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MAKING ORAL CARE TABLETS FROM LICORICE, CRANBERRY AND BLACKBERRY PLANT EXTRACTS

Zeynep Tuna Yurder[1], Melis Erdogan[2], Hulya Yonkes[3]

[1]Student, zeyneptunayurder@posta.eyuboglu.k12.tr, Eyuboglu Science & Technology High School, Istanbul

[2]Student, meliserdogan@posta.eyuboglu.k12.tr,Eyuboglu Science & Technology High School, Istanbul

[3]Biology Teacher, hulyayonkes@posta.eyuboglu.k12.tr

_____ ,Eyuboglu Science & Technology High School, Istanbul

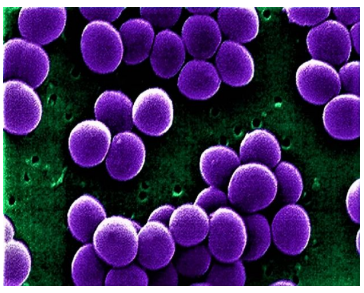
ABSTRACT

Although protecting oral health is important, we may not always be able to take care of our oral health in our daily life. Due to this problem our aim is to produce an oral care tablet which is easy-to-use and effective. The experiment applied for the project has four stages. In the first stage, the plants were powdered and macerated with 100 ml ethanol for 3 days at room temperature. The formed solutions were placed in the evaporator and ethanol was removed. The remaining extract was filtered by adding DMSO and distilled water, and the extract was created. In the second step, *S.aureus* and *E.coli* disc diffusion method was applied to bacteria. The antibacterial properties of the extracts were compared by measuring the inhibition diameters in the containers. In the third stage, tablets were produced from a mixture of three plant extracts. In the last step, the antibacterial properties of the tablet was analyzed. After all, black mulberry, liquorice and cranberry It has been obtained that the active ingredients of the plants, glycyrrhizin,which increase the contamination of oral and dental health of alkaloid, flavonoids and carotenoids *S.aureus and E.coli* antibacterial effects in. *E.coli* And *S.aureus* was observed that the inhibition diameters formed when the extracts of the plants were combined in the bacteria were higher than the inhibition diameter formed by the plants. The inhibition diameter of the tablet formed with black mulberry, cranberry and liquorice extracts is lower than the triple plant extract; Although it is less, it has been determined that it gives the desired effect. Since we observe that the tablet produced has antibacterial effects on gram (+) and gram (-) bacteria, we think that it will give similar antiviral results on viruses. Accordingly, we recommend that the concentrations of the oral tablet or the concentrations of the extract be changed and the doses that can be used in the pharmaceutical industry should be discovered and the extracts of the plants we work with should be used as alternative and natural antibiotics.

Keywords: plant, extraction, antibacterial, tablets, health

1. INTRODUCTION

Dental and oral diseases are among the most important health problems in the world. However, it does not directly threaten our lives so many people are not given enough attention. Since our mouth is the entrance of the digestive tract, it can lead to deterioration in dental health and negative problems in the digestive system. The mouth makes it ready to be swallowed by mixing the nutrients with saliva. In addition to breaking down the nutrients of our teeth, it has an important place in speech and appearance. People who have lost teeth due to tooth decay cannot properly make particular sounds and also can't break down food properly (Türkmen, Ayhan, & amp; Altuntaş, 2016, pp. 51-61).Impaired oral and dental health can become the focus of infections that cause serious problems in structures such as the heart, kidneys and joints. In addition to exaggerated tea, coffee, clenching and grinding and nutritional disorders, it also affects oral health. (Ntv, 2017).Not brushing your teeth creates a suitable environment for the bacteria in the mouth to reproduce, making your oral health a threat factor. Brushing teeth plays an important role to prevent the negative effects of bacteria in the mouth after eating. In addition, brushing teeth is important in shining teeth and eliminating bad breath. To protect the oral and dental health against these factors, it is necessary to brush teeth, use dental floss after eating. (Anadolu Medical Center Oral and Dental Health Experts, 2016). However, in the tempo of daily life, this may not be a suitable environment for the methods of protection considered. As a solution to this situation, there are pharmacological products such as lozenges and tablets used in the market. (Dyna Dental Implant., 2017), (Bilgiustam, 2019). One of the biggest factors that cause impaired oral and dental health is the bacteria that cause the formation of dental caries and mouth sores. For bacteria to be effective in tooth decay; they need to be able to adhere to the tooth surface, produce lactic acid, survive and reproduce at low pH, withstand high sucrose concentrations, and make extracellular and intracellular polysaccharides. In our mouth flora, *Streptococcus*, *Lactobacillus*, *Eshericia*, *Staphylococcus*, *Corynebacterium* and especially *Bacteroides* group bacteria are abundant. In addition, microorganisms frequently isolated from the oral cavity are given in table 1 (Özan & amp; Er, 2015, p. 36.). These microorganisms adhere to the tissues and colonize. This situation is prevented by some physical, chemical and biological mechanisms. The main bacteria that cause dental plaque and dental caries are *Staphylococcus aureus, Eshericia colisanguis* , *S. mutans* and *S. Streptococcus mutans are* the most important known cariogenic bacteria. It is approximately in the oral cavity of every person. *The ability of Streptococcus mutans* to attach to the tooth surface depends on the presence of acids released into the environment as a result of fermentation of sucrose and other sugars that can cause dental caries. The fact that microorganisms are so dense causes the soft tissues of the teeth and the tooth around to interact with bacteria in high concentrations and eventually the formation of teeth and gum diseases. (Turkmen, Ayhan, & amp; Altuntaş, 2016, pp. 51-61).



Licorice *(Glycyrrhiza glabra)* has been shown to be effective in diseases caused by weakening of the immune system. When sores such as aphthae and herpes in the mouth occur as a result of weakening of the immune system, licorice is used to prevent them. Licorice helps digestion and is an expectorant in respiratory diseases such as asthma and bronchitis. It is effective against that cause tooth decay, which is the causative agent of licorice root *Streptococcus mutans* (Ban AJ Sidkey, Afrah H. Omran, 2017) , (Vural, 2014).Black mulberry (*Morus nigra*) leaves and roots are used as tapeworm, diuretic and blood glucose regulator. Marmalade prepared from fruit, and molasses is used especially in removing tonsillitis and healing mouth and dental wounds. Thein alkaloids and carotenoid agents are used especially in the known as thrush in children and *Candida* treatment of infections caused by microorganisms oftype. (Turan Karadeniz, 2010). Cranberry *(Cornus mas*) fruits are generally used in the pharmaceutical industry as both traditional and modern drugs in the form of beverages, syrups, vinegar, marmalade and jams. It has many bioactivity. It has been reported to have antioxidant, antimicrobial, antiobesity, antihyperlipidemic, antidiabetic, antiatherosclerotic,antihypercholesterolemic,cytotoxic,cardioprotective,renalprotective,neuroprotective,hepatoprotective, anti-inflammatory, memory-enhancing,antiepileptic and diuretic effects. There are flavonoid (isoflavone) and carotenoid as active ingredients in cranberries.(Aykut & amp; Konuklugil, 2018). (Özlem Tokuşoğlu, 2017)

3. METHOD

3.1) Preparation of plant extracts

1. Licorice, black mulberry and cranberry plants were broken down with a blender and sieved to homogenize. (KULEAŞAN, 2016)

2.10 gr 3 plant samples were weighed on a sensitive scale.

3.100 ml of solvent (ethanol) were made for 3 days at room temperature. The reason for using ethanol is that ethanol and the active substances in plants have polar ends. In this way, the plants dissolve in ethanol. (Ban AJ Sidkey, Afrah H. Omran, 2017)

4. The created solutions were placed in the evaporator for 1.5-2 hours at 50 degrees.

(Budiman, Praditasari, Rahayu, & amp; Aulifa, 2019)

5. After evaporation process, the remaining extract was filtered by adding 2ml DMSO and 2ml distilled water and the extract was completed. (Nizioł-Łukaszewska, Wasilewski, Bujak, Gaweł-Bęben, Osika, & amp; Czerwonka, 2018).

Figure 3-4-5: Samples from the weighing stage of plants

3.2) Investigation of Antibacterial Properties of Extracts

Escherichia coli (ATCC 8739), *Staphylococcus aureus* (ATCC 6538) one aerobic In-vitro antibacterial investigation was performed on standard bacterial strains (Table 2).

Microorganism strains	**Gram +/-**
Eshericia coli (ATCC 8739)	Gram -
Staphylococcus aureus (ATCC 6538)	Gram +

Table 1: The strain microorganism used as follows

1) Bacterial strains (*Staphylococcus aureus, Escherichia coli)* Nutrient Broth It was incubated at 37 ± 0.1 ° C (Onur Tolga OKAN, Hülya VARLIBAŞ, Mehmet ÖZ, İlhan DENİZ, 2013).

2) Sterilized and cooled to 45-50 ° C, Müller-Hinton Agar (MHA) was poured homogeneously to 90mm petri dishes and solidified.

3) It is expected that the condensation liquid will fly for a day and at the same time, a sufficient number of empty MHA petri dishes are 37 according to the sampling method ^{0}incubated atC for one day.

4) It was prepared by suspending directly from saline from single fallen colonies in MHA medium.

5) The turbidity of the suspension was adjusted to the turbidity equivalent to the McFarland 0.5 standard. In addition, after eye adjustment, adjustment was made quantitatively with the Densitometer device .

6) Inoculum-wetted swab (swab stick) was applied to the surface of the dried MHA plate and the bacteria were inoculated on the entire agar surface.

7) The petri dish was rotated 60 ° each time during planting. The process was repeated 3-4 times with no planting on the agar surface .

8) In the last step, the swab (swab stick) was placed all around the edges of the plate and inoculation, ie planting was completed.

9) Standard antibiotic discs and ethanol impregnated discs were used as the control group. The control antibiotics used are Cephalothin (30µg) and Ceftriaxone (10µg)) -Bioanalyse products .

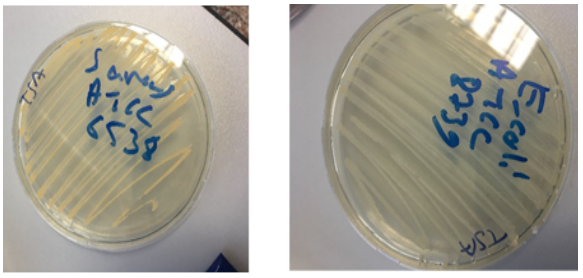


Figure 8-9: Microorganism strains used

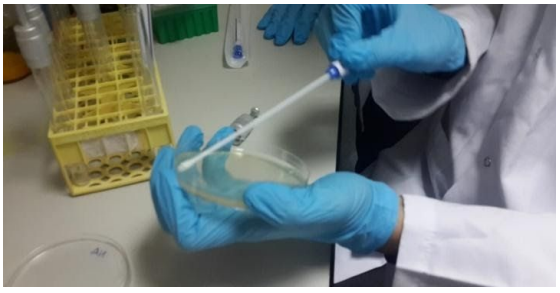


Figure 10: The stage of planting the medium

3.3) <u>Preparation of tablets from the extracts selected according to their antibacterial properties and tablet formulation</u>

After the antibacterial property analysis, it was decided that the mixture of the three extracts was suitable for the preparation of the tablet. The formulation created for the tablet manufacturing process is as follows;

Formulation Study:

Formula:

- Cranberry Extract ……. 250 microliters

-Liquorice Extract …... 250 microliters

-Black mulberry Extract … 250 microliters

-Syloid XDP 3150 (Silicon dioxide) (Grace) (Adsorbent) 500 mg

- Parteck M 200 (Mannitol 200) (Merck) 1043 mg

After determining the formulation, the steps applied for the tablet manufacturing process are as follows;

3) Mannitol (Mannitol 200) is added to the dried adsorbent-extract mixture and mixed becomes homogeneous until it.

4) At the end of the mixture, 2.2 grams of powder were obtained and this powder mixture was prepared in tablets of tablets weighing 275 mg.

5) The mixture (tablet powder) was pressed in a tablet press machine with a pressure of 400-500 psi .

3.4. Antibacterial analysis of the tablet obtained In the

In a microbiology laboratory *Staphylococcus aureus* ,ATCC 6538, *Escherichia coli* oral tablets were created in which the extracts would be included in three extracts according to the antibacterial status of the extracts in accordance with the in-vitro antibacterial research on ATCC 8739 standard bacterial strains.

In this tablet, in order to compare the effect of the extract with the triple mixture, bacteria were cultivated by disc-diffusion method and the procedures were repeated for the tablet.

1) Adsorbent (Syloid XDP 3150) is treated with extract and mixed until a homogeneous mixture is obtained.

2) After mixing, the alcohol was kept in the extract to remove some amount of alcohol.

Take the tablet to discs in 10 **µl** liquid In order to impregnate, the following processes have been carried out;

1. The tablet pressed by the compacting machine was pulverized in mortar.
2. Since the tablet is intended to be active in the mouth, a mixture of 0.16 g KCL, 0.16 NaHCO110 ml distilled water to create artificial saliva 3, 0.13 g KSNC, 0.05 g $NaH_2PO_4.H_2O$ is obtained in(Zeynep Duymuş, Nurdan Sağsöz, Ömer Sağsöz, Funda Bayındır, 2014).
3. A powdered tablet was added into the resulting artificial saliva liquid and mixed at 36 degrees for 20 minutes with a magnetic stirrer.
4. As a result of mixing, a homogeneous liquid was obtained.
5. 10two discs of this liquid **µl** of micropipette was impregnated with.
6. For each type of bacteria, the swab (swab stick) wetted with Inoculum was applied to the surface of the dried MHA plate and the bacteria were inoculated on the entire agar surface. These discs were then placed in agars.

5. RESULTS

5.1. Extracted and tablets of the bacteria are analyzed on

Results obtained by the analysis of disc diffusion method on aerobic bacteria Eshericia coli (ATCC 8739) and Staphylococcus aureus (ATCC 6538) tablets containing binary combinations and all at the same time of cranberry, licorice and black mulberry plant extracts.The inhibition diameters obtained in the analysis are as in the table.

Table 2. Inhibition diameters obtained by the disc diffusion method

Extracts and mixture extracts	***Staphylococcus aureus*** (ATCC 6538)	***Eshericia coli*** (ATCC 8739)
(E1) cranberry extract	3mm	9mm
(E2) blackberry extract	2mm	2mm
(E3) liquorice extract	5mm	6mm
(E1 + E2) cranberry + black mulberry extract	0mm (no growth under the disc)	2mm
(E1 + E3) cranberry + liquorice extract	4mm	3mm

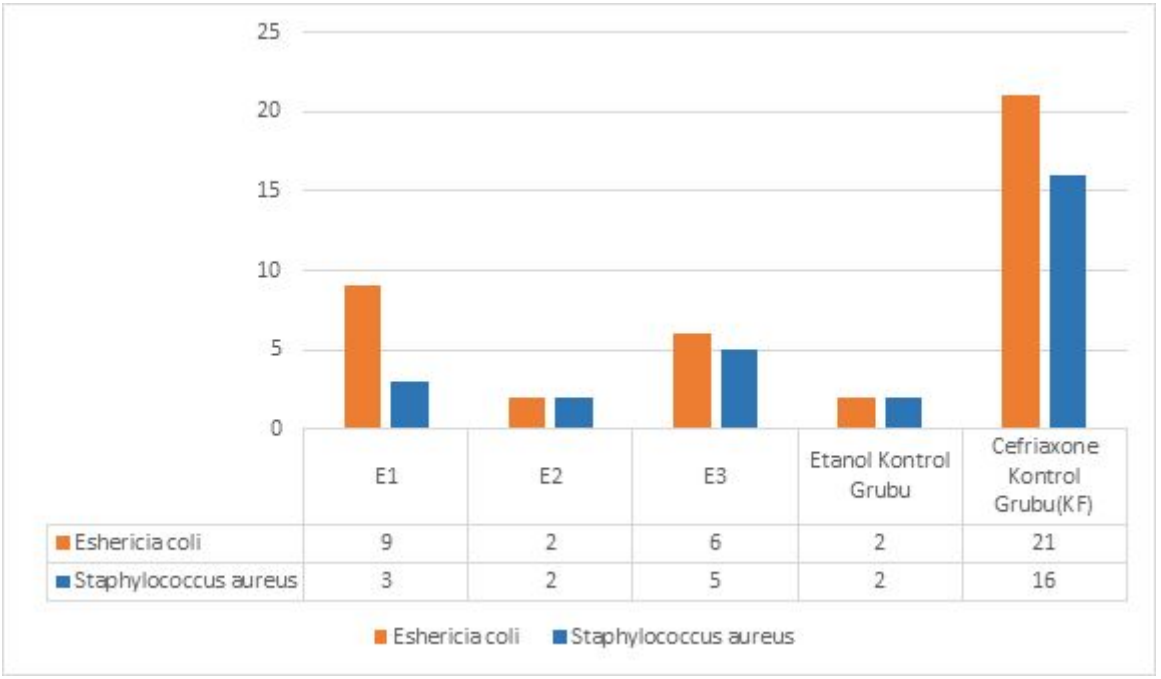
(E2 + E3) black mulberry + liquorice extract	1mm	2mm
E1 + E2 + E3 extract	11mm	13mm
E1 + E2 + E3 tablet	7mm	9mm
Ethanol Control Group	2mm	2mm
Ceftriaxone Control Group (KF)	16mm	21mm

6. CONCLUSION AND DISCUSSION

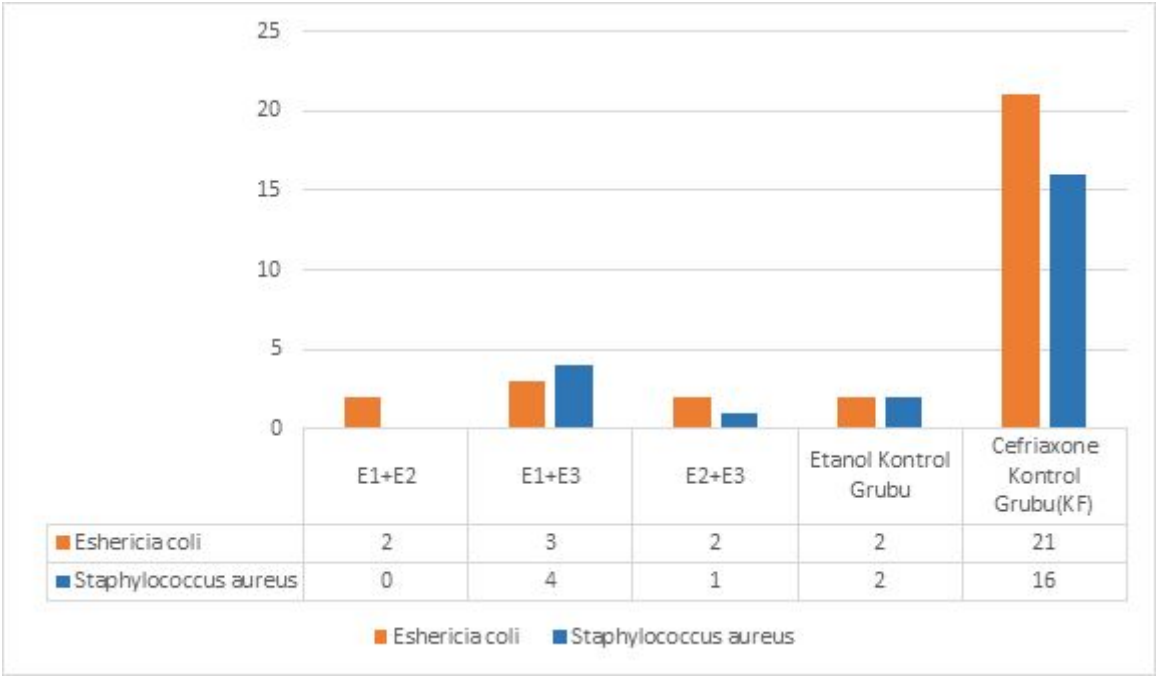
The Liquorice Grown in our country *(Glycyrrhiza glabra)*, black mulberry (*Morus nigra*) and cranberry *(Cornus mas antibacterial* effect of) plants was investigated. At the same time,active substances of these plants, glycyrrhizin,which increase the contamination of alkaloid, flavonoids and carotenoids in oral and dental health *S.aureus and E.coli* bacteria, information has been obtained that the antibacterial effects on.

The highest inhibition diameters are seen in the control group, ceftriaxone. This is an expected result. If the plant extracts are used individually, in combination with double and triple combinations, the results obtained; It was observed that when the extracts are applied one by one, the situation where the three come together is more effective than the results. It was found that the inhibition diameters were seen in their individual applications, but they did not show sufficient effect to wear the oral tablet. Licorice*(Glycyrrhiza glabra)*, black mulberry (*Morus nigra*) and cranberry *(Cornus mas*) plants mixture extract *S.aureus*(ATCC 6538) and *E.coli (ATCC 8739)* is effective on and. *ItS.aureus* 11 mm in bacteria and 13 mm in *E.coli* was observed that formed an inhibition diameter.

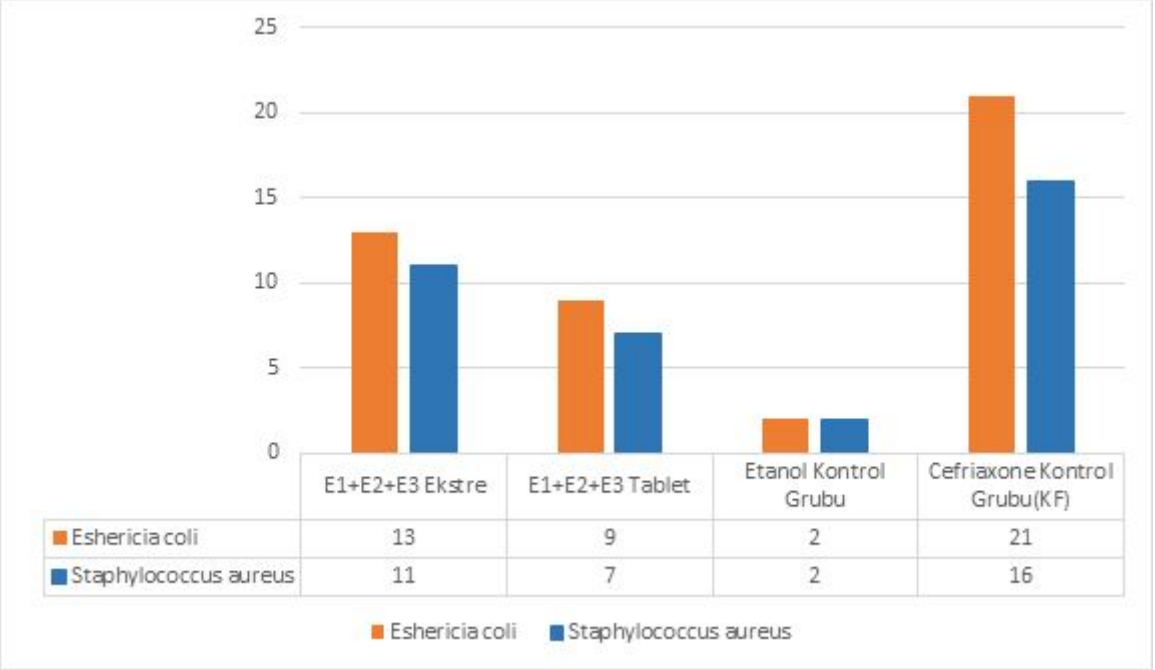
The inhibition effect of the tablet, including all three of the plant extracts, was observed to be less than the effect of the extract mixture but still effective. The tablet manufactured *S. aureus* 7 mm in bacteria and 9 mm in *E.coli* was observed that had an effect of bacteria (Chart 1-2-3).



Graphic 2: Inhibition analysis graphic created by the extracts one by one



Graphic 3. Graph of inhibition analysis formed by binary groups of extracts



Graphic 1: Inhibition analysis graphic of triple extract and tablet

7. SUGGESTIONS

1)licoriceour project.*(Glycyrrhiza glabra)*, black mulberry (*Morus nigra*) and cranberry *(Cornus mas*Gram (+)which has a negative effect on the oral and dental health of the mixture extract of) plants in *S.aureus,* andgram (-) *E.coli* its effect againstbacteria was quantitatively observed. While making this observation, it was quantitatively observed that the effect of each plant extract was low, but the effects increased when they were together. In the tablet obtained in line with the results obtained, a mixture extract showing antibacterial properties was created. Accordingly, it is recommended to study the doses that can be used in the medical pharmaceutical industry by changing the concentrations of the oral tablet or the concentrations of the extract.

2) Alternative ways to replace antibiotics are sought in order to increase the use of antibiotics and minimize antibiotic resistance. Welicorice*(Glycyrrhiza glabra)*, black mulberry (*Morus nigra*) and cranberry *(Cornus mas*recommend using the antibacterials of) plant extracts as an alternative and natural antibiotic to the pharmaceutical industry.

3) As a result of the experiments conducted throughout the project, it was found that licorice*(Glycyrrhiza glabra)*, black mulberry (*Morus nigra*) and cranberry *(Cornus mas*) plant extracts showed higher bactericidal (antibacterial) properties. It was determined that the produced oral tablet had an antibacterial effect on gram (+) and gram (-) bacteria. We think that these ointments will give similar results on viruses. We suggest studying the antiviral properties of the

triple mixture of plant effects studied for future research.

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