1. Introduction
DEVELOPMENT AND EVALUATION OF POLYCLONAL AND MONOCLONAL ANTIBODY BASED IMMUNO-ASSAYS FOR THE DIAGNOSIS OF WUCHERERIA BANCROFTI INFECTION IN HUMANS

1. INTRODUCTION

Lymphatic Filariasis (LF), also known as elephantiasis, is a widespread tropical disease caused by three species of nematode parasites namely *Wuchereria bancrofti*, *Brugia malayi*, and *Brugia timori* belonging to the Order, Filarridea, under the Superfamily, Filarioidea, the Family, Onchocercidae. Adult parasites of these species lodge themselves in lymphatic vessels, causing lymphatic damage that leads to elephantiasis and/or hydrocele and other symptoms such as tropical pulmonary eosinophilia, chyluria etc. The disease is transmitted by various genera of mosquitoes belonging to the Order Diptera, family Culicidae. The mosquito vectors, *Culex quinquefasciatus*, *Mansonia annulifera* and *Anopheles barbirostris* are responsible for transmitting the larvae of *W. bancrofti*, *B. malayi*, and *B. timori* respectively.

Nearly one billion people in 80 endemic countries and territories are at the risk of filarial infection and over 120 million have already been affected by it, 43 million (40 million with bancroftian filariasis
and 3 million with brugian or timorian filariasis) of them are incapacitated and disfigured by it (WHO, 2001).

One-third of the people infected with the disease live in India accounting for 42.8% of the global burden of the disease (WHO, 1998). It is estimated that 454 million people are at risk of infection of which 29.7 million are microfilaria (mf) carriers and 22.5 million are diseased (Datta, 2000).

LF has a broad clinical spectrum ranging from asymptomatic to chronic manifestation resulting in elephantiasis and/or hydrocele. The mf stage of the parasite circulate in the blood in a periodic pattern depending upon the species. In W. bancrofti infection the mf exhibit nocturnal periodicity and in W. bancrofti pacefica the mf exhibit diurnally sup-periodicty which occurs in many islands in South Pacific, small foci in Nicobar Island, in French Polynesia and in Thailand. In case of human B. malayi infection, the mf are nocturnally periodic, and nocturnally sub-periodic strain of mf of B. malayi is common in animal reservoirs. In case of B. timori the mf exhibits nocturnal periodicity.

Interest in filariasis elimination is growing among public health programmers around the world, partly because of the burden that filariasis and other chronic parasitic diseases place on the health
budget. The recent call from the 50th World Health Assembly to governments and policy makers to provide resources to eliminate lymphatic filariasis has given the political will to realize this goal by 2020 (WHO Fact sheet 1998). To achieve this goal the necessary tools for accurate and early detection of the disease is very important which in turn will lead to effective cure and control of filariasis. By the utility of these tools, in delimiting the endemic areas, also by the continuous monitoring of filariasis elimination programme, the certification of the areas as "disease free" can be achieved.

Currently diagnosis of the infection is carried out by parasitological examination of thick blood smear collected at night (because of the nocturnal periodicity of the parasite) for the presence of mf. This method has several drawbacks such as lower sensitivity, cumbersome, tedious and has poor community acceptance. Though the mf can be detected in blood of microfilaraemic asymptomatic individuals, the same cannot be found in blood in the later stages of this disease such as the manifestation of lymphedema (Manson-Bahr & Bell, 1987) and hence this method is not useful in detecting low level microfilaraemia and prepatent (L3 stage to adult stage) infection. Hence, for effective cure and control of filariasis, accurate and also an early diagnosis is important. To overcome this problem the need was to develop immunodiagnostic assays that could detect the infection in day
blood yet being simple and cost effective. Immunological tests, based on monoclonal antibodies (MAbs) to filarial parasites, have been developed elsewhere recently like the Og4C3 test (More & Copeman, 1990) and immuno-chromatographic test (ICT) cards (Weil et al., 1997) to detect *W. bancrofti* circulating filarial antigen (CFA) and are now commercially available. However these tests are expensive, costing approximately Rs. 80.00 and 150.00 per test respectively, and need to be imported and hence majority of the people cannot afford to use them. Therefore, there is a need to develop immuno-diagnostics indigenously, producing such kits will prevent logistics of import, apart from facilitating easy availability. With these principles in consideration the objectives of the present study were:

i) To develop poly/monoclonal antibodies for detecting circulating filarial antigens in individuals.

ii) To evaluate the specificity and sensitivity of the poly/monoclonal antibodies in detecting the filarial infection.

iii) To develop specific immunodiagnostics for early detection of developing stages of *W. bancrofti* such as L4.