INTRODUCTION:

Liver have a vital role in regulation of various physiological processes. It is implicated in numerous functions such as biotransformation, synthesis of several biomarkers like SGPT, SGOT, clotting factors and helps in storage of glycogen and so on. Besides, this it has role in detoxification of various drugs and xenobiotics. The bile synthesized by the liver has among other thing, an important function in digestion of fatty molecules. Liver diseases are among the most serious ailments in the world. They may categories as acute or chronic hepatitis (Inflammatory hepatocytes), hepatosis (Non inflammatory hepatocytes), NAFLD (Non Alcoholic Fatty Liver Diseases) NASH (Nonalcoholic steatohepatitis) and liver cirrhosis (Degenerative disorder leads to fibrosis of the liver). Hepatotoxicity is commonly known as toxicity of liver through various chemical entities (certain antibiotics, chemotherapeutics, aflotoxin, corbon-tetrachloride and chlorinated hydrocarbon) they are also known as hepatotoxins.

According to WHO, liver disorders accounted for 1.8% of all death in Europe (WHO,s wide geographical definition), causing around one lakh seventy thousand deaths per year. However, in recent years, a liver disorder has also be major health hindering problem in some western European countries. Over the one decade the associated mortality rate has incresesed. Approximately twenty thousand death caused by liver disorder found every year.

Due to westernization of lifestyles, the prevalence of nonalcoholic fatty liver disease (NAFLD) has been rising around the world in recent years. The predictable number of patients with NAFLD has reached 80-100 million in the U.S. and the corresponding number of patients in Japan has been estimated at 10-20 million. The prevalence of NAFLD and nonalcoholic steatohepatitis (NASH) is increasing and is becoming a major target disease not only in Western countries, but also in Japan.

Non-alcoholic fatty liver disease (NAFLD) is a common clinical situation which is fast assuming importance as a feasible precursor of more serious liver disorders, including cirrhosis of the liver and hepatocellular carcinoma. There are no data in the published in literature on the prevalence of NAFLD in India. Fatty liver was seen more commonly in
males (26.9%) than in females (13.8%). NAFLD is perhaps as common in developing world as in the developed countries despite a lower prevalence of obesity. Indian males may have a greater genetic predisposition to developing NAFLD. This non alcoholic fatty liver disease occurrence age group between 40-49. According to the recent WHO data available in April 2011 Liver disease deaths in India reached 2,08185 or 2.31% of total deaths and India ranked as 27 positions.

The foremost reason for recurrent episode of hepatotoxicity is because liver comes directly to contact with xenobiotics (drug/toxin) concentrated blood from gastrointestinal tract thus making it most exposed. Drug/toxin taken or generated in the body increase levels of Reactive Oxygen Species (ROS), which cannot quenched by non–enzymatic scavengers (antioxidants) as well as enzymatic system (Glutathione conjugation) are involved in the detoxification of reactive oxygen species. Oxidative stress, resulting from imbalance in the generation of free radical and antioxidant defense molecules, affects biological macromolecules causing their structural alterations that lead to cell damage and its death. This phenomena is considered to be a important factor in the pathogenesis of a numerous variety of liver diseases.

The management of liver disease is still hurdles for modern medication. No drug has been developed in modern system of medication which trigger the liver function, protect hepatocell from damage or helps in regeneration of hepatic cell. In the scarcity of a reliable liver protective drug and severe unfavorable side effects of synthetic drug, there fore raising focus to follow systematic research methodology and evaluate scientific root for traditional herbs and herbs derived medicines that are claimed to possess hepatoprotective activity.

In recent era, scientists having tremendous scope and evaluated hepatoprotective potential of traditionally used medicinal plants in various animal models. Thus herbs and herbs derived medicines are gradually gaining fame in the treatment of liver disease and its complications which associated with oxidative stress. The major characteristic of herbal medicine seems to be their supposed efficacy, low occurrence of serious adverse effects and more economic (Prasanna shama K et al. 2012). Naturally occurring products
and their derivatives have been a successful source of bioactive molecules in medicine before the advancement of other modern therapeutics in the post-economic era.

The few decades has seen the proliferation of a huge number of scientific studies paying attention towards functional food like polyphenols such as flavonoids, phytoalexins and phenolic acid. Functional foods are those food items which provide health benefits beyond their nutritive value and are considered as a suitable alternative for potent drugs with severe systemic toxicities such as chemotherapeutic agents. Of all functional foods, polyphenols attracted many researchers as they possess a wide spectrum of health benefits including anti-cancer activity. Flavonoids belong to a group of functional food with polyphenolic structures and are found in fruit, vegetables, grains, bark, roots, stems, flowers, tea, and wine. Flavonoids are classified into flavonols, flavones, catechins, flavonones and isoflavonoids. Many literatures reported that flavonoids posses the various biological activity like hepatoprotective, antioxidant, antimicrobial, antiviral, gastroprotective, cytotoxic, antineoplastic, anti-inflammatory, antidiabetics, antihypertensive and hypolipiedemic.

Despite having a wide spectrum of biological activity, medicinal value of flavonoids cannot be used due to poor biodisponibility, poor solubility and metabolism. Which contribute to reduced therapeutic hepatoprotective activity and. Therefore there is an extensive need for combinatorial and novel drug delivery system such as dual loaded Flavono-nanoparticulate system (FNPs).

Nanotechnology is an emerging field that manipulates materials in the atomic and molecular levels (1 to 100 nm). Nanotechnology finds potential applications in almost all fields wherein biomedical application plays a vital role in our day to day life. The herbal leaves and its extracts have many biomedical applications which are evident from our ancient time. The processing of herbal nanoparticles (dissolved, entrapped, encapsulated
or attached to a nanoparticle matrix) from medicinal plants shows increasing widespread medical applications.

In the past few decades, tremendous advancement has been made toward formulating Nanoparticles containing therapeutic products have gained much interest in scientific and commercial fields due to their potential for enhancing the biodisponibility and site specific drug delivery and accordingly the optimization of novel drug delivery. A 2006 European technological observatory survey shown that more than 180 pharmaceutical companies developing nanoscale formulation\(^\text{10}\). There are plenty of novel herbal based nanoparticulate formulations available in the market like biodegradable polymeric nanoparticles, nanocapsules, liposomes, \textit{}, nanoemulsions, microsphere, dendrimeric nanoparticles, transferosomes and ethosomes has been reported using plant extracts and its bioactive phytochemical constituents. Nowadays biodegradable polymeric NPs have attracted considerable attention as potential drug delivery system in view of their applications in the control release of drug moiety, their capable to target specific organ or tissues, as carriers of DNA in the gene therapy, and in their ability to deliver proteins, peptides and genes through a entral route of administration\(^\text{11}\). The majority of review article dealt with NPs of poly(D,L-lactide),poly(D,L-glycolide)PLG,poly(lacticid)PLA,poly(lactide-co-glycolide) poly(cyanoacrylate)PCA and EPO. Significant achievement in gaining the attention of researchers towards the loading of dual bioflavonoid into the polymeric chain network is increasingly addressed in recent while (Moorthi C. 2013).

Foremost concept of fabricating nanoparticles as a delivery system are to control particle size, surface properties and release of pharmacologically active agents in order to achieve the site-specific action of the drug at the therapeutically optimal rate and dose regimen.

The advantages of using hepatoprotective nanoparticles as a drug delivery system include the following:

1. Particle size and surface characteristics of nanoparticles can be easily manipulated to achieve both passive and active drug targeting after parenteral administration.
2. They control and sustain release of the drug during the transportation and at the site of localization, altering organ distribution of the drug and subsequent clearance of the drug so as to achieve increase in drug therapeutic efficacy and reduction in side effects.

3. Controlled release and particle degradation characteristics can be readily modulated by the choice of matrix constituents. Drug loading is relatively high and drugs can be incorporated into the systems without any chemical reaction; this is an important factor for preserving the drug activity.

4. Site-specific targeting can be achieved by attaching targeting ligands to surface of particles or use of magnetic guidance.

5. The system can be used for various routes of administration including oral, nasal, parenteral, intra-ocular etc.

A variety of herbal novel formulations containing nanoparticles of drugs like Silybum marianum, Ginkobiloba, Curcuma longa and soon have exposed their potential as controlled and targeted drug delivery systems. Several patents climbing the sustained release formulations of Boswellia serrata, Calendula officinalis, Commiphora wightii and some Chinese drugs using nanotechnology have been documented\textsuperscript{12-16}.

However a systematic study of fabrication, characterization and pharmacological evaluation of dual loaded flavononanoparticles for anti-oxidant and hepatoprotective study is not reported much in the literature.

In this context, the present study is aimed to encapsulate dual loaded selected flavonoids in polymeric nanoparticles to overcome the limitation of instability and improve their biodisponibility (bioavailability) and site based targeting. It is also proposed to evaluate the \textit{in vitro} and \textit{in vivo} anti-oxidant, pharmacological activity and hepatoprotective activity of selected dual loaded flavononanoparticles with the single loaded nanoparticles as well as the pure flavonoids.