**Potential Use of Cholecalciferol Polyethylene Glycol Succinate as a Novel Pharmaceutical Additive**

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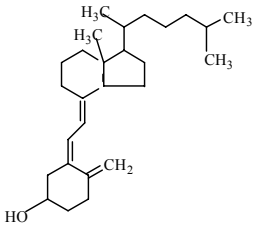
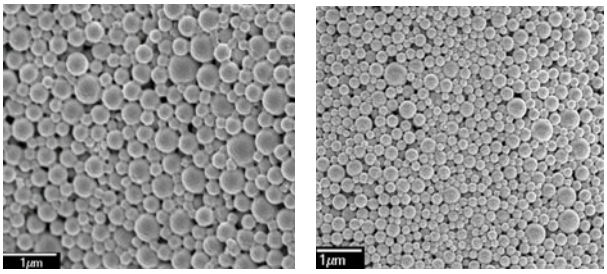
**Statement of Purpose:** D-α-tocopheryl polyethylene glycol succinate (TPGS) has been utilized in numerous drug delivery formulations in recent years [1]. Due to its amphiphilic structure, it can be used as emulsifier and vehicle for lipid-based drug delivery formulations. It is also an effective P-glycoprotein (P-gp) inhibitor. However, TPGS represents only one of the surfactants in the class of "Vitamin-PEG" conjugated surfactants. In order to determine the effectiveness of Vitamin-PEG conjugated surfactants as additives in formulating drug delivery carriers, we hypothesized that any hydrophobic vitamins combined with PEG may achieve similar effectiveness that TPGS has brought in drug delivery formulation. Vitamin D is of much interest in this study because of its cancer prevention functions [2]. A growing number of studies have suggested that Vitamin D can be effective in certain types of cancer chemoprevention and treatment [3]. To design a new adjuvant or additive, a conjugate made of Vitamin D (cholecalciferol) and PEG-cholecalciferol polyethylene glycol succinate (CPGS) was synthesized as an additive for the formulation of drug loaded PLGA nanoparticles, and the efficacy of the resultant drug carrier was tested in vitro.




Structure of vitamin D

**Methods:** CPGS was synthesized by conjugating vitamin D succinate with poly(ethylene glycol) methyl ether (single end capping). The resultant product was characterized with H-NMR, FTIR and HPLC. Doxorubicin (DOX) loaded PLGA nanoparticles with CPGS were then prepared by dialysis method and the morphology of the nanoparticles was visualized by SEM. Physical properties of the drug loaded nanoparticles including in vitro drug release were characterized. The cytotoxicity effect on Caco-2 cells was then studied. The accumulation of CPGS added PLGA nanoparticles in cells was also investigated using rhodamine and observed via confocal microscopy.

**Results/Discussion:**
Characterization of CPGS – Both H-NMR and FTIR show the successful conjugation of cholecalciferol and PEG. HPLC results confirm the purity of CPGS as only a single peak was observed.
Characterization of DOX loaded nanoparticles – The morphology of the nanoparticles is shown in Figure 1. It was found that with the presence of CPGS, the size of the nanoparticles reduces from approximately 300 to 200 nm in diameter.




(a) (b)

Figure 1. SEM images of (a) PLGA nanoparticles, (b) PLGA nanoparticles with 5% CPGS additives

Cytotoxicity study – Figure 2 shows that the presence of PLGA nanoparticles does not affect the cell viability when compared with free DOX administration. However, with the use of CPGS as additive, the cytotoxicity increases substantially, especially at the higher DOX concentration (> 1 μM DOX). The effect of CPGS in increasing cytotoxicity is comparable to that of TPGS (1, 10, 20 μM DOX) demonstrating that there is a good potential that CPGS can be used as an alternative in drug formulation.

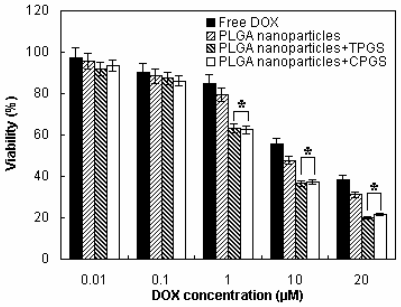



Figure 2: The cytotoxicity of DOX formulations in Caco-2 cells. Bars marked with * are significantly different from free DOX and DOX loaded PLGA nanoparticles at the same DOX concentration ($p<0.05$).

**Conclusions:** A new vitamin D-PEG conjugate - cholecalciferol polyethylene glycol succinate (CPGS) was synthesized as a new drug additive. The structure of CPGS suggests that it will have similar properties to TPGS. From our current study, CPGS could be used as a surfactant to prepare drug-loaded nanoparticles.

**References:**

1. Somavarapu S., Int. J. Pharm., 2005. 298: p. 344.
2. Donaldson M.S., Nutr. J., 2004. 3: p.19.
3. Weitsman G.E., Free Radic. Biol. Med., 2005. 39: p. 266.