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ORIGINAL ARTICLE

By-products of bioenergy systems (anaerobic digestion and gasification) as sources of plant nutrients: scope of processed application and effect on soil and crop

Sampriti Kataki[1] · Samarendra Hazarika[2] · D. C. Baruah[1]

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Abstract
Management of the by-products generated during bioenergy conversion technologies is essential for technology sustainability and due to strict adherence to waste minimisation legislation. We investigated the potential of four types of bioenergy by-products (BEBPs), i.e. char from rice husk and digestates from 3 types of feedstocks: (i) *Ipomoea carnea*:cow dung (ICD), (ii) rice straw:green gram:cow dung (RGC) and (iii) cow dung (CD) as nutrient input for *Zea mays* L. Digestates were applied in four application phases, i.e., whole, solid, liquid and ash from solid digestates. BEBPs provoked significant changes in soil pH, electrical conductivity, available NPK, organic carbon and micronutrients depending upon both feedstock and phase. Digestates in solid and whole phases were found better as an organic amendment, whereas RGC and ICD digestates were superior in maintaining higher soil available P and K, respectively. BEBP showed satisfactory performance compared to BEBP-untreated control in terms of crop growth and yield, but chemical treatment resulted in the highest yield. N preservation against volatilization loss may be required through appropriate timing and method of application in case of high-ammonia-N-containing ICD digestates. Outcomes of this investigation are expected to be useful to undertake selective utilization practices of BEBPs for better handling and management.

Keywords Anaerobic digestion · Gasification · Bioenergy · By-products · Digestate · Biochar · Fertilizer

Introduction

Adoption of renewable energy technology is being emphasized globally due to limited availability of conventional energy resources and their adverse effect on climate change [1]. In this context, demands for affordable, reliable and flexible energy conversion processes such as anaerobic digestion that produces biogas and gasification that produces producer gas are rapidly growing [2]. However, functioning of these processes is also associated with generation of their inevitable by-products, i.e. digestate from biogas generation process and char from gasification process [3, 4]. With faster development and expansion of these renewable energy technologies, it is expected that there will be a simultaneous increase in generation of their by-products. Hence, for acceptability and sustainability of bio-energy conversion technologies, management of these residues along with the main energy output has become essential [3, 4]. Further, due to increased pressure on environmental resources and strict adherence to waste minimization legislation, it is necessary to search for alternative options of valorization of these residues through reuse and recycling [5, 6]. Considering the physicochemical properties analogous to organic manures, basic direction of bioenergy by-product (BEBP) management is through their utilization as fertilizer [7]. Moreover, depleting sources of non-renewable chemical fertilizer, their increased cost of production and harmful

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✉ Sampriti Kataki
sam.kataki@gmail.com; sam_kat@tezu.ernet.in

Samarendra Hazarika
Samarendra.ches@gmail.com

D. C. Baruah
baruahd@tezu.ernet.in

1 Energy Conservation Laboratory, Department of Energy, Tezpur University, Tezpur, Assam 784028, India

2 ICAR Research Complex for North Eastern Hill Region, Umium, Meghalaya 793103, India

implications associated with their long-term usage necessitate exploring the appropriate fertilizer potential of these bioenergy by-products. The prospect of application of such alternative fertilizers is very significant and relevant in areas where requirements of energy and fertilizer are vital.

Anaerobic digestion (AD) and biomass gasification (BG) technologies are versatile in terms of feedstock of varying composition which leads to generation of by-products of varying characteristics, i.e. digestate and char, respectively [8, 9]. In previous literature, evaluations of the fertilizer values of digestates from various feedstocks (poultry slaughterhouse waste, municipal sludge, guinea pig manure, cow dung and chicken droppings, food waste, agro-residues, co-digested dairy manure, food waste, etc.) were reported [10–19]. The properties of digestates were shown to vary with respect to feedstock in these studies.

From application perspective, anaerobic digestates were reported to improve soil properties by reducing the bulk density, increasing saturated hydraulic conductivity, moisture retention capacity [20, 21], aggregate stability [21, 22], and increasing soil nutrient content and microbial content [4, 19]. Further, application of digestate has been proven to be beneficial than using no fertilizer [13, 23, 24]. A range of studies reported variation in relative performance of digestates compared to mineral fertilizer from no statistical difference in yield [23, 25–27] to better yield than mineral fertilizer [28, 29]. In relation to its undigested counterpart, digestate applications were found better in terms of crop yield mainly because of improved status of N nutrition to the crops [30]. There are also studies reporting similar performance of digested and undigested feedstocks on crop [27, 31, 32]. Further, digestate application in some form (separated liquid) may also affect seed germination requiring appropriate measure [17]. Similarly, it has been reported that biochar properties depend upon feedstock and production temperature [33, 34], which eventually affect its fertilizing properties [11]. There are also reports indicating significant enhancement of soil and crop property from application of biochar [35, 36]. Zhang et al. [36] reported increase in soil pH, soil organic carbon, total nitrogen and decrease in soil bulk density after biochar application.

Positive influences of applications of bioenergy by-products are almost conclusively evidenced; however, information concerning the effect of BEBPs as fertilizer is situation specific with limited general applicability. There are reports stating issues and concerns in different aspects such as (i) agronomic (low or imbalanced concentration of nutrients, high salinity), (ii) economic/managerial (cost of transport and handling) and (iii) environmental (gaseous emissions, phytotoxicity, nutrient leaching and pathogen spread) which may result due to applications of digestate without taking appropriate measures. Generation of such information would necessitate identification of optimum application route with respect to varied input feedstocks and available processing options for their appropriate and selective use. Characteristics of BEBPs, type of soil, climate, crop, user-specific requirements are some of the major factors to be considered for determining the suitable route of BEBP application, which may otherwise compromise the nutrient value [17, 37, 38].

Keeping in view of the above discussion, the present study aims to investigate the prospects of upgrading the by-products of bioenergy systems into acceptable crop fertilizer with proven value through their processed application and their subsequent interaction with soil and crop. The objectives set for the current study are to investigate the effect of anaerobic digestate (in different application phases) and gasification char generated from local surplus biomass on soil health as well as on crop (maize) growth. The findings of the present research work enable to identify biomass which could be processed for getting both bioenergy as well as by-product-based organic fertilizer generation. No literature could be found that discusses the potential application of bioenergy by-products available from the selected set of biomass feedstock combinations (cow dung, *Ipomoea carnea*, green gram, rice straw, rice husk) which are abundantly available in the study region. An elaborate discussion has been made on the variation of characteristics and effect of fertilization with selected by-products with respect to different application options (whole digestate, separated solid, separated liquid, ash from solid digestate) with an aim to assess relative merits of these sources as per user need. It is expected that the findings of the study would be useful for successful application and acceptance of bioenergy-based fertilizer and in making decisions about future directions of bioenergy by-product utilization and research.

Materials and methods

Bioenergy by-products: generation and processing

Biomasses considered for the present study were cow dung, *Ipomoea carnea*, rice straw, green gram stover for AD and rice husk for BG. Identified biomasses for AD were co-digested in some predetermined ratios such as (i) *Ipomoea carnea* leaves:cow dung (ICD) (60:40 dry weight) and (ii) rice straw:green gram stover:cow dung (RGC) (30:30:40 dry weight). Moreover, cow dung alone was also considered as control feedstock for AD. Further, four different application options or phases, viz., (i) whole digestate, (ii) separated solid digestate, (iii) separated liquid digestate and (iv) ash from solid digestates were considered for assessment of each type of digestate from the three feedstocks. Separation of digestate into solid–liquid fraction was considered as it was expected to ease handling of digestate. Production of ash from solid digestate was taken as another application option, considering the fact that nutrients in residual digestate ash may be recycled back to crop field if digestates are used as solid fuel for secondary energy extraction [39]. Cow dung was used as co-digestion substrate in the digestion of *Ipomoea carnea* leaves, rice straw and green gram stover to improve their anaerobic digestibility.

The AD feedstocks were digested in laboratory-based 0.25 m^3 Shakti Surabhi® type biogas reactor designed by Vivekananda Kendra—Natural Resources Development Project (VK-NARDEP), Kanyakumari, India, kept in the Department of Energy, Tezpur University (Supplementary material Fig. 1). After collection, 150-micron filter bags were used for separating whole digestate into solid and liquid fractions. Ashing of dried solid digestate was done by open-air combustion at 400–450 °C. Details of feedstock considered, anaerobic digestion regime followed, BEBP generation and processing have been reported in previous-related publication [15].

Rice husk char considered as the gasification by-product was obtained from a biscuit factory (Nebisco Biscuit Pvt. Limited, Tezpur, Assam, India) equipped with rice husk fired gasification unit. Figure 1 presents the overall picture of BEBP generation and processing considered for the present investigation.

BEBP application experiment on maize crop

Experimental location and treatments

The pot experiment of growing maize crop was carried out in the Department of Energy, Tezpur University, Assam (latitudes 26°41″ 56°69″N and longitudes 92°49″ 59°92E). The prevailing meteorological information (temperature and relative humidity) during the study period (Supplementary material Fig. 2) was collected from an Automatic Weather Station installed by National Institute of Wind Energy, Chennai, India, in the University campus.

In total, there are fifteen treatment combinations (Table 1) including the control and chemical (NPK) fertilizer treatments. In thirteen treatments, BEBP obtained through anaerobic digestion/gasification process were used as source of N, P (partially) and K (partially) for growing maize as test crop.

Description of experiments

Experiments were conducted under controlled conditions using an open net house. Plastic pots (30 cm top diameter, 20 cm bottom diameter and 30 cm height) containing 14 kg of air-dried soil were used for growing the crop. Maize variety 'PAC 740' collected from Assam Agricultural University, Jorhat, Assam, was used for the study. For the NPK treatment, urea (46% N), single super phosphate (SSP, 16% P_2O_5) and muriate of potash (MOP, 60% K_2O) were used as sources of N, P and K, respectively. The experimental soil was collected from farmer's field, air dried and ground prior to filling the pots. Representative soil sample was drawn from the bulk soil and preserved for analysis of physical and chemical properties. The experiment was laid out in a completely randomized design with three replications during the season June–September 2014. Important properties of the experimental soils are presented in Table 2.

Required doses of BEBP were estimated based on the recommended dose of N (60 kg N ha^{-1}) and the total N content of each of the BEBPs separately. Double the calculated doses of BEBPs were applied to the crops considering the availability of only 50% of the total N of BEBPs to crops during the period of their growth. Application of recommended dose of NPK (60-40-40 kg ha^{-1} as urea, SSP and MOP, respectively) to the crops was considered as one of the treatments for testing the relative efficacy of the BEBPs. Additional amount of P (40 kg ha^{-1}) and K (40 kg ha^{-1}) at recommended dose was added to the pot soil under all treatments through SSP and MOP, respectively. The pots under control treatment received only P (40 kg ha^{-1}) and K (40 kg ha^{-1}) through chemical fertilizers. The BEBP mixed

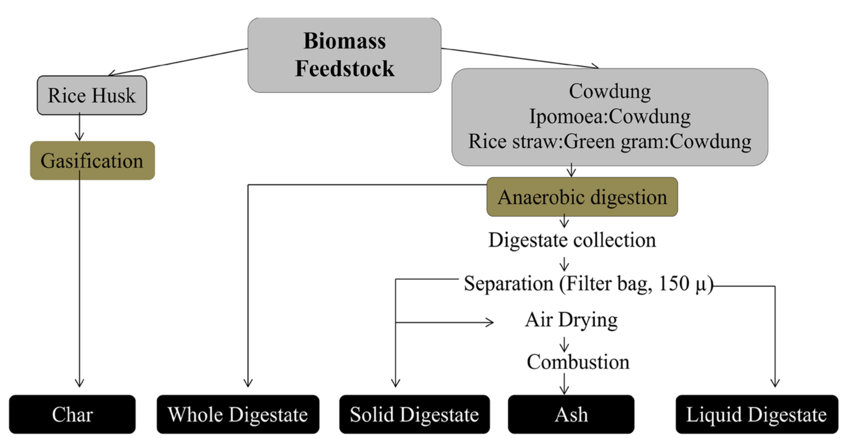

Fig. 1 Schematic diagram of bioenergy by-product generation and processing (adapted from [15])

Table 1 Description of the treatments

Treatment description				
S. no	Process	Material	Nature	Symbol
1	Anaerobic digestion	Cow dung	Whole	CD_W
2			Solid	CD_S
3			Liquid	CD_L
4			Digestate-derived ash	CD_A
5		Ipomoea carnea leaves:cow dung (60:40)	Whole	ICD_W
6			Solid	ICD_S
7			Liquid	ICD_L
8			Digestate-derived ash	ICD_A
9		Rice straw:green gram stover:cow dung (30:30:40)	Whole	RGC_W
10			Solid	RGC_S
11			Liquid	RGC_L
12			Digestate-derived ash	RGC_A
13	Gasification	Rice husk	Char	RH_C
14	Readymade fertilizers as source of nutrients	Chemical	NPK	NPK
15	Nil (control)	Control	Control	C

Table 2 Characteristics of soil used for growing maize crop

Parameter	Value
Soil texture	Clay loam (Silt: 23.95% Clay: 34.20% Sand: 41.85%)
Bulk density (Mg m^{-3})	0.90
Maximum water holding capacity (%)	66.83
Particle density (Mg m^{-3})	2.15
Total pore space (%)	57.85
pH (1:2.5)	5.43
Electrical conductivity (dS m^{-1})	0.22
Total carbon (%)	1.75
Soil organic carbon (%)	1.01
Total N (%)	0.13
Available N (mg kg^{-1})	125.00
Available P (mg kg^{-1})	10.39
Available K (mg kg^{-1})	67.50
DTPA extractable Cu (mg kg^{-1})	4.36
DTPA extractable Fe (mg kg^{-1})	189.00
DTPA extractable Zn (mg kg^{-1})	2.63
DTPA extractable Mn (mg kg^{-1})	46.10

as basal dose with the whole pot soil was allowed to equilibrate for 1 week prior to sowing of seeds. NPK fertilizers were added and thoroughly mixed with the soil 2 days prior to seed sowing.

In each pot, single plant was maintained throughout the period of the experiment. Further, water was added to the pots to maintain soil moisture at ~60% of the maximum soil water holding capacity and identical intercultural operations were done throughout the period of the experiment. The crops were harvested after attaining the physiological maturity.

Analytical methods

After crop harvesting, representative soil samples collected from each pot were air dried under shade and sieved through a 2-mm sieve for analysis. Texture, pH (1:2.5), electrical conductivity (1:2.5), maximum water holding capacity, total pore space, particle density, bulk density, organic carbon, available N, available P, available K and DTPA extractable Cu, Fe, Mn, Zn were the initial properties of soil determined prior to experimentation, whereas in soil samples collected after harvesting of the crops, following soil properties were determined, viz., pH, electrical conductivity, soil organic matter, available N, available K, available P and DTPA extractable Cu, Fe, Mn, Zn. Standard protocols and methodologies used for soil, plant and grain analyses are described in Table 3.

Plant height and leaf area were measured prior to the harvesting of the crops. Measurements of other parameters (ear length of cob, root length of plant) were taken immediately after crop harvesting. Measurement of the dry weight of roots, cob, and shoot was taken after drying the samples at 60 °C. Dried samples of shoot, grain and seed were used for different chemical analysis after grinding using an electrical grinder. Plant height was measured as the height from the base of each plant (soil surface) to the base of the tassel at physiological maturity. For the determination of leaf area, three full matured leaves were selected per plant at 50% silking (60 days from the date of emergence). Leaf

Table 3 Standard methods/protocol used for soil and plant analyses

Soil	Protocol/method	Plant/grain	Protocol/method
Water holding capacity Porosity Volume expansion Bulk density Specific gravity	Keen–Raczkowski Box Method [40]	Plant height Ear height Maximum root length Root weight	Physical measurement
pH	pH meter in a 1:2.5 (*w*/*v*) soil–water suspension	Total dry matter Grain yield 1000 kernel weight Ear length	Physical measurement after drying at 60 °C
Electrical conductivity	Conductivity meter in a 1:2.5 (*w*/*v*) soil–water suspension	Shoot/grain Total N	CHN analysis
Total N	CHN analysis	Shoot/grain P, K	Diacid digestion and ICP analysis
Soil texture	International Pipette method [41]		
Soil organic carbon	Walkley and Black rapid titration method [42]	Root:shoot ratio	Estimated from root:shoot weight
Available N	Subbiah-Asija method [43]	Grain protein concentration	Estimated from total N concentration of grain using Jones Factor (6.25) [44]
Available P	Bray and Kurtz method [45]	Crop NPK uptake	Estimated by NPK concentration of shoot and grain and crop total dry matter
Available K	Flame photometer	N harvest index	$\frac{\text{Total N uptake by grain}}{\text{Total crop N uptake}}$
Available micronutrients (Cu, Fe, Zn, Mn)	DTPA extraction and ICP analysis	Leaf area	LA = 0.75 × leaf length × maximum breadth [46]
		Leaf area index	$\frac{\text{Leaf area}}{\text{Sampled ground area}}$

area index was calculated by dividing the leaf area per sampled ground area (surface area of the pot) [47]. To estimate dry matter accumulation and partitioning in different parts of the maize crop, plant parts were weighed separately as roots, cob and above-ground biomass after drying at 60 °C. Root:shoot ratio was determined by taking into account the dry weight of above-ground biomass to the dry weight of root. The weight of the oven-dried grain of each replicated pot (single plant) was taken using electronic balance and averaged over replicated pots under each treatment to get the gain yield (g pot^{-1}).

Statistical analyses

The data related to soil and crop parameters were subjected to statistical analysis using SPSS 16.0 programme for Windows. Pearson bivariate correlation coefficients were calculated using SPSS to understand the levels of relationships among the selected parameters of BEBP, soil and crop. Soil and crop data corresponding to three replications were analysed by two-factorial analysis of variance. Means with statistically significant difference were indicated by different letters in tables after comparison using Duncan's multiple-range test at $p \leq 0.05$. In the tables presented in "Results and discussion", parameter values are mean of three replications ± standard deviation of observed values.

Results and discussion

Characteristics of bioenergy by-products

Detail characteristics of the selected by-products in terms of their plant nutrient contents (macro- and micronutrient), organic matter content (total organic carbon, lignin, cellulose, hemicelluloses, CN ratio), spectroscopic characterization (FTIR, XRD), morphology, phytotoxicity, heavy metal content as well as other physicochemical parameters (pH, EC, TS) were reported in our previous publications [15, 16]. Some of the major BEBP characterization parameters adapted from Kataki et al. [15] are presented in Table 4 and highlighted below. Ipomoea:cow dung (ICD) digestates showed significantly higher EC in all the application phases compared to the respective phases of digestates from cow dung (CD) and rice straw:green gram:cow dung (RGC) feedstocks, which may be due to the presence of rich alkaloid content (swainsonine and calystegines) of *Ipomoea carnea* leaves [48, 49]. In general, organic matter, macronutrients, micronutrients and heavy metal contents vary both with respect to types of feedstock and application options or phases of by-products. Ipomoea digestates in all four application phases contain significantly higher plant macronutrients (N, K, Ca and S), ammonia-N and micronutrients (Fe, Cu and Mo) compared to the respective phases of CD and RGC digestates. Rice husk char from gasification was

Table 4 Total solid, pH, electrical conductivity, macronutrients (TN, NH_4-N, P, K), total organic carbon (TOC) and TOC:TN of BEBPs (Adapted from [15])

Sample	TS, g kg^{-1}FM	pH	EC, mS cm^{-1}	TOC, % DM	TOC:TN	TN, g kg^{-1} FM	NH_4-N, mg kg^{-1} FM	K, g kg^{-1} FM	P, g kg^{-1} FM
CD_W	113.4 ± 2.5^{a}	7.3 ± 0.02^{a}	4.3 ± 0.01^{f}	27.42 ± 1.2^{ad}	16.03	1.94 ± 0.05^{ah}	123 ± 12.3^{ac}	0.85 ± 0.04^{a}	0.28 ± 0.02^{ad}
ICD_W	113.6 ± 1.8^{a}	7.8 ± 0.04^{b}	7.2 ± 0.02^{g}	40.16 ± 3.7^{b}	16.73	2.72 ± 0.17^{ab}	790 ± 50.0^{b}	1.53 ± 0.08^{a}	0.37 ± 0.01^{a}
RGC_W	135.2 ± 3.4^{a}	8.0 ± 0.01^{c}	3.8 ± 0.03^{e}	43.33 ± 2.8^{c}	44.21	1.30 ± 0.12^{ac}	116 ± 8.5^{a}	1.06 ± 0.08^{a}	0.19 ± 0.00^{cd}
Mean	120.7	7.7	5.1	36.97	25.65	1.98	343	1.14	0.28
CD_S	299.2 ± 1.5^{c}	8.1 ± 0.01^{d}	1.5 ± 0.10^{a}	25.82 ± 1.5^{d}	17.93	4.44 ± 0.37^{d}	165 ± 7.3^{c}	1.53 ± 0.03^{a}	0.65 ± 0.03^{e}
ICD_S	225.3 ± 1.3^{b}	8.0 ± 0.01^{e}	2.0 ± 0.00^{b}	38.49 ± 1.8^{b}	17.33	4.71 ± 0.73^{d}	399 ± 28.6^{d}	1.60 ± 0.01^{a}	0.87 ± 0.00^{f}
RGC_S	235.4 ± 2.0^{b}	7.0 ± 0.01^{f}	1.5 ± 0.01^{a}	44.54 ± 2.4^{c}	36.21	2.60 ± 0.31^{bh}	138 ± 29.6^{ac}	1.34 ± 0.06^{a}	0.52 ± 0.01^{g}
Mean	253.2	7.7	1.6	36.28	23.82	3.91	234	1.49	0.68
CD_L	31.0 ± 0.8^{d}	8.5 ± 0.01^{g}	4.3 ± 0.02^{f}	$0.55^{*} \pm 0.07^{e}$	7.85	0.85 ± 0.08^{ce}	109 ± 3.5^{a}	0.69 ± 0.05^{a}	0.17 ± 0.01^{cd}
ICD_L	20.3 ± 1.1^{d}	8.8 ± 0.02^{h}	9.3 ± 0.02^{h}	$2.00^{*} \pm 0.9^{e}$	9.09	2.18 ± 0.30^{bh}	935 ± 7.8^{e}	1.61 ± 0.04^{a}	0.19 ± 0.00^{cd}
RGC_L	7.2 ± 1.1^{d}	8.9 ± 0.03^{i}	3.4 ± 0.01^{d}	$0.18^{*} \pm 0.02^{e}$	7.20	0.25 ± 0.05^{e}	113 ± 10.9^{a}	0.81 ± 0.08^{a}	0.06 ± 0.01^{c}
Mean	19.5	8.7	5.6	0.91	8.04	1.09	386	1.03	0.14
CD_A	997.3 ± 3.2^{f}	9.9 ± 0.02^{j}	2.0 ± 0.05^{b}	6.92 ± 0.9^{f}	9.22	7.54 ± 0.41^{f}	164 ± 45.7^{c}	9.68 ± 0.61^{b}	8.42 ± 0.16^{h}
ICD_A	991.7 ± 2.7^{f}	10.1 ± 0.00^{k}	4.2 ± 0.03^{f}	11.85 ± 1.6^{g}	11.17	11.62 ± 0.93^{g}	423 ± 38.4^{f}	15.35 ± 0.13^{c}	5.00 ± 0.18^{i}
RGC_A	999.2 ± 1.5^{f}	9.7 ± 0.03^{l}	2.2 ± 0.01^{c}	19.48 ± 1.9^{h}	18.73	11.26 ± 0.85^{g}	109 ± 5.7^{a}	9.53 ± 0.17^{d}	3.78 ± 0.12^{j}
Mean	996	9.96	2.8	12.75	13.04	10.14	232	11.52	5.73
RH_C	999.7 ± 2.0^{f}	10.3 ± 0.01^{m}	2.0 ± 0.10^{b}	28.65 ± 2.9^{a}	143.25	1.8 ± 0.01^{a}	48 ± 3.2^{g}	8.02 ± 0.09^{e}	1.10 ± 0.01^{j}

Means in a column followed by a common letter are not significantly different at $p \leq 0.05$ based on Duncan's multiple-range test

*The values are expressed in fresh weight basis

found to be nutrient poor in comparison to the by-products of AD. Solid–liquid separation results in remarkable variation in distribution of plant nutrients. After separation, liquid fractions of all three digestates (ICD, CD and RGC) retain higher fraction of NH_4^+-N (61–91%) compared to the solid (12–41%) and ash phases (1.4–4.7%). It is also found that TOC contents are higher in solid digestates with a variation among feedstocks (RGC > ICD > CD) than the liquid phase of digestates. Data on BEBP micronutrient status (Ca, Mg, S, Cu, Fe, Mn, Zn) published in the previous publication of the authors [15] have been added in Appendix (Supplementary material Table 1).

It is important to evaluate how the nutrients are distributed between the separated solid and liquid fractions after separation of solid and liquid of digestate. Hence, after determining the value of each parameter (i.e. mass and major plant nutrients such as N, K, P, Ca, Mg, S) in solid and liquid fractions separately (Table 4), the values are expressed as percentage of the total content (i.e. total value of whole unseparated digestate), to understand what percentage of the total content is retained by each solid and liquid fraction. Figure 2 shows percentage distribution of mass and major plant nutrients (N, K, P, Ca, Mg, S) among solid and liquid fractions of digestate after separation.

Higher fraction (69–76%) of the total mass of digestate was distributed to liquid fraction, whereas separated solid fraction contained 24–31% of the total mass of

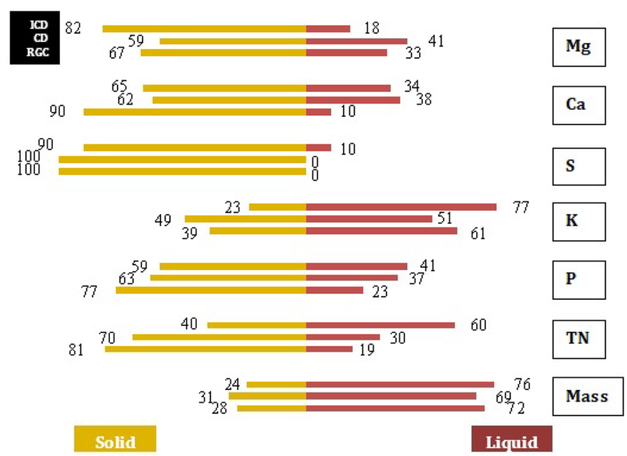

Fig. 2 Percentage distribution of mass and major plant nutrients among solid and liquid fractions of digestate

digestate. Again, after solid–liquid separation, higher fraction (59–77%) of P remains in solid phase with a variation depending upon the type of feedstock. On the other hand, liquid phases retain higher fraction of K (51–77%). In case of secondary macronutrients (Ca, Mg and S), in digestates from all three feedstocks, higher fraction of Ca (62–90%), Mg (59–82%) remained in solid fraction, while S remained typically in solid fraction (90–100%) after solid–liquid separation.

Variations in soil properties as affected by BEBP application

Soil pH and electrical conductivity

Application of BEBP was found to have a significant effect on pH of post-harvest soil. All treatments except ICD_L and ICD_A could maintain higher pH of the soil as compared to that under control and NPK treatment (Fig. 3a). Averaged over type of feedstock, application of digestates from RGC feedstocks resulted in highest pH rise (5.50–5.60) and pH under RGC_S and RGC_L remained significantly higher than control. Rise in soil pH of soil could be attributed to the microbial decomposition and de-carboxylation of organic acid anions [50, 51]. On the other hand, upon application of ICD_L digestates which had relatively higher NH_4^+ content (Table 4), nitrification of NH_4^+ might have taken place, in which, H^+ ions are released into soil solution, lowering soil pH [52]. Considering the abundance of acid soil in more than 80% soils of northeast India, application of BEBPs as soil amendment for improving the quality of acid soils of the region could have a good prospect. Application of BEBP was found to have a statistically significant impact on the soil EC (Fig. 3b). The DMRT test shows that rise in EC under CD_S, ICD_L, CD_A and RGC_A was significantly higher as compared to that under control as well as NPK fertilizer treatments.

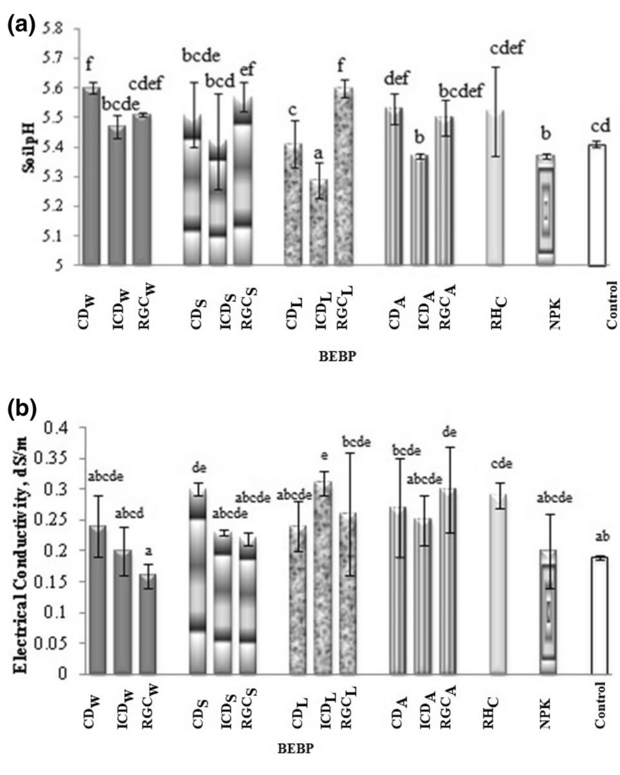

Fig. 3 Variation of **a** soil pH and **b** soil electrical conductivity under various treatments

Soil organic carbon (SOC) and nitrogen content

Application of BEBP showed a significant impact on organic carbon of soil with an increase in SOC in the range 1.78–32.62% (Table 5). SOC concentration was highest in CD_S (1.24 ± 0.00%) [significantly different than control soil (0.93 ± 0.02)] and was the lowest in RGC_A (0.94 ± 0.01%). It was observed that, in each feedstock, there was a variation of SOC with respect to the application phase of BEBP. Averaged over phase, highest SOC was observed under the application of digestates in solid phase (mean 1.19%) followed by whole digestates (mean 1.13%), which may be attributed to the presence of high molecular weight organic complex such as lignin and cellulose in these that remain undegraded during anaerobic digestion [53]. On the other hand, increase in SOC due to application of liquid (mean 1.02%) and ash (mean 1.02%) digestates was at par to that under control, which may be related to their lower TOC content. Though insignificant, the increase of SOC due to ash amendment may be explained by the presence of residual recalcitrant C of the ash [54]. In acid soils, abundantly available oxides of Fe and Al might constitute a major mechanism for organic matter stabilization forming aggregates and thus protecting from microbial degradation [55]. Presence of higher concentration of exchangeable and readily soluble Al in acid soil might have protected the added soil organic matter from rapid microbial degradation by forming organo-aluminium complexes and thereby enhanced the concentration of SOC as compared to that under control treatment [56]. Significant improvement of SOC (1.22 ± 0.02%) from application of balanced doses of chemical fertilizer could be attributed to higher biomass generation and better root mass development and subsequently higher return of plant residues to soil [57, 58]. The results showed that application of RH_C increased the SOC to the tune of 21% as compared to the control condition.

The effect of BEBP was found to be non-significant on soil TN (results not shown) and significant on soil available N ($p < 0.01$). In post-harvest soil, AN was enhanced by 29.06% (under CD_S, 164 ± 11.4 mg kg^{-1}) to 1.28% (under RGC_A, 129 ± 1.6 mg kg^{-1}) (Table 5). Increase in AN of soil could be related to increase in SOC concentration as evidenced by their statistically significant positive correlation ($r = 0.68$, $p < 0.01$). This result is in agreement with the general observation that the N content of soil parallels to that of organic carbon [59]. Increase in AN in soil under digestate application might also be attributed to the direct addition of N through digestate to the available pool of the soil [60, 61]. The increase in available nitrogen under RH_C treatment may

Table 5 Effect of BEBP application on soil organic carbon, available nitrogen, available P, available K and micronutrients (Cu, Fe, Mn, Zn) in post-harvest soil

BEBP	Soil organic carbon, %	Soil available N, mg kg^{-1}	Soil available phosphorus, mg kg^{-1}	Soil available potassium, mg kg^{-1}	Cu, mg kg^{-1}	Fe, mg kg^{-1}	Mn, mg kg^{-1}	Zn, mg kg^{-1}
CD_W	1.16 ± 0.00^{fg}	151.90 ± 4.90^{def}	12.45 ± 1.50^{abc}	83.65 ± 3.06^{bcde}	5.85 ± 0.89^{ab}	273.15 ± 61.88^{abc}	29.17 ± 7.62^{abc}	14.02 ± 2.70^{d}
ICD_W	1.19 ± 0.01^{gh}	138.83 ± 1.63^{bc}	14.90 ± 3.14^{cd}	92.72 ± 9.23^{e}	5.41 ± 1.56^{a}	273.88 ± 46.81^{abc}	30.24 ± 9.32^{abc}	7.63 ± 0.57^{ab}
RGC_W	1.02 ± 0.01^{bc}	143.73 ± 3.26^{bcd}	14.46 ± 1.58^{abcd}	78.42 ± 9.36^{bcd}	5.18 ± 0.50^{a}	282.15 ± 1.61^{bc}	37.30 ± 6.15^{c}	8.71 ± 0.17^{bc}
Mean	1.13	144.82	13.93	84.93	5.48	276.39	32.23	10.12
CD_S	1.24 ± 0.00^{h}	164.42 ± 11.47^{g}	17.82 ± 1.69^{de}	78.93 ± 2.52^{bcd}	7.30 ± 0.22^{c}	407.67 ± 2.89^{d}	38.50 ± 3.47^{c}	11.54 ± 2.57^{cd}
ICD_S	1.17 ± 0.01^{fg}	160.06 ± 6.53^{fg}	12.46 ± 1.24^{abc}	87.38 ± 6.45^{de}	5.29 ± 0.20^{a}	272.36 ± 18.07^{abc}	31.28 ± 0.18^{abc}	5.99 ± 0.83^{ab}
RGC_S	1.16 ± 0.02^{fg}	156.80 ± 0.00^{fg}	19.48 ± 3.52^{e}	77.10 ± 3.65^{abcd}	5.31 ± 0.38^{a}	270.34 ± 17.75^{abc}	31.29 ± 3.50^{ac}	11.63 ± 1.27^{cd}
Mean	1.19	160.42	16.58	81.13	5.96	316.79	33.69	9.72
CD_L	1.05 ± 0.00^{cd}	144.82 ± 8.22^{cde}	12.70 ± 1.00^{abc}	81.74 ± 2.81^{bcde}	5.39 ± 0.46^{a}	264.64 ± 3.08^{ab}	31.33 ± 14.25^{c}	7.44 ± 0.85^{ab}
ICD_L	1.03 ± 0.06^{bc}	129.13 ± 3.26^{a}	14.56 ± 0.78^{cd}	83.28 ± 5.97^{bcde}	7.04 ± 0.32^{bc}	390.18 ± 27.53^{d}	40.25 ± 8.18^{c}	8.60 ± 1.09^{bc}
RGC_L	1.00 ± 0.04^{b}	140.46 ± 3.26^{bc}	15.95 ± 1.75^{cde}	73.18 ± 2.42^{ab}	4.59 ± 0.17^{a}	229.59 ± 8.95^{a}	21.13 ± 2.26^{ab}	4.71 ± 2.46^{a}
Mean	1.02	138.13	14.40	79.40	5.57	288.13	30.90	6.91
CD_A	1.04 ± 0.01^{bc}	145.36 ± 1.63^{cde}	17.47 ± 2.45^{de}	67.02 ± 3.33^{a}	6.66 ± 0.58^{ab}	305.71 ± 5.19^{c}	34.03 ± 6.04^{bc}	8.01 ± 1.74^{ab}
ICD_A	1.09 ± 0.00^{de}	135.02 ± 4.98^{ab}	24.18 ± 1.88^{e}	80.78 ± 6.02^{bcd}	4.88 ± 0.19^{a}	231.74 ± 10.36^{a}	22.24 ± 4.46^{ab}	7.20 ± 2.57^{ab}
RGC_A	0.94 ± 0.01^{a}	129.03 ± 1.63^{a}	18.10 ± 1.84^{de}	81.85 ± 2.54^{bcde}	6.13 ± 0.50^{bc}	262.43 ± 27.04^{abc}	36.71 ± 2.03^{c}	8.61 ± 0.14^{bc}
Mean	1.02	138.74	19.91	76.55	5.89	266.62	30.99	7.94
RH_C	1.13 ± 0.01^{ef}	153.53 ± 3.26^{ef}	17.38 ± 2.76^{de}	85.20 ± 13.55^{cd}	5.47 ± 1.21^{a}	264.98 ± 34.53^{a}	30.45 ± 3.41^{abc}	7.93 ± 0.08^{ab}
NPK	1.22 ± 0.02^{h}	138.83 ± 1.69^{bc}	11.01 ± 0.61^{ab}	82.65 ± 2.80^{bcde}	5.78 ± 0.66^{a}	266.20 ± 10.38^{abc}	28.24 ± 0.25^{abc}	6.50 ± 3.01^{ab}
C	0.93 ± 0.0^{a}	127.39 ± 3.26^{a}	10.75 ± 1.63^{a}	75.43 ± 0.67^{cd}	5.14 ± 0.04^{a}	264.31 ± 2.26^{c}	28.14 ± 0.25^{abc}	5.04 ± 1.51^{a}
Level of significance	*	**	**	**	**	**	**	**

Different lowercase letters in same columns indicate significantly different means at $p \leq 0.05$ between the treatments

NS not significant

*, **Significant at $p < 0.05$ and $p < 0.01$ level, respectively

be attributed to higher nutrient retention capacity of biochar leading to accumulation of ammonia-N in soil [62].

There was variation in rise of AN with respect to both application phase and type of feedstock of BEBP. Maximum rise in AN was observed in case of soil treated with solid phase (by 27.7% compared to control). It is to be noted that in spite of higher content of mineralized N in separated liquid fractions of digestate compared to that of the separated solid (as mentioned in Characteristics of bioenergy by-products) for the same total N content, the rise in soil AN under liquid digestate treatment was not apparent. High temperature (22–37 °C, summer season) during the experimental period might have caused higher volatilization loss of N compounds through ammonia from the soil leading to lower accumulation of available N in soil treated with liquid digestates [63, 64]. Relatively lower rise in soil AN under ICD treatments (by 6%), compared to rise under CD (by 20%) and RGC (by 11%) may be attributed to the loss of N from soil after its application due to high temperature as discussed above since ICD digestates were found to have higher pH and ammonia-N compared to other respective phases. Immediate uptake by crop is another possibility, leading to minimum soil N accumulation in the post-harvest soil, as maize crop is an extensive feeder.

Soil available P, available K and micronutrient (Cu, Fe, Mn, Zn) concentration

The effect of different treatments on concentration of available P in soil was found to be significant ($p < 0.01$) with an increase in the range of 7.4–125% (Table 5). Soil available P was highest under ICD_A treatment (24.18 ± 1.88 mg kg^{-1}), which was significantly different from the rest of the treatment. Averaged over phase, concentration of available P in soil showed the following trend in decreasing order, viz., ash digestate ≥ char > solid digestate > liquid digestate > whole digestate > chemical > control. Feedstock wise, application of digestates from RGC feedstock resulted in highest soil available P (mean 16.96 mg kg^{-1}). RH_C also led to significantly ($p < 0.05$) higher (62% over control) accumulation of available P in soil than that under control treatment.

Organic fertilizer addition can significantly increase the availability of soil P to plants and decrease the P adsorption capacity of soils [65, 66]. When organic substrates are added to soil, they release organic acids, which have greater affinity towards Al, Fe and Mn abundantly available in acid soil and form insoluble complexes with these metal cations [66–68]. They can also be adsorbed to Al, Fe and Mn oxide surfaces consequently blocking P-adsorption sites that results in increased availability of P in soil [67]. The significant rise in soil pH due to the addition of the BEBP might also have played a significant role in increasing the availability of P in soil under BEBP treatments [68]. We found soil available P to significantly correlate with TS (=0.66, $p<0.05$), Ca ($r=0.68$, $p<0.01$) and Mg ($r=0.59$, $p<0.01$) of the BEBPs, indicating the presence of P in association with these fractions (Ca, Mg content of BEBPs are shown in supplementary material T1).

Magnitude of increase in available K due to BEBP treatment varied between 2.2 (under RGC_S) and 23% (under ICD_W). In general, available K of soil was enhanced to the maximum under ICD digestates, which may be related to its higher K concentration compared to the other two feedstocks. Barring CD_A and RGC_L treatments, soil available K under all BEBP was found to be at par or higher than that under fertilizer NPK treatment. RH_C also led to 13% increase of soil available K accumulation than that under control treatment.

Following BEBP application, the concentration of micronutrients (Cu, Fe, Mn, Zn) treated either with the solid or whole digestate or char were found to increase in soil (Table 5). The importance of micronutrient fertilization on soils is implicit from the fact that the primary nutrients unless supported by micronutrient supplementation may not be adequate to improve growth and productivity of micronutrient-sensitive crops. Apart from soil acidity, deficiency of micronutrient, particularly Zn, B and Mo, in soils of northeast India is one of the major factors constraining productivity of crops [69]. Application of BEBP would be of significant consideration particularly in the context of enhancing the Zn fraction of northeast soil, as Zn is one of the most deficient micronutrients in this region [69]. Zn deficiency is mainly attributed to a number of soil and climatic factors including high rainfall, light texture, abundance of Fe and Al oxides and low rate of organic matter decomposition. In the present study, Zn concentration in the post-harvest soil under selected BEBP treatments, particularly CD_W (by 115% over control), CD_S (by 77% over control) and RGC_S (by 79% over control) was enhanced to a level significantly higher ($p<0.05$) than control. The rise in micronutrient concentration under different treatments did not show any particular trend, and no general correlation was found with the micronutrient concentration of BEBP. Copper ($r=0.59$, $p<0.05$) and Fe ($r=0.44$, $p<0.05$) concentrations showed a positive correlation with electrical conductivity of soil.

Growth of maize as affected by BEBP application

The growth (leaf area, plant height, dry matter yield, maximum root length, root weight, root:shoot ratio) and yield (ear length, 1000 kernel weight, grain yield) parameters of maize observed in the pot experiment are presented below in 'Above-ground biomass and root development' and 'Yield attributing parameters and grain yield'.

Above-ground biomass and root development

Leaf area (LA) was significantly affected by different BEBP treatments (Table 6). Highest and significantly different LA was observed under ICD_W (537 ± 4.2 cm^2). ICD_W application also significantly increased leaf area index (LAI) to the maximum (0.76) as compared to minimum LAI under control (0.61). Nitrogen fertilization in the form of BEBP addition that has created a favourable growth environment in soil might have promoted organogenesis and growth and expansion of the aerial parts, leading to higher LA and LAI under BEBP treatments as compared to control plot [70, 71]. It has been reported that under limiting N (as in case of control soil) and under P deficiency (as in case of control soil, being more acidic as compared to BEBP applied), reduce the leaf production, individual leaf area and total leaf area [72]. Among BEBPs, higher and immediate availability of ammonia-N in all phases of ICD digestates may have contributed to its better leaf expansion under these treatments. Application of BEBP was also found to increase the plant height under all treatments compared to control. Magnitude of increase was in the range of 3.9–35%. Similar to the effect on leaf area, maximum plant height was recorded in ICD_W treatment (211 ± 5.08 cm), whereas it was minimum in control treatment (155 ± 4.24 cm). While comparing with NPK treatment, it was observed that height attained under CD- and ICD-based digestates was higher or at par with that of NPK treatment. Marked increase in plant height attained under different BEBP treatment can be explained by the overall improvement in soil fertility (e.g. higher availability of N, K, P, reduction in soil acidity and soil organic matter accumulation) that helped in balanced nutrition of the crop resulting in relatively better growth of maize crops as compared to control plots. There are reports on enhanced plant growth, increased number and length of the internodes resulting in progressive increase in plant height due to direct addition of N sources such as BEBPs [73, 74]. Moreover, a significant positive correlation ($r=0.57$, $p<0.05$) obtained between SOC and plant height indicates that higher buildup of SOC helped plant in attaining a better height. Among BEBPs, the height obtained under the treatment of ICD_W was maximum and significantly different from rest of the treatments. Higher leaf area obtained under this treatment may have helped in better utilization of nutrient resulting in better plant height, as there are reports that leaf area is a determinant factor in radiation interception and photosynthesis directly affecting biomass accumulation [75]. Therefore, it is an important parameter in determining plant growth and productivity. This may be further supported by a significant positive correlation as observed between leaf area ($r=0.59$,

Table 6 Effect of BEBP treatment on vegetative growth (plant height, leaf area, leaf area index, plant total dry matter, maximum root length, root dry weight, root:shoot ratio) of maize

Treatments	Plant height, cm	Leaf area, cm^2	Leaf area index	Total dry matter, g pot^{-1}	Max root length, cm	Root dry weight, g	Root:shoot ratio at harvest
CD_W	182.24 ± 1.91^{de}	501.14 ± 8.51^{e}	0.71 ± 0.01^{fg}	143.95 ± 0.44^{ef}	28.4 ± 4.06^{a}	17.07 ± 4.31^{bc}	0.13
ICD_W	210.82 ± 5.08^{g}	536.64 ± 4.27^{g}	0.76 ± 0.00^{i}	138.86 ± 12.70^{cdef}	34.3 ± 2.08^{c}	28.24 ± 7.63^{d}	0.25
RGC_W	170.39 ± 7.02^{bc}	481.54 ± 13.52^{cd}	0.68 ± 0.00^{cd}	124.08 ± 4.60^{ab}	27.9 ± 4.58^{a}	17.69 ± 2.35^{bc}	0.16
Mean	187.81	506.44	0.72	135.63	30.2	21	
CD_S	184.57 ± 9.36^{de}	482.92 ± 9.21^{cd}	0.68 ± 0.01^{de}	144.02 ± 3.76^{ef}	27.6 ± 0.58^{a}	11.31 ± 2.09^{a}	0.08
ICD_S	187.37 ± 8.03^{def}	492.75 ± 1.07^{de}	0.66 ± 0.00^{bc}	124.48 ± 2.93^{abcd}	29.2 ± 2.25^{a}	19.33 ± 2.41^{c}	0.18
RGC_S	161.92 ± 0.64^{ab}	475.33 ± 5.71^{bc}	0.67 ± 0.00^{cd}	141.18 ± 5.66^{def}	27.7 ± 3.15^{b}	16.48 ± 4.17^{bc}	0.13
Mean	177.95	483.66	0.67	136.56	28.16	15.70	
CD_L	189.23 ± 12.70^{def}	432.29 ± 3.30^{a}	0.62 ± 0.02^{a}	120.12 ± 13.32^{ab}	28.8 ± 0.79^{a}	12.53 ± 0.85^{ab}	0.14
ICD_L	186.23 ± 7.17^{de}	476.15 ± 0.39^{bcd}	0.65 ± 0.00^{b}	113.45 ± 7.62^{a}	31.8 ± 0.58^{a}	28.61 ± 3.80^{d}	0.33
RGC_L	182.52 ± 0.95^{de}	492.77 ± 8.95^{de}	0.70 ± 0.01^{ef}	129.74 ± 11.44^{abcd}	30.3 ± 5.81^{a}	17.22 ± 0.28^{bc}	0.15
Mean	185.99	467.07	0.66	121.10	29.96	19.45	
CD_A	184.57 ± 6.99^{de}	509.26 ± 14.08^{ef}	0.72 ± 0.00^{gh}	133.44 ± 4.58^{bcde}	29.8 ± 1.95^{a}	16.69 ± 1.81^{bc}	0.14
ICD_A	196.85 ± 2.54^{f}	501.03 ± 4.43^{e}	0.71 ± 0.00^{fg}	131.29 ± 1.75^{bcde}	29.0 ± 2.58^{a}	19.26 ± 2.92^{c}	0.17
RGC_A	167.00 ± 0.64^{bc}	461.47 ± 6.07^{b}	0.65 ± 0.00^{b}	124.32 ± 7.93^{abc}	30.0 ± 3.17^{a}	17.53 ± 5.86^{bc}	0.13
Mean	182.80	490.58	0.69	129.68	29.60	17.82	
RH_C	181.61 ± 3.36^{de}	519.90 ± 7.50^{f}	0.73 ± 0.01^{h}	115.66 ± 5.27^{ab}	27.5 ± 0.58^{a}	14.70 ± 0.59^{abc}	0.14
NPK	189.86 ± 1.91^{ef}	474.45 ± 11.75^{bc}	0.67 ± 0.00^{bcd}	155.44 ± 5.18^{f}	30.0 ± 0.61^{a}	20.43 ± 3.50^{c}	0.15
C	155.73 ± 4.24^{a}	433.21 ± 18.75^{a}	0.61 ± 0.00^{a}	108.35 ± 17.32^{a}	31.2 ± 0.82^{a}	17.99 ± 0.07^{c}	0.19
Level of significance	**	**	**	**	NS	**	**

Different lowercase letters in same columns indicate significantly different means at $p \leq 0.05$ between the treatments

NS not significant

**Significant at $p < 0.01$ level

$p < 0.01$) and leaf area index ($r = 0.57$, $p < 0.05$) with plant height.

Across the treatments, maize total dry matter yield varied with respect to both the origin and phase of BEBP (Table 6). Among the BEBP treatments, ICD_L (113 ± 7.6 g pot^{-1}) and both CD_S and CD_W digestates (144 ± 3.7 g pot^{-1}) produced maize with lowest and highest dry matter content, respectively, whereas it was significantly higher under NPK (155 ± 5.1 g pot^{-1}). Similar finding on maize was reported by Bachmann et al. [76] which was attributed to efficient utilization of the recommended dose of fertilizer. However, CD_W, CD_S, ICD_W and RGC_S also increased maize dry matter yields to the same extent as the mineral NPK fertilization with no statistical difference. RH_C application resulted in the production of dry matter statistically at par with that of control (0.93% higher over control). External addition of N in the form of BEBP have increased availability of soil available N and SOC as discussed before, along with increase in soil available fraction for P and K, which in turn may have helped for enhanced meristematic growth and higher accumulation of photosynthates from increased leaf area resulting in increased dry matter as compared to control treatment [77]. Increased dry matter could also be related to improved organic carbon of soil as evidenced by their significant positive correlation ($r = 0.72$, $p < 0.01$).

The effects of different treatments on root characteristics, viz., root length, root dry weight and root:shoot ratio, are presented in Table 6. ICD_W (34.3 ± 2.0 cm) digestates produced plants having longest (significantly different) root system. Again, both ICD_W and ICD_L treatments produced significantly higher root weight compared to rest of the treatments. Feedstock wise, gain of root mass was in the order ICD digestates (mean 23.8 g) > RGC digestates (mean 17.2 g) > CD digestates (mean 14.4 g) > RH_C (14.70 g).

Root length and proliferation depends on nutrient availability in the rhizosphere. It has been reported that, under nutrient stressed environment, plant has the ability to overcome these situations by extending root length further for acquisition of more nutrients from deeper layers of soil [78, 79]. In this context, with an exception to ICD_W and ICD_L, relatively lower root length observed under most of the BEBP treatments compared to that under control condition might be attributed to higher availability of nutrients and favourable soil conditions under these treatments. On the other hand, observation of significantly higher root length, root weight and root:shoot ratio under the treatment of ICD_W and ICD_L may be an indication of lower nutrient availability particularly soil available N in soil leading to higher root proliferation [80]. As discussed before, from the status of soil available N under various BEBP treatments, it was observed that soil available N, particularly under the treatment of ICDW and ICDL, was significantly low. Higher proliferation of root to soil under lower concentration of soil available N may be supported by statistically significant negative correlations observed between soil available N in post-harvest soil with maximum root length ($r = -0.63$, $p < 0.01$), root dry weight ($r = -0.53$, $p < 0.05$) and root:shoot ratio ($r = -0.55$, $p < 0.05$). An higher root:shoot ratio may lead to root redundancy, which impairs nutrient uptake affecting crop yield [81]. It was also evident from the study, as reflected by the significant inverse correlation between grain yield and root:shoot ratio, which is discussed in the next section.

Yield attributing parameters and grain yield

Data in Table 7 show that the longest ear was obtained from the application of CD_A (25.5 ± 1.02 cm) and under control condition, it was lowest (21 ± 0.20 cm). Data on thousand (1000) grain weight of maize revealed that grain weight was significantly affected by different BEBP treatments. Mean values indicated that maximum thousand grain weight was observed under CDs (303 ± 13 g) treatment and lowest in ICD_L (227 ± 19 g). Our study clearly demonstrated that 1000 grain weight has a significant positive correlation with grain yield ($r = 0.63$, $p < 0.05$).

Table 7 Effect of BEBP treatment on grain yield and yield attributing parameters (ear length and thousand grain weight) of maize

Treatments	Ear length, cm	1000 kernel wt, g	Grain weight
CD_W	22.2 ± 0.44^{a}	289.32 ± 55.07^{cdef}	20.28 ± 2.06^{cd}
ICD_W	23.0 ± 0.61^{ab}	255.38 ± 4.64^{abcd}	10.81 ± 0.53^{a}
RGC_W	21.5 ± 1.42^{a}	300.45 ± 5.78^{f}	19.88 ± 0.57^{cd}
Mean	23.20	281.71	16.99
CD_S	22.1 ± 0.60^{a}	303.54 ± 13.29^{f}	19.22 ± 1.86^{cd}
ICD_S	23.3 ± 1.63^{ab}	278.42 ± 14.12^{bcdef}	15.85 ± 0.65^{b}
RGC_S	21.3 ± 0.61^{a}	293.90 ± 13.23^{ef}	20.74 ± 2.01^{d}
Mean	22.73	291.62	18.60
CD_L	23.0 ± 0.81^{ab}	253.96 ± 20.13^{abc}	14.65 ± 2.43^{b}
ICD_L	22.8 ± 0.61^{ab}	227.28 ± 19.65^{a}	11.11 ± 1.17^{a}
RGC_L	22.3 ± 0.40^{a}	248.11 ± 2.03^{ab}	20.16 ± 1.45^{cd}
Mean	21.65	243.11	15.30
CD_A	25.5 ± 1.02^{b}	251.15 ± 6.25^{abc}	17.03 ± 0.81^{bc}
ICD_A	21.8 ± 0.61^{a}	244.46 ± 3.97^{ab}	14.82 ± 0.56^{b}
RGC_A	21.5 ± 0.20^{a}	292.22 ± 14.43^{def}	15.84 ± 0.79^{b}
Mean	21.65	262.61	15.89
RH_C	21.3 ± 1.22^{a}	261.07 ± 33.51^{abcde}	17.51 ± 1.43^{bcd}
NPK	25.3 ± 0.61^{b}	273.25 ± 16.10^{bcdef}	24.5 ± 4.40^{e}
C	21.0 ± 0.20^{a}	221.19 ± 14.62^{ab}	11.54 ± 0.84^{a}
Level of significance	*	**	**

Results on grain yield (Table 7) revealed that the application of the BEBP as a source of N stimulated the growth of maize over control in all the treatments except ICD_W and ICD_L. Among the BEBP treatments, highest grain yield was obtained under RGC_S (20.74 ± 2.0 g pot^{-1}) treatment, which was marginally higher than CD_W (20.28 ± 1.0 g pot^{-1}), RGC_L (20.16 ± 1.4 g pot^{-1}), RGC_W (19.87 ± 0.5 g pot^{-1}) and CD_S (19.22 ± 1.8 g pot^{-1}) with no statistical difference amongst them. In general, across all treatments, the highest and significantly different yield was found under NPK (24.50 ± 4.4 g pot^{-1}). The significantly higher yield under NPK treatment may be explained by the fact that, being readily available source of nutrients, it was taken up by crops immediately after application, whereas availability of nutrients from BEBP depends on the extent of rate of mineralization to transform into plant-available forms [82, 83]. Among BEBPs, averaged over type of feedstock, RGC digestates gave the maximum yield (mean 18.66 g pot^{-1}), ICD the lowest (mean 13.14 g pot^{-1}) and that of CD was intermediate (mean 17.79 g pot^{-1}).

Increased grain yield under RGC treatment may have resulted due to improvement in soil fertility in terms of both available N (23% over control) and P (81% over control), organic carbon (25% over control) and soil pH (0.20 unit over control). This indicates that the higher amount of nutrient, high pH and organic carbon content of under this treatment provided congenial soil environment to overcome the soil acidity-related constraints for crop production and contributed significantly towards increase in grain yield of the crop. To further support this, the grain yield showed significant positive correlation with available fractions of soil N ($r = 0.64$, $p < 0.05$) and soil pH ($r = 0.80$, $p < 0.01$) (Fig. 4a, b).

It has been reported that maize takes up N rapidly at the middle vegetative growth period and maximum rate of N uptake occurs near silking stage and uptake continues until near maturity [84, 85]. Availability of N in soil is important during grain filling stage, as 60% of grain N is contributed by the above-ground parts of the plant and the rest 40% comes from soils and roots [86]. In our experiment we found lower grain yield under ICD_W and ICD_L treatments as compared to control, though with no statistically significant difference. Though under these two treatments a better vegetative growth was achieved in terms of height and leaf area (discussed before) the same was not subsequently reflected in yield. This may be explained by the fact that ICD_W and ICD_L were the ones with maximum mineralized N and higher pH,

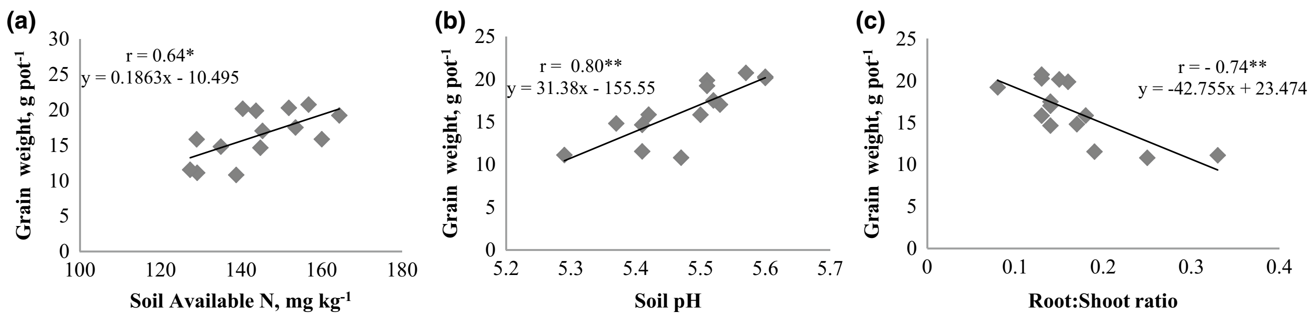

Fig. 4 Relationship of grain yield with **a** soil available N, **b** soil pH and **c** root:shoot ratio

which might have enhanced the vegetative growth through its immediate uptake. But, since the temperature during the experimental period was high (22–37 °C) there was a possibility of subsequent volatilization loss of N from soil [87, 88]. Volatilization loss from the application of high-pH digestate was also indicated by Formowitz and Fritz [89]. Volatilization loss under ICD_L and ICD_W treatment may have created N insufficiency during grain filling stage of the crop development ultimately affecting crop yield. Possible N loss under ICD_L and ICD_W treatments may also be supported by the lower accumulation of available N in post-harvest soil under these treatments (Table 5). It was also evident by significant inverse correlation observed between grain yield and root:shoot ratio ($r = -0.66$, $p < 0.01$) (Fig. 4c). Overall, these results suggests that, in case of application of BEBP having higher mineral N or higher pH, where there is possibility of nutrient loss in relation to atmospheric condition, measures should be taken through appropriate timing and method of application (split dosing or lower injection of fertilizer to soil) so as to obtain the maximum benefit. On the other hand, under RGC treatment, mineralization was slow and steady, releasing nutrients as and when required during grain filling giving better yield in all phases.

Biomass distribution in maize

Distribution of biomass in different plant parts, viz. aboveground parts (excluding cob weight), cob and root of maize, is shown in Fig. 5. There was variation in allocation of biomass with respect to digestate feedstock. For all digestates, the share of aboveground biomass in total biomass weight showed a narrow range of 66–67% (mean of four application option of each digestate). However, in case of distribution of root weight and cob weight, RGC and CD digestates showed a similar result, each showing 10% and 22% distribution of total biomass weight in root and cob, respectively. On the other hand, higher distribution of biomass in roots (15%) and lower in cob (18%) was observed under application of ICD digestates as compared to that under other BEBPs. Optimal partitioning theory predicts that, in response to a resource

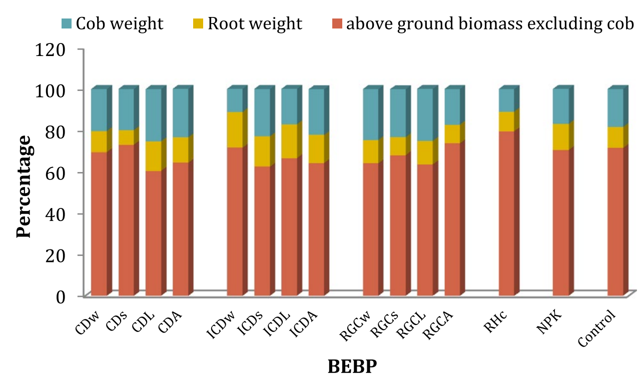

Fig. 5 Distribution of maize biomass among cob, root and aboveground biomass fraction

gradient, plants will optimize overall growth rate by making all resources equally limiting and adjust their biomass partitioning patterns to obtain the most limiting resource [90, 91]. It has been reported that plants that encounter limited nutrient or water supply are expected to partition more biomass to their roots and less to their stems and leaves [92]. Supporting our previous discussion, higher biomass allocation into root under ICD based digestate treatments, compared to that under RGC and CD treatments, indicates lower nutrient availability in soil under ICD based digestates treatments.

Crop nutrient (N, P, K) uptake, grain protein concentration and nitrogen harvest index of maize

Overall, the effect of BEBP application was found to have a statistically significant effect on N, P and K uptake (Table 8). Across all treatments, performance of NPK in enhancing NPK uptake was significantly superior over BEBPs and it remained lowest for control treatment. Among BEBPs, N uptake of maize in ICD_W (1.99 ± 0.16 g pot^{-1}) treatment was enhanced to the maximum. Barring two exceptions, i.e., RH_C and CD_W, P uptake in control treatment was significantly lower and it was maximum in CD_A by 87%. For K, uptake was found to be the highest and significantly different

Table 8 Total nutrient (N, P, and K) uptake, grain protein concentration and nitrogen harvest index in maize

Treatments	Crop total N uptake, g pot^{-1}	Crop total P uptake, g pot^{-1}	Crop total K uptake, g pot^{-1}	Grain protein, %	Nitrogen harvest index
CD_W	1.46 ± 0.12^{cde}	0.18 ± 0.00^{a}	0.70 ± 0.06^{a}	11.30 ± 2.59^{a}	0.24 ± 0.04^{def}
ICD_W	1.99 ± 0.16^{f}	0.25 ± 0.00^{cde}	1.36 ± 0.09^{e}	11.50 ± 2.30^{d}	0.10 ± 0.02^{a}
RGC_W	1.20 ± 0.06^{ab}	0.25 ± 0.01^{cde}	0.74 ± 0.10^{a}	12.60 ± 0.62^{cd}	0.33 ± 0.03^{g}
Mean	1.52	0.22	0.93	11.59	0.22
CD_S	1.75 ± 0.23^{e}	0.22 ± 0.02^{bc}	0.94 ± 0.10^{bc}	10.74 ± 0.21^{ab}	0.21 ± 0.04^{cde}
ICD_S	1.45 ± 0.02^{cde}	0.28 ± 0.01^{def}	1.14 ± 0.02^{d}	11.08 ± 0.85^{abc}	0.19 ± 0.02^{bcde}
RGC_S	1.32 ± 0.19^{bcd}	0.27 ± 0.01^{cde}	0.83 ± 0.02^{ab}	11.69 ± 1.88^{ab}	0.26 ± 0.06^{ef}
Mean	1.49	0.26	0.97	10.76	0.22
CD_L	1.29 ± 0.24^{bc}	0.22 ± 0.03^{bc}	0.70 ± 0.03^{a}	12.53 ± 1.11^{d}	0.23 ± 0.06^{cdef}
ICD_L	1.63 ± 0.13^{de}	0.25 ± 0.01^{cdef}	0.77 ± 0.02^{a}	12.30 ± 0.79^{d}	0.13 ± 0.01^{ab}
RGC_L	1.53 ± 0.23^{bcd}	0.25 ± 0.01^{de}	0.81 ± 0.19^{ab}	11.14 ± 0.84^{ab}	0.24 ± 0.02^{cdef}
Mean	1.42	0.24	0.76	11.98	0.20
CD_A	1.89 ± 0.06^{f}	0.30 ± 0.02^{f}	1.03 ± 0.06^{cd}	13.02 ± 1.25^{d}	0.18 ± 0.02^{bcd}
ICD_A	1.60 ± 0.27^{cde}	0.27 ± 0.03^{def}	1.09 ± 0.03^{d}	13.24 ± 1.40^{d}	0.19 ± 0.00^{bcde}
RGC_A	1.50 ± 0.11^{bcd}	0.25 ± 0.02^{cdef}	1.65 ± 0.03^{f}	10.58 ± 0.96^{a}	0.17 ± 0.02^{bc}
Mean	1.57	0.27	1.26	12.09	0.18
RH_C	1.11 ± 0.02^{a}	0.20 ± 0.03^{ab}	1.18 ± 0.09^{d}	11.06 ± 0.89^{bcd}	0.27 ± 0.01^{fg}
NPK	2.17 ± 0.07^{g}	0.29 ± 0.00^{ef}	1.09 ± 0.03^{d}	12.51 ± 1.88^{bcd}	0.22 ± 0.02^{cdef}
C	0.99 ± 0.02^{a}	0.16 ± 0.00^{a}	0.70 ± 0.07^{a}	10.79 ± 0.37^{ab}	0.18 ± 0.04^{bc}
Level of significance	**	**	**	NS	**

Different lowercase letters in same columns indicate significantly different means at $p \leq 0.05$ between the treatments. **Significant at $p < 0.01$ level, NS: not significant

in soil treated with ICD_W (1.36 ± 0.09 g pot^{-1}). Better uptake of nutrient under BEBP treatments particularly the digestates compared to control may be explained by lower nutrient immobilization in microbial biomass since less organic C is available for microbial growth through their application [93]. Higher uptake of N under ICD digestates can be related to higher biomass production under these treatments as seen from maize vegetative growth. Further to support this, N uptake of maize crop showed significant positive relationship with both plant height ($r = 0.59$, $p < 0.05$) and dry matter ($r = 0.72$, $p < 0.01$). Similarly, the protein content of maize grain grown with BEBP treatment clearly showed the superiority of the BEBP over control in improving protein content [94]. It can be assumed that BEBP addition increases the amount of accessible N in the plant and contributes to increase the qualitative yield in terms of N-associated components such as protein.

N harvest index (NHI) is used to describe the accumulation and redistribution of assimilates which shows the efficiency of crop to convert the fraction of photosynthates into grain yield (Table 8) [95]. Highest NHI was observed in RGC_W (0.33 ± 0.03) followed by RH_C (0.27 ± 0.01) and RGC $_S$ (0.26 ± 0.06), all showing significantly higher NHI than that under control (0.18 ± 0.04). Higher NHI under RGC can be related to higher grain production under these treatments. Lowest NHI under ICD digestates indicates lower translocation of N to grain and hence lower biomass partitioning to grain production. In our experiment, we found a significant negative correlation between grain N uptake and stover N uptake ($r = -0.51$, $p < 0.05$). Efficiency of N utilization depends on the timing of the plant demand with nutrient availability [96, 97]. Hence, it may be possible that, particularly under ICD treatments having higher ammonia-N due to mismatch of demand and availability, the plant prioritizes one component (vegetative growth) over the other during its development.

Conclusions

Our results clearly demonstrated that the fertilizing properties of the by-products (BEBP) are dependent on type of feedstocks as well as application phase of by-products. The application of BEBPs to soil significantly altered the pH, EC, available N, P and K, organic carbon and concentration of micronutrients in soil. Application of BEBP resulted in significant increase in grain yield as compared to that under untreated control which could be related to improvement in soil fertility (available fraction of N, P and K, organic carbon, pH and micronutrients) as well as crop physical growth. Conservation of nutrients, particularly nitrogen of high mineralized N containing by-products against volatilization loss should be ensured through appropriate timing and method of application. The outcomes are expected to stimulate the growth of integrated production of bioenergy and by-product-based organic fertilizer. However, future study is recommended considering agricultural application of bioenergy by-product at field scale, its long-term effects on different test crops and techno-economic assessment of by-product processing.

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