spread weekly on the litter surface and sprinkled with water to keep moist. Larvae were collected and reared in the laboratory at room temperature.

The lesser mealworms were identified and confirmed by Commonwealth Institute of Entomology as Alphitobius diaperinus (Panzer) belonging to family (Tenebrionidae). Since its occurrence has not previously been reported, it is believed to be the first record from pigeon houses in the Sudan but never encountered in poultry pens which may indicate a sort of environmental preference of the parasite. Alphitobius diaperinus (Panzer) is commonly recognized as a prolific adaptable pest of concern to certain aspects of agriculture.

The male and female of A. diaperinus possess a pair of reversible posterior abdominal glands. On examining the adults of A. diaperinus two conical organs covered with yellow oily substance were observed protruding from the posterior margin of the last abdominal sternum. The oily secretion has a pungent irritating odour. Wilson and Miner⁶ described the glands as follows: "Adults of both sexes possess a pair of large scent glands that protruded when pressure was applied to the posterior region of the abdomen. The fluid obtained by puncturing the extruded gland produced a musky odour and caused sexual excitement in both sexes".

Although the constituents of the reservoir are unknown, Yeuh-Chu et al.⁷ isolated 2-methyl-1, 4-benzoquinone and 2-ethyl-1, 4 benzoquinone. Other constituents are unknown.

Back and Cotton¹ recorded A. diaperinus as a general pest of stored grain. Although it has not been encountered in poultry houses during this survey, Gould and Moses⁵ claim that it can be found in broiler chicken houses. However, Harding and Bissell³ noticed that the larvae burrowed into dead chicks. A. diaperinus drew considerable notoriety when it was incriminated as a carrier of acute avian leukosis by Eidson et al.². Irving and Bayona⁴ proved that both adults and larvae of Alphitobius lavigatus were predators of oriental rat flea Xynopsylla cheopis (Rothschild) and cat flea Ctencephalides felis (Bouche) in Puerto Rico.

During this work the life-cycle has not been completely studied. However, the average larval and pupal periods were found to be 13 and 5 days respectively.

The authors are thankful to the Under-Secretary. Ministry of Animal Production for permission to publish this note. Thanks are due to the Chief Veterinary Research Officer for keen interest. Dr. Abdel Rahim Hashim, Head of Poultry Division for co-operation and El Sheikh M. El Sheikh for technical supervision.

Veterinary Research ALI I. YAGI.

Division, M. T. ABDEL RAZIG.

Ministry of Animal

Production,

Khartoum, Sudan, April 1, 1972.

- 1. Back, E. A. and Cotton, R., UASD, FARM Bull., 1962.
- 2. Eidson, C. S., Schmittle, S. C., Goode, R. B. and Lai, J. B., J. Vet. Res., 1966, 27, 1053.
- 3. Harding, W. C. and Bissell, T. L., J. Econ. Entomol., 1958, 51, 112.
- 4 Irving, F. and Bayona, I. G., Ibid., 1968, 61, 877.
- 5. Gould, G. E. and Moses, H. E., *Itid.*, 1951, 44, 265.
- 6. Wilson, T. H. and Miner, F. O., J. Kans. Entomol. Soc., 1969, 42, 294.
- 7. Yeuh-Chu, L. T., Davidson, J. A. and Menzer, R. E., J. Econ. Entomol., 1971, 64, 425.

A NOTE ON THE PRODUCTION OF DIAGNOSTIC ANTIGEN FOR AVIAN MYCOPLASMOSIS

Mycoplasma organisms have been shown to be associated with chronic respiratory disease (CRD) of chickens and infectious sinusitis of turkeys. The serological diagnosis of mycoplasmosis in fowls in this country is so far conducted with the help of antigen imported from other countries. On the basis of serological testing the incidence of the disease is reported to be very high (Malik and Verma, 1969 and others). The fastidious nature and strict growth requirements of the organism is mainly responsible for its poor growth on ordinary laboratory media, thus making it difficult to prepare the diagnostic antigen. The non-availability of luxuriant growth of the organism is also responsible for lack of information on the serological types prevalent in the country. Present study was undertaken to develop a medium which could support luxuriant growth of Mycoplasma for different types of studies including the preparation of diagnostic antigen.

Standard S₆ strain of Mycoplasma galliscricum originally isolated by Zander (1961) from the brain of a turkey poult was employed using two basal media, namely, ox-heart infusion and Hank's balanced salt solution. Different nutrients were incorporated in the above two basal media in various combinations. In all 28 media were prepared and tested for growth both at 37° C and 40° C incubation temperatures. The constituents included in the study were five different preparations of Difco peptones (Bacto-peptone, proteose peptone, tryptone, casitone and trypticase), two preparations of oxoid

yeasts (yeast extract paste and yeast autolysate-powder), thiamine hydrochloride, cystein hydrochloride, glucose and serum from ox, horse, pig and chicken. The growth was harvested at different incubation periods.

One hundred ml of each medium in 500 ml Erlenmeyer flasks was inoculated with 5 ml of 6 days old culture of M. gallisepticum strain S_6 . One set of inoculated flasks was incubated at 37° C and the other at 40° C for a period of 144 hours and growth determined by OD method with Bijou colorimeter at 625 m μ wavelength.

The optical density (OD \times 100) of cultures in various media at different incubation temperatures and periods varied from 2.5 to 13.1. The maximum OD was obtained in ox-heart infusion broth consisting of casitone 1%, sodium chloride 0.5%, horse serum 10%, yeast autolysate 2%, thallium acetate 1:3000 and penicillin 1000 units/ml both at 37°C and 40°C. Although growth at 40°C started earlier, there was no appreciable difference in growth at the end of 144 hours of incubation. In some cases, on prolonged incubation OD was found to reduce perhaps due to autolysis of the microorganisms.

The growth in the ox-heart infusion broth at the end of 144 hours of incubation was harvested by centrifugation at 12000 RPM for 30 minutes in ZENTZEKI Model K24 centrifuge at 0°C. The sediment was suspended in 0.5% carbol saline and recentrifuged at the same speed and time. In this way the washing of the cells was repeated three times before diluting the culture in carbol saline matching with Brown's opacity tube No. 4 thus making a total suspension of about 30 ml per litre of liquid culture. The suspension was then homogenized for 5 minutes at approximately 5000 RPM, in 'Metronex' universal laboratory homogenizer model 30. The homogenized cell suspension was used as diagnostic plate antigen and results compared with the antigen imported from Commonwealth Serum Laboratories, Melbourne, Australia. In all 305 individual or pooled poultry serum samples were tested. Out of these, 282 samples gave positive agglutination reaction with imported antigen and 285 with locally prepared antigen. All reactions correlated with each other except in 3 cases where imported antigen gave negative reactions while locally prepared antigen showed weakly positive reaction.

The above studies have given very encouraging results and have paved the way to prepare mycoplasma diagnostic antigen locally.

The authors are grateful to Dr. R. L. Kaushal, Dean, for his keen interest in the work.

Department of Microbiology, College of Veterinary Science and Animal Husbandry, Jabalpur, April 6, 1972.

B. S. Malik. N. B. Singh.

M. C. S. PARIHAR*.

N. S. DHANESAR.

* Present address: Veterinary Officer, Military Dairy Farm Jabalour.

1. Malik, B. S. and Verma, K. C., Avian Dis., 1969, 13, 695.

2. Zander, D. V., Ibid., 1961, 5, 154.

MUTAGENIC EFFECT OF UV AND GAMMA-RAYS ON ANTIBIOTIC PRODUCTION OF ASPERGILLUS CHEVALIERI

It is well established that mutagenic action exerts definite effect on the biosynthesis of metabolic products in micro-organisms. Savage¹, Dulaney et al.² and Chaudhury and Chakrabarty³ obtained mutants of Streptomyces species through irradiation which gave higher yield of antibiotic than the original culture. Mutations resulting in the increased antibiotic production by fungi have been reported by Demerec⁴ and Hollaender⁵. A strain of Aspergillus chevalieri (Mangin) which produced an antibiotic⁶ was selected for the study of the effect of UV and γ -rays on the survival and mutation frequency in terms of antibiotic production.

The spore suspension of A. chevalieri in Ringer's solution was irradiated with UV rays7. For γ-irradiation the spore suspension was kept in screw capped aluminium capsules and subjected to the rays, the doses being 10, 20, 30 and 40 Krad (energy 6.141 Krad/min.). The irradiated spore suspensions were plated in complete medium⁸ (CM), incubated at 28-30° C and the developed colonies were transferred to minimal agar⁹ (MM) plates. Biochemical and morphological mutants were determined according to Pontecorvo¹⁰ method. Control plates in triplicates were kept in similar conditions. Antibiotic production was measured by the agar cup method of assay against Escherichia coli and the percentages of mutants with affected or unaffected antibiotic productions were caluclated.

Increase in antibiotic production was noticed in a number of mutants (Table I). Of these two mutants were selected for further studies. Mutant UV-14 was deep yellow with medium sporulation at the margin while mutant Gm-12 was bluish green with heavy sporulation at the centre of the colony. A comparison of the antibacterial properties of the antibiotics produced by these two mutants and the parent strain showed wide differences. It was found that while the parent strain showed no inhibition against Bacillus cereus,