

## MORPHOLOGICAL AND PHENOLOGICAL VARIABILITY OF *Echinochloa* ACCESSIONS AND THEIR HERBICIDE SENSITIVITY

S. M. Masum<sup>1</sup>, M. Hasanuzzaman<sup>2</sup>, M. Malek<sup>3</sup>, M. J. Jalal<sup>4</sup> and M. A. Razzaque<sup>5</sup>

### ABSTRACT

*Echinochloa* species are the most problematic grass weeds in rice fields worldwide. The species of this genus have high intra- and interspecific variability, with many ecotypes. Based on this, the study was conducted to characterize the morphology and phenology of Bangladesh *Echinochloa* accessions and evaluate their herbicide sensitivity. The results showed a distinct variability of the growth characteristics among *Echinochloa* ecotypes. The four *Echinochloa* accessions, designated *Echinochloa crus-galli* ecotypes A, B, C, and D, were discovered in the various rice-growing regions of Bangladesh. The same ecotypes were treated with pre-and post-emergence herbicides in pot culture and the puddle and dry sowing conditions to determine the herbicide sensitivity. Among the pre-emergence herbicides, all except butachlor in dry sowing conditions and pyrazosulfuron ethyl in both puddle and dry sowing conditions were effective against ecotypes of *Echinochloa* detected. Bispyribac sodium and metamifop were effective in both the four-leaf and eight-leaf stages of *E. crus-galli* ecotype D. In contrast, fenoxaprop-p-ethyl was effective at the four-leaf stage. *E. crus-galli* ecotype B was effectively controlled by quizalofop-p-ethyl when sprayed at the eight-leaf stage and fenoxaprop-p-ethyl at the four-leaf stage. *E. crus-galli* ecotype C was less susceptible to all the herbicides. However, quizalofop-p-ethyl and fenoxaprop-p-ethyl, although effective at the four-leaf stage in reducing the weed persistence index (WPI). This information helps make weed management decisions and is informative in understanding the speciation and adaptation of weedy *Echinochloa* species.

**Keywords:** *Echinochloa* accessions, morphology, pre-emergence herbicides, post-emergence herbicides, weed persistence index

### INTRODUCTION

Knowledge on the basic biology of weeds is essential for developing sustainable weed management systems. *Echinochloa* is a genus belonging to Poaceae of about 50 weed species with a worldwide dispersion. It has been reported to be a serious weed in 36 crops (Holm *et al.*, 1991), particularly in rice, where its similar habit and appearance make it difficult to distinguish when young. *Echinochloa* species are self-pollinating, but gene flow by wind-mediated outcrossing can occur (Bagavathiannan *et al.*, 2012). Correctly identifying *Echinochloa* species is challenging because its morphological characteristics tend to intergrade. The widely distributed *E. crus-galli* is mainly known for its high phenotypic plasticity (Maun and Barrett, 1986; Masum *et al.*, 2016). As a result, synonyms were misclassified and used incorrectly at the species and intraspecies levels. (Michael, 2003). Numerous species of *Echinochloa* exhibit weed-like characteristics, including prolific seed generation, protracted emergence durations due to dormancy, photoperiod insensitivity, and rapid vegetative growth. *Echinochloa* is a C<sub>4</sub> plant that utilizes carbon dioxide more effectively than C<sub>3</sub> plants like rice and wheat. This characteristic, and various weedy traits, have made *Echinochloa* a great competitor in crop production systems in both flooded (e.g., rice) and non-flooded (e.g., soybean, maize, various others) fields (Bagavathiannan *et al.*, 2011). Consequently, proper identification of (sub) species is a prerequisite for optimization of weed control comprising an appropriate choice of herbicide and dose tailored to the weeds present. Therefore, a better knowledge of the distribution of variability within the genus *Echinochloa* would help in its taxonomic classification and in designing a rational strategy for herbicide testing.

Biological invasion severely threatens agro-biodiversity, ecological stability, and agricultural sustainability. Recently, herbicide resistance has become a conundrum that is haunting ecosystem

---

<sup>1&2</sup> Associate Professor, Dept. of Agronomy, <sup>3</sup> Deputy Chief Farm Superintendent, Farm Division, <sup>4</sup> Professor, Dept. of Agricultural Chemistry, Sher-e-Bangla Agricultural University, Dhaka-1207, <sup>5</sup> Crop Protection, Padma Oil Company Limited

stability, weed management, and sustainable crop production (Heap, 1999). Therefore, herbicide resistance in weeds necessitates the continued monitoring of herbicide resistance evolution in weed species (Asaduzzaman *et al.*, 2022). Nowadays, *E. crus-galli* is included among the most dangerous herbicide-resistant weeds in the world (Beckie and Reboud, 2009). It shows morphologic variability and differences in response to herbicides (Claerhout *et al.*, 2015). It has evolved both target-site and non-target-site herbicide resistance, due to which a quest for alternative herbicides is also in danger (Norsworthy *et al.*, 2014). Besides good management practices to reduce resistance evolution, the investigation into biological and ecological mechanisms is much needed. Various biotypes of *E. crus-galli* are now resistant to multiple herbicides (Wilson *et al.*, 2014; Masum *et al.*, 2019). Moreover, the continuous use of the same herbicides with a similar mode of action has evolved resistance in *E. crus-galli*. Undoubtedly, herbicide resistance in *E. crus-galli* is a severe threat to sustainable crop production in a large area globally.

Despite being one of the world's most significant warm-season annual grass weeds, *E. crus-galli*, relatively few researchers have examined the population plasticity of its growth and development and responses to herbicides. These studies are infrequent in Bangladesh. Therefore, these investigations aimed to obtain specific information on the growth and development of *E. crus-galli*, creating an emerging concern towards herbicide resistance. This knowledge may help develop more effective control methods for this weedy grass.

## MATERIALS AND METHODS

### Seed source

Existing *Echinochloa* spp. of Sher-e-Bangla Agricultural University (SAU) farm and farmer's field in Bangladesh were used as test species. A field survey was conducted in rice fields of SAU farm and different locations in Bangladesh to identify different *Echinochloa* spp.

### Growing of plants

*Echinochloa* accessions were sown in trays and grown under a plastic rain shelter. The soil was shielded from weed growth by geotextile membranes. After 18 days, plants at the four-leaf stage were planted outside in pots in a completely randomized block design with three replications. The experimental unit (pot) consisted of five plants of a given accession.

Manual labor was used to eradicate the remaining other weeds. All plants were screened for qualitative and quantitative parameters regarding vegetative and generative traits.

### Morphological and phenological observation

Growth traits such as plant type, plant height, number of tillers, flag leaf length and width, panicle length, spikelet weight, fresh weight of 100-seeds, and panicle awn were assessed at the maturity stages. Plant type (prostrateness, Fig. 1) and panicle awn were considered as qualitative characters visually rated on a scale from none (0), moderate (1) to severe (2) (Park *et al.*, 1995; Yamasue, 1997). In addition, phenological events such as emergence date, heading time, and growth duration were also evaluated.

### Whole-plant herbicide resistance assay

A pot culture study was conducted from March to June 2021 under a plastic rain shelter at SAU's Agronomy field laboratory. Plastic pots (50 cm x 40 cm) were used for the experiment, and 3/4th of the pots were filled with soil collected from the field. The soil was silty clay loam (sand 26%, silt 45%, and clay 29%) with pH 5.8, C 0.45%, N 0.12%,  $\text{HPO}_4^{2-}$  0.40 mg g<sup>-1</sup> soil, and K<sup>+</sup> 0.70 mg g<sup>-1</sup> soil. Each pot was sown with 25 seeds of *Echinochloa* spp. After seed germination, five seedlings were maintained per pot. The seeds were pretreated with 1M ethanol under darkness for three days to improve germination - a modification of the procedure suggested by Masum *et al.* (2016). A completely randomized design was adopted for experiments, and separate experiments were conducted for the different *Echinochloa* types with three replications for each treatment. The ecotypes of *Echinochloa* were treated with pre-emergence herbicide with a dose of 0.75 kg a.i. ha<sup>-1</sup> of pretilachlor, 1.25 kg a.i. ha<sup>-1</sup> of pyrazosulfuron ethyl, 1.5 kg a.i. ha<sup>-1</sup> of pendimethalin, (0.18+ 0.40) kg a.i. ha<sup>-1</sup> of (bensulfuron

methyl + pretilachlor), (0.04 + 0.14) kg a.i. ha<sup>-1</sup> of (bensulfuron methyl + acetachlor), 1.25 kg a.i. ha<sup>-1</sup> of butachlor, 0.10 kg a.i. ha<sup>-1</sup> of oxadiazon, (0.0015 + 0.06) kg a.i. ha<sup>-1</sup> of (pyrazosulfuron-ethyl + pretilachlor), along with untreated control. All the herbicides were applied in both puddle sowing and dry sowing conditions. Post-emergence herbicides were applied at four-leaf and eight-leaf stages of *Echinochloa* spp. and treated with 0.12 kg a.i. ha<sup>-1</sup> of quizalofop-p-ethyl, 0.024 kg a.i. ha<sup>-1</sup> of carfentrazone-ethyl, 0.025 kg a.i. ha<sup>-1</sup> of bispyribac sodium, 0.06 kg a.i. ha<sup>-1</sup> of fenoxaprop-p-ethyl, 0.015 kg a.i. ha<sup>-1</sup> of ethoxysulfuron, 0.125 kg a.i. ha<sup>-1</sup> of metamifop and untreated control. The quantity of herbicide was calculated based on the recommendation and the pot's diameter. Six ml of the solution was then carefully sprayed in each pot using a hand sprayer. The efficacy of pre-emergence herbicides was studied under both dry sowing and puddle sowing conditions. In dry sowing conditions, *Echinochloa* seeds were sown in pots filled with soil and irrigated lightly without flooding, simulating rainfall. Herbicides were sprayed on the day after sowing. In puddle sowing conditions, soil in the pots was puddled by hand, and excess water was removed after settling the clay. Twenty-five seeds were sown on the top layer and, after one day, again flooded to a depth of three centimeters. Water was drained after one day, and herbicides were sprayed on the third day after sowing. Flooding was again done after 24 hours and continued for the duration of the study.

Weed control efficiency (WCE) was calculated based on the number of surviving weed seedlings in each pot (Mani *et al.*, 1973) by using the following equation-

WCE =  $\frac{x-y}{x} \times 100$ , where x = Number of *Echinochloa* seedlings in unweeded control and y = Number of *Echinochloa* seedlings in the treated plot.

The efficacy of each herbicide was determined on the basis of seedling survival percentage and weed persistence index (WPI). WPI was estimated using the following formula of Mishra and Misra (1987).

WPI =  $\frac{(p/q) \times (b/a)}$ , where p = Dry weight of weeds in the treated plot, q = Dry weight of weeds in the control plot, a = Weed count in the treated plot, and b = Weed count in the control plot.

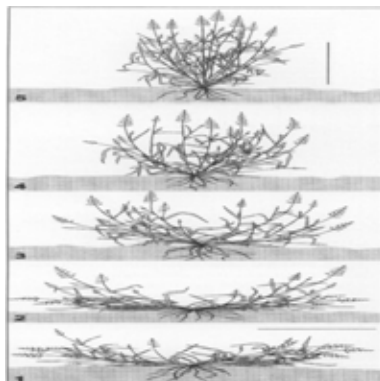
WPI indicates the degree of resistance of the plant that survived the herbicide and is expressed in terms of dry matter production. The data on seedling survival percentage and weed persistence index were transformed by angular transformation to normalize their distribution.

The statistical software 'STATIX 10' was used to analyze the data, and the means were compared based on the least significant difference (LSD) at a 0.05 level of significance.

## RESULTS AND DISCUSSION

### Identification of *Echinochloa crus-galli* accessions

Different types of *E. crus-galli* were collected from different locations based on their prostrate/erect habit (Fig. 1), having awn, and seed size. The growth form of individual *E. crus-galli* plants in the study population ranged from almost prostrate to upright.



**Fig. 1.** The collected ecotypes of *Echinochloa crus-galli* were selected as of the growth patterns displayed by Norris, 1996

Within the population, variation in prostrateness has been observed for *E. crus-galli* accession. According to Fig. 1, awn and seed size, *E. crus-galli* accessions are classified into four types (Fig. 2).

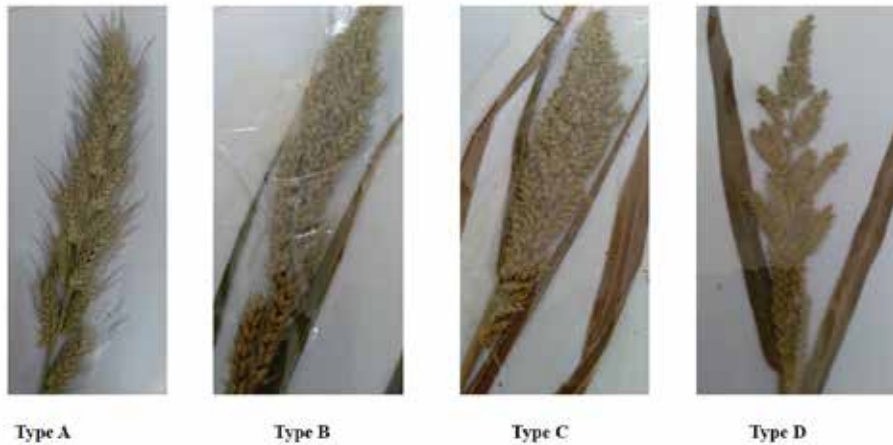


Fig. 2. Spikes of different ecotypes designated Type A, Type B, Type C, and Type D of *Echinochloa crus-galli* are detected in Bangladesh.

#### Variability in growth characteristics among *Echinochloa* ecotypes

Variability in growth characteristics among *Echinochloa* ecotypes is presented in Table 1. Results of the growth characteristics among *Echinochloa* ecotypes collected from different rice-growing areas of Bangladesh showed distinct variability. The seedling emergence period varies significantly among species, showing a wide range of days to emergence (Table 1). The range of emergence was from 3-7 days, and heading time was from 41 to 59 days. When the growth duration ranged from 86 days to 98 days. The diversity in emergence behavior across *Echinochloa* populations presents a big problem for weed management. These species can emerge across a wide window of time, allowing many to escape weed management tactics (Scursoni *et al.*, 2007). In general, growth characteristics variability among *Echinochloa* ecotypes might be affected by geographic origin. This observation was in line with the work of Johnson *et al.* (1989). They reported that the ecotype of some Sweetvetch (*Hedysarum boreale* Nutt.) differed in their taxonomic distance values, and the difference is not always closely related to specific characteristics of the collection sites. Widespread species tend to have more genetic variability than close relations with narrow distribution (Maki, 1999). Damalas *et al.* (2008) studied the morphological variation among 12 populations of *Echinochloa* species in the same environment in Greece.

Plant height differed across species. Type D was the shortest species compared to other species and produced significantly lower tillers than other species, but *Echinochloa* were equal producers of tillers. The high tillering capability allows *Echinochloa* ecotypes to produce many panicles (and seeds), contributing to its abundance. Competition drastically reduces the number of tillers in rice fields—generally less than ten productive tillers plant<sup>-1</sup> (Zhang *et al.*, 2021). However, with gaps in crop rows and field edges, *Echinochloa* species can quickly reach their tillering potential and maximum seed production. *Echinochloa* in this study produced fewer tillers, with high variability in tillering than *Echinochloa* in Greece (115-131 tillers) (Damalas *et al.*, 2008).

*E. crus-galli* type C had the longest flag leaf among the three species, significantly longer than other ecotypes. Big flag leaves provide a competitive advantage because of the light interception's large surface area and higher photosynthesis capacity. A big plant such as *E. crus-galli* type C needs to fix a large amount of carbon to develop a large plant canopy and biomass. Also, big flag leaves afford a higher capability to shade adjacent plants and gain a competitive advantage (Weiner and Thomas, 1986). Panicle lengths were significantly different among species. *E. crus-galli* type C had the longest

panicles, followed by *E. crus-galli* type B, and *E. crus-galli* type D. *E. crus-galli* type A had the shortest panicles at 12.83 cm (Table 1). The estimated total seed output plant<sup>-1</sup> differed significantly between species after accounting for the total number of panicles. *E. crus-galli* type C produced the highest amount of seed (533 seeds panicle<sup>-1</sup>), and *E. crus-galli* type A produced the lowest (240 seeds panicle<sup>-1</sup>). However, the *E. crus-galli* type A and B were found awns. The awn length depended on its habitat's relative humidity (Maun and Barrett, 1986). Thus, the awn length of *E. crus-galli* type A (21 mm) varied significantly, while *E. crus-galli* type B had 10 mm awns (Table 1). With such a considerable variation in this trait, one can only associate the most obvious generalities to a species, such as being predominantly awnless, predominantly long-awned, or having long awns only on apical spikelets in a panicle.

The spikelet characteristics (spikelet length and width) of the three species were significantly different. *E. crus-galli* type C had the longest (4.10 mm) and widest (2.13 mm) spikelets. There was a wide range of spikelet sizes among *Echinochloa* accessions in many plant studies (Gould *et al.*, 1972; Michael, 2003; Tabacchi *et al.*, 2006). As expected, greater root-to-foilage ratios were found for *E. crus-galli* type C plants.

**Table 1. Growth characteristics of collected *Echinochloa crus-galli* accessions**

<i>E. crus-galli</i> accessions	Emergence (days)	Average length of tiller (cm)	Tillers plant <sup>-1</sup> (Nos.)	Heading Time (Days)	Flag leaf length (cm)	Flag leaf width (cm)	Panicle length (cm)	Spikelet length (mm)	Spikelet width (mm)	Seeds panicle <sup>-1</sup>	Length of awn (mm)	Growth duration (Days)	Root foliage ratio
Type A	7 a	82.33 c	50.33 a	59.33 a	17.17 bc	1.13	12.83 b	2.63 b	1.40 ab	240.00 c	21 a	95.67 a	0.14 bc
Type B	3 b	130.33 a	44.00 a	41.67 b	23.40 ab	1.33	17.70 ab	3.53 a	1.70 ab	353.33 b	10 b	86.00 b	0.18 b
Type C	7 a	108.67 b	44.67 a	52.67 b	27.67 a	1.27	20.23 a	4.10 a	2.13 a	533.33 a	-	98.00 a	0.23 a
Type D	5 b	37.33 d	29.00 b	47.00 c	13.30 c	1.23	14.57 ab	2.50 b	1.37 b	205.00 c	-	93.33 a	0.11 c
SE(±)	0.75	6.761	4.055	1.73	2.94	0.13	2.88	0.31	0.19	43.58	0.57	2.29	0.01
CV%	4.23	15.59	9.35	3.99	17.67	13.56	6.66	1.72	1.45	19.53	1.33	5.30	12.06

In a column means having similar letter(s) are statistically similar, and those having dissimilar letter(s) differ significantly at a 0.05 level of probability.

### Herbicides susceptibility

In pots treated with pretilachlor, pendimethalin, bensulfuron methyl + pretilachlor, bensulfuron methyl + acetachlor, oxadiazon, and pyrazosulfuron-ethyl + pretilachlor, there was no seed germination under either puddle sowing or dry sowing conditions. These pre-emergence herbicides were thus 100% efficient in controlling all four types of *Echinochloa*. However, seedling emergence was observed in pots treated with butachlor in dry sown condition, whereas in puddle condition, no seedling was found. In the case of pyrazosulfuron-ethyl, seedling emergence was observed in three types (A, B, and C) of *Echinochloa*, whereas there was no germination of types D. So, butachlor could be applied in stagnant water, and pyrazosulfuron-ethyl is recommended to be used 6-9 days after sowing, which could be the reason for their poor performance in the present study where they were applied in dry condition and 3-6 days earlier, respectively. Plants grown in dry sown conditions were more susceptible to pyrazosulfuron-ethyl than those in wet sowing conditions. The susceptibility to pyrazosulfuron-ethyl was in the order *E. crus-galli* type D > *E. crus-galli* type C > *E. crus-galli* type A > *E. crus-galli* type B (Table 2). Post-emergence herbicides had varying effects on the different types of *Echinochloa*. Although herbicides that were known to be highly effective against the weed were evaluated, they were less effective in pot culture studies. The data on WPI are presented in Table 3.

Application of bispyribac sodium at the eight-leaf stage was highly effective against *E. crus-galli* type A as there was no seedling survival. Bispyribac sodium has also been reported to be effective in rice nurseries and the main field against *E. crus-galli* (Duary and Mukherjee, 2013). At the four-leaf stage, the survival seedlings' persistence was low (0.04). The survival percentage of the weed was less when quizalofop-p-ethyl, carfentrazone-ethyl, fenoxaprop-p-ethyl, and metamifop were applied at the four-leaf stage. At the eight-leaf stage, these herbicides performed poorly, resulting in high dry matter

**Table 2. Efficacy of butachlor and pyrazosulfuron-ethyl on *Echinochloa* spp.**

Ecotype of <i>Echinochloa</i>	Treatments	Dry sowing conditions		Puddle sowing conditions	
		Emergence (%)	WCE (%)	Emergence (%)	WCE (%)
Type A	butachlor	18.67	61.86	-	-
	Control	50.67	61.86	-	-
	pyrazosulfuron ethyl	24.00	63.55	26.67	55.51
	Control	54.66	63.55	62.67	55.51
Type B	butachlor	8.00	87.29	-	-
	Control	61.33	87.29	-	-
	pyrazosulfuron ethyl	22.67	71.99	21.33	66.75
	Control	60.00	71.99	66.67	66.75
Type C	butachlor	12.00	79.80	-	-
	Control	60.00	79.80	-	-
	pyrazosulfuron ethyl	20.00	66.87	28.00	48.73
	Control	58.67	66.87	54.67	48.73
Type D	butachlor	30.67	46.95	-	-
	Control	60.00	46.95	-	-
	pyrazosulfuron ethyl	0.00	78.611	42.67	24.49
	Control	58.67	78.611	57.33	24.49

**Table 3. Effect of post-emergence herbicides on Weed Persistence Index (WPI) of *Echinochloa* spp.**

Treatments	Dose (kg a.i. ha <sup>-1</sup> )	<i>Echinochloa</i> type A		<i>Echinochloa</i> type B		<i>Echinochloa</i> type C		<i>Echinochloa</i> type D	
		Four leaf stage	Eight leaf stage	Four leaf stage	Eight leaf stage	Four leaf stage	Eight leaf stage	Four leaf stage	Eight leaf stage
Quizalofop-p-ethylc	0.12	0.05	0.27	0.11	0.00	0.44	0.67	0.00	0.21
Carfentrazone-ethyl	0.024	0.06	0.86	0.41	0.65	0.69	0.92	0.81	0.80
Bispyribac sodium	0.025	0.04	0.00	0.37	0.34	0.41	0.54	0.00	0.42
Fenoxaprop-p-ethyl	0.06	0.02	0.64	0.08	0.27	0.19	0.53	0.09	0.94
Ethoxysulfuron	0.015	0.09	0.86	0.18	0.92	0.54	0.65	0.60	0.67
Metamifop	0.125	0.18	0.45	0.00	0.15	0.48	0.46	0.00	0.17

production by the surviving seedlings. At the eight-leaf stage, persistence (0.86) was higher when carfentrazone-ethyl was applied. Though an earlier application produced better results, the effect was seen to be inadequate, pointing to a likely need for either a higher dose of application or a still earlier application. Sharma *et al.* (2004) recommended a dose of 0.09 kg ha<sup>-1</sup> to control *Echinochloa* in the nursery. It was suggested that a higher amount is required for suppressing *Echinochloa* in an advanced stage. The time of application also has to be precise, as a slight variation in the age of the weed seedlings can render a herbicide ineffective. The application of metamifop killed off *E. crus-galli* type B at the four-leaf stage or quizalofop-p-ethylc at the eight-leaf stage.

Contrary to its effect on *E. crus-galli* type A and *E. crus-galli* type D, bispyribac sodium was seen to be ineffective against *E. crus-galli* type B at both leaf stages of application. Earlier application of bispyribac sodium was impractical in suppressing most of the seedlings, while at the later stage, the persistence of the surviving seedlings was high. Fenoxaprop-p-ethyl was a suitable herbicide for *E. crus-galli* type B at the four-leaf stage, as the seedling persistence was only 0.08. A similar result was reported by Singh *et al.* (2004). Chauhan and Abugho (2012) observed less effective control of *E. crus-*

*galli* when fenoxaprop + ethoxy sulfuron (150 + 18 g ha<sup>-1</sup>) was sprayed at the eight-leaf stage. In contrast, the weed control efficiency at the four-leaf stage was 68%. In the present experiment, the surviving seedlings showed high persistence at the eight-leaf stage.

Differences in anatomical, physiological, and biochemical traits may help make weeds best adapted to applied herbicides. *E. crus-galli* type C was significantly different from the other species of *Echinochloa* studied in its response to herbicides. Bispyribac sodium was less effective against this species at the four-leaf and eight-leaf stages, and the surviving seedlings were highly persistent. Azmi (2002) observed that certain weed species might establish when a particular herbicide is continuously used due to inherent properties or sublethal concentrations, leading to resistance. This may lead to changes in the weed spectrum and composition and distribution.

For *Echinochloa* type D, the application of quizalofop-p-ethyl and fenoxaprop-p-ethyl at the four-leaf stage was less effective. Fenoxaprop-p-ethyl performed better at the eight-leaf stage of the weed, suggesting that application at this stage would be more effective. Bispyribac sodium and metamifop were effective at both stages of the weed, indicating their superiority in controlling *Echinochloa* type A. Chauhan and Abugho (2012) reported that post-emergence application of bispyribac sodium at four-leaf stage reduced the biomass of *Echinochloa* up to 95%. Walia *et al.* (2008) noted that bispyribac sodium (0.4 kg ha<sup>-1</sup>) could significantly reduce *Echinochloa*'s biomass accumulation when applied 30 days after sowing. There was no seedling survival when quizalofop-p-ethyl, bispyribac sodium, and metamifop were used at the four-leaf stage. Ethoxysulfuron was the least effective among the herbicides applied as the seedling survival was 100%, and the surviving plants were highly persistent.

Detailed knowledge of how individual weeds grow and reproduce may improve the accuracy of predicting population dynamics and crop losses due to competition and may point to ways to improve control strategies. This comprehensive study of a single *Echinochloa* population emphasizes the extreme variability within a population. The findings imply that *Echinochloa crus-galli* ecotypes have developed and adapted in Bangladesh. Morphological variations among *E. crus-galli* ecotypes in Bangladesh are not very high. The ecotype variations will affect the successful management of *Echinochloa crus-galli* using a chemical or potential biocontrol agent. However, molecular traits analysis would be helpful to characterize the level of genetic variability among *E. crus-galli* ecotypes. Suppose new strains (biotypes) of the *Echinochloa* grass migrates from one to another regions and cross-pollinate. In that case, the genetic diversity may increase, and the control of *E. crus-galli* in rice production may become more complex.

## REFERENCES

- Asaduzzaman, M., Koetz, E., Wu, H. and Shephard, A. 2022. Paraquat resistance and hormetic response observed in *Conyza sumatrensis* (Retz.) E. Walker (tall fleabane) in Australian cotton cropping systems. *Phytoparasitica*, 50: 269–279.
- Azmi, M. 2002. Impact of continuous direct seeding rice culture on weed species diversity in the Malaysian rice ecosystems. Proc. Symposium on Environment and Nature Research, 10-11 April 2002. Kuala Lumpur. Malaysia.
- Bagavathiannan, M.V., Norsworthy, J.K., Smith, K.L. and Burgos, N. 2011. Seedbank size and emergence pattern of barnyardgrass (*Echinochloa crus-galli*) in Arkansas. *Weed Sci.*, 59: 359-365.
- Bagavathiannan, M.V., Norsworthy, J.K., Smith, K.L. and Neve, P. 2012. Seed production of barnyardgrass (*Echinochloa crus-galli* L.) in response to time of emergence in cotton and rice. *J. Agric. Sci.*, 150: 717-724.
- Beckie, H.J. and Reboud, X. 2009. Selecting for weed resistance: Herbicide rotation and mixture. *Weed Technol.*, 23: 363–370.
- Chauhan, B.S and Abugho, S.B. 2012. Effect of growth stage on the efficacy of post emergence herbicides on four weed species of direct-seeded rice. *Sci. World J.*, 2012: 1-7.

- Claerhout, S., Dewaele, K., De Riek, J., Reheul, D. and De Cauwer, B. 2015. Morphological and genetic variability of local *Echinochloa* accessions and the link with herbicide sensitivity. *Weed Res.*, 56: 137–148.
- Damalas, C.A., Dhima, K.V. and Eleftherohorinos, I.G. 2008. Morphological and physiological variation among species of the genus *Echinochloa* in northern Greece. *Weed Sci.*, 56: 416-423.
- Duary, B. and Mukherjee A. 2013. Distribution pattern of predominant weeds of wet season and their management in West Bengal, India. Proc. 24<sup>th</sup> Asian-Pacific Weed Science Society Conf. 191-199.
- Gould, F.W., Ali, M.A. and Fairbrothers, D.E. 1972. A revision of *Echinochloa* in the United States. *Am. Midl. Nat.*, 87: 36-59.
- Heap, I.M. 1999. International survey of herbicide-resistant weeds: lessons and limitations. Proc. Int. conf. 3: 769-776.
- Holm, L.G., Plunknett, D.L., Pancho, J.V. and Herberger, J.P. 1991. The World's Worst Weeds. Distribution and Biology. Krieger Pub., Malabar, Florida, 609pp.
- Johnson D.A., Ford, T.M.J., Rumbaugh, M.D. and Richardson, B.Z. 1989. Morphological and physiological variation among ecotypes of Sweetvetch (*Hedysarum horeale* Nutt). *J. Range Manag.*, 42(6): 496-501.
- Maki, M. 1999. Genetic diversity in the threatened insular endemic pert *Asterasa-grayi* (Asteraceae). *Plant Syst. Evol.*, 217: 1-11.
- Mani, V.S., Malla, M.L., Gautham, K.C., and Bhagwandas. 1973. Weed killing chemicals in potato cultivation. *Indian Farming*. 22: 17-18.
- Masum, S.M., Hossain, M.A., Akamine, H., Sakagami, J.I. and Bowmik, P.C. 2016. Allelopathic potential of indigenous Bangladeshi rice varieties. *Weed Biol. Manag.*, 16: 119-131.
- Masum, S.M., Hossain, M.A., Akamine, H., Sakagami, J.I., Ishii, T., Konno, T., Nakamura, I. Assaduzzaman, M and Bowmik, P.C. 2019. Performance of Bangladesh indigenous rice in a weed infested field and separation of allelopathy from resource competition. *Weed Biol. Manag.*, 19: 39-50.
- Maun, M.A. and Barrett, S.C.H. 1986. The biology of Canadian weeds. *Echinochloa crus-galli* L. Beauv. *Can. J. Plant Sci.*, 66: 739-759.
- Michael, P.W. 2003. *Echinochloa* P beauv. P. 390-403. In: M.E. Barkworth *et al.* (ed.). Flora of North America North of Mexico, vol. 25. Oxford University Press, New York.
- Mishra, M. and Misra, A. 1997. Estimation of IPM index in jute: a new approach. *Indian J. Weed Sci.*, 29: 39-42.
- Noris, R.F. 1996. Morphological and phenological variation in barnyardgrass (*Echinochloa crus-galli*) in California. *Weed Sci.*, 44(4): 804-814.
- Norsworthy, J.K., Wilson, M.J., Scott, R.C. and Gbur, E.E. 2014. Herbicidal activity on acetolactate synthase-resistant barnyardgrass (*Echinochloa crus-galli*) in Arkansas, USA. *Weed Biol. Manag.*, 14: 50-58.
- Park S.J., Kim K.U. and Shin, D.H. 1995. Systematical classification of *Echinochloa* species. Proc. 16<sup>th</sup> Asian-Pacific Weed Sci. Conf. 756-758.
- Scursioni, J.A., Forcella, F. and Gunsolus, J. 2007. Weed escapes and delayed weed emergence in glyphosate-resistant soybean. *Crop Prot.*, 26: 212-218.
- Sharma, S.D., Punia, S.S., Malik, R.K. and Narwal, S. 2004. Efficacy of cyhalofop butyl against weeds in rice nursery. *Indian J. Weed Sci.*, 36: 18-83.
- Singh, V.P., Govindra, S. and Mahendra, S. 2004. Effect of fenoxaprop-p-ethyl on transplanted rice and associated weeds. *Indian J. Weed Sci.*, 36: 190-192.
- Tabacchi, M., Mantegazza, R., Spada, A. and Ferrero, A. 2006. Morphological traits and molecular markers for classification of *Echinochloa* species from Italian rice fields. *Weed Sci.*, 54:1086-1093.

- Walia, U.S., Bhullar, M.S., Shelly, N. and Walia, S.S. 2008. Control of complex weed flora of dry-seeded rice (*Oryza sativa*) with pre-and post-emergence herbicides. *Indian J. Weed Sci.*, 40: 161-164.
- Weiner, J. and Thomas, S.C. 1986. Size variability and competition in plant monocultures. *Oikos*, 47: 211-222.
- Wilson, M.J., Norsworthy, J.K., Scott, R.C. and Gbur, E.E. 2014. Program approaches to control herbicide-resistant barnyardgrass (*Echinochloa crus-galli*) in Midsouthern United States rice. *Weed Technol.*, 28: 39-46.
- Yamasue, Y. 1997. Biometric analysis of *Echinochloa* weed complex. Proc. 16<sup>th</sup> Asian-pacific Weed Sci. Society Conf. 233-237.
- Zhang, Z., Cao, J., Gu, T., Yang, X., Peng, Q., Bai, L. and Li, Y. 2021. Co-planted barnyardgrass reduces rice yield by inhibiting plant above- and belowground-growth during post-heading stages. *Crop J.*, 9: 1198-1207.

## PREVALENCE AND RISK FACTORS ASSOCIATED WITH CLINICAL CASES OF PET ANIMALS IN DHAKA CITY, BANGLADESH

M. M. Hossain<sup>1</sup>, M. Islam<sup>2</sup>, N. Nazmunahar<sup>3</sup>, M. R. Islam<sup>4</sup> and A. N. M. I. Rahman<sup>5</sup>

### ABSTRACT

This study aimed to determine the prevalence of most common clinical cases of dog, cat and rabbit reported at MM Hossain's Pet Clinic, Dhaka, Bangladesh. One thousand and two hundred (n=1200) clinical cases were recorded during the period from March 2020 to July 2022. Among them, dog, cat, and rabbit covered 21%, 65.5% and 12.5% cases, respectively. Parasitic disease was the highest prevalent case (25%) in dogs followed by gastritis (11.51%), wound (10.71%), dermatitis (7.54%), conjunctivitis (6.75%), and kennel cough (5.16%) ( $p<0.001$ ). Likewise, parasitic disease followed by wound, viral disease, and diarrhea were the more prevalent clinical cases in cats (23.31%, 18.05%, 6.77%, and 5.64%, respectively) ( $p<0.001$ ). In case of rabbit, traumatic injuries and fracture was the highest reported case (16.67%) followed by parasitic infestation, diarrhea, and dermatitis (13.33%, 13.33%, and 12.00%, respectively) ( $p<0.001$ ). Clinical cases were more prevalent in male dogs than female dogs (56.75% vs 43.25%) ( $p<0.05$ ), and dogs more than 6 months of age had more reported clinical cases compared to 1-6 months of aged dogs (65.08% vs 34.92%) ( $p<0.05$ ). Local breed of cats showed higher prevalence of clinical cases than that of cross breed (66.54% vs 33.46%), male cats had higher prevalence compared to female cats (59.40% vs 40.60%), and cats at the age of 1-6 months represented higher prevalence than those more than 6 months of age (62.16% vs 37.84%) ( $p<0.05$ ). Male rabbits represented as higher reported clinical cases than female rabbits (72.67% vs 27.33%) ( $p<0.05$ ). Overall, Parasitic infection/infestation was the prominent reported clinical cases in pet dogs, cats, and rabbits. Proper prevention and therapeutic management should be carrying out to reduce the prevalence of parasitic diseases of pets.

**Key words:** clinical cases, pet animals, prevalence, risk factors

### INTRODUCTION

Day by day rearing pets is becoming very popular in Bangladesh. People care about caring for pets like dogs, cats and rabbits for passing their free time (Hossain and Kayesh, 2014). The tendency to keep companion animals is increasing daily among the people of modern society. Pets have become an essential part of the family, and are repeatedly considered as family members (Yadav *et al.*, 2017). In many households, pet animals contribute to the physical, social and mental well-being of children and their owners (Dohoo *et al.*, 1998; Robertson *et al.*, 2000; Parvez *et al.*, 2014). Dogs are serving as a companion and act as a workforce (Singh *et al.*, 2014). Dogs and cats are the most frequent household pets around the world, but there are also many other vertebrates that share our household environment (Chomel *et al.*, 1992). Pets keeping is usually related with certain responsibilities like housing, disease management and responsible for pet ownership with negative consequences for public health when neglected (William *et al.*, 2002). Pets some times are responsible for the transmission of zoonotic pathogens to human (Dada *et al.*, 1979; Robertson *et al.*, 2000; Molyneux, 2004). Since 19<sup>th</sup> century, rabbits became as pets in western nations. Recently, rabbit makes a mutual relationship with its owners through better understanding.

Several clinical diseases frequently affect pets. Among them, the viral diseases such as rabies, infectious canine hepatitis, canine distemper, canine parvovirus infections, feline Panleukemia, and feline calici viral infections are very common in the Indian sub-continent like Bangladesh (Biswas *et al.*, 1996; Samad, 2011). Bacterial diseases such as leptospirosis, brucellosis, kennel cough, clostridial infection etc are more common in pets. Furthermore, household pets are playing an important role in transmitting zoonotic diseases (Kornblatt and Schantz 1980; Plant *et al.*, 1996; Geffray, 1999).

---

<sup>1</sup>Associate Professor, <sup>3</sup>MS student, Dept. of Anatomy, Histology and Physiology, <sup>2</sup>Associate Professor, Dept. of Microbiology and Parasitology, <sup>4</sup>Associate Professor, Dept. of Surgery and Theriogenology, <sup>5</sup>Associate Professor, Dept. of Animal, Nutrition, Genetics and Breeding, Sher-e-Bangla Agricultural University, Dhaka-1207

Therefore, clinical diseases of pets especially in Dhaka city, the highest number of pets rearing city, is needed to investigate. Though the incidence of clinical diseases and conditions of pet animals has already been performed in different areas of Bangladesh (Tarafder and Samad, 2010; Mahmud *et al.*, 2014; Parvez *et al.*, 2014; Yadav *et al.*, 2017). However, the prevalence of clinical diseases in pet animals in Dhaka city is less documented. It was reported earlier that the overall prevalence of clinical diseases in the Dhaka city area in dogs, cats and rabbits were 71.50%, 18.70% and 9.80% respectively (Runa *et al.*, 2016). However, the risk factors associated with the clinical cases of pets were not understood well. Therefore, this study conducted to know the prevalence of clinical diseases of pet animals in Dhaka city, Bangladesh along with the assessment of risk factors affected systems of the body and the etiology of pet diseases.

## MATERIALS AND METHODS

### Research Area and Duration

The study was carried out in Dr. MM Hossain's Pet Clinic at Dhaka city from the period of March 2020 to July 2022. A total of 1200 cases were examined during the study period out of them 252 were dogs, 798 were cats and 150 were rabbits. The clinical examinations of diseased animals were performed on basis of complaints of owners of patients, history of clinical diseases and clinical examinations of patients.

### Data Collection

The data were collected according to the owner's complaints, visual examinations like body condition, behavior, gesture, posture, skin lesions, salivation, nasal and ocular discharge, distension of the abdomen, locomotor disturbances etc.

### Determination of Age of Animals

The age of individual animals was determined by interviewing the Owners of the animals and the examination of teeth of animals.

### Methods of diagnosis of diseases

Based on the history from owners, physical examination, clinical signs and clinical examination of animals, the presumptive diagnosis of different diseases or clinical cases were performed.

### Study Design

Clinical cages of three different groups of animals such as dogs, cats and rabbits were noted down.

### Clinical examination of patient:

Distant inspection started with the observation of the general attitude of the patient such as attentiveness, dullness, depression, and anorexia attentively inspected. After that, normal and abnormal posture and gait examined according to the condition of the dog, cat, and rabbit. Close inspection of the animals performed by the visual examination. Separation of hairs, light palpation and close direct inspection was performed to detect hair, coat and skin abnormalities. Skin lesions, nature of lesions such as foul odorous discharge, crusts, scale and dandruff, location and distribution of those lesions also performed. In addition, external parasites like ticks, lice, fleas, flies and larvae of flies identified during the close inspection.

### Physical examination

For the identification of wounds first of all visual inspection was performed and then categorizing the wound whether it septic, lacerated, incised, punctured, perforating, abrasions or hematoma for a more precise diagnosis. In required cases, needle punctures were also conducted. Visual inspection of pulse rate, respiration and rectal temperature was recorded and then different organs and systems of the body of animals were performed by using palpation, percussion and auscultation. Mouth gag and local anaesthesia were also used for the clinical examination of the animals. Extension and flexion, and needle puncture were also performed when required.

### Laboratory diagnosis

Faecal samples and skin scrapings were examined with a compound microscope at the department of Microbiology and Parasitology of Sher-e-Bangla Agricultural University (SAU), Dhaka. Collected

blood and urine samples were tested at the department of Anatomy, Histology and Physiology of SAU, Dhaka for routine and specific diagnosis. X-Ray or imaging was performed at Teaching and Training Pet Hospital and Research Centre at Purbachal, Dhaka to diagnose the musculoskeletal and chest diseases in required cases.

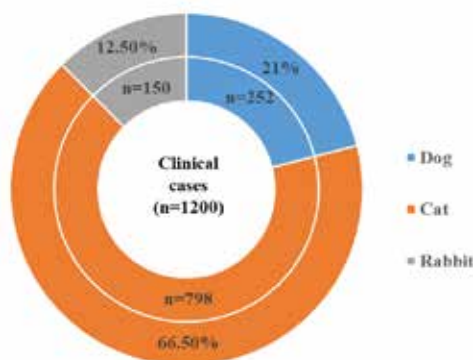
#### Statistical analysis

The data collected from each patient were entered into MS Excel (Microsoft office excel-2013, USA). All data obtained from this study were analyzed by Pearson's Chi-square test using the Minitab17 program (Minitab Ltd., UK). Significant differences were declared as  $p < 0.05$ .

## RESULTS AND DISCUSSION

### Prevalence of various clinical cases in pet dogs, cats and rabbits

Twelve hundred (1200) cases of different clinical cases were recorded in Dr. MM Hossain's Pet Clinic in Dhaka city, Bangladesh during the period from March 2020 to July 2022. The overall prevalence of clinical cases was 21.00%, 66.50% and 12.50% in dogs, cats and rabbits, respectively (Fig. 1). A similar study was conducted by Yadav *et al.* (2017), who reported that the overall proportional prevalence of clinical diseases and disease conditions in dogs, cats and rabbits were 44.32%, 34.28% and 21.39% respectively. Sarker *et al.* (2015) further reported that the overall proportional prevalence of clinical diseases in dogs and cats was 75.00% and 25.00%, respectively. The variation of finding among different studies might be due to other study areas and periods.



In this study, parasitic disease was the highest prevalent case (25%) in dogs followed by gastritis (11.51%), wound (10.71%), dermatitis (7.54%), conjunctivitis (6.75%), and kennel cough (5.16%) ( $p < 0.001$ ) (Table 1). Likewise, parasitic disease followed by wound, viral disease, and diarrhea were the more prevalent clinical cases in cats (23.31%, 18.05%, 6.77%, and 5.64%, respectively) ( $p < 0.001$ ) (Table 2). In case of rabbit, traumatic injuries and fracture was the highest reported case (16.67%) followed by parasitic infestation, diarrhoea, and dermatitis (13.33%, 13.33%, and 12.00%, respectively) ( $p < 0.001$ ) (Table 3). The findings of this study is in agreement with Parvez *et al.* (2014) who reported more parasitic cases in dogs (51.54%) and cats (54%). Yadav *et al.* (2017) further reported the parasitic diseases as the most frequent clinical cases in dog (24.42%) and cat (23.31%), while traumatic injuries and fracture was higher in rabbits (16.87%). In contrast, Sarker *et al.* (2015) reported low prevalence of parasitic diseases, which were 14.77% in dogs and 13.33% in cats. The reason might be due to performing de-worming reported by Sarker *et al.* (2015).

**Table 1. Occurrence of clinical cases of pet dogs according to their breed, sex and age.**

Cases	Overall			Breed				Sex				Age						
	No.	%	p-value	Local		Cross		p-value	Male		Female		p-value	>6 Months		p-value		
				No.	%	No.	%		No.	%	No.	%		No.	%			
Diarrhoea	15	5.95		8	3.17	7	2.78	0.796	8	3.17	7	2.78	0.796	8	3.17	7	2.78	0.796
Gastritis	19	11.51		17	6.75	12	4.76	0.353	18	7.14	11	4.37	0.194	7	2.78	22	8.73	0.005
Pneumonia	6	2.38		3	1.19	3	1.19	1.000	3	1.19	3	1.19	1.000	6	2.38	0	0.00	0.014
Kennel cough	13	5.16		5	1.98	8	3.17	0.405	8	3.17	5	1.98	0.405	8	3.17	5	1.98	0.405
Dermatitis	19	7.54		11	4.37	8	3.17	0.491	11	4.37	8	3.17	0.491	4	1.59	15	5.95	0.012
Conjunctivitis	17	6.75		5	1.98	12	4.76	0.090	9	3.57	8	3.17	0.808	4	1.59	13	5.16	0.029
Otitis	9	3.57		7	2.78	2	0.79	0.096	5	1.98	4	1.59	0.739	0	0.00	9	3.57	0.003
Canine distemper/Hepatitis	5	1.98		3	1.19	2	0.79	0.655	4	1.59	1	0.40	0.180	0	0.00	5	1.98	0.025
Bacterial diseases	12	4.76		7	2.78	5	1.98	0.564	8	3.17	4	1.59	0.248	6	2.38	6	2.38	1.000
Parasitic diseases	63	25.00	<0.001	34	13.49	29	11.51	0.529	36	14.29	27	10.71	0.257	20	7.94	43	17.06	0.004
Protozoal diseases	2	0.79		2	0.79	0	0.00	0.157	2	0.79	0	0.00	0.157	0	0.00	2	0.79	0.157
Fungal diseases	6	2.38		2	0.79	4	1.59	0.414	4	1.59	2	0.79	0.414	0	0.00	6	2.38	0.014
Endometritis & pyometra	4	1.59		3	1.19	1	0.40	0.317	0	0.00	4	1.59	ND	0	0.00	4	1.59	ND
Pregnancy detection	4	1.59		2	0.79	2	0.79	1.000	0	0.00	4	1.59	ND	0	0.00	4	1.59	ND
Wound	27	10.71		20	7.94	7	2.78	0.012	16	6.35	11	4.37	0.336	16	6.35	11	4.37	0.336
Abscess	2	0.79		2	0.79	0	0.00	0.157	2	0.79	0	0.00	0.157	2	0.79	0	0.00	0.157
Neuter	5	1.98		2	0.79	3	1.19	0.655	5	1.98	0	0.00	ND	0	0.00	5	1.98	ND
Parvo virus	7	2.78		2	0.79	5	1.98	0.257	4	1.59	3	1.19	0.705	7	2.78	0	0.00	0.008
Spaying	7	2.78		3	1.19	4	1.59	0.705	0	0.00	7	2.78	ND	0	0.00	7	2.78	ND
<b>Total</b>	<b>252</b>	<b>100.00</b>	-	<b>138</b>	<b>54.76</b>	<b>114</b>	<b>45.24</b>	<b>0.131</b>	<b>143</b>	<b>56.75</b>	<b>109</b>	<b>43.25</b>	<b>0.032</b>	<b>88</b>	<b>34.92</b>	<b>164</b>	<b>65.08</b>	<b>&lt;0.001</b>

ND= not done; No.= Number, %= percentage

Table 2. Occurrence of clinical cases of pet cats according to their breed, sex and age.

Cases	Overall			Breed				Sex				Age					
	No.	%	p-value	Local		Cross		Male		Female		1-6 months		>6 Months		p-value	
				No.	%	No.	%	No.	%	No.	%	No.	%	No.	%		
Diarrhoea	45	5.64		27	3.38	18	2.26	0.180	24	3.01	21	2.63	35	4.39	10	1.25	<0.001
Gastitis	33	4.14		20	2.51	13	1.63	0.223	22	2.76	11	1.38	20	2.51	13	1.63	0.223
Pneumonia	36	4.51		24	3.01	12	1.50	0.046	20	2.51	16	2.01	26	3.26	10	1.25	0.008
Urinary disorder	8	1.00		5	0.63	3	0.38	0.480	6	0.75	2	0.25	0	0.00	8	1.00	0.005
Poisoning	30	3.76		22	2.76	8	1.00	0.011	18	2.26	12	1.50	22	2.76	8	1.00	0.011
Dermatitis	28	3.51		18	2.26	10	1.25	0.131	16	2.01	12	1.50	8	1.00	20	2.51	0.023
Conjunctivitis	24	3.01		12	1.50	12	1.50	1.000	14	1.75	10	1.25	15	1.88	9	1.13	0.221
Otitis	24	3.01		14	1.75	10	1.25	0.414	14	1.75	10	1.25	14	1.75	10	1.25	0.414
Bacterial diseases	24	3.01	<0.001	15	1.88	9	1.13	0.221	16	2.01	8	1.00	4	0.50	20	2.51	0.001
Viral diseases	54	6.77		40	5.01	14	1.75	<0.001	36	4.51	18	2.26	44	5.51	10	1.25	<0.001
Parasitic diseases	186	23.31		132	16.54	54	6.77	<0.001	114	14.29	72	9.02	126	15.79	60	7.52	<0.001
Protozoal diseases	21	2.63		15	1.88	6	0.75	0.050	14	1.75	7	0.88	16	2.01	5	0.63	0.016
Fungal diseases	21	2.63		12	1.50	9	1.13	0.513	12	1.50	9	1.13	16	2.01	5	0.63	0.016
Pregnancy detection	12	1.50		12	1.50	0	0.00	ND	0	0.00	12	1.50	0	0.00	12	1.50	ND
Orthopedic surgery	54	6.77		36	4.51	18	2.26	0.014	36	4.51	18	2.26	42	5.26	12	1.50	<0.001
Wound	144	18.05		102	12.78	42	5.26	<0.001	82	10.28	62	7.77	108	13.53	36	4.51	<0.001
Spaying	24	3.01		10	1.25	14	1.75	0.414	0	0.00	24	3.01	0	0.00	24	3.01	ND
Neuter	30	3.76		15	1.88	15	1.88	1.000	30	3.76	0	0.00	0	0.00	30	3.76	ND
<b>Total</b>	<b>798</b>	<b>100.00</b>	-	<b>531</b>	<b>66.54</b>	<b>267</b>	<b>33.46</b>	<b>&lt;0.001</b>	<b>474</b>	<b>59.40</b>	<b>324</b>	<b>40.60</b>	<b>496</b>	<b>62.16</b>	<b>302</b>	<b>37.84</b>	<b>&lt;0.001</b>

ND= not done; No.= Number, %= percentage

**Table 3. Occurrence of clinical cases of pet rabbits according to their breed, sex and age.**

Cases	Overall			Sex				Age				
	No.	%	p-value	Male		Female		1-6 months		>6 Months		p-value
				No.	%	No.	%	No.	%	No.	%	
Diarrhoea	20	13.33		14	9.33	6	4.00	13	8.67	7	4.67	0.180
Dermatitis	18	12.00		13	8.67	5	3.33	10	6.67	8	5.33	0.637
Pneumonia	12	8.00		9	6.00	3	2.00	9	6.00	3	2.00	0.083
Conjunctivitis	8	5.33		6	4.00	2	1.33	4	2.67	4	2.67	1.000
Coccidiosis	12	8.00		10	6.67	2	1.33	8	5.33	4	2.67	0.248
Poisoning	4	2.67	<0.001	2	1.33	2	1.33	3	2.00	1	0.67	0.317
Parasitic infestation	20	13.33		15	10.00	5	3.33	8	5.33	12	8.00	0.371
Alopecia	10	6.67		7	4.67	3	2.00	4	2.67	6	4.00	0.527
Traumatic injuries and fracture	25	16.67		19	12.67	6	4.00	16	10.67	9	6.00	0.162
Abscess	5	3.33		3	2.00	2	1.33	2	1.33	3	2.00	0.655
Others	16	10.67		11	7.33	5	3.33	7	4.67	9	6.00	0.617
<b>Total</b>	<b>150</b>	<b>100.00</b>	-	<b>109</b>	<b>72.67</b>	<b>41</b>	<b>27.33</b>	<b>84</b>	<b>56.00</b>	<b>66</b>	<b>44.00</b>	<b>0.142</b>

ND= not done; No.= Number, %= percentage

## **Influence of breed, sex and age on the prevalence of clinical cases in pet animals**

Clinical cases were more prevalent in male dogs than female dogs (56.75% vs 43.25%) ( $p < 0.05$ ), and dogs more than 6 months of age had more reported clinical cases compared to 1-6 months of aged dogs (65.08% vs 34.92%) ( $p < 0.05$ ) (Table 1). Our findings are supported by Yadav *et al.* (2017) who revealed that clinical cases were higher in male than female dogs (58.72% vs 41.28%), and in dogs, more than 6 months compared to dogs less than 6 months of age (68.02% vs 31.98%). The findings of this study further are in partial agreement with the report of Tarafder and Samad (2010) who stated that the prevalence of clinical cases was higher in old (48.12%) followed by adults (34.33%) and young dogs (17.55%). The local breed of cats showed a higher prevalence of clinical cases than that of crossbreed (66.54% vs 33.46%), male cats had a higher prevalence compared to female cats (59.40% vs 40.60%), and cats at the age of 1-6 months represented higher prevalence than those more than 6 months of age (62.16% vs 37.84%) ( $p < 0.05$ ) (Table 2). Likewise, Yadav *et al.* (2017) showed that clinical cases of cats were higher in males (58.65%), in the local breed (79.70%), and in cats less than 6 months of age (51.13%). Male rabbits represented higher reported clinical cases than female rabbits (72.67% vs 27.33%) ( $p < 0.05$ ) (Table 3). This finding is in agreement with the results of Yadav *et al.* (2017) who reported more clinical cases in male than female rabbits (71.11% vs 22.88%). Likewise, Sarker *et al.* (2015) and Parvez *et al.* (2014) reported that male rabbits had higher clinical cases than female rabbits. The reason for higher recorded clinical cases in male dogs, cats, and rabbits might be due to the preference of males as pets by the pet owners to avoid the unnecessary stress of special care and management hazard for female pets during the gestation period along with the reproductive noise. Wound infection was reported as a higher percentage in male dogs compared to female dogs ( $p < 0.05$ ). This is due to the movement of male dogs are more frequent than female dogs. Parasitic diseases, gastritis, dermatitis, conjunctivitis, otitis, canine distemper/hepatitis, and fungal diseases were more prevalent in dogs more than 6 months of age; however, pneumonia, and parvovirus infection showed higher prevalence in 1-6 months of aged dogs ( $p < 0.05$ ). The reasons behind of this, some diseases are age specific. Parasitic diseases, wound infection, viral diseases, orthopaedic surgery, pneumonia, poisoning, and protozoal diseases were more reported in local breed cats compared to cross-breed cats ( $p < 0.05$ ). Male cats were more prone to parasitic diseases, viral diseases, and orthopaedic surgery than female cats ( $p < 0.05$ ). The more reported clinical cases of cats at the age of 1-6 months were parasitic diseases, wound infection, viral diseases, orthopaedic surgery, diarrhoea, poisoning, protozoal diseases, and fungal diseases; however, dermatitis, bacterial diseases, and urinary disorder were more prevalent in cats more than 6 months of age ( $p < 0.05$ ). Most of the pets Owners who have the high social standard they rear crossbred animals and so they medicate their pets properly as compared with others. Our study further reported that male rabbits were more prone to traumatic injuries and fracture, parasitic infestation, and coccidiosis compared to female rabbits ( $p < 0.05$ ). This is because of movement of male rabbits are more as compared with the female rabbits. All the pets such as dogs, cats, and rabbits of this study were infected with parasitic infections or infestations, which might be associated with poor hygiene and not performing regular de-worming. In conclusion, pet dogs, cats, and rabbits were infected by different clinical diseases and conditions along with the increased number of voluntary surgery in Dhaka city, Bangladesh. Parasitic infections and/or infestations appeared as the most significant clinical disease in all pets. Therefore, care should be taken by pet owners to prevent or reduce parasitic infection and/or infestation through regular deworming and good hygiene practices.

## **REFERENCES**

- Biswas, H.R., Hoque, M.M., Samad, M.A. and Rahman, A. 1996. Prevalence of inapparent rabies infection in street dogs. *Bangladesh Vet.*, 13: 29-31.
- Chomel, B.B. 1992. Zoonoses of house pets other than dogs, cats and birds. *Pediatr. Infect. Dis. J.*, 11: 479-487.

- Dada, B.J., Adegboye, D.S. and Mohammed, A.N. 1979. A survey of gastrointestinal helminth parasites of stray dogs in Zaria, Negeria. *Vet. Record*, 104: 145-146.
- Dohoo, I.R., McDonell, W.N., Rhodes, C.S. and Elazhary, Y.L. 1998. Veterinary research and human health. *Canadian Vet. J.*, 39: 548-556.
- Geffray, L. 1999. Infections associated with pets. *Rev. Med. Interne*, 20: 888 -901.
- Hossain, S.S.M.R. and Kayesh, M.E.H. 2014. Common diseases of pet animals in Dhaka city and their zoonotic importance. *Int. J. Nat. Soc. Sci.*, 1: 81-84.
- Kornblatt, A.N. and Schantz, P.M. 1980. Veterinary and public health considerations in canine roundworm control: a survey of practicing veterinarians. *JAVMA*, 177 : 1212-1215.
- Mahmud, M.A.A., Belal, S.M.S.H. and Uddin, F.M.J. 2014. Prevalence of protozoan diseases in pet dogs at district veterinary hospital, Sirajganj, Bangladesh. *Bangladesh J. Vet. Med.*, 12: 191-196.
- Molyneux, D.H. 2004. Neglected diseases but unrecognized successes challenges and opportunities for infectious disease control. *Lancet*, 364 : 380 -383.
- Parvez, M.A., Proshan, M.A.M., Das B.C. and Khatun, R. 2014. Prevalence of clinical conditions in dogs and cats at teaching veterinary hospital (TVH) in Chittagong Veterinary and Animal Sciences University, Bangladesh. *Res. J. Vet. Practi.*, 2: 99-104.
- Plant, M., Zimmerman, E.M. and Goldstein, R.A. 1996. Health hazards to humans associated with domestic pets. *Annu. Rev. Public Health*, 17: 221-245.
- Robertson, I.D., Irwin, P.J., Lymbery, A.J. and Thompson, R.C.A. 2000. The role of companion animals in the emergence of parasitic disease. *Int. J. Parasitol.*, 30: 1369-1377.
- Runa, N.S., Ahmed, S.U., Husna, A., Runa, N.Y., Yesmin, S., Badruzzaman, A.T.M., Islam, M.S., Noor, M., Alam, K.J. and Rahman, M.M. 2016. Prevalence of diseases in pet animals at Dhaka city of Bangladesh. *Annals Vet. Anim. Sci.*, 3: 88-92.
- Samad, M.A. 2011. Public health threat caused by zoonotic diseases in Bangladesh. *Bangladesh J. Vet. Med.*, 9: 95-120.
- Sarker, M.S., Ahduzzman, M., Kabir, M.N., Rahman, M.K., Hossian, Farhana, Nath, S.K. and Bupasha, Z.B. 2015. Prevalence of Clinical Conditions in Dogs and Cats at Central Veterinary Hospital (CVH) in Dhaka, Bangladesh. *Bangladesh Vet. J.*, 26: 101-105.
- Singh, S.K., Islam, M.K. and Hasan, M.T. 2014. The Prevalence of Clinical Diseases in Dogs of Sylhet Sadar, Bangladesh. *Int. J. Pure Appl. Sci. and Technol.*, 5: 41.
- Tarafder, M. and Samad, M.A. 2010. Prevalence of clinical diseases of pet dogs and risks perception of zoonotic infection by dog owners in Bangladesh. *Bangladesh J. Vet. Med.*, 82: 163-174.
- William, A., Chaudhari, S.U.R. and Atsandac, N.N. 2002. Prevalence of some diseases of dogs and cats at the state government veterinary clinic in Maiduguri (Nigeria). *Pak. Vet. J.*, 22: 56-58.
- Yadev, U., Zuhra, F.T., Rahman M.A. and Ahmed, M.S. 2017. Epidemiological investigation of clinical diseases and conditions of pet animals at Chittagong city area, Bangladesh. *Bangladesh J. Vet. Med.*, 15: 63-70

## LACTIC ACID BACTERIA AND THEIR BENEFICIAL EFFECTS IN FERMENTED DAIRY PRODUCTS

A. Rahman<sup>2</sup>, M. Akter<sup>3</sup>, M. S. Islam<sup>4</sup>, A. Rahman<sup>5</sup>, L. Biswas<sup>6</sup> and M. Asaduzzaman<sup>1</sup>

### ABSTRACT

The main focus of this review is the investigation of the roles of lactic acid bacteria (LAB) in fermented food prepared from dairy sources. LAB has been used extensively in food fermentation across the globe as potential microorganisms. Around the world, several species of LAB are employed as starter cultures in several fermented dairy products. Fermentation of milk and milk products involves the LAB genera *Lactobacillus*, *Streptococcus*, *Leuconostoc*, *Pediococcus*, and *Lactococcus*. LAB can be widely employed in developing novel fermented milk products, given the existing knowledge of its numerous health-promoting properties. LAB is widely known as a natural approach to the fermentation of food and enhancing a healthy life. In recent years, there has been a surge of interest in researching LAB to know more about their effects on starter culture or probiotic activity. From a microbiological viewpoint, the diversity of LAB involved in different fermented dairy products are extensive. LAB identified in fermented products could be assessed for their potential beneficial properties to enhance the quality of the product produced. So, the study's goals are to learn about the most recent studies on LAB, which is found in fermented dairy products, as well as their characteristics and health benefits.

**Keywords:** lactic acid bacteria, fermented dairy products, starter culture

### INTRODUCTION

Lactic acid bacteria (LAB) species are grouped into several genera under the Lactobacillaceae family. They are always an integral part of fermented dairy products and are widely utilized in the fermented food industry (Wang *et al.*, 2021). LAB ferments milk's lactic acid and acts as a probiotic, improving gut microbial balance (Bin Masalam *et al.*, 2018) and benefiting host health (Li *et al.*, 2020).

Dairy products are considered primary dietary sources of LAB, which can be found naturally or added afterwards (Reuben *et al.*, 2020). Although lactic acid bacteria include more than 60 genera, the most frequent genera in food fermentation generally include *Lactobacillus*, *Lactococcus*, *Leuconostoc*, *Pediococcus*, *Streptococcus*, *Enterococcus*, *Weissella*, (Mokoena, 2017., Li *et al.*, 2020). *Lactobacilli* such as *Lactobacillus acidophilus*, *L. casei*, *L. paracasei*, *L. rhamnosus*, *L. delbrueckii* subsp. *bulgaricus*, *L. brevis*, *L. johnsonii*, *L. plantarum* and *L. fermentum* are commonly used as probiotic products (Nawaz *et al.*, 2011, Panesar, 2011).

Exopolysaccharides produced by LAB are used in the food industry as emulsifiers, stabilizers, thickeners, gelling agents, and to keep in moisture, change rheology, firmness, and syneresis, and improve texture, mouthfeel, and sensory properties (Korcza and Varga, 2021). Many studies have shown that lactic acid bacteria may produce antioxidant metabolites. Antioxidant compounds produced by LAB in fermented foods are quite safe and may have several positive impacts on human health (Feng and Wang, 2020).

Scientists reveal LAB's health advantages. The health advantages include anti-allergic, anti-carcinogenic, prevention of gastrointestinal infection, constipation relief, immune system activation, and cholesterol-reducing (Panesar, 2011). In many aspects related to LAB functions, preservation and milk fermentation are explained in different research articles. The main target of this review paper is to provide the reader with summarized data related to the beneficial effect of LAB in fermented dairy products, its features and health benefits.

#### Different fermented dairy products

LAB is essential in the fermentation of milk, the process by which raw milk is transformed into

---

<sup>1</sup>Associate Professor, <sup>2</sup>MS Student, <sup>5</sup>Assistant Professor, <sup>6</sup>Lecturer, Dept. of Dairy Science, <sup>3</sup>Associate Professor, Dept. of Animal Nutrition, Genetics and Breeding, <sup>4</sup>Associate Professor, Dept. of Animal Production and Management, Sher-e-Bangla Agricultural University, Dhaka- 1207

fermented milk products. Various LAB strains have been discovered, and their distinctive traits and actions have been used in the industrial production of fermented milk products. LAB's proteolytic activities change milk protein, resulting in acidification and the addition of taste and texture. Yogurt and cheese are two examples of fermented milk products, and both of them have a pleasant, mild acid taste and a delightfully firm texture (Widyastuti *et al.*, 2014). Almost all domestic milch animals' milk (usually cow milk, buffalo milk, ewe milk, goat milk etc.) can be used to make fermented dairy products. Several unique fermented milk products are gaining rapid growth in popularity in many parts of the world. Milk fermentation involving LAB could occur naturally or be artificially induced. Several of these microorganisms are capable of colonizing new environments and transforming milk into an array of healthy fermented dairy products (yogurt, cheese, kefir, fermented milk, lassi, borhani, kumys etc.) (Garcia-Burgos *et al.*, 2020). Consumers' increasing demand for therapeutic goods has resulted in the widespread use of probiotic microorganisms in dairy products. Members of the bacterial genus *Lactobacillus* or *Bifidobacterium* are the most common types of probiotic bacteria found in today's commercial products (Panesar, 2011). Microorganisms used in fermented foods and beverages include bacteria (e.g. *Lactobacillus*, *Streptococcus*, *Enterococcus*, *Lactococcus* and *Bifidobacterium*), moulds (e.g. *Aspergillus oryzae*, *Aspergillus sojae*, *Penicillium roqueforti* and *Penicillium chrysogenum*), and yeasts (e.g. *Saccharomyces cerevisiae*, *Candida krusei* and *Candida humilis*) (Xiang *et al.*, 2019). They may be thoroughly investigated in the creation of novel fermented milk products since they are widely accepted as safe and possess a variety of health-promoting properties. Different popular fermented dairy products and LAB used in the preparation of these products will be discussed here.

#### **Fermented milk**

Fermented milk is a type of dairy product made by lowering the  $p^H$  of milk by the activity of appropriate microorganisms, with or without coagulation, in accordance with the legal restrictions (isoelectric precipitation). These inoculating microorganisms must remain alive and intact in the product over its minimum shelf life. There is a unique culture utilized in the fermentation of each type of fermented milk (Garcia-Burgos *et al.*, 2020).

Fermented milk products or cultured dairy products that have been fermented by a group of lactic acid bacteria cause milk curdling or sourness and also help milk retain its flavour and nutritional value (Mokoena, 2017). Starter cultures for fermented milk serve many purposes, including improving the sensory, textural properties, and nutritional qualities of the final product by increasing the production of bio-preservative compounds that preserve the milk and ensure it is safe to consume. Active starter cultures require strict adherence to standards of cleanliness and long-term viability if they are to serve their intended purpose (Surono and Hosono, 2011). *Lactobacilli* can grow from 1 to 10 million cells per millilitre by metabolizing and growing somewhat on milk's nutrients. During fermentation and storage, the microorganisms produce primary and secondary metabolites and enzymes, which are present in the fermented milk both as living cells and as autolyzed cells. Therefore, fermented milk's nutritional value is determined by the nutrient contents present, the ease with which they can be digested, and the alterations to these components that occur during the microbial growth and fermentation processes. (Xiang *et al.*, 2019). The production of fermented milk relies heavily on mesophilic cultures. Use of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* as a thermophilic starter culture, and *Lactococcus lactis* subsp. *cremoris* and *Leuconostoc* sp. as a mesophilic starter cultures, both are common and used for the fermentation of fermented drinks (Surono and Hosono, 2011). Different types of fermented milk are gaining popularity due to their numerous beneficial effects on human health.

#### **Cheese**

Cheese is made by a process of microbial fermentation in which certain lactic acid bacteria (LAB) convert lactose into lactic acid. This lowers the curd's pH to a particular degree (Hayaloglu, 2016). Various strains of lactic acid bacteria are widely used in the production of a wide variety of cheeses. LAB starter cultures may either be mesophilic from the genera *Lactococcus* and *Leuconostoc* or thermophilic from the genera *Streptococcus* and *Lactobacillus*. *Streptococcus thermophilus*, *Lactococcus lactis*, and *Lactobacillus helveticus* are the three species that have undergone the most

thorough research. *L. helveticus* plays a substantial part in the production of certain taste compounds in Italian artisan cheeses. (Widyastuti *et al.*, 2014) Protein catalysts produced by the microorganisms degrade complex compounds, such as the proteins and sugars in milk, into simpler molecules that can often be kept for longer periods than the original material via a process called microbial fermentation. Cheese made from fermented milk may be eaten months or even years after the milk itself would have gone bad. Cheeses that have been matured for 12 months or more have a more robust taste, aroma, and texture than their fresh counterparts, which may be served immediately after production. (Swanson *et al.*, 2014) Mesophilic lactobacilli such as *Pediococcus* spp., *Enterococcus* spp., and *Leuconostoc* spp. are the most frequent non-starter lactic acid bacteria (NSLAB) in white-brined cheeses. However, white-brined cheeses had negligible levels of *pediococci* and *enterococci*. A study found that the addition of *Enterococcus durans* and *E. faecium* enhanced the cheese's flavour and texture. NSLAB are present in all cheeses, even unripened cheeses, but only after they have been aged for some period of time (Hayaloglu, 2016). Mesophilic starters consisting of *L. lactis* subsp. *cremoris* and *L. lactis* subsp. *lactis* are employed as starters in the production of cheddar cheese, while the other strains of *lactobacilli* may be added just to aid in taste formation and hasten the cheese's ripening process (e.g., by using *L. helveticus*, *L. casei*). It has been suggested that *S. thermophilus* may be utilized for rapid acidification (Hayaloglu, 2016). Therefore, there is a use of different starter LAB and NSLAB in the production of various soft, semi-hard and hard types of cheese.

### **Dahi/ Yogurt**

Dahi is the oldest and most widely consumed fermented milk product. Dahi can be eaten plain or with the addition of salt or sugar. Usually, dahi is prepared using a simple art or technology following the traditions. It is either prepared in consumers' homes or in urban sweet maker businesses. The milk is heated in the home, then cooled to room temperature, and then added with a tablespoon of starter culture (previous day's dahi or buttermilk) (about 0.5-2.0 %) and allowed to set overnight (Shah, 2017). It is preferable to apply well-known starter mixtures for producing dahi on a big scale with known, uniform quality. *Lactococcus lactis* ssp. *lactis*, *L. lactis* ssp. *cremoris*, *L. lactis* ssp. *diacetylactis*, *Leuconostoc* spp., *Lactobacillus* spp., and *Streptococcus thermophilus* are the most used starter microorganisms in the preparation of dahi (Shah, 2017). Because of their greater acid tolerance, *lactobacilli* predominate in sour dahi and *streptococci* predominate in sweet dahi (Shah, 2017). *Lactobacillus* (62%) was the predominant bacterial species in the dahi sample, followed by *Streptococcus* (38%) (Nahidul-Islam *et al.*, 2018). The starter cultures, processing conditions, milk supply, chemicals and additives used to make yogurt all contribute to its final flavour. The starting cultures used in the fermentation process are a particularly important contributor to the development of taste components. Several countries only allow goods to be labelled "yogurt" if they are made with starters containing strains of both *S. thermophilus* and *Lb. delbrueckii* subsp. *bulgaricus*, the two bacteria that make up the conventional yogurt culture (Shah, 2017). Flavour compound production was evaluated by Beshkova *et al.*, (2012) during lactic acid fermentation using a mixed culture and they found that the highest concentration of flavour compounds occurred between 22 and 31 hours into the cooling stage. Yogurt often contains commercial probiotic microorganisms from the families *Lactobacillus* and *Bifidobacterium*. (Nyanzi *et al.*, 2021). Yogurt, Bulgarian buttermilk, and the whole spectrum of goods prepared with gut bacteria, principally *lactobacilli* and *bifidobacteria*, all employ thermophilic starting cultures. Yogurt of varying flavours relies on thermophilic starter cultures. They are added to yogurt for their purported health advantages, but they also change the flavour. When making stirred yogurt, mesophilic microorganisms are utilized (Surono and Hosono, 2011). Therefore, yogurt is very popular in many parts of the world as a dessert after heavy meals due to its efficiency in digesting foods and other beneficial health properties.

### **Borhani**

Borhani is a yogurt-based drink that is often offered during lavish gatherings like weddings and celebrations. Pepper, mint leaves, cumin seeds, green chillies, sugar, salt, and water are all added before being blended till smooth. After that, it's chilled by being stored in a fridge (Hossain and Kabir, 2016). Sour yogurt is an essential ingredient in making borhani. It's a great nutritional choice since it

has a wide variety of nutrients including protein, carbohydrates, fats, vitamins, and minerals (Bari *et al.*, 2020). *Lactobacillus* (43%), *Streptococcus* (20%), and *Pseudomonas* (3%), in addition to the most frequent LABs, was discovered as predominant bacteria in borhani (Nahidul-Islam *et al.*, 2018). Whole milk curd borhani had the most nutrient value and were the most popular with consumers, however, preparing borhani using skim milk curd or full cream powder milk curd was more cost-effective. It can be established, however, that borhani made with whole milk curd is superior to other forms of borhani in terms of both nutritional content and customer preference (Bari *et al.*, 2020). As borhani is made from dahi blended with mint leaves, it has an effect on digestion.

### **Kefir**

Kefir is a fermented milk drink similar to a thin yogurt which is prepared through the symbiotic fermentation of milk by lactic acid bacteria and yeasts contained within an exopolysaccharide and protein complex called a kefir grain (Garrote *et al.*, 2001). Kefir grains were first described by the tribes in the Northern Caucasian mountain region of Russia (Seydim, 2001). Kefir grains are generally symbiotic colonies of bacteria and yeast in a protein and lipid matrix. Kefir differs from other fermented products because of the particular characteristic of its starter culture special flavour and taste. LAB is the main population in kefir grains accompanied by acetic acid bacteria and yeasts (Dong *et al.* 2018). Lactic acid bacteria generally produced lactic acid, acetic acid and antimicrobial compounds that help the preservation of the kefir (John and Deeseenthum, 2015) and also organoleptic properties by producing volatile compounds (e.g. acetaldehyde), exopolysaccharides (Rimada and Abraham, 2003) or free amino acids (Guzel-Seydim *et al.*, 2011). Alcohol and carbon dioxide are produced by yeasts produced in the milk that contribute to the mouthfeel and taste of kefir (Rosa *et al.* 2017).

The bacterial genera most commonly found in kefir using culture-dependent techniques are *Lactobacillus*, *Lactococcus*, *Streptococcus*, and *Leuconostoc*. Among them, *Lactobacillus acidophilus* is one of the predominant species. (Simova *et al.*, 2002; Witthuhn *et al.*, 2004; Chen *et al.*, 2008). There were anywhere between 65% and 80% *Lactobacillus* and *Lactococcus* in kefir grains, with yeasts making up the rest of the microbial makeup. 80 percent of the *Lactobacillus* were identified as *Lactobacillus kefir*, while the remaining 20 percent were identified as one of the following: *Lactobacillus acidophilus*, *Lactobacillus paracasei* subsp. *paracasei*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Lactobacillus plantarum*, or *Lactobacillus kefirianofaciens* (Rosa *et al.*, 2017).

Scientists are interested in kefir because of its claimed health benefits, which include better digestion and lactose tolerance, antibacterial effect, hypocholesterolemic effect, control of plasma glucose, anti-hypertensive effect, anti-inflammatory effect, antioxidant activity, anti-carcinogenic activity, and anti-allergenic activity.

### **Role of LAB as a starter culture in fermented dairy products**

A "starter culture" is a microbial preparation comprising significant populations of cells of at least one microbe added to raw material to generate a fermented food, which enhances and facilitates the fermentation process. There is a long and safe history of using LAB in the manufacturing of fermented dairy foods and beverages, thus they are an integral part of these processes (Hati *et al.*, 2013). In a wide variety of processes involving the fermentation of milk and the storage of food, LAB is indispensable, and the importance of their contributions cannot be overstated. LAB have been used to improve product features and impart particular properties that promote customer satisfaction and interest. As a result, several traditional dairy products have been made utilizing lactic acid bacteria. The majority of the goods that are created via the utilization of LAB also provide greater health benefits to the customers, which is essential for the maintenance of a healthy digestive system (Ayivi *et al.*, 2020). The flavour and aroma of fermented items can be enhanced by the presence of LAB. They cause the food to become acidic, giving it a flavour similar to that of lactic acid, frequently engage in actions that are proteolytic and lipolytic, and create aromatic substances. As a result of their excellent synthesizing capacity and the ability to create aromatic compounds, wild strain starters and NSLAB play a crucial part in the process of flavour formation. Increased levels of amino acids, peptides, and liberated fatty acids are produced as a result of the incorporation of NSLAB as adjunct cultures during the production

of cheese. This results in a more robust flavour profile and quicker maturation of the cheese. (Hati *et al.*, 2013). LAB is responsible for the production of fermented foods as well as the development of their flavour through the processes of proteolysis, glycolysis and lipolysis. Despite the fact that the major metabolic action is the production of lactic acid from the fermentation of carbohydrates, also known as the acidification of the food, LAB are involved in the production of many bioactive components (Bintsis, 2018).

Gram-positive, non-spore-forming, catalase-negative, cytochrome-deficient, non-aerobic yet aerotolerant, fastidious, acid-tolerant, strictly fermentative bacteria called lactic acid bacteria create lactic acid as the principal end product of sugar fermentation (Konig and Frohlich, 2017). They can't make a proton gradient, which is necessary to produce ATP, since they can't make cytochromes and porphyrins (parts of respiratory chains). Fermentation of carbohydrates is the sole source of ATP for the lactics. Lactic acid bacteria thrive in anaerobic environments because they do not need oxygen for energy generation; yet, they may also expand when exposed to oxygen. Because they contain peroxidases, they are safe from oxygen radicals. The capacity to ferment hexoses to lactic acid is what sets these organisms apart from others, thus the name. Because of their differing carbohydrate fermentation capabilities, LAB is classified as either homofermentative or heterofermentative. One molecule of glucose is converted into two molecules of lactate by homofermentative LABs, such as *Lactococcus* and *Streptococcus*, whereas glucose is converted into lactate, ethanol, and carbon dioxide by heterofermentative LABs, such as *Leuconostoc*, *Wiessella*, and some *lactobacilli* (Mokoena, 2017). Because of their restricted metabolic capabilities, lactic acid bacteria must get their carbon and energy from external sources, such as sugar. It is common practice to use a nutrient-dense media for growing lactics. Due to their stringent needs, they can only live in places where certain compounds are plentiful (animals, plants, and other multicellular organisms). Most lactic acid bacteria strains can thrive in a pH range from 4.4 to 5.5, and they can survive temperatures ranging from 5 to 45 degrees Celsius (Konig and Frohlich, 2017). LAB has extensive effects on fermented food preparation, therefore it is very popular. The health effects of LAB will be discussed later on. The species of lactic acid bacteria which were commonly used in the preparation of fermented dairy products is given in the following table 1:

**Table 1. Starter cultures used in fermented dairy products**

Fermented Dairy Products	Starter Cultures	References
Cultured buttermilk	<i>Streptococcus lactis</i> subsp. <i>diacetylactis</i> , <i>S. cremoris</i>	Ayivi <i>et al.</i> , 2020
Dahi	<i>Lactobacillus fermentum</i> , <i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i> , <i>Streptococcus thermophilus</i> , <i>Lactococcus lactis</i> subsp. <i>cremoris</i> , <i>Lactococcus lactis</i> subsp. <i>lactis</i> biovar <i>diacetylactis</i> , <i>Leuconostoc mesenteroides</i> subsp. <i>mesenteroides</i> , <i>Streptococcus thermophilus</i>	Bhattarai <i>et al.</i> , 2021; Bangaragiri <i>et al.</i> , 2021; Rai, 2020
Hard Cheese	<i>Lactococcus lactis</i> subsp. <i>lactis</i> , <i>L. lactis</i> subsp. <i>cremoris</i> , <i>Streptococcus thermophiles</i> , <i>L. delbrueckii</i> subsp. <i>bulgaricus</i> , <i>Lactobacillus helveticus</i>	Hayaloglu, 2016; Gobetti <i>et al.</i> , 2018
Hard cheese without eyes	<i>Lactococcus lactis</i> subsp. <i>lactis</i> , <i>Lactococcus lactis</i> ssp. <i>cremoris</i>	Hayaloglu, 2016
Kefir	<i>Lactobacillus acidophilus</i> , <i>Lactobacillus kefir</i> , <i>Lactobacillus kefiranofaciens</i>	Chen <i>et al.</i> , 2008; Rosa <i>et al.</i> , 2017
Kumiss	<i>L. acidophilus</i> , <i>L. bulgaricus</i> , <i>L. casei</i>	Ghosh <i>et al.</i> , 2019; Panesar, 2011
Lassi	<i>Lactococcus lactis</i> ssp. <i>cremoris</i> , <i>Lactobacillus casei</i>	Adiver <i>et al.</i> , 2021
Leben	<i>S. lactis</i> , <i>S. thermophilus</i> , <i>L. bulgaricus</i>	Panesar, 2011
Shrikhand	<i>Lactobacillus bulgaricus</i> , <i>Streptococcus thermophilus</i>	Sahu, 2021
Surface mould cheese	<i>Lactococcus lactis</i> subsp. <i>lactis</i> and <i>L. lactis</i> subsp. <i>cremoris</i>	Panesar, 2011; Hayaloglu, 2016
Swiss cheese	<i>Lactobacillus delbrueckii</i> ssp. <i>lactis</i> , <i>Lb. helveticus</i> , <i>Streptococcus thermophiles</i>	(Hayaloglu, 2016)
Tofu	<i>Lactobacillus plantarum</i>	(Li <i>et al.</i> , 2020)
Yogurt	<i>Lactococcus lactis</i> ssp. <i>lactis</i> , <i>L. lactis</i> ssp. <i>cremoris</i> , <i>L. lactis</i> ssp. <i>diacetylactis</i> , <i>Leuconostoc</i> spp., <i>Lactobacillus</i> spp., <i>Streptococcus thermophiles</i> , <i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	(Shah, 2017; Chen <i>et al</i> 2017; Laino <i>et al.</i> , 2013)

### **Health benefits of LAB**

Some studies have shown that using LAB to make fermented dairy products has positive health effects. *Lactobacillus acidophilus* with dairy products may reduce blood cholesterol, alleviate lactose intolerance, and reduce colon cancer risk (Zhang *et al.*, 2019). Acidification of the starting materials is triggered rapidly by LAB due to the production of organic substances, most notably lactic acid. In addition to acetic acid and ethanol, other by-products include aroma compounds, bacteriocins, exopolysaccharides, and many enzymes (Hati *et al.*, 2013). LAB is used for its different beneficial effects such as bacteriocin production, antiviral activity, anti-diabetic activity, anti-allergic, anti-obesity anti-diarrheal activity, prevention of lactose intolerance and so on (Ayivi *et al.*, 2020). Several studies have shown that consuming fermented milk products improves human health. There are several ways in which LAB may be utilized as a probiotic to help keep us healthy. The starting culture of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* utilized in Koumiss has anti-allergic properties. Colon cancer prevention and therapy both benefit from the usage of LAB (Ayivi *et al.*, 2020). Here we discussed about numerous beneficial effects of different types of fermented dairy products.

### **Alleviation of lactose intolerance**

Intestinal lactase enzyme deficiency causes lactose intolerance in a large percentage of the world's population. Milk containing *L. acidophilus* helps these persons better digest lactose (Grover *et al.*, 2019). In some studies, it was found that about 60-70% of adult people suffering from lactose malabsorption and found that persons who are lactose intolerant may tolerate fermented dairy products like yogurt better than the same quantity of unfermented milk (Downs *et al.*, 2022; Sharanagouda *et al.*, 2022). The lactic acid bacteria used to manufacture yogurt generate lactase, an enzyme that breaks down lactose, and this has been shown to aid lactose digestion (Panesar, 2011). The lactase enzyme produced by *Lactobacillus bulgaricus* and *Streptococcus thermophilus* in yogurt is effective in treating lactose intolerance (Masood *et al.*, 2011). So, different fermented dairy products like yogurt, cheese, lassi, borhani, kumiss, kefir etc. could be a potential dietary use in the treatment of lactose intolerance patient.

### **Bacteriocin production**

The development of tiny proteins called bacteriocins that inhibit related bacteria is another starting enhancer that has been researched in depth by genetic modification in LAB. Nisin, an essential antibacterial component utilized as a natural preservative in food systems, is generated by the "food grade" starter strain, *Lactococcus lactis* ssp. *lactis* (Mokoena *et al.*, 2021). Nisin acts against food-borne bacteria, like *Staphylococcus* spp., *Listeria* spp. and *Clostridium* spp. (Banerji *et al.*, 2022). Increased Nisin production was seen in genetically modified *Lactococcus* spp. owing to the insertion of numerous copies of genes involved in Nisin production, such as nisZ, nisRK, or nisFEG (Grover *et al.*, 2019). Bacteriocins are a diverse category of antimicrobial peptides or proteins that are produced by ribosomes and secreted extracellularly, where they may kill other bacteria (Zhang *et al.*, 2022). There has been an increase in both basic and applied research on lactic acid bacterium (LAB) bacteriocins over the last two decades due to their potential application as bio-preservatives in food and food products to minimize the development of food-borne bacterial illnesses (Nawaz *et al.*, 2011). However, given ideal physical and chemical circumstances, bacteriocins by LAB may be generated in considerably greater quantities during in-vitro fermentations. Due to the complexity of the food matrix and the difficulty of identifying bacteriocin activity in meals, in-vitro research may mimic and analyze the in-situ functionality of bacteriocinogenic starters. The most promising method for achieving the desired fermentation and a safe end product is in-situ bacteriocin production since it allows for rapid, broad, and legal usage of bacteriocins. The generation of bacteriocins may be most important when starter or preservation cultures made from LAB are added to food. (Beshkova and Frengova, 2012). Different fermented dairy products like yogurt, cheese, lassi, borhani, kumiss, and kefir etc. can be a good source of bacteriocin and act as bio-preservatives of dairy products.

### **Anti-diabetic and anti-obesity**

Burakova *et al.*, 2022 show that LAB reduces the occurrence of diabetes and *Lactobacillus acidophilus* reduces type 2 diabetes risks and improves weight management. Species of probiotic bacteria as a

result of their ability to lower cholesterol levels, *Bifidobacterium longum*, *Lactobacillus casei*, and *Lactobacillus acidophilus* have also been associated with playing a role in the prevention of obesity (Ayivi *et al.*, 2020). Examples of special bacteria for obesity include *Lactobacillus acidophilus*, *Lactobacillus brevis*, *Lactobacillus casei*, *Lactobacillus fermentum*, *Lactobacillus paracasei*, *Lactobacillus rhamnosus*, and *Lactobacillus sakei*. These bacteria regulate lipid metabolism, inflammation, glucose metabolism, insulin sensitivity, and other metabolic processes (Wahid *et al.*, 2021). Therefore, fermented dairy products are popularly used for the prevention and treatment of diabetes and obesity.

#### **Anti-diarrheal**

Acute diarrhea in both children and adults may be helped by taking a probiotic. Probiotics were proven to be effective in preventing diarrhea in children, as they shortened the average duration of diarrhea by 13 hours, halves the rate of treatment failure, and so on (Ayivi *et al.*, 2020). There must be a steady population of microorganisms in the gut for the intestines to function normally. Several health issues affecting the digestive tract can be improved by eating fermented dairy products containing live lactic acid bacteria. These include lactose malabsorption, viral and drug-induced diarrhea, irritable bowel syndrome, inflammatory bowel disease, antineoplastic effects on human cell lines, normal blood insulin levels, and improved fat absorption (Heyman, 2000). Restoring normal intestinal flora, eliminating intestinal pathogens, bolstering intestinal barrier capacity to foreign antigens, stimulating nonspecific immunity such as phagocytosis, stimulating humoral immunity, and producing anti-inflammatory products are all ways, in which LAB accomplish these beneficial effects (Masood *et al.*, 2011). Researchers have shown that the *Lactobacillus* strain is effective in treating both viral and idiopathic diarrhea. The probiotic strain of *Lactobacillus rhamnosus*, *Lactobacillus plantarum*, *Bifidobacterium*, and *Enterococcus faecium*, has been shown to be useful in treating children with diarrhea. *Streptococcus faecium* was effective against respiratory tract infection-associated diarrhea (Liao *et al.*, 2021). It is important to encourage children to consume LAB containing foods like yogurt and fermented milk due to the beneficial effects of lactic acid bacteria in diarrheal illness, particularly in youngsters (Masood *et al.*, 2011).

#### **Anti-carcinogenic**

Multiple studies have demonstrated that probiotic bacteria may inhibit the development of cancer or even reverse its course. Since lowering glucuronidase and carcinogen levels and increasing *lactobacilli* and *bifidobacteria* in the stomach minimizes the absorption of harmful mutagens, a knowledge of this phenomenon is vital (Mendoza *et al.*, 2019). Intestinal instillation of probiotics, including *L. casei*, has been shown in several studies to prevent urinary bladder cancer recurrences (Ayivi *et al.*, 2020). Fermented milk products' probiotic content, which includes *Lactobacilli* and *Bifidobacteria*, reduces colon cancer risk by enhancing immune cell function and changing the activity of faecal enzymes such as glucuronidase, beta-glucuronidase, azotortase, and nitroreductase (Moreno *et al.*, 2007). It has been demonstrated that fermented milk products help prevent some types of cancer. Yogurt, Gouda cheese, and buttermilk all have protective effects against breast cancer (Cai *et al.*, 2021). In animal studies, lactic acid bacteria have been shown to have an anti-carcinogenic effect, meaning that they either inhibit the onset of cancer or slow its progression. There was evidence of anti-carcinogenic effects of *L. acidophilus*-fermented yogurt and several hypothesized mechanisms by which lactic acid bacteria may exert their antitumor effects have been proposed (Juraskova *et al.*, 2022). These include alterations in faecal enzymes thought to be involved in colon carcinogenesis, cellular uptake of mutagenic compounds, reduced mutagenicity of chemical mutagens, and tumor suppression by the improved immune response (Panesar, 2011).

#### **Anti-cardiovascular Diseases**

There has been a steady rise in the incidence of cardiovascular illnesses, making them one of the world's top killers. Most cardiovascular disorders may be avoided by taking any of the *lactobacilli* species, including *L. casei*, *L. plantarum*, *L. fermentum*, *L. rhamnosus*, *L. reuteri*, and *L. paracasei* (Forte *et al.*, 2021). By reducing cholesterol levels, most *lactobacilli* are able to stave against hyperlipidemia, hypercholesterolemia, and atherosclerosis (Wahid *et al.*, 2021). Probiotic *lactobacilli*

and metabolic by-products may have cardioprotective effects and reduce blood cholesterol levels. Recent clinical trials, particularly in the treatment of hypertension, have shown the therapeutic effects of probiotics in the management of cardiovascular illnesses. Patients with hypertension have shown a decrease in their systolic and diastolic pressures (an estimated 14-6.9 mm reduction) after taking probiotics (Ayivi *et al.*, 2020). Many probiotic strains have shown effects on alleviating hypertension, including *Lactobacillus plantarum*, *Lactobacillus casei*, *Lactobacillus helveticus*, and *Streptococcus thermophilus* (Thushara *et al.*, 2016).

#### **Antiviral Activity**

LAB is used to treat viral infections and illnesses. It is also worth mentioning that lactic acid bacteria have a wide range of health-promoting antiviral capabilities (Agagunduz *et al.*, 2022). Lactic acid bacteria are thus thought to be effective antidotes to a variety of viral diseases. In addition, the emergence of viral infections like Covid-19 has presented a formidable challenge to scientists as they search for an effective treatment to counteract this global menace. Traditional preventive antiviral medicines and drugs are often linked with a wide range of unwanted side effects, making it imperative to find a natural alternative treatment option for viral infections, such as probiotics and LAB (Ayivi *et al.*, 2020). The antiviral effects of probiotics and lactic acid bacteria are exerted through several methods. Direct viral contact, production of antiviral inhibitory chemicals, immune system regulation, and stimulation are some of the well-known probiotic antiviral mechanisms. Many studies have shown that probiotic lactic acid bacteria's antiviral properties are strain-specific and dependent. *L. acidophilus*, *L. plantarum*, *L. rhamnosus*, *Lactobacillus ruminis*, *Lactobacillus delbrueckii* ssp. *bulgaricus*, *L. fermentum* have antiviral properties (Quinto *et al.*, 2014).

## **CONCLUSION**

Lactic acid bacteria are beneficial starter organisms which are crucial in the fermentation of milk and milk products. Therefore, lactic acid bacteria exert a potent antimicrobial effect against numerous microorganisms, including milk-spoiling organisms and pathogens. Furthermore, it has been speculated that the bacteriocins produced by some strains aid in the long-term storage of fermented milk and milk products. The quality of milk may be improved by fermentation, and it can also be preserved and harmful bacteria removed from milk through fermentation. In addition to food safety, the nutritional and flavour profiles of the products must fulfil modern consumers' expectations. Fermentation with LAB is a cost-effective method for preserving milk that may be used even in rural or isolated areas, and it improves the texture, flavour, and nutritional content of numerous milk products. As LAB can be used as a probiotic, it has a vital role in building a healthy microbiome and enhanced immunity to illnesses and infections. Natural therapies that are ecologically friendly while preventing viruses and food deterioration organisms are currently advocated by the enlightened food safety culture. In recent years, several biotechnological tools are also used to improve the characteristics of lactic acid bacteria to improve the quality of the fermented products produced. Recent advancements in biotechnology may be used to acquire a better understanding of the influence of LAB in different body functions. Moreover, health education must include educating communities about the advantages of consuming fermented milk and milk products.

## **REFERENCES**

- Adiver, C.N. and Hiremath, J.P. 2021. Sensory and physico-chemical characteristics of probiotic goat (*capra hircus*) milk lassi. *IRJMETS*. 03(03).
- Agagunduz, D., Sahin, T.O., Ayten, S., Yilmaz, B., Gunesliol, B.E., Russo, P. and Ozogul, F. 2022. Lactic acid bacteria as pro-technological, bioprotective and health-promoting cultures in the dairy food industry. *Food Biosci.*, 101617.

- Ayivi, R.D., Gyawali, R., Krastanov, A., Aljaloud, S.O., Worku, M., Tahergorabi, R., Silva, R.C. da and Ibrahim, S.A. 2020. Lactic Acid Bacteria: Food Safety and Human Health Applications. *Dairy*, 1(3): 202–232.
- Banerji, R., Karkee, A. and Saroj, S.D. 2022. Bacteriocins against Foodborne Pathogens. *Appl. Biochem. Microbiol.*, 58(5): 518–539.
- Bangaragiri, D., Prabha, R. and Manjunatha, H. 2021. Characterization of Acid and Bile Tolerant Lactic Acid Bacteria Isolates from the Domestic Dahi Samples. Characterization of Acid and Bile Tolerant Lactic Acid Bacteria Isolates from the Domestic Dahi Samples, *JETIR*, 8(10).
- Bari, A., Hossain, M.A., Camy, M.L.Y., Nahid, M.I., Alam, M.A., Sarkar, S., Ahammad, G.S., Rahman, M.H., Wadud, A and Rashid, M.H.U. 2020. Evaluation of borhani prepared from whole milk, skim milk and full cream powdered milk curd. *Asian Australas. J. Food Saf. Secur.*, 4(2): 58–65.
- Beshkova, D. and Frengova, G. 2012. Bacteriocins from lactic acid bacteria: Microorganisms of potential biotechnological importance for the dairy industry: Bacteriocins from lactic acid bacteria. *Eng. Life Sci.*, 12(4): 419–432.
- Bhattarai, R.R., Gautam, N., Nawaz, M.A. and Das, S.K.L. 2021. Isolation and identification of dominant lactic acid bacteria from dahi: an indigenous dairy product of Nepal Himalayas. *J. Microbiol. Biotechnol. Food Sci.*, 2021: 358–363.
- Bin Masalam, M.S., Bahieldin, A., Alharbi, M.G., Al-Masaudi, S., Al-Jaouni, S.K., Harakeh, S.M. and Al-Hindi, R.R. 2018. Isolation, Molecular Characterization and Probiotic Potential of Lactic Acid Bacteria in Saudi Raw and Fermented Milk. *Evid. Based Complementary Altern. Med.*, 1–12.
- Bintsis, T. 2018. Lactic acid bacteria as starter cultures: An update in their metabolism and genetics. *AIMS Microbiology*, 4(4), 665–684. Dertli, E., and Çon, A.H. (2017). Microbial diversity of traditional kefir grains and their role on kefir aroma. *LWT - Food Sci. Technol.*, 85: 151–157.
- Burakova, I., Smirnova, Y., Gryaznova, M., Syromyatnikov, M., Chizhkov, P., Popov, E. and Popov, V. 2022. The effect of short-term consumption of lactic acid bacteria on the gut microbiota in obese people. *Nutrients*, 14(16): 3384.
- Cai, J.S., Feng, J.Y., Ni, Z.J., Ma, R.H., Thakur, K., Wang, S. and Wei, Z.J. 2021. An update on the nutritional, functional, sensory characteristics of soy products, and applications of new processing strategies. *Trends Food Sci. Technol.*, 112: 676–689.
- Chen, C., Zhao, S., Hao, G., Yu, H., Tian, H. and Zhao, G. 2017. Role of lactic acid bacteria on the yogurt flavour: A review. *Int. J. Food Prop.*, 20(1): 316–330pp.
- Choen, H.C., Wang, S.Y. and Chen, M.J. 2008. Microbiological study of lactic acid bacteria in kefir grains by culture-dependent and culture-independent methods. *Food Microbiol.*, 25(3): 492–501.
- Moreno de LeBlanc, A., Matar, C. and Perdigon, G. 2007. The application of probiotics in cancer. *Br. J. Nutr.*, 98: 105–10.
- Dong, J., Liu, B., Jiang, T., Liu, Y. and Chen, L. 2018. The biofilm hypothesis: The formation mechanism of Tibetan kefir grains. *Int. J. Dairy Technol.*, 71: 44–50.
- Downs, B.W., Banik, S.P., Bagchi, M., Ghosh, R.B., Kushner, S. and Bagchi, D. 2022. The microbiome, immunity, anaerobism, and inflammatory conditions: a multifaceted systems biology intervention. (pp. 205–216). *In: Microbiome, Immunity, Digestive Health and Nutrition*. Elsevier Science and Technology, San Diego.
- Feng, T. and Wang, J. 2020. Oxidative stress tolerance and antioxidant capacity of lactic acid bacteria as probiotic: A systematic review. *Gut Microbes*, 12(1): 1801944p.
- Forte, M., Schirone, L., Ameri, P., Basso, C., Catalucci, D. and Modica, J. 2021. The role of mitochondrial dynamics in cardiovascular diseases. *Br. J. Pharmacol.*, 178(10): 2060–2076.
- Garcia-Burgos, M., Moreno-Fernandez, J., Alferéz, M.J.M., Diaz-Castro, J. and Lopez-Aliaga, I. 2020. New perspectives in fermented dairy products and their health relevance. *J. Funct. Foods*, 72: 104059.

- Garrote, G.L., Abraham, A.G. and De Antoni, G.L. 2001. Chemical and microbiological characterisation of kefir grains. *J. Dairy Res.*, 68(4): 639–652.
- Ghosh, T., Beniwal, A., Semwal, A. and Navani, N.K. 2019. Mechanistic insights into probiotic properties of lactic acid bacteria associated with ethnic fermented dairy products. *Front. Microbiol.*, 10: 502p.
- Gobbetti, M., Cagno, R., Calasso, M., Neviani, E., Fox, P.F. and Angelis, M. 2018. Drivers that establish and assemble the lactic acid bacteria biota in cheeses. *Trends Food. Sci. Technol.*, 78: 244-254.
- Grover, S., Batish, V.K. and Reddy, V.P. 2019. Dairy Biotechnology. 245.
- Guzel-Seydim, Z.B., Kok-Tas, T., Greene, A.K. and Seydim, A.C. 2011. Functional properties of kefir. *Crit. Rev. Food Sci. Nutr.*, 51(3): 261-268.
- Hati, S., Mandal, S. and Prajapati, J. 2013. Novel Starters for Value Added Fermented Dairy Products. *Curr. Res. Nutr. Food Sci.*, 1(1): 83–91.
- Hayaloglu, A.A. 2016. Cheese: Microbiology of Cheese. In *Ref. Module in Food Sci.* (p. B9780081005965007000).
- Heyman, M. 2000. Effect of Lactic Acid Bacteria on Diarrheal Diseases. *J. Am. Coll. Nutr.*, 19(2): 137S-146S.
- Hossain, M. and Kabir, Y. 2016. Ethnic Fermented Foods and Beverages of Bangladesh. In *J. P. Tamang, Ethnic Fermented Foods and Alcoholic Beverages of Asia (73–89pp)*.
- John, S.M. and Deeseenthum, S. 2015. Properties and benefits of kefir-A review. *Songklanakarinn J. Sci. Technol.*, 37(3).
- Juraskova, D., Ribeiro, S.C. and Silva, C.C. 2022. Exopolysaccharides produced by lactic acid bacteria: From biosynthesis to health-promoting properties. *Foods*, 11(2):156.
- Konig, H. and Frohlich, J. 2017. Lactic Acid Bacteria. Biology of Microorganisms on Grapes, in Must and in Wine (3–41pp).
- Korz, E. and Varga, L. 2021. Exopolysaccharides from lactic acid bacteria: Techno-functional application in the food industry. *Trends in Food Sci. Technol.*, 110: 375-384.
- Laino, J.E., del Valle, M.J., de Giori, G.S. and LeBlanc, J.G.J. 2013. Development of a high folate concentration yogurt naturally bio-enriched using selected lactic acid bacteria. *LWT- Food Sci. Technol.*, 54(1):1-5.
- Li, J., Huang, Q., Zheng, X., Ge, Z., Lin, K., Zhang, D., Chen, Y., Wang, B. and Shi, X. 2020. Investigation of the Lactic Acid Bacteria in Kazak Cheese and Their Contributions to Cheese Fermentation. *Front. Microbiol.*, 11: 228.
- Liao, W., Chen, C., Wen, T. and Zhao, Q. 2021. Probiotics for the Prevention of Antibiotic-associated Diarrhea in Adults: A Meta-Analysis of Randomized Placebo-Controlled Trials. *J. Clin. Gastroenterol.*, 55(6): 469.
- Masood, M.I., Qadir, M.I., Shirazi, J.H. and Khan, I.U. 2011. Beneficial effects of lactic acid bacteria on human beings. *Crit. Rev. Microbiol.*, 37(1): 91–98.
- Mokoena, M.P. 2017. Lactic Acid Bacteria and Their Bacteriocins: Classification, Biosynthesis and Applications against Uropathogens: A Mini-Review. *Molecules*, 22(8): 1255.
- Mokoena, M.P., Omatola, C.A. and Olaniran, A.O. 2021. Applications of lactic acid bacteria and their bacteriocins against food spoilage microorganisms and foodborne pathogens. *Molecules*, 26(22): 7055.
- Nahidul-Islam, S.M., Kuda, T., Takahashi, H. and Kimura, B. 2018. Bacterial and fungal microbiota in traditional Bangladeshi fermented milk products analysed by culture-dependent and culture-independent methods. *Food Res. Int.*, 111: 431–437.
- Nawaz, M., Wang, J., Zhou, A., Ma, C., Wu, X., Moore, J.E., Cherie Millar, B. and Xu, J. 2011. Characterization and Transfer of Antibiotic Resistance in Lactic Acid Bacteria from Fermented Food Products. *Curr. Microbiol.*, 62(3): 1081–1089.
- Nyanzi, R., Jooste, P.J. and Buys, E.M. 2021. Invited review: Probiotic yogurt quality criteria, regulatory framework, clinical evidence, and analytical aspects. *J. Dairy Sci.*, 104(1):1–19.

- Panesar, P.S. 2011. Fermented Dairy Products: Starter Cultures and Potential Nutritional Benefits. *Food Nutr. Sci.*, 02(01): 47–51.
- Quinto, E.J., Jimenez, P., Caro, I., Tejero, J., Mateo, J. and Girbes, T. 2014. Probiotic Lactic Acid Bacteria: A Review. *Food Nutr. Sci.*, 05(18): 1765–1775.
- Rai, R. 2020. Diversity of lactic acid bacteria and their probiotic properties in some naturally fermented milk products of Sikkim. Ph.D. Thesis. Sikkim University, Sikkim, India.
- Reuben, R.C., Roy, P.C., Sarkar, S.L., Alam, R.U., A.S.M. and Jahid, I.K. 2020. Characterization and evaluation of lactic acid bacteria from indigenous raw milk for potential probiotic properties. *J. Dairy Sci.*, 103(2): 1223–1237.
- Rimada, P.S. and Abraham, A.G. 2003. Comparative study of different methodologies to determine the exopolysaccharide produced by kefir grains in milk and whey. *Le Lait*, 83(1): 79-87.
- Rosa, D.D., Dias, M.M.S., Grzeskowiak, L.M., Reis, S.A., Conceicao, L.L. and Peluzio, M.do C.G. 2017. Milk kefir: Nutritional, microbiological and health benefits. *Nutr. Res. Rev.*, 30(1): 82–96.
- Sahu, V. 2021. Optimization of starter culture to develop healthy goat milk shrikhand. *Pharma Innov.*, 10(10): 1473-1477.
- Seydim, Z.B. 2001. *Studies on fermentative, microbiological and biochemical properties of kefir and kefir grains.*
- Shah, N.P. 2017. Dahi-An Indian Naturally Fermented Yogurt. In *Yogurt in health and disease prevention* (353–369pp).
- Sharanagouda, B., Lal, H. and Mahajan, I.S. 2022. Study on simultaneous lactose hydrolysis and fermentation in the preparation of lactose hydrolyzed yogurt and its effect on physico-chemical properties.
- Simova, E., Beshkova, D., Angelov, A., Hristozova, T.S., Frengova, G. and Spasov, Z. 2002. Lactic acid bacteria and yeasts in kefir grains and kefir made from them. *J. Ind. Microbiol. Biotechnol.*, 28(1): 1-6.
- Surono, S. and Hosono, A. 2011. Fermented Milks | Starter Cultures. In *Encyclopedia of Dairy Sciences* ( 477–482pp). Elsevier.
- Swanson, M.S., Blaser, M.J., Bryant, D.A., Dermody, T. and Fink, G. 2014. Board of Governors, American Academy of Microbiology. 2014: 36.
- Thushara, R.M., Gangadaran, S., Solati, Z. and Moghadasian, M.H. 2016. Cardiovascular benefits of probiotics: A review of experimental and clinical studies. *Food Funct.*, 7(2): 632–642.
- Wahid, F., Huang, L.H., Zhao, X.Q., Li, W.C., Wang, Y.Y., Jia, S.R. and Zhong, C. 2021. Bacterial cellulose and its potential for biomedical applications. *Biotechnol. Adv.*, 53:107856.
- Wang, Y., Wu, J., Lv, M., Shao, Z., Hungwe, M., Wang, J., Bai, X., Xie, J., Wang, Y. and Geng, W. 2021. Metabolism Characteristics of Lactic Acid Bacteria and the Expanding Applications in Food Industry. *Front. Bioeng. Biotechnol.*, 9: 612285.
- Widyastuti, Y., Rohmatussolihat and Febrisiantosa, A. 2014. The Role of Lactic Acid Bacteria in Milk Fermentation. *Food Nutr. Sci.*, 05(04): 435–442.
- Witthuhn, R.C., Schoeman, T. and Britz, T.J. 2004. Isolation and characterization of the microbial population of different South African kefir grains. *Int. J. Dairy Technol.*, 57(1): 33-37.
- Xiang, H., Sun-Waterhouse, D., Waterhouse, G.I.N., Cui, C. and Ruan, Z. 2019. Fermentation-enabled wellness foods: A fresh perspective. *Food Sci. Hum. Wellness*, 8(3): 203–243.
- Zhang, R., Xu, L. and Dong, C. 2022. Antimicrobial peptides: An overview of their structure, function and mechanism of action. *Protein Pept. Lett.*, 29(8): 641-650.
- Zhang, T., Jeong, C.H., Cheng, W.N., Bae, H., Seo, H.G., Petriello, M.C. and Han, S.G. 2019. Moringa extract enhances the fermentative, textural, and bioactive properties of yogurt. *LWT*, 101: 276–284.

## CORRELATION COEFFICIENT AND PATH COEFFICIENT ANALYSIS IN DIFFERENT *Brassica rapa* GENOTYPES

N. J. Shelly<sup>1</sup>, M. S. R. Bhuiyan<sup>2</sup>, F. Mahmud<sup>3</sup>, J. Rahman<sup>4</sup> and M. A. Alam<sup>5</sup>

### ABSTRACT

A field experiment was conducted in the experimental field of Sher-e- Bangla Agricultural University, Dhaka, Bangladesh to study the correlation between pairs of different characters and the direct and indirect effect of different characters on seed yield plant<sup>-1</sup> in seven selected genotypes of *Brassica rapa* L. to select the plants with higher yield potential. Yield plant<sup>-1</sup> was highly significant and positively correlated with days to 50% flowering ( $r_g = 0.987^{**}$  and  $r_p = 0.989^{**}$ ) and 80% maturity ( $r_g = 0.989^{**}$  and  $r_p = 0.990^{**}$ ) at both genotypic and phenotypic level while the correlation was significant and positive with plant height ( $r_g = 0.802^*$  and  $r_p = 0.812^*$ ) and thousand seed weight ( $r_g = 0.770^*$  and  $r_p = 0.768^*$ ) indicating dependence of these characters on grain yield. Path coefficient analysis for yield plant<sup>-1</sup> revealed that days to 80% maturity (7.9587) exerted the highest direct effect on the yield followed by days to 50% flowering (5.4155) and plant height (2.4601). The indirect contribution of component characters viz. days to 80% maturity was high indirect effect via days to 50% flowering (5.3693) followed by number of secondary branches plant<sup>-1</sup> via days to 80% maturity (4.0874) towards seed yield plant<sup>-1</sup>. Thus, the yield plant<sup>-1</sup> in selected *B. rapa* genotypes could be improved by direct selection based on these traits.

**Keywords:** *Brassica rapa*, correlation coefficient, path coefficient analysis, residual effect.

### INTRODUCTION

In Bangladesh total cultivated area under rapeseed and mustard cultivation is 0.522 million hectares which produces 0.683 million metric tons in 2018-19 (AIS, 2020). *B. rapa* is the main oil yielding species in Bangladesh and occupies the 1<sup>st</sup> position in respect of area and production (Naznin *et al.*, 2015). Other two major local cultivar, *B. juncea* and *B. napus* are high yielding but not short durable so comparatively low yielding *B. rapa* is widely grown in the country for their short duration and fulfills our requirement approximately 50% (Islam, 2015). Although short durable, low yielding and pest susceptible variety Tori-7 of *B. rapa* is popular in Bangladesh but there is still lack of improved short durable variety with higher yield. Therefore, oilseed research should be directed towards the minimization of yield gap through the development of the high yielding short duration varieties to fit into the profitable cropping patterns (T. Aman-Mustard-Boro) with higher adaptability and stability which will ultimately increase the oil production in the country. Increased yield and improved quality are generally confronted with laborious analyses and long term breeding programs. The inter relationship of quantitative characters with yield may determine the efficiency of breeding program. Phenotypic correlation reflects the observed relationship, while genotypic correlation underline the true relationship among characters. Selection procedures could be varied depending on the relative contribution of each. The association of a particular character in relation to other traits contributing to yield of a crop would be of great importance in planning a successful breeding program (Mary and Gopalan, 2006). Thus determination of correlation coefficients is an important statistical procedure to evaluate breeding programs for high yield, as well as to examine direct and indirect contributions to yield variables (Ali *et al.*, 2003). On the other hand the path analysis helps to determine the direct and indirect contribution of traits towards the yield which cannot be differentiated from correlation studies. Unlike the correlation coefficient path coefficient measures the magnitude of direct and indirect contribution of a component character to a complex character and it has been defined as a standardized regression coefficient which splits the correlation coefficient into direct and indirect effects and thus

---

<sup>1</sup>Assistant Professor, Dept. of Biochemistry, <sup>2,3&4</sup>Professor, Dept. of Genetics and Plant Breeding, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka-1207, <sup>5</sup>Scientific Officer, Bangladesh Agriculture Research Institute, Gazipur.

enable the breeders to judge about the important component characters during selection (Sabaghnia *et al.*, 2010). Therefore, correlation in combination with the path coefficient analysis quantifies the direct and indirect contribution of one character upon another (Dewey and Lu, 1959). Hence the present study was carried out to understand the correlation and the direct and indirect effect of different characters on yield as selection criteria which will help for launching an effective breeding program to meet the existing demand.

## MATERIALS AND METHODS

Different *B. rapa* genotypes were collected from BARI (Bangladesh Agriculture Research Institute), BINA (Bangladesh Institute of Nuclear Agriculture) and SAU (Sher-e-Bangla Agricultural University). Selection was done on the basis of yield, duration and oil content of the collected genotypes. Then seven selected genotypes BARI Sar-6, BARI Sar-9 × BARI Sar-6 (F<sub>16</sub>), Yellow Special, Tori-7, BARI Sar-14, BARI Sar-15 and BARI Sar-17 were grown on the experimental plot. The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications at the experimental field of Sher-e-Bangla Agricultural University, Dhaka. Each plot was 2.5 m long with three rows. Distance between row- row 30 cm and plant-plant 10 cm and block-block 1 m were maintained. Recommended doses of fertilizers and standard cultural practices were carried out for better crop production. Data were recorded on days to flowering, days to maturity, plant height (cm), number of primary branches plant<sup>-1</sup>, number of secondary branches plant<sup>-1</sup>, number of siliqua plant<sup>-1</sup>, siliqua length (cm), number of seeds siliqua<sup>-1</sup>, 1000 seed weight (g) and seed yield plant<sup>-1</sup> (g). The mean values of ten randomly selected plants were computed for each of ten traits for each genotype in each replication and were subjected to statistical analysis. Mean, range and co-efficient of variation (CV %) were estimated using MSTAT-C software program. Both genotypic and phenotypic coefficients of correlation between two characters were determined by using the variance and covariance components as suggested by Al-Jibouri *et al.* (1958) and path coefficient analysis was done following the method outlined by Dewey and Lu (1959) and categories were determined according to Lenka and Mishra (1973).

## RESULTS AND DISCUSSION

### Analysis of variance

Analysis of variance was performed for ten quantitative characters including yield and yield attributing traits on seven genotypes of *B. rapa* (Table 1). From the analysis of variance it was observed that mean sum of squares due to genotypes were significant for all the studied traits, thus exhibiting the presence of considerable genetic variability. This finding was in confirmation with the findings of Singh *et al.* (2013), Tripathi *et al.* (2013) and Shekhawat *et al.* (2014).

### Correlation analysis

The genotypic and phenotypic correlation coefficient for ten yield and related characters in seven selected genotypes of *B. rapa* was studied and presented in Table 2 and 3.

### Days to 50% flowering

It showed high significant and positive correlation with seed yield plant<sup>-1</sup> ( $r_g = 0.987^{**}$  and  $r_p = 0.989^{**}$ ) and days to 80% maturity ( $r_g = 0.991^{**}$  and  $r_p = 0.990^{**}$ ) at both genotypic and phenotypic levels, so any change or variation for this trait will have considerable effect on seed yield plant<sup>-1</sup>. Hence, selection in the performance of one of the traits will result in the improvement of the other traits. The result was supported by Naznin *et al.* (2015), Jamali *et al.* (2016), and Siddique *et al.* (2017) while Zahan (2006) reported non-significant negative association of this trait with seed yield plant<sup>-1</sup>. Kahrizi and Alaahvarand (2012), Halder *et al.* (2014) reported seed yield plant<sup>-1</sup> had significant negative association with this trait. It presented significant and positive correlation with thousand seed weight ( $r_g = 0.840^*$  and  $r_p = 0.795^*$ ) and plant height ( $r_g = 0.791^*$  and  $r_p = 0.772^*$ ) at both genotypic and phenotypic levels while significant and negative correlation with number of siliqua plant<sup>-1</sup> ( $r_p = -0.784^*$ ) at phenotypic levels only. It also showed non-significant and negative correlation with number

of secondary branches plant<sup>-1</sup> ( $r_g = -0.515$  and  $r_p = -0.551$ ) at both genotypic and phenotypic levels, siliqua length ( $r_p = -0.019$ ) at phenotypic level and number of siliqua plant<sup>-1</sup> ( $r_g = -0.381$ ) at genotypic level only while non-significant and positive correlation with other remaining traits at both levels. Ejaz-Ul-Hasan *et al.* (2014) found a positive and high significant genetic relationship of days to 50% flowering with seeds siliqua<sup>-1</sup>.

**Table 1. Analysis of variance for seed yield and seed yield attributing traits in *Brassica rapa* genotypes**

Source of variation	Df	Days to 50% flowering	Days to 80% maturity	Plant height (cm)	No. of primary branches plant <sup>-1</sup>	No. of secondary branches plant <sup>-1</sup>	No. of siliqua plant <sup>-1</sup>	Siliqua length (cm)	No. of seed siliqua <sup>-1</sup>	1000 Seed Weight (g)	Seed yield plant <sup>-1</sup> (g)
Genotype	6	235.11**	311.89**	2101.65**	2.24**	44.13**	7726.9**	0.88**	101.46**	1.35**	2.96**
Replication	2	26.09	20.67	1.15	0.66	0.69	404.7	0.02	1.18	0.02	0.58
Error	12	0.09	0.08	10.24	1.02	1.24	124.2	0.01	0.91	0.08	0.03
CV (%)		0.66	0.32	3.05	13.69	28.06	6.54	1.97	4.41	7.20	3.09
LSD		0.52	0.51	5.70	1.80	1.98	19.83	0.13	1.70	0.50	0.29

Here, Df = Degree of freedom, CV = Co-efficient of variation, LSD = Least Significant Difference, \*\* = Significant at 1%

**Table 2. Genotypic correlation coefficients among different pairs of yield and yield contributing traits of selected *Brassica rapa* genotypes**

Yield contributing traits	Days to 50% flowering	Days to 80% maturity	Plant height (cm)	No. of primary branches plant <sup>-1</sup>	No. of secondary branches plant <sup>-1</sup>	No. of siliqua plant <sup>-1</sup>	Siliqua length (cm)	No. of seeds siliqua <sup>-1</sup>	1000 seed weight (g)
Days to 80% maturity	0.991**								
Plant height (cm)	0.791*	0.794*							
No. of primary branches plant <sup>-1</sup>	0.195	0.215	0.801*						
No. of secondary branches plant <sup>-1</sup>	-0.515	-0.513	-0.774*	-0.952**					
No. of siliqua plant <sup>-1</sup>	-0.381	-0.239	-0.091	-0.186	0.775*				
Siliqua length (cm)	0.082	0.131	0.849*	0.470	-0.197	0.235			
No. of seeds siliqua <sup>-1</sup>	0.560	0.469	0.219	0.095	-0.411	-0.775*	-0.034		
Thousand seed weight (g)	0.840*	0.843*	0.924**	0.799*	-0.871*	-0.348	0.780*	0.406	
Seed yield plant <sup>-1</sup> (g)	0.987**	0.989**	0.802*	0.213	-0.593	-0.485	0.144	0.571	0.770*

Here, \*\* = Significant at 1%; \* = Significant at 5%

**Table 3. Phenotypic correlation coefficients among different pairs of yield and yield contributing traits of selected *Brassica rapa* genotypes**

Yield contributing traits	Days to 50% flowering	Days to 80% maturity	Plant height (cm)	No. of primary branches plant <sup>-1</sup>	No. of secondary branches plant <sup>-1</sup>	No. of siliqua plant <sup>-1</sup>	Siliqua length (cm)	No. of seeds siliqua <sup>-1</sup>	1000 seed weight (g)
Days to 80% maturity	0.990**								
Plant height (cm)	0.772*	0.771*							
No. of primary branches plant <sup>-1</sup>	0.111	0.124	0.493						
No. of secondary branches plant <sup>-1</sup>	-0.551	-0.548	-0.865*	-0.806*					
No. of siliqua plant <sup>-1</sup>	-0.784*	-0.577	-0.566	-0.366	0.939**				
Siliqua length (cm)	-0.019	0.039	0.794*	0.321	-0.259	-0.073			
No. of seeds siliqua <sup>-1</sup>	0.630	0.534	0.321	0.122	-0.404	-0.871*	0.047		
Thousand seed weight (g)	0.795*	0.801*	0.885**	0.524	-0.912**	-0.846*	0.575	0.477	
Seed yield plant <sup>-1</sup> (g)	0.989**	0.990**	0.812*	0.116	-0.596	-0.781*	0.095	0.598	0.768*

Here, \*\* = Significant at 1%; \* = Significant at 5%

### Days to 80% maturity

Days to 80% maturity showed high significant and positive correlation with seed yield plant<sup>-1</sup> ( $r_g = 0.989$ \*\* and  $r_p = 0.990$ \*\*\*) at both genotypic and phenotypic levels hence direct selection for this trait

will be effective in improving the seed yield. Hosen (2008), Uddin *et al.* (2013) and Halder *et al.* (2014) also found high significant and positive correlation of seed yield plant<sup>-1</sup> with days to maturity but Naznin *et al.* (2015) and Kumari *et al.* (2017) revealed non-significant and positive interaction with seed yield plant<sup>-1</sup>. Zahan (2006) studied non-significant negative association with days to maturity. It also showed significant and positive correlation with thousand seed weight ( $r_g = 0.843^*$  and  $r_p = 0.801^*$ ) and plant height ( $r_g = 0.794^*$  and  $r_p = 0.771^*$ ) whereas non-significant negative correlation with number of secondary branches plant<sup>-1</sup> ( $r_g = -0.513$  and  $r_p = -0.548$ ) and number of siliqua plant<sup>-1</sup> ( $r_g = -0.239$  and  $r_p = -0.577$ ) at both genotypic and phenotypic levels while having non-significant positive correlation with remaining traits at genotypic and phenotypic levels. Ejaz-Ul-Hasan *et al.* (2014) found positive and highly significant genetic relationship of 80% maturity with thousand seed weight and seed siliqua<sup>-1</sup>.

### **Plant height**

Plant height showed high significant and positive correlation with thousand seed weight ( $r_g = 0.924^{**}$  and  $r_p = 0.885^{**}$ ) while it showed significant and positive correlation with seed yield plant<sup>-1</sup> ( $r_g = 0.802^*$  and  $r_p = 0.812^*$ ) and siliqua length ( $r_g = 0.849^*$  and  $r_p = 0.794^*$ ) at both genotypic and phenotypic levels and number of primary branches plant<sup>-1</sup> ( $r_g = 0.801^*$ ) at genotypic level only hence direct selection for this trait will be effective in improving the associated traits. Genotypic correlation coefficients were generally higher than their corresponding phenotypic correlations indicated strong hereditary association and the masking effects of environment on these traits. Joya *et al.* (2016), Afrin *et al.* (2017) and Siddique *et al.* (2017) reported that seed yield plant<sup>-1</sup> had high significant positive association with plant height. Parveen (2007) and Mekonnen *et al.* (2014) observed that seed yield plant<sup>-1</sup> had non-significant positive association with plant height while Kumari *et al.* (2017) observed that seed yield plant<sup>-1</sup> had non-significant negative association with plant height. It was significant and negatively associated with number of secondary branches plant<sup>-1</sup> ( $r_g = -0.774^*$  and  $r_p = -0.865^*$ ) at both genotypic and phenotypic level. Non-significant and negative association was observed for number of siliqua plant<sup>-1</sup> ( $r_g = -0.091$  and  $r_p = -0.566$ ) while the correlation of plant height with other remaining traits were positive but not significant at both genotypic and phenotypic level. Mekonnen *et al.* (2014) studied strong and positive correlation with number of siliqua plant<sup>-1</sup>. Ejaz-Ul-Hasan *et al.* (2014) found a positive and high significant genetic relationship of plant height with seeds siliqua<sup>-1</sup> while Siddique *et al.* (2017) observed negative and significant relationship of this trait with seeds siliqua<sup>-1</sup>.

### **Number of primary branches plant<sup>-1</sup>**

Number of primary branches plant<sup>-1</sup> showed high significant and negative correlation with number of secondary branches plant<sup>-1</sup> at genotypic level ( $r_g = -0.952^{**}$ ) but have significant and negative correlation at phenotypic level ( $r_p = -0.806^*$ ). It also showed significant and positive correlation with thousand seed weight ( $r_g = 0.799^*$ ) at genotypic level only. It had negative and non-significant interaction with number of siliqua plant<sup>-1</sup> ( $r_g = -0.186$  and  $r_p = -0.366$ ) at both genotypic and phenotypic level but had positive non-significant interaction with other remaining traits at both genotypic and phenotypic levels hence selection for this trait would not effective. Naznin *et al.* (2015), Singh *et al.* (2017) and Rauf and Rahim (2018) reported that seed yield plant<sup>-1</sup> had significant positive correlation with number of primary branches plant<sup>-1</sup> while Kumari *et al.* (2017) observed that seed yield plant<sup>-1</sup> had non-significant negative association with number of primary branches plant<sup>-1</sup>.

### **Number of secondary branches plant<sup>-1</sup>**

The number of secondary branches plant<sup>-1</sup> showed high significant and positive correlation with number of siliqua plant<sup>-1</sup> at phenotypic level ( $r_p = 0.939^{**}$ ) but had significant and positive correlation at genotypic level ( $r_g = 0.775^*$ ) while it showed highly significant and negative correlation with thousand seed weight at phenotypic level ( $r_p = -0.912^{**}$ ) but had significant and negative correlation at genotypic level ( $r_g = -0.871^*$ ). The correlation of this trait with other remaining traits were negative and non-significant at both genotypic and phenotypic levels. Uddin *et al.* (2013), Halder *et al.* (2014), Naznin *et al.* (2015) and Afrin *et al.* (2017) observed high significant positive association of this trait

with seed yield plant<sup>-1</sup>. Parveen (2007) and Mekonnen *et al.* (2014) observed non-significant positive association of this trait with seed yield plant<sup>-1</sup>.

#### **Number of siliqua plant<sup>-1</sup>**

It showed significant and negative correlation with number of seeds siliqua<sup>-1</sup> ( $r_g = -0.775^*$  and  $r_p = -0.871^*$ ) at both genotypic and phenotypic level. It also showed significant and negative correlation with thousand seed weight ( $r_p = -0.846^*$ ) and seed yield plant<sup>-1</sup> ( $r_p = -0.781^*$ ) at phenotypic level only and had positive and non-significant association with siliqua length at genotypic level while had negative and non-significant association with other remaining traits thus selection for this trait would not effective. Jamali *et al.* (2016), Afrin *et al.* (2017) and Siddique *et al.* (2017) revealed that it had significant positive association with seed yield plant<sup>-1</sup>. Parveen (2007) observed non-significant positive association with yield plant<sup>-1</sup>. Siddique *et al.* (2017) found significant negative association for this trait with seeds siliqua<sup>-1</sup>.

#### **Siliqua length**

It had a significant and positive association with thousand seed weight ( $r_g = 0.780^*$ ) at genotypic level while had non-significant and positive association with seed yield plant<sup>-1</sup> ( $r_g = 0.144$  and  $r_p = 0.095$ ) at both genotypic and phenotypic levels, number of seed siliqua<sup>-1</sup> ( $r_p = 0.047$ ) and thousand seed weight ( $r_p = 0.575$ ) at phenotypic level only but non-significant and negative correlation with number of seeds siliqua<sup>-1</sup> ( $r_g = -0.034$ ) at genotypic level only. The result was supported by Parveen (2007) and Kumari *et al.* (2017) where they observed seed yield plant<sup>-1</sup> had non-significant positive association with it but Singh *et al.* (2017) and Rauf and Rahim (2018) reported that seed yield plant<sup>-1</sup> had high significant positive association with this trait. Halder *et al.* (2014) reported seed yield plant<sup>-1</sup> had high significant negative association with siliqua length. Ejaz-Ul-Hasan *et al.* (2014) found positive and high significant genetic relationship between siliqua length and seeds siliqua<sup>-1</sup>.

#### **Number of seeds siliqua<sup>-1</sup>**

It showed non-significant positive interaction with thousand seed weight ( $r_g = 0.406$  and  $r_p = 0.477$ ) and seed yield plant<sup>-1</sup> ( $r_g = 0.571$  and  $r_p = 0.598$ ) at both genotypic and phenotypic levels. Non-significant association of these traits indicated large influences of environmental factors. The result was supported by Parveen (2007) and Kumari *et al.* (2017) while Jamali *et al.* (2016) and Rauf and Rahim (2018) observed positive and high significant association of this trait with seed yield plant<sup>-1</sup> at both genotypic and phenotypic levels.

#### **Thousand seed weight (g)**

It had significant and positive interaction with seed yield plant<sup>-1</sup> ( $r_g = 0.770^*$  and  $r_p = 0.768^*$ ) at both genotypic and phenotypic levels (Table 2 and 3), hence direct selection for this trait will be effective in improving the seed yield. Genotypic correlation coefficients were generally higher than their corresponding phenotypic correlations indicated strong hereditary association among the traits due to genetic factors such as linkage and/or pleiotropic effect enabling consistent performance and the masking effects of environment on these traits. The result was supported by Maurya *et al.* (2012), Hussain *et al.* (2014), Joya *et al.* (2016), Afrin *et al.* (2017) and Rauf and Rahim (2018) while Naznin *et al.* (2015) and Kumari *et al.* (2017), reported that it had non-significant positive interaction with seed yield plant<sup>-1</sup>. Yadava *et al.* (2011) and Kumari *et al.* (2017) found positive significant correlation of thousand seed weight with siliqua length and number of seeds siliqua<sup>-1</sup>.

Days to 50% flowering and 80% maturity showed high significant and positive correlation with seed yield plant<sup>-1</sup> at both genotypic and phenotypic level while plant height and thousand seed weight showed significant and positive correlation with yield plant<sup>-1</sup> at both genotypic and phenotypic level indicated yield plant<sup>-1</sup> would be increased if these associated traits increase, hence direct selection for these traits will be effective in improving the seed yield. Number of primary branches plant<sup>-1</sup>, siliqua length, Number of seeds siliqua<sup>-1</sup> showed non-significant and positive correlation with yield plant<sup>-1</sup> at both genotypic and phenotypic level while number of secondary branches plant<sup>-1</sup> showed non-significant and negative correlation at both genotypic and phenotypic level. Number of siliqua plant<sup>-1</sup> showed non-significant and negative correlation at genotypic level and significant negative correlation

at phenotypic level with yield plant<sup>-1</sup>. Non-significant association of these traits indicated large influences of environmental factors and therefore, selection for these traits would not be effective. However, such traits need further study on their direct and indirect effect on the grain yield plant<sup>-1</sup> by path coefficient analysis to remark as selection criteria for grain yield improvement programs.

### Path Coefficient Analysis

Path coefficient analysis was carried out in the present study (Table 4) considering yield plant<sup>-1</sup> as dependent character and its attributes as independent characters. Each component has two path actions viz. direct effect on yield and indirect effect through components which are not revealed by correlation studies.

**Table 4. Partitioning of genotypic correlations into direct (bold) and indirect effects of yield and yield related characters by path coefficient analysis in *Brassica rapa* genotypes**

Traits	Days to 50% flowering	Days to 80% maturity	Plant height (cm)	No. of primary branches plant <sup>-1</sup>	No. of secondary branches plant <sup>-1</sup>	No. of siliqua plant <sup>-1</sup>	Siliqua length (cm)	No. of seed siliqua <sup>-1</sup>	Thousand seed weight (g)	Genotypic correlation with yield
Days to 50% flowering	<b>5.4155</b>	-7.8909	-1.6033	-0.0211	0.9094	-0.2589	0.1365	-0.6117	-0.6571	0.987**
Days to 80% maturity	5.3693	<b>7.9587</b>	-1.7086	-0.0232	0.9062	-0.2203	0.2189	-0.5130	-0.6597	0.989**
Plant height (cm)	3.5295	-5.5276	<b>2.4601</b>	-0.0758	1.1725	-0.0838	1.2449	-0.2401	-0.8204	0.802*
No. of primary branches plant <sup>-1</sup>	1.0602	-1.7129	-1.7249	<b>-0.1081</b>	1.5037	-0.1713	0.7814	-0.1046	-0.6202	0.213
No. of secondary branches plant <sup>-1</sup>	-2.7909	4.0874	1.6346	0.0921	<b>-1.7646</b>	0.6213	-0.3284	0.4495	0.7731	-0.593
No. of siliqua plant <sup>-1</sup>	-1.5246	1.9063	0.2241	0.0201	-1.1920	<b>0.9197</b>	0.3909	0.7377	0.3089	-0.485
Siliqua length (cm)	0.4453	-1.0493	-1.8441	-0.0508	0.3490	0.2165	<b>1.6608</b>	0.0380	-0.4439	0.144
No. of seeds siliqua <sup>-1</sup>	3.0338	-3.7393	-0.5411	-0.0103	0.7264	-0.6214	-0.0579	<b>-1.0919</b>	-0.3602	0.571
Thousand seed weight (g)	4.0119	-5.9191	-2.2754	-0.0756	1.5380	-0.3203	0.8310	-0.4434	<b>-0.8871</b>	0.770*

Here, Residual effect = 0.094, \*\* = Significant at 1%, \* = Significant at 5%

### Days to 50% flowering

It showed very high positive direct effect (5.4155) towards yield plant<sup>-1</sup>. This result was supported by Uddin *et al.* (2013), Ejaz-Ul-Hasan *et al.* (2014) and Rashid *et al.* (2015) but Islam *et al.* (2016) and Rauf and Rahim (2018) who found that it had the negative direct effect on yield plant<sup>-1</sup>. Kumari *et al.* (2017) showed that it exerted negative indirect effect on seed yield but showed very high negative indirect effect towards yield plant<sup>-1</sup> via days to 80% maturity (-7.8909) and plant height (-1.6033) while negligible negative indirect effect via number of primary branches plant<sup>-1</sup> (-0.0211). It also showed highly positive indirect effect towards yield plant<sup>-1</sup> via number of secondary branches plant<sup>-1</sup> (0.9094) while high negative indirect effect via number of seeds siliqua<sup>-1</sup> (-0.6117) and thousand seed weight (-0.6571). However, moderate negative indirect effect was recorded towards yield plant<sup>-1</sup> via number of siliqua plant<sup>-1</sup> (-0.2589) and low positive indirect effect via siliqua length (0.1365). It had positive and high significant genotypic correlation (0.987\*\*) with yield plant<sup>-1</sup>.

### Days to 80% maturity

It showed very high positive direct effect (7.9587) towards yield plant<sup>-1</sup>. Ejaz-Ul-Hasan *et al.* (2014), Mekonnen *et al.* (2014), Naznin *et al.* (2015), Rashid *et al.* (2015) and Rauf and Rahim (2018) also found positive direct effect towards yield plant<sup>-1</sup> but Hussain *et al.* (2014) and Halder *et al.* (2016) reported direct negative association with yield plant<sup>-1</sup>. Further, it recorded high negative indirect effect towards yield plant<sup>-1</sup> via thousand seed weight (-0.6597) and plant height (-1.7086), negligible negative indirect effect via number of primary branches plant<sup>-1</sup> (-0.0232), moderate negative indirect effect via siliqua plant<sup>-1</sup> (-0.2203) and moderate positive indirect effect via siliqua length (0.2189). However, high negative indirect effect towards yield plant<sup>-1</sup> via number of seeds siliqua<sup>-1</sup> (-0.5130) and number of secondary branches plant<sup>-1</sup> (0.9062) and very high positive indirect effect via days to 50% flowering (5.3693). It showed positive and high significant genotypic correlation (0.989\*\*) with yield plant<sup>-1</sup>.

### **Plant height (cm)**

It exhibited very high positive direct effect (2.4601) towards yield plant<sup>-1</sup>. The result matched with Halder *et al.* (2016), Islam *et al.* (2016), Afrin *et al.* (2017), Karmokar (2018) and Rauf and Rahim (2018) while in the contrast, Uddin *et al.* (2013) and Rashid *et al.* (2015) found negative direct effect on yield plant<sup>-1</sup>. Here the genotypic correlation was positive and significant (0.802\*) with yield plant<sup>-1</sup> (Table 4). Further, very high positive indirect effect towards yield plant<sup>-1</sup> via days to 50% flowering (3.5295), siliqua length (1.2449) and number of secondary branches plant<sup>-1</sup> (1.1725) while very high negative indirect effect via days to 80% maturity (-5.5276) was recorded. Naznin *et al.* (2015) observed positive indirect effect on seed yield plant<sup>-1</sup> through siliqua length. It also showed high negative indirect effect towards yield plant<sup>-1</sup> via thousand seed weight (-0.8204). However, it was found negligible negative indirect effect towards yield plant<sup>-1</sup> via siliqua plant<sup>-1</sup> (-0.0838) and number of primary branches plant<sup>-1</sup> (-0.0758). It showed moderate negative indirect effect towards yield plant<sup>-1</sup> via seeds siliqua<sup>-1</sup> (-0.2401).

### **Number of primary branches plant<sup>-1</sup>**

It had low negative direct effect (-0.1081) towards yield plant<sup>-1</sup>. Alam (2010) and Islam *et al.* (2016) also recorded negative direct effect on yield plant<sup>-1</sup> but Halder *et al.* (2016), Afrin *et al.* (2017) and Rauf and Rahim (2018) reported that it had strong positive direct effect on seed yield. Further, it was recorded high positive indirect effect towards yield plant<sup>-1</sup> via days to 50% flowering (1.0602) and number of secondary branches plant<sup>-1</sup> (1.5037) while very high negative indirect effect via plant height (-1.7249) and days to 80% maturity (-1.7129). However, low negative indirect effect towards yield plant<sup>-1</sup> was recorded via seeds siliqua<sup>-1</sup> (-0.1046) and siliqua plant<sup>-1</sup> (-0.1713). It showed highly positive indirect effect towards yield plant<sup>-1</sup> via siliqua length (0.7814) and highly negative indirect effect via thousand seed weight (-0.6202). Here the correlation was non-significant and positive (0.213) with yield plant<sup>-1</sup>.

### **Number of secondary branches plant<sup>-1</sup>**

It showed very high negative direct effect (-1.7646) towards yield plant<sup>-1</sup>. Hussain *et al.* (2014), Islam *et al.* (2016) and Rauf and Rahim (2018) also found similar result but Naznin *et al.* (2015) and Rashid *et al.* (2015) reported that it had strong positive direct effect on seed yield. It also recorded high positive indirect effects to yield plant<sup>-1</sup> via seeds siliqua<sup>-1</sup> (0.4495), siliqua plant<sup>-1</sup> (0.6213) and thousand seed weight (0.7731) but had high negative indirect effect via siliqua length (-0.3284). However, it had very high positive indirect effect towards yield plant<sup>-1</sup> through days to 80% maturity (4.0874) and plant height (1.6346) while it had very high negative indirect effect via days to 50% flowering (-2.7909). It also showed negligible positive indirect effect towards yield via number of primary branches plant<sup>-1</sup> (0.0921). The genotypic correlation of this trait with yield plant<sup>-1</sup> (-0.593) was negative and non-significant.

### **Number of siliqua plant<sup>-1</sup>**

It exhibited high positive direct effect (0.9197) towards yield plant<sup>-1</sup>. This result was supported by Naznin *et al.* (2015), Rashid *et al.* (2015), Islam *et al.* (2016), Afrin *et al.* (2017) and Rauf and Rahim (2018) but Halder *et al.* (2016) reported that it had the greater negative direct effect on seed yield plant<sup>-1</sup>. Though it showed high positive direct effect on yield, but its correlation with yield was significant and negative at phenotypic level only. However, it showed highly positive indirect effect towards yield plant<sup>-1</sup> via siliqua length (0.3909), seeds siliqua<sup>-1</sup> (0.7377) and thousand seed weight (0.3089) while it had very high negative indirect effect via number of secondary branches plant<sup>-1</sup> (-1.1920) and days to 50% flowering (-1.5246). It also showed very high and moderate positive indirect effect towards yield plant<sup>-1</sup> via days to 80% maturity (1.9063) and plant height (0.2241) respectively but showed moderate positive indirect effect via number of primary branches plant<sup>-1</sup> (0.0201) (Table 4). The genotypic correlation of this trait with yield plant<sup>-1</sup> (-0.485) was negative and non-significant.

### **Siliqua length**

It showed very high positive direct effect (1.6608) towards yield plant<sup>-1</sup>. Hussain *et al.* (2014), Islam *et al.* (2016) and Rauf and Rahim (2018) also found positive direct effect but Rashid *et al.* (2015) and Kumari *et al.* (2017) found negative direct effect of it towards yield plant<sup>-1</sup>. High negative indirect

effect towards yield plant<sup>-1</sup> via thousand seed weight (-0.4439) and high positive indirect effect via number of secondary branches plant<sup>-1</sup> (0.3490) and days to 50% flowering (0.4453) while, very high negative indirect effects to yield plant<sup>-1</sup> via days to 80 % maturity (-1.0493) and plant height (-1.8441) was recorded. It also showed negligible positive and negative indirect effect via seeds siliqua<sup>-1</sup> (0.0380) and number of primary branches plant<sup>-1</sup> (-0.0508), respectively towards yield plant<sup>-1</sup> and moderate positive indirect effect via number of siliqua plant<sup>-1</sup> (0.2165) (Table 4). The genotypic correlation of siliqua length with yield plant<sup>-1</sup> (0.144) was positive and non-significant.

#### **Number of seeds siliqua<sup>-1</sup>**

It showed very high negative direct effect (-1.0919) towards yield plant<sup>-1</sup>. Hussain *et al.* (2014) and Rashid *et al.* (2015) also found similar result but Afrin *et al.* (2017) and Rauf and Rahim (2018) revealed greater direct positive contribution of it on seed yield. Further, it was recorded very high positive indirect effect towards yield plant<sup>-1</sup> via days to 50% flowering (3.0338) while very high negative indirect effect via days to 80% maturity (-3.7393). It also showed high negative indirect effect via thousand seed weight (-0.3602), plant height (-0.5411) and number of siliqua plant<sup>-1</sup> (-0.6214) while negligible negative indirect effect via number of primary branches plant<sup>-1</sup> (-0.0103) and siliqua length (-0.0579) towards yield plant<sup>-1</sup> while highly positive indirect effect towards yield plant<sup>-1</sup> via number of secondary branches plant<sup>-1</sup> (0.7264). It had non-significant and positive genotypic correlation (0.571) with yield plant<sup>-1</sup>.

#### **Thousand seed weight**

It showed high negative direct effect (-0.8871) towards yield plant<sup>-1</sup>. Alam (2010), Rashid *et al.* (2015) and Islam *et al.* (2016) also found negative direct effect of this trait towards yield plant<sup>-1</sup> while, Joya *et al.* (2016), Afrin *et al.* (2017) and Rauf and Rahim (2018) revealed the maximum direct positive effect towards yield plant<sup>-1</sup>. Naznin *et al.* (2015) found negative indirect effect for thousand seed weight towards yield plant<sup>-1</sup>. However, it showed high negative direct effect on yield but its correlation with yield was significant and positive (0.770\*). Further, it was recorded very high positive indirect effect towards yield plant<sup>-1</sup> via number of secondary branches plant<sup>-1</sup> (1.5380), siliqua length (0.8310) and days to 50% flowering (4.0119) while very high negative indirect effect via days to 80 % maturity (-5.9191) and plant height (-2.2754) but had high negative indirect effect via seeds siliqua<sup>-1</sup> (-0.4434) and number of siliqua plant<sup>-1</sup> (-0.3203) but negligible negative indirect effect via number of primary branches plant<sup>-1</sup> (-0.0756) towards yield plant<sup>-1</sup>.

#### **Residual effect**

The magnitude of residual effect (0.094) indicated that traits included in the path analysis explained about 90.6% of the variation in yield. However, the remaining variation in yield (9.4%) can be attained by incorporating other yield related traits in the path analysis as far as studies involving association of traits is concerned. Karmokar (2018) also found residual effect 0.091 while Naznin *et al.* (2015) found 0.450, Islam *et al.* (2016) found 0.430 and Ullah (2018) found 0.570 in case of yield plant<sup>-1</sup>.

Days to 50% flowering and 80% maturity, plant height, number of siliqua plant<sup>-1</sup>, siliqua length showed very high positive direct effect towards yield plant<sup>-1</sup> while thousand seed weight, number of primary and secondary branches plant<sup>-1</sup> and number of seeds siliqua<sup>-1</sup> had high positive indirect effect towards yield plant<sup>-1</sup> hence all the traits were the most important contributors to seed yield plant<sup>-1</sup> which could be taken in consideration for future hybridization program. Strong direct effect indicate selection of the traits might be effective for yield improvement. Low residual effect indicated respective traits of the study explained almost all the variability towards yield.

## **CONCLUSION**

Correlation analysis revealed that days to 50% flowering and 80% maturity, plant height and thousand seed weight had positive significant correlation with seed yield plant<sup>-1</sup> and path coefficient analysis indicated that days to 50% flowering and 80% maturity, plant height, number of siliqua plant<sup>-1</sup> and siliqua length showed very high positive direct effect towards yield plant<sup>-1</sup>, hence direct selection for such traits will be effective in improving the seed yield in *B. rapa*. The strong positive association

among the traits indicated that simultaneous selection for these characters would result in improvement of high yielding varieties.

## REFERENCES

- Afrin, F., Mahmud, F. and Islam, M.S. 2017. Genetic variability and character association in BC<sub>1</sub>F<sub>6</sub> population of *Brassica napus* L. MS. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.
- AIS. 2020. Agricultural Information Service. Krishi Diary (In Bangla). Khamarbari, Farmgate, Dhaka, Bangladesh, 16 pp.
- Alam, M.F. 2010. Variability studies in F<sub>4</sub> progenies of *Brassica rapa* obtained through inter-varietal crosses. M.S. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.
- Ali, N., Javidfar, F., Elmira, J.Y. and Mirza, M.Y. 2003. Relationship among yield components and selection criteria for yield improvement in winter rapeseed (*Brassica napus* L.). *Pakistan J. Bot.*, 35 (2): 167-174.
- Al-Jibouri, H., Miller, P.A. and Robinson, H.F. 1958. Genotypic and environmental variances and covariances in an upland cotton cross of interspecific origin. *Agron. J.*, 50 (10): 633-636.
- Dewey, D.R. and Lu, K.H. 1959. A correlation and path coefficient analysis of components of crested wheat grass seed production. *Agron. J.*, 51: 515-518.
- Ejaz-Ul-Hasan, Mustafa, H.S.B., Bibi, T. and Mahmood, T. 2014. Genetic variability, correlation and path analysis in advanced lines of rapeseed (*Brassica napus* L) for yield components. *Cercetări Agron.*, 1 (157): 71-79.
- Halder, T., Bhuiyan, M.S.R. and Islam, M.S. 2014. Variability and correlation study of some advanced lines of *Brassica rapa*. *Bangladesh J. Pl. Breed. Genet.*, 27 (1): 25-36.
- Halder, T., Bhuiyan, M.S.R., Islam, M.S. and Hossain, J. 2016. Analysis of relationship between yield and some yield contributing characters in few advanced lines of rapeseed (*Brassica rapa* L.) by using correlation and path analysis. *Adv. Agric. Bot.*, 8 (1): 32-42.
- Hosen, M. 2008. Variability, correlation and path analysis in F<sub>3</sub> materials of *Brassica rapa* L. M.S. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.
- Hussain, M.A., Hossain, M.S. and Bhuiyan, M.S.R. 2014. Genetic variability and character association of advanced lines in *Brassica rapa*. M.S. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.
- Islam, M.S., Haque, M.M., Bhuiyan, M.S.R. and Hossain, M.S. 2015. Estimation of genotypic and phenotypic coefficient variation of yield and its contributing characters of *Brassica rapa* L. *American-Eruasian J. Agril. Environ. Sci.*, 15 (10): 2029-2034.
- Islam, M.S., Haque, M.M., Bhuiyan, M.S.R. and Hossain, M.S. 2016. Path coefficient analysis and correlation coefficients effects of different characters on yield of *Brassica rapa* L. *Plant.* 4 (6): 51-55.
- Jamali, K.H., Mari, S.N., Soomro, Z.A., Soomro, S. and Khanzada, A. 2016. Correlation study on yield and yield contributing traits in *Brassica campestris* L. *Int. J. Life Sci.*, 10 (1): 1-7.
- Joya, S., Shamsuddin, A.K.M. and Nath, U.K. 2016. Genetic variability, heritability and characters associations in rapeseed (*Brassica napus* L.). *Bangladesh J. Pl. Breed. Genet.*, 29 (2): 11-16.
- Kahrizi, D. and Alaahvarand, T. 2012. Estimation and interrelationships of genetic variability parameters of some morpho-phenological traits in spring rapeseed (*Brassica napus* L.). *Asian J. Biol. Sci.*, 5: 358-364.
- Karmokar, D. 2018. Genetic study on the yield and quality traits of advanced breeding populations in *Brassica rapa* L. M.S. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.
- Kumari, S., Kumar, K. and Kumari, K. 2017. Study on correlation among different character pairs and path coefficient analysis in yellow sarson (*Brassica rapa*. Var. Yellow Sarson). *Prog. Agric.* 17 (1): 1-20.
- Lenka, D. and Mishra, B. 1973. Path coefficient analysis of yield in rice varieties. *Indian J. Agril. Sci.*, 43: 376-379.

- Mary, S.S. and Gopalan, A. 2006. Dissection of genetic attributes yield traits of fodder cowpea in F<sub>3</sub> and F<sub>4</sub>. *J Appl. Sci. Res.*, 2: 805-808.
- Maurya, N., Singh, A.K. and Singh, S.K. 2012. Inter-relationship analysis of yield and yield components in Indian mustard, *Brassica juncea* L. *Indian J. Pl. Sci.* 1 (23): 90-92.
- Mekonnen, T.W., Wakjira, A. and Genet, T. 2014. Correlation and path coefficient analysis among yield component traits of Ethiopian mustard (*Brassica carinata*). *Ethiopian J. Pl. Sci.* 2 (2): 89-96.
- Naznin, S., Kawochar, M.A., Sultana, S. and Bhuiyan, M.S.R. 2015. Genetic variability, character association and path analysis in *Brassica rapa* L. Genotypes. *Bangladesh J. Agril. Res.*, 40 (2): 305-323.
- Parveen, S. 2007. Variability study in F<sub>2</sub> progenies of inter-varietal crosses of *Brassica rapa* L. M.S. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.
- Rashid, M.H.U., Parveen, S. and Bhuiyan, M.S.R. 2015. Morphological attributes species identification of oleiferous *Brassica* species and better parents selection criteria for *Brassica juncea*. *Int. J. Cur. Res.*, 7 (9): 19847-19854.
- Rauf, M.A. and Rahim, M.A. 2018. Genetic variability studies among yield and its contributing traits in mustard (*Brassica napus* L.). *Adv. Zool. Bot.*, 6 (4): 101-108.
- Sabaghnia, N., Dehghani, H., Alizadeh, B. and Mohghaddam, M. 2010. Interrelationships between seed yield and 20 related traits of 49 canola (*Brassica napus* L.) genotypes in non-stressed and water-stressed environments. *Spanish J. Agric. Res.*, 8: 356-370.
- Shekhawat, G., Jadeja C., Singh, J. and Shekhawat, R.S. 2014. Character association studies among yield and its component characters in Indian mustard (*Brassica juncea* L. Czern and Coss). *The Biosean*, 9 (2): 685-688.
- Siddique, D.M., Chandio, S.A., Ahmed, N.S., Karloo, W.M., Pathan, K.A., Meghwar, L.B. and Laghari, M.A. 2017. Character association of *Brassica campestris* L. *J. Agril. Res.*, 55 (2): 249-265.
- Singh, P., Chauhan, V.V., Meena, J.S. and Mishra, D.C. 2013. Correlation and path coefficient analysis for yield and yield components in early generation lines of Indian mustard (*Brassica juncea* L.). *Curr. Adv. Agril. Sci.*, 5 (1):37- 40.
- Singh, A.P.K., Verma, O.P. and Kumar, K. 2017. Estimates of genetic variability parameters and interrelationships of morpho-physiological traits in yellow sarson (*Brassica rapa* L. var. yellow sarson). *Elect. J. Pl. Breed.*, 8 (2): 629 -635.
- Tripathi, N., Kumar, K. and Verma, O.P. 2013. Genetic variability, heritability and genetic advance in Indian mustard (*Brassica juncea* L. Czern and Coss) for seed yield and its contributing attributes under normal and saline/alkaline condition. *Int. J. of sci. Res.*, 6: 6-14.
- Uddin, M.S., Bhuiyan, M.S.R., Kabir, K., Shahjahan, M. and Rahaman, M.S. 2013. Variability study in F<sub>2</sub> progenies of *Brassica rapa*. *Int. J. Agric. Crop Sci.*, 6 (11): 676-683.
- Yadava, D.K., Giri, S.C., Vignesh, M., Vasudev, S., Yadav, A.K., Das, B., Singh, R., Singh, N., Mohapatra, T. and Prabhu, K.V. 2011. Genetic variability and trait association studies in Indian mustard (*Brassica juncea*). *Indian J. Agril. Sci.*, 81 (8): 712–716.
- Zahan, M.I. 2006. Morphological characterization and genetic diversity in oleiferous *Brassica* species. M.S. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.

## SHORT-TERM WATERLOGGING EFFECT ON GROWTH AND YIELD OF MUNGBEAN

N. Naher<sup>1</sup>, A.K.M.M. Alam<sup>2</sup> and O. Zannat<sup>3</sup>

### ABSTRACT

Waterlogging stress is one of the most atrocious environmental factors restricting the productivity of mungbean in tropical and subtropical region. So a pot experiment was conducted at Sher-e-Bangla Agricultural University, Dhaka, during April to July 2019 to evaluate the growth and yield attributes of mungbean under waterlogging condition. The experiment comprised of four mungbean varieties, Green Diamond, Crystal, Satin and Celera and two treatments control and waterlogging which was laid out in two factor Randomized Complete Block Design with three replications. The results showed that both the varieties and waterlogging treatment had significant influence on growth and yield traits of mungbean plant. Celera showed the highest (37.04 cm) plant height in both control and waterlogging condition (31.06 cm). Among the four varieties Crystal created the highest SPAD value (50.07 units) than other varieties. After waterlogging, new leaves initiated by Green Diamond in 39 DAS followed by Celera. Number of leaves was higher (11) in Green Diamond in control and it became 4.66 in waterlogging. Celera produced higher pod per plant both in control and waterlogging. The highest yield per plant containing variety was Celera both in control (5.30 g) and (4.19 g) in waterlogging. Whereas the lowest under control in Green Diamond (3.80 g) and under waterlogging in Crystal (1.02 g). The relative yield indicated that Celera is the most waterlogging tolerant (0.791), followed by Satin (0.563), Green Diamond (0.563) and Crystal (0.196).

**Key words:** water logging, growth, yield, Mungbean

### INTRODUCTION

Bangladesh is a low lying country and almost all of Bangladesh lies on the largest delta in the world. Sudden flooding is a common disaster in our country due to its geographical position. Around 6.77 lakh hectares of croplands have been damaged by the recent flood that affected about 61 lakh people in 28 districts. Crops on 5.32 lakh hectares were destroyed and the rest damaged (BBS, 2019). In this phenomenon waterlogging tolerant crop varieties which are nitrogen producing are a blessing for our country. In tropical and subtropical region, heavy rainfall in the rainy season frequently induces short time flooding in the crop field. Soil flooding occurs over vast regions throughout the world adversely affecting approximately 10% of the global land area (FAO, 2002). Soil flooding has long been identified as a major abiotic stress and the constraints it imposes on roots have marked effects on plant growth and development (Parent *et al.*, 2008). Pounding of water due to rainfall, particularly in clay soil hampers root respiration. The problem is wide spread under flash flood due to climate change. A complete crop failure due to flooding is not uncommon. The effect of flooding on plant is obviously a reduced exchange of gasses between the plants and the environment (Maberly and Spense, 1989). Oxygen deficiency is the main constraint for plants have to deal with in a flooded situation (Crawford and Brandle, 1996). Flooding-induced stress may affects directly on the guard cell causing stomatal closure and reduces photosynthetic capacity of plants (Bradford and Hsiao, 1982). Despite this fact, very little information is available on the physiological responses of mungbean to soil waterlogging. Mungbean [*Vigna radiata* (L.) Wilczek] is one of the important pulse crops of the world and is also known as greengram. Mungbean is a pulse crop under Fabaceae family, grown principally for its protein rich edible seeds. It is a short duration grain legume having wider adaptability and low input requirements. It is the third most popular pulse crop after chick pea and pigeon pea cultivated throughout India. Besides its utilization as food in many forms, haulms are used as fodder and green

---

<sup>1</sup> Professor, <sup>2</sup>MS Student, Dept. of Agroforestry and Environmental Science, Sher-e-Bangla Agricultural University, Dhaka-1207, <sup>3</sup> Principal Scientific Officer, PRC, BARI, Gazipur, Bangladesh

manure. It has the unique ability to fix the atmospheric nitrogen (58-109 kg ha<sup>-1</sup>) in symbiotic association with *Rhizobium* bacteria, which not only enables it to meet its own nitrogen requirement but also benefits the succeeding crops (Ali, 1992). Due to its short term growth, nitrogen fixing capability, soil reinforcement and prevention of soil erosion, it is superior to other legumes. Though, pulses have been playing a vital role in the diet, the per capita availability of pulses has declined from 60.7 g day<sup>-1</sup> in 1951 to 35.5 g day<sup>-1</sup> in 2007 as against the FAO/WHO's recommendation of 80 g day<sup>-1</sup>. Thus based on above statistics and the per capita availability, the production potential of pulses has to be increased substantially. It contains amino acids lysine, methionine and cysteine. It is rich in digestible protein (approximately 25–28 %) by virtue of N<sub>2</sub> fixation machinery and extensively grown in tropical and subtropical Asia because of its wider range of adaptability (Poehlman, 1991). This crop is fitted well in multi-cropping systems, because of its rapid growth and early maturity, results in the increase of small landholders' income and improvement of soil fertility (Nsoukpoe-Kossi *et al.*, 1999). In Bangladesh, mungbean is traditionally cultivated in the winter months, but may be cultivated year round if there any stress tolerance variety presence. But, now-a-days it is successfully growing in summer season (Kharif-I and Kharif II seasons). It contributed 6.5% of the total pulse production of the country. It ranks third both in acreage and production among the pulses (BBS, 2019)). The yield of mungbean is quite lower than other grain legumes. For increasing pulse production it is urgently needed to extent cultivation of pulse crops rapidly to all possible areas of Bangladesh. But the cultivation of pulse crop in that area is not easy because of the lack of flood tolerant varieties of this crop. It is needed to develop stress tolerant high yielding varieties of mungbean by combining together the tolerant character that may have been distributed sporadically in different related genotypes. With this consideration the present study was conducted to assess the growth and yield attributes of mungbean genotypes under imposed waterlogging conditions and to identify the relative flood tolerant variety of mungbean if any.

## MATERIALS AND METHODS

A pot was conducted at the research field of the Department of Agroforestry and Environmental Science, at Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka, Bangladesh with four mungbean varieties collected from Pulse Research Center, BARI, Gazipur and the experimental duration was April, 2019 to July, 2019. The experiment was laid out by two factors Randomized Complete Block Design (RCBD) with three replications such as factor A: mungbean varieties; V<sub>1</sub> - Green Diamond, V<sub>2</sub> - Crystal, V<sub>3</sub> - Satin and V<sub>4</sub> - Celera and factor B: Control (normal irrigation) and 72 hours artificial waterlogging condition. Each pot was filled with 8 kg previously prepared growth media (soil and cow dung mixture), compost (¼ of the soil volume) and 0.2g Urea 0.4g TSP and 0.12g MP. Eight seeds were sown in each pot at a depth of 1cm. Intercultural operations, weeding and other measures were taken when necessary. Pots were placed in the water chamber of brick built (4 × 1.5 × 1) meter<sup>3</sup>) after 24 days of germination for 72 hours. The water level was 3 cm high from the soil level. Data obtained for morphological and yield parameters were statistically analyzed by using Statistix 10 software to find out the significance of variation resulting from the experimental treatments. The mean values for all the treatments were accomplished by Duncan test. The significance of difference between pair of means was tested at 5% and 1% level of probability and means were compared using Least Significant Difference (LSD) test according to Gomez and Gomez (1984).

## RESULTS AND DISCUSSION

Waterlogging variably influenced the different mungbean varieties.

### **Plant height (cm)**

Waterlogging significantly influenced plant height of the different mungbean varieties (Table 1) and it was highest in Celera (31.06 cm). On the other hand Green Diamond, Crystal, Satin varieties respectively showed the height 27.65 cm, 26.64 cm, 18.48 cm at 38 DAS after tolerating 72 hours

waterlogging. After waterlogging, vegetative growth was being slow. Voesenek and Blom (1996) stated that the elongation of stems and petioles may enable plants to emerge from the water in aquatic and waterlogging tolerant terrestrial species. This variation in plant height might be recognized to the genetic characters. But if plants are reserved in the field for longer period the life span of plants are extended further 37 days giving new flowers and fruits. Waterlogging treatment caused reduction in plant growth in terms of leaf area and growth rate in all the genotypes and the level of reduction was more pronounced in sensitive genotypes, according to Solaiman *et al.* (2007); Pocięcha *et al.* (2008); Celik and Turhan (2011).

**Table 1. Effect of waterlogging on plant height (cm) of different mungbean varieties**

Variety	Waterlogging	
	Control	72 hrs waterlogging
Green Diamond	32.663	27.657
Crystal	28.477	26.647
Satin	35.077	18.487
Celera	37.040	31.067
CV (%)	3.02	
LSD (5%)	0.333	

#### Number of leaves plant<sup>-1</sup> and leaf length

The influence of waterlogging on the number of leaves plant<sup>-1</sup> and leaf length was significantly different in different mungbean varieties (Table 2). Highest numbers (11) of leaves were found at control in green diamond whereas lowest in Satin (8.00), which was statistically similar to that of Cerela (8.33) but significantly different from Crystal (9.00). In waterlogging, Crystal and Cerela showed the highest leaf number (5.00 for both) compared to that of Green Diamond and Satin (4.66 for both). The reason is that leaves are also very receptive to waterlogging stress; respiration changes in the leaf, leaf chlorophyll content, and photosynthetic assimilation have been detected during a waterlogging period (Parolin, 2000).

**Table 2. Effect of waterlogging on leaf number per plant and leaf length (cm) of different mungbean varieties**

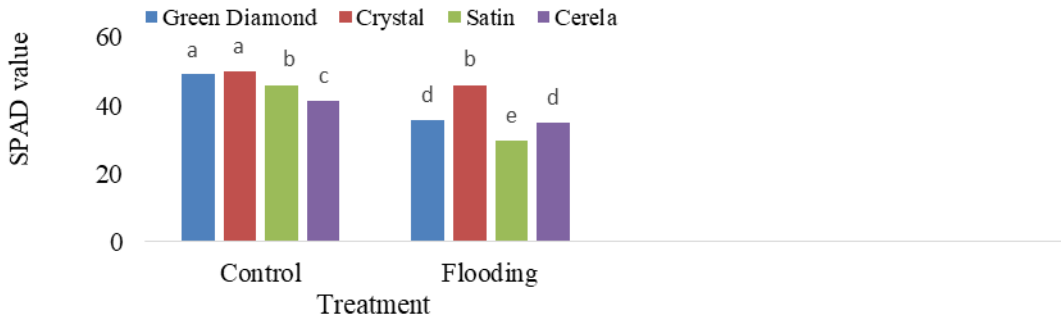
Variety × Treatments	No. of leaf at 38 days after waterlogging	Leaf length (cm)
Green Diamond × Control	11.00 a	9.15 b
Satin × Control	8.00 c	9.87 a
Crystal × Control	9.00 b	7.70 c
Celera × Control	8.33 bc	6.33 d
Green Diamond × Waterlogging	4.66 d	7.56 c
Crystal × Waterlogging	5.00 d	3.90 f
Satin × Waterlogging	4.66 d	7.26 c
Celera × Waterlogging	5.00 d	4.66 e
CV%	7.60	4.80

The highest leaf length (9.87 cm) was measured at control situation in Satin and lowest in Crystal variety (3.9 cm). Green Diamond, Satin, Celera gave 7.56, 7.26 and 4.66 cm leaf length respectively in waterlogging condition. Similar result was also observed by Islam (2005). This might be due to the senescence and abscission of lower leaves at maturity. Solaiman *et al.* 2007; Pocięcha *et al.* 2008;

Celik and Turhan (2011) reported that waterlogging treatment caused reduction in plant growth in terms of leaf area and growth rate.

### Leaf chlorophyll content

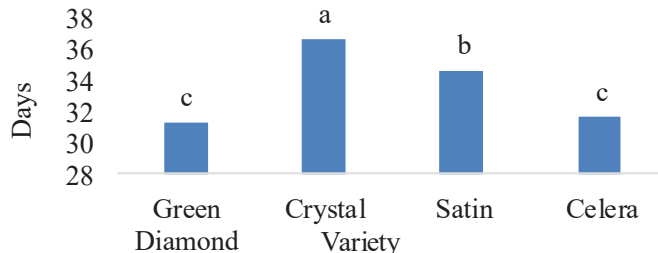
Waterlogging reduced chlorophyll content which was statistically significant (Fig.1). Green Diamond ( $V_1$ ), Crystal ( $V_2$ ), Satin ( $V_3$ ) and Celera ( $V_4$ ) contained 49.30, 50.07, 46.01 and 41.25 SPAD unit respectively, in control. On the other hand, after facing waterlogging stress chlorophyll content of each variety was reduced extremely. It was measured after waterlogging in mungbean varieties respectively 35.733, 46.03, 29.53, and 35.016 SPAD unit. Highest reduction percentage of chlorophyll content observed in Satin ( $V_3$ ), and less reduction detected in Crystal ( $V_2$ ) followed by Celera ( $V_4$ ).



**Fig.1.** Effects of waterlogging on leaf chlorophyll content of different mungbean varieties after 39 DAS [LSD<sub>0.05</sub> = 0.693]

### Days required to turning into normal condition of plant after waterlogging

After 72 hours waterlogging, plants became wilted. Each variety took different time duration to turn into normal condition from wilting which was significantly different. Celera and Green Diamond being normal in 31~32 DAS. On the other hand Crystal took 36 DAS and Satin took 34 DAS (Fig. 2). Some leaves were abscised for more water uptake. Plants had to operate their energies into renewed pigment production, and re-greened chlorotic leaves at the onset of recovery (Smethurst *et al.*, 2005). Relatively waterlogging-tolerant genotypes had an altered root distribution (i.e. near the soil surface) pattern while grown in waterlogged conditions as demonstrated by shallow root system-root length was short (~100 mm) in waterlogged plants. It is promising that during the recovery period the shallow root resumed growth and reached the same length as in the drained control, allowing access to soil moisture at depth as the soil profile dries later in the season (Malik *et al.*, 2001).



**Fig. 2.** Required days to turn into normal condition for mungbean varieties [LSD 0.05 = 1.89]  
Waterlogging effects on new leaf initiation

New leaf initiated after waterlogging condition and plants being vigorous day after day. The first day of initiated leaf was difference among varieties which was statistically significant (Fig. 3). Firstly in Green diamond leaf initiated at 39.66 days after emergence that was after 12 days of waterlogging then

in Celera, it took 40.33 days. Others two variety Crystal and Satin took 16 days and 19 days after waterlogging. Leaf initiation increase photosynthesis rate and its must be effective for yield.

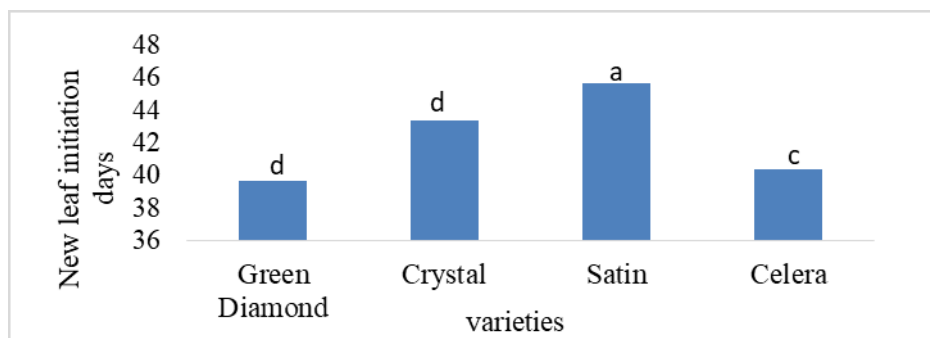


Fig. 3. Required days to initiation of new leaf after waterlogging condition [LSD 0.05 =0.490]

### Effects of waterlogging on 50% flowering of mungbean varieties

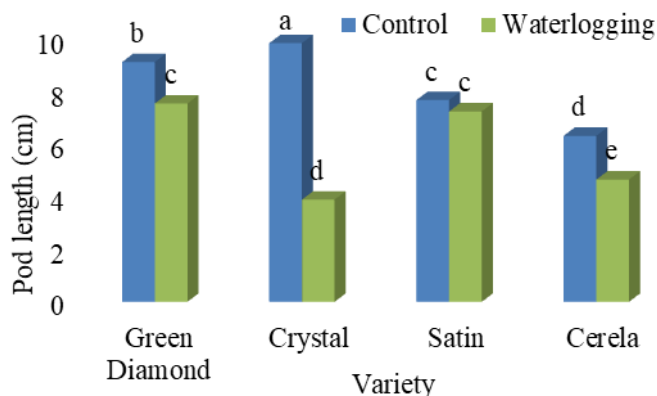
Effect of waterlogging on days to 50% flowering of four mungbean genotypes was significantly different from each other and they took 10-12 more days than control environment. Crystal and Satin took 34.33 and 34.3 days to 50% flowering, respectively (Table 3). Indeterminate plants flower until some environmental condition triggers them to stop. Too cold, too hot, too dry or too wet can trigger indeterminate plants to stop flowering. Similarly Kumar *et al.* (2013) reported that both tolerant and sensitive Mungbean genotypes showed the inhibition of flowering and pod setting under waterlogging.

Table 3. Effect of waterlogging on days to 50% flowering and pod number of different mungbean varieties

Variety × Treatments	Days of 50% flowering	Pod number
Green Diamond × Control	31.00 e	12.33 c
Satin × Control	34.33 d	10.33 d
Crystal × Control	34.33 d	14.00 b
Celera × Control	29.00 f	20.00 a
Green Diamond × Waterlogging	43.00 b	9.33 de
Crystal × Waterlogging	47.33 a	7.66 f
Satin × Waterlogging	47.66 a	8.66 ec
Celera × Waterlogging	41.00 c	12.33 c
CV%	2.63	7.33

### Effects of waterlogging on pod number and pod length of mungbean

The number of pods per plant was found significantly different for waterlogging. The highest (20) number of pods per plant was obtained from control (Table 3) in variety Celera and the lowest (7.66) from Satin after waterlogging. Similar result was also observed in legumes by Solaiman *et al.*, (2007), Pocięcha *et al.* (2008) under waterlogging condition. Islam (2005) reported that significantly reduced pods plant<sup>-1</sup> in mungbean and 36% more pods were produced in control plants than waterlogged plants. Varieties showed significant difference in pod length (Fig. 4). The longest pod length (9.87cm) was recorded in V<sub>1</sub> (Green Diamond) and the shortest (3.9 cm) in V<sub>2</sub> (Crystal) in waterlogging. These results have the agreement with the results of Sarkar *et al.* (2004) who described that pod length differed from varieties to varieties. The possible reason of this difference could be the genetic make-up of the varieties.



**Fig. 4.** Effect of waterlogging on pod length (cm) of different mungbean varieties [LSD<sub>0.05</sub>=0.173]

**Effect of waterlogging on number of seeds per pod, hundred fresh seed weight (g) and dry weight (g) of different mungbean varieties**

Waterlogging reduced the number of seeds per pod significantly over the control. Among the varieties, Celera recorded highest number of seeds (11) per pod. Without Celera in all other varieties seeds per pod reduced in the waterlogging (Table 4). Similar differences in genotypes were also observed in green gram (Laosuwan *et al.*, 1994 and Yadav and Saxena, 1998). The highest 100-seed weight was found in Crystal (8.00 g) at control. Green Diamond and Celera showed less reduction of 100-seed weight from control to waterlogging, this was respectively in control 3.97g, 3.71g and after waterlogging stress 2.59 g, 3.57 g. Genotypic variation in 1000-seed weight was observed by Tomar *et al.* (1996) in mungbean that also supported the present experimental results. The maximum 100 seed dry weight (7.25g) was observed in Crystal at control and minimum (2.03 g) in same variety by Green Diamond (2.25 g).

**Table 4.** Effect of waterlogging on seed per pod, 100 fresh seed weight, dry seed weight and yield of different mungbean varieties

Variety × Treatments	No. of Seed per pod	100 fresh seed wt. (g)	Oven dry wt. of 100 seed (g)	Yield/plant (g)
Green Diamond × Control	10.00 ab	3.97 b	3.59 b	3.80 d
Satin × Control	11.00 a	8.00 a	7.25 a	4.60 bc
Crystal × Control	10.00 b	7.85 a	7.12 a	5.20 ab
Celera × Control	11.00 ab	3.71 b	3.36 b	5.30 a
Green Diamond × Waterlogging	8.66 c	2.59 cd	2.25 c	1.23 f (0.324)
Crystal × Waterlogging	10.00 b	2.27 d	2.03 c	1.02 f (0.196)
Satin × Waterlogging	8.66 c	2.99 c	2.53 c	2.59 e (0.563)
Celera × Waterlogging	11.00 a	3.57 b	3.2 b	4.19 cd (0.791)
CV%	4.15	7.17	4.80	10.49

Figures in the parenthesis indicate relative seed yield

### **Effect of waterlogging on yield per plant (g) of mungbean**

The effect of waterlogging on yield per plant of different mungbean varieties was also varied significantly (Table 4). The highest yield was found in Celera (3.08 g), then satin (5.2 g), Crystal (4.6 g) and Green Diamond (3.80 g). In Control according to yield performance per plant the varietal series is Celera > Satin > Crystal > Green Diamond. On the other hand, in waterlogging, Celera also contained the highest yield (4.19 g) per plant and it was not much reduced like other varieties. The varietal series by yield per plant in waterlogging treatment is Celera > Satin > Crystal > Green Diamond. Similar results were also reported in greengram (Laosuwan *et al.*, 1994, and Ahmed *et al.* 2002); in blackgram (Pallavi *et al.*, 2004) and in soybean (Sorte *et al.*, 1996. Reduction in seed yield under waterlogged condition was due to oxygen deficiency and anaerobic conditions and less root activity. It was fundamentally due to diminish of water absorbing ability of the plants as specified by the reduction in leaf turgidity as well as translocation of dry matter from the vegetative growth to the reproductive structures (seeds) possibly due to damage caused to the root system. Such inhibition may also be due to adverse effects of waterlogging on water and mineral uptake. Reduction in seed yield was mainly due to impairment of water absorbing ability of the plants or inhibition of synthesis and transportation of photosynthetic assimilate (Kumar *et al.*, 2013).

## **CONCLUSION**

Waterlogging stress is one of the most atrocious environmental factors restricting the productivity of mungbean. Considering the findings it may be concluded that plant height, leaf number, seed moisture percentage and chlorophyll content reduced significantly due to waterlogging. Highest plant height was found in Celera (37.06 cm) and shortest was in Satin (18.48 cm) at waterlogging condition. Yield reduced significantly in waterlogging condition. Maximum number of pods (12.33) per plant obtained from Celera and the minimum was in crystal (7.66) at waterlogging. The highest 100-seed weight in Celera (3.57 g) and the lowest in Crystal (2.27 g) at waterlogging. The Celera was more waterlogging stress tolerant than Green Diamond, Crystal and Satin. Therefore, Celera can be added in the existing cropping pattern at short term waterlogging condition in tropical and subtropical regions.

### **Acknowledgement**

The PI gratefully acknowledge the financial support of SAURES, Sher-e-Bangla Agricultural University, Dhaka which enabled to conduct the research at Agroforestry and Environmental Science Department, Sher-e- Bangla Agricultural University, Dhaka, Bangladesh.

## **REFERENCES**

- Ahmed, S., Higuchi, H., Nawata, E. and Sakuratani, T. 2002. Effects on exogenous ABA and ethylene application and waterlogging on photosynthesis in mungbean (*Vigna radiata* (L.) Wilczak) *Japan J. Trop. Agric.* 46: 166–174.
- Ali, M. 1992. Weeds are a great threat to kharif pulses. *Indian Farming*, 42: 27–30 (Field crop Absts; 46: 3616; 1993).
- Bradford, K.J. and Hsiao, T.C. 1982. Stomatal behaviour and water relations of waterlogged tomato plants. *Plant Physiol.* 70: 1508–1513.
- BBS (Bangladesh Bureau of Statistics). 2019. Yearbook of Agricultural Statistics of Bangladesh, Ministry of Planning, Govt. People's Republic of Bangladesh, Dhaka, Bangladesh.
- Crawford, R.M.M. and Braendle, R. 1996. Oxygen deprivation stress in a changing environment *J. Exp. Bot.* 47: 145–159.
- Celik, G. and Turhan, E. 2011. Genotypic variation in growth and physiological responses of common bean (*Phaseolus vulgaris* L.) seedlings to flooding. *Afr. J. Biot.* 10: 7372–7380.

- FAO, 2002. Agriculture. <http://www.fao.org/waicent/FAOINFO/AGRICULT/ag1/gaez//nav.html>.
- Gomez, K.A. and Gomez, A.A. 1984. Statistical Procedures for Agricultural Research. 2. New York: John Wiley and Sons.
- Islam, M.R. 2005. Response of mungbean to flooding at vegetative stage I. Root and shoot growth. *Bangladesh Agron. J.* 11(1 & 2): 1-9.
- Kumar, P., Pal, M., Joshi, R. and Sairam, R.K. 2013. Yield, growth and physiological responses of mungbean [*Vigna radiata* (L.) Wilczek] genotypes to waterlogging at vegetative stage. *Physiol. Mol. Biol. Plants.* 19(2): 209-220.
- Laosuwan, P., Mekanawakul, M and Thongsomsri, A. 1994. The Effect of waterlogging on growth, development and yield of mungbean. *J. Sci. Tech.* 1: 9-14.
- Maberly, S.C. and Spence, D.H.N. 1989. Photosynthesis and photorespiration in freshwater organisms: amphibious plants. *Aquatic Botany.* 34: 267–286.
- Malik, A.I., Colmer, T.D., Lamber, H. and Schortemeyer, M. 2001. Changes in physiological and morphological traits of roots and shoots of wheat in response to different depths of waterlogging. *Aust. J. Plant. Physiol.* 28: 1121- 1131.
- Nsoukpoe-Kossi, C.N., AgIvanov, A.G., Veeranjaneyulu, K. and Leblanc, R.M. 1999. Protective action of abscisic acid against the inhibition of photosynthesis of barley leaves by bisulphate. *Photosynthetica.* 36: 51–60.
- Pallavi, E.B., Chore, C.N., Deotale, R.D., Ratnaparkhi, V.P., Phad, K.M and Yenpedriwar, M.N. 2004. Effect of waterlogging on biochemical, yield and yield contributing parameters in blackgram. *J. Soils and Crops.* 14(1):76-78.
- Parent, C., Berger, A. Folzer, H., Dat, J., Crevecoeur, M., Badot, P.M. and Capelli, N. 2008. A novel nonsymbiotic hemoglobin from oak: Cellular and tissue specificity of gene expression. *New phytol.* 177: 142-154.
- Parolin, P. 2000. Growth, productivity and use of trees in white water floodplains. In: The Central Amazon floodplain: actual use and options for a sustainable management. Backkhuy Publishers, Leiden. 375-391pp.
- Pociecha, E. Koscielniak, J. and Filek, W. 2008. Effect of root flooding and stage of development on the growth and photosynthesis of field bean (*Vicia faba* L. minor). *Acta Physiol. Plant.* 30: 529-535.
- Poehlman, J.M. 1991. The Mungbean. New Delhi: Oxford and IBH. 375 p.
- Sarker, M.A.R., M.H. Kabir, M. Begum and M.A. Salam, 2004. Yield performance of mungbean as affected by planting date and planting density. *J. Agron.*, 3: 18–24.
- Smethurst, C.F., Garnet, T. and Shabala, S. 2005. Nutrition and chlorophyll fluorescence responses of lucerne (*Medicago sativa*) to waterlogging subsequent recovery. *Plant Soil.* 270 (1–2): 31-45.
- Solaiman, Z., Colmer, T.D., Loss, S.P., Thomson, B.D. and Siddique, K.H.M. 2007. Growth responses of cool-season grain legumes to transient waterlogging. *Aust. J. Agric. Res.* 58: 406–412.
- Sorte, N.V., Deotah, R.D., Meshram, J.H. and Chanekar, M.A. 1996. Tolerance of soybean cultivars of waterlogging at various growth states. *J. Soil Crops.* 6: 68– 72.
- Tomar, S.S. and Tiwari. 1996. Response of green gram and black gram genotypes to plant density. *Gujrat Agric. Univ. Res. J.* 21: 88-92.
- Voesenek, L. and Blom. C.W.P.M. 1996. Plants and hormones: an ecophysiological view on timing and plasticity. *J. Ecol.* 84: 111-119.
- Yadav, R.S. and Saxena, H.K. 1998. Response of waterlogging on growth and seed yield of mungbean [*Vigna radiata* (L.) Wilczek]. *Ind. J. Plant Physiol.* 3: 71- 72.

## PROFITABILITY, PROBLEMS AND POSSIBLE SOLUTION OF BEAN CONTRACT FARMING IN BANGLADESH

M. R. Hasan<sup>1</sup>

### ABSTRACT

Contract farming may be solving production and marketing problems of many developing countries which gain importance. To investigate the profitability and problems of contract farming, 120 contract farmers and 180 non-contract farmers were randomly selected from Narsingdi district of Bangladesh. Descriptive statistics were used to analyze the data and conventional cost and revenue analysis were used to investigate the profitability of bean production. Result showed that bean contract farmers had 7.31 years of schooling whereas non-contract farmers had 2.75 years of schooling. Contract farmers got higher prices (Tk. 41.32) for their bean than for the non-contract farmers (Tk. 31.22). Net farm income of contract farmers was Tk. 789677.22/ha, whereas Tk. 457589.30/ha for non-contract farmers. Total 96.67% of contract farmers had verbal commitments with companies for the contract and only 3.33% of contract farmers had written document. Most of the farmers (82.5%) had marketing contract with different companies. 51.67% of contract farmers got cash support from the contract companies and 17.5% of contract farmers got training from different companies. 86.67% of the contract farmers were satisfied for their contract with different companies. Both contract and non-contract bean producing farmers mentioned their problems and they also suggested some solutions to solve their problems. Government monitoring and supervision are necessary to expand contract farming and to solve existing problems of contract farming in Bangladesh.

**Keywords:** contract farming, bean, profitability, price, problems

### INTRODUCTION

Bangladesh is highly densely populated country and total population of Bangladesh is 168.22 million. The growth rate of population of Bangladesh is 1.30 and population density is 1140 person per sq. km (BBS, 2021a). Bangladesh is predominantly an agrarian economy where most of the poor people live in rural areas and reliant on agriculture for their livelihood and food security (Alam *et al.*, 2018). The share of agriculture to GDP is decreasing but still agriculture is important sector in Bangladesh. According to the provisional calculation of BBS, the contribution of agriculture to the GDP in the fiscal year 2021-22 is about 11.50 percent (BER, 2022). Vegetables contain vitamins and minerals and vegetables is common for Bangladeshi meals. Bangladesh is suitable for producing various vegetables (more than 142 types of home-grown and exotic vegetables produced in the country) due to fertile land and environment (BARI, 2017-18). Bangladesh retained 3<sup>rd</sup> position in global vegetables production (FAO, 2017). Total area of vegetable production was 1121618.24 acres and total production was 4729441.73 metric tons (BBS, 2021b).

Contract farming gained importance in many developing countries, facilitating the coordination in modern agricultural supply chains (Mishra *et al.*, 2018). Contract farming is perceived as a strategy for agricultural transformation in developing countries because of its potential to address agricultural marketing and production challenges (Bidzakin *et al.*, 2020). Contract farming will necessarily to emerge when market failure may appears while uncertainty and commodity specificity are high, the trade products that are perishable and difficult to store and transport the products (Soullier and Moustier, 2018; Minot and Sawyer, 2016). Contract farming is an intermediary form of vertical coordination in agricultural production (Hoang, 2021). Contract farming is as an efficient mechanism to link smallholder farmers to high-value supply chains (Nguyen *et al.*, 2015). Allen and Lueck (1995) note that contract farming can be used to manage production and marketing risk and these risks are the

---

<sup>1</sup>Associate Professor, Dept. of Agribusiness and Marketing, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh

major risks that smallholder households face in developing and emerging economies. Contract farming is an agreement between farmers and firms in producing and providing agricultural products with a certain price (Eaton and Shepherd, 2001). Contract farming may be either oral or written agreement between growers and buyers or processors (Mulatu *et al.*, 2017). According to Freguin-Gresh *et al.*, (2012), the buyer can be a local or a transnational agribusiness (processor, exporter, retail outlet or shipper), a private plantation, or the local merchants (greengrocers, wholesalers, hawkers, brokers, etc.).

Contract farming has been criticized for agribusiness companies to exploit an unequal power relationship with farmers (Porter and Philips-Howard, 1995; Key and Runsten, 1999 and Singh, 2002). Criticism of contract farming is that the provision of input and a fixed price may be perceived as a disadvantage of contract farming that restricts farmers to accessing better sources of seed, fertilizer, credit, and technical assistance as well as selling in spot markets to obtain a higher price and income (Hoang, 2021). Market failures of contract farming due to the lack of legal enforcement of contracts observed in developing countries (McMillan and Woodruff, 1999; Fafchamps, 1996; Fafchamps and Minten, 2001). Bean is producing all over Bangladesh and it is a common vegetable for all consumers. Total area of bean production was 55076.58 acres and total production was 169655.90 metric tons in Bangladesh (BBS, 2021b).

Previous studies (Hasan *et al.*, 2014; Chowdhuri *et al.*, 2014) found that bean production was profitable in Bangladesh. However, no study investigated bean contract farming and find out contract farmers problems. The present study not only analyzes profitability of bean contract farming but also compare profitability of bean contract farming with non-contract farming. Bean contract farming is an emerging concept in Bangladesh and it is challenging to collect data. It is assume that contract farming assist farmers to market their product as well as ensure quality bean production. Comparing profitability, production problems and find out possible suggestions of contract and non-contract bean producing farmers are the new aspect of this research.

In this context, the study was conducted to investigate the socio-economic characteristics of bean producing contract and non-contract farmers. The research compares the profitability of bean producing contract and non-contract growers. Finally the study highlights major problems and find out possible solution both for contract and non-contract farmers.

## METHODOLOGY

### Study area and sample

Narsingdi district is the major vegetable producing area in Bangladesh with available country bean growers. Two Upazila namely Shibpur and Belabo Upazila of Narsingdi district were purposively selected for the present study. These two Upazila were famous for vegetable production and vegetable contract farming. Twelve villages were randomly selected from two Upazila and six villages were randomly selected from one Upazila. A full list of country bean producing farmers was collected from the Upazila agriculture office. Total 1181 bean producing farmers from 12 villages were listed, which served as a sampling framework for the study. First, divide two farmers group as contract and non-contract farmers. Then farmers were randomly selected by using random sampling. Total 120 contract farmers and 180 non-contract farmers were selected from the farmer's list. Finally, 300 country bean producing farmers were selected for the present study. Two pre-tested interview schedules were used for face-to-face data collection. One interview was for contract farmers and another for non-contract farmers. Different questions were asked to the farmers in a systematic and simple way. Data were collected from February to April, 2022 to achieve the objectives.

### Cost and revenue analysis

Descriptive statistics were mainly used to analyze the data. The revenues and costs of bean production were calculated for four months. The study used total variable cost, total fixed cost, total cost, total revenues, gross farm income, net farm income, revenues over variable cost and revenues over total cost to calculate profitability. This conventional farm income calculation was used by Hasan and Bai, (2016). Total variable cost was the sum of seed, power tiller, labor, fertilizer, cow dung, organic

fertilizer, pesticides, irrigation and cost for structure including bamboo. Fixed cost constituted by family labor cost, interest on operating capital and land rent. Total cost was the sum of total variable and fixed cost. Gross farm income was the outcome from total variable cost deducted from total revenue. Net farm income was the result from total revenue minus total cost. Revenue over variable cost was the ratio of total revenue and total variable cost and revenue over total cost was the ratio of total revenue and total cost. The interest on operating capital was calculated for four months considering 6.0% interest rate.

### Description of variables

Table 1 describes the variables used for farmer's characteristics and their measurement. Table 2 describes the variables used for production, inputs costs, revenue and their measurement.

**Table 1. Description of variables related with farmers characteristics**

Variables	Measurement
Age	Years
Schooling	Years
Farming experience	Years
Adult household member	Numbers
Household size	Numbers
Total operating land	Decimals
Bean cultivation land	Decimals
Credit amount	Taka
Extension contact	Numbers/Year
Training	Numbers

**Table 2. Description of variables related with production, inputs costs and revenue**

Variables	Measurement
Bean cultivation area	ha
Total production	Kg
Market price	Tk./kg
Seed	kg
Power tiller	Number of operation
Labor	Number
Fertilizer	Kg
Cow dung	Kg
Organic fertilizer	Kg
Pesticide	Liter
Irrigation	Number of operation
Interest on operating capital	four months
Land rent	Tk./ha/season
Average duration of the contract	Year

Note: Tk. means Taka which is the national currency of Bangladesh

## RESULTS AND DISCUSSION

### Personal characteristics of contract farmers and non-contract farmers

Significant t test indicated that there were significant differences between contract and non-contract farmer schooling, bean cultivation land and credit amount. The years of schooling were much higher for the contract farmers (7.31 years) than for non-contract farmers (2.75 years) indicating that educated farmers adopted more contract farming.

**Table 3. Personal characteristics of contract and non-contract farmers**

Items	Contract farmer		Non contract farmer		Mean difference
	Mean	S.D.	Mean	S.D.	T test
Age	45.55	11.97	45.28	12.95	-0.18
Schooling	7.31	2.51	2.75	1.92	-17.78***
Farming experience	20.47	11.71	21.73	12.42	0.88
Adult household member	3.70	1.44	3.72	1.42	0.082
Household size	6.0	1.73	5.86	1.91	-0.67
Total operating land	124.25	69.67	114.02	61.89	-1.33
Bean cultivation land	40.15	34.57	27.94	15.13	-4.17***
Credit amount	24541.67	41262.41	17477.78	26239.43	-1.81*
Number of extension contact	4.95	3.27	4.6	3.20	-0.94
Number of training	0.65	0.47	0.62	0.48	-0.39

Source: Farmer's household survey, 2022

Note: \*\*\*, \*\* and \* indicates significance at the 1%, 5% and 10% levels respectively

The bean cultivation land was higher for the contract farmers (40.15 decimals) than for non-contract farmers (27.94 decimals) imply that contract farmers cultivated more land for bean production. The credit amount was higher for contract farmers (Tk. 24541.67) than for non-contract farmers (Tk. 17477.78) indicating that contract farmers took more loan than non-contract farmers. Contract farmers had 20.47 years of farming experience whereas non-contract farmers had 21.73 years of farming experience. Contract farmers contacted with extension workers 4.95 times/year however contract farmers contacted with extension workers 4.6 times/year indicating that contact farmers more contacted with extension worker.

#### **Cost of bean production for contract farmers in the study area**

Contract farmers used 8.44 kg seed in a hectare of land and average seed price was Tk. 476.83 (Table 4). On average, farmer spent Tk. 11533.68 for power tiller, which was 2.88% of total cost. Farmers

**Table 4. Cost of per hectare bean production for contract farmers in the study area**

Cost items	Price	Total quantity	Total cost (Tk.)	%
Seed	476.83	8.44	4024.44	1.0
Power tiller	467.19	24.68	11533.68	2.88
Labor	-	-	157578.55	39.28
Family labor	506.75	164.17	83193.14	20.74
Hired labor	506.75	146.78	74385.41	18.54
Fertilizers	-	-	23014.81	5.73
Urea	20.13	157.17	3163.83	0.79
Triple Super Phosphate	25.0	310.98	7774.5	1.94
Diammonium Phosphate	34.16	83.77	2861.58	0.71
Muriate of Potash	19.81	104.39	2067.96	0.52
Zinc Sulfate	198.08	16.05	3179.18	0.79
Gypsum	10.30	79.16	815.34	0.20
Boric Acid	203.12	15.52	3152.42	0.79
Cow dung	1.61	6701.67	10789.68	2.69
Organic fertilizer	401.43	80.57	32343.21	8.06
Pesticides	438.10	68.25	29903.88	7.45
Irrigation	394.93	23.65	9343.56	2.33

Cost items	Price	Total quantity	Total cost (Tk.)	%
Cost for structure including Bamboo	-	-	44113.64	10.99
Total variable cost	-	-	322645.45	80.43
Interest	-	-	3226.45	0.80
Land rent	-	-	75265.27	18.76
Total fixed cost	-	-	78491.72	19.57
Total cost	-	-	401137.17	100.0

Source: Farmer's household survey, 2022

used 164.17 man-day family labor and 146.78 man-day hired labor in a hectare of land for bean production. They spent Tk. 83193.14 for family labor and Tk. 74385.41 for hired labor, which was 39.28% of total cost. Farmers used urea, triple super phosphate, diammonium phosphate, muriate of potas, zinc sulfate, gypsum and boric acid in their land. They spent Tk. 23014.81 for different fertilizers, which was 5.73% of total cost. Moreover, they spent Tk. 10789.68 for cow dung, which was 2.69 % of total cost. They used 80.57 kg organic fertilizers and average fertilizers price was Tk. 401.43/kg. They spent Tk. 32343.21 for organic fertilizers which were 8.06 % of total cost. They used 68.25 liters of pesticides and average price of pesticides was Tk. 438.10. They spent Tk. 29903.88 for different pesticides, which was 7.45% of total cost. Irrigation cost was Tk. 9343.56, which was 2.33% of total cost. Cost for structure including bamboo was Tk. 44113.64, which was 10.99% of total cost. Interest on operating capital (four months) was Tk. 3226.45 and land rent cost was Tk. 75265.27. Total variable cost was Tk. 322645.45, which was 80.43% of total cost. On the other hand, total fixed cost was Tk. 78491.72, which was 19.57% of total cost. Total cost of bean production was Tk. 401137.17/ha.

#### Cost of bean production for non-contract farmers in the study area

Non-contract farmers used 8.94 kg seed and they spent Tk. 3873.43 in a hectare of land (Table 5). On average, farmer spent Tk. 12012.04 for power tiller. They spent Tk. 84444.85 for family labor and Tk. 75436.99 for hired labor, which was 38.35% of total cost. They spent Tk. 24247.03 for different fertilizers, which was 5.82% of total cost. Moreover, they spent Tk. 12658.57 for cow dung, which was 3.04% of total cost. They used organic fertilizers and average fertilizers price was Tk. 417.55/kg . Farmers spent Tk. 37663.01 for organic fertilizers which were 9.04% of total cost. They used 74.05 liter of pesticides and average price of pesticides was Tk. 403.46. They spent Tk. 29880 for different pesticides, which was 7.16% of total cost. Irrigation cost was Tk. 10942.29, which was 2.63% of total cost. Cost for structure including bamboo was Tk. 46249.94, which was 11.09% of total cost. Interest on operating capital (four months) was Tk. 3374.08 and land rent cost was Tk. 76024.1. Total variable cost was Tk. 337408.15, which was 80.95% of total cost. On the other hand, total fixed cost was Tk. 79398.18, which was 19.05% of total cost. Total cost of bean production was Tk. 416806.33/ha.

**Table 5. Cost of per hectare bean production for non-contract farmers in the study area**

Cost items	Price	Total quantity	Total cost (Tk.)	%
Seed	433.27	8.94	3873.43	0.93
Power tiller	365.11	32.89	12012.04	2.88
Labor				
Family labor	502.11	168.18	84444.85	20.26
Hired labor	502.11	150.23	75436.99	18.09
Fertilizers	-	-	24247.03	5.82
Urea	20.08	162.56	3264.20	0.78
Triple Super Phosphate	25.08	341.29	8559.55	2.05
Diammonium Phosphate	34.81	88.60	3084.16	0.74
Muriate of Potash	19.92	111.68	2224.66	0.53

Cost items	Price	Total quantity	Total cost (Tk.)	%
Zinc Sulfate	200.11	16.24	3249.78	0.78
Gypsum	10.02	77.01	771.64	0.19
Boric Acid	202.16	15.30	3093.04	0.74
Cow dung	1.61	7862.47	12658.57	3.04
Organic fertilizer	417.55	90.20	37663.01	9.04
Pesticides	403.46	74.05	29880	7.16
Irrigation	313.90	34.85	10942.29	2.63
Cost for structure including Bamboo	-	-	46249.94	11.09
Total variable cost	-	-	337408.15	80.95
Interest on operating capital	-	-	3374.08	0.81
Land rent	-	-	76024.1	18.24
Total fixed cost	-	-	79398.18	19.05
Total cost	-	-	416806.33	100.0

Source: Farmer's household survey, 2022

### Profitability of bean production for contract and non-contract farmers

Significant t test indicated that there was significant difference between contract and non-contract farmer's cultivation area, total production, market price of bean, revenues from selling bean, total revenues, total variable costs, total cost, gross farm incomes, net farm incomes, BCR based on variable cost and BCR based on total cost. Contract farmers cultivated more land (0.162 ha) than the non-contract farmers (0.113 ha) and the difference was significant at 1% level. This indicated that contract farmer used more land than the non-contract farmer for bean production (Table 6). Contract farmers (28574.92 kg/ha) produce more bean than the non-contract farmers (27683.22 kg/ha) and the difference was statistically significant at 1% level. Contract farmers got higher prices (Tk. 41.32) for their bean than for the non-contract farmers (Tk. 31.22) and difference was statistically significant at 1% level. Revenues from selling bean was much higher for contract farmers (Tk. 1180715.69/ha) than the non-contract farmers (Tk. 864270.12/ha) and the difference was statistically significant at 1% level. Non-contract farmers (Tk. 10125.51) got more income from selling used bamboo than the contract farmers (Tk. 10098.7). The total revenue was much higher for the contract farmers (Tk. 1190814.39) than for non-contract farmers (874395.63) and the difference was statistically significant at 1% level. This result indicating that contract farmers got more revenue from selling bean than the non-contract farmers. Total variable cost of non-contract farmers (Tk. 337408.15) was higher than the contract farmers (Tk. 322645.45) and the difference was statistically significant at 5% level. Total cost of non-contract farmers (Tk. 416806.33) was higher than the contract farmers (Tk. 401137.17) and the difference was statistically significant at 5% level. This result indicating that non-contract farmers cost for producing bean was higher than the contract farmers. Gross farm income of contract farmers (Tk. 868168.94) was much higher than the contract farmers (Tk. 536987.48) and the difference was statistically significant at 1% level. Similarly, net farm income of contract farmers (Tk. 789677.22/ha) was much higher than the non-contract farmers (Tk. 457589.3/ha) and the difference was statistically significant at 1% level. This result indicated that contract farmers earn more profit than the non-contract farmers by producing bean in the study area. BCR considering variable cost of contract farmers (3.6) was higher than the non-contract farmers (2.59) and the difference was statistically significant at 1% level. BCR considering total cost of contract farmers (2.96) was higher than the non-contract farmers (2.09) and the difference was statistically significant at 1% level. BCR considering both variable and total cost indicated that bean production was profitable both for contract and non-contract farmers. However, contract farmers earn more profit from their investment for bean production than the non-contract farmers in the study area.

**Table 6. Profitability of bean production per hectare for contract and non-contract farmers**

Particulars	Contract farmers (n = 120)	Non-contract farmers (n = 180)	Mean difference (T-test)
Cultivation area	0.162	0.113	-4.19***
Total production	28574.92	27683.22	-3.15***
Market price	41.32	31.22	-21.86***
Revenues from selling bean	1180715.69	864270.12	-21.99***
Income from selling used bamboo	10098.7	10125.51	0.035
Total Revenues	1190814.39	874395.63	-21.88***
Total variable costs	322645.45	337408.15	2.52**
Total cost	401137.17	416806.33	2.02**
Gross farm incomes	868168.94	536987.48	-22.20***
Net farm incomes	789677.22	457589.3	-22.68***
BCR (Variable cost basis)	3.6	2.59	-17.23***
BCR (Total cost basis)	2.96	2.09	-18.11***

Source: Farmer's household survey, 2022

Note: \*\*\*, \*\* and \* indicates significance at the 1%, 5% and 10% levels respectively

#### Document and pricing method of contract farmers

Majority of the contract farmers (96.67%) had only verbal commitments about the contract and only 3.33% of the farmers had written document about the contract (Table 7). This result showed that contract farmers did not have strong document with different company about the contract. Farmers and company follows different pricing method for fix the bean price. Near about 80% of the farmers agreed to follows spot price and only 6.67% farmers agreed to follow forward price for their produced bean. Sometimes contract farmers (14.16%) agreed to follow fixed price determined by the company. Average duration of the contract between farmers and the company was 1.22 years, indicating that company do not contract with the contract farmers for long duration and they liked to contract for short duration.

**Table 7. Contract farmers document with the companies for the contract and pricing method**

Items	% of farmers
Document	
Only verbal	96.67 (116)
Written	3.33 (4)
Pricing method	
Spot price	79.17 (95)
Forward price	6.67 (8)
Fixed price determined by the company	14.16 (17)
Average duration of the contract	1.22

Source: Farmer's household survey, 2022

Note: Figure in the parentheses indicate number of farmers

#### Contract farmer's types of contract and support got from the companies

Contract farmers followed different types of contract viz. production, marketing and total contract (Table 8). About 17% of the contract farmers followed production contract and 82.5% of the contract farmers followed marketing contract. This result showed that most of the farmers liked marketing contract in the study area. Only 0.83% of farmers followed total contract. Contract farmers got

different types of support from the company. Only 1.67% of the contract farmers got inputs from the company and 51.67% of the contract farmers got cash support from the company. Some contract farmers (38.33%) got advice from the company and 17.5% of the contract farmers got training from the company. Companies marketed the bean with standard packaging and 4.17% of the companies gave packaging support to the contract farmers.

**Table 8. Different types of contract and support got from the companies**

Items	% of farmers
Types of contracts	
Production contract	16.67 (20)
Marketing contract	82.5 (99)
Total contract	0.83 (1)
Different support	
Inputs	1.67 (2)
Cash	51.67 (62)
Advice	38.33 (46)
Training	17.5 (21)
Packaging	4.17 (5)

Source: Farmer’s household survey, 2022

Note: Figure in the parentheses indicate number of farmers

**Satisfaction of contract farmers for their contract**

Most of the contract farmers (86.67%) were satisfied about their contract, indicating that contract farming improving production and marketing problems of the farmers (Table 9).

**Table 9. Satisfaction of contract farmers for their contract**

Items	% of farmers
Satisfied	86.67 (104)
Not satisfied	8.33 (10)
Neutral	5 (6)

Source: Farmer’s household survey, 2022

Note: Figure in the parentheses indicate number of farmers

Only 8.33% of the contract farmers were not satisfied with their contract, indicating that those farmers still faced problem with contract farming. Five percent of the contract farmers gave neutral opinion about the contract in the study area.

**Problems of bean producing contract farmers**

Contract farmers mentioned different types of problems and 64.17% of the contract farmers mentioned that some company did not strictly follow the contract (Table 10). Contract farmers desire training and 56.67 % of the contract farmers did not get training from the company. Contract farmers (76.67%) mentioned that company did not share risk with the farmers. Furthermore, contract farmers (64.17%) mentioned that some company did not give predetermined set price to them. Companies did not give inputs mentioned by 64.17 % of farmers. More than fifty percent contract farmers (53.33%) mentioned that contract farming was not benefited to them. Sometime companies did not take bean from contract farmers (67.5%). Contract farmers also mentioned that weak law enforcement to follow contract by the company. It was difficult to follow contract mentioned by 64.16% of the contract farmers.

**Table 10. Problems of bean producing contract farmers**

<b>Problems</b>	<b>% of farmers</b>
Companies do not follow contract	64.17 (77)
Companies do not give support	68.34 (82)
Companies do not advance cash	85.83 (103)
Companies do not give training	56.67 (68)
Companies do not share risk	76.67 (92)
Companies do not give set price	64.17 (77)
Companies do not give inputs	64.17 (77)
No benefit from contract farming	53.33 (64)
Sometime company do not take bean	67.5 (81)
Weak law enforcement to follow contract	72.5 (87)
Difficult to follow contract	64.16 (77)

Source: Farmer's household survey, 2022

Note: Figure in the parentheses indicate number of farmers

### **Contract farmer's suggestion to solve their existing problems**

Contract farmers gave some suggestions to solve their problems (Table 11). Almost 87% of the contract farmers suggested that the company need to follow the contract. Most of the contract farmers (89.16%) mentioned that they need cash in advance from the company they contracted. Contract farmers (80.83%) need training from the company. Seventy percent of the farmers mentioned that company need to give higher bean price to the farmers in compare to market price, indicating that the higher bean price will give incentive to them. Many contract farmers (71.67%) mentioned that they need inputs supply from the company, which solve farmers input crisis problem. Most of the farmers (92.5%) desired government monitoring for the contract, which compelled to the company to follow the contract properly.

**Table 11. Contract farmer's suggestion to improve existing problems**

<b>Suggestions</b>	<b>% of farmers</b>
Company need to follow the contract	86.67 (104)
Company need to give cash in advance	89.16 (107)
Farmers need training from company	80.83 (97)
Company need to give higher bean price in compare to market price	70.0 (84)
Company need to give inputs to the contract farmers	71.67 (86)
Farmers desire government monitoring for the contract	92.5 (111)

Source: Farmer's household survey, 2022

Note: Figure in the parentheses indicate number of farmers

### **Problems of bean producing non-contract farmers**

Low price of bean (83.89%) was the crucial problem of farmers for non-contract farmers in the study area (Table 12). Most of the non-contract farmers (97.78%) mentioned that price fluctuation of bean was their major problem. High price and unavailability of inputs was the problem of 86.11 % of the farmer. Lack of storage facilities was another problem of the non-contract farmers (77.22%). Almost similar percentage of farmers mentioned that insect and disease damage were their problem. About 63% of the non-contract farmers mentioned that lack of irrigation facility was their problem. High transportation cost was the problem of 72.78% of the farmers. Non-contract farmers (84.45%) also mentioned that lack of marketing facility was their problem, indicating that farmers faced problem to sell their bean in the study area. Lack of capital with low interest rate was the problem of 96.67% of the farmers, indicating that non-contract farmers needed capital for the production of bean in the study area.

**Table 12. Problems of bean producing non-contract farmers**

Problems	% of farmers
Low price of bean	83.89 (151)
Price fluctuation of bean	97.78 (176)
High price and unavailability of inputs	86.11 (155)
Lack of storage facilities	77.22 (139)
Insect damage	87.22 (157)
Disease damage	88.33 (159)
Lack of irrigation facility	62.78 (113)
High transportation cost	72.78 (131)
Lack of marketing facility	84.45 (152)
Lack of capital with low interest rate	96.67 (174)

Source: Farmer's household survey, 2022

Note: Figure in the parentheses indicate number of farmers

### Non-contract farmer's suggestion to solve their existing problems

Non-contract farmers (72.77%) had desire to engage with contract farming (Table 13). Most of the farmers (95.56%) needed capital with low interest rate from different sources, which will solve their capital problem. High price and unavailability of inputs was the problem of non-contract farmers and 85.56 % of the non-contract farmers needed reasonable price and availability of inputs. Farmers needed available market to sell their bean and 74.44 % of the non-contract farmers mentioned that they needed market in the district headquarter. Transportation facility was the problem of contract farmers and 74.44 % of the non-contract farmers needed railway service to transport bean in the study area. Irrigation was important for bean production and 86.11 % of the non-contract farmers need subsidy for electricity and diesel price. Most of the non-contract farmers (90.0%) were needed more facility in the market.

**Table 13. Non-contract farmer's suggestion to improve existing problems**

Suggestions	% of farmers
Farmers want to engage with contract farming	72.77 (131)
Need capital with low interest rate	95.56 (172)
Need reasonable price and availability of inputs	85.56 (154)
Need farmers market in the district headquarter	74.44 (134)
Need railway service to transport bean	74.44 (134)
Need subsidy for electricity and diesel price for irrigation	86.11 (155)
Need more facilities in the market	90.0 (162)

Source: Farmer's household survey, 2022

Note: Figure in the parentheses indicate number of farmers

## CONCLUSION AND RECOMMENDATION

Many farmers are producing bean in Narsingdi district and contract farming is becoming popular. Both contract and non-contract farmers are interviewed to achieve the objectives. The major objective of the study is to investigate the profitability of bean production. This research also identifies farmer's major problems and their suggestions are also documented. The study found that, country bean production is profitable both for contract and non-contract farmers. But country bean production is more profitable for the contract farmers. The research found that most of the farmers have verbal commitment with the companies for the contract. Most of the contract farmers had marketing contract and 51.67% of contract farmers get cash support from the companies. The study found that most of the contract

farmers are satisfied about their contract. Contract farmers faced different problems in the research area. Most crucial problems of contract farmers are contract companies do not advance cash to the farmers and company do not share risk. Contract farmers desire government monitoring for the contract. Contract farmers suggested that contract companies need to follow the contract. Most of the contract farmers suggested that company need to give cash in advance and training to the farmers. On the other hand, price fluctuation of bean and lack of capital with low interest rate are the crucial problem of non-contract farmers. Non-contract farmers suggest different solution to solve their problems. They need capital with low interest rate and they need more facilities in the market.

Based on the existing findings of the study, the following recommendation may be made to increase production and marketing of bean in the study area.

- In case of contract farming, some companies do not follow the contract. It is necessary to have written document of the contract and farmers and companies need to strictly follow the contract.
- Contract farmer's desire cash in advance from the company and company need to distribute cash in advance to the farmers during the bean production.
- Both contract and non-contract farmers need available inputs with reasonable price. They also need training about production and marketing of bean.
- Contract and non-contract farmers need capital with low interest rate and proper initiative must be taken for the institutional credit to the farmers.
- Finally, local agricultural office must monitor farmer's different agricultural contract which will solve different problems of farmers.

## REFERENCES

- Alam, G.M.M., Alam, K. and Mushtaq, S. 2018. Drivers of Food Security of Vulnerable Rural Households in Bangladesh. *South Asia Economic J.*, 19: 43–63.  
<https://doi.org/10.1177/1391561418761075>
- Allen, D.W. and Lueck, D. 1995. Risk Preferences and the Economics of Contracts, *American Economic Review*, 85: 447–451.
- BARI (Bangladesh Agricultural Research Institute). 2017-18. BARI Annual Report 2017–18, Bangladesh Agricultural Research Institute, Gazipur, Bangladesh.
- Bangladesh Bureau of Statistics (BBS), 2021. Statistical Pocketbook 2021a, Statistics and Informatics Division, Ministry of Planning, Government of the People's Republic of Bangladesh.
- Bangladesh Bureau of Statistics (BBS), 2021. Yearbook of Agricultural Statistics. 2021a, Statistics and Informatics Division, Ministry of Planning, Government of the People's Republic of Bangladesh.
- Bidzakin, J.K., Fialor, S.C., Awunyo-Vitor, D. and Yahaya, I. 2020. Contract farming and rice production efficiency in Ghana, *J. Agribusiness in Developing and Emerging Economies*, 10(3): 269-284.
- BER (Bangladesh Economic Review) 2022. Bangladesh Economic Review, Finance Division, Ministry of Finance, Government of the People's Republic of Bangladesh, Dhaka, Bangladesh.
- Chowdhuri, N.Y., Haque, S., Shammi, S.A., Jannat, A., Sannyashi, P.R., 2014. Profitability analysis of winter vegetables production in a selected area of Narshingdi district in Bangladesh, *Progressive Agriculture*, 25: 45-53.
- Eaton, C. and Shepherd, A.W. 2001. Contract farming: partnerships for growth. FAO Agricultural Services Bulletin 145. Food and Agriculture Organization of the United Nations, Rome, Italy.
- Fafchamps, M. 1996. The Enforcement of Commercial Contracts in Ghana. *World Dev.*, 24: 427–448.
- Fafchamps, M. and Minten, B. 2001. Property Rights in a Flea Market Economy. *Econ. Dev. Cult. Change*, 49: 229–267.

- FAO, 2017. Statistical Year Book 2017. Food and Agricultural Organization of the United Nations, Rome, Italy.
- Freguin-Gresh, S., D'haese, M. and Anseeuw, W. 2012. Demythifying contract farming: Evidence from rural South Africa, Agrekon: Agricultural Economics Research, Policy and Practice in Southern Africa, 51(3): 24-51.
- Hasan, M.R., Mutatisse, A.A., Nakamoto, E. and Bai, H. 2014. Profitability of cauliflower and bean production in Bangladesh- A case study in three districts, *Bangladesh J. Extension Edu.*, 26 (1 & 1): 63-75.
- Hasan, M.R. and Bai, H. 2016. Profitability of tomato production in three districts of Bangladesh, *Intl. J. BioRes.*, 21 (6): 1-8.
- Hoang, V. 2021. Impact of contract farming on farmers' income in the food value chain: A theoretical analysis and empirical study in Vietnam. *Agriculture*, 11: 797. doi: <https://doi.org/10.3390/agriculture11080797>
- Key, N. and Runsten, D. 1999. Contract farming, smallholders and rural development in Latin America: the organization of agro-processing firms and the scale of out grower production, *World Development* 27(2): 381-401.
- McMillan, J. and Woodruff, C. 1999. Inter-firm relationships and informal credit in Vietnam. *Q. J. Econ.*, 114: 1285-1320.
- Minot, N. and Sawyer, B. 2016. Contract Farming in Developing Countries: Theory, practice, and Policy Implications. In *Innovation for Inclusive Value Chain Development: Successes and Challenges*; International Food Policy Research Institute: Washington, DC, USA, 127-155.
- Mishra, A.K., Kumar, A., Joshi, P.K. and D'Souza, A. 2018. Production risks, risk preference and contract farming: Impact on food security in India. *Applied economic perspectives and policy* 40: 353-78.
- Mulatu, G., Haji, J., Legesse, B. and Ketema, M. 2017. Impact of Participation in Vegetables' Contract Farming on Household's Income in the Central Rift Valley of Ethiopia. *American J. Rural Develop.*, 5(4) : 90-96. doi: 10.12691/ajrd-5-4-1
- Nguyen, A.T., Dzator, J. and Nadolny, A. 2015. Does contract farming improve productivity and income of farmers?: A review of theory and evidence. *J. Developing Areas*, 49: 531-538.
- Porter, G. and Phillips-Howard, K. 1995. Farmers, labourers and the company: Exploring relationships on a Transkei contract farming scheme. *J. Develop. Studies*, 32(1): 55-73.
- Soullier, G. and Moustier, P. 2018. Impacts of contract farming in domestic grain chains on farmer income and food insecurity. Contrasted evidence from Senegal. *Food Policy*, 79: 179-198.
- Singh, S. 2002. Contracting out solutions: political economy of contract farming in the Indian Punjab, *World Development*, 30(9): 1621-1638.

## EFFECT OF COVID-19 PANDEMIC ON MARKET PRICE OF POULTRY AND POULTRY PRODUCTS IN BANGLADESH

M. A. H. Beg<sup>1</sup>, M. A. Raihan<sup>2</sup>, T. H. Beg<sup>3</sup> and M. Begum<sup>4</sup>

### ABSTRACT

The aim of the study was to investigate how the COVID-19 pandemic has impacted the wholesale price of chicken and chicken products. Data of one year were collected from online database of poultry news portal from which 6 months of pre-covid and 6 month of covid period. A structured questionnaire was prepared to put market price of chicken and chicken products from eight divisions of Dhaka, Khulna, Barishal, Mymensingh, Sylhet, Rangpur, Rajshahi and Chattogram of Bangladesh. Secondary data were collected for 12 months from each divisions for all parameters. The results showed that the average wholesale price of broilers, culled white chicken and Sonali chicken, shell eggs (brown and white) and day-old-chicks (brown chicks, white layer chicks and broiler chicks) were fluctuated due to COVID-19 pandemic, but no significant ( $p>0.05$ ) difference was found in prices except live culled red chicken. The average wholesale price of culled red chicken in Chattogram was  $171.12\pm 19.67$ Tk/kg on July to December 2019 and  $195.34\pm 35.70$ Tk/kg on July to December 2020. In Khulna and Rajshahi the wholesale price of brown shell eggs has descending order whereas in Dhaka, Barishal and Chattogram the price has a ascending order, but it remain unchanged in Mymensingh, Sylhet and Rangpur during COVID pandemic. The wholesale price of white shell eggs in Dhaka, Khulna and Rajshahi showed decreasing trend during pandemic but increasing trend in Chattogram. The wholesale price of white layer chicks was lowered by 21.43Tk, 23.79Tk and 31.03Tk/chick in COVID period than pre-COVID period in Dhaka, Barishal and Rangpur, whereas minor price fluctuation was observed in brown layer and broiler chicks. The extreme wholesale price of a Sonali chick was  $15.67\pm 3.50$ /chick in Rangpur only during pandemic. The study concluded that the COVID-19 pandemic had a negative impact on the poultry industry.

**Keywords:** poultry industry, COVID-19, wholesale price, poultry and poultry products<sup>7</sup>

### INTRODUCTION

Poultry business is a promising sector for reducing poverty in Bangladesh. The majority of the poultry produced in Bangladesh is chicken. There are about 311.8 million chicken and 63.84 million ducks in Bangladesh (DLS, 2022). Now the available meat consumption is 147.84g/day/head (DLS, 22) where as the people of developed country consume above 200g (UFAPRI, 2019). Same time our egg consumption is 136.01Nos./year/head (DLS, 2022) and the people of developed country intake above 250 eggs/year (The Business Standard, 2020). About 3.5 to 4.0 crore eggs are required daily to meet national demand (BEPA, 2022). The primary animal industry in Bangladesh is poultry, which plays a significant role in the nation's overall food security and economic growth (Hamid *et al.*, 2017). This industry employs more than 8.5 million people, making it the second-largest employer behind the apparel sector. *Poultry meat alone* contributes 37% of the total meat (Hamid *et al.*, 2017). In Bangladesh Contribution of livestock in national GDP is 1.9% and share of livestock in Agricultural GDP is 16.52% (DLS, 2022). One of the important livestock sub-sectors dedicated to giving the country cheap access to animal protein in the form of eggs and protein is the poultry business (Das *et al.*, 2008). According to the BPICC, Bangladesh has 18 grandparent and 216 parent stock corporations of which one hundred certified (BPICC, 2022). Bangladesh reported its first COVID-19 case on March, 2020 (WHO, 2020). The government implemented a statewide lockdown (known as the "General Holiday" from 26 March and 30 May as part of its efforts to stop the virus spread. Meanwhile the market of poultry products has fallen dramatically as a result of the corona virus. In addition, the output

---

<sup>1,2&4</sup> Dept. of Poultry Science, <sup>3</sup>Dept. of Management & Finance, Sher-e-Bangla Agricultural University, Dhaka-1207

of eggs has significantly decreased (Sattar *et al.*, 2021). In the meantime, more farms sold their older birds without bringing in a fresh batch of chicks. As hotels, restaurants, bakeries, and fast food outlets have been shut down since March 26 to stop the spread of the corona virus, the demand for poultry meat and eggs has continued to decline and about 50-60% of chicken farms have been temporarily closed (BPICC, 2020). More than half of the nation's poultry farms and hatcheries were forced to close as a result of the avian influenza epidemic in 2007. At the beginning of coronavirus, spread of misinformation and rumors driven by the fear of linking chicken meat and eggs in the transmission of this disease (COVID-19) among human population is witnessed in India ( Kolluri *et al.*, 2020). It seriously threatened the livelihoods of millions Bangladeshi backyard chicken raisers and small traders (Mahmud, 2020). If the supply chain for the poultry business is disrupted for an hour, a week, a day, or a month, it can have major economic repercussions, as it was happened pandemic time. A few studies on COVID-19 and poultry have been conducted. Taking into account the aforementioned concerns, the research project set out to determine the market fluctuation scenario for poultry and poultry products.

### **Objectives**

The present study has been taken to investigate the market fluctuation scenario of poultry and poultry products of Bangladesh before and during pandemic situations. However, the specific objectives are as follows:

- To find out the fluctuation of wholesale price of broilers, culled birds, shell eggs and day-old-chicks and
- To compare pandemic market price of chicken and chicken products with Pre-COVID and COVID market.

## **MATERIALS AND METHODS**

### **Study area**

The study has been conducted in Bangladesh. The country has eight administrative divisions. The study covered all division of Bangladesh.

### **Type and sources of data**

In the present study, secondary data were used only from wholesale market. The data were collected from relevant online database of poultry news portal Agricare24.com (2017) and Agrinews24.com (2017).Data of one year were collected of which six months from pre-COVID period (July-December/2019) and six months from COVID period (July-December/2020) to conduct the experiment.

### **Preparation of questionnaire**

A structured questionnaire was prepared to collect market price of live chicken and chicken products which includes the following products.

- A. Wholesale price of Live Birds: Broiler chicken, Culled red chicken, Culled white chicken and Sonali chicken.
- B. Wholesale price of eggs: Brown shelled eggs and White shelled eggs.
- C. Wholesale price of day-old-chicks: Brown layer chicks, White layer chicks, Broiler chicks and Sonali chicks.

### **Selection of months to collect the data**

The first known outbreak of COVID-19 started in Wuhan, Hubei, China, in November 2019. Many early cases were linked to people who had visited the Huanan Seafood Wholesale Market there, but it is possible that human-to-human transmission began earlier. However, in Bangladesh it was confirmed the first coronavirus case on 8 March 2020. In response to the COVID-19 pandemic, the Government of Bangladesh (GoB) declared special “general leave” from 26 March in the name of “lockdown” and extended it up to 30 May 2020 in seven different time slots. So, although it is COVID-19, but 2019 was totally free from COVID pandemic in Bangladesh. Therefore, last six months of 2019 i.e. July to December month was selected to collect pre-COVID data for the study. In Bangladesh lockdown

affects the poultry market seriously since July 2020. This is why July- December/2020 month was selected to collect data during pandemic.

#### **Data collection**

Data were collected from eight divisions (Dhaka, Khulna, Barishal, Mymensingh, Sylhet, Rangpur, Rajshahi and Chattogram) of Bangladesh. To collect the data 4 days of a month were considered randomly taking a day from each week. A Total of (4 days x 6months) 24 days from pre-COVID and 24 days from COVID period i.e. 48 days data were collected from each division for each parameters for the study.

#### **Collection of wholesale price data of live birds**

All categories of data were not available in all divisions for pre-COVID and COVID period. Only those divisions' data were considered which were available in the both period. In case of live broiler, Khulna and Rangpur division's data were not available during pre-COVID and COVID period. For culled red birds Khulna, Mymensingh, Sylhet, Rangpur's data were not available. For culled white only Dhaka (Gazipur), Khulna and Rajshahi's data were available in both period. Khulna, Sylhet and Rangpur divisions' data were not available for Sonali chicken.

#### **Collection of wholesale price data of eggs**

In case of brown shelled eggs all data were available in all divisions during COVID and pre-COVID period. But in case of white shelled eggs the divisions of Barishal, Mymensingh, Sylhet and Rangpur's data were not available in both period.

#### **Collection of wholesale price data of day-old-chicks**

Wholesale price of chicks were available in four divisions, Dhaka, Barishal, Rangpur and Chattogram for brown, white and day-old-broiler chicks. Day-old-Sonali chicks' wholesale price was not available in all divisions of Bangladesh during pre-COVID period except Rangpur division.

#### **Statistical analysis**

The collected data and information were scrutinized, classified, edited and coded. Information of questionnaire was transferred into a master sheet for entering the data in the computer. Microsoft Excel computer software package programs were used to find out average data of live birds, eggs and day-old- chicks. Finally the SPSS computer package program was used for data analysis. To compare the data of two period of same divisions paired t-test was used.

## **RESULTS AND DISCUSSION**

### **Wholesale Price of Live Birds**

Generally the price of live birds varied in different market place of Bangladesh. This is due to production cost, seasons, festival and demand of the customers. But during current pandemic it was seriously affected poultry industry and as well as price of live birds.

#### **Wholesale price (Taka/Kg) of broiler chicken in different divisions in Bangladesh**

Table 1 gives the mean wholesale price of broiler chicken that was delivered by several agricultural marketplaces of different divisions both before and during COVID-19. Data from 12 months were used in the analysis, representing 6 divisions. In most cases the data were gathered from 6 months prior to COVID and 6 months during COVID. The mean wholesale price has increased trend during the COVID-19 period as compared to the same period last year. No statistical ( $p>0.05$ ) difference was found in the wholesale price of broiler chicken before COVID in 2019 and during COVID in 2020. But, during COVID 2.0 to 3.35Tk were increased in Barishal, Dhaka and Chattogram; the price was unchanged in Sylhet; 1.0Tk difference was observed in Mymensingh and Rajshahi division. The findings of the current analysis indicated that 2020 was seeing a modest increased in wholesale prices compared to 2019. The Business Standard (2020) reported that monthly broiler production fell to 25000 from 27000 tones and around 50% to 60% poultry farms shuttered across the country due to COVID-19 pandemic. Furthermore, Berkhout (2020) reported that these production levels decreased by at least 50% after March 2020.

**Table 1. Average wholesale price (Taka/Kg) of broiler chicken in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	93.80 $\pm$ 7.64	96.88 $\pm$ 6.12	0.516 <sup>NS</sup>
Barishal	92.92 $\pm$ 3.93	95.00 $\pm$ 7.52	0.565 <sup>NS</sup>
Mymensingh	96.63 $\pm$ 8.60	97.18 $\pm$ 5.31	0.907 <sup>NS</sup>
Sylhet	97.91 $\pm$ 7.21	97.33 $\pm$ 6.48	0.888 <sup>NS</sup>
Rajshahi	98.55 $\pm$ 9.04	97.98 $\pm$ 4.60	0.899 <sup>NS</sup>
Chattogram	96.87 $\pm$ 6.22	100.22 $\pm$ 6.13	0.433 <sup>NS</sup>

Here, SD = Standard Deviation; NS = Non-significance; Tested at 5% level of significance ( $p < 0.05$ )

### Average whole price (Taka/Kg) of culled red chicken in different divisions

Table 2 gives the mean wholesale price of culled red chicken of Dhaka, Barishal, Rajshahi and Chattogram that was delivered by several agricultural marketplaces of different divisions both before and during COVID-19. The results revealed that there was no significant ( $p > 0.05$ ) difference between the wholesale price of culled red chicken in 2019 and 2020 at Dhaka, Barishal and Rajshahi divisions, but at Rajshahi 22.0Tk was increased/kg, whereas 4.0Tk was at Barishal. At Chattogram division 24.0Tk was increased significantly ( $p < 0.05$ ) per kg of culled red chicken during COVID than pre-COVID period.

**Table 2. Monthly average wholesale price (Taka/Kg) of culled red chicken in different divisions**

Division	Pre-COVID Period ( July-Dec/19)	During COVID Period July-Dec/20)	Level of significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	157.86 $\pm$ 10.21	172.83 $\pm$ 31.92	0.160 <sup>NS</sup>
Barishal	178.12 $\pm$ 10.89	182.08 $\pm$ 29.22	0.659 <sup>NS</sup>
Rajshahi	160.05 $\pm$ 19.77	182.08 $\pm$ 31.09	0.083 <sup>NS</sup>
Chattogram	171.12 $\pm$ 19.67	195.34 $\pm$ 35.70	0.044 <sup>*</sup>

Here, SD = Standard Deviation; NS= Non-significance \*means significant at 5% level of significance ( $p < 0.05$ ).

Disruption of poultry supply chain and state lockdown severely affects poultry industries and this is the main cause of increasing price of culled red chicken during COVID in 2020 compared to pre-COVID. Another reason of increasing price of culled red chicken is only the rich persons (Sattar *et al.* 2021) purchase it. The distribution of chicken and poultry products is hampered by market risks such value chain fragmentation and high intermediation costs (MoF, 2020).

### Average wholesale price (Taka/Kg) of culled white chicken in different divisions

From Table 3, it is noted that the mean wholesale price of culled white chicken increased 15.0Tk at Dhaka division and decreased 10.0Tk at Khulna, but it remain unchanged at Rajshahi division during COVID period. The result revealed that the wholesale price of culled white chicken was fluctuated between pre-COVID and during COVID period, but, there was no significant ( $p > 0.05$ ) difference between the wholesale price of culled white chicken in 2019 and 2020.

This finding was an agreement with Sattar *et al.* (2021); a notable COVID-19 effect on the poultry industry was a change in live chicken and egg prices before, during, and after the lockdown from January to June 2020. As a major factor in the decline in consumer demand, the rumors were also cited in other news papers (Sharma *et al.*, 2020).

**Table 3. Monthly average whole price (Taka/Kg) of culled white chicken in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	115.34 $\pm$ 12.12	130.56 $\pm$ 26.25	0.09 <sup>NS</sup>
Khulna	146.64 $\pm$ 13.28	136.69 $\pm$ 25.72	0.310 <sup>NS</sup>
Rajshahi	135.77 $\pm$ 12.92	135.91 $\pm$ 27.09	0.988 <sup>NS</sup>

Here, SD = Standard Deviation; NS= Non-significance; Tested at 5% level of significance ( $p < 0.05$ )

#### Average wholesale price (Taka/Kg) of Sonali chicken in different divisions

The wholesale price of Sonali chicken is presented in Table-4, which was not significantly ( $p > 0.05$ ) different. Compared to broilers, Sonali chickens make up a lower percentage of the chicken marketed in Bangladesh, and its producer fetch a higher price from (mainly) richer buyers (Sattar *et al.*, 2021). It is observed that in COVID period its wholesale price at Barishal, Rajshahi and Chattogram division declined to 7.0, 32.0 and 7.0Tk respectively compared with pre-COVID price; although its price remains unchanged at Dhaka and Mymensingh division.

**Table 4. Monthly average wholesale price (Taka/Kg) of Sonali chicken in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period( July-Dec/20)	Level of significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	171.32 $\pm$ 15.19	171.04 $\pm$ 30.45	0.988 <sup>NS</sup>
Barishal	173.12 $\pm$ 7.57	166.66 $\pm$ 24.48	0.621 <sup>NS</sup>
Mymensingh	173.96 $\pm$ 14.15	173.29 $\pm$ 30.17	0.970 <sup>NS</sup>
Rajshahi	175.79 $\pm$ 15.61	143.33 $\pm$ 75.53	0.412 <sup>NS</sup>
Chattogram	184.79 $\pm$ 14.99	176.09 $\pm$ 29.11	0.608 <sup>NS</sup>

Here, SD = Standard Deviation; NS= Non-significance; Tested at 5% level of significance ( $p < 0.05$ )

The result is an agreement with Sattar *et al.* (2021) and showed that Sonali's price swings were noticeably less pronounced than those of broilers, and they remained at or below pre-lockdown levels even throughout lockdowns.

#### Wholesale Price of Shell Eggs

The devastating impact of COVID-19 on poultry industry, specially egg supply chain seriously disrupted and egg price fluctuated after and before COVID pandemic. The fluctuation of the wholesale price of shell eggs has been studied here.

#### Average wholesale price (Taka/Piece) of brown shell eggs in different divisions

The mean wholesale price of brown shell eggs at Dhaka, Khulna, Barishal, Mymensingh, Sylhet, Rangpur, Rajshahi and Chattogram is presented in Table 5. Although the wholesale price of brown shell eggs little bit fluctuated between after and during COVID-19 but there were no significant ( $p > 0.05$ ) difference of the wholesale price of brown shell eggs between pre-COVID and during COVID pandemic. It is revealed from the findings that in Khulna and Rajshahi the wholesale price was decreased. Mahmud (2020) noted similar opinion that people have started avoiding consumption of chicken and eggs. But in Dhaka, Barishal and Chattogram the price was upward, but it was remain unchanged in Mymensingh, Sylhet and Rangpur divisions during pre-COVID and COVID pandemic period. According to BPICC (2020) due to the closure of 32–35% egg-producing farms, egg output had also dramatically decreased during the pandemic.

**Table 5. Monthly average wholesale price (Taka/Piece) of brown shell eggs in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	7.28 $\pm$ 0.54	7.39 $\pm$ 0.76	0.496 <sup>NS</sup>
Khulna	7.61 $\pm$ 0.71	7.45 $\pm$ 0.96	0.438 <sup>NS</sup>
Barishal	7.01 $\pm$ 0.78	7.31 $\pm$ 0.85	0.10 <sup>NS</sup>
Mymensingh	7.37 $\pm$ 0.53	7.37 $\pm$ 0.79	0.988 <sup>NS</sup>
Sylhet	7.50 $\pm$ 0.57	7.53 $\pm$ 0.81	0.896 <sup>NS</sup>
Rangpur	7.17 $\pm$ 0.49	7.17 $\pm$ 0.66	0.964 <sup>NS</sup>
Rajshahi	7.11 $\pm$ 0.51	7.03 $\pm$ 0.60	0.566 <sup>NS</sup>
Chattogram	7.46 $\pm$ 0.61	7.55 $\pm$ 0.81	0.476 <sup>NS</sup>

Here, SD = Standard Deviation; NS= Non-significance; Tested at 5% level of significance ( $p < 0.05$ )

#### Average wholesale price (Taka/Piece) of white shell eggs in different divisions

The mean wholesale price of white shell eggs at Dhaka, Khulna, Rajshahi and Chattogram is displayed in Table 6. The results revealed no significant difference ( $p > 0.05$ ) of wholesale price of white shell eggs between pre-COVID and COVID period. The wholesale price in the division of Dhaka, Khulna and Rajshahi were decreasing trend during pandemic, but it was observed to increase only at Chattogram division.

**Table 6. Monthly average wholesale price (Taka/Piece) of white shell eggs in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of Significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	7.05 $\pm$ 0.56	7.01 $\pm$ 0.84	0.885 <sup>NS</sup>
Khulna	7.42 $\pm$ 0.67	7.23 $\pm$ 0.74	0.369 <sup>NS</sup>
Rajshahi	6.88 $\pm$ 0.64	6.48 $\pm$ 0.63	0.067 <sup>NS</sup>
Chattogram	7.12 $\pm$ 0.60	7.43 $\pm$ 0.92	0.119 <sup>NS</sup>

Here, SD= Standard Deviation; NS= Non-significance; Tested at 5% level of significance ( $p < 0.05$ )

The findings of this study in an agreement with the researcher (Kolluri *et al.* 2020) reported that the lowest egg prices were observed during the COVID-19 pandemic at USD 2.04 compared to a USD 5.0 manufacturing cost. Furthermore, Egg prices are an indirect indicator of the supply-demand situation in the market, and examining their trajectory in relation to time series offers a chance to adapt to shifting market conditions (Yuhuan and Fu, 2018). Bangladesh Poultry Industries Central Council President told (Dhaka Tribune, 2020) that the consumers cannot go to the kitchen markets due to the government-imposed lockdown, significantly bringing down the demand for meat, egg, and other processed food.

#### Wholesale price (Taka/chick) of day-old-chicks

People avoided consumption of chicken and egg as the COVID-19 outbreak had started. Besides, consumers were unable to go to the markets due to the lockdown which significantly reduced demand for meat, egg, and other processed food. As a result, a large number of poultry farmers shuttered down their poultry farm which effected on the price of day-old-chicks.

### Average wholesale price (Taka/chick) of brown layer chicks in different divisions

From the Table 7, it is illustrated that the non-significant ( $p>0.05$ ) mean of wholesale price of brown layer chicks at Barishal, Rangpur and Chattogram were decreased to  $36.20\pm 7.54$ ,  $36.91\pm 5.90$  and  $35.31\pm 6.60$  Tk during pandemic from pre-pandemic price of  $37.00\pm 7.06$ ,  $38.70\pm 6.90$  and  $41.58\pm 12.04$  respectively. The minor price hike (1.59Tk) was found at Dhaka division during COVID period. According to FAO (2020) reports, the prices of day-old layer chicks dropped by 75% and 40–50% of newly born DOCs get unsold due to COVID-19 pandemic.

**Table 7. Monthly average wholesale price (Taka/chick) of brown layer Chicks in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of Significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	38.12 $\pm$ 9.70	39.71 $\pm$ 6.60	0.476 <sup>NS</sup>
Barishal	37.00 $\pm$ 7.06	36.20 $\pm$ 7.54	0.748 <sup>NS</sup>
Rangpur	38.70 $\pm$ 6.90	36.91 $\pm$ 5.90	0.559 <sup>NS</sup>
Chattogram	41.58 $\pm$ 12.04	35.31 $\pm$ 6.60	0.223 <sup>NS</sup>

Here, SD= Standard Deviation; NS= Non-significance; Tested at 5% level of significance ( $p<0.05$ )

The number of day-old chicks produced has decreased by the breeders. According to the report (BPICC, 2020) over 50% of hatcheries have reportedly ceased production.

### Average wholesale price (Taka/chick) of white layer chicks in different divisions

The mean wholesale price of white layer chick in Dhaka, Barishal and Rangpur division were about 66.00Tk in pre-COVID time but it fall down to 35.0 to 44.0 Tk during pandemic period. The values showed no significant ( $p>0.05$ ) difference between in pre-COVID and COVID period (Table 8). In COVID-19, it was a great impact on poultry industry, basically on the price of white layer chicks. Due to COVID-19 pandemic the wholesale price of white layer chicks was decreased dramatically in different division of Bangladesh.

**Table 8. Monthly average wholesale price (Taka/chick) of white layer chicks in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of Significance
	Mean $\pm$ SD	Mean $\pm$ SD	
Dhaka	66.41 $\pm$ 12.98	44.98 $\pm$ 7.27	0.08 <sup>NS</sup>
Barishal	66.14 $\pm$ 14.20	42.35 $\pm$ 5.41	0.08 <sup>NS</sup>
Rangpur	66.55 $\pm$ 19.23	35.52 $\pm$ 6.68	0.05 <sup>NS</sup>

Here, SD= Standard Deviation; NS= Non-significance; Tested at 5% level of significance ( $p<0.05$ )

The result is an agreement with the previous researcher Sattar *et al.* (2021) showed that March and April, 2019 was a beneficial time of year for chicks traders and producers, because chicks prices are often slightly higher during these months. The situation, however, dramatically changed on March 26 when the pandemic hit the nation and the authorities declared a state of lockdown.

### Average wholesale price (Taka/chick) of broiler chicks in different divisions

No significant ( $p>0.05$ ) difference was observed in wholesale price of broiler chicks between pre-COVID and COVID period among different divisions (Table 9). The wholesale price of Dhaka,

Barishal, Rangpur and Chattogram was lower in 2020 than 2019 by difference of 2.62Tk, 4.89Tk, 7.92Tk and 5.38Tk, respectively. Many poultry farmers shuttered down their broiler farms due to decline of consumers demand in COVID period were the main cause of decreasing the price of broiler chicks. The study is an agreement with the report of IDLC (2020) that for many farmers, the business was unprofitable due to the difference between production costs and wholesale prices.

**Table 9. Monthly average wholesale price (Taka/chick) of broiler chicks in different divisions**

Division	Pre-COVID Period (July-Dec/19)	During COVID Period (July-Dec/20)	Level of Significance
	Mean ±SD	Mean ±SD	
Dhaka	25.96±4.80	23.34±4.02	0.269 <sup>NS</sup>
Barishal	26.27±5.12	21.38±4.47	0.139 <sup>NS</sup>
Rangpur	31.25±7.73	23.33±3.77	0.079 <sup>NS</sup>
Chattogram	27.90±6.17	22.52±3.08	0.135 <sup>NS</sup>

Here, SD= Standard Deviation; NS= Non-significance; Tested at 5% level of significance (p<0.05)

Ali (2020) also reported that the production of chicken meat dropped quickly from 90,000 to 25,000 tons/month with small-scale farms being particularly badly impacted while many large-scale farms cut their flock size. So at that time demand of chicks naturally decreased as well as chick's price was decreased.

#### **Average wholesale price (Taka/chick) of Sonali chicks in different divisions**

The data of the wholesale price of Sonali chicks was not available in poultry web portal on 2019 at different divisions of Bangladesh, but it was available only at Rangpur division in 2020. So the value was not statistically comparable. However, in Rangpur the average mean wholesale price of Sonali chicks was only 15.67±3.50Tk/chick on July to December 2020. Islam *et al.* (2021) also found day-old Sonali chick at low cost of 17.73 Tk/chick in winter season at Rajshahi division.

## **CONCLUSION**

Poultry sector play an important role in the economy of Bangladesh. Many people maintain their livelihood through involve this sector. This study attempted to assess the effect of COVID-19 pandemic on market price of poultry and poultry products. It was observed that the price of poultry and poultry products varies division to division a bit. There was no significant difference between before and at the time of COVID pandemic regarding poultry and poultry products price. However, people involved with this sector greatly hampered due to COVID-19 pandemic. The policymakers should consider the facts and take measures to improve the sector.

**Acknowledgement:** The authors are grateful to the Sher-e-Bangla Agricultural University Research System (SAURES) authority for providing financial support to complete the research work

## **REFERENCES**

- Agricare24.com*. 2017. A specialized online portal related to agriculture, livestock, poultry, fisheries, Environment and Agricultural technology activities. Sector-5, RD-1, H-39, Uttara Model Town, Dhaka-1230, Bangladesh.
- Agrinews24.com*. 2017. An online portal related to agriculture, livestock, poultry, fisheries. House-54, Road-01, Washpur, Hazaribagh, Dhaka, Bangladesh.

- Ali, S. 2020. Poultry Production Falls Alarmingly. <https://tbsnews.net/coronavirus-chronicle/covid-19-bangladesh/poultry-productionfalls-alarmingly-80857> (accessed December 28, 2020).
- BPICC, 2020. Bangladesh Poultry Industries Central Council. Press Conference at Dhaka Office. Rupayan Shopping Complex, Bashandhara Residential Area, 300 Foot, Dhaka-1229.
- BPICC, 2022. Report of Bangladesh Poultry Industries Central Council. Dhaka Office. Rupayan Shopping Complex, Bashandhara Residential Area, 300 Foot, Dhaka-1229.
- BEPA, 2022. Bangladesh Egg producers Association. About 3.5 to 4.0 crore eggs are required daily to meet national demand.
- Berkhout, N. 2020. Bangladesh Poultry Production Plumets. Available online at: <https://www.poultryworld.net/Meat/Articles/2020/5/Bangladesh-poultryproduction-plumets-585903E/>.
- Das, S.C., Chowdhury, S.D., Khatun, M.A., Nishibori, M. and Yoshimura, Y. 2008. Poultry production profile and expected future projection in Bangladesh. *J. World Poultry Sci.* 64: 98–118.
- Dhaka Tribune, Coronavirus. 2020. Local Poultry Industry Facing a Massive Loss. <https://www.dhakatribune.com/health/coronavirus/2020/04/03/coronavirus-local-poultry-industry-facing-a-massive-loss>. (Accessed 24 November 2020).
- DLS, 2022. Livestock Economy at a Glance (Provisional). Prepared by Planning Section, Department of Livestock Services (DLS), Khamarbari, Farmgate, Dhaka,
- FAO, 2020. Food and Agriculture Organization. Mitigating the Impacts of COVID-19 on the Livestock Sector. <http://www.fao.org/3/ca8799en/CA8799EN.pdf>
- Hamid, M., Rahman, M., Ahmed, S. and Hossain, K.M. 2017. Status of poultry industry in Bangladesh and the role of private sector for its development. *Asian J. Poult. Sci.*, 11:1–13.
- IDLC, 2020. “Feed industry of Bangladesh: sustaining COVID-19 and potentials in upcoming days,” in IDLC Monthly Business Review. Available online at: <https://idlc.com/mbr/images/public/xWV4Ylp7Dg1TyTKLENXGmW.pdf> (accessed December, 2020).
- Islam, S., Hossen, F. and Rahman, M.M. 2021. Productivity and Profitability Estimates for Sonali Chicken Farm in Rajshahi, *Bangladesh. J.Sci.Res.* 13(3):989-998. DOI:10.3329/jsr.v13i3.52768.
- Kolluri, G., Tyagi, J.S. and Sasidhar, P.V.K. 2020. Research Note: Indian poultry industry vis-à-vis coronavirus disease 2019: a situation analysis report. *Poult Sci.* 2021 Mar; 100(3): 100828. Published online 2020 Nov 19. doi: 10.1016/j.psj.2020.11.011
- Mahmud, N. 2020. Coronavirus: Local Poultry Industry Facing a Massive Loss. <https://www.dhakatribune.com/health/coronavirus/2020/04/03/coronavirus-local-poultry>
- Mahmud, R. 2020. One health poultry hub, COVID-19 and the future for Bangladesh’s poultry sector. <https://www.onehealthpoultry.org/blog-posts/covid-19-and-the-future-for-Bangladeshs-poultry-sector/>.
- MoF, 2020. Ministry of Food. Bangladesh Second Country Investment Plan Nutrition-Sensitive Food System(CIP2 2016-2020) Monitoring Report. Available online: <https://mofood.portal.gov.bd/sites/default/files/mofood.portal.gov.bd/policies>.
- Sattar, A.A., Mahmud, R., Mohsin, M.A.S., Chisty, N.N., Uddin, M.H., Irin, N., Barnett, T., Fournie, G., Houghton, E. and Hoque, M.A. 2021. COVID-19 Impact on Poultry Production and Distribution Networks in Bangladesh. *Front. Sustain. Food Syst.* 5:714649.
- Sharma, S. 2020. Poultry Sector Loses Hope as Sales Hit More by Rumours Than Lockdown; Govt Says, Eating Chicken Safe. Available online at: <https://www.financialexpress.com/economy/poultry-sector-looses-hope-as-sales-hitmore-by-rumours-than-lockdown-govt-says-eating-chicken-safe/1917922/> (accessed December 28 2020).
- The Business Standard. 2020. Deadly Covid-19 Pandemic Impact: Poultry Sector Stares at over Tk1, 150 Crore Losses. <https://tbsnews.net/economy/industry/poultry-sector-stares-over-tk1150-crore-losses-63472>. (Accessed 3 May 2020).

- UFAPRI, 2019. The US Food and Agricultural Policy Research Institute. A report on average per capita meat consumption per person in the world.
- WHO, 2020. World Health Organization. Bangladesh COVID-19 Situation Report No. 10. Available online at: <https://www.who.int/docs/default-source/searo/bangladesh/covid-19-who-bangladesh-situation-reports/who-ban-covid-19-sitrep-10.pdf?sfvrsn=c0aac0b84>.
- Yuhuan, W. and Fu, Q. 2018. Analysis of egg price fluctuation and cause. *J. Agric. Sci.* 10:581–587.

## MATRIX METALLOPROTEINASES (MMPS) EFFECT ON ENDOMETRIUM IN CYCLIC COWS

A. N. M. I. Rahman<sup>1</sup>, L. Y. Asad<sup>2</sup>, B. F. Amy<sup>3</sup> and M. M. Hossain<sup>4</sup>

### ABSTRACT

The aim of this study was to investigate the probable hormonal influence on the expression of matrix metalloproteinases (MMPs) in cow endometrial tissue. The cow uterus of different follicular and luteal stage was collected from an abattoir in Dhaka city. In the laboratory, the tissue was separated from the caruncular and intercaruncular regions of the cow endometrium. MMPs were isolated from the endometrial tissue by using tissue lysis buffer and gelatin zymography have done. Zymograms of the tissue supernatant showed that MMP-9 and MMP-2 both are differentially expressed in the follicular and luteal group. Total MMP-9 and MMP-2 protein expression were found higher in caruncular tissue in the estrous cycle group. Caruncular tissues have more potential to respond to hormonal-induced MMP secretion. Reproductive hormone estrogen (E2) and progesterone (P4) seems to be influenced the clearance of MMPs since E2 and P4 is the prime hormone of the follicular and luteal stage respectively. In conclusion, hormonal contact of the endometrial tissue during the cyclic stage control the MMP secretion and play regulatory activity of extracellular matrix (ECM) remodeling in endometrium during cyclic stage of cows.

**Keywords:** cow, endometrium, estrogen, progesterone, MMP

### INTRODUCTION

The endometrium, a highly specific layer that surrounds the mostly muscular uterine body, has evolved to sustain the dynamic events necessary to establish as well as maintain pregnancy. Throughout the breeding cycle, the mammalian endometrium undergoes morphological and structural changes. The structural rearrangement of living tissue, which depends on the breakdown and reformation of the extracellular matrix (ECM), is commonly referred to as tissue remodeling of the endometrium (Pinet *et al.*, 2019). ECM is a non-cellular three-dimensional macromolecular network composed of collagens, proteoglycans/glycosaminoglycans, elastin, fibronectin, laminins, and several other glycoproteins. Matrix components bind each other as well as cell adhesion receptors forming a complex network into which cells reside in all tissues and organs (Theocharis *et al.*, 2016). The proteolytic destruction of ECM components is mediated by a number of proteases, although the matrix metalloproteinase (MMPs) family members are the most prevalent. MMPs is a large family of zinc-dependent endopeptidases which include the collagenases, gelatinases, stromelysins, metalloelastases and membrane-type MMPs (Birkedal-Hansen *et al.*, 1993; Nagase and Woessner, 1999). For the window of implantation in ruminants, reproductive uterine remodeling before to implantation is crucial, and it has been revealed that MMP plays a crucial function during this time for a healthy pregnancy (Hashizume, 2007). Structural reformation of bovine endometrium during estrous stage also reported by Arai *et al.* (2013). MMPs have a wide range of possible uses, however the connection between MMPs and tissue remodeling in the ruminant endometrium is still unclear. It was reported that, MMPs are regulating the release or activation of several biological factors thus participating in physiological processes (Loffek *et al.*, 2011). Understanding the function of tissue remodeling in endometrial physiology is made possible by the elucidation of the pattern of MMPs secretion in bovine endometrium, which must be crucial for the development of bovine pregnancy. It was reported that MMPs are spatiotemporally expressed in the uterus of cyclic female of mammals and confirm the structural change for maintain the

---

<sup>1</sup>Associate Professor, <sup>2</sup>Professor, <sup>3</sup>Assistant Professor, Dept. of Animal Nutrition, Genetics and Breeding, <sup>4</sup>Associate Professor, Dept. of Anatomy, Histology and Physiology, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh

pregnancy (Zhang *et al.*, 2007). Balanced secretion of this enzyme is important to maintain the surroundings of the endometrium. Most MMPs act extracellularly and at neutral pH and are released as latent precursors requiring activation by proteases such as plasmin, trypsin and elastase, as well as by other activated MMPs. MMPs can be regulated at a number of levels. Transcription is modified by growth factors, cytokines and steroid hormones, the actions of which are tissue- and cell-type specific and vary among the enzymes (Salamonsen and Woolley, 1999). By considering the other studies and their effects, the current work focused on the pattern of expression of MMPs in bovine endometrial tissue to show the likely expressional pattern that regulates tissue remodeling in endometrium during the estrous cycle of cows.

## MATERIALS AND METHODS

The experiment was conducted at the Genetics and Breeding laboratory under the Department of Animal Nutrition, Genetics and Breeding at Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.

### **Preparation of the laboratory**

Before starting the experiment, all the necessary electrical power driven or digital equipment were properly installed and checked for good condition. If needed, these were repaired, reinstalled and finally cleaned and sterilized with 70% alcohol. All the reusable equipment were properly washed, dried, covered with aluminum foil, sterilized and finally kept in a cleaned and sterilized chamber until use and same procedure was applied before reuse. All the essential disposable equipment as well as media, chemicals and reagents were made readily available before starting the experiment.

### **Collection of endometrial tissue from estrous cycle group**

Cow uteri and endometrium were collected from Mohammadpur local slaughterhouse of Dhaka city. Total 30 uterus were collected from slaughter house and among them 16 numbers of uterus were found suitable for sample collection and rest are unsuitable due to degraded or missing ovaries. Missing ovaries or reproductive part found difficult to identify the cyclic stage. This unsuitable uterus found 14 in number. The follicular and luteal stage of the estrus cycle was determined by ovarian morphology with consideration of follicular size and corpus luteum (Ireland *et al.*, 1980). During follicle stage, the ovary contains at least one large follicle and a regressed corpus luteum with no vasculature was visualized on its surface, whereas during luteal stage, a corpus luteum is fully formed with vasculature visible around its periphery. Among 16 number of good uterus 5 number was properly identified the cyclic stage by ovarian morphology. Hence, cow uteri from estrus cycle groups, follicular stage (n=5) and luteal stage (n=5), were selected. In the laboratory, reproductive tract of each cow was removed and trimmed of extraneous tissue. The uterus was immersed in 70% ethanol and then washed two times with Phosphate buffer solution (PBS) at 38.5°C. The uterus was opened longitudinally, and caruncular and intercaruncular part of the endometrium were carefully cut from the lamina propria of the endometrium with scissors and transferred in to serum tubes. On the luminal surface of the mature bovine uterus, four irregular rows of oval caruncles can be observed running the length of each horn. These samples were then stored at refrigerator until further processing.

### **Protein extraction and gelatin zymography**

After collection, cow uterine tissues was homogenized using tissue homogenizer. Briefly, put 50–100 mg of caruncular and intercaruncular tissue sample from each follicular and luteal stage tissue to disrupt in the tube separately. Add 250 µl of lysis buffer [20 mM Tris-HCl (pH 7.5)/150 mM NaCl/2 mM EDTA] in each tube. After fine chopping with sharp scissor insert the pestle into the tube and disrupt the tissue until it appears completely fine in structure. Discard the pestle and add 750 µl of lysis buffer again to increase the volume of the supernatant. After that, vortex it and centrifuged at 15,000 × g for 30 sec. Extra amount of tissue supernatant after centrifuge collect in separate tube, marked it and stored in the refrigerator for further use. To determine the enzymatic activity of MMPs, 1 µg of extract protein samples or 10 µl of harvested medium was subjected to Sodium Dodecyl Sulphate-

Polyacrylamide Gel Electrophoresis (SDS-PAGE) with 10% (w/v) acrylamide gel containing gelatin (0.6 mg/ml) under non-reducing condition.

To make gelatin solution, it was heated at 37°C to properly dissolve the gelatin in the water. After making the gel it was run in 1×SDS running buffer [3.03gm Tris/14.4 gm glycine/1gm SDS] with 10µl of loaded sample mixed with sample buffer. Finally, run the electrophoresis machine for protein separation from the sample. The machine was run for 2hr with 80 voltage until the sample buffer reached in the bottom of the gel. After running, the gel was washed with a washing buffer [50 mM Tris-HCl (pH 7.5)/5 mM CaCl<sub>2</sub>/1 µM ZnCl<sub>2</sub>/2.5% (v/v) Triton X-100/0.02% (w/v) NaN<sub>3</sub>] to remove extra SDS and then incubated at 37°C for 18 hr in an incubation buffer [50 mM Tris-HCl (pH 7.5)/5 mM CaCl<sub>2</sub>/1 µM ZnCl<sub>2</sub>]. Thereafter, the gel was stained with staining buffer [0.1% (w/v) coomassie brilliant blue R-250/ 50% (v/v) methanol /20% (v/v) acetic acid] for 1 hr and destained it by using decoloration liquid. Gelatinolytic activity was detected as unstained bands on blue background of the gel. Densitometric analysis was performed using Image J 1.48 software.

#### **Statistical analysis**

Single factor analysis of variance (ANOVA) was used to analyze the statistical differences where significances was considered at P<0.05. Furthermore, the Student-Newman-Keuls test was used to compare two groups and the differences was considered significant at the level of P<0.05.

## **RESULTS AND DISCUSSION**

In the slaughter house most of the animal found non cyclic and their reproductive history was unknown. After slaughtering most of the reproductive part discarded from the edible portion. During slaughtering, reproductive part as well as uterus was harshly treated and it makes removal of the ovary or part of the uterus from the reproductive organ. As a result it makes difficult to identify the cyclic stage of the uterus and damages occur in the uterine tissues. In this study lot of uterus counted as unsuitable to collect sample due to miss handling of the uterus during slaughter in the slaughter house shown in Table 1. and Table 2. After collection the suitable tissue, enzymatic activity of MMP-9 and

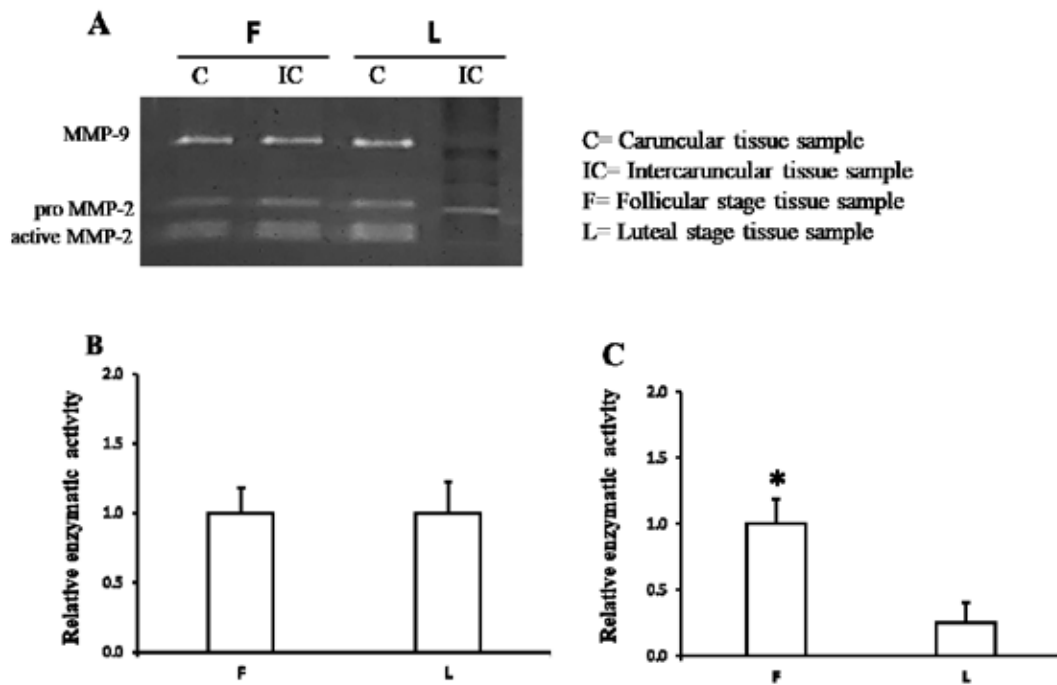
**Table 1. Total number of cows uterus collected from slaughter house**

<b>Total Number of cow uterus collected (n=30)</b>	
No. of uterus suitable for sample collection (n)	No. of uterus not suitable for sample collection (n)
16	14

**Table 2. Numbers and parameters of identified cyclic stage uterus.**

<b>Types of uterus in cyclic stage</b>	<b>Number (n)</b>	<b>Parameter</b>
Follicular stage	5	-Large follicle present -Regressed corpus luteum present -No vasculature in ovarian surface
Luteal stage	5	-Small follicle present -Fully formed corpus luteum present -Fully formed vasculature visible in ovarian surface

MMP-2 in cow endometrial tissue were analyzed by gelatin zymography (Fig. 1). Pro-form of MMP-9 was detected in caruncular tissue of both follicular and luteal stage. On the other hand pro-form MMP-9 was only detected in intercaruncular tissue in case of follicular stage but absent in luteal stage. Furthermore, MMP-2 enzymatic activity was detected during all stage of estrous cycle. Both MMP-2 activity of the precursor and mature forms were strongly detected at follicle stage in caruncular and intercaruncular tissue, then the activity become weak at luteal stage. Relative enzymatic activity of



**Fig. 1.** Expression of MMPs in cow endometrium. Enzymatic activity of MMP-9 and MMP-2 in cow caruncular and intercaruncular tissue during estrous cycle (A). Total enzymatic quantification in caruncular (B) and intercaruncular tissue (C).

caruncular tissue for follicular and luteal stage found no significant ( $P > 0.05$ ) difference. Whether as significance differences ( $P < 0.05$ ) were observed in intercaruncular tissue in between follicular and luteal stage. It was reported that during estrous cycle, the ECM component collagen type-I and -IV exhibit changes clearly visible both in structures and amounts in bovine endometrium (Boss, 2000). In cattle, the role of ECM remodeling in reproductive process is poorly understood as only few studies were performed in ruminants at the implantation period (Mishra *et al.*, 2010). For the process of ECMs remodeling, MMPs is considered as an important mediators (Visse and Nagase, 2003). To unravel the regulatory mechanisms of MMPs in the endometrium, in vitro culture models were typically used. Previous reports suggested that the expression of several MMPs was controlled by steroid hormone (Vassilev *et al.*, 2005) and by soluble local regulators such as hormone and other factors (Pretto *et al.*, 2008). In this study, tissue were collected from slaughter house uterus of different estrous group where E2 and P4 was key regulator of this stage which regulate different biomolecule including MMP. The expression of MMP-9 and MMP-2 has also been reported within the uterus of domestic animals including bovine (Hashizume *et al.*, 2003), goats (Uekita *et al.*, 2001) and sheep (Salamonsen *et al.*, 1995) respectively. Although, the information in this regard is very limited. It was reported that, MMP-2 were secreted by cultured ovine endometrial stromal, but not epithelial cells whereas MMP-2 production is constitutive (Salamonsen *et al.*, 1995). Our report also support the previous statement where MMP-2 consecutively secrete follicular and luteal tissue. Although the patterns of specific MMP expression that are regulated by estrogens or progesterone are not yet fully established, since lot of factor is involved in in vivo condition. Accumulated research suggests that ovarian steroids affect the gene expression of the MMP system in the uterus. P4 appears to down-regulate some MMPs in the uterus. Though MMP-9 and MM P-2 secrete in luteal stage but in this study MMP-9 and MMP-2 seems to be less secret in luteal stage intercaruncular tissue. It might be due to P4 effects. Additionally,

findings of MMPs expression in cow endometrial tissues in this study also supported this suggestion because enzymatic activity of MMP-9 was hardly detected in all stages. The cellular difference of caruncular and intercaruncular area is well documented. Caruncular and intercaruncular area are covered by dense stroma and large number of branched glands respectively. This structural distinction also indicate the functional inequality of the endometrium also. The difference of enzymatic secretion of caruncular and intercaruncular region confirm the cellular dissimilarity of the specific region of the endometrium. The alteration of the MMP-9 and MMP-2 secretion may also regulated by the unknown cell secretory factor. However, from this data it is assumed that after synthesis of MMP-9 and MMP-2 it does not release in the tissue environment immediately, somehow it stored in the tissue which can be detected through zymography otherwise it cannot be detected after long time of tissue processing. These results suggested that other factors except E2 and P4 might have another effects on the expression of MMP-9 and MMP-2 mRNA in cow endometrium. Since different form of MMP-9 and MMP-2 are differentially expressed in the tissue; it is possible to predict that there activation from pro to mature form is regulated by some inherent factor from the tissue cells which regulate the activation for specific function of the endometrium. In summary, this study revealed that hormonal influence regulated the MMPs clearance and that released MMPs induced ECMs remodeling in cow endometrium.

## CONCLUSION

In summary, this study revealed that steroid hormone regulated MMPs clearance and that released MMPs induced ECMs remodeling in cow endometrium. Furthermore, caruncular and intercaruncular site of the endometrium differentially expressed the MMPs. Since placentation is occurred in caruncular site hence MMPs role is to remodel the implantation site for successful implantation and this remodeling occurs continuously in each cycle of cow. However, to explore more in MMPs expression; mimic physiological condition is necessary. That's why spheroids are more suitable for analyzing the regulatory mechanisms of ECMs remodeling and the related factors in the cow endometrium compared with monolayer cell culture models should applied in future.

## ACKNOWLEDGEMENTS

Authors are grateful to Sher-e-Bangla Agricultural University Research System (SAURES), Dhaka, Bangladesh for giving fund to conduct the research program.

### Compliance with ethical Statements

Conflict of interest: The authors declare that they have no conflict of interest.

## REFERENCES

- Arai, M., Yoshioka, S., Tasaki, Y. and Okuda, K. 2013. Remodeling of bovine endometrium throughout the estrous cycle. *Anim. Repro. Sci.*, 142(1-2): 1-9.
- Birkedal-Hansen, H., Moore, W.G., Bodden, M.K., Windsor, L.J., Birkedal-Hansen, B., DeCarlo, A. and Engler, J.A. 1993. Matrix metalloproteinases: A review. *Crit. Rev. Oral. Biol. Med.* 4:197–250.
- Boos, A. 2000. Immunohistochemical assessment of collagen types I, III, IV and VI in biopsy samples of the bovine uterine wall collected during the oestrous cycle. *Cells. Tiss. Org.* 167(4): 225-238.
- Hashizume, K. 2007. Analysis of uteroplacental-specific molecules and their functions during implantation and placentation in the bovine. *J. Repro. Dev.* 53(1): 1-11.
- Hashizume, K., Takahashi, T., Shimizu, M., Todoroki, J., Shimada, A., Hirata, M., Sato, T. and Ito, A. 2003. Matrix-metalloproteinases-2 and-9 production in bovine endometrial cell culture. *J. Repro. Dev.* 49 (1): 45-53.

- Ireland, J.J., Murphee, R.L. and Coulson, P.B. 1980. Accuracy of Predicting Stages of Bovine Estrous Cycle by Gross Appearance of the Corpus Luteum. *J. Dairy Sci.* 63(1): 155-160.
- Löffek, S., Schilling, O. and Franzke, C.W. 2011. Biological role of matrix metalloproteinases: a critical balance. *Euro. Resp. J.* 38(1): 191-208.
- Mishra, B., Kizaki, K., Koshi, K., Ushizawa, K., Takahashi, T., Hosoe, M. and Hashizume, K. 2010. Expression of extracellular matrix metalloproteinase inducer (EMMPRIN) and its related extracellular matrix degrading enzymes in the endometrium during estrous cycle and early gestation in cattle. *Repro. Biol. Endocrin.* 8(1): 60.
- Nagase, H. and Woessner Jr, J.F. 1999. Matrix metalloproteinases. *J. Biol. Chem.* 274(31): 21491–21494.
- Pretto, C.M., Gaide, Chevronnay, H.P., Cornet, P.B., Galant, C., Delvaux, D., Courtoy, P.J., Marbaix, E. and Henriët, P. 2008. Production of interleukin-1alpha by human endometrial stromal cells is triggered during menses and dysfunctional bleeding and is induced in culture by epithelial interleukin-1alpha released upon ovarian steroids withdrawal. *J. Clin. Endocri. Meta.* 93: 4126–4134.
- Pinet, K. and McLaughlin, K.A. 2019. Mechanisms of physiological tissue remodeling in animals: Manipulating tissue, organ, and organism morphology. *Devel. Biol.*, 451(2): 134-145.
- Theocharis, A.D., Skandalis, S.S., Gialeli, C. and Karamanos, N.K. 2016. Extracellular matrix structure. *Adva. Drug. Deli. Revie.*, 97: 4–27.
- Salamonsen, L.A., Nagase, H. and Woolley, D.E. 1995. Matrix metalloproteinases and their tissue inhibitors at the ovine trophoblast-uterine interface. *J. Repro. Fertil. Suppl.*, 49: 29-38.
- Salamonsen, L.A. and Woolley, D.E. 1999. Menstruation: induction by matrix metalloproteinases and inflammatory cells. *J. Repro. Immun.* 44(1-2): 1-27.
- Uekita, T., Tanaka, S.S., Sato, H., Seiki, M., Tojo, H. and Tachi, C. 2001. Expression of membrane-type 1 matrix metalloproteinase (MT1-MMP) mRNA in trophoblast and endometrial epithelial cell populations of the synepitheliochorial placenta of goats (*Capra hircus*). *Arc. Hist. and Cyto.*, 64 (4): 411-424.
- Vassilev, V., Pretto, C.M., Cornet, P.B., Delvaux, D., Eeckhout, Y., Courtoy, P.J., Marbaix, E. and Henriët, P. 2005. Response of matrix metalloproteinases and tissue inhibitors of metalloproteinases messenger ribonucleic acids to ovarian steroids in human endometrial explants mimics their gene- and phase-specific differential control in vivo. *J. Clin. Endocri. Meta.* 90:5848–5857.
- Visse, R. and Nagase, H. 2003. Matrix metalloproteinases and tissue inhibitors of metalloproteinases: structure, function, and biochemistry. *Circul. Res.* 92(8): 827-839.
- Zhang, X., Christenson, L.K. and Nothnick, W.B. 2007. Regulation of MMP-9 expression and activity in the mouse uterus by estrogen. *Mol. Repro. Devel. Inco. Gam. Res.*, 74(3): 321-331.

## PRODUCTION STRATEGY AND ITS EFFECT ON SPOTTED DEER AT NATIONAL ZOO IN BANGLADESH

M. R. Islam<sup>1</sup>, M. J. Alam<sup>2</sup> and A. K. M. B. I. Talukder<sup>3</sup>

### ABSTRACT

The production and reproduction profile, feeds and feeding and herd management of spotted deer studied in captive condition at Bangladesh National Zoo in this experiment. For these purposes, fifteen (15) deer's from each group i.e. adult male (Stag or buck), adult female (Doe), juvenile and infant were used during January 15<sup>th</sup> to December 15<sup>th</sup> 2018 for a period of eleven months. Available feeds that were supplied to the spotted deer such as maize fodder, Jambu grass and Gourd Spinach (Kolmi grass), cabbages, cucumber, gourd, pumpkin, grain, wheat bran and soybean meal. Amount of CP and metabolic energy was 13.63% and 14.38 MJ ME per day respectively. From the study it was observed that average males and females' birth weights were 2.97 and 2.73 kg, adult males and females weights 78.08 and 60.97 kg, males and females weaning weights were 19.05 and 18.49 kg respectively. It was also observed that average weaning age was 5.19 months, length of estrous was 17.40 days, age at first fawning was 14.48 months and gestation lengths were 232.69 days respectively. It is concluded that recommended rations need to be fed with proper management systems to keep balanced nutritive conditions along with productive and reproductive well-being of the spotted deer at Bangladesh National Zoo.

**Key words:** *Axis axis*, chital deer, chittra deer, spotted deer, zoo

### INTRODUCTION

Spotted Deer is widely distributed in all habitats of the Sundarbans. The natural distribution of spotted deer (*Axis axis*) is limited in to the Indian sub-continent, although successful introductions occurred elsewhere (Geist, 1999; Seidensticker *et al.* 1999). The Spotted Deer ranges between 52600 (Khan 1986) and 83000 (Dey 2007). The relative abundance of Spotted Deer varies from habitat to habitat, a gradual reduction from west to east (Deodatus and Ahmed, 2002) and deer density increases with the habitat ensuring fresh drinking water (Feeroz, 2001). Apart from the Sundarbans, several islands in the Bay of Bengal also support large population of Spotted Deer. The availability of drinking water, trees for shade, grass for forage, and in presence of the high rugged terrain are four factors that influence the spotted deer concentration in certain areas. Deer have been morpho-physiologically classified as ruminants. Their main fodder in the forest are the leaves and fruits of keora, new leaves of passur, gewa, and various grasses, but they met their calcium requirement by chewing fallen deer antler and eating crab, shrimp etc. In Bangladesh National Zoo (BNZ), the number of spotted deer & barking deer are 241 and 11 respectively. To keep them nutritionally sound everyday a balanced ration is supplied containing green fodders (Maize fodder, Jambu grass, and gourd spinach), vegetables (cabbage, cucumber, pumpkin) and concentrate feeds (Grain, Soybean meal, wheat bran, vitamin-mineral and common salt). Deer is highly sensitive, agile and powerful. They are hardly an easy experimental animal to work with. Nevertheless, feeding of captive deer is now greatly simplified by the

---

<sup>1</sup>MS, <sup>2</sup>Professor, Dept. of Animal Production & Management, Sher-e-Bangla Agricultural University, <sup>3</sup>Senior Scientific Officer, Animal Nutrition Section, Livestock Research Institute, Dept. of Livestock Services, Farmgate Dhaka, Bangladesh

development of a pelleted ration which seemingly provides the essential nutrients for optimum growth and maintenance. In future it could be an expedient to supply complete pelleted ration for free roaming deer that are faced with acute browse shortage and severe environmental pressure. Deer farming is an economically promising industry. To establish deer farming for the production of venison and other by-products, it is essential to understand and develop effective ex-situ management systems of spotted deer in Bangladesh. Conservation of nature and natural resources are now a global concern. Without proper scientific knowledge on conservation management, natural resource utilization is very difficult. Therefore, the present study was undertaken to find the feeding management strategy to explore the production and reproduction profile of deer at Zoo condition.

## MATERIALS AND METHODS

### Location, environmental condition, animals and study period

The study was conducted with the spotted deer at the Bangladesh National Zoo. The experiment was carried out from January 15<sup>th</sup> to December 15<sup>th</sup>, 2018 for a period of eleven months. The production of spotted deer in a Zoo, tourist and their interaction are greatly influenced by the local environmental condition. Therefore, environmental condition of the study area is overviewed. Maximum and minimum temperatures as observed in May and January ranged between 33°C and 15-20°C. Summer season continued from April to June 25- 30°C and winter lasted from December to February. Rainfall started in May and continued up to September. About 5% of the annual rainfall occurred during the monsoon. The maximum humidity was observed 96% from July to September and the minimum about 45% from January to April (BBS, 2000). The necessary data for this experiment was collected from the following numbers of deer such as adult male, adult female, young (Juvenile) and infants as 15, 15, 15 and 15 respectively at Bangladesh National Zoo.

### Parameters studied

The present study covered the following aspects of spotted deer such as feeds and feeding, production and reproduction profile and herd management.

### Feeds and feeding

*Axis axis* consume an extremely wide range of forage items throughout their native range and in introduced locales. They eat over 75 species of plants, as well as the full spectrum of plant parts including leaves, stems, fruits, seeds, flowers and bark. Their diet consists largely of grasses in all seasons, augmented with browse. Green grasses less than 10cm high are preferred.

Chital deer are ruminant herbivores. (Sub order Ruminantia). Deer have a four-chambered stomach. The first chamber, called the rumen, is for storage. The rumen allows for the deer to gather a lot of food at once and then digest it later. The deer bring the food back up into their mouth and chew it again. This process is called chewing their cud. It is also called ruminating, named after the rumen. Animals that can do this are called ruminants. The reticulum is the second stomach chamber. This is where the microorganisms live. The microorganisms attack the chewed food that the deer has eaten. This process is called fermentation. This helps to break the cellulose down into simpler substances that can be absorbed by the deer and the microorganisms. Fermentation produces a gas (methane), which the deer must discharge very regularly. When deer chew their cud again, mixed in with the digested food are microorganisms. The deer chew the microorganisms and a lot of deer's nutrition comes from them. There are plenty of microorganisms left in the reticulum. This time when it goes back down, the chewed food goes to the third chamber the omasum. This is where water is absorbed. Finally, the resulting cud enters the last chamber, (the abomasum), where gastric juices continue digestion. Gastric juices are liquids, like the acids in your stomach that help digest food. Last, it moves on to the intestines. This is where the food is absorbed by the animal's body. This is where the animal receives the nutrients for his body. The deer's intestines are 28 feet long. When the animal has absorbed everything it can use, everything that isn't digested is passed off as waste droppings.

Deer seek out the highly palatable feed in preference to feed of medium or low palatability. Their feed utilization is influenced considerably by the season. Highest feed intake occurs during the spring when

plants contain highest protein and medium intake occurs during the winter when feed is scarce. During the rut, males ingest little or no feed was reported by Vos (1982). Deer have been morpho-physiologically classified as ruminants, which readily browse shrubs, forbs and grasses (Jean *et al.*, 1995). Deer can eat and digest any kind of feedstuffs used for more traditional domestic ruminants. In digestibility trials deer have been found to digest poor quality roughage diets less completely but high quality concentrate diets more than sheep. This part covered for feed sources, chemical composition of feed, water sources, supplemental feeding and seasonal effect on feed intake. Chemical composition of the supplied feed were analyzed according to AOAC (2005).

### **Production profile**

This part covered mainly for the birth weight of male and female, adult weight to female, weaning weight of male, weaning weight of female, weaning age, length of estrous, age at first fawning, gestation length and sex ratio.

### **Housing Requirements**

The position of the enclosure is irrelevant as long as the deer have shade from large trees and have a sheltered area for poor weather conditions.

The enclosure needs to have a sheltered area for the deer to retreat to in poor weather. There are no size requirements but will need to protect the total number of deer's within the exhibit. There are no temperature requirements. Deer prefer warmer environments with cool areas and this can be provided with trees as it wood replicates the forest. Preferred substrate materials: mulch and pine chips, dolerite other suitable materials: Grass Materials to avoid: Cement (as the harshness to hoofs and joints). Straw and saw dust are the preferred materials for bedding. Which should be completely changed at least once a week? Heat lamps offered in bedding area in the cooler months will provide deer with extra warm, especially when fawn are in the herd. Bedding areas should be under shelter to avoid wet materials. Suitable furnishings would include large tall trees. Mud wallows and scratching poles would offer enrichment to the chital deer. Large logs and nature items.

### **General Husbandry**

#### **Hygiene and Cleaning**

Daily cleaning tasks: Spot cleaning feces, cleaning drinking water, removing old browse and left over foods; Weekly cleaning tasks: Complete change of bedding material; Monthly cleaning tasks: Scrub any cement areas; Half yearly: New substrate – mulching; Cleaning agents suitable: Animal house bleach 4% - cleaning holding and cement areas.

Pest control boxes should be placed in areas in and around enclosure and checked weekly and are refilled and any pest removed. Disposing of pests-follow intuitions procedures. Taranga's – all dead animals are to be taken to VQC for post mortem.

#### **Record Keeping**

All animals should be individually identifiable, with the use of colored or numbered ear tags. Each animal should have its own file which contains identification numbers or tags of animals, ARKS number, Health problems, Veterinary examinations, Veterinary treatments, Behavioral data, Reproductive stages, condition and behaviors, Gene pool information, Parents, Birth dates, Changes in diet, Movements within and between institutions, Body mass and measurements, History of animal, Enrichment behaviors and reactions, Transfer details, individual characteristics etc.

#### **Routine Data Collection**

Most information is on deer farming. Some of this information does work for deer in captive intuition but most is for the production of venison. There have been many studies on wild deer and many on farming deer's. Information that may be acquired for long term studies includes Weights/growth–development charts, Contraception/genes/parenting behaviors, Blood biochemistry

#### **Reproduction and selection**

The mating system of these deer is not known. However, in other, related deer, the most common mating system is polygyny. Males compete with one another for access to estrous females. Competition often involves sparring and vocalizing. Successful males are typically older and larger, and able to drive away younger, smaller males. These successful males are the ones who mate with the females. It

is likely that Visayan spotted deer have a similar mating system. In the wild, *Axis axis* bucks are found throughout the year with hardened antlers and in rutting condition. The reproductive cycle of each individual is not synchronized with that of other males in the herd. Concurrently, throughout the year, some bucks are coming into rut, while others are going out of rut, or are in a non-breeding condition. Females also experience non-synchronized estrous cycles, with each cycle lasting about 3 weeks. Bucks do not retain harems of does, but instead mate with does in each herd as they become receptive. One fawn is typically produced per pregnancy and gestation lasts 210-238 days. Following parturition, females again mate during the subsequent breeding period. Adult females tend to produce one fawn each year. The current reproduction system may be defined as a controlled, single-sire and natural mating:

#### **Restraint Systems**

Mechanical, manual and chemical restraint systems were followed for control and handling the animals.

#### **Weighing Animals**

The systems available are electronic or mechanical and are suspended or platform. All of the restraints can be mounted on either system. Using the restraint as a weight crate means the animals can be weighed each time they are in the restraint. It also means they have to enter the restraint each time they are weighed. The alternative is to put a separate weigh crate in the tunnel, most easily with platform scales, with the facility to draft the animals after the weigh scales and before the restraint. This is most easily achieved with a long tunnel.

#### **Health Management**

One key component of herd management is disease control. It is the responsibility of each farmer to manage his animals in such a manner that they are healthy and in good condition because it makes the animals more resistant to infections and disease. Prevention of disease is better than any possible cure. It is important to establish a good working relationship with a veterinarian. Together, the farmer and the veterinarian can work out a program tailored for any individual farm to control disease and to maximize the health and productivity of the animals. The causes of diseases can be categorized as Intrinsic flaws: Nutritive deficiencies; Exogenous poisons: Trauma, Tumours (neoplasms) and Parasitic or other living organisms (i.e. parasites, bacteria, fungi, viruses, mycoplasma and rickettsia).

Deer at low population densities on natural range are generally not affected by disease to any significant extent. Deer in their first 12-15 months of life are more susceptible to disease than adults. Treatment of sick deer is analogous with that of domestic animals. Prevention of diseases by nutritional management, testing, vaccination, drenching and dipping, is more important than treatment. The deer's in Bangladesh National zoo, veterinary facility is provided periodically or when necessary. A vaccine is given which contains BQ, HS, and FMD within 3-4 days of newly born deer. After the birth of a baby deer, the workers put a tag on the body of the spotted deer within 2-3 days of birth.

#### **Statistical analysis of data**

Data recorded for different parameters were compiled and tabulated in proper way for statistical analysis. Analysis of mean, standard error of mean, standard deviation and variance was done with the help of SPSS (Version 20.0).

## **RESULTS AND DISCUSSION**

The results of following parameters such as feeds and feeding, production and reproduction profile and herd. management have been presented.

#### **Feeds and feeding**

The chemical composition of the feeds supplied to the deer are showed in the (Table-1). From the table 2, it was observed that the total amount of feed intake was 8.12 kg as fresh basis and 2.079 kg as DM basis. During the experimental period maize fodder (*Zea mays*) and gourd spinach (*Impomoea aquatica*) as leafy vegetable, pumkin (*Cucurbita moschata*) and cucumber (*Cucumis Sativus*) as

**Table 1. Chemical composition of feed consumed by experimental spotted deer at Bangladesh National Zoo (AOAC, 2005)**

Name of feed		% DM	% CP	ME (Kcal/kg)
Local name	Botanical name			
Maize fodder/Jambu grass	<i>Zea mays/Sesbania bispinosa</i>	18	7.91	230
Gourd spinach	<i>Impomoea aquatica</i>	15	8.1	131
Cabbage	<i>Brassica oleracea</i>	10	1.3	260
Pumkin	<i>Cucurbita moschata</i>	14	2.1	430
Gourd	<i>Lagenaria siceraria</i>	14	1.9	420
Cucumber	<i>Cucumis sativus</i>	14.1	1.8	410
Grain	<i>Cicer arietinum</i>	12.51	19.68	720
Soybean meal	-	88	45	2500
Wheat bran	-	88	14	1600
Common salt	-	90		-
Vit-min premix	-	-	-	-

vegetable, grain (*Cicer Arietinum*) and as concentrate soybean and wheat bran were supplied to the deer at BN Zoo. The percentage of crude protein (CP) and amount of metabolisable energy of the supplied ration for an adult deer were 13.63% and 14.38 MJ per day (Table 2).

Spotted deer that are free browsers intake a various types to grasses, creepers, shrubs, tree leaves, vegetables etc. But deer at Bangladesh National Zoo, that are confined can not take any feed other than supplied. A little amount of grasses was browse there. This amount is very negligible. From the data of BN zoo authority it was observed that green fodder, leafy vegetables and succulent vegetable were changed in the various seasons and depends on availability in the market. Khira (*Cucumis sativas*), Sasa (*Saha senanensis*), Misty Cumra (*Cucurbita moschata*), Cabbage (*Brassica oleracea*) were supplied in the various seasons of the year.

**Table 2. Feed with nutrient intakes by the spotted deer at Bangladesh National Zoo**

Name of feed		Amount supply (kg)/deer	DMI (kg)/deer	% CP intake/deer	ME (kcal/kg)/deer
Local name	Botanical name				
Maize fodder /Jambu grass	<i>Zea mays Sesbania bispinosa</i>	3.0	0.54	2.07	690
Gourd spinach grass	<i>Impomoea aquatica</i>	2.0	0.3	1.15	262
Cabbage	<i>Brassica oleracea</i>	0.5	0.05	0.033	130
Pumkin	<i>Cucurbita moschata</i>	0.6	0.084	0.086	258
Gourd	<i>Lagenaria siceraria</i>	0.5	0.07	0.063	210
Cucumber	<i>Cucumis sativus</i>	0.4	0.056	0.048	164
Grain	<i>Cicer arietinum.</i>	0.2	0.177	1.68	144
Soybean meal		0.26	0.229	4.95	650
Wheat bran		0.6	0.528	3.55	960
Common salt		0.05	0.045		-
Vit-min premix	-	0.01			
Total		8.12	2.079	13.63	14.38 MJ

The crude protein is far below than the requirement to the findings of Moon et al. (2000) but metabolisable energy is more or less near to the requirement. They investigated that DMI is around 2.0% in winter increasing to 2.3% in summer for adult spotted deer. DM is around 2.7% to 3.8% for fallow deer (Wilson,1979). Adam (1994) stated that in autumn, winter, spring and summer DMI is 1.7,

2.0, 2.3, 3.0 kg/d/hinds where as 1.4, 1.3, 2.0, 2.2 kg/d/growing calves. Fennessy (1981) stated that an adult spotted deer required for maintenance energy was 16-18 MJ where as a protein diet in the 13-16% range is required for the successful growth, antler development and reproduction (Verme and Ullrey, 1972).

**Table 3. Nutrient balances for the spotted deer**

Characteristics	CP %	ME (MJ)
Requirement	14.5	16.
Supplied	13.63	14.38
Balance	-0.87	-1.62

A high level of crude protein (approximately 16%) is advantageous for fallow fawns to achieve their target live weights (Malley et al., 2001). Adam (1994) stated that in calves, hinds and stags for the season of autumn, winter, spring and summer CP is required as 17, 10, 12-17; 10, 14, 17 and 10, 12% respectively. Female fawns (white-tailed deer) required 13% crude protein for maximum growth (Ullrey et al., 1967). Denholm (1984) stated that estimates of dietary crude protein requirements for optimum growth vary within the range of 13 to 15%. French et al. (1956) concluded that young male fawns (white-tailed deer) required 13% to 16% crude protein.

The energy concentration for empty body weight is 17.5 MJ/kg and 21.7 MJ/kg and protein content is 150 g/kg and 146 g/kg (ARC, 1980). From Adam (1994) standard of nutritional requirement of spotted deer, it was observed that crude protein deficiency was 4.5% and the ME deficiency was 1.66 MJ per day that are supplied to the deer (Table 3).

There is an imbalance of crude protein and energy supply to the deer of BN zoo. Proper growth and development both in productive and reproductive may not be perform due to insufficient protein supply and in balance of CP and ME. Recommended ration for deer is shown in Table 4.

**Table 4. Recommended rations for spotted deer**

Name of feed		Amount supply (kg/deer)	DMI (kg) deer	% CP intake / deer	ME (kcal/kg) / deer	
Local	Botanical					
Maize fodder	Jambu grass	<i>Zea mays</i>	3.00	0.54	2.07	690
Gourd	spinach	<i>Impomoea aquatica</i>	2.00	0.30	1.15	262
Cabbage		<i>Brassica oleracea</i>	0.5	0.05	0.033	130
Pumkin		<i>Cucurbita moschata</i>	0.6	0.084	0.086	258
Gourd		<i>Lagenaria siceraria</i>	0.6	0.08	0.066	252
Cucumber		<i>Cucumis sativus</i>	0.5	0.060	0.058	164
Grain		<i>Cicer arietinum.</i>	0.3	0.187	1.78	216
Soybean meal			0.3	0.239	0.500	750
Wheat bran			0.7	0.548	3.65	1120
Common salt			0.05	0.045		-
Vit-min premix		-	0.001	-	-	-
Total			8.5	2.088	13.893	15.82 MJ

### Production and reproduction profile

Body weight of spotted deer at BN Zoo is presented in table 5. The average birth weight of males and females spotted deer fawn were 2.97±0.17 kg and 2.73±0.17 kg. The highest and lowest birth weights of them were 4.10 kg to 1.80 kg and 3.90 kg to 1.70 kg respectively. The average birth weight of males and females fawn of spotted deer were more or less similar to the findings of Mulley (1984). He

reported that the mean birth weight of male and female spotted deer fawn was 4.5 kg and 4.2 kg respectively. Ables, (1977) reported that the average birth weight of males and females spotted deer fawn were 3.5 kg and 3.0 kg respectively. Fawn birth weight is generally 10% of their mother's weight reported by Kay and Staines (1981); Mulley *et al.* (1990).

In the present study, the adult males and females' weights were  $78.08 \pm 2.17$  kg and  $60.97 \pm 1.77$  kg respectively. The highest and lowest adult males and females weights were 88.90 kg to 62.20 kg and 74.50 kg to 51.30 kg respectively.

**Table 5. Body weight of spotted deer at Bangladesh National Zoo**

Traits	No. of animal	Mean	Standard error mean	Standard deviation	Maximum	Minimum
Male birth wt. (kg)	15	2.97	0.17	0.67	4.10	1.8
Female birth wt. (kg)	15	2.73	0.17	0.65	3.90	1.70
Adult male wt. (kg)	15	78.08	2.17	8.39	88.90	62.20
Adult female wt. (kg)	15	60.97	1.77	6.87	74.50	51.30
Male weaning wt. (kg)	15	19.05	0.76	2.96	24.30	15.30
Female weaning wt. (kg)	15	18.49	0.58	2.24	22.30	15.20

The average adult males and females weights were more or less similar to the findings of Ables (1977). He concluded that the average adult males and females weights must be attained within 85.00 kg and 70.00 kg. There is strong relationship between body weight and reproduction. Sharman (1978) stated that young hinds weighing less than 60.00 kg at rutting did not produce calves. Kelly and Moore (1978) showed that the percentage of red deer calves dropped by yearling hinds less than 65 kg in liveweight was only 50%, while above 65kg it could be about 90%. Mitchell and Brown (1974) suggested that a liveweight of 75 kg for hinds should give 80% of calving percentages.

Table 5 showed that the average males and females weaning weights were  $19.05 \pm 0.76$  kg and  $18.49 \pm 0.58$  kg respectively. The highest and lowest weights of them were 24.30kg to 15.30 kg and 22.30kg to 15.20 kg respectively. The average males and females weaning weights in this study were more or less similar to the findings of Mulley (1984) who stated that the mean males and females weaning weights were 19.4 kg and 18.5 kg. Weaning generally takes place at an average weight of 18.00 to 21.00 kg reported by Mulley *et al.* (1990).

**Table 6. Reproductive performance of spotted deer at Bangladesh National Zoo**

Traits	No. of animal	Mean	Standard error mean	Standard deviation	Maximum	Minimum
Length of estrus (Days)	15	17.40	0.54	2.10	21.10	14.50
Age at 1 <sup>st</sup> estrus (Months)	15	14.48	0.43	1.65	18.30	11.50
Gestation period (Days)	15	232.69	1.48	5.73	241.10	223.70
Age at 1 <sup>st</sup> fawning (Months)	15	14.58	0.43	1.68	18.20	12.00
Weaning age (Months)	15	5.19	0.18	0.68	6.20	4.00

In the present study, the average length of estrous cycle was  $17.40 \pm 0.54$  day. The highest and lowest lengths of estrous cycle were 21.10 day and 14.50 day respectively. The mean length of estrous cycle was more or less similar to the findings of Chappelle *et al.* (2003). They reported that mean duration of estrous cycle of chital hinds was  $19.30 \pm 1.30$  day with a range of 17-21 days. Vos (1982) showed that the mean length of estrous cycle in spotted deer was 18.20 days. The mean length of estrous cycle was  $18 \pm 0.7$  days with a range of 12-23 days concluded by Mylrea *et al.* (2004).

From the table-6 showed that the average duration of gestation length was 232.69±1.48 days. The highest and lowest lengths of gestation period were 241.10days and 223.70 days. The average length of gestation period in this study were almost similar to the findings of The mean duration of gestation period was 234.5±3.0 days at chital hinds reported by Chapple *et al.* (2003), that is more or less similar to the present study. Ables (1977) showed that only one fawn is produced per pregnancy after a gestation period of 210-238 days. Vos (1982) reported that only one fawn and rarely twin is produced per pregnancy after a gestation period of 226-233 days.

From the table-6 was observed that the average age at first fawning was 14.58±0.43 months. The highest and lowest age at first fawning was 18.20 months and 12.00 months respectively. In the present study the average age at first fawning were almost similar to the findings of Ables (1977) who reported that does may first bred at an age of 14-17 months. Vos (1982) showed that does may be first bred within 1.5-2 years.

From the table 6 it has been also observed that the average weaning age was 5.19±0.18 months. The highest and lowest weaning age was 6.20 months and 4.0 months respectively. This is an agreement with the findings of Mulley *et al.* (1990). They reported that weaning generally takes place between 14-21 weeks. Flesch *et al.* (1999) showed that weaning age ranged between 12-20 weeks. Mulley and Flesch (2001) cited that deer must be weaned within 16 weeks of its age.

### Health Status of Experimental Animals

#### Deer Disease:

Yersiniosis and malignant catarrhal fever have not been recorded in fallow deer farmed in BN Zoo. Deer develop severe clinical illness very quickly in these serious diseases, which can rapidly progress to decumbency and death. The progress of these diseases is exacerbated by their nervous temperament, making these diseases potentially very dangerous.

During the examination period, no deer was sick in the Bangladesh National Zoo. The deer we identified did not see any disease in the year (table-9). But we tried very well to observe it clearly. Here we have learned from past history that due to the absence bacteria, viruses, parasites and nutrition there usually diseases. However, during the examination, we did not use any medicines in deer. Management of good deer does not cause in disease. So, the deer is a great gain animal.

**Table 7. Common parasites reported in deer**

<b>Internal parasites reported in deer</b>	
<b>Lungworm Nematodes</b>	<b>Large Intestine/Cecum</b>
Dictyocaulus	Chabertia
Elaphostrongylus	Oesophagostomum
Protostrongylus	Trichuri
<b>Gastroenteric Nematodes Abomasum</b>	<b>Other Nematodes</b>
Haemonchus	Elaeophora (circulatory system-arterial worm)
OstertagiaRinadia	Parelaphostrongylus (nervous systemmeningeal worm)
Skrjabinagia	<b>Trematodes (liver flukes)</b>
Spiculopteragia	FasciolaFascioloides
Trichostrongylus	Paramphistomum
<b>Small Intestine</b>	Dicrocoelium
Bunostomum	
CapillariaCooperia	
Nematodirus	
Strongyloides	
Trichostrongylus	

**Table 8. Common Deer Diseases**

Diseases reported in deer	
Bluetongue Brucellosis Clostridial infections: ▪blackleg, ▪Malignant Edema, ▪Enterotoximia Facial Eczema Louping ill Pneumonia	Foot Disorders: ▪foot scald ▪foot abscess ▪foot rot Kerato conjunctivitis Leptospirosis Ryegrass staggers Salmonellosis Tuberculosis

**Table 9. Experimental Deer Diseases**

Identify of Experimental Animals	Name of diseases														
	Deer 1	Deer 2	Deer 3	Deer 4	Deer 5	Deer 6	Deer 7	Deer 8	Deer 9	Deer 10	Deer 11	Deer 12	Deer 13	Deer 14	Deer 15
Calves (Fawn)	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×
Buck or Stags	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×
Doe or Hinds	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×
Pregnant Deer	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×
Weaning Animals	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×

**Herd management**

Deer rapidly adapts to the presence of man and machinery when they are enticed with palatable feeds. When deer are confined, they should not be over-crowded, so that a handler can move amongst them and carry out drenching, ear tagging, vaccination and pour-on dipping. Normally a supervisor of deer section maintains the following activities:

**Table 10. Schedule of day-to-day operations on spotted deer premises**

Approx. time (hrs)	Farm operations
7.00-9.00 A.M	<ul style="list-style-type: none"> <li>❖Check all fences surrounded by deer premises</li> <li>❖Count the total numbers of deer and adjust previous numbers</li> <li>❖Find out or observe if any mortality occurred</li> <li>❖Inspection within the area if any hazard happened at sight</li> <li>❖Clean out the dumping wastage material such as cons, bottle or any plastic product which was thrown by visitors</li> <li>❖Cleaning all the equipment's such as feeder, waterer</li> <li>❖Cleaning the entire feed supply zone</li> <li>❖Arrange the necessary vehicle to transport feed or other's</li> <li>❖Cleaning farm premises</li> </ul>
9.00-12.00 A.M	<ul style="list-style-type: none"> <li>❖Feeding of concentrate feed to deer herd</li> <li>❖Feeding of dry/green fodder</li> <li>❖Isolation of sick deer</li> <li>❖Treating sick deer</li> </ul>
12.00-3.00 P.M	<ul style="list-style-type: none"> <li>❖Lunch cum rest period for labors</li> </ul>
3.00-4.00 P.M	<ul style="list-style-type: none"> <li>❖Miscellaneous jobs of deer premises such as, periodical vaccinations, Repair of farm fences, Fittings and repair of equipment, weekly scrubbing and white washing of drinking water tank, attending to sale of deer and their transportation, periodical spraying of deer premises with suitable pesticides</li> </ul>
4.00 P.M-7.00 A.M	<ul style="list-style-type: none"> <li>❖A Night watchman on duty.</li> </ul>

For the determination of group size and composition, only visual observation techniques were used in this study. Data on group composition were recorded to age-sex classes, which were previously categorized.

## CONCLUSION

At Bangladesh National Zoo, the total amount of feed potted deer was 8.12 kg on fresh basis and 2.079 kg on DM. The percentage of crude protein and amount of met-abolishable energy in supplied feed were 13.63% and 14.38 MJ per day. There was variation of supplied green fodder, leafy vegetables and succulent vegetables with availability in the market and seasons. From the results it could be concluded that to keep balanced nutritive conditions along with productive wellbeing of the spotted deer in the zoo, recommended rations need to be fed with effective management systems to be build up permanently. Then it will largely contribute to the different zoos of Bangladesh.

## REFERENCES

- Ables. E.D. (Ed.) 1977. The Axis Deer in Texas, College station, Texas A & M Agricultural Experimental Station, USA.
- Adam, C.L. 1994. Feeding. In: Management and Disease of deer, 2nd Edn. (Ed. T. L. Alexander and D. Buxton). The Veterinary Deer Society. London, Great Britain. pp. 44-54.
- Adam, C.L. and Fletcher, T.J. 1994. Orphan calves. In: Management and Disease of deer, 2nd Edn. (Ed. T. L. Alexander and D. Buxton). The Veterinary Deer Society. London, Great Britain. pp. 55-58.
- AOAC. 2005. Official method of Analysis. 18th Edition, Association of Officiating Analytical Chemists, Washington DC, Method 935.14 and 992.24.
- ARC. 1980. The Nutrient Requirements of Ruminant Livestock. Commonwealth Agricultural Bureaux, England, UK.
- Azad, M.A.K., Hossain, M.M. and Bhuiyan, A.F.K.H. 2005. Asian Network for Scientific Information. Department of Animal Breeding & Genetics, Faculty of Animal Husbandry, Bangladesh Agricultural University, *Intl. J. Zoological Res.*, 1(1):48-52.
- Chapple, R.S., English, A.W. and Mulley, R.C. 2003. Characteristics of the estrous cycle and duration of gestation in chital hinds (*Axis asis*). *J. Reproduction and Fertility*, 98: 23-26.
- Denham, L.J. 1984. The nutrition of fanned deer. In: Deer Refresher Course for Veterinarians. Proc. No.72. The University of Sydney, Sydney, Pp. 662-691.
- Dey, T.K. 2007. Deer Population in the Bangladesh Sunderbans. Ad Communication. 60, Jamal Khan Road, Chittagong, Bangladesh. Pp1-83.
- Fennessy, P.F., Moore, G.H. and Corson, I.D. 1981. Energy requirements of red deer. Proc. N. Z. Soc. Anim. Prod. 41: 167-173.
- Feeroz, M.M. 2001. Species diversity and population density of non- human primates in north-east and south-east of Bangladesh. *Ecoprint* 8 (1): 53-57.
- French, C.E., Meewan, L.C., Magruder, N.D., Ingram, H.H. and R.W. Swift. 1956. Nutrient requirements for growth and antler development in the white-tailed deer. *J. Wildl. Manage.* 20:221.
- Geist, V. 1999. Deer of the world; their evolution, behaviour and ecology. Swann Hill Press, Shrewsbury, Gennany.
- Jean, B.T., Kim, K.H., Kim, U.S., Lee, S.M. and Kim, C.W. 1995. Deer farming survey on feed and feeding system in Chung-buk province. *Kor. J. Anim. Nutr. Feed*, 19: 235-241.
- Kay, R.N.B. and Staines, B.W. 1981. The nutrition of the red deer. In: Nutr. Abstr. Rev. (B) 51: 601-622.

- Kelly, R.W. and Moore, G.H. 1978. Reproductive performance in farmed red deer. In: D.R. Drew, Ed. *Advances in deer farming* 46 pp. Editorial services Ltd., Wellington.
- Khan, M.A.R. 1986. Wildlife in Bangladesh mangrove ecosystem. *J. Bombay nat. Hist. Soc*, 83:32-40.
- Mitchell, B. and D. Brown. 1974. The effects of age and body size on fertility in female red deer. XIth *Int. Congr. Game Biol.*, S. N. V. 1974. 13E: 89-98.
- Moon, S.H., Jeon, B.T., Lee, S.M., Kim, K.H. and Hudson, R.J. 2000. Seasonal comparison of voluntary intake and feeding behaviour in Korean spotted deer (*Cervus nippon*). *Asian-Aust. J. Anim. Sci*, 13 (10): 1394-1398.
- Mulley, R.C. 1984. Reproduction in fallow deer. In deer Refresher Course for Veterinarians. Proc. No. 72, The University of Sydney, Sydney, pp. 463-464.
- Mulley, R.C. and Flesch, J.S. 2001. Nutritional requirements for pregnant and lactation red and fallow deer. A Report for the Rural Industries Research and Development Corporation. R1RDC Publication No. 01/0095. Canberra, Australia.
- Mulley, R.C., English, A.W. and Kirby, A. 1990. The reproductive performance of farmed fallow deer in New South Wales. *Australian Vet. J.*, 67: 281-286.
- Mylrea, G.E., Mulley, R.C., English, A.W. and Evans, G. 2004. Reproductive cycles of farmed female chital deer (*Axis axis*). *CISRO Publishing-Reproduction, Fertility and Development*, 11(3): 167-174.
- Seidensticker, J., Christie, S. and Jackson, P. 1999. Introducing the tiger. In. *Riding the tiger: Tiger conservation in human-dominated landscapes*. Cambridge University Press, London, UK. pp. 1-3.
- Sharman, G.A.M. 1978. Red deer farming. *Ann Appl. Biol.* 88: 347- 350.
- Ullrey, D.R., Youatt, W.G., Johnson, H.E., Fay, L.D. and Bradley, B.L. 1967. Protein requirements of white-tailed deer fawns. *J. Wildl. Manage*, 3: 679-685.
- Verme, L.J. and Ullrey, D.E. 1972. Feeding and nutrition of deer. In: *Digestive Physiology and Nutrition of Ruminants*. Volume 3-Practical Nutrition (Ed. D.C. Church). The OSU. Bookstores, Inc., Corvallis, Oregon, USA. pp. 275-291.
- Vos, A.D. 1982. Deer farming guidelines on practical aspects Food and Agriculture Organization of the United Nations. Rome, Italy.
- Wilson, P.R. 1979. Nutrition and Reproduction of farmed deer. In: deer Refresher Course for Veterinarians. Proc. No.72, The University of Sydney, Sydney, pp. 463-464.

## ANTIDIABETIC EFFECT OF ALOE VERA IN ALLOXAN INDUCED DIABETIC RAT

M. S. Islam<sup>1</sup>, S. K. Sarkar<sup>2</sup>, M. M. Hossain<sup>3</sup>, M. A. Masum<sup>4</sup> and T. Rahman<sup>5</sup>

### ABSTRACT

Oral anti-diabetic agents have a number of serious adverse effects, thus, managing diabetes without any side effects is still a challenge. Therefore, the search for many effective and safer hypoglycemic agents has continued to be an important area of investigation. Besides, drug classically used for the treatment of diabetes (insulin, sulphonylureas, biguanides and thiazolidinediones), several species of plants have been described in the scientific and popular literatures having a hypoglycemic activity. The aim of this research was to investigate the effect of Aloe vera on blood glucose level in rats in diabetic condition. In this study a total of 150 rats (50 normal rats and 100 alloxan induced diabetic rats) were used for five trials. The rats were divided into three groups for each trial, each containing 10 individuals as follows: Group A is the control, Group B as diabetic control and Group C were diabetic rat received Aloe vera. Then alloxan injection was injected at a dose rate of 100 mg/kg body weight through intra-peritoneal route to each rat to induce diabetes in groups B and C. Aqueous extract of Aloe vera were fed by gavage at a dose of 300 mg/kg body weight daily for 21 days in group C. On the 10th day blood glucose level and the body weights were measured for the first time to ensure diabetic induction. Then all the rats of this group were kept for more 21 days for the treatment of diabetic condition. During experimental period, day 0, 7, 14 and 21 blood samples were collected from all groups and determined their blood sugar level using diabetic kit. After 3 weeks of treatment the blood glucose level significantly ( $P < 0.001$ ) reduced in the group C compared to B from  $260 \pm 11.53$  mg/dl to  $91.21 \pm 3.67$  mg/dl. On the other hand, the average body weight was increased significantly ( $P < 0.001$ ) from  $260 \pm 3.95$  gm to  $279.88 \pm 7.13$  gm in the group C compared to that of B. From our findings, it is concluded that Aloe vera can be used as anti-diabetogenic agent in food.

**Keywords:** Aloe vera, anti-diabetic, anti-hyperglycemic, rat

### INTRODUCTION

Diabetes is one of the major degenerative diseases in the world today. It is considered as one of the five leading causes of death in the world. Diabetes mellitus (DM) is characterized by elevated plasma glucose concentrations resulting from insufficient insulin, insulin resistance or both leading to metabolic abnormalities in carbohydrates, lipids and proteins (Hernandez-galicia *et al.*, 2002). It is a major risk factor for the development of cardiovascular disease. About 70-80% of deaths in diabetic patients are due to vascular disease. In particular, hyperglycemia, the primary clinical manifestation of diabetes, is thought to contribute to diabetic complications by altering vascular cellular metabolism, vascular matrix molecules and circulating lipoproteins. It can be hereditary and environmental which leads to metabolic abnormalities mainly characterized by hyperglycemia resulting from defects in insulin secretion, insulin action or both. Being a major degenerative disease, diabetes is found all over the world, and it is becoming the third most lethal disease of mankind and increasing rapidly (King and Herman 1998). It is the most common endocrine disorder, affecting 16 million individuals in the United States and as many as 200 million individuals worldwide (IDF, 2019).

For instance hyperglycemia increases diacylglycerol levels and activates protein kinase C activity in the aorta of streptozotocin induced diabetic rats (Inoguchi *et al.*, 1994) and dogs (Xia P *et al.*, 1994). Thickening of the basement membranes in renal glomeruli and peripheral capillaries has been observed in stz. induced diabetic rats (Olgemoller *et al.*, 1993) and hyperlipidemia is a feature of drug induced diabetes in rats (Still *et al.*, 1964) and rabbits (Nordestgaard *et al.*, 1988; Miller and Wilson, 1984).

Although, oral hypoglycemic agents and insulin are the main treatment of diabetes with prominent side effects and fail to significantly alter the course of diabetic complications. The common side effects

---

<sup>1</sup>Professor, <sup>2</sup>Assistant Professor, <sup>3</sup>Associate Professor, <sup>4</sup>Associate Professor, <sup>5</sup>MS. Student, Department of Anatomy, Histology and Physiology, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh

associated with oral hypoglycemic agents are hypoglycemia, weight gain, gastrointestinal disorders, peripheral edema and impaired liver function, in addition to the cost of treatment. Since natural remedies are somehow safer and more efficacious than pharmaceutically derived remedies. Complementary and alternative medicine involves the use of medicinal plant alternatives to mainstream treatment. A recent study has estimated that up to 30% of patients with DM use complementary and alternative medicine (Mukharje and Karati 2023; Villarruel-López *et al.*, 2018). According to the World Health Organization, more than 70% of the world's population must use traditional medicine to satisfy their principal health needs (Farnsworth *et al.*, 1985). A great number of medicinal plants used in the control of the DM have been reported (Baily and Day 1989). Aloe vera (synonym *A. barbadensis* Miller) (Liliaceae) is a fleshy plant filled with a clear viscous gel and it may exert its blood glucose lowering effect by preventing the death of  $\beta$ -cells and/or recovering the destroyed  $\beta$ -cells. It may also have initiated cell proliferation (Noor *et al.*, 2013). Five phytosterols in Aloe vera gel like lophenol, 24-methyl-lophenol, 24-ethyl-lophenol, cycloartanol, and 24-methylene-cycloartanol are believed to have anti-hyperglycemic effect in diabetes (Tanaka *et al.*, 2006). The aim of the present research proposal is to elucidate the possible antidiabetic activity of *Aloe vera* and its medicinal potency responsible for the protection of different organs.

## MATERIALS AND METHODS

This study was conducted in the Department of Anatomy, Histology and Physiology, Sher-e-Bangla Agricultural University, Dhaka to evaluate the efficacy of Aloe vera extract in diabetic rats.

### **Collection and acclimatization of rats:**

Total 150 mixed male albino rats (aged 2-3 months) were collected from Jahangirnagar University, Savar, Dhaka. For five experimental trials, all the rats were grouped into three each containing 50 rats. Each group of rats was housed at serene bottomed wire cages arranged in rows and kept in the animal house of this department. The animals were fed with pellet at a recommended dose of 100 gm/kg. Drinking water was supplied *ad libitum*. The rats were reared in this condition for a period of three weeks to acclimatize them prior to experimental uses.

### **Induction of diabetes:**

To induce diabetes mellitus, alloxan injection was given through intra-peritoneal route and this increased the blood glucose level ( $P < 0.05$ ) and at the same time body weight was decreased also. Single dose of alloxan administered intraperitoneally @100 mg/kg body weight (Junod *et al.*, 1996). In this experiment, polyuria, polydipsia and polyphagia after 24 hours of alloxan injection were observed. Rats with serum glucose level ranging between 150 mg/dl or above considered as hyperglycemic (Reeves *et al.*, 1993).

### **Experimental design:**

In that study, a total of 150 rats (50 normal rats and 100 alloxan induced diabetic rats) were used for five trials. The rats were divided into three groups for each trial, each containing 10 individuals (N=10) as follows:

**Group A:** Normal control group

**Group B:** Diabetic control group

**Group C:** Diabetic with Aloe vera treated group

After 18 hours of starvation, body weights and blood glucose were measured after acclimatization of rats. Then alloxan injection was injected at a dose rate of 100 mg/kg body weight through intra-peritoneal route to each rat to induce diabetes in groups B and C. All the groups of rats were reared under normal diet and water *ad libitum* from Day 1-10, on 10th day blood glucose level and the body weights gaining were measured for the first time to ensure diabetic induction. Then all the rats of this group were kept for more 21 days for the treatment of diabetic condition. During that period on Day 0, 7, 14, and 21st the body weight and blood glucose level were measured. Aqueous extract of Aloe vera fed by gavage at a dose of 300mg/kg body weight daily for 21 days in group C.

### Statistical analysis:

All data were expressed as mean  $\pm$  SEM (N=10) and differences among the groups of animals were compared using one-way analysis of variance (ANOVA) test. P-values less than 0.05 were considered significant.

## RESULTS AND DISCUSSION

To the best of our knowledge, this is the first study of anti-hyperglycemic activity of aqueous extract of Aloe vera in normal and alloxan diabetic rat. The study was carried out to evaluate the effects of Aloe vera on blood glucose and body weight. Changes in blood glucose level of rats were summarized in Table 1 describing that treatment of diabetic rats with Aloe vera induced a significant decrease in fasting blood glucose levels compared with diabetic untreated group. At the day of 21, aqueous solution of Aloe vera extract was significantly ( $P < 0.001$ ) found to reduce blood glucose levels in group C compared to B from  $260 \pm 11.53$  to  $91.21 \pm 3.67$  mg/dl.

**Table 1. Effect of Aloe vera on studied blood sugar in diabetic rats, compared to normal control** (Values are expressed in Mean  $\pm$  SEM, N= 10 for each group)

Grs	Day 0	Day 7	Day 14	Day 21
	ABSL (mg/dl)	ABSL (mg/dl)	ABSL (mg/dl)	ABSL (mg/dl)
A	95.50 $\pm$ 5.21 <sup>c</sup>	90.15 $\pm$ 5.4 <sup>c</sup>	91.89 $\pm$ 5.5 <sup>c</sup>	92.98 $\pm$ 9.5 <sup>c</sup>
B	188.00 $\pm$ 2.55 <sup>a***</sup>	193.15 $\pm$ 5.7 <sup>a***</sup>	210.68 $\pm$ 1.74 <sup>a***</sup>	260 $\pm$ 11.53 <sup>a***</sup>
C	94.55 $\pm$ 2.88 <sup>b***</sup>	96.5 $\pm$ 1.3 <sup>b***</sup>	94.1 $\pm$ 3.1 <sup>b***</sup>	91.21 $\pm$ 3.67 <sup>b***</sup>

a\*\*\*Significantly different from control at  $P < 0.001$ , b\*\*\*Significantly different from DM (Diabetic mellitus) at  $P < 0.001$

**Legends:** Grs: Group, Group A: Normal Control; Group B: Diabetic control; Group C: Alloxan + Aloe vera treated; ABSL: Average blood sugar level

Aloe vera treatment induced a significant increase in body weight compared with diabetic untreated group (Table 2). The average body weight was increased significantly ( $P < 0.001$ ) from  $260 \pm 3.95$  to  $279.88 \pm 7.13$  gm in group C compared to B after 3 weeks treatment.

**Table 2. Effect of Aloe vera on studied body weight in diabetic rats, compared to normal control** (Values are expressed in Mean  $\pm$  SEM, N= 10 for each group)

Grs	Day 0	Day 7	Day 14	Day 21
	ABW (gm)	ABW (gm)	ABW (gm)	ABW (gm)
A	243.24 $\pm$ 4.15 <sup>c</sup>	246.5 $\pm$ 5.50 <sup>c</sup>	251.75 $\pm$ 5.96 <sup>c</sup>	265.51 $\pm$ 1.5 <sup>c</sup>
B	271.55 $\pm$ 2.11 <sup>a***</sup>	270.44 $\pm$ 6.51 <sup>a***</sup>	264.5 $\pm$ 4.21 <sup>a***</sup>	260 $\pm$ 3.95 <sup>a***</sup>
C	266.82 $\pm$ 8.44 <sup>b***</sup>	271.43 $\pm$ 7.5 <sup>b***</sup>	275.37 $\pm$ 9.63 <sup>b***</sup>	279.88 $\pm$ 7.13 <sup>b***</sup>

a\*\*\*Significantly different from control at  $P < 0.001$ , b\*\*\*Significantly different from DM (Diabetic mellitus) at  $P < 0.001$

**Legends:** Grs: Group, Group A: Normal Control; Group B: Diabetic control; Group C: Alloxan + Aloe vera treated; ABW: Average body weight

Diabetes mellitus is probably the fastest growing metabolic disease in the world. As the knowledge of multifactorial nature of this disease increases so does the need for more challenging and appropriate therapies (King *et al.*, 1998; Dans *et al.*, 2007). Alloxan is known for selective pancreatic islet  $\beta$ -cell cytotoxicity and has been extensively used to induce diabetes mellitus in animals (Fernandes *et al.*, 2007). Generalized increase in the level of blood glucose during diabetes have been consistently

reported both in animal models and humans especially those suffering from insulin dependent diabetes mellitus (Mathew *et al.*, 1973; Lorenzati *et al.*, 2010).

In the present study we found that Aloe vera extract reduced the blood glucose in diabetic rats. Regarding serum glucose level, treatment of diabetic rats with Aloe vera caused significant decreases in fasting and post-prandial serum glucose levels as compared to the diabetic untreated group. There are two possible explanations for this finding. First, Aloe vera may exert its effect by preventing the death of  $\beta$ -cells or it may permit recovery of partially destroyed  $\beta$ -cells. Like *Momordica charantia* (Akhter *at al.*, 2018a) Aloe vera may also have initiated cell proliferation. These results are in accordance with the previous findings in Aloe vera (Noor *et al.*, 2013; Govindarajan *et al.*, 2021), Neem (Chattopadhyay *et al.*, 1987), Bitter melon (Akhter *et al.*, 2018a), Garlic (Akhter *et al.*, 2018b), Drumstick (Villarruel-López *et al.*, 2018) and Telakucha (Sarkar *et al.*, 2020).

## CONCLUSION

In conclusion, the present study calls attention to the therapeutic use of A. vera in DM. The results of the current study demonstrated that Aloe vera was effective in lowering hyperglycemic activity in alloxan induced rats. Further studies are necessary in terms of different doses and longer duration of A. vera to be considered as better therapeutic option for DM.

### Conflict of interest:

The authors declared there is no conflict of interest.

## REFERENCES

- Akhter, R., Rasel, I.H. and Islam, M.S. 2018a. Antidiabetic effect of bitter melon/Kerala (*Momordica charantia*) in alloxan induced diabetic rat. *Res. Agric. Livest. Fish.*, 5(3): 373-379.
- Akhter, R., Rasel, I.H. and Islam, M.S. 2018b. Effect of bitter melon and garlic on blood glucose level and blood cholesterol level in rats in diabetic condition. *Res. Agric. Livest. Fish.*, 5(3): 359-363.
- Bailey, C.J. and Day, C. 1989. Traditional Plant Medicines as Treatments for Diabetes. *Diabetes Care*, 12: 553-564.
- Chattopadhyay, R.R., Chattopadhyay, R.N., Nandy, A.K., Poddar, G. and Maitra, S.K. 1987. Preliminary report on antihyperglycemic effect of a fraction of fresh leaves of *Azadirachata indica* (Beng. Neem). *Bull. Calcutta School Trop. Med.* 35: 29-35.
- Dans, A.M., Villarruz, M.V., Jimeno, C.A., Javelosa, M.A., Chua, J., Bautista, R. and Velez, G.G. 2007. The effect of *Momordica charantia* capsule preparation on glycemic control in type 2 diabetes mellitus needs further studies. *Medizinische Monatsschrift für Pharmazeuten.*, 30 (4): 131-137.
- Fernandes, N.C., Lagishetty, C.V., Panda, V.S. and Naik, S.R. 2007. An experimental evaluation of the antidiabetic and antilipidemic properties of a standardized *Momordica charantia* fruit extract. *BMC Complementary Alternative Medicine*, 7: 29.
- Farnsworth, N.R., Akerele, O., Bingel, A.S., Soejarto, D.D. and Guo, Z. 1985. Medicinal plants in therapy. *Bulletin of the World Health Organization*, 63(6): 965-981.
- Govindarajan, S., Babu, S., Vijayalakshmi, M., Manohar, P. and Noor, A. 2021. Aloe vera carbohydrates regulate glucose metabolism through improved glycogen synthesis and downregulation of hepatic gluconeogenesis in diabetic rats. *J. Ethnopharmacol.*, 281:114556.
- Hernandez-Galicia, E., Aguilar-Contreras, A., Aguilar-Santamaria, L., Roman-Ramos, R., Chavez-Miranda, A.A., Garcia-Vega, L.M., Flores-Saenz, J.L. and Alarcon-Aguilar, F.J. 2002. Studies on hypoglycemic activity of Mexican medicinal plants. *Proc. Western Pharmacol. Society*, 45: 118-124.
- IDF (International Diabetic Federation) Diabetes Atlas, 2019. Ninth edn. <https://www.diabetesatlas.org/en/>.

- Inoguchi, T., Xia, P., Kunisaki, M., Higashi, S., Feener, E.P. and King G.L. 1994. Insulin's effect on protein kinase C and diacylglycerol induced by diabetes and glucose in vascular tissues. *American J. Physiology.*, 267: E369-E379.
- Junod, A., Lambert, A.E., Stauffer, W. and Renold, A.E. 1996. Diabetogenic action of streptozotocin. Relationship of dose to metabolic response., *J. Clinical Invest.*, 48: 2129-2139.
- King, H. Aubert, R. E. and Herman, W.H. 1998. Global burden of diabetes, 1995-2025: prevalence, numerical estimates and projections., *Diabetes Care.*, 21: 1414-1431.
- Lorenzati, B., Zucco, C., Miglietta, S., Lamberti, F. and Bruno, G. 2010. Oral Hypoglycemic Drugs: Pathophysiological Basis of Their Mechanism of Action. *Pharmaceuticals (Basel, Switzerland).*, 3(9): 3005-3020.
- Mathew, P.T. and Augusti, K.T. 1973. Studies on the effect of allicin (diallyl disulphide-oxide) on alloxan diabetes I. Hypoglycaemic action and enhancement of serum insulin effect and glycogen synthesis. *Indian J. Biochem. and Biophysics.*, 10: 209-212.
- Miller, R.A. and Wilson, R.B. 1984. Atherosclerosis and myocardial ischemic lesions in alloxan-diabetic rabbits fed a low cholesterol diet. *Arteriosclerosis.*, 4(6): 586-591.
- Mukherje, S. and Karati, D. 2023. Exploring the Phytochemistry, Pharmacognostic Properties, and Pharmacological Activities of Medically Important Plant *Momordica Charantia*. *Pharmacological Research-Modern Chinese Medicine.*, 100226.
- Nordestgaard, B.G.S., Stender, S. and Kjeldsen, K. 1988. Reduced atherogenesis in cholesterol-fed diabetic rabbits. Giant lipoproteins do not enter the arterial wall. *Arteriosclerosis.*, 8: 421-428.
- Noor, A., Bansal, V.S. and Vijayalakshmi, M.A. 2013. Current update on anti-diabetic biomolecules from key traditional Indian medicinal plants. *Current Science.*, 104(6): 721-727.
- Olgemoller, B. and Schleicher, E. 1993. Alterations of glomerular matrix proteins in the pathogenesis of diabetic nephropathy. *Clinical Investigation.*, 71: S13 – S19.
- Reeves, P.G., Nielsen, F.H. and Fahey, G.C. 1993. AIN-93 purified diets for laboratory rodents: final report of the American institute of nutrition Ad Hoc writing committee on reformulation of the AIN-76 rodent diet. *J. Nutrition.*, 12: 23-56.
- Sarkar, S.K., Uddin, M., Hossain, M.M., Masum, M.A. and Islam, M.S. 2020. Hematobiochemical Effects of *Telakucha (Coccinia Indica)* in Alloxan Induced Diabetic Rats. *Research in Agriculture Livestock and Fisheries.*, 7(3): 431-438.
- Still, W.J.S., Martin, J.M. and Gregor, W.H. 1964. The effect of alloxan diabetes on experimental atherosclerosis in the rat. *Experimental Molecular Pathology.*, 3:141-147.
- Tanaka, M., Misawa, E., Ito, Y., Habara, N., Nomaguchi, K., Yamada, M., Toida, T., Hayasawa, H., Takase, M., Inagaki, M. and Higuchi, R. 2006. Identification of five phytosterols from *Aloe vera* gel as anti-diabetic compounds. *Biological and Pharmaceutical Bulletin.*, 29 (7): 1418-22.
- Villarruel-López, A., López-de la Mora, D.A., Vázquez-Paulino, O.D., Puebla-Mora, A.G., Torres-Vitela, M.R., Guerrero-Quiroz, L.A. and Nuño, K. 2018. Effect of *Moringa oleifera* consumption on diabetic rats. *BMC complementary and alternative medicine.*, 18(1): 127.
- Xia, P., Inoguchi, T., Kern, T.S., Engerman, R.L., Oates, P.J. and King, G.L. 1994. Characterization of the mechanism for the chronic activation of diacylglycerol-protein kinase C pathway in diabetes and hypergalactosemia. *Diabetes.*, 43(9): 1122-1129.

## INCIDENCE OF SURGICAL CASES AT MIRZAPUR UPAZILA IN TANGAIL DISTRICT OF BANGLADESH

M.M. Hossain<sup>1</sup>, N. Nazmunahar<sup>2</sup>, R. Akter<sup>3</sup> and M. Islam<sup>4</sup>

### ABSTRACT

This study was to determine the incidence of most common surgical cases in cattle, calves and goats at Mirzapur Upzilla Veterinary Hospital, Tangail during the period from January 2021 to December 2022. The total number of recorded cases were 900 (Cattle=328, Calves=472 and Goat=472) during the study period. The incidence rate of surgical affections in recorded is castration (23.77%), abscess (12.80%), wound (10.97%), fracture (9.76%), Navel ill (8.54%), Vaginal prolapsed (8.54%), Hernia (7.93%), Myiasis (6.71%), Atresia ani (5.49%), ovariectomy (3.05%) and upward patellar fixation (2.44%). In calves, the rate was umbilical hernia (40%), umbilical abscess (15%), atresia ani(12%), naval myiasis and abscess (8%), intestinal prolapsed and fracture of jaw (2%). In small ruminants especially in goat the incidence rate was coenurosis (19.07%), subcutaneous cyst (13.98%), castration (13.56%), abscess (11.86%), fracture (8.90%), myiasis (8.47%), wound (6.36%), atresia ani (5.51%), urolithiasis (5.51%), hernia (4.24%) and ovariectomy (2.54%). The incidence rate was higher in goat (52.5%) , cattle (36.4%) and in calves (11.1%) .Moreover, the incidence rate was higher in male compared with female. In addition, the incidence rate was higher in summer (36.2%) followed by rainy season (32.01%) and winter season (31.2%). Myiasis and arthritis were the most common cases in summer whereas urolithiasis was more common in winter. However, the study might be helpful to compare the incidence rate of surgical cases in other areas of Bangladesh and take necessary action to minimize the incidence of surgical cases.

**Key words:** surgical diseases, animals, surgery, prevalence, Mirzapur.

### INTRODUCTION

Bangladesh is an agricultural country where more or less 80% of the populations live on agriculture and livestock farming. Domestic animals especially goats and cattle are an important part of livestock and plays a crucial role in the conventional economy of Bangladesh (Alam *et al.*, 2015). Among the various shortfall to goats and cattle production, diseases are the most important hindrance, which tarnished the productivity of these animals (Sarker *et al.*, 1999). Farmers are interested in rearing goats and cattle as because they are very docile, the availability of feed, good fertility rate and multiple kidding efficiency. Economy of Bangladesh is vibrant due to sustainable agricultural and livestock production. There are about 14.8 million goats and 25.7 million cattle in Bangladesh (DLS, 2019-2020). The contribution of Livestock in Gross Domestic Product (GDP) is about 3.47% in Bangladesh (BBS, 2019).The faulty management practices of animals as well as climatic condition of Bangladesh are also encouraging for the incidence of various surgical affections. The occurrence of diseases and disease conditions varies with the species, ages, sex of the animals and time of the year (Hoaque and Samad, 1996; Samad, 2001; Islam *et al.*, 2013). Most of the animals are weak, emaciated and non-satisfactory productive performance mainly due to malnutrition and diseases. Veterinary hospital is an ideal and reliable source of information about animal diseases and their solution. People from the neighboring areas bring their sick animals to the veterinary hospital every day. Analysis of the case record gives a comprehensive idea about the disease problems at local areas. The importance of surgery is to save or prolong the life of an animal and to hasten the recovery from an injury (Sarker *et al.*, 2014). Additionally, surgery is performed to stop the progression of a disease process, for aesthetic purposes, to cure abnormalities or deformities, to replace a part with an artificial one, for financial reasons, or to make an animal more sociable. Surgery is crucial to aid in the diagnosis of a potential pathological process as well as for research job investigation. For the treatment of both large and small animals, including birds, developed countries have excellent modern surgical facilities. However, these opportunities are less common in

<sup>1</sup>Associate Professor, <sup>2&3</sup>MS student, Dept. of Anatomy, Histology and Physiology <sup>4</sup>Associate Professor, Dept. of Microbiology and Parasitology, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh

developing countries due to restricted access to operating rooms, a lack of specialized surgical instruments, and a lack of anesthetic devices needed for both anesthesia induction and maintenance. Despite challenges, veterinarians frequently perform minor surgical procedures on goats and cattle in the field in our country. A few works on surgical affections in goats and cattle were done in Bangladesh Agricultural University Veterinary Clinic (Rahman *et al.*, 1972; Hossain *et al.*, 1986; Das and Hashim, 1996; Samad, 2001; Samad *et al.*, 2002), Haluaghat Upazila Veterinary Hospital, Mymensingh (Sarker *et al.*, 1999) and Dairy Cooperatives in Pabna district (Pharo, 1987), Ulipur Upazila Veterinary Hospital, Kurigram (Kabir *et al.*, 2010), Chandanaish Upazila of Chittagong District, Bangladesh (Pallab *et al.*, 2012) and Patuakhali Science and Technology University Veterinary Clinic (Rahman *et al.*, 2012). Therefore, this research work helps to determine the incidence of most common surgical cases recorded in Upazila Livestock hospital at Mirzapur in Tangail district of Bangladesh during the period of from January 2021 to December 2021.

## MATERIALS AND METHODS

### 2.1: Research Area and Duration

This study was performed at the Upazila Livestock Office and Veterinary Hospital, Mirzapur, Tangail from the period of January 2021 to December 2022. A total 900 cases were examined during the study period out of them 472 were goats, 328 were cattle and 100 were calves.

### 2.2: Data Collection

The data were collected by prescribed questionnaires through owner's complaints as well as visual observations.

### 2.3: Determination of Age of Animals

The age of individual animal was determined by the examination of teeth and interviewing the owner's of the animals.

### 2.4: Methods of diagnosis of disease and disorder

The presumptive diagnosis was performed based on history from Owner's, physical examination, clinical signs and clinical examination of animals.

### 2.5: Study Design

Surgical cases categorized into three groups such as cattle, calves and goats. The recorded cases were further characterized based on the species, age, sex, breed, seasons etc.

### 2.2. Physical examination

Examination of different parts and systems of individual sick animals were performed through palpation, percussion, auscultation, needle exploration, extension and flexion of limbs, use of mouth gag, local anesthesia and walking of animals as per methods described by Kelly (1979) and Samad *et al.* (1988).

### 2.3. Clinical examination

Clinical examinations of 472 goats, 328 cattle and 100 calves of different ages were conducted based on history of the diseases, owner's complaint and symptoms to diagnose. History of individual case provides a guideline for examination of the animals. On the basis of the merit of individual case, general clinical examination were conducted and owner's complaint, symptoms and techniques such as microscopic examination, common laboratory techniques used by Rosenberger (1979) and Samad *et al.* (1988) for the diagnosis of the diseases. The temperature, pulse, and respiratory rate from individual sick animal were recorded.

## RESULTS AND DISCUSSION

### 3.1. Incidence of various surgical cases in cattle, calves and goats

The detail result on the occurrence of surgical cases in cattle, calves and goats were shown in Table 1, 2 and 3. During the study period, a total 328 numbers of cattle, 100 numbers of calves and 472 numbers goats were examined for surgical purpose to determine the prevalence of surgical cases in cattle, calves and goats as well as to find out the effect of some factors (age, sex, and breed) on that case.

**Table 1. Incidence of surgical cases in cattle**

Surgical cases	Incidence rate (%)
Castration	23.77 (78)
Abscess	12.80 (42)
Wound	10.97 (36)
Fracture	9.76 (32)
Navel ill	8.54 (28)
Vaginal prolapse	8.54 (28)
Hernia	7.93 (26)
Myiasis	6.71 (22)
Atresia ani	5.49 (18)
Ovariohysterectomy	3.05 (10)
Upward patellar fixation	2.44 (8)
Total	328

**Table 2. Incidence of surgical cases in calves**

Surgical cases	Incidence rate (%)
Umbilical hernia	46(40)
Umbilical abscess	18(15)
Atresia ani	12(12)
Naval myiasis	10(8)
Naval abscess	10(8)
Intestinal prolapse	2(2)
Fracture of jaw	2(2)
Total	100

**Table 3. Incidence of surgical cases in goats**

Surgical cases	Incidence rate (%)
Gid disease	90 (19.07)
Subcutaneous cyst	66(13.98)
Castration	64(13.56)
Abscess	56 (11.86)
Fracture	42 (8.90)
Myiasis	40(8.47)
Wound	30 (6.36)
Atresia ani	26 (5.51)
Urolithiasis	26 (5.51)
Hernia	20 (4.24)
Ovariohysterectomy	12 (2.54)
Total	472

The most prevalent surgical cases in cattle at Mirzapurr Upazila Veterinary Hospital from highest to lowest is castration (23.77%), abscess (12.80%), wound (10.97%), fracture (9.76%), Navel ill (8.54%), Vaginal prolapse (8.54%), Hernia (7.93%), Myiasis (6.71%), Atresia ani (5.49%), Ovariohysterectomy (3.05%) and Upward patellar fixation (2.44%). In calves the most common surgical cases were umbilical hernia (40%), umbilical abscess (15%), atresia ani(12%), naval myiasis and abscess (8%), intestinal prolapsed and fracture of jaw (2%). In goat, most common surgical cases from highest to lowest is coenurosis (19.07%), subcutaneous cyst (13.98%), castration (13.56%), abscess (11.86%),

fracture (8.90%), myiasis (8.47%), wound (6.36%), atresia ani (5.51%), urolithiasis (5.51%) hernia (4.24%) and ovariohysterectomy (2.54%).

### 3.2. Effects of various factors on the incidence of surgical cases in cattle

#### 3.2.1. Effect of age and sex

The effect of age and sex on the occurrence of surgical diseases in cattle are shown in Table 4. In male animals, the highest incidence (74.6%) of the disease occurred in cattle of 1 year plus old. Like male cattle, the incidence rate (25.4%) of the disease in the female was also recorded in one year plus age group. The highest incidences (65.3%) was observed in cattle under one year of age, while the lowest incidence rate (34.7%), was observed in same age group.

**Table 4. Effect of age and sex on the incidence of surgical cases in cattle**

Age	Male	Female	Total (%)
<1 year (n=188)	122(64.9)	66(35.7)	188(57.1)
1 year plus (n=140)	102(72.9)	38(27.1)	140(42.8)
Total	224(68.9)	104(31.1)	328(100)

#### 3.2.2. Effect of breed

The effect of breed on the occurrence of surgical cases in cattle is shown in Table 5. Out of 328 affected cattle, 50 were indigenous and 278 were crossbred, the incidence of surgical affections in indigenous male (64%), female (36%), crossbred male (69.5%), and in female (30.5%). From this observation, it is clear that crossbred cattle are more susceptible for surgical affections compared to indigenous cattle. This observation about age and sex is agreeable to earlier reports (Rahman *et al.*, 2001).

**Table 5. Effect of breed on the incidence of surgical cases in cattle**

Breed	Male (No. %)	Female (No. %)	Total (No. %)
Indigenous (n=50)	32(64)	18(36)	50(15.1)
Cross (n=278)	192(69.1)	86(30.9)	278(84.9)
Total	224(68.3)	104(31.7)	328(100)

### 3.3. Effects of different variables on the incidence of surgical affections in goat

#### 3.3.1. Effect of age, sex and breed

The effect of age and sex on the incidence of surgical cases in goat is presented in Table 6 and Table 7. In male animals, the highest incidence (67.4%) of the disease occurred in goat of 1-24 months of age, and the low incidence rate (32.6%) of the disease in the female was recorded in 1-24 months age group. In an average more surgical affections occurred in 6-24 months age (54.7%). In goats gid disease and subcutaneous cyst is more prevalent than other surgical cases because the parasitic infection is more in Mirzapur Veterinary Hospital like Bangladesh (Rahman *et al.*, 1983). The present study is closely related with this information.

**Table 6. Effect of age and sex on the incidence of surgical cases in goats**

Age	Male	Female	Total (%)
<1 month (n=86)	60(69.8)	26(30.2)	86(18.2)
1-6 months (n=128)	90(70.3)	38(29.7)	128(27.1)
6-24 months (n=258)	110(42.6)	148(57.4)	258(54.7)
Total	260(67.4)	212(32.6)	472(100)

**Table 7. Effect of breed on the incidence of surgical cases in Goats**

Breed	Male (No. %)	Female (No. %)	Total (No. %)
Local (n=400)	220(55)	180(45)	400(84.7)
Jamunapari (n=72)	40(55.6)	32(44.4)	72(15.3)
Total	260(78)	212(22)	472(100)

### 3.4. Effects of different variables on the occurrence of surgical affections in calves

#### 3.4.1. Effect of age, sex and breed

The effect of age and sex on the incidence of surgical cases in calves is presented in Table 8 and Table 9. In male animals, the highest incidence (68%) of the disease occurred in calves of 1-3 months of age, and the low incidence rate (32%) of the disease in the female was recorded in 1-3 months age group.

**Table 8. Effect of age and sex on the incidence of surgical cases in calves**

Age	Male	Female	Total (%)
<1 month (n=64)	44(68.8)	20(31.3)	64(64)
1-3months (n=36)	24(66.7)	12(33.3)	36(36)
Total	68( 68)	32(32)	100(100)

**Table 9. Effect of breed on the incidence of surgical cases in calves**

Breed	Male (%)	Female (%)	Total (%)
Indigenous (n=20)	14(70)	6(30)	20(20)
Cross (n=80)	64(80)	16(20)	80(80)
Total	78(78)	22(22)	100(100)

**Table 10. Effect of season on surgical affections in goats at Mirzapur Upazila, Tangail**

Diseases	Summer	Winter	Prevalence rate (%)
Gid disease	60	30	19.07 (90)
Subcutaneous cyst	46	20	13.98 (66)
Castration	24	40	13.56 (64)
Abscess	40	16	11.86 (56)
Fracture	22	20	8.90 (42)
Myiasis	30	10	8.47 (40)
Wound	14	16	6.36 (30)
Atresia ani	14	12	5.51 (26)
Urolithiasis	10	16	5.51 (26)
Hernia	8	12	4.24 (20)
Ovariohysterectomy	4	8	2.54 (12)
Total	272	200	472

**Table 11. Effect of seasons on surgical cases in cattle at MirzapurUpazila, Tangail**

Diseases	Summer	Winter	Incidence rate (%)
Castration	30	48	23.77 (78)
Abscess	32	10	12.80 (42)
Wound	16	20	10.97 (36)
Fracture	12	20	9.76 (32)
Navel ill	22	6	8.54 (28)
Vaginal prolapse	12	16	8.54 (28)
Hernia	14	12	7.93 (26)
Myiasis	18	4	6.71 (22)
Atresia ani	10	8	5.49 (18)
Ovariohysterectomy	4	6	3.05 (10)
Upward patellar fixation	2	6	2.44 (8)
Total	172	156	328

**Table 12. Effect of seasons on surgical cases in calves at Mirzapur Upazila, Tangail**

Diseases	Summer	Winter	Incidence rate (%)
Umbilical hernia	34	12	40 (46)
Umbilical abscess	14	4	15 (18)
Atresia ani	8	4	12 (12)
Myiasis	8	2	18 (10)
Naval abscess	8	2	8 (10)
Intestinal prolapse	2	0	2 (2)
Fracture of jaw	2	0	2(2)
Total	76	24	100

## CONCLUSION

It might be concluded that the abscess in cattle, umbilical hernia in calves and gid disease in goats were most prevalent at Mirzapur Upazila Veterinary Hospital. Most Surgical cases occurred mostly in cattle of less than one-year age group, less than one-month age group of calves and 6-24 months age group in goats. The incidence of surgical cases was more common in male cattle, calves and goats than that in females. The higher incidence of surgical cases was encountered in the crossbred cattle in contrast to indigenous cattle. In goat, gid disease was the most incidence rate at Mirzapur Upazila Veterinary Hospital of Tangail, District.

## REFERENCES

- Alam, M.A., Amin, M.R., Paul, T.K., Saha, T.K., Rahman, M.K. and Rizon, M.K. 2015. Prevalence of clinical diseases and disorders of goats at Upazila Livestock Development Center, Kapasia, Gazipur. *Asian J. Medical and Biolog. Res.*, 1(1): 47-52.
- BBS. 2019. Statistical year book on Bangladesh. Bangladesh Bureau Statistics, Ministry of Planning, Dhaka.
- Das, B.C. and Hashim, M.A. 1996. Studies on surgical affections in calves. *Bangladesh Vet. J.*, 30: 53-57.
- DLS. 2019-20. Annual Report of Directorate of Livestock Services, Bangladesh.
- Hoque, M.S. and Samad, M.A. 1996. Prevalence of clinical diseases in dairy cross-bred cows and calves in the urban areas in Dhaka. *Bangladesh Vet. J.*, 30: 118-129.
- Hoque, M.S. and Samad, M.A. 1997. Present status of clinical diseases of goats in the urban areas in Dhaka. *Bangladesh Vet. J.*, 31: 35-40.
- Hossain, M.A., Shaidullah, M. and Ali, M.A. 1986. A report on surgical diseases and reproductive disorders recorded at the Veterinary Hospital of Bangladesh Agricultural University, Mymensingh. *Bangladesh Vet. J.*, 20: 1-5.
- Islam, M., Khan, M., Kader, H., Begum, M. and Asgar, M. 2013. Prevalence of PPR of goat and their response to antibiotic treatment at Mirzaganj Upazila of Patuakhali District. *J. Environ. Sci. and Natural Resour.*, 5:181-184.
- Kabir, M.H., Reza, M.A., Razi, K.M.A., Parvez, M.M., Bag, M.A.S. and Mahfuz, S.U. 2010. A report on clinical prevalence of diseases and disorders in cattle and goat at the Upazila Veterinary Hospital, Ulipur, Kurigram. *Intl. J. Biolo. Res.*, 2: 17-23.
- Kelly, W.R. 1979. Veterinary clinical diagnosis, second edn.1974. Bailliere, Tandall and Cassel Ltd. London. 39-102.
- Pallab, M.S., Ullah, S.M., Uddin, M.M. and Miazi, O.F. 2012. A cross sectional study of several diseases in cattle at Chandanaish Upazila of Chittagong district. *Sci. J. Vet. Advan.*, 1: 28-32.

- Pharo, H.J. 1987. Analysis of clinical case records from dairy co-operatives in Bangladesh. *Trop. Anim. Health Prod.*, 19: 136-142.
- Rahman, M.A., Islam, M.A., Rahman, M.A., Talukder, A.K., Parvin, M.S. and Islam, M.T. 2012. Clinical diseases of ruminants recorded at the Patuakhali Science and Technology University Veterinary Clinic. *Bangladesh J. Vet. Medicine*, 10(1&2): 63-73.
- Rahman, M.A., Ali, K.M. and Rahman, A. 1972. Incidence of diseases of cattle in Mymensingh. *Bangladesh Vet. J.*, 6: 25-30.
- Rahman, A., Samad, M.A. and Huq, M.M. 1983. Clinico-pathological studies on psoroptic mange in a goat. *Bangladesh Vet. J.*, 13: 53-55.
- Rosenberger, G. 1979. Clinical Examination of Cattle. 2nd edn., Verlag Poul Parey, Germany.
- Samad, M.A. 2001. Observations of clinical diseases in ruminants at the Bangladesh Agricultural University. Veterinary Clinic. *Bangladesh Vet. J.*, 35: 93-120.
- Samad, M.A. 2002. Clinical surgery In: Veterinary Practitioner's Guide. LEP publication, Dhaka. 399-412.
- Samad, M.A., Bari, A.S.M. and Bashir, S.A. 1988. Gross and histopathological studies on bovine babesiosis in Bangladesh. *Indian J. Animal Sci.* 58: 926-928.
- Sarker, N.U., Samaddar, K., Haq, M.M. and Rahman, M.M. 2014. Surgical affections of cattle in the milk-shed areas of Bangladesh. *The Bangladesh Veterinarian*, 31: 38 – 45.
- Sarker, M.A.S., Hashim, M.A., Rahman, M.B. and Begum, H. 1999. Studies on bovine lymphadenitis syndrome. *Bangladesh Veterinarian*, 10: 6-8.