







Review

Foams in Wastewater Treatment Plants: From Causes to Control Methods

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Received: 21 March 2020; Accepted: 11 April 2020; Published: 15 April 2020



Abstract: The formation of persistent foams can be a critical problem in wastewater treatment plants (WWTPs) as it could lead to a series of operational problems, especially the reduction of the overall system performance. To date, the effects of foaming in the WWTPs are a problem that is currently very common and shared, but which to date is treated mainly only at the management level and still too little studied through a globally shared scientific method: the complexity of the phenomenon and the systems have led to numerous partially contradictory descriptions and hypotheses over the years. The goal must be to suggest future research directions and indicate promising strategies to prevent or control the formation of foams in WWTPs. This study examines and investigates the problem of foams by a methodological approach of research through a review on the state of the art: the factors influencing the formation of foams are described first (such as surfactants and/or extracellular polymeric substances (EPSs)), then the known methods for the evaluation of foaming, both direct and indirect, are presented, with the aim of identifying the correct and best (from the management point of view) control and/or prevention strategies to be applied in the future in WWTPs.

Keywords: foaming; chemical and biological foams; evaluation method; foam test; control method; WWTP

1. Introduction

Chemical and biological foaming in conventional activated sludge (CAS) plants [1] has been under study since the 1960s [2], and, in general, in wastewater treatment plants (WWTPs) is an issue that adversely affects worldwide the aeration tank and/or the final clarifier [3–11]. Foams are a set of stable bubbles, produced when air or other gases are introduced below the surface of the liquid that expands to enclose the gas with a liquid film called “lamellae” of the foams [12,13]. A condition necessary for foaming is the presence of surface-active components or surfactants in the liquid [14] because these substances settle on the interfaces between the gas and liquid and reduce the surface tension [15]. Gas bubbles in the system are originated from aeration and mixing in oxidation tanks and from gas production in denitrification reactors, anaerobic digesters [16] and in nitrification/anammox reactors [17–19]. The formation of persistent foams may be a critical issue as it could lead to a series

of operative problems including: (i) reduction in oxygen transfer, (ii) decrease in the concentration of biomass in the biological reactor, (iii) odor problems and (iv) increase of the management and maintenance costs [12,20,21]. The production of uncontrolled foams in large volumes causes a physical hazard for operators due to exposure to pathogens, obstruction of passageways and formation of slippery surfaces causing dangers of falling [20,21].

In recent years, foaming has also been studied in membrane bioreactors (MBRs) [2,21–23]. Membrane processes, by replacing the gravitational sedimentation tank of a CAS process, allow retaining all solids, with a size greater than the porosity of ultrafiltration or microfiltration, inside the reactor. The main management problems that derive from this are (i) the fouling of the membranes and (ii) the formation of biological foams [24,25]. The recirculation of the mixed liquor at the head of the membrane bioreactor worsens these conditions and the tank in which the MBR unit is submerged may become a real trap for foams [5,21].

Foaming represents also one of the most common operating problems in anaerobic digesters (ADs) of WWTPs and the main critical aspect deriving (in addition to those previously described) is the reduction of digester performance, which results in a lower production of biogas and volatile solids degradation [26]. Other serious operational problems that can be encountered are (i) gas mixing blocks, (ii) fouling of the gas collection pipes, (iii) covering of the digester wall with solid foams, (iv) proliferation of pathogenic bacteria due to the reduction of the active volume in the digester, (v) maintenance interventions for cleaning biogas pipes and (vi) foam spills with the formation of slippery areas [26,27]. From the AD management point of view, the economic losses deriving from the phenomenon of foams are another important and not negligible aspect. The estimation of the costs is not easy to quantify because different parameters must be included in the count: the cleaning costs of the digester, the repair costs and the personnel costs for increased monitoring and maintenance are equally essential [28–30]. The aspect of worker safety must not be underestimated. For instance, very dangerous explosion phenomena have also been observed in some barns in the USA following the formation of stable and thick foams in ADs [28].

The formation of foams can also reduce the overall WWTP performance, therefore in order to optimize its management, causes, quantification and removal of foams must be investigated [12,20]. As already expressed in literature [31,32] for drinking water treatment plants, also for WWTPs the same approach of monitoring and optimization could be applied. This would guarantee to cope with the increasing presence of industrial contaminants in wastewaters [33–35] and would allow producing a sludge that respects strict limits for the reuse in agriculture [36,37].

This review aims at examination and investigation of the problem of foams through the methodological approach of research to identify the correct control and/or prevention strategies in WWTPs; therefore suggested future outlooks on the basis of previous studies in this topic area are presented and discussed at the end of this work.

2. Bibliometric Research and Structure

In order to focus the problems of foams in wastewater with the causes and circumstances associated, the literature search was performed in the Scopus and Google Scholar databases by entering “foam wastewater” and “foaming wastewater” as keywords. To get a review on the currently known methods for the evaluation of foaming and the most applied strategies for controlling foaming directly in WWTPs, “foam test wastewater” or “foam control wastewater” were used as keywords in the same databases. In Scopus database the analysis has been conducted searching the keywords on fields “Article title, Abstract, Keywords” to focus attention on works closely related to the management application field in WWTPs. The research focused mainly on works published since 1990. A first screening of the literature was carried out, discarding the articles and reviews that do not analyze (i) the causes and inconveniences related to the formation of foams in WWTPs, (ii) the evaluation methods or (iii) the control mechanisms that operators can apply to manage the problem in WWTPs. At the end of this screening, about 120 papers were found. Focusing only on the field of environmental

science and engineering rather than on chemical engineering and chemistry, almost 90 articles were selected for the review.

The spatial and temporal distribution of publications concerning the foam formation in the WWTPs carried out since 1990 is shown below. In Figure 1 the period from 1990 to today is divided into time intervals of 5 years and for each one, the total number of international scientific publications on foaming in WWTPs is reported. It is possible to notice how over the years there has been a growing interest in the problem, this has led the world of research to focus more on the issue. In fact, since 2015 there have been 30 publications, 20 more than the first five years considered.

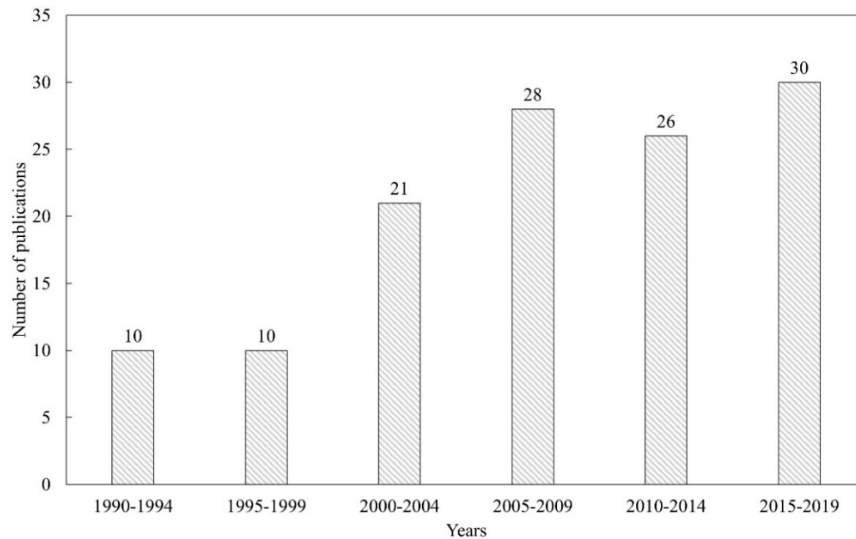


Figure 1. Number of publications per year on Scopus introducing the words “foam wastewater” and “foaming wastewater”.

The map of Figure 2 shows the origin of the scientific publications on foams in the WWTPs. The country reported in a scientific publication indicates the nationality of the first author. The largest number of research and review papers on foams in WWTPs was written in the United States and China, respectively 21 and 20. Italy and Australia are placed in third and fourth position with 11 and 10 publications, respectively.

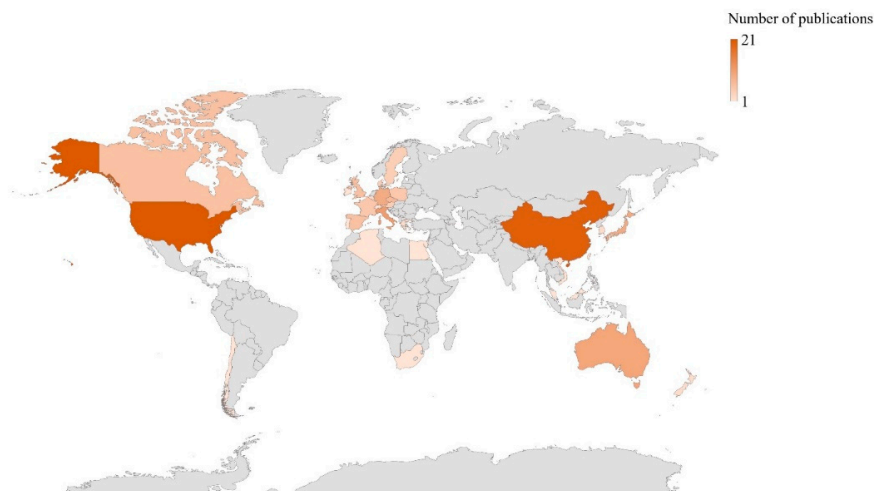


Figure 2. Number of publications by country on Scopus introducing the words “foam wastewater” and “foaming wastewater”.

In conclusion, it is possible to state that this topic is not yet very studied in the world, but over the years a greater interest is developing more and more. Publications are on the rise and on a geographical level this issue is being addressed in more and more countries. Foaming in the WWTPs is a very sensitive issue that is only dealt with at the management level, so in recent years there has been a growing perception of the need to deepen the study through a methodological research approach.

The review consists of four parts; Section 3 reports the causes of the formation of foams, focusing mainly on the biological ones in CAS, MBR processes and ADs. Instead, in Section 4 the main methods of evaluation of the foams are explored, describing both the direct methodologies (including tests applicable directly on-site) and the indirect ones of characterization of the mixed liquor. Section 5 discusses the application of foam control methods, mainly describing the physical and chemical ones most used on the real scale. Finally, in Section 6 a brief discussion is presented on the main advantages and disadvantages of the evaluation and control methods, focusing on possible future outlooks.

3. Causes

Foaming formation occurs either chemically or biologically [2,14,23,38] and in both cases, a necessary condition is the dispersion of a gas in a liquid [12,13,26]. The chemical foams are caused mainly by excess surfactants (white foams), which have many industrial applications such as washing and cleaning (detergents), pharmaceuticals, cosmetics, textiles, tanneries, agriculture, biotechnology, food, paints, microelectronic, metallurgy, oil recovery and paper, among others [35,39–41]. The presence of surface-active compounds affects the rheology of the fluid interfaces because they can cause variable viscosity, elasticity and surface tension gradients [42]. The factor that contributes to the arising of the biological ones (brown foams) is the growth of particular filamentous bacteria, the so called “foam-former”, and/or the presence of hydrophobic high-molecular-weight substances synthesized by bacteria present in mixed liquor, although not the foam-former [2,4,14,43–45]. Biological foams are more viscous and stable than foams induced by synthetic surfactants and the most foam problems in WWTPs are associated with them: a three-phase phenomenon that involves a liquid, a solid and a gas [23,26]. The hydrophobic products described above are extracellular polymeric substances (EPSs), which have properties of surface-active agents [22]. The EPSs are present both outside of cells (bound EPS (EPS_b) and soluble EPS (EPS_s)) and in the interior of microbial aggregates. Their major components are carbohydrates and proteins; but humic substances, lipids, nucleic acids, uronic acids and some inorganic components can also be found in EPSs [46]. In general, their concentration plays a key role in making the foams more stable [47,48]. The bacteria in the mixed liquor can produce EPSs through a variety of biological processes (including active secretion, substrate degradation, biomass decay, shedding of cell surface material and bacterial self-hydrolysis of floc-associated EPS) [46,49,50] or under biological or mechanical stress conditions [51]. These disturbing conditions for biomass can be caused by the presence of toxic substances in wastewater, such as surfactants, that can damage the cell membrane or by a strong imbalance between organic matter and nutrients with consequent production of EPSs [2,38,46]. A deviation from the correct balance (BOD:C:P = 100:5:1 in CAS processes), such as an important lack of phosphorus significantly influence EPSs production [2,46]. The EPS concentration is increased by the presence of foam-forming and filamentous microorganisms in the mixed liquor of which they cause a worsening of the potential foaming phenomenon [52–54]. Among the causes of the formation of filamentous bacteria, in addition to a low ratio between organic matter and microorganisms [2,5,23], there may also be some particular operative conditions such as low concentrations of dissolved oxygen (indicatively < 2 mg L⁻¹) in the biological reactor and high retention times of solids that create selective conditions for the growth of foam-former microorganisms [23,55,56].

In CAS processes, the most observed filamentous taxa seen are *Candidatus* ‘*Microthrix parvicella*’, a gram-positive filamentous bacterium, and members of the *Mycolata*, bacteria containing mycolic acids in their cell walls. All are members of the order Actinomycetales in the phylum Actinobacteria [57–60]. Several mycolic acids containing Actinomycetes (*Mycolata*) have been found in biological foams and

within this group, *Gordonia amarae*, *Skermania piniformis* and *Rhodococcus rhodochrous* are the main organisms previously identified in foaming microbial communities [61].

Although foaming originally was associated with CAS plants, recently this phenomenon has been observed also in other treatment processes such as MBRs and ADs [2,21–23,46]. There is little information available concerning the microbial community of MBR processes and almost nothing is known about the potential micro-organisms that attend the foam formation [23]. The predominant filamentous bacteria in the foaming MBR process appear to be the HGC bacteria, including Actinomycetes, *Nostocoida limicola* III, *Microthrix parvicella*, Eikelboom type 1851, Eikelboom type 0041, *Gordonia* and *Cytophaga* [54]. All these bacterial species play a fundamental role in stabilizing the foams because they are hydrophobic as in their cell walls there is the mycolic acid [62]. *Microthrix parvicella* is one of the most frequently observed filamentous bacteria in foaming at activated sludge and the results obtained by Fan et al. [63] indicate that a low sludge load condition could induce a significant increase in these bacteria. In MBR systems, foams can also form in the absence of surfactants from the incoming wastewater and without a large concentration of hydrophobic bacterial strains (foam-forming bacteria). The amount of foams, in this case, is related to the concentration of EPSs in the mixed liquor, which influenced the formation of foams but not its stability [23]. As stated jointly by the scientific community [22,49,51,52,64–66], in MBR plants, EPSs cause membrane fouling, which can also lead to the obstruction of the membrane thus blocking the operation of the whole plant. Therefore, it is advisable to study the formation of foams in MBR processes with the aim of keeping membrane fouling under control. The EPS, therefore, in addition to causing the fouling of the membranes are also foaming agents [67]. The presence of filamentous organisms and surfactants in the incoming discharge can cause an increase in the concentration of extracellular substances, leading to worsening of both membrane fouling and foaming phenomena [53]. The mechanisms that lead to the formation of foams in MBR and CAS systems are the same, the main difference between the two systems lies in the fact that MBR process tends to retain most of the EPSs, so as to become a real and own “foam trap” [21].

The ADs provide a favorable environment for the formation of three-phase foams as all the components are present: liquid, surface active agents and solid particles and gases (biogas). In their review, Subramanian and Pagilla [26] reported that the foaming of a three-phase system is directly proportional to the concentration of particles and inversely proportional to the particle size: in their three-phase foams scheme, the particles and gas bubbles are relative of the same dimensional order. Instead, in the biological foams of the CAS process, the gas bubbles are much larger than the particles. Foam formation in anaerobic digesters is still a poorly understood mechanism and the studies in this regard are less thorough than those carried out for the CAS and MBR processes: there is, therefore, an urgent need for research in this field to clarify the conditions that trigger the generation of foams in biogas reactors and to develop new solutions for the prediction and prevention of foam formation [28].

4. Evaluation Method

Despite the range of methods that have already been used to evaluate foaming characteristics, to date a shared procedure has yet to be developed to assess the potential risk of formation of stable foams on the surface of the aeration basin of a WWTP [62]. There are several different methods for generating and characterizing foams on a small scale (including whipping, stirring, plunging, shaking, oscillating, pouring and bubbling) [20,68] and the most used are (i) sparging or diffusing gas (gas is sparged inside a tube with a glass sinter at the bottom into which a known volume of test liquid is introduced) and (ii) shaking (liquid/gas mixture in a bottle is shaken for a specified period of time). To date, the most widely used procedure consists in the bubbling of gas through the liquid, which consists of the basic practice that characterized the Bikerman test, one of the first experiments on foams: introduction of air at a fixed flow rate through a disc of porous glass on the bottom of a glass column [20,69]. In recent years, the scientific community has been moving in this direction, mainly carrying out a series of foamability tests that simulate the aeration conditions in a plant [21]. This approach allows obtaining

a series of useful information on the quantitative and qualitative characteristics of foams that are produced following aeration.

Foams evaluation methods include direct methodologies (analysis of the foams following their formation), including those that can be applied directly on-site at an aeration basin. There are also other methods that allow the assessment of foams by indirect means: quantification of surface tension, evaluation of the concentration of EPS and filamentous bacteria and the characterization of hydrophobicity and viscosity.

4.1. Direct Methods

4.1.1. Foam Rating (FR)

The first foam-based test is the foam rating (FR) introduced by Blackall et al. [13], according to which the foams have been classified on the basis of four parameters: foam volume, bubble size, foaming speed and time required for foam collapse after aeration has ceased. Foams generated are classified based on the foam rating, which can take a value from 0 to 7. When the mixed liquor does not produce foams and the reaction to aeration as for pure water, the value of FR is zero; on the other hand, the value 7 is associated to a situation with a formation of dense and very stable foams [21,70]. Therefore, FR is a subjective measure of the quality and type of foams. Blackall et al. [13] proposed a foaming apparatus with a flow rate of air of 200 mL min^{-1} : 50 mL of activated sludge sample to be tested was poured into a glass cylinder (500 mm high, 40 mm diameter) and, through a sintered glass diffuser with porosity in the range 40–90 μm , the gas bubbles went up the liquid when the aeration was started. Foams produced were also rated by Stratton et al. [71] and Hug [16] on the arbitrary scale proposed by Blackall et al. [13], and also Di Bella and Torregrossa [21] applied the FR to define the foam generation and stability in MBR processes.

4.1.2. Volume

Patist et al. [72] adopted two different mechanisms for the foam production based on the techniques proposed by Blackall et al. [13]: formation of foams by agitation and formation of foams by controlled aeration, as described below.

- Shaking: a 100 mL graduated cylinder containing 15 mL of sample for 10 s was shaken vigorously. Foaming was recorded as the volume of foams produced immediately after shaking.
- Air bubbling: an apparatus consisting of a 50 cm high quartz column with a diameter of 3.5 cm and a 0.25 cm capillary on the bottom, was used to generate bubbles. Fifty milliliters of sample were poured into the column using a long funnel that reached the bottom in order to ensure that the cylinder walls remained dry (in this way the matrix was only on the bottom and not on the walls). The airflow was constant at a flow rate of $158.2 \text{ mL min}^{-1}$. The foam volume produced after 2 min was recorded.

Both methods were used to study the properties of foams generated by sodium dodecyl sulphate (SDS) mixed with long-chain alcohols (C_nOH).

The Waring blender method is an assessment method for the foaming capacity proposed by Zhao et al. [73] and similar to the test adopted by Patist et al. [72]. The water used in the experiment of primary selection of foaming agent is the formation water from an oil production plant and the foaming properties of six kinds of foaming agent (lauryl sodium sulfate, sodium dodecyl benzene sulfonate, HY-2, SJ-6, SJ-8 and DY-1) were evaluated. The procedure was the following: 100 mL of solution to be tested was placed in a beaker and mixed at high speed ($>1000 \text{ r min}^{-1}$) for 3 min. At the end of the agitation, the volume of foams produced was measured, correlated with the foaming capacity of the tested material.

4.1.3. Foam Power (FP)

The foam power test proposed by Nakajima and Mishima [22] is a direct measure of the foaming potential and it was developed in accordance with the concepts used in food science [74–76]. This test is among those most used in the foaming phenomenon in WWTPs and in the scientific literature it has almost always been applied following the measurement procedures reported by Nakajima and Mishima [22]. In their experiments, a 100 mL sample of activated sludge was placed vertically in a transparent acryl cylinder (30 mm diameter × 1000 mm height). The tests were conducted with an airflow of 5 L min⁻¹ supplied from the bottom of the cylinder during 20 s or 30 s. Albumin solution and EPS extracted from the activated sludge were used as the samples for foaming examinations and the activated sludge was from small scale domestic WWTPs with an MBR process.

The other researchers, in the experiments carried out in the following years, have applied the same procedure by varying only some operating conditions such as the volume of the sample or the airflow. The foaming power index (FP) is determined as the volume of sample consumed by the foams per liter supplied with air through Equation (1) [22]:

$$FP \left[mL L_{air}^{-1} \right] = \frac{(H_0 - H_1) * S}{Q * \Delta T} \quad (1)$$

where:

H_0 : the level of the interface between air and the matrix before aeration (cm);

H_1 : the level of the interface between the matrix and the foams after an aeration interval ΔT (cm);

S : the cylinder area (cm²);

Q : the aeration flow rate (L_{air} min⁻¹);

ΔT : the aeration period (min).

For the determination of foaming scum index (FSI; which is described below), Fryer and Gray [62] proposed an alternative approach to the quantification of foaming potential. Instead of the level of the interface between the matrix and the foams after aeration, the height reached by the foams in the column following aeration is measured. In this case the foam power, called FP2, can be quantified using Equation (2):

$$FP2 \left[mL L_{air}^{-1} \right] = \frac{(H_2 - H_0) * S}{Q * \Delta T} \quad (2)$$

where:

H_2 : the level of the interface reached by the foams at the end of the aeration (cm);

H_0 : the level of the interface between air and the matrix before aeration (cm);

S : the cylinder area (cm²);

Q : the aeration flow rate (L_{air} min⁻¹);

ΔT : the aeration period (min).

In the study of Fryer and Gray [62], a sample of mixed liquor (150 mL) was aerated for a period of 1 min at a controlled flow rate of 0.5 L min⁻¹ during which time the foaming potential was determined as a percentage of the maximum expansion of foams achieved in relation to the starting height.

4.1.4. Foam Stability (FS)

Another method to characterize foams is to assess their stability. Foam stability (FS) is the ability of the foams to resist overtime after interrupting the aeration. It was evaluated by Nakajima and Mishima [22] taking into consideration the fact that the level of foams decreased very quickly at first and then decreases more slowly. In fact, the total height of foams $H(t)$ is divided into two phases, slow and fast, as follows in Equation (3) [22]:

$$H(t) = H_1 - k_1 t + H_2 - k_2 t \quad (3)$$

where H_1 and H_2 are the initial heights of the slow and fast process respectively. $H_1 + H_2 = H_0$ and $k_1 > k_2$. The height H_1 is usually measured every 10 s until the liquid–foam interface level reaches H_0 . FS is estimated using the higher speed constant k_1 since only the change of $H(t)$ of the rapid phase is caused by the breaking of the foams, according to Equation (4) [22]:

$$FS [min] = \frac{\ln 2}{k_1} \quad (4)$$

where k_1 is identified by relating the logarithm of $H(t)$ to time and considering only the values of the first-time interval (rapid decrease). $H(t)$ is to be considered the difference between the level of the liquid before aeration and the level of the liquid at time t after turning off the aeration. In Nakajima and Mishima [22] experiments, the same samples and the same operative conditions as those already described for the Foam Power test were used.

The stability of the foams is usually assessed together with the foaming potential to describe the foams produced from a qualitative and not just a quantitative point of view, for example, FS was measured by Fryer and Gray [62] as the time taken by all bubbles to collapse following a one-minute aeration period and all tests were conducted under ambient temperature conditions (20 °C) using activated sludge samples at a predetermined solids concentration of 3 g L⁻¹.

4.1.5. Scum Index (SI)

The scum index (SI) is used to control biological foams in CAS and its quantification is based on the flotation induced by standardized and constant aeration, as proposed by Pretorius and Laubscher [77]. In these tests, an aliquot of mixed liquor equal to 2 L was placed in the flotation cell (80 mm internal diameter, 500 mm height) and aerated with an intensity of 10 L_{air} L⁻¹ h⁻¹ for 15 min. During the aeration a stable foams layer could be formed. The aeration was then suspended, and the foam layer was separated from the mixed liquor by draining the latter. The remaining foam layer was removed from the flotation cell and collected. The removed non-floating mixed liquor was placed again in the flotation cell and the separation–flotation process was repeated until no more foams were formed (aeration times of 15 min in the first hour and subsequently at 60-min intervals). Since some non-foaming microorganisms were separated with the foaming microorganisms, the foaming microorganisms (which were collected) were further purified as follows: these foams were resuspended in tap water and the foam separation process was repeated until the concentration of suspended solids (SS) in the non-floating part was less than 3 mg L⁻¹. The dry mass of the recovered/purified foams was determined, and the scum index was calculated as follows in Equation (5) [21,77]:

$$SI [-] = \frac{\text{mass of recovered foam}}{\text{mass of suspended solids (SS) initially presents}} * 100 \quad (5)$$

The original method proposed by Pretorius and Laubscher [77] has been slightly modified to define the specific role of EPS concentration in MBR foam formation: the results of the flotation operation alone, without the subsequent dilution and purification, were also considered as a modified scum index test since these last steps can negatively influence the effects of the EPS [21,23].

4.1.6. Foaming Scum Index (FSI)

Fryer and Gray [62] proposed a global approach specifically for the characterization of CAS foaming through the foaming scum index (FSI). FSI has been formulated as an algorithm using data produced from a series of individual tests normally employed to investigate foaming such as foam potential, foam stability and filamentous bacteria. Seven parameters were used in FSI formulation: foam color, bubble size, foam stability, foam potential, coverage and filament abundance within foams. To increase the robustness of the model, foam potential (FP2) has been measured using the glass column sintered disc method following procedures described by Fryer et al. [20]. Hypotheses have been put

forward on the influence of each variable on the others and field studies have been conducted in 10 different WWTPs, which employed activated sludge as the main treatment process. A preliminary weighting scheme was proposed in order to convert measured values into standardized scores so that parameters were comparable on the same scale: lower scores reflect a smaller contribution of each parameter to activated sludge foaming. Multivariate analysis and structural equation modeling (SEM) have been used to weight each parameter within the FSI, considering the relevance of each to foaming in CAS plants. FSI is calculated as follows in Equation (6) [62]:

$$FSI = \left(\sum_{n=7}^{n=1} p_i w_i \right)^2 \quad (6)$$

where *FSI* is foaming scum index (from 0 to 25), p_i is the score value of the variable from 1 to 5, w_i is the weighting attributed to every parameter. Fryer and Gray [62] declare that high FSI values indicate the presence in the plant of persistent and stable foams that can cover the entire aeration basin. Instead, low FSI values refer to foams that collapse quickly and occupy only a small part of the surface of the basins.

The equipment and the operative conditions of foams direct evaluation methods are summarized in Table 1. Figure 3 shows the pilot plant used by the researchers for aeration tests on foaming described above.

Table 1. Instrumentation and operative conditions of foam direct assessment methods.

Direct Methods	Equipment	Operative Conditions	References
Foam Rating (FR)	Glass tube (50 cm high, 4 cm diameter), air diffuser (porosity 40–90 μm) on the bottom	Sample volume: 50 mL, Airflow: 200 mL min^{-1}	[2,13,23]
Shaking	100 mL graduated cylinder	Sample volume: 14 mL, Stirring time: 10 s	[72]
Air bubbling	Quartz column (50 cm high, 3.5 cm diameter), 0.25 cm capillary on the bottom	Sample volume: 50 mL, Airflow: 158 mL min^{-1} for 2 min	[72]
Waring Blender	High speed mixer	Sample volume: 100 mL, Mixing speed >1000 r min^{-1} for 3 min	[73]
Foam Power (FP)	Vertical acrylic cylinder (100 cm height, 3 cm diameter, 7 cm^2 of cross section)	Sample volume: 100 mL, Airflow: 5 L min^{-1} for 20 s or 30 s	[22]
Foam Power 2 (FP2)	Glass tube (1 L, 6 cm diameter), borosilicate sintered disc (porosity 160–250 μm , 100–160 μm , 40–100 μm) on the bottom	Sample volume: 150 mL, Airflow: 0.5 L min^{-1} for 1 min	[20,62]
Foam Stability (FS)	Graduated cylinder, sintered glass diffuser on the bottom	Sample volume: 150 mL, Airflow: 0.5 L min^{-1} for 1 min	[22,62]
Scum Index (SI)	Flotation cell (50 cm height, 8 cm internal diameter)	Sample volume: 2L, Airflow: 10 L (L h) $^{-1}$ for 15 min	[2,21,78]

4.1.7. Methods Applicable on Site

Two methods of direct measurement of foams applied directly on-site are described below. The results of these procedures must be interpreted and evaluated with caution, attention and critical spirit since above all the apparent degree of foams produced is probably influenced by the layout of the plant, the configuration of the process equipment and the operating parameters (for example, entrapment and accumulation of foams in certain areas of the reactors) [21,62].

Foam Surface Covered (FSC)

Foam surface covered (FSC) is a percentage index of the surface of the aeration basin covered by foams. To achieve this, the aerobic reactor is photographed daily with an instrument orthogonal to the surface, connected to a digital image processing system (CAD Software). The areas covered by the foams are thus digitized [23]. This method was also previously applied by Torregrossa et al. [78]: it was possible to evaluate the foam-covered surface of aeration tank because they were equipped with mechanical aerators, although with an approximated method proposed by Kocianova et al. [79].

In some cases, there was an excellent correlation between the foam coverage and the foaming potential. However, it is preferable to use the result on the foam coverage together with other methods for assessing the extent of foaming in the aeration basins [62,78].

Foam Volume (FV)

The second method examined is the foam volume (FV). In order to quantify the foam volume, its thickness must be determined as the arithmetic mean of ten measurements taken at different points of the aeration basin operating on a pre-established horizontal grid [23]. Note the percentage of the covered area, it is possible to estimate the total apparent volume: in the study of Torregrossa et al. [78], the foam thickness was measured with a hydrometer and the estimation of the total apparent foam volume has been expressed as foam volume ratio parameter, defined as the per-thousand part of the total volume of the aerobic reactor. Fryer and Gray [62] applied a new method based on the observation of the difference in conductivity to identify the interface between the foam layer (with lower conductivity) and the underlying sludge (high conductivity). Measurements were performed using a WTW LF91 conductivity meter.

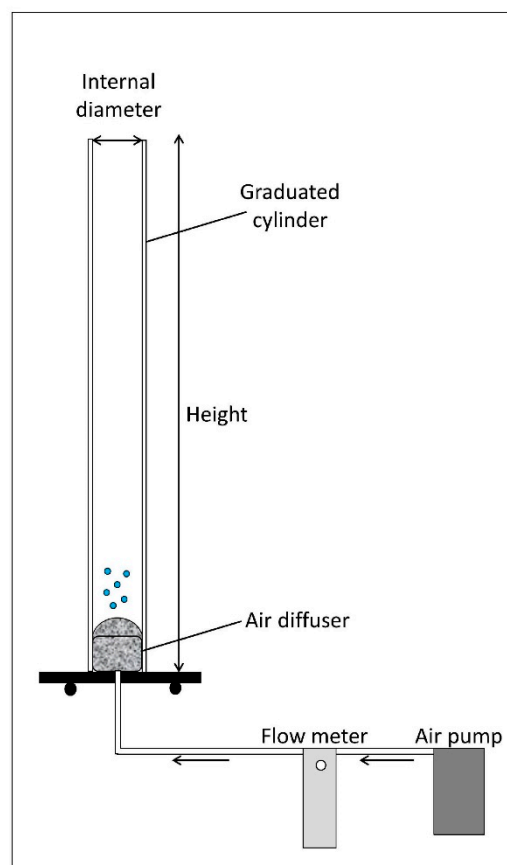


Figure 3. Pilot plant for aeration tests.

4.2. Indirect Methods

4.2.1. Surface Tension

One parameter that could potentially be used to evaluate the potential foaming is the surface tension, which can be quantified with the application of the Wilhelmy plate method [12,13,72]. The procedure consists of the measurement of the surface tension overtime, during immersion and lifting of a paper plate into and out of solution using a tensiometer and the sample must be solids free [15]. In a solution, different compounds tend to change the surface tension of the solution due

to their molecular weight and/or physical–chemical properties: in particular, the surfactants tend to lower the surface tension of a solution, increasing the surface activity, which facilitates the initiation of foaming [30]. The Wilhelmy plate method has been applied to monitor and demonstrate the production of biosurfactants by foam foaming bacteria observed as a lowering in surface tension [62]. Using this technique, Blackall et al. [13] assessed the effects on the surface tension of the temperature and concentration of the SS, by diluting the sample. The effects of the presence of additives on the surface tension of the liquid phase obtained by filtering the mixed liquor on 0.3 μm membranes were also monitored. As additives both *Nocardia amarae* cells and synthetic surfactants Tween 40, Tween 60, Teric 402, Teric 405, oleic acid and cooking oil have been tested. With this technique, it was possible to correlate the formation of foams with low surface tension values, both by working with chemical surfactants and with pure cultures. However, Blackall et al. [13] stated that this method is not suitable for evaluations in activated sludge plants where, in the face of high surface tension, close to that of water (approximately 72 mN m^{-1} at 20°C [30,80]), foams may still occur; therefore, surface tension values could not be correlated with foaming at the plant.

Heard et al. [12] also applied the Wilhelmy plate method using an analyte surface tension meter (40 mL of filtrate in a 100-mm diameter petri dish) and investigated three strains of the filamentous bacterium *Gordonia amarae*, isolated from WWTPs, to determine their effect on foam formation and stabilization. The filtrate was obtained by filtering the whole culture through 0.45 μm glass fiber membrane filters. The results of the experiments showed that the foam persistence of the filtrate was inversely related to the surface tension: a surface tension less than 60 mN m^{-1} was required to generate stable foams, with or without bacteria. Inclusion of bacteria substantially increased the foam persistence but the presence of bacteria in a non-foaming sample was not enough to induce foaming.

The plate method with centrifugation was applied by Verma et al. and Nitschke and Pastore [81–83] for the determination of surface tension in sludge samples. However, to date, an adequate method that allows one to determine the surface tension in the AD sludge has not been developed because the main difficulty is related to the fact that the surface tension measurements can be performed only in samples without solids [30]. The data obtained in the work of Ganidi et al. [30] allowed one to study the effect of proteins and carbohydrates and dissolved organic substances on the surface tension in sludge samples.

4.2.2. EPS Concentration

The abbreviation EPS indicates different classes of macromolecules such as polysaccharides, proteins, nucleic acids, lipids and other polymeric compounds that can be found in the intracellular zone of microorganisms, in particular near or outside the cell surface [84]. EPSs are produced by bacteria during the biological process as a result of stress conditions (e.g., nutrient deficiency and high sludge age) and make the cell walls of the filamentous microorganisms present more or less hydrophobic. Therefore, EPSs take on a crucial role in the formation of biological foaming [85]. EPSs can be divided into EPS_b , which is the fraction contained inside the cell or attached to it, and EPS_s , which is the fraction secreted into the mixed liquor [2,21,23,86]. As suggested by [50] EPS_s can be considered as the generic SMP, therefore, the total EPSs are calculated in accordance with Equation (7):

$$\text{EPS} = \text{EPS}_b + \text{SMP} \quad (7)$$

Heating Method

The first method described to evaluate the concentration of EPS is the heating method, which was applied by Le-Clech et al., Zhang et al., Morgan et al. and Cosenza et al. [87–90] to extract EPSs from activated sludge. According to the procedure proposed by Capodici et al. [2], after concentrating the sample by centrifugation, the supernatant was filtered to obtain the SMP fraction. The residue was resuspended in ultra-pure, heated water. The separation of EPS_b by the heated sludge mixture was carried out by ultracentrifugation and following filtration. The total EPS concentration was calculated as the sum of EPS_b and SMP. The following quantification methods were then applied to the

aqueous phases: for both SMP and EPS_b analysis, the “Lowry’s method” [91] to determine the total concentration of proteins and the “Anthrone method” [92] to quantify carbohydrates. Therefore, EPS_b and SMP were calculated with Equations (8) and (9):

$$EPS_b = EPS_p + EPS_c \quad (8)$$

$$SMP = SMP_p + SMP_c \quad (9)$$

where:

EPS_p: protein fraction of bound extracellular polymeric substances;

EPS_c: carbohydrate fraction of bound extracellular polymeric substances;

SMP_p: protein fraction of soluble microbial product;

SMP_c: carbohydrate fraction of soluble microbial product.

Steaming Extraction Method

Nakajima and Mishima [22] proposed a steaming extraction method, which was a modification of that proposed by Brown and Lester [93]. In their work, Brown and Lester [93] stated that the steaming treatment was the most effective extraction method for the activated sludges since it released a significant quantity of extracellular polymers from the flocs and caused less cellular disruption than other treatments described by them. The procedure implemented by Nakajima and Mishima [22], in their experiments, is as follows: a volume of 5–10 mL of activated sludge sample was placed in a centrifuge tube and centrifuged at 3000 rpm for 10 min. The solid residue was suspended in 30 mL of distilled water. The centrifuge tube was placed in an autoclave at 105 °C for 30 min, then it was cooled, centrifuged at 14,000 rpm for 10 min and the supernatant was filtered. Protein and carbohydrate quantification in this EPS extract was carried out using the “Lowry’s method” and the “Anthrone method”, respectively.

India Ink Reverse Stain Method

Given the importance attributed to EPSs, the simplest method for detecting the excess presence in cases of viscous bulking is the India ink reverse stain method proposed by Jenkins et al. [5] and a similar procedure has also been implemented by Capodici et al. [2] ten years later. The procedure applied by Capodici et al. [2] in their experiments is as follows: prepare an aqueous suspension of mixed liquor and India ink (lampblack, or carbon black, mixed with water to form a liquid) in 4:1 ratio, pour 20 µL of this mixture in the centre of the slide, cover the slide with the square slide cover (2 cm × 2 cm), caring that the mixture is spread uniformly on the surface, quickly observe optical phase-contrast stereoscope at 10× magnification and acquire the digital image. When this ink is mixed with activated sludge, it penetrates the flakes, coloring them. If there are EPSs inside the flake, the areas occupied by it remain clear (not colored). The method was also considered able to estimate the abundance of EPSs in the activated sludge, using an optical phase-contrast microscope to measure the extent of non-colored areas, within a well-defined region. The post-processing of the digital image acquired through the microscope with a special digital acquisition system allows to evaluate the areas “including ink” (colored) or “not included” (not colored) and provides an estimate of the abundance of EPS in the sludge [2].

4.2.3. Hydrophobicity

The bacteria causing foaming in activated sludge plants are hydrophobic, and their hydrophobicity is assumed to be a crucial factor in their foam-forming ability. The method of cell surface hydrophobicity (CSH) involves partitioning the mixed liquor biomass in a hydrophobic organic solvent and the absorbance of the water layer is measured before and after treatment: the difference is used to calculate the hydrophobicity [62]. Therefore, there is a relationship between CSH, as determined by microbial adherence to hydrocarbons, and the ability of microorganisms isolated from activated sludge foams

to produce stable foams [71,94]. Capodici et al. [2], to evaluate the hydrophobicity of cell surfaces, estimated the microbial adherence to hydrocarbons (MATH) according to the modified Rosenberg's method, using n-hexadecane as the solvent [95]. Torregrossa et al. [78], applied the same method, extending the measure of hydrophobicity not only to the superficial cells of mixed liquor (CSHml) but also to those in foams (CSHf).

4.2.4. Viscosity

To assess the correlation between the foaming phenomenon and the change in sludge rheology, viscosity is a very important parameter as it is strongly correlated with the EPS concentration. Capodici et al. [2] and Di Bella et al. [23] measured sludge viscosity using a Brookfield digital viscometer (model DV-E) equipped with concentric cylinders and an adapter for low viscosity. The measurements were conducted at a constant temperature of 20 °C and two different viscosity data were used to describe the rheological characteristics of the activated sludge: the limit viscosity μ_{∞} , which describes the rheological condition of the sludge with a constant viscosity value reached after the rheofluidification phenomenon (less than the apparent initial viscosity after the first Newtonian plateau) and the apparent viscosity η_a depending on mixed liquor SS concentration as described by Pollice et al. [96,97].

4.2.5. Filamentous Bacteria

Another parameter that plays a key role in the formation of biological foams is the greater or lesser presence of filamentous bacteria. A method based on Gram or Neisser staining was developed and tested, aimed at the identification of dominant filamentous microorganisms, which can be carried out on a morphological basis (recognition of filamentous bacteria) using a phase-contrast microscope [2,21]. It is possible to identify the quantitative information about the presence of filaments of a certain genus or type through the normalized intersections number (NIN). Explained in detail by Di Bella and Torregrossa [21], a sample of mixed liquor (50 mL), after Gram or Neisser staining, was observed by an optical microscope following a default path and counting the number of intersections between the path and the filamentous bacteria. The average number of counted intersections, X_{AVG} , was determined by counting the number of intersections on five slides and averaging the five numbers. This average was related to the total mass of volatile suspended solids (VSS) in the sample ($50 \mu\text{L} \times \text{VSS concentration in the mixed liquor}$) amplifying it in proportion to the number of fields of view covered by the sample at 1000× magnification (equivalent to 220 of view fields). The following equation was used to calculate the NIN [21]:

$$NIN = \frac{X_{AVG} * 220}{VSS * 50} \quad (10)$$

5. Foam Control Methods

For the detection of foams contact and contactless sensors can be used. The contact sensors work as capacity or conductivity sensors, while the non-contact detectors are based on ultrasounds that detect the pressure variation in the reactor [80]. Once the presence of biological foams is identified, physical (e.g., mechanical), chemical and biological control mechanisms are activated. Specific and non-specific methods can be applied for foaming control: control strategies are preferable because they guarantee a permanent solution, on the contrary, non-specific methods (such as chlorine or hydrogen peroxide) offer only temporary and non-definitive solutions [98]. It is preferable to apply specific mechanisms because they are selective for the specific filamentous microorganism and do not damage other bacteria, while non-specific methods can be potentially harmful to all the biomass present in the reactor [99].

5.1. Physical Methods

Examples of specific approaches are classifying selectors (physical methods), and modifications of plant configuration or operating conditions [100]. The selector forms the initial part of a biological reactor and consists of a mechanism for selecting the activated sludge microorganisms against foam-former bacteria [101] and bench-scale anaerobic selector experiments were conducted by Pitt and Jenkins [6] to assess the ability of an anaerobic selector to control a *Nocardia* population. Since biological foaming is difficult to control in the intense development phase, the control strategy in its initial phase is important: a pre-alarm system should be established, useful for intervening on the formation of foams by blocking the proliferation of foam-former microorganisms from the beginning and avoiding the strong expansion [56]. Various methods have been developed to achieve mechanical foam control, including injectors, impellers and centrifuges. The most efficient mechanical systems, however, are too complicated, consume a lot of energy, and do not guarantee ease of use [80]. For the foaming control due to excessive growth of *Microthrix parvicella*, there are a series of effective strategies such as improving the concentration of dissolved oxygen (maintaining a concentration indicatively $> 2 \text{ mg L}^{-1}$ at all places in the aerobic zones), increasing the ratio between organic carbon and biomass or reducing sludge age or methods of chemical precipitation and chlorination at the moment of maximum growth of filamentous bacteria [5,56,99]. Noutsopoulos et al. [102], with bench-scale continuous-flow experiments, has shown that the reduction of sludge age to block the growth of *Microthrix parvicella* is an effective method, but this solution can no longer be used in those CAS processes where it must also take place nitrification, as it can lead to the elimination of nitrifying bacteria.

5.2. Chemical Methods

The chemical control of the foams takes place through the dosage of antifoaming agents, to which a series of advantages (simplicity, ease of use, improving flocculation with the use of coagulants, removing of filamentous microorganisms that are embedded in the flakes) and disadvantages (reduced mass transfer rate, inhibition both nitrification and organic matter removal, floc break-up due to the high chlorine doses, toxicity and production of additional chemical sludge) are associated [56,98,103]. Chlorination is one of the most popular applications for the rapid and effective control of activated sludge foaming [98], but Mamais et al. [103] have also shown the efficiency of some coagulants (such as ferric chloride, ferrous chloride, polyaluminium chloride, hydrated aluminum sulphate and cationic polymer) for the control of foaming in bench-scale batch experiments. The addition of polyaluminium chloride (e.g., PAX-14) is an effective method of controlling *Microthrix parvicella* in WWTPs and Rossetti et al. [99] stated that it seems to be ineffective against other filamentous bacteria. Mamais et al. [103], in their work, have instead shown that both *Microthrix parvicella* and *Gordonia amarae* filamentous organisms were embedded inside the floc material, making access to particulate and colloidal substrates more difficult due to increased diffusional resistance. These chemical treatments can also be very expensive and, more importantly, they are only a temporary solution since the foaming will resume when the addition of the chemicals is stopped [104]. Another principal disadvantage of chlorination is the production of undesirable disinfection by-products, such as the significant formation of trihalomethanes (THMs) in the effluent [100,105,106].

Although it is indisputable that the chemical methods are those mainly used, it is also interesting to apply biological methods for the control of foams, that is, the use of specific bacteriophages that act only on filamentous bacteria: even if their impact on the sludge structure active is still largely unknown, the characterization of the genome of various filamentous bacteriophages is made available by Petrovski et al. [107–109] studies.

6. Discussion and Future Outlooks

The discomfort generated by foaming in the WWTPs represents a very common problem, but to date, it is treated mainly only at the management level and still too little studied through a globally

shared scientific method. In fact, years of research have not been able to completely clarify the key processes underlying this phenomenon. The complexity of the issue, combined with inaccurate measurement methods, inaccurate definitions of terms and confusion of phenomena have led to numerous partially contradictory descriptions and hypotheses [16]. To date, the most used procedure for assessing foaming in wastewater treatments is the air method, used above all to define the foaming potential and foam stability. As stated by Fryer and Gray [62], the principal advantage of the air method is that it is relatively easy to carry out and produces comparative results in a short period of time. However, FP indices give only information on how much foams are produced in terms of height or volume but not on consistency and stability of foams. Therefore, FPs indices should not be used individually but integrate with other values to better define the tendency for foaming of wastewaters. It is not easy to obtain a complete, exhaustive and subjective characterization of a foaming phenomenon. Firstly, apply a method for the evaluation of the foaming potential that allows obtaining careful and constant results on equal conditions is necessary. However, this is not enough. Other tests must be used to further deepen the assessment, such as the stability of the foams, but not only. The ideal goal is to define a mathematical model based on easily and directly measurable on-site parameters. The variables of the model could be the airflow rate, the aeration time and the initial height of the liquid before aeration: their monitoring is not complicated for the operators in WWTPs. Through the model, significant and simple to interpret data/results should be obtained. For example, the parameter resulting from the model could be the percentage increase in the height of the mixed liquor in the aeration basin following the formation of foams. It is good to avoid subjective procedures with little correlation between the parameters considered and hardly applicable on-site by the operators. It is strongly suggested to develop and validate mathematical models that allow evaluating the real weights of the operative parameters on which the formation of foams depends, as different degrees of dependence on the variables analyzed in the model could result.

Regarding the foams control mechanism, the mechanical control methods are preferable to overcome the disadvantages associated with antifoam chemical agents. However, mechanical devices alone are generally not totally effective, and their action is enhanced by simultaneously using chemical antifoam agents at the lowest possible concentration. Although foam control systems are widely used, when possible, it is preferable to adopt management interventions preventive of foam formation in the reactor [80]. The goal shared by the scientific community must be to clarify the phenomenon as much as possible in order to identify future research directions: (i) develop a shared protocol for the monitoring of foaming phenomena in WWTPs through evaluation procedures and (ii) indicate promising strategies to prevent or control the formation of foams in WWTPs especially of easy applicability from a management and operative point of view. In fact, to date, there is not a standardized assessment and control method available.

7. Conclusions

Although foaming originally was associated with CAS plants, in recent years, it has also been studied more thoroughly in MBRs processes and in ADs. Years of research have not been able to completely clarify the processes that underlie this phenomenon because the causes of such foaming events are often unclear. The aim shared by the scientific community must be to lead to a better understanding of the mechanisms involved in stable foams production in order to identify common hypotheses. Moreover, the evaluation methods proposed by the scientific community and the potential control measures possibly applicable in those WWTPs subject to serious problematic foaming phenomena are described. It is important and useful to deepen the studies on foaming, clarifying the causes and effects that affect the phenomenon: above all (i) to identify a common and shared method of evaluation of the foams and as objective and stable as possible, (ii) to simplify the control mechanisms from a management point of view and earlier (iii) to predict and subsequently apply preventative measures directly in WWTPs.

Author Contributions: Conceptualization, M.C.C., M.B. and M.C.M.; Methodology, M.C.C., M.B., A.A., E.C.R. and V.T.; Writing—Original Draft Preparation, F.M.C. and M.C.M.; Writing—Review and Editing, M.C.C., A.A., E.C.R. and V.T.; Visualization and supervision, M.C.C. and M.B. All authors have read and approved the final manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

Nomenclature

AD	Anaerobic digester
CAS	Conventional activated sludge
CSH	Cell surface hydrophobicity
EPS	Extracellular polymeric substances
EPS _b	Bound extracellular polymeric substances
EPS _s	Soluble extracellular polymeric substances
EPS _p	Protein fraction of bound extracellular polymeric substances
EPS _c	Carbohydrate fraction of bound extracellular polymeric substances
FP	Foaming power index calculated as consumed sample per liter of supplied air
FP ₂	Foaming power index calculates as foam volume produced per liter of supplied air
FR	Foam rating
FS	Foam stability
FSI	Foaming scum index
FV	Foam volume
FSC	Foam surface covered
MBR	Membrane bioreactor
NIN	Normalized intersections number
SDS	Sodium dodecyl sulphate
SEM	Structural equation modeling
SI	Scum index
SMP	Soluble microbial product
SMP _p	Protein fraction of soluble microbial product
SMP _c	Carbohydrate fraction of soluble microbial product
SS	Suspended solids
THM	Trihalomethane
VSS	Volatile suspended solids
WWTP	Wastewater treatment plant

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