***Salmonella dublin* infection in adult cattle: a slaughterhouse and knackery survey in South West Wales**

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SUMMARY

A survey was carried out between June 1970 and December 1971 in which gall bladders from cattle either slaughtered for food consumption or disposed of through knackeries were cultured for salmonellas. *Salmonella dublin* was isolated from 28 of 1617 gall-bladders of adult cattle killed in the slaughterhouse at Carmarthen, and from 23 of 197 gall-bladders obtained from adult cattle sent to a knackery at Abergwili, near Carmarthen. The results are compared with a similar survey carried out in 1947 and 1948.

Between June and November 1971 gall-bladders and uteri were also obtained from a slaughterhouse in Newcastle Emlyn, North Carmarthenshire, and a knackery in Tanygroes, South Cardiganshire. *S. dublin* was cultured from two gall-bladders but from none of the uteri of the 161 slaughterhouse cases. Seven of the 46 cows from the knackery yielded *S. dublin* on culture: 2 in the gall-bladder only, 3 in the uterus only and 2 in both gall bladder and uterus.

INTRODUCTION

In 1947 and 1948 Field (1949) performed a survey in which he cultured the gall-bladders of cattle, disposed of through nine knackeries in Wales, for *Salmonella*. The investigation was carried out during the months June to November in each of the two years. In an earlier investigation into bovine salmonellosis Field (1948) had found that the majority of cattle died when affected with clinical salmonellosis so that his isolation rate of salmonella from cattle carcasses in knackeries gave an indication of the incidence of clinical salmonellosis in the area served by the knackery.

He recovered salmonellas from 58 of 414 (14 %) gall-bladders obtained from the knackery at Abergwili, Carmarthenshire and from 9 of 205 (4.4%) gallbladders from that at Tanygroes (The counties referred to were those in existence before the reorganisation of the county boundaries in 1974). The majority of the strains were *S. dublin*. Nineteen of the 176 salmonellas isolated in the survey were four other serotypes although no information was given on their distribution.

This report gives the results of a survey carried out in South West Wales between June 1970 and December 1971 and includes samples obtained from the knackeries at Abergwili and Tanygroes.

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MATERIAL AND METHODS

Gall-bladder survey

Between June 1970 and December 1971 gall-bladders from adult cattle slaughtered by one wholesale butcher were collected once or twice a week from the slaughterhouse in Carmarthen, usually on the day after the animals were killed, and also at less frequent intervals from a knackery at Abergwili, near Carmarthen. In all, a total of 1716 gall-bladders in 131 batches were examined from the slaughterhouse and 197 gall-bladders in 55 batches from the knackery. In neither instance was any reference made to the breed, age or sex of the beast of origin.

At the laboratory the gall-bladders were incised with sterile instruments and 5 ml of bile withdrawn in a sterile pipette and added to 5 ml of double-strength selenite F broth (Oxoid). One subculture was made after overnight incubation on deoxycholate citrate agar (Hynes modification) (D.C.A.) (Oxoid) and also on MacConkey medium (Oxoid) containing 1/25,000 brilliant green (B.G.M.) during most of the survey period. All cultures were incubated at 37° C in air.

The salmonellas were identified by the slide agglutination test using group specific antisera. Many of the strains were sent to W. J. Sojka, Central Veterinary Laboratory, Weybridge, for confirmatory tests.

Uterus and gall-bladder survey

This was carried out as part of an investigation of *S. dublin* abortion in cattle and covered the period June to November 1971, a time of year when this cause of abortion is most prevalent (Hinton, 1975). The gall-bladder and uterus of individual cows were examined. A total of 161 sets of specimens were obtained from cows killed at a slaughterhouse in Newcastle Emlyn, Carmarthenshire, and 46 sets from a knackery at Tanygroes, Cardiganshire. The slaughterhouse specimens were collected on the day after slaughter and the knackery specimens, which were stored in a cold-room, were picked up once a week. Three additional specimens were also examined from the Abergwili knackery.

The gall-bladders were cultured as already described: the surface of the uterus was seared with a hot iron, incised with a sterile scalpel and a swab taken of the mucosal surface. This was then placed in 10 ml of single-strength selenite F broth and subcultured in the same way as the bile samples.

The validity of the survey technique

The unsocial hours of slaughtering cattle and the sporadic delivery of carcasses to knackeries meant that trained personnel could not be made available to collect samples as soon as the abdomen was opened. As a consequence, the specimens had to be collected and stored in batches and not individually, and this meant that there was a risk of cross-contamination occurring between evisceration and the examination at the laboratory.

In five batches from the Carmarthen slaughterhouse 50 of 99 gall-bladders yielded *S. dublin* on culture. This was considered to represent severe cross-contamination and consequently these batches have been excluded from the results,

Table 1. *Isolations of Salmonella dublin from bovine gall-bladders obtained from the Carmarthen slaughterhouse*

	Month											
	J	F	M	A	M	J	J	A	S	O	N	D
1970												
No. of batches	—	—	—	—	—	6	7*	6	2*	6*	8	8
No. of gall bladders examined	—	—	—	—	—	105	89	61	29	35	108	108
No. of positive <i>S. Dublin</i>	—	—	—	—	—	0	6	0	1	0	2	0
1971												
No. of batches	8	8	6	8	7	6	6*	7	7	7	7	6*
No. of gall bladders examined	108	101	110	146	71	55	77	54	99	53	154	54
No. of positive <i>S. dublin</i>	0	0	1	2	0	0	0	0	6	2	6	2

* One batch examined in each of these months has been excluded as there was evidence of gross contamination.

which are consequently based on the examination of 1617 gall-bladders in 126 batches.

Some cross-contamination probably occurred occasionally in some of the other batches in which more than one of the specimens was positive. However, it was considered that this was not a serious problem and could be discounted as its effect on the general conclusions would have been negligible.

RESULTS

Gall-bladder survey

The results are listed in Tables 1 and 2 and are summarized in Table 3, together with some additional data from other diagnostic specimens for comparison. In the survey period of 19 months *S. dublin* was isolated from 28 of 1617 (1.7%) bladders obtained from the slaughterhouse and 23 of 197 (11.7%) bladders from the knackery. In 1970 and 1971 the proportion of gall-bladders yielding *S. dublin* during the months June to November (the same period as Field's survey) were 2.1% and 2.9% for the Carmarthen slaughterhouse and 3.7% and 23% for the Abergwili knackery. The difference in isolation rate between the two years was significant for the knackery ($P = < 0.01$).

When the 2 years are combined there were significantly more ($P = < 0.001$) positive gall-bladders in the knackery samples than in those from the slaughterhouse.

During 1971 additional gall-bladders were examined from the slaughterhouse at Newcastle Emlyn and the knackery at Tanygroes (see Table 4) and consequently a comparison between all four sources can be made for that year. There were no significant differences in the isolation rate in either the two slaughterhouses or the two knackeries but as expected when the two slaughterhouses and

Table 2. *Isolations of Salmonella dublin from bovine gall-bladders obtained from the Abergwili knackery, June 1970 to December 1971*

	Month											
	J	F	M	A	M	J	J	A	S	O	N	D
1970												
No. of batches	—	—	—	—	—	1	2	3	3	3	4	3
No. of gall bladders examined	—	—	—	—	—	4	5	7	9	15	14	15
No. of positive <i>S. dublin</i>	—	—	—	—	—	0	0	0	2	0	0	0
1971												
No. of batches	3	4	3	3	3	3	4	3	3	3	2	2
No. of gall bladders examined	13	18	12	11	10	7	10	8	11	13	8	7
No. of positive <i>S. dublin</i>	0	2	2	0	0	0	3	2	4	4	0	4

Table 3. *A summary of isolations of Salmonella dublin from various bovine sources between June and November in 1970 and 1971*

	Proportion of samples (%) positive for <i>S. dublin</i>		
	June–Nov. 1970	June–Nov. 1971	June–Nov. 1970/71
Gall-bladders (Carmarthen slaughterhouse)	2.1	2.85	2.5
Gall-bladders (Abergwili knackery)	4	22.8	13.5
Adult faeces swabs (Carmarthen V.I. Centre)*	35.9	18.3	27.3
Placentas and fetuses (Carmarthen V.I. Centre)*	11.3	6.3	8.3

* Carmarthen V.I. Centre (unpublished data).

the two knackeries together are compared the isolation rates were significantly higher in the latter ($P = < 0.001$).

A comparison between the combined results from the Abergwili knackery in June to November 1970 and 1971 and those of Field in 1974 and 1948 during the same months showed that the total isolation rates were similar (13.5% and 14%). However if the years are considered separately there were significantly fewer ($P = < 0.05$) isolations in 1971 than in 1947 and 1948, though in 1972 the difference was not significant.

With regard to Tanygroes it was only possible to compare the findings in 1971 with those of Field and in this year the difference was not significant.

Uterus and gall-bladder survey

The results of examining uteri and gall-bladders from the Newcastle Emlyn slaughterhouse and the two knackeries are listed in Table 4. *S. dublin* was not

Table 4. *Isolation of Salmonella dublin from the uterus and gall-bladder of adult cattle killed in the Newcastle Emlyn slaughterhouse or sent to the knackeries at Tanygroes and Abergwili, June to November 1971*

	Source of material			Slaughter-house Newcastle Emlyn
	Knackery			
	Tanygroes	Abergwili	Total	
No. of cattle examined	46	3	49	161
No. of batches	13	3	14	16
No. of cattle pregnant (%)	8	1	9 (18.5%)	44 (27.5%)
Isolation of <i>S. dublin</i>				
Gall-bladder + ve uterus + ve	2	1	3	0
Gall-bladder + ve uterus - ve	2	0	2	2
Gall-bladder - ve uterus + ve	3	1	4	0
Proportion of cows with positive gall-bladders (%)	8.5	33	10	1.24

isolated from the gall-bladder, uterus or fetal stomach of any of the 53 pregnant cows, although it was isolated from several of the knackery cases, namely the gall-bladder and uterus in three, the gall-bladder only in two and the uterus only in four.

DISCUSSION

S. dublin was the only salmonella serotype isolated in this survey. However, this is not surprising as this serotype accounted for over 95% of the salmonellas isolated from diagnostic material submitted to this laboratory from cattle in the former counties of Breconshire, Carmarthenshire and Pembrokeshire during 1970 and 1971 (Carmarthen V.I. Centre, unpublished data).

Inspection of the extensive data on the incidence of salmonellosis in cattle in England and Wales provided by Sojka & Field (1970), Hughes *et al.* (1971) and Sojka, Wray, Hudson & Benson (1975) reveals that during the 1960s there was a dramatic increase in the number of incidents in which *S. dublin* was isolated from adult cattle. This reached a peak in 1969 and began to fall thereafter to the 1968 figure by 1973.

The isolation rate from gall bladders obtained from the knackeries in 1970/1 was shown to be similar to that in 1947/8. However, the similarity in these findings is probably coincidental in that the effects of the increase in incidence of clinical salmonellosis will have to be set against the considerable reduction in mortality rate but not in the convalescent carrier rate brought about by more effective therapy (Hughes *et al.* 1971).

Cows recovering from *S. dublin* dysentery usually remain life-long excretors of the organism in their faeces (Field, 1948) and consequently the improved recovery rate in clinical salmonellosis will result in an increase in the number of carrier cows in the population. It was the purpose of the slaughterhouse survey to make an estimate of this incidence.

At present there are no absolute criteria available for defining a carrier and these may exist as either constant or intermittent faecal excretors or as latently infected animals. When the survey was planned a consideration of the reports of Field (1948, 1949) suggested that the examination of gall-bladders of clinically normal cows would give a reasonable estimate of the incidence of carrier cows. However, papers subsequently published by Watson, Wood & Richardson (1971) and McCaughey, McClelland & Hanna (1971) indicate that the culture of gall-bladders alone will only identify half the carriers so in order to obtain a realistic figure it is necessary to double the numbers recorded in our survey. Nevertheless the incidence in S.W. Wales still appears low compared with the situation in Northern Ireland, where Murdock & Gordon (1953) and McCaughey *et al.* (1971) record an incidence of 7-8 %.

The examination of the uteri and gall-bladders formed part of a detailed study of *S. dublin* abortion in cattle (Hinton, 1974). As abortion is frequently the only clinical sign and the mortality rate is very low the affected animal will rarely be sent to the knackery for disposal. On the other hand, abortion is a common sequel to *S. dublin* dysentery in the pregnant cow (Field, 1948) and consequently the uterine infections noted in the knackery specimens are probably fatal infections of this type. The fact that the gall-bladder was only positive in five of the nine infected cows identified in the knackeries suggests that the examination of the gall bladder alone is also unsatisfactory for identifying cows dying of clinical salmonellosis and this means that Field's (1949) estimate of mortality rate was probably too low.

The main conclusions to be drawn from this survey are, firstly, that despite the change in the clinical situation between 1947/8 and 1970/1 the introduction of modern therapeutic measures has meant that the proportion of cattle infected with *S. dublin* entering knackeries has remained much the same, while, secondly, in spite of the inevitable increase in the number of carriers in the healthy cattle population that probably has occurred in recent years the proportion of carriers remains at a relatively low level even though S.W. Wales is an area in which *S. dublin* infection has been endemic for many years.

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