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“Methano-compost”, a booster and restoring agent for thermophilic anaerobic digestion of energy crops

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ABSTRACT

The influence of plant litter-compost of the hot rotten-phase as additional inoculum for anaerobic batch digestion of sugar beet silage (SBS) was studied. Four simultaneously driven batch-fermenters were inoculated with sewage sludge. Two of the fermenters were inoculated additionally with the same amount of organics by compost of the hot rotten-phase. Two of the fermenters were mesophilic (40 °C) and the other two were thermophilic (60 °C). The impact on the gas production rate and gas yield was observed to be boosted for thermophilic (60 °C) and only a minor effect of 6–13% for mesophilic (40 °C) digestion. The gas yield increased considerably up to 26.5% at 60 °C (batch). Also the methane content increased from 57.4% to 62.3% by adding compost (continuously run mesophilic digestion). Fluorescence In Situ Hybridization (FISH) indicated that a microbial effect was responsible for the observed stimulation of gas production rates, but not simply by increasing the bacterial counts. By analysing each fermenter for its mineral and trace element content a mineralic effect could be excluded. However, the bacterial counts by FISH of 10 different groups were somewhat ambiguous. But an effect on the presence of Chloroflexi could be demonstrated. They nearly doubled to 15–16% by supplementation with compost. Furthermore, under thermophilic conditions, the added compost induced a significant shift in the microbial composition towards hydrogenotrophic Methanobacteriales. The suggestive conclusion drawn is that this explicitly increase in hydrogenotrophic activity could alone or in combination with accompanying fermentative bacteria forces the microbial food chain towards stimulation of methane generation.

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1. Introduction

A remarkable publication in the last 25 years referring to anaerobic digestion was that Methanoarchaeota were also discovered in aerobic environments [1]. The Dutch colleagues found in aerated mushroom compost of the hot rotten-phase by classical methods high numbers of methanogen count of

2e8 g⁻¹ fresh compost weight, mainly *Methanobacterium thermoautotrophicum*. Such high numbers of methanogens seemed to be quite astonishing for this habitat. Furthermore, there was a report of two unknown practically orientated German scientists who published that they could repeatedly restore an acidified anaerobic digester with municipal sewage sludge simply by the addition of compost made from plant litter.

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Surprisingly, the acidified mesophilic sewage sludge became alkaline after addition of plant litter compost [2]. Later, Thummes investigated in her Ph. D. thesis the microbial composition of compost made from bio-waste of the separate civic collection and found also a similar number of *Methanothermobacter* than Derikx et al. [1], but additional *Methanosarcina thermophila* in compost [3,4]. Based on the microbial investigations with mushroom compost by Derikx et al. [1], a hot rotten compost was applied generally as inoculum in previous studies in the HAW Hamburg to obtain high digestion rates [5,6], but its necessity was not proved beside the simultaneous use of swine manure and sewage sludge. The addition of a suspension of hot rotten compost alone had a poor effect on the start-up phase. Therefore, generally, sludge of a mesophilic sewage sludge digester was used in addition to compost as an inoculum.

“Hot rotten” compost was successfully and routinely used for the thermophilic digestion of the organic fraction of municipal “grey waste” [5] as well as the mesophilic and thermophilic anaerobic digestion of beet silage [6]. As shown by Scherer et al. [6], the thermophilic fermenter (65 °C) started by this inoculation procedure instantly and reached the full organic loading rate (OLR) in just four days. Unfortunately, the 65 °C beet fermenter acidified after 85 days as the propionate level increased slowly, but after decreasing the temperature to 60 °C the automatically fed fermenter is still running for more than 10 years since 2001 with beet silage as substrate without disturbance. If there was a disturbance due to acidification of an anaerobic digester at the HAW Hamburg, in general, the fermenter was successfully restored with the help of a suspension made from the hot rotten phase of compost as described under Materials and methods.

The recovering of acidified, large scale biogas plants is an unsolved problem as the disturbance is often accompanied by significantly high ammonium concentrations. Therefore, the commonly applied second inoculation with manure or sewage sludge (sewage sludge is not allowed in agricultural biogas plants in Germany) is critical. The alternative supplementation by lime to neutralize the acidified reactor could have a toxic side effect. The supplementation with “methano-compost” seems to be easier and cheaper to use than specially pre-cultivated microorganisms [7,8]. It was called “methano-compost” as it contained besides the aerobic microflora, a methanogenic population. The bioaugmentation by nearly ammonium-free compost of anaerobic digesters was first presented on the 11th World Congress for anaerobic digestion in Brisbane, 2007 [9]. It was the starting point to proof the stimulating effect in lab-scale fermenters under exactly driven automated conditions [10]. Aim of the present study was to further enlighten in more detail the role of the participating microbial groups for the observed remarkable effects.

2. Material and methods

The effect of hot rotten compost on anaerobic digestion was investigated in 2-L batch fermenters at 40 °C as well at 60 °C in water baths (Fig. 1). Each fermenter was started with the same substrate from the same harvest. An amount of 160 g

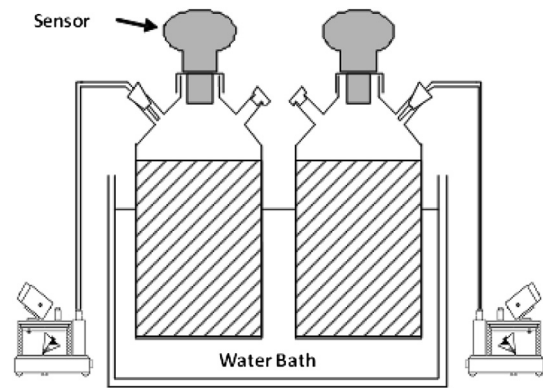


Fig. 1 – Setup of the anaerobic batch mini-fermenters with different inoculi and the operational temperature 40 °C and 60 °C, respectively. The net gas production under standard temperature and pressure (STP) was calculated according to the instruction manual of the used milligascounters (Ritter GmbH, Bochum, Germany, www.ritter.de). The gas production was recorded online by a self constructed data logger as outlined in [11]. On top of the mini-fermenters online IR-sensors were placed to record the methane concentration (Bluesens GmbH, Herne, Germany).

of 1 + 3H₂O diluted mashed sugar beet was used as sole substrate [solid + dissolved VS = 8.792 g/assay, carbon mass fraction 42.1%, 0.006–0.012% ammonium, 0.5–0.6% as ash (TS-VSS), pH 3.8]. Each fermenter vessel was fed with slightly pre-fermented fresh sugar beet silage containing 2.67% (generally 4–8%) dissolved organic acids and alcohols (nearly exclusively acetic acid, < 0.01% of lactic acid and 0.06% ethanol). For further details see Ref. [28]. After 27 d, the same substrate supply was repeated by using a syringe through the rubber stoppers fitted on the fermenters. This interrupted and diminished the value of the gas yield. To compensate for the low buffer capacity of the substrate, 0.04 mol L⁻¹ NaHCO₃ final concentration was added. The combination of 1.0% volatile solids (VS) compost and 1.0% VS of sewage sludge was compared with 2.0% VS sewage sludge alone as inoculum. The experimental setup is shown in Fig. 1.

The sewage sludge was obtained from the municipal waste treatment plant in the city Geesthacht (30 km away from Hamburg), Germany. The 35 °C thickened sewage digester had a retention time of 20–30 days and a TS content of about 5% with 68% VS. The compost (54% VS of the TS content) derived from the organic waste (grass, leaves, flowers and branches) from landscaping of the Hamburg-Bergedorf cemetery precisely at August-Bebel Str. 200, had a temperature of about 50–70 °C (inside) and was between 3 and 4 weeks old. The heaps were usually about 5 m high and only changed every two weeks by a payloader. After sieving the compost with a 1 mm sieve, a suspension of about 1.0% was made with nearly oxygen-free water (boiled water and cooled down). The VS content of the inoculum was adjusted with demineralised anaerobic water to 1.0%, as mentioned above.

The net gas production under standard temperature (0 °C) and pressure (101.3 kPa) was calculated from the measured

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