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RESEARCH ARTICLE

PROSPECTIVE OF AGRICULTURAL WASTES AS BASE RESOURCES FOR MASS MULTIPLICATION OF *TRICHDERMA* SPECIES WORLDWIDE: AN OVERVIEW

*Bhale, U. N.

Department of Botany, Arts, science and Commerce College Naldurg, Tq. Tuljapur Dist. Osmanabad 413602 (M.S.)

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ABSTRACT

The environmental infectivity caused by extreme use of chemical pesticides increased the interests in integrated pest management (IPM), where chemical pesticides are substituted by bio-pesticides to control plant pests and plant diseases. Genus *Trichoderma* has high ability to inhibit the growth of pathogenic fungus by secreting some enzymes. *Trichoderma* is an asexually reproducing filamentous fungus and a species aggregate. An effective biological control agent should be genetically stable, effective at low concentrations, easy to mass produce in culture on inexpensive media and be effective against a wide range of pathogens. The mechanisms of *Trichoderma* include direct competition with the target organism, antibiosis and parasitism of the target organism and induce resistance of the host plant. Therefore various agricultural by products were evaluated for mass production of *Trichoderma* species worldwide. *Trichoderma haarzianum* and *T. viride* was found suitable carrier materials to formulate various agro wastes. Biological control of plant pathogens through antagonists is a potential, ecofriendly and a sustainable approach apart them being a promising alternative to the use of chemicals. The major issue involved in mass production and utilization of biocontrol agents are selection of effective strains, development cost effective methods, for mass multiplication, effective methods for storage, consignment and its formulation. Abiotic factors also have a profound effect on the production and activities of enzymes and antibiotics associated with biocontrol by *Trichoderma* species.

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INTRODUCTION

Trichoderma have often been used in the management of crop plant diseases. *Trichoderma* is a genus of asexually reproducing fungi that is present in all types of soils. *Trichoderma* species have been recognized as antagonists of soil-borne and foliage pathogens and as efficient decomposers of cellulosic waste materials**.** *Trichoderma* is an antagonistic free living fungus found in the soil. Desirable quality of their properties such as quick growth and multiplication, antagonism, growth promotion, competition with other microbes, hyperparasitsm etc., these suppress the pathogenic fungi and protect the plants from diseases. *Trichoderma* is widely used as bio-control agent against several root pathogenic fungi throughout the world (Chet *et al*., 1979) Elad *et al*. (1980), Sivan *et al*. (1984).

**Corresponding author: Bhale, U. N.*
Department of Botany, Arts, science and Commerce College Naldurg, Tq. Tuljapur. Dist. Osmanabad 413602 (M.S.)

Trichoderma spp. is free living fungi that are highly interactive in root, soil foliar and environments. In disease management, the increased use of chemicals have caused negative impact on environmental quality and resulted in grow of many living forms which are resistant to the chemicals. A variety of alternative approaches have also been considered which include the use of biocontrol agents. *Trichoderma* spp. is biocontrol agents widely used in management of fungal diseases of crop plants exhibiting mycoparasitism against a wide range of plant pathogens. *Trichoderma* spp is a potential fungal biocontrol agent against a range of plant pathogens. Biological control is nature friendly approach that uses specific micro-organisms, which hamper with plant protection. Biological control by an antagonism is a potential, no chemical and eco-friendly approach for managing plant diseases (Bailey, 2004). Indiscriminate use of fungicide, pesticides has resulted in emergence of resistant pathogen strains overtime and has increased the environmental pollution level in soil and water, and adverse effect on food quality and human health. Thus now day's *Trichoderma* is most consistent to increase the population for using every crop fields. Therefore this review article is introduced the current status and mass multiplication of *Trichoderma* as a biocontrol agent on agro waste materials.

Methodological approach

Different sources viz. rhizospheric soil of irrigated and non irrigated plants, crop seeds, grains, fruit rinds air etc were used for isolation of *Trichoderma* species. The isolation of *Trichoderma* species was done by dilution plate method (Waksman, 1922; Papavizas and Lumsden, 1982) and purified by *Trichoderma* selective medium (TSM) (Elad and Chet, 1983). The *Trichoderma* species were identified by determining their colony characters with the help of manuals (Gams and Bisset,1998). Spore suspension of *Trichoderma* species from 7 days old culture was prepared. One ml spore suspension was inoculated in presterilized and cool polythene bags containing agrowaste materials, by disposable syringe. Initial weight of bags was noted. After the incubation period at room temperature for 12 days, the final weight was taken and biomass of *Trichoderma* species was obtained. *Trichoderma* species spores count on each base materials were determined by using haemocytometer (Singh and Singh, 2007; Valdez and Picolo, 2007; Vijayan *et al.*, 2002).

Control mechanism

Trichoderma-based biocontrol agents possess better ability to promote plant growth and soil remediation activity compared to their counterparts (virus, bacteria, nematodes and protozoa etc.). *Trichoderma* spp. is acclaimed as effective, ecofriendly and cheap, nullifying the ill effects of chemicals. Therefore, of late, these biocontrol agents are identified to act against on array of important soil borne plant pathogens causing serious diseases of crops. Therefore considering the cost of chemical pesticides and hazardous involves, biological control of plant diseases appears to be an effective and ecofriendly approach being practice world over. Different biological control agents (BCAs) can be used for the control of diseases. These include bacteria, fungi and actinomycetes. Weindling (1932) ascribed biocontrol by *T. lignorum* of citrus seedling disease, incited by *Rhizoctonia solani*, to mycoparasitism. Weindling described in detail the mycoparasitism of *R. solani* hyphae by the hyphae of the biocontrol agent, including coiling around pathogen hyphae, penetration, and subsequent dissolution of the host cytoplasm. There are four different mechanisms by which BCAs control other microorganisms. The genus *Trichoderma* comprises a large number of species some of which act as biological control agents through one or more mechanisms. These mechanisms include competition for space and nutrients (Elad, 1999). *Trichoderma* species are generally considered to be aggressive competitors, grow very fast and rapidly colonize substrates to exclude pathogens such as *Fusarium* spp. (Papavizas, 1985). Rhizosphere competence, following seed treatment is an important strategy to create a zone of protection against pathogens (Howell, 2003). Mycoparasitism role of *Trichoderma* grows tropically toward hyphae of other fungi, coil around them in a lectin mediated reaction, and degrade cell walls of the target fungi by the secretion of different lytic enzymes. The antagonist attached to the pathogen and secreted glucanase and chitinase enzymes that act through the cell wall (Elad, 1983; Haran, 1996; Lorito, 1996), production of inhibitory compounds (Sivasithamparam and Ghisalberti, 1998). Induced resistance of specific strains of fungi in the genus *Trichoderma* colonize and penetrate plant root tissues and initiate a series of morphological and biochemical changes in the plant, considered to be part of the plant defense response, which subsequently leads to induced systemic resistance (Bailey and Lumsden, 1998;Kapulnik and Chet, 2000). *Trichoderma* strains exert control against fungal phytopathogens either indirectly by competing for nutrients and space, modifying the environmental condition, promoting plant growth, plant defensive mechanisms and antibiosis, or directly by mechanisms such as mycoparasitism. Activation of each mechanism implies the production of specific metabolites, such as plant growth factors, hydrolytic enzymes, siderophores, antibiotics, and permeases. The application of *Trichoderma* species can control a large number of foliar and soil borne fungi i.e. *Fusarium* spp*., R. solani, Pythium* spp*., S. sclerotium*, *S. rolfsii*, in vegetables, field, fruit and industrial crops (Tran, 1998; Ngo *et al*., 2006). *Trichoderma* .spp. was demonstrated to be very efficient producers of extracellular enzymes with cellulases as the first example. Later, these fungi were found to produce a wide range of other extracellular enzymes and some of these were implicated in the biological control of plant diseases (Elad *et al*., 1982). Once *Trichoderma* hyphae penetrate the roots, a series of bioactive metabolites from the fungus is produced that induces walling off and biochemical mechanisms that limit growth of the *Trichoderma* to a small area. This reaction may not always occur; for example, there now are known endophytic *Trichoderma* strains that colonize vascular systems of certain plants.

Control strategies include biological control, breeding of resistant varieties, improvement in cultural practices, storage conditions to those less favorable for pathogen attack and survival, application of chemical fungicides and using integrated pest management (IPM). *Trichoderma harzianum* is the most effective strains among the various species of the *Trichoderma* that is used for biocontrol mechanisms (Gao *et al*., 2002). The loss can occur in the field or in the store and at any time between sowing and consumption of the harvest. Over 30000 plant diseases are recorded from different countries, of which about 5000 are present in India (Rangaswamy, 1988). *Trichoderma* spp. Is a potential biocontrol agent for the management of various plant diseases like, sapota (Wagh and Bhale, 2011; Bhale, 2013), Leafy vegetables (Rajkonda *et al*., 2012), spinach (Bhale, 2012), brassicas (Cheah and Marshall, 1995).

Strain improvement

In the nature, numbers of factors as well as chemical compounds are mutagenic agents. They caused their effects on microorganisms. Their effects may be found to be harmful as well as significant up to some extent. Many mutagenic agents showed their effects resulting the improvement of the resistant microorganisms. Mutation is one of the important strategies for strain improvement programme of bicontrol agents. Papavizas *et al*. (1882) studied UV induced mutants of *T. harzianum* tolerant to benomyl fungicides and suppressed the growth of *Trichderma* have been developed. Earlier workers made their efforts on strain improvement of *Trichoderma* species by the treatment of mutagenic agents. Combined treatment of ultra-violet radiation and sodium nitrate (0.5mg/ml.) for 45 minutes to *Trichoderma* species (*T. reesei* QM9414) was resulted in hyperproduction of cellulase up to 1.5-1.75fold (Gadgil *et al*., 1995). Strains of *Trichoderma* species such as *T. harzianum, T. koningii, T. pseudokoningii* and *T. viride* were mutated for tolerance to the fungicide Benomyl. These species were not found to be tolerant to the Benomyl but *Trichoderma* species were induced by mutation to increase their linear growth rate (Ahmad and Baker, 1988). Chand *et al*. (2005) employed two methods for mutation, first in which germinating spore suspension was treated with 0.1 and 0.2mg/ml of 1-methyl-3-nitro-1-nitrosoguanidine (MNNG) and Ethydium bromide(EtBr) and uv rays for 30 min. and 1 hour duration and second in which the same mutagens were incorporated in the selective media in sublethal concentration (5µg/ml). They concluded that mutation using sublethal concentrations of mutagens for a prolonged period of growth had yielded mutants which can produce more cellulase. Mutant strains of *T. viride* those not producing cellulase were induced and found that mutant strains lack the ability to hydrolyze both crystalline and soluble cellulose (Nevalainen and Palva, 1978). Four mutant strains from *T. viride* were prepared by UV irradiation of conidia (Farkas *et al*., 1981). Some mutants showed morphological differences with the parent strain. The mutants produced 2 to 3 times higher levels of β- glucosidase than the parent strain. From the mutant strains of *T. pseudokoningii*, secretion of 3.3 times the extracellular protein and 3 times the endoglucanase was reported (Zaldivar *et al*., 1987). A wild strain of *Trichoderma viride* was subjected to successive mutagenic treatment with UV irradiation, low energy ion beam implantation, Atmospheric Pressure Non-Equilibrium Discharge Plasma (APNEDP) and N-methyl-N-nitro-Nnitrosoguanidine to generate about 3000 mutants (Xu *et al*., 2011). Among these mutants *T. viride* N879 exhibited the greatest activity like 2.38 fold filter paper activity, 2.61 fold carboxy methyl cellulase, 2.18 fold β-glucosidase and 2.27 fold cellobiohydrolase activities. Abou Sereih *et al*. (2007) used chitosan in five concentrations such as 0.38, 0.75, 1.50, 3.00 and 4.50 µg/ml for study of its effect on growth of *T. harzianum*. However, two doses 3.00 and 4.50 µg/ml resulted in changing the growth colour from green to yellow. It was reported that chitosan in the media enhanced the antagonistic properties of *T. harzianum* against *Fusarium oxysporum* f. sp. *sesami*. Gamma ray exposure was found to be induced mutation in *T. harzianum* resulted two stable salt tolerant mutants (Mohamed and Haggag, 2006). UV-irradiated strain of *T. harzianum* in pellet form comparable to the chemical fungicide Mancozeb against damping-off of vegetables was applied 2 weeks before planting in a farmers' field in Laguna province, Philippines (Cuevas *et al.*, 2005).

Biomass production on agricultural wastes

Techniques for mass multiplication of *Trichoderma* have been developed which are cheap, simple, easy and less time consuming. Commercial and practical success of mass multiplication of a biocontrol agent depends on its efficacy, self life, eco-friendly and its mass production capacity on suitable and easily available substrates. The efforts of mass multiplication were made since 30-40 years. Several substances have been used as substrates including agro waste materials, fruit wastes and industrial byproducts for mass multiplication of *Trichoderma* species. Clay impregnated with 10 percent molasses was found to be maximum production of *Trichoderma* spores (Blackman and Kabana, 1975). Similarly yeast medium containing molasses were found to be very good for mass multiplication of *Trichoderma* (Papavizas *et al*., 1984; Prasad and Rangeshwaran, 2000). The molasses & jaggery broth also significant for substrate for mass production of *Trichoderma* (Sawant and Sawant, 1996). Many workers were tried the various agrowaste materials such as bran of cereal crops, husks of pulse crops, fruit rinds, saw dust etc. as a medium for mass multiplication and viability of biocontrol agenmt *Trichoderma* species worldwide (Table 1). Among the agrowaste materials, wheat bran was found as effective medium for mass multiplication of *T. harzianum* (Heins *et al*., 1978; Martin *et al*., 1984). Similarly wheat bran and saw dust combination proved to be good substrate for *Trichoderma* mass multiplication (Elad *et al*., 1980; Mukhopadhyay *et al*., 1986). Combination of wheat bran with maize bran also showed comparative effects for *Trichoderma* mass production and found to be good medium (Kapoor and Kumar, 2004). Similarly pulse bran with saw dust was found to be better than the wheat bran as a substrate for mass production (Dubey and Patel, 2002). Use of corn as a substrate for *Trichoderma* mass production has been tested by many pathologists (Lewis and Papavizas, 1980). Barley was also used as a medium by Abd El Moity and Shatala (1981). Similarly sorghum was tested as a substrate for mass production of *Trichoderma* species (Padmanabhan and Alexander, 1984; Upadhyay and Mukhopadhyay, 1986). Among the agowaste materials, many workers reported that sugarcane bagasse, rice straw and groundnut shell also a good substrate. Dubey and Patel (2002) used wheat straw, groundnut shells, and mushroom bed straw. Similarly decomposed coffee pulp (Sangle *et al*., 2002), coffee waste (Saju *et al*., 2002), agroindustrial cellulosic waste (Tewari *et al*., 2004). It was suggested that the byproducts of sugar industry was the best media for mass multiplication of *T. harzianum* (Mev and Meena, 2003).

Other agrowaste materials used as asubstrate for mass production of *Trichoderma* species were straws (Davet *et al*., 1981), Topioca (Kousalya and Jeyarajan, 1988), FYM – organic wastes (Jacob and Sivaprakasam, 1993) and vermiculite (Lewis *et al*., 1991). For the mass multiplication of *Trichoderma* species a biolocontrol agent, the role of environmental factors are important along with substrates. Temperatureis one of the most important factors caused effects on mass multiplication of *Trichoderma* species. The species of *Trichoderma* grow maximum at 25±2°C while poor growth at 10±2°C and 35±2°C was recorded (Bhatnagar, 1996). *Trichoderma* species have been grown on wide range of grains viz. maize,sorghum, pearl millet, wheat, wheat bran, waste tea leaves, banana fruit rinds,paddy straw (Zaidi and Singh, 2004). Press mud is good substrate and its composition suits well for developing good compostby *Trichoderma* (Singh and Joshi, 2007). Press mud helps in establishment of *Trichoderma* in soil and provides protection against different diseases. *Trichoderma harzianum* is reported to multiply well on press mud under laboratory and compost pits at farmer fields.

Table 1. Mass production of the *Trichoderma* species using various agro base materials worldwide

S.No	Agro waste materials	*Trichoderma* species	References
1	Black gram shell,shelled maize cob, coir pith, peat,gypsum, barley grains.	*T. viride,T. harzianum*	Kumar and Marimuthu, 1997.
2	Coffee fruit skin + biogas slurry	*T. harzianum*	Sawant and Sawant, 1996.
3	Coffee husk	*T. harzianum,T. viride, T. virens.*	Bhai et al., 1998.
4	Coffee berry husk	*T. harzianum,T. viride, T. virens*	Sawant and Sawant, 1989
5	Fruit skin and berry mucilage	*T. harzianum,T. viride, T. virens*	Sawant and Sawant, 1989
6	Soil	*T. viride,T, harzianum*	Singh, 2002
7	Mustard oil cake	*T. viride.*	Singh, 2002
8	Sorghum grains	*T harzianum,T. virens*	Singh, 2002.
9	Sugarcane straw	*T. viride,T. harzianum, T. koningii,*	Upadhyay and Mukhopadhyay, 1986; Mishra, 1998
10	Wheat bran	*T. reesei*	Singh et al., 2004
11	Rice husk, maize cob powder,	*T. virens*	Singh et al., 2002
12	tea leaves, wheat bran, citrus fruit pulp	*T. harzianum* (MTCC-3843)	Tripathi, 1998.
13	Saw dust, Rice bran, Coir pith, Sorghum grains, Coir pith + Neem cake, Cow dung + neem cake	*T. harzianum T. viride*	Rini and Sulochana,2007
14	Carrot peel, Cucumber peel, Potato peel, Cabbage waste, Pea husk, Sugarecane waste, Orange peel, Papaya Peel	*T. harzianum, T. viride*	Simon andAnamika,2011
15	Vegetable waste, Fruit juice waste, Sugarcane baggase, Rotten wheat grains	*T. viride*	Chaudhari et al.,2011
16	Sorghum grains, Pearlmillet grains,Groundnut cake, mushroom byproduct	*T. harzianum*	Sharma and Sain,2006
17	Sugarcane baggase, Waste tea powder, Wood chips, Maize bran + saw dust	*T. viride,*	Rajkonda,2012
18	Maize bran + saw dust ,Sugarcane baggase, Waste tea powder, Moth seeds, Gram seeds	*T.harzianum*	Rajkonda,2012
19	Rice bran + saw dust, Maize bran, Wood chips, Pigeon pea bran + saw dust, Sugarcane baggase, Waste tea powder	*T koningii*	Rajkonda,2012
20	Wood chips, Maize bran + saw dust	*T. pseudokoningii*	Rajkonda,2012
21	Gram husk, Sugarcane baggase, Maize bran	*T. virens*	Rajkonda,2012
22	Rice husk,wheat straw+ Rice husk,wheat straw+readgram straw	*T.viride*	Patale,2005
23	Sorghum grain	*T. harzianum*	Upadhyay and Mukhopadhyay,1986
24	Wheat bran-saw dust modified medium	*T. harzianum*	Mukhopadhyay *et al*., 1986
25	Tapioca rind, Tapioca refuse, FYM, press mud		Kousalya Gangadharan & Jeyarajan, 1990
26	FYM, wheat bran, rice bran, peat soil, rice straw	*Trichoderma* spp	Sangeetha Panicker *et al*., 1993
27	Groundnut shell medium	*Trichoderma* spp	Raguchander *et al*., 1993
28	Spent tea leaf waste and coffee husk	*Trichoderma* spp	Bhai *et al*., 1994
29	Wheat bran and biogas manure	*T. harzianum*	Jagadeesh & Geetha ,1994
30	Pigeonpea husk, tapioca waste (after starch extraction) and press mud.	*T. harzianum*	Jayaraj & Ramabadran, 1996
31	Decomposed Coconut Coir pith	*T.viride*	Kumar & Marimuthu, 1997
32	Spent malt	*Trichoderma* spp	Gopalakrishnan *et al.* 2003
33	Rice barn	*T. harzianum*	Tiwari and Bhanum,2004
34	Tea waste	*T. harzianum, T. virens*	Prakash,1999
35	Sugarcane baggasse, Maize bran, Wood chips, Maize bran + saw dust,	*T. viride, T. harzianum,T. koningii, T. pseudokoningii, T. virens*	Rajkonda and Bhale,2012
36	Sugarcane baggase, Vegetable waste	*Trichoderma candidum*	Babu and Pallavi,2013
37	Rice husk + sorghum, Saw dust + sorghum Rice bran + sorghum	*Trichoderma viride*	Pandey,2009
38	Pulses	*T. viride*	Khandelwall et al.,2012
39	Vegetable wastes, fruit wastes, crop wastes, FYM and Poultry manure	*T. viride* and *T.harzianum*	Siman and Anamika,2011
40	Rice husk, saw dust, maize husk and wheat bran	*T. viride and T. harzianum*	Yadav,2012
41	Sugarcane baggase followed by vemicompost, talcum powder and paddy straw	*T. harzianum*	Subash et al.,2014
42	Spent mushroom compost, Vermicompost ,farmyard manure, wheat grain, sorghum grain, broken maize grain	*T. harzianum*	Singh et al.,2014
43	Spent mushroom compost	*Trichoderma longibrachiatum, T. harzianum, T. viride*	Kaviyarasan and Siva (2007)

Continue………….

44	Wheat straw, tea wastes, vegetable wastes, coffee husk and barley bran	*Trichoderma* spp	Mamo and Alemu,2012
45	Corn fiber dry mass, sewage sludge compost	*T. viride*	Taweil et al., 2009
46	Sawdust, paddy straw	*T. harzianum*	Kumar and Palakshappa, 2009
47	cow dung, rice bran	*Trichoderma* spp	Rini and Sulochana, 2007
48	Vegetable waste, fruit juice waste and rotten wheat	*Trichoderma viride*	Chaudhari et al., 2010
49	Cowdung, banana leaf, arecanut leaf, coconut leaf, neem cake,vermicompost, rice barn and rice straw	*T. viride*	Chakrabarty et al., 2014
50	Sugarcane bagasse ,Apple peel , Mustard,Wheat , Coriander ,Dry leaves	*T. viride and T. harzianum*	Gautam and Gupta,2014
51	Molasses yeast medium, broken sorghum grains, whole sorghum grains and broken maize grains	*T. harzianum*	Pramod Kumar and Palakshappa,2009
52	Sugarcane baggase, fruit juice waste, vegetable waste, rotten wheat grains	*T. viride*	Esposito, da Silva, (1998)
53	Broken corn seed medium, sugar beet molasses medium, Tragacanth gum, Maltodekstrin, whey powder	*T.harzianum (1211), T.virense (9011) T.atroviride (6022), T.koningii (iso2)*	Panahian et al.,2012
54	Biowastes of mango, carrot, papaya, banana and chukandar	*T. harzianum*	Mucksood et al.,2013
55	Molasses,tea waste, paddy husk, coconut poonac & poultry manure	*T. koningii*	Nishantha et al.,2001
56	Pigeonpea leaves, and urd bean straw, wheat straw and saw dust	*T.harzianum*	Prajapati, et al.,2012
57	Tea waste, wheat barn, sorghum straw, cotton seed and cotton cake	*T.harzianum T. viride,T.virens*	Sharma and Trivedi,2005
58	Orange husk, tangerine husk , Mixture , potato media	*T.hamatam*	Heba et al.,2013
59	Organic amendment and oil cakes	*T.harzianum T. viride*	Jahagirdar et al.,1998

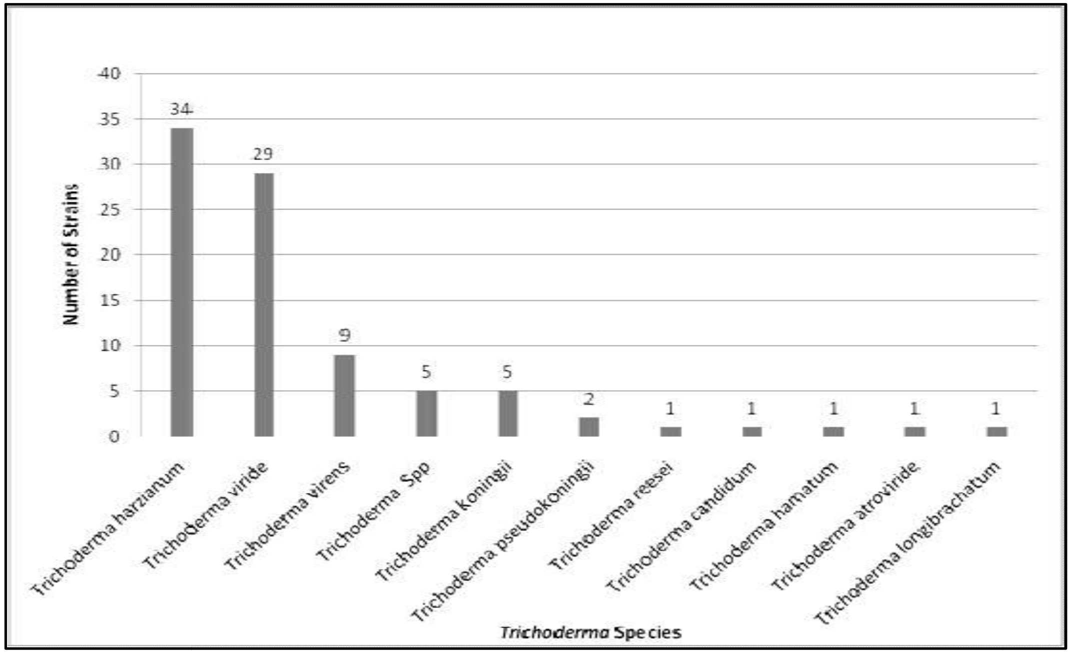

Figure 1. Potential of *Trichoderma* species for mass multiplication on agro waste materials worldwide

Thangavelu *et al*. (2004) tested five different organic substrates viz. rice bran, rice chaffy grain, farm yard manure, banana pseudostem and dried banana leaf for mass multiplication of *T. harzianum.* Various low-cost agricultural waste products were screened for mass multiplication of *T. harzianum* under normal storage condition and most effective formulated product of T. harzianum was evaluated in vivo and under field condition (Bhagat and Pan, 2007). A tea waste was the best substrate for mass multiplication of the bioagents and the mass multiplied cultures could be stored for 3 months without much reduction in the population of the biocontrol agents. Among the substrates, the most beneficial substrate was sorghum sp. found to be best for mass multiplication of *Trichoderma viride* (Singh and Singh, 2007). To overcome cost limitation, alternative substrates such as rice husks, coffee, sugarcane waste, rice bran, corn meal were used (Ha, 2010) and these materials found to be good for mass production of *Trichoderma* species.

DISCUSSION

Biocontrol agents like *Trichoderma* species are acclaimed as effective, ecofriendly and cheap, nullifying the ill effects of chemicals. Therefore these biocontrol agents are identified to act against as array of important plant pathogens causing serious diseases to crops. *Trichoderma* is so popular that its commercial formulations are available for field application and is not generally known to affect human health. However, it is also known that these biocontrol agents should be native for their good efficacy against their target pathogens. A biological control of plant disease appears to be an effective and ecofriendly approach being practice world over. Further biological control strategy is highly compatible with sustainable agriculture and has a major role to play as a component of integrated pest management. Large scale production, along with shelf life and establishment of bioagents in tartgeted niche, determine the success of biological control. Therefore cost effective large scale production, shelf life of formulation, establishment of bioagent in to targeted niche and consistency in disease control are the primary concern. Development of acceptable easily prepared and cost effective formulations for delivery should be major goal.

At present, *Trichoderma* species has emerged as a potential biological control agent which has been found effective against many economically important plant pathogenic fungi. It is a pre-requisite for biological control application to develop mass production that should be easy for operation. The mass production of *Trichoderma* species is now important task for the research work. Their mass production practices should be economical, easy to handle, less time consuming and safe. Other important factors are necessary to determine and proper study ofnutritional requirements must be essential. The common property of *Trichoderma* is its mycoparasitic and saprophytic mode of nutrition. The nutritional requirements of the *Trichoderma* species should be considered for their effective mass multiplication. Therefore in this investigation, efforts on mass production of *Trichoderma* species on various substrate materials were evaluated. It was also considered that *Trichoderma* species preferred the carbohydrate and protein materials for their growth and sporulation. The success of biological control depends not only the isolation, characterization and pathogenicity, but on the successful mass production of the fungal agent in laboratory.

Many workers made their efforts in mass multiplication of *Trichoderma* species and the present study was in accordance with these findings. *Trichoderma harzianum* and *T. virens* were grown on tea waste and found to be best media for mass multiplication (Prakash *et al*., 1999). Higher spore count of *T. harzianum* was obtained in molasses yeast medium followed by sorghum grains and broken maize grains (Pramodkumar and Palakshappa, 2009). Sawant *et al*. (1995) reported coffee wastes and poultry manures were excellent substrates for the growth of *T. harzianum*. Sethuraman (1991) reported that increase in *Trichoderma* population was due to application of organic amendments viz., rice bran and farm yard manure (FYM) in soil. Various low cost agricultural waste products were screened for mass multiplication of *T. harzianum* by Katiuar *et al*. (2007) and reported that wheat bran + mustard oilcake was the best substrate supporting the growth of antagonist under normal condition. Swankhi (*Sorghum* sp.) local millet of Jammu was found to be best substrate for *T. viride* and provided maximum spore concentration, spore viability and total biomass of fungal antagonist (Tiwari *et al*., 2004). Wheat bran and saw dust with two percent molasses was the best medium for mass multiplication of *T. viride* and *T. harzianum* (Mathur and Sarbhoy, 1978). Kousalya (1989) mentioned that sand maize medium, sorghum grain medium, wheat bran, rice bran and wheat grains were good for mass multiplication of *Trichoderma* species. Panicker and Jayarajan (1993) tested five substrates among which farm yard manure was the best for mass production of *T. viride* and *T. harzianum* followed by wheat bran and rice bran. Patel and Mishra (1994) evaluated cereal grains and agricultural wastes as substrates for mass multiplication of biological agent *T. harzianum*. Kalpana *et al*. (2007) reported that agrowaste materials such as farm yard manure, vermicompost, poultry manure, decomposed coir pith and combination of these materials proved to be more promising for the growth and sporulation of *T. viride*.Cow dung, neem cake, coir pith, sorghum grains, saw dust, and rice bran, either alone or in certain combinations, with orwithout additives such as jaggery and wheat flour, and having differential moisture levels were evaluated as substrates for mass production of *Trichoderma harzianum* and *T. viride (*Rini and Sulochana,2007). Fittingness of the wheat bran for the growth of *Trichoderma* has been reported by Lumsden and Lewis (1988) and Singh (1994) and Rama *et al*. (2001). Bheemaraya *et al*. (2011) tested various agrowaste materials for mass multiplication of *T. viride, T. harzianum* and *T. piluliferum* and reported that rice husk was found to be superior substrate.

Conclusion

The solid agrowaste materials were suitable for mass production of *Trichoderma* species. By adopting proper care, the process of mass multiplication was very easy to handle. The inoculums of potential biocontrol agent can be maintained in the field forever. The mass multiplication practice gave the local people opportunities to reduce health risks, costs and environmental damages. Among all *Trichoderma* species, *T.harzianum* and *T.viride* have better potential to colonize the agrowaste materials worldwide. Many strains were isolated because they were observed parasitizing plant pathogens or inhibiting their growth in culture. The enzymes and antibiotics produced by *Trichoderma* species that appear to be involved in biocontrol are strongly influenced by the substrate on which the fungus is grown. The farmers should not depend on the commercial products because these had no longer performance. In order to tackle these global problems, effective alternatives to chemical control are being investigated and the use of antagonistic microbes as biocontrol agent seems to be one of the promising approaches. With the advent of biocontrol as a potential approach to Integrated Pest Management (IPM) in the area of fungi-mediated plant disease control, the genus *Trichoderma* has gained considerable importance. Increasing production and productivity of the crops is need of the hour to feed the increasing population and also to overcome the problem of decreasing land for agriculture.

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