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Antimicrobial Activities of *Psidium guajava, Citrus lemon, Alium sativum, Azadiracha indica* and *Pondias mombin* in the South–Eastern Nigerian against *Aeromonas hydrophila* Isolate from *Clarias gariepinus* (Catfish)

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Abstract: *Aeromonas hydrophila*is a heterotrophic, gram-negative, rod-shaped pathogen (bacterium), mostly found in areas with a warm climate. An investigation into the antimicrobial activity of some medicinal plants in the South-Eastern Nigerian against *Aeromonas hydrophila* isolated from catfish. A total of 28 strains of *A. hydrophila*were isolated from thirty catfish samples from a fish farm in Abakaliki, Ebonyi State and the antibacterial activities of crude extracts of *Psidium guajava*, *Citrus lemon*, *Allium sativum*, *Azadiracha indica* and *Pondias mombin*against the strains studied. Results showed that the extracts of *Spondias mombin* displayed a high level of antibacterial activity, followed by *Psidium guajava, Allium sativum* and *Azadiracha indica*respectively in a descending order against all the twenty eight strains. More so, the extracts at 100% concentration were shown to be more effective than most conventional antibiotics used in the study. However, extract of*citrus lemon* did not show any antibacterial effect against any of the isolates. Introducing the leasves of *Spondias mombin*, *Psidium guajava, Allium sativum*and *Azadiracha indica*into fish ponds could therefore, be suggested to help curtail the harmful effects of *A. hydrophila*in fishes produced in such ponds.

Key words: Plant Extracts · African Catfish · *Aeromonas hydrophila* · Antibacterial Activity

INTRODUCTION

Fish parasitismare common natural occurrences that could be either internal (endoparasites) or external (ectoparasites). *Aeromonas sp*. is a group of microorganisms commonly found in aquatic environments [1, 2]. *Aeromonas hydrophila* are examples of internal fish parasitewhich affect the ovary of female catfish [3]. The adult female parasite is about 40 centimetres in length, 1.6 mm diameter and red in colour while the males are tiny. Fish pathogens like *Aeromonas hydrophila* are widely distributed in nature, leading to a lot of morbidity as well as mortality [4].

These microorganisms have wide host selection and have frequently been isolated from people with diarrhoea [5]. Crude extractsof different species ofplants have been utilized for therapeutic purposes for hundreds of years; many plants now used as culinary herbs and spices have been used as medicines from prehistoric times. Some plants (spices) have been used partly to counter food spoilage bacteria, especially in hot climates [6-8]. These plants that are used as medicine are cheaper and most are home grown and could be gotten at no cost. Moreover, pharmaceutical companies have exploited the knowledge of herbal plants of indigenous peoples across the globe in the research to create new drugs [9]. In India, where Ayurveda (tradition of alternative medicine) has been observed for a very long time, the use of medicinal plants to cure illness are championed by the government [10, 11]. Most developed countries screen these plants using bioassay-guided fractionation to isolate active particles that, in most cases, are developed into different medicinal active products that are used either as the natural products or a synthetic modification or a

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synthesised analogue with added medicinal action or reduced adverse side effects [10]. The use of medicinal plants in the treatment of diseases are seen by some as a preferable when compared to pure industrial medical compounds produced [5, 12].

Presently, more than eighty percent of the world population depend on medicinal plants and herbs as their source of medication in meeting their different medication need. These medicinal plants and herbs are believed to be safer and have proved to be effective against specific ailments [13]. They are extensively used in African and other developing countries of the world. However, in recent time,the use of spices/herbs is gradually gaining ground in developed countries too, owing totheir obvious beneficial effects. In this study, we investigated the antibacterial efficacy of the crude extractsof five different well-known medicinal plants in the South-Eastern Nigeria (*Psidium guajava, Citrus lemon, Allium sativum, Azadiracha indica* and *Spondias mombin)* against twenty-eight distinct strains of *Aeromonas hydrophila* isolates from *Clarias gariepinus*.

MATERIALS AND METHODS

Sample (Fish) Collection and Processing: A total of thirty (30) catfish samples including(15 males and 15 females) were purchased from the Chi-boy farms in Abakaliki, Ebonyi state andwere taken in a sterile container to the microbiology laboratory of Ebonyi State University, Abakaliki. Each of the fish samples were dissected using a surgical blade and samplecollected from the intestinal tracts. About 2g each were weighed, ground in sterile mortar and transferred into 5 test tubes. Thereafter, 10ml of alkaline peptone water was added into each of the test tubes and incubated at 37°C for 24h to get the stock culture.

Serial Dilution and Isolation of the *Aeromonas Hydrophila*: Exactly10^3 serial dilution was used duringthe plating on the *aeronomas* agar base media. Distilled water (0.9ml) was transferred into 3 sterile test tubes and 0.1ml of stock culture added to the first test tube. After this, 0.1ml was transferred from the first test tube to the second test tube and down to the third test tube. Finally, 0.1ml aliquot was aseptically drawn from the third test tube ($10^{\wedge 3}$) of each sample and inoculated on clean sterile petri dishes containing *Aeromonas* agar base media using spread plate method. The plates were incubated at 37°C for 24hours and culture observed for yellow colonies.

Purification of the Mixed Culture: The yellow colonies obtained from the initial culture, were sub-cultured on nutrient agar using streak plate method and later sub-cultured back on the *aeromonas* agar base selective media.

Agar Haemolysis Test: Blood agar haemolysis test was also carried out on the pure isolate to assay for the presence of haemolysin gene.

Preparation of Plant Extracts: The fresh samples of the medicinal plants were sourced within Abakaliki and washed withdistilled water. About 120g of theplant leaves wereground and extracted using ethanol and hot water respectively and then filtered using membrane filtration (0.45 micron sterile filter). Three different concentrations: 50%, 75% and 25% were made by diluting the 100% concentrated extract with right volumes of sterile water.

Garlic extract was prepared in an alternative manner due tosome difficulties of filtering the crushed materials. About 100 grams of the cleaned garlic was taken and sterilized using ethanol. A laminar flow chamber was used to evaporate the ethanoland the garlic homogenised aseptically using a sterile mortar and pestle. The extract was aseptically squashed out using sterile cheesecloth.

Antibacterial Activity Testing Using Agar Well Method: The *A. hydrophila* strains were inoculated in 10 mlof sterile nutrient broth and incubated at 37°C for 16-18 hours. A sterile cotton swab was used to swab the surface area of sterile nutrient agar plates and agar wells were done with the aid of sterilizedcorkborer of 10mm diameter. A micropipette was used to transfer dilutions (100%, 75 %, 50 % and 25%) into the various wells in the plate. The plates were then incubated in an erect position at 37°C for 25hrs. The diameter of inhibition zones was assessed in mm and the outcome noted. The inhibition zones with diameter under 12mm were noted as having zero antibacterial activity.

Antibacterial Sensitivity Testing Using Filter Paper Method: Filter paper discs of seven mm diameter were prepared and sterilised. Forceps that were sterilised using ethanol and flame were used to place the discs on the nutrient agar plates inoculated with the *A. hydrophila* strains in aseptic conditions[4]. One 100 microliters of the different plant extracts (100%, 75 %, 50 % and 25%) was aseptically transferred to the discs and the plates were then incubated in an erect position at 37C for 24 hours. The diameter of inhibition zones was assessed in mm and

the result noted. Inhibition zones with diameter under twelve mm were noted as having zero antibacterial activity while those> 16mm were notedas extremely active.

Antibiotic Sensitivity Testing: The *A. hydrophila* test microorganisms were further analysed for their sensitivity to the conventional antibiotics such asampicillin, augmentin, gentamycin, pefloxacin, tarivid, streptolysin, septrin, chloranphenicol, sparfloxacin, ciprofloxacin and amoxicillin by the disk diffusion technique (3). The *A. hydrophila* strains were inoculated in 10 ml of sterile nutrient broth and incubated at 37°C for 16-18 hours. A sterile cotton swab was used to swab the surface area of sterile Aeromonas Agar Base (AAB) plates. Forceps that were sterilised using ethanol and flame were used to place the discs on the nutrient agar plates inoculated with the *A. hydrophila* strains in aseptic conditions and adequately separated from one another to avoid overlapping of the inhibition zones. The diameter of the inhibition zones was assessed in mm. The AAB selective media used in this study was obtained from ZigmaAndrich USA.

RESULTS

Microorganism: Twenty-eight strains of pathogenic *Aeromonas hydrophila* both from male and female African catfish were isolated in this study. The pathogenic strains were: AH01, AH02, AH03, AH04, AH05, AH06, AH07, AH08, AH09, AH10, AH11, AH12, AH13, AH14, AH15, AH16, AH17, AH18, AH19, AH20 AH21 AH22, AH23 AH24, AH25, AH26, AH27 and AH28.

THE ANTIBACTERIAL ACTIVITY OF THE PLANT EXTRACTS (*Azadiracha indica, Allium sativum* and *Citrus lemon)* against different strains of of *A. hydrophila* Isolated from Male and Female Africancatfish: *A. hydrophila* responded differently on the *Azadiracha indica* extract at quite different levels. At 100% concentration, *A. hydrophila* strain AH1 and AH22were extremely sensitive (30mm and 29mm) respectively, followed by AH8, AH16 and AH27 (28mm, 28mm and 27mm respectively). Ah28 and serogroup AH4 was the least susceptible (20 mm). *Azadiracha indica* extract showed higher antibacterial activity at 100 % and 75% concentrations. However, 50 % concentration of the extract showed moderate antibacterial activity against the strains and none at 25%. Nevertheless, *Azadiracha indica* extract was able to inhibit the growth of several of the strains at various concentrations.*Allium sativum* extract at levels (100%, 75%, 50 % and 25%) was linear. Nevertheless, AH26 and AH27 were not inhibited by *Allium sativum* extract at this particular concentration. AH12 and AH2 had moderate sensitivity of 13mm and 12mm respectively at 75% concentration. For *Citrus lemon* extracts, all the *A. hydrophila* strains zero inhibition at different concentrations hence failed to prevent the growth of all of the test organisms as shown in Table 1.

The Antibacterial Activity of the Plant Extracts (*Spondia Smombin* and *Psidium guava)* Against Different Strains of *A. hydrophila* Isolated from Male and Female African Catfish: The antibacterial impact of *Spondias mombin* extract against strains of *A. hydrophila* showed that majority of them was highly sensitive to the extract at 100% and 75% concentrations (Table 2). At 25% concentration, the extract didn't show anyantibacterial activity. While the AH26 was extremely sensitive to 100% concentration of the *Spondias mombin* extract by 30mm diameter of inhibition, AH12 wasn't extremely affected. The diameter of inhibition zone was high against *Aeromonas hydrophila* serogroup AH26. The growth of serogroups AH30, AH28, AH22 and AH24 was reasonably inhibited at 100% concentration of the extract.The inhibition zone obtained for the *Aeromonas hydrophila* sero groups AH30 and AH16 was 24mm and 23mm respectively at 75% concentration, 20mm and 15mm at 50% concentration; 9mm and 7mm at 25 % concentration and the antibacterial impact of *Psidium guava* extract against strains of *A. hydrophila*showed that most *spp*. of *A. hydrophila*were highly sensitive to the extract at hundred % and also seventy five % concentrations. At 25 % concentration, the extract didn't show anyantibacterial effect, while the AH5 was highly sensitive to 100% concentration of the *Psidium guava* extract by 20mm diameter of inhibition, AH9 wasnotmuch affected. The diameter of inhibition zone was high against *A. hydrophila*serogroup AH1, AH5, AH19,AH11,AH12 and AH18. The growth of serogroups AH2, AH3, AH27 and AH9 was reasonably inhibited at100% concentration of the *Psidium guava* extract.

The Inhibitory Effect of the Plant Extracts When Compared with That of 10 Conventional Antibiotics (Ampicillin, Augmentin, Gentamycin, Pefloxacin, Tarivid, Streptolysin, Septrin, Chloranphenicol, Sparfloxacin, Ciprofloxacin and Amoxicillin): The diameter of the inhibition zone gotten from different plant extracts at 100 % concentration was greater than most of

Table 1: Antibacterial activity of different concentrations of *Azadiracha indica, Allium sativum* and Citrus lemon extracts

	Diameter of inhibition zone (in mm) against various concentrations of plant extracts											
	Azadiracha indica extract				*Allium sativum* extract				*Citrus lemon* extract			
A. hydrophila												
strains	100%	75%	50%	25%	100%	75%	50%	25%	100%	75%	50%	25%
AH1	30	25	12	3	18	10	3	0	0	0	0	0
AH2	24	22	10	0	17	12	0	0	0	0	0	0
AH3	23	21	15	5	20	6	2	0	0	0	0	0
AH4	20	19	14	7	16	0	0	0	0	0	0	0
AH5	23	21	15	0	16	13	0	0	0	0	0	0
AH6	25	22	17	0	17	10	0	0	0	0	0	0
AH7	26	23	15	8	20	0	2	0	0	0	0	0
AH8	27	24	18	9	14	0	2	0	0	0	0	0
AH9	23	21	17	0	16	10	0	0	0	0	2	0
AH10	25	23	14	0	22	6	4	0	0	0	0	0
AH11	23	22	18	0	20	5	0	0	0	0	0	0
AH12	28	21	15	0	22	13	0	2	0	0	0	0
AH13	24	21	15	2	16	10	0	0	0	0	0	0
AH14	29	24	16	4	18	10	3	0	0	0	2	0
AH15	25	20	15	0	18	0	5	0	0	0	0	0
AH16	26	22	15	0	17	12	0	0	0	0	0	0
AH17	27	22	17	0	19	8	0	0	0	0	0	0
AH18	22	20	16	4	18	0	2	5	0	0	0	0
AH19	28	19	15	0	21	0	0	0	0	0	0	2
AH20	25	22	18	3	20	0	4	0	0	0	0	0
AH21	28	24	15	5	16	0	2	0	0	0	0	0
AH22	29	25	18	0	14	7	0	0	0	0	0	0
AH23	23	20	17	0	12	4	0	0	0	0	0	1
AH24	27	24	16	0	16	10	0	0	0	0	0	0
AH25	29	24	15	2	14	10	0	3	0	0	0	0
AH26	27	23	16	0	0	5	2	0	0	0	0	0
AH27	28	22	17	0	0	0	0	0	0	0	0	0
AH28	20	19	17	3	12	0	3	0	0	0	0	0
AH29	27	20	15	2	18	10	0	0	0	0	0	0
AH30	23	20	18	0	13	9	2	0	0	0	0	0

Less than (<) 12 mm means resistance to the plant extracts and greater than (>) 12 mm means susceptibility to the plant extracts.

the ones obtained against the conventionalantibiotics. *Psidium guajava, Allium sativum, Azadiracha indica* and *Spondias mombin*) extract showed larger diameter of inhibition zones than gentamycin, pefloxacin, streptolysin, septrin, ciprofloxacin and amoxicillin antibiotics respectively as shown in Table 3 when compared with that of the 100% plant extracts in Table 1 and Table 2.

DISCUSSION

Total of five plants were analysed, four (*Psidium guajava*, *Allium sativum*,*Citrus lemon, Azadiracha indica* and *Spondias mombin*) of them showed antibacterial activity. The results of the antibacterial activity of the strains of *A. hydrophila* isolated from male as well as female African catfish samples were as shown in tables one and two respectively.

*Azadiracha indica*extract showed very high antibacterial activity atvarious concentrations (100 %, 75 %, 50 % and 25%). The activity was a linear function of concentrations, various strains of the *A. hydrophila* responded differently on the *Azadiracha indica* extract at quite different levels. At 100% concentration, *A. hydrophila* strain AH1 and AH22 were extremely sensitive (30mm and 29mm) respectively, followed by AH8, AH16 and AH27 (28mm, 28mm and 27mm respectively). Ah28 and serogroup AH4 was the least susceptible (20 mm). *Azadiracha indica* extract showed higher antibacterial activity at 100 % and 75% concentrations. However, 50 % concentration of the extract showed moderate antibacterial activity against the strains and none at 25%. Nevertheless, *Azadiracha indica* extract was able to inhibit the growth of several of the strains at various concentrations. Thoughthese observations are not in line with those of earlier researchers [1, 12, 14, 15] who reported that

Table 2: The Antibacterial Activity of the Plant Extracts (*Spondia Smombin* and *Psidium Guava)* Against Different Strains of *A. hydrophila* Isolated From Male and Female African Catfish

A. hydrophila strains	Diameter of inhibition zone (in mm) against various concentrations of speces extract							
	Spondias mom extract				*Psidium guaja* extract			
	100%	75%	50%	25%	100%	75%	50%	25%
AH1	22	21	14	10	20	15	5	0
AH2	21	20	13	0	18	14	2	0
AH3	21	20	14	0	19	11	0	0
AH4	25	22	16	9	20	12	0	0
AH5	22	20	15	10	20	12	3	0
AH6	23	21	14	0	18	10	4	0
AH7	28	22	19	10	17	14	0	0
AH8	22	21	20	12	15	10	0	0
AH9	21	20	18	10	18	17	0	0
AH10	20	19	16	10	21	14	0	0
AH11	22	20	19	9	20	10	0	0
AH12	17	15	14	0	20	15	4	0
AH13	19	16	13	0	15	12	3	0
AH14	24	20	18	8	17	15	0	0
AH15	23	22	18	10	16	12	0	0
AH16	28	24	20	7	19	15	0	0
AH17	21	20	19	0	16	13	0	0
AH18	22	21	17	0	20	9	0	0
AH19	24	20	19	0	20	12	0	0
AH20	24	21	18	6	19	10	0	0
AH21	25	20	16	5	19	11	0	0
AH22	23	20	18	9	0	0	0	0
AH23	24	21	16	7	0	0	0	0
AH24	25	22	18	0	15	12	5	0
AH25	29	23	17	0	16	11	0	0
AH26	30	25	19	10	19	10	0	0
AH27	21	20	17	8	15	10	0	0
AH28	23	20	16	0	14	8	0	0
AH29	25	22	18	0	14	12	0	0
AH30	25	23	15	9	17	10	0	0

Less than (<) 12 mm means resistance to the plant extracts and greater than (>) 12 mm means susceptibility to the plant extracts.

Table 3: The Inhibitory Effect of the Plant Extracts When Compared with That of 10 Conventional Antibiotics (Ampicillin, Augmentin, Gentamycin, Pefloxacin, Tarivid, Streptolysin, Septrin, Chloranphenicol, Sparfloxacin, Ciprofloxacin And Amoxicillin).

A. hydrophila strains	Antibiotics	Antimicrobial resistance	Antimicrobial susceptibility	Zone of inhibition in diameter
AH28,AH27,AH15	Au(25µg)	ApSp	AmGe	14
AH20,AH19	Ge(10µg)	ApCh	AmGePe	15
AH3,AH5,AH10	Pe(10µg)	ApChSp	AmPeSt	21
AH1,AH12,AH16	Ta(30µg)	ApTaSp	AmStCi	20
AH14,AH11	St(30µg)	ApSp	AmCi	16
AH8,AH9,AH13,AH17	Se(30µg)	ApChSp	AmStCi	18
AH4,AH6	Ch(30µg)	ApAuTa	AmGeSt	17
AH2	Sp(10µg)	ApSp	AmStCi	16
AH22,AH24,AH26	Ci(10µg)	ApCh	AmCiPe	19
AH25,AH23,AH21,AH1	Am(30µg)	ApSpTaAu	AmStGePe	17

Test for ampicillin(Ap), augmentin(Au), gentamycin(Ge), pefloxacin(Pe), tarivid(Ta), streptolysin(St), septrin(Se), chloranphenicol(Ch), sparfloxacin(Sp), ciprofloxacin(Ci) and amoxycillin(Am).

Less than (<) 12 mm means resistance to the antibiotics and greater than(>)12 mm means susceptibility to the antiobiotics

Azadiracha indica showed no significant antimicrobial activity, howeverour results were much like those of Sumner and Janda and Abbot [8, 9] who reported decreased activity of *Azadiracha indica* extract on strains of *A. hydrophila* when the concentration of the extract decreases. In addition, it was recently comfirmed by theIndu *et al.* [4] that *Azadiracha indica*has high antimicrobial activity at high concentration.Furthermore, it was shown that at 100% concentrations of crude extract of *Azadiracha indica* it showed higher antimicrobial activity more the ten different conventional antibiotics and finding s of Thongson *et al.* [16] who revealed that the neem (*Azadiracha indica)* leaf extract significantly reduces the bacterial pathogens and their infection in marine ornamental fishes and neem (*Azadiracha indica*) has found enormous medicinal applications making it a green treasure [17].

The antibacterial activity of *Allium sativum* extract is actually due to the activity of allicin or maybe diallylthiosulphinic acid [2]. In this study, *Allium sativum* extract showed reasonable antibacterial activity at various levels (100 %, 75%, 50 % and 25%) to the strains of *A. hydrophila* and the activity was a linear function of the concentrations.Various strains of the *A. hydrophila* responded in different ways to the *Allium sativum* extract at quite different levels of dilution. At 100 % concentrations *A. hydrophila* strain AH7 and AHI0 were extremely sensitive (22mm) and (20mm) respectively, this supports the report of Indu *et al.* [4] who found out that antibacterial activity of *Allium sativum* extract at levels (100%, 75%, 50 % and 25%) was linear however AH26 and AH27 were not inhibited by *Allium sativum* extract at this particular concentration; AH12 and AH2 had moderate sensitivity of 13mm and 12mm respectively at 75% concentration which is in line with the report of Thongson *et al.* [16] who listed *Allium sativum* as among many medicinal plants that possessed antimicrobial activities. Itis postulated that antifungal and antibacterial properties of *Allium sativum* extract are actually due to the inhibition of succinic dehydrogenase through the inactivation of thiol group [4]. The results revealed variations in the sensitivities of various strains of *A. hydrophila* to *Allium sativum* extract, hinting that systems of resistance are actually building in this specific organism. Garlic could be utilized as a powerful inhibitor of pathogens. Taking of garlic *(Allium sativum*) would boost the shelf life and also reduce the options of spoilage and food poisoning in prepared foods [4].

Citrus lemon extracts at different concentrations failed to prevent the growth of almost the test organisms. The differences might be due to a positive change in the assortment of the citrus lemon applied to this study. It is worthy to note that, very little literature on the antibacterial activity of citrus lemon is actually available 4]. ean while *Citrous lemon* was not included in the work of Thongson *et al.* [16] who reviewed the different types of medicinal plants that have been shown by different research works so far and this was further confirmed by theIndu *et al.* [4] who also reported that *Citrouslemon*has zero effect on the fish species he researched on.

The results of the antibacterial impact of *Spondias mombin* extract against strains of *A. hydrophila* showed that majority of them were highly sensitive to the extract at 100% and 75% concentrations (Table 2). At 25% concentration, the extract didn't show anyantibacterial activity. While the AH26 was extremely sensitive to 100% concentration of the *Spondias mombin* extract by 30mm diameter of inhibition, AH12 wasn't extremely affected. The diameter of inhibition zone was high against *Aeromonas hydrophila*serogroup AH26. The growth of serogroups AH30, AH28, AH22 and AH24 was reasonably inhibited at 100% concentration of the extract.The inhibition zone obtained for the *Aeromonas hydrophila*serogroups AH30 and AH16 was 24mm and 23mm respectively at 75% concentration, 20mm and 15mm at 50% concentration; 9mm and 7mm at 25 % concentration. These observations proved that antibacterial impact of *Spondias mombin* decreases with concentration in line with the report of Indu *et al.* [4]. The inhibitory impact on*Aeromonas hydrophila* was observed at 100%, 75% and 50 % concentrations. However, no activity was observed at 25% concentrations. Outcomes of the antibacterial activities of*Spondias mombin* extract showed fascinating observations. Although the antibacterial activity of this extract on the growth of *A. hydrophila*serogroups was reduced in comparison with *Azadiracha indica* extract, it'd considerable inhibitory impact on all of the strains of *A.hydrophila* under research at hundred %, seventy five % and also fifty % concentrations and in support ofour result, the findings of De, Krishna Deand Banerjee [18] suggested that *Spondias mombin* contain some compounds that can enhance the effect of conventional antibiotics and antifungal agents in inhibiting the growth of pathogens.

Results of the antibacterial impact of *Psidium guava* extract against strains of *A. hydrophila* showed that most *spp*. of *A. hydrophila*were highly sensitive to the extract at hundred % and also seventy five % concentrations (Table 2). At 25 % concentration, the extract didn't show anyantibacterial effect, thereby supporting the reportof Cohen [19] whose study suggests that *P. guajava* leaf extract has the potential to control fish diseases caused by A. hydrophila.While the AH5 was highly sensitive to 100% concentration of the *Psidium guava* extract by 20mm diameter of inhibition of AH9 wasnotmuch affected that of *A. hydrophila* serogroup AH1, AH5, AH19, AH11, AH12 and AH18 showed high effect (Table 2) this observation is similar to the earlier report ofIndu *et al*. [4]. The growth of serogroups AH2, AH3, AH27 and AH9 was reasonably inhibited at100% concentration of the *Psidium guava* extract. However, *P. guava* extract couldn't prevent the growth of AH23 and AH22 at100% concentration and these observations also supported the report of Indu *et al.* [4].

The diameter of the inhibition zone gotten from different plant extracts at 100 % concentration was greater than almost those obtained against the antibiotics (Tables 3).

Psidium guajava, Allium sativum, Azadiracha indica and *Spondias mombin*) extract showed larger diameter of inhibition zones than gentamycin, pefloxacin, streptolysin, septrin, ciprofloxacin and amoxicillinantibiotics respectively. This observation is similar to the report ofIndu *et al.* [4]. Extracts of plant origin have played role in the findingdrugs like quinine from Cinchona [6]. Finding fresh antimicrobial herbs are important at present, owing to the escalating amounts of antibiotic resistance among pathogenic bacteria [20]. Shobana and Naidu [13]. Enhanced animal husbandry methods have added additional gravity to this issue. The outcomes from this study can be encouraging, since the potency of several of the four out the five plant extracts is very high.

REFERENCES

1. Arora, D.S. and J. Kaur, 1999. Antimicrobial activity of spices. International Journal of Antimicrobial Agents, 12(3): 257-62.
2. Avato, P., E. Tursil, C. Vitali, V. Miccolis and V. Candido, 2000. Allylsulfide constituents of garlic volatile oil as antimicrobial agents. Journal of Phytomedicine, 7(3): 239-243.
3. Moravec, F. and J.L. Justine, 2014. Philometrids (Nematoda: Philometridae) in carangid and serranid fishes off New Caledonia, including three new species. Journal of SystematicParasites 21: 21.
4. Indu, M.N., A.A.M. Hatha, C. Abirosh, U. Harshaand G. Vivekanandan, 2006. Antimicrobial activity of some of the south-Indian spices against serotypes of *Escherichia coli*, *Salmonella*, *Listeria monocytogenes* and *Aeromonas hydrophila*. Journal of Microbiology, 37(2): 50-64.
5. Fabricant, D.S. and N.R. Farnsworth, 2001. The value of plants used in traditional medicine for drug discovery. Journal of Environmental Health, 109(1): 69-75.
6. Tapsell, L.C., I. Hemphill and L. Cobiac, 2006. Health benefits of herbs and spices: the past, the present, the future. Medical Journal of Australia, 185(4): 4-24.
7. Sherman, P.W. and G.A. Hash, 2001. Why vegetable recipes are not very spicy. Journalof Evolution and Human Behaviour, 22(3): 147-163.
8. Sumner, J., 2000.The Natural History of Medicinal Plants. Timber Press. pp: 16.
9. Janda, J.M. and S.L. Abbot, 1998. Evolving concepts regarding the genus *Aeromonas*: an expanding panorama of species, disease presentations and unanswered questions. Journal of ClinicalInfection Disease, 27(2): 40 52.
10. Kala, C. and S. Prakash, 2007. Revitalizing Indian systems of herbal medicine by the National Medicinal Plants Board through institutional networking and capacity building. Journal of Current Science, 93(6): 797-806.
11. Bassam, A.R.H., 2012. Medicinal Plants (Importance and Uses).Journal ofPharmaceuticaAnalytica, 3: 10.
12. Lichterman, B.L., 2004. Aspirin: The Story of a Wonder Drug. British Medical Journal, 329(7479): 1408.
13. Shobana, S. and K.A. Naidu, 2000. Antioxidant activity of selected Indian spices. Journal of Prostaglandins Leukotology and Essential Fatty Acids, 62(2): 107-110.
14. Elnima, E.I., S.A. Ahmed, A.G. Mekkawi and J.S. Mossa, 1983. The antimicrobial activity of garlic and onion extracts. Journal of Phytomedicine, 38(11): 747-748.
15. Grutter, A.S., 1994. Spatial and temporal variations of the ectoparasites of seven reef fish species from Lizard Island and Heron Island, Australia. Journal of Marine Ecology, 115: 21-30.

16. Thongson, C., P.M. Davidson, W. Mahakarnchanakul and J. Weiss, 2004. Antimicrobial activity of ultrasound-assisted solvent-extracted spices. Journal of Applied Microbiology, 39(5): 401-406.
17. Gold, S.G. and R.C. Moellering, 1996. Antimicrobial drug resistance. Journal of Medicine, 335: 1445-1453.
18. De, M., A. KrishnaDe and A.B. Banerjee, 1999. Antimicrobial screening of some Indian spices. Journal of Phytomedicine, 13(7): 616-618.
19. Cohen, M.L., 1992. Epidemiology of drug resistance, implications for a post antimicrobial era. Journal of Science, 257: 1050-1055.
20. Hill, A.F., 1952. Economic Botany. A textbook of useful plants and plant products. 2nd edition. McGarw-Hill Book Company Inc, New York.