The effect of chlorination on chicken carcasses infected with Salmonellae

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INTRODUCTION

Salmonellae have been found in the environment of poultry-processing factories in the United States (Galton, Mackel, Lewis, Haire & Hardy, 1955; Brobst, Greenberg & Gezon, 1958; Morris & Ayres, 1960) and in Great Britain (Dixon & Pooley, 1961). Poultry has been incriminated as the cause of many outbreaks of human food poisoning in the U.S.A. (Galton & Arnstein, 1960), and in view of the remarkable increase in the consumption of chickens in this country in the last 6 years (Pendry, 1960) it is possible that similar incidents may occur in Britain (Walker, 1960). There is little evidence of this at present (Mann, 1961), although Spink (1960) has suggested that an outbreak of illness in Lancashire due to *Salmonella thompson* might have been due to the consumption of infected broiler chickens.

It was decided to investigate the possibility of reducing the number of salmonellae on infected carcasses by a suitable chlorination procedure. Birds that have been cross-contaminated from other birds in processing factories might be particularly suited to this form of treatment because the contaminating organisms would be confined to the surfaces of the carcasses. Most carcasses that harbour salmonellae after processing have probably been infected in the factory; only the small number that were infected during rearing are likely to harbour salmonellae deep in the tissues where chlorination would be ineffective. Chlorination at some stage in the processing of poultry was advised by Atkinson (1957) in the U.S.A. and by Wadhams (1960) in Great Britain, but there is little information available with regard to its effect on carcasses infected with salmonellae.

A trial was made to determine the effect of chlorine solutions on broiler chicken carcasses that had been artificially infected with mixtures of chicken faeces and salmonella cultures.

MATERIALS AND METHODS

Organisms

Salmonellae that had been isolated from factories that process broiler chickens were used. The serotypes were *Salm. enteritidis, Salm. kentucky, Salm. menston, Salm. montevideo, Salm. thompson, Salm. typhimurium* and *Salm. worthington.*

Inocula

The salmonella strain was grown in nutrient broth at 37°C. for 18 hr. and then at room temperature for a further 24 hr. before use. One ml. of the culture was mixed with an approximately equal volume of chicken faeces in 20 ml. quarter-strength Ringer solution. Further serial dilutions were made in 20 ml. or 100 ml. quantities of Ringer solution, using a fresh pipette for each step; the insides of chicken carcasses were inoculated with either 1 ml. or 0·1 ml. of one of these dilutions. A viable bacterial count of the inoculum was made by inoculating drops of ten-fold dilutions on to the surface of MacConkey agar plates using a method similar to that of Miles & Misra (1938).

Chlorine solutions

'Delsanex' (Delsanex Ltd, Fenner Works, Great Yarmouth, Norfolk), a preparation containing 10–12 % available chlorine, was the chlorinating agent. Volumes of a 1 % solution of Delsanex were added to about 8 l. of water in a polythene pail to produce concentrations of chlorine within the range 50–200 p.p.m. Chlorine concentrations were determined by the potassium iodide method, using a Lovibond All-Purposes Comparator with disk 3/2 I. The test is rapid and capable of use by relatively unskilled workers.

Procedure

Chickens were selected at random from those that had been routinely eviscerated and chilled at the factory. Inocula of 0·1 ml. or 1·0 ml. were pipetted into carcasses and then distributed over the inside surfaces by gauze swabs mounted on steel wire. Carcasses were then immersed, with intermittent agitation to ensure removal of all air, in a polythene pail containing a chlorine solution. The temperature of the solutions was that of the mains water supply and was approximately 10°C. After the appropriate time had elapsed the birds were removed and allowed to drip for 15 min. The carcasses used in the early part of the investigation were then packed in polythene bags and frozen rapidly in the routine manner adopted at the factory; 2 days later the chickens were thawed and the inner surfaces of the birds swabbed with gauze swabs that were subsequently incubated in selenite broth. Freezing was omitted in the latter part of the study because of practical difficulties, and instead the birds were swabbed on the day following chlorination, after storage overnight at room temperature. Trials showed no obvious difference between results obtained by the two swabbing procedures.

Bacteriology

The swabs of the birds were incubated at 37°C. in selenite F broth (Leifson, 1936; Hobbs & Allison, 1945) and subcultured after 24 hr. on to Wilson and Blair's medium and MacConkey agar to which 1:25,000 brilliant green had been added. The plates were examined after incubation at 37°C. for 24 and 48 hr. and colonies thought likely to be salmonellae were examined by the usual serological and biochemical tests.

RESULTS

One hundred and sixteen chicken carcasses that had been infected with salmonellae were treated with concentrations of chlorine between 50 and 200 p.p.m. for periods not exceeding 10 min. The results of examination of swabs taken after various chlorination procedures are presented in Table 1; the effect of chlorination on carcasses inoculated with various numbers of salmonellae is indicated. The minimum concentration of chlorine found to have any regular effect in freeing carcasses from salmonellae was 200 p.p.m. when applied for 10 min., and even this was irregular in its action when more than 1000 organisms were involved. Higher concentrations of chlorine were not used because some observers considered that the flavour of chickens was altered after treatment with more than 200 p.p.m.

Table 1. *The effect of chlorine treatment on chicken carcasses infected with various numbers of salmonellae*

Chlorine treatment		No. of salmonellae inoculated	No. of chickens	
Conc. (p.p.m.)	Time (min.)		Treated	Negative after treatment
200	10	$< 10^3$	19	18
		10^3-10^5	14	9
		$> 10^5$	13	1
200	5	$< 10^3$	5	0
		10^3-10^5	3	0
150	10	$< 10^3$	13	0
		10^3-10^5	7	0
150	7	10^3-10^5	1	1
		$> 10^5$	3	0
100	10	$< 10^3$	13	4
		10^3-10^5	14	1
		$> 10^5$	3	0
100	5	$> 10^5$	6	0
50	5	$> 10^5$	2	0

The results indicated that 200 p.p.m. chlorine, the highest concentration that did not taint carcasses, would, when applied for 10 min. under the conditions described, make it difficult or impossible to recover salmonellae when less than 1000 organisms had been distributed over the inside of a carcass in a faecal suspension. Heavily infected carcasses, such as those harbouring 100,000 or more salmonellae, were not rendered free from salmonellae by any of the chlorination procedures used. Chlorine concentrations of 150 p.p.m. and below had no consistent effect on carcasses infected with even small numbers of salmonellae.

DISCUSSION

Control of salmonellae in broilers may be attempted either before or after the birds reach the factory. Eradication of infection from breeding stock, hygienic handling of eggs (Buxton & Gordon, 1947), elimination of salmonellae from poultry feeding-

stuffs, some of which have been shown to contain the pathogens (Report, 1959), and prophylactic addition of antibacterial drugs to feeds may all help to reduce the number of infected birds that arrive for processing. It is unlikely, however, that these steps will be entirely successful, and each day a few birds carrying salmonellae in the gut, gall bladder or, more rarely, tissues of the body (Wannop, 1960) may enter factories that process up to 30,000 birds daily. Spread of salmonellae could then occur at two stages of the processing. Dixon & Pooley (1961) have recorded the isolation on two occasions of salmonellae from the gloves of persons engaged in evisceration, and so doubtless cross-contamination occurs at this stage. The second point of possible contamination is during the cooling process in water tanks. These are of various types, but Dixon & Pooley were able to demonstrate salmonellae in 21 of 87 samples of water taken from spin-chillers through which water was flowing continuously; birds immersed in such water very probably become infected (Atkinson, 1958).

In the factory premises there are, apart from general hygiene and multiple washing procedures, two specific measures that could be used to reduce the number of salmonellae harboured on carcasses. Dipping the carcasses in antibiotic solutions, such as tetracycline, will reduce the number of salmonellae and many other organisms but is open to the objection that the procedure tends to favour any antibiotic-resistant pathogens that may be present (Barnes, 1957). For this reason it has not been recommended in Britain (Hobbs, Reeves, Garside, Gordon, Barnes, Shrimpton & Anderson, 1960). Chlorination is the other possible specific control measure and has been studied in this work.

It is difficult to determine the number of salmonellae present on a naturally infected carcass and so wide variations in the number of salmonellae inoculated were used in this work. The concentrations of chlorine employed were limited by the risk of tainting the birds. A number of observers could not distinguish between treated and untreated chicken, either before or after cooking, when up to 200 p.p.m. had been used; chickens treated with higher concentrations could be detected by some persons. The duration of the chlorination was limited to 10 min. because longer treatment periods would be difficult to incorporate in routine processing at most poultry packing factories, many of which deal with 1500 or more birds each hour. The degree of chlorination mentioned by Atkinson (1957) as effective in decreasing viable bacterial counts of carcasses was 10–20 p.p.m. for at least 5 min. In the present work, however, considerably higher concentrations were necessary to eliminate even small numbers of viable salmonellae from broiler carcasses. The reduction in numbers of viable salmonellae that occurred with the different chlorination procedures was not studied, as the aim of the experiments was to find a method that would free infected carcasses from salmonellae. It is impossible to assert that salmonellae are absent from a bird unless the whole carcass is homogenized and cultured, but the swabbing technique used for recovery of the pathogens was found to yield a positive result when less than twenty salmonellae had been distributed inside a carcass, and was therefore considered sufficiently sensitive for the purpose.

Salmonellae can survive on the skin of frozen poultry (Galton & Arnstein, 1960),

and it is desirable to prevent infected carcasses from appearing on the market. The main risk of such carcasses is that salmonellae may be spread to other foods, particularly those which are likely to allow bacterial multiplication and are not subsequently cooked before being eaten. There may, however, be some risk from the bird itself, particularly if it be cooked and then left at room temperature before consumption. Hussemann & Wallace (1951), for example, reported that though roasting and broiling would reduce the number of salmonellae present in carcasses that had been infected artificially with large numbers of salmonellae, in no instance was a bird completely freed from infection by such cooking.

Chlorination, to be effective, must be applied at a stage when there is no further possibility of contamination of the birds. The carcasses must therefore be treated after they leave the chilling tanks. A quantitative study of the degree of contamination of chill-tank water with salmonellae is in progress, and from the preliminary results we consider that carcasses becoming infected in such water would be unlikely to harbour more than a thousand salmonellae. It is suggested, therefore, that routine chlorination at 200 p.p.m. for 10 min. after the carcasses have left the spin-chillers would greatly reduce the number of salmonella-infected broiler chickens that leave processing factories. No detailed study was made of the effect of this degree of chlorination on the keeping quality of carcasses, but a small number of observations indicated that treated carcasses remained in good condition for a longer period than untreated birds when stored at room temperature.

SUMMARY

A study was made of the effect of chlorine solutions on broiler chicken carcasses that had been artificially infected with salmonellae. One hundred and sixteen carcasses were infected with widely varying numbers of seven different salmonella serotypes.

Treatment of carcasses with 200 p.p.m. of chlorine for 10 min. usually prevented the subsequent recovery of salmonellae when fewer than 1000 organisms had been inoculated. When larger numbers of organisms were inoculated or when lower concentrations of chlorine were used, salmonellae were usually recovered from the treated carcasses.

It is suggested that the incorporation of chlorination with 200 p.p.m. for 10 min. into the routine processing of poultry carcasses would greatly reduce the number of contaminated birds leaving processing factories.

REFERENCES

Atkinson, J. W. (1957). Trends in poultry hygiene. *Publ. Hlth Rep., Wash.*, **72**, 949.

Atkinson, J. W. (1958). Poultry processing sanitation under official programs. *J. Milk Tech.* **21**, 38.

Barnes, E. (1957). New methods of food preservation. (a) Antibiotics. *R. Soc. Hlth J.* **77**, 446.

Brobst, D., Greenberg, J. & Gezon, H. M. (1958). Salmonellosis in poultry and poultry processing plants in Western Pennsylvania. *J. Amer. vet. med. Ass.* **133**, 435.

Buxton, A. & Gordon, R. F. (1947). The epidemiology and control of *Salmonella thompson* infection of fowls. *J. Hyg., Camb.*, **45**, 265.

Dixon, J. M. S. & Pooley, F. E. (1961). Salmonellae in a poultry-processing plant. *Mon. Bull. Minist. Hlth Lab. Serv.* **20**, 30.

Galton, M. M. & Arnstein, P. (1960). *Poultry Diseases in Public Health—Review for Epidemiologists.* U.S. Publ. Hlth Serv. publ., no. 767, Washington, D.C.

Galton, M. M., Mackel, D. C., Lewis, A. L., Haire, W. C. & Hardy, A. V. (1955). Salmonellosis in poultry and poultry processing plants in Florida. *Amer. J. vet. Res.*, **16**, 132.

Hobbs, B. C. & Allison, V. D. (1945). Studies on the isolation of *Bact. typhosum* and *Bact. paratyphosum B. Mon. Bull. Minist. Hlth Lab. Serv.* **4**, 12.

Hobbs, B. C., Reeves, J. C., Garside, J. S., Gordon, R. F., Barnes, E. M., Shrimpton, D. H. & Anderson, E. S. (1960). Antibiotic treatment of poultry in relation to *Salmonella typhimurium. Mon. Bull. Minist. Hlth Lab. Serv.* **19**, 178.

Hussemann, D. L. & Wallace, M. A. (1951). Studies on the possibility of the transmission of Salmonella by cooked fowl. *Food Res.* **16**, 89.

Leifson, E. (1936). New selenite enrichment media for isolation of typhoid and paratyphoid (Salmonella) bacilli. *Amer. J. Hyg.* **24**, 423.

Mann, T. B. (1961). Letter to editor. *Lancet*, i, 226.

Miles, A. A. & Misra, S. S. (1938). The estimation of the bactericidal power of the blood. *J. Hyg., Camb.*, **38**, 732.

Morris, T. G. & Ayres, J. C. (1960). Incidence of *Salmonellae* on commercially processed poultry. *Poultry Sci.* **39**, 1131.

Pendry, A. R. (1960). The broiler industry: broiler production and processing in Britain. *Sanitarian, Lond.*, **69**, 120.

Report (1959). Salmonella organisms in animal feeding stuffs and fertilisers. *Mon. Bull. Minist. Hlth Lab. Serv.* **18**, 26.

Spink, M. S. (1960). Letter to editor. *Lancet*, ii, 707.

Wadhams, E. H. C. (1960). The broiler industry: an approach to the public health aspect. *Sanitarian, Lond.*, **69**, 128.

Walker, J. H. C. (1960). The broiler industry—transmission of Salmonella infection. *R. Soc. Hlth J.*, **80**, 142.

Wannop, C. C. (1960). The broiler industry: diseases of the broiler chicken of possible public health importance. *Sanitarian, Lond.*, **69**, 124.