Investigations of the Pathology and Chemotherapy of Infectious Enterohepatitis (Blackhead) in Turkeys

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In 1967 considerable economic losses, of the order of several million Forints were caused by infectious enterohepatitis in turkey flocks. This was partly due to inadequate knowledge of effective control measures, but above all to lack of an efficient specific drug such as has been widely applied in other countries where turkeys are bred on a large scale. During the first half of 1967 the sole available chemotherapeutic in this country was furazolidone (Tikofuran) which has been found in our country to have only weak antiblackhead action.

During the second half of the year a large amount of 2-amino-5-nitrothiazole, widely used against blackhead, was imported. A field trial of this drug in the form of the premix Hepamix which contains it at a concentration of 10% was carried out in nearly 300,000 turkeys and provided abundant material for investigation. This communication comprises a review of relevant observations which both confirm and complement the known facts about blackhead.

1.) Gross lesions in the liver. In most textbooks the sites of gross lesions of blackhead are given as the caeca, caecal walls, peritoneum adjoining the caeca, and liver. In practice, however, exclusively caecal lesions have often

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been found on post-mortem examination, liver lesions being either absent or recognized only with difficulty. It is, therefore, not particularly surprising that because of their similarity to caecal coccidiosis of chickens, the caecal lesions have frequently been confused with coccidiosis.

Fig. 1. Turkey liver with lesions characteristic of blackhead from the material designated "Tiszaroff". This liver is about 1/3 original size.

Jellegzetes blackheades elváltozásokat feltüntető pulyka-máj a "Tiszaroff" elnevezésű anyagból. Az eredeti nagyságnak kb. az egyharamda.

The material designated "Tiszaroff" provides a good example of the inexplicable alternating incidence of lesions involving either the caeca alone or both caeca and liver. The turkey flock of a collective farm, totalling approximately 15,000 birds, was housed in two premises several kilometres apart. Conditions of management and feeding were similar in the two places and medication with Tikofuran against blackhead was carried out to an identical schedule. The outbreak of the disease occurred simultaneously in both premises with comparable morbidity. Yet
necropsy material from one place showed, in every case, distinctive lesions of both caeca and liver, with typical concentric hepatic foci, whereas in material from the other farm characteristic caecal lesions, enlargement of the liver with indistinct discolorations, but no focal lesions were found.

Several transitional forms were found to exist between hepatic changes that macroscopically were scarcely discernible and the focal lesions considered pathognomic of blackhead. Enlargement of the liver was a typical finding in turkeys that died or were killed at the height of infection. In survivors regression of the liver to normal size took place over several weeks. On the enlarged liver the least conspicuous lesions were poorly demarcated discolorations of varying size, paler in colour than normal liver tissue, and seen only at high illumination (in sunshine) after the liver surface had been smoothed.

In other — supposedly more chronic — cases the considerably enlarged liver showed round foci of coin size slightly depressed beneath the surface of the organ, yellowish or greenish in colour and of concentric pattern (Fig. 1). In some cases these foci penetrated deeply into the parenchyma. Such lesions are not readily confused with those due to other diseases.

The third type of lesion was encountered in the livers of turkeys that had recovered from blackhead, the concentric foci described above having been replaced by cicatricial tissue.

Regeneration of the necrotic areas by cicatrization produced sharply contoured discoloured spots on the surface of the liver. The cicatricial tissue was paler than the surrounding hepatic tissue, yet the more advanced the process of regeneration the less marked was the difference. Since, however, regeneration of liver cells was incomplete, the larger areas of scar tissue imparted a spotted appearance to the liver throughout the lifetime of the bird.

2.) Microscopic lesions. Histological examinations were performed on a total of 50 turkey livers, partly to
identify by morphological and staining properties the animal, (Histomonas) or plant (Candida), origin of the etiological agent, and partly to examine the histopathology of livers with various lesions. For this purpose, paraffin embedded sections were stained with hematoxylin and eosin, Mallory’s stain and periodic acid-Schiff (PAS).

The initial hypothesis assumed that the two morphologically distinct lesions were due to two different etiological agents of blackhead. This assumption was supported by findings in the liver (designation "Boldogasszonyfa") of a turkey which on postmortem examination showed typical caecal lesions, but only enlargement and diffuse discoloration of the liver. Microscopic examination of this liver revealed large numbers of conspicuous round homogenous bodies (Figs. 2a, 2b), 7-10 μ in diameter, staining pale pink with eosin, vivid red with Mallory’s stain and deep lilac with PAS. The parasites, clustered densely together, occupied the spaces between the rows of liver cells, sometimes giving the impression of being intracellular in location. A similar intracellular position was previously described by CHESTER and ROBIN (1900) although later TYZZER (1920) advocated the intercellular location of the parasites. In the particular case being described only a few parasites other than those mentioned above were found; they also stained pale pink but showed an internal structure.

Fig. 2a. Causative agents of blackhead in a section from turkey liver. a: The round structureless bodies resemble fungi. Hematoxylin and eosin. X 300

Fig. 2b. Magnified part of Fig. 2. PAS. X 1000

Fig. 3a. Causative agents of blackhead in turkey liver. a: Bodies resembling to Histomonas. Hematoxylin and eosin. X 300

Fig. 3b. Magnified part of Fig. 3a. Hematoxylin and eosin. x 800
Their shape was generally round, but often ovoid or ameboid.

In later studies on both types of liver lesion one or other parasite was noted without apparent relationship to the type of lesion.

The macroscopically distinguishable liver lesions showed no corresponding variation of microscopic structure. In the liver parenchyma necrotic foci with karyolysis and subacute inflammation of the adjoining tissue were found. The foci did not become encapsulated by fibrous tissue; only a slight increase of connective tissue being noted in inflammatory areas around the blood vessels and biliary ducts. Besides infiltrating lymphocytes and mononuclear cells these inflamed areas contained many eosinophilic and pseudoeosinophilic cells. Granulocytes and numerous giant cells were also found. In the affected areas the lumina of blood vessels and sinusoids were distended with red blood cells (Fig. 5).

In the present examinations the parasites considered to be the etiological agent of blackhead were of two morphologically distinct types. They occurred either singly or in groups (nests), most of them intercellularly. One type, a round structureless body of 7-11 μ diameter, stained vivid red with Mallory's stain. Staining positively with PAS, it was very likely a fungus. The other type, staining pale pink with eosine, appeared as single or multiple roundish bodies surrounded by a clear ring (Figs. 3a, 3b, 4). Mallory's stain imparted to them an uncertain greyish hue. They were larger than the fungus-like organisms, of slightly ovoid shape and some had diameters above 40 μ. Occasionally they were seen in an apparently intracellular position, adjacent to the enlarged and flattened nucleus of the host cell. Some of the parasites showed one or more tiny bright ruby-red granules, surrounded by a clear halo, situated close to the nucleus. The occurrence of such granules seemed to be associated with division of the parasite. Single granules were usually seen close to the nucleus, whereas when several granules were present they were distributed in the cytoplasm, sometimes with obvious signs of cell division around them. In larger parasites the clear halo was more
distinct and a thin filament was seen to connect its border with the body of the parasite. The morphological appearance and particularly the filament suggested the parasite to be a *Histomonas meleagridis*.

In healed lesions the liver cells were arranged more densely and their nuclei stained darker with hematoxylin than did healthy liver tissue (Fig. 6). In addition, a few residual granulocytes and structures resembling the parasites were encountered in paler areas where the liver trabeculae had been replaced by disorderly aggregates of liver cells.

Thus histological examination failed to provide evidence as to the precise nature of the etiological agent of blackhead. The PAS-positive structureless organisms were probably fungi, whereas the pale pink structured organisms closely resembled *H. meleagridis*. It seems feasible, therefore, that both organisms and, according to more recent findings, even certain bacteria, play a role in the causation of blackhead (TALL and FRANKER, 1963).

3.) Action of *Hepamix*. Mixed in the diet of turkey flocks severely affected with blackhead (10-20% losses), 1% *Hepamix* feed additive stopped the outbreak in 5 days. Later deaths were mostly due to intercurrent diseases.

Given in the initial stage of the outbreak, treatment with *Hepamix* minimized losses. Naturally, early medication requires early correct diagnosis. The experienced veterinarian, and even technical assistants can recognize blackhead by the clinical symptoms prior to occurrence of losses. Diseased birds lose

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**Fig. 4.** Histomonas-like parasites in the liver of a turkey affected with blackhead. Adjacent to the nucleus rubin granules are apparent. Oilimmersion. X 1500

Histomonas-szerű élősködők blackheads pulyka májában. A mag mellett a rubinszemcsék is feltűnnek. Immerziós felvétel. 1500 X
appetite, are sleepy, lag behind the rest and have diarrhoea, the yellowish pasty droppings of sick turkeys being immediately noticed. In such circumstances blackhead is easily diagnosed by postmortem examination of the most ailing birds.

Delayed treatment for blackhead is often due to erroneous diagnosis. Considerable distention of the caeca of affected birds and caseous-fibrinous plugs in the caecal lumina superficially resemble lesions encountered in the caecal coccidiosis of chickens. In fact, like most domesticated animals, the turkey has several coccidian parasites. Yet oocysts are always demonstrable in the droppings, often even in the pasty stool seen during the early phase of blackhead. Although the presence of oocysts apparently supports the diagnosis of caecal coccidiosis no turkey coccidium is known to cause such extensive caecal lesions.

Good durable therapeutic results can be obtained if 1 % Hepamix is given in the diet for 7-10 days. Subsequently treatment must not be stopped but continued at half the dose rate for at least as long a time. As indicated in the literature, the disease is liable to recur even then, indicating that the immune response to blackhead is very low (for ref. see BIESTER and SCHWARTE, 1965).

4.) Prophylaxis of blackhead. In the spread of blackhead the most important role has been attributed to the eggs of Heterakis gallinarum. The etiological agents of blackhead are carried either by these eggs, or by the larvae hatched from them, through the wall of the caecum and invade the host. Therefore, turkeys should be housed and managed so that they remain free of infection with Heterakis eggs. In this respect, Heterakis eggs shed with chicken droppings seem to be even more dangerous than those excreted by the turkeys themselves. It is known that blackhead also occurs in gallinaceous birds, but affects them only slightly or not at all. Nevertheless, the Heterakis eggs shed with the droppings of gallinaceous birds may infect relatively large areas for long periods of time.

The first and most important rule of blackhead prophylaxis is to prevent all contact of the turkeys with Heterakis ova. These pa-
Rasite eggs may remain viable in the soil for as long as 2 years although the ovum-transmitted agent of blackhead deteriorates in a few months. Therefore, care should be taken to place the turkey flock in "virgin" premises, where neither turkeys nor chickens have been kept for the preceding two years, or at least one. Experience with material designated "Orosháza" emphasise the importance of this particular requirement. Within a given area, containing farms not far distant from each other, blackhead occurred only where chickens and turkeys had been kept during the preceding year. Farms where this was not the case remained free of blackhead.

Another method of preventing blackhead is to rear turkey poult's up to 3-4 weeks' age in isolation on a deep litter not infected with Heterakis eggs.

If subsequent exposure is inevitable, losses can be minimized by starting preventive medication at the proper time.

Summary

A field trial of the feed additive H e p a m i x, containing 2-amino-5-nitrothiazole at a concentration of 10 %, was carried out.

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Fig. 5. Microscopic lesions in the liver of a turkey affected with blackhead. Inflammatory foci containing detritus, granulocytes and fungus-like parasites. Hematoxylin and eosin. X 150

Fig. 6. Microscopic picture of liver in the convalescent stage after blackhead. Note healthy liver in the centre. Hematoxylin and eosin. X 150

A blackhead gyógyuló esete, a középen az érintetlen májsszövet. Hem-eozin. 150 X
on nearly 300,000 turkeys. Detailed examination of gross and microscopic lesions of the liver indicate that in the etiology of blackhead both presumed causative agents, viz. Histomonas meleagridis and Candida albicans, are implicated. Hepamix proved to be very efficient against blackhead; when mixed in the diet at a concentration of 1%, it stopped the outbreak in 3-5 days. Practical experience suggests that planned prevention of blackhead remains to be established.

PELLÉRDY, L.: Patológiás és terápiás vizsgálatok fertőző enterohepatitisben beteg (blackheads) pulykaállományokban

A blackhead Magyarországon 1967-ben több millió Ft kárt okozott. Ennek az volt az oka, hogy egyrészt nem rendelkeztünk kellő tapasztalattal a betegségnek időben történő felismeréséhez, másrészt pedig nem volt birtokunkban blackhead ellen biztos hatású gyógyszer. Amikor 2-amino-5-nitrotiazol-t nagyobb mennyiségben importáltunk és azt a Phylaxia Hepamix nevű takarmánygyógyszerkeverékben elkészítette, alkalmam nyilott kórbonctani és kórszövettani megfigyelésekre végzésekre.

A kórbonctani elváltozások számos esetben csak a vakbelekben voltak feltűnők, azonban a máj mindig megmagyarázatlanul volt és az erős megvilágítással (napon) a máj lesimitása után foltos színűüléseket lehetett találni az egyes területek között. A blackheadre típusos májelváltozásokon (1. kép) kívül a gyógyszer esetekben a májénál jelentősen színű gócos elváltozásokat találtunk, amelyek mindenek szerint regenerálódó hegszövetek feleslegesek voltak. Ez a szövettani vizsgálatok is igazolták. Jóllehet a blackhead két feltételezett kórokozójának, a Histomonas meleagridisnek és a Candida albicansnak a májában jelentősen kiterjedt változásai voltak, a szövettani vizsgálatok alapján arra az eredményre kellett jutnom, hogy a blackhead előidézésében a C. albicansnak (2a, 2b. kép) és a H. meleagridisnek (3a, 3b, 4. kép) egyaránt szerepe lehet. A betegség okozói és megállapítására további vizsgálatok vannak folyamatban.
A Hepamix kitűnő hatású gyógykeverék. Közel 300,000 pulykára kiterjedő megfigyelésem megerősítette, hogy a Hepamix-nak 1 %-ban több napig történő etetése után a blackhead 3-5 napon belül megszűnik. A biztos siker alapja a gyógyszer alkalmazásánál a mielőbbi és helyes diagnózis. Pulykában a vakbél-elváltozások nem téveszthatók össze a csirkék vakbélcoccidiosisa során észlelhető elváltozásokkal, de egyébként sincs a pulykának olyan coccidiuma, amely az Eimeria tenella-hoz hasonló patogén hatású volna. A blackhead elleni védekezés és a gyógyszer sikeres alkalmazásának előfeltétele az, hogy kellő időben avatkozzunk be és hogy a tervszerű megelőzésre nagy súlyt helyezzünk. Közismert tény, hogy a blackheadet a vakbélférgek (Heterakis gallinarum) terjesztik oly módon, hogy petéiket a pulykák felszedik. A Heterakis-ok ferfőzött pulykából származhatnak, de a tyúkban élő Heterakis-ok is a blackhead közvetítői lehetnek. Fontos tehát, hogy az állatokat biztosan Heterakis-tól mentes környezetben neveljük.

References


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