COMPARATIVE ANALYSIS OF THE EFFICACY OF ALLIUM SAVITUM (GARLIC) AND ERYTHROMYCIN ON STREPTOCOCCUS PYOGENES

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ABSTRACT

Comparative efficacy of garlic and erythromycin on *streptococcus pyogenes* was carried out in vitro using agar-well diffusion technique. The *streptococcus pyogenes* used were isolated from the conjunctiva of infected patients that visited Abia State University, Optometry clinic. The isolated microorganisms were identified and characterized using standard laboratory methods like gram stain, catalase test and bacitracin test. Diameter of zone of inhibition was measured for different concentrations of garlic and erythromycin. The data collected were tabulated and analyzed using student's t-test at 0.05 level of significance. The results of the study showed that *streptococcus pyogenes* was sensitive to both garlic and erythromycin with the latter being more efficacious. The effect of garlic on the microorganism was also found to be concentration dependent. The minimum inhibitory concentration was found to be 31.25mg/ml. The antibacterial activity of garlic could be used as a preventative measure rather than curative in the management of microorganisms like *streptococcus pyogenes*.

KEYWORDS: *streptococcus pyogenes*, erythromycin, *allium savitum*, agar-well diffusion technique, minimum inhibitory concentration.

INTRODUCTION

The use of higher plants and their extracts to treat infections is an age-old practice in the traditional African medicine and other parts of the world[1]. Traditional medicine as defined by the World Health Organization (WHO) refers to the health practices, approaches, knowledge and beliefs incorporating plant, animal and mineral-based medicine, spiritual therapies, manual techniques and exercises applied singularly or in combination to treat, diagnose and prevent illness or maintain well being[2]. Roots, barks and leaves of various plants are employed in ethonomedicine. Plants extracts are given singly or as concoctions for treatment of diseases[3].

Garlic (*Allium Savitum*), a perennial plant in the family of Alliacae and genus Allium[4] has long been used as additives and medicinal herbs. It has been revered for its medicinal properties, which have escalated in recent years as a result of data indicating that garlic may influence the risk of health diseases and cancer[5]. The therapeutic efficacy of garlic is attributable not only to few isolated constituents, but to the overall compound of the plant with a wide range of substances such as vitamins, nicotinamide, hormones, choline, thiocyanic acid, iodide and saponinons[6].

Researchers have indicated that garlic has some health benefits such as reduction of platelet aggregation[7], serum cholesterol level[8], risks of thrombosis and hypertension[9,10], diabetic mellitus[11]; artherosclerosis inhibition[12] and exhibits broad antibiotic spectrum[13]. It has been suggested that garlic decreases fatigue and improves memory[14].

Erythromycin is a macrolide antibiotic, which has antimicrobial spectrum similar to or slightly wider than that of penicillin and is often used for people who are allergic to penicillin. The structural components of erythromycin (14 numbered lactone rings with ten asymmetric center and two sugars) make it difficult for the compound to be produced via synthetic methods[15]. Erythromycin prevents bacteria from growing by interfering with their protein synthesis. Gastric acids easily inactivate it; hence, all orally administered formulations are given as either enteric coated or as more stable salts or esters.

Streptococcus pyogenes is a gram positive, non-motile, non-spore forming coccus that

occurs in chains or in pairs of cells. Individual cells are round to ovoid cocci, about 0.6 1.0 micrometer in diameter. Streptococci divide in one plane and thus occur in pairs or in chains of varying lengths[16]. *Streptococcus pyogenes* is one of the most frequently occurring pathogens of humans. It is estimated that between 5 15% of normal individual harbour the bacterium, usually in the respiratory tract, without signs of disease. As normal flora, *streptococcus pyogenes* can infect when defenses are compromised or when the organisms are able to penetrate the constitutive defense[17]. Diagnosis of streptococcal infection is based on both clinical and laboratory findings.

METHOD

This research was carried out at the microbiology laboratory of Abia State University, Uturu with the assistance of a microbiologist. Sterile swabs were used to collect loads of bacteria from the conjunctiva of patients of Abia State University, Optometry clinic suffering from conjunctivitis. Cultures of the bacteria were formed in pre-prepared Petri dishes containing blood agar. *Streptococcus pyogene* organisms were identified and characterized using standard laboratory methods like gram stain, catalase test and bacitracin test. The organisms so identified were used to test for the efficacy of *Allium Savitum* and erythromycin.

Garlic used for this study was purchased from Eke Okigwe market of Imo State and authenticated by a botanist in the Department of Plant Science, of the same University. The garlic bulbs were peeled, washed and mashed into a paste. Five grams of the paste was weighed out and mixed with 10ml of ethanol and allowed to stand for 6hrs[4]. The mixture was then filtered out to get the garlic extract. This extract was recorded as the stock. Other concentrations of garlic extract used were obtained through double dilution method[17]. The erythromycin used was manufactured by Abbot Pharmaceuticals, Pakistan with NAFDAC no. 3205. Different concentrations of erythromycin used were also obtained through double or serial dilution.

The isolated *streptococcus pyogenes* were subjected to sensitivity test using *allium savitum* extract and erythromycin eye drop. The minimum inhibitory concentration (MIC) of garlic extract on *streptococcus pyogenes* was determined according to Baron et al[16]. The various dilutions, the stock and the erythromycin eye drop were used to carry out sensitivity test using agar-well diffusion technique. The zones of inhibition were measured horizontally and vertically with the help of millimeter rule and recorded as diameter of zone of inhibition. The measured diameters of zones of inhibition of the various concentrations of the garlic extract and erythromycin were then compared.

The data obtained were tabulated and statistically analyzed using student's t-test.

RESULTS

The efficacy of garlic and erythromycin on streptococcus pyogenes was studied using zones of inhibition. Garlic extract (stock) was found to have zone of inhibition of 25mm while erythromycin had 34.5mm (table 1). These results showed the activities of the two substances to be comparable. Table 2 shows the different concentrations of erythromycin derived through serial dilution and their zones of inhibition. The efficacy of different garlic concentrations obtained from doubling dilution and their zones of inhibition on *streptococcus pyogenes* were tabulated in table 3. The MIC of garlic was found to be 31.25mg/ml.

Statistical analysis using student's t-test showed no significant difference ($p<0.05$) hence, garlic (*allium savitum*) is not more effective on streptococcus pyogenes than erythromycin in vitro (t_{tab}, 1.860 > t_{cal}, 1.530). The results of zones of inhibition showed also that the diameters were greater for erythromycin than garlic indicating the efficacy of erythromycin to be greater than that of garlic.

DISCUSSION

The findings in tables 1, 2 and 3 showed that the test organism *streptococcus pyogenes* was sensitive to both garlic and erythromycin in vitro. The finding is consistent with the report of Donnefield and Pessy[18], who said that erythromycin is a standard, widely used antibiotic because of its relatively lack of toxicity and good activity against *streptococci, staphylococci, haemphilus influenza, legionella pneumohila and chylamydia trachomatis*. It also agrees with the report of Sivam[13] that garlic exhibits a broad spectrum against both gram-positive and gram-negative bacteria.

Erythromycin showed a relatively higher antibacterial activity when compared with garlic (stock) with a zone of inhibition of 34.50mm and

25.0mm respectively (see table 1). The minimum inhibitory concentration (MIC) of the garlic extract was determined using double dilution method of Baron et al[16]. It was found that the zone of inhibition of garlic on *streptococcus pyogenes* was concentration dependent. The zone of inhibition steadily decreased from 22.50mm at a concentration of 125mg/ml to 12.0mm at a concentration of 31.25mg/ml, below this concentration, garlic had no effect on *streptococcus pyogenes*.

Statistical analysis of the data showed no significant different ($p>0.05$) of garlic antibacterial activities when compared with that of erythromycin, though the results of MIC and diameters of zones of inhibition showed greater inhibition of the microorganism with erythromycin than garlic. Hence, erythromycin was found to be more efficacious in the management of the test organisms than garlic. Inspite of the above observation, garlic has been found to be a powerful antibiotic, albeit broad spectrum and not targeted. The body does not appear to build up resistance to garlic so that its positive health benefits continue over time. On the other hand, researchers have found erythromycin resistant group of streptococci microorganisms especially the group A streptococci[19, 20, 21]. The compound allicin found in garlic had been found to be responsible for the antibacterial properties of garlic. Allicin inhibits the growth of pathogenic microorganisms and has a wide spectrum of antibacterial activity against gram-positive and gram-negative bacteria[22].

Garlic though exhibits antibiotic activities; it cannot be used to replace orthodox therapy for conditions like conjunctivitis but could be used as preventative and adjuvant measures.

REFERENCES

1. Sofowora, A. (1984): Medicine plants and traditional medicine in Africa. John Wiley and Chichester, Spectrum books limited, 8-76.
2. Baggozzi, A. (2002): Increase in Traditional Medicine Practice. Russ. Acad. Med. Sci., 120:189-90.
3. Kimbi, H. K. and Fagbenro-Beyioku, B. K. (1996): Efficacy of Cymbopogom gigantous plasmodia. East Afr. Med. J., 12:636-8.
4. McGee, K. and Harold, C. (2004): Garlic; food and cooking. Revised edition. Scribner Books Co., 316-23.
5. Ishikawa, K., Katsuzaki, H., Ohta, R. and Yoshida, S. (1999): An organosulphur compound isolated from oil macerated garlic extract and its antimicrobial effect. Bio. Sci. Biotech. Biochem., 63: 588-90.
6. Limson, K. (2001): The science behind the legendary healing properties of garlic. Sci. in Afr., 17:10.
7. Steiner, M. and Li, R. S. (1998): Changes in platelet function and susceptibility of lipoproteins to oxidation associatedwith administration of aged garlic extract. J. Cardiovasc. Pharmacol., 31: 904-8.
8. Neil, H. A., Silagy, C. A., Lancaster, T., Hodgeman, J., Vos, K., Moore, J. W., Jones, L., Cahill, J. and Fowler, G. H. (1996): Garlic powder in the treatment of moderate hyperlipidaemia a controlled trial and meta-analysis. J. R. Coll. Physicians, London, 30: 329-34.
9. Petkov, V. (1979): Plants and hypotensive, antiatheromatous and coronarodilating action. Am. J. Clin. Med., 7:197-236.
10. Ali, M. and Thomson, M. (1995): Consumption of garlic clove a day could be beneficial in preventing thrombosis. J. Tradit. Chin. Med., 53: 211-2.
11. Zhang, X., Lowe, D., Giles, P., Fell, S., Connock, M. J. and Maslin, D. J. (2001): Gender may affect the action of garlic oil on plasma cholesterol and glucose levels of normal subjects. J. Nutri., 131: 1471-8.
12. Campbell, J. H., Efendy, J. L., Smith, N. J. and Campbell, G. R. (2001): Molecular basis by which garlic suppresses atherosclerosis. J. Nutri., 131: 10065-95.
13. Sivam, G. (2001): Antibacterial properties of garlic. J. Nutri., 131(3): 1106.
14. Milner, J. A. (1996): garlic- its anticarcinogenic and tumorigenic properties. Nutri. Rev., 54:581-4.
15. Lewin, C. S. (1993): Use of Macrolides in treatment of Group A streptococcal infections in children. J. R. Coll. Physcian, Lond., 28: 39 45.
16. Baron, E. J., Lance, R. P. and Sydney, M. F. (1994): Diagnostic Microbiology. 9th edn. The C. V. Mosby Publishers, pp 203-958.
17. Prescott, L. M., Harley, J. P. and Wein, D. A. (2002): Microbiology. 5th edn. The C. V.

Mosby Publishers, pp 984 -5.
18. Donnenfield, E. D. and Pessy, H. D. (1994): Antibacterial effectiveness of clarithromycin and azithromycin. Am. J. Ophthalmol., 101: 902 5.
19. Martin, J. M., Green, M., Barbadora, K. A. and Wald, E. R. (2002): Erythromycin-Resistant Group A Streptococci in school children in Pittsburgh. The New Eng. J. Med., 346 (16): 1200-6.
20. Katz, K. C., McGeer, A. J., Duncan, C. L., Ashi-Sulaiman, A., Willey, B. M., Sarabia, A., McCann, J., Pong-Porter, S., Rzayer, Y., de Azavedo, J. S. and Low, D. E. (2003): Emergence of Macrolide Resistance in Throat Culture isolates of Group A streptococci in Ontario, Canada in 2001. Antimic. Agents Chemo., 47 (7): 2370-2.
21. Low, D. E. (2003): Emergence of macrolide resistance in throat culture isolates of group A streptococci in Ontario, Canada in 2001. Antimicr. Agents Chemother., 47(7): 2370-2.
22. Uchida, Y., Takahashi, T. and Sata, N. (1975): The characteristics of antibacterial activity of garlic. Jap. J. Antibiotic, 28: 638-42.

TABLE 1: GARLIC AND ERYTHROMYCIN SENSITIVITY WITH STREPTOCOCCUS PYOGENES.

	Zones of inhibition		
.	**Vertical diameter (mm)**	**Horizontal diameter (mm)**	**Average (mm)**
Garlic extract (stock)	26	24	25
Erythromycin	35	34	34.50

TABLE 2: DIAMETER OF INHIBITION (IN MM) PRODUCED BY ERYTHROMYCIN

	Zones of inhibition		
Concentration (mg/ml)	**Vertical diameter (mm)**	**Horizontal diameter (mm)**	**Average (mm)**
50.0	35	34	34.50
5.0	34	33	33.50
0.50	29	28	28.50
0.05	20	18	19
0.005	11	10	10.50

TABLE 3: DIAMETER OF ZONES OF INHIBITION (IN MM) PRODUCED BY GARLIC EXTRACT ON STREPTOCOCCUS PYOGENES

	Zones of inhibition		
Concentration (mg/ml)	**Vertical diameter (mm)**	**Horizontal diameter (mm)**	**Average (mm)**
250	26	24	25
125	22	23	22.50
62.50	18	17	17.50
31.25	12	12	12.0
15.625	0	0	0