

Conclusions. The experiments carried out showed that adriamycin significantly depressed the level of circulating antibodies; its action depends on the total dose of the

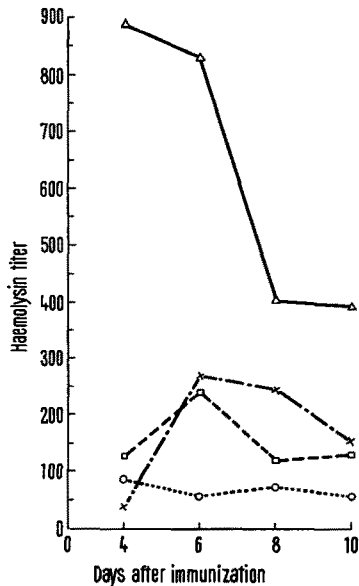


Fig. 3. Effect of adriamycin on the primary response of mice immunized with sRBC. The total dose of adriamycin administered was 9 mg/kg for the 3 different experimental groups of mice. Δ - Δ , control group. \times - \times , treatment at days -3, -2, -1 (3 mg/kg/die i.v.). \square - \square , treatment at days -3, -2, -1, +1, +2, +3 (1.5 mg/kg/die i.v.). \circ - \circ , treatment at days +1, +2, +3 (3 mg/kg/die i.v.). \circ , day of immunization.

drug administered. The highest immunodepressive effect was observed when the treatment followed the antigen administration. This finding suggests that adriamycin does not interfere with the very early steps of immunological response.

The immunodepressive effect of this antibiotic probably depends on its action on lymphatic tissue and on highly proliferating cell systems¹⁴. In fact, as chronic toxicity studies on the dogs demonstrated¹⁵, adriamycin causes a marked reduction of lymphatic tissue of thymus, lymphnodes, spleen and provokes a blood lymphocytopenia. Besides, adriamycin, interfering with nucleic acid synthesis¹², has an inhibiting effect on bone marrow cells, and on blood lymphocytes stimulated by PHA¹¹; a similar action can be supposed on immunocompetent cells after antigenic stimulation.

Riassunto. Studi sperimentali hanno messo in evidenza che l'adriamicina possiede un'azione immunodepressiva in quanto determina una inibizione significativa del titolo degli anticorpi emolitici ed emoagglutinanti in topi immunizzati con globuli rossi di montone.

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¹⁴ C. BERTAZZOLI, T. CHIELI, M. GRANDI and G. RICEVUTI, *Experientia* 26, 389 (1970).

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Observations on Non-Specific Reactions to Tuberculin in Sheep and Goats with *Corynebacterium ovis*

The problem of non-specific reactions to tuberculin amongst cattle has been very commonly met with in many countries where the campaign for tuberculosis eradication has been taken up on a large scale. In areas relatively free from bovine tuberculosis, the problem of non-specific reactions was mostly confronted. Various causative organisms, namely actinomyces, actinobacillus, *Johnes' Bacillus faciola*, brucella species, and pyogenic infections have been reported to elicit non-specific reactions to tuberculin in cattle (KLEBERG¹, RUSFORD², and TRANTWEIN³). CANHAM⁴ also reported sensitivity to tuberculin in guinea-pigs infected with the *Corynebacterium pyogenes*. But from the literature it is evident that non-specific sensitization to the tuberculin in ovines and caprines has not been reported so far.

The present paper includes the study of a naturally infected goat and a sheep which reacted to tuberculin in routine testing of the herds. These reactors were sacrificed for the isolation of the causative organisms and their characterisation.

Material and methods. Case No. 1. An indigenous goat No. 426 of Katula goat farm at Indian Veterinary Research Institute, Mukteswar-Kumaon, was subjected to double intradermal test with tuberculin and johnin simultaneously. The animal reacted to the tuberculin but not to johnin. This goat was sacrificed and on thorough post-mortem examination an abscess of 2" diameter was found in the diaphragmatic lobe of right lung which had thick hard capsule and was filled with soft caseated yellowish

pus. No other lesions were seen in other visceral organs. Smears were also made from ileo-caecal gland for detection of *Johnes' bacillus*.

A direct attempt was made to isolate culture from the abscess. The smears were made and stained with Zeihl-Neelson's and Gram's stains. The pus material along with fibrous capsule was triturated aseptically in sterile saline and was inoculated into 2 guinea-pigs by i.m. route, 3 fowls by i.v. route and 2 kids by subcutaneous route in neck. Each animal was inoculated with 1 ml of suspension.

Kids were also infected with new isolates by different routes. Infected animals were tested with tuberculin at varying intervals of time.

Case No. 2. A Bikaneri sheep of the Genetics Division, I.V.R.I., Izatnagar, elicited reaction to tuberculin and was negative to johnin. Sheep was suspected for tuberculosis and subjected to s.c. tuberculin test. Significant rise in temperature of 3.5° F to 4° F was noted at 12-15 h after s/c test. Sheep was sacrificed and on post-mortem examination 3-4 abscesses of 1' to 3' diameter were found in the spleen. Abscesses were like that of goat.

¹ H. H. KLEBERG, *Jl. S. Afr. vet. med. Ass.* 35, 103 (1964).

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³ K. TRANTWEIN, *Bull. Off. Int. Epizoot.* 50, 305 (1958).

⁴ A. A. CANHAM, *Onderstepoort J. vet. Sci. Anim. Ind.* 19, 29 (1944).

Observations on non-specific reaction to tuberculin in sheep and goats infected with *Corynebacterium ovis*

No. of animals	Material inoculated	Dose, route and sites of inoculation (1 ml)	Readings of tuberculin test (mm)			
			5 weeks interval		70 days interval	
			Original	Finalreading	Original	Finalreading
Kid No. 74	Pus of goat 426	s.c. neck	2.3	7.9	2.4	11.0
Kid No. 75	Pus of goat 426	s.c. neck	2.2	6.0	2.3	7.5
Kid No. 391	48 h old culture isolate I	i.t.	2.9	8.6	3.0	9.5
Kid No. 392	48 h old culture isolate I	i.t.	2.5	10.2	2.5	10.9
Kid No. 394	48 h old culture isolate II	i.t.	2.6	11.3	2.5	9.6
Kid No. 395	48 h old culture isolate II	s.c.	2.7	9.8	2.5	9.8
Kid No. 390	Standard strain 48 h culture	i.t.	3.0	8.1	2.5	8.0
	Standard strain 48 h culture	s.c.	2.7	9.2	3.5	7.9
Average increase in thickness of skin (mm)			2.6	8.9	2.7	9.25
Lamb No. 12	Pus of sheep 311	s.c. neck	1.5	10.8	1.5	12.0
Lamb No. 11	Pus of sheep 311	s.c. neck	2.0	11.6	1.6	14.0
Lamb No. 15	Isolate I 48 h old culture	s.c. neck	1.7	13.0	1.6	14.0
Lamb No. 16	Isolate I 48 h old culture	s.c. neck	1.5	12.0	1.3	13.6
Lamb No. 17	Isolate II 48 h old culture	s.c. neck	1.4	12.0	1.5	15.7
Lamb No. 18	Isolate II 48 h old culture	s.c. neck	1.3	11.0	1.4	14.6
Lamb No. 13	Standard strain	s.c. neck	1.5	9.8	1.5	10.0
Lamb No. 14	Standard strain	s.c. neck	1.6	10.0	1.4	11.0
Average increase in thickness of skin (mm)			1.5	11.2	1.4	14.6

Organisms were isolated and sheep were infected with 48 h old glucose broth culture as well as with the suspension of the abscess. These were tested with tuberculin before infection as well as after infection. Biochemical morphological characters of the new isolates were compared with the standard strain of *Corynebacterium ovis*.

Results. Smears from the abscesses of both the cases did not reveal any acidfast bacteria but Gram positive, pleomorphic coccobacillary micro-organisms were seen in teeming numbers. Similar bacteria were also seen in the abscess material of artificially infected kids and lambs with new isolates.

Guinea-pigs and fowls were also sacrificed but they did not reveal any gross pathological lesion and smears from different visceral organs were negative for the presence of either Gram positive or acid fast organisms.

Kids and lambs infected with either original abscess material or new isolates developed local abscesses at the sites of inoculation. Lymphatics of neck showed marked cording which was more pronounced in the animals infected with new isolates than in those infected with standard strain. The cording of lymphatics was more severe in lambs than in kids. Abscesses at the site of inoculations in the animals infected with standard strain subsided after 45 days while with new isolates they persisted as long as 150 days. Goat No. 391, which was infected intra-tracheally with new isolates, died on 40th day of infection and had multiple suppurative abscesses in the lungs with severe congestion of diaphragmatic and intermediate lobes. Smears from these lesions revealed similar organisms.

Kids and lambs infected with new isolates as well as with standard strain were subjected to tuberculin test at varying intervals after infection. They elicited significance reaction at 5 weeks interval which was more marked at later intervals. The details of the tuberculin test are given in the Table. It is apparent from the Table that sensitization of the animals infected with standard strain was less than that of new isolates, the former exhibited less of tuberculin reaction than the latter. However, it was interesting to note that tuberculin reaction was more pronounced in lambs than in kids. The average increase

in the thickness of skin at the site of tuberculin test was 3.4 times in kids and 7.4 times in lambs at 5 weeks interval, and 3.9 times and 9.1 times respectively at 70 days interval.

Biochemical characters of the new isolates were compared with that of *C. ovis* standard strain. Both isolates could ferment glucose, galactose, trehalose, but there was no change in litmus milk, lactose maltose, manitol, sucrose and dextrose. Both were found negative for NH₃, H₂S, Indol, Nitrate, M.R. and V.P. There was no growth on gelatin but pellicle formation on broth like standard strain. New isolates were diphtheroid in appearance and closely resembling the standard strain of *C. ovis*.

Discussion and conclusion. On the basis of biochemical, morphological and pathogenicity tests, the 2 new isolates were proved to belong to *Corynebacterium* species. These strains were found to be pathogenic to lambs and kids but not to guinea-pigs and fowls. Both naturally as well as artificially infected animals reacted to tuberculin. The reaction was more severe in lambs than in kids, probably this was due to loose texture of skin. This clearly indicated that *C. ovis* elucidates non-specific reaction in sheep and goats to tuberculin in naturally infected animals as well as in experimental ones. This might be either due to sharing of some common antigenic components between 2 organisms, or even due to severe necrosis of animal tissues⁵.

Résumé. La tuberculine provoque une réaction non-spécifique sur les moutons et les chèvres qui sont, soit naturellement, soit artificiellement, infectés avec *Corynebacterium ovis*.

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