PATHOMORPHOLOGICAL CHANGES INDUCED BY VARIOUS MANAGEMENT CONDITIONS IN ARCTIC FOXES (*ALOPEX LAGOPUS*)

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Abstract

The studies aimed to evaluate the pathomorphological changes induced by various management conditions in arctic foxes. Healthy pups aged about eight weeks were randomly assigned to two groups, 20 animals each. The foxes of the control group were housed on a farm, while the experimental group comprised of foxes raised in a confined space. Throughout the rearing period, air quality monitoring of the environment of both groups was performed. During the autumn slaughter (the experiment lasted for 7 months), sections of the liver, kidneys, lungs, ovaries, and uterus obtained from all the foxes were evaluated pathomorphologically. Pathological changes were observed in lung sections collected from the animals of the experimental group. During the air monitoring, higher concentrations of pollutants were identified in the experimental group environment.

Key words: arctic fox, air pollution, pathomorphological analysis.

Recently, a number of EU-based fur sector enterprises, including fur animal farms have been brought to Poland, which induced the development of fur farming in this country. A pressure of numerous ecologic organisations and an extensive public debate imposed the improvement of animal management conditions so that fur farming system could ensure a high standard of animal welfare and an appropriate “odour image” maintained around the farm. The intensive-confinement breeding increases potential animal exposure to hazardous chemical or biological agents. The exposure may implicate adverse progressive involvement of animal body systems and compromise their function that lead to lowered breeding and utility value and as a direct result, serious economic losses (15, 18). So far, this problem has not been addressed in full; therefore, the studies were performed to assess the impact of gaseous air pollutants released in the animal farm environment on polar fox organism.

Material and Methods

The investigations were carried out on the polar fox (*Alopex lagopus*) farm where ca 50 animals of the basic stock were housed during the research. The control (K) comprised 20 pups reared in the conventional pavilion system. The experimental group (D) included 20 pups raised in the confined chamber with natural ventilation, where air movement did not exceed 0.1m/s. Throughout the study period (May-December), air quality in the environment of both groups was monitored with chromatographic methods once a month. A percentage of the identified groups of gaseous contaminants are compared below (17).

The foxes were fed the same rations in compliance with the feeding standards for fur animals satisfying the nutritional needs of the animals according to their age (4). A periodical clinical check-up and daily animal inspection did not show any deviation from the standard of good health. However, a change in the management conditions likely decreased the final value of the foxes. In late December, the animals from both groups were slaughtered and the carcasses underwent the post mortem examinations. The sections of the liver, kidneys, lungs, ovaries, and uterine horns were collected for histopathological analysis. The tissue material was fixed in 10% neutral buffered formalin for 24 h, then the tissues were processed on automatic tissue processor (Leica TP-1020) for 24 h, and embedded in paraffin blocks. The obtained sections were stained with haematoxylin and eosin. The additional frozen sections of the liver and kidneys were stained according to Daddi`s Sudan IV stain for neutral fats. Parts of the liver
samples were fixed in an absolute alcohol and paraffin sections stained with PAS method (21) were prepared.

Results

The pathological lesions were detected in the lung sections collected from the experimental group foxes. Macroscopically, the animals exhibited light red and foamy lungs, whereas microscopically, besides the normally developed alveoli, there were visible numerous deformed alveoli of irregular shapes. A large number of air-vesicles were markedly distended, their walls lost their elasticity and occasionally ruptured forming the sinusal spaces towards the lumen with projected pointed stumps of thinned interalveolar septa. The image exhibited the above mentioned numerous large air spaces formed by a coalition of air cells after atrophy of the alveolar septa and walls (Fig. 1).

Besides, the lumen of bronchioli of these animals was filled with substantial amount of mucus together with desquamated cells of the respiratory epithelium. All the macro and microscopic changes suggested alveolar emphysema condition (Fig. 2). Emphysematous foci were shown to occur uniformly in the pulmonary lobules with extensive portions of the lung parenchyma involved.

Fig. 1. Large spaces formed by atrophy of alveolar walls. H.E., ca 240x (experimental group)

Fig. 2. Pulmonary emphysema with noticeable bronchiolus and its lumen filled with mucus and desquamated epithelial cells. H.E., ca 120x (experimental group)

Fig. 3. Droplet deposits of sudanophilic bodies in the cytoplasm of few hepatocytes. Sudan IV + H., ca 120x (experimental group)

Fig. 4. Liver congestion. Ample erythrocytes in the central vein lumen and dilated sinusoids. H.E., 120x (experimental group)

Fig. 5. Granular reaction to glycogen in hepatocytes. (PAS), ca 320x (experimental group)

Fig. 6. Droplet deposits of sudanophilic bodies in the cytoplasm of epithelial cells of renal tubules. Sudan IV. ca 240x (experimental group)
Microscopically, the liver presented a slightly differentiated morphological structure for both groups. In the majority of the animals, the liver tissue sections appeared to have the morphological structure of hepatic lobules similar to normal ones. The trabeculae-forming hepatocytes were arranged radially showing the normal convergence towards the central veins. The cytoplasm of hepatocytes contained basophilic granules and only few cells manifested some signs of acidophilic degeneration and small vacuoles that after Sudan staining proved to be of the lipid nature (Fig. 3).

Between hepatocytes, a moderate number of Browicz-Kupffer cells (stellate cells) were noted. In a part of the experimental foxes, there was detected the blurred liver cell trabecula arrangement as well as some signs of congestion manifested by an excessive number of erythrocytes in the central vein lumen and dilated sinusoids. In hepatocytes, more abundant lipid vacuoles were observed (Fig. 4).

Although the presented cases were characterised with an increased fat content, no changes of fat degeneration were determined in hepatocytes, that indicated the enhanced lipid metabolism. Glycogen present in all the sections occurred as fuchsinophilic granules that filled uniformly the whole plasma of most cells (Fig. 5). The reaction of the same intensity was developed across the whole hepatic lobule. However, especially in the experimental group, cells of the lobule with an elevated fat level gave a weaker reaction for glycogen.

Microscopic study of the kidney sections from both groups presented normal architecture of the cortical and medullary layers. There were found plentiful well-developed renal glomeruli with adjacent corresponding fragments of the nephron. The proximal tubules were lined by cubic epithelium with the brush border. In the cytoplasm of epithelial cells of tubule segments, there was shown the presence of ample small vacuoles that gave a colour reaction with Sudan IV (Fig. 6). The other elements of the secretory and excretory part of renal tubules exhibited the normal structure. The other analysed organs (ovary, uterus) did not exhibit any pathomorphological changes.

During air monitoring in the K and D groups, there were identified substances whose concentration reached the following values: sulphur compounds: K – 10.98 µg/m³, D - 79.62 µg/m³; alcohols and aldehydes: K – 193.68 µg/m³, D - 340.92 µg/m³; hydrocarbons: K – 128.94 µg/m³, D – 171.98 µg/m³; and nitrogen compounds: K – 1,820 µg/m³, D – 2,390 µg/m³. A percentage of each compound is summarised in Fig. 7. The highest content in all the groups of the identified substances was determined for the nitrogen compounds, with dominant ammonia. As for alcohols and aldehydes, the presence of pentanoles, butanoles, and acrolein was established, whereas out of the hydrocarbon group – benzene, ethylbenzene, toluen, xylenes, naphthalene, 3-karen were detected.

**Discussion**

The present researches have revealed the most advanced morphological lesions in the pulmonary tissue, which occurred as pulmonary emphysema. Aetiopathogenesis of emphysema has not been
explained explicitly yet, still some pathogenetic mechanisms are known to be primary contributors to this degenerative condition. Expiratory airflow obstructions cause the distension of the peripheral lung tissue, from a site of increased resistance to airflow. Under normal conditions, bronchioli without a cartilaginous ring are kept open at the expiration by the elasticity of their walls and adhesion to air-filled alveoli. The inflammatory lesions in small peripheral bronchi may develop emphysema due to the obliteration of the bronchioli (19).

Another approach to emphysema aetiopathogenesis implies that all the processes resulting in the accumulation and enhanced activity of leukocytes and pulmonary macrophages in alveoli, bronchioli, and interstitial tissue may be major aetiological factors in this disease development (9, 19). Estimation into the potential effects of human exposure to gaseous pollutants emitted to the air has remained a challenge and is usually based on some damage events reported in industry (6). In humans, most data refer to harmful effect of tobacco smoke, including large number of free radicals (13, 19). Airborne gaseous pollutants that penetrate the respiratory system may also induce the pathological processes leading to emphysema (6, 19).

Out of air contaminants released in the chamber, the highest concentrations of nitrogen compounds, particularly ammonia, were detected. The intensity of injury at ammonia exposure is proportional to its air concentration and corresponds to the microclimatic parameters (6, 14, 20).

Effects of acrolein exposure were most thoroughly analysed in the experimental animals, which inhaled it with the air. Mice and rats developed the changes in the respiratory system: inflammation, ulceration, necrosis; the intensity of changes increased with the rise of an acrolein concentration. In rabbits and hamsters however, the changes appeared to be slightly different, that is associated with some species differences. No impact of inhaled acrolein on animal reproduction was reported (11, 12, 14). Lung response occurring after inhaled ethylbenzene exposure is markedly visualised in a lung image as 40-60% of this compound is retained in the pulmonary tissue. However, rat and mouse exposure to this compound (1.000 ppm) did not produce any histopathological lesions (10). Rats exposed to acetaldehyde showed some degenerative changes in the olfactory epithelium, whereas in hamsters, the trachea was affected. The progression of the changes was related to an increase in its concentration (5). As for toluene exposure, the experimental animals were shown to have enlarged liver, kidneys, brain mass, but any histopathological changes were reported (1, 7). Depending on a kind of gas mixture and its concentration as well as animal species, age, sex, and management conditions, a various response may occur. According to Woto-Gaye et al. (20) and Nimmermark (15), hydrogen sulphide, ammonia, benzene, and formaldehyde may cause harmful reactions even at low concentrations. Some other gaseous pollutants identified in the investigated object also included xylene and phenol. A percentage of inhaled xylene retained in the pulmonary tissue is 60% and it undergoes rapid biotransformation. Toxicity, however, appears at low concentrations (critical dose for rat – 200 ppm) (3). Under the influence of phenol, the laboratory animals developed neurotoxic, immunotoxic, and hepatotoxic lesions (2).

The changes in the respiratory system of the experimental group appear to be the response to the impact of mixture of air gaseous pollutants released in the chamber. The raising of animals in the most extreme confinement system with no appropriate inspection and a lack of efficient ventilation system leads to the chamber microclimate disturbance. The disturbed microclimatic balance means higher susceptibility of animals to various pathogenic infections that in turn makes them an easy target of attack and penetration of both, biological and chemical agents.

Fat deposition in the liver and kidneys as compared to the previous experiments in which the animals were exposed to the elevated level of gaseous contaminants, proved to be lower (16). Fat amount determined in the present researches, despite some differences between animals, did not produce any degenerative lesions in the liver or renal tubule epithelium. While the glycogen presence in the liver, according to Elumalai and Balasubramanian (8) report, depends on the strength of a toxicant, which may block glycogogenesis process. An animal organism, to satisfy its energy requirements under stress or response to a chemical activity, is capable of stimulating glycogenolysis that results in glycogen breakdown. The aforementioned authors have detected a decreased glycogen level after the direct administration of naphthalene.

The Scandinavian countries made some recommendations on the housing and management of fur animal species caged in closed facilities mainly due to an increased regulatory pressure concerning environmental protection issues. The introduction of the total confinement housing system may minimise the potential of any adverse environmental burden. However, as its implementation is associated with some new hazards and risks, the recommendations should be addressed as a priority research question.

References

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