months of the year. Available evidence suggests that significant increases in neutralizing antibodies to BA types 1, 2 and 3 occur in the sera of cattle involved in respiratory disease outbreaks in Britain[6] and that it is also possible that further, undefined serotypes of bovine adenoviruses may be involved.

J. H. Darbyshire

Central Veterinary Laboratory,
Weybridge.

A. R. Jennings
A. R. Omar

School of Veterinary Medicine,
University of Cambridge.

P. S. Dawson
P. H. Lamont

Central Veterinary Laboratory,
Weybridge.

[1] Darbyshire, J. H., Dawson, P. S., Lamont, P. H., Ostler, D. G., and Pereira, H. G., *J. Comp. Path. Ther.*, **75**, 327 (1965).
[2] Pereira, H. G., *Virology*, **6**, 601 (1958).
[3] Darbyshire, J. H., Jennings, A. R., Dawson, P. S., Lamont, P. H., and Omar, A. R., *Res. Vet. Sci.* (in the press).
[4] Darbyshire, J. H., and Pereira, H. G., *Nature*, **201**, 895 (1964).
[5] Darbyshire, J. H. (unpublished results).
[6] Darbyshire, J. H., and Pereira, H. G. (unpublished results).

Hepatic Tumours in Ducks fed a Low Level of Toxic Groundnut Meal

The toxicity of certain samples of groundnut meal has been shown to be due to fungal metabolites, collectively known as aflatoxin, produced by strains of *Aspergillus flavus*[1]. The carcinogenic effect of such meals for rats, a species relatively resistant to the acute effect of aflatoxin, has been demonstrated[2–4] and it has since been shown that crystalline aflatoxin is carcinogenic for these animals[5,6].

Young ducklings are the most susceptible animals to the acute effect of aflatoxin[7]. This communication describes the change found in ducks fed for a prolonged period on a ration containing a low level of toxic groundnut meal.

Thirty-seven 7-days-old Khaki Campbell ducklings (group A) were fed a commercial poultry ration free of aflatoxin to which was added 0·5 per cent toxic Brazilian groundnut meal which contained approximately 7 p.p.m. aflatoxin (assayed as aflatoxin B_1). Sixteen control ducklings (group B) of the same age and breed were fed the commercial ration without added groundnut meal. All birds received tap-water *ad lib*. They were brooded for the first three weeks in electrical tier brooders and thereafter housed in arks on pasture. Weights were recorded weekly for the first 14 weeks of the experiment. Deaths in both groups were recorded and tissues retained for histological examination. The survivors in both groups were killed fourteen months after commencement of the feeding trial.

Tissues were fixed in formal-saline, embedded in paraffin wax, sectioned at 7μ and stained with haematoxylin and eosin. Liver sections were, in addition, stained with Masson's trichrome stain and frozen sections were prepared and stained with oil red O.

Plotting the mean weight of the birds in the two groups against time showed no significant differences in the slope of the two regression lines, but the differences between means for the groups were significant at week 3 (group A 11·6 oz., group B 15·2 oz. ($P < 0.05$)), week 4 (group A 16·0 oz., group B 22·3 oz. ($P < 0.01$)) and week 5 (group A 23·5 oz., group B 27·9 oz. ($P < 0.05$)) but not at any other time to week 11. From weeks 11 to 14 there was no longer a linear weight increase in either group; the controls remained at a higher mean level than the treated birds and this difference was highly significant ($P < 0.01$) on mean values alone.

During the first four weeks of the experimental period nineteen birds in group A died. On histological examination lesions consistent with poisoning due to aflatoxin were present in the liver of all of them although in three birds death was ascribed to other causes. In the following 7 months a further seven birds in group A died, or were killed, and in each of these hepatic lesions consistent with aflatoxicosis were present.

In group B four ducks died due to bacterial diseases and two were killed because of severe trauma inflicted by their pen-mates. On histological examinations of the livers from these birds there was no evidence of aflatoxicosis.

Fourteen months after commencement the survivors of both groups, consisting of eleven in group A and ten in group B, were killed. Macroscopic lesions were confined to the livers; those from birds in group A were atrophied, putty coloured and contained numerous yellowish focal lesions 1–2 mm diameter. In four of the six drakes there were, in addition, solid yellow-coloured nodules varying in size from 1 to 3 cm diameter. In four of the five ducks, in addition to the small focal lesions, there were nodules in the livers between 0·5 cm and 2·0 cm in diameter. In group B, comprising six drakes and four ducks, one drake had a number of small irregularly-shaped necrotic lesions in the liver. Apart from this there were no other macroscopic lesions in the organs of the birds in this group.

The small focal lesions present in the livers of all birds in group A were lymphoid foci composed mainly of mature lymphocytes surrounding degenerating hyperplastic bile duct epithelium. There were no mitotic figures but, peripherally, there was infiltration, in some cases extensive, of lymphocytes between the hepatic cells. The larger nodules were liver tumours of two types: thickly encapsulated, vacuolated hepatomata composed of hypertrophic ballooned cells occasionally containing giant vesicular nuclei with very prominent nucleoli, and cholangiomata composed of dense masses of well-differentiated bile ducts (the cells of which showed very few mitotic figures) surrounded by a thin fibrous capsule. This variation in cell type is seen in hepatic tumours in rats fed aflatoxin and other carcinogens[5]. In the liver of one duck tumours of both types were present, in three ducks and two drakes there were hepatomata, and in two of the other drakes cholangiomata occurred.

After 14 months eight of eleven birds fed a ration containing 0·5 per cent toxic groundnut meal developed hepatic tumours whereas none occurred in ten birds fed the same ration without groundnut meal. The aflatoxin B_1 content of the tumour-inducing ration for ducks was approximately 0·03 p.p.m. compared with 0·4 p.p.m. required to produce hepatic tumours in rats within 13–18 months[5]. The single oral LD_{50} dose of aflatoxin for male rats is 7·2 mg/kg[8] compared with 0·4 mg/kg for male ducklings[9]. The relative susceptibility of ducks and rats to both the acute and hepato-carcinogenic effect of aflatoxin is therefore comparable.

I thank Dr. J. G. Campbell of the British Empire Cancer Campaign for Research, Edinburgh, for his advice and Miss C. N. Hebert of this Laboratory for statistical analysis.

R. B. A. Carnaghan

Central Veterinary Laboratory,
Weybridge.

[1] Sargeant, K., Sheridan, A., O'Kelly, J., and Carnaghan, R. B. A., *Nature*, **192**, 1096 (1961).
[2] Lancaster, M. C., Jenkins, F. P., and Philp, J. McL., *Nature*, **192**, 1095 (1961).
[3] Schoental, R., *Brit. J. Cancer*, **15**, 812 (1961).
[4] Le Breton, E., Frayssinet, C., and Boy, J., *C.R. Acad. Sci., Paris*, **225**, 784 (1962).
[5] Butler, W. H., and Barnes, J. M., *Brit. J. Cancer*, **17**, 699 (1963).
[6] Dickins, F., and Jones, H. E. H., *Brit. J. Cancer*, **17**, 691 (1963).
[7] Asplin, F. D., and Carnaghan, R. B. A., *Vet. Rec.*, **73**, 1215 (1961).
[8] Butler, W. H., *Brit. J. Cancer*, **18**, 756 (1964).
[9] Carnaghan, R. B. A., Hartley, R. D., and O'Kelly, J., *Nature*, **200**, 1101 (1963).