https://doi.org/10.26651/allelo.j/2024-62-1-1488 0971-4693/94 Euro 20.00
Allelopathy Journal 62 (1): 85-96 (May, 2024) *International Allelopathy Foundation* 2024
Table: 4, Figs: 1

Effects of leaf extracts of five tree species on germination and seedling growth of millets, *Paspalum scrobiculatum* and *Panicum miliaceum*

C. Swaminathan*, K. Sangeetha[1], P. Nivethadevi[2], M.M. Yassin and R. Renuka[3]
Tamil Nadu Agricultural University, Coimbatore-641003, India
[1,2]Research scholars in Agronomy, Agricultural College and Research Institute, TNAU, Madurai
E. Mail: brownrevolution@yahoo.com

(Received in revised form : February 22, 2024)

ABSTRACT

We analyzed the biochemical compounds by GCMS and did bioassay of leaf extracts of five tree species viz., *Aegle marmellos* (L.) Correa, *Annona squamosa* L., *Mangifera indica* L., *Morinda tinctoria* Roxb and *Moringa oleifera* Lam. on germination and associated characteristics of kodomillet (*Paspalum scrobiculatum* L) and prosomillet (*Panicum miliaceum* L.). Response Index was used for comparison, and by GCMS, the compounds found in the leaves were determined and studied their effects on germination and seedling growth. The 5 % *Moringa oleifera* leaf extract, followed by extracts from *A. squamosa* and *Mangifera indica*, improved millets' seed germination. Perhaps because the extracts contained the dominant compounds 2-Piperidinone and 2-Pyrrolidinone. Tree leaf extracts from the above-donor species had a synergistic effects in prosomillet, stimulating germination by 14-18 %, than kodomillet. However, *Morinda tinctoria* and *Aegle marmellos* leaf extracts inhibited the germination of both millets than control.

Key words: *Aegle marmellos*, *Annona squamosa*, Bioassay, GCMS, *Mangifera indica*, *Morinda tinctoria*, *Moringa oleifera*, seed germination, tree leaf extracts

INTRODUCTION

The main millet producing nations are in Asia and Africa. One of the earliest crops known to mankind prosomillet (*Panicum miliaceum* L.) is most extensively produced (6). *Paspalum scrobiculatum* L, (kodomillet) has originated in India. The emerging practice of organic farming across the globe offers a lot of scope for alternatives to synthetic products to promote crop growth and development, and natural biostimulants (plant-derived substances) are one such alternative that can promote growth, seed germination, foliar nutrition, resistances to abiotic stresses, and post-harvest quality with no adverse effects (10,33). Seed soaking before planting mitigates the detrimental effects of high temperatures and increases seed germination. It provides enough moisture to the seed, which may allow it to germinate more rapidly. Though seed soaking with tree extracts (botanicals) has been employed in recent times, the extracts of few species have negative and inhibitory effects on seed germination and growth due to presence of phytotoxins present in the extracts (19) rather than osmotic inhibition since the extract offers low osmolality (28). Gas chromatography-mass spectrometry (GC-MS), is used to determine such components and their role (26) in germination and growth of plants.

M. oleifera leaf extract (MLE) is gaining popularity as a biostimulant due to its superior efficacy to synthetic growth promoters, low cost (2,13,15) and presence of hormones (auxins, gibberellins and cytokinin), as well as minerals and antioxidants. It

*Correspondence author, [3]Professor (Biotechnology), Agricultural College and Research Institute, TNAU, Madurai

protects cells from the damaging effects of reactive oxygen species on ageing by enhancing antioxidant activities of various enzymes (31). In irrigated cotton, *A. squamosa* leaf extract had a comparable impact (8). The barnyard millet produced the largest amount of leaf chlorophyll, when *A. squamosa* leaf extract was applied as a foliar spray at the flowering stage; *A. squamosa* leaf extract applied as a 5 % spray in okra (*Abelmoschus esculentus*) produced higher yield, and when *M. indica* leaf extract was sprayed on foxtail millet at a dosage of 5 %, the highest growth and physiological indicators were noted (27,28). In contrast, prosomillet millet residue extracts inhibited the test crop seed germination and seedling growth (radicle length and plumule length). According Abadalla *et al.* (1) maize and other millets' seed germination and seedling growth were hindered by the shoot, root, and biomass residue extract of prosomillet because phytochemicals regulated the water-nutrient interactions and heat responses.

This study aimed (i) to test the influence of 5-trees leaf extracts based on two decades of research (27) on seed germination and seedlings growth of kodomillet and prosomillet and (ii) to identify the principal compounds causing positive or inhibitory effects of these donor tree leaf extracts, which could be further screened for preparations of biostimulants on a large scale.

MATERIALS AND METHODS

The laboratory experiments were setup in Seed laboratory, Agricultural College and Research Institute, Tamil Nadu Agricultural University, Madurai, India.

I. Bioassay

***Test crops*:** Kodomillet cv. CO3 and Prosomillet cv. ATL 1

Collection and preparation of botanicals/leaf extracts: Fresh, spotless, clean leaves from the campus orchard were gathered, pulverized in 1:1 ratio with distilled water, and the extracts were filtered before being preserved as stock solution. Kodomillet and prosomillet seeds were soaked in this 5 % solution for 12 h (27). The intensity of inhibition *vs.* stimulation in the bioassay was compared (24), which indicated that fresh leaf extract concentrations > 10 % inhibited seed germination; hence, this investigation was done with 5 % concentration.

Donor Tree species

The tree species' leaf extracts such as *Aegle marmellos* (L.) Correa, *Annona squamosa* L., *Mangifera indica* L., *Morinda tinctoria* Roxb. And *Moringa oleifera* Lam. were used as they may affect the physiological and biochemical changes during germination and growth

Setup of lab experiment

According to ISTA (12), roll towel method was used to assess germination, which involves placing seeds on germination paper and rolled up in vertical standing rolls. Twenty five seeds of each, kodomillet and prosomillet were placed on each germination paper, and stored vertically in sterilised conical flasks with 5-leaf aqueous extractions at the bottom (22). Simultaneously, a separate germination test was done in petriplates (9 cm dia) lined with germination paper to study speed of germination. For both tests, distilled water served as control. Three replications were used in the experiments set up in

a Completely Randomized Design (CRD). Germination and related attributes in treated and control seedlings were investigated till 10 days. At end of the study, total seedling length was recorded and germination (%) was calculated. A daily count of germinants was used to construct a different germination metrices, including germination energy and emergence energy value. The data observed on germination and growth parameters were analysed statistically and subjected to ANOVA (Analysis of Variance) at 5 % probability level for their differences (23).

Measurement and analysis of data

Germination and seed quality traits were evaluated in both treated and control seeds. From the start of germination to 10 days, the number of germinants were counted every day. Per cent germination, shoot length, root length and vigour index I (% germination x total seedling length) were calculated from the daily count of seedling (3). Other germination parameters (i) germination energy (GE) and (ii) emergence energy value (EEV) were evaluated. Using a single index, does not accurately describe the allelopathic effects on the germination process. A comparison of different indices can make allelopathy a more accurate science.

Germination Energy

Germination energy (GE) measures the seed ability for faster germination and calculated by the formula proposed by (17)

$$GE = \frac{X1}{Y1} + \frac{X2-X1}{Y2} + \ldots\ldots\ldots\ldots + \frac{Xn-Xn-1}{Yn}$$

Where, X_n : number of germinant on n^{th} counting date; Y_n : number of days from sowing to n^{th} count.

***Emergence Energy Value* (5)**

The seed germination capacity and germination energy decide the early emergence, establishment and survival of seedlings. It represents both the emergence of the seed and the energy within the seed. If the seed has a higher germination energy this will be reflected in its early seedling emergence with higher values. Germination (%) was calculated daily, this was then divided by the number of days since the start of test. This calculation is done daily until the end of test. The highest obtained value is EEV.

Response index

In bioassays, magnitude of inhibition vs. stimulation was assessed using response index (RI) (24, 29), which was calculated as under:

if T >C, RI=1-(C/T)
if T = C, then RI = 0
if T<C, then RI= (T/C) -1

Where, T: Treatment mean and C: Control mean. A negative RI reflects proportional disparity in output (germination, radicle length or plumule growth), a positive RI reflects the stimulation in output of test crop in treatment relative to output in control.

II. GC-MS analysis

Five g of powdered leaf sample was extracted with 50 ml milli-Q water in a mechanical shaker at 250 rpm for 3 days continuously. To concentrate the sample, the

extracted material was filtered through Whatman No. 40 filter paper and kept under boiling water bath. To re-dissolve this sample, 2 ml of methanol solvent was used and then filtered through a Paul membrane filter by injecting the sample into 2 ml syringe. The sample was used for GC-MS analysis using GC-MS QP 2020 coupled with MSD. The GC fortified with Rxi-5 Sil MS fused silica capillary column (30mL x 0.25 D x 0.25 µm thickness). Interpretation on GCMS was drawn using database of national institute standard and technology (NIST) having more than 62,000 patterns. The spectrum of unknown component was compared with spectrum of known components stored in NIST library. Peak compounds were identified based on retention time and area spread.

RESULTS AND DISCUSSION

I. Bioassay

(i). Germination, shoot length, root length and vigour index

Kodomillet and prosomillet germination was found to be greatest in *Moringa oleifera* leaf extract (92 and 78.67 %, respectively), which is 19 % and 14 % higher than control (86 %, 57.3 %, respectively for kodomillet and prosomillet), and statistically significant at 5 % probability level (Table 1). The critical value for significant difference is 5.81. Both crops showed equal response to *A. squamosa* and *M. indica* leaf extracts in terms of germination. The presence of active ingredients, micronutrients, and biochemical components such as phenolic compounds, organic acids, proteins, and alkaloids (25) in botanical leaf extracts favours higher germination (%), which is crucial factor in determining seed volume and viability (21). This lends credence to recent findings. Leaf extracts of *M. tinctoria* and *A. marmellos* decreased germination in kodomillet as compared to control, demonstrating a negative trend. This may be caused by the presence of toxic phytochemical substances that hinder germination (19,22). The results of this study show that kodomillet had slower seedling development but better germination. Prosomillet had less germination than kodomillet, but it developed seedlings more successfully regardless of how the seeds were treated with various tree leaf extracts. The vigour index and seedling characteristics both showed this behaviour. *M. oleifera* leaf extract registered maximum root length (9.21 cm), shoot length (7.75 cm), and vigour (1560) in kodomillet and 10.93 cm, 10.93 cm, and 1718 cm, respectively, in prosomillet. The corresponding values in control were 7.73, 6.46, 1220 in kodomillet and 9.4, 9.4, 1076 in prosomillet, respectively. *M. indica* leaf extract came in second place. *M. oleifera* leaf extract produced longest roots in millets as evidenced from findings of (11,31). This might be owing to germination inhibition is presence of inappropriate phytochemical compounds in Moringa leaves (19,22). For root length and shoot length, the critical value for significant variations is 0.55 and 0.56, respectively.

With the exception of *M. tinctoria* and *A. marmellos*, tree leaf extracts generally had a positive effect on augmenting the germination and seedling characteristics. These two species had suppressed millet seed germination and decreased the vigour index. The vigour index, as well as germination and seedling growth, were increased by the other three leaf extracts in both millets, with *M. indica* and *M. oleifera* having the maximum effects.

Table 1. Influence of tree leaf extracts on germination, seeding length and vigour index of millets

Donor Plant	Leaf Extract Conc (%)	Germination (%)	Root length (cm)	Shoot length (cm)	Vigour Index
	Recipient Kodomillet (*Paspalum scrobiculatum* L.)				
Control		86[b]	7.73[c]	6.46[c]	1220[b]
Annona squamosa	5 %	90[ab]	7.82[b]	7.1[b]	1313[a]
Aegle marmelos	5 %	66[c]	7.4[c]	5.7[c]	865[b]
Mangifera indica	5 %	88[a]	8.08[b]	8.4[a]	1452[a]
Moringa oleifera	5 %	92[a]	9.21[a]	7.8[b]	1560[a]
Morinda tinctoria	5 %	68.6b	7.27c	6.0c	911b
	Recipient Prosomillet (*Panicum miliaceum* L.)				
Control		57.3[c]	9.4[d]	9.4[c]	1076[c]
Annona squamosa	5 %	73.3[ab]	11.3[a]	11.1[a]	1639[a]
Aegle marmelos	5 %	66.7[c]	10.8[a]	10.8[a]	1442[b]
Mangifera indica	5 %	76.0[a]	10.0[c]	9.7[c]	1497[b]
Moringa oleifera	5 %	78.7[a]	10.9[a]	10.9[a]	1718 [a]
Morinda tinctoria	5 %	68.0[b]	9.8[c]	9.7[c]	1327[b]

(ii). Germination attributes

Germination energy measures how quickly seeds germinate and how that affects the viability and vigour of seedlings (12). For kodomillet and prosomillet seeds, respectively, the leaf extract from *Moringa oleifera* showed a significant effect and maximal germination energy of 2.31 and 2.27 (Table 2). Leaf extracts from *A. squamosa* and *M. indica* came next. It's interesting to notice that the leaf extracts from *M. tinctoria* and *A. marmellos*, which had weak effects on millets' germination per cent and seedling growth, had more of an impact on the germination's initiation by initially reflecting larger emergence energy values (EEV) but failed to maintain them. It might be caused by the molecule responsible for an early impact volatilizing with time, which needs further research. This may be because tree leaf extracts include a variety of phytochemicals, including terpenoids, phytosteroids, fatty acids, glycosides, essential oils, saponins, phenols, and flavonoids, which function as a precursor to GA3 due to saponins (25).

Table 2. Influence of tree leaf extracts on germination related indices

Donor plant	Germination Energy		Emergence Energy Value	
	Kodomillet	Prosomillet	Kodomillet	Prosomillet
Aegle marmellos	2.23*[c]	1.92[d]	18.7[b]	15.6[b]
Annona squamosa	2.30[b]	2.00[c]	25.0[a]	13.8[c]
Mangifera indica	2.00[d]	2.08[b]	16.1[c]	15.6[b]
Moringa oleifera	2.31[a]	2.27[a]	16.7[c]	19.3[a]
Morinda tinctoria	2.23[c]	1.83[c]	18.0[b]	14.4[c]

- The coefficient of variation (CV) for the Germination Energy was 6.54 and 8.31 for kodomillet and prosomillet respectively, indicating that the reliability of the calculated values and. Similarly the values for Emergence Energy are 18.84 and 13.58. The lower the value of the coefficient of variation (< 20 %), the more precise the estimate and the numerical differences reflect the influence of treatments on the parameter studied.

(iii). Effects on Response Index

Individual tree leaf extracts changed both millets' germination and seedling growth (Table 3), resulting in a range of response index values compared to the control at 5 % probability level. Except for negative responses in *M. tinctoria* and *A. marmellos* leaf extracts, the Response Index for the three parameters viz., germination, root length, and shoot length were primarily positive and higher. Kodomillet seed germination was decreased by *M. tinctoria* and *A. marmellos* leaf extracts and was little influenced by other leaf extracts with a per cent increase of 2.3 to 6.5. While prosomillet seed germination showed the highest Response Index with *M. indica* leaf extract and 24.6 % higher germination compared to control. The least increase in seed germination of prosomillet was spotted with *A. marmellos* leaf extract**.** Despite having a higher response index for prosomillet seed germination, *M. oleifera* leaf extract had no effect on seedling growth. However, *A squamosa* demonstrated a higher and more favourable response index for prosomillet seedling growth. The tree leaf extracts had a poor response index for kodomillet in all the parameters examined when compared to prosomillet, at 5 % probability level. Three of the five tree leaf extracts used in this investigation had positive RI values, indicating that all extracts are natural growth stimulants (Figure 1).

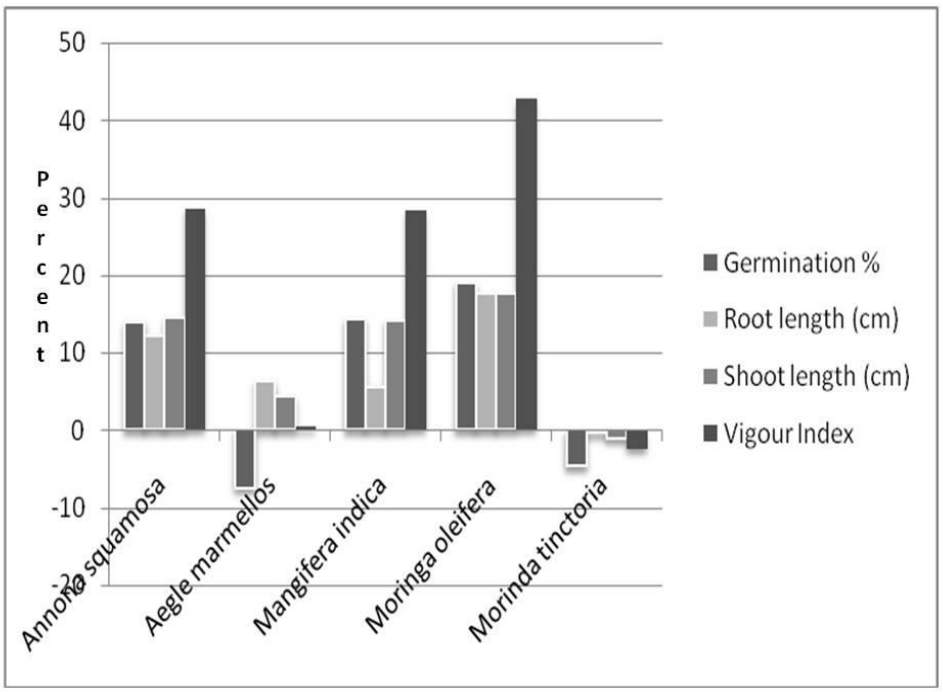


Figure 1. Effects of tree leaf extracts germination and seedling growth of millets compared to control

Table 3. Response index for germination and seedling growth of millets in response to tree species leaf extract

Donor Plant	**Leaf Extract Conc (%)**	**Germination (%)**	**Root length (cm)**	**Shoot length (cm)**
	Recipient Kodomillet (*Paspalum scrobiculatum* L.)			
Control				
Annona squamosa	5 %	+0.044*	+0.012	+0.086
Aegle marmelos	5 %	-0.902	-0.043	-0.118
Mangifera indica	5 %	+0.023	+0.043	+0.233
Moringa oleifera	5 %	+0.065	+0.161	+0.166
Morinda tinctoria	5 %	-0.902	-0.060	-0.071
	Recipient Prosomillet (*Panicum miliaceum* L.).			
Control				
Annona squamosa	5 %	+0.218	+0.168	+0.148
Aegle marmelos	5 %	+0.140	+0.130	-0.0276
Mangifera indica	5 %	+0.246	+0.060	+0.119
Moringa oleifera	5 %	+0.271	+0.140	+0.137
Morinda tinctoria	5 %	+0.157	+0.041	-0.0106

(*Calculated values and hence data not statistically analysed)

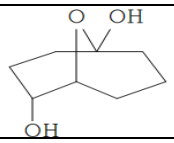
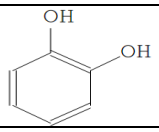
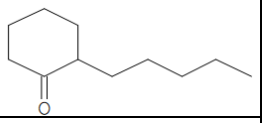
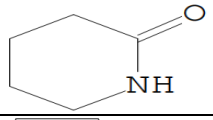
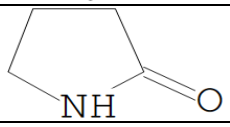
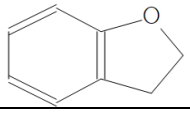
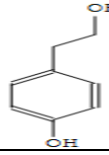
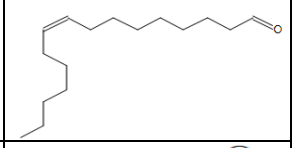
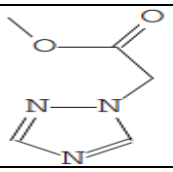
II. Identification of primary compounds in tree leaf extracts by GC-MS

The principal compounds found in tree leaves were identified via GC-MS analysis have a scientific explanation of cause and effect by revealing the chemistry behind the beneficial influence of tree leaf extracts on seed germination and associated attributes. According to Lopez-Amoros and coworkers (16), germination alters the quantitative and qualitative phenolic compounds of legumes, with the alterations varying depending on the kind of legume and germination conditions. As a result of variations in antioxidant activity, these alterations have an impact on the functional mechanisms of legumes. Terpenoids are the most abundant compounds reported in *Glochidion ellipticum* leaves next by alkaloid compounds and esters of fatty acid compounds (14). The current study scientifically proves that the oldest Indian agricultural system relied on natural compounds found in plant and tree leaves for food production, and that a concept of positive allelopathy should be developed and studied further to develop biostumulants to produce high-quality food grains without the use of modern pesticides, herbicides, and agrochemicals.

GC-MS analysis

Using Gas-Chromatography and Mass Spectrometry (GCMS) we identified the bioactive chemicals (14,32) leaf extracts of 5-donor tree species. The solvent Milli-Q water was used for chemical extraction and qualitative analysis.

Table 4. Major compounds identified in the five tree leaf extracts

S. No.	Tree species	Compound name	R TIME (min.)	Area %	Height %	Formula	Molecular structure
1	*Morinda tinctoria*	9-Oxa-bicyclo[3.3.1]nonane-1,4-diol	22.660	15.90	10.77	$C_8H_{14}O_3$	
2		1-Heptatriacotanol	23.962	9.14	8.07	$C_{37}H_{76}O$	
3	*Mangifera indica*	Catechol	13.241	15.50	9.36	$C_6H_6O_2$	
4		Cyclohexanone, 2-pentyl-	16.986	15.05	18.53	$C_{11}H_{20}O$	
5	*Moringa oleifera*	2-Piperidinone	12.571	26.50	10.96	C_5H_9NO	
6		2-Pyrrolidinone	9.464	11.92	6.34	C_4H_7NO	
7	*Annona squamosa*	Benzofuran, 2,3-dihydro	13.550	21.48	16.29	C_8H_8O	
8		Benzene ethanol, 4-hydroxy	19.408	13.13	9.15	$C_8H_{10}O_2$	
9	*Aegle marmellos*	cis-9-Hexadecenal -	24.730	14.08	10.88	$C_{16}H_{30}O$	
10		1H-1,2,4-Triazole-1-acetic acid, methyl ester	18.001	11.99	9.62	$C_5H_7N_3O_2$	

A. squamosa leaf Milli Q water extracts had the highest concentrations of the phytochemicals (27,28) such as Benzofuran, 2,3-dihydro (21.48 %) and Benzene ethanol, 4-hydroxy (13.13 %), with retention times of 13.550 and 19.408 minutes, respectively (Table 4). With retention times of 24.730 and 18.001 minutes, respectively, the top chemical components found in *A. marmellos* were cis-9-Hexadecenal (14.08 %) and 1H-1,2,4-Triazole-1-acetic acid, methyl ester (11.99 %). 2-Piperidinone (26.50 %) and 2-Pyrrolidinone (11.9 %) had the largest area percentage compound found in *M. olerifera* leaf extracts, and their respective retention times were 12.571 and 9.464 minutes. The highest area percentage compound identified from *M. indica* leaf was Catechol (15.50 %) and 2-pentyl cyclohexanone (15.05 %) with the retention time of 13.241 and 16.986 minutes, respectively. The peak bioactive compounds identified in Milli Q water extracts of *M. tinctoria* leaf were 9-Oxa-bicyclo[3.3.1]nonane-1,4-diol (15.90 %) and 1-Heptatriacotanol (9.14 %) with the retention time of 22.660 and 23.962 minutes, respectively.

CONCLUSIONS

Millet seeds fortified with 5 % leaf extract of *M. oleifera* and *A. squamosa* had higher germination, seedling length and seedling vigour index. Three of 5-donor tree leaf extracts stimulated response index (RI) values for all the parameters evaluated. *M. tinctoria* and *Aegle marmellos* extracts had effects on both kodomillet (*Paspalum scrobiculatum*) and prosomillet (*Panicum miliaceum* L.) seed germination but stimulated seedling development at later stage. The *Moringa oleifera* and *Annona squamosa* leaf extracts at 5 % concentration stimulated the seed germination and seedling growth of kodomillet (*Paspalum scrobiculatum* L) and prosomillet (*Panicum miliaceum* L.). But aqueous leaf extract of *M. tinctoria* and *A. marmellos* leaf extracts were inhibitory to germination of kodomillet (*Paspalum scrobiculatum*) Seed germination stimulation showed positive allelopathic activity. The GC-MS study detected the abundance of Benzofuran, 2,3-dihydro (21.48 %) and Benzene ethanol, 4-hydroxy in *Annona squamosa* leaf extract and 2-Piperidinone (26.50 %) and 2-Pyrrolidinone (11.9 %) had the largest area percentage compound found in *Moringa oleifera* leaf extracts and their respective retention times were 12.571 and 9.464 minutes. After more research, such tree botanicals could be developed as biostimulants to improve the seed germination and seedling growth to replace synthetic agents for organic farming.

ACKNOWLEDGMENTS

The senior authors wish to thank for analytical facilities provided by Centre of Innovation, and GOI-DST-FIST agronomy labs of Agricultural College and Research Institute, TNAU, Madurai, India.

DECLARATION

C. Swaminathan, designed and planned and conducted experiment and wrote the article K. Sangeetha and P. Nivethadevi involved in supervising the experiment and data collection and analysis. M.M. Yassin assisted in editing the draft. R. Renuka involved in Biochemical analysis.

CONFLICT OF INTEREST

The authors announce that they have no conflict of interest.

ETHICAL APPROVAL

The authors declare that the study was carried out following scientific ethics and conduct.

REFERENCES

1. Abdalla. I.A. Ahmed and Hou, F.J. (2021). Allelopathic effects of Prosomillet (*Panicum miliaceum* L) extracts on seed germination and seedling growth of other millets and maize. *Allelopathy Journal* **54 (2)**: 157-168.
2. Abdel-Rahman, S.S.A. and Abdel-Kader A.A.S. (2020). Response of Fennel plants to foliar application of moringa leaf extract and benzyladenine. *South African Journal of Botany* **129**: 113-122.
3. Abdul-Baki A.A. and Anderson J.D. (1973). Physiological and biochemical deterioration of seeds. In: *Seed Biology* (Ed., T.T. Kozlowski), Academic press, New York Vol. **2**: 283-315
4. Ahmed, A., and El-Mahdy, A. (2022). Improving seed germination and seedling growth of maize (*Zea mays* L.) seed by soaking in water and Moringa oleifera leaf extract. *Current Chemistry Letters* **11(2)**: 147-156
5. Bahuguna, V.K, Pyare L. and Dhawan, V.K. (1987). Standardisation of nursery techniques (seed sowing methods and watering schedules) of Eucalyptus FRI- 4 under North Indian moist tropical climatic conditions. *Indian Forester* **113**: 541-549
6. Baltensperger, D.D. (1996). Foxtail and prosomillet. In: *Progress in New Crop* (Ed., J. Janick). ASHS Press, Alexandria, Pp. 182-190.
7. Basra, M.A.S. and Lovatt, C.J. (2016). Exogenous applications of moringa leaf extract and cytokinins improve plant growth, yield, and fruit quality of cherry tomato. *Hort Technology* **26**: 327-33.
8. Chandrasekaran, H. and Venkatesan, M. (2018). Evaluation of intercropping system, nutrient management and tree leaf extract spray on irrigated cotton. *Asian Journal of Biological Sciences* **11**: 217-222
9. Changmei, S. and Dorothy, J. (2014). Millet-the frugal grain. *International Journal of Scientific Research and Reviews* **3 (4):** 75-90. https://www.ijsrr.org/down_394.php.
10. Del, B.D., Bartucca, M.L., Ballerini, E., Senizza B., Lucini, L. and Trevisan, M. (2021). Physiological and biochemical effects of an aqueous extract of *Lemna minor* L. as a potential biostimulant for maize. *Journal of Plant Growth Regulation* **14**: 1-10.
11. Iqbal, J., Irshad, J., Bashir, S., Khan, S., Yousaf, M. and Shah, A.N. (2020). Comparative study of water extracts of moringa leaves and roots to improve growth and yield of sunflower. *South African Journal of Botany* **129**: 221-224.
12. ISTA, (2006). ISTA Method validation for seed testing. ISTA Secretariat, Switzerland pp. 1-70.
13. Jain, P., Farooq, B., Lamba, S. and Koul, B., (2020). Foliar spray of moringa leaf extracts (MLE) enhances stevioside, zeatin and mineral contents in *Stevia rebaudiana*. *South African Journal of Botany* **132**: 249-257
14. Jayashree, I., Geetha, D. and Rajeswari, M. (2015). GC-MS analysis of bioactive constituents of *Glochidion ellipticum* Wt. *International Journal of Pharmaceutical Sciences and Research* **6 (6)**: 2546-2550.
15. Khan, S., Basra, S.M.A., Afzal, I., Nawaz, M. and Rehman, H.U. (2017). Growth promoting potential of fresh and stored moringa leaf extracts in improving seedling vigor, growth and productivity of wheat crop. *Environmental Science and Pollution Research* **24**: 27601-27612
16. Lopez-Amoros, M.L., Hernandez, T. and Estrella, I. (2006). Effects of germination on legume phenolic compounds and their antioxidant activity. *Journal of Food Composition and Analysis* **19**: 277-283
17. Maguire, J.D, (1962). Speed of germination aid in selection and evaluation for seedling emergence and vigour. *Crop Science* **2**: 176-177.
18. Maitra, S. and Shankar, T. (2019). Agronomic management in little millet (*Panicum sumatrense* L.) for enhancement of productivity and sustainability. *International Journal of Bio Resource Science* **6 (2):** 91-96. https://DOI. 10.30954/2347-9655.02.2019.9
19. Mariyappillai, A., Gurusamy, A., Swaminathan, C. and Duraisingh, R. (2022). Allelopathic chemical screening of aqueous extracts of coir pith on seedling growth of black gram and green gram. *Legume Research-An International Journal* **10**: 1-6. DOI:10.18805/LR-4703

20. Mohan, V., Unnikrishnan, R., Shobana, S., Malavika, M., Anjana, R.M. and Sudha, V. (2018). Are excess carbohydrates the main link to diabetes & its complications in Asians. *The Indian Journal of Medical Research* **148 (5):** 531. DOI: 10.4103/ijmr.IJMR_1698_18.
21. Nikishina, T.V., Popov, A.S., Kolomeitseva, G.L. and Golovkin, B.N. (2001). Effects of cryoconservation on seed germination of rare tropical Orchids. *Russian Journal of Plant Physiology* **48 (6)**: 810-815.
22. Nivethadevi, P., Swaminathan, C., Kannan, P. and Tamilselvi, E. (2022). Seed fortification and foliar spraying with moringa leaf extract enhances yield and yield attributes in blackgram. *Legume Research* **45(5)**: 352-356.
23. Panse, V.G. and Sukhatme, P.V. (1967). *Statistical Methods for Agricultural Workers*. Indian Council of Agricultural Research, New Delhi.
24. Richardson, D.R. and Williamson, G.B. (1988). Allelopathic effects of shrubs of sand pine scrub on pines and grasses of sand hills. *Forest Science* **34(3):** 592-605.
25. Senthilkumar, S.K., Deepa, T., Suganya, M., Janakarajan, J. and Muralidhar, V.P. (2016). Therapeutic properties of noni and its products. *International Journal of Science, Environment and Technology* **5**: 1496-1502
26. Stashenko, E. and Martínez, J. (2012). GC-MS analysis of volatile plant secondary metabolites. In: Gas chromatography in plant science, wine technology, toxicology and some specific applications. Dr. Bekir Salih (Ed.), InTech Europe, Crotia. 262-264. ISBN: 978-953-51-0127-7
27. Sangeetha, K., Swaminathan, C., Subramanian, E., Sivasankari, B. and Kannan, P. (2023). Influence of tree botanicals on seed germination and enzyme activity in blackgram (*Vigna mungo* L.) and cowpea (*Vigna unguiculat*a L.). *Allelopathy Journal* **59** (1): 49-67
28. Swaminathan, C. and Nandakumar, M.R. (2017). *Vrikshayurvedic farming. The Traditional Indian Agriculture*, Daya publishing House, New Delhi.
29. Swaminathan, C., Vinaya Rai, R.S. and Suresh, K.K. (1990). Allelopathic effects of *Parthenium hysterophorus* on germination and seedling growth of a few multipurpose trees and arable crops. *International Tree Crops Journal* **6** : 142-150
30. Wang, J., Vanga, S.K., Saxena, R., Orsat, V. and Raghavan, V. (2018). Effects of climate change on the yield of cereal crops: A review. *Climate* **6(2)**: 41. DOI: 10.3390/cli6020041
31. Yasmeen, A., Basra, S.M.A., Wahid, A., Nouman, W. and Hafeez-ur-Rehman. (2013). Exploring potential of moringa leaf extract as a seed priming agent in improving wheat performance. *Turkish Journal of Botany* **37**: 512-520.
32. Zhang, X. and Ervin, E.H. (2004). Cytokinin-containing seaweed and humic acid extracts associated with creeping bent grass leaf cytokinins and drought resistance. *Crop Science* **44**: 1737-1745.
33. Zulfiqar, F., Casadesus, A., Brockman, H. and Munne-Bosch, S. (2020). An overview of plant based natural biostimulants for sustainable horticulture with a particular focus on moringa leaf extracts. *Plant Science* **295**: 110194.