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Screening of Antibacterial Efficacy of Aloe Barbadensis Miller (*Aloe Vera*) Using Leaf Extract against Different Bacterial Strains

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Abstract: ***The current research was aimed to evaluate in vitro antibacterial activity of crude extract prepared from leaves of Aloe vera plant. The extract of Aloe vera was prepared in Methanol. Two extracts were prepared, one from whole leaf powder and the other from fresh Aloe vera gel. To determine the antibacterial efficacy of the given plant, Kirby-bauer disk diffusion method was performed. The standard antibiotic used was Gentamicin. Gentamicin showed significant antimicrobial efficacy against E.coli and Bacillus subtilis forming a zone of inhibition of 34mm in both. This research provides information about antibacterial susceptibility of aloe vera leaves (leaf powder and fresh gel) against two different bacteria: E.coli which is a gram-negative bacteria and Bacillus subtilis which is a gram-negative bacteria. The antibacterial susceptibility test showed that the extract of leaf and the gel of Aloe vera inhibited the growth of both microorganisms during test. Growth of E,coli strain were inhibited more as compared to Bacillus subtilis because gram negative bacteria do not have lipopolysaccharide layer in their cell wall. Also the fresh gel Extract was more effective as compared to Aloe vera leaf powder.***

Keywords: Aloe vera, antibacterial, disc diffusion, inhibition, Aloe vera gel, leaf extract, antibiotics.

I. INTRODUCTION

Infectious diseases are a major cause and concern for silent deaths all over the world. There are many antibacterial medicines and antibiotics are available in the market but development of resistance against them is a major concern. Plants are a great source of many compound shaving antibacterial properties, which can be observed and used in future for development of various antibacterial medicines. *Aloe vera* is a succulent plant species belongs to Asphodelaceae family. It is a great medicinal plant having enormous properties. This plant have water storage tissue inside the leaves of the plant, so that it can survive in areas. Inside of the plant leaf, a clear liquid is found which is soft and slippery[1]. Fleshy leaves of the plant not only contains cell wall carbohydrates like cellulose and hemicellulose but also storage carbohydrates such as acetylated mannans[2]. Aloe Leaf can be divided into two portions, each of them have different composition and therapeutic properties. Inner part have clear jelly like material which is mucilaginous pulp called Aloe gel. The Aloe gel is made up of 96% of water, rest of the 4% includes vitamins, amino acids, calcium and enzymes. Beneath the green rind of the leaf, it produces a bitter yellow exudate[3],[4]. Aloe vera is being cultivated from many years in huge quantity because of its high demand in Pharmacy, Industries, Cosmetics sectors. Pharmacological actions of this great plant consists of some Anti-inflammatory and Anti-arthritic activities. It also shows antibacterial properties and hypoglycaemic effects. This plant is depicted as healing plant or silent healer as it can heal wounds and burns[5]. *Aloe vera* is called Ghrit kumari in sanskrit. Ayurveda delineates *Aloe vera* plants multi-functions such as anti-inflammatory, laxatives, blood purifier, diuretic, uterine tonic, spermatogenic, fever reliever etc. Also it plays significant role in purgative, used as appetite regulator, effective in cold and cough, can help in piles, dyspnoea, jaundice and asthma[6]. *Aloe vera* is the most commercialized plant. In food industries, it is used as a healthy ingredient of food products, Aloe drinks are quite famous for their health benefits hence the gel of aloe plant is used in the preparation of various beverages. It has been used in cosmetics and toiletry industries in the manufacture of soaps, creams, handwashes, lotions, cleansers and many more products. They are also used in the manufacture of antibacterial creams and ointments due to their considerable efficacy against pathogenic microbes[7],[8].

Many scientific studies been carried out to determine the use of *aloe vera*, some of them conflicting. Inspite of these constraints, there is some prefatory proofs that *Aloe vera* extracts may be useful in the medical treatment of wound and burn healing, skin infections, cyst, diabetes, and elevated blood lipids in humans. The productive effects are thought to be due to the presence of compounds such as polysaccharides, mannans, anthraquinones, and lectins.

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II. MATERIALS AND METHODS

A. Sample Collection

Aloe vera plant is a medicinal plant and is of great importance. Researches have shown its different properties and efficacies in treatment of various diseases. Its farming and maintenance are quite genuine. Study of Antibacterial efficacy was done in Institute of Applied Medicines & Research, Duhai, Ghaziabad. The Aloe vera plant was Harvested from pot which was kept at home. Plant was 2 years old, having mature leaves. Only green, uninfected mature leaves were harvested for research purpose.

1) Chemicals Used: Nutrient Agar media, methanol, Antibiotic discs, Dimethyl sulfoxide (DMSO), filter paper disc.

B. Preparation of Leaf Extract

Mature fresh *Aloe vera* leaves were harvested and washed until clean with fresh water. Dead parts and thorns were removed with a sterile cutter. Leaves were then chopped into extremely thin slices including the gel within. On a clean sheet of paper, the thin slices of leaves were spreaded carefully and covered with a thin cotton cloth to prevent any dust sitting on it. The preparation was kept in shed at 25°C for 40 days to completely dry. After drying, the dried leaves were grounded into powder in a clean mixer grinder and the powder was kept in a clean container to avoid any contaminants.

C. Methanol Extract

For extract preparation, About 50g of leaf powder was weighed in electronic weighing machine in a clean beaker, and was dissolved in 200ml of methanol i.e, 1:4 concentration. This mixture was kept for 72 hours at room temperature and shaken twice a day. After the given time, the content was then filtered using whattman filter paper no.4, so that the compounds which were soluble in the solvents can be filtered. Again, the filtrate was kept undisturbed for 72 hours to evaporate methanol (this can be done by using rotatory evaporator). After evaporation, the semisolid extract is then weighed. Four different Dilutions of were prepared in DMSO. DMSO is used as a solvent as it do not have any antimicrobial effect of its own. 800mg aloe extract dissolved in 1ml Dimethyl sulfoxide (DMSO), 400mg aloe extract in 1ml DMSO, 200mg in 1ml DMSO , 100mg aloe extract in 1 ml of DMSO in separate vials. Mixed properly and stored at room temperature. Disc of whattman filter paper were prepared and dipped for 24 hours in all 4 concentrations kept in vials and capped tightly.

D. Preparation of Aloe vera gel

Mature leaves of *Aloe vera* were harvested and washed with clean water. Their thick epidermis layer (outer green leafy part) was removed with knife and solid mucilaginous gel was collected in a sterile beaker. Slurry was formed with the help of Mortar and pestle. Four different concentrations of aloe vera gel were also prepared. 800mg of gel in 1ml DMSO, 400mg in 1ml DMSO, 200mg in 1ml DMSO and 100mg in 1ml DMSO. Disc of filter paper no.4 were dipped in all 4 vials and kept undisturbed for 24 hours.

E. Microorganisms

The test microorganisms includes one gram negative bacteria *Escherichia coli* **(*E.coli*)** and one gram positive bacteria *Bacillus subtilis*. They were previously isolated and identified. Stored in refrigerator in the Department of life sciences at Institute of Applied Medicines & Research, duhai, Ghaziabad.

F. Disc Diffusion Method

The antibacterial activity of of the leaf and gel of the plant was tested by using disc diffusion method. A total of 20 sterile Nutrient Agar Plates were prepared in Laminar Air Flow. The bacterial test organism, E.coli and *Bacillus subtilis* were spread all over the plate on media with the help of a sterile cotton swab simultaneously. The plates were kept for 5 minutes in the Laminar so that the bacterial culture can be dried. After the lawn preparation, the disc of different concentrations of leaf extract and gel extract were placed on the organism inoculated plates. 1 plate for both organisms were kept as control in which antibiotic discs were placed at equal distance of other discs. Here, antibiotic disc Gentamicin used for *E.coli Bacillus subtilis* were used. All the plates were kept in the BOD Incubator and incubated for 24 hrs at 27°C. The diameter of Zone Of Inhibition was measured in mm(millimeter).

III. RESULTS

The current study carried out on *Aloe vera* plant whole leaf and gel alone. It revealed the antibacterial capacity of the plant against two common bacteria. The leaf extract and gel extract both were prepared using methanol. Methanol is used as solvent to strain all the required compounds from the plant leaf to get better results. Aloe vera leaf extract Inhibited the growth of both bacteria, was more effective on Bacillus as compared to *E.Coli*. but the opposite was seen in the case of *Aloe vera* gel extract.

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The gel alone showed high activity against *E.coli* in comparison to Bacillus strain. This study can be taken as comparison between the aloe vera powder and fresh gel. Fresh gel of the plant shows great efficacy than the powdered leaf.

Table 1: Antibacterial Activity of *Aloe vera* leaf powder

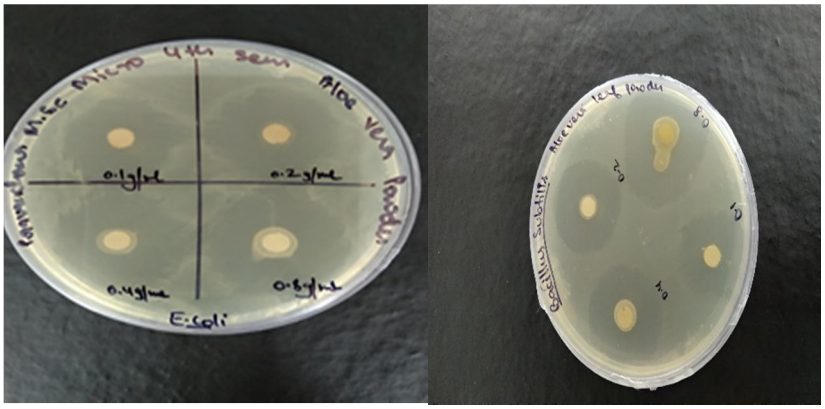
Concentration of Aloe vera leaf extract (g/ml)	Zone of Inhibition (diameter in mm)		Control
	E.Coli	*Bacillus subtilis*	*Gentamicin*
0.1g/ml	16mm	18mm	34mm
0.2g/ml	20mm	21mm	
0.4g/ml	25mm	26mm	
0.8g/ml	29mm	30mm	

Table 2: Antibacterial activity of *Aloe vera* gel

Concentration of Aloe vera gel extract (g/ml)	Zone of Inhibition (diameter in mm)		Control
	E.Coli	*Bacillus subtilis*	*Gentamicin*
0.1g/ml	20mm	19mm	34mm
0.2g/ml	26mm	25mm	
0.4g/ml	28mm	27mm	
0.8g/ml	31mm	32mm	

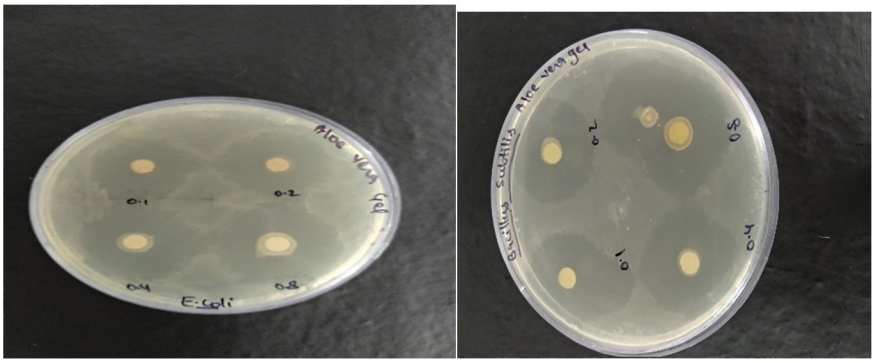
Fig 1: Plates showing Zone of inhibition

A. Aloe Vera Leaf Powder Extract Plates



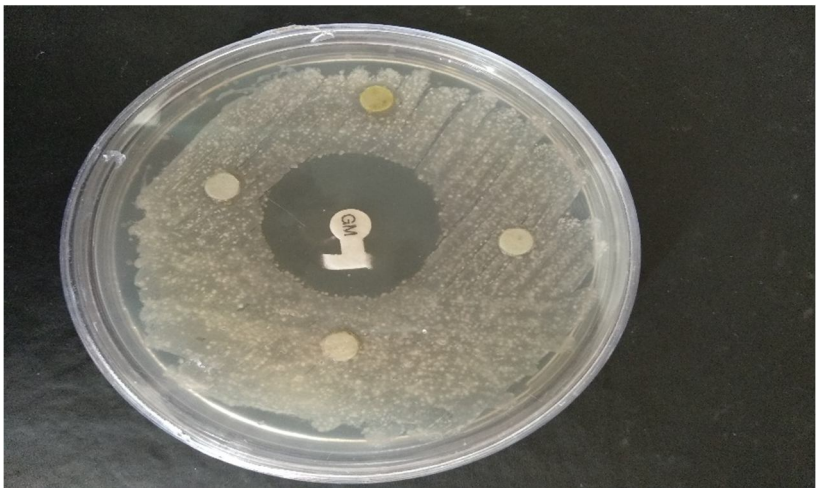
E.Coli *Bacillus subtilis*

B. Aloe Vera Gel Extract Plates



E.Coli *Bacillus subtilis*

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Gentamicin control

IV. DISCUSSION

The current study had shown the antibacterial efficacy of *Aloe vera* leaf powder and *Aloe vera* gel by the presence of Zone of Inhibition. Both the extracts were prepared using ethanol, it has been confirmed that only alcoholic extracts are better as a solvent for preparing the extract to study antimicrobial activity[9]. Fresh aloe vera gel extract was proven more effective against both the bacteria. When the zone of inhibition was compared with the standard drugs, gentamicin, it was observed that the resulted plant extract zones are very close to standard antibiotics which is a good result. The above result data supports strong scientific data and can be used to develop the treatment of microbial diseases. Many antibacterials can be produced using the natural plant and its product. Apart from diseases treatments, the data can be used for the production of industrial edible suppliments and toiletries. Hence it can be conclude from the above study that aloe vera plant contains strong antibacterial agents which have the ability to replace most of the medicines of this era and many small diseases and infections can be treated naturally with the help of this wonder plant.

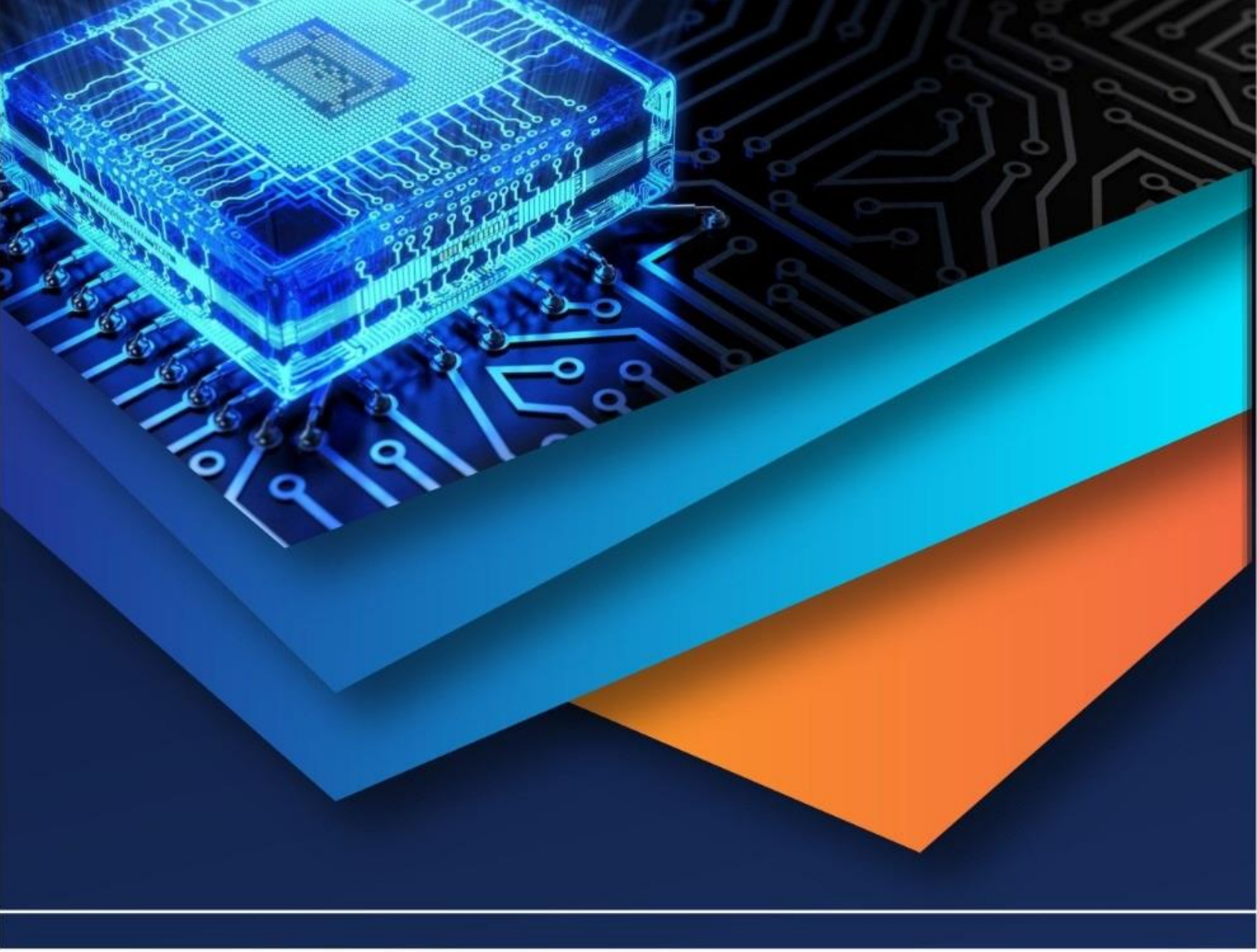
V. CONCLUSION

The results from the study suggested that Aloe vera is a sublime plant, which shows marvelous Antibacterial properties against pathogenic gram positive and negative microbes. The results and the potential of aloe vera as an antimicrobial plant can be used further to develop antibiotics against the bacterias causing common diseases. Also this efficiency can be utilized in industries to make products which can help in killing these microbes before entering into the body. Aloe vera is being used and can be used further to manufacture antibacterial lotions, soaps, creams, serums for healthy skin as well as drinks and edibles to make body healthy and disease free**.**

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