

## Review Article

# Biostimulatory Impacts of Seed Priming through Botanical Extracts on Crop Production-A Critical Review

## ABSTRACT

Rapid human population increase, food scarcity, and climate change are some of the global challenges in the current scenario. Among these, climate change adversely impacts crop production by creating abiotic stresses such as drought, salinity, and metal toxicity. Seed emergence, the critical stage in crop development, is highly vulnerable to these abiotic stresses. Therefore, short-term approaches to increase the sprouting of seeds, **seed germination** and the initial growth of plants were investigated in advance as a solution. Seed priming is **indeed** a pre-sowing treatment of seeds. **It is a technique used to enhance the germination and early growth of seeds by exposing them to controlled hydration and sometimes other treatments before planting.** Seed priming considered a low-cost, eco-friendly, and sustainable technique to promote seed germination and the initial growth of plants. Though various priming methods are available, this review discusses only botanical extracts (both seaweed and plant extracts) used as priming agents in seed priming. Among the seaweed extracts, green and brown algae species such as *Ascophyllum nodosum*, *Sargassum spp.*, and *Ulva spp.* enhanced the crop performances even under stress conditions. In addition, leaf extracts derived from **Drumstick** (*Moringa oleifera*) and **Neem** (*Azadirachta indica*) are widely used to increase the **crop production** productivity of vegetable and field crops under stress and non-stress environments. Furthermore, in this review we discussed about priming mechanism, different extraction methods and bioextracts behavior under diverse agroecosystems are discussed in detail.

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**Keywords:** Abiotic stresses, antioxidant, bioextracts, biostimulant, seed priming

## 1. INTRODUCTION

The challenge of a rising population affects the world. Due to population expansion, more individuals would require assistance with food security and supply. However, climate change, environmental pollution and abiotic stressors significantly impact on seed germination, emergence, seedling vigour, and crop productivity **production** [1]. These undesirable impacts on crop production and performance **of** significantly influence the food and agricultural systems. Several approaches, such as conventional breeding and advanced technologies (e.g., genetic engineering, mutation breeding, and polyploidy breeding) are being assessed to enhance crop production under environmental challenges. However, these techniques have significant drawbacks, including sizeable human resource requirements, biosafety apprehensions, and ethical concerns [1]. Thus, it is necessary to develop simple, efficient, and sustainable technology to solve these issues [2]. In this regard **regards**, seed priming is an alternative approach to getting around these constraints and increasing the capacity of plants to withstand stress [3,4].

Seed priming is a simple and an efficient method that would enhance **plants** **plant's** physiological and biochemical responses to abiotic stresses; thus, regulates hydration of seeds that permits pre-germination of metabolic activities without causing the radicle to emerge [5]. Basically, seed priming improves the seed germination or seedling growth rate through entrusting uniformity of seedling establishment [6]. Besides, priming process ensures early emergence, efficient water usage, mitigation of excessive fertilizer usage, promote roots growth, initiate the growth of reproductive organs, early flowering and maturity, resistance to abiotic stresses and soil-borne pathogens [2,6]. According to the way of conducting priming procedure, seed priming methods can be classified as hydro-priming, halo-priming/chemo-priming, osmo-priming, osmo-hardening, on-farm priming, matrix priming, nutri-priming, nano-priming, bio-priming, thermo-priming, UV priming and hormone priming [2,7]. Hydro-priming is achieved by soaking seeds in clean water and re-drying them to their initial moisture content before planting [8]. In this regard, hydro-priming shortens the lag phase of the germination process and facilitates rapid germination for vigorous development [9,10]. In osmo-priming, a low water potential osmotic solution is used to soak seeds rather than pure water [1].

Numerous materials are used in osmo-priming, including mannitol, sorbitol, glycerol, and inorganic salts [7].

Osmo-hardening includes re-drying of seeds after soaking them in tap water for 24 hours, then hardening them with  $\text{CaCl}_2$  Calcium chloride and  $\text{KCl}$  Potassium chloride solutions [2]. In solid matrix priming a solid or semi-solid media is employed instead of a liquid medium. This method involves combining seeds with a solid or semi-solid substrate with certain quantity of water [9]. Solid matrix priming would regulate water absorption by properly adjusting of moisture content [7]. In nutrient priming, seeds are exposed to a particular concentration of macro or micronutrients for a predetermined duration before planting. One of the novel seed priming techniques is nano-priming which uses nanoparticles (NPs), such as zinc oxide, iron oxide, titanium dioxide, silver nano-particles etc. [2]. In bio-priming, specific microorganisms or bioactive molecules are used for seed treatment [7]. Thermo-priming involves treating seeds at various temperatures while minimizing the influence on their critical physiological functions [7]. In hormonal priming plant growth regulators like auxins, gibberellins, kinetin, salicylic acid, abscisic acid, ethylene, and ascorbic acids are widely utilized [9]. Apart from these methods, in chemical priming seeds are treated with either organic (e.g., organic acids, botanical extracts, chitosan, polyamines, mannose, and trehalose) or inorganic (e.g., sodium nitroprusside, and sodium hypochlorite) substances [11]. As organic substances, botanical extracts are significant for seed priming. The main types of botanical extracts are being obtained from seaweed and plant extracts. Hence, in this review we would discuss the numerous botanical extracts, extraction methods, and their significance under abiotic stress conditions.

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## 2. SEAWEED EXTRACTS (SWEs) USED IN SEED PRIMING

Seaweed, often known as marine macroalgae, comprises a large portion of the world's marine life resources [12]. Most of their distribution recorded in shallow coastal waters, estuaries, and backwaters; but relatively small quantity has been found on solid surfaces like rocks, dead coral, and pebbles [12]. Approximately 10,000 species of macroalgae exist in globally which account for 10% of marine productivity [13]. According to presence of pigments, marine macroalgae would divide into three categories namely; (a) Chlorophyta or green algae, (b) Rhodophyta or red algae, and (c) Phaeophyta or brown algae respectively [14]. Brown algae mainly encompasses chlorophyll a and c with carotenoids and fucoxanthin while green algae contain pigments that are identical to the chlorophyll a, b and carotenoids which found in terrestrial plants [14]. Red algae contains both chlorophyll a, b, and phycoerythrin pigment which is responsible for the color of the red algae [15, 16].

Seaweed extracts (SWEs) have been used as a seed treatment to improve the seed germination growth and development [17]. These are widely recognized to include a number of bioactive compounds [18]. A variety of biologically active components including polyphenols (e.g., phenolic acids, flavonoids, cinnamic acid, isoflavones, benzoic acid, lignans, quercetin), polysaccharides (e.g., galactan, fucoidan, alginate, laminarin), proteins (e.g., lectins), polyamines, pigments, free amino acids, vitamins, micronutrients (e.g., B, Co, Cu, Fe, Mn, Mo, Ni, Si, Zn), macronutrients (e.g. Ca, K, Mg, Na, P, S), and natural phytohormones (e.g., cytokinins, auxins, gibberellins, abscisic acid), antioxidants, osmo-protectants betains, antimicrobial compounds are consist in SWEs [16, 18, 19]. Furthermore, seaweeds have elicitor chemical precursors that aid for seed germination [20]. Several Seaweed extracts (SWEs) used for seed priming are summarized in table 1.

**Table 1:**Seed priming through seaweed extract

No	Seaweed extract type	Crop type	Priming condition	Impact of seed priming	Country of study	Stress condition	Reference
1	<i>Ascophyllum nodosum</i>	Spinach	Seeds were soaked in solutions ranging from 0.15% to 1.2%	A concentration of 0.3% treatment showed the highest seed germination percentage and seedling	Brazil	Thermal stress	[21]
		Moth bean	Seeds were soaked with various concentrations (0%, 0.01%, 0.05%, 0.10%, 0.50% and 1.0%) for different periods ranging from 0 to 24 hours	Improved the seed germination, shoot and root length, fresh and dry weight, free radical scavenging, whereas alpha-glucosidase inhibitory action at low concentrations	India	Non-stress	[14]
		Onion	Seeds were soaked for 6 hours in different concentrations ranging from 0ppm to 7500ppm	Improved the growth of onion seedlings	India	Non-stress	[22]
		Pepper	Seeds were primed for 1 to 3 days in an incubator at 20°C using concentrations ranging from 1:1 to 1:1000 dilutions of seaweed extract	Increased total seed germination rate and decreased the mean germination time	Turkey	Non-stress	[23]

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	<i>Colpomeniasinuosa</i> <i>Sargassum vulgare</i>	Fenugreek	and water Seeds were soaked for 12 hours with various concentrations ranging from 5% to 25%	Enhanced the seed germination, biochemical constituents under low concentration	Egypt	Non-stress	[24]
	<i>Codium</i> <i>taylorii</i> <i>Pterocladia</i> <i>capillacea</i>	Radish	Sterilized seeds Were soaked for 2 hours in water	Enhanced radish water content and increased plant length significantly	Egypt	Salinity Stress	[25]
	<i>Codium tomentosum</i>	Wheat	Seeds were soaked in Different concentrations ranging from 10% to 50% at 12 hours	Increased shoot length, root length, fresh and dry weights of seedlings	Egypt	Non-stress	[26]
2	<i>Cystoseira</i> <i>compressa</i>	Cowpea/ Maize	Seeds were soaked in a concentration of 20 g L <sup>-1</sup> for 2 hours at 25 °C	Increased the germination rate of both crops, vigorous coleoptile elongation of maize, increased biomass weight and guaiacol peroxidase activity	Egypt	Salinity stress	[27]
3	<i>Ecklonia maxima</i>	Okra	Seeds were immersed for 24 hours in Kelpak® solution under 1:20, 1:40 and 1:100 v/v levels	Seed germination parameters were increased from 1:100 solution			[28]
		White foxglove	Seeds were treated with different biostimulants for 48 hours at 25 °C	Enhanced the seed germination and seedling growth	South Africa	Heat stress	[29]

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Comment [AR11]: South Africa

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	Maize	Soaked seeds in aqueous solution of seaweed extracts for 24 hours	Induced seed germination and shoot weight	Poland	Non-stress	[30]
4	<i>Ecklonia maxima</i> <i>Janiaadhaerens</i>	Tomato	Seeds were immersed in concentrations of 2.5, 5.0, and 10.0 mg mL <sup>-1</sup> overnight at room temperature	Extracts provide a sound protection from controlling of <i>Rhizoctonia solani</i> on tomato plants	Italy	Non-stress [31]
	<i>Gracilariatextorii</i> J. Agardh and <i>Hypneamusiformis</i> (Turner)	Brinjal, Chilly and Tomato	Seeds were soaked in the various ratios (1:2, 1:4, 1:6, 1:8, 1:10, 1:20,1:30) for 12 hours	Increased seed germination and crop yield	India	Non-stress [32]
5	<i>Gracilaria edulis</i>	Chilli	Seeds were primed for various time periods (1:25 for 48 hours, 1:5 for 24 hours and 1:5 for 72 hours)	Improved the vigor index and seedling weight	India	Non-stress [33]
6	<i>Janiarubens</i>	Maize	N/A	Germination rate, seedling length and weight, and number of lateral radicles were increased	Morocco	Non-stress [34]
7	<i>Kappaphycusalvarezii</i> <i>/Gracilaria edulis</i>	Rice	Seaweed extracts were added to rice seeds at concentrations of 2.5%, 5%, 7.5%, 10% and 15% for 24	Rice seed germination and seedling vigor index were increased at 2.5% and 5% concentrations	India	Non-stress [35]

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8	<i>Laurencia obtusa</i>	Cowpea/ Maize	hours Seeds were soaked for 2 hours at 25 °C in a concentration of 20 g L <sup>-1</sup>	Higher germination rate vigorous coleoptile elongation, higher biomass weight and guaiacol peroxidase activity were observed in both crops	Egypt	Salinity stress	[27]
	<i>Padina gymnospora</i>	Tomato	Seeds were treated with different concentrations (0.2, 0.4 and 1.0 %) for 24 hours	Enhanced germination, rate of germination, lower mean germination time, high germination index, greater seedling vigor, greater plumule and radicle length	Mexico	Non-stress	[36]
9	<i>Solieria spp.</i>	Kale	Seeds were soaked for 22 hours at 20°C	Seed primed with seaweed extract has not interfered with seed health	Brazil	Non-stress	[37]
	<i>Sargassum polycystum</i>	Red gram	Seeds were treated with different concentrations of water extracted and alcohol extracted seaweed (0.1, 0.25, 0.50, 0.75, 1.0 and 1.5%) for every 12 hours	The lowest concentration showed higher seed germination, shoot length, root length, fresh weight and dry weight	India	Non-stress	[38]
	<i>Sargassum tenerrimum</i>	Tomato	Seeds were immersed in a variety seaweed aqueous extract concentration,	Seed germination rate was high at 0.8%	India	Non-stress	[39]

**Comment [AR21]:** Which Concentration

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		including 0.2%, 0.4%, 0.6%, 0.8%, and 1.0%				
<i>Sargassum vulgare</i>	Wheat	Seeds were soaked in different concentrations ranging from 10% to 50% for 12 hours	Increased seed germination, growth and yield	Egypt	Non-stress	[26]
	Tomato	Seeds were treated with 0.2% and 0.5% concentration of seaweed extract and then exposed for 14 days photoperiod	Increased seed germination percentage and the growth of the radicle	Morocco	Salt stress	[40]
	Wheat	Seeds were treated at different concentrations (0.2, 0.5, 25 and 50 %) and kept under photoperiod for 14 days	Enhance the germination rate with lower mean germination time, greater seedling vigor and greater radicle length	Tunisia	Salt stress	[41]
<i>Sargassum vulgare</i> , <i>Colpomeniasinuosa</i> , <i>Pandiapavonica</i>	Pea	Sterilized seeds were soaked in seaweed extracts or distilled water for two hours	Increased the production of photosynthetic pigments, new peptides, and improved germination, seedling length, and fresh weight	Mention country name	Salt stress	[42]
<i>Sargassum wightii</i> Grev.	Green gram	Seeds were soaked with concentrations of 0.5%,	In comparison to the high concentration (2.0%), 6 hours of	Mention country	Non-stress	[43]

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		1.0%, and 2.0%, for 6, 12 and 24 hours	soaking in the 0.5% concentration led to higher seed germination and earlier growth	name				Comment [AR28]: ???
								Comment [AR27]: Which concentrations
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<i>Sargassum wightii</i>	Wheat	Seeds were soaked with different concentrations ranging from 5% to 50% for 12 hours	Increased the germination rate, number of lateral roots, shoot length, number of kernels per plant, and kernal length	India	Non-stress	[44]		Comment [AR31]: Ranged
<i>Thalassiahemprichii</i>	Green gram	Seeds were treated with 50 ppm and 100 ppm of seagrass extract	In comparison to the control treatment, the root: shoot ratio has risen for all of the treatments	India	Non-stress	[45]		
<i>Ulva</i> , Polysiphonia, Cladophora	Garden cress	Seeds were treated with concentration of 0.5, 2.5 and 10%	Chlorophyll content was higher compared to non-treated sample	Mention country name	Non-stress	[19]		Comment [AR33]: Seed germination
<i>Ulva fasciata</i>	Bell pepper	for 24 hours, seeds were soaked in seaweed extract at various concentrations (2, 4, 6, 8 and 10%).	The phytochemical content and germination were enhanced at 8% concentration	India	Non-stress	[46]		Comment [AR32]: Which concentration
<i>Padina gymnospora</i>								Comment [AR34]: ???
<i>Gracilaria edulis</i>								Comment [AR35]: Which various concentration
12 <i>Ulva fasciata</i> , <i>Padina gymnospora</i> , <i>Gracilaria edulis</i>	Chilli	Seeds were treated in various concentrations range from 2% to 10% for 24 hours	Phytochemical content and germination were enhanced by <i>Padina gymnospora</i> at 8% concentration level	India	Non-stress	[15]		

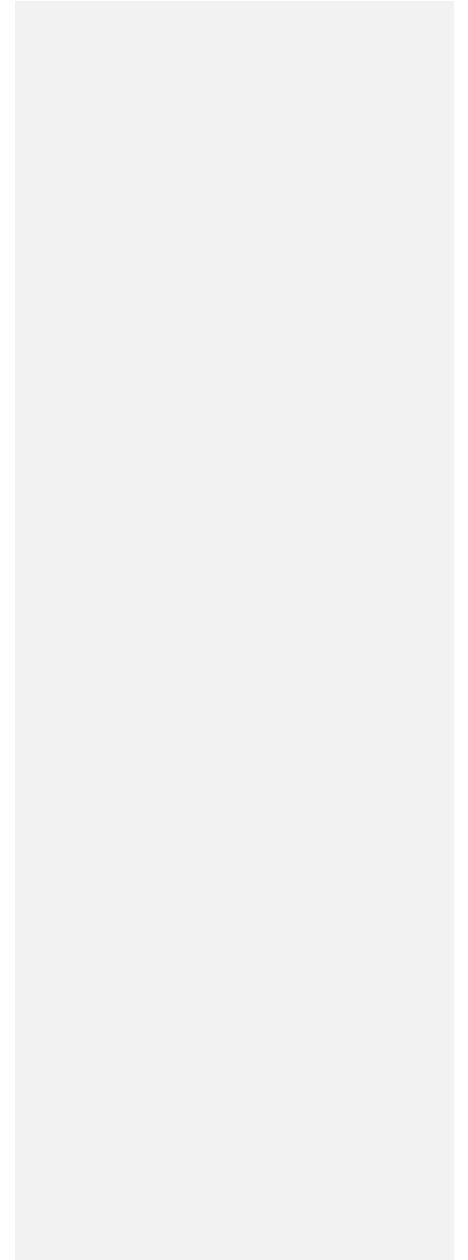
13 *Ulva lactuca*

Onions	Solid matrix priming using a 2:1:3 mixture of seed, vermiculite, and seaweed extract were carried out at 15 °C for 2 days, seaweed extract was applied at 5% concentration	Enhanced the seed germination and seedling quality	Mention country name	Drought and salinity stresses	[47]
Carrot	N/A	Enhanced the germination fresh and dry weight of seedling	Turkey	Salinity stress	[48]
Tomato	Seeds were soaked for 24 hours at 25°C in a 1 mg mL <sup>-1</sup> solution	Increased the tomato fresh weight, glycine betaine, total phenols and soluble sugars concentration, significantly decreased the hydrogen peroxide (H <sub>2</sub> O <sub>2</sub> ) concentration	Morocco	Salinity stress	[49]
Cowpea and Maize	Seeds were soaked for 2 hours at 25 °C in a concentration of 20 g L <sup>-1</sup>	Observed higher germination rate of both crops, vigorous coleoptile elongation of maize, increased biomass weight and guaiacol peroxidase activity	Egypt	Salinity stress	[27]
Tomato	Soaked for 24 hours	Enhanced germination, rate of germination, lower mean germination time, high germination	Mexico	Non-stress	[36]

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			index, greater seedling vigor, greater plumule, radicle length			
	<i>Ulva lactuca</i> ,	Green	Seeds were soaked for 12	The 0.3% concentration has the	India	Non-stress [50]
	<i>Caulerpa scalpelliformis</i>	gram	hours with concentrations	lowest germination percentage and		
	<i>Sargassum plagiophyllum</i>	( <del>Vigna</del> <del>radiata</del> )	ranging from 0.1% to 0.5%	maximum germination (100%) has		
	<i>Turbinaria conoides</i>		seaweed extracts	0.4% <i>Caulerpa scalpelliformis</i> and		
	<i>Padina Tetrastrumatica</i>			0.1% <i>Padina tetrastrumatica</i>		
	<i>Dictyota dichotama</i>			extractions		
14	<i>Ulva linza</i>	Wheat	Seeds were soaked for 12	Increased seedling growth at low	Egypt	Non-stress [51]
	<i>Corallina officinalis</i>		hours in aqueous extract at	concentration		
			different concentrations			
			range from 5% to 30%			

UNDER PEER REVIEW



The commercial **Seaweed extracts** **SWEs** sector employs various exclusive extraction methods to rupture the cells and release beneficial components into the extract. These extraction techniques are further classified as enzyme-assisted extraction, microwave-assisted extraction, pressurized liquid extraction, supercritical fluid extraction, ultrasound-assisted extraction, soxhlet extraction, alkali extraction, acid extraction, and cell bust extraction [13, 18, 19]. In this regard, a variety of solvents, including ethanol, acetone, methanol-toluene, methanol, petroleum ether, ethyl acetate, dichloromethane, and butanol can be utilized for an effective extraction process. However, these procedures are sometimes considered costly and hazardous [19]. Hence, boiling and soaking extraction techniques with distilled water are used to overcome conventional extraction techniques' drawbacks; which are eco-friendly and require no organic solvents [19].

### 3. PLANT EXTRACTS (PEs) USE IN SEED PRIMING

Another effective method to promote germination is soak the seeds in PEs before planting [52]. Numerous chemical compounds identified in various plant species that improve the growth, development, and productivity of crops grown under adverse conditions. PEs modify the physiological and biochemical processes such as stomatal conductance, phytohormone metabolism, absorption of water and nutrients, antioxidant defensive systems, and photosynthesis [53]. Different PEs used for seed priming are summarized in table 2.

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Comment [AR38]: Write full name

Comment [AR39]: Write full name

UNDER PEER REVIEW

**Table 2:** Seed priming through plant extracts

No	Plant extract type	Crop type	Priming condition	Impact of seed priming	Stress Condition	Country of study	Reference
1	Moringa leaf extract (MLE)	Maize	Seeds were soaked (0.25, 0.5 and 1.00 g) at the ratio of 1:10 concentrate for 12 to 20 hours	Improved the germination and seedling growth	Non-stress	Egypt	[52]
		Rice	Seeds were soaked in an aerated solution of MLE at 3% for 8 hours	Improved the speed and spread of emergence of seedlings (e.g., Time to start emergence, emergence index, mean emergence time, final emergence percentage)	Drought stress	Pakistan	[53]
			Seeds were soaked in the MLE (seed weight: MLE, 1:5) at room temperature in dark conditions under continuous shaking, for 24 hours	Improved seed germination and the plant growth	Arsenic stress	India	[58]
		Onion	Seeds were soaked in 12.5% and 25% of MLE for 6 and 12 hours respectively	Seeds soaked in MLE for 6 hours gave higher seed emergence	Non-stress	Nigeria	[60]
		Wheat	Seeds were soaked in MLE 1:10 concentration for 12 hours	Improved seed germination and seedling growth	Salinity stress	Libya	[61]

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Raddish	Seeds soaked in 3 and 5% for 6 and 9 hours under continuous aeration	Increase growth with improved biochemical and antioxidant attributes	Non-stress	Pakistan	[62]
Cucumber	Aerated fresh moringa leaf extract for 18 hours	Improved germination and seedling growth	Non-stress	Nigeria	[63]
Pea seeds	Seeds were primed with 3% moringa leaf extract and in combination of magnetized water and moringa leaf extract for 12 hours	Improved germination capacity and seedling vigor	Non-stress	Pakistan	[64]
Cowpea	Primed with 5% and 2% moringa leaf extract for 12 hours	Increase germination percentage with better root and shoot length	Non-stress	Pakistan	[65]
Linola	Primed with 3.3% moringa leaf extract for 12 hours	Improve crop growth and yield	Non-stress	Pakistan	[66]
<i>Sesamum indicum</i> L.	Primed with 2, 4 and 6% solutions of fresh moringa leaf extract for 6 hours	Improve the seedling emergence and growth	Non-stress	Pakistan	[67]
Spring maize	Priming 3% moringa leaf extract for 18 hours	Increased root and shoot lengths, the ratio of root:shoot, length and maximum seedling fresh and dry weights	Under cool condition	Pakistan	[68]
Hybrid maize	Primed with 1:30 and 1:40 moringa leaf extract for 18 hours	Higher emergence rate and better early seedling	Non-stress	Pakistan	[69]

**Comment [AR45]:** What is 3.3% ?

			growth				
2	Neem leaf extract (NSE)	Bread Wheat	Seeds were soaked for 12 hours	Enhanced plant growth and reduced nematode infestation	Non-stress	Pakistan	[57]
		Rice	Seeds were primed with NSE concentrations 25%, 50% and 100% for 24, 48, and 72 hours respectively	Increased the seed germination	Non-stress	Sri Lanka	[70]
		Chickpea	Seeds were primed in 5% and 10% NSEs for 10 hours	10% NSE significantly affected for the emergence percentage, primary branches per plant, pods/plant, seeds per pod, and seed yield per plant	Non-stress	India	[59]
		Lentil	Seeds were primed in 50% NSE solution for overnight	Observed higher germination percentage, number of branches per plant, seeds per pod, 1000 seeds weight, and yield	Non-stress	India	[55]
3	Garlic extract	Brinjal	Seeds were primed in 100, 200, and 300 $\mu\text{g ml}^{-1}$ for 4, 8, and 12 hours respectively	Enhanced the seed germination and initial growth	Non-stress	China	[54]
4	Sorghum water extract	Camelina	Seeds primed in 5% solution for 24 hours at 25°C	Increased the emergence percentage and ionic homeostasis. Enhanced seedling growth, biomass	Salinity stress	Pakistan	[56]

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**Comment [AR47]:** Mention in which concentration maximum seed germination

**Comment [AR48]:** Just 50%

				production, and chlorophyll content			
		Sunflower/ Maize	Seeds were primed with 1%, 1.5%, 2%, 2.5% and 3% concentrations for 8 and 14 hours, respectively	Primed seeds were showed higher seedling development	Non-stress	Pakistan	[71]
6	Arappu extract	Okra	Seeds were kept in 3% solution for 12 hours	Increased the germination and seedling vigor	Non-stress	India	[72]
7	Sugar beet extract	Wheat	Seeds were primed at different concentrations (10%-50%) for 12 hours	Increased biomass, photosynthetic pigments, and antioxidant defense mechanism	Drought stress	Pakistan	[73]
	<i>Eclipta alba</i>	Sorghum	Seeds soaked in the extract for 6 hours	Increase of yield, emergence and suppression of pathogenic fungi	Non-stress	Denmark	[74]
	Prosopis ( <i>Prosopis juliflora</i> )	Black gram	Seeds were soaked in leaf extracts at 1% solution for four hours	Enhance the seed quality characters	Non-stress	India	[75]
		Pigeon pea	Priming with 2% leaf extract for one hour soaking	Enhanced the seed and seedling quality characters	Non-stress	India	[76]
	Marigold	Pepper ( <i>Capsicum baccatum</i> var. <i>pendulum</i> )	18 ml of priming solution at 25 °C for 24 hours in the dark	Improved the emergence, and in the repair of undeveloped seeds	Non-stress	Turkey	[77]

**Comment [AR49]:** which concentration is record maximum results

**Comment [AR50]:** not related with

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		<i>Willd.</i> )				
White musale, Periwinkle, Neem, Wood apple, Lantana and White cedar	Tomato	Seeds soaked at room temperature 2% and 4% solution four hours	Improvement in seed and seedling growth parameters in presence of pathogen	Non-stress	India	[78]
<i>Acacia nilotica</i> (L.) and <i>Sapindus mukorossi</i> (L.)	Peanut, chickpea, sunflower and okra	Seeds primed with for 5, 10, 20 and 40 minutes	Control of root rot fungi and growth and productivity	Non-stress	Pakistan	[79]
<i>Cyperus esculentus</i> (Della) <i>Axonopus compressus</i> (Itsit), <i>Convolvulus arvensis</i> (Lehli) and <i>Parthenium</i>	Rice	Seeds soaked in the weed extract for 30 minutes	increased germination rate and germination percentage	Non-stress	Pakistan	[80]

*hysterop  
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	Aloe vera, <i>Moringa olifera</i> or sugar beet aqueous extracts	Lentil ( <i>Lens culinaris</i> Medik)	Seeds soaked in aerated for 14 h using five concentrations (1%, 2%, 3%, 4% and 5%)	Improved seedling growth	Chilling stress	Pakistan	[81]
	pungam ( <i>Pongamia pinnata</i> ) and arappu ( <i>Albizia amara</i> )	Cluster bean	SeedSoaked in 1and 2% for 3 hours and 6 hours	Enhanced germination, vigour and field emergence	Non-stress	India	[82]
9	<i>Cyperus rotundus</i> L.	Drumstick	Seeds were soaked in four aqueous extracts of <i>Cyperus rotundus</i> concentrations (0%, 25%, 50% and 100%)	Seed priming with 50% concentration resulted in an increase in initial shoots length and photosynthetic pigments accumulation	Non-stress	Brazil	[83]
10	Tomato leaf	Sugar beet	Seeds were primed for 2 days at 25 °C	Increased seed germination	Non-stress	Turkey	[84]

Comment [AR52]: what mean field emergence

	extract						
11	Carrot leaf extract	Pea	Seeds were primed 75 ml L <sup>-1</sup> concentration for 10 hours	Observed higher plant height	Drought stress	Egypt	[85]
12	Chicory, clerodendron, noni and calotropis leaf extract	Maize	Seeds were soaked in 5%, 10% and 15% chicory extract and 1, 2 and 3 % of clerodendron, noni and calotropis for 12 hours	Increased the seedling vigor and yield parameters	Non-stress	India	[86]

**Comment [AR53]:** what about germination results

According to the literature, several PEs are widely used in crop cultivation such as *Allium sativum* (Garlic), *Zingiber officinale* (Ginger), *Azadirachta indica* (Neem), *Codiaeum variegatum*, (Crouton), *Moringa oleifera* (Moringa/Drumstick), *Datura stramonium* (Jimsonweed), *Aloe vera* (Aloe), *Ricinus communis* (Castor), *Allium cepa* (Onion), *Helianthus annuus* (Sunflower), *Sorghum bicolor* (Sorghum) etc. [54,55,56,57,58]. PEs are rich in macro and micronutrients, antioxidants, amino acids (e.g., alanine, glycine, leucine and proline), ascorbate, zeatin, growth hormones, vitamins (e.g., B-complex, C,  $\beta$ -carotene,  $\alpha$ -tocopherol), allelochemicals, enzymes (e.g., amylase, catalase, lipase, oxidase, superoxide dismutase) and other organic compounds (e.g., triglycerides, triterpenoid, gibberellin, potassium sorbate and salicylic acid) [52,53,59]. However, PEs would have a variety of compositions depending on the plant species.

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PEs are commonly prepared as fresh or dry basis [52, 53]. For instance, Khan et al. [53] prepared the plant extract using fresh mature leaves. According to the described method, the extract was extracted from overnight frozen leaves using locally built equipment, and predetermined concentration was prepared by mixing it with distilled water. Ahmed and El-mahdy [52] described the procedure to prepare the extract by air-drying fresh moringa leaves and to store the dried powders at room temperature. The powdered moringa leaves were immersed in distilled water on a weight/volume basis for 24 hours with periodic shaking. The liquid was filtered through four cheesecloth layers and Whatman filter paper to remove fibre debris. Moreover, in some experiments centrifuging process was also carried out as an additional step [57].

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#### 4. MECHANISMS OF SEED PRIMING

There are three main phases in the seed priming procedure namely; seed imbibition (phase I), activation phase (phase II) and growth and cell elongation phase (phase III). In phase I, water uptake occurs according to the water potential gradient [11]. In activation phase, most metabolic and repairing actions occur in the cytosol (e.g., protein synthesis, DNA repairing and stimulation of enzymes and antioxidants). During this phase, water imbibition is also continued and numerous cellular activities such as production of ATP, essential lipids and antioxidants take place [87]. Among these activities, DNA repairing is an essential process which protects the cell from oxidative injuries during seed germination [88]. According to latest literature, germination-related enzymes are activated at a higher rate in this phase. For instance,  $\alpha$ -amylase activity amplified in wheat seeds after the seed was primed with sorghum water extract [88]. Moreover, seed priming boosted protein synthesis by improving rRNA synthesis and ribosome integrity [11]. Simultaneously, antioxidant enzymes such as peroxidase (POD), superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione reductase (GR) and catalase (CAT) ensure cellular protection from reactive oxygen species (ROS) [8]. Stage III consists of rapid water uptake, and the radicle's protrusion indicates that the germination process has advanced to the development and cell elongation phases [87]. Figure 1 illustrates the seed priming mechanism.

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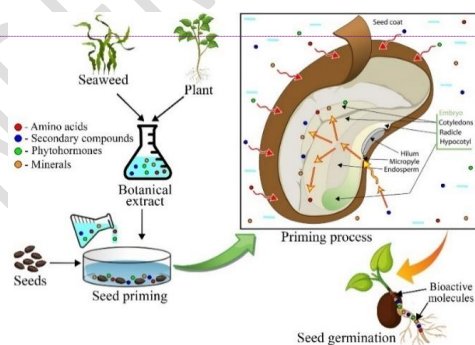


Figure 1: Seed priming mechanism

#### 5. SEED PRIMING THROUGH BOTANICAL EXTRACTS UNDER NON-STRESS CONDITION

Seed germination is the most crucial step in crop cultivation which is considered as an internal process aided by a series of enzymatic reactions, mainly governed by  $\alpha$ -amylase [28]. Furthermore,  $\alpha$ -amylase

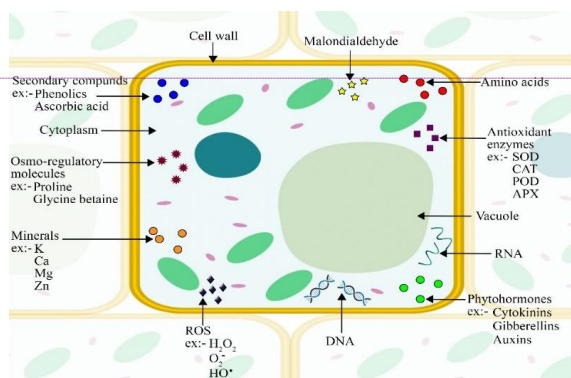
gelatinizes the starch in the endosperm, thus supplying ATP energy for the growing embryo [88]. However, presently, commercial crop growers might face numerous consequences related to poor performances in the seed germination process due to non-synchronized germination, an uneven growth pattern, and insufficient seedling emergence [28]. Therefore, to mitigate these constraints, seed priming with numerous composites has been introduced as a viable approach [88].

As a biostimulant, SWEs can be incorporated into the seed coat through priming, stimulating the seed germination rate and initial growth [28]. SWEs promote seed germination mainly due to the presence of various compounds in the solution. For instance, at different concentrations, SWEs containing various phytohormones would result the germination and initial seedling development process [16]. However, Nonogaki et al. [89] further reported that auxins had not affected seed germination directly, even though they enabled gibberellic acid biosynthesis. Thus, they promoted seed sprouting through activating of  $\alpha$ -amylase [89]. Concurrently, primed seed germination enhancement might be impacted by solution-retention effects in the pre-germination phase [20]. Furthermore, some mineral nutrients in the seaweed extract might positively affect the plants seed germination rate and vigor index [33]. Seed priming through seaweed extraction directly impacts the alteration of seed biochemical properties which accelerates seed germination [33]. According to recent findings, the cell membrane permeability has been reduced in seaweed extract-primed chilli seeds. In contrast, electric conductivity is also reduced in the cytosol, resulting from optimum cell functions avoiding damage or solute leakage from membranes [33]. Furthermore, numerous macro and microelements in the SWEs, such as N, P, K, Ca, Mg and Zn, promote initial plant growth and development [25]. These elements act as building blocks of proteins and other essential substances in the cytoplasm, which play a critical role in osmotic adjustment [25]. Similarly,  $Ca^{2+}$  and  $K^+$  ions included in SWEs catalyze some enzymatic reactions related to seed germination and growth, thus, ultimately enhancing the final yield [36]. The impacts of SWEs on seed priming have been investigated through several experiments (table 2). In addition, SWE-primed seeds contained a higher concentration of antioxidants; thus, enhancing the metabolic activities and mitigating ROS production would affect seed growth and development [33].

Similar like to SWEs, plant extracts also contain numerous bioactive compounds. [52, 56]. However, the plant type and area where these plants are being grown altered the bioactive compounds' concentration and composition [53]. Seed priming through leaf extractions enhanced plant growth under normal and stress conditions [53]. For example, moringa leaf extraction (MLE) consisted of a mixture of mineral nutrients including K, Ca, Mg, vitamin C, and plant growth regulators (e.g., cytokinin), which expedited the cell division, delayed leaf senescence, expanded the leaf area and increased chlorophyll pigments [56]. Furthermore, plants' biochemical attributes would increase after seed priming through MLE due to various allelochemicals, including secondary composites such as phenolic and ascorbic acid [53]. Simultaneously, with the enhancement of chlorophyll content, it has been observed that mineral nutrients in MLE accumulate in the seed embryo during the seed priming process and facilitate the growth and development of plants [28, 53].

## 6. SEED PRIMING WITH BIOEXTRACTS UNDER STRESS CONDITIONS

As a result of extreme environmental factors such as drought, salinity, temperature and trace metal (loid) s physicochemical reactions in the plant cells are highly impacted [90]. For instance, Siddiqui al. [90] reported that salinity stress enhanced leaf chlorosis by accelerating chlorophyllase activity which depreciates pigment proteins or enhances the enzymatic chlorophyll degradation process. When plants are exposed to stress conditions, they would stimulate to react to the stress by producing reactive oxygen species (ROS) such as singlet oxygen molecules, hydrogen peroxide ( $H_2O_2$ ), hydroxyl radicals (OH), superoxide anion ( $O_2^-$ );[33,42; Figure 2).



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**Figure 2: Different cellular substances present in the cytosol after seed priming under stress condition**

These ROS are involved in the degradation of the thylakoid membrane and hinder the chlorophyll synthesis process [90]. Furthermore, Szafrńska et al. [91] observed poor expression of several Mg-chelatase subunit-encoding genes, resulting in low chlorophyll synthesis. In addition, under the salinity stress, water absorption, stomatal conductance nutrient translocation and assimilation [56] were hindered, and this leads to the malfunctioning of cellular activities. In addition, higher salinity level has altered the plant's morphological characteristics by reducing leaf size, developing thicker leaves, and dwarf plants [56]. ROS substances down-regulated gibberellin genes, thus, resulting from low gibberellic acid production and reduction of  $\alpha$ -amylase activity [91]. Overall, seed germination and initial plant growth would retard drastically. Apart from ROS, Malondialdehyde (MDA) synthesis is also induced due to lipid peroxidation, and higher accumulation of MDA in plant cells eventually leads to malfunctioning cell functions [92].

However, to combat with ROS compounds plants produce other osmo-regulatory molecules such as proline and glycine betaine [42]. Since proline is an essential amino acid, it makes a defense against oxidative damage that occurs from ROS [93]. Hence, proline accelerates protein hydrolysis and ensures osmotic balance [42]. Further, glycine betaine production in the cytosol also increased and mitigated the most adverse effects of salinity stress [93]. For example, glycine betaine protects cell membranes and proteins, triggers coenzyme activation, and removes excess ROS compounds from the cell [93]. Interestingly, with elevated saline levels, plants would enhance carotenoid production [53]. Hence, salinity stress triggers overexpression of genes relevant to carotenoid accumulation under stress conditions, directly involved in reducing free radicals in the cytosol [92].

Seaweed and plant extracts contain unique bioactive compounds (e.g., growth regulators, micronutrients, osmo-protectants, and amino acids) that are essential to accelerate defense mechanisms under abiotic stress environments [56]. The protection mainly occurs due to numerous minerals, amino acids, proline and glutathione induce antioxidant enzyme production; thus, reducing ROS levels in stressed plants [93]. Moreover, the exogenous application of SWEs and PEs further increases the antioxidant concentration in cells, which mitigates ROS production [53]. In this regard, the most common antioxidant enzymes, namely, POD, SOD, APX, GR and CAT concentration, have increased after the priming, which protects the plants from oxidative stress by scavenging ROS [8]. In addition, previous research details provide evidence that even under abiotic stress circumstances, seed priming with SWEs would mitigate the harmful effects of those adverse conditions [29]. For instance, seed priming with MLE boosted the initial maize growth due to the presence of phenols and ascorbates [69]. Moreover, Tahira et al. [94] further noticed that MDA production had been reduced after the seed priming.

Abiotic stress conditions widely affect the protein synthesis process in plants. For example, salinity stress-induced *Vigna mungo* varieties showed different protein profiles, known as "new stress-proteins" which are highly important to manipulate osmotic potential [93]. The elevated ROS production in cell resulted DNA alternations and produced DNA fragments, resulting in genome instability [95]. The presence of bioactive substances in SWEs would upregulate the polypeptide synthesis by activating specific enzymes [42]. In addition, salinity stress might involve in occurring of mutations, chromosomal rearrangements, and alternations in DNA base arrangement that leads to genotoxic impairment and structural modifications [95]. Hence, seed priming through SWEs ensures DNA, protein, and RNA repair and synthesis throughout germination phase II [42].

## **7. CONCLUSION AND FUTURE PERSPECTIVES**

Climate change and other numerous challenging factors negatively influence on the agricultural production. The sustainability of crop production in a changing environment depends on improved seed quality through using environmentally friendly method like seed priming with botanical extracts. Hence, without endangering the environment or human health, it may be possible to promote crop growth and development, boosting of yield and protecting against numerous abiotic stressors. In addition, crop cultivation under stress and non-stressed conditions, seed priming would become more effective tool.

Although seed priming with bio-extracts would have given positive results, further researches are needed to understand the physiological and biochemical impact of bioextracts at seed germination and seedling growth stages. In this regard, future experiments should target on molecular mechanisms

engage in seed priming process. Furthermore, studies on undiscovered mechanisms of bioextracts on growth promoting processes would be essential to increase usage of these extracts in crop cultivation. Future researches on direct role of botanical extracts in mobilizing seed reserves and the partitioning of biomass in seedlings is also required. Moreover, SWEs differ from one another even processing the same raw material using different extraction methods. Hence, further crop-specific experiments are necessary to maximize the application of botanical extracts to achieve optimum results. The intensity of this impact varies depending on the species and crop/plant treated. However, the bioextract source varies according to these several factors such as maturity, leaves or shoots contained, location etc. Thus, these factors should be more considered in future experiments. Comprehensive studies should be needed to investigate the biological actions of botanical extracts during seed germination, including the control of ROS, their scavenging by antioxidant enzymes. Hence, more studies are required in future to assess the effectiveness of botanical extracts in minimizing the adverse effects of soil salinization on plants, as well as the ideal dose of application.

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