

ORIGINAL ARTICLE

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# Foam formation in full-scale biogas plants processing biogenic waste

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## Abstract

**Background:** The proportion of biogas in the mix of renewable energies is still remarkably high. The process of anaerobic digestion (AD) provides the basis of biogas production but often leads to excessive foaming. Identifying the reasons for foaming is difficult for biogas plant operators because many factors may play a role. It is therefore difficult for laboratory research to give answers to this specific problem, as the consistency of the digestate itself plays a crucial part in the foam formation process. Hence, careful investigation of foaming in full-scale biogas plants is important in order to identify the main causes and to develop strategies for the prevention of foaming.

**Methods:** Fifteen operators of biogas plants treating biogenic waste have been reviewed in order to estimate the frequency of foaming events in full-scale biogas plants. Samples from foaming digestates were subsequently analyzed. Seven foaming periods in five biogas plants were investigated closely in order to ascertain the causes of foaming events.

**Results:** It was noted that 80% of surveyed biogas plants have had excessive foam formation during the AD process. The foam of two wastewater-treating biogas plants contained filamentous microorganisms. An abrupt temperature increase and the use of grain products and yeasts were identified to be the reason for foaming in four cases. It was, however, not possible to identify the real causes of the foaming event in two cases.

**Conclusions:** Foam formation is a common phenomenon in waste-processing biogas plants. It is important to identify the reasons for foaming because this knowledge helps biogas plant operators to prevent foam formation in the future.

**Keywords:** Full-scale biogas plants; Biogenic waste; Foam formation; Process disturbances

## Background

Biogas plays now an important role in the mix of renewable energies in Germany. This is reflected in the high number of biogas plants. For 2013, the German Biogas Association estimated that there were a total of 7,850 biogas plants and 3,543 MW of total installed electric output [1]. According to the Renewable Energy Statistic Working Group of the German Federal Ministry for Economic Affairs and Energy, biogas accounted for 18.3% of the total electricity generated from all renewable energy sources in 2013 [2]. In order to operate the biogas plants effectively, the identification of problems during all stages of biogas production is becoming an important issue.

The anaerobic digestion (AD) of organic matter is the core of the overall biogas production process, the main problems of which are over-acidification [3,4], forming of floating layers [5], and excessive foam formation. While the causes of over-acidification and floating layer formation are relatively well understood, the causes of foam formation have remained unidentified to a large extent in practice. There are very few references in the literature concerning this issue. The majority of these publications deal with foam formation in full-scale digesters in wastewater treatment e.g. [6-11]. Nevertheless, in the course of AD of sewage sludge, the causes of the foaming events are mostly of another nature than in the course of AD of either biogenic wastes or renewables. Recently, two research papers have been published dealing with foam formation in manure digesters due to organic overloading [12,13]. To our knowledge, there exist only

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two papers dedicated to foam formation in a full-scale biogas plant treating biogenic waste until now [5,14]. Lienen et al. [5] analyzed the formation of floating layer and foam in a full-scale biogas plant treating biogenic waste. The authors demonstrated that both phenomena can be reduced by proper stirring and well-controlled substrate feeding. Kougias et al. [14] described a survey of manure-based full-scale biogas plants in Denmark concerning foaming. The authors reported that foam formation is a widespread problem in Denmark affecting 15 of the 16 biogas plants examined. No similar survey has been carried out for German biogas plants so far.

Excessive foaming can cause substantial costs for biogas plant operators due to the need to add anti-foaming agents, for repairing the broken parts of the system, for the cleaning, and for the additional working hours of the staff caused by an enhanced manpower requirement [8–10]. The follow-up costs of a foam event differ from case to case. Westlund et al. [9] described a ten-week foam event in spring 1996 in the digestion tower of a Swedish waste water treatment plant that produced 2,000 m<sup>3</sup> biogas per day. The foam problem has led to a reduction of gas production by 40%. Due to the reduction of electricity production, the additional personnel costs, and the increased oil consumption and polymer use in the dewatering phase, a total damage of \$150,000 was estimated. Kougias et al. [14] reported a loss of about 20% to 50% biogas production during foaming periods in full-scale biogas plants in Denmark. In extreme cases, the foaming has led to a total process failure [14].

Foaming is not only influenced by the physicochemical properties of the substrates and/or of the fermentation material, but also by microbial effects, the process management (such as the feeding and stirring cycle), and the geometry of the digester itself [6,15]. The mostly described reason for foaming is the organic overload of the digester [6,13,14]. Furthermore, the presence of specific substances such as protein [14], fat, oil, and grease [11] as well as suspended particles [6] was found to contribute to foam formation during the AD. Also, the AD of some substrates such as sugar beet pulp [16,17], acidic whey in connection with chicken manure [14], and slaughterhouse waste [18] is accompanied by enhanced foam formation. Kougias et al. [14] reported that one-fifth of the interviewed biogas plant operators could not identify the real causes of foaming in their digester. The inability to find the foaming causes has ‘the consequence’ that the measures against foaming in biogas reactors are mostly of empirical nature. The common methods are the decrease of the organic load of the digester, the addition of anti-foaming agents, and the optimization of the stirring period [14,18]. Unfortunately, there existed no early warning indicator for the prediction of foam formation until now [11]. The only method for estimating the foaming

propensity of substrates is the foaming test described in Moeller et al. [19]. The laboratory research of foaming causes and mitigation strategies has many limits as the down-scaling of biological processes may lead to inaccuracy and several effects, e.g. an influence of the agitation devices, which is not transferable to the full-scale plants.

While Germany is the worldwide leader in biogas production, the examination of foaming in full-scale fermenters has been rare so far. Thus, the aim of this report is to provide an insight into the research on the causes of foaming based on full-scale biogas plants that utilize biogenic waste. The investigation was carried out on two levels. Firstly, biogas plant operators in Saxony, Saxony-Anhalt, and Thuringia have been reviewed. The aim of this exercise was to survey the experience of a significant number of plant operators with excessive foaming in their digestates. In addition, foaming biogas plants were visited and sampled in order to examine the foaming causes as well as the possible impact of foaming on the anaerobic digestion process. The present work provides new aspects for the research of foaming in the course of anaerobic digestion in biogas production by showing the abundance of effects that may play a role in the formation and stabilization of foam. This is the first publication that presents a systemic study of foaming causes and mitigation strategies in German full-scale biogas plants.

## Methods

### Survey of biogas plant operators

As clear information with regard to the issues of foaming in full-scale biogas reactors has been rare in the literature, a survey of operators of biogenic waste-utilizing biogas plants was carried out in order to monitor the circumstances of foaming in practice. The interviews are based on a qualitative research design in which the subjective actor statements were placed in the foreground. The interviews were carried out according to a guideline. The range of topics encompassing the main questions covered questions about the frequency of foaming, the supposed causes, and consequences of foam formation as well as measures that were usually applied against foaming. The biogas plant operators were defined to be interlocutors, as they are responsible for the management of the particular biogas plant and are aware of the problems, decisions, and economic consequences of the process set-ups in the digesters.

Operators of all waste-utilizing biogas plants based on liquid fermentation that are operated in Saxony (6), Saxony-Anhalt (6), and Thuringia (6) were contacted. Fifteen biogas plant operators were willing to share their experiences.

Prior to the interviews, e-mails introducing the survey were sent to the operators. Ten interviews were carried

out by phone; five biogas plant operators were visited. The names of the plants were coded for data protection reasons in order to ensure a free information flow. During the survey analysis, only the technical content of the interviews was evaluated; the assessment of the nonverbal communication was omitted.

#### Analyses of foaming causes in full-scale fermenters

Five foaming full-scale biogas plants were sampled in order to examine the causes of foaming more closely. The operational data of the sampled biogas plants are summarized in Table 1. Approximately 1 L of digestate was withdrawn via a tap at the side of the fermenter. In the case of BP B, the foam was sampled using an inspection shaft, whereas in the case of BP C, both the outlet of the recirculation pump and the drain of every digester were employed. BP D over-foamed, and foam was collected by its escape from the fermenter. The samples were withdrawn by the biogas plant operators, cooled immediately, and sent in boxes equipped with thermal packs to the laboratory by courier services within 1 day. The analyses were carried out immediately after their arrival.

The samples were pre-treated in order to guarantee their sufficient homogeneity for the analyses. Because the analyses were refined on an ongoing basis, the type of pre-treatment used is indexed in the tables. The original sample without pre-treatment is marked as 'O'.

The sample was either passed through a sieve with a mesh size of 0.75 mm (marked as 'S') or homogenized using a commercially available blender (marked as 'B'). The sieved sample was centrifuged (20 min, at 5,300 rpm and 20°C, Avanti 30 Centrifuge, Beckman, Brea, USA; marked as 'CE') and filtered afterwards (pressure filtration device SM 16 249, Sartorius, Göttingen, Germany; nylon membrane filter: pore size 0.45 µm, Whatman, Germany; marked as 'F'). For the analysis of fatty acids, the sample was extracted according to Bligh and Dyer [20] and Morrison and Smith [21] (marked as 'E'). The calculated values are labeled with 'CA'.

The methods used for the analyses of digestates and their foams are summarized in Table 2. VFA/TIC, pH, and concentrations of NH<sub>4</sub>-N and of volatile fatty acids (VFAs) were determined as quantities which are able to confirm the stability of the biogas process according to Switzenbaum et al. [22], Chen et al. [23], and Eder and Schulz [24]. The well-known foam-triggering substances are proteins [6], polysaccharides [25], and lipids [14] as it was reported for sewage sludge AD and AD in the rumen. For this reason, the contents of these substances were analyzed. Furthermore, Miltimore et al. [26] assumed that several elements such as calcium, nickel, and zinc influenced the foam formation and stabilization in ruminant AD. For this reason, the analyses of elements that are soluble with water were carried out in digestates.

**Table 1 Operating data of the investigated waste utilizing biogas plants**

Biogas plant	Substrate	Organic loading rate	Mode of operation	Installed electric capacity	Retention time	Number of digesters	Agitation
BP A	50 thousand t/a swine and cattle manure, 30 thousand t/a biogenic industrial wastes from trade and industries	2 to 2.5 kg VS/(m <sup>3</sup> × day)	Mesophilic, two-stage (with an upstream hydrolysis stage)	2 × 469 kW <sub>el</sub> , (7.4 mil. kWh <sub>el</sub> /a)	25 days	Two hydrolysis digesters (520 m <sup>3</sup> each) 2 × digesters (2,300 m <sup>3</sup> each)	Continuous stirring with two paddles
BP B	8.320 t/a commercial food waste, 3.070 t/a vegetable materials, 3.040 t/a grease separator contents and flotation tailings, 1.400 t/a pastry wastes, 170 t/a miscellaneous (dairy wastewater, potato wastes, old bread grain sieving wastes)	2.8 kg VS/(m <sup>3</sup> × day)	Mesophilic, one-stage	860 kW <sub>el</sub>	29 days	Two digesters (1,000 m <sup>3</sup> each)	Recirculation and pneumatic
BP C	Sewage sludge, approximately 480 to 640 t grease separator contents in December 2009 to March 2010	2.5 to 3 kg VS/(m <sup>3</sup> × day)	Mesophilic, one-stage	No information	20 days	Four digesters (8,000 m <sup>3</sup> each)	Recirculation
BP D	36.500 t/a wastes and sludge from potato processing	2.8 kg VS/(m <sup>3</sup> × day)	Thermophilic, one-stage	400 kW <sub>el</sub> ; the biogas is burned and heat is used for the preparation of potato products	36 days	One digester (3,600 m <sup>3</sup> )	Recirculation
BP E	22.800 t/a grain waste products, 1.200 t/a grease separator contents	No information	Mesophilic, two-stage (with open mash and hydrolysis stage)	No information	30 to 35 days	One digester (5,000 m <sup>3</sup> )	Hydraulic

**Table 2 Parameters and analytical methods for the evaluation of the fermentation material**

Parameter	Sample pre-treatment	Analytical methods and instruments
Total solids (TS)	O	DIN 12880
Volatile solids (VS)	O	DIN 12879
pH value	CE/S	Microprocessor pH meter pH 95 (WTW, Germany)
VFA/TIC	CE/S	Titration method according to Nordmann [27]
Ammonium-nitrogen (NH <sub>4</sub> -N)	F	DIN 38406 E5, Spektroquant® test kit (measuring range 0.01 to 3 mg/L NH <sub>4</sub> -N, Merck, Germany), photometric measurement with MultiLab P5 (WTW, Weilheim, Germany)
Chloride	F	Spektroquant® test kit (measuring range 2.5 to 250 mg/L Cl <sup>-</sup> , Merck, Darmstadt, Germany), photometric measurement with SpektralPhotometer CADAS 200 (Dr. Lange, Düsseldorf, Germany)
Volatile fatty acids (acetate, propionate, butyrate)	F	High-performance liquid chromatography (Shimadzu, Japan); detector: RID-10A; column: VA 300/7.8 Nucleogel Ion 300 OA; eluent: 0.01 N H <sub>2</sub> SO <sub>4</sub> [28]
TOC/IC/TC/TN	B/S	TOC-VCSH/CSN with a TN unit (Shimadzu, Japan)
Crude protein	CA	Calculated according to Dumas method [29]: Crude protein = 6.25 × ([TN] – [NH <sub>4</sub> -N] – [NO <sub>3</sub> -N] – [NO <sub>2</sub> -N]) (The concentrations of nitrate- and nitrite-nitrogen were neglected due to their low concentration in the digestates as measured previously.)
Water-soluble elements	F	Inductively coupled plasma atomic emission spectrometry ICP-AES (Spectroflame, Spectro Int., Kleve, Germany)
Pattern of fatty acids	E	GC (System 5890 Series II GC, Hewlett-Packard, Wilmington, USA) after extraction according to Bligh and Dyer [20] and Morrison and Smith [21]
Surface tension	CE	Drop volume tensiometer (Lauda TV T-1, Lauda Dr. R. Wobser GmbH and Co. KG, Lauda-Königshofen, Germany)

## Results and discussion

### Survey of foam formation in full-scale biogas plants

The results of the survey on operators of full-scale biogas plants are summarized in Table 3. Fifteen out of eighteen biogas plant operators were willing to provide information about foam formation in their digesters. Twelve operators (i.e. 80% of informants) had experience with foaming in their digesters. Two respondents reported foam formation in the biological desulfurization step. Fifty percent of biogas plant operators regularly observed foaming in biogas digesters (Figure 1a), but 42% had no more problems with foaming at the time of the survey. Twenty-five percent of the operators could not identify the reason for foaming (Figure 1b), and 46% recognized specific substrates as being responsible for foaming in their fermenter. Substrates that contained proteins, fat, and grease were indicated as foam-forming agents.

Foam was generally suppressed by changing the process management strategy such as increasing the stirring intensity and adjustment of the substrate loading (Figure 1c). Furthermore, diverse additives such as micronutrient mixtures were used to bring excessive foaming under control. The most frequent foam control measures were the lowering of the organic loading rate (OLR) and the addition of anti-foaming agents.

In general, no information could be given about the economic consequences caused by foaming. Only one plant operator reported that foam damaged the roof construction

of the digester during an extreme foaming event. The cost of the damage here was estimated at €500,000.

The survey showed that foaming is a common problem not only in biogas digesters but also in the process of biological desulfurization of biogas. Similar observations had also been made by Kougiass et al. [14] who reported about foam formation in both the substrate storage and pre-digesters. The biogas plant operators mostly combated foam after it had appeared which is in agreement with the data published by Kougiass et al. [14]. Only a few preventive methods such as the addition of nutrient mixture were carried out. The most common cause of foaming according to Kougiass et al. [14] was the organic overload (44%) followed by feedstock which was high in protein and fat (31% in total) which contradicts the results of the survey described in this paper. A relatively high number of biogas plant operators cannot recognize the foaming causes in their digester (19% in [14], 23% in this study). For this reason, several extensive analyses of foaming causes in full-scale digesters were carried out (see Analysis of foaming causes in full-scale digesters).

### Analysis of foaming causes in full-scale digesters

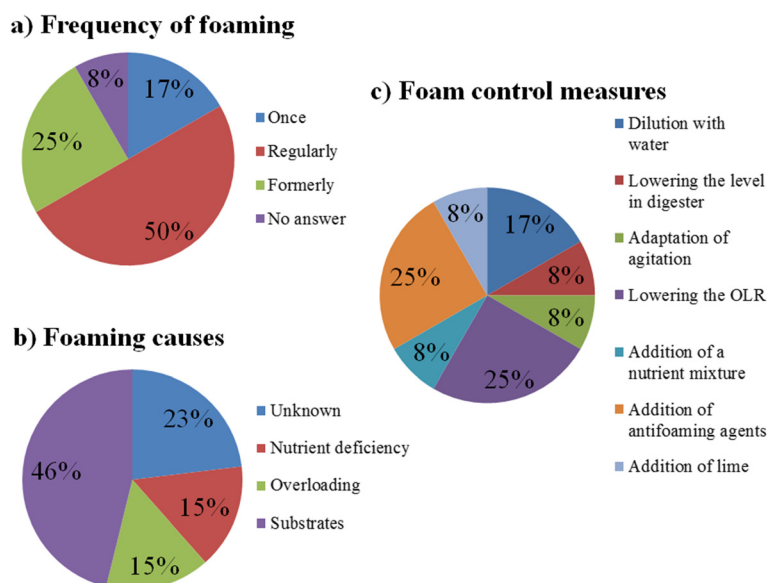
#### Biogas plant BP A

The biogas plant BP A was sampled every second week during 1 year. The aim of the intensive sampling was to study the differences in the behavior of physicochemical

**Table 3 Results of the survey of operators of biogenic waste-treating biogas plants regarding foam formation in digesters**

Biogas plant	Description of surveyed biogas plants			Survey results				
	Substrates	Capacity		Foam formation	Frequency of foam formation	Suspected causes of foaming	Foam control measures	Application of anti-foaming agents
Saxony								
1	Sewage sludge, separately collected biowaste and commercial waste	55,000 t/a	Two digesters á 2,300 m <sup>3</sup>	Yes	Once a year	Surfactants, animal protein, etc.	Dilution, lowering the level in digester, addition of lime	Yes
2	See information to BP E in Table 1			Yes	Always	Unknown	Agitation	No
3	Cattle manure, poultry manure, grease separator contents	28,000 t/a	Two-stage, mesophilic	Yes	Only at the beginning	Unknown	NA	NA
4	See information to BP D in Table 1			Yes	Formerly	Nutrient deficiency, overloading	'Starvation diet', adjustment of the feeding system	No
5	Poultry manure, green waste, kitchen waste, grease separator contents, old food, paper sludge	40,000 t/a	Two-stage, mesophilic	NA				
6	NA			NA				
Saxony-Anhalt								
7	Biowaste, food waste from discounter	50,000 t/a	Coupled dry and liquid production	Yes	Once	No more information available		
8	Biogenic waste	30,000 t/a	Single-stage, thermophilic	No				
9	Remains of the bioethanol production	100,000 t/a	NA	No (foaming problems only in the desulfurization step)				
10	Mash from a distillery, process wastewater from a potato peeling plant	2,400 m <sup>3</sup> /a	Single-stage (1,200 m <sup>3</sup> )	Yes	NA	Tensides	NA	NA
11	Poultry waste (litter from the poultry keeping, flotation tailings, and wastewater from the slaughter), grease and pizza residues	12,400 t/a	Two-stage, mesophilic	Yes	Twice per year	Nutrient deficiency	Addition of a nutrient mixture	Yes
12	Green waste, commercial waste, biowaste	21,000 t/a	Single-stage, mesophilic	Yes	Regularly	Unknown	Dilution of substrate with water	Yes
13	Food, commercial waste	46,000 t/a	NA	Yes	Once	Overfeeding	Reduction of the substrate loading	No
Thuringia								
14	Leftovers	72,000 t/a	Single-stage, mesophilic	Yes	Sometimes	NA	NA	NA
15	NA	NA	NA	NA				
16	Swine manure, paper fiber residual materials, slaughterhouse waste	NA	Single-stage, mesophilic	Yes	Formerly	Protein-containing slaughterhouse waste, cellulosic wastes from a paper mill	The operator tried miscellaneous, but nothing helped considerably	NA
17	Process wastewater from paper processing	NA	NA	No (foaming problems only in the desulfurization step)				
18	Cattle manure, biowaste, grease separator contents	30,000 t/a	Single stage (2× 800 m <sup>3</sup> ), thermophilic	Yes	Regularly	Grease separator contents	Reduction of the substrate loading	NA

NA, no answer.



**Figure 1** Survey results: frequency of foaming events in digesters (a), most common foaming causes (b) and foam control measures (c) in 15 biogas plants utilizing biogenic waste in Saxony, Saxony-Anhalt, and Thuringia.

parameters in a stable compared to an instable state. The foaming period occurred in the course of the seventh month of the sampling period and lasted 3 days. The biogas plant operator assumed that the reason for foam formation was an abrupt temperature increase from 35°C to 38°C in both digesters. A part of the digestate was pumped out in order to lower the level in the digesters, so that the foam could be stirred in by fixed-

positioned agitators. The foam disappeared after 1 week. No foam sample could be withdrawn from the digesters, so that only digestates were analyzed as described in 'Analyses of foaming causes in full-scale fermenters'.

Data on the chemical analysis of samples which were withdrawn before, during, and after the foaming period are presented in Table 4; the substrate charging during 16 weeks including the foaming period are presented in

**Table 4** Comparison of data from the foaming digesters D1 and D2 (biogas plant BP A)

Time of sampling		Two weeks before foaming		Day 1 of foaming		One week after foaming	
Digester	Index	D1	D2	D1	D2	D1	D2
TS [%]	O	3.53	1.77	4.17	3.45	3.25	2.85
VS [% TS]	O	56.5	33.5	66.0	64.4	52.1	45.6
pH [–]	CE	8.08	8.06	8.12	8.03	7.87	8.12
VFA/TIC [–]	CE/CA	0.16	0.13	0.11	0.12	0.09	0.11
NH <sub>4</sub> -N [g/L]	F	2.81	3.09	2.50	2.06	2.04	2.31
Total organic carbon [g/L]	B	6.52	5.44	11.1	8.68	9.10	11.26
Total nitrogen [g/L]	B	4.31	4.32	4.62	4.29	4.23	4.31
Crude protein [g/L]	CA	9.34	7.62	13.2	13.9	13.6	12.5
Acetic acid [mg/L]	F	159	182	31.4	26.5	29.5	76.7
Propionic acid [mg/L]	F	<1	<1	<1	<1	<1	<1
Butyric acid [mg/L]	F	<1	<1	<1	<1	<1	<1
Calcium [mg/L]	F	12.0	48.0	41.0	58.6	43.5	21.0
Magnesium [mg/L]	F	1.41	7.23	5.05	6.06	5.91	2.31
Phosphorus [mg/L]	F	300	284	316	305	248	190
Potassium [mg/L]	F	1,148	1,188	1,130	1,159	1,096	1,190
Sulfur [mg/L]	F	301	297	379	332	345	324
Surface tension [mN/m]	CE	57.7	57.0	60.7	60.2	58.0	57.0



Figure 2. The VFA/TIC factor expresses the ratio between the VFA content and total inorganic carbonate buffer, being in fact an indicator for the robustness of the system at high VFA concentrations. Each biogas digester has an own optimum value of VFA/TIC so that one isolated value can give us only little information and, as a consequence, only the comparison of values within one biogas digester makes sense. The VFA/TIC factor was only found to be slightly different in the period before, during, and after foaming and found to show a declining tendency from values of 0.16 (D1) and 0.13 (D2) down to 0.09 (D1) and 0.11 (D2), respectively. The acetic acid concentration decreased in both digesters during the sampling period. Both propionic and butyric acid were below the limit of detection (i.e. <1 mg/L) in all samples. Findings addressing the optimum VFA concentration in the literature have been rare. Hill et al. [30] reported that only concentrations of acetate higher than 13 mM (i.e. approximately 930 mg/L) indicate process imbalances. Thus, the acetate content in BP A was uncritical during the whole monitoring periods.

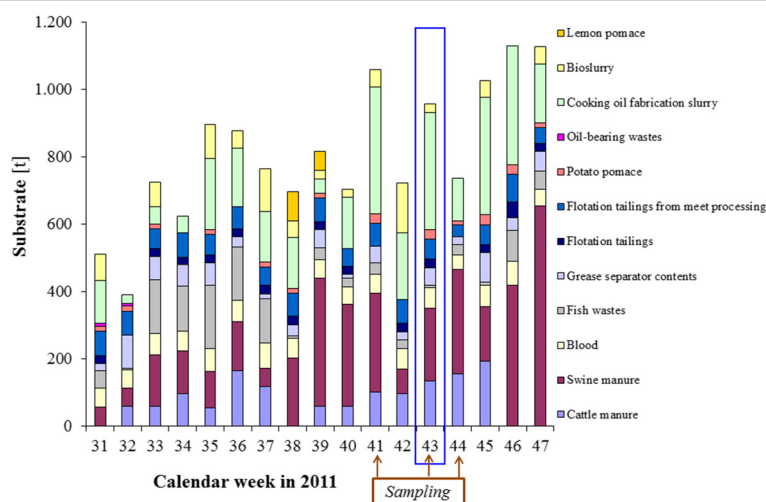
With regard to water-soluble elements, the calcium, phosphorous, and sulfur concentrations had slightly higher values during the foaming period as compared with the rest of the samples from non-foaming periods (Table 4). The magnesium concentrations had an opposite tendency in both digesters, while the potassium content stayed constant during the whole sampling period. The iron, zinc, and nickel concentrations were under the detection limit (i.e. <1.5 mg/L Fe, <2.5 mg/L Zn, and <2 mg/L Ni). As described by Miltimore et al. [26], calcium, nickel, and zinc were found to be associated with bloat in the rumen, whereas magnesium was not related to bloat. The authors also demonstrated that contrary to

nickel and zinc, calcium was associated with Fraction I protein which is assumed to be the cause of pasture bloat of ruminants [31]. On the other side, Moeller et al. [32] reported about a stabilizing effect of an addition of calcium and magnesium during sugar beet-based foaming in AD. The concentrations of elements as displayed in Table 4 do not allow us to draw conclusions about their influence on foam formation or stabilization as no maximum could be observed during the time of foaming.

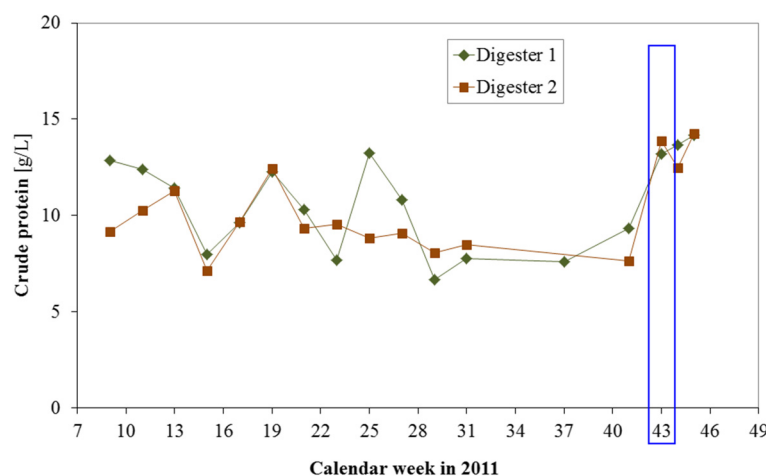
Ammonium nitrogen ( $\text{NH}_4\text{-N}$ ) declined during the sampling period from 3 g/L to 2 g/L. There are many studies concerning the effect of ammonium nitrogen concentration on the AD stability (see [23] for an overview). However, like in the case of VFA, no general optimum could be confirmed by the authors. On the other side, the decrease in the  $\text{NH}_4\text{-N}$  concentration shows that this parameter could not have a decisive effect on the foam-forming procedure.

The crude protein content increased from 9.3 (D1) and 7.6 (D2) to 13.2 (D1) and 13.9 (D2) during foaming as it is illustrated in Figure 3. In the 14 proceeding weeks before the foaming occurred, the crude protein concentration did not exceed 10 g/L. The crude protein content remained high after the foam has disappeared. The role of protein in foam formation has already been recognized by many authors (e.g. Ganidi et al. [6], Kougias et al. [14], and Clarke and Reid [31]).

The substrate charging in the period before, during, and after foaming is presented in Figure 2. There is a peak in the substrate feeding 2 weeks before foaming in the digester just as in the case of the foaming period described by Lienen et al. [5]. As the hydraulic retention time of the hydrolytic reactors was 11.2 days, the foam-producing agents reached the digesters during the second



**Figure 2** Substrate feeding before, during, and after the foaming period in BP A. The blue rectangle indicates the period when both digesters foamed.



**Figure 3** Crude protein concentrations before, during, and after the foaming period in BP A. The blue rectangle indicates the period when both digesters foamed.

week after their feeding into the biogas plant. The enhanced load during the 41st week may lead to an overload of the digester AD. The overloading was detected as one of the most common causes of foaming in AD as described by Kougias et al. [14] and Ganidi et al. [33]. Nevertheless, the concentrations of volatile organic acids and other chemical parameters (Table 4) did not support the theory of overloading. Thus, it is plausible that the excessive foaming was a consequence of the sudden temperature increase. Three effects may have played a role in foam formation and stabilization in BP A. The first effect is the change in the solubility of gas at higher temperatures when large amounts of gases (mainly carbon dioxide) are released [34]. Secondly, a sudden change of cultivation conditions may lead to higher mortality of microbial cells as already has been described by Eder and Schulz [24]. This would explain the higher protein concentrations in the digestate as a consequence of cell decomposition. Thirdly, the enhanced addition of cooking oil in week 41 could contribute to foam stabilization. According to Kougias et al. [35], both gelatine and sodium oleate have formed foam in the AD of manure-based biogas reactors. This theory supports the steep increase in total organic carbon (TOC) concentration in digestate during the foaming period (Table 4). As the TOC content remained at high levels, its effect on foaming cannot be confirmed.

The most probable theory is that the temperature increase caused the release of high gas amounts, which have been stabilized by protein and oil that have been loaded in high amounts into the digester.

#### **Biogas plant BP B**

The two egg-shaped digesters of the biogas plant BP B originally served as digesters in a former wastewater

treatment plant. Both digesters are connected together by using three overflows and can thus be operated as one tank due to the mixing of substrate by level adjustment during the feeding. The feed changed every 2 h from one digester to the other automatically.

The biogas plant operator often had problems with foam formation. Due to the egg shape of the digesters, the foam had not enough space in the headspace so that it tended to overflow. The foaming events were accompanied with high cleaning efforts as a consequence. According to the operator, the costs were estimated to be €500 to €600 for each foaming event. For this reason, both digesters were equipped with emergency overflows so that the foam can escape in the case of excessive foaming.

The biogas plant foamed regularly twice a week at the time of the first visit. The biogas plant operator identified dairy flushing water as the cause of the foam formation. The foam was reduced by the addition of plant oil. Three liters of rapeseed oil together with water were sprayed into the head space of the digesters. After performing this measure, the foam disappeared within a short time. Both the digestate and foam caused by loading of dairy flushing water were sampled and analyzed in August 2011.

In spring 2012, no more regular foaming was observed in BP B that could be connected with the loading of the digesters with dairy flushing water. The biogas plant operator believed that the reason for this was the stabilization of the fermentation material by the addition of old bread. Nevertheless, in February and mid-March, excessive foaming occurred that was definitely not associated with feeding the digesters with dairy flushing water. The operator supposed that the foam was caused by a particular charge of grease separator contents. However, the operator could



not identify the real cause of strong foam formation. The grease separator content was sampled. Furthermore, several other samples of grease separator contents of various origins were withdrawn. The contents of water-soluble elements as well as the chloride concentration were analyzed as described in Table 2 in order to search for the causes of foaming.

The next excessive foaming event occurred in May 2012. The foam formation was accompanied by a considerable decrease of 50% in biogas production. This time, the grease separator content from a particular restaurant was suspected to be the cause of the problems. The biogas plant operator assumed that prohibited chloride-containing disinfectants were used in the restaurant which entered into the grease separator contents. For this reason, the grease separator content was analyzed as described above. Furthermore, the digestate was sampled and analyzed as described in 'Analyses of foaming causes in full-scale fermenters'. This time, no foam was delivered by the operator to the laboratory.

The results of the analysis of foams and digestates are presented in Table 5, whereas the substrate charging during the foaming periods are shown in Figures 4 and 5.

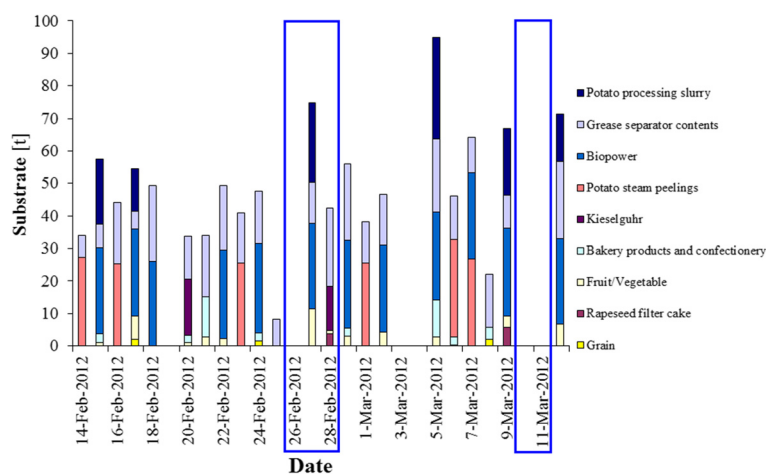
The first sampling occurred in August 2011 after the addition of dairy flushing water. The difference in crude protein concentration in digestate (15 g/L protein) and foam (2.8 g/L protein) showed that the foam was not

formed by protein molecules. On the other hand, the total organic carbon content in foam increased. Propionate and butyrate were the only carbonic substances analyzed which had higher concentrations in the foam than in the digestate, but the VFA concentration measured by using the titration method (see VFA/TOC in Table 2) reached lower values in the foam than in the digestate (1.34 g/L in foam vs. 1.61 g/L in digestate). Thus, it is highly probable that grease or its components played a certain role in foam formation and stabilization.

The causes of the foaming event in March 2012 were investigated. The separator content was analyzed regarding the elements which are soluble with water. The analysis data are shown in Table 6 in comparison with grease separator contents from other sources gained from BP B and from a canteen kitchen. The high variability in the content of elements especially in the case of calcium and potassium below the single samples is notable. Nevertheless, the analysis of the grease separator content, which was suspected to be a cause of foam formation, showed a slightly enhanced concentration of aluminum (1.42 mg/L) and zinc (10.5 mg/L). The effect of aluminum on biogas microbiology was studied by Cabirol et al. [36]. The authors found that this element inhibits bacterial activity. An addition of aluminum to the form of 1 g/L aluminum hydroxide led to a reduction in microbial activity of 50% in the case of methanogenic microorganisms

**Table 5 Analysis data of the digestates and foams of biogas plant BP B**

	Index	Digestate August 2011	Foam	Digestate April 2012	Digestate May 2012	Digestate June 2012	Foam	Digestate July 2012
		Foaming		Foam-free	Foaming	Foaming		Foam-free
TS [%]	O	4.82	4.70	3.87	4.87	n. d.	n. d.	4.68
VS [% TS]	O	72.1	79.9	51.2	55.6	51.9	50.9	53.1
pH [–]	CE	7.70	7.50	8.20	7.87	8.06	8.02	7.89
VFA/TIC [–]	CA	0.19	0.24	0.14	0.18	0.16	0.17	0.20
NH <sub>4</sub> -N [g/L]	F, S	1.38 (F)	1.89 (F)	1.58 (S)	2.55 (S)	n. d.	n. d.	3.95 (F)
Total organic carbon [g/L]	B	11.1	13.7	17.0	20.6	12.8	8.62	14.0
Total nitrogen [g/L]	B	3.77	2.36	5.43	8.81	5.86	5.12	6.36
Crude protein [g/L]	CA	14.9	2.83	24.0	39.1	n. d.	n. d.	15.0
Acetic acid [mg/L]	F	583	564	16.9	601	446	71.0	1,061
Propionic acid [mg/L]	F	63.5	162	0	81	17.6	0	125.9
Butyric acid [mg/L]	F	0	39.6	0	0	0	0	13.8
Calcium [mg/L]	F	85.2	38.8	97.1	119	67	31.6	158
Iron [mg/L]	F	4.42	4.69	14.1	4.44	5.82	5.02	6.00
Magnesium [mg/L]	F	10.1	7.02	15.8	34.2	11.7	29.0	39
Nickel [mg/L]	F	<2	<2	8.71	<0.8	<0.8	<0.8	2.4
Phosphorus [mg/L]	F	12.8	19.7	70.4	68.8	38.4	6.92	47.4
Potassium [mg/L]	F	1,700	1,190	1,602	1,606	1,500	1,546	1,762
Sulfur [mg/L]	F	30.8	28.2	59.2	62.2	33.8	33.6	38.6



**Figure 4** Substrate supplies in biogas plant BP B during February and March 2012. The blue rectangles indicate the foaming events.

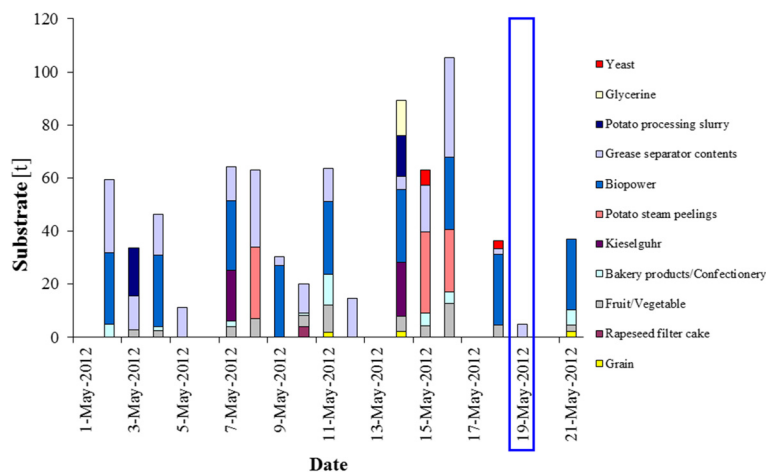
and of 72% in the case of acetogenic bacteria. The aluminum concentration in the grease separator content was indeed much lower at a value of 1.42 mg/L. Zinc also has an inhibitory effect on anaerobic digestion - in concentrations from 400 mg/L in the form of free ions and from 160 mg/L as zinc carbonate [37]. However, it is questionable whether the contents of both elements can cause negative effects after dilution in the digesters.

The investigation of the causes of foaming also included the analysis of the substrate supplies to the biogas plants during February and March (Figure 4). It is noticed that grain and bakery products were delivered 2 days before foaming. 'Grain' means grain sieve wastes that contained grain corn as well as coarse grain milled at various degrees of milling coarseness. Moeller et al. [38] described foam formation in the anaerobic digestion process due to the loading with coarse grain. The milling coarseness played a crucial role in the foaming propensity of grain: the finer

the grain was milled, the higher was the foam content in the foaming tests. Moreover, Moeller et al. [32] demonstrated that sucrose can also cause foaming in AD. Thus, it is very possible that the combination of these two components caused excessive foaming in the biogas plant BP B.

The analyses of the suspicious grease separator content showed no abnormalities as compared to other substrate samples (Table 6). Only the calcium concentration at 210 mg/L was somewhat higher than in the case of the majority of the samples. The analysis of chloride content showed that the chloride concentration in this grease separator content of 120 mg/L was the lowest of all of the tested substrates (Table 7). On the other hand, the chloride concentration in the digestate was higher at the time of foam formation than in older samples.

Considering the substrate supplies in May 2012, the delivery of baker's yeast before the foaming event is



**Figure 5** Substrate supplies in biogas plant BP B during May 2012. The blue rectangle indicates the foaming event.

**Table 6 Water soluble elements in grease separator contents used as substrate in BP B**

	Grease separator contents used at biogas plant BP B				Grease separator content from a canteen kitchen
	March 2012 (foaming event)	May 2012 (foaming event)	4th June 2012	18th June 2012	
Aluminum [mg/L]	1.42	<1.2	<1.2	<1.2	<1.2
Calcium [mg/L]	125	210	67.0	586	6.67
Iron [mg/L]	16.9	<0.6	5.82	37.8	1.53
Magnesium [mg/L]	18.8	41.0	11.7	80.4	6.37
Manganese [mg/L]	<0.2	<0.2	<0.2	2.32	<0.2
Phosphorus [mg/L]	31.7	66.6	38.4	130	14.6
Potassium [mg/L]	52.8	77.8	1,500	143	28.2
Sulfur [mg/L]	26.1	46.6	33.8	53.2	39.4
Zinc [mg/L]	10.5	<1.2	<1.2	<1.2	<1.2

Grease separator content from a canteen kitchen served as reference.

noteworthy (Figure 5). It is known that yeast supports foam formation [39]. For this reason, yeast is normally loaded in small amounts into the digester. Nevertheless, in this case, an employee at BP B was inattentive and loaded two pallets of yeasts into the substrate storage container at once.

#### Biogas plant BP C

The characteristics of biogas plant BP C have already been published in [11]. The four digesters were operated in cascades (digesters D1 and D3 in one line, digesters D2 and D4 in the second line). A mixture of primary sludge and surplus sludge served as the substrate for biogas production and was fed into the digesters D1 and D3. In addition, grease separator contents were fed into one of the digesters (D3).

Foam formation was detected in digester D3, where grease separator contents were co-digested over a period of 2 months (December 2009 and January 2010). The digesters D1, D2, and D3 were sampled at the time of foam occurrence in digester D3. Additionally, the foam was sampled in the overrun of the foaming digester. The samples were analyzed as described in 'Analyses of

foaming causes in full-scale fermenters'. Unlike the analysis of samples of the other biogas plants, the fatty acid patterns were analyzed in digestates and foams from all digesters. This analysis was performed in order to identify the influence of the grease separator contents on the digestate composition and to determine the foam-forming compounds.

The analysis data of the digestate and foam samples are summarized in Table 8. The digestate from the foaming

**Table 8 Analysis data of samples of the biogas plant BP C**

	Index	Digester D1	Digester D2	Digester D3	
		Digestate	Digestate	Digestate	Foam
TS [%]	O	3.55	3.44	3.20	3.43
VS [% TS]	O	n. d.	69.3	69.8	70.7
pH [–]	CE	7.33	7.24	7.36	7.64
VFA/TIC [–]	CA	0.05	0.06	0.08	0.06
NH <sub>4</sub> -N [g/L]	F	0.86	0.94	0.99	1.20
Total organic carbon [g/L]	CE	1.20	1.11	2.13	1.22
Total nitrogen [g/L]	CE	1.54	1.46	2.01	1.81
Acetate [mg/L]	F	2.2	21	30	59
Propionate [mg/L]	F	<1	5.5	3.6	10
Butyrate [mg/L]	F	<1	<1	<1	<1
Palmitic acid [%]	E	36.8	39.2	28.4	27.7
Stearic acid [%]	E	28.7	29.9	19.8	19.3
Oleic acid [%]	E	21.7	18.1	43.5	43.2
Linoleic acid [%]	E	10.9	9.6	5.8	6.8
Linolenic acid [%]	E	1.9	3.2	2.5	2.9
Surface tension [mN/m]	CE	72	72	71	71

Taken on 14th December 2009.

**Table 7 Chloride concentrations in samples of grease separator contents and digestates from the biogas plant BP B**

	Date	Chloride [mg/L]
Grease separator content (origin: foaming BP B)	March 2012	191
Grease separator content (origin: foaming BP B)	May 2012	120
Grease separator content (origin: restaurant in Saxony); serves as reference	May 2012	607
Digestate (origin: foaming BP B)	May 2012	3,266
Digestate (origin: non-foaming BP B)	November 2011	2,420
Digestate (origin: non-foaming BP B)	November 2011	2,370

digester had the highest VFA/TIC of 0.08, but the VFA/TIC values were in general very low in all digestates, when compared with samples from other biogas plants (e.g. Tables 4 and 5). The concentrations of ammonium-nitrogen and acetate, propionate, and butyrate were also comparatively low. The low VFA content shows that over-acidification can be excluded as the foaming cause in D3.

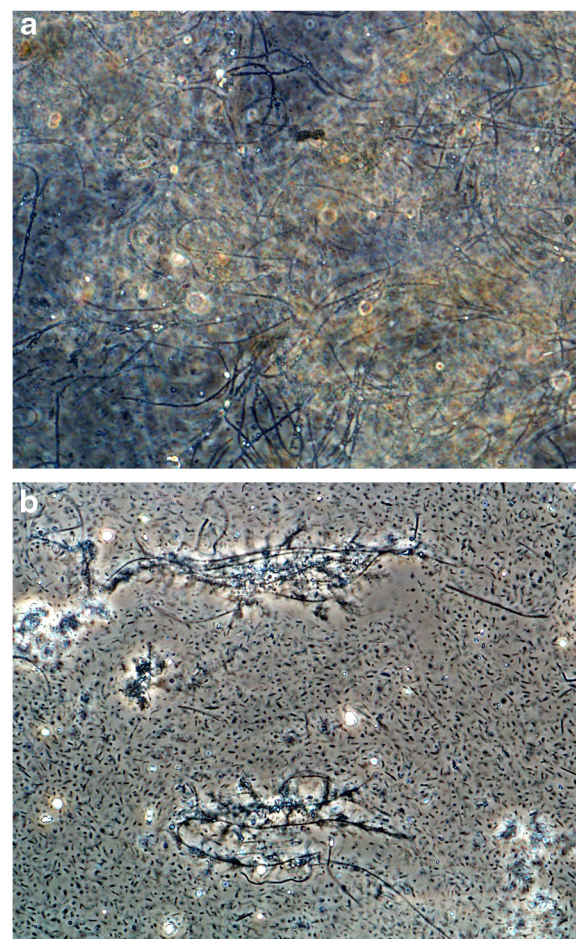
On the contrary, the concentrations of TOC, TN, and oleic acid were higher in digester D3 when compared to digesters D1 and D2, which can be traced back to the feeding of grease separator contents into D3.

Comparing the data of foam and digestate of D3, a slight accumulation of ammonium-nitrogen, acetate, and propionate in the foam fraction was observed. However, the content of long-chain fatty acids was not higher in the foam fraction, so it can be concluded that the foam was not produced or stabilized solely by the components of the grease separator contents. For this reason, the foam of D3 was evaluated under the microscope. This measure ultimately proved to be effective, as filamentary microorganisms were found to be responsible for foam formation in D3 (Figure 6a). Further closer microbiological analyses, including genetic fingerprinting and quantitative PCR (qPCR), have been carried out at the German Research Centre for Geosciences and are described by Lienen et al. [11]. The authors found that a higher abundance of the filamentous bacterium *Microthrix parvicella* occurred in D3 during two foaming periods than in both digesters D1 and D2.

#### Biogas plant BP D

The biogas plant BP D was visited for the first time within the scope of the survey of biogas plant operations. There were no problems with foaming at that time. The biogas plant operator stated that there was long-term foaming in the start-up stage, when biogas production also declined simultaneously. This problem was solved by adding a nutrient solution.

One year after the first visit, the biogas plant operator reported *about* excessive foam formation. According to the operator, several events occurred before foaming appeared: a new method of digestate processing was tested during full-load operation. The digestate was separated into liquid and solid phases; the solid phase was fed back into the digester. Changes in the digestate color from a light to a dark tone were observed during this phase. In addition, the wastewater treatment plant had been rebuilt with the aim of improving the dewatering of the sewage sludge. As a side effect of this, the phosphate concentration in the sewage sludge that was fed into the biogas plant was increased. After the implementation of the new technologies, the biogas plant started to foam. This problem was combated by a starvation diet and by



**Figure 6 Filamentous microorganisms in foam.** Filamentous microorganisms in foam from digester D3 of the biogas plant BP C (200x magnification) (a) and from the biogas plant BP D (400x magnification) (b).

pumping water into the digester. Unfortunately, the latter measure led to a considerable reduction in temperature of more than 10 K inside of the digester. The subsequent foaming was so strong that the foam overflowed. The biogas plant operator sampled the digestate and foam twice in an interval of 1 month and delivered the samples to the laboratory for analysis.

As the foaming problem was very serious, the biogas digester content had to be pumped out and the AD process had to be restarted.

The analysis data of these samples are displayed in Table 9. The acetate concentration in digestate was 95 and 109 mg/L showing a balanced AD [30]. The ammonium-nitrogen content in both digestates was lower than in all other biogas plants investigated in this study. For this reason, there was no indicating device for a disturbance of the AD.

The foaming cause could be the temperature fluctuations as reported in the case of BP A. According to Eder



**Table 9 Analysis data of samples from the biogas plants BP D and BP E**

	Biogas plant BP D					Biogas plant BP E			
	Index	Digestate	Foam	Digestate	Foam	Index	Substrate	Digestate	Recirculate
		First sampling (17th July 2012)		Second sampling (27th August 2012)			8th September 2011		
TS [%]	O	2.17	1.60	1.98	1.93	O	14.3	10.7	8.08
VS [% TS]	O	72.2	69.4	63.8	74.4	O	69.5	77.0	76.3
pH [–]	S	7.93	8.22	7.96	8.67	CE	8.23	7.18	8.10
VFA/TIC [–]	CA	0.24	0.19	0.16	0.20	CA	1.39	0.16	0.20
NH <sub>4</sub> -N [g/L]	F	0.55	0.33	0.64	0.67	F	5.05	4.01	4.07
Total organic carbon [g/L]	S	3.69	4.33	6.12	5.76	B	43.5	30.9	28.6
Total nitrogen [g/L]	S	1.03	1.23	1.81	1.68	B	7.78	8.63	6.62
Crude protein [g/L]	CA	2.99	5.65	7.36	6.30	CA	16.6	28.5	15.5
Acetate [mg/L]	F	95	27.9	109	30	F	1,762	143	51.6
Propionate [mg/L]	F	<1	<1	15.7	<1	F	868	7.60	0
Butyrate [mg/L]	F	<1	<1	<1	<1	F	313	0	0
Calcium [mg/L]	F	40.8	45.8	53.8	46.2	F	91.8	221	112
Iron [mg/L]	F	1.08	1.18	2.12	1.48	F	17.0	11.0	15.5
Potassium [mg/L]	F	2,140	2,220	2,560	2,500	F	3.98	4.08	4.70
Magnesium [mg/L]	F	11.6	9.82	12.6	7.94	F	211	84.8	139
Manganese [mg/L]	F	0.36	0.34	0.94	0.74	F	<0.5	<0.5	<0.5
Phosphorus [mg/L]	F	47.8	42.6	66.6	51.2	F	138.9	210.5	84.8
Potassium [mg/L]	F	2,140	2,220	2,560	2,500	F	3,905	3,775	3,085
Sulfur [mg/L]	F	14.0	23.6	18.4	24.2	F	117	106	110
Surface tension [mN/m]		n. d.	n. d.	n. d.	n. d.	CE	53.6	55.5	n. d.

and Schulz [24], sudden temperature changes of more than 1 K are very problematic, especially for thermophile biogas processes. Moreover, one other aspect was assumed to play a role in the foaming of BP D. Because the biogas plant was loaded with sewage sludge rich in phosphate, among other components, it was suspected that filamentous microorganisms could play a role in foam stabilization as described by Pagilla et al. [8], Lienen et al. [11], and Westlund et al. [9]. A microscopic inspection of foam did indeed show a lot of filamentous structures (Figure 6b) which could probably be the foam-forming agents in this case.

#### Biogas plant BP E

The biogas plant BP E had major problems with foaming. Foam was formed at all its process stages. The biogas plant operator did not use anti-foaming agents to remove the foam because no success had been achieved with the commercial defoamers. Thus, all stages were equipped with stirrers that operated continuously. Even the digestate storage had to be stirred continuously to avoid over-foaming.

The substrate mixture, the digestate, and the recirculate were sampled and analyzed (Table 9). The ammonium-

nitrogen concentration in the samples was very high; even in the recirculate 4.07 g/L NH<sub>4</sub>-N was measured. The reason for this lies in the digestion of the grain husk in high amounts. On the other hand, the concentration of acetate showed a balanced digestion process as described by Hill et al. [30]. The surface tension was lower here than in samples of all other biogas plants described in this paper.

As the NH<sub>4</sub>-N concentration was very high, the ammonia inhibition of the AD process can be assumed. Kroiss [40] showed in laboratory experiments that there has occurred a beginning limitation of the AD at ammonium concentrations of 3 g/L. On the other side, Chen et al. [23] demonstrated that there are many different data addressing the inhibitory effect of the total ammonia concentration in the AD ranging from 1.7 to 14 g/L. The authors also described an effect of acclimatization of the AD microbiology on high ammonia concentrations. Eder and Schulz [27] reported that the ammonia inhibition is often accompanied by a strong foaming. They suspected that the foam formation is caused by protein-degrading compounds. Furthermore, they showed that the recirculation of the digestate can cause an accumulation of ammonium in the digester. In fact, in the BP E phase, the separation of the digestate is carried out and the liquid phase aimed

at mashing the substrate. No ammonia stripping is carried out. On the other side, the low VFA concentrations do not strengthen the view that an inhibition of ammonia has started.

The foam formation is a common phenomenon in grain AD as described in Moeller et al. [38]. The authors reported that triticale and wheat formed more foam than rye, barley, and oats. The measured ammonium concentration of 2 g/L was not as high as in BP E. Both protein and starch were identified to be the foam-forming agents.

For this reason, it can be presumed that the strong foaming in BP E is caused by the digestion of grain waste products and the ammonia inhibition may not be assumed being the direct foaming cause in this case. Nevertheless, more investigations will be required to define the most proper foaming mitigation strategy for this biogas plant.

#### **Importance of the results for the research of foaming causes in full-scale reactors**

The survey of biogas plant operators showed that foam formation is a well-known phenomenon in full-scale biogas plants. Only little information was given by biogas plant operators about the costs for foaming. Excessive foaming can have serious economical consequences as described in 'Survey of foam formation in full-scale biogas plants'. On the other side, the costs can be limited to several hundred euros. According to the operator of BP B, the costs were estimated to be several hundred euros for each foaming event. However, the operators do not mostly know the real costs of a foaming event if no damage of biogas plant devices occurred.

The closer analysis of foaming events in five biogas plants further revealed that the search for foaming causes is often a very hard job as many factors may play a role. For example, physical effects such as temperature fluctuations are accompanied by foaming (BP A). Moreover, the composition of the substrate mix is of high importance as substrates containing proteins (BP B and BP E), grease (BP B), and filamentous microorganisms (BP C and BP D) were identified as foaming causes. The physicochemical analyses used in this study confirmed that the most common indicators of unbalanced AD did not indicate a process failure in the majority of foaming events. On the other side, the analyses of the foam and digestate compositions contributed to the indication of foaming causes. Unfortunately, the foam cannot be sampled in most of the biogas digesters as no access to the digestate surface is possible.

This study showed that the laboratory analyses have to be completed by the operational data of the biogas plants in the search for the causes of foaming. Primarily, the substrate loading diagram is a very important support.

Apart from that, the use of batch foaming tests as described by Moeller et al. [19] may be helpful. This method is suitable for both laboratory research and for on-site estimation of the foaming propensity of substrates. As the filamentous microorganisms are well visible under the microscope, the microscopic observation of foam may also assist the examination of foaming causes. In this way, the biogas plant operators are able to identify the most foaming causes by themselves. After that, they can apply some of mitigation strategies that are suitable for the specific foaming cause. As an example, in the case of the foaming due to the presence of filamentous microorganisms, methods of ultrasonic pre-treatment of feeding sludge were developed by Barjenbruch et al. [10]. However, new methods of foaming mitigation have to be developed for the use of 'risky' substrates like for example grain and yeast.

#### **Conclusions**

This study displayed a high frequency of foaming events and identified the main causes of foaming in full-scale biogas plants. A survey of biogas plant operators in three German federal states showed that 80% of biogas plants had problems with foaming. A closer analysis of foaming events in five biogas plants enabled the identification of the causes of foaming in five cases. In general, a correlation was found between the use of substrates for biogas production and foam formation. The foam-provoking substrate mixtures contained not only grain, bakery, and confectionery products but also yeasts and filamentous microorganisms. Furthermore, sudden temperature increases by 3 K had provoked foaming in one biogas plant. While excessive foaming is a frequent phenomenon that often has serious consequences for biogas plants, the causes of foaming usually remain unidentified in practical applications. In addition, it was not possible to determine the reasons for foaming in our research in some cases. Foam formation in the anaerobic digestion process has begun to attract increasing attention within the context of the improvement of the efficiency of the entire biogas production process. This is the first publication concerning the establishment of the causes of foaming in full-scale biogas plants which process biogenic waste that shows an abundance of factors playing a role in foam formation and stabilization based on real data.

The next research study should focus on foaming mitigation strategies in full-scale fermenters to explore the effects of specific substrates as for example on grain AD. In addition, new methods of substrate pre-treatment before its loading into the fermenter should be developed in order to prevent the foam formation in biogas digesters.



## Abbreviations

BP: biogas plant; CA: calculated; CE: centrifuged; D: digester; E: extracted; F: filtered; IC: inorganic carbon; O: origin sample; P: pureed sample; S: sieved sample; TC: total carbon; TIC: total inorganic carbonate buffer; TOC: total organic carbon; TN: total nitrogen; TS: total solid content; VS: volatile solid content; VOA: volatile organic acids.

## Competing interests

The authors declare that they have no competing interests.

## Authors' contributions

LM led the project, collected and sorted the publication and information material, and prepared the manuscript. KG surveyed the operators, analyzed the samples, contributed with critical reading of the manuscript, and provided input for the final version. Both authors read and approved the final manuscript.

## Acknowledgements

This project was funded by the German Federal Ministry for the Environment, Nature Conservation and Nuclear Safety on the basis of a decision of the German Bundestag and the Initiating and Networking Fund of the Helmholtz Association. The authors would like to thank all the biogas plant operators who provided us with substrates and fermentation material. The authors would also like to extend their best wishes to Prof. Dr.-Ing. Andreas Zehnsdorf on the occasion of his 50th birthday.

Received: 17 June 2014 Accepted: 11 December 2014

Published online: 17 January 2015

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