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The survival of salmonellas on finger-tips and transfer of the organisms to foods

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SUMMARY

The survival of salmonellas on the finger-tips is considered with reference to the ease with which they can be transferred to food by handling.

Escherichia coli and several *Salmonella* serotypes were shown to survive on the finger-tips for various periods of time, for example, *S. anatum* could be recovered 3 hr. after artificially contaminating them with between 500 and 2000 organisms. *S. anatum* could also be recovered from the finger-tips after contaminating them with more than 6000 organisms followed by a 15 sec. hand-wash 10 min. later. Similarly, the survivors from minimal inocula of less than 100 *S. anatum*/finger-tip were, after 10 min., still capable of infecting samples of corned beef and ham. *E. coli* was isolated from the finger-tips of 13 of 110 butchers soon after they had left the meat line at a meat products factory, but was not detected on the finger-tips of 100 volunteers at the Central Public Health Laboratory.

The implications of the present findings to the spread of salmonellas from raw to cooked foods, and the relevance of this to outbreaks of *Salmonella* infection in the general population and in hospitals, are discussed.

INTRODUCTION

It is not always clear how salmonellas of animal origin reach prepared foods and give rise to infection in man. Raw foods of animal origin, especially meat and poultry, are often contaminated with salmonellas, but outbreaks of infection may follow the consumption of adequately cooked foods. The circumstances of many outbreaks indicate that salmonellas may have been transferred from a raw to a cooked food by means of equipment, working surfaces or by the hands when both foods have been handled by the same person. There is still much emphasis on the spread of salmonellas from human faecal excretors to foods, but hitherto little attention has been paid to the ability of the human hand to transfer these organisms from one food to another. The aim of the present work was, therefore, to study the survival of salmonellas on the finger-tips and the ease with which they could be transferred to food by handling.

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MATERIALS AND METHODS

Volunteers

Preliminary tests to detect the presence of *Escherichia coli* on the finger-tips were made on 100 members of the staff at the Central Public Health Laboratory. These were 50 laboratory and 25 secretarial staff, and 25 persons who washed glassware usually wearing rubber gloves. In a similar experiment, the finger-tips of one hand of 110 butchers at a large meat products factory were sampled about 5 min. after they had left the meat line and before handwashing.

In experiments involving the artificial contamination of finger-tips with *E. coli* or salmonellas, tests were first made on the unwashed finger-tips of one or both of the authors. Similar experiments were then made on volunteers (9 male, 12 female) from the staff at the Central Public Health Laboratory. Volunteers did not use their contaminated hands during the period of each experiment, and after all the finger-tips had been sampled by the elution technique, the volunteers were instructed to wash their hands thoroughly with soap and water for at least 1 min. immediately, and again about 10 min. later.

Bacteria

The cultures used for the artificial contamination of finger-tips were *E. coli* (O-group 88), one culture each of *Salmonella anatum*, *S. derby*, *S. infantis*, *S. meleagridis* and *S. panama* that had been isolated during 1967–1968 from various foods of animal origin and stored on Dorset egg medium, and *S. senftenberg* 775W (N.C.T.C. 9959).

Media

MacConkey agar was used for the isolation of *E. coli* from the finger-tips in both the impression and elution methods described below.

Bismuth sulphite agar (Oxoid) and deoxycholate citrate lactose agar (Hynes, 1942) modified by the addition of 1% sucrose, were used for the isolation of salmonellas from the contaminated finger-tips of volunteers. These media were also used for subcultures from liquid enrichment of corned beef and ham previously touched with contaminated fingers. In these experiments 25 ml. of lactose broth was used as an enrichment medium for *Salmonella* from corned beef, and selenite F medium (Leifson, 1936) modified by replacement of lactose with mannitol and sterilization by Seitz filtration, for recovery of *Salmonella* from ham. Different enrichment media were employed because corned beef is a sterile product and ham is not.

Diluent

The diluent used throughout the work was 0·045 M phosphate buffer containing 0·1% peptone and 0·1% Triton X-100 (a non-ionic detergent, Rohm and Hass Limited), at pH 7·9.

Finger impression plates for Escherichia coli

The finger-tips of one hand were gently pressed on the surface of a MacConkey agar plate for about 2 sec. The plates were incubated at 37° C. for 24 hr. and colonies morphologically resembling *E. coli* were picked for confirmation by biochemical tests.

Contamination of finger-tips with Escherichia coli *and salmonellas*

The tips of the fingers of volunteers were artificially contaminated at various times during the normal working day. Each finger-tip received one drop (0·0067 ml.) of a convenient dilution of an overnight broth culture of the test organism delivered from a no. 20 needle mounted on a syringe; the drop was spread over an area of about 0·5 cm.^{2} The number of organisms in the diluted culture was estimated by the method of Miles & Misra (1938).

Elution of organisms from finger-tips

To elute surviving organisms, the tip of each finger was rubbed against the bottom of a 2 oz. jar containing 1 ml. of diluent and then wiped on the rim to leave as much diluent in the jar as possible. Tenfold dilutions were prepared from 0·5 ml. samples of eluate and 0·02 ml. drops of each dilution plated onto selective media which were incubated at 37° C. for 48 hr. Colonies of *E. coli* or *Salmonella* were confirmed by biochemical or serological tests.

In experiments to determine the presence or absence of salmonellas, 1 ml. of warm (37° C.) double-strength lactose broth was added to the diluent in the sampling jars; the jars were incubated at 37° C. and plated after 24 and 72 hr. on bismuth sulphite and deoxycholate citrate sucrose agars.

Recovery of Escherichia coli *and salmonellas from the finger-tips after artificial contamination*

In a series of tests on different days, various inocula of *E. coli* were applied to the finger-tips of both authors; separate finger-tips were sampled after 2, 5, 10, 15 and 25 min. These experiments were repeated with 19 other volunteers with inocula in the same range, with sampling times of 5, 15, 30, 45 and 60 min.

Similar experiments were carried out with *S. anatum*, first on one of the authors, using a wide range of inocula, and then on eleven other volunteers with inocula ranging from 280 to 3500 organisms per finger-tip. The presence or absence of *S. anatum* in eluates from finger-tips, after contamination with low inocula, was also determined. In these tests various inocula from *ca.* 27 to 1800 were applied to the finger-tips of both hands of one of the authors; separate finger-tips were sampled at intervals up to 180 min.

The survival of different serotypes of *Salmonella* on finger-tips was also studied. One finger-tip on each hand of one of the authors was artificially contaminated with *S. anatum*, *S. derby*, *S. infantis*, *S. meleagridis*, *S. panama*, or *S. senftenberg* (twice). Inocula ranging from 3300 to 9500 organisms per finger-tip were used, and fingers were sampled 15 and 60 min. after contamination.

Effect of hand washing on the removal of salmonellas from contaminated finger-tips

In a series of tests on different days, various inocula of *S. anatum* from 830 to a million were applied to the finger-tips of one person. After 10 min. exposure, the hands were washed with soap and running warm water for 15 sec. in a standard manner and dried with individual paper towels. Each finger-tip was then rubbed against the bottom of an individual jar containing diluent, double-strength lactose broth was added and the jars incubated at 37° C. The experiments were repeated with seven other persons with inocula of *S. anatum* of *ca.* 250/finger-tip.

Artificial contamination of food with salmonellas from the finger-tips

Approximately 5 g. samples of corned beef from a freshly opened can were placed in 2 oz. jars. The finger-tips of 17 persons were artificially contaminated with inocula of *S. anatum* ranging from less than 10 to 6000. After 10 min. exposure, each finger-tip was pressed for about 5 sec. on an individual sample of corned beef. Twenty-five ml. of lactose broth were then added and the jars incubated at 37° C.; subcultures were made after 24 and 72 hr. on bismuth sulphite and deoxycholate citrate sucrose agars. The experiments were repeated on 14 persons using samples of ham; the inocula of *S. anatum* per finger-tip ranged from less than 10 to 330, and selenite F medium was used as the enrichment medium.

RESULTS

Although *E. coli* was not isolated from any of the finger-tips on the sampled hands of 100 of the staff at the Central Public Health Laboratory, it was detected on the finger-tips of the sampled hands of 13 of 110 butchers in the meat products factory.

Table 1 shows the percentage recovery of *E. coli* from the finger-tips after artificial contamination with various inocula. The percentage recovery was somewhat variable, but in general the range and median values obtained in replicate tests on the two authors were similar. After a contact time of 15 min. usually less than 1 % of the initial inoculum was recovered. Yet, in single tests on the finger-tips of nineteen other persons the percentage recoveries of *E. coli* were generally greater and for ten persons the initial inoculum was not reduced to 1 % until 60 min.

Table 2 shows the recovery of *S. anatum* from the finger-tips after artificial contamination with various inocula. In general, the percentage recoveries obtained from nine tests on one of the authors and one test on eleven other persons were similar. The percentage recovery of *S. anatum* fell sharply during the period 0–5 min. and then more slowly during the subsequent 55 min.

Table 3 shows the recovery of *S. anatum* from the finger-tips of one volunteer after contamination with inocula of different sizes. With the larger inocula, *S. anatum* was recovered at least 180 min. later, but with the smaller (less than 100 organisms per finger-tip) this time was reduced to 75 min. or less.

Table 4 shows the percentage recovery of various *Salmonella* serotypes from artificially contaminated finger-tips. The results show that after 15 or 60 min. the percentage recovery rates for *S. anatum*, *S. derby*, *S. infantis*, *S. meleagridis*, and *S. panama* were similar and all were greater than those for *S. senftenberg*.

Table 5 shows the effect of hand washing on the removal of *S. anatum* from contaminated finger-tips. The standardized hand washing procedure used did not

Table 1. *Recovery of* Escherichia coli *from artificially contaminated finger-tips*

No. of persons		1	1	19
No. of tests/person		7	13	1
Initial inoculum per finger-tip ($\times 10^4$)		15–180	3·5–180	32–46
Percentage recovery of *E. coli* after (min.)	2	82·2* (15–100)†	29·7 (11·2–96)	ND
	5	8·2 (0·46–13·6)	2·0 (0·27–9·0)	87·5 (6·2–100)
	10	0·70 (0·20–1·91)	1·1 (0·09–1·91)	ND
	15	0·42 (0·27–0·68)	0·36 (0·07–1·47)	13·8 (0·3–100)
	25	0·31 (0·03–1·05)	0·11 (0·03–0·68)	ND
	30	ND	ND	6·0 (0·02–100)
	45	ND	ND	8·7 (0·02–93·8)
	60	ND	ND	1·1 (0·02–40)

* Median value.
† Range (minimum–maximum).
ND = not done.

Table 2. *Recovery of* Salmonella anatum *from artificially contaminated finger-tips*

No. of persons		1	11
No. of tests/person		9	1
Initial inoculum per finger-tip ($\times 100$)		9·3–3900	2·8–35
Percentage recovery of *S. anatum* after (min.)	5	7·4* (2·36–14·7)†	4·4 (1·08–100)
	15	2·7 (0·43–10·7)	2·8 (1·08–18·3)
	30	1·1 (0·43–11·3)	2·4 (< 0·36–15·4)
	45	0·86 (0·21–8·3)	1·4 (< 0·36–3·9)
	60	0·40 (0·21–4·0)	0·72 (< 0·36–3·9)

* Median value.
† Range (minimum–maximum).

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Table 3. *Recovery of* Salmonella anatum *from the contaminated finger-tips of a single volunteer*

Initial inoculum /finger-tip	Presence (+) or absence (−) of *S. anatum* in cultures from finger-tips after various time intervals (min.)											
	5	10	15	30	45	60	75	90	105	120	150	180
1800	ND	ND	+	+	+	+	+	+	+	+	+	+
530	ND	ND	+	+	+	+	+	+	+	+	+	+
80	+	+	+	+	+	+	+	−	−	−	ND	ND
36	ND	ND	+	−	−	−	−	+	−	−	−	−
27	+	+	+	−	+	−	−	−	−	−	ND	ND

ND = not done.

Table 4. *Recovery of salmonellas from finger-tips after contamination with different serotypes*

Salmonella serotype	Initial inoculum /finger-tip $\times 10^3$	% recovery of salmonellas from finger-tips after two time intervals (min.)	
		15	60
S. anatum	9·5	2·7	0·59
S. derby	3·4	3·3	1·8
S. infantis	3·3	3·3	1·1
S. meleagridis	7·6	3·9	1·7
S. panama	6·5	5·3	3·7
S. senftenberg 775W	6·6	0·24	0·18
	7·5	0·50	0·16

Table 5. *Recovery of* Salmonella anatum *from finger-tips after hand washing*

Initial inoculum/finger-tip	Proportion of finger-tips positive for *S. anatum*
1,000,000	10/10 (one person)
6,400	3/10 (one person)
830	0/10 (one person)
250	0/70 (seven persons)

Table 6. *Recovery of* Salmonella anatum *from corned beef* after contact with contaminated finger-tips*

Inoculum of *S. anatum*/ finger-tip	No. of finger-tips contaminated	No. of corned beef samples +ve for *S. anatum*	% of corned beef samples +ve for *S. anatum*
6000	60	60	100
600	40	40	100
60	40	36	90
7	25	4	16

* Plate count at 35° C. = < 100 organisms/g. *Salmonella* not found in 50 g. − before contamination.

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remove an inoculum of 10^6 *S. anatum*/finger-tip and all ten finger-tips remained contaminated. With inocula between 10^3 and 10^4 some finger-tips remained positive, but with inocula below 10^3 no salmonellas were grown after hand washing. The results from Table 2 indicate that a considerable reduction in numbers of salmonellas could be expected during the 10 min. period between contamination and hand washing.

Table 6 shows the recovery of *S. anatum* from corned beef after contact with contaminated finger-tips. All, or nearly all samples were contaminated from fingers with the three largest inocula, and only with the small inoculum of *ca.* 7 organisms/finger-tip did the positive samples fall to 16%. Similar results are shown with cooked ham in Table 7.

Table 7. *Recovery of* Salmonella anatum *from cooked ham* after contact with contaminated finger-tips*

Inoculum of *S. anatum*/ finger-tip	No. of finger-tips contaminated	No. of ham samples +ve for *S. anatum*	% of ham samples +ve for *S. anatum*
330	40	39	98
30	50	43	86
8	50	7	14

* Plate count at 35° C. = 3×10^5 organisms/g. *Salmonella* not found in 50 g. – before contamination.

DISCUSSION

The survival of bacteria on human skin depends on factors such as humidity, pH, the presence of antibacterial substances in the skin secretions and the presence of competitive organisms. In general, Gram-negative bacilli are more susceptible to desiccation than are Gram-positive cocci and most of them do not survive long on the skin (Ricketts, Squire & Topley, 1951; McDade & Hall, 1964). Payne (1949), for example, showed that the death rate of *E. coli* on skin depended on the rate of drying and was at a maximum between relative humidities of 40 and 50%. Coliform bacilli are not commonly found on normal skin although few workers have made an extensive or careful search for them. Williams & Miles (1949) reported the isolation of coliforms from 18 (3·6%) hands from 500 normal individuals. In contrast, Horwood & Minch (1951) using a hand-rinse technique reported that coliform bacilli were present on the hands of 22 (65%) of 34 food handlers; *E. coli* was found on 12 (38%) of the hands tested. In the present study *E. coli* was not isolated from the finger-tips of 100 of the staff at the Central Public Health Laboratory, but the organism was isolated from the finger-tips of 13 (11·8%) of the 110 butchers tested soon after leaving duty. The difference in the isolation rates of *E. coli* is probably attributable to the fact that the butchers' hands were continuously exposed to contamination from the meat they were handling. The isolation rate from the butchers would probably have been much higher if a finger-rinse technique had been used instead of the finger impression technique and if sampling had been done immediately after the butchers had left the meat line. We are unaware of any surveys on the frequency of isolation of

salmonellas from the hands, but as meat and poultry are important sources of these organisms occasions will arise when the finger-tips of butchers and food handlers are contaminated. Tables 6 and 7 show that cooked foods are very vulnerable if they are touched by fingers that have been contaminated by low numbers of salmonellas. Furthermore, any cooking process will have reduced the numbers of competitive organisms and thus the opportunity for subsequent multiplication of salmonellas is probably provided.

In the experiments to study the survival of *E. coli* and *S. anatum* on contaminated finger-tips there was a sharp fall in the percentage recovery of organisms in the 0–15 min. period after contamination. It was during this period, and usually within 5 min. that the inocula dried on the finger-tips. In replicate tests on the finger-tips of both authors the percentage recoveries of *E. coli* for various inocula were similar with respect to the range and median values obtained (Table 1). Similar tests, on the finger-tips of nineteen other persons gave different results; the percentage recovery rates and median values were significantly greater. Nevertheless, the numbers of organisms had fallen sharply 1 hr. after contamination. For *S. anatum* there was good agreement in the percentage isolation from finger-tips obtained in nine replicate tests on one of the authors and single tests on eleven other persons (Table 2). The results in Table 3 show that *S. anatum* could be isolated from the finger-tips for at least 3 hr. after contamination with an inoculum of about 500 organisms. It is also evident (Table 4) that there is no appreciable difference in the survival rates on skin of five of the six *Salmonella* serotypes studied.

Cooke *et al.* (1970) have reported that *E. coli* was present in 78 of 873 samples of food served to hospital patients. They suggested that many of the strains of *E. coli* had entered the hospital kitchen on raw meat and poultry and other foodstuffs, and had subsequently contaminated other raw foods and also cooked foods. We believe that salmonellas and *E. coli* can be and are transferred on many occasions by the hands from raw to raw and from raw to cooked or processed foods. Our results (Table 5) show that hand-washing with soap and water, followed by drying with paper towels, reduces the risk of transient skin carriage of salmonellas unless the initial contamination is very heavy. Much has been done in the past to encourage food handlers to wash their hands after visiting the w.c. A similar effort to encourage all food handlers to wash their hands after touching raw foods of animal origin is equally essential.

The results may also have some bearing on the mode of spread of *Salmonella* infection in hospitals. Short explosive outbreaks of foodborne *Salmonella* infection occasionally afflict hospitals, but more often the outbreaks run a protracted course over a variable period of time and therefore are less likely to have involved the continuous contamination of food and subsequent growth in it (Taylor, 1963; Williams, Blowers, Garrod & Shooter, 1966). However, even if multiplication in foods is not necessary, the salmonellas probably enter their new host by the mouth. The routes by which salmonellas travel from one patient to another have not been extensively studied, but these routes probably include the transient contamination of the hands of staff or patients (Mackerras and Mackerras, 1949; Watt *et al.* 1958),

and environmental contamination, e.g. fomites, dust and surfaces, which can act as secondary reservoirs (Rubbo, 1948; Parker, 1954; Bate & James, 1958; Rowe, Giles & Brown, 1969).

The ability of salmonellas to survive under various environmental conditions will affect their spread. Our results show that salmonellas can survive on the finger-tips for several hours and that during this time the hands can transmit infection.

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