

Brief Communication

**ISOLATION OF CAMPYLOBACTER FETUS SUBSP. JEJUNI
FROM THE GALLBLADDER OF NORMAL SLAUGHTER
PIGS, USING AN ENRICHMENT PROCEDURE**

Campylobacter fetus subsp. *jejuni* has been reported as a cause of enteritis in man (*Skirrow* 1977) and animals (*Hastings* 1978). There are also reports of disease transmission from animals to man (*Skirrow et al.* 1980, *Svedhem & Norkrans* 1980) and via foodstuffs to man (*Tiehan & Vogt* 1978, *Robinson et al.* 1979). It is therefore of great epidemiological significance to define the reservoir of this bacterium. Using the methods described by *Skirrow* and *Lauwers et al.* (1978), *Campylobacter fetus* subsp. *jejuni* has been isolated from the intestinal contents in 72.1 % (*Jørgensen* 1979) and 60.7 % (*Oosterom* 1980) of clinically healthy pigs. A survey of the prevalence of *Campylobacter* species in the gallbladder of normal sheep and cattle showed that 16 % of the sheep and 17 % of the cattle were infected (*Bryner et al.* 1972).

In the present investigation the gallbladders together with 1—2 cm of the associated main bile ducts from 50 clinically healthy pigs were removed immediately after slaughter. The samples were collected at an approved slaughterhouse, and represented a total of 22 herds in the Oslo area. Six of the pigs were sows, while the remainder were ordinary bacon pigs. In addition, swabs from the surface of the liver and gallbladder of 30 other bacon pigs were taken before the gallbladders were removed during the routine slaughter process. The samples were taken on 4 different days, and investigation of the samples started within 3 h after collection. Material from the internal wall of the gallbladder and some bile, were seeded directly onto the surface of selective agar (*Skirrow*). The gallbladders with associated bile ducts were then added to 100 ml of enrichment medium, and the swabs were transferred to test tubes containing 5 ml of the same medium. The enrichment medium consisted of: Pepton (Difco 0118) 10 g, "Lab lemco" powder (Oxoid L 29) 8 g, Yeast extract (Oxoid L 21) 1 g, NaCl 5 g, Rezasurin 0.025 % 16 ml, Vancomycin 10 mg, Trimethoprim Lactate 5 mg, Polymyxin B 2500 IU, Distilled water to 1000 ml. All the samples were incubated at 42°C in a microaerophilic atmosphere created by

means of Gas-Pak sachets in anaerobic jars without catalysts. Secondary seeding was carried out from the enrichment medium onto selective agar after 24 and 48 h. Typical colonies were examined morphologically and for typical motility under the phase contrast microscope and tested for catalase and oxydase reactions, growth under aerobic and anaerobic conditions at 42°C, growth under microaerophilic conditions at 25°C, and for sensitivity to nalidixic acid.

Table 1 shows, that *Campylobacter* bacteria were isolated from 58 % of the samples from the bile/gallbladders and bile ducts after 48 h incubation in the enrichment medium. These bacteria could not be isolated after direct seeding on selective agar plates. Swabbing of the livers before the gallbladders were removed resulted in isolation of *Campylobacter* bacteria from 7 out of 30 samples (23 %). All the strains were catalase and oxydase positive. The bacteria did not grow under aerobic or anaerobic conditions at 42°C or microaerophilic at 25°C. Three of the strains were not sensitive to nalidixic acid. The results of morphological and biochemical tests of the isolated strains are in accordance with the characteristics of *Campylobacter fetus* subsp. *jejuni*.

Table 1. The prevalence of *Campylobacter fetus* subsp. *jejuni* in the gallbladders/bile ducts of slaughter pigs.

	Number of samples	Number of isolates	Isolates, %
Direct seeding	50	0	0
24 h enrichment	50	21	42
48 h enrichment	50	29	58

The high prevalence of *Campylobacter fetus* subsp. *jejuni* in gallbladders/bile ducts of clinically healthy slaughter pigs indicates that these sites are important reservoirs of these bacteria. During the routine slaughter process and post mortem examination, the main bile ducts are incised and some bile can often be spilt onto the liver. This could explain the occurrence of *Campylobacter* on the surface of the liver. The enrichment medium described appears suitable for the investigation of *Campylobacter* from gallbladders and bile ducts. The medium has also been found useful in the isolation of this bacterium from foodstuffs (Rosef, unpublished).

O. Rosef

The Department of Food Hygiene, Veterinary College of Norway, Oslo.

REFERENCES

- Bryner, J. H., P. A. O'Berry, P. J. Estes & J. W. Foley*: Studies of vibrios from gallbladder of market sheep and cattle. *Amer. J. vet. Res.* 1972, 33, 1439—1444.
- Hastings, D. H.*: *Campylobacter* enteritis in pets. *Lancet* 1978, 2, 1249—1250.
- Jørgensen, K.*: Forekomst af *Campylobacter* hos svin. (The occurrence of *Campylobacter* in swine). *Nord. Vet.-Med.* 1979, 31, 534.
- Oosterom, J.*: Het voorkomen van *Campylobacter fetus* subspecies *jejuni* bij normale slachtvarkens. (The presence of *Campylobacter fetus* subspecies *jejuni* in normal slaughtered pigs). *T. Diergeneesk.* 1980, 105, 49—50.
- Lauwers, S., M. De Boeck & J. P. Butzler*: *Campylobacter* enteritis in Brussels. *Lancet* 1978, 1, 604—605.
- Robinson, D. A., W. J. Edgar, G. L. Gibson, A. A. Matchett & L. Robertson*: *Campylobacter* enteritis associated with consumption of unpasteurised milk. *Brit. med. J.* 1979, 1, 1171—1173.
- Skirrow, M. B.*: *Campylobacter* enteritis: A "new" disease. *Brit. med. J.* 1977, 2, 9—11.
- Skirrow, M. B., G. L. Turnbull, R. E. Walker & S. E. J. Young*: *Campylobacter jejuni* enteritis transmitted from cat to man. *Lancet* 1980, 1, 1188.
- Svedhem, A. & G. Norkrans*: *Campylobacter jejuni* enteritis transmitted from cat to man. *Lancet* 1980, 1, 713—714.
- Tiehan, W. & R. L. Vogt*: Waterborne *Campylobacter* gastroenteritis—Vermont. *MMWR* 1978, 27, 207.

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Reprints may be requested from: O. Rosef, the Department of Food Hygiene, Veterinary College of Norway, P.O. Box 8146 Dep., Oslo 1, Norway.