Mucositis is the painful inflammation and ulceration of the mucous membranes. Mucosal toxicity, post cancer chemotherapy, manifested by the disruption of mucosal barrier all along the orodigestive tract is referred to as alimentary mucositis. It affects the mucosa, causing painful ulcerations, dysphagia, abdominal pain, bloating, vomiting, and diarrhoea (1-3). Diarrhoea associated with gastrointestinal mucositis (GIM) has been shown to be related to both structural and functional changes in the small intestine and colon such as crypt hypoplasia and altered absorptive function (4, 5). It also alters the normal flora of the gastrointestinal tract (6), mucin composition and goblet cell distribution (7). Even though the general mechanisms of mucositis are thought to be similar irrespective of the location, the kinetics of symptom development is region specific which is probably governed by the local epithelial cells turnover (1, 8). Previously chemotherapy induced gastrointestinal toxicity was thought to be an epithelial phenomenon but now it is vastly acknowledged that mucositis pathobiology is multifactorial including the connective tissue elements and the epithelium (9). The mucosal immune system also plays a vital role in mucositis by mediating the release of pro-inflammatory cytokines (10). The development of effective palliative or preventive measures is essential for the treatment of mucositis.

Oral mucositis affects approximately 40% of patients after the first chemotherapy cycle (11). The global prevalence of mucositis is around 500 000 patients annually (12). Mucositis increases the economic burden, hospital stay, readmission rates and morbidity of the patients (13, 14). Moreover, specific cytotoxic chemotherapeutic agents such as 5-fluorouracil (5-FU) and etoposide are associated with more severe mucositis (15). Mucositis was identified as the single most debilitating side effect in a patient who underwent bone marrow transplantation followed by chemotherapy (16).

Etoposide (VP-16-213), an epipodophyllotoxin interferes with DNA topoisomerase II and prevents resealing of the DNA break, which results in cell death (17). Cells in the S and G2 phases are highly susceptible to etoposide. It is administered orally or through intravenous injection. Approximately 40% of administered dose is excreted intact through urine. Etoposide is primarily used for treatment of testicular tumors. It
Introduction

is also combined with bleomycin and cisplatin or ifosfamide and cisplatin for small cell lung carcinoma. Etoposide is effective against non-Hodgkin's lymphomas, acute nonlymphocytic leukemia and kaposi sarcoma associated with acquired immunodeficiency syndrome. Etoposide is frequently used together with ifosfamide and carboplatin for high-dose chemotherapy in treatment of leukemia & lymphoma (18). Besides the general toxicities, nausea, vomiting, stomatitis and diarrhoea complicate treatment in nearly 15% of patients (19).

Many strategies such as antibiotic administration, parenteral analgesia and nutrition supplementation are used for management of mucositis (20, 21). Nonetheless, switching over to an alternate regimen remains the most effective means to limit the actual incidence and severity. This strategy is attendant with diminished efficacy of anti-cancer therapy, hence may not be viable, particularly when salvage or total ablation therapy is required. Currently available treatments such as topical anaesthetics, mucosal barriers, immune-modulatory agents and mucosal coating agents like sucralfate can limit the severity of mucositis but cannot prevent it completely. Excess liberation of free radicals coupled with induction of inflammation trigger the early events of mucositis. Prophylactic therapy with antioxidants or anti-inflammatory agents can partially limit some of these ill effects (12).

In 2006, the mucositis study section of the Multinational Association of Supportive Care in Cancer and the International Society for Oral Oncology (MASCC/ISOO) advocated the use of cryotherapy, and/or laser therapy for management of melphalan, 5-fluorouracil induced alimentary mucositis (22). In addition, some innovative new drugs including palifermin, the recombinant keratinocyte growth factor 1 (KGF-1) is used to treat high dose chemotherapy associated mucositis (23). Furthermore, opioid analgesics used in the management, hamper the quality of life because of their untoward effects (16). Recent studies emphasized the use of hormones, cytokines, growth factors that alter epithelial metabolism, thereby reducing its susceptibility to mucositis (12). A Cochrane review published in February 2013 showed that coating agent such as sucralfate is effective in reducing the severity of mucositis. Amifostine, granulocyte growth factor, polymixin, tobramycin, amphotericin, laser, aloe vera and honey prevented oral mucositis in patients receiving cancer treatment (24). Vis-a-vis
with the progress achieved over the years, management of chemotherapy induced mucositis prevention still remains as a challenge for medical practitioners (24, 25). Hence there is a necessity to develop novel, promising anti-mucositis entities for the prevention / treatment of this debilitating adverse effect.

Almost 80% of cancer patients depend on complementary and alternative medicine remedies along with the conventional anticancer therapies (26, 27) and many of them use plant derived products (28, 29). Indian traditional medicine offers empirical herbal formulae for treating mouth ulcers, stomatitis and diarrhea which have frequently been used in complementary treatment of oral mucositis in the last decades without solid scientific rationale (30). Preliminary studies with PV701- a milk-derived growth factor extract, vitamins (A, B12, E), licorice and curcumin could not provide enough support to incorporate them as alternative agents (22). The incomplete evidence coupled with the rising importance of alternative medicine provides impetus for further studies (31). However, above said treatments have only been reported to partially prevent intestinal mucositis. As such more effective treatment strategies are sought.

*Spondias pinnata* is a deciduous tree distributed in India, Sri Lanka and South-East Asian countries. In India it is commonly seen in the semi-evergreen forests of the Western Ghats. The genus *Spondias* includes 17 described species, 7 of which are native to the neotropics and about 10 are native to tropical Asia. Phytochemical screening of methanol extract of *Spondias pinnata* bark powder showed the presence of reducing sugars, flavonoids, peptides, phenolic compounds and tannins with high antioxidant and free radical scavenging activities (32).

Whey is the liquid remaining after milk has been curdled and strained to remove the casein (curds). It contains proteins, lactose, vitamins, minerals and traces of fat. Whey protein, which represents 20% of the total milk protein, is sold as a nutritional supplement. Whey is a source of major proteins like lactoglobulin, lactalbumin, glycomacropeptide, proteose, immunoglobulins and serum albumin which together make up 85% of whey protein. Whey proteins have been reported to have effects on bone, muscle, blood, brain, pancreas, immune system, cancer, infection, metabolism, wound healing, learning and aging (33).
Introduction

Hence the present study hypothesizes that the constituents of the whey preparation and *Spondias pinnata* bark extract would be effective in etoposide induced intestinal damage in a rat model of mucositis potentially through their modulatory effects on inflammation and other actions.