5. Discussion

Plants are important source of potentially useful structures for the development of new chemotherapeutic agents. The first step towards this goal is the *in vitro* antibacterial activity assay (Tona *et al.,* 1998). Many reports are available on the antiviral, antibacterial, antifungal, anthelmintic, antimolluscal and anti-inflammatory properties of plants (Samy and Ignacimuthu, 2000; Palombo and Semple, 2001; Kumaraswamy *et al.,* 2002; Stepanovic *et al.,* 2003; Bylka *et al.,* 2004; Behera and Misra, 2005; Govindarajan *et al.,* 2006). Some of these observations have helped in identifying the active principle responsible for such activities and in the developing drugs for the therapeutic use in human beings.

Many studies have been conducted to establish the antimicrobial effect of the medicinal plants (Habsah *et al*., 2000; Sudhakar *et al.,* 2006). The results of different studies provide evidence that some medicinal plants might indeed be potential sources of new antibacterial agents even against some antibiotic-resistant strains (Indu *et al*., 2006). Significant increase in the inhibitory zone was observed by increasing the concentration of the extract. Specifically, saponin has been reported to have antimicrobial effects (Mahato *et al*., 1992) and could serve as precursors of steroidal substances with a wide range of physiological activities (Madusolomuo *et al*., 1999).

According to Alam *et al.,* (2000), reported that the leaves of *Anisomeles indica* consists of diterpenoids, ovatodiolide and its derivatives that are used as HIV inhibitors.

The ethanomedicinal plant *Boswellia serrata*, 3-O-Acetyl-11-keto-beta-boswellic acid (AKBA) is the most active compound of *Boswellia* extract and is a potent inhibitor of 5-lipoxygenase (5-LOX), a key enzyme in the biosynthesis of leukotrienes from arachidonic acid in the cellular inflammatory cascade (Safayhi *et al.,*1992; Sailer *et al.,* 1996; Amar *et al.,* 2011).

Chlorophytum borivilianum has mannose and glucose which makes a mucilaginous layer around the urinogenital, gastrointestinal and respiratory tract when consumed orally. The layers trap the microbial flora and make them unable to invade the system (Sundaram *et al.,* 2011).

The ethanolic and petroleum ether flower extracts of *Euphorbia hirta* influences lipid parameters in alloxan induced diabetic mice (Kumar *et al.,* 2010). The preponderance of saponin in the extracts of *E. hirta* that reported by previous researchers (Johnson *et al*., 1999; Tona *et al*., 2004; Sudhakar *et al*., 2006) could then justify the use of these plants in the treatment of some microbial infections mentioned earlier. The results obtained indicated that the ethanolic extract of the *Euphorbia hirta* plant inhibited the growth of the test isolates except *Salmonella typhi.* This therefore shows that the extract contains substance(s) that can inhibit the growth of some microorganisms. Other workers have also shown that extracts of plants inhibit the growth of various microorganisms at different concentrations (Akujobi *et al.,* 2004; Esimone *et al.,* 1998; Nweze *et al*., 2004; Ntiejumokwu and Alemika, 1991; Osadebe and Ukwueze, 2004). The observed antibacterial effects on the isolates is believed to be due to the presence of alkaloids, tannins and flavonoids which have been shown to posses antibacterial properties (Cowan, 1999; Draughon, 2004). Some workers have also attributed their observed antimicrobial effects of plant extracts to the presence of these secondary metabolites (Nweze *et al.,* 2004). Some workers have also identified tannins, flavonoids and alkaloids in the extracts of the plant (Yoshida *et al.,* 1990; Blanc and Sacqui-Sannes, 1972; Abo, 1990; Baslas and Agarwal, 1980). The large zones of inhibition exhibited by the extract on *S. aureus* and *P. aeruginosa* justified their use by traditional medical practitioners in the treatment of sores, bores and open wounds. *S. aureus* and *P. aeruginosa* have been implicated in cases of boils, sores and wounds (Ogueke *et al.,* 2007). Also the moderate growth inhibition on *E. coli* justifies its use in the control of diarrhoea and dysentery. *E. coli* is the common cause of travelers diarrhoea and other diarrhoeagenic infections in humans (Ogueke *et al.,* 2007). The low MIC exhibited by the extract on *S. aureus* is of great significance in the health care delivery system, since it could be used as an alternative to orthodox antibiotics in the treatment of infections due to this microorganism, especially as they frequently develop resistance to known antibiotics (Singleton, 1999; Ogueke *et al.,* 2007). Their use also will reduce the cost of obtaining health care. The relatively high zone of inhibition exhibited by the extract on *E. coli* is also of significance, since *E. coli* is a common cause of diarrhea in developing countries (Ogueke *et al.,* 2007).

Evolvulus alsinoides contains alkaloids: betaine, shankhapushpine and evolvine. Fresh plant contains volatile oil. It also contains a yellow neutral fat, an

organic acid and saline substances (Goyal *et al.,* 2005; Singh, 2008). Universal secondary metabolites present in plants have been details by Rabe, (2000) and Omogbai and Eze, (2011) to be responsible for therapeutic activity. Singh and Bhat (2003); Omogbai and Eze, (2011) reported that flavonoids are dependable for the antibacterial activity which is related to various medicinal plants. The essential or volatile oils and their chemical components of plant have been used in bacterial infections for medicines purpose.

Moreover, hydrocarbon and terpene constituents of these oils having antibacterial properties. (Amit and Shailendra, 2006; Omogbai and Eze, 2011). Alkaloids and glycosides presence in *Evolvulus alsinoides* which is used in morden healthcare systems in the treatment of fever, cold, cough and veneral diseases. In morden research areas the ethanol extract of medicinal plants that having excellent antimicrobial activity compare to another solvent because the principles of antimicrobial activity were either polar or non polar components were more extracted throughout the medium of organic solvent. (Omogbai and Eze, 2011).

Microorganisms widely used in their susceptibility to antimicrobial agents. A high MIC value show low activity. The gram- negative organisms having lowest MICs and MBCs. The ethanol extract of *Evolvulus alsinoides* show higher susceptibility. Therefore, the ethanol extract of plant can be used to control of infections which caused by *Escherichia coli, Salmonella typhi, Staphylococcus aureus* and *Klebsiella pneumonia.* (Omogbai and Eze, 2011).

The *in vitro* study useful for antimicrobial substances against microorganisms. The Flavonoids substance response to the bacterial infection. The activity of substance to their ability to complex with bacterial cell walls, extracellular and soluble proteins. (Marjorie, 1999; Mohamed Sham Shihabudeen *et al.*, 2010). The *S. aureus* show highest sensitivity that response to its outer membrane and cell wall structure. (Mohamed Sham Shihabudeen *et al.*, 2010). Saponin substance having antimicrobial properties due to ability to the cause leakage of enzyme and proteins from the cells. (Zablotowicz *et al.,* 1996; Mohamed Sham Shihabudeen *et al.*, 2010).

Medicinal plants are used by large proportion of Indian population. The reasons for this include true improvement, absence of harmful side effects and the

high cost of other forms of treatment. Urinary Tract Infections (UTIs) are a leading cause of morbidity and involve high health care expenditure in persons of all ages. Sexually active young women are disproportionately affected, but several other populations including elderly persons and those undergoing genito-urinary instrumentation or catheterization, are also at risk. An estimated 40 percent of women reports having had a UTI at some point in their lives. UTIs are the leading cause of gram-negative bacteria. Several plants have been used in traditional medicines to combat UTI. From the earlier studies it is obvious that most of the plants differ significantly in their antimicrobial property. These differences may be attributed to the differences in the cell wall constituents of bacteria which vary among the gram positive and gram negative ones (Yao and Moellering, 1995; Balasundaram *et al.,* 2011).

In general, alcoholic extracts exhibit highest degree of antimicrobial activity as compared to aqueous extract fractions. The inhibitory activity was found to be maximum in the methanolic extract**s** of Euphorbiaceae plants against urinary tract pathogens such as *E. coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* (Ramesh *et al*., 2001; Balasundaram *et al.,* 2011).

The study indicated that, *P. aeruginosa* was more susceptible to the ethanol extract followed by *S. aureus.* This is in contrary with the earlier reports, which have shown that, most antibacterial medicinal plants attack gram positive strains than gram negative strains because of their permeability differences (Priscilla *et al.,* 2007; Pavithra *et al.,* 2010; Ramya and Devi, 2011). The possible mechanism for their broad spectrum activity against both gram positive and gram negative bacteria may due to their ability to complex with cell wall (Cowan, 1999; Ramya and Devi, 2011).

The maximum zone of inhibition was observed in the methanolic extract of *Thevetia peruviana* against *Klebsiella*, *Proteus* & *E.coli*, *MRSA* and *Pseudomonas* respectively work done by Jaryal *et al.,* 2012.

According to Pathak *et al.,* (2010), the methanolic extract of the leaves was effective against *Staphylococcus aureus, Proteus vulgaris, Klebsiella pneumonia* & *Bacillus subtilis.* The Comparative antibacterial activity between Methanolic extract of *Annona muricata* and standard antibiotic Streptomycin was studied. The

Methanolic extract showed significant antibacterial efficacy as compared to the standard antibiotic, Streptomycin. The beneficial effects of treatment can be achieved with the treatment with the leaves of *Annnona muricata* in various bacterial infectious diseases like Pneumonia, Diarrohea, Urinary tract infection, & even some skin disease.

According to Panghal *et al.,* 2011, the bacteria contain outer membrane made up of lipopolysaccharide that permeability give the naturally resistance to antibiotics. The biofilm of colonize surfaces the cells resistance to the concentration sof antibiotics. *P. aeruginosa* resistance to antibiotics and particularly hazardous and anxiety pathogen. (Iinuma *et al.,* 1994; Kim *et al.,* 1995).

Virtual screening has developed to an essential part of modern drug research. A multiplicity of computational tools is developed and to spend for speedy screening methods to protein hits yielding. The last few years successful application methods utilize pharmacophore based search, machine learning tools and ranging methods, etc. modern drug discovery pharmacodynamic, pharmacokinetic and toxicity aspects consider in the virtual screening approaches. (Daisy *et al.,* 2011).

According to Johnson and Wolfgang, (2000), the chemical compounds have assured property to be traditional as drug. It is a rule of thumb to evaluate drug likeness, or to determine if a chemical compound with a certain pharmacological or biological activity has properties that would make it a likely active drug. The Lipinski's Rule of Five requirements with no more than one violation. All the compounds were found to have no more than one violation in terms of hydrogen bond donors (<5 hydrogen bond donor) and hydrogen bond accepter (<10 hydrogen bond acceptor), Molecular weight was in the range of 160 to 500, Partition coefficient log *P* in -0.4 to +5.6 ranges, Molar refractivity from 40 to 130, Topological Polar Surface Area (TPSA) no greater than 140 $Å^2$ (Ghose *et al.,* 1999; Daisy *et al.,* 2011)

Using a simplified, yet efficient version of the QSAR paradigm for structure-permeability (Van de Waterbeemd *et al.,* 1996; Daisy *et al.,* 2011) suggested that poor absorption or permeation is more likely to occur when the molecular weight (MW) is over 500, the calculated (Leo, 1993; Daisy *et al.,* 2011) octanol/water partition coefficient (CLOGP) is over 5, there are more than 5 H-bond (hydrogen bond) donors

(HDO-expressed as the sum of O-H and N-H groups) and there are more than 10 H-bond acceptors (HAC-expressed as the sum of N and O atoms). Lipinski *et al.,* 2001 (Doddareddy *et al.,* 2006; Daisy *et al.,* 2011) suggested that any pair wise combination of the following conditions: MW > 500, LOGP > 5, HDO > 5, and HAC > 10, may result in compounds with poor permeability (exceptions are actively transported compounds and peptides) (Daisy *et al.,* 2011).

According to Egan and Lauri, 2002, ADMET predicts the Human Intestinal Absorption (HIA) after oral administration. Intestinal absorption is defined as a percentage absorbed rather than as a ratio of concentrations (cf. blood-brain penetration). A well-absorbed compound is one that is absorbed at least 90% into the bloodstream in humans. Cheng and Merz, 2003, suggested that, ADMET describes the aqueous solubility (Aq.Sol.Lev) using linear regression to predict the solubility of each compounds in water at 25˚C. ADMET-Blood brain barrier is a model that predicts blood-brain penetration (blood-brain barrier, BBB) of the compounds after oral administration. ADMET-The cytochrome P450 2D6 model predicts CYP2D6 enzyme inhibition using 2D chemical structure as input. Dixon and Villar, 1999 developed the ADMET hepatotoxicity model which predicts potential organ toxicity for a wide range of structurally diverse compounds. Dixon and Merz, 2001 developed the ADMET-plasma protein binding model that prediction of a chemical compound expected to extremely bind to the proteins in the blood. (Daisy *et al.,* 2011).