Introduction

Epilepsy is one of the oldest medical disorders known. The word epilepsy derived from the Greek word *epilambanein*, meaning to be seized or to be overwhelmed by surprise. Epilepsy is one of the most common serious disorders of the brain, affecting at least 50 million people worldwide. It knows no geographical, racial or social boundaries. Epilepsy accounts for 1% of the global burden of disease, determined by the number of productive life years lost as a result of disability or premature death. Among primary disorders of the brain, epilepsy ranks with depression and other affective disorders, Alzheimer’s disease, other dementias and substance abuse. Among all medical conditions, it ranks with breast cancer in women and lung cancer in men. Eighty per cent of the burden of epilepsy is in the developing world, where 80–90% of people with epilepsy receive no treatment at all. It is also necessary to recognize that epilepsy consists of more than seizures for the affected individual and affects his or her family. Epilepsy leads to multiple interacting medical, psychological, economic and social repercussions, all of which need to be considered. (WHO epilepsy Atlas 2005)

Epilepsy is characterized by a tendency to recurrent seizures. The word ‘seizure’ is derived from the Latin word *sacire* meaning ‘to take possession of’ is the clinical manifestation of an abnormal, excessive, hypersynchronous discharge of a population of neurons. The hypersynchronous discharges that occur during a seizure may begin in a very discrete region of brain and then spread to neighbouring regions. Seizure initiation is characterized by two concurrent events: 1) high-frequency bursts of action potentials and 2) hypersynchronization of a neuronal population. The synchronized bursts from a sufficient number of neurons result in ‘spike discharge’ on the EEG. At the level of single neurons, epileptiform activity consists of sustained neuronal depolarization resulting in a burst of action
potentials, a plateau-like depolarization associated with completion of the action potential burst and then a rapid repolarization followed by hyperpolarization. This sequence is called the paroxysmal depolarizing shift. The bursting activity resulting from the relatively prolonged depolarization of the neuronal membrane is due to influx of extracellular Ca\textsuperscript{2+}, which leads to the opening of voltage-dependent Na\textsuperscript{+} channels, influx of Na\textsuperscript{+} and generation of repetitive action potentials (Schiller et al., 2004). Seizure propagation is the process by which a partial seizure spreads within the brain, occurs when there is sufficient activation to recruit surrounding neurons. This leads to a loss of surrounding inhibition and spread of seizure activity into contiguous areas via local cortical connections and to more distant areas via long association pathways such as the corpus callosum.

Because of the well organized and relatively simple circuits within the entorhinal-dentate-hippocampal loop, the limbic system has been intensively studied in experimental models of epilepsy. These investigations have led to two theories regarding the cellular network changes which cause the hippocampus, among the most common sites of origin of partial seizures, to become hyperexcitable. The first proposes that a selective loss of interneurons decreases the normal feed-forward and feed-back inhibition of the dentate granule cells, an important group of principal neurons (Stief et al., 2007). The other theory suggests that synaptic reorganization follows injury and creates recurrent excitatory connections, via axonal "sprouting," between neighboring dentate granule cells (Cohen et al., 2003). More recently, it has been proposed that the loss, rather than being of GABAergic inhibitory neurons, is actually of excitatory neurons which normally stimulate the inhibitory interneurons to, in turn, inhibit the dentate granule cells. These mechanisms of hyperexcitability of the neuronal network are not mutually exclusive, could act synergistically and coexist in the human epileptic brain.
Temporal lobe epilepsy (TLE) is one of the most common forms of intractable epilepsy. Patients affected often have similar clinical history, including an initial precipitating injury such as childhood febrile convulsions, status epilepticus (SE) or trauma. Between this injury and the emergence of recurrent complex partial seizures, there is usually a latent period of several years (Turski et al., 1989). Frequently associated with this epilepsy is the presence of hippocampal sclerosis (HS). HS is defined by specific neuronal loss throughout the hippocampus, with severe damage in the prosubiculum. CA1, CA4 and hilus in contrast with slighter damage in granule cells and relative sparing of CA3 and especially CA2 region. Human studies strongly support the view that HS probably initiates or contributes to the generation of most TLEs (Engel et al., 1996). However, there is a growing body of evidence that amygdala, limbic thalamus and entorhinal cortex may be injured in TLE (Jutila et al., 2001). The respective role of various hippocampal or extrahippocampal structures in the genesis of the disease remains unknown.

Induction of SE by systemic application of pilocarpine and subsequent occurrence of spontaneous seizures is probably the most attractive animal model, for the study of temporal lobe epilepsy. Pilocarpine treatment is characterized by generalized convulsive SE in rodents, which represents the initial precipitating injury. After a latent period, adult rats exhibit spontaneous recurrent seizures (SRS) during the remainder of their life. The EEG and behavioral features of these seizures resemble those of complex partial seizures (Leite & Cavalheiro, 1995). This model shares many histopathological and molecular changes that have been characterized in neurosurgical resections and post mortem specimen from TLE patients. Surprisingly little is known on the molecular and cellular signaling during induction of SE and the role of muscarinic and glutaminergic functional regulation in chronic epilepsy models.
Bacopa monnieri (Brahmi) has been used by Ayurvedic medical practitioners in India for almost 3000 years. The earliest chronicle mention is in the Ayurvedic treatise, the Charaka Samhita (100 A.D.), in which Brahmi is recommended in formulations for the management of a range of mental conditions including anxiety, poor cognition, lack of concentration and epilepsy. According to Charaka, Brahmi acts as an effective brain tonic that boosts one's capabilities to think and reason. The Sushruta Samhita (200 A.D.) attributes the plant with efficacy in maintaining acuity of intellect and memory. Pharmacologically, it is understood that Brahmi has an unusual combination of constituents that are beneficial in mental inefficiency and illnesses and useful in the management of convulsive disorders like epilepsy. Bacosides, Brahmi's active principle component responsible for improving memory related functions, are attributed with the capability to enhance the efficiency of transmission of nerve impulses, thereby strengthening memory and cognition (Kishore et al., 2005).

The present work is to understand the alterations of total muscarinic, muscarinic M1 and glutamate receptors in the brain regions of pilocarpine induced epileptic rats. The work focuses on the evaluation of the antiepileptic activity of extracts of Bacopa monnieri, Bacoside A and Carbamazepine in vivo. The molecular changes in the muscarinic M1 receptors in the pre- and post-treated epileptic model with Bacopa monnieri, Bacoside A and Carbamazepine were also studied. These studies will help us to elucidate the functional role of muscarinic and glutamate receptors in epilepsy.
OBJECTIVES OF THE PRESENT STUDY

1. To study the antiepileptic activity of whole plant extract of *Bacopa monnieri* and Bacoside A in pilocarpine induced Temporal Lobe Epileptic rat model.

2. To study the cholinergic activity using acetylcholine esterase assay in the brain regions- hippocampus and brainstem in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, post-treated epileptic rats.

3. To study the muscarinic general receptor binding parameters in the hippocampus, cerebellum and brainstem in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, post-treated epileptic rats.

4. To study the muscarinic M1 receptor binding parameters in the hippocampus, cerebellum and brainstem in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, pre- and post- treated epileptic rats.

5. To study the muscarinic M1 receptor gene expression in the hippocampus in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, pre- and post-treated epileptic rats.

6. To study the muscarinic M1 receptor gene expression in the Cerebellum and Brainstem in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, post-treated epileptic rats.

7. To study the glutamate dehydrogenase activity in the brain regions- hippocampus, cerebellum and brainstem in epilepsy, *Bacopa monnieri* post-treated epileptic rats.

8. To study the glutamate receptor binding parameters in the hippocampus, cerebellum and brainstem in epilepsy, *Bacopa monnieri* post-treated epileptic rats.

9. To study the NMDA R1 and metabotrophic glutamate 8 receptor gene expression in the hippocampus, cerebellum and brainstem in epilepsy, *Bacopa monnieri* post-treated epileptic rats.

10. To perform the Neo-Timm Staining in the hippocampus in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, post-treated epileptic rats to observe reversal of mossy fibre sprouting.
11. To perform neurophysiologic analysis of the electrical activity of the brain using electroencephalogram in epilepsy, Carbamazepine, *Bacopa monnieri* and Bacoside A, pre- and post-treated epileptic rats.

12. To study the spatial learning ability by Morris water maze experiment in epilepsy, *Bacopa monnieri* post-treated epileptic rats.