

substance was effected by dilution of plasma with 10 volumes of water and acidification with CO_2 to pH 5.8 or with 1 per cent. acetic acid to pH 5.3. The precipitate was collected by centrifuging. After resuspension in 0.85 per cent. NaCl solution to the volume of the original plasma, this substance was as effective as its parent plasma in reducing the clotting time of hemophilic blood. When the precipitate was dried *in vacuo* at room temperature a brown-grey amorphous mass was obtained. Approximately 450 to 600 mgm were derived from 100 cc of citrated plasma. The dried substance was suspended in 0.85 per cent. NaCl solution and centrifuged. The clear supernatant fluid likewise was effective in reducing the clotting time of hemophilic blood both *in vitro* and *in vivo*.

When the above procedure was applied to hemophilic plasma, an approximately equivalent amount of the dried substance was produced, the saline suspension of which, however, had little if any ability to hasten the clotting of hemophilic blood.

Tested against a freshly prepared calcium-fibrinogen system without other coagulation factors, both the normal and hemophilic precipitates were equally active as prothrombin. Our observations seem to indicate that the difference between normal and hemophilic blood is due either to a qualitative difference of their prothrombins or to other substances probably associated with prothrombin. Studies leading to the identification of this substance or to the nature of the prothrombin modification are being made.

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THE ETIOLOGY OF ULCERATIVE ENTERITIS IN UPLAND GAME BIRDS

ULCERATIVE enteritis, or so-called "quail disease," first came to the attention of sportsmen early in 1900 through the exceedingly heavy losses encountered in importations of Mexican quail. The steadily increasing demand for quail for restocking game and shooting preserves has resulted in an increased number of game farms, with a consequent concentration of birds on limited areas. Such concentrations provide favorable conditions for the transmission of the disease, and the majority of game breeders are familiar with the devastating results associated with an outbreak. Although little is known about the prevalence of the disease in wild birds, observations on native ruffed grouse in Minnesota indicate that it may be of wide-spread occurrence.

Ulcerative enteritis is a highly infectious and rapidly fatal disease that affects most species of native upland game birds. Characteristic lentiform ulcers in the intestines are the principal lesions, and the infection

is readily transmitted by means of droppings. The average incubation period is about 4 days, though death may occur in 24 to 48 hours and before symptoms or lesions other than those of a toxemia have developed. A diagnosis of the disease can be established by feeding infective intestinal material to test birds. Ordinary cultural methods for isolation of the causative organism yield consistently negative results. A number of contaminating organisms, pathogenic on injection, have been isolated from diseased birds. Since natural infection, however, is contracted by the ingestion of infective material, the feeding test has remained the criterion in attempting to incriminate any organism as the causative agent.

Though a number of investigators, Morse,¹ Barger, Park and Graham,² Shillinger and Morley,³ Pickens, DeVolt and Shillinger,⁴ and Levine,⁵ have reported on the disease in quail and grouse, no conclusive evidence has thus far been advanced to establish the etiology.

The results of our work, as reported on February 6, 1936, to the North American Wildlife Conference, at Washington, D. C., record for the first time the isolation of an organism from the liver and spleen of diseased birds that is capable of producing death and typical lesions of ulcerative enteritis when fed in pure culture to susceptible quail. Eight strains of the organism, six from bobwhite quail, one from valley quail and one from ruffed grouse, have been studied. Only slight variations in the morphological and cultural characteristics have been noted, but it appears that there may be two or more serological types. The characteristics of the organism causing ulcerative enteritis in quail are as follows:

Corynebacterium perdicum, nov. sp.

Morphology: Primary growth in nutrient broth or agar, short thick rods 0.5 to 0.8 by 2 microns occurring in clumps. Non-motile. Gram-positive. In the tissues and older cultures the organisms are gram-variable. In secondary cultures the rods are of variable dimensions, straight or slightly curved, and show metachromatic granules. Some strains exhibit a marked tendency toward pleomorphism and under certain conditions produce branching filaments, cocci, and intermediary forms.

Cultural and physiological characters: Nutrient broth: Slow growth, with increasing cloudiness and formation of viscid sediment in older cultures. Agar media: No surface growth from tissues on agar media. Primary cultures can be started in semi-solid agar or broth and

¹ G. B. Morse, *U. S. Dept. Agr. Bur. Anim. Ind. Cir.* 109, 11 pp., 1907.

² E. H. Barger, S. E. Park and Robert Graham, *Jour. Am. Vet. Med. Assoc.* 84 (n.s. 37): 776.

³ J. E. Shillinger and L. C. Morley, *Jour. Am. Vet. Med. Assoc.*, 84 (n.s. 37): 25, 1934.

⁴ E. M. Pickens, H. M. DeVolt and J. E. Shillinger, *Maryland Cons.*, spring issue, p. 18, 1932.

⁵ P. P. Levine, *Amer. Game Conf.*, 19th, Trans., p. 437, 1932.

transplanted to plates or slants. Small circular, smooth, grayish white colonies appear in 72 hours from new cultures. When growth is established, the colonies are large, moist, and convex, with yellow pigment. The rough types are large white umbonated, leathery colonies with irregular margins that become powdery white on top in 48 hours and show no tendency to spread with age. Gelatin stab: Slight growth, no liquefaction. Potato: Good growth with yellow pigment. Loeffler's blood serum: Small, circular, colorless colonies, with slow liquefaction. Serum enhances growth of organism. Litmus milk: Unchanged. Kligler agar: Negative to slight hydrogen sulphide production. Indol test: No production in tryptophane broth. Nitrates: No reduction. Fermentation with cresol red and bromthymol blue, used as indicators: Acid produced in galactose, dextrose, levulose, maltose, sucrose, and salicin. Arabinose variable. Alkaline reaction produced in adonitol, dextrin, inulin, lactose, mannitol, raffinose, and xylose. Aerobic, facultative. Optimum hydrogen ion concentration of media: Ph. 7.6. Optimum temperature: 37° C.

Habitat: Isolated from liver, spleen, and heart blood of infected birds. Pathogenic for quail and grouse with the formation of intestinal ulcers when given per os.

The organism quickly loses its virulence following

isolation, and ordinarily most of the strains become non-virulent when growth is established on culture media. The symptoms, the sudden death and the character of the lesions associated with acute cases of the disease indicate that the organism produces a potent toxin, but thus far we have been unable to demonstrate toxin production.

Under conditions on game farms there is usually a sudden onset of an epizootic with a large percentage of the birds succumbing in a short time. The disease may entirely subside or it may be followed by an occasional case of chronic infection. It is as a rule difficult to transmit the disease with infective material from a chronically affected individual. Repeated transmission of the disease from one bird to another, under laboratory environment in every instance, has tended to decrease rather than increase the virulence of the infection. All attempts to increase the virulence under natural conditions have failed.

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VINEGAR AS A SOIL DISINFECTANT

ACETIC acid, like formaldehyde, is for some purposes an effective soil disinfectant.¹ Neither remains in the soil long enough to prevent all reinfestation of soil and resulting late infection of plants by fungi, but either is useful in protecting seedlings, of all but slowly germinating species, against damping-off. Applied to soil in too large amounts and not a long enough time before seeding, either may be toxic to plants. Any delay between soil treatment and seeding is considered objectionable by plantmen, and the relatively general use of formaldehyde dust is due as much to the fact that it may be safely applied to soil immediately before seeding as to its effectiveness.

An acetic acid dust, containing about 23 per cent. of acetic acid in a carrier of powdered wood charcoal, is apparently no less effective in controlling damping-off of seedlings caused by species of *Pythium* and *Rhizoctonia* and no less safe to plants. This has been applied to and thoroughly mixed with sandy soils, at the rate of 42 grams per square foot of soil surface. With seeds sowed immediately after this treatment of the soil, and with soil well watered at the time of seeding, acetic acid dust has had no harmful effect on growth of seedlings of *Centaurea Cyanus*, *Bellis perennis*, *Cerastium Biebersteinii*, *Lupinus polyphyllus*, Zinnia, China aster, *Laburnum alpinum*, cress

(*Lepidium sativum*), beet and five species of *Rhododendron*.

Neither acetic acid nor formaldehyde is always at hand when needed by the plantsman, and this is one principal reason why seeds are often sowed without any protection against soil fungi, for convenience or the lack of it is an important consideration in practice.

Cider vinegar, as usually sold, contains 4 to 5 per cent. acetic acid or not far above the minimum required by law. There are few substances more readily available or more generally at hand than vinegar, and the writer has found it reasonably effective in the protection of seedlings against some of the damping-off fungi and at the same time relatively or quite harmless to plants, even when applied to soil immediately before seeding.

Four brands of cider vinegar have been used. Although their acetic acid contents varied from 4.1 to 5.2 per cent., with an average of 4.3 per cent., they have not differed significantly in effectiveness.

Vinegar, without dilution, was applied to and thoroughly mixed with somewhat dry and definitely sandy soils. Soils used were either naturally infested with species of *Pythium* and *Rhizoctonia* or were, some days before treatment, inoculated with these fungi. Seeds were sowed immediately or within a few hours after soil treatment and the soils were then well watered.

The degrees of protection resulting from this treat-

¹ *Jour. Agr. Res.*, 44: 571-578, 1932.