2. Review of Literature

2.1. Ethnobotanical study

Ethnobotanical knowledge is proving to be in valuable for drug discovery in the wake of effective prospecting from biodiversity (Barkatullah 2015). On the other hand, the escalating human pressure is threatening the endogenous flora. Situated at the foot hill of the Himalayas, Pakistan boasts of rich floristic distribution. However, many lush yet imperiled regions of this country has never been explored. It inspired us to evaluate and document the taxonomic composition, significance of medicinal plants and associated traditional knowledge in the District of Malakand, Khyber Pakhtunkhwa Province. Vegetation growing in Malakand pass hills, Pakistan was studied and data were collected using an open-ended questionnaire, in addition to interviewing the local elderly, knowledgeable persons, and herbal practitioners. Relative Frequency Citation (RFC) and Use Value (UV) of the medicinal plants were calculated and their correlation was determined by Pearson correlation coefficient. This study encompasses 92 plant species belonging to 56 families thriving in the study area. The information gathered includes ethnobotanical inventory and their pharmacological uses. Quantitative analysis throws light on the consistence of RFC and UV. Asteraceae and Lamiaceae were the most abundant families represented by 6 species each. Shoots were the most used parts (23.6%) and wound healing (7.91%) was the most common therapeutic use. The result obtained from the study implies that local in habitants rely on these plants for their medicinal requirements. Also, the statistics reveal that, the vegetation can be assessed for potential drug leads. However, urban expansion is threatening the existence of indigenous flora and old generation with ancient herbal wisdom is perishing. So, it appears imperative to preserve the traditional knowledge. This survey is expected to contribute to the discovery of novel bioactive constituents, stimulate conservation efforts of the perturbed flora and promote sustainable exploitation of the medicinal bounty.
Xavier et al, (2014) documented the traditional healers who are practicing herbal medicine among the Kani tribals in Thodu hills of Kerala, India and quantitatively document their indigenous knowledge on the utilization of medicinal plants, particularly most common ethnomedicinal plants. A field study was carried out over a period of 1 year in Thodu hills. The ethnomedicinal information was collected through interviews among the Kani traditional healers. The collected data were analyzed through use value (UV), informant consensus factor (Fic), fidelity level (FL) and relative importance (RI). Results: During the present study a total of 35 medicinal plant species belonging to 28 families and 34 genera have been documented. These plants were used to treat various diseases and ailments grouped under 14 disease categories, with the highest number of species (7) being used for liver problems, circulatory system and dermatological disorders, followed by skeleto muscular system disorders (6), and fever (5). In the study area the informant consensus about usages of medicinal plants ranges from 0.70 to 1 with an average value of 0.83. Herbs (46%) were the primary source of medicine, followed by shrubs (23%). *Plumbago zeylanica* (UV of 1.86) and *Ocimum tenuiflorum* (UV of 1.57) are the most frequently and popularly used medicinal plant species in the study area. *Aristolochia tagala* is rare and a vulnerable climber, *Curculigo orchioides, Elephantopus scaber, Helicteres isora, Smilax zeylanica* and *Strychnos nux-vomica* are rare species which need to be conserved for future use. The high degree of consensus among the informants suggests that the current use and knowledge is still strong. The efficacy and safety of all the reported ethnomedicinal plants needs to be evaluated for phytochemical and pharmacological studies, especially the plants with high informant consensus factor, use value and fidelity level should be given priority to carry out bioassay and toxicity studies. They recommend the plants *Plumbago zeylanica, Ocimum tenuiflorum, Artocarpus hirsutus, Andropogon muricatus, Helicteres isora, Coscinium fenestratum* and *Justicia adhatoda* with high UV and RI values. *Biophytum sensitivum, Curculigo orchioides, Strychnos nux-vomica, Gossypium hirsutum, Artocarpus heterophyllus, Elephantopus*
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scaber, Pergularia daemia and Pyrrosia heterophylla (newly reported claims with highest FL) for further ethnopharmacological studies for the discovery of potential new drugs.

Baceta et al, (2014) reported that the recorded and analyzed medicinal plants used in the folk medicine of the Northwest of the Basque Country focusing on how medicinal plants knowledge and practices evolve. Field work consisted of 265 orally consented semi-structured interviews with 207 informants about medicinal uses of plants. Interviews were conducted between September 2008 and January 2011. Informants were on average 76 years old (minimum 45, maximum 95), being more than half of them (112) men. Data collected were structured in use reports (UR). Following informants 0 comments, medicinal use reports were classified as abandoned-UR, when the informants reported that the use was only practiced in the past, and prevalent-UR, when the informants reported to continue the practice. A total of 2067 UR for 139 species that belong to 58 botanical families were recorded, being the most important families Asteraceae, Liliaceae sensu latu and Urticaceae. Some of the most important species are commonly used in other European areas (e.g., Chamaemelum nobile, Urtica dioica and Chelidonium majus). However, there are also plants commonly used in the area such as Helleborus viridis or Coronopus didymus, that are scarcely used in other areas, and whose record is an original contribution of the local pharmacopeia. It is also the case of remedies such as the use of Plantago leaves against strains in a local remedy called zantiritu. Overall, and for all variables analysed (total UR, medicinal use-categories, drug preparation and administration), the percentage of UR being currently practiced (prevalence ratio) was very low (near 30%) suggesting a strong decay in the use of traditional medicinal plants. Exceptionally, some species (Chamaemelum nobile, Verbena officinalis or Anagallis arvensis) have a high prevalence ratio, reflecting the fact that this erosion process is not evolving homogeneously. Informants also reported that new species and medicinal plant uses were entering into the local pharmacopeia via non-traditional sources such as books, courses, or the internet. These modern ways are now being used to spread some traditional remedies that in the past were only orally transmitted. This study shows that traditional knowledge is
continuously changing, evolving and adapting to the new social and environmental conditions. The image of the local folk medicine as a dying reality doomed to disappear should be reviewed. It also shows the need of a culturally sensitive approach by the official health systems to these practices.

Different communities throughout the world have specialized and profound knowledge on the use medicinal plants for various diseases. However, the detailed information on the respective use may extinct in near future as this knowledge is passed only orally among generations in most of the communities. Omwenga et al, (2015) pointed out the use of medicinal plants by traditional healers from the Kisii community, Borabu sub-county in Nyamira county, Kenya, to treat infections of the urinary tract, oral cavity, gastrointestinal system and the skin and to evaluate the social context in which the healers work and practice. Validated questionnaires were applied to 50 traditional healers in the study region, followed by interviews and structured conversations. Information on the relevant traditionally used medicinal plants and their use were documented, including sampling and identification of voucher specimens. The ethnopharmacological survey revealed 25 medicinal plant species belonging to19 families. It got evident that most of these species will be extinct in the near future unless appropriate measures are taken, as it turned out difficult to collect some of the wild growing species. *Elaeodendron buchananii* Loes, *Erlangea marginata*, *S. Moore*, *Acacia gerrardii* Benth., *Balanites orbicularis* Sprague, *Solanum renschii* Vatke and *Orthosiphon hildebrandtii* Vatke have not been described before for its medicinal use. Among the 25 species collected from the various regions of Borabu sub-county *Urtica dioica* L. was the only medicinal plant that was collected from all regions. In contrast *Erythrina abyssinica* and *Rhus natalensis* were found in only two regions of the study area. The traditional medicinal use of the reported plants for infections should be documented and a great need of awareness from scientists and local government for improved preservation or field cultivation of some species is obvious.
Islam et al. (2014) reported the Madhupur forest area, Tangail is one of early human settlements in Bangladesh. Having abode in the vicinity of the forest, a strong ethnobotanical practice has prevailed in this area since ancient time. Due to the rapid deforestation during the last few decades, many plants have already disappeared or are facing extinction. Thus, they attempted to document the medicinal plant use of Madhupur forest area with a view to preserve the ethnobotanical knowledge and in order to protect the biodiversity of this area. The field work was conducted during a period of 1 year. Data was collected by interview, questionnaire, and group discussion with randomly selected informants including indigenous, tribal people, and Traditional Health Practitioners (THPs) living in the study area. Recorded plants are listed along with their indication, part used, form of preparation and use value (UV). Results were also analyzed to determine informant consensus factor (ICF) and fidelity level (FL) of the plants on the basis of their use under various ailment categories. The present study has documented 78 medicinal plant species from 45 families used for the treatment of at least 77 different major and minor ailments and conditions. Medicinal plant species were categorized as tree, shrub, tuber, herb, and climber. Leaves were found to be the most frequently used plant part while decoction is the major form of preparation. In most cases preparations are either administered orally or applied topically. Plants with high ICF and FL values can be subjected to bioassay guided investigation while plants which scored low UV require bioactivity screening to justify their use for the reported ailment.

The use of medicinal plants in human health has been documented since ancient times and they provide a useful source of new therapeutics. In Singapore, despite the accessibility to modern health care, there still exist pockets of the population who choose to use locally grown fresh medicinal plants for health promotion and even therapeutic purposes. However to date, there is no published report of first-hand account of their usage in Singapore. As land is scarce and rapidly used for re-development, such important knowledge may be lost if not properly documented in time. This work regards the local folk knowledge, and provides information on common and scarcely reported fresh
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medicinal plants. The objective of their study is together information regarding the usage of fresh medicinal plants in Singapore through face-to-face interviews. Information on demographic data and plant use methods were collated via face- to-face interviews of 200 fresh medicinal plant users who have used fresh medicinal plants in the last five years. The survey protocol was approved by the National University of Singapore Institutional Review Board and informed consent was obtained from every participant. A total of 414 plants represented by 104 plant species from 44 families were reportedly used by the 200 participants. The five most commonly used plants were Clinacanthus nutans (34 users), Strobilanthes crispus (31 users), Pereskia bleo (25 users), Aloe vera (18 users) and Zingiber officinale (16 users). Leaves were the most commonly used plant part while preparing a decoction was the most common method of preparation. The majority of interviewees used plants for general health purposes and to treat diseases related to the respiratory system and cancer. Siew et al, (2014) documented the rich wealth of traditional usage and knowledge on 414 fresh medicinal plants grown in Singapore through face-to-face interviews with 200 users.

Sri Lanka has rich traditional systems of medicine, which cater to 60-70% of the rural population’s primary health care needs. However, development of existing systems has been hindered by the unavailability of up-to-date information on medicinal materials and other related issues. For streamlining purposes, we investigated the present-day scenario of country's medicinal plant industry by gathering up-to-date information on the types of raw materials required, their aggregate quantities, heavily used and rare materials, family wise distribution, challenges faced by stakeholders as well as other pertinent issues. Kankanamalage et al., 2014 selected government Ayurveda hospitals, traditional and Ayurveda practitioners, large and small-scale herbal drug and cosmetic manufactures, importers, collectors and Ayurveda commissioners throughout the country. A systematic questionnaire was distributed and face-to-face interviews were conducted. A diverse range of medicinal materials, including 290 species (64.73%) from dried plants, 59 (13.17%) from fresh plants, 69 (15.40%) from minerals, 18 (4.02%) from animal sources and 12
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(2.68%) from other sources were recorded. A total of 302 plant species belonging to 95 families, dominated by Leguminosae family, was listed. Out of these, 46 species belonging to 35 families were used intensively. A large portion of herbal materials was of completely local origin (71.13%) while 26% were imported and the rest (2.87%) can be obtained by both routes. Leaves were the most highly used part of the plant (22.2%). High price, poor quality, insufficient or totally absence of continuous supply and adulteration were the main constraints faced by the stake holders. The unavailability of systematic cultivation and processing protocols, in correct identification, and lack of proper quality control methodologies were identified as major challenges of the industry.

Savikin et al, (2013) documented the ethnobotanical information on medicinal plant uses in the Zlatibor district, South-Western Serbia. A survey was performed using questionnaires with 220 informants (mean age 47, 79% female, 21% male). In addition, the use value and the relative importance of species were determined and the informant consensus factor was calculated for the medicinal plants included in the study. Intended plants usage was compared with previous ethnobotanical literature, with reference to the neighboring areas of Zlatibor district. The informants provided data for 69 medicinal plants belonging to 36 families. Rosaceae, Lamiaceae and Asteraceae were the predominant locally used families. The species with the highest use Value were Mentha piperita, Matricaria chamomilla, Hypericum perforatum and Achillea millefolium. The most frequently reported medicinal uses were ones for treating gastrointestinal ailments, respiratory problems and skin diseases. Usually, the administration was primarily oral followed by topical applications. All different plant parts were utilized, however leaves were the most exploited parts of the plants. Folk medicine in South-Western Serbia, Zlatibor district is intended mainly as a mode of primary health care in healing of minor illnesses. The results indicate a slight reduction in the ethnobotanical and medical knowledge in this area, when compared with neighboring regions.
Boudjelal et al, (2013) pointed out large number of wild medicinal plants used in the M’Sila region (northern Algeria) for the treatment of several human pathologies. Another more ambitious aim is to contribute to overcoming the limits of an orally transmitted pharmacopoeia, attempting to exploit the large ethnopharmacology patrimony of the region for further pharmacological purposes. Our field study was carried out over a period of three years (2008–2010). During this period, herbalists were interviewed using semi-structured questionnaires investigating the herbalist as a holder of information (gender, age and educational level) and about wild medicinal plants (local name, uses and part used). In addition, the relative importance value of the species was determined and informant consensus factor (ICF) was calculated for the medicinal plants included in the study. A total of 83 herbalists were interviewed; men dominate the practice of traditional medicine in the region. About 41% of them are between 31 and 40 years, and about a third (34%) is illiterate. The traditional herbal knowledge is passed from generation to generation in the verbal form, a writing tradition being almost totally absent. The interviewed herbalists identified and recorded 58 plants species and 50 genera belonging to 27 plant families. Lamiaceae and Asteraceae were the most represented plant families. The aerial parts were the most commonly used plant part, while infusion and decoction were the most common method of traditional drug preparation. The survey provides avertable source of information on the herbalists and wild medicinal plants. Plants which are used in different parts of the world for the treatment of similar diseases may be deemed to be effective in pharmacological terms. These medicinal plants may be incorporated into the healthcare delivery system of the country.

Choudhury et al, (2012) reported the medicinal plants used by the Chorei tribe residing in Southern Assam part of North Eastern India in the treatment of various ailments. Systematic and intensive field surveys were conducted in Chorei inhabited parts of Southern Assam part of North East India to collect information on medicinal plants used by them in treatment of various ailments. Data was collected through structured questionnaires and personal observations made during the field visit. A total of 53
different medicinal plants were recorded along with their vernacular names, parts used and mode of utilization by the Chorei tribes. Each of the plants was categorized according to their use in treatment of particular disease. The Chorei tribe is primarily dependent of medicinal plant for treatment of various ailments.

Sharmila et al, (2014) pointed out the folklore medically important plants frequently used to treat different ailments by Toda tribe living in Thiashola, Manjoor, Western Ghats. During survey, the experienced old folk of these communities were interacted for gathering information. A total of 57 herbaceous medicinal plants from 54 genera under 29 families were enumerated. The above enumerated species are being practiced by the Tribals for treatment of various diseases like skin diseases (*Centella asiatica, Erigeron karvinskianus, Helichrysum hookeriana*), cancer (*Cayratia pedata var. glabra*), rheumatism (*Anemone rivularis* and *Toddalia asiatica* var. floribunda), jaundice (*Physalis peruviana* and *Cynoglossum zeylanicum*) and so on. The treatment given by them is found to be effective. Plants of Asteraceae (10 species) was largely represented followed by Poaceae and Urticaceae (5 species). The plants may be useful under rural healthcare system and for herbal drug industry.

Prasad and Shyma, (2013) documented the traditional herbal drugs in the treatment of human and veterinary ailments by the tribal communities residing in Vythiri taluk of Wayanad district, Kerala in India. An ethno-medico-botanical survey was carried out in 6 randomly selected panchayaths of Vythiri taluk of Wayanad district, Kerala investigating the tribal healing systems and herbal treatments used for various ailments. Semi structured questionnaires were used for data collection. A total of 67 medicinal preparations involving 67 species belonging to 44 families, used by the Kurichia, Kuruma, Adiyan, Kattunaika and Paniya tribes to treat 33 human and 7 veterinary ailments were documented. Among these, the use of dried powdered form of *Amorphophallus paeoniifolius* and the leaf powder of *Memecylon umbellatum* as an antidiabetic drug and the whole plant of *Hydrocotyle javanica* for the treatment of fits and asthma are noteworthy. The plants used for the same disease is distinct from one tribe to another of
the same locality. The documentation of this information will be vital in future for sustainable utilization and conservation.

Rafael et al, (2014) reported that vaginal health, defined as the vaginal state in which the physiological condition remains stable, being protected from the onset of symptoms and facilitating a satisfying sex life, is one of the most common and less valued concerns in postmenopausal women. Many of the condition that affect the vagina are related to its trophism and susceptibility to infection by unusual germs, which are phenomena strongly influenced by estrogen impregnation and the microbiota composition, ultimately affecting sexuality and the quality of life. An expert panel of the Spanish Menopause Society met to establish criteria for diagnosing and treating the processes that affect overall vaginal health and to decide the optimal timing and methods based on the best evidence available.

Naidoo et al, (2013) investigated the in vitro antimicrobial activity and cytotoxicity profiles of a selection (individual plants and selected combinations) of traditionally used plants in this study area. Aqueous and organic (dichloromethane: methanol, 1:1) extracts were prepared. Antimicrobial activity was assessed using the minimum inhibitory concentration (MIC) assay against the STI associated pathogens; *Candida albicans* ATCC 10231, *Ureaplasma urealyticum* clinical strain, *Oligella ureolytica* ATCC 43534, *Trichomonas vaginalis* clinical strain, *Gardnerella vaginalis* ATCC 14018 and *Neisseria gonorrhoeae* ATCC 19424. For the combination study, interactions were assessed using the fractional inhibitory concentration (ΣFIC). The plant species were assessed for safety using the fractional inhibitory concentration (ΣFIC). The plant species were assessed for safety using the 3-[4, 5-dimethyl-2-thiazol-yl]-2,5-diphenyl-2H-tetrazolium bromide (MTT) cellular viability assay on the human embryonic kidney epithelial (Graham, HEK-293) cell line. *U. urealyticum* was the most sensitive of the six test organisms, with the aqueous extract of *Ranunculus multifidus* (0.02 mg/ml) and the organic extract of *Peltophorum africanum* (0.04 mg/ml) being the most antimicrobially active plant species studied. *Sclerocarya birrea* was found to have the broadest spectrum of activity (mean MIC of 0.89 mg/ml). The only plant species to exhibit some degree of cytotoxicity against the
kidney epithelial cell line was *Kigelia africana* (100 µg/ml), with 22% and 16% cell death for the aqueous and organic extracts, respectively. Of the 13 combinations studied, several synergistic combinations were evident, the most prominent being the combination of *Albizia adianthifolia* and *Trichilia dregeana* (aqueous extract) with an ΣFIC value of 0.15 against *O. ureolytica*. Synergistic interactions were observed regardless of the ratio of the aqueous mixtures of the two plants. *Syzygium cordatum* and *S. birrea* (aqueous extract) was also a combination of interest, demonstrating synergistic (ΣFIC=0.42) interactions against *O. ureolytica*. This combination, however, also displayed some cytotoxicity towards the human epithelial cell line.

Plants with numerous efficacious observations have historically been used as a starting point in the development of new drugs, and a large percentage of modern pharmaceuticals have been derived from medicinal plants. Hugo and Crystle, (2014) stated that the all plant use mentioned specifically for female healthcare, such as medicine to increase fertility, induce menstruation or abortion, ease pregnancy and parturition, reduce menstrual bleeding and postpartum hemorrhage, alleviate menstrual, parturition and postpartum pain, increase or inhibit lactation, and treat mastitis and uterine prolapse, in 200 studies focusing on medicinal plant use, either general studies or studies focusing specifically on women’s healthcare. Nearly 2000 different plant species are reported to be used in over 5000 combinations. Most common are *Achyranthes aspera*, *Artemisia vulgaris*, *Blumea balsamifera*, *Carica papaya*, *Curcuma longa*, *Hibiscus rosa-sinensis*, *Leonurus japonicus*, *Psidium guajava* and *Ricinus communis* each of these species had been reported in more than 10 different scientific articles.

Wet et al, (2012) conducted the ethnobotanical study on plants used for the treatment of sexually transmitted infections was undertaken to document the knowledge by lay people in a rural community in northern Maputaland, South Africa. The focus was on the medicinal plants which are growing in and around the immediate vicinity of the homesteads. Thirty three plant species were recorded as being used for the treatment of
sexually transmitted infections such as gonorrhoea (drop or ugcusulu), external and internal sores caused by sexually transmitted infections, genital warts (cauliflower or umhluma) and syphilis. Nine plants (*Bridelia cathartica* subsp. *cathartica*, *Cladostemon kirkii*, *Erianthemum dregei*, *Euphorbia hypericifolia*, *Ipomoea batatas*, *Krauseola mosambicina*, *Mimusops caffra*, *Opuntia stricta* and *Sarcophyte sanguinea* subsp. *sanguinea*) were recorded for the first time in the literature world wide as a treatment for sexually transmitted infections. Five new vernacular names were documented for *B.cathartica* subsp. *cathartica*, *Bryophyllum pinnatum*, *Clematis brachiata*, *E. hypericifolia* and *Pyrenacantha kaurabassana*. The 33 plant species are used in 23 different combinations of two or more plants per herbal remedy. The three most frequently used plant species in the study area for the treatment of sexually transmitted infections are; *Hypoxis hemerocallidia*, *Senecio serratuloides* and *Ranunculus multifidus*. Roots are mostly prepared, as a decoction which is taken orally or used as an enema. All eighty of the interviewees preferred traditional medicine as the first therapeutic choice for treating sexually transmitted infections. The wide variety of plants that are used to treat sexually transmitted infections in this area emphasises the importance that medicinal plants can have in the primary health care system of the rural people in northern Maputaland (KwaZulu–Natal).

Srithi et al, (2012) documented the traditional medical practices and determined which of the species used are culturally important among the Hmong. They interviewed six key informants and 147 non-specialist informants about their traditional knowledge of medicinal plants used in Hmong women’s healthcare. They are selected nine species that were known in all three villages as the domain for questionnaire interviews with 181 additional and randomly selected non-specialist informants. They calculated the Cultural Importance index (CI) for each species and use category. They tested normality of the data, age correlations, and gender correlation with Kolmogorov-Smirnov tests, Spearman’s rank correlation coefficient, Kruskal-Wallis test, an Mann–Whitney tests. They are documented traditional knowledge of 79 medicinal plants used in women’s healthcare. Of
these, three species were culturally important to the Hmong. Our questionnaire interviews revealed significant difference in traditional medicinal plant knowledge between genders and age groups. The Hmong people in northern Thailand possess large amounts of traditional knowledge related to women’s healthcare and plants used for this purpose. However, this knowledge, even for the culturally important species, is not possessed by all Hmong and there were signs of knowledge erosion. Preservation of the Hmong intellectual heritage related to medicinal plants used in women’s healthcare requires intensive traditional knowledge dissemination to the young Hmong generation.

Nagata et al, (2011) documented and identified the herbal medicines used by persons living with HIV/AIDS on Mfangano Island, Suba District, Nyanza Province, Kenya. We interviewed herbalists and knowledgeable mothers to obtain information regarding medicinal plants, particularly for HIV/AIDS-related symptoms, HIV/AIDS, and chira (an illness concept with similarities to HIV/AIDS regarding sexual transmission and wasting symptoms). Using systematic sampling, 67 persons living with HIV/AIDS (49 of whom were receiving ART) were selected from an Mfangano Island health clinic and participated in semi-structured interviews. Interviews with herbalists and mothers identified 40 plant species in 37 genera and 29 families that a person with HIV/AIDS or chira could use for herbal remedies. Overall, 70.1% of persons living with HIV/AIDS had used medicinal plants after HIV diagnosis, most commonly to treat symptoms related to HIV/AIDS. In addition to common vegetables and fruits that can serve medicinal purposes, Azadirachta indica A. Juss. (Meliaceae), Carissa edulis (Forssk.) Vahl (Apocynaceae), and Ximenia Americana L. (Olacaceae) were the most frequently cited medicinal plants used by persons living with HIV/AIDS. Collaboration and communication between biomedical clinicians and herbalists should be encouraged given high rates of concomitant ART-herb usage. Pharmacological, toxicological, and ART herb interaction studies based on the plants identified in this study and their constituent ingredients should be considered.
Telefo et al, (2011) reported that collecting and documenting information on herbal remedies traditionally used for the treatment of women infertility in Baham subdivision (Western Region of Cameroon). Fieldwork was undertaken as an ethnopharmacological survey involving thirty-two traditional medicine practitioners interviewed in 8 villages of the Baham sub-division. Personal information on interviewees as well as issues related to medicinal use of plants were recorded using structured questionnaires. A literature investigation on the therapeutic or pharmacological properties of recorded medicinal plants was further undertaken. From this inventory, a total of 46 plant species belonging to 43 genera and 26 families have been registered. These plants are used in 32 recipes and prepared as maceration (43%) or decoction (40%) of only one plant (25%) or of the mixture of two (22%), three (28%), four (22%) or even seven (3%) medicinal plants. Globally, they are given orally during 30 days, at an average dosage of two glasses per day. The literature confirms the use of the majority of these plants for the treatment of the woman infertility and illnesses that are associated to it.

2.2. Antimicrobial assay

Mulaudzi et al, (2013) reported that the anti-bacterial, anti-fungal, anti-gonococcal, HIV-type 1 reverse transcriptase (RT) and to determine phenolic content of twelve medicinal plants used by the Venda people to treat venereal diseases. Twelve medicinal plants were extracted with petroleum ether (PE), dichloromethane (DCM), 80% ethanol (EtOH) and water. The extracts were evaluated for their antimicrobial properties against two Gram-positive (Bacillus subtilis and Staphylococcus aureus), three Gram-negative (Neisseria gonorrhoeae, Escherichia coli and Klebsiella pneumoniae) bacteria and a fungus Candida albicans. The phenolic contents including total phenolics, flavonoids, gallotannins and condensed tannins of the methanolic extracts of the same plants were also determined. DCM and EtOH extracts of Bolusanthus speciosus bark and stems exhibited good activity (<0.1 mg/ml) against Escherichia coli and Staphylococcus aureus with MIC values ranging from 0.098 to 0.012 mg/ml. The DCM extracts of Bolusanthus speciosus
bark showed the best activity against *Candida albicans* with MIC and MFC values of 0.012 mg/ml. Water and 50% methanol extracts of *Acacia karroo* bark, *Bolusanthus speciosus* stems and *Ximenia caffra* roots and leaves showed good RT inhibition percentages (>70%) at 1 mg/ml. All tested extracts exhibited dose dependent IC50 values ranging from (0.1 to 0.6 mg/ml). Almost all plant species investigated contained phenolic compounds, which were greater than 5 mg/g with the exception of *Adansonia digitata* bark and *Aloe chabaudii* roots. The highest level of flavonoids (11.9 μg/g) were detected in *Ximenia caffra* leaves. Whereas the highest amount of gallotannins were detected in *Ekebergia capensis* bark (69 μg/g). Condensed tannins were higher in *Ekebergia capensis* bark and *Ximenia caffra* roots (0.47 and 0.48% respectively). *Adansonia digitata* bark and *Aloe chabaudii* roots exhibited low levels of phenolic compounds as well as antimicrobial activities.

Jarriyawattanachaikul et al, (2016) reported the identifying natural compounds with anti-bacteria activity from twenty-six Thai-herbal plants. Antimicrobial activity was evaluated by an agar diffusion method, which allowed for the determination of the minimum inhibitory concentration (MIC). The results indicated that Thai-herbs have potent antimicrobial activity against *E. coli, S. aureus* and *C. jejuni* at bacteria suspensions of 2.0-3.0x10^9 CFU/ml. Interestingly, *C. formosum* had the highest antimicrobial activity against the three food-borne pathogens of *E. coli, S. aureus* and *C. jejuni*, which were isolated from the chicken-caecum. MIC values of *C. formosum* against *E. coli, S. aureus* and *C. jejuni* were 3.0 mg/ml, 3.0 mg/ml and 0.3 mg/ml, respectively. Other herbal plants also had antimicrobial activity against the three food-borne pathogens in this study. The herbal plants provide not only a natural source of anti-bacterial activity, but also antioxidant activity and anticancer properties. The application of using Thai-herbal plants compounds by adding them in animal feed is proposed. This may be a safe means of enhancing health and production of livestock and thus benefits humans and animals. Consequently, the selection of herbal plants, for use in preventing food-borne bacterial infection, is both interesting and worthwhile for food safety.
Galappathie et al., (2014) pointed out the traditional claims about medicinal plants collected by the Traditional knowledge documentation program at the Sarawak Biodiversity Centre on the island of Borneo. The majority of the medicinal plants are utilized as traditional therapies for various diseases, including diarrhoea, food poisoning, vaginomycosis, sexually transmitted infections (gonorrhoea) and furunculosis. Six medicinal plants used as indigenous herbal medicines were individually screened for antimicrobial and antifungal effects using their crude extracts and were found to inhibit a broad range of pathogenic microorganisms. Plant extracts derived from *Fibraurea tinctoria*, *Polyalthia hookeriana*, *Pyrenaria* sp., *Baccaurea lanceolata*, *Goniothalamus tapisoides* and *Goniothalamus velutinus* were demonstrated to have the highest antimicrobial activities. *Pyrenaria* sp. showed significant antifungal activity against *Candida albicans* with minimum inhibitory and minimum fungicidal concentrations of 25 g/mL and 50 g/mL, respectively.

Gupta et al., (2016) studied the antimicrobial activity and phytochemicals of extracts from 5 different medicinal plants, as well as to evaluate the synergistic activity of potent plant extracts with suitable antibiotic discs and antibiotics susceptibility of tested microorganisms. The antimicrobial activities of different extracts were evaluated by using agar well diffusion method and antibiotics susceptibility of five selected microorganisms was tested by using disc diffusion method. For determination of synergistic activities of the potent plant extracts along with antibiotic discs, agar well diffusion and disc diffusion methods were combinedly used. Their research investigation, the maximum in vitro inhibition of tested microorganisms, *Escherichia coli*, *Pseudomonas aeruginosa*, *Candida albicans* and *Staphylococcus aureus* were recorded in chloroform and methanol extracts of *Terminalia arjuna*, ethanol extract of *Camellia sinensis*, and petroleum ether extract of *Polyalthia longifolia* which offered inhibition zone ranged from 11 to 18 mm. The maximum antibacterial efficacy was exhibited by levofloxacin with an inhibition zone of 35 mm against *Escherichia coli*. The potent plant extracts showed positive synergistic effects against *Staphylococcus aureus* with lincomycin. The phytochemical analysis of the
potent plant extracts revealed the presence of saponin, tannin, protein, carbohydrate, flavonoid, terpenoids and glycosides.

Aumeeruddy-Elalfi et al, (2016) pointed out the antimicrobial properties of essential oils (EOs) extracted from 10 common medicinal plants of Mauritius. Eighteen microorganisms (bacterial and fungal isolates) were used to evaluate the antimicrobial potential of the EOs as well as their ability to potentiate conventional antibiotics. The phytochemical profile was established using Gas Chromatography-Mass Spectrometry method. Antibacterial activities were recorded with low minimal inhibitory concentration for 4 of the EOs using the microbroth dilution assay. A synergistic effect of the EO of Citrus hystrix D.C., Citrus reticulate (Blanco) and Melaleuca quinquenervia S.T. Blake (Cav.) were observed against *E. coli* (ATCC 25922) and *S. epidermidis* (ATCC 12228) when combined with gentamicin. Fungicidal and fungistatic effects of the EOs were observed among all the fungi irrespective of the family except for *T. mentagrophytes*. Twenty eight major compounds were identified and predominantly composed of monoterpenes hydrocarbons at a dose ranging from 0.68 to 88.58%. It provided key information on the antimicrobial property and phytochemical composition of some tropical medicinal plants. Hence, EOs studied in the present investigation may be considered as potential medicinal candidates that could be exploited as complementary and alternative therapies for the treatment and management of infectious diseases.

Sanjesh et al, (2013) studied the antifungal and antibacterial activity of six nature plants like *Sapindus emarginatus*, *Hibiscus rosa-sinensis*, *Mirabilis Jalapa*, *Euphorbia tirucalli L.*, *Vitex negundo L.*, and *Saussurea costus* of Methanol, Chloroform, N-Hexane and Water extracts on various *Aspergillus flavus*, *Candida albicans* and *Candida glabreta*, fungal strains *Bacillus subtilis*, *Escherichia coli* and *Staphylococcus epidermidis* bacterial strains using standard methods Agar tube dilution for antifungal, Agar diffusion assay for antibacterial as per the screening for extraction and evaluation of nature plants for anti-fungal and anti-bacterial activities for Minimum inhibitory concentration (µg/ml).
Among the six plants *Sapindus emarginatus, Hibiscus rosa-sinensis, Mirabilis jalapa, Euphorbia tirucalli L, Vitex negundo L* and *Saussurea costus* used four different solvents at antimicrobial strains for antibacterial and antifungal activity it showed the best report on *Sapindus emarginatus* and *Saussurea lappa Costus*, on n-hexane extract according to Minimum inhibitory concentration leaf it showed satisfactory result against various gram +ve and gram –ve bacteria and fungi on n-hexane extract showed the highest Minimum inhibitory concentration (µg/ml) 15-18 (µg/ml) while minimum inhibitory concentration showed the lowest report then other remained crude extract Minimum Inhibitory concentration for antimicrobial activity.

Agarwal and Prakash, (2014) studied the antibacterial activity of the aqueous and solvent extract of Red flowers of *Hibiscus rosa-sinensis*. The extract contains large amounts of phenolic compounds and flavonoids. The aqueous and solvent extracts of flowers of *H. rosa-sinensis* were screened for antibacterial activity by using disc diffusion method. The flower material can be taken as an alternative source of antibacterial agent against the human pathogens.

Al-Alak et al, (2015) studied the antibacterial activity of *Hibiscus rosa-sinensis* extract and Synergistic Effect with Amoxicillin against some human pathogens. Test microorganism for antibacterial assay (Agar well diffusion method) (Influence of combination between antibiotics and plant extracts against selected multi drug resistant strains). A total of five clinical isolates (*P. aeruginosa, Serratia, Micrococcus, Enterobacter and Salmonella*) belonging to different bacterial species were collected from the department of biology, college of science, Al-Mustansirya university, Baghdad, Iraq. Co-trimoxazol, Tetracycline, Lincomycin, *Enterobacter* isolate was resistant to Amikacin. All bacterial isolates were sensitive to Ciprofloxacin, Norfloxacin. The minimum inhibitory concentrations (MICs) of Amoxicillin most effective antibiotics were tested against the five MDR isolates. Their study only the susceptibilities of clinical bacterial isolates to the extracts of *H. rosa-sinensis* leaves and flowers have been examined. Therefore, further microbiological studies would be carried out to determine the minimal
inhibitory concentrations (MICs) of the extracts prepared from *H. rosa-sinensis* against (*P. aeruginosa, Serratia, Micrococcus, Enterobacter* and *Salmonella*). The water extracts of *H. rosa-sinensis* was evaluated against isolates and the results showed that the crude extracts of them with boiling water were more effective than the cold watery extracts. Of this work the combination between *H. rosa-sinensis* and Amoxycillin showed synergistic effect against the tested bacteria than each of them alone.

*Hibiscus rosa sinensis* is member of the family Malvaceae and grows as an evergreen herbaceous plant in tropical regions. Hibiscus species are used in the treatment of various diseases. They have many pharmacological properties including antipyretic, antispasmodic, hypotensive, antifungal, anti-inflammatory and many more. Antibacterial activity of methanolic extract from the leaves and flowers of *H. rosa sinesis* and identification of secondary metabolites in these extracts. The phytochemical analyses showed the presence of alkaloids, glycosides, flavonoids, tannin and phenols in hibiscus leaf extract, while hibiscus flower extract contained alkaloids, protein, steroid and carbohydrate. Antibacterial activity has been checked in terms of zone of inhibition by disc diffusion method against microorganisms *E.coli* and *S. aureus* for different concentrations of methanolic leaf and flower extracts ranging from 31.25 to 500 mg/disc which were compared with positive control gentamicin (1mg/disc). Both extracts showed increasing antibacterial property with increase in the extract concentration. Maximum zone of inhibition observed for both methanolic leaf and flower extracts of *H. rosa sinesis* at concentration 500 mg for *E. coli* 23+1.01 mm and 13.75+0.99 mm, respectively. However, for *S.aureus* methanolic leaf and flower extracts of *H. rosa sinesis* at concentration 500 mg showed maximum zone of inhibition 19.33+0.29 mm and 9.75+0.76 mm (Tiwari et al., 2015).

Ferreira et al, (2013) reported the anti-fungal activity of 10 medicinal plants from northeastern Brazil, traditionally used as anti-infective agents. The activity of 30 crude extracts (water; ethanol:water, 1:1; acetone:water, 1:1) against four standard species of Candida yeasts (*Candida albicans* ATCC 90028, *Candida dubliniensis* ATCC 7289,
Candida glabrata ATCC 2001 and Candida krusei ATCC 6258) was investigated by the Minimal Inhibitory Concentration (MIC), using the microdilution method and the working range used was from 1.95 to 1000 μg/mL. Extracts from leaves of Eugenia uniflora (Myrtaceae), stem bark of Caesalpinia ferrea (Caesalpinaceae) and leaves of Psidium guajava (Myrtaceae) showed significant activity against all yeasts evaluated. The best antifungal activities were achieved against C. glabrata and C. krusei by E. uniflora extract (MIC = 15.62) and followed by extracts from C. ferrea and P. guajava (MIC ranged from 15.62 to 250 μg/mL). E. uniflora also showed fungicidal properties against all yeasts, especially against Candida dubliniensis.

Uddin et al, (2013) studied the antibacterial activities of some traditional medicinal plants on several pathogenic bacteria, which can cause diseases in human. Thirty four medicinal plants belonging to twenty-four families were selected based on medicinal reports practiced by the indigenous people and screened for their antibacterial activity against eight human pathogenic bacteria (Bacillus subtilis, B. megaterium, B. cereus, Staphylococcus aureus, Escherichia coli, Vibrio cholerae, Salmonella typhi and Shigella dysenteriae) by disc diffusion and agar cup methods. Among them Psidium guajava, Terminalia arjuna, Phyllanthus embelica, Terminalia chebula, Justicia adhatoda, and Ocimum sanctum showed significant antibacterial activity against the human pathogenic bacteria. The largest zones of inhibition (22 mm in diameter) were recorded against S. dysenteriae and B. cereus with the fruit extracts of O. sactum.

Khan et al, (2013) studied the plants Bergenia ciliata, Jasminum officinale, and Santalum album for their potential activity against human bacterial pathogens. B. ciliata, J. officinale, and S. album extracts were prepared in cold and hot water. The activity of plant extracts and selected antibiotics was evaluated against five bacterial pathogens including Staphylococcus aureus, Bacillus subtilis, Proteus vulgaris, Pseudomonas aeruginosa, and Escherichia coli using agar well diffusion method. Among the three medicinal plants, B. ciliata extracts displayed potential activity against bacterial pathogens. Cold water extract of Bergenia ciliate showed the highest activity against B. subtilis, which is comparable
with a zone of inhibition exhibited by ceftriaxone and erythromycin. *J. officinale* and *S. album* extracts demonstrated variable antibacterial activity. Further studies are needed to explore the novel antibacterial bioactive molecules.

### 2.3. Gas chromatography and Mass spectroscopy (GC-MS)

Paramanantham and Murugesan, (2014) investigated the phytoconstituents of ethyl acetate extract of *Holarrhena antidysenterica* using GC-MS. GC-MS analysis of flowers extract were performed using a Thermo GC-Trace Ultra VERSION:5.0 interfaced to a Thermo Mass Spectrometer DSQ II, fused Silica Capillary Column (30mmX 0.25mm 1DX1 eMdf, composed of 100% Dimethyl polysiloxane). The results of the GC-MS analysis confirmed the presence of thirty compounds. The most prevailing compounds are- 1-Eicosanol, Hexadecane, Sigmastane 1,3-dione, Behenic alcohol etc. From the results, it can be concluded that the flowers extract show the presence of 30 phyto compounds.

Wei et al, (2014) documented the essential oil composition of the flower of *Jasminum officinale* L. var. *Grandiflorum* L. (*Jasminum grandiflorum*) by gas chromatography-mass spectrometry (GC-MS). The optimum GC-MS conditions used for the analysis were 250°C inlet temperature, 150°C MSD detector temperature, and GC oven temperature program as follows: 100°C initial temperature, increased to 270°C at 4°C/min, final temperature 270°C and held for 7.5 min. Thirty compounds were identified, representing 99.28 % of the oil content. The major volatile components of the flower were 3,7,11,15- tetramethyl-2-hexadecen-1-ol (phytol) (25.77 %), 3,7,11- trimethylidodeca -1,6,10-trien-3-ol (12.54 %) and 3,7,11,15- tetramethyl -1-Hexadecen-3-ol (12.42 %). The results show that phytol is the major volatile component of *Jasminum grandiflorum*.

Inikpi et al, (2014) reported the chemical compositions of essential oil obtained from the hydrodistillation of air-dried flowers of *Hibiscus sabdariffa* L (Malvaceae) are reported. The components of the essential oils were analysed by means of gas chromatography (GC) and gas chromatography coupled with mass spectrometry (GCMS). The major compounds identified in the essential oil were hexadecanoic acid (64.3%) and linoleic acid (22.7%).
**Calotropis gigantea** white (Asclepiadaceae), is a weed plant commonly known as giant milk weed. It has one of the important traditional medicines to treat various ailments. Rajamohan et al, (2014) reported that the phytochemicals present in the flower extract by GC-MS analysis. The results showed the presence of phytochemicals of alkaloids, tannins, phenol, flavonoids, sterols, anthraquinones, proteins and quinones in the flower extract. The GC-MS analysis of the extract revealed the presence of 4 major compounds.

Kanthal et al, (2013) reported the phytochemical constituents from species of the Compositae (Asteraceae). Hitherto no reports exist on the phytochemical components and biological activity of *Lactuca runcinata* DC. Phytochemical screening of the entire herb of *Lactuca runcinata* DC revealed the presence of some bio-active components. Gas chromatography-mass spectrometry (GC-MS) analysis of the whole plant methanol extract of *Lactuca runcinata* was performed on a GC-MS equipment (Thermo Scientific Co.) Thermo GC-TRACE ultra ver.: 5.0, Thermo MS DSQ II. Results: The phytochemical tests showed the presence of alkaloids, cardiac glycosides, flavonoids, phenols, phlobatannin, reducing sugars, saponins, steroids, tannins, terpenoids, volatile oils, carbohydrates, and protein/amino acids in methanolic extract of *Lactuca runcinata*. The GC-MS analysis has shown the presence of different phytochemical compounds in the methanolic extract of *Lactuca runcinata*. A total of 21 compounds were identified representing 84.49% of total methanolic extract composition.

Medicinal plants are at great interest to the researcher in the field of biotechnology, as most of the drug industries depend in medicinal plants for the production of pharmaceutical compounds. Plants are the traditional sources for many chemicals used as pharmaceutical biochemicals, fragrances, food colours and flavours in different countries especially in India. Most herbal medicines and their derivative products were often prepared from crude plant extracts, which comprise a complex mixture of different phytochemical constituents (plant secondary metabolites). The chemical features of these constituents differ considerably among different species. GC-MS method used for the analysis of the obtained extracts can be an interesting tool for testing the amount of some
Identification and characterization of antimicrobial compounds from selected ethnomedicinal plants of Silent Valley (Western Ghats, Kerala) with emphasis on Venereal diseases

active principles in herbs used in cosmetic, drugs, pharmaceutical or food industry. Gomathi et al, (2013) screened the identification of bioactive compounds from the whole plant ethanolic extract of *Evolvulus alsinoides* by Gas chromatography and Mass spectroscopy (GC-MS). GCMS analysis of ethanolic extract was done by standard protocol using the equipment Thermo GC-Trace Ultra Version: 5.0, Thermo MS DSQ II. The GCMS analysis revealed the presence of various compounds like piperine, octodeconoic acids, hexadecanoic acid and squalene in the ethanolic extract of *Evolvulus alsinoides*. Hence, the *Evolvulus alsinoides* may have chemopreventive, anticancer, antimicrobial activity, antioxidant and antidiabetic activity due to the presence of secondary metabolites in the ethanolic extract. Due to the presence of esters which can be used as a flavoring agent in food industries.

Nandagopalan et al, (2015) pointed out the possible chemical components from *Hibiscus tiliaceus* by GC-MS technique. This analysis revealed that the methanol extract contains N, N- Dimethylglycine (83.97%), 3, 7, 11, 15-Tetramethyl-2-hexadecen-1-ol (2.94%) and 4H-Pyran-4-one, 2, 3-dihydro-3, 5-dihydroxy-6-methyl- (2.69%). These constituents of plant extract provide the scientific evidences for the antimicrobial, anticancer, antioxidant and antidiabetic properties of the plant.

Wu et al, (2010) reported that the *Chrysanthemum indicum* flower is a traditional Chinese medicine with strong aroma and many previous studies focused on its essential oil. GC/MS and HPLC were used to determine its volatiles, flavonoids and flavonoid glycosides. Sixty three volatiles were detected and the abundant volatiles included 2,6,6-trimethyl-bicyclo[3.1.1]hept-2-en-4-ol, 2-(2,4-hexadiynylidene)-1,6-dioxaspiro[4.4]non-3-ene, germacrene D, á-neoclovene, eucalyptol, á-pinene. Ten flavonoids were identified. Quercitrin, myricetin and luteolin- 7-glucoside were abundant flavonoids. The bioactivities of the abundant components in *Chrysanthemum indicum* flower were discussed. It is considered that *Chrysanthemum indicum* flower is a good source of natural quercitrin and myricetin, which is significant for the development of potential pharmaceuticals.
2.4. Molecular Docking analysis

Archana et al, (2014) reported the malaria remains one of the major public health problems when *Plasmodium falciparum* is one of the causative agents. The dihydrofolate reductase (DHFR) is one of the well-defined targets of *P. falciparum* which is involved in the reproduction of this parasite. Proguanil is a prophylactic antimalarial drug; it stops the malaria parasite, from reproducing once it is in the red blood cells. It does this by inhibiting the enzyme, dihydrofolate reductase. The side effects of these drugs make the need for the necessity of new improved drugs. The present paper was framed to find active components through GC MS analysis and *insilico* docking studies with the identified compounds in the methanolic extract of *Chromolaena odorata* leaves to validate the antimalarial potential of these phytocompounds. There are 33 phytocompounds derived through GC-MS analysis, out of these 4 compounds satisfied the Lipinski’s properties. The docking studies of these compounds were done using commercial tool Accelyrs Discovery Studio 2.1. Among these compounds, Falcarinol showed the highest dock score of 71.128 with more hydrogen bond interactions.

Priya et al., (2014) studied the angiotensin converting enzyme (ACE) catalyses the conversion of angiotensin I to angiotensin II a potent vasoconstrictor in a substrate concentration dependent manner and degrades bradykinin a potent vasodilator and other vasoactive peptides which leads to increase in blood pressure. Prolonged increase in blood pressure condition increases the risk of heart attacks, heart failure, and stroke or kidney failure. Naturally occurring proteins acts as angiotensin converting enzyme inhibitors. Inhibition of ACE by angiotensin converting enzyme inhibitors results in the decreased of formation of angiotensin II and decreased metabolism of bradykinin leading to systematic dilation of the arteries and veins and a decrease in arterial blood pressure. The molecular docking analysis done indicates that the receptor of human angiotensin converting enzyme through an interaction with the chemical bonds. Herbal drugs were safe and milder with few or no side effects than the drugs currently used in the treatment lessening high blood
pressure which can be better used for the development of new therapeutics to decrease the formation of angiotensin II and to decrease the activation of bradykinin.

Vijayalakshmi et al, (2014) reported the in silico analysis to elucidate the inhibitory activity of retinol binding protein 4 (RBP4) using compounds from GC-MS analysis of Bauhinia variegata and Garcinia cambogia. These compounds were docked with Retinol binding protein 4. These chemical components having anti-lipidemic activity were subjected to in silico analysis. This study evaluates the inhibitory activity of compounds with Retinol binding protein 4 using Argus lab software. Compounds like Octadecanoic acid, Tertradecanoic acid, Hexadecanoic acid and Trans-Geranylgeraniol of Garcinia cambogia and compounds like phytol and Hexadecanoic acid of Bauhinia variegata were docked with RBP4. GC-MS analysis reveal 31 compounds from Garcinia cambogia while, Bauhinia variegata exhibit 9 compounds. In silico analysis showed that Tetradecanoic acid has interaction energy -9.08kcal/mol, followed by Hexadecanoic acid (-9.64Kcal/mol) and then Octadecanoic acid has -9.88Kcal/mol. Trans-geranylgeraniol showed -16.45Kcal/mol. But there was no interaction with protein and ligands of Bauhinia variegata like Hexadecanoic acid and Phytol were studied for insilico analysis. Phytol has -3.78Kcal/mol of interaction energy similarly hexadecanoic acid has -9.88Kcal/mol. But phytol did not show any hydrogen bond between the protein and ligand but Hexadecanoic acid shows hydrogen bond. While comparing these compounds Octadecanoic acid, hexadecanoic acid and tetradecanoic acid shows better interaction with RBP4 compared to Phytol and Trans-geranylgeraniol. Molecular docking analysis proved that secondary metabolites of Garcinia cambogia had better inhibitory activity against RBP4 than Bauhinia variegate and so these compounds from Garcinia cambogia may act as better drug models for obesity.

Millingtonia hortensis was examined for their antibacterial activity particularly against Mycobacterium leprae by Kumar et al, (2014). Initially, the solvents such as methanol, acetone, benzene and petroleum ether were utilized to extract the chemical components from Millingtonia hortensis. Subsequently, the components were identified by
means of GC-MS techniques. Moreover, molecular docking techniques were employed to determine the antibacterial activity of these compounds particularly against *Mycobacterium leprae*. Finally, Molinspiration and OSIRIS program were utilized to investigate the bioavailability and toxicity of the selected compounds. The results indicates that compounds such as Dl-alpha-tocopherol, Vitamin E, Squalene isolated from *Millingtonia hortensis* could be the potential molecule for the treatment of new as well as dapsone resistance cases of leprosy.

Gaddaguti et al, (2014) reported the compound identification from *Ocimum tenuiflorum* var. CIM-AYU leaves by Gas chromatography-Mass spectrometry (GC-MS) analysis and molecular docking studies of identified compounds along with known synthetic repellent DEET were made using Schrödinger maestro. Molecular docking results reveal that three compounds viz. Dl-alpha-Tocopherol, Gamma-Sitosterol and Lycopersin exhibit high binding affinity against Odorant binding receptor protein 3Q8I of *Anopheles gambiae*.

Selvamangai and Bhaskar, (2013) analyzed the phytochemical constituents of *Eupatorium triplinerve* using GC-MS and to study the ability of the metabolites to serve as an antagonist to caspase 3 receptors to ascertain its anticancer properties. Ten grams of the powdered sample was subjected to column chromatography over silica gel (100-200 mesh) and eluted with n-hexane, chloroform, ethanol and methanol respectively. n- Hexane and Chloroform did not elute much of the compounds. The methanol fraction of the *Eupatorium triplinerve* was taken for GCMS analysis. The analysis was carried out on a GC Clarus 500 GC system with a column packed with Elite – 1 (10% dimethyl poly siloxane, 30 x 0.25 mm ID x 1 EM df), the compounds are separated using with Helium as carrier gas at a constant flow 1ml/min. sample extract (2 μL) injected into the instrument was detected by Turbo gold mass detector (Perkin Elmer) with the aid of the Turbo mass 5.1 software. The important compounds obtained from GC-MS were further studied *in silico* to study its anticancer activity by docking with caspase 3 receptor of four important metabolites neophytadiene, nitrocyclohexane, octadecane and tetradecanoic acid. The GC
MS analysis provided peaks of eleven different phytochemical compounds namely hexadecanoic acid (14.65%), 2,6,10-trimethyl,14-ethylene-14-pentadecene (9.84%), Bicyclo [4.1.0] heptane, 7-butyl- (2.38%), Decanoic acid, 8-methyl-, methyl ester (3.86%), 1-undecanol (7.82%), 1-hexyl-1- nitrocyclohexane (2.09%), 1,14-tetradecanediol (6.78%), Octadecanoic acid, 2-hydroxy-1,3-propanediyl ester (19.18%) and 2- hydroxy-3-[(9E) -9-octadecenoyloxy] propyl(9E)-9-octadecenoate (8.79%). From the docking assay it was found that nitrocyclohexane and neophytadiene compounds exhibited good docking score. The bioactive compounds in the methanolic extract of *Eupatorium triplinerve* have been screened using this analysis. Isolation of individual components would however, help to find new drugs.

Gaddaguti et al, (2012) reported the plants constitute major source of drugs for prevention and spread of wide range of pathogenic carriers and also treating various diseases of human beings. Modern people increasingly prefer drugs of natural origin mostly from plant origin due to abundant accessibility and fewer side effects. Whereas synthetic drugs and antibiotics often cause wide spread toxicity and harmful side effects to the end user other than targeted health condition / pathogen carrier. In search of novel active compounds from plant origin, and to assess the efficient thereupatic properties with minimum side effects, application of advanced methods like GC MS and computational techniques play a crucial role in designing and development of drug of interest. 13 compounds were identified in aerial parts of *Hyptis suaveolens* L. methanolic extracts. Of the 13 compounds identified in the methonolic extract, Stigmast-5-en-3-ol, oleate, and Gamma-sitosterol and Butyl 11-eicosenoate found to represent 51.7% of the 13 compounds. Molecular docking studies were performed for all 13 compounds along with commercially known mosquito repellent compounds including DEET, Prallathrin, and Permithrin against Odorant Binding Protein (3N7H) of Anopheles *gambiae* using Schrodinger Maestro software. The binding affinities for compounds of *Hyptis suaveolens* were compared with known mosquito repellents for its ability to suppress human seeking
behavior of mosquitoes and further possibility for designing of potential mosquito repellent natural compounds were discussed.

Hariprasath, (2012) studied the molecular docking was carried out against two target proteins which are helpful for MRSA infection with the help of accelrys discovery studio to understand the applicability of the method to differentiate between the active and inactive compounds. Molecular docking of thirty two structurally diverse inhibitors were carried out and it was observed that though some false positives were also obtained; considering the limitations of the available docking programs, the results were promising. The high molecular weight compounds with heterocyclic rings showed very low binding energy, but did not comply with Lipinski’s rule. The active constituents that were docked with the protein are Baicalein, Biochanin, Carnosol, Genistein, Orobol, Resveratrol, Rhein, Gallic acid, Pyrithione, Resveratrol and Linozolid. The compound Orobol was found to interact more towards the target protein like showing highest Dock score.

### 2.5. Phytocompound isolation

Jameel et al, (2015) studied about the plant *Lens culinaris* Medik (Leguminosae). It is an annual, bushy and herbaceous plant cultivated globally for its edible seeds. A methanolic extract of the seeds contained four new antioxidant compounds, namely β-sitosteryl-3-(20-n-eicosanyloxy)-benzoate (3), n-octadec-9-enoyl-1-blucurano pyranoside (4) α-D-galactopyranosyl-(6′′-1′′′)α-D-galactopyranosyl-(6′-1′′′)-α-D-galactopyranosyl-(6′′-1′′′)-α-D-galactopyranoside (5) and benzoyl-O-α-D-glucopyranosyl-(2a-1b)-O-α-D-glucopyranosyl-(2b-1c)-O-α-D-glucopyranosyl-(6c-1d)-O-α-D-glucopyranosyl-(6d-1e)-O-α-D-gluco-pyranoside (6) along with two known compounds n-heptadecanyl n-octadec-9-enoate (1) and β-sitosterol (2) on the basis of chromatographic and spectral data analytical techniques. Compound 3 showed significant antioxidant activity compared to compounds 4, 5, and 6.

Ford et al, (2015) studied about the plant *Rubia tinctorum* L. It has been exploited as a dye throughout history. The roots of the plant are very rich in the highly coloured glycosidic compounds ruberythric acid and lucidin primeveroside, alongside the
corresponding aglycons which can be readily formed by deglycosylation, particularly during extraction. Supported by 1H and 13C NMR data, the conclusive X-ray crystal structure of the natural dye ruberythric acid is presented for the first time. The solid state structure revealed extensive intermolecular hydrogen bonding interactions between the sugar moieties in the unit cell, but only intramolecular hydrogen bonding through the hydroxyquinone groups. There is also some additional p–p stacking from the anthraquinone moiety.

Madikizela et al., (2014) studied the isolated compounds responsible for such activities and to isolate compounds responsible for antimicrobial activities from the crude extracts of *Terminalia phanerophlebia* leaves. *Terminalia phanerophlebia* crude extracts obtained from 80% methanol was successively extracted with hexane, dichloromethane (DCM), ethyl acetate (EtOAc) and n-butanol. The fractions obtained and isolated compounds were tested for their antibacterial activities against *Mycobacterium aurum*, *Mycobacterium tuberculosis*, *Staphylococcus aureus* and *Klebsiella pneumoniae*. Bio guided fractionation of the EtOAc fraction afforded two bioactive compounds. Structure elucidation was carried out using NMR (1Dand2D) spectroscopic methods. EtOAc fraction exhibited highest antimicrobial activities and its fractionation afforded methyl gallate (methyl-3,4,5-trihydroxybenzoate) (1) and a phenyl propanoid glucoside, 1,6-di-O-coumaroyl glucopyranoside (2). These compounds are reported from *Terminalia phanerophlebia* for the first time. Both compounds showed good antimicrobial activity against all bacterial strains tested with minimum inhibitory concentration (MIC) values ranging from 63 to 250 mg/mL. Inhibition of *Mycobacterium tuberculosis* by 1,6-di-O-coumaroyl glucopyranoside (2) at a MIC value of 63 mg/mL was noteworthy, as this bacterial strain is reported to be the leading cause of tuberculosis worldwide. Good antimicrobial activities exhibited by the compounds isolated from *Terminalia phanerophlebia* authenticate the traditional use of this plant in treating tuberculosis and its related symptoms. Compound (2), 1,6-di-O-coumaroyl glucopyranoside could serve as a lead compound for tuberculosis drug discovery.
Itharat et al., (2014) studied the rhizomes of *Dioscorea membranacea* Pierre (DM) which have been used as ingredients in anticancer herbal formulations in Thai Q2 traditional medicine (TTM). The active constituents of DM for cytotoxic activity in order to support its TTM use. A bioassay-guided isolation procedure was used to separate the cytotoxic constituents from ethanolic extract of *Dioscorea membranacea* rhizomes by testing against five human cancer cell lines, i.e. large cell lung carcinoma, COR-L23; liver cancer cells, HepG2; prostate cancer cells, PC3; breast cancer cells MCF 7; cervical cancer cells, Hela; and one normal human lung cell line (MRC5) using the SRB assay. Two known dihydrophenanthrene compounds [2,4 dimethoxy-5,6-dihydroxy-9,10-dihydrophenanthrene(1) and 5-hydroxy-2,4,6-trimethoxy-9,10-dihydrophenanthrene (2)], and a new dihydrophenanthrene compound, 5,6,2-tri hydroxy3,4-methoxy,9,10-dihydrophenanthrene(3) were isolated and fully characterized. 1 showed the highest cytotoxic activity against COR-L23, MCF-7 and PC3 cell lines (IC50=14.89, 17.49 and 19.04 mM, respectively), and 2 showed selective cytotoxic activity against PC3 (IC50=23.54 mM). The new compound 3 showed selective cytotoxic activity against only MCF-7 cells (IC50=31.41 mM). Interestingly the crude extract of DM was much less toxic to the normal cell line (MRC-5) (IC50=450 mg/ml) compared to the five cancer cell lines, (IC50 value ranged between 6 and 29 mg/ml). The phytochemicals isolated from DM may serve as lead compounds for the design of new anticancer agents with better selective cytotoxic indices.

Hooper et al., (2015) studied the plant genus *Desmodium*, especially *D. uncinatum*, are used on sub-Saharan small-holder farms as intercrops to inhibit parasitism of cereal crops by *Striga hermonthisca* and *Striga asiatica* via an allelopathic mechanism. The search for *Desmodium* species which are adapted to more arid conditions, and which show resilience to increased drought stress, previously identified *D. intortum*, *D. incanum* and *D. ramosissimum* as potential drought tolerant intercrops. Their potential as intercrops was assessed for resource poor areas of rain-fed cereal production where drought conditions can persist through normal meteorological activity, or where drought may have increasing
impact through climate change. The chemical composition of the root exudates was
categorized and the whole exudates biological activity was shown to be active in pot
experiments for inhibition of Striga parasitism on maize. Furthermore, rain fed plot
experiments showed the drought tolerant Desmodium intercrops to be effective for Striga
inhibition. This work demonstrates the allelopathic nature of the new drought tolerant
intercrops through activity of root exudates and the major compounds seen in the exudates
are characterized as being C-glycosylflavonoid. In young plants, the exudates show large
qualitative differences but as the plants mature, there is a high degree of convergence of
the C-glycosyl flavonoid exudates chemical profile amongst active Desmodium intercrops
that confers biological activity. This defines the material for examining the mechanism for
Striga inhibition.

Amin et al, (2016) studied the anti-leishmanial activity of plants collected from the
Egyptian flora, twenty extracts from fifteen plants growing in Egypt have been
investigated for in vitro leishmanicidal activity against Leishmania donovani
promastigotes. Among the tested extracts, the methanol extract of Euphorbia peplus aerial
parts exhibited a significant anti-leishmanial activity as it produced 100% inhibition of
growth with activity similar to amphotericin B. The total extract was subjected to liquid-
liquid fractionation using solvents of different polarities, followed by testing the
antileishmanial activity of the successive fractions. Phytochemical exploration of the
active n-hexane fraction (which produced 75% inhibition of growth) led to isolation of
four compounds: simiarenol (1), 1-hexacosanol (2), b-sitosterol (3), and b-sitosterol-3-O-
glucoside (4) from the biologically active sub-fractions. Structure elucidation was aided by
1D and 2D NMR techniques. In conclusion, E. peplus plant has many non-polar secondary
metabolites that can be used as drug leads for treatment of leishmaniasis.

Sanadhya and Durve, (2014) stated the isolated and characterized antimicrobial
compound from fruits of Anthocephalus indicus A. Rich. and Anthocephalus indicus A.
Rich. crude ethanol fruit extract and different solvent fractions of fruit extract were
screened for its antimicrobial activity against Gram negative and Gram positive bacterial
cultures by MIC and agar well diffusion method. TLC bioautography method was used for isolation of compound from fraction of fruit extract showing maximum antimicrobial activity. Screening of isolated compound was carried out by preliminary phytochemical test and characterization by FTIR, MS and 13. Crude ethanol fruit extract showed significant antimicrobial activity with MIC values of 12.5-6.25 mg/ml and zone of inhibition from 19.0-24.66 mm. amongst the different solvent fractions used aqueous fraction showed maximum antimicrobial activity with MIC values of 0.78-3.12 mg/ml and zone of inhibition from 26.33-35.0mm. TLC bioautography method helps in the isolation of antimicrobial compound from aqueous fraction. Characterization of isolated compound by FTIR, MS and NMR detects it as iridoid glucoside. The results revealed that the fruit extract possess significant antimicrobial activity due to the presence of iridoid glucoside as a bioactive compound which can be used as a source of safe herbal antimicrobial agent.

Mohammed and Bayati, (2009) studied the aerial parts of Thymus kotschyanus Boiss. and (Lamiaceae) and flower buds of Dianthus caryophyllus L. (Caryophyllaceae) have been traditionally implemented in the treatment of wounds, throat and gum infections and gastro-intestinal disorder by the indigenous people of northern Iraq, although the compounds responsible for the medicinal properties have not been identified. In this study, antibacterial compounds from both plants were isolated and characterized, and the biological activity of each compound was assessed individually and combined. Compounds were isolated and characterized from the extracted essential oils of both plants using different spectral techniques: TLC, FTIR spectra and HPLC. The minimum inhibitory concentrations MIC values for the compounds were assessed individually and combined based on a microdilution and the checker board method in 96 multi-well microtiter plates. Two known compounds were isolated from the essential oils of both plants and were identified as thymol and eugenol. The isolated compounds were investigated for their single and combined antibacterial activities against seven selected pathogenic bacteria; Staphylococcus aureus, Bacillus cereus, Listeria monocytogenes, Proteus mirabilis, Escherichia coli, Klebsiella pneumoniae and Pseudomonas aeruginosa.
Thymol MIC values ranged from 15.6 to 250.0 mg/ml and B. cereus was found to be the most sensitive pathogen with a MIC value of 15.6 mg/ml. Eugenol achieved stronger MIC values against most tested pathogens and the best MIC value (15.6 mg/ml) was observed against B. cereus, L. monocytogenes and K. pneumoniae whereas, S. aureus, P. mirabilis and E. coli were inhibited with a MIC value of 31.2 mg/ml. Combination results had antibacterial enhancement against most pathogens and the best synergistic result was seen against P. mirabilis and E. coli. The isolation of two antibacterial compounds from Thymus kotschyanus aerial parts and Dianthus caryophyllus flower buds validates the use of these species in the treatment of throat and gum infections, wound healing and gastro-intestinal disorder.

Huang et al, (2013) reported the anti-inflammatory activities of Chaenomeles speciosa (Sweet) Nakai (Rosaceae) to investigate the main components of 10% ethanol fraction of the crude extract of C. speciosa fruit in an attempt to find bioactive compounds or new compounds from this medicinal plant. The phytochemical investigation succeeded in isolating two new phenolic compounds, specpolyphenol A (1) and specphenoside A (2), together with three known phenyl glycosides (3–5) from the fraction. The structures of the new compounds were deduced from comprehensive spectroscopic analyses including IR, EI-MS, $^1$H NMR, $^{13}$C NMR, DEPT, COSY, HMBC and HMQC. The structures of the three known compounds 3, 4 and 5 were identified by comparison of their spectral data with those reported in the literature.

2.6. Antimicrobial activity of isolated compound

Pendota et al, (2017) reported Pappea capensis is a medicinal plant widely used in the management of eye infections, sexually transmitted infections and as an aphrodisiac in South Africa. Two flavonoids quercetin-3-O-rhamnoside (1) and epicatechin (2) were isolated through bio-assay guided fractionation. The extracts and isolated compounds were evaluated for antimicrobial, antagonococcal, antioxidant and cytotoxicity potential. The ethyl acetate fraction exhibited a broad spectrum of activity against Bacillus subtilis, Staphylococcus aureus, Escherichia coli and Candida albicans with MIC values ranging
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from 0.39 to 0.78 mg/mL. In the DPPH radical scavenging assay, extracts and isolated compounds showed free radical scavenging activity with EC50 values ranging from 6.34 to 158.00 μg/mL. The isolated compound epicatechin (2) exhibited higher antioxidant activity in the β-carotene-linoleic acid model system (EC50=9.64 μg/mL) than the ascorbic acid standard (EC50=13.37 μg/mL). Cytotoxicity of the samples was determined using the MTT (3-(4,5-dimethylthiazol)-2,5-diphenyl tetrazolium bromide) assay against Vero monkey kidney cells. Little to no cytotoxicity of the extracts and isolated compounds was observed. These results may help to discover new chemical classes of natural antimicrobial and antioxidant substances that could serve as selective agents for infectious microbial diseases.

Mabona et al, (2013) analyzed the antimicrobial properties of southern African medicinal plants against dermatologically relevant pathogens. The study also aimed at providing a scientific rationale for the traditional use of plant combinations to treat skin diseases and the isolation of the bioactive compound from the most active species, Aristea ecklonii (Iridaceae). Organic and aqueous extracts (132) were prepared from 47 plant species and screened for antimicrobial properties against dermatologically relevant pathogens using the microlitre plate dilution method. Four different plant combinations were investigated for interactive properties and the sum of the fractional inhibitory concentration (ΣFIC) calculated. Isobolograms were used to further investigate the antimicrobial interactive properties of Pentanisia prunelloides combined with Elephantorrhiza elephantina at varied ratios. A bioactivity-guided fractionation process was adopted to fractionate the organic leaf extract of Aristea ecklonii. Plants demonstrating notable broad-spectrum activities (MIC values ≤1.00mg/ml) against the tested pathogens included extracts from Aristea ecklonii, Chenopodium ambrosioides, Diospyros mespiliformis, Elephantorrhiza elephantina, Eucalyptus camaldulensis, Gunnera perpensa, Harpephyllum caffrum, Hypericum perforatum, Melianthus comosus, Terminalia sericea and Warburgia salutaris. The organic extract of Elephantorrhiza elephantina, a plant reportedly used to treat acne vulgaris, demonstrated
noteworthy antimicrobial activity (MIC value of 0.05 mg/ml) against *Propionibacterium acnes*. Similarly, *Diospyros mespiliformis* reported for its traditional use to treat ringworm, also displayed noteworthy antimicrobial activity against *Trichophyton mentagrophytes* (MIC 0.10mg/ml) and *Microsporum canis* (MIC 0.50mg/ml). The aqueous root extracts of *Pentanisia prunelloides* combined(1:1) with *Elephantorrhiza elephantina* displayed synergistic interactions (ΣFIC values 0.31–0.38) against *Staphylococcus aureus*, *Gentamycin methicillin* resistant *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Candida albicans*. Fractionation of *Aristea ecklonii* resulted in the isolation of the known bioactive compound, plumbagin, displaying noteworthy antimicrobial activity (MIC range between 2.00 μg/ml and 16.00 μg/ml). Most of the plant extracts demonstrated pathogen specific antimicrobial effects with a few exhibiting broad-spectrum activities. Positive antimicrobial effects noted for plants such as *Elephantorrhiza elephantina* and *Diospyros mespiliformis* used for acne vulgaris and ringworm infections, respectively, give some validation to their reported traditional uses. Synergistic interactions noted for *Pentanisia prunelloides* combined with *Elephantorrhiza elephantina* validate an enhanced antimicrobial effect when used in combination. Noteworthy antimicrobial activities (MIC range between 2.00 μg/ml and 16.00 μg/ml) were observed for plumbagin isolated from *Aristea ecklonii*.

Christiana et al, (2014) reported the isolated compounds from the petroleum spirit, chloroform and methanol crude leaf extracts of the white specie of the leaf of the *Sesamum indicum* used by traditional medicinal practitioners for the management of infectious diseases were investigated for *in vitro* antimicrobial activity against some organisms. The various extracts afforded compounds which had significant antimicrobial activities. The antimicrobial screening showed that all the pure isolates from the different solvent extracts were active against the organisms, *Staphylococcus aureus*, *Streptococcus pyogenes*, *Salmonella typhi*, *Candida albicans*, *Candida krusei*, and *Candida tropicalis* at various MIC’s and (MBC/MFC)’s. The pure isolates from methanol and ethyl acetate fractions were all inactive against *Bacillus cereus*, *Corynebacterium ulcerans*, and *Escherichia coli*. 
Only the pure isolates from petroleum spirit and chloroform fractions showed activity against *Bacillus cereus*, *Corynebacterium ulcerans*, and *Escherichia coli* at an MIC of 0.5mg/ml and MCB/MFC of 1mg/ml. Chromatographic techniques such as analytical TLC, Purification by dry vacuum liquid chromatography (DVLC) and Preparative TLC were used.

Verma et al, (2016) reported the chemical composition and antibacterial activity of the primary essential oil (PEO) and secondary essential oil (SEO) derived from steam distillation (SD) and hydro distillation (HD) of *Mentha citrata* Ehrh. Were investigated using gas chromatography-flame ionization detector (GC-FID), GC–mass spectrometry (GC–MS), disc diffusion assay and micro dilution broth assay. The oils isolated from herb and aqueous distillate using different methods showed substantial variations in their composition and antibacterial activity profile. Major constituents of the oils were linalool (33.9–77.1%), linalyl acetate(2.2–45.4%), α-terpineol (2.3–24.9%),α -caryophyllene (0.0–3.2%) and nerol (0.5–3.1%). The SEOs showed activity against all eight tested bacterial strains (MIC: 250–1000 µg/mL), while PEOs were active against seven strains (MIC: 250→1000µg/mL).

Niu et al, (2016) reported the two new compounds, (R)-4-(2-methylpentyl)-4H-dithieno[2,3-b:30,20-e]pyran (1) and 4-(2-ethylbutyl)- 4H-dithieno[2,3-b:30,20-e] pyran (2) were extracted from the seeds of *Voacanga africana*. The molecular structures of these compounds were measured with the help of broad spectroscopic (1D and 2D-NMR, IR, ESI-TOF-MS, HR-MS) analyses. The primary pharmacological operations of these compounds were brought to evaluation by applying the antibacterial extra somatic test. The results revealed that compound 1 and 2 were in an effective position to stop the growth of *Escherichia coli*, *Streptococcus aureus* and *Salmonella typhi*. On the other hand, the inhibitory impacts on *Staphylococcus aureus* as well as *Pseudomonas aeruginosa* were not apparent.

Chen et al, (2013) studied the identified antimicrobial components of *Araucaria cunninghamii* with an activity-guided purification process. Eight compounds were
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obtained from the most active n-BuOH fraction and identified as the new compound 4-n-butoxyl-phenylpropanetriol (1), together with seven known compounds (2–8). These compounds were tested for antimicrobial activities against five bacteria and four plant pathogenic fungi. Within the series of compounds tested, compound 2 was the most active, particularly displaying moderate antibacterial activities against *Erwinia carotovora* and *Bacillus subtilis* with MICs 7.8 and 15.5 mg/ml. Moreover, this compound exhibited inhibitory activities against four plant pathogenic fungi: *Helminthosporium sativum*, *Rhizoctonia solani*, *Fusarium oxysporum* f. sp. Niveum and *Fusarium oxysporum* f. sp. Cubense, with EC50 values of 42.3, 90.0, 62.7 and 100.2 mg/ml. To our knowledge, this is the first report that the n-BuOH fraction and compound 2 from *A. cunninghamii* showed inhibitory activity against plant pathogenic fungi.

Kirmizibekmez et al, (2015) reported the new prenylated isoflavan, iconisoflavan (1), and a newprenylated isoflav-3-ene, iconisoflaven (2) were isolated 23 from the roots of *Glycyrrhiza iconica* together with four known ones namely (3S)-licoricidin (3), licorisoflavan A 24 (4), topazolin (5) and glycycoumarin (6). The structures were elucidated on the basis of extensive spectroscopic analysis including 1D and 2D NMR as well as HR–MS. Furthermore, the absolute configurations of compounds 1, 3 26 and 4 were established by electronic circular dichroism (ECD). All the isolated compounds (1–6) were evaluated for their in vitro antimicrobial activities against five pathogenic bacteria and one yeast (Candida albicans) 28 using an in vitro microdilution method. Compounds 1 and 3–5 displayed significant activity against *Salmo-29 nella typhimurium* ATCC 13311 with MIC values ranging from 2 to 8μg/mL. Additionally, all compounds 30 were screened for their in vitro free radical scavenging activities using an in vitro microdilution DPPH 31 assay spectrofotometrically. The tested compounds exhibited IC50 values in the range of 0.18–0.56 mg/mL, suggesting an activity comparable with that of ascorbic acid (IC50: 0.07 mg/mL). To the best of our knowledge, 33 the present study constitutes the first phytochemical and bioactivity investigation on *G. iconica*. 
Aziz et al, (2012) studied the chemical constituents of the pseudo stems and rhizomes of Malaysian *Alpinia conchigera* and to evaluate the antimicrobial activity of the dichloromethane (DCM) extracts of the pseudostems, rhizomes and the isolated compounds against three selected fungi and five strains of *Staphylococcus aureus*. The dried and ground pseudo stems (0.8kg) and rhizomes (1.0kg) were successively extracted in Soxhlet extract or using n-hexane, dichloromethane (DCM) and methanol. The n-hexane and DCM extracts of the pseudo stem and rhizome were subjected to isolation and purification using column chromatography on silica gel using a stepwise gradient system (n-hexane to methanol). Briefly, a serial two fold dilutions of the test materials dissolved in DMSO were prepared prior to addition of 100 ml over night microbial suspension (108cfu/ml) followed by incubation at 37°C (bacteria) or 26°C (dermatophytes and candida) for 24 h. The highest concentration of DMSO remaining after dilution (5%,v/v) caused no inhibition to bacterial /candida/ dermatophytes’ growth. Antibiotic cycloheximide was used as reference for anti-candidal and anti-dermatophyte comparison while oxacilin was used as reference for antibacterial testing. DMSO served as negative control. Turbidity was taken as indication of growth, thus the lowest concentration which remains clear after macroscopic evaluation was taken as the minimum inhibitory concentration (MIC). The isolation of n-hexane and DCM extracts of the rhizomes and pseudostems of *Alpinia conchigera* via column chromatography yielded two triterpenes isolated as a mixture of stigmasterol and b-sitosterol: caryophylleneoxide, chavicolacetate1, p-hydroxycinnamaldehyde2,10S-10- acetoxychavicicolacetate3, trans-p-coumaryldiacetate4,10S-10-acetoxyeugenolacetate5,10-hydroxy- chavicol acetate6, p-hydroxycinnamylacetate7 and 4-hydroxybenzaldehyde. The DCM extract of the rhizome of *Alpinia conchigera* indicated potent antifungal activity against *Candida albicans*, *Microsporum canis* and *Trycophyton rubrum* with MIC values of 625 mg/ml, 156 mg/ml and 156 mg/ml, respectively. It also showed significant inhibitory activity with MIC values between 17.88 and 35.75 mg/ml against the mutant Staphylococci isolates MSSA, MRSA and Sa7. Amongst the isolated compounds, the lowest inhibition observed were of
10S-10-acetoxyeugenol against the dermatophytes (MIC313 mg/ml) followed by trans-p-coumaryldiacetate against both dermatophytes and candida (MIC625 mg/ml). The compound p-hydroxycinnamylacetate strongly inhibited *Staphylococcus aureus* strain VISA (MIC39 mg/ml) followed by trans-p-coumaryldiacetate and 10-hydroxychavicolacetate with MIC value of 156 mg/ml. In conclusion, the observed antibacterial, anticandidal and antidermatophyte activity of the extracts and compounds obtained from the rhizome confirm the traditional use of *Alpinia cochigera* rhizome in the treatment of skin infection.

Karunai Raj et al, (2012) reported the various extracts of the leaves and the isolated and identified Flindersine, a quinolone alkaloid as the major active principle. In the present study, they report the antibacterial and antifungal activities of Ulopterol, a coumarin isolated as another major active antimicrobial principle. The leaves were successively extracted with hexane, chloroform, ethyl acetate, methanol and water. The extracts were studied for their antimicrobial activity against selected bacteria and fungi by using disc-diffusion method. The ethyl acetate extract which was found to possess highest antimicrobial activity was subjected to activity guided fractionation by column chromatography over silica gel. This resulted in the isolation of the coumarin, Ulopetrol, an active principle besides Flindersine which was reported by us earlier. The structure of the compound was elucidated using physical and spectroscopic data. Flindersine and Ulopterol were quantified by HPLC. Ulopterol showed activity against the bacteria viz. *Staphylococcus epidermidis, Enterobacter aerogenes, Shigella flexneri, Klebsiella pneumoniae* (ESBL-3967), *Escherichia coli* (ESBL-3984) and fungi viz. *Aspergillus flavus, Candida krusei* and *Botrytis cinerea*. Quantification by HPLC showed the content of Flindersine and Ulopterol to be 0.361% and 0.266% respectively on dry weight basis of the leaves. Ethyl acetate extract (successive extraction) contained Ulopterol, a coumarin, besides Flindersine, a quinolone alkaloid, as a major active principle in the antimicrobial studies.
Viswanathan et al, (2012) studied the effect of different concentrations of the solvent extracts of *Jatropha tanjorensis* leaves and four isolated compounds were tested against human pathogenic microorganisms such as gram-positive bacteria of *Bacillus cereus*, *Bacillus subtilis*, *Staphylococcus aureus* and *Staphylococcus epidermis*, gram-negative bacteria of *Aeromonas hydrophila*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Proteus vulgaris*, *Salmonella paratyphi*, *Salmonella paratyphi* A, *Vibrio alcaligenes*, *Vibrio cholerae* and fungi of *Aspergillus fumigatus*, *Candida albicans*, *Microsporum gypseum* and *Trichophyton rubrum* by agar-well diffusion and disk diffusion methods. In agar-well diffusion method, maximum activity was recorded in a concentration dependent manner. The extracts recorded activity against bacteria such as 17–26, 15–25 and 13–24 mm to methanol extract and 15–24, 14–23 and 12–22 mm to chloroform extract at 50, 25 and 12.5 mg/ml respectively and fungi such as 9–15 mm to *A. fumigatus* and 5–16 mm to *T. rubrum*. Maximum activity was 30–46, 27–43 and 17–40 mm to friedelin and 23–46, 28–44 and 18–41 mm to R (+) 4-hydroxy-2-pyrrolidinone against bacteria and 12–37, 8–34 and 31–33 mm to friedelin and 12–40, 11–35 and 10–33 mm to R (+) 4-hydroxy-2-pyrrolidinone against fungi at 10, 5 and 2.5 mg respectively. Their finding concludes that friedelin, β-amyrin, stigmasterol and R (+) 4-hydroxy-2-pyrrolidinone present in the methanol extract could be responsible for the broad spectrum of antimicrobial activity and provide scientific evidence.