ABSTRACT

The research work embodied in this thesis entitled "**PHYSICOCHEMICAL CHARACTERIZATION OF LIPIDIC COMPONENTS WITH SPECIAL REFERENCE TO MONOLAYER, BYLAYER AND SOLID LIPID NANOPARTICLE**" is primarily focused on the three different important aspects of application of lipid mixtures. The first part of the thesis is completely focused on the effect of different additives, *viz.*, cholesterol, low density lipoprotein (LDL) and serum protein at the air – solution interface and in the solution (dispersion) phase in the form of vesicles on the functionality and structure of lung surfactant (BLES). The second part is concerned about another vital application of lipids in the form of solid lipid nanoparticles (SLN), in order to develop novel drug delivery systems. Followed by the thesis excluding the brief introduction and current literature review and works delineated herein has been divided into four chapters.

Chapter I describes the influence of, cholesterol, low density lipoprotein (LDL) and serum proteins on bovine lipid extract surfactant (BLES). Different biophysical tools such as Langmuir Blodgett balance, Raman spectroscopy, mass spectroscopy, atomic force microscopy (AFM). While small amount of cholesterol (10 wt %) and LDL did not significantly affect the adsorption and surface tension lowering properties of BLES, however serum lipids, whole serum as well as higher amount of cholesterol and LDL drastically altered the surface properties of BLES films, as well as gel-fluid structures formed in such films observed using atomic force microscopy (AFM). Raman spectroscopic studies revealed that serum proteins, LDL and excess cholesterol had fluidizing effects on BLES bilayers dispersion, monitored from the changes in hydrocarbon vibrational modes during gel-fluid thermal phase transitions. The studies clearly suggest that pathophysiological amounts of serum lipids (and not proteins) significantly alter the molecular arrangement of surfactant in films and bilayers, and can be used to model lung disease.

Chapter II describes the effect of hydrocarbon chain length of nonionic surfactants (Tween 40 and Tween 60) on the physicochemical properties of nanostructured lipid carriers (NLCs). Two local anaesthetics, lidocaine (LIDO) and procaine hydrochloride (PRO.HCl), were incorporated in the NLCs. NLC formulations were prepared using sorbitantristearate (Span 65), soy lecithin (SLC) and stearic acid (SA) in 2:2:1 mole ratio employing the hot homogenization technique. Systems were

characterized by combined dynamic light scattering (DLS), transmission electron microscopy (TEM), differential scanning calorimetry (DSC) and spectroscopic studies. The developed NLCs were considered to have prospects as novel drug carriers for controlled/sustained release to improve the duration of anaesthesia, especially for topical application.

Chapter III describes the impact of saturation/unsaturation in the fatty acyl hydrocarbon chain on the physicochemical properties of nanostructured lipid carriers (NLC) in order to develop anticancer drug (ursolic acid, UA) loaded delivery systems. Mutual miscibility of the components at the air-water interface was assessed from the surface pressure-area isotherms. NLCs were characterized by combined dynamic light scattering, differential scanning calorimetry, TEM, AFM, encapsulation efficiency, pay load, *in vitro* drug release and *in vitro* cytotoxicity studies. All UA loaded formulations exhibited superior anticancer activity compared to free UA against human leukemic cell line K562 and melanoma cell line B16.

Chapter IV explored the orcinol glucoside-loaded nanostructured lipid carrier (NLC) coated with polyethylene glycol (PEG) for oral delivery of orcinol glucoside (OG) to improve the *in vitro* cytotoxicity against GIT cell lines such as Hepatocellular carcinoma (HepG2), hepatocyte-derived carcinoma (Huh-7), colorectal carcinoma (HCT-116) and gastric adenocarcinoma AGS cells. Orcinol glucoside loaded PEG-NLC (OG-PEG-NLC) comprising tristearin, oleic acid and PEG - 25/55 - stearate were prepared by hot homogenization followed by ultrasonication technique. Non PEGylate NLC (OG-NLCs) was also prepared as control. Characteristics of OG-PEG-NLC and OG-NLC, such as particle size, zeta potential, morphology (TEM, SEM, and AFM), entrapment efficiency and drug loading were investigated in detail. The OG-PEG-NLC exhibited superior anticancer activity compared with OG-NLC and OG solution against GIT cancer cell lines. This is first report demonstrating a practical approach for oral delivery of OG for GIT cancer targeting, warranting further *in vivo* cancer study for superior management of GIT cancer.

The thesis then follows comprehensive summary and conclusion followed by the cited references and off-prints of the published journal articles.

PREFACE

The work presented in this thesis is a gleaning discourse on two very important aspects of lipid mixtures/aggregates: (1) functionality of lung surfactant (Bovine Lipid Extract Surfactant or BLES), and (2) Solid Lipid Nanoparticles (SLN) as drug delivery system. The structure of this thesis begins with a detailed introduction involving corresponding literature survey; the reported information on understanding the inhibitory effect of the lung BLES and SLN as improvised drug delivery systems are hereby presented in a generalized manner after careful and exclusive review of the same, which further follows a description on the scope and perspective. Although, many of previous reported studies have dealt with the interactions between lipid monolayer and the disrupting agents, however the exact mechanism of film dysfunctionality, induced by different additives, remains still unclear till date. For decades now, several attempts to develop novel drug delivery systems, having all the advantages such as improved bioavailability, increased therapeutic activities and sustained release have succeeded to a very little extent. Systematic investigations on the dysfunctionality of lung surfactant by different additives such as cholesterol, LDL and serum protein are not so common in the literature. Different kinds of monoglycerides, diglycerides, triglycerides, waxes, phospholipids and fatty acids have extensively been used to develop NLCs. However, the use of sorbitantristearate (Span 65) as one of the lipidic components in preparing NLCs have not been studied extensively. In addition, the effect of stabilizers on the solution phase behavior and thermal properties of NLC have not been meticulously investigated. Lidocaine (LIDO) and procaine hydrochloride (PRO.HCl) are frequently used as local anaesthetics for topical application. Both the drug were loaded in NLCs, in order to prolong the anaesthetic effect and reduce dose frequency as well as the skin irritation caused by the high dose of anaesthetics, such formulations are considered worthy to be investigated. The impact of saturation and unsaturation on the physicochemical properties of nanostructured lipid carriers (NLCs) of fatty acyl hydrocarbon chain was investigated to develop novel delivery systems loaded with the anticancer drug, ursolic acid (UA). The aim of this study was to evaluate and compare the saturated and unsaturated lipids comprising NLCs to determine if differences in composition could alter the performance of the systems. Orcinol glucoside (OG) incorporated into the conventional nanostructured lipid carriers (OG-NLC) and PEG-coated orcinol glucoside-loaded nanostructured lipid carriers (OG-PEG-NLC), with the purpose of targeting gastrointestinal tract cancer with

enhanced anticancer activity for its oral delivery were explored. Finally, findings based on different experimental observations were summarized which followed some concluding remarks. The thesis then follows the off-prints of the published journal articles.