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RESEARCH ARTICLE

A new dawn for coriander seeds: Overcoming dormancy through innovative and comprehensive enhancement techniques

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Abstract

Coriandrum sativum an important spice exhibits lower seed germination. That may be due to phenolic compounds that induce dormancy in seeds. This study aimed to develop a comprehensive seed enhancement technique to overcome dormancy and improve germination and seedling growth. Seeds were subjected to GC-MS analysis to identify bioactive components. Seeds were leached for 6 to 30 h (Leaching). Following leaching, hydropriming for 3 to 18 h to standardize the priming duration. At the standardized priming duration, seeds were primed with hydrogen peroxide, nitric oxide, salicylic acid and 2,4-D (leaching + priming). Additionally, leached seeds were coated with polymer and plant growth-promoting microorganisms such as *Azospirillum*, *Trichoderma viride* and *Bacillus subtilis* (leaching + coating). Finally, the best-performing treatments from these experiments were compared with comprehensive technique. The results revealed that coumarin was the primary inhibitor. Leaching (24 h) significantly improved germination by 25.8 % over control. The duration of priming was standardized to 6 h after leaching. Leaching (24 h) + priming with 25 mM H_2O_2 (6 h) significantly increases germination percentage by 30.7 % over control. Leaching (24 h) + coating with polymer and *Azospirillum* at 100 g/kg of seeds significantly increased germination by 32.8 % over control. Assessment of the comprehensive techniques revealed that seed leaching (24 h) + priming with 25 mM H_2O_2 (6 h) + coating with polymer and *Azospirillum* at 100 g/kg significantly increased the germination percentage by 39 % over control. Results show that a comprehensive technique breaks dormancy and improves germination and seedling growth.

Keywords

dormancy; coumarin; seed leaching; seed priming; seed coating; *Coriandrum sativum*

Introduction

Coriander (*Coriandrum sativum* L.) belongs to the family Apiaceae and grows worldwide. It is widely cultivated for its nutritional benefits, biological properties and culinary purposes (1). The leading countries in

coriander production were Canada, China, Bulgaria, India, Morocco, Syria, Egypt and Romania. Among these, India contributes to 80 % of total coriander production in the world (2). In the Indian subcontinent, coriander leaves were used as a flavoring agent in curries, soups, boiled items and added in the final stage of cooking (3). It has multiple medicinal benefits, such as anti-atherogenic, hypolipidemic, antiarrhythmic and antihypertensive effects and helps in cardio protection (4).

Coriander seed is a dry schizocarp and ovate globular in shape with 2 mericarps (5). The schizocarp does not split into the 2 mericarps naturally but these can be achieved by manual splitting with fingers. Moreover, in some genotypes fruits split on maturity spontaneously (6). The Indian Minimum Seed Certification Standards for the germination percentage of coriander seeds were 65 %, as it has a low seed germination rate (7). Coriander seeds show lesser germination percentages even under favourable conditions. There by it exhibits poor efficiency in production and yield as it has low and inconsistent rate of seed germination (8). The seed quality has a reflective influence on the seedling emergence and productivity of agricultural crops economically for all species (9).

Seed germination is a physiological process that begins with the water uptake by the quiescent seeds and starts with the protrusion of the radicle from the covering layers of the seed. On the other hand, seed dormancy is an adaptive trait that blocks seed germination even under favourable conditions (10). Chemical inhibitors such as phenols present in the pericarp and seed coat also inhibit seed germination (11).

Seed leaching is a technique that involves washing off the inhibitors present within the seed or seed coat by soaking it in water or keeping it under running water. In cases of prolonged soaking, the water must be changed frequently to restrict the fermentation or decay of seeds (11).

Seed priming process involves the hydration followed by dehydration of seeds prior to germination of seeds in order to improve the metabolic process of seeds. This technique speed up the germination process, growth of seedlings and yield under normal and stressful conditions (12). Several priming agents, such as hydrogen peroxides (H_2O_2), salicylic acids (SA), nitric oxides (SNP), gibberellic acids (GA_3), water (hydropriming), etc., can be used to increase seed germination (13-15).

Seed coating is the modification of the physical properties of seeds by exogenously applying certain physical, chemical or biological compounds to the natural surface of the seed coat. Seed coating improves germination, fastens phenological events, enhances physiological and morphological attributes and ultimately improves yield (16).

Considerable levels of dormancy due to the presence of inhibitor are the major problem encountered in coriander seed testing. So, there is a necessity to overcome the seed dormancy and enhance the seed performance. It has been hypothesized that developing a comprehensive seed management technique that encompasses removal of inhibitors and techniques that enhances the seed quality.

The objectives of this study were to (i) standardize the leaching duration for removal of inhibitors from the seed. (ii) identifying the effect of different seed enhancement technique such as priming and coating (iii) development of comprehensive seed management technique by combining all the techniques for improved germination, seedling growth, vigour and biochemical activities

Materials and Methods

Study area

Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India is located at 11.0139° N, 76.9338° E.

Source of seed

Coriander seeds (Var. CO 5) with an initial moisture content of 9.8 % were obtained from the Department of Spices and Plantation Crops, Tamil Nadu Agricultural University, Coimbatore. Coriander seeds are botanically schizocarp, which has been split into two as mericarp seeds. Mericarp seeds were used in this experiment.

Gas chromatography Mass spectroscopy:

Sample extraction

Coriander seeds were macerated using mortar and pestle and 100 g of powdered sample was taken and extracted with 500 mL of 80 % methanol in ultra sonicator for 90 min. Obtained extract was filtered using 0.45 µ syringe filter.

Gas chromatography Mass spectroscopy analysis

GC-MS analysis of the seed extracts of *C. sativum* was performed using GC-MS Instrument: Agilent GC 7890A / MS5975C. This equipment has Agilent DB5MS capillary column with dimensions of Column Length: 30 m / 0.25 mm internal diameter / 0.25 micron film thickness. Helium is the carrier gas, with a modest flow rate of 1.0 mL/min. The temperature of the oven was programmed as follows, with the injector running at 250 °C: 15 min at 60 °C, followed by a progressive increase to 280 °C in 3 min. For component identification and comparison of the retention indices NIST libraries are used.

Seed leaching

Mericarp coriander seeds were tied in cloth bags and kept in running water for 6 h, 12 h, 18 h, 24 h and 30 h to standardize the best leaching duration. After leaching, seeds were shade-dried to bring back their original moisture content.

Seed leaching + Priming

After leaching treatment best performing leaching seeds were primed with different priming agents 2,4-D at 100 ppm and 150 ppm, H_2O_2 at 25 mM and 50 mM, NO (Nitric oxide) at 25 µM and 30 µM, Salicylic acid at 25 µM and 30 µM at a 1:1 v/v seed-to-water ratio at a standardized

priming duration. Seeds were primed with water for a period of 3, 6, 9, 12, 15 and 18 h to standardize the priming duration.

Seed leaching + Coating

After leaching treatment best performing leaching seeds were coated with TNAU seed coating polymer (Vithai Amirtham) along with different plant growth promoting microorganisms such as *Azospirillum* at 100 g / kg of seed, *B. subtilis* at 10 g/kg of seed, *T. viride* at 4g/kg of seed. TNAU seed coating polymer (Vithai Amirtham) was purchased from the Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore. Quantity of TNAU Seed coating polymer required to coat one kg of seed was standardized and a concentration of 10 g/kg of seeds with 292 mL water was used.

Seed leaching + priming + coating (Comprehensive approach)

Comprehensive seed quality enhancement technique for enhanced germination was developed by combining the best performing seed treatment from seed leaching, seed coating and seed priming through assessing the seed quality parameter.

Germination test

The germination test was conducted in a germination room maintained at 25 ± 1 ºC and 95 ± 2 % RH. Mericarp seeds were used for germination in roll towel with 400 seeds. According to International seed testing Association the final count day for coriander is 21 days. After 21 days seedling was evaluated for various physiological parameter (17).

Germination percentage

Germination % of coriander seeds were calculated using the formula (18):

Germination % = (Number of normal seedlings / Total number of seeds sown) × 100 (Eqn. 01)

Root length and Shoot length

Ten normal seedlings were randomly selected in each of the replications and the length from the base of the hypocotyl to the tip of the primary root was measured using a measuring scale, and the mean was expressed as root length in centimetres. The seedlings measured for root length were again measured for shoot length from the base of the hypocotyl to the tip of the primary leaves and the mean was expressed as shoot length in centimetres.

Dry matter

Following the seedling measurement, 10 typical seedlings were dried for a total of twenty-four hours at 80 °C in a hot air oven and their dry weight was determined.

Vigour index

The seedling vigour index was determined using the method (19):

Vigour index I = Germination % × Total seedling length (cm) (Eqn. 02)

Vigour index II = Germination % × Dry matter production (g per 10 seedlings) (Eqn. 03)

Determination of α-amylase activity

To determine the α amylase activity weigh 500 mg of pre-germinated seed and homogenised using 1.8 mL of cold 0.02 M sodium phosphate buffer (pH 6.0) and later centrifuged at 20000 rpm for 20 min, for enzyme extraction. From the enzyme extract, take 0.1 mL and add 1 mL of 0.067 % starch solution. Then add 1 mL of iodine HCl solution (60 mg of KI and 6 mg of I_2 dissolved in 100 mL of 0.05 N HCl) and incubate at 25 ºC for 10 min. The colour change was measured at 620 nm using a double beam spectrophotometer. The amylase activity was calculated by using the following formula and the mean values were expressed in mg maltose min^{-1} (20).

α- amylase content (mg maltose min^{-1}) = (OD Value / Volume of sample pipetted out) × (1000/500) (Eqn. 04)

Determination of dehydrogenase activity

Mericarp coriander seeds were preconditioned by soaking in water for 6 h. Seeds were bisected longitudinally to expose the embryo. The bisected seeds were steeped in a 0.1 % solution of 2, 3, 5-triphenyl tetrazolium chloride and kept in the dark for 6 h at 40 °C for staining. After staining, the excess solution was decanted and the seeds were washed thoroughly with distilled water and transferred to a 25 mL conical flask containing 10 mL of 2-methoxy ethanol (methyl cellosolve). The conical flask was closed airtight and allowed to remain in the incubator in darkness overnight for extracting the red-coloured formazan. The coloured solution was decanted and the color intensity was measured in an ELICO UV-VIS spectrophotometer (Model SL-159) using a blue filter (470 nm) and methyl cellosolve as the blank. The optical density value obtained was reported as dehydrogenase activity (21).

Determination of total phenols

Coriander seeds were grinded with pestle and mortar using 10 times volume of 80 % ethanol. The homogenized solution was centrifuged at 3000 rpm for 10 min and supernatant was collected. Pipette out 1 mL of supernatant and the volume were made up to 3 mL with distilled water. To that, 0.5 mL of folin - ciocalteau reagent was added. After 3 min, 2 mL of 20 % Na_2CO_3 solution was added and mixed well. Then, the sample was placed in a boiling water bath for 1 min. After cooling, the absorbance was measured at 650 nm in an UV-VIS spectrophotometer. Finally, the concentration of total phenols was calculated by using the standard curve and the mean value was expressed in mg GAE/g (22).

Statistical analysis

The lab experiment was laid out in a Completely Randomized Block design (CRD). Data are given as the mean ± SE of 4 replicates. All data were subjected to statistical analysis using AGRES software. An analysis of variance (ANOVA) was conducted at a significance level of $P < 0.05$, followed by comparisons of treatment means using Duncan's Multiple Range Test (DMRT). Graphical data analysis for, total polyphenols, dehydrogenase activity and α-amylase activity were conducted using the Graph pad software.

Results

Gas Chromatography - Mass Spectroscopy analysis of *C. sativum*

This analysis was conducted to identify the bioactive components of *C. sativum* seeds using GC-MS. The active compounds with their molecular formula, retention time, area and area % was given in Table 1. which indicates the presence of 5 phenolic compounds, 6 terpenes and terpenoids, 5 aldehydes and ketones, 4 carboxylic acids and derivatives, 2 nitrogenese compounds, 1 cyclic compound, 3 other compounds were identified. Among the identified compounds 2H-1-benzopyran-2-one with 49.6 % peak area is the inhibitor of seed germination (Fig. 1).

Effect of leaching duration on physiological and biochemical parameters of coriander seeds

Results observed from the seedling evaluation done after 21 days from the date of sowing, includes germination percentage (G %), root length (RL), shoot length (SL), dry matter production (DMP), vigor index I (VI-I) and vigor index II (VI-II) and biochemical parameters α-amylase and dehydrogenase are given in Table 2. The results revealed that seeds leached for 24 h significantly increased the germination percentage and seedling growth of coriander seeds over control. Leaching (24 h) increases the G %, RL, SL, DMP, VI-I and VI-II over control by 25.8 %, 63.5 %, 18.8 %, 51.2 %, 81.5 % and 91.7 % respectively. And biochemical parameters such as α- amylase, dehydrogenase also significantly increased over control by 31.2 % and 37.6 % respectively (Fig. 2 A). All the seedling parameters increase with an increase in leaching duration up to 24 h. The total phenol content significantly decreases with an increase in leaching duration (Fig. 2 B). The results suggested that Leaching (24 h) will enhance germination percentage and seedling growth and reduce the phenol content that inhibits seed germination.

Table 1. GC-MS spectral analysis of methanolic extract of coriander seeds.

Sl. No.	RT	Compound Name	Molecular formula	Area	Area (%)
		Phenolic Compounds			
1	13.363	2-hydroxyphenethyl alcohol	$C_8H_{10}O_2$	11253	0.44
2	9.897	Ethyl 4-ethoxybenzoate	$C_{11}H_{14}O_3$	70611	2.75
3	9.275	2H-1-benzopyran-2-one	$C_9H_6O_2$	1284954	49.96
4	13.230	Benzenemethanol	C_7H_8O	6167	0.24
5	6.509	Benzeneacetic acid	$C_8H_8O_2$	8640	0.34
		Terpenes and Terpenoids			
1	5.353	1-bromo-2-ethylhexane	C_8H_{17Br}	3082	0.12
2	5.898	1,6-octadien-3-ol, 3,7-dimethyl	$C_{11}H_{18}O_2$	705769	27.44
3	7.420	2,6-octadien-1-ol, 3,7-dimethyl	$C_{10}H_{18}O$	12545	0.49
4	7.175	2,7-octadiene, 4-methyl	C_9H_{16}	12877	0.50
5	8.308	7-methyl-Z-8,10-dodecadienal	$C_{13}H_{22}O$	7105	0.28
6	6.820	Butanoic acid, 3-hexenyl ester	$C_{10}H_{18}O_2$	7817	0.30
		Aldehydes and Ketones			
1	5.131	4-hydroxy-3-methyl-2-butanone	$C_5H_{10}O_2$	4582	0.18
2	8.064	1-hydroxy-2-acetyl-4-methylbenzene	$C_9H_{10}O_2$	26185	1.02
3	12.441	Cyclopentane acetaldehyde, 2-formyl-3-methyl-α-methylene	$C_{10}H_{14}O_2$	8708	0.34
4	10.941	2-quinolinecarboxaldehyde, 8-hydroxy-, oxime	$C_{10}H_8N_2O_2$	7615	0.30
5	12.741	m-nitro benzaldehyde acetyl hydrazone	$C_9H_9N_3O_3$	4351	0.17
		Carboxylic Acids and Derivatives			
1	12.997	Diphenyl sulfone	$C_{12}H_{10}O_2S$	8268	0.32
2	6.742	Oxalic acid	$C_2H_2O_4$	11023	0.43
3	6.998	2-methyl-4-oxopentanoic acid	$C_6H_{10}O_3$	5594	0.22
4	12.808	Methyl isohexadecanoate	$C_{17}H_{34}O_2$	9931	0.39
		Nitrogenous Compounds			
1	5.809	Dimethylamine, N-(neopentyloxy)	$C_7H_{17}NO$	1309	0.05
2	4.320	3-amino-5-methylisoxazole	$C_4H_6N_2O$	75886	2.95
		Cyclic Compounds			
1	9.119	1,2,4,5-cyclohexanetetrol	$C_6H_{12}O_4$	60578	2.36
		Other Compounds			
1	6.453	2-pentyn-1-ol	C_5H_8O	4438	0.17
2	8.886	2,5-dimethyl-2-vinyl-4-hexenenitrile	$C_{10}H_{15}N$	4245	0.17
3	7.286	1-dimethyl(isopropyl) silyloxypropane	$C_8H_{20}OSi$	10117	0.39

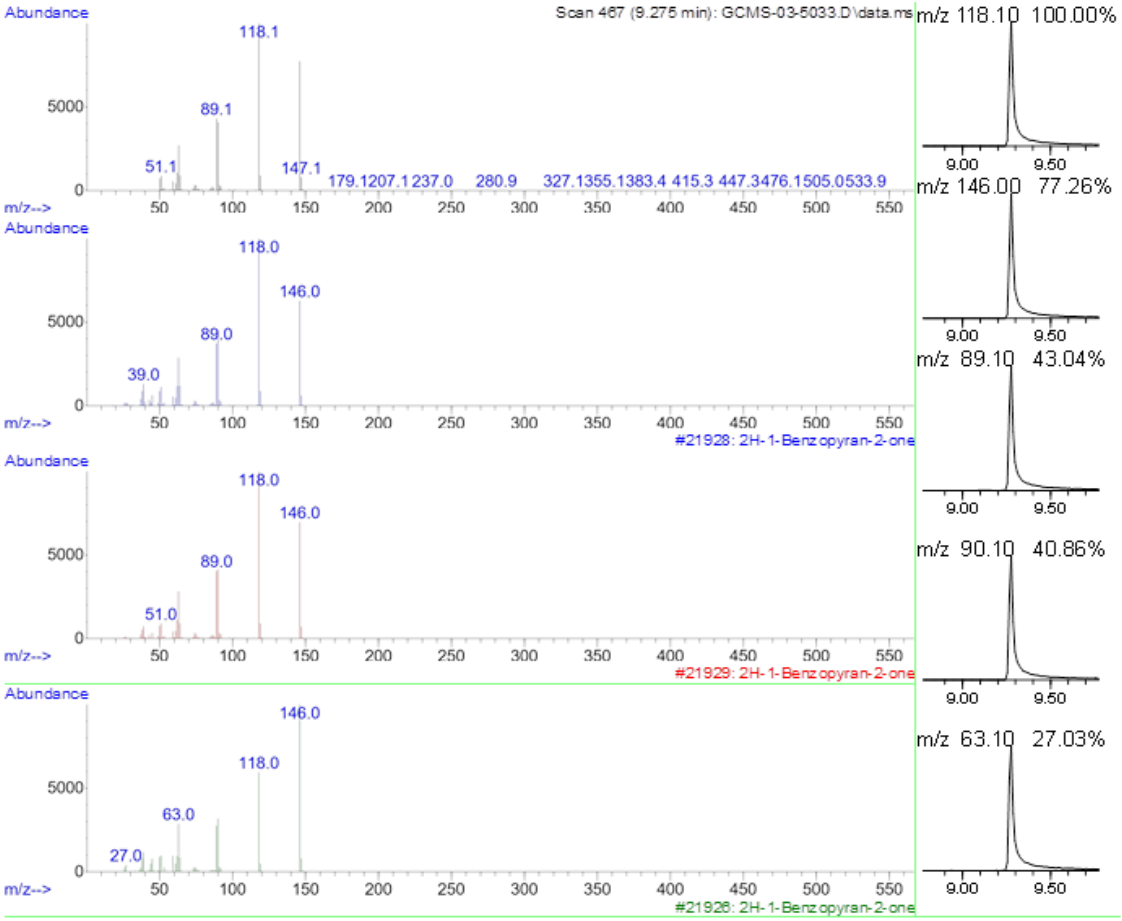

Fig. 1. Mass spectra of coumarin compound from methanolic extract of coriander.

Table 2. Effect of leaching duration on physiological parameters of *Coriandrum sativum.* In each column means followed by the same letter are not significantly different at the P < 0.05 level.

Treatment	Germination (%)	Root length (cm)	Shoot length (cm)	Dry matter production	Vigour index I	Vigour index II
Control	62 ± 6.21^{c}	10.35 ± 0.75^{b}	7.36 ± 0.21^{b}	0.041 ± 0.0016^{d}	1106 ± 139.0^{b}	2.54 ± 0.27^{c}
Leaching (6 h)	64 ± 1.63^{c}	12.98 ± 1.60^{b}	7.68 ± 0.05^{b}	0.048 ± 0.0054^{cd}	1323 ± 112^{b}	3.09 ± 0.42^{bc}
Leaching (24 h)	65 ± 3.79^{c}	12.99 ± 1.01^{b}	7.99 ± 0.10^{b}	0.049 ± 0.0022^{bcd}	1375 ± 151^{b}	3.23 ± 0.19^{bc}
Leaching (18 h)	67 ± 1.00^{bc}	13.04 ± 1.23^{b}	7.99 ± 0.20^{b}	0.054 ± 0.0010^{abc}	1411 ± 99^{b}	3.63 ± 0.06^{b}
Leaching (24 h)	78 ± 1.16^{a}	16.93 ± 1.34^{a}	8.75 ± 0.42^{a}	0.062 ± 0.0063^{a}	2006 ± 156^{a}	4.87 ± 0.54^{a}
Leaching (30 h)	76 ± 1.63^{ab}	16.93 ± 0.97^{a}	8.70 ± 0.15^{a}	0.060 ± 0.0017^{ab}	1952 ± 118^{a}	4.61 ± 0.07^{a}

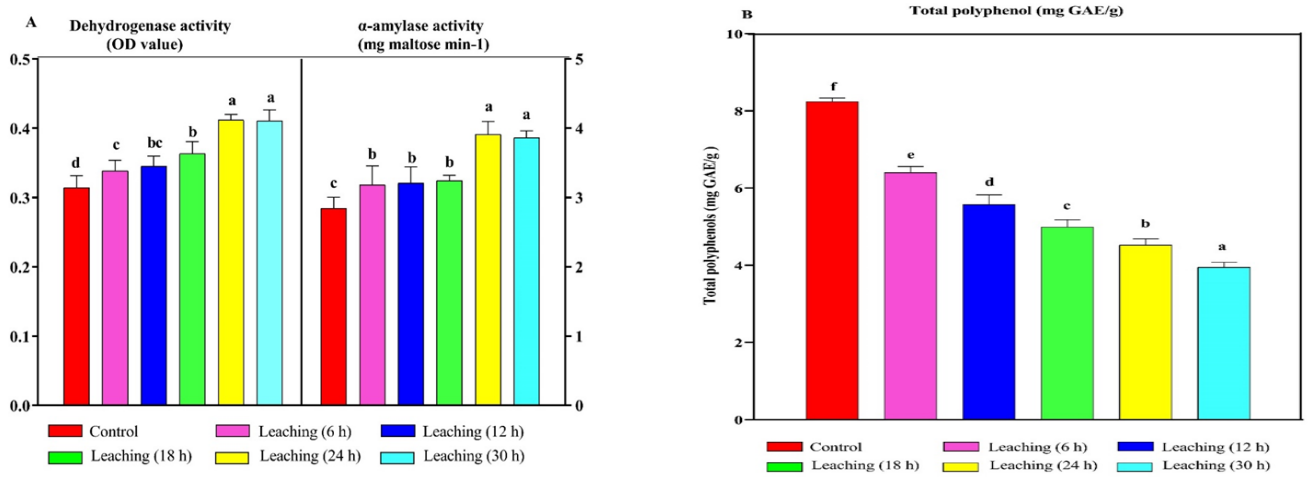

Fig. 2. Effect of leaching duration on biochemical parameters on *Coriandrum sativum* seeds. The same letters are not significantly different at the P < 0.05 level. A - Dehydrogenase and α-amylase activity, B - Total polyphenol.

Standardization of priming duration for enhanced seed germination and seedling growth in coriander

Seed priming experiment was conducted to standardize the duration of seed priming. Results obtained from the seedlings evaluated after 21 days reveals that the seedling parameters such as germination percentage (G %), root length (RL), shoot length (SL), dry matter production (DMP), vigor index I (VI I) and vigor index II (VI II) are given in Table 3. Leaching (24 h) + hydropriming (6 h) significantly increases the germination % over control and leaching (24 h) by 25.7 % and 6.4 % respectively. Root length significantly increases over control and leaching (24 h) by 39.4 % and 6.2 % respectively. Shoot length in leaching (24 h) + hydropriming (6 h) significantly increases over control and leaching (24 h) by 15.3 % and 7.1 % respectively. Dry matter production follows the same trend as root and shoots length. In leaching (24 h) + hydropriming (6 h) vigour index I significantly increase over control and leaching (24 h) by 64.8 % and 13.4 % respectively. Vigour index II significantly increases over control and leaching (24 h) by 105.2 % and 33.1 % respectively. Biochemical parameters α-amylase and dehydrogenase, are given in Fig. 3. Dehydrogenase activity significantly increases in leaching (24 h) + hydropriming (6 h) over control and leaching (24 h) by 47.2 % and 11.2 % respectively. Leaching (24 h) + hydropriming (6 h) significantly increases the α-amylase activity over control and leaching (24 h) by 55.8 % and 17.8 % respectively. The results suggest that seeds after leaching for 24 h subjected to hydropriming for 6 h shows better performance over control and leaching (24 h). Hence 6 h of priming is the best duration of priming for leached coriander seed.

Effect of different priming agents on physiological and biochemical parameters of coriander seeds

The results of seed priming with different priming agents revealed that seed priming significantly increased the

Table 3. Standardization of priming duration of *Coriandrum sativum*. In each column means followed by the same letter are not significantly different at the P < 0.05 level.

Treatment	Germination (%)	Root length (cm)	Shoot length (cm)	Dry matter production (g/10 seedlings)	Vigour index I	Vigour index II
Control	66 ± 2.58^{c}	12.81 ± 0.58^{e}	6.60 ± 0.07^{cd}	0.040 ± 0.003^{d}	1286 ± 92^{e}	2.66 ± 0.18^{d}
Leaching (24 h)	78 ± 1.15^{ab}	16.81 ± 0.30^{abc}	7.10 ± 0.08^{b}	0.052 ± 0.0013^{b}	1866 ± 41^{bc}	4.10 ± 0.13^{b}
Leaching (24 h) + hydropriming (3 h)	78 ± 1.15^{ab}	17.28 ± 0.89^{ab}	7.24 ± 0.08^{ab}	0.054 ± 0.0014^{b}	1912 ± 73^{abc}	4.27 ± 0.05^{b}
Leaching (24 h) + hydropriming (6 h)	83 ± 1.91^{a}	17.86 ± 0.86^{a}	7.61 ± 0.02^{a}	0.065 ± 0.0017^{a}	2120 ± 122^{a}	5.46 ± 0.21^{a}
Leaching (24 h) + hydropriming (9 h)	80 ± 2.30^{ab}	17.45 ± 0.62^{ab}	7.11 ± 0.05^{b}	0.051 ± 0.0003^{b}	1962 ± 47^{ab}	4.12 ± 0.12^{b}
Leaching (24 h) + hydropriming (12 h)	78 ± 1.15^{ab}	16.06 ± 0.45^{bcd}	7.08 ± 0.15^{bc}	0.044 ± 0.0013^{c}	1805 ± 48^{bcd}	3.47 ± 0.13^{c}
Leaching (24 h) + hydropriming (15 h)	78 ± 2.58^{ab}	15.18 ± 0.63^{cd}	6.82 ± 0.31^{bcd}	0.043 ± 0.0010^{c}	1713 ± 68^{cd}	3.37 ± 0.12^{c}
Leaching (24 h) + hydropriming (18 h)	76 ± 1.63^{b}	14.94 ± 0.22^{d}	6.57 ± 0.28^{d}	0.039 ± 0.0032^{cd}	1636 ± 39^{d}	3.03 ± 0.29^{cd}

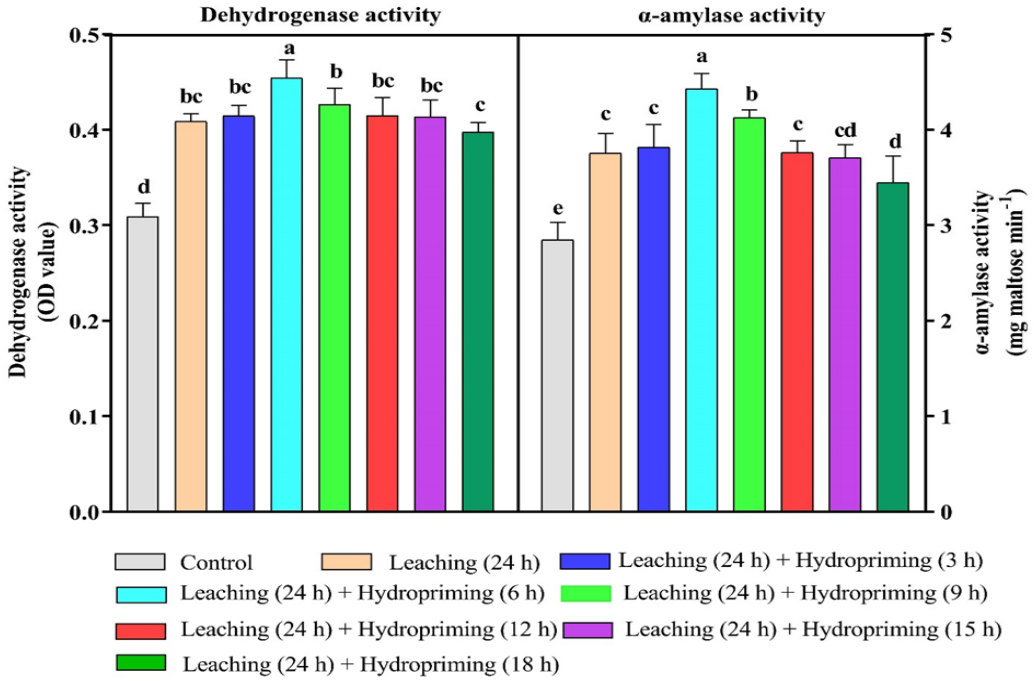

Fig. 3. Standardization of priming duration on biochemical parameters of *Coriandrum sativum*. The same letters are not significantly different at the P < 0.05 level.

germination and seedling growth of coriander seeds in all the treatments over control (Table 4). Leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) shows significantly higher germination % over control by 30.7 %. The root and shoot length were also significantly increases in leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) over control by 59.25 % and 17.1 % respectively. Dry matter production also follows the same trend. Vigour index I was significantly higher in leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) over control by 88.13 %. The dehydrogenase activity was significantly higher in seed priming at 25 mM H_2O_2 (6 h) over control by 58 %. The α- amylase activity increases significantly over control 76.5 % (Fig. 4). The results shows that leaching (24 h) + seed priming at 25 mM H_2O_2 (6h) increased the seedling characters and biochemical parameters in leached coriander seed.

Effect of seed coating with seed coating polymer and different plant growth promoting microorganism on physiological and biochemical parameters of coriander seeds

The results of seed leaching followed by seed coating with polymer and plant growth promoting microorganisms reveals that germination percentage was significantly increases in leaching (24 h) + seed coating with polymer +

Table 4. Effect of priming agent on physiological parameters of *Coriandrum sativum*. In each column means followed by the same letter are not significantly different at the P < 0.05 level.

Treatment	Germination (%)	Root length (cm)	Shoot length (cm)	Dry matter Production (g/10 seedlings)	Vigour index I	Vigour index II
Control	65 ± 1.29^{d}	10.97 ± 1.40^{e}	6.41 ± 0.16^{e}	0.041 ± 0.0003^{g}	1129 ± 99^{e}	2.69 ± 0.06^{e}
Leaching (24 h) + hydropriming (6 h)	82 ± 1.16^{ab}	17.30 ± 0.19^{ab}	7.19 ± 0.13^{ab}	0.061 ± 0.0013^{b}	2009 ± 27^{a}	5.02 ± 0.15^{a}
Leaching (24 h) + priming with H_2O_2 at 25 mM (6 h)	85 ± 1.92^{a}	17.47 ± 0.08^{a}	7.51 ± 0.14^{a}	0.064 ± 0.0012^{a}	2124 ± 51^{a}	5.49 ± 0.22^{a}
Leaching (24 h) + priming with H_2O_2 at 30 mM (6 h)	80 ± 1.63^{bc}	16.17 ± 0.24^{abc}	7.03 ± 0.13^{bc}	0.056 ± 0.0006^{c}	1856 ± 30^{b}	4.52 ± 0.12^{b}
Leaching (24 h) + priming with 2,4D at 100 ppm (6 h)	80 ± 1.63^{bc}	14.94 ± 0.41^{cd}	6.40 ± 0.15^{e}	0.047 ± 0.0010^{ef}	1706 ± 26^{cd}	3.77 ± 0.05^{cd}
Leaching (24 h) + priming with 2,4D at 150 ppm (6 h)	77 ± 1.00^{c}	14.33 ± 0.39^{d}	6.56 ± 0.03^{d}	0.045 ± 0.0011^{f}	1609 ± 39^{d}	3.46 ± 0.09^{d}
Leaching (24 h) + priming with NO at 25 µM (6 h)	80 ± 1.63^{bc}	15.46 ± 0.18^{cd}	6.85 ± 0.03^{cd}	0.048 ± 0.0003^{e}	1785 ± 37^{bc}	3.87 ± 0.07^{bc}
Leaching (24 h) + priming with NO at 30 µM (6 h)	78 ± 1.15^{bc}	15.72 ± 0.34^{cd}	6.51 ± 0.09^{de}	0.051 ± 0.0005^{d}	1735 ± 43^{bcd}	4.03 ± 0.08^{bcd}
Leaching (24 h) + priming with SA at 25 µM (6 h)	80 ± 1.63^{bc}	15.60 ± 0.19^{cd}	6.82 ± 0.05^{cd}	0.052 ± 0.0009^{d}	1793 ± 17^{bc}	4.20 ± 0.07^{bc}
Leaching (24 h) + priming with SA at 30 µM (6 h)	80 ± 1.63^{bc}	15.94 ± 0.17^{bc}	6.65 ± 0.17^{de}	0.053 ± 0.0009^{d}	1807 ± 31^{bc}	4.30 ± 0.16^{bc}

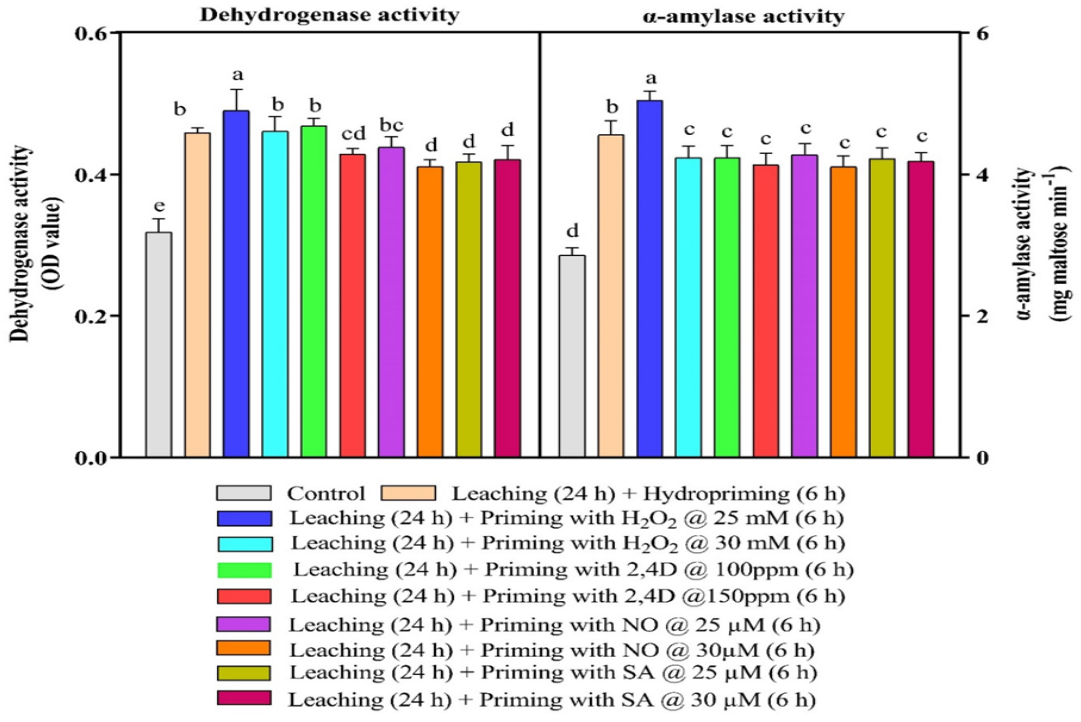

Fig. 4. Effect of priming agent on biochemical parameters of *Coriandrum sativum* seed. The same letters are not significantly different at the P < 0.05

Azospirillum over control by 32.8 %. When compare to control germination percentage of leaching (24 h) + seed coating with polymer + *Trichoderma viride* and leaching (24 h) + seed coating with polymer + *Bacillus subtilis* recorded higher by 26.5 % and 25.0 % respectively. The root length was significantly higher in leaching (24 h) + seed coating with polymer + *Azospirillum,* leaching (24 h) + seed coating with polymer + *Trichoderma viride* and leaching (24 h) + seed coating with polymer + *Bacillus subtilis* over control by 63.7 %, 45.9 % and 45.8 % respectively. The shoot length of leaching (24 h) + seed coating with polymer + *Azospirillum* recorded significantly higher than the control by 21.5 %. Leaching (24 h) + seed coating with polymer + *Trichoderma viride* and leaching (24 h) + seed coating with polymer + *Bacillus subtilis* shows higher shoot length than control by 4.1 % and 9.2 % respectively. Dry matter production was higher in seed coating with leaching (24 h) + seed coating with polymer + *Azospirillum* over control and all other biocontrol agent coatings. Vigour index I was higher in leaching (24 h) + seed coating with polymer + *Azospirillum* over control by 96.9 %. Whereas leaching (24 h) + seed coating with polymer + *Trichoderma viride* and leaching (24 h) + seed coating with polymer + *Bacillus subtilis* recorded higher than control by 65.2 % and 65.4 % respectively. Vigour index II was also higher in seed coating with polymer + *Azospirillum* over control and other biocontrol agent coatings was given in Table 5. The α-amylase activity indicator of dehydrogenase activity was significantly higher in leaching (24 h) + seed coating with polymer + *Azospirillum* over control by 58.9 %. Leaching (24 h) + seed coating with polymer + *Trichoderma viride* and leaching (24 h) + seed coating with polymer + *Bacillus subtilis* recorded higher α-amylase activity than control by 45.9 % and 42.6 % respectively. The dehydrogenase activity shows significantly higher in leaching (24 h) + seed coating with polymer + *Azospirillum* over control by 58.9 %. Leaching (24 h) + seed coating with polymer + *Trichoderma viride* and leaching (24 h) + seed coating with polymer + *Bacillus subtilis* observed higher dehydrogenase activity than control by 32.8 % and 32.1 % respectively. (Fig. 5). The results shows that leaching (24 h) + seed coating with polymer + *Azospirillum* enhanced the

Table 5. Effect of coating with TNAU Vithai amirtham polymer along with different plant growth promoting microorganisms on physiological parameters of *Coriandrum sativum.* In each column means followed by the same letter are not significantly different at the P < 0.05 level.

Treatment	Germination (%)	Root length (cm)	Shoot length (cm)	Dry matter Production (g / 10 seedlings)	Vigour index I	Vigour index II
Control	64 ± 0.81^{d}	10.98 ± 0.35^{d}	6.27 ± 0.05^{d}	0.027 ± 0.0015^{c}	1105 ± 26^{e}	1.73 ± 0.09^{c}
Leaching	81 ± 1.00^{bc}	16.86 ± 0.11^{bc}	7.07 ± 0.03^{bc}	0.052 ± 0.0011^{b}	1939 ± 28^{c}	4.21 ± 0.10^{b}
Leaching + coating with polymer	84 ± 1.63^{ab}	17.04 ± 0.07^{b}	7.04 ± 0.24^{b}	0.054 ± 0.0045^{ab}	2047 ± 24^{b}	4.60 ± 0.40^{ab}
Leaching + coating with polymer and *Azospirillum*	85 ± 1.00^{a}	17.98 ± 0.30^{a}	7.62 ± 0.08^{a}	0.060 ± 0.0029^{a}	2176 ± 46^{a}	5.08 ± 0.24^{a}
Leaching + coating with polymer and *Bacillus subtilis*	80 ± 1.63^{bc}	16.01 ± 0.51^{c}	6.85 ± 0.05^{bc}	0.052 ± 0.0011^{b}	1828 ± 52^{d}	4.14 ± 0.03^{b}
Leaching + coating with polymer and *Trichioderma viride*	81 ± 1.00^{bc}	16.02 ± 0.28^{c}	6.53 ± 0.13^{cd}	0.054 ± 0.0013^{ab}	1826 ± 17^{d}	4.35 ± 0.09^{b}

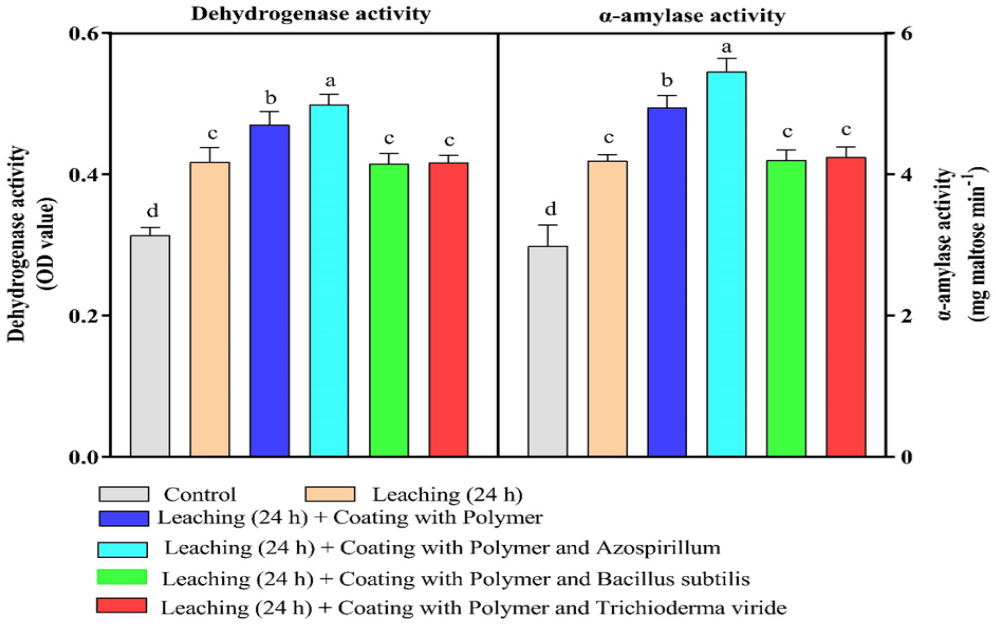

Fig. 5. Effect of coating with polymer along with different plant growth promoting microorganisms on biochemical parameters of *Coriandrum sativum* seed. The same letters are not significantly different at the P < 0.05 level.

germination and biochemical parameters than other biocontrol agents used for coating.

Assessing the effect of different seed management techniques on physiological and biochemical parameters of coriander seeds

The results from the comparison of the 4 established seed management strategies are given in Table 6. Seed germination percentage of leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* shows significant increase in germination percentage (89 %) whereas leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) recorded 84 %, leaching (24 h) + seed coating with polymer + *Azospirillum* recorded 84 %, leaching (24 h) recorded 79 % and control recorded 64 %. The root length of leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* recorded 18.25 cm were significantly higher than all other treatments and control. The shoot length was also significantly higher in leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* (9.51 cm) than all other treatments and control. Leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* recorded higher dry matter production (0.061 g/10 seedlings) than all other treatments and control. The vigour index I recorded 2471 in leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* which is higher when compared to the control and all other treatments. The vigour index II was significantly higher in leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* (5.50) than the control and all other treatments. The dehydrogenase activity in leaching (24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* was recorded significantly higher. α-amylase activity recorded significantly higher in leaching

Table 6. Effect of comprehensive seed enhancement technique on physiological parameters of Coriandrum *sativum* seed. In each column means followed by the same letter are not significantly different at the P < 0.05 level.

Treatment	Germination (%)	Root length (cm)	Shoot length (cm)	Dry matter Production (g / 10 seedlings)	Vigour index I	Vigour index II
Control	64 ± 1.63^{d}	7.39 ± 0.24^{e}	6.57 ± 0.14^{e}	0.029 ± 0.0013^{e}	895 ± 37^{d}	1.89 ± 0.08^{e}
Leaching (24 h)	79 ± 1.91^{c}	13.18 ± 0.13^{d}	8.12 ± 0.11^{d}	0.038 ± 0.0033^{d}	1683 ± 47^{c}	3.04 ± 0.12^{d}
Leaching (24 h) + priming with H_2O_2 at 25 mM (6 h)	84 ± 1.63^{b}	16.18 ± 0.15^{c}	8.51 ± 0.17^{c}	0.046 ± 0.0022^{c}	2075 ± 48^{b}	3.86 ± 0.07^{c}
Leaching (24) + coating with polymer and *Azospirillum*	84 ± 1.63^{b}	17.03 ± 0.28^{b}	9.07 ± 0.02^{b}	0.052 ± 0.0017^{b}	2193 ± 50^{b}	4.43 ± 0.13^{b}
Leaching (24 h) + priming with H_2O_2 at 25 mM (6 h) + coating with polymer and *Azospirillum*	89 ± 1.00^{a}	18.25 ± 0.22^{a}	9.51 ± 0.07^{a}	0.061 ± 0.0017^{a}	2471 ± 35^{a}	5.50 ± 0.10^{a}

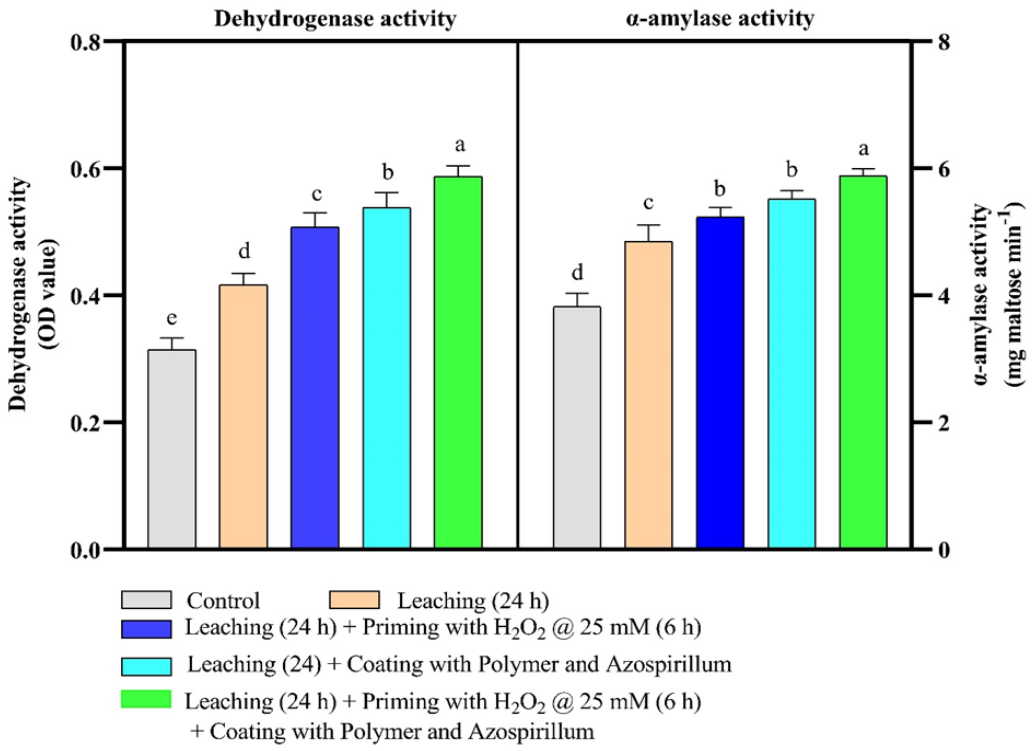

Fig. 6. Effect of comprehensive seed enhancement technique on biochemical parameters of *Coriandrum sativum* seed. The same letters are not significantly different at the P < 0.05 level.

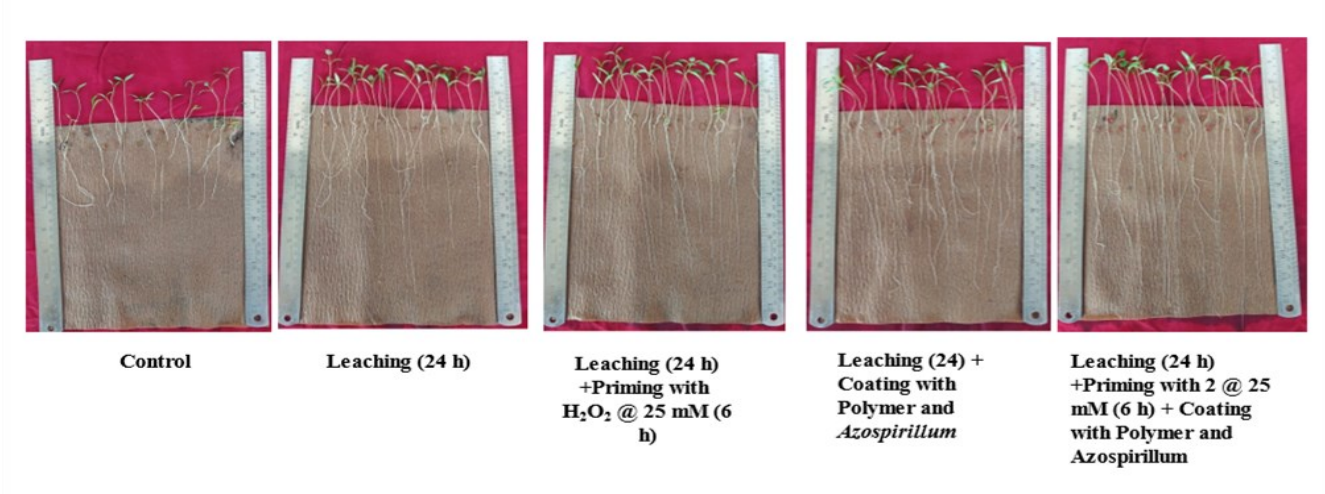

Fig. 7. Effect of comprehensive seed enhancement technique on seed germination and seedling vigour *Coriandrum sativum*.

(24 h) + seed priming at 25 mM H_2O_2 (6 h) + seed coating with polymer + *Azospirillum* (5.89 mg maltose min^{-1}) (Fig. 6).

Discussion

Among the identified compounds coumarin (2H-1-benzopyran-2-one) benzopyrones that are present abundantly in nature and has a physiological role in plant growth (antagonism of auxin), seed germination and dormancy (23). These coumarins may be the reason for inhibition in seed germination.

Phenolic compounds are secondary metabolites derived from phenylpropanoid pathway and these phenolic compounds are present in seed coat and pericarps are responsible for seed dormancy (24). Our results from GC-MS show that presence of coumarin in seeds of coriander. Similarly in other reports also found the presence of coumarin in coriander seeds (25). Coumarin reduces the bioactive GA_4 content, GA_4 is known for its positive regulation in seed germination (26). Leaching of seeds removes the inhibitory substance such as phenols from the seeds thereby improving seed germination. Similar findings were also reported in *Moringa peregrina* where seed leaching for 48 h increased germination percentage and polyphenol content in carob seeds decreased due to leaching which resulted in increased germination (27, 28). Naturally occurring inhibitors such as coumarin content in coriander seeds may be the reason for reduction of root and shoot lengths, dry matter production and vigour index. Similarly, coumarin reduces the root and shoot length of sorghum and wheat seedling (29). Coumarin affects the alpha amylase activity through its inhibitory effect on the expression of amylase genes (30). Our findings show that leaching reduces the concentration of inhibitors present in the seeds. This will improve the seed germination, seedling growth and vigour.

Seed priming increases α-amylase activity (31). That enhances starch degradation process (32, 33). These improvements help in strong and uniform establishment of seedlings. Seed priming enhances protein synthesis, DNA repair, RNA stabilization (34). Similar results were obtained in maize seeds where 36 h of priming increases the germination (35). In bitter gourd 72 h of priming enhances seedling length, dry weight, vigour over control (36). In sweet basil hydro priming for 18 h increased the germination percentage significantly whereas higher concentration 24 h decreases the germination (37). Similarly In Punjab canola and Faisal canola seed priming for 24 h shows shoot length significantly increases over control (38). This may be attributed to seed priming with several chemicals and by various methods leads to significant improvement in seed germination percentage, uniform seedling emergence in field through controlled hydration of seeds. Further this activates several metabolic processes such as enzyme activation, mobilization of resources for early sprouting and radical emergence (39). Seed priming with H_2O_2 improves germination by disturbing the hard seeds and allowing imbibition of water (40). H_2O_2 is a strong oxidizing agent (41). These phenolic contents can be oxidized by the H_2O_2 leads to removal of inhibitors (42). ABA catabolism has been up-regulated by the H_2O_2 through NO signalling and also promotes biosynthesis of gibberellic acids (43). H_2O_2 increases the concentration of ascorbate peroxidase (APX), ascorbate oxidase (AAO) and peroxidase. An increase in these enzymes shows increased seedling growth, and the redox state of the ascorbate decreases. Cytosolic and stromal APX transcript levels were increased due to the increase in APX activity (44).

Seed coating is the process of delivering the plant growth promoting substances or micro-organisms to the seeds for improving the seed quality and plant growth (45). Similar findings such as, seed coating with bio stimulants improves the shoot and root length as compared to control in broccoli (46). In red clover (*Trifolium pratense* L.) and perennial ryegrass, shoot length, dry matter production and vigour of coated seeds were significantly higher than control (47). Increase in root growth might be due to the production of bacterial phytohormones especially biosynthesis of indole-3-acetic acid (IAA) (48). Seeds treated with *Azospirillum* enhance the amylase activity during germination (49). Gibberellins secreted by the bacteria may be the reason for increase in amylase content. Seed treatment with beneficial microorganism has increased the germination, vigour and

biomass, mitigates stress at the time of emergence and after emergence also (50).

Conclusion

Based on the results of all-developed management techniques, 24 h of leaching followed by 6 h of priming with H_2O_2 at 25 mM concentration and seed coating with polymer + *Azospirillum* overcome the dormancy caused by the coumarin through leaching and priming whereas coating increases the seedling growth and stability of seed and seedling establishment. This comprehensive enhancement technique increased the germination and seedling growth in coriander seeds. The influence of comprehensive enhancement technique of coriander seeds on seed longevity and their performance on field remains unexplored. Future research in these areas promises to advance our understanding of coriander cultivation, contributing to food security and sustainable agricultural practices.

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Authors' contributions

SP-V carried out the seed treatments, participated in the statistical analysis and preparation of the manuscript. KS carried out the setup of experimental details. UR coordinated the entire research. RV and UD participated in the evaluation of seedlings. MK and TH carried the manuscript editing and preparation of final draft. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this manuscript, the authors did not use any generative AI or AI-assisted technologies. The content is totally the result of the authors original work and they take full responsibility for the content of the publication.

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