A Study of the Presence of Antibiotic Resistant Bacteria in Fitness Centers and Their Susceptibility to a Variety of Antibiotics

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Abstract:
The purpose of this project was to see if there was a presence of antibiotic resistant bacteria in fitness centers and also to determine their susceptibility to a variety of antibiotics. The hypothesis was that there would be a significant amount of the bacteria encountered in fitness centers and they would exhibit a degree of resistance. Also, it was further hypothesized that greater numbers of bacteria would be resistant to penicillin, since it is the oldest known antibiotic.

Ten machines were swabbed from the fitness center and then the bacteria were swabbed onto LB plates. Then five colonies were chosen from each plate and cultured. The cultures were then put onto LB plates and antibiotic disks from six different antibiotics were placed onto the plates.

After 24 hours the zones of inhibition from the antibiotic disks were measured and classified as resistant, intermediate, and susceptible according to the zone radii. The results showed that there were more colonies resistant to the penicillin and ampicillin as was hypothesized.

Introduction:

Imagine contracting “something” and doctors are frantically trying to figure out what the source of it is. Eventually, they determine it is an infection caused by antibiotic resistant bacteria. Now what? Where does one go from there? It is not as if the doctors can pick from an array of antibiotics for a cure. These questions among others go through a person’s head when diagnosed with an infection which is antibiotic resistant disease. The most powerful antibiotic used as a “last resort” is Vancomycin. What happens, however, if the bacteria become resistant to Vancomycin? Current research is being done on these problems. Scientists question where people are getting these antibiotic resistant diseases. The scary part is that a lot of it can be contracted at any given public facility, even upper class facilities are at risk.
Penicillin was the first antibiotic that was developed. In 1928, Sir Alexander Fleming isolated it from the mold found growing in his lab experiments (2). This revolutionized medical practice and improved the health of people all over the world. Since that time numerous antibiotics have been developed, but with each bacteria have mutated and developed resistance to many of them.

Vancomycin was introduced into the medical community about forty years ago, and it has played a very important role since. It is often been referred to as the “antibiotic of last resort (1).” Doctors turn to Vancomycin when no other antibiotics will work. An increasing number of diseases however are beginning to become resistant to even Vancomycin. Doctors wonder what to do? If an antibiotic is introduced into a hospital, it can be as little as a few months before resistant strains of bacteria can appear (1). When Vancomycin was first introduced doctors thought of it as a cure-all antibiotic. This thought ended in 1987 when the first strains of Vancomycin resistant bacteria appeared in hospitals.

Now cases of Vancomycin resistant bacteria are as frequent as ever, and serious research is being done to determine how to handle this growing problem. Scientists are a closer to understanding how antibiotic resistance spreads because the genome of the two main antibiotic resistant bacteria have been sequenced. Dr. Keiichi Hiramatsu and other colleagues at Juntendo University sequenced the genome of Staphlococcus aureus, one of the two main antibiotic resistant bacteria (2). Dr. Hiramatsu said “To avoid emergence of resistance, we should restrict use of antibiotics (2). Scientists say that part of the problem is that bacteria are very resilient and can develop ways to survive drugs that should kill them. Studies show
that about 70% of bacteria are resistant to at least one of the drugs most commonly used to treat infections (3). To try to help this problem doctors should avoid giving antibiotics if possible. If doctors continue to prescribe so many antibiotics then the problem of antibiotic resistant bacteria will continue to rise steadily.

To fully understand antibiotic resistant bacteria, scientists must question “where does it come from?” Fitness centers provide an excellent site for bacterial transmission and growth. They provide warm, moist environments for bacteria to grow. To prevent contracting bacteria while using a fitness center, one should always use a barrier between them and the equipment.

The purpose of this project was to determine whether patrons of fitness centers are being exposed to antibiotic resistant bacteria during their routine workouts. How many antibiotic resistant bacteria are present in fitness centers? If there are any antibiotic resistant bacteria, to which antibiotic are they most resistant?

The hypothesis for this project was that a significant amount of the bacteria encountered in fitness centers will exhibit some degree of resistance. It was further hypothesized that greater numbers of bacteria will be resistant to penicillin, since it is the oldest known antibiotic.

**Methods and Materials:**

**Materials:**

10 LB Agar Plates

Sterile swabs

Antibiotic disks (ampicillin, tetracycline, streptomycin, penicillin, kanamycin, and chloramphenicol)
LB Broth
Sterile Water
Inoculating loops
Bacteria spreader
Disinfectant used at fitness center (100 mL)
10 Ziploc bags
50 Falcon Tubes (15 mL)
Antimicrobial susceptibility test guide

Procedure:
1. After choosing a fitness center location, choose 10 machines to test.
2. Moisten sterile swab with sterile water and then thoroughly swab the handle area of the machine. (repeat with each machine)
3. Streak an LB plate for each machine. Let grow for 24 hours at 37 degrees Celsius.
4. Using an inoculating loop, select 5 colonies from each plate and start different cultures of each in 15 mL Falcon Tubes of LB broth. Let grow for 24 hours at 37 degrees Celsius in a shaking water bath.
5. Prepare pour plates by adding 150 uL of each bacteria culture into 3 mL tubes of melted R-top soft agar and vortex.
6. Pour onto LB agar plate and swirl to cover entire plate.
7. Place each of the 6 different antibiotic disks onto the agar plate in a circular pattern, separated equally from each other.
8. Seal with para-film and incubate.
After 24 hours, measure the zones of inhibition in millimeters for each antibiotic disk.

Results:

Bacteria were isolated from 10 different fitness machines. Five morphologically different colonies from each machine’s plates were cultured in tubes of LB broth for a total of 50 cultures. After plating the bacteria onto LB plates, the six antibiotic disks were placed on each of them. Examination after 24 hours of incubation revealed considerable differences between the antibiotics and also differences in the degree of susceptibility to the antibiotic. The greatest number of plates were most resistant to ampicillin. 34% of all the cultured bacteria plates were highly resistant to it. There was also a high degree of resistance, 32%, to penicillin. On the remaining plates, less than 10% were highly resistant to streptomycin, kanamycin, chloramphenicol, and tetracycline.
Many of the bacteria cultures exhibited high susceptibility to the antibiotics. Interestingly enough, 56% and 44% of the plates showed high sensitivity to ampicillin and penicillin respectively. Tetracycline, streptomycin, and kanamycin all ranged from 26% up to 58% that were susceptible.
The graphs below show the degree of susceptibility that the bacteria cultures had to all six antibiotics.
Analysis and Conclusion:

The results of this experiment provide evidence that people are being routinely exposed to antibiotic resistant bacteria in public facilities. It is concluded that there are considerable differences among the resistance of the bacteria to the different antibiotics.

It was found that the greatest amount of bacterial resistance was to ampicillin and penicillin. This is likely due to the fact that penicillin is the oldest and most widely used antibiotic, and ampicillin is a derivative of penicillin. However, penicillin and ampicillin were quite effective in destroying many of the cultured bacteria, thus implying that they do continue to be an effective antibiotic. The high resistance percentages for penicillin and ampicillin were 32% and 34% respectively. The low susceptibility percentages were 44% and 56% respectively. This shows that the antibiotic is at one extreme or the other. This is somewhat of a double-edged sword.

The results of this project do support the hypothesis that there are significant numbers of bacteria that are highly resistant to typical antibiotics. Also, greater numbers of bacteria were resistant to both ampicillin and penicillin, further supporting the hypothesis.

One possible source of error is the likelihood that the same bacteria were present on the different machines and may have been used as a sample more than once. Without further identification of these bacteria, this would be difficult to overcome this problem.

The results also reaffirm the concern for overuse of antibiotics. The more that antibiotics are used the greater risk there will be for resistance. This project taught
me the significance of antibiotics and also the dangers they can posses. I learned that they are not to be used generously so to keep resistance levels low. If I could have done this project differently, I would have tested more of a variety of machines.

Possible future studies for this project might include testing other fitness centers and other public facilities to compare percentages of antibiotic resistant bacteria. Also, it would be extremely interesting to run other tests like Gram staining to determine more specifically what these bacteria are.

Acknowledgements:

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References:


**Data Table:**

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