Mycobacteria in water, feedstocks and food: analysis of publications

M. Kaevska, K. Hruska

Veterinary Research Institute, Brno, Czech Republic

ABSTRACT: Papers on mycobacteria in food, feed and water, published between 1945 and 2010 and indexed in the database Web of Science® (Thomson Reuters) were ranked according to authors, institutions, countries and source titles. The total number of papers on mycobacteria and food and mycobacteria and water were 1486 and 1419, respectively. More than 40% of papers have been published in the last five years. In addition to publications in peer reviewed journals the archives of ProMED-mail and the Rapid Alert System for Food and Feed of the European Union were also searched. It is evident that much attention is being paid to mycobacteria in food, feed and water as they likely pose a public health risk.

Keywords: food safety; public health; milk; meat; beef; cheese; fish; fruits; vegetables

There is no doubt that *Mycobacterium bovis*, a known member of the obligatorily pathogenic group of mycobacteria, can be shed in milk and can cause tuberculosis as a food pathogen. A globally disseminated species, contaminating milk, is *Mycobacterium avium* subsp. *paratuberculosis*. Many other mycobacteria are present in the environment and in food and potable water, and high numbers of cells are often consumed daily over an extended period of time. Generally, although highly resistant to higher temperatures and some disinfectants, mycobacteria are not yet considered to pose a risk as food pathogens. Nevertheless, non-tuberculous mycobacterioses represent serious infections for immunodeficient persons and food or water can be a vector for their transmission. A basic consideration is that of dead mycobacterial cells as source of immunomodulators and it is generally accepted that muramylpeptides, originating from peptidoglycans in bacterial cell walls, are very effective triggers of different immunoprotective and inflammatory pathways. Unfortunately, it is not known how many mycobacterial cells surround us and how easy it is to ingest thousands of cells with contaminated food, water, biofilms, aerosols or air. Greater interest should concern the presence of mycobacteria in baby food, because formula fed babies may be under the influence of immunoactive components of mycobacterial cells (Hruska et al., 2005). A recently published book has summarised the impact of mycobacteria in the environment on human and animal health (Kazda et al., 2009). Some reviews pertaining to mycobacteria in food, feed and water are listed in Table 1.

The Web of Knowledge® portal (Thomson Reuters) has already been used to analyse the search results of publications pertaining to paratuberculosis (Hruska, 2004; Kaevska and Hruska, 2010a) and *Mycobacterium avium* (Kaevska and Hruska, 2010b). The aim of this analysis is to assess the trend in research on mycobacteria in water, feedstocks, and food and participation of authors, institutions and countries in the published results.

MATERIAL AND METHODS

The Web of Science, Science Citations Index Expanded, Social Sciences Citation Index and Art and Humanities Index databases® (Thomson

Supported by the Ministry of Agriculture of the Czech Republic (Grant No. MZE 0002716202) and the Ministry of Education, Youth and Sports of the Czech Republic (AdmireVet; Grants No. CZ.1.05/2.1.00/01.0006 and No. ED 0006/01/01).

Table 1. Selected review articles published in recent years

Food as a source of exposure to mycobacteria	Anonymous, 2010; Nacy and Buckley, 2008
Food safety interventions in primary production of beef and lamb	Adam and Brulisauer, 2010
Mycobacteria from foods as possible exposure sources	Argueta et al., 2000
Mycobacterium paratuberculosis: a potential food-borne pathogen?	Collins, 1997
Mycobacterium avium subsp. *paratuberculosis* in food products	Eltholth et al., 2009
Mycobacterium avium subsp. *paratuberculosis* in food	Cirone et al., 2007
Mycobacterium avium subsp. *paratuberculosis* in milk and cheese	Grant, 2006
Emerging bacterial pathogens in meat and poultry	Mor-Mur and Yuste, 2010
Isolation of mycobacteria from porcine lymph nodes	van Ingen et al., 2010
Mycobacterium avium from patients and foods	Yoder et al., 1999
Infections due to *Mycobacterium avium* complex	Kasperbauer and Daley, 2008; McGrath et al., 2010
Mycobacterium bovis as a zoonosis	Thoen et al., 2006
Tuberculosis: a re-emerging disease in animals and humans	Thoen et al., 2009
Nontuberculous mycobacteria causing disease in humans	van Ingen et al., 2009
Mycobacteria and HIV-Infected Individuals	Yajko et al., 1995
Free-living amoebae – a vector for mycobacteria	Salah et al., 2009
Mycobacteria in water and biofilms	Falkinham 2009a,b; Falkinham, 2010
Mycobacteria in tap water	Chang et al., 2002
A way towards detection techniques	Nayak et al., 2009
Detection of *M. a. paratuberculosis* in milk and milk products: a review	Slana et al., 2008

Reuters) were used to search articles published between 1945 and 19 November 2010.

The database **Mycobacteria and water** covered records, resulting from using the search profile (mycobact* AND water).

The database **Mycobacteria and food** covered records, resulting from using the search profile [mycobact* AND (food OR feed OR milk OR meat OR cheese OR fish OR fruit* OR vegetable*)]. The search results have been refined using key words

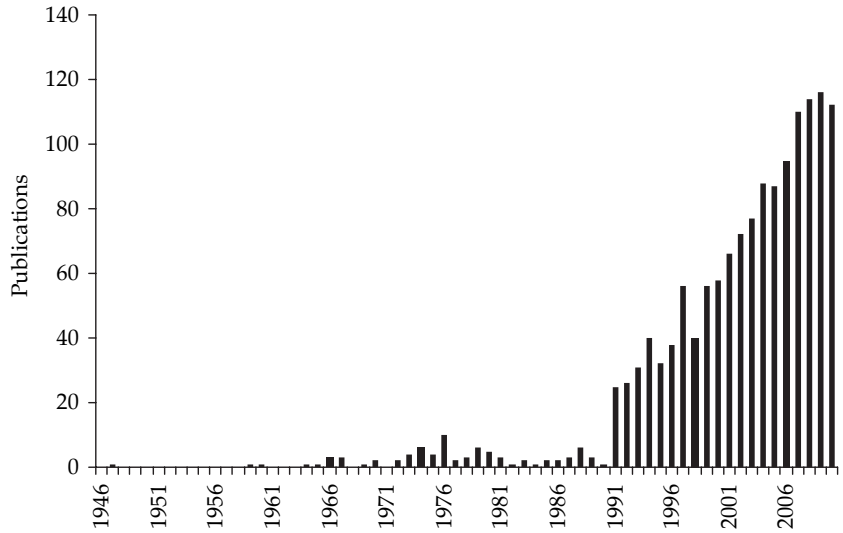

Figure 1. Papers on mycobacteria and water published from 1945 and indexed in the Web of Science database

Table 2. Search results on mycobacteria and food or water

	All years	2006–2010
Mycobacteria and water	1419	558
Authors	5315	
Institutions	1601	
Country	79	
Source	564	
Mycobacteria and food	1486	668
Mycobact* AND		
Milk	626	307
Fish	381	151
Meat	98	45
Fruit*	65	39
Cheese	37	20
Vegetable*	11	6
Food	352	166
Feed	119	35
Authors	4814	
Institutions	1512	
Country	84	
Source	531	

food, feed, milk, meat, cheese, fish, fruit*, and vegetable*.

Both databases have been analyzed according to the year of publication, author, institution, country and source titles.

Additionally, the web archive ProMED-mail (the Program for Monitoring Emerging Diseases, http://www.promedmail.org) and the Rapid Alert System for Food and Feed (RASFF) of the European Union

Table 3. Authors with 10 or more publications on mycobacteria and water (total number of authors 5315)

	Authors	Number of records
1	Falkinham, JO	29
2–3	Katila, ML	16
2–3	Pavlik, I	16
4	Portaels, F	15
5	Wallace, RJ	14
6–7	Matlova, L	11
6–7	Zhang, YS	11
8–10	Groves, MJ	10
8–10	Iivanainen, E	10
8–10	Vincent, V	10

https://webgate.ec.europa.eu/rasff-window/portal/ were searched for records and notifications on mycobacteria.

RESULTS AND DISCUSSION

Mycobacteria and water

In total 1419 papers have been published between 1947 and 2010 (Figure 1, Table 2), 558 (39,3%) during the last five years, indexed from 2006 to November 2010.

Authors who published ten or more papers, collected in the database for mycobacteria and water are presented in Table 3, the institution of authors in Table 4, and countries in Table 5. From the number of published papers it can be seen that research on mycobacteria in water is being

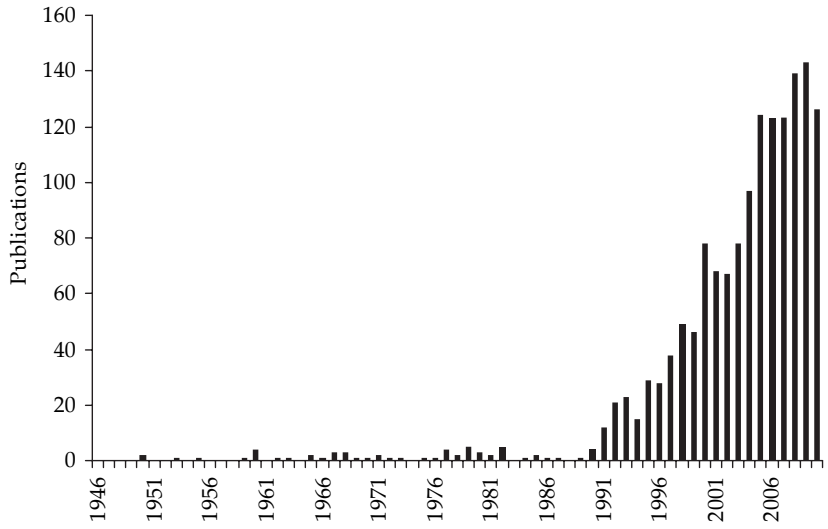

Figure 2. Papers on mycobacteria and food published from 1945 and indexed in the Web of Science database

Table 4. Institutions with 10 or more publications on mycobacteria and water (total number of institutions 1601)

	Institution	Number of records
1	Institute Pasteur	33
2	Centre of Disease Control & Prevention	30
3	Virginia Polytechnical Institute & State University	28
4	National Public Health Institute	26
5	University of Texas	21
6	University of Illinois	20
7	US Environmental Protection Agency	20
8	University of Helsinki	19
9–10	Veterinary Research Institute	17
9–10	Institute of Tropical Medicine	16
11	University of Colorado	16
12–13	Harvard University	15
12–13	Kuopio University Hospital	14
14	University of Michigan	12
15–19	University of Sao Paulo	12
15–19	University of Wurzburg	12
15–19	Veteran Affairs Medical Centre	12
15–19	Michigan State University	11
15–19	University of Kuopio	11
20–26	University of Tennessee	11
20–26	University of Washington	11
20–26	Institute Francais Petr	10
20–26	Queens University Belfast	10
20–26	University of California San Francisco	10
20–26	University of Cincinnati	10

Table 5. Countries with 10 or more publications on mycobacteria and water (total number of countries 79)

	Country	Number of records		Country	Number of records
1	USA	535	16	Czech Republic	27
2	France	142	17–18	Peoples Republic of China	25
3	England	122	17–18	Greece	20
4	Germany	94	19	South Korea	20
5	Canada	66	20–21	Portugal	19
6	Japan	63	20–21	Sweden	19
7	Spain	50	22–24	Denmark	15
8–9	India	46	22–24	Ghana	15
8–9	Australia	43	22–24	Scotland	15
10	Finland	41	25	Turkey	15
11–12	Switzerland	40	26–28	North Ireland	12
11–12	Italy	39	26–28	South Africa	12
13	Netherlands	39	26–28	Taiwan	12
14	Belgium	36	29	Poland	11
15	Brazil	35			

Table 6. Journals (Source Titles) with 10 or more publications on mycobacteria and water (total number of journals 564)

	Source	Number of records
1	Applied and Environmental Microbiology	101
2	Journal of Clinical Microbiology	44
3	Journal of Hospital Infection	32
4	Journal of Biological Chemistry	27
5	Clinical Infectious Diseases	22
6	Journal of Applied Microbiology	20
7–8	Biochemistry	19
7–8	Journal of Molecular Biology	17
9–11	Environmental Science & Technology	16
9–11	Applied Microbiology and Biotechnology	15
9–11	Journal of Microbiological Methods	15
12–14	Water Research	14
12–14	Emerging Infectious Diseases	12
12–14	FEMS Microbiology Letters	12
15	Infection Control and Hospital Epidemiology	12
16	Veterinarni Medicina	12
17	American Review of Respiratory Disease	10

pursued most with particular focus in the United States (Centre of Disease Control and Prevention, Virginia Polytechnical Institute and State University, National Public Health Institute, University of Texas, University of Illinois and US Environmental Protection Agency) with 20 and more published papers. Institute Pasteur, Paris, is the only institute outside the United States, which published more than 20 papers. More than 10 papers were published in 26 institutions from 29 countries (Tables 4 and 5).

The total number of authors was 5315, institutions 1601, and countries 79 (Table 2). Papers have been published in 564 journal titles (Table 6), with more than 30 in the following journals: Applied and Environmental Microbiology, Journal of Clinical Microbiology and Journal of Hospital Infection.

Table 7. Authors with 10 or more publications on mycobacteria and food (total number of authors 4814)

	Authors	Number of records		Authors	Number of records
1	Pavlik I	49	14–15	Singh SV	13
2	Grant IR	31	16–17	Godden SM	12
3	Rowe MT	28	16–17	Treml F	12
4–5	Collins MT	24	18–22	Adams A	11
4–5	Nielsen SS	24	18–22	Portaels F	11
6–7	Kent ML	17	18–22	Singh AV	11
6–7	Wells SI	17	18–22	Singh PK	11
8–9	Matlova L	16	18–22	Sohal JS	11
8–9	Stabel JR	16	23–27	Hejlicek K	10
10–13	Ball HI	14	23–27	Moravkova M	10
10–13	Colorni A	14	23–27	Richards RH	10
10–13	Delahay RJ	14	23–27	Smith SA	10
10–13	Dvorska L	14	23–27	Thompson KD	10
14–15	Bartos M	13			

Table 8. Institutions with 10 or more publications on mycobacteria and food (total number of institutions 1512)

	Institutions	Number of records
1	Veterinary Research Institute	54
2	Queens University Belfast	38
3	US Department of Agriculture Animal Research Service	32
4	University of Wisconsin	30
5	University of Minnesota	29
6	Oregon State University	26
7	University of Guelph	23
8	US Department of Agriculture	21
9–10	Michigan State University	19
9–10	University of Maryland	19
11	Cornell University	18
12–13	University of California Davis	17
12–13	University of Copenhagen	17
14	Central Science Laboratory	16
15–19	Centre of Disease Control & Prevention	14
15–19	Royal Veterinary & Agricultural University	14
15–19	Texas A&M University	14
15–19	University of Florida	14
15–19	US Food and Drug Administration	14
20–26	Colorado State University	13
20–26	Institute of Tropical Medicine	13
20–26	National Veterinary Institute	13
20–26	University of Pennsylvania	13
20–26	University of Stirling	13
20–26	University of Washington	13
20–26	Virginia Polytechnical Institute & State University	13
27–30	Animal Research Service	12
27–30	Pennsylvania State University	12
27–30	University of Colorado	12
27–30	University Prince Edward Island	12
31–36	Central Institute for Research on Goats	11
31–36	Iowa State University	11
31–36	Purdue University	11
31–36	University of California San Francisco	11
31–36	University College Dublin	11
31–36	Veterans Affairs Medical Centre	11
37–40	Institute Pasteur	10
37–40	Israel Oceanographic & Limnological Research	10
37–40	University of Tennessee	10
37–40	University of Zurich	10

Table 9. Countries with 10 or more publications on mycobacteria and food (total number of countries 84)

	Country	Number of records		Country	Number of records
1	USA	516	34–36	Greece	10
2	England	134	34–36	Iran	10
3	Germany	67	34–36	Portugal	10
4	Canada	65	16	Japan	33
5	Czech Republic	63	17–18	Belgium	29
6	Netherlands	55	17–18	Ireland	29
7	France	52	19	Brazil	26
8–9	Denmark	49	20–21	New Zealand	22
8–9	Italy	49	20–21	Turkey	22
10	Spain	48	22–24	Israel	20
11–12	North Ireland	45	22–24	Norway	20
11–12	Scotland	45	22–24	South Africa	20
13	India	43	25	Austria	18
14	Australia	41	26–28	Argentina	16
15	Switzerland	39	26–28	Mexico	16
30–31	Thailand	13	26–28	Poland	16
32–33	Peoples Republic of China	12	29	Ethiopia	15
32–33	Sweden	12	30–31	Finland	13

Table 10. Journals (Source Titles) with 10 or more publications on mycobacteria and food (total number of journals 531)

	Source	Number of records
1	Journal of Dairy Science	60
2	Applied and Environmental Microbiology	57
3	Veterinarni Medicina	44
4	Preventive Veterinary Medicine	42
5	Veterinary Microbiology	39
6	Journal of Fish Diseases	27
7–8	Diseases of Aquatic Organisms	23
7–8	Journal of Clinical Microbiology	23
9–11	American Journal of Veterinary Research	19
9–11	Infection and Immunity	19
9–11	International Journal of Food Microbiology	19
12–14	International Journal of Tuberculosis and Lung Disease	17
12–14	Journal of Food Protection	17
12–14	Letters in Applied Microbiology	17
15	Veterinary Record	15
16	Journal of Veterinary Diagnostic Investigation	14
17	Journal of The American Veterinary Medical Association	12
18–21	Clinical Infectious Diseases	11
18–21	Irish Veterinary Journal	11
18–21	Journal of Applied Microbiology	11
18–21	Veterinary Journal	11
22–24	FEMS Microbiology Letters	10
22–24	Tropical Animal Health and Production	10
22–24	Veterinary Immunology and Immunopathology	10

Table 11. ProMED-mail archive records, reporting mycobacteria as food risk for consumers (http://www.promedmail.org, accessed 25 November 2010)

Tuberculosis linked to *Mycobacterium bovis* in cheese
ProMED-mail (2005): MYCOBACTERIUM BOVIS, CHEESE – USA (NY): Alert. ProMED-mail 2005, 16 Mar: 20050316.0760
ProMED-mail (2005): MYCOBACTERIUM BOVIS, CHEESE – USA (NEW YORK) (02). ProMED-mail 2005, 18 Mar: 20050318.0785
ProMED-mail (2005): MYCOBACTERIUM BOVIS, CHEESE – USA (NEW YORK) (03). ProMED-mail 2005, 26 Jun: 20050626.1791
ProMED-mail (2008): MYCOBACTERIUM BOVIS, CHEESE – USA: (CALIFORNIA EX MEXICO). ProMED-mail 2008, 8 Jun: 20080608.1825
***Mycobacterium avium* risk to HIV positive and immunocompromized people**
ProMED-mail (2003): MYCOBACTERIUM AVIUM, SWINE – USA EX CANADA. ProMED-mail 2003, 3 Feb: 20030203.0295
***Mycobacterium bovis*, drug resistant**
ProMED-mail (2000): MYCOBACTERIUM BOVIS, DRUG RESISTANT – SPAIN (ZARAGOZA). ProMED-mail 2000, 3 May: 20000503.0677
***Mycobacterium simiae* in drinking water**
ProMED-mail (1998): MYCOBACTERIUM SIMIAE, HUMAN (02). ProMED-mail 1998, 24 Aug: 19980824.1679
***Mycobacterium avium* subsp. *paratuberculosis* in milk**
ProMED-mail (1998): MYCOBACTERIUM PARATUBERCULOSIS, MILK SURVEY – UK. ProMED-mail 1998, 11 Aug: 19980811.1580
ProMED-mail (1998): MYCOBACTERIUM PARATUBERCULOSIS, MILK SURVEY – UK (02). ProMED-mail 1998, 13 Aug: 19980813.1594
ProMED-mail (1998): MYCOBACTERIUM PARATUBERCULOSIS, MILK SURVEY – UK (03). ProMED-mail 1998, 15 Aug: 19980815.1628

Mycobacteria and food or feedstocks

In total 1486 papers have been published between 1950 and 2010 (Figure 2, Table 2), 668 (45%) during the last five years, indexed from 2006 to November 2010. Most papers were concerned with milk (626) and fish (381). Table 2 summarized papers sorted according to the key words used. Attention has been paid also to meat, cheese and vegetables. Mycobacteria in feed have been mentioned in 119 papers.

Authors who published ten or more papers, collected in the database for mycobacteria and food or feed are presented in Table 7, the institution of authors in Table 8, and countries in Table 9. It is evident, that according to the number of published papers research on mycobacteria is proceeding most intensely in the institutions of the US Department of Agriculture, Veterinary Research Institute, Brno, Queens University, Belfast, and the University of Wisconsin, from which more than 30 papers were published. The total number of authors was 4814, institutions 1512, and countries 84 (Table 2). Papers have been published in 531 journal titles (Table 10), more than 40 in the Journal of Dairy Science, Applied and Environmental Microbiology, Veterinarni Medicina and Preventive Veterinary Medicine.

A search of the ProMED-mail archive retrieved 60 records, some reporting mycobacteria as food risk for consumers (Table 11).

Mycobacteria in food and water are not under regulation according to EU legislation. Nevertheless, a report is present in the RASFF database, describing *Mycobacterium tuberculosis* in cattle from which milk was used to produce raw milk cheese from Germany, distributed to France, United Kingdom and the United States (RASFF Notification 2009.1227).

From the increasing number of publications it is evident that mycobacteria in food, feed and water are the subject of intense research. An understanding of how mycobacteria participate in human and animal infections and how they participate in immunomodulation and contribute to autoimmune and autoinflammatory diseases will likely lead to the implementation of regulatory measures to protect consumers against this public health risk.

REFERENCES

Adam K, Brulisauer F (2010): The application of food safety interventions in primary production of beef and lamb: A review. International Journal of Food Microbiology 141, S43–S52.

Anonymous (2010): Assessment of food as a source of exposure to Mycobacterium avium subspecies paratuberculosis (MAP). Journal of Food Protection 73, 1357–1397.

Argueta C, Yoder S, Holtzman AE, Aronson TW, Glover N, Berlin OGW, Stelma GN, Froman S, Tomasek P (2000): Isolation and identification of nontuberculous mycobacteria from foods as possible exposure sources. Journal of Food Protection 63, 930–933.

Chang CT, Wang LY, Liao CY, Huang SP (2002): Identification of nontuberculous mycobacteria existing in tap water by PCR-restriction fragment length polymorphism. Applied and Environmental Microbiology 68, 3159–3161.

Cirone K, Morsella C, Romano M, Paolicchi F (2007): Mycobacterium avium subsp. paratuberculosis in food and its relationship with Crohn's disease (in Spanish). Revista Argentina de Microbiologia 39, 57–68.

Collins MT (1997): Mycobacterium paratuberculosis: a potential food-borne pathogen? Journal of Dairy Science 80, 3445–3448.

Eltholth MM, Marsh VR, Van Winden S, Guitian FJ (2009): Contamination of food products with Mycobacterium avium paratuberculosis: a systematic review. Journal of Applied Microbiology 107, 1061–1071.

Falkinham JO (2009a): Surrounded by mycobacteria: nontuberculous mycobacteria in the human environment. Journal of Applied Microbiology 107, 356–367.

Falkinham JO (2009b): The biology of environmental mycobacteria. Environmental Microbiology Reports 1, 477–487.

Falkinham JO (2010): Impact of human activities on the ecology of nontuberculous mycobacteria. Future Microbiology 5, 951–960.

Grant IR (2006): Mycobacterium avium ssp. paratuberculosis in foods: current evidence and potential consequences. International Journal of Dairy Technology 59, 112–117.

Hruska K (2004): Research on paratuberculosis: Analysis of publications 1994–2004. Veterinarni Medicina 49, 271–282. http://www.vri.cz/docs/vetmed/49-8-271.pdf

Hruska K, Bartos M, Kralik P, Pavlik I (2005): Mycobacterium avium subsp. paratuberculosis in powdered infant milk: paratuberculosis in cattle – the public health problem to be solved. Veterinarni Medicina 50, 327-335. http://www.vri.cz/docs/vetmed/50-8-327.pdf

Kaevska M, Hruska K (2010a): Analysis of publications on paratuberculosis from 1995 to 2009 with emphasis on the period from 2005 to 2009. Veterinarni Medicina 55, 43–54. http://www.vri.cz/docs/vetmed/55-02-43.pdf

Kaevska M, Hruska K (2010b): Research on *Mycobacterium avium* during the period 1995 to 2009. Veterinarni Medicina 55, 473–482. http://www.vri.cz/docs/vetmed/55-10-473.pdf

Kasperbauer SH, Daley CL (2008): Diagnosis and treatment of infections due to Mycobacterium avium complex. Seminars in Respiratory and Critical Care Medicine 29, 569–576.

Kazda J, Pavlik I, Falkinham III JO, Hruska K (eds.) (2009): The Ecology of Mycobacteria: Impact on Animal's and Human's Health. 1st ed. Springer. XVIII, 565 pp.

McGrath EE, Blades Z, McCabe J, Jarry H, Anderson PB (2010): Nontuberculous mycobacteria and the lung: From suspicion to treatment. Lung 188, 269–282.

Mor-Mur M, Yuste J (2010): Emerging bacterial pathogens in meat and poultry: An overview. Food and Bioprocess Technology 3, 24–35.

Nacy C, Buckley M (2008): Mycobacterium avium paratuberculosis: Infrequent human pathogen or public health threat? A report from the American Academy of Microbiology, 1–37.

Nayak M, Kotian A, Marathe S, Chakravortty D (2009): Detection of microorganisms using biosensors-A smarter way towards detection techniques. Biosensors & Bioelectronics 25, 661–667.

Salah IB, Ghigo E, Drancourt M (2009): Free-living amoebae, a training field for macrophage resistance of mycobacteria. Clinical Microbiology and Infection 15, 894–905.

Slana I, Paolicchi F, Janstova B, Navratilova P, Pavlik I (2008): Detection methods for Mycobacterium avium subsp paratuberculosis in milk and milk products: a review. Veterinarni Medicina 53, 283–306. http://www.vri.cz/docs/vetmed/53-6-283.pdf

Thoen C, Lobue P, de Kantor I (2006): The importance of Mycobacterium bovis as a zoonosis. Veterinary Microbiology 112, 339–345.

Thoen CO, Lobue PA, Enarson DA, Kaneene JB, de Kantor IN (2009): Tuberculosis: a re-emerging disease in animals and humans. Veterinaria Italiana 45, 135–181.

van Ingen J, Boeree MJ, Dekhuijzen PNR, van Soolingen D (2009): Environmental sources of rapid growing nontuberculous mycobacteria causing disease in hu-

mans. Clinical Microbiology and Infection 15, 888–893.

van Ingen J, Wisselink HJ, Solt-Smits CB, Boeree MJ, van Soolingen D (2010): Isolation of mycobacteria other than Mycobacterium avium from porcine lymph nodes. Veterinary Microbiology 144, 250–253.

Yajko DM, Chin DP, Gonzalez PC, Nassos PS, Hopewell PC, Reingold AL, Horsburgh CR, Yakrus MA, Ostroff SM (1995): Mycobacterium-avium complex in water, food, and soil samples collected from the environment of hiv-infected individuals. Journal of Acquired Immune Deficiency Syndromes and Human Retrovirology 9, 176–182.

Yoder S, Argueta C, Holtzman A, Aronson T, Berlin OGW, Tomasek P, Glover N, Froman S, Stelma G (1999): PCR comparison of Mycobacterium avium isolates obtained from patients and foods. Applied and Environmental Microbiology 65, 2650–2653.

Received: 2010–10–29
Accepted after corrections: 2010–11–28

Corresponding Author:

Professor Karel Hruska, Veterinary Research Institute, Hudcova 70, 621 00 Brno, Czech Republic
E-mail: hruska@vri.cz