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Influence of Microgravity on the Physiology, Pathogenicity and Antibiotic Efficacy of Microorganisms

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ABSTRACT

Exploration beyond low earth orbit is a major challenge during space missions. The journey brings deleterious changes to the composition of bacterial flora of the spacecraft and compromises the immune system of the crew members significantly. Space exploration reduces immunological competence in crew members and is expected to create harmful alterations in the bacterial flora of the nasal, gastrointestinal, and respiratory tracts, increasing the susceptibility to disease. The pathogenicity character traits of bacteria and other microorganisms that pollute the material of the International Space Station as well as other flight platforms may be modified by the space flight environment,which may affect their vulnerability to antibiotics, which are important ingredients of flights medical setups. In conclusion susceptibility of microbes to antibiotics was affected and measures need to be studied in order to establish precautionary methods for future space missions. In this review we discussed the effect of spaceflights on microbial physiology; various challenges faced by the crew members and spacecraft equipment, and also highlighted methods toovercome these challenges during space flights for ensuring safety of the crew.

Keywords- Microgravity, Microbiome, Secondary metabolism, Pathogenicity, Antibiotic efficacy.

I. INTRODUCTION

Space is a difficult and expensive atmosphere to operate in. It has always been the domain of governments, many of whom are currently facing budget cuts and severe doubts about the worth of their space initiatives.

The most significant thing that might help us accelerate our development toward realizing this capability is funding. Not just total research funding, but funding to ensure the availability of all aspects that influence the success of the research. From frequency, dependability and availability to microgravity research platforms, coherence of funding is needed to promote robust universal programming that will attract top scientific talent. The desire of governments, institutions, and private companies to invest will be determined by a number of factors which will change with time and hence, we need to adapt with those changes as well.

For the future of humanity in space, we have reached a tipping point. Many private companies are successfully pioneering transport services to and from space, and the potential applications of space are expanding further than communication systems, imagery, and weather inference to include research, advancement, maintenance, and discovery on space-based platforms where humans can reside and operate (DiFrancesco & Olson, 2015).With the advent of frequent space mission all over the world, understanding the role of Microorganisms under the effect of microgravity and radiation is now possible at a much faster scale than it was in the 90's or early 20's. The low gravity and highly radiated environment provides a special ecological niche to undertake scientific studies that decode how vigorous space flights affect living beings in non-Earth environments.

Extensive investigations have been performed on living organisms in several space missions worldwide (Horneck et al., 2010; Koenig & Pierson, 1997; Milojevic & Weckwerth, 2020). However, micro-

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organisms come out to be the most desired subjects because of their short life span and ease of utility. Furthermore, with an aim to enhance certain microbial activities as well as increase metabolite production for economical advantages, space exploration studies offer a new terrain that has immense potential in the future.

Studies have shown that there has been an increase in pathogenicity, growth rate, and secondary metabolite production of microorganisms under the influence microgravity (Horneck et al., 2010; Taylor, 2015).

While the idea to study the micro-organisms under the influence of microgravity seems a very alluring, due to several instrumental and logistical hurdles, it wasn't till 1960's that several unresolved questions pertaining to effect of microgravity and microbial deportment could be resolved (Nickerson et al., 2000). Lack of instrumentation and technology made it difficult to get an in-depth analysis of the effects of microgravity on the genetics and phenotypic responses of microbes. In terms of their responses to microgravity, only a few microbes had been investigated, including *streptomycetes* till 20's (Senatore et al., 2018). With the advent of space flight exploration for commercial purposes, this area is now open for a comprehensive and deep investigation which will expand our knowledge pertaining to physiology and metabolism of living organisms.

In this review, we explore the effects of microgravity on microbial physiology (like effects on pathogenicity, and growth rates, etc.), production of secondary metabolism and antibiotic susceptibility (Huang et al., 2018) and how microbial contamination of spacecraft affects the life of crew members and causes threats to their survival by contaminating their food resources and damaging the essential equipments of spacecraft. We will also discuss the methods whichwere employed to deal with problems such as microbial contamination, increased pathogenicity, biofilms formation, etc. With deep space explorations as well as landings on several other planets, it is safe to presume that scientific discoveries in the field of space microbiology will only see a rise and also augment human capabilities in the annals of time.

Effects of simulated microgravity and spaceflight on microbial secondary metabolism

In ground-based and spaceflight microgravity analogue tests, microgravity has been proven to alter metabolism. Although microgravity and its counterparts have been investigated in relation to microorganisms, there have been numerous reports of contradictory and inconsistent results, particularly in the areas of secondary metabolism. In general, environmental cues and pressures outside of the cell, such as nutrients, osmotic stress, heat and shear stress, affect the biosynthesis and yield of microbial secondary metabolites (Berdy, 2005; Viollier et al., 2003). As a result, it's crucial to look at the yield of secondary metabolites produced in microgravity cells. Conditions have an effect on the rate of microbial growth in the same way that they have an effect on the rate of microbial growth, a variety of findings involving secondary metabolite yields have been published frequently (Demain & Fang, 2001; Gao et al., 2011). Scientists considering using simulated microgravity on the land to study the effects of the weightless environment in space on secondary metabolites utilised in human and veterinary medicine should look into these effects. (i.e. immunosuppressive drugs, antibiotics and anti-tumor agents).

During the US Space Shuttle mission STS-80, *Streptomyces plicatus* WC56452 enhanced its basic productivity (pg/CFU) of actinomycin D. (Lam et al., 2002). The creation of monorden by *Humicolafuscoatra* WC5157 cultivated on two kinds of agar plates (T8 and PG) were boosted throughout spaceflight onboard the US Space Shuttle Mission STS-77, according to another study (Lam et al., 2002) (Lam and colleagues, 1998). Luo et al., 1998 found that during 15 days of spaceflight aboard a spacecraft, the productivity of nikkomycins by *Streptomyces ansochromogenus* increased by 13–18%. Xiao et al., 2010 found that SMG, or simulated microgravity, was used as a new environmental indication, increased the growth rate of the toxin microcystin (MC) by the Cyanobacterium *Microcystis aeruginosa* PCC7806, while inhibiting its growth. Liu et al., 2011 found that under a high magnetic field and in a changed gravity situation approximated by diamagnetic levitation, in a solid culture, *Streptomyces avermitil*is increased the progression of the major anthelmintic agent avermectin.

Fang et al. 2001 (Demain & Fang, 2001) found that low shear modeled microgravity (LSMMG) prevented *S. clavuligerus* ATCC 27064 and *S. hygroscopicus* ATCC 29253 from developing the beta-lactam antibiotic cephalosporin and the polyketide macrolide rapamycin, respectively. Growth under LSMMGcircumstances favoured rapamycin extracellular proliferation, according to further studies. Moreover, the site of rapamycin accumulation was transferred to an extracellular position to a modest degree, while overall rapamycin yields were reduced. LSMMG inhibited the multiplication of the peptide antibiotic microcin B17 by *E. coli* ZK650 in HARVs, according to Fang et al (Koenig & Pierson, 1997), If *E. coli* was grown in shaking flasks or RWBs, it was discovered that the place of microcin aggregation differed significantly. The majority of the microcin in bacteria grown in flasks was located in the extracellular medium, whereas the extracellular medium contained the most of the microcin in HARVs. It's worth noting that the switch in microcin localization from intracellular to extracellular was most likely caused by the bioreactors' reduced shear stress, as a single glass bead added to the RWB medium was enough shear to transfer microcin aggregation from the media to the cells (Huang et al., 2018).

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Effect of space flight on the physiology of the bacteria:

The analysis of bacterial activity in space is critical for early detection of changes in bacterial populations and bacteria with medical, environmental, or life support implications for the crew's sustainability in closed space environments. To protect astronauts' health, several experiments were conducted to determine physiological changes in possible pathogens during space missions. The Cytos 2 experiment was carried out during the French– Soviet space mission in July 1982, in which the minimum inhibitory concentration of various antibiotics in *Staphylococcus aureus* was determined *E.coli, S.aureus* (Tixador et al., 1985). The results showed that both microorganisms were more resistant to all antibiotics analysed. To corroborate this impact, similar investigations were performed in future flight experiments, and they consistently demonstrated increasing resistance to antibiotics in these bacteria (Thévenet et al., 1996). Bacteria with biocorossive and biodegradative properties can jeopardise the credibility of the spatial hardware and, by causing infections, may have a negative impact on the crew's health (Senatore et al., 2018).

Microgravity affects bacteria only indirectly (Benoit & Klaus, 2007; Klaus et al., 1997), due to the quiescent fluid state circling the cells in liquid suspension culture. In microgravity, cell settling by liquid media and the capacity forbuoyant convection of less dense fluid in the vicinity of suspended bacteria are greatly reduced, and diffusion becomes the primary mode of nutrient and metabolic waste transfer toward and away from the cell. The hypothesis that microgravity-induced variations in liquid-culture growth kinetics are caused by fluid dynamics and extracellular transport phenomena rather than cellular dynamics is confirmed by findings that bacteria such as *E.coli* and *Bacillus subtilis* cultured on solid medium across flight grow at the very same rate and to the same degree as terrestrial control (Kacena et al., 1997; Pollard, 1965). The effect of space flight on growth kinetics and bacterial motility has been found to have a strong connection, which helps to explain differences within flown experiments in this field. Thus, variations among microgravity-induced growth effects and ground controls seem to be most noticeable when the bacteria under study are flagellate: obviously, motile cells have the ability to search out microclimates in liquid cultures that have not been drained of nutrients, and flagellar activity will mix the quiescent layer across the cell. Although no conclusive experiments to support this claim have been conducted, studies with microgravity analogues such as clinostats and the high aspect ratio vessel (HARV), a rotating wall bioreactor, support the idea that mixing microgravity-grown cultures to remove differences in fluid dynamics abrogates these growth kinetic effects (Taylor, 2015).

Effect of Microgravity on the pathogenicity of bacteria:

Pathogenicity is defined as the degree of virulence of a pathogen i.e. bacteria and this is adjudicated by its potential to invade and multiply within the host. Immune system plays a crucial role in protecting us from these pathogens by forming Lymphocytes i.e. white blood cells that are further of two types, the T- lymphocytes which help destroying cancerous or infected cells and the B- lymphocytes which forms antibodies that wipe out toxins and bacteria (Crucian et al., 2018). Previous studies have proved that space flight environment weakens the immune system of human and animals to great extent. Therefore, protection of humans and animals against various infectious diseases is a big challenge during extended space flight. Also it has been noticed that bacteria are acclimatized into new environment very quickly, some pathogenic bacteria shows an increase in their pathogenicity and anti- microbial resistance in microgravity and it has also been proved that bacteria in microgravity showed an increase in production rate (i.e. increase in cell population, etc) due to decrease in various stress factors (Huang et al., 2018). The detection of biofilms in the water system as well as other surfaces has been observed as a result of spacecraft pollution (Novikova, 2004; Ott et al., 2004; Schultz et al., 1989; Song & Leff, 2005). Some research has focused on this microbial element, which may play a role in enhanced pathogenesis and is influenced by space conditions. *Salmonella typhimurium* cultured in space revealed considerable variability in cell aggregation and thickness, according to Wilson et al. (Wilson et al., 2007), which they linked to extracellular matrix aggregation, believed to be an indicative of biofilm development (Nickerson et al., 2001).

Further research has shown that cultivating *S.typhimurium* under LSMMG increased the pathogen's virulence (Nickerson et al., 2000). As compared to species grown under normal gravity, S.*typhimurium* grown under LSMMG hada lower LD50, a shorter host time-to-death, and increased colonization of the murine liver and spleen following oral infection. This research was the first to show that LSMMG plays a role in microbial virulence, and it also shows that LSMMG has a global effect on bacterial physiology. As a result, LSMMG can be implemented in terms of ecological indicators known to modulate the expression of virulence determinants in *Salmonella*, which already includes osmolarity, malnutrition, oxidative stress, pH, and growth process (Foster & Spector, 1995; Mahan et al., 1996)

Kim et al., 2013 on the other hand, found no cell clustering in *Penicillium aeruginosa* cultured in space, which they speculated was due to variations in the strain's extracellular polymeric substances (EPS). *P.aeruginosa* PA14 cultured during spaceflight, unlike *P.aeruginosa* PAO1 cultured in simulated microgravity, in planktonic cultures, no cellular aggregates were

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formed. Variations in the EPS that produce cellular clustering are most likely to blame for this finding. (Kim et al., 2013). In addition, the difference in finding between spaceflight and simulated microgravity settings could be explained by the fact that the spaceflight mechanism has significantly less shear than LSMMG in modeled microgravity (Crabbe et al., 2011).

P.aeruginosa was cultivated throughout two Space Shuttle missions (STS-132 and STS-135) by (Kim et al., 2013), and the biofilms developed during spaceflight were characterized. The biofilms had an unusual structure consisting of columns overlaid with a canopy that had never been seen on earth before.

Effect of spaceflight on fungal physiology

From HEPA filters in the US Laboratory module of the International Space Station, possible toxin or allergen producers like *Penicillium chrysogenum* and *Penicillum brevicompactum*, as well as potential opportunistic pathogens like *Aspergillus niger* and *Aspergillus flavus*, have been isolated Inflammatory and cytotoxic responses can be triggered by these toxin producers (Vesper et al., 2008). Biodegradation and a direct hazard to crew health are also possible outcomes of colonised microbial consortia (Leys et al., 2009). The capacity of astronauts' neutrophils and monocytes to phagocytize after a mission has been confirmed to be reduced (Kaur et al., 2004, 2005). As a result of this situation, these opportunistic pathogens could become a significant threat to astronauts' health. Furthermore, fungal mould growth may affect the organic substrates of space travel, causing damage and affecting its activity. Thus, phenotypic characterization of these filamentous fungi in microgravity must take precedence (Sathishkumar et al., 2014).

Scanning electron microscopy is regarded as a promising advanced method for observing the development of fungal hyphae, which can be influenced by growth conditions, as well as a possible indicator for monitoring the impact of microgravity on fungal physiology. The morphology of fungal hyphae can be used as a possible selective predictor to investigate the effects of microgravity on fungal physiology. When comparing the *A. niger* mycelium grown LSMMG condition to the regular and static conditions, homogeneous hyphae with smooth cell walls were observed. Although the effects of microgravity on *P. chrysogenum* mycelium were similar to those of *A. niger*, the findings for *P. chrysogenum* mycelium were not. Some studies looked at the impact of laboratory-scale LSMMG on microbial growth over a short period of time. A study (Purevdorj-Gage et al., 2006) found that yeast cells *S. cerevisiae* (1,400 min old culture) grown under LSMMG conditions grew at the same rate, scale, shape, and viability as controls. Overall, SEM findings show that the LSMMG did not stress *A. niger* and *P. chrysogenum* mycelia.

Internal subcellular organelles, vacuolation sequence, irregular polysaccharide distribution, autolysis and degradation of cytoplasmic contents can all be studied using transmission electron microscopy (Ahmad & Khan, 2012). It has been found that, except for the increased distribution of extracellular melanosome-like structure along the cell wall, intracellular observations *of A. niger* hyphae grown under LSMMG revealed no differences in subcellular distribution relative to normal and static conditions. Furthermore, no woronin bodies were found near the septa, indicating that *A. niger* was not stressed by microgravity. In comparison to microgravity and static conditions, *A. niger* grown in normal gravity showed normal cytoplasm granulation and a small increase in the number and size of vacuoles, as well as electron dense gamma particles in the cytoplasm.

Let's take on *P. chrysogenum*, the increased number of mitochondria with membranous cristae was observed all along the cytoplasm and cell wall *of P. chrysogenum* seemed to be stable, as well as the rate of spore germination was enhanced under microgravity conditions, according to a previous study (Sathishkumar et al., 2014). *P. chrysogenum*, a filamentous fungus can be chosen as a strong candidate for producing silver nanoparticles. *P. chrysogenum* has evolved various unique adaptations to live in such microgravity environments, including improvements in secreting metabolites, membrane function, and enzyme function, as compared to normal gravity (1 g control). The colour shift in the medium containing 1 mM silver nitrate could be used to visually infer the synthesis of silver nanoparticles. The medium changed colour from colourless to dark brown, suggesting a reduction in silver ions. In microgravity, colour shift was noticeably quicker and more rapid than in normal gravity. For AgNPs synthesised in microgravity (MG-NPs) and AgNPs synthesised in normal gravity (NG-NPs), the maximum colour change was observed after 8 and 24 hours, respectively. With the exception of the rapid reduction of silver ions by culture filtrate of microgravity-grown biomass, it is worth noting that; the colour was also more intense, suggesting a high yield of silver nanoparticles. Hence it has been concluded that there is a positive impact of the microgravity on the synthesis of silver nanoparticles and this application of *P. chrysogenum* in microgravity can be used for the production of silver nanoparticles efficiently, as these particles have antibacterial and cytotoxicity activity (Sheet et al., 2017).

Effect of space flight on fungal virulence:

With the advancement of space technology in recent years, microbial space protection has become a research hotspot. Microorganisms such as fungi have been found in abundance on the International Space Station (Novikova et al., 2006). Let's take a look on the *C. albicans; C. albicans* is a widespread contingent pathogen that feeds on the skin, mouth, urinary tract, and reproductive system of humans (McCullough et al., 1996). *C. albicans*

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pathogenicity can change in response to changes in the external environment (Mayer et al., 2013). These microorganisms have been found to proliferate more rapidly in the International Space Station (Rosenzweig et al., 2010) (Sugita et al., 2016), increasing the risk of onboard cross-contamination, colonisation, and infection.

It has been proved by an experiment where *C. albicans* was recovered in the sabouraud-dextrose broth (SDB) medium after exposure to the spaceflight environment, and their survival was measured using the OD600 method. The same tests were performed on non-exposed control *C. albicans* that were grown on the ground. The spaceflight group had an 8-hour growth lag after being inoculated into medium, which was shorter than the control group's (10- hour lag). In the sabouraud-dextrose broth (SDB) medium containing hydrogen peroxide, the survival rate of the spaceflight group was higher than that of the control group, according to the environmental resistance assessment results. However, there was no discernible difference in acid, alkali, alcohol, or salt tolerance between the two groups of *C. albicans*. The results of the virulence experiment revealed that the spaceflight group had a lower threshold and a more substantial decrease in the normalised cell index (NCI) than the control group, indicating that the spaceflight group is more cytotoxic. Overall, exposure to the spaceflight environment increased *C. albicans* proliferation, biofilm formation, antioxidant ability, and cytotoxicity.

Furthermore, the astronaut's immunological investigations revealed many dysregulations, including lymphocyte proliferation, cytokine synthesis (Sonnenfeld Gerald et al., 2003), and leukocyte subset redistribution (Crucian et al., 2015). *C. albicans* development in simulated microgravity results in an increase in filamentous types of the organism, which is consistent with increased pathogenicity (Altenburg et al., 2008). According to those sources, the presence of this microbe in the space environment could endanger astronauts' health. And it has been discovered that the genes involved in oxidative stress tolerance were up-regulated in the spaceflight (Crabbé et al., 2013).

Effect of space flight on fungal secondary metabolite production

Secondary metabolite production is one of the most important properties of microorganisms like fungi. It has been stated that fungi's secondary metabolites, such as fungal poisons and material corrosion, could pose a severe threat to the ISS's long-term function. As a result, the astronauts' lives could be jeopardized while they deal on them. So, we need to look into the potential behaviors of fungi in the production of secondary metabolites in order to find a solution to these issues, as they may be life-threatening to astronauts in space. However, microbial development of antibiotics through secondary metabolism is one of the main areas of applied microbiology; secondary metabolites can be used to make antibiotics and other useful products that can be used to combat a range of pathogens that can pose a serious threat to crew members. Since secondary metabolite development varies depending on the climate, it's possible that microgravity may be an appropriate environment for the production of these useful products. So, in order to investigate these various fungi behaviors in the development of secondary metabolites, we must examine the various species in greater detail, since it is possible that different species may act differently in microgravity and produce a different variety of metabolites with greater efficiency than on the earth, which could be extremely useful and informative to mankind.

Spaceflight and simulated microgravity have been shown to elicit morphological alterations in *Ulocladium chartarum*, *A. carbonarius*,*P. chrysogenum*, and *A. niger* mycelia and colonies (Gomoiu et al., 2013). Let's take a look on *A. carbonarius*, in comparison to those in the control condition; MG resulted in a greater colony centre of *A. carbonarius*. As detected by high-performance liquid chromatography, the accumulation of ochre-toxin A (OTA), oxalic acid, and citric acid was significantly increased in the MG treatment group compared to the control group (HPLC). By using high-resolution gaschromatography (GC)-time of flight (TOF) mass spectrometry, the internal and extracellular metabolites of *A. carbonarius* were identified individually (MS). In the cells and the potato dextrose agar (PDA) media, there were 858 and 613 peaks, respectively. Amino acids, organic acids, carbohydrates, fatty acids, and lipids were shown to be involved in a variety of metabolic reactions in *A. carbonarius*. The three metabolites with the consequent amount in expression in the MG group compared to the control group were D-altrose (1,286,796-fold), isocitric acid (5.77-fold), and 3-hydroxyflavone (5.23-fold). After clinostat rotation, the expression of certain metabolites that have protective and antioxidant effects on fungal cells, such as trehalose and lipoic acid, rose dramatically. It should be noted, however, that D-altrose is extremely rare in nature and has only been discovered in bacteria. Lactamide, Maltitol, cellobiotol, dehydroepiandrosterone, tagatose and saccharic acid are also described as fungal metabolites on a rare occasion. As a result, additional research is required. Using the respective pure chemicals, validate the presence of these products (Chunmei et al., 2021).

Let's take a look on *Ulocladium chartarum*, the existence of long submerged, completely alive mycelium and the formation of microcolonies at the edge of the culture plates in the flight samples are the most noticeable differences between flight and ground samples. Healthy hyphae were shown to develop and branch in both horizontal and vertical directions, orienting themselves towards the substrate's depth (Gomoiu et al., 2013). The very long, nearly unbranched hyphae developed quickly away from the main

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colony, towards the border of the culture plates. It is well known that in nutrient-rich places, mycelium branches and grows slowly, increasing the amount of nutrients it can absorb; in nutrient-poor places, hyphae grow significantly and with minimal branching. Factors such as hazardous chemical biosynthesis, CO2 accumulation, and the virtual absence of air convection in space could encourage the growth of submerged mycelium with extreme apical dominance and, eventually, the formation of microcolonies due to heavy random lateral branching, as a strategy to ensure colony survival and locate less harsh growing conditions. In all experimental settings, exudates or liquid droplets were observed on the surface of colonies, but they were larger and more abundant in flight than on the ground. Exudates are connected with actively expanding mycelia, according to Colotelo, 1978, and the process of exudation is physiologically important and intimately related with colony ageing, according McPhee & Colotelo, 1977. *Ulocladium chartarum*, as well as other fungal species such as *Sclerotinia sclerotiorum* (Liang et al., 2010), exude liquid droplets often. Proteins found in exudates were divided into numerous functional categories, including secondary metabolism, according to a proteome-level analysis. Botralin (*Ulocladium botrytis*), curvularins *Ulocladiumatrum*), ulocladol A and ulocladol B (*Ulocladiumchartarum*) (Andersen & Hollensted, 2008; Höller et al., 1999; Sviridov et al., 1991) are examples of secondary metabolites isolated from *Ulocladium* species. Space circumstances and the biocontainer's isolated environment may cause an increase in secondary metabolism and metabolite excretion, as well as faster buildup and higher concentrations near the colony, all of which operate as stressors/toxins.

Effect of space flight on the virulence of the virus:

The analysis of the influence of microgravity on virus pathogenicity is critical because astronauts interact directly with the viruses on the space station and must understand how they behaves in that environment because some viruses can increase their pathogenicity in order to properly manage conditions inmicrogravity**.**

NASA has investigated that any dormant viruses have the potential to reactivate when exposed to intense environments such as microgravity. NASA is continuing to research how the human immune system responds in space in order to prepare for future human spaceflight missions to the Moon and Mars. Certain viruses, such as the *Varicella Zoster Virus* (VZV), which causes chickenpox in children and shingles in adults, do not completely clear the body.The immune system keeps them in check and keeps the virus inactive in the body's spinal cord nerve cells. These viruses can “awaken,” or reactivate, at any time, and begin to develop and multiply. The person usually has no symptoms or signs of illness when this happens; however, it is more common among older people and people with weakened immune systems for the reactivation to cause disease. The reactivation of the virus is thought to be caused by stressful life conditions (Rooney et al., 2019).

Previously, astronauts on short-duration (10–16 day) space shuttle flights showed increased reactivation of various naturally occurring latent herpes viruses such as *Epstein–Barr virus* (EBV), *Varicella Zoster Virus* (VZV) and *cytomegalovirus* (CMV). 1. However, viruses were lost in body fluids during reactivation, and the astronauts were normally asymptomatic. 2. Stress responses linked with spaceflight involve stimulation of the hypothalamic- pituitary-adrenal and sympathetic-adrenal-medullary axis 3, which can lead to the reactivation of latent herpes viruses, putting astronauts at risk of transmitting live, infectious viruses while in space. (4and5). Cortisol and dehydoepiandosterone (DHEA) may influence cellular immune regulation, causing dormant viruses to reactivate. 4. Herpes virus reactivation can cause silent, debilitating, or perhaps even life-threatening reactions. Isolating crew members before a flight has little effect on the reactivation of dormant viruses.Even a thorough quarantine isn't enough to keep viruses from reactivating in space. 5. NASA has investigated that any dormant viruses have the potential to reactivate when exposed to intense environments such as microgravity. NASAis continuing to research how the human immune system responds in space in order to prepare for future human spaceflight missions to the Moon and Mars.

Most astronauts who reactivated one or more of the target herpes viruses experienced significant changes in cell-mediated immunity. Glaser previously discovered a link between EBV reactivation and weakened cell-mediated immunity (Mehta et al., 2017).

II. TOOLS AND TECHNIQUES

Nanopore sequencers:

Portable technologies are required for remote molecular diagnostics on Earth and in space (McIntyre et al., 2016). Microbes demonstrate higher virulence in microgravity (Altenburg et al., 2008; Klaus & Howard, 2006; Wilson et al., 2007) while humans show immunological deregulations (Mermel, 2013; Sonnenfeld & Shearer, 2002). This is a dangerous combination onboard tight vessel, where medical specialists are scarce and supplies are limited. Sequencing technologies could be crucial for quick reactions to medical illnesses in space, such as determining whether or not to use antibiotics and, if so, which ones to use. Single-molecule approaches can also detect changed nucleic acids, which could be useful for crew health monitoring (Li & Mason, 2014; Rand et al., 2016; Saletore et al., 2012; Simpson et al., 2016). As generic current sensing devices, nanopore sequencers could help in the hunt for alien intelligence by expanding the range of detectable polymers beyond the conventional nucleobases of DNA and RNA (Garalde et al., 2018; Rezzonico, 2014).

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Oxford Nanopore Technologies' (Jain et al., 2016) commercialised nanopore sequencing is a promising approach that is now widely used in laboratories as well as in the field. McIntyre et al. showed a solitary mapped read obtained by nanopore sequencing throughout parabolic flight spanning numerous parabolas (Carr et al., 2020). Flow cell vibrations advised that 70% of pores should oppose launch, which is compatible with subsequent successful nanopore sequencing on the International Space Station (ISS) (McIntyre et al., 2016).

The group evaluated the Oxford Nanopore Technologies MinION on a parabolic flight to understand the impact of varying gravity on the instrument and dataas a first step toward sequencing in space and aboard the International Space Station (ISS). The Oxford Nanopore Technologies (ONT) MinION sequencer is a compact sequencing device (4"1.5"1", 100 g) that gets power from and sends information to a server via a solitary USB 3.0 connection (Loman & Watson, 2015; Carr et al., 2020). Double-stranded DNA molecules with a hairpin adapter connecting the strands around one end as well as a motor protein linked at another end make up libraries. When the template strand, hairpin adapter, and complementary strand all pass through the pore in the same order, this arrangement allows both strands of the library templates to be sequenced. Base calls are more accurate when the consensus information from the "2D" reads is used instead of the template or complement strands alone.

At any given time, the nucleotides in the pore impede flow of current with a signal unique to their identity. At time points where raw electric current measurements change dramatically, which should reflect the entry of a single new nucleotide into the pore, these are referred to as "events." The current detection procedure, on the other hand, is noisy and depending on reaction circumstances such as temperature. Traditional alignment software failed to map most reads in previous versions of the pipeline from ONT, which used a hidden Markov model (HMM) algorithm with a Metrichor Viterbi decoder algorithm to call bases from event data (Jain et al., 2015; Mikheyev & Tin, 2014) for the newest version of the pore (‘R9'), Metrichor implements a recurrent neural network for improved base calling. Because of the high error rates (15 percent for earlier 2D reads13), producing and interpreting nanopore sequencing data remains a significant difficulty. Long reads of tens of thousands of bases or more, on the other hand, are frequently enough to allow taxonomic classification at the species or genus level (Juul et al., 2015).

Microscope for space studies:

Human health risks in space are well-known, but the underlying biological mechanisms are poorly understood and require further investigation. Microorganism survival research would improve space exploration in a variety of ways, including assisting in the creation of planetary preservation measures. The smallest microscopes examined were not stand-alone instruments, and the microscopes previously used in space were quite huge. There is a demand for more self-contained and tiny space microscopes. NIZEMI, a microscope- based biological space research mission that flew on the Spacelab mission in 1994, was the first of its kind. Its goal was to investigate how gravity affected small biological systems such miniature plants, fungus, spores, and cell cultures. It was made up of three parts: a control module, an experiment control unit, and an experiment module. Because the modules' aggregate mass was 98 kg, the system was relatively huge. A centrifuge was included in the experiment module, capable of accelerations ranging from 0.001g and 1.5g. The microscope was placed on top of the centrifuge and revolved alongside the sample. Bright field, dark field, and phase contrast imaging modes were all possibilities (Friedrich et al., 1996).

Microscopes, which are integrated parts of the facilities, are also available in the laboratory modules on the ISS. The Light Microscopy Module (LMM) is a multifunctional sub-rack of the Destiny module's Fluids Integrated Rack. Multiple imaging techniques, such as bright field, dark field, phase contrast, differential interference contrast, and confocal microscopy, are included inLMM, which was originally modified from a commercial light microscope. This microscope is controlled from ground (Lant & Resnick, 2000). Despite the fact that the LMM was created primarily for fluid physics studies, this has also beenemployed in biological research (Ferl & Paul, 2016).

Microscope for biological research in space

On the International Space Station (ISS), FLUMIAS-DEA, a firm high-resolution 3D fluorescence microscope, photographed two scientific data, one with fixed cells and the other with living human cells, in a temperature-controlled environment. The FLUMIAS-DEA microscope amalgamate the advantages of a high-resolution 3D fluorescence microscope relying on structured illumination microscope (SIM) technology accompanied by hardware designs to fulfill the needs of a space equipment. The FLUMIAS technology was successfully proved to be capable of acquiring, transmitting, and storing high-resolution 3D fluorescence images of fixed as well as living cells, allowing mathematical and dynamic analysis of subcellular structures including the cytoskeleton (Thiel et al., 2019). The ability of real-time analysis methods on the International Space Station will significantly increase our understanding of the kinetics of cellular reactions and modeling to the space environment, which is not only a choice, but a prerequisite of evidence-based medical risk evaluation, tracking, and mitigation advancement for exploration category missions.

Waste management in space:

The International Space Station, like any other home or business, gathers a lot of trash that is no longer

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needed by its inhabitants. Because there is no rubbish collection in space, the scientists on board rely on docked cargo ships, space shuttle missions (STS-105 and STS-108)

Wet trash returned from the STS-105 and STS-108 shuttle missions servicing the International Space Station was sampled and analyzed microbiologically (ISS). Plate waste and accompanying food packaging (which made up the majority of the garbage collected), sanitary waste, and free liquid inside the waste container were all used to gather samples. Microbial loads cultivated on selective and non-selective media, as well as total bacterial counts using acridine orange direct count (AODC) techniques, all revealed substantial microbial densities in the waste container liquid. Isolates like *Klebsiella pneumoniae, Bacillus spp., Escherichia coli*, etc were found. To evaluate the water and organic content of the materials, dry and ash weights were taken for each sample. The biostability of trash from shuttle flights will be evaluated in the future to establish a baseline measure of waste content, labile organics, and microbial load. The goal is to identify the waste stream's substance, as well as possible stabilization and recovery methods that could be used for long- term missions. There are space shuttles like STS-30, STS-29, and STS-35 that are used to remove rubbish from the International Space Station, and they had their trash evaluated at Johnson Space Centre (JSC), while some other shuttles like STS-99 and STS-101 had trash evaluations at Kennedy Space Centre (KSC). The goal of these studies was to determine the physical state of the various waste stream items recovered aboard the shuttle orbiter, with the goal of creating waste treatment methods for future lengthier spaceflights. Trash, solid food system waste, human solid waste, and experimental waste were the four categories of solid waste. Water contributed for nearly 30% (by mass) of the typical 9.89 kg per crew day of waste created on orbit for STS-30, STS-29, STS35, STS-101, and STS-99 waste assessments (Maxwell & Drysdale, 2001), while product packaging contributed for the bulk (80%) of the trash volume.

The orbiter's waste storage comprises of a volume F container for wet trash and a volume B compartment for dry rubbish. Discarded office supplies, plastic containers, and research wastes are examples of dry garbage. Mealtime wastes such as residual foods and beverages (plate waste) and the accompanying food packaging, personal hygiene goods, and toilet wipes are all collected in volume F. Because of its angular shape, the toilet wipes are discarded of next to the Waste Collection System (WCS) (toilet) in a plastic dumping bag known as a"elbow pack." Individual trash liners are used to keep food and personalhygiene items in places of high garbage creation within the crew compartment,such as the galley. When the liners are full, they are gathered and deposited in the proper storage container. Storage is sufficient for STS missions lasting less than 30 days because garbage spillage is stowed in the airlock.

Because the International Space Station (ISS) is a permanently operational on- orbit research facility, it has distinct waste disposal requirements than STS missions. Solid garbage is usually held on the International Space Station until it can be transported to Earth. The amount of rubbish that can be removed from the station this way is determined by the return method. The majority of the station's garbage (1600 kg) is evacuated by automated Russian Progress modules being sent to the station once every month. When the space shuttle undocks from the station on ISS servicing flights, it can also remove large amounts of waste (up to 9000 kg), along with an Italian-built Multi-Purpose Logistics Module (MPLM) payload for ferrying equipment and research to the station (Kish et al., 2002). STS missions to the International Space Station, on the other hand, are not spaced evenly with Progress launches. As a result, wastes are held aboard the space station for variable durations of time, which has ramifications for bacterial development and trash decomposition.

III. CONCLUSION

The review will encompass the importance of spaceflights and simulated microgravity on microbial physiology, virulence, and secondary metabolite production and also highlights various tools and techniques used for analysis of environmental variations on microorganisms.

As we travel into deep space, the risk of serious infection for spaceflight crew members will increase. Changes in human physiology and bacterial morphologies generated by the specific qualities of the space flight environment may hamper our ability to treat illnesses on these missions. Under the effect of microgravity, opportunistic diseases' resistance to conventional drugs may shift, and virulence-related properties of bacteria — fellow passengers on board the spacecraft – may change. The harsh climate, extremely radiated terrain and imbalanced atmosphere are set to pique the curiosity of scientists from all over the worlds. Differences in metabolite production and physiology of the micro-organisms have shown us that in terms of exploiting microbes for our benefit, we were limited by our current environment. Taking our research to the space has given us a chance to expand our knowledge and simultaneously maintain the desire to help mankind.

As a result, ground-based analogues will continue to be a promising technique of understanding how microorganisms' basic physiology adjusts to the space environment in the near future. However, all of the analogues will cause artefacts in simulation, which requires extra care. The essential difficulty is determining how to examine the data obtained in order to eliminate the artifact. Short-term space flight may enhance secondary metabolite production, but long-term

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space flight may impair secondary metabolism, according to preliminary results on the impact of space flight on secondary metabolism. Future research is required to clearly illustrate the mechanism.

Therefore, to conclude the subject, space exploration has opened a new leeway for scientists all over the world to take their research to a new level both in terms of quality and quantity. The plethora of options available when venturing in deep space establishes a completely new domain of research.

AUTHORSHIP

All authors have contributed to this manuscript.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

[1] Ahmad, I. and Khan, M.S.A. (2012). Microscopy in mycological research with especial reference to ultrastructures and biofilm studies. Current microscopy contributions to advances in science and technology. *Formatex Research Center, Spain*, 646-659.

[2] Altenburg, S.D., Nielsen-Preiss, S.M. and Hyman, L.E. (2008). Increased filamentous growth of Candida albicans in simulated microgravity. *Genomics, proteomics & bioinformatics*, 6(1), 42-50.

[3] Andersen, B. and Hollensted, M. (2008). Metabolite production by different Ulocladium species. *International journal of food microbiology*, 126(1-2), 172-179.

[4] Benoit, M.R. and Klaus, D.M. (2007). Microgravity, bacteria, and the influence of motility. *Advances in Space Research*, 39(7), 1225-1232.

[5] Berdy, J. (2005). Bioactive microbial metabolites. *The Journal of Antibiotics*, *58*(1), 1–26.

[6] Carr, C. E., Bryan, N. C., Saboda, K. N., Bhattaru, S. A., Ruvkun, G., & Zuber, M. T. (2020). Nanopore sequencing at Mars, Europa, and microgravity conditions. *Npj Microgravity*, *6*(1), 24. https://doi.org/10.1038/s41526-020-00113-9

[7] Chunmei, J., Dan, G., Zhenzhu, L., Shuzhen, L., Junling, S., Dongyan, S., & S., D. I. (2021). Clinostat Rotation Affects Metabolite Transportation and Increases Organic Acid Production by Aspergillus carbonarius, as Revealed by Differential Metabolomic Analysis. *Applied and Environmental Microbiology*, *85*(18), e01023-19. https://doi.org/10.1128/AEM.01023-19

[8] Colotelo, N. (1978). Fungal exudates. *Canadian Journal of Microbiology*, *24*(10), 1173–1181.

[9] Crabbé, A., Nielsen-Preiss, S. M., Woolley, C. M., Barrila, J., Buchanan, K., McCracken, J., Inglis, D. O., Searles, S. C., Nelman-Gonzalez, M. A., Ott, C. M., Wilson, J. W., Pierson, D. L., Stefanyshyn-Piper, H. M., Hyman, L. E., & Nickerson, C. A. (2013). Spaceflight Enhances Cell Aggregation and Random Budding in Candida albicans. *PLOS ONE*, *8*(12), e80677. https://doi.org/10.1371/journal.pone.0080677

[10] Crabbé, A., Schurr, M. J., Monsieurs, P., Morici, L., Schurr, J., Wilson, J. W., Ott, C. M., Tsaprailis, G., Pierson, D. L., & Stefanyshyn-Piper, H. (2011). Transcriptional and proteomic responses of Pseudomonas aeruginosa PAO1 to spaceflight conditions involve Hfq regulation and reveal a role for oxygen. *Applied and Environmental Microbiology*, *77*(4), 1221–1230.

[11] Crucian, B. E., Choukèr, A., Simpson, R. J., Mehta, S., Marshall, G., Smith, S. M., Zwart, S. R., Heer, M., Ponomarev, S., & Whitmire, A. (2018). Immune system dysregulation during spaceflight: potential countermeasures for deep space exploration missions. *Frontiers in Immunology*, *9*, 1437.

[12] Crucian, B., Stowe, R. P., Mehta, S., Quiriarte, H., Pierson, D., & Sams, C. (2015). Alterations in adaptive immunity persist during long-duration spaceflight. *Npj Microgravity*, *1*(1), 15013. https://doi.org/10.1038/npjmgrav.2015.13

[13] Demain, A. L., & Fang, A. (2001). Secondary metabolism in simulated microgravity. *The Chemical Record*, *1*(4), 333–346.

[14] DiFrancesco, J. M., & Olson, J. M. (2015). The economics of microgravity research. *Npj Microgravity*, *1*(1), 1–6.

[15] Ferl, R. J., & Paul, A.-L. (2016). The effect of spaceflight on the gravity-sensing auxin gradient of roots: GFP reporter gene microscopy on orbit. *Npj Microgravity*, *2*(1), 1–9.

[16] Foster, J. W., & Spector, M. P. (1995). How Salmonella survive against the odds. *Annual Review of Microbiology*, *49*(1), 145–174.

[17] Friedrich, U. L. D., Joop, O., Pütz, C., & Willich, G. (1996). *The slow rotating centrifuge microscope NIZEMI—a versatile instrument for terrestrial hypergravity and space microgravity research in biology and materials science*. Elsevier.

[18] Gao, H., Liu, Z., & Zhang, L. (2011). Secondary metabolism in simulated microgravity and space flight. *Protein & Cell*, *2*(11), 858–861.

[19] Garalde, D. R., Snell, E. A., Jachimowicz, D., Sipos, B., Lloyd, J. H., Bruce, M., Pantic, N., Admassu, T., James, P., & Warland, A. (2018). Highly parallel direct RNA sequencing on an array of nanopores. *Nature Methods*, *15*(3), 201–206.

[20] Gomoiu, I., Chatzitheodoridis, E., Vadrucci, S., & Walther, I. (2013). The Effect of Spaceflight on Growth of Ulocladium chartarum Colonies on the International Space Station. *PLoS ONE*, *8*(4). https://doi.org/10.1371/JOURNAL.PONE.0062130

[21] Höller, U., König, G. M., & Wright, A. D. (1999). A new tyrosine kinase inhibitor from a marine isolate of Ulocladium botrytis and new metabolites from the

ISSN: 2583-4053

Volume-1 Issue-5 || December 2022 || PP. 24-35

https://doi.org/10.55544/jrasb.1.5.3

marine fungi Asteromyces cruciatus and Varicosporina ramulosa. *European Journal of Organic Chemistry*, *1999*(11), 2949–2955.

[22] Horneck, G., Klaus, D. M., & Mancinelli, R. L. (2010). Space microbiology. *Microbiology and Molecular Biology Reviews*, *74*(1), 121–156.

[23] Huang, B., Li, D.-G., Huang, Y., & Liu, C.-T. (2018). Effects of spaceflight and simulated microgravity on microbial growth and secondary metabolism. *Military Medical Research*, *5*(1), 1–14.

[24] Jain, M., Fiddes, I. T., Miga, K. H., Olsen, H. E., Paten, B., & Akeson, M. (2015). Improved data analysis for the MinION nanopore sequencer. *Nature Methods*, *12*(4), 351–356.

[25] Jain, M., Olsen, H. E., Paten, B., & Akeson, M. (2016). The Oxford Nanopore MinION: delivery of nanopore sequencing to the genomics community. *Genome Biology*, *17*(1), 1–11.

[26] Juul, S., Izquierdo, F., Hurst, A., Dai, X., Wright, A., Kulesha, E., Pettett, R., & Turner, D. J. (2015). What's in my pot? Real-time species identification on the MinION™. *BioRxiv*, 30742.

[27] Kacena, M. A., Leonard, P. E., Todd, P., & Luttges, M. W. (1997). Low gravity and inertial effects on the growth of E. coli and B. subtilis in semi-solid media. *Aviation, Space, and Environmental Medicine*, *68*(12), 1104–1108.

[28] Kaur, I., Simons, E. R., Castro, V. A., Ott, C. M., & Pierson, D. L. (2004). Changes in neutrophil functions in astronauts. *Brain, Behavior, and Immunity*, *18*(5), 443–450.

[29] Kaur, I., Simons, E. R., Castro, V. A., Ott, C. M., & Pierson, D. L. (2005). Changes in monocyte functions of astronauts. *Brain, Behavior, and Immunity*, *19*(6), 547–554.

[30] Kim, W., Tengra, F. K., Shong, J., Marchand, N., Chan, H. K., Young, Z., Pangule, R. C., Parra, M., Dordick, J. S., & Plawsky, J. L. (2013). Effect of spaceflight on Pseudomonas aeruginosa final cell density is modulated by nutrient and oxygen availability. *BMC Microbiology*, *13*(1), 1–10.

[31] Kim, W., Tengra, F. K., Young, Z., Shong, J., Marchand, N., Chan, H. K., Pangule, R. C., Parra, M., Dordick, J. S., & Plawsky, J. L. (2013). Spaceflight promotes biofilm formation by Pseudomonas aeruginosa. *PloS One*, *8*(4), e62437.

[32] Kish, A. L., Hummerick, M., Roberts, M. S., Garland, J. L., Maxwell, S., & Mills, A. (2002). Biostability and microbiological analysis of Shuttle crew refuse. *SAE Technical Paper*, 1–2356.

[33] Klaus, D. M., & Howard, H. N. (2006). Antibiotic efficacy and microbial virulence during space flight. *Trends in Biotechnology*, *24*(3), 131–136.

[34] Klaus, D., Simske, S., Todd, P., & Stodieck, L. (1997). Investigation of space flight effects on Escherichia coli and a proposed model of underlying physical mechanisms. *Microbiology*, *143*(2), 449–455.

[35] Koenig, D. W., & Pierson, D. L. (1997). Microbiology of the space shuttle water system. *Water Science and Technology*, *35*(11–12), 59–64.

[36] Lam, K. S., Gustavson, D. R., Pirnik, D. L., Pack, E., Bulanhagui, C., Mamber, S. W., Forenza, S., Stodieck, L. S., & Klaus, D. M. (2002). The effect of space flight on the production of actinomycin D by Streptomyces plicatus. *Journal of Industrial Microbiology and Biotechnology*, *29*(6), 299–302.

[37] Lam, K. S., Mamber, S. W., Pack, E. J., Forenza, S., Fernandes, P. B., & Klaus, D. M. (1998). The effects of space flight on the production of monorden by Humicola fuscoatra WC5157 in solid-state fermentation. *Applied Microbiology and Biotechnology*, *49*(5), 579–583.

[38] Lant, C. T., & Resnick, A. (2000). Multi-function light microscopy module for the International Space Station. *AIP Conference Proceedings*, *504*(1), 324–329.

[39] Leys, N., Baatout, S., Rosier, C., Dams, A., s'Heeren, C., Wattiez, R., & Mergeay, M. (2009). The response of Cupriavidus metallidurans CH34 to spaceflight in the international space station. *Antonie Van Leeuwenhoek*, *96*(2), 227–245.

[40] Li, S., & Mason, C. E. (2014). The pivotal regulatory landscape of RNA modifications. *Annual Review of Genomics and Human Genetics*, *15*, 127–150.

[41] Liang, Y., Strelkov, S. E., & Kav, N. N. V. (2010). The proteome of liquid sclerotial exudates from Sclerotinia sclerotiorum. *Journal of Proteome Research*, *9*(6), 3290–3298.

[42] Liu, M., Gao, H., Shang, P., Zhou, X., Ashforth, E., Zhuo, Y., Chen, D., Ren, B., Liu, Z., & Zhang, L. (2011). Magnetic field is the dominant factor to induce the response of Streptomyces avermitilis in altered gravity simulated by diamagnetic levitation. *PLoS One*, *6*(10), e24697.

[43] Loman, N. J., & Watson, M. (2015). Successful test launch for nanopore sequencing. *Nature Methods*, *12*(4), 303–304.

[44] Luo, A., Gao, C., Song, Y., Tan, H., & Liu, Z. (1998). Biological responses of a Streptomyces strain producing-nikkomycin to space flight. *Hang Tian Yi Xue Yu Yi Xue Gong Cheng= Space Medicine & Medical Engineering*, *11*(6), 411–414.

[45] Mahan, M. J., Slauch, J. M., & Mekalanos, J. J. (1996). Environmental regulation of virulence gene expression in Escherichia, Salmonella, and Shigella spp. *Escherichia Coli and Salmonella: Cellular and Molecular Biology*, *2*, 2803–2815.

[46] Maxwell, S., & Drysdale, A. E. (2001). *Asssessment of Waste Processing Technologies for 3 Missions*. SAE Technical Paper.

[47] Mayer, F. L., Wilson, D., & Hube, B. (2013). Candida albicans pathogenicity mechanisms. *Virulence*, *4*(2), 119–128. https://doi.org/10.4161/viru.22913

[48] McCullough, M. J., Ross, B. C., & Reade, P. C. (1996). Candida albicans: a review of its history, taxonomy, epidemiology, virulence attributes, and methods of strain differentiation. *International Journal*

ISSN: 2583-4053

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https://doi.org/10.55544/jrasb.1.5.3

of Oral and Maxillofacial Surgery, *25*(2), 136–144. https://doi.org/https://doi.org/10.1016/S0901-5027(96)80060-9

[49] McIntyre, A. B. R., Rizzardi, L., Yu, A. M., Alexander, N., Rosen, G. L., Botkin, D. J., Stahl, S. E., John, K. K., Castro-Wallace, S. L., McGrath, K., Burton, A. S., Feinberg, A. P., & Mason, C. E. (2016). Nanopore sequencing in microgravity. *Npj Microgravity*, *2*(1), 16035. https://doi.org/10.1038/npjmgrav.2016.35

[50] McPhee, W. J., & Colotelo, N. (1977). Fungal exudates. I. Characteristics of hyphal exudates in Fusarium culmorum. *Canadian Journal of Botany*, *55*(3), 358–365.

[51] Mehta, S. K., Laudenslager, M. L., Stowe, R. P., Crucian, B. E., Feiveson, A. H., Sams, C. F., & Pierson, D. L. (2017). Latent virus reactivation in astronauts on the international space station. *NPJ Microgravity*, *3*(1), 1–8.

[52] Mermel, L. A. (2013). Infection prevention and control during prolonged human space travel. *Clinical Infectious Diseases*, *56*(1), 123–130.

[53] Mikheyev, A. S., & Tin, M. M. Y. (2014). A first look at the Oxford Nanopore MinION sequencer. *Molecular Ecology Resources*, *14*(6), 1097–1102.

[54] Milojevic, T., & Weckwerth, W. (2020). Molecular mechanisms of microbial survivability in outer space: a systems biology approach. *Frontiers in Microbiology*, *11*, 923.

[55] Nickerson, C. A., Goodwin, T. J., Terlonge, J., Ott, C. M., Buchanan, K. L., Uicker, W. C., Emami, K., LeBlanc, C. L., Ramamurthy, R., & Clarke, M. S. (2001). Three-dimensional tissue assemblies: novel models for the study of Salmonella enterica serovar Typhimurium pathogenesis. *Infection and Immunity*, *69*(11), 7106–7120.

[56] Nickerson, C. A., Ott, C. M., Mister, S. J., Morrow, B. J., Burns-Keliher, L., & Pierson, D. L. (2000). Microgravity as a novel environmental signal affecting Salmonella enterica serovar Typhimurium virulence. *Infection and Immunity*, *68*(6), 3147–3152.

[57] Novikova, N. D. (2004). Review of the knowledge of microbial contamination of the Russian manned spacecraft. *Microbial Ecology*, *47*(2), 127–132.

[58] Novikova, N., De Boever, P., Poddubko, S., Deshevaya, E., Polikarpov, N., Rakova, N., Coninx, I., & Mergeay, M. (2006). Survey of environmental biocontamination on board the International Space Station. *Research in Microbiology*, *157*(1), 5–12.

[59] Ott, C. M., Bruce, R. J., & Pierson, D. L. (2004). Microbial characterization of free floating condensate aboard the Mir space station. *Microbial Ecology*, *47*(2), 133–136.

[60] Pollard, E. C. (1965). Theoretical studies on living systems in the absence of mechanical stress. *Journal of Theoretical Biology*, *8*(1), 113–123.

[61] Purevdorj-Gage, B., Sheehan, K. B., & Hyman, L. E. (2006). Effects of low-shear modeled microgravity on cell function, gene expression, and phenotype in Saccharomyces cerevisiae. *Applied and Environmental Microbiology*, *72*(7), 4569–4575.

[62] Rand, A. C., Jain, M., Eizenga, J., Musselman-Brown, A., Olsen, H. E., Akeson, M., & Paten, B. (2016). Cytosine variant calling with high-throughput nanopore sequencing. *BioRxiv*, 47134.

[63] Rezzonico, F. (2014). Nanopore-based instruments as biosensors for future planetary missions. *Astrobiology*, *14*(4), 344–351.

[64] Rooney, B. V., Crucian, B. E., Pierson, D. L., Laudenslager, M. L., & Mehta, S. K. (2019). Herpes virus reactivation in astronauts during spaceflight and its application on earth. *Frontiers in Microbiology*, *10*(feburay). https://doi.org/10.3389/FMICB.2019.00016/FULL

[65] Rosenzweig, J. A., Abogunde, O., Thomas, K., Lawal, A., Nguyen, Y.-U., Sodipe, A., & Jejelowo, O. (2010). Spaceflight and modeled microgravity effects on microbial growth and virulence. *Applied Microbiology and Biotechnology*, *85*(4), 885–891. https://doi.org/10.1007/s00253-009-2237-8

[66] Saletore, Y., Meyer, K., Korlach, J., Vilfan, I. D., Jaffrey, S., & Mason, C. E. (2012). The birth of the Epitranscriptome: deciphering the function of RNA modifications. *Genome Biology*, *13*(10), 1–12.

[67] Sathishkumar, Y., Velmurugan, N., Lee, H. M., Rajagopal, K., Im, C. K., & Lee, Y. S. (2014). Effect of low shear modeled microgravity on phenotypic and central chitin metabolism in the filamentous fungi Aspergillus niger and Penicillium chrysogenum. *Antonie van Leeuwenhoek*, *106*(2), 197–209.

[68] Schultz, J. R., Taylor, R. D., Flanagan, D. T., Gibbons, R. E., Brown, H. D., Sauer, R. L., & Pierson, D. L. (1989). biofilm formation and control in a simulated spacecraft water system: Interim results. *SAE Transactions*, 964–973.

[69] Senatore, G., Mastroleo, F., Leys, N., & Mauriello, G. (2018). Effect of microgravity & space radiation on microbes. *Future Microbiology*, *13*(07), 831–847.

[70] Sheet, S., Sathishkumar, Y., Sivakumar, A. S., Shim, K. S., & Lee, Y. S. (2017). Low-shear-modeled microgravity-grown Penicillium chrysogenum-mediated biosynthesis of silver nanoparticles with enhanced antimicrobial activity and its anticancer effect in human liver cancer and fibroblast cells. *Bioprocess and Biosystems Engineering*, *40*(10), 1529–1542.

[71] Simpson, J. T., Workman, R., Zuzarte, P. C., David, M., Dursi, L. J., & Timp, W. (2016). Detecting DNA methylation using the oxford nanopore technologies MinION sequencer. *BioRxiv*, 47142.

[72] Song, B., & Leff, L. G. (2005). Identification and characterization of bacterial isolates from the Mir space station. *Microbiological Research*, *160*(2), 111–117.

[73] Sonnenfeld Gerald, Butel Janet S., & Shearer William T. (2003). Effects of the Space Flight Environment on the Immune System. *Reviews on Environmental Health*, *18*(1), 1–18. https://doi.org/doi:10.1515/REVEH.2003.18.1.1

ISSN: 2583-4053

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https://doi.org/10.55544/jrasb.1.5.3

[74] Sonnenfeld, G., & Shearer, W. T. (2002). Immune function during space flight. *Nutrition*, *18*(10), 899–903.

[75] Sugita, T., Yamazaki, T., Makimura, K., Cho, O., Yamada, S., Ohshima, H., & Mukai, C. (2016). Comprehensive analysis of the skin fungal microbiota of astronauts during a half-year stay at the International Space Station. *Medical Mycology*, *54*(3), 232–239. https://doi.org/10.1093/mmy/myv121

[76] Sviridov, S. I., Ermolinskii, B. S., Belyakova, G. A., & Dzhavakhiya, V. G. (1991). Secondary metabolites of Ulocladium chartarum. Ulocladols A and B—New phytotoxins of terpenoid nature. *Chemistry of Natural Compounds*, *27*(5), 566–571.

[77] Taylor, P. W. (2015). Impact of space flight on bacterial virulence and antibiotic susceptibility. *Infection and Drug Resistance*, *8*, 249.

[78] Thévenet, D., D'ari, R., & Bouloc, P. (1996). The SIGNAL experiment in BIORACK: Escherichia coli in microgravity. *Journal of Biotechnology*, *47*(2–3), 89–97.

[79] Thiel, C. S., Tauber, S., Seebacher, C., Schropp, M., Uhl, R., Lauber, B., Polzer, J., Neelam, S., Zhang, Y., & Ullrich, O. (2019). Real-time 3D high-resolution microscopy of human cells on the international space station. *International Journal of Molecular Sciences*, *20*(8), 2033.

[80] Tixador, R., Richoilley, G., Gasset, G., Templier, J., Bes, J. C., Moatti, N., & Lapchine, L. (1985). Study of minimal inhibitory concentration of antibiotics on bacteria cultivated in vitro in space (Cytos 2 experiment). *Aviation, Space, and Environmental Medicine*, *56*(8), 748–751.

[81] Vesper, S. J., Wong, W., Kuo, C. M., & Pierson, D. L. (2008). Mold species in dust from the International Space Station identified and quantified by mold-specific quantitative PCR. *Research in Microbiology*, *159*(6), 432–435.

[82] Viollier, P. H., Kelemen, G. H., Dale, G. E., Nguyen, K. T., Buttner, M. J., & Thompson, C. J. (2003). Specialized osmotic stress response systems involve multiple SigB-like sigma factors in Streptomyces coelicolor. *Molecular Microbiology*, *47*(3), 699–714.

[83] Wilson, J. W., Ott, C. M., Zu Bentrup, K. H., Ramamurthy, R., Quick, L., Porwollik, S., Cheng, P., McClelland, M., Tsaprailis, G., & Radabaugh, T. (2007). Space flight alters bacterial gene expression and virulence and reveals a role for global regulator Hfq. *Proceedings of the National Academy of Sciences*, *104*(41), 16299–16304.

[84] Xiao, Y., Liu, Y., Wang, G., Hao, Z., & An, Y. (2010). Simulated microgravity alters growth and microcystin production in Microcystis aeruginosa (cyanophyta). *Toxicon*, *56*(1), 1–7.