INTRODUCTION
An Endemic Medicinal plant C. neilgherrensis reported from Araku valley and also from Tirumala and Talakona of Seshachalam Hill Ranges along the Eastern Ghats. C. neilgherrensis commonly called as “Adavi Pasupu,” in Telugu “Manjakova” “Katterkalavazh” by the Pallyyar Tribes of Tamil Nadu. The plant has lot of medicinal properties as anti inflammatory, Cholagogue, hepatoprotective, blood purifier, antioxidant, toxifier, anti-asthmatic, antitumour, stomachic, carminative and regenerator of liver tissue. It is also used for chronic hepatitis, antiarthritic activity. It is also reported as the liver tissue. It is also used for multiple therapeutic activities by the herbalists and the tribes. To prove its antibacterial activity rhizomes and leaf extracts were tested against both gram +ve and gram -ve bacteria of human pathogens.

MATERIALS AND METHODS
The rhizomes and leaves were collected and the species was identified as per the standard methods of Jain and Rao and the species was identified as per the standard methods of Jain and Rao and Kokate, antibacterial activity was performed by Agar well diffusion method of Perez and MIC values by Volkova and Usman. The data were subjected to standard deviation. All the results were given in the respective tables, figures and graphs.

Results: Antibacterial activity of C. neilgherrensis showed more susceptible activity on Staphylococcus aureus with rhizome cold water extracts at 10mg/well than the standard drug Ampicillin. All extracts of rhizome and leaf expressed most promising activity against human pathogens. Minimum Inhibitory Concentrations ranges from 0.078 to 2.5mg.

Conclusion: Antibacterial activity and the MIC values of the C. neilgherrensis rhizome and leaf extracts are comparable to that of Clonga, Cedoaoaria, C. Xanthorrhiza on S. aureus and other mouth and dental pathogens and other human pathogens like Klebsiella, Vibrio, Bacillus, Streptococcus, Azotobacter, Enterobacter, Pseudomonas and Ecoli.

ABSTRACT
Background: Curcuma longa (Zingiberaceae) is an import spice ingredient of Indian food which is also a major export and economic source of foreign exchange. Clonga is also having many traditional and herbal medicinal uses having Curcuminoids and volatile oils. C. neilgherrensis (Adavi pasupu) a wild species and also an endemic to Eastern Ghats also used for multiple therapeutic activities by the herbalists and the tribes. To prove its antibacterial activity rhizomes and leaf extracts were tested against both gram +ve and gram -ve bacteria of human pathogens.

Materials and Methods: The rhizomes and leaves were collected and the species was identified as per the standard methods of Jain and Rao and extracts were prepared as per the methods of Kokate, antibacterial activity was performed by Agar well diffusion method of Perez and MIC values by Volkova and Usman. The data were subjected to standard deviation. All the results were given in the respective tables, figures and graphs.

Results: Antibacterial activity of C. neilgherrensis showed more susceptible activity on Staphylococcus aureus with rhizome cold water extracts at 10mg/well than the standard drug Ampicillin. All extracts of rhizome and leaf expressed most promising activity against human pathogens. Minimum Inhibitory Concentrations ranges from 0.078 to 2.5mg.

Conclusion: Antibacterial activity and the MIC values of the C. neilgherrensis rhizome and leaf extracts are comparable to that of Clonga, Cedoaoaria, C. Xanthorrhiza on S. aureus and other mouth and dental pathogens and other human pathogens like Klebsiella, Vibrio, Bacillus, Streptococcus, Azotobacter, Enterobacter, Pseudomonas and Ecoli.

INTRODUCTION
An Endemic Medicinal plant C. neilgherrensis reported from Araku valley and also from Tirumala and Talakona of Seshachalam Hill Ranges along the Eastern Ghats. C. neilgherrensis commonly called as “Adavi Pasupu,” in Telugu “Manjakova” “Katterkalavazh” by the Pallyyar Tribes of Tamil Nadu. The plant has lot of medicinal properties as anti inflammatory, Cholagogue, hepatoprotective, blood purifier, antioxidant, toxifier, anti-asthmatic, antitumour, stomachic, carminative and regenerator of liver tissue. It is also used for chronic hepatitis, antiarthritic activity. It is also reported as the liver tissue. It is also used for multiple therapeutic activities by the herbalists and the tribes. To prove its antibacterial activity rhizomes and leaf extracts were tested against both gram +ve and gram -ve bacteria of human pathogens.

MATERIALS AND METHODS
Plant Material Collection
Plant material C. neilgherrensis (Zingiberaceae) was collected from Tirumala, Talakona along the Seshachalam Hill Ranges during the months of April – September, 2011, the plant material was authenticated by Prof. N.Yasodamma and a voucher specimens DC 921, DC 922 were prepared and preserved in the herbarium Department of Botany, S.V.University, Tirupati as per the standard method [9]. Rhizomes and leaves were collected, thoroughly washed, cut in to pieces and further dried under shade at 28 ± 2 °C for about 10 days. The dried parts were ground well in to a fine powder in a mixer grinder and sieved to particle size of 50 – 150mm. The powders were stored in a polythene bags at room temperatures.

Antibacterial Activity
Extracts preparation
Shade dried leaf and rhizome powders were subjected to soaked extractions with Methanol, Ethanol and Hydro Alcohol. Simultaneously cold water and hot water extracts also prepared. The above obtained semisolid extracts were preserved in airtight bottles at 4°C in the refrigerator until further use.

Bacterial cultures
The bacterial strains Bacillus subtilis (MTCC- 441) causes Pneumonia, diarrhoea. Staphylococcus aureus (MTCC- 737) causes bone and joint pains, skin infections, boils; Escherichia coli (MTCC- 443) causes urinary tract infections, Pneumonia, Pseudomonas aeruginosa (MTCC- 741) causes urinary infections and Pneumonia. The bacterial strains were procured from the Department of Microbiology S.V.University Tirupati.

Preparation of inoculums
Stock cultures were maintained at 4°C of nutrient agar slants. Active cultures were prepared by transferring a loop full of cells from the stock cultures to test tubes containing nutrient agar medium and were incubated without agitation for 24 hrs at 37°C.

agar well diffusion assay
The antibacterial activity of the leaf and rhizome extracts (10mg/well) was determined by using agar well diffusion method by Perez with slight modifications [10]. Nutrient agar was inoculated with the selected microorganisms by spreading the bacterial inoculums on the media. Wells (9 mm diameter) were punched in the agar and filled with plant extracts. Control wells contained with the standard antibiotic positive control Ampicillin (10 mg/well). The plates were incubated at 37°C for 18hrs. The antibacterial activity was assessed by measuring the diameter of the zone of inhibition of the respective drug. The data of crude drug activity is given in the mean of quadruplicates along with the standard error.

Minimum Inhibitory Concentration
The Volkova method modified by Usman was employed [11-12]. In this method the broth dilution technique was used, where the leaf and rhizome extracts were prepared to the highest concentration of 10mg (stock concentration). By adding sterile distilled water and serially diluted (two fold dilution) using the nutritive broth and later inoculated with 0.2ml standardized suspension of the test organisms. After 18hrs of incubation at 37°C, the test tubes were observed for turbidity. The lowest concentration of the tube that did not show any visible growth can be considered as the Minimum Inhibitory Concentration.
RESULT

Antibacterial activity [Table: I], [Figure: I]

Crude drug mean concentration was assessed by studying antibacterial activity with 10mg/well, 50mg/well, 100mg/well and 150mg/well concentration. And observed the mean inhibition Concentration as 10mg/well an effective concentration against all pathogens with all extracts.

Antibacterial activity of leaf and rhizome with the Cold water, Hot water, Alcohol, Methanol and Hydro alcoholic extracts on the selected four strains resulted as *S. aureus* is most susceptible with rhizome cold water extracts at 10mg/well with 30.9mm zone of inhibition when compared to that of the control *Ampicillin* with 15.96 mm. But with leaf extracts also on *S. aureus* shows more effective inhibition zone with all the extracts than the control drug. Effective inhibition was observed against all selected bacterial strains with the leaf and rhizome extracts in order of Alcohol> Methanol > Hot water > Cold water > Hydro alcoholic extracts respectively.

Minimum Inhibitory Concentration [Graph: I]

All extracts of leaf and rhizomes are showing Minimum Inhibitory Concentrations at a very low against all pathogens ranging between 0.078 to 2.5 mg. The rhizome extracts showed the lowest concentrations against *Staphylococcus aureus* at 0.078mg with cold water and on *Pseudomonas aeruginosa* with hot water extracts at 0.156mg and 0.312 mg against *B. subtilis* and *E.coli*.

Table I: Antibacterial Activity of Leaf and Rhizome extracts (10 mg/well)

<table>
<thead>
<tr>
<th>Organism</th>
<th>Leaf</th>
<th>Rhizome</th>
<th>AMP</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>B. subtilis</em></td>
<td>17.7±</td>
<td>18.0±</td>
<td>22.7±</td>
</tr>
<tr>
<td><em>S. aureus</em></td>
<td>26.3±</td>
<td>22.8±</td>
<td>27.8±</td>
</tr>
<tr>
<td><em>P. aeruginosa</em></td>
<td>0.84±</td>
<td>0.16±</td>
<td>0.28±</td>
</tr>
<tr>
<td><em>E. coli</em></td>
<td>2.11±</td>
<td>23.1±</td>
<td>23.0±</td>
</tr>
</tbody>
</table>

Values are mean Inhibition zone (mm) ± SD of quadruplicate.

C. W = Cold Water; H. W = Hot Water; A = Alcohol; M = Methanol; H. A = Hydro Alcoholic; AMP = Ampicillin.

![Cold Water](image1.jpg)

![Hot Water](image2.jpg)

![Alcohol](image3.jpg)
Methanol

Hydro Alcoholic

Antibacterial Activity of Rhizome Extracts (10mg/Well)

Cold Water

Hot Water

Alcohol
Methanol

Hydro Alcohol

Antibacterial Activity of the Control drug Ampicillin (10mg/Well)

Ampicillin:

A: B. subtilis; B: E. coli; C: P. aeruginosa; D: S. aureus

1, 2, 3, 4: Quadruplicates for 10mg/well.

Fig. I: Antibacterial Activity of Leaf Extracts (10mg/well)

Graph I: Minimum Inhibitory Concentration
DISCUSSION
Antibacterial activity of Xanthorrhizol from *C. xanthorrhiza* rhizome extract against oral microorganisms resulted highest activity on four *Streptococcus* species which causes dental carries and also against *Actinomycyes viscosus* and *Porphyromonas gingivalis* responsible for periodontitis. Whereas on *Callicans* and *Lacto bacillus* somewhat resistant to Xanthorrhizol ranging MBC 4 to 8 µg/ml and MIC 2-4 µg to that of Chlorhexidine 2-4 µg/ml MBC and 1-4 µg MIC [13]

*C. zedoaria* and *C. malabarica* rhizome hexane and acetone extracts against six bacterial and two fungal strains exhibited effective activity than other extracts and *Cedulaaria* is most efficient with MIC values ranging from 0.01 to 0.094 µg/ml and MIC 2-4 µg to that of Chlorhexidine 2-4 µg/ml MBC and 1-4 µg MIC [13].

Clonga ethyl acetate extracts on MRSA showed highest antibacterial activity than other extracts and the MIC also very low to that of the control drug Ampicillin and Oxacillin at 0.125 to 2mg. [15] Aqueous rhizome extracts of *Clonga* on *E.coli*, *Saureus*, *S. epidermidis* and *K pneumonia* resulted effective inhibitory activity with MBC ranges 16-32 g L-1 and 4-16 g L-1 MIC to that of Gentamicin (0.5 mg L-1) [16]. Curcuminoid extracts of *C. longa* against 24 pathogenic bacterial strains isolated from shrimp and chicken resulted an effective activity with hexane extracts on 13 bacterial strains such as *Vibrio*, *Streptococcus agalactiae*, *Staphylococcus*, *Bacillus* and *Edwardsiella tarda*. With Curcuminoinds extracts only on 8 bacterial strains activity as on *A. hydrophila*, *Streptococcus, B. subtilis* and *E. coli* were observed. The MIC value of ethanol, Curcuminoinds hexane extracts ranges from 3.9 to 125; 391 to 500 and 125 to 1000 ppt respectively. Ethanol extracts of *C indica* proved as most effective drug than Curcuminoinds and hexane extracts of *Clonga* compare to Tetracycline the control drug. [17].

Crude ethanol extracts of Curcuminoinds and hydro methanol extracts of essential oils of *C. longa* from the varieties of Kasur, Fazirabad and Banu areas the antibacterial activity against three *Bacillus* spp and one Azobacter resulted effective activity with Kasur variety than others and *B subtilis* was the most sensitive to Curcuminoinds and oils of turmeric. [18]. *Clonga* volatile oils and Curcuminoinds hydro methanol extracts were tested for antibacterial activity against *B subtilis*, *K pneumonia*, *E.coli*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Saureus* and *E. Morboribilis* strains and on two fungal strains. *Aniger* and *Callicans* effective activity observed with Curcuminoinds extract than volatile oils on both bacterial and fungal strains to that of the standard drugs *Kanamycin* and *Flucanazole* respectively [19]. Aqueous and Hydro alcoholic extracts of *Clonga* on *Saureus* and *Callicans* proved very effective. [20].

Antibacterial activity of *C zedoaria* rhizome methanolic extracts of Malaysian variety both in vivo and in vitro conditions on four *Bacterial* strains of *B cereus*, *Saureus*, *E.coli* and *Pseudomonas* where in *in vitro* conditions inducing with growth hormones 0.25 to 3.5mg/Lt IBA and 0.5 to 4mg/Lt of BAP resulted effective inhibition than in *in vivo* conditions except on *B cereus* [21]. Essential oils isolated from *C unthorrhiza* as Xanthoriol, Camphere, Curcumin, α-pinene, β-pinene, Myrecene, Linalool antimicrobial activity against bacterial and fungal strains showed most effective on *E.coli*, *B. amylolyquefaciens*, *K pneumoniae*, *P. aeruginosa* followed by others. Strains and relatively less activity are observed on fungal strains. [22].

CONCLUSION
Rhizomes of *Cneiligherrensis* consists a good number of secondary metabolites like alkaloids, flavonoids, phenols, steroids, tannins, lignins, indoles, glycosides, carbohydrates, proteins, amino acids and proved their effective antibacterial activity against all pathogens especially which causes arthritis, skin diseases, diarrohea, urinary tract infections and Pneumonia; These extracts may also inhibit MRSA as that of *Clonga* apart from other bacterial strains also supports the antibacterial activity of *Clonga*, *C zedoaria* and *C xanthorrhiza*. Hence the herbal drug may be recommended as the best drug to cure bone fractures, skin diseases, boils and ulcers. Further studies may recommend for the isolation of bioactive constituents and biological assay methods for the standard drug preparations.

ACKNOWLEDGEMENTS
The authors are grateful to the University Grants Commission (UGC) New Delhi for financial assistance. We are also indebted to the Department of Botany, S.V.U College of Sciences, Sri Venkateswara University, Tirupati, Andhra Pradesh, India for providing the space and facilities to complete the above Research work. The authors are grateful in this regard.

REFERENCES

