université PARIS-SACLAY
FACULTÉ DE PHARMACIE

Graduate School: Health and Drug Research
Master 2 Pharmaceutical Technology and Biopharmacy

Company/laboratory/public institution: Pasut's Lab, University of Padova

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Position:

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Internship period: 20 January - 18 July 2025

Title of the project: Preparation and characterization of immunoliposomes for cancer therapy

Liposomes play a pivotal role in drug delivery due to their biocompatibility and versatility as carriers. A major advancement in liposome technology was the development of stealth liposomes, which are modified with hydrophilic polyethylene glycol (PEG) molecules on their surface. This innovation led to significant improvements in the pharmacokinetic profile compared to unmodified ("naked") liposomes and has been widely adopted in clinical practice for many years, proving their efficacy and safety compared to free drug therapies.

However, despite these benefits, stealth liposomes lack the ability to specifically target diseased tissues, resulting in non-specific toxicity to healthy tissues. This remains a critical challenge, particularly in cancer therapy. The development of effective targeted liposomes has encountered numerous obstacles, including clinical trial failures. Active targeting, which involves guiding liposomes to specific cells or tissues, continues to be a major hurdle.

Nonetheless, various studies have focused on creating cancer cell-selective liposomes by functionalizing their surface with antibodies or antibody fragments, resulting in the formation of "immunoliposomes." In this context, our research group has developed novel Super Stealth Immunoliposomes (SSIL), incorporating a polyethylene glycol (PEG)-bi-phospholipid derivative as a shielding polymer on the liposome surface. By using a polymer with two phospholipid units per chain, we have improved the stability of the polymer-liposome interaction, thereby preventing premature PEG detachment during in vivo circulation.

Moreover, a fraction of the PEG-bi-phospholipid derivative can be modified at the polymer's terminal end to facilitate the coupling of fragment antigen-binding (Fab') regions from monoclonal antibodies. This produces Fab'-PEG-bi-phospholipids, which are stably anchored to the liposome surface and ensure proper outward orientation of the target antigen. The effectiveness of these targeted immunoliposomes in delivering anticancer drugs has been demonstrated using trastuzumab-derived Fab' fragments [1].

Objectives of the internship:

1) Acquire expertise in the preparation and characterization of liposomes;

2) Acquire expertise in modification of proteins by generating Fab' moieties and conjugating them to polymers;
3) Acquire expertise in cell testing with drug-loaded liposomes.

The student will work on a project focused on the development of drug-loaded immunoliposomes. Initial experiments will explore various methods for preparing conventional liposomes, followed by their thorough characterization. Next, a specific monoclonal antibody will be modified to generate the corresponding Fab' fragment. PEG-bi-phospholipids will be synthesized in the lab and subsequently coupled with the Fab' fragment. The student will then optimize the procedures for immunoliposome preparation and drug loading.

The resulting immunoliposomes will be tested in vitro against different cell lines that overexpress the target antigen. The in vitro evaluation will focus on characterizing the activity of the liposomes and their internalization within the cells.

References:

1. Canato E, Grigoletto A, Zanotto I, Tedeschini T, Campara B, Quaglio G, Toffoli G, Mandracchia D, Dinarello A, Tiso N, Argenton F, Sayaf K, Guido M, Gabbia D, De Martin S, Pasut G. Anti-HER2 Super Stealth Immunoliposomes for Targeted-Chemotherapy. Adv Healthc Mater. 2023 Nov;12(29):e2301650. doi: 10.1002/adhm.202301650