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Anaerobic membrane reactor with phase separation for the treatment of cheese whey

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Abstract

Two-phase anaerobic digestion of cheese whey was investigated in a system consisting of a stirred acidogenic reactor followed by a stirred methanogenic reactor, the latter being coupled to a membrane filtration system to enable removal of soluble effluent whilst retaining solids. The acidogenic reactor was operated at a hydraulic retention time (HRT) of one day, giving maximum acidification of 52.25% with up to 5 g/l volatile fatty acids, of which 63.7% was acetic acid and 24.7% was propionic acid. The methanogenic reactor received an organic load up to 19.78 g COD/l d, corresponding to a HRT of 4 days, at which 79% CODs and 83% BOD_5 removal efficiencies were obtained. Average removals of COD, BOD_5 and TSS in the two-phase anaerobic digestion process were 98.5%, 99% and 100%, respectively. The daily biogas production exceeded 10 times reactor volume and biogas methane content was greater than 70%.

Keywords: Cheese whey; Acidogenesis; Methanogenesis; Cross-flow microfiltration

1. Introduction

Whey is a by-product of the dairy industry in which the principal components are lactose, proteins and mineral salts (Vasala et al., 2005). Approximately 47% of the 115 million tons of whey produced world-wide every year are disposed of in the environment (Leite et al., 2000; Zhou and Kosaric, 1993; Siso, 1996). This represents a significant loss of resources and causes serious pollution problems since whey is a high strength organic pollutant with high BOD_5 and COD, with values of 40,000–60,000 mg/l and 50,000–80,000 mg/l, respectively (Ben-Hassan and Ghaly, 1994; Fournier et al., 1993). More than 90% of whey BOD_5 is due to lactose (Kisaalita et al., 1990).

Currently, the whey production in Tunisia is estimated at 35,000 tonnes/year. During the last few decades, this production has increased very rapidly with the development of the dairy industry. Thus, the problem of whey disposal will worsen. Indeed, the continuous discharge of whey onto land can endanger the chemical and physical structure of the soil, reduce crop yields and lead to serious groundwater pollution problems (Ben-Hassan and Ghaly, 1994).

For medium size cheese factories, that have growing disposal problems and cannot afford high investment costs for whey valorisation technologies (such as whey protein and lactose recovery, spray drying, etc.), physico-chemical and/or biological treatment of this effluent is imperative. Due to the high organic content of whey, the basic biological treatment process to be used can only be anaerobic digestion, whereas regular treatment processes such as the

Abbreviations: AD, anaerobic digestion; AR, acidogenic reactor; BOD_5, biochemical oxygen demand; COD, chemical oxygen demand; CODs, soluble chemical oxygen demand; CSTRs, continuous stirred tank reactors; CSMR, continuous stirred methanogenic reactor; HRT, hydraulic retention time; TMP, trans-membrane pressure; TN, total nitrogen; TP, total phosphorous; TS, total solids; TSS, total suspended solids; VCR, volumetric concentration factor; VFAs, volatile fatty acids; VS, volatile solids; VSS, volatile suspended solids; MR, methanogenic reactor; CFM, cross-flow microfiltration; MS, mineral solids.

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activated sludge process are completely inappropriate (Gavala et al., 1999).

Anaerobic digestion of cheese whey offers an excellent solution in terms of both energy saving and pollution control (Ergüder et al., 2001). The major advantages of this process are low cost, high energy efficiency and process simplicity compared to other waste treatment methods.

However, despite these advantages, anaerobic digestion is not widespread in the dairy industry, largely due to the problems of slow reaction, which requires longer HRT, and poor process stability, especially for effluents rich in components that are subject to rapid acidification. Indeed, Malaspina et al. (1996) stated that raw whey is a quite problematic substrate to treat anaerobically because of very low bicarbonate alkalinity (50 meq l^{-1}), high COD concentration (70 g COD l^{-1}) and a tendency to acidify very rapidly.

The idea of developing anaerobic digestion as a two-phase process originated from the view that it is generally a process involving two different sets of activities. Overall, the two-phase process takes advantage of the phase separation phenomenon, deriving naturally from different kinetic rates. This provides separate acidogenic and methanogenic reactors to decrease the cost, and to improve treatment efficiency, energy production and process stability of anaerobic systems (Ke and Shi, 2005).

Anaerobic digesters are widely used for treatment of agro-industry by-product wastewaters. These digesters are single pass reactors with no selective solids recycle. This limits the organic loading rates and operating biomass concentrations (Pillay et al., 1994). One way to overcome these problems is to include a membrane to enable independent control of hydraulic and solid retention times (Dhouib et al., 2003). Indeed, in recent years, considerable attention has been focused on development of a novel anaerobic process in which a membrane separation unit is incorporated in place of a settlement system. So far, several investigators have studied anaerobic-membrane processes for treatment of wastewaters such as wine distillery effluents (Ross et al., 1990), palm oil mill effluent (Fakhru'l-Razi and Noor, 1999) and dairy wastes (Li et al., 1985).

This study examined the feasibility of applying an anaerobic membrane bioreactor with phase separation (acidogenesis/ methanogenesis) to treat cheese whey.

2. Methods

2.1. Whey

The whey used in this study was obtained from the "Tunisian Cheese Factory" (Sfax, Tunisia) which used traditional technologies for cheese manufacture. The whey samples were drained directly from the cheese vats, collected in 20 l tanks and transported to the laboratory freezer and stored there at a temperature of −20 °C to avoid the acidification and the chemical composition modification of cheese whey. About one week before it was needed, a proportion of the frozen whey was moved into a cold room at 4 °C for defrosting. During the adaptation phase diluted whey at pH 6.5 was fed into the reactor.

Table 1
Chemical characteristics of raw cheese whey

Characteristics	Sample
COD (g/l)	68.6 ± 3.3
BOD_5 (g/l)	37.71 ± 2.84
COD/BOD_5	1.83 ± 0.05
TSS (g/l)	1.35 ± 0.06
Lactose (g/l)	45.9 ± 0.88
Proteins (g/l)	2.71 ± 0.05
TS (%)	5.93 ± 0.38
VS (%)	5.61 ± 0.36
MS (%)	$0.31 \pm 6.3 \times 10^{-4}$
Fat (g/l)	9.439 ± 1.14
pH	4.9 ± 0.27
TKN (g/l)	1.12 ± 0.01
TP (g/l)	$0.5 \pm 1.8 \times 10^{-3}$

The chemical composition of cheese whey is shown in Table 1. The notable characteristics of this effluent are the high COD and especially BOD_5 values. Indeed, more than 90% of whey BOD_5 is due to lactose (Kisaalita et al., 1990).

2.2. Experimental apparatus

The experimental set-up used in this study is shown schematically in Fig. 1. It consisted of a continuously stirred reactor used as an acidogenic reactor and a continuously stirred reactor coupled to a membrane module used as a methanogenic reactor.

The seed sludge for both reactors was obtained from a full-scale anaerobic wastewater treatment plant.

2.3. Acidogenesis

The acidogenic phase was carried out in a 7 l stirred reactor (diameter 16.4 cm; height 33 cm), with 5 l working volume. The acidogenic reactor was kept at room temperature (37 ± 2 °C). Agitation was provided by a magnetic stirrer. The pH of the feed was regulated at the beginning of the tests at 6.5.

2.4. Methanogenesis

The methanogenic phase was carried out in a 20 l (diameter 22 cm; height 52 cm) Biolafite thermostatic reactor with a working volume of 15 l and a stirring speed of 200 rpm. The temperature of the reactor was maintained constant at 37 °C by circulating water through the thermostatic column in the reactor.

Solids (anaerobic sludge) separation prior to recycling was achieved by gravity settlement using a conventional decanter during the first 25 days. After this period, a membrane module was attached to the methanogenic reactor and the retentate was recycled into the reactor.

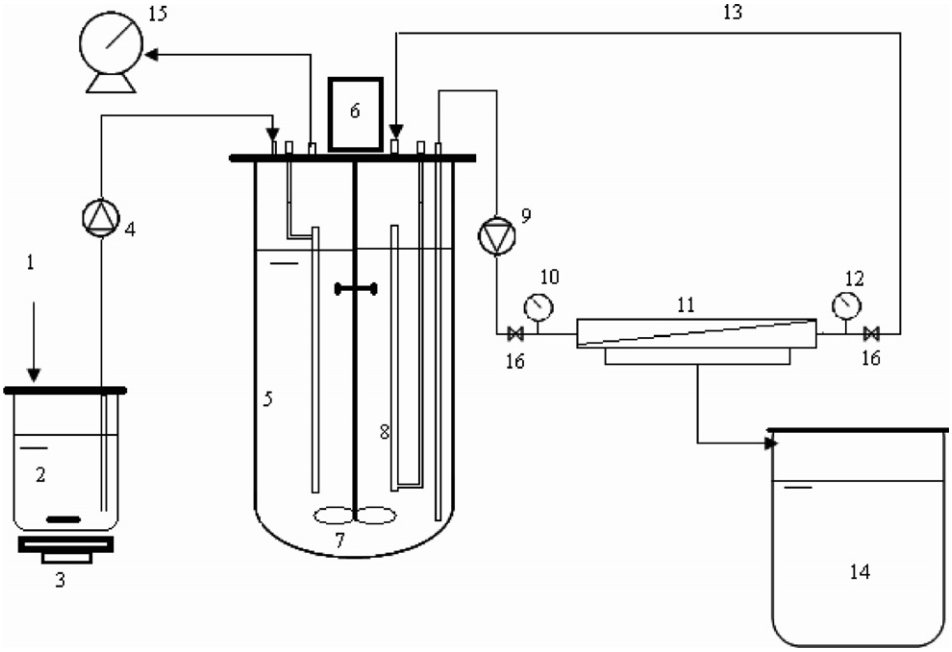

Fig. 1. Schematic diagram of laboratory-scale two-phase anaerobic digestion system. 1. Raw cheese whey, 2. acidogenic reactor, 3. magnetic stirrer, 4. peristaltic pump, 5. methanogenic reactor, 6. moto-regulator with variable speed, 7. agitation propeller, 8. thermostatic column, 9. peristaltic pump, 10. manometer, 11. microfiltration membrane, 12. manometer, 13. biomass recycling, 14. permeate tank, 15. gas flow meter, 16. valves.

2.5. Microfiltration (MF) membrane

A lab-scale microfiltration (MF) system comprising a membrane module (Membralox, Tarbe, France) was used to separate sludge solids from permeate. The ceramic membrane (α-alumin) was 0.4 m^2 in area and had a 0.2 μm cut-off. The cross-flow velocity was fixed at a value of 5 m/s and the trans-membrane pressure was varied from 1.25 to 1.75 to 2.25 bar. Pressure changes were adjusted by the use of valves before and after the microfiltration membrane. Membrane permeates were measured by a handy measure of the flow rate without using a flow meter.

2.6. Analytical methods

COD was estimated using the method described by Knechtel (1978). The percentage of acidified COD was estimated following Hajipakkos (1987) relationship:

$$\text{Percentage of acidified COD}(\%) = \frac{\text{COD of VFA}(\text{mg/l})}{\text{Soluble COD}(\text{mg/l})} \times 100$$

BOD_5 was determined by the manometric method with a respirometer (BSB-Controller model 620 T (WTW)).

Total soluble proteins were determined according to the Bradford method (1976).

TP was determined by the Dabin (1967) method. TKN was determined by the Kjeldahl (1883) method.

Soxhlet solvent extraction was used to determine the fat content of cheese whey. A mass (m) of the total solids (TS) of a volume (V) of cheese whey was placed in a cavity that was gradually filled with liquid hexane by condensation of vapors from a distillation flask (m_0 is the mass of the empty flask). When the liquid reached a preset level, a siphon pulled the contents of the cavity back into the distillation flask, thus carrying the extracted fat into the bulk liquid. This procedure was repeated for 2 h to achieve complete extraction. After that, the hexane was evaporated under vacuum using a rotary evaporator (Rotavap). The flask was dried at 105 °C for 24 h then weighed (m_1).

Fat content = $(m_1 - m_0)/m$ (mg Fat/g TS) and then fat concentration was calculated (g Fat/l cheese whey).

TS, VS, TSS and VSS were determined according to Standard Methods (APHA, 1992).

To get the gas composition, gas samples were taken with a syringe from the tank of biogas and analysed with a gas chromatograph (DELSI Model: IGC11) equipped with a thermal conductivity detector and a concentric Alltech CTRI column. Column temperature was 60 °C. Helium was used as the carrier gas at a flow rate of 35 ml/min. Volatile fatty acids (VFA) were analysed by a gas chromatograph (SHIMADZU GC-9A) equipped with a flame ionisation detector (SHIMADZU CR 6A). A Nukol capillary silica column (30 m × 0.32 mm) was used (Mechichi and Sayadi, 2005).

The measurement of turbidity was based on comparison of the intensity of light scattered by the effluent compared to the light scattered by a reference suspension under the same conditions. The turbidity was determined using a turbidimeter (WTW, turb 551 IR).

The conductivity and the pH were determined using a conductivimeter, model CONSORT C 831, and a pH meter, model Metrohm 744.

3. Results and discussion

3.1. Acidogenesis stage

The stirred tank acidogenic reactor was operated at a HRT of one day. The pH of raw cheese whey was regulated at the beginning of the tests at an average value of 6.5 (Fig. 2); this is the optimal pH range for acidogenic metabolism (Kisaalita et al., 1987). Anaerobic digestion is a complex process consisting of a series of microbial transformations of organic materials to methane and VFAs such as acetate, propionate, butyrate, *iso*-butyrate, valerate and *iso*-valerate. These VFAs have long been recognized as the most important intermediates in the anaerobic process and have been proposed as a control parameter (Mechichi and Sayadi, 2005). Therefore, changes in VFA concentration can be in response to variations in temperature, organic loading rates or levels of toxicants.

The VFA concentrations in the crude whey were less than 1 g/l with about 0.4 g/l of acetic acid. The aim of this part of the study was to obtain maximum acidification. The method used for the assessment of acidification using the COD equivalent of each VFA was adapted from Hajipakkos (1987). The COD equivalents of the four VFAs are presented in Table 2.

At an HRT of 24 h, the pH decreased considerably with increasing VFA concentration. The maximum acidification was 52.25%. VFA concentration in the acidified effluent was up to 5 g/l, with acetic and propionic acids the main products of this treatment phase. Valeric, butyric and *iso*-butyric acids were also present, but in substantially lower quantities (Fig. 3). Acetic acid and propionic acid made up on average 63.7% and 24.7% of the total VFAs, respectively (Fig. 3(b)).

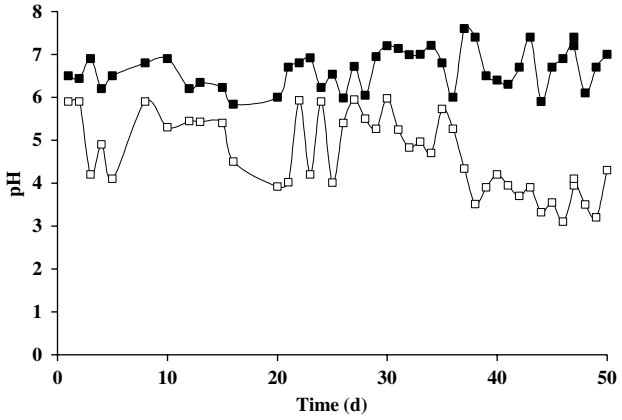

Fig. 2. pH values observed during the acidification of cheese whey: (■) raw cheese whey pH; (□) acidified cheese whey pH.

Table 2
COD equivalents of volatile fatty acids

VFA	COD equivalent
Acetic acid	1.066
Propionic acid	1.512
Butyric acid	1.816
Valeric acid	2.036

During whey fermentation most of the lactose is transformed into lactic acid, acetic acid and other VFAs. The majority of lactose (62%) was converted into VFA (5 g/l) and lactic acid (18 g/l) (Fig. 3(c)). Profiles of influent and effluent COD of the acidogenic reactor are shown in Fig. 3(d). COD removal during this step of bioconversion was 3–9 g/l, corresponding to a COD removal of about 18% (Fig. 3(d)). In fact, the acidogenic phase is characterised by its low pH, high VFAs and low COD reduction (Yan et al., 1992). The composition of gas from the acidogenic reactor was 35% CO_2, 65% N_2 and 0% methane.

Yilmazer and Yenigün (1999) have reported that with an HRT of 24 h, the maximum acidification was about 50%. Acetic acid comprised 52% of the total VFA produced, while propionic and butyric acids were 14% and 27%, respectively. More recently, Yu and Fang (2001) used an up-flow reactor to treat dairy wastewater, and they reported that, at pH 6.5, acetate, propionate and butyrate represented 79% of the total VFA.

3.2. Methanogenesis stage

In order to investigate the performance of the methanogenic phase of cheese whey treatment, effluent from the acidogenic reactor operating at an HRT of one day was fed to the continuously stirred methanogenic reactor (CSMR). The loading rate in the methanogenic reactor ranged from 3 to 19.78 g COD/ld (Fig. 4(a) and (b)). The effect of influent COD concentration and organic loading rate was examined under the operating conditions of HRT 4 days and SRT between 29.7 and 78.6 days.

Due to the separation between the acidogenic and the methanogenic steps, VFA production was not significant in the methanogenic reactor. The VFA concentration was less than 1 g/l. So VFA concentrations were always below the inhibitory limits, permitting the methanogenic process to be established progressively. The pH of the methanogenic effluent ranged from 7.29 to 8.51. The methanogenic reactor yielded an average COD removal rate of 79% (Fig. 4(b)). Also, the BOD_5 removal rate reached 83% (data not shown). Thus, in this study, the quality of the effluent after anaerobic digestion would not allow its direct disposal to the environment. Further treatment would be needed to meet the standard required.

Studies on anaerobic digestion of cheese whey are generally conducted with a single-phase digestion system (Bradford et al., 1986; Yan et al., 1989; Schroder and de Haast, 1989; Malaspina et al., 1995). Patel et al. (1995) investigated anaerobic digestion of high strength cheese whey with COD of 70,000 mg/l, using an upflow fixed film reactor with various support media, obtaining a maximum COD removal of 81%. A high COD removal of 98% was

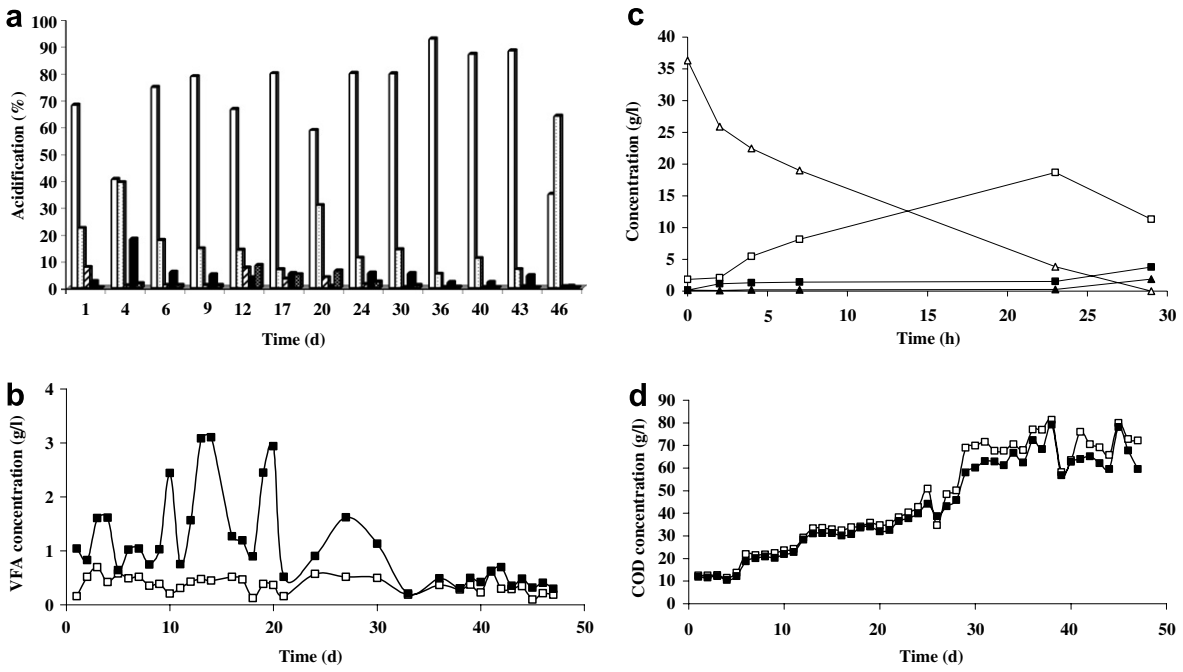

Fig. 3. (a) Rate of acidification of cheese whey expressed as percentage of acetic acid, propionic acid, *iso*-butyric acid, butyric acid and valeric acid produced. □: Acetic acid, ⬚: propionic acid, ▨: *iso*-butyric acid, ■: butyric acid, ▩: valeric acid. (b) Total volatile fatty acids in raw (■) and acidified (□) cheese whey. (c) Lactose, lactic acid, acetic acid and total volatile fatty acid concentrations in the acidogenic reactor. (△): lactose, (■): total volatile fatty acids, (□): lactic acid, (▲): acetic acid. (d) COD concentrations in the feed and in the effluent of the acidogenic reactor. (□): Feed COD concentration, (■): Effluent COD concentration.

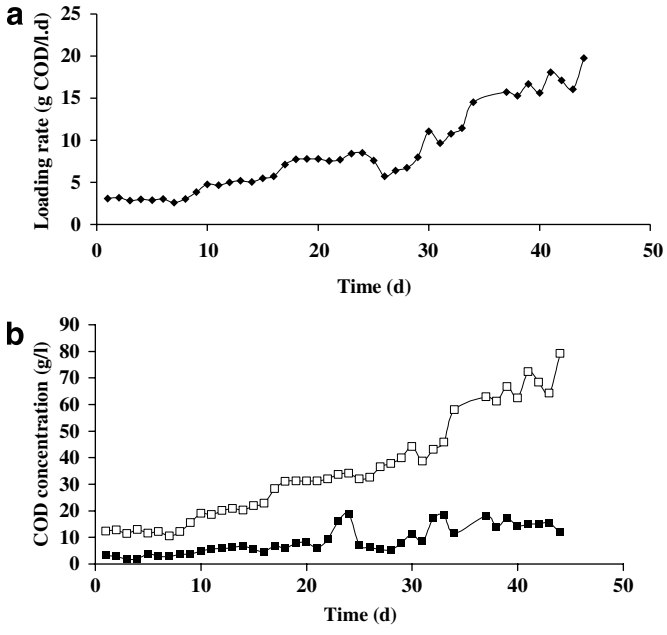

Fig. 4. (a) Organic loading rate introduced into the methanogenic reactor. (b) COD concentrations before and after methanogenesis. (□): raw whey, (■): effluent.

reported by Malaspina et al. (1996) using a downflow–upflow hybrid reactor. More recently, Yilmazer and Yenigün (1999) obtained high COD removal, over 90%, in an up-flow anaerobic filter operated at a HRT of 4 days with a maximum biogas yield of 0.55 m^3/kg COD removed.

3.3. Methanogenesis stage coupled to cross-flow microfiltration

Membrane separation is an effective method to achieve complete separation of solids from the effluent and this method allows operation at high sludge ages.

The tests were carried out under optimal conditions for treated whey microfiltration at a cross-flow velocity of 5 m/s, a VCF of 1 and a biomass concentration of 8.5 g/l (Dhouib et al., 2003). The main parameter varied for optimisation was TMP. The results of these tests are shown in Fig. 5. Permeate flux increased with increasing TMP from 1.25 to 1.75 bar. A plateau with 1.75 bar pressure was

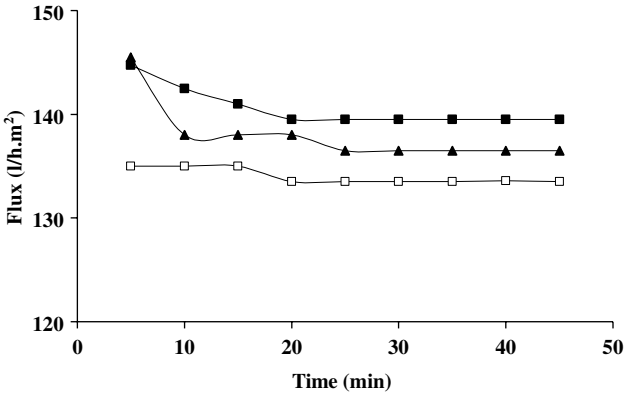

Fig. 5. Flux variation at TMP = 1.25, 1.75 and 2.25 bar. (□): TMP = 1.25 bar, (■): 1.75 bar, (▲): 2.25 bar.

obtained after 20 min at a flux of 139.5 l/h m^2. Increasing the TMP to 2.25 bar caused a decline of the flux to 136.5 l/h m^2. The flux decline can be explained by membrane fouling caused by the formation and compaction of a cake layer on the membrane surface (Hassairi et al., 2001) or by partial breakdown of the cake layer and continuous infiltration of the particulate matter inside the porous membrane. To conclude, the results of the optimisation of the treated whey cross-flow microfiltration show that the optimal TMP was 1.75 bar which resulted in a flux of 139.5 l/h m^2 (Fig. 5).

The reactor performance during a period of 45 days continuous operation is given in Fig. 6(a)–(d). The methanisation phase coupled to the cross-flow microfiltration improved the organic content and the turbidity reduction of the effluent (14.5 NTU). The average COD removal in the whole process was 98.5% throughout the whole experimental period (Fig. 6(a)). The BOD_5 of the raw cheese whey fed in the first step (acidogenic reactor) was between 5 and 35 g/l and less than 0.1 g/l after integrated methanisation-microfiltration, so that the average BOD_5 removal was 99.2% for the last ten days of the experiment. Permeate quality indicated that the removal of suspended solids was 100%. The data clearly indicated that anaerobic treatment coupled to cross-flow microfiltration of cheese whey was successful, the quality of the permeate effluent being acceptable for disposal via public drains. Indeed, in the Tunisian wastewater standards the COD, BOD and SS concentrations are 90, 30 and 90 mg/l, respectively (Tunisian Standard, 1989).

The calculated methane yield expressed as the volume of methane produced per g of COD removed is presented in Fig. 6(b). The average was 0.3 l CH_4/g $COD_{removed}$. Fig. 6(c) shows that the biogas productivity increased with increasing loading rate. In addition, combining the membrane system with the methanogenic reactor for the acidified cheese whey treatment led to an improvement in the daily biogas production, which exceeded 10 times the reactor volume at HRT of 4 days. The biogas production increased steadily with the increase in organic loading rate. The methane content was more than 70%. This proportion of methane is considered indicative of good performance (Strohwald and Ross, 1992).

As can be seen in Fig. 6(d), during the first 25 days where the biomass was recycled after decantation, the biomass concentration was constant at a value of 6.4 g VSS/l, corresponding to a SRT of 31.6 days. The combination of the methanogenic reactor with membrane microfiltration allowed complete retention of the microorganisms within the system. This led to an increase of the VSS from 6.4 to 10 g/l.

4. Conclusions

The study of two-stage anaerobic digestion of cheese whey was undertaken. During the acidogenesis step, a maximum acidification of 52.25% was achieved in the completely mixed acidification reactor at an HRT of 24 h. COD removal was low. The AR did not produce methane and achieved bioconversion only.

During the methanogenesis step, about 79% COD removal was achieved. The optimisation of cross-flow microfiltration at a VCF = 1 gave a TMP and permeate flux of about 1.75 bar and 139.5 l/h m^2, respectively.

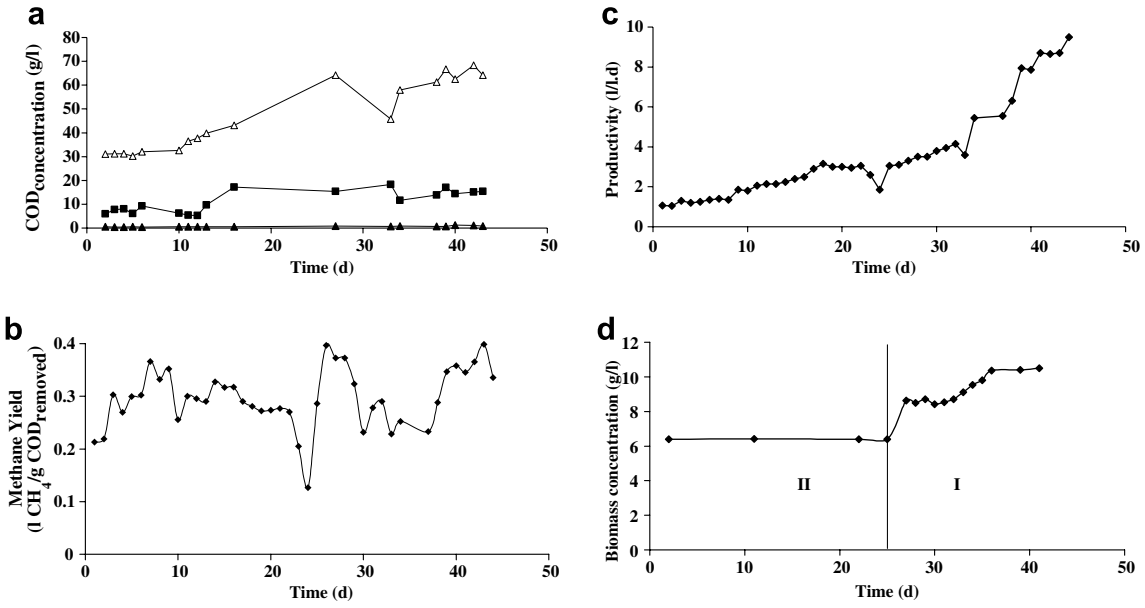

Fig. 6. (a) COD concentrations in raw whey (△), in the effluent leaving the methanogenic reactor (■) and in the permeate leaving the membrane module (▲). (b) Variation of the methane yield (l CH_4/g $COD_{removed}$) during the methanisation of the acidified cheese whey. (c) Biogas productivity in the methanogenic reactor coupled to cross-flow microfiltration. (d) Variation of biomass concentration in the methanogenic reactor. (I) Biomass recycled after decantation. (II) Biomass recycled after microfiltration ($V = 5$ m/s and TMP = 1.75 bar).

In the methanogenic reactor coupled to cross-flow microfiltration, the COD removal was about 98.5% throughout the whole experimental period. The BOD_5 removal was 99.2%. The daily biogas production exceeded 10 times the volume of the reactor at HRT of 4 days. The methane yield was up to 0.3 l CH_4/g COD removed. The membrane-anaerobic process consistently removed about 98.5%, 99% and 100% of the COD, BOD and TSS, respectively.

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