Observations on the distribution of monophasic Salmonella Typhimurium on pig farms in Great Britain

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Abstract
Ten pig herds were visited and intensively sampled to determine the within group prevalence, distribution of contamination and numbers of Salmonella organisms excreted by infected pigs. The distribution of infection was highly variable but on all farms with breeding pigs the breeding herd was involved, even though the occurrence of the organism was greater in growing and fattening pigs. Infection was less common in farrowing areas. Involvement of wild birds and contamination of soil on outdoor units was frequently found. Numbers of organisms excreted were typically low but levels of up to 106 cfu/g were found in a small number of samples. The role of breeding pigs was clearly illustrated by the finding of five different serovars, including monophasic S.Typhimurium, in a single batch of replacement gilts delivered to one outdoor breeding farm.

Introduction
Monophasic variants of Salmonella Typhimurium (mST) have emerged in pigs and other species in many countries during the last twenty years, but the emergence of phage type DT193 variants with resistance to ampicillin, streptomycin, sulphonamide and tetracycline in most European countries has been particularly dramatic, and has resulted in a substantial number of human cases (EFSA, 2010). The reason for the emergence and rapid spread of mST in UK pigs since 1996 is unknown but one hypothesis could be increased involvement of breeding pigs or higher numbers of organisms shed in faeces, leading to more rapid spread of infection. This study was therefore begun in order to investigate qualitatively and quantitatively the distribution, and subsequent persistence, of infection on a series of different types of pig farms.

Materials and Methods
Ten commercial pig herds (3 outdoor breeding, 3 indoor breeder finisher, 1 outdoor grower, 4 outdoor finisher, 2 indoor finishers) in which monophasic S.Typhimurium S.4(5)12:i:- or S.4,12:i:- had been isolated or suspected were visited by the authors. A combination of naturally pooled faeces (taken with a large gauze swab), individual faeces (60 per epidemiological group or if less than 60 pigs in the group a number of faeces equivalent to the number of pigs was taken) and wildlife and environmental samples were taken and returned to the laboratory on the day of collection. Culture of the swab samples was begun on the day of collection but individual samples were held at 4oC until the next day when 2 g was aliquoted and tested. The remainder of the individual samples was retained until a Salmonella result was obtained then a maximum of 40 samples from each farm as semiquantified by a dilution-enrichment technique as described by Wales et al (Wales et al., 2006). Salmonella culture was carried out using a modification of ISO6579 : Annex D, using Rambach agar as the single isolation medium. Salmonella isolates were serotyped and a selection were also phage typed.

Results
A mixture of Salmonella serovars was found in pig areas on all farms except E and F, where only monophasic S.Typhimurium (mST) was present. Only the qualitative results from breeding farms are shown in table one because of space limitations but other data will be presented at the conference. In larger farms both regular mST and 'Copenhagen' variants were sometimes found and phage types DT193 and DT120 could be present concurrently. Monophasic and regular S.Typhimurium was more likely to be found in weaned and fattening pigs rather than the breeding herd but in most cases a low to moderate prevalence was also found in breeding pigs, particularly gilts. Involvement of wild birds, particularly on outdoor units, and rodents was identified. Pooled water, transport vehicles and various environmental samples were

often positive. On Farm C, which held a newly established breeding herd set up on new ground, five different serovars of Salmonella (S.4,5,12:i:- DT193, ST DT untypable, S.Anatum, S.Infantis, S.Derby) were previously found in batches of gilts delivered from a breeding company. Some, but not all, of these serovars were later found amongst maiden gilts and boars sampled on the farm. Prevalences of mST excretion in batches of animals varied from less than 2% to 100% (in growing pigs) and estimated numbers of organisms per gram of faeces ranged from 1-106, with highest numbers more likely to be found in gilts and pigs in service areas. A wide range of serovars other than mST was found on most farms and these often dominated, especially in adult pigs.

Similar serovars were found in wild birds, rodents and environmental samples as were found in individual pig faeces samples. On outdoor units soil and wallows were particularly likely to be contaminated if there was significant infection in the pigs. Salmonella was also found in empty pens that had been cleaned and disinfected, illustrating the need for improved procedures.

Discussion

The observations from this series of visits confirm that mST is likely to behave in a similar way to regular ST (Davies and Wray, 1997), although the high level of involvement in human cases in the absence of a regular poultry host is unusual (VLA, 2010). Some of these farms have been visited on a second occasion, 3 - 4 months after the first visit, and mST has persisted on all of these except Farm J (data not shown) which was a batch farm in which the replacement batch of pigs carried serovars other than mST, although this was still found in the environment around the pig houses as well as in the range area of two free-range broiler **f**ocks on the same premises. Spread from pigs into other species is a major concern, especially for chicken breeding and laying **f**ocks which are likely to be slaughtered if such strains are found. To date, a small number of conventionally-housed chicken breeding **f**ocks, free-range laying **f**ocks and free-range broiler **f**ocks in GB have been found with mST and the organism has also been reported from numerous cattle herds (VLA, 2010). All of the poultry and cattle herds with mST that have been visited by the project team have had pigs on the same or adjacent holdings. Pig-related serovars of Salmonella have also sometimes been found in wild bird faeces on such farms. Unlike the situation in pigs, mST in cattle herds appears to reduce rapidly, especially in closed herds when ST vaccine has been used. There are however indications that procedures such as moving outdoor herds to new land, acidification of feed and use of liquid feed may be associated with a reduction in the prevalence of infection in positive herds. Further studies will aim to follow the course of infection over a longer time period and will attempt to link the characteristics of the farm infections with management practices and molecular genetic profiles of the strains.

Conclusions

Monophasic Salmonella Typhimurium in pig populations appears to be a significant potential new public health threat and a source of infection for other species. Information on effective means of control is urgently required.

References

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Table 1: Breeding Herds – pooled faeces / environmental samples – no.mST(no. other serovars)/no. samples taken [%mST](% other serovars.)

	A[O]	B[O]	C[O]	D[I]	E[I]	F[I]	Total
maiden gilts	2(4)[V]/8[25](50)	0(0)/4[0](0)	33(10)[D](4)[r](2)[V](2)[R](2)/46[71.7](21.7)	1(4)[D]/12[8.3](33.3)	0(0)/1[0](0)	2(0)/2[100](0)	38(14)/65[**58.5**](21.5)
in pig gilts	0(2)[V]/2[0](100)	1(1)[L]/14[7.1](7.1)	-	5(4)[D]/14[35.7](28.6)	0(0)/1[0](0)	4(0)/4[100](0)	10(5)/33[**30.3**](15.2)
service area	0(6)[V]/12[0](50)	0(1)[L]/5[0](20)	-	0(12)[D]/14[0](85.7)	2(0)/8[25](0)	-	2(13)/27[**7.4**](48.1)
dry sows	0(22)[V](20)[V](2)/22[0](100)	1(3)[R,V]/17[5.9](17.6)	-	3(30)[D]/44[6.8](68.2)	0(0)/4[0](0)	8(0)/10[80](0)	12(33)/75[**16**](44)
farrowing sows	5(13)[V](2)[V](11)/21[23.8](61.9)	0(2)[L]/64[0](3.1)	-	0(5)[D]/18[0](27.8)	0(0)/21[0](0)	5(0)/10[50](0)	5(7)/113[**4.4**](6.2)
weaners	-	3(0)/28[10.7](0)	-	0(7)[D]/37[0](18.9)	0(0)/8[0](0)	3(0)/3[100](0)	6(7)/76[**7.9**](9.2)
growers	-	-	-	19(0)/49[38.8](0)	18(1)[T]/43[41.9](2.3)	4(0)/4[100](0)	41(1)/96[**42.7**](1)
finishers	-	-	-	7(5)[D]/24[29.2](20.8)	-	9(0)/9[100](0)	16(5)/33[**48.5**](15.2)
wild birds	7(20)[V]/43[16.3](46.5)	0(0)/17[0](0)	17(0)/22[77.3](0)	1(3)[D]/9[11.1](33.3)	0(0)/4[0](0)	1(0)/2[50](0)	26(23)/97[**26.8**](23.7)
rodents	-	-	-	3(1)[D]/6[50](16.7)	1(0)/6[16.7](0)	1(0)/1[100](0)	5(1)/13[**38.5**](7.7)
transport	-	0(0)/3[0](0)	2(0)/4[50](0)	-	-	10(0)/10[100](0)	12(2)/19[**63.2**](10.5)
pooled water	-	0(0)/4[0](0)	-	3(5)[D]/8[37.5](62.5)	0(0)/8[0](0)	1(0)/1[100](0)	4(5)/21[**19**](23.8)
misc env.	-	1(1)[L]/41[2.4](2.4)	5(3)[D](2)[R]/15[33.3](20)	-	0(2)[r]/8[0]0(25)	4(0)/9[44.4](0)	10(6)/73[**13.7**](8.2)
						Overall total:	187(122)/699[**26.8**](17.5)

General key: mST = monophasic Salmonella Typhimurium; Salm. = Salmonella; O outdoor herd; I indoor herd; env. = environmental sample
Serovar key: T S.Typhimurium; d S.Derby; L S.London; V S.Virchow; r S.Reading; m S.Bovismorbificans

Table 2: Breeding Herds – individual faeces samples – no.mST(no. other serovars)/no. samples taken [%mST](% other serovars)

	A[◊*]	B[◊]	C[◊]	D[‡]	E[‡]	F[‡]	Total/Mean
maiden gilts **prev.**	4(0)/19[21.1]	4(1)[N]/60[6.7](1.7)	167(0)/364[45.9](0)	ND	ND	0(0)/4[0](0)	171(1)/424[**40.3**](0.2)
mean **cfu score** (range)	1.75(1-2)	1.2(1-4)	1.42(1-4)	ND	ND	0	1.09(1-4)
in pig gilts **prev.**	22/22(100)	3(0)/60[5](0)	-	ND	ND	3(0)/11[27](0)	6(0)/71[**8.5**](0)
mean **cfu score** (range)	18(3-6)	1.3(1-2)	-	ND	ND	1.0(1)	6.77(1-6)
service area **prev.**	51/59[-](86.4)	1(1)[-]/60[1.7](1.7)	-	0(7)[D]/50[0](14)	0(0)/9[0](0)	-	1(8)/119[**0.8**](6.7)
mean **cfu score** (range)	3(1-6)	1.0(2)	-	1.4(1-2)	0	-	1.35(1-6)
dry sows **prev.**	59)/59(100)	0(1)[P]/60[0](1.7)	-	0(15)[D]/60[0](25)	ND	2(0)/38[5.3](0)	2(16)/158[**1.3**](10.1)
mean **cfu score** (range)	4.5(3-4)	1.0(1)	-	1.8(1-4)	ND	1.0(1)	2.08(1-4)
farrowing sows **prev.**	51/80(63.8)	0(0)/60[0]0(0)	-	0(1)[D]/60[0](1.7)	0(0)/19[0](0)	1(0)/6[16.7](0)	1(1)/145[**0.7**](0.7)
mean **cfu score** (range)	1.4(1-3)	0	-	NA	0	1.0(1)	0.6(1-3)
weaners **prev.**	-	4(0)/60[6.7](0)	-	0(5)[D]/60[0](8.3)	1(0)/50[2](0)	30(0)/30[100](0)	35(5)/200[**17.5**](2.5)
mean **cfu score** (range)	-	1.25(1-3)	-	1.8(1-4)	1.0(1)	2.3(1-4)	1.59(1-4)
growers **prev.**	-	-	-	14(0)/60(23.3)(0)	7(0)/101[7](0)	9(0)/9[100](0)	30(0)/170[**17.6**](0)
mean **cfu score** (range)	-	-	-	1.2(1-3)	2.0(1-5)	1.1(1-2)	1.43(1-5)
finishers **prev.**	-	-	-	5(0)/60[8.3](0)	see growers	41(0)/60[68.3](0)	46(0)/120[**38.3**](0)
mean **cfu score** (range)	-	-	-	1.0(1)	-	1.8(1-3)	1.4(1-3)
* not included in prevalence totals as serotyping not done on all isolates							292(31)/1407[**20.8**](2.2) MEAN 2.04(1-6)