Micro Environer **2021, 1(1): 54-62**
https://doi.org/10.54458/mev.v1i01.6673

Research Article **Open Access**

Analysis of antimicrobial activity of Ashurak extract prepared with different solvents on *Klebsiella pneumoniae* and *Shigella dysentery* isolated from poultry feces

Mehdi Jahantigh[1*]. Hassan ahmadi[2]

Article info
Received: 23 May 2021
Revised: 05 Sep 2021
Accepted: 19 Oct 2021

Keywords:
Shigellosis, Flavonoids, Ethyl acetate, Hydroalcohol, Methanol, Proanthocyanidin

ABSTRACT

Bacteria are more likely to cause disease than foodborne pathogens. Articles have shown that most plant extracts have antimicrobial properties. The aim of this article was to analyze the antimicrobial activity of Ashurak extract prepared with different solvents on *K. pneumoniae* and *Sh. dysentery* isolated from poultry feces. In this article, antimicrobial effects of various extracts (ethyl acetate, aqueous, ethanolic, methanolic and hydroalcoholic) on *K. pneumoniae* bacteria *Sh. dysentery* isolated from poultry feces was obtained by diffusion method in Müller Hinton agar medium using 6 mm paper discs according to the instructions. Mean comparison was performed using two-way analysis of variance and post hoc test with the least significant difference. The lowest inhibitory concentrations of aqueous, ethanolic, methanolic and hydroalcoholic extracts of Ashurak on *K. pneumoniae* were 12.5, 12.5, 12.5 and 3.1 ppm, respectively. The lowest inhibitory concentration of aqueous, ethanolic, methanolic and hydroalcoholic extracts of Ashurak on SchiegelDysentery was equal to 12.5, 12.5, 25 and 25 ppm. Among the mentioned extracts, the aqueous extract had the greatest effect on the inhibition of *K. pneumoniae* , but there was no difference between the extracts. According to the results of the present study and the increasing resistance to synthetic antibacterial substances, it seems that Ashurak can be considered as effective plants in cleansing some bacteria, including *K. pneumoniae* and *Sh. dysentery* . In addition, the results of this study showed that Ashurak plant is more effective In contrast; *K. pneumoniae* had dysentery compared to Shigella.

1. Introduction

One of the challenges of the food and drug industry is the development of microbial indices resistant to drugs and antibiotics. Today, due to the toxic and carcinogenic properties of chemical and synthetic compounds, the use of medicinal plants to treat chronic diseases has attracted the attention of many researchers. Therefore, the use of natural antimicrobial and antioxidant compounds such as organic acids, essential oils and plant extracts can be a suitable and safe alternative in food [1]. Also, in problems related to oral and arthritis that collagen It is exposed to degradation, phenolic compounds and antioxidants of these plants can prevent it [2]. Flavonoids are phenolic compounds that directly inhibit the active molecules of superoxide, hydrogen peroxide, hydroxyl and peroxyl radicals[3].

Numerous studies have shown that plants containing flavonoid compounds exhibit high antioxidant activity [3]. This antioxidant activity is usually related to the ability of these compounds to give electrons or hydrogen atoms, and therefore. They are medically important and significant [4]. It is noteworthy that the seeds and skin parts of some fruits have more antioxidant activity even than meat. For example, grape seeds and pomegranate peel have more antioxidant activity than meat and are rich in

[1] *Associate professor of clinical pathology , Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Zabol, Iran*
[2] *Agricultural Biotechnology Institute, University of Zabol, Zabol, Iran*
***Corresponding Author:** Mehdi Jahantigh (mjahantig@yahoo.com)*

proanthocyanidins, which are strong inhibitors of reactive oxygen radicals [5].

Reactive oxygen radicals can attack the best cell compounds such as fatty acids, proteins, nucleic acids and pigments [6] to neutralize the toxic effect. Reactive oxygen radicals, antioxidant compounds are needed. Plant cells from two enzymatic antioxidant systems (superoxide dismutase, catalase, peroxidase, antioxidant metabolites phenyl, chenyl anhydride, carotene, phenyl, carthenol) [7, 8].

Since the second half of the last century, extensive research has been done on medicinal plants in most countries of the world, and after that, many herbal medicines have been prepared and marketed, due to the rich flora of Iran, which is more than 7500 plant species and ان They are medicinal [9, 10]. The need to study the antibacterial properties of plants such as Ashurak on *K. pneumoniae* and Shigella has been important.

Rhazya stricta is an important drug that is rich in alkaloids containing anti-cancer alkaloids. Herbal medicines are used locally to treat various diseases in South Asia (Pakistan, India and Afghanistan) and the Middle East (for example, Saudi Arabia, Qatar, the United Arab Emirates (UAE), Iran and Iraq). Some of its alkaloids also have anti-cancer propertiesHas been reported. More than 100 alkaloids have been isolated from *R. stricta* leaves, but their medicinal activities are known only for a few of these compounds. Very few ethnographic studies have been performed on *R. stricta* [11].

K. pneumoniae is an important opportunistic pathogen that frequently causes urinary tract infections and pneumonia in immunocompromised individuals. After Escherichia coli, the most common cause of sepsis is gram-negative bacteria and nosocomial infections. Global spread of multidrug-resistant strains of *K. pneumoniae* K is an important cause for concern. Antibiotic resistance is the ability of a microorganism to stop the effect of an antibiotic and is a major cause of failure in the treatment of infectious diseases, which increases the incidence of disease, mortality and health care costs. High resistance of Klebsiella to antibiotics and their rapid spread in different parts of the hospital cause major problems in treatment and cause septicemia and death of patients [12, 13].

Shigellosis is an intestinal infection whose symptoms range from watery diarrhea to severe inflammation. This disease with pain severe abdominal pain, fever, bloody stools and mucus are known. This disease is usually self-limiting, except in cases where the patient has an immunodeficiency or primary medical treatment is not available [14]. *Sh. dysentery*, buidi and Sonei are different species of this bacterium [15].

Shigella is highly contagious and is easily transmitted from person to person through feces and mouth. Eating contaminated food and water and human feces is the primary source of Shigella. Specific symptoms of dysentery include: anorexia, fever, intestinal inflammation, bloody stools, purulent, abdominal pain And the feeling of incomplete emptying of the intestine is anal pain [16].

Antioxidants have many uses in addition to the treatment and prevention of cancer and atherosclerosis, for example, natural antioxidant compounds of olive leaf to increase the storage of fats and oils, almond peel extract due to its antioxidant activity is used to preserve chips and non-chips [17]. On the other hand, due to the fact that different extracts have different ability to extract active substances and of course have different properties on the lack of bacterial growth, so for this purpose In the present study, we will try to extract the extract of Ashurak plant with different solvents and then focus on the inactivity of *K. pneumoniae* and Shigella bacteria

2. Materials and methods of work

2.1. Herbal materials

R. stricta was collected from Saravan (Coordinates: 27 ° 22′15 ″ N 62 ° 20′03 ″ E) and their species were identified in the botanical laboratory of University of Zabol (Fig. 1).

After collecting these samples in natural conditions and in dry shade and then crushed,

to prepare the extract, 40 g of dry plant powder was placed in half-liter Erlenmeyer flakes containing 200 ml of 96% ethanol, methanol, ethyl acetate, hydroalcoholic and water. The contents of the Erlenmeyer flask were mixed at room temperature for 24 hours with a shaker (at 130 rpm) and then filtered through Whatman 2 paper. The solvent was separated from the extract by a rotary apparatus using a vacuum pump (distillation in vacuum). The weighted extracts were then dissolved in DMSO solvent

Figure 1. Appearance characteristics of leaves (A, B), Stem (B), and flowers (A, C) of *R. stricta* plant [18]

2.2 Culture and identification of *K. pneumoniae* and *Sh. dysentery*

To identify *K. pneumoniae* , the samples were cultured on agar medium by flame and incubated for 37-24 hours at 37 °C. After bacterial growth, the strains of *K. pneumoniae* were identified based on microbiological methods including hot staining and microscopic observation of bacilli, OF-TSI-SIM-MR-VP-citrate-urea-lysine decarboxylase and orrentine deoxyribase [19] tests. To identify *Sh. dysentery* from Salmonella Shigella agar specific culture mediaAnd selenite F is used [20].

Determining the level of inhibitory concentration and minimum lethal concentration of Ashurak extract prepared with different solvents: Sensitivity of bacterial isolates with multiple resistance to the extract was determined using the dilution method in the well. Seven wells were made in the solid culture medium and 100 µl of Müller-Hinton nutrient medium (MHB) was added to each well. To the first well, 100 ml of diluted solution of essential oil or extract was added, and after mixing, 100 µl of the first well was removed and added to the second well.This was done until the last well was removed from the last well of 100 µl of culture medium. 100 µl of microbial suspension containing 107 units / ml equivalent to 0.5 McFarland was added and incubated at 37 ° C. It was exposed to Celsius for 24 hours. The first tube to prevent the growth of bacteria after incubation is considered and to ensure clear wells, 10 microliters are taken and transferred to the Müller Hinton agar medium, and after 24 hours, the first dilution that can be 99.9.The percentage of bacteria killed is shown as the minimum lethal concentration

3. Results

The results of this paper showed that the lowest inhibitory concentration of Ashurak ethyl acetate extract against *K. pneumoniae* was 25 ppm, in which 2 strains were inhibited. The lowest inhibitory concentrations of aqueous, ethanolic, methanolic and hydroalcoholic extracts were 12.5, 12.5, 12.5, 3.1 ppm, respectively, in which 2-3-1 and 2 strains were inhibited at these concentrations (Table 1).

The lowest inhibitory concentration of ethyl acetate against *Sh. dysentery* samples was 25 ppm. While the lowest inhibitory concentration of aqueous extract, ethanol, Methanolic and hydroalcoholic was equal to 12.5, 12.5, 25, 25 ppm. The highest inhibitory concentrations of ethyl acetate-aqueous-ethanol-methanolic and hydroalcoholic were 50, 25, 25, 50 and 50 ppm (Table 1).

The lowest lethal concentrations of ethyl acetate, aqueous, ethanolic, methanolic, hydroalcoholic extracts against *K. pneumoniae* were 50, 25, 25, 25 and 6.25 ppm, in which 2, 2, 3, 1 and 2 strains were inhibited at this concentration (Table 2).

Analysis of variance diameter of growth inhibition zone due to the effect of different solvents showed that among the mentioned extracts of the extractAqueous had the greatest effect on the inhibition of *K. pneumoniae* but there was no difference between the extracts in the inhibition of *Sh. dysentery* (Fig. 2).

Table 1. Minimum bactericidal concentrations of Ashurak extract with different solvents on Klebsiella and Shigella (ppm)

Bacteria Strain	Ethyl acetate	Aqueous extract	Ethanolic	methanolic	hydroalcoholic
K. neumoniae 1	50	25	12.5	12.5	12.5
K. neumoniae 2	50	25	12.5	50	25
K. neumoniae 3	50	25	25	50	12.5
K. neumoniae 4	50	12.5	25	50	25
K. neumoniae 5	50	25	25	50	3.1
K. neumoniae 6	50	25	25	50	3.1
K. neumoniae 7	25	25	12.5	25	25
K. neumoniae 8	25	12.5	25	25	50
Sh. Dysentery 1	25	25	12.5	25	25
Sh. Dysentery 2	25	25	25	25	25
Sh. Dysentery 3	50	12.5	25	50	50

Table 2. Minimum lethal concentration of Ashurak extract with different solvents on Klebsiella and Shigella (ppm)

Bacteria Strain	**Ethyl acetate**	**Aqueous extract**	**Ethanol**ic	**methanolic**	**hydroalcoholic**
K. neumoniae 1	100	50	25	25	25
K. neumoniae 2	100	50	25	100	50
K. neumoniae 3	100	50	50	100	25
K. neumoniae 4	100	25	50	100	50
K. neumoniae 5	100	50	50	100	6.25
K. neumoniae 6	100	50	50	100	6.25
K. neumoniae 7	50	50	25	50	50
K. neumoniae 8	50	25	50	50	100
Sh. Dysentery 1	50	50	25	50	50
Sh. Dysentery 2	50	50	50	50	50
Sh. Dysentery 3	100	25	50	100	100

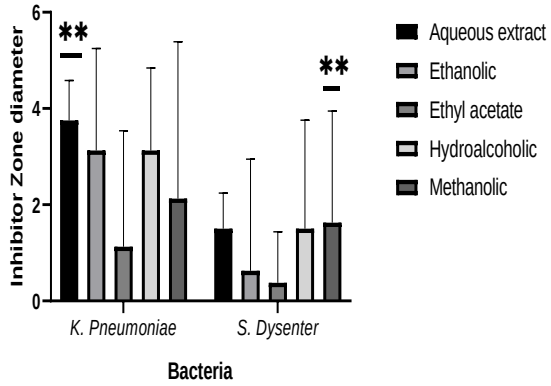

Fig. 2. Inhibitory halo diameter of Ashurak extract on Klebsiella and Shigella bacteria (mm)

4. Discussion

The antibacterial effects of whole fruit powder extract and pomegranate and pumpkin juice (Bottle Gourd) on gram-positive and gram-negative bacteria such as Salmonella typhi, Escherichia coli, *K. pneumoniae* , Bacillus cereus, Bacillus subtilis and *Sh. dysentery* have been investigated. It was concluded that pomegranate powder extract with 250 micrograms per disc could slightly inhibit the growth of the studied bacteria except Bacillus subtilis. Pomegranate juice and pumpkin and pumpkin powder extract did not show antibacterial activity [21]. However, in the present study, it was found that the hydroalcoholic extract of Ashurak had the greatest effect on *K. pneumoniae* and *Sh. dysentery* , which showed that Ashurak was more effective than pomegranate peel.

The antimicrobial effect of aqueous and alcoholic extracts of chamomile, turmeric, peppermint and nettle on *K. pneumoniae* and comparing their effect with common antibiotics, and concluded that the alcoholic extract of chamomile with a maximum

diameter of no growth zone of 14 mm against Klebsiella Has shown the best effect. The bacteria in question showed resistance to aqueous and alcoholic extracts of deciduous plants and also ethanolic extract of nettle and chamomile plants in the amount of 0.39 mg/ml had the best antibacterial effect. The minimum growth inhibitory concentration of blueberry extract against *K. pneumoniae* was 12.5 mg/ml and lower than other types of extracts [22]. In the present study, it was found that the greatest effect of Ashurak plant was obtained from hydro-alkaline extract with 10 mm diameter growth inhibition zone. And indicates greater power of growth inhibitionBacteria were caused by chamomile compared to Ashurak. Investigation of zinc oxide nanoparticles with antimicrobial and antibiotic activity against *K. pneumoniae* and concluded that antibiofilm activity and biofilm removal by zinc oxide nanoparticles were obtained at 50 and 500 µg/ml, respectively [23]. The effect of iron oxide nanoparticles on the expression of biofilm production genes and antibiotic resistance in *K. pneumoniae* strains has been investigated and it has been concluded that both types of iron oxide nanoparticles have an important effect in inhibiting biofilm formation and resistantThey had antibiotics for this bacterium [24]. In the present study, it was found that the minimum inhibitory concentration of hydrochloric acid extract of Ashurak against *K. pneumoniae* was equal to 3.1 ppm, which had a greater effect than the mentioned plants.

The antiplasmid effects of extracts of safflower, safflower, licorice, eucalyptus on resistant strains of *K. pneumoniae* have been investigated and it has been concluded that all extracts have antimicrobial properties and have been able to prevent the growth of resistant strains of *K. pneumoniae* . Among these are eucalyptus leaf extractIt has the highest antimicrobial properties (average MIC 0.1 mg/ml) [25]. In the present study, it was found that the minimum inhibitory concentration of hydrochloric acid extract of Ashurak against *K. pneumoniae* was equal to 3.1 ppm, which was less effective than eucalyptus extract.

It has been reported that the ethanolic extract has a positive effect on Staphylococcus aureus [26, 27] but the affected oil has shown a greater effect [27]. Also, the positive effect of other extracts of chloroform, ethyl acetate and methanol on the leaves and also the dependence of these extracts on the concentrationThey were more effective on Staphylococcus aureus [28] and more effective than methanolic extract than ethanolic extract of the leaves and branches of the plant on the bacteria Lestria monocytogenes, Sodomonas aeruginosa and Staphylococcus aureus[29].. In one study, it was found that although methanolic extract of the plant did not show a high effect on the diffusion activity of Escherichia coli, but the minimum lethal concentration for Escherichia coli was reported above 40 mg / ml [30] and in other studies, noThe effect of the case extract on Escherichia coli has been reported [27, 29]. In a study, Houshmand et al. Had the highest effect of the plant extract on Pseudomonas aeruginosa at a concentration of 2.5% with a growth inhibition zone diameter of about 16 mm and the lowest effectiveness of the plant extract at a concentration of 5% on lactobacilli with a growth inhibition zone diameter of about 6 mm reported and in general concluded that the extract with different concentrations has different effects on bacteria [31]. In general, it has been found that different extracts have different effects depending on the type of plantThey inhibit bacteria [32, 33]. In the present study, it was found that the most effective solvent for extracting the active ingredients was the hydrocarbon solvent.

The study of Kouitcheu, reports the *in vitro* and *in vivo* anti-shigellosis activity of the methanol extract of this plant on rats. The result show that *in vitro*, the extract had an antimicrobial effect on 11 out of the 17 pathogenic strains tested. The values of CMI and CMB obtain against *Shigella dysenteriae* type I were 800 and 6400 µg/ml respectively*. In vivo*, diarrhoea induction was effective and we notice an increase in faeces frequency and weight ($p < 0.05$), increase in the percentage of diarrheic stool released as well as the mucus contained in stool ($p < 0.05$), an increase in bacterial population in stool ($p < 0.05$). *Picralima nitida* extract, like

ciprofloxacin markedly reduces the frequency faeces released and sd1 density from 100% (diarrheic rats) to 47.22 and 61.69% (500 mg/kg) respectively. It also slowed down the movement of charcoal meal through gastro-intestinal tract with the percentage of intestinal length covered of 60.54 (500 mg/kg)[34].

The study of René, investigated the acute toxicity and effect of the aqueous ethanol extract of the plant on gastrointestinal propulsion, in vitro bacterial growth and in vivo bacillary dysentery. The aqueous ethanol extract of E. prostrata was not toxic. In vitro, the minimal inhibitory and minimal bactericidal concentrations of the extract were 3,500 and 12,000 µg/ml, respectively. In vivo, diarrhea went along with increase in faeces frequency ($P < 0.01$ by the 3rd day), increase in the bacterial population to a maximum on the 2nd day after infection ($P < 0.01$). The death rate in diarrheic control group was 100% by day 6. E. prostrata extracts (20 and 40 mg/kg), like norfloxacin, reduced the bacterial growth ($P < 0.01$), so that by the 6th day Sd1 density was < 0.01) reduction in faeces frequencies. The extract exhibited notable ($P < 0.01$) inhibition of intestinal propulsion[35].

The study of Khosravi was to investigate the antibacterial effects of *Tecomella undulata* and *Momordica charantia* extracts on *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Shigella dysenteriae*. The result show that the minimum inhibitory concentrations of *Tecomella undulata* extract against the studied bacteria were 1.87 to 3.75 mg/ml. The highest lethal concentration of leaf extract was 15 mg/mL and the lowest lethal concentration of *Momordica charantia*extract was 1.87 to 3.75 mg/mL. The highest *Momordica charantia* extract concentration was 15 mg/mL and *Shigella dysenteriae* were eliminated at this concentration[36].

The study of Saravani was to investigate the antimicrobial effects of Momordica charantia against pathogenic bacteria. The results of this study showed that the lowest inhibitory concentration of extract against bacteria was 12.5 ppm, (Vibrio cholera, Pseudomonas aeruginosa and E. coli) the results also showed that only one inhibited. However, the highest inhibitory concentration was estimated 25 ppm against Shigella dysenteriae and Bacillus cereus and highest bactericidal concentration was estimated 25 and 50 ppm[37].

5. Conclusion

Overall, the results of this study showed that Ashurak had a greater effect against inhibition of *K. pneumoniae* than *Sh. dysentery* . Among the mentioned extracts, hydro-alcoholic extract had the most effect.
Considering that every research has its shortcomings and the present research is no exception, so it is suggested that in the next stage, the active ingredients of Ashurak, which has played an effective role on antimicrobial properties, beBe evaluated directly to present the final conclusion more accurately

Abbreviation

Not applicable

Conflict of Interest

Authors declare there is no conflict of interest in this research study

Consent for publications

All authors read and approved the final manuscript for publication.

Availability of data and material

All data generated during this study are included in this published article.

Ethics approval and consent to participate

No human or animals were used in the present research.

Ethics declarations.

The authors declare no conflict of interest in financial or any other sphere. All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

Source of Funding

No funding was received against this research study.

References

1. Negi P S. (2012). Plant extracts for the control of bacterial growth: Efficacy, stability and safety issues for food application. International Journal of Food Microbiology, 156(1): 7-17. https://doi.org/10.1016/j.ijfoodmicro.2012.03.006
2. Kwok C Y, Wong C N Y, Yau M Y C, Yu P H F, Au A L S, Poon C C W, Seto S W, Lam T Y, Kwan Y-W, Chan S W. (2010). Consumption of dried fruit of *Crataegus pinnatifida* (hawthorn) suppresses high-cholesterol diet-induced hypercholesterolemia in rats. Journal of functional foods, 2(3): 179-186. doi: 10.1016/j.jff.2010.04.006
3. Sharma R, Samant S, Sharma P, Devi S. (2012). Evaluation of antioxidant activities of *Withania somnifera* leaves growing in natural habitats of North-west Himalaya, India. Journal of Medicinal Plants Research, 6(5): 657-661. doi: 10.5897/JMPR11.257
4. Shrivastava A, Roy S. (2013). *Cucurbitaceae*: A ethnomedicinally important vegetable family. Journal of Medicinal Plants Studies, 1(4): 16-20.
5. Bagchi D, Bagchi M, Stohs S J, Das D K, Ray S D, Kuszynski C A, Joshi S S, Pruess H G. (2000). Free radicals and grape seed proanthocyanidin extract: importance in human health and disease prevention. Toxicology, 148(2): 187-197. doi: 10.1016/S0300-483X(00)00210-9
6. Jafari R, Manochehri Kalantari K, Ahmadi Mousavi A. (2007). Effect of paclobutrazol on accumulation of antioxidants in tomato seedlings under cold stress. Iranian Journal of Biology, 20: 206-216 (In Persian).
7. Chen Y, Zhang M, Chen T, Zhang Y, An L. (2006). The relationship between seasonal changes in anti-oxidative system and freezing tolerance in the leaves of evergreen woody plants of Sabina. South African Journal of Botany, 72(2): 272-279. doi: 10.1016/j.sajb.2005.09.004
8. Mollá S, Villar-Salvador P, García-Fayos P, Rubira J L P. (2006). Physiological and transplanting performance of *Quercus ilex* L.(holm oak) seedlings grown in nurseries with different winter conditions. Forest ecology and management, 237(1): 218-226.
9. Zakizadeh M, Nabavi S, Nabavi S, Ebrahimzadeh M. (2011). In vitro antioxidant activity of flower, seed and leaves of Alcea hyrcana Grossh. European review for medical and pharmacological sciences, 15(4): 406-412.
10. Mozdastan S, Ebrahimzadeh M A, Eslami S. (2015). Effect of Increasing the Polarity of Solvent on Total Phenol and Flavonoid Contents and Antioxidant Activity of Myrtle (Myrtus communis L.). [Research(Original)]. Journal of Mazandaran University of Medical Sciences, 25(126): 68-81 [Farsi with abstract English].
11. Gilani S A, Kikuchi A, Shinwari Z K, Khattak Z I, Watanabe K N. (2007). Phytochemical, pharmacological and ethnobotanical studies of *Rhazya stricta* Decne. Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives, 21(4): 301-307. https://doi.org/10.1002/ptr.2064
12. David S, Reuter S, Harris S R, Glasner C, Feltwell T, Argimon S, Abudahab K, Goater R, Giani T, Errico G. (2019). Epidemic of carbapenem-resistant *Klebsiella pneumoniae* in Europe is driven by nosocomial spread. Nature microbiology, 4(11): 1919-1929. https://doi.org/10.1038/s41564-019-0492-8
13. Martin R M, Bachman M A. (2018). Colonization, infection, and the accessory genome of *Klebsiella pneumoniae*. Frontiers in cellular and infection microbiology, 8: 4. https://doi.org/10.3389/fcimb.2018.00004
14. Clarkson K A, Frenck Jr R W, Dickey M, Suvarnapunya A E, Chandrasekaran L, Weerts H P, Heaney C D, McNeal M, Detizio K, Parker S. (2020). Immune response characterization after controlled infection with lyophilized *Shigella sonnei* 53G. Msphere, 5(5): e00988-00919. https://doi.org/10.1128/mSphere.00988-19
15. Niyogi S K. (2005). Shigellosis. Journal of microbiology, 43(2): 133-143.

16. Sur D, Ramamurthy T, Deen J, Bhattacharya S. (2004). Shigellosis: challenges & management issues. Indian J Med Res, 120(5): 45-54.
17. Rehman Z-U. (2003). Evaluation of antioxidant activity of methanolic extract from peanut hulls in fried potato chips. Plant Foods for Human Nutrition, 58(1): 75-83. https://doi.org/10.1023/A:1024031522588
18. Beigomi M, Shahraki-Mojahed L, Heydari-Sadegh B, Dahmardeh N, Rouhani R, Javadian F. (2021). Evaluation of Antimicrobial Activity of *Rhazya Stricta* (Apocynaceae) Extract Prepared with Different Solvents on *Staphylococcus Aureus* (Staphylococcaceae) Isolated from Humans. Int. J. Adv. Biol. Biomed. Res., 9(3): 241-253. https://doi.org/10.22034/ijabbr.2021.528883.1357
19. Mahon C R, Lehman D C, Manuselis G. (2018). Textbook of diagnostic microbiology-e-book: Elsevier Health Sciences. 1088 pages,
20. Bennish M L, Salam M A, Hossain M A, Myaux J, Khan E H, Chakraborty J, Henry F, Ronsmans C. (1992). Antimicrobial resistance of Shigella isolates in Bangladesh, 1983–1990: increasing frequency of strains multiply resistant to ampicillin, trimethoprim-sulfamethoxazole, and nalidixic acid. Clinical infectious diseases, 14(5): 1055-1060. https://doi.org/10.1093/clinids/14.5.1055
21. Ghodratollah N, Hassanpour-Fard M, Bodhankar S, Dikshit M. (2011). Pomegranate, Bottle gourd, Antibacterial activity, Tetracycline. [Original Article]. Journal of Birjand University of Medical Sciences, 17(4): 257-264.
22. Azizi Alidoust F, Anvari M, Ataei Jaliseh S. (2020). Antimicrobial Activity of Aqueous and Alcoholic Extracts of Chamomile, Fleawort, Aquatic Pennyroyal and Nettle Plants on Klebsiella pneumoniae and Comparing Their Effects with Common Antibiotics. Iranian Journal of Medical Microbiology, 14(4): 361-373. http://dx.doi.org/10.30699/ijmm.14.4.361
23. Ahmadinasab R, Aghaei S, Gasemzadeh M A. (2018). Synthesis of zinc oxide nanoparticles with antimicrobial and antibiofilm activity against *Klebsiella pneumonia*. Applied Biology, 8(31): 107-119.
24. Masoudian S, Hosseini F, Amini K. (2021). Investigating the Effects of Iron Oxide Nanoparticles on the Expression of Biofilm Production Genes and Antibiotic Resistance in *Klebsiella pneumoniae* Strains. Biological Journal of Microorganisms, 10(38): 17-26. 10.22108/bjm.2020.123026.1300
25. Shakibaiee M, Heydari M, Ahmadinezhad M, Mohammadi M. (2000). Plasmid Curing Activity of Five Plant Extracts on Multiple Resistant Plasmid Bearing *Klebsiella Pnumoniae* Strains. [Research]. Journal of Guilan University of Medical Sciences, 9(33): 1-10.
26. Alem G, Mekonnen Y, Tiruneh M, Mulu A. (2008). Invitro antibacterial activity of crude preparation of myrtle (*Myrtus communis*) on common human pathogens. Ethiopian medical journal, 46(1): 63-69.
27. Salvagnini L E, Oliveira J R S, Santos L E d, Moreira R R D, Pietro R C L. (2008). Evaluation of the antibacterial activity of *Myrtus communis* L.(Myrtaceae) leaves. Revista Brasileira de Farmacognosia, 18(2): 241-244.
28. Gholamhoseinian-Najar A, Mansouri S, Rahighi S. (2009). Effect of sub-inhibitory concentrations of myrtus communis leave extracts on the induction of free radicals in *Staphylococcus aureus*; A possible mechanism for the antibacterial action. Asian Journal of Plant Sciences, 8(8): 551-556.
29. Amensour M, Bouhdid S, Fernandez-Lopez J, Idaomar M, Senhaji N S, Abrini J. (2010). Antibacterial activity of extracts of *Myrtus communis* against food-borne pathogenic and spoilage bacteria. International Journal of Food Properties, 13(6): 1215-1224.
30. Ghasemi Pirbalouti A, Jahanbazi P, Enteshari S, Malekpoor F, Hamedi B. (2010). Antimicrobial activity of some Iranian medicinal plants. Archives of Biological Sciences, 62(3): 633-641.
31. Houshmand B, Mortazavi H, Alikhani Y, Abdolsamadi H, AhmadiMotemayel F, ZareMahmoudabadi R. (2011). In Vitro

Evaluation of Antibacterial Effect of *Myrtus* Extract with Different Concentrations on Some Oral Bacteria. Journal of Mashhad Dental School, 35(2): 123-130.
32. Fazeli-nasab B, Moshtaghi N, Forouzandeh M. (2019). Effect of Solvent Extraction on Phenol, Flavonoids and Antioxidant Activity of some Iranian Native Herbs. Scientific Journal of Ilam University of Medical Sciences, 27(3): 14-26 https:doi.org/10.29252/sjimu.27.3.14
33. Fazeli-Nasab B, Rahnama M, Mazarei A. (2017). Correlation between Antioxidant Activity and Antibacterial Activity of Nine Medicinal Plant Extracts. J Mazandaran Univ Med Sci 27(149): 63-78.
34. Kouitcheu L B M, Tamesse J L, Kouam J. (2013). The anti-shigellosis activity of the methanol extract of Picralima nitida on *Shigella dysenteriae* type I induced diarrhoea in rats. BMC Complementary and Alternative Medicine, 13(1): 1-11. https://doi.org/10.1186/1472-6882-13-211
35. Rene K, Hortense G K, Pascal W, Alexis M N J, Vidal P E, Archange F T M, Christine F M. (2007). Activity of aqueous ethanol extract of Euphorbia prostrata ait on *Shigella dysenteriae* type 1-induced diarrhea in rats. Indian Journal of pharmacology, 39(5): 240-244.
36. Khosravi H, Solouki M, Ganjali S. (2020). Investigating Antibacterial Properties of Tecomella undulata and Momordica charantia Plant Extracts on Some Pathogenic Bacteria. Gene, Cell and Tissue, 7(1): e94960. https://dx.doi.org/10.5812/gct.94960
37. Saravani K, Javadian E, Mohsenbeygi M, Hassanshahian M. (2019). Study the Antimicrobial Effects of *Momordica Charantia* on Pathogenic Bacteria. Journal of Medical Bacteriology, 8(5-6): 1-7.