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Investigation of risk factors for *Salmonella* on fattening-turkey farms

C. A. FEATHERSTONE[1]*, R. REICHEL[1], L. C. SNOW[2], R. H. DAVIES[3], K. H. CHRISTIANSEN[2], J. J. CARRIQUE-MAS[3] AND S. J. EVANS[3]

[1] *Veterinary Laboratories Agency – Thirsk, West House, Station Road, Thirsk, North Yorkshire, UK*
[2] *Veterinary Laboratories Agency – Centre for Epidemiology and Risk Analysis, New Haw, Addlestone, Surrey, UK*
[3] *Veterinary Laboratories Agency – Food & Environmental Safety, New Haw, Addlestone, UK*

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SUMMARY

A cross-sectional study into risk factors for *Salmonella* was undertaken using data gathered from 252 fattening turkey flocks in the UK. The data was derived from the EU baseline survey conducted during 2006 and 2007, in addition to a voluntary questionnaire. Multivariate logistic regression models identified significant risk factors for *Salmonella* spp. and *Salmonella* Typhimurium. A decreased risk of *Salmonella* spp. infection was associated with a history of intestinal illness in the sampled flock (OR 0·17), the use of wood shavings as litter (OR 0·21), use of disinfectant in the cleaning process (OR 0·25), incineration of dead birds on farm (OR 0·29), seasonal production (OR 0·31), farm staff also working with cattle (OR 0·31), and the presence of pigs on neighbouring farms (OR 0·38). The risk of isolating *Salmonella* spp. varied according to the company from which the poults were sourced. A reduced risk of *S.* Typhimurium infection was associated with the use of wax blocks to control rodents (OR 0·09), using mains water (OR 0·19) and having a *Salmonella* test programme (OR 0·23). An increased risk of *S.* Typhimurium infection was associated with storage of items around the turkey house (OR 5·20), evidence of mice (OR 4·71) and a soil surface surrounding the turkey house (OR 2·70). This study therefore identifies a number of important practical measures which can be implemented by farmers and veterinarians within the turkey industry to assist in the control of salmonellosis at the farm level.

Key words: Disinfection, risk factors, rodents, *Salmonella*, *Salmonella* Typhimurium, survey, turkey.

INTRODUCTION

Salmonella spp. are an important cause of human illness. In 2007, there were 13 213 laboratory-confirmed cases of *Salmonella* in humans in the UK, although the true number of cases is considered to be four times higher [1]. Consumption of poultry products is well documented as a major source of infection, with the public health risk arising from accidental undercooking of meat or cross-contamination of other foods [1–3]. The contribution of turkey meat is, however, unknown. Each year about 17 million turkeys are raised in the UK, 10 million of these for the Christmas market [4]. Currently, most major turkey

* Author for correspondence: Mrs C. A. Featherstone, VLA Thirsk, West House, Station Road, Thirsk, North Yorkshire, YO7 1PZ, UK.
(Email: c.featherstone@vla.defra.gsi.gov.uk)

companies undertake voluntary monitoring for *Salmonella* and most *Salmonella* isolations are not associated with clinical disease. In 2007, there were 112 reports of *Salmonella* in turkey flocks in Great Britain [1].

Salmonella in poultry traditionally arouses much political interest, with pressure to reduce contamination at the farm level. October 2006 marked the start of a European Union (EU)-wide baseline survey over one year, to determine the prevalence of *Salmonella* spp. in breeding and fattening turkey (*Meleagris gallopavo*) flocks [5]. Results of the survey have been used to set targets to reduce the prevalence of *Salmonella* in both breeding and fattening turkey flocks, as part of EU-wide National Control Plans that come into force from 2010.

The pathways of *Salmonella* dissemination on poultry units are dynamic and complex, and researchers have identified numerous potential sources and risk factors for *Salmonella* infection on poultry farms [6–10]. Only a few studies have looked at the risk factors in turkey flocks specifically, which differ from chicken broiler flocks in their seasonality of production and longer growing period. Arsenault and others [11] reported that turkey flocks were more likely to be infected if they originated from a particular hatchery, or were raised in houses where two or more persons had access. They did not detect association between *Salmonella* infection at flock level and variables related to pest control, downtime period, manure disposal, and poultry-house cleaning and disinfection practices. Analysis of data collected during the EU baseline survey in 2006–2007 revealed increased risk of *Salmonella* in fattening turkey flocks with a larger number of turkeys on the holding. The presence of breeding turkeys on the same unit and having a free-range flock also increased the risk. While the risk of *Salmonella* infection varied throughout the survey period, *Salmonella* vaccination and subdividing the birds on a holding into a relatively large number of flocks was found to decrease the risk [12].

A greater understanding of farm husbandry and management practices which are associated with *Salmonella* infection in turkeys would assist in the challenge of reducing *Salmonella* prevalence, as required by the EU, in a realistic, practical, and cost-effective way. The baseline survey provided the opportunity to gather information on turkey fattening farms to investigate potential risk factors for *Salmonella* infection, as recommended to member states by the European Food Safety Authority (EFSA). This paper describes the results of the UK investigation.

METHODS

Baseline survey

The survey in the UK was carried out between October 2006 and September 2007 as part of the EU survey to establish the baseline prevalence of *Salmonella* in commercial turkey flocks. A detailed description of the survey design, the sampling and bacteriological testing is given in the document of European Commission, Directorate General for Health and Consumer Affairs (DG SANCO) [13]. A total of 317 holdings were selected at random from a list of commercial fattening turkey holdings in the UK, stratified by region (England, Wales, Scotland, Northern Ireland) and number of turkeys on the holding (holding size). Holdings with <500 fattening birds were excluded from the sampling frame. From each holding one flock was sampled within 3 weeks of slaughter/depopulation. Five pooled environmental faecal samples were collected from each sampled flock, using boot swabs pre-moistened with sterile water and worn over disposable plastic over-boots. Samples were despatched to the laboratory on the day of collection, and examined on the day of arrival. Samples from England, Wales and Scotland were submitted to the Veterinary Laboratories Agency (VLA), Weybridge and samples from Northern Ireland to the Agri-Food and Biosciences Institute (AFBI), Belfast. The isolation method used a modified semi-solid Rappaport–Vassiliadis medium (MSRV) as the single selective enrichment medium, as described in Annex D of ISO 6579 [14].

Data collection

Basic farm-level information was required by the EU and gathered for all member states. This included details of the production type and size of holding, as well as data relating to the sampled house (number of birds, age, crops per year, expected age at slaughter, *Salmonella* vaccine and recent medicine use). In addition, flock owners in the UK were requested to complete an additional voluntary questionnaire, by interview with the animal health officer taking the samples. This questionnaire was designed to collect information on additional flock, house and farm-level factors that may be related to the risk of *Salmonella*

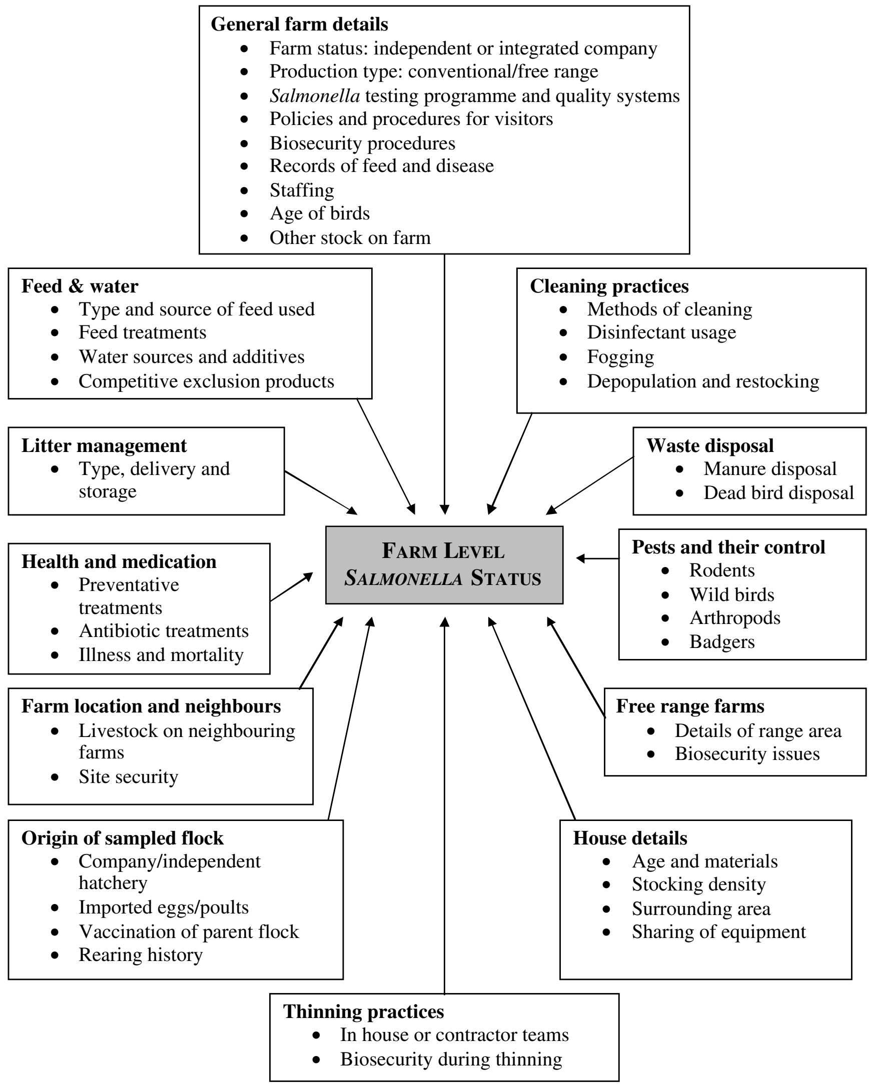

Fig. 1. Farm and house factors on which data were gathered using the two questionnaires.

(Fig. 1). All data from the two questionnaires and test results from the sampled holdings in the UK were collated, entered and validated by trained data entry staff into a Microsoft Access 2000 database at the Centre for Epidemiology and Risk Analysis, VLA, Weybridge.

All statistical analyses were conducted using Stata/IC version 10.0 for Windows (Stata Statistical Software Release version 10.0, StataCorp LP, USA) using the survey commands for analysing complex survey design data [15, 16].

Statistical methods

A holding was classified as *Salmonella*-positive if *Salmonella* spp. was cultured from at least one of the five pairs of boot-swab samples. Similarly, a holding was classified as positive for *Salmonella* Typhimurium (ST) if ST was cultured. All observations were weighted by the inverse of the selection probability in each of four holding size strata [15, 17] and a finite population correction was also applied to adjust standard errors for sampling a large proportion of each strata. Two separate models were developed. For the first model, the outcome was the detection of *Salmonella* (any serovar). In the second model the outcome was detection of ST *vs.* farms where no ST was detected. These two outcomes were considered of most interest to the turkey industry in terms of realistic farm-level interventions.

The questions that formed the questionnaires were critically evaluated and a total of 420 variables were created. Univariate analysis was carried out to examine the potential association of each variable with

Salmonella status based on Pearson's χ^2 statistic with the Rao & Scott second-order correction [16]. Any biologically plausible variables significant at $P \leqslant 0{\cdot}10$ for the *Salmonella* spp. model, or $P \leqslant 0{\cdot}20$ for the ST model, were then assessed for inclusion in the multivariate models to estimate the association of holding-level *Salmonella* status with various risk factors. Variables that showed collinearlity (determined using the _RMCOLL command in Stata) were not assessed in the model at the same time and variables with insufficient observation numbers, which would have compromised the power of the model, were excluded. Holding size, flock type, belonging to one of the 10 largest UK turkey-producing companies and seasonal production (defined as fattening a single crop of turkeys per year) were included in both models as *a priori* potential confounders because it was considered that these four variables may be associated independently with the outcomes and potential risk factors. Due to the large number of variables being considered, a forwards stepwise approach introducing variables in order of significance was used. Variables were included or excluded from the model based on the adjusted Wald test statistic and only variables with $P_{\text{Wald}} < 0{\cdot}05$ were retained in the final model [18]. As a final step, variables that were not selected initially were added back individually and retained if significant at $P_{\text{Wald}} < 0{\cdot}05$. Model fit was assessed using the Hosmer–Lemeshow goodness-of-fit test after removing the weighting [18]. Where more than one combination of variables was possible, the best-fit model which demonstrated the most useful combination regarding farm interventions for *Salmonella* control was selected.

RESULTS

Response to voluntary questionnaire

A total of 252 (79·5%) of the 317 fattening turkey holdings enrolled in the EU survey completed the additional questionnaire on risk factors. Analysis of the farms which responded, compared to those that did not respond, indicated no significant difference in terms of production type, *Salmonella* status, *Salmonella* vaccination status of the flock, or whether the farm was a seasonal producer. Respondents included representation from all four regions of the UK, with no significant difference in response between region (response rate between 76% and 100%). Smaller farms were less likely to respond to the additional questionnaire, with a response rate of 72% in farms with <5000 birds; this rate of response increased as holding size increased, with a 100% response rate from the largest holdings of >50 000 birds. This trend was statistically significant (Pearson's $\chi^2 = 18{\cdot}9$, $P < 0{\cdot}001$), but was not considered to present unacceptable bias.

Table 1. *Frequency of isolation of serovars of* Salmonella

Salmonella serovars isolated	Number (%) of positive holdings* ($n = 252$)
Kottbus	45 (17·9)
Typhimurium	15 (6·0)
Derby	13 (5·2)
Indiana	6 (2·4)
Kedougou	4 (1·6)
Newport	3 (1·2)
Saint Paul	2 (0·8)
Agama	2 (0·8)
Anatum	2 (0·8)
13,23:I:-	2 (0·8)
Stourbridge	1 (0·4)
Senftenberg	1 (0·4)

* Some holdings had more than one serovar.

Prevalence of *Salmonella* spp. and ST

Salmonella was isolated from 86 of the 252 holdings giving a *Salmonella* spp. prevalence of 34·1%. Table 1 shows the serovars of *Salmonella* isolated. The most common serovar isolated was *S*. Kottbus, occurring in 45 (52%) of the 86 positive holdings. The second most common serovar was ST, isolated from 15 (17%) of the 86 positive holdings, giving an overall ST prevalence of 6·0%.

Factors associated with farm-level *Salmonella* status

Univariate analysis

Of the variables initially considered, 54 variables significant at $P \leqslant 0{\cdot}10$ (*Salmonella* spp. model) and 50 variables significant at $P \leqslant 0{\cdot}20$ (ST model) were considered for inclusion in the multivariate models. Tables 2 and 3 show the variables significantly associated ($P < 0{\cdot}05$) with *Salmonella* spp. and ST in the univariate analysis, in addition to the univariate results of those variables which were included in the multivariate models.

Table 2. *Results of univariate analysis of factors significantly associated* ($P<0{\cdot}05$) *with* Salmonella (*all serovars*)

Variable	Level	No. farms sampled	% positive	χ^2 *P* value	OR (95% CI)
Farm details and policies					
Number of birds on holding	<4999	118	42		Baseline
	5000–9999	36	39	0·741	0·90 (0·47–1·72)
	10 000–49 999	68	22	0·007	0·40 (0·21–0·78)
	>50 000	30	27	0·027	0·51 (0·28–0·93)
Part of one of 10 largest UK turkey-producing companies	No	134	44		Baseline
	Yes	118	23	0·010	0·50 (0·30–0·85)
Seasonal production (1 crop per year)	No	130	27		Baseline
	Yes	122	42	0·020	1·83 (1·10–3·05)
Farm has a *Salmonella* testing programme	No	131	46		Baseline
	Yes	120	22	<0·001	0·38 (0·22–0·65)
Staff trained in standard operating procedures	No	28	57		Baseline
	Yes	185	31	0·041	0·45 (0·21–0·97)
Standard operating procedures independently audited	N	76	43		Baseline
	Yes	134	28	0·028	0·52 (0·29–0·93)
Wood shavings used as litter on farm	No	136	46		Baseline
	Yes	116	21	<0·001	0·30 (0·17–0·51)
Straw used as litter on farm	No	68	16		Baseline
	Yes	184	41	<0·001	4·95 (2·84–8·64)
Formaldehyde products used in feed	No	211	36		Baseline
	Yes	36	22	0·020	0·37 (0·16–0·85)
Dead birds disposed of by incineration on farm	No	217	36		Baseline
	Yes	35	23	0·048	0·44 (0·19–0·99)
Disinfectant foot dips used	No	57	51		Baseline
	Yes	195	29	0·008	0·46 (0·26–0·82)
Separate boots used for each house	No	159	38		Baseline
	Yes	91	25	0·030	0·54 (0·31–0·94)
Visitors park vehicles outside farm gate	No	164	36		Baseline
	Yes	83	29	0·016	0·50 (0·29–0·88)
Public footpath through farm	No	153	27		Baseline
	Yes	98	45	0·035	1·73 (1·04–2·90)
Staff also work with pigs	No	231	32		Baseline
	Yes	21	57	0·041	2·41 (1·03–5·61)
Staff also work with cattle	No	175	38		Baseline
	Yes	77	26	0·026	0·52 (0·30–0·92)
Pigs present on contiguous farms	No	213	37		Baseline
	Yes	39	21	0·007	0·33 (0·15–0·74)
Dogs on neighbouring farms	No	146	41		Baseline
	Yes	106	25	0·006	0·47 (0·28–0·81)
House and bird details					
Flock type sampled	Conventional	205	28		Baseline
	Free range	47	60	<0·001	3·27 (1·75–6·11)
Flock origin	Independent hatchery or other	116	43		Baseline
	Company hatchery	121	24	0·014	0·51 (0·30–0·87)
Origin of sampled flock	Company A	42	76		Baseline
	Company B	39	26	<0·001	0·08 (0·03–0·20)
	Company C	40	10	<0·001	0·05 (0·01–0·16)
	Company D	26	42	<0·001	0·19 (0·08–0·46)
	Other specified company	58	17	<0·001	0·06 (0·02–0·15)
	Did not specify company	47	40	0·001	0·23 (0·10–0·54)
Were preventative treatments used during the first few days?	No	166	37		Baseline
	Yes	75	27	0·039	0·54 (0·30–0·97)

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Table 2 (*cont.*)

Variable	Level	No. farms sampled	% positive	χ^2 *P* value	OR (95% CI)
History of intestinal illness in sampled flock	No	195	41		Baseline
	Yes	50	10	0·000	0·16 (0·06–0·42)
Has any antibiotic been given during the life of the flock?	No	153	41		Baseline
	Yes	91	25	0·011	0·49 (0·28–0·85)
Age of oldest birds in sampled flock	< 112 days	56	21		Baseline
	112–133 days	68	28	0·136	1·85 (0·82–4·18)
	134–146 days	59	41	0·008	2·99 (1·33–6·69)
	⩾ 147 days	69	45	0·007	2·90 (1·33–6·30)
Litter type in sampled house	Straw only	151	43		Baseline
	Wood shavings only	73	18	< 0·001	0·27 (0·14–0·51)
	Straw and wood shavings	18	33	0·650	0·79 (0·28–2·20)
	Other type	10	20	0·299	0·47 (0·11–1·97)
House walls made of metal	No	221	32		Baseline
	Yes	31	48	0·044	2·14 (1·02–4·47)
Cleaning practices					
Manure disposed of on adjacent arable land	No	157	27		Baseline
	Yes	95	46	0·001	2·53 (1·50–4·24)
Manure disposed of on distant grazing land	No	218	37		Baseline
	Yes	34	15	0·009	0·29 (0·12–0·73)
Cleaning practices following depopulation (defined sequentially)	Litter removed only	23	57		Baseline
	Floors and walls washed only	6	50	0·944	0·94 (0·18–4·96)
	Wash and disinfected only	75	44	0·143	0·52 (0·22–1·25)
	Wash and disinfect, including fittings and outside area	146	25	0·005	0·29 (0·13–0·69)
Disinfectant used during cleaning processes	No	12	67		Baseline
	Yes	238	33	0·012	0·23 (0·07–0·72)
Formaldehyde used for Disinfection	No	207	37		Baseline
	Yes	33	15	0·003	0·25 (0·10–0·63)
Houses are fogged	No	172	37		Baseline
	Yes	68	25	0·048	0·54 (0·30–0·99)
Time between disinfection and laying new bedding	1 day	6	50		Baseline
	> 1 day	176	28	0·038	0·20 (0·04–0·91)
Pests					
Wild birds access poultry houses	No	147	27		Baseline
	Yes	103	45	0·014	1·91 (1·14–3·19)
Game birds seen regularly around houses	No	152	28		Baseline
	Yes	93	44	0·014	1·94 (1·14–3·28)
Free-range farms only ($n = 55$): game birds considered a problem on range areas?	No	48	56		Baseline
	Yes	7	86	< 0·001	23·3 (5·61–97·1)
Thinning					
Thinning carried out	No	149	42		Baseline
	Yes	103	22	0·003	0·44 (0·25–0·75)
Thinning team	Contractors	59	17		Baseline
	Company	38	32	0·036	2·79 (1·07–7·26)
Protective clothing provided for the thinning team	No or sometimes	50	10		Baseline
	Always	52	33	0·046	3·47 (1·02–11·8)
Number of additional people entering the house for thinning	< 3	21	52		Baseline
	> 3	81	14	< 0·001	0·13 (0·04–0·39)
Thinning performed late evening	No	74	26		Baseline
	Yes	28	11	0·030	0·24 (0·06–0·87)

OR, Odds ratio (weighted by sampling probability of the holding); CI, confidence interval.

Table 3. *Results of univariate analysis of factors significantly associated* ($P<0{\cdot}05$) *with* Salmonella *Typhimurium and those included in the multivariate model*

Variable	Level	No. farms sampled	% positive	χ^2 *P* value	OR (95% CI)
Number of birds on holding	<4999	118	8		Baseline
	5000–9999	36	6	0·621	0·71 (0·18–2·75)
	10 000–49 999	68	6	0·644	0·76 (0·23–2·48)
	>50 000	30	0		n.a.
Part of one of 10 largest UK turkey-producing companies	No	134	7		Baseline
	Yes	118	5	0·409	1·53 (0·55–4·21)
Seasonal production (1 crop per year)	No	130	6		Baseline
	Yes	122	6	0·601	0·77 (0·28–2·09)
Contract rearer for a turkey company	No	169	5		Baseline
	Yes	83	8	0·046	2·79 (1·02–7·64)
Comprehensive written hygiene and staff training procedures and records on medication, illness, feed and sample analysis	No	114	10		Baseline
	Yes	138	3	0·033	0·27 (0·08–0·90)
Farm has a *Salmonella* testing programme	No	131	8		Baseline
	Yes	120	3	0·156	0·43 (0·13–1·38)
Water supply	Borehole/spring	43	9		Baseline
	Mains	209	5	0·024	0·28 (0·09–0·85)
Flock type sampled	Conventional	205	6		Baseline
	Free range	47	6	0·964	0·97 (0·28–3·34)
Salmonella found in sampled house in the last crop	No	111	3		Baseline
	Yes	4	25	0·009	27·4 (2·29–327)
Litter type in sampled house	Woodchip/straw	164	8		Baseline
	Shavings	88	2	0·024	0·20 (0·05–0·81)
House walls made of brick	No	125	8		Baseline
	Yes	127	4	0·037	0·32 (0·11–0·93)
Soil surface surrounding turkey house	No	215	5		Baseline
	Yes	37	11	0·028	3·45 (1·14–10·4)
Stored items in area surrounding turkey house	No	246	6		Baseline
	Yes	6	17	0·173	3·98 (0·54–29·1)
Evidence of mice (droppings, tracks or live mice seen)	No	172	3		Baseline
	Yes	78	10	0·003	5·05 (1·73–14·8)
Use wax blocks in rodent control programme	No	189	7		Baseline
	Yes	63	2	0·039	0·11 (0·01–0·89)
Use traps in rodent control programme	No	235	5		Baseline
	Yes	17	18	0·048	3·80 (1·01–14·2)

OR, Odds ratio (weighted by sampling probability of each holding); CI, confidence interval.

Multivariate results

Table 4 shows the final multivariate model with adjusted odds ratios (OR) and 95% confidence intervals (CI) for factors associated with *Salmonella* spp. at the farm level. Table 5 shows a similar model, but specific to ST.

Factors associated with farm-level *Salmonella* status (all serovars)

Of the *a priori* confounding variables, only on farms with seasonal production was there a significantly reduced risk of *Salmonella* (OR 0·31), compared to those which rear multiple crops of turkeys per year.

The majority (58%) of turkey flocks in this study were sourced from four companies. The risk of isolating *Salmonella* varied between these companies, as shown in Table 4. For example, in comparison to company A there was a significant decrease in risk for farms which sourced their poults from companies B or C (OR 0·05 and 0·15, respectively), but there was no significant difference between companies A and D.

The use of wood shavings as the sole type of litter had a protective effect for *Salmonella* (OR 0·21)

Table 4. *Multivariate analysis of factors associated with holding level* Salmonella (*all serovars*) *status* (*n* = *243*)

Variable	Level	No. farms sampled	% positive	P_{Wald}	OR (95% CI)
Number of birds on holding	< 4999	118	42		
	5000–9999	36	39	0·857	1·11 (0·34–3·58)
	10 000–49 999	68	22	0·493	0·68 (0·22–2·08)
	> 50 000	30	27	0·129	0·18 (0·02–1·64)
Part of one of 10 largest UK turkey-producing companies	No	134	44		Baseline
	Yes	118	23	0·126	0·46 (0·17–1·24)
Flock type sampled	Conventional	205	28		Baseline
	Free-range	47	60	0·797	0·87 (0·32–2·38)
Origin of sampled flock	Company A	42	76		Baseline
	Company B	39	26	< 0·001	0·05 (0·02–0·15)
	Company C	40	10	0·035	0·15 (0·03–0·87)
	Company D	26	42	0·927	1·12 (0·09–13·7)
	Other specified company	58	17	< 0·001	0·04 (0·01–0·14)
	Did not specify company	47	40	< 0·001	0·17 (0·06–0·46)
History of intestinal illness in sampled flock	No	195	41		Baseline
	Yes	50	10	< 0·001	0·17 (0·06–0·45)
Litter type in sampled house	Straw only	151	43		Baseline
	Wood shavings only	73	18	< 0·001	0·21 (0·09–0·49)
	Straw and wood shavings	18	33	0·646	1·33 (0·39–4·50)
	Other type	10	20	0·244	0·48 (0·14–1·65)
Disinfectant used during cleaning processes	No	12	67		Baseline
	Yes	238	33	0·028	0·25 (0·07–0·86)
Dead birds disposed of by incineration on farm	No	217	36		Baseline
	Yes	35	23	0·037	0·29 (0·09–0·92)
Seasonal production (1 crop per year)	No	130	27		Baseline
	Yes	122	42	0·025	0·31 (0·11–0·86)
Staff also work with cattle	No	175	38		Baseline
	Yes	77	26	0·002	0·31 (0·15–0·64)
Pigs present on neighbouring farms	No	213	37		Baseline
	Yes	39	21	0·031	0·38 (0·15–0·92)

OR, Odds ratio; CI, confidence interval.
Hosmer–Lemeshow test: χ^2 (8 d.f.) = 5·57, P = 0·6948.

compared to using straw only, but this effect was lost when wood shavings were used in combination with straw as bedding. The use of disinfectant in cleaning following depopulation also significantly reduced the likelihood of finding *Salmonella* on a holding (OR 0·25) compared to those farms which did not use disinfectant.

If a flock had a history of intestinal illness prior to sampling, the likelihood of isolating *Salmonella* was significantly reduced (OR 0·17) compared to those which had no such history. While birds which had suffered intestinal illness prior to sampling were significantly more likely to have received antibiotic treatment (OR 20·8, 95% CI 8·75–48·7, P < 0·001), the use of antibiotic medications itself did not show any significant association with *Salmonella* in the final model. There was significantly less risk of isolating *Salmonella* from a holding if dead birds were disposed of by incineration on farm (OR 0·29) compared to farms which used off-site carcase disposal methods.

The presence of pigs on neighbouring farms was shown to confer a significant protective effect (OR 0·38) compared to holdings without a contiguous pig farm. Having staff that also worked with cattle was shown to reduce the likelihood of isolating *Salmonella* from a holding (OR 0·31) compared to those farms whose staff did not work with cattle.

Factors associated with farm-level ST status

Size of holding, production type, being part of one of the 10 major UK turkey-producing companies and seasonal production were included in the final model

Table 5. *Multivariate analysis of variables associated with holding level* Salmonella *Typhimurium status* (*n* = *249*)

Variable	Level	No. farms sampled	% positive	P_{Wald}	OR (95% CI)
Number of birds on holding	<4999	118	8		
	5000–9999	36	6		
	10 000–49 999	68	6		
	>50 000	30	0	0·137	0·57 (0·27–1·20)
Part of one of 10 largest UK turkey-producing companies	No	134	7		Baseline
	Yes	118	5	0·071	3·35 (0·90–12·5)
Seasonal production (1 crop per year)	No	130	6		Baseline
	Yes	122	6	0·134	0·31 (0·06–1·44)
Farm has a *Salmonella* testing programme	No	131	8		Baseline
	Yes	120	3	0·040	0·23 (0·05–0·93)
Water supply	Borehole/spring	43	9		Baseline
	Mains	209	5	0·009	0·19 (0·05–0·65)
Flock type sampled	Conventional	205	6		Baseline
	Free-range	47	6	0·221	0·47 (0·14–1·59)
Soil surface surrounding turkey house	No	215	5		Baseline
	Yes	37	11	0·043	2·70 (1·03–7·04)
Stored items in area surrounding turkey house	No	246	6		Baseline
	Yes	6	17	0·002	5·20 (1·31–20·6)
Evidence of mice (droppings, tracks or live mice seen)	No	172	3		Baseline
	Yes	78	10	0·008	4·71 (1·51–14·7)
Use wax blocks in rodent control programme	No	189	7		Baseline
	Yes	63	2	0·038	0·09 (0·01–0·87)

OR, Odds ratio, CI, confidence interval.
Hosmer–Lemeshow test: χ^2 (8 D.F.) = 11·28, P = 0·1863.

as before, although none were significantly associated with presence of ST on the holding compared to flocks which were either positive for other *Salmonella* serovars or negative.

Those farms which had a *Salmonella* testing programme in place were significantly less likely to test positive for ST (OR 0·23) compared to those farms which had no *Salmonella* test programme. Use of mains water also significantly reduced the likelihood of testing positive for ST (OR 0·19), the source of water for the majority (85%) of non-mains water supplied farms was a borehole. A soil surface and storage of items in the area surrounding the turkey house increased the risk of isolating ST from a flock (OR 2·70 and 5·20, respectively) compared to those flocks which had no soil surface or stored items surrounding the turkey house.

The presence of mice (either live mice, mice droppings or tracks observed) on a holding resulted in more than a fourfold increase (OR 4·71) in the probability of isolating ST compared to those farms which had no evidence of mice. The use of wax blocks in the farm rodent control programme as opposed relying solely on other means (e.g. other bait types, contact rodenticides or trapping) was protective against ST (OR 0·09).

DISCUSSION

This study has identified a number of risk factors for *Salmonella* infection on fattening turkey units. As a cross-sectional study, the analysis was not able to differentiate factors related to introduction of *Salmonella* onto a holding from factors that facilitate the persistence of *Salmonella* infection on a holding. The large number of variables analysed may increase the potential of type I error and measures were therefore taken to reduce the likelihood of this occurrence. These included consideration of the biological importance and plausibility of variables prior to inclusion in the models, and their significance at the univariate level.

Seasonally produced flocks were shown to be around three times less likely to have *Salmonella* than those flocks on holdings rearing more than one crop of birds per year. To our knowledge, this has not been previously demonstrated in turkey meat production. An extended period between batches of birds is conducive to comprehensive cleaning and disinfection, a reduction in environmental contamination and environmental reservoirs of infection and it interrupts the carry-over of infection that may occur between successive batches.

The risk of isolating *Salmonella* varied significantly between the companies from which the poults originated. Other studies have also identified the source company or hatchery of the flock as a risk factor for *Salmonella* in poultry [11, 19–21]. This reinforces the importance of *Salmonella* control at each level in the production pyramid. It was not possible to perform separate analyses for each *Salmonella* serovar due to the small numbers of farms with some of these serovars. However, in the UK, at the time of this study, *S*. Kottbus was the predominant isolate from both breeding and fattening flocks [12].

The use of wood shavings alone as litter was found to have a significant protective effect on *Salmonella* infection in this study, yet this effect was lost when wood shavings were used in combination with straw. This is an interesting finding as there has been little research to substantiate the persistence of *Salmonella* in different types of poultry litter. It is possible that properties intrinsic to wood shavings may inhibit *Salmonella* organisms, or that superior drainage offered by shavings as opposed to straw may encourage *Salmonella* persistence on the latter. It is also possible, given its favourability by rodents as harbourage, that straw may become contaminated in the field and during storage.

Incineration of dead birds on farm is a marker for good practice for disease control. On-farm incineration is likely to reduce environmental contamination and attraction of pests that can be associated with carcases, especially if carcases are stored insecurely on the farm for any length of time prior to removal. A history of intestinal illness in the sampled flock was an unexpected protective factor, but the variable showed moderate correlation with the use of antibiotics in the flock. The use of such agents, targeted at coliform-induced intestinal disease, is likely to have reduced excretion of *Salmonella*.

It is widely reported that pigs may act as a source of *Salmonella* infection [22, 23]. It is therefore surprising that this study identified the presence of pigs on neighbouring farms as a protective factor. It is possible that turkey farms situated close to pig units may employ stricter biosecurity measures although no correlation could be determined from our data. Farms where staff also work with cattle were less likely to have *Salmonella*, but there does not appear to be a ready biologically plausible explanation for this. It may, however, be linked with other unidentified protective farm management factors. It is also possible that both these variables are negatively correlated with poultry density and further analysis of location data and population distribution may be of use.

The UK Code of Practice for the Control of *Salmonella* in Commercial Turkey Flocks [24] underlines the importance of thorough cleaning and disinfection between flocks and this study confirmed the value of disinfection in reducing the risk of *Salmonella* infection. This is in accord with other studies, although many workers have emphasized the importance of thorough cleaning prior to disinfection as most disinfectants are readily inactivated by residual organic matter [25–27]. Persistent contamination of the environment with *Salmonella* is a major problem on poultry units and a source of infection to new batches of birds [28–30].

Holding size, while significant at the univariate level, did not retain association with farm *Salmonella* status in the final models. This is contrary to risk factor studies in laying hens [27] and to the overall EU baseline turkey survey, where larger holdings of birds were more likely to be *Salmonella* positive [12]. Similarly in contrast to the EU survey, our study did not identify free-range production as a particular risk factor for *Salmonella* infection on fattening turkey holdings. Being part of one of the 10 major turkey-producing companies in the UK was not significant in the multivariate models, but retained as a potential confounder.

ST accounted for 13·4% of the cases of salmonellosis in humans in the UK in 2007 and is considered to be one of the priority *Salmonella* serovars by EFSA [1]. The EU has set a reduction target for the priority serovars, *S*. Enteritidis and ST of no more than 1% of flocks to test positive by 31 December 2012 [31]. It is therefore of particular importance to consider specific risk factors for ST. ST is also one of the main serovars that can sometimes cause clinical disease in turkeys [32] and is often associated with multiple antibiotic resistance [33, 34].

Having a *Salmonella* testing programme, using mains water and using wax blocks to control rodents were all shown to reduce the risk of testing positive for ST. A testing programme should not only enable units to promptly detect and respond to *Salmonella* infection, before it spreads and becomes persistent, but also help in developing action plans to eliminate infection. Mains water will have undergone statutory purification and testing to ensure it complies with certain health and safety standards and therefore have a much lower risk of being contaminated with *Salmonella* – although water is unlikely to be a source of *Salmonella*, the lack of mains water may be an indication for other reduced husbandry standards. The provision of rodenticide via wax blocks is currently considered one of the most effective methods of mouse control, compared to whole wheat bait which is normally only suitable for rats.

The role of mice in *Salmonella* epidemiology on poultry farms is well documented in the literature [28, 29, 35]. In our study the presence of mice on a holding was significantly associated with ST infection specifically. Infected mice can act as a vector of *Salmonella*, amplifying and spreading the organism between houses, consecutive flocks and possibly even to neighbouring units [29]. In our study, rats were not found to pose a statistically significant risk for *Salmonella* infection on turkey farms, but rats are more likely to be observed and effectively controlled, minimizing breeding populations.

Both the storage of items around the turkey accommodation and a soil surround were identified as risk factors for ST. Soil, vegetation and clutter around houses are likely to increase the risk of infestation of houses by rodents. Such surfaces and items are also difficult to disinfect and while not significant in the final model, disinfection of areas surrounding the house was shown to reduce the risk of *Salmonella* (all serovars) in the univariate analysis.

The comprehensive questionnaire examined a very wide variety of issues relating to turkey production and farm management. A number of variables that were significant in the univariate analysis and provided biologically plausible explanations of farm-level *Salmonella* status were not confirmed in the multivariate models, although the power of detection may have been compromised by the small sample. Variables specific to free-range farms or those farms which operate thinning policies had to be excluded from the multivariate models for this reason. Similarly, the EU survey demonstrated that vaccination conferred significant protection against *Salmonella* [12]. Of the respondents in this study, only one farm indicated that the sampled flock was vaccinated against *Salmonella* and therefore it could not be used as a study variable, although it is an intervention step which may be considered as part of the package of control measures to limit ST infection in turkeys.

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DECLARATION OF INTEREST

None.

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