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Salmonella in Sheep in Iceland

By *S. Hjartardóttir*[1], *E. Gunnarsson*[1], and *J. Sigvaldadóttir*[2]

[1]Institute for Experimental Pathology, University of Iceland, Keldur and [2]Chief Veterinary Office, Iceland.

Hjartardóttir S, Gunnarsson E, Sigvaldadóttir J: Salmonella in sheep in Iceland. Acta vet. scand. 2002, 43-48. – In 1995 several outbreaks of food poisoning in humans occurred in Iceland, that were traced to salmonella contamination of singed sheep heads. This prompted us to study the prevalence of salmonella infection in sheep and to trace where and how infection might have occurred. Faecal, intestinal contents and tonsillar samples were collected in the spring and autumn from sheep on 50 farms in the southwestern part of the country, where salmonellosis had been detected and from 5 farms in the northwestern part of the country. All faecal samples from the southwest were negative, whereas samples from 3 farms obtained in the autumn in the northwest were positive. Tonsillae taken in the autumn were positive in sheep from 3 farms in the southwest and 2 in the northwest. Our results show that salmonella infection is rare in Icelandic sheep but healthy carriers may harbour the bacteria in tonsillae. Salmonella was not detected in drainage from slaughterhouses nor in singed sheep heads.

faeces; food poisoning; gull; raven; salmonella; sheep; tonsillae.

Introduction

Infection by salmonella is a common cause of food poisoning in humans (*Hobbs et al.* 1989). The genus Salmonella comprises approximately 2,200 serotypes and although infection by *Salmonella typhimurium* is the most common cause of food poisoning in Europe, America and Australia, *S. enteritidis* has exceeded those due to *S. typhimurium* in Britain (*Gilbert et al.* 1990). The severity of symptoms varies with the infecting serotype. Most patients recover after approximately 5 days but more serious illness can occur, especially in children and elderly people (*Prescott et al.* 1996).

Food-animals such as intensively reared pigs, poultry and calves are probably the major reservoir of salmonella infection in humans and outbreaks of food poisoning caused by salmonella are most frequently associated with consumption of products from these species. Contamination of the products can often be traced to slaughtering and processing. Salmonella infection of sheep and food poisoning through consumption of sheep products is apparently rare. However, 3 years ago several outbreaks of food poisoning in Iceland caused by salmonella could be traced to consumption of singed sheep heads, a speciality of the country. This caused considerable alarm as the Icelandic sheep is considered to be a wholesome product, living on the clean highland crop. A limited survey of salmonella infection of singed sheep heads from several slaughterhouses was undertaken 1995 and salmonella could be isolated from approximately 20% of the specimens studied. The serotypes found were *S. thompson, S. oranienburg* and *S. montevideo* (unpublished results).

To obtain more information of the prevalence of salmonella infection of sheep a more extensive study was initiated. In addition to obtaining better data on the prevalence of salmonella infection of sheep we were also interested in tracing the origin of the infection, i.e. whether the

singed sheep heads got contaminated in the slaughterhouses during the processing or if the sheep were infected in the field and carried the salmonella in tonsillae. The food poisoning could be explained by insufficient boiling of singed sheep heads contaminated by salmonella in tonsillae.

Materials and methods

Sampling

In 1996 a screening for salmonella infection was done in sheep from 56 farms, 50 farms from 4 districts in the southwest and 6 farms from a single district in the northwest (Fig.1). Samples were collected from sheep in late pregnancy in spring before they were driven to pastures and at slaughter in the autumn.

Faeces: In the spring, faecal samples were obtained from approximately 50 pregnant ewes from each of 50 farms in the southwest, 2,592 ewes altogether. Samples from 6-8 ewes from the same farm were mixed into a single sample of 10 grams, making a total of 437 cultures.

In the autumn, intestinal samples were collected at slaughter from lambs from most of the same 50 farms in the southwest, 38 out of 50, 1,372 samples altogether and from 60 lambs from 5 farms in the northwest, 1,432 lambs altogether. The samples were taken from caecum content with aseptic precautions. Samples from 6 lambs from the same farm were thoroughly mixed and a spoonful put into a sterile plastic container, making a total of 437 cultures.

Heads: Samples were taken from lambs at slaughter in the autumn. In general, samples were taken from 2 heads from each farm, one singed and one untreated. Seventy two tonsillar cultures were made from untreated sheep heads from 39 farms. Twenty four tonsillar cultures were made from singed sheep heads from 12

Figure 1. Icelandic districts.

farms. For bacterial culture from untreated heads, tonsillae were removed aseptically but singed heads were washed in Buffered Pepton Water and a sample of water taken for culture.

Washwater: Salmonella contamination in 2 slaughterhouses was studied by putting tampons in drainage pipes in the morning and removing them in the evening. This was done 4 times during the slaughtering period lasting from September to October. The samples were taken from drainage pipes where the sheep are eviscerated. Seventy two tonsillar cultures were made from untreated sheep heads from 39 farms. Twenty four tonsillar cultures were made from singed sheep heads from 12 farms.

Culture methods

Pre-enrichment: Pre-enrichment was performed in a non-selective medium (Buffered Peptone Water, Oxoid CM 509) at 37 °C for 18-24 h. A faecal sample of 10 grams was pre-enriched in 100 ml of buffered peptone water. Singed heads were placed in a plastic bag and washed in 250 ml of buffered peptone water. A sample of 100 ml was pre-incubated. Tonsils from 2 lambs from the same farm were minced and incubated together in 100 ml of buffered peptone water. The tampons from slaughterhouses were incubated separately, each in 200 ml of buffered peptone water.

Enrichment: A 0,1 ml of pre-enriched broth was transfered to 10 ml of RV selective broth (Rappaport-Vassiliatis Enrichment Broth, Oxoid CM 669) which had been pre-warmed to the incubation temperature of 42 °C.

Spreading on agar plates: 10 μl of the enrichment broth was spread on the surface of a BG plate (Brilliant green, Oxoid CM 263). The brilliant green agar contained sulphadiazinum, 1 ml of 0.8% solution in 100 ml of agar (Sulphadiazinum, Norsk Medisinaldepot 31 94 91). Plating was made from the enrichment broth after 24 h and 48 h of incubation. The BG plates were incubated inverted at 37 °C for 18-24 h.

Table 1. Screening for *Salmonella* in faeces of pregnant ewes in spring in SW Iceland.

District	Farms	Samples	Cultures	Positive cultures
Rangárvallasýsla	18	984	165	0
Árnessýsla	19	999	167	0
Mýra- and Borgfj.s.	11	535	92	0
Kjósarsýsla	2	74	13	0
Total	50	2,592	437	0

Confirmation: Suspected salmonella colonies were picked from the BG agar and tested with biochemical and serological methods. Biochemical tests used for identification of Salmonella were TSI (Triple Sugar Iron agar, Difco 0265-01-9), LIA (Lysine Iron Agar, Difco 9849-01) and urea agartest (Urea agar base, Difco 0283-01-7). Questionable colonies were studied further by API-tests (API 20 E, bio Mérieux 20 100).
Serotyping was done at the Department of Bacteriology of the National Hospital, Reykjavík.

Results

Faecal samples from spring

The results of 437 cultures for salmonella from faecal samples from 2,592 pregnant ewes collected in spring are shown in Table 1. All cultures were negative.

Intestinal samples from autumn

The results of cultures from intestinal samples from 1,432 sheep are shown in Table 2. All samples from 38 of the 50 farms studied in spring were negative, whereas *S. montevideo* was isolated from sheep in 3 out of 5 farms in

Table 2. Screening for *Salmonella* in caecum of lambs at slaughter.

District	Farms	Samples	Cultures	Positive cultures	Positive cultures (%)
SW					
Rangárvallasýsla	14	632	104	0	0
Árnessýsla	13	362	62	0	0
Mýra- and Borgarfj.s.	10	348	58	0	0
Kjósarsýsla	1	30	5	0	0
NW					
V-Barðastrandasýsla	5	60	10	4	40
Total	43	1,432	229	4	2

the northwest (V-Barðastrandasýsla).

Sheep heads

Salmonella was isolated from 6 out of 72 tonsillar cultures from untreated sheep heads (Table 3). The highest frequency of positive cultures were found in samples from Barðastrandasýsla in the northwest, where *S. montevideo* was isolated from sheep from 2 out of 6 farms. S. thompson was isolated from 1 out of 6 farms in Mýra- and Borgarfjarðarsýsla and 2 out of 16 in Árnessýsla, a single sample from each farm. Cultures from singed sheep heads were all negative (Table 4).

Drainage from slaughterhouses

Cultures obtained from 2 slaughterhouses were negative (data not shown).

Discussion

Our findings of salmonella infection in sheep, support and extend the results of our earlier pilot study. The prevalence of the infection was low, i.e. 1.3% of attempted cultures. There are some sporadic reports of the occurrence of salmonella infection in sheep (*Coulson et al.* 1983, *Gitter et al.* 1970, *Brogden et al.* 1994, *Norris et al.* 1989a,b,c, and 1990) but a comparable survey has apparently not been done in other countries. There was, however, a marked difference in the prevalence of the infection in different areas. Thus the prevalence was much higher in the northwestern part of the country. In autumn percentange of positive cultures was

Table 3. Screening for *Salmonella* in tonsils of lambs at slaughter.

District	Farms	Samples	Cultures	Positive cultures	Positive cultures (%)
SW					
Rangárvallasýsla	13	30	16	0	0
Árnessýsla	17	362	28	2	7
Mýra- and Borgarfj.s.	3	348	15	1	7
Kjósarsýsla	0	30	0	0	0
NW					
V-Barðastrandasýsla	6	60	13	3	23
Total	39	1,432	72	6	8

Table 4. Screening for *Salmonella* in lambs at slaughter, singed sheep heads.

District	Farms	Cultures	Positive cultures
SW			
Rangárvallasýsla	2	4	0
Árnessýsla	1	2	0
Mýra- and Borgarfjarðarsýslur	8	16	0
Kjósarsýsla	1	2	0
NW			
V-Barðastrandasýsla	0	0	0
Total	12	24	0

30.4% in the nothwest but but only 1.1% in the southwest. This difference may be due to smaller, more densely populated pastures in the northwest.

Stress is known to lead to activation of latent salmonella infection and shedding in faeces (*Norris et al.* 1989a,b,c, 1990). In our study faecal samples were taken under stressful conditions, i.e. in the spring from ewes in late pregnancy and in the autumn after driving the sheep from mountain-pastures, which supports that the ewes are not infected before they are driven to the mountain pastures but get infected there. The higher prevalence of salmonella infection in the northwest can be explained by smaller pastures which are more densely populated by grazing sheep, which would favour spreading of the infection. Another factor that might contribute to the higher prevalence of salmonella in the northwest is the existence of wild birds, especially gulls and ravens, that may lead to contamination of pastures. *Salmonella spp.* have been found in approximately 5% of gulls in Iceland (unpublished results) which is similar to that reported in other countries (*Bø* 1980, *Fenlon* 1981, *Butterfield et al.* 1983, *Coulson et al.* 1983, *Girdwood et al.* 1985). A much higher frequency, i.e. approximately 67%, was found in gulls and ravens in a small community in the south where a serious epidemic of salmonellosis in horses was traced to contamination of drinking water (unpublished results). The number of gulls, e.g. *Larus marinus* and *L. fuscus,* has been increasing in recent years and they are more frequently found far inland on mountain pastures (*Ingólfsson* 1982, *Hersteinsson* 1989). Gulls are especially abundant around fishprocessing-plants, which are numerous in the northwest and distance to summer pastures is short. Salmonella is very hardy and can survive for more than 2 years in bird faeces (*Fiennes* 1982). A further indication that gulls and possibly other wild birds may spread the infection to sheep is that the same serotype, i.e. *S. thompson,* has been detected in gulls (unpublished results).

We did not find evidence of salmonella contamination in slaughterhouses. Thus, salmonella was neither detected in singed sheep heads nor in drainage pipes in slaughterhouses. However, the reservation must be made that we got very few samples from drainage pipes.

It is well known that animals can be healthy carriers of salmonella (*Brogden et al.* 1994, *Gitter et al.* 1970). Our findings that the bacterium can be harboured in tonsillae are in agreement with Gitter's results (1970).

In short, our results have shown a low prevalence of salmonella infection in Icelandic sheep and that infection apparently occurs on mountain-pastures that may be contaminated by wild birds, especially gulls that carry the infection.

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Sammendrag

Salmonella i får i Island.

I året 1995 forekom i Island nogle tilfælde af madforgiftning i mennesker, som stammede fra salmonellasmitte i svedne fårehoveder. Dette motiverede os til at undersøge periodetallet af salmonellasmitte i får og hvor og hvordan smitten kunne have fundet sted. Ekskremental, tarmindeholds- og mandelprøver blev samlet i foråret og efteråret fra får på 50 gårde i sydvest Island, hvor salmonellasmitte havde blevet opdaget, og fra 5 gårde i nordvestlandet. Alle ekskrementale prøver fra sydvestlandet var negative, men prøver fra 3 gårde, som blev taget i efteråret i nordvestlandet, var positive. Mandelprøver som blev taget i efteråret var positive i får fra 3 gårde i sydvestlandet og 2 i nordvestlandet. Salmonella fandtes hverken i afløb fra slagterier eller i svedne fårehoveder. Vores resultater viser at salmonellasmitte er sjælden i islandske får, men sunde lam kan bære bakterien i mandlen.

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Reprints may be obtained from: S. Hjartardóttir, Institute for Experimental Pathology University of Iceland, Keldur, Iceland. E-mail: sigrhj@hi.is (S. Hjartardóttir), tel: +354-5674700, fax: +354-5673979.