

ISOLATION OF CAMPYLOBACTER COLI FROM A COW WITH MASTITIS

K. Serdar Diker¹

Jale Erdeğer²

Levent Keskintepe³

Mustafa Arda⁴

Mastitisli bir inekten *Campylobacter coli* izolasyonu

Özet: Mastitisli bir ineğin iki meme lobundan *Campylobacter coli* izole edildi. İneğin dışkı kültürü campylobacterler yönünden negatif bulundu. Mikroorganizma, ilk örneğin alınmasından 15 gün sonra aynı meme loblarından yeniden izole edildi. İzole edilen *C. coli* suşu ampisillin, penisillin ve chloramphenicol'e dirençli, erythromycin, kanamycin, neomycin, streptomycin ve tetracyclin'e duyarlı bulundu. İzole edilen organizma 10^8 koloni oluşturan ünite miktarında pastörize edilmemiş süte inoküle edildiğinde, 37 °C de 5 gün ve 4 °C de 18 gün canlı kaldı.

Summary: *Campylobacter coli* was isolated from two quarters of a cow with mastitis. Faecal culture of cow was negative for campylobacters. Organism was reisolated from same two quarters 15 days after first sampling. *C. coli* isolated was resistant to ampicillin, penicillin and chloramphenicol but sensitive to erythromycin, kanamycin, neomycin, streptomycin and tetracycline. Organism survived 5 days at 37 °C and 18 days at 4 °C in unpasteurised milk after inoculation of 10^8 colony forming unit of bacteria.

The recent reports of experimental infection of the bovine mammary gland with *Campylobacter jejuni* raised the possibility of a naturally occurring campylobacter mastitis (3). Subsequently this was confirmed by the isolation of *C. jejuni* (2, 7) and aerotolerant campylobacter (6) from the field cases of bovine mastitis. *C. jejuni* can also be isolated from normal milk (1). However, many surveys have found the incidence of contaminated milk samples to be very low (2, 8). Although *C. jejuni* is documented in cases of bovine mastitis, there is no report on the isolation of *C. coli* from bovine mastitis.

1 Dr., A.Ü. Veteriner Fakültesi, Mikrobiyoloji Anabilim Dalı, Ankara.

2 Arş. Gör., A.Ü. Veteriner Fakültesi, Mikrobiyoloji Anabilim Dalı, Ankara.

3 Arş. Gör., A.Ü. Veteriner Fakültesi, Doğum ve Reprodüksiyon Hastalıkları Bilim Dalı, Ankara.

4 Prof. Dr., A.Ü. Veteriner Fakültesi, Mikrobiyoloji Anabilim Dalı, Ankara.

Several freshly calved cows in a dairy herd showed signs of clinical mastitis. Mastitic condition was confirmed by using California Mastitis Test (CMT). Thirty-seven milk samples from affected quarters were collected after washing and drying of the gland, discarding of the fore-milk and sterilising the teat apex with alcohol. Samples were inoculated onto blood agar plates for common bacteria and incubated in aerobic and carboxyphilic conditions at 37 °C for 48 hours. In addition to the direct culture, 1 ml of milk sample was also cultured for campylobacter in 9 ml of brucella broth (Difco) containing Preston selective supplement (Oxoid) and incubated in microaerophilic atmosphere at 42 °C for 18 hours. After enrichment procedure, 0.1 ml of this culture was seeded onto 7 % defibrinated sheep blood agar containing same supplement and incubated in microaerophilic condition at 42 °C for 48 hours. *C. coli* strains were identified on the basis of criteria described by Lior (5).

Antibiotic susceptibility of two strains of *C. coli* was tested by agar disk diffusion technique. The survival of isolates was tested in unpasteurised milk held in refrigeration temperature and at 37 °C.

Two *C. coli* strains were isolated in pure culture on selective medium from left-fore and right-hind quarter of a cow with mastitis. Both isolates were negative in hippurat hydrolysis and rapid H₂S tests, sensitive to nalidixic acid (30 mcg), resistant to cephalothin (30 mcg). Since all phenotype characteristics of both isolates were same, they were considered as a single strain. No microorganisms other than *C. coli* were isolated from affected quarters on other media. The cow was a Swiss Brown at the age of 9 and did not show the signs of systemic illness. The faeces of this cow was found negative for campylobacters. The milk samples from affected quarters were positive (+ 3) in CMT. Two weeks after first sampling, milk samples were collected from all quarters of this cow and reexamined for campylobacter and other bacteria. Same *C. coli* strain was isolated in pure culture from left-fore and right-hind quarters.

Both *C. coli* isolates survived 5 days at 37 °C and 18 days at 4 °C after inoculation of 10⁸ colony forming unit of bacteria into 1 ml of unpasteurised milk. Both strains were resistant to ampicillin, penicillin, chloramphenicol and sensitive to erythromycin, kanamycin, neomycin, streptomycin and tetracycline.

Lander and Gill (3) produced clinical mastitis in cows by intramammary inoculation of *C. coli/jejuni* in doses ranging from 2.6 to 3.8

X 10⁹ colony forming unit. However, *C. coli* and *C. jejuni* were considered as a single species in that study, and it was not stated whether this strain was *C. coli* or not. Before the establishment of current classification (5), the distinction between *C. coli* and *C. jejuni* was not clear and *C. coli* was shown to be a biotype of *C. jejuni*. Although *C. coli* and *C. jejuni* have many phenotypic characteristics in common, DNA homology studies have shown that they are separate species (4).

Isolation of *C. coli* in pure culture from milk samples of affected quarters and negative findings for other known causes (*Streptococcus sp.*, *Staphylococcus sp.*, *Corynebacterium pyogenes* and coliforms) of bovine mastitis strongly suggested that *C. coli* was the aetiological agent of this case. Reisolation of *C. coli* from same quarters at the second sampling supported this suggestion. The source of *C. coli* could not be identified in this case. Since *C. coli* was not isolated from the faeces of cow the source of this infection can not be considered as a contamination from faeces. As searching the literature related with both *C. coli* and bovine mastitis it could not be found any. In the view of these data, it can be concluded that as far as is known this is the first report of naturally occurring bovine mastitis caused by *C. coli*.

Experiments to produce mastitis in mice are in progress.

References

1. **Humphrey, T.J. and Beckett, P.** (1987). *Campylobacter jejuni* in dairy cows and raw milk. *Epidemiol. Infect.*, 98: 263-269.
2. **Hutchinson, D.N., Bolton, F.J., Hinchcliffe, P.M., Dawkins, H.C., Horsley, S.D., Jessop, E.G., Robertshaw, P.A. and Counter, D.E.** (1985). Evidence of udder excretion of *Campylobacter jejuni* as the cause of milk-borne campylobacter outbreak. *J. Hyg.*, 94: 205-215.
3. **Lander, K.P. and Gill, K.P.W.** (1980). Experimental infection of the bovine udder with *Campylobacter coli*/*jejuni*. *J. Hyg.*, 84: 421-428.
4. **Leaper, S. and Owen, R.J.** (1982). Differentiation between *Campylobacter jejuni* and allied thermophilic campylobacters by hybridization of deoxyribonucleic acids. *FEMS Microbiol. Lett.*, 15: 203-208.
5. **Lior, H.** (1984). New extended biotyping scheme for *Campylobacter jejuni*, *Campylobacter coli* and *Campylobacter lariidis*. *J. Clin. Microbiol.*, 20: 636-640.
6. **Logan, E.F., Neill, S.D. and Mackie, D.P.** (1982). Mastitis in dairy cows associated with an aerotolerant campylobacter. *Vet. Rec.*, 110: 229-230.
7. **Morgan, G., Chadwick, P., Lander, K.P. and Gill, K.P.W.** (1985). *Campylobacter jejuni* mastitis in a cow: a zoonosis related incident. *Vet. Rec.*, 116: 111.
8. **Waterman, S.C., Park, R.W.A. and Bramley, A.J.** (1984). A search for the source of *Campylobacter jejuni* in milk. *J. Hyg.*, 93: 333-337.