Abstract
Across the globe and in India, cancer drug delivery has focused using novel carrier system due to their effective encapsulation and release controlled property with the minimum side-effect. The flexibility of modification in liposomes makes them a useful drug delivery system in various fields like protein /drug delivery, antiviral therapy, tumor therapy, gene delivery, vaccine delivery, cosmetics, dermatology and others. However, there are some stability issues and incomplete drug releases at target site are attached with liposome. In order to achieve maximum or appropriate therapeutic response, the drug in required quantity should be available to the target cells in the disease like cancer. Therefore, considering the example of cancer, delivery of drug with liposome to the site should be released in therapeutic concentration. For this purpose, internal stimuli can be used effectively to enhance the release of drug from liposome.
Artesunate (ART) and curcumin (CUR) are proved natural herbal lead chemotherapeutics used as Chinese and Indian food respectively. But due to its low absorption and the poor bioavailability limits its clinical efficacy. The object of the present work was to investigate the cytotoxic potential of these herbal lead molecules in the co-encapsulated liposome formulation. It is hypothesized that liposomes co-encapsulating Artesunate and Curcumin in a synergistic ratio would be an appropriate drug delivery system to treat breast cancer effectively.
First, we docked the Artesunate and Curcumin with two pro-carcinogenic ligands, to check the binding energy and interaction between them. We confirmed the cell viability of free drug combination before formulation and determined the IC\textsubscript{50} values on two cell lines (MCF-7 and MDA-MB-231). IC\textsubscript{50} value for ART and CUR was found to be 0.65 ± 0.22μg/ml and 3.09 ± 0.84 μg/ml respectively.
Artesunate and Curcumin co-encapsulated conventional and PEGylated liposomes were formulated and characterized. Lipid and cholesterol molar ratio were optimized on the basis of particle size and entrapment efficiency (% EE). Size of formulated liposome was around 119-125 nm and with maximum percentage entrapment efficiency of 85-88 %. Co-encapsulation in nanometer size is beneficial for uptake and stability of a drug. Further study included \textit{in vitro} release from liposomes. PEGylated liposomes showed the remarkable release of ART and CUR as compared to the conventional liposomes. TUNEL assay and Western blot assay was performed to find approx 50% apoptosis of prepared formulations and PEGylated liposome showed
improved mRNA expression for TRAIL and DR5 compared to conventional liposomes. The formulation was tested for storage stability and found stable by protected from light at 4°C. These results were supported by *in vivo* study using an animal model. Healthy Swiss albino female mice were used and MCF-7 cell line was used to induce breast cancer. Animals were treated with PEGylated and conventional liposomes to find their efficacy against induced cancer. Different antioxidant stress parameters were estimated and significant (p<0.05) improvement was observed with PEGylated liposomes treated animals. The research concluded ART plus CUR co-encapsulated and PEGylated liposome had a synergistic cytotoxic, and apoptotic effect on MCF-7 cell lines by *in-vitro* and *in-vivo* studies. The results from these studies warrant further testing of this liposome formulation in clinical settings.