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The first visually-guided bronchoscopy in European bison (Bison bonasus) – An additional tool in the diagnosis of bovine tuberculosis?

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ABSTRACT

The European bison (Bison bonasus, EB) is an endangered species, and as about 1/3 of its global population is found in Poland, it is particularly important that Polish herds should be monitored. One particular concern is tuberculosis, which is not a marginal problem in wildlife in Poland, and has been microbiologically confirmed in EB, wolves (Canis lupus) and wild boar (Sus scrofa). However, ante mortem diagnosis of tuberculosis in EB is troublesome. Therefore, the present paper evaluates the potential of bronchoscopy as a diagnostic tool. Seven EB were studied, four of which were found to be naturally infected with M. caprae; in two of these, endoscopy identified abnormalities in the respiratory tract ante mortem. Therefore, despite some limitations, endoscopy can be an additional tool for diagnosing tuberculosis in EB, especially in highly valuable animals, and to assess the stage of the disease.

The European bison (Bison bonasus, EB) is an endangered species protected by international law: it is currently categorised as vulnerable by the International Union for Conservation the Nature (IUCN). Its conservation requires a comprehensive approach based around a number of factors, such as setting up new herds, managing environmental hazards and health monitoring protocols (Didkowska et al., 2018; Klich, Olech, Łopucki & Danik, 2018, 2020; Olech, Klich & Perzanowski, 2019). In the last 30 years, bovine tuberculosis (BTB) has proved to be one of the most dangerous diseases in the Polish EB population (Krajewska et al., 2015; Welz et al., 2005). BTB is a chronic infectious disease caused by Mycobacterium bovis or Mycobacterium caprae. In addition, to cattle and other livestock, it is also known to threaten wildlife and humans (O'Reilly et al., 1995). Infection most commonly occurs through the aerogenic route, but some alimentary infections also occur; the primary focus is most often observed in the lungs, but also in the thoracic lymph nodes or mesenteric lymph nodes, depending on the route of infection (Pollock & Neill, 2002).

There is hence a great need to improve the methods of material collection and diagnostic tests for bovine tuberculosis, also in EB (Anusz et al., 2017; Didkowska et al., 2020). While most ante mortem tests are based on detection of the cell-mediated immune response (Bezos et al., 2014), serological tests have also recently become available and have yielded reliable results (Krajewska-Wędzina et al., 2020).

Alternatively, imaging could be used for diagnosis. However, while a

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number of imaging methods, such as X-Rays, computer tomography, magnetic resonance imaging and bronchoscopy, are used for tuberculosis diagnosis in human medicine, they are generally impractical for use in animals (Davies, Lalvani, Muhunthan & Korzeniewska-Koseła, 2018; Skoura, Zumla & Bomanji, 2015). Such examinations are difficult to perform in free-ranging animals, mainly due to the need for technically-advanced equipment that is often not adapted to field work. The use of such tests is typically restricted to zoos or breeding centres used to provide ante mortem diagnostics of infectious diseases and to deepen knowledge of the anatomy and physiology of these animals (Wojick, Langan, Terio, Righton & Drees, 2018). Nevertheless, imaging tests have previously been used in both domestic and wild animals to diagnose tuberculosis, e.g. in rhesus macaques (Macaca mulatta) (Gong et al., 2017) and cats (Felis catus) (Major et al., 2018) and so may also be of use in European bison.

The aim of the current study was to confirm whether any abnormalities present in the lower respiratory tract of European bison infected with Mycobacterium caprae are detectable ante mortem, and to evaluate the potential of endoscopic examination in the diagnosis of bovine tuberculosis (BTB) in these animals. The bronchoscopy procedure was carried out in seven captive EB: two male, five female, aged five to 19 years (Table 1). All were resident at the Bison Breeding Centre in central Poland ($n = 6$) and Zoo ($n = 1$). Although only one animal showed clinical symptoms, in the form of cough and dyspnoea, all were suspected of being BTB-positive due to direct contact with other positive individuals.

Before bronchoscopy and material collection for BTB diagnostics, the animals were immobilized pharmacologically using Palmer's weapon as described previously (Krzysiak & Larska, 2014). Briefly, anaesthesia was performed with etorphine hydrochloride (Captivon, 9.8 mg / ml, Wildlife Pharmaceuticals, South Africaand) and xylazine (Nerfasin, 100 mg / ml, Livisto). In total, the entire bronchoscopy and material collection procedure lasted 20–40 min. Ante mortem procedures were conducted by qualified veterinarians. The welfare of the animals was ensured, in accordance with Polish regulations (Polish Animal Protection Act). Four EB were confirmed post mortem to be M. caprae-positive and three as M. caprae-negative by culture as described previously (Didkowska et al., 2020). Euthanasia was conducted in accordance with the Decision of The Polish Ministry of The Environment number DOP-WPN.286.219.2018.MŚ.

The examination of the upper and lower respiratory tract was performed with a Fujinon EC-200WM2 flexible endoscope (length 170 cm, diameter 13 mm, Fujinon EVE Processor EPX-201 videoprocessor) (Fig. 1). The power source was a combustion generator. The animals were placed in lateral recumbency, or in sternal recumbency if possible. The endoscope was passed through the right or left nostril, depending on which side the head was lying: the endoscope was inserted through the nostril farther from the ground. No lubricant was necessary as large amounts of secretions were present in the respiratory tract. After passing through the larynx isthmus, the trachea and main bronchi were imaged. After the endoscope reached the bronchi, the catheter was inserted through the endoscope canal, a heated 0.9% sodium chloride solution was injected into the respiratory tract and then aspirated to 50 ml; the solution was then taken for further diagnostic tests to diagnose BTB (Didkowska et al., 2020). The image was recorded on a portable computer (Dell Computer Corporation, US). Between each examination, the working parts of the endoscope were thoroughly washed with water and disinfected with Cidezyme enzymatic detergent (Johnson & Johnson, New Brunswick, NJ, US) and glyrasldehyde Lysoformin 3000 solution (Medilab, Bialystok, Poland). The greatest difficulty encountered during the study was delivering the equipment to the European bison in the forest: the examination itself did not present any complications.

No abnormalities were observed in five of the EBs, including three M. caprae-negative and two M. caprae-positive animals. However, were found in two EBs, both M. caprae-positive. In the first animal, the tracheal mucosa was hyperaemic with thick discharge. Purulent discharge was observed in the area of the tracheal carina and main bronchi, an oedema of the carina was detected (Fig. 2). During the examination, cough reactions occurred, in which main bronchi contracted. In the second animal, the nasal tracts were unobstructed and their mucosa was slightly hyperaemic. Fluff mucosa was visible in the throat cavity. No macroscopical lesions were found in the rest of the respiratory tract. This is the first positive identification of tuberculosis lesions in European bison by bronchoscopy.

Although our initial findings indicate that bronchoscopy has the potential to be a very valuable tool for identifying traces of BTB in European bison ante mortem, the usage of such delicate and uncomfortable equipment in the field can be very difficult, especially in the case of free-ranging EB. In addition, the potential for performing anaesthesia is limited by the low temperatures in the area. It should be also noted that bronchoscopy is quite an invasive method. Therefore, endoscopy can only be considered as an additional examination in valuable individuals from captive breeding canters and zoos, where it can be used to assess the severity of tuberculous lesions in individuals rather than make diagnosis. Also it should be highlighted that due to its considerable body weight, it is not safe for an EB, the largest free-ranging mammal in Europe, to remain immobilised in a laying position: amongst others, it carries the risk of aspiration pneumonia, which can be fatal. For EB, the ideal solution would involve the implementation of non-invasive methods of material sampling and diagnostics, without the need to perform pharmacological immobilization. This procedure is currently recommended for ante mortem diagnosis of infectious diseases in free-ranging animals (JWD Wildlife Welfare Supplement Editorial Board, 2016). However, in the case of tuberculosis, the diagnosis and collection of appropriate clinical material from a living wild animal is very difficult without pharmacological immobilization, and the chance of isolating mycobacteria is small.

Our initial findings regarding bronchoscopy in EB may open the door to further research and the use of this method in the diagnosis of diseases other than tuberculosis. Such imaging testes may play a crucial role in conserving endangered species by allowing ante mortem detection of various medical conditions.

Summing up, our findings confirm the ante mortem detection of abnormalities in respiratory tract for the first time in EB, thus demonstrating the potential value of bronchoscopy in the diagnosis of tuberculosis. However, the procedure has a number of technical limitations regarding its use in the field, and may not detect highly-specific lesions (Fig. 3).

Table 1
Description of European bison tested.

ID	Gender	Year of birth	Place
1	Male	2013	Bison breeding centre
2	Male	2011	Bison breeding centre
3	Female	2011	Bison breeding centre
4	Female	2008	Bison breeding centre
5	Female	2012	Bison breeding centre
6	Female	2012	Bison breeding centre
7	Female	1999	Zoo

Fig. 1. Endoscopic examination in European bison.

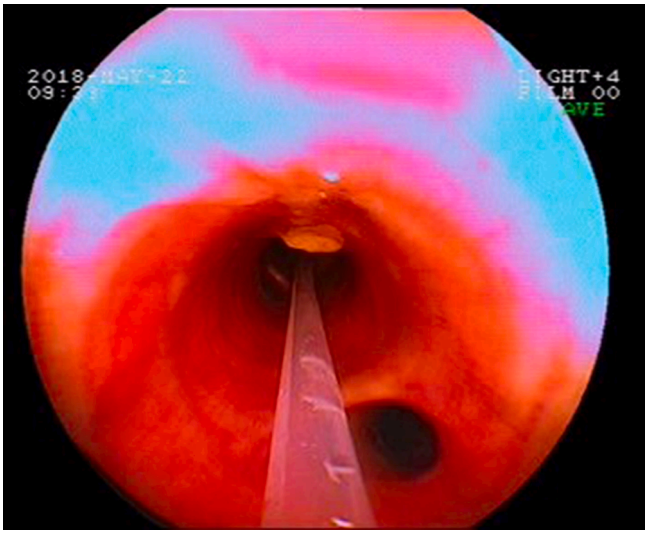

Fig. 2. Endoscopic image of the Mycobacterium caprae-positive European bison – an oedema of the tracheal carina, the airway mucosa is swollen and hyperaemic, mucopurulent discharge visible in the lumen above the catheter.

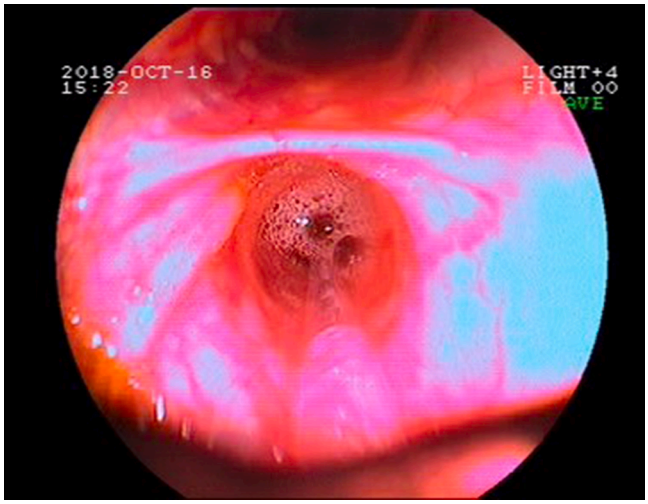

Fig. 3. Endoscopic image of the respiratory tract of a European bison with no changes during wash collection – healthy bronchus image, catheter, foam (sodium chloride which was already administrated for procedure purposes).

Conflict of interest statement

None of the authors of this paper has a financial or personal relationship with other people or organisations that could inappropriately influence or bias the content of the paper.

Ethical statement

Ante-mortem procedures in European bison does not need approval decision due to Polish law and was conducted by qualified veterinarians maintaining animals welfare.

Anna Didkowska

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Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Wanda Olech reports financial support was provided by Forest Found (Poland). This author has no additional relationships to disclose. This author has no patents to disclose. The corresponding author reports no additional activities to disclose.

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