GENERAL INTRODUCTION

Regarding inventions, Indians had been the forefathers in almost all the fields. By casting an eye on the manuscripts written on palm leaves, we can understand that the priority that they given was medicines. By strictly following their own inventions they had cherished their lives on this beautiful earth more than hundred years.

Autoimmune diseases are defined as a group of disorders in which tissue injury is caused by humoral or cell mediated immune response to self antigens. Rheumatoid arthritis is a health condition that causes pain, swelling, stiffness and loss of function in the joints (Waller, 1983; Goldenberg, 1999; Bertolini et al., 2001 and White et al., 2007). Conventional treatments are available for rheumatoid arthritis. There are about 100 types of arthritis but most common are osteoarthritis and Rheumatoid arthritis. In osteoarthritis, the common sings and symptoms are stillness in a joint after getting out of led or sitting for a long time, swelling in joints, crunching feeling or the sound of bone rubbing on bone (Walker et al., 2006). Rheumatoid arthritis is a long lasting disease that can affect joints in any part of the body most commonly the affects the wrists of hands and knee of legs. The immune system mistakenly attacks itself and causes the joint lining to swell by targeting the thin membrane (synovium) that lines the joints. The inflammation then spreads to the surrounding tissues and can eventually damage cartilage and bone (Hand out on health 2004). Catabolic reactive protein (CRP) was originally discovered by Tillett and Francis in 1930 as a substance in the serum of patients with acute inflammation that reacted with the polysaccharide of Pneumococcus (Tillett and Francis, 1930). Initially it was thought that CRP might be a pathogenic secretion as it was elevated in people with a variety of illnesses including cancer (Pepys and Hirschfield, 2003). However, discovery of hepatic
synthesis demonstrated that it is a native protein. CRP is a member of the class of acute – phase reactants, as its levels rise dramatically during inflammatory processes occurring in the body. This increment is due to a rise in the plasma concentration of IL – 6, which is produced predominantly by macrophages (Pepys and Hirschfield, 2003) as well as adipocytes, (Lau et al., 2005). CRP is used mainly as a marker of inflammation.

Inflammation is the complex biological response of vascular tissues to harmful stimuli including pathogens, irritants or damaged cells. It is a protective attempt by the organism to remove the injurious stimuli as well as initiate the healing process for the tissue (Denko, 1992) The process of inflammation is necessary in healing of wounds, inflammation however, if runs unchecked, lead to onset of disease like vasomotor rhinnorhoea, rheumatoid arthritis and atherosclerosis (Henson and Murphy, 1989).

The modern system of medicine had always been enthusiastic to evoke non-specific defence mechanisms of human physiology, which led to the discovery of active immunization using microbial preparations to enhance the host defense against infection. Recently, the same enthusiasm has taken an important leap towards exploring a novel group of substances from natural resources that modulate the immune response of living systems (Gutali et al., 2002). The immune system is involved in the etiology as well as pathophysio logic mechanisms of many diseases. Modulation of the immune responses to alleviate the diseases has been of interest for many years (Sharma 1983).

Medicinal plants are a rich source of substances which are claimed to induce paraimmunity, the non-specific immunomodulation of essentially granulocytes, macrophages, natural killer cells and complement functions (Sainis et al., 1997). In India,
knowledge and wisdom have been passed on from one generation to the next through songs and poems, which scholars and physicians have to learn and recite by heart. The Veda is an ancient text in four parts (Rig Veda, Sama Veda, Yajur Veda and Atharva Veda), the earliest of which date back to 2000 years BC. The principles of Ayurvedic medicine and the medicinal uses of plants are contained in thousands of poetic hymns in the Rig Veda. The first school to teach Ayurvedic medicine is at the University of Banaras in 500 BC where the great Samhita (encyclopedia of medicine) is written. Another great encyclopedia is written 700 years later, and these two together form the basis of the Ayurveda (Chopra, 2000).

A number of traditions come to dominate the practice of herbal medicine in the western world at the end of the twentieth century. The western based on Greek and Roman sources also called Arabic Medicine (WHO, 2002). A large number of traditional herbal remedies in Europe have become widely known as a result of commercialization and a number of active compounds are isolated from medicinal plants and are used today as single chemical entities (Pieroni, 2000).

Throughout the ages, human beings have relied on nature for their basic needs, for the production of food, shelter, clothing, transportation, fertilizers, flavours and fragrances, and medicines (Cragg and Newman, 2005). Plants have formed the basis of sophisticated traditional medicine systems that have been in existence for thousands of years and continue to provide mankind with new remedies. Although some of the therapeutic properties attributed to plants are erroneous, medicinal plant therapy is based on the empirical findings of hundreds and probably thousands of years of use. The first records, written on clay tablets in cuneiform, are from Mesopotamia and date from about 2600 BC (Heinrich et al., 2004).
There are many factors that lead to the process of increasing the use of medical plants in treating different diseases. First of all the presence of new disease that some of which have not been got rid of till now and secondly the belief of the societies, members that the natural substances have good effects. Finally the appearance of ecological movements all over the world that call for paying attention to the medical use of plants since people believed strongly that plant medicine is safer and more successful than the manufactured synthetic drugs (Shulz et al., 1998 and Tyler, 1999).

A vast majority of people on this planet still rely on their traditional "materia medica" (medicinal plants and other materials) for their everyday health care needs. It is also a fact that one quarter of all medical prescriptions are formulations based on substances derived from plants or plant-derived synthetic analogs. According to WHO (2000), 80% of the world's population, primarily those of developing countries rely on plant-derived medicines for their healthcare. It is likely that the profound knowledge of herbal remedies in traditional cultures developed through trial and error over many centuries, and that the most important cures were carefully passed on verbally from one generation to another. The hidden resources of ethnobotany are brought to light by many workers to get a cure without side effects.

A number of undesired side effects sometimes occur during chemotherapy in treating diseases. Natural therapies, such as the use of plant-derived products, may reduce adverse side effects. (WHO, 2005). Because of the side effects caused by conventional drugs, and the resistance that pathogenic micro organisms build against antibiotic, the use of natural products as an alternative to conventional treatment in healing and treatment of
various diseases has been on the rise in the last few decades (Fong, 2002, Chung et al., 2004 and Essawi et al., 2005).

India has 2.4% of world's area with 8% of global biodiversity. It is one of the 12 mega-diversity hot-spot regions of the world, other countries being Brazil, Colombia, China, South Africa, Mexico, Venezuela, Indonesia, Ecuador, Peru, USA and Bolivia. Across the country, the forests of India are estimated to harbour 90% of India's medicinal plants diversity in the wide range of forest types that occur (Pushpangadan and Atal, 1984). Only about 10% of the known medicinal plants of India are restricted to non-forest habitats. The estimated numbers of plant species and those used for medicinal purpose vary. One fifth of all the plants found in India are used for medicinal purpose. The world average stands at 12.5%, while India has 20% plant species of medicinal value and which are used to treat various diseases (Balick and Cox, 1997). Although it is difficult to estimate the number of medicinal and aromatic plants present worldwide, the fact remains true that India with rich biodiversity ranks first in medicinal flora, containing myriads of active medicinal ingredient.

Medicinal plants serve as therapeutic alternatives, safe choices, or in some cases, as the only effective treatment. A large number of plants and their isolated constituents have shown beneficial therapeutic effects, including anti-oxidant, anti-inflammatory, anticancer, antimicrobial and anti immunomodulatory effects (Thatte et al., 1992, Salem and Houssain, 2000, Huffman, 2003 and Miller et al., 2004). Some of the plants with established immunomodulatory activity are Viscum album, Panax ginseng, Asparagus
Though several works on the phytochemistry, separation of active components, antimicrobial efficacy and immune enhancement of various medicinal plants and their use in therapeutical applications have been carried out, no detailed study was made in this line by selecting tender fruit of *A. esculentus*. Considering this fact the present investigation is aimed to probe the therapeutic value of *A. esculentus* for sustainable disease management.

The study comprises,

- Chapter I provide the basic information about phytochemistry and identification of active principles present in tender fruits of *A. esculentus*.
- Chapter II describes the anti bacterial effect of tender fruits of *A. esculentus* against human bacterial pathogenic organisms. It helps to develop plant based pharmaceutical products.
- Chapter III focuses on animal, Swiss albino mice, the immune modulatory effect of tender fruits of *A. esculentus* and it deals about the immunology such as B and T cell count, antibody titration, hematological parameters and Delayed type of Hypersensitivity (DTH).
- Chapter IV provides the therapeutical assay of chosen plant against the rheumatoid antigen (RA) and catabolic reactive proteins (CRP).
- Chapter V explains microbiological aspects in view to find out the effect of extracts of *A. esculentus* on the health status mice.
- Chapter VI focuses about the extent of damage in histology of tissues, namely liver, spleen and muscle due to rheumatoid antigen and to test the repairing efficiency of tender fruit of *A. esculentus* in the experimental animal.
Scope of the Study

Arthritis and rheumatism are well defined diseases of the musculoskeletal system. Before the availability of synthetic drugs, man was completely dependant on medical plants for curing diseases. With synthesis of aspirin in 19th century, a new era started in the history of anti-inflammatory and analgesic drugs (Vane and Bolting 1971). It is well known that several physiological changes play key role in initiating information in disorders like arthritis and rheumatism. These responses to inflammation include hyperpyrexia, increased leucocyte count and differential leucocyte count. This disease has a worldwide distribution but its pathogenesis is not clearly understood (Harris et al 1990). Natural products have long been recognized as an important source of therapeutically effective medicine (Cragg and Newman, 2003).

The vast majority of people on this planet still relay on their traditional material medica for their everyday health care needs. One quarter of all medical preparations are formulations based on substance derived from plants or plant derived synthetic analogs (Kannan et al., 2007) A large number of plants and their isolated constituents have shown beneficial therapeutic effects, including anti-oxidant, anti-inflammatory, anticancer, antimicrobial and anti immunomodulatory effects (Huffman, 2003 and Miller et al., 2004). The ethanol extract Justica gendarussa leaf showed significant antiarthritic activity (Paval et al., 2009).

A.esculentus showed positive influence against genito – urinary disorders, spermatorrhoea and chronic dysentery (Nandkarni 1927) and ulcers and relief from hemorrhoids (Adams, 1975). However, there is no detailed information on the phyto constituents of the plant A.esculentus and its effect against export rheumatoid antigen
(RA), in an animal model swiss albino mice. The results accrued through the plant
A. esculentus may pave way for the formulation of new drug, relating to RA, one of the
stressing problem facing by the people all over the country.
Objectives of this study

• To screen the phytochemicals present in tender fruit of A. esculentus.

• Isolation and identification of biologically active compound(s).

• *In vitro* testing of various extracts of A. esculentus against human bacterial pathogens.

• To evaluate immunomodulatory efficiency of A. esculentus in the test animal, Swiss albino mice.

• To screen the therapeutic value of various extracts of A. esculentus against rheumatoid antigen and production of catabolic reactive protein.

• To analyse the influence of various extracts of A. esculentus on the gut microbial population of Swiss albino mice.

• To find the impact of ethanol extract of A. esculentus on the recovery of rheumatoid Swiss albino mice by histological observation.
Description of the test plant

Plant bhendi *A. esculentus* L. (SPHB-9) – Malvaceae (Plate-1) was chosen for present investigation. It is being used for a long time as an edible vegetable in many countries because of its nourishment component. Traditionally it is believed that the plant is useful in the treatment of inflammatory disorders, conceptions, retention of urine etc (Nandkarni, 1927).

It is grown commercially in India, Turkey, Iran, Western Africa, Yugoslavia, Bangladesh, Afghanistan, Pakistan, Burma, Japan, Malaysia, Brazil, Ghana, Ethiopian, Cyprus and the southern United States. India ranks first in the world with a production of 3.5 million tones (70% of the total world production) of Okra (FAOSTAT 2008). In some regions leaves are also used for human consumption. This vegetable provides an important input of vitamins and mineral salts including calcium, which are often lacking in diet of developing countries (IBPGR, 1990).

Classification of test plant

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<tr>
<td>Kingdom</td>
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<td>Division</td>
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<td>Order</td>
<td>Malvales</td>
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<tr>
<td>Family</td>
<td>Malvaceae</td>
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<tr>
<td>Genus</td>
<td><em>Abelmoschus</em></td>
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<tr>
<td>Species</td>
<td><em>esculentus</em></td>
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Other names of *A. esculentus*

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<th>Language</th>
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<tbody>
<tr>
<td>Tamil</td>
<td>Vendi</td>
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<tr>
<td>American English</td>
<td>Okra</td>
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<tr>
<td>British English</td>
<td>Lady’s finger</td>
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<tr>
<td>Hindustan</td>
<td>Bhendi and Gumbo.</td>
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The Species *A. esculentus* (Plate 1.1) is an annual growing to 2 m tall. The leaves are 10-20 cm long and broad, palmately loved with 5-7 lobes. The flowers (Plate 1.2) are 4-8cm diameters with five white to yellow petals, after with a red or purple spot at the base of each petal. The fruit (Plate 1.3) is a capsule up to 18cm long, containing numerous seeds (Martin., 1978).

**Traditional and other uses**

*A. esculentus* helps to lubricate the large intestines due to its bulk laxative qualities. The *A. esculentus* fibre absorbs water and ensures bulk in stools. This helps to prevent constipation. *A. esculentus* is non – toxic, has no adverse side effects, is full of nutrients, and is economically within reach of most. Its mucilage is a suspending agent and tablet binder (Deveswaran *et al.*, 2011).

The roots and stems of Okra are used for cleaning the cane juice from which gur or brown sugar is prepared (Chauhan, 1972) It’s ripe seeds are roasted, ground and used as a substitute for coffee in some countries. Mature fruits and stems containing crude fiber are used in the paper industry. Extracts from the seeds of the Okra is viewed as alternative source for edible oil. The greenish yellow oil has a pleasant taste and odor, and is high in unsaturated fats such as Oleic acid and linolenic acid. The oil content of the seed is quite
high at about 40%. The composition of edible portion of Okra is reported (Gopalan et al., 2007).

However, biochemical characterization of tender fruits of *A. esculentus* and its therapeutical effects, particularly against arthritis was not studied. Therefore, in the present study is aimed to test the effect of various extracts of tender fruits of *A. esculentus* in curing the arthritis in rheumatoid antigen treated Swiss albino mice.