

TED ANKARA COLLEGE FOUNDATION HIGH SCHOOL

Investigating a possible relationship between
adolescent acnes and carrying *Staphylococcus
aureus* on skins

BIOLOGY EXTENDED ESSAY

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ABSTRACT

It is estimated that 80 percent of all people between the ages of 11 and 30 have acne outbreaks. While especially teenagers are very concerned with the problem, the reasons for it can be further investigated. Very little research has been done on possible bacterial interference on skin between *Propionibacterium acnes*, which causes acne and *Staphylococcus aureus*, an opportunistic bacteria that lives on skin. Therefore, the objective of this extended essay is to investigate a possible relationship between acnes and carrying *Staphylococcus aureus* on skins of teenagers. This essay examines to what extent carrying *Staphylococcus aureus* on skin affects the number and distribution of acnes and pimples.

The research question being investigated was: “Is there any positive correlation between having *Staphylococcus aureus* on skin of adolescent people and formation of acne outbreaks on their skin?” It was hypothesized that; the number and dispersal of adolescent acnes on skin doesn’t change the probability to carry *Staphylococcus aureus* on skin.

In order to test the hypothesis and to answer the research question, volunteer students were grouped into 4 categories considering their gender, age and whether they have acnes on skin or not. Bacterial samples were collected from the skin flora of these students, *Staphylococcus aureus* population was cultured on blood agar plate and left for incubation of 24 hours in laboratory conditions.

Resultantly, five out of twenty students were carriers of *Staphylococcus aureus* on skin. Four of the carriers were selected as students without acne on skin, whereas only one belonged to the group that represented students with acnes. Also, four of them were male where one was a female. The results and statistical analysis revealed by the chi-square test showed that there was no correlation between having acnes on skin and carrying *Staphylococcus aureus* on skin.

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INTRODUCTION

The first time I came across with the topic that this extended essay is going to focus on was after I had a foodborne illness. The main reason for the foodborne disease I caught couldn't be designated, thus I felt obliged to determine the source to avoid possible foodborne illness in the future both for me and for my family. However, I was told that the source can't be surely defined besides, the contamination could have been occurred and spread from anywhere. Even there is a type of bacteria which lives in the skins of warm-blooded animals including humans can cause foodborne illness, which shows that sources for it is very prevalent in our environment.

The response given to me was not satisfactory, however the fact that a bacteria which can live on our skins and can lead to foodborne illness caught my attention, since I was surprised how easily a pathogen can affect our health. Highly interested on the topic, I began researching the issue. The very first thing that my research focused on was to determine the types of bacteria that caused foodborne illness and the enterotoxins they produce. Most abundant species that cause food infection is *Salmonella*, *Campylobacter jejuni*, *Eschericia coli*, *Listeria monocytogenes*. In addition, some foodborne illnesses are caused by enterotoxins rather than by direct bacterial infection. Staphylococcal enterotoxins, especially Staphylococcal Enterotoxin A and Staphylococcal Enterotoxin B are the most commonly reported enterotoxins which are produced from *Staphylococcus aureus*. Throughout my researches, *Staphylococcus aureus* was determined as the bacteria which lives on human's skin which is mentioned previously. After an extended process of researching the bacteria *Staphylococcus aureus*, I have found out that this type of bacteria also can lead to a huge variety of diseases and infections, including pimples and acnes on skin, which is a common problem of people on my age which is seventeen. The fact that *Staphylococcus aureus* can cause pimples on skin, influenced me to further inquire whether the adolescent acnes on skins of teenagers, that are seventeen years old, are formed as a result of activities of *Staphylococcus aureus* and its enterotoxins, or not.

Staphylococcus aureus is a bacteria of the order Bacillales, the family Staphylococcaceae and the genus *Staphylococcus*. It is an ideal species for the experiment since it is rapid growing which makes the experiment to be carried out in a short time period. In addition, they are very abundant in the environment, such as skins of people, which makes it easier to congregate for the experiment. Also, they can be easily seen and analyzed on blood agar plates.

The name *Staphylococcus* is derived from the Greek word staphyle or "bunch of grapes" because of the characteristic cluster-like appearance of the bacteria under the microscope. *Staphylococcus aureus* is a bacterium that is a member of the phylum Firmicutes, and is frequently found in the human nasal passages and on the skin but can also be found in the environment and in contaminated food. *Staphylococcus aureus* are gram-positive, catalase

positive cocci. They are approximately 0.5-1.5 μm in diameter, nonmotile, non-spore-forming, facultative anaerobes with the exception of *S. aureus anaerobius* that usually form in clusters. *S. aureus* can cause a range of sicknesses, from skin infections, such as pimples, impetigo, boils, cellulitis folliculitis, blisters, scalded skin syndrome, and abscesses, to life-threatening diseases such as pneumonia, meningitis, osteomyelitis, bacteremia, and sepsis. Furthermore, *S. aureus* causes foodborne illness by growing in temperatureabused food and producing a heat stable toxin. *S. aureus* are very abundant in the environment and can contaminate food by direct bodily contact, through skin fragments, or through respiratory droplets produced when people cough and sneeze. Its symptoms are nausea, vomiting, abdominal pain, cramps, and diarrhea. There are 32 species of staphylococci, but only 17 are indigenous to humans.

The presence of *S. aureus* does not always indicate infection. Depending on the strain, *S. aureus* is capable of secreting several exotoxins, which is the reason for the symptoms mentioned above. To date, 14 different Staphylococcal Enterotoxin types have been identified, which share structure and sequence similarities. They are highly stable, resist most proteolytic enzymes, such as pepsin or trypsin, and thus keep their activity in the digestive tract after ingestion although the superantigen Staphylococcal Enterotoxin is a protein. *Staphylococcal enterotoxin B (SEB)* is the most abundant enterotoxin which is a common cause of food poisoning. Enterotoxins can produce illness even when the microbes that produced them have been killed.

It is also worth noting that, *Staphylococcus aureus* can pose a great threat to human health as the article of World Health Organization (WHO) shows¹, case fatality rates in some *S. aureus* infections today still can reach 30%.

Consequently this paper will focus on the research question: “Is there any positive correlation between having *Staphylococcus aureus* on skin of adolescent people and formation of acne outbreaks on their skin?”, and designates how the experiment was planned and carried out. Also, discuss the results obtained and last but not least derive possible consequences.

¹ http://www.who.int/vaccine_research/diseases/soa_bacterial/en/index2.html

HYPOTHESIS

Staphylococcus aureus (Staph) is a common bacterium found on the skin and in the noses of up to 25% of healthy people and animals. They are very prevalent in the environment. According to research carried out in Ankara, Turkey from June 2006 – 2007 shows², among patients and hospital staff in a tertiary referral center of 438 people, 106 (24.2%) were nasal carriers of *S.aureus*. There was no statistically significant difference between the hospital staff, in-patients and out-patients regarding prevalence of nasal *S. aureus* carriers ($p>0.05$). There was also no significant difference between the genders regarding nasal *S. aureus* carriage ($p>0.05$). Although this research focuses on the existence of *Staphylococcus aureus* in nasal passages and do not consider skin types of people, it still provides a good knowledge to the probability of a people to carry *Staphylococcus aureus* on skin.

The aim of this essay focuses on is a possible relationship between acnes on skin and presence of *Staphylococcus aureus*. Therefore, the major reasons for pimples and adolescent acnes were also considered. *Propionibacterium acnes* is a gram-positive human skin commensal that prefers anaerobic growth conditions and several studies have indicated that specific strains of *P. acnes* bacteria are associated with acne vulgaris. Although it is known that, damage caused by *P. acnes* and the associated inflammation make the affected tissue more susceptible to colonization by opportunistic bacteria, such as *Staphylococcus aureus*, whether this is a root causality, just opportunistic and a side effect, or a more complex pathological duality between *P. acnes* and this particular Staphylococcus species is not definitely known. However, another research article suggests that³ fermentation of glycerol with *Propionibacterium acnes* (*P. acnes*), a skin commensal bacterium, can function as a skin probiotic for in vitro and in vivo growth suppression of USA300, the most prevalent community-acquired methicillin-resistant *Staphylococcus aureus* (CA-MRSA).

With regarding the fact that there has never been a proven bacterial interference between *Propionibacterium acnes* and *Staphylococcus aureus* that increases their activity, it can be hypothesized that carrying *Staphylococcus aureus* on skin doesn't increase the number and dispersal of adolescent acnes on skin. It is expected that after experimental groups are created as the ones that have pimples on skin and the ones that don't have any, both of the groups will show equal or very close possibility levels.

² African Journal of Microbiology Research Vol. 5(13), pp. 1615-1618, 4 July, 2011

³ Fermentation of *Propionibacterium acnes*, a Commensal Bacterium in the Human Skin Microbiome, as Skin Probiotics against Methicillin-Resistant *Staphylococcus aureus*, Plos One, journal 0055380, Feb 06, 2013

METHOD DEVELOPMENT AND PLANNING

Designing a scientific method for the experiment is essential to be able to conduct the experiment successfully and to reach results accurately. Before carrying out the experiment, the species of the bacteria that is appropriate for this experiment and the factors that affect the reproduction rate of the bacteria should be determined. With considering all these aspects, the most suitable method should be decided on.

However, designing an appropriate method that would support or reject the previously mentioned hypothesis, brought several problems and limitation with it. One of them was with collecting bacteria into a sterilized place and fixing them to this sterilized place until they reproduce. After further research, the problem was overcome when I decided to use sterilized swabs and stuart transport medium. I used sterilized swabs for collecting bacteria from people's skins with the help of their cotton tip. However, collected samples should have been protected from environmental factors, such as exposure to sunlight or changes in temperature, until they are delivered to the laboratory for incubation. Thus, I used stuart transport medium which is used for the transport of bacteriological specimens, including *Staphylococcus aureus*. This special medium was perfectly suitable not only it maintains the organism's viability without significant multiplication, it also prevents microbial proliferation. In addition, with the help of its ingredients, it retards oxidation and maintains osmotic equilibrium in the medium.

Another problem was with finding the most suitable medium for implanting the collected *Staphylococcus aureus*. While searching for the right medium, I have considered the pH values, organic and inorganic growth factors of various of growth mediums. Also, how the collected samples are seen on that certain growth medium was important. After research, blood agar plates were suitable for *Staphylococcus aureus* since it is a differential media which allows the detection of hemolysis by cytolytic toxins secreted by some bacteria including *Staphylococcus*, and distinguish one microorganism type from another growing on the same media. Furthermore, *Staphylococcus aureus* can grow at pH levels of 4.2 to 9.3 but grows best at pH levels of 7-7.5. Blood agar plates' pH value is adjusted to 7.3 which is another factor that supports *Staphylococcus aureus* growth in the medium.

My aim with the experiment was to find a possible relationship with acnes and presence of *Staphylococcus aureus* on skin. Thus, after my friends on 11-U, who were volunteer to be a part of my experiment, were selected I rubbed the sterilized swab to the person's nose and right and left upper cheeks since these areas are the most oily parts of the skin and hence most suitable areas for pimples. Also, they were told not to wear any make up on their face, such as but not limited to, foundation, powder, sunscreen or any kind of cream. Furthermore, they were also told that they shouldn't wash their face just before the experiment was carried out. Necessarily these precautions were taken to prevent any chemical factor to affect the presence of *Staphylococcus aureus* on a person's face.

It was essential to make sure that all variables were being controlled. Environmental factors of temperature and light exposure were the most apparent ones. Besides, time was another important factor while conducting the experiment, especially when the swab was rubbed against the person's face and also when the collected samples were left for incubation. Thus, these factors were dealt accordingly to gather accurate data. It was decided to collect samples in a room where there was no window to keep temperature constant as much as possible at 26 °C, which also the light to be also controlled while collecting process. Also, while collected samples were transported to my house for the implementation of the bacteria to blood agar plates and then delivery of these plates to the laboratory for incubation, expandable polystyrene boxes were used to prevent light exposure to the collected and implanted samples and also to keep temperature constant. Although it is impossible to take equal amounts of bacteria from each and every people's face, we used the chronometer to keep the time for rubbing a person's face constant to take sample amounts close to each other. Also, in the laboratory the blood agar plates were left for incubation for a constant time period of 24 hours.

Staphylococcus aureus can live in a temperature range of 6-48 °C. However, 37 °C is the optimum temperature for this species to reproduce. For this reason the blood agar plates were left for incubation at 37°C for 24 hours. However, after 24 hours some samples didn't show the necessary growth, thus the blood agar plates that the expected growth was not reached, left for incubation for an additional 24 hours.

The experiment was done in Reference Laboratory of Republic of Turkey Ministry of Food, Agriculture and Livestock, with the help of Dr. Ahmet Koluman and Dr. Nazan Akçelik. They helped me while conducting the experiment like incubation of blood agar plates. Moreover, they supported me with a rich supply of material, such as but not limited to blood agar plates, expandable polystyrene boxes and incubator.

While carrying out the experiment, samples from 21 people were collected and the experiment was based on these trials. However, one of the blood agar plate dried out, thus it was not considered while data analysis and evaluation process.

METHOD

Materials:

- 20 blood agar plates
- 30 sterilized swabs
- 30 stuart transport medium
- incubator
- 2 expandable polystyrene boxes (EPS)
- 2 cold gel stipes
- chronometer
- thermometer

- 2 pair of gloves
- labels
- fridge

Method:

- 1) In a class atmosphere (11-U), 20 students were selected that were volunteer to be a part of the experiment, with considering their year of birth, gender and their skin type, whether has adolescent acnes on face or there are no acnes, and they were labelled from A to V.
- 2) A sterilized swab was taken out of its protective plastic bag and it was rubbed against the person's face, that has been selected, to the areas of nose and left and right upper cheeks for one minute, with ensuring that the swab's cotton tip and its long stick doesn't touch anywhere.
- 3) The swab was put into one of the stuart transport medium. (see appendix 2)
- 4) The tube was labelled with a letter that represented the person.
- 5) Steps two to four were repeated for nineteen other selected students under the same room conditions. (temperature, light intensity, pressure, humidity, etc.)
- 6) All the swabs inside stuart transport mediums were taken and they were put inside an expandable polysterene box to avoid any light exposure.
- 7) Meanwhile, blood agar plates were waited inside a fridge that had a temperature of 6 °C for 24 hours.
- 8) After waiting for sixteen hours, stuart transport mediums were taken out of the expandable polysterene box and one of the swabs was taken out of its protective tube.
- 9) The lid of a blood agar plate was taken up and the cotton tip of the swab was rubbed on the surface of the agar plate.
- 10) The plate was reversed and put on the lid.
- 11) The lid was labelled with the letter that represented the person.
- 12) Steps eight to ten were repeated for other nineteen swabs under same room conditions.
- 13) The agar plates and cold gel stupes that waited also in fridge at 6 °C for 24 hours, were put into an expandable polystrene box and were taken to Reference Laboratory of Republic of Turkey Ministry of Food, Agriculture and Livestock.
- 14) In this laboratory, the blood agar plates were left for incubation under 37°C for 24 hours.
- 15) The blood agar plates were taken out and the ones that *Staphylococcus aureus* have reproduced were determined.
- 16) The blood agar plates were scanned.
- 17) A survey composed of 5 yes/no questions was carried with the volunteer students who took part in the experiment. (see appendix 1)

DIAGRAMS



Figure 1. Samples are collected from the volunteer students' noses and right and left upper cheeks with the help of a swab and this swab is put into a stuart transport medium.



Figure 2. Blood agar plates before plantation of bacteria *Staphylococcus aureus* on it



Figure 3. The cotton tip of the swab is rubbed on the surface of the blood agar plate.



Figure 4. Labeled blood agar plates are put into an expandable polystyrene box.

RESULTS

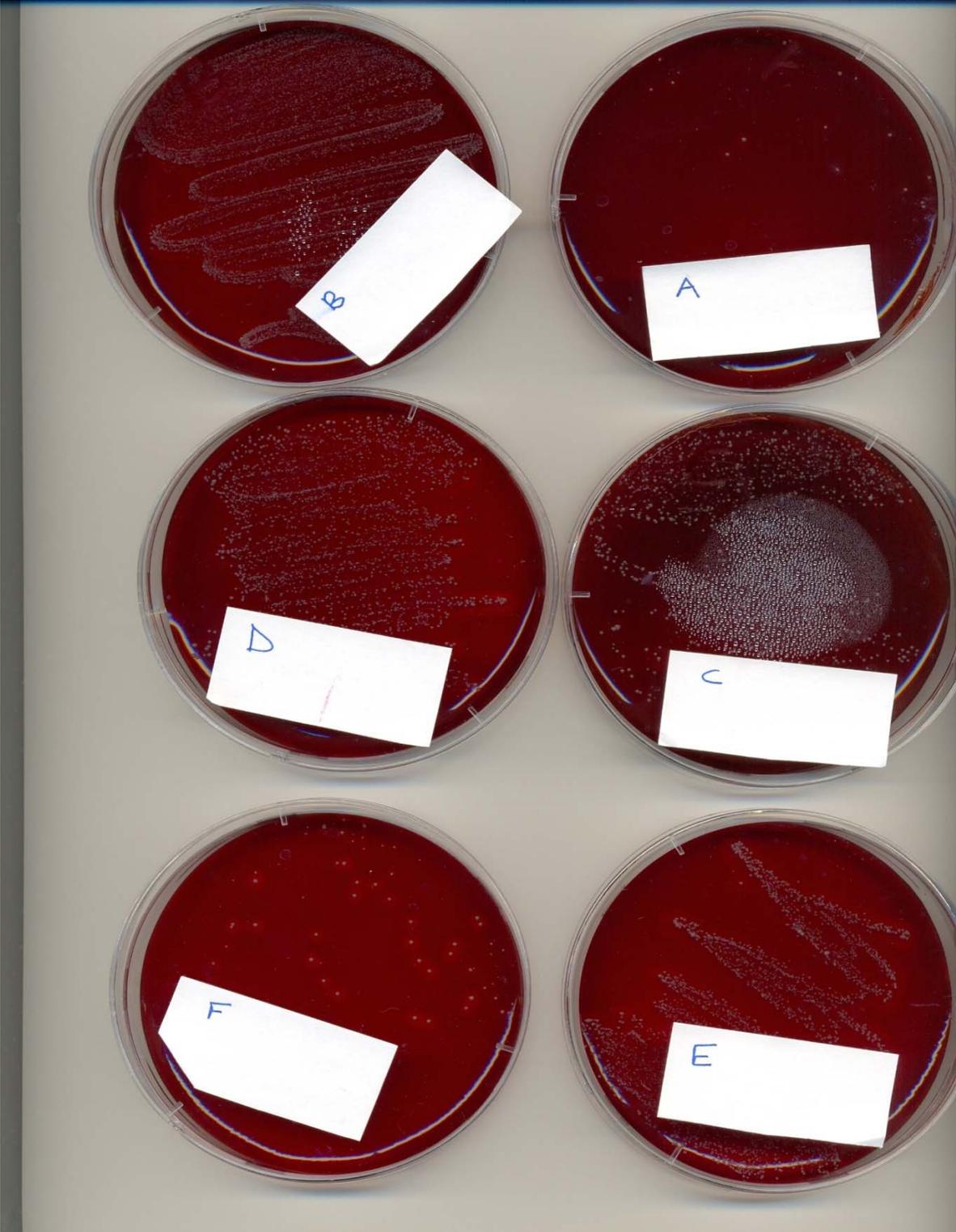




Figure 5. Blood agar plates from A to F after 24 hours of incubation

Figure 6. Blood agar plates from G to L after 24 hours of incubation



Figure 7. Blood agar plates from M to R after 24 hours of incubation

Group number	Letter it is represented by	Age	Gender	Presence of pimples on skin	Presence of <i>Staphylococcus aureus</i> on skin
1	A	17	Male	-	+
	B	17	Male	-	-
	C	17	Male	-	-
	E	17	Male	-	+
	U	17	Male	-	+
2	F	17	Male	+	+
	G	17	Male	+	-
	H	17	Male	+	-
	I	17	Male	+	-
	J	17	Male	+	-
3	K	17	Female	-	-
	L	17	Female	-	-
	M	17	Female	-	-
	N	17	Female	-	+
	O	17	Female	-	-
4	Q	17	Female	+	-
	R	17	Female	+	-
	S	17	Female	+	-
	T	17	Female	+	-
	D	17	Female	+	-

Table 1: Raw data table showing the relationship between presence of *Staphylococcus aureus* on skin and presence of pimples on skin considering the volunteer students' genders and age

Control group number 1 represents male students without acnes or pimples on skin

Control group number 2 represents male students who have acnes and pimples on skin

Control group number 3 represents female students without acnes or pimples on skin

Control group number 4 represents female students with acnes and pimples on skin

	Male	Female
Has pimples	%20	0
No pimples	%60	%20

Table 2: Table showing the varying probability to carry *Staphylococcus aureus* on skin with considering gender and skin type of the person

Group Number	Letter presented with	Do you wash your face everyday?	Did you get an acne this week?	Do you use cosmetic products like foundation or sunscreen on daily basis?	Do you go to a dermatologist regularly?	Do you pop your acnes?
1	A	Yes	No	No	No	No
	B	Yes	No	No	No	No
	C	Yes	No	No	No	No
	E	Yes	Yes	No	No	Yes
	U	Yes	No	No	No	No
2	F	Yes	Yes	No	No	Yes
	G	Yes	Yes	No	No	Yes
	H	Yes	Yes	No	No	Yes
	I	Yes	Yes	No	No	No
	J	Yes	Yes	Yes	No	No
3	K	Yes	No	No	No	No
	L	Yes	No	No	No	No
	M	Yes	No	No	No	No
	N	Yes	No	Yes	No	Yes
	O	Yes	No	No	No	No
4	Q	Yes	Yes	Yes	Yes	No
	R	Yes	Yes	Yes	No	Yes
	S	Yes	Yes	Yes	No	Yes
	T	Yes	Yes	Yes	Yes	Yes
	D	Yes	Yes	No	No	No

Table 3: Raw data table showing the results obtained from the survey with the students that took part in the experiment

	Answer is YES	Answer is NO
Question no.1	%100	0
Question no.2	%50	%50
Question no.3	%30	%70
Question no.4	%10	%90
Question no.5	%40	%60

Table 4: Table showing the percentages of answers “yes” and “no” to the questions of the survey with the students that took part in the experiment

	Answer is YES	Answer is NO
Question no.1	%100	0
Question no.2	%20	%80
Question no.3	%20	%80
Question no.4	0	%100
Question no.5	%60	%40

Table 5: Table showing the percentages of answers “yes” and “no” to the questions of the survey of the students who carry *Staphylococcus aureus* on skin

DATA ANALYSIS

Statistical Analysis 1: A crosstabulation showing the overall results of presence of pimples on skin and *Staphylococcus aureus* on skin regardless the genders

		Presence of <i>Staphylococcus aureus</i> on skin		Total
		None	Present	
Presence of pimples on skin	None	6	4	10
	Present	9	1	10
Total		15	5	20

Statistical Analysis 2: Chi-Square Tests examining a possible positive correlation between having acnes on skin and *Staphylococcus aureus* regardless the genders

	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	2,400(b)	1	,121		
Continuity Correction(a)	1,067	1	,302		
Likelihood Ratio	2,532	1	,112		
Fisher's Exact Test				,303	,152
Linear-by-Linear Association	2,280	1	,131		
N of Valid Cases	20				

a Computed only for a 2x2 table

b 2 cells (50,0%) have expected count less than 5. The minimum expected count is 2,50.

Statistical Analysis 3: A crosstabulation showing the overall results of presence of pimples on skin and *Staphylococcus aureus* on skin with taking into consideration the genders

Gender		Presence of <i>Staphylococcus aureus</i> on skin		Total	
		None	Present		
Male	Presence of pimples on skin	None	2	3	5
		Present	4	1	5
	Total		6	4	10
Female	Presence of pimples on skin	None	4	1	5
		Present	5	0	5
	Total		9	1	10

Statistical Analysis 4: Chi-Square Tests examining a possible positive correlation between having acnes on skin and *Staphylococcus aureus* with taking into consideration the genders

Gender		Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Male	Pearson Chi-Square	1,667(b)	1	,197		
	Continuity Correction(a)	,417	1	,519		
	Likelihood Ratio	1,726	1	,189		
	Fisher's Exact Test				,524	,262
	Linear-by-Linear Association	1,500	1	,221		
	N of Valid Cases	10				
	Female	Pearson Chi-Square	1,111(c)	1	,292	
Continuity Correction(a)		,000	1	1,000		
Likelihood Ratio		1,498	1	,221		
Fisher's Exact Test					1,000	,500
Linear-by-Linear Association		1,000	1	,317		
N of Valid Cases		10				

a Computed only for a 2x2 table

b 4 cells (100,0%) have expected count less than 5. The minimum expected count is 2,00.

c 4 cells (100,0%) have expected count less than 5. The minimum expected count is ,50.

CONCLUSION

My research question, “Is there any positive correlation between having *Staphylococcus aureus* on skin of adolescent people and formation of acne outbreaks on their skin?” is answered in the light of the results of the experiment. As mentioned in my hypothesis it was expected that the experimental groups that are created as the ones that have pimples on skin and the ones that don’t have any, both of the groups would show equal or very close possibility levels. However, only 1 out of 5 students that carry *Staphylococcus aureus* on skin had acnes on skin, other 4 people were students without acnes. Hence, the results of the experiment and statistical analysis obtained from the chi-square tests showed that there was no positive correlation between having acnes and presence of *Staphylococcus aureus* on skin. Besides the fact that the method can be modified for more realistic results, the results are still accurate with literature. Thus, I consider the study successful.

It is worth mentioning that there is a difference between the results of female and male students that carry *Staphylococcus aureus* on skin since the results showed that 4 students who were carriers of *Staphylococcus aureus* on skin were male where only 1 was a female. Thus, this study can lead us to: “Is there any difference on carrying *Staphylococcus aureus* on skin between male and female teenagers?” To conclude, the reason why I chose this topic was to examine a possible correlation between acnes and *Staphylococcus aureus* on skin. The results of the experiment showed that there was not such correlation and partially supported my hypothesis. So, my research question is completely answered and my aim is fulfilled.

EVALUATION

The aim of the experiment was to find a possible relationship between adolescent acnes and presence of *Staphylococcus aureus* on skin. My hypothesis was “Carrying *Staphylococcus aureus* on skin doesn’t increase the number and dispersal of adolescent acnes on skin.” In order to reach my goal of answering the research question and obtaining accurate data, I set up a scientific method. First of all, 21 students were selected from class 11-U, considering their gender, age and whether they have apparent pimples and acnes or skin or not. Then, with using stuart transport medium, samples were collected from selected students’ skins. After, they were rubbed on surfaces of blood agar plates. Then, they were taken to Reference Laboratory of Republic of Turkey Ministry of Food, Agriculture and Livestock, where they were left for incubation of 24 hours.

As first table shows, 5 out of 20 students of 11-U were carriers of *Staphylococcus aureus* on skin which corresponds to a probability of 25%. According to second table, within the students who carry *Staphylococcus aureus* on skin, 20% is female whereas 80% of them are male. Only, 20% of the students who carry *Staphylococcus aureus* on skin were selected from the control groups which are composed of students who have apparent pimples on skin. 80% of them belonged to the control groups that don’t have pimples on skin.

According to the results of the survey that was made with the students who took part in the experiment, it is seen that all the students wash their face at least once in a day. Half of the students got a pimple within a week before the experiment date. This half part belongs to the control groups of 2 and 4, which represent male and female groups that have pimples on skin. Nobody belongs to control groups of 1 and 3 had a pimple that week. 30% of the students use cosmetic products on a daily basis. Within this 30%, 5 of the students were females whereas 1 was a male. Also, 4 females out of that 5 belonged to control group 4, which represents female students with pimples. Only two students, 10% of the students go to dermatologist regularly and both of these students are female and has pimples on skin. 40% of the students admit that they pop their pimples. This percentage corresponds to 8 person out of 20. 6 people out of that 8 belonged to control groups 2 and 4, representing the students that have pimples on skin. Although, this survey doesn't provide absolute certainty about the habits of the people with acnes, it still can be considered as an indicator that popping the acnes increases the dispersal of acnes on skin and impedes amelioration.

In order to compare the results of the students who carry *Staphylococcus aureus* on skin with the overall results of the whole students, table 5 and table 4 can be compared. Table 5 shows that all the students that carry *Staphylococcus aureus* on skin wash their face everyday, which don't make any difference from the results of the table 4. 20% of the students that carry *Staphylococcus aureus* on skin got a pimple within the week before the experiment date and this student belonged to control group 2, which represents male students with pimples. This percentage is relatively low when compared to the overall probability of all the students. 20% of the carriers of *Staphylococcus aureus* on skin use cosmetic products on a daily basis. None of the carriers go to dermatologist regularly and lastly, 60% of them have an habit of popping their pimples, which is a higher value when compared to overall results of all the students.

In light of the obtained datas and analyses, my hypothesis was partially supported. The results showed that there was no correlation between presence of acnes and carrying *Staphylococcus aureus* on skin. However, the control groups that represents students with pimples and students without pimples, didn't show equal or close possibilities for carrying *Staphylococcus aureus*. 5 of the students carry *Staphylococcus aureus* on skin and within this 5 students, 4 of them belongs to control groups 1 and 3 which represent students who don't have adolescent acnes on skin. Only 1 of them has acnes. Moreover, the Chi Square Test (χ^2) has been applied with the help of the statistical programme SPSS 13.0. With the help of the test, we examined a possible correlation between having acnes and presence of *Staphylococcus aureus* on skin. The predetermined alpha level of significance is 0.05. When we look at our Pearson Chi Square value (the second Chi Square table), it is 0.197. Since a p-value of 0.197 is greater than the conventionally accepted significance level of 0.05, we fail to reject the null hypothesis. In other words, there is no statistically significant correlation between having acnes and carrying *Staphylococcus aureus* on skin.

Despite the fact that my hypothesis is not fully supported, the experiment can appear as a successful one since its results maintain the literature value. According to an article,

Melissa Conrad Stöppler⁴ writes that *Staphylococci* can be found normally in the nose and on the skin of around 25%-30% of healthy people. In my experiment, 5 out of 20 students were carriers of *Staphylococcus aureus* on skin flora which corresponds to a percentage value of 25% which is coherent with the literature.

While carrying out the experiment I have met with several limitations which might have affected the results. Initially, the major limitation of the experiment was the number of people that took part in the experiment, which was relatively small. Also, the process of collecting samples from students' skins were carried out during the breaks. I had to divide 21 people into 3 groups and samples of these groups were collected in consecutive breaks. Thus, there have been a time difference between these groups. Lastly, while the incubation of bacteria on blood agar plates, one of the plates dried out.

To gain more accurate and relevant data and improve the experiment done, small changes in the method can be useful. Firstly, in order to alleviate the time difference while collecting samples two more people can help and follow the same method. Moreover, selected students can wear bonnets and all accessoires and glasses can be removed in order to minimize the affect of other factors that may carry bacteria on it. It is worth noting that the dried out blood agar plate did not contribute to the results and analyses of the experiment. Finally, the experiment should have carried with a higher number of people, in order to obtain more accurate and realistic results. Also, the statistical analysis obtained from the Chi Square Test would be more reliable if there were more students that contributed to the experiment.

⁴ http://www.medicinenet.com/staph_infection/article.htm

APPENDIX 1

Figure 1: An example from the survey carried out with the volunteer students that helped me with the experiment

1) Do you wash your face every day?

Yes

2) Did you get an acne this week?

No

3) Do you use cosmetic products like foundation or sunscreen on daily bases?

No

4) Do you go to a dermatologist regularly?

No

5) Do you pop your acnes?

No

APPENDIX 2

STUART TRANSPORT MEDIUM

Typical Formula*	gm/litre
Sodium glycerophosphate	10.0
Sodium thioglycollate	0.5
Cysteine hydrochloride	0.5
Calcium chloride	0.1
Methylene blue	0.001
Agar	5.0
pH 7.4 ± 0.2 @ 25°C	

Table 1: The table showing the ingredients of Stuart transport medium

Stuart transport medium is a non-nutrient soft agar gel containing a reducing agent to prevent oxidation, and charcoal to neutralise. This non-nutritional semi-solid substrate is used for the preservation of *Neisseria* species and other fastidious organisms during their transport from clinic to laboratory.

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