Plant Archives Vol. 18, Special Issue (ICAAAS-2018), 2018 pp. 376-381 *ISSN 0972-5210*

EFFECT OF VARIOUS SPAWN BASE SUBSTRATES ON THE SPOROPHORE YIELD OF MULTISPORE ISOLATE OF *PLEUROTUS SPP.*

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Abstract

Mushrooms have achieved significant importance in many countries due to their high nutritive and genuine medicinal values as well as an income generative venture. Blessed with varied agro-climates, Indian weather is aptly suitable for the cultivation of edible mushrooms. Among the various spawns supplemented with pulse flour the spawn of the isolate *Pe* x *Po* supplemented with horsegram flour at three per cent level recorded the minimum spawn run days (9.86 days), maximum yield (625.45 g/bed) and bio-efficiency (125.09%). The spawn of *Pe* x *Po* supplemented with cowpea flour recorded the minimum yield (588.38g/bed) and bio-efficiency (117.68%). The isolates *Pc* x *Pe, Pc* x *Pfl, Pf* x *Po* and *P. eous* came next in the order of merit with the same trend as that observed in the isolate *Pe* x *Po.* Generally all the gram flours increased the sporophore production when compared to control. Among the gram flours, supplementation of spawn base with horsegram flour at 4 per cent level registered the maximum yield of all the isolates.

Keywords: *Pleurotus spp.,* sporophore,multispore, spawn base and pulse flour.

Introduction

Mushrooms have achieved significant importance in many countries due to their high nutritive and genuine medicinal values as well as an income generative venture. Blessed with varied agro-climates, Indian weather is aptly suitable for the cultivation of edible mushrooms. The entire coastal belts of India running in to thousands of kilometers is a potent place to produce low cost speciality mushrooms which could supplement the protein deficiency and malnutrition, besides bringing in a sky – rocketing export market of a kind which is incomparable to any single cell protein (SCP) product (Kohlii, 2000).

It is estimated that about 355 million tonnes of crop residue is generated annually and about 170 million is left out posing problems for disposal (Tewari and Pandey, 2002). Even if one per cent of this agricultural waste is used to produce mushrooms, India will soon become a major mushroom producing country in the world. Mushroom production is the only biotechnological means available to convert these agricultural wastes into highly valuable edible proteins. So far around 5658 species of mushroom in 230 genera have been recorded from all over the world; whereas from India 850 species spread over 115 genera have been reported. Of this 850 species about 20 are being commercially cultivated (Saini and Atri, 1995).

Among these, the white button mushroom(*Agaricus bisporus*), oyster mushroom (*Pleurotus* spp.), paddy straw mushroom (*Volvariella volvacea*) and milky mushroom (*Calocybeindica*) are popular among the commercial growers in India as the techniques for their cultivation have been well developed (Vijaya Khader *et al.,* 1998). World mushroom production at present is estimated to be around 5 million tonnes/annum and is increasing @ 7 per cent/annum. The total mushroom production in India has increased from 4000 tonnes in 1955 to 30,000 tonnes in 1995 and it is estimated to be around 50,000 tonnes / annum (Tewari, 2004).

Agaricus bisporus is highly temperature specific, and its cultivation is restricted to temperate regions. But oyster mushrooms can be cultivated easily in tropical and subtropical regions. Hence, it is rightly named as "the crop of the future". *Pleurotus* spp. has the ability to degrade most of the lignocellulosic agro wastes, thus the cultivation of this mushroom is an efficient means for the conversion of agricultural wastes in to valuable edible proteins (Deepika Sud and Sharma, 2005).

The farmers and consumers have also developed preference towards *Pleurotus* spp. in recent years because of its advantages *viz.,*high nutritive value and easiness in cultivation using the farm wastes (Eswaran, 1998). Among the thirty eight species of *Pleurotus* existing in nature, only nine species are being cultivated under artificial condition (Jandaik, 1987). Every species has its own attributes and each is known for its yield, substrate utilization and wide temp. adoption (Ravichandran, 2001). Inspite of its easy

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cultivation methods and adaptation to wide range of temp., the production of *Pleurotus* spp. is very less when compared to button mushroom production in India. Hence, a need was felt for up scaling the yield potential of *Pleurotus* spp. for large scale production.

Materials and Methods

Organism

The pure culture of *Pleurotus* spp. (*Pleurotus citrinopileatus* (Fr.) Singer, *P. djamor* (Rumph.) Boedijn, *P. eous* (Berk) Sacc*, P. flabellatus* (Berk and Br.) Sacc., *P. florida* (Eger) and *P. ostreatus* (Jacq.Fr.) Kummer) were obtained from National Centre for Mushroom Research (NCMR) Chambaghat, Solan, Himachal Pradesh. The sub cultures were maintained on oat meal agar (OMA) medium.

Pc- Pleurotus citrinopileatus

Pd- Pleurotus djamor

Pe- Pleurotus eous

Pf- Pleurotus flabellatus

Pfl- Pleurotus florida

Po- Pleurotus ostreatus

Isolation and purification

The mushroom tissue was cut at the junction of the pileus and stipe using a sterile scalpel and surface sterilized with 95 per cent ethyl alcohol for one min. These bits were placed on OMA in sterile Petri dishes and incubated at room temp. (28 ± 2° C) for seven days. The isolates were then purified by single hyphal tip method and maintained on OMA slants.

Preparation of spawn

Sorghum grain spawn was prepared by adopting the method described by Sivaprakasam (1980). Sorghum grains were partially cooked in water for 40 min. After draining the excess water, the grains were mixed with calcium carbonate at two per cent (w/w) to prevent adhesion of the grains and for optimizing pH. The grains were filled up to two-third volume of glass glucose drip bottles plugged with non-absorbent cotton wool; the mouths were wrapped and sterilized at a pressure of 15 psi. for two h. The grains were inoculated with pure cultures of the fungus and incubated at room temp. (28 ± 2° C). All these were carried out under aseptic condition. The nature of the growth and time taken for complete colonization of the spawn were recorded.

Effect of pulse flour supplementation to spawn base on the sporophore yield

Various pulse grains *viz*., redgram, blackgram, greengram, horsegram, bengalgram and cowpea were sun dried, ground to powder form and added with the spawn substrate @ 30 g/kg level along with calcium carbonate @ 20g/kg of the substrate (Kalaiselvi, 2003). Then these substrates were filled in poly bags and sterilized. After sterilization the bottles were inoculated with stock culture of 9 mm mycelial disc and incubated at room temp. (28 ± 2^0C) for 10 – 15 days. This spawn was used for bed preparation and all the observations on yield attributes as already described were recorded. The effective supplement (horsegram flour) identified in the preset study was further evaluated at graded levels *viz*., 1, 2, 3, 4, 5 and 6 per cent. IFPg spawn without any supplementation served as control for both the experiments.

Results

Effect of supplementation of spawn base with various pulse flours on spawn growth and sporophore yield

Generally supplementation of spawn base with pulse flours hastened the spawn run period and increased the sporophore yield of all the isolates tested when compared to control. Among the various pulse flours used at three per cent level, horsegram flour exerted a significant influence on the spawn growth which recorded the minimum spawn run days and maximum sporophore yield in all the isolates tested. This was followed by bengalgram, greengram, blackgram and redgram flours. Cowpea flour was found as the least effective among the pulse flours tested as it recorded higher spawn run days and minimum sporophore yield in all the isolates. The spawn with out any supplementation recorded the maximum spawn run days and minimum yield .

Among the various spawns supplemented with pulse flour the spawn of the isolate *Pe* x *Po* supplemented with horsegram flour at three per cent level recorded the minimum spawn run days (9.86 days), maximum yield (625.45 g/bed) and bio-efficiency (125.09%). The spawn of *Pe* x *Po* supplemented with cowpea flour recorded the minimum yield (588.38g/bed) and bio-efficiency (117.68%). The isolates *Pc* x *Pe, Pc* x *Pfl, Pf* x *Po* and *P. eous* came next in the order of merit with the same trend as that observed in the isolate *Pe* x *Po.*

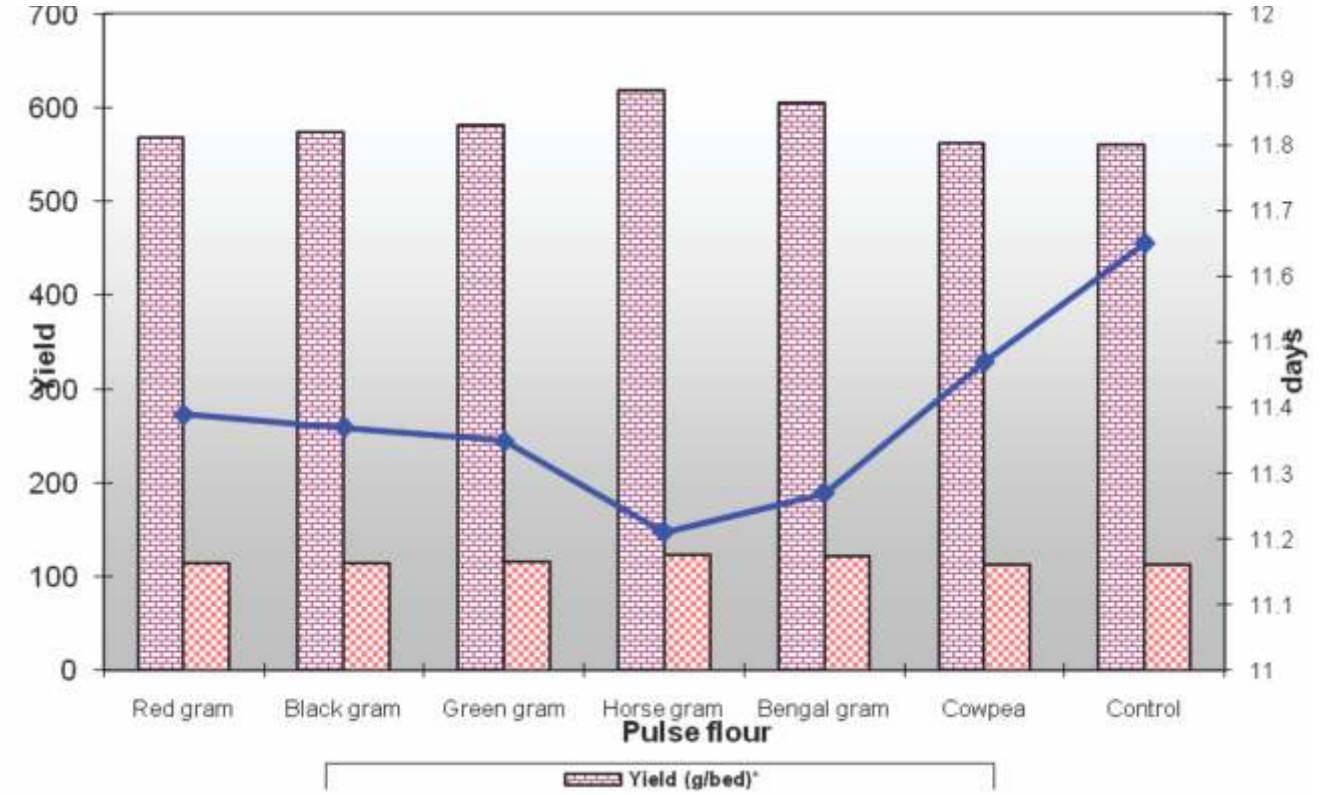

Fig. 1. Effect of pulse flour on spawn growth and yield potential of Pc x Pe

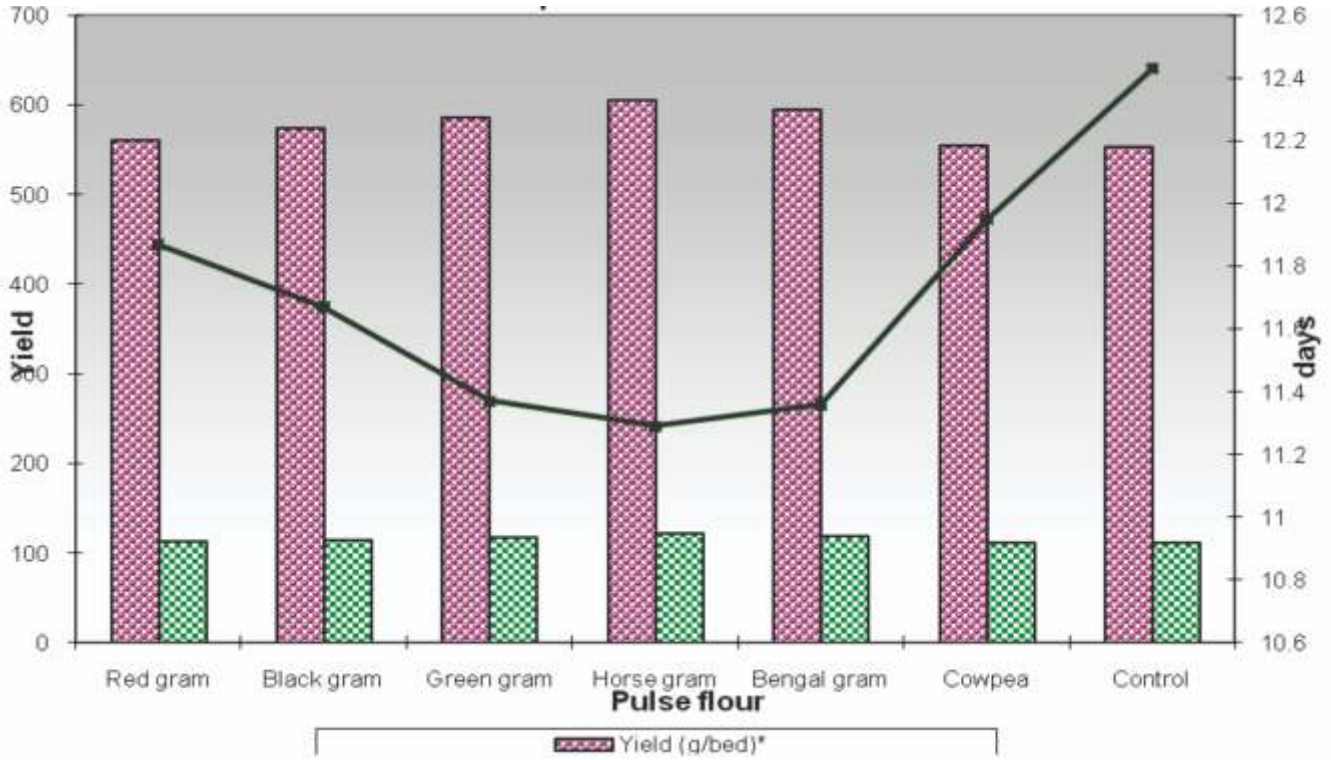

Fig. 2. Effect of pulse flour on spawn growth and yield potential of Pc x Pfl

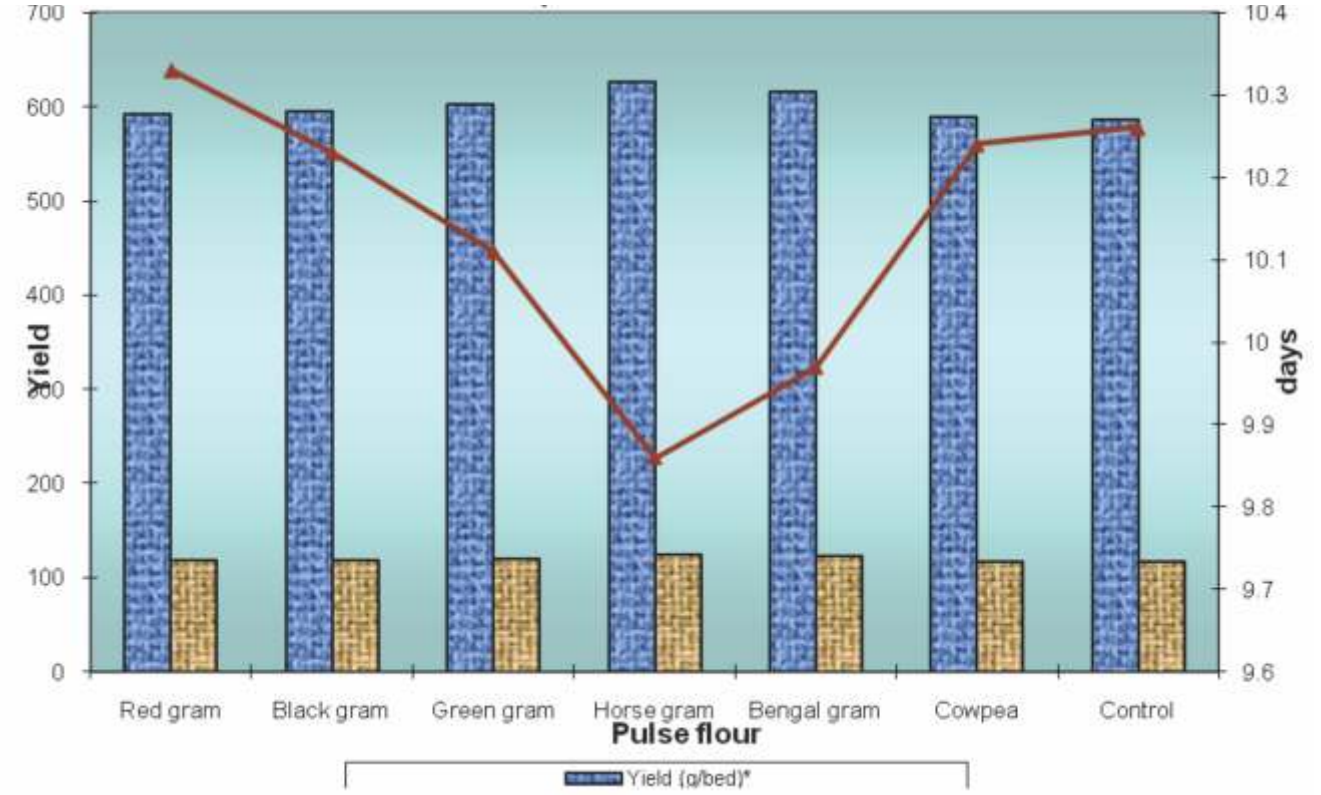

Fig. 3. Effect of pulse flour on spawn growth and yield potential of Pe x Po

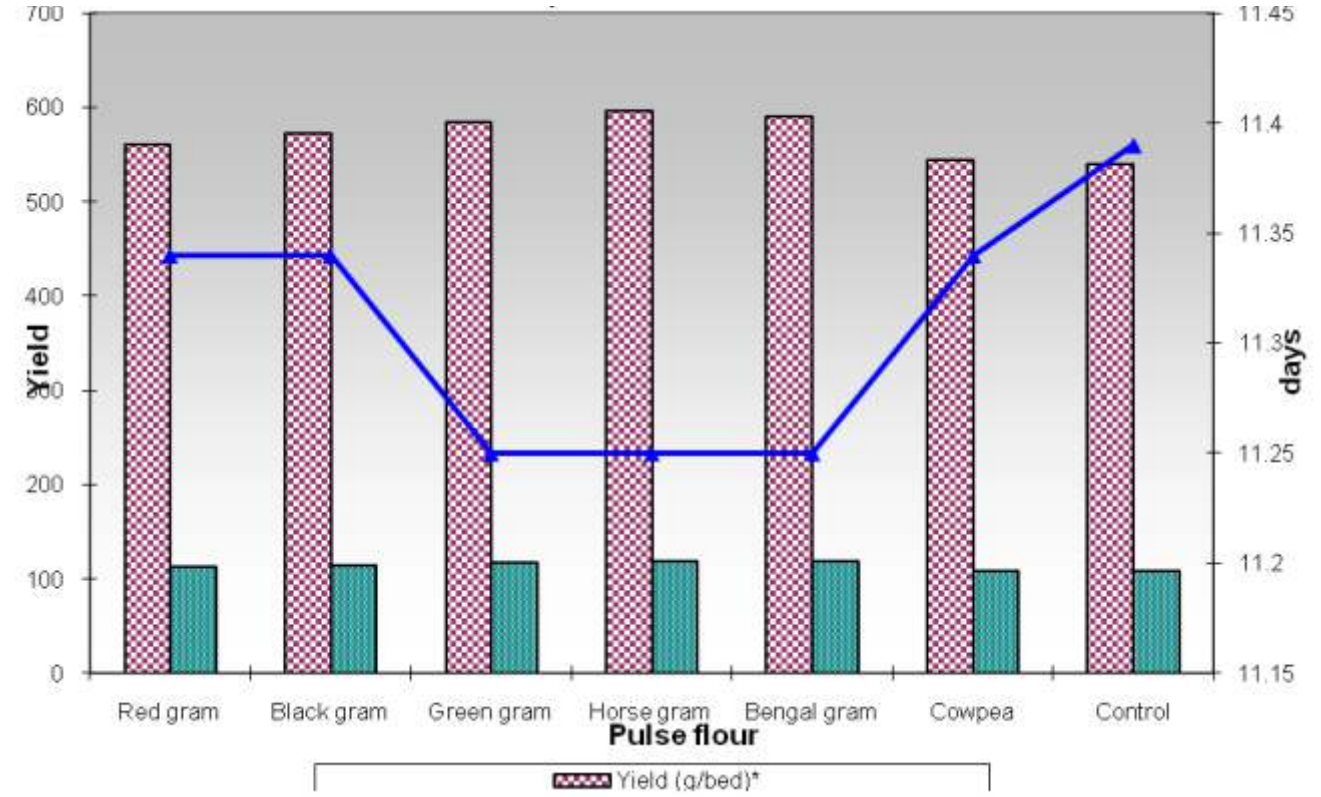

Fig. 4. Effect of pulse flour on spawn growth and yield potential of Pf x Po

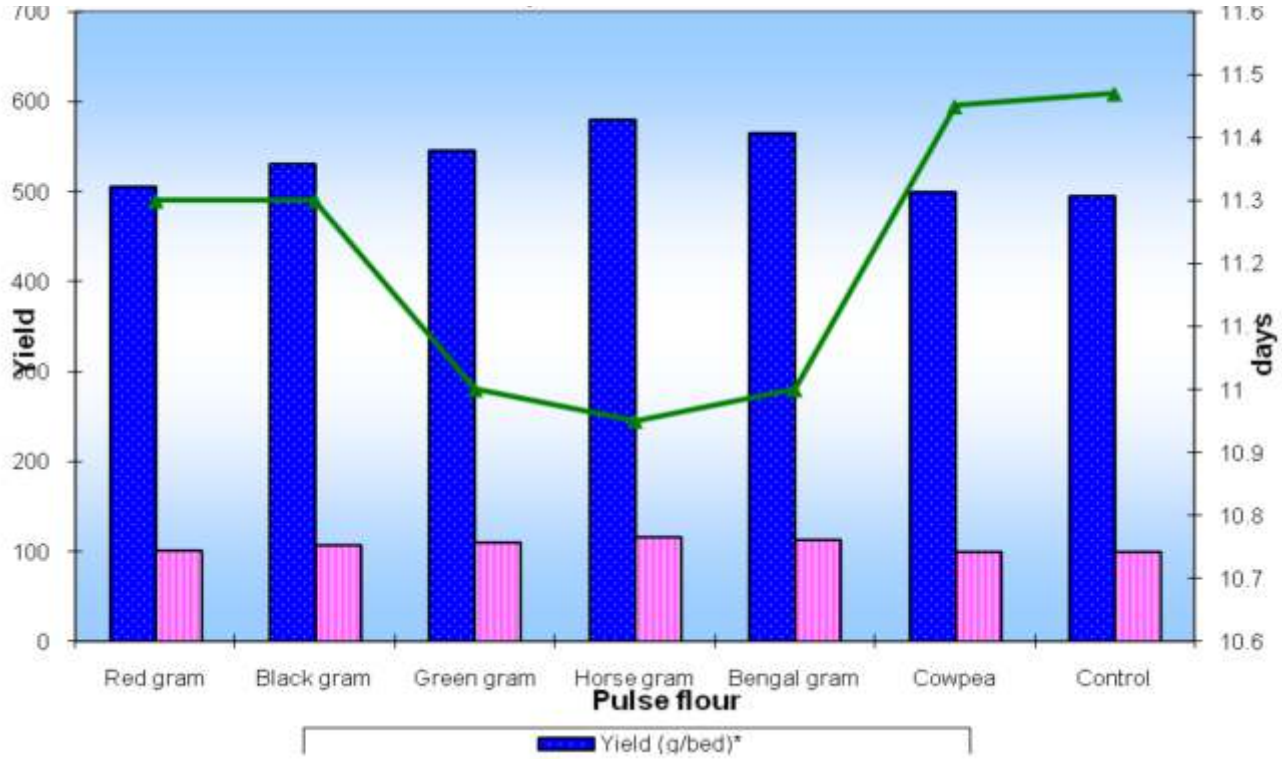

Fig. 5. Effect of pulse flour on spawn growth and yield potential of *P. eous*

Discussion

The present study revealed that supplementation with pulse flour on spawn production increased the yield of multispore isolates of *Pleurotus* spp. Supplementation with horsegram flour hastened the days required for the spawn growth and first harvest and also increased the yield in all the four multispore isolates viz., *Pc x Pe, Pc x Pfl, Pe x Po, Pf x Po* and standard parent when compared to control (Fig. 1 to 5).

Similar results were reported by earlier workers (Jaganathan, 1972; Eswaramurthy *et al.,* 1983). Kathe *et al.* (1996) observed that supplementation ofsoybean flour at 3 per cent concentration was found to be better than other flours. The spawn base supplemented with horsegram flour 3 per cent + Gypsum 1 per cent recorded the maximum sporophore numbers and higher yield of *Pleurotus* spp. (Eswaran, 1998; Senthil kumar, 2004; Senthilmurugan, 2004).

Sorghum grain added with gram flour as spawn substrate hastened the yield and sporophore maturity of *P. sapidus* (Sanjeevkumar, 2002). All these reports confirm the present findings. The stimulation activity of horsegram flour supplements might be due to increase in the activity of beneficial saprophytic bacteria which can help in biodegradation of organic substances and thereby increasing the nutrient availability in the substrates (Krishnamoorthy and Narasimhan, 1994).

The addition of supplements *viz.,* horsegram flour to IFPg spawn alters the structure of the substrate and therefore its gas exchange ability. Higher O_2 and lower CO_2 levels were present in substrates with larger particles (Donoghue and Denison,1995) which stimulate and inhibit the mycelial growth (Donoghue and Denison, 1996). Ivan Henrique Rossi *et al.* (2003) showed the importance of O_2 supply during mycelial growth by obtaining higher lignocellulolytic enzyme activities during the first 20 days, a time during which the conc. of the element is higher.

The study has brought out a useful finding that ill-filled paddy grains can be used as a substrate for spawn preparation with supplementation. The IFPg spawn supplemented with horsegram flour at 4 per cent conc. was found to be better than the other treatments and will be inexpensive as compared to the conventional sorghum grain spawn.

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