Salmonellas of subgenus III (Arizona) isolated from abattoirs in England and Wales

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Salmonellas that belong to subgenus III, often termed Arizona strains, have attracted little attention in the United Kingdom, although these organisms can sometimes produce severe and fatal infections in man. The most likely reason for this is the rarity of isolation of these organisms.

A human infection with a subgenus III strain has, however, been recorded in Europe under conditions in which there was little doubt as to the pathogenicity of the organism (Edwards, Kauffmann & Stucki, 1960). This infection occurred in a man returning from holiday in the Balearic Islands.

Late-lactose-fermenting strains of subgenus III may easily be mistaken for salmonellas of sub-genus I. Antigenic overlapping between subgenus III and subgenus I is common with both O and H antigens. The H antigens may be diphasic or monophasic and there is a tendency to regard diphasic varieties as relatively non-pathogenic to man. Rapid lactose fermentation is often recorded in diphasic strains.

During an investigation into the salmonella content of Indian crushed bone several subgenus III strains were isolated (Harvey & Price, 1962). All were diphasic and required 40–64 hr. to ferment 1% lactose peptone water. On brilliant green MacConkey agar (Harvey, 1956), however, some serotypes produced apparently non-lactose fermenting colonies, some were frank lactose fermenters and some produced a mixture of both lactose-fermenting and non-lactose-fermenting colonies, which were sharply differentiated one from the other. One serotype found had previously been encountered in Indian monkeys imported into Germany, Canada and America (P. R. Edwards, personal communication).

Strains of subgenus III were noticeably absent from a recently published list of salmonellas isolated from human, animal and other sources and identified in Great Britain between 1951 and 1963 (Taylor *et al.* 1965). The few isolations recorded here may therefore be of interest.

MATERIALS

The observations on Indian bone, at one time an ingredient of animal feeding stuffs distributed in South Wales, prompted us to examine native animal material. Because the incidence of subgenus III salmonellas was expected to be very low, a

pooling procedure had to be used (Harvey, 1957; Newell, 1959). Four abattoirs were examined using gauze swabs placed in drains receiving material from cattle, pig and sheep slaughter. These swabs were selectively cultured with a view to isolating salmonellas (Moore, 1948; Harvey & Phillips, 1961). Two of the abattoirs (Cardiff and Barry) were in South Wales and two were in England (Beccles I and Beccles II). Some details of the proportions of animals killed in these slaughter houses have already been published (Report, 1964).

METHODS

The techniques used in both laboratories for the isolation of subgenus III strains of salmonellas were very similar. Selenite F broth was used as enrichment medium for culturing the Moore's swabs. Enrichment broths were incubated at 37 or 43° C. or sometimes at both temperatures (Harvey & Thomson, 1953). Subcultures were made at various times from 24–72 hr. on de Loureiro's (1942) modification of Wilson and Blair's medium, on which strains of subgenus III formed characteristic black colonies with surrounding sheen. Other selective plating media were occasionally used successfully, but Wilson and Blair's medium was found to be the most satisfactory. Plates were examined after incubation at 37° C. for 24 and 48 hr. Suspicious colonies were picked and examined for the biochemical properties of members of the genus *Salmonella*. The antigenic structure of the strains isolated was kindly determined by Dr P. R. Edwards and by Dr Joan Taylor.

Table 1. *Details of abattoirs observed*

Abattoir	County	Animals killed in month of isolation of strain			Year and month of isolation	Total period of observation
		Pigs	Cattle	Sheep		
Cardiff	Glamorgan	2386	836	2757	1965 Apr.	1957–65
Barry	Glamorgan	323	276	1877	1961 Jan.	1960–65
Beccles I	Suffolk	82	684	279	1962 Feb.	1961–62
Beccles II	Suffolk	65	27	17	1962 Feb.	1961–62

Table 2. *Isolations of subgenus I and subgenus III from abattoirs*

Abattoir	Total swabs examined	Swabs positive for subgenus I	Isolations of subgenus III		Serotype of subgenus III
			Swabs positive	Class of animal sampled	
Cardiff	880	347 (39)	3 (0.3)	Cattle + sheep	26:29–30
Barry	434	122 (28)	1 (0.2)	Cattle + pigs + sheep	26:29–30
Beccles I	150	110 (73)	1 (0.7)	Cattle + pigs + sheep	26:30
Beccles II	177	34 (22)	1 (0.6)	Cattle + pigs + sheep	26:30
Total	1641	613 (37)	6 (0.4)		

Figures in brackets are percentages.

RESULTS

Details of the four abattoirs from which subgenus III strains were isolated are given in Table 1.

Details of the isolation of subgenus III strains are given in Table 2. Four of the strains were diphasic and two were monophasic.

No strains of subgenus II were encountered in these four abattoirs. The six swabs, from which salmonellas of subgenus III were cultured, sampled drains receiving material from the slaughter of cattle, pigs and sheep, or of *cattle and sheep only*.

DISCUSSION

It must be emphasized that very few isolations of subgenus III salmonellae were made. In a period of 9 years in which 1641 abattoir swabs were examined only six were found positive. The only point common to the six positive samples was that the drains concerned received material from cattle and from sheep. Cattle are presumed to be one of the main sources of salmonella in British abattoirs. Sheep are regarded as being relatively infrequently infected (Report, 1964). It was, therefore, natural to think that the subgenus III salmonellas might have a bovine origin. Reference to the Communicable Diseases Centre, Atlanta, Georgia, however, showed that serotype 26:29-30 (isolated four times) was most commonly found in sheep. Of thirty-five strains identified by the Communicable Diseases Centre to 30 June 1965, twenty-three came from sheep and four from other animals. It was interesting to note that on three occasions serotype 26:29-30 was cultured from man and, in two of these incidents, those infected belonged to sheep-herding tribes of American Indians (W. H. Ewing, personal communication). No useful information was available on the monophasic serotype.

Culturally all the abattoir strains of subgenus III appeared as non-lactose fermenting colonies when plated on lactose-containing selective media. In fluid media (1% lactose peptone water), however, the diphasic strains fermented lactose in 48-52 hr, while the monophasic varieties failed to ferment lactose in 7 days. We confirm that the most useful differential medium for subgenus III salmonellas is bismuth sulphite agar, on which, in our experience, the colonial appearance is consistent.

Lastly, this investigation demonstrates the sensitivity of the Moore's swab as an instrument of survey. It is very probable that without this technique the presence of subgenus III salmonellas in native abattoirs could not have been demonstrated.

SUMMARY

Using Moore's swabs in four abattoirs, six strains of subgenus III salmonellas were isolated—four diphasic strains belonging to one serotype and two monophasic strains belonging to another. The serotypes differed in the length of time needed to ferment lactose. There was some evidence that the diphasic serotype could have had an ovine origin.

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