http://www.bpasjournals.com
Print version ISSN 0970 0889
Online version ISSN 2320 3161
DOI 10.5958/2320-3161.2019.00004.X
Original Research Article
Bio-Science Research Bulletin
Vol. 35 No. 1, January - June 2019: P.18-25

# Management of Post Harvested Paddy Crop Residues by *Aspergillus* Species for Sustainable Agriculture

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**Received: Dec 18, 2018**
**Revised: Mar 11, 2019**
**Accepted: May 28, 2019**
**Published: June 20, 2019**

**ABSTRACT**

Rice, *Oryza* sp. (Angiosperm: Gramineae) is the main staple crop in the World and 493 million tons of rice was produced world-wide, which supposed to be a source of main crop residue as per the report of IGC (International Grain Council) during 2017-18. Therefore, it was an urgent need to management of the residue by the best suitable method i.e. biodegradation using potent fungal species. The present study was conducted to evaluate the potential mycobiota and analyze the mechanism of action for the management of post harvested crop residues for the sustainable growth. The findings of the current investigation reflected the pectolytic, lignolytic and cellulolytic activities of *Aspergillus* species. The observed worked out may also prove that these mycoflora played a key role for biodegradation of paddy residues. During the course of study a sum of nine fungal isolates were recovered from paddy residues (semicompost resource). Out of them, six isolates were selected on the basis of their enzymatic activities against lignin, pectin and cellulose constituents of paddy straw. All the six fungal isolates like *A. nidulans*, *A. wentii*, *A. tamarii*, *A. fumigatus*, *A. flavous*, and *A. sydowii* showed optimum lignocellulolytic activities, which were further used for *in vitro* decomposition of fresh paddy residue by the application of 10ml sample as spray on two gram of fresh paddy residue and incubated at 28 ± 1ºC for 30 days. At the end of 30 days experiment, the rate of decomposition was measured by ADR (Absolute decomposition rate) and RDR (Relative decomposition rate). It was found that *A. flavous* showed highest ADR and RDR; however, *A. fumigatus* represented the lowest ADR and RDR. Thus, from the findings authors are supposed to proposed and formulate a strategy for sustainable management of paddy crop residues. The practical application of the present study is assessed to be a mile stone for the sustainable environment and natural resources management to ecofriendly agricultural activities and livelihood sustainability.

**KEYWORDS:** *Aspergillus* sp., Paddy crop residues, Biodegradation, Pectolytic, Lignolytic, Cellulolytic.

## INTRODUCTION

Rice is the main staple crop in the world, where 486 million tons of rice was produced in 2016 – 2017 and 493 million tons in 2017-18 (IGC, International grain council). Also, in India 111.52 million tons of rice was produced in 2017-18 (Department of economics and statistics). Annually a large amount of straw is accumulated as a byproduct from rice cultivation, as straw makes up about 50% of the dry weight of the rice plant. Farmers do not incorporate rice straw in the crop field because of its slow degradation rate, disease infestation, unstable nutrients, and reduced yield caused by the short-term negative effect of nitrogen immobilization (Pandey *et al*., 2009). They usually dispose of it through open field burning. As a consequence, carbon dioxide, carbon monoxide, methane, nitrous oxide, and sulphur dioxide are emitted into the atmosphere. This process also emits harmful air pollutants such as polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs), which have toxic properties and are, notably, potential carcinogens that can cause severe impacts on human health (Gadde *et al*., 2009).

Thus, proper management and disposal of bulky rice straw is a serious concern all over the world. Attention has been focused on nonhazardous, environment friendly and sustainable techniques for safe disposal of rice straw in a short period of time (Singh *et al.,* 2018a). Microbial composting is an effective environmentally sound alternative for the recycling of rice straw into compost. It promotes sustainable agriculture and environmental protection, improving the soil's physical, chemical, and biological properties (Perez-Piqueres *et al*., 2006; Rasool *et al*., 2008; Mylavarapu and Zinati, 2009), which ultimately results in better plant growth and yield. Composting of lignocellulosic rice straw requires a process that ensures rapid biodegradation despite the fact that the lignin matrix shields cellulose and hemicelluloses from biodegradation. Naturally a few microbes have the potential to depolymerize lignin. Fungi have an advantage in the composting of lignocellulosic waste because they are filamentous and have the ability to produce prolific spores, which can invade substrates quickly. Moreover, mixed cultures can better influence colonization of the substrate through increased production of enzymes as well as resistance to contamination by other microbes.

The most important determinant in mixed cultures is strain compatibility, which influences the organization, distribution, and density of the micro habitat population and the ecological balance of the communities (Gutierrez-Correa and Tengerdy, 1997; Molla *et al*., 2001). Hence, a compatible lignocellulolytic fungal consortium might play a vital role in the rapid disposal of rice straw. In order to address the fore going issues, this study was undertaken to isolate, screen, and evaluate the compatible lignocellulolytic fungal consortium from ecologically related habitats for rapid and environmentally friendly composting of rice straw. It is clear from the above discussion that despite a number of studies already undertaken on the fungal deposition of paddy straw and its components, a number of aspects of this process still lie unresolved. The present study was, therefore undertaken which embodies an attempt at: (a) Isolation and identification of the fungi colonizing, (b) Assessing the rate of decomposition, (c) Biodegradation of the paddy residue.

## MATERIALS AND METHODS

Some bundles of harvested paddy straw were collected from the field of village Garhi Birbal, Karnal, Haryana. Different components *i.e.* internodes, leaves, kernels (spikes) of these plants were collected. Two-year older sample was collected, which was stored for some household purposes like fodder for animal or for packaging purpose. Collected sample was stored in open exposed to each environmental condition for growth of biodegradable substances. Collected paddy straw was partially bio-degraded by natural action of environment and microbial actions, having some-what blackish and whitish colonies and easily breakable into small particles (partially decomposed). The sample was collected and stored into white polythene zip bags, so that micro-flora of other areas do not disturb the *in vitro,* decomposition process of fresh paddy residue.

Serial dilution plate method (Waksman, 1927) was used for qualitative and quantitative assessment of the fungi colonizing decomposing paddy crop residue. After spreading the inoculum, the petri dishes containing the medium and inoculum were incubated at 28 ± 1°c for 3-5 days. Identification of the fungus was done on the basis of their morphological and cultural characteristics following Gilman, (1957), Barnett and Hunter, (1972), Subramanian, (1971), Ainsworth *et al.,* (1973) Von Arx, (1974) and Singh and Charaya, (2003).

## RESULTS

### The decomposer mycobiota

A total of nine forms of fungi have been isolated from the paddy crop partly decomposed residue. A list of nine species of fungi which sporulated and could be identified is given below. The broad taxonomic arrangement is based upon that proposed by Ainsworth (1966), for the use in the "Dictionary of fungi" (Ainsworth, 1971) and subsequently followed in "The fungi: An Advanced Treatise" Vol. IVA and B (Ainsworth *et al*., 1973). The genera and species with in a family are arranged and presented below:-

Deuteromycotina
Hyphomycetales
Moniliaceae
- *A. nidulans* Winter
- *A. wentii* Wehmer
- *A. tamari* Kita
- *A. fumigatus* Fresenius
- *A. flavous* Link
- *A. sydowii* Thom and Church (Bainier and Sartory)
- *A. fumiculosus* G. Smith
- *A. variecolor* Thon and Raper

Tuberculariales
Tuberculariaceae
- *Fusarium* sp.

The population ecology of the recovered fungal flora during the investigation against paddy crop residues decomposition was summarized in Table 1 showing a sum of 59 isolates with frequency percentage between 09 to 54%.

**Table 1: Frequency class, Frequency percentage, Total no. of isolates and percentage isolates of fungal sp. isolated from decomposing paddy straw.**

| Fungal sp. | Frequency class | Frequency % | Total isolates | Percentage isolates |
|---|---|---|---|---|
| *A. sydowii* | I | 27 | 9 | 15.25 |
| *A. fumiculosus* | II | 27 | 8 | 13.56 |
| *A. nidulans* | III | 54 | 11 | 18.64 |
| *A. flavous* | II | 36 | 9 | 15.25 |
| *A. fumigatus* | II | 27 | 3 | 5.08 |
| *A. tamarii* | I | 18 | 7 | 11.86 |
| *A. wentii* | I | 18 | 4 | 6.78 |
| *A. variecolor* | I | 09 | 6 | 10.16 |
| *Fusarium* sp. | II | 36 | 2 | 3.38 |

### Degradative enzyme potential of *Aspergillus* sp.:

One of the most important prerequisite for successful colonization of a substrate of a given fungi has its ability to produce various enzymes *in vitro* which can degrade the cell wall materials (cellulose and lignin) as well as the substances involved in the cementing together of the cells (pectin). Therefore, it was decided to study the potential of selected fungi to decompose carboxymethylcellulose (CMC), pectin as well as lignin for cellulolytic, pectolytic and lignolytic activities respectively. The six fungal species *viz. A. flavous, A. fumigatus, A. nidulans, A. sydowii, A. wentii,* and *A. tamarii* was selected for the study (Fig. 1).

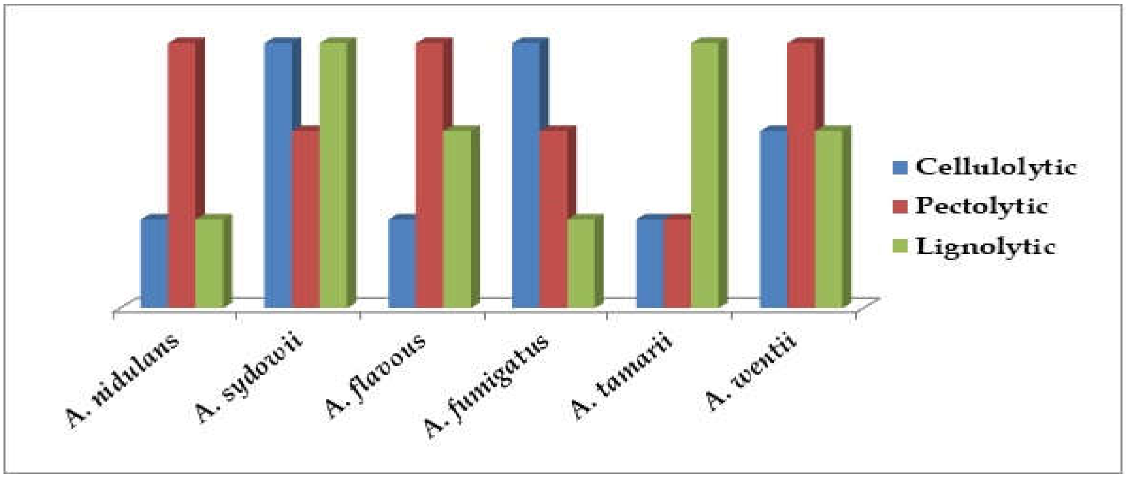


**Figure 1: *In vitro* enzymatic potential of selected *Aspergillus* sp.**

***In vitro* decomposition of paddy residues:**

All the six species of *Aspergillus* were tested for in vitro decomposition of paddy residues by spraying on fresh paddy samples and were allowed to decompose for one month. The results are present in Figs. 2-4.

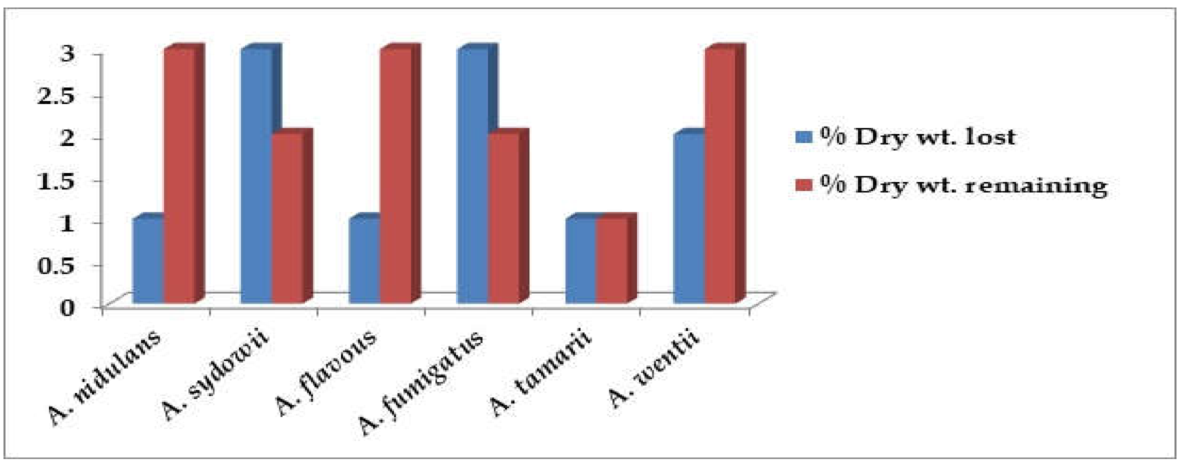


**Figure 2: Rate of decomposition of paddy crop residues by different *Aspergillus* species.**

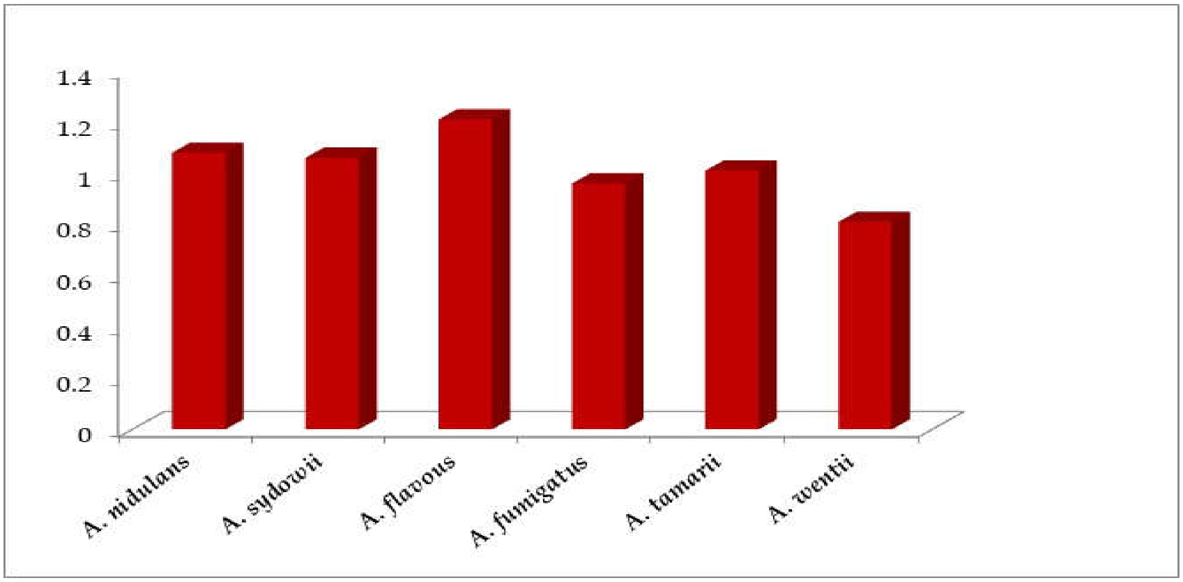


**Figure 3: Absolute decomposition rate (ADR) of paddy crop residues by different *Aspergillus* species.**

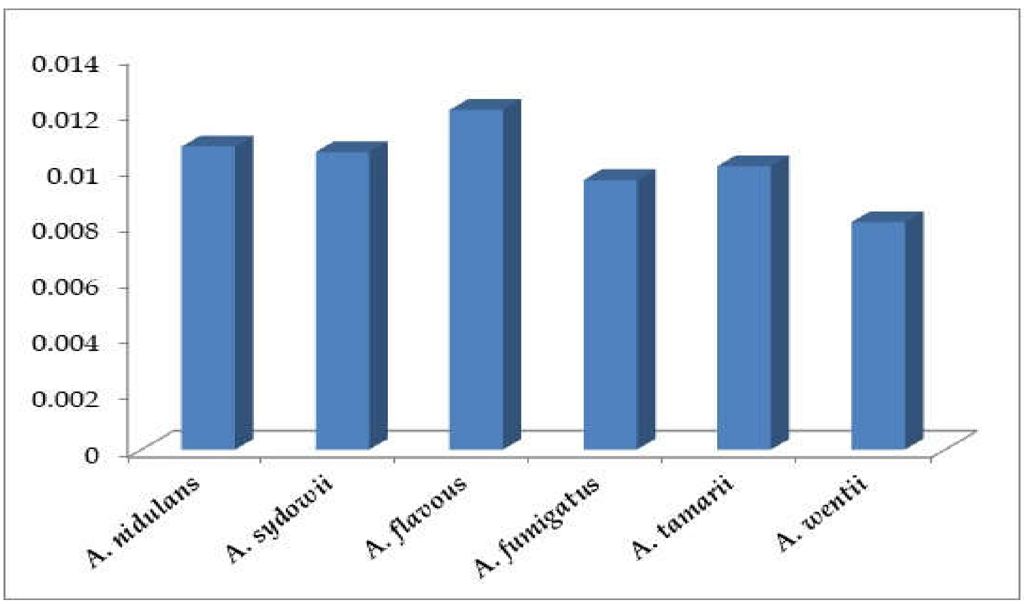


**Figure 4**: **Relative decomposition rate (RDR) of paddy crop residues by different *Aspergillus* species.**

## DISCUSSION

The process of decomposition of different components of paddy straw could help in devising better and more efficient method for their management. The present study was, therefore, conducted to have an understanding of the mycobiota decomposing paddy crop residues, the patterns of colonization of these resources by the mycobiota and chemical changes occurring in the resources during decomposition. Attempts were also made to have an assessment of the capability of selected fungi to decompose these resources *in vitro.*

**The decomposer mycobiota:**
A total of nine forms of fungi were isolated from decomposing paddy crop residue in the present study. All of them were identified and belong to Deuteromycotina, out of them eight fungi belong to order Hyphomycetales and one from Tuberculariales. The dominance of Deuteromycotina observed in the present study is in full conformity with the earlier reports (Hudson, 1968; Dickinson and Pugh, 1965; Hayes, 1979; Charaya, 1985; Singh, 2004; Singh and Charaya, 2003; Singh *et al.,* 2019). Along with them the presence of Tuberculariales in the present study is also in full conformity with the earlier report (Kadarmoidheen *et al*., 2012).

**Enzymatic activities**:
The bioconversion of cellulose, lignin and pectin containing raw material is an important problem of current biotechnology, due to increase demand for energy, food and chemicals (Solovyeva *et al.,* 1997). Cellulases are enzymes which hydrolysis the β-1-4 glycosidic linkage of cellulose and are synthesized by microorganisms during their growth on cellulose material (Lee and Koo, 2001; Singh *et al.,* 2017b). The pectins and lignins are also an integral constituent of cell wall which provides plant strength and resistant to microbial degradation (Singh *et al*., 2018b. In the present study, six fungal isolates were recovered and identified as: *A. nidulans, A. sydowii, A. wentii, A. fumigatus, A. flavous* and *A. tamarii.* The fungal isolates were examined to produce lignocellulase enzyme were cultured on Hankin and Anagnostakis media (modified), 1971 for carboxymethylcellulase and Waksman agar medium (modified) (Aggarwal, 1969) for showing pectin and lignin biodegradation activity. During the present discourse, *A. fumigatus* showed best CMCase activity (forms a clear zone), on contrary to Kadarmoidheen *et al.,* 2012. In his work *Trichoderma viride* was found to be the best in degrading CMC constituents of paddy crop residue but in present study *A. fumigatus* showed best activity against CMCase of paddy residues.

The biodegradation of crop residue by fungi is dependent on the plant and fungal species. There is a good relation between residue biodegradation and fungi lignocellulolytic enzyme activities (Singh and Upadhyay, 2019). The Hyphomycetes grew fast and produce more lignocellulolytic enzymatic activities on paddy crop residue (Sinegani *et al.,* 2005; Singh *et al.,* 2015). According to Sinegani *et al.,* 2005, *A. terreus* show best result against tannic acid activity, but in present study among the six fungal isolates *A. flavous* show best activity (form dark brown color zone) on paddy crop residue for degradation of lignin molecules. The pectins are a heterogeneous group of polysaccharides that compose the plant cell wall (Loperena *et al.,* 2012; Singh *et al.,* 2017a) characterized the production of pectolytic activity in several Antarctic fungi, using semi aquatic plate analysis method but did not find any fungi producing pectolytic activity. On the contrary of this Poveda *et al.* (2018) found eight strains of fungi producing pectolytic activity. In the present study among the six fungal isolates *A. flavous* represents best pectolytic activity against paddy crop residue (form clear zone). The appearance of this clear zone and dark brown zone represents the enzymatic activities of particular fungus against paddy crop residue. On basis of their action they are further more used for biodegradation of paddy crop residue *in vitro* conditions.

**Rate of decomposition**:
A number of workers in the past have tried to device suitable model to describe the rate of decay of natural materials under a variety of conditions. The most-simple one was linear model which considered that ADR (Absolute decomposition rate) was constant throughout the decomposition process for each fungus but the value of ADRs did not justified same. Similar observation was recorded by Charaya (1985).

There was sufficient variation in the capability of different fungus species to decompose the resources. The rate of decomposition varied not only from species to species but from resource to resource also. Interestingly, in present study of paddy crop residue decomposition by Hyphomycetes fungus *A. flavous* showed maximum 36.5% dry weight lost along with highest 1.21 ADR value after decomposition for 30 days and *A. fumigatus* showed least record with 29% dry weight lost and 0.96 ADR value. The present study data is contrary to Sannathimmappa *et al.* (2015). The result of the present investigation revealed the activity of decomposition rate of various fungi for microbial biodegradation which can be furthermore increased by addition of various reagents.

**ACKNOWLEDGEMENT**

Authors are very thankful to Dr. Anil K. Sharma, Prof. & Head, Department of Biotechnology, Maharishi Markandeshwar (Deemed to be University), Mullana-Ambala, Haryana, India for perpetual moral assistance and critical suggestions during compilation of research findings.

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