EFFECT OF THE COMPOSITION OF LIPOSOMES ON DEXAMETHASONE ENCAPSULATION AND ITS KINETICS OF RELEASE

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1. INTRODUCTION

Liposomes are tiny vesicles with a lipid bilayer primarily made up of cholesterol (Chol) and phospholipids. Hydrophobic active ingredients (API) are thought to be encapsulated in this lipidic membrane, while hydrophilic APIs would be encapsulated in the aqueous core of the liposome [1]. As smart nanocarriers, liposomes can selectively address the API to its site of action and regulate its release, making them a good option to reach difficult targets such as inner ear hair cells in the treatment of sensorineural hearing loss [2]. Developing a sustained release (SR) formulation would maintain a steady concentration of API in the cochlea while reducing the frequency of administrations through transtympanic injection. According to literature, release kinetics may be influenced by the lipid composition of the liposome. For instance, the incorporation of saturated lipids in the formulation, such as DPPC or DSPC, along with an increase in the percentage of Chol, are known to stabilize the lipid bilayer and slow down API release [3]. The aim of this work is therefore to investigate the influence of various liposome compositions on the release kinetics and encapsulation efficiency (EE) of dexamethasone (Dexa) or dexamethasone sodium phosphate ($DexaPO_4$) used as APIs. These will be compared to identify the best therapeutic option.

2. MATERIALS & METHODS

a. Liposomes composition and production

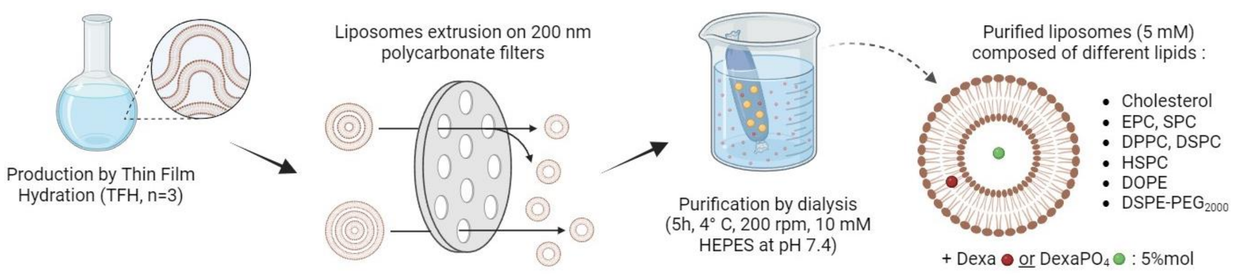

Figure 1. Production, extrusion and purification of liposomes composed of different types of lipids [4].

b. Release test

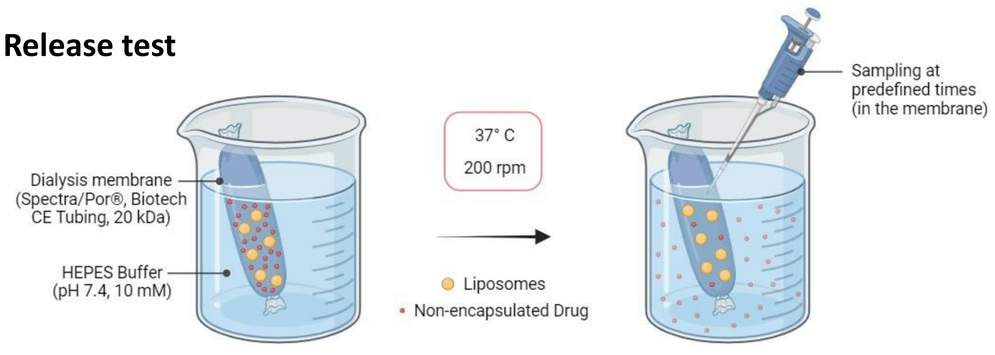

Figure 2. Release test based on a dialysis method [4].

3. RESULTS & DISCUSSION

The highest results in terms of EE were obtained with 10% Chol for Dexa (Figure 3). Chol will thus be used at a low percentage due to its structural analogy with Dexa and their competitive effect for space in the liposome lipid bilayer. Among the various lipids tested, EPC exhibited significantly higher EE for Dexa-liposomes, while DSPC showed the best EE for $DexaPO_4$-liposomes. The longest release kinetics for Dexa-liposomes were obtained with EPC as the main lipid and 10% Chol (Figure 4). However, these components do not appear to substantially improve the SR, since the API is released over the same period of 3 hours for Dexa and 24 hours for $DexaPO_4$ (Figure 5). Similarly, addition of DSPE-PEG_{2000} or presence of DOPE in Dexa-liposomes did not greatly affect SR (Figure 4). In terms of kinetics, the use of $DexaPO_4$ thus appears to be more interesting, since its release occurs over 24 hours. However, in terms of EE, results are significantly higher for encapsulated Dexa than for encapsulated $DexaPO_4$. The most promising approach therefore seems to be the use of Dexa, as it would allow reaching therapeutic concentrations more easily than with $DexaPO_4$. New strategies will then be required to sustain the release of Dexa (Figure 7). The impact on HEI-OC1 cell viability was then assessed for a few formulations, HEI-OC1 being a model for inner ear hair cells (Figure 6). Results show that EPC-liposomes (F1, F2) significantly reduce viability when applied at a minimal concentration of 0.5 mM, while liposomes containing saturated lipids (DSPC, DPPC) do not show any cytotoxic effect at the concentrations studied.

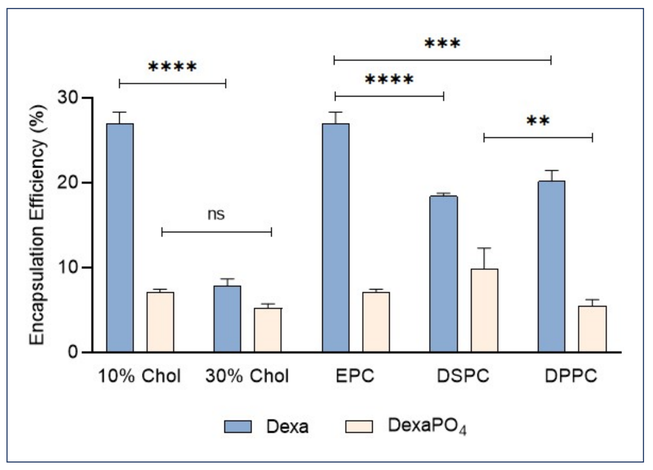

Figure 3. Comparison of Dexa and $DexaPO_4$ encapsulation efficiencies.

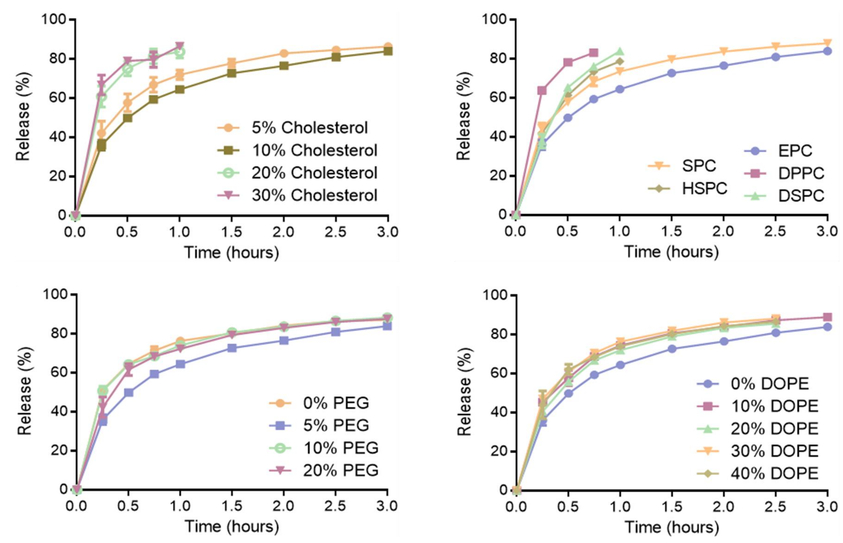

Figure 4. Impact of lipid composition on the release of **Dexa** encapsulated in liposomes in function of time.

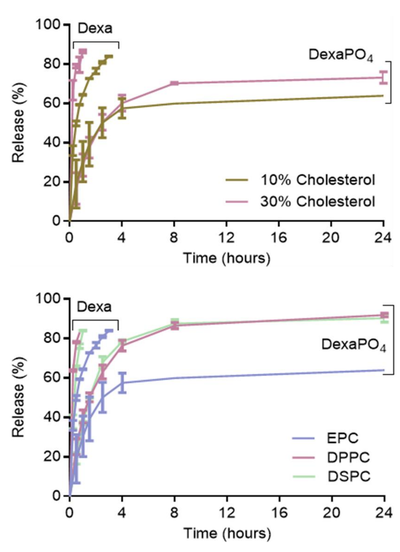

Figure 5. Comparison of Dexa- and $DexaPO_4$-liposomes SR.

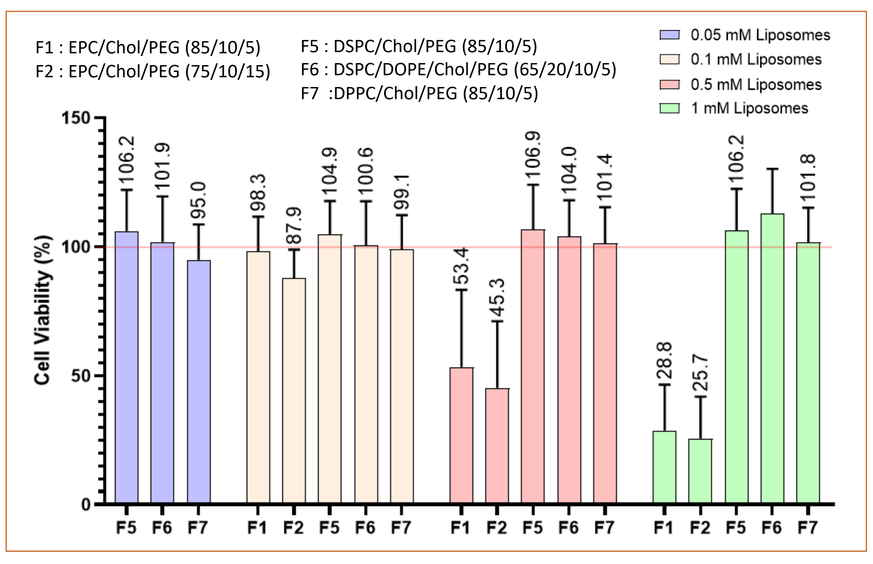

Figure 6. Comparison of the viability of HEI-OC1 cells exposed for 24h to different liposome compositions (drug free).

4. CONCLUSION & PERSPECTIVES

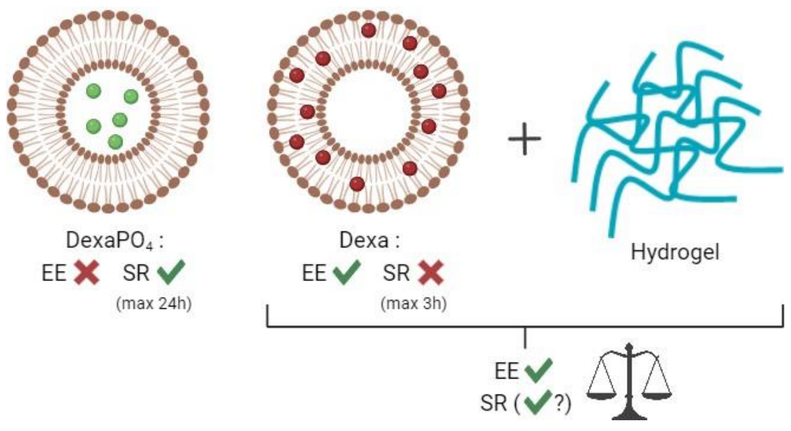

Figure 7. Dexa-liposomes will be dispersed in a hydrogel to improve the SR effect [4].

Changes in liposome composition

The lipids considered for liposome formulation optimization showed no improvement in the release kinetics of APIs. Some components, though, seemed useful to significantly improve EE (10% Chol, EPC, etc.). Some formulations have been tested *in vitro* for cytotoxicity on HEI-OC1 cells. DSPC- and DPPC-liposomes appear to be more biocompatible, but the impact of all these formulations has yet to be assessed on hSAE cells, which will be modeling the round window membrane of the inner ear. Most biocompatible lipids will be considered for the final liposome formulation.

5. REFERENCES

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[4] Created with BioRender.

6. ACKNOWLEDGMENTS

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