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ISOLATION OF *CORYNEBACTERIUM EQUI* FROM MILK

Corynebacterium equi, first described by Magnusson⁴ in 1923, is chiefly involved in suppurative pneumonia of horses and abscess formation in the swines^{1-5,7}. The organism has also been isolated from the genital tracts of mares, aborted equine fetuses and an aborted buffalo fetus⁶. Perusal of literature does not reveal the isolation of *C. equi* from cow's milk. The present report is to put on record the isolation of *C. equi* from a milk sample of a cow which had mastitis due to *Klebsiella*.

During an investigation of mastitis in a dairy herd, *Klebsiella pneumoniae* was isolated from 5 cases out of the 6 cows affected. In one case, *C. equi* was isolated along with *K. pneumoniae*. The corynebacterium had the following characteristics: The colonies on tryptose agar were round convex with regular margin. They developed a bright pink colour in 10-12 days incubation at 37° C. The pigment production was faster when the plates were incubated at room temperature. Microscopic examination of the organisms from solid medium revealed them to be gram positive bacilli, Neisser's staining showed 2-3 metachromatic granules in 50% of the cells. The isolate was non-motile, capsulated and did not ferment any of the sugars tested. Nitrates were reduced to nitrites within 24 h. Litmus milk was unchanged. Gelatin was not liquified. Methyl red and Voges-Praskauer tests were negative. It withstood 2.5% oxalic acid treatment for 45 min. The isolate conformed to all the characteristics of *Corynebacterium equi* as described in the *Bergey's Manual of Determinative Bacteriology* (1974).

As already mentioned, the presence of *C. equi* in the udder of cattle has not been reported in the literature. The present isolation of this organism from a bovine udder affected with *Klebsiella* mastitis is unusual and could be explained by the fact that the dairy had a small piggery unit attached to it and workers were common to both the units. Contamination from the exterior of the udder was eliminated by collecting the milk with strict aseptic precautions. It is likely in this particular case that *C. equi* was transmitted from pigs, possibly harbouring it, to the cows through common workers.

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DIFFERENTIAL EFFECTS OF PROSTAGLANDINS ON CARTILAGE

THE growth and maturation of cartilage is a complex process and is influenced by a number of hormonal and metabolic regulatory factors^{1,2}. The mechanism of action of various factors on cartilage varies with the type of cartilage used, the nature of the incubation medium and the age of the animals, etc. Prostaglandins have been suggested to influence macromolecular synthesis in chick embryonic and hypophysectomized rat cartilage³. Various prostaglandins (PGA, PGB, PGE₁, PGE₂ and PGF₁α) have been shown to elevate cyclic 3', 5'-adenosine monophosphate (cyclic AMP) levels in chick embryonic cartilage. However, only PGA and PGB caused inhibition of synthesis of DNA, RNA, proteins, and proteoglycans in cartilage³. PGE₁ and theophylline, which raise intracellular levels of cyclic AMP, also increased transport of α-amino-isobutyric acid in chick embryonic cartilage⁴. In the present communication, data on the effects of prostaglandins (E₁, E₂, F₁α and F₂α) on the macromolecular synthesis in cartilage is presented. Uptake of ³H-proline in the chick embryonic cartilage was studied which represents the synthesis of collagen, an important constituent of the cartilage matrix.

Chick embryonic cartilage (Pelvic rudiments, 12-days age) was incubated in tissue culture medium-199⁵ at 37° C. 0.5 μci/ml of ³H-proline (Amersham, 1 ci/m mole) was added either in the beginning or after preincubation with various prostaglandins. The reaction was terminated by chilling the tubes in ice and washing the tissue (5-6 times) with ice cold medium

TABLE I
Effect of prostaglandins on incorporation of ³H-proline into chick embryonic cartilage

Sl. No.	Additions	Concentration (μg/ml)	Experiment 1		Experiment 2	
			DPM/mg cartilage ± SE	% change	DPM/mg cartilage ± SE	% change
1.	None		4162 ± 788		5210 ± 146	
2.	NHS	10%	7177 ± 172	72 ↑		
3.	PGE ₁	25	..		4943 ± 741	
		50	5074 ± 402	22 ↑	6241 ± 422	20 ↑
		100	5055 ± 328	21 ↑	6043 ± 675	16 ↑
4.	PGE ₂	50	..		3938 ± 1370	24 ↓
		100	3358 ± 56	19 ↓	3988 ± 154	23 ↓
		200	1707 ± 147	59 ↓	..	
5.	PGF _{1α}	50	..		6492 ± 958	25 ↑
		100	4525 ± 154	9 ↑	5451 ± 987	
		200	4993 ± 516	20 ↑	..	
6.	PGF _{2α}	50	..		4822 ± 1585	
		100	5092 ± 44	22 ↑	6712 ± 512	29 ↑
		200	5008 ± 407	20 ↑	..	

Pelvic rudiments from chick embryos (12 days) were incubated in Medium - 199 (2.0 ml) supplemented with ³H-proline (1 μCi). In the experiment 1, ³H-proline was added after 2 hours of preincubation with various prostaglandins and further incubated for 6 hours at 37°C in 'Dubnoff Metabolic Shaker' whereas in Experiment 2 no preincubation was carried out. Reaction was terminated by chilling the tubes and washing thoroughly with PBS containing nonradioactive L-proline. Procedure for measuring the radioactivity is described in the text. Vertical arrows, ↑ or ↓, stand for increase or decrease respectively. Values are mean of 3-5 observations ± SE.

containing 1 mM L-proline. Cartilages were weighed and dissolved in Soluene-350 (Packard) and radioactivity determined after adding 10.0 ml scintillation mixture [4g, 2, 5-diphenyloxazole and 0.5 g 1,4-Di (2-(5-phenyloxazolyl)) benzene per 1000 ml toluene]. Before counting, the vials were kept in a cool dark place for stabilization. Quenching correction was made using automatic standardization in the counter (Packard Tri-Carb Liquid Scintillation Spectrometer) and counts per minute (CMP) were converted into disintegrations per minute (DPM).

Results of uptake and incorporation of ³H-proline into chick embryonic cartilage are presented in Table I. Stimulation of uptake of ³H-proline by normal human serum can be attributed mainly, to the growth hormone (GH) dependent serum somatomedins which mediate actions of GH on bones and cartilage⁶. Among various prostaglandins tested PGE₁, PGF_{1α} and PGF_{2α} showed marginal stimulation whereas PGE₂ inhibited the uptake and incorporation of ³H-proline in the cartilage. Physiological significance of these data is however, not clear. Whether differential effects of prostaglandins are reflected in the cyclic AMP levels remains to be tested.

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