Introduction
Parasitic diseases have been responsible for major health problems in developing and underdeveloped countries of tropical and subtropical belts. Helminth parasitic diseases, viz., Filariasis, Ascariasis and Hookworm infections contribute significantly to the problem. Human Lymphatic filariasis (LF) is a mosquito borne disease of tropics and subtropics (Sasa, 1976). It is one of the oldest and most debilitating diseases in the world (Nelson, 1979; Dean, 2001). Over 120 million people in 80 countries are afflicted with disease, with 20% of the world’s population living at the risk of this disease (Fig. 1) (WHO, 1997a). LF is caused by three species of nematode parasites, namely *Wuchereria bancrofti*, *Brugia malayi* and *Brugia timori* belonging to the Phylum: Nematoda, Class: Secernentea, Order: Spirurida, Sub-order: Spirurina, Family: Filaroidae (Yamaguti, 1961). *W. bancrofti* accounts for 90% of LF infections worldwide, while *B. malayi* is prevalent only in some parts of South & South East Asia, and *B. timori* is found only in Indonesia (WHO, 2002). India alone contributes to about 40% of the total global burden of this disease and there are approximately 21 million people with symptomatic filariasis and 27 million who have asymptomatic microfilaraemia, while a total of 553 million people are at risk of infection (Ramaiah et al., 2000). A total of 289 districts in India were surveyed for filariasis until 1995; out of which 257 were found to be endemic. *W. bancrofti* is the predominant species accounting for about 98% of the national burden, widely distributed in 17 states and union territories of the country (Sabesan et al., 2000; Das and Ramaiah, 2002).

Globally, today, LF remains the second leading cause of permanent and long-term disability (Evans, et al., 1993). The disfigurement of body parts caused by chronic disease manifestations of LF such as lymphedema and elephantiasis in men and women along with hydrocele in men inflict stigma and disability on the affected people (Evans, et al., 1993). Adult parasites of these species lodge themselves in lymphatic vessels, causing lymphatic damage that leads to elephantiasis, hydrocele and other symptoms such as Tropical Pulmonary Eosinophilia (TPE), and chyluria. Nocturnally periodic
Fig. 1. Lymphatic filariasis – endemic countries and territories, 2004.
Source: (www.filariasis.org/docroot/docs/7_Media/worldmapPDF.pdf)
W. bancrofti (Cobbold et al., 1877) is transmitted by the tropical house mosquito, Culex quinquefasciatus (Say, 1823) which is the most formidable and widely prevalent vector. Though there are several species of vectors like Culex, Anopheles, Aedes and Mansonia. However, more than 50% of the LF infections worldwide are transmitted only by C. quinquefasciatus (Southgate, 1984).

LF impairs the mobility, social life, education, employment, marriage prospects and also marital relations and sexual life. It restricts the occupational activities leading to lower productivity by as much as 27%, (Ramu et al., 1996) leading to significant economic loss (Sabesan et al., 1992; Ramaiah et al., 2000). India alone suffers an economic loss of 1 billion US $ annually resulting from decreased productivity in LF infected people (Ramaiah et al., 2000). The number of infected individuals worldwide is on the increase due to rapid urbanization and poor sanitation that serves as a good breeding ground for the growth of vectors. There are two ways for controlling the disease, one is vector control and the other is chemotherapy. The biocides, like Bacillus sphaericus and Bacillus thuringiensis, are alternatives but have limited options for vector control in a longer period of time (Porter et al., 1993). The development of new insecticides, transgenic mosquitoes in future may be useful for control of the disease. Several findings during the last decade and a half, particularly the discovery of the effect of DEC (6 mg/kg body weight) and Ivermectin (200-400 μg/kg body weight) in single dose (Ottesen, 1997), have brought about newer tools to manage morbidity (Dreyer, et al., 2002) and raise the hopes of LF control. Co-administration of either of these drugs with albendazole (400 mg) is recommended for mass treatment under the LF elimination programme (Ottesen, 2000). DEC-salt trials in India (Rao et al., 1981; Subramanyam and Venkateshwaralu, 1996) indicated that consumption of the salt for a minimum period of 6-9 months decreases mf-prevalence by 70–100% (Gelband et al., 1994). New diagnostic tools such as immunological tests, based on monoclonal antibodies (Mabs) to filarial parasites, have been developed recently, such as the Og4C3 (More and Copeman, 1990) and
Immuno-Chromatographic Test (ICT) (Weil et al., 1997), which detect W. bancrofti circulating filarial antigens (CFA). Additionally, DNA based diagnostics for species specific PCR to detect vectors infected with W. bancrofti has strengthened the hopes (Ramzy et al., 1997; Hoti et al., 2001) and led to development of a global programme to eliminate LF as a Public health problem by the year 2020.

Rationale and objectives of present study

As stated above W. bancrofti is the major cause of human lymphatic filariasis, an important public health problem in India, accounting for 40% of the global burden (Ramaiah et al., 2002). Furthermore, Mass Drug Administration (MDA) is on-going in several states of India to eliminate lymphatic filariasis, which might be putting intense selective pressure on the parasite populations. This situation warrants assessing the genetic structure of W. bancrofti parasite populations existing across the country and also to monitor genetic changes that may take place in future. When this study was initiated there was no reproducible robust molecular markers available to study the genetic diversity among populations of this parasite occurring in different geo-climatic endemic zones of India.

Prevention and control of parasitic diseases in humans will always be associated with problems of variation in outcome or failure and solutions to such problems may require knowledge of geographical distribution of the parasite genotypes. In India, occurrence of different physiological strains of W. bancrofti parasite, based on the periodicity of microfilaria (mf) in the peripheral blood of man, viz., nocturnally periodic, nocturnally sub-periodic and diurnally sub-periodic has been reported, as also the occurrence of ecological races of the species (Sasa, 1976). Variations in the spectrum of disease and its symptoms, varying mf counts in patients, and surface immunoreactivity of mf from region to region has been reported, indicating the occurrence of different strains/genotypes of this parasite (Rao et al., 1977a; Ravindran et al., 1994). These findings indicate, the occurrence of different
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genotypes/strains of *W. bancrofti* in India and this has important implications in control operations. Understanding heterogeneity of parasites across the country would yield benefits in the long run. Therefore, there is a need to develop tools to differentiate genotype\strains of *W. bancrofti* at spatially hierarchical levels. *W. bancrofti* manifests a spectrum of pathological conditions as stated above and it is reported that exist variations in the clinical spectrum in the different geographic regions of India (Rao et al., 1977a). Since, the pathological conditions in the host are determined by the molecules of the parasite, the spectrum of clinical manifestation might be due to their polymorphic nature. One of the parasite molecules that is reported to be involved in pathogenesis of LF is polyprotein antigen (gp15/400), a homologue of ABA polyprotein of *Ascaris* species (Kennedy, 2000a). It might be interesting to see if there is any correlation between polymorphism of this antigen of *W. bancrofti* from different climatic regions and their clinical spectrums. Keeping these issues in mind, the following objectives were set for this thesis work.

Objectives

1. To develop suitable marker(s) for studying the genetic diversity and population genetics of *W. bancrofti*.
2. To understand the genetic diversity of *W. bancrofti* populations at spatially hierarchical levels.
3. To deduce phylogeography of *W. bancrofti* in India.
4. To study the polymorphism of the polyprotein (gp15/400) gene of *W. bancrofti* populations from different geoclimatic regions of India.