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Evaluation of Antimicrobial Properties of Acalypha Indica Extract Against Gastrointestinal Infection Causing Organisms

[1]Vishnu Priyanka K, [2]Poovendiran P, [3]SudharsanK, [4]SornambigaRavi, [5]Archana S Nair

[1,2,3,4&5]PG Students, Post graduate Research Department of microbiology, Dr.N.G.P. Arts and science College, Coimbatore, Tamilnadu, India.

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ABSTRACT: There is potential for the normal faecal flora of humans to be augmented by resistant strains of bacteria, acquired from food. The frequency of resistance in the aerobic Gram-negative faecal flora is often very high. The purpose of this study was to find out whether food strains contribute to this resistance. Stool samples were studied, five different strains were found. From these samples, E. coli, Pseudomonas spp, Shigella spp, Salmonella spp, Klebsiella sppdifferent strains of bacteria belonging to the family Enterobacteriaceae were isolated. Enterobacter spp. were most frequent, E. coli was rare. Sensitivity testing was undertaken only for isolates with different biotypes and antibiograms. Acalypha indica and commercially available drugs resistance was found in 12% and 14% of isolates, respectively. The only statistically significant differences between the Finnish and imported strains were for these two; the Finnish isolates were more resistant to plant extract, whereas the imported ones were more resistant to commercially available drug. Consequently, bacteria from vegetables are not responsible for the high prevalence of resistant Enterobacteriaceae in faecal flora. They are in fact unusually susceptible to the antibiotics studied. Multi resistance profiles, typical of strains associated with human activities, were not identified in these isolates.

KEYWORDS: Enterobacteriaceae, E. coli, Antimicrobial activity, Acalypha indica.

I. INTRODUCTION

The Enteric diseases are caused by the membersof bacteria family and Vibrionaceae. These pathogens are named as enteric pathogens that belong to the genera that initiate infection by offensive the intestinal epithelial tissue. They are predominantly facultative anaerobic bacterial flora of large intestine of human beings. In most cases, infections of the intestines result in diarrhoea, dysentery, nausea, vomiting, or abdominal cramping. The enteric disease-causing members are mainly E. coli, Shigella spp, Salmonella spp, Pseudomonas spp, Klebsiella pneumonia etc. Gastrointestinal, or stomach disorders are comprised of a variety of ailments that have many different types of symptoms. The gastrointestinal tract, also known as the GI tract or gut, starts at the exophages, and includes the stomach, small intestine (also known as the duodenum), large intestine (also known as the colon), rectum, and anus. The chemical commercial drugs are creating a lot of side effects in human body. The kidney failure due to side effects of chemical medicines is improving. But in fact, even people are aware of this also, they are not at all ready move to natural medicines, because the way and mode of action is very lagging for them in our body. So, the curing time exceeds. All these leads the society to opt chemical medication more than natural medicines. In such a reality the implementation of naturo-chemical drugs may have a significant role. Here they can have the fast mode of action just as a chemical but immunizing activity will be of natural extracts. The health and time of an individual during treatment will be saved. The level of application of the plants and chemicals will be 9:1 and that won ‘t be a marginal level of immunizing for human. The body can quickly accept the medicine and the very minute level of chemical may not make any loss in body. There by plant extracts can refill the new generation ‘s health problems for a huge extend. A vital task of the clinical microbiology laboratory is that the performance of antimicrobial susceptibility testing of significant microorganism isolates. The goals of testing are to discoverattainable drug resistance in common pathogens and to assure susceptibility to medicine of selection for specific infections. The foremost widely used testing methods include broth microdilution or fast automatic instrument

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strategies that use commercially marketed materials and devices. Manual methods that offer flexibility and possible cost savings embrace the disk diffusion and gradient diffusion methods. Every methodology has strengths and weaknesses, including organisms that will be accurately tested by the strategies. Some methods provide quantitative results (example: minimum inhibitory concentration), and all provide qualitative assessments using the categories susceptible, intermediate, or resistant. In general, current testing strategies offercorrect detection of common antimicrobial resistance mechanisms. However, newer or rising mechanisms of resistance need constant vigilance reading to the flexibility of every test method to accurately detect resistance. In this study with a view to establishing the evaluation study of commercial drugs and Acalypha indica extract against gastrointestinal tract infection causing organisms.

II. STUDY OBJECTIVES:

- To isolate and characterize pathogens in infected stoolsample
- To test the antimicrobial activity of commercial drugs against organisms such as E. coli, Pseudomonas spp, Salmonella spp, Shigella spp, and Klebsiellaspp.
- To test antimicrobial activity of plant extract againstmicroorganisms.
- To perform comparative study between commercial drugs and plant extract against microorganisms.
- To perform comparative study between commercial drugs and a mixture of broad-spectrum antibiotic and plant extract againstmicroorganisms.

III. MICROORGANISMS INVOLVED IN GASTROINTESTINAL TRACT INFECTIONS:

- E. coli
- Pseudomonas spp
- Salmonella spp
- Shigella spp
- Klebsiella spp

IV.ANTIMICROBIAL ACTIVITY WITH MEDICINAL PLANT EXTRACTS:

Medicinal plants represent a good supply of antimicrobial natural products. The use of medicinal plants all over the world predates the introduction of antibiotics and other modern drugs into African continent (Haslam et al., 1989). Plants have been used in traditional medicine for many centuries as abortifacients, contraceptives, for menstrual regulation, fertility control, as well for the treatment of ailments of both microbial and non-microbial origins (Gill and Akinwunmi, 1986). The Nigeria flora is wealthy in medicinal plants that are sometimes exploited by herbaldoctors otherwise known as native doctor. The indigenous population in Southwest, Nigeria for example has developed a vast knowledge on the use of plants as traditional remedies (Ekundayo 1986). Some of the plant's collections are used against a variety of diseases such as typhoid fever gastroenteritis, dysentery, malaria and others which are typical diseases of tropical countries (Sofowora 1993; Nick et al., 1995).

V. MATERIALS AND METHODS:

Specimen Collection And Processing:

A total of 5 stool samples were collected from hospital patients who were suffering signs and symptoms of gastrointestinal tract infections attending PSG hospital, Coimbatore from December 2019 to January 2020, and processed for isolation of bacterial pathogens. Freshly voided faecal samples were collected into sterile glass containers and stored aseptically. Following collection, these stool samples were transported immediately to microbiology laboratory for culture and antimicrobial susceptibility testing. All collected samples were identified according to the morphological characteristics, cultural characteristics, and biochemical characteristics.

IDENTIFICATION OF BACTERIAL SPP.:

Identification of bacterial sample is being processed through the below mentioned examinations.

MICROSCOPIC EXAMINATION:

The isolated colonies should be stained by Gram stain to observe their response to stain, shapes and their arrangement.

GRAM STAINING:

It is very useful to identify and classify bacteria into two major groups such as Gram-positive and Gram-negative. To follow the Christian gram (1884) gram staining procedure and microscopic observation was performed.

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CULTURING OF ORGANISMS:

NUTRIENT AGAR:

Nutrient agar is a common medium used for the culturing of all types of organisms. It is basal medium which supplies the basic requirements of carbon, nitrogen and mineral source for growth. Nutrient agar was prepared and plating was done.

EOSIN METHYLENE BLUE:

Eosin methylene blue {EMB} agar is a selective and differential medium used to isolate faecal coliforms such as E. coli. Eosin methylene blue are the pH indicators. Escherichia coli, a full of life fermenter produces a green sheen. Slow or weak fermenters will produce mucoid pink colonies. Colourless colonies indicate that the organism is not a faecal coliform. E. coli was inoculated in this media contained plate.

Salmonella spp andShigella spp:

The selective Medias were prepared such as Salmonella Shigella agar, Deoxycholate citrate agar, Xylose lysine Deoxycholate agar and Salmonella spp and Shigella spp was inoculated to plates contained media.

MACCONKEY AGAR:

MacConkey agar is a selective and differential media used for the isolation and differentiation of non-fastidious gram-negative species, significantly members of the family Enterobacteriaceae and also the genus
Pseudomonas spp. The organism was inoculated.

MUELLER HINTON AGAR (MHA) (pH-7.3):

Mueller Hinton agar medium was used to check the antibacterial activity. Medium was prepared and the antibacterial activity was checked.

ISOLATION OF COLONIES:

The colonies were isolated from reputed selective Medias.

VI.BIOCHEMICALCHARACTERIZATION:

INDOLE PRODUCTION TEST:

Tryptophan is decomposed in to its metabolic products like indole, pyruvic acid and ammonia by the enzymes, tryptophanase. The indole is detected by calorimetric reaction by p – dimethyl amino benzaldehyde (kovac ‘s reagent). The indole was performed and obtained the result.

METHYL RED TEST:

Enteric organisms are oxidizing glucose and the end products vary based on the specific pathway of metabolism. The methyl red test is employed to detect the ability of microorganisms to oxidize glucose. MR test was done and the color formation was observed.

VOGES – PROSKAUER TEST:

The VP test was performed and the result was observed.

CITRATE UTILIZATION TEST:

During this reaction, the medium becomes alkaline as the carbon dioxide combines with sodium and water to form sodium carbonate changes the bromothymol blue from green to deep Prussian blue. A positive tube shows a blue color on the streak of growth. Retention of original green colour and no growth on the line of streak indicates a negative reaction.

HYDROGEN SULPHIDE PRODUCTION:

Hydrogen Sulphide production indicated by a blackening of the butt. This is due to the reaction of hydrogen Sulphide with the ferrous ammonium sulphate to form black ferrous Sulphide.

CATALASE TEST:

Accumulation of hydrogen peroxide and super oxide leads to the death of the organisms unless they are degraded enzymatically, organisms capable of producing catalase or peroxidase. 1ml of 3% hydrogen peroxide was added to the culture after 5 minutes the culture was examined immediately for the evolution of bubbles, which indicates a positive test.

OXIDASE TEST:

During aerobic respiration, oxidase enzymes play a vital role in the operation of electron transport system. Cytochrome oxidase the catalyzes the oxidation of a reduced cytochrome by molecular oxygen, resulting in the formation of water or hydrogen peroxide. This test depends on the presence of certain oxidases in bacteria that will catalyzes the transport of electrons between electron donors in the bacteria and a redox dye – Tetramethyl p-Para phenylene diamine dihydrochloride. The dye is reduced to a deep purple color.

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UREASE TEST:

Urease is an enzyme produced by few microorganisms. Urease is a hydrolytic enzyme that attacks the nitrogen and carbon bond in amide compounds like urea and forms an alkaline end product such as ammonia. A positive urease reaction is indicated by a change in the colour of the medium from yellow to purple color.

VII.ANTIBIOTIC SUSCEPTIBILITYTESTING:

ANTIBIOTIC SUSCEPTIBILITYTESTING WITH COMMERCIALDISCS:

Select the pure cultures from each organism to be tested. Aseptically emulsify a colony from the plate in the sterile saline solution. Mix it thoroughly to confirm that no solid material from the colony is visible in the saline solution. Repeat till the turbidity of the saline solution visually match that of the standard turbidity.Take a sterile swab and dip it into the broth culture oforganism.Gently squeeze the swab against the inside of the tube in order to get rid of excess fluidin theswab.Take a sterile Mueller-Hinton agar (MHA) plate. Use the swab with the test organism to streak an MHA plate for a lawn ofgrowth.Once the streaking is complete, allow the plate to dry for 5minutes.Antibiotic discs ampicillin(10mcg/disc), tetracycline (30mcg/disc), gentamycin (10 mcg/disc), kanamycin (30 mcg/disc), and nitrofurantoin (300mcg/disc)) can be placed on the surface of the agar using sterilizedforceps.Gently press the discs onto the surface of the agar by using flame sterilized forceps or inoculation loop.Carefully invert the inoculated plates and incubate for 24 hours at 37°C.Once incubation, use a metric ruler to measure the diameter of the zone of inhibition for every antibioticused.

ANTIBIOTIC SUSCEPTIBILITY TESTING WITH MEDICINAL PLANT EXTRACT OF Acalypha indica:

PLANT LEAF COLLECTION AND EXTRACTPREPARATION

Plant leaves of Acalypha indica are collected and air-dried in the dark at room temperature (RT) and then grind to powders. Exactly each 10g of dried powdered plant material was macerated separately in 100ml of concentrated acetone, methanol, ethanol, and water in large labeled glass bottles and put in an orbital shaker for 24h at room temperature. The solvent was then removed under reduced pressure in a rotary evaporator at 55°C approximately 50–60 min. The procedure was repeated 4 times. Extracts were first filtered using Whatman No.1 filter papers, filtrates were evaporated to dryness at RT in a steady air current. All dried crude extracts were collected in clean universal bottles and were keep at −20 °C tillneeded for testing. Stock solutions were prepared by dissolving the extracts in 1ml dimethyl Sulphide (DMSO) before use.

The inoculum was evenly spread on the MHA plate from pure broth cultures of bacteria by sterile cotton swab and allowed to dry for 5–8 min. Wells (6 mm in diameter) were punched in the agar using a sterile well cutter and filled with 10µL of each extract (acetone, methanol, ethanol, and water) on inoculated plates. The plates were incubated at 37˚Cfor couple of days.Zone of inhibition should be recorded.

COMPARISON BETWEEN EXTRACT OF ACALYPHA INDICA AND COMMERCIAL DRUGS

The inoculum was evenly spread on the MHA plate from pure broth cultures of bacteria by sterile cotton swab and allowed to dry for 5–8 min.Wells (6 mm in diameter) were punched in the agar using a sterile well cutterFill with 10µL of extracts such as acetone, methanol, ethanol, and water.And antibiotic discs of tetracycline(30mcg/disc), gentamycin(10mcg/disc), kanamycin (30 mcg/disc), ampicillin (10 mcg/disc), and nitrofurantoin (300 mcg/disc) was added.The plates were incubated at 37˚c for 2 days.Zone of inhibition should be recorded.

COMPARISON BETWEEN COMMERCIAL DRUGSAND A MIXTURE OF BROAD-SPECTRUM ANTIBIOTIC AND PLANT EXTRACT:

Inoculum was evenly spread on the MHA plate from pure broth cultures of bacteria by sterile cotton swab and allowed to dry for 5–8 min. Wells (6 mm in diameter) were punched in the agar using a sterile well cutter and filled with a mixture of broad-spectrum antibiotic and plant extract at a concentration of 1: 9. The plates were incubated at 37˚c for 24 hours. Zone of inhibition should be recorded and compared.

VIII.RESULT AND DISCUSSION:

Using bacteriological, Gram staining and biochemical characterization, the result obtained from the data shows that the main bacterial isolates found in the stool samples were Escherichia coli, Salmonella spp., Klebsiella spp., Pseudomonas

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spp., Citrobacter spp., and Shigella spp., with the following prevalence 22%, 19%, 14%, 12%, 14%, 19% respectively. The result of the study implies that Escherichia coli has the best prevalence with 22%, followed by Salmonella spp and Shigellaspp with 19% each, whereas Pseudomonas spp has the smallest occurring with prevalence percentage of 12%. The findings of the research indicated that the enteric bacteria are associated with the diarrheic stool infection.

Identification Of Bacterial Colonies:

Significant bacterial cultures like E. coli, Salmonella spp., Pseudomonas spp., Shigella spp., and Klebsiella spp. were determined with the colony growth. Pure growth of isolates was examined for the following microscopic, culture morphology and cultural characteristics. Bacterial pathogens from stool samples were identified.

GRAM STAINING AND HANGING DROP METHOD:

- E. coli - gram negative rods, motile
- Pseudomonas spp - gram negative rods, non-motile
- Salmonella spp - gram negative rods, motile
- Shigella spp - gram negative rods, non-motile
- Klebsiella spp - gram negative rods, non-motile

CULTURAL CHARACTERISTICS:

Isolation of E. coli colonies:

Grow well on nutrient agar media forming smooth, colorless colonies. It can withstand the temperature with 15-45˚C.On EMB agar plate, a green metallic sheen was observed.

Isolation of Pseudomonasspp colonies:

It forms round colonies with fluorescent greenish color, sweet odor on nutrient agar media. On blood agar it shows **β**- hemolysis. Citrimide agar gave the cultural characteristics with the production of Pseudomonas spp pigments pyocyanin and fluorescein, which show a character of yellow green color.

Isolation of Salmonella spp colonies:

On nutrient media, Salmonella spp isolates were observed as translucent, colorless and smooth round colonies.

On SS agar, Salmonellaspp gives black opaque moist colonies. Dark straw-colored colonies were observed on DCA media and it specifies the presence of Salmonellaspp on the medium.

Isolation of Shigella spp colonies:

Nutrient agar gave a colourless, translucent, rough colonies were observed. On XLD agar medium, dark pink or red colored colonies, with opaque nature were observed. On SS agar, white colored, waxy, pointed transparent colonies were observed.

Isolation of Klebsiellaspp colonies:

On nutrient agar media, moist, white colored, opaque colonies were observed. The colonies of Klebsiella spp on MacConkey agar media were observed as pink colored colonies due to lactose fermentation.

SPECIMEN COLLECTION:

fig1: Stool sample ofinfectedperson**.**

COLLECTION OF LEAF:

fig2: Acalypha indica

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fig3: grinded form of leaves of Acalyphaindica

EXTRACT OREPARATION PROCESS

fig4: ethanol, methanol, water, acetone

fig5: filtering process of water extract

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BIOCHEMICAL ANALYSIS:

After biochemical test, the results for various microorganisms were tabulated.

Table1: Biochemical test results

Organisms	Glucose	Sucrose	Mannitol	Lactose	TSI	Indole	MR	VP	Citrate	Urease	Catalase	Oxidase
E. coli	+	variable	+	+	A/A Gas	+	+	-	-	-	+	-
Pseudomonas spp	+	-	-	-	AK/AK No gas	-	-	-	+	-	+	+
Salmonella spp	+	-	+	-	AK/A Gas H2S	-	+	-	-	-	+	-
Shigella spp	+	-	+	-	AK/A No gas	+	+	-	-	-	+	-
Klebsiella spp	+	+	+	+	A/A Gas	-	-	+	+	+	+	-

ANTIBIOTIC SUSCEPTIBILITY TESTING USING COMMERCIAL DRUGS:

Table2: result for antibiotic susceptibility testing using commercial drugs

S.no	Organisms	Amp (mm)	Gen (mm)	Tet (mm)	Nf (mm)	K (mm)
1	E. coli	7	8	21	22	23
2	Pseudomonass pp	0	17	0	0	0
3	Salmonellaspp	19	24	25	25	26
4	Shigellaspp	21	26	27	25	25
5	Klebsiella spp	0	21	24	20	20

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To check antibiotic susceptibility of isolated organisms, and taken antibiotics such as tetracycline(30mcg/disc), ampicillin(10mcg/disc), gentamycin(10mcg/disc),nitrofurantoin(300mcg/disc) and kanamycin(30mcg/disc),then observed that all the E. coli and Salmonella isolates from stool samples were most susceptible to kanamycin with a diameter of 23mm and 26mm respectively, Pseudomonas isolates were most susceptible to gentamycin with a diameter of 17mm and Shigella and Klebsiella isolates were most susceptible to tetracycline with a diameter of 27mm and 24mm respectively.

Antimicrobial Activity Testing Using Plant Extracts

E. coli and Salmonella spp, having highest susceptibility to methanol extract with a diameter of 17mm and 19mm respectively. Pseudomonas spphaving high susceptibility to acetone extract with a diameter of 18mm. Klebsiella spp having 23mm diameter of inhibition zone on ethanol extract and that shows as most susceptible. Shigella spphaving highest susceptibility to both methanol and ethanol extracts with a diameter of 23mm and 26mm.

Table 3: antimicrobial activity testing using plant extracts

S. No	Organisms	Meth(mm)	Eth(mm)	Wat(mm)	Ace(mm)
1	E. coli	17	9	2	22
2	Pseudomonas spp	16	18	0	18
3	Salmonella spp	19	26	0	28
4	Shigella spp	23	26	0	22
5	Klebsiella spp	7	23	3	24

COMPARISON BETWEEN PLANT EXTRACT AND COMMERCIAL DRUGS:

Table 4: comparison between plant extract and commercial drugs

S.no	Organisms	Plant extracts (in µL)				Commercial drugs (in mcg)				
		Meth	Eth	Wat	Ace	Amp	Gen	Tet	Nf	K
1	E. coli	17	9	2	22	7	8	21	22	23
2	Pseudomonas spp	16	18	0	18	0	17	0	0	0
3	Salmonella spp	19	26	0	28	19	24	25	25	26
4	Shigella spp	23	26	0	22	21	26	27	25	25
5	Klebsiella spp	7	23	3	24	0	21	24	20	20

In this study theplant extracts are more active on microorganisms than on commercially available drugs. The comparison of given table leads us the result that obviously shows higher zone of inhibition of plant extraction. The above table shows that.

COMPARISION BETWEEN COMMERCIAL DRUGS AND A MIXTURE OF BROAD SPECTRUS ANTIBIOTIC AND PLANT EXTRACT:

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Table 5: comparison between commercial drugs and a mixture of broad-spectrum antibiotic and plant extract

S.no	Organisms	Extracts + Gentamycin (in µL)				Commercial drugs (in mcg)				
		Met	Eth	Wat	Ace	Amp	Gen	Tet	Nf	K
1	E. coli	14	12	6	9	7	8	21	22	23
2	Pseudomonas spp	15	11	6	22	0	17	0	0	0
3	Salmonella spp	11	19	4	13	19	24	25	25	26
4	Shigella spp	12	20	19	8	21	26	27	25	25
5	Klebsiella spp	11	17	17	13	0	21	24	20	20

According to the comparative study of chemical medicines are less effective compare to herbal medicine. To that the introduction of naturo-chemical medicines may give an evolved change in medicinal habits of people. Thereby the side effects can be nullified for the maximum extend. We had taken the antimicrobial activity of chemical drugs and plant extracts. Then compared each of them and analyzed that chemical drugs are leading far better than antimicrobial activity of plant extract. After that 90% plant extract with 10% of chemical drugs and through this research found that the antimicrobial activity of the new mixture had a greater improvement. Through this study able to summarize that the minor mixing up of chemical drugs with maximum quantification of plant extract can nullify the chemical side effects of chemicals and could give a better improvement in killing the microbes and curing of disease. This can preserve the health of each individual from side effects causing chemicals.

XI.CONCLUSION:

The mode of pure medication is only been happened through natural plants and extractions. In the conclusion naturo-chemical medicines which are having 90% natural extracts and 10% chemicals. The chemicals along with the extracts only for the fast mode of action. The minor quantity of chemicals will be nullified in their action of side effects and should pointed to the only level of catalyzing the treatment. The plant extracts will have a highest mode of action with the help of chemical drugs will be able to act with higher frequency without any side effects. Along with that natural extracts are being quicker in their action. The people when become free from side effects of kidney and brain will be able to work much better for the well-being of nation.

SOME OF THE ADVANAGES FROM THE ABOVE RESULTS

a) **Reduced risk of side effects:** well tolerated by the patient, with fewer unintended consequences than pharmaceutical drugs.

b) **Effectives with chronic conditions:** Herbal medicines tend to be more effective for long-standing health complaints that don't respond well to traditional medicine.

c) **Lower cost:** Another advantage to herbal medicine is cost. Herbs cost much less than prescription medications. Research, testing, and marketing add considerably to the cost of prescription medicines.

d) **Widespread availability:** Yet another advantage of herbal medicines is their availability. Herbs are available without a prescription.

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REFERENCE:

[1]. Abdulsalam A., Alhendi B., 1999. Nasal microflora of camels (Camelus dromedarius) under two different conditions. Pakistan Veterinary Journal, 19(4):164-167.

[2]. Adak, G. K, Long S. M. and O`Brien, S.J.

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(2002). Trends in indigenous food borne disease and deaths, England and Wales, 1992-2000. Gut, 51:832-841.

[3]. Adesiyun AA, Dasuki MO and Ibrahim GA (1988). Occurrence and antibiograms of Salmonellae isolated from slaughter sheep and goats in Zaria, Nigeria and Israel. Journal of Veterinary Medicine 44:248-255.

[4]. Al-Muhannak FHN. Spread of some extended spectrum beta lactamases in clinical isolates of Gram-negative bacilli in Najaf. M.Sc. Thesis College of Science, University of Kufa; 2010.

[5]. Al-Tarazi Y.H., 2001. Bacteriological and Pathological Study on Pneumonia in the One-humped Camel (Camelus dromedarius) in Jordan. Revue Élev. Méd. Vét. Pays trop., 54(2):93-97

[6]. Angulo FJ, Tippen S, Sharp DJ, et al. A community waterborne outbreak of salmonellosis and the effectiveness of a boil water order. Am J Public Health **1997**; 87:5804.

[7]. Anyinam C 1995. Ecology and ethnomedicine: Exploring links between current environmental crisis and indigenous medical practices. Soc Sci Med, 4: 321–329.

[8]. Arora R., Kalra D.S., 1973. A note on isolation of Klebsiella peumoniae and diplococci from cases of broncho-pneumonia in camels. Indian J. Anim. Sci.43:1095-1096.

[9]. Arshad, R., Farooq, S. and Shahid-Ali, S. 2006. Manipulation of different media and methods for cost-effective characterization of Escherichia coli strains collected from different habitats. Pakistan Journal of Botany 38: 779-789.Blacklow NR, Greenberg HB. Vira gastroenteritis. New Engl J Med 1991; 325: 252–264.

[10]. Boeckxstaens G, Hashem B, André J, Peter J (2014) Symptomatic reflux disease: The present, the past and the future. Gut 63: 1185-193.

[11]. Bonacorsi S, Bingen E. Molecular epidemiology of Escherichia coli causing neonatal meningitis. Int J Med Microbiol 2005; 295: 373-38.

[12]. Boyce TG, Koo D, Swerdlow D, et al. Recurrent outbreaks of Salmonella Enteritidis infections in a Texas restaurant: phage type 4 arrives in the United States. Epidemiol Infect **1996**; 117:29– 34.

[13]. Brisse, S., F. Grimont, and P.A. Grimont, the genus Klebsiella, in The Prokaryotes. 2006, Springer. p.159-196.

[14]. Buxton A and Fraser G (1977). Animal Microbiology. Blackwell Scientific Publications, Oxford.Vol.1:85-110.

[15]. Cakilcioglu U, Turkoglu I 2010. An ethnobotanical survey of medicinal plants in Sivrice (Elazig- Turkey). J Ethnopharmacol, 132: 165-175.

[16]. Carter M.E., Chengappa M.M., 1990. Enterobacteria, In: Diagnostic Procedures in Veterinary Bacteriology and Mycology, Carter, G.R., Cole Jr, J.R., 5th Edition, Academic Press.Boyce TG, Koo D, Swerdlow D, et al. Recurrent outbreaks of Salmonella Enteritidis infections in a Texas restaurant: phage type 4 arrives in the United States. Epidemiol Infect **1996**; 117:29– 34.

[17]. Boyce TG, Koo D, Swerdlow D, et al. Recurrent outbreaks of Salmonella Enteritidis infections in a Texas restaurant: phage type 4 arrives in the United States. Epidemiol Infect **1996**; 117:29– 34.

[18]. Brisse, S., F. Grimont, and P.A. Grimont, the genus Klebsiella, in The Prokaryotes. 2006, Springer. p.159-196.

[19]. Buxton A and Fraser G (1977). Animal Microbiology. Blackwell Scientific Publications, Oxford.Vol.1:85-110.

[20]. Cakilcioglu U, Turkoglu I 2010. An ethnobotanical survey of medicinal plants in Sivrice (Elazig- Turkey). J Ethnopharmacol, 132: 165-175.

[21]. Carter M.E., Chengappa M.M., 1990. Enterobacteria, In: Diagnostic Procedures in Veterinary Bacteriology and Mycology, Carter, G.R., Cole Jr, J.R., 5th Edition, Academic Press.

[22]. CDC. 2008. Escherichia coli 0157:H7 infections in children associated with raw milk and raw colostrum from cows--California. Morbidity and Mortality Weekly Report 57(23): 625-8.

[23]. Chalker RB, Blaser MJ. A review of human salmonellosis. III. Magnitude of Salmonella infection in the United States. Rev Infect Dis **1988**; 10:111–24.

[24]. Chompook P. (2011) Shigellosis. Encyclopedia of EnvironmentalHealth.pp.26-32

[25]. Cohen ML, Tauxe RV. Drug-resistant Salmonella in the United States: an epidemiologic perspective. Science**1986**; 234:964–9.

[26]. Deshmukh, A.S, Karpe, A.G. 2006. In vitro transfer of plasmids in E. coli. IndianJournal ofComparative Microbiology Immunology and Infectious Disease, 27: 32 -34.
[27]. Dogan Y 2012. Traditionally used wild edible greens in the Aegean Region of Turkey. Acta Soc Bot Pol, 81(4): 245-255.
[28]. Dogan Y, Baslar S, Ay G, Aydin H, Yorek N, Mert HH 2005. Poisonous plants distributed naturally in Turkey. Pharmacia, 52: 50-55.
[29]. Dogan Y, Baslar S, Celik A, Mert HH, Ozturk M 2004b. A study of the roadside plants of West Anatolia, Turkey. Nat Croat, 13(1): 63-80.
[30]. Dogan, Y., S. Baslar S, H. H. Mert HH, and G Ay G 2003. Plants used as natural dye sources in Turkey. Econ Bot, 57(4): 442-453.
[31]. Dogan Y, Mert HH 1998. An autecological Study on the Vitex agnus-castus L. (Verbenaceae) distributed in West Anatolia. Turk J Bot, 22: 327-334.
[32]. Dogan Y, Nedelcheva AM, Obratov-Petkovic D, Padure IM 2008. Plants used in traditional handicrafts in several Balkan countries. Indian J Tradit Know, 7 (1): 157-161.
[33]. Dogan Y, Ugulu I, Baslar S 2010b. Turkish red pine as a bio monitor: A comparative study of the accumulation of trace elements in needles and barks. Ekoloji, 19 (75): 88-96.