International Journal of Environment and Climate Change

Volume 13, Issue 12, Page 1017-1023, 2023; Article no.IJECC.111222
ISSN: 2581-8627
(Past name: British Journal of Environment & Climate Change, Past ISSN: 2231–4784)

Compatibility Analysis of Mushroom with different Botanicals in *In vitro* Condition

Rohit Kumar [a++*], Rinku Bhaskar [b++], Pankaj N. Mapari [a++], Shivani Datre [a++], Faijuddin Ahammad [a++], Prafull Patil [a++] and Sobita Simon [a#]

[a] Department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj (U.P.), 211007, India.
[b] Department of Plant Pathology, Acharya Narendra Deva University of Agriculture and Technology, Kumarganj, Ayodhya -224229, (U.P.), India.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJECC/2023/v13i103765

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/111222

Original Research Article

Received: 16/10/2023
Accepted: 22/12/2023
Published: 24/12/2023

ABSTRACT

White button mushroom [*Agaricus bisporus* (Lange) Imbach] is the most popular cultivated edible mushroom, fetching high price and still dominating in Indian and International market. However, the limiting factor for its successful cultivation is the occurrence of competitor moulds. The present investigations were carried out in the laboratory Department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, Uttar Pradesh. In this experiment different botanical (seed extract) *viz., Trachyspermum ammi* (Ajwain), *Foeniculum vulgare* (Saunf), *Anethum graveolens* (Soa), *Trigonella foenumgraecum* (Methi) were evaluated to test their

---

[++] M.Sc. Research Scholar;
[#] Professor;
**Corresponding author: E-mail: rohitksup01@gmail.com;*

compatibility with *Agaricus bisporus*. The botanicals (seed extract) were evaluated *in vitro* through poison food technique at 10, 20 and 30% concentrations and 24, 48 & 72 hours of incubation. The maximum radial growth of *Agaricus bisporus* was observed in T_0-Control (*Agaricus bisporus*) (16.45mm), (17.12 mm) and (17.22 mm) and minimum in T1 - *Trachyspermum ammi* (Ajwain) (1.46mm), (0.83mm) and (0.60mm) at 10, 20 and 30 percent concentration respectively. Maximum percentage of inhibition was observed in T1 - *Trachyspermum ammi* (Ajwain) (91.11%), (95.13%) and (96.50%) at 10, 20 and 30 percent concentration respectively.

Keywords: Agaricus bisporus; Anethum grveolens; Foeniculum vulgare; Trachyspermum ammi.

1. INTRODIUCTION

The white button mushroom (*Agaricus bisporus*) is very popular throughout the world and is the most important mushroom of commercial significance in India [1]. Mushrooms, classified in Phylum Basidiomycota, class Agaricomycetes, are revered as the "Flower of God" in India, known by diverse names. Thriving in varied agro-climatic conditions, these saprophytic fungi absorb nutrients from organic matter, flourishing in natural settings like decaying matter, leaves, droppings, and dead wood. [2]. A Frenchman achieved the initial success in commercial mushroom production in 1780 by cultivating *A. bisporus* underground in quarries near Paris [3]. Significant advancements in mushroom cultivation occurred in France around 1600 A.C., with the first cultivation of *Agaricus bisporus* on specially prepared agricultural media [4]. Compost, essential for growing white button mushrooms, results from breaking down organic waste by tiny organisms. Composting involves these organisms breaking down organic material, making protein, and preparing fibrous materials to hold water. Microbes not only change compost's properties but also limit the growth of other competing microbes. China initiated the first cultivation of mushrooms, and in India, commercial production began in hilly areas like Chail (Himachal Pradesh), Kashmir, and Ooty (Tamil Nadu) [5]. In the northern region, commercial cultivation primarily occurs during the winter season [1]. India has approximately 198,000 hectares under mushroom cultivation, yielding an annual production of 487,000 metric tons [6]. Among the total mushroom production in India, white button mushrooms constitute 73% (Sharma et. al. 2017). In the 2021-2022 period, Bihar leads in mushroom production, followed by Maharashtra, Orissa, Haryana, and Uttarakhand, as per the Agricultural and Processed Food Products Export Development Authority of India (APEDA), [7]. Fungal pathogens such as (Verticillium) Lecanicillium fungicola, *Mycogone perniciosa*, (Dactylium) Cladobotryum spp. and *Trichoderma spp*. afflict the cultivated mushroom *Agaricus bisporus* (Lange) Imbach, causing its most serious fungal diseases, dry and wet bubble, cobweb disease and green mould, respectively Potocnik et al. 2008a, [8,9,10]. 4 Several diseases of cultivated mushrooms are caused by bacteria and viruses (Grogan, 2008; Geels et. al. 2008). The most common bacterial disease, distributed worldwide, is the bacterial brown blotch caused by *Pseudomonas tolaasii* [11]. Various botanicals have proved useful source of fungitoxic substances that are rather harmless compared to synthetic chemical fungicides, which often impose undesirable side effects. Several plants have been reported to posses' substances, which are toxic to microbial pathogens and serve as protective barrier to infection. Keeping above in view, an effort was made to evaluate the efficacy of leaf extracts of angiospermic plants as compared to control of pathogenic fungi of mushrooms under field conditions.

2. MATERIALS AND METHODS

The present investigations were carried out in the laboratory, Department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, Uttar Pradesh (Year 2022-23) to test the compatibility of different treatments with *Agaricus bisporus* under *in vitro* conditions. In order of find out the compatibility of various plants extracts *viz., Trachyspermum ammi* (Ajwain), *Foeniculum vulgare* (Saunf), *Anethum garaveolens* (Soa), *Trigonella foenumgraecum* (Methi) with *Agaricus bisporus* were used. Seeds of all botanicals were collected and washed thoroughly in clean water. Equal number of washed seeds were grinded in mortar and pestle by adding same amount of sterilized water (1:1 w/v) and boiled at 80^0 C for 10 minutes in hot water bath. The extract was filtered by double layer muslin cloth followed by sterilized Whatman No.1 Filter paper [12]. Aqueous extract of 10, 20, 30 % was prepared according to the treatment by mixing 10, 20 and

30 ml of spices seed extract with 90, 80, 70 ml PDA respectively in separate conical flask. The flasks were thoroughly shaken to ensure an even mix of the extract under aseptic conditions. Twenty ml of sterilized melted PDA was aseptically poured in sterilized Petri dishes and allowed to solidify. After solidification of media 5mm disc of 7 days old subculture of *Agaricus bisporus* were placed in the centre of the Petri plates and one control plate which has only the PDA medium inoculated with culture disc and used as control. Each treatment and control were repeated three times to make three replications. Replicates were maintained for each test and those plates were incubated at 27 ± 1^0 C at incubator. The radial growth of mycelium was measured at different intervals of 24, 48 and 72 hrs. The radial growth of mycelium of each plate was measured by taking average of the two diameters taken right angles for each colony. Percent inhibition in growth was calculated in relation to growth in control using the following formula of Vincent (1947). The experiment was conducted in completely randomized block design (CRD) with three replications in each treatment. The conclusion was arrived at after statistical analysis of the data. The Completely Randomized Design (CRD) method recommended by Goon *et. al*. was used to conduct the statistical analysis of laboratory experiments (1931). The variance ratio test at the 5% level of probability was used to determine the significance of treatment differences. The observation of per cent inhibition of mycelial growth, were transformed in to "Arc sin Transformation" = $\sin^{-1}\sqrt{p}/100$ used for statistical analysis.

Mycelial inhibition = (Radial growth in control – Radial growth in treatment) / (Radial growth in control) ×100

3. RESULTS AND DISCUSSION

3.1 *In vitro* effect of botanicals (seed extract) on mycelial growth and per-cent inhibition of *Agaricus bisporus* at 10 % concentration at 24hrs, 48hrs and 72hrs

As shown in Table 2 and depicted in Fig. 1 reveals that at 10% concentration, after 24hrs, 48hrs and 72hrs incubation, the maximum radial growth of *Agaricus bisporus* was observed in T_0-Control (*Agaricus bisporus)* (16.45mm) followed by T_2- *Foeniculum vulgare* (Saunf) (4.90mm), T_4 - *Trigonella foenumgraecum* (Methi) (2.11mm) T_3- *Anethum grveolens (Soa)* (1.95mm), and T_1 **–** *Trachyspermum ammi* (Ajwain) (1.46 mm). The treatment T_0- *Agaricus bisporus* (Alone) was significant over all the treatments. The result showed that maximum percentage of inhibition was observed in T_1 **–** *Trachyspermum ammi* (Ajwain) (91.12%) followed by T_3- *Anethum grveolens (Soa)* (88.17%), T_4 - *Trigonella foenumgraecum* (Methi) (88.15%), T_2-*Foeniculum vulgar* (Saunf) (70.21%) and T_0-Control (*Agaricus bisporus*) Table 3.

3.2 *In vitro* effect of botanical (seed extract) on mycelial growth and per-cent inhibition of *Agaricus bisporus* at 20 % concentration at 24 hrs, 48 hrs and 72 hrs

As shown in Table 2 and depicted in Fig.1 reveals that at 20% concentration, after 24hrs, 48hrs and 72hrs incubation, the maximum radial growth of *Agaricus bisporus* was observed in T_0-Control (*Agaricus bisporus*) (Alone) (17.12 mm) followed by T_2- *Foeniculum vulgare* (Saunf) (3.54 mm), T_4 - *Trigonella foenumgraecum* (Methi) (1.17 mm), T_3- *Anethum grveolens (Soa)* (0.96 mm), and T_1 **–** *Trachyspermum ammi* (Ajwain) (0.83 mm). The treatment T_0- *Agaricus bisporus* (Alone) was significant over all the treatments. The result showed that maximum percentage of inhibition was observed in T_1 **–** *Trachyspermum ammi* (Ajwain) (96.04%) followed by T_3- *Anethum grveolens (Soa)* (95.55%), T_4 - *Trigonella foenumgraecum* (Methi) (93.63%), T_2- *Foeniculum vulgare* (Saunf) (80.12%) and T_0- Control (*Agaricus bisporus*) (0.00 %). Table 3.

3.3 *In vitro* effect of botanical (seed extract) on mycelial growth and per-cent inhibition of *Agaricus bisporus* at 30% concentration at 24 hrs, 48 hrs and 72 hrs

As shown in Table 2 and depicted in Fig. 1 reveals that at 30% concentration, after 24 hrs, 48 hrs and 72 hrs incubation, the maximum radial growth of *Agaricus bisporus* was observed in T_0-Control (*Agaricus bisporus*) (17.22 mm) followed by T_2- *Foeniculum vulgare* (Saunf) (2.56 mm), T_4 - *Trigonella foenumgraecum* (Methi) (0.84 mm), T_3- *Anethum grveolens (Soa)* (00.60 mm), and T_1 **–** *Trachyspermum ammi* (Ajwain) (0.50 mm). The treatment T_0- *Agaricus bisporus* (Alone) was significant over all the treatments. The result showed that maximum percentage of inhibition was observed in T_1 **–** *Trachyspermum ammi* (Ajwain) (97.10%) followed by T_3- *Anethum*

Table 1. List of botanicals and their scientific names

Serial. No.	Common name	Botanical name	Plant part used
1	Ajwain	*Trachyspermum ammi*	Seed
2	Saunf	*Foeniculum vulgare*	Seed
3	Soa	*Anethum grveolens*	Seed
4	Methi	*Trigonella foenumgraecum*	Seed

Table 2. Mycelial growths of *Agaricus bisporus* at 10 %, 20 % and 30 % concentration along with 24, 48 and 72 hours of incubation

	Mycelial growth (mm)								
	10 %			20 %			30 %		
	24 Hours	48 Hours	72 Hours	24 Hours	48 Hours	72 Hours	24 Hours	48 Hours	72 Hours
Agaricus bisporus (Alone)	5.92	10.72	16.45	6.35	10.98	17.12	6.12	10.87	17.22
Trachyspermum ammi (Ajwain)	0.7	1.08	1.46	0.49	0.63	0.83	0.32	0.47	0.50
Foeniculum vulgare (Saunf)	1.96	3.38	4.9	1.58	2.48	3.54	1.03	1.72	2.56
Anethum grveolens (Soa)	0.93	1.59	1.95	0.56	0.67	0.96	0.34	0.50	0.60
Trigonella foenumgraecum (Methi)	0.93	1.58	2.11	0.51	0.8	1.17	0.42	0.63	0.84
F- test	S	S	S	S	S	S	S	S	S
S. Em	0.010	0.007	0.011	0.013	0.008	0.013	0.012	0.009	0.012
CD (0.05)	0.033	0.023	0.035	0.040	0.026	0.041	0.039	0.027	0.040

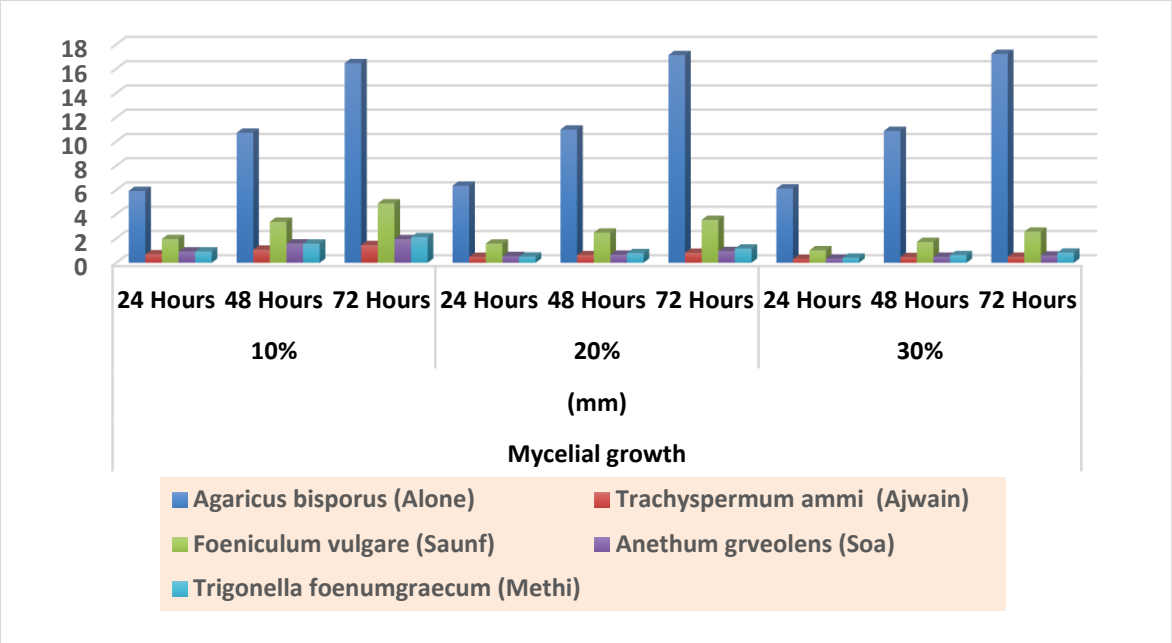

Fig. 1. Mycelial growth of *Agaricus bisporus* at 10 %, 20 % and 30 % concentration along with 24, 48 and 72 hours of incubation

grveolens (Soa) (96.52%), T_4 - *Trigonella foenumgraecum* (Methi) (95.12%), T_2- *Foeniculum vulgare* (Saunf) (85.13%) and T_0- *Agaricus bisporus* (0.00 %). Table 3.

Similar findings of botanicals, and fungicides on mycelial growth and percent inhibition were reported by Jahan et. al., Singh et. al. [13], Sailja and Radhika.

Table 3. Mycelial growth inhibition of *Agaricus bisporus* at 10 %, 20 % and 30 % concentration along with 24, 48 and 72 hours of incubation

Treatments	Per-cent inhibition of mycelial growth (mm)								
	10%			20%			30%		
	24 Hours	48 Hours	72 Hours	24 Hours	48 Hours	72 Hours	24 Hours	48 Hours	72 Hours
Agaricus bisporus (Alone)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Trachyspermum ammi (Ajwain)	88.18 (69.86)	89.93 (71.46)	91.12 (72.63)	92.28 (73.84)	94.26 (76.11)	96.04 (78.49)	94.77(76.75)	95.40 (77.58)	97.10(80.15)
Foeniculum vulgare (Saunf)	66.89 (54.84)	68.47 (55.81)	70.21(56.90)	75.12 (60.05)	77.41 (61.59)	80.12 (63.50)	83.17(65.75)	84.18 (66.53)	85.13(67.29)
Anethum grveolens (Soa)	84.29 (66.62)	85.26 (67.39)	88.17 (68.98)	91.18 (72.69)	93.90 (75.66)	95.55 (77.79)	94.44(76.34)	95.68 (77.96)	96.52(79.21)
Trigonella foenumgraecum (Methi)	84.29 (66.62)	85.17 (67.32)	88.15 (69.83)	92.07 (73.61)	92.71(74.31)	93.63 (75.35)	93.14(74.78)	94.20 (76.04)	95.12(79.21)
C.D	0.35	0.14	0.14	0.54	0.22	0.96	0.71	0.17	0.32

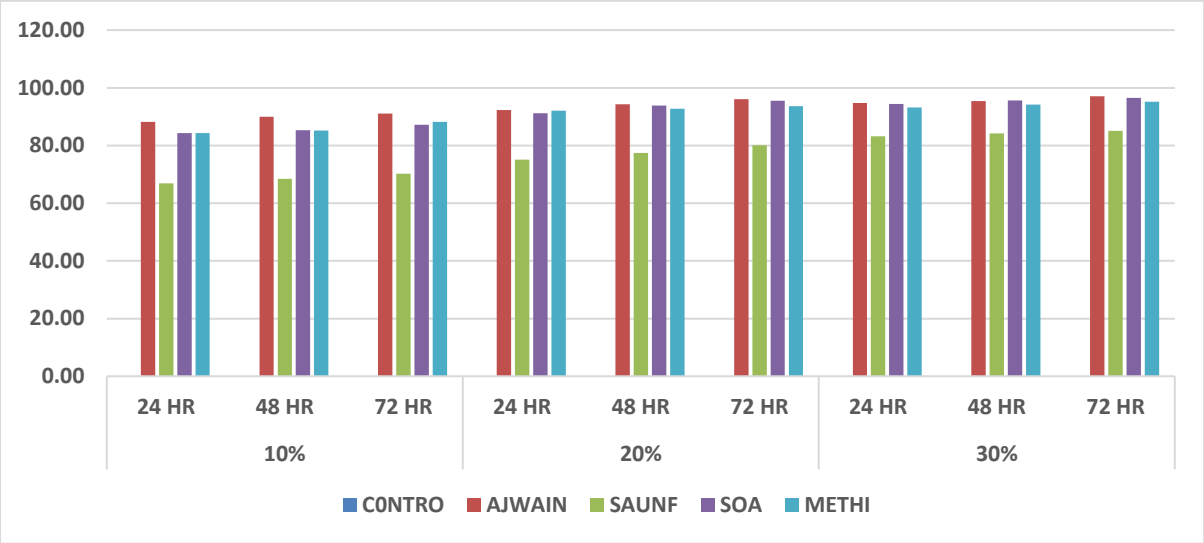

Fig. 2. Mycelial growth inhibition of *Agaricus bisporus* at 10 %, 20 % and 30 % concentration along with 24, 48 and 72 hours of incubation

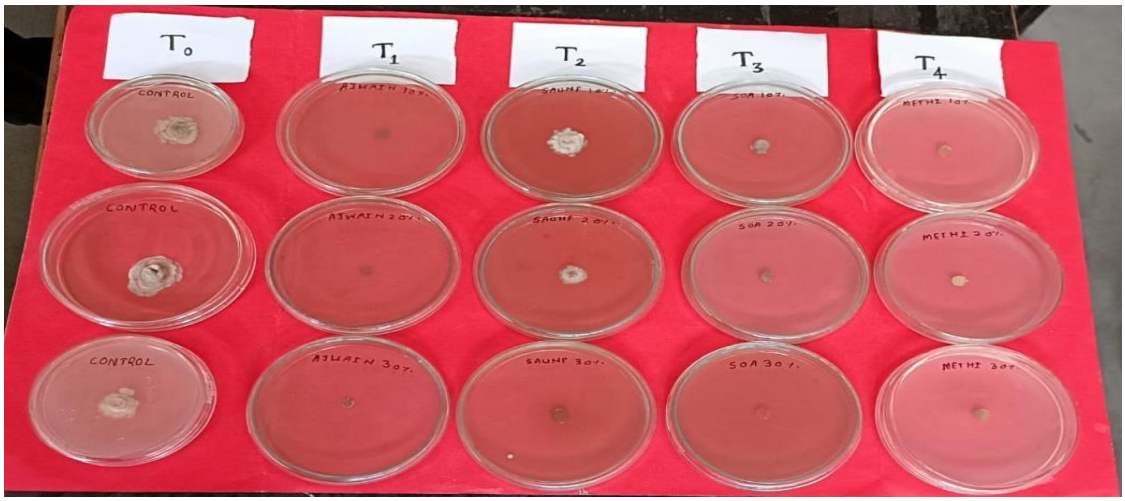

Fig. 3. Response of botanicals against *Agaricus bisporus* on mycelial growth

4. CONCLUSION

Among the tested botanicals, it was found that all the botanical more or less inhibited the growth of *Agaricus bisporus* as compared to growth in control-T_0 [*Agaricus bisporus* (Alone)]. With the increase in concentration there was increase in the inhibition of mycelial growth of *Agaricus bisporus.* The results revealed that botanical seed extracts were found incompatible with *Agaricus bisporus* under *in vitro* condition.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERECES

1. Maheshwari S. A Guide for White Button Mushroom (*Agaricus bisporus*) Production. Open Access Scientific Reports. 2013;(3):2-3.
2. Kumar K, Biswas SK, Hussain A, Baboo D. Kumar S. Influence of different casing material on growth parameters and yield of white button mushroom (*Agaricus bisporus),* (Longe. Imboch). Journal of Pharmacognosy and Phytochemistry. 2020;(1):902-905.
3. Beyer DM. Basic procedures for Agaricus mushroom growing. Penn State College of Agricultural Sciences Research,

Extension, and Resident Education Programs, Pennsylvania, Cheste. 2003; (69):254-259.
4. Chang ST, Miles PG. Mushrooms cultivation, nutritional value, medicinal effect and environmental impact, CRC Press. 2004;(2):22-25.
5. Jain SK, Mehta BK, Sharma GP, Doshi A, Jain HK. Cultivation of button mushroom and its processing: a techno-economic feasibility. International Journal of Advanced Biotechnology and Research. 2011;(1):201-207.
6. Anonymous. Horticulture statistics at a glance. National Horticulture Board Database; 2017.
Available:https://www.nhb.gov.in.
7. Agricultural and processed food products export development authority of India (APEDA); 2021.
Available:www.apeda.in
8. Potocnik I, Vukojevic J, Stajic M, Tanovic B, Todorovic B. Fungicide sensitivity of selected *Verticillium fungicola* isolates from *Agaricus bisporus* farms. Archives of Biological Sciences. 2008b;(60):151-158.
9. Potocnik I, Vukojevic J, Stajic M., Rekanovic E, Stepanovic M, Milijasevic S, Todorovic B. Toxicity of biofungicide Timorex 66 EC to *Cladobotryum dendroides* and *Agaricus bisporus*. Crop Protection. 2010a;(29):290-294.
10. Potocnik I, Vukojevic J, Stajic M, Tanovic B, Rekanovic E. Sensitivity of *Mycogone perniciosa*, pathogen of culinary medicinal button mushroom *Agaricus bisporus* (J. Lange) Imbach (Agaricomycetideae) to selected fungicides and essential oils. International Journal of Medicinal Mushrooms. 2010b;(12):91-98.
11. Todorovic B, Milijasevic-Marcic S, Potocnik I, Stepanovic M, Rekanovic E, Nikolic-Bujanovic LJ, Cekerevac M. *In vitro* activity of antimicrobial agents against *Pseudomonas tolaasii*, pathogen of cultivated button mushroom. Journal of Environmental Science and Health, Part B: Pesticides, Food Contaminants and Agricultural Wastes. 2012;(47):175-179.
12. Bhaskar R, Kumar R, Singh J, Kumar R, Chandra S, Chand R, Vimal SC. *In vitro* Evaluation of Botanicals, Bio-agents and Fungicides against *Rhizoctonia solani* Causing Web Blight Disease of Mungbean. Int. J. Environ. Clim. Change. 2023;13(11):3546-3552.
13. Singh S, Lal AA, Simon S. Efficacy of some botanicals in controlling dry bubble (*Verticillium fungicola)* disease in white button mushroom (*Agaricus bisporus*) cultivation. An International Quarterly Journal of Life Sciences. 2017;(4):2969-2973.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/111222