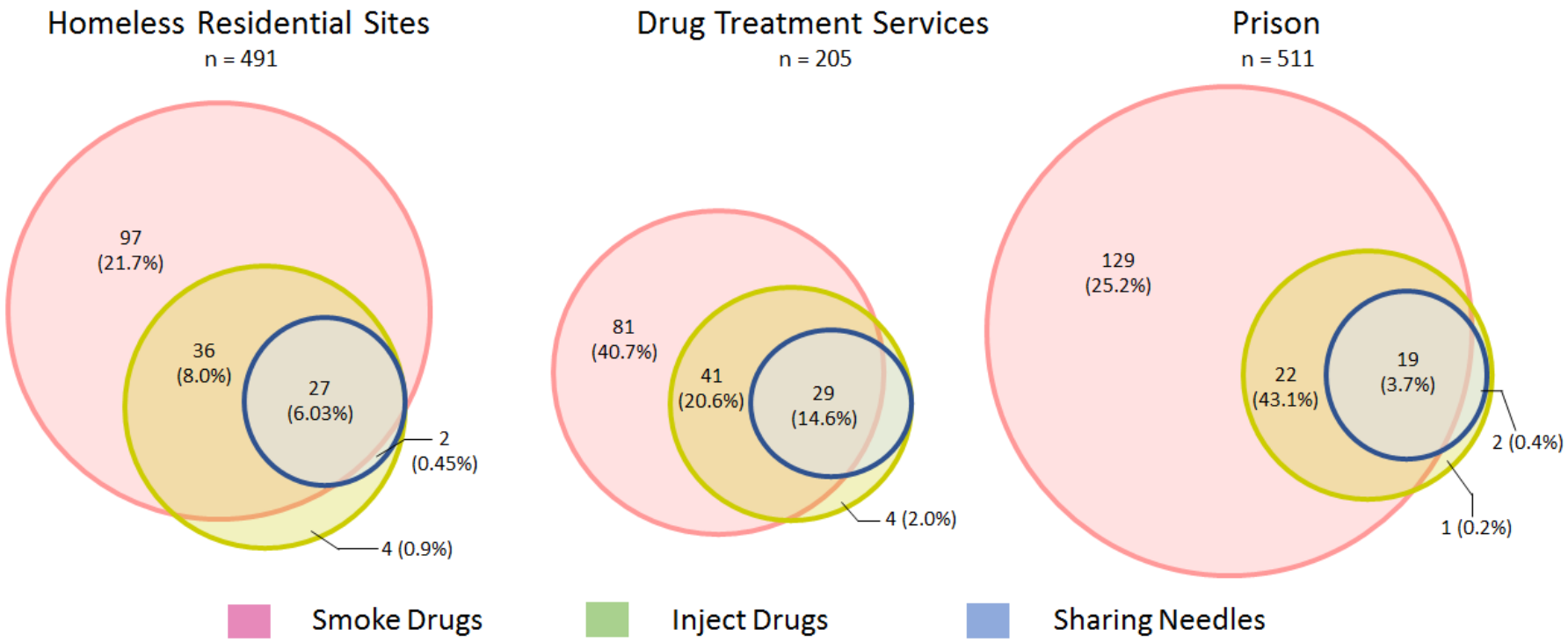

Figure 2.23 Drug Use Behaviour among participants recruited in homeless residential sites, drug treatment services and prison

5.3.3 HCV RISK FACTORS

Risk Factors for HCV Infection

80% (109/136, 95% CI: 72.3% - 86.3%) of HCV infected participants in this study reported injecting drug use. The univariate analysis suggested that many factors were associated with HCV infection including: age >30 years, ethnicity (UK born non-white and non-UK born non-white), imprisonment outside UK, having a history of homelessness, illicit drug use, duration of injecting, smoking and alcohol drinking behaviour (Table 5.2). In the adjusted analysis, only longer duration of injection drug use, being aged more than 30 years and UK born non-white ethnicity were strongly associated with HCV infection. Compared to 18-29 years old, the odds of infection were five-fold greater in participants who were 50 years or older (OR=5.55, 95% CI: 2.25-13.70). Those who were born in UK and non-white were less likely to be infected with HCV (OR=0.38, 95% CI: 0.15-0.99) compared to those who were born in UK and white. The odds of HCV infection were very strongly associated with duration of injecting. The longer duration of injecting, the higher HCV risk of infection found (OR=12.62, 95% CI: 6.22-25.57 for those who injected drugs less than 1 year, OR=50.04, 95% CI: 24.80-100.95 for those who injected drugs for 2-9 years, and OR=67.34, 95% CI: 32.29-140.46 for those who injected drugs more than 10 years compared to the odds of infection in those who were non-injectors.

Table 2.7 Univariate and Multivariate Logistic Regression Risk Factor Analysis of Hepatitis C Past & Current Infection

Risk Factors		n (total)	%	n HCV (+)	%	Past and Current HCV Infection	
						Univariable OR	Multivariable OR
Sex	Female	110	9.1	17	15.5	1	
	Male	1093	90.6	118	10.8	0.67 (0.398, 1.16)	
Age group	18-29 years	362	30.0	10	2.8	1	1
	30-49 years	614	509.0	92	15.0	**6.20 (3.19, 12.08)**	**3.00 (1.33, 6.76)**
	50+ years	228	18.9	33	14.5	**5.92 (2.86, 12.28)**	**5.55 (2.25, 13.70)**
Ethnicity	UK-white	542	44.9	86	15.9	1	1
	UK-others	231	19.1	12	5.2	**0.29 (0.16, 0.54)**	**0.38 (0.15, 0.99)**
	Non UK-white	187	15.5	29	15.5	0.97 (0.61, 1.53)	1.78 (0.96, 3.31)
	Non UK-others	199	16.5	8	4.0	**0.22 (0.11, 0.47)**	0.89 (0.36, 2.19)
Ever been in prison	Had been in prison before	374	31.0	83	22.2	1	
	Currently in prison	511	42.3	20	3.9	1.16 (0.77, 1.76)	
Ever been in prison outside UK	No	1069	88.6	101	9.5	1	
	Yes	83	6.9	16	19.3	**2.31 (1.29, 4.15)**	
Rough sleeping/homeless	Never	464	38.4	30	6.5	1	
	< 1 year	350	30.0	39	11.1	**1.81 (1.10, 2.97)**	
	> 1 year	237	19.6	43	18.1	**3.20 (1.95, 5.26)**	
	Unknown Duration	106	8.8	6	5.7	0.86 (0.35, 2.13)	
Illicit drug use	Neither	676	56.0	16	2.4	1	
	Has ever smoke heroin/crack only	317	26.3	10	3.2	1.34 (0.60, 2.99)	
	Inject drugs - no sharing needles	128	10.6	64	50.0	**41.78 (22.80, 76.56)**	
	Inject drugs with sharing needles	84	7.0	45	53.6	**48.70 (25.23, 93.99)**	
Duration of injecting	Non-injectors	993	82.3	26	2.6	**1**	**1**
	Injecting for <1 year	67	5.6	18	26.9	**13.90 (7.14, 27.10)**	**12.62 (6.22, 25.57)**
	Injecting for 2-9 years	61	5.1	34	55.7	**48.49 (25.51, 92.16)**	**50.04 (24.80, 100.95)**

Risk Factors		n (total)	%	n HCV (+)	%	Past and Current HCV Infection	
						Univariable OR	Multivariable OR
	Injecting for ≥10 years	57	4.7	40	70.2	**87.24 (43.83, 173.63)**	**67.34 (32.29, 140.46)**
Smoker	No	227	18.8	8	3.5	1	
	Yes	980	81.2	128	13.1	**4.12 (1.99, 8.58)**	
Has alcohol problem	No	799	66.2	72	9.0	1	
	Yes	408	33.8	64	15.7	**1.88 (1.31, 2.70)**	

Risk factors for HCV in Those who Reported not Injecting Drugs.

Since 20% of HCV cases in this study occurred in people who reported not having used drugs I undertook a separate analysis of risk factors for HCV in those who had never injected drugs. Univariate analysis suggested that many factors were associated with HCV infection among non-injectors including: age 30-49 years old (OR=10.35, 95% CI: 1.36-79.11), age >50 years old (OR=20.07, 95% CI: 2.57-156.66), history of imprisonment (OR=0.43, 95% CI: 0.20-0.94), history of homelessness for over a year (OR=2.90, 9%% CI: 1.03-8.12), and alcohol problem (OR=3.59, 95% CI: 1.61-8.01). Separate multivariate analysis showed that alcohol problems (OR=2.92, 95% CI: 1.24-6.89), increasing age (age 30-49 years old (OR=8.29, 95% CI: 1.06-64.73), and age more than 50 (OR=13.85, 95% CI: 1.67-114.85)) increased the risk of HCV infection among individuals who reported not injecting drugs (Table 5.3).

Table 2.8 Univariate and Multivariate Logistic Regression Risk Factor Analysis of Hepatitis C among Non-Injecting Drugs Individuals

Risk Factors		n (total)	%	n HCV (+)	%	Past & Current HCV infection for Non-Injectors	
						Univariable OR	Multivariable OR
Sex	Female	110	9.1	2	1.8	1	
	Male	1093	90.6	24	2.2	1.16 (0.27, 5.00)	
Age group	18-29 years	362	30	1	0.3	1	1
	30-49 years	614	509	14	2.3	**10.35 (1.36, 79.11)**	**8.29 (1.06, 64.73)**
	50+ years	228	18.9	11	4.8	**20.07 (2.57, 156.66)**	**13.85 (1.67, 114.85)**
Ethnicity	UK-white	542	44.9	14	2.6	1	
	UK-others	231	19.1	1	0.4	0.14 (0.02, 1.04)	
	Non UK-white	187	15.5	5	2.7	0.99 (0.35, 2.79)	
	Non UK-others	199	16.5	6	3.0	0.93 (0.35, 2.47)	
Ever been in prison	Had been in prison before	374	31	11	2.9	1	
	Currently in prison	511	42.3	14	2.7	**0.43 (0.20, 0.94)**	
Ever been in prison outside UK	No	1069	88.6	22	2.1	1	
	Yes	83	6.9	2	2.4	**0.44 (0.20, 0.99)**	
Rough sleeping/homeless	Never	464	38.4	7	1.5	1	1
	< 1 year	350	30	8	2.3	1.77 (0.63, 4.93)	1.11 (0.39, 3.17)
	> 1 year	237	19.6	8	3.4	**2.90 (1.03, 8.12)**	1.18 (0.39, 3.52)
	Unknown Duration	106	8.8	2	1.9	1.26 (0.26, 6.14)	0.64 (0.13, 3.23)
Smoker	No	227	18.8	4	1.8	1	
	Yes	980	81.2	22	2.2	1.55 (0.53, 4.55)	
Alcohol Problem	No	799	66.2	10	1.3	1	1
	Yes	408	33.8	16	3.9	**3.59 (1.61, 8.01)**	**2.92 (1.24, 6.89)**

Risk Factors for Spontaneous Clearance

38/136 (27.9%) participants with evidence of past HCV no longer had detectable HCV RNA. Given that none had been treated this enabled an analysis of risk factors for clearance. In the univariate analysis, HCV infected individuals who had a history of injecting drugs with sharing needles (OR=0.22, 95% CI: 0.07-0.75) or without sharing needles (OR=0.28, 9%% CI: 0.09-0.87) were less likely to achieve clearance than non-injectors. In the multivariate analysis only illicit drug use with needle sharing reduced the likelihood of achieving Spontaneous Viral Clearance/SVC (OR=0.27, 95% CI: 0.08, 0.95) – see table 5.4 below.

Table 2.9 Univariate and Multivariate Logistic Regression Risk Factor of Hepatitis C Spontaneous Clearance – 38 participants with spontaneous clearance included in the analysis below. Spontaneous clearance is defined by the disappearance of HCV RNA in the serum (indicated by a negative HCV-RNA and a positive antibody-HCV test)

Risk Factors		n (total)	%	n HCV (+)	%	HCV Spontaneous Clearance	
						Univariable OR	Multivariable OR
Sex	Female	110	9.1	6	5.5	1	
	Male	1093	90.6	32	2.9	0.68 (0.23, 2.90)	
Age group	18-29 years	362	30.0	2	0.6	1	
	30-49 years	614	509.0	26	4.2	1.58 (0.31, 7.92)	
	50+ years	228	18.9	10	4.4	1.74 (0.31, 9.69)	
Ethnicity	UK-white	542	44.9	26	4.8	1	1
	UK-others	231	19.1	1	0.4	0.21 (0.03, 1.71)	0.23 (0.03, 1.95)
	Non UK-white	187	15.5	7	3.8	0.73 (0.28, 1.93)	0.80 (0.29, 2.19)
	Non UK-others	199	16.5	4	2.0	2.31 (0.54, 9.94)	1.66 (0.32, 8.56)
Ever been in prison	Had been in prison before	374	31.0	25	6.7	1	
	Currently in prison	511	42.3	4	2.0	0.85 (0.33, 2.16)	
Ever been in prison outside UK	No	1069	88.6	31	2.9	1	
	Yes	83	6.9	4	4.8	0.75 (0.23, 2.52)	
Rough sleeping/homeless	Never	464	38.4	10	2.2	1	

Risk Factors		n (total)	%	n HCV (+)	%	HCV Spontaneous Clearance	
						Univariable OR	Multivariable OR
	< 1 year	350	30.0	10	2.9	0.69 (0.24, 1.96)	
	> 1 year	237	19.6	13	5.5	0.87 (0.32, 2.36)	
	Unknown Duration	106	8.8	3	2.8	2.00 (0.34, 11.76)	
Illicit drug use	Neither	676	56.0	9	1.3	1	1
	Has ever smoke heroin/crack only	317	26.3	2	0.6	0.19 (0.03, 1.22)	0.23 (0.04, 1.53)
	Inject drugs - no sharing needles	128	10.6	17	13.3	**0.28 (0.09, 0.87)**	0.38 (0.11, 1.27)
	Inject drugs with sharing needles	84	7.0	10	11.9	**0.22 (0.07, 0.75)**	**0.27 (0.08, 0.95)**
Duration of injecting	Non-injectors	993	82.3	11	1.1	1	
	Injecting for <1 year	67	5.6	3	4.5	0.27 (0.06, 1.18)	
	Injecting for 2-9 years	61	5.1	11	18.0	0.65 (0.23, 1.88)	
	Injecting for ≥10 years	57	4.7	11	19.3	0.52 (0.18, 1.47)	
Smoker	No	227	18.8	3	1.3	1	
	Yes	980	81.2	35	3.6	0.63 (0.14, 2.76)	
Has alcohol problem	No	799	66.2	19	2.4	1	
	Yes	408	33.8	19	4.7	1.18 (0.56, 2.49)	

5.4 DISCUSSION

5.4.1 SUMMARY OF FINDINGS

The study confirmed high risk of past or current HCV infection when screening in drug treatment services (25%, 95% CI: 19%-31%), homeless hostels (13%, 95% CI: 10%-17%) and prison (4%, 95% CI: 2%-6%). The risk in prisoners was likely underestimated due the recruitment process reducing the likelihood of patients in the detoxification wing of the prison being offered radiographic screening (which was an inclusion criteria for the study). There was a very high degree of overlap between these populations. The risk of HCV was driven primarily by injecting drug use. Past or current HCV infection was found in 27% of those injecting less than a year, 56% of those injecting between one and 10 years and 70% of those injecting for over 10 years compared to 3% of those reporting never having injected. Nevertheless 20% of past or current HCV infections were in those who reported not having injected drugs. In these patients HCV infection was more likely in those who were older and those who had alcohol problems, possibly reflecting differential reporting of injecting drug use in these groups. 28% of those with HCV antibody no longer had detectable HCV RNA suggesting spontaneous clearance (none had been treated). This is similar to findings in Chapter 2. Clearance was least likely in those who reported needle sharing. This supports the hypothesis that spontaneous clearance rates are low in PWIDs because of reinfection.

5.4.2 STUDY STRENGTHS AND LIMITATIONS

The strength of this study is it was able to recruit vulnerable populations in London by capitalising on a well-established network through the F&T service. The questionnaire used was piloted with the targeted population. Furthermore, this study was performed by skilled team members who have experience working with vulnerable populations, which should help to improve representativeness. Other strengths of the study were the wide range of risk factors assessed and low levels of missing data. Because the level of missing data is less than 5%, I can treat those with pairwise deletion to maximise data availability for analysis. It is also a strength that I was able to assess a wide range of confounders in this study.

A major challenge when undertaking studies recruiting hard to reach populations is selection bias. The study was only able to recruit individuals who were in contact with drug treatment services, homeless shelters or prisons. This may have affected

the estimates of HCV prevalence as individuals who are not in contact with services may have a higher burden of undiagnosed HCV.

In all settings, recruitment was restricted to those who had a recent Chest X-ray to exclude active tuberculosis). Although Find & Treat target all hostel residents and all those attending drug treatment services on the day of screening, this requirement may have introduced additional biases although it is uncertain in what direction these would affect prevalence estimates. A further limitation is that as the recruitment was alongside high throughput outreach based mobile X-ray screening, it was difficult for staff to obtain records on recruitment rates amongst those eligible or to compare the characteristics of those recruited and eligible people who were not recruited. This makes assessment of the impact of selection bias challenging. However, the perception in hostels and drug treatment services was that recruitment was primarily restricted by the capacity of the team to recruit and obtain blood samples rather than by the characteristics of those eligible.

In prison, the testing was alongside an initiative to screen for active TB using radiography. Since prisoners undergoing drug detoxification were located in another part of the prison (who were unable to access easily the testing facility), estimates of disease prevalence exclude many of these higher risk prisoners. Furthermore, the study only assessed inmates in one prison which might not be representative among 14 prisons in London although this prison had the 3rd largest capacity. This was a high throughput prison with many inmates being on remand or having short sentences, rates in prisons for different offender types may have different infection rates. Another challenge was because participants were asked for self-reported history of homelessness, drugs used, and imprisonment, these reports may be affected by recall bias or reluctance to report these risk factors. In addition, information bias might have occurred because the definition of alcohol problem used in this study was whether participants had ever been concerned about their drinking or had had a health worker express concern about their alcohol consumption, thus I could not measure how much alcohol drinking behaviour participants had.

5.4.3 Discussion of the Results

PWID

This study's estimate of the prevalence of HCV among PWID's is comparable to the prevalence estimate reported by Public Health England (approximately 50% in

England, 32% in Northern Island and 47% in Wales).(30) A multi-centre study published in 2007 reported a wide range of HCV prevalence among PWID across England varying from 27% in Middlesborough, 34% in Exeter, 51% in Reading, 54% in Plymouth, 65% in Bristol, 66% in Central Manchester, to 74% in Greater Manchester, with a total of 1058 participants.(19) Possible explanations for the variation in prevalence estimates include differences in how individuals were identified, study population age and injecting behaviour, such as duration (31) and frequency of injection drug-use (32) as well as needle sharing (33) or sharing drug preparation equipment behaviour (34, 35). Despite the wide-range of prevalence estimates, these studies highlight the importance of focusing efforts on PWIDs if we are to reduce the burden of HCV (36). However, the study identified that homeless hostels are also an important venue to screen for HCV since many homeless people have a history if injecting drug use and these people may not be in contact with drug treatment services.

This study suggested duration of injecting drug use was an important determinant of risk, but that even those injecting for less than 12 months had high levels of HCV. This shows the need to screen drug users early in the course of their injecting history to ensure access to treatment and reduce onwards infection – In addition to the importance of offering access to drug rehabilitation And harm minimisation services. This confirms the well-established link between HCV and injection drug use.(6, 35, 37, 38) The longer duration of injection, the higher risk of infection found. This hypothesis was supported by Lamden et.al (39) who found injecting drugs more than 3 years increased risks up to 3 times higher (40) and Miller et.al study where injecting drugs for 2-3 years increased risk of infection 2 times higher, even up to 10 folds if the duration of injection were longer than 6 years (6).

Some studies also investigated the association of age and the risk of infection. Nyamati et.al found older age than 40 years old (13) increased risk of infection up to 5 folds whereas Miller at.al suggested older age increased risk 1.29 times higher (6). The fact that the association with age remains after controlling for duration of injecting either suggests residual confounding through imprecise measurement of injecting duration or may suggest cohort effects with higher rates of transmission of transmission of HCV in the past (older cohorts) than in more recent (younger cohorts). The specific association between country of birth - ethnicity and HCV infection likely reflects the varied prevalence of HCV internationally (see chapter 1). In this study, 22% of UK born-white were PWID whereas it was only 0.07% among UK born non-white group. In the foreign-born HCV infection may be more likely to

be driven by iatrogenic exposures rather than injecting drug use compared to in the UK born.

The spontaneous clearance found in this study was 27.9% (95% CI: 20.8%-36.4%) which was similar to that found in the meta-analysis reported in chapter 2 which found clearance rates of 19.8% (95% CI: 2.6-47.5%), 27.9% (95% CI: 17.2-41.8%), 36.1% (95% CI: 23.5-50.9%), and 37.1% (95% CI: 23.7-52.8%) within 3, 6, 12, and 24 months after infection respectively. The lower estimate found might be due to it did not take into account the initial time of infection which might be underestimated the Spontaneous Viral Clearance (SVC) and introduced bias. Based on multivariate analysis on SVC, I found that injecting drugs with sharing needles reduced the odds of clearance, which might be related with re-exposure and reinfection.(41) This supports the hypothesis that PWIDs are less likely to clear virus than other groups due to repeated reinfections, reinforcing the need for harm minimisation such as needle exchange and use of oral substitution therapy.

Homeless

This study revealed that 13.2% of people who are homeless were infected with HCV. This was similar to a 2002 study conducted in Oxford (26.5%) (23) and pooled prevalence estimates from a meta-analysis of 43 studies reported in 2012 (20.3%) (22). Whereas this study recruited individuals from homeless hostels where the prevalence of PWID was 17.8%, the Oxford study recruited individuals who were not in contact with services. These individuals are likely to be even more vulnerable, supported by the fact that more than half of the participants in this study were PWIDs. This again highlights the importance of intersecting risk factors and provision of better services and access to the service in London. The study demonstrates that screening homeless people for HCV is worthwhile, but the fact that none of those identified had been treated shows the need for increased efforts to ensure treatment.

Prison

This study is likely to have underestimated the prevalence of HCV in prison populations (3.9%) because it largely excluded prisoners undergoing drug detoxification. It was low compared to a study conducted in Scottish prison where the prevalence was 19% (26) or a cross-sectional study in Dartmoor prison where the prevalence was 12.6% (42). In Scottish prison, 53% of prisoners had injected drugs, whereas in our study only 8.6% of prisoners were PWIDs.

Being homeless, PWID and imprisoned may increase vulnerability to infection. For example, Homeless Drug Users (HDUs) have been described as experiencing 'double jeopardy' given the large number of life and health issues they encounter.(43) Over half of the individuals in this study had a history of homelessness, drug use and imprisonment. A study in South Wales that recruited participants from treatment services, needle and syringe exchange services, homeless hostels and the street, showed that being homeless increased the risk of HCV about 4 fold (OR=4.41, 95% CI: 1.6-12.5).(44) Furthermore, a study conducted by Vescio, et.al. estimated risk of infection with HCV among inmates who were PWIDs was 24 times higher compared to non-PWID inmates.(45)

I found that 50.7% of HCV infected individuals had co-infection with other blood borne viruses (HBV or HIV) and this is likely to be driven by needle sharing behaviour.(46) A study performed in two Spanish prisons showed a higher prevalence of HCV-HBV co-infection (42.5%) and HIV-HBV-HCV coinfection (37.3%), and mono-infections were uncommonly found (overall 13%).(47) On the other hand, a study in Iran performed in treatment and harm reduction facilities and drug user hangout public areas showed a similar prevalence of HCV-HBV coinfection (21%), HCV-HBV-HIV coinfection (6.5%) and higher prevalence of HCV-HIV coinfection (8.7%).(48) Needle sharing is the major high risk of HCV co-infection.(46) Another study also suggested sexual activity and duration of injection were associated with HVC-HIV coinfection.(49) 6.6% of HCV patients were coinfected with LTBI only (14.0 % had triple infection of HCV and LTBI and HIV/HBV).

5.4.4 Implication for Policy and Practice

Overall, this study provides evidence of a high burden of HCV among PWIDs and homeless populations, as well as a higher prevalence among prisoners compared to the general population. The high degree of overlap of these populations indicates the need for HCV screening and treatment services to engage with services for all of all of these groups. This study's findings also support the requirement for accessible screening program, intensive case management, preventative interventions, and ongoing support so we can reach and treat many of infected individuals in vulnerable populations. In addition, with the arrival of new drugs, treatment for these high risk groups should be prioritised as they pose a higher risk of onward transmission.(18) The high level of infection highlights the importance of drug treatment and harm minimisation activities to reduce the risk of injecting in

these settings. Outreach services for vulnerable groups such as those provided by the Find & Treat tuberculosis team should also include HCV screening.(50) (51)

This study also highlighted the strong association of injecting drug behaviour and its duration with HCV infection. Thus, it is important to intervene early to minimise risk of transmission. However, given that 20% of hepatitis C infected patients in the study did not report injecting, it is reasonable to screen in these settings regardless of reported drug use. The advent of Direct Acting Antivirals (DAAs) offers new opportunities to expand treatment in these high-risk populations but integrated models of screening for blood borne viruses, managing addiction and managing infections need to be developed and evaluated. Future research should focus on how screening, treatment and prevention services can be integrated for vulnerable populations to maximise treatment access and reduce reinfection. The aspiration to eliminate HCV cannot be achieved without such integrated services. Given the high rates of infection amongst groups who are traditionally thought of hard to access this will present a major challenge to efforts to eliminate hepatitis C through broadening access to screening and treatment. In the following chapter I assess the level of treatment coverage that is likely to be needed to achieve elimination targets.

5.4.5 My Role In This Study

In this study, I conducted secondary analysis from of an existing survey. The primary data collection was conducted by TB Reach team alongside with the F&T service between May 2011 and June 2013 as part of an NIHR programme grant. From the data collected, I cleaned and extracted the data, performed the descriptive analysis, and examined the overlapping risk factors between three different vulnerable populations in London. I assessed the risk factors associated with HCV infection among recruited participants and among non-injectors. I also investigated the risk factors associated with spontaneous clearance. I was responsible for drafting the manuscript to be submitted to peer-reviewed international journal. This work has been published in the Journal of Viral Hepatitis (DN Aisyah, L Shallcross, A Hayward, RW Aldridge, S Hemming, S Yates, G Ferenando, L Possas, E Garber, JM Watson, AM Geretti, TD McHugh, M Lipman, A Story. 2018. Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, People Who Inject Drugs and Prisoners in London. Journal of Viral Hepatitis. May 2018. DOI: 10.1111/jvh.12936)

Key Points:

- This study provides evidence of a high burden of HCV among PWIDs and homeless populations, as well as a higher prevalence among prisoners compared to the general population
- More than half of HCV infected individuals had intersecting risk factors of homelessness, prison and drug use, which implies those group are important as population target for HCV prevention and screening program.
- I also found strong association of injecting drug behaviours and its duration with HCV infection which underlined the important of early intervention to minimise the risk of transmission.
- Alcohol problems and age ≥ 30 years old increased the risk of HCV infection among individuals who reported not injecting drugs; whereas illicit drug use with needle sharing reduced the likelihood of achieving spontaneous clearance.
- This study also supports the requirement for accessible screening program, intensive case management, preventative interventions, and ongoing support so we can reach and treat many of infected individuals in vulnerable population.

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6. Modelling Hepatitis C Direct Acting Antiviral (DAA) Treatment Scale Up among People Who Inject Drugs in London: Working towards WHO Incidence Elimination Target by 2030

Chapter's Aim:
The aim of this chapter is to estimate the Direct Acting Antiviral (DAA) treatment needed among People Who Inject Drugs (PWID) to achieve WHO's HCV incidence elimination target by 2030.

6.1 INTRODUCTION

In most developed countries, people who inject drugs (PWID) are the main source of hepatitis C virus (HCV) transmission, accounting for 50-90% of all HCV infections in several European countries, the United States, and Australia.(1) Based on a systematic review in 2011, more than 50% of all ever-infected HCV cases in developed countries occur in patients with a history of injection drug use.(2, 3) Worldwide estimates of the seroprevalence of anti-HCV (a marker of ever infection) amongst drug-users vary widely with an average of 60-80% and the lowest seroprevalence was 9.8% in Paraguay to 97.4% in Mexico (4). PWID have been identified as the key population with primary mode of transmission for hepatitis C in the developed nations.(5)

In the UK, an estimated 92.5% of HCV infections were caused by injecting drug use.(3) Based on the Unlinked Anonymous Survey (UAS) in 2017, the seroprevalence of hepatitis C infection among PWID in London is 63%.(6) UAS is an annual cross-sectional survey coordinated by Public Health England, with support from Public Health Wales and Public Health Agency Northern Ireland. This survey is targeted at people who inject psychoactive drugs and they were recruited from drugs and alcohol treatment services across UK.

New direct-acting antiviral agents (DAA) represent a major advance in hepatitis C treatment, with cure rates of more than 90%, shorter duration of treatment, less toxicity and fewer side effects compared to interferon based therapy.(7-9) There is major potential to substantially reduce the future burden of HCV cases if treatment can be targeted effectively to high-risk individuals. This treatment revolution offers

an opportunity to eliminate hepatitis C in the future.(10) In 2016, the World Health Organisation (WHO) published hepatitis B and C elimination as public health targets, including a 90% reduction in new chronic infections and 65% reduction in mortality by 2030 compared with a scenario in which interventions would continue at the 2016 level.(11)

Mathematical epidemic modelling plays a large and important role in the planning of public health interventions. The application of modelling of infectious diseases has been widely used to study the mechanisms by which diseases spread (12, 13), epidemic surveillance (14-17), to monitor, identify, and forecast disease outbreaks (17-20), and to evaluate strategies to control an epidemic or evaluate public health programmes/interventions (21-24). In relation to hepatitis C prevention, mathematical modelling has been largely utilised to evaluate the potential effectiveness of HCV treatment as prevention among PWID (10, 25-31) as well as evaluating the effectiveness of harm reduction among this high risk group (29, 32-34). In this chapter, I model the impact of DAA therapy to reduce HCV incidence among PWID in London. The aim of this study is to assess the DAA treatment scale up needed among PWID in London to achieve the WHO incidence elimination target by 2030.

6.2 METHODS

6.2.1 MODEL STRUCTURE

We used a dynamic deterministic compartmental model previously developed (30) describing HCV transmission among PWID. A dynamic model was used which allowed us to include the indirect effects of interventions such as treatment, by assuming that the force of infection is proportional to the prevalence of HCV infection among PWID, not the number of infected individuals per se, especially if the population size changes over time. The model is deterministic, in that it produces the same results if the initial conditions and parametrization remain unchanged as the model does not use any degree of randomness or uncertainty in its computation.(35)

In this study, the model was divided into 5 infection states, which are (S) susceptible, (A) acute HCV infection, (C) chronic HCV infection, (T) individuals on DAA treatment, and (TF) treatment failure (Figure 1). In this model, new people who inject drugs continually enter the model at a constant rate (ϕ) as susceptible. Susceptible PWID can become infected with hepatitis C virus and become acutely

infected (λ). This study uses a dynamic model, thus the risk of HCV infection among PWID is proportional to hepatitis prevalence and changes over time. A proportion of the acutely infected individuals may clear the virus spontaneously without treatment (σ) after some duration of time (ξ). The remaining proportion who do not experience spontaneous clearance (1- σ) will become chronically infected. A fixed proportion (Δ) of PWID with chronic infection starts the DAA treatment and will exit after the DAA treatment duration (ω). A proportion of treated individuals will achieve Sustained Virologic Response (SVR) and return to the susceptible compartment (θ), and the remainder will experience treatment failure, (1-θ). PWID exit all of the compartments caused by permanent cessation of drugs (d_1) or death due to drug or non-drug-related cause (d_2).

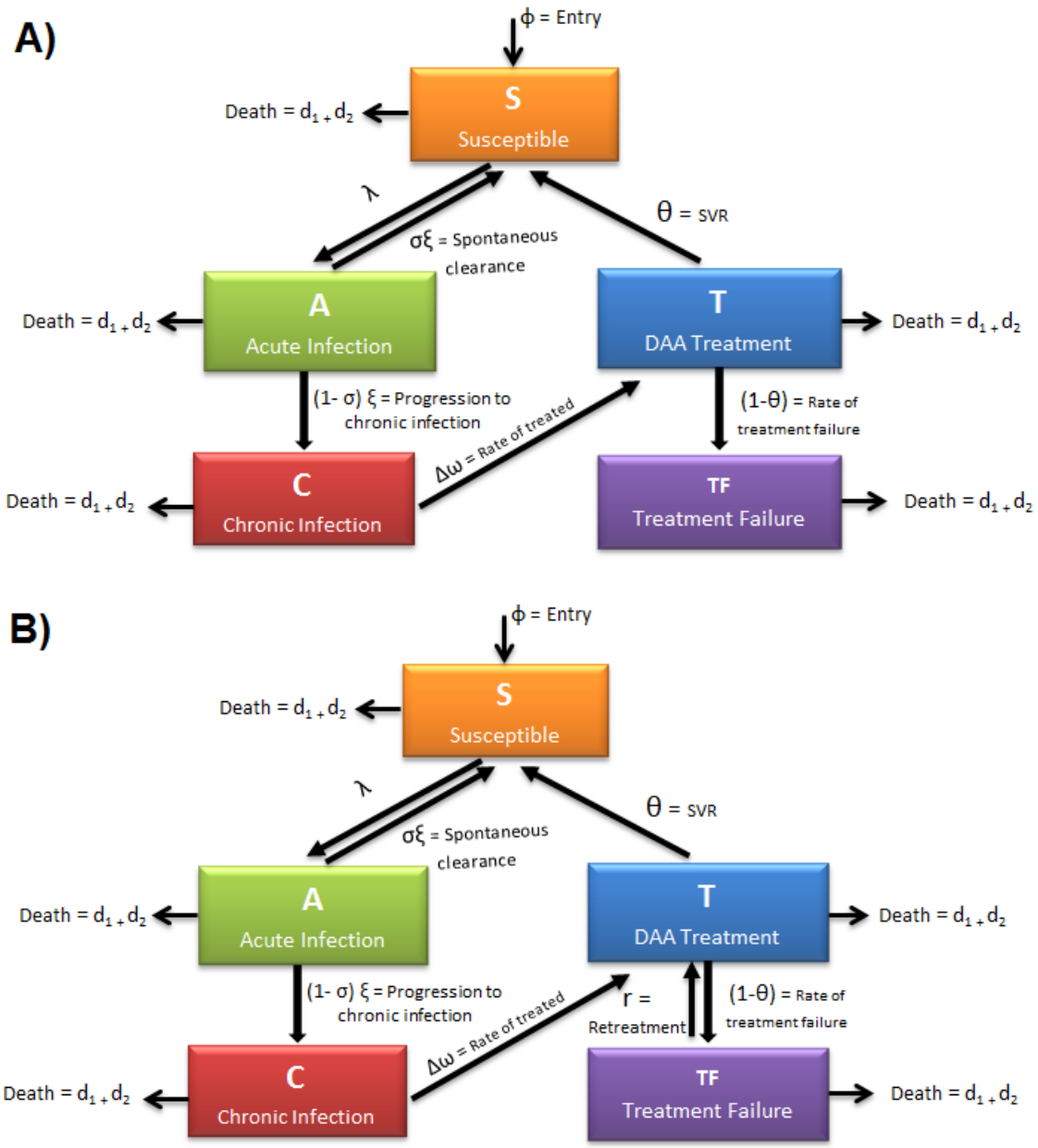

Figure 6.24 Model structure showing Hepatitis C transmission (A) without re-treatment and (B) allowing re-treatment. Five infection states included: (S) susceptible, (A) acute HCV infection, (C) chronic HCV infection, (T) individuals on DAA treatment, and (TF) treatment failure

In our model, DAA treatments were not given to individuals with acute infection because PWID are usually diagnosed with HCV during chronic infection and acute infections are usually asymptomatic (36, 37). Our initial model assumed that those who fail treatment could not be treated because of potential resistance and that those who fail treatment are not retreated. We also used the assumption that those who are on treatment are not infectious. The corresponding equation for the above structure is:

$$1)\ \frac{dS}{dt} = \phi - \lambda S(A+C+TF)/(S+A+C+T+TF) + \sigma\xi A + \theta\omega T - (d1 + d2)S$$

$$2)\ \frac{dA}{dt} = \lambda S(A+C+TF)/(S+A+C+T+TF) - (1-\sigma)\xi A - \sigma\xi A - (d1 + d2)A$$

$$3)\ \frac{dC}{dt} = (1-\sigma)\xi A - \Delta C - (d1 + d2)C$$

$$4)\ \frac{dT}{dt} = \Delta C - \theta\omega T - (1-\theta)\omega T - (d1 + d2)T$$

$$5)\ \frac{dTF}{dt} = (1-\theta)\omega T - (d1 + d2)TF$$

We also explore a second model allowing re-treatment (at a rate r) of treatment failures, because the effectiveness of new treatments for retreatment is promising.(38-40) (see figure 1B):

$$1)\ \frac{dS}{dt} = \phi - \lambda S(A+C+TF)/(S+A+C+T+TF) + \sigma\xi A + \theta\omega T - (d1 + d2)S$$

$$2)\ \frac{dA}{dt} = \lambda S(A+C+TF)/(S+A+C+T+TF) - (1-\sigma)\xi A - \sigma\xi A - (d1 + d2)A$$

$$3)\ \frac{dC}{dt} = (1-\sigma)\xi A - \Delta C - (d1 + d2)C$$

$$4)\ \frac{dT}{dt} = \Delta C - \theta\omega T - (1-\theta)\omega T - (d1 + d2)T + r*TF$$

$$5)\ \frac{dTF}{dt} = (1-\theta)\omega T - (d1 + d2)TF - r*TF$$

6.2.2 MODEL PARAMETERISATION

The model was parameterised to London, United Kingdom, using data from several sources that are shown in Table 1. A previous modelling study conducted at multiple sites in the UK showed low treatment rates at baseline (28), for simplicity, given the extremely low proportion of drug users who were treated prior to 2017 we used the assumption of no treatment prior to 2017. The PWID population size was estimated by applying the PWID prevalence estimation by Hickman et.al for London, UK (41), 1.2% (95% CI: 1%-1.6%) and multiplying by the number of population aged 15-44 years old in London in 2017 based on data from Greater London Authority (GLA). Based on Unlinked Anonymous Survey data, the HCV seroprevalence in PWID in London was 63% in 2016.(6) By subtracting the proportion of those who were expected to spontaneously clear the virus (36% - see chapter 2), we estimated a chronic HCV prevalence of 40% among PWID (63%-(63*26%)=40%). We assumed the proportion of HCV infected individuals who achieved sustained virological response using DAA treatment used in this model was 90%.(42) We assume the entry rate (number of people who were started injecting) represented by ϕ, is equal with the number of people who exit all of the compartment either due to permanent cessation of drug use (d1) or death due to drug or non-drug related cause (d2).

Table 6.10 Model Parameters and Sources

Parameters	Symbol	Value	Units	References
Entry rate	ϕ	d1+d2		We assumed the number of people who were started injecting equals with number of people who exit all of the compartment either due to permanent cessation of drug use (d1) or death due to drug or non-drug related cause (d2)
HCV chronic prevalence among PWID in 2016	φ	40%		Using seroprevalence number from Unlinked Anonymous Survey 2017 (6), we deducted with 36% spontaneous clearance (63%-(63*36%)) leading to the HCV chronic prevalence of 40%
PWID population size 2017		49,771		PWID prevalence by Hickman et. al (2004) (41) multiplied by population size of adults in

Parameters	Symbol	Value	Units	References
				London (43)
Death due to drug or non-drug-related cause	d2	0.0075	Per year	Hickman, et.al (2009) (44)
Proportion of spontaneously clear in acute stage	σ	36%	%	Aisyah, et.al (2018) (45) – based on result from chapter 2
Duration of clearance	12/ξ	12	months	Aisyah, et.al (2018) (45)
Duration of injecting until final cessation	1/d1	11	years	Sweeting et.al (46)
SVR	θ	90	%	Dore et.al (42)
Duration of treatment	52/ω	12	weeks	Dore et.al (42)

6.2.3 MODEL CALIBRATION

The model was calibrated at equilibrium of 40% chronic prevalence among PWID through varying the parameter lambda (λ), which resulted in a value of 0.244. I assumed that the number of PWID population was constant, thus the number of people who enter the compartments (people who start injecting) was equal with the number who exit the compartments. The chronic prevalence was assumed based on steady seroprevalence in the UK and I assume there was no treatment at baseline.

6.2.4 MODEL SCENARIOS

First, I ran the model with no treatment, allowing us to see what the HCV prevalence and incidence is in the future when there was no treatment intervention Then, I ran the model with treatment scale up from 10%, 20%, 30%, 40%, and 50% of HCV infected PWID per year started from 2017 until 2030. I also examined what level of treatment is needed to achieve the WHO incidence elimination target of 90% reduction from 2017 to 2030.

6.2.5 MODEL OUTPUTS

I calculated the prevalence (A) by dividing the number of those who are infected with hepatitis C (including those who were in acute, chronic, and treatment failure) by the number of total population of PWID. The incidence (B) was assessed by calculating the instantaneous incidence, which is done by dividing the number of incident infections at a given time point by the number at risk (susceptible population) at that time point. To obtain the relative difference (%) of incidence or

chronic prevalence, I subtracted the incidence/prevalence in 2030 by incidence/prevalence in 2017 then divided by incidence/prevalence in 2017.

$$(A)\ PREV = (A+C+TF)/(S+A+C+T+TF)$$
$$(B)\ INC = (lambda*S*(A+C+TF)/(S+A+C+T+TF))/S$$

6.2.6 Sensitivity Analysis

To evaluate the impact of the model assumption on treatment rate needed to achieve the WHO incidence elimination target by 2030, multiple univariate sensitivity analyses were performed to assess the impact of: shorter or longer duration of injecting, lower or higher proportion of spontaneous clearance, and lower or higher death rate due to drug or non-drug-related causes. The duration of injecting was assessed for 8 or 14 years versus 11 years at baseline, the spontaneous clearance was modelled for 26% or 46% versus 36% at baseline, and death rate assessed for 0.0025 or 0.0125 versus 0.0075 at baseline.

6.3 Results

6.3.1 Base Case with No Re-Treatment of Treatment Failure

Our model predicted that without treatment, the incidence of hepatitis C in PWIDs in London is 9.76% in 2017. This translates to 2654 incident infections in 2017. The model is well calibrated to the estimated chronic prevalence of 40% in 2017 (figure 7.2).

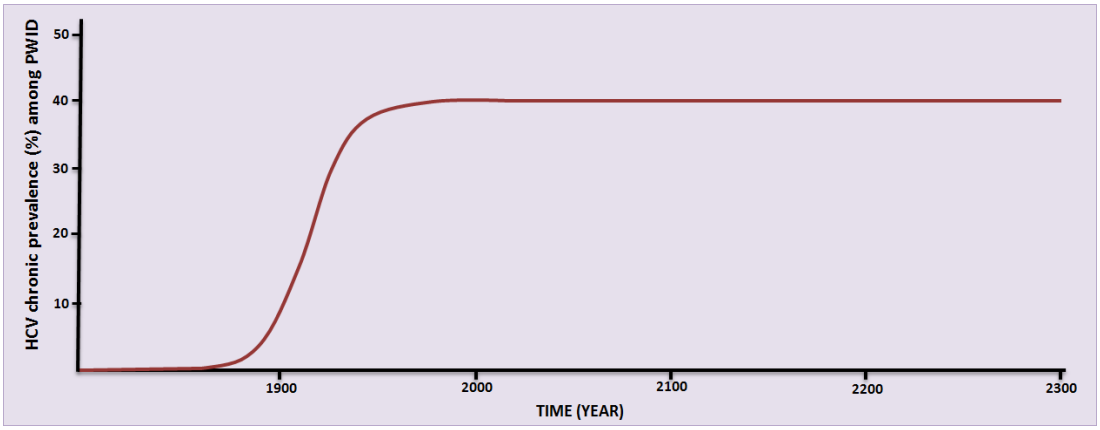

Figure 6.25 HCV Chronic Prevalence of 40% at Equilibrium – The model was calibrated at equilibrium of 40% chronic prevalence among PWID through varying the parameter lambda (λ), which resulted in a value of 0.244

By providing 10%, 20%, 30%, 40%, and 50% of chronically infected PWID with DAA treatment annually started from 2017, it could lead to a 50.2% (95% CI: 20%-80.3%), 73.4% (95% CI: 37.8%-93.7%), 84.4% (95% CI: 48%-98.1%), 88.5% (95%

CI: 52.1%-99.2%), and 90.8% (95% CI: 54.4%-99.6%) relative reduction in incidence by 2030, respectively. This would translate to relative reductions in chronic prevalence of 50.1% (95% CI: 34.2%-66.1), 73.4% (95% CI: 56.8%-85.6%), 84.4% (95% CI: 68.8%-93.4%), 88.5% (95% CI: 73.6%-95.9%), and 90.8% (95% CI: 76.4%-97.2%).

The WHO elimination target aims to have 90% reduction of incidence by 2030, corresponding to an incidence target of 0.98%. Based on the model, to achieve this aim, we need to treat at least 46% of chronically infected PWID with DAA treatment annually from 2017 until 2030 (incidence reduction of 90.1% (95% CI: 53.7%-99.5%).

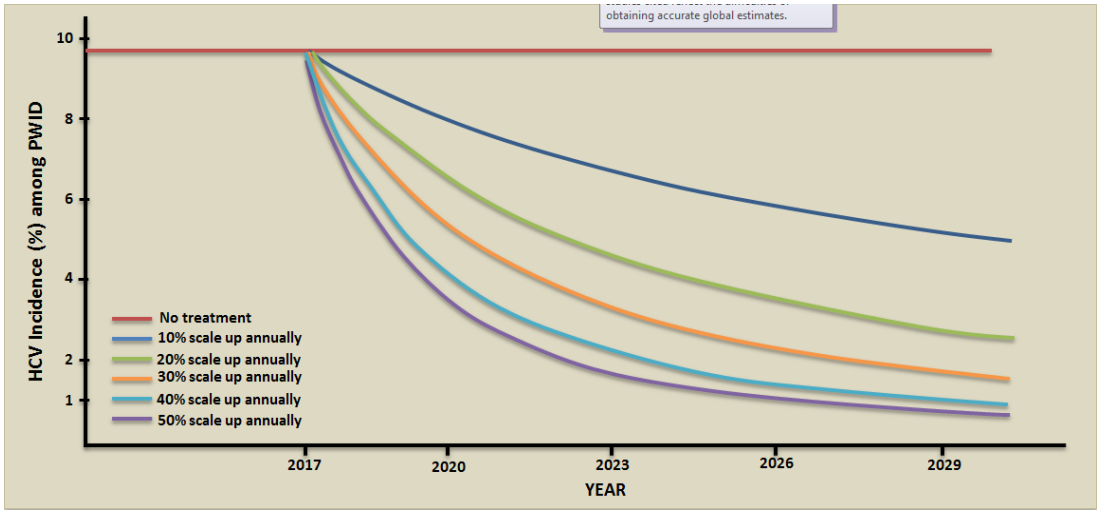

Figure 6.26 Model predictions of absolute HCV Incidence among PWID in London with no treatment, 10%, 20%, 30%, 40%, and 50% annual treatment rates among HCV-infected PWID annually from 2017

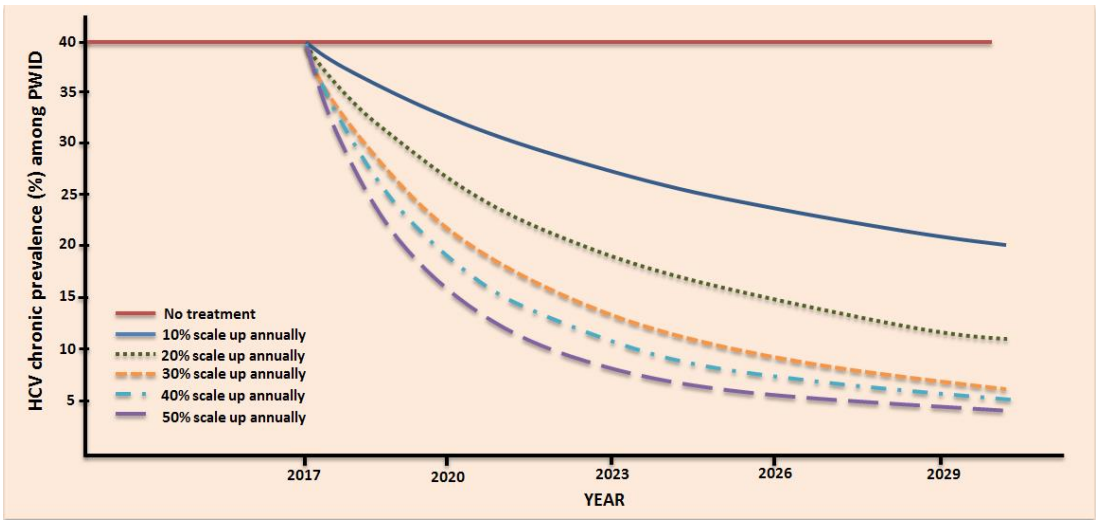

Figure 6.27 Absolute Chronic Prevalence among PWID in London with no treatment, 10%, 20%, 30%, 40%, and 50% annual treatment rate among HCV-infected PWID annually since 2017

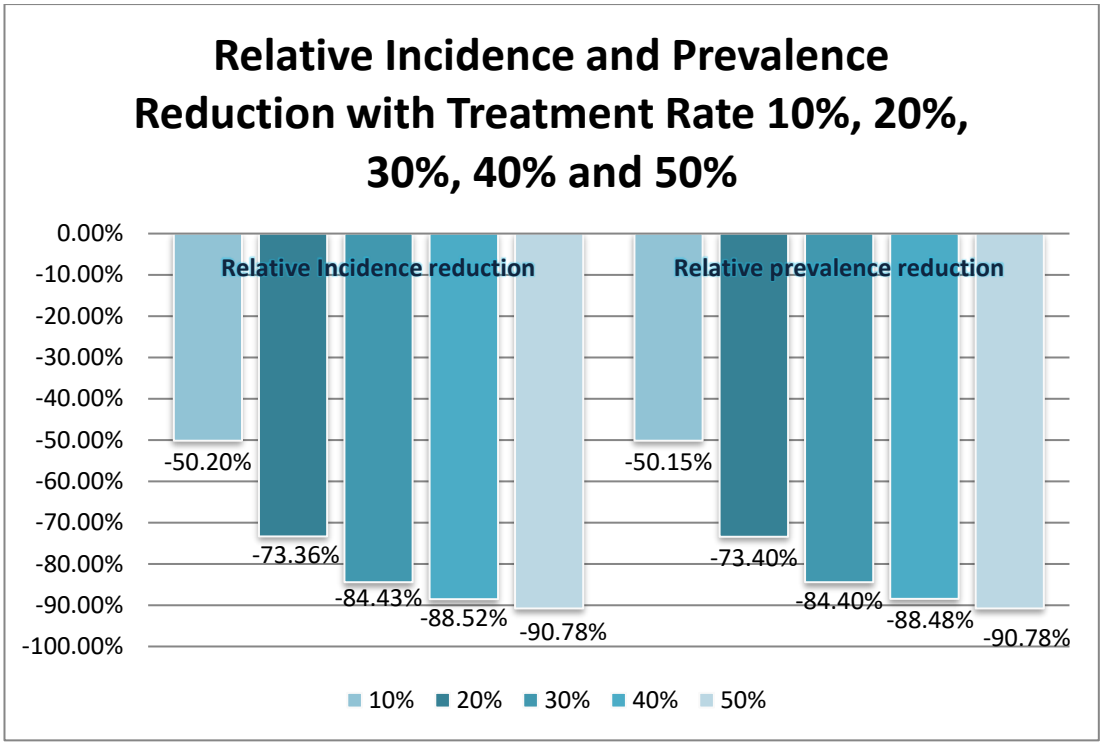

Figure 6.28 Relative Incidence and Prevalence Reduction Achieved by 2030 with Treatment Rate of 10%, 20%, 30%, 40%, and 50%

6.3.2 *Sensitivity Analysis of No Re-Treatment of Treatment Failures Model*

6.3.2.1 *Using Higher/Lower SVR Assumption*

The SVR used for our base case was 90%. In some studies, higher SVR was found among HCV patients.(9, 47) Thus, we also want to assess the treatment scale-up needed if the SVR assumption was higher than the base case. In figure 7.6 we can see that using assumption of DAA treatment efficacy for 95%, a 33% annual treatment of chronically infected PWID is required until 2030 to reach the WHO target. On the other hand, when we assume the SVR of DAAs is 100%, only 27% of chronically infected PWID population need be treated each year to reach the target. However, if we assumed the SVR was lower (e.g. DAA efficacy is 85% when rolled out to increasingly socially complex groups), it will require treatment coverage of 93% infected PWID annually between 2017 and 2030.

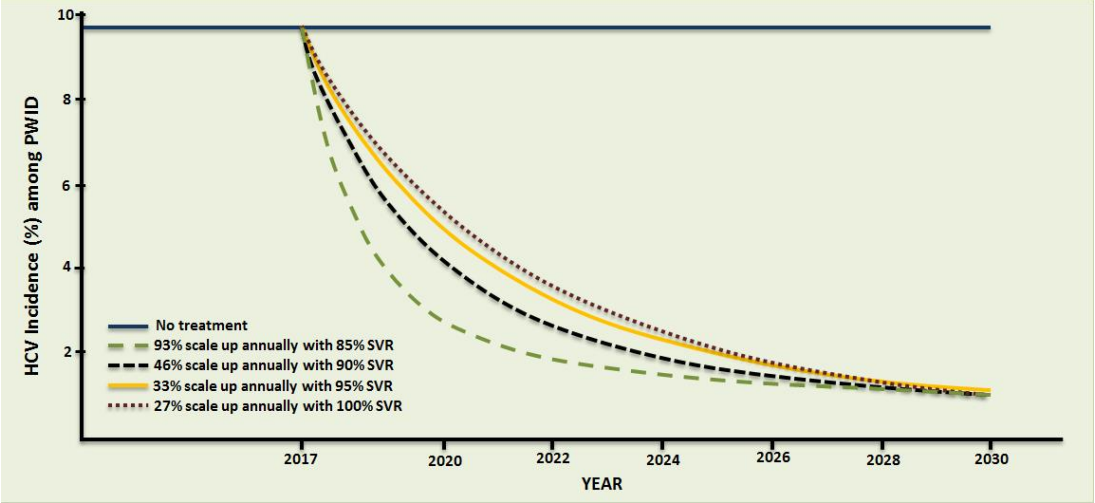

Figure 6.29 Annual Treatment Scale-Up Required to Reduce Incidence within 13 Years (by 2030) when the SVR is 85%, 90%, 95% and 100%.

Table 6.11 incidence in 2030 when the SVR was 85%, 90%, 95%, 100% (the yellow block showed treatment rate needed to achieve WHO incidence elimination target by 2030).

Annual Treatment Rate	Incidence in 2030 when SVR 85%	Incidence in 2030 when SVR 90%	Incidence in 2030 when SVR 95%	Incidence in 2030 when SVR 100%
0%	9.76	9.76	9.76	9.76
10%	5.13	4.86	4.58	4.29
20%	2.99	2.60	2.18	1.77
27%	2.23	1.81	1.37	**0.97***
30%	2.02	1.52	1.14	
33%	1.84	1.41	**0.97***	
40%	1.55	1.12		
46%	1.39	**0.97***		
93%	**0.97***			

**Yellow shading highlights threshold level for achieving 90% incidence reduction target*

6.3.2.2 *Using Higher/Lower Duration of Injecting, Death Rate, and Spontaneous Clearance Assumption*

Several sensitivity analyses were performed to examine changes in the required treatment rate to achieve the WHO incidence target when duration of injecting, death rate, and proportion of spontaneous clearance getting lower or higher. This sensitivity analysis showed that duration of injecting had the highest impact on treatment scale up needed to achieve the elimination target. With scenario of 3 years shorter duration of injecting compared to base case scenario, the treatment scale up required decreased to 19%. On the other hand, when the duration of

injecting was 3 years longer than the base case scenario, the treatment scale up required increased to 100% (see table 7.3).

Table 6.12 Impact of various sensitivity analyses on the treatment rate needed to achieve the WHO incidence elimination target by 2030 using the retreatment model.

Variable		Treatment rate needed to achieve WHO incidence elimination target
Duration of injecting	8 years	19%
	14 years	100%
Death rate	25/10000	53%
	125/10000	41%
Spontaneous clearance	26%	65%
	46%	32%
SVR	85%	93%
	95%	33%
	100%	27%

When the death rate was 0.0025 and 0.0125 (compared to the base-case of 0.0075), the treatment scale up increased by a relative 15.2% and decreased by a relative 10.9% respectively (i.e. the higher the death rate the lower the proportion who need to be treated to eliminate). I also varied the proportion of spontaneous clearance among PWID. With the assumption of 10% lower spontaneous clearance from 36% in the base case, the treatment scale up needed was increased by 41.3%. Using 10% higher assumption of spontaneous clearance, the treatment scale up needed was decreased by a relative 30.4% (see figure 7.7). The treatment scale up needed was decreased by 28.3% and 41.3% when we used higher assumption of SVR 95% and 100%. However, the treatment scale up needed was significantly increased by 130.4% when we used lower assumption of SVR 85%.

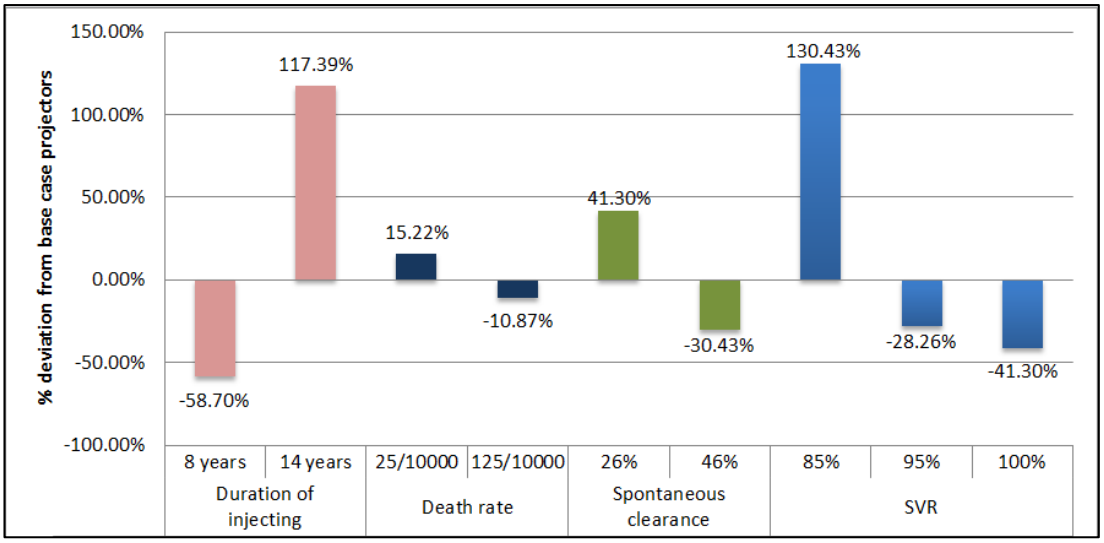

Figure 6.30 Sensitivity analyses for duration of injecting, death rate, spontaneous clearance and sustained virological response. The figure shows percent changes of treatment threshold needed to achieve WHO incidence elimination target from base case scenario based on sensitivity analyses with no-retreatment of treatment failure model

6.3.3 Results For Model Incorporating Re-Treatment for Treatment Failures

We also assessed the potential impact of DAA re-treatment of treatment failures. When we allow re-treatment at an equal rate as the treatment rate, a 29.5% and 28% per year treatment rate among chronically infected PWID is required to achieve WHO elimination target with assumption of 90% SVR and 95% SVR, respectively. Furthermore, using the assumption of 85% DAA's treatment efficacy, and assuming retreatment achieving the WHO elimination target requires 31.3% treatment rate annually among chronically infected PWID in London from 2017. The estimate incidence reduction based on treatment and re-treatment strategy can be seen below in Table 7.4.

Table 6.13 Absolute Incidence reduction at 2030 by conducting treatment and re-treatment strategy (with the similar rate with treatment rate) when the SVR was 90% and 95%

SVR	Treatment Rate	Absolute incidence reduction (%) without re-treatment of TF	Absolute incidence reduction (%) with re-treatment of TF	Relative incidence reduction (%) with re-treatment of TF
85%	0%	9.76	9.76	
	10%	5.15	4.86	-50.20%
	20%	3.00	2.31	-76.33%
	30%	2.03	1.07	-89.04%
	31%	1.97	0.99	-89.86%
	31.3%	1.95	0.97	-90.06%
	32%	1.91	0.92	-90.57%
90%	0%	9.76	9.76	
	10%	4.87	4.66	-52.25%
	20%	2.60	2.11	-78.38%
	25%	1.99	1.41	-85.55%
	29%	1.65	1.01	-89.65%
	29.50%	1.62	0.974	-90.02%
	30%	1.59	0.94	-90.37%
95%	0%	9.76	9.76	
	10%	4.58	4.47	-54.20%
	20%	2.18	1.93	-80.23%
	25%	1.55	1.26	-87.09%
	28%	1.29	0.97	-90.06%

**Yellow shading highlights threshold level for achieving 90% reduction target*

6.3.4 Sensitivity Analysis for Re-Treatment Failures Model

Sensitivity analyses were performed to examine changes in the required treatment rate to achieve the WHO incidence target using lower or higher duration of injecting, death rate, and proportion of spontaneous clearance. Similar to the sensitivity analysis performed for no treatment model, this sensitivity analysis showed that duration of injecting had the highest impact on treatment scale up needed to achieve the elimination target. With scenario of 3 years shorter duration of injecting compared to base case scenario, the treatment scale up required decreased to 17%. On the other hand, when the duration of injecting was 3 years longer than the base case scenario, the treatment scale up required increased to 34.2% (see table 7.5).

Table 6.14 Impact of various sensitivity analyses on the treatment rate needed to achieve the WHO incidence elimination target by 2030 using the retreatment model.

Variable		Treatment rate needed to achieve WHO incidence elimination target
Duration of injecting	8 years	17%
	14 years	34.2%
Death rate	25/10000	30.8%
	125/10000	28.2%
Spontaneous clearance	16%	33.8%
	36%	24.1%
SVR	85%	31.3%
	95%	28.0%

When the death rate was 0.0025 and 0.0125 (compared to the base-case of 0.0075), the treatment scale up increased by a relative 4.4% and decreased by a relative 4.4% respectively. Furthermore, with the assumption of 10% lower spontaneous clearance from 36% in the base case, the treatment scale up needed was increased by as relative 14.6%. Using 10% higher assumption of spontaneous clearance, the treatment scale up needed was decreased by a relative 18.3% (see figure 4). The treatment scale up needed was decreased by a relative 5.1% and increased by a relative 6.1% when we used higher assumption of SVR 95% and 85%.

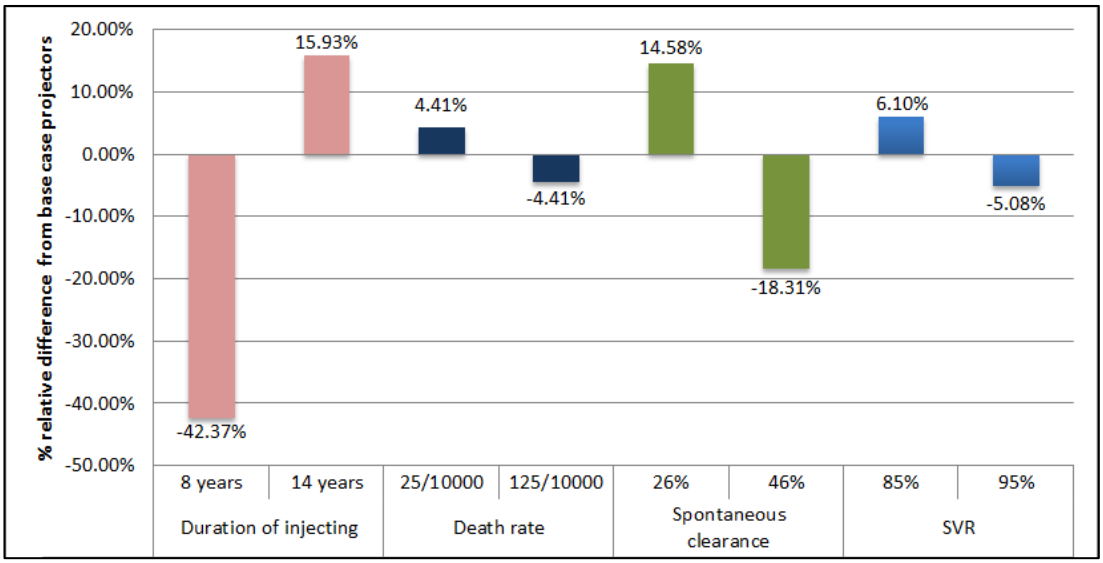

Figure 6.31 Percent changes of treatment threshold needed to achieve WHO incidence elimination target from base case scenario based on sensitivity analysis with retreatment of treatment failure model

6.4 DISCUSSION

6.4.1 SUMMARY OF FINDINGS

This study examined the DAA treatment scale up that would be needed among chronically infected PWID in London to achieve WHO's elimination target by 2030. In the base case scenario to achieve 90% reduction of HCV new cases in London among chronically infected PWID by 2030, a 46% treatment coverage would be needed annually starting from 2017 if treatment failures are not retreated. If treatment failures are retreated, then only an annual treatment rate of 29.5% among chronically infected PWID will be needed to achieve the target. We assumed a 90% SVR at baseline, but our results show fewer treatments are required if SVR is higher. Conversely, substantially higher numbers of treatment courses are required if SVR is lower. For example, if SVR is 95%, then only 33% of chronically infected PWID need to be treated per year if there are no retreatments (compared to 46%). But if SVR is 85% then 93% coverage is needed. Retreatment strategies substantially lessened the dependence on high SVR.

Our sensitivity analysis underlined the importance of duration of injecting as it has the highest impact on treatment scale up needed. This highlights a clear public health message for reinforcing the importance of screening drug users and interventions to stop injecting behavior as a tool to prevent HCV transmission. The model demonstrates that treatment coverage will need to be considerably scaled up to achieve elimination targets. The model also demonstrates the critical importance of ensuring high SVR (which may require adherence support as treatment is rolled out to increasingly socially complex cases). The model also shows the importance of retreating treatment failures. Finally the model shows the importance of effective drug treatment services to reduce injecting duration.

6.4.2 STUDY STRENGTHS AND LIMITATIONS

The strength of our study is that we adapted an existing, HCV transmission model for PWID and parameterised it specifically for London. We examined different scenarios of DAA's treatment efficacy using 85%, 95% and 100% SVR. We also performed sensitivity analysis by varying the duration of injecting drug use, death rate, and spontaneous clearance to see how these impacts on the number of DAA treatment courses required.

As the projections are based on theoretical assumption based on previous published papers, the uncertainty of parameters was one of the limitations of this

study. Studies of DAA treatment outcomes among PWID are very limited. In a study conducted by Dore et.al SVR, among individuals taking opioid therapy who we treated with elbasvir–grazoprevir (42) was estimated to be 91.5%, which is close to the parameter used in the base case. I also did not take into account any co-infection, HCV infection stages (fibrosis, cirrhosis, decompensated cirrhosis), and different genotypes as those factors might affect the results. For example, the effectiveness of treatment might be reduced in those who were cirrhotic or had decompensated cirrhosis (48-52) thus increasing the treatment rate needed to achieve WHO incidence elimination target. However, most PWID will be young with less advanced of liver disease and will not likely fall into these categories because it takes many decades to progress and PWID only inject on average for 10 years.

From the literature search, there are several factors that may influence the ability of infected patients to be cured such as genotype 1 or 3 infection (47, 53, 54), cirrhosis or advanced fibrosis (55-57), and/or prior non-responders to pegylated IFN–based treatment (58, 59). I also did not incorporate harm reduction interventions into this model which have been shown to lower the possibility of reinfection and transmission (60) thus decreasing the treatment rate needed to achieve WHO's incidence elimination target.

6.4.3 DISCUSSION OF THE RESULTS

Our model predicted that without treatment, the incidence of hepatitis C among PWID in London is 9.76% in 2017. This prediction is lower compared to a study conducted in 2004 where the incidence of antibody to hepatitis C virus was 41.8 cases per 100 person years among PWID in London (61). On the other hand, our estimate is higher compared to a study in 2009 where the incidence of primary HCV infection varied from 2.7–3.2 per 100 person years among people who had ever injected drugs in the UK (62). The study reported in chapter 5 found that 24.9% of participants recruited in drug treatment services were infected with HCV. The varied estimates might be affected by the outcome used (HCV antibody positive, chronic HCV), different study populations, regional variation in HCV prevalence, categorisation of drug injecting status (currently injecting, ex-injectors, ever injected) and how this information is elicited, and year of the study.

The result from the analysis showed that 46% treatment coverage would be needed annually to achieve the WHO target it treatment started in 2017 (the coverage needed was lower, 29.5%, in the scenario of retreatment of treatment failures). This target seems quite ambitious considering the historically limited budget available for

DAA treatment. Using population data in 2005, there were approximately 150,000 individuals with chronic hepatitis C infection in England, 66,000 (44%) of whom were attributable to current PWID and 64,500 (43%) attributable to ex-PWID (63). Considering this huge number, treating 10,000 patients with DAA's in 2016-2017 (only 8%) was far from sufficient. The reasons for this include, the large numbers of potential patients and the very high aggregate cost of the treatments involved.(64) If we assume that 46% treatment coverage would be needed for current PWID chronically infected with HCV, we would need to provide DAA therapy for approximately 30,360 PWID in 2017, almost 6 times higher than budgeted for in 2016-17. Under a strategy of re-treatment for treatment failures, 29.5% DAA treatment would be required for 19,470 current PWID chronically infected with HCV in 2017. Achieving WHO's incidence elimination target would require significant budget and strong commitment from local government. We also need to consider the difficulties of accessing this population and how to increase their engagement with HCV screening and treatment services and with drug treatment and harm minimization services.

Other modelling studies also have been done to evaluate the effectiveness of HCV treatment in European countries (26), United Kingdom (27, 28), and other cities in Australia (Melbourne) and Canada (Vancouver) (65). A study published in 2015 assessed the impact of treatment among PWID in seven cities in the UK including Bristol, East London, Manchester, Nottingham, Plymouth, Dundee and North Wales.(28) Based on the study (28), a 10% absolute reduction in chronic HCV prevalence could be achieved in East London (22–36%) within 10 years, if genotype 1 patients were treated with DAAs (with genotype 2/3 remaining on pegIFN/RBV with the lower ITT SVR). Furthermore, the model predicted a 15% absolute reduction in chronic prevalence within 10 years in East London (13–20%) if treatment rates were scaled up to 26 per 1000 PWID annually and DAAs were made available for all genotypes in early 2016. These studies have evaluated the impact of DAA treatment among PWID population, however, to the best of my knowledge, this study is the first to model the treatment rate that would be required in order to achieve WHO's incidence elimination target by 2030 in a UK setting.

In Europe (Czech Republic, Slovenia, and Amsterdam) modelling studies based on current usage of direct-acting antivirals (DAAs) have estimated reductions in chronic HCV prevalence that range from 38–63% in 10 years. Doubling the HCV treatment rates has been estimated to reduce prevalence by 12–24% (Belgium/Denmark/Hamburg/Norway/Scotland), but was predicted to have minimal

impact in Sweden and Finland due to its high HCV chronic prevalence.(26) In the USA, a modelling study estimated 120 per 1000 PWID would need to be treated annually to eliminate HCV within 10 years when baseline HCV prevalence is 60% or lower.(66) We can't directly compare our results with this study because the function of their treatment model was based as a fixed number per year instead of treating a proportion of chronic infections. One study in Australia found that DAA treatment among PWID was cost effective, with a discounted average gain of 2.98 (95% confidence interval 2.88–5.22) QALYs[2] per person for an additional cost of $15,132 ($11,246–18,922), giving an ICER[3] of $5078 ($2847–5295) per QALY gained.(68)[4]

While HCV treatment is one of public health strategies to control hepatitis C among PWID, harm reductions intervention also play an important role for intervention and prevention of HCV in this high-risk group population. A modelling study predicted that modest reductions in syringe sharing frequency (<25%) will reduce the HCV seroprevalence in newly initiated PWIDs (injecting less than four years) but much larger and sustained reductions (>50%) are required to reduce the HCV seroprevalence in long-term PWID (injecting more than 8 years) in London.(70) Another study assessing the combination of DAA treatment and harm reductions found that scaling up OST (Opioid Substitution Therapy) and high-coverage needle and syringe programs (HCNSP) by 40% would halve prevalence for 20%, 40%, or 60% baseline chronic HCV prevalences by treating 10, 23, or 42 per 1000 PWID annually over 10 years, respectively.(29) A modelling study in Europe also found that by scaling-up OST and needle and syringe programmes (NSP) to 80% coverage, current treatment rates using DAAs could achieve observable reductions in HCV prevalence (18–79%) in all study sites.(26) Moreover, a modelling study in the United Kingdom estimated that without current coverage levels of OST and 100% NSP the chronic HCV prevalence could be 25% higher than the existing

[2] QALY (Quality Adjusted Life-Year) is a measure of the state of health of a person or group in which the benefits, in terms of length of life, are adjusted to reflect the quality of life.(NICE) It is developed as a measure of health effectiveness for cost-effectiveness analysis, a method intended to aid decision-makers charged with allocating scarce resources across competing health-care programs. 67. NICE. Glossary: National Institute for Health and Care Excellence 2018 [Available from: https://www.nice.org.uk/glossary?letter=q.

[3] ICER (Incremental Cost-Effectiveness Ratio) is a summary measure representing the economic value of an intervention, compared with an alternative (comparator). An ICER is calculated by dividing the difference in total costs (incremental cost) by the difference in the chosen measure of health outcome or effect (incremental effect) to provide a ratio of 'extra cost per extra unit of health effect' – for the more expensive therapy vs the alternative.

[4] In Australia, the threshold base-case reference for ICER was estimated at AUD28,033 per QALY gained. 69. Edney LC, Afzali HHA, Cheng TC, Karnon J. Estimating the Reference Incremental Cost-Effectiveness Ratio for the Australian Health System. PharmacoEconomics. 2018;36(2):239-52.

40%. However, increasing OST and 100% NSP coverage further is unlikely to reduce chronic prevalence to less than 30% over 10 years unless coverage becomes ≥80%.(34) These studies show the critical importance of scaling up HCV treatment in PWID and scaling up access to drug treatment services and needle exchange. However, elimination strategies appear to emphasise HCV treatment above measures to address injecting risk. It should also be noted that interventions to address injecting will have numerous additional health benefits beyond HCV reduction (e.g. reductions in other Blood Borne Virus - BBVs, skin and soft tissue infections, osteomyelitis, endocarditis, overdose etc.).

The model in this study showed that scaling up treatment could theoretically reduce the number of HCV new cases considerably, but there are considerable logistical challenges to achieving this aim. For example, the first challenge is identifying high-risk individuals which would require an increase in the uptake of testing among high risk individuals by providing screening and testing that is accessible for vulnerable groups. The uptake of hepatitis C testing among PWID in England and Wales was poor with less than two-fifths having ever been tested, and only half of those tested having been tested recently.(71) In chapter five I show the importance of screening in venues such as drug treatment services, prisons and homeless hostels. A mobile specialist outreach screening team with an extensive network is one potential model to improve case finding amongst “hard to reach” individuals. The Find & Treat Service is an example of such an approach – this target homeless groups and drug users for TB screening and is now starting to screen for hepatitis C.(72) This model is currently largely restricted to London. Approaches such as this and other innovative screening approaches would need to be considerably scaled up to achieve elimination targets.

Several studies have confirmed that HCV treatment is safe and effective among PWID.(73, 74) However, to improve the benefit of the treatment, we need to ensure good adherence to treatment (e.g. by providing interventions to support adherence) to maximise SVR and prevent resistance. Although the rate of resistance is currently low (75-79), there is a risk that resistance will emerge particularly if high-risk individuals (who are at greatest risk of re-infection) fail to adhere to treatment. The emergence of resistance could have a major impact on treatment coverage needed to achieve WHO’s elimination target.

Currently, treatment may be focused on relatively well engaged PWIDs in whom high SVR is possible but as treatment is expanded the social complexity of those

treated may increase such that adherence is poorer. This could have a major impact on the coverage needed and also increase the risk of treatment failure due to drug resistance. This emphasises the importance of understanding the extent to which poor adherence affects SVR and measures to increase adherence. The implementation of technological solutions such as Video Observed Therapy (VOT) may provide opportunities to improve adherence in high risk populations. Some studies have showed evidence of asynchronous VOT's acceptance and improve adherence in the similar populations.(80, 81) The applications of VOT have been used for patients with tuberculosis, and studies have shown that VOT is feasible and acceptable as monitoring adherence tools.(82-84)

Another key parameter affecting the proportion of individuals who need to be treated is the average duration of injecting. Reducing this can lead to major reductions in the threshold who need to be treated. In recent years there have been major cuts to local authority funding and consequent reductions in drug treatment services. This coincides with increased NHS spending on Hepatitis C treatment. The results show that it is very important that a national hepatitis C control strategy addresses both injecting behaviour and provides access to HCV treatment.

6.4.4 IMPLICATION FOR POLICY PRACTICES

Overall, this study provides evidence that achieving the WHO HCV elimination incidence target among PWID in London is possible by 2030 but will require a step change in the proportion of those infected who are treated and ideally supported by a range of other strategies to reduce duration of injecting drug use. To the best of my knowledge, this is the first analysis which has modelled the prevalence of treatment that would be required among PWID in London to achieve WHO target. A lot of budget (4-6 times higher than the allocation for 2016/2017) needs to be allocated in order to provide DAA treatment for people who inject drugs.

Even if financial resources were made available to cover the cost of DAA's, there are a number of barriers to achieving the WHO target: 1) Increase case finding by providing screening and testing to diagnose hepatitis C among high risk individuals 2) ensure cases who are found move on to treatment 3) ensure treatment is effective e.g. by supporting adherence, 4) establish measures for retreating treatment failures 5) ensure those treated also have addiction problems and injecting behaviour addressed, 6) improve access to effective drug treatment services. Strong collaboration and active engagements with drug and alcohol

treatment services is needed to reduce the risk of transmission and reinfection among PWID.

6.4.5 My Role In This Study

I devised the idea for the study in collaboration with an infectious disease modeller from San Diego University and my supervisors. I drew the model structure, performed literature search for parameters, and conducted model calibration. I ran the analysis to assess the treatment scale up needed to achieve WHO incidence elimination target by 2030. I also examined the results using several scenarios and performed sensitivity analysis of the study. I drafted the manuscript and plan to be submitted into international peer-reviewed journal.

Key Points:

- This study suggested 46% of HCV infected PWID need to be treated each year in London to achieve the WHO elimination target by 2030 if treatment failures are not retreated
- If treatment failures are retreated, the study suggests that treating 29.5% of current PWID chronically infected with HCV and 29.5% of those who failed treatment will achieve the WHO elimination target by 2030 in London.
- The sensitivity analysis showed that duration of injecting (suggesting the need for active engagement and collaboration with drug treatment services) and sustained virological response rate (suggesting need to support adherence and prevent resistance) had the highest impact on treatment scale up needed to achieve the elimination target. The model also shows the importance of retreating treatment failures.
- Several aspects may hamper the elimination program: the need for increased budget allocation for DAA treatment for PWID, increased case finding, ensured treatment adherence to maximise SVR and prevent drug resistance, and improved access to drug treatment and needle exchange services.

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7. DISCUSSION

7.1 SUMMARY OF KEY FINDINGS AND THEIR IMPLICATIONS FOR PUBLIC HEALTH POLICY

The introduction of Direct Acting Antiviral (DAAs) treatment means that it may become feasible to eliminate hepatitis C in the future. However, more research is required to understand factors related to infection, diagnosis and treatment of HCV (Figure 7.1) in order to develop evidence-based public health strategies to work towards the aim of eliminating[5] HCV. In this final chapter, I explain how my findings could support efforts to work towards the elimination of HCV and discuss the implications of my findings for public health policy. The diagram below describes the structure of my thesis and illustrates how I have addressed some of the important research questions relating to HCV in each of my thesis chapters.

[5] Elimination is defined as reduction to zero of the incidence of disease or infection in a defined geographical area 1. Dowdle WR. The principles of disease elimination and eradication. Bulletin of the World Health Organization. 1998;76(Suppl 2):22.

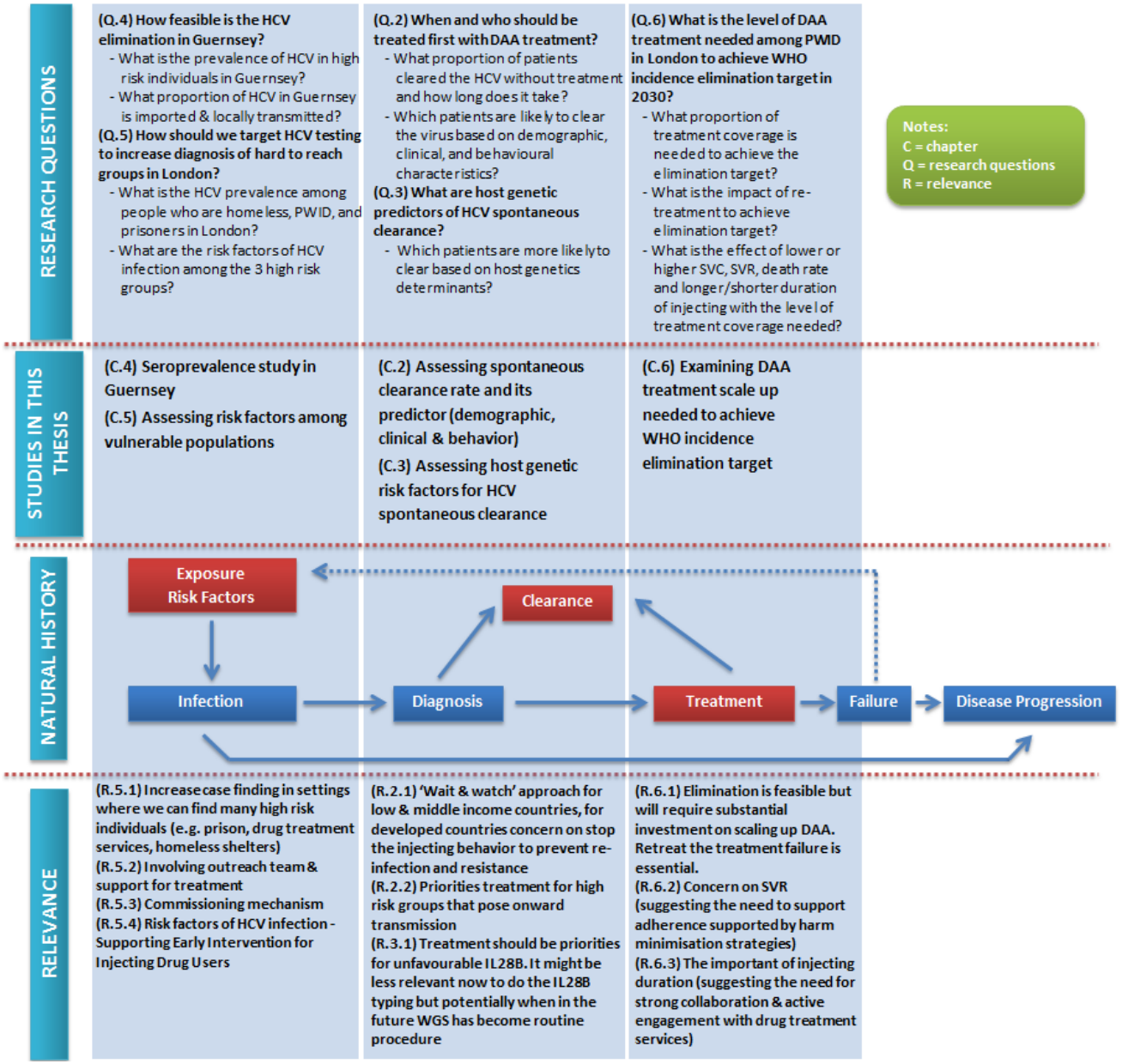

Figure 7.1 Research questions addressed in each of my 5 thesis chapters and how the relevance of my findings to health policy. Q = questions addressed by this thesis; C = chapter; R = relevance.

7.1.1 Exposure and Risk Factors for Infection

The first step to assessing the feasibility of eliminating HCV is to understand the burden of disease in the population. I set out to estimate the prevalence of HCV in a specific population (Guernsey study) and in vulnerable groups including People Who Inject Drugs (PWID), who account for approximately 90% of HCV infections in England.

In chapter 4, I outline a protocol for a seroprevalence study of HCV in Guernsey. My aim was to measure the prevalence of HCV and quantify the relative importance of local transmission versus importation of infection, then to assess the feasibility of eliminating or reducing HCV transmission through treatment with DAAs in the context of a relatively closed population (population of Guernsey is 62,229 with limited migration). Guernsey was arguably ideally suited for this exercise because it has close inter-agency collaboration, well-established health care services, a well-characterized high-risk population, and funding opportunities to treat infected high-risk individuals using the DAA treatment.

Unfortunately, we were unable to undertake the study having co-developed the protocol with the local PI and obtained ethical approvals. Since July 2016, I have been unable to contact the local PI in Guernsey leaving us no choice but to abandon the study. Nevertheless, I gained valuable experiencing in how to design a prospective research study and the processes for obtaining ethical approvals. I also learnt lessons in relation to building a collaboration and research team. This experience also taught me that communication, including face to face meetings and visits to research sites is important in collaboration.

A large number of studies in hepatitis C have focused on high-risk groups such as people who inject drugs (2-9), but fewer studies report the prevalence of HCV among prisoners (10-13) and people who are homeless (14-17). Knowing the burden of HCV among these groups is important to target screening programmes, design effective interventions, and calculate the cost-effectiveness of the intervention programs.

In chapter 5, I investigated the association between overlapping risk factors (drug use, homelessness, imprisonment) and HCV. My work provided evidence of a high burden of HCV among PWIDs and homeless populations, as well as a higher prevalence among prisoners compared to the general population. Furthermore, more than half of HCV infected individuals had intersecting risk factors of

homelessness, prison and drug use, which implies those group are important as population targets for HCV prevention and screening. This highlights the need for interventions that tackle these overlapping issues and the importance of prioritising treatment for these individuals. I also found strong association between HCV and injecting drug behaviours including duration of previous drug use.

However, this study is potentially limited by the way in which individuals were recruited. Individuals were recruited through drug treatment services, homeless shelters and a prison. This may have affected my estimates of HCV prevalence as individuals in the community who are not in contact with any of these services may actually have a higher burden of undiagnosed HCV. In prison, the testing was conducted alongside an initiative to screen for active TB using radiography. Since prisoners undergoing drug detoxification were located in another part of the prison (who were unable to access easily the testing facility) my estimates of disease prevalence exclude these higher risk prisoners and are thus very likely to underestimate the burden of HCV for this group. Despite these limitations, this study represents a valuable addition to the research literature because it is very challenging to gain valid estimates of the prevalence of HCV in vulnerable populations.

Study Relevance

7.1.1.1 Increase Case Finding in Settings Where We Can Find Many High-Risk Individuals

The work in chapter 5 highlights that people who are homeless, PWID, and prisoners have a very high burden of undiagnosed HCV compared to the rate in the general population. Consequently, there is a need to invest time and effort in case finding in the types of settings where there are large numbers of vulnerable people. In addition to screening in drug treatment services, homeless shelters and prisons should be prioritised for screening. Other settings where case finding might work include haemodialysis centres, hospital's department where we can screen transfusion-dependent patients or sexual health clinics where we can screen men who have sex with men (MSM) patients. However, as mentioned in chapter 1, the prevalence of hepatitis C might be lower compared to PWID, people who are homeless or prisoners. Accident and Emergency Departments also often treat homeless people and Injecting drug users and are a potential venue for active case finding.

Increasing case finding in the setting where vulnerable populations can be identified is reasonable given the highly dynamic and overlapping nature of these three risk groups. The work presented in chapter 5 shows that almost 60% of HCV infected individuals have overlapping risk factors (e.g. have been ever homeless, have been in prison, have been injecting/using drugs in their lifetime). Thus, providing screening and access to treatment and intervention for these groups is crucial if we want to eliminate HCV in the future. Modelling studies have also indicated that intervention among prisoners such as screening and provision of HCV treatment (18, 19) are cost-effective. Furthermore, a study has demonstrated the effectiveness of an interdisciplinary, community-based, comprehensive Nurse Case Managed HCV education intervention that addressed psychosocial issues common to homeless adults such as self-esteem building, and behavioural factors preventing adoption of safe injection practices, in addition to providing standard risk factor education.(20)

7.1.1.2 Involving Outreach Team and Support for Treatment

The first step in public health strategy working towards HCV elimination is by identifying undiagnosed HCV cases. This can be achieved by increasing the uptake of testing among high-risk individuals and by providing screening and testing for all vulnerable groups. The uptake of hepatitis C testing among PWID in England and Wales was poor with less than two-fifths having ever been tested, and only half of those tested having been tested recently.(21) Mobile screening and active case finding in places where we can reach high-risk individuals is crucial in order to provide access to testing and treatment.

In reality, some challenges found in the real world are how to increase the engagement of high-risk individuals with treatment services and how to decrease the number of people who are lost to follow up. Due to the nature of high-risk groups who do not have permanent housing, my findings also support the requirement for accessible screening programmes, intensive case management, preventative interventions, and ongoing support so we can reach and treat many of infected individuals in vulnerable populations. In the field of tuberculosis, which also affects similar high-risk populations, outreaching screening and intensive treatment support through the Find & Treat programme has been an effective strategy.(22) (23) There is a potential role for a similar model for mobile hepatitis C screening and treatment support, especially for vulnerable populations. The team which consists of social and outreach workers as well as former patients who work as peer advocates may

improve the engagement. Otherwise, in a setting where outreach services for vulnerable groups such as those provided by the Find & Treat tuberculosis team are already established, it should also include HCV screening and probably other blood-borne infections testing (e.g. HBV, HIV) in their routine program. The aspiration to eliminate HCV is unlikely to be achieved without such integrated services to maximise case detection, address underlying social issues, reduce high risk injecting and maximise adherence.

7.1.1.3 Commissioning to promote harm minimization

In recent years there have been major cuts to local authority funding and consequent reductions in drug treatment services. It has led to reductions in availability of drug treatment services and needle exchange services, which is likely to make elimination much harder. This coincides with increased NHS spending on Hepatitis C treatment. In early 2018, the NHS set out plans to be first in the world to eliminate Hepatitis C by 2025.(24) The procurement in February 2018 was the single largest medicines procurement ever done by the NHS. Over 25,000 patients have already been treated to date and this number is expected to rise to 30,000 later this year. The results from my study show that it is very important that a national hepatitis C control strategy addresses both injecting behaviour and provides access to HCV treatment. The NHS team can provide support for screening and DAA treatment provision while drug treatment services can provide support by incorporating harm minimisation strategies and strengthening engagement with target populations to prevent treatment failures and reinfection among high-risk individuals. My modeling suggests clinical Hepatitis C treatment programmes are likely to fail to eliminate Hepatitis C unless adequate drug treatment and harm minimization services are also commissioned.

7.1.1.4 Risk Factors of HCV Infection – Supporting Early Intervention for Injecting Drug Users

The works in chapter 5 suggest that longer duration of injecting drug use was an important determinant of risk of HCV infection but that even those who have injected for less than 12 months are at high risk.(7, 25-27). My work suggests the need to screen drug users early in the course of their injecting history to ensure access to treatment and reduce onward transmission and to minimize the lifetime duration of injecting through early intervention and access to drug treatment services. The work underlines the high prevalence of infection in groups who are traditionally considered hard to reach and difficult to engage in treatment. Although the advent of

DAAs has led to optimism about the prospects for HCV elimination, this will be impossible to achieve if HCV services, do not develop effective means of outreaching screening, and engaging patients in care. This will require a shift in emphasis towards outreach and highly patient centred care that addresses broader social and addiction related problems. This results was supported by Lamden et.al (28) who found injecting drugs for more than 3 years increased risks up to 3 times higher and Miller et.al study where injecting drugs for 2-3 years increased risk of infection 2 times higher, even up to 10 fold if the duration of injection was longer than 6 years (7). Moreover, results from chapter 6 also suggested that duration of injecting plays an important role in determining DAA treatment scale-up needed to achieve WHO elimination target. With scenario of 3 years shorter duration of injecting compared to base case scenario, the treatment scale-up required decreased to 19%. This highlights that changing the average duration of injection may have a very large impact on the treatment scale-up needed to achieve the elimination target.

7.1.2 Diagnosis and Clearance of HCV

As I outline in my first chapter, DAA's represent a major advance in hepatitis C treatment, but the drugs are currently very expensive which means in some settings there is a need to prioritise who gets treated first. Prioritisation would arguably be most relevant for low-income countries where cost represents the greatest barrier to treatment. In all settings policies of delayed treatment may be made more challenging by the fact that for many patients the timing of initial infection is unknown. I have explored spontaneous viral clearance (SVC) and demographic and behavioural characteristics of patients who achieve SVC in chapter 2. Low-risk individuals who are likely to achieve SVC could potentially defer HCV treatment for 12 months to give them an opportunity to clear the virus without drug treatment, enabling more individuals to be treated.

My work showed that approximately 35% of HCV infected patients will clear the virus spontaneously at 1-year post infection and those who have not cleared by this point are unlikely to do so. This is important both for informing mathematical models of HCV elimination (see chapter 6) as well as for considerations about the relevant advantages and disadvantages of delaying treatment. This is higher than previous estimates, and highlights that more than one third of patients may not require treatment. However, putting this finding into practice is challenging in high-income countries because most individuals with HCV who attend clinic will have already

been infected with HCV for >12 months and will be unaware of when they became infected. Consequently, the proportion for whom treatment could be delayed is likely to be very small. By contrast in low-income settings where access to treatment and healthcare services may be limited, there is a stronger argument for delaying treatment for low-risk individuals to maximise effective use of limited resources. In order to adopt this type of risk stratification it is helpful to be able to identify the types of patients who are most likely to achieve SVC.

My analysis of risk factors for SVC demonstrated significantly reduced clearance rates in individuals with: HIV co-infection, active intravenous drug-use and excessive alcohol intake. These findings are likely to be applicable to both high and low-income settings and provide support for a strategy of early treatment for high-risk groups who are less likely to achieve SVC. In high-income settings where intravenous drug use is the dominant mode of HCV acquisition, these findings emphasise the need to prioritise treatment for PWID to prevent onward transmission of HCV, particular given that PWID are at greater risk of being lost to follow up if treatment is delayed. However, this needs to be balanced against the problem that reinfection rates may be higher in these groups. A wide range of factors need to be assessed in determining the desirability of delayed treatment in different settings which may be best addressed through transmission modelling with linked economic analyses to assess the potential effectiveness and cost effectiveness of different strategies. It is hoped that the results of this work will inform such approaches.

In addition to investigating clinical and epidemiological risk factors for HCV, I also assessed the host genetic predictors of hepatitis C virus (HCV) spontaneous clearance to provide information that may help understand mechanisms of clearance and therefore potentially inform development of future approaches to treatment and or vaccination. In a similar way to the work presented in chapter 2, the genetic predictors could also suggest groups who should be prioritised for early treatment. However, an additional issue to consider here is the cost of screening for such genetic factors and the limited availability of technology to achieve this outside of research settings and particularly in resource poor countries. The results are presented in chapter 3. In line with previous work my analysis confirmed the importance of IL28B (IL28B rs8103142 TT genotype, IL28B rs12979860 CC genotype, and IL28B rs8099917 TT genotype) as a predictor for SVC. The results suggest that patients with unfavourable IL28B genotypes could be prioritised for having early treatment, given their low likelihood of spontaneous clearance. The

results also suggest that polymorphism in IL28B region is a very strong predictor of clearance compared to demographic, clinical and behavioural factors. Although IL28B genotyping has a potential role in terms of individualised treatment decisions it seems unlikely that a genotypic test will be widely used in the short term, particularly in low-income countries.

Study Relevance

7.1.2.1 Determining When and to Whom the Treatment Should be Given

New DAAs were initially very expensive at approximately $53,600-$84,000 for a 12-week course of treatment (29) and their availability continues to be limited in middle and low-income countries due to highly constrained healthcare budgets. By contrast, among hepatitis C patients around 15-35% of acute HCV patients will clear the virus spontaneously without treatment.(30) The result from chapter 2 in my thesis found that 36% of HCV infected patients will clear spontaneously 12 months after initial infection. Considering the limited availability of DAA treatment in some resource constrained settings, the question of which patients may spontaneously clear without treatment remains relevant, to treatment prioritisation particularly in low income settings.(31)

A previous systematic review that was published in 2006 estimated that 26% of HCV infected patients achieved spontaneous viral clearance (32), yet the study population was very heterogeneous and did not consider time since infection which potentially introduces bias in the estimate of prevalence. In chapter 2, I conducted an extensive systematic review and meta-analysis to ascertain precise estimates of viral clearance rates where time of infection was clearly reported to inform clinical decision making regarding the use of antiviral agents for HCV.

My work suggests that patients with HCV continue to spontaneously clear HCV for at least 12 months following initial infection but those who have not cleared by this point are unlikely to do so. Thus, given the high costs of treatment and the potential of new drug resistance (33-35), it may be important to give sufficient time for patients to spontaneously clear infection before initiating treatment in settings where cost limits treatment availability. This thesis provides support for a strategy of observation for a year before instigating treatment for low-risk patients thought to have been recently infected in these settings.

However, implementing this might be challenging because the majority of HCV patients are asymptomatic; thus the acute infection phase is usually undetected.(36,

37) Hepatitis C often does not have any noticeable symptoms until the liver has been significantly damaged. Many infected people may have active normal life for decades, without realising that they have been infected and physicians are usually unable to determine the initial time of infection. Consequently, a strategy of observation for one year before instigating treatment might be most applicable for individuals who are routinely screened for HCV, and/or low-risk individuals who have been infected through iatrogenic transmission which is most common in low and middle-income countries. In settings where DAA treatment is widely available, where injecting drug use is the predominant risk factor (where the chances of spontaneous clearance are much lower) and where injecting drug users pose an ongoing risk of onward transmission, treating this group should be a priority.

Considering the fact that hepatitis C disease progression is slow with 4-20% of individuals with HCV developing cirrhosis over a 20 year period (38, 39) and limited availability of DAAs treatment, it is thought to be reasonable to make decisions about prioritising treatment based on disease severity. For example, to defer treatment for certain patients with mild liver disease.(40) Recent guidelines developed by European Association for the Study of the Liver (EASL) suggested that treatment should be given without delay for patients with significant fibrosis or cirrhosis (METAVIR score F2, F3 or F4), including decompensated (Child-Pugh B or C) cirrhosis, in patients with clinically significant extra-hepatic manifestations (e.g. symptomatic vasculitis associated with HCV-related mixed cryoglobulinaemia, HCV immune complex-related nephropathy and non-Hodgkin B cell lymphoma), and in patients with HCV recurrence after liver transplantation.(41)

However, some studies conclude that DAAs treatment should be delivered to all HCV infected patients including those with stage F0-F2 fibrosis (F0 = no fibrosis; F1 = mild fibrosis; F2=moderate fibrosis) with or without evidence of coexisting liver disease.(42, 43) It is thought that early treatments of HCV infected individuals would be even more efficacious than waiting particularly if it includes all cases from F0-F4 hepatic disease.(43)

7.1.2.2 Treatment Prioritisation for High Risk Group that Pose Higher Risk of Onward Transmission

Currently in England decisions around treatment prioritisation have been made by limiting the number of available treatment courses and only providing access to individuals who are in contact with healthcare services. Given that the major burden

of disease is in PWID who are less likely to be in contact with healthcare services, and that these individuals pose the greatest risk of onward transmission of infection and have the greatest theoretical risk of drug resistance (poor compliance with treatment leading to treatment failure), there is an argument that PWID should be the priority group for treatment. However, accessing PWID is challenging and there may be political sensitivities about prioritizing treatment to these individuals.

There is now a growing consensus that high-risk individuals should be prioritised for treatment given the risk of onward HCV transmission. The recent EASL (European Association for the Study of Liver) guideline also recommends treatment should be prioritised for individuals at risk of transmitting HCV (active injection drug users, men who have sex with men with high-risk sexual practices, women of child-bearing age who wish to get pregnant, haemodialysis patients, incarcerated individuals).(41) My work in chapter 2 supports this strategy by providing evidence that people who inject drugs are less likely to achieve SVC. Besides PWID, patients with HIV co-infection also found less likely to clear. This thesis supports for a strategy of treating high-risk groups who are less likely to clear spontaneously, may pose a higher risk of onward transmission and who may be more likely to be lost to follow up.

Prioritising people who pose higher risk of onward transmission for early treatment is important as they have higher chance to transmit the infection, for example by practicing unsafe injecting behaviours among PWID. The fact that our meta-analysis result also found that people who inject drugs also tend to be less likely to have spontaneous clearance supports the strategy for providing early treatment for this high-risk group. By providing early treatment for them, it is expected that we do not only cure them but also cut the chain of HCV transmission, especially in condition where their injecting behaviours are also addressed well. In the past there has been reluctance to treat such groups because the risk of reinfection is high such that if patients are reinfected the original treatment is considered not to have been worthwhile. However, this needs to be balnced against the impact of early treatment on reducing onward transmission. Transmission modelling studies linked to economic analyses are likely to be the best approach for assessing these issues. The data from this work will inform such studies.

7.1.2.3 Treatment Prioritisation Based on Host Genetic Factors

Results in chapter 3 suggest that patients with unfavourable IL28B genotypes could be prioritised for having early treatment, considering their low likelihood of spontaneous clearance. In addition, studies found that patients with favourable

IL28B rs12979860 genotype CC were found to have better response to PEGylated interferon and ribavirin treatment.(44, 45) Thus, in the DAA treatment era, *IL28B* typing might help to inform the decision to start therapy in patients with chronic genotype 1 HCV infection, particularly in the context of the impending availability of DAA therapy.(46) In patients with the good response *IL28B* genotype, peg-IFN and RBV therapy can be considered because it is associated with high rates of sustained virological response (SVR), although the side effect profile is less favourable than direct acting agents. Although IL28B genotype is a very strong predictor of clearance, it seems unlikely that genotypic testing would be included as part of routine clinical care, particularly when there is insufficient funding to support wider use of DAA's. The benefits of genotyping would need to be considered in the context of the falling costs of DAA's treatment.

7.1.3 Treatment

The overarching goal of my thesis is to consider the feasibility of eliminating HCV and how we might work towards this goal in the future. My work in Chapter 5 emphasised that understanding the needs of vulnerable groups is fundamental to how we might address this question. Therefore, in my penultimate chapter, I developed a mathematical model which focuses on HCV treatment for PWID. Using estimates derived from my research and previous studies, I explore the feasibility of eliminating HCV among PWID in London by applying the World Health organization (WHO) targets for HCV treatment and elimination. I built a dynamic deterministic compartmental model, and parameterised the model using data from London. I conducted model calibration, examined the model using several scenarios and explored the impact of varying estimates of spontaneous clearance, duration of injecting, death rate, and SVR.

The results of the modelling study showed that achieving the WHO incidence elimination target is possible by 2030 but will require a major scale-up of DAA treatment in London, which must be supported by the effective treatment management and harm minimisation strategies outlined earlier in this chapter. Forty-six percent of PWID would need to be treated annually to achieve the WHO target started from 2017 until 2030 (or by providing 29.5% annual treatment with retreatment of treatment failures). Furthermore, the sensitivity analysis showed that duration of injecting (suggesting the need for active engagement and collaboration with drug treatment services) and sustained virological response rate (suggesting need to support adherence and prevent resistance) had the highest impact on

treatment scale up needed to achieve the elimination target, with shorter duration injecting and higher SVR leading to reductions in required treatment rate to achieve the WHO targets. However, infectious disease modelling hugely depends on the assumptions used. As the projections are based on theoretical assumption based on previous published papers, the uncertainty of parameters was one of the limitations of this study.

It seems unlikely that this target will be achieved in London, given the current availability of DAA treatment and the lack of infrastructure that exists to promote treatment for vulnerable groups. In addition, existing data on the effectiveness of DAA's is largely derived from clinical trials. Until DAA's are routinely used in more chaotic PWID who are less likely to adhere to treatment and have a higher risk of re-infection, it remains to be seen whether drug resistance will emerge as an important issue.

Study Relevance

7.1.3.1 Hepatitis C Elimination is Feasible but Will Require Substantial Investment on Scaling up DAA

So far, only one infectious disease, smallpox, has been successfully eradicated from the world.(47) In the UK, WHO has confirmed that the country has achieved measles elimination in 2016. The elimination strategies differ for each infectious disease and depend on their transmission and available interventions. For example, elimination of measles can be achieved through high rate of vaccination. For malaria as a vector-borne disease, the existence of disease reservoirs makes it unlikely to be totally eliminated. On the other hand, tuberculosis is as an airborne disease making its transmission difficult to control and the long duration of treatment making multidrug resistance a major problem in the context of poor treatment adherence. As for hepatitis C, the major advancement of hepatitis C drugs has made elimination of HCV a potentially achievable goal.(48-50) In high-income countries Hepatitis C is concentrated in PWID. The absence of a non-human reservoir also increases the feasibility of elimination. Despite the absence of an effective vaccine, the revolution of DAAs therapy is considered as a viable means to make elimination possible. My work in chapter 6 found that it may be possible to achieve WHO hepatitis C incidence elimination target in 2030 by providing DAAs treatment and targeting current PWID chronically infected with HCV in London. Nevertheless, the target is extremely ambitious and it would require a step change in the proportion of those

infected to be treated. My data also shows the importance of addressing other factors that will foster HCV elimination, including identifying undiagnosed HCV cases, treating identified cases, ensuring treatment adherence, and harm minimisation strategies.

Having identified HCV cases, the next step is to ensure that cases are treated. In the era of pegylated interferon and ribavirin treatment in England, around 20% of those testing positive for HCV RNA are thought to have accessed treatment.(51). However, as a result of DAA introduction, the recent Public Health England report shows significantly more people have accessed treatment than in earlier years (show data by FY 2015/2016 and 2016/2017 (15,506 in total compared to mean 5100 per year in 2008-2014).(52) This equates to a 19% increase in the number of individuals who were treated in 2015/2016 compared to 2008-2014 levels and a 56% increase in 2016/2017 compared to 2015/2016. This is primarily due to the introduction of the DAAs drugs in the UK since 2014/2015. Nevertheless, we can still see major barriers for treatment among vulnerable populations in the UK as the latest Unlinked Anonymous Survey revealed that the treatment uptake only 21% among high-risk individuals.(53) The study presented in chapter 5 shows that none of those identified with HCV had been treated; it indicates the need for increased efforts to ensure treatment.

Several studies have explored factors associated with barriers to treatment. Lack of awareness, fear of side effects, poor adherence and economic and social pressure may prevent HCV patients accessing treatment.(54) Among PWID, some factors related to decreased treatment uptake include current heroin use and HIV/HCV co-infection (55). Based on a qualitative study, barriers to HCV care included perceptions of HCV infection as relatively benign, fear of investigations and treatment, and feeling well.(56) A study underlined the key facilitating factors to treatment access include: combination intervention approaches encompassing social as well as biomedical interventions, low threshold access to opiate substitution therapy, and integrated delivery of multidisciplinary care.(57)

7.1.3.2 Treatment Adherence Supported by Harm Minimisation Strategies

Another consideration regarding the DAA treatment management for high-risk individuals is the need to ensure treatment adherence to minimise the risk of treatment failures and drug resistance. Treatment adherence is one of the challenges among patients treated with Pegylated Interferon due to longer duration of treatment and side effects of the therapy including influenza-like symptoms,

neuropsychiatric symptoms, hematologic abnormalities (58-60), and depression which can occur in up to 30% of patients (58). With the introduction of DAA therapies which are better tolerated, have high rate of efficacy, and offers shorter duration of treatment, some studies found increasing treatment completion among HCV patients. A study published in 2016 assessing DAA treatment effectiveness among PWID found that 95% of participants had treatment completion (61), whereas study published in 2017 found 87% treatment completion among people who are homeless (62). I identified, no studies that have assessed the DAAs treatment adherence among inmates in prison, however, there was an estimate of 79% treatment completion among prisoners.(63) It should be noted however, that unless doses have been observed, it is not possible to be sure that treatment has in fact been completed. In tuberculosis treatment, it is common to use direct observation of treatment to maximise adherence in socially complex groups. A trial of smartphone-enabled Video Observed therapy has also recently shown major increases in the ability to observe treatment successfully and at lower cost. Pilots of the intervention in Hepatitis C patients have also started. It remains unclear the extent to which strict adherence to new treatment regimens is necessary to ensure viral clearance, prevent relapse and prevent resistance. However, with increasing scale up of treatment to socially complex groups it is highly likely that treatment adherence will be poor. This may lead to increased transmission, clinical failures and resistance over time. Further research is needed to understand the role of adherence and of supportive interventions such as Video Observed Therapy in HCV.

Harm minimisation strategies also play a pivotal role to support the HCV elimination program.(64) Ensuring injecting drug use behaviours are addressed and addictions are managed is important to minimise transmission and the risk of reinfection. Modelling studies have shown that harm reductions programs including needle and syringe programs in combination with DAA treatment strategy will lead to significant reductions in HCV prevalence among PWID.(65-67) My thesis underlined the feasibility of hepatitis C elimination through treatment, but we also need to consider some aspects for strengthening drug treatment management (especially among high-risk individuals) to achieve the elimination target, set by WHO in 2030.

7.1.3.3 Strong Collaboration and Active Engagements with Drugs and Alcohol Treatment Services

Another factor that needs to be considered for intervention strategies is strong collaboration and active engagement with drugs and alcohol treatment services to

help reduce unsafe injecting drugs practice, addiction control, and maintain adherence among PWID. Based on the results in chapter 5, people who inject drugs with or without sharing needles are at risk of HCV infection. Moreover, injecting drugs even for less than a year have 14 times higher risk of HCV infection compared to non-injectors. This highlights the need to work with drugs treatment services to maximise the intervention effects for drugs injectors. If we are to meet the WHO target and eliminate HCV as a major public health threat by 2030, it is essential that local authorities, drug services and the NHS work together to improve testing and treatment for individuals at risk.

7.1.4 Case Studies of HCV Elimination

In order to assess the feasibility of hepatitis C elimination among populations, we need to first test whether this can be achieved in a defined population. Currently, some countries around the globe are working on the hepatitis elimination program. During the World Hepatitis Summit (WHS) in Sao Paulo Brazil 2017, it was announced that nine countries are on course to eliminate hepatitis C by 2030, including Australia, Brazil, Egypt, Georgia, Germany, Iceland, Japan, the Netherlands and Qatar.(68, 69) Some key countries which were being highlighted at the summit for their innovative work to eliminate viral hepatitis were Georgia, Brazil, Australia, and Egypt.

In Georgia, as a form of partnership with the US Centres for Disease Control and Prevention and Gilead Sciences in April 2015, the country launched the world's first national HCV elimination programme, aiming to reduce HCV prevalence by 90% by 2020. After 2 years since the launch of the programme, almost 40,000 patients have started treatment with new antiviral medicines, out of which almost 32,000 have already completed the treatment successfully.(70, 71) Large-scale activities are taking place to ensure at-risk groups are screened, including key populations, medical personnel and all hospitalised people. Meanwhile, Brazil has committed to gradually lift treatment restrictions in 2018, meaning that the country will be able to treat all people infected with hepatitis C, ensuring it is on target to eliminate hepatitis C.(72) Previously, treatment was restricted to only the sickest patients with advanced liver disease. In Australia, the Australian government responded to the call for universal access to the hepatitis C DAAs with an AUS $1billion dollar investment over 5 years. This risk-sharing agreement with pharmaceutical companies provides government-funded treatment to all adults without restriction and has paved the way for the elimination of hepatitis C by 2030. More than 30,000

patients with hepatitis C were treated and cured in 2016.(73, 74) As for Egypt, in 2017, the country pledged to test 30 million for hepatitis C by the end of 2018 by implementing mass screening initiatives (including assistance from the military), as well as mass producing generic copies of DAA drugs for under the US $200 per 12-week course.(68, 75)

There is still a lot of work to do to achieve global elimination of hepatitis C. The different geographic location, culture, burden of disease among populations, dominance of PWID versus iatrogenic transmission by setting, health systems, budget allocation, and access to vulnerable populations adds to the complexity of strategies that need to be developed for each country. Nevertheless, this thesis has tried to develop evidence to inform public health strategies to work towards this elimination in a range of settings.

7.2 Identifying Areas for Further Research

Although some work has been done to establish the public health strategies towards hepatitis C elimination, there are some areas where further research is required. First, findings from chapter 2 might be helpful in the context of low and middle-income countries, but further research to look at how this might work to support treatment prioritisation in these setting would be required. More research is needed to assess the feasibility and cost effectiveness of adopting approach delaying treatment for one year in low-income settings. In terms of identifying individuals who are most likely to achieve SVC, meta-regression analysis would be valuable to adjust for potential confounders in individual-level data. Moreover, to improve the meta-analysis performed in chapter 3 to assess the host genetics predictors of HCV spontaneous clearance, it would be interesting to analyse data from a large and well characterised population as so far, the studies which have been done focus on limited host genetic predictors only.(76-78) This could be achieved, for example through large scale Genome Wide Assossiation Studies (GWAS) rather than targeting individual genetic elements.

Despite the fact that some hepatitis C control programs towards elimination have been ongoing in some countries as mentioned in section 7.1.5 above, future research should focus on how screening, treatment and prevention services can be integrated for vulnerable populations to maximise treatment access and reduce reinfection. Studies need to be context specific to take account of the effectiveness in different settings. The study should start by focusing on a specific geographic

area with a well-characterised high-risk population and well-established healthcare services.

Learning from the study conducted in chapter 5 where we estimated the prevalence of hepatitis C among vulnerable populations (people who are homeless, people who inject drugs, and prisoners), further research should include larger number of population recruited which represents these vulnerable groups in a range of study areas in different countries. More research is also required to work out how to engage with these populations to promote uptake of treatment.

The study presented in chapter 6 (modelling treatment scale-up needed for PWID in London to achieve WHO incidence elimination target by 2030) could be improved by incorporating parameters of HCV infection stages (acute, fibrosis F0-F4, cirrhosis, decompensated cirrhosis), HCV co-infection (e.g. HBV, HIV, TB), harm minimisation strategies, different genotypes, and varied rate of treatment adherence (which might be the case for high-risk groups) which is more realistic considering the complexity faced in the real world. In addition, health economic modelling studies need to be conducted to find the most cost-effective pathway for screening, diagnosing, and managing treatment among high-risk individuals. Operational research is needed to understand how best to scale up HCV screening and effective treatment in vulnerable populations and to reduce transmission through addressing addiction and injecting behaviour in high income countries.

Finally, currently treatment may be focused on relatively well-engaged PWIDs in whom high SVR is possible but as treatment is expanded the social complexity of those treated may increase such that adherence is poorer and DAAs drug resistance and reinfection occur more frequently. This could have a major impact on efforts to work towards elimination of HCV This emphasises the importance of understanding the extent to which poor adherence and drug resistance affects SVR and measures to increase adherence and decrease reinfection.

In summary, a wide range of factors impact on the ability of a single country to work towards elimination of HCV. The work presented here improves our understanding of spontaneous clearance, which may inform treatment prioritisation and modelling and underlines the critical importance of intensifying and scaling up control measures in socially complex groups if elimination is to be achieved.

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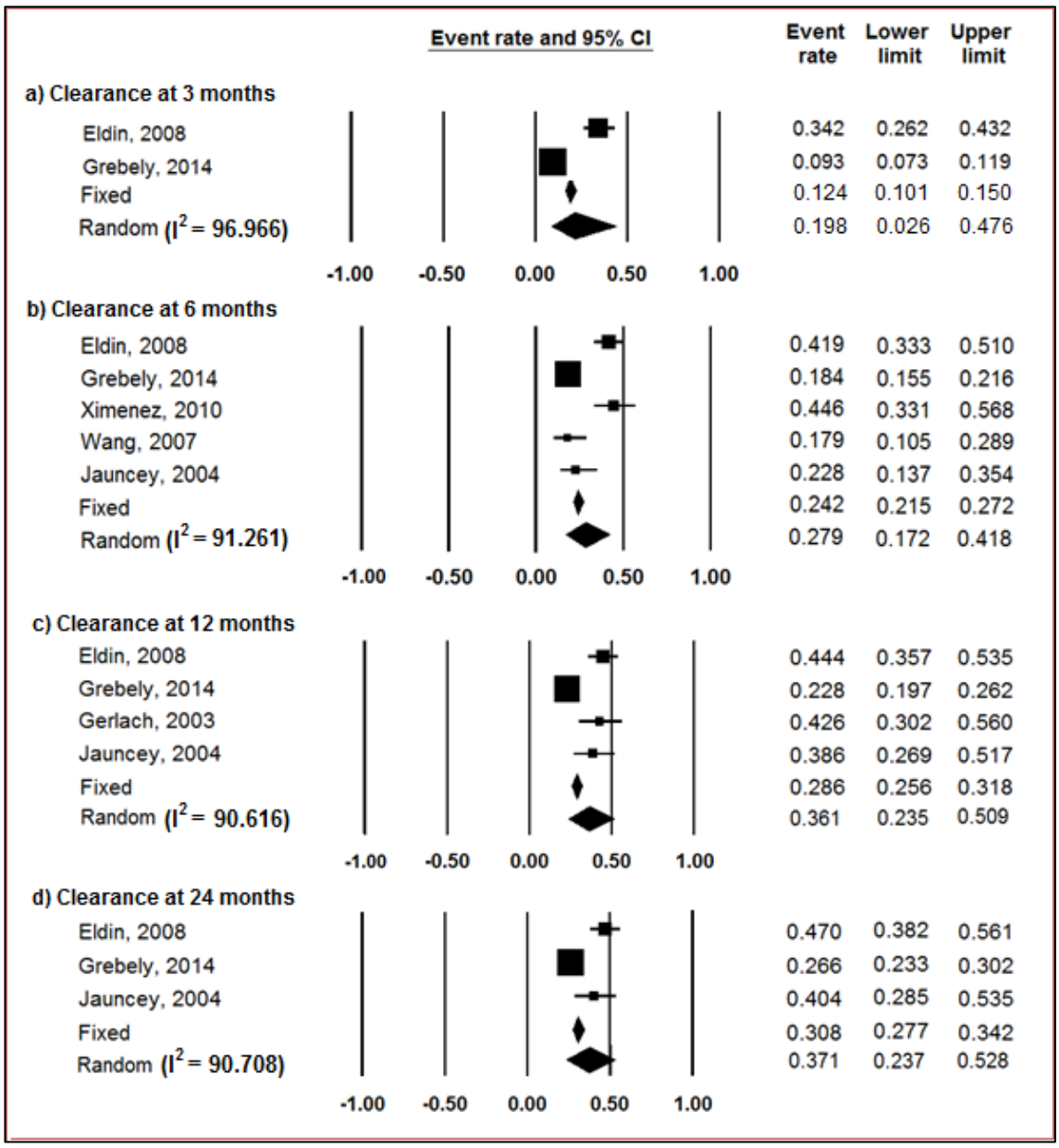
APPENDIX 1. RATE OF SPONTANEOUS CLEARANCE IN SEVERAL TIME POINTS



APPENDIX 2. RESULT OF META-ANALYSIS EXAMINING DEMOGRAPHIC, CLINICAL, AND BEHAVIOUR FACTORS ASSOCIATED WITH HCV SPONTANEOUS CLEARANCE - ALL STUDIES VS STUDIES WITH MINIMUM 12 MONTHS FOLLOW UP

Determinants	All Studies					>12 months study				
	∑ study	I^2	OR	Lower CI	Upper CI	∑ study	I^2	OR	Lower CI	Upper CI
Demographic Determinants										
Male vs Female‡	8	0·00	0·68	0·57	0·81	6	0·00	0·61	0·48	0·78
Age ≥ 30 yo vs <30 yo	6	37·96	0·86	0·68	1·09	3	0·00	0·77	0·54	1·09
Age ≥ 40 yo vs <40 yo	5	81·33	0·52	0·26	1·07	1	NA	NA	NA	NA
Age ≥ 45 yo vs <45 yo	4	33·71	0·79	0·64	0·97	2	21·87	1·03	0·49	2·16
Black vs Non-Black	3	57·58	0·38	0·20	0·75	2	46·19	0·30	0·13	0·69
Non-Aborigin vs Aborigin	5	43·63	0·47	0·36	0·62	3	58·07	0·55	0·28	1·07
African American vs Non-African American	5	95·33	0·29	0·08	1·05	0	NA	NA	NA	NA
White vs Others	5	60·38	0·94	0·53	1·48	2	0·00	1·53	0·70	3·34
Clinical Determinants										
Asymptomatic Infection	7	0·00	0·38	0·27	0·56	6	0·00	0·41	0·28	0·62
HIV Co-infection‡	4	0·00	0·50	0·37	0·66	2	0·00	0·54	0·40	0·75
Non HBV co-infection‡	7	49·45	0·21	0·14	0·32	2	47·68	0·22	0·11	0·45
Non Genotype 1 vs Genotype 1	4	0·00	0·63	0·45	0·89	4	0·00	0·63	0·45	0·89
Behaviour Determinants										
IDU vs Non-IDU	7	82·88	0·59	0·37	0·93	2	75·14	1·04	0·24	4·44
Alcohol*	5	72·15	0·67	0·47	0·95	2	38·99	0·50	0·26	0·96

‡Multivariate analysis

*Alcohol drinker or had history of drinking excess alcohol

APPENDIX 3. NON META-ANALYSIS OF HOST GENETIC DETERMINANTS ON HCV SPONTANEOUS VIRAL CLEARANCE

Other Gene Candidates						Notes
IL1B-511	1	-	0.80	0.59	1.09	CC vs CT/TT
IL1B+3954	1	-	1.10	0.74	1.61	AA vs AG/GG
IL1RN+2018	1	-	0.98	0.69	1.38	AA vs AG/GG
IL1A+4845	1	-	1.10	0.77	1.55	AA vs AC/CC
IL6-174	1	-	1.41	0.88	2.26	GG vs GC/CC
IL10-1082	3	-				AA vs AA/GA. IL10-1082, IL10-819, IL10-592 have no association with HCV clearance
IL-18 +105C allele with haplotype GCC	1	-	*0.59*	*0.41*	*0.84*	
IL-22-rs1012356 AA vs non AA	1	-	*0.26*	*0.08*	*0.86*	
IL-10 ATA	1	-				The IL-10 ATA haplotype was more frequent in patients with spontaneous HCV RNA clearance (36.0%) than in patients with persistent infection (23%) (p = 0.009, p corrected = 0.036)
IL28B rs11881222	1	-	*3.07*	*1.52*	*6.20*	AA vs AG/GG
IL10RB-rs2834167 AG IL28RA-rs11249006 GG IL28RA rs10903035 AA	1	-				No association with HCV clearance in Chinese population
TLR7 rs179016 C allele	1	-	0.75	0.53	1.07	
TLR7 rs1634323 G allele	1	-	*1.79*	*1.12*	*2.87*	
TLR7 rs3853839	1	-	*0.25*	*0.17*	*0.37*	GG vs GC/CC
Immunoglobulin GM ff (+) & HLA C1C2 (-) or GMff (-) & HLA C2C2 (+)	1	-				HLA C1C1 genotype, in the absence of GM ff, were more than seven times [odds ratio (OR) 7.15, 1.54-33.20] as likely to have persistent infection

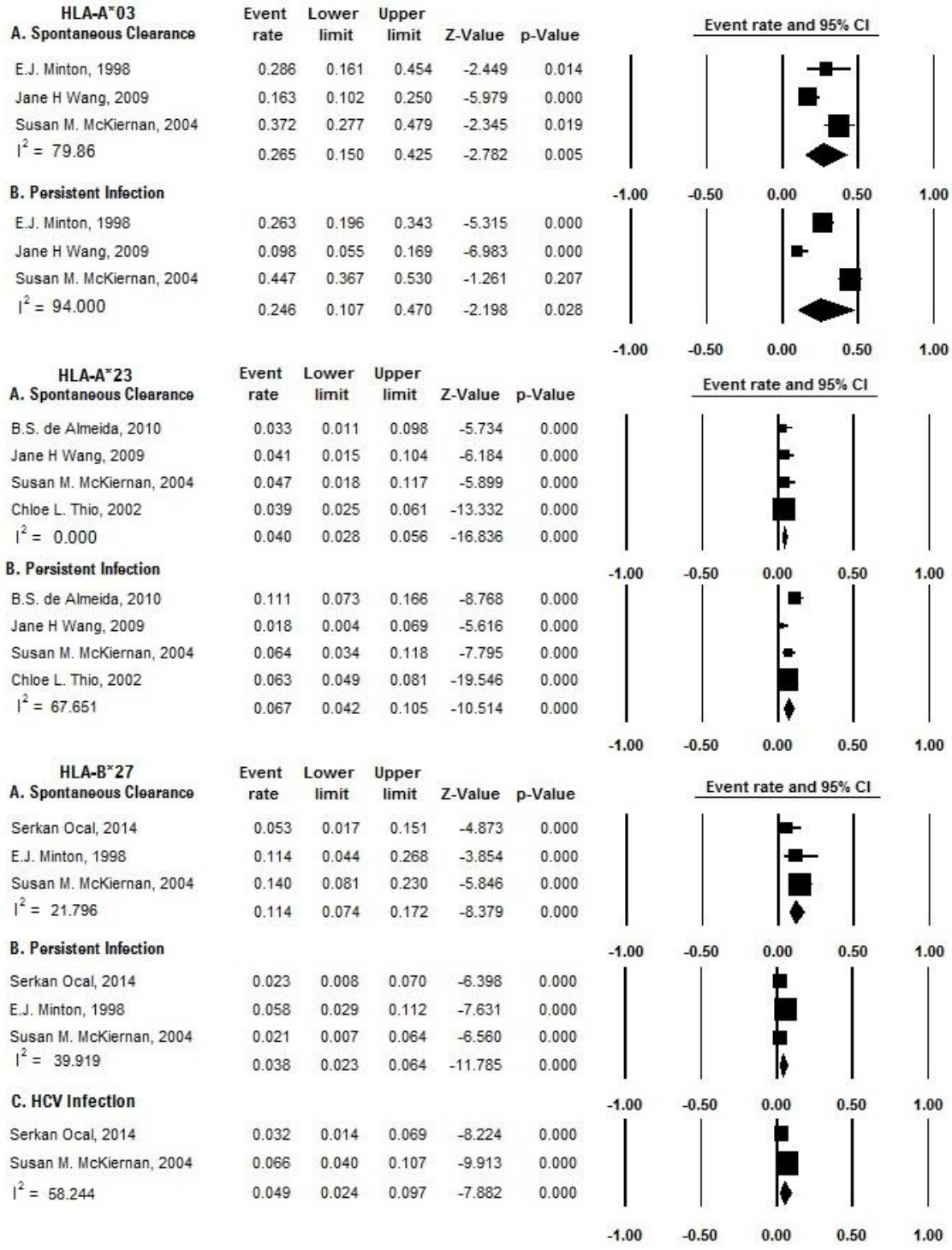
Other Gene Candidates						Notes
Immunoglobulin GM & KM	1	-				None of the GM or KM phenotypes by itself was associated with the clearance or persistence, but the combination of phenotypes has strong association. Eg. GM 1,17 5,13 and KM 1,3 support clearance, GM 1,3,17 23 5,13 phenotype (in the absence of KM 3) was associated with persistence.
LMP2 rs17587 A	1	-	*2.87*	*1.27*	*6.50*	in Chinese population
LMP7 genes codon 145 **Gln/Lys** **Lys/Lys** **Gln/Lys+Lys/Lys**	1	-	*0.61* *0.42* *0.57*	*0.39* *0.19* *0.37*	*0.96* *0.92* *0.87*	Gln/Gln as reference
IFLN4 TT/TT **IFLN4 TT/ΔG**	1	-	3.59 0.95			Significant result was only found among black people. IFNL4-ΔG/ΔG = 11.9% as reference
ESR rs2228480 A allele	1	-	*0.83*	*0.64*	*0.87*	in Chinese population
ESR2 rs4986938 AA genotype	1	-	0.27			Females carrying the rs4986938 AA genotype appeared to clear HCV spontaneously
TAP rs9277972 TT vs AA	1	-	2.63	0.46	15.14	in Chinese population
MDA-5 R843H (rs3747517) C/T	1	-				Melanoma Differentiation-Associated gene 5 support HCV clearance
SUMO1 rs10185956T	1	-	*2.71*	*1.33*	*5.87*	Small Ubiquitin-Like Modifier (SUMO) in Morrocan population
P170 (CD24 Ala57Val)	1	-	*2.11*	*1.19*	*3.73*	in Chinese population
APOE*E2 **APOE*E4**	1	-	*0.39* *0.60*	*0.21* *0.38*	*0.73* *0.96*	Apolipoprotein E
TNF-α -863A	1	-	*1.92*	*1.075*	*3.45*	
TNF- α -308, TNF- α 238	1	-				No significant difference in distribution of any of polymorphisms was found

Other Gene Candidates						Notes
IFN-α rs2069707 C/G	1	-	3.51	1.0	12.5	
L SIGN genotype 7/4 **4-L-SIGN**	1	-	*4.00* *3.28*	*1.359* *1.45*	*11.70* *7.45*	

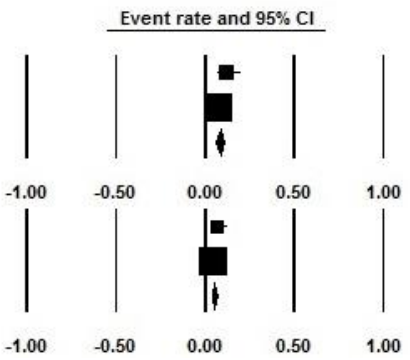
Italic typing : statistically significant based on 1 study

APPENDIX 4. FOREST PLOT OF EACH HOST GENETIC DETERMINANTS ASSOCIATED WITH HCV SPONTANEOUS CLEARANCE

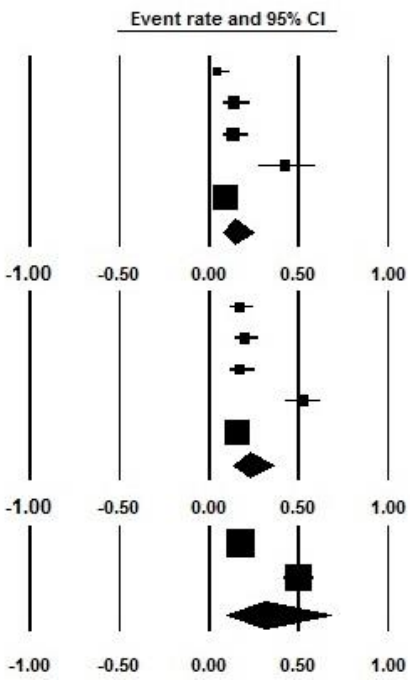
HLA Class I



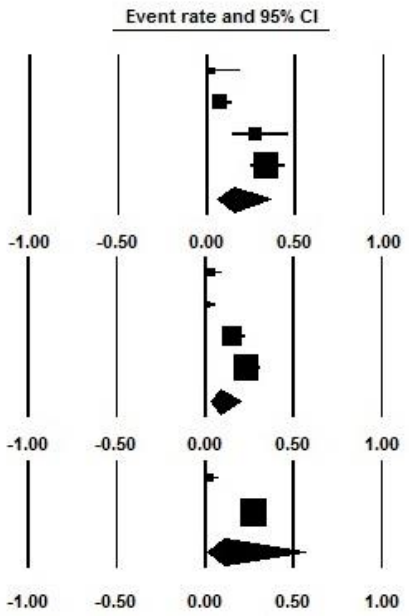
HLA-B*57 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Susan M. McKiernan, 2004	0.116	0.064	0.203	-6.029	0.000
Chloe L. Thio, 2002	0.074	0.053	0.101	-14.215	0.000
I^2 = 43.093	0.081	0.061	0.108	-15.383	0.000
B. Persistent Infection					
Susan M. McKiernan, 2004	0.071	0.039	0.127	-7.842	0.000
Chloe L. Thio, 2002	0.046	0.034	0.062	-18.937	0.000
I^2 = 35.804	0.050	0.038	0.065	-20.458	0.000



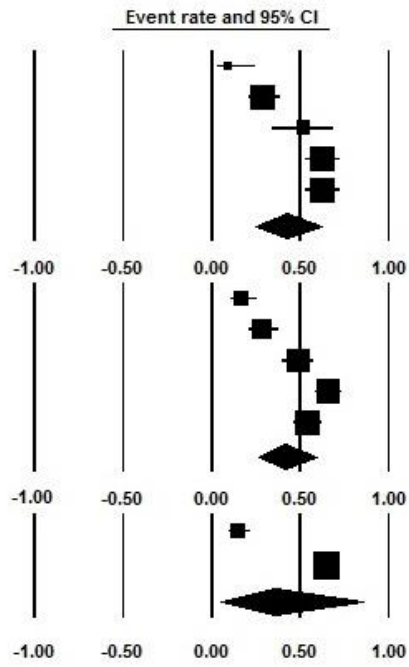
HLA-Cw*04 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Liam J. Fanning, 2004	0.047	0.018	0.117	-5.899	0.000
Susan M. McKiernan, 2004	0.140	0.081	0.230	-5.846	0.000
Jane H Wang, 2009	0.133	0.079	0.215	-6.305	0.000
Serkan Ocal, 2014	0.424	0.270	0.595	-0.867	0.386
Chloe L. Thio, 2002	0.095	0.072	0.126	-14.204	0.000
I^2 = 86.674	0.138	0.073	0.247	-5.035	0.000
B. Persistent Infection					
Liam J. Fanning, 2004	0.173	0.118	0.245	-6.982	0.000
Susan M. McKiernan, 2004	0.199	0.141	0.273	-6.609	0.000
Jane H Wang, 2009	0.170	0.111	0.251	-6.308	0.000
Serkan Ocal, 2014	0.521	0.421	0.619	0.408	0.683
Chloe L. Thio, 2002	0.153	0.131	0.178	-18.352	0.000
I^2 = 93.818	0.225	0.132	0.356	-3.759	0.000
C. HCV Infection					
Susan M. McKiernan, 2004	0.176	0.132	0.231	-8.853	0.000
Serkan Ocal, 2014	0.496	0.411	0.582	-0.088	0.930
I^2 = 97.368	0.315	0.093	0.672	-1.021	0.307



HLA-Cw*05 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Serkan Ocal, 2014	0.030	0.004	0.186	-3.413	0.001
Jane H Wang, 2009	0.071	0.034	0.142	-6.539	0.000
E.J. Minton, 1998	0.276	0.144	0.462	-2.323	0.020
Susan M. McKiernan, 2004	0.337	0.245	0.443	-2.963	0.003
I^2 = 86.776	0.153	0.055	0.357	-2.983	0.003
B. Persistent Infection					
Serkan Ocal, 2014	0.031	0.010	0.092	-5.854	0.000
Jane H Wang, 2009	0.009	0.001	0.061	-4.688	0.000
E.J. Minton, 1998	0.149	0.096	0.224	-6.828	0.000
Susan M. McKiernan, 2004	0.234	0.171	0.311	-5.961	0.000
I^2 = 87.509	0.083	0.031	0.202	-4.558	0.000
C. HCV Infection					
Serkan Ocal, 2014	0.031	0.012	0.080	-6.776	0.000
Susan M. McKiernan, 2004	0.273	0.219	0.335	-6.571	0.000
I^2 = 95.382	0.103	0.010	0.562	-1.757	0.079

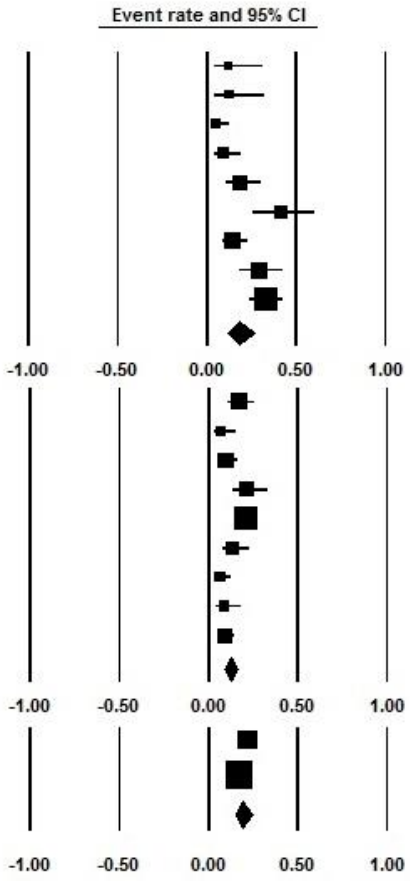


HLA-Cw*07 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Serkan Ocal, 2014	0.091	0.030	0.247	-3.803	0.000
Jane H Wang, 2009	0.286	0.205	0.383	-4.098	0.000
E.J. Minton, 1998	0.517	0.341	0.689	0.186	0.853
Susan M. McKiernan, 2004	0.628	0.521	0.723	2.345	0.019
Liam J. Fanning, 2004	0.628	0.521	0.723	2.345	0.019
I^2 = 90.511	0.422	0.246	0.621	-0.762	0.446
B. Persistent Infection					
Serkan Ocal, 2014	0.167	0.105	0.255	-5.877	0.000
Jane H Wang, 2009	0.286	0.210	0.376	-4.381	0.000
E.J. Minton, 1998	0.488	0.400	0.576	-0.273	0.785
Susan M. McKiernan, 2004	0.660	0.578	0.733	3.721	0.000
Liam J. Fanning, 2004	0.540	0.456	0.621	0.932	0.351
I^2 = 93.959	0.418	0.262	0.591	-0.928	0.354
C. HCV Infection					
Serkan Ocal, 2014	0.147	0.096	0.219	-7.068	0.000
Susan M. McKiernan, 2004	0.648	0.583	0.707	4.379	0.000
I^2 = 98.551	0.362	0.053	0.852	-0.478	0.633

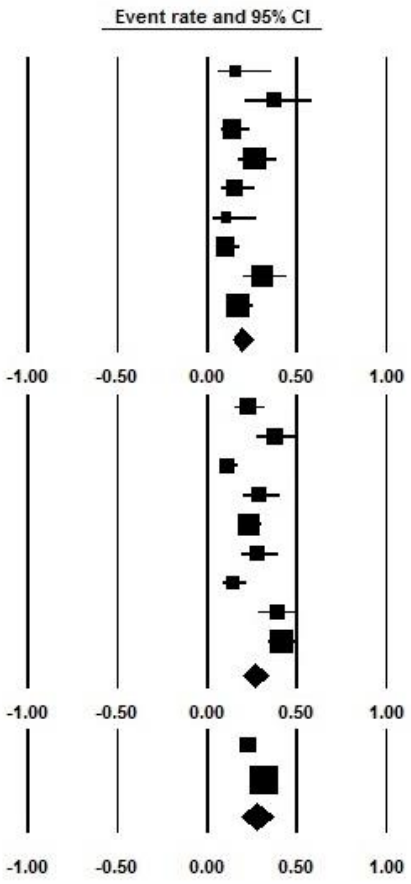


HLA Class II

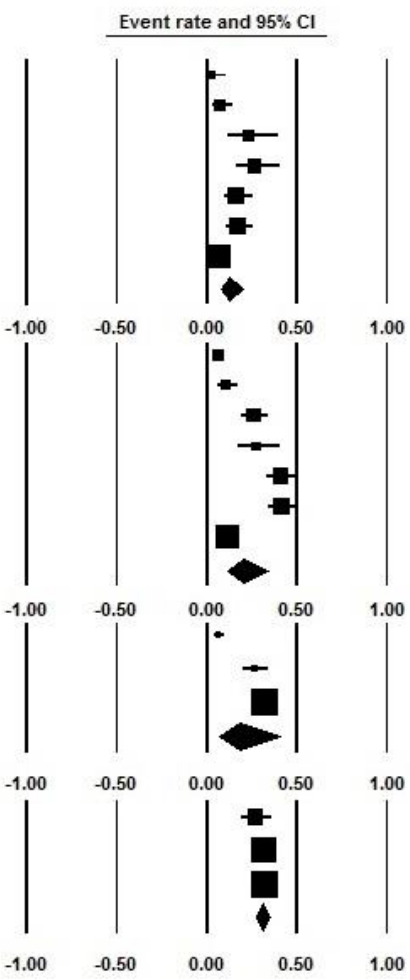
DRB1*01 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Laurent Alric, 1997	0.120	0.039	0.313	-3.237	0.001
Leila K. Cheikrouhou, 2007	0.125	0.041	0.324	-3.153	0.002
Jose Azocar, 2003	0.050	0.019	0.126	-5.740	0.000
Liam J. Fanning, 2000	0.094	0.043	0.193	-5.290	0.000
L. Alric, 2000	0.186	0.106	0.306	-4.407	0.000
Jeny R. C. Santos, 2007	0.414	0.252	0.596	-0.924	0.356
Jane H Wang, 2009	0.143	0.086	0.227	-6.207	0.000
S Barret, 2001	0.288	0.182	0.425	-2.950	0.003
Susan M. McKiernan, 2000	0.326	0.240	0.427	-3.313	0.001
I^2 = 77.697	0.180	0.116	0.268	-5.805	0.000
B. Persistent Infection					
Laurent Alric, 1997	0.167	0.106	0.252	-6.058	0.000
Leila K. Cheikrouhou, 2007	0.067	0.028	0.150	-5.701	0.000
Jose Azocar, 2003	0.097	0.058	0.157	-7.923	0.000
Liam J. Fanning, 2000	0.217	0.136	0.330	-4.389	0.000
L. Alric, 2000	0.206	0.152	0.273	-7.117	0.000
Jeny R. C. Santos, 2007	0.133	0.073	0.230	-5.510	0.000
Jane H Wang, 2009	0.063	0.030	0.125	-6.937	0.000
S Barret, 2001	0.087	0.040	0.180	-5.504	0.000
Susan M. McKiernan, 2000	0.088	0.052	0.145	-8.059	0.000
I^2 = 68.428	0.122	0.088	0.165	-10.882	0.000
C. Healthy Control					
L. Alric, 2000	0.218	0.190	0.247	-14.939	0.000
Susan M. McKiernan, 2000	0.171	0.155	0.189	-25.963	0.000
I^2 = 87.476	0.192	0.151	0.242	-9.677	0.000



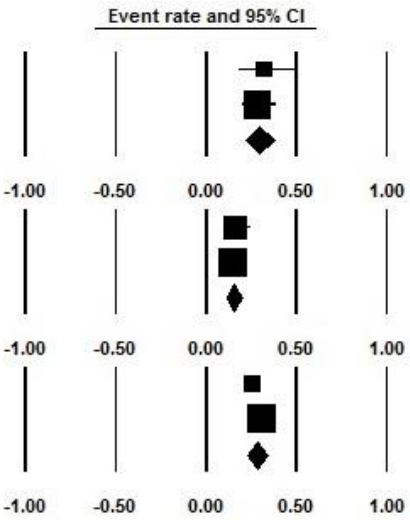
DRB1*03	Event rate	Lower limit	Upper limit	Z-Value	p-Value
A. Spontaneous Clearance					
Laurent Alric, 1997	0.160	0.061	0.357	-3.040	0.002
Leila K. Cheikrouhou, 2007	0.375	0.208	0.578	-1.212	0.226
Jose Azocar, 2003	0.138	0.078	0.231	-5.656	0.000
Liam J. Fanning, 2000	0.266	0.172	0.386	-3.593	0.000
L. Alric, 2000	0.153	0.081	0.268	-4.736	0.000
Jeny R. C. Santos, 2007	0.103	0.034	0.276	-3.542	0.000
Jane H Wang, 2009	0.102	0.056	0.179	-6.517	0.000
S Barret, 2001	0.308	0.198	0.445	-2.699	0.007
Susan M. McKiernan, 2000	0.168	0.106	0.257	-5.825	0.000
I^2 = 60.525	0.188	0.137	0.252	-7.665	0.000
B. Persistent Infection					
Laurent Alric, 1997	0.225	0.155	0.317	-5.208	0.000
Leila K. Cheikrouhou, 2007	0.373	0.272	0.487	-2.170	0.030
Jose Azocar, 2003	0.111	0.069	0.174	-7.842	0.000
Liam J. Fanning, 2000	0.290	0.195	0.407	-3.377	0.001
L. Alric, 2000	0.229	0.172	0.299	-6.642	0.000
Jeny R. C. Santos, 2007	0.280	0.190	0.392	-3.672	0.000
Jane H Wang, 2009	0.143	0.089	0.221	-6.635	0.000
S Barret, 2001	0.391	0.284	0.510	-1.791	0.073
Susan M. McKiernan, 2000	0.412	0.336	0.493	-2.126	0.034
I^2 = 84.488	0.261	0.194	0.341	-5.388	0.000
C. Healthy Control					
L. Alric, 2000	0.226	0.199	0.257	-14.551	0.000
Susan M. McKiernan, 2000	0.318	0.297	0.339	-15.545	0.000
I^2 = 95.594	0.271	0.190	0.369	-4.260	0.000



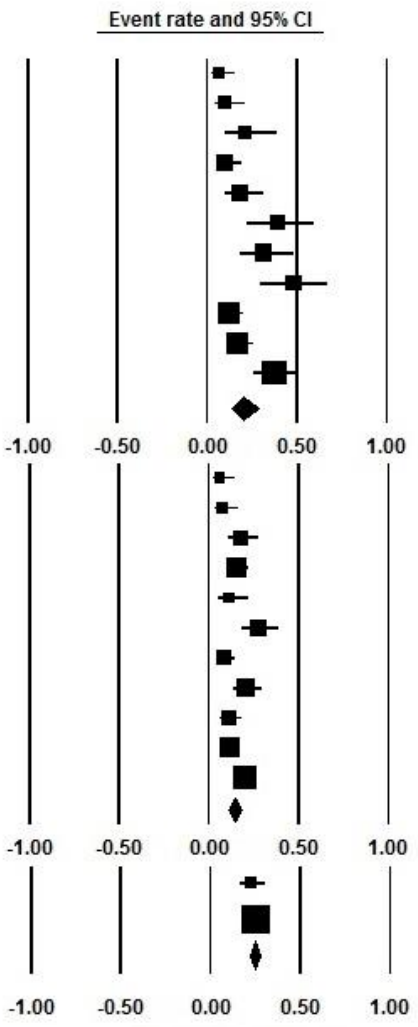
DRB1*03011 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 1999	0.029	0.007	0.107	-4.915	0.000
Alessandra Mangia, 2011	0.071	0.034	0.142	-6.539	0.000
E.J. Minton, 1998	0.229	0.119	0.395	-3.022	0.003
Matthew E. Cramp, 1998	0.265	0.161	0.405	-3.148	0.002
Susan M. McKiernan, 2004	0.163	0.099	0.256	-5.606	0.000
Susan M. McKiernan, 2000	0.168	0.106	0.257	-5.825	0.000
Chloe L. Thio, 2001	0.070	0.049	0.100	-13.200	0.000
I^2 = 81.370	0.126	0.076	0.202	-6.752	0.000
B. Persistent Infection					
Alessandra Mangia, 1999	0.064	0.041	0.098	-11.332	0.000
Alessandra Mangia, 2011	0.103	0.062	0.166	-7.672	0.000
E.J. Minton, 1998	0.259	0.192	0.340	-5.345	0.000
Matthew E. Cramp, 1998	0.273	0.172	0.404	-3.240	0.001
Susan M. McKiernan, 2004	0.411	0.333	0.494	-2.094	0.036
Susan M. McKiernan, 2000	0.412	0.336	0.493	-2.126	0.034
Chloe L. Thio, 2001	0.116	0.095	0.141	-17.781	0.000
I^2 = 95.952	0.203	0.112	0.338	-3.842	0.000
C. Healthy Control					
Alessandra Mangia, 1999	0.063	0.043	0.091	-13.110	0.000
Matthew E. Cramp, 1998	0.261	0.194	0.342	-5.287	0.000
Susan M. McKiernan, 2000	0.318	0.297	0.339	-15.545	0.000
I^2 = 97.635	0.184	0.070	0.402	-2.670	0.008
D. HCV Infection					
Matthew E. Cramp, 1998	0.269	0.193	0.362	-4.517	0.000
Susan M. McKiernan, 2004	0.317	0.260	0.381	-5.376	0.000
Susan M. McKiernan, 2000	0.317	0.261	0.378	-5.571	0.000
I^2 = 0.000	0.309	0.272	0.348	-8.913	0.000



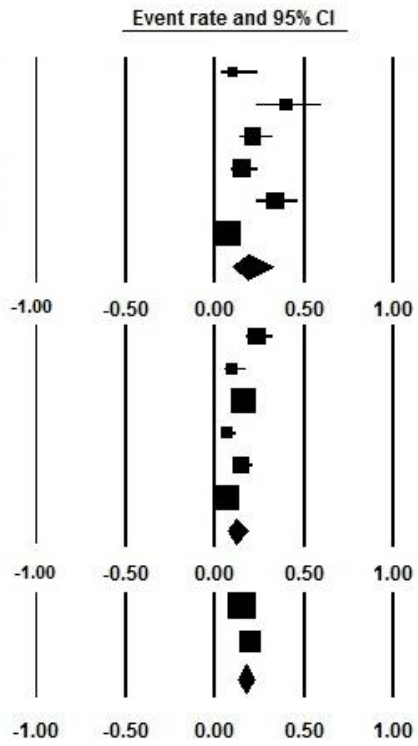
DRB1*0401 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Laurent Alric, 1997	0.321	0.176	0.511	-1.847	0.065
Susan M. McKiernan, 2000	0.284	0.203	0.383	-4.061	0.000
I^2 = 0.000	0.293	0.219	0.379	-4.445	0.000
B. Persistent Infection					
Laurent Alric, 1997	0.157	0.098	0.241	-6.177	0.000
Susan M. McKiernan, 2000	0.149	0.100	0.215	-7.553	0.000
I^2 = 0.000	0.152	0.113	0.202	-9.756	0.000
C. Healthy Control					
Laurent Alric, 1997	0.253	0.224	0.284	-13.336	0.000
Susan M. McKiernan, 2000	0.306	0.285	0.327	-16.512	0.000
I^2 = 87.086	0.280	0.231	0.335	-7.135	0.000



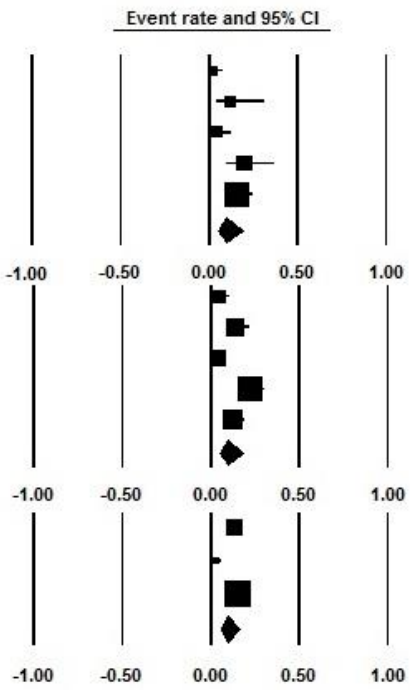
DRB1*11 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Liam J. Fanning, 2000	0.063	0.024	0.155	-5.244	0.000
S Barret, 2001	0.096	0.041	0.211	-4.763	0.000
Jeny R. C. Santos, 2007	0.207	0.096	0.390	-2.931	0.003
Jose Azocar, 2003	0.100	0.051	0.187	-5.896	0.000
Matthew E. Cramp, 1998	0.184	0.098	0.317	-4.043	0.000
Rebecca A. Haris, 2008	0.391	0.218	0.598	-1.034	0.301
E.J. Minton, 1998	0.314	0.183	0.483	-2.143	0.032
Laurent Alric, 1997	0.480	0.296	0.669	-0.200	0.842
Jane H Wang, 2009	0.122	0.071	0.203	-6.391	0.000
B.S. de Almeida, 2010	0.167	0.103	0.258	-5.690	0.000
L. Alric, 2000	0.373	0.260	0.502	-1.931	0.053
I^2 = 78.793	0.202	0.136	0.289	-5.675	0.000
B. Persistent Infection					
Liam J. Fanning, 2000	0.058	0.022	0.144	-5.412	0.000
S Barret, 2001	0.072	0.030	0.163	-5.490	0.000
Jeny R. C. Santos, 2007	0.173	0.103	0.276	-5.121	0.000
Jose Azocar, 2003	0.153	0.103	0.221	-7.395	0.000
Matthew E. Cramp, 1998	0.109	0.050	0.222	-4.855	0.000
Rebecca A. Haris, 2008	0.271	0.180	0.387	-3.674	0.000
E.J. Minton, 1998	0.081	0.046	0.141	-7.700	0.000
Laurent Alric, 1997	0.206	0.138	0.295	-5.513	0.000
Jane H Wang, 2009	0.107	0.062	0.179	-6.940	0.000
B.S. de Almeida, 2010	0.117	0.077	0.172	-8.719	0.000
L. Alric, 2000	0.200	0.147	0.267	-7.230	0.000
I^2 = 66.321	0.139	0.107	0.179	-11.956	0.000
C. Healthy Control					
Matthew E. Cramp, 1998	0.224	0.161	0.302	-5.999	0.000
Laurent Alric, 1997	0.253	0.224	0.284	-13.336	0.000
I^2 = 0.000	0.249	0.222	0.277	-14.606	0.000



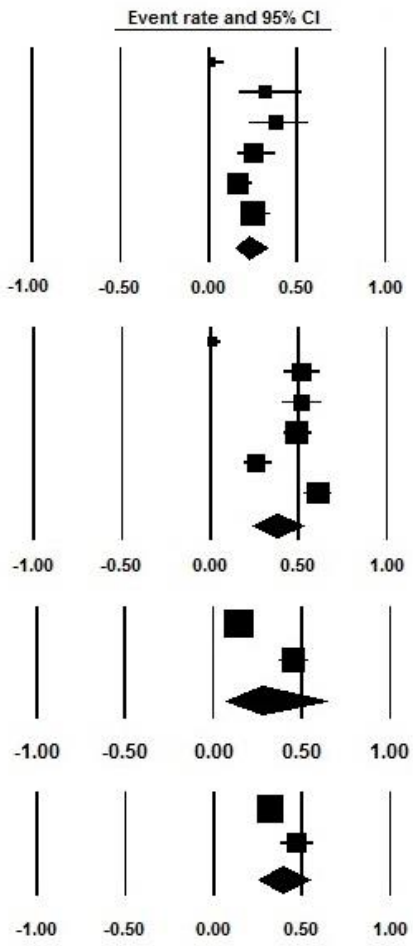
DRB1*1101 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Viviana Romero, 2008	0.103	0.039	0.243	-4.110	0.000
Laurent Alric, 1997	0.400	0.230	0.597	-0.993	0.321
Alessandra Mangia, 1999	0.214	0.134	0.326	-4.460	0.000
Alessandra Mangia, 2011	0.153	0.094	0.239	-6.098	0.000
L. Alric, 2000	0.339	0.230	0.468	-2.428	0.015
Chloe L. Thio, 2001	0.073	0.051	0.102	-13.219	0.000
I^2 = 89.172	0.187	0.100	0.323	-3.953	0.000
B. Persistent Infection					
Viviana Romero, 2008	0.240	0.172	0.324	-5.421	0.000
Laurent Alric, 1997	0.098	0.054	0.173	-6.665	0.000
Alessandra Mangia, 1999	0.164	0.127	0.211	-10.402	0.000
Alessandra Mangia, 2011	0.066	0.035	0.122	-7.674	0.000
L. Alric, 2000	0.147	0.101	0.209	-8.117	0.000
Chloe L. Thio, 2001	0.063	0.048	0.083	-17.935	0.000
I^2 = 89.675	0.119	0.074	0.187	-7.380	0.000
C. Healthy Control					
Laurent Alric, 1997	0.153	0.129	0.179	-17.440	0.000
Alessandra Mangia, 1999	0.200	0.164	0.242	-11.090	0.000
I^2 = 76.605	0.174	0.132	0.225	-9.501	0.000

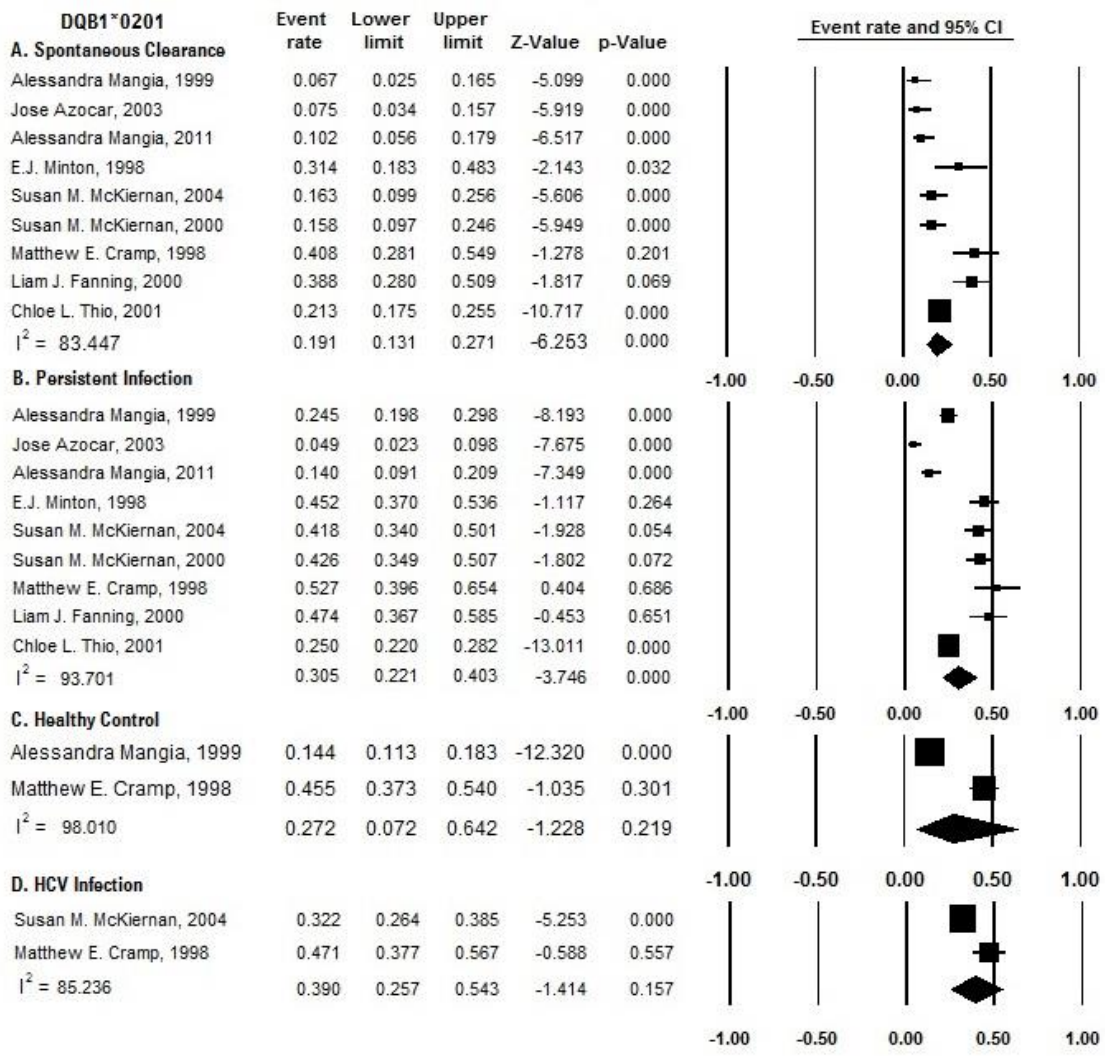


DRB1*1301 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Alessandra Mangia, 2011	0.020	0.005	0.078	-5.419	0.000
Laurent Alric, 1997	0.120	0.039	0.313	-3.237	0.001
Alessandra Mangia, 1999	0.043	0.014	0.125	-5.263	0.000
E.J. Minton, 1998	0.200	0.098	0.364	-3.281	0.001
Susan M. McKiernan, 2000	0.158	0.097	0.246	-5.949	0.000
I^2 = 71.023	0.094	0.044	0.187	-5.568	0.000
B. Persistent Infection					
Alessandra Mangia, 2011	0.051	0.025	0.104	-7.508	0.000
Laurent Alric, 1997	0.137	0.083	0.219	-6.389	0.000
Alessandra Mangia, 1999	0.037	0.021	0.065	-10.616	0.000
E.J. Minton, 1998	0.222	0.160	0.300	-6.051	0.000
Susan M. McKiernan, 2000	0.122	0.078	0.185	-7.862	0.000
I^2 = 88.814	0.098	0.050	0.184	-5.972	0.000
C. Healthy Control					
Laurent Alric, 1997	0.135	0.113	0.160	-17.953	0.000
Alessandra Mangia, 1999	0.033	0.019	0.055	-12.035	0.000
Susan M. McKiernan, 2000	0.154	0.138	0.171	-26.877	0.000
I^2 = 94.207	0.098	0.059	0.158	-7.907	0.000

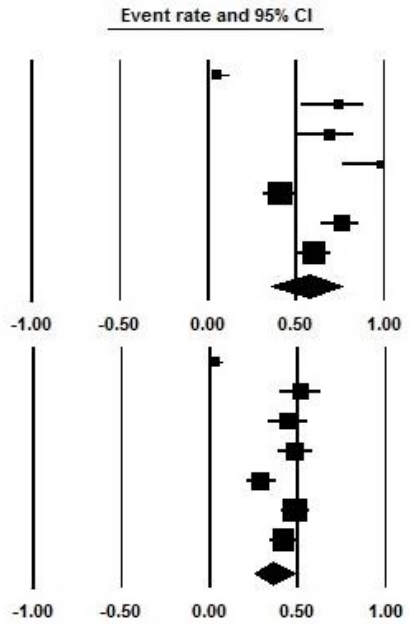


DQB1*02	Event rate	Lower limit	Upper limit	Z-Value	p-Value
A. Spontaneous Clearance					
Jose Azocar, 2003	0.013	0.002	0.083	-4.342	0.000
Laurent Alric, 1997	0.320	0.169	0.522	-1.758	0.079
Jeny R. C. Santos, 2007	0.379	0.224	0.564	-1.287	0.198
L. Alric, 2000	0.254	0.159	0.380	-3.599	0.000
Jane H Wang, 2009	0.163	0.102	0.250	-5.979	0.000
Susan M. McKiernan, 2000	0.253	0.175	0.349	-4.594	0.000
I^2 = 71.095	0.226	0.147	0.332	-4.532	0.000
B. Persistent Infection					
Jose Azocar, 2003	0.014	0.003	0.054	-5.986	0.000
Laurent Alric, 1997	0.516	0.415	0.617	0.314	0.753
Jeny R. C. Santos, 2007	0.520	0.408	0.630	0.346	0.729
L. Alric, 2000	0.490	0.413	0.568	-0.239	0.811
Jane H Wang, 2009	0.259	0.186	0.348	-4.875	0.000
Susan M. McKiernan, 2000	0.608	0.527	0.683	2.609	0.009
I^2 = 92.340	0.374	0.240	0.531	-1.583	0.113
C. Healthy Control					
Alessandra Mangia, 1999	0.144	0.113	0.183	-12.320	0.000
Matthew E. Cramp, 1998	0.455	0.373	0.540	-1.035	0.301
I^2 = 98.010	0.272	0.072	0.642	-1.228	0.219
D. HCV Infection					
Susan M. McKiernan, 2004	0.322	0.264	0.385	-5.253	0.000
Matthew E. Cramp, 1998	0.471	0.377	0.567	-0.588	0.557
I^2 = 85.236	0.390	0.257	0.543	-1.414	0.157

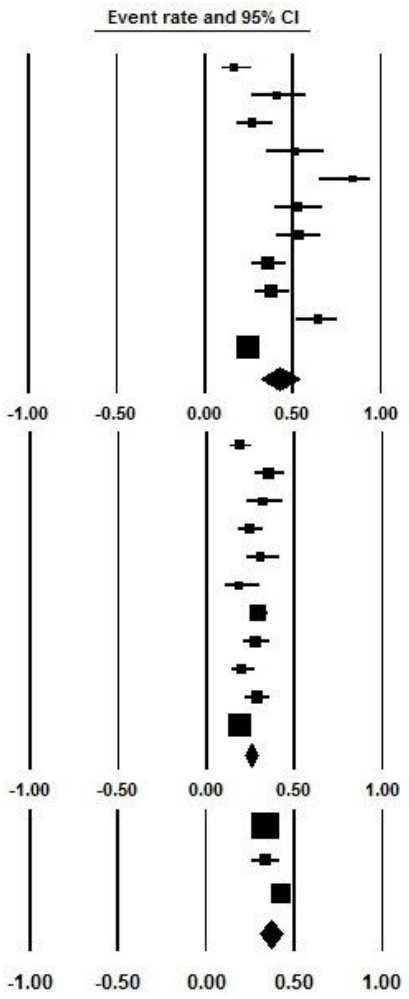



DQB1*0201
Event rate
Lower limit
Upper limit
Z-Value
p-Value
Event rate and 95% CI
A. Spontaneous Clearance
Alessandra Mangia, 1999 0.067 0.025 0.165 -5.099 0.000
Jose Azocar, 2003 0.075 0.034 0.157 -5.919 0.000
Alessandra Mangia, 2011 0.102 0.056 0.179 -6.517 0.000
E.J. Minton, 1998 0.314 0.183 0.483 -2.143 0.032
Susan M. McKiernan, 2004 0.163 0.099 0.256 -5.606 0.000
Susan M. McKiernan, 2000 0.158 0.097 0.246 -5.949 0.000
Matthew E. Cramp, 1998 0.408 0.281 0.549 -1.278 0.201
Liam J. Fanning, 2000 0.388 0.280 0.509 -1.817 0.069
Chloe L. Thio, 2001 0.213 0.175 0.255 -10.717 0.000
I² = 83.447 0.191 0.131 0.271 -6.253 0.000
B. Persistent Infection
Alessandra Mangia, 1999 0.245 0.198 0.298 -8.193 0.000
Jose Azocar, 2003 0.049 0.023 0.098 -7.675 0.000
Alessandra Mangia, 2011 0.140 0.091 0.209 -7.349 0.000
E.J. Minton, 1998 0.452 0.370 0.536 -1.117 0.264
Susan M. McKiernan, 2004 0.418 0.340 0.501 -1.928 0.054
Susan M. McKiernan, 2000 0.426 0.349 0.507 -1.802 0.072
Matthew E. Cramp, 1998 0.527 0.396 0.654 0.404 0.686
Liam J. Fanning, 2000 0.474 0.367 0.585 -0.453 0.651
Chloe L. Thio, 2001 0.250 0.220 0.282 -13.011 0.000
I² = 93.701 0.305 0.221 0.403 -3.746 0.000
C. Healthy Control
Alessandra Mangia, 1999 0.144 0.113 0.183 -12.320 0.000
Matthew E. Cramp, 1998 0.455 0.373 0.540 -1.035 0.301
I² = 98.010 0.272 0.072 0.642 -1.228 0.219
D. HCV Infection
Susan M. McKiernan, 2004 0.322 0.264 0.385 -5.253 0.000
Matthew E. Cramp, 1998 0.471 0.377 0.567 -0.588 0.557
I² = 85.236 0.390 0.257 0.543 -1.414 0.157
-1.00 -0.50 0.00 0.50 1.00
DQB1*0202
Event rate
Lower limit
Upper limit
Z-Value
p-Value
Event rate and 95% CI
A. Spontaneous Clearance
Alessandra Mangia, 2011 0.020 0.005 0.078 -5.419 0.000
Jose Azocar, 2003 0.125 0.069 0.217 -5.756 0.000
Susan M. McKiernan, 2000 0.105 0.058 0.185 -6.401 0.000
Liam J. Fanning, 2000 0.388 0.280 0.509 -1.817 0.069
I² = 91.078 0.121 0.040 0.310 -3.284 0.001
B. Persistent Infection
Alessandra Mangia, 2011 0.051 0.025 0.104 -7.508 0.000
Jose Azocar, 2003 0.146 0.097 0.213 -7.487 0.000
Susan M. McKiernan, 2000 0.243 0.181 0.319 -5.924 0.000
Liam J. Fanning, 2000 0.474 0.367 0.585 -0.453 0.651
I² = 93.851 0.191 0.082 0.384 -2.917 0.004
-1.00 -0.50 0.00 0.50 1.00

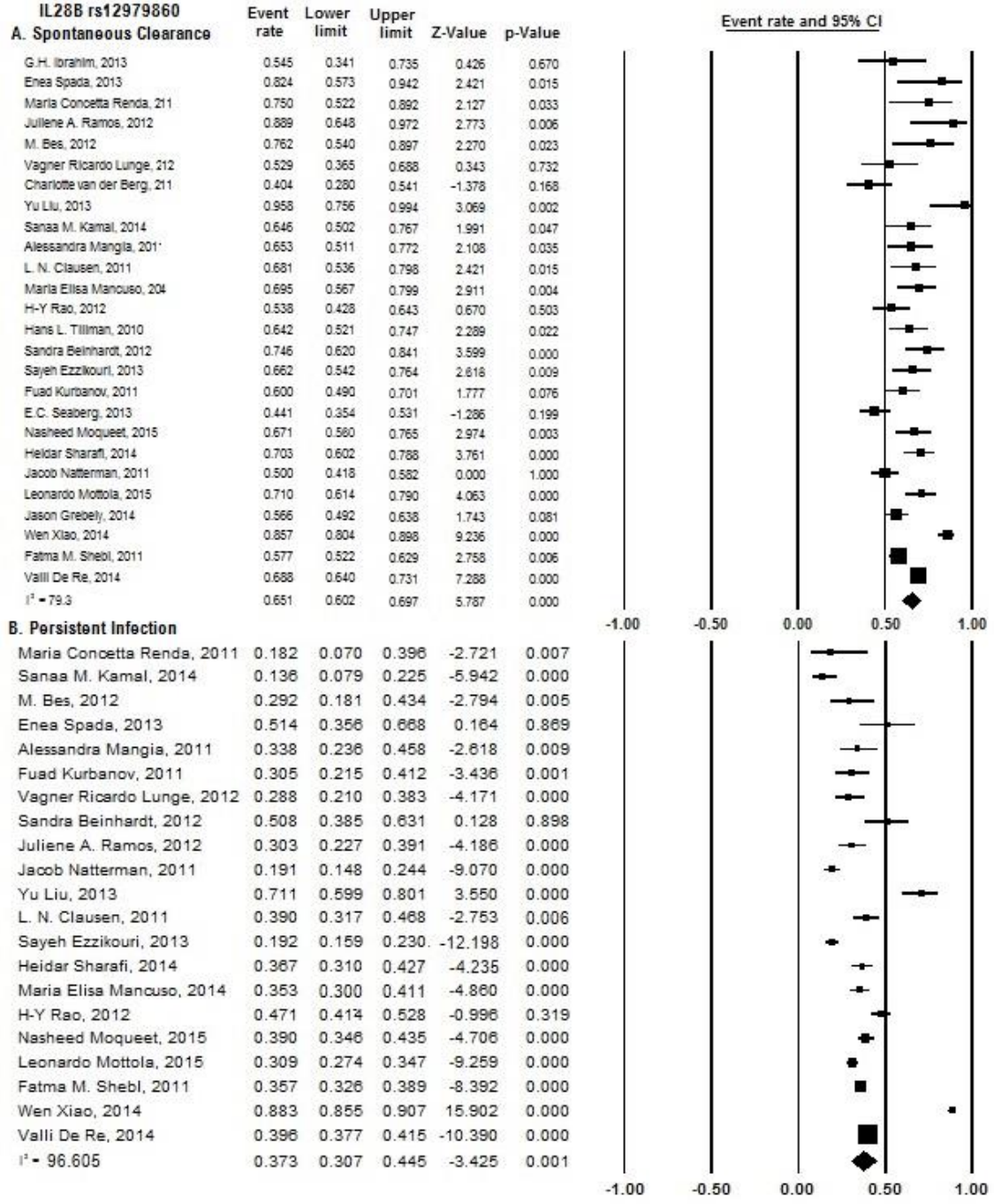
DQB1*03 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	0.050	0.019	0.126	-5.740	0.000
Rebecca A. Haris, 2008	0.739	0.528	0.878	2.193	0.028
Jeny R. C. Santos, 2007	0.690	0.503	0.830	1.989	0.047
Laurent Alric, 1997	0.981	0.756	0.999	2.753	0.006
Jane H Wang, 2009	0.408	0.316	0.508	-1.808	0.071
L. Alric, 2000	0.763	0.638	0.854	3.815	0.000
Susan M. McKiernan, 2000	0.600	0.499	0.693	1.936	0.053
I^2 = 91.233	0.574	0.359	0.764	0.664	0.507
B. Persistent Infection					
Jose Azocar, 2003	0.035	0.015	0.081	-7.305	0.000
Rebecca A. Haris, 2008	0.514	0.399	0.629	0.239	0.811
Jeny R. C. Santos, 2007	0.440	0.332	0.553	-1.037	0.300
Laurent Alric, 1997	0.484	0.383	0.585	-0.314	0.753
Jane H Wang, 2009	0.286	0.210	0.376	-4.381	0.000
L. Alric, 2000	0.484	0.407	0.562	-0.399	0.690
Susan M. McKiernan, 2000	0.419	0.342	0.500	-1.964	0.050
I^2 = 89.696	0.357	0.252	0.477	-2.325	0.020



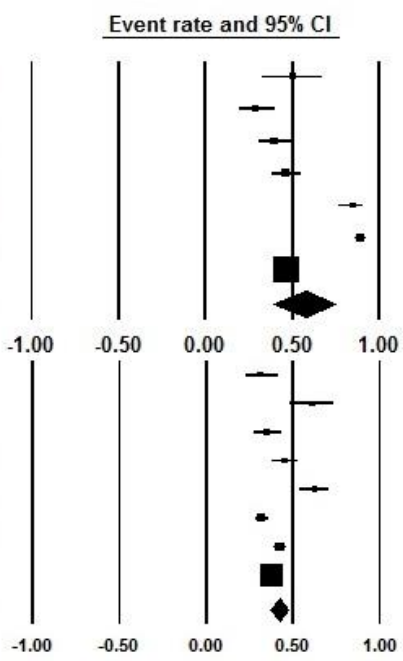
DQB1*0301 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Jose Azocar, 2003	0.163	0.097	0.260	-5.411	0.000
Viviana Romero, 2008	0.410	0.269	0.568	-1.115	0.265
Liam J. Fanning, 2000	0.269	0.176	0.387	-3.634	0.000
E.J. Minton, 1998	0.514	0.353	0.673	0.169	0.866
Laurent Alric, 1997	0.840	0.643	0.939	3.040	0.002
Matthew E. Cramp, 1998	0.531	0.392	0.665	0.428	0.668
Alessandra Mangia, 1999	0.533	0.408	0.655	0.516	0.606
Susan M. McKiernan, 2000	0.358	0.268	0.459	-2.731	0.006
Alessandra Mangia, 2011	0.378	0.287	0.477	-2.399	0.029
L. Alric, 2000	0.644	0.515	0.755	2.181	0.000
Chloe L. Thio, 2001	0.245	0.205	0.290	-9.681	0.000
I^2 = 88.845	0.425	0.322	0.535	-1.343	0.179
B. Persistent Infection					
Jose Azocar, 2003	0.188	0.132	0.260	-6.868	0.000
Viviana Romero, 2008	0.355	0.275	0.444	-3.135	0.002
Liam J. Fanning, 2000	0.321	0.227	0.431	-3.097	0.002
E.J. Minton, 1998	0.244	0.179	0.324	-5.635	0.000
Laurent Alric, 1997	0.308	0.222	0.410	-3.570	0.000
Matthew E. Cramp, 1998	0.182	0.101	0.306	-4.302	0.000
Alessandra Mangia, 1999	0.290	0.241	0.345	-6.865	0.000
Susan M. McKiernan, 2000	0.277	0.211	0.355	-5.223	0.000
Alessandra Mangia, 2011	0.199	0.140	0.274	-6.492	0.000
L. Alric, 2000	0.287	0.221	0.362	-5.166	0.000
Chloe L. Thio, 2001	0.189	0.162	0.218	-15.615	0.000
I^2 = 71.898	0.255	0.218	0.296	-10.267	0.000
C. Healthy Control					
Laurent Alric, 1997	0.338	0.306	0.371	-9.020	0.000
Matthew E. Cramp, 1998	0.336	0.261	0.420	-3.728	0.000
Alessandra Mangia, 1999	0.428	0.379	0.478	-2.833	0.005
I^2 = 79.317	0.368	0.306	0.435	-3.794	0.000



IL28B rs12979860

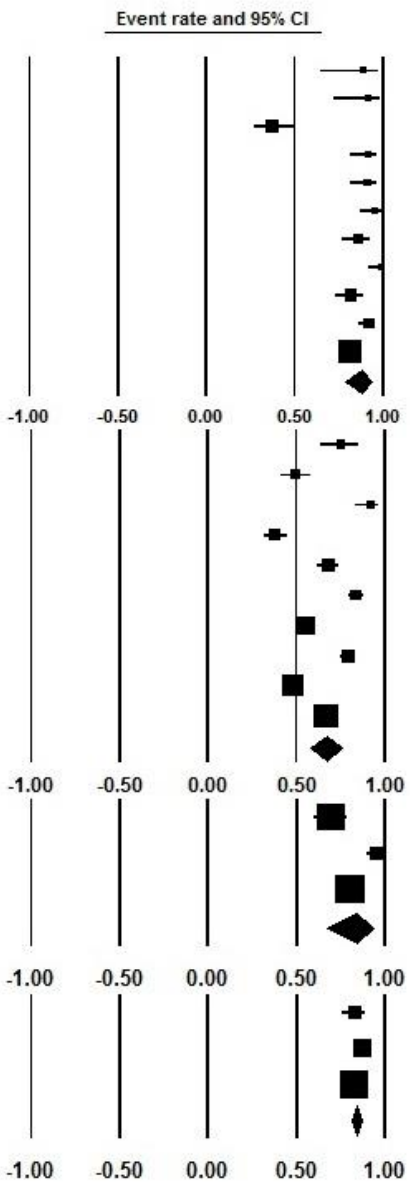


IL28B rs12979860 C. Healthy Control	Event rate	Lower limit	Upper limit	Z-Value	p-Value
M. Bes, 2012	0.500	0.328	0.672	0.000	1.000
G.H. Ibrahim, 2013	0.286	0.192	0.402	-3.463	0.001
Sandra Beinhardt, 2012	0.396	0.303	0.497	-2.026	0.043
Sayeh Ezzikouri, 2013	0.464	0.382	0.547	-0.851	0.395
Yu Liu, 2013	0.847	0.767	0.903	6.488	0.000
Wen Xiao, 2014	0.893	0.858	0.921	12.704	0.000
Valli De Re, 2014	0.462	0.436	0.488	-2.811	0.005
I^2 = 97.36	0.579	0.389	0.748	0.808	0.419
D. HCV Infection					
G.H. Ibrahim, 2013	0.312	0.226	0.413	-3.536	0.000
Enea Spada, 2013	0.611	0.476	0.731	1.619	0.105
Vagner Ricardo Lunge, 2012	0.348	0.273	0.431	-3.517	0.000
Fuad Kurbanov, 2011	0.451	0.376	0.528	-1.255	0.209
Sandra Beinhardt, 2012	0.625	0.535	0.707	2.709	0.007
E.C. Seaberg, 2013	0.318	0.280	0.359	-8.157	0.000
Jason Grebely, 2014	0.421	0.383	0.460	-3.961	0.000
Valli De Re, 2014	0.373	0.358	0.388	-15.639	0.000
I^2 = 88.149	0.419	0.365	0.474	-2.854	0.004



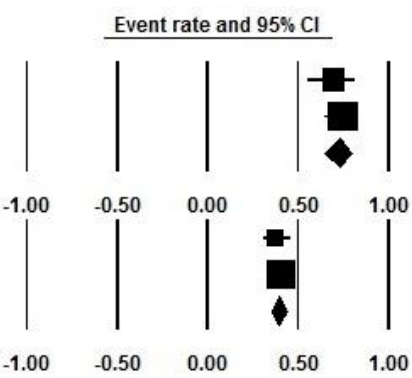
IL28B rs8099917

IL28B rs8099917 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Juliene A. Ramos, 2012	0.889	0.648	0.972	2.773	0.006
Yu Liu, 2013	0.917	0.721	0.979	3.247	0.001
Charlotte van der Berg, 2011	0.371	0.267	0.490	-2.127	0.033
Sandra Beinhardt, 2012	0.915	0.812	0.964	5.090	0.000
Sayeh Ezzikouri, 2013	0.912	0.817	0.960	5.462	0.000
Ming-Lung Yu, 2014	0.956	0.872	0.986	5.209	0.000
Nasheed Moqueet, 2015	0.861	0.766	0.921	5.605	0.000
H-Y Rao, 2012	0.987	0.915	0.998	4.316	0.000
Leonardo Mottola, 2015	0.820	0.732	0.884	5.826	0.000
Kohei Oda, 2014	0.919	0.864	0.954	8.089	0.000
Fatma M. Shebl, 2011	0.813	0.767	0.852	10.434	0.000
I^2 = 90.22	0.878	0.788	0.933	5.857	0.000
B. Persistent Infection					
Sandra Beinhardt, 2012	0.754	0.631	0.846	3.769	0.000
Juliene A. Ramos, 2012	0.496	0.412	0.581	-0.087	0.931
Yu Liu, 2013	0.921	0.835	0.964	5.775	0.000
Ming-Lung Yu, 2014	0.377	0.312	0.446	-3.437	0.001
Sayeh Ezzikouri, 2013	0.681	0.618	0.738	5.385	0.000
H-Y Rao, 2012	0.838	0.791	0.876	10.205	0.000
Nasheed Moqueet, 2015	0.554	0.508	0.599	2.322	0.020
Kohei Oda, 2014	0.790	0.745	0.830	10.150	0.000
Leonardo Mottola, 2015	0.483	0.444	0.522	-0.843	0.399
Fatma M. Shebl, 2011	0.666	0.634	0.696	9.677	0.000
I^2 = 96.258	0.669	0.574	0.752	3.404	0.001
C. Healthy Control					
Sandra Beinhardt, 2012	0.698	0.599	0.781	3.767	0.000
Yu Liu, 2013	0.955	0.896	0.981	6.673	0.000
Sayeh Ezzikouri, 2013	0.804	0.730	0.862	6.588	0.000
I^2 = 89.613	0.843	0.672	0.934	3.420	0.001
D. HCV Infection					
Sandra Beinhardt, 2012	0.833	0.756	0.890	6.571	0.000
Ming-Lung Yu, 2014	0.875	0.829	0.910	10.297	0.000
Kohei Oda, 2014	0.829	0.793	0.859	13.307	0.000
I^2 = 29.024	0.842	0.816	0.864	17.985	0.000



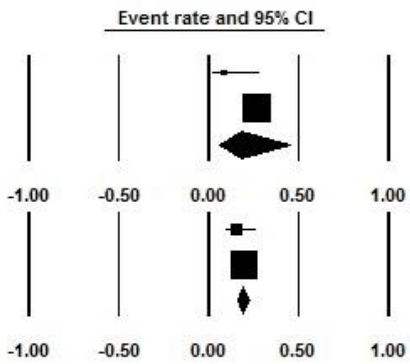
IL28B rs8103142

IL28B rs8103142 A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
L. N. Clausen, 2011	0.696	0.549	0.811	2.580	0.010
Nasheed Moqueet, 2015	0.747	0.640	0.830	4.181	0.000
I^2 = 0.000	0.727	0.643	0.798	4.874	0.000
B. Persistent Infection					
L. N. Clausen, 2011	0.376	0.304	0.454	-3.079	0.002
Nasheed Moqueet, 2015	0.403	0.359	0.448	-4.160	0.000
I^2 = 0.000	0.396	0.358	0.435	-5.142	0.000



IL18-317C

IL18-317C A. Spontaneous Clearance	Event rate	Lower limit	Upper limit	Z-Value	p-Value
Leila K. Cheikrouhou, 2011	0.083	0.021	0.279	-3.247	0.001
Ping An, 2008	0.269	0.210	0.338	-5.975	0.000
I^2 = 70.719	0.180	0.055	0.452	-2.243	0.025
B. Persistent Infection					
Leila K. Cheikrouhou, 2011	0.158	0.092	0.258	-5.321	0.000
Ping An, 2008	0.195	0.158	0.239	-10.716	0.000
I^2 = 0.000	0.189	0.155	0.228	-11.941	0.000



APPENDIX 5: INFORMATION LEAFLET

INFORMATION SHEET

Title of Project: Study to Assess the Prevalence and Risk Factors for Hepatitis C Infection in Guernsey

This study has been approved by the UCL Research Ethics Committee (Project ID Number): 6988/001

Guernsey Health and Social Services Department are conducting a survey on Hepatitis C across the island to understand how many people have the infection and how people become infected. You are being invited to take part in this research study because you could be infected with hepatitis C. If you are infected with the virus then you will be assessed by the medical team who may be able to offer you treatment to get rid of the virus. Before agreeing to participate, it is important that you read and understand the following information about the study. It is up to you to decide whether you join the study. If you agree to take part, we will then ask you to sign a consent form. You are free to withdraw at any time, without giving a reason.

Background

Hepatitis C is a virus that can infect and damage the liver. If it is not treated, it may eventually lead to liver disease, liver cancer, and liver failure. You can become infected with Hepatitis C if the blood of someone who has Hepatitis C enters your body. Most cases of hepatitis C occur in people who share needles or injecting equipment contaminated with traces of blood. In about one quarter of people who are infected the infection goes away without any treatment. Many people with Hepatitis C don't know they have it because they feel entirely well and have few or no symptoms.

Because Hepatitis C often causes no obvious symptoms, testing is highly recommended if you are at high-risk of infection. This could be because you inject drugs, or have injected drugs in the past, or if you have had long kidney dialysis. You are also at risk if you have had unprotected sexual contact with a person who has Hepatitis C. There are six types of Hepatitis C Virus (HCV) and if you are infected, it is important to know which type you have because different types need different treatments. The blood test will tell us what type of hepatitis C you have and whether you may need treatment. You can be treated for hepatitis C but until recently many people have found it difficult to take the drugs because of side effects like headache, fatigue, loss of appetite, nausea and vomiting, etc. Now we have new drugs with few side effects so we hope many more people can be treated.

This study will help Guernsey Health and Social Services Department to determine how many people may require treatment with the new drugs. Further information about Hepatitis C can be found in NHS website: http://www.nhs.uk/conditions/Hepatitis-C/Pages/Introduction.aspx

The Purpose of the Study

You are being asked to participate in a research study which will help to estimate the number of people with Hepatitis C across the Bailiwick of Guernsey.

Why have you been invited?

You have been chosen to participate in this study as you are accessing the services in Drug Concern, CDAT, Orchard Clinic or Prison and you could be infected with Hepatitis C. Approximately 250-350 people are being invited to participate in the study.

What will you be asked to do?

You will be asked to have a rapid screening test for Hepatitis C and to complete a 2 page research questionnaire with a nurse or doctor. For individuals in prison, a team from Drug Concern with assistance from Prison Healthcare Services will do the screening test. This will take about 15 minutes. The nurse or doctor will take a swab from your top and bottom gums then test it for Hepatitis C. We will tell you your test results on the same day and arrange follow up if the screening test is positive. We will also give you some general information about Hepatitis C.

What will happen if a test is positive?

It takes about 20-40 minutes to get the result of the HCV rapid screening test. We will tell you your test result after administering the questionnaire. If you have a positive test result, a blood test will be performed to confirm this result. If the blood test is positive, you will be given an opportunity to join a further study. The further study will tell us what type of hepatitis C you have and whether you may need treatment. It can be done by analysing the blood samples that previously have been collected. In addition, you will be given an opportunity to go to the Orchard Clinic for further clinical assessment and so that further management of hepatitis C infection can be discussed with you. For individuals in prison who wish to join the further study, a medical team from the Orchard Clinic with assistance from Prison Healthcare Services will collect the blood sample and undertake a clinical assessment.

Risks

In addition to your time and inconvenience, you may feel mild discomfort during the oral swab and/or when your blood sample is taken.

Benefits

You may benefit from the study because you will find out if you have hepatitis C. Your doctors will use this to decide whether you need treatment. You will also help researchers and doctors learn about how people become infected with hepatitis C. Improving your knowledge of hepatitis C will also make you aware of how to stop hepatitis C being spread in Guernsey.

Compensation

You will receive no payment or compensation for your participation.

Confidentiality

Your data and personal information will be recorded by Guernsey Health and Social Services Department. Anonymised information (with your name, address and date of birth removed) will be sent to University College London where researchers will use it to learn about hepatitis C. Your name or other identifying information will not be reported in any publications or presentations.

Voluntary

Participation is voluntary. If you choose to take part in this study, you may stop at any time during the study. You may skip any questions you do not wish to answer. If you do not want to participate in this study, it will not affect your current or future treatment in any way. You may also choose to have a clinical assessment without participating in the further study.

Contact Information

If you have any questions related to the study, please contact **Marianne Duquemin** by telephone: 01481-707707. If this study has harmed you in any way or if you wish to make a complaint about the conduct of the study you can contact UCL using the details below for further advice and information:

The Chair of UCL Research Ethics Committee,
2 Taviton Street, Room 3.03 (3rd floor)
Tel: 020 7679 8876 (ext. 28876)
Email: ethics@ucl.ac.uk

All data will be collected and stored in accordance with the Data Protection Act 1998.

Thank you for reading this information sheet and for considering takes part in this research.

Research Investigators:

Dr. Laura Shallcross, Prof. Andrew Hayward, Dr. Zisis Kozlakidis, Dewi Nur Aisyah
UCL Infectious Disease Informatics, Farr Institute of Health Informatics Research
222 Euston Road, London NW1 2DA

APPENDIX 6: INFORMED CONSENT

Patient ID:

INFORMED CONSENT FORM

Title of Project: Study to Assess the Prevalence and Risk Factors for Hepatitis C Infection in Guernsey

This study has been approved by the UCL Research Ethics Committee (Project ID Number): 6988/001

I have read or have had read to me the information provided version 2 date 18/11/2015 about the study.

Staff participating in the Guernsey Hepatitis Project have explained the study to me and answered all of my questions. I have been told about the possible risks, as well as the possible benefits, of the study.

I understand that I do not have to take part in this study and my refusal to participate or my decision to withdraw will not affect my current or future treatment in any way. I am also aware that the staff involved in doing the study may choose to stop my participation at any time.

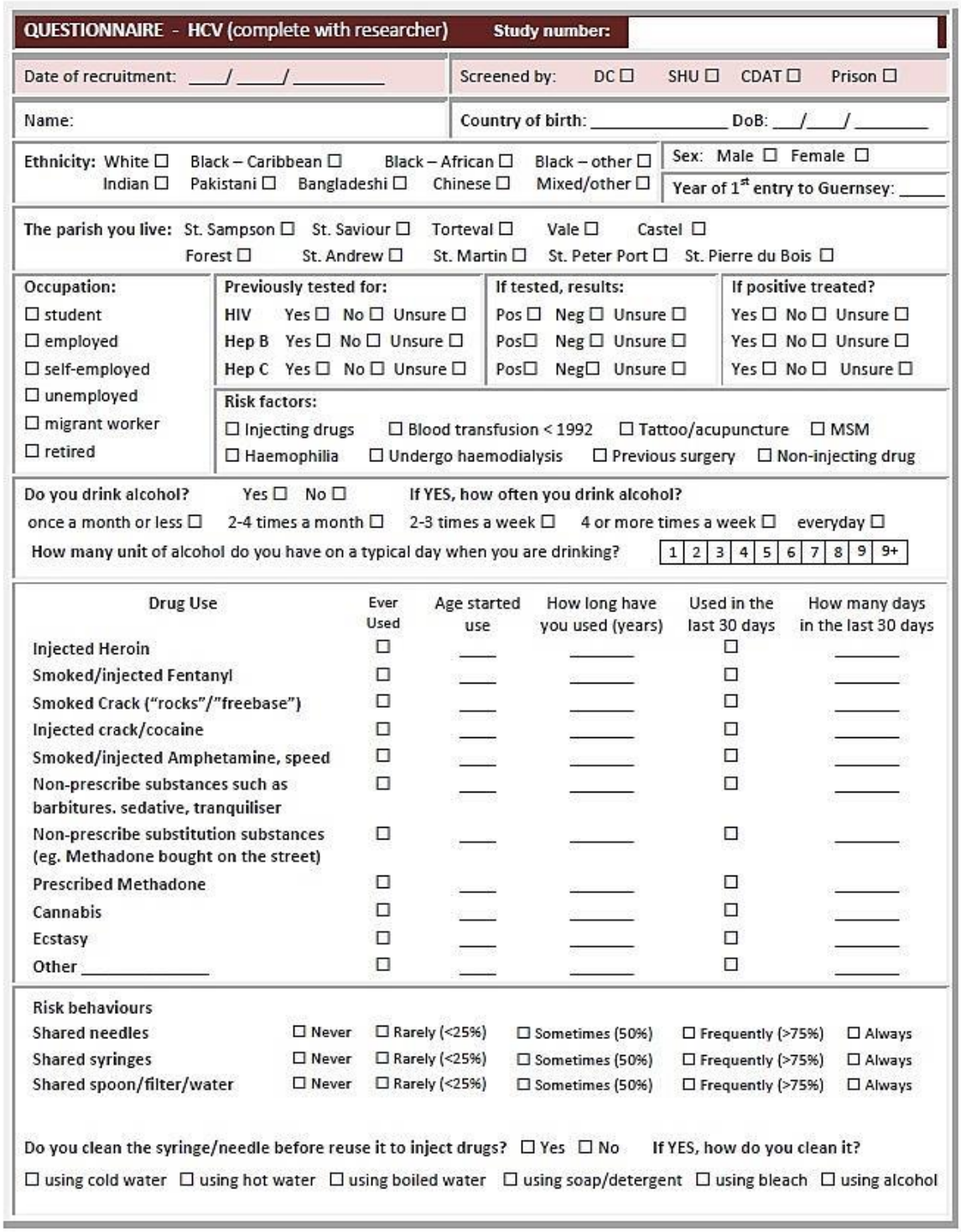
I understand that any of my information will be treated as strictly confidential and handled in accordance with the provisions of the Data Protection Act 1998.

In case I have any questions about the study, I have been told I can contact Marianne Duquemin on 01481-707707.

	Yes	No
I agree that the research project named above has been explained to me to my satisfaction and I agree to take part in this study.	☐	☐
I consent to being tested for Hepatitis C	☐	☐
I consent for my information to passed onto the Orchard Clinic so that they can contact me for further testing and management if my first test is hepatitis C positive	☐	☐
If I am already known to be hepatitis C positive, I consent for any stored blood samples that have previously be taken from me to be used for the study	☐	☐
I consent for any blood samples taken from confirmatory test may be used for further study.	☐	☐

Name of Patient	**Date**	**Signature**
Name of Person taking consent	**Date**	**Signature**
Name of Chief Investigator	**Date**	**Signature**

APPENDIX 7: QUESTIONNAIRE



QUESTIONNAIRE - HCV (complete with researcher) **Study number:**

Date of recruitment: ____/____/_____ Screened by: DC ☐ SHU ☐ CDAT ☐ Prison ☐

Name: Country of birth: _____ DoB: ___/___/_____

Ethnicity: White ☐ Black – Caribbean ☐ Black – African ☐ Black – other ☐
Indian ☐ Pakistani ☐ Bangladeshi ☐ Chinese ☐ Mixed/other ☐

Sex: Male ☐ Female ☐

Year of 1st entry to Guernsey: _____

The parish you live: St. Sampson ☐ St. Saviour ☐ Torteval ☐ Vale ☐ Castel ☐
Forest ☐ St. Andrew ☐ St. Martin ☐ St. Peter Port ☐ St. Pierre du Bois ☐

Occupation:	Previously tested for:	If tested, results:	If positive treated?
☐ student	HIV Yes ☐ No ☐ Unsure ☐	Pos ☐ Neg ☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ employed	Hep B Yes ☐ No ☐ Unsure ☐	Pos☐ Neg ☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ self-employed	Hep C Yes ☐ No ☐ Unsure ☐	Pos☐ Neg☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ unemployed			
☐ migrant worker			
☐ retired			

Risk factors:
☐ Injecting drugs ☐ Blood transfusion < 1992 ☐ Tattoo/acupuncture ☐ MSM
☐ Haemophilia ☐ Undergo haemodialysis ☐ Previous surgery ☐ Non-injecting drug

Do you drink alcohol? Yes ☐ No ☐ If YES, how often you drink alcohol?
once a month or less ☐ 2-4 times a month ☐ 2-3 times a week ☐ 4 or more times a week ☐ everyday ☐
How many unit of alcohol do you have on a typical day when you are drinking? 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 9+

Drug Use	Ever Used	Age started use	How long have you used (years)	Used in the last 30 days	How many days in the last 30 days
Injected Heroin	☐	____	_____	☐	_____
Smoked/injected Fentanyl	☐	____	_____	☐	_____
Smoked Crack ("rocks"/"freebase")	☐	____	_____	☐	_____
Injected crack/cocaine	☐	____	_____	☐	_____
Smoked/injected Amphetamine, speed	☐	____	_____	☐	_____
Non-prescribe substances such as barbitures. sedative, tranquiliser	☐	____	_____	☐	_____
Non-prescribe substitution substances (eg. Methadone bought on the street)	☐	____	_____	☐	_____
Prescribed Methadone	☐	____	_____	☐	_____
Cannabis	☐	____	_____	☐	_____
Ecstasy	☐	____	_____	☐	_____
Other _____	☐	____	_____	☐	_____

Risk behaviours

Shared needles	☐ Never	☐ Rarely (<25%)	☐ Sometimes (50%)	☐ Frequently (>75%)	☐ Always
Shared syringes	☐ Never	☐ Rarely (<25%)	☐ Sometimes (50%)	☐ Frequently (>75%)	☐ Always
Shared spoon/filter/water	☐ Never	☐ Rarely (<25%)	☐ Sometimes (50%)	☐ Frequently (>75%)	☐ Always

Do you clean the syringe/needle before reuse it to inject drugs? ☐ Yes ☐ No If YES, how do you clean it?
☐ using cold water ☐ using hot water ☐ using boiled water ☐ using soap/detergent ☐ using bleach ☐ using alcohol

Reason why sharing needle/syringe/equipment:

☐ It was not possible to get new syringe/needle

☐ Next opportunity to exchange syringes was too far away

☐ I had no money to buy new syringes or needles

☐ I was in prison and sterile syringes or needles were not available

☐ Other reason _____

Have you ever:

Been in prison in UK/Guernsey? Yes ☐ No ☐ Currently in prison ☐

How many times? 1 2 3 4 5 6 7 8 9 10 (>10times) ☐

How long? (< 1 year) ☐ 1 2 3 4 5 6 7 8 9 10 (> 10 years) ☐

Been in prison outside the UK? Yes ☐ No ☐

How long? (< 1 year) ☐ 1 2 3 4 5 6 7 8 9 10 (> 10 years) ☐

Slept rough? ☐ No ☐ Yes (< 1 year) ☐ Yes (> 1 year)

Lived in a hostel? ☐ No ☐Yes(< 1 year) 1 2 3 4 5 ☐ Yes (> 5 years)

Lived in a squat or on someone's floor or sofa? ☐ No ☐Yes(< 1 year) 1 2 3 4 5 ☐ Yes (> 5 years)

Sex partner: male ☐ female ☐ both ☐

Number of male partner in the last 12 months

0 ☐ 1 ☐ 2-5 ☐ 6-9 ☐ 10-13 ☐ >13 ☐

Number of female partner in the last 12months

0 ☐ 1 ☐ 2-5 ☐ 6-9 ☐ 10-13 ☐ >13 ☐

Participate in: Anal sex Yes ☐ No ☐

Oral sex Yes ☐ No ☐

Vaginal sex Yes ☐ No ☐

Have ever sex with someone infected with:

Hepatitis B ☐ Hepatitis C ☐ HIV ☐ STD ☐ Not sure

Have you ever sex with injecting drug users?

Yes ☐ No ☐ Unsure ☐

Have you ever paid for sex or traded sex for drugs, food, clothing, etc.?

Yes☐ No ☐

How often use condom when have sex?

Always ☐ Most of the time ☐ sometimes ☐ Not that often ☐ Never ☐

Drug Services have used in the last 6 months:

The provision of drug use equipment:

☐ Syringe & needle ☐ Sterile alcohol swabs ☐ Sterile/new filters ☐ Sterile water

☐ Crack pipe ☐ Foil ☐ Cooker/spoon

The education in correct handling of drug use equipment:

☐ Syringe & needle ☐ Sterile alcohol swabs ☐ Sterile/new filters ☐ Sterile water

☐ Crack pipe ☐ Foil ☐ Cooker/spoon

APPENDIX 8: CLINICAL ASSESSMENT FORM

QUESTIONNAIRE - HCV Genomic Study (complete by doctor) **Study number:**

Date of recruitment: ____/ _____/ _____ | Blood sample number:

Name: | Place of recruitment: Orchard Clinic ☐ Prison ☐

Street Address: _____ House #: _____ | DoB: ___/____/ _____

City: _____ Postcode: _____ | Phone number: _____

For how long have you known infected with Hepatitis C?
☐ Don't know
☐ One year
☐ 2-5 years
☐ more than 5 years

Does patient have any other significant chronic viral infection? ☐ Yes ☐ No
If yes, please specify: _____
Does the patient suffer from any other significant medical condition? ☐ Yes ☐ No
If yes, please specify: _____

Previous Test Results:		Date of latest result	Results
HCV PCR test	☐ Not done ☐ Not known	___/___/ _____	☐ Pos ☐ Neg Viral load _____ Genotype ____
Liver function test	☐ Not done ☐ Not known	___/___/ _____	☐ normal ☐ abnormal ☐ unknown
			ALT _____ Bilirubin _____ Albumin _____
Haematology test	☐ Not done ☐ Not known	___/___/ _____	INR/PTT: _____ Platelets: _____
Liver biopsy	☐ Not done ☐ Not known	___/___/ _____	☐ normal ☐ abnormal ☐ unknown
Fibroscan	☐ Not done ☐ Not known	___/___/ _____	Fibroscan score: ____ kPa range (____ - _____)

Symptoms felt by the patients:	Date of 1st onset		Date of 1st onset
☐ high temperature (38°C) or above	___/___/ _____	☐ stomach pain or diarrhoea	___/___/ _____
☐ tiredness	___/___/ _____	☐ feeling sick and vomiting	___/___/ _____
☐ loss of appetite	___/___/ _____	☐ aching joints	___/___/ _____

Liver Disease Symptoms:
Spider naevi ☐ Hepatomegaly ☐ Splenomegaly ☐ Jaundice ☐ Encephalopathy ☐
Varices ☐ Bleeding varices ☐ Liver tumor ☐ Liver palms ☐ Ascites ☐

Diagnosis: Acute hepatitis ☐ Chronic hepatitis ☐ Cirrhosis ☐ Hepatocellular carcinoma ☐ Asymptomatic ☐

Antiviral Treatment
Currently treated for Hepatitis C Yes ☐ No ☐ List of medication: _____
Date started: ___/___/____ Date finished: ___/____/_____ Weeks of treatment completed: ____ weeks
Response for the latest treatment: _____ Receive any other treatment (e.g herbal medicine) Yes ☐ No ☐
Previously treated for Hepatitis C Yes ☐ No ☐ List of medication: _____
Date started: ___/___/____ Date finished: ___/____/_____ Weeks of treatment completed: ____ weeks
Response for the latest treatment: _____ Receive any other treatment (e.g herbal medicine) Yes ☐ No ☐

Drug Allergies:	Responder Status:	Participant examination:
☐ NKDA	☐ Naive patient	Pregnant Yes ☐ No ☐ Due date:
☐ Known Drug Allergies	☐ Null responder	Obese (BMI>30) Yes ☐ No ☐ ___/____/____
Specify _____	☐ Relapser	Diabetes Yes ☐ No ☐

Sexual health examination:
☐ Syphilis (bad blood) ☐ Genital/Sex warts ☐ Trichomonas (trich)
☐ Gonorrhoea (clap) ☐ Herpes ☐ Chlamyidia
☐ Women infection in your tube/womb (PID) ☐ Men burning or drip from penis (not gonorrhoea or chlamydia)

APPENDIX 9: ETHICS APPROVAL DOCUMENTATION FROM GUERNSEY

HEALTH AND SOCIAL SERVICES
A STATES OF GUERNSEY GOVERNMENT DEPARTMENT

IJG/C5.4

Health and Social Services
Corporate Headquarters
Rue Mignot
St Andrew's, Guernsey
GY6 8TW
Tel +44 (0) 1481 725241
www.gov.gg

Dr N Brink
Consultant Virologist and Assistant Director in Medical Public Health
Health and Social Services Department
Corporate Headquarters
Rue Mignot
St Andrew's
GY6 8TW

2 October, 2015

Dear Nikki

Study to assess the prevalence and risk factors for Hepatitis C infection in Guernsey

Thank you for attending the Ethics Committee meeting on Thursday 4 June, 2015 to discuss your application for the above research project. At the meeting, the following documents were reviewed by the Ethics Committee:

a) Proforma Ethics Committee: Researcher – Nicola Brink;

b) Appendix A – Study Protocol;

c) Appendix B - Information Sheet;

d) Appendix C – Informed Consent Form;

e) Appendix D - Questionnaire;

f) CV – Nicola Brink.

Members of the Committee were pleased to have had an opportunity to meet you, to hear your summary of the proposed project and to ask questions.

At the meeting, the Ethics Committee approved the project, subject to minor changes requested during discussion, including:

i) the reference to looking at case-notes be removed;

ii) consideration be given to using an alternative font for the study document;

iii) minor amendment to Appendix D (Questionnaire).

In order to track progress of approved studies, the Ethics Committee would be grateful if you could provide regular progress reports for the project (at least annually) and / or a final report following its completion. This should be forwarded to: Ian Gaudion, Executive Assistant (Committees), Health and Social Services Department, Corporate Headquarters, Rue Mignot, St Andrew's, GY6 8TW.

The Committee and I wish you every success with your project.

Yours sincerely

Mrs Tracey McClean
Chairman, Ethics Committee

APPENDIX 10: UCL ETHICS APPROVAL

UCL RESEARCH ETHICS COMMITTEE
ACADEMIC SERVICES

23 November 2015

Dr Laura Shallcross
Farr Institute of Health Informatics Research
UCL

Dear Dr Shallcross

Notification of Ethical Approval
Project ID: 6988/001: Study to assess the prevalence and risk factors for Hepatitis C infection in Guernsey

Further to your satisfactory response to the committee's comments, I am pleased to confirm in my capacity as Chair of the UCL Research Ethics Committee (REC) that your study has been approved by the REC for the duration of the project i.e. until November 2016.

Approval is subject to the following conditions:

1. You must seek Chair's approval for proposed amendments to the research for which this approval has been given. Ethical approval is specific to this project and must not be treated as applicable to research of a similar nature. Each research project is reviewed separately and if there are significant changes to the research protocol you should seek confirmation of continued ethical approval by completing the 'Amendment Approval Request Form': http://ethics.grad.ucl.ac.uk/responsibilities.php

2. It is your responsibility to report to the Committee any unanticipated problems or adverse events involving risks to participants or others. The Ethics Committee should be notified of all serious adverse events via the Ethics Committee Administrator (ethics@ucl.ac.uk) immediately the incident occurs. Where the adverse incident is unexpected and serious, the Chair or Vice-Chair will decide whether the study should be terminated pending the opinion of an independent expert. The adverse event will be considered at the next Committee meeting and a decision will be made on the need to change the information leaflet and/or study protocol.

   For non-serious adverse events the Chair or Vice-Chair of the Ethics Committee should again be notified via the Ethics Committee Administrator (ethics@ucl.ac.uk) within ten days of an adverse incident occurring and provide a full written report that should include any amendments to the participant information sheet and study protocol. The Chair or Vice-Chair will confirm that the incident is non-serious and report to the Committee at the next meeting. The final view of the Committee will be communicated to you.

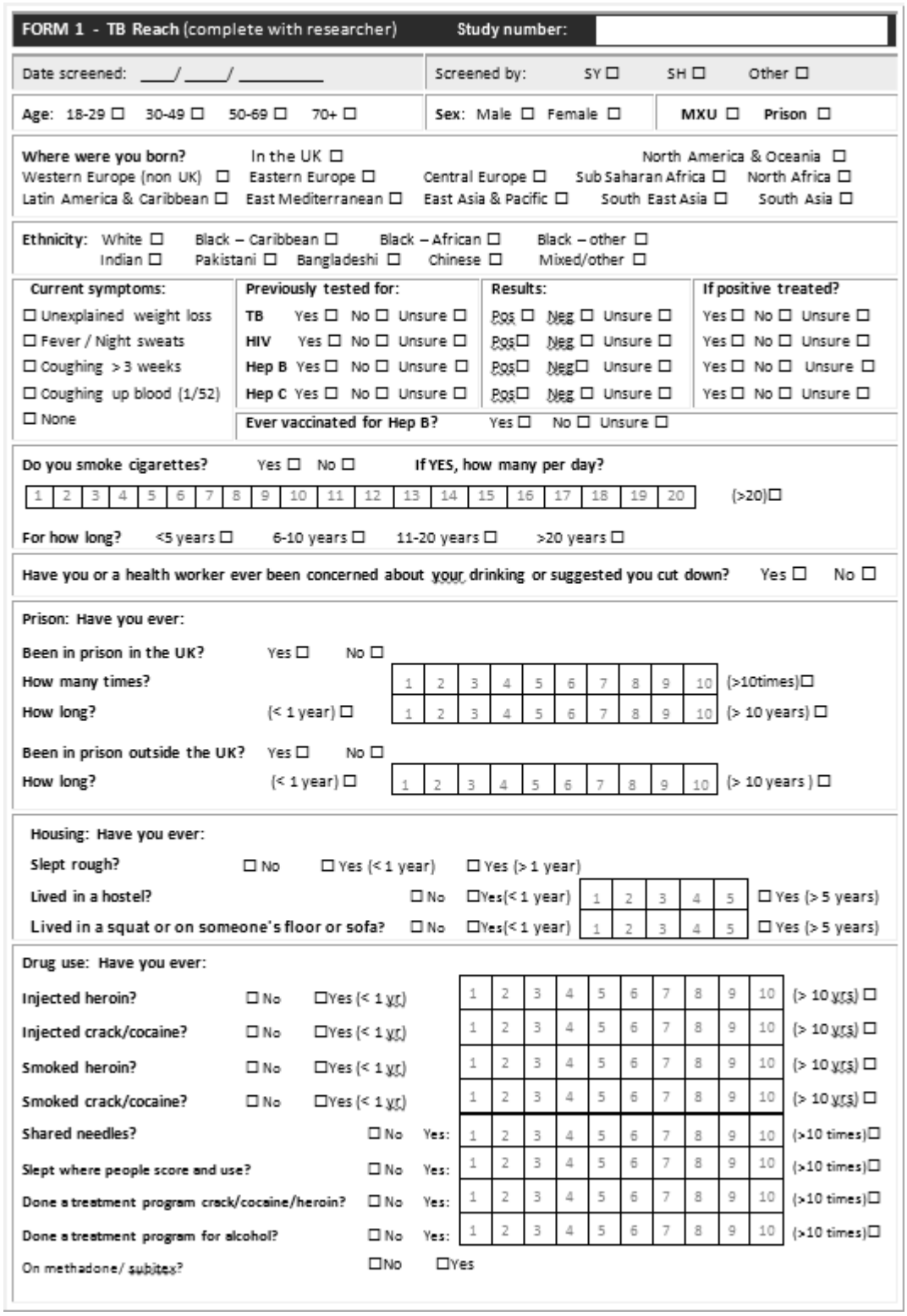
On completion of the research you must submit a very brief report of your findings/concluding comments to the Committee, which includes in particular issues relating to the ethical implications of the research.

Yours sincerely

Professor John Foreman
Chair of the UCL Research Ethics Committee

Academic Services, 1-19 Torrington Place (9th Floor),
University College London
Tel: +44 (0)20 3108 8216
Email: ethics@ucl.ac.uk
http://ethics.grad.ucl.ac.uk/

APPENDIX 11: QUESTIONNAIRE OF TB REACH STUDY



FORM 1 - TB Reach (complete with researcher) **Study number:**

Date screened: ____/ ____/ _____ Screened by: SY ☐ SH ☐ Other ☐

Age: 18-29 ☐ 30-49 ☐ 50-69 ☐ 70+ ☐ Sex: Male ☐ Female ☐ MXU ☐ Prison ☐

Where were you born? In the UK ☐ North America & Oceania ☐
Western Europe (non UK) ☐ Eastern Europe ☐ Central Europe ☐ Sub Saharan Africa ☐ North Africa ☐
Latin America & Caribbean ☐ East Mediterranean ☐ East Asia & Pacific ☐ South East Asia ☐ South Asia ☐

Ethnicity: White ☐ Black – Caribbean ☐ Black – African ☐ Black – other ☐
Indian ☐ Pakistani ☐ Bangladeshi ☐ Chinese ☐ Mixed/other ☐

Current symptoms:	Previously tested for:	Results:	If positive treated?
☐ Unexplained weight loss	TB Yes ☐ No ☐ Unsure ☐	Pos ☐ Neg ☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ Fever / Night sweats	HIV Yes ☐ No ☐ Unsure ☐	Pos☐ Neg ☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ Coughing > 3 weeks	Hep B Yes ☐ No ☐ Unsure ☐	Pos☐ Neg☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ Coughing up blood (1/52)	Hep C Yes ☐ No ☐ Unsure ☐	Pos☐ Neg ☐ Unsure ☐	Yes ☐ No ☐ Unsure ☐
☐ None	Ever vaccinated for Hep B? Yes ☐ No ☐ Unsure ☐		

Do you smoke cigarettes? Yes ☐ No ☐ If YES, how many per day?

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20

(>20)☐

For how long? <5 years ☐ 6-10 years ☐ 11-20 years ☐ >20 years ☐

Have you or a health worker ever been concerned about your drinking or suggested you cut down? Yes ☐ No ☐

Prison: Have you ever:

Been in prison in the UK? Yes ☐ No ☐
How many times? 1 2 3 4 5 6 7 8 9 10 (>10times)☐
How long? (< 1 year) ☐ 1 2 3 4 5 6 7 8 9 10 (> 10 years) ☐

Been in prison outside the UK? Yes ☐ No ☐
How long? (< 1 year) ☐ 1 2 3 4 5 6 7 8 9 10 (> 10 years) ☐

Housing: Have you ever:

Slept rough? ☐ No ☐ Yes (< 1 year) ☐ Yes (> 1 year)
Lived in a hostel? ☐ No ☐Yes(< 1 year) 1 2 3 4 5 ☐ Yes (> 5 years)
Lived in a squat or on someone's floor or sofa? ☐ No ☐Yes(< 1 year) 1 2 3 4 5 ☐ Yes (> 5 years)

Drug use: Have you ever:

Injected heroin? ☐ No ☐Yes (< 1 yr) 1 2 3 4 5 6 7 8 9 10 (> 10 yrs) ☐
Injected crack/cocaine? ☐ No ☐Yes (< 1 yr) 1 2 3 4 5 6 7 8 9 10 (> 10 yrs) ☐
Smoked heroin? ☐ No ☐Yes (< 1 yr) 1 2 3 4 5 6 7 8 9 10 (> 10 yrs) ☐
Smoked crack/cocaine? ☐ No ☐Yes (< 1 yr) 1 2 3 4 5 6 7 8 9 10 (> 10 yrs) ☐
Shared needles? ☐ No Yes: 1 2 3 4 5 6 7 8 9 10 (>10 times)☐
Slept where people score and use? ☐ No Yes: 1 2 3 4 5 6 7 8 9 10 (>10 times)☐
Done a treatment program crack/cocaine/heroin? ☐ No Yes: 1 2 3 4 5 6 7 8 9 10 (>10 times)☐
Done a treatment program for alcohol? ☐ No Yes: 1 2 3 4 5 6 7 8 9 10 (>10 times)☐
On methadone/ subitex? ☐No ☐Yes

APPENDIX 12: ICONIC SHIPPING SAMPLES STANDARD OPERATIONAL PROCEDURES

InfeCtion respONse through vIrus genomiCs

ICONIC

Shipping Samples for RNA extraction at UCLH, shipment of RNA to the Sanger Institute and full-Length Sequencing

Version 1.0
Date: 20th February 2014

Name and Title of Author: Dr Z. Kozlakidis, Project Manager

Signature of Author:

Name and Title of Approver: Prof P. Kellam, Prof. A. Hayward, ICONIC co-leads.

Signature of Approver:

Effective Date: 20th February 2014

Revision History

Version	Author	Date	Reason for Revision
1.0	Zisis Kozlakidis	20th February 2014	Initial Document
1.1	Spela Binter	25th September 2014	Addition of Sample Management processing of samples at WTSI

TABLE OF CONTENTS

1. PURPOSE

The purpose of this document is to ensure that adequate procedures are in place for the shipment of all study samples which will be analysed within the ICONIC project network.

2. RESPONSIBILITIES

2.1 The principal investigators (PIs) and/or Lead Clinical Virologist of each NHS laboratory, NHS Trust, accredited pathology service laboratory or Public Health England (collectively 'Sample Providers') contributing samples are responsible for ensuring that the relevant agreements and when necessary research ethics approvals are in place.
2.2 Sample Providers contributing samples must ensure that copies of all research ethics documentation, and any other relevant documentation (Materials Transfer Agreements (MTAs), Confidentiality agreements (CDAs), Memorandums of understanding (MOU), research collaboration agreements (RCA) relating to the samples are sent to the ICONIC project manager.
2.3 Sample Providers are responsible for delegating the responsibility for sample shipments to a member of their team. The delegate's information will be included in this document.
2.4 The delegate is to inform the project manager of any sample storage issues and of previous shipments which may have impacted on the quality of the study samples.
2.5 The delegate is responsible ensuring that the correct samples are retrieved for the shipment.
2.6 The delegate is responsible for filling in the sample shipment form (Appendix 2).
2.7 The delegate is responsible for overseeing sample shipments, ensuring that all relevant approval and shipping documentation is in place and a reliable courier service is being used.
2.8 The delegate is responsible for ensuring the samples are in the correct plates/tubes for processing at the Wellcome Trust Sanger Institute.
2.9 The Sample Providers carry overall responsibility for points 2.2 to 2.8 above.
2.10 At the receiving WTSI laboratory, the Sample Management Team are responsible for receiving the specimen. The relevant names and contact details are provided in this document.
2.11 Sample Management at the receiving laboratory must verify the sample shipment against the storage documentation received and will record storage location of samples.
2.12 A delegate at the receiving laboratory is responsible for accurate sample retrieval and record keeping, and for the timely processing of study samples. The relevant names and contact details are provided in this document.
2.13 The project manager is responsible for ensuring that an adequate framework for the processes outlined above is in place.

3. SAMPLE SHIPPING OVERVIEW

Residual diagnostic plasma samples are provided to University College London Hospitals NHS Foundation Trust (UCLH) for RNA extraction OR residual nucleic acid from diagnostic workflows are retrieved from UCLH, Barts Health NHS Trust's (Barts Health), Royal Free London NHS Foundation Trust (Royal Free) and Public Health England (PHE) for the ICONIC project. For ICONIC appropriate nucleic acid is RNA.

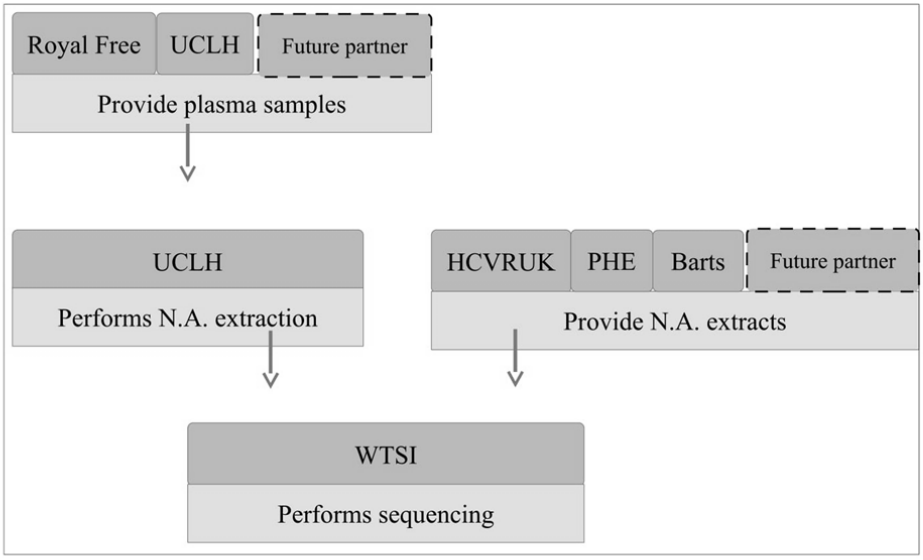
All centres will perform appropriate nucleic acid (N.A.) extraction before sending the samples to the WTSI in Cambridge, UK [see: ICONIC Sample selection and RNA extraction SOP]. The WTSI is only able to receive samples where the nucleic acid has already been extracted prior to shipment. Therefore, all samples that are to be sequenced at the WTSI must be processed before shipment.

The WTSI is not able to receive infectious materials and ONLY NUCLEIC ACID EXTRACTs can therefore be sent to this sequencing centre.

For clarity, nucleic acid is extracted from serum, plasma (for HIV, Hepatitis C virus and Measles virs), stool (for Norovirus), BAL, CTNS or NPA (for Influenza) samples, whereas all specimens received at the WTSI must be non-infectious.

Samples selected from HCV research UK Biobank (HCVRUK) and Public Health England (PHE) will be extracted as nucleic acid and stored at the original location before being shipped to WTSI.

Figure 1: Sample Shipment Overview.



4. CONTACT DETAILS

4.1 Courier contact details

All parties participating in the ICONIC project should use their preferred courier service for sample shipments. However, it is recommended that an alternative courier service is being used where difficulties with previous sample shipments were encountered.

4.2 Contact details of shipping delegates

ICONIC partner	Name	Email	Phone Number
UCLH	Bridget Ferns	r.ferns@ucl.ac.uk	+44 207 3447 8998
BartsHealth	Duncan Clark	Duncan.Clark@bartsandthelondon.nhs.uk	+44 203 246 0327
Royal Free	Daniel Webster	danielwebster@nhs.net	+44
PHE	Richard Myers	richard.myers@phe.gov.uk	+44 208 327 6614
HCRUK	John McLauchlan	john.mclauchlan@glasgow.ac.uk	+44 141 3304028

4.3 Contact details and shipping address of central sample extraction centers

4.3.1 University College London Hospital (UCLH)

Address: Clinical Microbiology and Virology
University College London Hospital
NHS Foundation Trust
60 Whitfield Street
London W1T 4EU

Key Personnel:

Name	Email	Telephone
Bridget Ferns	r.ferns@ucl.ac.uk	+44 207 3447 8998
Elisabeth Gyimah	Elizabeth.Gyimah@uclh.nhs.uk	+44 207 3447 8998
Shelley Wilson	Shelley.Wilson@uclh.nhs.uk	+44 207 3447 8998
Eleni Nastouli	Eleni.Nastouli@uclh.nhs.uk	+44 207 3447 8998

4.3.2 Bart's Health NHS Trust (Barts Health)

Address: Specialist Virology Centre
Department of Virology
Barts and the London NHS Trust
Pathology and Pharmacy Building
80 Newark Street
London E1 2ES

Key Personnel:

Name	Email	Telephone
Duncan Clark	Duncan.Clark@bartsandthelondon.nhs.uk	+44 203 246 0327

4.3.3 Royal Free London NHS Foundation Trust (Royal Free)

Address: Virology Department

Royal Free London Hospital
NHS Foundation Trust
Pond Street
London NW3 2QG

Key Personnel:

Name	Email	Telephone
Daniel Webster	danielwebster@nhs.net	+44 207 7931373

4.3.4 Public Health England (PHE)

Address: Virus Reference Department
Microbiological Services Colindale
Public Health England (PHE)
61 Colindale Avenue
Colindale, London, NW9 5EQ

Key Personnel:

Name	Email	Telephone
David Brown	david.brown@phe.gov.uk	+44 208 3276018
Richard Myers	richard.myers@phe.gov.uk	+44 208 3276614

4.3.5 Hepatitis C Virus Research UK Biobank (HCVRUK)

Address: MRC Glasgow
University of Glasgow Centre for Virus Research
8 Church Street
Glasgow, G11 5JR
Scotland

Key Personnel:

Name	Email	Telephone
John McLauchlan	john.mclauchlan@glasgow.ac.uk	+44 141 3304028
Will Irving	will.irving@nottingham.ac.uk	+44 115 8230752
Sarah McDonald	sarah.mcdonald@glasgow.ac.uk	+44 141 3304028

4.4 Contact details and shipping address of sequencing center

4.4.1 Welcome Trust Sanger Institute

Only non-infectious samples, i.e. extracts can be shipped to this sequencing centre and all key personnel listed below must be informed via email about each shipment.

Address: Sample Management team 178, ICONIC
Wellcome Trust Sanger Institute
Wellcome Trust Genome Campus
Hinxton, Cambridge, CB10 1SA, UK

Key Personnel:

Name	Email	Telephone
Paul Kellam	pk5@sanger.ac.uk	+44 1223 494940
Simon Watson	sw10@sanger.ac.uk	+44 1223 495367
Astrid Gall (for HIV)	ag8@sanger.ac.uk	+44 1223 495367
Stephanie Edwards	sf12@sanger.ac.uk	+44 1223 495367
Spela Binter	sb34@sanger.ac.uk	+44 1223 495367

Sample Management	sample-management@sanger.ac.uk	+44 1223 494990

4.5 Other contact details

Name	Role	Email	Telephone
Zisis Kozlakidis	Project Manager	z.kozlakidis@ucl.ac.uk	TBC
Paul Kellam	Executive	pk5@sanger.ac.uk	+44 1223 494940
Andrew Hayward	Executive	a.hayward@ucl.ac.uk	+44 0207 4726777
Dan Frampton	Bioinformatician	d.frampton@ucl.ac.uk	TBC
Bridget Ferns	Senior Scientist	r.ferns@ucl.ac.uk	+44 2073 4478998
TBC	Database Manager	TBC	TBC
Eleni Nastouli	Consultant Virologist	Eleni.Nastouli@uclh.nhs.uk	+44 207 3447 8998

5. SHIPPING DETAILS

All shipments should be on dry ice according to national regulations. The shipper is always responsible for the shipment.

To assist with the preparation of sample extract shipments, the shipment checklist provided as Appendix 1 should be completed.

All shipments require the shipment preparation steps outlined below:

- Samples must arrive at the WTSI no later than 16:00 on a Friday, if you have arranged courier transportation during the latter part of the week please confirm expected delivery times with your chosen courier company. Please note the sample management facility is unmanned over the weekend and any samples arriving during this time will not be accepted.
- Prior to sending samples to the Sanger Institute an electronic manifest and adhesive barcodes will be provided by WTSI Sample Management, so that sample information can be collected and corresponding plates/tubes correctly labelled..
- Adhesive barcodes will be sent by Royal Mail for domestic recipients, overseas suppliers will receive barcodes via FedEx. Included with the barcodes will be an adhesive address label for shipping purposes and a laboratory guide for preparing samples.
- The electronic manifest must be filled out with the sample details in accordance with Appendix 3. Once completed the manifest should be returned to sample-management@sanger.ac.uk.
- An excel document containing the sample details outlined in section 6 below should be provided to the receiving laboratory and the project manager in accordance with Appendix 2. The document **must not** contain any patient names.
- The appropriate paperwork for sample shipments must be prepared in advance of each shipment, including:
  - shipment inventory
  - courier reference number
  - other permits and Material Transfer Agreements (MTAs), as applicable
- Electronic copies of the paperwork must be provided to the receiving laboratory, the project manager and the database manager before each shipment. Hard copies must be retained at sending and the receiving laboratory.

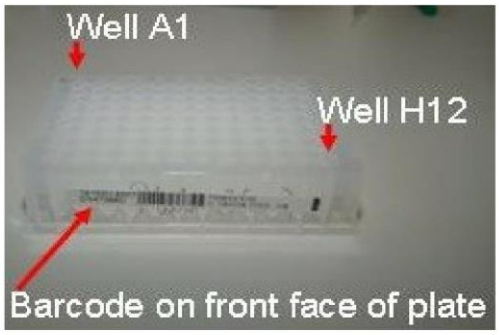
- On the day of shipment, the specimens are packaged according to national shipping regulations into appropriate shipping boxes. All samples should be shipped on dry ice.
- Upon receipt, staff at the receiving laboratory must notify the sending laboratory and the project manager of the following:
  - that the shipment has been received
  - whether the shipment was complete (as indicated on the inventory log). Any discrepancies should be noted.
  - whether or not the shipment was in good order.
- The sending laboratory is expected to respond to any sample queries within one week of receiving the shipment.

6. SAMPLE DETAILS

At the WTSI, each batch of extracts will be analysed as batches of 88 samples in a 96 well format with the final column of 8 wells left blank [These are used for QA/QC steps]. Each column (of 8 wells) must contain samples of the same viral agent, i.e. all HIV, all HCV, etc. Sample Management at WTSI require samples to be sent in the following plates or tubes:

ABgene AB_0800 0.2ml full skirted 96 well plates (volumes <100µl)
ABgene AB_0765 0.8ml Storage plate (volume >100µl)
FluidX 0.75ml 2D barcoded tube rack
FluidX 1.1ml 2D barcoded tube rack
Plates should be sealed using either a removable heat seal or adhesive seal capable of withstanding dry ice conditions.

Each plate should be identified by attaching the adhesive barcode to the middle of the front side of the plate, orientated so that well A1 is at the top left hand corner when viewed from above (See diagram below).



All collaborators should therefore ONLY send 88 samples per plate (RNA extracts), each column containing samples of the same viral agent.

An excel document (Appendix 2; "From_plates_v1_ICONIC.xls") containing the information outlined in Appendix 2 should be provided to the receiving laboratory and the individuals listed in section 4.5, other contact details. All fields should be completed to the best of the staff ability.

The file needs to be saved in the format YYYY_MM_DD_ICONIC_SITE_1 where:

- YYYY; is the year
- _; is an underscore
- MM; is the month
- _; is an underscore
- DD; is the day
- _; is an underscore
- ICONIC; is the project
- _; is an underscore
- SITE; is the unique identifier for the site or cohort or supplying centre. The unique identifiers are: UCLH or RoyalFree or BartsHealth or PHE or HCVRUK.
- _; is an underscore
- 1; is the first plate in the series from the site, cohort or supplying centre

The form should be filled in as outlined in Appendix 2.

7. APPENDICES

Appendix 1: ICONIC Shipment Checklist

Appendix 2: ICONIC Sample data form

Appendix 3: Wellcome Trust Sanger Institute Sample Management manifest

Appendix 1: ICONIC Shipment Checklist

Shipment Date:	_____	Shipment No:	_____
Courier Name:	_____	Receiving Lab Name:	_____
# of Specimens:	_____	# of Plates:	_____

A. PRIOR TO SHIPPING DAY

- ☐ 1. Have all specimens been 100% checked against Shipment Inventory
- ☐ 2. Is the material to be shipped nucleic acid?
- ☐ 3. Have all discrepancies been rectified?
- ☐ 4. Has each specimen been labelled on the tube?
- ☐ 5. Are appropriate shipping containers on-site?
- ☐ 6. Are all necessary shipping supplies on-site?
- ☐ 7. Has the courier been contacted and a tentative shipment date established?
- ☐ 8. Has the receiving lab been contacted to ensure that the tentative shipping date is acceptable?
- ☐ 9. Has an appropriate amount of dry ice been ordered for the shipment date?
- ☐ 10. Has the shipment inventory been forwarded to the recipient party and the project manager?
- 11. Has a sample log been created and forwarded to the recipient party and the project manager?

B. ON THE SHIPPING DAY

- ☐ 1. Has all shipment documentation been printed?
- ☐ 2. Are all applicable stickers/labels on the outside of the shipping container(s)?
- ☐ 3. Is sufficient amount of dry ice used for the frozen shipment?
- ☐ 4. Is the shipment packed in compliance with all national regulations?
- ☐ 5. Has all shipping documentation (including shipping inventory) been included in the shipment?
- ☐ 6. Have copies of the shipping documentation been filed appropriately?

C. UPON RECEIPT OF SHIPMENT

- ☐ 7. Has confirmation that the shipment has been received been obtained?
- ☐ 8. Were all sample queries answered?

Appendix 2: ICONIC Sample data form

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
1	sample data															
2	Well position	Sample name	Organism	GC content	Public Name	Common Name (species name)	Strain	virus	iconic identifier	plate_date	well_location	viral_load	ct	lab id	type	date_lab_receipt
3	A1	LEAVE BLANK	Human immunodeficiency Virus Ty	42%	LEAVE BLANK	Human immunodeficiency Virus Type 1	IC1413u581966-Syyyy	hiv	350	20/11/2013	b1	940	test not done	13c581966	b	19/11/2013
11	A2		Influenza A virus	43%		Influenza A virus	IC1414u135947-Syyyy	resp	572	31/01/2014	c3	test not done	25.26	14u135947	fluA	31/01/2014
19	A3		Norovirus	49%		Norovirus	IC1414U128059-Syyyy	GI	573	28/01/2014	d2	test not done	39	14U128059	Noro G1	28/01/2014
27	A4		Hepatitis C Virus	58%		Hepatitis C Virus	IC1414u128121-Syyyy	hcvg	548	29/01/2014	d8	3200000	test not done	14u128121	1a	27/01/2014
28	A5		Measles Virus	56%		Measles Virus	IC1414L134567-Syyyy	MeV	666	22/02/2014	e5	test not done	test not done	14L134567	A	24/02/2014
91	A12	PLEASE LEAVE THIS W	PLEASE LEAVE THIS WELL EMPTY		PLEASE LEAVE THIS WELL EM	PLEASE LEAVE THIS WELL EMPTY										

The attributes of the columns shown in the Excel spreadsheet with the ICONIC sample information are as follows:
Columns A – G are WTSI related, while columns H – P are hospital laboratory related.

- Column A: The position of the sequenced sample in the well at WTSI.
- Column B: To be left Blank.
- Column C: Names the virus contained in each one of the samples.
- Column D: Indicates the GC content for each virus, it remains as a constant attribute for that particular virus.
- Column E: To be left Blank.
- Column F: Names the common name of the virus in each one of the samples.
- Column G: Contains the WTSI-relevant ICONIC identifier in the format IC14xxYxxxxxx-Syyyyy, where
  - IC is the ICONIC project code
  - 14 is the two digit year
  - xxYxxxxxx is the nine digit WinPath number shown in column N [or any other equivalent laboratory pseudonymisation number]
  - - is a machine readable break and
  - Syyyyy is the Sanger added sample code generated from Sanger LIMS.
- Column H: Names the virus contained in each one of the samples, as designated in the hospital laboratory systems.
  - Designations: hiv for HIV, hcvg for HCV, resp for Influenza, GI for Norovirus samples and MeV for Measles samples respectively.

- Column I: Contains the laboratory- relevant ICONIC identifier for the sample in the form xxxxx where x are numbers and will be filled where possible in a strict ascending numerical order.
  - Designations: the ICONIC sample numbers from UCLH will start from 1, not from 00001, from Barts will start from number 20001, from the Royal Free will start from number 40001, from HCVRUK will start from 60001 and from PHE will start from 80001.
  - This designation system allows the quick visibility of the relative sample contribution from each partner and the addition of future partners using an odd-number initial, e.g. 30001, 70001.
  - The total number of samples expected in ICONIC are 20000 in total from all partners, hence any potential conflict arising from the use of an identical number for two different samples is not possible.
- Column J: Contains the date the samples were tested at each centre in the format: DD/MM/YYYY. It is not necessarily the same as the date they were received in the lab or collected in the field.
- Column K: Contains the original well location from which the sample was retrieved in the laboratory.
- Column L: Contains the Viral Load information for the sample. Not all viruses are tested for viral load, in which case 'test not done' is entered.
- Column M: Contains the RT-PCR derived Ct value or the designation 'test not done'.
- Column M: Contains the sample Lab_Id number. In the case of UCLH, the Royal Free and BartsHealth this number corresponds to the unique WinPath number for the sample and has a specific format of xxYxxxxxx, where x are numbers and Y a single letter.
  - Designations: The first two digits correspond to the two-digit year the sample was received in the lab.
  - The single letter to the Collection Centre of origin, specifically U stands for UCLH, V for BartsHealth and X for the Royal Free.
  - The six digits following the letter serve as a lab-created unique sample identifier. The 6 digit max number allows for 999999 samples in total.
  - The Winpath number is sample specific, not patient specific. However, samples of interest to ICONIC are not collected longitudinally on a routine basis, hence the potential of two Winpath numbers belonging to the same individual is very small.
- Column O: Contains the subtype of the virus for the samples as a further characterization of the infectious agent.
  - Designations: For HIV it contains the HIV clade designation, e.g. B, C, including recombinant clades. For HCV it contains the genotype designation, e.g. 1b, 2a etc. For Influenza it contains either FluA or FluB. For Norovirus it contains either NoroG1 (type 1) or NoroG11 (type 2)
- Column P: Contains the date the sample was received in the lab in the format: DD/MM/YYYY, not necessarily the date that it was physically taken in the field.

ONLY SEND 88 samples per sample box or plate for RNA extracts (last column of 8 wells left empty for QC/QA)

Appendix 3: Wellcome Trust Sanger Institute Sample Management manifest

A	B	C	D	AA	AB
Study:	XXXXSTDY				
Supplier:					
No. Plates Sent:	6				

SANGER PLATE ID	WELL	SANGER SAMPLE ID	SUPPLIER SAMPLE NAME	TAXON ID	COMMON NAME
DN329395J	A1	XXXXSTDY5573566	IC14xxYxxxxxx	11676	Human Immunodeficiency virus type 1
DN329395J	B1	XXXXSTDY5573567	IC14xxYxxxxxx	11320	Influenza A virus
DN329395J	C1	XXXXSTDY5573568	IC14xxYxxxxxx	11520	Influenza B virus
DN329395J	D1	XXXXSTDY5573569	IC14xxYxxxxxx	11103	Hepatitis C virus
DN329395J	E1	XXXXSTDY5573570	IC14xxYxxxxxx	142786	Norovirus
DN329395J	F1	XXXXSTDY5573571	IC14xxYxxxxxx	11234	Measles virus

The attributes of the columns shown in the Excel spreadsheet with the ICONIC sample information are as follows:
Columns A – C are pre-populated by the WTSI Sample Management, only columns D, AA and AB need to be filled out.

- Column A: Sanger Plate ID, DNXXXXX, will correspond to the barcodes provided. Pre-populated by WTSI Sample Management.
- Column B: The position of the sample in the well. Pre-populated by WTSI Sample Management.
- Column C: Sanger Sample ID. Pre-populated by WTSI Sample Management.
- Column D: Supplier Sample Name. Contains the WTSI-relevant ICONIC identifier in the format IC14xxYxxxxxx, where
  - IC is the ICONIC project code
  - 14 is the two digit year
  - xxYxxxxxx is the nine digit WinPath number
- WinPath number has a specific format of xxYxxxxxx, where x are numbers and Y a single letter.
  - Designations: The first two digits correspond to the two-digit year the sample was received in the lab.
  - The single letter to the Collection Centre of origin, specifically U stands for UCLH, V for BartsHealth and X for the Royal Free.
  - The six digits following the letter serve as a lab-created unique sample identifier. The 6 digit max number allows for 999999 samples in total.
  - The Winpath number is sample specific, not patient specific. However, samples of interest to ICONIC are not collected longitudinally on a routine basis, hence the potential of two Winpath numbers belonging to the same individual is very small.

- Column AA: Taxon ID.
- Column AB: Common name of the virus (Human Immunodeficiency virus type 1, Influenza A virus, Influenza B virus, Hepatitis C virus, Norovirus or Measles virus).

APPENDIX 13: UCL DOCTORAL TRAINING SCHOOL ATTENDED

Your Training

This page shows all the training you have logged.

Total Points Gained: 211

Confirmed Training

You have logged points for the following training:

Description	Points to Gain	Registration Date	
Statistics for Researchers - University College London Observational Studies: 16 and 23 Jan 2015 Start Date: 16-01-2015 End Date: 23-01-2015	2 Points	17 Nov 2014	
Statistics for Researchers - University College London Design of Experiments: 6, 13 and 27 Feb 2015 Start Date: 30-01-2015 End Date: 13-02-2015	3 Points	17 Nov 2014	
Introduction to Statistics with R - University College London 27 Feb, 2 and 3 Mar 2015 Start Date: 27-02-2015 End Date: 03-03-2015	6 Points	17 Nov 2014	
Systematic reviews and meta-analysis for healthcare and allied healthcare professionals - University College London 20 Feb 2015 Start Date: 10-02-2015 End Date: 10-02-2015	2 Points	17 Nov 2014	
Advanced and systematic literature searching using biomedical databases - University College London 26 Jan 2015 Start Date: 26-01-2015 End Date: 26-01-2015	1 Point	17 Nov 2014	
A Short Introduction to Bioinformatics - University College London 5 Dec 2014 Start Date: 05-12-2014 End Date: 05-12-2014	2 Points	17 Nov 2014	
Understanding Statistical Concepts in Research - University College London 27, 28 May, 8-10 June 2015 Start Date: 02-06-2015 End Date: 10-06-2015	9 Points	06 Jan 2015	
Hugh Kearns workshops - University College London 26 Jan 2015: The seven secrets of highly successful research students Start Date: 26-01-2015 End Date: 26-01-2015	1 Point	07 Jan 2015	
Statistical Analysis Methods for Epidemiology and Social Sciences - University College London BLOCK 1: 24 Mar-14 Apr 2015 Start Date: 24-03-2015 End Date: 14-04-2015	5 Points	22 Jan 2015	
Statistical Analysis Methods for Epidemiology and Social Sciences - University College London BLOCK 2: 21 Apr-5 May 2015 Start Date: 21-04-2015 End Date: 05-05-2015	6 Points	22 Jan 2015	
Statistical Analysis Methods for Epidemiology and Social Sciences - University College London BLOCK 4: 19 May-26 May 2015 Start Date: 19-05-2015 End Date: 26-05-2015	4 Points	22 Jan 2015	
Your PhD Part 3 - Managing and Producing Your Thesis and Reports - University College London 11 Feb 2015 Start Date: 11-02-2015 End Date: 11-02-2015	2 Points	02 Feb 2015	
Your PhD Part 2 - Management Skills for Researchers - University College London 10 Feb 2015 Start Date: 10-02-2015 End Date: 10-02-2015	2 Points	05 Feb 2015	
Practical SPSS - University College London Log points Start Date: 23-03-2015 End Date: 23-03-2015	2 Points	30 Mar 2015	
Using Word 2013 for Dissertations/Theses - University College London Log points Start Date: 19-03-2015 End Date: 19-03-2015	1 Point	30 Mar 2015	
Understanding Statistical Concepts in Research - University College London SPSS Add-on: 2-7 July 2015 Start Date: 02-07-2015 End Date: 07-07-2015	4 Points	30 Mar 2015	
Bioinformatics - University College London 22-26 June 2015 Start Date: 22-06-2015 End Date: 26-06-2015	10 Points	13 Apr 2015	
Statistical Analysis Methods for Epidemiology and Social Sciences - University College London BLOCK 3: 12 May 2015 Start Date: 12-05-2015 End Date: 12-05-2015	2 Points	05 May 2015	
Introduction to Dealing with Missing Data (2 Days) Start Date: 15-07-2015 End Date: 16-07-2015	4 Points	27 Jul 2015	Remove
Critical appraisal of a systematic review for life and medical sciences students - University College London 25 Feb 2016 Start Date: 25-02-2016 End Date: 25-02-2016	1 Point	11 Jan 2016	
Longitudinal Data Analysis - University College London 20-21 Jun 2016 Start Date: 20-06-2016 End Date: 21-06-2016	4 Points	23 May 2016	
Conference - Attendance EASL Special Conference: New Perspectives in Hepatitis C Virus Infection - The Road Map of Cure Start Date: 23-09-2016 End Date: 24-09-2016	2 Points	12 Oct 2016	Remove

Conference - Poster Presentation EASL Special Conference. Paper Title: Understanding Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis Start Date: 23-09-2016 End Date: 24-09-2016	1 Point	12 Oct 2016	Remove
Submitting an Application to an Ethics Committee for Review (per application) Study to assess the prevalence and risk factors for Hepatitis C infection in Guernsey Start Date: 23-11-2015 End Date: 31-12-2016	1 Point	12 Oct 2016	Remove
Attending Training - 3 days Pathogen Genomics Short Course at LSHTM Start Date: 14-09-2015 End Date: 16-09-2015	6 Points	12 Oct 2016	Remove
Writing a Paper for submission to a Journal - Per Paper Identification and Distribution of Pathogens in a Major Tertiary Hospital of Indonesia Start Date: 01-02-2016 End Date: 15-03-2016	4 Points	12 Oct 2016	Remove
Writing a Paper for submission to a Journal - Per Paper Phylogenetic characterisation of circulating, clinical Influenza isolates from Bali, Indonesia: preliminary report from the BaliMEI project. Start Date: 01-02-2016 End Date: 30-04-2016	4 Points	12 Oct 2016	Remove
Being Post-Graduate Representative on Staff / Student Consultative Committee Student Representative for PhD program in Institute of Health Informatics UCL Start Date: 01-09-2015 End Date: 30-09-2016	1 Point	12 Oct 2016	Remove
Conference - Poster Preparation Understanding Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis Start Date: 23-09-2016 End Date: 24-09-2016	2 Points	12 Oct 2016	Remove
Conference - Attendance Indonesian Scholars International Convention (ISIC) 2015 Start Date: 03-10-2015 End Date: 04-10-2015	2 Points	12 Oct 2016	Remove
Writing a Paper for submission to a Journal - Per Paper Assessing Hepatitis C Spontaneous Clearance and Understanding Associated Factors: A Systematic Review and Meta-Analysis Start Date: 01-07-2016 End Date: 31-10-2016	4 Points	12 Oct 2016	Remove
Contribution to Programme / Event Reviewer for Indonesian Scholars International Convention (ISIC) 2015 on population health topic Start Date: 03-10-2015 End Date: 04-10-2015	2 Points	12 Oct 2016	Remove
Contribution to Programme / Event Reviewer for Indonesian Scholars International Convention (ISIC) 2016 on population health topic Start Date: 01-10-2016 End Date: 02-10-2016	2 Points	12 Oct 2016	Remove
Attending Training - 1 day An Introduction to Infectious Disease Modelling to Inform Policy-Making Start Date: 17-03-2016 End Date: 17-03-2016	2 Points	12 Oct 2016	Remove
Writing a Paper for submission to a Journal - Per Paper Surveillance and characterisation of influenza viruses among humans in Bali, Indonesia, July 2010-June 2014 Start Date: 01-10-2015 End Date: 30-10-2016	4 Points	12 Oct 2016	Remove
Conference - Poster Preparation UCL Populations & Lifelong Health Domain Symposium - The future health of the public: Towards transdisciplinary research; "Identification and Distribution of Pathogens in a Major Tertiary Hospital of Indonesia" Start Date: 17-01-2017 End Date: 17-01-2017	2 Points	23 Jan 2017	Remove
Conference - Poster Presentation UCL Populations & Lifelong Health Domain Symposium - The future health of the public: Towards transdisciplinary research; "Identification and Distribution of Pathogens in a Major Tertiary Hospital of Indonesia" Start Date: 17-01-2017 End Date: 17-01-2017	1 Point	23 Jan 2017	Remove
Student Mentor Scheme with Volunteering Services Unit Involve in PhD buddy scheme as a mentor for Myrto Kremyda-Vlachou Start Date: 26-10-2017 End Date: 25-10-2018	5 Points	23 Jan 2017	Remove
Contribution to Programme / Event Reviewer for The 3rd Asian Academic Society International Conference (AASIC) Start Date: 13-05-2015 End Date: 14-05-2015	2 Points	23 Jan 2017	Remove
Being a Facilitator (e.g. on Facilitaions Skills Course) Systematic Review course, Farr Institute Start Date: 01-11-2016 End Date: 01-11-2016	5 Points	23 Jan 2017	Remove
Conference - Attendance Asia Pacific Association for the Study of the Liver Annual Meeting, in Shanghai, China Start Date: 15-02-2017 End Date: 19-02-2017	2 Points	01 Aug 2017	Remove
Conference - Poster Preparation APASL Annual Meeting, Host Genetic Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis, Shanghai China Start Date: 15-02-2017 End Date: 19-02-2017	2 Points	01 Aug 2017	Remove
Conference - Poster Presentation APASL Annual Meeting, Host Genetic Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis, Shanghai China Start Date: 15-02-2017 End Date: 19-02-2017	1 Point	01 Aug 2017	Remove
Conference - Attendance International Liver Congress, Amsterdam, Netherland Start Date: 19-04-2017 End Date: 23-04-2017	2 Points	01 Aug 2017	Remove
Conference - Poster Preparation ILC, Understanding Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis, Amsterdam Netherland Start Date: 19-04-2017 End Date: 23-04-2017	2 Points	01 Aug 2017	Remove
Conference - Poster Presentation ILC, Understanding Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis, Amsterdam Netherland Start Date: 19-04-2017 End Date: 23-04-2017	1 Point	01 Aug 2017	Remove
Attending Training - 5 days or more Taming The BEAST, London School of Hygiene and Tropical Medicine, London, United Kingdom Start Date: 24-07-2017 End Date: 28-07-2017	10 Points	01 Aug 2017	Remove

Attending Training - 5 days or more Introduction to Infectious Disease Modelling and its Applications, London School of Hygiene and Tropical Medicine, London, United Kingdom Start Date: 19-06-2017 End Date: 30-06-2017	10 Points	01 Aug 2017	Remove
Contribution to Programme / Event Reviewer for Indonesian Scholars International Convention (ISIC) 2017 on population health topic Start Date: 24-07-2017 End Date: 26-07-2017	2 Points	01 Aug 2017	Remove
Contribution to Programme / Event Helping and coordinating the banner activities for children during IHI Summer Party Start Date: 26-07-2017 End Date: 26-07-2017	2 Points	01 Aug 2017	Remove
Writing a Paper for submission to a Journal - Per Paper Big data analytics, infectious diseases and associated ethical impacts, submitted to Philosophy and Tecnology Journal. Start Date: 01-05-2017 End Date: 30-07-2017	4 Points	01 Aug 2017	Remove
Conference - Attendance European Scientific Conference on Applied Infectious Disease Epidemiology Start Date: 06-11-2017 End Date: 08-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Preparation European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), Title: Surveillance and characterisation of influenza viruses among patients with influenza-like illness in Bali, Indonesia, July 2010-June 2014 Start Date: 06-11-2017 End Date: 08-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Presentation European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), Title: Surveillance and characterisation of influenza viruses among patients with influenza-like illness in Bali, Indonesia, July 2010-June 2014 Start Date: 06-11-2017 End Date: 08-11-2017	1 Point	29 Nov 2017	Remove
Conference - Poster Preparation European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), Title: Identification and Distribution of Pathogens in a Major Tertiary Hospital of Indonesia Start Date: 06-11-2017 End Date: 08-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Presentation European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), Title: Identification and Distribution of Pathogens in a Major Tertiary Hospital of Indonesia Start Date: 06-11-2017 End Date: 08-11-2017	1 Point	29 Nov 2017	Remove
Conference - Poster Preparation European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), Title: Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, IDU and Prisoners in London Start Date: 06-11-2017 End Date: 08-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Presentation European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), Title: Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, IDU and Prisoners in London Start Date: 06-11-2017 End Date: 08-11-2017	1 Point	29 Nov 2017	Remove
Conference - Attendance UK Public Health Science Conference Start Date: 24-11-2017 End Date: 24-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Preparation UK Public Health Science Conference. Title: Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, IDU and Prisoners in London Start Date: 24-11-2017 End Date: 24-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Presentation UK Public Health Science Conference. Title: Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, IDU and Prisoners in London Start Date: 24-11-2017 End Date: 24-11-2017	1 Point	29 Nov 2017	Remove
Conference - Attendance UCL Global Engagement Reception Start Date: 28-11-2017 End Date: 28-11-2017	2 Points	29 Nov 2017	Remove
Conference - Poster Presentation UCL Global Engagement Reception. Title: Characterisation of circulating, clinical Influenza isolates from Bali, Indonesia: preliminary report from the BaliMEI project Start Date: 28-11-2017 End Date: 28-11-2017	1 Point	29 Nov 2017	Remove
Submitting an Application to an Ethics Committee for Review (per application) Ethics application for research title: Sensitivity and Specificity Analysis of TB DeCare (Tuberculosis Detect and Care): An Improved Automated Acid Fast Bacilli Test for Tuberculosis Detection Start Date: 16-11-2017 End Date: 16-11-2017	1 Point	29 Nov 2017	Remove
Writing a Paper for submission to a Journal - Per Paper Hepatitis C among Vulnerable Populations: A Seroprevalence Study of Homeless, People Who Inject Drugs and Prisoners in London Start Date: 10-09-2017 End Date: 30-04-2018	4 Points	01 May 2018	Remove
Writing a Paper for submission to a Journal - Per Paper Host Genetic Factors Associated with Hepatitis C Spontaneous Viral Clearance: A Meta-Analysis Start Date: 08-06-2017 End Date: 02-04-2018	4 Points	01 May 2018	Remove
Writing a Paper for submission to a Journal - Per Paper Assessing HCV Distribution among Vulnerable Populations in London Using Whole Genome Sequencing: Report from TB REACH Study Start Date: 10-01-2018 End Date: 30-04-2018	4 Points	01 May 2018	Remove
Significant Contribution to the Writing of Research Grant Applications (per Grant) TB Reach Wave 6 Application: Digital Adherence Technologies for Tuberculosis in Indonesia (DATTII) - A Cluster Randomised Controlled Trial comparing digital adherence interventions (99DOTs and VIdeo Observed Therapy) with usual care (Family DOT) in Indonesia Start Date: 30-11-2017 End Date: 25-03-2018	1 Point	01 May 2018	Remove

Co-ordinating / Convening a Conference or Workshop Workshop entitled "Defining the opportunity for mobile health technology implementation in Indonesia" Start Date: 05-01-2018 End Date: 30-04-2018	5 Points	01 May 2018	Remove
Writing a Paper for submission to a Journal - Per Paper Modelling Hepatitis C Direct Acting Antiviral (DAA) Treatment Scale Up among People Who Inject Drugs in London: Working towards WHO Incidence Elimination Target by 2030 Start Date: 10-05-2018 End Date: 20-07-2018	4 Points	11 Jun 2018	Remove
Peer Reviewing a Paper for a Journal Reviewer for BMC Public Health Journal - Factors associated with participation in counseling services for HIV out-patients of an HIV/AIDS national referral hospital in Jakarta, Indonesia: a cross sectional study Start Date: 06-06-2018 End Date: 21-06-2018	2 Points	11 Jun 2018	Remove
Peer Reviewing a Paper for a Journal Reviewer at Epidemiologic Reviews - Hepatitis C and Homelessness in the United States: A Systematic Review Start Date: 06-06-2018 End Date: 27-06-2018	2 Points	11 Jun 2018	Remove
Conference - Paper Preparation 27th International Joint Conference on Artifical Intelligence (IJCAI), Evaluating the Performance of Automated Classification of Sputum Smear Slides for TB Diagnostics, Stockholm Sweden Start Date: 13-07-2018 End Date: 18-07-2018	2 Points	07 Jul 2018	Remove