Chapter 6
Spacecraft Microbiology

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6.1 Introduction

During spaceflights, the immune system is one of the most affected systems of the human body (Ullrich and Paulsen 2011). To determine the medical risks of long-term spaceflights and to develop prophylactic and therapeutic arrangements, it is important to know the microbial flora on board of a spacecraft or space station and its specific factors influencing this microflora. It is well known from several space missions that crew members suffered from bacterial and viral infections like influenza, *Pseudomonas aeruginosa*, and B streptococci. Also on long-term habitation on space station Mir and ISS, astronauts suffered from acute airway infections, conjunctivitis, and dental infections, and also reactivation of the Epstein-Barr virus was observed. An overview of microbial infections, pathogens, and general observations is given in Table 6.1. Figure 6.1 shows the variables impacting the risk of infections and their transmission during space travel, on which the following headings are related to.

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Table 6.1 Microbiological situation aboard of spacecrafts and space stations

Microbial pathogens, infections, and colonizations	Spacecraft
Occurrence of conjunctivitis, dental infections, acute respiratory infections (Ball and Evans 2001)	Mir
Occurrence of different bacterial and viral infections with influenza, *Pseudomonas aeruginos*a, B streptococci (Sonnenfeld 2002)	Apollo
Reactivation of Epstein-Barr virus (Pierson et al. 2005)	Space Shuttle
Changes in intestinal (Lencner et al. 1984), oral (Brown et al. 1976), and nasal (Decelle and Taylor 1976) microflora	Skylab Apollo Soviet Cosmonauts
Decrease of apathogen and increase of pathogen bacteria in nasal flora (Nefedov et al. 1971)	Soviet Cosmonauts
Breathing air, *Staphylococcus* sp., *Aspergillus* sp., *Penicillium* sp.; water, *Sphingomonas* sp., *Methylobacterium* sp.; surfaces, *Staphylococcus* sp., *Aspergillus* sp., *Cladosporium* sp. (Novikova et al. 2006)	ISS
Gram-positive and gram-negative microorganisms, Actinomyces and fungi; potable water, evidence of DNA pathogenic microorganisms (La Duc et al. 2004)	ISS
Activation of opportunistic pathogens, increased density of aerobic gram-negative bacteria and staphylococci on skin, in upper airways and colon (Ilyin 2005)	Salyut, Mir
58 forms of bacteria, 36 forms of molds and yeasts, significant percentage of pathogenic microorganisms, many of them with biodestructive properties (Viktorov et al. 1992)	Mir
Condensed water: *Serratia liquefaciens*, *Yersinia enterocolitica*, *Pseudomonadaceae*, *Stenotrophomonas maltophilia* (Harada 2001)	Mir
Condensed water: *Escherichia coli*, *Serratia marcescens*, amoeba resembling *Acanthamoeba* or *Hartmannella species* (Ott et al. 2004)	Mir
108 forms of bacteria, 206 forms of fungi, among them pathogenic and material destructive forms (Novikova 2004)	Mir
Expansion of *Penicillium chrysogenum* (Viktorov et al. 1998)	Mir
Potential biodegradation of polymers (Novikova 2004)	Mir
Higher caries incidence in astronauts (*Streptococcus mutans*) (Cheng et al. 2014)	ISS

ISS International Space Station, *DNA* deoxyribonucleic acid

6.2 Microbial Colonization of Spacecrafts

In an orbiting spacecraft, airborne microorganisms (and dust) do not settle due to the absence of gravity, and thermodiffusion or electrostatic forces gain in importance. This results in a more persistent (bio)aerosol and higher microbial contamination level in cabin air, and thus a continuous active removal of the aerosols from the air is necessary (Van Houdt et al. 2012).

On board of the Soviet space station Mir, several microbial studies were established during its operating time from 1986 to 2001 (Table 6.1). In one study, 58 forms of bacteria and 36 forms of mold and yeast forms were found, of which a significant part were pathogen microorganisms (Viktorov et al. 1992). Fungi types with material destructive properties were also identified. Another study found 108 types of bacteria

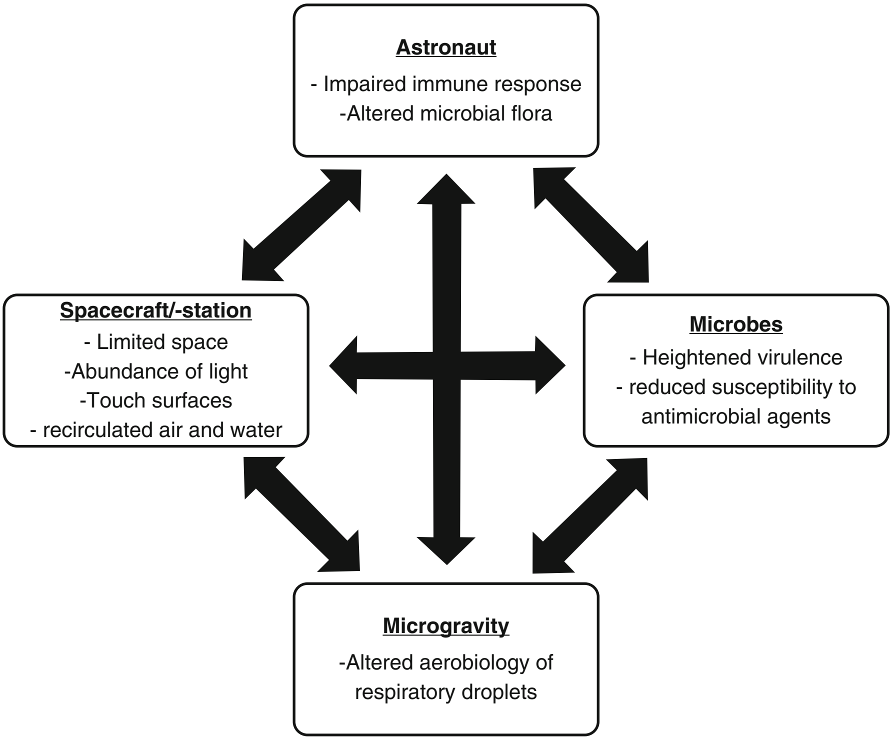

Fig. 6.1 Variables impacting the risk of infections and their transmission during space travel (Modified from Mermel 2012)

and 206 types of fungi, again with many pathogen and/or material destructive types among them (Novikova 2004). Condensed water was contaminated with *Serratia liquefaciens*, *Yersinia enterocolitica*, and *Stenotrophomonas maltophilia* and even radioresistant bacteria (Ott et al. 2004). Examination of optically hazy condensed water from behind instrumental panels aboard of the Mir revealed enterobacteria, *Escherichia coli*, *Serratia marcescens*, *Legionella* sp., spirochetes, protozoa, and mites (Ott et al. 2004). Bacteriofungal associations primarily resided on surfaces and structural materials of space interiors and equipment which gather anthropogenic organic compounds and air condensate enough to allow a full vegetative cycle and reproduction of heterotrophic microorganisms and molds (Novikova 2004). The microbial loading dynamic did not have linearly progressing character within the isolated environment of the Mir, but it was a wavy process of alternations of the microflora with changes of the dominating species (Novikova 2004). Fluctuating alterations in solar activity, degree of radiation, and gradients of magnetic fields can be considered parameters capable of initiating quantitative variations in the microflora of the space station (Novikova 2004). Also the ISS is by now severely colonized by microorganisms (Table 6.1): a 6-year study about the microbial environment on board of the ISS revealed that *Staphylococcus* sp., *Aspergillus* sp., and *Penicillium* sp. within the breathing air, *Sphingomonas* sp. and *Methylobacterium* sp. within the

Fig. 6.2 Mold on panel (ISS) (Ott et al. 2004)

Fig. 6.3 Dust mites, free condensate (Mir) (Ott et al. 2004)

water, and *Staphylococcus* sp., *Aspergillus* sp., and *Cladosporium* sp. on surfaces were dominating, respectively (Novikova et al. 2006). Examinations with cultivation-independent verification procedures revealed many gram-positive and gram-negative microorganisms, *Actinomyces* and fungi (Ullrich and Paulsen 2011). Even within the drinking water, pathogenic microorganisms were found. The identified spectrum of bacterial and fungal species of the ISS was very similar to the spectrum on board the Mir (Table 6.1). Additionally, the main species of bacteria and fungi as found on 15-year-old Mir are the same as those on board of the space shuttle (Pierson 2001). It must be assumed that each and every spacecraft or station is microbially contaminated with comparable spectra as soon as it was in contact with human beings. This leads to the conclusion that the primary source of the contamination is neither the spacecraft nor the food or water brought along but mainly the endogen flora of the astronauts (Ullrich and Paulsen 2011) (Figs. 6.2, 6.3, 6.4, and 6.5).

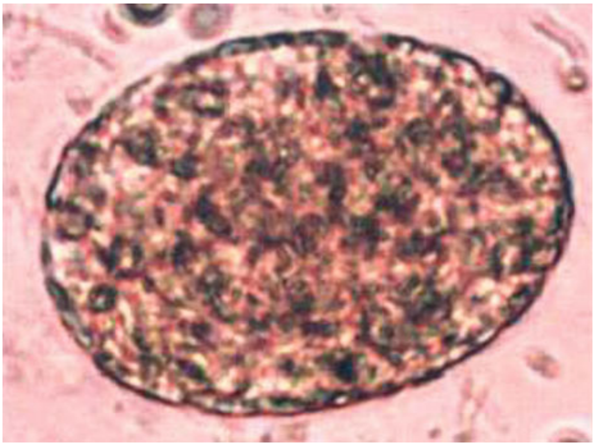

Fig. 6.4 Amoeba, free condensate (Mir) (Ott et al. 2004)

Fig. 6.5 Ciliated protozoa, free condensate (Mir) (Ott et al. 2004)

6.3 Alterations Within the Microflora of Crew Members

Investigations on crew members of the Apollo and Skylab missions and also on Soviet cosmonauts revealed that there were fundamental changes in the intestinal, oral, and nasal microflora under spaceflight conditions. Within the nasal flora, a decrease of apathogen and an increase of pathogen bacteria were found (Nefedov et al. 1971). The causation of this change can be found on the one hand by the crew members themselves, because under the condition of the isolation on board, an obviously considerable mutual exchange of microorganisms occurs between them. This exchange is not only affecting the upper respiratory tract but also intestinal

Table 6.2 Effects of space flight conditions on microorganisms

Effect	Spacecraft/Mission
Increased growth rate of *Chlamydomonas monoica* (Van den Ende and Van den Briel 1997)	Foton 1
Shortened lag phase of *Escherichia. coli* (Bouloc and D'Ari 1991)	STS-65, IML-2
Increased virulence of *Salmonella typhimurium* (Wilson et al. 2007)	STS-115
Faster growth rate, increased virulence, and raised resistance of *Salmonella typhimurium* (Nickerson et al. 2000)	Simulated microgravity
Increased virulence and resistance against antibiotics, tetracycline resistance of coliform bacteria (Klaus and Howard 2006)	Several
Development of resistant *E. coli* (Tixador et al. 1992)	Salyut 7
Faster growth and raised resistance against antibiotics of *E. coli* (Tixador et al. 1985)	STS-61-A, Spacelab D1
Severe increase of denseness of the cell wall of *Staphylococcus aureus* (Lapchine et al. 1986)	STS-61-A, Spacelab D1
Significantly increased mutation rate of bacterial ribosomal genes (Fukuda et al. 2000)	Mir
Enhanced acid tolerance ability, modified biofilm architecture and extracellular polysaccharide distribution of *Streptococcus mutans*, increase of proportion of *S. mutans* within dual-species biofilm (Cheng et al. 2014)	Simulated microgravity
Adoption to anaerobic mode of growth with denitrification of *Pseudomonas aeruginosa* (Crabbe et al. 2011)	ISS

STS Space Transport System, *ISS* International Space Station

bacteria (Taylor and Sommer 2005). The intestinal flora changes a lot, and after 2 weeks of space flight, the amount of detectable bacteria from the gastrointestinal tract is decreasing significantly (Taylor and Sommer 2005). The food on board could be a further reason, because the continuous consumption of sterilized, dehydrated nourishment leads to a rapid decrease in the amount of bifidobacteria and lactobacilli and is therefore promoting the expansion of resistance against antimicrobial drugs and the infection with opportunistic agents (Taylor and Sommer 2005). The construction of spacecraft and components of space stations under clean room conditions is followed by a settlement of the flora brought in by the crew. This is proven by the fact that microorganisms in the air and on surfaces are derived from the crew members (Makimura et al. 2011).

6.4 Raised Resistance Under Space Flight Conditions

Space flight conditions seam to alter the properties of many microorganisms (Table 6.2): on board of spacecrafts, an enhancement of the microbial proliferation, an altered microbial flora, an increased virulence, and a decreased effectiveness of antimicrobial drugs can be reported (Juergensmeyer et al. 1999; Leys et al. 2004). The alteration of susceptibility or resistance to antibiotics is very different, and the resistance effect is quickly lost upon return to earth. Each bacterial species responds differently to the suite of antibiotics, frequently becoming less resistant but occasionally even more resistant to antibiotics (Juergensmeyer et al. 1999). Bacteria seem better to be able to protruse stressors like changes in osmolarity, pH, temperature, and antimicrobial substances in absence of gravity (Rosenzweig et al. 2010). In weightlessness, a thickening of the cell wall of bacteria could be observed, which showed reversible after returning to terrestrial environment. The decreased stress on surfaces of microorganisms in microgravity can directly alter gene expression and affect physiological functions. In *Salmonella typhimurium*, for example, mechanisms associated with microgravity are mediated by the RNA chaperone. Hfq is a global transcriptional regulator, which plays an important role in the translation in answer to “envelope stress” and environmental stress (Wilson et al. 2007; Crabbe et al. 2011) (see also chapter 1). This chaperone is evolutionary highly conserved and could absolutely be one of the basic principles of the molecular mediation of changes in gravity on cells. Hfq even represents the first spaceflight-induced regulator acting across bacterial species (Crabbe et al. 2011). In addition to the influence of gravity acting on microorganisms on board of a spacecraft, also high doses of cosmic rays do cause an increase in mutation frequency (Horneck et al. 2010). In general it can be said that space conditions may significantly increase the mutation frequency of certain genes in microorganisms (Su et al. 2013). Spaceflight conditions therefore lead most likely to increased proliferation and selection of bacteria that are better adapted to microgravity and to the special environment of a spacecraft or space station (Juergensmeyer et al. 1999; Leys et al. 2004). In addition to these processes of adaptation, the bacterial phenotype trained in weightlessness seems to be particularly resistant to environmental influences. Unlike human cells, such as cells of the immune system (Ullrich and Thiel 2012), bacteria seem to be well prepared for a life under space conditions.

6.5 Material Damage Due to Microbial Contamination

Not only human health is affected by the microbial flora on board, but also the spacecraft, equipment, and different materials can be colonized or even degraded or inhibited in function by fungi or bacterial biofilms. An overview is summarized in Table 6.1, contaminations and control mechanisms in the chapter about contamination monitoring and control. Among the proven microorganisms on the Mir were many species with biodestructive properties that significantly damaged the cabin interior, the plastic seals, cables, and lighting (Novikova 2004; Van Houdt et al. 2012; Viktorov et al. 1992). For example, an expansion of *Penicillium chrysogenum* was observed, a material degrading, and biodestructive fungus (Viktorov et al. 1998). Thin biofilms, which are able to degrade many materials occurring on the ISS, are mostly formed at interfaces (Gu et al. 1998). Bacteria organized in biofilms show a very solid resistance against antibiotics (Mah and O’Toole 2001).

6.6 Conclusion

The ultimate target and attraction to explore the universe remain in human beings to discover and experience space, despite the benefits of using robots. The spaceship or space station that will be the home for a quite long time for future astronauts and the understanding of its microbial environment play a crucial role in making any space intention a success (Nicogossian and Gaiser 1992).

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