Traditional medicine and biological applications of *Aegle marmelos* (*L.*) -An updated review

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***Abstract*—** *Aegle marmelos (L.)* or golden apple has long been used in traditional medicinal system around Southeast Asia, and also Ayurveda and may contain valuable compounds for industrial applications. The various health benefits of Bael, traditional, therapeutic potential including antidiabetic, toxicity activity, anticancer activity, wound healing activity, antioxidant and hepatoprotective, antimicrobial activity, SARS-COVID-2, diarrhea and dysentery capabilities, are attributed to its biologically active constituents, including alkaloids, coumarins, flavonoids, and essential oils. Bael's therapeutic capability in various diseases in the pharmaceutical industries is to develop Bael extracts and compounds as drugs for the treatment of diabetes, gastrointestinal problems, or skin diseases. The present information presented here will be useful for researchers, healthcare professionals, and pharmaceutical companies to design and development of effective biologically active ingredient application drugs against antibiotics, antimicrobials, and anticancer activities.
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***Keywords*—** Antidiabetic, Antimicrobial, Traditional medicine, Golden Apple

I. INTRODUCTION

Medicinal plants are used in traditional medicine in India and other Asian countries. Among the medicinal plants, *Aegle marmelos (L.)* Correa (family Rutaceae), commonly known as pal in North India and Soth India, has been used in traditional medicine in India for over 5000 years[1]. Traditional medicine in India has a long history. Indian medical literature contains a wealth of knowledge about folklore and traditional properties of natural ingredients that are therapeutically beneficial. Ayurveda, Siddha and Unani are some of the systems used in Indian traditional medicine[2-13]. It is native to India, Nepal, Bangladesh and Pakistan, but is also found in Sri Lanka, Thailand, Malaysia, Cambodia, the Myanmar, Java, Philippines and other Southeast Asian countries [14-15]. Pal has different names in various Indian languages, including Bilva, Shivaphala in Sanskrit; Belo in Oriya; Bel Beli, Belgiri in Hindi; Bel in Assamese and Marathi; and in India, Vilva Maram in Tamil. Other names for Bael include Be Li in Sinhalese, Matoom in Thai, and Bela in Spanish. There are many more vernacular names for A. mermalos (L.) in various countries[16] (Rasool & Dehghan, 2022). The word "bael" has many meanings and names in various countries. "Bilva," "Shivaphala," "Belo," "bel," "Vilvamaram," "Golden Apple," "A wooden Apple," "Bengal Quince," and "Indian Quince" are some examples[17]. According to legendarily, it also has great religious significance in Hindu rituals. The Triphadra, or "sacred leaf," of this sacred tree has made significant contributions to Lord Mahadeva[18].

A. marmelose contains various phytochemicals such as alkaloids, flavonoids, saponins, marmeline, limolin, tannins, phenolics, coumarins, etc. Compounds extracted from A. marmelos have various applications such as treating diarrhea, fighting cancer, preventing diabetes, healing ulcers, and preventing obesity. Almost all parts of the plant have been found to participate in the production of various pharmacological activities. Traditional medical practice, widespread throughout the Asian continent, uses this plant to prevent and cure various types of diseases. In addition, this plant exhibits antidiarrheal, anti-

inflammatory, anticancer, antimicrobial, antiviral, antidiabetic, and antioxidant properties. Compounds extracted from *A. marmelos* have various applications such as treating diarrhea, fighting cancer, preventing diabetes, healing ulcers, and preventing obesity. Unripe pele fruit juice helps in the treatment of hemorrhoids and piles[19]. Ear infections, persistent inflammation, and pus discharge can all be treated with roots soaked in neem oil[19] Sesilin isolated from the leaves of A. marmelos has the potential to treat COVID-19 [20].

Methodology

Research papers were searched using various keywords such as *Aegle marmelos*', 'traditional medicines', conventional medicine, folk medicine, 'phytochemicals', 'bioactive compounds', 'therapeutic', 'pharmacology', 'bale', 'nutritional value', ' 'anticancer', 'antidiabetic' and 'antimicrobial' in various databases such as Google Scholar, Science Direct, Scopus, Springer Link and Web of Science. The papers considered for review included papers that discussed the therapeutic potential of bael, the phytochemical composition of various plant parts and its phytochemical potential. Articles published in reputed journals such as Springer, Science Direct, Taylor & Francis, Wiley Online Library and Societies in the last decade were included in the discussion, while those published by other lesser-known publishers were not considered a priority. Although, some important articles published before the period under consideration, but essential to our discussion, were also included. Information was extracted from the original articles to understand the research in this field, however, some notable review articles were also included. Only the most relevant articles that met the purpose of this article were considered for discussion.

Traditional uses of *A. marmelos*

All parts of the A. marmelos plant, such as the fruit, stem, bark and leaves, are useful because they have therapeutic and nutritional properties and are of commercial importance. Its fruit, bark, leaves, flower, root and seed are effective in treating a wide range of diseases in humans. Several types of tablets, pastes and powders are prepared from this plant. Bael is an important ingredient in the preparation of Dashmula (meaning 'ten roots'; it cures colitis, dysentery, flatulence and fever), Chyawanprash etc.,[21]. The unripe fruit and leaves are important components in the treatment of conditions such as recurrent diarrhea, dysentery, and diabetes insipidus in Ayurveda. Diabetics take the juice of the Bael leaves of the plant early in the morning on an empty stomach, and it is said to reduce sugar levels within a month[22]. A well-balanced combination of the five components of *Aegle marmelos* namely leaf, flower, fruit, root and bark, can be found to be a powerful treatment for many mental illnesses. The fruit powder also has anti-proliferative and anti-cancer properties[23-25]. The unripe mature fruits are astringent, and when they ripen, the edible pulp inside the fruit changes its color to yellow/orange, producing a significantly sweeter taste with a floral aroma and sweet taste, and is consumed raw [26]. In South India, the juice is given to patients to relieve shortness of breath and respiratory distress. The ripe fruit is used to make sharbat[22].

Therapeutic potential of *A. marmelos*

A. marmelos is one of the primary medicinal and nutritional plants. Studies have also shown its multiple medicinal uses such as antibacterial, antiviral, antidiarrheal, antiulcerative, antioxidative, hepatoprotective, antidiabetic, and anticancer effects. The use of medicinal plants such as AM in traditional medicine is guided by its protective factor.

Antidiabetic activity of *A. marmelos*

In a streptozotocin-induced diabetic rat model, the aqueous extract of A. marmelos fruits reduced blood sugar. It increased insulin secretion by partially regenerating β-cells of pancreatic islets[27–34]. The effects observed in the treated fruit juice were superior to those observed in animals treated with glibenclamide. In vitro evaluation of the current study showed a potent antidiabetic effect from the lectin extract, as measured by glucose uptake in yeast cells[35]. A fruit lectin extract with an IC_{50} of 3.36 µg/ml was more effective than the standard drug metformin in increasing glucose uptake by yeast cells. In this study, A. marmelos fruit extract was found to reduce hypoglycemia, which may be due to its antioxidant activity and high content of active components [36]. Consequently, different parts of the A. marmelos

plant may be beneficial as part of a healthy diet and in the development of antidiabetic drugs. The active components in the leaf and callus extracts reduced blood sugar levels in STZ-diabetic rabbits, and the methanol extract of A. marmelos callus powder was as potent as the leaf extract in the treatment of diabetes [37]. This studypreferable that aqueous seed extract of A. marmelos reduced blood glucose levels in normal and severe diabetic rats and improved glucose tolerance in sub and mild diabetic rats and also consistently reported as tolbutamide. Alcoholic extract of A. marmelos leaves significantly reduced α-amylase and α-glycosidase enzymes with IC_{50} values of 46.21 and 42.07 µg/ml, respectively. A. marmelos significantly reduced the increased ROS levels due to high glucose and increased glucose consumption[38].

Toxicity study of *A. marmelos*

A. marmelos dry fruit pulp is being investigated for its topical properties. Swiss albino mice were tested for acute oral toxicity with ethanolic extract of A. marmelos dry fruit pulp at doses of 550 and 1250 mg/kg. The test results should indicate that the extract is not toxic at these doses. The behavior and physiological activity of the mice remained unchanged throughout the test (14 days)[39]. The findings show that the LD_{50} of the test extract is very significant. No toxic signs, behavioral changes, or mortality were shown in the oral acute toxicity study at doses of 1250 mg/kg. Therefore, the ethanolic extract of A. marmelos dry fruit pulp extract did not have a biologically significant toxic effect on mice below the LD_{50}.

Anticancer activity of A. marmelos

The cytotoxicity of methanol and acetone extracts of A. marmelos against HEp-2, MDA-MB-231 and Vero cells was investigated. The IC_{50} of methanol extract of A. marmelos was 47.08 g/ml and the IC_{50} of acetone extract of A. marmelos was 79.62 g/ml, indicating that HEp-2 cells were more sensitive to it. Both extracts of A. marmelos were toxic to cancer cells; although, Vero cells survived for 24 hours[40–41]. MTT assays confirmed the in vitro anticancer activity in the human breast cancer cell line MCF-7 at difference concentrations. Flavonoids in fruit extract act as a potential reducing agent and are reasonable for the formation of gold nanoparticles[42]. The aqueous fruit pulp extract from A. marmelos caused the most effective MCF7 cell death at 100 g/ml and IC_{50} at concentrations of 47.92 g/ml [43]. In an in-vivo study, Swiss albino mice infected with Ehrlich ascites carcinoma were given an intraperitoneal injection of 400 mg/kg hydroalcoholic extract of A. marmelos. The median survival time after tumor inoculation was significantly increased to 28 days compared to the control group injected with saline[44]. The ethanolic extract of A. marmelos fruit pulp has antiproliferative effects by inhibiting the proliferation of breast carcinomas in a rat model. Both breast tumor volume ($p < 0.05$) and different blood biomarkers ($p < 0.0001$) were significantly reduced after A. marmelos treatment[45].

Wound Healing Activity of *A. marmelos*

There are several stages in wound healing, including inflammation, cell proliferation and stiffness during the growth of the collagen matrix. Reddening, pain, and swelling are some of the most common symptoms associated with wounds, and they are also present when inflammation is present. One protective mechanism has been found to involve the release of reactive species. *Aegle marmelos* fruit extract reduced wound size and improved tissue regeneration by inhibiting beta-catenin, Akt signaling, and ERK (extracellular signal-regulated kinase) pathway-regulated keratinocyte migration in Aegle marmelos fruit extract-treated rats. A. marmelos fruit extract and its active ingredient induce mRNA expression, reduce nitric oxide and PGE2 production, and promote the motility of HaCaT keratinocytes during wound healing in cultured rat [46]. When applied to wounds in rats, A. marmelos fruit extract significantly reduced wound size. Re-epithelialization of skin lesions occurred more rapidly than expected. Further investigation of the effects of bale extract using exudate and cut models showed that bael has a positive effect on wound healing. Essential oils, bioflavonoids, and nitrogen-containing organic compounds such as alkaloids and sterols are some of the phytochemical compounds found in bale. These phytocomponents are responsible for the wound healing properties of bael, accelerated epithelialization, wound tightening, flexible power, and hydroxyproline composition. Balsam's ability to promote wound healing is comparable to the ability of the drug nitrofurazone.

Antiulcer activity of *A. marmelos*

Methanolic and aqueous extracts of *A. marmelos* seeds were tested for antiulcer activity in indomethacin-induced ulcer, stress induced ulcer and pyloric ligation-induced ulcer using ranitidine as a standard (50 mg/kg)[47]. Peptic ulcers are caused by the bacterium H. pylori. There is little or no literature on the effect of *A. marmelos* on Helicobacter pylori, so further research is needed to determine its effect on H. pylori. If it positively reduces AMR, it could be an effective herbal medicine to treat ulcers without adverse effects[48]. *A. marmelos* is frequently used in Ayurveda to treat ulcers and related diseases, and oral administration of methanolic extract of *A. marmelos* to rats with lipopolysaccharide induced gastric ulcers caused by Helicobacter pylori has been observed[49]. It was shown in the experiment that 500mg/kg of methanolic extract reduced gastric ulcers by 93.98%. Gastric secretion parameters such as free and total acidity, gastric extract, acid output volume and pepsin concentration were inhibited, resulting in a reduction in gastric ulcers.

Antioxidant and hepatoprotective activity of A. marmelos

Oxidative stress is the result of changes in DNA and lipids caused by the production of various reactive free radicals, which are induced by various chemical and environmental variables[50–62]. The reducing capacity of a chemical is a key indicator of its potential antioxidant activity[1]. The hepatoprotective activity of *Aegle Marmelos* is attributed to its strong antioxidant activity. The liver is a crucial organ in the human body and is involved in the detoxification and elimination of harmful chemicals. Hepatotoxins, xenobiotics, and different chemotherapeutic drugs compromise liver function. Bael leaves have excellent hepatoprotective properties against alcohol-induced liver damage measured using biochemical markers in albino rats[63-64]. This was demonstrated by a decrease in lipid peroxidation products, an increase in antioxidant enzyme activity, and glutathione [65]. Similarly, extracts from the pulp and seeds of the bael fruit have been reported to protect against CCl_4-induced liver cytotoxicity. These fruits and seeds, when administered in an aqueous preparation, induced 80% and 70% protection against CCl_4 toxicity, respectively. Rajashree et al.,[66] reported that the consumption of a herbal medicine derived from Aegle marmelos had a protective effect by inhibiting oxidative stress induced by free radicals and enhancing the elevated catalase activity in liver tissue[67].

Antimicrobial activity of *A. marmelos*

Antimicrobial drugs are a class of drugs commonly used to treat water- and food-borne diseases. Substances with antibacterial properties that occur in many therapeutic plant extracts can target pathogens. Various studies have demonstrated the antibacterial activity of A. marmelos extract. The bael extract also showed activity against *Proteus vulgaris*, *Staphylococcus epidermidis*, and *S. aureus*. Leaf and flower extracts of *A. marmelos* were tested at various concentrations (50, 100, and 200 ppm) against five clinical pathogens (S. *aureus, P. aeruginosa, Proteus mirabilis, E. coli*, and *Salmonella typhi*). E. coli was the most sensitive pathogen to the treatment[68–78]. Furthermore, bael leaf oil showed variable levels of inhibition against various fungal isolates, including the resistant fungus *Fusarium udum*, in a concentration- and time-dependent manner. Bael leaf essential oil may interfere with the metabolism of Ca+2 dipicolonic acid. Therefore, it inhibits the growth of fungal vegetative bodies on solid media or within the host, which suppresses fungal activity[79]. Antimicrobial and anthelmintic properties have also been reported against non-saponifiable matter and pure *Aegle marmelos* oil[80]. According to Kanal et al., extracts obtained from various parts of bael have been shown to inhibit the growth of several bacterial strains. Similarly, Choudhury et al., [81] evaluated the antifungal effects of *A. marmelos* leaves and fruits. The new findings support the ancient value of *A. marmelos* fruit as an herb for the treatment of various diseases, which was confirmed by a recent study that showed that the crude methanolic extract of the *A. marmelos* fruit exhibited potent and effective antimicrobial activity against various Gram-positive and Gram-negative bacterial pathogens [82].

Activity against SARS-COVID-2

Seselin, a chemical compound derived from the leaves of *A. marmelos*, was tested against several SARS-CoV-2 targets, including the viral spike protein S2, the COVID-19 major protease, and the non-enzymatic enzyme SARS-CoV-2 (2019-nCoV). Seselin leap to receptors with a binding energy of 6.3 kcal/mol, spike protein S2 with a binding energy of 6.9 kcal/mol, and the COVID-19 major protease with a binding energy of 6.7 kcal/mol. As pridict by a docking study with three definite receptors, intermolecular hydrogen bonds and stacking interactions are responsible for stabilizing the complexes with the lowest planned energy. Results from molecular dynamics simulation and MM/PBSA (molecular mechanism/Poisson Boltzmann surface area) study confirmed that seselin binds to its target receptors and has therapeutic potential for COVID-19. Marmin, isolated from A. marmelos, was shown by an in silico investigation to be a potential inhibitor of RNA-dependent RNA polymerase and 3CLpro of SARS-COVID-2[20,83].

Diarrhea and dysentery

Both ripe and unripe bael fruits are considered to be excellent products for treating diarrhea and dysentery. Constipation, diarrhea, and chronic diarrhea can all be usefully treated with the fruit powder[84]. A. marmelos decoction can help manage numerous infectious diarrheal diseases caused by various microorganisms, as well as rotavirus infections and giardiasis to a certain extent. At 400 mg/kg, A. marmelos showed significant antidiarrheal effects in rats with castor oil-induced diarrhea, which was almost equivalent to the common drug loperamide [85]. The primary antidiarrheal components found in the fruits of the bael, such as tannins, flavonoids, and mucilaginous compounds, have been shown to have antibacterial, intestinal, antisecretory, motility properties[86]. Antidiarrheal activity was demonstrated by the ethanolic extract from the dried fruit pulp of Aegle marmelos. While S. dysenteris showed moderate activity, Shigella boydii, S. sonnei, and S. flexneri showed significant activity. When evaluated in vitro, the chloroform extract of Aegle marmelos root showed strong bactericidal activity similar to ciprofloxacin [87]. Moreover, by inhibiting intestinal peristalsis, diarrhea was reduced in rats.

Conclusion: In the current paper, *Aegle marmelos* is discussed in terms of its traditional uses, therapeutic and biological applications studies. A. marmelos is a used in ayurvedic and traditional medicine to treat diseases like therapeutic potential including antidiabetic, toxicity activity, anticancer, wound healing activity, antioxidant and hepatoprotective, antimicrobial activity, SARS-COVID-2, diarrhea and dysentery. Therefore, a few researchers available for this species in terms of traditional, therapeutic and biological and pharmacological evaluation of antidiabetic, antioxidant, antimicrobial, anticancer, SARS-COVID-2 activity.

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