

Avian Tuberculosis

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Summary of general activities related to the disease

1. Test(s) in use/or available for the specified disease at your laboratory

Test (Total)	For	Specificity	Total
ELISA (120)	Antibodies	Sonicated antigen of <i>M. avium</i> subsp. <i>avium</i> (serotype 2)	120 domestic pigs
ELISA (120)	Antibodies	Sonicated antigen of <i>M. avium</i> subsp. <i>hominissuis</i> (serotype 8)	120 domestic pigs
Rapid agglutination (936)	Antibodies	Corpuscular antigen of <i>M. avium</i> subsp. <i>avium</i> (serotype 2)	931 domestic pigs 0 wild boar 4 wild ruminants 1 exotic birds
Rapid agglutination (174)	Antibodies	Corpuscular antigen of <i>M. avium</i> subsp. <i>hominissuis</i> (serotype 8)	169 domestic pigs 0 wild boar 4 wild ruminants 1 exotic birds
Rapid agglutination (6)	Antibodies	Corpuscular antigen of <i>M. intracellulare</i> (serotype 19)	2 domestic pigs 0 wild boar 4 wild ruminants 1 exotic birds
Culture examination (2 819)	Mycobacteria	Conventional culture technique (media without Mycobactin J)	1 011 environmental samples 1 328 pig tissues 88 tissues of other mammals 176 pig faeces 27 bird tissues 33 bird faeces 85 small terrestrial mammals tissues 71 poikilothermic animals
PCR Quadruplex (198)	Isolates	Quadruplex system for <i>M. avium</i> species differentiation ¹	16 <i>M. avium</i> subsp. <i>avium</i> 96 <i>M. avium</i> subsp. <i>hominissuis</i> 86 <i>M. avium</i> subsp. <i>paratuberculosis</i>
16S rRNA PCR (357)	Isolates	16S rRNA PCR system for <i>Mycobacterium</i> sp. and <i>M. avium</i> complex members identification ²	151 <i>M. avium</i> sp. 10 <i>M. intracellulare</i> 141 other atypical mycobacteria ³ 55 other bacteria
IS1245 RFLP (14)	Isolates	Standardized method ⁴	14 <i>M. avium</i> subsp. <i>hominissuis</i>
IS901 RFLP (7)	Isolates	Standardized method ⁵	5 <i>M. avium</i> subsp. <i>avium</i>

HAIN test (21)	Isolates	HAIN Lifescience, Nehren Germany	1 <i>M. peregrinum</i> 6 <i>M. szulgai/intermedium</i> 1 <i>M. fortuitum</i> 1 <i>M. chelonae / immunogenum</i> 7 <i>M. chelonae</i> 1 <i>M. kansasii</i> 3 <i>M. marinum</i> 1 <i>M. interjectum</i> 0 other atypical mycobacteria ³
16SrDNA sequencing (139)	Isolates	Differentiation of <i>Mycobacterium</i> sp. Harmsen et al. ⁶	1 <i>M. abscessus</i> 6 <i>M. arupense</i> 1 <i>M. celatum</i> 1 <i>M. engbaekii</i> 12 <i>M. fortuitum</i> 13 <i>M. gordonae</i> 1 <i>M. hiberniae</i> 6 <i>M. chelonae</i> 3 <i>M. chitae</i> 3 <i>M. interjectum</i> 12 <i>M. intracellulare</i> 4 <i>M. kansasii</i> 2 <i>M. kumamotoense</i> 1 <i>M. malmoense</i> 12 <i>M. marinum</i> 1 <i>M. nebraskense</i> 1 <i>M. neoaurum</i> 5 <i>M. nonchromogenicum</i> 1 <i>M. paraffinicum</i> 10 <i>M. peregrinum</i> 2 <i>M. porcinum</i> 1 <i>M. senegalense</i> 1 <i>M. simiae</i> 2 <i>M. szulgai</i> 7 <i>M. terrae</i> 4 <i>M. triviale</i> 1 <i>M. vaccae</i> 5 <i>M. xenopi</i> 1 <i>M. peregrinum/septicum</i> 3 <i>M. farcinogenes/ senegalense/fortuitum</i> 1 <i>M. porcinum/fortuitum</i> 1 <i>M. terrae</i> complex 14 <i>M. sp.</i>

¹ Home made technique for the detection and differentiation of members of *M. avium* species: Moravkova, M., Hlozek, P., Beran, V., Pavlik, I., Prezioso, S., Cuteri, V., Bartos, M.: Strategy for the detection and differentiation of *Mycobacterium avium* species isolates and heavily infected tissues. *Research in Veterinary Science*, paper in press.

² Wilton, S., Cousins, D.: Detection and identification of multiple mycobacterial pathogens by DNA amplification in a single tube. *PCR Methods Applications*, 1992, 1, 269-273.

³ Non-identified mycobacterial species other than members of *M. avium* species, *M. avium* complex and *M. tuberculosis* complex.

⁴ Van Soolingen, D., Bauer, J., Leão, S., Pavlik, I., Vincent, V., Rastogi, N., Gori, A., Bodmer, T., Garzelli, C., Garcia, M. J.: *IS1245* Restriction fragment length polymorphism typing of *Mycobacterium avium* isolates: proposal for standardization. *Journal of Clinical Microbiology*, 1998, 36 (10), 3051-3054.

⁵ Dvorska, L., Bull, T. J., Bartos, M., Matlova, L., Svastova, P., Weston, R. T., Kintr, J., Parmova, I., Van Soolingen, D., Pavlik, I.: A standardised restriction fragment length polymorphism (RFLP) method for typing *Mycobacterium avium* isolates links *IS901* with virulence for birds. *Journal of Microbiological Methods*, 2003, 55 (1), 11-27.

⁶ Harmsen, D., Dostal, S., Roth, A., Niemann, S., Rothganger, J., Sammeth, M., Albert, J., Frosch, M., Richter, E., 11-11-2003. RIDOM: comprehensive and public sequence database for identification of *Mycobacterium* species. *BMC Infect.Dis.* 3, 26

2. Production and distribution of diagnostic reagents

Antigens and PCR kits for *M. avium* complex and *M. avium* species members were produced (see the Table).

Antigens and PCR kits for *M. avium* complex and *M. avium* species members were produced for our laboratory (see the Table).

Activities specifically related to the mandate of OIE Reference Laboratories

3. International harmonisation and standardisation of methods for diagnostic testing or the production and testing of vaccines

During the year 2007 we have cooperated with Dr. Dr. Henk Wisselink Ph.D. (Research Scientist, Animal Sciences Group, Wageningen UR, Division of Infectious Diseases, LELYSTAD, The Netherlands) in the field of serological diagnostics of *M. avium* complex infections in pigs.

4. Preparation and supply of international reference standards for diagnostic tests or vaccines

None

5. Research and development of new procedures for diagnosis and control

Conventional “quadruplex” PCR system for the identification of members of *M. avium* species was routinely used. The paper is in press.

Quantitative real time PCR “triplex” system with an internal amplification control for the detection of *M. avium* subsp. *avium* and *M. avium* subsp. *hominissuis* is in testing now.

6. Collection, analysis and dissemination of epizootiological data relevant to international disease control

IS901 RFLP analysis was carried out on the 7 *M. avium* subsp. *avium* isolates from the following hosts in the Czech Republic: 3 domestic pig (*Sus scrofa domestica*), 1 sea eagle (*Haliaeetus albicilla*), 1 pigeon (non specified), 2 sparrow hawk (*Accipiter nisus*).

7. Provision of consultant expertise to OIE or to OIE Member Countries

None

8. Provision of scientific and technical training to personnel from other OIE Member Countries

MVSc. Edmealem Shitaye Jembere (Senior Epidemiologist responsible for exotic infectious diseases at Animal Health Department, Ministry of Agriculture Addis Ababa, Ethiopia), University of Veterinary and Pharmaceutical Sciences Brno, Faculty of Veterinary Medicine, 31.10.2004-31.8.2007: Study of epizootiology of infections caused by the members of *Mycobacterium avium* species in animals and birds.

Marija Kaevska, Faculty of Natural Sciences and Mathematics, Sts. Cyril and Methodius University, Skopje, Macedonia, 1.9.2006-30.6.2007: PCR detection of *Mycobacterium avium* subsp. *avium* in pig tissue samples.

Mgr. Tsilla Boisselet, Vienna University, 10.9.2007-31.3.2008: Diagnosis of mycobacterial infections and study of the distribution of different mycobacterial species in the environment

9. Provision of diagnostic testing facilities to other OIE Member Countries

none

10. Organisation of international scientific meetings on behalf of OIE or other international bodies

none

11. Participation in international scientific collaborative studies

■ *National grants*

Healthy animals and safety food as a basis for the health of human population, part: Mycobacterial infections of animals. Institutional Research Plan, 15.1.2004-31.12.2008. Ministry of Agriculture, the Czech Republic, No.: MZE 0002716201, Project leader: Prof. MVDr. Jiri Rubes, CSc.

Avian tuberculosis and mycobacterioses: The development of methods for their prevention and eradication by study of mycobacterial sources and pathogenesis of infection. National Program of Research (NPV), No. 1B53009, 1.1.2005-31.12.2008, Project leader: Prof. MVDr. Ivo Pavlik, CSc., VRI Brno, Czech Republic.

Efficiency of alternative methods for prevention of enteric diseases in pigs and determination of risks from an aspect of food safety. National Agency for Agricultural Research (NAZV), No. QH71054, 1.1.2007-31.12.20011, Project leader: Prof. MVDr. Ivo Pavlik, CSc., VRI Brno, Czech Republic.

Paratuberculosis in the Czech Republic: introduction of new methods for the detection of causative agent and monitoring of its spread and survival in studs, foodstuff and environment. National Agency for Agricultural Research (NAZV), No. QH81065, 1.1.2008-31.12.20012. Project leader: Prof. MVDr. Ivo Pavlik, CSc., VRI Brno, Czech Republic.

■ *European Union Grants:*

Veterinary Network of Laboratories Researching into Improved Diagnosis and Epidemiology of Mycobacterial Diseases. Project Acronym: VENOMYC, No. SSPE-CT-2004-501903, EU, Brussels, 1.10.2003-30.9.2008, Co-ordinator: Prof. Lucas Dominguez, Dpt. Patologia Animal I, Sanidad Animal Facultad de Veterinaria U.C.M., Madrid, Spain.

Control and prevention of emerging and future pathogens at cellular and molecular level throughout the food chain. Integrated Project. Project Acronym: PathogenCombat. No. FOOD-CT-2005-007081. Framework VI Programme. EC Brussels, 1.4.2005-31.3.2009. Co-ordinator: Prof. Mogens Jakobsen, The Royal Veterinary and Agricultural University, Dept. of Food Science, Food Microbiology, Copenhagen, Denmark.

Development of improved tools for detection of paratuberculosis in livestock, *M. avium* subsp. *paratuberculosis* in food and for the assessment of the risk of human exposure. Acronym: ParaTBTools. No. FP6-2004-FOOD-3B-023106. STREP project, 2006-2009. Co-ordinator: Dr. Douwe Bakker, DLO Institute for Animal Science and Health, Lelystad, Netherlands.

12. Publication and dissemination of information relevant to the work of OIE (including list of scientific publications, internet publishing activities, presentations at international conferences)

■ *Presentations at international conferences and meetings*

Kralik, P., Tesinska, I., Pavlik, I.: Molecular identification and detection of *Mycobacterium avium* complex and other mycobacteria. Seminar on paratuberculosis, sheep and goats, situation in Cyprus and relation with Public Health, Veterinary Services, Nicosia, Cyprus, 18.7.2007.

Pavlik, I., Mrlik, V., Matlova, I., Trcka, I., Beran, V., Kralik, P., Moravkova, M., Parmova, I., Svobodova, J., Mezensky, L., Slosarek, M., Havelkova, M., Kaustova, J., Mullerova, M., Holcikova, A., Capovova, I., Sterba, J.: Mycobacterial infections in children: the impact of *Mycobacterium avium* complex members and potentially pathogenic mycobacteria. Paediatrician Seminary, Hotel „Zlaty Andel“, Cesky Krumlov, Czech Republic, 24.4.2007 (invited to give lecture).

Pavlik, I., Tesinska, I., Kralik, P.: Paratuberculosis and other mycobacterial infections in domestic and wild

animals. Seminar on Paratuberculosis, sheep and goats, situation in Cyprus and relation with Public Health, Veterinary Services, Nicosia, 18.7.2007

■ **Scientific publications in peer-reviewed journals**

- Dvorska, L., Matlova, L., Ayele, W. Y., Fischer, O. A., Amemori, T., Weston, R. T., Alvarez, J., Beran, V., Moravkova, M., Pavlik, I.: Avian tuberculosis in naturally infected captive water birds of the Ardeidae and Threskiornithidae families studied by serotyping, IS901 RFLP typing and virulence for poultry. *Veterinary Microbiology*, 2007, 119, 366-374.
- Moravkova, M., Bartos, M., Dvorska-Bartosova, L., Beran, V., Parmova, I., Ocepek, M., Pate, M., Pavlik, I.: Genetic variability of *Mycobacterium avium* subsp. *avium* of pig isolates. *Veterinarni Medicina*, 2007, 52 (10), 430-436.
<http://www.vri.cz/docs/vetmed/52-10-430.pdf>
- Pavlik, I., Matlova, L., Gilar, M., Bartl, J., Parmova, I., Lysak, F., Alexa, M., Dvorska-Bartosova, L., Svec, V., Vrbas, V., Horvathova, A.: Isolation of conditionally pathogenic mycobacteria from the environment of one pig farm and the effectiveness of preventive measures between 1997 and 2003. *Veterinarni Medicina*, 2007, 52 (9), 392-404.
<http://www.vri.cz/docs/vetmed/52-9-392.pdf>
- Shitaye, J.E., Matlova, L., Horvathova, A., Moravkova, M., Dvorska-Bartosova, L., Treml, F., Lamka, J., Pavlik, I.: *Mycobacterium avium* subsp. *avium* distribution studied in a naturally infected hen flock and in the environment by culture, serotyping and IS901 RFLP methods. *Veterinary microbiology*, 2008, 127, 155-164.
- Skoric, M., Shitaye, J. E., Halouzka, R., Fictum, P., Trcka, I., Heroldova, M., Tkadlec, E., Pavlik, I.: Tuberculous and tuberculoid lesions in free living small terrestrial mammals and the risk of infection to humans and animals: a review. *Veterinarni Medicina*, 2007, 52 (4), 144-161.
<http://www.vri.cz/docs/vetmed/52-4-144.pdf>

■ **Other communications**

Organisation of workshops:

„Application of real time PCR in practice“, 11. – 12. 10. 2007, Veterinary Research Institute, Brno, Czech Republic, 15 presentation/6 presentation were performed by members from our lab, 70 participants.

Participation in workshop organised by SAFOODNET „Detection and identification of harmful microbes“ 10. – 12. 12. 2007, Veterinary Research Institute, Brno, Czech Republic, practical demonstration of the molecular methods used in our laboratory.

Moravkova, M., Pavlik, I.: Molecular epidemiology of mycobacterial infections. „Safoodnet“ workshop, 10. - 12. 2007, Veterinary Research Institute, Brno, Czech Republic.

■ **Other publications related to mycobacteriology**

Ikonomopoulos, J., Balaskas, C., Kantzoura, B., Fragiadaki, E., Pavlik, I., Bartos, M., Lukas, J. C., Gazouli, M.: Comparative evaluation of positive tests to *Mycobacterium avium* subsp. *paratuberculosis* in clinically healthy sheep and goats in South-West Greece using molecular techniques, serology, and culture. *The Veterinary Journal*, 2007, 174, 337-343.

Mrlik, V., Pavlik, I.: Bovine tuberculosis in badgers in Ireland. *Veterinarstvi*, 2007, 57 (1), 55-56 (in Czech).

Shitaye, J. E., Tsegaye, W., Pavlik, I.: Bovine tuberculosis infection in animal and human populations in Ethiopia: a review. *Veterinarni Medicina*, 2007, 52 (8), 317-332.

<http://www.vri.cz/docs/vetmed/52-8-317.pdf>

Trcka, I., Lamka, J., Kopecna, M., Pavlik, I.: Tuberculosis in wild boar in the Czech Republic. *Lesu zdar*, 2007, 1, 28-29 (in Czech).