# CHAPTER 1

# LITERATURE SURVEY

## 1.1. The problem of antibiotic resistance

Antibiotics are substances that suppress the growth of other microorganisms eventually killing them (Goodman and Gilman, 1998). However, common usage often extends the term "antibiotics" to include products derived from plants as well as synthetic antibacterial agents, such as the sulfonamides and quinolones, which are not products of microbes. Antibiotics differ in chemical, physical, and pharmacological properties, antibacterial spectra, and mechanisms of action.

The development of antimicrobial drugs represents one of the most important advances in therapeutics, both in the cure and control of serious infections and in the prevention and treatment of infectious complications arising from other forms of medical therapy such as cancer chemotherapy and surgery, (Katzung, 1998). The discovery of the antimicrobial activity of penicillin turned the attention of investigators to antibiotics as potentially useful chemotherapeutic compounds. In the 1940's and 1950's streptomycin, chloramphenicol, tetracyclines, polymyxin, bacitracin and neomycin greatly increased the range of effectiveness of antibacterial chemotherapy (Clark *et al.*, 1992). However, there is increasing evidence that antimicrobial agents are vastly over-prescribed in many outpatient settings globally (Katzung, 1998). This as well as the availability of antimicrobial agents without a prescription in many developing countries has facilitated the development of resistance, severely limiting the therapeutic options in the treatment of life-threatening infections.
In a small study at Pretoria Academic Hospital, it was established that 60% of patients were prescribed antimicrobials as part of their treatment regimen (Serage *et al.* 1999, unpublished). In addition, as the population increases, more funds are spent on antimicrobial therapy alone. Worldwide spending on finding new anti-infective agents is expected to increase by 60% from the spending levels in 1993. The public is becoming increasingly aware of problems with the over-prescribing and traditional misuse of antibiotics (Cowan, 1999).

The cornerstone of treatment for an infected patient is the isolation and identification of the microorganism involved. The susceptibility of the microorganism(s) to the antimicrobial agents is then determined. However, because of time lag involved in this process, often about 48 hours, the initial treatment must frequently be based entirely on clinical impression. This is deduced from the history of the patient, physical examination, symptomology, microscopic examination, epidemiology and, if possible, rapid laboratory tests. The antimicrobial therapy is started with agents that cover the suspected microorganism (Wiener and Pepper, 1985). It is also therapeutically recommended to treat with the single agent that is most specific for the infecting organism. This strategy reduces the emergence of resistance and minimizes toxicity (Mycek, Harvey and Champe, 2000).

The rational use of antimicrobials must involve a consideration of several aspects. Antimicrobial agents should be indicated based on thorough clinical findings. Clinical specimens should be obtained in order to establish the sensitivity of the suspected pathogens to the available antimicrobial regimen. The etiologic factors for the patient's illness should be established. Lastly, the individuals exposed to the index case should be protected in order to prevent secondary or nosocomial infections. Unfortunately antimicrobial agents are usually used before the causative pathogen for a particular illness or the susceptibility to a particular antimicrobial agent is known. This use of antimicrobial therapy is called empirical (presumptive) therapy, and is based upon the experience with a particular clinical entity (Katzung, 1998). The principal justification for empirical therapy is that infections are best treated early. To withhold therapy until the results on culture and susceptibility tests are available may expose the patient to risks of serious morbidity or death. Initiation of empirical therapy to a certain extend should however conform to a well-defined protocol (Barrie and Jacobs, 1996).

Several schemes have been proposed to classify and group antimicrobial drugs by their mode of action, (Katzung, 1998). Some of the proposed mechanisms of action are as follows:

- Agents that inhibit the synthesis of bacterial cell walls, e.g. the penicillins and cephalosporins.

- Agents that act directly on the cell membrane of the microorganism, affecting permeability and thus leading to leakage of intracellular constituents; e.g. detergents, polymyxin, and the polyne antifungal agents, nystatin and amphoteracin B, that binds to cell wall sterols.
- Agents that affect the function of the 30s or the 50s ribosomal subunits to cause a reversible inhibition of protein synthesis, this includes chloramphenicol and the tetracyclines.
- Agents that affect the nucleic acid metabolism and thus inhibiting DNA-dependent RNA polymerase, such as rifampicin.
- Antimetabolites, which block specific steps that are essential to the growth of microorganisms, eg. trimethoprim and sulfonamides. They may also be nucleic acid analogues, which inhibit viral enzymes that are essential for DNA synthesis thus hindering viral replication. e.g. zidovudine, acyclovir and vidarabine.

Microorganisms can adapt effectively to environmental pressures in a variety of ways. Their response to antibiotics is no exception. It is reported that penicillins are by far the most widely used antibiotics and therefore account for some of the most drastic consequences of antibiotic misuse (Jawetz 1996). Although pneumococci have long been considered an example of the total and regular susceptibility to penicillins, this is no longer entirely true. In Papua New Guinea and South Africa, outbreaks of meningitis and pneumonia due to penicillin-resistant pneumococci have been observed (Jawetz, 1996). The increase in resistant strains to current regimens of antimicrobials has led to increased efforts in the search of new antimicrobial agents.

An inevitable consequence of antimicrobial usage is the selection of resistant microorganisms. Thus, an important part of the pharmacology of antibiotics is the study of mechanisms of resistance to them. Some of the ways through which resistance is brought about are as follows:

- The increased production of enzymes by the microorganisms. The β-lactam ring of the penicillins and cephalosporins stimulate production of the enzyme β-lactamase, which inactivate the antimicrobials,

- Changing of the target site, e.g. penicillin binding site by a single mutation,
- Antibiotic exclusion, resulting from the inability of the drug to penetrate the outer membrane of the organisms.
- The cell wall of Gram-negative organisms is more difficult to penetrate than those of the Gram-positive, so in general the former present greater problems with resistance.

### 1.2. Can plant constituents play a role in resolving the problem of antibacterial resistance?

Man has largely depended on plants as the main source of medicine for thousands of years (Husain, 1991). It has been estimated that approximately 25 % of modern drugs are originally derived from higher plants. The use of plant extracts (botanicals or phytomedicines) and other forms of alternative medicinal treatments has gained popularity in the late 1990's. In 1996 alone sales of phytomedicines for antimicrobial usage increased by 37% over sales in 1995 (Kinghorn and Balandrin, 1993).

It was only since the Second World War that scientists embarked on the synthesis of medicine at a pharmaceutical level. The use of herbal medicines has however remained the mainstay of health-care in most of the world (Husain, 1991). Noristan Pharmaceutical company was involved in the screening of South African medicinal plants. Many of the plants investigated did exhibit a potential antimicrobial activity, (Fourie *et al.* 1992). Plants can be used either as dried or as fresh material depending on the type of cure they are intended for. The leaves, roots and bark are the most useful parts of the plants because of their accessibility.

Early man developed the doctrine of signatures; this states that God created plants so that the natural appearance of plant parts signifies what they can cure. For example, plants with heart-shaped leaves would cure heart-related disorders while kidney-shaped leaves will be useful for kidney malfunctions. As expected, scientific evidence could not confirm this general theory although in some cases there was efficacy, e.g. digitalis for cardiac failure (Huxley, 1984). Some plants are used to cure multiple diseases, effective against many ills e.g. *Ephedra* species. When the twigs are made into tea they allay fever and coughs, and may increase blood pressure. Reserpine derived from *Rauwolfia serpentina* lowers blood pressure in hypotensive patients, controls arrhythmia and calms the central nervous system.

Herbal medicines may have little or no pharmacological effect and could be mere placebos that might do little good but do not cause any harm, in contrast to the pharmaceuticals (Tyler and Robbers, 2000). However, one major threat to the use of plants in herbal industry is if, the natural vegetation fails to cope with the ever-increasing demand. A good portion of currently used medicines is derived either directly or indirectly, from active principles that have been isolated from plants [Table 1.1]. Most of these substances do not occur in plants individually but in groups of compounds, such as caffeine in the group of methylxanthines, digoxin in the group of cardiac glycosides, and morphine in the group of opium alkaloids.

Table 1.1 Active principles presently used for medicinal purposes and the plants from which they are derived (Husain, 1991 and Cowan, 1999).

| **Plant constituent** | **Plant raw material** | **Pharmacological activity** |
|---|---|---|
| Cocaine | *Erythrophyllum coca* | Local anaesthetic, Analgesic |
| Digoxin lanatosides | *Digitalis lanata* | Cardiotonic |
| Emetine | *Cephaelis ipecacuanha* | Emetic |
| Ephedrine, Pseudoephedrine | *Ephedra species* | Antihypertensive, Sympathomimetics |
| Ergotamine | *Claviceps purpurea* | Migraine |
| Quinidine | *Cinchona species bark* | Antiarrythmic |
| Quinine | *Cinchona species bark* | Antimalarial |
| Reserpine | *Rauwolfia serpentina* | Antihypertensive |
| Tannin | *Eucalyptus globulus* | Antibacterial, antiviral |
| Totarol | *Podocarpus nagi* | *Pseudomonus acne* |
| Curcumin | *Curcuma longa* | Antiprotozoal, antibacterial |
| Thymol | *Thymus vulgaris* | Antibacterial, antiviral, antifungal |
| β-Resercyclic acid | *Cannabis sativa* | Antibacterial, antiviral |
| Catechin | *Camellia sinesis* | *Shigella, Vibrio, Streptococcus mutans* |
| Combretastatins | *Combretum caffrum* | Anti-angionesis |

Plants have been used for centuries to treat infections and other illnesses in humans. In some cases traditional healers working together with scientists were keeping records on the safety and effectiveness of phytochemical treatments, but these were generally uncontrolled and unrandomised studies. However, a few reviewed trials of the use of phytochemicals are briefly reported e.g. mouth-rinses containing antimicrobials were evaluated in humans and compared with listerine or chlorhexidine. The investigation of plant extracts effective against methicillin-resistant *S. aureus* provides an example of prospects in the search for new compounds which may be effective against infections that are currently difficult to treat (Cowan, 1999). In the next chapter the possible use of members of Combretaceae as the source of antibacterial compounds will be discussed.