ENVIRONMENT AND HEALTH

Influence of Resident Salmonella on Contamination of Broiler Flocks

C. LAHELLEC, P. COLIN, G. BENNEJEAN

Ministere de l'Agriculture, Direction de la Qualite, Services Veterinaires, Station Experimentalte d'Aviculture, B.P. 9, 22440 Ploufragan, France

J. PAQUIN, A. GUILLERM, and J. C. DEBOIS

Etablissements Guyomarc'H, B. P. 234, 56006 Vannes, Cedex, France

(Received for publication August 29, 1985)

ABSTRACT An epidemiological survey was made of 5329 samples from 10 poultry operations to determine the relationship between total poultry farm environment and incidences of *Salmonella* contamination of broiler flocks. Samples were analyzed from walls, drinkers, feeders, litter, insects, water, chicks, broilers, and feed to determine the effect of common sanitary practices on *Salmonella* contamination of flocks.

Results indicated that although similar hygienic practices had been taken on the 10 poultry farms examined, great variation exists in *Salmonella* contamination among the farms. Among the sources studied, the most important source of contamination was determined to be the resident *Salmonella* of the flock *i.e.*, the strain isolated on chicks' first day in the poultry house. This source was more important than *Salmonella* isolated during the rearing period. However, the precise conditions of *Salmonella* contamination in poultry flocks remain to be elucidated.

(Key words: Salmonella, broilers, environment, contamination, poultry farms)

1986 Poultry Science 65:2034-2039

INTRODUCTION

One of the most perplexing problems in the control of Salmonella in foods is the role of carriers in animal and human populations. Barnes (1972) indicated that no definitive solution to poultry carcass contamination could be found unless the flocks were Salmonella free. Recently, much attention has been directed to the examinations of sources and routes of Salmonella contamination of poultry flocks. Sources of contamination implicated include chicks (Snoeyenbos et al., 1967; Ehrsam and Spillman, 1981; Bolder et al., 1982), feed (Snoeyenbos et al., 1967; Mackenzie and Bains, 1976), equipment, rodents, insects, and humans (Morgan-Jones, 1982). Although Spillman and Ehrsam (1983) emphasized the need to have clean environment and hygienic conditions to prevent Salmonella contamination of flocks, it is difficult to ascertain whether such measures alone are adequate to prevent contamination. The purpose of this epidemiological investigation was to determine the incidence of different serotypes of Salmonella in poultry farms with similar hygienic practices at the beginning and the end of the rearing period to ascertain the source of Salmonella contamination in flocks.

MATERIALS AND METHODS

Ten poultry houses were selected for investigation using the following criteria: 1) isolation from other industrial farms, 2) protection against birds and rodents, 3) motivation and efficiency of the farmers, and 4) ease of cleaning and disinfection of silos. Buildings had dirt floors with walls made of asbestos cement and conglomerate.

Houses were washed before manure removal, followed by sweeping and spraying with a phenol-base disinfectant. Formaldehyde fumigation (10 to 15 kg of formaldehyde powder/1000 m^2) was done 48 hr before arrival of the chicks. Manure was removed after each brood and, on average, the houses remained vacant for 2 weeks between broods. Sixteen to 21 successive flocks from each of 9 poultry houses were examined; in a 10th house, (C) only six flocks were studied. Samples were collected at the time of chick arrival and at the end of the rearing period to ascertain the possible succession of *Salmonella* during rearing period. Birds were about 42 days of age when slaughtered.

Bacteriological samples were taken at the time of arrival of chicks and at the end of the

rearing period. Environmental swabs (500 cm² each) were made by use of sterile linen (40 \times 40 cm) moistened with sterile buffered peptone solutions (peptone 10 g, NaCl 5 g, NaH₂PO₄ 9.0 g, KHPO₄ 1.5 g, distilled water 1000 ml, pH 7.2). For each house three swabs each were taken from walls, drinkers, and feeders.

Litter samples (50 g) in triplicate were taken from three to four places in the broiler house and collected in sterile plastic bags. Insects were also analyzed in one experiment.

Water samples (50 to 100 ml) were taken from four or five fountains with a sterile syringe. Skin and viscera of day-old chicks (5 per house per sampling time) were studied. Feed samples (200 g) were collected from trucks before feed was stored in bulk feeders. Numbers examined varied according to poultry house.

A total of 5329 samples was examined: 2291 from the first day, 2105 at the end of the rearing period, 557 from feed, and 376 from carcasses examined in the processing plant.

Salmonella were isolated by enrichment serology (Sperber and Deibel, 1969; Boothroyd and Baird-Parker, 1973) as adapted for poultry products by Lahellec and Colin (1977) and by conventional method. The conventional method entailed preenrichment at 37 C for 18 h in buffered peptone solution followed by enrichment in tetrathionate broth (no. 41302, Institut Pasteur, Lille) and then plating on brilliant green agar (Kristensen medium, no. 64467, Institut Pasteur, Paris). Strains were identified by their biochemical properties using the miniaturized methods of Fung and Hartman (1975) as adapted by Lahellec and Colin (1981).

RESULTS AND DISCUSSION

Table 1 indicates that 34 flocks out of 180 were found to be contaminated on the first day. These results include chicks (in 6 cases), samples from the environment (in 25 cases), or chicks and environment (in 3 cases). Table 1 shows that the percentage of samples found to be contaminated by resident strains on the first day in the house is guite low, but it increased significantly at the end of the rearing period. Absence of detectable Salmonella on the first day does not necessarily mean that contamination will not occur during the rearing period. In addition, serotypes identified the 1st day may remain in the environment and can be found at the end of the rearing period of successive flocks even if not recovered in intermediate flocks, as

shown in Table 2. In fact, when examining different serotypes isolated from the 10 poultry houses (Table 3), it becomes apparent that the resident *Salmonella* in the poultry farms is far more important than *Salmonella* isolated later during the rearing period in terms of contamination of the flocks.

Nevertheless, additional serotypes are recovered during the rearing period, as shown in Table 1. Of course, there is a possibility that sample size and numbers were not large enough to detect the presence of *Salmonella* on the 1st day, but other possible sources have also to be taken under consideration.

In a separate experiment, feed contamination by *Salmonella* was examined. From a total of 557 samples only 7 were contaminated. The strains included *S. saint paul, S. senftenberg, S. livingstone,* and *S. infantis,* recovered from the feed delivered to 4 farms. Identical serotypes were found in only two cases in both the feed and the poultry house at the end of the rearing period. This excludes feed as a major source of contamination of *Salmonella* in poultry houses.

Other sources, such as the farmers themselves, should be considered, such an approach was not possible during the present experiment.

Samples were examined from poultry farms for sources of *Salmonella* contamination in the environment and chicks (Table 4) on the 1st day of chicks arrival. Walls of poultry houses provided the highest frequencies of recovery of *Salmonella*. The contamination of walls might have come from soils, insects, etc. In general, at the end of the rearing period drinkers had the highest incidence of *Salmonella* contamination, although in one house chicks had the highest and in another feeders had the highest.

It is important to note that although all houses practice the same hygienic procedures, high variations of contamination occur among houses at the end of the rearing period.

This was a qualitative *Salmonella* investigation (i.e., all or none in each sample) only. When quantitative investigations are made (i.e., number of *Salmonella* in each sample) other facets of *Salmonella* contamination may be discovered.

In conclusion, in this study resident Salmonella, and not other serotypes isolated during the rearing period, constituted the most important contamination source of Salmonella in the flock. However, other studies indicate that newly placed chicks may also be an important source of contamination. The conditions of colonization

22.9	20/237 481/2105 9	20/237 9	85/224 2,5,7,10,13	37/224 1,2,3,4,5,11	42/223 1,4,6,7,10	31/239 54/238 4,7,9 1,7,9	31/239 4,7,9	58/189 3,7	6/72 8	63/236 1,3,4,5,6,7	85/223 1,2,3,4,5,6	no. of samples examined Serotypes ¹
53.3	7/20 96/180	7/20	14/19	5, 2016 1/2	9/19	11/21	11/20 11/21	11/16	1/6	11/20	14/20	No. of flocks contaminated/ no. of flocks examined No. of samples contaminated/
		:	5,10	1,2,5%,12		1,14	4,7	2	•	1,2,3,5,7	2,4,5,6	Serotypes Last day
2.2	0/260 50/2288	0/260		7/240	3/236	3/253 6/266	3/253	4/202	0/80	6/249		no. of samples examined
18.9	34/180	0/20	8/19	7/1%	2/19	3/21	2/20	4/16	9/0	5/20	3/20	no. of flocks examined No. of samules contaminated/
(0/)				erno Lib								First day No. of flocks contaminated/
	Total	ſ	Ι	sity (I II	IJ	F	Е	D	U	В	A	
				niver	in France	Poultry farm in France	Po					
			aring period	oe end af the re	st day and at th	on the fir	try farms	on of pou	ıtaminatı	almonella con	TABLE 1. S	
				nals.org/ at Penn S								
				jour								

LAHELLEC ET AL.

S. beidelberg	S. beidelberg (walls, feeders)	S. infantis		S. saint paul				
1/12	2/13	8/12	0/13	6/12	0/11	0/12	0/13	6
S. petaelberg	S. <i>Detaelberg</i> (walls)			5. saint paul		o. wompson		
12/12	1/13	0/0	0/13	6/12	0/13	7/12	0/12	ŝ
S. Detaelberg		016		3. saint paut	o. saint paut (soil)	s. wompson		
11/12	0/13	@/10	0/13	9/12	1/13	5/9	0/13	4
3. nemenery	S. veuceneerg (soil)	tober		o. sum pum		o. wompson		
11/12	1/12	0/12 0/12	0/13	1/11	0/13	10/12	0/13	6
S. heidelberg		o) oi		S. saint paul		S. thompson		
5/12	0/15	<u>0</u> /11	0/13	1/12	0/12	1/10	0/10	2
S. heidelberg	S. <i>beildelber</i> g (walls, soil)	an infantis outa	S. <i>infantis</i> (chicks)	S. saint paul		S. thompson	S. thompson (feeders)	
8/12	3/13	у 10	2/11	4/12	0/13	6/9	1/11	1
		ersit	- (No. positive samples/no. samples examined) -	. positive samples/n	0N)			
Last day	First day	Fast day	First day	Last day	First day	Last day	First	Flock ²
	I	State	Η		D		Α	
		enn S	s in France	Poultry Farms in France				
		als.org/at	e of resident Salmone	owing the importanc	ALE 2. Examples sh	TA		
		ttp://ps.oxfordjournals	ttp://ps.oxfordjournals					
		n ł						

¹ The serotype isolated on the first day in poultry house samples is defined as resident Salmonella.

² The flocks numbered were reared in succession in the same poultry house.

SALMONELLA CONTAMINATION OF BROILERS

Poultry farm	Total of <i>Salmonella</i> - contaminated samples	Samples contaminated by serotypes isolated the first day	Serotypes isolated the first day/total serotypes
		(no.)	(%)
А	84	57	67.9
В	62	53	85.5
С	6	0	0
D	56	49	87.5
E	30	17	56.7
F	53	30	56.6
G	42	19	45.2
н	37	34	14.6
1	85	70	82.4
J	20	0	0
Total	475	329	69.3

TABLE 3. Influence of Salmonella serotypes isolated the first day on the final contamination of poultry farms

TABLE 4. Salmonella isolations on the first day according to sources

Poultry farms in			Source of S	Salmonella cor	Itamination		
France	Drinkers	Feeders	Walls	Soil	Feed	Insects	Chicks
			· - · ·	- (no. flocks)			
Α		1	1				1
В		1	1			1	2
С							
D	1	1		1	1		
Е			1				1
F			1	11			21
G	1			1			1
Н			5	1			1
I	1	1	7	3			1
J							
Total	3	4	16	7	1	1	9

¹One flock was positive for chicks and soil.

of poultry guts by *Salmonella* remain to be elucidated.

ACKNOWLEDGMENTS

The authors wish to acknowledge with gratitude technical assistants Allo and Michel. They are indebted to G. Snoeynebos and D.Y.C. Fung for reading the manuscript.

REFERENCES

- Barnes, E. M., 1972. Food poisoning and spoilage bacteria in poultry processing. Vet. Rec. 90:720–722.
- Bolder, N. M., M. C. Van Der Hulst, and R. W.A. W. Mulder, 1982. Salmonellae in broiler chicks. Zootec. Int.

12:20-34.

- Boothroyd, M., and A. C. Baird-Parker, 1973. The use of enrichment serology for *Salmonella* detection in human foods and animal feeds. J. Appl. Bacteriol. 36:165– 172.
- Ehrsam, M., and S. Spillman, 1981. Salmonelles chez le poulet de chair: Aspects epidemiologiques. Page 72 *in:* C. R. VII Congr. Int. World Poult. Sci. Assoc., Oslo, Norway.
- Fung, D.Y.C., and P. A. Hartman, 1975. Miniaturized microbiological techniques for rapid characterization of bacteria. Pages 347–370 in: C. G. Heden and T. Illeni, ed. New Approaches to the Identification of Micro-organisms. John Wiley and Sons, Inc., New York, City, NY.
- Lahellec, C., and P. Colin, 1977. Application de la technique dite de "sero-enrichissement" a la mise en evidence de *Salmonelles* a partir de carcasses de volailles et de

certains produits transformes. Bull. Inf., Sta. Exp. Avic. Ploufragan (Cotes-du-Nord). 17(2):76-78.

- Lahellec, C., and P. Collin, 1981. Miniaturized methods in poultry microbiology. Pages 189–190 in: Rapid Methods and Automation in Microbiology. R. C. Tilton, ed., Am. soc. Microbiol., Washington, DC.
- MacKenzie, M. A., and B. S. Bains, 1976. dissemination of *Salmonella* serotypes from raw feed ingredients to chicken carcasses. Poultry Sci. 55:957–960.
- Morgan-Jones, S. C., 1982. Breaking the *Salmonella* cycle in deep litter houses. Poult. World. 1982 October: 17-

18.

- Snoeyenbos, G. H., V. L. Carlson, B. S. McKensie, and C. F. Smyser, 1967. An epidemiological study of Salmonellosis of chickens. Avian Dis. 11:653–667.
- Sperber, W. H., and R. H. Deibel, 1969. Accelerated procedure for Salmonella detection in dried food and feeds, involving only broth cultures and serological reactions. Appl. Microbiol. 17:533–539.
- Spillman, S., and M. Ehrsam, 1983. Zur Epidemiologie der Salmonelleninfe ktionen beim Mastgeflugel. Schweiz. Arch. Tierheilkd. 125:423–431.