1. INTRODUCTION

Lectins are a diverse class of sugar-binding proteins of non-immune origin that are known to reversibly bind to specific carbohydrates [1]. They were initially discovered in castor beans in 1988 and have since then been purified from a wide variety of natural sources including plant roots, seeds, barks, animals, bacteria, viruses, fungi, molluscs, seaweeds, etc. As most lectins are multivalent in nature, their interaction with the sugar residues present on the surface of the RBCs can cause clumping of these cells. RBC agglutination which is a major attribute of lectins is routinely used in their detection and characterization.

As knowledge regarding their molecular properties of lectins is essential for understanding their activities at the molecular level, the amino acid sequences and three-dimensional conformation of a number of lectins, isolated from widely varied sources have been elucidated. Further, the chemical groups and bonds that are responsible for diverse interactions between the lectins and the specific carbohydrates they bind to have also been identified by high-resolution X-ray crystallographic studies. The conformational stability of various lectins have been determined by analyzing the denaturant or thermal-induced unfolding transition [2,3]. Studying the molecular basis of stability and assembly of a lectin will help to further understand the structure-function relationship and to broaden the horizon of its applications.

Accumulating evidence suggests the involvement of lectins in biological processes such as facilitating attachment of infectious agents to host cell surface, mediating cell to cell and cell to matrix interactions, mediating endocytosis and regulating cellular growth [4]. Besides, the ability of lectins to specifically recognize cancer cells by binding to aberrant glycosylation present on the tumor cell surface is being exploited in diagnosis and prognosis of various cancers [5]. Lectins also exhibit potential to be developed into effective anticancer agent by virtue of their ability to selectively instigate autophagy and/or apoptosis in cancer cells. Various lectins isolated from diverse families

with different sugar-binding specificities are known to possess significant antitumor effects *in vitro*, *in vivo* and also in case studies involving humans [6]. While some lectins act as non-cytotoxic inhibitors of cancer cell proliferation, others such as ricin, wheat germ agglutinin (WGA) and mistletoe lectins (MLs) induce apoptosis of cancer cells. Also, few lectins were found to be internalized into the tumor cells after which they induce autophagy [7]. Several plant lectins also exert potential immunomodulatory effects which are attributed to their ability to cross-link glycoproteins present on the immune cell surface [8]. Since many decades, plant lectins have been recognized as immunomodulatory agents and are known to modulate immune mechanisms including effector functions and inflammatory reactions. Apart from modulating the production of cytokines, certain lectins also influence the production of other mediators of immune response such as nitrogen species and reactive oxygen species. The ability of lectins to modify the immune responses to combat pathological conditions such as cancer and other microbial infections have been extensively reported [9].

1.1 DISTRIBUTION OF LECTINS

Lectins are a heterogeneous class of sugar-binding proteins that have been isolated from plants, microorganisms, animals and marine sources such as algae, fishes and marine cyanobacteria. Lectins have also been isolated from lesser known sources such as slime molds and protozoa.

1.1.1 Microbial lectins

The main function of lectins or glycan-binding proteins expressed by microorganisms is to facilitate their adhesion to the host cell surface. The first viral lectin referred to as a hemagglutinin was identified in influenza virus in the early 1950s. The viral hemagglutinin was shown to interact with sialic acid residues present on the erythrocytes resulting in agglutination.

Bacterial lectins were initially identified in the 1970s and are present in pili or fimbriae which interact with the glycoproteins present on the surface of

the host cells. Lectins expressed on the surface of pathogenic bacteria such as *Helicobacter pylori* and *E. coli* are considered as one of the virulence factors as they facilitate attachment of the bacteria on to the host cell surface [10].

Though the occurrence of lectins in fungi was first reported as early as 1891, it was only in recent years that fungal lectins attained wide attention due to their various biological properties. Majority of fungal lectins have been isolated from the fruiting bodies [11,12]. However, lectins have also been isolated from the conidia [13], mycelium [14,15]. and sporomes [16]. Some fungal species such as *Conidiobolus lampraugus* and *Neurospora sitophila* produce two agglutinins, one associated with the cell wall and the other secreted into the culture filtrate [17,18]. Most of the lectins isolated from fungus that are pathogenic to humans have been implicated in their adherence to the host cell surface; for instance, an extracellular agglutinin isolated from a dermatophyte, *Trichophyton rubrum* mediated attachment on to the host cell surface which is a prerequisite for its entry into the cells [19].

1.1.2 Animal lectins

Initially, most animal lectins were isolated from invertebrates and lower vertebrates and were reported to play a vital role in innate immune response. The galactose-specific family of animal lectins known as the galectins are the most commonly found family of animal lectins and have been isolated from mammals, sponges and worms. They are found in almost all types of cells and are found both inside and outside the cells. Within the cells, galectins occur in the cytosol and nucleus while they may also occur in the intercellular space or on the cell surface [20]. While some animal lectins act as molecular chaperones during glycoprotein synthesis, others may be involved in cell-cell interactions, mediation of endocytosis and cellular growth regulation [21].

1.1.3 Plant lectins

In plants, apart from seeds which are the major sources, lectins alos occur in other vegetative tissues including barks, corns, flowers, fruits,

bulbs,ovaries, roots,rhizomes and leaves. Lectins are the major proteins present in most of the leguminous seeds and storage tissues of plants. The seed lectins mainly occur in the cotyledons during the later stages of maturation and in some cases, they are also found in the embryo and seed coat [22]. They constitute 1 to 10% of total protein content in seeds; for instance, phytohaemagglutinin (PHA) constitutes 10% of the total protein present in the seeds of *Phaseolus vulgaris* [23]. In some species, the seed lectin constitutes 50% of the total protein content of the seed. In case of bulbs, bark, tubers, corn and rhizomes, the lectin accounts for 1-20% of the total protein content.

Some non-seed lectins also occur in various tissues of the same plant; for instance, although the lectin from snowdrop and daffodil are most abundant in bulbs, they are also present in all other vegetative tissues [24]. Also, the lectins isolated from the tubers and pericarp of potato were similar but differed considerably from the seed lectin [25]. Apart from protecting the plants from predatory animals and phytopathogens, plant lectins also mediate symbiotic relationship between the leguminous plant and rhizobia.

1.2 CLASSIFICATION OF PLANT LECTINS

Plant lectins were broadly classified into polyspecific or monospecific lectins based on whether they interact with one or more than one sugars [26,27]. Besides, plant lectins have also been classified on the basis of different criteria such as their overall structure, carbohydrate specificity, and evolutionary relatedness.

1.2.1 Classification based on carbohydrate specificity

Based on the specific sugars they bind to, lectins were initially classified into varied groups such as mannose binding, mannose/maltose binding, mannose/glucose binding, galactose/N-acetylgalactose binding, N-acetylglucosamine binding, L-fucose specific lectins and sialic acid specific lectins [28]. The sugar-binding specificity of lectins is generally determined by

the hapten-inhibition technique [29]. Likewise, other physical methods like fluorescence spectroscopy and equilibrium analysis have also been used to determine the carbohydrate specificity [30]. However, as more lectins from varied sources were identified, other classification systems evolved.

1.2.2 Classification based on structure

Based on their overall structure, mature plant lectins have been classified into the following four groups; merolectins, hololectins, chimerolectins and superlectins [31].

Merolectins

Merolectins are small proteins that consist of a single sugar binding site. As they are monovalent in nature, they are neither able to cause agglutination of cells nor precipitation glycoconjugates; e.g., the mannose-binding lectin isolated from leaves of orchid twayblade [32].

Hololectins

Hololectins consists of two or more, similar or almost similar sugar-binding domains that specifically bind to the same or structurally similar sugars. Most plant lectins are hololectins and are capable of agglutinating cells. Concanavalin A (Con A) isolated from *Canavalia ensiformis* is a well-known example of a hololectin [33].

Chimerolectins

Chimerolectins consists of a carbohydrate-binding domain and another unrelated domain which possess some biological activity. The activity of the latter domain is independent of the Carbohydrate recognition domain (CRD). Chimerolectins with a single CRD do not agglutinate cells and behave like a merolectin while those with more than one CRDs act like a hololectin; for instance, the type 2 Ribosome Inactivating Proteins (RIP) has two carbohydrate-binding domains that are present on the B chain and can agglutinate cells and precipitate glycoconjugates [31].

Superlectins

Superlectins consist of two CRDs which recognize two different, unrelated sugars; e.g., the lectin TxLCI isolated from tulip bulbs consists of two tandemly arrayed domains that possess specificity toward mannose and N-acetylgalactosamine (GalNAc) respectively [34].

1.2.3 Classification based on domain structure and binding capabilities

Depending on their domain structure and binding capabilities, plant lectins were initially classified into the following seven families; legume lectins, jacalin related lectins (JRLs), chitin-binding lectins, Galanthus Nivalis Agglutinin (GNA)-related lectins, type 2 Ribosome Inactivating Proteins (RIP II), Amaranthin lectin family and the Cucurbitaceae phloem lectins [35].

Legume lectins

Legume lectins include a large family of extensively studied sugar-binding proteins, most of which have been purified from plants that belong to the Leguminosae family. In spite of their sequence similarity and evolutionary relatedness, they differ in their quaternary structure and also exhibit wide range of sugar specificities [36]; for instance, Con A isolated from *Canavalia ensiformis* and PSA isolated from *Pisum sativum* are mannose/glucose-specific legume lectins while peanut agglutinin (PNA) isolated from *Arachis hypogaea* and Soybean agglutinin (SBA) isolated from *Glycine max* are galactose/N-acetylgalactosamine-binding lectins. Others such as Ulex europaeus agglutinin I (UEA I) isolated from *Ulex europaeus* and lectin from *Lotus tetragonolobus* are fucose-specific legume lectins.

Majority of legume lectins are dimers or tetramers, that largely consists of antiparallel β sheets. Each of the 2 or 4 subunits which are about 25-30 kDa include a carbohydrate-binding site and binding sites for transition metals (Ca^{2+} and Mn^{2+}). The amino acid residues found in the metal-binding site are highly conserved and the binding of metals is requisite for lectin-carbohydrate interaction [37]. Depending on their quaternary structure, legume lectins are subdivided into two groups; one group includes lectins in which the subunits

are identical or nearly homologues as in the case of PHA, Con A and SBA. Another group includes lectins made up of distinct subunits; lectins isolated from *Pisum sativu*m and *Lens culinaris* that consists of a larger β subunit and a smaller α subunit are relevant examples of this group [38,39].

Jacalin related lectins (JRLs)

JRLs are a group of sugar-binding proteins that exhibit sequence similarity with the lectin jacalin. Based on the specificity, JRLs are further divided into two subfamilies: Galactose-specific JRLs (gJRL) and the Mannose-specific JRLs (mJRL). The Gal-specific homologs of jacalin have been identified only from other Artocarpus species and in seeds of *Maclura pomifera* [40]. Unlike the gJRLs that are confined to the family of Moraceae, the mJRLs have been isolated from species that belong to many taxonomic groups; for instance, the mJRLs have been isolated and characterized from the pulp of banana, the leaves of salt stressed rice, the rhizomes of hedge bindweed, the tubers of Jerusalem artichoke, the seeds of *Parkia platycephala* and from the Japanese chestnut [41-45].

Chitin-binding lectins

This family of lectins consists of structurally conserved GlcNAc-binding domains and are found in plants belonging to families such as Solanaceae, Gramineae, Urticaceae, Phytolaccaceae, etc. [46]. They are made up of two identical subunits and are found to be rich in cysteine.

Galanthus Nivalis Agglutinin (GNA)-related lectins

This family of glycoproteins were initially found to occur only in plants from monocot families including Araceae, Alliaceae, Liliaceae, Bromeliaceae, Amaryllidaceae, and Orchidaceae [28]. However, lectins with GNA domains were later identified in fish, liverwort and the gymnosperm *Taxus media* [47]. Most GNA-related lectins share sequence similarity and contain 1, 2 or 4 subunits of 12,000 kDa M_r [48].

Type-2 RIPs

Type 2 RIPs are a superfamily of evolutionarily related glycoproteins that consists of an A-chain which is enzymatically active and is bound to a B-chain through a disulfide bridge. The larger B chain is the lectin subunit that exhibits specificity towards galactose and N-acetylgalactosamine [49]. The type 2 RIPs are that found in plant families such as Euphorbiaceae, Fabaceae, Sambuceae, Leguminosae, etc., are usually located in all plant parts including roots, shoots, seeds, leaves, bulbs, fruits and bark [50]. All type 2 RIPs share a similar 3D structure with an approximate M_r of 56-65 kDa. Some type 2 RIPs such as modeccin, ricin, abrin, lanceolin, volkensin, aralin, riproximin, etc., are potent toxins. These proteins possess distinct cytotoxic properties, substrate specificity and catalytic activity. Their cytotoxicity counts on the binding of the B-chain to the specific sugars present on the cell surface thereby facilitating uptake of these proteins into the cells.

Amaranthin lectin family (Amaranthins)

The Amaranthin family includes GalNAc-specific lectins isolated from the seeds of plants such as *Amaranthus caudatus*, *A. cruentus*, *A. spinosus* and *A. leucocarpus*, all of which belong to the Amaranthaceae family. The Amaranthins are homodimeric proteins that are about 30 kDa and exhibit specificity towards GalNAc and the disaccharide, Thomsen-Friendenreich (TF).

Cucurbitaceae phloem lectins

These are a small family of carbohydrate-binding proteins that are confined to the phloem of some species belonging to the Cucurbitaceae family. This family includes chitin-binding lectins that were isolated from phloem exudates of Cucurbita, Sechia, Luffa, Citrullus, etc. Most lectins belonging to this family are known to occur as dimers in solution and exhibit specificity towards the oligomers of GlcNAc.

1.2.4 Other systems of classification of plant lectins

Based on their sequence, structure homology and their evolutionary relatedness, lectins were further classified into the following 12 families which included almost all the identified and well-characterized plant lectins; Amaranthin, Agaricus Bisporos agglutinin, Chitinase-related agglutinin, Cyanovirin, GNA-related lectins, *Euonymus europaeus* agglutinin, Hevein, JRLs, Lysine motifs, legume lectins, Nicataba and Ricin-B families [51].

1.3 CLASSIFICATION OF ANIMAL LECTINS

Initially, animal lectins were classified into two main groups, namely S-type or the thiol dependent lectins and the C-type or Ca^{2+} dependent [52]. In addition to the previous two groups, animal lectins were further divided into other families such as I-type, C-type, P-type, pentraxins, calreticulin and calnexins, annexin lectins, discoidins, eel agglutinins, etc. [21].

1.4 BIOLOGICAL PROPERTIES OF LECTINS

Agglutination

The property of lectins to agglutinate the RBCs differentiates them from other sugar-binding macromolecules. This unique property of lectins is being exploited in neurosciences, blood typing and other biomedical applications. Further, they have also been utilized in identifying the molecular basis behind blood group specificity. The lectins isolated from *Ulex europaeus* (anti H) and *Dolichos biflorus* (anti A1) are being effectively used in blood banks to differentiate between blood groups [53].

Antitumor properties

The antitumor properties of lectins isolated from varied sources have been explored based on their ability to specifically recognize the glycan structures on the cancer cell surface. Also, preclinical and clinical trials of a number of plant lectins have been implemented so as to determine their potential as antineoplastic drugs for cancer treatment [54].

Anti-bacterial effects

Plant and animal lectins are known to play a natural role in host defense against microorganisms. Plant lectins strongly interact with the bacterial cell wall peptidoglycans, thus preventing the entry of bacteria into the cytoplasm [55]. While the antibacterial effects of some animal lectins rely on their pore-forming ability resulting in permeabilization of bacterial membrane, others cause bacterial autophagy [56,57].

Antiviral effects

Lectins are potent inhibitors of animal and human viruses, *in vitro*. Several plant lectins such as WGA, Con A, PSA, Vicia faba agglutinin were found to interact with the Human immunodeficiency virus 1 (HIV-1) gp120 envelope molecule, thereby, inhibiting fusion of HIV infected cells with the $CD4^+$ cells [58]. Likewise, animal lectins inhibit the pervasion and replication of virus by recognizing the pathogen associated molecular patterns (PAMPs) on the virus.

Antifungal effects

Most plant lectins interact with the chitin present on the fungal cell wall and are known to inhibit growth of several non-pathogenic and phytopathogenic fungi [59-61]. In some cases, plant lectins were shown to bind to the hyphae resulting in prevention of spore germination [62].

Mitogenic stimulation of lymphocyte

Lectins are capable of stimulating the non-dividing lymphocytes to proliferate. PHA isolated from red kidney bean was the first lectin that was shown to stimulate lymphocytes to undergo mitosis [63]. While PHA and Con A were found to selectively stimulate the proliferation of T lymphocytes, pokeweed mitogen induces the proliferation of both T and B lymphocyte [64].

Toxicity

Certain lectins such as SBA, Con A, PHA and WGA have been foundto be toxic to mammalian cells, both *in vitro* and *in vivo*; for instance, WGA was

found to exhibit anti-nutritive effects and induce hyperplastic growth of rat gut and pancreas [65]. Moreover, WGA and PHA were also found to have detrimental effects on the intestine [66].

1.5 DIETARY LECTINS

Lectins that found in dietary sources such as jackfruit, peanuts, Kidney bean, soybean, tomato, banana, pea lentils, wheat, potato, mushroom and corn are referred to as dietary lectins. They belong to a unique group of carbohydrate-binding proteins which possess the ability to agglutinate the RBCs from diverse species [1]. Dietary lectins are known to increase the permeability of the gut, thereby allowing transit of gut-derived and dietary bacterial antigens into the periphery. As most dietary lectins possess a tightly globular structure that exhibit resistance to heat, extreme pH and to digestion by proteases, they are capable of retaining their biologically active form, even after traversing via the systemic circulation, eg., active form of PNA was identified in the human blood, minutes after consumption and has also been recovered from fecal sample [67]. Also, biologically intact form of WGA, an N-acetylglucosamine binding lectin, has been detected in fecal collections and in the ileostomy effluent.

As plant lectins are consumed as part of our food, it is essential to figure out the outcome of their binding on to the surface of the mammalian cells. Many lectins isolated from different plant sources exhibit specificity towards different carbohydrates and have been shown to have a pronounced effect on cancer cell proliferation [68]. Hence, after ingestion, it is of utmost significance to know as to what will be the direct and indirect effects of these lectins.

A number of studies are underway to reveal the consequences of binding of dietary lectins to normal and malignant cells. These lectins are known to non-covalently bind to the specific glycoconjugates present on normal and transformed cell surface, thereby affecting the cell proliferation. The binding of these lectins will have a pronounced effect on the subsequent

downstream signaling of molecules which are essential for proliferation of cells.

1.6 THE THOMSEN-FRIEDENREICH DISACCHARIDE (TFD) BINDING LECTINS

The TF-antigen (Galβ1-3GalNAcα1-Ser/Thr) also known as T antigen, is known to be expressed only during the development of fetus. The expression of T antigen in case of normal, healthy adults is still unclear. In normal epithelium, the TF-antigen is usually masked by sialic acids or sulfate and is expressed on the glycoproteins and mucins present on the tumor cells [69]. The TF-antigen has been shown to be overexpressed in 90% of all human cancers including the cancers of the breast, liver, stomach, colon, prostate and bladder. In most cases, the increased expression of TF-antigen corresponds to tumor progression and metastasis [70]., for instance, patients with colon cancer who are tested positive for TF-antigen are at an increased risk of liver metastasis. Likewise, the expression of TF-antigen was shown to be higher in invasive bladder cancer as compared to non-invasive cancer [71]. The potential use of TF-antigen in cancer immunotherapy and cancer diagnosis have also been explored. In several clinical trials, vaccination using synthetic TF were demonstrated to induce complement-mediated killing of tumor cells and also resulted in prolonged survival in cancer patients [72].

The TFD-binding lectins are a remarkable class of sugar-binding proteins that are known to selectively bind to the TF-antigen. Jacalin, PNA, *Agaricus bisporus* lectin (ABL), Maclura pomifera agglutinin, amaranthine and the heat labile *E. coli* enterotoxin are some of the well-known TFD-binding lectins. These lectins are given substantial emphasis due to their potential diagnostic and therapeutic value. The binding of these lectins on the surface of the cancer cell membranes regulate a number of signaling pathways resulting in mitogenic, antiproliferative, autophagic or apoptotic effects on cancer cells both *in vitro* and *in vivo*. Certain TFD-binding lectins such as Moringa G and jacalin have also been used to specifically target the sugar-binding receptors present on the tumor cells during photodynamic treatment

of cancer cells. Jacalin is one such dietary, TFD-binding lectin which has been the focus of several studies mainly because of its ease of purification, abundance of source material, yield and stability.

1.7 JACALIN

Jacalin is a tetrameric lectin purified from the seeds of jackfruit (*Artocarpus integrifolia*). It is known to specifically bind to the tumor-associated disaccharide, TF-antigen [73]. It possesses the ability to agglutinate red blood cells of different species including human, mouse, buffalo, rabbit, pigeon and duck [74]. At the monosaccharide level, though jacalin essentially was known to be galactose-specific, it has also been shown to readily interact with mannose, glucose, N-acetylmuramic acid and N-acetylneuraminic acid [75].

1.7.1 Structure of jacalin

Jacalin is a tetramer that is made up of a heavy α chain and a light β chain of 133 and 20 amino acids respectively [76]. The α and β chains are non-covalently associated with each other forming a β-prism monomeric structure αβ [77]. Approximately one-third of α subunit is glycosylated and is referred to as α' subunit. The molecular weight of jacalin was reported to be 66,000 Da, as determined by native PAGE at pH 4.3 and analytical ultracentrifugation studies [78,79]. The amino acid sequences of α and α' chains were almost similar and produced two distinct bands when separated by SDS-PAGE [80,81]. The molecular mass of α' chain as estimated by mass spectrometry was found to be 15,880 Da [82], while the molecular mass of the glycosylated α subunit was found to be 14,662 Da as calculated based on the amino acid residues. Further, the electron micrograph of jacalin suggests that it occurs in the form of the usual symmetrical tetramer even in solution [79].

1.7.2 Applications of jacalin

Owing to its diverse biological applications, jacalin has been the focus of various studies. As jacalin is selectively mitogenic for $CD4^+$ T cells, it has been used in evaluating the immune status of patients with HIV-1 infection [73,83]. Jacalin has further been used in isolation of IgA1 and other plasma glycoproteins. Moreover, Jacalin-based ELISA was developed to determine the level of IgA1 in biological samples. The ability of jacalin to bind to porphyrin rings has been exploited in photodynamic therapy [84]. Jacalin has further been used as a carrier protein for delivery of anti-cancer molecules that were coupled to nanoparticles [85,86]. Besides, jacalin has also been used as a tool in histochemistry to detect tumors and glycoconjugates.

1.8 RATIONALE OF THE STUDY

Lectins are abundantly present in foods such as legumes and grains and hence are an inevitable part of our diet. As dietary lectins retain their intact, biologically active form even after traversing through the gastrointestinal barrier [66], once ingested, the consequences of their effects are unavoidable, eg., the capability of kidney bean lectin to function as a toxin is dependent on the increased rate of uptake of the lectin by the epithelial cells [87]. Further, tomato lectin was detected in the capillary vessels of small intestinal villi, thereby signifying that the lectin enters into the circulation from the gut lumen [88]. Also, biologically functional form of some lectins have been recovered from the feces of humans and rats that were fed with diet rich in lectin [89]. The presence of lectins in our everyday diet emphasizes the need to understand the functional relationship between lectins and their effects on normal and transformed cell proliferation. Further, upon interacting with the components of the immune system, dietary lectins have been shown to modulate the secretion of inflammatory cytokines [90]. Henceforth, it is significant to further investigate and gain knowledge regarding the mechanisms underlying the effects of dietary lectins on cell proliferation and immunomodulation.

1.9 GENESIS OF THE CURRENT STUDY

Jacalin is known to undergo irreversible thermal denaturation, resulting in aggregation. Considering its potential applications, it becomes important to sustain the functional stability of the protein at the time of storage, transport and for longer use. Henceforth, the primary focus of the current research was to identify additives that can prevent thermal aggregation of jacalin thus preventing loss of its activity. Further, in view of the contrastive effects of the TFD-binding lectins, the effects of jacalin on cancer cells of different lineages were analyzed and a possible mechanism underlying the mitogenic facet of jacalin has been proposed. As dietary lectins modulate the immune functions and are capable of polarizing the immune response to inhibit or stimulate tumorigenesis, the ability of peripheral blood mononuclear cells (PBMCs) to proliferate and produce cytokines in response to jacalin treatment were also investigated.

1.10 OBJECTIVES OF THE CURRENT STUDY

The specific objectives of the current research work were:

1. Screening various additives for their ability to prevent thermal aggregation and retaining the hemagglutination activity of jacalin, purified from jackfruit seeds.
2. To study the effects of jacalin on cancer cells that belong to different lineages and to elucidate the underlying mechanism.
3. To study the effects of jacalin stimulation on the PBMCs.
4. To review the immunomodulatory effects of jacalin-stimulated PBMCs on the cancer cells.

1.11 OUTLINE OF THE THESIS

The contents of the thesis has been divided into nine chapters

Chapter 1 – Introduction

In this chapter, a detailed account of the fundamental concepts followed by the rationale behind the present research work is described.

Chapter 2 – Literature overview

This chapter provides a detailed review of literatures pertaining to the various biological effects of lectins, including anticancer and immunomodulatory effects.

Chapter 3 – Screening for additives for their ability to enhance stability of jacalin, against thermal aggregation

This chapter provides a detailed account of affinity purification of jacalin from jackfruit seeds followed by an investigation of the effects of various additives on thermal-induced unfolding and aggregation of jacalin.

Chapter 4 – In vitro effects of jacalin on cancer cells that belong to different lineages

In this chapter, the consequences of binding of jacalin to cancer cells of different lineages were assessed, with emphasis on the mitogenic effects of jacalin on K562 erythroleukemia cells.

Chapter 5 – An investigation into the mechanism underlying the mitogenic effects of jacalin

As most studies lay emphasis on identifying the molecular mechanism that is responsible for the anti-proliferative effects of lectins, it was intriguing to shed light on the molecular mechanism underlying the mitogenic activities of jacalin on K562 cells. Thus, in this chapter, the potential target of jacalin was identified and the possible mechanism that may be responsible for the mitogenic effects of jacalin has been elucidated.

Chapter 6 – Immunomodulatory effects of jacalin on the PBMCs

This chapter gives a detailed account of the immunomodulatory effects of jacalin on PBMC proliferation and cytokine production. Further, the effects of jacalin-prestimulated PBMCs on cancer cell proliferation have been investigated.

Chapter 7 – Summary and Conclusions

This is the concluding chapter which briefs the significant observations of the research work.

Chapter 8 – Scope for future work

This chapter provides a detailed account of the future prospects of the present work.

Chapter 9 – References

This chapter provides the compiled list of references.