

The former posterior pole of the terminal organ of the graft, now anteriorly directed, may at the same time give rise to a chain of organs extending anteriorly in the rapidly growing graft. From the 2 poles in such a terminal organ the line of new organs may equal in number that of the primary organs found in the entire tail on one side of a normal young larva of *A. punctatum*.

The terminal organ of the host may give rise to a chain of organs extending posteriorly into the region of the graft, a reaction similar to simple amputation of the tail tip. The organ of the graft nearest the host tissue may also give off bud-organs anteriorly (from former posterior pole) to meet such a chain from the host. This seems to depend upon the direction and extent of the early regeneration in host and graft tissue.

8259 C

Caseation and Softening in Tuberculosis of Vaccinated and Unvaccinated Guinea Pigs.

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There is much evidence that vaccination with the BCG strain of the tubercle bacillus produces a certain degree of resistance to reinfection with virulent tubercle bacilli. The question has been raised, however, as to whether the allergy resulting from such vaccination might be a factor in producing a more extensive and more destructive type of tuberculosis. In an experiment to ascertain if any relationship exists between allergy and the character of the tuberculous lesions, one series of guinea pigs was injected subcutaneously in the left inguinal region with 5 mg. of the BCG culture, a second series was injected in a similar manner with 30 mg. of the virulent Ravenel strain of bovine tubercle bacillus killed by heating for 1 hour at 70°C., and a third group of unvaccinated guinea pigs were used as controls.

Six weeks after vaccination the sensitivity of these animals to old tuberculin was determined by injecting into the skin of each animal, at widely separated points, 1, 5 and 10 mg. of old tuberculin, and reading the results 48 hours later. It was found that among the 14 guinea pigs of the first series, injected with BCG, 9 reacted to 1.0 mg. of tuberculin and 5 to 5.0 mg. Of the 15 guinea pigs of the

second series, injected with the heat-killed tubercle bacilli, 3 reacted to 1.0 mg. of tuberculin, 6 to 5.0 mg., 5 to 10.0 mg., and one failed to react. The 11 untreated control guinea pigs did not react to the injection of tuberculin. The vaccinated as well as the unvaccinated control animals were then injected intratracheally while under ether anesthesia with approximately 100 tubercle bacilli of the virulent Ravenel bovine strain. Six weeks later all of the animals were killed and necropsied.

Results in Guinea Pigs Vaccinated with BCG Cultures. The cervical lymph nodes were tuberculous in but one animal of this group. The hilum lymph nodes, which were enlarged in all the pigs, measured on an average 12x7x4 mm. They were firm and fibrous, except in an occasional animal in which they were fibrocaseous. Microscopically these nodes showed marked increase of the stroma and the capsule was thickened and fibrous. Widely separated small areas of caseation were noted in the sections of most animals, but in an occasional animal caseation was extensive and confluent.

The lungs contained a small number of discrete tubercles ranging from less than 1 to 2 mm. in diameter. These tubercles were most numerous in the lower lobe, and only occasionally was caseation noted macroscopically. Microscopically these tubercles consisted of either a diffuse intra-alveolar infiltration of large mononuclear cells or of aggregations of these cells about the smaller bronchi. In 7 of the 14 guinea pigs of this series small areas of caseation surrounded by epithelioid cells were observed.

The spleens, slightly larger than normal, measured on an average 27x14x2 mm. In 3 of the animals a small number of discrete minute tubercles were seen macroscopically. Microscopic examination of the spleens showed that in many instances the Malphigian corpuscles and germinal centers were hypertrophied. The splenic pulp contained an increased number of large mononuclear cells. In the spleen of 3 animals a number of epithelioid tubercles were observed, with small caseous centers. In the liver of one animal a small tubercle was noted, while in another animal the periportal lymph node was fibrocaseous.

Tubercle bacilli were not demonstrable in any of the lesions, nor was softening observed in any of the tuberculous tissue studied.

Results in Guinea Pigs Vaccinated with Heat-killed Tubercle Bacilli. The cervical lymph nodes were found to be enlarged and caseous in 5 of 15 guinea pigs. The hilum lymph nodes, which were enlarged and caseous, measured on an average 23x12x7 mm. Mi-

Microscopically these nodes showed extensive and confluent caseation associated in 13 instances with softening. This softening was characterized by definite dissolution and separation of the necrotic material from the adjacent tissue, resulting in an abscess-like formation. In 8 instances tubercle bacilli were found. In the lungs there were observed numerous tubercles 3 to 5 mm. in diameter, many of which were caseous. Microscopically the tubercles, usually peribronchial, consisted of extensive areas of caseation surrounded by numerous epithelioid cells and a small layer of lymphocytes at the periphery. The alveoli were filled with epithelioid cells, the lymphatics were dilated, and conspicuous aggregations of lymphocytes were noted about the larger blood vessels. In a small number of animals extensive edema was present. Softening of the caseous areas was observed in the lungs of 11 animals, and tubercle bacilli were found in 8 instances.

The spleens were markedly enlarged, averaging 38x20x4 mm. in size. They were dark red or purplish and their surfaces were mottled by grayish areas of caseation. Microscopically a marked infiltration of epithelioid cells and numerous giant cells were noted. Caseation was extensive and, in some instances, confluent. Softening was observed in 3 instances, while tubercle bacilli were demonstrated in the spleens of 6 animals. In 13 of the 15 pigs, the liver was found to have an occasional tubercle, and in the remaining 2 animals tuberculosis was extensive. The periportal lymph nodes were enlarged and caseous in every animal.

Results in Unvaccinated Guinea Pigs. The cervical lymph nodes were enlarged and caseous in 6 of the 11 unvaccinated control animals. The enlarged caseous hilum lymph nodes averaged 21x12x7 mm. Microscopically the pathological findings resembled those noted in the hilum lymph nodes of the animals injected with the heat-killed tubercle bacilli, but the caseation was more extensive. Softening was observed in 8 instances and tubercle bacilli were demonstrable in 6 instances. In the lungs a number of tubercles ranging in size from 3 to 5 mm., and in some instances larger wedge-shaped confluent tubercles measuring 10 to 15 mm., were noted. Microscopic examination revealed areas of extensive caseation, some of which were confluent. Epithelioid cells were noted in large numbers about the caseous centers and in the alveoli. The tubercles were located about the smaller bronchi and beneath the pleura. Softening and tubercle bacilli were observed in 7 instances. In some animals there was noted a diffuse edema. In general the tuberculous process was more extensive in this series of guinea pigs than among those

vaccinated with either the BCG culture or with the heat-killed cultures of bovine tubercle bacilli.

The spleens, which were all enlarged, measured on the average 30x23x3 mm. In most instances they were studded with macroscopic tubercles of varying size. Microscopically numerous tubercles consisting of extensive areas of caseation were found; many of them confluent. Numerous epithelioid cells were observed about the caseous areas, as well as in other parts of the spleen. Softening of the tubercles was observed in 2 instances, and tubercle bacilli were demonstrable in the spleen of 6 animals.

Summary. Guinea pigs vaccinated with the attenuated BCG strain of tubercle bacillus, and thereby made allergic, were reinfected intratracheally with a small number of virulent tubercle bacilli. Examination of the animals 6 weeks after reinfection showed a small number of fibrous or fibrocaceous tubercles in the organs and lymph nodes in most instances, but failed to show any evidence of softening, nor could tubercle bacilli be demonstrated in the sections. Guinea pigs vaccinated with heat-killed tubercle bacilli, although less allergic, as well as control, unvaccinated guinea pigs that were not allergic, when reinfected with virulent tubercle bacilli showed 6 weeks after reinfection extensive tuberculosis of the organs and lymph nodes with well marked softening; and in most instances numerous tubercle bacilli were demonstrable in the sections. Thus, no relationship could be established between the presence of allergy at the time of reinfection and the subsequent softening of the tuberculous process.

8260 P

Nitrogen Content of the Bile of Normal and Diseased Gall Bladders.*

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A series of recent papers have indicated the possibility that simple bacterial infection is not sufficient to explain the onset of at least part of the clinical cases of cholecystitis. Not only has the diseased gall bladder in about half the cases defied the attempts of bacteriol-

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