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Impact of climate change and globalization on the use of plant protection products and food safety in fresh produce

Thesis submitted in fulfillment of the requirements
for the degree of Doctor (PhD) in the Applied Biological Sciences

Dutch translation of the title:

Impact van klimaatsverandering en globalisering op het gebruik van GBM en de voedselveiligheid van verse groenten en fruit.

Cover picture: A flooded meadow in Belgium (2013), caused by heavy rainfall.

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VOORWOORD - PREFACE

Na vier lange jaren (en een lange pauze van bijna een jaar), heb ik eindelijk een degelijk antwoord klaar op de vraag: ‘Ah en waarover gaat jouw onderzoek dan wel?’. In plaats van telkens een uitleg te proberen formuleren in lekentaal en keer op keer te moeten nadenken over synoniemen voor woorden zoals globalisatie en pesticiden, kan ik nu gewoon een boekje in hun handen geven en de kous is af. Dit lijkt me wel plezant en klinkt ook eenvoudig. Het achterliggende werk, daarentegen, was helemaal niet zo eenvoudig en zou ik op mijn eentje nooit gekund hebben. Daarom mijn dankbetuiging aan iedereen die me gesteund heeft en aan iedereen die een handje (of een heleboel handen) heeft toegestoken om alles tot een goed einde te brengen. In eerste instantie, mama, een hele dikke dankjewel voor alle hulp bij de kleine en iets minder kleine problemen het voorbije jaar, het vele werk die je voor ons gedaan hebt en niet in zijn minst je controleer- en opzoekwerk die ervoor gezorgd hebben dat alles op tijd klaar geraakt is. Zonder al deze hulp, denk ik niet dat dit een afgesloten hoofdstuk geworden zou zijn. Papa, bedankt voor het nalezen en inschakelen van talloze mensen die het, volgens jou dan toch, iets beter wisten dan jijzelf. Daarbij zou ik zonder de vele babysit-uurtjes ook nooit voldoende tijd gehad hebben om rond te komen. Bij deze ook een dikke dankjewel, Lena, Geert en Tineke; door de afstand (en die eeuwige wegenwerken) was het niet altijd zo evident om in Gent te raken, maar om te komen oppassen was niks jullie teveel.

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Alle schrijfsels hier, zijn slechts een klein onderdeeltje van een groots project, met vele partners en veel nieuwe gezichten. Aangezien ik zoveel mensen heb leren kennen en om zeker niemand te vergeten, zal ik hier geen lijst met namen opsommen, met uitzondering misschien van Mieke en Liesbeth, vooral jullie hebben Veg-i-Trade gemaakt tot wat het geworden is. De ervaringen, uitstappen en vergaderingen waren altijd pico-bello georganiseerd en er was altijd ruimte voor een leuke noot. Natuurlijk hebben alle aanwezigen daar ook hun steentje aan bijgedragen, vandaar dat ik jullie allemaal heel erg dankbaar ben om van die vier jaar zo een mooie tijd te maken.

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Ilse

SAMENVATTING

Dit onderzoek kadert in het FP7 Veg-i-Trade project getiteld 'Impact van klimaatverandering en globalisering op de voedselveiligheid van verse producten – handhaving van een soevereine voedselketen'. De doelstelling van dit werk is inzicht te verkrijgen in de invloeden van internationale handel en klimaatverandering op het gebruik van gewasbeschermingsmiddelen (GBM) en het risico die de consument ten gevolge daarvan loopt bij het consumeren van verse groenten en fruit.

De handel in landbouwproducten kent reeds enkele decennia een sterke globalisering. Het voedsel dat wij op heden consumeren is afkomstig van over de hele wereld. Er wordt verwacht dat de klimaatswijziging een belangrijke impact zal hebben op de agrarische productie door invloeden op zowel bodem, plant als gewasbeschadigers. Doordat de productie van bepaalde gewassen sterk belemmerd kan worden, zullen veel mensen nog sterker afhankelijk worden van de internationale handel in voedsel. Daarenboven zullen veel landbouwers extra inputs, waaronder meststoffen en GBM, nodig hebben om hun productieniveau toch enigszins op peil te kunnen houden.

Gewasbeschermingsmiddelen behoren al lang tot de belangrijkste hulpmiddelen in de landbouw om hoge opbrengsten te verzekeren. Daarom worden ze ook, al dan niet op een correcte manier, wereldwijd toegepast. Deze GBM kunnen echter toxisch zijn voor zowel mens als milieu en het gebruik ervan dient dus strikt gereguleerd en gecontroleerd te worden. De wetgeving en de controle op de toepassing van GBM verschillen echter sterk van regio tot regio. Hierdoor kan het gebruik van producten die in het ene land vaak toegepast worden, in een naburig land verboden zijn. Dit zorgt uiteraard voor problemen wanneer geoogste producten verhandeld worden in één van deze landen. De controles op het productgebruik zijn ook niet uniform. Zo wordt in vele ontwikkelingslanden enkel toezicht gehouden op producten bestemd voor de export, terwijl op de lokale markten vaak voedsel

verkocht wordt met residuen van verboden GBM, al dan niet in hoge concentraties. Daarenboven is er een groot verschil in analytische mogelijkheden in ontwikkelingslanden, ten opzichte van bijvoorbeeld Europa en kunnen normen sterk verschillen. Hierdoor komt het vaak voor dat producten uit derdewereldlanden aan de grens geweigerd worden, terwijl deze perfect voldoen aan de wetgeving en limieten van het land van oorspong.

Niet enkel aan grenzen worden residucontroles uitgevoerd, in Europa gebeurt dit ook op vele andere plaatsen in de voedselketen, onder meer in verwerkende bedrijven. Deze bedrijven importeren vaak verse producten uit verschillende regio’s en werken ook met vele leveranciers. Aangezien het onmogelijk is alle producten te controleren op (verboden) residuen, dient een prioritering toegepast te worden. Het risico om GBM residuen te vinden op verse producten is sterk afhankelijk van het specifieke product en zijn oorsprong. Processen op het bedrijf zelf kunnen echter ook een invloed hebben op het residugehalte op deze producten bij het verlaten van het bedrijf. Een risico-gebaseerde staalname indicator kan berekend worden op basis van vijf parameters: (1) de garanties die een leverancier kan bieden; (2) het land van oorsprong; (3) het productrisico; (4) de getroffen maatregelen om kruiscontaminatie op het bedrijf te vermijden; en (5) mogelijke productverwerking op het bedrijf. Deze risico-indicator kan samen met de batchgrootte en de totale staalnamecapaciteit van het bedrijf gebruikt worden om te berekenen welke batchen het grootste risico inhouden en dus prioritair gesampled dienen te worden.

Het bereiden of verwerken van verse producten (parameter 5), heeft een invloed op de residuconcentratie in deze producten. Vanuit het oogpunt van blootstellingberekeningen, is het dan ook belangrijk te weten wat het effect van zulke bereidingsmethoden op het residugehalte van groenten en fruit precies is. Over standaard bereidingsmethoden zoals koken en stomen, is geweten dat deze de residuconcentratie over het algemeen doen dalen. Momenteel kennen grillen, wokken en bereidingen met behulp van een microgolfoven een sterke opgang. Daarom werd het effect van deze bereidingsmethodes op het residugehalte in bepaalde groenten nagegaan. Algemeen bleek dat deze methodes in bepaalde gevallen (specifieke gewas-PPP combinaties) voornamelijk een concentratie van de residuen tot gevolg hadden, terwijl koken en stomen resulteerden in een concentratiedaling. De gebruikte matrices en werkzame stoffen hadden echter een grote invloed op de resultaten. Dit was ook het geval bij het berekenen van de procesfactoren (PF). Daarom is het belangrijk om niet met algemene, maar gewas en PPP specifieke PF te werken. De blootstellingberekeningen voor wortelconsumptie, toonden dat bereidingsmethodes effectief een invloed hebben op de blootstelling van de consument. Het gebruik van conversie- en procesfactoren bij blootstellingberekeningen is daarenboven belangrijk bij het benaderen van de actuele blootstelling door middel van modellen. Dit bleek uit het feit dat de berekende blootstellingen, met inachtname van de PF en CF, soms hoger lagen dan de waarden die op een conservatievere manier berekend werden.

Op vandaag wordt bijna overal erkend dat het klimaat aan het veranderen is. Wetenschappers zijn het erover eens dat deze klimaatverandering op vele plaatsen zal resulteren in een stijgende temperatuur en extremere neerslaghoeveelheden. Het spreekt voor zich dat zulke wijzigingen een belangrijke weerslag zullen hebben op het leven op aarde en vooral op de landbouwgewassen. Aangezien landbouwgewassen rechtstreeks beïnvloed worden door omgevingsomstandigheden, zullen wijzigingen in gewasbeschermingsmiddelengebruik aan de orde zijn om mogelijke negatieve effecten op te vangen. Zo kunnen een hogere groeisnelheid, verminderde opname onder stress (bijvoorbeeld droogtestress) en verhoogde plaaggevoeligheid, de efficiëntie van een toepassing verminderen met een toename van de afhankelijkheid van GBM tot gevolg. Anderzijds kunnen een toegenomen gevoeligheid voor deze middelen, verhoogde wortelopname en infectieresistentie een lagere bespuitingintensiteit of het overschakelen naar minder fytotoxische producten vereisen. Door de sterke gewas-plaag-interactie, kunnen ook hier belangrijke gevolgen verwacht worden, voornamelijk qua timing en frequentie van applicaties. De overwintering, verspreiding, reproductie en ontwikkelingssnelheid van de meeste organismen is namelijk sterk gecorreleerd met bijvoorbeeld de temperatuur, wat in de meeste gevallen resulteert in een toename van de GBM behoefte. Deze effecten kunnen echter erg verschillen van plaag tot plaag. Wanneer in detail het effect van klimaatverandering (gebaseerd op CCSM-4 klimaatsprojecties) op het fruitmotje (*Cydia pomonella*) bestudeerd wordt, blijkt dat de temperaturen in de toekomst, theoretisch gezien, het voorkomen van meerdere generaties fruitmotjes per teeltseizoen toelaten.

Het is belangrijk te vermelden dat ook GBM en hun efficiëntie beïnvloed worden door omgevingsomstandigheden. Specifieke processen zoals vervluchtiging, runoff, uitloging, chemische en microbiële degradatie, zullen bevorderd worden als de klimaatsprojecties voor Europa kloppen. Dit blijkt eveneens uit een casestudy over het effect van een wijzigende regenintensiteit op de residuen van mancozeb en triadimenol in een appelboomgaard. Hieruit bleek dat de triadimenol SC formulering een lagere regenvastheid had dan de EC formulering. Tussen een WP en WG bestaande uit mancozeb, werd geen significant verschil in regenvastheid gevonden. Deze twee formuleringstypes waren, in tegenstelling tot de EC en SC formuleringen, wel gevoelig voor de wachttijd tussen GBM applicatie en regenval. Als een GBM toepassing uitgevoerd dient te worden kort voor een regenbui, is het dus aangewezen goed na te denken welke formulering het efficientst (meest regenvast) is in die situatie. De ecotoxiciteit van GBM is daarenboven omgevingsgerelateerd en omvat vooral een temperatuurseffect op de acute toxiciteit voor verschillende organismen. Een mogelijke oplossing hiervoor is niet in se een toename van het GBM-gebruik, maar voornamelijk een wijziging in de keuze van het type en toepassingstijdstip.

Dit onderzoek illustreert op verschillende vlakken voornamelijk de grote variatie aan effecten en invloedsfactoren op het gebruik van GBM. Daarom is het niet zinvol om de geïllustreerde effecten eenvoudigweg te veralgemenen. Dit werk kan als basis gebruikt worden om alle elementen verder uit te diepen. Het is echter aangewezen om daarbij binnen een specifieke geografische en tijdspanne te werken.

ABSTRACT

This research was conducted within the FP7 Veg-i-Trade project, titled “Impact of climate change and globalization on safety of fresh produce – Governing a supply chain of uncompromised food sovereignty”. The aim was to evaluate the effects of climate change and the global trade system on food safety, specifically caused by plant protection product (PPP) use.

For several decades, trade of agricultural produce has been characterised by a strong globalization. The food that we eat daily, originates from all over the world. Research shows that climate change will affect the agricultural productivity in many regions. Therefore, the number of people that depend on the international trade in food, will increase. In addition, many farmers will have to rely on extra inputs to maintain a certain level of production and yields, such as fertilisers and PPPs.

Plant protection products (PPPs), have long been one of the most important means in agricultural production to ascertain high yields. Here for, they are – either correctly or illegally – used worldwide. Unfortunately, PPPs can be very toxic to human beings, as well as the environment and therefore, their use should be strictly regulated and monitored. Pesticide legislation and monitoring differ strongly between regions and countries. Hence, it frequently occurs that common PPPs from one country are unauthorized in a neighbouring country. This obviously results in trade issues, when produce from one of these countries is exported to the other. Plant protection product monitoring also differs strongly between regions and countries. In many developing countries only produce intended for the export market is supervised. Hence, food with high levels or illegal PPPs can regularly be found on local markets. In addition, the analytical capacity and facilities are distinct from these in e.g. Europe and standards might differ. Produce from developing countries is, consequently,

often rejected at the border, even though it perfectly complies with the legislation and standards of the country of origin.

Residue monitoring is not only conducted at countries' borders. In Europe this is a requirement at many other stages of the food chain, for example, at processing and trading companies. These firms import fresh produce from different regions and collaborate with different suppliers worldwide. Sampling and analysing all products that enter a company is unfeasible, therefore, a prioritisation method is required. In a risk-based approach, companies can calculate a risk indicator for each batch, based on five parameters: (1) the guarantees a supplier can offer; (2) the products' country of origin; (3) the commodity risk; (4) the measures taken to avoid cross-contamination at the company; and (5) possible processing steps at the company. This risk indicator, the batch sizes and a company's total sampling capacity, can be combined to assess which batches should preferably be sampled.

Preparing or processing fresh produce (parameter 5), influences the residue concentration on a commodity. In view of exposure assessments, it is important to exactly know their effect on the residue levels in fruit and vegetables. In many studies about peeling, washing, cooking and steaming fresh produce, a decreasing effect on the residue levels is indicated. At present, grilling and using a wok pan or a microwave oven, are common food preparation methods. The effect of those modern preparation methods on the residue concentrations in certain vegetables, was assessed in this study. On average, a concentration of the residues due to these modern preparation methods and a decline due to cooking and steaming, were noted. The results were, however, strongly matrix and PPP dependent. This was also the case for the processing and conversion factors (PF, CF). Therefore, using the correct PF for a specific crop-PPP combination is important. A risk assessment for carrot consumption showed that processing influences consumer exposure. Using PF and CF is also important when the actual exposures want to be assessed by using models. This showed by calculated exposures that were higher when considering the PF and CF, than outputs from more conservative calculations.

Climate change is, at present, recognised all over the world. Scientists tend to agree that climate change will result in rising temperatures and more extreme precipitation events.

Such changes will obviously affect all life on earth and not the least the agricultural crops. Therefore, PPP use is also expected to be influenced in this situation.

Agricultural crop growth and yields strongly rely on environmental conditions. Adaptations of the current PPP use are, therefore, necessary to counteract possible negative effects. An increased growth rate, a stress related hindered PPP uptake and increased sensitivity to pests and diseases, all reduce the efficiency of PPP applications, resulting in an increased PPP dependence. On the contrary, in case of an increased sensitivity to PPPs, root uptake and infection resistance, lower application intensities are required or a shift to other less phytotoxic products is needed. Due to the strong crop-pest-interaction, climate effects will also have important consequences for the timing and frequency of PPP applications. Overwintering, spreading, reproduction and development, are population characteristics that are strongly correlated with e.g. temperature. However, the effects can differ strongly between pests, a higher need for PPPs is expected. When the effect of climate change on the codling moth (based on CCSM-4 climate projections) is studied in detail, it seems that future circumstances will (theoretically) strongly benefit its occurrence.

Realising that also PPP efficiency is influenced by environmental conditions, is important. Specific processes such as evaporation, runoff, leaching, chemical and microbial degradation will be enhanced, if the climate projections for Europe are correct. This was exemplified in a case study on the rainfastness of mancozeb and triadimenol in an apple orchard, subject to high rain intensities. A lower rain fastness of the triadimenol SC than the EC formulation was observed. There was no significant difference between the mancozeb WP and WG formulations. Those formulation types were, in contrary to the EC and SC, strongly liable to the time between application and rainfall. Therefore, different formulations should be considered when a PPP application is necessary close to a rainfall event. Plant protection product ecotoxicity is also correlated with the environmental conditions and mainly encompasses a temperature effect on the acute toxicity towards several organisms. Therefore, a change in the type of products and timing of the applications is appropriate.

This study illustrated that there is a large variation in effects and influencing factors for PPP use, so generalizing the effects is not meaningfull. However, this study can be used as a basis to elaborate on all separate subjects, for a specific geographical and time frame.

x

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List of Symbols

Symbol	Description	Unit
BW	bodyweight	kg
n	number of active ingredients	-
q	error	-
B_i	batch i	-
I_i	batch importance	-
A_i	batch fractional sampling assignment	-
c_i	commodity risk	-
s_i	supplier risk	-
v_i	batch size or volume	-
$\log P_{o/w}$	octanol water partition coefficient	-

List of Abbreviations

Abbreviation	Description
ADI	Acceptable Daily Intake
ANVISA	National Sanitary Surveillance Agency
ARfD	Acute Reference Dose
BRC	British Retail Consortium
CCSM	Community Climate System Model
CF	Conversion Factor
CS	Microencapsulated particles
DAFF	Department of Agriculture, Forestry and Fisheries
DD	Degree-Days
DoH	Department of Health
EC	Emulsifiable Concentrate
ECMP	EU Coordinated Monitoring Program
EFSA	European Food Safety Authority
EU	European Union
FAO	Food and Agriculture Organization

FASFC	Federal Agency for the Safety of the Food Chain
FTB	Fresh Trade Belgium
GAP	Good Agricultural Practices
GBM	Gewasbeschermingsmiddelen
GC-ECD	Gas Chromatography - Electron Capture Detector
GHG	GreenHouse Gas
HPLC	High Pressure Liquid Chromatography
ISO	International Standards Organization
L	Larval stage
LOD	Limit Of Detection
LOQ	Limit Of Quantification
LOR	Reporting Limit
MAPA	Ministry of Agriculture, Livestock and Food Supplies
MRL	Maximum Residue Level
P	Percentile
PARA	Program on pesticide Residue Analysis in Food
PF	Processing Factor
PNCRC	National Residue and Contaminant Control Program
PPP	Plant protection product
QMS	Quality Management Systems
RASFF	Rapid Alert System for Food and Feed
RCP	Representative Concentration Pathway
RF	Radiative Forcing
SC	Suspension Concentrate
SD	Standard Deviation
SL	Soluble Concentrate
TF-VS	Turbo Floodjet wide angle nozzle type
TMDI	Theoretical Maximum Daily Intake
TVI	Hollow cone anti-drift nozzle type
VBT	Association of Belgian Horticultural Cooperatives
WG	Water dispersible Granule
WHO	World Health Organization
WP	Wettable powder

CHAPTER 1 BACKGROUND AND GENERAL INTRODUCTION

This chapter introduces the Veg-i-Trade framework, in which all research was conducted and elaborates on chemical crop protection with plant protection products. Furthermore, the basic principles of chemical exposure assessments are introduced and the concepts of globalization and climate change are defined. Finally, the research objectives and thesis outline are presented.

1.1 VEG-I-TRADE FRAMEWORK

This study was conducted as part of the Seventh Framework Programme Veg-i-Trade project (www.veg-i-trade.org), titled 'Impact of climate change and globalization on safety of fresh produce - Governing a supply chain of uncompromised food sovereignty'. Veg-i-Trade provides platforms to identify impacts of anticipated climate change and globalization on food safety, microbiological and chemical hazards of fresh produce and derived food products. Focus lies on hazards, intervention technologies and best practices in the fresh produce chain, thus providing input of microbial and chemical risk assessment to elaborate support to risk-based sampling plans and evaluate the risks of newly identified threats as affected by the global trade system and climate change. This thesis focusses on the work about chemical hazards and more specifically plant protection products (PPPs).

1.2 CHEMICAL CROP PROTECTION

Plant protection products (PPPs) remain the most widely used means in agriculture to support food production by protecting plants or plant products against diseases, insects and other harmful organisms (Amr 1999; Vlaamse Overheid 2007). Plant protection products are used to increase the yield, improve the quality and extend the storage life of food crops (Cho *et al.* 2009; Kmellar *et al.* 2010). Furthermore, PPPs can improve the nutritional value of food and its safety (Cooper and Dobson 2008). They contribute to our food supplies, the amelioration of the public health, general hygiene and indirectly enhance the working conditions in the agricultural and horticultural sector (Vlaamse Overheid 2007). Today's PPPs are considered to be safe (Cho *et al.* 2009) and if they are applied according to Good Agricultural Practices (GAP), it is unlikely that regulatory or toxicological limits will be exceeded (Kmellar *et al.* 2010). Nevertheless, the potential risks from the use/abuse of PPPs, are among the most important environmental and health problems (Amr 1999; Wauchope *et al.* 2004; Xu *et al.* 2008). Due to misunderstanding or misuse, several potentially hazardous situations may occur during the distribution or application of PPPs and this especially in third world countries (Amr 1999). In addition, residues of these PPPs may be present on food (EFSA 2011; Tait and Bruce 2001).

1.2.1 RESIDUES IN FOOD

Food is, for consumers, the most important exposure route to PPPs, with exposures about five orders of magnitude higher than by, for example, air and drinking water (Claeys *et al.* 2011; Juraske *et al.* 2009). Fruits and vegetables make up 30 % (weight) of the food consumption and are the most frequently consumed food group (Claeys *et al.* 2011; WHO 2003). Considering that this group is also treated with the largest number of PPPs, consumption implies an uptake of higher residue levels compared to other food of plant origin (Claeys *et al.* 2011; EFSA 2011; Knežević and Serdar 2009).

1.2.2 SAMPLING AND ANALYSIS OF RESIDUES

Sampling is an essential element of residue monitoring in food. Two different methods of sampling can be distinguished: the targeted follow-up enforcement sampling and the more randomised surveillance sampling (Commission of the European communities 2008; EFSA 2011). Enforcement samples are taken in case of suspicion or as a follow-up for previously found violations and are directed to a specific grower/producer, or a specific consignment (Commission of the European communities 2008; EFSA 2010a). Surveillance samples are collected without suspicion towards a particular producer, food sample or consignment (Commission of the European communities 2008; EFSA 2011). Although the surveillance output is useful, it lacks information necessary for a proper interpretation in terms of food safety (Claeys *et al.* 2011).

A description of primary samples and the minimum size of lab samples for national monitoring is provided in, for example, the Codex Alimentarius. In case of small sized fresh products like berries, where units generally weigh less than 25 g, whole units or packages are required. For medium sized fresh products such as apples, where units generally weigh up to 250 g, whole units are sampled. A lab sample of at least 10 units, is required here. In both cases, the sample has to weigh minimum 1 kg. For large sized fresh products that weigh over 250 g such as grapes, whole bunches are sampled. In this case, the lab sample has to weigh 2 kg and contain at least 5 units. A bulk sample is then composed of 1, 3, 5, 10 or 15 primary samples, taken from an equal number of randomly chosen positions in a lot (Codex Alimentarius Commission 1999; EFSA 2010a). From this bulk sample, equal parts are selected as lab samples and parts not to be analysed, are removed (Codex Alimentarius Commission 1999; Noble 1995).

In residue analysis, the limit of quantification (LOQ), limit of detection (LOD) and measurement uncertainty are vital to make a sound interpretation of the results.

- The LOQ is the lowest concentration of analyte in a test sample that can be determined with acceptable precision (repeatability) and accuracy under the stated test conditions (Codex Alimentarius Commission 1993).
- The LOD is the smallest concentration of analyte in a test sample that can be measured with a stated probability that this analyte is present at a concentration above that in the blank sample (Codex Alimentarius Commission 1993).
- The measurement uncertainty estimate describes the range around a reported or experimental result, within which the true value is expected to lie with a defined level of probability and is influenced by external operations (e.g. sampling) and the measurement process (e.g. cleanup) (Codex Alimentarius Commission 2006).

The results of the analytical portions, considering the accuracy and precision of analysis, are used to make a decision on the compliance of the whole lot (Codex Alimentarius Commission 1999).

Monitoring data often consider a Limit Of Reporting (LOR), which is the minimum concentration (mg/kg) of a residue used for reporting purposes. Results of analyses lower than the LOR, are generally not included in monitoring reports. The LOR can be set at, for example, 10 - 20 % of the respective MRL (1.2.3), as is the case in South Africa (DAFF 2009).

1.2.3 RESIDUE ANALYSIS END POINT: THE MAXIMUM RESIDUE LEVEL (MRL)

The MRL is the maximum allowed residue of a plant protection product that can be present on a commodity when harvested (EFSA 2010a; Stephenson *et al.* 2006). This MRL is a condition for the authorization of a PPP in a specific crop and is determined in two steps (Codex Alimentarius Commission 1993; EC 2008). First, the critical GAP that equals the largest residue concentration, left on a commodity under GAP, is determined for each crop and region, based on the pre harvest interval, application dose and number of applications (De Cock 2011; MacLachlan and Hamilton 2010). Secondly, residue experiments are conducted to statistically calculate an MRL for each crop, through extrapolation. In these experiments, PPP application is consistent with the critical GAP of the respective region (De Cock 2011).

The MRL of the fresh product, taking into account concentration or dilution factors in processing, generally applies (Commission of the European communities 2008). If analysis results indicate residues above the MRL, the identity and concentration must be verified (Codex Alimentarius Commission 1999). The MRL is not a toxicological limit. Consequently, a residue concentration may be above the MRL without representing a risk to consumers (Claeys *et al.* 2011). Nevertheless, foods derived from commodities complying with the respective MRLs, are intended to be toxicologically acceptable (Codex Alimentarius Commission 1999; De Cock 2011). Therefore, a Threshold Residue Level, the theoretical maximum acceptable residue in the edible part of the crop, corresponding to an intake of 100 % of the Acute Reference Dose (ARfD), was introduced. A threshold for the Raw Agricultural Commodity (RAC) as it moves in trade, was also established and needs consideration of the processing factor (PF) for derivation (Commission of the European communities 2008; EFSA 2011). MRLs are region specific and are mostly set by national governments. The FAO, WHO and European Commission also provide MRLs for use in trade. Despite this harmonisation attempt, regional differences remain a trade barrier (De Cock 2011).

1.2.4 CHEMICAL EXPOSURE ASSESSMENT

Chemical exposure assessment is an inherent part of risk assessment. This is a process intended to estimate the risk to a given organism following exposure to a particular substance. Exposure assessment of chemicals provides an estimate of the amount of a particular substance that reaches a target organism within a specific time frame (OECD 2003). Consumers' intake of a certain substance per kg bodyweight is, therefore, compared to a toxicological limit: the Acute Reference Dose (ARfD) for acute exposure or the Acceptable Daily Intake (ADI) for chronic exposure (http://ec.europa.eu/sanco_pesticides).

- The ARfD is an estimate of the amount of substance, expressed on a body weight basis that can be ingested in a period of max. 24 h, without appreciable health risk to the consumer, based on all known facts at the time of evaluation (Solecki *et al.* 2005).
- The ADI is an estimate of the amount of substance in food that is daily ingestible over a lifetime, without appreciable risk to any consumer, based on all known facts at the time of evaluation and considering a population's sensitive groups (Renwick 2002).

Acute toxicity indicates the occurrence of adverse effects caused by a single, momentary exposure to a high dose of a substance. Chronic toxicity of a substance refers to effects that occur in the long run, or after a lifelong exposure to small amounts of that substance (Mileson *et al.* 1998). Both acute and chronic exposure, can be calculated by combining consumption data and dietary exposure models. In this thesis, consumption data from the Belgian national consumption database of 2004 (De Vriese *et al.* 2005) was used. This database contains consumption data of 3200 individuals for a period of 24 hours and two non-consecutive days. Residue data were obtained from the Belgian Federal Agency for the Safety of the Food Chain (FASFC).

The use of Conversion Factors (CFs) and Processing Factors (PFs) allows to respectively calculate the actual consumed weight and the active ingredient intake, when consuming prepared/processed foods.

1.2.4.1 Conversion factor

A conversion factor (CF) considers weight changes of the vegetables during processing. It fills the gap between food consumption data, expressed as raw products and actual food intake (weight of prepared food) and can be calculated with Equation 1.1 (Boon *et al.* 2009).

$$CF = \frac{Weight\ of\ Raw\ Agricultural\ Commodity\ (kg)}{Weight\ of\ product\ after\ processing\ (kg)} \qquad \text{Equation 1.1}$$

1.2.4.2 *Processing factor*

A Processing Factor (PF) considers changes in the amount of active ingredient that occur during preparation or processing of food. A PF is calculated by comparing the concentration of active ingredient (mg/kg vegetable) after processing with the initial concentration on those vegetables as presented in Equation 1.2 (Bonnechère 2012). This factor is useful in diet studies that consider exposure changes through home preparation of food, since the actual residue uptake can now be calculated (Balinova *et al.* 2006; Bonnechère *et al.* 2012b).

$$PF = \frac{Concentration\ of\ active\ ingredient\ in\ processed\ food\ \left(\frac{mg}{kg}\right)}{Concentration\ of\ active\ ingredient\ in\ raw\ food\ \left(\frac{mg}{kg}\right)} \qquad \text{Equation 1.2}$$

1.2.4.3 *Deterministic approach*

Deterministic modelling provides a single value for the average exposure to active ingredients due to consumption of a food (Claeys *et al.* 2011; Scientific Committee and Scientific Secretariat of the FASFC 2005). Therefore, point-estimates such as the average are multiplied for both consumption and residue levels (Equation 1.3). Variability and uncertainty are covered by calculating worst-case scenarios (P 97.5) and applying safety factors. An estimate of the total exposure to residues, is then given by a sum of the exposures to all residue/commodity combinations (Claeys *et al.* 2008).

$$Daily\ intake = \frac{Average\ daily\ consumption\left(\frac{kg\ food}{day}\right) \times Average\ residue\ level\left(\frac{mg\ a.i.}{kg\ food}\right)}{BW(kg)} \qquad \text{Equation 1.3}$$

Wherein *BW* is the bodyweight of the consumer, expressed in kg and a.i. stands for ‘active ingredient’.

Several deterministic models have been developed, of which a detailed description is given by Keikotlhaile (2011).

1.2.4.4 *Probabilistic approach*

Probabilistic modelling combines both intake and residue distributions into one risk distribution by executing a numerical or analytical technique, such as a Monte Carlo simulation (Boon *et al.* 2008; Claeys *et al.* 2011; Hamilton *et al.* 2004). The total exposure considers potential correlations between the daily consumption of different commodities.

1.3 Globalization

Globalization, or the world-wide circulation of goods, implies an increased interdependence of nations' economies and an integration of production systems.

Agriculture and the food industry are two intrinsically linked sectors that are part of a supply chain in which value-adding activities take place (Gereffi *et al.* 2001; Nomisma 2008). This agri-food chain is economically very important on a global level, because of the close link with other sectors and the high level of integration (Nomisma 2008). It is a strongly consumer-driven market, which has its consequences (De Rosa *et al.* 2008; Gereffi *et al.* 2001). In developed countries, consumers are used to a high food safety and quality. The strict regulatory systems increase production costs. Developing regions are interesting expansion areas due to lower labour costs and economies of scale. Therefore, globalisation of trade in food products increases (Nomisma 2008). High intensity inputs of fertilizers and PPPs are vital to maintain a high food productivity in these countries (Dasgupta *et al.* 2001; Jorgenson and Kuykendall 2008; Nomisma 2008; Shiva *et al.* 2002). These inputs are mainly organised on a transnational basis and supported by foreign investments (Gereffi *et al.* 2001; Jorgenson and Kuykendall 2008). Despite the positive correlation between PPP use and a region's development, human health problems and environmental damage seem prominent in developing countries (Dasgupta *et al.* 2001; Jorgenson and Kuykendall 2008).

PPP residues in food, can be seen as a traded product-based risk, which crosses national borders due to commercial transactions (Tait and Bruce 2001). Trade liberalisation and the global character of the food market, strongly increased the importance of international food safety standards (De Rosa *et al.* 2008; Shiva *et al.* 2002; Tait and Bruce 2001). Therefore, food safety policies can be used as risk regulation tools (De Rosa *et al.* 2008; Tait and Bruce 2001). This implies that, in case of public health concerns, trade barriers can be imposed. In the past, however, risk regulation has often been challenged by governments as a restriction of trade (Tait and Bruce 2001). This is still possible since (1) the high food safety and quality standards, applied in for example Europe, are not applicable in developing countries; (2) food safety policies also include socio-cultural concerns, traditions and the feasibility of controls; and (3) risk assessments are not purely objective scientific methods (De Rosa *et al.* 2008).

1.4 CLIMATE CHANGE

Climate change is defined as a change in the statistical properties of the climate system, when considered over long periods of time and regardless of cause (ENSAA 2011; IPCC 2001). The term may refer specifically to climate change caused by human activity, as opposed to changes that result from the earth's natural processes (UNFCCC 1994). Modelling of only natural processes and their effects on the climate, has not been able to explain the warming over the past century (Figure 1.1) (USGCRP 2009). Scientists assume that most of the warming since the 1950s results from an increase in concentrations of greenhouse gasses (GHG), caused by emissions from human activities (EEA 2012; Intergovernmental Panel On Climate Change 2007; NASA ; US EPA ; USGCRP 2009), as the rapid rise in global mean temperatures since 1985, of which the 14 warmest years all occurred since 1990, cannot only be ascribed to solar activity (Wang *et al.* 2009a). In addition to GHG, changes in land and water use can also influence the earth's energy balance (Harvell *et al.* 2002; Intergovernmental Panel On Climate Change 2007). The key elements in this energy balance that affect the amount of heat retained by the atmosphere, are (1) the variation in sun energy that reaches the earth; (2) changes in reflectivity of the earth's atmosphere and surface; and (3) changes in the greenhouse effect (USGCRP 2009).

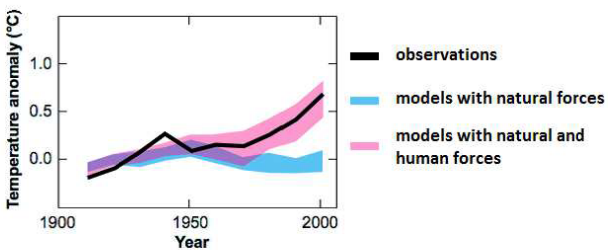


Figure 1.1: Change in surface temperature: results simulated by climate models using only natural or a combination of natural and human forces versus the average decadal observed effect. Source: Intergovernmental Panel On Climate Change (2007).

The solar intensity follows an eleven-year cycle of small alterations, due to changes that occur in the sun itself (Lean 2010; Lockwood 2009; NRC 2010; USGCRP 2009). Together with the earth's orbit and axis tilt, this can affect the amount of solar energy that reaches the earth (Jansen *et al.* 2007; NRC 2010). This sunlight can be reflected by snow, clouds and some aerosols (albedo). The albedo of the earth is 30 %, implying that 70 % of the sunlight is absorbed (NRC 2010). Absorption is enabled by dark surfaces, such as oceans, forests and soils, but also aerosols and GHG (Forster 2007; Solomon 2007; UNEP/WMO 2011; US EPA). These GHG act like a blanket, absorbing energy and inhibiting heat losses to space (EEA 2012; NASA). This so called, greenhouse effect, causes the earth to be warmer than it would naturally be (Solomon 2007; US EPA).

Greenhouse gases are emitted through natural processes, but mainly human activities (Table 1.1). Water vapour (H_2O), carbon dioxide (CO_2) and methane (CH_4) are the main GHG, of which the last two are directly linked with human activity (Solomon 2007; US EPA). The main sources of CO_2 emissions are burning of fossil fuels and deforestation, while most CH_4 emissions are caused by agriculture and land filling or waste (EEA 2012).

Table 1.1: Main characteristics of the most important greenhouse gasses (Lean 2010; NASA ; NRC 2010).

GHG	**Characteristics**
Water vapour (H_2O)	is the most abundant GHG quickly responds physically or chemically to changes in temperature acts as a feedback in the climate system
Carbon dioxide (CO_2)	is a major GHG is released through natural processes and human activities remains semi-permanently in the atmosphere does not respond physically or chemically to changes in temperature is the most important long-lived “forcing” of climate change
Methane (CH_4)	is a more powerful GHG than carbon dioxide is less abundant in the atmosphere is released through natural processes and human activities
Nitrous oxide (N_2O)	is a powerful GHG is produced by soil cultivation practices, fuel burning and nitric acid production
Chlorofluorocarbons (CFCs)	are synthetic compounds of industrial origin are able to contribute to ozone layer destruction are largely regulated in production and release

There is concordance amongst scientists that climate change encompasses atmospheric CO_2 variations, altered worldwide temperatures and precipitation variation, all directly or indirectly influencing sea levels and salinity, alterations in arable land, crop yields, changes in soil quality, nitrogen deposition and plant diversity (Fontaine *et al.* 2009; Harvell *et al.* 2002; Jackson *et al.* 2011; Miraglia *et al.* 2009; Myneni *et al.* 1997). The extensively differing impact on nature, human health and even the economy, implies that climate change is both spatially and temporally heterogeneous (EEA 2012; Fontaine *et al.* 2009; Harvell *et al.* 2002). The expected changes in climate variables are shown in Figure 1.2.

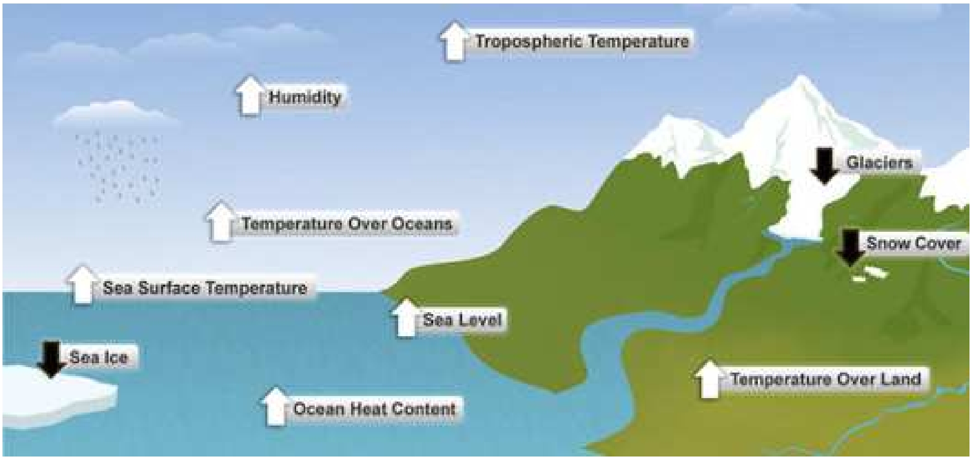


Figure 1.2: Change of the climate indicators. Source: ENSAA (2011).

A main effect of the increased GHG and CO_2 concentrations in the atmosphere, is an increased average earth temperature (EEA 2012; Fontaine *et al.* 2009; Jackson *et al.* 2011; NASA ; Scherm 2004) and the corresponding timing of seasonal climate conditions, which can vary widely among locations (Fontaine *et al.* 2009). It is expected that night time minima will increase more than daytime maximum temperatures. The effect on winter temperatures will also be larger than on those in summer (Harvell *et al.* 2002; Jackson *et al.* 2011) and regional warming is predicted to increase with latitude (Wang *et al.* 2009a). These warmer conditions can alter the intensity and/or frequency of the El Niño Southern Oscillation (Scherm 2004) and induce more evaporation and precipitation. A consequent shift in rainfall patterns causes additional flooding in wet regions and drought or desertification in dry regions (EEA 2012; Fontaine *et al.* 2009; Harvell *et al.* 2002; NASA ; Turner *et al.* 1994; Wang *et al.* 2009a). An increase in soil erosion and salinization with consequent depletion of the limited water resources can also be expected (Turner *et al.* 1994).

Short term precipitation intensity has been increasing over the last three decades. Additionally, there is evidence that the spatial extent of recently increased extreme precipitation variability and uncertainty, exemplified by changed monsoon and tropical rain patterns on one hand (Turner *et al.* 1994) and severe soil moisture deficits and frequency of short-term droughts on the other hand (Wang *et al.* 2009a), will double in the future (Costa and Soares 2009; EEA 2012; Harvell *et al.* 2002). Another extreme event is storminess, which has undergone substantial long time scale fluctuations. Storminess is also characterized by considerable seasonal and regional differences, exemplified by the storm track that seems to be shifting Poleward in winter (Fontaine *et al.* 2009; Wang *et al.* 2009a). A final effect of the GHG is warmer waters, with glacier and snow melting as a result. Warming water expands and can, alongside melting ice caps, contribute to the global mean sea level rise (EEA 2012; NASA). The last years seem to have established all-time records in terms of glacier and sea-ice melting (Wang *et al.* 2009a), with their expected effects on biodiversity.

Temperature, light and water are the key elements that control growth and development of organisms (Harvell *et al.* 2002; Rosenzweig *et al.* 2001). Consequently, biodiversity responses that depend on these environmental variables can be expected (Lepetz *et al.* 2009). For example, altered precipitation patterns and cultivation practices can create a thriving environment for insect and pathogen attacks (Roos *et al.* 2011) or corresponding advances in phenology (Fontaine *et al.* 2009). Moreover, the increasing climate variability (Wang *et al.* 2009a), can induce alterations in interspecific relationships between organisms such as competition or predation (Lepetz *et al.* 2009), possibly resulting in a decrease in food supplies and an increase in microbial and toxic contaminants in food (Hall *et al.* 2002). Consequently, not only ecosystems but also human society will be impacted through effects on energy supplies, human health and agricultural crop yields (US EPA, 2014).

These possible agricultural yield losses should be countered, hence, an effect on PPP use is expected. Additionally, climate change has a powerful effect on the environmental fate and behaviour of PPPs by altering fundamental mechanisms of partitioning between the environmental compartments (Noyes *et al.* 2009).

1.5 RESEARCH OBJECTIVES AND THESIS OUTLINE

In view of the resolute importance of PPPs in agriculture, the research objective of this thesis was to assess the effect of climate change and globalization on PPP use and human health. This general objective is treated in two more specific chapters (Chapter 2 and Chapter 5), each covering one aspect of influence. Next to these two key chapters, a few basic case studies were conducted and included, to elaborate on or exemplify some of the main issues or conclusions in the previous chapters. The structure of this thesis is shown in Figure 1.3.

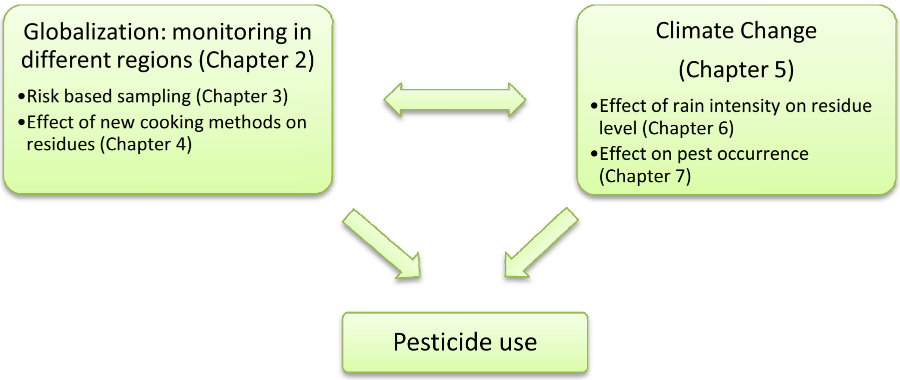


Figure 1.3: Structure of PhD thesis

In view of globalization, an evaluation of the current pesticide legislation and monitoring approach in different climate regions, is presented (Chapter 2). In Chapter 3, the development of a risk based sampling tool is explained. In view of the practical use, and validation options with Veg-i-Trade partner companies, this sampling plan was developed for processing companies. Chapter 4 elaborates on one of the parameters of the proposed sampling plan, namely the effect of processing on PPP residues in food. Chapter 5 is dedicated to a review of the influencing factors and effects of climate change on PPP use. A first case study considering the effect of climate change on PPP fate is elaborated on in Chapter 6. This chapter studies the effect of rain intensity and the possibility to counteract the effects by using different formulation types. Chapter 7 is dedicated to a second case study, considering an indirect effect of climate change on PPP use. Herein, an approach to quantitatively assess the effect of climate change on an insect pest with forecasting models, is presented. The last chapter (Chapter 8) provides a discussion and the general conclusions of this thesis, with suggestions for further research. All chapters start with a limited literature review on their specific subject.

CHAPTER 2 CURRENT PESTICIDE LEGISLATION AND MONITORING IN DIFFERENT SOCIO-ECONOMIC REGIONS

This chapter reviews the pesticide legislation and monitoring approach in different Veg-i-Trade countries. This comparison was executed in order to gain insight in pesticide issues due to globalization.

2.1 INTRODUCTION

Registration and authorisation of pesticides require many scientific studies, which is very time consuming and expensive. Given the potential risk of pesticides for public health, training programs, licensing and enforcement schemes are common post-registration activities, together with constant monitoring of pesticide use in agriculture (Amr 1999; Claeys *et al.* 2011).

Pesticide residue analyses are mostly conducted by governments for regulatory purposes (Codex Alimentarius Commission 1993). Objectives are checking for compliance with the maximum residue levels (MRLs) in food and assessing a population's exposure to pesticides (Andersen and Poulsen 2001). Another objective is meeting consumer expectations and counteract the rising pressure on quality assurance and growing numbers of residue cases (Van den Idsert 2009). Monitoring can provide indications of important problems in the supply chain (McMahon and Burke 1987; Wermund 2009) and is generally used to exclude food that contains illegal pesticide residues, from the market (Van den Idsert 2009).

The situation in the Veg-i-Trade partner countries Brazil, Egypt and South Africa, was compared to Europe and Belgium in specific. In this study, regulations, responsible authorities and monitoring programs were presented and discussed; specific authorisations in a fruit (apple) and vegetable (carrot) crop were compared; and sampling and analysis methods were described. In addition, general results of the monitoring programs were presented and if available, compared to results in traded and imported foods.

2.2 EUROPE

2.2.1 LEGAL FRAMEWORK

In the European Union (EU), Member States are obliged to comply with EU and national MRLs by at least check sampling products. In 2009, Regulation EC/1107/2009, Directive 2009/127/EC and Directive 2009/128/EC were in place. These respectively concerned the placing of pesticides on the market, requirements for application machinery and a framework for Community action to achieve a sustainable use of pesticides. Harmonisation was achieved through mutual approval in the zones of reciprocal authorisation (EFSA 2010a).

The political task of risk management lies with the European Commission, the European Parliament and the Council of the EU, responsible for ensuring a proper implementation of the EU-law. Risk assessment and communication are tasks assigned to the European Food Safety Authority (EFSA). Inspection and control are conducted by the Food and Veterinary Office, which assesses compliance of food moving within the EU, according to EC/669/2011 and directs national authorities. It also sets up standards for testing, routine procedures and reliable methods and organizes comparative tests and analysts training. The European Commission coordinates the Rapid Alert System for Food and Feed (RASFF) that notifies international bodies of food safety emergencies. Consumer protection through food safety risk assessment, management and communication resides within the jurisdiction of national governments (BfR 2011).

2.2.2 MONITORING PROGRAMS

2.2.2.1 EU COORDINATED PESTICIDE MONITORING PROGRAM

In 1996 an EU coordinated monitoring program (ECMP) was initiated, to provide statistically representative data regarding pesticide residues in food, in order to estimate the actual dietary pesticide exposure throughout Europe (Commission of the European communities 2008; EFSA 2010a). For that, a rolling program covering major pesticide-commodity combinations was developed. Commodity selection considered the major components of the WHO standard European diet and rules for monitoring provisions of Regulation EC/645/2000 (Commission of the European communities 2008). In 2008, the commodities covered by the ECMP contributed for approximately 30% of the total food intake, excluding products of animal origin (EFSA 2010a).

2.2.2.2 National monitoring programs

National monitoring programs serve to ensure compliance with food legislation and are complementary to the controls performed in the context of the ECMP (EFSA 2011; Keikotlhaile 2011). Although the provisions on national monitoring programs are defined in the MRL Regulation (EFSA 2010a), Member States are free to decide on their design (EFSA 2011). In most cases the national program includes samples for the ECMP with an additional range of authorised substances on locally consumed materials (Andersen and Poulsen 2001).

2.2.2.3 Monitoring of traded and imported commodities

MRL analyses of imported commodities have been disquieting in the past (Malcorps 2002). Therefore, import tolerances on maximum levels of pesticide residues in foods, set in Regulation No. 396/2005/EG, remained valid for products originating from non-EU countries, even though MRL harmonization took place in the EU (De Cock 2011). The currently required phytosanitary document replaces former import certificates and further enhances import control harmonisation in the different Member States (DG Controlebeleid 2005). This regulation additionally permits free circulation within the EU, provided that there is compliance with the MRLs (Codex Alimentarius Commission 2009).

2.2.3 Results of the monitoring program

2.2.3.1 General results

In the ECMP of 2006, 769 different active ingredients were sought for in surveillance samples, ranging from 45 till 683 per country. In 4 % of the samples with detectable residues (46 %), an MRL was exceeded (not yet harmonised at that time). When considering fruit and vegetable samples only, 49 % contained detectable residues of which 5 % exceeded an MRL (3 % EC-MRLs). In addition, residues were detected more often in fruit (68 %) than in vegetables (21 %) (EFSA 2011). Overall, a decreasing trend in MRL violations, ranging from 0 to 5 % in the reporting countries, was noted for all commodities, except wheat (Commission of the European communities 2008; EFSA 2011). Considering pesticide/crop combinations, MRL violations were most frequently detected with ethefon in figs, tetramethrin and nicotine in wild mushrooms and dithiocarbamates in passion fruits (Commission of the European communities 2008).

2.2.3.2 PESTICIDE RESIDUES IN TRADE

In the 2008 ECMP, imported products accounted for 22 % of all samples (EFSA 2011). MRL violations occurred in 2 % of the EU samples and 8 % of imports, while in 2009 this was respectively 2 and 7 % (Commission of the European communities 2008; EFSA 2010a; EFSA 2011). Within the EU the highest percentage of MRL violating samples was identified on products originating from Cyprus, Portugal, Belgium and Lithuania. Most non-conformities in surveillance samples were found in processed vegetables (5 %), followed by unprocessed other plant products (4 %). The MRL violation rate in enforcement samples was logically higher (maximum 24 % in unprocessed vegetables) (EFSA 2011).

2.3 BELGIUM

2.3.1 REGULATIONS AND RESPONSIBLE AUTHORITIES

The Royal Decree on self-control, registration and traceability in the food chain, published in the Belgian Bulletin of Acts, Orders and Decrees (14/11/2003), gives liability to the Food Agency to require registration of products and check for compliance with the law.

The Residue Control Service was founded in 1978 as a voluntary safety system for auction vegetables and fruits and checked for "environmental friendly production" and residue tolerances (Vlaamse Overheid 2007). The Belgian Organization for Accreditation is responsible for quality assurance and checks for pesticide conformity. The Scientific Institute of Public Health (WIV-ISP) conducts scientific research to support health policy and provides public service. The Federal Public Service for Health, Food Chain Safety and Environment is the EFSA Focal and Codex Contact Point in Belgium and responsible for policy and legislation concerning pesticides. The Federal Agency for the Safety of the Food Chain (FASFC), was set up in 2000, under the responsibility of the Federal Minister of Health, Food Chain Safety and Environment (EFSA 2011; FAVV 2010b; Vlaamse Overheid 2007). FASFC oversees all official pesticide residue controls from farm to fork, based on multi-annual control plans (Claeys *et al.* 2011; EFSA 2011); it stipulates target pesticides for each sample type in application of EC/396/2005, allocates samples to different labs and is the RASFF Contact Point for Belgium (BfR 2011; EFSA 2011; FAVV 2010b). FASFC also defines and enforces operational standards, applicable during food production and is supervised by the European Food and Veterinary Office.

2.3.2 MONITORING PROGRAM

The national program includes the ECMP as well as targeted sampling on products from specific countries. The control program is risk based and follows a general statistical approach, in which pesticide toxicity, violations in previous years, RASFF messages, food consumption figures, authorized pesticides and the budgetary potential are considered (Claeys *et al.* 2011; EFSA 2010a; EFSA 2011; FAVV 2010a; FAVV 2010b). All groups of fruits and vegetables are included and a rolling program is applied for less important commodities (EFSA 2010a; EFSA 2011; FAVV 2010b).

Some organizations impose extra requirements on farmers in order to obtain a certificate or label (e.g. Flandria, GlobalGAP). To obtain a certificate, producers accept additional pesticide use restrictions on top of the legal obligations and frequent checks by independent control bodies (Vlaamse Overheid 2007).

2.3.3 AUTHORISATIONS IN APPLES AND CARROTS

In Belgium, each different formulation of an active ingredient is given a different authorisation number. Overall, 219 formulations were authorised in apple production, all containing one, two (19 formulations) or more (3 formulations) of the 82 active ingredients that were authorised in this crop. The main active ingredients were mancozeb, present in 11 % of the formulations and sulphur, bifenthrin and difenoconazole, which were present in 5 % of all formulations for apple production. The most common formulation types were wettable granules (WG) (25 %), wettable powders (WP) (15 %) and emulsifiable concentrates (EC) (14 %). On carrots, 26 active ingredients were authorised, present as 73 different formulations. Here, 13 formulations were based on a combination of two different active ingredients, reducing resistance induction. Pyrethrins (11 %), difenoconazole (10 %) and dimethoate (10 %) were the main ingredients for use in carrot cultivation. In this case, EC (43 %), followed by WG (19 %) and suspension concentrates (SC) (14 %) were most popular. Based on the number of formulations that were available, the main targeted pests and diseases in apple orchards were *Venturia inaequalis*, aphids, *Podosphaera leucotricha*, *Carpocapsae* and *Tetranychidae*. In carrot cultivation, annual dicotyledonous weeds, aphids and *Erysiphe heraclei* were mainly targeted.

2.3.4 SAMPLING AND ANALYSIS

The FASFC specifies the target pesticides for each sample and samples are mainly taken at auctions, import points, wholesalers and processors by trained officers according to Dir. 2002/63/EG (Claeys *et al.* 2011; EFSA 2011; FAVV 2010b). Auctions and certification bodies also take samples themselves (De Clercq 2011). In case of a non-compliant sample, follow-up action is taken to identify the cause and the responsible company/person (De Clercq 2011; EFSA 2010a; EFSA 2011; FAVV 2010b). The producer/importer is then subject to enhanced controls and a fine is proposed by the FASFC legal department (EFSA 2010a; EFSA 2011; FAVV 2010b). When the MRL violation lies within the analytical uncertainty, a warning is issued. When intake calculations (according to SANCO/3346/2001) indicate a consumer risk, an international alert will be issued by RASFF and protective measures will be taken (EFSA 2010a).

Since 2009, all five Belgian analytical labs are ISO 17025 certified and take part in proficiency tests (FAVV 2010b). Quality control procedures specified in SANCO/3131/2007 are applied and samples are analysed by means of multi- and single-residue methods, covering more than 375 active ingredients. The analytical uncertainty is specified in the guidelines (SANCO) and a default uncertainty figure of 50 % of the results is used in cases of enforcement decisions (EFSA 2011; FAVV 2010b). The MRL is the key-value and end-point in use, without considering consequences on the food chain. Furthermore, customer demands play an important role as often just a specific set of residues is required to check for (De Clercq 2011).

2.3.5 RESULTS OF THE MONITORING PROGRAM

2.3.5.1 GENERAL RESULTS

In 2008, FASFC took 1602 samples of which 1413 in fruit and vegetables (EFSA 2010a). Of the 349 analysed active ingredients, 138 were found at least once. Residues were detected in 72 % of the samples but only 6 % was non-compliant (Claeys *et al.* 2011). The pesticide residues that were found most often were iprodione, boscalid and dithiocarbamates (EFSA 2010a).

The main fruit groups for which residue levels exceeded the MRL, were exotic fruits, citrus fruits, berries and small fruits. The main groups of non-compliant vegetables, were fruiting vegetables, bulb vegetables and legume vegetables. Potatoes, lettuce, tomatoes and apples are generally the most important contributors to the exposure to dithiocarbamates, while apples are amongst others for difenoconazole, thiabendazole, chlorpyriphos, λ-cyhalothrin and thiacloprid (Claeys *et al.* 2011). In 2009, 2112 ECMP-samples of fruits, vegetables, cereals, animal products and processed products were taken in Belgium (EFSA 2011). Of the 1871 fruit and vegetables samples, 96 % complied to the MRLs set in legislation (EFSA 2011; FAVV 2010b).

2.3.5.2 PESTICIDE RESIDUES IN TRADE

Of all commodities analysed in 2009, 42 % was grown in Belgium, 21 % came from the EU and 31 % was imported from third countries. Similar to previous years, import from third countries (12 %) exhibited proportionally more MRL violations than local (5 %) or EU (3 %) produce (Claeys *et al.* 2011; EFSA 2011; FAVV 2010b). MRL violations were mainly seen in strawberry, currant, celery, leek, spinach and parsley samples (FAVV 2010b). Products from Thailand (chili peppers) and the Dominican Republic (lauki and wild mushrooms) were targeted in 2009 (241 samples), because of suspicion, complaints or previously found violations. About 74 % of these samples was conform to EC-MRLs (EFSA 2011; FAVV 2010b).

2.4 BRAZIL (SAO PAULO)

Fruit production is one of the most important areas of agricultural activity and places Brazil on a third position, while ranked 12th in world fruit exports. Almost 60% of its fruit and vegetable production is exported to Europe (Ciscato *et al.* 2009).

2.4.1 REGULATIONS AND RESPONSIBLE AUTHORITIES

The basis for the Brazilian pesticide regulation was enacted in 1989 and is now replaced by Acts 4074/2002 and 5981/2006 of the National Sanitary Surveillance Agency (ANVISA) (Jardim and Caldas 2012). In the 1990s, Brazil started the “Integrated Production of Fruits-Program” for apple production, for a more effective use of land and chemicals (Ciscato *et al.* 2009). Recently, the integrated production of fruits-program was extended to ten other fruit crops. The acceptable residue levels are specified in the Federal `Lei de Agrotóxicos e Afins Nº 7802’ of July 1989` (Ciscato *et al.* 2009; Jardim and Caldas 2012; Tondo 2011). To minimize non-authorized pesticide use on minor crops, MRLs were extended (2010) to all crops within a group (Jardim and Caldas 2012). Since June 2011, MRLs are established for 343 active ingredients on a variety of food commodities in the country (Jardim and Caldas 2012).

The Ministry of Agriculture, Livestock and Food Supplies (MAPA) is responsible for the certification of (new) pesticides (Ciscato *et al.* 2009), defines legal pesticide/crop combinations, evaluates pest control efficacy and approves product labels. ANVISA is responsible for pesticide registration (Ciscato *et al.* 2009; Jardim and Caldas 2012), human health impact evaluation of pesticide use and establishing MRLs based on national GAP and supervised residue trials conducted throughout the country. They also make recommendations for surveillance. The pesticide Residue Laboratory del Instituto Biológico is supervised by MAPA, evaluates fruit products for residues and verifies compliance with national and international MRLs (Ciscato *et al.* 2009; Tondo 2011). The organisation of pesticide legislation in Brazil is displayed in Figure 2.1.

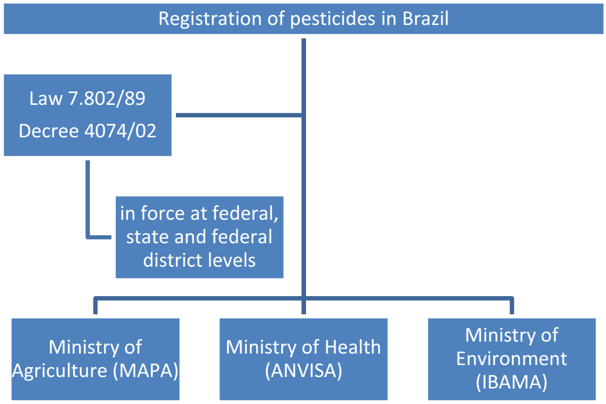


Figure 2.1: Organigram of involved bodies in pesticide registration in Brazil (Rangel 2011).

2.4.2 Monitoring programs

Two permanent nationwide monitoring programs for pesticide residues in food of vegetal origin are in place in Brazil: the Program on pesticide Residue Analysis in Food (PARA), coordinated by ANVISA and the National Residue and Contaminant Control Program (PNCRC), coordinated by the Coordination of Residues and Contaminants, structurally linked to MAPA (Jardim and Caldas 2012; Rangel 2011). PARA was initiated to guarantee compliance to an integrated fruit production, safety and commercialisation (Ciscato *et al.* 2009; Jardim and Caldas 2012). Goals were, measuring pesticide levels in Brazilian food, verifying MRL violations in practice, detecting pesticide abuse and supplying data for daily intake estimations (Jardim and Caldas 2012). Action plans for pesticide reduction have been developed with cooperation of the producers (Ciscato *et al.* 2009). PNCRC for products of vegetal origin was initiated in 2006. Its main objectives were to verify and evaluate good agricultural practices, check food safety and supply guarantees equivalent to international requirements (Codex Alimentarius, WTO, FAO, WHO) (Rangel 2011). Crop selection in these programs varies according to the objectives of the surveillance services and Government (Tondo 2011). In addition to these nationwide monitoring programs, state laboratories also analyse fruit and vegetables from local markets and distribution centres.

2.4.3 Authorisations in apple and carrots

In Brazil, all products containing one specific active ingredient are assigned the same authorisation number, even when formulated differently. On apples, a total of 204 formulations was authorised, of which 10 were not available on the Brazilian market and 5 others were not registered in apples. All these formulations consisted of one, two (11 formulations) or more (1 formulation) active ingredients of the 90 that were authorised. The main active ingredients were glyphosate, mancozeb and thiophanate methyl, respectively present in 10, 7 and 5 % of all authorised formulations. The most popular formulation types were EC (27 %), WP (24 %) and microencapsulated particles (CS) (16 %). For carrots, only 9 active ingredients and formulations were authorised. Most available formulations targeted pests and diseases in apple orchards: *Venturia inaequalis*, *Tetranychidae*, *Colletotrichum gloesporioides* and fruit moths. In carrot cultivation, *Alternaria douci*, *Erwinia carotovora*, *Cercospora carotiae* and *Meloidogyne javonica* were mainly targeted.

2.4.4 Sampling and analysis

Sampling at state level is done according to the Codex Alimentarius for fruits in natura and residue analysis is conducted with a multiresidue method (127 active ingredients) (Gebara *et al.* 2008). Samples for PARA are randomly collected by state surveillance agencies at local supermarkets and food distributors (Jardim and Caldas 2012; Tondo 2011). In 2009, PARA monitored 234 active ingredients in 20 kinds of food, chosen according to consumption habits and food presence in different States and resulting in approximately 600 collected samples (Tondo 2011). PNCRC initially analysed export samples of apple and papaya, but was expanded to samples from local markets and other crops. Federal agriculture inspectors collected 934 (63 %) samples of export products at packing houses and 550 samples for domestic consumption at a key national fresh food distributor (Jardim and Caldas 2012).

Four of the five analysing labs within PARA are state labs, inspected and ISO/IEC17025 certified by ANVISA. PNCRC-samples are analysed at two accredited labs. In both programs, a multi-residue method with a scope of 140 substances is used. LOQs were generally 0.01 mg/kg, but this varies among laboratories, matrices and compounds (Jardim and Caldas 2012).

Brazilian labs analyse authorized, as well as illegal pesticides (Tondo 2011). Each lab in each quarter of a year analyses only one previously chosen agricultural product to maximize analytical productivity and minimize costs. The analysis includes the peel, seeds and pulp of fruits (Ciscato *et al.* 2009).

2.4.5 Results of the monitoring program

A state study in Sao Paolo, showed a high percentage of detectable residues in strawberries (71 %), apples (64 %) and pears (54 %). The ingredients that occurred most were captan, chlorpyrifos and dimethoate. Of all fruit samples, 27 % contained detectable residues, 20 % violated the legislation and 9 % contained multiple residues. In general, the monitoring program revealed a great incidence of residues that were not permitted on fruits, but the chronic dietary intake calculations did not exceed the toxicological acceptance level (Gebara *et al.* 2008). Between 2001 and 2010, a total of 13556 samples were analysed under PARA (12072 from 2001 till 2009) and PNCRC (1484 from 2006 till 2010) (Jardim and Caldas 2012).

2.4.5.1 PARA

Within PARA, 350 tomato and strawberry samples were analysed. Before 2006, less than half of the analysed samples contained residues, while from 2006 until 2010, at least one active ingredient was found in almost every sample. Samples containing residues of unauthorized active ingredients or violating the Brazilian MRL, accounted for 13 % of all samples and 27 % of the positive samples. The highest percentage of irregular samples (18 %) was recorded in 2009, mostly related to extended use of unauthorized substances (72 %) (Jardim and Caldas 2012). Dithiocarbamates and pyrethroids accounted each for about 20 % of all detected residues (Ciscato *et al.* 2009; Oliva *et al.* 2003) and apple was the crop with the highest number of analysed samples (80 % positive samples) (Jardim and Caldas 2012).

2.4.5.2 PNCRC

Analysis results showed that 75 % of the samples at the national distribution centre and 89 % at packing houses were positive. Contrarily, only 3 % of the packing house samples were irregular, while at the distribution centre, this was 11 %. Respectively 2 and 6 % of the samples indicated non-authorised use. One third of all positive samples contained organophosphates. Chlorpyrifos, methamidophos, acephate and dimethoate, represented 15 % of the residues and were the most frequently found compounds within the PNCRC sampling plan (Jardim and Caldas 2012).

Apples accounted for 77 % of the samples with carbaryl residues, which makes them the most important source of carbamate residues within PNCRC (Jardim and Caldas 2012).

2.4.5.3 PARA VERSUS PNCRC

An overall comparison of both programs is impossible since the number of samples analysed within PNCRC was much smaller than in PARA. However, similar frequencies of positive samples were noted for apple and papaya. The majority of the irregular apple samples in both programs contained unauthorized substances (67 % in PARA and 72 % in PNCRC). Overall, a lower frequency of positive samples but a higher frequency of irregular samples (18 %) was noted in the PARA-program, when compared to the PNCRC-program (8 %).

2.4.5.4 PESTICIDE RESIDUES IN TRADE

The analytical results indicated an important difference between production for domestic consumption and for export which is subject to more restricted pesticide use (Jardim and Caldas 2012). The monitoring of export products in 2006 was conducted on a total of 112 fruit samples, following Codex procedures. The samples of avocado, cherimoya, mulberry, fig, grape, guava, lemon, mango, papaya and persimmon, all had a destination in Europe. Only 4 % of them contained pesticide residues exceeding the Brazilian MRL. In contrast, 13 % of those samples exceeded the EC-MRLs. This indicates an important difference between Brazilian and European legislation, possibly causing trade restrictions (Ciscato *et al.* 2009).

2.5 EGYPT

In Egypt, pesticides remain the most widely used tool of controlling the majority of agricultural pests. Unfortunately, the risk to the population caused by the (ab)use of those agrochemicals is among the most important environmental problems in the country. Also human health problems, such as acute pesticide poisoning, occur frequently (Amr 1999).

2.5.1 REGULATIONS AND RESPONSIBLE AUTHORITIES

Most agricultural pesticides are imported into Egypt. The ministry of Agriculture and Land Reclamation is responsible for the Agricultural pesticide Committee (APC - Ministerial Decree 2188/2011) and the Central Agricultural pesticides Lab that governs the official Egyptian pesticides database. The pesticide Committee evaluates the efficacy, safety and performance of pesticides before registration and is liable for pesticide regulation (Ministerial Decree 1018/2013) and monitoring (Yehia 2014). It considers many international reference guides and guidelines for policy setting, registration requirements and procedures, in order to avoid obstruction of trade with other nations (APC 2010; Yehia 2014). The ministry of Health checks for illegal pesticide use and limit violations. The most common post-registration activities include training programs, licensing and enforcement schemes and also monitoring (El Tahan 2012). Figure 2.2 displays the organigram for the Egyptian pesticide legislation.

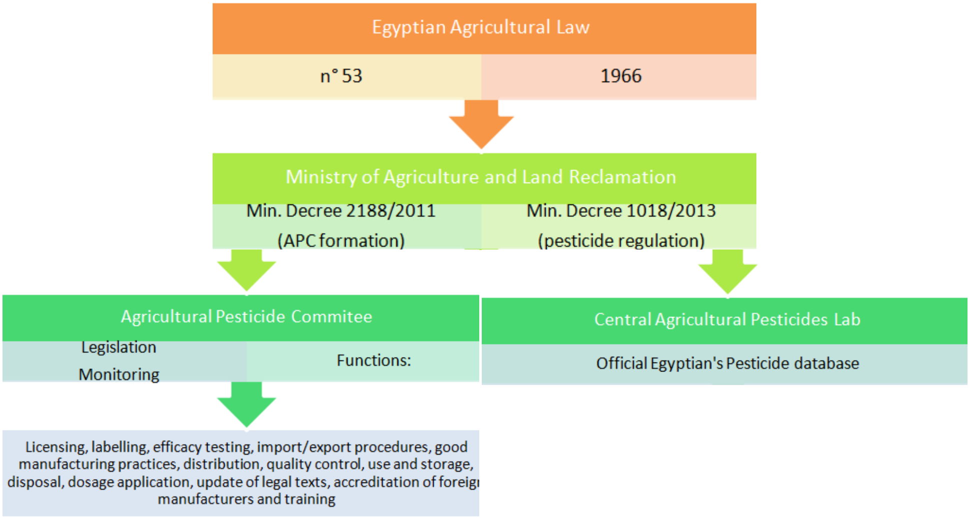


Figure 2.2: Organigram of Egyptian bodies involved in pesticide legislation, registration and monitoring.

2.5.2 AUTHORISATIONS IN APPLE AND CARROTS

A total of 44 formulations (27 registered active ingredients), was authorised for use on apples. The main active ingredients were glyphosate, carbendazim and sulphur, respectively present in 27, 9 and 7 % of all authorised formulations. The most popular formulation types were EC (30 %), WP (21 %) and soluble concentrates (SL) (18 %). In carrot production, only promethryn was registered. The most common ingredients for apple orchards (number) targeted *Podosphaera leucotricha*, annual and perennial weeds, *Venturia inaequalis* and *Tetranychidae*. In carrot cultivation, only annual broadleaf and grassy weeds were targeted.

2.5.3 SAMPLING AND ANALYSIS

Sampling can be done on three levels in Egypt. At first, exporters can take and send samples to the analysis lab. The sample volume in this case, is regulated for each commodity. Secondly, customers can require analyses, but then, the lab is responsible for sampling. Finally, samples can be taken by the government.

The quality control through pesticide residue analysis in Egypt is, for 95 %, conducted on export products and is not obligatory. This explains the limited research and few samples taken at retailers. Labs are accredited for pesticide residue analysis of exported fruits and vegetables and participate in proficiency tests. The scope of every analysis is defined by the customer, or sometimes all possible residues are searched for. In case of imported products, each commodity has a specific scope for which analysis is done (El Tahan 2012).

2.5.4 MONITORING RESULTS

In 2001, 2318 domestic samples of fruit and vegetables were collected from eight Egyptian local markets. Residue analysis was conducted for 54 substances and residues were detected in 18 % of all samples. Root and leafy vegetables showed the lowest contamination rates (respectively 2 and 5 %), marginally exceeding the MRL in leafy vegetables. Overall, 2 % of the contaminated samples showed MRL violations. Fruit samples showed a slightly higher proportion of contamination than vegetables (29 and 14 %, respectively) and exhibited a similar level of violation (± 2 %). Residues of dicofol and dimethoate occurred most frequently here (Dogheim *et al.* 2002). Nevertheless, the decrease of contamination and violation rates compared to previous recordings indicates a positive trend (Dogheim *et al.* 2004; Dogheim *et al.* 1996).

2.6 SOUTH AFRICA (WESTERN CAPE)

Marketing of fresh fruits in South Africa is mostly oriented towards exports. The top-five of exported agricultural products are oranges, wine, apples, grapes and pears (USAID 2006). South Africa covers 2-3 % of the global agrochemical market, with the highest market share for herbicides (39 %) (Department of Agriculture Forestry and Fisheries 2009).

2.6.1 REGULATIONS AND RESPONSIBLE AUTHORITIES

Several national standards are in use for the 3000 approved pesticide formulations. In general, the pesticide industry has a well-structured agricultural “dealer network” of accredited and officially trained pesticide salesmen (Pest control operators). In order to establish international benchmarked MRL values, South Africa relies on the Codex, which accounts for regional variations in pest control requirements (Korsten 2012).

South Africa’s food control system is a typical multiple agency food control system. The responsibility for pesticide legislation, regulation, human health, product quality and safety and efficacy assessment, is shared by the Department of Agriculture, Forestry and Fisheries (DAFF), the Department of Health (DoH) and the Department of Trade and Industry (Bruckner *et al.* 1998; Department of Agriculture 2005). DAFF, through the Directorate Food Safety and Quality Assurance, is responsible for the certification and registration of (new) pesticides under the Fertilizers, Farm Feeds, Agricultural and Stock Remedies Act 36/1947. It also develops and implements policies and guidelines related to pesticide management, such as the Agricultural Products Standards Act (No. 119 of 1990). DoH is responsible for setting and approving MRLs for agricultural food products under the Foodstuffs, Cosmetics and Disinfectants Act (No. 54 of 1972). DAFF and DoH also conduct surveillance and monitoring, as well as compliance checks and enforcement. Local inspections and auditing are performed by the Agricultural Product Inspection Services and inspection agencies such as the Perishable Product Export Control Board (DAFF 2012; Korsten 2012; Mutengwe 2011).

2.6.2 MONITORING PROGRAM

Only a limited pesticide monitoring program exists in South Africa. However, unofficial monitoring is done by industries as part of their approach to product stewardship. The pesticide industry should also report factual information regarding pesticides that may cause unreasonable adverse effects on human health, safety or the environment. However, this is currently not a requirement and therefore not officially done (Korsten 2012).

2.6.3 Authorisations in apples and carrots

In South Africa, a total of 425 formulations was authorised on apples. They consist of one, two (6 formulations) or three (2 formulations) active ingredients (125 authorised). The main active ingredients were deltamethrin, bitertanol and triadimenol, respectively present in 3, 2 and 2 % of all authorised formulations. The most popular formulation types were EC (18 %), WP (15 %) and SC (13 %). In carrot production, only 9 active ingredients were authorised and used in 29 different formulations. The most frequently occurring active ingredients here, were chlorpyrifos, parathion and difenoconazole, respectively present in 24, 21 and 17 % of the formulations. Twenty one percent of the formulations in use were EC, while granules and WP both accounted for 11 %. Based on the number of available active ingredients, the main targeted pests and diseases in apple orchards seemed to be fruit moths, *Tetranychidae*, aphids and beetles. In carrot cultivation, annual and perennial weeds were mainly targeted, followed by sucking and chewing insects.

2.6.4 Sampling and analysis

Samples of fresh fruits and vegetables are drawn from local fresh produce markets and retailers according to Codex guidelines, with replicates at the beginning of the season, peak and late season. Targeted commodities are table grapes, apples, pears, nectarines, peaches, plums, cherries, apricot, mangoes, cabbage, lettuce and spinach. Sampling is conducted according to the Standard Operating Procedure (SOP) on “Sampling and Analysis of Agricultural Products of Plant Origin” to determine agrochemical residue levels as part of export inspection and certification in terms of the Agricultural Product Standards Act, 1990. At ports, samples are randomly extracted from different crates within consignments, at specified rates (Korsten 2012; Mutengwe 2011).

Analyses are conducted by Government Labs in Pretoria or Stellenbosch and also in Private Labs by methods, provided by DoH. However, Act No. 36 of 1947 does not require testing at an accredited lab, labs follow strict regulations and procedures and analysis quality is checked through ring tests. Currently, only two labs are certified for pesticide testing according to EU-GLP (Good Laboratory Practices) requirements. Pesticide detection is done by multi-residue analysis and monitoring is focused on a wide spectrum of substances that include pesticide residues from registered spraying programmes, lists with requirements of importing countries and banned or illegal substances (Korsten 2012; Mutengwe 2011).

2.6.5 PESTICIDE RESIDUES IN TRADE VERSUS LOCAL MARKETS

South Africa approves Codex MRLs for import purposes but those are only applicable when no South African MRLs have been established. The second default choice is the LOD as required in the South African Food Code. Finally, if none are applicable, MRLs set by other countries are considered. Import inspection and sampling is the responsibility of the municipalities or Port Health inspectors but is done inconsistently throughout the country. Port authorities report that it has been five years since the last MRL testing was done because it is an expensive procedure and it is not easy to prioritize. However, they do react on threatening health alerts and rely on reports from other international organizations by organizing contamination analysis and detainment of imported products until further analysis can be conducted (Korsten 2012).

Twenty fresh produce markets in South Africa are registered for export according to international standards required by for example the Organization for Economic Cooperation and Development or Codex Alimentarius. All exported food products of plant origin are inspected for quality and safety issues as required by Act No. 119 of 1990. On the contrary, agricultural fresh produce sent to the local Fresh Produce Markets together with some of the imported fresh produce are not analysed for chemical residues. Some are not even inspected for compliancy with food safety and quality assurance as stipulated in local regulations for fresh vegetables and fruits (Korsten 2012).

2.7 COMPARISON BETWEEN COUNTRIES AND DISCUSSION

Table 2.1 shows the pesticide legislative and monitoring differences for the four Veg-i-Trade countries under consideration.

2.7.1 LEGAL FRAMEWORK

The considered countries all have a different legislative basis, but Belgium and Egypt both mainly use the EU-MRLs as limit. In the other countries, national MRLs are in place, often combined with the values stated in the Codex Alimentarius.

Table 2.1: Detailed overview of all pesticide legislation, sampling, analysis and monitoring properties for Belgium, Brazil, Egypt and South Africa.

Country	Belgium	Brazil		Egypt	South Africa
Food Legislation Basis	EC/178/2002				
pesticides	EC/1107/2009, phytosanitary import control	Act 5981/2006, international legislation			Act 119 (1990), Act 54 (1972)
MRLs	European Commission + national	ANVISA (national)		Europe + other export countries	national + Codex (import)
1 Institution for RA/RM[i]	no	yes		no	yes
Monitoring					
Basis	ECMP, national program, rolling program	2 national programs: PARA	PNCRC	limited implementation, not obligatory	limited program, unofficial monitoring by industries, only export products
Prioritisation	pesticide toxicity, previous violations, RASFF, consumption habits, authorised active ingredients, budget	objectives surveillance services and government, consumption habits, legal and illegal active ingredients		human resources, technical requirements, analytical facilities	registered spraying programs, import requirement lists, banned/illegal active ingredients
Sampling	risk based	ad random		not risk based	ad random
Sampling plan	Dir. 2002/63/EG	SOPs, Codex Alimentarius		random, Codex Alimentarius	Codex guidelines, Act 119 (1990)
Who	trained FASFC officers, auctions, certification bodies	state sanitary surveillance agencies	federal agricultural inspectors	exporters, customers, government	agri product inspection services, port health inspectors, perischable product export control board
Where	auctions, processors, border inspection posts, retail	local supermarkets, food distributors	packing houses	local markets	ports, local fresh produce markets, retail
Samples in fruit and vegetables	(2009) 1871: 241 targeted	(2009) 600	(2009/2010) 934 export; 550 domestic	(2001) 2318 domestic	(2009) 162 fruit; 54 leafy vegetables
Analysis					
Laboratories	#5: ISO17025 accredited	#4 ISO17025 #2 national labs	#2: ISO17025	accredited	South African National Accreditaion System: #2 (=EU good lab practices)
Ring-tests	EU + Food analysis performance assessment scheme		annually	yes	yes
Analysis methods	LC/MS or GC/MS		GC/FPD, ECD, NPD, MS-MS or LC/MS-MS	multi-residue	multi-residue
Scope; N° of active ingredients	defined by customer 349	234 (20 crops) + Mini Luke 140		customer defined, specific for imports 54	
Consumption data	national data		national statistics institute	national	

[i] RA: Risk assessment; RM: Risk management

2.7.2 Monitoring programs

2.7.2.1 Essentials of a good Monitoring Program

A good basis for a national control program is essential (Wauchope 2010). At first, a selection of active ingredients that can be used safely on specific crops should be defined. This can be done by conducting a realistic exposure assessment of the population based on decent national dietary characteristics and toxicological details, environmental fate and climatic conditions as well as the efficacy of each substance (EFSA 2011; Wauchope 2010). Secondly, a listing of all authorised substances and their application specifications is needed. Third, a Quality Assurance program should be in place, allowing a permanent monitoring of formulation quality conformity. For that, specialized analytical laboratories and a regular, randomized sampling of products are needed. Next, producer awareness about the need to use pesticides according to the indications given on the label, should be enhanced and users should be persuaded to act according to it. Finally, the implementation efficiency should be checked through monitoring of pesticide residues. Nevertheless, this is only the final verification of the application of Good Agricultural Practices (GAP) in a country (Wauchope 2010). According to EFSA (2011), the costs of the analysis, the lab capacity and national budgets should also be taken into account.

2.7.2.2 Situation anno 2012

Table 2.2 gives an overview of the implementation of these 'essentials' in the selected countries according to literature.

Table 2.2: Overview of the application of "Essentials of a good Monitoring Plan" (Wauchope 2010) in the Veg-i-Trade countries. (■ fully implemented; ■ with remarks; □ no data)

Essentials / **Country**		Belgium	Brazil	Egypt	South Africa
Defined active ingredient - crop combinations		fully implemented	fully implemented	with remarks	with remarks
Listed specifications/restrictions for all authorised active ingredient		fully implemented	fully implemented	fully implemented	fully implemented
Quality assurance of pesticides		fully implemented	fully implemented	with remarks	fully implemented
Enhancing producer awareness:	- education	with remarks	with remarks	with remarks	with remarks
	- surveillance	fully implemented	fully implemented	with remarks	with remarks
	- sentence system	fully implemented			
Efficiency monitoring		fully implemented	fully implemented	with remarks	with remarks

The Belgian control program is risk based and particular attention is given to foodstuffs, susceptible to exceed the MRLs. The only discrepancy with the 'essentials' is that education was not properly tackled in the past. Since 2011, however, a new regulation imposes several obligatory training/education sessions for pesticide applicators and salesmen. The mandatory scheme will be effective as from november 2015 (De Cock 2011). Jardim and Caldas (2012) indicate that most Brazillian food growers have low levels of education and receive limited technical support, do not read the pesticide labels or do not understand their content and are economically vulnerable. In many cases, decisions regarding which pesticides to use, rely strongly on farmers' previous experience, product costs and availability on the farm. Oliva *et al.* (2003), describe only a minimal phytosanitary support given to certain crops. They also state that measures are particularly required to counteract the frequent illegal use of certain pesticides in fruit and vegetable production for domestic consumption. Due to the profile of the country's agricultural population, an extreme difference in product quality of export and domestic products was noted (Jardim and Caldas 2012; Oliva *et al.* 2003). For Egypt, Amr (1999) mentions a discrepancy between government recommendations for specific pesticide/crop combinations and reality, where products are registered for a specific pest. A lack of legislative basis for pesticide production or formulation was also mentioned, since all pesticides are imported from abroad. Consequently, producers are relied on for product quality assurance. In general, everything is allowed and there is an important difference between facts and regulations. Because of the large number of small farms, scattered over the countryside it is very difficult to police the use of pesticides. The only way of detecting misuse is through pesticide residue analysis (Amr 1999). Nevertheless, quality control through pesticide residue analysis was not obligatory and conducted for 95% on export products (El Tahan 2012). South Africa recently implemented crop specific registration and authorisation resulting in many GlobalGAP-certified companies losing their certificate. Education was tackled but not in a sustainable way. Without easy access to training, farmers lack knowledge to recognize pests and predators, select products and apply them properly. In addition, quality and safety surveillance of local produce was only rarely done (Korsten 2012; Mutengwe 2010).

2.7.3 AUTHORISATIONS

The active ingredients and formulated pesticides, used in the considered countries, differ substantially for each commodity. The number of active ingredients and formulated products allowed on a specific crop are also variable. Pesticide availability in apple cultivation of all four countries can indicate that this is a valuable crop. Carrot production on the other hand is probably only of consideration in Belgium and South-Africa, since these are countries with at least some diversity in available pesticides for this crop. Formulation preference does not show any link with the local climate, however, such an effect was expected here (see Chapter 5 & 6). The scope of targeted pests in apple cultivation also did not differ substantially between the regions. In both Brazil and Belgium, most pesticides were fungicides due to frequent humid conditions, followed by insecticides. In South Africa, insecticides were most common, while in Egypt insects were least targeted. If this was a good representation of plague occurrence, different climates do not necessarily seem to imply other infestation problems. However, sales figures are not available, so the effective use and importance of pesticides for the different commodities are unknown. Carrot production in all countries was mainly hampered by weed occurrence. An important remark is that the three most frequently occurring active ingredients in authorised formulations on for example Brazilian apples, are only authorised in Belgium. Both in Egypt and South Africa, at least one of those three active ingredients could not be used in apples (illegal). Hence, problems can occur when commodities with residues of those substances are imported in one of those countries.

2.7.4 SAMPLING AND ANALYSIS

Considerations regarding analysis are that it was not always determined which part of the crop should preferably be analysed (Braeckman 2009), nor were there corrections made for recovery or analytical uncertainty of the results (Codex Alimentarius Commission 1993; EFSA 2010a). When looking at monitoring in trade, important issues arise. Preferably, the share of domestic and imported samples, analysed within a monitoring program, should reflect the situation in the respective national market to be representative. In addition, the sampling method, often not random in imported products, can distort the view on the conformity of traded products.

Overall, there are several factors that cause differences in national monitoring programs. Firstly, the scope of active ingredients, investigated in different commodities, may vary between countries and in time. Secondly, the sampling method and commodity selection also have an important influence on the monitoring results. Third, differing analytical and technical laboratory capabilities can cause a wrong interpretation of the results. This showed, for example in Belgium, where the analytical performance improvement of the labs over the last ten years allowed for a doubling of the substances to be analysed (EFSA 2010a). The differences in national monitoring programs, are likely to account for an important part of the variation in monitoring results (Commission of the European communities 2008). Nevertheless, it seems that fruit samples generally contain more residues than vegetables. The frequent MRL violations do, however, not mean that consumer health is always at risk. In addition, the presence of residues should be relativized as foods often contain higher quantities of natural carcinogens than synthetic pesticide residues (Noble 1995).

2.7.5 MONITORING IN TRADE

MRL violations in imports often seem to originate from a difference in national (exporter) and EU-MRLs (importer). The global nature of food trade and increasing globalization, obviously require harmonisation in one way or another, to avoid future trade problems. A first attempt came from the Codex Alimentarius Commission, who proposed a guideline for food import control systems (Codex Alimentarius Commission 2009).

Monitoring results for Brazil show that it will be important to evaluate the extreme difference in product quality between export and domestically consumed products. According to Oliva *et al.* (2003), this is caused by the uncontrolled use of low-quality/low-price pesticides, smuggled across Brazilian borders by small and uneducated local producers.

In Egypt, increased dimensions of food contamination in both domestic and imported products were noted. The biggest problem there, seems to be the shortage of some food commodities, which has led to increased fraudulent trading of contaminated produce that enters the Egyptian market after having been rejected in other countries. In addition, there is an important difference in end points for local and imported produce. For imported produce, the MRL applies, while in other cases customers can decide what is acceptable. In a report by Salam (2010), it was, however, concluded that measures are currently being taken to tackle the issue.

In South Africa, most of the supply of fresh fruits and vegetables for domestic markets and export to overseas markets, is provided by commercial farmers, together with importers. The number of small-scale farmers producing for both markets, is increasing due to the liberalisation of agriculture in South Africa. Quality standards and traceability requirements together with a year-round demand of exotic products are also increasing. This makes it very difficult for small-scale farmers to continue complying with GAP (Korsten 2012). Firstly, stricter registration requirements in the EU, have resulted in companies not re-registering existing pesticides, phasing out more than 500 active ingredients. Secondly, EU-countries often maintain lower MRLs than those defined in the Codex and finally, certain retailers even require a zero limit. The lack of accredited lab-capacity in terms of MRL testing, also directly impacts the country's ability to effectively trade and supply safe food for consumers (Korsten 2012; Mutengwe 2010).

The main issue, when substantial differences in compliance and analysis of local and imported produce are noted, seems a lack of government interference with monitoring and sampling. In countries where government agencies are not responsible for monitoring, independent agencies or industry delegate the analyses. In this case, the targeted products are often only high-value products and almost exclusively for export.

Finally, pesticide monitoring in trade is an important issue in view of increased globalization. Literature provides several reasons why this is difficult, especially in developing countries (Amr 1999). A first issue, is the large number of small farms scattered over the countryside, which makes it very difficult to monitor pesticide use. Secondly, residue analysis under stringent working conditions as enforced in for example the EU, are unrealistic in other parts of the world (Amr 1999; Codex Alimentarius Commission 1993). Third, residue analysis in labs with an irregular electricity supply or limited budget, do not allow the use of e.g. HPLC or mass spectrometry (MS) detection (Ambrus *et al.* 2005). Finally, to be effective, monitoring of pesticide use and residue levels has to rely on legislation and sufficiently trained personnel, to enforce these laws (Keikotlhaile 2011; Wauchope 2010).

2.8 CONCLUSIONS

Important dissimilarities in legal framework, which were mentioned in literature (Keikotlhaile 2011; Wauchope 2010), were confirmed in the comparison between the considered Veg-i-trade countries in this chapter. The different approach and requirements, hamper the comparability of monitoring results of the selected countries. Key differences can be seen in Belgium and Brazil, who execute an extended national monitoring program, while in Egypt and South Africa only basic monitoring principles are stipulated. In addition, it was mentioned that these principles are often only rarely applied. Monitoring in these countries is often driven by international trade, whose standards are incorporated in the control programs. Hence local produce is often excluded from food safety analyses. Egypt and South Africa are two large nations, who are important players on the food (production) market. This shows that the application of chemical food safety still has a long way to go. In addition, the different approach of food safety and monitoring in diverse world regions is still causing important trade issues.

CHAPTER 3 DEVELOPMENT AND EVALUATION OF A RISK BASED PESTICIDE RESIDUE MONITORING TOOL TO PRIORITIZE THE SAMPLING OF FRESH PRODUCE[ii]

This chapter describes the development of a theoretical risk based sampling plan that omits trade barriers based on prejudice and hence, tries to counter the differences in plant protection product legislation and the monitoring approach described in Chapter 2. In addition, a tool for sample size calculation is presented.

3.1 INTRODUCTION

During the last decade, Europe was not only subject to increasing food safety concerns, but also consumer awareness of food safety issues (EFSA 2010b; Van Boxstael *et al.* 2013). This has resulted in increasing demands for operators in the food chain and an augmented importance of food safety and quality controls. A sampling plan - defined as a detailed overview that allows an analyst to make decisions on criteria that are applicable to a batch - is an inherent part of a good quality system in the food industry. In Belgium, official pesticide residue controls according to Directive (EC) No 2002/63/EEG are conducted (EFSA 2011). In addition, Belgian companies are obliged to design and implement a sampling plan following the Royal Decree of November 14th, 2003 concerning "self-control, obligatory notification and traceability in the food chain". In a joint initiative of the sector associations Belgapom, Fresh Trade Belgium and Vegebe with Ghent University, self-checking guidelines were developed for use in trade and processing of vegetables, fruits and potatoes and were accepted by the competent authority. These guidelines include several parameters to

[ii] *This chapter has been compiled from*:

Delcour, I., Rademaker, M., Jacxsens, L., De Win, J., De Baets, B., & Spanoghe, P. (2015). A risk-based pesticide residue monitoring tool to prioritize the sampling of fresh produce. *Food Control*, *50*, 690–698.

prioritize in the sensibility of fresh produce to pessticides and describe all demands and recommendations concerning food safety and quality that apply in processing companies and trade. However, these sampling plans only apply to common risks for all operators and are not company specific. Therefore, companies should consider additional parameters in function of their own risk analysis (Belgapom 2010).

The ability to detect and quantify residues on fresh produce is important to guarantee food safety. The FAO and WHO provide instructions considering where and how many samples should be taken, but none of them helps companies to prioritize between batches (Food & Agriculture Organization 2000). More stringent food safety policies imply higher costs for sampling and analysis. A scientific risk-based sampling plan that considers both risk and economic issues to validate food safety in an efficient manner is desirable. Unfortunately, due to a multitude of suppliers, products, exporting countries and the unknown concomitant and variable prevalence of residue excesses, a statistically founded sampling plan cannot be designed at the company level. Accordingly, the primary goal of this chapter is to develop a monitoring tool for prioritization of fresh produce sampling with concerns for PPP residues and risk. Additionally, a method for sample size calculation based on the developed monitoring tool is provided.

3.2 METHODOLOGY

3.2.1 PARAMETER SELECTION

In a risk based sampling plan, monitoring should be proportionate to the attributed level of risk of particular commodities and processing methods and should be well balanced. In addition, precedence should be given to consumer health protection and assurance of fair practices in food trade (Codex Alimentarius Commission 2009). The guidance document for national control programs advises to consider the use patterns of PPPs, active ingredient toxicity, RASFF notifications, laboratory capacity, budgets and other relevant issues for national control activities (EFSA 2010a; EFSA 2011). Furthermore, the sampling plans of four Belgian sector organizations (Fresh Trade Belgium - FTB, Association of Belgian Horticultural Cooperatives - VBT, Belgapom and Vegebe) and the competent authority (Federal Agency for the Safety of the Food Chain - FASFC) were examined. The parameters that were relevant for monitoring and sample size calculation were combined into a risk-based prioritisation tool.

For calibration of the prioritisation method, Veg-Trade experts (17 in total) with relevant knowledge on PPP residues, were consulted. They were selected to represent the different actors in PPP monitoring within this specific part of the food chain: the government (two Belgian respondents), producer organizations (two Belgian and one Norwegian respondent), auctions (two Belgian respondents), researchers (three Belgian, one Spanish and one South-African respondent) and companies (three Belgian and two Spanish respondents). All contact and questionnaire administration procedures were managed electronically. Data collection was performed in March 2013.

3.2.2 QUESTIONNAIRE AND DATA ANALYSIS OF THE EXPERT OPINIONS

Participants were asked to complete a self-administered electronic questionnaire. The items of interest related to (a) perception of parameter contribution to total PPP residue risk, (b) perceived risk levels of the provided parameters and the associated options and (c) perceived risk of fifteen scenarios that combined different parameter options. The scores assigned by the experts to these scenarios, allows weighing the parameters in relation to each other. Each parameter's contribution to the total risk of purchasing/marketing non-conform produce (a), could be indicated by dividing 100 points over the five parameters, taking into account their judged importance to food safety. Risk level perception (b) was measured on five-point scales ranging from low risk (1) to high risk (5). The fifteen scenarios (c) consisted of different combinations of parameter values, thus presenting an actual situation to be assigned a risk level by the experts (low risk - high risk).

The median risk levels assigned by the experts for (b) and (c), were used for further calculations. Parameter weights were computed based on (c), to allow for the calculation of a risk level for new scenarios. These parameter weights were chosen so as to yield risk levels close to the median assigned risk levels.

3.2.3 PROPOSED RISK-BASED APPROACH

Given a collection of equally sized and risky batches, the sampling capacity assigned to each batch should of course be equal. If one batch is characterized by a greater risk or by a larger size, everything else remaining equal, more sampling capacity should be assigned to that particular batch. In general, a sampling plan for a given collection of batches will require a proper description of the batches (see Figure 3.1 for a general schematic representation).

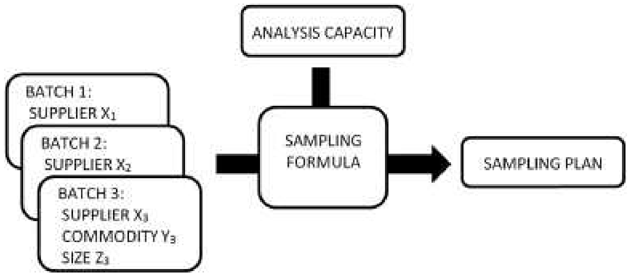


Figure 3.1: Schematic representation of a general approach to constructing a sampling plan.

Unfortunately, it is not possible to objectively and exactly calculate how much more capacity should be assigned to the larger or riskier batch without assuming or estimating prevalence of violations for each batch. Such an estimated or assumed risk could be computed as a function of the inherent commodity risk. Unfortunately, preliminary questionnaires conducted in this work, showed widely varying estimations by experts from both academia and industry. Indeed, it is exactly this large variance that implied the use of risk scores as an alternative.

Hence, a function that takes as input a description of a collection of batches and generates as output a distribution of the total sampling capacity over all batches, was searched for. Important batch characteristics are the scenario risk level, commodity risk and the size of each batch. How to assess scenario risk and commodity risk, was discussed in Section 3.3 of this chapter.

However, if one batch is 100 times larger than another batch, it is probably not desirable to assign 100 times as much capacity to the large batch. As a related example, consider estimating the mean of a population on the basis of a number of samples: the size of the confidence interval associated with the estimated mean is inversely proportional to the square root of the number of samples. Similarly, the square root of the batch size was proposed to mitigate the effect of batch size in this study.

It is important to consider that the exact value of the constructed scenario and commodity risk indicators has no objective physical meaning. This implies that, how to use them, in order to assign sampling capacity, is not objectively established. With an eye toward applicability and practical use, it was decided to simply assign sampling capacity proportionally to the geometric mean of all three indicators. In light of the somewhat arbitrary nature of this decision, support from industry, governing bodies and experts is especially important.

Supplier risk scores calculated with the developed tool, the commodity risk and batch sizes are thus combined in order to yield an indicator for the sampling importance. For a batch B_i, let c_i denote the commodity risk, s_i the scenario risk and v_i the batch size or volume (Equation 3.1). Using these, a batch importance I_i and a batch fractional sampling assignment A_i can be computed (Equation 3.2.). While translating all of these dimensions to a commensurate numerical scale is not necessarily mathematically valid, the following ad-hoc constructed formulae yielded plausible values in practice for batch i=1,...,n:

$$I_i(c_i, s_i, v_i) = c_i \times s_i \times SQRTv_i \quad \text{Equation 3.1}$$

$$A_i(I_1, \dots, I_n) = \frac{I_i}{\sum_{i=1}^{n} I_i} \quad \text{Equation 3.2}$$

$$for\ batch\ i = 1 \dots n$$

How to assess commodity risk is explained in detail in Section 3.3.6, while scenario risk is discussed in Section 3.3.7.

3.3 RESULTS

3.3.1 SELECTION OF RELEVANT PARAMETERS

Examination of six existing monitoring plans at the European or Belgian governmental level and by sector associations, resulted in the identification of the parameters listed in Table 3.1. This shows that the examined (Belgian) sampling plans mainly focus on food commodities with high residues/non-compliance rates in preceding years and crops with high RASFF notification rates. Food originating from producers (or countries) with multiple non-compliances in the past, is also targeted. Additionally, food consumption figures and processing before consumption, are considered (EFSA 2011).

The parameters shown in Table 3.1, were summarized and combined into the following five key parameters: Supplier conformity guarantee, country of origin, processing, cross-contamination and commodity risk (combination of consumption figures, PPP use and PPP risk). These parameters were selected based on their occurrence in the consulted sampling plans, EFSA guidelines and expert panel input. They should all be considered within a risk based sampling plan and are described in more detail below.

Table 3.1: Parameters included in the consulted sampling plans

	Sampling plan					
Parameter	**EFSA**	**FASFC**	**FTB**	**VBT**	**Belgapom**	**Vegebe**
Supplier guarantees	x	x	x	x	x	x
Country of origin	x	x	x	n.a.°	x	x
Product risk		x				
Consumption figures		x	x	x	x	x
pesticidePPP use	x					
pesticidePPP risk (non-compliances and toxicity)	x*	x	x*	x*	x*	x*
Contamination risk				x		
Peeling before consumption (processing)			x		x	x

° this sampling plan is used for Belgian produce exclusively

* this parameter is combined with product risk as mainly RASFF notifications and MRL violations of commodities are considered

3.3.2 Parameter 1: Supplier conformity guarantee

Quality management systems are essential to provide safe and high quality food (Filipovic *et al.* 2008). The most important quality management systems in food industry are ISO 22000 (International Standards Organisation) and the Global Food Safety Initiative benchmarked food safety scheme, in which the British Retail Consortium (BRC) and International Food Standards are included (Filipovic *et al.* 2008; GFSI 2011; Luning *et al.* 2006). The objective of all these quality management systems, is to certify food production processes in an internationally accepted way (Filipovic *et al.* 2008; GFSI 2011). Together with residue analysis reports and PPP residue issues in the past, quality management systems are the key elements in assessing supplier risk. This parameter is taken into consideration in all consulted sampling plans.

3.3.3 Parameter 2: Country of origin

Residue monitoring indicates that countries mostly have a very different compliance percentage for PPP residues, with EU-countries performing better compared to non-EU countries (EFSA 2010a; EFSA 2011). Also noted in developing countries, is the use of hazardous products that have been banned or restricted in developed countries long time ago, such as DDT (Amr 1999; Wilson and Otsuki 2004). The RASFF combined with historical national and governmental data, can be a useful tool. Importing companies should accordingly consider this element to limit the risk of supplying non-conforming produce to the market. Country of origin is also included in all consulted sampling plans, except VBT, which is an auction association dealing with only Belgian and locally grown produce.

3.3.4 PARAMETER 3: PROCESSING

The impact of processing on PPP residue levels, is included in half of the consulted sampling plans. In this case, only consumer actions (such as peeling before consumption) are considered, while other processing activities can also strongly influence residues at the company level. Washing fruit and vegetables, for example, does not change the edible part of the raw commodity, but has an effect on residue levels. This should, consequently, be taken into account when considering consumer risk. In general, PPP residue levels are shown to be significantly lower on processed forms than in fresh produce. The average processing factor for peeling is 0,41 (0,30-0,54 95% confidence interval, 13 studies) and for washing 0,68 (0,52-0,82 95% confidence interval, 25 studies) (Keikotlhaile 2010). Nevertheless, these results can vary significantly, depending on the processing conditions and the physicochemical characteristics of the residues (Claeys *et al.* 2011; Kaushik *et al.* 2009; Keikotlhaile 2011).

3.3.5 PARAMETER 4: CROSS-CONTAMINATION

The transfer of commodities' residues to packaging materials, other commodities, conveyor belts and water basins can result in residues on those items and is gaining increasing attention in food safety. Currently, a single German study on lemons by Wojzich (2010), demonstrates that transfer of imazalil on fruit and vegetables to a packing or transport line and vice versa, is possible. However, the detected concentrations are low. Since these are stand-alone lab results, the risk in practice cannot be ruled out completely. Consequently, although cross-contamination of PPPs in practice is not sufficiently studied yet, this risk is potentially present and listed by the experts as 'important'. Therefore, this parameter is also considered in the tool. Additionally, this inclusion is useful, if the tool will be adapted for other food safety hazards (for example viruses), with cross-contamination occurrence, in the future.

3.3.6 PARAMETER 5: COMMODITY RISK

Legislation provides crop-specific authorisations of active ingredients, implying that on different crops, a different collection of PPPs can be applied. Due to globalization, many of the more than 1000 worldwide used PPPs can occur in our food (EFSA 2010a; EFSA 2011). Accordingly, all consulted sampling plans consider RASFF notifications, MRL violations and also consumption figures separately. In this study, *residue risk*, *toxicity* and *consumption* were combined in the parameter 'Commodity risk'. To quantify this parameter, a case study on apples, strawberries, lettuce, grapes and carrots was conducted.

For each commodity, the ten key PPP residues were selected, based on the Belgian residue monitoring data of 2005 till 2009 (Table 3.2). For this selection, the five-year monitoring data of each commodity were scanned separately for active ingredients that (in order of importance): (1) violated the MRL; (2) were not authorised in the year of analysis; (3) were detectable in the highest number of samples (in percentage terms); and (4) were detected in consecutive years (with increasing residue levels). Next, a qualitative ranking of the active ingredients was conducted. Thus, active ingredients with recent and/or a high number of MRL violations were ranked first. This ranking was then combined with the occurrence of illegal residues. Further ranking was executed by including the percentage of samples with (high levels of) detectable residues and in consecutive years. This way, the selected active ingredients have the highest risk of being (illegally) present at high levels.

Table 3.2: Ten key PPPs for the commodities included in the case study (apples, strawberries, grapes, lettuce, carrots)

Apples	Strawberries	Grapes	Lettuce	Carrots
daminozide	pyraclostrobin	fenazaquin	mepronil	linuron
phosalone	ethion	methamidophos	azoxystrobin	boscalid
pirimicarb	methomyl	fenhexamid	mandipropamid	tebuconazole
dithianon	carbendazim	cyprodinil	pymetrozine	azoxystrobin
boscalid	cyprodinil	famoxadone	iprodione	dimethoate
pyraclostrobin	fludioxonil	sulphur	propamocarb	oxadixyl
cyprodinil	boscalid	imidacloprid	boscalid	chlorpyrifos
flufenoxuron	fenhexamid	azoxystrobin	dimethomorph	metalaxyl
triadimefon	kresoxim-methyl	fludioxonil	deltamethrin	omethoate
carbendazim	mepanipyrim	triadimefon	tolclophos	pirimiphos-methyl

Food consumption distributions were extracted from the Belgian national consumption database of 2004 (De Vriese *et al.* 2005). These were used to calculate an adapted Theoretical Maximum Daily Intake (T*h*MDI), by multiplying the MRLs (mg active ingredient/kg food) by the actual consumption distribution for each commodity (*consumption*). The difference with a TMDI as defined in the WHO Guidelines for predicting dietary intake of pesticide residues (1989 – available online)., is the use of actual daily intakes of fruit/vegetable consumers, instead of l national consumption data that describe long term average intakes. By using the ThMDI, the worst case PPP residue loading of realistic daily menus of actual fruit/vegetable consumers can be assessed. The ADI was then compared with these ThMDIs for the ten key residues on that specific commodity.

To give an indication of the fraction of consumers that is at risk in a worst case scenario for a specific active ingredient and commodity, the percentage of fruit/vegetable consumers that is exposed to different percentiles of the ADI, was computed. An example of the output is shown in Figure 3.2.

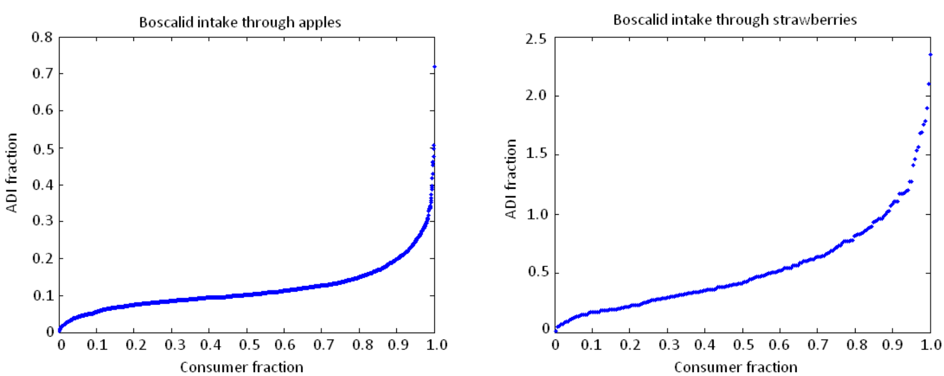


Figure 3.2: Consumer and reached ADI percentiles exemplified by boscalid for apples and strawberries.

Fruit/vegetable consumer percentages for which an ADI violation occurred in these worst case scenarios, are presented in Table 3.3. If, on at least one of the two reporting days in the consumption database, the ADI was exceeded for at least one PPP, the consumer was considered to be exposed to an excessive amount of residues. To this end, only effective consumers were considered, meaning that respondents who did not consume the considered commodities on either of the days, were not included in the corresponding column. The combined exposure to multi-commodity pesticides has not been taken into account in Table 3.3.

Table 3.3: Worst case consumer percentages (effective consumers only) exposed to ADI violations for the selected commodities and over all included pesticides.

	Apples	Strawberries	Grapes	Lettuce	Carrots	All commodities
% of commodity consumers for which ThMDI > ADI at least once (one day and 1 active ingredient)	18.7 %	12.8 %	4.2 %	7.0 %	1.6 %	14.4 %

From the "Apples" column in Table 3.3, we can for example conclude that 18.7% of all consumers that ate (a part of) an apple, were (worst case) exposed to the residue of at least one active ingredient, present on that apple, that resulted in an ThMDI value that exceeded the ADI. The last column corresponds to examining each commodity for each consumer and indicates that 14.4% of all consumers in the database were (worst case) exposed to, at least one, active ingredient-commodity combination, of which the ThMDI exceeded the ADI.

3.3.7 SCORING OF THE PARAMETERS

In the questionnaire, the experts were asked to indicate their perception of each parameter's contribution to total pesticide residue risk (a). The panel indicated that the absence or presence of supplier guarantees contributed on average most (30 %) to the total risk on company level, followed by commodity risk (23 %), processing (18 %), country of origin (16 %) and cross-contamination (13 %) (Figure 3.3).

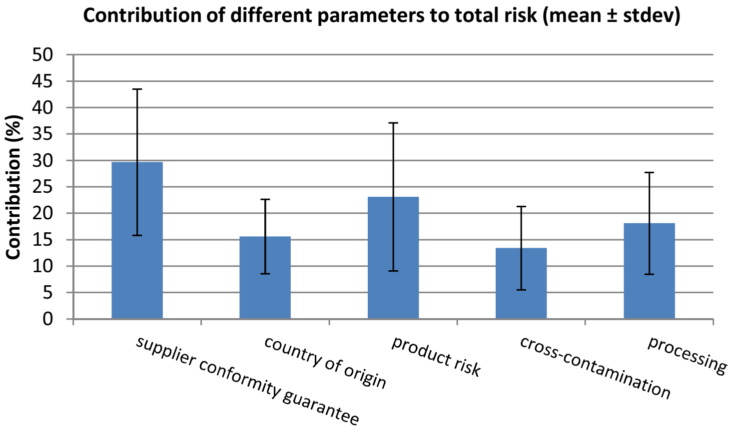


Figure 3.3: Perceived contribution (%) of the 5 selected parameters to the total risk, according to the experts (mean ± stdev).

Another part of the questionnaire inquired about the perceived risk levels of the provided parameters and the associated options (b). Figure 3.4 shows the variation in assigned risk levels for each parameter value (1: low risk - 5: high risk), for the seventeen respondents. The normality of the option medians was assessed and the values were classified using a one-way ANOVA test, accordingly combined with the Dunnets T3 or Scheffe post-hoc test. The risk classification, based on expert opinions and resulting from these tests, is presented in Table 3.4. The final risk scores were calculated by multiplying each option's median risk level with the weighing factor of this parameter. This factor was based on the importance of this parameter compared to the least important (perceived by experts) parameter, which was cross-contamination (Table 3.6).

There was no significant difference in assigned risk levels between experts with a different background for any of the parameter options. Hence, the response of the groups (government, researchers, auctions, company's and company organisations) was not biased by the different numbers of participants in each group.

The fifth parameter, commodity risk, was calculated as described in 3.3.6 and is used further on in the sample size calculation. Accordingly, it was not scored by the expert panel, nor was it included in the calculation of the risk score. We chose to handle the commodity risk parameter separately in order to focus on the controllable aspects of the processing chain.

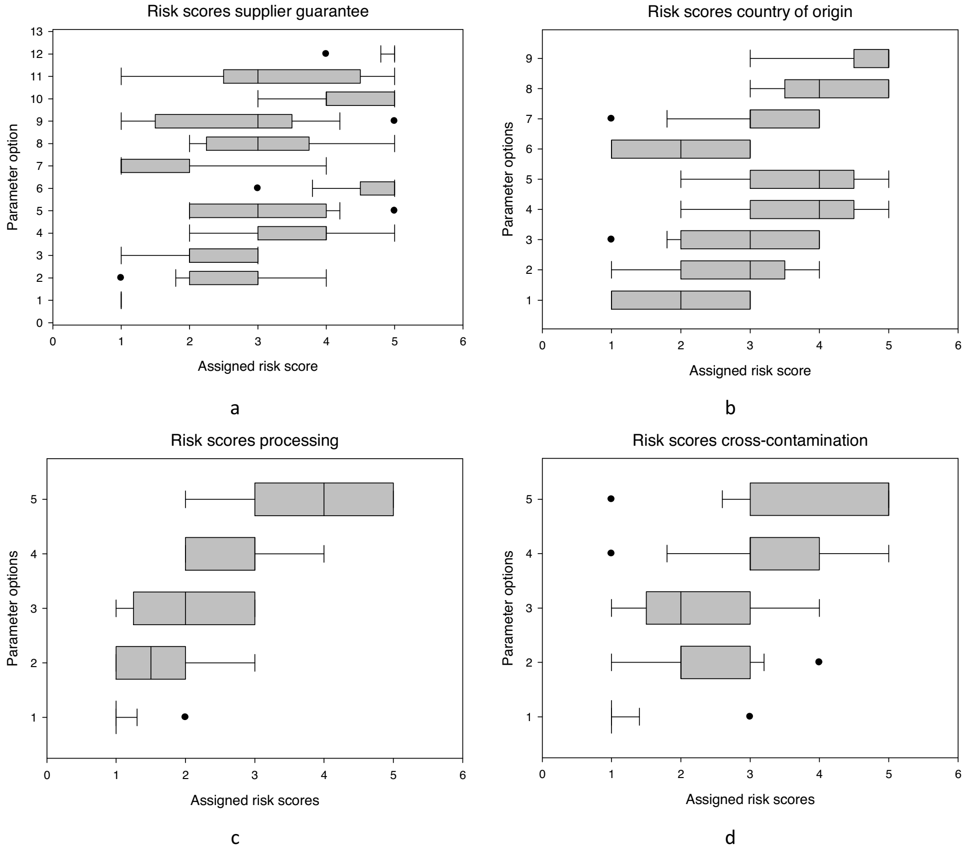


Figure 3.4: Variation in assigned risk levels for each value of the parameters (a) supplier conformity quarantee, (b) country of origin, (c) processing and (d) cross-contamination; in which the assigned risk scores range from 1 (low risk) till 5 (high risk).

Table 3.4: Parameter options, assigned risk levels (low, medium-low (m-low), medium, medium-high (m-high), high) and weighed risk scores (median expert score x weight) for each option.

Option	Description	Risk Level	Risk score*
Parameter 1: Supplier conformity guarantee			
The supplier can provide analysis results for the delivered batch and …			
a	is certified for a QMS (GlobalGAP, ISO, BRC…)	low	11
c	does not have a QMS certification but meets certain FS specifications	m-low	22
e	does not have a QMS certification, nor does meet FS specifications	medium	33
g	has never delivered contaminated produce in the past	low	11
i	has delivered contaminated produce once in the past	medium	33
k	has often delivered contaminated produce in the past	medium	33
The supplier cannot provide analysis results for the delivered batch and …			
b	is certified for a QMS (GlobalGAP, ISO, BRC…)	medium	33
d	does not have a QMS certification but meets certain FS specifications	m-high	44
f	does not have a QMS certification, nor does he meet FS specifications	high	55
h	has never delivered contaminated produce in the past	medium	33
j	has delivered contaminated produce once in the past	m-high	44
l	has often delivered contaminated produce in the past	high	55
Parameter 2: Country of origin			
The product is grown in a country …			
a	that didn't occur in the RASFF last year	m-low	8
b	for which RASFF-News occurred last year	medium	12
c	for which RASFF-Information occurred last year	medium	12
d	for which RASFF-Alerts occurred last year	m-high	16
e	for which RASFF-Border rejections occurred last year	m-high	16
f	where in the past, no residue problems occurred at import	m-low	8
g	where in the past, some residue problems occurred at import	medium	12
h	where residue problems occurred recently at import	m-high	16
i	where in the past, residue problems often occurred at import	high	20
Parameter 3: Processing			
Products are …			
a	peeled and washed	low	4
b	peeled (>50% PPP residue reduction)	low	4
c	washed (±25% PPP residue reduction)	m-low	8
d	cleaned (e.g. exterior leaves of lettuce removed)	medium	12
e	only repacked	m-high	16
Parameter 4: Cross-contamination			
The company …			
a	disinfects all transport equipment and storages before a batch is imported	low	1
b	cleans thoroughly every two days to limit cross-contamination	m-low	2
c	has limited concerns for cross-contamination but washes all re-usables	medium	3
d	only washes the floors and machines	m-high	4
e	doesn't take any measures to prevent cross-contamination with PPP residues	high	5

* Risk score = median risk assigned by experts x weights assessed in the linear model (Table 3.6)

The final part of the questionnaire queried the perceived risk of fifteen scenarios that combined different parameter options from Table 3.4 and presented the product chain of a commodity. The risk level of all possible scenarios was estimated and for each (estimated) risk level, three random scenarios were selected. These (fictional) scenarios, presented to the expert panel for scoring, are listed in Table 3.5. The scoring of scenarios was conducted before the separate parameters, as opposed to the order in which they are discussed here. This choice was made to prevent bias or impelling of the experts (Arnold *et al.* 2000).

Table 3.5: Scenarios presented to the expert panel, based on different combinations of values of the selected parameters. The shown risk level (low, medium-low (m-low), medium, medium-high (m-high) represents the median assigned risk of all experts; the weighed parameter scores, the risk calculated with the outcomes of the linear model

Scenarios	Combination of parameter values				Risk level (by experts)	∑ weighed parameter scores (linear model)
Scenario 1	1a	2b	3d	4a	low (1)	36
Scenario 2	1i	2g	3a	4c	m-low (2)	52
Scenario 3	1a	2f	3e	4d	m-low (2)	39
Scenario 4	1c	2f	3e	4e	m-low (2)	51
Scenario 5	1g	2g	3b	4e	m-low (2)	32
Scenario 6	1i	2f	3e	4e	medium (3)	62
Scenario 7	1h	2b	3d	4a	medium (3)	58
Scenario 8	1i	2i	3a	4d	medium (3)	61
Scenario 9	1a	2h	3a	4e	m-low (2)	36
Scenario 10	1k	2f	3c	4e	medium (3)	54
Scenario 11	1j	2g	3e	4a	m-high (4)	73
Scenario 12	1k	2h	3c	4b	m-high (4)	59
Scenario 13	1j	2d	3e	4e	High (5)	81
Scenario 14	1l	2h	3a	4a	m-high high (4.5)	76
Scenario 15	1j	2i	3b	4c	High (5)	75

The designed monitoring tool aims to calculate risk scores that provide an insight how to prioritise between batches for sampling in several situations. Due to a required level of simplicity and ease of use, it was decided to use a simple linear additive model (risk score = ∑ weighed parameter scores) for calculation of these risk scores. When using the average parameter importance as weights in the additive model, there were some discrepancies between resulting additive model yielded risk scores and the risk scores expressed by the experts. As a consequence, certain scenarios received higher risk scores according to the additive model than others, while the opposite was true for the assignments made by the experts. A simple linear program in Maple 17.0 (Maplesoft, Waterloo Maple Inc.) (code presented in appendix) allowed computing parameter weights that respected, as much as possible, the experts' original risk class assignments. The resulting weighing factors are shown in Table 3.6.

Table 3.6: Weighing factors obtained for each parameter with a simple linear additive model based on scenario scores.

	Parameter 1 Supplier	**Parameter 2 Country**	**Parameter 3 Contamination**	**Parameter 4 Processing**	**Parameter 5 Commodity risk**	**Error q**
Weighing factor	0.589	0.188	0.053	0.170	n.a.	0.125
Relative weighing factor*	11	4	1	4	-	-

* To avoid decimal risk scores, only integers were used.

The error term "q" was introduced (and minimized in the linear program) to minimize the extent of contradiction between the experts' opinions and the simple linear additive model (see code in appendix). The non-zero error term "q" in Table 3.6 quantifies the extent to which the linear additive model is unable to completely respect the median assigned classes based on expert evaluations. More to the point, scenarios 14 and 15, respectively belonging to risk class 4.5 and 5 according to the median assigned class based on expert evaluation (Table 3.5), received lower risk scores 3.92 and 3.80, respectively, when computed with the linear additive model (low risk score 1 - high risk score 5). Clearly, scenario 14 receiving a higher risk score (3.92) than scenario 15 (3.80) contradicts the expert's original opinions and points to the limited expressive power of a linear additive model. Correspondingly, scenario 14 receives a higher risk score (76) than scenario 15 (75) in Table 3.5. Risk classes after application of the additive model (sum of the weighed parameter values) and calculated weights are given in Table 3.7.

Table 3.7: Classification of total risk scores in risk classes

∑ Risk scores (all parameters)	**Risk class**
24 - 36	low risk
37 - 52	medium-low risk
53 - 65	medium risk
66 - 74	medium-high risk
75 - 96	high risk

3.4 SAMPLE SIZE CALCULATIONS

3.4.1 GENERAL SETTING

Sample sizes of regular statistical sampling plans can be calculated in many ways (attributive, variable acceptance sampling). However, in a risk-based sampling plan, prior knowledge of batches and commodity risk should also be considered. When the analysis costs and possibly, the indirect costs of erroneously accepting a faulty batch for a given or supposed prevalence of contaminations in different situations, are known, sample size calculations for optimal sampling plans are rather easy (Champernowne 1953; Guthrie Jr. and Johns 1959; Hald 1960). Unfortunately, general prevalences of violations for PPP residues in different situations are unknown. Furthermore, in a setting where batches originate from a multitude of producers and countries of origin, the corresponding increase in unknown variables makes this problem even more complex. Hence, it seemed impossible to compute an optimal sample size in the current setting. The example of an alternative approach in this chapter, was intended to assist food processing or trading companies in dividing their total analysis capacity (number of analyses in preceding years, government/sector organization requirements, costs) over batches of incoming products.

3.4.2 CASE STUDY

To provide an example of the risk-based monitoring tool and combined sample size calculation, a simple fictive purchase scheme of a Belgian processing company is considered below. The company bought 7 tons of apples (Batch 1) and 6 tons of strawberries (Batch 2) from a QMS certified Belgian auction where samples of this batch have already been taken. The analysis results did not indicate any MRL violation and the reports accompanied the batch. Additionally, another 6 tons of strawberries (Batch 3) were bought from a local organic farmer. This farmer has been delivering produce for some time and problems never occurred. To transport the commodities from farm to company, the company utilises its own re-usable plastic palloxes. Once they are empty, they are disinfected and stored for reuse. At the processing company, the apples are transported to a peeling machine in a water current before being cut into small pieces by a cutting machine. The crown of the strawberries is removed manually after which they are fed through the same cutting machine. Finally the apple and strawberry pieces are mingled, weighed and packed in small plastic bowls.

Table 3.8 to Table 3.10 present the parameters that are applicable to the batches in the case study.

Table 3.8: Risk calculation applied for Batch 1 (apples auction).

Parameter	Parameter option	Corresponding parameter value (from Table 3.4)		Risk level (score)
Supplier	Accreditation + analysis results	1a		low (11)
Origin	Belgium did not occur on RASFF last year	2a		medium-low (8)
Product	Apples	-		n.a.
Processing	Washing (transport bath) and peeling	3a		low (4)
Contamination	Disinfection of re-usable materials	4c		medium (3)
Scenario risk			∑=	low risk* (26)

*according risk class from Table 3.7 for the calculated scenario risk

Table 3.9: Risk calculation applied for Batch 2 (strawberries auction).

Parameter	Parameter option	Corresponding parameter value (from Table 3.4)		Risk level (score)
Supplier	Accreditation + analysis results	1a		low (11)
Origin	Belgium did not occur on RASFF last year	2a		medium-low (8)
Product	Strawberries	-		n.a.
Processing	Removal of crowns	3d		medium (12)
Contamination	Disinfection of re-usable materials	4c		medium (3)
Scenario risk			∑=	low risk* (34)

*according risk class from Table 3.7 for the calculated scenario risk

Table 3.10: Risk calculation applied for Batch 3 (strawberries organic farmer).

Parameter	Parameter option	Corresponding parameter value (from Table 3.4)		Risk level (score)
Supplier	Historically no issues, no analysis reports	1h		medium (33)
Origin	Belgium did not occur on RASFF last year	2a		medium-low (8)
Product	Strawberries	-		n.a.
Processing	Removal of crowns	3d		medium (12)
Contamination	Disinfection of re-usable materials	4c		medium (3)
Scenario risk			∑=	medium risk* (56)

*according risk class from Table 3.7 for the calculated scenario risk

The commodity risk also has to be considered in order to assign sample sizes. In this case study only strawberries and apples are present (n=2). From Table 3.3, it can be concluded that apples were more problematic regarding commodity risk than strawberries, due to the higher proportion of consumers possibly exposed to ADI violations. Hence, the assigned commodity risk rank for strawberries is 1 (lowest risk) and apples 2 (highest risk = n). Finally, the batch sizes can be considered and integrated in the calculation.

Table 3.11: Input and calculated sample spreading (according to Equation 3.1 and 3.2)

Input				Output	Sample spreading
Batch B_i	Scenario risk score s_i	Batch size v_i (unit weight)	Commodity risk rank c_i (Lowest risk = 1; n=2)	Batch importance I_i	Batch fractional sampling assignment A_i
Batch 1	26	7 (tons)	2	138	39 %
Batch 2	34	6 (tons)	1	83	23 %
Batch 3	56	6 (tons)	1	137	38 %

In this case study, the batch importance formula suggests assigning 38 % of the sample capacity to the strawberries originating from the local organic farm. The comparable fraction (39 %) of the total sampling capacity assigned to the apples (batch 1), despite the larger batch size and higher commodity risk, is caused by the lower scenario risk score when compared to strawberries. Hence, in this case the perceived risk is indeed more important than the batch size.

3.5 DISCUSSION

Only limited research has been published on how to assign sample sizes in a scientifically founded and risk-based manner that also allows differentiating between various batches, suppliers and exporting countries at a governmental or company level. Sample sizes of regular statistical sampling plans can easily be calculated when prevalences for PPP residues in different situations are known. Unfortunately, such data was not available in the current setting. An alternative approach to assist companies in dividing their total analysis capacity over all incoming batches was constructed and an additive model for computing risk scores was chosen. The total analysis capacity depends on the type of company and the according governmental demands. More specifically, it can be based on the number of analyses in preceding years, government/sector organisation requirements and/or costs and should currently be defined by each individual company.

While the generated scenario ranking closely approximated the risk ranking by experts, such an additive model has some shortcomings, such as the inability to model interaction between different parameters. However, the approach is considered to be a practical and useable tool in which possible further improvements have to be made without sacrificing the ease-of-use. An example of a possible improvement, would be the ability to reserve a portion of the sampling capacity for emerging hazards and surveillance samples.

Discussions with industry and regulatory organizations will have to show if it is possible to introduce (slightly) more advanced techniques, such as robust ordinal regression or dominance-based rough set approaches (e.g., the work by Greco *et al.* (2008 and 2010)). Such techniques are better able to deal with non-linear behaviour of risk factors, a limitation which was encountered in the current work.

According to the expert panel and the consultation of sampling plans, five parameters, consistent with the consulted monitoring plans, were considered to be essential in a risk-based sampling plan. However, some of the mathematically generated weight combinations indicated that, based on our expert panel input, only one parameter (supplier guarantee) could currently be sufficient to classify scenarios into risk classes. If it is desirable to further simplify the methodology, it will have to be evaluated by future research and practitioners. In any case, looking at multiple parameters simultaneously, rather than only depending on supplier guarantee, makes the methodology more robust, and can help when assessing new scenarios.

A possible point of criticism regarding the method, concerns the consumption data on which the commodity risk calculations were performed. Here, the 48 hour recall Belgian consumption database was used (De Vriese *et al.* 2005) as two linked sets of 24 hour data, without taking into account combined exposure. Furthermore, by using the ThMDI, residue levels were assumed to equal the MRLs. Both choices imply that a distorted view of norm (ADI) violations is possible.

Finally, it needs to be stressed that the constructed “batch importance formula” for sample size calculations is one of the possible ways to do so. Consequently, it could be interesting to further develop, validate and adjust this formula in practice. Especially the way in which the commodity risk parameter is handled, by ranking the batches according to the risk of each commodity, rather than scoring these risks absolutely, is a point of attention. What, if anything, would change if this risk parameter were processed in the same way as the other parameters, remains to be discussed. For now, we chose to present the experts with general scenarios that made no mention of a specific commodity, so as to not confound the analysis of the resulting scores.

3.6 CONCLUSIONS

A risk assessment tool based on experts' opinions in order to conduct sampling of fresh produce was developed. The tool calculates a risk score on the basis of information regarding the supplier guarantees, country of origin, commodity processing and vulnerability to cross-contamination. Next, a commodity risk indicator was established, based on an estimated prevalence of ADI infraction in a realistic daily menu. Risk scores calculated with the developed tool, the commodity risk, batch sizes and the importance a company assigns to these batch sizes, were combined here in order to yield an indicator for the sampling importance, even though translating all of these dimensions to a commensurate numerical scale is not necessarily mathematically valid. Nevertheless, the method provides an empirical and easily comprehensible/applicable alternative for governments or at company level.

The presented monitoring tool can be used by Belgian companies and companies from neighbouring countries. Furthermore, with limited adaptations, it could also be of use to governments in order to indicate problem areas. To use it globally, however, adaptations, extended verification in other countries and elaborated expert input from these countries are required. To this end, the outlined protocol in this chapter can help future researchers in collecting the expert input required in order to adapt the proposed approach for use in their countries and with their commodities of interest.

CHAPTER 4 THE INFLUENCE OF MODERN COOKING METHODS ON RESIDUES OF PLANT PROTECTION PRODUCTS ON VEGETABLES

This chapter elaborates on processing parameters, with the aim of covering the effect of only marginally studied cooking methods on plant protection product residues in food. Additionally, a risk assessment for carrot consumption is presented to illustrate the effect of the different processing methods on consumer exposure.

4.1 INTRODUCTION

Consumers can reduce residues of plant protection products (PPPs) on their food by simple processes such as washing (Chauhan *et al.* 2012) or peeling fruits (Bonnechère *et al.* 2012a; Hamilton and Crossley 2004), or preparing vegetables (Keikotlhaile *et al.* 2009; Hamilton and Crossley 2004). The latter has been shown in multiple studies considering the effect of cooking (Aguilera *et al.* 2012; Holland *et al.* 1994; Kar *et al.* 2012; Kaushik *et al.* 2012; Kontou *et al.* 2004; Lentza-Rizos and Balokas 2001; Ling *et al.* 2011; Rasmusssen *et al.* 2003), steaming (Bajwa and Sandhu 2011; Keikotlhaile 2011) and microwaving (Bonnechère 2012; Kaushik *et al.* 2012; Ling *et al.* 2011; Sood *et al.* 2004) of vegetables on residues. However, increasing residue levels were also noted for cooking (Rasmusssen *et al.* 2003) and microwaving (Bonnechère 2012). In Table 4.1, results from literature are catalogued for the processing methods and active ingredients that were considered in this study.

Table 4.1: Summary of consulted literature, dealing with effects of the food processing methods considered in this study, on deltamethrin, lambda-cyhalothrin or tebuconazole. The displayed concentrations and percentages represent the residue on the raw agricultural commodity.

Process	Active ingredient	Matrix	Initial active ingredient concentration (mg/kg) (±SD)	Concentration after processing (mg/kg) (±SD)	Reduction after processing (%)	Reference
cooking	deltamethrin	spinach			-20 %	Holland *et al.*, 1994
		green beens			-50 %	Holland *et al*,.1994
	deltamethrin	apples	0,020 ± 0,004	0,020 ± 0,003		Rasmusssen *et al.,* 2003
	λ-cyhalothrin	apples	0,015 ± 0,003	0,016 ± 0,004		Rasmusssen *et al.,* 2003
steaming	deltamethrin	snow pea			-34 % till -48 %	Bajwa and Sandhu, 2011
	tebuconazole	carrots			-60 %	Keikotlhaile, 2011
		broccoli			-30 %	Keikotlhaile, 2011
microwaving	deltamethrin	spinach	0,183 ± 0,018	0,178 ± 0,011		Bonnechère, 2012
		tea leaves			-24 % ± 7 %	Sood *et al.,* 2004

Plant protection product residues in food are a main consumer concern (Van Boxstael *et al.* 2012) and home cooking is popular (Euromonitor International 2012). Therefore, gaining insight into the effects of different processing methods on the residues of PPPs, present in food, is interesting. A lot of research has already been done in this area of study. However, the use of a wok or grill pan and a microwave oven, has only marginally been covered. Therefore, this study did not only consider the effect of cooking and steaming, but also grilling, wokking and microwaving on PPP residues. Adding salt to the cooking water, was a common instruction in the consulted cookery books. Since Lalah and Wandiga (2002), Radwan *et al.* (2005) and Zhang *et al.* (2007) indicated that this might affect the residue concentration on the vegetables, this was also included in this study. The results were then used to calculate the processing factors as described by Balinova *et al.* (2006) and Bonnechère *et al.* (2012b) and the conversion factors as explained by Boon *et al.* (2009). The results of this study should enable a higher tier determination of consumer exposure to PPPs, through vegetable intake (Hamilton and Crossley 2004). A case study on carrots was conducted to demonstrate the effect of considering processing factors in exposure assessment, for both a deterministic and probabilistic approach.

4.2 MATERIALS AND METHODS

4.2.1 PLANT PROTECTION PRODUCTS

Batches of broccoli, carrots, courgettes and green beans, purchased from an organic vegetable shop, were dipped in a solution of 1 % deltamethrin (Decis plus - Bayer CropScience N.V., Belgium.), 0.1 % tebuconazole (Horizon EW - Bayer CropScience N.V., Belgium) and 0.1 % λ-cyhalothrin (Karate Zeon - Syngenta Crop Protection, Belgium) for three seconds. This was repeated five times (4 replicates + blanco) for each vegetable and processing method. Deltamethrin and tebuconazole are both non-systemic insecticides, while λ-cyhalothrin is a systemic fungicide. The vegetables were then stored in aluminium grill plates and covered with tinfoil, to protect them against light. The vegetables were allowed to dry for 70 hours before processing (19°C, 55 % RH). The green beans were put up in a glass cabinet, with pins on a string, in an attempt to simulate the field situation. They were also covered with a sheet of tinfoil.

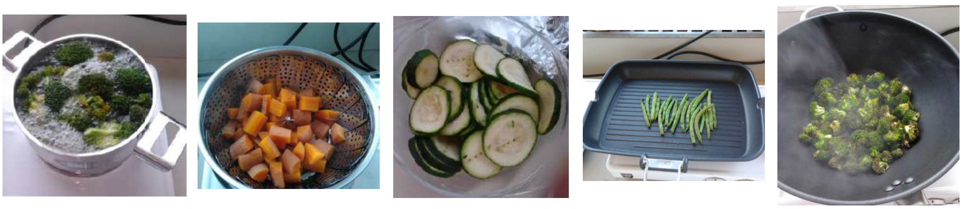
4.2.2 PROCESSING METHODS

Broccoli and courgette samples consisted of one unit, carrots of three units and green bean samples consisted of 50 grams. Before processing, non-edible parts of the vegetables were removed and the vegetables were chopped and weighed. In addition, a sample of each unit was taken to analyze (Appendix B) the residue concentration of each replicate, before processing. Processing was conducted until the vegetables were tender. Processing times follow the instructions from cookery books and literature and are shown in Table 4.2.

Table 4.2: Processing times (minutes) and unit sizes used in this study for the considered vegetables (broccoli, courgettes, carrots and green beans) and for each processing method (cooking, steaming, microwaving, grilling, wokking).

Process	Crop	Broccoli	Courgette	Carrots	Green beans
Cooking	Duration	6 min	10 min	22 min	8 min
	Unit size	florets ± 30 g	slices 0.5 cm	¼ x 3 cm	one bean
Steaming	Duration	7 min	10 min	20 min	7 min
	Unit size	florets ± 30 g	slices 0.5 cm	¼ x 3 cm	one bean
Microwaving	Duration	5 min	4 min	4.5 min	4 min
	Unit size	florets ± 30 g	slices 0.5 cm	¼ x 3 cm	one bean
Grilling	Duration	12 min	4 + 4 min	5 + 5 min	7 + 7 min
	Unit size	florets ± 12 g	slices 0.5 cm	slices 0.5 cm	one bean
Wokking	Duration	11 min	6 min	8 min	9 min
	Unit size	florets ± 12 g	pieces 1 x 1 cm	pieces 0.5 x 0.5 cm	pieces 3 cm

None of the vegetables were washed before processing. Cooking was conducted by placing the vegetables in 1 L of already boiling water. To assess the effect of using salted water, half a broccoli was cooked, while the other half was cooked in salted water (5 g salt/ 1.5 L water). Steaming was carried out by means of a net that hung above the boiling water, without any direct contact with the liquid. Microwaving was performed in a simple microwave oven (Moulinex MO 20 MS) at 750 watt and except for courgettes, 10 mL of water was added to avoid extreme dehydration of the vegetables. Grilling was carried out without addition of grease, which was also the case when using a wok pan. All processes are illustrated in Figure 4.1.



a b c d e

Figure 4.1: Illustration of vegetable processing: (a) cooking broccoli; (b) steaming carrots; (c) courgettes for microwaving; (d) grilling green beans and (e) wokking broccoli.

Dry matter contents of the vegetables were assessed by putting a 20 gram vegetable sample in a stove at 70°C for 48 hours and comparing the weight before and after drying.

Sample extraction and analysis methods for the considered active ingredients are extensively described in Appendix B: Applied sample extraction and analysis methods for deltamethrin, lambda-cyhalothrin and tebuconazole.

4.2.3 Data analysis

Four replicates of each vegetable were analysed for each processing method. Data normality was checked and the applying parametric (e.g. independent t-test or ANOVA) or non-parametric tests (Kruskal-Walllis), were executed. Homoscedasticity was tested with the Modified Levene test and significance of the differences was assessed with the Tukey or Dunnet’s T3 test. Normality assessment may have been influenced because only four replicates were analysed. Therefore, a fifth replicate was considered in the analysis of deltamethrin on beans. For this experiment, normality was proven, so it was assumed that the limited number of replicates did not bias the previous normality assessment, similar to Krol and Arsenault (2000) and Aguilera and Valverde (2012).

4.2.4 CASE STUDY ON CARROTS: TEBUCONAZOLE EXPOSURE MODELLING

Consumer exposure can be assessed by combining residue and consumption data:

Exposure = {residue concentration$_{active\ ingredient}$ (mg$_{active\ ingredient}$/kg$_{vegetable}$) x consumed amount$_{vegetable}$ (kg$_{vegetable}$/person/day)}/ body weight (kg) (Keikotlhaile 2011)

In risk evaluation, exposure is compared to toxicity limits, such as the Acceptable Daily Intake (ADI) and Acute Reference Dose (ARfD) (http://ec.europa.eu/sanco_pesticides). The defintions of these toxicological limits were specified in Chapter 1 (section 1.2.4).

Residue data on carrots for 2009 (Figure 4.2), were obtained from the Belgian Federal Agency for the Safety of the Food Chain (FASFC). Data on deltamethrin and λ-cyhalothrin were limited, so only tebuconazole was considered in this case study. It was assumed that these residue data represented the residue concentration on the raw commodities. Non-detects in the residue database were treated as concentrations at ½ LOR, therefore, this was also done in this study.

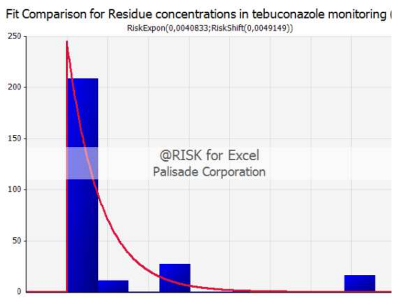


Figure 4.2: Residue concentrations of tebuconazole in monitoring of carrot samples and distribution fit (@Risk). Values on the x-axis were not displayed because of data confidentiality.

Carrot consumption distributions were extracted from the Belgian national consumption database of 2004 (De Vriese *et al.* 2005). Microwaving carrots was not included in the database, but 'reheated carrots' were used as a substitute. The data were converted with the conversion factors from this study, to express every portion as raw agricultural commodity.

4.2.4.1 Deterministic exposure calculation

Deterministic exposure calculations provide a low tier assessment, to assess if there is a course for concern for the defined exposure. The acute exposure (section 1.2.4) to tebuconazole was calculated by multiplying the consumption of the 97.5 percentile consumers ($kg_{carrots}/kg_{bodyweight}$/day) with the 97.5 percentile tebuconazole concentrations ($mg_{active\ ingredient}/kg_{carrot}$) for each processing method (Keikotlhaile 2011). Chronic exposure was calculated similarly, but with the average consumption and residue concentrations, as advised by EFSA (2009).

A slightly higher tier assessment was conducted, by considering the actual intake of residues due to consumption of processed carrots. This was done by multiplying the residue concentrations with the specific processing factors, calculated in this study.

4.2.4.2 Probabilistic exposure calculation

Probabilistic exposure assessment was conducted in the software program @Risk 5.7 for Microsoft Excel 2010 (Palisade corporation, US), wherein the consumption and residue distributions were combined into an exposure distribution ($\mu g_{active\ ingredient}/kg_{body\ weight}$). The number of iterations in the Monte Carlo simulation, was obtained by running five simulations per number of iterations. The variation of the average and percentiles were then compared, favouring 100 000 iterations.

4.3 Results

4.3.1 Concentration after processing

The average (n = 4) active ingredient concentrations for the different cooking processes, expressed as raw agricultural commodity, are shown in Figure 4.3**Fout! Verwijzingsbron niet gevonden.**. Overall effects are difficult to extract, since both the active ingredients and the vegetables often differ in their responses to the applied cooking processes. An unambiguous decline in residues due to cooking broccoli and a residue concentration increase due to microwaving green beans, could, however, be noted.

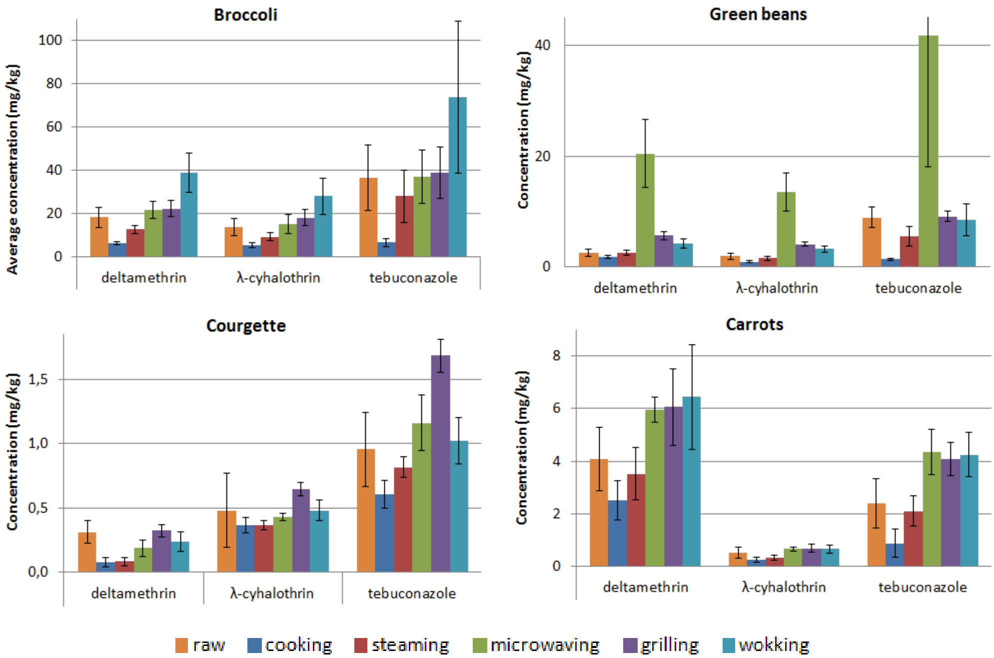


Figure 4.3: Average residue concentrations (±SD), expressed as mg active ingredient/kg raw commodity of deltamethrin, λ-cyhalothrin and tebuconazole for the different processing methods in the four matrices: broccoli, beans, courgette and carrots (n=4).

4.3.1.1 Effect of salt

Cooking a broccoli in salted water, seemed to cause a decrease in deltamethrin and λ-cyhalothrin concentrations, when compared to cooking without addition of salt. The effect of salted water for each active ingredient concentration, is illustrated by an estimated marginal means plot (Figure 4.4). The interaction between the residue concentration and salted water was not statistically significant (p= 0.10).

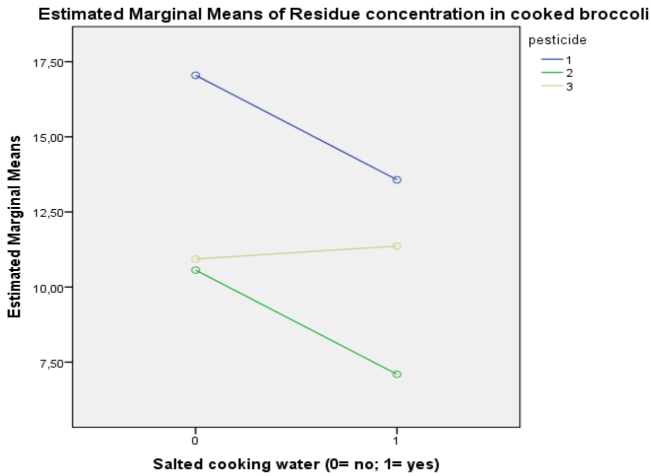


Figure 4.4: Interaction between the active ingredients, (1) deltamethrin; (2) lambda-cyhalothrin and (3) tebuconazole and the addition of salt to the cooking water of broccoli, assessed with a factorial analysis of variance.

4.3.2 CONVERSION FACTORS

The weight of the vegetables generally decreased during processing, except in case of cooking broccoli, where a weight increase was noticed (CF < 1). Conversion factors for cooking with or without salt, were also not statistically different (p = 0.33). Therefore, only one conversion factor was calculated for cooking. The calculated conversion factors (± SD) for all vegetable/processing method combinations, are presented in Table 4.4**Fout! Verwijzingsbron niet gevonden.**.

Table 4.3: Average conversion factors (CF) with standard deviations (SD), for all process - vegetable combinations considered in this study (n=4).

Vegetable Process	Green beans (CF ± SD)	Broccoli (CF ± SD)	Courgette (CF ± SD)	Carrot (CF ± SD)
Cooking	0.97 ± 0.09	0.72 ± 0.06	1.21 ± 0.02	1.04 ± 0.04
Steaming	1.15 ± 0.03	0.96 ± 0.03	1.14 ± 0.03	1.09 ± 0.05
Microwave	3.16 ± 0.40	1.21 ± 0.04	1.19 ± 0.10	1.28 ± 0.04
Grill	1.45 ± 0.03	1.34 ± 0.06	1.48 ± 0.02	1.23 ± 0.03
Wok	1.36 ± 0.06	1.35 ± 0.19	1.30 ± 0.17	1.44 ± 0.07

4.3.3 PROCESSING FACTORS

The Processing Factors (PF) are presented in Figure 4.5**Fout! Verwijzingsbron niet evonden.**. There were no significant differences in PF between the different matrices for cooking, steaming and grilling. Therefore, calculating a mean PF for these processing methods, was meaningfull. The results for microwaving green beens, were significantly different from these for broccoli and courgette.

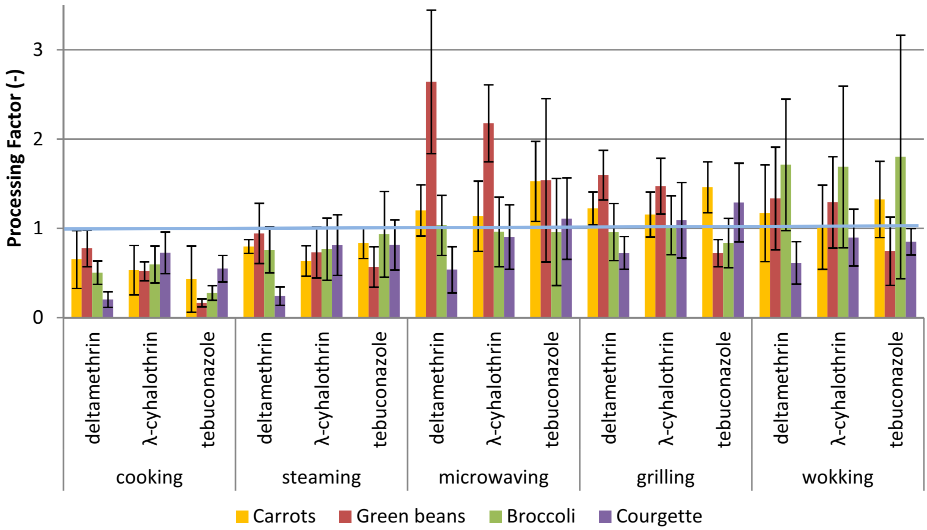


Figure 4.5: Processing factors (± SD) for cooking, steaming, microwaving, grilling and wokking for deltamethrin, λ-cyhalothrin and tebuconazole in different matrices (carrot, green bean, broccoli, courgette) (n=4).

The standard deviation of the average concentration on microwaved green beans (0.72 ± 0.25), was very high, in comparison with the other matrices. Therefore, concentrations on green beans were not included to calculate an average PF for microwaving. Results for wokking of broccoli differed significantly from the other three matrices and was, therefore, not considered in the calculation of an overall PF. The calculated average processing factors (± SD) for the processes in this study, are presented in Table 4.6. The averages are only calculated to give an overview of the overall effect of a processing method. For exposure assessments, PF for the specific matrix-active ingredient combinations should be used.

Table 4.4: Average processing factors (± SD) for cooking, steaming, microwaving, wokking and grilling for the matrices in this study; based on residue concentrations of deltamethrin, lambda-cyhalothrin and tebuconazole. (*excluding green beans; ** excluding broccoli).

Process	PF		SD
cooking	0,49	±	0,19
steaming	0,74	±	0,19
microwaving*	1,04	±	0,27
grilling	1,13	±	0,29
wokking**	1,03	±	0,27

Processing factors for cooking and steaming, lay below the value of 1, indicating a decrease in active ingredient concentration due to those processes. Excluding the high PF for microwaving green beans (2.12 ± 0.55) and wokking broccoli (1.73 ± 0.06), those food preparation methods had little effect on the residue concentration (PF ≈ 1).

4.3.4 DETERMINISTIC EXPOSURE ASSESSMENT FOR TEBUCONAZOLE IN CARROTS

The chronic exposure to tebuconazole, due to carrot consumption, is shown in Figure 4.6. In the conservative approach, monitoring data were used te assess the average and P97.5 residue concentrations. The actual intakes were a combination of these data with the PFs (4.3.3), assessed in this study.

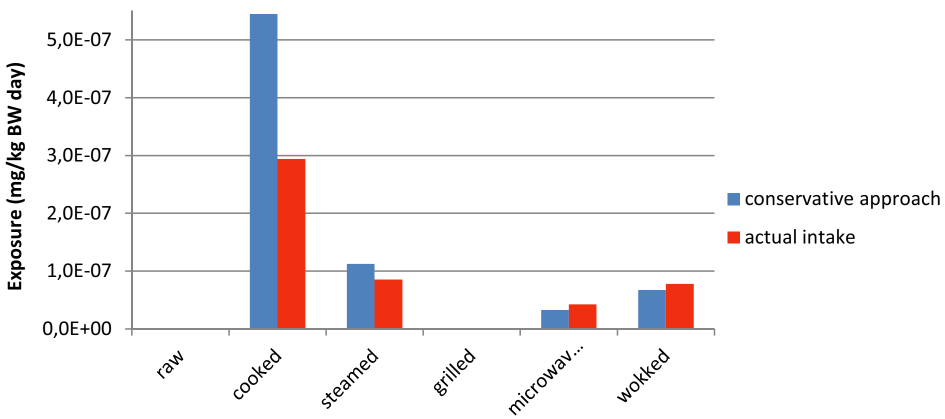


Figure 4.6: Chronic exposure to tebuconazole due to consumption of raw or processed carrots; deterministic calculation (conservative approach based on monitoring data – actual intake assessed by integrating processing factors).

Acute exposures were ten times higher than the chronic exposure that was shown in Figure 4.6. None of the exposures was higher than the toxocological limits for tebuconazole on carrots, which is due to the low amounts of carrots that were consumed and the low number of samples with detected residues.

4.3.5 PROBABILISTIC EXPOSURE ASSESSMENT FOR TEBUCONAZOLE IN CARROTS

The use of distributions and Monte Carlo simulations instead of discrete numbers, resulted in the (example) exposures shown in Figure 4.7. These values are not fixed, since every simulation will result in slightly different figures.

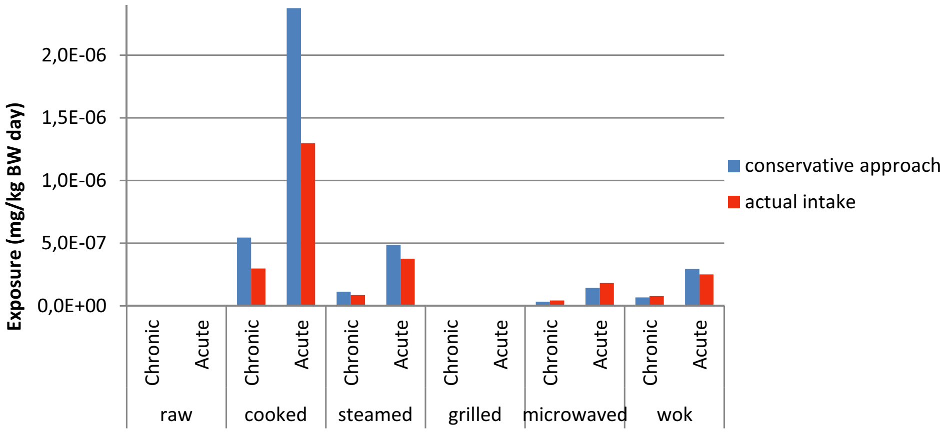


Figure 4.7: Chronic and acute exposure to tebuconazole due to consumption of raw or processed carrots; probabilistic calculation with @Risk (conservative approach based on monitoring data – actual intake assessed by using processing factors).

The actual exposure to tebuconazole was generally lower than the calculations with the conservative approach for cooking (Figure 4.7) and steaming. For the other processes, the actual exposure was slightly higher than calculated conservatively.

Chronic risk assessment of tebuconazole on carrots due to different processing methods, indicated an elevated exposure for all processing methods, except for microwaving, in comparison with consumption of raw carrots. Nevertheless, only 0.05 % or 0.02 % of the ARfD was maximally reached in those cases, respectively calculated with the conservative approach and the actual intake. An example of the tebuconazole exposure due to cooked carrot consumption is shown in Figure 4.8.

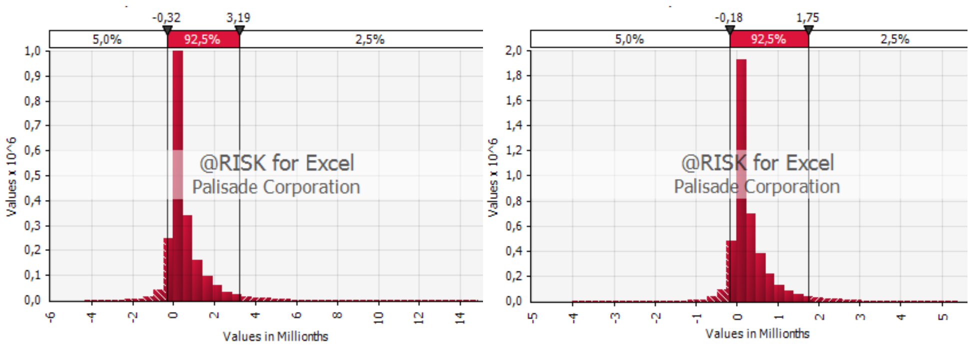


Figure 4.8: Example of @Risk output: exposure to tebuconazole (mg/kg BW day) due to consumption of cooked carrots; (a) conservative calculations, (b) actual intake by considering the PF.

4.4 DISCUSSION

When comparing the concentrations after processing, it was noted that distinct differences between processes did not always show in the statistical output files, even though a clear effect was expected. This shows that the power of these tests was low, probably due to the limited replicates for each processing method in this study. Therefore a critical analysis of the results was necessary. In case of illogical results, the data were reorganised and different statistical tests were conducted (eg. Independent samples t-test instead of an ANOVA when variances were equal).

In this study, the addition of salt before cooking broccoli did not cause a significant effect on the residue concentrations, in comparison with processing without salt. This contrasts with the study of Lalah and Wandiga (2002), who indicated an obvious residue decrease of malathion on maize and beans, caused by added salt. Malathion is a contact insecticide so the results should be comparable with these of deltamethrin and tebuconazole. Besides the effect of the active ingredient properties, it is likely that the difference in results was caused by the fact that the added amount of salt was five times lower in our experiment.

Our results showed a concentration decrease due to cooking and steaming. Residual concentrations after cooking were eacht time lower than after steaming, which was caused by the intense contact of the vegetables with the boiling water which allow the active ingredients to dissolve. In for example broccoli, this was confirmed, since the decrease in tebuconazole (polar, Log $P_{o/w}$ 3.7) concentrations was larger than for deltamethrin (Log $P_{o/w}$ 4.6) and λ-cyhalothrin (Log $P_{o/w}$ 4.6 – 7.0). This demonstrates the results of Ozbey and Uygun (2007), who concluded that a higher water solubility and lower partition coefficient result in a larger residue transfer. Even though this is true for several process-matrix combinations, it was not possible to confirm that the concentrations of active ingredients with a higher Log $P_{o/w}$ were less affected by processing, as opted by Guardia-Rubio *et al.* (2007) and Keikotlhaile (2011).

Processing factors (PFs) for the considered methods were assessed. However, since a difference between matrices was noticed, the calculation of overall processing factors should be nuanced. In this study, the PF for microwaving green beans and wokking broccoli, were significantly higher than in the other matrices.

The conversion factors (CF) were crop specific and generally indicated a weight decrease due to processing. Weight changes for broccoli during cooking, were an exception. This is probably due to the structure of these florets, which can retain some cooking fluid.

The factors (CF, PF) assessed in this study, were used in an exposure assessment to tebuconazole in carrots. The assessment approaches, were slightly improved by considering the conversion and processing factors. Especially the latter allows to calculate more accurate exposures, by considering the actual intake of residues, when consuming processed carrots. The effect and importance of considering processing factors in exposure assessment, was shown by the occurrence of higher actual exposure levels, when compared to the more conservative approach.

4.5 CONCLUSIONS AND FURTHER RESEARCH

The present study demonstrated that: (1) cooking vegetables particularly caused a decline in active ingredient residues; (2) increased residues after using a microwave oven, grill or wok pan, were only distinct in some combinations of active ingredients and vegetable matrices; and (3) weight and concentration changes due to processing are important parameters to consider in exposure assessments.

For further research, field applications of the active ingredients with consideration of the waiting period before harvest is an interesint approach. This might affect the dislodgeable foliar residue on the vegetables and hence the reductive effect of cooking on the residue concentrations. This way, exposure assessments can even be improved to represent the actual risks.

CHAPTER 5 LITERATURE REVIEW: IMPACT OF CLIMATE CHANGE ON PLANT PROTECTION PRODUCT USE[iii]

This chapter reviews scientific literature about climate change, in order to reveal the climate aspects that can directly or indirectly influence plant protection product use and hence, human exposure.

5.1 INTRODUCTION

Current plant protection product (PPP) efficiencies and use, can be influenced in many ways and not in the least, by environmental conditions. Given the general acceptance of major climate change effects, an effect on PPP use is obviously expected. The direction of this effect is, however, uncertain and has not yet been thoroughly investigated. Research for the effects of climate change generally considers multiple aspects and is, consequently, not very detailed, in a way that only limited influencing factors or effects are described. In this review, current knowledge on possible climate change effects on PPP use, was combined and a detailed effect on PPP use, was distilled. Implications of an adapted PPP use for consumer exposure to residues is important at the end of the food supply chain.

In contrast to natural contaminants, produced by micro-organisms or fungi, pesticide residues in food, can be controlled by human actions. Highly toxic crop protection chemicals can, for instance, be replaced by less dangerous or more human and environmental friendly alternatives. In this respect, the food safety issues related to an increased exposure to pesticide residues, as a consequence of climate change, might not occur.

[iii] This chapter was compiled from:

Delcour, I., Spanoghe, P., Uyttendaele, M. (2014). Literature review: Impact of climate change on pesticide use. *Food Research International*, DOI: 10.1016/j.foodres.2014.09.030.

5.2 INFLUENCING FACTORS FOR PLANT PROTECTION PRODUCT USE

Given the multivariate nature of climate change and nonlinear thresholds in natural processes, it is difficult to consider all the links between climate change and PPP use (Harvell *et al.* 2002). Six aspects that directly impact a farmer's use of PPPs, were selected (Figure 5.1). Amongst those six aspects, technological progress, legislation and the economic situation are not directly influenced by the climate, while crop characteristics, pest occurrence and severity and PPP efficiency, are directly influenced by climate. in the following paragraphs, the interactions between those six key aspects and PPP use, are discussed in detail.

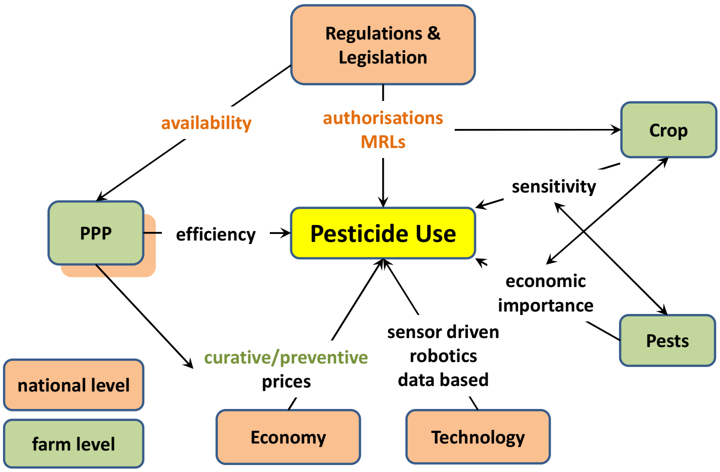


Figure 5.1: Influencing factors for plant protection product (PPP) use and the level at which they have an effect (national or farm level).

5.2.1 REGULATIONS, ECONOMIC SITUATION AND TECHNOLOGICAL PROGRESS

Plant protection product use is strongly controlled through several Regulations included in legislation, which define authorized active substance/crop combinations. The concentration of PPP residues that may remain on the crops after harvest, is regulated by setting Maximum Residue Levels (MRLs). At national level, legislation is, consequently, an important influencing factor for PPP use, as it clearly limits the number and scope of PPP a farmer has at his disposal.

With regard to the economic situation, Eid *et al.* (2007), indicated that high temperatures can constrain agricultural production. Additionally, Turner *et al.* (1994), have shown that a shift in precipitation can slow down the economic development of some nations and particularly affects agricultural production. The economic climate also strongly influences on-farm decisions, by limiting a farmer's pest control options. Firstly, PPP producing companies decide what (authorised) products, active substances and formulations will be marketed in a specific country. To increase a company's profits, focus lies on PPP production for major crops, which is an important issue. This limits the scope of available products for small-scale crops. Secondly, farmers can use preventive or curative PPPs in accordance with the advice of an officer or seller, but in reality, this choice is largely influenced by the purchase and application costs. A good example, is the use of illegal products or obsolete stocks in developing countries, because of the small scope of available products and their high prices (Wassie 2012).

Humans adapt their behaviour to the pressures of climate change (IPCC 2014; Kirezieva *et al*. 2014). An example of adaptation scenarios for pesticides is shown in Figure 5.2. According to Kirezieva *et al*. (2014), an extended use of resistant cultivars, integrated pest management or land-use changes are additional responses that can occur at farm level.

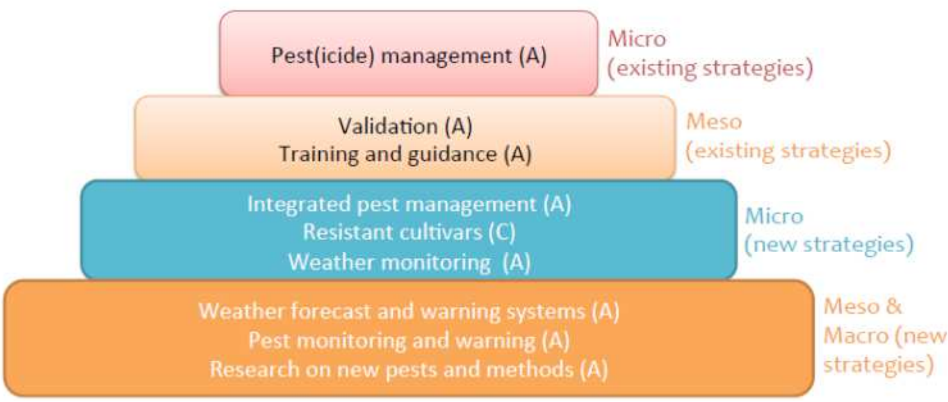


Figure 5.2: Final adaptation scenarios for pesticides, assessed within Veg-i-Trade (directly from Kirezieva *et al*. 2014).

Technological progress also strongly influences PPP application and the residues. Advanced agricultural technologies can be illustrated by, for example, GPS guided field sprayers in modern farming systems. These sprayers are equipped with sensors to determine their location and plant numbers, coverage levels, the amount of biomass or infection levels. A combination of these robotics' recordings, with additional information on required pesticide doses, leads to precision farming, which promotes a more efficient pest/disease control (Dworak 2013). Such targeted applications decrease the required PPP volumes, needed to maintain the same level of crop protection that is achieved with current practices.

5.2.2 CULTIVATED CROPS

Plant protection product adhesion and uptake into a plant is driven by plant growth and soil properties, both strongly liable to climatic influences. Species all have clear characteristic climatic requirements for growth, survival and reproduction that limit their geographic distribution (e.g. need of vernalization), agricultural value, abundance and interaction with other species (Bloomfield *et al.* 2006; Gutierrez *et al.* 2008; Harvell *et al.* 2002). Most food crops are sensitive to direct effects of high temperatures and precipitation extremes or indirect effects of the climate on soil processes, nutrient dynamics and pest organisms (Rosenzweig *et al.* 2001).

Higher temperatures and increased CO_2 concentrations, associated with a substantial change in photosynthetic activity, promote plant growth and expansion (Gutierrez *et al.* 2008; Reilly *et al.* 2003). A high growth rate causes a dilution of the absorbed PPPs in plants, decreasing the PPP residue concentration (Holland and Sinclair 2004; Zongmao and Haibin 1997). In addition, the roots are able to reach deeper soil layers, thus preventing uptake of PPPs that are present in the top layers. If the high temperatures cause a decrease in soil organic matter and an elevated evaporation rate, root uptake is enhanced (Miraglia *et al.* 2009). In some cases, warm and dry conditions increase resistance to plant infections, resulting in a reduced fungicide need (Patterson *et al.* 1999), which is also the case with high atmospheric CO_2 concentrations (Scherm 2004). Physiological plant stress increased host susceptibility and PPP dependency, especially in sensitive crop developmental stages is also possible (Harvell *et al.* 2002; Rosenzweig *et al.* 2001). A lengthening of the active growing season potentially allows for increased farming, introduction of new crops and a northward crop expansion. This ability to grow new or more crops, results in increased PPP use and their introduction in naïve ecosystems (Noyes *et al.* 2009). A temperature variability increase can also adversely affect crops growing at low or high mean temperatures, due to diurnal and seasonal canopy temperature fluctuations that exceed the crop's optimum range (Rosenzweig *et al.* 2001). In conclusion, increased temperatures affect plant productivity (Rosenzweig *et al.* 2001; Scherm 2004; Woodruff *et al.* 2009), giving rise to a potential increase in volume and array of PPPs used (Noyes *et al.* 2009) and a shift in cropping patterns exemplified by, for example, a due change in sowing dates (Eid *et al.* 2007).

Precipitation is the other major determining factor of crop productivity, influencing variations in crop yields, yield quality and pests in both a positive and negative way (Gadgil *et al.* 2002; Gutierrez *et al.* 2008; Patterson *et al.* 1999; Reilly *et al.* 2003; Rosenzweig *et al.* 2001). This duality lies in increased yields, due to more precipitation during the growing season, while intense rainfall can damage younger plants and be detrimental to crop productivity (Rosenzweig *et al.* 2001). Here for, the use of early maturing varieties and crop varieties with high water use efficiency will possibly blossom (Eid *et al.* 2007; Gadgil *et al.* 2002). Plant protection product uptake and transport in plants, are affected by precipitation and are limited in case of decreased transpiration under dry circumstances (Keikotlhaile 2011).

5.2.2.1 Crop-pest interactions

Climatic variation influences the physiology and phenology of the host species, host resistance and growth, all affecting the synchrony between host and parasite (Harvell *et al.* 2002; Perarnaud *et al.* 2005; Rosenzweig *et al.* 2001). In contrast, the most severe and least predictable disease outbreaks occur when altered geographic ranges cause formerly disjunctive species and populations to converge (Harvell *et al.* 2002). This effect is enhanced when a maladjusted population of compatible beneficial insects, to diminish the pest infestations, is present (Rosenzweig *et al.* 2001). Finally, due to cropping intensification, crop rotation reductions, increased areas of perennial crops, introduction of new species or varieties and autumn sowing, the rural landscape changes. These changes influence the location and availability of host plants for pest species and provide a green bridge for pests during winter (Noyes *et al.* 2009; Reilly *et al.* 2003; Roos *et al.* 2011; Rosenzweig *et al.* 2001).

5.2.2.2 Adaptation measures

Simulations show that production in developed countries benefits from the projected climate change, whereas production in developing nations is expected to decline (Rosenzweig *et al.* 2001). In response to climate change and a pronounced decrease in area for specific crops, a change in planting date, shortened maturity dates and an expanded use of better adapted cultivars are decent adaptation measures (Shakhramanyan *et al.* 2013, Reilly *et al.* 2003). In addition, the introduction of more genetically modified organisms might be a solution (Hall *et al.* 2002).

Because of the reduced PPP tolerance of crops under stress, the use of another range of new PPPs will possibly be needed. On the other hand, a shift in the use of certain classes of current PPPs, is the most probable evolution.

5.2.3 OCCURRING PESTS (WEEDS, INSECTS AND DISEASES)

In terms of climate change, temperature increases and precipitation changes, are the main pest infection determinants. Other influencing aspects are dew, atmospheric CO_2 concentration and radiation (Bloomfield *et al.* 2006; Jackson *et al.* 2011; Noyes *et al.* 2009; Rosenzweig *et al.* 2001; Scherm 2004).

Crop damage by pests and diseases is a result of complex ecological dynamics between two or more organisms and are difficult to predict exactly (Rosenzweig *et al.* 2001; Scherm 2004). There are indications that climate change causes phenology and geographic distribution changes in a wide range of ecosystems (Greco *et al.* 2011; Scherm 2004; Seeland *et al.* 2012). As a result of the expected response of single species and communities to climate change (Müller *et al.* 2010), a disturbed temporal synchrony of pest, host and bio-control agents with according yield losses is expected (Fontaine *et al.* 2009; Gutierrez *et al.* 2008; Patterson *et al.* 1999; Perarnaud *et al.* 2005; Scherm 2004). Research shows that pest infestations often coincide with modifications in climatic conditions (Rosenzweig *et al.* 2001), so changes are species and region specific (Noyes *et al.* 2009).

In general, many pest and pathogen species are favoured by warm and humid conditions, of which the latter is the main confounding factor affecting crop-pest interactions (Bloomfield *et al.* 2006; Delorenzo *et al.* 2009; Rosenzweig *et al.* 2001).

5.2.3.1 INSECT PESTS

Although insects flourish in all climates, research reports an earlier appearance and activity in warmer circumstances (Bloomfield *et al.* 2006; Jackson *et al.* 2011; Rosenzweig *et al.* 2001). This is not illogical as temperature affects not only the availability of host plants and refuges, but also improves overwintering, dispersal, migration and population characteristics such as reproduction and growth rates (Jackson *et al.* 2011; Macdonald *et al.* 2005; Patterson *et al.* 1999; Perarnaud *et al.* 2005; Roos *et al.* 2011; Rosenzweig *et al.* 2001). In addition, Harvell *et al.* (2002) reported a negative correlation between temperature and the severity of fungal pathogens of insects. For instance, a surplus of five generations a year of aphids is expected with climate change (Roos *et al.* 2011).

Wet conditions bring on severe insect and plant pathogen infestations (Rosenzweig *et al.* 2001) or effect a geographical shift of some insects (Bloomfield *et al.* 2006). Increases in CO_2 concentration, wind induced dispersal of pests, differences in soil nitrogen content and population density, determine insect abundance (Patterson *et al.* 1999; Roos *et al.* 2011). Finally, extremes seem to have a divergent effect by reducing some species longevity, while others thrive in these circumstances (Perarnaud *et al.* 2005; Rosenzweig *et al.* 2001).

Climate change promotes distribution and abundance of pests due to migration and range shifts, increases pest outbreaks and alters the dissemination of vectors (Gutierrez *et al.* 2008; Hall *et al.* 2002; Jackson *et al.* 2011; Macdonald *et al.* 2005; Midgley *et al.* 2002; Miraglia *et al.* 2009; Noyes *et al.* 2009), all favouring pests compared to crops (Müller *et al.* 2010; Roos *et al.* 2011).

5.2.3.2 Diseases

Plant diseases are mainly affected by temperature, rainfall, humidity, radiation and dew (Patterson *et al.* 1999). Plant diseases can be influenced by altering biological processes of the host, pathogen or disease-spreading organisms. Different life stages may vary in their weather susceptibilities but, the direct effects on pathogens are likely to be strongest (Burdon and Elmqvist 1996). Wet conditions promote the germination of spores, the spread and activity of zoospores and the proliferation of fungi and bacteria (Roos *et al.* 2011; Rosenzweig *et al.* 2001). This is also the case for extreme events and rainfall in particular, which aid the dispersal of diseases (Hall *et al.* 2002; Jackson *et al.* 2011).

Directional climate warming effects are expected to improve pathogen overwintering, development and dispersal, resulting in an elevated disease severity and crop losses (Harvell *et al.* 2002; Roos *et al.* 2011). In case of soil-borne pathogens, a pre-seasonal sclerotial stage was observed after mild winters, leading to more frequent root infections. In addition, these infected plants became more sensitive to above ground pathogens.

Oliveira *et al.* (2009) demonstrated a season-specific response of fungal spore dispersal and concentration to climate aspects. While *Aspergillus* and *Penicillium* spore concentrations were not affected by meteorological factors, divergent responses were found for other fungi. For example, spring-autumn spores of *Pleospora* were negatively correlated with temperature and positively correlated with relative humidity and rainfall. On the other hand, inverse correlations of late spring till summer spores of *Alternaria* with the respective climate elements, were recorded (Oliveira *et al.* 2009). Single abiotic stress factors also seem to impact fungal diseases differently than a combination of factors. Pathogens causing overwintering diseases are affected differently, since milder winters and less snow cover decrease the importance of these pathogens. Late blight incidence on potato on the other hand, is expected to increase in case of warmer springs, summers and more humid conditions in the future. The main cause is a shift to populations with a larger spore production capacity and a shorter infection to symptom interval (Roos *et al.* 2011). Moreover, these short generation times permit a quick adaptation of the fungus to increased host resistance (Scherm 2004).

Given the high degree of complexity in plant-pathogen systems and the extremes, occurring in both climate and disease processes, it is difficult to completely seize the links between both processes (Harvell *et al.* 2002; Roos *et al.* 2011). Nevertheless, an increased disease pressure contributes to population declines, especially for pathogens infecting multiple host species (Harvell *et al.* 2002).

5.2.3.3 Weeds

Because of the susceptibility of crop-weed interactions, local environmental factors benefit either crop or weed (Jackson *et al.* 2011). A temperature increase causes fundamentally altered weed communities and a geographic niche expansion of many species (Jackson *et al.* 2011; Patterson *et al.* 1999). Research also demonstrated that an increased atmospheric CO_2 concentration directly increased weeds' herbicide tolerance and severity (Gutierrez *et al.* 2008), because of the higher carbon dioxide fertilization effect and improved water use efficiency in comparison with agricultural crops (Patterson *et al.* 1999; Rosenzweig *et al.* 2001). In addition, increasing leaf thickness and the partial stomatal closure in this case, reduce herbicide absorption and efficacy (Jackson *et al.* 2011).

5.2.3.4 Plant protection product use

The challenge of pests to agriculture will rise due to increased prevalences of pests, diseases and weeds, which affects PPP efficiency (Miraglia *et al.* 2009; Müller *et al.* 2010; Ntonifor 2011; Patterson *et al.* 1999). In the past, application rates and total amounts of applied herbicides, exceeded insecticides or fungicides (Probst *et al.* 2005), which will probably shift because of pest population favouring climate changes (Goel *et al.* 2005). Weed resistance to herbicides and the according decline in efficacy, can influence the balance (Bailey 2003). A compensatory increased use of agricultural chemicals seems necessary (Hall *et al.* 2002; Rosenzweig *et al.* 2001). In first instance, a shorter infection-symptom interval causes the need for more frequent PPP sprayings to prevent infection (Noyes *et al.* 2009; Roos *et al.* 2011). Secondly, the augmented evolutionary rate of genetically different strains and quick PPP resistance development under warm conditions, might be insufficiently covered by current pest management strategies (Jackson *et al.* 2011). Improved biological control is a solution for populations that become resistant to PPPs (Jackson *et al.* 2011).

In developing countries, easily available, biodegradable, low cost and low risk PPPs are needed for low income peasant farmers and organic farmers (Ntonifor 2011). Especially since those countries will presumably suffer most from climate change. Some countries might even re-introduce or increase the use of banned or restricted PPPs (Macdonald *et al.* 2005).

5.2.4 Plant protection product fate

The level of plant protection is determined by the PPP residue level on that plant, which is in turn influenced by a product's formulation, concentration, dose and application timing and method (5.2.4.1). A PPP can interact with the plant surface, but is also exposed to environmental influences such as wind, radiation and rainfall (Keikotlhaile 2011; Steurbaut 2009). Transport (5.2.4.2) and degradation (5.2.4.3) are the two main routes that affect PPP availability and efficacy. In case of systemic products, a transfer inside plants is essential for pest control (Keikotlhaile 2011). Plant protection product transfer includes volatilization, wash- and runoff and leaching processes, while PPP degradation encompasses photolysis, chemical and microbial breakdown (Figure 5.3). The ecotoxicity (5.2.4.4) and dissipation of PPPs determine the effectiveness of an applied dose and influence the PPP use.

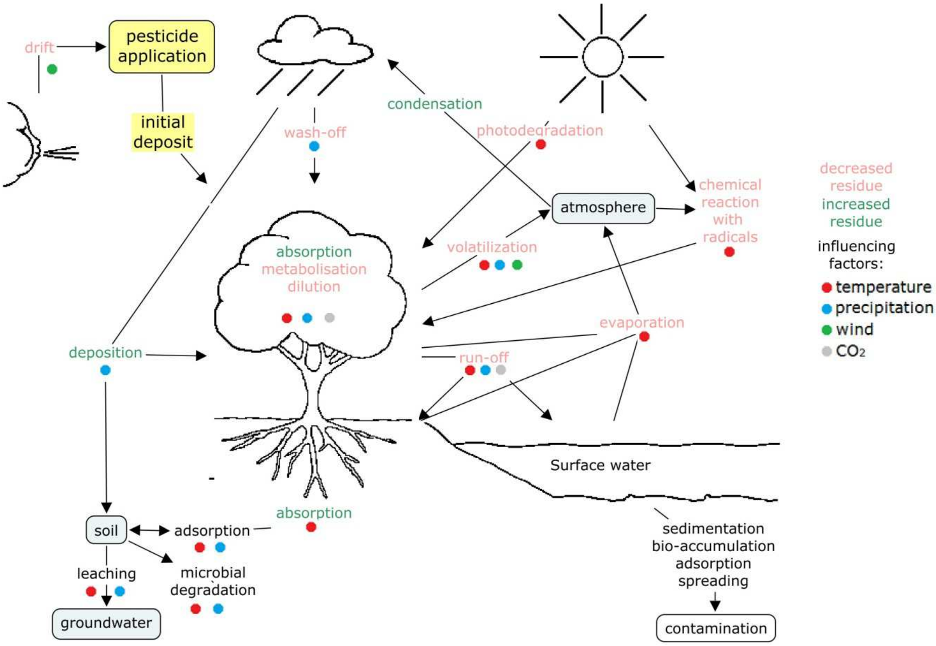


Figure 5.3: Illustration of the environmental factors (temperature, precipitation, wind, CO_2) that influence the plant protection product residue and fate after application and the direction of this influence.

5.2.4.1 Plant protection product application

During PPP application, a large amount of spraying liquid ends up on the soil, depending on drop size, crop density and maturity. The link with extreme events, additionally influences the timing of PPP applications (Johnson *et al.* 1995; Otieno *et al.* 2013). For example, predicted higher soil moisture deficits in autumn, can limit field work or move it to an earlier date than now, while high soil moisture in humid areas, can also hinder field operations (Chen and McCarl 2001; Miraglia *et al.* 2009; Rosenzweig *et al.* 2001). This can oblige farmers to apply autumn herbicide treatments earlier, resulting in a more difficult winter weed control (Bailey 2003).

5.2.4.2 Plant protection product transport

5.2.4.2.1 Volatilization, aerial inputs and wet deposition

Volatilization from soil and vegetation is one of the main causes of PPPs in the atmosphere (Yeo *et al.* 2003) and takes place when a liquid or solid substance transfers to the gaseous phase. Rapid volatilization is mainly due to elevated temperatures, direct exposure to sunlight and a high soil moisture content (Johnson *et al.* 1995; Otieno *et al.* 2013).

Unfortunately, an unambiguous climate change effect on the exchange processes at the interface of the air, soil and vegetation cannot be characterized (van Pul *et al.* 1999). This is exemplified by the studies of Bossi *et al.* (2008), who noted no clear seasonal variation in the concentrations of organochlorine PPPs, while van Dijk and Guicherit (1999) pointed out that this was actually the case for more persistent PPPs.

Studies in the USA and Korea reported higher volatilization rates of organochlorine PPPs during warmer weather (Nations and Hallberg 1992; Yeo *et al.* 2003). In spite of an opposite result for fenhexamid (Schummer *et al.* 2010), this finding has more recently been confirmed for other substances, of which the atmospheric concentrations show a significant positive correlation with temperature (Bloomfield *et al.* 2006; Holland and Sinclair 2004; Navarro *et al.* 2007; Steurbaut 2009). A humid soil after rainfall, favours PPP volatilization (Navarro *et al.* 2007), while a study on the effect of relative humidity, did not record any correlation (Schummer *et al.* 2010).

After volatilization, compounds can be dispersed (van Dijk and Guicherit 1999) from areas with high concentrations and be distributed widely at low concentrations in form of aerial inputs or wet deposition in rain (Bloomfield *et al.* 2006; De Rossi *et al.* 2003; Donald *et al.* 2007; Polkowska *et al.* 2000; van Dijk and Guicherit 1999; Dubus *et al.* 2000). Characteristics of this wet deposition, are an elevated concentration of residues at the beginning of the rain event (Goel *et al.* 2005) and an increased amount of contaminants due to higher intensity and frequency rain and storm events (Noyes *et al.* 2009). Additionally, multiple applied PPPs are less influenced than single ones (Zhang *et al.* 2006) and fluxes linked with the application season of PPPs, were higher in wetter years (Goel *et al.* 2005).

5.2.4.2.2 DRIFT AND RUNOFF

Drift and runoff are the most important transfer pathways of PPPs to other sites or surface waters (Johnson *et al.* 1995; Keikotlhaile 2011; Otieno *et al.* 2013). PPP emission and damage by droplet spray drift, is defined as the amount of PPP that is deflected out of the treated area by the action of air currents (De Schampheleire *et al.* 2007). Farmers can check the wind speed and apply PPPs during drift reducing conditions.

Precipitation is the main driving factor for agricultural runoff and soil erosion (Cryer *et al.* 1998; Ficklin *et al.* 2010; Johnson *et al.* 1995; Otieno *et al.* 2013; Steurbaut 2009). Several studies indicate that increased precipitation enhances runoff contaminated with PPPs (Probst *et al.* 2005; Reilly *et al.* 2003; Turner *et al.* 1994, Carere *et al.* 2011; Oliver *et al.* 2012). Herbicide fluxes were studied separately and were shown to be (Paetzold *et al.* 2007) or not to be (Goel *et al.* 2005) influenced by precipitation volumes.

Effects of higher temperatures were shown by Carere *et al.* (2011), who revealed an altered distribution and partitioning of contaminants in water and by Ficklin *et al.* (2010), who demonstrated a decreasing agricultural runoff load. An unambiguous effect of increasing CO_2 levels on several active ingredients, could not be proven (Ficklin *et al.* 2010).

5.2.4.2.3 LEACHING

Leaching is the downward movement of a chemical through the soil, eventually reaching the groundwater (Keikotlhaile 2011). According to Bloomfield *et al.* (2006), there is an equilibriium with the adsorption to soil particles. Blenkinsop *et al.* (2008) and Nolan *et al.* (2008) concluded that the transfer of PPPs to depth via leaching and to surface water via drainage, were mostly influenced by interactions between weather and soil-PPP combinations. Several studies reported an enhancing effect of precipitation volumes (Bloomfield *et al.* 2006; Roy *et al.* 2001; Woodruff *et al.* 2009) of variable duration (Bloomfield *et al.* 2006; Nolan *et al.* 2008), rainfall seasonality (Bloomfield *et al.* 2006; Loewy *et al.* 2006), intensity (Bloomfield *et al.* 2006) and timing in relation with PPP application (Lewan *et al.* 2009).

Temperature is a main driver for leaching (Bloomfield *et al.* 2006) because it affects soil mineralogy and geochemistry (Woodruff *et al.* 2009). Research describes a negative correlation with leaching (Beulke *et al.* 2004; Bloomfield *et al.* 2006; Nolan *et al.* 2008), often caused by desorption (Stenrod *et al.* 2008). Temperature not only causes a seasonal effect on PPP transport in leaching (Nolan *et al.* 2008), but also reduces the influence of winter rainfall (Blenkinsop *et al.* 2008). This winter rain exhibits an overall strong influence on the more retained and less degraded residues of spring or autumn applications (Blenkinsop *et al.* 2008; Bloomfield *et al.* 2006; Nolan *et al.* 2008).

5.2.4.3 PLANT PROTECTION PRODUCT DEGRADATION

Plant protection product dissipation is not only influenced by PPP transport but also degradation. Degradation of PPPs in the soil or atmosphere is realized by phototransformation or microbial breakdown, while degradation on plant surfaces is caused by photodegradation, evaporation, rainfall elution and growth dilution (Zongmao and Haibin 1997) (Figure 5.3, p 80).

Global warming is acknowledged to accelerate the degradation of chemical components due to higher microbial and chemical reaction rates and may reduce concentrations of PPPs in the environment (Ahmad *et al.* 2003; Athanasopoulos *et al.* 2004; Beulke *et al.* 2004; Bloomfield *et al.* 2006; Borgå *et al.* 2010; Caceres *et al.* 2008; Ismail and Azlizan 2002; Kookana *et al.* 2010; Noyes *et al.* 2009; Singh *et al.* 2003; Stenrod *et al.* 2008; Wang *et al.* 2009b). However, exceptions were discovered in alkaline soils (Caceres *et al.* 2008) and also in case of 2,4-D, which degraded faster at 20°C than 40°C (Shymko *et al.* 2011). Elevated soil moisture contents and increased precipitation, also enhance PPP degradation and accordingly, persistence (Bailey 2003; Ismail and Azlizan 2002; Noegrohati *et al.* 2008; Noyes *et al.* 2009). Furthermore, a higher relative humidity was proven to induce a faster environmental PPP degradation, despite the more difficult initial degradation in this case (Athanasopoulos *et al.* 2004).

5.2.4.3.1 Phototransformation

Photolysis or phototransformation occurs when a molecule absorbs energy from sunlight (UV-irradiation), resulting in a chemical alteration of that molecule (Rosenzweig *et al.* 2001). Because of the kinetic characteristics of these reactions, a temperature effect on phototransformation, is expected (Bloomfield *et al.* 2006; Noyes *et al.* 2009; Rosenzweig *et al.* 2001). A study by Hebert *et al.* (2000), demonstrated the temperature independence of for example chlorpyrifos degradation. However, an effect was shown in a study on triadimefon in which an increased phototransformation rate in water, compared to on-leaf residues, was noted (Nag and Dureja 2003).

5.2.4.3.2 Microbial degradation

The presence of soil micro-organisms also plays an important role in PPP dissipation and transformation (Ahmad *et al.* 2003; Caceres *et al.* 2008; Ismail and Azlizan 2002; Singh *et al.* 2003; Wang *et al.* 2009b). Biological and chemical reaction rates tend to rise at increased temperatures, which is also the case for microbial activity (Chang *et al.* 2007; Ismail and Azlizan 2002; Kookana *et al.* 2010; Wang *et al.* 2009b). The soil moisture content enhances microbial activity, but in lesser extent than the temperature effect (Bailey 2003; Ismail and Azlizan 2002).

5.2.4.4 *Plant protection product ecotoxicity*

Ecotoxicity can be influenced by many processes. For example, adsorption and the formation of non-extractable residues, can be seen as a decrease in toxicity or bioavailability of PPPs, resulting in a higher need for PPP inputs (Barriuso *et al.* 2008).

A positive correlation between increased temperatures and ecotoxicity was reported (Noyes *et al.* 2009; Seeland *et al.* 2012), except for pyrethroids and DDT, which are thought to be more toxic under low temperature conditions (Noyes *et al.* 2009). The temperature effect on PPP ecotoxicity can be clarified by a changing toxicokinetic profile, resulting in an altered absorption and substance elimination (Seeland *et al.* 2012). Studies on *Chironomus riparius*, *Palaemonetes pugio* and *Mya arenaria* demonstrated this increased acute toxicity of applied PPPs at higher temperatures (Delorenzo *et al.* 2009; Greco *et al.* 2011; Seeland *et al.* 2012). For *Physella acuta*, a complex PPP-temperature interaction with a life stage specific and temperature dependent ecotoxicity was discovered. Finally, a combined effect of fungicide exposure and thermal stress was proven to increase the average mortality of *Daphnia magna* (Seeland *et al.* 2012). Indirectly, increasing temperatures can alter the ability of species to respond to PPP exposures or alter PPP uptake and metabolism, thus assumedly increasing PPP ecotoxicity (Greco *et al.* 2011; Patterson *et al.* 1999).

5.2.5 Plant protection product fate under climate change

A combination of the PPP fate and more specifically, the environmental factors that influence the initial PPP deposit, can be seen in Figure 5.3 (p 80).

Climate change can reduce concentrations of PPPs, due to a combination of increased volatilization and accelerated degradation (Noyes *et al.* 2009; Zhang *et al.* 2006), both strongly affected by a high moisture content, elevated temperatures and direct exposure to sunlight (Johnson *et al.* 1995; Otieno *et al.* 2013). The last two elements also influence the chemical alteration of PPPs (Rosenzweig *et al.* 2001). In general, PPP dissipation seems to be benefitted by higher amounts of precipitation, in addition to temperature, degradation and sorption (Stenrod *et al.* 2008). Within leaves, the uptake and release equilibrium of semi-volatile PPPs is reached faster at higher temperatures (Bloomfield *et al.* 2006) and transport through the atmosphere, is predominantly impacted by local surroundings (van Dijk and Guicherit 1999; Yeo *et al.* 2003). The timing and intensity of rainfall, mainly influences PPP persistence and efficiency (Bailey 2003; Rosenzweig *et al.* 2001). In addition, temperature and light affect PPP persistence through chemical alteration. This results in an effect on the PPPs used to control and/or prevent pest outbreaks (Rosenzweig *et al.* 2001). In general, a warmer climate may necessitate an increased PPP usage (Noyes *et al.* 2009; Rosenzweig *et al.* 2001). Even though the lifetime of some products might be increased (Jager *et al.* 1998), a reduced period of effect, results in more applications that are necessary to sufficiently protect crops.

5.3 GENERAL EFFECT OF CLIMATE CHANGE ON PLANT PROTECTION PRODUCT USE

Several elements that can influence PPP use, have been presented. In first instance, PPP producing companies will strive to supply optimal products. Active ingredients will have to be formulated in a rain-fast product for agricultural use. For farmers, the season and timing of the PPP application, seasonal precipitation and temperature in relation to environmental factors, will strongly influence management decisions (Nolan *et al.* 2008; Reilly *et al.* 2003).

Secondly, climate change affects crop characteristics and appearance due to a lengthening active growing season (Myneni *et al.* 1997). Corresponding advances in phenology are to be expected (Fontaine *et al.* 2009; Lepetz *et al.* 2009), while climatic variation can alter plant resistance for pests but also for PPPs (Harvell *et al.* 2002). According to Reilly *et al.* (2003), overall climate change will be beneficial to crop productivity, despite the risks at regional levels. Local climates, will strongly determine which areas will remain suitable for a crop to be cultivated (Jackson *et al.* 2011; Myneni *et al.* 1997).

Third, the key factor for PPP use is the presence and severity of weeds, pests and diseases in a crop. These organisms are affected by climate change in a similar way as crops are. There is also a high likelihood of genetic adaptation (Bloomfield *et al.* 2006; Lepetz *et al.* 2009), although the first response will probably be a phenology alteration or geographical redistribution (Scherm 2004). Pest and disease invasions are aided by mostly temperature effects. Finally, PPP efficiency, represented by the initial deposit, PPP fate and (eco-) toxicity, also has a major impact on PPP use.

In general, PPP losses of mobile active substances are mainly influenced by the time gap between extreme weather events and PPP application (Blenkinsop *et al.* 2008; Nolan *et al.* 2008). In soil transport of PPPs is mainly driven by rainfall seasonality, intensity and temperature increases, but also land-use changes (indirect long term impact) (Bloomfield *et al.* 2006). The soil microbial activity is affected by moisture content and soil temperature (Barriuso *et al.* 2008). Even though some reducing effects were found, increasing temperatures will overall result in higher volumes of PPPs that will have to be applied more often. An increased intensity of PPP use is expected in form of higher amounts, doses, frequencies and different varieties or types of applied products (Bloomfield *et al.* 2006; Goel *et al.* 2005; Hall *et al.* 2002; Miraglia *et al.* 2009; Noyes *et al.* 2009; Rosenzweig *et al.* 2001).

CHAPTER 6 EFFECT OF RAIN INTENSITY ON FUNGICIDE RESIDUES AND DIFFERENT FORMULATIONS IN AN APPLE ORCHARD

This chapter starts with a literature review of different crop, PPP and rain characteristics that affect rain fastness, or the ability to withstand rainfall. Then the effect of climate change induced rain intensity changes on different formulation types are assessed, to reveal solutions for possible PPP efficiency decreases.

6.1 INTRODUCTION

Current climatic projections indicate an increase in precipitation intensity in the future due to climate change. Since PPP efficiency is sensitive to not only plant characteristics, but also environmental influences, an important impact of precipitation shifts is expected (Heijne and Anbergen 1996; Hunsche *et al.* 2007; Northover *et al.* 1986; Schepers 1996; Smith and MacHardy 1984; Willis and McDowel 1987; Xu *et al.* 2008).

Rain fastness affects the the efficacy of foliar-applied PPPs. Generally, rain fastness is achieved when PPPs have dried, or have been absorbed by the plant (Wells & Fishel 2011). The effect of specified rain characteristics on the initial deposit and rain fastness has already been extensively studied. Firstly, the large effect of small volumes was shown by Pick *et al.* (1984) and Xu *et al.* (2008). Additionally, a hyperbolic and linear decrease up to 80 % of the initial deposit, with increasing volumes, were depicted by respectively Smith & MacHardy (1984) and Hunsche *et al.* (2007). Secondly, studies on the influence of different drying times generally revealed a positive effect of increased drying periods (Bruhn and Fry 1982; McDowell *et al.* 1985; Pick *et al.* 1984; Schepers 1996), however this effect was not always seen (Hunsche *et al.* 2007). Third, rain intensity experiments revealed an increased impact on crops (Kudsk and Mathiassen 1991; Simmons 1980) and larger deposit decreases when rain intensities were high (Bruhn and Fry 1982; Hunsche *et al.* 2007; Kudsk and Mathiassen 1991; Schepers 1996; Smith and MacHardy 1984). In some experiments, however, different rain intensities did not affect the PPP deposits (Fife and Nokes 2002; Pick *et al.* 1984). As suggested by Chen & McCarl (2001), the overall reducing effect of all these rainfall properties will probably cause an increase in use of PPPs and hence, impact food safety.

The effect of different plant characteristics, mentioned by Hull (1970), has been studied by Cabras *et al.* (2001), who showed that PPP deposits on leaves were twice as high as those on fruits. Other studies indicated the importance of the sampling zone within a tree (Hall *et al.* 1997; Poulsen *et al.* 2012; Smith and MacHardy 1984; Xu *et al.* 2008).

Finally, PPP properties, such as the active ingredient, formulation type and adjuvants, were mentioned to influence residue deposits (Hull 1970). Cabras *et al.* (2001) demonstrated a higher rain fastness of systemic, than contact products and Ebeling (1963), van Bruggen *et al.* (1986) and Kudsk & Mathiassen (1991) revealed a higher rain fastness of liquid (EC, SC) than solid (WP, WG) formulations. Unfortunately, a combination of varying rain intensities with some of the above mentioned aspects, has only marginally been studied. An overview of rain intensity studies and their resuls, is given in Table 6.1.

This study focussed on the effect of varying rain intensities on the difference in rain fastness between different formulation types of an active ingredient. Focus lay on fungicides (captan, mancozeb and triadimenol), because of the prominent share of fungicides in the total PPP use in Western Europe and more specifically, apple orchards in Flanders (Lenders *et al.* 2008). The first experiment handled the differences in initial deposit and influence of rain intensity for captan (WG) and triadimenol (EC, SC) for: (1) samples taken from the inside and the outside of the tree canopy; (2) apples versus leaves; and (3) an EC versus SC formulation. To reduce result variability, a second experiment was conducted on individual clipped branches. This experimental set-up served to compare the effects of rain intensity on (1) a WP and WG formulation of mancozeb; (2) an SC and EC formulation of triadimenol; and (3) leaves versus apples, in a better controllable lab environment. The third and fourth experiment ran simultaneously. The respective goals of these tests were (1) to assess the effect of drying time and (2) to check whether lower rain intensities would have affected the previously obtained results.

Table 6.1: Overview of the studies considering rain intensity effects on PPP residues, the rainfall characteristics and their main conclusions.

Crop		Plant protection products		Precipitation				Other conclusions	Reference
Crop	**Tested tissues**	**Active ingredients**	**Formulation**	**Applied volume**	**Intensity**	**Drying period**	**Residue after rainfall**		
apple	seedlings	captan	WP	5 - 8 mm (lab)	60 mm/h	24h	± 20 % (hyperbolic decrease)		Smith & MacHardy (1984)
	whole plant			> 25 mm (orchard)	-	< 48h	± 20 % (linear decrease)		
apple, grapes, pear	whole plant	captan	WP	0 - 94 mm (spread over 18 days)	-	-		Natural rainfall events result in contrasting effects: no indistinct direction distillable	Frank, *et al.* (1985)
cherries, peaches	fruit	captan	WP	8 - 51 mm	-	-	< 50 %		Northover, *et al.* (1986)
apple	whole plant	captan	WP	6 - 28 mm	-	-	-	Decreased efficiency of active ingredients towards *V. inaequalis* after both natural and simulated rain	Heijne & Anbergen (1996)
		dithianon							
apple	fruit, leaves	captan	WG	1, 2, 3, 6, 9 and 12 mm	1.5 mm/h	24h	1 mm: 50 %	Environment controlled in climate room	Xu, *et al.* (2008a)
apple	whole plant	captan	WG	-	-	-	Decrease (%) depends on initial deposit	Field tests resulted in a high result variability. A difference between inner and outer canopy was seen.	Xu, *et al.* (2008b)
potatoes	whole plant	mancozeb	WP and EC, SC	3.7 mm (pH 4.6)	11 mm/h	-	19 %	Retention WP < EC, SC	Van Bruggen *et al.* (1986)
		maneb					18 %		
		chlorathalonil					77 %		
		triphenyloxide					42 %		
		copperhydroxide					73 %		
peas and apple	whole plant, leaves	mancozeb	SC, WP	3 mm	3, 9 and 27 mm/h	24h	Wash-off: 3 < 9 < 27 mm/h	Retention WP < SC	Kudsk & Mathiassen (1991)
		maneb							
grapes	fruit, leaves	folpet + phthalimide	WG	15, 30 and 45 mm	60 mm/h	1, 3, 5 and 10 days	grapes: 100-85 % leaves: 100-90 %	Retention folpet > mancozeb	Cabras, *et al.* (2001)
		mancozeb	WP				grapes: 60-65 % leaves: 80-70%		

Crop (continued)		Plant protection products		Precipitation				Other conclusions	Reference
Crop	Tested tissues	Active ingredients	Formulation	Applied volume	Intensity	Drying period	Residue after rainfall		
apple	seedlings	mancozeb	WG	5, 10, 20 and 30 mm	0.5, 5 and 48 mm/h	2, 4 and 24h	Hyperbolic decrease		Hunsche, *et al.* (2007)
cotton	whole plant	endosulfan	EC	1, 2, 5 and 10 mm	15-18 mm/h	1, 4, 7, 24 and 72h	1 h (1-10 mm): 79-28 % 5 mm (1-72u): 38-90 %	Rain intensity has no significant effect on active ingredient residues on the plant	Pick, *et al.* (1984)
		carbaryl	WP				1 h:58-30 % 5 mm: 39-86 %		
		carbaryl	SC				1 h: 96-49 % 5 mm: 56-100 %		
		cypermethrin	EC				1 h: 66-56 %		
	whole plant	parathion	EC, WP	1, 3 and 5 mm	20 and 56 mm/h	0.5, 1.5 and 2.5h	1 h (1-5 mm): 58-27 % 3 mm (0.5-2.5u): 15-50 % 1 h:32-20 % 3 mm: 35-34 %		
cotton	whole plant	parathion	EC	51 mm	53 mm/h	2, 6, 29, 50, 98, 146h	2-146 h: 27-99 %	Large variability in results	McDowell, *et al.* (1985)
		toxaphene	EC				2-146 h: 86-92 %		
		fenvalerate	EC				2-146 h: 88-95 %		
potatoes	whole plant	fluazinam	SC	10 mm	8 and 38 mm/h	4h, 4d and 6d		Longer drying times increased efficiency of residues. Rain induced redistribution of deposit with increased protection against *P. infestans* (with bio-assays).	Schepers (1996)
		maneb + fentinacetate	WP						
tomato	leaves	chlorothalonil		10, 20, 30, 68 and 150 min	13, 25, 51 and 76 mm/h	5h		Rainfall duration and intensity correlated with residue after rainfall event	Fife & Nokes (2002)
apple	leaves	copper oxychloride	WP	1, 2, 3, 4 and 5 mm	5 mm/h	3h	1 mm: 65 %	Additives significantly affect active ingredient adhesion	Hunsche, *et al.* (2011)
		copper hydroxide	WP				1 mm: 40 % 2 mm: 40 % 5 mm: 25 %		

6.2 MATERIALS AND METHODS

6.2.1 FUNGICIDES

Three fungicides, of which two different formulations were authorized in Belgian apple orchards, were selected: triadimenol suspension and emulsifiable concentrates (Exact 312 SC and Shavit 250 EC - Makhteshim-Agan Chemical works Ltd., the Netherlands), mancozeb water dispersible granules and wettable powder (Prozeb 75 WG and Prozeb 80 WP - Belchim Crop Protection, Belgium) and captan water dispersible granules (Merpan 80 WG - Makhteshim-Agan Chemical works Ltd., the Netherlands). The captan wettable powder was not available on the Belgian market and hence omitted from the trial. The captan WG formulation was retained, to allow a comparison with other rain intensity studies conducted with captan. Triadimenol is the only systemic active ingredient and mainly targets powdery mildew diseases, while both captan and mancozeb are contact fungicides, preventing apple scab caused by *Venturia inaequalis*.

6.2.2 RAINFALL SIMULATION

The experiments were conducted with the rainfall simulator of the Ghent University department at Bottelare (Belgium), represented in Figure 6.1. Briefly, the rain simulator consists of a large metal tray above which a horizontal spraying boom, with three nozzles (60 cm apart), can move at 1.5 m height. The spraying boom is driven by an engine connected chain, which allows a uniform distribution of the spraying liquid. Rain intensities are regulated by setting the volume, pressure and duration of the rain application. The lowest technically possible rain intensities with this simulator, were achieved by utilizing nozzle types TF-VS-10 ('torrential rain'), TF-VS-4 ('heavy rain') and TF-VS-2 ('light rain'), giving different droplet sizes, at a constant volume of 4 L/m². This resulted in rain intensities of respectively 720, 360 and 180 L/h m². Rain quantities were controlled each run with rain gauges. Plants were left on the spraying tray until dry and subsequently transported to the lab, where residue analyses were conducted the next day.

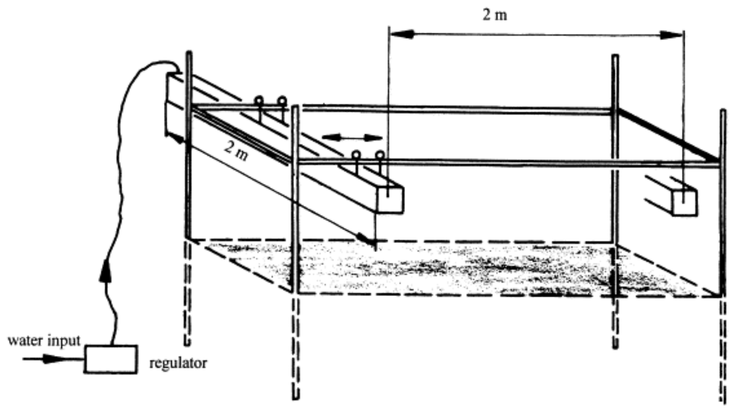


Figure 6.1: Schematic representation of the rain simulator of the University College of Ghent (Singh *et al.* 1997), with own adaptations.

6.2.3 EXPERIMENTS

6.2.3.1 EXPERIMENT 1: FIELD TRIAL

The field trials were conducted in a fifteen-year old Golden Delicious apple orchard, on a sandy soil type in Lokeren, Belgium on September 4th (triadimenol and captan) and October 2nd (mancozeb), 2012. Weather characteristics for both days are given in Table 6.2. Trees (3 per formulation type) were sprayed at 1 pm with a manual knapsack sprayer, equipped with an Albuz TVI 80-025 lilac air induction hollow cone nozzle at a pressure of approximately 4 bar. The trees were sprayed until runoff, which equaled about 1 liter of spraying solution. Concentrations of active ingredient in the spraying solutions were: 8 g/L captan WG, 0.5 g/L triadimenol EC, 0.624 g/L triadimenol SC, 3.75 g/L mancozeb 75 WG and 4 g/L mancozeb WP, respectively. Each formulation was sprayed in another row of trees and treated rows were at least 15 meter apart. After a waiting period of 24 hours, the branches (3 x 3 per formulation type and rain intensity + 3 blanks) and apples (3 x 1 per formulation type and rain intensity + 3 blanks) were clipped and transported for rainfall simulation, as described in 6.2.2. Since a different initial deposit on the interior and exterior of the tree canopies was expected (Hall *et al.* 1997; Poulsen *et al.* 2012; Smith and MacHardy 1984; Xu *et al.* 2008), both locations were sampled separately.

Table 6.2: Local weather characteristics in the apple orchard during the field trials on September 4th and October 2nd (at 1 pm).

Weather characteristics	**September 4th**	**October 2nd**
Temperature (°C)	26.2	17.1
Relative humidity (%)	58	71
Dew point (°C)	17.3	11.8
Air pressure (hPa)	1020.2	1011.3

6.2.3.2 Experiment 2: Lab experiment with branches

In the second experiment, we attempted to attain a more uniform initial deposit, by adapting experiment 1 to a laboratory environment. Clipped branches (± 60 cm - 3 x 3 per formulation type and rain intensity + 3 x 3 blanks) were placed in glass bottles filled with water, to avoid wilting. Fungicide application took place in the open air, with a small spray bottle with pressure regulation. The spraying solutions contained respectively 1.8 g/L Prozeb, 2 mL/L Shavit and 2 mL/L Exact and were sprayed until runoff. Merpan 80 WG was not utilized, because of the possible interference with the residues from the grower's captan applications. After four hours of drying in the open air, the branches were stored in the lab (20 °C, 50 % RH) for another 20 hours before the rainfall simulation. The experiment with mancozeb took place after harvest (October 8th), so there were no apples present anymore.

6.2.3.3 Experiment 3 & 4: Lab experiments with apples

Organic Jonagold apples, with an average weight of 100 g, were purchased and treated by immersing the apples (3 x 1 apple per formulation type and drying time/rain intensity + 3 x 1 blank) in a fungicide solution of 4 g/L captan WG, 1 g/L triadimenol EC, 1 g/L triadimenol SC, 2.4 g/L mancozeb WG and 2.4 g/L mancozeb WP. The treated apples were put on a string with pins and the fungicide deposit was allowed to dry in the lab (18 °C, 55 % RH) until rainfall simulation (next day).

In experiment 3, the effect of waiting period, or time between fungicide application and rainfall event, was assessed. After a waiting period of 2, 4 and 24 hours, the apples were exposed to a rainfall intensity of 12 L/h, obtained by spraying the apples for 9 seconds with 30 mL of tap water, from a small spray bottle at constant pressure (pressure not adjustable).

In experiment 4, the effect of rain intensity was assessed under well controlled lab conditions to complement the long drying period and high intensities, obtained in experiment 1. A waiting period of 4 hours was selected and the apples were exposed to rain intensities of 0.5 L/h (light rain) and 12 L/h (heavy rain).

6.2.3.4 Fungicide analysis

In each experiment, three blanks (3 x 3 branches or 3 x 1 apple) were sampled and analysed before the fungicide application to assess the concentration already present on the leaves and apples. If the fungicides, used in the described experiments, were detected on the blank samples, concentrations were recalculated to assess the actual applied initial deposit. The fungicide residues on the apples and leaves, were analyzed as described in Appendix C.

6.2.4 Statistical analysis

Experiments were conducted with three replicates per treatment. A Kolmogorov-Smirnov test was used to check data normality. A factorial analysis of variance was conducted and the applying parametric (e.g. independent t-test or ANOVA) tests, were executed. Homoscedasticity was tested with the Modified Levene test and significance of the differences was assessed with the Scheffe or Dunnet's T3 test. To allow comparison of the different samples, the residue after rainfall was expressed in relation to the initial deposit before the rain event.

6.3 Results

6.3.1 Experiment 1: Field trial

Residue concentration measurements on brances from the inside and outside of the orchard trees, showed a significantly higher initial concentration of fungicides at the outside of the tree canopies for all analysed active ingredients, formulations and plant tissues (apple versus leaves) (Table 6.3, Table 6.4). The concentrations of initial deposits of the different fungicides, could not be directly linked to the applied amounts of active ingredient (g/tree). However, when compared relative to each other, a good repeatability of the fungicide application method showed.

Rainfall caused an average decline of 29 ± 7 % of the captan concentration on leaves ($p = 0.001$), while the difference between the three simulated rain intensities was not statistically significant for the inner, nor the outer canopy samples ($p = 0.83$). On apples, an average decline of 58 ± 14 % of the captan residue after rainfall, was detected ($p < 0.001$). In these samples, a significant effect ($p = 0.01$) of rain intensity on rain fastness was also noted. The concentration difference after the rain treatments was, however, only statistically different between light and heavy rainfall. On average, rain fastness of residues under the lowest rain intensity and when assessed on the inside of the trees, was highest.

Table 6.3: Residue concentrations (± SD) of captan, formulated as a wettable powder, on leaves and apples from the inner/outer of the canopy after exposure to a light (180 L/h m²), heavy (360 L/h m²) or torrential (720 L/h m²) rain (n = 3).

		Captan (WP)			
		Leaves		Apples	
Sampled zone	**Rainfall treatment**	**Absolute residue concentration (mg/g) (±SD)**	**Relative residue (%) (±SD)**	**Absolute residue concentration (mg/kg) (±SD)**	**Relative residue (%) (±SD)**
branches inner canopy	control	0.65 ± 0.10		14.74 ± 1.22	
	light	0.46 ± 0.10	74.2 ± 28.6	9.29 ± 1.22	62.8 ± 4.4
	heavy	0.51 ± 0.10	82.6 ± 39.0	4.84 ± 1.22	32.3 ± 7.7
	torrential	0.44 ± 0.10	69.1 ± 19.1	4.53 ± 1.22	31.8 ± 20.8
branches outer canopy	control	1.29 ± 0.10		34.45 ± 1.22	
	light	0.83 ± 0.10	65.1 ± 23.7	19.44 ± 1.22	56.6 ± 8.0
	heavy	0.81 ± 0.10	64.2 ± 17.4	10.10 ± 1.22	29.0 ± 7.0
	torrential	0.91 ± 0.10	69.5 ± 8.1	12.96 ± 1.22	37.5 ± 2.6

The residue analyses for triadimenol are presented in Table 6.4. The formulation type (EC versus SC) did not significantly affect the initial deposit.

Table 6.4: Residue concentrations (±SD) of triadimenol, formulated as a emulsifiable and suspension concentrate, on leaves and apples from the inner/outer of the canopy after exposure to a light (180 L/h m²), heavy (360 L/h m²) or torrential (720 L/h m²) rain (n = 3).

		Triadimenol (EC)				Triadimenol (SC)			
		Leaves		Apples		Leaves		Apples	
Sampled zone	**Rainfall treatment**	**Absolute residue concentration (mg/g) (±SD)**	**Relative residue (%) (±SD)**	**Absolute residue concentration (mg/kg) (±SD)**	**Relative residue (%) (±SD)**	**Absolute residue concentration (mg/g) (±SD)**	**Relative residue (%) (±SD)**	**Absolute residue concentration (mg/kg) (±SD)**	**Relative residue (%) (±SD)**
inner canopy	control	0.15 ± 0.01		1.73 ± 0.20		0.16 ± 0.01		1.53 ± 0.24	
	light	0.14 ± 0.01	93.8 ± 4.6	1.45 ± 0.18	73.9 ± 30.5	0.12 ± 0.01	75.7 ± 6.3	1.17 ± 0.08	77.3 ± 9.2
	heavy	0.14 ± 0.01	91.0 ± 6.9	1.49 ± 0.08	86.9 ± 9.0	0.13 ± 0.01	82.0 ± 2.1	1.15 ± 0.05	75.6 ± 8.7
	torrential	0.12 ± 0.01	80.0 ± 1.4	1.33 ± 0.13	76.8 ± 4.3	0.10 ± 0.01	62.6 ± 8.3	1.27 ± 0.09	84.9 ± 18.2
outer canopy	control	0.17 ± 0.01		2.13 ± 0.12		0.19 ± 0.01		2.62 ± 0.05	
	light	0.15 ± 0.01	88.8 ± 8.5	1.51 ± 0.18	70.9 ± 6.4	0.15 ± 0.02	80.5 ± 10.6	2.27 ± 0.36	86.8 ± 14.3
	heavy	0.14 ± 0.02	83.9 ± 11.6	1.55 ± 0.11	72.7 ± 3.3	0.15 ± 0.02	78.3 ± 4.6	2.16 ± 0.22	82.4 ± 9.2
	torrential	0.13 ± 0.01	79.4 ± 9.6	1.64 ± 0.12	76.8 ± 2.1	0.11 ± 0.01	64.8 ± 8.6	2.04 ± 0.30	77.6 ± 10.1

Rainfall caused an average decline of 21 % of the initial triadimenol deposit ($p < 0.001$). Torrential rain caused a significantly lower residual concentration of the triadimenol EC formulation on leaves than light rainfall. The differences for the triadimenol SC formulation were significant between torrential and light rain and torrential and heavy rain. The difference between both formulation types was also calculated and revealed a lower rain fastness of the SC than the EC formulation ($p < 0.001$). On average, rainfall resulted in a 22 % decline of triadimenol residue on apples ($p < 0.001$). The apple samples did not reveal a significant ($p_{EC}= 0.25$, $p_{SC}= 0.83$) effect of rain intensity, nor an effect of formulation type on rain fastness ($p = 0.78$).

Finally, a comparison of the initial deposits of captan and triadimenol revealed that the triadimenol deposit on leaves was at least twice as large as the captan deposit ($p < 0.001$), while on apple samples no difference was seen.

6.3.2 Experiment 2: Lab experiment with branches

Mancozeb residues on the individually treated branches showed a comparable initial deposit of Prozeb 80 WP and Prozeb 75 WG before rainfall ($p = 0.71$), when the initial spraying concentration was taken into account. Application of 4 mm of rain, resulted in an average decrease of mancozeb residue of 23 % for the WP ($p = 0.02$) and 14 % ($p = 0.03$) for the WG formulation. The difference between both formulations was, however, not significant ($p = 0.11$). Residual mancozeb after rain simulation is shown in Figure 6.2 and was statistically different for heavy and torrential rain ($p = 0.03$).

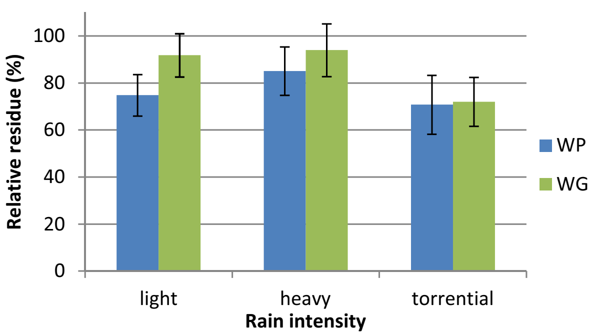


Figure 6.2: Relative mancozeb residue concentrations (±SD) of a wettable powder and water dispersible granules, after exposure to a light (180 L/h m²), heavy (360 L/h m²) or torrential (720 L/h m²) rain (4 mm), expressed as a percentage of the initial mancozeb deposit on the leaves (n = 3).

Measurement of the initial triadimenol deposit on clipped branches and apples revealed a comparable concentration ($p_{leaf}= 0.25$ and $p_{apple}= 0.89$) for both formulation types. The general effect of rain on this initial concentration was statistically higher on leaves, when compared to apples ($p = 0.01$) and revealed a respective decrease of 33 % and 23 % of the initial deposit after rainfall. The relative residue concentration after rain simulation (Figure 6.3) was very similar for all three rain intensities and both formulation types. This was confirmed by statistical analysis, which indicated no significant differences in rain fastness.

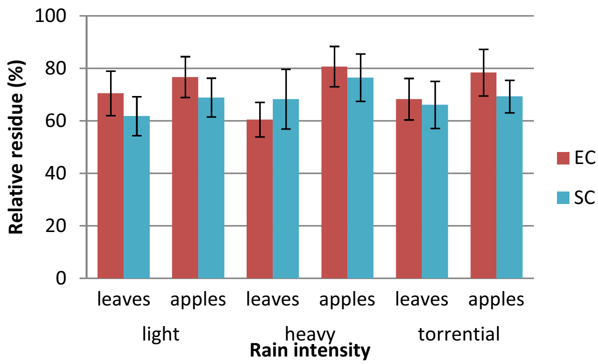


Figure 6.3: Relative residue concentrations (±SD) of triadimenol emulsifiable and suspension concentrates, after exposure to a light (180 L/h m²), heavy (360 L/h m²) or torrential (720 L/h m²) rain, expressed as a percentage of the initial triadimenol deposit on the leaves ($n = 3$).

Finally, a comparison of the initial deposits of mancozeb and triadimenol revealed that the triadimenol deposit on leaves was at least three times the mancozeb deposit, when the concentration difference in the spraying liquid was accounted for ($p < 0.001$).

6.3.3 Experiment 3 and 4: Apple lab experiments

6.3.3.1 Effect of waiting period

The effect of a waiting period of 2, 4 or 24 hours was assessed (Figure 6.4) and revealed a significantly different effect on rain fastness for the mancozeb WP and WG formulations ($p < 0.001$). Rain fastness of those solid formulation types almost doubled after 24 hours, when compared to a drying time of 2 hours. Average reductions of 64, 50 and 28 % of the initial mancozeb and captan deposits were found for respective drying times of 2, 4 and 24 hours. Conversely, the rain fastness of the liquid formulation types (EC, SC) did not exhibit a significant influence of drying time ($p = 0.64$). In this case, an average initial deposit decrease of 18 % was noted.

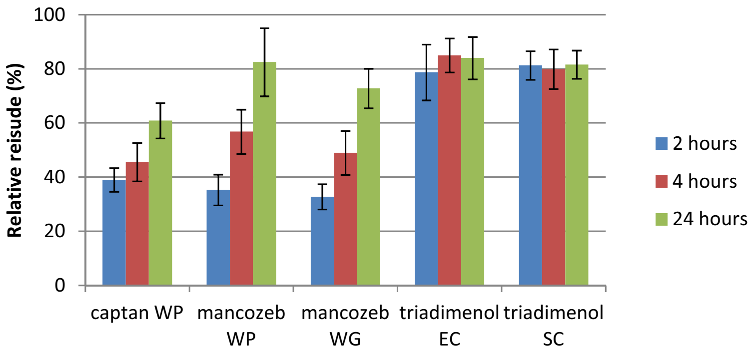


Figure 6.4: Relative residue concentrations (±SD) of captan, mancozeb and triadimenol after simulation of a rainfall intensity of 12L/h, with drying times of 2, 4 or 24 hours, expressed as a percentage of the initial residue concentration (n = 3).

6.3.3.2 Effect of rain intensity

Mancozeb residues after exposure to two realistic rain intensities and a drying time of four hours, showed a significant difference of 17 % between both rain intensities ($p = 0.01$) and for both formulations tested (Figure 6.5), but there were no statistically relevant effects of formulation type ($p = 0.73$).

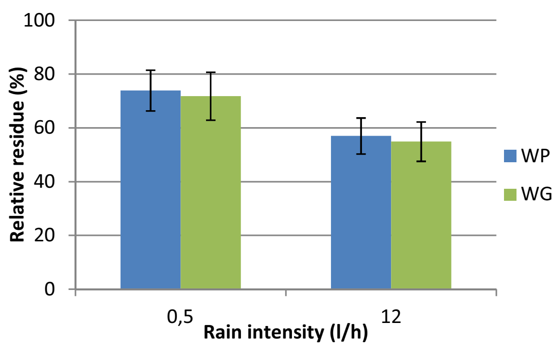


Figure 6.5: Relative residue concentrations (±SD) of a mancozeb wettable powder and water dispersible granules, after simulation of a rainfall intensity of 0.5 L/h and 12 L/h, expressed as a percentage of the initial residue concentration (n = 3).

6.4 Discussion

The first two experiments conducted in this study, did not reveal any clear trend in rain intensity effect, except for a larger effect of the highest rain intensity on the fungicide deposit. This deviation of the expected effect of rain intensity, can be a consequence of the experimental set up in this study. Firstly, in experiment 1, shielding might have had an important effect on the differences, noted between the two sampling zones. In practice, sprayers with air-support, allowing for a better penetration into the foliage, are commonly used. Therefore, the difference in initial residue deposit on apples from the inside versus the outside of the tree canopies, could be an overestimation. In contrast to the apples, this difference was not significant for the leaves, probably because one leaf sample consisted of three branches, versus only 1 apple. Thus, for leaves, this limitation did not considerably influence the results.

Secondly, rainfall simulation prevented the underside of the leaves from extended exposure to the simulated rain, while this side also received a part of the PPP spray. In addition, Bondada *et al.* (2007) illustrated a 40 % higher retention of captan on the abaxial, compared to the upper side of the leaves. Therefore, it is likely that the rain fastness was overestimated in this experiment. Nevertheless, the difference between both formulations was statistically significant. This difference can be explained by the absorption of both formulations into the plant tissues: emulsions have a high affinity for the waxy layer of leaves and are absorbed faster than non-oily formulations such as an SC. The diverse cuticula characteristics of fruits and leaves might explain the differences in rain fastness, similar to Cabras *et al*. (2001). Since this consideration is valid for all leaf samples, it does not influence the results of the applied rain intensities.

Third, the rain intensities that were technically possible, were very high when compared to a real situation. In addition, the very short rainfall duration might have inhibited residues from dissolving in the simulated rain. This can explain why a clear effect of rain intensity was not observed. Nevertheless, most samples exhibited the highest residue reduction after exposure to the highest rain intensity. Experiment 4 was conducted to assess whether these very high rain intensities might have masked the effect of rain intensity in experiments 1 and 2. For both the WP and WG formulation of mancozeb, a higher rain intensity resulted in a lower rain fastness. A difference between both formulations, on the contrary, was not observed. This indicates that an effect of rain intensity cannot be ignored, even though not all results in this study unambiguously confirmed such an effect.

Fourth, the samples were allowed to dry for 24 hours in experiments 1 and 2, which may have masked the effect of the different rain intensities, as proven by Hunsche *et al.* (2007).

This was confirmed in experiment 3 that depicted a doubled rain fastness of the WP and WG formulations after 24 hours in comparison with two hours. Contrarily, the liquid EC and SC formulations (systemic), exhibited little effect of drying time. Cabras *et al.* (2001) already proved that systemic PPPs are more rain resistant due to plant uptake, which explains why the triadimenol residues were less affected by the drying period than captan and mancozeb residues. However, since the experiment was conducted with harvested apples, it can be questioned if systemic transport through the apples actually occurred during this experiment. Nevertheless, these products are designed to penetrate the plant's tissues, which probably happened here.

Finally, result variability was substantially different in all experiments. This can be explained by the different sampling method for apples and leaves in the first experiment. In the second experiment, the adapted experimental set-up in which the branches were treated separately allowed for a more uniform initial deposit, decreasing result variability.

6.5 CONCLUSIONS AND FURTHER RESEARCH

This study showed that: (1) drying time is an important parameter in rain fastness; (2) EC formulations are preferred to SC, WP or WG formulations when rainfall is expected within a short time after application; and (3) an effect of increasing rain intensities due to climate change is expected, but could not be quantified in this study.

Further research might focus on the causes of differences in the rain fastness of fruits and leaves. This can have major implications for not only product choice and efficiency (high residue levels preferred), but also food safety (low residues levels preferred). Finally, it would be interesting to examine whether rain fastness could be quantified based on effective disease prevention or reduction through bio-assays instead of residue levels, as those allow a better reflection on the needed application adaptations under future climate conditions.

CHAPTER 7 ESTIMATING THE EFFECT OF CLIMATE CHANGE ON THE CODLING MOTH (*CYDIA POMONELLA*) AND PLANT PROTECTION PRODUCT USE, WITH A FORECASTING MODEL

Chapter 5 mentioned an expected but widely varying effect of climate change on pest occurrence and generation times. This chapter presents theoretical work on forecasting models with special attention to the effects of climate change on a specific pome fruit orchard pest, the codling moth (*Cydia pomonella*).

7.1 INTRODUCTION

Conventional agriculture often uses 'calendar sprayings' to fight pests and diseases. In that case, PPP treatments are scheduled periodically, depending on weather conditions. Until recently, this provided a sufficient level of pest management. However, this approach does not fit into the current European Integrated Pest Management strategy, implied by Directive 2009/128/EC, which aims to establish a framework for Community action to achieve sustainable use of PPPs from 2014 on.

Within an integrated pest management strategy, information on the life cycle of organisms and their interactions with the environment are generally considered to combine all appropriate pest management options, with the least impact on environmental and human health. Treatments with chemicals are only justified when the population density of the pest exceeds an action threshold and no other management options are available, which is both economically and ecologically beneficial (European Crop Protection 2010).

In Flemish pome fruit production, integrated pest management was already dominant in practice prior to its legal description in 1996 and forecasting models are commonly used tools (Vlaamse Overheid 2006). Forecasting models are decision support systems that assist in crop protection management. They are used to predict pest occurrence (and density) under certain circumstances, providing insights in the need for PPP applications and their optimal timing (Shaffer and Gold 1985). In practice, two different types of forecasting systems are applied: (1) systems in which the development of pests is observed in the field, to provide recommendations on the right timing of crop protection applications; and (2) systems in which pest pressure is modelled, based on weather data and that calculate action thresholds at which crop protection treatments should start (PCF 2013).

In Flemish apple cultivation, warning messages were issued for mainly *Cydia pomonella*, *Venturia inaequalis* and *Podosphaera leucotricha* (PCF 2013). Currently, the Podem, Mills A3, Maryblyt, Cougarblight and Billings models are being used in Belgium, together with degree-day models for the three above mentioned pests. Also extensively used by Belgian growers, is the Dutch program: 'RIMpro', which collects simulation models for important pests and diseases in fruit production. These models are generally more complicated than the simple degree-day models and require detailed local weather and input data (Trapman *et al.* 2008).

Forecasting models are available for sectors with high-quality produce that demand qualitative products with little PPP residues after harvest (Zadoks 1990). Some of those models are universally known and intensively used worldwide (Jones *et al.* 2013; Roubal and Rouzet 2003; Trapman *et al.* 2008). However, all models need validation in the local climate and for possible local pest varieties. The importance of forecasting models has increased and they are currently the third most important strategy for PPP reduction (Madden *et al.* 2000; Roubal and Rouzet 2003; Shaffer and Gold 1985; Trapman *et al.* 2008).

Many studies focus on the impact of climate change on pests and diseases, often linked with crop yields (Bergant *et al.* 2005; Kocmankova *et al.* 2010; Roos *et al.* 2011). However, little research on the impact of climate change on crop protection, has been conducted (Hirschi *et al.* 2010). The purpose of this study, was to evaluate the impact of climate change on pest pressure and PPP applications against *C. pomonella* in Flemisch apple orchards, based on forecasting and climate models.

This study considers *C. pomonella* because of the worldwide importance of this pest (Alston *et al.* 2010; Helsen *et al.* 2009; Jones *et al.* 2013; Roubal and Rouzet 2003; Shaffer and Gold 1985; Trapman *et al.* 2008) and the relative simplicity of the degree-day forecasting model. This limitation was necessary since projected future climate data do not yet consist of sufficiently detailed data, which are needed in the more complex forecasting models.

7.2 MATERIALS AND METHODS

7.2.1 CYDIA POMONELLA

C. pomonella is a major pest in apple and pear production and also attacks walnuts, apricots, plums, quince and sometimes peaches (Alston *et al.* 2010; Boivin *et al.* 2001; INRA 2013; Jones *et al.* 2013).

Fruit penetration of *C. pomonella* larvae shows by entrance and exit holes filled with excrements, called 'frass' (Figure 7.1a). Tunnelling into the fruit centre (Figure 7.1b) and feeding on seeds by the larvae may cause an altered hormone production, resulting in prematurely dropping fruits in which larval development can be completed (Alston *et al.* 2010). Obviously, bite marked and smeared fruits are impossible to sell (Alston *et al.* 2010; INRA 2013). Codling moth damage increased over the last two decades (Helsen *et al.* 2009) and was already mentioned to damage 85 % of the fruits, in a study by Moffitt and Westigard (1984).

(a) (b)

Figure 7.1: (a) Apple with frass; (b) tunneling by a larvae. Source a: Alston *et al.* (2010).

7.2.1.1 LIFE CYCLE

C. pomonella larvae (Figure 7.2) overwinter in a sheltered cocoon until diapause termination under influence of temperature and day-length. Then they pupate and a first generation of adults emerges, currently in the month of May for Flanders and the Netherlands (Helsen *et al.* 2009). Adults show a medium longevity of 15 to 18 days and peak activity occurs a few hours before and after twilight, when temperatures are at least 13°C. Mating and egg deposition occur during sundown and an average of 30 to 55 eggs are laid singly on the upper surface of leaves, near fruits (INRA 2013). Both processes are positively correlated with temperature, especially in the months of May and June (Helsen *et al.* 2009). The pre-oviposition period was longer for a first than for later generations, probably since suitable flight conditions occur later in spring than in summer. In addition, the flight is inhibited during rainfall, independent of temperature. Embryonic development occurs in 1 to 2 weeks, depending on temperature and hatching generally occurs between June 20th and July 20th. After hatching, the emerged caterpillar penetrates a fruit and tunnels to the core, where it feeds on the developing seeds and flesh. Larvae pass through five instars in 4 to 8 weeks, until they are fully grown and leave the fruit for a sheltered spot, to weave a cocoon to overwinter or pupate to a next generation of adult moths. This process is day length dependent, so for half of the 5th instar larvae that did not pupate by August 1st, diapause is induced (Helsen *et al.* 2009). However, higher temperatures (> 17°C) might counteract the effect of declining day-lengths, allowing a further development of a part of the larvae (Trapman 2013).

(a) (b) (c) (d)

Figure 7.2: Illustration of the different life stages of C. pomonella: (a) eggs, (b) first instar larva, (c) pupa, (d) adult female. Source a - c: INRA (2013); d: Alston *et al.* (2010).

7.2.1.2 WEATHER DEPENDENCY: THE DEGREE-DAY MODEL

Codling moth damage can be prevented by PPP applications targeting the eggs, larvae or adults, but none of the PPPs suffices on its own. A cumulative strategy, in which different PPPs are combined, can manage the population growth in consecutive years. Authorised active ingredients and their targeted stages in Belgian apple orchards are shown in Table 7.1. Timing is crucial for maximal PPP effectiveness because of the narrow exposure time frame of sensitive stages (Helsen *et al.* 2009) and can be modelled by combining adult occurrence in pheromone trap catches with phenology models (Jones *et al.* 2013; Roubal and Rouzet 2003; Shaffer and Gold 1985; Trapman *et al.* 2008). These traps are mainly a useful tool for insect detection and monitoring, since mass trapping is not effective enough to control the pest, in case of high pest pressures (Charmillot *et al.* 2000). Mating disruption in combination with insecticide sprayings is currently the most common practice (Fytoweb 2013; Jones *et al.* 2013). Model application is possible in small scale areas, when calibrated with local meteorological data (Roubal and Rouzet 2003).

Table 7.1: PPPs authorised in Belgium for use against C. pomonella in apple orchards. Source: www.fytoweb.org

Active ingredient	Targeted stage
fenoxycarb	eggs
diflubenzuron	eggs
chlorantraniliprole	eggs, larvae
emamectin benzoate	larvae
thiacloprid	larvae
granulose virus	larvae
Bacillus thuringiensis	larvae
methoxyfenozide	larvae
tebufenozide	larvae
indoxacarb	larvae
deltamethrin	adults
codlemone, tetradecene-1-ylacetate, dodecanol	adults

The biology of the codling moth can be divided into five life stages and temperature dependent developmental processes (Trapman *et al.* 2008). The lower developmental threshold is considered to be 10°C, while the maximum developmental speed is achieved at 28°C, with an upper threshold at 31°C. The duration of every life stage, is location and temperature dependent and can be expressed as the product of time (days) and the temperature above the lower threshold. The accumulated heat over time, or Degree-Days (DD) models, can be linear or more sophisticated, as described in other studies (Helsen *et al.* 2009; Jones *et al.* 2013; Roubal and Rouzet 2003; Trapman *et al.* 2008). The developmental stages and an indication of the developmental time in DD for *C. pomonella* are shown in Table 7.2.

Table 7.2: Degree-days needed for the individual developmental stages of C. pomonella, according to Alston *et al.* (2010).

Developmental stage/process		Accumulated heat (°C) Needed	Total
larvae	diapause termination	100 DD*	100 DD
	pupation	160 DD	260 DD
adults	pre-oviposition	50 DD	310 DD
eggs	embryonic development	100 DD	410 DD
larvae	larval development (L1-L5)	300 DD	710 DD
	pupation	160 DD	870 DD
adults	adults: 2^{nd} flight		
generation time		610 DD	

*Diapause termination after 98.8 DD, according to Trapman *et al.* (2008) or 96.7 DD, according to calculations with the method of Jones *et al.* (2013).

The model simulation for *C. pomonella* always runs from January 1st (Helsen *et al.* 2009; Shaffer and Gold 1985). After an empirically derived number of DD, adult emergence occurs. According to Alston *et al.* (2010), this occurs at 100 DD. However, a Dutch publication (Trapman *et al.* 2008) reports adult emergence on April 13th. When Ukkel (Belgium) temperature data for the period 2000 till 2008 were considered, this corresponded with 98.8 DD. Calculations with the method, described by Jones *et al.* (2013), result in an estimated adult occurrence after 96.7 DD. Nevertheless, it was chosen to work with 100 DD (Alston *et al.* 2010) here, since this value was also used in past model simulations in Flanders and this was the only source providing DD for every developmental stage.

Peak catches of *C. pomonella* are then used to determine when mating occurs. From that moment on, the model calculates when eggs and larvae are expected (based on Table 7.2) and crop protection treatments should be conducted. Individual growers can adapt the calculated accumulated heat over time to different climate types and local environmental conditions (Jones *et al.* 2013; Trapman *et al.* 2008). In this study, one of the a linear degree-day models was used, considering only a lower threshold of 10°C. Since PPP impact models generally require hourly data and most of the weather data are available on a daily timescale, DD calculation was based on daily minimum and maximum temperatures as described in the adapted single sine wave method by Watanabe (1978) (Case A and B). Hence, the other requirements mentioned in the life cycle paragraph, such as a sufficently high evening temperature, precipitation and day lenght, were not considered.

7.2.2 Weather data and climate projections

In this study, weather data registrations of the Royal Belgian Meteorological Institute from 1981 till 2008, were used. The meteorological institute provided both the minimum and maximum daily temperatures at their weather station in Ukkel, for the whole period. These data were used to compare the modelling results for *C. pomonella* with the actual occurrence of this pest in the past and to calculate climate projections for the future.

Climate projections were based on the latest assessment report of the International Panel on Climate Change and provide plausible descriptions of what may happen in the future. In this report, four representative concentration pathways (RCPs), covering the wide range of (atmospheric geenhouse gas concentration) emission scenarios that were published in literature, were identified. These RCPs are defined in radiative forcing, which is the difference between the radiant energy that enters and the energy leaving the atmosphere. They lead to respective radiative forcing (RF) levels of 8.5, 6, 4.5 and 2.6 W/m² by the end of the century (Moss *et al.* 2010; van Vuuren *et al.* 2011). These RCPs were used as input for the General Circulation Models, which then provide projections for the future climate. However, because of their large spatial and temporal resolution, the data were processed by Wageningen University, before they were useful in this study.

A detailed explanation of the modelling and downscaling is described by Liu *et al.* (In Preparation). The data from one earth-system model, the Community Climate System Model version 4 (CCSM4 - described by Gent *et al.* (2011)) was used here, because this model provides sound temperature and precipitation data, which are important variables for food contamination risk modelling (Liu *et al.* 2013). Since the output of general circulation models is gridded, downscaling of the spatial resolution was done by producing point data with local weather station data (for Ukkel, Belgium) as a reference for variability. The data provided by Wageningen University currently includes daily values from 2006 to 2100, divided in scenario periods, defined as 1981-2000 and 2031-2050 for the near past and future and 2080-2100 for the far future, to account for inter annual variability (Liu *et al.* In Preparation).

7.3 Results and discussion

7.3.1 Average yearly temperature

The average yearly temperatures projected for the near and far future (reference years 1981-2000), is illustrated in Figure 7.3. The temperature shows a similar trend for all simulations, since the projections were all calculated from the original data of 1981-2000. Projections for the near future indicated higher average temperatures for a RF level of 4.5 W/m² than 8.5 W/m² and 2.6 W/m² than 6.0 W/m². This deviation of the expected order, is due to the assumptions in greenhouse gases in these scenario's, which strongly influence the temperatures (Liu *et al.* In Preparation). In detail, the RCPs with radiative forcing of 6.0 W/m² (RCP26) and 8.5 W/m² (RCP85) first show a lower increase in greenhouse gases than the one with a radiative forcing of 4.5 W/m² (RCP45). For the far future, the trends followed the expectations. Overall, an increase in average temperature in the future, calculated with the CCSM-4 model, was projected, when compared to past temperature recordings. Since the

RCP26 and RCP85 respectively represented the lowest and highest temperature change, these two scenarios will be discussed.

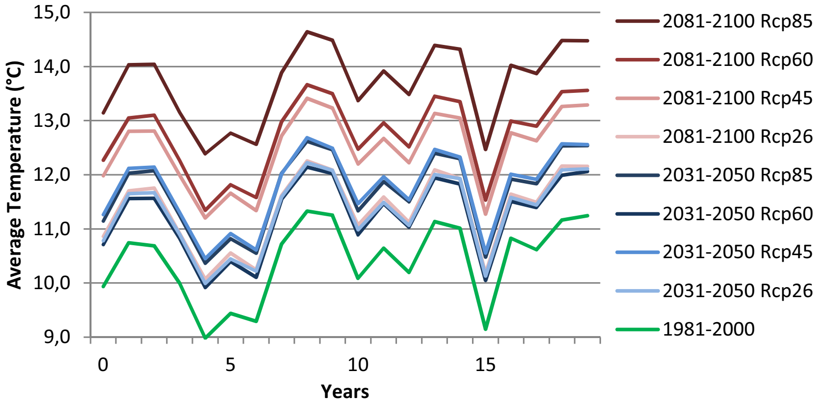


Figure 7.3: Average yearly temperature for time periods of 20 years (near past from 1981-2000, near future from 2031-2050 and far future from 2081-2100) and different radiative forcing levels.

Table 7.3: Theoretical timing of the different developmental stages, calculated with DD for the three twenty-year periods: 1981-2000, 2031-2050 and 2081-2100 and according to four different representative forcing pathways (RCPs).

Time frame / Developmental stage	1981-2000 (day-month)	2031-2050 RCP26	2031-2050 RCP45	2031-2050 RCP60	2031-2050 RCP85	2081-2100 RCP26	2081-2100 RCP45	2081-2100 RCP60	2081-2100 RCP85
diapause termination	08-05	25-05	23-05	24-05	21-05	15-05	08-05	30-04	27-04
egg deposition	21-05	06-06	03-06	04-06	02-06	25-05	19-05	14-05	09-05
egg hatching	09-06	23-06	20-06	23-06	19-06	12-06	02-06	30-05	25-05
L5 leaving fruits	21-07	25-07	22-07	26-07	21-07	18-07	07-07	04-07	26-06
2nd flight	07-08	06-08	03-08	08-08	03-08	01-08	21-07	19-07	10-07
egg deposition	13-08	10-08	07-08	12-08	06-08	05-08	26-07	23-07	13-07
egg hatching	28-08	18-08	14-08	20-08	13-08	14-08	03-08	31-07	21-07
L5 leaving fruits	11-10	16-09	08-09	19-09	07-09	17-09	28-08	23-08	10-08
3rd flight		04-10	23-09	11-10	22-09	30-09	13-09	08-09	21-08
egg deposition		10-10	28-09	22-10	27-09	05-10	19-09	13-09	25-08
egg hatching		17-10	12-10	23-10	10-10	30-10	02-10	26-09	02-09
L5 leaving fruits			08-11		31-10		09-11	06-11	03-10
4th flight									16-10
egg deposition									19-10
egg hatching									08-11
L5 leaving fruits									

7.3.2 TIMING OF DEVELOPMENTAL STAGES

An indication of the average timing for all developmental stages for the near past, near future and far future time periods, is presented in

Table 7.3. These dates roughly indicated occurrence of the first individuals in this stage, since calculations were conducted for pupated larvae (from which the first adults emerged, once diapause termination was reached). Fifth instar larvae from a previous year only started pupating at that time, so adult emergence is, in fact, spread over a longer period. These adults, on average, emerge 260 DD after January 1st, but were not considered here.

7.3.2.1 NEAR PAST: 1981-2000

Calculations of the temperature sum, based on the DD of Alston *et al.* (2010), indicated that the first flight of adults occurred between April 26th (1990, 1991, 1993) and May 26th (1984), resulting in a 20-year average of May 8th. Egg deposition was calculated to occur on average on May 21st, which is early, when compared to the actual deposition that occurred in the Netherlands on June 2nd, June 1st, June 14th and May 23rd, between 2004 and 2007 (Helsen *et al.* 2009). When considering the acutal temperatures in Uccle for these years, egg deposition was calculated to occur on May 21st, May 17th, May 17th and April 24th.

Since the study of Alston *et al.* (2010) did not specify any restrictions for timing of pupation and simulations were conducted for Belgium, this was assumed to be similar as in the Netherlands: diapause induction if L5 exit the fruits after August 1st. Calculations showed a second generation of codling moths in every year within this period, except for 1984 and 1987, where the fifth instar larvae were not fully developed before August 1st. This contrasts with literature considering the North-West of Europe that observed only one complete generation in this region (Jones *et al.* 2013; Trapman *et al.* 2008). On average, the calculated second generation of adults emerged around August 7th and diapause of the subsequent larvae was induced on October 11th. However, this only occurred in the years 1982, 1983, 1989, 1990, 1994, 1995, 1997 and 1999, if assumed that the declining day length did not inhibit completion of any previous stage. A fully developed L5 are expected to increase pest pressure the next year due to presence of cocoons (1st generation) and diapausing L5 of both the first and second generation of moths. However, literature indicated that the second generation has, until now, never been able to reach maturity (Helsen *et al.* 2009).

The differences between the calculated dates for egg deposition and number of completed generations, indicate the limitations of the simple linear DD model that was used. Such early (calculated) flights could theoretically occur, if they were not hindered by precipitation (on average 33 days in this period) and if temperatures only slowly dropped at evenings. According to Helsen *et al.* (2009), the differences can also indicate the importance of the photoperiod for this organism.

7.3.2.2 NEAR FUTURE: 2031-2050

In the near future projections, the first flight of adult codling moths is expected between May 8th and June 13th for the RCP26 scenario and May 6th and June 10th for RCP85. Both simulations indicated a possibility to complete the fifth larval stage in most years of the considered time frame. This occurred between July 18th and August 6th in the RCP26 scenario and four days earlier in the RCP85 scenario. The larvae that completed their development before August 1st induce a second generation of adults, on average on August 6th (RCP26) and August 3rd (RCP85). The development of L5 in this generation was on average completed on September 16th and September 7th, respectively in the RCP26 and RCP85 scenarios. When considering the percentage of larvae that did not enter diapause here, a third generation could emerge on October 4th (RCP26) or September 22nd (RCP85). According to this linear DD model, these individuals could theoretically reach enough DD for hatching in RCP26. Nevertheless, they would not contribute to next year's population. In the RCP85 scenario, larval development would theoretically be completed between October 29th and November 2nd. Overall, the DD model showed the possibility for an extra (completed) generation of codling moths to occur in Flanders, depending on the radiative forcing level.

7.3.2.3 FAR FUTURE: 2081-2100

Temperature projections for the far future indicated a large temperature increase when compared with the two other time frames, especially in the RCP85 scenario. The time frame in which the first adults were expected to emerge, lay in the month of May for RCP26 and between March 31st and May 11th for RCP85. Pupation of the first larvae was predicted to occur on July 18th and June 26th in respectively the RCP26 and RCP85 scenarios. Larval development in the second generation was predicted to be terminated between September 3rd and October 16th in RCP26 and even from August 3rd in the RCP85 scenario. The higher temperatures allowed for egg hatching and even a fully developed L5 and 4th generation of adults in Belgium, for the respective radiative forcing scenarios. Currently, there are already countries where multiple generations of codling moths are noted, such as large parts of the United States, England, Northern Italy and Switzerland (Jones *et al.* 2013; Trapman *et al.* 2008).

7.3.2.4 Evolution and considerations

With increasing temperatures, an earlier occurrence of the first adults was expected. This is consistent with earlier flowering as a consequence of mild winter temperatures, which was already noticed in the past (ESA 2014). Modelling of the simulated scenarios here, did not show this trend in the near future, nor the far future, except for the RCP60 and RCP85 scenarios. When the individual developmental stages are compared, the 2031-2050 simulations show an almost identical trend with the 1981-2000 period. However, once the second generation is reached, the developmental speed was a little higher in the near future than it was in the past. This is probably due to the gradual increase of greenhouse gases assumed in the projections and the influence of extremes, which will occur both in winter and summer. Here for, it is likely that higher peak temperatures in summer cause the gradually faster development from the second generation on. In addition, high evening temperatures, intensifing mating and oviposition, are more likely to occur during summer. Overall, little difference can be seen between these two time frames.

In the far future, an effective shift forward is predicted for all developmental stages. In this time period, a second flight is expected every year and even a theoretical, but limited, third generation is possible. When considering the RCP85 scenario, an effective third generation of moths showed, followed by a theoretical fourth generation, if the current day length restriction can be surpassed. This is likely, since high temperatures seem to counteract the diapause induction due to decreasing day lenghts (Helsen *et al.* 2009). Literature shows a structurally advancing flowering date since the 1980s, in concordance with increasing average temperatures in the months of February, March and April (ESA 2014). If this trend continues in the future, picking dates of the apples are also expected to shift forward. On average, Belgian apple picking begins on September 16th (average of 12 apple varieties). After the mild winters of, for example, 2007 and 2011, picking started respectively one and two weeks earlier (VBT 2007; VBT 2008; VCBT 2014). Hence, the fourth generation of codling moths will probably not completely emerge, since the timing of the larval development termination (September 16th - November 11th) implies that most apples will already be harvested. Hence, larvae lose their food source and some of them will be eliminated from the field with the harvested fruits. In the future, more generations of codling moths will generally be possible in Belgium, but there are still many unconsidered restrictions and influencing factors, which can strongly influence the actual number of completed generations.

Temperature sums were based on the DD described by Alston *et al.* (2010), even though Dutch publications cited other DD for some developmental stages, for example, 80-90 DD for egg development (Helsen *et al.* 2009). However, since there is no reason to assume that the codling moth reacts differently on temperature in any part of the world, the Alston DD were supposed to be valid for Belgium. Temperature sums assume a linear relationship between developmental time and temperature, which is not the case in nature. In addition, other inhibiting factors, such as rainfall events, were not considered. Here for, the temperature sums will overestimate the developmental rate. In addition, the measured air temperature can strongly differ from the actual temperature to which an organism is exposed (sunny trunk, inner of apple). This overestimation already showed, when comparing the calculated dates with data from literature for the near past. Nevertheless, Shaffer & Gold (1985) indicated that simulated times in their models were longer than observed, which can counteract the overestimation due to the linearity assumption. Since the purpose of this study was to reveal a possible evolution, comparing only calculated data decreases the importance of these limitations.

7.3.2.5 Insecticide applications

Currently, the codling moth is controlled with a combination of pheromone mating disruption and treatments with ovicides and larvicides.

Trapman *et al.* (2008), suggested that a link between the timing, at which larvae are fully developed in summer and enter diapause, with the timing of pupation in the following year, can be important in cases where multiple generations of codling moths occur. The projected evolution in the near and far future shows that more generations of codling moths could occur in Belgium. In this situation, pest pressure can indeed be higher, since more generations are able to develop cocoons with overwintering larvae that contribute to a next year's generation. An easy deduction would be to expect that insecticide use will have to increase. Nevertheless, two other considerations should be made. Firstly, temperatures at sundown and rainfall strongly influence adult moth activity (Trapman 2013), but were not modelled. However, these elements can directly influence the required insecticide applications. When weather conditions are less favorable for the codling moth, oviposition will occur over an extended period of time, hence, requiring additional ovicide applications. In contrast, when evening temperatures are high, a peak activity of adults and oviposition in a small time frame, are expected. In that case, Trapman *et al*. (2008) suggested that only one well timed ovicide application can suffice to eradicate a large part of the population. Secondly, in practice, a good management of the first generation is currently sufficient to strongly reduce codling moth occurrence (second generation) in apple orchards. This is also necessary, since management actions against the codling moth seem to be less effective when targeted against a second generation of codling moths. A possible cause is an increased insecticide tolerance or even resistance, induced by accelerated development rates. Therefore, a further reduction of the effects of later insecticide treatments might arise, which was already stated by Shaffer and Gold (1985) and Roubal and Rouzet (2003). Hence, it will remain important to sufficiently control the first generation of codling moths, preventing a population built-up further in the season.

7.4 CONCLUSIONS AND FUTURE RESEARCH

This study considered *C. pomonella* because of the worldwide importance of this pest and the relative simplicity of its forecasting model.

Weather generator outputs statistically behave like weather data, but any particular simulated sequence should not be considered as an observation at a given time. The selection of only a small set of best performing models, is not the best approach because there is no robust process for selecting a few "best" models. Certainly, the use of only one model as was done here, implies a large uncertainty of the results.

The presented results, showed future temperature conditions that enable multiple generations of codling moths in Belgium. However, field registrations remain important to cover the aspects that are not considered in the models. Detailed input data requiring phenology models, such as RIMpro, can then provide more accurate projections.

Even with additional generations of moths in the future, it is likely that a good management of the first generation will be sufficient to restrict codling moth occurrence in apple orchards. Hence, insecticide applications against this pest are assumed not to increase.

Currently, malformed or damaged apples are throw on the soil during picking, sustaining the L5 population. It would be interesting to study the feasibility and effect of removing these apples from the ochard, on the codling moth pest pressure in the next season.

CHAPTER 8 GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS FOR FUTURE RESEARCH

The fact that our climate has changed and that this will continue in the future, is practically indisputable. The effects of climate change occur all over the world, are various and important. Not only natural systems, such as coastal systems and low-lying areas are vulnerable, but also our energy supplies and transport can be strongly influenced by climate change (IPCC 2014). One specific effect considers food production systems and food safety, which was the focuss of the Veg-i-Trade project.

Food can contain different contaminants. Their origin can be either natural, such as microorganisms, mycotoxins or natural plant toxins; or introduced by humans, such as additives and agrochemicals (e.g. pesticides, growth regulators or veterinary drugs). Presence of natural toxins in food is difficult to control, but toxin introduction by humans can be controlled. This dissertation focusses on one group of agrochemicals: plant protection products.

Globalization, or the process of international integration, is another aspect with worldwide consequences. Increasing trade of, for example, fresh produce between countries, is made possible by a combination of lower transport costs, technological progress and policy liberalisation (EC 2012). With respect to plant protection products, the legislative basis, authorisations and monitoring in four countries (Belgium, Egypt, Brazil and South Africa), were reviewed and compared in Chapter 2. Trade can be impeded when authorised residue levels in the importing country are lower than in the exporting country. In addition, the observed differences can affect food safety, in a way that imported produce might contain high residue concentrations. A tool to detect where these food safety risks are most likely to occur, is required. An example of such a tool, was described in Chapter 3.

Another result of globalization is the introduction of more exotic products in our daily meals and foreign cooking methods. An example is the use of a wok pan, which originated in Asia. Since processing has an effect on the plant protection product residues in food, the effects of different processing methods should be assessed. The effects of cooking, steaming, grilling, microwaving and wokking were described in Chapter 4. This is important to provide more accurate risk assessments for human exposure to plant protection products in food.

The aim of this study was to enhance the knowledge on the effects of climate change on food safety, specifically caused by plant protection product use. Research was started by investigating the important aspects that affect the use of plant protection products and are directly or indirectly influenced by climate change (Chapter 5). It was carried on into a more practical oriented experiment, considering rain intensity (chapter 6); and a theoretical approach of changed pest pressure (chapter 7).

8.1 GENERAL DISCUSSION

When using climate change models, it is customary to define time and geographical frames, as was exemplified in Chapter 7. In this work, it was opted to describe the effect of specific changes in relation to the current situation, because this approach involves less uncertainties. This had some implications. Many of the effects, described in Chapter 5 are likely to occur, but the likelihood of these effects will differ at least regionally. This was exemplified by the adaptation scenario's, in which the likelihood of, for example, the introduction of new pests, was evaluated 'very likely' by experts of the global south, while 'neutral', by experts in the North (Kirezieva *et* al. 2014). In addition, several statements in Chapter 5, referred to only one study and might, hence, be anecdotal.

The use of simple models as in Chapter 3, 4 and 7 is acceptable for giving an indication on the output. Advantages are that these models do not require a lot of data and are generally quite easy to use. Disadvantages are the assumptions and simplification of, for example, pest development requirements (Chapter 7). However, within the scope of this work, this was sufficient, as it aimed to increase the knowledge on climate change effects on pesticide use.

The conclusions on future pesticide use, must be placed in the right context. Technological progress, nor other crop protection methodology developments were considered in this study, even though it is very likely that current developments and improved new technologies might replace some of the current plant protection products. In addition, increased use of (adapted) biological crop protection methods, can also offer solutions in a changing climate.

The innovation of this work lies in (1) the comparison between used/authorised pesticides in different countries; (2) the evaluation of new processing methods in comparison with common practices; (3) the development of a risk based sampling plan; (4) a demonstration of the necessity of formulations and (4) the description of the climate effect on the use of plant protection products.

8.2 General conclusions and recommendations for future research

This study showed that pesticide use will change in two ways due to climate change: (1) residues on crops decline faster due to temperature and precipitation increases; and (2) the pest/disease pressure of some pests/diseases will increase in the future.

The impact of globalization is linked with an altered plant protection product risk due to (1) imports of fresh produce from countries with less strict plant protection product requirements; and (2) the introduction of new cooking methods that have a different impact on the residues in food.

This study provided a clear indication of the limitations of current knowledge and research in this area, but also a way to go for the future.

In first instance, gathering more (detailed) data on consumption of processed food, residues, and residue prevalences, is required to elaborate on the work in this dissertation. In addition, the described experiments should be repeated for other processes, matrices and active ingredients (Chapter 4); with other rain intensities and formulations (Chapter 6); and for other pests (Chapter 7). Elaborating on all these aspects should enhance the knowledge and reveal clear trends. Finally, each chapter on its own revealed additional links and influencing aspects that should be considered to gain insights in the future situation. A case study for a specific location and time frame that combines all these separate chapters would provide insight into what to expect for a specific location. This would preferably be conducted in different countries.

APPENDIX A: LINEAR PROGRAM CODE FOR COMPUTING PARAMETER WEIGHTS

$with(Optimization):$

Minimize the tolerance "q" (=the biggest error "a should be <= b, but it is not, with an error a-b").

$$NLPSolve\left(q, \left\{u + v + x + y = 1, a = 1 \cdot u + 3 \cdot v + 1 \cdot x + 3 \cdot y, b = 3 \cdot u + 3 \cdot v + 3 \cdot x + 1 \cdot y, c = 1 \cdot u + 2 \cdot v + 4 \cdot x + 4 \cdot y, d = 2 \cdot u + 2 \cdot v + 5 \cdot x + 4 \cdot y, e = 1 \cdot u + 3 \cdot v + 5 \cdot x + 1 \cdot y, f = 3 \cdot u + 2 \cdot v + 5 \cdot x + 4 \cdot y, g = 3 \cdot u + 3 \cdot v + 1 \cdot x + 3 \cdot y, h = 3 \cdot u + 5 \cdot v + 4 \cdot x + 1 \cdot y, i = 1 \cdot u + 4 \cdot v + 5 \cdot x + 1 \cdot y, j = 3 \cdot u + 2 \cdot v + 5 \cdot x + 2 \cdot y, k = 4 \cdot u + 3 \cdot v + 1 \cdot x + 4 \cdot y, l = 3 \cdot u + 4 \cdot v + 2 \cdot x + 2 \cdot y, m = 4 \cdot u + 4 \cdot v + 5 \cdot x + 4 \cdot y, n = 5 \cdot u + 4 \cdot v + 1 \cdot x + 1 \cdot y, o = 4 \cdot u + 5 \cdot v + 3 \cdot x + 2 \cdot y, a \le c + q, a \le b + q, a \le i + q, a \le e + q, a \le d + q, c \le g + q, c \le h + q, c \le j + q, c \le f + q, b \le g + q, b \le h + q, b \le j + q, b \le f + q, i \le g + q, i \le h + q, i \le j + q, i \le f + q, e \le g + q, e \le h + q, e \le j + q, e \le f + q, d \le g + q, d \le h + q, d \le j + q, d \le f + q, g \le l + q, g \le k + q, h \le l + q, h \le k + q, j \le l + q, j \le k + q, f \le l + q, f \le k + q, l \le m + q, l \le n + q, l \le o + q, k \le m + q, k \le n + q, k \le o + q, n \le o + q, n \le m + q, p \ge \left(\left(u - \frac{.296875}{0.76875}\right)^2 + \left(v - \frac{.15625}{0.76875}\right)^2 + \left(x - \frac{.134375}{0.76875}\right)^2 + \left(y - \frac{.18125}{0.76875}\right)^2\right)^{\frac{1}{2}}\right\}, assume = nonnegative\right)$$

$$[0.124999999999999667, [a = 1.71428571428571, b = 2.66071428571429, c = 1.85714285714286, d = 2.50000000000000, e = 1.58928571428571, f = 3.08928571428571, g = 2.89285714285714, h = 3.08928571428571, i = 1.77678571428571, j = 2.75000000000000, k = 3.65178571428572, l = 2.96428571428571, m = 4.05357142857143, n = 3.91964285714286, o = 3.79464285714286, p = 1.07190425099899, q = 0.125000000000000, u = 0.589285714285715, v = 0.187500000000000, x = 0.0535714285714285, y = 0.169642857142857]] \quad (1)$$

APPENDIX B: APPLIED SAMPLE EXTRACTION AND ANALYSIS METHODS FOR DELTAMETHRIN, LAMBDA-CYHALOTHRIN AND TEBUCONAZOLE

8.2.1.1 PROCESSED VEGETABLES

For sample extraction, 50 g of the mixed, processed samples (4 replicates) was diluted with 200 mL of hexane/acetone mixture (1:1). After a second mixing, the contents were poured on a Büchnerfilter and the filtrate was put in a separation funnel with 200 mL of water and 25 mL of saturated sodium chlorine solution. The funnel was firmly shaken and after layer separation, the bottom layer was removed. This procedure was repeated twice after which the remaining layer was dried over sodium sulphate and dissolved in hexane.

8.2.1.2 COOKING WATER

For residue extraction from the preparation liquids in the cooking and steaming processes, the liquids (4 replicates) were put in a separation funnel with 25 mL of saturated salt-solution and 100 mL of hexane. The funnel was firmly shaken and after layer separation, both layers were collected in a beaker. This process was repeated with the collected water layer. The hexane layers were filtered over a sodium sulphate filter and the filtrate was collected in vials for residue analysis.

8.2.1.3 RESIDUE ANALYSIS

Deltamethrin residue concentrations were analyzed with an Agilent 6890N GC-ECD system (Agilent Technologies), equipped with an HP-5MS column (30 m x 250 µm x 0.25 µm) at an initial temperature of 130°C and increasing up to 280°C, resulting in a retention time of 17.67 minutes. To analyse the λ-cyhalothrin and tebuconazole residue concentrations, an Agilent 689N/5973 inert GC-MS system was used (HP-5MS column) at an initial temperature of 70°C, increasing to 150°C (25°C/min), 200°C (3°C/min) and 280°C. Retention times were 28.15 minutes for λ-cyhalothrin and 24.99 minutes for tebuconazole.

Concentration series for deltamethrin, λ-cyhalothrin and tebuconazole were prepared from standard solutions and served for establishing the calibration curves. Correlation coefficients for all three curves were sufficiently high and recoveries (8 replicates) ranged from 55 % ± 4 % (broccoli) till 103 % ± 6 % (carrots) for deltamethrin, from 78 % ± 9 % (courgette) till 112 %

± 18 % (broccoli) for λ-cyhalothrin and from 54 % ± 9 % (courgette) till 70 % ± 20 % (beans) for tebuconazole.

APPENDIX C: APPLIED SAMPLE EXTRACTION AND ANALYSIS METHODS FOR CAPTAN, TRIADIMENOL AND MANCOZEB

8.2.1.4 CAPTAN AND TRIADIMENOL

Each sample for analytical determination comprised of the leaves of three branches and a total of three replicates per treatment were analyzed. Apples were individually considered as one sample and again three replicates per treatment were analyzed. Leaves were plucked from the clipped branches and grinded with a mixer. Subsamples of 10 g were transferred into a glass bottle with screw cap and 50 mL of ethyl acetate was added. Bottles were put on an orbital shaker at a speed of 200 rpm for 60 minutes. The bottle content was then filtered through a Buchner-filter, dried over 30 g of $NaSO_4$ and collected in a flask of which 1 mL was transferred into a vial. Each individual apple was also grinded with a mixer and a subsample of 20 g was mixed with 100 mL of ethyl acetate in a grass bottle with screw cap. Bottles were put on an orbital shaker at a speed of 200 rpm for 60 minutes. The bottle content was then dried over 30 g of $NaSO_4$ and collected in a flask of which 1 mL was transferred into a vial. The captan concentration in the samples was analyzed with an Agilent GC-ECD (6890N, Agilent Technologies) in split mode at a temperature of 280°C and pressure of 102.7 kPa equipped with an HP-5MS column (30 m x 250 um x 0.25 µm) and oven program ranging from 80 till 290°C. The triadimenol levels were analyzed with an Agilent GC-MS (6890N) with injection in splitless mode at 280°C and 1.365 bar equipped with an HP-5MS column (30 m x 0.25 mm x 0.25 µm) and initial oven program ranging from a temperature of 70°C till 280°C. Retention times of 12.9 minutes for captan and 18.3 and 18.7 minutes for the two triadimenol isomers were found.

Concentration series for captan and triadimenol were prepared from standard solutions and served for establishing the calibration curves. Correlation coefficients for both curves were sufficiently high and recoveries (8 replicates) ranged from 80.5 % ± 6.3 % for apples till 94.6 % ± 5.1 % for captan on leaves and 95.8 % ± 4.4 % for apples till 104.7 % ± 4.7 % triadimenol on leaves.

8.2.1.5 MANCOZEB

Mancozeb analysis is based on the degradation of dithiocarbamates into CS_2 in an acid environment. The formed CS_2 is collected in a solution of di-ethanolamine and copper acetate in ethanol, which results in a yellowish-colored complex that can be measured spectrophotometrically. For each treatment, the leaves of three branches were plucked and cut up and homogenized. A subsample of 5.0 g was taken and transferred into the distillation equipment. Each apple was considered an individual sample and as such cut up and homogenized to provide a subsample of 50 g. A solution of 31.3 g $SnCl_2$ and 0.5 L HCL was diluted with 2.5 L of water and 130 mL was added to the subsamples. The two absorption bottles were respectively filled with 30 mL of NaOH (6.5 %) and 30 mL of a 12 mg copper(II)acetate and 25 g di-ethanolamine solution in 69 % ethanol. The mixture was cooked for 30 minutes after which the content of the last absorption bottle was transferred into a volumetric flask of 50 mL and diluted with 96 % ethanol. Extinction was measured at 435 nm in a UV-VIS spectrophotometer. A concentration series for mancozeb was prepared from standard solutions and served for establishing the calibration curves. The correlation coefficient was 0.998 and the recovery (8 replicates) ranged from 70.9 % ± 3.9 % for apples till 89.0 % ± 3.0 % for mancozeb on leaves.

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CURRICULUM VITAE

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EDUCATION

2009 – 2014	Doctoral Schools of Bioscience Engineering, Ghent University, Ghent
2004 – 2009	MSc in Bioscience Engineering, option Agriculture, Faculty of Bioscience Engineering, Ghent University, Ghent
2002 – 2004	Secondary school, Science– Mathematics, Pleinschool, Kortrijk
1998 – 2002	Secondary school, Modern languages– Mathematics, Sint-Amandscollege, Harelbeke

PROFESSIONAL CAREER

2010 – 2014 Researcher at Ghent University, department of Crop Protection,
Laboratory of Crop Protection Chemistry

‘Impact of Climate Change and Globalization on Safety of Fresh Produce - Governing a Supply Chain of Uncompromised Food Sovereignty’.
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2009 – 2010 Researcher at Ghent University, department of Crop Protection, Laboratory of Crop Protection Chemistry

Tasks:
Updating the pesticidePPP Occupational Risk Indicator (POCER)
POCER and Seq calculations for the Flemisch Environment Agency (VMM)
Risk calculations for moss biocides (Test-Aankoop)
Risk calculations for parasiticides for pets (Test-Aankoop)
POCER calculations for ‘Dual indicator set for sustainable crop protection’ (ILVO).

PUBLICATIONS

8.2.1.6 INTERNATIONAL JOURNALS WITH PEER REVIEW

Delcour, I., Rademaker, M., Jacxsens, L., De Win, J., De Baets, B., & Spanoghe, P. (2015). A risk-based pesticide residue monitoring tool to prioritize the sampling of fresh produce. *Food Control*, *50*, 690–698.

Delcour, I., Spanoghe, P., Uyttendaele, M. (2014). Literature review: Impact of climate change on pesticide use. *Food Research International*, DOI: 10.1016/j.foodres.2014.09.030.

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Delcour, I., Van Boxstael, S., Jacxsens, L., Amuoh, C.N., Uyttendaele, M., Spanoghe, P. (In preparation). Health and food safety risks related to PPP use: a different context in EU and developing countries. *Food Security.*

8.2.1.7 CONFERENCE PROCEEDINGS (FIRST AUTHOR)

Delcour, I., De Win, J., Jacxsens, L. and Spanoghe, P. (2012). "Development and validation of a prioritisation methodology for sampling and analysis of PPP residues in fresh fruit and vegetables." *Presented at 64th International symposium on Crop Protection*.

Delcour, I., Houbraken, M., Gobin, A., Steurbaut, W. and Spanoghe, P. (2011). "The influence of climate change on the incidence of plagues and diseases." *Presented at 63rd International symposium on Crop Protection*.

Delcour, I., Vanhonacker, F., Rosseneu, F., Verbeke, W., Steurbaut, W., Spanoghe, P. and Van Klaveren, J. (2011). "Acropolis: cumulative and aggregate risk assessment of PPPs in Europe." *Presented at 63rd International symposium on Crop Protection*.

Delcour, I., Spanoghe, P., Jacxsens, L. and Steurbaut, W. (2010). "Impact on PPP application and PPP risk assessment by climate changes and globalization." *Presented at 62nd International symposium on Crop Protection*.

SCIENTIFIC ACTIVITIES

8.2.1.8 ORAL PRESENTATIONS AT CONFERENCES

2013 ‘Prioritisation methodology for sampling and analysis of PPP residues in fresh fruit and vegetables.’
65th International Symposium on Crop Protection, May 21st, Ghent, Belgium.

‘A Prioritisation method for PPP monitoring.’
Veg-i-Trade communication session to the industry, June 11th, Ghent, Belgium.

2012 ‘Monitoring of PPP residues in trade.’
Veg-i-Trade workshop for the industry, March 8th, Lokeren, Belgium.

‘Methodology for PPP residue monitoring in fresh fruit and vegetables.’
Innovation for Sustainable Production 2012 (i-SUP), May 9th, Bruges, Belgium.

8.2.1.9 POSTER PRESENTATIONS AT INTERNATIONAL CONFERENCES

2013 ‘The effect of using a grill, wok or microwave oven on PPP residues on vegetables.’
65th International Symposium on Crop Protection, May 21st, Ghent, Belgium.

2012 ‘Prioritisation methodology for sampling and analysis of PPP residues in fresh fruit and vegetables.’
64th International Symposium on Crop Protection, May 22nd, Ghent, Belgium.

2011 ‘Cumulative and Aggregate Risk Assessment of PPPs in Europe.’
63rd International Symposium on Crop Protection, May 24th, Ghent, Belgium.

‘The influence of climate change on the incidence of pests and diseases: prediction models.’
63rd International Symposium on Crop Protection, May 24th, Ghent, Belgium.