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PHOTOTHERAPY AGAINST CANCER AND EFFECTS OF GLYCATED CHITOSAN ON SOMATIC CELLS

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Abstract

Immunoadjuvant is a molecule or solution that enhances the immune response to a stimuli or vaccine. N-dihydrogalactochitosan (glycated chitosan, GC) is an immunoadjuvant used in combination with photothermal therapy to treat late-stage, metastatic cancers. After tumor cells release damage-associated molecular patterns (DAMPs) from laser irradiation, GC is uptaken by antigen-presenting cells (APCs) at the treatment site. These APCs can differentiate into their inflammatory state and then activate the adaptive immune system. This chain reaction will then eliminate the residual tumor, as well as metastatic cancer cells. GC can also be combined with antigens in an intra-nasal vaccine formulation against respiratory infections. The use of GC in the formulation promotes the number of recruited T cells and IgA at the delivery site. The reaction of immune cells to GC is well studied but its direct effects on cancer and epithelial cells have not been fully explored. In this study, we investigated the potential effect of GC on nasal epithelial cells and on the cancer cells in the anti-viral and anti-tumor contexts. We used viability assays, confocal imaging, and flow cytometry to determine GC's toxicity on different cell lines, epithelial cells' intake of GC, and activations induced by GC. Understanding GC's interaction with non-immune cells can advance our knowledge of GC's mechanism and potentially find novel applications for cancer therapies and vaccinations. Chen lab combined the use of GC with photothermal therapy. However, photothermal is not the only form of therapy using laser power. Photodynamic therapy is another potential approach. It generates reactive oxygen species (ROS) using the energy of the laser accumulated by a photosensitizer (PS). These ROS then provoke the death of the cancer cells. Photochemical internalization combined encapsulated cancer toxic molecules with the use of a PS to deliver these toxic compounds in the cytosol of the cancer cells. We will also review the current treatment modalities used for cancer patients by clinicians and compare their efficacy with photodynamic therapy, photochemical internalization and photothermal therapy.

Keywords: N-dihydrogalactochitosan, laser therapy, intranasal vaccine, cancer treatment

Chapter 1. Introduction

1.1. N-dihydrogalactochitosan – a potent immunostimulant/adjuvant

N-dihydrogalactochitosan, also called glycated chitosan (GC), is a biopolymer composed of chitosan with galactose attached to the amine groups (**Figure 1**)[1]. Studies on GC demonstrated its immune enhancing properties, similar but stronger than chitosan's. When Antigen Presenting Cells (APC) phagocytose GC and antigens, their type I IFN pathway of activation is stimulated.

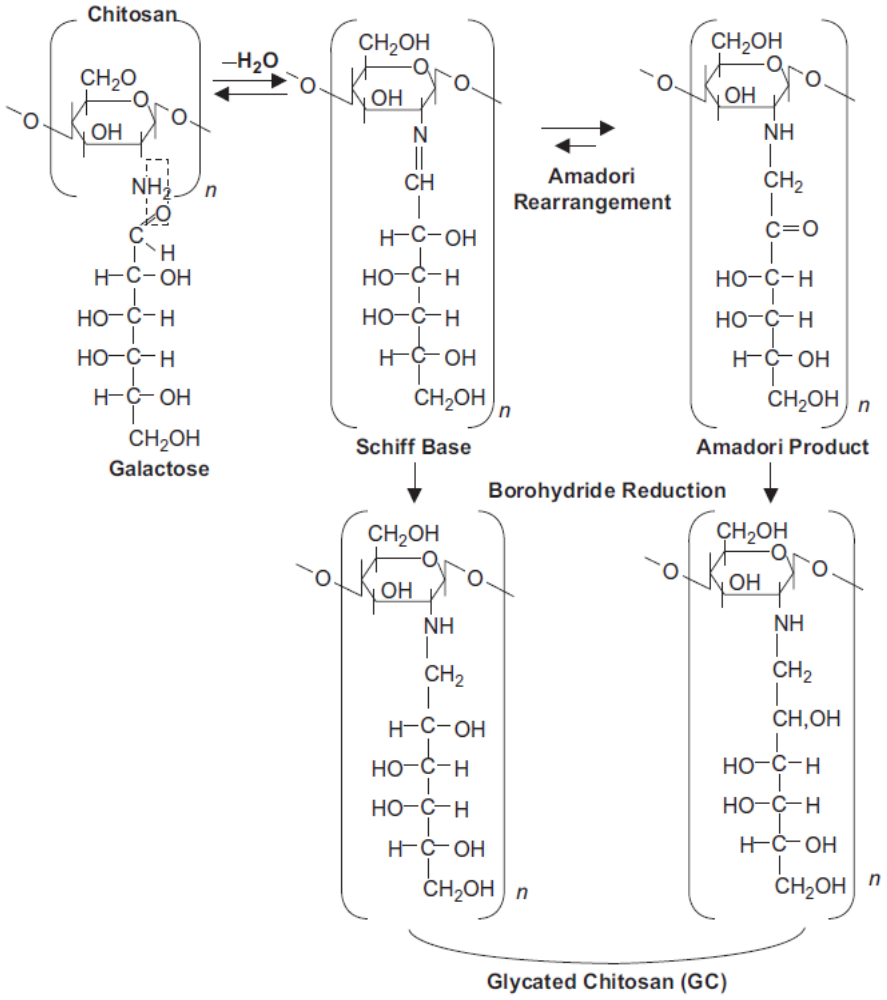


Figure 1 Structure of the Glycated Chitosan[1].

More precisely, GC provokes inflammatory cell death through the activation of the STING pathway inside dendritic cells (DC).[2] Due to this immune-adjuratory effects, GC has been combined with photothermal therapy to treat metastatic cancer. The heat produced by the laser provokes the death of tumor cells and the release of damaged associated molecular patterns (DAMPs), which serve as antigen in cancer therapy. APCs and Th1 immune cells invade and completely eliminate the tumor and the potential metastasis[3]. Although a few studies have examined the effects of GC on immune cells in a cancer treatment context, its direct effect on the

tumor remains unknown. Therefore, we aim to investigate the primary functions GC may have when in contact with cancer cells.

New cancer treatments based on the use of a laser (phototherapy) are being investigated in clinical trials in various countries[4–7] but this number is still relatively low compared to more traditional treatment protocols such as surgery, chemotherapy, and/or radiotherapy. These conventional treatments demonstrate limitations, as cancer-related mortality rates remain high worldwide[8,9]. As a result, a review of the success rate of the current treatment —assessed through patient life expectancy, cure rates, and time to cure— will be compared with the preliminary results from clinical trials using one of those three types of phototherapy: photodynamic therapy, photothermal therapy and photochemical internalization. We will highlight the potential of laser treatments for different types of cancer (including the organs affected and their stage of development) and compare these results to those from current clinical treatments.

GC is also investigated as a potential adjuvant in intranasal vaccine formulations[10]. *In situ* mucosal vaccines have the potential to be more efficient than intradermal vaccine because they induce a localized immune response, mimicking the viral infection route, and activating mucosal-specific adaptive immunity (e.g., IgA producing B cells). Additionally, mucosal vaccines also have the advantage of not requiring specific administration training or needles[11]. While the effect of GC on the immune system is well-studied in the context of intranasal vaccination, its impact on the mucosa itself remains unknown. Therefore, preliminary investigations were conducted to examine the potential adjutancy of GC on the nasal epithelial cells of the mucosa. In the following section of this introduction, we provide more detailed definitions and current knowledge relevant to these investigations.

1.2. Phototherapy and cancer treatment

According to the American Cancer Society, the second most common cause of death in the United States is cancer with a 69% overall survival rate between 2013 and 2019: specifically, 99% for thyroid cancer but only 22% survival rate for esophageal cancer. Thanks to recent advances in immunotherapy, the survival rate increased from 18% in 2009 to 38% in 2015 in cases of metastatic melanoma. The overall survival also has improved over the last few decades due to new early diagnostic methods, but there is still thousands of people who died from cancer every year around the globe.[8]

Many treatments have been developed to treat cancer, such as chemotherapy, radiotherapy, and immunotherapy. However, the success rate of these treatments is not 100%, especially concerning aggressive, high risk types of cancers and advanced stages[12,13]. Also, these treatments provoke a lot of sides effects in patient such as immune-mediated damage to healthy tissue for immunotherapy[14], damage to the DNA or nephrotoxicity for chemotherapy[15], and pain or nausea due to radiotherapy or chemotherapy[16]. Researchers and physicians continue to explore different combinations of treatments and develop new therapies to fight cancer.

In this context, phototherapy has emerged as a new treatment strategy in synergy with existing treatments[17,18]. Phototherapy has the potential to be a targeted and non-invasive treatment [18], and

in the past decades, different types of therapy using light have been developed.

In the following section, we will define and review the current advancements in research on phototherapy, but also the advancement in their use in clinical trials. We will also review the current treatment protocol used by clinicians to help extend the survival of cancer patients and compare those protocols with the results obtained in clinical trials with phototherapy. These comparisons will also help us evaluate the potential of phototherapy as a cancer treatment and the limitations encountered that still require optimization in preclinical study.

1.3. Clinical Trials

Looking into the current research conducted in clinical trials is a good way to get a general view of the advancement made in a field. But before looking for clinical trials, we need to understand how they are designed, the criteria to start and by validated and the existing specific cases applicable to our subject.

The World Health Organization (WHO) defines clinical trials as "a type of research that studies new tests and treatments and evaluates their effects on human health outcomes. People volunteer to take part in clinical trials to test medical interventions including, drugs, cells and other biological products, surgical procedures, radiological procedures, devices, behavioral treatments, and preventive care."[19] A treatment (i.e. drug, biological product, procedure, device, any type of care) tested in clinical trials is first tested for safety and effectiveness in preclinical trials (studied using animals).

In the United States of America (USA), if the results are considered favorable by the Food and Drug Administration (FDA), it approves the beginning of tests in humans.[20] The majority of clinical trials in the USA are also approved and monitored by an Institutional Review Board, which ensures that all risks are reduced and outweighed by the benefits the treatment can provide.[21] In the European Union (EU), each country has its own regulation and committee reviewing all the clinical trial application. However, since January 2022, every new clinical trial EU-based (i.e. performed in one or several EU countries) needs the approval of the Clinical Trials Regulation.[22]

A clinical trial is composed of four phases, each phase has its own objectives, group size range and requires the validation of the previous phase to be conducted. The objective of phase I is to test a new treatment on a small sized group of people (around a dozen people) to ensure the safe dosage range and the potential side effects of the treatment. Phase II evaluates the short-term therapeutic efficacy and potential adverse events of the different dosages in a larger group of people (between 20 and 150 people). Phase III confirms the results of the treatment on a large group (hundreds of participants) and a randomized placebo, or similar standard treatment (used by practicians), will be included in the study. The purpose is to generate statistically significant data on the treatment outcomes and determine the most beneficial dosage (better outcome with fewer side effects). At the end of Phase III, the FDA (European Medicines Agency, EMA, in EU) will deliver a license or not for the public usage of the treatment in cases similar to the one evaluated during the clinical trial. Phase IV designated the collection of data within the population when the treatment is prescribed by clinicians. Long-term effects of the drug are evaluated.[23,24]

Some Treatment are not evaluated in all four phases. Special cases of treatments, such as those for rare diseases, a disease without current effective available treatment or treating a worldwide pandemic, can go through the clinical trials process faster by four approaches: Priority Review, Breakthrough Therapy, Accelerated Approval and Fast Track. The Priority Review goal is to apply the treatment within 6 months. A priority evaluation and overall attention from the FDA will be direct to this treatment. It can be applied to treatment proving "significant improvements in the safety or effectiveness of the treatment, diagnosis, or prevention of serious conditions when compared to standard applications. The Accelerated Approval can be applied to treatment "for serious conditions that filled an unmet medical need to be approved based on a surrogate endpoint." A surrogate endpoint is a measure that can predict a clinical benefit, but not the measure of the clinical benefit itself. This type of early approvement of drug usage can be applied to cancer treatments capable of extending cancer patients' survival. The Breakthrough Therapy applies to drugs intended to treat serious conditions and there is preclinical evidence of the drug demonstrating significant improvement over available therapy. The Fast Track is similar to the Breakthrough Therapy but applying to drugs treating serious disease without current treatment available.[25] Treatment intending to treat rare disease (like rare form of cancer) can go through an accelerated version of the clinical trials, with the Phase 1 and 2 combined into one due to a fewer number of patient meeting the recruitment requirement.[26]

In Chapter 2, we will review the current phototherapy-based treatment for cancer and compare their efficacy to current treatments for phase III trials. This will allow us to identify the form of cancers not covered by phototherapy research.

1.4. The Chemokine path of communication between cancer cells and their environment.

Some polymers have shown antitumor activity and effects on the production of chemokines by cancer cells[27–29]. Cytokines and chemokines are proteins secreted by different types of cells as communication vectors and regulators. They are particularly used by the immune system to regulate the response of various cells. Chemokines can be over- or under- expressed when cells become cancerous, increasing their growth, inhibiting immune cells, and inducing metastasis. They are either secreted by the cancerous cells or the tumor microenvironment, creating communication between both, which decreases the levels of pro-apoptotic proteins and favors growth.[30] Therapies targeting their regulation have been developed as cancer treatments[30–32]. Each chemokine has a different role, and their regulation and effects are cell type dependent.

RANTES (CCL5) has been shown to both tumor progression promotion and recruits anti-tumor immune cells in different studies, especially in melanoma[30–32].MIP-2α (CXCL2) is involved in inflammatory response, leukocyte and neutrophil chemotaxis[33]. It is a prognostic marker in several types of cancer[34], is correlated with neutrophil invasion[35,36] and promotes the generation of monocytic myeloid-derived suppressor cells[37]. MIP-1β (CCL4) receptor is present on T-cell and plays a role in their recruitment and differentiation[35,38]. It is also a chemoattractant for NK cells and B cells, promoting their infiltration into tumors in the case of pancreatic cancer[39]. MCP-1 (CCL2) is known to attract monocyte and dendritic cells, but also tumor-promoting myeloid cells[32]. MCP-3 (CCL7) is a chemoattractant for various leukocytes, monocytes and NK cells; it

can promote tumor progression and metastasis[40,41]. MIP-1α (CCL3) stimulates tumor growth[31,32]. GROα (CXCL1) is a tumor promoting inflammatory chemokine[32]; it is upregulated in melanoma and is implicated in tumor growth[30]. IP-10 (CXCL10) can suppress angiogenesis and attract T cells, limiting tumor growth[30].

Eotaxin (CCL11) recruits eosinophils into the tumor microenvironment, playing an anti-tumor role. It is related to tumor suppression in breast cancer patients[42].

As chemokines are part of the communication network between the immune system and tumors and promote tumor growth, it remains interesting to study the effects of our treatment (mainly GC effects on cancer cells directly) on chemokine regulation to see if it can favor the recruitment of anti-tumor immune cells and reduce the tumor growth signals.

1.5. Objectives

Overall, three different objectives were defined, with different questions and hypotheses developed based on the current research on GC and phototherapy. In this study, we will first review the different clinical trials using phototherapy as a cancer treatment, their progress, and results. Then, as GC has been studied in cancer combinations, we will investigate the toxicity and effects on chemokine regulation of GC in cancer cell lines and some immune cells. Finally, in addition to the previous studies on the functions of GC on immune cells, we will also study the effects of GC on the activation of epithelial cells, in order to extend our understanding of the immunological mechanism of GC, which coulg further enhance cancer therapy and antiviral effects.

Chapter 2. Intranasal vaccination, a new route of immunization

2.1. Vaccines and immune adjuvant

To understand what an immune adjuvant is, we first need to understand how a vaccine works and what it is composed of. A vaccine is a solution mainly composed of an element derived from a pathogen administered to a person, along with an adjuvant. This element can be an attenuated form of the pathogen, a portion of its membrane (or capsule), proteins or, in the most recent developments, RNA or DNA. All of these elements can be recognized as one or several antigens targeting the pathogen of interest. The administration of this component in the body allows the immune system to recognize and generate memory cells targeting the pathogen. Attenuated forms and membrane portions are easily recognized by the immune system as an intruders due to the abundance and size of possible antigens. However, vaccines composed only of small elements, like proteins, RNA or DNA, are less readily recognized and phagocytosed by APCs. Therefore, another component is added to the vaccine composition to elicit the innate immune system to recognize the antigens: the adjuvant.[11,43,44]

An adjuvant can be an emulsion, a polymer or a molecule. These adjuvants create an "immunocompetent" environment at the injection site, resulting in increased recruitment and activation of the innate immune system[45]. MF59 and AS03 are two examples of oil-in-water emulsions introduced and used as adjuvants for the influenza vaccine in 2009[43,46]. The mechanism behind the action of an adjuvant might be unique to each one, but we can categorize these modes of action according to their origin (**Figure 2**), all leading to an increased activation of the immune system at the site.

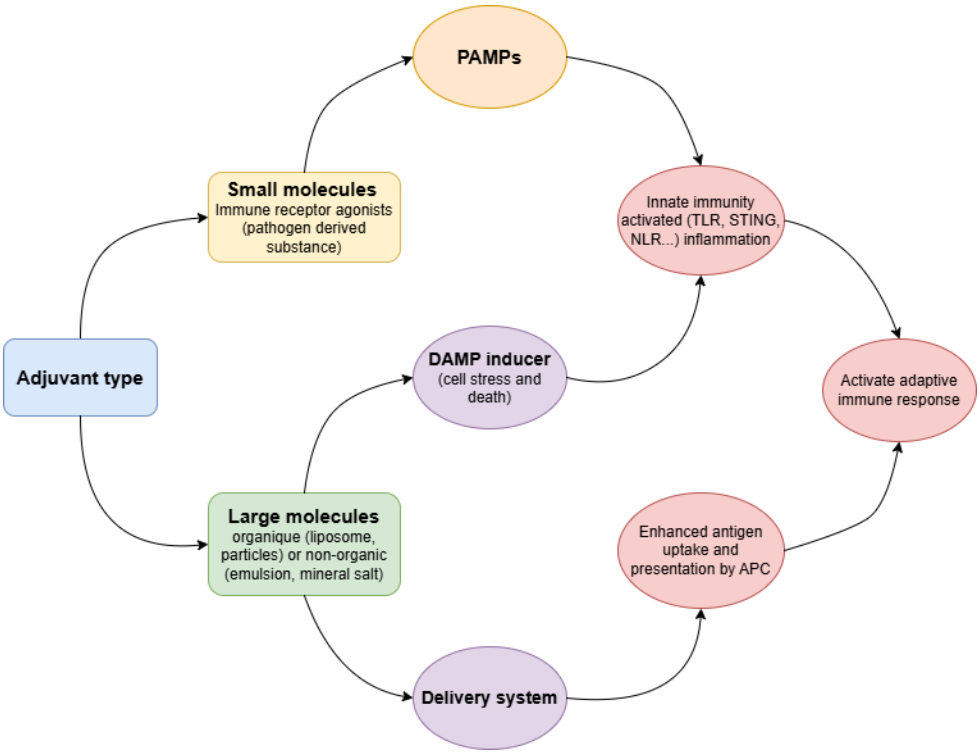


Figure 2. Different types of adjuvants and their mechanism of action.

Traditionally, the vaccines are developed and stored in solution and administered through one or several injections in people. The components are directly in contact with the tissue-resident immune cells and activate the adaptative immune system. A vaccine's efficacy is evaluated clinically through its ability to provoke a systemic immune response and the level of antibodies generated targeting the antigens.[11]

However, this method of immunization presents several issues. First, a major part of the population is afraid of needles, causing them to refuse vaccination. Second, injections require specific training, so they can only be performed by certified caregivers, limiting the accessibility of the vaccine.

Another limitation of these vaccines is the non-specificity of the response. Unless the patient is contaminated surgically or through perfusion, the pathogen always enters the body through a wound or a mucosa. The vaccination route does not reflect the reality of the contamination, and the immune system's memory of the pathogen might not be completely adapted to response to the real pathogen.[11]

Therefore, new vaccination routes have been developed to address these limitations.

2.2. A new vaccination route, the mucosa

The mucosa is the first line of defense against pathogen in our body and the major route of infection. It is the first physical barrier to pathogens but also the residence of specific immune cells

and proteins, like IgA[47]. The immunization route can be adapted according to the pathogen's route of infection. For example, vaccines against airborne pathogens can be introduced through a liquid sprayed in the nose, while immunization against water and foodborne pathogens can be stimulated through pills. Introducing the vaccine via the same route the pathogen uses to enter the body allows the immune system to be more efficient in its response as the required and specialized adaptative immune cells are already in memory[43,44,48]. APCs can recognize pathogens with their receptors, but the epithelial cells composing the mucosa also possess these receptors and other mechanisms to recognize pathogens[49,50].

Chitosan has been studied as an adjuvant and has demonstrated immune stimulation associated with biosafety, either in form of salt or as the major component of a nanoparticle to deliver antigens[51,52]. As GC is a modified version of chitosan, we can easily hypothesize that GC could be a promising immune adjuvant for an intranasal vaccine. A preliminary study using a SARS-COV-2 proteins - GC vaccine formula was published previously, showing promising results with the intranasal route of immunization[10]. We have various results showing how the immune system is stimulated by GC, but no results are available regarding the direct reaction of the mucosa.

Chapter 3. Current treatment protocol for Cancer

3.1. Life Expectancy, cure fraction and Time to cure

The European Cancer Register (EUROCARE) is a project that brings together cancer-related data from all participative institutes, research laboratories, and care centers across Europe. The data from EUROCARE-5[9] includes "population-based indicators of cancer cure" from people diagnosed between 1990 and 2007. EUROCARE-6 is still ongoing but has already published collected data which compared with EUROCARE-5[53]. Different parameters can be cited when we want to search for the efficacy of a treatment or the fatality of cancer types.

First, Life Expectancy can be defined as the estimated time left for a patient between diagnosis and death[9,54]. The EUROCARE-5 study gives us an estimation of the life expectancy of patients according to the type of cancer and their age at diagnosis. The most fatal cases (with less than a year of estimated life expectancy) are those with liver, pancreas, gallbladder, lung, esophagus, stomach and brain (after 45-year-old) cancers. The patients with the longest life expectancy (more than 5 years) are those diagnosed with breast, testicular (between 45- and 54-year-old), prostate (after 65-year-old), non-Hodgkin lymphoma (after 45-year-old), leukemia (between 55- and 64-year-old) and small lymphocytic lymphoma/chronic lymphocytic leukemia.

Second, the Cure Fraction is the proportion of patients declared as "cured" of their condition and expected to have a longevity similar to the general population[9,55]. The time to cure is the duration between diagnosis and when the probability of death due to the cancer becomes negligible[9]. The overall cure fraction is 39% for men and 51% for women. The cure fraction for one type of cancer varies between men and women but we can still observe that the cases with the smaller life expectancy correspond to the smallest percentage of cure fraction for both sexes (
).

Finally, Time To Cure can be defined as "the number of years after cancer diagnosis when the excess mortality due to cancer becomes negligible". According to the report, the different forms of leukemia, lymphoma, myeloma and laryngeal cancer can take more than 25 years to cure and are the longest to cure. The shortest Time To cure (less than or around a year) are testicular cancer (for men younger than 65), and thyroid cancer (for men younger than 45 and women younger than 65).[9]

All of these numbers still highlight the need for more efficient diagnosis and treatment plans to decrease the number of deaths from cancer and increase the lifespan of cancer patients. The cancer statistics report published each year by the American Cancer society provides different statistics, such as the number of new cases and deaths due to cancer by state, sex, ethnicity, and age, and tries to estimate statistics for the following years. Their conclusion is similar: cancer is still a leading cause of death despite a decline in mortality. Cancer prevention also needs improvement, even though the decline of smoking since 1991 has contributed to a reduction in new cases.[8]

Each type of cancer and its stages is treated differently to adapt the genetic profile of the primary tumor and the side effects.

Table 1. Cure Fraction by type of cancer and sex[9].

Cure Fraction (%)	Men	Women
75-100	Testis cancer Skin melanoma	Thyroid cancer Skin melanoma Corpus uteri cancer
50-75	Follicular non-Hodgkin lymphoma Connective tissue Prostate cancer Hodgkin lymphoma Thyroid cancer	Connective tissue Colorectal cancer Follicular non-Hodgkin lymphoma Colon cancer Rectum cancer Vagina cancer Cervix uteri cancer Breast Cancer Hodgkin lymphoma
25-50	Oral cavity and pharynx cancer Larynx cancer Diffuse large-B cell non-Hodgkin lymphoma Kidney cancer Rectum cancer Bladder cancer Colorectal cancer Colon cancer	Non-Hodgkin lymphoma Ovarian cancer Diffuse large-B cell non-Hodgkin lymphoma Kidney cancer Oral cavity and pharynx cancer Bladder cancer
0-25	Pancreatic cancer Liver cancer Lung cancer Small lymphocytic lymphoma /Chronic lymphocytic leukemia Esophagus cancer Multiple myeloma Brain cancer Gallbladder cancer Leukemia Stomach cancer Non-Hodgkin lymphoma	Pancreatic cancer Liver cancer Multiple myeloma Gallbladder cancer Esophagus cancer Small lymphocytic lymphoma /Chronic lymphocytic leukemia Brain cancer Leukemia Stomach cancer Larynx cancer

3.2. Different cancer treatments adapt to each case: surgery, chemotherapy, radiotherapy, immunotherapy and combinations

The course of treatment is determined according to the type, stage and genetic profile of the cancer. We can distinguish four main categories of treatment: surgery, chemotherapy, radiotherapy and immunotherapy. These treatments can also be combined to achieve a higher regression of the tumor and/or increase the lifespan.[15]

Surgery is primarily used to treat early stages of cancer by partially or completely removing the primary tumor. For example, stage 0 breast cancers are removed by surgery and might be combined with radiotherapy or endocrine therapy according to the tumor profile. In more invasive early stages, a combination therapy including chemotherapy or immunotherapy may be used as a phase 2 treatment, followed by tumor resection.[56] Lung cancer is rarely treated using surgery, it is primarily uses in early stages.[57] Some cases of thyroid cancer (e.g. papillary microcarcinoma, where treatment gives a relapse-free survival after 5 years of 78.6%) and prostate cancer (which benefits over time with a significant reduction in metastases and a reduced need for other therapies) can also be treated with surgery[58,59].

Chemotherapy refers to the use of a broad range of drugs that target the DNA, causing damage and cell death in tumors. These drugs are administered to patients intravenously or orally. However, most of these drugs do not specifically target cancerous cells, leading to significant damage to healthy cells in the body and causing a variety of side effects.[15] For cases of hepatocellular carcinoma (liver cancer), chemotherapy is one of the best options for advanced stages, increasing survival. However, some cases develop drug resistance after 6 months of treatment (e.g., with sorafenib).[60] Chemotherapy is also an option for breast cancer patients in both early and advanced stages (Stage II to III), with some combinations of chemotherapy drugs showing a few months' increase in overall survival.[56,61] However, for metastatic thyroid carcinoma, chemotherapy has shown a poor response rate.[62] Thus, it is not always the best treatment, and new combinations of chemotherapy drugs are being studied to increase survival. For example, numerous new chemotherapy drug combinations are being explored in clinical trials to prolong the life expectancy of stage IV colorectal cancer patients[63].

Radiotherapy uses radiation to kill tumors. It is used as frequently as chemotherapy to treat advanced lung cancers (about 40% of cases).[57,64] Radiotherapy is widely used in combination with chemotherapy to treat pancreatic cancers[65].

Immunotherapy refers to treatments that use the tumor as a source of antigens, which the immune system will recognize and use to completely remove the remaining cancerous cells. Monoclonal antibodies are a type of immunotherapy. These antibodies either target receptors on the tumor or are used as immune checkpoint inhibitors. Immunotherapy can be used for advanced stages of breast cancer[56] and liver cancer[60]. It has been extensively explored in combination therapies, especially with new treatment techniques like phototherapy[1,7,66,67].

3.3. Conclusion

Current cancer treatments are not efficient for every type of cancer, and some forms still lack effective treatments to significantly decrease mortality rates. The combination of existing treatments is one avenue being explored clinically to improve overall survival, enhance treatment response, and overcome drug resistance without exacerbating side effects. However, new treatment strategies are also being investigated in both pre-clinical and clinical studies, which combine existing therapies with innovative technologies or introduce entirely new approaches, such as phototherapy.

Chapter 4. Principle of phototherapies

4.1. Photodynamic Therapy

Photodynamic therapy (PDT) is a non-invasive type of therapy where a photosensitizer (PS) is administered to the treatment site and then excited to create reactive oxygen species (ROS) by energy transfer [68,69]. It is used in dermatology, chronic inflammation, drug-resistant bacterial infection and as cancer therapy[69]. The PS is able to specifically accumulate into the targeted tissue, so it is administered into the system then the targeted tissue is exposed to light corresponding to the PS excitation wavelength. In presence of oxygen, the energy generated by excitation will be transferred to the oxygen molecule that will become a reactive oxygen species that will cause cell death in the tumor and an inflammation at the local site[68,70] (**Figure 3**).

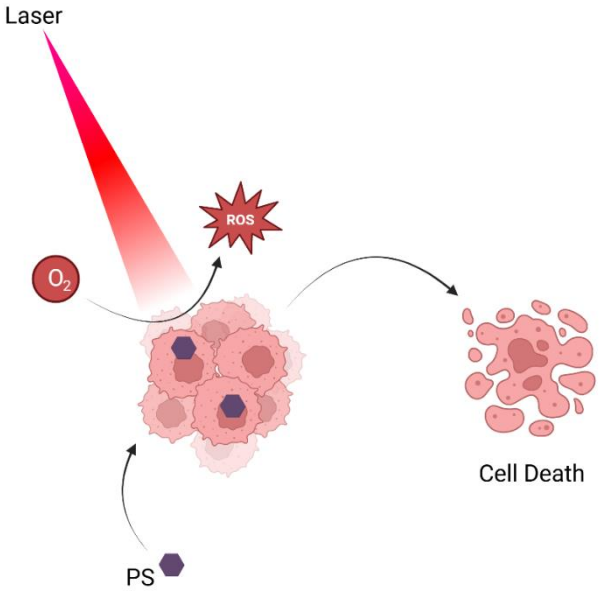


Figure 3 Photodynamic Therapy mechanism. PS: photosensitizer, ROS: Reactive Oxygen Species. Created with Biorender.

The first key challenge of this technique was to specifically accumulate the PS in the tumor instead of healthy tissues to prevent harmful side effects[69]. Recent technological developments enable a better selectivity of the treatment against tumors.

The use of new PS with minimal side effects through specific receptor targeting is one of these developments. For example, studies show the tendency of PS to accumulate in tumors cells by transport in low density lipoproteins (LDL). PS accumulate in LDL preferentially[71,72] and tissues with important mitotic activity, like tumors, show an increase concentration of LDL receptors on their surface, hence the higher concentration of PS in tumors[72,73]. However, this does not entirely explain the PS biodistribution. More research is required on this part of the PDT mechanism. The second limitation of this technique is the light. The excitation light needs to penetrate deep enough in the tissue to be absorbed by the PS and produce ROS. However, tissues are not a completely

transparent material, a portion of the light is absorbed by every tissue layer or it can be scattered or reflected between layers, and only a portion of the light reach the tumor site.[74]

Studies using infrared light have more successful rate due to the depth penetration into tissues using longer wavelengths[75]. Some other systems have been developed to enhance the penetrability of the light and reduce the proportion of light scattered by the tissue, like chemiluminescence combination with PDT[76], fiber optic[77], self-luminescence[78,79] or X-ray[78,80].

While *in vitro* studies can identify suitable PS agents, successful animal studies are required for treatment approval in clinical studies. While subcutaneous models provide an indication for success probability, orthotopic models are more reliable in a clinical efficacy point of view[69]. In clinical trials, 7 PS are evaluated across different studies in 2023 and dozens of others are still studied *in vitro* and *in vivo*[81]. PDT treatments are available in some specialized treatment, but its application is still restricted to a limited number of cases. These treatments are not suitable for disseminated tumors[69], proving the need of further studies to increase its application range.

4.2. Photothermal Therapy

Photothermal therapy (PTT) is a type of therapy where heat is produced by light exposure, with or without the use of photothermal molecules or particles. PTT can use a PS that generate heat out of the absorbance of light, generally near infrared light for their penetrability[3,82,83]. When the heat attain a range around 50-70°C, it induces immunogenic cell death and the release of heat shock proteins (HSP) which are recognized by the immune system as damage-associated molecular patterns (DAMP) and tumor associated antigens [3,18,82]. Above 80°C, vaporization starts to happen and every structure, protein and cell component are destroyed by heat without generation of DAMPs[84]. Cancer cells are more sensitive to elevated temperature than normal cells so this strategy of treatment is consistent with the disease[83](
Figure 4).

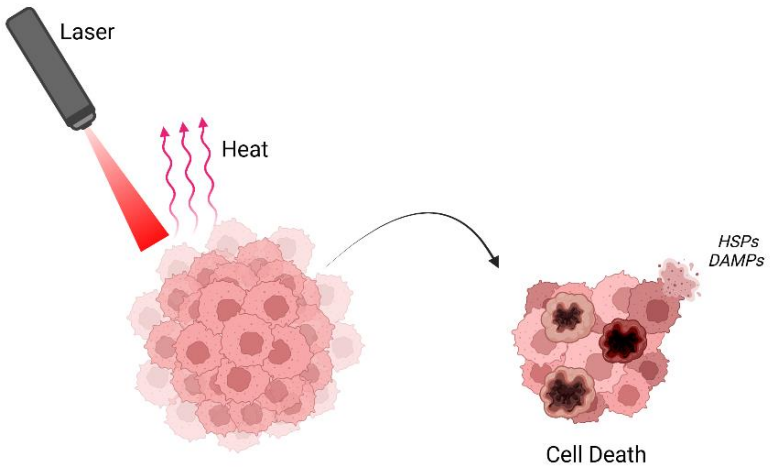


Figure 4. Photothermal Therapy Mechanism. HSP: Heat Shock Protein, DAMPs: Damaged-associated Molecular Patterns. Created with Biorender.

PTT application is mainly explored in cancer therapy but it can also be applied to tissue regeneration or drug delivery[85].

However, the short range of temperature and laser penetration depth limit the good results of this type of therapy. Controlling the temperature range inside the body to keep the tumor within the immunogenic cell death temperature range and limiting the increase of temperature in the healthy surrounding tissue remain significant challenges. The 100% cancer cell death is not guaranteed, and the tumor microenvironment can inhibit the immune system, preventing the uptake of the DAMP by APC and the activation of the immune system[18]. PTT itself is not effective enough to completely remove a tumor in a patient[82].

4.3. Photochemical Internalization

Photochemical internalization (PCI) is similar to PDT from the use of photosensitive agent. For this case, these agents are used to facilitate the transport of molecules in the cytosol. The PS is entrapped by endocytosis inside a vesicle, or in the vesicle membrane, with the molecule used as a treatment, then the vesicle is broken by the formation of ROS due to light exposure of the PS. The molecule will then be released inside the cytosol to interact with proteins within the cytoplasm or migrate to the nucleus (
Figure 5)[67,86,87]. It is a technique also used for other purpose drug delivery, like intracellular bacterial infection, or vaccination[86].By definition, PCI is never employed by itself due to its mechanism of action, it is always coupled with a drugs like chemotherapeutic (chemo) agents or antigens. Thanks to PCI, the molecule can be concentrated inside the cancer cells and achieve the same effect at lower dose than alone. It also minimizes the side effects from the interaction of the molecule with healthy tissue.

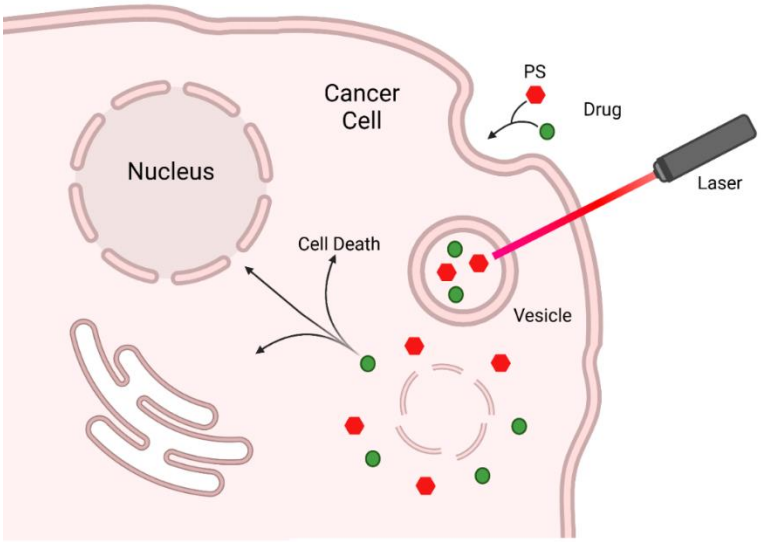


Figure 5. PhotoChemical Internalization mechanism of action. PS: photosensitizer. Created with Biorender.

The PS needs to accumulate preferentially in vesicles and not diffuse across the cell membrane to correctly activate PCI. Hence, it limits the PS's choice to those with a high molecular weight and a particular pKa to not escape acidic environment[87]. Another limit is the degradation of the associated drug. The light exposure can destabilize the molecular structure of the chemo and break some liaison inside the molecule, decreasing its activity[88].

The PS can also be a targeted toxin. Targeted toxins are molecules engineered as the combination of two molecules, one binding to the cell (like an antibody or a receptor ligand) and one killing the cell (like chemo). However, this type of PS has poor tumor penetration due to its size.

PCI is not a technique commonly tested in preclinical studies. Between 2009 and 2020, only 16 preclinical studies (4 only *in vitro*) and 2 clinical studies were reported for cancer treatment. It shows promising results, especially in cases of chemo resistance, but more studies evaluating the application of this technique in combination with other methods are necessary to improve it[67,89,90].

4.4. Combination Therapy

Combination therapy is also a part of the recent advances made to increase the success rate of the treatment. One combination treatment explored by different groups is PDT with immunotherapy and immune vaccines. PDT triggers the immune system by releasing tumor antigens with the cell death, and immunotherapy enhances the immune response in tumors and the rest of the body. Dendritic cells and macrophages, also known as antigen-presenting cells (APCs), present these antigens to CD4+ and CD8+ T cells that will induce cell death to the residual and metastatic cancer cells. Checkpoint inhibitors, immune adjuvants, or toll-like receptors agonists combined with PDT have been explored and they have shown promising results *in vivo*[68,91]. Combination of PDT and photothermal therapy is another advancement made in the field. Some PS can also act like a photothermal agents (PTAs), meaning converting light into heat, hence combining both techniques with one molecule[92].

The result can be increased by combining PTT with other techniques like immunotherapy or nanomedicine. The combination of PTT with an adjuvant increase the survival and assure the resistance against tumor rechallenge in *in vivo* model[3].

Nanomaterials, organic or metallic, can also be used in replacing the PTA to induce more local heat with the laser. Nanomaterials have the ability to accumulate into specific tissue due to their nature or their structure, concentrating the heat into the tumor tissue[66]. They can also be applied to the body by the use of a hydrogel containing the PTA and other drugs[82,85]. Combination of PTT with nanomedicine is largely explored in case of bone tumors because the hypoxic environment in the bone limits the application of PDT for this organ due to the poor production of ROS[83]. The use of carbon nanotubes is also mainly explored in combination with PTT due to their biodegradability and the ability of these carbon-based nanomaterials to convert light into heat. Several studies show good survival results, above 50% in *in vivo* studies, to these combined treatment with an activation of the adaptive immune system at the tumor site[82,93,94].

PTT was also combined with immunotherapy using GC to stimulate the immune response after

the release of antigen by PTT.[1,95,96]

In this review[97], 5 *in vivo* studies are presented combining immunotherapy and PTT with the use of nanomaterials, and survival of animals with the treatment to certain form of skin cancer are shown. Other previous reviews show promising results in combination therapy *in vivo* and clinical trials[98].

The PS can also be a nanoparticle engineered to contain everything required for the treatment (PS, delivery system and chemo drug)[87].

PCI can be combined with an adjuvant, or a checkpoint inhibitor use in immunotherapy to stimulate the immune system after tumor cell death. It can also directly target APCs by delivering the PS coupled with tumor-specific antigens and applying the light to force the antigen uptake by the proteasome and then MHC I[67,88].

4.5. Conclusion

PDT, PTT and PCI hold promising results as cancer treatment, alone or in combination with immunotherapy. The use of light to provoke immunological cell death (directly or indirectly with the transport of chemo drugs) allows the production of DAMPs recognizable by the immune system enabling an immune response targeting the rest of the cancerous cells and the potential metastasis. GC was combined with PTT (PTT +GC = laser immunotherapy, LIT) to enhance the immune response against cancer, resulting in the complete recession of the primary tumor and metastasis. Cured Mice also survived a rechallenge, proving an immune memory against cancer.
Even if there are still some limitations to the techniques, clinicals trials were already held to test those techniques in humans.

Chapter 5. Ongoing clinical trials treating cancer using phototherapy

5.1. Objectives

In this chapter, the objective is to report on previous, ongoing and completed clinical trials found on the website clinitrials.gov and to compare their results to current treatment. With these data, we can emphasize the potential of phototherapy, and LIT, in cancer treatment but also identify types of cancer left out of those studies, needing more attention for the scientific community.

5.2. Methods

On this website, clinicaltrials.gov, queries were made using the key words “cancer” for the condition, and “photothermal therapy”, “photodynamical therapy”, photochemical therapy” or “phototherapy” for the treatment. 466 trials were collected with these terms. From this set, the studies were separated according to the type of treatment applied. The groups were realized using the filters of the website primarily, then by searching and selecting the name of the three methods in the “interventions” or “brief summary”, because certain studies only named the drug in the intervention section, information. We obtained 320 PDT studies, 4 PTT studies and 8 PCI studies. Finally, studies in double and not concerned by cancer type of disease were removed to obtain 209 PDT studies, 4 PTT studies and 6 PCI studies.

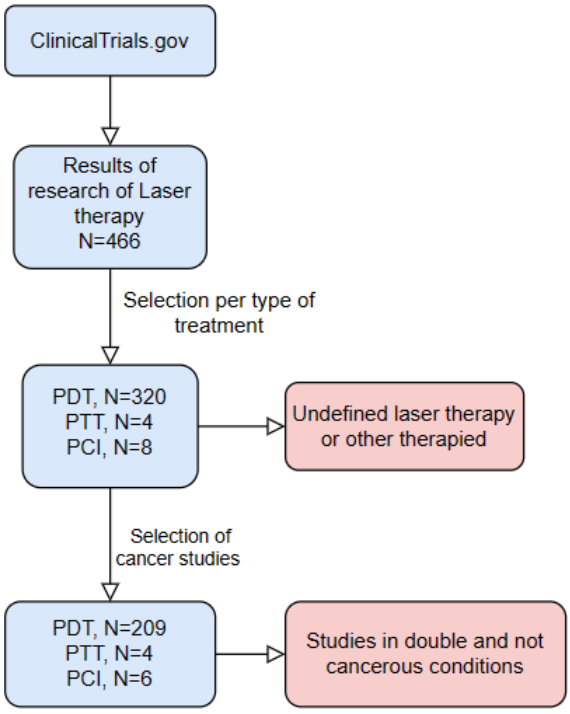


Figure 6 Flowchart of the studies collected.

Cervical dysplasia premalignant was removed due to the description of the disease stating that it cannot become cancer, Paget disease, periodontitis and oral leukoplakia were removed because it

cannot become cancerous. High grade dysplasia was kept because it is considered stage 0 of cancer. Actinic keratosis, port-wine stain and warts were removed because it is not cancerous.

Additionally, the selection process was summarized in the flow Chart **Figure 6**. The list of every study included in this review is given in supplementary.

For every study, the NCT number, the study title, and the URL were extracted to retrieve more data later from the website, if needed. The status, results, condition, intervention, phase, and type of study were used for the statistical analysis. Finally, the primary outcome measures, the collaborators, and the brief summary constitute more details about the trials. Some studies treating conditions like dermatitis, actinic Keratosis, wart were removed because the conditions are not defined as a type of cancer. Categories of conditions were created to regroup by type according to the concerned system

Table 2.

Table 2 Categories of cancer types

Category	Type of cancer
Digestive system	oral, liver, pancreas, esophagi, rectal, colon, anal, bladder, renal and bile.
Reproductive system	Prostate and breast.
Skin	basal cell, skin, Bowen's disease, melanoma and intraepithelial
Respiratory system	lung and Peritoneum
Neural system	Brain, neuro, glioblastoma, cervical and neck
Lymphoma	T-cell lymphoma, mycosis fungoides and lymphoma

One last category was specifically created to count the number of studies treating cancer with metastasis. Categories were also made according to the study status
Table 3.

Table 3 Categories of status

Category	Status
Stopped	Withdrawn, terminated, not yet recruiting, no longer available, suspended, unknown
Ongoing	Active not recruiting
Completed	Completed
Recruiting	Recruiting, enrolling by invitation

The "Stopped" category regroups all studies that were stopped or not started due to different reasons, or the data was not updated within the 2 required years after the completion date. All the studies were separated and counted by treatment type followed by a count of the completed studies,

specifically by treatment type. For the phase, “blank” means not specified - or not applicable - on the website, meaning the study was approved by the FDA as one that does not follow the classic process per phase due to its design or purpose. The last research was done on 8 February 2024 for PDT and PTT and on 3 April 2024 for PCI.

5.3. Results

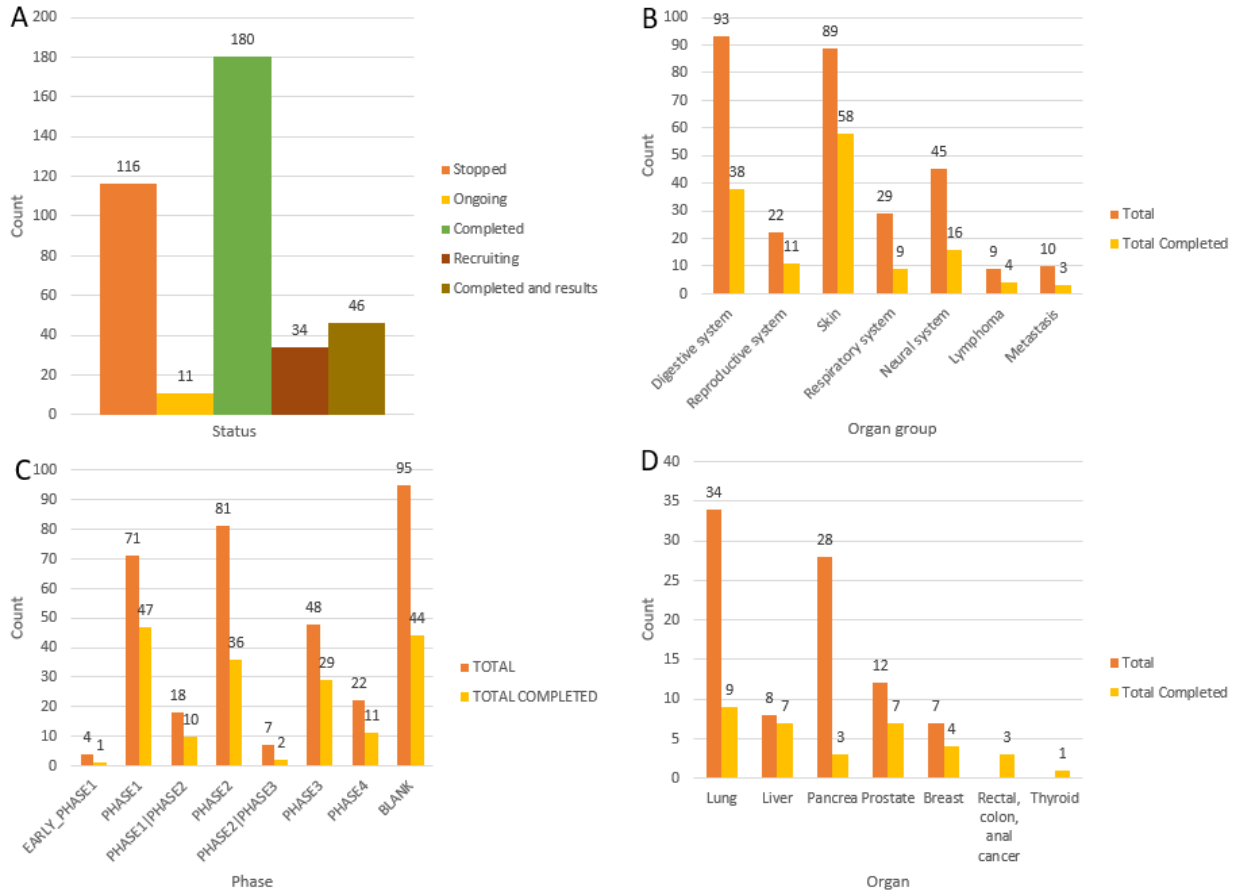


Figure 7 Total clinical trials inventory in the dataset per field. A: Total per status, **B**: Total and completed per conditions, **C**: Total and completed studies per phase, **D**: Total and completed studies by conditions belonged to the skin category.

According to **Figure 7**A, on a total of 387 studies, 46% are completed, 30% have been stopped and only 12% published results on the website, representing a really small portion of all the studies but it does not seem that surprising when most of the clinical trials fail due to failing treatment or budget issues.

Of all the types of conditions, most of the studies were carried out on the digestive system or on different types of skin cancer. However, on the completed studies, the majority involve skin type of cancer and there is a few studies on the different types of lymphoma and thyroid cancer, the respiratory system and on metastatic forms of cancer.

5.3.1. *Result per Treatment*

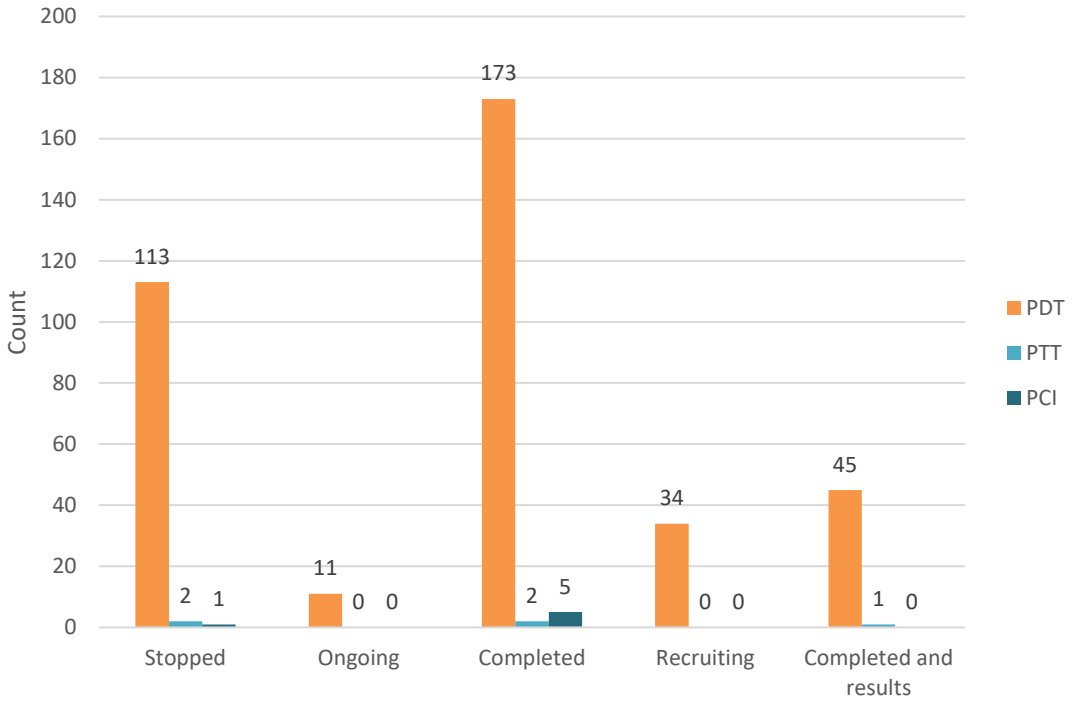
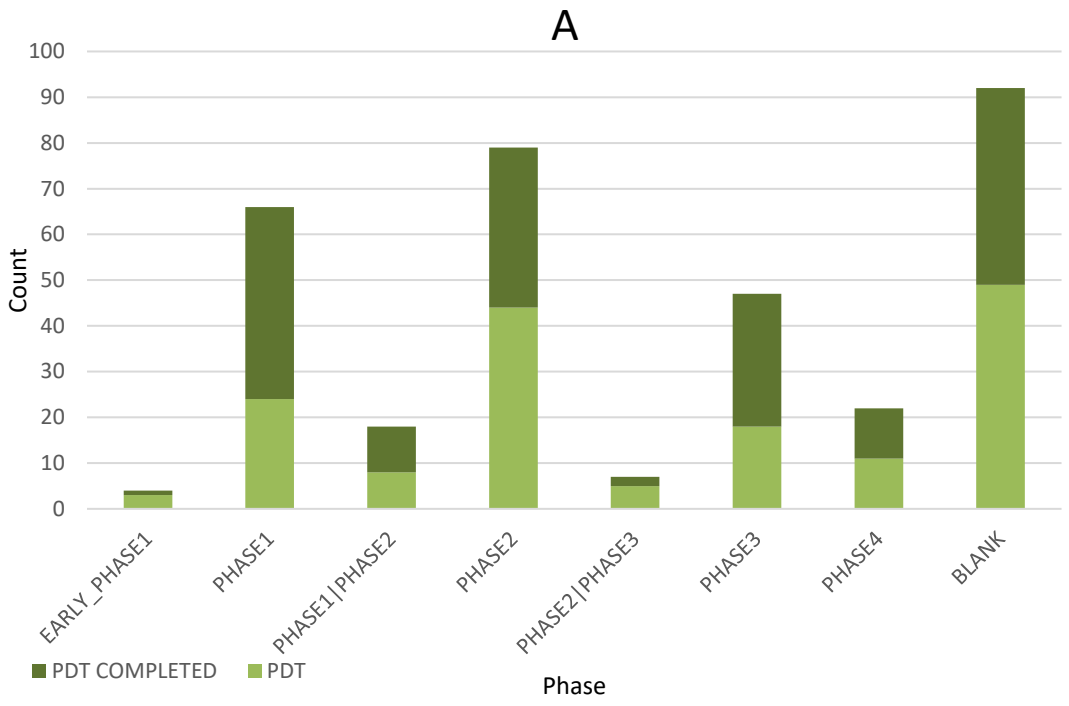


Figure 8 Repartition of the studies by status and type of treatment.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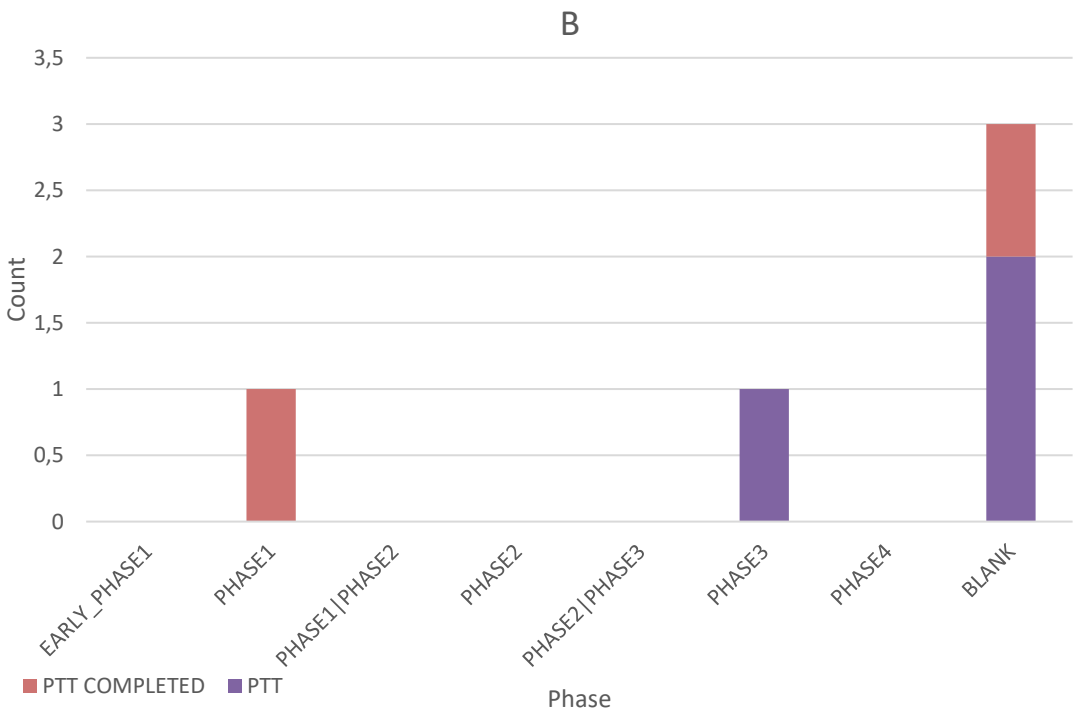
B
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2,5
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EARLY_PHASE1
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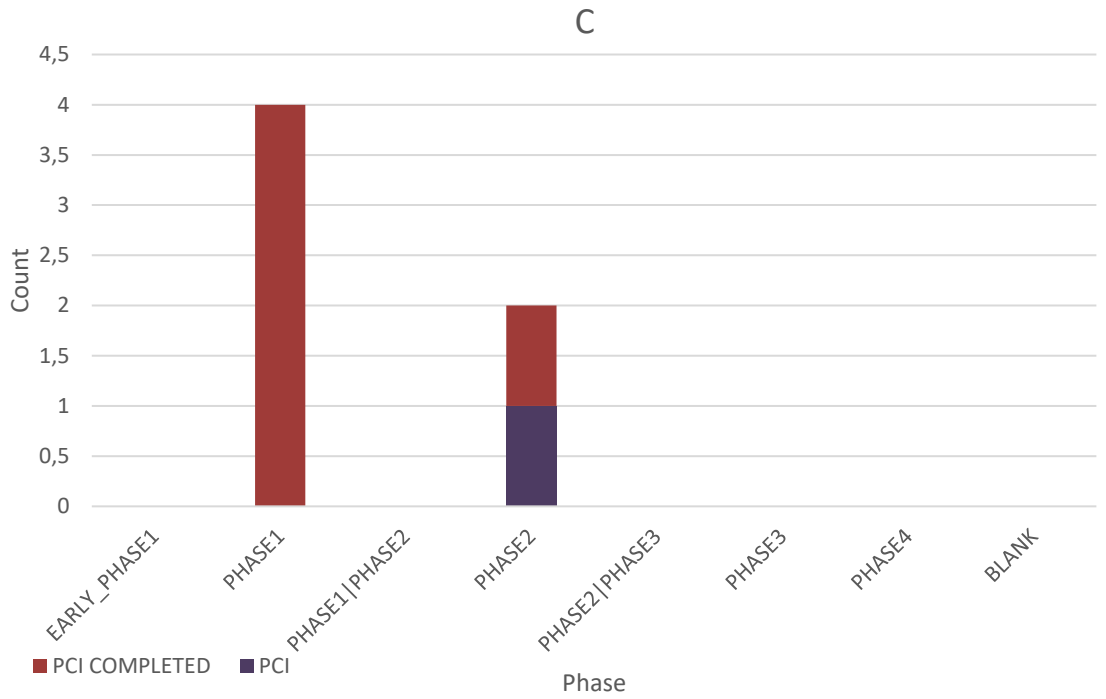

Figure 9. Number of Studies by Treatment (status completed separated from the others). A: Photodynamical therapy, B: Photothermal therapy, C: Photochemical internalization.

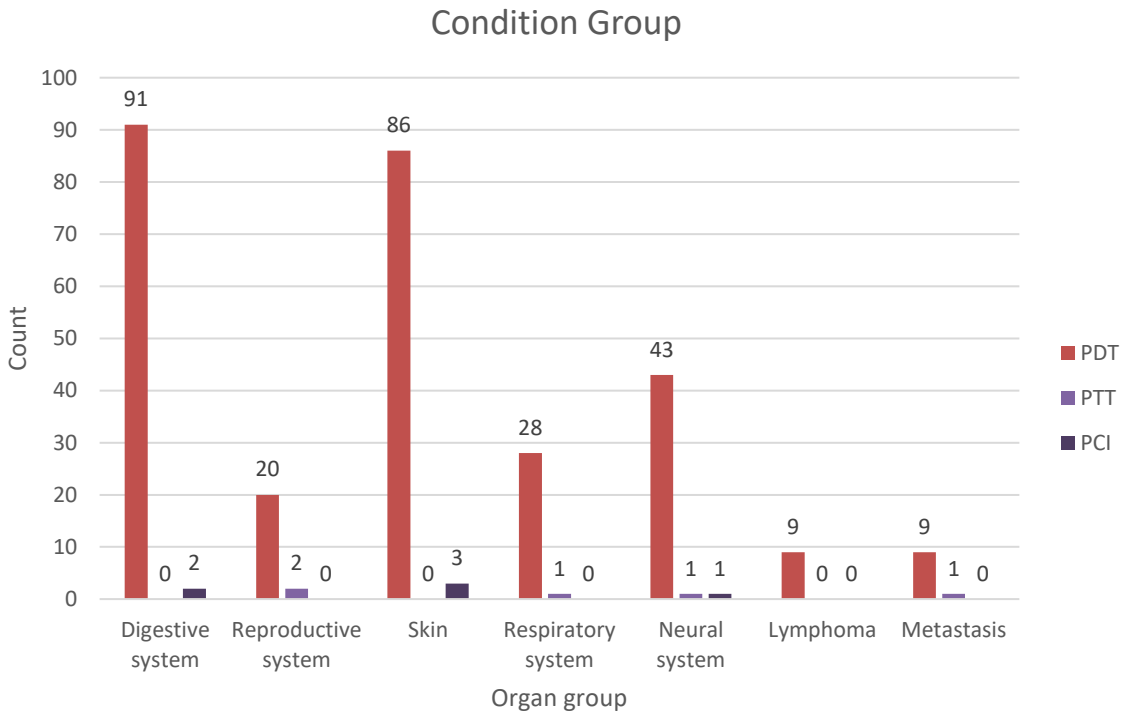

Figure 10. Number of studies by organ group by treatment.

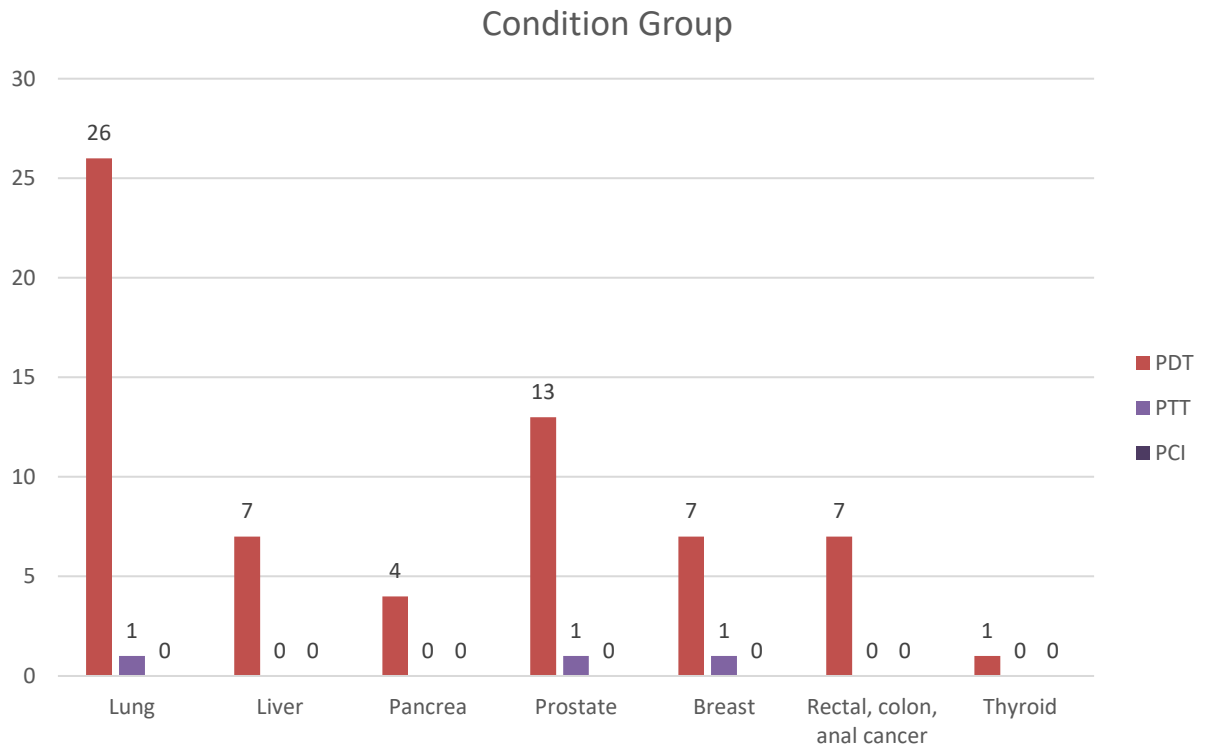

Figure 11. Number of Studies by treatment for cases taken as an example in chapter 2.2. and there is a few studies on the different types of lymphoma and thyroid cancer, the respiratory system and on metastatic forms of cancer.

5.3.2. Result per Treatment

From the total data collected, most of the studies used PDT as a treatment. According to **Figure 8**, almost half of the PDT studies are completed but only 5% published their results and only 1 PTT study published its results. 2 studies marked as complete without results still have publications with promising results published online[99,100]. The investigators may not have published their results on the website and preferred writing a publication instead. The same phenomenon happened for one PTT study[7]. According to **Figure 7**C and **Figure 9**, most of PDT studies are still in PHASE1, meaning most of the clinical trials are recent. **Figure 10** confirms that the majority of the studies for the different groups of organs are represented by PDT studies. **Figure 9** shows that less than 10 trials are available to the public as a treatment option in phase 4. It shows the novelty of this treatment strategy and the need to continue trials.

As for the organs used as examples in chapter 2.2 (**Figure 11**), there are only studies using PDT and PCI.

5.3.3. Comparison between phototherapy and current treatment. LIT results in clinical trials.

The PDT clinical trial NCT 00083785 in phase 1 treating colorectal liver metastases published its results[101]. It observed a tumor response rate of 33%, which is in the range of the current response to treatment for resectable cases[102].

LIT was studied in a phase 3 clinical trial against late-staged breast cancer[7]. Ten patients were enrolled, and preliminary toxicity tolerance and adverse events were studied. The results are that no serious side effects were assessed, some adverse events were observed at the treatment site due to the local administration of the treatment. The response rate was 62.5% and the beneficial response rate was 75% and one patient achieved a complete cure for the primary tumor and lung metastasis. This study holds great potential as the results are better than any existing treatment.

5.4. Conclusion

Phototherapy is a type of therapy with a high potential to cure patients with cancer and with advantages from traditional treatments. Their function can be used as treatment for a lot of diseases, conditions, or pain. Depending on the results, dosage of the PS and power of the laser can be adjusted for biological function modulation or cell death induction.

An increasing number of preclinical studies is observed in previous years but there is still a gap with the number converted into clinical trials to treat human patients. Some clinical trials show promising results on patients with good responses and a decreased number of adverse events due to the treatment like LIT.

Chapter 6. GC safety and regulation on chemokines' production in a cancer treatment context

6.1. Objectives

The safety and effect of GC on the tumor directly were never studied. Therefore, we first look into the viability of different immune and cancer cell lines to know whether GC is toxic or not for those types of cells. Then, as described in the introduction, chemokines are a path of communication between the tumor and its environment, the associated cells and the immune cells recruited on site by LIT treatments. Chemokine can also be a prognostic marker for some cancers. Therefore, we studied by multiplex whether GC can have an up or down regulation of the cancer cells' chemokine production**.** This results will extend our knowledge of GC's immunological mechanism to further enhance cancer therapy.

6.2. Materials

The cell lines used were BlaER1 Human B-cell Precursor Leukemia, THP-1 Human Monocyte (ATCC TIB-202), Panc02H7 (7^{th} metastases of Panc02 Mouse Pancreatic cancer cells), B16-F10 Mouse Melanoma (ATCC CRL-6475), 4T1 Mouse breast cancer (ATCC CRL-2539).

Aqueous solution of N-dihydrogalactochitosan, also referred to as glycated chitosan (GC), (10mg/mL) was provided by ImmunoPhotonics, Inc.

The Cell Counting Kit 8 (CCK8) was purchased from Dojindo Laboratories (CAT#CK04-13). The absorbance was measured using the BioTek Synergy Neo2 Multimode Reader. The chemokines were measured using a ProcartaPlex Mouse Chemokine Panel 1, 9plex purchased from Invitrogen (CAT#EPX090-20821-901)

6.3. Methods

6.3.1. Cell Viability Assessment

3000 cells were seeded in 90µL of media per well in 96-well plate. The plate was incubated overnight at 37°C, 5%CO_2. The next day, 10µL of the different concentrations of GC were added in each well in triplicate per time point, giving a total of 9 wells per cell line per concentration of GC. Blank wells were also prepared with only media and GC at the different concentration in triplicate for each time point. After 24h,72h or 1 week, 10µL of CCK8 was added in each well, then the plate was incubated for 1 hour. After 1 hour, the plate was agitated for a minute to homogenize the color then read with a microplate reader at 450nm. Every sample absorbance value was subtracted with the absorbance of the corresponding blank. This equation was then applied to the values:

$$\text{Cell viability (\%)} = \frac{As - Ab}{Ac - Ab} * 100$$

As = absorbance of the experimental well
Ab = blank well absorbance
Ac = control well absorbance
Every negative value was set at 0 because a negative viability does not represent a biological reality.
The statistical analysis was performed using GraphPad Prism. A two-way ANOVA with multiple comparison was performed to evaluate the statistical significance (α=0.05).

6.3.2. Evaluation of the chemokine production by cancer cells by Multiplex

The multiplex was performed by OUHSC following the manufacturer instruction. No statistical analysis was performed on the dataset due to lack of biological repetition. Those results can be confirmed later by ELISA.

6.4. Results

6.4.1. GC is nontoxic for cancer cells, monocyte and B-cells

The potential direct effect of GC on cancer cells was tested. First, GC's toxicity was assessed by cell viability. Breast cancer, pancreatic cancer and melanoma cells were incubated with different GC concentrations. A broad spectrum of concentrations and 3 different timepoints were assessed to obtain a general idea of how the cells react to GC. The results can be observed on the (**Figure 12**).

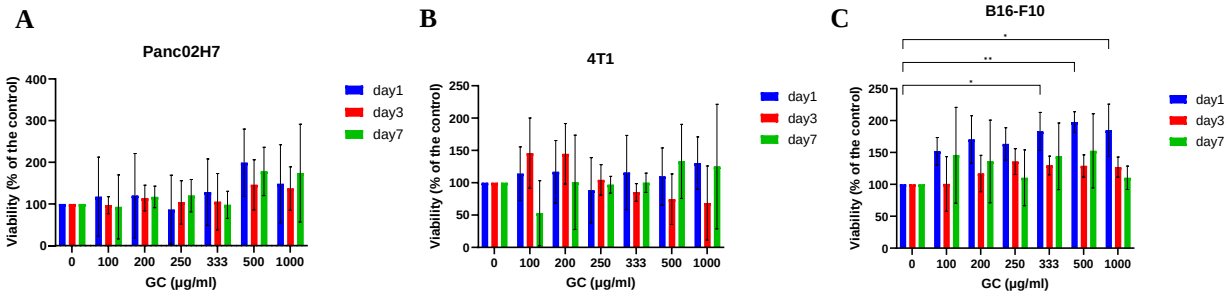

Figure 12. GC cytotoxicity assessment on 3 cancerous cell lines. (A)Panc02H7 is a mouse pancreatic cell line, (B) 4T1 a mouse breast cancer cell line and (C) B16-F10 mouse melanoma. *p: <0.05, **: 0.005 non-significant comparisons are not drawn.

The control (0µg/mL GC) was set at 100% viability for each timepoint, and all the values were scaled accordingly. For Panc02H7 on day 1, we first observed a small increase of the viability until 250µg/mL. At 250µg/mL, the viability decreased under 100% and then the viability increased above 100% for the higher concentrations. On day 3, the viability increased with the concentration of GC but not as much as on day 1. On day 7, the viability is lower than 100% for 100, 250 and 333µg/mL of GC but increased two times the control for the two highest GC concentration. 4T1

viability trends seem constant when we look at the overall graph, but in detail, the viability is very low for 100µg/mL GC on day 7 then increased until being higher than 100% for 500 and 1000µg/mL GC. On day 3, the viability decreased with the increase of GC concentration. For B16-F10 viability, it increased at least 50% on day 1 for all GC concentrations and 10% on day 3. On day 7, the viability increased by 50% for 100µg/mL GC and decreased until a little less than 100% for 250µg/mL and increase again above 100% for the highest concentrations.

None of the viability variability are statistically different from the control so we can hypothesize that these are due to biological variability between repeats and overall, GC is not toxic for these cancer cell lines. This observation can be correlated to the previous survival studies performed with GC where the tumor growth and the survival rate of the group of mice treated only with GC were not significantly different from the control[3].

GC's cytotoxicity was previously tested on dendritic cells but not on other immune cell types potentially recruited at the tumor site by the APCs. The viability of monocytes and B cells in the presence of GC was also assessed (**Figure 13**).

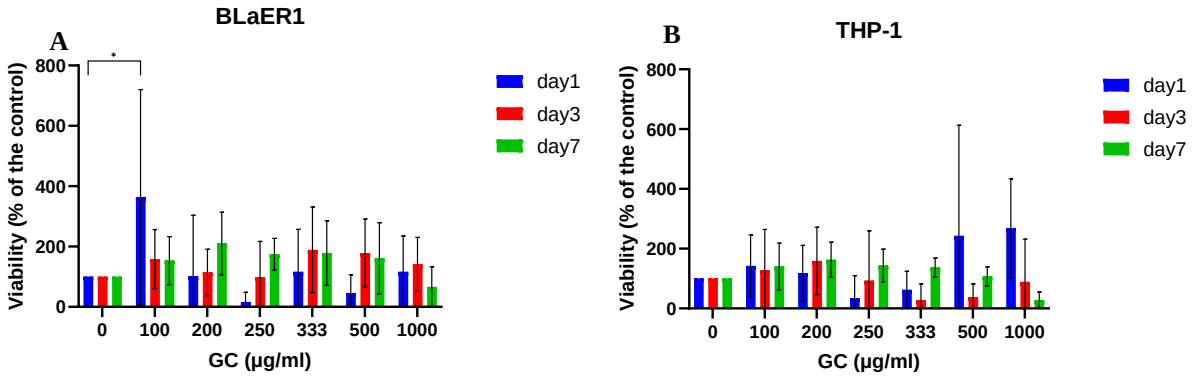

Figure 13. GC cytotoxicity assessment on 2 immune cell lines. BLaER1 is a human B-cell line, THP-1 human Monocyte. *p<0.05, non-significant comparisons are not drawn.

For BlaER1, the viability significantly increased on the day 1 with 100µg/mL GC, then it decreased at 100% or lower for the other GC concentrations. On day 3, the viability is higher than the control for all GC concentrations, with a slight decrease between 100 and 250µg/mL GC and another decrease between 333 and 1000µg/mL GC. On day 7, the viability increased slightly with GC concentration until 250µg/mL GC and then decreased until being under the 100% reference at 1000µg/mL GC. Overall, these variations are not significantly different from the control so it might only be biological variations between the repeats with an overall viability unchanged with GC, with the exception of day 1 100µg/mL GC.

An increased viability is hard to interpret because this assay does not allow us to precisely quantify the growth rate or the number of cells after treatment, it might just be the upregulated metabolites reacting with the cell counting reagent.

For THP-1, on day 1, the viability increased near the 200% for 100µg/mL GC and decreased until reaching around 50% for 250µg/mL and then increased with GC concentration, reaching 300% for

500 and 1000µg/mL GC. On day 3, the viability increased above the control for 100 and 200µg/mL GC, decreased under 100% between 250 and 500µg/mL and increased again to 90% for 1000µg/mL. On day 7, the viability was stable around 100-110% for the concentrations of GC between 100 and 333µg/mL and decreased under the 100% for the two highest GC concentrations. Overall, these variations are not significantly different from the control so it might only be biological variations between the repeats with an overall viability unchanged with GC.

Then, we evaluated the chemokine release by the same cancer cells by multiplex.

6.4.2. GC can influence cancer cells' chemokine release

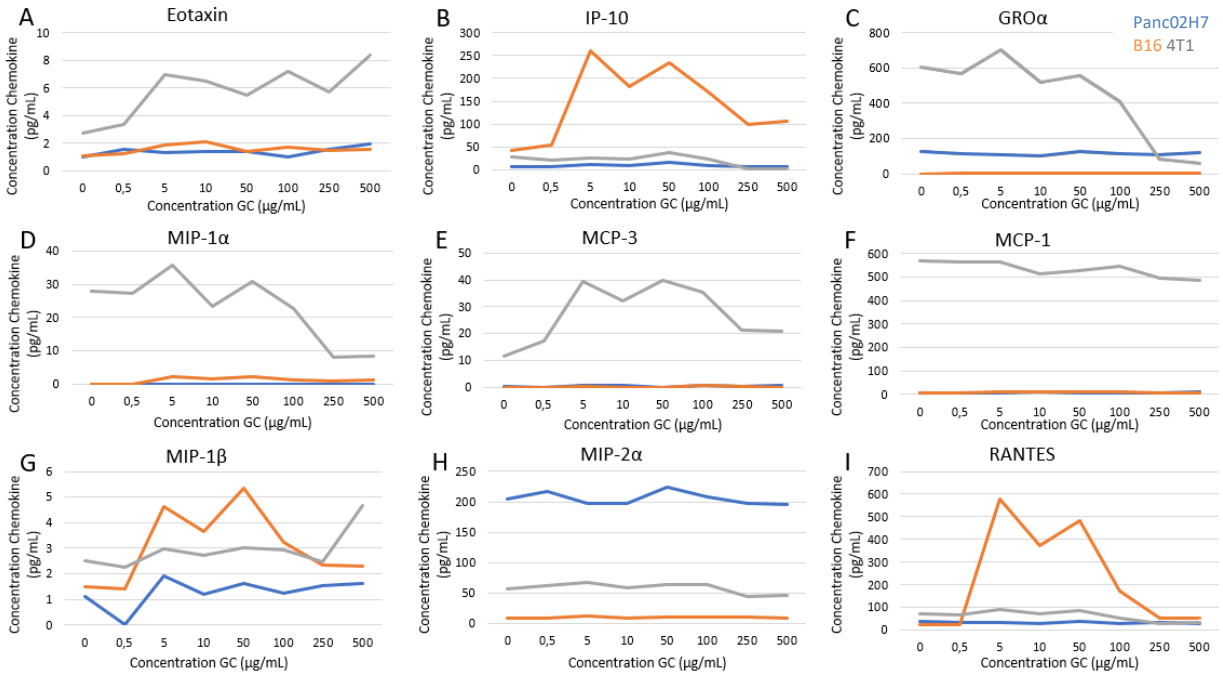

Figure 14. Regulation of chemokines production in cancer cell lines by GC. Panc02H7 (Blue) is a mouse pancreatic cancer cell line, 4T1 (Gray) mouse breast cancer and B16-F10 (Orange) a mouse melanoma. A: Eotaxin (CCL11), B: IP-10 (CXCL10), C: GROα (CXCL1), D: MIP-1α (CCL3), E: MCP-3 (CCL7), F: MCP-1 (CCL2), G: MIP-1β (CCL4), H: MIP-2α (CXCL2), I: RANTES (CCL5). No statistical analysis was performed due to a lack of biological repetition.

As we can see on **Figure 14**, not every chemokine evaluated was affected by GC. First, we can observe that B16-F10 and Panc02H7 don't produce MCP-3, MIP-1α and MCP-1 and Panc02H7 doesn't produce MIP-2α. Secondly, for Panc02H7 (Blue line), MIP-1β was inhibited with 0.5µg/mL GC and then its concentration slightly increased for higher GC concentration. For B16-F10 (Orange line), The evolution of the chemokine concentration according to GC concentration is similar for IP-10 (CXCL10), MIP-1β (CCL4) and RANTES (CCL5). It is increased between 0.5 and 100µg/mL GC, with a small decrease at 10µg/mL, and even after 250µg/mL GC, the evolution of the chemokine concentration remains constant but higher than without GC. The maximum concentration of chemokine released is at 5µg/mL GC for IP-10 and RANTES, and at 50µg/mL GC for MIP-1β. Finally, for 4T1 (Gray), Eotaxin and MCP-3 production are increased with the

increased concentration of GC, and MIP-1α and GROα concentration decreased with GC concentration decreased increasing. MCP-1 and MIP-2α productions are not affected by GC. All of these variations of concentrations enhance tumor inhibition of growth, except MCP3 high expression enhance the tumor growth in breast cancer.

6.5. Conclusion

GC can be considered safe for B cells and monocytes because it does significantly reduce their viability, but also the viability of the tested cancer cell lines is not reduced by GC, correlating with the *in vivo* tumor growth and survival studies previously performed.

Chapter 7. GC can activate the inflammatory state of nasal epithelial cells

7.1. Objectives

GC's mechanism of action to increase the immune response is well known. However, we want to extend our knowledge of its mechanism to further enhance cancer therapy and antiviral effects. A mucosal adjuvant is more efficient if it can also activate the reaction of the epithelial cells forming the mucosa in contact with the intra-nasal vaccine formulation. Therefore, the potential antiviral effects GC can have on these epithelial cells are being studied. First, we look into the potential toxicity GC can cause to the cells. We also look for any morphology and activation profile change that can indicate a form of reaction from the epithelial cells when in contact with GC. The activation profile we look for is the regulation of the inflammatory receptors present on the cell membrane and if there is a communication between the epithelial cells and DCs that will activate DCs inflammatory state. These results will give us some indication on how GC's immunological mechanism is applied to the mucosa and how it can enhance cancer therapy and antiviral effects.

7.2. Materials

The RPMI 2650 cell line (EC) was purchased from ATCC (Human nasal septum Squamous Cell Carcinoma CCL-30) and THP-1 Human Monocyte (ATCC TIB-202) were used for the co-culture protocol.

Aqueous solution of N-dihydrogalactochitosan, also referred to as glycated chitosan (GC), (10mg/mL) was provided by ImmunoPhotonics, Inc.

A GC-FITC aqueous solution (10mg/mL) was provided by ImmunoPhotonics.

The Cell Counting Kit 8 (CCK8) was purchased from Dojindo Laboratories (CAT#CK04-13). The absorbance was measured using the BioTek Synergy Neo2 Multimode Reader.

The imaging was performed using the Leica SP8 confocal microscope. The membrane staining was realized using wheat germ agglutinin (WGA) conjugated with CF®405S (stock at 1mg/mL) (Biotium, CAT#29027-1).

The monocyte differentiation was induced with Human recombinant IL-4 (Stemcell Technologies, CAT #78045) and Human recombinant GM-CSF (Stemcell Technologies, CAT #78015).

Here is the list of Antibodies used to stain the cells for the Flow Cytometry:

- Brilliant Violet 605™ anti-human CD11b Antibody (Biolegend, CAT#301332)
- PE Annexin V (BD Pharmingen™, CAT#556421)
- Pacific Blue™ anti-human CD326 (EpCAM) Antibody (Biolegend, CAT#324217)
- Alexa Fluor® 700 anti-human CD206 (MMR) Antibody (Biolegend, CAT#321131)
- PerCP/Cyanine5.5 anti-human HLA-DR, DP, DQ (MHC II) Antibody (Biolegend, CAT#361709)

- APC/Cyanine7 anti-human CD83 Antibody (Biolegend, CAT#305329)
- PE/Dazzle™ 594 anti-human CD11c Antibody (Biolegend, CAT#301641)
- FITC Anti-Human CD40 (G28.5) (TONBO, CAT#35-0410)
- APC anti-human CD80 Antibody (Biolegend, CAT#305220)
- PE/Cyanine7 anti-human CD86 Antibody (Biolegend, CAT#374210)
- Ghost Dye™ Violet 510 (TONBO, CAT#13-0870)
- Human Trustain FcX (Fc receptor Blocking Solution) (Biolegend, CAT#422301)

The Staining buffer is a mix of 5mL 0.5M Ethylenediaminetetraacetic acid (EDTA), 0.5g Sodium Azide (NaN_3) and 20mL of Fetal Bovine Serum (FBS).

The Annexin Buffer is a mix of 5mL 1M HEPES, 14mL 5M NaCl, 125µL 1M $NaCl_2$ in 50mL milli-Q Water (recipe for 10X Annexin Buffer, dilute in milli-Q water to obtain 1X buffer to use for the staining protocol). The flow measurement was performed using the Cytek™ Northen Lights and the analysis using the software FlowJow™ V10.10.

7.3. Methods

7.3.1. Microscopy Imaging

Approximately 5.10^6 EC were collected in media a 15mL tube. WGA staining was added at a final concentration 3µg/mL. The tube was covered in aluminum foil and incubated for 10min at room temperature. The cells were then centrifuged at 1200rpm, 5min. The supernatant was discarded, and the cells were resuspended in 5mL fresh media. This washing step was repeated 3 times. The cells were then resuspended in 5mL fresh media, and 4µL of the GC-FITC stock was added (final concentration 8µg/mL). The cells were incubated for 18h at 37°C, 5%CO_2. After 18h, The cells were washed 2 times, and a drop of cells was placed on a glass slide with a cover slide for imaging. A control was prepared with only the cells stained with WGA without GC-FITC.

The imaging was performed using a confocal microscope under Dr. Gu's supervision.

The argon laser was used to excite the WGA for the membrane dye with the emission detection window set between 415 and 487nm. The 450 Diode laser was used to excite the FITC conjugated to GC, the emission detection window was set between 498 and 650nm. The objective used was a 63 x - 1.40 NA oil. The PMT Transmitting detector was turned on to get an image of the cells with the transmitting light. A z-stack was performed with a 0.2µm step size.
The image analysis and orthogonal section was performed using the software Fiji.

7.3.2. Cell Viability Assessment

Approximately 3.10^3 EC were seeded in 90µL of media per well in 96-well plate. The plate was incubated overnight at 37°C, 5%CO_2.

The next day, 10µL of the different concentrations of GC were added in each well in triplicate per time point, giving a total of 9 wells per cell line per concentration of GC. Blank wells were also

prepared will only media and GC at the different concentration in triplicate for each time point.

After 24h,72h or 1 week, 10µL of CCK8 was added in each well, then the plate was incubated for 1hour at 37°C 5% CO_2. After 1 hour, the plate was agitated for one minute to homogenize the color then read with a microplate reader at 450nm. Every sample absorbance value was subtracted with the absorbance of the corresponding blank.

This equation was applied to the results:

$$\text{Cell viability (\%)} = \frac{As - Ab}{Ac - Ab} * 100$$

As = absorbance of the experimental well
Ab = blank well absorbance
Ac = control well absorbance (Media only)
The statistical analysis was performed using Graph Pad prism. A two-way ANOVA with multiple comparisons was performed to evaluate the statistical significance (α=0.05).

7.3.3. Monocyte differentiation into inactivated DCs.

The protocol was designed based on previous work[103,104].

On day 1, THP-1 cells were collected and seeded in T-75 flasks at 2.10^5 cells/mL in 20mL of media supplemented with 10% FBS, 1% P/S,100ng/mL IL-4 and 100ng/mL GM-CSF to start their differentiation into inactivated dendritic cells.

On day 4, the media was collected in 50mL tube, centrifuged at 1200rpm for 5min to pellet the cells and resuspended into fresh media.

On day 6, the non-adherent and adherent cells were either collected for the co-culture (see chapter 6.3.4) or left in the flask until day 7 to be used as reference groups for the flow cytometry.

7.3.4. Co-culture

On day 4 of the differentiation, 6.10^5 RPMI 2650 cells were seeded in 2mL EMEM media per well into a 6-well following the plate map in **Figure 15**.

A 100-mm petri dish was seeded with 2.10^6 EC cells to use for the reference group for the flow cytometry.

On day 5, GC was added in the EC+GC wells at different concentrations (same concentration on 1 plate, different depending on the purpose of the repeat).

On day 6, the media was removed, and the cells washed with 2mL PBS. Then, 1.10^6 differentiated cells (iDC) were added with 2mL fresh RPMI 1650 media in the corresponding wells. The media was also changed for RPMI1650 in the A1 well. GC was added in the B2 well at the same

concentration as on day 5.

On day 7, all the cells were collected to be stained for evaluation using flow cytometry.

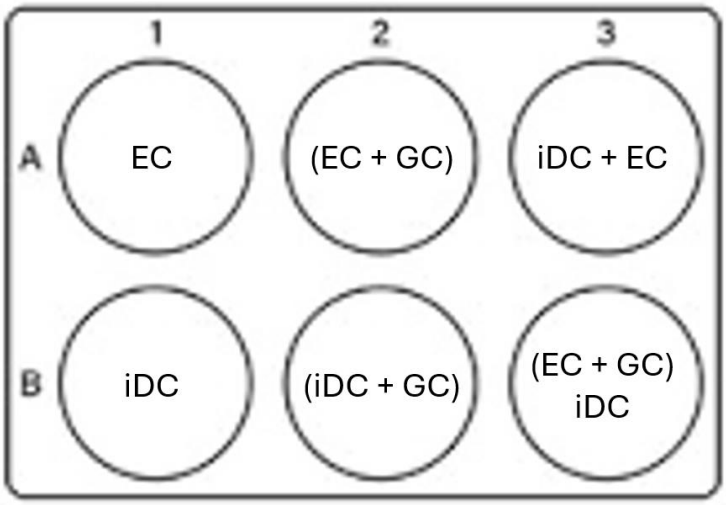

Figure 15. Co-culture plate map.

Here is a summary of the complete timeline for the THP-1 cells differentiation and the co-culture:

Table 4. Co-Culture protocol timeline.

Days	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
iDC	THP-1 in RPMI 1650 +IL-4 + GM-CSF 2.10^5 cells/mL in 20mL			Change medium		Aspirate media, wash with PBS, add iDC 1.10^5 cell/mL GC treatment	Label for flow cytometry.
EC				Seed 6-well plate 6.10^5cell	GC treatment		

7.3.5. Flow cytometry

Samples preparation

The cells of the co-culture different wells were collected into labeled 15mL tube (non-adherent in the supernatant and adherent cells detached with trypsin). Two additional tubes were prepared with a mix of 1.10^6 iDC and 1.10^6 EC each for the unstained group and the live/dead control positive.

The tubes were centrifuged at 2800rpm for 5min. The supernatant was discarded, the cells were resuspended into 1mL staining buffer, transferred into flow tube and centrifuged at 2500rpm for 5min. The supernatant was discarded and 0.5µL of Fc block was added in the remaining volume of liquid (around 100µL left in the tube due to capillarity). The cells were resuspended and incubated on ice for 20min.

During this time, the antibodies for the sample staining were mixed. 0.5µL of each antibody (except Annexin-V) was added into 100µL staining buffer.

After 20min, 100µL of the mix was added into the samples tube. The tubes were vortexed then incubated on ice for 30min in the dark.

After 30 min, 2mL of Annexin buffer was added into the tube and they were centrifuge at 2500rpm for 5min. The supernatant was discarded and 0.5µL of the Annexin-V antibody was added in the tube. The tubes were vortexed and incubated on ice for 10min.

At the end, 300µL of Annexin buffer were added into the tubes and they were vortexed prior to data acquisition.

Controls preparation

The positive staining controls were prepared with one drop of beads and 0.5µL of the tested antibody in a flow tube. These tubes were vortexed then incubated 45min on ice in the dark. 300µL of staining was added after the incubation and the tubes were vortexed one last time before the flow measurement.

The unstained control was prepared with a mix of 1.10^6 EC and 1.10^6 iDC in a tube. The cells followed the same preparation step as the samples except the adding of the antibodies.

The live and dead control was also prepared with 1.10^6 EC and 1.10^6 iDC in a tube, washed with 1mL staining buffer, then half of the cells were separated in another tube with ethanol for 5min to provoke their death. The dead were washed with staining buffer and centrifuged at 2500rpm for 5min then transferred back in the live/dead tube for the Fc block staining (step similar to the samples preparation). Then 0.5µL of the Ghost Dye antibody was added in the tube and the cells were vortexed and incubated 30min on ice in the dark. 300µL of staining buffer was finally added in the tube and vortex for the flow measurement.

The Annexin V positive control was prepared using a mix of 1.10^6 EC and 1.10^6 iDC, half dead cells and half live cells prepared in a similar way to the live/dead control in a tube. The cells were centrifuged and resuspended in annexin buffer, then stained with 0.5µL Annexin-V antibody, vortex and incubate 30min on ice in the dark. 300µL of annexin buffer was added for the flow measurement.

The flow measurement was performed following the manufacturer instruction, the number of positive events out of the 50000 events red by the machine was counted (Cellularity). The

statistical analysis was performed using one-way (for epithelial cells inflammation marker) or two-way ANOVA.

7.4. Results

7.4.1. GC is not toxic for Nasal Epithelial cells

The cytotoxicity was assessed on a broad range of GC concentration and timeline. The control (0µg/mL GC) was set at 1 (100% viability) and all the values were scaled accordingly (**Figure 16**).

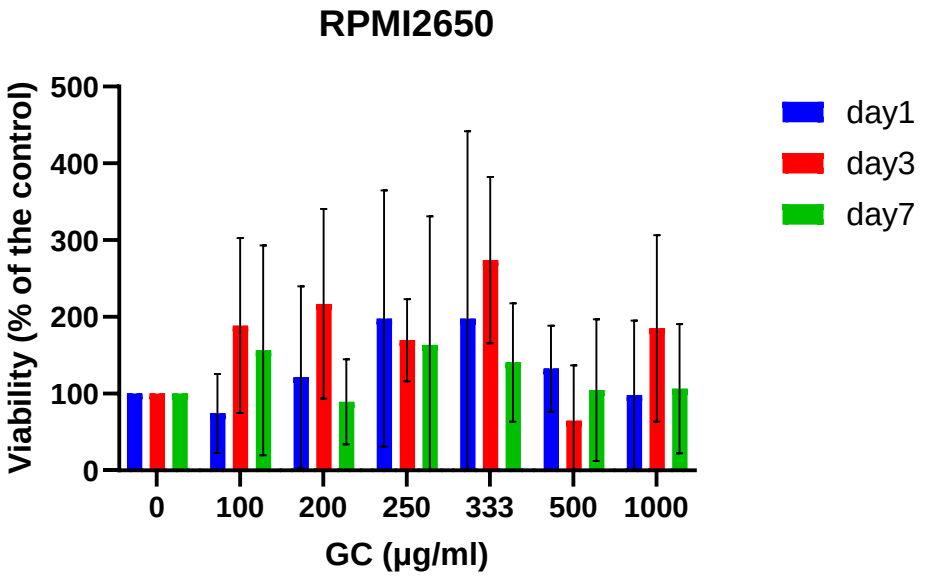

Figure 16 Nasal Epithelial cell viability is not affected by GC. *p<0.05, non-significant comparisons are not drawn.

On day1, the viability is decreased to 50% for 100µg/mL GC and increased to 100% for 250µg/mL GC, decreased again to 60% for 333µg/mL GC and increased with GC increased concentrations. On day 3, the viability is above 100% for all GC concentrations except 500µg/mL. On day 7, the viability variates up and down around 100% with a peak at 250µg/mL GC. These viability variations are not statistically different from the control, so they might represent little biological variations between the different repeats of the experience.

These results are encouraging because we do not want to cause any trouble to the person we want to vaccinate. Disrupting the integrity of the mucosa might cause the entrance of unwanted viruses or bacteria in the system and cause infections or other side effects. A decreased viability of the EC is not the only factor that can cause side effects, but it is a first preliminary good indicator of GC's safety.

7.4.2. Observation of epithelial cells' morphology by confocal microscopy

We also observed the morphology and localization of GC when it is in contact with EC for 18h.

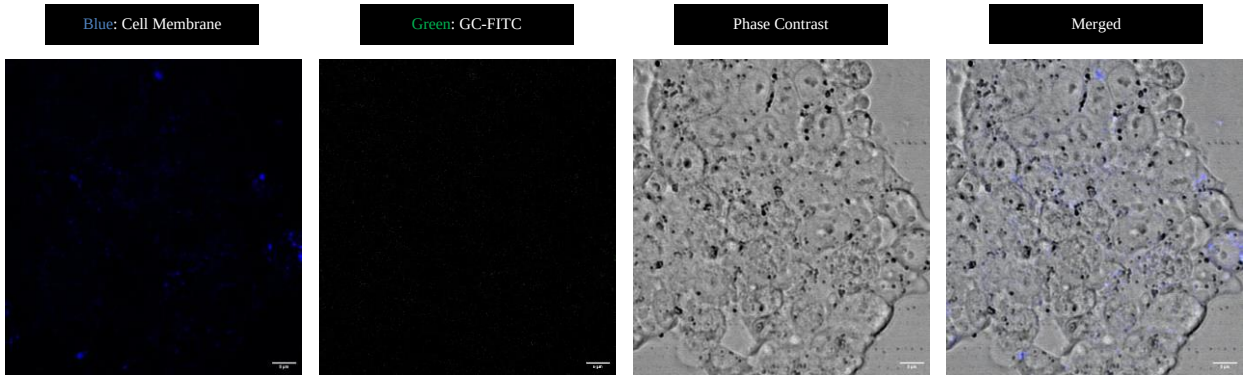

Figure 17 Confocal imaging RPMI6250 morphology in absence of GC. RPMI2650 were stained with WGA405S to color the cell membrane in Blue, the phase contrast channel was turned on to get a better visual of the cell morphology.

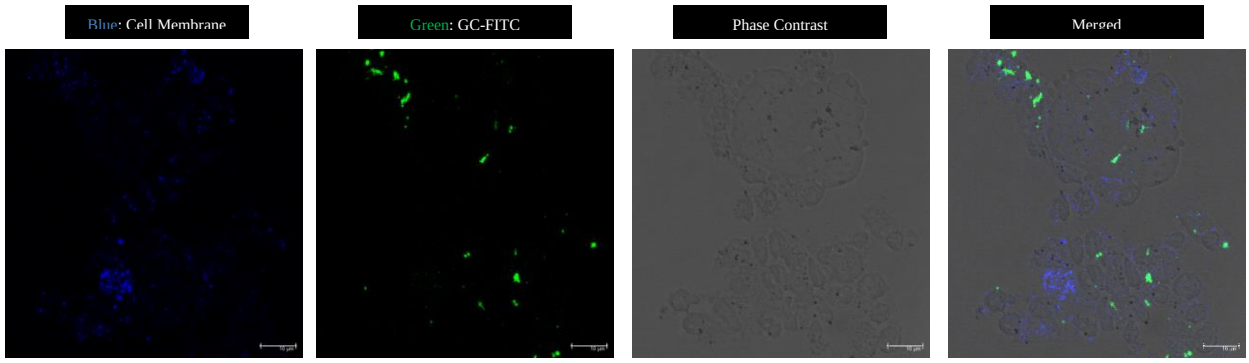

Figure 18 Confocal imaging of RPMI2650 in presence of GC-FITC. RPMI2650 were stained with WGA405S to color the cell membrane in Blue, GC was associated with FITC to localize it in green, the phase contrast channel was turned on to get a better visual of the cell morphology. White arrows points out what is suspected to be a stress reaction from the cells.

If we compare the morphology of the cells with and without GC(**Figure 17** and **Figure 18**), we can see that, without GC, the cell keeps a very spheric shape and no vesicles are visible around the cell membranes. Whereas, with GC, the cells are surrounded by vesicles, suspected to be apoptosis vesicles, and the shape is not particularly different from the cells not in contact with GC. It might be because all the cells are connected to each other, so the shape of the membrane is conditioned by the inter-cell connections. When we look at an orthogonal section of the groups of cells with GC (
Figure 19), we can distinguish GC's signal coming from inside the cell, as we can observe the cell membrane surrounding the signal in both XZ and YZ section.

We look into stress markers caused by GC using flow cytometry and the Annexin V staining (**Figure 20**). The group Ghost Dye- Annexin V+ groups the live stress cells. We can observe a small number of stress cells but no significant difference with and without GC so we can't validate our apoptotic vesicle hypothesis from the confocal image.

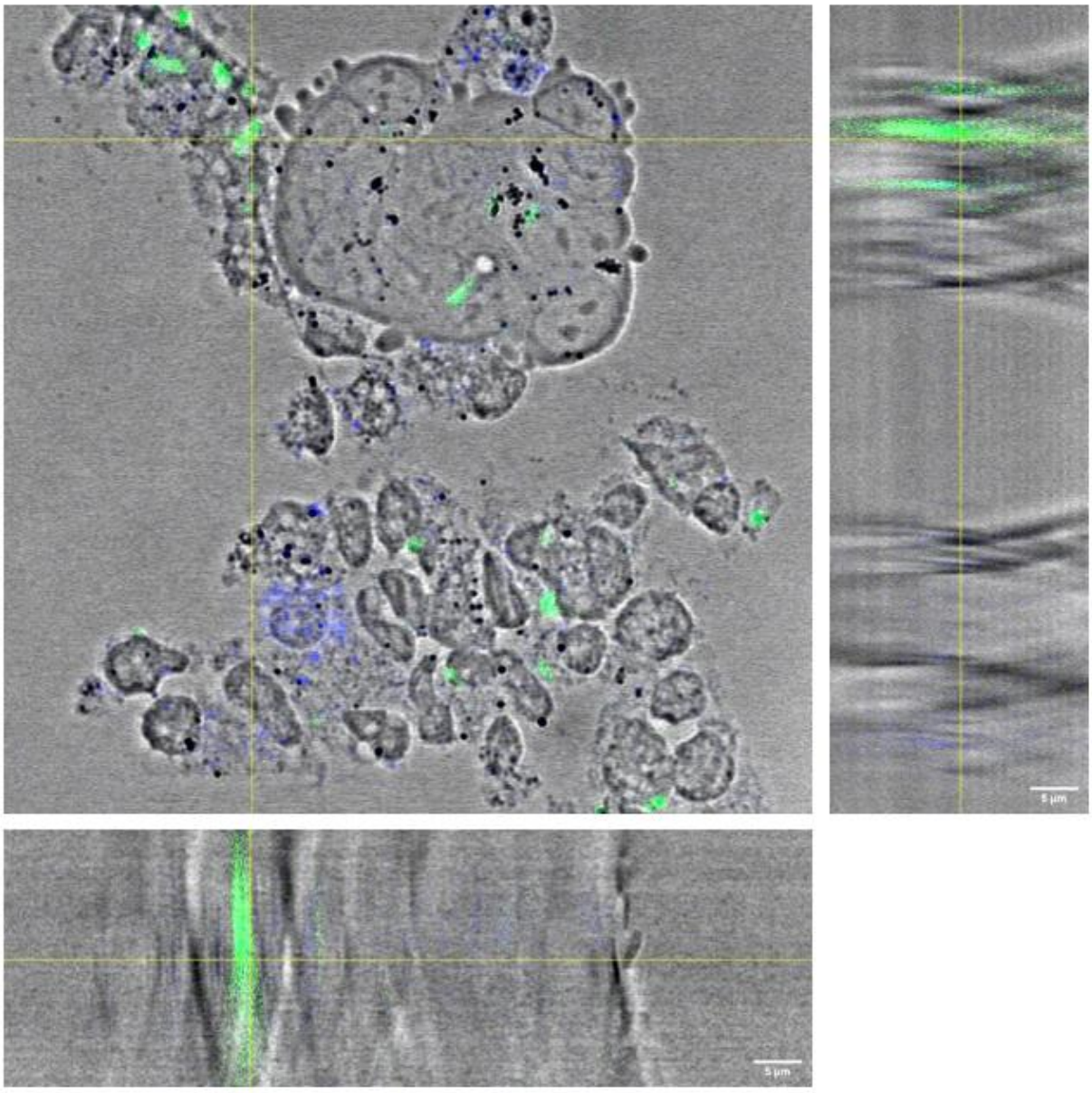

Figure 19. Orthogonal Section of RPMI2650 in presence of GC-FITC. The localization of GC can be visualized relative to the cell membrane (appearing in black with the phase contrast channel).

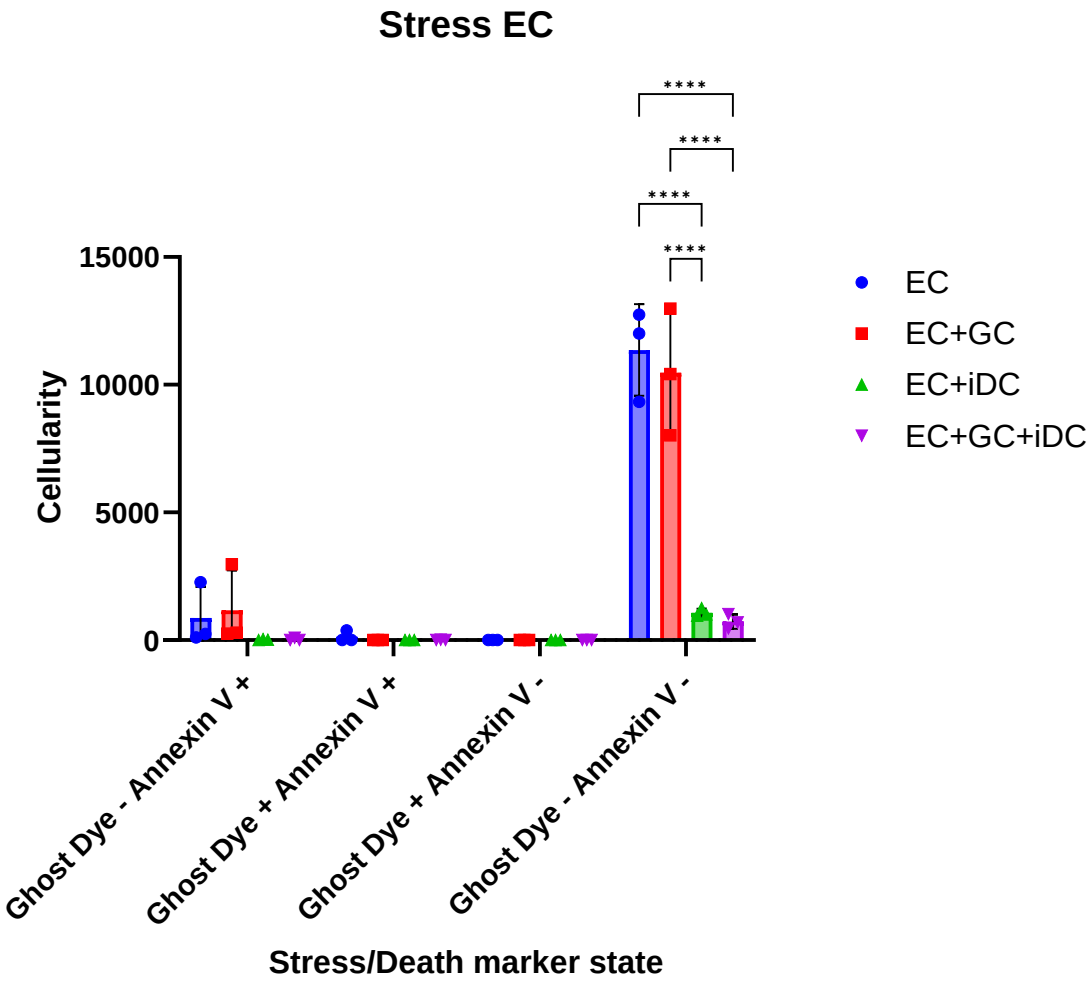

Figure 20. Graph proportion of apoptotic cells in each group. EC: Epithelial cells, DC: dendritic cells, GC: EC treated with 50µg/mL glycated chitosan. Ghost Dye-, Annexin V+: Apoptosis; Ghost Dye+, Annexin V+: Dead cells; Ghost Dye+, Annexin V-: Dead cells; Ghost Dye-, Annexin V-: Live cells. Two-way ANOVA performed, $p<0.0001$

7.4.3. Expression of inflammatory activation markers at the surface of epithelial cells and dendritic cells profile.

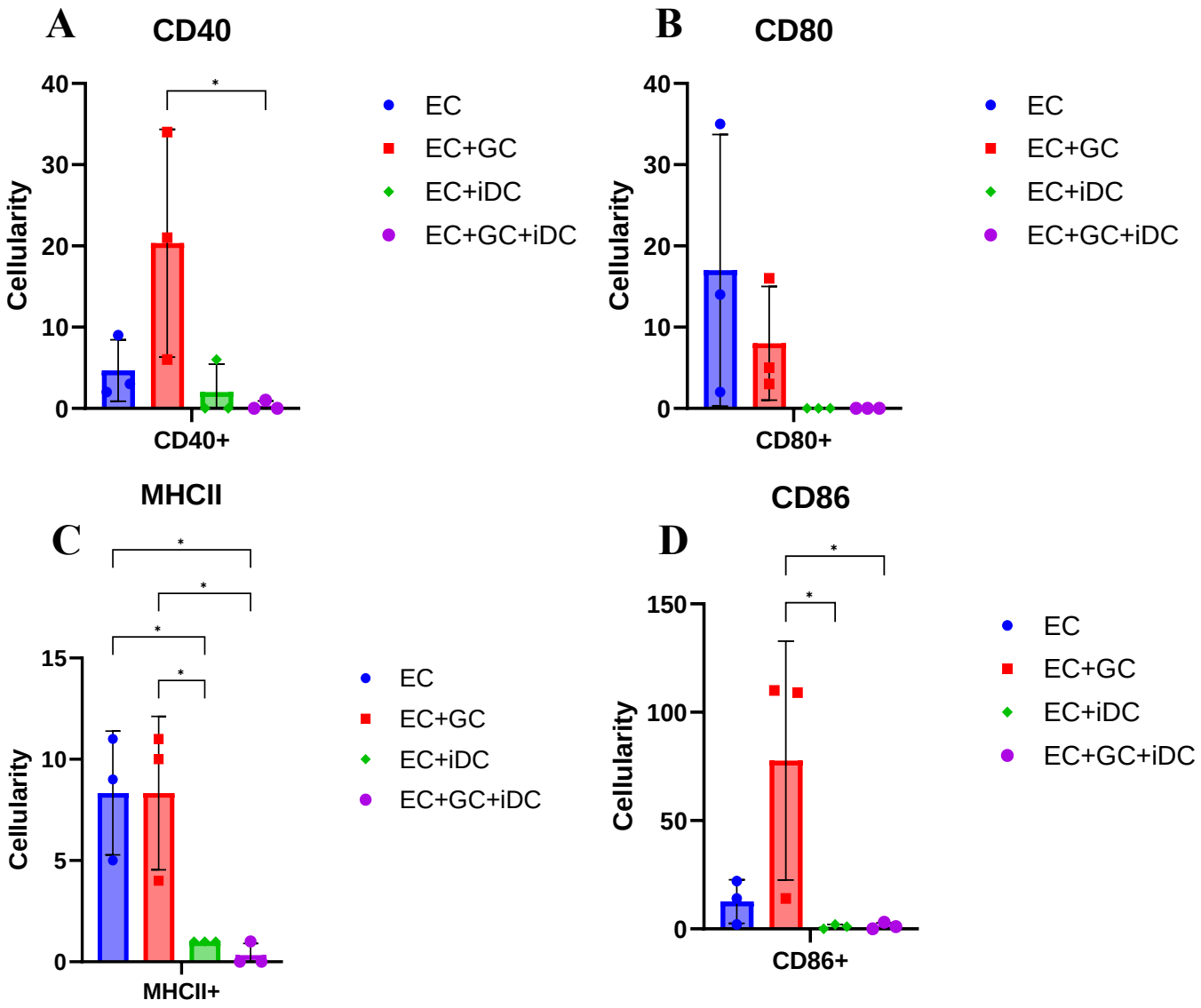

Figure 21 Activation of maturation markers on Epithelial cells (EpCAM+). EC: Epithelial cells, DC: dendritic cells, GC: EC treated with 50µg/mL glycated chitosan. A: CD80 expression, B: CD40 expression, C: MHC II expression, D: CD86 expression. One-way ANOVA performed, *: $p<0.05$

As we can see on **Figure 21**, we don't have significant change in the inflammatory receptors' expression with and without GC. But we also observe a significant decrease when the cells are in co-culture with dendritic cells. However, if we report these expressions as a percentage of the total population of EC (**Figure 22**), we obtained a significant increased expression of CD86 on ECs in presence of GC and a decrease in presence of DCs. These interpretations need to be assessed carefully due to an error in the experimental design. Indeed, the co-culture was realized with a higher concentration of DCs compared to the number of ECs, which is not representative of a

biological reality. The reference used for the protocol was seeding ECs and DCs with a proportion of 5 ECs for 1 DCs. Also, if we compare the raw numbers of each biological repeats, the third measure detected a higher number of ECs compared to the others biological repeats, which influence greatly the standard deviation on **Figure 21**.

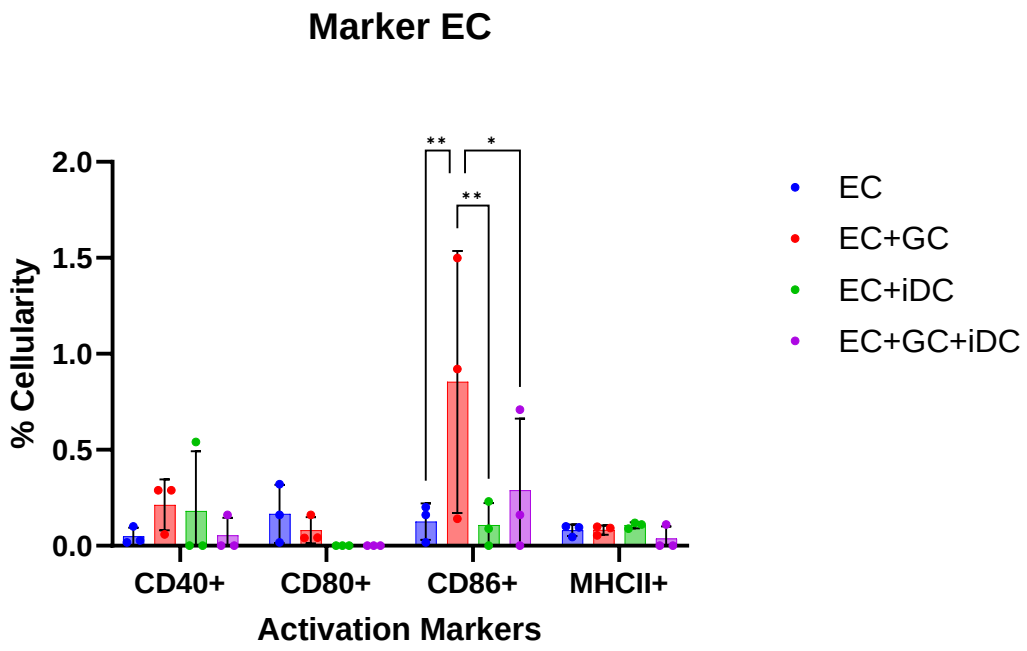

Figure 22. Percentage of EC population expressing inflammatory markers. EC: Epithelial cells, DC: dendritic cells, GC: EC treated with 50µg/mL glycated chitosan. A: CD80 expression, B: CD40 expression, C: MHC II expression, D: CD86 expression. Two-way ANOVA performed, *: $p<0.05$, **: $p<0.005$.

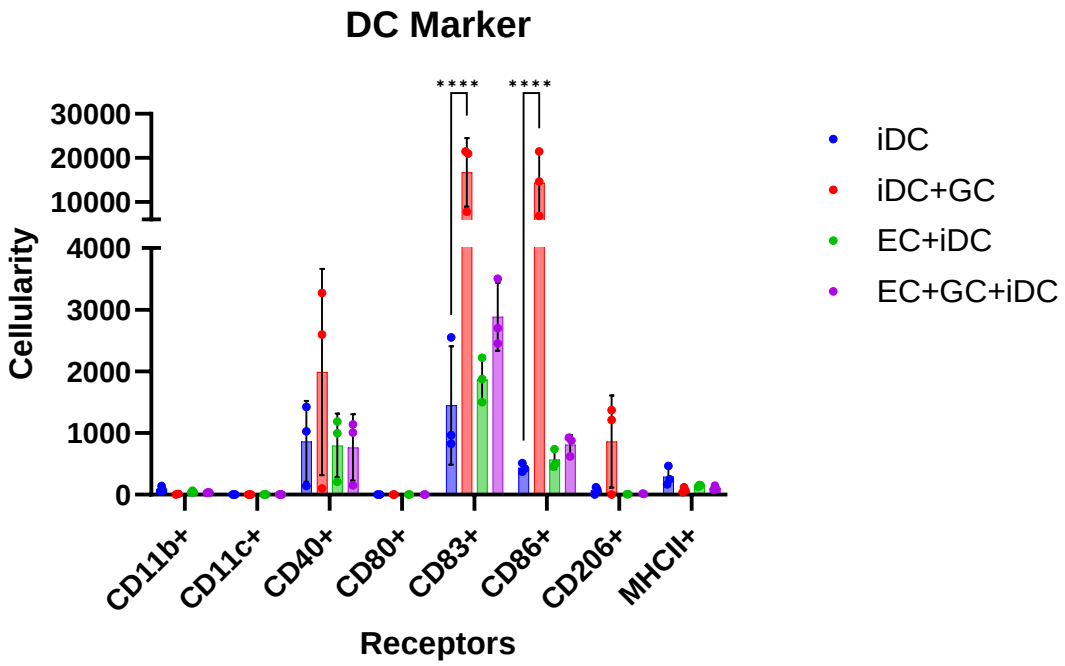

Figure 23. Maturation Marker present on the dendritic cells (EpCAM -) in co-culture with epithelial cells. EC: Epithelial cells, DC: dendritic cells, GC: EC treated with 50µg/mL glycated chitosan. ****: $p<0.0001$ two-way ANOVA was performed.

Figure 23 assess the expression of different markers on the surface of DCs and non-differentiated monocytes (EPCAM- cells). With GC, we can observe a significant upregulation of CD83, the activation marker of DCs, and CD86, similarly to ECs.

7.5. Conclusion

GC does affect the inflammatory profile of nasal epithelial cells. It can be uptake by those cells but doesn't affect their viability significantly. Also, there is no significant proof of GC induced apoptosis of these cells, so the vesicles observed by microscopy are still unidentified. The hypothesis of culture contamination was assessed but no mycoplasma was detected in the cell culture of the culture medium. GC can also increase the expression of CD86 on epithelial cells which can provide a T cells activation profile to the ECs. This interaction can further enhance the adaptive immune response to a viral or cancerous challenge in the environment of the mucosa.

Chapter 8. Discussion and Future Directions

Phototherapies are still underexplored in clinical trials. As the number of preclinical studies continues to rise, it is essential to deepen our focus on these treatments in clinical settings to better understand how the human body responds to them. Although these three phototherapies each have advantages and limitations, their targeting potential positions them as promising candidates for the next generation of cancer therapies.

Cancer cells are not the only components of a tumor. They exist within a microenvironment that includes fibroblasts, tumor-associated immune cells, and the extracellular matrix. These microenvironmental cells also contribute to chemokine communication and play critical roles in tumor growth and the establishment of an immune-suppressive environment around the tumor. Studying the effects of GC on these surrounding cells could offer valuable insights. Tumor-associated cells may be converted to an inflammatory state by GC, similar to the immune cells recruited to the tumor site during treatment. Fibroblasts influence tumor progression through their production of receptors, cytokines, and chemokines, so it would be worth investigating how GC impacts these cells. Chemokines are downstream effectors in signaling pathways within cells, making it particularly relevant to explore how GC might regulate cytokine and chemokine-associated pathways in cancer cells.

Nasal epithelial cells also express receptors involved in pathogen detection, such as toll-like receptors. Investigating the regulation of these receptors following treatment with GC could provide further understanding of GC's potential as an immune adjuvant. Additionally, deeper exploration of the pathways regulating inflammatory receptors could be informative. Other dyes, which highlight cell death-associated vesicles under the microscope, can be used to identify specific features observed using confocal microscopy.

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Supplemental Materials

Supplement Table 1 List of Clinical Trials.

NCT Number	Study Title
PDT	
NCT02698293	PDT Plus Vitamin D3 for Anal Dysplasia
NCT01966120	Safety and Efficacy Study for the Field-directed Treatment of Actinic Keratosis (AK) With Photodynamic Therapy (PDT)
NCT02799082	Evaluation of Efficacy and Safety of BF-200 ALA Used With Photodynamic Therapy in Patients With Actinic Keratosis.
NCT02281682	IM Versus 5-FU Versus IMI Versus MAL-PDT in Treatment of Actinic Keratosis
NCT00472108	Photodynamic Therapy (PDT) With Metvix Cream 160 mg/g Versus PDT With Placebo Cream in Patients With Primary Nodular Basal Cell Carcinoma
NCT01556009	Trial Comparing the Effects of Intermittent Vismodegib vs. PDT in Patients With Multiple Basal Cell Carcinomas
NCT02872909	Randomized Comparison of Low and Conventional Irradiance PDT for Skin Cancer
NCT00926952	Short Incubation Methylaminolevulinate Photodynamic Therapy Without Occlusion
NCT00028782	EF5 in Detecting Oxygen Level and Blood Vessels in Tumor Cells of Patients Undergoing Photodynamic Therapy for Intraperitoneal or Pleural Cancer
NCT03571282	Safety and Effectiveness of Intravitreal Conbercept for Exudative Circumscribed Choroidal Haemangioma
NCT03322293	Daylight Photodynamic Therapy for Actinic Keratosis
NCT00028405	Photodynamic Therapy System for Patients With Refractory/Unresponsive Solid Tumors
NCT00002963	Photodynamic Therapy in Treating Patients With Skin Cancer
NCT02631863	Aminolaevulinic Acid Photodynamic Therapy for HPV+ Low Grade Cervical Intraepithelial Neoplasia (LSIL;CIN1)
NCT05374915	Efficacy and Safety Study of REM-001 Photodynamic Therapy for Treatment of Cutaneous Metastatic Breast Cancer (CMBC)
NCT00218868	Cytodiagnosis of Basal Cell Carcinoma and Actinic Keratosis Using Papanicolaou and May-grunwald-giemsa Stained Tissue Smear
NCT01292668	Photodynamic Therapy Using Methyl-5-Aminolevulinate Hydrochloride Cream in Determining Pain Threshold in Patients With Skin Cancer
NCT00060268	Photodynamic Therapy Using HPPH in Treating Patients With Obstructive Esophageal Tumors
NCT01506115	Safety and Efficacy of Photodynamic Therapy for Bile Duct Invasion of Hepatocellular Carcinoma
NCT02916745	Feasibility Study of Using Navigational Bronchoscopy to Perform PDT-Photofrin® in Unresectable Peripheral Lung Cancer
NCT00068068	Photodynamic Therapy With Talaporfin Sodium (LS11) in Treating Patients With Refractory Colorectal Liver Metastases

NCT05363826	Intracavitary Photodynamic Therapy as an Adjuvant to Resection of Glioblastoma or Gliosarcoma Using IV Photobac®
NCT02639117	Photodynamic Therapy and Vismodegib for Multiple Basal Cell Carcinomas
NCT00469417	Metvix PDT Versus Cryotherapy in Patients With Primary Superficial Basal Cell Carcinoma
NCT02451579	A Clinical Trial Evaluating the Role of Systemic Antihistamine Therapy in the Reduction of Adverse Effects Associated With Topical 5-aminolevulinic Acid Photodynamic Therapy
NCT04429139	Photodynamic Therapy With Visudyne for Human Retinoblastoma: A Preliminary Study
NCT02018679	Er:YAG Ablative Fractional Laser Assisted-Photodynamic Therapy Versus Photodynamic Therapy for Basal Cell Carcinoma
NCT01475071	Intra-individual Comparison of Efficacy and Safety of Metvix® Natural Daylight Photodynamic Therapy Versus Conventional Metvix® Photodynamic Therapy in Subject With Mild Actinic Keratoses
NCT02955771	Efficacy and Safety Study of PDT Using Deuteporfin for Unresectable Advanced Perihilar Cholangiocarcinoma
NCT05736406	A Dose-escalation Clinical Study of Intraoperative Photodynamic Therapy of Glioblastoma
NCT02464709	Daylight PDT for Actinic Keratoses: a Multicentre Study Comparing Two Photosensitizers (BF-200 ALA Versus MAL)
NCT01303991	Hexvix Photodynamic Therapy in Patients With Bladder Cancer
NCT00784108	Optimizing Photodynamic Therapy of Cutaneous Neoplastic Diseases Via Structured Illumination and Real-time Dosimetry.
NCT02239679	Controlled Study of the Occurrence of Actinic Keratosis on the Face After Cryotherapy + Aminolevulinic Acid (ALA) Photodynamic Therapy
NCT03897491	PD L 506 for Stereotactic Interstitial Photodynamic Therapy of Newly Diagnosed Supratentorial IDH Wild-type Glioblastoma
NCT00975039	Study Using WST11 in Patients With Non-Resectable or Inoperable Cholangiocarcinoma
NCT02373371	Actinic Keratoses Treatment With Metvix® in Combination With Light
NCT00513539	Biliary Stenting With or Without Photodynamic Therapy in Treating Patients With Locally Advanced, Recurrent, or Metastatic Cholangiocarcinoma or Other Biliary Tract Tumors That Cannot Be Removed by Surgery
NCT04620239	ENdoluminal LIGHT ActivatED Treatment of Upper Tract Urothelial Cancer (ENLIGHTED) Study
NCT00054171	Photodynamic Therapy in Treating Patients With Lymphoma or Chronic Lymphocytic Leukemia
NCT00304239	Metvix PDT Versus Vehicle PDT With Aktilite CL128 Lamp in Patients With Actinic Keratosis on the Face and Scalp
NCT00005808	Photodynamic Therapy Using Lutetium Texaphyrin in Treating Patients With Cervical Intraepithelial Neoplasia
NCT02354391	Ingenol Mebutate (Picato®) With Methyl Aminolevulinate Photodynamic Therapy for the Treatment of Actinic Keratosis

NCT04429308	PDT vs Peels for AKs
NCT00253617	Stent Placement With or Without Photodynamic Therapy Using Porfimer Sodium as Palliative Treatment in Treating Patients With Stage III or Stage IV Cholangiocarcinoma That Cannot Be Removed By Surgery
NCT03511326	PRO With Luxerm® in the Field Treatment of Thin and Non-hyperkeratotic Non-pigmented AK
NCT03833726	Light Emitting Diode for the Treatment of Genitourinary Syndrome of Menopause Associated With Breast Cancer Treatment
NCT04400539	The IMmunotherapy Pleural 5-ALA PDT
NCT00025571	Photodynamic Therapy With HPPH in Treating Patients With Non-Small Cell Lung Cancer
NCT01821391	Phase 3b Study of Metvix NDL-PDT Versus Metvix c-PDT in Subjects With Actinic Keratoses
NCT02631993	Photochemotherapy and Graft-versus-leukemia in Acute-leukemia
NCT01875393	Efficacy,Safety and Quality of Life After TOOKAD® Soluble VTP for Localized Prostate Cancer
NCT02519816	Continuous Alloreactive T Cell Depletion and Regulatory T Cell Expansion for the Treatment of Steroid-refractory or Dependent Chronic GVHD
NCT01203163	Case Analysis on Real Life Incidence of Photodynamic Therapy (PDT) Safety Outcomes
NCT05547516	Blue Laser -5ala Photodynamic Therapy (PDT) in High-Risk Non-muscle Invasive Bladder Cancer (NMIBC) Patients
NCT05688904	The Effect of Topical Imipramine on Pain and Effectiveness of Topical Photodynamic Therapy
NCT01482104	New Versus Approved Methyl-aminolevulinate Photodynamic Therapy (MAL-PDT) Regime in Basal Cell Carcinoma (BCC)
NCT01043016	Safety Study of Photodynamic Therapy Using Photocyanine Injection in Treating Patients With Malignant Tumors
NCT00472706	Trial of Excision Versus Photodynamic Therapy in the Treatment of Bowen's Disease
NCT03012009	Laser Assisted Drug Delivery in the Treatment of Superficial Non Melanoma Skin Cancer: a Randomized Controlled Trial
NCT02976727	Efficacy of Topical Calcipotriol-assisted AFL-PDT in Actinic Keratosis
NCT01872104	Safety and Efficiency of Photodynamic Therapy for Rectal Cancer
NCT01966809	Photodynamic Therapy (PDT) For Recurrent High Grade Gliomas
NCT02647151	Efficacy and Safety of Treatment of Actinic Keratoses With Photodynamic Therapy Between MAL Cream and ALA Gel
NCT03067051	Clinical Study to Assess the Safety and Efficacy of the SpectraCure P18 System
NCT02751151	Photodynamic Therapy for Prevention of Nonmelanoma Skin Cancer in Organ Transplant Recipients
NCT02867722	Daylight Photodynamic Therapy for the Treatment of Actinic Keratoses in the Northeast United States
NCT02082522	Efficacy and Safety Study of PDT Using Photofrin in Unresectable Advanced Perihilar Cholangiocarcinoma (OPUS)

NCT03320447	Long-term Efficacy of Ablative Fractional Laser-assisted Photodynamic Therapy for Treatment of Lower Extremity Bowen's Disease
NCT02367547	Superficial Basal Cell Cancer's Photodynamic Therapy: Comparing Three Photosensitizers: HAL and BF-200 ALA Versus MAL
NCT00587600	Biomarkers in Phototherapy of Barrett's Esophagus
NCT00217087	Endoscopic Therapy of Early Cancer in Barretts Esophagus
NCT00524485	Photodynamic Therapy Using Topical Aminolevulinic Acid in Treating Patients With Actinic Keratosis
NCT01675219	Fluorescence Cystoscopy and Optimized MMC in Recurrent Bladder Cancer (FinnBladder 9)
NCT00601848	Photodynamic Therapy in Treating Patients With Resectable Non-Small Cell Lung Cancer That Has Spread to the Pleura
NCT02674048	Metvix Daylight PDT in Actinic Keratosis
NCT00083785	Study of the Litx™ System Combined With Chemotherapy in Patients With Colorectal Liver Metastases
NCT01439685	Comparison of Biliary Stenting Alone vs. Stenting With Photodynamic Therapy (PDT) During ERCP
NCT02137785	Safety and Efficacy Study of Photodynamic Therapy With Levulan Kerastick + Blue Light for Actinic Keratoses on the Upper Extremities
NCT01458587	Levulan PDT Versus Vehicle for Extremity Actinic Keratoses (AK)
NCT03638622	Low-cost Enabling Technology for Image-guided Photodynamic Therapy (PDT) of Oral Cancer Cancer.
NCT02702700	Photo-induction as a Means to Improve Cisplatin Delivery to Pleural Malignancies
NCT01053000	Photodynamic Therapy With Levulan® +/- Topical Retinoid Pre-Treatment In The Treatment Of Actinic Keratoses
NCT02520700	A Comparison of White-light and Daylight Topical Methyl 5-aminolaevulinic Acid Photodynamic Therapy for Actinic Keratoses
NCT03606122	5-ALA Patch-PDT of Actinic Keratosis on the Upper Extremities
NCT00118222	High Light and Low Light Dose PDT in Glioma
NCT05359419	Comparison of Two Modes of Photodynamic Therapy for the Treatment of Actinic Keratosis on the Upper Extremities
NCT01606566	A Study to Evaluate the Safety and Efficacy of Amphinex Induced PCI of Bleomycin for Recurrent Head and Neck Cancer.
NCT00014066	Photodynamic Therapy Plus Brachytherapy in Treating Patients With Lung Cancer
NCT04842266	IlluminOss Photodynamic Bone Stabilization System for the Treatment of Impending and Actual Pelvis Fractures
NCT05519319	Safety and Efficacy of PDT vs RFA vs PDT+RFA for the Treatment of Extrahepatic Cholangiocarcinoma
NCT01148966	Aminolevulinic Acid During Surgery in Treating Patients With Malignant Brain Tumors
NCT06027619	Short Contact Protocols to Reduce Pain During 10% ALA Gel Red-light Photodynamic Therapy of Actinic Keratoses
NCT00308919	Study of Photodynamic Therapy in Patients With Prostate Cancer Following Radiation Therapy

NCT00057954	Reduced-Intensity Regimen Before Allogeneic Transplant for Patients With Relapsed Non-Hodgkin's or Hodgkin's Lymphoma
NCT00265603	Safety Study: Combined Modality Treatment for Non-Small Cell Lung Cancer
NCT01573156	Vascular Targeted Photodynamic Therapy T1a Renal Tumours
NCT00003856	Photodynamic Therapy in Treating Patients With Recurrent, Refractory, or Second Primary Head and Neck Cancer That Cannot Be Treated With Surgery or Radiation Therapy
NCT02999854	Safety and Efficacy of ATIR101 as Adjunctive Treatment to Blood Stem Cell Transplantation From a Haploidentical Family Donor Compared to Post-transplant Cyclophosphamide in Patients With Blood Cancer
NCT03757754	HPPH Photodynamic Therapy for Patients With Esophageal Cancer
NCT03589456	Establishment of a PDT Patient Registry
NCT03325803	Efficacy in Ablative Fractional Laser Assisted Photodynamic Therapy According to Ablative Depth for Actinic Keratosis
NCT00473343	Metvix PDT in Participant With "High Risk" Basal Cell Carcinoma
NCT04085367	Multicenter Study to Assess the Efficacy and Safety of Methyl Aminolevulinate Hydrochloride (MAL) 16.8% Cream (CD06809-41) Versus Vehicle Cream for Actinic Keratosis of the Face
NCT00002167	Photodynamic Therapy Clinical Trial For Cutaneous AIDS-Related Kaposi's Sarcoma Study Summary.
NCT05918783	Padeliporfin VTP Using Robotic Assisted Bronchoscopy in Peripheral Lung Cancer
NCT01812837	The Use of Microneedles in Photodynamic Therapy
NCT02124733	Protocols for Painless Photodynamic Therapy (PDT) of Actinic Keratoses
NCT01236443	Study of Photodynamic Therapy (PDT) Using HPPH in Barrett's Esophagus
NCT00155337	Photodynamic Therapy for Oral Leukoplakia and Erythroleukoplakia
NCT06052033	Comparison of 5-ALA Photodynamic Therapy and CO2 Laser for Treating Persistent Low-Grade Cervical Lesions With High-Risk HPV Infection
NCT00670397	Photodynamic Therapy Using HPPH in Treating Patients With Recurrent Dysplasia, Carcinoma in Situ, or Stage I Oral Cavity Cancer
NCT05333367	MORPHEE : Mechanisms of Cell Death Induced by Extracorporeal Photochemotherapy
NCT00243490	Photodynamic Therapy in the Treatment of Malignant Intracranial Tumors
NCT03697590	Photodynamic Therapy of Actinic Keratosis of the Face and Scalp With and Without Prior Curettage
NCT00005067	Photodynamic Therapy With Lutetium Texaphyrin in Treating Patients With Locally Recurrent Prostate Cancer
NCT05386056	Pembrolizumab and Photodynamic Therapy in Previously Treated Metastatic Esophageal Squamous Cell Carcinoma
NCT04860154	Photodynamic Therapy for Cholangiocarcinoma
NCT00308854	Photodynamic Therapy With PD P 506 A Compared With Placebo-PDT for the Treatment of AK
NCT00675233	Photodynamic Therapy Using HPPH in Treating Patients With Dysplasia, Cancer in Situ, or Invasive Cancer of the Larynx

NCT05060237	Study to Evaluate Safety and Tolerability of BF-200 ALA (Ameluz®) for Photodynamic Therapy in the Treatment of the Expanded Field of Actinic Keratosis on Face and Scalp
NCT00587314	The Effect of Ablation Therapy on Barrett's Esophagus
NCT01035437	Study for Determination of Feasibility and Toxicity of Pre-Treatment With HPPH (2-1[1-Hexyloxyethyl]-2 Devinyl Pyropheophorbide-a) Photodynamic Therapy Prior to Chemoradiation in Non-Operable Patients With Obstructive Esophageal Cancer
NCT00984243	Photodynamic Therapy (PDT) in Lung Cancer
NCT00472043	PDT With Metvix 160 mg/g Cream Versus PDT With Placebo Cream in Patients With Primary Nodular Basal Call Carcinoma
NCT03066843	Photodynamic Therapy Incubation Times for Actinic Keratosis
NCT02106559	Photodynamic Therapy During Surgery in Treating Patients With Pleural Malignancy
NCT00049959	Two Studies to Determine if Verteporfin PDT is Effective & Safe in Treating Multiple Basal Cell Carcinoma of the Skin.
NCT03564054	Trial of Photodynamic Therapy Versus Argon Plasma Coagulation for Lung Cancer With Endobronchial Obstruction
NCT02585856	Efficacy and Safety of Photodynamic Therapy for Unresectable Cholangiocarcinoma
NCT00023790	Photodynamic Therapy in Treating Patients With Skin Cancer or Solid Tumors Metastatic to the Skin
NCT00472459	PDT With Metvix® 160 mg/g Cream in Organ Transplant Recipients With Non-melanoma Skin Cancer
NCT00747903	Photodynamic Therapy Using Aminolevulinic Acid in Treating Patients With Skin Cancer
NCT00308867	PD P 506 A-PDT Versus Placebo-PDT and Cryosurgery for the Treatment of AK
NCT03110159	DUSA: Cyclic PDT for the Prevention of AK & NMSC in Solid Organ Transplant Recipients
NCT05208775	A Comparative Study of Endoscopic Submucosal Dissection and Photodynamic Therapy for Early Esophageal Cancer
NCT00002975	Photodynamic Therapy in Treating Patients With Skin Cancer
NCT00528775	Photodynamic Therapy Using HPPH in Treating Patients With Advanced Non-Small Cell Lung Cancer That Blocks the Air Passages
NCT04319159	Study to Evaluate the Safety of BF-200 ALA (Ameluz®) for Photodynamic Therapy (PDT) in the Treatment of Expanded Fields of Actinic Keratosis (AK)
NCT03678350	Light Dosimetry for Photodynamic Therapy With Porfimer Sodium in Treating Participants With Malignant Mesothelioma or Non-Small Cell Lung Cancer With Pleural Disease Undergoing Surgery
NCT00707356	Study of WST11 in Patients With Localized Prostate Cancer
NCT03133650	A Trial of a New Type of Photodynamic Therapy (VTP) in the Treatment of Patients With Cancer of the Esophagus Who Have Trouble Swallowing

NCT02500550	Safety and Efficacy of Two Doses of ATIR101, a T-lymphocyte Enriched Leukocyte Preparation Depleted of Host Alloreactive T-cells, in Patients With a Hematologic Malignancy Who Received a Hematopoietic Stem Cell Transplantation From a Haploidentical Donor
NCT01481155	Comparative Study of Photodynamic Therapy vs. CO2 Laser Therapy in Treatment of Actinic Keratoses
NCT01842555	Photodynamic Therapy (PDT) Oncology Registry
NCT00535080	Compassionate Use of Metvix® (Methyl Aminolevulinate) Photodynamic Therapy (PDT) in Subjects With Field Actinic Keratoses, Large/Multiple Superficial Basal Cell Carcinomas (BCCs), or Bowen's Disease
NCT03573401	Study to Evaluate the Safety and Efficacy of BF-200 ALA (Ameluz®) and BF-RhodoLED® in the Treatment of Superficial Basal Cell Carcinoma (sBCC) With Photodynamic Therapy (PDT).
NCT04269395	A Study to Assess Recurrence of Actinic Keratosis in Participants Treated With Methyl Aminolevulinate Hydrochloride Cream or Vehicle Cream Who Achieved Complete Response to Treated Lesions in Earlier Study
NCT03492424	Ablative Therapy in the Management of Prostate Cancer
NCT01538901	Imiquimod Versus Photodynamic Therapy of Actinic Keratoses in Organ Transplant Recipients
NCT02666534	Efficacy of AFL-assisted PDT in Microinvasive Squamous Cell Carcinoma
NCT01256424	Dose-finding Study of Hexaminolevulinate (HAL) Photodynamic Therapy (PDT) to Treat Cervical Neoplasia
NCT00470496	Photodynamic Therapy Using HPPH in Treating Patients Undergoing Surgery for Primary or Recurrent Head and Neck Cancer
NCT03053635	Intravesical Photodynamic Therapy (PDT) in BCG Refractory High-Risk Non-muscle Invasive Bladder Cancer (NMIBC) Patients
NCT03025724	Photodynamic Therapy for Treatment of Cutaneous Squamous Cell Carcinoma in Situ
NCT03048240	INtraoperative photoDYnamic Therapy of GliOblastoma
NCT02068157	Interstitial Photodynamic Therapy in Treating Patients With Recurrent Head and Neck Cancer
NCT02504957	Photodynamic Therapy With Polyhematoporphyrin for Malignant Biliary Obstruction
NCT01718223	Photodynamic Therapy Using Temoporfin Before Surgery in Treating Patients With Recurrent Oral Cavity or Oropharyngeal Cancer
NCT01854684	Photodynamic Therapy During Surgery in Treating Patients With Non-small Cell Lung Cancer That Can Be Removed by Surgery
NCT01541228	Photodynamic Treatment of Actinic Keratoses With Different Light Doses
NCT01000636	Gene Expression in Renal Transplant Patients With Field Actinic Keratosis Undergoing Metvix® Photodynamic Therapy (PDT)
NCT01673074	HPPH and PDT for Pleural Malignancy
NCT01015898	Short Term Effects of Photodynamic Therapy in Basal Cell Carcinoma
NCT00869635	S-1 and Photodynamic Therapy in Cholangiocarcinoma

NCT06159842	Evaluating the Use of Photodynamic Therapy to Treat Facial Cutaneous Squamous Cell Carcinoma in Situ (isSCC)
NCT02840331	Pilot Study on Intra-abdominal Photodynamic Diagnosis in Peritoneal Carcinosis
NCT04824742	Neoadjuvant PDT in the Treatment of Cholangiocarcinoma
NCT03467789	Vitamin D as a Nutritional Neoadjuvant During Photodynamic Therapy of Basal Cell Carcinoma
NCT02159742	Prospective Follow-up of Outcomes in Patients Receiving Photodynamic Therapy for Neoplastic Diseases
NCT03327831	Study of Daylight Photodynamic Therapy With Aminolevulinic Acid for Actinic Keratoses
NCT02149342	Daylight-mediated Photodynamic Therapy of Actinic Keratoses:Comparing 0.2%HAL With 16%MAL
NCT03849365	Study of Erectile Dysfunction, Urinary Incontinence and Related QoL After TOOKAD® VTP for Low Risk Prostate Cancer
NCT02984072	Menthol for PDT Pain Relief
NCT05919238	Padeliporfin VTP Treatment for Unresectable Pancreatic Adenocarcinoma
NCT00530088	Photodynamic Therapy Using Porfimer Sodium in Treating Patients With Recurrent Mouth or Throat Dysplasia or Recurrent In Situ Cancer or Stage I Cancer of the Mouth or Throat
NCT01086488	Foscan®-Mediated Photodynamic Therapy Versus Brachytherapy in Patients With Nasopharyngeal Carcinoma
NCT00868088	Photodynamic Therapy to Treat Actinic Damage in Patients With Squamous Cell Carcinoma (SCC) of the Lip
NCT00558688	A Photodynamic Therapy for Treatment of Actinic Keratoses
NCT01800838	Silicon Phthalocyanine 4 and Photodynamic Therapy in Stage IA-IIA Cutaneous T-Cell Non-Hodgkin Lymphoma
NCT00708942	Hexaminolevulinate (HAL) Photodynamic Therapy (PDT) of Cervical Intraepithelial Neoplasia (CIN) Grade 1
NCT00312442	Phase II/III Study of WST09 in Prostate Cancer After Radiation Therapy
NCT00756288	Photo-therapy With a Topical Retinoid Versus Photo-therapy Alone for Actinic Keratoses
NCT04099888	PCI Treatment/Gemcitabine & Chemotherapy vs Chemotherapy Alone in Patients With Inoperable Extrahepatic Bile Duct Cancer
NCT00030589	Chemotherapy and Photodynamic Therapy in Treating Patients With Cutaneous T-Cell Lymphoma
NCT03731988	Efficacy of Ablative Fractional Laser-assisted Photodynamic Therapy According to the Laser Density for Actinic Keratosis
NCT03945162	Intravesical Photodynamic Therapy ("PDT") in BCG-Unresponsive/Intolerant Non-Muscle Invasive Bladder Cancer ("NMIBC") Patients
NCT00754910	Photodynamic Therapy Using Porfimer Sodium in Treating Patients With Non-Small Cell Lung Cancer And Bronchial Disease
NCT01682746	Photodynamic Therapy (PDT) for Recurrent Pediatric Brain Tumors
NCT01524146	Photodynamic Therapy (PDT) Cholangiocarcinoma Registry
NCT03167762	Photographing the Skin During Photodynamic Therapy

NCT00306800	Metvix PDT Versus Vehicle PDT With Aktilite CL128 Lamp in Patients With Actinic Keratosis on the Face and Scalp
NCT00017485	Photodynamic Therapy in Treating Patients With Basal Cell Skin Cancer
NCT00713687	Gemcitabine/Oxaliplatin and Photodynamic Therapy in Cholangiocarcinoma
NCT02644187	Pain Relief During Photodynamic Therapy for Actinic Keratoses With a New Irradiation Protocol
NCT01260987	Fractional CO2 Laser Assisted Photodynamic Therapy
NCT00002647	Photodynamic Therapy With Porfimer Sodium in Treating Patients With Refractory Brain Tumors
NCT02872064	Treatment of Primary Breast Cancer Using PDT
NCT01522677	A Study of Neoadjuvant Photodynamic Immunomodulation for Colon Cancer
NCT00305929	Study of WST09 in Prostate Cancer After Radiation: Repeat Procedure
NCT06044142	Curcumin VS Photo-bio-modulation Therapy of Oral Mucositis in Pediatric Patients Undergoing Anti-Cancer Non-invasive Treatment
NCT00526461	Photodynamic Therapy Using HPPH in Treating Patients With Stage 0 Non-Small Cell Lung Cancer
NCT01525329	Combination Therapy With 5-Fluorouracil and Photodynamic Therapy in Post-transplant Premalignant Skin Disease
NCT05488860	Piezoelectric Drived Microneedling in Treating Refractory Skin Diseases
NCT00985829	Evaluation of Efficacy of Photodynamic Therapy in Basal Cell Carcinoma
NCT00218829	DMSO-PDT of BCC - A 6 Year Follow up
NCT04391062	Dose Finding for Intraoperative Photodynamic Therapy of Glioblastoma
NCT00974662	Study Using WST11 in Patients With Obstructing Endobronchial Non-Small Cell Lung Cancer
NCT02728388	Photodynamic Therapy for Benign Dermal Neurofibromas- Phase II
NCT00003788	Surgery, Radiation Therapy, and Chemotherapy With or Without Photodynamic Therapy in Treating Patients With Newly Diagnosed or Recurrent Malignant Supratentorial Gliomas
NCT03003065	Safety and Tumoricidal Effect of Low Dose Foscan PDT in Patients With Inoperable Bile Duct Cancers
NCT01739465	Comparison of Endoscopic Radiofrequency Ablation Versus Photodynamic Therapy for Inoperable Cholangiocarcinoma
NCT02628665	Clinical Study of Time Optimizing of Endoscopic Photodynamic Therapy on Esophageal and/or Gastric Cardiac Cancer
NCT05020912	Alteration of the Immune Microenvironment in Basal Cell Carcinoma Following Photodynamic Therapy
NCT03090412	Photodynamic Therapy With HPPH Compared to Standard of Care Surgery in Treating Patients With Oral Cavity Cancer
NCT03319251	Biomarker Database Registry for Photodynamic Therapy
NCT01497951	Photodynamic Therapy for Oral Precursor Lesions
NCT01475955	Short-incubation Levulan Photodynamic Therapy Versus Vehicle for Face/Scalp Actinic Keratosis (AK)
NCT03076892	Evaluation of Non-inferiority and Tolerability of the Device PHOS-ISTOS

NCT03973125	Application of Intravitreal Conbercept Injection as a Primary Treatment for Exudative Circumscribed Choroidal Haemangioma
NCT02685592	Photodynamic Therapy for Lentigo Maligna Using 5-aminolevulinic Acid Nanoemulsion as a Light Sensitizing Cream
NCT04140292	Vitamin D Supplementation as a Neoadjuvant for Photodynamic Therapy of Actinic Keratoses
NCT03596619	The Study of Plum-blossom Needling Enhancing Efficacy of Aminolevulinic Acid-photodynamic Therapy for Actinic Keratosis
NCT04552990	Use of Jet-injection in Photodynamic Therapy for Basal Cell Carcinoma
NCT00237107	Debulking Effect of Curretage on Basal Cell Carcinomas - a Histological Assessment.
NCT03013647	Daylight Photodynamic Therapy for Actinic Keratosis and Skin Field Cancerization
NCT01415986	Interstitial Photodynamic Therapy (PDT) With Temoporfin for Advanced Head and Neck Cancers
NCT04779255	Pain Management During a Photodynamic Therapy Session on the Vertex for Actinic Keratosis: Tumescent Anesthesia Interest
NCT02799030	A Clinical Trial of Topical Photodynamic Therapy With 5-aminolevulinic Acid for the Treatment of Actinic Keratosis
NCT03033225	Ultrasound-Guided Verteporfin Photodynamic Therapy for the Treatment of Unresectable Solid Pancreatic Tumors or Advanced Pancreatic Cancer, VERTPAC-02 Study
NCT02209012	Twelve Month Follow-Up of CP0108
NCT02670655	Efficacy of Iontophoresis-assisted AFL-PDT in Actinic Keratosis
NCT00988455	Evaluation of Efficacy of Photodynamic Therapy in Basal Cell Carcinoma With 6 Months Follow-up
NCT02939274	An Open Label, Phase II Trial of Continuous Low-Irradiance Photodynamic Therapy (CLIPT) Using Verteporfin (Visudyne®) for the Treatment of Cutaneous Metastases of Breast Cancer
NCT01032044	CLE for Assessment of Neoplasia After Mucosal Ablation or Resection of Gastrointestinal Neoplasia
NCT03643744	Photodynamic Therapy-Induced Immune Modulation: Part III
NCT02594644	The Use of Microneedles to Expedite Treatment Time in Photodynamic Therapy
NCT00978081	Photodynamic Therapy in Treating Patients With Premalignant or Early Stage Head and Neck Tumors
NCT00571974	Treatment of Oral Premalignant Lesions With 5-ALA PDT
NCT01637376	Photodynamic Therapy (PDT) With Temoporfin for Non-Resectable Non-Small-Cell Lung Cancer
NCT04469699	Stereotactical Photodynamic Therapy With 5-aminolevulinic Acid (Gliolan®) in Recurrent Glioblastoma
NCT01859169	Safety and Efficiency of Photodynamic Therapy for Blie Duct Carcinoma
NCT03735095	Endobronchial Ultrasound Guided Interstitial Photodynamic Therapy in Treating Patients With Locally Advanced Lung Cancer

NCT00322699	Sequential Whole Bladder Photodynamic Therapy (WBPDT) in the Management of Superficial Bladder Cancer
NCT05551299	Treatment of Non-resectable Bile Duct Cancer With Radiofrequency Ablation or Photodynamic Therapy
NCT04301999	Clinical Effect and Safety of PDT and RFA for Unresectable EHCC
NCT00122876	Tumor Ablation With Talaporfin Sodium and Interstitial Light Emitting Diodes Treating Hepatocellular Carcinoma (HCC)
NCT02725073	Photodynamic Therapy in Locally Advanced Hilar Cholangiocarcinoma
NCT04836429	Photodynamic Therapy to Amplify the Response to Immunotherapy in Patients With Non-small Cell Lung Cancer With Pleural Disease
NCT02144077	Safety and Efficacy Study for the Treatment of Non-Aggressive Basal Cell Carcinoma With Photodynamic Therapy
NCT02070432	Photodynamic Therapy With LUZ11 in Advanced Head and Neck Cancer
NCT02662504	Intrapleural Photodynamic Therapy in a Multimodal Treatment for Patients With Malignant Pleural Mesothelioma
NCT03346304	Photodynamic Therapy for the Prevention of Lung Cancer
NCT03315754	Study of the Efficacy, Safety and Quality of Life After TOOKAD® Soluble (VTP) for Intermediate Risk Prostate Cancer.
NCT01459393	Comparison Between 5-aminolevulinic Acid Photodynamic Therapy Versus Cryotherapy for Actinic Keratosis Treatment
NCT01253759	Long Term Results of Combined Transpupillary Thermotherapy (TTT) Indocyanine Green (ICG) Based Photodynamic Therapy (PDT) in Choroidal Melanoma
NCT02464761	Photodynamic Therapy for the Treatment of Vertebral Metastases
NCT03727061	Porfimer Sodium Interstitial Photodynamic Therapy With or Without Standard of Care Chemotherapy in Treating Patients With Locally Advanced or Recurrent Head and Neck Cancer
NCT01686594	PUVA Maintenance Therapy in Mycosis Fungoides
NCT02622594	Bilateral Comparison of Treatment of Facial Actinic Keratoses Using Microneedling and Photodynamic Therapy With Aminolevulinic Acid and Blue Light Versus Photodynamic Therapy With Aminolevulinic Acid and Blue Light Using Two Incubation Times
NCT02153229	Phase II Trial of Radical Pleurectomy With or Without Intraoperative PDT for Malignant Pleural Mesothelioma
NCT00975429	Study Using WST11 in Patients With Localized Prostate Cancer
NCT05923060	Imaging Techniques to Monitor Photosensitizer and sO2 Levels During Photodynamic Therapy of Actinic Keratoses
NCT05937529	Impact of Madecassoside and 5 % Panthenol Cream in Post Photodynamic Therapy for Actinic Keratosis
NCT04943094	Clinical and Functional Consequences of Photodynamic Diagnosis (PDD) and Intravesical Instillation Therapy
NCT05850377	5-Aminolevulinic Acid (5-ALA) Gliolan®: Usage Increase Proposal for Neurosurgical Procedures in High-Grade Gliomas

NCT02632110	Microneedle Lesion Preparation Prior to Aminolevulinic Acid Photodynamic Therapy (ALA-PDT) for AK on Face
NCT03909646	Surgical Excision Versus Photodynamic Therapy and Topical 5-fluorouracil in Treatment of Bowen's Disease
NCT00103246	Photodynamic Therapy Using Silicon Phthalocyanine 4 in Treating Patients With Actinic Keratosis, Bowen's Disease, Skin Cancer, or Stage I or Stage II Mycosis Fungoides
NCT03076918	Evaluating the Device FLEXITHERALIGHT Compared to the Conventional Photodynamic Therapy
NCT03344861	Safety of PDT-Photofrin® Prior to Lung Surgery
NCT01349361	Treatment of Basal Cell Carcinomas With Methyl Aminolevulinate and Daylight
NCT01016002	Single-arm Study of Photodynamic Laser Therapy Using Foscan for Non-curatively-resectable Bile Duct Carcinoma
NCT04753918	Novel Light Delivery Methods for Lung Cancer Photodynamic Therapy - A Pilot Study
NCT01203878	Treatment of Actinic Keratoses of the Face With Imiquimod 3.75% Cream Followed by Photodynamic Therapy
NCT00453336	Photodynamic Therapy With Porfimer Sodium in Treating Patients With Precancerous Lesions, Cancer, or Other Disease of the Aerodigestive Tract
NCT05456334	New Treatments for Actinic Keratoses of the Scalp
NCT02281136	Maximal Use Systemic Exposure (MUSE) Study of Levulan Kerastick
NCT00281736	Photodynamic Therapy in Treating Patients With Precancerous Esophageal Conditions or Early Stage Esophageal Cancer
NCT02119728	Photodynamic Therapy With HPPH in Treating Patients With Squamous Cell Carcinoma of the Oral Cavity
NCT05662202	Study to Evaluate the Safety, Tolerability and Efficacy of BF-200 ALA (Ameluz®) in the Field-directed Treatment of Actinic Keratosis (AK) on the Extremities and Neck/Trunk With Photodynamic Therapy (PDT) Using a RhodoLED Lamp
NCT00054002	Surgery and Photodynamic Therapy in Treating Patients With Malignant Mesothelioma
NCT00843323	Photodynamic Therapy (PDT) Effect on Large Surface Photodamaged Skin
NCT01140178	A Trial of Photodynamic Therapy With HPPH for Treatment of Dysplasia, Carcinoma in Situ and T1 Carcinoma of the Oral Cavity and/or Oropharynx
NCT00711178	A Randomized Multicenter Study of Daylight Mediated Photodynamic Therapy (PDT)
NCT03211078	ENB and Photodynamic Therapy in the Treatment for Early Lung Cancer
NCT00571558	Photodynamic Therapy Using Aminolevulinic Acid in Treating Patients With Oral Leukoplakia
NCT05580328	An Exploratory Clinical Study of Photodynamic Therapy Combined With Sonodynamic Therapy in Cholangiocarcinoma
NCT00814528	Photodynamic Therapy Mediated by Topical Application of 5- ALA for the Treatment of Actinic Keratoses
NCT02157623	Blue vs Red Light During Levulan Based Photodynamic Therapy in Patients With Basal Cell Nevus Syndrome

NCT01770132	Ultrasound-Guided Photodynamic Therapy With Photofrin & Gemcitabine for Patients With Locally Advanced Pancreatic Cancer
NCT01898936	Fractional Laser-assisted Daylight Photodynamic Therapy Versus Daylight Photodynamic for Treatment of Actinic Keratoses
NCT05522036	Clinical Evaluation of a Short Illumination Duration (35 Minutes) When Performing PDT of AK Using the Dermaris ®
NCT03990636	Photodynamic Therapy With Metil 5-aminolevulinate for Actinic Cheilitis - Phase 2 Clinical Trial
NCT02628236	Maximal Use Systemic Exposure Study of Levulan Kerastick (MUSE 2)
NCT01571336	Photodynamic Therapy of Actinic Keratoses With Alacare®
NCT00003923	Photodynamic Therapy in Treating Patients With Cancer of the Bile Duct, Gallbladder, or Pancreas
NCT04396184	Conventional Photodynamic Therapy vs. Painless Photodynamic Therapy for Actinic Keratosis
NCT00946881	Safety and Tolerability Study Using WST11 In Patients With Localized Prostate Cancer
NCT00369018	A Dose-finding Study of MAL and HAL Photodynamic Therapy of Cervical Premalignant Lesions.
NCT02555501	Oral Mucositis and Laser Therapy Associated With Photodynamic Therapy
NCT00862901	Safety Study Using Photodynamic Therapy Light Therapy for Patients With Chest Wall Progression of Breast Cancer and Satellite Metastases of Melanoma
NCT01668823	Photodynamic Therapy in Treating Patients With Lung Cancer
NCT00269074	Effect of IDEA-070 on Pain and Inflammation Induced by PDT
NCT00432224	Treatment of Actinic Keratoses With Photodynamic Therapy Using Sunlight Versus Red Light
NCT02248298	Efficacy of AFL-assisted PDT With Short Incubation Time in Actinic Keratosis
NCT04225299	Evaluation of Efficacy of TOOKAD® (VTP) Versus Active Surveillance for Intermediate Risk Localized Prostate Cancer
NCT02799069	Evaluation of Safety and Efficacy of BF-200 ALA for the Treatment of Actinic Keratosis With Photodynamic Therapy
NCT00007969	Photodynamic Therapy in Treating Patients With Stage III or Stage IV Melanoma
NCT00907413	Photodynamic Therapy (PDT) Trial for Palliation of Cholangiocarcinoma
NCT00716469	Phase I Clinical Study of the Safety of Photodynamic Therapy (PDT) Using LS11 in Children With Plexiform Neurofibromas
NCT00540735	Efficiency Evaluation of Photodynamic Therapy With Photofrin® on Unresectable Type III or IV Cholangiocarcinomas
NCT01912976	Study of Methyl Aminolaevulinate Photodynamic Therapy With and Without Er:YAG Laser in Bowen's Disease
NCT03453476	Effect of Photodynamic Therapy on Gingival Crevicular Cytokines in Periodontitis Patients
NCT01755013	Photodynamic Therapy (PDT) for Palliation of Cholangiocarcinoma

NCT01794299	Safety and Efficacy of Donor T-lymphocytes Depleted ex Vivo of Host Alloreactive T-cells (ATIR) in Patients With a Hematologic Malignancy Who Received a Hematopoietic Stem Cell Transplantation From a Haploidentical Donor
NCT02100111	An Analysis of Treatment Patterns and Outcomes for Basal Cell Carcinoma (BCC) Cancer Participants
NCT02304770	Aminolaevulinic Acid Photodynamic Therapy of Cervical Persistent HPV Infection and Cervical Neoplasia
NCT00002935	Photodynamic Therapy in Treating Patients With Early Esophageal Cancer
NCT03281811	Photodynamic Therapy in Treating Patients With Refractory Mycosis Fungoides
NCT01209013	Safety of Photodynamic Therapy (PDT) in the Ablation of High-grade Dysplasia (HGD) in Barrett's Esophagus (BE)
NCT01491711	Superficial Basal Cell Carcinoma Treatment With Topical Photodynamic Therapy With Fractionated 5-aminolevulinic Acid 20% Versus Two Stage Methylaminolevulinate
NCT01682811	Phase I Photodynamic Therapy (PDT) for Benign Dermal Neurofibromas (NF1)
NCT05725213	ADL-PDT Under Routine Clinical Conditions in Patients With Actinic Keratosis
NCT03642535	Aminolevulinic Acid-photodynamic Therapy for Facial Actinic Keratosis Treatment and Prevention
NCT01292668	Photodynamic Therapy Using Methyl-5-Aminolevulinate Hydrochloride Cream in Determining Pain Threshold in Patients With Skin Cancer
NCT00005808	Photodynamic Therapy Using Lutetium Texaphyrin in Treating Patients With Cervical Intraepithelial Neoplasia
NCT00253617	Stent Placement With or Without Photodynamic Therapy Using Porfimer Sodium as Palliative Treatment in Treating Patients With Stage III or Stage IV Cholangiocarcinoma That Cannot Be Removed By Surgery
NCT01481155	Comparative Study of Photodynamic Therapy vs. CO2 Laser Therapy in Treatment of Actinic Keratoses
NCT01898936	Fractional Laser-assisted Daylight Photodynamic Therapy Versus Daylight Photodynamic for Treatment of Actinic Keratoses
NCT02555501	Oral Mucositis and Laser Therapy Associated With Photodynamic Therapy
PTT	
NCT00805883	MRI Targeted Focal Laser Thermal Therapy of Prostate Cancer Followed by Radical Prostatectomy
NCT00848042	Pilot Study of AuroLase(tm) Therapy in Refractory and/or Recurrent Tumors of the Head and Neck
NCT01679470	Efficacy Study of AuroLase Therapy in Subjects With Primary and/or Metastatic Lung Tumors
NCT03202446	Randomized Clinical Trial Evaluating the Use of the Laser-Assisted Immunotherapy (LIT/inCVAX) in Advanced Breast Cancer
PCI	

NCT00993512	Safety Study of Amphinex Based Photochemical Internalisation (PCI) of Bleomycin in Patients With Cutaneous Cancer
NCT00586040	Photochemical Tissue Bonding
NCT01872923	Dose Escalating Study for Amphinex-based PCI of Bleomycin.
NCT01900158	A Phase I/II Safety and Efficacy Study of PCI of Gemcitabine and Chemotherapy in Patients With Cholangiocarcinomas
NCT02947854	Study to Assess Safety, Tolerability and Immune Response of Fimaporfin-induced Photochemical Internalisation of Antigen/Adjuvant
NCT04099888	PCI Treatment/Gemcitabine & Chemotherapy vs Chemotherapy Alone in Patients With Inoperable Extrahepatic Bile Duct Cancer