Biological agents as occupational hazards – selected issues

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Abstract
There are two main groups of biological agents regarded as occupational hazards: allergenic and/or toxic agents forming bioaerosols, and agents causing zoonoses and other infectious diseases. Bioaerosols occurring in the agricultural work environments comprise: bacteria, fungi, high molecular polymers produced by bacteria (endotoxin) or by fungi (β-glucans), low molecular secondary metabolites of fungi (mycotoxins, volatile organic compounds) and various particles of plant and animal origin. All these agents could be a cause of allergic and/or immunotoxic occupational diseases of respiratory organ (airways inflammation, rhinitis, toxic pneumonitis, hypersensitivity pneumonitis and asthma), conjunctivitis and dermatitis in exposed workers. Very important among zoonotic agents causing occupational diseases are those causing tick-borne diseases: Lyme borreliosis, anaplasmosis, babesiosis, bartonellosis. Agricultural workers in tropical zones are exposed to mosquito bites causing malaria, the most prevalent vector-borne disease in the world. The group of agents causing other, basically not vector-borne zoonoses, comprises those evoking emerging or re-emerging diseases of global concern, such as: hantaviral diseases, avian and swine influenza, Q fever, leptospiroses, staphylococcal diseases caused by the methicillin-resistant Staphylococcus aureus (MRSA) strains, and diseases caused by parasitic protozoa. Among other infectious, non-zoonotic agents, the greatest hazard for health care workers pose the blood-borne human hepatitis and immunodeficiency viruses (HBV, HCV, HIV). Of interest are also bacteria causing legionellosis in people occupationally exposed to droplet aerosols, mainly from warm water.

Key words
biological agents, occupational hazards, bioaerosols, allergy, toxins, viruses, bacteria, fungi, infectious diseases, zoonoses, tick-borne diseases

INTRODUCTION

There are two main groups of biological agents regarded as occupational hazards:
1) allergenic and/or toxic agents forming bioaerosols (bacteria, endotoxin, fungi, mycotoxins, β-glucans, particles of plant and animal origin) causing occupational diseases of the respiratory tract, conjunctiva and skin, mostly in agricultural workers; and 2) agents causing zoonoses and other infectious diseases that could be spread by tick or insect vectors, by the airborne route, by the alimentary route, or immediately by contact with skin.

ALLERGENIC AND/OR TOXIC AGENTS IN BIOAEROSOLS

General. Bioaerosols are biological particles of organic dust and/or droplets suspended in the air - viruses, bacteria, endotoxin, fungi, β-glucans, secondary metabolites of fungi, plant particles, particles of faeces and the bodies of mites and insects, particles of epithel, feather, hair, faeces, and urine of birds and mammals. They are often respirable, and in humans may induce disorders of the respiratory system (inflammation of the airways, rhinitis, toxic pneumonitis, hypersensitivity pneumonitis and asthma), conjunctivitis and dermatitis [1-3]. Bioaerosols are a main health problem in agriculture and the agricultural industry [4-6], and may be also an occupational risk factor in many other work environments, such as: medical and veterinary facilities, diagnostic laboratories [7], sewage treatment plants [8], waste sorting plants [9,10], plants producing biofuel from rape [11], the metallurgical industry [12], libraries [13], art conservation [14], and many others. Recently, the total inflammatory potential of bioaerosols could be assessed on the basis of granulocyte or whole blood response to bioaerosol or organic dust sample [15,16].

Wood dust, mostly from deciduous wood, is a known inducer of cancer in the nasal cavity [17]. Jacobsen et al. [17,18] in their thorough reviews demonstrate that wood dust exposure is also a risk factor for the development of non-malignant respiratory disorders, such as asthma, bronchitis, rhino-conjunctivitis, and acute and chronic impairment of lung function.

Bacteria and endotoxin. The air of agricultural settings could be heavily polluted with bacteria and endotoxin, the high molecular, heteropolymer lipopolysaccharide present in the outer membrane of Gram-negative bacteria [4,6,11,19]. Pantoaea agglomerans, a Gram- negative bacterium common on the surface of plants, has been experimentally identified as a producer of extremely strong allergens and endotoxin [20-22]. High levels of endotoxin have been recorded at grain threshing and sieving [4,23], flax threshing [4,24], herb processing [4,11,23], wood processing [25], waste collection and sorting [10], handling dry sludge [26], on pig farms [6,27,28], in cucumber and tomato nurseries [29], in plants using straw and wood chips as biofuel [30], and in many other work environments [2]. In the lungs of exposed workers, endotoxin evokes an inflammatory process mediated through the CD14 receptor and Toll Like Receptor 4 (TLR-4), leading to impairment of lung function, bronchitis, asthma, and toxic pneumonitis (organic dust toxic syndrome) [2,3].
β-1,3-glucan is an immunologically-active glucose polymer that occurs in the cell wall of mould fungi [2,3]. It is an important agent causing the development of pulmonary diseases, both of an inflammatory and an allergic nature [3]. β-1,3-glucan can induce Th1 as well as Th2-driven immune responses [3]. Recently, Wu et al. [55] demonstrated that automobiles are potentially a significant source of β-1,3-glucan and endotoxin exposure that could be of importance for asthmatics.

Recent papers also provide new data on the lesser known potentially pathogenic fungal species and toxins. Thus, the species Botrytis cinerea, in spite of low prevalence in ambient air, should be regarded as an important hazardous agent because of its strong allergenic properties [36]. This species infects the grapevine, and was identified as a cause of specific form of hypersensitivity pneumonitis described as “wine-grower’s lung” [56,57].

Pollen. Exposure to pollen grains, mainly to those produced by grasses, trees, and weeds, may be a cause of IgE-mediated pollinosis in occupational and general populations. The concentration of pollen in the air depends on geographic location, season of the year, and meteorological parameters [58-61]. Cases of occupational pollinosis have been described, mainly in greenhouse workers exposed to the pollen of bell pepper [62], strawberries [63], chrysanthemum [64], and various vegetables, such as sugar beet [65], cauliflower and broccoli [66]. The disease may occur also in agricultural workers toiling in open fields, such as those cultivating rice in India [67].

Arthropods. Mites are the most important source of allergens, which are mostly enzymes occurring in faecal pellets. The Acaridae species living in stores evoke rhinoconjunctivitis, dermatitis and asthma in agricultural workers, while the Pyroglyphidae species occurring in the dust of dwellings, evoke similar symptoms in home inhabitants [68-70]. Allergy to insects may appear as a reaction to inhaled particles or a reaction to venom, mostly after bee or wasp stings. Karakiş et al. [71] showed that allergy to hymenoptera venom has a genetic background and may be associated with classes of human leukocyte allergy (HLA).

AGENTS CAUSING VECTOR-BORNE ZOONOSSES

Among the zoonotic agents causing occupational diseases, those transmitted by ticks, are very important, in particular the sphaeriditessel Borrelia burgdorferi which are the common cause of occupational Lyme borreliosis in forestry and agricultural workers [72-75]. The other potentially occupational tick-borne diseases are: tick-borne encephalitis, anaplasmosis, ehrlichiosis, Rocky Mountains spotted fever, babesiosis, and bartonellosis [73,76-81]. The reservoir of tick-borne pathogens are wild and domestic mammals and birds. In Europe, the main vector of these pathogens is Ixodes ricinus, whereas in North America the main vectors are Ixodes scapularis and Dermacentor andersoni. A new epidemiological problem for European and North American countries is the growing import of exotic reptiles, mainly from Africa, which are carriers of novel species of ticks that could be potentially infected with novel pathogens [82,83].
Agricultural workers in tropical zones are exposed to mosquito bites causing malaria, the most prevalent vector-borne disease in the world, or infection with parasitic nematodes.

*Borrelia burgdorferi* is a spirochete causing Lyme borreliosis, the most common vector-borne disease in both Europe and North America [73,74,84,85]. In Poland in 2009, it caused as many as 10,333 cases, of which 664 were certified as an occupational disease [75]. The physiological flexibility of *B. burgdorferi* enables its reproduction in many organs and tissues of infected mammals and birds [74,84,86,87]. Changes in the synthesis of outer proteins (Osp) are the main strategy of *B. burgdorferi* to evade host response are associated also with the large number of plasmids (in spite of small genome), rare in other bacteria, which show a high variability as a result of recombination with each other [74]. Mosquitoes are considered an accessory vector of *B. burgdorferi*, but examination of adult specimens and larvae showed only a very low prevalence of infection [89].

*B. burgdorferi sensu lato* is a collective species which comprises a number of genospecies, of which the most important are *B. burgdorferi sensu stricto*, *B. afzelii* and *B. garinii* that in humans cause arthritis, skin infections and neural infections [73,74,85]. The pathogens that cause Lyme borreliosis, human anaplasmosis, babesiosis and tick-borne encephalitis, may coexist in *Ixodes* ticks and different animal species [79-81,90-92].

**Babesia spp.** These are intraerythrocytic protozoa of the Phylum Apicomplexa causing a zoonotic disease – babesiosis. Infection may lead to lysis of erythrocytes resulting in haemolytic anaemia which, in severe cases, may lead to organ failure and death, particularly in immunocompromised individuals [101]. Human infections are caused mostly by the species *Babesia microti* and *Babesia divergens*. The disease occurs mostly in the USA, but cases have also been reported in several European countries, in Egypt, India, Japan, Korea, Taiwan, and South Africa [101]. Of importance in veterinary medicine is *B. canis*, causing canine babesiosis [102].

**Bartonella spp.** These are intracellular Gram-negative bacteria that show preference for erythrocytes and endothelial cells in humans. The genus *Bartonella* currently comprises over 20 identified species of which about half are known to infect humans causing bartonellosis [103-105]. Members of this genus are involved in a variety of human diseases, such as Carrion’s disease, cat scratch disease, trench fever, bacillary angiomatosis, endocarditis, pericarditis and neuroretinitis. Humans are natural hosts for two species: *Bartonella bacilliformis* and *Bartonella quintana*, but many other animal-associated *Bartonella* strains can also cause disease in humans [103]. The evidence for ticks as a vector of *Bartonella* is circumstantial but fairly strong. Cotté et al. [106] infected experimentally *Ixodes ricinus* with *B. henselae* and demonstrated transmission of the bacterium across developmental stages, migration or multiplication of *B. henselae* in salivary glands, and transmission of viable and infective *B. henselae* from ticks to blood. These results provide evidence that I. ricinus is a competent vector for *B. henselae* [106].

**Agents causing other vector-borne zoonoses.** In the tropical zone, protozoa *Plasmodium* spp. transmitted by mosquito vectors cause malaria, the most common vector-borne disease in the world with 247 million cases, and nearly one million deaths in the year 2008 [107]. Performing agricultural work is considered to increase the risk of contracting malaria [108], but there are no exact data on this subject. Promising effects have been shown in combating malaria in Benin with use of an integrated vector management concept, which includes control of mosquito larvae with bio-insecticides obtained from the *Bacillus* species [109].

Agricultural workers in tropical zones are also under risk of contracting diseases caused by filarial nematodes transmitted by mosquitoes or blackflies (*Wuchereria bancrofti*, *Brugia malayi*, *Onchocerca volvulus*) [29]. In central and eastern Europe, cases of human dirofilariosis caused by the species *Dirofilaria repens* and *Dicrofilaria immitis* are noted as an emerging zoonosis [110]. Dirofilariosis, transmitted by mosquitoes, is a common infection in dogs and cats in southern Europe, Asia and America. In humans, immature worms of *D. immitis* are responsible for pulmonary dirofilariosis, whereas subcutaneous or ocular changes are caused by *D. repens* [111]. Due to global warming, the disease is spreading northwards and has been reported from Hungary, Switzerland, Russia and Slovakia [110].

**AGENTS CAUSING OTHER ZOONOSES**

To this group belong agents causing emerging or re-emerging zoonoses of global concern, such as those discussed...
individually below: hantaviral diseases, avian and swine influenza, Q fever, leptospiroses, staphylococcal diseases caused by the methicillin-resistant *Staphylococcus aureus* (MRSA) strains, cryptosporidiosis and toxoplasmosis.

**Hantaviruses.** Infections caused by these RNA-viruses pose an increasing global problem and are regarded as 'emerging infectious diseases' [112]. In Eurasia, two forms of the disease are distinguished: 1) severe Haemorrhagic Fever with Renal Syndrome (HFRS), caused by Hantaan virus (HTNV), Dobrava virus (DOBV) and Seoul virus (SEOV), with the mortality rate amounting up to 20%; and 2) the considerably milder Nephropathia Epidemica (NE) caused mainly by Puuimala virus (PUUV) [112].

In America, the very severe Hantavirus Cardio-Pulmonary Syndrome (HCPS) occurs, caused mainly by Sin Nombre virus (SNV) and Andes virus (ANDV). Small rodents are both reservoir and vectors of the disease. It has been demonstrated that the increase of HDV cases in particular years is preceded by rodent propagation associated with rich abundance of beechnast, a source of their food. Recently, the first focus of hantavirus disease (HDV) has been established in the Carpathian mountains in southeast Poland, close to foci described earlier in Slovakia [113]. A total of 13 serologically-confirmed clinical cases of HDV were described, of which 10 were HFRS cases caused by DOBV, and 3 were NE cases caused by PUUV [112].

**Zoonotic influenza viruses.** The highly pathogenic avian influenza A(H5N1) virus caused between 2003 – 9 August 2011 a total of 564 human infections in 15 countries of Asia and Africa, of which 330 (58.5%) resulted in death [114]. To date in Europe, no infections caused by avian influenza virus have been registered, except for the occupational epidemic among poultry breeders that occurred in the year 2003 in the Netherlands, caused by a highly pathogenic virus of another subtype A(H7N7) [115]. The highly pathogenic avian influenza A(H5N1) virus caused infections in wild birds and birds kept on farms in many European countries, including Poland, but laboratory tests did not detect any human cases, in spite of confirmed occupational exposure to infected poultry [116]. Nevertheless, there is a strong necessity for the awareness of medical and laboratory staff, as well as the awareness of some occupational groups about human infections with avian influenza viruses, including the importance of seasonal influenza vaccination [116].

In April 2009, an epidemic outbreak caused by the swine A(H1N1) influenza virus occurred in Mexico that rapidly spread worldwide. It was determined that the emerging virus was, in fact, a mutant, the so-called ‘triple reassortant virus’ containing swine, avian and human influenza A genes. It is estimated that during the first months of this epidemic the A(H1N1) influenza virus caused over 200,000 human infections and circa 2,000 deaths [117]. In June 2009, the World Health Organization (WHO) declared a global pandemic caused by the A(H1N1) influenza virus. International collaboration brought good results and the following year, in August 2010, the WHO announced the end of pandemic. In most cases, the pandemic A(H1N1)/2009 influenza virus caused milder illness compared to seasonal influenza viruses [118]. The occupational groups exposed to infection with the pandemic virus were mostly medical and veterinarian workers and swine breeders [119].

**Coxiella burnetii.** This small Gram-negative bacterium, causing Q fever and now classified within Legionellales, occurs in a great range of mammals, birds and arthropods [120]. In humans, the acute stage of the disease develops mostly as influenza-like fever, in 30-50% accompanied by interstitial pneumonia, while the chronic stage may appear as a high risk endocarditis [121]. The most important source of this bacterium for humans are domestic ruminants (cattle, sheep, and goats) from which pathogens are transmitted by the airborne route [120,122]. Dorko et al. [123] found a high prevalence of antibodies to *C. burnetii* among sheep in eastern Slovakia, and an increase of seropositivity in the course of 10 years. The potential reservoirs of *C. burnetii* may be also wild ruminants living in zoos [124]. Q fever - because of a recent epidemic in the Netherlands in 2007-2010 - is currently of great concern in Europe [125]. With over 4,000 human cases it is regarded as the greatest epidemic ever recorded [122]. The disease is spread mostly by infectious airborne particles released during abortions in goats [125]. Q fever is regarded as a neglected disease [120] which is properly diagnosed only in a small percent of cases, estimated by Knap [121] in Poland as only 1%. This is a typical occupational disease occurring mostly in farmers, veterinarians, tanners, furriers, butchers and dairy workers [120,121]. Monno et al. [126] found a very high prevalence of antibodies to *C. burnetii* among animal breeding farmers and veterinarians in southern Italy.

**Leptospira spp.** Leptospiroes are thin, helical bacteria classified into at least 12 pathogenic and 4 saprophytic species, with more than 250 pathogenic serovars [127]. Leptospirosis is regarded as the most widespread zoonosis in the world, and a re-emerging public health problem, particularly in the large urban centres of developing countries [128,129]. Depending on the serovar, the disease could be a mild, flu-like illness, or a severe disorder characterized by multi-organ system complications leading to death. Yet, the disease is severely neglected and the estimated incidence of about half a million severe human cases annually is probably an underestimate [130]. Humans most commonly become infected through occupational, recreational, or domestic contact with the urine of carrier animals, either directly or via contaminated water or soil [127]. *Leptospira* spp. present an occupational risk to agricultural workers, slaughterhouse workers, sewage workers, veterinarians, and other professions [1]. The mechanisms of pathogenesis of *Leptospira* are largely unknown [128]. With global climate change, extreme weather events such as cyclones and floods are expected to occur with increasing frequency and greater intensity, and may potentially result in an upsurge in the disease incidence as well as the magnitude of leptospirosis outbreaks [131].

**Methicillin-resistant *Staphylococcus aureus* (MRSA).** MRSA infection can cause severe health problems in health care workers that may lead to long-term incapacity [132]. People occupationally exposed to livestock are under risk of nasal colonization with MRSA strains [133]. In the Netherlands, the prevalence of MRSA in pig farmers was estimated to be 29% [134] compared to circa 0.1% in general population. In the USA, however, the exposure of pig farmers to MRSA is much lower [133].

**Cryptosporidium spp.** This protozoan causes cryptosporidiosis, a common zoonosis occurring as diarrhea.
in humans and animals [135]. It has been estimated that worldwide over 90% of the infections in people are caused by *C. hominis* and *C. parvum* [136]. In immuno-compromised individuals infection can become chronic, and potentially deadly. *C. parvum* infections are the direct cause of death for 15% of HIV-infected persons [137].

Transmission of the parasite can occur from person-to-person, by ingestion of contaminated water and food or by contact with contaminated surfaces. Transmission can be via direct contact with animals or indirect, such as that following the contamination of drinking water. Among animal species, cattle are the main reservoir for humans. It has been estimated that in 50% or more of all sheds for dairy calves there are detectable numbers of infective oocysts, and the parasite was present on more than 90% of all US dairy farms. Thus, *Cryptosporidium* may represent an occupational hazard to farmers, particularly to those working with calves [138].

Wild animals (small rodents, beavers, deer, wolves) represent a potentially significant source of environmental contamination and reservoir of the disease for domestic livestock and humans [139]. Application of molecular methods makes possible the estimation of the zoontic sources of infection for humans. PCR and sequence analyses allow genetic characterization of the *Cryptosporidium* species, which in turn helps to determine the source of infection for humans and animals [140].

**Toxoplasma gondii.** This is the agent of toxoplasmosis, and is a parasitic protozoan widespread in the environment. The course of *T. gondii* infection in healthy humans is mostly subclinical, manifesting only with the presence of specific antibodies. *T. gondii* infection may be especially dangerous for pregnant women with respect to the possibility of occurrence of congenital defects in the foetus, as well as for people with decreased immunity, in whom it may lead to severe systemic changes, or even death. In a mild form, toxoplasmosis may manifests as retinitis or chorioiditis [141,142].

Cats are definitive hosts of the parasite and play a special role in the spread of *T. gondii* infection, contaminating the environment with resistant oocysts, the dispersive forms of this parasite. As a large number of species of mammals and birds can be intermediate hosts of *T. gondii*, the possible sources of infection are multiple. Humans acquire toxoplasmosis mostly via the alimentary route, by eating raw meat (mainly pork) containing cysts of *T. gondii*, or by consuming other foods or water contaminated with oocysts of the parasite [143]. A number of waterborne cases of toxoplasmosis in humans have also been described. The results of a study from rural areas of eastern Poland proved that shallow household wells, which are often the only source of water and are not covered by examinations for pathogenic protozoans, pose a potential risk of infection [144,145].

Toxoplasmosis may constitute a serious epidemiological problem, particularly in the rural environment, where there are many cross-routes of *Toxoplasma* transmission [144,145]. The high risk of *T. gondii* infection for rural inhabitants can result from frequent contact with soil, and other potential sources of *Toxoplasma*. The results of seroepidemiological study in the Lublin region of eastern Poland [144,145] showed that people living on farms were infected with *Toxoplasma gondii* significantly more often than urban dwellers, and that the infection rate in farm inhabitants increased significantly with age. Other results of this work indicate that one of the reasons for the common occurrence of *Toxoplasma* infection among farm inhabitants may be the close contact with domestic animals (and with their tissues) which could be infected with *T. gondii* at high rates (pigs - 17.9%, cats - 75.0%, goats - 71.4%) [145].

Additional routes of *T. gondii* transmission should be also considered as a threat to human health, including infection by arthropods. In the study of *Ixodes ricinus* ticks collected from Lublin region and Szczecin region (western Poland), the prevalence of tick infection with *T. gondii* was 12.6% and 12.7%, respectively [146,147]. RFLP PCR test detected that type I and atypical *T. gondii* strains were the most common in ticks [147].

**AGENTS CAUSING OTHER INFECTIOUS DISEASES**

In these group of agents, the greatest problem for health care workers is posed by the blood-borne human hepatitis and immunodeficiency viruses (HBV, HCV, HIV) which were discussed in an earlier article [148]. Among other infectious agents causing occupational non-zoonotic diseases, of interest are the bacteria causing legionellosis in humans occupationally exposed to droplet aerosols, mainly from warm water. These infectious agents are described below.

**Legionella spp.** *Legionella* is a fastidious Gram-negative bacterium developing mostly in water, less often in soil and organic matter. Some species are pathogenic for humans and may cause respiratory disease (legionellosis, Legionnaires’ disease), appearing as an atypical pneumonia or influenza-like illness (Pontiac fever) after inhalation of bacteria-laden droplet aerosol or dust [149]. *Legionella* occurs commonly in water distribution and air conditioning systems. The highest levels of colonization by *L. pneumophila* were found at water temperatures from 30-35°C, whereas at temperatures ranging from 55-60°C the concentrations of these bacteria dropped below the threshold limit values [150].

People exposed to an infectious aerosol when bathing or performing various occupations are liable to contract legionellosis. The increased risk of exposure to *Legionella* has been reported among workers employed at cooling towers, oil drilling platforms, automotive plants, plastics factories, ship repair facilities, sewage treatment plants, and dental facilities, as well as among gardeners, miners, turbine operators, plumbers, construction workers, subway personnel, and railway conductors [149]. Recent research carried out in Turkey and Japan demonstrated that vehicle drivers, in particular professional bus drivers, who are exposed for a long time to car air-conditioning and air-circulating systems, are under increased risk of contracting legionellosis [151,152].

**REFERENCES**


