6.1. Summary

From ancient times plants are being used for treatment of various illnesses. Presence of various bioactive molecules are considered key attribute to the medicinal properties of these plants. In order to study antimicrobial activity of plant extracts various methods like agar diffusion and broth dilution methods are used and bioactive principle is isolated from the potent extract using bioassay guided fraction technique. Information gathered through ethnomedicinal survey of plants used in primary health care by traditional healers and local people of villages has led to the development of increased interest among the researchers of present time to study the bioactivity of these plants. Although there is a diverse variety of medicinal flora and immense use folk medicine practice, the documentation of traditional use of medicinal plants is very limited in North East India. In recent time, a few researchers have reported medicinal use of plants of North East India including Assam. In this research, information were gathered through personal communication with the traditional herbal healers and local people of different parts of Assam. Various plant parts belonging to 21 plant species that were used by traditional healers for infectious disease treatment were collected and their vernacular names were recorded in the field. Different extracts prepared with different solvents were evaluated to study their antibacterial activity. The mechanism of action of the plant extract on the bacterial cells was also studied.
Some of the collected plant part extracts showed antibacterial activity screened by disc diffusion method. Only *L. salicifolia*, *M. pruriens*, *Xanthium strumarium* showed strong antibacterial activity, a few showed moderate activity and most of the extracts showed weak antibacterial activity. In this research, we selected *L. salicifolia* and *M. pruriens* for further analysis because, the extracts of these two plants showed broad spectrum activity.

PE and CHF extract of *M. pruriens* showed activity against all the four bacteria in dose dependant manner. MT extract was active against *S. pneumoniae* and *P. aeruginosa* whereas water extract was active only against *P. aeruginosa*. We carried out further analysis of CHF extract because CHF extract showed lowest MIC value against *S. aureus* and *E. coli*. Moreover, TLC analysis showed the presence of various chemical constituents in CHF extracts compared to other extracts. GC-MS analysis of the crude chloroform extracts revealed the presence of various chemical components with α-Linolenic acid as the major component. Mechanism of action of the CHF extract on bacterial cells showed that treatment with the extract resulted in generation of reactive oxygen species (ROS) and also there was considerable cell and DNA damage of both *E. coli* and *S. aureus*. Purification of the crude CHF extract using bioactivity guided fractionation resulted in decrease in activity of the fractions. This suggested synergistic activity of the chemical components of the crude extracts.

The CHF extract of *L. salicifolia* showed broad spectrum activity by disc diffusion method. The MIC value of CHF extract against *S. aureus* and *E. coli* appeared to be 0.076±0.023 mg/ml and 0.096±0.041 mg/ ml which suggested strong antibacterial activity.
of the chloroform extract. Study of the mechanism of action of the extract on bacterial cells indicated generation of oxidative stress and also damage of cell structure and DNA. Time kill kinetic study of the treated bacterial cells suggested bactericidal activity of the chloroform extract against *E. coli* and *S. aureus*. When the crude extract was purified using column chromatography five fractions were obtained. Out of the five fractions, fraction 1 showed strong antibacterial activity against *S. aureus* with MIC value 0.08± 0.017 mg/ ml. From fraction 1, a compound namely β- sitosterol was isolated. However, the antibacterial activity of β- sitosterol was much lower than fraction 1 and the crude extract. This suggested that in the antibacterial activity of *L. salicifolia* extract compounds other than β-sitosterol was also responsible.

6.2. Conclusion

In recent times, numerous studies on pharmacological properties of medicinal plants have been accomplished and many plant based drugs have also been produced that are commercially available in the market such as Taxol, Artemisinin etc. Development of plant based antimicrobial agents is an alternative tool to fight infectious diseases, which would probably decrease the cost and improve the quality of treatment.

In our study, among all the plant extracts tested, petroleum ether and chloroform extract of *M. pruriens* and *L. salicifolia* possessed good antibacterial activity. *S. aureus* was found to be most sensitive bacterial strains against the plant extracts. Polar plant extracts such as methanol and water extracts were less effective towards the bacterial species compared to the non polar extracts. In this research, extract treated cells showed higher
level of ROS production compared to untreated control, which indicated that ROS production could be the possible cause or consequence of cellular damages. For *M. pruriens*, the activity of the fractions was much lower than crude extracts against the test bacterial species. For *L. salicifolia*, both crude and Fraction 1 showed strong antibacterial activity, but the pure compound isolated from the active fraction did not turn out to be the major active antibacterial compound suggesting involvement of other compounds present in the fraction. The plants that showed antibacterial activity in this study scientifically validate the ethnomedicinal use of these plants in treatment of infectious diseases including pneumonia and these plants may also provide clues to prepare new potent antibacterial variants through synthetic route.