Introduction
and Literature review
Introduction and Literature Review:

The homeopathic mode of treatment was first discovered by a German physician named Samuel Hahnemann (1755-1843). Dr. Hahnemann conducted some experiments on himself. He noticed that after taking malarial remedy, China (Peruvian bark), he experienced symptoms similar to those of patients of malaria. He repeated the experiments, which were later termed as "provings" on his family members and friends and found that the substance that produces symptoms in healthy subjects can also cure such symptoms when applied in ultra-low doses after serial dilution and succussion of that substance. Thus, he apparently confirmed the basic principle of homeopathy to be "Like cures like" or "Similia Similibus Curentur". He further noticed with great interest that diluting and vigorously shaking his remedies, (a process that was later termed as "potentization" or "potentiation") often rendered the remedy more potent in terms of clinical response. This method of homeopathic treatment soon became very popular and spread all over the country because this system was very simple and less painful than the other prevailing systems like blood letting, crude surgery etc.

However, at the end of the nineteenth century homeopathy became a subject of vehement attack for the following reasons:

i) The potentized homeopathic drugs above 12C did not theoretically speaking contain even a single molecule of the original drug substance.

ii) The homeopathic doctrine of "more diluted" the remedy, the "more strong and long lasting the effect" could not be scientifically explained within available knowledge.

iii) The mechanism of action of ultra-high dilution used in microdoses could not be explained with a firm scientific basis.

Hahnemann defined the "Law of Similes" as the central principle of homeopathy, which was known to cause symptoms similar to a particular disease when given to a patient in an extreme diluted form (Khuda-Bukhsh,
This was believed to induce a restorative process in the body that would counteract the effects of disorder being treated. Homeopathic dilutions can be distinguished into two types based on the degree of dilution applied to the starting material (Jutte and Relay, 2005). One type of traditional homeopathy involves diluting natural compounds extensively, but low, presumably biologically relevant levels of active ingredients remain in solution (Moffett et al., 2006). This type has been referred to as "Hormetic method" (Arndt-Schultz law) (Jutte and Relay, 2005). Another method of homeopathy that is extensively practiced uses remedies diluted beyond the Avogadro's limit. These potentized medicines are referred to as BRAN (beyond the reciprocal of Avogadro's number), (Moffett et al., 2006). According to the homeopathic clinicians and researchers, the homeopathic preparations are made more potent, or are "potentized", by this kind of extreme dilution process. In the homeopathic potentization procedure, 1 ml of mother tincture is generally diluted with 99 ml of ethanol and given 10 jerks or succussions (a vigorous type of shaking either by hand or machine) to produce the potency 1C. Similarly, 1 ml of the drug solution at potency 1C is again added with 99 ml of ethanol and followed by 10 succussions to produce the potency 2C and in this way by successive dilutions and succussions, further potencies like 30C, 200C, and beyond are produced. Therefore, at high dilutions, say beyond potency 12C (beyond Avogadro's limit i.e., $6.023 \times 10^{23}$ approximately), the solution is unlikely to contain even a single molecule of the original drug substance (i.e., initial drug substance). It is claimed that the succussion process is critical for "potentizing" homeopathic preparation and that further the solution is diluted and succussed, the more effective it will be. This is one of the controversial subjects that cannot be resolved within the present state of our scientific knowledge. Therefore, some people are somewhat inclined to believe in the homeopathic remedies used in crude form or in dilution not that high to cross the Avogadro's limit while some people will not accept the remedies diluted beyond Avogadro's limit. For this reason, the initial researches were done with the primary goal of demonstrating the efficacy particularly of the potentized forms of medicines in various organisms.
Further, controlled experiments were set up in which a placebo fed control group was also maintained. This was to reveal if the effects of homeopathic drugs were decisively greater or better than the placebo. A large number of experiments have been carried out to prove the efficacy of homeopathic drugs, both \textit{in vivo} and \textit{in vitro}.

\textbf{In vitro laboratory experiments and a few hypotheses on biological response}

Many of the prominent studies aimed at demonstrating the biological action have been conducted in toxicology and immuno-allergology (Poitevín, 1990). The term "Hormesis" was proposed by Southam and Ehrlich (1943) to describe "a stimulatory effect of sub-inhibitory concentrations of any toxic substance on an organism." The literary meaning of this Greek word is "excitation by impulse." Subsequently, Stebbing (1982) used this term to describe the stimulation of growth by low levels of inhibitors. Wiegant \textit{et al} (1997) have done considerable work on the hormetic effect. According to them, the stimulation of a disturbed self-recovery by the application of the similia principle is considered to be the essence of homeopathy. In this experiment, human fibroblasts were exposed to a threatening condition, chemical (arsenite and cadmium) or a physical (increased temperature) nature, and extensive analyses were made with regard to processes which might be considered as self-recovery, i.e., cells' potential of temporal increased proliferation and additional development of resistance to the threatening condition. Further specific analysis was made of a number of molecular processes particularly on the stress proteins or heat shock proteins. Heat shock proteins are a special class of protein molecules which have a function in the prevention of damage and assistance in the recovery. They showed that small doses of threatening condition (increased temperature of arsenite) would bring about a clear improvement of the activation of synthesis of hsp70 (Van Wijk and Wiegant, 1994, 1998). Therefore, ultra low doses of the same toxic substances could evoke suitable recovery responses at the molecular and physiological levels. Studies on crystal induced inflammation as well as...
insulin receptor activation by oxy-anions have been utilized to understand how the hydrate structure of certain types of silica could activate or some anesthetic agents could activate specific types of cell surface protein directly or indirectly due to their co-incidental complementary structure (Matsumoto, 1994, 1995) Researchers conducted by several others have tested the biological effects of I$_E$ crystals and have found remarkable effects. Lo and Bonavida (1998) tested a sample of crystals on blood and found a 2-100 fold increase in cytokine activity (mediators of immune function that protects against infection and tumor growth). Important signal transduction studies are also being carried out in other institutes (e.g., Samueli Institute, USA). Davenas et al. (1988) showed that very diluted anti-serum against Ig-E triggered human basophil de-granulation. However, Maddox et al. (1988) could not verify the result of Davenas et al. (1988) although some other researchers from different countries replicated the study and obtained results similar to that of Davenas et al. (Belon et al. 1999, 2004) Recently immunological implications of homeopathic remedies on both animals and humans have been thoroughly reviewed by Bellavite et al. (2005, 2006a, 2006b, 2006c, 2006d) which have elaborately discussed various immunological aspects involved in homeopathic studies.

**In vivo laboratory studies**

Many *in vivo* studies were designed with the primary objective of demonstrating the efficacy of homeopathic remedies in respect of their physiological, biochemical, cytogenetical and molecular changes. Many of these studies were carried out on mice and rats and also occasionally on other higher mammals like cattle and horse. Khuda-Bukhsh and his collaborators (Khuda-Bukhsh et al. 1982, 1983 1986, 1995, Khuda-Bukhsh and Maity, 1991, Khuda-Bukhsh and Banik, 1991, Banik and Khuda-Bukhsh, 1991, 1995, 1996) studied the efficacy of some potentized homeopathic drugs in reducing genotoxicity and clastogenicity induced by X- irradiation in mice, keeping suitable controls. Further modulation of cytogenetical effects.

Cancer is a dreadful disease that kills a large number of people every year If cancer progresses to an advanced condition practically there is no chance of survival of the patient, no matter what kind of advanced treatment is offered But, if cancer is detected at early stage it can be tackled in a better way and can be cured even But, the problem is in many cases by the time when cancer is detected, it often has progressed to a certain stage from where the eradication of the disease becomes quite difficult, and often needs palliative care to alleviate the sufferings of victim In recent years supportive CAM therapies are being advocated as these seem to offer better results (Ernst 2003, 2006, Ricotti and Delanty 2006, Simpson 2006, Balzarnin et al 2000, Richardson and Stross, 2000, Shen et al 2002, Chrystal et al 2003, Molassitus et al 2005,2006) Homeopathy is being explored for screening
those drugs which may have anti-cancerous activity, by subjecting them to rigorous scientific tests, often by utilizing *in vivo* mammalian or *in vitro* cancer cell lines. Although there has already been some published account on the efficacy of some homeopathic drugs in amelioration of cancer in animals (Choudhury 1980, Biswas and Khuda-Bukhsh, 2002, 2004, Biswas *et al* 2005, Pathak *et al* 2006) and in human beings (Sen Pathak *et al* 2003), more such studies are necessary with drugs having proven ability to ameliorate symptoms of some specific types of cancer. Some nosodes derived from cancer cells or tissues have also been successfully used in clinical practices, (e.g., Carcinosin and Cholestennum prepared from either breast cancer tissue by potentizing them through homeopathic procedure (Boencke *et al* 1976).

*Mynca cenfera* Linn is a shrub belonging to family Myricaceae. It is also known as Southern Bayberry or Southern Wax Myrtle. Native Americans use a decoction of the leaves and stems of *Mynca cenfera* to treat fever and roots to treat inflamed tonsil and stomach ache, and as a stimulant (Moerman, 1986). This plant can be easily grown because it can tolerate a variety of landscape settings from full sun to partial shade, wet swamp lands or high, dry and alkaline areas (Gilman and Watson, 1994). Bayberry is known to contain various phytochemical agents including saponins, resins, albumin, aluminium, chromium, cobalt, flavonoids etc (Blake, 2004). *Mynca cenfera* in homeopathic potencies is used in disease where thick, viscid secretions of the mucous membrane or catarrhal state of the intestinal tract and liver disorder occur (Clarke 2003, Vol-II, Materia Medica). In the present study an induced *in vivo* liver cancer model (Kitagawa *et al* 1979, 1984, Uchida and Hirono, 1979, 1981, Biswas and Khuda-Bukhsh, 2002, 2004, Biswas *et al*, 2004, Pathak *et al* 2006a, 2006b) has been used. Mice were chronically fed with an azo-dye N-N' dimethyl-4-Phenyldiazobenzene (p-DAB) which initiates liver cancer on long term feeding, which is further accelerated by 5-ethyl-5 phenyl 2, 4-6 (IH3H5H) pyrimidinetrione commonly known as Phenobarbital (PB) that acts as promoter.
The main objectives of the present study are

1) To evaluate if *Myrica cenfera* has any anti-tumorigenic, anti-genotoxic and anti-clastogenic effects in mice chronically fed with carcinogens against suitable controls, by using relevant cytogenetical and biochemical protocols,

2) To study the effect of the carcinogens on some organs including liver, the target organ of the carcinogens by utilizing electron microscopic methodologies and to study the positive changes, if any, brought about by the homeopathic medicines.

3) To correlate the effects, if any, of the different potencies of *Myrica cenfera* namely, Myrica- 30 and Myrica- 200 and to compare them with that of Myrica mother tincture.