EFFECT OF LINE AND PACKET SPACING ON GROWTH AND YIELD OF OYSTER MUSHROOM (*Pleurotus ostreatus*)

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CERTIFICATE

This is to certify that the thesis entitled, **EFFECT OF LINE AND PACKET SPACING ON GROWTH AND YIELD OF OYSTER MUSHROOM (*Pleurotus ostreatus*)"** submitted to the Faculty of **AGRICULTURE**, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE IN HORTICULTURE** embodies the result of a piece of bona fide research work carried out by **MD. SHAHIN SHAKAWAT, Registration No. 26276/00559** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that any help or source of information, received during the course of this investigation have been duly acknowledged.

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Supervisor

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Author

Effect of spacing on growth and yield of Oyster mushroom (*Pleurotus ostreatus*)

ABSTRACT

An experiment was conducted at National Mushroom Development and Extension center at Savar, Dhaka during the period from October 2006 to January 2007 with a view to determine the influence of line to line and packet spacing on growth yield and quality of mushroom. Four levels of line spacing viz. 14, 17, 20, 25cm and six levels of packet spacing viz. 10, 11, 13, 14, 17, 20 cm were taken as two factors of the experiment for the study. The line spacing showed significant influence on the mushroom growth and pileus shape and size. After that both mushroom growth and size of pileus did not show significant variation at days after opening. Number of primordia per packet was also significantly influenced by line and packet spacings. But the Number of effecting Fruiting body, Diameter of pileus, Thickness of pileus, Length of stalk, Diameter of stalk, Individual Fruiting body weight, yield per packet and per square meter yield were significantly influenced by the line and packet spacings. It was observed that wider line and packet spacings had better response on growth, and quality per packet while closer line and packet spacings showed higher yield per unit area. The interaction effect of line and packet spacing had significant influence on yield and yield parameters. The highest (11.7 kg or 11770 g) yield per unit area (sq.m) was recorded from the combination of 14cm line and 10cm packet spacing which also did not perform the highest benefit cost ratio. The highest (2.98) benefit cost ratio was found from spacing 20cm×11cm. While the lowest (1.91) BCR was obtained from the spacing of 25cm×20cm spacing. Considering the yield of mushroom per square meter, cost of production, benefit cost ratio and net return,the20cm×11cmspacin appeared to be recommendable for the cultivation of Oyester mushroom in Bangladesh.

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ACRONYMS

Abbreviation	Full words
NP	Number of primordia
NEFB	Number of effective fruiting body
DP	Diameter of pileus
TP	Thickness of pileus
LS	Length of stalk
DS	Diameter of stalk
DPI	Days to primordia initiation
DMRT	Duncan's Multiple Range Test
BCR	Benefit Cost Ratio
et al	Associate with others
EY	Economic Yield
BY	Biological Yield
PDA	Potato dextrose agar
NMDEC	National Mushroom Development and Extension Centre
MCH	Mushroom Culture House
PTC	Plant Tissue Culture
g	gram

DEDICATED

TO

MY PARENTS AND TEACHER

CHAPTER I
INTRODUCTION

CHAPTER I

INTRODUCTION

Mushrooms are large reproductive structures of edible fungi belonging to either Ascomycotina or Basidiomycotina Class. Since earliest times, mushrooms have been treated as a special kind of food. In recent years, it has occupied a very important place in man's dietary as a food item and has always been appreciated for its delicacy, nutritional value and medicinal properties. Mushrooms are recognized as the alternate source of good quality protein and are capable of producing the highest quantity of protein per unit area and time from the worthless agro-wastes (Chadha and Sharma, 1995). Compared to most other crops mushrooms contain 20-35% protein (dry weight basis) which is higher than vegetables and fruits. They are good source of protein vitamins and minerals (Khan *et al.* 1981) and contain about 85-95% water, 3% protein, (wet basis) 4% carbohydrates, 0.1% fats and 1% minerals and vitamins (Tewari, 1986). Mushroom protein quality is intermediate between that of animal and vegetable (Kurtzman 1974). Detrimental cholesterol is absent in mushroom but essential ergosterol is present (Chadha and Sharma, 1995). The present nutritional status of Bangladesh is a matter of great regrets concern. Because they produce large quantities in a short time (Gupta, 1986). Mushrooms have medicinal properties like anti-cancerous anti-cholesterol and anti-tumorous activities and useful against diabetic, ulcer and lung diseases (Quimio, 1976). In the era of healthy eating by cutting down calories, saturated fat and cholesterol, mushrooms are bound to attract the attention. Environmental factors (Air, temperature, relative humidity) play important role on mushroom production. The environmental factors of Bangladesh are suitable for the production of Oyster mushroom (*Pleurotus* spp.) A temperature range of 20-26°C and relative humidity of 70-90% is ideal for *Pleurotus sajor-caju* and fairly good crop can be obtained up to 30°C. Fresh air or in other hand oxygen is essential for the proper growth and development of Oyster mushroom .Mushroom cultivation is measured in terms of carbon dioxide concentration in the growing room which is a measure of difference, between carbon dioxide produced in the growing room and the carbon dioxide removed through ventilation. Relative humidity of the growing room should be ranged from 85-90 percent during spawn run.

The economy of Bangladesh is mainly agro based. Nearly all the arable areas of the country have been brought under plough and further agricultural expansion is almost impossible. On the other hand the unemployed population in our country is 2.2 million out of which 0.8 million is female (Anon., 2002). Mushrooms cultivation is labor intensive but land saving.It is also a short duration crop, and can be welcomed by the poor farmers of Bangladesh since it is a profitable business and does not require fertile land. But mushrooms have to be grown semi-controlled condition in a room. For the efficient use of space it is very important to determine the optimum spacing for mushroom cultivation.

Mushrooms produce huge amount of carbon dioxide which slowes the growth and twistes, distortes and stuntes the fruiting body (Nagaya and Kapoor, 1990) during cultivation and it is important to release the carbon dioxide from the surroundings of mushroom spawn packets. Light is an important factor for the development of the pinhead of the Oyster mushroom. It requires about 80 to 210 lux light intensity for maximum number of fruiting body per unit area. Mushroom emits heat during growing period and requires high relative humidity. These factors influenced by the line to line and packet to packet spacing of the Oyster mushroom (Alicbusan and santiago, 1975). Therefore, it is very important to know the effect of spacing to obtain the desired yield of mushroom. Since mushroom is a newly introduced crop in Bangladesh, the information regarding the influence of spacing on its growth and yield is very limited.

Considering the facts, the present study was undertaken with the following objective.

- To determine optimum line to line and packet to packet spacing for highest growth of Oyster mushroom in poly bag.
- To determine optimum line to line and packet to packet spacing for highest yield of Oyster mushroom in poly bag.

CHAPTER II
REVIEW OF LITERATURE

CHAPTER II

Review of literature

A number of literatures relating to different substrate and plant growth regulators on mushroom cultivation are available but literature on spacing of mushroom are not available. Literature review is given below based on the effects of different spacing on mushroom cultivation.

Effect of spacing on growth and yield

Gilbert (1988) observed that highest growth due to low density (space efficiency) and high ventilation has limited use for most growing during the spawn run. It may be more efficient to use more humid conditions during fruiting periods.

Uphoff (2004) reported that if the beds were appropriately laid out, they can become permanent with System of Rice Intensification (SRI) rice planted in two or three rows on the beds in the spring, after mushroom harvesting ends in April. This was an interesting no-till cultivation system. Since the soil was very rich and deep, placement of seedlings into the soil of mushroom beds were very easy. The recommended spacing was 40cm x 45 cm, which he thought at first would be too wide for best yield. He also mentioned that mushroom beds were so organically rich and fertile, that wide spacing worked fine.

Nagaya and Kapoor (1990) reported that high availability of water was necessary because mushrooms were approximately 90% water by weight. Disrupting the water supply can result in stunted, disproportioned, or aborted fruit bodies. Constant ventilation was necessary because mushroom produce carbon dioxide. Carbon dioxide is not vented or mixed with fresh air, thus the carbon dioxide, which is heavier than the other gases in the air, will accumulate at the bottom of the growing space around the cake culture and cause growth to be slowed and/or the fruit bodies to be twisted, distorted or stunted.

Alicbusan and Santiago (1967) mentioned that the convexed roof is better for air circulation at about 2.4 m of height from the floor to the rafter. Too ventilation with high covers properly screened to keep out insects. The length of the house was 7.5 m in length and it was 4.5 m wide. There were fine rows of one foot high cement blocks spaced 60 cm apart where 150 mushroom boxes could be accommodated. This type of growing house can be constructed singly or in a series of five or more units.

Alicbusan and Santiago (1975) reported that the planting were 5-8 cm from the edge of the straw and 5 cm apart. He also recommended during the dry season a four layer bed be used to counter lower relative humidity. Beds of six or more layer were possible in the wet season.

Adams (2000) observed that tube could be created using tubing up to 16 inches in diameter and 8 feet long. The next step is very important. Using a 16 unit, punch holes into the bag, spacing them about 15.24 cm apart.

Leatham (1982) mentioned that enough holes were drilled in the log to provide spacing about 500 cm sq. per hole.

Benefit cost ratio

Pungputinum (1995) conducted an experiment to know the general characteristics of the mushroom producer cost and return of each mushroom production as well as the production function and efficiency of input in producing Oyster mushroom, *Pleurotus* spp. collecting primary data from the crop year 1993, conducted by interviewing producers in 4 regions: Northern, Northeastern, Central and Southern through questionnaires. From the study of 122 samples, the result showed that most of the mushroom producers have approximately 5 years experience in producing mushrooms and fifty seven percent of the sample have been producing mushroom as their main career. There are 7 kinds of mushroom being produced: Oyster mushroom, *Pleurotus* spp, Ear mushroom balone mushroom, *Lentinus squarrouslus*, *Lentinus polychrous* and shiitake. These mushrooms can be cultured in plastic bag containing sawdust. An analysis of cost and return found that the average cost of producing mushroom per kg ranged from 5-8 baths and the producer gets the net profit ranging from 8-12 baths per kg depending on the kind of mushroom. The average cost of producing spore bag was 3 bath per bag and the net profit from selling spore bag ranged from 0.08-1.45 baht per bag. The estimation of Oyster mushroom, *Pleuribus sp*, production function by quadratic from revealed that there are only 2 input factors that have significant impact on production namely, sawdust and labour. The total elasticity of the production at 1.17 indicated the increasing returns to scale. To maximize profit from these two kinds of mushroom production, the increases in the use of sawdust and decreases in the use of labour must be adjusted to reach the optimum level.

Sarker et al (2004) studied the performances of different substrates and compared benefit cost ratio (BCR) of mushroom production. The highest BCR (6.50) was estimated when wheat straw was used as substrate followed by sugarcane bagasse (5.90), waste paper (5.65), rice straw (5.58) and kansh (5.25). The lowest BCR was obtained from water hyacinth (1.05).

CHAPTER III
MATERIAL AND METHODS

CHAPTER III

MATERIAL AND METHODS

The experiment was conducted in the Plant Tissues Culture (PTC) laboratory and the mushroom culture house of National Mushrooms Development Centre, Sobhanbug, Savar, Dhaka during the period of October 2006 to January 2007. Different steps involved in the implementation of the experiment are described below.

Maintaining Environmental condition

The temperature of the laboratory was maintained within the range of 20-22°C by air cooler. The temperature and relative humidity of the mushroom culture house was maintained by spraying water. The mean maximum temperature was 27°C and the mean minimum temperature was 18°C and maximum relative humidity was 90% and the mean minimum relative humidity was 41%. The mean maximum and minimum light intensities were 231 and 50 lux respectively. The environmental data: temperature, relative humidity and light intensity in culture house are given in Appendix-I.

Source of spawn collection

A spawn culture of *Pleurotus ostreotus* a widely cultivated species of Oyster mushroom in Bangladesh (Amin, 2002) was collected from the National Mushroom Development and Extension Centre (NAMDEC), Sobhanbug, Savar, Dhaka.

Preparation of pure culture

Pure culture of the selected mushroom fungus was prepared on Potato Dextrose Agar (PDA) media containing infusion of 200 g of peeled and sliced potato, 20 g of dextrose and 18 g of agar. The mixture was boiled on gas burner until the agar dissolved. Then the medium was poured into glass Petri dishes (90 mm diameter) of 15 ml/dish volume. The medium in Petri dish was sterilized in an autoclave for 15 minutes at 120±1°C under 1 kg/cm^2 pressure. After sterilization and solidification the plates were inoculated with tissue from full grown fruiting body of Oyster mushroom *(Pleurotus ostreatus)*. A fresh and full grown sporophore of Oyster mushroom was surface sterilized with 70% ethanol by rubbing cotton soaked in the alcohol. The stalk was peeled from the external side. Tissues were collected from the inner region of stalk of the sporophore. The tissues were cut into small pieces and placed on the solidified Petri dish containing PDA. After inoculation the PDA plates were covered with cellophane paper. All operations were done under sterile condition in a clean bench. The inoculated Petri dishes were transferred to a growth chamber maintaining temperature 27°C and incubated for 8 days. This 8 day old PDA culture was used for inoculation of mother culture.

Preparation of mother culture

The Mother culture was prepared by mixing sawdust and wheat bran at the ratio of 2:1. Calcium carbonate was used at the rate of 0.2% of the mixture. The moisture level of the mixture was maintained at 65% by adding tap water.

Polypropylene bags of 18 × 25 cm size were filled with 200 g of the above prepared mixture and packed tightly. The neck of the bag was made of heat resistant plastic tube. A hole of about 2/3 deep of the volume of the bag was made at the centre with a sharp end stick for space to put inoculums. The neck was plugged with cotton and covered with brown paper and tied with a placing rubber band. The packets were sterilized in an autoclave for one hour at 120°C under 1 kg/cm^2 pressure. After sterilization the packets were cooled for 24 hours and transferred into a clean bench. A piece of pure (PDA) culture medium containing mycelium of Oyster mushroom was placed aseptically in the hole of mother culture packet and again plugged the packet as mentioned before. Then the inoculated packets were placed on metal rack in the plant tissue culture laboratory at 25±2°C temperature for incubation. The medium of the mother culture was colonized by fungus as manifested by white colony growth mycelium within 15-16 days of inoculation. The fully colonized packets were used for spawning.

Preparation of spawn packets

Sawdust and wheat bran were used at the ratio of 2:1. Water was added to make the moisture content 65% and $CaCO_3$ was added at the rate of 0.2% of the total mixture. Polypropylene bags of 22.5 cm × 30 cm size were filled with 500 gm of substrate mixture. The procedure of packet preparation, plugging, sterilization and incubation were the same as mentioned under the section preparation of mother culture. Each spawn packet was inoculated with the mother culture at the rate of two teaspoonfuls per packet. After inoculation, the packets were incubated in the laboratory. Growth of mycelium in the spawn packets was completed within about 25 days.

Culture house activities

The brown paper, rubber bands, cotton plug and plastic neck of the spawn packets were removed and the mouths of polypropylene bags were wrapped and tied with rubber bands. Two ends, opposite to each other of the upper position of the plastic bag were opened by removing the plastic sheet with a scalpel making "D" shape opening. The opened surface of the substrate was scraped slightly with a teaspoon for removing the thin whitish mycelia layer. The spawn packets were soaked in water for 15 minutes and inverted for 15 minutes to remove excess water. The packers were placed on the floor culture house maintaining spacing of relevant treatment. The relative humidity and temperature of culture house were maintained at optimum level by watering thrice a daily. Light intensity and proper ventilation in culture house were maintained for fruiting body development.

Harvesting

The mushroom fruiting body was harvested by gentle twisting before the mushroom showed slightly curled edges. After first harvest the scraping and soaking were done again as during first opening of the packet for subsequent harvest. Scraping, soaking, remove excess water, packet placing and watering were done as mentioned before.

Data collection

Data were collected from 3 packets of each plot from the middle rows to avoid boarder effect following standard procedures as described hereafter. The following parameters were record

a. Days to primordial initiation (DPI)
b. No. of primordia (NP)
c. No. of effective fruiting body (NEFB)
d. Length of stipe (LS)
e. Diameter of stipe (DS)
f. Diameter of pileus (DP)
g. Thickness of pileus (TP)

Data calculation

Data were calculated from the randomly selected 3 packets of each unit area of the middle rows to avoid boarder effect following standard procedures and data also calculated from each unit area. The following parameters were record

a. Biological yield (BY)/packet
b. Economic yield (EY)/packet
c. Biological yield per square meter
d. Economic yield per square meter

No. of effective fruiting body: Number of well developed fruiting body was recorded. Dry and pinheaded fruiting body was discarded. But twisted and tiny fruiting body was included during counting.

Dimension of pileus and stalk: Thickness of the pili of four randomly selected fruiting bodies were recorded using a slide caliper. Diameter of pileus, length and diameter of stalk were recorded.

Biological yield: Biological yield, in g/500 packet, was recorded by weighing the whole cluster of fruiting body without removing the lower hard and dirty portion.

Economical yield: Economical yield in g/500 packet was recorded by weighting all the fruiting bodies in a packet after removing the lower hard and dirty portion.

Benefit cost ratio: The Benefit Cost Ratio (BCR) for different treatments were computed based on present market price of mushroom and cost of required packet used in the study/treatment.

Treatments

Factor A: Line to line distance (cm)

1. LD_1 :14cm
2. LD_2 :17cm
3. LD_3 :20cm
4. LD_4 :25cm

Factor B: Packet to packet distance (cm)

1. PD_1:10cm
2. PD_2:11cm
3. PD_3:13cm
4. PD_4:14cm
5. PD_5:17cm
6. PD_6:20cm

Design and layout of the experiment

The experiment was laid out in Randomized Complete Block Design (RCBD). The number of replication was 3 and the numbers of subplot were 72. Treatments were randomly placed on the floor of culture house. Dimension of each subplot was 1 m × 1 m. The unit plots and blocks were separated by a spacing of 0.2 m and 0.5 m respectively.

Statistical analysis

Data on various parameters were statistically analyzed by using the 'MSTAT-C' program. All the characters were subjected to analysis for variance (ANOVA). Means were compared using Duncan's Multiple Range Test (DMRT) (Gomez and Gomez 1984).

Economic analysis

The cost of production was analyzed in order to find out the profitability of the treatment combination. All the non material cost and interest on running capital were considered for computing the cost of production. The interest on running capital was calculated at the rate of 12% per year for three months. The rent of the house was considered as per place and the price of mushroom at harvest was considered as Tk. 140/kg. The benefit cost ratio was calculated as follows:

$$\text{Benefit cost ratio (BCR)} = \frac{\text{Gross return per unit area}}{\text{Total cost of production per unit area}}$$

CHAPTER IV
RESULTS AND DISCUSSION

CHAPTER IV

Results and discussion

The experiment was carried out to investigate the effect of the line to line and packet to packet distance on growth, yield and yield exponents of Oyster mushroom (*Pleurotus ostreatus)*. The results obtained in the study have been described and discussed in this section.

Days to primordia initiation

Days to primordia initiation (DPI) ranged from 2.78 to 4.11 at different spacing. The highest (4.11) days to primordial initiation was found from 25 cm×20 cm spacing. Days required for stimulation of primordia initiation were minimum (2.67 days) from the spacing 17 cm × 14 cm which was statically similar to sracing of 17 cm ×11cm and 14 cm ×11 cm (Table 1). This study showed that Days to primordia initiation (DPI) were higher at wider spacing and lower at lower spacing. This result is in agreement with the finding of Gilbert (1988) who also found the similar results, reported that wider spacing was more useful for primordial initiation.

Table 1. Effect of line to line and packet to packet distance on days to primordia initiation of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
	Days to primordia initiation			
PD_1	3.22 b_e	3.10 cde	3.67 abc	3.67 abc
PD_2	2.78 e	2.89 e	3.55 a_d	3.78 ab
PD_3	3.11 cde	3.00 de	3.77 ab	3.77 ab
PD_4	3.11 cde	2.66 e	3.55 a_d	3.66 abc
PD_5	3.00 de	3.11 cde	3.89 a	3.60 abc
PD_6	3.00 de	3.11 cde	3.77 ab	4.11a

Means within the column and rows, under a parameter, having a common letter do not differ significantly (P = 0.05).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Number of primordia/packet

The number of primordia/packet was significantly influenced by different line and packet spacing. The combined effect between line and packet spacing showed statistically significant difference on the number of primordial per packet and per unit area (sq.m) of space (Table 2). The maximum (66) number of primordia per packet was recorded from 17cm×10 cm spacing. The minimum (38.89) number of primordia per packet was recorded from the 14 cm×13 cm spacing (Table 2), which was statistically similar to 14 cm×10 cm, 14 cm ×14 cm, 14 cm ×17 cm, 14 cm × 20 cm, 20 cm ×11 cm, 20cm ×14 cm, 20 cm × 17 cm, 20cm × 20 cm, 25 cm × 10 cm, 25 cm × 11 cm, 25 cm × 13 cm, 25 cm × 14 cm, 25 cm × 17 cm, 25 cm ×20 cm spacing.

This study showed that the number of primordia per packet increased with the increased of spacing. But after certain limit of spacing it was decreased, because most fungi require adequate aeration for vegetative growth.Proper spacing is very important for the growth of fruting body. The result of the present study is similar to the finding of Gilbert (1988) who found that lower density was useful for fruiting body growth.It might be due to closer spacing may have an inhibitory role in number of primordial per packet.

Number of primordia per unit area of space

The number of primordia per unit area was statistically found to be influenced by the line to line and packet to packet distance. The combination of line to line and packet to packet spacing significantly influenced the number of primordia per unit area.

The highest number (3960) of primordial per plot was recorded from the 17cm ×10 cm and lowest (908.9) number of primordia per unit area was recorded from the 25cm ×20 cm spacing respectively (Table 2). The reason lies behind the fact that wider line to line and packet to packet wide spacing, which affected on micro environment of the surrounding of the packet which influenced the number of primordial of the packet. The result of the present study for this characters are in agreement with the findings of Gilbert (1988) who stated that number of primordia per unit area was decreased with the close spacing.

Table 2. Effect of line to line and packet to packet distance on the number of primordia per packet and per unit area of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
	Number of primordia per packet			
PD_1	39.67 gh	66.00 a	56.11 a-e	**48.33 c-h**
PD_2	42.89 efgh	54.56 a-f	47.33 d-h	41.22 fgh
PD_3	38.89 h	61.67 abc	60.89 abc	**44.00 e-h**
PD_4	48.67 c-h	57.89 a-d	50.11 c-h	46.67 d-h
PD_5	48.67 c-h	64.22 ab	46.78 d-h	**45.67 d-h**
PD_6	48.78 c-h	53 b-g	49.33 c- h	45.44 d-h
	Number of primordia per unit area(sq. m)			
PD_1	2777 bcd	3960 a	2806 bc	1933 f-i
PD_2	2702 bcd	2946 b	2130 efg	**1484 i-l**
PD_3	2178 efg	2960 b	2436 cde	1408 jkl
PD_4	2385 c-f	2431 cde	1754 g-j	**1307 j-m**
PD_5	2044 e-h	2312 def	1403 jkl	1096 lm
PD_6	1707 g-k	1590 h-k	1233 klm	**908.9 m**

Means within the column and rows, under a parameter, having a common letter do not differ significantly (P = 0.05).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Number of effective fruiting body per packet and per square meter

The number of fruiting body per packet was counted at different spacing and the data were analyzed statistically. The results showed that there was significant variation due to line to line and packet to distance. The maximum (41.00) number of effective fruiting body per packet was recorded from the line spacing 17 cm and packet spacing 10 cm, which was statistically similar to 17 cm×11 cm and 17cm × 13 cm, 14 cm × 20 cm spacing. The minimum number of effective fruiting body was recorded from 14 cm × 13 cm spacing (Table 3). The number of fruiting body increased with the increase of spacing up to 17 cm×10 cm and decreased there after. The combined effect of line to line and packet to packet distance on the number of effective fruiting body was significant. The widest and the closest spacing have inhibitory role on NEFB per packet.

Fruiting body per unit area of space

The interaction effect between line to line and packet to packet spacing significantly influenced on number of effective fruiting body per unit area . The highest (2460) number of fruiting body per plot was recorded from the 17cm × 10 cm spacing and minimum number (548.9) of fruiting body per plot was recorded from the 25cm ×20 cm spacing which was the widest spacing among all (Table3). The result of the present study for this character is in agreement with the findings of Gilbert (1988) who stated that low distance and high ventilation are more useful for the growth of fruiting body.

Table 3. Effect of line to line and packet to packet distance on the number of fruiting body per packet and per unit area of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
	Number of fruiting body per packet			
PD_1	29.89 c-g	41.00 a	33.22 b-g	28.44 d-g
PD_2	31.67 b-g	35.22 abc	30.22 c-g	29.89 c-g
PD_3	27.22 g	36.67 ab	33.89 b-f	30.22 c-g
PD_4	33.00 b-g	35.33 bc	29.33 c-g	29.89 c-g
PD_5	34.44 bcd	34.22 b-e	29.22 c-g	327.89 efg
PD_6	35.33 abc	30.00 c-g	30.00 c-g	27.44 fg
	Number of fruiting body per unit area (sq. m)			
PD_1	2092 b	2460 a	1661 de	1138 hij
PD_2	1995 b	1902 bc	1360 fgh	1076 ijk
PD_3	1524 def	1760 cd	1356 fgh	967.10 jkl
PD_4	1617 de	1484 ef	1027 ijk	836. 90 klm
PD_5	1447 efg	11232 ghi	876.70 klm	669.30 mn
PD_6	1237 ghi	900 j-m	750 lmn	548.90 n

Means within the column and rows, under a parameter, having a common letter do not differ significantly ($P = 0.05$).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Main effect of line and packet spacing on biological and economic yield per packet and per unit area

Significant variation in per packet yield of Oyster mushroom was observed due to line and packet spacing. From line spacing the highest (194 g) biological yield per packet was obtained from the 20 cm line spacings and the closest line spacing 14 cm produced the lowest (180 g/packet) yield (Table 4).Between packet spacing the highest biological yield (191 g) per packet was recorded from 13 cm packet spacing which was statistically identical to all other packet spacing except 10 cm. Almost similar trend was observed in case of economic yield per packet.

The highest (9427 g) biological yield per sq. m. was observed in the closest line spacing 14 cm which was significantly higher as compare to other line spacings. An inverse relationship was observed between the line spacing and biological yield per plot. In case of packet spacing a similar trend was observed. The minimum (5116 g/plot) yield was recorded in highest 20cm packet spacing which was statistically different from all other packet spacing. Exactly similar trend was observed in the case of economic yield per unit area (Table 4).

Table 4. Main effect of line and packet spacing on biological and economic yield of Oyster mushroom (*Pleurotus ostreatus*)

Line spacing	Biological yield g/packet	Biological yield g/unit area	Economic yield g/packet	Economic yield g/unit area
LD_1	180 c	9427 a	166 d	8560 a
LD_2	189 b	8532 b	177 b	8032 b
LD_3	194 a	7307 c	182 a	6846 c
LD_4	186 b	5598 d	173 c	5220 d
Packet spacing				
PD_1	183 b	10013 a	171 b	9221 a
PD_2	190 a	9395b	175 ab	8638 b
PD_3	191 a	8399 c	189 a	7884 c
PD_4	186 ab	7165 d	177 ab	6805 d
PD_5	187 ab	6209 e	173 b	5719 e
PD_6	186 ab	5116 f	171 b	4729 f

Means within the column and rows, under a parameter, having a common letter do not differ significantly (P = 0.05).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Interaction effect on Biological yield per packet and per unit area

The combined effect of line to line and packet to packet spacing had also significant influence on biological yield per packet (Table 5). The highest (203 g) yield per packet was obtained from 20cm × 13 cm spacing which was statistically similar to 14 cm×17cm, 17 cm×11 cm, 17 cm×13 cm, 20 cmx10 cm, 20 cm×11 cm, 25 cm×13 cm, and 25 cm×14 cm spacing. The lowest (167 g) yield was obtained from the closest spacing 14 cm ×10 cm which was significantly lower than all other spacing except 14 cm × 14 cm spacing. The result of this study was in agreement with the findings of Uphoff (2004) up to certain limit who obtained high yield with the higher spacing of 45 cm ×40 cm in case of soil culture. The combination of widest 25cm line and packet 20cm spacing produced lowest (182.30 g) yield per packet. From the study, it is clear that the highest biological yield per packet was obtained from 20 cm×13 cm spacing and the yield decreased with the increased or deceased of spacing. The lower yield in closer spacing may be due to the insufficient light and oxygen and excessive CO_2. On the other hand the lower yield at higher spacing may be due to lack of optimum moisture level in the surrounding the packets.

Biological yield per unit area

There was a significant influence of line to line and packet to packet distance on yield of mushroom per plot (sq.m). It was observed that with the increase of line to line and packet to packet spacing, the yield of mushroom decreased gradually. The maximum (11770 gm) yield of mushroom was obtained from 14 cm ×10 cm spacing. The widest line to line and packet to packet spacing 25 cm × 20 cm produced the lowest amount of mushroom (3647g per unit area) (Table 5). It was observed that the yield of oyster mushroom per unit area was inversely related to spacing. The higher yield of fruiting body was mainly contributed by the higher packet population per unit area in closer spacing. There was a great variation in the packet population of the treatment combination. It was observed that there were 70 packets were required for the spacing combination of 14 cm×10 cm which was 3.5 times higher (number packets) than the widest 25 cm×20 cm spacing required (number of packet 20).

Table 5. Effect of line to line and packet to packet distance on the biological yield (g) per packet and per unit area of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
		Biological yield per packet		
PD_1	167 e	187 bcd	192 abc	184 cd
PD_2	183 cd	192 abc	198 ab	185 bcd
PD_3	180 cd	193 abc	203 a	190 a-d
PD_4	178 de	189 bcd	189 bcd	190 a-d
PD_5	190 a-d	186 bcd	188 bcd	186 bcd
PD_6	183 cd	185 bcd	193 abc	182 cd
		Biological yield per unit area (sq. m)		
PD_1	11770 a	11270 a	9639 cd	7373 h
PD_2	11550 a	10410 b	8940 ef	6680 i
PD_3	10090 bc	9280 de	8124 g	6101 jk
PD_4	8738 f	7966 g	6638 i	5320l m
PD_5	8003 g	6708 i	5660 kl	4467 n
PD_6	6417 ij	5560 ij	4842 mn	3647 o

Means within the column and rows, under a parameter, having a common letter do not differ significantly ($P = 0.05$).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Interaction effect of economic yield per packet and per unit area

The interaction effect of line to line and packet to packet was significantly influenced on the yield mushroom. The highest (189.1 g) yield was recorded per packet from the spacing 20 cm × 13 cm. The lowest (154.40g) yield was obtained the spacing 14 cm×10 cm which was statistically similar 14 cm×11 cm and 25 cm × 20 cm spacing. The result of this study was in agreement with the findings of Uphoff (2004) who obtained the highest yield from the wider spacing 45 cm×40 cm incase of soil.

Economical yield per unit area

It was observed that the yield of mushroom per unit area was inversely related to the packet to packet and line to line spacing. The interaction effects between line to line and packet to packet spacing also showed significant influence and that was statistically identical to all other line to line and packet to packet spacing (Table 6).

The combination of 17cm × 10 cm line to line and packet to packet spacing produced higher (10610 g) yield. The line and packet spacing might have ensured the availability of uniform space in all directions essential for proper growth and development of above parts of the packet which also provided the available light, moisture, and relative humidity and thus favored better growth of the mushroom producing more number of bigger fruiting body and ultimately resulting better yield in this spacing. The lowest (3300 g) yield was obtained from the widest 25 cm×20 cm spacing

On the other hand widest line and packet spacing might have ensured more aeration, which could have helped to dry away moisture from the packet and surroundings. This unfavorable condition ultimately hampered the yield.

Table 6. Effect of line to line and packet to packet distance on the economic yield (g) per packet and per unit area of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
	Economical yield per packet			
PD_1	154.40 h	176.90a-f	180.20 a-e	176.10 b-f
PD_2	162.30 gh	181.10ab	184.40 abc	173.60 b-g
PD_3	168.00 efg	185.60ab	189.10 a	176.90 a-f
PD_4	171.90 c-g	180.00 a-e	180.90 a-d	175.30 b-f
PD_5	170.00 d-g	172.20 c-g	179.20 a-e	173.30 b-g
PD_6	171.00d-g	171.20 d-g	179.90 a-e	165 .00 fgh
	Economical yield per unit area (sq. m)			
PD_1	10180 ab	10610 a	9011 d	7044 g
PD_2	10230 ab	9780 bc	8300 f	6248 h
PD_3	9408 cd	8907 de	7564 g	5660 ij
PD_4	8423 ef	7560 g	6331 h	4909 kl
PD_5	7140 g	6200 h	5377 jk	4160 m
PD_6	5985 hi	5137 k	4497 lm	3300 n

Means within the column and rows, under a parameter, having a common letter do not differ significantly (P = 0.05).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Stalk length

There was significant variation in the stalk length of oyster mushroom due to line to line and packet to packet spacing. The longest (2.467 cm) stalk was obtained from the from the 14 cm×13 cm spacing which was statistically similar to 14 cm×14 cm, 17 cm×10 cm, 17 cm×13 cm, 17 cm×20 cm, 20 cm×13 cm, spacing. The lowest (1.653 cm) stalk length was observed in 25cm × 20 cm spacing which was statistically identical to all other spacing combination except 14 cm × 13 cm (Table 7). The result of the present study showed that the stalk length decreased with the increase or decrease of spacing.

Stalk diameter

Diameter of stalk was influenced by the spacing combination. The highest (0.7633 cm) stalk diameter was observed from the 14cm ×11cm spacing which was statistically identical to all other spacing combination except 17 cm ×11cm spacing combination. The lowest (0.6467 cm) stalk diameter was found from the 17 cm × 11 cm, spacing which also statistically identical to all other spacing combination except 14 cm ×11 cm, spacing combination (Table 7).

Table 7. Effect of line to line and packet to packet distance on the length and diameter of stalk (cm) of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
		Length of stalk		
PD_1	1.717 fg	2.110 a-f	1.927 d-g	2.033 b-g
PD_2	2.293 a-d	2.020 b-g	2.037 b-g	1.953 c-g
PD_3	2.467 a	2.343 abc	2.123 a-e	1.787 e-g
PD_4	2.373 ab	1.953 d-g	1.867 e-g	1.890 e-g
PD_5	1.937 d-g	1.950 c-g	1.900 d-g	1.797 e-g
PD_6	1.777 e-g	2.107 a-f	1.713 fg	1.653 g
		Diameter of stalk		
PD_1	0.7200 ab	0.6800 ab	0.6667 ab	0.6867 ab
PD_2	0.7633 a	0.6467 b	0.6867 ab	0.6933 ab
PD_3	0.7033 ab	0.6667 ab	0.6967 ab	0.7100 ab
PD_4	0.6900 ab	0.6967 ab	0.6600 ab	0.7467 ab
PD_5	0.6933 ab	0.6667 ab	0.6400 b	0.6667 ab
PD_6	0.7100 ab	0.6833 ab	0.6667 ab	0.6833 ab

Means within the column and rows, under a parameter, having a common letter do not differ significantly (P = 0.05).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm. Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Effect of line and packet spacing on diameter of pelius

There were no significant variation was observed in the diameter of pileus (Table 8).

Effect of line and packet spacing on thickness of the pileus

A significant variation in the thickness of pileus was observed due to line to line and packet to packet spacing (Table 8). The highest (5.10 mm) thickness was observed from 14 cm × 20 cm spacing which was statistically identical to all other spacing combination except 20 cm ×10 cm. The lowest (4.40 mm) thickness was recorded from the spacing at 20 cm × 10 cm.

Table 8. Effect of line to line and packet to packet distance on the diameter and thickness of pileus of Oyster mushroom (*Pleurotus ostreatus*)

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
		Diameter of pileus (cm)		
PD_1	5.54 a	5.39 a	5.56 a	5.66 a
PD_2	5.51 a	5.79 a	5.49 a	5.64a
PD_3	5.63 a	5.46 a	5.71 a	5.44 a
PD_4	5.47 a	5.56 a	5.52 a	5.77 a
PD_5	5.31 a	5.49 a	5.42 a	5.64 a
PD_6	6.08 a	5.47 a	5.62 a	5.51 a
		Thickness of pileus (mm)		
PD_1	4.60 ab	4.63 ab	4.40 b	4.80 ab
PD_2	4.83 ab	4.93 ab	4.80 ab	4.56 ab
PD_3	4.86 ab	4.63 ab	4.86 ab	4.70 ab
PD_4	4.97 ab	4.90 ab	4.80 ab	5.03 ab
PD_5	4.96 ab	4.46 ab	4.56 ab	4.63 ab
PD_6	5.10 a	4.50 ab	4.66 ab	5.00 ab

Means within the column and rows, under a parameter, having a common letter do not differ significantly (P = 0.05).

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.
Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

Economic analysis

The economic analysis was done with a view to compare the cost and benefit under different line and packet spacing. For this purpose the input cost for house rent, price of spawn, management cost, watering and interest of capital were considered against the treatment combination. It was observed that there was a great variation in the cost of production due to different treatment combinations. Because greater area was needed (Table 9). The benefit came only from selling the mushroom. The price of spent substrate was ignored. The maximum (2.98) benefit cost ratio was obtained from 20 cm × 11 cm spacing while the minimum (1.91) was found from the widest spacing at 25 cm×20 cm. Considering the yield of mushroom per unit area cost of production and benefit cost ratio, spacing combination 20 cm × 11 cm could be recommended for cultivation of Oyster mushroom (*Pleurotus ostreatus*).

Table 9. Effect of line to line and packet to packet distance on Benefit cost ratio of Oyster mushroom (*Pleurotus ostreatus)*

Packet to packet distance (cm)	Line to line distance (cm)			
	LD_1	LD_2	LD_3	LD_4
	Benefit cost ratio			
PD_1	2.10	2.51	2.78	2.36
PD_2	2.32	2.54	2.98	2.29
PD_3	2.36	2.56	2.54	2.28
PD_4	2.38	2.44	2.37	2.20
PD_5	2.30	2.27	2.28	2.10
PD_6	2.25	2.18	2.20	1.91

Details in Appendix III

Line to line distance (cm) LD_1 :14cm, LD_2 :17cm, LD_3 :20cm and LD_4 :25cm.

Packet to packet distance (cm) PD_1:10cm, PD_2:11cm, PD_3:13cm, PD_4:14cm, PD_5:17cm and PD_6:20cm

CHAPTER V
SUMMARY AND CONCLUSION

CHAPTER V

Summary and Conclusion

The experiment was conducted at the National Mushroom Development and Extension Center, Sobhanbagh, Savar, Dhaka during the period of October 2006 to January 2007 with Oyster mushroom (*Pleurotus ostreatus*) to standardize optimum line to line and packet to packet spacing for obtaining best yield of the mushroom.

There were two factors in this experiment namely, line spacing and packet spacing .The levels of line spacing were 14, 17, 20, 25 cm and the levels of packet spacing were 10, 11, 13, 14, 17, 20 cm. Thus there were 24 combinations of line and packet spacing. The experiment was laid out in the Randomized Complete Block Design (RCBD) with three replications. The unit plot size was 1m ×1m. Packets were laid on 29 October 2006 on the floor of culture house as per the treatments. Data were collected from three randomly selected packet of each unit area. Data were collected on days to primordia initation, number of primordia per packet, primordia per unit area , number of fruiting body per packet, No of fruiting body per unit area, biological yield per packet, biological yield per unit area, economical yield per packet, economical yield per unit area, length and diameter of stalk (cm) and thickness, diameter of pelius (cm). The collected data were analyzed by computer following MSTATC programme, and the means were separated by DMRT.

Effect of spacing was significant on growth, yield and yield components. It was observed from the results that significant increase in the growth parameters and yield per packet was obtained with the increase of spacing up to certain limit and then decreased gradually.

The highest (189.10 g) economic yield per packet was obtained from 20cm × 13 cm spacing where as the highest (10610 g) economic yield per unit area was obtained from 17 cm × 10 cm spacing. But highest (2.98) benefit cost ratio was obtained from 20 cm ×11 cm spacing.

Considering the yield, cost of production per unit area, net return per unit area and benefit cost ratio the spacing 20 cm × 11 cm was the best for Oyster mushroom cultivation on floor.

Further studies are needed to find out the optimum line and packet spacing for the production of different Oyster mushroom species on different steps and height of racks of culture house.

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CHAPTER VII

APPENDIXES

Appendix I. Environmental data during growing period of Oyester mushroom

Month	Decades	Temperature		Relative Humidity		Light Intensity	
		Maximum	Minimum	Maximum	Minimum	Maximum	Minimum
November'06	1*	27.7	24.02	81.8	71.3	183.2	43.5
	2	27.36	22.24	88.2	64.36	231.4	48.3
	3	25.15	20.84	85.7	43.8	198	60
December'06	1	25.00	18.15	87.1	51.3	185.37	56.54
	2	24.90	18	86.5	41.07	196.42	51.53
	3	24.22	17.8	89.5	48.4	176.41	29.97
January'07	1	20.88	20	88.15	61.45	179.14	41.74
	2	23.45	21	86.35	50.25	186.21	37.59

1* stands for day 01 to 10, 2 for day 11 to 20 and 3 for day 21 to rest of the month.

Appendix II. Disease and insect pest infestation data during growing period of Oyster mushroom.

Treatment	Plot No	Number of insect pest infested packet	Number of disease infected packet
14 X 10	1	6	30
14 X 11	2	2	24
14 X 13	3	2	18
14 X 14	4	-	13
14 X 17	5	1	12
14 X 20	6	-	4
17 X 10	7	1	16
17 X 11	8	-	12
17 X 13	9	-	14
17 X 14	10	-	8
17 X 17	11	1	9
17 X 20	12	-	8
20 X 10	13	1	17
20 X 11	14	2	11
20 X 13	15	2	12
20 X 14	16	-	3
20 X 17	17	-	5
20 X 20	18	-	4
25 X 10	19	-	9
25 X 11	20	-	6
25 X 13	21	-	4
25 X 14	22	-	3
25 X 17	23	-	2
25 X 20	24	-	-

Appendix III. Benefit cost ratio of oyester mushroom production at different spacings

Treatment	No. spawn per plot	Unit price of spawn TK.	Total price of spawn Tk.	Rent of room (1 sq.m) Tk.	Watering cost	Scrap. Harvest. Proces Market	Total invested capital	Inter. of capital (3% for 3 mon)	Total cost (Tk)	Economicyield kg/pl	Unit price of mushroom	Total price of mushroom/ plot	BCR
14 X 10	70	8.00	420.00	20.00	45.00	175.00	660.00	19.80	679.80	10.18	140.00	1425.20	2.10
14 X 11	63	8.00	378.00	20.00	45.00	157.00	600.00	18.00	618.00	10.23	140.00	1432.20	2.32
14 X 13	56	8.00	336.00	20.00	45.00	140.00	541.00	16.23	557.23	9.40	140.00	1316.00	2.36
14 X 14	49	8.00	294.00	20.00	45.00	122.00	481.00	14.43	495.43	8.42	140.00	1178.80	2.38
14 X 17	42	8.00	252.00	20.00	45.00	105.00	422.00	12.66	434.66	7.14	140.00	999.60	2.30
14 X 20	35	8.00	210.00	20.00	45.00	87.00	362.00	10.86	372.86	5.99	140.00	838.60	2.25
17 X 10	60	8.00	360.00	20.00	45.00	150.00	575.00	17.25	592.25	10.61	140.00	1485.40	2.51
17 X 11	54	8.00	324.00	20.00	45.00	135.00	524.00	15.72	539.72	9.78	140.00	1369.20	2.54
17 X 13	48	8.00	288.00	20.00	45.00	120.00	473.00	14.19	487.19	8.90	140.00	1246.00	2.56
17 X 14	42	8.00	252.00	20.00	45.00	105.00	422.00	12.66	434.66	7.56	140.00	1058.40	2.44
17 X 17	36	8.00	216.00	20.00	45.00	90.00	371.00	11.13	382.13	6.20	140.00	868.00	2.27
17 X 20	30	8.00	180.00	20.00	45.00	75.00	320.00	9.60	329.60	5.13	140.00	718.20	2.18
20 X 10	50	8.00	250.00	20.00	45.00	125.00	440.00	13.20	453.20	9.01	140.00	1261.40	2.78
20 X 11	45	8.00	202.00	20.00	45.00	112.00	379.00	11.37	390.37	8.30	140.00	1162.00	2.98
20 X 13	40	8.00	240.00	20.00	45.00	100.00	405.00	12.15	417.15	7.56	140.00	1058.40	2.54
20 X 14	35	8.00	210.00	20.00	45.00	87.50	362.50	10.88	373.38	6.33	140.00	886.20	2.37
20 X 17	30	8.00	180.00	20.00	45.00	75.00	320.00	9.60	329.60	5.37	140.00	751.80	2.28
20 X 20	25	8.00	150.00	20.00	45.00	62.50	277.50	8.33	285.83	4.49	140.00	628.60	2.20
25 X 10	40	8.00	240.00	20.00	45.00	100.00	405.00	12.15	417.15	7.04	140.00	985.60	2.36
25 X 11	36	8.00	216.00	20.00	45.00	90.00	371.00	11.13	382.13	6.24	140.00	873.60	2.29
25 X 13	32	8.00	192.00	20.00	45.00	80.00	337.00	10.11	347.11	5.66	140.00	792.40	2.28
25 X 14	28	8.00	168.00	20.00	45.00	70.00	303.00	9.09	312.09	4.90	140.00	686.00	2.20
25 X 17	24	8.00	144.00	20.00	45.00	60.00	269.00	8.07	277.07	4.16	140.00	582.40	2.10
25 X 20	20	8.00	120.00	20.00	45.00	50.00	235.00	7.05	242.05	3.30	140.00	462.00	1.91

Appendix IV. Experimental view