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INTRODUCTION

Cancer is an abnormal cell growth that is considered as the prime cause of mortality worldwide and it decreases the life span of people in all the countries. According to the survey of World Health Organization in the year 2020, it is estimated that the cancer is the leading cause of death in most of the countries and it holds third or fourth place in the cause of mortality in 23 countries (WHO, 2021).

Leukaemia is a type of cancer which leads to the increase in the number of leucocytes either in the bone marrow or in blood. There are four different types of leukaemia based on its origin, pathogenesis and prognosis. They are, "Acute Lymphoblastic Leukemia (ALL), Chronic Lymphoblastic Leukemia (CLL), Acute Myeloid Leukemia (AML) and Chronic Myeloid Leukemia (CML)" (Kipps *et al.,* 2017). The most common type of cancer among children is Acute Lymphoblastic Leukaemia and it accounts for about 25% of all cancers in children who are below the age of 15. Patients with blastic transformation of T and B cells are more prone to ALL (Chennamadhavuni, 2021). In most of the cases, ALL is treatable only if the adequate facilities available as in developed countries. Developing countries are still facing difficulty in treating ALL due to the lack of facilities to carry out modern treatment strategies (Abdelmabood *et al.,* 2020).

An atom or a molecule with minimum one or more unpaired ellectron is considered as free radicals. Since they contain unpaired electrons they are highly reactive. They have the ability to accept or donate electrons and hence play role as reductants or oxidants (Bo *et al.,* 2019). They take part in various cellular processes including the redox system alteration, induce DNA damage and also induce the activation of procarcinogens which ultimately results in cancer (Maddu, 2019). Free radicals are mainly categorized into two types namely Reactive Oxygen Species (ROS) which are oxygen derived and Reactive Nitrogen Species (RNS) which are nitrogen derived. The examples of ROS includes hydroxyl radicals (•OH), Peroxyl radical (ROO•), superoxide anion ($O^{\bullet-}_2$), alkoxyl radical (RO•), singlet oxygen ($^1O_2$) and hydrogen peroxide (H_2O_2) (Ramos and Muriel, 2019). The main RNS that is involved in various free radical related diseases is nitric oxide

(NO•). The excessive production of these toxic free radicals in the body leads to oxidative damage which results in various oxidative stress mediated diseases (Masuko *et al.,2021*).

Over the last few years, various anticancer drugs or agents are employed for cancer management which includes hormone therapeutics, alkylating agents, biological response modifiers, antibiotics, platinum compounds, antimetabolites and mitotic inhibitors. The major disadvantage of these anticancer agents is due to their adverse effects, which include fatigue, immunosuppression, liver damage, severe nausea and vomiting, lack of appetite, abdominal pain, back pain and weakness (Sagbo and Otang, 2021). The main reason for these side effects is that these anticancer agents or drugs lack appropriate specificity and sensitivity. Therefore, an anticancer drug with minimal or no side effects is the need of the hour (Schirrmacher, 2019).

Antioxidants are the molecules that act as a protective shield for the body by preventing the damage caused by the free radicals. The actual mechanism of antioxidants is that, they share their extra electrons to the free radicals which prevent them from damaging the cellular components (Jamshidi *et al.,* 2020). The antioxidants are broadly classified into two categories on based on their nature; they are enzymic antioxidants and non- enzymic antioxidants. The examples of enzymic antioxidants are catalase, glutathione peroxidase, superoxide dismutase and glutathione reductase. The major non enzymic antioxidants are flavonoids, ascorbic acid, β carotene and α-tocopherol. To maintain a healthy homeostasis, the body always maintains a balance between the level of free radicals and antioxidants (Anju *et al.,* 2019). Various parts of the plants like barks, roots, leaves, nuts, seeds, vegetables and fruits are the major source of natural antioxidants. The phytochemicals or secondary metabolites present in these plants acts as natural antioxidants (Glucin, 2020).

Ancient ayurveda and Indian traditional medicine uses plants for the treatment of several ailments. The plants possess medicinal properties due to the presence of abundant phytochemicals and their derivatives. One of the important properties of these phytochemicals is that they can act as anticancer agents. These phytochemicals are biologically active compounds that are present in nature with considerable anticancer potential. The major benefit of these phytochemicals is that they produce very less or minimal side effects. So the purification of these phytochemicals from the plants and using them as anticancer drugs will be the best adverse effect free treatment for cancer (Choudhari *et al.,* 2020).

Nanotechnology has gained much importance among the scientific community since the last century. Scientists have created a lot of revolution in the field of nanotechnology. Materials of size less than 100nm are considered as nanoparticles. The major advantage of these nanoparticles is their size, because the size of the nanoparticles influences the physiochemical properties of a substance. So different sized nanoparticles show variation in their shape, size and color (Ibrahim *et al.,* 2019). Nanoparticles can be synthesized in various shapes, size, and dimensions which shows diverse physiochemical properties. Based on the application, the nanoparticles can be customized in various shapes like wires, rods, sheets and particles (Modi *et al.,* 2022). Based on the chemical composition used for the preparation of nanoparticles they are divided into four types: i) Organic nanomaterials which include nanoconjugates, hydrogels, polymerosomes, dendrimers and micelles. ii) Inorganic nanomaterials which include ceramic nanomaterials, metal oxides and metals. iii) Composite nanostructures. iv) Carbon based nanomaterials which include graphene, carbon nanotubes, carbon nanofibres and fullerenes (Gaur *et al.,* 2021 and Sawy *et al.,* 2021)

Nanoparticles have a wide range of applications since they are prepared by converting the bulk materials into nanosize which makes changes in their physiochemical properties. It has enormous application in various fields like medical and pharmaceutical sciences, molecular biology, material science, physical, chemical and biological sciences (Ruqayah *et al.,* 2021). The nanomaterials are found to possess high surface to volume ratio which facilitates them to interact with cellular and molecular processes. This property attracted the biomedical scientists to apply them in several biomedical researches (Chandrakala *et al.,* 2022).

Metallic nanoparticles have paved its way in the field of nanotechnology because of its diverse properties. It has a remarkable application especially in the field of targeted drug delivery. They are able to carry various therapeutic agents like peptides, chemotherapeutic drugs, nucleic acids and antibodies to the target site. This targeted drug delivery is achieved by the metallic nanoparticles, since they develop a strong interaction with the therapeutic agent through electrostatic interaction, covalent bonding and hydrogen bonding. The optical properties of various metallic nanoparticles like gold, copper, zinc, silver, palladium and titanium nanoparticles can be modified for different applications (Chandrakala *et al.,* 2022).

Among the various metallic nanoparticles available, the silver nanoparticles attracted many researchers because of its unique magnetic, electrical and optical properties which leads to its

diverse applications. Silver nanoparticles have various applications in cosmetic products, food industry, electronic components, biosensors, composite fibers, catalysts, optical elements, textile industry and also they are found to possess anti inflammatory, antiviral, antifungal, antibacterial and antiseptic properties (Arif *et al.,* 2021). In recent reports, silver nanoparticles found a remarkable application in the field of cancer therapy. They are given role in the treatment and diagnosis of cancer and also they are used as the drug delivery carrier (Raj *et al.*, 2022). The major advantage of these silver nanoparticles is that they exert minimal damage to the mammalian cells when compared to other metallic nanoparticles (Sonika *et al.,* 2021).

Enormous and less expensive routes are available for the synthesis of AgNPs like physical, chemical, electrochemical and sonochemical methods (Khandel *et al.,* 2018; Fahmy *et al.,* 2019). But the major disadvantage of these methods is that they are hazardous to the environment especially when the synthesis is carried out with the use of chemicals and also the AgNPs are synthesized using these methods will tend to agglomerate easily because of their monodispersity property (*Gulati et al.,2018).* Among the various methods available for the synthesis of AgNPs, the plant mediated green synthesis is found to be very effective, easy to carryout, eco- friendly and cost effective. The phytochemicals available in the plant extracts acts as a capping agent that prevents them from agglomeration and this makes them very stable for a longer period (Bukhari *et al.,* 2019). These silver nanoparticles are used as anticancer agents due to their antiproliferative and apoptosis inducing properties (Almashgab *et al.,* 2020).

Liposomes are spherical vesicles made up of lipid bilayer. The major composition of liposomes is cholesterol and phospholipids. They can encapsulate both hydrophilic and hydrophobic drugs (Beltrán *et al.,* 2019). Alec D. Bangham discovered the liposomes in 1965 and they are the first approved therapeutic nanocarrier for the treatment of cancer (Bourquin *et al.,* 2018). They are widely used in nanotherapeutics because they are non immunogenic, biocompatible, non toxic and also biodegradable (Zamani *et al.,* 2018). The liposomes are successful carriers because their phospholipid bilayer resembles the mammalian cell membrane, which eventually facilitates the efficient cellular uptake. So the liposomes can reach the cells with high concentration which reduces unnecessary side effects and enhances the efficacy (Gonda *et al.,* 2019).

Based on the number of phospholipid bilayer present, the liposomes are classified into four types. They are unilamellar, multilamellar, oligolamellar, and multi vesicular vesicles. Various methods are available for the preparation of liposomes which include injection technique, microfluidic method, heating method, reverse phase evaporation technique, hydration methods, detergent depletion, thin film hydration method, membrane extrusion, freeze drying and are well documented (Has and Sunthar, 2019).

The liposomes of size ranging between 50 to 200 nm are preferable for drug delivery applications. Size of the liposomes is a major factor which influences the drug delivery to the targeted cells. Liposomes less than 200 nm possess enhanced drug release, high circulation time and accumulation in the target site. There are many routes where liposomes can be administered which include oral, ocular and intravenous for treating the variety of diseases (Leitgeb *et al.,* 2020).

Tabebuia pallida often called as "Pink trumpet tree" that belongs to the family "Bignoniaceae" is one of the vast species that is distributed throughout South America, Central America and Western region of India (Bussmann, 2018). It is usually grown as an ornamental tree that reaches upto 25-30 meter height. *Tabebuia pallida* is the candidate plant of the present study to synthesize the AgNPs. Tabebuia species are mainly employed in Ancient folk medicine for treating various diseases including allergies, syphilis, constipation, malaria, prostatitis, cutaneous infections, diabetes, stomach disorders, depression, cancer, irritability, inflammation, poor memory, pain, anxiety, fungal and bacterial infections (Ferraz *et al.,* 2017 and Regalado *et al.,* (2017). Since this plant is employed to cure various diseases in ancient folk medicine especially for curing cancer, there are very less scientific evidences available, and hence this plant is chosen for the present study.

Hypothesis of the study

The present study was framed to analyze the following hypothesis:

- Null hypothesis (H_0): *Tabebuia pallida* silver nanoparticles loaded liposomes do not possess drug release and anticancer properties
- Alternate hypothesis (H_A): *Tabebuia pallida* silver nanoparticles loaded liposomes possess drug release and anticancer properties

Thus, the present study was framed with the following objectives to test the above mentioned null and alternate hypothesis

- To synthesize and characterize AgNPs and AgNPs loaded Liposomes from *Tabebuia pallida* leaf extract
- To evaluate the *invitro* drug release of AgNPs loaded Liposomes and its validation through mathematical models
- To evaluate antioxidant and anticancer potential of AgNPs of *T. pallida* leaves and their liposomes

The review of literature relevant to the present study is presented in the next chapter.