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TRAVELING WITH STOWAWAYS: THE HUMAN MICROBIOME AND TRAVEL

Alisha Nicole Hughes Communications May, 2016

Faculty Adviser: Dr. David Hirschberg

Essay completed in partial fulfillment of the requirements for graduation with Global Honors, University of Washington, Tacoma

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Approved:

Faculty Adviser

Date

Director, Global Honors

Date

1. Introduction

In our globalized world people travel back and forth on a daily basis. According to the International Air Transport Association (*IATA*) on average, 8 million people per day use airplanes to reach their destination. This also means that they are exposed to bacteria, viruses and other microorganisms that linger on surfaces, are carried around by fellow passengers or float around in the air. An example for this is a person from New York City traveling to Cabo San Lucas in Mexico. After arriving in Mexico, the person from the U.S. decides to eat and drink local food and beverages. However, he or she soon feels sick because the organisms in New York City are different from the ones in Cabo San Lucas. The U.S. citizen's body is not accustomed to these stowaways and thus, this influences the host's health in a negative way.

Based on this scenario, the following thesis aims inform and, along with the proposed pilot case study, investigate how the human microbiome changes when the host travels and that those changes affect the individual's health. The next sections will first explain what the microbiome is, how it relates to human health and why conducting further research is relevant, before moving on to a proposed pilot case study.

2. The Human Microbiome

The word *microbiome* really just means that small, naturally occurring organisms live together, as a community, in a major habitat. "The assemblage of microorganisms themselves is referred to as the microbiota or microbial community" (Cox, Cookson & Moffatt, 2013, p. 2). The grouping of organisms can really be anything from bacteria, viruses and fungi to archaea, "phage, fungi and other microbial eukarya" (Cox, Cookson & Moffatt, 2013, p. 2). They all coexist with us human beings and in many cases we

actually depend on them. We are slowly starting to understand the importance of the human microbiota and some scientists even point out that there is "good evidence that humans co-evolved a requirement for their microbiota" (Marchesi, et al., 2015, p. 1).

When trying to understand what role these communities of tiny organisms play in regard to humans, researchers have to focus on more than just the individual itself. Outside factors, such as the location, influence and shape humans and their stowaways. When looking at our earth's geographical regions, one will find different habitats – deserts, oceans, tropical rainforest, and so forth. Within them, organisms have evolved, become extinct, learned to adapt, or even mutated. Thus, depending on the habitat, one will find different living beings. The same principle can be applied to the human body. We have different types of microbiomes in or on different body parts. One of the tasks of the research initiative *Human Microbiome Project* (HMP) is to map out which microbial communities are found in and on specific regions of the human body. Their analysis showed that, among many smaller regions, there are five major regions in which different communities of microbiomes can be found: the gut, oral, skin, and for females the placenta and vagina microbiome (Belizário & Napolitano, 2015, p. 2-4).

2.1 Microbiome Regions

Generally speaking, one has to realize that the five major regions can also be categorized in to sub-regions. Thus, when looking at the microbiome found in and around vaginas, it is necessary to focus on multiple areas. Belizário and Napolitano (2015) state that three different sites were chosen for swabs. They analyzed the organisms found in the mid-vagina, cervix and introitus and found over 200 phylotpyes.

As organisms in the vagina produce lactic acid and hydrogen peroxide, the pH value of this sexual organ is fairly low. Most organisms found in this area "belong to the phyla Firmicutes, Bacteroidetes, Actinobacteria, and Fusobacteria" (p. 5). Along with this analysis, scientists were able to correlate changes in the composition of the microbiome to possible infections, which will be discussed in *2.3 Microbiome and Health* of this thesis.

Medical researchers assumed that the microbiome of a fetus, the placenta and newborn all developed in a sterile environment. With the advent of culture free analysis techniques, such as shotgun DNA sequencing, it is evident that the developing fetus, the supporting organs' microorganisms originate from the mother. However, studies revealed that unique microbiota are found in the placenta. A healthy placenta is typically composed of 320 different microbial communities, which include "non-pathogenic commensal microbiota from the Firmicutes, Tenericutes, Proteobacteria, Bacteroidetes, and Fusobacteria phyla" (Belizário & Napolitano, 2015, p. 6).

Scientists were also able to see differences in preterm and term deliveries. An example for this is the ram-negative bacillus *Durkholderia*, which is "associated with preterm delivery and the gram-positive, rod-shaped, facultative anaerobic bacteria *Paenibacillus*" (Belizário & Napolitano, 2015, p. 6), which is linked to the term delivery. Along with this information, researchers noticed that the placenta microbiome is similar to the oral microbiome. A possible explanation for this overlap is the exchange of fluids between mother and the fetus (Belizário & Napolitano, 2015, p. 6). In addition to that, Belizário and Napolitano (2015) note that research done with mice has shown that the way the newborns were delivered matters as well. When comparing the

microorganisms of babies born via cesarean section with the ones born virginally, one will notice that the ones delivered by cesarean section show similarities to the skin microbiome. The newborns delivered vaginally on the other hand have a more diverse gut microbiome (Belizário & Napolitano, 2015, p. 6).

Just like the vagina microbiome, the oral microbiome can be parted into multiple regions as well. Belizário and Napolitano (2015) write that scientists looked at nine oral regions, which range from the cheek tissue to the tonsils, over the saliva, the tongue and the throat. The results showed over 300 different genera, however, the author points out that many are not able to colonize in other regions of the mouth – for example in the root canal system. Generally, the microbial communities vary, depending on the individual's diet. This is only one factor, among many, that influence the microorganisms. In the sections *2.2 Shaping the Microbiome* and *2.3 Microbiome* and *Health* of this thesis, the importance of diet will also be analyzed, as they do not only play a role in the oral, but also in the gut microbiome. (Belizário & Napolitano, 2015, p. 3-5).

As the skin is the largest organ of the human body, it harbors a large number of different microbial communities. It coexists with the host and for the most part, contains organisms that are harmless to the individual. Belizário and Napolitano (2015) point out, that in some cases they can even have a positive effect on the host. The type of microbial community found on the skin, depends on the layer of epidermis one is looking at. Thus, like other microorganisms on and in the human body, the exact types vary from individual to individual, but are still influenced by other factors as well (p. 6). Among the microorganisms found, is the *Actinobacteria phylum*. Additionally, the

"Gram-positive *Staphylococcus epidermidis* and *Propionibacterium acnes* are predominant on human epithelia" (Belizário & Napolitano, 2015, p. 6).

Despite the fact that the skin is the largest organ and also functions as a habitat for a large number of microorganisms, the gut has the most complex microbial communities. In an adult, analysis show, that the gram-negative Bacteroidetes and the gram-positive Firmicutes, are the most abundant stowaways. Along with them, the Actinobacteria, Fusobacteria, and Verrucomicrobia phyla are also found in the gut microbiome, but Belizário and Napolitano (2015) point out that this depends on the host. As the gut offers the microorganisms an anaerobic habitat, only certain species are able to live there. In addition to that, the various bacteria found in the gut have different functions, which will be discussed in 2.3 Microbiome and Health of this thesis (Belizário & Napolitano, 2015, p. 3). To date, most of our knowledge of the microbiome comes from analysis of fecal samples. They "contain between 1,000 and 1,150 bacterial species", however, "up to 55% are uncultivable and thus uncharacterized" (Belizário & Napolitano, 2015, p. 4). When it comes to other regions of the gastrointestinal tract, scientists can only assume that the communities there vary as well. Belizário and Napolitano (2015) note that researchers are limited in accessing other parts of the gut. Thus, the large number of uncharacterized microorganisms is mainly due to sampling limitations (p. 4).

2.2 Shaping the Microbiome

As previously mentioned, the microbiome is dependent on the host. This means the host's lifestyle, in terms of location, diet, race, and gender, influences what microbial communities are found in and on the individual. Konkel (2013) points out that scientists

have noticed changes in the gastrointestinal tract, depending on the host's age. The first notable differences in microbial communities can be detected after the first few months of life. Next, when babies transition from breast milk to solid food, the gut microbiome changes significantly as well. Konkel (2013) states that once a child is approximately two years old, "the gut microbial composition (...) resembles that of an adult" (Konkel, 2013, p. 2). Thus, when it comes to the host's age influencing the microbiome, only slight changes, for example during puberty, are expected. Next, Dey, et al. (2015) have concluded that the host's diet influences the microorganisms living in or on him or her significantly (p. 95). Along with that, Marchesi et al. (2015) write that the microbial "community evolved to work in partnership with its host's digestive system to derive energy and carbon from complex plant polysaccharides which would otherwise be lost in faeces" (p. 3). This means the host supplies the microbes through the type of food consumed and in return, the microbial community in the gut helps the host with digestion. They change polyphenols "into absorbable biological active species" (Marchesi, et al., 2015, p.3). How well this relationship functions depends on the hosts diet.

Another factor that influences the occurring microorganisms is the various different bacteria, fungi, phage, viruses and more interacting with each other. This influences what composition of communities is found in or on the host. Depending on the species, they can live together or not (Estrela, Whiteley & Brown, 2015, p. 1-2). For example two communities can coexist, compete with each other or even exploit each other. One scenario Estrela, Whiteley and Brown (2015) describe is microorganisms living in aerobic and anaerobic conditions. Anaerobe and aerobe cannot coexist and this

creates "competitive interactions (mediated by shared consumption of glucose)" (Estrela, Whiteley & Brown, 2015, p. 3).

Apart from the microorganisms' direct environment, the host's surroundings also influence which microbes are found. Betts (2011) writes that toxicologists have conducted research on how the environment - particularly dangerous toxins in the air affects the microbial communities on and in humans. Their research revealed "one family of microbes may biotransform contaminants into a new form, whereas another group may transform that new form back into its previous form or into something completely new with completely different biological activity" (Betts, 2011, p.

4).

Like animals and plants, not every microbial species can survive under certain conditions. The skin's communities are directly exposed to the environment and thus, different climate zones and the host's general surrounding environment can lead to shifts. Thus, when comparing people living in tropical regions to those living in arctic regions, one will find different microorganisms. In addition, researchers noticed that race is also a factor that influences the skin microbiome. When comparing the microbes of Europeans to Africans or Asians, one will see other microbial communities (Belizário & Napolitano 2015, p. 6).

2.3 Microbiome and Health

This section will show how the microbiome does more than coexist with an individual, but also influences its host's health. Owyang and Wu (2014) write that the gut microbiome in particular influences the individual's metabolism. The microorganisms provide a "wide range of otherwise inaccessible metabolic capabilities" (Owyang & Wu,

2014, p. 3). As previously mentioned, some microorganisms can prevent others from coexisting with them. It is however also worth mentioning that in some cases, microbes can be overrepresented. This in turn leads to a shift in balance that again can cause other chain reactions. The microbial communities that were once considered 'good' can "induce inflammation and trigger disease throughout the body, even in the nervous system" (Konkel, 2013, p. 1).

In addition, the individual's immune system also has a direct connection to the microbial communities. Scientists conducted research on germ-free animals to analyze the relationship between gut microbiome and immune system. The results showed unusual gut and immune activities, which led to scholars' assumption that the gut microbiome plays an important role to the host's immune system. The organisms living in the intestines interact with the immune system, which causes it to build up immune resistance. However, it is important to also realize that the individual's immune system also influences the microbial communities found in the gastrointestinal system. Thus, when this system gets out of balance, the host may have an increased chance of developing pathologies such as inflammatory bowel diseases (IBD) or metabolic diseases (Belizário & Napolitano, 2015, p. 3; Owyang & Wu, 2014, p. 3).

Furthermore, scientists found out that the gut contains microorganisms that help with digestion. Research revealed that diet plays an enormous role when it comes to pathogenic diseases. Depending on the host's food consumption, the composition of the gut microbiome can change and thus, can lead to negative changes in the host's health. Information about the gut microbiome's composition is also used to prevent and treat disorders. Researchers are finding strategies to use "full-spectrum microbiota therapies

and defined microbiota ecosystems" (Owyang & Wu, 2014, p. 4) as ways of treating illnesses. Based off of this information, Marchesi et al. (2015), write that scientists' attention was drawn to the possibility of the gut microbiome influencing obesity (p. 4). This growing body of evidence has lead to a better understanding of obesity arising from several metabolic phenotypes, rather than one.

The microorganisms in the gut also influence an individual's liver. 70% of the blood the liver gets comes from the gut through the portal vein, which means the microbial communities in the gut are transported this way as well. Researchers were able to detect the close relationship between incidences of liver disease and changes in the gut microbiome. This observation has let to the development of new approaches in microbiome therapy to treat liver disease (Marchesi, et al., 2015, p. 4).

Along with this information, research in Russia has proven that the gut's microbial communities also influence the host's brain. Scientists realized that this connection between gut microbiome and the brain can also be used to help fight senility (Konkel, 2013, p.1). Additionally, scholars learned that the host's sex, in combination with the microbiome, could also influence the individual's wellbeing. Scientists wanted to understand why men, in comparison to women, were less likely to suffer from celiac disease. Studies conducted with mice revealed that their sex hormones, in combination with the microbes, may be the cause for this. It turns out that prior to puberty, the gut microbiota between males and females is very similar but then changes. Thus, the microbial communities existing in the gut prior to puberty, before the sex hormones affect the host significantly, enable both male and female to be equally protected (Konkel, 2013, p. 2).

In addition to the consumption of food, it is also worth mentioning that medication also affects the microbiome. Most research is focused on antibiotics. As they are used to combat bacteria, it appeared to be logical that this would also affect the 'good' bacteria, specifically in the intestine. Therefore, the studies with antibiotics revealed that they are in fact harmful to the host's natural gut microbiome. Konkel (2013) writes that upon taking the antibiotics, instant reduction and thus, instability are the consequence. Furthermore, sequencing four years after the consumption showed that the microbial communities only partially recover (p. 4).

Like the gut microbiome, the skin microbiome is also linked to the host's immune system. Similar to the microorganisms in the intestine, overpopulation of certain species, mainly influenced by the environment or the host's diet, can cause an imbalance. This in turn leads to clogged pores and inflammatory conditions of the skin, such as atopic dermatitis. The imbalance caused by the microbial communities usually shows as "abnormal flaking and itching of the scalp" (Belizário & Napolitano, 2015, p. 6).

Moving on to the connection between the microbiome of the mother and her offspring, this paragraph will examine how birth and diet matter for the newborn. As mentioned in the previous section, Belizário and Napolitano (2015), point out that there are measurable differences between newborns delivered via vaginal and Cesarean section. Based on this information, scholars noticed that babies born via the Cesarean section were more likely to develop "type 1 diabetes, celiac disease, asthma, and obesity" (Belizário & Napolitano, 2015, p. 6). The reason for this is that these newborns lack the microorganisms coming from the mother's vagina microbiome. In addition to

that, whether the mother breast feeds her child or not also impacts the baby's health, as important microbial communities are also transferred from the mother's milk. This adds to the child's microbiome, which in turn help the child's immune system (Belizário & Napolitano, 2015, p. 7).

3. Relevance

After explaining what the microbiome is and how it affects humans' health, the next question that arises is: Why should further research be conducted? The answer to this is that the proposed pilot case study would give insight on *what* kind of changes the microbial communities undergo when being introduced to new, foreign microorganisms and also help understand *how* these changes affect the host. Furthermore, the results of the research can be used to find ways of boosting the individual's immune system and in turn, this would prevent or decrease the host's chance of getting ill. Along with that, the scenario described in the introduction of this thesis, about a person from the U.S. getting sick from consuming food and water in Mexico, could be avoided.

In addition to that, the U.S. Army has shown interest in this field as well. They have started research on how the human microbiome affects soldiers' performance. Soldiers, like other military or government personnel, travel on a regular basis. On average, a service member is relocated every two to three years. In addition to that, they have to attend training camps for two to six weeks and leave to warzones for four to twelve months (Benson, 2014). Therefore, a soldier's body is put under a lot of stress and thus, preventing illnesses – especially the ones related to traveling to foreign countries – is in the Army's interest.

Currently, the U.S. Army's research focuses on the gut microbiome and its

effects on the host's health; however, the results from this pilot case study could help improve military service members' health by boosting their immune system as well.

4. Pilot Case Study

The case study proposed here aims to analyze the effects that travel can have on the human microbiome. The goal is to determine whether or not the composition of the microbial communities changes, if these shifts are significant and lastly, how they affect the host's health. To ensure that the data resulting from this analysis is accurate, a group of at least 40 people is necessary. Of course, larger numbers would be preferred. Nevertheless, 40 participants would be enough to answer the basic questions and encourage further, more detailed research.

Next, the entire group of people has to travel to the same location, at the same time and be under the same conditions. This means, they all have to be on the same, or at least a very similar diet and be exposed to the same weather conditions. To receive a contrast in environment, climate, diet and so forth, the participants should be flying overseas. Lastly, the amount of stress that each individual experiences should also be as comparable as possible.

Moving on to the sample giving part, all the participants need to be equipped with a swab kit suitable for sequencing 16S rRNA, which will be explained in more detail in the methods section. In addition to that, the individuals need a notebook, voice recorder or video camera, which they will use as diary for documentation purposes. Each member in the group should swab her or his skin on the inside of the wrist. When first starting with giving samples, each person should be swabbed one week prior to departure. It is recommended to take samples at least once a day, but it would be ideal

to repeat the process three times a day. In addition to giving samples, the individuals should keep track of time and repeat the swabbing every day at the same time. After taking their sample, the participants are asked to write down or record the time and date. In addition to that, they should list what they ate since giving their last sample, how they feel emotionally (are they stressed, tense, relaxed, and so forth), rate their overall health (do they have any complaints, such as feeling sick or being in pain), and lastly, if they have been in contact with other individuals, animals or plants. For convenience purposes, the sample would be sent back to the laboratory periodically, depending on how long the group is overseas. After their travel, when they return back to the starting point, the participants should immediately take another sample and then continue with their routine for an additional week. Thus, to make it short: People would start swabbing at the starting point one week before departure and end one week after their return.

4.1 U.S. Military

When it comes to receiving samples from living beings as described above, researchers usually have difficulties with finding a group that match these conditions. However, with the U.S. Army showing interest in the field of human microbiomes, the case study could be conducted with active duty military service members. People serving the country are usually relocated – they undergo a Permanently Change of Station (*PCS*) - after two to three years. This means they leave their current base and move to a different one, which can be within the United States and its territories or somewhere overseas, for example Europe. In addition to 'PCSing', soldiers, airmen, seamen, and marines are sent to training stations or camps on a regular basis.

Depending on the service branch, this temporary change in duty station can range from a few days to a few weeks or even months. Lastly, deployments are also part of military service members' lives. This too means that they are relocated for months or even an entire year to a destination anywhere in the world.

Not only would the military, as participants of the case study, ensure travel, it would also mean that one could focus on a specific operational unit, such as a *Platoon*, which consists of 16-40 soldiers. Furthermore, the case study could be conducted with a Platoon deploying overseas, which means they would all leave from the same starting point: their base. Also, when it comes to the soldiers' diet, Meal Ready to Eat (MRE) or canteen food are usually very similar which again would come closest to the case study's requirements (*Go Army*). Worth mentioning is the fact that the soldiers that deploy together do not just have the same destination, but are also likely to encounter the same or similar stress levels.

Based on this information, the U.S. military would be the perfect group for this case study. Not only because their structure gives each individual's similar conditions, but also because of their already existing interest and research on the human microbiome. The data and conclusions resulting from this case study would be valuable to the military as an organization and to other health related organizations.

4.2 16S rRNA Sequencing

There are various different types of microorganisms living on and in humans and in some cases the exact species is not known. This means it is hard to analyze and label the samples properly. Nevertheless, researchers have come to know more species and have improved their methods of analysis. Databases were created that

harbor the known data on different types of microorganisms. To combat the lack of knowledge, researchers have been able to find an alternative that allows them to at least pinpoint a larger microbial community to study them. By doing so, they are now able to "gather substantial detail about individual organisms and entire communities, whilst circumventing the selection biases inherent in isolation by culture" (Cox, Cookson & Moffatt, 2013, p. 90).

As previously mentioned in the text, the quickest and most inexpensive way to analyze the samples collected from the soldiers, is 16S rRNA sequencing. Therefore, the probes will be sent to a laboratory where this method of sequencing will show which microorganisms are present. 16S rRNA allows scientists to get a quantitative description of the bacterial composition of the sample.

The probes collected from the U.S. soldiers are in fact a very complex biological mixture. Thus, to understand the process of 16S rRNA gene sequencing, it is important to comprehend *what* 16S rRNA is and what it means. According to Sanschargin and Yergeau (2014), RNA, which is short for ribonucleic acid, can be found in all microorganisms. In addition to that, RNA can be used to determine whether or not these organisms are related; this makes RNA essential. 16S rRNA functions as a key to unlock information. It is a gene used to encode RNA, in this case RNA of microbial communities found on the soldiers.

When the samples arrive in the laboratory the fist step to finding out what they are composed of, is to apply primers. This means, a starting point is set on the genes. The samples will be purified, quantified and attached to sequencing spheres. Thus, they can be duplicated with Polymerase Chain Reaction (PCR). The clones experience

changes in temperature for a designated time period over and over. The next step is to sequence the probes before they can be analyzed. The data information resulting from the sequencing shows what species of bacteria are found by comparing the results to data in the already existing database on microorganisms (Sanschargin & Yergeau, 2014; Cox, Cookson & Moffatt, 2013, p. 90 & 91).

By following these steps, one can compare the occurring microbial communities of soldiers before, during and after their deployment. The 16S rRNA sequencing will show if new microorganisms have emerged, previously existing ones have disappeared and overall how the balance of the microbial communities has shifted.

4.3 Expected Outcomes

As this study has not yet been conducted with humans, one can only assume how travel would affect the microbiome and its host's health. To gain insight on what the results could look like and how humans could benefit from them, the results of similar case studies will be explored below.

4.3.1 Mice and Travel

Dey, et al. (2015) write about an experiment conducted with germ-free mice in their article. Though it is important to note that the mice did not actually change their environment and thus, did not travel in reality. Nevertheless, Dey, et al.'s (2015) journal article focuses on how changes in diet, resulting from travel, influence the gut microbiome.

Dey, et al. (2015) write that the first step of this study was to colonize the healthy male mice with the gut microbiome of also healthy humans from different parts of the world. The goal was to create the conditions of global travel, focusing on the host's

eating habits. By doing so, the researchers ensured a high diversity and thus, high contrasts.

The case study consisted of two experiments: short-term and long-term travel. With the short-term travel the host would only have a brief change in diet, which means the mice were fed different food for eight days. For the long-term travel, the diet was changed for 14 days (Dey, et al., 2015, p. 95 & 96).

Once the mice received their human donor microorganisms, the scientists ensured that the bacteria would differ and yet still be able to grow and coexist within the mice's own gut organisms. In regard to parallels between the case study described by Dey et al. (2015) and the pilot proposed in this paper, the aim was to find out how changes in the host's life affect the microbial communities living on, or in this case in the host. In Dey et al.'s (2015) article the scholars seeked to examine how different foods affect diet trends, how long it would take for the microbiota to adjust and how this too could potentially influence other health aspects of the mice (p. 96).

As mentioned, to start the experiment human microorganisms were necessary. Thus, fecal samples from six adults were obtained, which were then injected into six germ-free mice. Next, the new hosts were fed the same types of food for eight days to simulate a *home diet*. The following days, their meal plan shifted, which should resemble the change that comes with travel and consumption of different foods. For the first experiment, the short-term, this new diet lasted for eight days and for the second experiment, it lasted for 14. After that, the scholars ensured that the mice's eating habits reverted back to their *home diet* (Dey, et al., 2015, p. 96-99).

The method used to analyze these mice was 16S rRNA sequencing and Dey et

al. (2015) write that the results of this experiment showed that mice with microbial communities originating from Bangladeshi donors differ significantly to the ones with microorganisms from the U.S. In addition, the mice with the U.S. donor microbiota "had significantly faster motility (i.e., shorter transit times) when consuming the Bangladeshi diet compared to primal diet" (p. 96).

In conclusion to this case study on mice, types of food consumed by the host did have an effect on the microorganisms. Thus, one can assume that the same or similar results can be seen when humans travel and consume food from different parts of the world to which their microbial communities are not accustomed to (Dey, et al., 2015, p. 95-97).

4.3.2 Microbial Cloud

As the previously described case study only focuses on the gut microbiome and not the entire biome, the results are not fully applicable to the pilot case study proposed in this thesis. The study proposed in this text, with the U.S. soldiers, aims to analyze how travel affects the host's health and his or her skin microbiome. As mentioned in the literature review, the skin microbiome is also linked to the gut microbiome and thus, changes in diet do affect the composition of the microorganisms found on the skin. In addition to that, these shifts in balance also affect the individual's health. In both cases, the research described by Dey et al. (2015) does not consider these changes and effects. Furthermore, when traveling around the world it is not just the host's diet that changes. Environmental changes, such as climate, plants, animals and other people surrounding the host can also affect the microbiome and thus, the individual's health.

Therefore, to get a better understanding of what the results of the proposed pilot

study might show the following text will elaborate on another project. The purpose of this one is to show how responsive the human microbiome is and that mere interaction with other individuals and overall changes in the host's surrounding affect the microbial communities. The next study is presented by Meadow, et al. (2015) and focuses on proving the existence of a microbial cloud that surrounds each individual. In the journal article, Meadow et al. (2016) write that the scholars' goal was to not just find out if the cloud existed but also if it was unique to each individual, like a finger print (p. 3).

In order for them to examine this, a chamber was completely sterilized and essential artifacts in the room were made of plastic to reduce bacteria. Furthermore, "continuous fan powered supply air was used to maintain positive pressure in the chamber" (Meadow et al., 2015, p. 4). Along with that, the filters too were placed to ensure that dust and bacteria within the small room were controlled and kept at a minimum. To make sure that the conditions were tracked and remained stable, a ventilation meter and a thermoanemometer probe were put in place as well (Meadow et al., 2015, p. 4).

Prior to the experiment, the contestants were examined and considered healthy. Also, it was important to the researchers that testers did not take antibiotics at least four months prior to the experiment, as this could interfere with the natural composition of the microbiome of the individual (Meadow et al., 2015, p. 3 & 4).

Once the instruments for measurement were set up, "each occupant sat in a disinfected plastic rolling chair surrounded by 12 sterile (...) air filters that were arranged equidistant from the seat" (Meadow et al., 2015, p. 4). However, only one individual at a time occupied the chamber. Each person was in it for intervals of 240 minutes and then

120 minutes (p. 3).

After the experiment was completed, the information collected from the ventilation system was analyzed. As in the previous case study with the mice, here too, 16S rRNA sequencing was the method of choice. The results showed that one could tell, simply by looking at the air samples, when the room was occupied and when it was not. Also worth mentioning is the fact that scientists were able to see whether or not the room had been occupied for up to four hours after the occupant had left. After four hours, the ruminants became harder to detect (Meadow et al., 2015, p. 8). In addition to that, the results enabled researchers to see a distinct difference between the individuals. This means that people do emit a microbial cloud and that they are unique to the hosts, just like a fingerprint (p. 11).

4.4 Pilot Case Study Conclusion

In conclusion, these two case studies only partially help predict the results from the pilot case study. In both studies, the scientists only focused on specific regions of the host but they neglected to view the entire picture. Furthermore, in the case of the study conducted with mice, the scholars only changed the diet. However, as mentioned, in reality, travel affects more than just the eating habits of the host. Nevertheless, these two case studies still showed that there are differences between individuals in regard to their microbial cloud. Thus, when being around people, it is possible for the organisms to interact with each other. This in turn means that when traveling to a different culture, one is exposed to foreign microorganisms that can lead to a shift in balance of the microbiome. Along with that information, Dey et al.'s (2015) article proves that the changes in diet affect the microbiome and thus, scientists should be able to see a

significant difference in microbial communities when comparing the samples collected from soldiers at the home base with the ones from overseas. Most importantly, both case studies prove that the microbiome is responsive and thus, is able to shift in balance. This means that, as discussed in the literature review of this thesis, those shifts in balance can affect the host's health.

5. Conclusion

In conclusion, this thesis shows that more research, in regard to the human microbiome and travel, needs to be conducted in order to fully understand the microorganisms' effects on the host. The case studies conducted by Dey et al. (2015) and Meadow et al. (2015), along with the general information on the human microbiome provided by Belizário and Napolitano (2015) and others, only provide a skeleton. However, the proposed pilot case study could be used to fill this skeleton and help answer questions not only in respect to travel and health, but also set a starting point for finding cures to illnesses.

Looking at the example of the person traveling from New York City to Cabo San Lucas, the pilot case study's results could prevent this individual from feeling sick after consuming drinks and foods in Mexico. It could be possible to add to our already existing stowaways to prevent illnesses or even cure them.

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