Mycobacterial infections in horses: a review of the literature

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ABSTRACT: Mycobacterial infections are rarely diagnosed in horses. Nevertheless, it was possible to obtain noteworthy information on diagnosis, manifold clinical signs and epidemiological relationships from available literature. It has been more than 60 years since a review dealing with this topic was published. Based on literature analysis, it was found that after bovine tuberculosis control in cattle and other animals in Central Europe, *Mycobacterium avium* complex (*MAC*) members were the most commonly found causal agents of mycobacterial infections in horses. At present, mycobacterial infections caused both by *M. tuberculosis* complex and *MAC* are occasionally diagnosed in horses in Central Europe. Finally, under certain conditions, horses may become a source of mycobacterial infections for other animal species or for immunocompromised humans.

Keywords: avian mycobacteriosis; zoonosis; vector; Czech Republic; PCR; RFLP; IS*901;* IS*1245*; *M. a. hominissuis*; *M. a. paratuberculosis*; *M. intracellulare*; *M. bovis*

Contents

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1. Introduction

Horses are considered naturally highly resistant to mycobacterial infections (Schmidt, 1930; Griffith, 1937; Innes, 1949; Luke, 1958; Drazan et al., 1962; Muser and Nassal, 1962; O'Reilly and Daborn, 1995; Thorel et al., 1997). Nevertheless, tuberculosis in horses is a disease which has been known for a long time. One of the earliest references is the description of avian tuberculosis in a horse in England at the beginning of last century (M'Fadyean, 1918). In the year 1921, Wester (1921) described in detail clinical signs of tuberculosis caused by members of *Mycobacterium tuberculosis* complex (*MTC*) in 26 horses. The first review summarizing isolation and characterization of mycobacteria from horses during the period 1896 to 1942 was written by Verge and Senthille (1942). More than 60 years later, this review summarizes knowledge from currently avialable literature from the standpoint of currently accepted taxonomy of mycobacteria.

Tuberculous lesions in horses of various breeds were detected in different continents (Tables 1 to 5). Data obtained by veterinary-hygienic inspections in slaughterhouses are mostly based on gross findings of tuberculous lesions without laboratory examination (Anonymous, 1960, 1961). In some cases, mycobacteria were demonstrated only by light microscopy based on the detection of acid-fast bacilli (AFB) in tuberculous lesions after staining according to Ziehl-Neelsen (Z-N); further examination by culture was not performed (Oikawa et al., 1976). Isolated bacteria in many other cases were identified only according to the existing knowledge at the time of their isolation or their identification was not accurate. However, it was possible to obtain a lot of noteworthy information on diagnosis, manifold signs of the disease and also the relationships to epidemiology (Tables 1 to 5).

High natural resistance of horses to mycobacterial infections was obviously also the reason for rare findings of tuberculous lesions in slaughtered horses even in the period of high prevalence of bovine tuberculosis in cattle in the first two thirds of last century in Europe (Thoen and Steele, 1995). Tuberculous lesions were detected only in 0.3% slaughter horses in West Germany in the year 1951 (Schutzler, 1954). In the following two years, the incidence of tuberculous lesions without culture examinations in slaughter horses was low: tuberculosis was diagnosed in 220 (0.3%) of 66 124 slaughtered horses in the year 1958 and in 234 (0.3%) of 76 440 slaughtered horses in the year 1959 (Anonymous, 1960, 1961). Although incidence of bovine tuberculosis in animals from the Czech Republic in the middle of last century was relatively high (Pavlas, 1999), tuberculous lesions were detected only in 5 (2.3%) of 214 slaughter horses during the years 1956 to 1958 (Krejci, 1958).

Bovine tuberculosis was put under control as early as in 1980s in Central Europe (Pavlik et al., 1998, 2002a,b). Incidence of the infection caused by *M. bovis* in the other species of domestic and wild animals and in human population also declined in the last decade of last century (Machackova et al., 2003; Pavlik et al., 2002c,d,e, 2003a,b).

2. Taxonomy of mycobacterial complexes of *M. tuberculosis* and *M. avium* isolated from horses

2.1. *M. tuberculosis* complex

The members of the *M. tuberculosis* complex (*MTC*) which contain the specific insertion sequence IS*6110* are now classified as follows: *M. tuberculosis*, *M. africanum*, *M. bovis*, *M. bovis* BCG, *M. microti*, *M. canetti*, and the recently described *M. caprae* (Aranaz et al., 2003; Erler et al., 2004) and *M. pinnipedii* (Cousins et al., 2003).

2.2. Occurrence of respective members of *M. tuberculosis* in horses

M. bovis and *M. tuberculosis* from *MTC* members were isolated from horses (Table 1). As in the time of isolation *M. caprae* (Aranaz et al., 2003), had not been described yet, it is not possible to find out with absolute certainty whether that *MTC* member affected horses or not.

2.3. *M. avium* complex

Tuberculous lesions may also be caused by the members of the *M. avium* complex (*MAC*) in susceptible hosts. The *MAC* comprises 28 serotypes (Wolinsky and Schaefer, 1973) and according to the currently accepted taxonomy, it has been divided into three groups by affiliation with serotypes and/or the contents of specific insertion sequences and/or

Table 1. Spontaneous infection in horses caused by *Mycobacterium tuberculosis* complex members

Reference	No. of horses	Samples	*Post mortem* findings	Notes
Infection caused by *M. tuberculosis*				
Verge and Senthille (1942)	11	?	Tuberculous lesions in unspecified tissue	
Muser and Nassal (1962)	1	?	Tuberculous lesions in unspecified tissue	
Infection caused by *M. bovis*				
Verge and Senthille (1942)	90	?	Tuberculous lesions in unspecified tissue	
Krejci (1958)	1	Pooled tissue	Multiple tubercles found in pleura, lung tissue and bronchial ln[b]	AFB[a] found in all samples
	1	Pooled tissue	Multiple tubercles found in lung tissue and bronchial ln[b]	AFB[a] found in all samples
Muser and Nassal (1962)	2	?	Tuberculous lesions in unspecified tissue	
Pavlik et al. (1998)	1	?	Tuberculous lesions in unspecified tissue found in 1976	Intimate contact of infected cattle with *M. bovis* on the farm
Monreal et al. (2001)	1	Spleen, mesenteric ln[b]	Multifocal granulomas in spleen, liver, lungs, pancreas and kidney, enlarged mesenteric, hepatic and mediastinal ln[b]	AFB[a] massively found in spleen by biopsy

[a]acid-fast bacilli (AFB) detected after the Ziehl-Neelsen staining; [b]lymph nodes; ? = data not given

virulence for birds (Runyon et al., 1986; Wayne and Kubica, 1986; Thorel et al., 1990; Kunze et al., 1992; Guerrero et al., 1995; Pavlik et al., 2000a; Mijs et al., 2002) as follows: *M. avium* subsp. *avium*, *M. a. hominissuis* and *M. intracellulare*. *M. chimaera* sp. *nov.* (a new member of the *MAC*) had been recently described (Tortoli et al., 2004).

2.3.1. *M. avium* subsp. *avium*

The first *MAC* member *M. a. avium* (serotypes 1 to 3 and genotype IS*901*+ and IS*1245*+) causes avian tuberculosis in birds. The field isolates were fully virulent for birds (Pavlik et al., 2000a; Dvorska et al., 2003).

2.3.2. *M. avium* subsp. *hominissuis*

The second *MAC* member *M. a. hominissuis* (serotypes 4 to 6, 8 to 11 and 21 and genotype IS*901*– and IS*1245*+; Piening et al., 1972; Mijs et al., 2002) is "partially virulent for pullets", in which it produces lesions only at the site of inoculation (e. g. in pectoral muscles). This member was most often isolated from pigs (Pavlik et al., 2003c; Matlova et al., 2004a,b,c,d), samples from external environment (Horvathova et al., 1997; Kazda, 2000; Matlova et al., 2003; Trckova et al., 2004) and from humans (Pavlik et al., 2000a; Dvorska et al., 2002), and less frequently from non-vertebrates (Fischer et al., 2001, 2003a,b, 2004a,b), small terrestrial mammals (Fischer et al., 2000), wild boar (Machackova et al., 2003) and wild ruminants (Machackova et al., 2004).

2.3.3. *M. intracellulare*

The third *MAC* member *M. intracellulare* (serotypes 7, 12 to 20 and 22 to 28 and genotype IS*901*– and IS*1245*–) "is non-virulent for pullets" (Wayne and Kubica, 1986). It occurs less frequently in the environment than *M. a. hominissuis* (Matlova et al., 2003) yet it was isolated from some people affected by mycobacteriosis (Kulski et al., 1995; Wallace et al., 2002).

Table 2. Spontaneous infection in horses caused by *Mycobacterium avium* subsp. *avium*

Reference	Horses		Sero-type[b]	Examined tissue samples	*Post mortem* findings	Notes
	No.	Age[a]				
Verge and Senthille (1942)	10	?	Nt	?	Tuberculous lesions in unspecified tissue	
Foschi (1946)	1	12	Nt	Lung tissue	Miliary lung tuberculosis	
Lesslie and Davies (1958)	1	8	Nt	Pooled tissue	Generalised granulomatous lymphadenopathy, miliary liver and lung lesions, abdominal mass	
Nielsen and Spratling (1968)	1	10	Nt	Abdomen, thorax, bones	Nodular spleen lesions, granulomatous lymphadenopathy in abdomen and thorax cavities, discrete lesions of vertebrae and ribs	
Binkhorst et al. (1972)	1	1.6	Nt	Cervical vertebrae	Granulomatous lymphadenopathy (head and neck), discrete lesions in cervical vertebrae	AFB[d] not found in tissue, Shetland pony
Baker (1973)	1	Aged	Nt	Udder – pus	Granulomatous lesions in udder and associated ln[c], miliary liver lesions	AFB[d] massively found in pus from udder
			Nt	Udder – ln[c]		
	1	4	Nt	Cerebrospinal liquor, spleen, liver, kidney, bones, ln [c]	Granulomatous lymphadenopathy (head, neck, thorax and abdomen), splenic enlargement, discrete lesions of all vertebrae, sternum and ribs	AFB[d] not found in tissue, Welsh pony
	1	?	Nt	Cervical, thoracic, lumbar vertebrae	Tuberculous lesions present in vertebrae, abdomen and thorax cavities including spleen, liver and kidney	Shetland pony
Cimprich (1974)	1	2	Nt	Intestine	Granulomatous enteritis/colitis, regional lymphadenopathy	
			Nt	Mesenteric ln[c]		
			Nt	Faeces		
Dolfijn and Van der Kamp (1975)	1	5.6	Nt	Cervical abscess	Cervical abscess	AFB[d] found in abscess
Merritt et al. (1975)	1	2	Nt	Intestinal tract	Granulomatous enteritis/colitis, regional granulomatous, lymphadenopathy, arthritis of thoracic vertebral joints	
Van Dijk et al. (1975)	1	Aged	Nt	?	Miliary lesions and exudative foci in lungs, liver, kidneys, spleen, intestine, bone marrow, and ln[c]	
Jorgensen (1978)	1	?	2	?	Tuberculous lesions in unspecified tissue	
Mair et al. (1986)	1	9	1/2	Pooled tissue	Chest wall and diaphragm were covered by soft nodular thickenings, ln[c] in thorax and abdomen were enlarged, spongy and oedematous	Granulomatous lesions in liver and lung biopsies

Buergelt et al. (1988)	1	2	1	Faeces	Miliary and lung tuberculosis, mesenteric ln[c] enlargement	Intimate contact with farm chicken and barn pigeons
			MAC	Mesenteric ln[c]		
			1/8, 5	Mesenteric ln[c]		

[a]age given in years; [b]*MAC Mycobacterium avium* complex virulent for pullets without any serotyping; [c]lymph nodes; [d]acid-fast bacilli detected after the Ziehl-Neelsen staining; Nt = not tested; ? = data not given

Table 3. Spontaneous infection in horses caused by *Mycobacterium avium* subsp. *hominissuis*

Reference	Horses		Sero-type	Examined tissue samples	*Post mortem* findings	Notes
	No.	Age[a]				
Buergelt et al. (1988)	1	7	8	Liver	Miliary liver tuberculosis, mesenteric ln[c] enlargement	Imported from Argentina to Japan
	1	6	8	Colon	Miliary tuberculosis of colon, mesenteric ln[c] enlargement	
			8	Mesenteric ln[c]		
Lofstedt and Jakowski (1989)	1	1.2	8	Ulcerative colitis	Diffuse granulomatous enterocolitis and hepatitis with associated granulomatous lymphadenitis involving mesenteric and hepatic ln[c]	AFB[d] detected in the granulomas of all tissue samples including liver biopsy
Flores et al. (1991)	1	1.5	8	Mesenteric ln[c]	Subserous nodules in enlarged mesenteric ln[c], tuberculous bronchiolitis, miliary pleuritis and peritonitis	
Gunnes et al. (1995)	1	2	4	Lung tissue	Tuberculous lesions found in lungs and mesenteric ln. in standardbred colt with a 12-month history of weight loss, intermittent diarrhoea and anorexia	AFB[d] were not detected in examined tissue of standardbred colt
				Mesenteric ln[c]		
				Liver		
				Spleen		
Leifsson et al. (1997)	1	6	Nt[b]	Pooled tissue sample	Systemic tuberculosis: mycobacterial bilateral uveitis, disseminated granulomatous foci in the myocardium, lungs, bronchial ln[c], kidneys, liver, pancreas, colon and visceral serosal surfaces (AFB[d] in heart, lungs, bronchial ln[c], kidneys, nodules on the pleural and splenic surfaces and eyes)	Stabled on straw and on wood shavings last 10 months of the life

[a]age given in years; [b]exact subtype given personally by Dr. Steen Giese (Danish Veterinary Laboratory, Copenhagen, Denmark); [c]lymph nodes; [d]acid-fast bacilli detected after the Ziehl-Neelsen staining; ? = data not given; Nt = not tested

Table 4. Spontaneous infection in horses caused by *Mycobacterium avium* complex isolates without exact typing

Reference	Horses		Examined tissue samples	*Post mortem* findings	Notes
	No.	Age[a]			
Muser and Nassal (1962)	63	?	?	Tuberculous lesions in unspecified tissue	
Anzai et al. (1989)	1	4	Pooled samples	Numerous tubercles in lungs, liver, enlarged kidney, ln[c], atrophy of thymus	Many AFB[b] found in tubercles in thoroughbred racehorse
Sills et al. (1990)	1	4	Retropharyngeal ln[c]	Diffuse chronic granulomatous lymphadenitis of the mandibular, retropharyngeal, and cervical ln[c], diffuse granulomatous and interstitial pneumonia, granulomatous hepatitis and colitis with focal erosions and ulcerations, multifocal ulceration and caseous exudation of the guttural pouches, multifocal erosions and ulcerations of colonic mucosa	AFB[b] were detected in all examined tissues except the liver and lung
			Mandibular ln[c]		
			Cervical ln[c]		
			Colon		
			Lung		
Cline et al. (1991)	1	17	Mare – faeces	Mare: granulomatous lesions with ulceration in colon, enlargement of mandibular, mesenteric and tracheobronchial ln[c]; Foetus: granulomatous lesions in small intestine	Rough colonies non-typeable with antisera for 1–2, and 4–20 serotypes
			Mare – ileocecal ln[c]		
		(160)	Foetus – intestine		
Helie and Higgins (1996)	1	6	Allantochorion	Placentitis and abortion of 30 kg female foetus (approximately 300 days of gestation)	AFB[b] were seen in allantochorion, not in liver or lung

[a]age is given in years and (160) is the estimation of age of aborted foetus in days; [b]acid-fast bacilli detected after the Ziehl-Neelsen staining; [c]lymph nodes; ? = data not given

2.3.4. *M. avium* subsp. *paratuberculosis*

The causal agent of paratuberculosis is classified as *MAC* and subspecies *M. avium* subsp. *paratuberculosis* according to the currently accepted taxonomy. Its identification is possible by the specific sequence IS*900* (Green et al., 1989) or by pseudo-IS*901* PCR (Svastova et al., 2002). Larsen et al. (1972) induced clinical signs of body weight loss by experimental intravenous and oral infection of the horse by *M. a. paratuberculosis* and recovered the causal agent of paratuberculosis from intestinal mucosa; however spontaneous paratuberculosis in horses has not been diagnosed after that year.

2.4. Occurrence of the members of *M. avium* in horses

Based on the knowledge of serotypes, virulence of *MAC* isolates for pullets and information given in available publications (Table 2), it is possible now to more accurately specify respective *MAC* members isolated from horses during 60 years.

Muser and Nassal (1962) isolated the members of *M. avium* from 63 (91.3%) of 69 horses, among which 55 isolates were virulent for birds and hence they were likely *M. a. avium* subspecies. Only 8 remaining isolates were partially virulent for birds, hence they were *M. a. hominissuis* subspecies (Table 4).

2.5. Occurrence of atypical mycobacteria in horses and their clinical significance

Among atypical mycobacteria (conditionally pathogenic) *M. smegmatis* was detected in a dermal abscess in one pony (Booth and Wattret, 2000) and *M. terrae* complex in stomach content, liver and lung tissues of an aborted foetus at day 250 of gestation (Tasler and Hartley, 1981). Among the other species of atypical mycobacteria, *M. fortuitum* and *M. chelonae* were isolated from nasal swabs of clinically normal horses kept on straw or sawdust bedding (Mair and Jenkins, 1990), (Table 5).

3. Occurrence of respective mycobacterial species in tuberculous processes in horses

Our knowledge of incidence of mycobacterial species in horses at the beginning of the 20th century is based on the review of Verge and Senthille (1942) which analysed 24 publications from the years 1896 to 1942, which describe 112 cases of tuberculosis in horses. The authors found that the isolates of *M. bovis* from tuberculous lesions (90 horses/80.4%) predominated. Among other mycobacterial species, *M. tuberculosis* (11/9.8%), *M. a. avium* (10/8.9%) and atypical mycobacteria (1/0.9%) were isolated from horses. However, unfavourable epidemiological situation of bovine tuberculosis in cattle began to change in the second half of last century due to the successfully introduced national sanitation programmes (Thoen and Steele, 1995).

Two decades later, Muser and Nassal (1962) recorded that among 69 isolates, 63/91.3% were *M. a. avium* and merely 2/2.9% isolates were *M. bovis* species, 3/4.4% *M. tuberculosis* species and 1/1.4% isolates were atypical (scotochromogenic) mycobacteria. In the Czech Republic, only one isolate of *M. bovis* in the horse was detected in the year 1976 (Pavlik et al., 1998).

Table 5. Spontaneous mycobacterial infection in horses caused by atypical (conditionally pathogenic) mycobacteria

Reference	Horses		Species of *Mycobacterium*	Examined tissue samples	*Post mortem* findings	Notes
	No.	Age[a]				
Verge and Senthille (1942)	1	?	Atypical mycobacteria	?	Tuberculous lesions in unspecified tissue	
Muser and Nassal (1962)	1	?	Scotochromogenic mycobacteria	?	Tuberculous lesions in unspecified tissue	
Tasler and Hartley (1981)	1	250 days	*M. terrae* complex	Stomach content	No autolysis or gross lesions were found in the foetus	AFB[b] were detected in cultured samples, thymus and spleen
			M. terrae complex	Liver		
			M. terrae complex	Lung		
Mair and Jenkins (1990)	15	1 to 17	*M. fortuitum*	Nasal swabs	Horses with no history of respiratory disease in normal clinical status	Horses stabled on straw or shavings
	3		*M. terrae*			
	1		*M. chelonae*			
	2		*M.* spp.			
Krejci (1958)	1	?	*M.* sp.	Tubercle in myocardium	Tubercle of the nut size found in the myocardium	AFB[b] found in the sample
Booth and Wattret (2000)	1	?	*M. smegmatis*	Abscess	Stifle abscess	

Explanations see in Table 4

3.1. Localization of pathologic process and gross changes observed in horses with tuberculosis

In the second half of last century, tuberculous lesions caused by *M. bovis* detected in horses were localized in various organs, most frequently in lungs, less frequently in liver, spleen and kidney (Krejci, 1958; Muser and Nassal, 1962; Zaharija et al., 1974, 1981). The infection site was most commonly the gastrointestinal tract, particularly cecum and colon. The infection was probably facilitated by ulcerations of the large intestine by invasion of parasites. Infection of mesenteric lymph nodes formed a complete primary complex (Godgluck, 1967). Oral infection is the cause of a more frequent non-complete primary complex with the affection of retropharyngeal and mesenteric lymph nodes without evident tuberculous lesions in the drained part of mucosa. Provided *MAC* was the cause of the infection, proliferative enteritis develops in small intestine which resembles Johne's disease (paratuberculosis) of cattle both by gross and microscopic examinations. If the primary lesions are found in the large intestine, they usually appear like tuberculous ulcerations (Dungworth, 1993).

If pulmonary tuberculosis is diagnosed, bronchial lymph nodes are always affected by a diffuse process with gross findings similar to those detected in the lungs. Tubercles in spleen (more frequently) and in liver are usually of a large shapeless formation character (Rooney and Robertson, 1996).

Hematogenous dissemination of mycobacteria to lungs, liver, spleen, and serosa is most common and is registered as miliary granuloma formation (Peel, 1983). Skin is another frequently affected organ by mycobacteria (Nieland, 1938; Buss, 1954; Pinkiewicz et al., 1963; Dukic and Putnik, 1971; Flores et al., 1991; Sevilla et al., 1995). Foetal infection caused by *MAC* (without exact typing of isolate) followed by abortion was also described (Cline et al., 1991; Helie and Higgins, 1996).

3.2. Clinical signs in infected horses

Diversity of clinical signs in horses affected by mycobacterial infection depends on the extent of affection of respective organs. They may share characteristics with a number of other diseases which may result in difficult diagnosing of mycobacterial infection in horses. Clinical course of tuberculosis in 26 horses with different localizations (e.g. *glandula parotis*, lymph nodes, lungs, liver and spleen) was described as early as in the year 1921 by Wester (Wester, 1921). Clinical signs varied, such as weight loss, pyrexia, chronic cough, mastitis, diarrhoea, and neck stiffness.

In horses the most frequent presenting complaint is chronic weight loss, weakness and lethargy. Horses terminally affected by the pulmonary form are febrile and dyspnoeic and have a cough (Peel, 1983; Beech and Sweeney, 1991).

4. Diagnostic methods of mycobacterial infections

Mycobacterial infections in horses may be considered as occasional in Central Europe (Cimprich, 1974; Mair et al., 1986; Buergelt et al., 1988; Pavlik et al., 1998). Their diagnosis is difficult because of both diversity of their clinical signs and atypical morphological lesions and because of low specificity and sensitivity of the skin test (Muser and Nassal, 1962; Konyha and Kreier, 1971). In case of suspected intestinal tuberculosis Pearson and Heidel (1998) recommended to perform also rectum or distal colon biopsy to confirm the diagnosis. In the case of a lower intensity of mycobacterial infection, staining for AFB and culture examination should complete histopathological examination to perform accurate diagnosis.

4.1. Histopathological examination

Microscopically characteristic granulomatous reaction in horses is abounding in epitheloid cells, giant cells, lymphocytes and fibroblasts (M'Fadyean, 1918; Rooney and Robertson, 1996).

Mycobacteria are sometimes detected in tuberculous lesions after Ziehl-Neelsen staining. It should be considered that AFB-like formations may be observed in the tissues when members of *Nocardia, Rhodococcus*, and *Corynebacterium* genera are present. From 1965 to 1983 *Nocardia asteroides* infection was diagnosed in 16 horses in the Veterinary Teaching Hospital University of California (Davis, California, USA). In 14 immunocompromised horses pulmonary or disseminated infections ended fatally (Biberstein et al., 1985).

4.2. Culture examination

For laboratory examination, approximately 1 g of tissue is collected, homogenized and decontaminated (Corner, 1994; Fischer et al., 2000; Pavlik et al., 2000b).

After decontamination samples are centrifuged and the sediment is inoculated to egg based agar media e.g. according to Stonebrink, Löwenstein-Jensen, liquid serum medium according to Sula (Sevac Prague, Czech Republic), Ogawa and liquid serum media by Banic (Votava, 2000) used for the detection of members of *MAC* and *MTC.* For *M. a. paratuberculosis* detection, the sediment is inoculated onto Herrold egg yolk medium (HEYM) containing growth stimulator Mycobactin J. Inoculated media are incubated at range from 24°C to 37°C. Mycobacterial growth is checked after the first week of incubation and then every second week for at least two months (Kubin et al., 1986; Wayne and Kubica, 1986; Allen, 1991; Votava, 2000).

4.3. Investigation of epidemiology of mycobacterial infections by molecular-genetic methods

Nowadays, various molecular techniques are used for the study of epizootiology and epidemiology of the mycobacterial infections. The techniques are based on DNA segments specific for respective taxons of mycobacteria. For the *MAC* members which are most consequential to the health, it is possible to use the standardized restriction fragment length polymorphism (RFLP) methods with IS*901* probe for the isolates of *M. a. avium* (Dvorska et al., 2003, 2004) and with IS*1245* probe for the isolates of *M. a. hominissuis* (Van Soolingen et al., 1998).

5. The risk factors of mycobacterial infection transmission from horses to humans

Although more than 0.5 million horses are kept in Central Europe, it can be concluded that the risk of transmission of tuberculous infection caused by members of *MTC* and *MAC* from horses to humans is very low. This is primarily because of the control measures for bovine tuberculosis in cattle and other animals monitored, which are in the countries of Central Europe (Czech Republic, Poland, Hungary, Slovakia, Bosnia and Herzegovina, and Croatia).

The incidence of mycobacterial infections caused by *M. a. avium* members in horses can also be considered occasional. The numbers of diagnosed cases of mycobacterial infection in Central Europe were very low as summarized in Tables 1 to 5. The incidence of infections caused by *M. a. avium* in the other animals (such as pigs), e.g. in the Czech Republic also declined significantly (Pavlik et al., 2003c).

Although, there is a low prevalence and incidence of tuberculosis in human population (Pavlik et al., 2003b), there is a risk of infection for human population is quite low, except for immunocompromised patients who are particularly susceptible to mycobacterial infections (Dvorska et al., 2002) and under certain conditions, particularly for *HIV/AIDS* patients, horses may be the source of mycobacterial infection.

According to our previously published results (Pavlik et al., 2000a; Dvorska et al., 2003) and based on this review of the literature we can make following conclusions:

1. While *M. bovis* may have been a significant pathogen earlier last century its role has been superseded by "opportunistic mycobacteria" in recent years.
2. Mycobacterial infections caused by *M. avium* largely occur sporadically and horses do not constitute a reservoirs host.
3. Atypical mycobacteria (*M. terrae, M. chelonae,* and *M. smegmatis*) may cause colonisation and minor disease in housed animal.

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