intake. The concentration of the vitamin A in the liver, therefore may be compared with that in the blood; in the latter a maximal level was reached with an intake of 50 units, whereas in the liver the concentration continued to mount as the intake was increased.

Blood concentrations of less than 37 units per 100 cc of plasma were associated with no liver storage; those between 37 and 53 units were accompanied by little or no liver storage; whereas blood levels above 53 units were always associated with vitamin A in the liver, the amount stored, however, varying considerably and having no relation to the absolute blood level. These findings therefore indicate that the blood level may be of value in ascertaining whether there is any storage of vitamin A in the liver, the chief reservoir of this vitamin in the body. Further experiments concerning the relationship of blood level to storage are being carried out and will be reported at a later date.

We wish to express our appreciation to Mr. Bertrand Flusser and to Miss Mildred Weinstock for their coöperation in this study.

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Specific Cause and Nature of Ulcerative Enteritis of Quail.*

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In a former communication[1] I have identified a previously unrecognized, Gram-negative, anaerobic bacillus as the specific cause of ulcerative enteritis of quail (so-called "quail disease"). I also gave details of the technic by which the organism may be demonstrated in suitable material, directed attention to the chronic carrier as a factor in maintaining and spreading the infection and recorded observations indicating transmission of the infection to young quail chicks through eggs laid by carriers.

The purposes of the present communication are: (a) to correct and contradict a statement in the previous paper relative to spore

* Studies on which this report is based have been aided by donations from Mr. A. B. Freeman and Mr. C. S. Williams of New Orleans, and by a grant from the Special Research Fund of the Tulane University of Louisiana.

1 Bass, Charles C., PROC. SOC. EXP. BIOL. AND MED., 1939, **42**, 377.

formation, (b) to give technic for isolation of the organism in pure culture, (c) to call attention to coprophagy as an important source of transmission in captive quail, and (d) to record successful immunization of birds by vaccination with killed cultures of the specific organism.

(a) The statement previously made that "occasionally spores can be found on slide preparations made direct from lesions" has been found to be incorrect. Examination of hundreds of specimens made from pure cultures and from lesions has failed to show spores. Reëxamination of some of the original specimens on which the first statement was based reveals that what were taken for spores were something else, therefore contradicting the previous statement relative to spores.

(b) Pending development of differential culture media and cultural technic, and possibly more favorable culture media than is now available, attention is called to a medium and technic which are entirely satisfactory for isolation and maintenance of pure cultures of the organism This medium is the thioglycollate medium of J. H. Brewer,[2] which I modify slightly by the addition of 0.1% agar for semisolid cultures or 1% for solid cultures. The composition of the Brewer medium is as follows:†

Infusion from 37.5 g pork per 100 cc.	
Peptone	**1.0%**
Dextrose	1.0
Sodium chloride	0.5
Sodium thioglycollate	0.1
Agar	0.05
Methylene blue (1 to 500,000)	0.0002

The medium is tubed and autoclaved in the usual manner. The sodium thioglycollate establishes and maintains a state of anaerobiosis except at the surface which is exposed to the air and where partial aerobic condition is indicated by a green color imparted by the methylene blue.

The medium keeps well at room temperature for a month or two. It permits cultivation of strict anaerobes, such as the quail disease bacillus seems to be, without the aid of seals, special tubes or other complicated equipment.

It is difficult to secure material from intestinal lesions that is not contaminated with other bacteria which greatly increase the diffi-

2 Brewer, J. H., *J. Bact.*, 1940, **39**, 10.

† A powdered concentrate of the solids of this medium can be obtained from Baltimore Biological Laboratory, Baltimore, Md. To prepare, it is only necessary to dissolve the proper amount of the powder (and for our purpose, the additional agar) in distilled water, dispense in tubes and autoclave.

culty of isolating pure cultures from such material. On the other hand, lesions in the liver (which are found in more than 10% of diseased birds) usually have only the specific bacillus.

The necrotic plug in a very small lesion should be removed with teasing needles and inoculated directly into the medium. Good growth occurs after 24 to 48 hours' incubation and the pure culture can be maintained indefinitely if transplanted at least every 3 or 4 days. Cultures in the semi-solid medium should be transferred with capillary pipettes instead of the platinum loop.

(c) Quail disease can be transmitted by forced feeding of fresh droppings from diseased birds. On the other hand, dry or old droppings are innocuous. I have often observed coprophagy in birds crowded together in small pens. Several quail breeders have confirmed this observation from their own experience. It is likely that coprophilia, or unnatural tendency to eat feces, develops in such birds as a result of dietary or environmental faults or deficiencies associated with close confinement.

I have penned clean birds with carriers and have repeatedly observed that transmission often occurs if the pen is so constructed and arranged as to favor accessibility of their droppings to the birds. On the other hand, many times I have kept infected and susceptible birds together in the same pen without spread of the infection, provided the pen is so constructed as to prevent exposure of the droppings to the birds.

The disease spreads in cages with wire bottoms made of hardware cloth with spaces one-third inch or less, but does not if the mesh is one-half inch or larger; provided there is no other opportunity for droppings to be accessible to the birds.

When wide distribution has occurred in a quail stock, it takes from 6 to 10 months for all infected birds to die out and the disease to disappear, even under the above indicated precautions.

(d) Vaccination with killed cultures of the quail disease bacillus produces a high degree of immunity against experimental inoculation with live cultures. Exact dosage and methods have not been satisfactorily established. However, I have obtained adequate protection by giving to half grown birds, 2 intramuscular injections at 5-day intervals, of 0.5 cc vaccine made from 24- to 36-hour cultures in the above described semi-solid medium and killed with merthiolate, 1 to 10,000. Birds thus immunized survive feeding of several hundred times the infectious (and usually fatal) dose for non-immune birds.