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Research Article

Excellence in Excrements: Upcycling of Herbivore Manure into Nanocellulose and Biogas

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ABSTRACT: The demand for animal products has significantly increased over the past decades as a result of the growing population and the heightened standards of living. Increased livestock farming does not only yield desired products but also significant quantities of wastes, particularly manure whose storage and application are being monitored with a tightening network of regulations. The problem is that manure is considered merely as a substrate for biogas production or as a fertilizer, whereas the substantial portion of fibers residing in herbivore manure has remained underutilized. Here, we propose a manure management system, in which not only biogas and fertilizer precursors but also high-value materials in the form of (nano)cellulose are produced. We show that high biogas yields can be achieved for elephant manure and the remaining substrate enables effortless isolation of cellulose nanofibers, leading to a significant reduction of the environmental impact compared with traditional systems based on wood.

KEYWORDS: *elephant manure, biogas, nanocellulose, manure management, tensile properties*

INTRODUCTION

Climate change has bestowed unforeseen engineering challenges upon the modern society. The most pressing of those challenges have been connected to meat and dairy production systems. In 2018, 436 Mt CO_2 equiv of greenhouse gases (GHG) were emitted by agricultural operations in the EU-27 alone, thus coming only second after the energy sector,[1] with manure being one of the main emitters of non-CO_2 GHG within agriculture, particularly rife in methane and nitrous oxides.[2] Additionally, this nutrient-rich material, riddled with pathogens, also adds to the eutrophication and contamination of soil and surface water if its release into the environment is uncontrolled.[3,4] What is often overlooked is that herbivore manure encompasses useful fibrous material because of its notoriously inefficient digestion. Depending strongly on the feed and digestion system of the animal, herbivore's manure can still contain 50−80% intact lignocellulosic material.[5,6] While livestock manure is already utilized as a fertilizer and in biogas production, it contains a lot of fibers that remain idle within these applications.[7] Indeed, anaerobic digestion (AD) is able to convert much of the compounds, like proteins and small-molecular carbohydrates, into CH_4, which can be used as a biofuel or source of electricity in a combined heat and power plant. Nevertheless, recalcitrant polymers, such as cellulose, are generally not converted completely and remain in the fermentation residue. In addition, there is simply an overabundance of manure in intensive livestock farming areas that are situated away from crop farming regions. For example, in the Netherlands, 17.9 Mt of the total 68.6 Mt animal manure produced in 2011 had to be exported, resulting in additional environmental burden and costs.[8]

Here, we propose a solution to elevate the use of manure into a completely new level as a source of high-value-added materials. Although much of the fibers pass the digestive system seemingly unchanged, their ultrastructural morphology has been markedly changed, resulting in an altogether more refined structure that better enables the disintegration of the fibers into nanocellulose. This apparently trivial procedure is a major engineering challenge in modern materials science concerned with renewable materials. For the best part of the current century, cellulose nanofibers (NFCs) have been touted as a promising sustainable alternative in, e.g., composite manufacturing,[9,10] membrane technology,[11,12] and medical applications,[13,14] but the high price and/or energy consumption of their preparation has impeded the genuine

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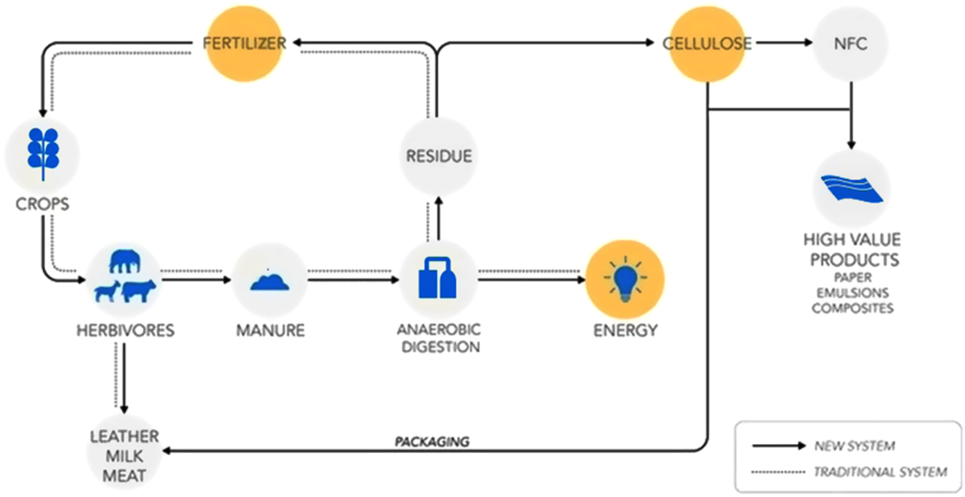

Figure 1. Graphical description of the envisioned production chain of value-added products from animal manure: conventional use of manure (traditional system, dotted line) and new production system (new system, solid line).

industrial triumph of NFCs so far.[15] According to Global Market Insights, by 2026, the nanocellulose business is anticipated to reach $418 million, but to fulfill the potential, the production costs must be decreased by new solutions.[16]

In this study, we are set to solve the very different problems of underutilized manure and energy consumption in NFC production. As a model system, we have used elephant manure. It has one of the highest cellulose contents among herbivore manure, as they only digest 30−40% of their feed, and comprises approximately 40 wt % cellulose, 23 wt % lignin, and 10 wt % hemicellulose. The elephant's digestive tract is representative of all large animals with monogastric digestion system.[6,17] For example, horse manure contains a similar amount of cellulose (20−30%), and therefore, the findings reported here are also applicable to horse manure.[18] It is estimated that 40−70 Mio tons per year is produced in Europe alone.[19] We show how animal manure as well as fermentation residues can be utilized to their full potential by generating products, such as NFCs, biogas, and fertilizer precursors (Figure 1). The quality of NFC was ascertained by characterizing the physicochemical and mechanical properties of cellulose nanopapers prepared thereof. The envisioned production chain not only upcycles manure and alleviates NFC production but also outsources the plant harvesting and processing to herbivores, thus reducing the effort and environmental costs of cutting down trees as well as pulping, bleaching, and defibrillation operations, which are only set to grow in demand in a modern society hungry for more renewable materials solutions. The environmental impact of the proposed production cycle was evaluated by lifecycle assessment and juxtaposed with the traditional paper production from wood and will be reported in Krexner et al.[20]

MATERIALS AND METHOD

Materials. Fresh elephant manure was provided by the Vienna Zoo "Tiergarten Schönbrunn". Different batches of manure (ca. 10 kg each) were collected in April, June (2018), and January (2019). If not mentioned otherwise, the discussed results were generated with material collected in April 2018. Sodium hypochlorite (NaOCl, 14% active Cl, W. Neuber's Enkel, Austria), sodium hydroxide (NaOH, 99.6%, Sigma-Aldrich), and sulfuric acid (95%, Sigma-Aldrich) were used as received. Neutral detergent solution concentrate, acid detergent solution powder, and triethylene glycol were purchased from Ankom Technology (Macedon). Decahydronaphthalene was supplied by Merck (Darmstadt, Germany). Microcrystalline cellulose was purchased from Sigma-Aldrich. As a reference, birch kraft pulp (KP) was used. For all experiments, distilled water was used. The inoculum for biogas experiments consisted of two inocula from a biogas plant located in Margarethen am Moos and Ziersdorf (Austria), respectively.

Specific Methane Yield and Degradation Kinetic Studies of Elephant Manure. Anaerobic digestion trials of elephant manure were performed in triplicate according to standard VDI 4630 (VDI—The Association of German Engineers 2016). Eudiometer batch fermenters (250 mL) were filled with 200 mL of inoculum in a 3:1 ratio based on volatile solid (VS) content and stirred continuously at 37.5 °C (mesophilic conditions) over the course of 40 days. Microcrystalline cellulose was used as a control. The methane and biogas production were monitored on a daily basis. Gas volumes are reported at 273.15 K and 101.33 kPa per kilogram of volatile solids (L_N kg^{-1} VS). The methane (CH_4) and carbon dioxide (CO_2) contents were analyzed with a portable gas analyzer (Dräger X-am 7000, Dräger, Lübeck, Germany), calibrated weekly with a gas standard consisting of 33% CH_4 and 33% CO_2 (Messer, Gumpoldskirchen, Austria).

Investigation of the degradation kinetics of structural compounds of elephant manure was performed following the procedure described previously.[21] Samples were taken after 5, 10, 20, 30, and 40 days of AD, respectively, and washed thoroughly with deionized water. The samples were dried at 50 °C until constant weight.

Chemical Analysis of Manure and Fermentation Residue. Elephant manure and fermentation residues were investigated with regard to their water, cellulose, hemicellulose, and acid detergent lignin (ADL) contents using the method of Naumann and Bassler.[22,23] The material was first ground using a cryomill (CryoMill, Retsch, Haan, Germany) at a frequency of 28 Hz for 14 min after precooling with liquid nitrogen at a frequency of 5 Hz for 2 min. The water content was evaluated using a Karl Fischer titrator (Mettler Toledo V20, Columbus, Ohio). To analyze the influence of anaerobic digestion on the structural fiber components, degradation yield ($\text{yield}_{\text{degradation}}$) was calculated based on the biomass before biogas production and the corresponding fiber contents of cellulose, hemicellulose, and ADL after days 0, 5, 10, 20, 30, and 40 of AD (eq 1).

$$\text{yield}_{\text{degradation}}\ (\%) = \frac{\text{mass biogas residue (g)} \cdot \text{fiber content (\%)}}{\text{mass before biogas production (g)}} \quad (1)$$

Elemental analysis was performed using an EA 3000 CHNS-O elemental analyzer (Eurovector, Italy) equipped with a high-

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temperature pyrolysis furnace (HT, Hekatech, Germany). Fourier transform infrared (FT-IR) spectra were recorded on a spectrometer (Carry 630, Agilent Technology, Austria).

Extraction of Cellulose Fibers from Elephant Manure and Biogas Residue. Dried manure or biogas residue, respectively, were washed over a 300 μm mesh sieve, and the sand was removed by sedimentation. Alkaline treatment (20 g manure/AD residue L^{-1}) was conducted in 0.1 M NaOH for 2 h at 80 °C.

The liquid residue of the alkaline treatment was neutralized and dried. $\text{Yield}_{\text{extract}}$ was calculated based on the received dry mass in the extract and the mass before biogas production in dry mass (eq 2)

$$\text{yield}_{\text{extract}}\,(\%) = \frac{\text{received material of alkaline extraction (g)}}{\text{mass before biogas production (g)}} \times 100\% \quad (2)$$

The residue was rinsed with water over a 75 μm mesh sieve. Bleaching was performed using 4 g of dry matter (DM) of alkaline-pretreated manure stirred overnight (17 h) in 660 mL of a 0.4 M NaOCl solution. The suspension was washed over a 75 μm mesh sieve, dewatered, and stored at 4 °C.

The total yield ($\text{yield}_{\text{total}}$) was calculated based on the mass of the lignocellulosic material received after the extraction process over the mass before biogas production in dry mass using eq 3.

$$\text{yield}_{\text{total}}\,(\%) = \frac{\text{material after extraction (g)}}{\text{mass before biogas production (g)}} \times 100\% \quad (3)$$

Nanocellulose Production. To produce nanocellulose, the extracted cellulose fibers (50 g DM as 3.5% suspension in water) were blended for 4 min at 1000 rpm (JB 3060, Braun) and subsequently passed through a disk mill (Granomat JP 150, Fuchs, Switzerland). Extracted fibers from elephant manure without prior biogas production were used to investigate the influence of grinding cycles on the material. The material was ground 1, 2, 5, 7, and 10 times, respectively. This procedure was done for two different batches. Extracted fibers from biogas residue were ground 10 times. The resulting gels were dewatered over a textile cloth to a consistency of approximately 2 wt % and stored at 4 °C. As a reference, nanocellulose was produced from kraft birch pulp by grinding 10 times. The energy consumption was monitored using an energy logger (Voltcraft 4000) and a high-voltage meter (Swissnox SX-3M).

(NFC)Paper Production. For papers with a grammage of 100 g m^{-2}, 1.22 g (DM) extracted fibers were blended for 3 min at 1000 rpm with 300 mL of water (JB 3060, Braun) and filtered over a Büchner filter funnel (VitraPOR, 100 mL, Por. 3, Robu, Hattert, Germany) lined with a filter paper (VWR 413, 125 mm in diameter). The filter cake was pressed between two sheets of blotting and one sheet of baking paper in a heated hydraulic press (model 412 6CE, Carver) two times at 20 °C at a 20 kN force and subsequently at 120 °C for 15 min applying a 20 kN force.

Tensile Properties of (NFC)Papers. For the determination of the tensile properties of the produced papers dog bone-shaped specimens (shape after type 1BA, EN ISO 527-2) with 5 mm parallel width and 75 mm overall length (Zwick ZCP 020 Manual Cutting Press, Zwick, Ulm, Germany) were cut. Tensile tests were performed using a universal test frame (Model 5969, Dual Column Universal Testing System, Instron, Darmstadt, Germany) equipped with a 1 kN load cell and a noncontact video extensometer (Gig ProE, iMETRIUM, Bristol, U.K.) at a temperature of 26 °C and a relative humidity of 50% with a testing velocity of 1 mm min^{-1}. The gauge length was set to 25 mm. The tensile strength (σ) was calculated from the maximum load and the cross-sectional area of the specimen. Young's modulus (E) was analyzed in the linear elastic region of the stress−strain curve as secant between strength values separated by 0.2% strain (ε).

Morphology of (NFC)Papers. Scanning electron microscopy (SEM) images of the (NFC)papers were obtained with a Zeiss Supra 55VP at an accelerating voltage of 2 kV and a working distance of 7.2 mm. The samples were coated with a 10 nm gold layer (Leica scd 050/EM QSG 100) at 60 mA for 67 s. The fiber diameters were determined with the operating software for field emission scanning electron microscope SmartSEM V05.04.

RESULTS AND DISCUSSION

Elephant Manure as Feedstock for Biogas Production. The collected and dried elephant manure had cellulose, hemicellulose, and ADL degradation yields of 31, 28, and 15 wt % of dry mass, respectively (see Supporting Information Table S1). The high share of lignocellulosic material of 74% highlights the inefficient digestive system of the elephants and signifies the suitability of manure as a raw material for the proposed system. With an ash content of 5 wt % representing the inorganic content, the organic residue can be estimated to be 8 wt %. Thus, it had a similar cellulose content to wheat straw (35−39 wt %), approaching that of wood (45−50 wt %) and comparable to cow manure (30%).[24,25] The difference being that elephants are monogastric herbivores, while cattle are ruminants. With straw and hay being the main feed of elephants kept in captivity, it signifies that the cellulose yield was not compromised by the elephant's digestion system but had the advantage of collection as well as mechanical and enzymatical pretreatment by the animal.[26] Manures collected at different times (April (0-1), June (0-2), and January (0-3)) varied in their composition (see Supporting Information Table S1). The raw material 0-2 collected in June exhibited the highest cellulose degradation yield (57%), and the material 0-3 collected in January had the highest ADL degradation yield (29%). This variation occurs because of differences in seasonal food supply for the elephants.

The volatile solid content (VS) corresponds to the organic dry matter in the material without inorganic matter and gives an orientation of how much organic material can be converted into biogas. With 89%, a high content of organic biomass is convertible into methane, indicating that elephant manure is a promising feedstock for the anaerobic digestion process. Elephant manure had a VS comparable to wheat straw (83−88%)[27,28] and other agricultural biomass (maize silage 96%,[29] grass silage 86%[29]). The C/N ratio (46:1; see Supporting Information Table S2) on the other hand is a crucial parameter for the process stability during AD. A low C/N ratio leads to ammonia intoxication of the fermentation system and inhibits the AD process, whereby a too high ratio microbial biomass cannot be maintained. The optimal C/N ratio for biogas production is widely discussed in the literature and is found to be optimal at approximately 30:1, but no ammonia intoxication is expected at a C/N ratio at 46:1.[30]

During the first 10 days of anaerobic digestion, the biogas and methane yields were 66% of the total biogas (262 L_N kg^{-1} VS) and 64% (156 L_N kg^{-1} VS) of total methane. After 40 days of fermentation, the final specific biogas yield was 399 L_N kg^{-1} VS with a methane content of 61% (244 L_N kg^{-1} VS) (Figure 2). A previous study reported a similar amount of methane produced from zoo animal manure in 12 days.[31] Using hay as biogas substrate resulted in almost the same methane yield (243 L_N kg^{-1} VS) but a lower methane concentration (58%) at an overall biogas yield slightly higher than in our study (420 L_N kg^{-1} VS).[32] Most of the biogas and methane is produced during the first 10 days of AD by preferential conversion of hemicellulose and cellulose (Figure 2);[33] their yield decreased by 55 and 54%, respectively. Between day 10 and day 40 in the fermenter, the yield of both structural polymers decreased further by approximately 30%. The ADL yield remained almost

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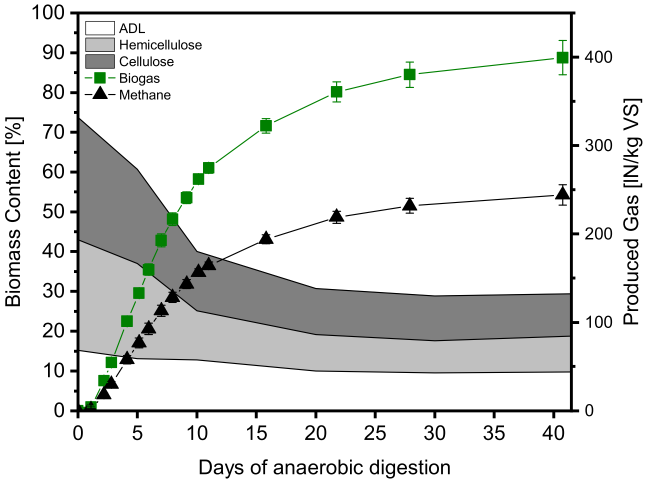

Figure 2. Biogas (green square) and methane (black triangle) yields (L_N kg^{-1} VS) as a function of fermentation time using elephant manure as feedstock. ADL (white), hemicellulose (gray), and cellulose (dark gray) contents in manure (day 0) and the digestate after 5−40 days of anaerobic digestion, respectively.

constant, showing that lignin was converted into biogas to a much smaller extent and hence up-concentrated in the material.[34]

An alkaline treatment was performed using 0.1 M NaOH to separate the fibrous material from nitrogen-containing compounds, such as proteins, keratinized tissue, and dead cells, which can potentially be used as a fertilizer and originated from the elephant's digestion process. The liquid residue after alkaline treatment was neutralized and dried to be considered as a fertilizer precursor. $\text{Yield}_{\text{extract}}$ ranged here from 25% for material without any prior anaerobic digestion and decreased to 11% for AD of 40 days. All liquid extracts after AD contained between 20 and 28% carbon and <1% nitrogen, which is in accordance with the fertilizer regulation (BGBl. II Nr. 100/2004) (see Supporting Information Table S4).[35] Optimization regarding nitrogen, phosphorus, and mineral content to meet the soils need would be possible through additional purification and up-concentration processes.

To come up with white cellulose pulp, a bleaching treatment was conducted. $\text{Yield}_{\text{total}}$ of lignocellulosic fibers after alkaline and bleaching treatment of elephant manure without prior anaerobic digestion was 41% (Table 1) and represents only the yield after the chemical treatment. AD prior to chemical extraction resulted in a yield reduction (21% for 5 days AD to 12% for 20 and 30 days of AD). Wood as raw material using the established Kraft process has yields of 46−50%, which seems significantly higher.[36,37] But it needs to be pointed out that already biogas in high yields is produced from the fermentation residue and that this is a low-grade biomass compared with wood. Cellulose could be extracted from cow manure with a yield of 11% without prior AD, which is similar to elephant manure after 20−40 days of anaerobic digestion.[38]

Mechanical Properties of Papers Prepared from Biogas Residues. The tensile properties of papers produced from extracted fibers were evaluated to assess the impact of biogas production and hence the altered composition of the raw material (Table 1, representative stress−strain curves in Supporting Information Figure S1) and degradation of the fibrous material. Papers produced from lignocellulosic fibers extracted from biogas residue (5 days AD) had even higher tensile strength (74 MPa), Young's modulus (11 GPa), and strain to failure (1.4%) than the reference paper produced from elephant manure extract without prior anaerobic digestion (60 MPa). The tensile strength of papers produced from lignocellulosic fibers extracted from unfermented manure or fermentation residue after 5 and 10 days outperformed ordinary copy paper (20−50 MPa) with the advantage of utilizing more sustainable raw material than wood and that a further value-added product (biogas) was also obtained in this process.[39,40]

For papers obtained from fibers that were retrieved after longer fermentation times (20−40 days), a significant decrease in tensile strength (~30 MPa), modulus (~6 GPa), and strain to failure (0.6−0.7%) was observed. These lower tensile properties of the produced papers can be attributed to the varying composition of the material during AD: With increasing fermentation time, lignin is up-concentrated, while the hemicellulose content decreases, but the molecular weight of the extracted lignocellulosic fibers did not follow a specific trend and is therefore more influenced on the variety of the biological material than influenced by the anaerobic digestion process (see Supporting Information Table S3). Lignin, due to its chemical structure, including hydrophobic aromatic rings, weakens particularly the hydrogen-bonding network between the cellulose fibrils and thus the paper strength decreases.[41] After extended AD (20, 30, and 40 days), between 8 and 10% lignin remains in the extracted fibers (Table 1). Only a minor increase in nitrogen content from <0.05 to 0.08 wt % indicates remaining compounds after the chemical extraction (see Supporting Information Table S3).

SEM images of the produced papers showed separated cellulose fibers with diameters of 13 μm with a high density of connection points between the fibers. The average fiber diameter was not affected by increasing the fermentation time. However, it appears that the fibers were embedded in a matrix

Table 1. Tensile Strength (σ, MPa), Elastic Modulus (E, GPa), Strain to Failure (ε, %), Fiber Diameter (μm), and ADL Content (wt %) of the Produced Paper of Cellulose Extracted from Fermentation Residue of Elephant Manure after 0−40 Days of Anaerobic Digestion[a]

days of AD	σ [MPa]	E [GPa]	ε [%]	$\text{yield}_{\text{total}}$ [%]	fiber diameter [μm]	ADL [wt %]
0	60.4 ± 4.9	9.1 ± 0.7	1.22 ± 0.12	41	11 ± 5	2.3 ± 0.4
5	74.0 ± 2.3	11.1 ± 1.2	1.37 ± 0.11	21	13 ± 8	4.3 ± 0.03
10	66.2 ± 6.8	9.7 ± 0.6	0.92 ± 0.08	17	13 ± 8	5.2 ± 0.5
20	32.2 ± 2.3	6.2 ± 0.8	0.68 ± 0.04	12	15 ± 10	8.3 ± 0.3
30	31.2 ± 8.4	5.7 ± 0.8	0.70 ± 0.13	12	13 ± 8	8.9 ± 0.2
40	27.4 ± 3.4	5.7 ± 0.3	0.58 ± 0.07	12	13 ± 7	10.4 ± 1.4

[a] $\text{Yield}_{\text{total}}$ (%) of received lignocellulosic fibers after biogas production and extraction.

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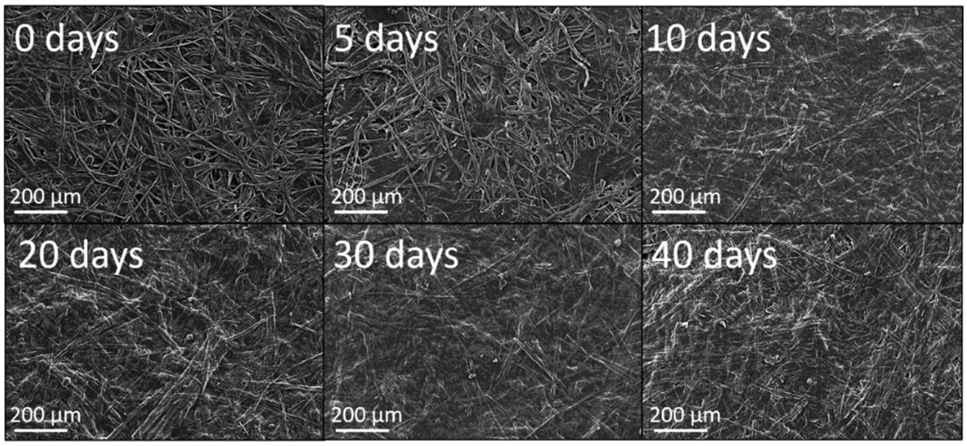

Figure 3. SEM micrographs (100× magnification) detailing the surface morphology of the produced papers using lignocellulosic microfibers extracted from elephant manure (0 days) and fermentation residues.

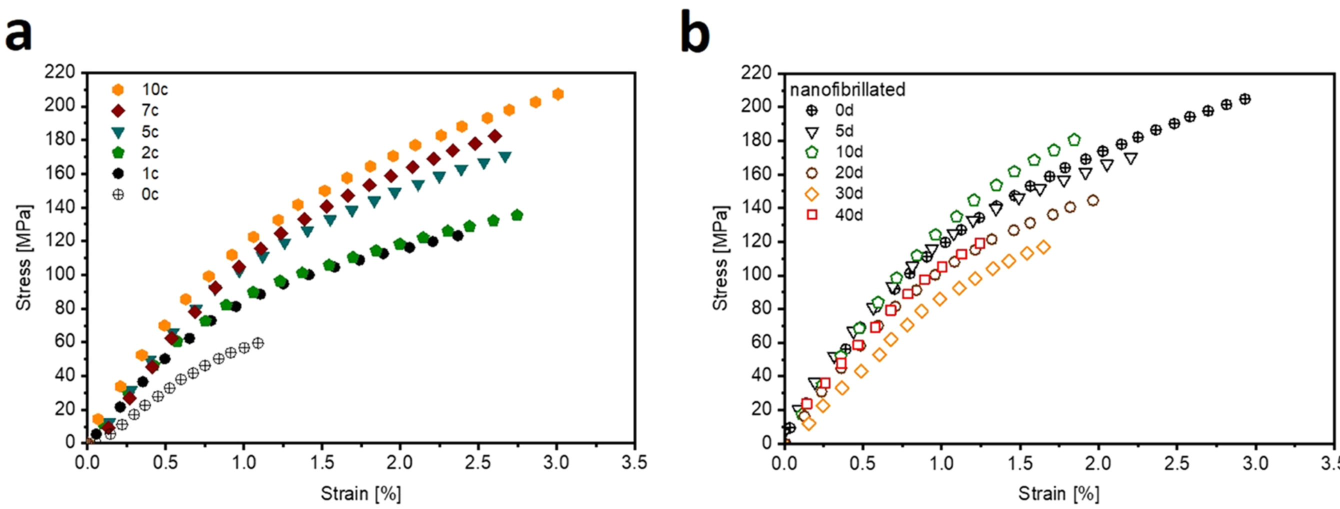

Figure 4. (a) Representative stress−strain curves of nanopapers manufactured using NFC produced by grinding extracted lignocellulose fibers from elephant manure without prior anaerobic digestion after 1 (1c, black dot), 2 (2c, green triangle), 5 (5c, blue triangle), 7 (7c, red diamond), and 10 (10c, orange hexagon) grinding cycles, as well as reference paper produced from elephant manure cellulose fibers (0c, crossed dot). (b) Representative stress−strain curves of nanopapers produced by grinding lignocellulosic fibers extracted from elephant manure 10 times (crossed circle) and from fermentation residue after 5 (black triangle), 10 (green pentagon), 20 (brown circle), 30 (orange diamond), and 40 (red square) days of anaerobic digestion (nanofibrillated).

of lignin, causing a reduction in connectivity between the fibers (Figure 3).

Nanofibrillation for Improved Paper Tensile Properties. Already after one grinding cycle (0.3 kWh kg^{-1}), the tensile strength of the papers increased to 126 MPa thus doubling compared to microfiber papers (64 MPa) (Figure 4a and Table 2). The modulus increased by 173% (from 7.1 to 12.3 GPa), and the strain to failure increased by approximately 50% (2.1%). Fiber agglomerates with diameters of 7−0.5 μm were present in the material, but the majority of fibers was already in the nanometer range (see Supporting Information Table S5). After 10 grinding cycles (5.2 kWh kg^{-1} total energy input), the tensile strength improved further, reaching ∼200 MPa. The increased tensile strength with increasing energy input can be attributed to the higher homogeneity of the material and decreased fiber diameter, leading to increased bonding between the fibers.[42] Reference paper fabricated from Kraft pulp had significantly lower tensile strengths of 16, 30, and 57 MPa after one, two, and five grinding cycles, respectively (see Supporting Information Table S5), which might be associated with larger fiber diameters that were still in the micrometer range after the two first grinding cycles (19 and 15 μm) (see Supporting Information Table S5). Nanometer-scale fibers were only obtained after the fifth grinding cycle, but they were still 50% bigger than fibrils produced by grinding of elephant manure fibers.

These NFC papers produced highlight the material's potential of elephant manure even without the previous production of methane from the material. The tensile properties are on par or even outperformed nanocellulose papers produced from high-grade biomass, such as wood as well as NFC papers from agricultural waste reported in the literature.[43−46]

The impact of variations in feed and thus seasonal influences, in particular the significantly higher ADL contents of 22 and 34 wt %, were confirmed by producing nanopapers from elephant manure collected in June and January exhibiting tensile strengths of 130 and 96 MPa, respectively (Table 2, Supporting Information Table S5).[17] After 10 grinding cycles, nanofibrils derived from the material collected in June and

Table 2. Tensile Properties: Tensile Strength (σ, MPa), Elastic Modulus (E, GPa), and Strain to Failure (ε, %) of NFC Papers as well as Average Fiber Diameter (nm) of Lignocellulosic Fibers Extracted from Elephant Manure with and without Prior AD[a]

days of AD	cycles	σ [MPa]	E [GPa]	ε [%]	fiber diameter [nm]
0	1	123 ± 6	12.3 ± 0.8	2.1 ± 0.3	36 ± 17
0	2	142 ± 11	12.6 ± 0.9	3.2 ± 0.4	32 ± 24
0	5	171 ± 23	11.7 ± 0.7	2.9 ± 0.5	27 ± 6
0	7	181 ± 16	11.9 ± 1.0	3.2 ± 0.6	33 ± 14
0	10	196 ± 12	11.6 ± 1.2	3.6 ± 0.6	22 ± 4
5	10	173 ± 7	14.5 ± 0.6	2.7 ± 0.6	22 ± 7
10	10	185 ± 7	14.2 ± 1.0	2.2 ± 0.4	32 ± 8
20	10	144 ± 7	13.5 ± 0.3	1.9 ± 0.2	27 ± 6
30	10	115 ± 5	13.3 ± 1.3	1.7 ± 0.2	27 ± 6
40	10	125 ± 9	12.9 ± 0.6	1.6 ± 0.3	31 ± 6
KP	10	102 ± 6	9.7 ± 3.4	3.7 ± 0.5	43 ± 13

[a]Values for the reference material from Kraft pulp (KP) are provided as well.

January had fiber diameters of 40 and 44 nm, respectively, which is ~50% higher than those produced from raw material collected in April.

Papers produced from nanofibrillated lignocellulosic fibers extracted from fermentation residue after 10 days of AD had a tensile strength of 185 MPa and a modulus of 14.2 GPa. The tensile strength was slightly lower, but the modulus was higher than that for nanopapers produced from fibers extracted from unfermented manure, with the major benefit that additionally biogas was produced. The reduction of the paper tensile strength from nonground lignocellulosic fibers extracted after 0−40 days of AD was 57%, whereas for the NFC papers, it was only 47%. The reduction of tensile strength of the papers produced using nonfibrillated fibers was caused by the up-concentration of lignin on the fibers (Figure 4b), which was partially mitigated by the nanofibrillation process. Papers produced from ground lignocellulosic material obtained after 40 days of fermentation had a tensile strength of 125 MPa, a modulus of 13 GPa, and a strain to failure (1.6%), which was still higher than the tensile properties of nanopapers from Kraft pulp. Nanocellulose papers from Norway spruce with 4 and 14% residual lignin had similar tensile strengths of 156 and 125 MPa, respectively.[47] Nanocellulose papers produced from *Miscanthus* straw, which had been anaerobically digested for 4 months, had a tensile strength of 155 MPa.[48] This is comparable to NFC papers from elephant manure after 20 days of anaerobic digestion, although the cellulose from *Miscanthus* straw was extracted with a harsher chemical procedure (extraction with organic solvents such as hexane and dichloromethane followed by multiple bleaching steps using $NaClO_2$).

CONCLUSIONS

Elephant manure was used as a model system to demonstrate the usefulness of livestock wastes as a raw material for the simultaneous production of biogas and (nanofibrillated) lignocellulosic fibers to yield high-performance (nano)papers and fertilizer precursors. After 10 days of AD, a biogas yield of 262 L_N kg^{-1} VS with 64% methane content was obtained, while (nano)papers produced from extracted lignocellulosic material had tensile properties (185 MPa at 100 g m^{-2}) similar to or exceeding those of papers produced from conventional raw materials, such as wood pulp. This shows that a multiple-output system for the utilization of animal manure as a resource can improve the sustainability of livestock waste management systems by reducing waste and energy consumption while simultaneously yielding high-quality products.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acssuschemeng.1c05175.

Additional supporting results, such as elemental composition and degradation yield of fermentation residue, elemental composition of extracted fibrous material from fermentation residue and elephant manure, elemental composition of neutralized dried residue after alkaline treatment, and mechanical properties of the produced paper of cellulose extracted from elephant manure and fermentation residue (PDF)

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Notes

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