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EFFECT OF DIFFERENT COMBINATION OF BUFFALO DUNG WITH SUGARCANE BAGASSE ON REPRODUCTION AND GROWTH OF *LAMPITO MAURITII* KINBERG DURING VERMICOMPOSTING

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ABSTRACT

The earthworm *Lampito mauritii* have been known to useful in management of different organic wastes and for production of organic fertilizer. In the present work sugarcane bagasse (SCB) *i.e.*, waste of sugar industry, was fed to *Lampito mauritii* with buffalo dung (BD) support as feed material at various ratio i.e. BD+SCB (1:1), BD+SCB (2:1), BD+SCB (1:2), BD+SC (3:1), BD+SCB (1:3) as well as BD alone. The significant initiation of clitellum development, initiation of cocoon production, period of cocoon production, incubation period, cocoon/worm, hatchling/cocoon, hatchling/worm were observed in all the combinations of feed material of buffalo dung with sugarcane bagasse. In binary combinations of buffalo dung with sugarcane bagasse in 1:1 ratio, development of clitellum was significantly faster and showed significant period of initiation of cocoon. The result demonstrated that significantly high number of cocoon productions was observed in combination of BD+SCB in 1:1 ratio (23.67±0.31SE cocoon/worm) followed by BD+SCB in 2:1 ratio (21.67±0.48SE cocoon/worm). The higher number of hatchlings were observed per worm produced in BD+SCB in 1:1 ratio (51.36 hatchling/worm) and showed maximum growth rate (9.15±0.45SE mg /worm/day) of *Lampito mauritii*. The result observed from the present study indicate that the combination of buffalo dung with sugarcane bagasse in 1:1 ratio is a best feed material for better growth and development of *Lampito mauritii* and thus play a major role in industrial waste management.

Keywords: *Buffalo Dung, Growth and Reproduction, Lampito mauritii, Sugarcane Bagasse, Vermicomposting*

INTRODUCTION

In the Indian economy, sugar mills play an important role and contribute significantly to export earnings. India is the world's second largest producer of sugar and its byproducts, among the 83 sugarcane-producing countries (Rao, 2005). Duque *et al.* (2015) reported that the globe produces 175.1 million metric tonnes of sugarcane, with each tonne yielding 280 kg of SCB (Restrepo-Serna *et al.*, 2018). There are numerous byproducts created throughout the manufacturing process, including bagasse, pressmud, and sugar cane residue. During the sugarcane juice extraction process, fibrous waste known as bagasse (B) is produced. Bagasse is a substance that contains between 30 - 40 percent pith fibres. It usually contains high levels of lignin (21%), cellulose (44%) and hemicellulose (28%), ashes (5%) and extractive (2%) (Karp *et al.*, 2013). Sugarcane bagasse has a higher water retention capacity due to its lignified surface area. Bagasse can change soil physico-chemical parameters, improve soil quality, and boost crop output (Xu *et al.*, 2021). Dumping bagasse is an unattractive process because to the large land need and pollution issues. One issue facing people today is waste management, often referred to as waste disposal

(Fataei and Seiied Safavian, 2017; Amirfazli *et al*., 2019; Samadi Khadem *et al*., 2020; Singh and Singh, 2023d). Technology for vermicomposting offers two main benefits; it reduces the generation of organic waste and, on the other hand, turns it into valuable product (Daghestani and Niknam, 2019). Vermicomposting is an achievable method that may be used to get rid of lignocellulosic waste and make useful products. On the basis of reports, cattle dung makes the best primary substrate for vermicomposting (Doan *et al.,* 2013).

Cattle dung can be used as the only substrate for vermicomposting, or it can be combined with other organic solid wastes as a bulking agent to create more stabilized, nutrient-rich vermicompost, or it can be used to study how earthworms reproduce and grow (Yadav *et al.,* 2013; Bhat *et al.,* 2016; Yuvaraj *et al.,* 2020). By the action of earthworms and bacteria, waste is ingested, broken down, and digested by them into finer, humified, microbially active material (Khwairakpam and Bhargava, 2009). Earthworms boost soil fertility by enhancing its physical, chemical, and biological properties (Singh *et al*., 2021; Siddiqui *et al*., 2022; Singh *et al*., 2022; Singh and Singh, 2023a, b, c; Fatima *et al*., 2023; Singh *et al.*, 2024). Worm castings improve soil fertility and health (Papafilippaki *et al*., 2015). Vermicompost, the finished product is a granular substance with a high porosity and water-holding capacity. Anecics' geophytic nature allows them to break tougher substrates than epigeics (Gajalakshmi *et al.*, 2001a, b, 2002). Gergs *et al.* (2022) suggest that anecic species are ideal for the treatment of organic waste from canteens, houses, towns, farms, and other sources. *Lampito mauritii* can vermicompost a variety of organic wastes and has a high rate of waste decomposition (Suthar and Singh, 2008). The current study aimed to enhance the growth and development of *Lampito mauritii* in a specific combination that is important in increasing the conversion of organic waste into effective vermicompost.

MATERIALS AND METHODS

Collection and rearing of the earthworm Lampito mauritii:

The cultured earthworm, *Lampito mauritii* from the Vermibiotechnology Laboratory of the Department of Zoology, Deen Dayal Upadhyay, Gorakhpur University, Gorakhpur were used for the experiment. In the laboratory vermibed were prepared by using garden litter with buffalo dung on a cemented surface for the experiment.

Collection of buffalo dung and sugarcane bagasse:

Buffalo dung was collected from the farm houses located at different places of Gorakhpur. Cane sugar bagasse was collected from Saraiyan Distillery Mill, Saradar Nagar, Gorakhpur, U.P. Cane sugar bagasse was used for vermicomposting as well as feeding material for earthworms. For up to 10 days, these organic wastes were spread out in a layer and exposed to sunlight to get rid of the various harmful organisms and noxious gases (Garg *et al.*, 2005).

Experimental setup:

A cemented earth surface was used for the experiment. Two kilograms of binary combination of buffalo dung and sugar cane bagasse in different ratio i.e. BD+SCB (1:1), BD+SCB (2:1), BD+SCB (1:2), BD+SC (3:1), BD+SCB (1:3) as well as BD alone were prepared in beds of (30cm x 30cm x 10cm) at room temperature in the dark. The experiment to study the growth and development of earthworm *Lampito mauritii* was performed by the method of (Garg and Kaushik, 2005). The vermicomposting beds were turned over manually every 24 hours for 10 days in order to eliminate volatile substances. After this, 20 newly hatched ones of *Lampito mauritii* (obtained from same combination) were introduced into each bed. Each experiment was replicated at six times.

Reproduction and growth rate of the earthworm Lampito mauritii:

Every week, cocoons were collected using a 0.5 mm mesh sieve and gently washed, and their numbers were recorded on an individual basis. Before weighing, the cocoons were lightly washed in distilled water to eliminate any debris stuck to the sticky hull. The culture sets were carefully observed daily for cocoons, if any. The cocoons were immediately isolated and incubated. In both experiments, old cultural media was replaced with the same amount of fresh media on a weekly basis, thus food was not a limiting factor. After isolation, each cocoon was freshly put in a petri dish containing moist filtered paper at 30±2 ^{0}C and 70±5% RH. The hatching of cocoons was determined to estimate incubation, and the number of progenies emerged per cocoon were recorded. The growth rates (GRs) were calculated for complementary growth measure, for each 15-day growth interval. Their values are obtained by calculating the change in individual's growth during an infinitely short time interval. The GR was calculated GR= $(W_f - W_i)/\ \delta t$, where, W_i = initial earthworm mass (mg), W_f = final earthworm mass (mg), respectively, and δt = time interval measured in days. Biomass gained was recorded after 15 days interval up to 90 days in each bed.

Statistical Analysis:

All the experiments were replicated six times to ensure consistency in the results and figure out the mean with standard error. Analysis of variance was used to analyze the significant difference between the combinations and *t* test ($p < 0.05$) used to identify the homogeneous type of bedding in terms of reproduction and growth compared to the control (Sokal and Rohlf, 1973).

RESULTS

The result demonstrated that the period of initiation of clitellum development and formation of first cocoon and number of hatchlings emerged out of cocoon of *Lampito mauritii* in binary combination of buffalo dung with sugarcane bagasse in different ratio (1:1, 2:1, 1:2, 3:1, 1:3). Significant variations in the first appearance of clitellum, cocoon formation and hatchling emerged in *Lampito mauritii* was observed in binary combination of buffalo dung with sugarcane bagasse from the dung alone and its growth (Table1-2, Fig. 1-3).

The binary combination of buffalo dung with sugarcane bagasse in (1:1 ratio) hasten the development of clitellum than other combination and took only 16.17±0.54 days followed by buffalo dung with sugarcane bagasse in 3:1 ratio (18.33±0.76 day) and buffalo dung with sugarcane bagasse in 2:1 ratio (19.00±0.58 day) as compared to 24.67±0.43 day in buffalo dung alone. The combination of buffalo dung with sugarcane bagasse in 1:1 ratio showed significant period of initiation of cocoon production (29.50±0.43 day) than control set up (37.50±0.43 day). There was no significance difference in period of cocoon formation (14.17±0.26 to 18.00±0.37 day). Significantly high number of cocoon production was observed in combination of buffalo dung with sugarcane bagasse in 1:1 ratio (23.67±0.31 cocoon/worm) followed by buffalo dung with sugarcane bagasse in 2:1 ratio (21.67±0.68 cocoon/worm) with respect to BD alone (13.83±0.65 cocoon/worm). The higher numbers of hatchlings were observed per worm produce in buffalo dung with sugarcane bagasse in 1:1 ratio (51.36 hatchling/worm) than buffalo dung alone (23.00 hatchling/worm) (Table 1, Fig. 2). The binary combination of buffalo dung with sugarcane bagasse in 1:1 ratio showed maximum growth rate (9.15±0.45 mg/worm/day) of *Lampito mauritii* followed by buffalo dung with sugarcane bagasse in 3:1 ratio (8.95±0.33mg/worm/day) and buffalo dung with sugarcane bagasse in 2:1 ratio (8.65±0.43 mg/worm/day) as compared to buffalo dung alone (7.09±0.23mg/worm/day) (Table 2, Fig. 3). Data illustrated in Fig. 4 demonstrated that the growth rate per 15 days reached its maximum after 15 days in all combinations while it reached maximum at 45th day in buffalo dung alone. Thereafter, the growth rate decreased. It implies that the combination of buffalo dung with sugarcane bagasse cause maximum. The growth of binary combinations just after 15 days while without combination is need atleast 45 days.

Table 1: Effect of different combinations of buffalo dung with sugarcane bagasse on the production of cocoon and hatchling of *Lampito mauritii*.

Combi-nations	Initiation of clitellum (in days)	Initiation of cocoon production (in days)	Period of cocoon formation	Incubation period (days)	Cocoon/ worm	Hatchling /Cocoon	Reproduction rate (hatchling/ worm)
BD	24.67 ± 0.43a	37.50± 0.43a	18.00 ± 0.37a	30.17 ± 0.54a	13.83 ± 0.65c	1.67 ± 0.21a	23.00
BD+SCB(1:1)	16.17 ± 0.54c	29.50 ± 0.43c	15.20± 0.40a	26.83 ± 0.26a	23.67 ± 0.31a	2.17 ± 0.17a	51.36
BD+SCB(2:1)	19.00 ± 0.58b	32.00 ± 0.45b	14.17 ± 0.26a	27.17 ± 0.48a	21.67 ± 0.68b	2.00 ± 0.21a	43.34
BD+SCB(1:2)	20.17 ± 0.60b	33.50 ± 0.76b	16.67 ± 0.42a	29.17 ± 0.48a	19.67 ± 0.48b	1.67 ± 0.22a	32.84
BD+SCB(3:1)	18.33 ± 0.76b	32.83 ± 0.40b	15.50 ± 0.56a	27.83 ± 0.60a	21.00 ± 0.52b	1.83 ± 0.17a	38.43
BD+SCB(1:3)	23.17 ± 0.48a	34.00 ± 0.37b	16.83 ± 0.60a	29.33 ± 0.88a	16.17 ± 0.88c	1.17 ± 0.31a	18.92

Each value is the mean ± SE of six replicates. BD = Buffalo Dung, SCB= Sugarcane bagasse
**Mean differences in column followed by common letter are not significant at $p < 0.05$ (DMRT) in 30.0x30.0x10.0 cm^3 area of vermicompost bed.*

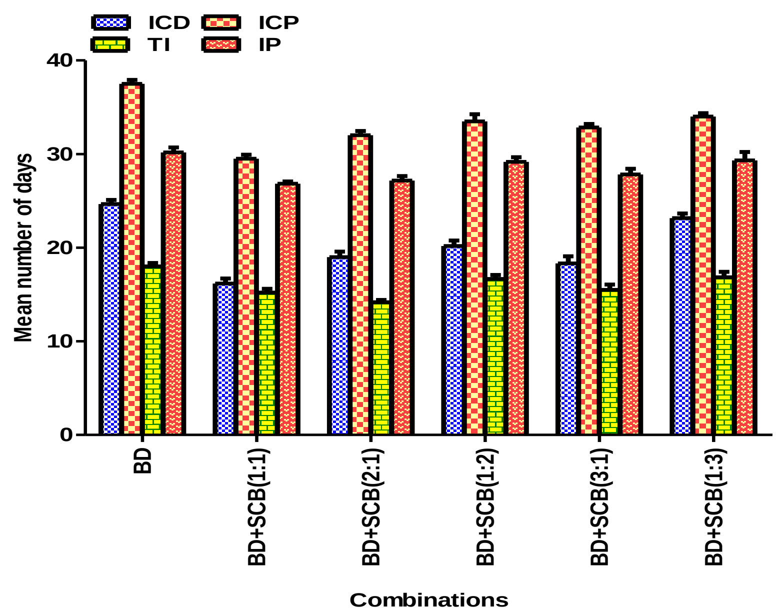

Figure 1: Effect of different combinations of buffalo dung with sugarcane bagasse on the initiation of clitellum development and cocoon production of *Lampito mauritii*. ICD= Initiation of clitellum (in days), ICP= Initiation of cocoon production (in days), TI= Time taken for cocoon production after initiation of clitellum development, IP=Incubation period, BD= buffalo dung, SCB=sugarcane bagasse.

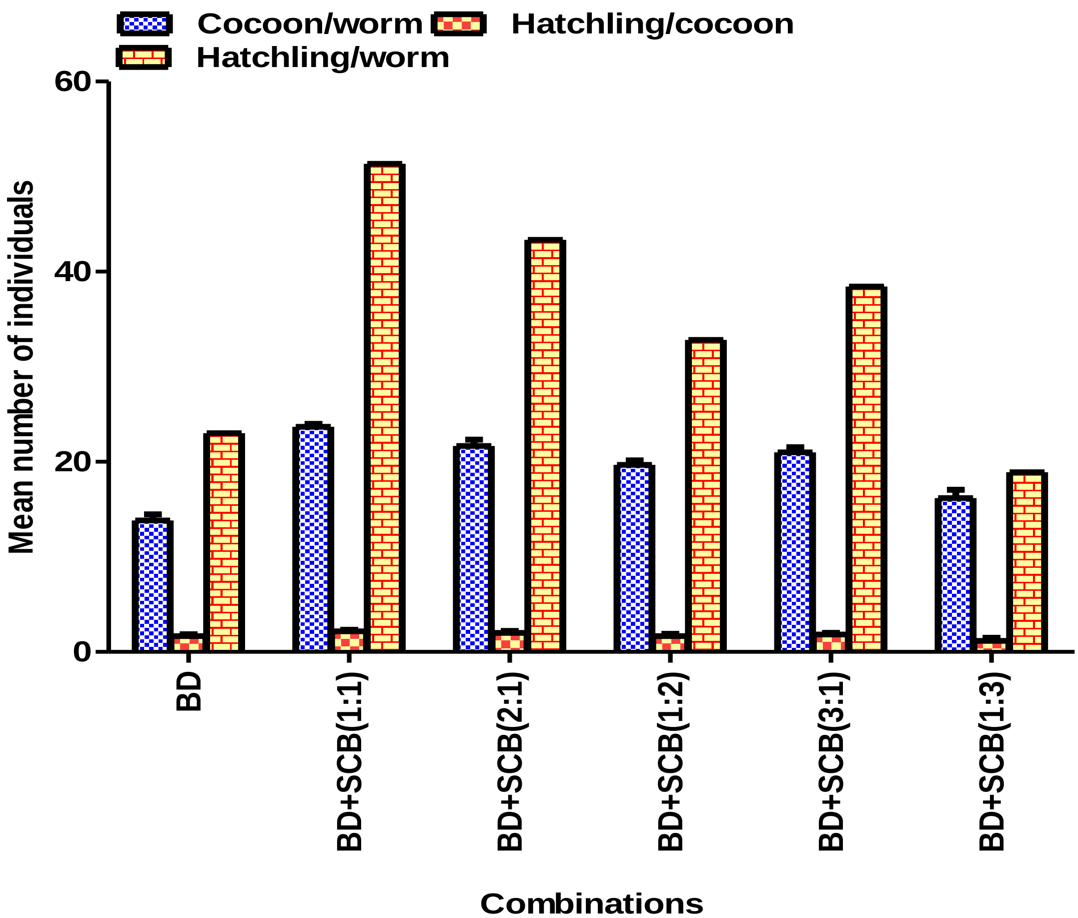

Figure 2: Effect of different combinations of buffalo dung with sugarcane bagasse on the production of cocoon and hatchling of *Lampito mauritii*. BD= buffalo dung, SCB= sugarcane bagasse.

Table 2: Growth rate of *Lampito mauritii* in different combinations of buffalo dung with sugarcane bagasse.

Combinations	Initial weight (mg)	Max. weight (mg)	Net weight (mg)	Growth rate (mg/worm/day)
BD	222.81 ± 6.40	860.98 ± 4.83d	638.17 ± 5.59c	7.09 ± 0.23
BD+SCB (1:1)	251.17 ± 5.81	1074.55 ± 5.28a	823.33 ± 4.09a	9.15 ± 0.45
BD+SCB (2:1)	249.28 ± 5.64	1027.78 ± 7.61b	778.50 ± 6.52b	8.65 ± 0.36
BD+SCB (1:2)	245.50± 5.89	886.17 ± 3.38c	640.67 ± 6.20d	7.12 ± 0.42
BD+SCB (3:1)	239.33 ± 5.62	1044.83 ±4.36b	805.50 ± 8.01a	8.95 ± 0.33
BD+SCB (1:3)	243.83 ± 3.36	855.83 ± 4.41d	612.00 ± 5.01c	6.80 ± 0.25

*Each value is the mean ± SE of six replicates. BD = Buffalo Dung, SCB = Sugarcane bagasse *Mean differences in column followed by common letter are not significant at $p < 0.05$ (DMRT) in 30.0x30.0x10.0 cm^3 area of vermicompost bed.*

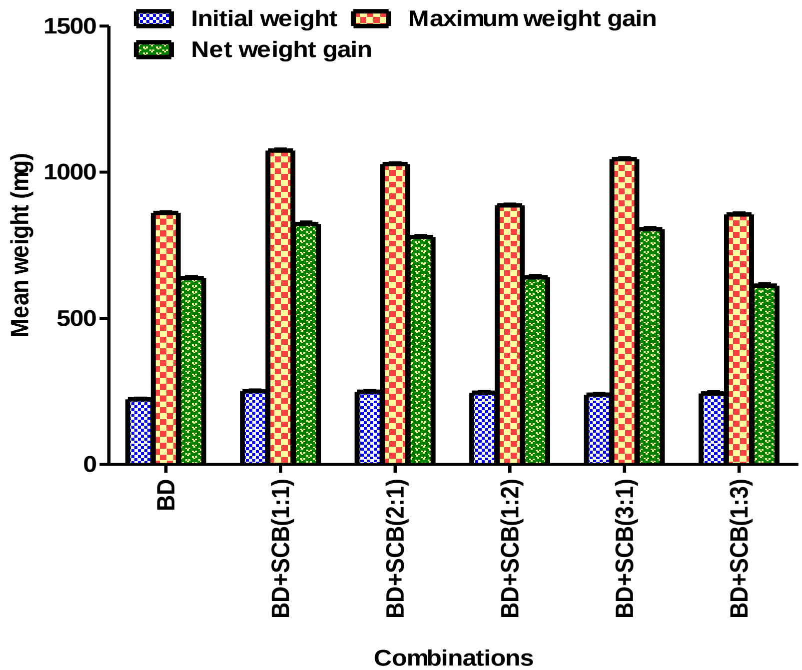

Figure 3: Effect of different combinations of buffalo dung with sugarcane bagasse on the growth of *Lampito mauritii*. BD= buffalo dung, SCB= sugarcane bagasse.

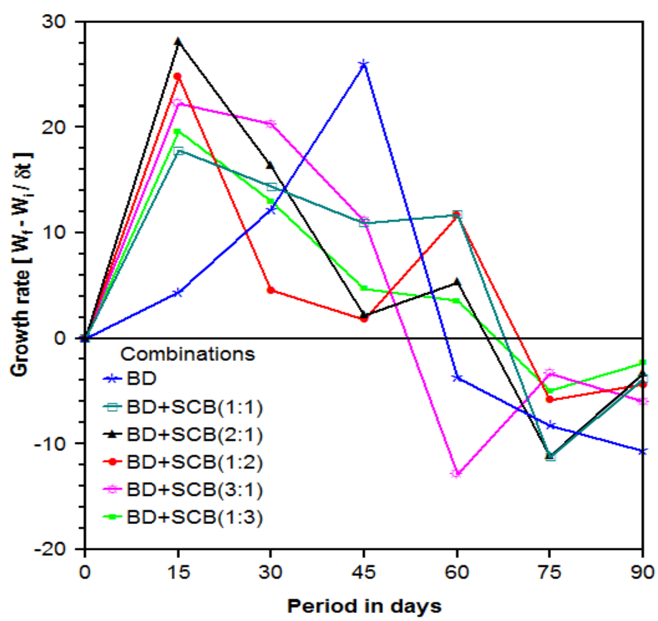

Figure 4: Growth curve of *Lampito mauritii* in binary combination of buffalo dung with sugarcane bagasse. BD= buffalo dung, SCB= sugarcane bagasse.

DISCUSSION

Organic carbon was the most essential feed material for earthworm growth and development, which might be attributed to its utilization in metabolism, resulting in a reduction in the final vermicompost. During vermicomposting, organic carbon content decreased substantially from the substrate for up to 90 days, and this carbon was used by earthworms in growth and development (Suthar, 2007; Yadav and Garg, 2010; Chauhan and Singh, 2013). According to Flack and Hartenstein (1984), one of the important factors for better clitellum growth in earthworms is the biochemical composition of the feed. Reinecke and Hallatt (1989) found that *Perionyx excavatus* produced more cocoons (1.4/worm/day, or 9.8/worm/week) starting on day 70 and continuing until the completion of the experiment. Elvira *et al*. (1998) found that combining paper mill effluent with cattle faeces increased the number of earthworms by 22 to 36-fold, as well as total biomass by 2.2 to 3.9 times. Food type, palatability, and quality have an impact on earthworm survival, growth rate, and reproductive potential (Tripathi and Bhardwaj, 2004; Gajalakshmi *et al.*, 2005). Our results support the findings of Bhat *et al.*, (2014), who found that an increase in pressmud content in the feed mixture led to a decrease in earthworm population. The optimal proportion of bagasse to buffalo dung was 1:1 ratio, since this was determined to be appropriate for *Lampito mauritii* growth and population development and the vermicompost's final granulating on its surface at its earliest stage. The amount of nitrogen in the substrates is an important factor in the formation of cocoons (Suthar, 2007). Food sources are also important for the production of cocoons, as noted by Fayolle *et al.*, (1997). In 1997, Ramalingam studied the growth, reproduction, and life cycle of *Eudrilus eugeniae* and *Lampito mauritii* utilizing pressmud. Higher concentrations significantly slowed down the rate of degradation and had a delayed and reduced effect on the production of cocoons in earthworms. Chauhan and Singh (2012) observed that there was a considerable increase in *Eisenia fetida* growth and cocoon formation when different binary combinations of buffalo dung with agro-wastes were used. The current study's increased hatchling formation is corroborated by research done by Chauhan and Singh, (2013) and Kaur *et al.*, (2010). The present study found that, with the exception of buffalo dung mixed in 1:3 ratios with sugarcane bagasse, worm biomass increased in all bagasse feed mixes. *Eudrilus eugeniae* feeding on various organic substrates showed an increase in body weight (Rathinamala *et al.*, 2008). The presence of fungi during the vermicomposting process provides the worms with more food, which increases their weight (Pramanik and Chung, 2011). Sogbesan *et al*. (2007) found that soil substrate had the lowest specific growth rate of 0.62% and cellulose substrate had the highest, at 0.76%. According to Pramanik *et al.*, (2007) and Manna *et al.*, (2003), sugarcane processing byproducts are excellent substrates for earthworm rearing. They also produce a product that is rich in chelating and phytohormonal components, has a high microbiological content, and stabilized humic compounds (Atiyeh *et al.*, 2001).

CONCLUSION

The earthworms are ecological engineers; they recycle the organic waste in rich organic vermicompost and improve the soil fertility as well as crop productivity. Vermicomposting is tool for waste management through earthworms and production and marketing of vermicompost. The vermicomposting is less expensive, non-hazardous, easily preparable, eco-friendly and safe for human, animal and environment. More earthworms convert more organic waste into rich organic manure. For the better growth and reproductive potential of earthworm *Lampito mauritii* need the best feed materials. On the basis of findings, among all the combination of buffalo dung with sugarcane baggase, the combination of BD + SCB (1:1) is the best combination of feed material for improvement of growth and reproductive potential of earthworm *Lampito mauritii*.

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