EXTENDED ESSAY

Research Question: How do five different types of countertops: polished chipboard, granite, marble, formica and laminate affect the total number of different types of bacteria on these surfaces which is indicated by viable cell count (colony counts) method?

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ABSTRACT

Since we live in an open ecosystem; we always interact with so many different living things and mostly we are not able to see what organism we are interacting like bacteria. Some of these bacteria do not affect human body but some of them can be really harmful for human-being. Most of the foods like vegetables contain bacteria that could be pathogenic and we may be infected because of these bacteria. Even we wash them very carefully; bacteria can locate in our kitchen countertops and can be transmitted to us. So the focus of this study is to find out which type of five different countertops provides the most hygienic environment for mankind.

The aim of this experiment is to compare total number of different types of bacteria due to different types of countertops. Since infeasibility to examine every types of countertop in the world, five most common types of countertops which are; granite, laminate, polished chipboard, marble and formica are choosen for the experiment.

In this experiment; the mean total number of different types of bacteria that was grown over granite, laminate, marble, formica and polished chipboard is respectively 203.2, 77.2, 76.8, 59.8 and 35.6. So polished chipboard provides the most hygienic environment as a countertop within the group while granite provides the least.

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A. INTRODUCTION

I really love to make dinner for myself and I really enjoy while eating them. Like all other cooks, I want my kitchen sterile and I am very rigorous about my meals. Especially I really like to make exotic salads in my kitchen: Caesar, chopped, Greek, Hawaiian, Italian, Antillaise, Pittsburgh... Although I really like to eat them, I am worried about their hygiene since, these kinds of vegetables used in salads like lettuce, cucumber, radish, cabbage…etc grow nearer to the soil; they have high potential to serve as a nutrient and accommodation source for the bacteria. If those bacteria are not cleaned carefully, they can cause serious bacterial infection in human body. I have known that risk, so I was very careful while washing salad materials but I learned in our biology lesson that even we wash vegetables very carefully; bacteria can locate in our kitchen countertops before we wash them and can be transmitted to us by touching. By the agency of this information I found out my research question. I wonder that what type of kitchen countertop provides more anti-bacterial environment. To find an answer to this question I chose to prepare my extended essay in the subject of biology.

Since I decided to examine total number of bacteria on the different types of countertops, I made researches about bacteria and how they increase in number. As a result of my researches, I obtained these informations:

- Under optimal conditions bacteria can grow and divide extremely rapidly by binary fission (a form of asexual reproduction). Bacterial population can double as quickly as every 9.8 minutes.[1, 2]

[1] Koch A (2002). "Control of the bacterial cell cycle by cytoplasmic growth". *Crit Rev Microbiol* **28** (1): 61–7

[2] Eagon RG (1962). "Pseudomonas natriegens, a marine bacterium with a generation time of less than 10 minutes".Journal of Bacteriology 83 (4): 736–7.

- We are always in an interaction with lots of bacteria in our daily life but not all of them cause infection in human. A bacterium which causes bacterial infection is called Pathogenic bacteria.[3] These kinds of bacteria give harm to host organisms when they come into contact with. [4] They can enter our body through cut skin, food, water, air, saliva and other body fluids.[5] As we are able to see bacteria can interact with us very easily. Also they can survive in many environments for an extended time, lasting months or years until they interact with host.[6] So they can settle in our kitchen countertops and contaminate our food. Since their productivity capacity is very high, even one bacterium which locates on the surface of countertop can produce very rapidly and form lots of bacterial colony. So it is an important issue to use countertop which provides the least suitable environment for bacterial growth.

There are lots of countertop types in the world and it is impossible to use each of them in one experiment. So I decided to work with only five types of countertops. I want to make my decisions from various types of countertop. I use all wooden such as laminate and stone products like marble or granite. My choices are laminate, granite, polished chipboard, formica and marble. All of them are commonly used in manufacture of countertops.

There are different methods for calculating the total number of bacteria which will be discussed under the method development and planning title and I had to choose one of them for my experiment. I chose viable cell count (colony counts) method to calculate the total number of different types of bacteria.

[3] http://bacteriamuseum.org/cms/Pathogenic-Bacteria/pathogenic-bacteria.html
[4] http://www.helium.com/items/1588712-characteristics-of-pathogenic-bacteria
[5] http://www.buzzle.com/articles/pathogenic-bacteria-resistant-to-antibiotics.html
[6] http://www.leptospirosis.org/topic.php?t=27

To resolve my curiosity I decided to find out which of the countertops causes the least bacterial reproduction and I determined my research question as: "**How do five different types of countertops: polished chipboard, granite, marble, formica and laminate affect the total number of different types of bacteria on these surfaces which is indicated by viable cell count (colony counts) method?"** and throughout this paper answer of this question will be discussed briefly.

B. HYPOTHESIS

Bacteria can locate and grow more easily into the pits of countertops because they will be well protected from external forces, like cleaning swabs and sunlight, in these pits. Their total number will change due to different types of countertops because each countertop has different number and size of pits. So growth and reproduction of bacteria on the countertops are related with the number of pits on the countertop. More pits on the countertop will cause more production and location of bacteria.[7]

To create my hypothesis I did some researches about physical characteristics of countertop materials. These were laminate, granite, polished chipboard, formica and marble. As a result of my researches I found out granite has the biggest number of pits while polished chipboard has the least. If we order them descending way: number of pits over the surface would be granite> marble>laminate>formica>polished chip board.[8,9, 10,] while doing my researches, I hoped to find a numerical value of pits per square but there were any source in the web or in the local library in my city. Also size of the pits is as important as number of them but I had no opportunity to examine these values.

Since more pits on the countertop will cause more production and location of bacteria as we discussed in the first paragraph my hypothesis is: "**The total number of different types of bacteria over the granite countertop will be the most and the total number of different types of bacteria on the polished chipboard countertop will be the least."**

[7] http://www.greatlakesgranite.com/faq/more-granite-faqs
[8] http://www.ssdionline.com/characteristics_of_granite
[9] http://www.ask.com/question/physical-properties-of-marble
[10] http://www.builddirect.com/Laminate-Flooring/Laminate-Floors-Articles/Laminate_Flooring_Defined_Characteristics_of_Laminate_Floors.aspx

C. METHOD DEVELOPMENT AND PLANNING

In my experiment I will try to find out which countertop provides the least bacterial division. In the beginning, I thought to count just one type of bacteria, *Clostridium Perfringens,* but after my researches I found out that procedure would be completed in a very long time period (at least in 2 years). So I decided to count only the total number of bacteria.

To find out an answer to my research question: "**How do five different types of countertops: polished chipboard, granite, marble, formica and laminate affect the total number of different types of bacteria on these surfaces which is indicated by viable cell count (colony counts) method?"** total number of bacteria number should be observed from different kinds of countertops in a constant environmental conditions.

I chose to do my experiment with laminate, granite, polished chipboard, Formica and marble as countertops because these are commonly used in our daily life.

i. Stabilizing variables for countertops

Total number of bacteria in a countertop surface depends on so many variables like surface area or environmental conditions. So controlling these kinds of variables were extremely important for my experiment. In the beginning I thought to take samples from different countertops from different homes but this procedure was not practical because to take samples from different houses would take very long time and most importantly environmental conditions such as the detergents to use to clean countertops, temperature, humidity… etc were not stable. Also surface areas of the countertops were varying in this procedure. So I decided to buy countertops

from the store and cut it in the same size 125(25x25) cm² surface area and 1 cm height]. By this way, samples can be taken more easily.

The other important thing for my experiment was to stabilize environmental conditions for the counter tops. I used sterilized countertops to sustain same conditions from the beginning of this experiment. I decided to keep sterilized countertops together for 30 days so I put them in a carton box homogenously. By this way; all have been kept in the same environmental conditions.

ii. Finding laboratory for experiment

To count total number of bacteria on these countertops, after it is waited in the same environment for 30 days, I had to use microbiology laboratory. So I asked for permission from the dean of the pharmacy faculty Prof. Dr. Maksut COŞKUN and head of the department of Pharmaceutical Microbiology Dr. Bahar Bozkılınç to use laboratory of the department of Pharmaceutical Microbiology in the faculty of pharmacy in Ankara University. After he had heard my aim of experiment; he gave me permission to carry out my experiment in the faculty.

iii. Choosing appropriate method

Although; I did so many researches about taking bacterial samples from a surface and growing them in Petri dishes, my mind was still confused. There were lots of options to measure rate of bacterial growth but I could not understand which way is the most appropriate for my experiment. Below table includes some methods to measure the total number of bacteria.

Method	Application	Comments
Direct microscopic count	Counting the number of bacteria in milk or vaccines	**Cannot separate living cells from dead ones**
Viable cell count (colony counts)	Counting the number of bacteria in milk, foods, laboratory cultures, etc.	Very sensitive if plating conditions are optimal
Turbidity measurement	Counting large numbers of bacteria in clear liquid media and broths	Fast and non-destructive, but **cannot detect cell densities less than 10^7cells per ml**
Measurement of Biochemical activity e.g. O2 uptake CO2 production… etc.	Microbiological assays	Requires a fixed standard to relate chemical activity
Measurement of dry or wet weight of cells or volume of cells after centrifugation	Measurement of total cell yield in cultures	more sensitive than total number of bacteria measurements

Table-1: Some Methods used to measure bacterial growth[11]

There were six ways that I found and I had to choose one of them. Firstly; the method, that I choose, had to be able to count only living bacterial cells. So I eliminated direct microscopic count method because it cannot distinguish living cells from nonliving cells. Secondly; I did not know how will be the density of bacteria in a countertop. It could be less than 10^7cells per ml so I eliminated Turbidity measurement method too. I also eliminated measurement of biochemical activity and measurement of dry or wet weight of cells or volume of cells after centrifugation method because these methods were too complicated to maintain in a short period of time. The most suitable method was viable cell count (colony counts) method. This method is sensitive enough to measure the total number of bacteria and it can be carrying out in Pharmaceutical Microbiology laboratory easily.

[11] http://textbookofbacteriology.net/growth_2.html

iv. Viable cell count (colony counts) method

In this method we count number of colonies of bacteria in the medium. To do this method; I had to prepare three main things: Saline(APPENDIX-A), Tryptic Soy Broth (TSB) (APPENDIX-B) solution and Tryptone Glucose Extract Agar(TGEA)(APPENDIX-C) solution. Saline is used to maintain dilution process. It stabilizes osmotic pressure of water for the bacteria because osmotic pressure of pure water is too much for the bacteria and causes the bacteria to explode.[12] TSB is needed to collect bacteria from the countertop surface and TGEA provides an environment to grow in Petri dishes. In this method results are expressed as colony-forming units(CFU).(APPENDX-G)

[12] http://wiki.answers.com/Q/Why_is_saline_used_in_bacterial_serial_dilution?#slide=2

D. MATERIALS

- 15 pcs sterile Petri dishes
- 110 ml sterile saline solution in the sterile Erlenmeyer flask with cork stopper
- 4.8 g sterile Tryptone Glucose Extract Agar(TGEA)
- 3 g sterile Tryptic Soy Broth(TSB)
- 2 pcs sterile 500 ml capacity Erlenmeyer flasks with cork stoppers
- 10 pcs 2 ml pipette
- 15 pcs sterile borosilicate glass tube which have 150 mm height, 8mm radius with cork stoppers
- 5 pcs sterile 18 cm swab
- 1 pcs sterile 125(25x25) cm^2 laminate(1 cm height)
- 1 pcs sterile 125(25x25) cm^2 granite(1 cm height)
- 1 pcs sterile 125(25x25) cm^2 polished chipboard(1 cm height)
- 1 pcs sterile 125(25x25) cm^2 formica(1 cm height)
- 1 pcs sterile 125(25x25) cm^2 marble(1 cm height)
- 1 pcs carton box which has 3750(50x75) cm^2 surface area and 10 cm height.
- 1 pcs sterile 7L iron bowl which has 10 cm radius
- 5 L tap water
- 300 ml pure water
- borosilicate glass tube holder
- a lighter
- a chronometer
- a glass graduated cylinder
- an acetate pen
- a 40 watt lamp

E. TOOLS

- An incubator(APPENDIX-D) at 37°C 20%humidity
- Precision weighing
- gas stove
- A vortex mixer(APPENDIX-E)
- An autoclave(APPENDIX-F)

F. METHOD

1. Buy 125(25x25) cm^2 laminate, granite, polished chipboard, marble and Formica.
2. Sterile all countertops for 15 minutes at 394 Kelvin in autoclave.
3. Keep all sterile countertops in the same temperature, pressure, light intensity, humidity conditions. Wait them for 30 days to provide growing of some bacteria.
4. Sterile all the equipments listed in the material list(except 5 L tap water, borosilicate glass tube holder, pure water, lighter, chronometer, acetate pen and lamp) for 15 minutes at 394 Kelvin in autoclave
5. Place all the materials to the laboratory.
6. Carry the countertops that you kept in the same conditions to the laboratory.
7. Prepare TSB solution.(APPENDIX-H)
8. Prepare TGEA solution.(APPENDIX-I)
9. Prepare TGEA medium for bacterial growing. (APPENDIX-J)
10. Take sample for laminate countertop that is in the laboratory by swabbing them with the swab which is soaked in TSB solution. Put the swab into the

prepared saline solution. In this way, 1/1 bacterial solution (APPENDİX-K) is prepared.

11. Prepare 1/10 and 1/100 bacterial solutions by doing some serial dilutions.(APPENDIX-L)
12. Put 1 ml of 1/1, 1/10 and 1/100 bacterial solution samples by using 2 ml pipette in to the TGEA medium that is prepared in step 9.
13. Wait the mediums in the incubator which is set at 37°C and 20%humidity for 24 hour to sustain optimal conditions for bacterial growing.
14. After 24 hours in the incubator, take out all bacterial mediums and carry them to the laboratory.
15. Open the 40 watt lamp and hold a 1/1 bacterial medium (Petri dish) over the lamp. By this way, it is easier to see bacterial colonies and count them.
16. Count all the little dots in 1/1 bacterial medium and if it is over 300, count 1/10 bacterial medium and if it is over 300 too, count 1/100 bacterial medium.
17. Note the data.
18. Repeat steps from 10 to 17 for granite, polished chipboard, marble and Formica countertop that are mentioned in step-6 separately.
19. Repeat all steps (1to 18) for 4 more times for each type of countertops to have 5 different trial value.

Formulas:

Mean

To calculate mean of the variables:

$$mean = \frac{\sum_{k=1}^{n} trial\text{-}k}{n} = \frac{trial\text{-}1 + \cdots trial\text{-}n}{n}$$

Where;
Trial-k: Total number of bacteria in colony forming unit (cfu) for trial number
n: total number of trial
For example:

$$mean\ for\ laminate = \frac{trial\text{-}1 + \cdots trial\text{-}5}{5}$$

$$mean\ for\ laminate = \frac{78 + 72 + 84 + 70 + 82}{5} = 77.2$$

Median

To calculate median of the variables; all values of trials for each countertop must be listed from lowest to highest value. The middle of the numbers will be the medial.
For example;
Trials' values for laminate are; 78, 72, 84, 70 and 82 cfu
Median of these variables: 70-72-**78**-82-84 is 78 cfu

Standard deviation

$$s = \sqrt{\frac{1}{N-1}\sum_{i=1}^{N}(x_i - \overline{x})^2}$$ [13]

Where;
s: standard deviation of the variables
N: total trial number for each independent variable (in this experiment N is 5)
$\bar{x}$: mean of the variables
x: Total number of bacteria in cfu for trial number
i: trial number
For example;
Trials' values for laminate are; 78, 72, 84, 70 and 82 cfu
Standard deviation of these variables:

$$s\ for\ laminate = \sqrt{\frac{1}{5-1}\sum_{i=1}^{5}(trial\text{-}i - \bar{x})^2}$$

$$s\ for\ laminate = \sqrt{\frac{1}{4}(78-77.2)^2 + (72-77.2)^2 + (84-77.2)^2 + (70-77.2)^2 + (84-77.2)^2}$$

$$s\ for\ laminate = 6.6$$

[13] http://www.mathsisfun.com/data/standard-deviation.html

Standard error

$$standard\ error = \frac{s}{\sqrt{N}}$$

Where;

s: standard deviation of the variables
N: total trial number for each independent variable (in this experiment N is 5)
For example;

Trials' values for laminate are; 78, 72, 84, 70 and 82 cfu

S=6.6 and N=5

$$standard\ error\ for\ laminate = \frac{6.6}{\sqrt{5}} = 2.95$$

Variance

$$variance = \frac{1}{N-1}\sum_{i=1}^{N}(x_i - \bar{x})^2$$

Where;

N: total trial number for each independent variable (in this experiment N is 5)
$\bar{x}$: mean of the variables
x: Total number of bacteria in cfu for trial number
i: trial number
For example;

Trials' values for laminate are; 78, 72, 84, 70 and 82 cfu

Standard deviation of these variables:

$$s\ for\ laminate = \frac{1}{5-1}\sum_{i=1}^{5}(trial\text{-}i - \bar{x})^2$$

$$s\ for\ laminate = \frac{1}{4}(78-77.2)^2 + (72-77.2)^2 + (84-77.2)^2 + (70-77.2)^2 + (84-77.2)^2$$

$$s\ for\ laminate = 43.56$$

Range

$$range = T_{MAX} - T_{MIN}$$

Where;
T_{MAX}: Maximum value of the trials
T_{MIN}: Minimum value of the trials
For example;

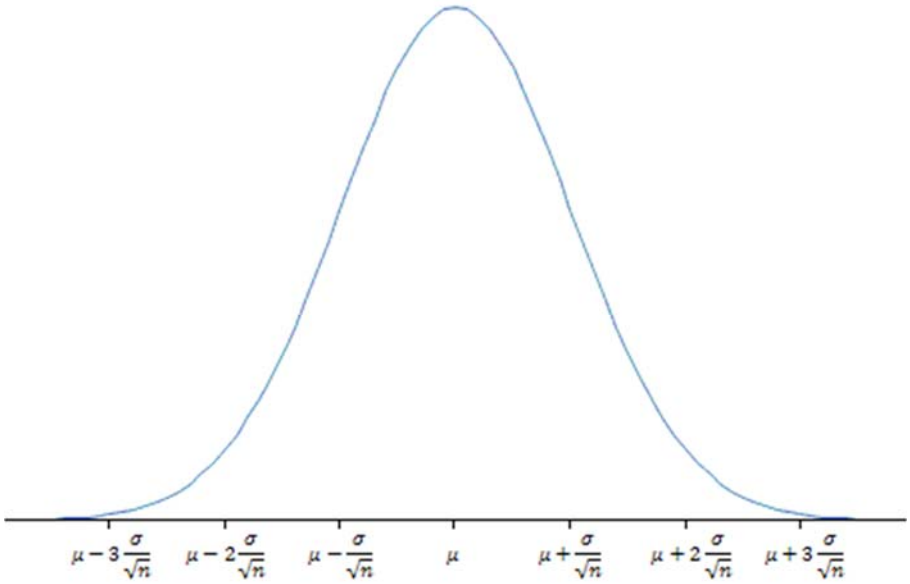
Trials' values for laminate are; 78, 72, 84, 70 and 82 cfu

$T_{MAX} = 84$ $T_{Min} = 70$

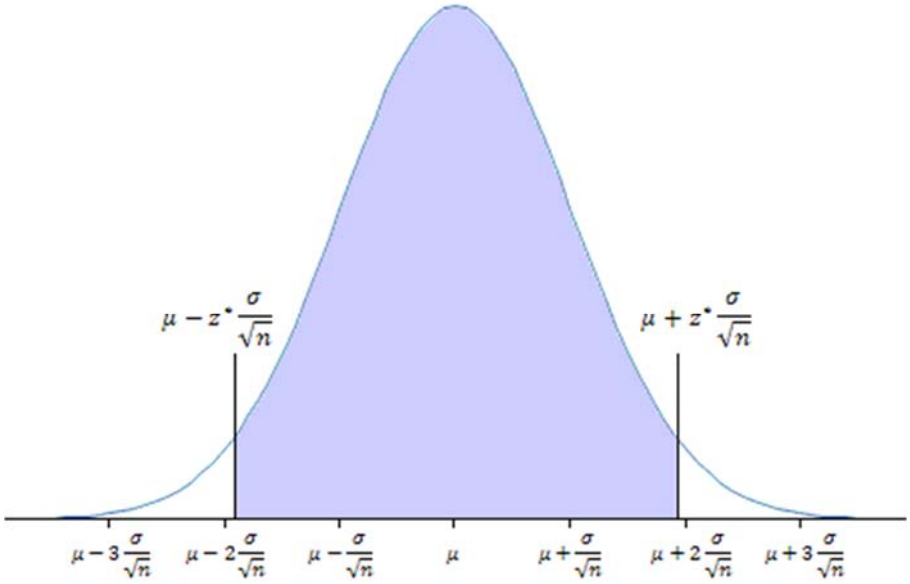
$$range\ for\ laminate = 84 - 70 = 14$$

Confidence interval

The distribution of sample means for samples of size n can be illustrated as follows:



For any given value of z^* the probability that a sample mean lies within z^* standard deviations of the mean can be calculated using ordinary left-tail probability tables. Let's call this probability C.



$$P\left(\mu - z^*\frac{\sigma}{\sqrt{n}} \leq \bar{x} \leq \mu + z^*\frac{\sigma}{\sqrt{n}}\right) = C$$

Notice, in particular, that this probability tells us something about the sample means but nothing about the population mean. Now let's consider the inequality:

$$\mu - z^* \frac{\sigma}{\sqrt{n}} \leq \bar{x} \leq \mu + z^* \frac{\sigma}{\sqrt{n}}$$

Write the resulting inequality in an alternate form:

$$\bar{x} - z^* \frac{\sigma}{\sqrt{n}} \leq \mu \leq \bar{x} + z^* \frac{\sigma}{\sqrt{n}}$$

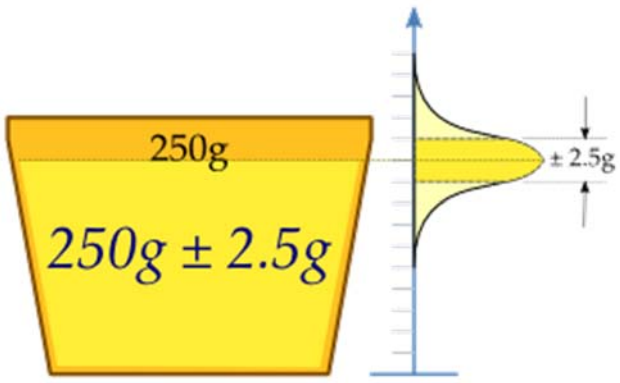
Substituting this result into our original probability, we obtain:

$$P\left(\bar{x} - z^* \frac{\sigma}{\sqrt{n}} \leq \mu \leq \bar{x} + z^* \frac{\sigma}{\sqrt{n}}\right) = C$$

In this form, C is called the confidence level and indicates how confident we are that the population mean lies within the indicated confidence interval. For example, if C = 0.95 then z* = 1.96. We say that we are 95% confident that the population mean lies within the interval:

$$\bar{x} - 1.96 \frac{\sigma}{\sqrt{n}} \leq \mu \leq \bar{x} + 1.96 \frac{\sigma}{\sqrt{n}}$$ [14]

For example;



A machine fills cups with a liquid, and is supposed to be adjusted so that the content of the cups is 250 g of liquid. As the machine cannot fill every cup with exactly 250 g, the content added to individual cups shows some variation, and is considered a random variable X. This variation is assumed to be normally distributed around the desired average of 250 g, with a standard deviation of 2.5 g. To determine if the machine is adequately calibrated, a sample of n = 25 cups of liquid are chosen at random and the cups are weighed. The resulting measured masses of liquid are X_1, ..., X_{25}, a random sample from X.

With the values in this example, the confidence interval is:

[14] http://dsearls.org/courses/M120Concepts/ClassNotes/Statistics/530G_Derivation.htm

$$0.95 = P\left(X - 1.96 \times 0.5 \leq \mu \leq X + 1.96 \times 0.5\right)$$

$$= P\left(\bar{X} - 0.98 \leq \mu \leq \bar{X} + 0.98\right).$$

This might be interpreted as: with probability 0.95 we will find a confidence interval in which we will meet the parameter μ between the stochastic endpoints

$$\bar{X} - 0.98$$

and

$$X + 0.98.$$

This does not mean that there is 0.95 probability of meeting the parameter μ in the interval obtained by using the currently computed value of the sample means,

$$(\bar{x} - 0.98, \bar{x} + 0.98).$$

Instead, every time the measurements are repeated, there will be another value for the mean X of the sample. In 95% of the cases μ will be between the endpoints calculated from this mean, but in 5% of the cases it will not be. The actual confidence interval is calculated by entering the measured masses in the formula. Our 0.95 confidence interval becomes: [15,16]

$$(\bar{x} - 0.98; \bar{x} + 0.98) = (250.2 - 0.98; 250.2 + 0.98) = (249.22; 251.18).$$

[15] Fisher, R.A.(1956) *Statistical Methods and Scientific Inference.* Oliver and Boyd, Edinburgh.

[16] Freund, J.E. (1962) *Mathematical Statistics* Prentice Hall, Englewood Cliffs, NJ.

Name of the counter top		Laminate	Granite	Polished chip board	Formica	Marble
Total number of bacteria in colony forming unit (cfu) (cfu) ± 2	Trial-1	78.0	202.0	30.0	60.0	78.0
	Trial-2	72.0	194.0	40.0	54.0	74.0
	Trial-3	84.0	202.0	36.0	60.0	82.0
	Trial-4	70.0	208.0	38.0	62.0	74.0
	Trial-5	82.0	210.0	34.0	62.0	76.0
Mean		77.2	203.2	35.6	59.6	76.8
Median		78	202	36	60	78
Range		14	16	10	8	10
Variance		43.56	39.20	14.80	10.79	17.19
Standard deviation		6.6	6.3	3.8	3.3	4.1
Standard error		2.95	2.80	1.72	1.47	1.85
95% confidence interval		5.79	5.52	3.33	2.89	3.59

Table-3: the raw data table of changes of mean, median, standard deviation, standard error, variance, 95% confidence interval and range value of total number of different types of bacteria in colony forming unit due to different kinds of countertops.

	laminate	granite	Chip board	formica	marble	Total
Number (n)	5	5	5	5	5	25
∑ x	386	1016	178	298	384	2262
Mean	77.2	203.2	35.6	59.6	76.8	90.48
∑ x^2						
Variance	37.20	39.20	14.80	10.80	11.20	
Std. Dev.	6.099	6.261	3.847	3.286	3.347	
Std. Err.	2.728	2.800	1.720	1.470	1.497	

ANOVA Result :

	SS	df	MS	F	P
Between	85173.4399	4	21293.3600	940.5194	<0.0001
Within	452.799999	20	22.6400		
Total	85626.2399	24			

Table-4: descriptive statistics and ANOVA results of total number of different types of bacteria in colony forming unit due to different kinds of countertops.

H. CONCLUSION AND EVALUATION

In this experiment my aim was to find an answer to my research question: "**How do five different types of countertops: polished chipboard, granite, marble, formica and laminate affect the total number of different types of bacteria on these surfaces which is indicated by viable cell count (colony counts) method?"** To find an answer for this question I did so many researches and experiments. To do my experiments: I had to choose which types of countertops to examine. I chose laminate, granite, polished chip board, marble and formica since they are commonly used as a countertop.

Before I started my experiments, I had to stabilize variables for countertops because total number of bacteria in the surfaces depends on so many different variables such as surface area of countertops, humidity of the room, temperature of the room...etc. So to control these kinds of variables were extremely important for my experiment. Therefore I put them in a carton box homogenously. I kept them away from the light source by putting into the box in a dark room because I could not able to stabilize the light source all the time. I kept the box in that room for 30 days.

In this experiment, another essential thing was choosing an appropriate method for my experiment. After my researches; I chose viable cell count method (colony counts) to find the total number of different types of bacteria because it was a sensitive method, can distinguish between living and non-living cells and it can be carrying out in Pharmaceutical Microbiology laboratory easily. After one month; I performed my experiment in the laboratory of the department of Pharmaceutical Microbiology in the faculty of pharmacy in Ankara University with the help of Dr. Bahar Bozkılınç.

In the graph-1 at page 20 can be seen the mean value of total number of different types of bacteria. Due to the graph; the highest value of total number of different types of bacteria was found on granite because it has more pits than the others due to its nature. The lowest value of total number of different types of bacteria was found on polished chip board since polishing material fills the pits of the surface and reduces the number of pits. Therefore bacteria cannot find suitable places to reproduce and protect from the external factors. These results also support my hypothesis: **"The total number of different types of bacteria over the granite countertop will be the most and the total number of different types of bacteria on the polished chipboard countertop will be the least."** Also the study, which is maintained by Ph.D. Pete Snyder in Hospitality Institute of Technology and Management, "The reduction of *E.coli* on various countertop surfaces" supports my results. In this experiment the reduction of *E.coli* on granite and laminate surfaces was examined when they are washed and rinsed or exposed vinegar. When the vinegar was applied the overall reduction of bacterial counts was about 500,000 to 1 for laminate and about 80,000,000 to 1 for granite.[17] The literature value of this experiment shows that granite surface has extreme amount of bacteria when it is compared to other surfaces. Since; granite has the highest total number of different types of bacteria in my experiment; literature supports my experiment's results.

At table-4; there is ANOVA results of my experiment. Before we interpret P value of my experiment, we should consider what P value is. When P value is very small (smaller than 0.5), it suggests that the results of samples that is observed cannot

[17] http://www.mbstone.com/HH_promo/articles/Bacteria_In_Granite_new.htm

caused by random sampling.[18] Since P value in this experiment is very small, my hypothesis is proven to be true.

The Error and Uncertainties

In this experiment; there were some mistakes due to equipments that were used and environment where the experiment has accomplished. So we had to calculate some error values for each experiment. I calculated standard deviation and standard error value for each type of countertop. Before we discussed result of these values, we have to know what these values represent. Standard deviation is a measure of the spread of scores within a set of data[19] and standard error is a statistical term that measures the accuracy with which a sample represents a population.[20] Standard deviation for laminate is 6.6, for granite 6.3, for polished chip board 3.8, for formica 3.3 and for marble 4.1. According to this; obtained data of formica and polished chipboard is close to their mean value while laminate and granite has wider data range. So results value for formica and polished chipboard is more reliable than the other results. The reason for this phenomenon could be: I examined these two surfaces before the other surfaces so my attention could be higher at the beginning and this may cause less random error for these countertops. Standard error value for laminate is 2.95, for granite2.80, for polished chip board1.72, for formica 1.47 and for marble1.85. Since formica and polished chipboard has lower standard error, their accuracy is higher.

As a result of this experiment; the mean total number of different types of bacteria for granite is 203.2, for laminate is 77.2, for marble is 76.8, for formica is 59.8 and for chip board is 35.6 as can be seen at table 3 and 4. I also calculated 95%

[18] http://www.graphpad.com/guides/prism/6/statistics/index.htm?f_ratio_and_anova_table_(one-way_anova).htm
[19] https://statistics.laerd.com/statistical-guides/measures-of-spread-standard-deviation.php
[20] http://www.investopedia.com/terms/s/standard-error.asp

confidence interval for each type of countertop too as can be seen at table-3. Confidence interval can be used to determine the reliability of a calculation. Confidence interval for these values are 5.79 for laminate, 5.52 for granite, 3.33 for chip board, 2.89 for formica and 3.59 for marble. In this experiment, all 95% confident interval values are low. This shows that the results of this experiment are accurate and reliable.

The errors in this experiment were decreased by stabilizing environmental factors and controlled variables such as:

- ✓ The countertops' surface area remained constant for each trial. By this way bacteria had the equal area for reproduction in each trial.
- ✓ Sterilization process is done and after this process each countertop is waited for 30 days. Consequently; the time that is given were constant for each trial and results were affected by bacteria which were placed before the experiment.
- ✓ The humidity, pressure and temperature of the room that the sterilized countertops kept for 30 days were kept constant. So these factors didn't affect the reproduction pattern of the bacteria.
- ✓ The volume of the TGEA solution in the Petri dish medium that sustain nutrition for bacteria were remained constant. In each trial; bacteria has the same amount of nutrient source.
- ✓ Volume of the bacterial solution that is put in to the TGEA medium was constant for each trial. Therefore; total number of different types of bacteria wasn't affected volume.
- ✓ The time that the bacterial samples in TGEA medium spent in the incubator was remained constant for each trial.

- ✓ Also humidity and temperature of the incubator were constant.

Even though so many external factors remained constant in this experiment, there are still some errors that should be fixed in the next experiments. Some of them are random and some of them are systematic but we have to fix most of the errors to reach the most accurate value. I listed below limitations of this experiment that can be the cause of these errors and suggestions to fix them.

Limitations and Suggestions

- ❖ Air may contain some bacteria as well as surfaces and when liquid TGEA or bacterial solution was pouring in to the Petri dish, some of these bacteria that are in the air may fall into the Petri dish. Since fire kills bacteria; these processes could be done near the Bunsen burner.
- ❖ In this experiment I waited only 24 hours to count bacterial colony number but some bacteria may need more than 24 hours to create a colony. So I could dismiss some of the bacteria. To correct this mistake; counting procedure can be done in different time intervals. For example; we can count colonies after 12^{th}, 18^{th}, 24^{th}, 30^{th}, 48^{th} and 72^{nd} hours.
- ❖ In this experiment; only five types of countertops were examined and since the results differ from the countertop to countertop, we do not have enough data to make a comparison. In further experiments; there should be more types of countertop like stainless steel, wood, soapstone, butcher block or paper composite.
- ❖ In this experiment; only one type of cell counting method was used. (Viable cell count method) and this can be a limiting factor to calculate bacteria number. In further experiments; different types of counting methods like direct microscopic count method should be used too.

- In this experiment; only one type of medium (TGEA-appendix C) was used as a nutrient source and this may affect the results because some types of bacteria may need different nutrient source. So in further experiments; different types of nutrient mediums like buffered charcoal yeast extract agar should be used.

After observing the results of the research question: "**How do five different types of countertops: polished chipboard, granite, marble, formica and laminate affect the total number of different types of bacteria on these surfaces which is indicated by viable cell count (colony counts) method?"** new research question comes to mind to find out the most hygienic environment in the kitchen: "**How does the total number of different types of bacteria, which is indicated by viable cell count (colony counts) method, changes on polished chipboard countertop due to different types of environment such as moist, cold, hot, dry and compressive room?"**

Countertops can be found so many places at our home like bathrooms or kitchen. So its hygiene is an essential for our health. However; some bacteria including pathogenic bacteria can locate on our countertop. Since we prepare our foods in kitchen, kitchen countertop's hygiene is particularly important. This research demonstrates; granite countertops have the highest number of bacteria and polished chipboard surface has the least. Therefore using polished surfaces as our kitchen countertop can decrease the risk of bacterial infections and provide more hygienic environment for people.

I. APPENDICES

APPENDIX-A

Saline: solving 4 g NaCl in 100 ml pure water

APPENDIX-B

TSB: In clinical microbiology, it is used for the suspension, enrichment and cultivation of strains isolated on other media.[21]

Figure-1: TSB material that is used in this experiment.

APPENDIX-C

Tryptone Glucose Extract Agar is used for cultivating and enumerating microorganisms in water[22]

Figure-2: TGEA material that is used in this experiment.

APPENDIX-D

An **incubator** is a machine that allows us to control environmental conditions such as humidity, temperature…etc. In this experiment; it is set at 37°C 20%humidity to maintain the conditions in human body.

Figure-3: incubator machine that is used in this experiment.

[21] http://www.bd.com/europe/regulatory/Assets/IFU/HB/CE/BA/BA-257107.pdf
[22] http://www.bd.com/europe/regulatory/Assets/IFU/Difco_BBL/223000.pdf

APPENDIX-E

- A vortex mixer is a machine that is used to mix sample liquids or solutions which is found in borosilicate tube.

Figure-4: vortex mixer that is used in this experiment

APPENDIX-F

- An autoclave: is a device used to sterilize equipment and supplies by subjecting them to high pressure saturated steam at 121 °C, 1 atm for around 15–20 minutes[23]

Figure-5: autoclave that is used in this experiment

APPENDIX-G

CFU is used to determine the number of viable bacterial cells in a sample per mL. Hence, it tells the degree of contamination
in samples of water, vegetables, soil or fruits, or the magnitude of the infection in humans and animals.[24]

[23] http://en.wikipedia.org/wiki/Autoclave
[24] http://www.biology-online.org/dictionary/Colony-forming_unit

APPENDIX-H and APPENDIX-I

To prepare TSB and TGEA solution:

1. Put 3 g TSB it in to an empty Erlenmeyer flask. Then put 100 ml pure water in to the same Erlenmeyer by using graduated cylinder.
2. Put 4.8 g TGEA into an empty Erlenmeyer flask. Then put 200 ml pure water in to the same Erlenmeyer by using graduated cylinder.
3. Place 5 L tap water in to the iron bowl.
4. Open the gas stove, light it with the lighter, and place the iron bowl.
5. Place 2 Erlenmeyer flasks, those have TGEA and TSB solutions, in to the iron bowl.
6. 40 minutes later; take the iron bowl over the gas stove and put it to the table.

Figure-6: TSB and TGEA solutions that is used in this experiment

APPENDIX-J

To prepare TGEA medium:

Open an Erlenmeyer flask which has TGEA solution in it and graduate 13.6 ml TGEA solution by using a glass graduated cylinder. Take one of the empty sterile Petri dishes which are put to the laboratory at step 4, open it and put 13.6 ml TGEA solution then close it immediately and put it back. Repeat this process for other 14 empty Petri dishes which are put to the laboratory before.

Figure-7: TGEA mediums

APPENDİX-K

To prepare 1/1 bacterial solution:

1. Open Erlenmeyer flask which has TSB solution in.
2. Take a sterile swab and put it in to TSB solution.
3. Then take a sample countertop and rub it with that swab homogonously.
4. Take the borosilicate glass tube that has 1 ml saline, open it, put the swab and close the tube.
5. Then put the tube to the vortex, set it for one minute in medium and start the vortex.

Then open the tube, throw out the swab and close it again.

APPENDIX-L

Serial dilution is used to decrease number of bacteria in a solution. When 1ml of the sample specimen is mixed with 9 ml of saline it becomes 1/10 diluted bacterial solution. If we take one ml from 1/10 diluted solution and add it to 9 ml saline then becomes 1/100 diluted bacterial solution.

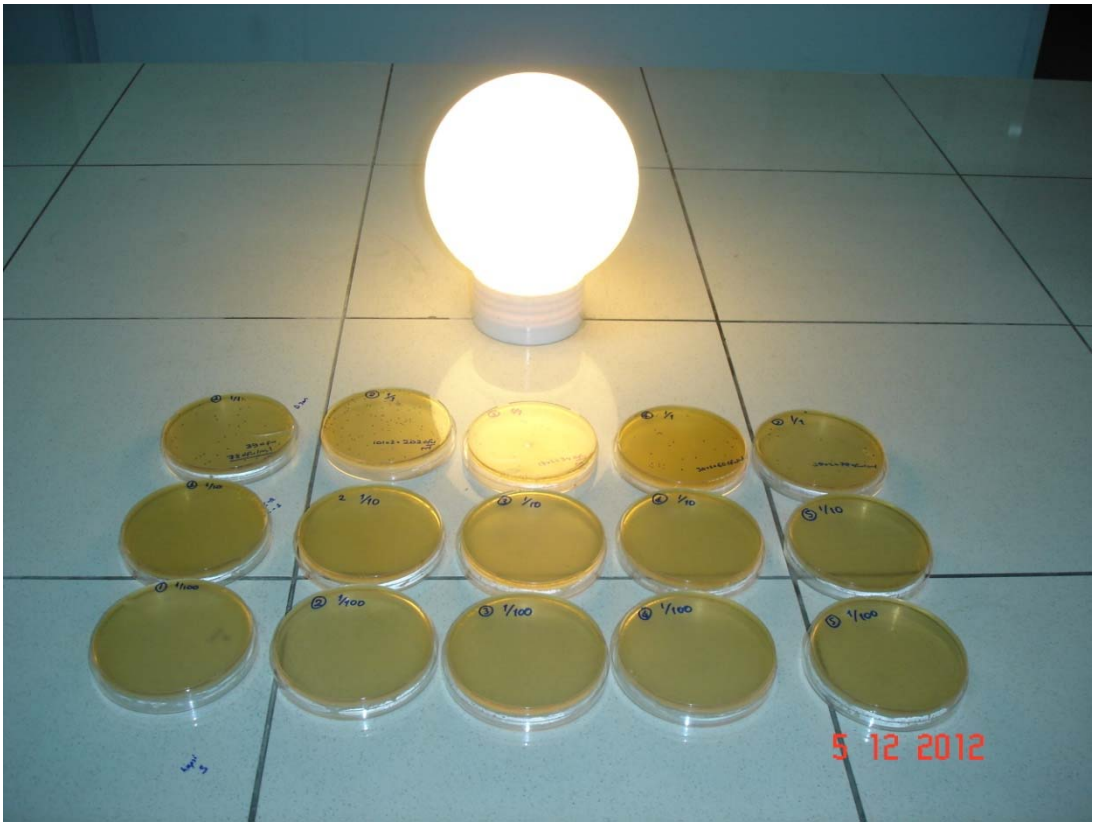


Figure-8: 1/1, 1/10 and 1/100 diluted bacterial samples of this experiment.

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