Overview on agricultural potentials of biogas slurry (BGS): applications, challenges, and solutions

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Abstract

The residual slurry obtained from the anaerobic digestion (AD) of biogas feed substrates such as livestock dung is known as BGS. BGS is a rich source of nutrients and bioactive compounds having an important role in establishing diverse microbial communities, accelerating nutrient use efficiency, and promoting overall soil and plant health management. However, challenges such as lower C/N transformation rates, ammonia volatilization, high pH, and bulkiness limit their extensive applications.

Here we review the strategies of BGS valorization through microbial and organomineral amendments. Such cohesive approaches can serve dual purposes viz. green organic inputs for sustainable agriculture practices and value addition of biomass waste. The literature survey has been conducted to identify the knowledge gaps and critically analyze the latest technological interventions to upgrade the BGS for potential applications in agriculture fields.

The major points are as follows: (1) Bio/nanotechnology-inspired approaches could serve as a constructive platform for integrating BGS with other organic materials to exploit microbial diversity dynamics through multi-substrate interactions. (2) Advancements in next-generation sequencing (NGS) pave an ideal pathway to study the complex microflora and translate the potential information into bioprospecting of BGS to ameliorate existing bio-fertilizer formulations. (3) Nanoparticles (NPs) have the potential to establish a link between syntrophic bacteria and methanogens through direct interspecies electron transfer and thereby contribute towards improved efficiency of AD. (4) Developments in techniques of nutrient recovery from the BGS facilities' negative GHGs emissions and energy-efficient models for nitrogen removal. (5) Possibilities of formulating low-cost substrates for mass-multiplication of beneficial microbes, bioprospecting of such microbes to produce bioactive compounds of anti-phytopathogenic activities, and developing BGS-inspired biofertilizer formulations integrating NPs, microbial inoculants, and deoiled seed cakes have been examined.

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Abbreviations

AAOB	Ammonia-oxidizing bacteria
ACD	Anaerobic co-digestion
ACE	Abundance-based coverage estimators
ACM	Acetoclastic methanogens
AD	Anaerobic digestion
ADDC	Agricultural digestate dry condition
ADG	Anaerobic digester
AIM	Agriculturally important microbes
ANOB	Anaerobic-oxidizing bacteria
AQS	Aqueous solubility
AR-W	Agricultural residue waste
ATP	Adenosine triphosphate
BCL	BGS cultivated lands
BD	Buffalo dung

BGS	Biogas slurry/bioslurry	NEDOCs	Non-edible deoiled cakes
BGSY	Biogas yield	NGS	Next-generation sequencing
BSM	Biostimulation	NOB	Nitrite-oxidizing bacteria
C/N	Carbon-to-nitrogen ratio	NPs	Nanoparticles
CAGR	Compound annual growth rate	nZVI	Nanosized zero-valent iron
CANON	Completely autotrophic nitrogen removal over nitrite process	OTUs	Operational taxonomic units
CBY	Cumulated biogas yields	PB	Plant biomass
CCL	Chemically cultivated lands	PSB	Phosphate-solubilizing bacteria
CD	Cow dung	QIIME	Quantitative insights into microbial ecology
CFs	Chemical fertilizers	SBAD	Substrate for anaerobic digestion
CM	Cattle manure	SC	Slurry compost
CMM	Cellular membrane	SI	Shannon index
CNPs	Conductive nanoparticles	SNA	Syntrophic acetogens
CS	Cattle slurry	SOC	Soil organic carbon
DBS	Digested biogas slurry	SPB	Secondary fermenting bacteria
DIET	Direct interspecies electron transfer	TAN	Total ammonia nitrogen
DOCs	Deoiled cakes	TKN	Total Kjeldahl nitrogen
DOM	Dissolved organic matter	TNC	Total nutrient content
DP	Dominant phylum	TS	Total solids
DPA	Dipicolinic acid	USD	United States dollar
DVM	Digestate with vermiculite	UTC	Coordinated universal time
EET	Extracellular electron transfer	V-GPM	Vacuum-assisted gas-permeable membrane
EU	European Union	VMD	Vacuum membrane distillation
FAO	Food and Agriculture Organization	VOA	Volatilization of ammonia
FYM	Farm yard manure	VS	Volatile solids
GD	Goat dung		
GHGs	Greenhouse gases		
HACs	High-ammonia conditions		
HED	Hen droppings		
HGM	Hydrogenotrophic methanogens		
HM	Horse manure		
HOD	Horse dung		
HRT	Hydraulic retention time		
IDR	Indonesian rupiah		
IHT	Interspecies H_2 transfer		
INR	Indian rupees		
J-DOC	Jatropha deoiled cake		
K-DOC	Karanja deoiled cake		
LCA	Life cycle assessment		
LFD	Liquid fraction of the digestate		
LR	Laser irradiation		
LSS	Livestock slurry		
MBRs	Metal-binding receptors		
MCNs	Micronutrients		
Mg ha^{-1}	Mega gram per hectare		
MIET	Mediated interspecies electron transfer		
MME	Microbial metabolic efficiency		
MMTs	Million metric tons		
MNPs	Magnetite nanoparticles		
MT	Million ton		
NAR	Nijhuis ammonium recovery		
NBS	Nutrient subsidy rates		

1 Introduction

1.1 Background of the study

The prevalence of various types of abiotic and biotic stress leads to a decline in crop productivity and consequently accelerates the application of different types of chemical fertilizers and pesticides in agriculture fields. Such chemical inputs not only impose a threat to human life, but environmental consequences are more pronounced. The application of nitrogenous chemical fertilizers has three important concerns, i.e., increasing cost (due to hikes in petroleum prices); economic loss (over 50% of applied nitrogenous fertilizers lost in different agriculture processing); and environmental hazards [1]. There is a gap of about 10 million ton (MTs) of plant nutrients between removal by crops and replenishment through fertilizers, presently in India [1]. Such figures necessitate transformation in our ideology from conventional practices to environmental-friendly approaches, promoting minimal or optimum use of chemical fertilizers or encouraging organic farming.

Organic farming as a sustainable agriculture practice has gained global attention in the present-day scenario; however, certain challenges to widen the scope of such practices need to be addressed. As per the data published in "The World of Organic Agriculture (Statistics and emerging trends 2020)"

(FiBL survey 2020), there are 186 countries involved in organic activities in the year 2018 with 71.5 million hectares of land covered under organic agriculture. However, as far as the worldwide organic producers are concerned, India (1,149,371) (2018 data) has maximum numbers followed by Uganda (210,352) (2016 data) and Ethiopia (203,602) (2015 data) (FiBL Survey 2020) [2]. The USA is the worldwide leader of the organic market followed by Germany and France with retail sales of 40559, 10919, and 9139 million euros respectively, while in terms of per capita consumption Denmark and Switzerland top the chart (FiBL-AMI survey 2020, 2018 data) [2]. Besides offering numerous benefits to the environment, issues like low productivity and higher produce cost are some of the key challenges that limit the interest of organic producers at large. A survey study by Ramesh et al. [3] also put forward similar concerns.

The amendments in biological resources (principally those considered as biomass waste or byproducts) to develop organomineral biofertilizers through the implementation of technological interventions such as microbiology, biotechnology, and nanotechnology are the key areas that must be looked upon to further strengthen organic agriculture practices. Biogas slurry/bioslurry (BGS) is one such residual biomass, which hosts a diverse microbial population, and accumulates secondary metabolites, carrying essential micro/macro-nutrients. Cow dung (CD), one of the common biogas feedstock and BGS source, is rich in mineral elements like N, P, Ca, Zn, Mg, S, Fe, Cu, Co, and Mn [4] and harbors diverse microbial population [5]. The full potential of CD in areas such as agriculture, energy, environment, and human health is still unexplored [5]. BGS demonstrates potential

applications as a soil conditioner, fertilizer, and bio-pesticide in agriculture. The detailed discussion over different applications of BGS in crop fields is presented in Section 2.2. Taking account of shortcomings, it has been observed that BGS alone is unable to fulfill the entire nutrient demand due to its bulkiness, N loss as ammonia volatilization, reduced rates of C/N transformation, etc. [6–8]. The major limitations and emerging challenges for BGS applications in the agriculture fields have been outlined in the next section.

1.2 Limitations of BGS: need for value addition

BGS obtained from animal dung such as cattle and livestock have nutrient profile contributed as a potential source of fertigation in agriculture fields. However, BGS has various limitations such as bulkiness, low C/N ratio, nitrogen loss through volatilization of ammonia (VOA), and high pH (Fig. 1). A biogas plant with 800 m^3 of working volume is estimated to be discharged 15 tons of BGS per day [9]; hence, generated volumes of BGS need to be managed appropriately to avoid any environmental consequences, as a result thereof. The bulkiness in BGS is due to the high water content (~93%) in it [10–12], which further complicates its transport and utilization [13]. The bulkiness of BGS also limits its potential to fulfill the entire nutrient demand [7] in agriculture fields.

Loss of nitrogen as VOA from liquid slurry depends upon different physical (liquid temperature, viscosity, contents, and distribution of particle size in solids), chemical (pH, ionic strength, and total ammonical nitrogen), and environmental (temperature, humidity, direction, and speed of the

Fig. 1 Limitations of BGS as a potential nutrient input in agriculture fields (contamination include pathogens, heavy metals, organic pollutants, pesticides, antibiotics, etc.) (conceptualized from [6–8, 21])
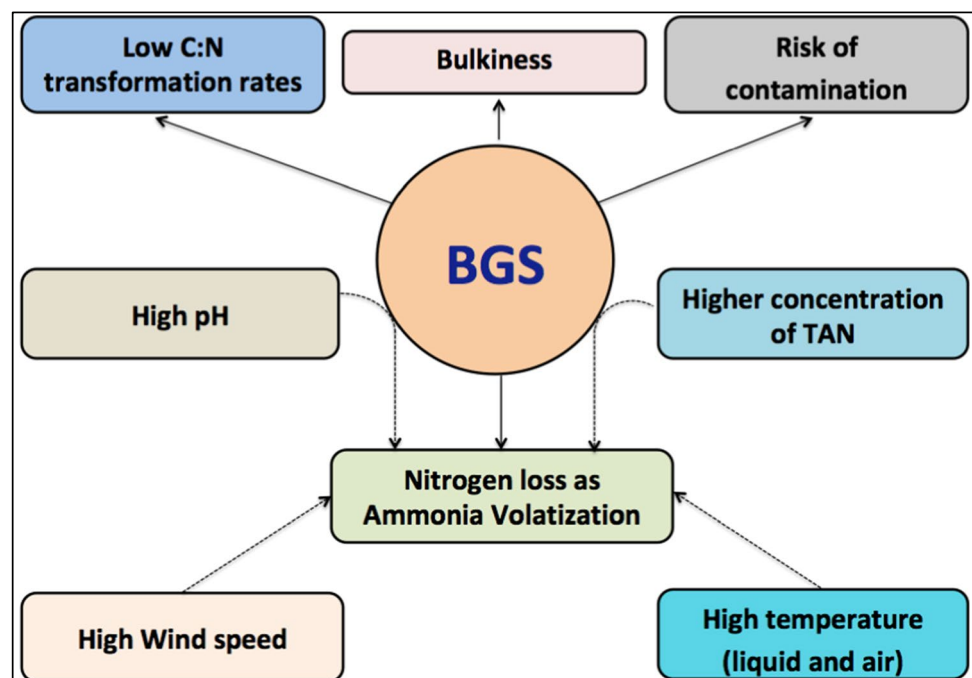
wind, etc.) factors [8]. Such losses were also observed at the time of drying BGS or soon after its application in agriculture fields [14]. BGS have pH values usually in the alkaline range [11, 14, 15], and as per the reports, the higher pH (>7) promotes N loss through VOA [16].

Nitrogen loss from BGS in the form of VOA imposes complexity during its handling, storage, and field applications [14]. Storage limitations such as lack of bottom liners or impermeable surfaces can have the risk of nitrogen and potassium leaching [14]. Under circumstances where the proper covering of recovered BGS is not available, the risk of ammonia emissions can further increase [14]. For convenient handling, BGS often dried or composted, which can also end up with N losses as VOA [14]. However, such losses can be minimized with the use of nitrogen-rich organic substrates during composting of BGS. High air temperature can also increase the N loss through ammonia emissions [16]. Studies mentioned that the available nitrogen in BGS is comparatively less efficient than mineral fertilizers due to such N losses [17]. The ammonia deposition as a consequence of increasing VOA is reportedly associated with the risk of acidification in sensitive ecosystems [18], besides this, ammonia also act as precursor for secondary aerosols production [19].

Zheng et al. [6] reported low C/N transformation rates in biogas digestate obtained from farm manure (pig manure and urine). The low carbon and high nitrogen contents in anaerobic digestates [14] are primarily attributed to the conversion of carbon-rich substrate into methane. Studies reported mixed responses (i.e., positive, negative as well as no effects) of N availability on mineralization of soil organic carbon (SOC) [20]. As cited by Insam et al. [21] and Stumpe et al. [22] and mentioned in the study of Senbayram et al. [23], the elevated levels of nitrogen in digestates can increase the mineralization of carbon (called as priming effect). High and low priming effects were reportedly observed under lower (higher C/N ratio) and higher N availability (low C/N ratio) respectively [24]. Liu et al. [24] reported higher SOC stocks and lower respiration activity of soil microbes under nitrogen deposition conditions. Researchers also speculate the role of soil microbial community in the degradation of BGS-derived recalcitrant organic carbon present in topsoil, hence, mounting the SOC stocks [25]. Liao et al. [20] mentioned that the decrease in priming effect under the conditions of higher nitrogen availability is majorly controlled by "microbial metabolic efficiency" (MME). Similar findings were reported earlier, and Chen et al. [26] also concluded that instead of microbial activity (extracellular enzymatic activities), the dynamics of carbon in topsoil under higher nitrogen availability is regulated by MME. As mentioned by Groot and Bogdanski [19], in spite of the higher mineralization of nitrogen at a low C/N ratio, the susceptibility towards emissions of ammonia increases.

To further explore this, we have reviewed the study of Senbayram et al. [23]. The application of organic matter in combination with ammonium sulfate initially increases the readily available SOC. Following the rapid decomposition of easily degradable carbon compounds, the ratio of nitrate nitrogen and available carbon increases, and once reached threshold levels, it may induce N_2O emissions under the conditions of high soil moisture [23].

The direct application of liquid BGS in agriculture fields (such as foliar spray) is also an important consideration. To avoid the toxicity risk on plant growth as a result of higher concentration of ammonia and soluble P, it has been recommended to dilute the liquid BGS before applying on standing crops based on the type of biodigester (like floating drum/fixed dome type; tubular/plug-flow type, etc.) [17]. The application of recovered digestates can also induce phytotoxicity effects precisely when used as a peat substitute in nursery/horticulture plantations, which more likely depends on the content of soluble salts in digestates [14].

The BGS concentration is another important aspect and as reported by Warnars and Oppenoorth [17]; it depends upon the absorption rate of plants at the time of slurry application. Niyungeko et al. [15] observed an increase in colloidal phosphorous leaching from paddy (rice) topsoil at medium (673 m^3 ha^{-1}: 165 kg N ha^{-1}, 52 kg P ha^{-1}) and high (1350 m^3 ha^{-1}: 330 kg N ha^{-1}, 104 kg P ha^{-1}) application rate of BGS. Warnars and Oppenoorth [17] recommended lower application doses of BGS to limit the N loss as nitrate leaching. Over-fertilization with BGS can risk the environment (VOA causes atmospheric pollution) and also incurred with financial losses (loss of fertilizer nitrogen) [19].

Reports indicated that the possibilities of pathogens and heavy metal contaminants in BGS are lesser in the case of using manure over other waste streams as feedstock material for AD [14]. However, it is noteworthy that the degradation of heavy metals does not occur during AD [27]. Hence, it is important to study the fate of heavy metal transfer on receiving environment as a consequence of BGS fertilization, more precisely the long-term application effects. Bian et al. [28] treated farmland soil with BGS (pig manure and straw as feedstock material for AD) and investigated the concentrations of heavy metals in different soil and plant samples. They observed higher levels of Cd, Pb, and Zn (mean concentrations) in soil amended with BGS (which exceeds standard safety limits as referred to in the study), and the plants were found to have heavy metals (under investigation) above the standard safety limits as referred to in the study (except Cu). Overall, leafy vegetables showed higher heavy metal uptake as compared to the non-leafy vegetables. Risk assessment through plant ingestion displayed higher "noncarcinogenic risk" with heavy metals As, Cd, and Pb; while the carcinogenic risk (CGR) was found to be associated with Cr>As>Cd (decreasing order), out of them, CGR through

food chain was primarily ascribed to Cd and As [28]. Tang et al. [27] found concentrations of heavy metals (Cd, Cu, Pb, and Zn) in soil and crop plants (Rice and Wheat) after 5 years following repeated fertilization with BGS (pig manure as feedstock material for AD) within the standard safety limits as laid down by different international bodies (China, EU, and FAO). In another interesting study by Lu et al. [29], the effect of BGS (called anaerobically digested slurry by the authors) (pig farm waste as feedstock material for AD) fertigation on rice grain yield and accumulation of heavy metals in harvested rice grains was investigated. They have recorded the heavy metal concentration in BGS as Cu>Cd>Pb>Cr>Hg (decreasing order), and observed levels of heavy metals in soil (before transplantation of rice and after harvesting) as well as in rice grains were found lower than the standard critical levels as referred to in the study, and the heavy metal concentration in harvested grains were independent of the dose of BGS application in fields.

In addition to this, it is also important that the BGS must be well-treated to be free from contaminations such as pathogens [17]. That could be of concern in scenarios where stringent measures (such as optimum retention time, pH, digester temperature [21]) during AD of feedstock materials will not be followed. Studies mentioned that managing such consistencies in operational conditions might be more challenging for developing countries [14]. Though the occurrence of pathogens in BGS is comparatively lesser than FYM, however, the risk cannot be ignored completely as the process of AD does not kill the entire range of pathogens [17]. In addition to pathogens and heavy metals, the presence of antibiotics and different organic pollutants (including pesticides) is some of the emerging challenges while using the BGS for agriculture applications [21]. Pointing to the existing challenges with the application of BGS as a rationale behind the present study and prioritizing the concept of sustainable agriculture practices, we have attempted to highlight the latest technological developments fostering the value addition of BGS.

2 BGS: source, composition, and agricultural applications

2.1 Source and composition

Biogas is comprised of methane (major component), carbon dioxide, nitrogen, hydrogen sulfide, hydrogen, and traces of ammonia [30]. Hydrolysis, acidogenesis, acetogenesis, and methanogenesis are the key steps of biogas production in which each step is catalyzed by the diverse microflora [31]. Different substrates for AD to produce biogas include animal (manure, dung, fodder residue, etc.), vegetable (grass, straw, etc.), and household (night soil, garbage, etc.) waste

[32]. Livestock dung (such as cattle manure) is the most commonly available substrate for biogas production [33], precisely in rural areas.

Discussing the Indian scenario, from 730 MT of dung produced by animals annually, bovine dung alone (cattle and buffalo dung) accounts for 256.2 MT from which estimated annual BGS production is 76.8 MT (at 1 kg cattle dung= ~0.3 kg BGS) [10]. In another study by Thiruselvi et al. [34], the amount of CD generated in India has been reported around 335 MT per month (out of which ~110 MT is considered to be waste during collection and transportation). Referring to Kumar et al. [10], we can estimate the monthly BGS recovery rates after AD of ~225 MT CD. Rath and Joshi [35] mentioned the estimated annual bioslurry recovery of 299 MT from ~995 MT of bovine dung. Such values direct the extent of digestate availability from the biogas plants. As per the case study conducted in Rajasthan (India) [36], the majority of biogas plant owners preferred to use recovered BGS as fertilizer. Considering this awareness among the farmers, the application of integrated approaches leading to the value addition of BGS to develop low-cost Bio-Agri inputs will be of great significance to further promote its full potential with anticipations of reducing our dependence on chemical fertilizers.

Biogas slurry or bioslurry abbreviated as BGS in the present study recovered as the spent from the AD of organic feeds during biogas production. The bulk of BGS is contributed by water (~93%) and the remaining part (~7%) is comprised of organic (4.5%) and inorganic (2.5%) matter [10–12]. pH value lies above the neutral range [37]. It is rich in NPK with other nutrients (Ca, Mg, Fe, Mn, Cu, and Zn) essential for plant growth [38]. One of the latest studies mentioned 2.55%, 0.57%, and 1.77% of N, P, and K respectively in BGS [33]. Sharma et al. [39] in sun-dried BGS reported C, N, P, and K content of 41.6±2.1%, 0.72±0.12%, 0.59±0.02%, and 0.91±0.04% respectively.

Mdlambuzi et al. [33] observed a direct relationship between the NPK content of cattle manure (CM) and recovered BGS, i.e., higher N, K, and lower P content. It is worth mentioning here that the nutritional status of recovered BGS relies upon several aspects such as the source of feedstock substrate, operational conditions in AD, and native microflora. Hence, it is important to optimize the substrate combination coupled with operational conditions during AD, which can yield twin benefits of higher biogas yields and nutrient-enriched BGS. Organic matter has a low C/N ratio, which limits the process of AD [40]. As the animal manures have low C/N content, it is ideal to perform co-digestion with the substrates having high carbon to low nitrogen content such as plant biomass. Co-digestion studies using plant biomass with livestock wastes such as CD [41, 42] and poultry litter [43] are well documented. This will facilitate C/N balance, favoring microbial breakdown of substrate mixture

in the digester [44]. Rodríguez-Abalde et al. [45] reported higher methane yields at a C/N ratio of 20–30. Wei et al. [46] optimized the combination of CM, maize straw, and sewage sludge (35:35:30) for higher methane yields. Priya et al. [47] performed co-digestion of plant biomass with organic wastes and found higher levels of volatile solids (VS) leading to improved levels of biogas yield.

Interestingly, the anaerobic digestate obtained from digestion (of feed substrate, i.e., CM, pig slurry, and poultry litter) or co-digestion (with onion waste) shows similarity in their structural characteristics, and the recovered co-digestate showed improvement in the availability of nutrients [48]. Concluding this, the C/N content is an important consideration while selecting substrates and optimizing their different combinations. The strategy of co-digestion with other feedstocks has also been reported to recuperate the process of AD [40]. We suggest further readings on the co-digestion of livestock manure with different feedstocks [31, 45, 49–51].

2.2 Agricultural applications

2.2.1 Is BGS a preferred source of fertilizer over raw manure or traditional composts?

Anaerobic digestates such as BGS comparatively offer low nitrate leaching and lesser nitrous oxide emissions [18]. Digestates from organic matter such as animal slurry reportedly contains more mineral nitrogen as compared to the undigested ones [52]. According to Aminul Haque [12], anaerobically decomposed BGS has a higher nutrient value than aerobically decomposed BGS. Smith et al. [53] observed higher nitrogen losses in composting (26–51%) over AD (5–10%). In comparison to traditional compost, BGS is an easily available form of compost [17] and is also reported to have stronger plant growth-promoting activities than raw slurries due to higher concentrations of ammonical nitrogen [54]. As compared to CD, BGS have lower C/N content, thus enabling higher fertilization potential, and anaerobic digestion aids mineralization effects, hence increasing the bioavailability of nutrients to the plants [16], though the susceptibility towards N loss due to VOA can be increased [19]. Tumuhimbise [55] recorded higher biofertilizer potential of CM bioslurry as compared to the sundried slurry, fresh CM, and NPK treatments on the overall growth of radish in ferralsol soil. On reviewing the literature, BGS appears as a preferred source of nutrients (like N) with significant fertilizer potential [17, 53–56]. Research and development in the area of BGS enrichment to meet the current nutrient demands in agriculture fields coupled with searching opportunities to develop effective bioformulations against a wide range of pests will be of great significance to meet the objectives of organic farming and sustainable development goals.

2.2.2 Nutrient profile of BGS

As compared to CM, Mdlambuzi et al. [33] found 0.65%, 0.133%, and 0.10% higher values of N, P, and K content (% wt) in BGS respectively. According to Devarenjan et al. [11], $2m^3$ of the biogas plant can produce around 50 kg of BGS every day and $1m^3$ of BGS contains about 0.16 to 1.05 kg of nitrogen which is equal to approximately 0.35 to 2.5 kg of urea. Animal urine added to BGS increases nitrogen levels that accelerate the compost-making process and optimize the C/N ratio in BGS that aids nutrient availability to soil biota and plants [10]. Jared et al. [38] recorded higher levels of Ca, Mg, Fe, Mn, and Zn in slurry compost as compared to BGS. However, from Fig. 2, it can be noticed that in terms of NPK values, BGS is preferred over slurry compost and farm yard manure (FYM). Incorporation of nutrient-rich organic amendments such as non-edible DOCs (NEDOCs) as a strategy to facilitate multi-substrate interaction of beneficial microbes and thereby to ameliorate micronutrient status of BGS has been proposed through the present review and will be discussed further upon.

2.2.3 BGS as an organic fertilizer

BGS offers various advantages as an organic fertilizer like improvement in soil nutrient profile and their availability to plants; improves soil structure, its water holding capacity, and cation exchange capacity; improves soil microbiota (nitrogen-fixing bacteria, phosphate-solubilizing bacteria (PSBs)); etc. [10]. Such organic amendments can further prevent or reduce soil erosion through improvements in the physical properties of soil (such as water-holding capacity [10], air permeability [6], aggregate stability, penetration resistance [57], etc.) along with an increase in soil organic carbon content [57]. BGS is a rich source of slow-release minerals [58] and provides nutrients in balanced proportions [59]. It can be utilized directly or indirectly as a fertilizer with other organic materials [10]. BGS can be applied as a foliar spray (onto crops), diluted liquid (onto roots), and dry composted form (combined with irrigation techniques to ensure sufficient water to crop plants) [37]. Zhao et al. [60] reported the preferential application of BGS in agriculture application as compost instead of its direct application. Referring to Table 1, it has been observed that the BGS have a potential role in plant growth–promoting activities such as overall yield and productivity, improving nutrient contents in soil, and maintaining soil structure, effectivity against soil-borne, and plant pathogens, etc. Beneficial effects of BGS fertilization leading to the overall soil and plant health management are represented schematically in Fig. 3.

Plant growth and productivity Ferralsol soil fertilized with CM bioslurry promotes plant height and total biomass
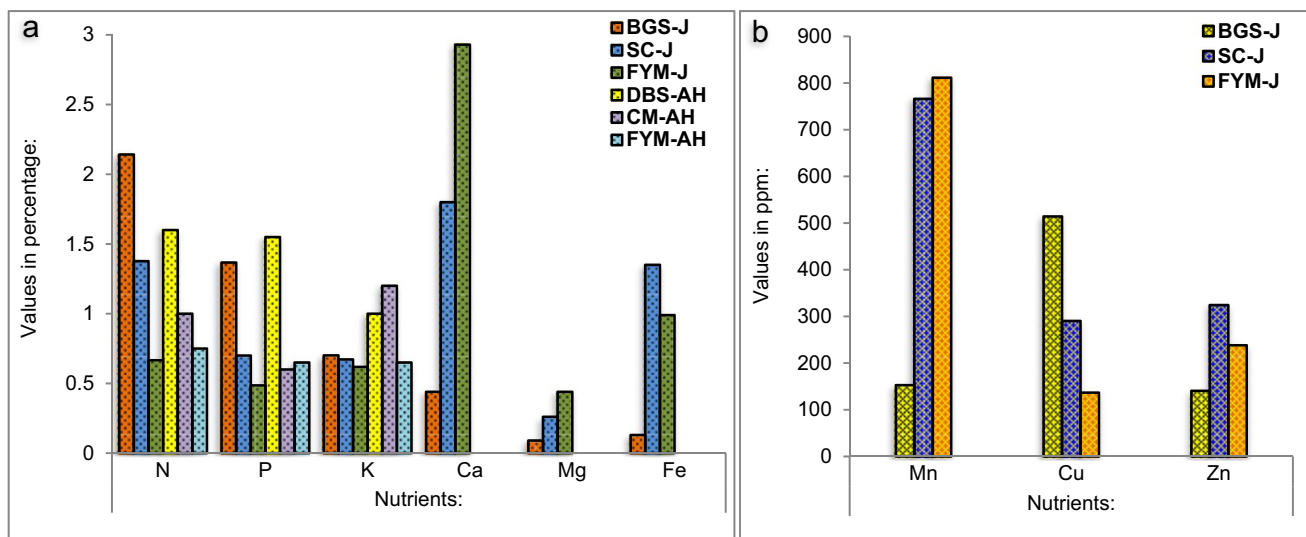
Fig. 2 Comparative analysis of nutrient composition in different organic inputs. **a** Macronutrients along with Fe; **b** micronutrients Mn, Cu, and Zn (note: DBS-AH, CM-AH, and FYM-AH represent average data from three different regions/sample matrices; BGS-J, SS-J, and FYM-J represent average data derived from the range of values provided; the values of Ca, Mg, and Fe were not reported in DBS-AH, CM-AH, and FYM-AH; DBS, digested biogas slurry; CM, compost manure; FYM, farm yard manure; BGS, biogas slurry; SC, slurry compost; J, reported by Jared et al. [38]; and AH, reported by Aminul Haque [12])

content, and improves growth parameters such as root diameter, and leave number and weight, in radish [55]. Xu et al. [61] recorded the highest yields in rice and rape at an application rate of 165.1 and 182.1 t ha^{-1} BGS respectively. They [61] have observed comparatively better results than those obtained from chemical fertilizers treatment. Besides this, You et al. [58] fertilized red clay soil with BGS (at 30 and 40 kg/plant/year) and noticed an improvement in fresh fruits yield, oil yield, and seed rate (fresh and dry) per plant in *Camellia oleifera*. Ferdous et al. [62] supplemented CD BGS with synthetic fertilizer (NPK, S, Zn, B) at an application rate of 5 t ha^{-1} and reported increasing tomato yields. Results of Xu et al. [37] showed an increase in overall growth, stomatal conductance, concentration of intercellular CO_2, water-use efficiency, rate of transpiration, photosynthesis, etc., in *Perilla frutescens* seedlings. Aminul Haque [12] reported a higher manure value of BGS in Rabi crops than Kharif crops and mentioned that nitrogen content was higher in air-dried BGS than sun-dried BGS.

Jothi et al. [63] noticed an increase in fruit number (3 fruits/plant) and yield of tomato (35.2 g/plant) with the application of BGS (obtained from the AD of a mixture containing CD and water at 1:1). Garg et al. [64] recovered the higher maize grain yields (6.21 Mg ha^{-1}) and improvement in root length density (1.10 cm cm^{-3}) with the treatment of BGS (at 15 Mg ha^{-1}). Zhao et al. [65] in laboratory investigations recorded the highest germination potential, germination rate, and germination index in *Vicia faba* (L.) seeds soaked in 75% BGS (w/v in distilled water). Du et al. [66] worked on Hoggery BGS and observed a substantial

increase in wheat (23.47%) and maize (15.46%) yields. Some of the studies such as Coelho et al. [67] demonstrate that the biofertilization potential of anaerobic digestates outweighs the effect of undigested cattle slurry.

Taking a brief account of soil aggregation properties, Zheng et al. [6] experimentally examined improvement in soil structure, fertility, and C/N distribution in red soil (Ultisol) treated with BGS (obtained from AD of pig manure and urine) and chemical fertilizer (0.45:0.55). The application of BGS can reduce the use of synthetic fertilizers up to 15–20% [10]. Lu et al. [29] found a reduction in the chemical application (NPK by 100, 100, and 26% respectively) and irrigation water use (by 45.5%) without affecting rice grain yields in the field experimental study. Xu et al. [37] observed variable concentrations of mineral elements in different organs of *P. frutescens*. Koszela and Lorencowicza [68] fertilized soil with BGS and reported higher N (3.66%), P (0.30%), K (1.96%), Ca (1.96%), and Mg (0.38%) content in *Alfalfa* leaves as compared to treatment that involved chemical fertilizer application. Yu et al. [69] with the application of concentrated BGS have seen improvements in the soil available NPK content while working on the tomato plant. In a similar study, Thomas et al. [70] found enhanced nitrogen and phosphorous uptake efficiency in Barley forage utilizing anaerobic beef cattle digestate (solid). In experiments of Coelho et al. [67] on ryegrass swards, soil treated with anaerobic co-digestate of food waste and pig slurry showed higher P (4.6 mg/L) than the soil treated with undigested CS (1.5 mg/L). They have also observed enhanced levels of nutrients such as B, Cu, Zn, and Mn.

Table 1 Agricultural applications of BGS obtained from different biodegradable feedstocks

| Treatment	Agriculture application	Observation	Reference
Ferralsol soil fertilized with CM bioslurry (fresh at 300 mL).	Improves overall growth in radish.	Increase in plant height, total biomass, and other parameters such as root diameter, leaves number, and weight.	Tumuhimbise [55]
Soil fertilized with BGS and CM (40 kg N ha^{-1}). BGS and CM were supplemented with straight fertilizer to achieve desired levels of phosphorus.	Higher grain yield with BGS treatment as compared to CM.	Equalizing the fertilization potential of BGS and CM can serve dual benefits, i.e., biogas production from CM followed by utilization of recovered BGS as a potential fertilizer.	Mdlambuzi [33]
BGS application rate 165.1[a] and 182.1[b] t ha^{-1}.	Higher yield of rice and rape.	Highestyield (in kg ha–1) of rice(8255.5±533.6)[a] and rape (3226.7±175.7)[b] as compared to other treatment doses, control, and chemical fertilizer applications.	Xu et al. [61]
Red clay soil fertilized with BGS (at 30 and 40 kg/plant/ year).	Improves fruit yield and seed rate in *Camellia oleifera* trees.	Increase in yield of fresh fruits, seed rate (fresh and dry), and oil yield per plant.	You et al. [58]
Soil treated with Hoggery BGS.	Improves wheat and maize yield.	Increase in wheat (23.47%), maize (15.46%) yield, starch, and crude protein content as compared to control (conventional fertilizer).	Du et al. [66]
Soil fertilized with BGS (AD of Pig manure and Urine) and Chemical fertilizer (0.45:0.55).	Soil aggregation properties and C/N distribution in red soil (Ultisol).	Improve soil structure and fertility.	Zheng et al. [6]
Soil amended with Solid digestate (AD of beef CM).	Improves barley forage yield and nutrient uptake efficiency.	Higher barley forage yield (31–50%) as compared to other treatments and control. Enhanced nitrogen and phosphorous uptake.	Thomas et al. [70]
Soil fertilized with digestate obtained from biogas plant.	Promotes higher nutrient content in *Alfalfa* leaves.	Higher N (3.66%), P (0.30%), K (1.96%), Ca (1.96%), and Mg (0.38%) content in *Alfalfa* leaves as compared to treatment involve chemical fertilizer application.	Koszela and Lorencowicza [68]
BGS at concentrations 0, 25, 50, 75, and 100% w/v in distilled water.	Improves seedling growth in *Vicia faba* (L.).	Seeds of *Vicia faba* (L.) soaked in 75% BGS for 4 h showed the highest germination potential, germination rate, and germination index.	Zhao et al. [65]
BGS at different concentrations (1:1, 1:2, and 1:4)	Promotes plant growth, nutrient levels, and water-use efficiency.	Promotes growth (root length, plant height, root and shoot biomass, etc.) in P.*frutescens* seedlings.	Xu et al. [37]
Concentrated BGS (livestock farm with cattle in majority over pigs).	Improves quality of tomato fruit.	Improves NPK content (soil available), the amino acid content in tomato fruit, and total microbial count (rhizospheric and non-rhizospheric soil).	Yu et al. [69]
BGS at 70 kg N ha^{-1}.	Improves nutrient content and biomass production in maize fodder (*Zea mays*).	Highest crude protein content (11.91%) and biomass yield (54.12 t ha^{-1}).	Islam et al. [74]
BGS at 15 Mg ha^{-1}.	Improves root growth and grain yield in maize.	Higher maize grain yields (6.21 Mg ha^{-1}) and improves root length density (1.10 cm cm^{-3}).	Garg et al. [64]
BGS (AD of a mixture containing cow dung and water at 1:1) application at 10% w/w.	Improves tomato yield and shows anti-nematodal properties.	Increase in number (3 fruits/plant) and yield (35.2 g/ plant) of tomato fruit. Nematicidal properties against *Meloidogyne incognita*.	Jothi et al. [63]
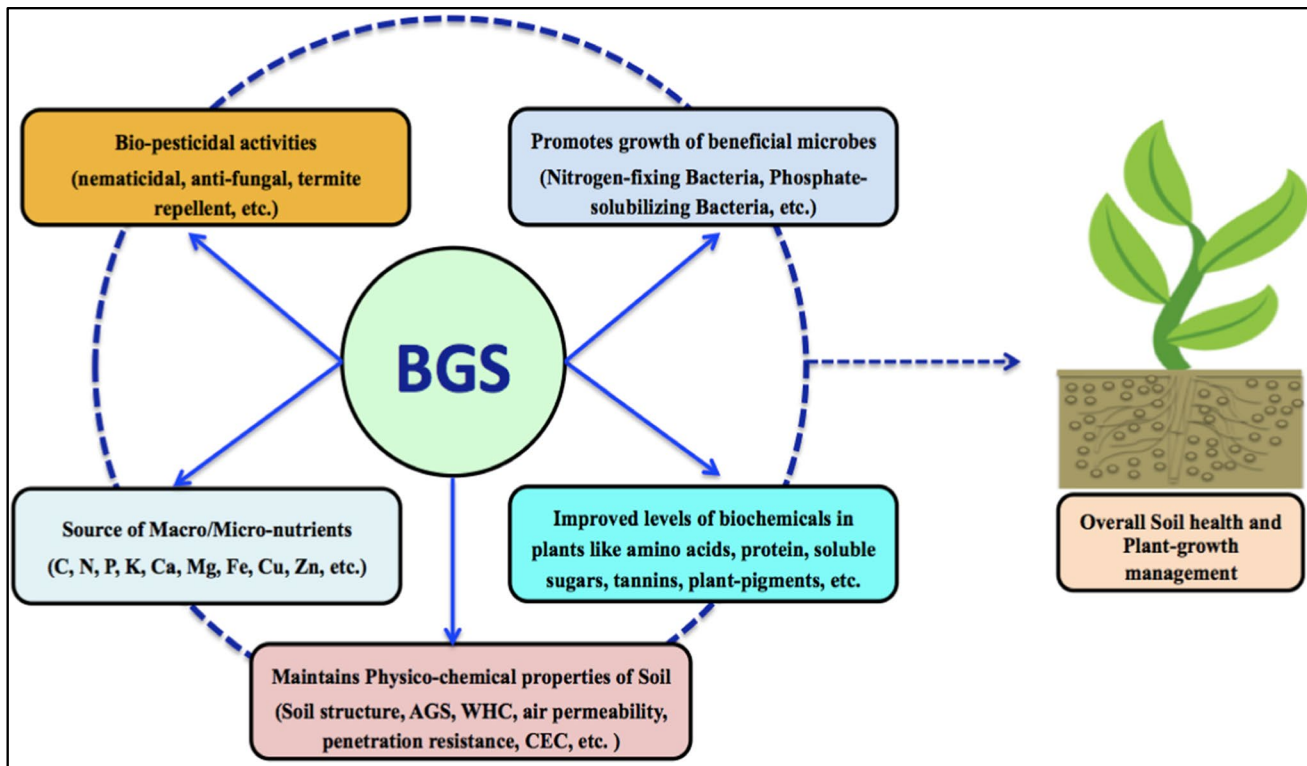
Fig. 3 Schematic representation of beneficial effects of BGS application in agriculture (source: conceptualized from the multiple authors referred in the manuscript) (AGS, aggregate stability; WHC, water-holding capacity; CEC, cation exchange capacity)

Biopesticidal activities Moving to biopesticidal potential, BGS successfully investigated for nematicidal, anti-fungal, termite repellent, and other similar activities. BGS (called effluent by the author) repels termites, and reduces weed growth by about 50%, and its compost was reported to increase 10–30% cereal crop production as compared to FYM if stored and applied properly [12]. AD of dairy manure promotes the multiplication of *Bacillus* species (*Bacillus subtilis*-B11; *Bacillus licheniformis*-B59) and suppressed late blight of potato [71]. Min et al. [72], utilizing anaerobically digested slurry, reported nematode control in tomato and radish. Jothi et al. [63] also observed nematicidal properties of BGS (obtained from AD of CD and water mixture in 1:1) applied at 10% w/w in tomato against *Meloidogyne incognita*. Kupper et al. [73] formulated Bio1 biofertilizer from anaerobically digested CM supplemented with micronutrients (and additives for promoting fermentation) and found effectivity (at 10% concentration) in control of citrus black spot caused by *Phyllosticta citricarpa* in Natal oranges, grafted on Cravo lemon trees.

Improvement in biochemicals and other utilities Du et al. [66] have seen improved levels of starch and crude protein in wheat and maize with the application of Hoggery BGS. Islam et al. [74] applied BGS (at 70 kg N ha^{-1}) and reported

the highest crude protein content of 11.91% and biomass yield of 54.12 t ha^{-1} in *Zea mays*. Yu et al. [69] applied concentrated BGS and observed a higher amount of amino acids, proteins, vitamin C, soluble sugars, tannins, and beta carotene in tomato fruits. They have also reported higher counts of culturable microbes (actinomycetes, bacteria, and fungi) in Rhizospheric and non-rhizospheric soils. In addition to this, BGS also contributes towards mitigating climate change through sequestration of Carbon in the soil [75]. Soil fertilized with anaerobic digestates offers low CO_2 emission as compared to manure [48]. BGS can also serve as an organic carrier material to deliver agriculturally important microbes (AIMs). In particular, this area has been further reviewed in the Section 3 (solutions) of the manuscript.

3 Solutions

Various limitations associated with the BGS like low C/N transformation rates, high TAN content, ammonia volatilization, high pH, bulkiness, and risk of contamination imposed challenges like handling, storage, transport, and further application in agriculture fields to meet the desired benefits. Efforts have been made by the researchers to investigate the different approaches to overcome

such ambiguities. To facilitate the systematic assessment of the available and prospective solutions, the present section has been discussed under nanotechnology-based interventions, nutrient recovery from BGS, microbially inspired, and integrated approaches. Microbes are the key drivers of agroecosystems and are known for increasing nutrient availability to plants and conferring pest and disease resistance as well. NEDOCs, apart from being a rich source of essential nutrients for crop plants, act as substrates for the beneficial microbial community that facilitates their decomposition followed by the nutrient release in soil [76]. BGS enrichment through NEDOCs and AIMs can serve multiple benefits such as balanced C/N ratio, availability of nutrients, and growth-promoting effects to the plants. NGS platform allows comprehensive assessment of the microbial diversity in solid and liquid fractions of the digestate. Screening and selection of beneficial microbes can further be subjected to the BGS-inspired multi-substrate interaction with different organic substrates for mass multiplication of such microbes. These integrated approaches can materialize to develop different biofertilizer and organomineral-based bio-formulations. The additional processing of slurry dewatering, separation of LFD, and decontamination can also be minimized following such integrated approaches. In recent years, nanotechnology-based approaches have also been explored to bring more opportunities for the stakeholders associated with the agriculture sector. Nanoparticles are reported to have a significant influence on the microflora favoring soil nutrient mobilization [77] and recovery of nutrient-rich BGS [78]. The scope of nanotechnology-based interventions has also been explored to upgrade the fertilization potential of recovered BGS from the digesters. To provide state-of-the-art information, this section has been taken in detail into the following sub-sections: role of nanoparticles in AD of biomass, nanoparticle-microbe interaction, nanoparticle co-aggregation, and environmental impacts of nanoparticle introduction into the functional system and environment. Afterward, the different methodologies of nutrient recovery from the LFD have been reviewed and recent technological developments in the area of nitrogen recovery as free ammonia and its subsequent utilization as a renewable CO_2 adsorbent for biogas upgradation were also discussed. The manuscript is concluded with an overview of commercial viability and economic estimates concerning BGS. The study will be of great significance for the researchers aiming to encourage environmental-friendly methods of developing organic formulations from biomass wastes/generated end-products to meet the current fertilizer demands and promote sustainable and circular bioeconomy through a zero-waste approach.

3.1 Microbially inspired approaches for the enrichment of BGS

The successful investigations on mass multiplication of AIMs in a variety of organic substrates such as DOCs, CM, and BGS not only promote the novel routes of microbial introduction in the agriculture fields but also showcase the possibilities of value addition of different organic inputs of low nutrient value. Different strategies of BGS enrichment through microbially inspired routes have been reviewed in this section. Taking account of another key aspect, i.e., the microbial community dynamics of methanogens, little information is available on the unculturable microbes supporting AD of organic matter leading to biogas production [40]. However, advancement in molecular biology techniques such as NGS will be useful in depicting microbial community structures (diversity and richness) through taxonomic assignments (QIIME analysis), alpha diversity analysis (Shannon index, Simpson index, ACE, Chao1 index, and observed species), and others. The microbial community analysis reported in CD, CM, and BGS and the potential role of the identified microflora in crop improvement have been reviewed. Furthermore, integrated approaches of combining BGS with other organic inputs (NEDOCs) to exploit the microbial community dynamics through multi-substrate interactions have also been discussed.

3.1.1 Microflora associated with biogas feedstocks and BGS

Microbial diversity analysis 16S ribosomal sequencing studies allow diversity and dynamics analysis of uncultured microbes inhabiting the environmental niches [79, 80]. Integration of advanced "Omics" technologies permits detailed investigation of microbial community structures in varied habitations including biogas digesters containing biomass feedstocks. Han et al. [81] reported bacterial and archaeal community diversity in household rural biogas digesters fed with different raw materials such as vegetable waste, pig manure, and a mixture of other waste materials. The most abundant phyla reported were *Bacteroidetes*, *Firmicutes*, and *Proteobacteria*.

Application of BGS has substantial effects on the microbial population (Rhizospheric and non-rhizospheric soil) [69]. CD, one of the common feed substrates in biogas production, harbors diverse microbiota. CD consists of 80% water, 14.4% undigested matter, and 5.6% of a microbial community comprising fungi, bacteria, and actinomycetes [82]. Sun et al. [83] reported *Firmicutes* as the dominant phylum in cow manure and the operational taxonomic units (OTUs) were 270–274. The Shannon Diversity, Chao1, and Simpson indices recorded by them were 6.361 to 6.167, 358

to 420, and 0.957 to 0.971 respectively for the bacterial population and 2.081 to 2.404, 22 to 27, and 0.648 to 0.693 for Archaeal population in cow manure. BGS reportedly support bacterial populations such as *Proteobacteria*, *Chloroflexi*, *Firmicutes* and *Bacteroidetes* [40], *Bacillus* spp., *Carnobacterium* spp., [60], *P. fluorescence*, *Nomuraea rileyii*, *Verticillium lecani*, *Fusarium-15*, and *Trichoderma viride* [59]. Christy et al. [84] isolated *Acetobacter syzygii*, *Bacteroides nordii*, *Clostridium perfringens*, *Methanobacterium formicicum*, *Lactobacillus acidophilus*, *Methanosarcina siciliae*, *Prevotella bivia*, *Porphyromonas asaccharolytica*, and *Prevotella bivia* from bioreactor (fed with CD slurry).

Xu et al. [61] performed alpha diversity analysis of bacterial population in soil fertilized with BGS, chemical fertilizer (NPK for rice: 0.006, 0.10, 0.07 t ha^{-1} respectively; NPK and boron for rape: 0.25, 0.27, 0.19, 0.007 t ha^{-1} respectively), and control (soil without treatment). They have recorded higher Chao1 index values in sampled soil enriched with a low dose of BGS (59.9 t ha^{-1}) as compared to its higher dose (264.4 t ha^{-1}). The decreasing order of Chao1 index in sampled soil was as follows: BGS low dose 59.9 t ha^{-1} (39,552–44828)> chemical fertilizer treatment (38,869–41,143)> control (37,068–41,756). The values obtained by them for Shannon Index were 10.80–12.22 for BGS (at 59.9 t ha^{-1}), 10.97–12.05 for chemical fertilizer treatment, and 10.44–12.46 for control. Compared to control, the abundance of *Acidobacteria*, *Chloroflexi*, *Planctomycetes*, *Verrucomicrobia*, *Nitrospirae*, and AD3 was relatively higher in BGS treatments. The alpha diversity indices for microbial community analysis associated with BGS are in Table 2.

Application of microbes (associated with BGS and biogas feedstocks) in agriculture Lu et al. [88] identified 17 bacterial strains from CD ((*Alcaligenes faecalis* CD232, CD243, CD257, CD261, CD205), *Bacillus cereus* (CD9, CD98, CD260), *Microbacterium aerolatum* (CD142), *P. otitidis* (CD237), *P. beteli* (CD204), *P. aeruginosa* (CD245), *Providencia rettgeri* (CD256), *Proteus penneri* (CD129, CD212), *Staphylococcus xylosus* (CD250), and *Staphylococcus sciuri* (CD97)) (out of 219 isolates) that showed anti-nematodal activity (greater than 90% within 1 h) against *Caenorhabditis elegans*. Except *Alcaligenes faecalis* CD205, *P. otitidis* CD237, and *P. aeruginosa* CD245, volatile compounds of the remaining 14 bacterial isolates exhibited nematicidal activity against *Meloidogyne incognita*. *Bacillus licheniformis* isolated from CD (fermented with basalt dust and eggshell) showed IAA production (1.75 µgmL^{-1}), phosphorous solubilization (2.37% in 10 days), and antagonistic activity against plant fungal pathogen (*Rhizoctonia bataticola*) [89].

Bioinoculants with multiple properties can be preferred in agriculture fields. Paikray and Malik [90] in a US patent application disclose a synergistic composition of at least seven beneficial bacteria (*P. fluorescens*, *P. striata*, *Azospirillum*, *Azotobacter*, *Bacillus subtilis*, *Bacillus polymyxa*, and *Lactobacillus*); at least two beneficial fungi (*Trichoderma herzianum, T. viride*); at least one yeast (*Saccharomyces cerevisiae*); and at least one compound which extends the effective lifetime of said formulation. Game et al. [91] studied the efficacy of microbial consortium (Bacterial isolate B-28 (*Bacillus* spp.), fungal isolate F-13 (*Aspergillus terreus*), and *Actinomycetes* isolate A-40 (*Streptomyces* sp.)) for composting of rural (farm waste, animal litter, animal fodder waste, dung) and urban wastes (vegetable waste, kitchen waste, roadside waste, papers, etc.) and reported the reduction in composting time coupled with nutrient enrichment in compost derived from both the waste types as compared to the commercial consortium and uninoculated controls.

BGS in combination with different organic amendments reported to support the population of bio-control agents such as *P. fluorescence*, *Verticillium lecani*, *Trichoderma viride*, and *Metarhizium anisopliae* [59]. Kavya et al. [92] reported an increase in the growth of beneficial microorganisms (*Rhizobium*, *Pseudomonas*, *Azotobacter*, *Azospirillum*) on different combinations of substrates (CD, press mud, poultry litter, kitchen wastes, maize stalks, and fruit wastes) utilized for biogas production and observed that the different organic substrates supported the microorganism's growth for at least 3 to 4 weeks of duration. Karmegam and Rajasekar [93] studied the strategies for enriching BGS vermicompost with *Azotobacter chroococcum* and *Bacillus megaterium*.

Soil available nutrient through the application of organic matter relies upon the catabolic potential of microflora [94]. The viability and an adequate load of microorganisms are important to get appropriate results in the agriculture fields. Bioinoculant success depends upon the survival of microbial strain in the soil, more prominently under adverse agro-climatic conditions such as temperature variations and pH range. Scientists and biofertilizer producers are also facing the same challenges to increase the effectiveness of biofertilizers under variable geographic and agro-climatic conditions [1]. The development of stress resistance in microbial strains through phenotypic adaptations (permanent and non-revertible) is preferred over genetically manipulated strains (gene transfer) due to environmental hazards associated with the introduction of genetically modified organisms [1]. Thus, studies pertaining to the selection and application of suitable bio-inoculants (with respect to soil nature, agro-climatic condition, crop variety, etc.) and microbial formulations coupled with soil texture profiling to tune up with microbial growth must be taken up further [1].

Table 2 Alpha diversity analysis of microbial community associated with BGS

Sample	Microbial community	Alpha Diversity analysis	Reference
AD of municipal wastewater activated sludge using ZnO NPs (at different concentrations, i.e., CA=30, CB=90, and CC=150 mg g^{-1}-VSS; while C serves as control).	Archaeal domain: dominant phylum (DP): *Euyarchaeota* (CA= 93.52%, CB=91.59%, CC=89.58%, and C=97.74%) Dominant class *Methanomicrobia* (CA= 80.36%, CB=78.04%, CC=76.64%, and C=90.16%) Dominant genus *Methanosaeta* (CA= 68.86%, CB=58.87%, CC=61.20%, and C=67.64%)	Bacterial population: OTU: 4353 (CB), 4445 (CC), 4963 (CA), 5617 (C) Shannon Diversity Index: 5.577 (CC), 5.720 (CB), 5.904 (C), 5.939 (CA). Chao1 Index: 19891.13 (CB), 20421.16 (CC), 20664.90 (CA), 26153 (C). Archaeal population: OTU: 2024 (C), 2433 (CA), 2576 (CB), 2888 (CC). Shannon Diversity Index: 2.426 (CA), 2.609 (C), 2.676 (CB), 2.703 (CC). Chao1 Index: 15904.09 (C), 19511.67 (CB), 20351.29 (CA), 25474.98 (CC).	Wang et al. [85]
Anaerobic digestate of cow manure and corn straw mixture (methanogenic phase, temperature varies from 20 to 35°C.	DP at all the temperatures: Thaumarchaeota Abundant Genus at all temperatures: *Cenarchaeum*	Shannon Diversity (4.03 to 5.09) and Chao1 (696 to 894) index varies with the temperature.	Wang et al. [49]
Anaerobic digestate of cow manure and corn straw mixture (acidogenic phase, temperature varies from 20 to 35°C.	Abundant genera at temperature range 25 to 35 °C: *Acinetobacter, Acetitomaculum,* and *Bacillus.* Abundant genera at 20°C temperature: *Cryptanaerobacter* and *Solibacillus.*	Shannon Diversity (4.51 to 6.46) and Chao1 (932 to 1214) index varies with the temperature.	
Anaerobic co-digestate of pre-treated corn straw and pig manure (C/N ratio varies 20, 25, 30, 35, and 300).	DP: *Bacteroidetes, Firmicutes, Proteobacteria, Spirochaetes, Chloroflexi,* and *Cloacimonetes.* Abundant Genus: *Rikenellaceae.*	Shannon Diversity (6.1742 to 7.5944) and Chao1 (1162.2264 to 1960.6667) index varies with the C/N ratio.	Ning et al. [86]
Sludge (obtained from membrane bioreactor) containing different doses of ZnO NPs, i.e., C (no ZnO NPs addition), T1 (ZnO NPs at 1 mgL^{-1}), T2 (ZnO NPs at 5 mgL^{-1}), T3 (ZnO NPs at 10 mgL^{-1}), T4 (ZnO NPs at 4 mgL^{-1}), and T5 (ZnO NPs were no longer added to the bioreactor).	Relative abundance: T5 (*Nitrosomonas*); C (*Nitrosococcus, Nitrosospira*); T5 (*Candidatus brocadia*); T1 (*Candidatus kuenenia*); C (*Nitrobacter*); T1 and T2 (*Nitrospira*); C and T1 (*Denitratisoma*).	Shannon Diversity: 4.27 (C), 4.39 (T1), 4.34 (T2), 4.24 (T3), 4.30 (T4), and 3.66 (T5).	Zhang et al. [87]
Anaerobic digestate of Cow manure at day 42[a], 85[b], and 148[c].	DP: *Firmicutes.*	Shannon Diversity (6.355[a], 6.292[b], 5.819[c]) and Chao1 (487[a], 553[b], 440[c]) index of bacterial population.	Sun et al. [83]
Anaerobic digestate of cow manure at 37°C (constant temperature).	DP: *Firmicutes.*	Shannon Diversity (4.896–5.550) and Chao1 (320–371) index of bacterial population at day 9.	

3.1.2 NEDOCs: potential substrates for mass multiplication of AIMs

BGS can combine with the organic substrates of high nutrient values like NEDOCs. The combination of BGS and NEDOCs complements each other in terms of improving their properties (such as balanced C/N ratio and moisture content), which can also be productive for the mass multiplication of AIMs. Besides this, the addition of NEDOCs potentially reduces the requirement of dewatering in BGS, hence aiding convenience in storage and handling for field applications. This is a key aspect as the process of drying BGS to reduce water content is reported to have low nitrogen levels as compared to the fresh BGS (wet) [9]. Thus, the addition of NEDOCs to some extent can reduce the necessity of additional treatments of BGS (to reduce water content) and could be beneficial to sustain the nutrient levels in BGS, which are probably lost at the time of drying.

The major fraction of seed cakes is reportedly rejected as toxic, and remains unutilized [95]. India has over 100 species of non-edible oil seed-bearing plants [96, 97], out of which presently 10–12 varieties have been explored until now [97]. Around 70% of oiled cake has been recovered after expelling oil from the tree-borne oilseeds [96]; hence, the availability of the DOCs can be estimated from such figures. Marathe and Deshmukh [96] while highlighting the oilseed production potential in India mentioned the non-edible oilseed cakes quantity of about 2.812 MMTs. However, as cited by Prasad [98], an evaluation of the exact quantity of non-edible oilseed cakes (NECs) is insignificant.

Table 3 shows the nutrient compositions of different non-edible seed cakes published in the previous reports. NEDOCs of *Azadirachta indica* contain N, P, K, Ca, Zn, Fe, etc., [99]; similarly, nutrients reported in *P. pinnata* DOCs were C, N, P, K, Ca, Mg, S (macronutrients), Zn, Fe, Cu, and B (micronutrients) [39, 100]. Jared et al. [38] found higher Mn, Zn, Ca, and Fe in slurry compost as compared to BGS, though the NPK levels were greater in BGS. As the microbes play an important role in the process of composting [101], the higher nutrient availability can be attributed to the undergoing microbial actions. Microbial biofertilizers have significant potentials for biological nitrogen fixation:

nutrient recycling, solubilization, and mobilization, etc. [102]. NEDOCs have also been investigated for supporting the population of agriculturally beneficial microbes [103, 104]. Optimization of BGS and DOC combination for mass multiplication of AIMs followed by application of developed biofertilizer formulations in agriculture fields can strategically favor the soil nutrient profile and higher crop yields.

The presence of toxic compounds in NEDOCs limits their applications in the agriculture fields. Decomposition substantially reduces the presence of toxic compounds present in DOCs. Das et al. [106] through co-composting of Jatropha DOC (J-DOC) and animal dungs considerably reduced the Phorbol ester content. Incorporating AIMs further improves the efficiency of decomposition followed by the metabolization of such toxic compounds. Researchers successfully utilized the NEDOCs as low-cost substrates for the multiplication of biocontrol agents, growth-promoting microbes, and developing different bioformulations [39, 76, 108–115]. According to Singh et al. [69], the utilization of DOCs as substrates for the multiplication of beneficial microbes facilitates soil fertility under field conditions coupled with reducing the risk of disease occurrence. Tomer et al. [112] mentioned the role of allelochemicals behind the prevention of various soil-borne diseases through the combined application of DOCs and microbial bio-control agents.

Studies in the area of screening, identification, and separation of bioactive compounds produced by the AIMs utilizing DOCs and BGS could be beneficial in developing formulations of potential biocontrol activities. The approach seems to be more conceivable if we look upon the findings of Sharma et al. [108], Arora et al. [109], and Sharma et al. [116]. Sharma et al. [108] multiplied *Purpureocillium lilacinum* 6029 on Karanja DOC (K-DOC) and the metabolites of potential biocontrol activity identified by them were benzene acetic acid, benzoic acid, phenyl ethyl alcohol, 2-ethyl butyric acid, and 3,5-di-t-butylphenol. The first report on the presence of butyric acid (possesses nematicidal activities) in *P. lilacinus* filtrate was predicted to be the result of metabolic pathways elicited in response to their growth (during submerged fermentation) on K-DOC [116]. Arora et al. [109] optimized K-DOC, J-DOC, and dextrose combination (4.5, 12.5, and 10 gL^{-1} respectively)

DOCs	Nutrient composition
Karanja	C: 42.26±2.4%; N: 4.87±0.87%; P: 0.89±0.03%; K: 1.3±0.02% [39]; Ca: 0.25%; Mg: 0.17%; Zn: 59 ppm; Fe: 1000 ppm; Cu: 22 ppm; Mg: 74 ppm; B: 19 ppm; and S: 1894 ppm [100]
Neem	N: 4.51%; P: 0.79%; K: 1.40%; Zn: 57 ppm; Ca: 1.40 ppm; Fe 640 ppm [99]
Mahua	C: 44.93±0.08%; N: 3.15±0.02% [105]
Jatropha	N: 2.93±0.06%; P: 9623±109 ppm; K: 8563±157 ppm; C/N: 10.2±0.10 [106]
Castor	N: 75.4 gkg^{-1}; P: 31.1 gkg^{-1}; K: 6.6 gkg^{-1}; Ca: 7.5 gkg^{-1}; Mg: 5.1 gkg^{-1} [107]

Table 3 Nutrient compositions of different non-edible seed cakes

for the growth of *Paecilomyces variotii* and reported dipi-colinic acid (DPA) production 2.5 times higher as compared to the basal medium. The effectiveness of DPA in regulating rhizospheric phytopathogens has also been reported. In this way, not only the wide range of NEDOCs; the other biomass wastes such as BGS can also be exploited to develop a rich repository of compounds derived from AIMs through multi-substrate interactions.

The low levels of NPK or other nutrients in the cattle feed could have been reflected in the CM and consequently in the recovered BGS. Under such conditions, BGS alone will be unable to fulfill the entire nutrient demands as a fertilizer. To balance the phosphorus levels in BGS, Mdlambuzi et al. [33] added superphosphate. However, possibilities of such amendments can also be possible through the enrichment with DOCs and AIMs (such as phosphate-solubilizing bacteria). The C/N content is critical in the investigations dealing with the suitability of different substrate(s) to achieve higher microbial growth. Sharma et al. [110] optimize the C/N ratio of 40:1 (pH=7) in K-DOC (using sucrose as carbon source) for the growth of *P. lilacinus*. Sharma et al. [39] have attempted to replace the sucrose with BGS (sun-dried) as a potential carbon source (required for microbial growth under SSF). The recorded values of C/N ratio for K-DOC and BGS (sun-dried) were 8.68 ± 0.11 and 57.78 ± 0.19 respectively [39]. In concerned with their suitability favoring microbial growth, the C/N ratio appears to be high in K-DOC and low in sun-dried BGS. The authors [39] reported K-DOC/BGS ratio of 60/40 as optimal for *P. lilacinum* 6029 multiplication under SSF. In co-composting of Jatropha DOCs and rice straw mixture (1:9) (OM) with different animal dungs (buffalo dung (BD), cow dung (CD), goat dung (GD),

horse dung (HOD), and hen droppings (HED)), Das et al. [106] recorded the highest C/N ratio in OM+CD (20:1). Upon optimizing the OM and animal dung proportions, the OM+BD, OM+GD, OM+HED, and OM+HOD in the combination of 20:1, 10:1, 5:1, and 5:1 respectively showed the highest C/N ratio (Fig. 4). Thus, as compared to other animal dungs, CD contributes maximum C/N ratio and undoubtedly emerges as an ideal ingredient to directly or indirectly (after resource generation such as BGS) utilized with other organic substrates for the mass multiplication of AIMs. Such studies certainly pave the way ahead to explore the diverse range of carbon-rich biomass wastes with the potentials of replacing expensive carbon sources.

3.2 Integrated approaches

This section entails the utilization of BGS for agricultural applications through enrichments via multi-component organic substrates, vermicomposting, NPK amendments, etc. Alongside, methodologies to overcome the contamination risks (pathogens, heavy metals, pesticides, and other organic pollutants) and handling a liquid fraction of BGS were also taken into consideration; hence, the said subsection is entitled as integrated approaches.

Taking account of growth-promoting activities of the BGS-AIM combination, Hamid et al. [117] recorded an overall improvement in growth, yield, and physiology of sunflower under moderate salinity stress with the application of BGS enriched with *B. subtilis* Y16 strain. Ahmad et al. [118] also noticed improvements in maize growth under salt stress using BGS supplemented with biofertilizer and 100% recommended dose of nitrogen. Gao et al. [119] reported an

Fig. 4 C/N ratio under optimized combinations of OM with different animal dungs (refer text for the details of abbreviations used) [106]
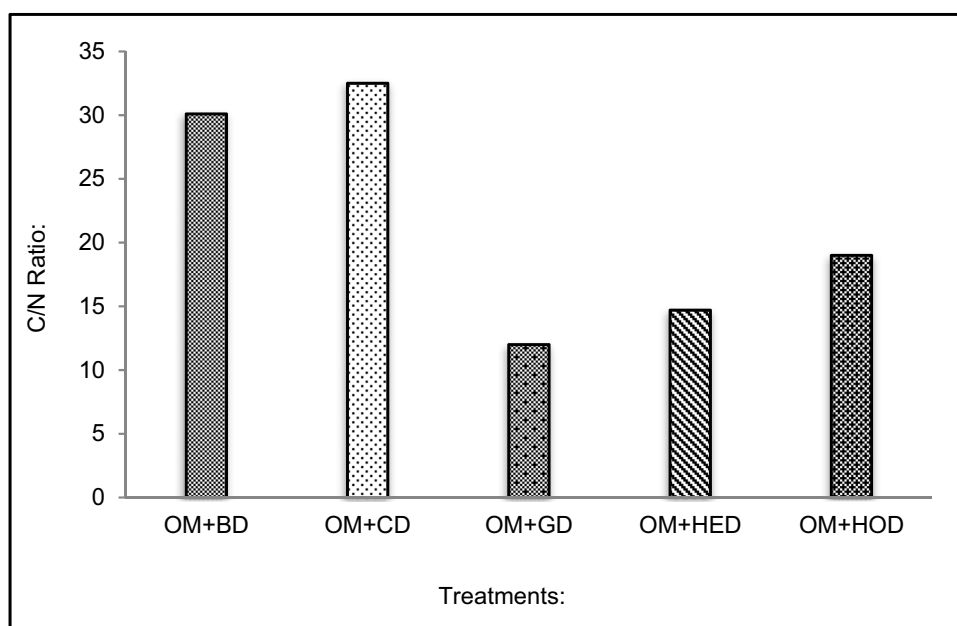
increase in mycorrhizal colonization, rhizospheric bacterial count, and overall growth in maize using formulation containing BGS, biofertilizer, and 50% NPK. Sharma et al. [39] developed a fungal formulation (*P. lilacinum*) using K-DOC and sun-dried BGS (60:40) as a substrate for nematode control. Devakumar et al. [59] also utilized multi-substrate-enriched BGS for the multiplication of biocontrol agents such as *P. fluorescence* and *Trichoderma viride*. In an experiment, soil samples were collected from BGS-cultivated (paddy) (BCL) and chemically cultivated lands (CCL) and found 23.3% higher bacterial counts in BCL as compared to CCL [36]. Thus, BGS application supports a favorable environment for microbial growth in agriculture fields.

Treatment of BGS has a significant influence on enzymatic regulation, cellular development, and photosynthetic induction [119]. Organic or mineral enrichments could be helpful to reduce the bulkiness of BGS and increase its application as fertilizer in agriculture [7]. Notably, performing anaerobic co-digestion (ACD) of CM with other carbon-rich organic substrates (balanced C/N ratio) promotes microbial activity, thereby improving biogas yield and recovery of nutrient-rich digestate.

Vermicomposting of BGS significantly reduces the C/N ratio (above 80% as compared to an untreated BGS recorded by Yadav et al.) [120], which supports its application in agriculture as a potential organic nutrient input. They have reported higher TKN values (g kg^{-1}) with vermicomposted BGS (29.90±2.10) as compared to an untreated slurry (6.20±0.25). The microbial diversity of vermicompost can also be explored for the value addition of BGS. Hao et al. [121] in their investigation observed that the bacterial communities associated with the vermicompost showed metabolic competence for the upgradation of swine manure compost. Decline in *Stenotrophomonas* and *Acinetobacter* (genera of phylum Proteobacteria known for pathogenic bacteria) and rise in *Chryseolinea* and *Actinomadura* genera (beneficial microbes) were also noted. The 30th-day inoculation of *A. chroococcum* at the rate of 30 mL, 35 mL, and 40 mL per 175 g of vermibed substrate (composted *BGS*) showed survival up to 105 days (cfu 2×10^7 g^{-1}), 135 (cfu 1×10^7 g^{-1}) days, and 135 days (cfu 2×10^7 g^{-1}) respectively [93]. In the case of *B. megaterium*, they found that the 30th-day inoculation at the rate of 30 mL, 35 mL, and 40 mL per 175 g of vermibed substrate showed survival up to 150 days (cfu 2×10^7 g^{-1}), 165 (cfu 1×10^7 g^{-1}) days, and 165 days (cfu 2×10^7 g^{-1}) respectively.

Extracts of *T. brownii* leaves and barks of *Acanthaceae* spp. (at 5% v/v separately) added to biogas substrate (67% kitchen waste and 33% of CD and goat dung in 1:1) reported increasing biogas yields and nutrient content (TKN, TAN, TS, TP, etc., in the case of *T. brownii* leaves extract; and K, Ca, NO$_3^-$, PO$_4^{3-}$, SO$_4^{2-}$ in the case of *Acanthaceae* spp. bark extract) in BGS [122]. Zhang et al. [123] performed

ACD of rice straw and combined swine manure-urea mixture in 7:3 (C/N ratio 24.23) and observed higher biogas production coupled with improving fertilization value of recovered digestate as compared to other treatments and control. The authors stated that the urea addition could foster the lignin degradation, hasten the process of hydrolysis, and improve N content in the digestates.

Partial replacement of chemical fertilizers with BGS has also been reported in a few studies for fertigation purposes. BGS enriched with variable doses of NPK promotes growth and pod yield in the pea plant [124]. As compared to fertilizer control, Jamison et al. [125] observed a slight increment in nutrient uptake and use efficiency in *Brassica juncea* using lignocellulosic biomass digestate supplemented with commercial liquid fertilizer. Do and Scherer [126] witnessed higher ryegrass yield using hygromull-enriched anaerobic digestate. Fraire García et al. [127] observed an increase in maize and tomato yields in Ultisol soil fertilized with BGS (enriched with recommended fertilizer dose). Xu et al. [9] found improvement of forage and biomass quality in *Lolium multiflorum* with partial replacement of chemical fertilizers (CFs) with BGS. Supplementation of BGS (100.5 kg ha^{-1}) with CF (37.5 kg ha^{-1}) was reported to increase crude fiber and crude protein content in *L. multiflorum* by 10.00% and 10.35% respectively as compared to CF treatment alone (112.4 kg ha^{-1}) (NPK content in BGS: 0.23%, 0.05%, 0.32% respectively; in CF: 11.34%, 7.56%, 7.56% respectively). Refer to Table 4 for more applications of enriched BGS in agriculture.

Apart from this, while handling the BGS, its management is also important to prevent the loss of nutrients at the time of application [53]. The BGS applied in agricultural fields is majorly acquired from biogas digesters operated with feedstock sources like livestock slurry (LSS), anaerobic co-digestates of LSS, and plant biomass or biomass waste such as agriculture residues. The recovered BGS is either applied directly or in supplementation with micronutrients, chemical fertilizers, etc. In such instances, the likelihood of meeting expected benefits primarily depends upon the handling, storage, and mode of the BGS application. The effectiveness of different application modes (such as root irrigation and spraying) also needs to be investigated [69]. Higher nutrient concentration can result in the accumulation of ions at the intercellular level, damage chloroplast structure, and consequently, decrease in chlorophyll content and photosynthetic rate [37]. Therefore, further studies leading to the optimization of effective dose with reference to the application mode, soil nutrient profile, plant's requirement, etc., are hereby suggested.

Another emerging challenge with the BGS (precisely obtained from animal manures, urine, and different waste streams as feedstock material for AD) is the likelihood of pathogens, heavy metals, pesticides, etc., which further

Table 4 Applications of enriched BGS in agriculture

Value addition	Application	Reference
Deoiled Karanja cake and BGS (sun-dried) (60:40).	Utilized as a substrate for the development of fungal formulation (*Purpureocillium lilacinum* 6029 based) for Nematode (*Meloidogyne incognita*) control (96.8% inhibition of egg mass, 376.65 Ug⁻¹ protease activity, and on the 3rd day 100% egg mass colonization observed).	Sharma et al. [39]
BGS enriched with different doses of NPK.	BGS (8 t ha^{-1}) enriched with variable doses of NPK (t ha^{-1}) promotes growth and pod yield in pea (*Pisum sativum* L.) in the following order: 60:135:135>40:90:90>80:180:180> 100:225:225.	Jilani et al. [124]
BGS enriched with *B. subtilis* Y16 strain.	Improves physiology (10–84%), growth (11–179%), and yield (106%) of sunflower under the conditions of moderate salt stress (as compared to the respective control experiments).	Hamid et al. [117]
Digestate of lignocellulosic biomass (semi-continuous AD of *Pennisetum purpureum*) enriched with liquid fertilizer (6-4-4) (GrowBig®, FoxFarm, Samoa, CA, USA) (1:1 and 1:9 w/w).	Digestate combination with liquid fertilizer showed a slight increase in nutrient uptake, nutrient (nitrogen) use efficiency, and growth in *Brassica juncea* (Kai Choy) as compared to fertilizer control (100% fertilizer).	Jamison et al. [125]
BGS (substrate feed rabbit manure) + biofertilizers [*A. chroococcum* SARS 101+ AMF (spores of *Glomus clarum*, *Gigaspora margarita*, *Glomus mosseae*) + *B. circulans* ARC-SWERI-2] + 50% NPK.	Maximum leaf area (1238.2 cm^2 and 1147.7 cm^2), dry weight of root (17.11 g and 18.94 g), and leaves (44.85 g and 45.59 g) in maize during both the growing seasons. Increase in mycorrhizal colonization and rhizospheric bacterial count.	Gao et al. [119]
Anaerobic digestate (substrate feed: 27% CD slurry + 8% *Vitis vinifera* (L.) stalks + 43% *Zea mays* (L.) silage + 22% *Triticale* (X. triticosecale, Wittmack) silage) in liquid, solid, and pellet form. Liquid digestate enriched with agriperlite, while solid and pelleted digestates were separately enriched with standard solution (contains NPK and acidifying agent).	Promotes growth of baby leaf lettuce (Chiara cultivar) in hydroponic cultivation.	Ronga et al. [128]
Ultisol soil fertilized with BGS (25t/ha) enriched with the recommended dose of inorganic fertilizer (1:1).	Increase in maize (208%) and tomato (169%) yield as compared to other treatments, i.e., recommended dose of inorganic fertilizer.	Fraire García et al. [127]
BGS (feed substrate buffalo manure) (field application at 270 kg N ha^{-1}) enriched with different doses of ZnO NPs (average size ≤40 nm; conc. 20% w/v in distilled water) (1.4, 2.8 and 3.6 mg kg^{-1} soil).	Improves soil nitrogen content.	Aziz et al. [77]
CD BGS (at 5000 kg ha^{-1}; NPK content of CD BGS was 1.3%, 2.8%, and 0.8% respectively) supplemented with synthetic fertilizer (N (137), P (33), K (115), S (30), Zn (1), B (1) kg ha^{-1}) (application rate 5 t ha^{-1}).	Highest fruit yield in tomato (*Solanum lycopersicum* L.) plant.	Ferdous et al. [62]
BGS (0.45) combined with chemical fertilizer (0.55) (based on total nitrogen content).	Promotes soil fertility and improves soil health.	Zheng et al. [6]
50% BGS enriched with 50% of nitrogen (recommended dose, i.e., NPK 150:60:60 kg ha^{-1}).	Increase 60% and 20% cob yield in baby corn (Syngenta G-5414) as compared to control and recommended urea dose respectively.	Malav et al. [129]
BGS enriched with Panchagavya, Jeevamrutha, compost extract, and press mud (1:1:1:1:1).	Multiplication of biocontrol agents *Metarhizium anisopliae*, *P. fluorescence*, *Nomuraea rileyii*, *Verticillium lecani*, *Fusarium-15*, and *Trichoderma viride*.	Devakumar et al. [59]
BGS (at 600 kg ha^{-1}; NPK content of BGS was 0.78%, 1.01%, and 0.91% respectively) + 100% of Recommended Nitrogen + Biofertilizer (*P. fluorescens* and *Enterobacter aerogene*).	Improves maize growth under salt stress conditions. Maximum plant height (171.33 cm), shoot weight (212.80 g), root length (33.50 cm), root weight (26.47 g), cob length (19.44 cm), cob weight (164.13 g), and 100-grain weight (24.73 g).	Ahmad et al. [118]

Table 4 (continued)

Value addition	Application	Reference
Vermicomposting of BGS (using *E. fetida*).	Higher values of TKN (g kg^{-1}) 29.90±2.10 as compared to untreated Biogas slurry (6.20±0.25).	Yadav et al. [120]
Biogas residues (substrate feed pig manure and maize) from biogas plant enriched with hygromull.	Higher yield of rye grass. Promotes nutrient uptake.	Do and Scherer [126]
Anaerobic digestate (piggery excreta and wastewater from piggery cleaning) with NPK content 540, 17.1, and 317.8 kg ha^{-1} respectively.	Improves rice grain yield (11.3 × 10^3 kg ha^{-1}), 24% and 4.9% higher yield as compared to control (no fertilizer) and chemical fertilizer (urea=270 N kg ha^{-1}, superphosphate=39.3 P kg ha^{-1}, KCl-150 K kg ha^{-1}) treatment respectively.	Lu et al. [29]
BGS enriched with Panchagavya (5 kg CD/BS, 12 ripe bananas, 3 L tender coconut water, 1 L cow ghee/butter clarified, 3 L cow urine, and 2 L each of cow milk and curd).	Highest grain yield in Maize ([a]3.14 t ha^{-1}) and Sunflower ([a]3.14 and [b]3.24 t ha^{-1}). a= growing year 2001. b= growing year 2002.	Somasundaram et al. [130]
Bio1 biofertilizer (anaerobically digested CM supplemented with micronutrients and additives for promoting fermentation) at 10% concentration.	Controls citrus black spot (caused by *Phyllosticta citricarpa*) in Natal oranges, grafted on Cravo lemon trees.	Kupper et al. [73]

contaminates soil and plants when applying directly to the crop fields. It is suggested that in cases where BGS applied to the vegetables and fruits that are consumed fresh, they must be washed well before consumption [17]. The risk of pathogens can be reduced through proper treatment of BGS (optimal retention time, temperature, etc.) [14]. Appropriate dilutions before field applications can further reduce the toxicity risk as a consequence of the higher concentration of ammonia and soluble P, presence of heavy metals, pathogens, and other contaminants. As mentioned earlier, the process of AD does not degrade heavy metals [27]; however, their solubility and availability can decrease (due to precipitation, struvite production, etc., depending upon metal type) [21]. With BGS application, the use of metal-immobilizing materials can play a substantial role in mitigating the potential risk of heavy metal pollution [27]. However, further studies are duly recommended to explore such possibilities.

3.3 Nanotechnology-based interventions

Nanoparticles offer a large surface-area-to-volume ratio, thus are reported to have no impact on chemical properties of soil (such as pH, electrical conductivity) when applied in combination with organic matter (such as anaerobic digestates) or alone [77]. Nanoparticles are also found to influence the microbiota and extracellular enzyme secretion in the soil thus favoring soil nutrient mobilization [77, 131]. Nanoparticle treatment has been reported to enhance biogas production through the increased activity of bacteria degrading organic matter anaerobically and the resulting waste can be utilized as quality compost [78]. Table 5 shows the role of NPs in biogas production from organic feedstocks such as livestock, phyto-biomass, and sludge/wastewater. The idea was to give an overview (based on the latest findings) of how nanoparticle dose, type, and their interaction with feedstock substrates and microbial community during AD affect the yield and quality of biogas production.

Apart from NPs, trace elements/bulk materials/mineral accelerants (such as iron oxide, copper oxide, zinc oxide, copper chloride, cobalt chloride, iron oxide-zeolite, magnetite, Fe^{2+} (as $FeCl_2 \cdot 4H_2O$), Ni^{2+} (as $NiCl_2 \cdot 6H_2O$), Fe–Co, Fe–Ni, Fe–Co–Ni, cupric sulfate, cupric glycinate, vermiculite, limonite, bentonite, waste iron powder, zvi powder, fly ash, clinoptilolite) have also reported improvements in the biogas/methane yield during AD of biomass [132–147]. We suggest further readings [148] to get more information on the role of trace elements (Cu, Co, Fe, Mn, Mo, Ni, Se, W, Zn, V) in different enzymes that take part in AD. The addition of trace elements/mineral accelerants during AD was also reported to improve fertilization values of recovered digestate, as observed by Xu et al. [134] in the case of vermiculite addition (as mineral accelerant). The biostimulation (BSM) of methanogens during AD through laser irradiation

Table 5 Applications of Nanoparticles in an anaerobic breakdown of organic substrates

Substrate for AD (SBAD)	Nanoparticle (s)	Observations	Reference
Livestock dung (SBAD)			
CM + NiNPs	NiNPs (4 mg L^{-1})	Higher biogas yield (792.0 mL/g VS) as compared to CM alone (589.2 mL/g VS).	Abdelwahab et al. [151]
	NiNPs (2 mg L^{-1})	Increase methane yield by 70.46% as compared to CM alone. Higher removal efficiency of Hydrogen sulfide (H$_2$S) gas (47.5%).	
CD + nanoferrites of Ni, Fe, and Co (NiFe$_2$O$_4$, Fe$_3$O$_4$, and CoFe$_2$O$_4$ respectively)	Same concentrations of metal nanoferrites in a different set of experiments	Cumulative volumes of biogas follow the trend: CoFe$_2$O$_4$> Fe$_3$O$_4$> NiFe$_2$O$_4$.	Sliem et al. [152]
CM + (FeNPs + NiNPs)/(FeNPs + NiNPs + CoNPs)	FeNPs (30 mg L^{-1}) + NiNPs (2 mg L^{-1})	Higher biogas yield (14.61%) as compared to control (CM alone).	Abdelwahab et al. [153]
	FeNPs (30 mg L^{-1}) + NiNPs (2 mg L^{-1}) + CoNPs (1 mg L^{-1})	Higher methane yield (19.30%) as compared to control (CM alone). Higher reduction in H$_2$S (35.10%) from biogas as compared to control.	
CM + FeNPs	FeNPs (15 mg L^{-1})	Higher biogas yield (953.41 mL/g VS) as compared to CM alone (589.2 mL/g VS).	Abdelwahab et al. [154]
Sludge/wastewater (SBAD)			
Wastewater + FeNPs/CuNPs/NiNPs/FeNiCu NPs	FeNPs	Higher cumulated biogas yields (CBY) at concentration of 1 g (13 mL day^{-1}) as compared to 2 g (6 mL day^{-1}). 100% methane yield at lower FeNP concentration (1g).	Amo-Duodu et al. [155]
	CuNPs	Higher CBY at concentration of 1 g (9 mL day^{-1}) as compared to 2 g (8 mL day^{-1}). 100% methane yield at 1 g CuNP concentration.	
	NiNPs	Similar CBY at concentrations of 1 and 2g (6.5 mL day^{-1}). 100% methane yield at higher NiNP concentration (2g).	
	FeNiCu NPs	CBY and methane yield at 2g of concentration was 10.5 mL day^{-1} and 100% respectively.	
Sewage sludge + AgNPs	AgNPs	Higher methane yield and a significant reduction in H$_2$S from biogas. The abundance of Genus *Methanosarcina* as compared to control (without AgNP treatment).	Grosser et al. [156]
Sewage sludge (semi-continuous AD) + nZVI	Zero valent FeNPs (nZVI)	Improves methane content in biogas (abundance of acetoclastic species of *Methanothrix* genus).	Barrena et al. [157]
Phyto-biomass (SBAD)			
Durum wheat + ZnO NPs	Chemically and biologically (durum wheat extract) synthesized ZnO NPs (10 mg L^{-1})	Higher biogas yield from durum wheat treated with biologically (457 mL/g VS) and chemically (422 mL/g VS) synthesized ZnO NPs as compared to control (353 mL/g VS) (without NPs treatment).	Hassaan et al. [158]

Table 5 (continued)

Substrate for AD (SBAD)	Nanoparticle (s)	Observations	Reference
Pre-treated Slurry (using NPs and calcium hydroxide) mixed with wheat straw	Graphene Oxide	Average methane yield of 65% (HRT was 40 days).	Kaushal and Baitha [159]
Alcaligenes aquatilis and *Providencia alcalifaciens* (isolated from municipal wastewater treatment plant) + *Pistia stratiotes* and *Potamogeton pectinatus* (enriched with NiO NPs)	NiO NPs (2 ppm)	Improve biogas production.	Salama et al. [160]
Chlorella pyrenoidosa + α-Fe$_2$O$_3$ NPs (at the 12th day of growth period)	α-Fe$_2$O$_3$ NPs (30 mg L^{-1})	Increase in biogas (25.14%) and methane (22.40%) yield.	Rana et al. [161]
Barley straw (*Hordeum vulgare* L.) (0.4 mm) + ZnO NPs	Chemically and Biologically (*Antithamnion plumula* extract) synthesized ZnO NPs (10 mg L^{-1})	Higher biogas yield in barley straw (0.4 mm) treated with biologically (390.5 mL/g VS) and chemically (385.0 mL/g VS) synthesized ZnO NPs as compared to barley straw (mechanically pretreated) without enrichment of ZnO NPs (340 mL/g VS) and control (untreated biomass) (279 mL/g VS).	Hassaan et al. [162]
Enteromorpha (microwave pretreated) + MgO NPs	MgO NPs (5 and 15 mg L^{-1})	*Enteromorpha* (microwave pretreated at 800W, 4 min) + MgO NPs (15 mg L^{-1}) = 342 mL (biogas yield). *Enteromorpha* (microwave pretreated at 600W, 6 min) + MgO NPs (5 mg L^{-1}) = 320 mL (biogas yield).	Shi et al. [163]
Phragmites australis (Inoculated with sugar beet factory digestate) + Fe$_3$O$_4$ NPs	Ascorbic acid capped Fe$_3$O$_4$ NPs (100 ppm)	Higher biogas yield (31.10 mL/3.4g of *P. australis*) as compared to control (18.07 mL/3.4g of *P. australis*).	El-Borady et al. [164]

(LR) has also been described to enhance the microbial actions, and in the latest developments the LR is coupled with NP treatment for improving the biogas production [149, 150] and LCA assessment performed by Samer et al. [150] asserted that the application of LR with NPs for BSM during AD have minimal adverse effects on the environment as compared to control.

While reviewing the literature, we have encountered a study where nanoparticles (zinc oxide NPs, average size ≤40 nm; conc. 20% w/v in distilled water) were supplemented with recovered BGS to investigate the effect of developed formulations on soil nutrient availability and uptake by plants [77]. BGS (biodigester feed substrate was buffalo manure) enriched with ZnO NPs (1.4, 2.8, and 3.6 mg kg^{-1} soil) at a field application rate of 270 kg N ha^{-1} showed improved soil nitrogen content as compared to the lone application of BGS. They have found decreasing soil microbial carbon and C/N ratio with increasing ZnO NP concentration (amended with BGS), which is attributed to the increasing bacterial population. On reviewing the work of Aziz et al. [77] and multiple citations of peer-reviewed studies mentioned in their work, the role of NPs in increasing soil N and P for plant uptake is principally due to their influence on soil microbial population and consequently the secretion of extracellular enzymes like phosphatase (phosphorous availability) and urease (nitrogen availability) in soil. The increasing nutrient immobilization (such as N and P) in soil has associated risks of decreasing uptake by plants. It is also important to take note that, instead of increasing soil-N content, Aziz et al. [77] have not observed any significant increase in N uptake by plants with ZnO NPs + BGS treatments as compared to lone BGS, which necessitates further studies to investigate the other factors affecting shoot N uptake. Possibilities of biotransformation of ZnO NPs to zinc phosphate or phytate that can limit translocation of zinc into shoots also need to be studied further.

3.3.1 Nanoparticles in AD of biomass

The assessment of AD of biomass signifies the biogas yields coupled with the quality of recovered digestate. Trace elements are reported to have stimulatory effects on methanogens activity during the AD of biomass. It is interesting to compare the influence of NPs and their bulk counterparts on the AD during biogas production. We have reviewed the work of Abdelsalam et al. [165, 166] and Juntupally et al. [167], to get a better understanding of the subject matter in question.

Beginning with the findings of Abdelsalam et al. [165], in the case of cobalt treatments, the maximum biogas yield (BGSY) (HRT 31–35 days) was reported at 1 mgL^{-1} CoNPs. At the same concentration and HRT, CoCl$_2$ treatment gives lower BGSY, which was even lesser than the recorded yield

at 0.5 mgL^{-1} CoNPs. If we consider the risk of NPs toxicity at higher concentrations, the obtained results at low CoNPs concentration (0.5 mgL^{-1}) are comparable and better than those observed at higher concentrations of their bulk counterparts (1.0 mgL^{-1} CoCl$_2$) (Fig. 5a). Likewise, in the case of NiNP treatment, the BGSY (HRT 26–30 days) follows the increasing trend with increase in concentration (0.5 mgL^{-1} <1.0 mgL^{-1} <2.0 mgL^{-1}). However, the maximum BGSY (HRT 31–35) with 1.0 mgL^{-1} NiCl$_2$ was lesser than the recorded yields at all the concentrations of NiNPs (Fig. 5b). Overall, the higher BGSY and lower HRT to achieve maximum yields were noticed with NiNP treatment as compared to CoNPs. Hijazi et al. [168] made similar observations with bulk counterparts (mentioned in brackets) of NiNPs (NiCl$_2$), CoNPs (CoCl$_2$), and FeNPs (FeCl$_3$), and recorded maximum biogas production from manure treated with NiNPs (1 gm^{-3}) followed by CoNPs (1 gm^{-3}) and FeNPs (1 gm^{-3}) (~7.82% and ~18.87% higher as compared to CoNPs and FeNPs treatments respectively).

Abdelsalam et al. [166] performed another study on Fe-NPs (nZVI), Fe$_3$O$_4$ magnetite NPs (MNPs), and their bulk counterpart (FeCl$_3$). They have observed the highest BGSY in 20 mgL^{-1} Fe-NPs as compared to other treatments (Fig. 5c). Similar observations were recorded for Fe$_3$O$_4$ MNPs at the same concentration (Fig. 5d). The HRT was comparatively lesser and BGSY yield was higher in the latter case (Fe$_3$O$_4$ MNPs) at all the treatment concentrations. BGSY with bulk Fe (FeCl$_3$) at 10 mgL^{-1} was not comparable with those from NP treatments at all the investigated concentrations. Taking note of the size of synthesized NPs in both the studies, it was higher with CoNPs (28±0.7 nm) than NiNPs (17±0.3 nm) in the first study, and in the second study Fe$_3$O$_4$ MNPs (7.0±0.2 nm) were slightly smaller than Fe-NPs (9.0±0.3 nm). Thorough investigations are required to evaluate the effect of NP size on their performance during AD. Juntupally et al. [167] compared the biogas production potential of Co (CO$_3$O$_4$ NPs), Fe (Fe$_3$O$_4$ NPs), Mo (MoO$_3$ NPs), and Ni (NiO NPs) nanoparticles with their bulk counterparts, i.e., CoCl$_2$, Fe$_2$O$_3$, (NH$_4$)$_6$Mo$_7$O$_{24}$·4H$_2$O, and NiCl$_2$ respectively. All the digesters fed with NPs showed higher biogas production over their bulk partners, with maximum values recorded for NiNPs (21.2 nm) followed by FeNPs (5.6 nm), and CoNPs (37 nm), and MoNPs (Fig. 5e).

FeNPs have been investigated in most of the studies in the area of AD of biomass and subsequent biogas production [154]. Fe-based additives were reported to influence microbial activity during AD [154, 166, 169]. Yang et al. [170] performed microbial diversity analysis (nZVI supplemented AD of swine manure) and found an abundance of hydrogenotrophic *Methanomassiliicoccus* (39.2–92.0%). nZVI facilitates the reduction of "oxidation-reduction potential," which is reported to promote AD [155, 171]. Ye et al. [171] also mentioned that the strong
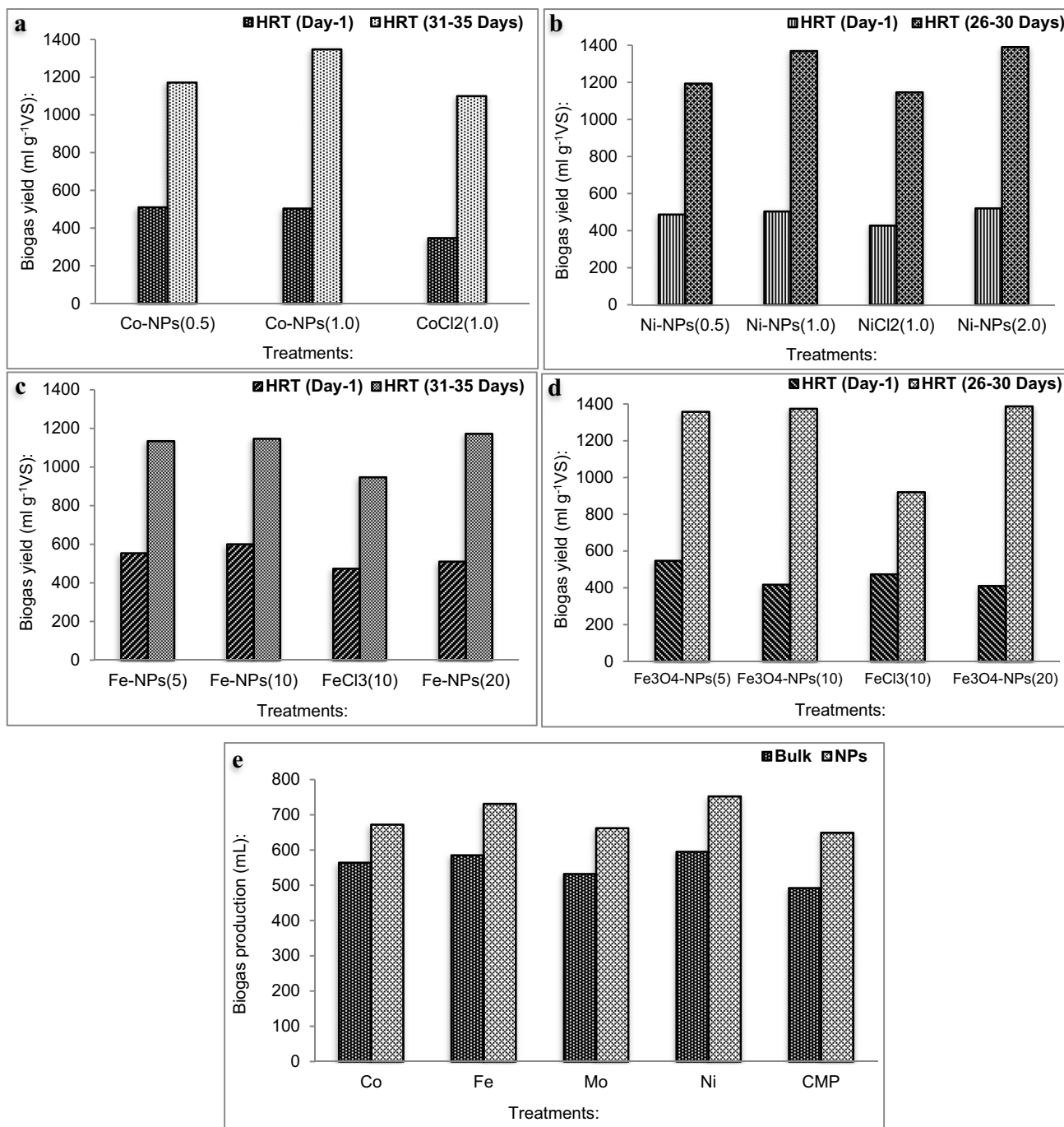
Fig. 5 Comparative analysis of the treatment of NPs and their bulk counterparts on AD of biomass leading to biogas production **a** Co-NPs and $CoCl_2$; **b** Ni-NPs and $NiCl_2$ [165]; **c** Fe-NPs (nZVI) and $FeCl_3$; **d** Fe_3O_4-NPs and $FeCl_3$ [166]; **e** CO-NPs: CO_3O_4, CO-Bulk: $CoCl_2$; Fe-NPs: Fe_3O_4, Fe-Bulk: Fe_2O_3; Mo-NPs: MoO_3, Mo-Bulk: $(NH_4)_6Mo_7O_{24}.4H_2O$; Ni-NPs: NiO, Ni-Bulk: $NiCl_2$; CMP: Composite treatment of Bulk/NPs (Bioreactor feed include 600 mL CM + 10 mg of NPs or bulk form or CMP of NPs/bulk form + 200 mL of microbial culture MMC-A) [167] (In graphs **a** to **d**, enclosed brackets in treatment categories on the X-axis indicates dose in mgL^{-1})

reduction ability of nZVI facilitates pollutant removal during AD via direct electron transfers. NPs such as Zn and Ni are involved in multi-enzyme complexes, thus becoming one of the preferred candidates for improvements in AD leading to higher biogas production. Ni is either associated with the cofactor F_{430} or bound to FE-S clusters, while Co is mentioned to take part in methyl group transfer [168, 172].

Amo-Duodu et al. [155] recorded higher methane yields (100%) from NP treatments (Fe-1g, Cu-1g, Ni-2g) as

compared to control (63%) (without NP treatment, wastewater as feed substrate only). NPs such as NiNPs alone or in combination with FeNPs and CoNPs reduce the H_2S content [151, 153], thus yielding good-quality fuel. Not only this; in the same study, researchers mentioned the increase in TS and VS removal efficiencies in the biodigester supplemented with NiNPs at 2 mgL^{-1} dose. Similar observations were reported by Abdelwahab et al. [151] with FeNPs (at 15 mgL^{-1}). pH was also recorded at the ideal range for optimal activity of microbes during the digestion process in both the studies at selective treatments. Juntupally et al. [173] found higher methane production from AD of pre-treated horse manure (HM) (dilute acid thermal pretreatment) added with 40 mgL^{-1} Fe_3O_4 NPs as an accelerant, though in untreated HM, they have found maximum yields with 60 mgL^{-1} Fe_3O_4 NPs. Overall, using the strategy of biomass pretreatment, the dose of NPs can be reduced as an accelerant during AD, with higher methane yields and reduced HRT. The addition of nickel NPs in biogas feed (CM) can improve the fertilizer potential of recovered effluents [151]. BGS (feed substrate buffalo manure) (field application at 270 kg N ha^{-1}) enriched with different doses of ZnO NPs (average size ≤40 nm; conc. 20% w/v in distilled water) (1.4, 2.8, and 3.6 mg kg^{-1} soil) improves soil nitrogen content [77]. The application of nanofertilizers can reduce nutrient leaching loss, thus promoting the efficiency of nutrient use [174].

Not only the elemental nanoparticles; other particles/materials of nano-dimensions have also been reported to have potential application in biomass degradation. Lignin degradation is one of the challenging tasks during biofuel production from the lignocellulosic feedstocks. Yang et al. [175] in an innovative approach reported 10% higher lignin breakdown (initial concentration of 50 mgL^{-1}) using nitrogen nanobubble water as compared to the control (during ACD with activated water sludge). An increase in activity of methanogenic bacteria during ACD is attributed to higher methane production as well. They have anticipated the role of hydroxyl ions generated by nitrogen nanobubble water behind such observations.

3.3.2 Nanoparticle-microbe interaction

As reviewed by Abdelsalam and Samer [172], the interaction of NPs with the biological systems can involve either direct uptake (via ATP dependent system; depends upon size and speciation of the NPs) or complexation with the dissolved organic matter (DOM) (on cell membrane DOM-NP complex binds with the MBRs). NP uptake by methanogens involved "metalloenzyme, electron transfer, and reduction pathways" during methane production [172].

Kumar et al. [176] reviewed the applications of conductive NPs (abbreviated as CNPs) to improve the efficiency of AD leading to higher methane production. The relatively low

efficiency of mediated interspecies electron transfer (MIET) as a consequence of diffusion limitations with H_2 as an electron carrier during AD limits the process of methanogenesis. In contrary to this, direct interspecies electron transfer (DIET) enables direct interaction (cell-to-cell electrons transport) between the methanogens and bacteria during AD and facilitates metabolic conditions favored higher rates of methane production.

Cruz Viggi et al. [146] via theoretical valuations cited that DIET permits higher electron transfer rates among syntrophic bacteria as compared to IHT (Interspecies H_2 transfer)/MIET. Basically, in IHT, the hydrogen acts as an electron carrier and mediates the transfer of electrons to the methanogens from secondary fermenting bacteria (SPB). Hence, the involvement of SPB in generating hydrogen as reducing equivalents requires energy and is thermodynamically not favored under standard conditions. Instead in DIET, the electron transfer is accomplished either via conductive pili or membrane-bounded cytochromes (c-type) without the requirement of such electron carriers; thus, DIET is more feasible in terms of energy requirements [137].

CNPs have been reported to play an important role in such microbial interactions for electron transfer during AD [176]. Zhang et al. [177] also investigated the role of magnetite-NP treatment favoring DIET and FE-S precipitation leading to improved methane yields. Through microbial community analysis, they have also predicted the role of magnetite-NPs in increasing methanogenesis over acetogenesis. For methane production, Yang et al. [170] found the addition of nZVI more beneficial during the methanogenic stage as compared to the acidogenic stage. Farghali et al. [178] reported enhanced methanogenic activity during AD of CM as a result of TiO_2-NP-assisted extracellular electron transfer (EET), and Ti–S clusters also brings reductions in H_2S levels. Wang et al. [179] characterize the Tungsten-based nanomaterials to establish DIET for improving AD of biomass. Attributes such as stable chemical properties during AD, zero band gap (results in higher electrical conductivity, potential to act as electron carriers during DIET), and co-existing of syntrophic bacteria and methanogens in biodigester as revealed from pyrosequencing studies yield conceivable pieces of evidence in support of tungsten-based NPs as one of the potential accelerants of AD. Van Steendam et al. [180] for improving AD via DIET recommends the development of appropriate characterization methods.

Bi et al. [139] found the increasing activity of hydrogenotrophic (HGM) and acetoclastic methanogens (ACM) during solid AD of chicken manure supplemented with Ni^{2+} and Fe^{2+} treatments. Yang et al. [141] with ZVI powder supplementation (5 gL^{-1}) found a higher relative abundance of *Methanoculleus* species (39.9%) as compared to the control experiment (34.6%) and these species belongs to hydrogenotrophic methanogens reported to thrive under high ammonia conditions (HACs)

during AD. ACM has higher substrate utilization potential; hence, a possibility of sustaining higher ACM activity during HACs through ZVI enrichment also existed that favors fixation of CO_2 into methane and sustained performance of anaerobes could also be attributed to the adaptive biochemical plasticity of these microbes under such stressed conditions [141]. The researchers also recovered ~66.0% of ZVI powder from the bioreactor after the experiment.

Kato et al. [181] observed that supplementing methanogenic microbes with hematite or magnetite nanoparticles (10–50-nm size) hasten methanogenesis (in terms of production rate and lag time) from ethanol and acetate. A schematic representation highlighting the role of Fe-NPs in AD of biomass is included in Fig. 6. The different routes leading to methane production has been reviewed keeping in view of both IHT and DIET process. Fe^{2+} ions through Fe–S precipitation also pave the route for H_2S mitigation. Methylotrophic, hydrogenotrophic, and acetoclastic are the three pathways of methane production during AD, and enzymes catalyzed in each step have specific metal requirements [182]. Hence, it is proposed to develop nano-elemental formulations to fulfill such requirements of participating

enzymes in all three production pathways and optimize the methanogenic activity for improved methane production.

NGS allows analysis of microbial diversity and dynamics during AD. Such investigations are vital to screen the most efficient microbes during the AD utilizing different biomass feedstocks and accelerants including trace metals and nanoparticles. In one such study, Eduok et al. [183] found abundance of *Methanosarcina* in the digesters spiked (at ~0.67 mL^{-1} L day^{-1} for 315 days) with the mixture of Ag NPs (20 nm) (0.01 mgL^{-1}), ZnO NPs (20 nm) (0.12 mgL^{-1}), and TiO_2 (21 nm) (0.08 mgL^{-1}). As per the previous reports, this archaeal genus possesses remarkable properties of multi-substrate utilization for biogas production, adaptations to NPs presence [183], and most important the capacity to catalyze all the major pathways leading to methane production during AD (acetoclastic, hydrogenotrophic, methylotrophic, and methyl reduction pathways) [183, 184].

Zhang et al. [87] reported nitrogen removal applying nanoparticle treatment, in which ZnO NPs at low concentrations (up to 5 mgL^{-1}) increase the activity of anaerobic-oxidizing bacteria (ANOB) and ammonia-oxidizing bacteria (AAOB)
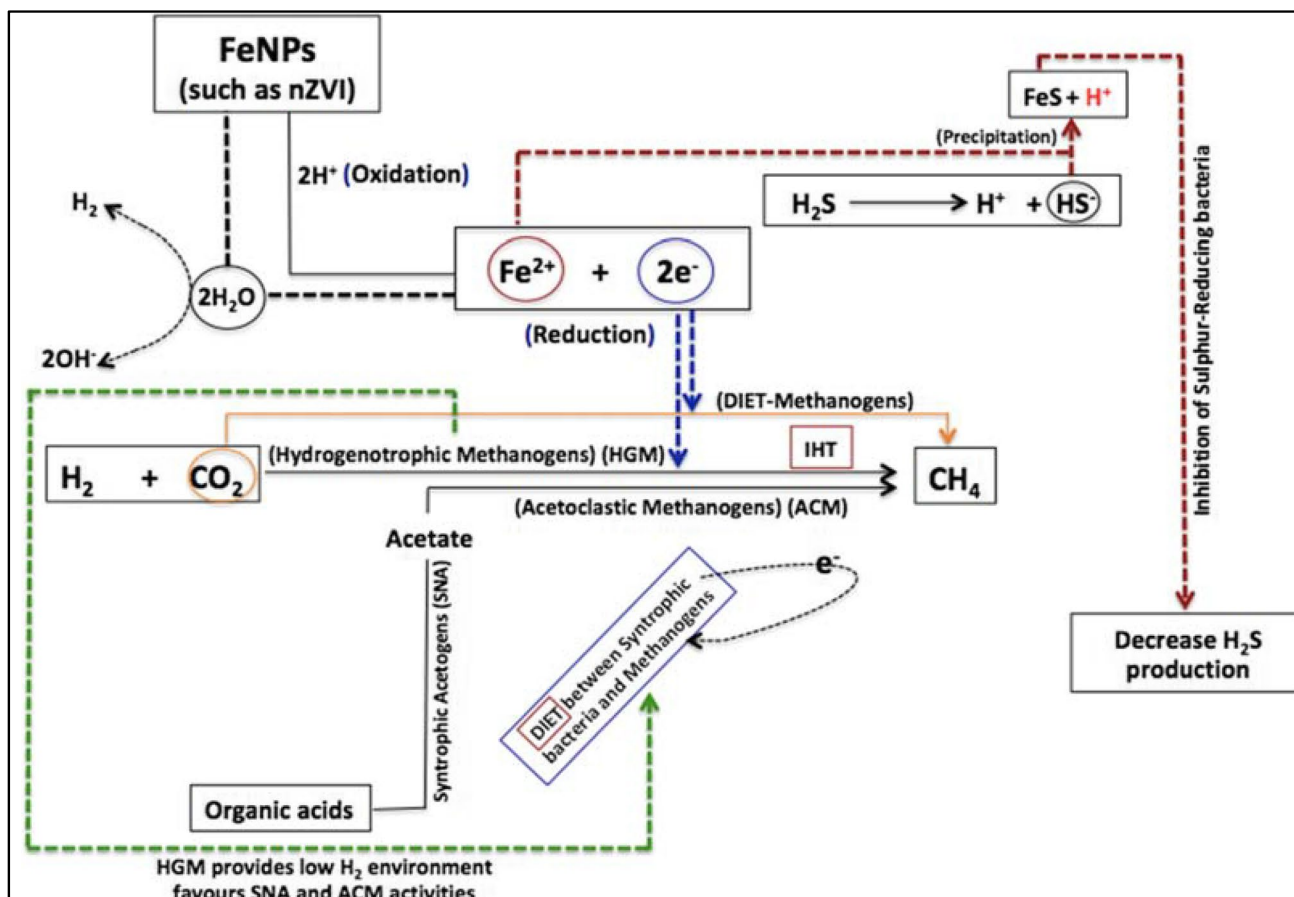

Fig. 6 Interaction between Fe-NPs and microbes during AD of organic substrates (source: conceptualized from the multiple authors referred in the manuscript)

to remove nitrogen from the sludge (Fig. 7). The activity of these bacteria inhibits nitrate formation through the suppression of nitrogen-oxidizing bacteria (NOB) activity. They have noticed the increase in microbial diversity at low concentrations of ZnO (Fig. 8). The OTU count and Shannon Index (SI) were highest at ZnO NPs 1 mgL^{-1} (OTU=998,
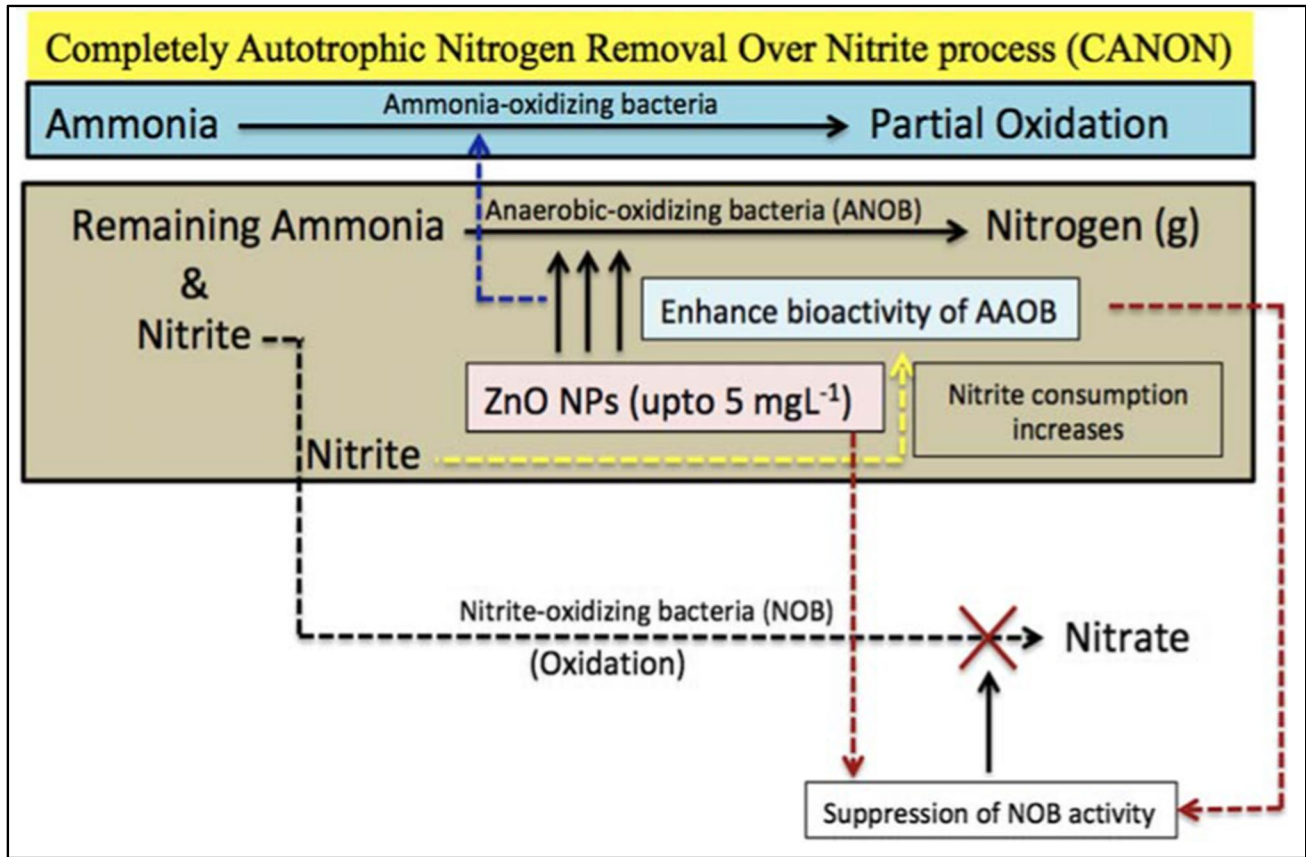

Fig. 7 Role of ZnO NPs in the removal of nitrogen from sludge during the CANON process (Conceptualized from [87])

Fig. 8 Effect of ZnO NP concentration on the relative abundance of bacteria participating in AD [87] (C=control experiment without ZnO NP addition; T1, T2, T3, and T4 are treatments with ZnO NPs concentrations 1, 5, 10, and 20 mgL^{-1} respectively, and T5 represents treatment in which no further addition of ZnO NPs done; B1: ammonia-oxidizing bacteria; B2: anaerobic-oxidizing bacteria; and B3: nitrite-oxidizing bacteria)
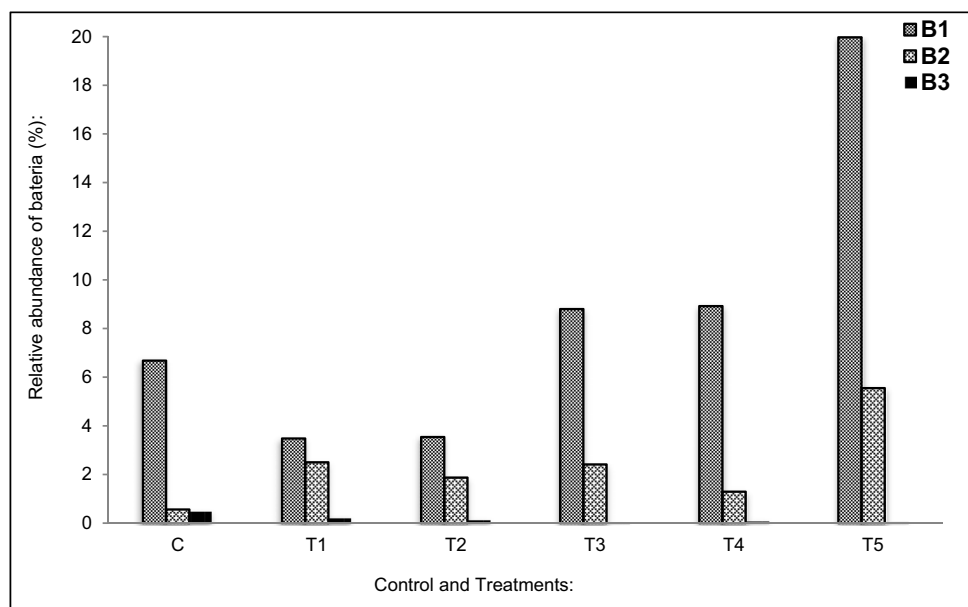
SI=4.39) followed by 5 mg L^{-1} (OTU=958, SI=4.34). They have also observed the improvements in nitrogen removal at these treatment doses. Lower concentrations of nanoparticles (Fe, Cu, and Ni NPs) favor nutrient supply for microbially inspired AD [155]. He et al. [185] in their investigation found higher sensitivity of microbes towards ZnO NPs at genus and phylum levels as compared to class level. Thus, nanoparticle treatment significantly alters the microbial community structure during AD of biogas feed substrates [86].

As stated above, nanoparticles play a significant role in the AD of biomass; this could serve as an opportunity to generate nutrient-rich digestate in addition to improved bioenergy efficiency. To achieve this objective, studies are warranted to advance our understanding of how nanoparticles interact with the microbes during AD, and the role of NPs to elicit the metabolic pathways in microbes followed by the screening and characterization of bioactive molecules was thereby produced. To further strengthen this, in our point of view, studies exploring the microbial diversity and dynamics in varied biomass feedstock and anaerobic digestates are vital, in which NGS could have an important contribution.

3.3.3 Nanoparticles co-aggregation

ZnO NPs are one of the highly used engineered nanomaterials [187]. AD systems encountered toxicity at higher doses of ZnO NPs. Tong et al. [188] in a phototoxicity analysis, reported ~40% disruption of the cellular membrane (CMM) in *E. coli* at 25 mgL^{-1} concentration of ZnO NPs, and such damaging effects were not visible at a lower dose of ZnO NPs (1 mgL^{-1}). Ye et al. [189] reported denitrification suppression by ZnO NPs in a dose-dependent manner (1, 25, and 50 mgL^{-1}) owning to a decrease in total nitrogen removal rates and N_2O emissions. They observed higher sensitivity of denitrification as compared to the nitrification process to ZnO nanotoxicity. However, to extenuate the effect of ZnO nanotoxicity (reported to be due to free Zn^{2+} and generation of reactive oxygen species like H_2O_2 and ·OH) on anaerobes during AD, Zhang et al. [190] performed a combined toxicity assessment of ZnO and TiO_2 NPs and observed decrease in Zn^{2+}. The previous studies also found a decrease in ZnO NP toxicity as a result of Zn^{2+} absorption in presence of TiO_2 NPs, as cited by [190].

Despite phototoxicity of TiO_2 NPs at 10 mg L^{-1} to *E. coli* (~65% CMMs damage), their treatment in combination with ZnO NPs (TiO_2 NPs 10 mgL^{-1} + ZnO NPs 1 mgL^{-1}) reported no toxicity, which appears to be eliminated by the ZnO NPs (ZnO NPs avoids close surface interaction of TiO_2 NPs/aggregates with bacterial CMMs) [188]. In another interesting observation, the authors reported that unlike in the case of phototoxicity, the TiO_2 and ZnO NP combinations (TiO_2 NPs 10 mgL^{-1} + ZnO NPs 1 mgL^{-1}) showed an additive effect in generating ROS. Now the question is despite generating higher ROS during combined treatment as a consequence of additive effect, how does this combination minimize the toxicity damage? They have proposed some possibilities behind such observations. One of these is, in case of combined treatment with concentrations, i.e., TiO_2 NPs 10 mgL^{-1} + ZnO NPs 1 mgL^{-1}, the formation of ZnO–TiO_2 aggregates established diminished contact with the CMMs of bacteria; thus, generated ROS in the majority did not make contact with bacterial CMMs, despite additive effects. Moving ahead, the authors also found that the combined treatment effect (in terms of phototoxicity) of TiO_2 NPs 10 mgL^{-1} + ZnO NPs 25 mgL^{-1} was comparable with that of their individual treatment dose (i.e., TiO_2 NPs at 10 mgL^{-1} and ZnO NPs at 25 mgL^{-1}); for this, the authors have mentioned that the larger ZnO NPs blocked the contact of several TiO_2 NPs with bacterial CMMs.

Further studies are duly recommended to examine the role of ZnO NP interaction with different components of AD (such as IET, syntrophic bacteria and methanogen activity, H_2S mitigation) for improved levels of produced methane coupled with the nutrient profile of recovered digestate (solid and liquid fractions). Besides this, integrated approaches must be explored to reduce the possibilities of ZnO NP toxicity interfering with AD of biogas feedstocks (precisely while using the wastewater sludge, or similar waste of anthropogenic origin). As stated earlier, the ZnO NP toxicity is mainly attributed to their dissolution and availability as free Zn^{2+} in the system [187, 188, 190, 191]; thus, investigating the suitable adsorbents or trace element/NPs co-aggregates will provide an opportunity to recover zinc in the digestate, which can be further utilized as a potential fertilizer in the zinc-deficient soil.

3.3.4 Environmental impacts

Nanotechnology-driven AD of biomass reported to enhance biogas/methane production; however, utilization of spent BGS must be investigated for toxicity analysis (on plants, beneficial microbes, etc.) before using as a fertilizer source for agriculture applications due to the associated risk of NP accumulation [78]. Hijazi et al. [192] performed the life cycle assessment (LCA) study to estimate the environmental impact of NP introduction (CoNPs: 1 g m^{-3}, NiNPs: 2 g m^{-3}, FeNPs: 15 g m^{-3}, Fe_3O_4 NPs: 20 g m^{-3}, and without NP treatment served as a control) during AD of manure for biogas production. For the analyzed six parameters (in reference to the electricity production by NPs treated and control biomass), CoNP treatment was best in terms of lowest: GHG emission, acidification, eutrophication, and human toxicity potential. NiNP treatment has the lowest risk of resource and ozone layer depletion. The representative view highlighting trends in the variation of environmental

indicators associated with the electricity generation from the NPs treated biomass and control experiment is shown in Table 6. It has been clearly observed that the untreated biomass (control) presented with the highest risk with reference to all the investigated environmental indicators. Considering an overall environmental impact, Hijazi et al. [192] recommended CoNPs for biomass treatment (AD) and electricity generation through obtained biogas.

Nguyen et al. [193] performed toxicity analysis of metallic nanoparticles on *E. coli* before and after AD and observed higher toxicity in samples before AD. Garcia et al. [194] observed 100% and 33–50% toxicity of CeO_2 NPs and Ag NPs on biogas production respectively. However, there was no or negligible toxicity of Au NPs, and TiO_2 NPs were reported towards oxidative heterotrophic organisms, ammonia-oxidizing bacteria, and anaerobic biomass. Dispersion of ZnO NPs caused 99% *E. coli* cell death at 100 mgL^{-1} [193]. CeO_2 NPs at l0 mgL^{-1} showed a positive effect on the *E. coli* population [193]. Zhang et al. [186] in a study on AD of sludge using NPs (carbon NPs, ZnO NPs, CuO NPs, and Al_2O_3 NPs) reported an abundance of mobile genetic elements in Phylum *Proteobacteria* followed by *Firmicutes*. The nZVI (55±11 nm) at 30 mM concentration was reported to inhibit methanogenesis as a result of disruption of the microbial cell membrane and rapid H_2 production [195]. Ma et al. [196] found an nZVI concentration of 160 mgL^{-1} suitable for reducing antibiotic resistance genes during AD of CM and also reported an increase in methane yield (6.43%). As per Zheng et al. [197], the variations in bioavailable fractions of heavy metals (like Zn, Ni) during AD (swine manure was feed substrate) depend upon three important digestion variables, i.e., concentration of ammoniacal nitrogen <methane content in daily biogas yield <pH (arranged as per the order of importance reported in the study).

Hassanein et al. [198] utilized the recovered effluent from the digester as a fertilizer source for *Lactuca sativa*. Despite

approximately similar levels of N, P, K, Zn, Mn, Cu, and Na in both the digester effluents, i.e., NP fed (NPD) (continuous digester containing NP mixture, i.e., FeNPs 1000 mgL^{-1}, NiNPs 120 mgL^{-1}, and CoNPs 54 mgL^{-1} fed twice on days 84 and 202) and control (CND) (biogas feedstock was cow dairy manure in both the digesters), the *L. sativa* fertilized with NPD effluent showed higher amount of these nutrients in its biomass as compared to those fertilized with CND effluent (both the digester effluents were diluted to 1:13.8 using deionized water (7.6 L) divided into five loadings over the growth period of 57 days). This could be attributed to the increasing nutrient uptake and translocation efficiency as a consequence of NP treatment. The recorded values of Fe, Ni, and Co in *L. sativa* were below the reported toxic limits. However, further investigations are much indeed in the area of NP introduction to the various levels of the food chain and in-depth assessment of their toxicity leading to the health and environmental implications. The authors have recommended such studies as well.

The non-biodegradable nature of metallic particles possesses a risk of toxicity [147] in the concerned environment. In the investigation of Luna-delRisco et al. [147], it has been observed that one of the reasons behind the toxicity of such particles is attributed to their aqueous solubility (AQS). They have reported poor AQS of CuO NPs, ZnO NPs, and their bulk counterparts, though it was noticed higher AQS of CuO NPs than their bulk counterpart that can partially be contributed to the higher toxicity potentials. The order of toxicity to anaerobic bacteria was reported in the following order: CuO NPs (~30 nm)> ZnO NPs (~50–70 nm)> ZnO Bulk> CuO Bulk. Taking account of recovery of Cu^{2+} ions (at higher CuO NP treatment doses, i.e., 120–480 mgL^{-1}) in liquid fraction of digestates, the proportion was highest at CuO NP concentration of 240 mgL^{-1} (~14.5%) followed by 480 mgL^{-1} (~9.94%) and 120 mgL^{-1} (~6.58%). Hence, the concentrations and types of NPs can significantly affect the microbial diversity during AD. As compared to control (without the addition of NPs), Hui and Kui [199] observed a higher sludge humification index (20.7–49.6%) during the process of vermicomposting in treatments with ZnO and TiO_2 NPs, which is attributed to the rich microbial diversity supported by NPs, as evidenced from the presence of organic acids, aromatic compounds, and polysaccharides in the final vermicompost product. The higher nanoparticle concentrations will also affect the process of AD of sludge (methanation, hydrolysis, acidification) as observed by Mu et al. [200] in the case of increasing ZnO NP dose. They have also noticed the inhibition of enzymatic activities at such doses. Furthermore, to explore this area in greater detail, studies related to the environmental exposure of nanoparticles and their toxicity analysis are important. Nanotoxicity studies (phytotoxicity, cytotoxicity, and genotoxicity) must be accomplished with thorough investigations on the

Table 6 Trends in the variation of environmental indicators associated with the electricity generation (bioenergy) from the NPs treated biomass and control experiment [192]

Environmental indicators	CoNPs	NiNPs	FeNPs	Fe$_3$O$_4$ NPs	Control
GHGs emission	1	2	4	3	5
Acidification	1	4	3	2	5
Eutrophication	1	3	2	4	5
Resource depletion	3	1	4	2	5
Ozone layer depletion	2	1	4	3	5
Human toxicity potential	1	3	4	2	5

*Numbers from 1 to 5 indicate an increasing trend of the environmental indicators impact, i.e., 1<2<3<4<5.

impact of nanoparticle types along with their shape, size, and concentration.

3.4 Nutrient recovery from BGS

Direct applications of BGS in agriculture fields have certain concerns regarding its physical (presence of debris such as plastics, glass, etc.), chemical (pH, nutrient composition, presence of heavy metals), and biological qualities (presence of pathogens) [13]. This will consequently entail some pre-conditioning or value additions. On surveying literature, different methods of nitrogen removal/recovery from the liquid fraction of BGS such as Anammox, membrane separation, absorption, and struvite crystallization, were encountered [13]. Solid digestate can be exploited directly or subjected to composting or drying and used as a rich source of phosphorous [13]. On the other hand, the liquid fraction of the digestate (LFD) is of great interest for researchers as it provides N and K. The difference between nitrogen removal (converting ammonium into nitrogen gas) and recovery (ammonium fixation to be further utilized as N source in agriculture fields) from the LFD is imperative. Menkveld and Broeders [201] described the Nijhuis Ammonium Recovery System (NAR) based on chemical stripping and asserted removal efficiency of ammonia between 85 and 90% from AD manure or organic wastewaters. Schematic representation of the NAR system has been mentioned in Fig. 9a. Here strategically, CO_2 stripping has been done through the introduction of fresh air; consequently, the pH increases and the requirement of NaOH decreases. Higher pH in ammonia stripper increase ammonia volatilization and ammonium phosphate recovery can reach 25–40% under optimized conditions, as stated.

Nitrogen recovery using chemical stripping is an energy-extensive process with high temperature and chemical treatments (NaOH and H_2SO_4) [202]. Battista et al. [202] in an innovative step performed recovery of nitrogen as ammonium sulfate (~2M, yield under optimized process) from ADDC using solar energy-driven lab-scale greenhouse (Fig. 9b). The recovered solid digestate was evaluated as P-source on growth in maize and results were comparable to commercial triple superphosphate fertilizer under observed greenhouse pot experiments. Xu et al. [134] used vermiculite (P content 0.1%) as an accelerant during ACD of *Aloe* peel waste and dairy manure for biogas production and found maximum N (%), K (%), and total nutrient content (TNC) (%) in digestate with vermiculite (0.3%) (DVM) as compared to control and other treatments (digestate with 0.1%/0.6%/0.9% vermiculite). The accelerant property of vermiculite favoring biomass decomposition could be attributed to the release of trace elements such as Fe^{3+}, Ca^{2+}, Mg^{2+}, Zn^{2+} and Al^{3+}, as cited by them. Compared to control, DVM-0.3% have ~12.57%, ~15.07, ~25.71, and ~15.42% higher N, P, K, and TNC respectively. Ehmann et al. [203]

also recovered P from anaerobic digestates. Gienau et al. [204] in an optimized membrane treatment process (centrifugation, ultrafiltration, and reverse osmosis) recovered nutrient-rich solid (total nitrogen: 8.2–12.0 kg/t, P_2O_5: 5.6–10.4 kg/t) digestate and LFD (NH_4^+–N: 2.9–5.6 kg/t, K^+: 6.2–9.2 kg/t).

He et al. [205] reported optimum pH, removal temperature, and pressure of 13.04, 35°C, and 15 kPa respectively for the removal of ammonia-nitrogen (AMN) through vacuum distillation. They have concluded that under the conditions of high pH, vacuum distillation of BGS can yield higher removal rates of AMN. In a "vacuum membrane distillation" (VMD) process, under alkaline pH (10) of bioslurry, He et al. [206] recorded over 90% conversion of TAN into free ammonia. They also observed that the alkaline treatment during the VMD process substantially reduces the BGS phytotoxicity. In both studies, recovered ammonia can act as a potential CO_2 absorbent. In another study, He et al. [207] recovered free ammonia from BGS using the VMD process, and the recovered ammonia showed better performance as a CO_2 absorbent in upgrading biogas as compared to the other tested absorbents. Chen et al. [208] used vacuum-assisted gas-permeable membrane (V-GPM) for ammonia recovery from LFD and reported over 80% removal (In 6 h, 70 °C temperature, and 30 kPa pressure). The technique was reported to have negative GHG emissions [209]. García-González et al. [210] recovered ammonia from digestate (obtained from ACD of pig manure and agro-food waste) using gas-permeable membranes with 71% efficiency of N recovery. The carbonaceous materials are retained with the digestate using this technology as cited by García-González et al. [210]. The ammonium ions in LFD can also be transformed to the gaseous phases such as hydrogen, ammonia, and N_2O through hydrolysis, stripping, and Nitrification-denitrification process respectively, for further utilization as an energy source [211]. For recovery of ammonia and biogas (up-gradation) from the wastewater digestates of high strength, a hydrophobic membrane-based ammonia recovery system link coupled with an electromethanogenic microbial electrolysis cell has been reported [212].

Taking account of the adsorption process for nutrient removal, biochar has been explored as one of the low-cost adsorbing materials with promising outcomes [213]. Heterogeneity in biochar surface facilitates different adsorption mechanisms [214]. Adsorption mechanisms for nutrient removal from BGS using iron modified biochars (pyrolysis of corn straw at 690°C temperature) include flocculation, electrostatic attraction, functional group reaction, ion exchange in case of nitrogen, flocculation, co-precipitation, ligand exchange, and surface complexation in the case of phosphorous [213]. Maximum adsorption values recorded using iron-modified biochars were 11.68 and 26.14 mg g^{-1} for nitrogen and phosphorous respectively [213]. Kubar
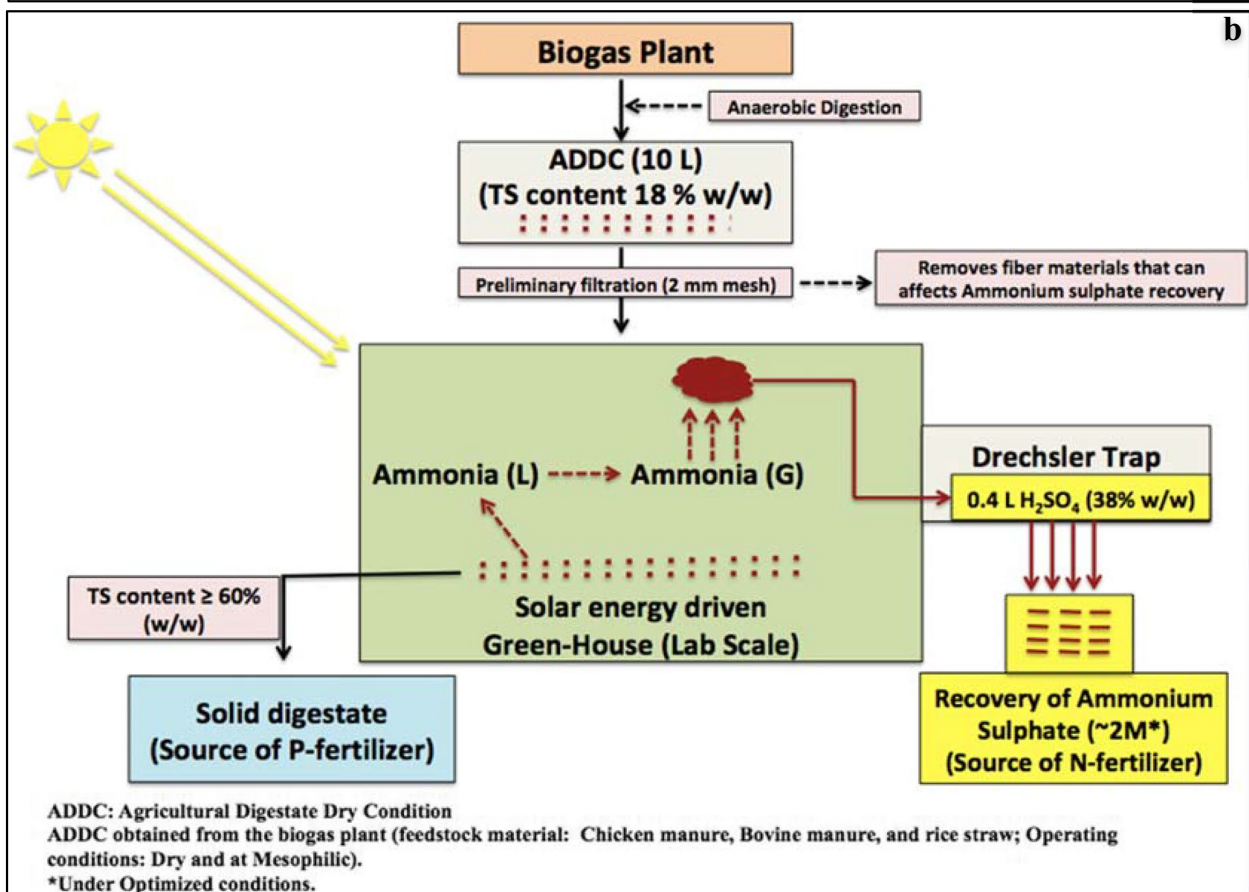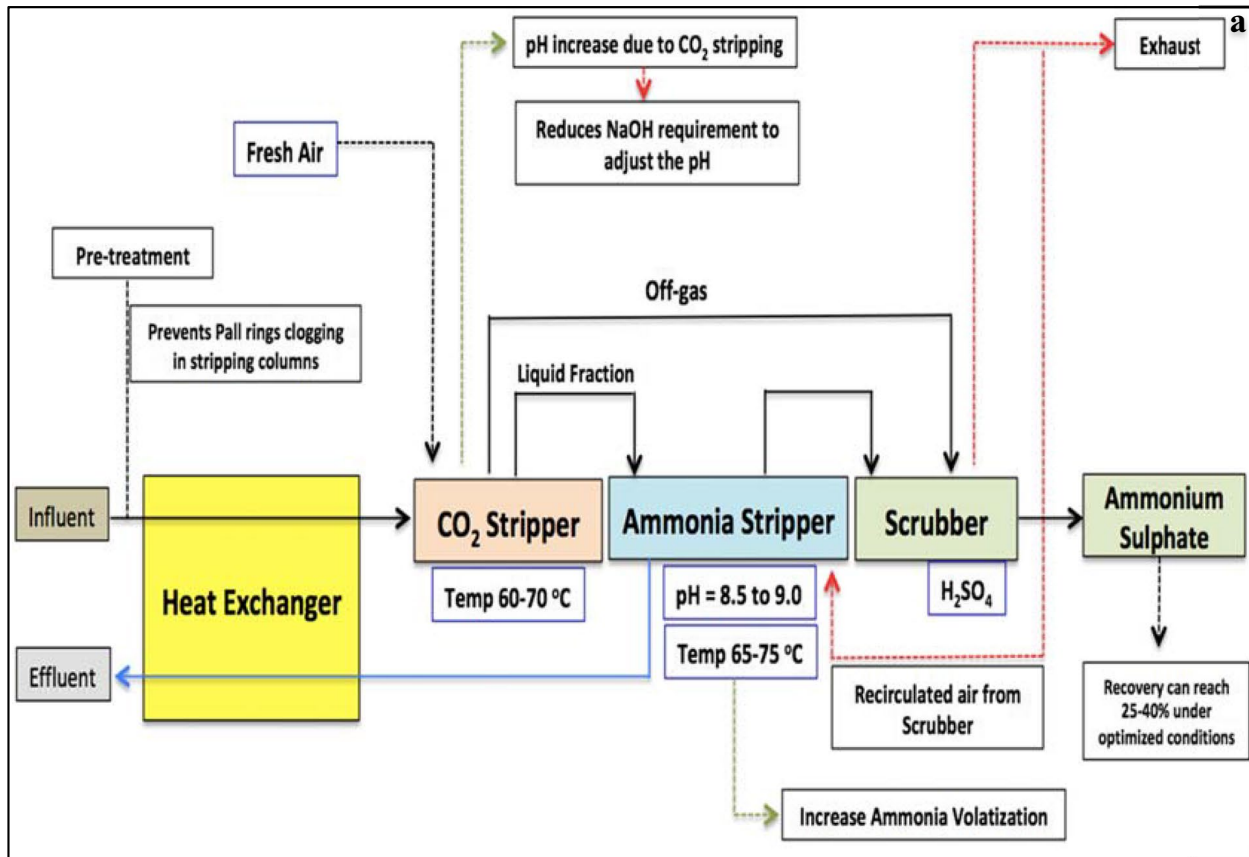
et al. [215] in a biochar and struvite co-precipitation method recorded maximum recovery of P (45.36% with $ZnCl_2$) and NH_4^+ (33% with $NaHCO_3^-$) using rice biochar and Tongan sludge urban biochar respectively (adsorbent dose = 0.2 g).

Biochar can be modified through different acid, alkaline, coating, magnetic, steam, and nanoparticle-based treatments for recovery of heavy metals, organic compounds, and nutrients such as nitrogen and phosphorous [214]. Biochar obtained from pyrolysis of anaerobic digestate of algae dairy manure upon treatment with 2M KOH showed higher sorption (50.71 mg g^{-1}) of Cu(II) as compared to untreated biochar (21.12 mg g^{-1}) [216]. Hardwood biochar (114.2 mg g^{-1}) showed maximum sorption capacity of ammonium as compared to corncobs (108.9 mg g^{-1}) and mixed sawdust pellets (24.7 mg g^{-1}) biochar from swine BGS (anaerobic digestate) (influent ammoniacal nitrogen concentration of 500 mg L^{-1}, inflow rate 15 mL min^{-1}) (pyrolysis temperature of 600 °C at 10 °C/min for 8 h) [217]. Kizito et al. [218] in another study reported maximum sorption capacity of ammonium by wood biochar (44.64±0.602 mg g^{-1}) followed by rice husk biochar (39.8±0.54 mg g^{-1}) from piggery bioslurry (anaerobic digestate) (influent ammoniacal nitrogen concentration of 1400 mg L^{-1}) (pyrolysis temperature of 600 °C for 10 h). Luo et al. [219] recovered 20% of nitrogen (30 mg L^{-1} day^{-1}) and 80% of phosphorous (7 mg L^{-1} day^{-1}) from BGS (piggery) using a novel photobioreactor under optimum operating conditions.

BGS have solid and liquid fractions, and to maximize their full potential as a nutrient source, the focus must be paid towards the exploitation of both these fractions. The LFD also aids bulkiness to the BGS and thereby limits its extensive applications in the agriculture fields as a consequence of challenges such as space limitations, long-term storage, and transportation. In general, LFD has low nutrient contents; hence, its further concentration or transforming to solid compounds has been recommended to facilitate the ease of transport [211]. It can also be amended with the mineral elements [35] and AIMs to develop value-added products. Additionally, the LFD can also be processed for the recovery of nitrogen; aiding to this, the latest technological developments have been reviewed and discussed [87, 201, 202]. The microalgal biomass harvested from such liquid fractions can also bioaccumulate N, P, and traces of micronutrients as well [13]. Sobhi et al. [220] explored heterotrophic indigenous microflora for the simultaneous achievement of biomass production (>6 gL^{-1} with lipid, crude protein, ash content, and gross energy of 10.9%, 65%, 10.7%, and 19.6 MJ kg^{-1} respectively) and nutrient recovery (~68% phosphate and ~97% of total nitrogen) from the LFD.

Apart from the nutrient recovery, the resultant biomass can be further exploited as an animal feed or bioenergy feedstock. Thus, microalgal and microbial-based biosorption methodologies will be of great interest to serve multiple purposes such as wastewater treatment, generation of nutrient-rich biomass, and potential feedstock for biofuel production.

4 Review of commercial viability and economic estimates

Interpretations of the BGS values can have multifaceted outlooks, such as economic fertilizer to agronomic estimates (Fig. 10). In beginning, the biofertilizer market has been reviewed to provide a brief overview of the existing market for organic agri-inputs. As per the biofertilizer market report (May 2021), the global market in 2021 for biofertilizers is estimated to be 2.6 billion USD, which is expected to be 4.5 billion USD by the year 2026 [222]. In another report published in June 2020 [223], the global biofertilizer market valued at 1.0 billion USD in 2019 is projected to achieve a CAGR of 12.8% from 2020 to 2027.

Moving to field valuations, an estimated return of INR 32400 can be obtained from the 10.8 ton of dried (25% moisture) BGS (at INR 3 per kilogram) (biogas plant of 2 m^3 capacity with daily feeding of 50 kg cattle dung and an equal amount of water, can produce 10.8 ton of dried BGS annually), as mentioned by [36]. Shaibur et al. [4] in their survey study on the utilization of CD in Ziala village (Satkhira District, Bangladesh) by sampled owners (twelve) estimated yearly savings of 10447 kg of chemical fertilizers, which is equal to 470115 Bangladeshi Taka per year (~ 5547.97 USD, as of 17 May 2021, 14:32 UTC, Xe Currency converter). Rachmah et al. [224] reported the cost of bioslurry fertilizer between 520 and 1000 IDR/kg (equivalent to 0.037–0.070 USD, as of 15 May 2021, 18:12 UTC, Xe Currency converter) in their study in central Java (Indonesia) region during January–February 2020. While making economic estimates, an important factor is the handling and transportation cost associated with the BGS. We have reviewed some of the related studies to explore the transport options and related costs, though such costs vary and depend upon multiple factors such as available infrastructure, technologies for conditioning, processing, and management of slurry. In one of the earlier studies of Ghafoori et al. [225], the on-road transport cost of liquid digestate through truck ("standard 40 t tandem trailer truck") (in Alberta, Canada) was estimated as 3.9 USD t^{-1} (distance fixed-cost) and 0.064 USD t^{-1} km^{-1} (distance variable cost) (refer to [225] for more details). Taking into account the cost associated with the feedstock material transport to the biogas plant, we have encountered a few studies. Zheng et al. [226] reviewed the economic scenario of one of the representative bio-natural

Fig. 10 A schematic view representing prospective values of BGS (conceptualized from [221])
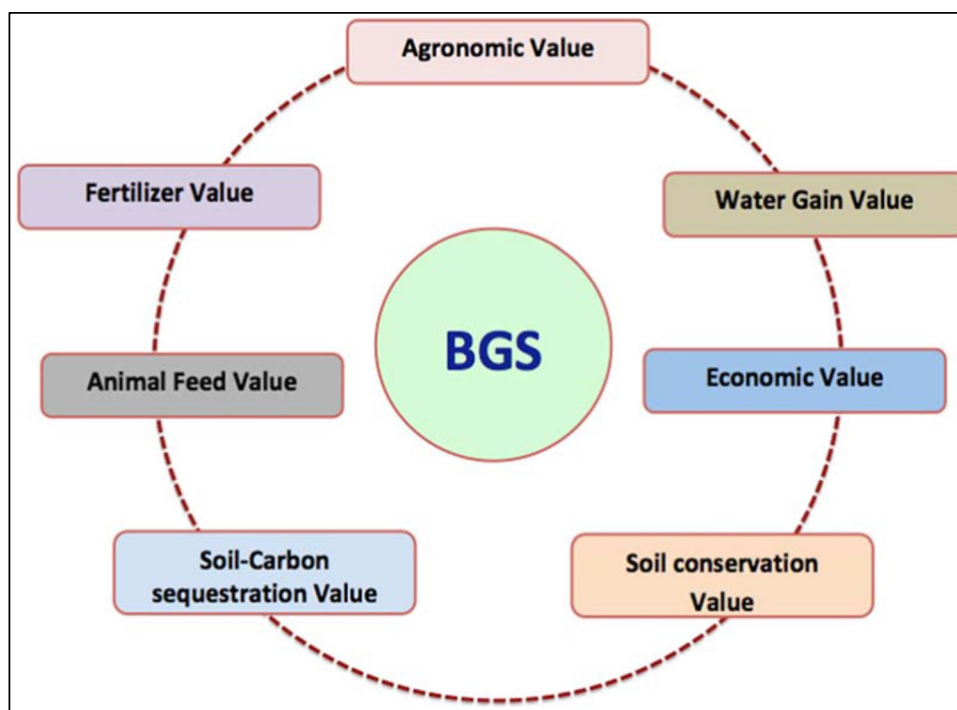
gas production projects in Europe; here the estimated cost for feedstock transport was mentioned as 6 Euro/t. According to Ghafoori et al. [225], transportation cost to bring feedstock (beef-cattle manure) material to a large-capacity centralized digester plant can be reduced using the pipeline as compared to the truck-based transport systems.

Utilization of LFD for recovery of nutrients can reduce the bulkiness of BGS and somehow the higher transportation cost as well [211]. The BGS enrichment studies and developing value-added products can be a considerable alternative to further reduce transport costs. Romero-Güiza et al. [13] reviewed the pelleting process for BGS solid fraction and cited that the price for the finished product can reach up to 250 euros per ton. Chen et al. [208] through vacuum-assisted gas-permeable membrane process of ammonia recovery estimated an additional benefit of 0.51 USD per kilogram of N. Menkveld and Broeders [201] estimated the nitrogen recovery cost per kilogram ranges from 1.0 to 3.0 euros using the NAR system (efficiency between 80 and 90%), while for Anammox (efficiency up to 75%) and MBR (efficiency up to 75%), they have mentioned the figures ranges from 1.5 to 3.0 and >5.5 euros respectively.

The highest benefit to cost ratio of 3.01 was reported in BGS enriched with Panchagavya (5 kg CD/BGS 12 ripe banana, 3 L tender coconut water, 1 L cow ghee/butter clarified, 3 L cow urine, and 2 L each of cow milk and curd) as compared to other organic treatments [130]. Rana et al. [227] performed bio-fortification of Cu, Zn, Fe, and Mn with combined inoculation of bacterial and cyanobacterial strains in rice-wheat cropping sequence and recorded yield benefits

of 40–60 kg N ha^{-1} savings (applied as urea) equivalent to 924–1386 USD (based on conservative estimates of area under rice and wheat crops in India and China).

Based on the NPK content in BGS, CM [33], N-DOC [228] (average value calculated from the multiple reported data), K-DOC [21], J-DOC, and J-BGS [229], economic values have been estimated theoretically (conceptualized from [221]). The price of N, P, and K fixed by the Department of Fertilizers, Ministry of Chemicals and Fertilizers, Govt. of India, under nutrient subsidy rates (NBS) per kilogram of N/P/K for the year 2020–2021 (Department of Fertilizers, Ministry of Chemicals and Fertilizers, Govt. of India) [230] was considered for the calculation. As observed from Fig. 11, value addition of BGS with DOCs resulted in higher N and P content, ensuring its higher price values as well.

Abdelwahab et al. [154] reported ~38.20% higher biogas production from CM treated with FeNPs (15 mgL^{-1}) during AD, as compared to the control (CM without NP treatment). An increase in ~1.16% N and ~3.63% K was also noticed in recovered digestate (treated with FeNPs) as compared to control, though the P content was 0.94% less in treated digestate. Hassaneen et al. [182] recovered nutrient-enriched digestate from the AD of NP-treated (zinc ferrite, average crystallite size 6.22 nm) CM. Interestingly, the recovered digestate (HRT-50 days) has higher P content (3.50%) along with N and K content of 3.40% and 1.30% respectively. Abdelwahab et al. [231] in cost analysis for biogas production reported net gains of 20.6, 19.7, and 13.5 USD with NiNP supplementation at 2.0 (NiNPs-2.0), 1.0 (NiNPs-1.0), and 0.5 (NiNPs-0.5)

Fig. 11 An economic value analysis of Organic manures based on the NPK content (A1: BGS, A2: CM; A3: BGS + N-DOC; A4: BGS+K-DOC; A5: BGS + J-DOC; A6: J-BGS; CM: cattle manure; N-DOC: neem deoiled cake; J-DOC: Jatropha deoiled cake; K-DOC: Karanja deoiled cake; BGS: biogas slurry from CM; J-BGS: BGS from the AD of J-DOC; combinations in A3 to A6 (50:50); MT: metric ton; INR: Indian rupees)
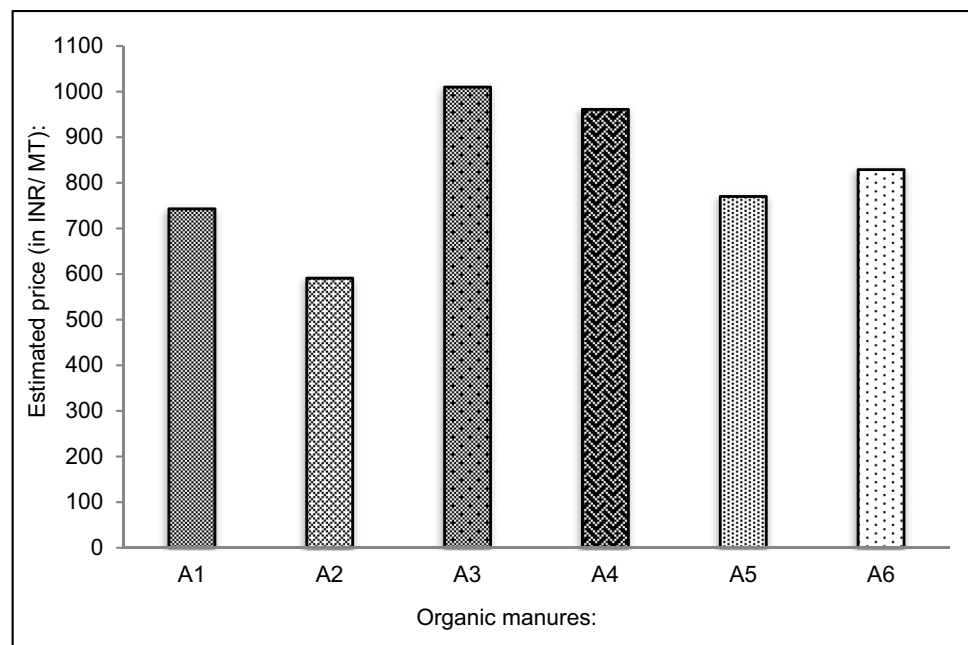
mgL^{-1} concentrations respectively. The estimated costs in NP synthesis (including chemicals) were 0.7, 1.5, and 3.1 USD with NiNPs-0.5, NiNPs-1.0, and NiNPs-2.0 respectively. Studies on ACD of BGS and DOCs coupled with NP supplementation will be of great interest for in situ recovery of nutrient-rich digestate and improved biogas yields. The recovered digestates can be further exploited as a suitable substrate for the mass multiplication of AIMs, owning to which bioformulations of the broad spectrum will be developed to meet the agriculture requirements. Additionally, the liquid fraction of digestate can also be processed for the recovery of nitrogen; aiding to this, the latest developments such as solar-driven technology [202], energy-efficient chemical stripping [201], and nanoparticle-assisted CANON process [87] have been discussed. The area is much promising and has immense potential to demonstrate multifaceted opportunities of generating greater revenues for the rural community where these resources (CD, BGS, DOCs) are in surplus availability. To further extend this, it is recommended to explore other associated values of BGS (mentioned in Fig. 10), as this will encourage the diverse stakeholders concerned to develop BGS-inspired value-added products that can act as a catalyst for rural development in terms of sustaining virtuous livelihood.

Some of the patents related to the recovery of resources from the BGS are mentioned below:

1. Patent title: Producing liquid fertilizer in a biogas plant. Publication number: US9957201. Publication date: 01 May 2018. Inventor: Liebeneiner, Rolf. Current Assignee: Bekon GMBH.

2. Patent title: Method for cycling biomasses between mushroom cultivation and anaerobic biogas fermentation, and for separating and drying a degassed biomass. Publication number: WO/2015/007290. Publication date: 22 Jan 2015. Inventors: Hoff, Svend Kristian and Pedersen, Lars Jørgen. Applicants: Advanced Substrate Technologies A/S.

3. Patent title: Lightweight assemblable appliance and respective method for production of biogas and liquid fertilizer. Publication number: WO/2014/203047. Publication date: 24 Dec 2014. Inventors: Efrati, Oshik Moshe; Teller, Yair; Lanzer, Erez; Miller, Yariv; Eilon, Tal; Zak, Shoham. Applicants: Ecogas Israel Ltd.

5 Summary, perspectives, and conclusions

The present review aims to cater the wider reader base, and efforts have been made to pay more attention towards the technical aspects linking to the conventional approaches to benefit the rural community and concerned stakeholders at large. The agricultural utilities of BGS have been covered in the post-introduction section of the manuscript. While gathering preliminary data during the literature survey, we have encountered some challenges that limit BGS applications in agriculture fields, despite the immense potential to compete with the available chemical and organic fertilizers and formulations of biocontrol activities. Here, we have reviewed the available literature and technological interventions in the area of bioslurry management and proposed some solutions with special reference to the bio/nanotechnology-based approaches aiding improvements and value addition to the

BGS. BGS is the by-product of anaerobic digestion, where microbes are the key players, and hence we have focused our review towards microbially inspired approaches to upgrade the BGS. In situ value addition designated to the methodologies operated within the digester, while ex situ approaches deal with the recovered digestates.

To enhance the microbial activity during AD of biomass, the C/N ratio is critical, which can be balanced through opting ACD of livestock manure with other organic materials such as NEDOCs. This, in addition, facilitates nutrient-rich media for the microbial and enzymatic activities during digestion, which could serve twin benefits of improved BGSY and recovery of nutrient-enriched digestate. The mechanism of nanoparticle-microbe interaction was also taken into consideration and the possible routes to develop an understanding of how NPs influence the interspecies H_2 transfer, and direct interspecies electron transfer, has been discussed. As per the reviewed literature, NPs such as FeNPs (nZVI) have been reported to have a potential role in establishing direct interspecies electron transfer between syntrophic bacteria and methanogens. FeNP interaction during AD also has considerable effects on reducing H_2S levels. Collectively, NP treatment facilitates higher biomethane yields with quality improvements and reduction in HRTs. In recent developments [149, 150], biomass material has been treated with laser irradiation coupled with nanoparticles during AD to achieve biostimulation of methanogens for improved biogas production.

To maximize the full potential of BGS as a nutrient source, both solid and liquid fractions need to be exploited. Recovery of nitrogen from the LFD is also a promising area to generate revenues. Several chemical methods have been reported for such recoveries; we have reviewed some of the latest and energy-efficient developments in the area (NAR system; V-GPM; solar energy-driven lab-scale greenhouse setup; hydrophobic membrane-based ammonia recovery system link coupled with an electromethanogenic microbial electrolysis cell; microalgal and microbial-based biosorption methodologies, etc.). Not only the biogas production; NPs can also be investigated for the recovery/removal of nitrogen from the digestates. In one such study, Zhang et al. [87] studied the role of ZnO NPs on the removal of nitrogen from the sludge in the CANON process. Here they have found ZnO NPs enhance the functioning of ANOB and AAOB, and suppress the activity of NOB, leading to improvements in nitrogen removal. After recovery/removal of nitrogen from the LFD, the solid digestate can serve as a rich source of phosphorus fertilizer.

Taking the account of post-recovery amendments in the BGS, we have reviewed the research efforts in the area of biomass conversion and waste valorization to develop conceptual understanding and technical realizations of the plausible strategies. For developing BGS-derived products,

another key challenge is the consistent quality of the slurry. There must be quality parameters for the procurement of BGS from various stakeholders. Rath and Joshi [35] recommended electrical conductivity and dissolved solid contents as two preferable parameters for on-site quality assessment of BGS. However, extensive studies are required to standardize the methods of BGS quality assessment and consequently the cost as per quality.

Referring to Fig. 11, it has been demonstrated through the theoretical calculations that adding NEDOCs can improve nutrient levels (such as N and P) in BGS. The combined formulations of BGS and NEDOCs (optimized ratios) can be utilized as a nutrient-enriched low-cost substrate for the mass multiplication of AIMs. Such investigations can potentially be replaced the requirements of expensive carbon sources such as sucrose to achieve the ideal C/N ratio for microbial growth during solid-state fermentation [39]. Bioprospecting of the beneficial microbes through multi-substrate interactions can also be done to produce bioactive compounds offering anti-phytopathogenic activities. Further research is recommended in the area of developing sustainable feedstock combinations (BGS amended with different NEDOCs) through optimization studies to produce platform chemicals. The expense of resources in phase fractionation (separation of solid and liquid phases) of BGS can also be reduced through the enrichment strategies using DOCs and AIMs.

The role of NPs in biostimulation of microbial activities during AD with opportunities and challenges to improve biogas yields coupled with the recovery of quality digestates is discussed in Section 3.3. Interestingly, we have also found reports on the synthesis of nanoparticles from NEDOCs such as *Madhuca latifolia* [232] and *Jatropha curcas* [233]. Hence, NEDOCs and BGS can be further explored as low-cost reducing and capping agents for the synthesis of nanoparticles. Application of such biofabricated nanomaterials in the improvement of AD of biomass waste, product recovery, and value addition of BGS will substantially encourage greener, eco-friendly, and economically viable routes to transform challenges into opportunities in the real world. Adding to this, several studies are available that report application of NPs (at low/optimized dose) in promoting microbial activity. Hence, it could be an innovative approach for the researchers to explore the possibilities of utilizing BGS enriched formulations (a) to deliver AIMs and nanoparticles to the plants, (b) to stimulate the process of vermicomposting, etc. However, the valorization methodologies inspired by nanotechnology-based approaches have the risk of nanotoxicity on the environment and ecosystem concerned. In this regard, we have reviewed the life-cycle analysis studies to access the environmental impacts of such introductions. In one such study, Hijazi et al. [192] reported that the environmental impact of electricity production from

the nanoparticle (CoNPs, NiNPs, FeNPs, and Fe_3O_4 NPs)-treated biomass was lesser as compared to the untreated biomass (control). Hassanein et al. [198] fertilized *Lactuca sativa* with the diluted effluent recovered from the digester fed with NP mixture (FeNPs 1000 mgL^{-1}, NiNPs 120 mgL^{-1}, and CoNPs 54 mgL^{-1}), and reported higher levels of N, P, K, Zn, Mn, Cu, and Na in the plant biomass which could be due to the increased nutrient uptake and translocation efficiency as a consequence of NP treatment. The recorded values of Fe, Ni, and Co in the plant biomass were below the reported toxic limits.

While reviewing the available reports, it has been observed that one of the reasons behind the toxicity of NPs is attributed to their aqueous solubility (AQS) [147]. To extenuate the effect of nanotoxicity during AD, nanoparticle co-aggregation can be explored, which is reported to decrease the availability of free ions in the aqueous medium, thus capable of reducing toxic effects. We have also compared the influence of nanoparticles and their bulk counterparts on the AD during biogas production. Studies favored NP application, though it has also been realized that certain aspects of NPs such as concentration, size, type, and biomass material to which NPs acted upon and their interaction with other trace elements/NPs (hetero-aggregation) in the system are of key significance, and hence application-based optimization studies are critical to prevent their (NPs) overexploitation and related environmental consequences thereof. In AD systems, the presence of antibiotics (norfloxacin, and sulfamethazine) can escalate the toxicity potential of NPs (such as ZnO NPs) [234]. Hence, apart from nanotoxicity, the risk of other emerging complex pollutants will also be concerned and investigated further. Tang et al. [27] reported a low risk of pollution with the repeated applications of BGS in crop fields. However, detailed studies are warranted for comparative assessment of heavy metals, pathogens, pesticides, and other contaminants present in digested (BGS) and undigested manures.

The valuation of BGS has multifaceted outlooks, ranging from agronomic to economic values. BGS and its derived products (such as solid pellets, liquid fertilizer, organic composts, and other value-added products) are in great demand as they offer multiple benefits to the soil and plants, hence considered one of the preferred bio-agri input for organic farmers. Some of the bioslurry-based products are already available in the market [35]. The economic benefits associated with the BGS application in agriculture fields have been reviewed [3, 13, 36, 130, 224]. Talking about commercial viability, competition with the available conventional fertilizers and some consumers also has negative perception towards digestates [13] limits their market acceptance. However, the technical limitations for such observations are covered in earlier sections. There is a need to develop more robust models for the economic and agronomic assessments of such biomass wastes. Integrated studies based on bio/nanotechnology interventions can promote niche development for generating the value-added products from different waste biomass of agriculture origin, thus promoting rural development and benefiting society at large. Overall, it has been concluded that chemical fertilizers are considered a readily available form of nutrients to the crop plants, though their extensive usage in agriculture fields reportedly has adverse impacts on normal ecological functioning at various levels. The area of organic farming is traditionally in use, much promising, and diverse but emerging challenges like effectivity, productivity, and commercial exploitation precisely with the use of organic manures derived from livestock wastes processing such as BGS need further attention. Here, based on an extensive literature survey and critical assessment of available information to the best of our access, we have proposed integrated approaches for improving the agriculture potential of BGS, which is principally an amalgamation of applied aspects of bio/nanotechnology.

Declarations

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