Review Article

Moringa oleifera: Resource Management and Multiuse Life Tree

ABSTRACT

Moringa oleifera Lamarck (*Moringaceae* family) is a plant native from the Western and sub-Himalayan parts of Northwest India, Pakistan and Afghanistan. This species is widely cultivated across Africa, South-East Asia, Arabia, South America and Caribbean Islands. *M. oleifera* culture is also being distributed in the Semi-Arid Northeast of Brazil. It is a multiuse life tree with great environmental economic importance in industrial and medical areas. This review reports different purposes of *M. oleifera* including sustaining environmental resources, soil protection and shelter for animals. This plant requires few cares and distinct parts have bioactive compounds. Moringa tissues used in human and animal diets, also withdraw pollutants from water. The seeds with coagulant properties applied to water treatment for human consumption, remove waste products like surfactants, heavy metals and pesticides. The oil extracted from seeds is used in cosmetic production and as biodiesel. *M. oleifera* tissues also contain proteins with different biological activities, including lectins, chitin-binding proteins, trypsin inhibitors, and proteases. The lectins are reported to act as insecticidal agents against *Aedes aegypti* (vector of dengue, chikungunya and yellow fevers) and *Anagasta kuehniella* (pest of stored products) and also showed water coagulant, antibacterial and blood anticoagulant activities. The presence of trypsin inhibitors has been reported in *M. oleifera* leaves and flowers. The inhibitor from flowers is toxic to larvae of *A. aegypti*. The flowers also contain caseinolytic proteases that are able to promote clotting of milk. In this sense, *M. oleifera* is a promising tree from a biotechnological point of view, since it has shown a great variety of uses and is source of several compounds with a broad range of biological activities.

Keywords: Moringa oleifera; water treatment; bioactive proteins; lectins; trypsin inhibitor; proteases.

1. INTRODUCTION

Moringa oleifera (Figure 1A and 1B) is distributed over countries around the world and widely cultivated across Africa (e.g., Nigeria, Senegal, Tanzania), South America (e.g., Semi-Arid Northeast of Brazil), Central, Southeast and South Asia (e.g., Afghanistan, Malaysia, Indonesia, Pakistan, Bangladesh), India, Arabia, Pacific and Caribbean Islands [1, 2, 3, 4]. It is an arboreal, perennial and fast growing plant which can reach 7-12 m of height, sometimes even 15 m [5, 6]. Moringa flowers (Figure 1C) and seeds (Figure 1D) are produced from the first year and there may be multiple seed harvests in many parts of the world [7]. *M. oleifera* has white flowers, with unequal petals and slight odor [8]; the pollination, pollen germination and stigma receptivity of *M. oleifera* flowers were studied. Successful pollination of moringa flowers requires large number of insects' visitations; among them are individuals from the orders Thysanoptera (*Haplothrips ceylonicus*), Hymenoptera (*Xylocapa* sp. and *Apis* sp.), Lepidoptera (Pappilionidae and Pieridae) and Coleoptera. In addition, biochemical studies of stigmas reveal that there are an over expression of proteins and secretion of esterases in receptive stigmas [8].

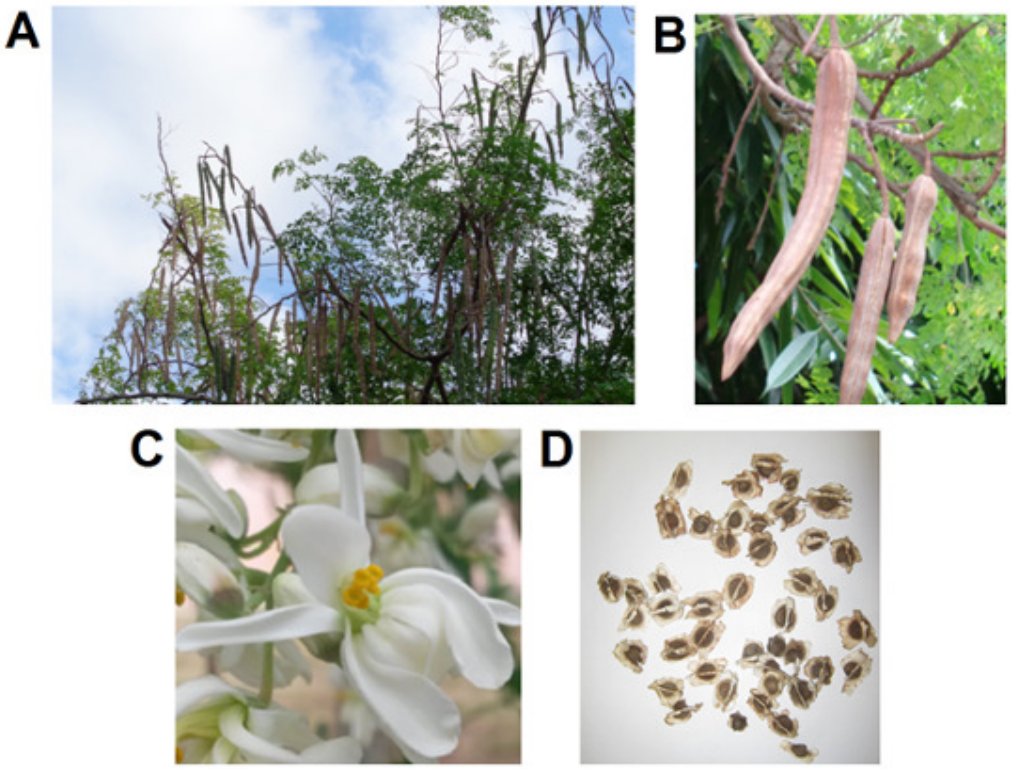

Fig. 1. Aspects of *Moringa oleifera.*
(A) A view of the top of tree crown. (B) Fruits. (C) Flowers (D) Seeds.

Moringa is considered as a pan tropical tree of hot Semi-Arid Regions (annual rainfall 250-1500 mm), which is adaptable to a wide range of environmental situations: from hot dry to hot, humid and wet conditions. It is resistant to light frosts but does not survive under freezing condition. This tree is quite drought tolerant and is well adapted for a wide range of adverse environments that would not be suitable for other fruit, nut and tree crops [9]. The flowers and the fruits from moringa appear twice a year, and seeds (Figure 1D) or cuttings can propagate the tree [6]. India was considered the largest producer of moringa with 42,613 ha, 90% located in the Southern States of Tamil Nadu, Karnataka, Kerala and Andhra Pradesh [10].

2. Moringa oleifera, A MULTIUSE LIFE TREE

M. oleifera is a very important plant whose different properties have promoted its widespread applications. Distinct tissues of this plant can be used for several purposes (Table 1).

Seeds of *M. oleifera* contain water-soluble substances that are undoubtedly the most studied natural coagulants [11]. The specific denomination *oleifera* is due to a 35-45% oil content in the seeds [10]. Moringa tree can produce about 2000 seeds per year. The number of seeds could handle about 6,000 liters of water using a dose of 50 mg/L. The trees, however, can be cultivated to produce about five to ten times this yield (i.e. 10,000-20,000 seeds). This would produce up to 60,000 L of water treated per year [12]. When fully mature, the dry seeds are round or triangular in shape and the kernel is surrounded by a shell with three wings [13].

Moringa seed oil has been applied in cosmetics and is considered a great natural emollient with almost total absence of color and odor, and high oleic acid concentration (>73%) [10]. Also, the seed oil is used as raw material for production of a biodiesel with properties that follow the international biodiesel standards. Studies showed that moringa oil can be used as a fuel in diesel engines, mainly mixed with petrodiesel. The performance of moringa biodiesel is comparable to palm-oil blends biodiesel and petrodiesel fuel. In addition, moringa biodiesel

produced lower exhaust emissions than petrodiesel fuel, so this fuel can replace petrodiesel in unmodified engines to reduce the global energy demand and exhaust emissions to the environment [14]. The oil from moringa seeds, as well as the moringa leaves can be used as a source of antioxidant additives for biodiesels with low oxidation stability [15].

Table 1. Application areas of *M. oleifera* tissues

Tissue	Applications
Seeds	Emollients Cosmetics Biodiesel production Water treatment Fertilizers Animal diet Culinary
Fruits	Human diet
Flowers	Cosmetics Human diet Medicinal Milk clotting
Leaves	Human diet Animal diet Forage Medicinal

The leaves of this plant are historically used as nutritious food and traditional medicine in Asia and Africa; moringa leaves have isothiocyanates that attenuated in vivo inflammation [16]. Also, this tissue showed in vivo and in vitro antioxidant activities suggesting that the regular intake of moringa leaves through diet can protect normal as well as diabetic patients against oxidative damages [17].

M. oleifera leaf extract can mitigate the effects of salinity and cadmium in bean (*Phaseolus vulgaris* L.) plants; these are the most serious abiotic stress factors causing environmental problems and limiting growth and crop productivity [18].

Moringa is able to propagate from seeds even in soils destitute of nutrients with plants requiring minimum attention; as a drought tolerant species, it plays an important role protecting poor soils from the Semiarid Northeastern Brazil [7]. Rivas et al. [19] studied that young *M. oleifera* plants originated from seeds subjected to moderate water deficit showed increased ability for drought tolerance.

The flowers of *M. oleifera* have several medicinal applications and are rich in calcium, potassium and antioxidants, such as α- e γ-tocoferol [20, 5, 6]. In addition, they contain proteases with milk-clotting ability [21]. A water extract from *M. oleifera* flowers, containing tannins, saponins, flavones, flavonols, xanthones and trypsin inhibitor activity, showed molluscicidal activity against embryos and adult snails of the schistosomiasis vector *Biomphalaria glabrata* [22].

3. *Moringa oleifera,* SOURCE OF HUMAN AND ANIMAL FOOD

M. oleifera has been widely used in human and animal diets (Table 1). Different parts of this tree are applied as food to combat malnutrition especially among infants and breastfeeding woman in many developing countries, particularly in India, Pakistan, the Philippines, Hawaii and many parts of Africa [9].

The young leaves, flowers and green pods are common in the diet at Philippines [23]. In Malaysia, the young tender pods, cut into small pieces are added to curries [13]. The young leaves and seeds are rich sources of calcium, iron and vitamin C serving as nutrients for various

communities [24]. Ethanolic and saline extracts from different tissues of *M. oleifera* are potential sources of antioxidants [25]. Moringa fresh seeds after roasting make a palatable dish; also, seeds are consumed after frying and taste like peanuts [6].

Moringa leaves are eaten in Nigeria as vegetables. Leaves are consumed by infants and children in South India since their high content of beta-carotenes could help to prevent the development of blindness by vitamin A deficiency [26]. This tissue, besides being an excellent source of vitamin A, is also rich in vitamins B, C, proteins and minerals [27]. The content of amino acids such as methionine and cysteine is high and the contents of carbohydrates, fats and phosphorous are reported to be low [28]. Teixeira et al. [29] showed that whole leaf flour contained 28.7% of crude protein, 7.1% of fat, 44.4% of carbohydrate, 3.0 mg/100 g of calcium and 103.1 mg/100 g of iron. The protein profile revealed levels of 3.1% of albumin, 0.3% of globulins, 2.2% of prolamin, 3.5% of glutelin and 70.1% of insoluble proteins. Ethanolic extract from *M. oleifera* leaves showed antioxidant activity that was stable in pH 4 and 9; when the extract was stored in the dark at 5 and 25ºC during a 15-day period, it did not show any significant change in its antioxidant property. Therefore, this plant extract is a potential source of dietary antioxidants [30].

Sánchez et al. [31] studied the effect of feeding with different levels of foliage from *M. oleifera* to dairy cows. This study showed that the inclusion of moringa as a protein supplement improved dry matter intake, digestibility of the diet and increased milk production without altering milk composition. Richter et al. [32] suggested that moringa leaf meal could substitute up to 10% of dietary protein in Nile tilapia without significant reduction in growth. Qwele et al. [33] reported that the meat from goat whose diet was supplemented with *M. oleifera* leaves had higher concentrations of total phenolic content and higher antioxidant activity. Nkukwana et al. [34] investigated the effects of dietary supplementation with *M. oleifera* leaf meal as an improving agent on the growth performance, apparent digestibility, digestive organ size, and carcass yield of broiler chickens; the supplementation up to 25 g per kg of feed did not impair nutrient utilization efficiency, but enhanced bird genetic potential for growth performance.

Defatted *M. oleifera* seed meal was used as an additive in sheep diets based on soybean meal suggesting that seeds have potential to improve rumen fermentation without altering the intake and digestibility; the authors found better growth results using 4 g of moringa seed meal per 100 g of soybean meal [1].

Elemental analysis of shelled and non-shelled *M. oleifera* seeds showed that organic matter consists of the six main elements: carbon (C), oxygen (O), hydrogen (H), nitrogen (N), phosphorus (P) and sulphur (S). The shelled seeds contain 55% carbon, 8.5% hydrogen and 6% nitrogen. The remaining 31% consists of oxygen and trace elements. The non-shelled seeds trail closely the shelled seeds in all the elements analyzed with inferior percentage. The shelled and non-shelled seeds contain about 37% and 27% of proteins and 35% and 21% of lipids, respectively; carbohydrates (as oligosaccharides) represent about 5% of the shelled and non-shelled seeds [35].

In many parts of the world, such as in Haiti, the moringa seed oil is used in general culinary and salads, being reported that it has a pleasant taste. The moringa oil resembles olive oil in its fatty acid composition. Moringa oil is highly unsaturated because of the high percentage of oleic acid. Other prominent fatty acids found in oil from moringa were palmitic, stearic and behenic acids. It is liquid at room temperature, pale-yellow in colour and had flavor similar to that of peanut oil. The oil also contains 36.7% triolein as the main triacylglycerol [13].

4. REMOVAL OF POLLUTANTS FROM WATER BY *M. oleifera* SEEDS

Water is an essential and restrictive substance to human life and environmental equilibrium. In addition, it is a central point in a wide cycle that links human beings health and education [36].

The unsustainable use of biological resources and the indiscriminate application of pesticides are dangerous for the environment, human and animal health. Polluted water has frequently led to waterborne disease outbreaks with acute and long-term health effects ranging from diarrhea

to death; also, it is often the main human exposure pathway to carcinogenic organic and inorganic contaminants. During the last few decades, the increase in human population and the several aspects arising from globalization have introduced several additional water pollutants such as, pharmaceuticals, hormones, endocrine disrupting chemicals, viruses, and toxins [37]. These emerging pollutants are a rising problem nowadays, especially due to the fragility of water resources [36].

M. oleifera seeds have been used to remove different pollutants from water (Table 2). Seed proteins are among the molecules responsible for water clarification [38]. The functional groups in the amino acid side chains of these proteins contribute to water clarification and the mechanism of coagulation consists in adsorption and neutralization of the colloidal positive charges that attract the negatively charged impurities in water [39]. Gassen [40] and Gassenschmidt [41] reported that the coagulant active component of *M. oleifera* could be a cationic peptide with a molecular weight between 6 and 16 kDa and isoelectric point at pH 10. Gassenschmidt [42] analyzed the primary structure of this peptide, showing large amounts of glutamine, proline, arginine in a total of 60 residues. Muyibi and Okufu [43] reported that *M. oleifera* coagulant was not effective for treatment of water with low turbidity.

Table 2. Pollutants removed from water by *M. oleifera*.

Plant tissue	Pollutants	References
Seeds	Surfactants	[36]
	Cadmium	[44, 45]
	Arsenic	[46]
	Silver	[47]
	Anionic dyes	[48]
Pods; wood	Zinc	[49, 50]
Wood	Copper	[50]
	Nickel	[50]
Leaves	Lead	[51]
Husks	Chromium	[52]
Pods	Methyl parathion (pesticide)	[53]

Ndabigengesere et al. [35] studied the efficiency and the mechanisms of coagulation promoted by *M. oleifera* seeds in turbid water. The active component was a dimeric protein more efficient in coagulation than aluminum salt and the organic residue formed after the treatment was safe for the environment and 4 to 5 times lower than that found in water treated by aluminum. The moringa coagulant did not alter the pH, was soluble in water, and had a molecular weight of 13 kDa with isoelectric point between pH 10-11. In addition, it was reported that *M. oleifera* seeds may be used shelled and non-shelled; however, shelled seeds were more effective in coagulation. Ndabigengesere and Narasiah [54] observed that the optimal dosage was 0.5 to 1.0 mg/L and the protein was completely soluble in water. Another component was extracted from seeds of *M. oleifera* by using phosphate buffer and ion exchange chromatography [42]. This flocculant is a protein with molecular weight of 6.5 kDa and isoelectric point above pH 10. Comparison of the primary structure with known protein sequences revealed no significant homology.

Okuda et al. [55] extracted a coagulant from *M. oleifera* seeds with 1 M NaCl and coagulation properties 7.4 times higher than the coagulant extracted in water. In 2001, Okuda et al. [56] isolated another component with coagulant properties from saline extracts; this component was not a protein, polysaccharide or lipid but a polyelectrolyte with a molecular weight around 3.0 kDa and optimum pH for coagulation above 8. This coagulant did not increase the concentration of residual organic carbon. Proteins called lectins are also involved in the coagulant activity of *M. oleifera* seeds [38, 57] and will be discussed ahead.

The coagulant power of *M. oleifera* seeds has been applied to remove different components in aqueous solutions and suspensions. Santos et al. [58] reported that the saline extract from *M. oleifera* seeds at a low protein concentration (1 mg/L) can be an interesting natural alternative

for removing humic acid from water. This extract dose did not impart odor or color of treated water. Sengupta et al. [59] studied an aqueous extract from moringa seeds that was effective in reducing the number of helminth eggs in water with high turbidity.

M. oleifera seed extract can also be considered a competitive coagulant agent for the removal of anionic surfactants from aqueous effluents, especially those with long carbon chains. Surfactants are one of the main dangerous and noxious contaminants [36]. Meneghel et al. [44] and Sharma et al. [45] studied cadmium removal from contaminated water using moringa seeds and their cake (obtained after oil extraction) as biosorbents and found that they were effective in remediation of solutions containing Cd. The use of seed cake is a low cost viable option since it is a byproduct. Kumari et al. [46] showed that flour of *M. oleifera* shelled seeds removed arsenic from water bodies. Seeds of *M. oleifera* were also tested as adsorbents for the removal of silver ions (Ag) [47]. A moringa seed coagulant protein was purified and used for treating textile wastewater effluents and was efficient in the removal of an anionic dye, namely Acid Red 88 without increase the total organic carbon (TOC) concentration in the treated water [48].

Biomass from *M. oleifera* pods removed Zn(II) ions from aqueous solutions [49]. Pods of moringa removed methyl parathion pesticide from water; this pesticide is acutely toxic in small amounts and may cause serious health disorders leading to death by failure of respiratory system [53]. Kalavathy and Miranda [50] described that an activated carbon preparation obtained from *M. oleifera* wood removed copper, nickel and zinc from synthetic wastewater; the authors stated that this material has good potential in treating metal laden industrial effluents. Alves and Coelho [52] developed a method for selective extraction and a pre-concentration of chromium in water using moringa husks.

Although moringa seeds have been used as a coagulation reagent for drinking water purification, Al-Anizi et al. [60] studied that significant cytoxicity effects by *Acinetobacter* were observed when the powdered seed concentrations are from 1 to 50 mg/L. The main toxicity is from the insoluble fatty acid components, which would remain in the supernatant. In addition, Rolim et al. [61] reported that the moringa seed extract showed a mutagenic effect by Kado and Ames assays when evaluated at concentration 3-fold higher than that popularly used to treat water; for this reason, the authors highlighted that it is not recommended to increase the amount of material used for water treatment. In other study, Araújo et al. [62] showed that the aqueous seed extract, as utilized by people to treat water, did not cause systemic toxicity to mice at the dose of 2,000 mg/kg; however, extract that is more concentrated was cytotoxic to peripheral blood mononuclear cells. Thus, further investigation of appropriate water purification techniques for rural areas in developing countries should be performed.

5. BIOACTIVE PROTEINS FROM *M. oleifera* TISSUES

It is well known that *M. oleifera* tissues contain proteins with different biological activities, including lectins, trypsin inhibitors and proteases (Figure 2). Different biological activities have been reported for these proteins (Table 3).

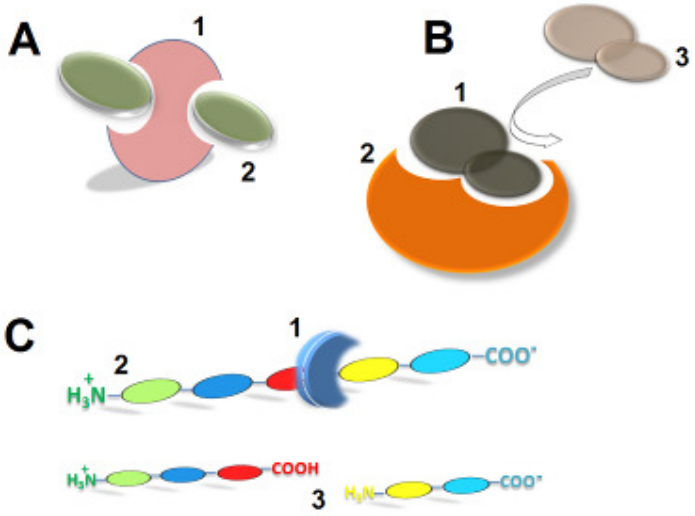

Fig. 2. Bioactive proteins found in *M. oleifera* tissues.
(A) Lectin (1) is a protein able to interact with carbohydrates (2) through reversibly molecular cooperation at specific carbohydrate-binding sites. (B) A protease inhibitor (1) is a protein able to blocking the active site from a proteolytic enzyme (2) and thus preventing the entering of the substrate (3). These molecules can also bind in other regions of the enzyme structure. (C) A protease (1) catalyzing the hydrolysis of a peptide bond, breaking down a protein (2) into smaller peptides (3).

Table 3. Proteins from *M. oleifera* and their biological activities.

Protein	Tissue	Matrix for Isolation	Biological activities	References
Lectins				
WSMoL	Seed	Chitin	Insecticidal activity Capture of *A. aegypti* eggs Antibacterial activity Water coagulant activity	[63] [64] [65] [57]
cMoL	Seed	Guar gel	Water coagulant activity Insecticidal activity Blood anticoagulant	[38] [66] [67]
MoL	Seed	Ion exchange matrices	-------	[68]
Trypsin inhibitors				
Leaf inhibitor	Leaf	Sephadex G75	Antifungal activity	[69]
MoFTI	Flower	Trypsin-Agarose	Insecticidal activity Antibacterial activity	[70] [71]
Chitin-binding proteins				
MoCBPs	Seed	Chitin	Antifungal activity	[72]

Lectins are proteins of non-immune origin, containing two or more binding sites for carbohydrates. These molecules have the ability to agglutinate cells such as erythrocytes (hemagglutination), lymphocytes, fibroblasts and bacteria, being also able to precipitate glycoconjugates [73] The lectins, first identified in plants, are widely distributed in nature, including prokaryotic and eukaryotic organisms [74]. In addition to plants, lectins can be found in

animal venoms [75], bacteria [76], viruses [77] and fungi [78]. Plant lectins have been isolated from tissues such as seeds [38, 63], leaves [79], bark [80] and roots [81].

Seeds of *M. oleifera* constitute a rich source of bioactive proteins, including lectins with various biological activities. These lectins were purified through protein precipitation techniques followed by affinity or ion exchange chromatographies (Figure 3).

Santos et al. [82] reported for the first time the presence of lectin in seeds of this plant. The work demonstrated the presence of a water-soluble lectin (WSMoL) in preparations obtained through immersion of intact seeds in water after 5, 15 and 37 h. The preparations were particularly active with rabbit erythrocytes at pH 4.5 and showed affinity to fructose and porcine thyroglobulin. WSMoL was isolated by chromatography on chitin and the sequence (QAVQLTHQQQGQVGPQQVR) of a peptide (2,130 Da) obtained after in gel digestion of this protein showed significant similarity (70%) with M02.1 and M02.2, which are coagulant proteins from *M. oleifera* seeds [63].

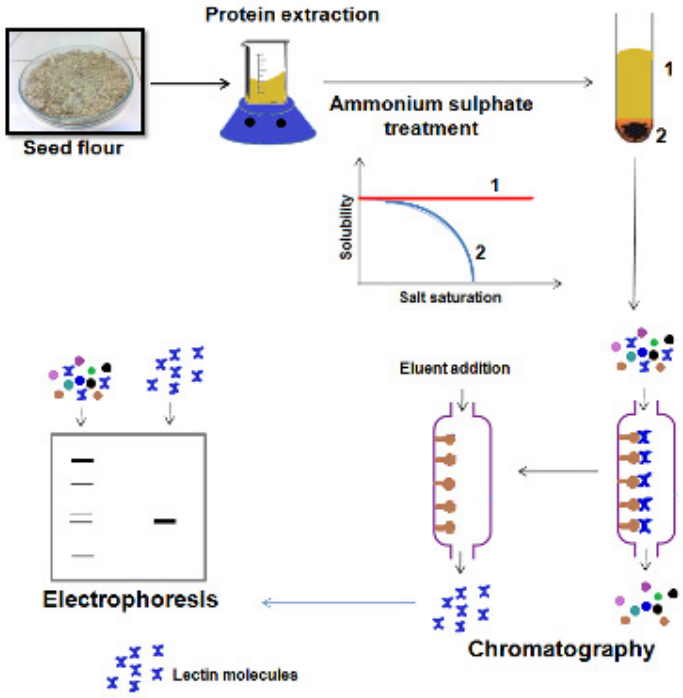

Fig. 3. A general scheme for purification of lectins from *M. oleifera* seeds.
The seed flour is used as starting material. The first step (protein extraction) corresponds to the homogenization of the seed flour with the extraction solution (distilled water, saline solution of buffers). Next, the proteins in the crude extract are precipitated by treatment with ammonium sulphate at different saturations. After centrifugation, this procedure results in separation of contaminants in the supernatant (1) and precipitated proteins (2) that had their solubility reduced by the increase of ammonium sulphate saturation. The protein fraction is then loaded onto affinity (chitin and guar gel for WSMoL and cMoL isolation, respectively) or ion exchange (for MoL isolation) chromatographies. The homogeneity of the lectins is investigated through polyacrylamide gel electrophoresis.

Katre et al. [68] reported the presence of another lectin from moringa seeds named MoL (*M. oleifera* Lectin); this protein is cationic with two subunits of 7.1 kDa in the presence of 2-mercaptoethanol; however, in the absence of 2-mercaptoethanol, two bands of 13.6 and 27.1 kDa appeared. The lectin was isolated by chromatography on DEAE-cellulose and CM-Sephadex and was inhibited by the glycoproteins thyroglobulin, fetuin and holotransferin indicating the complex sugar specificity of the lectin. The secondary structure elements of MoL are α-helix, 28%; β-sheet, 23%; turn 20%; and unordered structures, 28%.

Santos et al. [38] purified another lectin named cMoL (coagulant *M. oleifera* lectin) by affinity chromatography on guar gel. The cMoL is a cationic, heat-stable and pH resistant protein, with water coagulant activity and constituted by subunits of 26.5 and 14.9 kDa. Structural studies revealed that cMoL has 101 amino acids, 11.67 theoretical pI and 81% similarity with a *M. oleifera* flocculent protein. Secondary structure content revealed 46% α-helix, 12% β-sheets, 17% β-turns and 25% unordered structures. In addition, the tertiary structure of this protein belongs to the α/β class. cMoL significantly prolonged the time required for blood coagulation, affecting both the activated partial thromboplastin (aPTT) and prothrombin times (PT) [67].

WSMoL showed insecticidal activity against the dengue mosquito, *Aedes aegypti* [63]. WSMoL killed fourth-stage larvae of *A. aegypti* promoting morphological alterations in their digestive tract such as hypertrophy of segments and disruption of the epithelial layer [63]. Agra-Neto et al. [83] reported that the mechanism of larvicidal activity of WSMoL might involve deregulation of digestive processes due to a stimulatory effect on protease, trypsin, and α-amylase activities at larvae gut. WSMoL also showed stimulatory effect on oviposition by *A. aegypti* females and reduced the hatchability of the eggs by killing the embryos [64]. The same effect on oviposition was demonstrated when the lectin was tested in ovitraps at semifield conditions, revealing that the lectin has potential to be used for capture of eggs from *A. aegypti* [65].

cMoL also showed insecticidal activity, being active against the moth *Anagasta kuehniella* [66]. cMoL caused nutritional disorders and delayed the development of *A. kuehniella* larvae as well as reduced the weight and the survival of the pupae [66]. cMoL did not promote death of *A. aegypti* larvae but showed a remarkable inhibitory effect on superoxide dismutase of organophosphate-resistant larvae, which suggests future investigations on the use of this lectin as a synergist for larvicides [83].

Ferreira et al. [57] reported that WSMoL also has antibacterial and coagulant activities. Rolim et al. [61] investigated the genotoxic and mutagenic effects of the lectin and found that it was not mutagenic neither genotoxic by Kado/Ames and cell-free plasmid assays, respectively.

Seeds of *M. oleifera* were also reported to contain a family of chitin-binding proteins called MoCBPs. The $MoCBP_3$ is a 14 kDa protein that was able to inhibit the growth of the phytopathogenic fungi *Fusarium solani, Fusarium oxysporum, Colletotrichum musae* and *Colletotrichum gloesporioides* [72]. A study identified this protein as a member of the 2S albumin family, which is a class of seed storage proteins, and that fragments of its amino acid sequence aligned with stretches of the primary structure of cMoL (with homology between 75.7 and 94.2%); however the lectin cMoL possess a large amount of extra residues at the C-terminal end [84].

It has also been reported that the *M. oleifera* leaves and flowers contain protease inhibitors, which are proteins that inhibit the catalytic activity of enzymes able to cleave peptide bonds. A trypsin inhibitor was isolated from the leaves and showed a molecular mass of 23.6 kDa and a K_i value of 1.5 nM [69]. The inhibitor was more active against trypsin, but also showed high inhibitory activity toward serine proteases thrombin, elastase, chymotrypsin and the cysteine proteases cathepsin B and papain. The protease inhibitor also showed inhibited proteases from *Bacillus licheniformis* and *Aspergillus oryzae* proteases. It was also demonstrated that this inhibitor has potential to be used as food preservative because it prevented proteolysis in stored shrimps *Penaeus monodon*, which may reduce product deterioration [85].

Water extract from *M. oleifera* flowers was also source of a proteinaceous trypsin inhibitor (deemed MoFTI). This extract, which also contained triterpene, sterol and flavonoids, was able to promote mortality of *A. aegypti* larvae at second, third and fourth stages (LC_{50} of 1.72%, 1.67%, and 0.92%, respectively). Both the trypsin inhibitor activity and the larvicidal effect were abolished after heating (100 ºC, 12 h) of the flower extract; also, larvae exposed to the extract showed a progressive reduction of gut trypsin activity along the time. Together, the results suggested MoFTI as an active principle for the larvicidal activity of the extract [70].

In a further study, Pontual et al. [71] reported the isolation of MoFTI, which showed a molecular mass of 18.2 kDa and a K_i of 2.4 μM against bovine trypsin. The isolated inhibitor was able to kill *A. aegypti* newly hatched larvae and promoted arrest of larval development. In addition, the

authors showed that MoFTI exerted antimicrobial effect against the microbiota found at the gut of fourth-stage larvae (minimal inhibitory concentration of 0.031 mg/mL and minimal bactericidal concentration of 1.0 mg/mL). According to the authors, this is an interesting result because it is known that bacteria present in gut of *A. aegypti* larvae and adults are involved in their susceptibility to the dengue virus.

Finally, the proteases are another type of bioactive proteins found in *M. oleifera*. The flowers are source of proteases of interest to dairy industry. A flower protein preparation, obtained after treatment of a saline extract with ammonium sulphate, showed protease activity and was able to promote clotting of milk. The study revealed that flower proteases extensively cleaved the κ-casein but promoted low hydrolysis of the α_s- and β-caseins. The milk-clotting activity was Ca^{2+}-dependent and aspartic proteases are probably the main class of proteases involved in this activity [21].

6. *M. oleifera* AS A MEDICINAL PLANT

Traditionally, almost all tissues of *M. oleifera* are used in natural medicine to treat diseases such as abdominal tumors, hysteria, scurvy, attacks of paralysis, prostate and bladder diseases, wounds and skin infections [86]. In Thailand, moringa earned the name of “wonder tree” due to its potential therapeutic values to treat cancer, diabetes, rheumatoid arthritis and other diseases [87]. In India this plant is incorporated in various commercial herbal formulations, such as Rumalya and Septilin (The Himalaya Drug Company, Bangalore, India), and Orthoherb (Water Bush-nell Ltd., Mumbai, India), which are available for various disorders [88].

Extracts from moringa leaves are used for different medicinal purposes. Ghasi et al. [26] and Jaiswal et al. [88] studied that extracts of this tissue have hypocholesterolemic and hypoglycemic activities and Chumark et al. [87] found that this extract possesses antioxidant, hypolipidemic and antiatherosclerotic activities with therapeutic potential for the prevention of cardiovascular diseases. In addition, the aqueous leaf extract was able to inhibit the proliferation of human tumor cells (KB) in a dose-dependent manner as well as inducing cellular apoptosis [86]. Ouedraogo et al. [89] indicated that aqueous-ethanolic extract of *M. oleifera* leaves attenuates renal injury in rabbits treated with gentamicin, possibly by inhibiting lipid peroxidation. Jaiswal et al. [17] reported that aqueous extract of moringa young leaves showed antioxidant activity at in vivo and in vitro conditions; so, the regular intake of its leaves through diet could protect normal and diabetic patients against oxidative damage. Also, ethanolic extract of the leaves is used for hypertension [90] and promotes axodendritic maturation, as well as provides neuroprotection suggesting a promising pharmacological importance of moringa for the well-being of nervous system [91]. The hydroalcoholic extract of *M. oleifera* leaves showed therapeutic potential against vascular intimal damage and atherogenesis that lead to various types of cardiovascular complications. Therefore, this extract has being prescribed as food supplement for patients with coronary artery disease [92].

The oil from seeds is applied externally for skin diseases [93]. Guevara et al. [23] isolated from the ethanolic extract of *M. oleifera* seeds an antitumor promoter capable of inhibiting the progression of skin cancer in mice. Seed powder presented therapeutic efficacy against post arsenic exposure protecting animals from arsenic-induced oxidative stress and in depletion of arsenic concentration [94]. Aqueous seed extract showed anti-inflammatory activity in vivo in a model of carrageenan-induced pleurisy; the anti-inflammatory effect was linked to reduction of myeloperoxidase activity and nitric oxide, TNF-α and IL-1β levels [62].

Essential oil from moringa leaves and extract from seeds showed anti-fungal activities against dermatophytes such as *Trichophyton rubrum*, *Trichophyton mentagrophytes*, *Epidermophyton Xoccosum* and *Microsporum canis* and thus can be used in the future for development of anti-skin disease agents [95].

Aqueous and alcoholic extracts of *M. oleifera* root-wood reduced significantly the elevated urinary oxalate, showing a regulatory action on endogenous oxalate synthesis. Then, moringa root-wood showed antiurolithiatic activity [96]. Root extracts also had cytotoxic activity against leukemia cells (HL-60 and CEM) and melanoma [97].

Phenolic glycosides from *M. oleifera* fruits showed potent nitric oxide inhibitory activity. Nitric oxide is one of the inflammatory mediators causing inflammation in many organs [98]. Phytochemicals from *M. oleifera* pod husks, which are usually considered as agri-residues, exhibited antimicrobial potential against a variety of medically important pathogens. Therefore, this part of moringa has the potential for development of drugs of broad spectrum activity [99].

7. CONCLUSIONS

Environmental management as well as water and food supply are increasing challenges in the world. The protection of soil and water as vital natural resources as well as generation of biodiversity could certainly improve life conditions. *M. oleifera* has broad resource contributions due to its multiuse such as water purification, source of food nutrients and medicinal compounds. Thus, this plant is a powerful tool for humans in their constant task to guarantee adequate conditions for the life in our planet.

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