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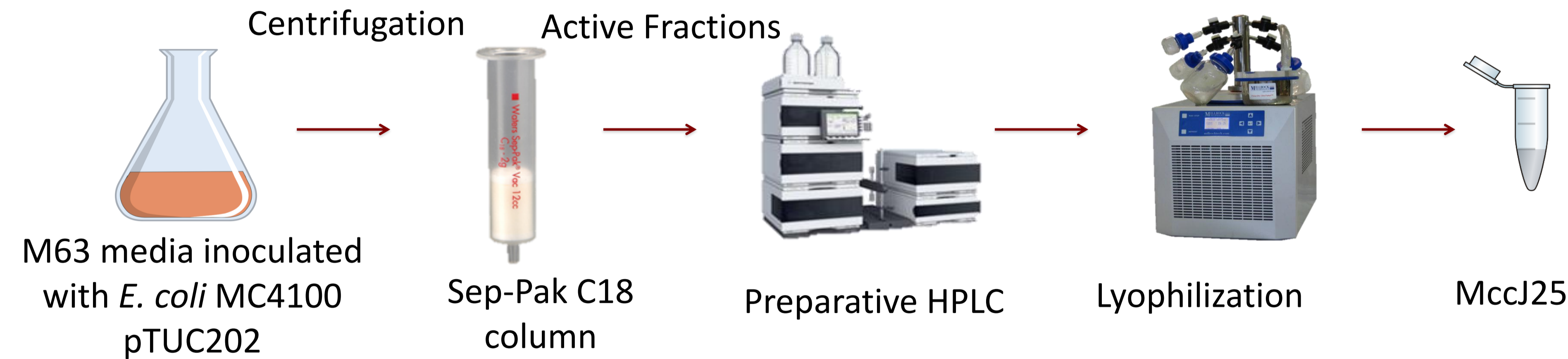
Introduction

Encapsulation is one of many strategies that can be implemented to achieve controlled release of the encapsulated active ingredient and to maintain antimicrobial activity and stability of the active ingredients in complex systems. Liposomes have the advantage of being able to trap large quantities of hydrophilic or hydrophobic substances. However, applications are limited because of their low physical and digestive stabilities when passing through the gastrointestinal tract. The bile salts are harmful to them because they accelerate the hydrolysis of the lipid bilayer by increasing the fluidity of the membrane and the low pH, for example gastric conditions, lead to changes in the surface charge of the liposomes. Nevertheless, the stability of the liposomes to gastrointestinal conditions can be increased by using coating materials.

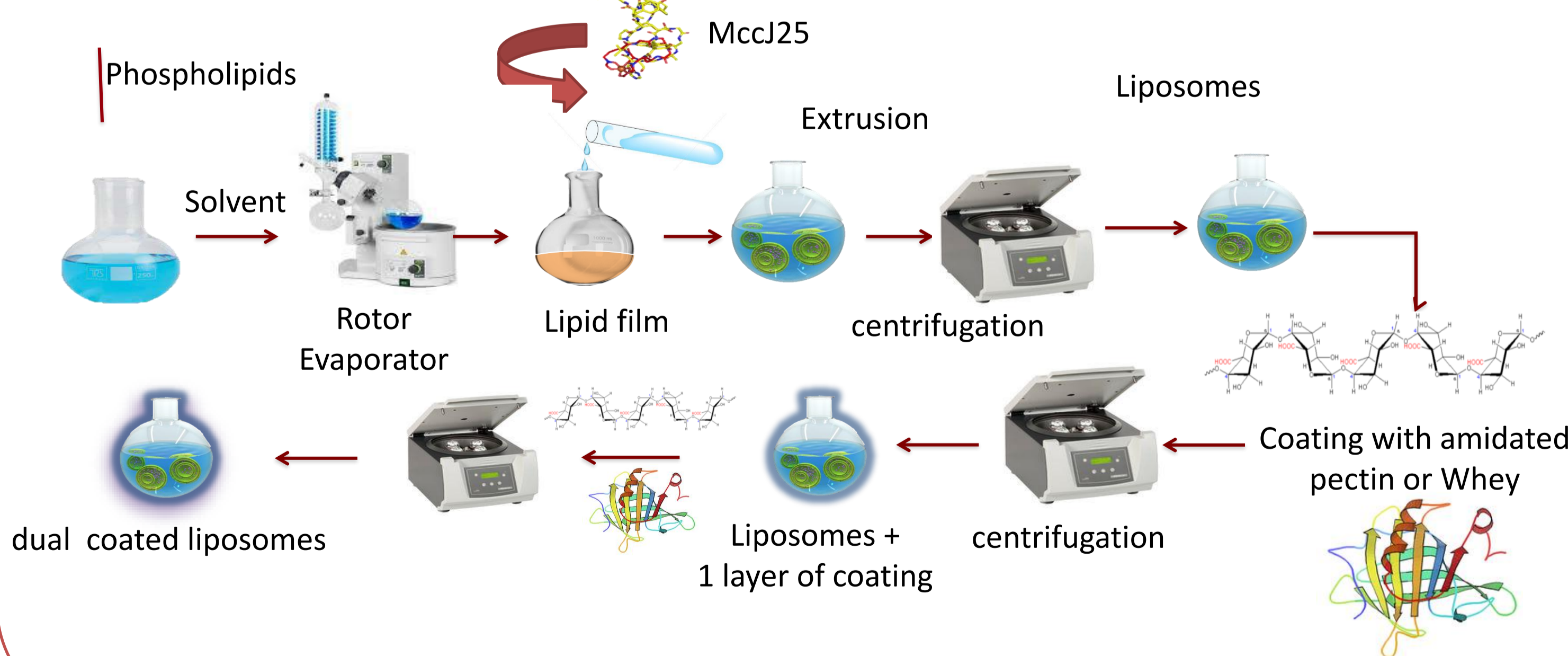
The objective of this study is to develop food-grade liposome formulations that allow for a sufficient improvement in the stability and controlled release of bacteriocins during their oral assimilation. The formulas are elaborated from anionic phospholipids as a negative liposome model and cationic as a positive liposome model using the method of hydrating a phospholipid film. Thereafter, the liposomes are coated with a network of biopolymers (pectin) and / or whey proteins (WPI) in order to further improve the stability and gradual release of the developed liposomes. The muco-adhesive properties of the liposomes can be improved by coating with pectin^{3,4}. Recently, it has been shown that the coating of liposomes by WPI makes it possible to reduce their semipermeability and the protections of osmotic forces. Thus, WPI and pectin prove to be two very interesting materials in order to improve the encapsulation and release of antimicrobial peptides.

Methods

1) Large Scale preparation of microcin J25 (mccJ25):



2. Preparation of liposomes by lipid film hydration method:



3. Analysis:

- Zeta potential
- Particle size distribution
- FTIR
- TEM
- Encapsulation Efficiency %
- In-vitro release Model

Results & Discussion

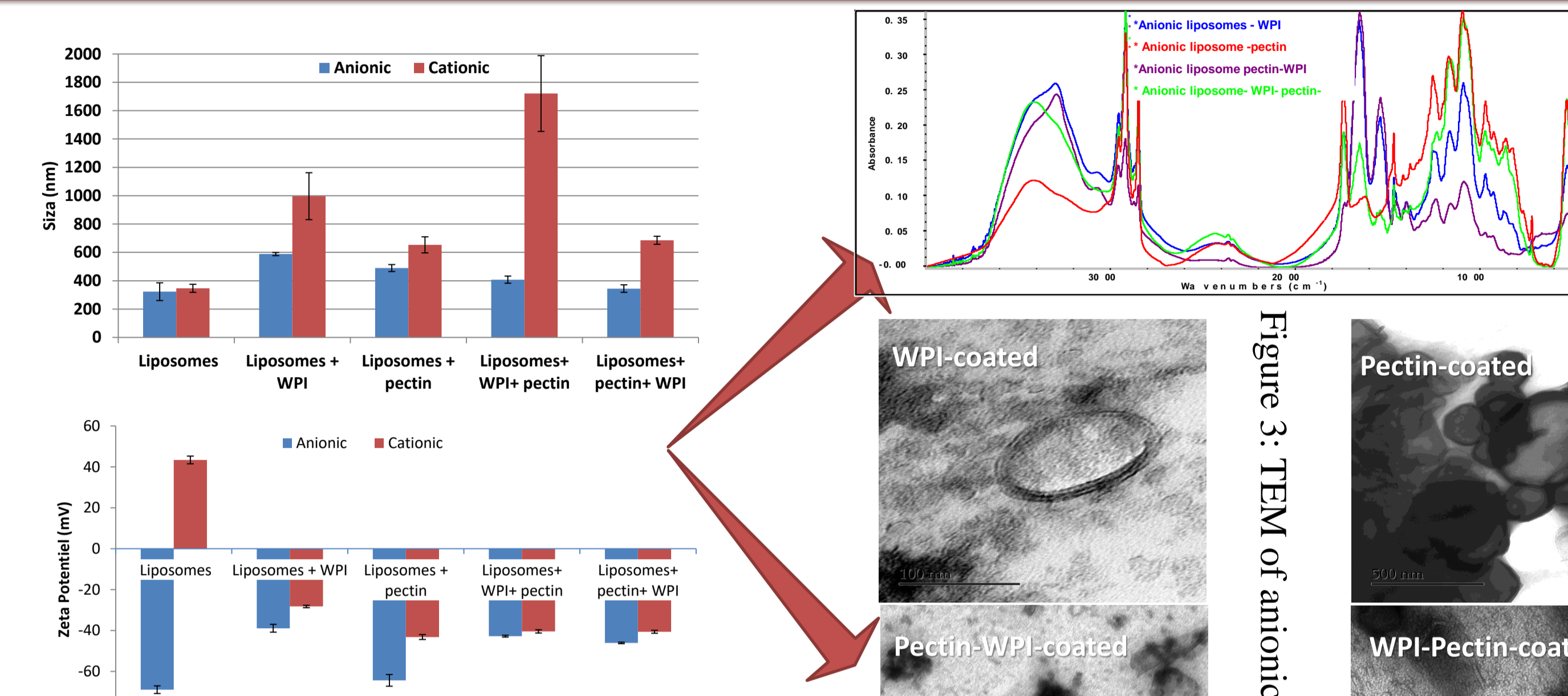


Figure 1: Particle size distributions (a) and of the zeta potential (b) of positive and negative liposomes coated with WPI and/or pectin.

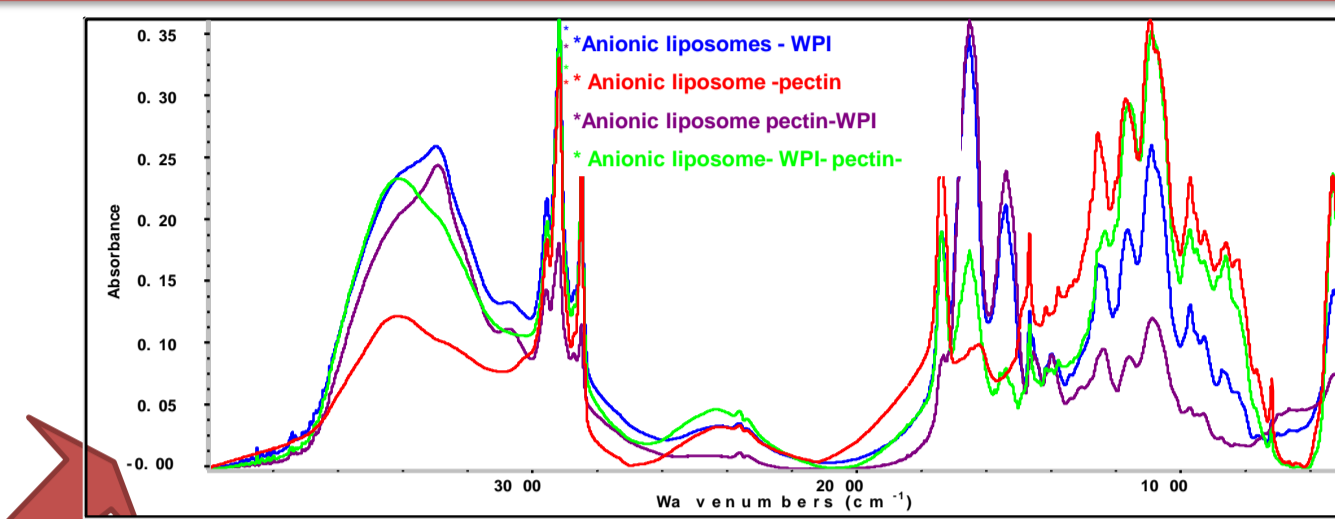


Figure 2: FT-IR spectra of anionic liposomal formulas.

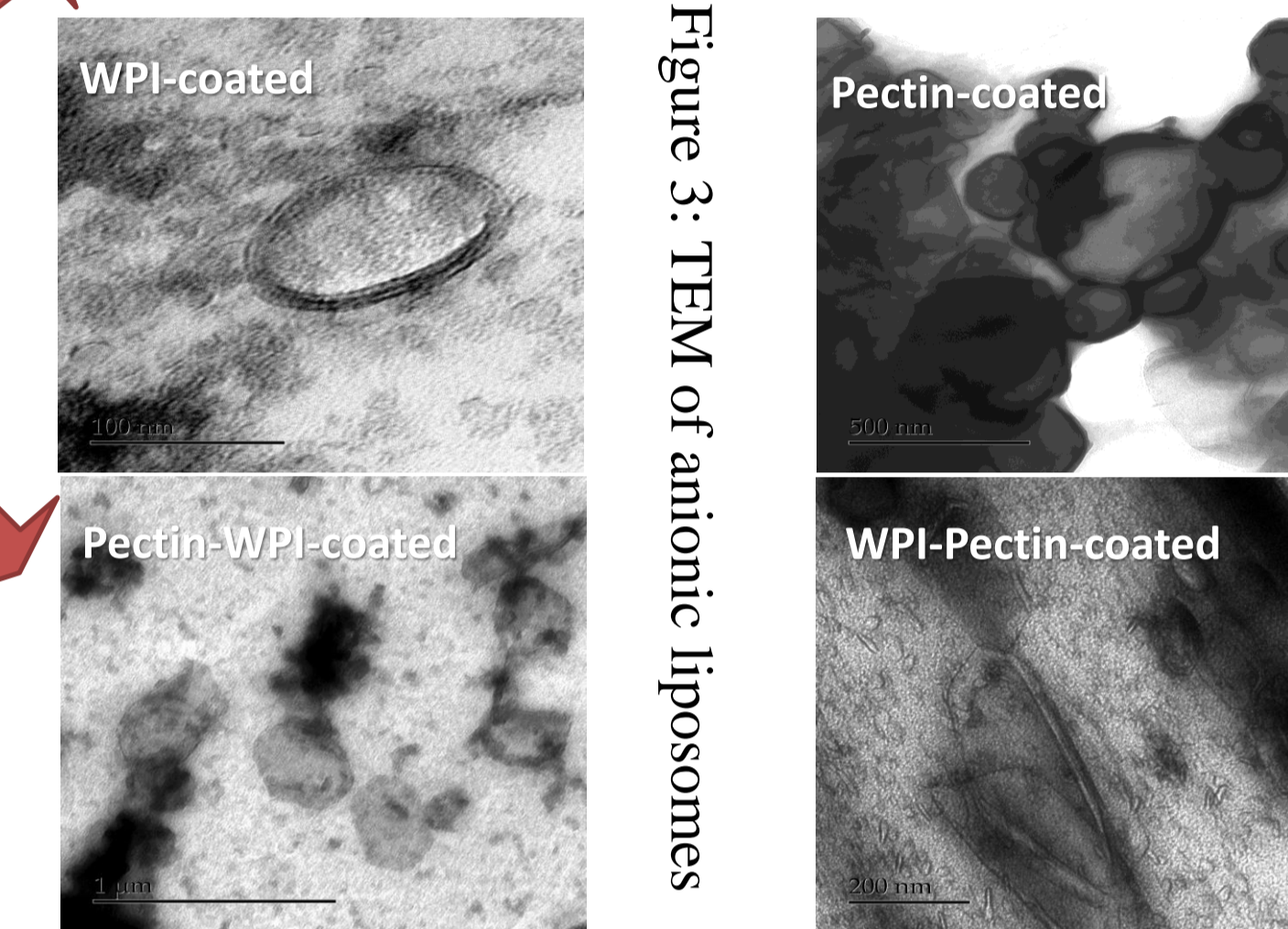


Figure 3: TEM of anionic liposomes

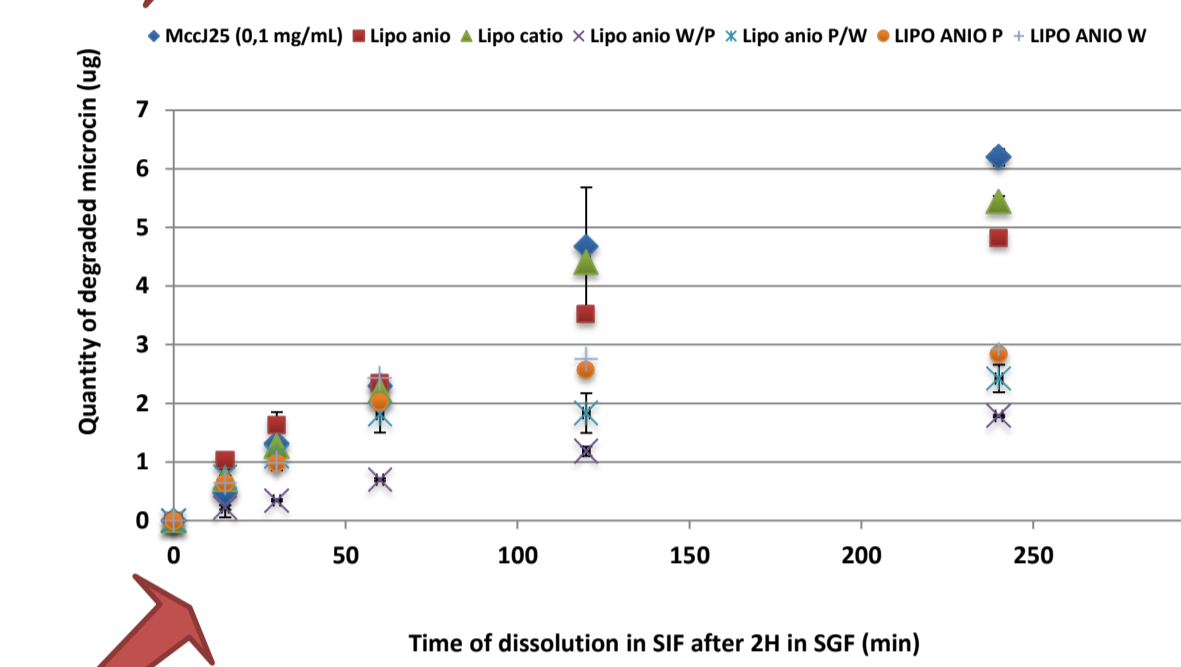


Figure 4: Dissolution of encapsulated MccJ25 in simulated gastrointestinal conditions.

The production of MccJ25 was monitored by determining the protein concentration and bactericidal activity. Size and zeta potential analysis showed an increase in the size with the coating layers and a change in the zeta potential close to the value of the used coating matrix. Anionic liposomes coating with whey could be explained by electrostatic interactions between positive charge of the whey and the negative charge of the liposomes. While, Efficient coating of anionic liposomes with pectin could be explained by the interactions between amide groups of pectin and liposomes. Cationic liposomes were successfully coated with whey at pH above IEP and pectin with electrostatic interactions. Liposomes coated with two layers of whey and pectin improved the stability of liposomes.

Conclusion

In this study the MccJ25, an antimicrobial peptide, was encapsulated in positively or negatively charged liposomes that subsequently coated with pectin and/or WPI. Coating process was optimized to improve the EE% and the protection of microcin against gastrointestinal digestion. The dual coating liposomes provided additional protection to the microcin during simulated gastrointestinal digestion.

References

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