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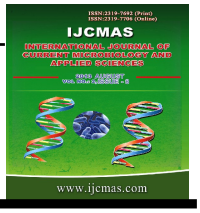

Original Research Article

Assessment of groundnut shells as a carrier material for starter cultures of fungal inoculum designed for rapid composting

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Groundnut shells; composting; carrier material; starter cultures; cellulolytic *Chaetomium.*

A B S T R A C T

Modern composting technologies make the best use of efficient microorganisms with noteworthy cellulolytic capabilities, for reducing the usually long durations of the traditional processes. Complications and difficulties in safe packaging, transportation and handling of conventional microbial cultures of superior cellulolytic organisms in laboratory vessels by the users, comprising mostly laymen with little or no technical expertise in management of such cultures, at faraway site locations are some of the commonly encountered 'on site' difficulties which have been hampering the widespread usage of efficient fungal isolates in composting. Development of user- and environment-friendly starter cultures of proven, superior cellulolytic microorganisms for composting, based on compatible carrier materials is a practicable solution to these problems. The communication deals with utilization of groundnut based agro-wastes viz. groundnut shells, as carrier material for development of starter culture formulations of efficient cellulolytic fungal inoculum for applications in composting. Suitability of the chosen agricultural residue as carrier material, compatibility trials with efficient cellulolytic fungal inoculum, usage of the carrier material based starter cultures in composting and shelf life of the carrier based culture formulations is discussed. Pulverized groundnut shells were appraised for their suitability as carrier material for starter cultures of cellulolytic fungal inoculums, which were successfully tested for composting of religious refuse. Groundnut shells were found extremely suitable as carrier material for fungal inoculum comprising species of the cellulolytic genus *Chaetomium*.

Introduction

The recent past has witnessed exemplary shifts towards sustainability accompanied by a rise in the popularity of sustainable organic agriculture (Falvey, 2002), one of the consequence of which has been a renewal of interests in the recycling of organic wastes, a long-standing but nevertheless neglected concept, which has once again regained recognition. India, with its traditional agriculture based economy, has always had a vast potential of local renewable organic resources; both

agricultural and agro- industrial wastes (Gaur *et al.*, 1995) that can be profitably utilized for enhanced soil improvement and crop production through recycling; a time-tested and environment friendly practice. The world is currently facing herculean difficulties related to disposal of the tremendous quantities of wastes generated by its ever growing population. Composting has been advocated and promoted as an effective and economical answer for dealing with solid organic wastes (Hubbe *et al*., 2010). Their utilization for preparation of organic manure not only provides essential plant nutrients and organic matter to soil, but also facilitates hygienic disposal of the wastes and residues, which would otherwise cause pollution and sanitation hazards.

Current research attention has been dedicated to reducing the usually lengthy periods associated with conventional composting techniques, by employment and exploitation of active decomposer microorganisms to accelerate the process; and also, on improving the quality of the product (Taiwo and Oso, 2004, Gautam *et al*., 2011). While both cellulolytic fungi as well as bacteria can effectively carry out the process of composting, the fungi, owing to their mycelial structure, are known to be more efficient. Some of the hurdles encountered in the popularizing of composting technology, involving efficient fungal organisms, are the safe packaging and transportation of the starter inoculum to the end users; mostly comprising laymen and farm labourers, having little or no expertise in sophistication required for handling of laboratory fungal cultures. Suspensions, cultures and combinations of efficient inoculants, housed in conventional laboratory vessels, need effective modification, facilitating packaging for safe transportation and usage by this clientele. A few decades ago, the need for development of compatible carrier materials for formulations of efficient decomposer inoculants was felt (Subba Rao, 1984), resulting in search and recognition of some classic carrier materials (Smith, 1992). Carrier based formulations of efficient microorganisms for rapid composting (Rasal and Patil, 2001) are generally based on cereals; jowar grains being the material of choice as carrier (Nakasone *et al*., 2004). Scanty and scattered information exists on the hunt for novel carrier materials for starter cultures of cellulolytic inoculum for composting and there is plenty of scope for promotion of many more, better and cheaper carrier materials. The present investigation was initiated to develop an effective and user friendly carrier based delivery system for efficient composting organisms. Groundnut wastes, namely, shells in pulverized form, were evaluated for their suitability as carrier material for starter cultures of cellulolytic fungal inoculum designed for rapid composting of organic residues.

Materials and Methods

The groundnut residues selected for evaluation were abundantly available in the areas of study, viz. Mumbai (the financial capital of India) and the adjoining city of Thane in the state of Maharashtra, situated on the western coast of India. After procurement, they were heated to 80^0C for 4 hours; pulverized and brought to a uniform 20 mesh size (Nakkeeran *et al*., 1997) as pre-treatment. Fifteen grams of pre- treated carrier materials in separate glass bottles of 100 ml capacity each, were then adjusted to 60% moisture (Mishra, 2002) with Reese

liquid medium, and autoclaved at 20 lbs psi pressure for 1 hour followed, on cooling, by inoculation with 2ml of spore suspension (10^6 spores/ ml) of the respective cellulolytic isolate. The experiment was carried out in triplicate with control. Five superior isolates, belonging to the cellulolytic genus *Chaetomium*, namely, *Chaetomium globosum* Kunze, *C.crispatum* Fuckel , *C.olivaceum* Cooke and Ellis , *C.nigricolor* Ames *and C. virginicum* Ames, isolated from various deteriorated cellulosic samples (Kolet, 2009; 2010a-c) were used as test inoculum. The bottles were capped under aseptic conditions and incubated at room temperature (28^0C) for 15 days, during which period the mycelial and ascomatal growths of the respective isolates on the carrier material were visually monitored and recorded.

On completion of the incubation period, the carrier based starter culture formulations were stored for a period of 4 months at room temperature in sealed polythene bags (Sethi and Adhikary, 2012). At the end of this period, 200 ascomata each, of the respective isolates, growing on the carrier material were plated out on PDA medium. Their ability to grow out and form colonies was recorded and interpreted as viability of the carrier based starter culture inoculum.

The starter cultures obtained, were used as test cultures for composting of religious refuse (floral wastes), carried out in the laboratory in triplicate with control. Individual starter culture based inoculum of the 5 isolates was mixed in equal proportions by weight and applied @ 100g/100kg raw material. The progress was monitored for a period of 4 weeks.

Results and Discussion

Groundnut being a prominent crop and commodity of trade in and around the areas of study, the carrier material selected for the current study was abundantly available. Large quantities of groundnut shells are obtained during shelling operations. Pre-treatment of the selected carrier material was carried out in accordance with the recommendations of Wahal (1998).The characteristics of the pre-treated carrier material are presented in Table 1. The pre-treated material exhibited high organic matter and water holding capacity; and a C: N ratio favourable for growth and multiplication of cellulolytic inoculum. It satisfied the pre- requisites of a good carrier base; findings of which are in agreement with Malusa *et al*., (2012). The cellulolytic fungal inoculum was multiplied, maintained and stored in the carrier material after the initial incubation period. The pattern of initial 15 days growth of the inoculum on the carrier material is presented in Table 2.

Carrier material comprising pulverized groundnut shells displayed excellent growth and multiplication of the cellulolytic fungal inoculum, which can be clearly interpreted as compatibility of the carrier material with the starter organisms; a phenomenon most probably due to the groundnut shells being rich in essential plant nutrients (Prasad and Kumar, 1998) and the favourable C: N ratio exhibited by them. There were no lump formations in the pulverized material throughout the period of study, thereby facilitating proper aeration and maintaining favourable conditions for growth.

Table.1 Characteristics of the Pre-treated Carrier Material

Characteristic	**Carrier Material: Groundnut shells**
Colour	Pale Brown
Texture	Fine powder
Particle size	Passed through 20 mesh size
Moisture (%)	4.5
Organic Matter (%)	71.02
Organic Carbon (%)	41.20
Nitrogen (%)	1.62
C:N Ratio	25.4 : 1
Water Holding Capacity (water held by 10 g sample) (ml)	29.5
Effective water holding capacity (%)	295

Table.2 Fifteen days growth of test organisms on the carrier material for starter cultures

Test Organism	**Pattern of growth on Groundnut shells**		
	5D	10D	15D
Chaetomium globosum	+++/+	+++/+	+++/+++
C. crispatum	+++/+	+++/+++	+++/+++
C. olivaceum	++/+	+++/+++	+++/+++
C. nigricolor	++/++	+++/+++	+++/+++
C. virginicum	+++/+++	+++/+++	+++/+++

Mycelial development/ ascomatal development; '-': absence of growth, '+': *poor*, '++': moderate, '+++': rich growth

Table.3 Viability of starter cultures based on groundnut shells after 120 days of storage

Test Organism	**Percent viability** (%)
Chaetomium globosum	90.6
C. crispatum	94.8
C. olivaceum	91.4
C. nigricolor	99.0
C. virginicum	98.6

The shelf life of the starter cultures was monitored after 4 months of storage. The fungal inoculum multiplied and was maintained in the favourable environment provided by the carrier material. Results depicted in Table 3 reveal high percentage of viability of the inoculum after the period of storage. The findings are in conformity with those of Abdel-Kader *et al*. (2012).

The application of starter cultures based on the carrier material at the rate of 100g/ 100kg (1kg/ ton) raw composting material effected faster degradation of religious floral refuse vis-à-vis control set and brought about 75.60 per cent and 74.00 per cent reductions in volume and weight respectively, in the substrate, as against 43.26 per cent and 49.7 per cent respectively in the control set. The C: N ratio dropped by 37 per cent and narrowed down to 15.13 from the initial 24.05 in the composting period of 4 weeks as against a modest 11 per cent drop in the control set. The influence of the carrier based starter inoculum on the process of composting is presented in Table 4. The results are in agreement with Subba Rao (1997) and Jusoh *et al*., (2013). A variety of carrier materials have been evaluated for various purposes (Yardin *et al*., 2000; Simons *et al*., 2012; Selvi, 2013). Presently, agro-wastes are also being assessed as carriers of microbial inoculant formulations (Covino *et al*., 2010, Kaljeet *et al*., 2011). Lignite and peat based bacterial activators of composting as well as, fungal inoculants for composting, based on cereal grains are also available (Bashan, 1998; KKV, 2003).

The utilization of agro-residues has multiple advantages. Apart from abundant availability, they are annually renewable, extremely cost-effective; their use revolves around simple processes and equipments and does not damage the environment. Their ligno-cellulosic nature and favourable carbon: nitrogen ratio makes them ideal substrates for mass production of cellulolytic inoculum for composting. While some amount of the groundnut residues may be put to some use, a major quantity is wasted or burned and the ash used as manure (CSIR, 1948).

Table.4 Influence of the carrier based starter culture inoculum on hastening the process of composting of religious floral refuse

Treatment	Compost sampling interval: 4 weeks					Reduction in OC (%)	Reduction in volume (%)	Reduction in weight (%)
	OC (%)	N (%)	C:N ratio	Volume (Cm^3)	Weight (g)			
Control: only raw material	48.60	2.27	21.40	14595	2515	10.59	43.26	49.7
Raw material + carrier based starter inoculum	35.72	2.36	15.13	6275	1300	34.28	75.60	74.0

Initial values- OC: 54.36; N: 2.26; C:N ratio: 24.05; volume: 25725 cm^3; weight: 5kg

Groundnut shells have also been used for various purposes such as fodder (Aregheore, 2000, Ketkar, 2001), industrial fuel (Srikanth *et al*., 2004), in manufacture of particle boards (Chawla, 1980), low cost adsorbent (Malik *et al*., 2007), soil conditioner and stabilizer (Zende, 2002; Oriola and Moses, 2010), substrate for production of enzyme (Couto and Sanroman, 2006) and as alternative source of carbon for growth of fungal cultures (Khetmaldas *et al*., 1984) among several other usages. The commercial usage of this substrate as a carrier material for development of a delivery system of fungal starter inoculum for composting is awaited. A delivery system comprising starter cultures for composting was developed from a novel carrier material namely groundnut shells, tested and found effective. Apart from gainfully utilizing this ligno-cellulosic residue which otherwise is wasted, the cost-effective starter cultures would be beneficial in the easy distribution of efficient starter cultures for faster composting. Groundnut shells were found to be extremely suitable as carrier material as shown by their compatibility with the cellulolytic fungal inoculum.

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