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PhD THESIS TITLE

**NOVEL LAGER BEER CLARIFICATION
AND STABILISATION PROCESS
USING CERAMIC TUBULAR
MICROFILTRATION MEMBRANES**

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ABSTRACT

Novel Lager Beer Clarification and Stabilisation Process Using Ceramic Tubular Microfiltration Membranes

This PhD thesis dealt with the development of a novel combined system for lager beer clarification and stabilization using ceramic tubular microfiltration membranes to avoid the safety and environmental problems associated with the use and disposal of conventional filter-aids and to minimise the environmental impact of the current industrial beer downstream processing. The optimal operating conditions for this system were assessed on a bench-top plant, appropriately designed. The data obtained allowed the determination of the predominant fouling mechanism and its reduction through appropriate centrifugal and enzymatic pretreatments on rough beer. The assessment of the operating cost of this novel procedure, as well as its environmental impact on climate change, was finally carried out by referring to an industrial brewery capacity.

Key words: CO₂ backflushing; Carbon Footprint; ceramic tubular membranes; clarification; fouling mechanisms; lager beer; microfiltration; optimal operating conditions; permeation flux; stabilization.

RIASSUNTO

Nuovo processo per chiarificare e stabilizzare la birra lager tramite membrane ceramiche tubolari di microfiltrazione

Questa tesi di dottorato ha riguardato lo sviluppo di un nuovo sistema per chiarificare e stabilizzare la birra lager, che prevede l'impiego di membrane tubolari ceramiche sia per evitare i problemi ambientali e di sicurezza associati all'uso ed allo smaltimento di farine fossili, sia per ridurre l'impatto ambientale dei processi industriali di finitura della birra. Le condizioni operative ottimali per questo sistema sono state determinate in un impianto da banco specificatamente progettato.

I risultati ottenuti hanno permesso di determinare e minimizzare il prevalente meccanismo di sporramento, utilizzando opportuni pretrattamenti (centrifugazione e trattamento enzimatico) sulla birra torbida, si sono studiati anche i costi operativi e l'impatto ambientale per un ipotetico impianto di chiarificazione e stabilizzazione di birra lager industriale

Parole chiave: birra lager; chiarificazione; condizioni operative ottimali; controlavaggio con CO₂; flusso di permeazione; impronta del carbonio; meccanismi di sporramento; membrane tubolari ceramiche; microfiltrazione; stabilizzazione.

Novel Lager Beer Clarification and Stabilisation Process Using Ceramic Tubular Microfiltration Membranes

1. Introduction

In accordance with the PhD thesis project previously described (Cimini, 2011), this oral communication reports the main results of the following four activities directed to:

- A1) Assess the optimal operating conditions (*i.e.*, feed superficial velocity, v_s ; transmembrane pressure difference, TMP; membrane porosity; CO₂ backflushing strategy) for the Cross Flow Microfiltration (CFMF) process of rough beers obtained from commercial hopped-malt extracts using a bench-top plant, appropriately designed and equipped with ceramic tubular membrane modules, as well as their main beer characteristics accordingly to EBC (2010).
- A2) Validate the above optimal operating conditions in terms of the average permeation flux ($J_{v,av}$) and colloidal stability of microfiltered beer at 0 °C by using rough malt beers obtained in the pilot-scale brewery by the Italian Brewing Research Centre (CERB, Perugia, Italy), and a pale lager produced in an industrial brewery (Birra Peroni Spa, RM, IT) as such or after a few preliminary treatments.
- A3) Assess the fouling mechanisms for beer CFMF on the basis of the main dead-end filtration models.
- A4) Develop a novel combined system for clarifying and stabilizing lager beer at a commercial chill haze of 0.6 EBC unit and assess the specific operating costs and global warming potential.

2. State of the Art

The clarification and pasteurization of rough beer are the final and most important processing steps in order to obtain a bright and microbiologically stable beer with no temporary haze after cold storage and/or serving temperatures. Green beer is currently filtered in presence of filter-aids (mainly diatomaceous earth, DE, or kieselguhr) and polyvinylpyrrolidone (PVPP) and then pasteurized. Since the crystalline silica present in DE is regarded as a cause of lung disease, DE use and recovery for either recycling or sludge disposal are not only problematic, but also expensive, DE sludge disposal costs being ~ €170/Mg (Fillaudeau *et al*, 2006).

CFMF appears to be the only alternative to get rid of filter-aid disposal problems, reduce beer losses, enhance solid-handling capacity, and replace heat pasteurisation. Unfortunately, membrane fouling limits the average permeation flux ($J_{v,av}$) to about one fifth of that (250-500 L m⁻² h⁻¹) achievable with powder filters (Buttrick, 2007). Moreover, unsatisfactory separation properties are responsible for some negative quality aspects, uncertainty in productivity, and large flux/quality variations among different beer brands as filtered on the same membrane system. Thus, more than 85% of all brewers still perform beer clarification with DE (Fillaudeau *et al*, 2007).

There is however a great deal of interest for this technique. Three commercial CFMF systems using hydrophilic, polyethersulphone (PES) membranes, with porosity of 0.45-0.65 µm and hollow-fibre (Norit and Pall) or flat-sheet (Alfa Laval) modules, are currently available for beer clarification. In particular, both Pall and Alfa Laval processes rely on a pre-centrifugation step to remove yeast cells and larger aggregates, and thus minimise membrane fouling.

The main environmental issues associated with brewing include material, water and energy consumption, as well as waste production. Brewery processes are intensive users not only of both electrical and thermal energy, but also of good-quality water, their consumption being 8-12 kWh, 100-200 MJ and 4-7 hL per hL of beer produced, respectively (Olajire, 2012). Additionally, wastewater generation ranges from 1 to 5% of total beer production. Several studies have been so far carried out to check for the environmental impact generated through the entire life cycle of some types of beer (Cordella *et al*, 2008; Koroneos *et al*, 2005; Talve, 2001). Certification of the environmental performance of lager beer packed in disposable 33-cL glass bottles, 25-L steel kegs, or 20-L plastic drums was recently reported (EPD, 2011). The greenhouse gas (GHG) emissions due to the production process represented 15-18% of the overall ones, those associated to the upstream agriculture production or downstream processes, when including glass bottle production or steel keg distribution, being the greatest ones (Cordella *et al*, 2008; Koroneos *et al*, 2005; Talve, 2001).

2. Materials and Methods

Three different types of beer were used in this work. The rough beer labelled A was produced in the laboratory-scale, that labelled B, in the pilot-scale brewery at CERB (Perugia, Italy) and finally that labelled C at the brewery plant of Birra Peroni (Roma, Italy). Rough beer A and B were produced in 25-L lots from worts (density: 1.045 kg L^{-1}) obtained by diluting hopped-malt extracts (Pils, Brewferm, Beverlo, B) with tap water at 80°C or by mashing 100% pils malt (Durst-Malz, Bruchsal-Heidelberg, Bruchsal, D) and hopping with traditional bitter Hallertau Magnum hop pellets. Fermentation was started by adding 11.5 g of dry yeast of type Safale S-04 or Saflager W-34/70 (Fermentis, Marcq-en-Barœul, F), respectively. The corresponding fermentation temperature was kept constant at ~ 20 or 12°C for about 4 or 10 days, then was gradually lowered to 15 or $2 - 4^\circ\text{C}$ over the following 4 days. After racking, all rough beers were stored in stainless-steel maturation vessels and kept at 4°C for about 15 or 30 days, respectively. The optimal operating conditions for beer clarification via CFMF were assessed using rough beer A as such, and were confirmed using rough beer B, as such or after pretreatments. Some samples were clarified using a laboratory centrifuge (Beckman mod. J2-21) at 6000xg and less than 4°C for 10 min, once collected in 0.3-L plastic bottles. Others were mashed by adding 0.15 mL of a commercial Beerzym PENTA preparation (Erbslöh Geisenheim AG, Geisenheim, Germany) per L of rough beer at 4°C for 24 h to degrade almost all the pentosans and β -glucans present, and then centrifuged as reported above.

Beer clarification was carried out using a typical temperature- and pressure-controlled bench-top CFMF plant, appropriately designed and assembled (Fig. 1). It was equipped with ceramic tubular membrane modules (US Filter, Warrendale, PA, USA) with 6-mm inside diameter, 500-mm length, 94.2-cm^2 effective membrane surface area, and porosity of 0.4, 0.8, or $1.2 \mu\text{m}$. As suggested by Gan *et al* (1999), membrane cleaning was sequentially carried out by caustic and oxidation, and acidic cleanings. The water permeability at $20.0 \pm 0.1^\circ\text{C}$ for the 0.4-, 0.8-, or $1.2\text{-}\mu\text{m}$ membrane module was 521 ± 37 , 773 ± 17 , or $1716 \pm 44 \text{ L m}^{-2} \text{ h}^{-1} \text{ bar}^{-1}$ ($r^2 = 0.99$), respectively. A stainless steel Lowara centrifugal pump (Montecchio Maggiore, I) was piloted using a 0.75 kW electric motor via a frequency inverter Commander SK (Control Techniques, Powys, UK). Feed and retentate pressure and flow rates were monitored using digital and analogical pressure and flow rate sensors.

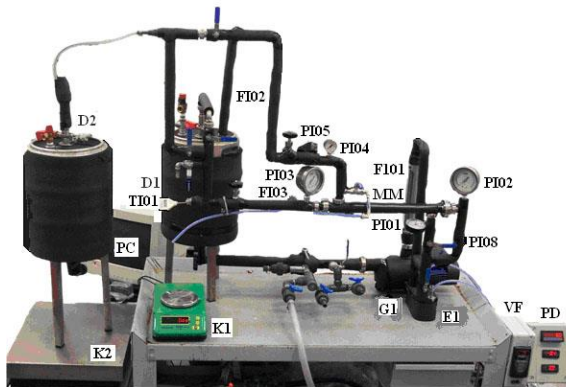


Figure 1 Photograph of the bench-top CFMF plant used in this work. Equipment identification items: D, tank; E1, plate heat exchanger; FI, flow meter; G1, centrifugal pump; K, balance; MM, CFMF membrane module; PC, personal computer; PD, soft instrument panel; PI, manometer; TI, temperature indicator; VF, frequency inverter.

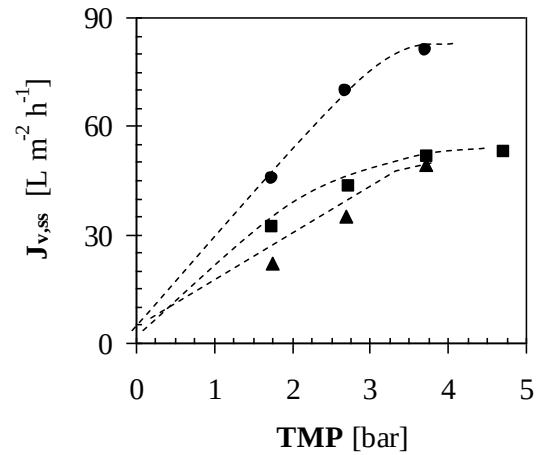


Figure 2 Effect of TMP on the quasi steady-state permeation flux ($J_{v,ss}$) of rough beer A samples with turbidity > 7 EBC unit at $v_s = 6 \text{ m s}^{-1}$ and 10°C using ceramic tubular modules of different porosity: $\blacktriangle$, $0.4 \mu\text{m}$; $\blacksquare$, $0.8 \mu\text{m}$; $\bullet$, $1.2 \mu\text{m}$.

The permeate flow rates were assessed via electronic balances K1 and K2, both connected to a PC. Total recycle runs were carried out at ~ 10 °C by varying TMP and v_s in the ranges of 1-5 bar, and 2-6 m/s, respectively. Membrane cleaning with CO₂ backflushing was periodically carried out at a backflush pressure difference between the permeate and retentate sides of +3 bar for 2 min. The green beers and permeates were assayed for pH; density; viscosity; turbidity or haze (H) at 20 or 0 °C; colour; β -glucans, real or original extract, ethanol content, and total polyphenol concentration (Analytica EBC, 2010).

3. Results and Discussion

3.1 Assessment the Optimal Operating Conditions for CFMF of Rough Beer A

Previously (Cimini, 2012; Cimini and Moresi, 2013), the CFMF performance of samples of green beer A with initial turbidity of 1.1-12.0 EBC unit was studied in the bench-top plant, equipped with the 0.8- μ m membrane module, at ~ 10 °C by varying v_s and TMP in the ranges of 2-6 m s⁻¹, and 0.96-4.73 bar, respectively. In all conditions tested and independently of beer turbidity, the instantaneous permeation flux (J_v) showed a drastic reduction and tended to a quasi steady-state value ($J_{v,ss}$) after about 1 h. By operating at TMP=3.73 bar, $v_s=6$ m s⁻¹, and turbidity of 1 or 4 EBC unit, an average permeation flux of 258 or 199 L m⁻² h⁻¹ was respectively achieved, thanks to periodic CO₂ back-flushing.

The effectiveness of such operating conditions was established with samples of rough beer A at hazes of 1.1-1.27 or >7 EBC unit using the above three membrane modules at 10 °C and TMP values progressively increasing from ~ 1 to 5 bar under a constant v_s of 2, 4 or 6 m s⁻¹. As the porosity was increased from 0.4 to 1.2 μ m, $J_{v,ss}$ exhibited an increasing limiting permeation flux for TMP >2 bar at $v_s=2$ m s⁻¹ or for TMP ≈ 3 -4 bar for $v_s=4$ -6 m s⁻¹ (see for instance Fig. 2). In terms of beer quality, the smaller the pore size the higher the membrane retention for density and colour was found, whereas that for haze and viscosity was about constant. Despite its smaller $J_{v,ss}$, the 0.8- μ m module was selected owing to its greater haze retention with just 6% reduction in the permeate colour against the 18% reduction obtained with the 0.4- μ m module, as well as to a presumed higher microorganism retention *versus* the 1.2- μ m module. Such a choice will be further validated (see below).

3.2 Establishment the Optimal Operating Conditions for CFMF of Rough Beer B

The effectiveness of the aforementioned operating conditions (TMP=3.74 bar, $v_s=6$ m s⁻¹, T=10 °C) for the 0.8- μ m membrane module was further tested with rough beer B, as such or after a few pretreatments. Also in this case, the permeation flux reached the $J_{v,ss}$ value after ~ 1 h whatever the beer turbidity (Cimini *et al*, 2013). For H ranging from 0 to 2-3 EBC unit, $J_{v,ss}$ drastically reduced and tended to an asymptotical value of 91 ± 8 L m⁻² h⁻¹ for H >7 EBC unit. Figure 3A compares the time course of the experimental permeation fluxes (J_v) under periodic CO₂ backflushing.

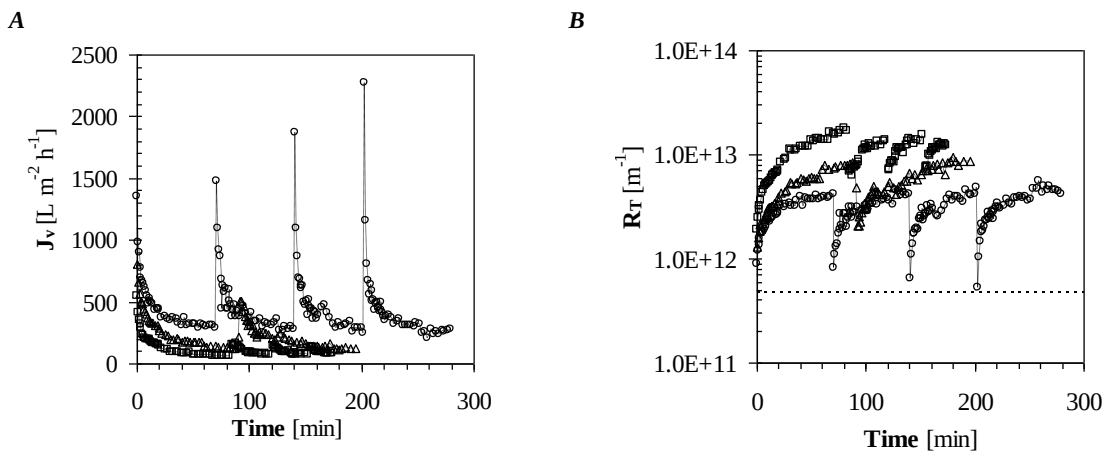


Figure 3 Time course of (A) the permeation flux (J_v) and (B) overall hydraulic resistance (R_t) under constant TMP (3.74 bar), v_s (6 m/s), T (10 °C) and periodic CO₂ back-flushing when using rough beer B samples as such ($\square$, 18 EBC unit), precentrifuged ($\triangle$, 1.5 EBC unit) or after enzymatical and centrifugal pretreatments ($\circ$, 0.9 EBC unit). The horizontal broken line refer to the clean membrane hydraulic resistance (R_m).

Thanks to the removal of most of suspended matter, the centrifugation step resulted in ~50 or 75% increase in $J_{v,ss}$ ($137 \pm 13 \text{ L m}^{-2} \text{ h}^{-1}$) or $J_{v,av}$ ($205 \text{ L m}^{-2} \text{ h}^{-1}$). The use of the enzyme preparation lowered the β -glucan content from ~140 to 1.3 mg L^{-1} , this avoiding gel layer formation or adsorption of finer aggregates within the membrane pores. The observed $J_{v,ss}$ ($294 \pm 30 \text{ L m}^{-2} \text{ h}^{-1}$) or $J_{v,av}$ ($336 \text{ L m}^{-2} \text{ h}^{-1}$) values were more than two or three fold higher than those detected after the only centrifugation

Like the great majority of DE-filtered beers, those submitted to CFMF even at 0°C still retained the soluble haze precursors responsible for post-filtration hazes. These are generally adsorbed with PVPP, alone or combined with selected carrageenan or silica xerogel (Rehmanji *et al*, 2005), or with agarose beads (Taylor *et al*, 2006).

As the permeates of beer B had been cooled to 0°C , their chill haze ranged from 1.5 to 7.6 EBC unit. To obtain a brilliant, chill-proof and microbiologically stable beer, ready-to-be aseptically packaged, samples of beer B, after enzymatic mashing and centrifuging, were added with 0.3 or 0.5 g L^{-1} of regenerable PVPP (size: $50\text{--}250 \mu\text{m}$) and fed to the CFMF module. Their initial total polyphenol content ($224\text{--}297 \text{ mg L}^{-1}$) was reduced by 20 or 40% and the permeate turbidity at 0°C to 0.74 or 0.6 EBC unit, respectively; but $J_{v,av}$ fell to $84 \pm 4 \text{ L m}^{-2} \text{ h}^{-1}$

3.3 Assessment of the Main Membrane Fouling Mechanisms

To discriminate the membrane fouling mechanisms, the time course of the overall resistance (R_T) to filtrate flow was assessed in accordance with Darcy's law (Bolton *et al*, 2006). As shown in Figure 3B, during the CFMF of rough beer B the overall resistance was definitively greater than that of clean membrane [$R_m = (4.7 \pm 0.1) \times 10^{11} \text{ m}^{-1}$]. The pre-centrifugation step not only reduced the growth rate of R_T , but also approximately halved its maximum value to $0.88 \times 10^{13} \text{ m}^{-1}$. The enzymatic pre-treatment further reduced R_T to $\sim 0.41 \times 10^{13} \text{ m}^{-1}$. Moreover, CO_2 backflushing lowered R_T to $(5.2\text{--}8.1) \times 10^{11} \text{ m}^{-1}$, these values being quite near to R_m . In all cases, the time course of R_T exhibited a downward concavity, this being typical of intermediate blocking or external fouling due to cake filtration. According to Cimini *et al* (2014), the time course of the experimental filtrate volumes collected during constant TMP operations was satisfactorily reconstructed by using the cake fouling model (Bolton *et al*, 2006). This fouling mechanism congruently explained the efficacy of CO_2 backwashing to restore the original permeation flux.

3.4 Final validation tests for the combined stabilization and crossflow microfiltration of an industrial rough pale lager

Different lots of a pale lager, produced in the industrial brewery Birra Peroni (Rome, Italy), were used to confirm the optimal operating conditions previously established. A sequential clarification, PVPP stabilization and CFMF process was assessed using this industrial rough lager beer, as such or pre-centrifuged (C) to remove yeast cells and larger aggregates.

Contrary to previous testing on a malt-based rough beer containing as much as $140\text{--}250 \text{ mg L}^{-1}$ of β -glucan (Cimini *et al.*, 2013ab; 2014), the rough pale lager under study contained as little as $(16.8 \pm 0.9) \text{ mg}$ of β -glucan L^{-1} . In the circumstances, the β -glucanase and pentosanase pretreatment previously used appeared to be useless. It was possible to limit the fouling contribution of yeast cells and aggregates during beer CFMF by the sequential steps of centrifuging, PVPP stabilization and cartridge filtration. In fact, the resulting average value of the permeation flux was about $(337 \pm 1) \text{ L m}^{-2} \text{ h}^{-1}$ thanks to the periodic CO_2 backflushing. To assess the economic feasibility of this novel CFMF process with respect to the conventional powder filters (DEF) when using single-use (SU) or regenerable (R) PVPP, a rough-grade feasibility study was carried out by referring to an industrial plant with an annual production capacity of about $3 \times 10^6 \text{ hL}$ of lager beer, based on three shifts per day, 300 days per year. The use of regenerable PVPP resulted to be beneficial whatever the filtration procedure used. Nevertheless, the use of CFMF together with regenerable PVPP reduced not only the overall beer clarification and stabilization costs (C_o) by ~36%, that is from $\text{€}0.32$ to $\text{€}0.12/\text{hL}$; but also the overall environmental impact by ~27%, that is from 2.25 to $0.60 \text{ kg CO}_2\text{e}/\text{hL}$. The CFMF in conjunction with single use PVPP resulted in the minimum global warming potential ($0.25 \text{ kg CO}_2\text{e}/\text{hL}$), but the overall operating costs increased to $\text{€}0.39/\text{hL}$.

3.5 A novel beer clarification and stabilisation process using CFMF membrane

To minimise the fouling contribution of yeast cells, aggregates and polysaccharides during CFMF and maximize the effectiveness of PVPP stabilization and regeneration, the process flow sheet shown in Fig. 4 was outlined. The same cylindro-conical tank D1 used to ferment the wort allows the yeast to settle to its bottom at the end of fermentation so as to be collected in tank D4. By feeding the rough beer to the centrifuge C1, a yeast cream is recovered and conveyed to D4, while the centrifuged beer is fed to tank D2. A thick slurry of regenerable PVPP is then added at a rate of 20 to 50 g per hL by a proportioning pump. An inert gas (CO_2 or N_2) is also sparged to ensure thorough dispersion and minimum O_2 pick-up.

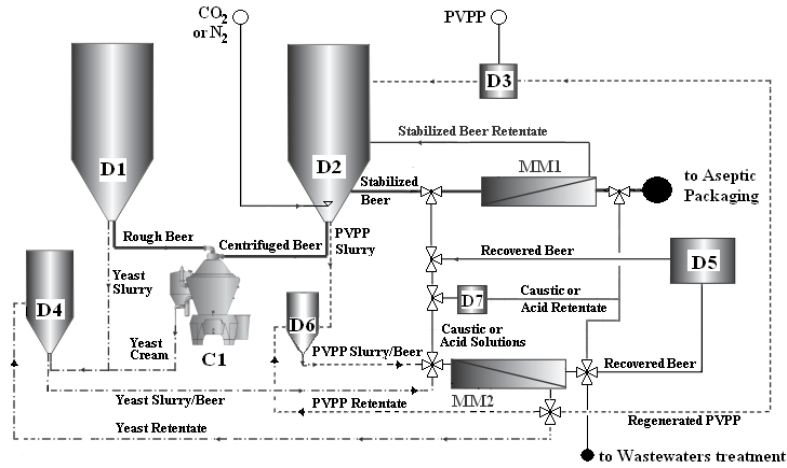


Figure 4 Schematic diagram of the novel lager beer clarification and stabilisation process using tubular ceramic microfiltration membranes developed in this work. Equipment identification items: C, centrifuge; D1, fermentation and maturation uni-tank; D2, PVPP treatment tank; D3, PVPP dosing tank; D4, yeast tank; D5, recovered beer tank; D6, PVPP slurry tank; D7, tank for cleansing solutions; MM1, CFMF membrane module for beer clarification; MM2, CFMF membrane module for beer recovery from yeast or PVPP slurry or for PVPP regeneration. Product, by-product and ancillary identification lines: Beer, —; Yeast, · · · · ·; PVPP, - - - -.

After a contact time of 24-48 h at $\sim 0^\circ\text{C}$, spent PVPP-polyphenol complexes are primarily removed by settling and accumulated in tank D6. Stabilized beer is pumped to the CFMF module MM1 (pore size: 0.8 or 1.2 μm). By setting the aforementioned operating variables, it is expected an average permeation flux of $\sim 200 \text{ L m}^{-2} \text{ h}^{-1}$, and a bright, stabilised permeate ready for aseptically packaging with no supplementary pasteurization step.

Another CFMF system MM2, equipped with ceramic tubular membrane modules too, is used to recover sequentially not only the remaining beer from either yeast or PVPP slurry, while its corresponding retentate is recirculated through tank D4 or D6; but also to regenerate used PVPP (Gopal and Rehmanji, 2000).

4. Conclusions and Future Perspectives

A combined clarification and PVPP stabilisation procedure using 0.8- μm ceramic tubular membrane modules was developed and tested on a bench-top plant scale. By avoiding DE disposal problems and heat pasteurisation, and minimising beer losses, such a novel process appeared to be not only a reliable alternative to conventional powder filters in terms on the average permeation flux, but also a cost-effective one capable of mitigating the related life cycle GHG emissions. By referring to an industrial plant capacity of 2×10^6 hL of lager beer, the estimated overall costs and global warming potential for lager beer clarification and stabilization were about 30% smaller than those associated with the current industrial DE-filtration and regenerable PVPP stabilisation procedures.

INTRODUCTION

Beer clarification, stabilization, and pasteurization are the final and most important processing steps in order to obtain a bright and microbiologically stable beer with no temporary haze after cold storage and/or at serving temperatures.

Beer haze may be of the biological or non-biological type. The former is due the growth of either bacteria or wild yeasts, while the latter depends on some polypeptides and polyphenols originating from barley and hops that give rise to flocs responsible for the so called *chill* or *permanent haze* provided that it disappears or is persistent at room temperature (McMurrough *et al.* 1992; Siebert *et al.* 1996).

The conventional beer clarification process employs filter presses or pressure vessel filters, which are commonly pre-coated with filter aids in the form of porous particles, mainly of diatomaceous earth (DE or kieselguhr), but also of perlite, cellulose or active carbon, which play an important role not only in acting as a second filtration barrier, but also in adsorbing the chill haze components (Buttrick, 2007). This procedure has been successfully used for decades in beer filtration, but use of this material, as well as disposal of spent filter sludge, needs ensuring safe working conditions, having the World Health Organization ascertained that the crystalline silica present in DE causes lung disease. Thus, DE sludge disposal is currently the main cost (ca. € 170 per ton of DE purchased) of beer filtration (Fillaudeau *et al.* 2006; Knirsch *et al.* 1999).

Alternatively, a single step method based on cross-flow microfiltration (CFMF) might be used. The first research applying CFMF in the brewing industry dates back to the early 1980s (Gir & Leeder, 1992). Since then, this topic has been an area of intense research in order to get rid of filter-aid handling and disposal problems (Freeman & McKechinie, 1995), reduce beer losses (Lee, 1987), handle high suspended solid liquors, and replace heat pasteurization (Czekaj *et al.* 2000; Fillaudeau *et al.* 2006). Unfortunately, membrane fouling limits the average permeation flux ($J_{v,av}$) to about one fifth of that (250-500 L m⁻² h⁻¹) practically achievable with powder filters (Buttrick, 2007; Fillaudeau *et al.*, 2006). Moreover, unsatisfactory separation properties result in poor beer quality and uncertain productivity, especially when different beer brands are filtered on the same membrane system. Thus, nearly 99% of all brewers still perform beer clarification with DE (Anonymous, 2007).

Beer is a complex fluid, with multiple compounds (i.e., yeast cells, proteins, polysaccharides and polyphenols) leading to different kinds of fouling. Generally

speaking, yeast cells tend to form a cake layer over the membrane surface, the thickness of which being controlled by the beer crossflow velocity. The internal fouling of membranes is attributed to arabinoxylans, β -glucans, and hydrophilic (haze-forming) proteins and polyphenols (Gan *et al.* 1997, 2001; Taylor *et al.* 2001) as they penetrate inside the membrane itself and are adsorbed onto the membrane pore walls, this leading to pore constriction or blocking. The polysaccharides may also form complexes with proteins or gels (Güell and Davis, 1996), these entrapping other macromolecules or aggregates either in the cake layer or inside the membrane itself. Also, the complex interactions between fouling molecules and membrane material should be taken into account (Czekaj *et al.* 2000). Yeast cells may sometimes play an antifouling role by forming a less compact deposit at the membrane surface that acts as a secondary membrane, the latter retaining protein aggregates and protecting the membrane itself against internal fouling (El Rayes *et al.* 2011; Guëll *et al.* 1999). Whereas the cake layer and some of the aggregates accumulated within the porous membrane structure can be removed by short-term back-flushes, most of the proteins and aggregates adsorbed onto or inside the membrane are only removable by acid and/or alkaline cleaning enriched with oxidative agents (Gan *et al.*, 1999).

Whilst the mechanisms of fouling and flux decline in beer microfiltration are currently better understood (Blanpain-Avet *et al.* 1999; Burrell *et al.* 1994; Gan *et al.*, 1997; van der Sman *et al.* 2012), there is no systematic analysis of the dynamic and inter-dependent relationships between permeate quality, flux and membrane fouling.

Membrane porosity affects not only the permeation flux (Stopka *et al.* 2000), but also the head retention values (Gan *et al.* 1997). By increasing the transmembrane pressure difference (TMPD), the beer permeation flux increases, but not proportionally (Thomassen *et al.* 2005). Feed superficial velocities of 0.87 - 7.2 m s⁻¹ exerted a strong effect on the reversible external membrane resistance (Filladeau & Lalande, 1998). Generally, membrane fouling dramatically reduces the flux to far below the critical steady-state flux of 100 L m⁻² h⁻¹ (Filladeau *et al.* 2006). There is also detectable change in the beer quality after filtration (e.g., retention of flavor-active constituents and decrease in foam stability).

Several hydrodynamic techniques, using fluid instabilities (such as co-current mode, pulsating flow, periodic stop of TMPD, periodic back-flush or back-shock

process, generation of Dean or Taylor vortices, introduction of turbulence promoters as baffle channel or stamped membrane), two-phase flow (gas-liquid, liquid-solid), or rotating and vibrating filtration devices, have been so far investigated with poor results (Blanpain-Avet *et al.* 1999; Fillaudeau *et al.* 2007; Gan *et al.* 1997; Kuiper *et al.* 2002). To increase the permeate flux, a back-flushing with permeate was employed (Gan *et al.* 2001; Sondhi & Bhave, 2001) and a 10-h average flux of $22 \text{ kg m}^{-2} \text{ h}^{-1}$ was achieved. Nevertheless, the industrial plants using CFMF do not rely yet on such complex dynamic filtration technologies. In fact, at the Heineken brewery in Zoeterwoude (NL) about $10 \text{ m}^3 \text{ h}^{-1}$ of rough beer are filtered through $0.45\text{-}\mu\text{m}$ polyethersulphone (PES) hollow-fiber (HF) modules accordingly to the so called Norit process at 0°C , 2 m s^{-1} crossflow velocity, and TMPD up to 1.6 bar (Noordman *et al.* 2001). By applying 10-min periods of back-flushing every 2 h during filtration, the permeation flux was kept at $100 \text{ L m}^{-2} \text{ h}^{-1}$, while the beer permeated accomplished the EBC standards in terms of turbidity near to 0.6 EBC unit, bitterness, total extract, color, and protein content.

Nowadays, besides the above Norit process that makes use of a cylindro-conical tank to allow yeast to settle to its bottom before recirculating the rough beer through the CFMF modules, two other combined beer filtration processes, consisting of a high-performance centrifuge and $0.65\text{-}\mu\text{m}$ PES, HF (Pall) or flat-sheet (Alfa Laval) membrane modules, are commercially available (Buttrick, 2007). It is still unclear whether the centrifugation stage is effectively convenient to minimize membrane fouling by yeast cells and larger aggregates, especially because of its quite high additional investment and operation costs.

Thus, the complexity of beer clarification makes the optimal performance of CFMF still far away to be assessed.

In synthesis, the main aims of this PhD thesis were:

- A1) to assess the optimal operating conditions (*i.e.*, feed superficial velocity, v_s ; transmembrane pressure difference, TMPD; membrane porosity; CO_2 backflushing strategy, BF) for the Cross Flow Microfiltration (CFMF) process of rough beers obtained from commercial hopped-malt extracts by using a bench-top plant, appropriately designed and equipped with ceramic tubular membrane modules, as well as the main beer characteristics accordingly to Analitica EBC (2010);

- A2) to validate the above optimal operating conditions in terms of the average permeation flux ($J_{v,av}$) and colloidal stability of microfiltered beer at 0 °C by using either rough malt beers obtained in the pilot-scale brewery by the Italian Brewing Research Centre (CERB, Perugia, Italy), or pale lagers obtained in the industrial brewery of Birra Peroni (Roma, Italy) as such or after a few pretreatments including centrifugation with or without enzymatic hydrolysis of the constitutive gel forming polysaccharides;
- A3) to assess the main fouling mechanisms of rough beer CFMF by referring to the most important classical dead-end filtration models;
- A4) to develop a novel combined system for clarifying and stabilizing rough lager beers at a commercial chill haze of 0.6 EBC unit and assessment of the operating costs and carbon footprint of a functional unit of 1 hL of microfiltered lager beer.

CHAPTER 1:
The State of the Art on Beer Production,
Quality and Environmental Impact

1.1 – BEER PRODUCTION, CONSUMPTION AND CONSUMER

Beer is the alcoholic beverage mostly consumed in the world. It is a very popular drink, not only because of its low alcohol content, but also for the extensive marketing and promotion activities carried out by multinational companies in almost every country in the world.

In the food industry, the brewing sector holds a strategic economic position, the annual world beer production having exceeded 1.8 billion hL in 2010 (Barth-Haas Group and Germain Hansmaennel, 2010)

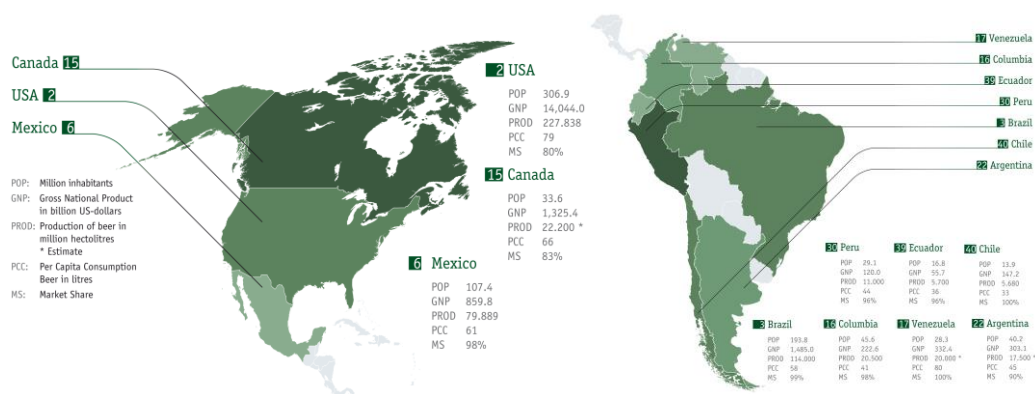


Figure 1.1 Beer production in the North, Central and South America. (Haas Group and Germain Hansmaennel, 2010)

In spite of the general economic and financial crisis, in 2008 the worldwide beer market marked a growth of 1.6%, quite a half of the average growth rate of about 3% per year of previous years. In 2009, the global volumes of beer production and consumption were about at the same levels of the previous year. In 2010, the latest Barth Reports indicated a slight recovery in the world production of beer to 1.846 billion hL (Table 1), while the average per capita consumption was around 27 L/year.

According to the latest report *Global Beer Market Trends* (www.canadean.com), the worldwide beer market will grow until 2015 at an average annual rate of around 2.8%. However, global data mask large regional differences. In Asia, beer consumption is growing in a more accelerated way than in other continents. Asia has just conquered the world leadership with approx. 630 million hL produced, although per capita consumption is at the level of 15 L/year.

The top 10 brewing nations represented one third of the world population, but two thirds of global production of beer. The first nation is China, that produced 448 million hL of beer in 2010, with a per capita consumption of only 33 L/year, and just passed the historical primacy of the U.S. market.

According to the *Global Beer Market Trends*, the Chinese beer market is expected to reach 573 million hL by 2015, this representing over one quarter of all beer consumption in the world. Over the past 10 years, China has more than doubled the production and consumption of beer.

The U.S. market with 228 million hl of production in 2010 occupies the second place in the ranking of the world brewing nations.

Table 1.1 *The 40 largest brewing groups in the world as of December, 31st,2010 (Haas Group and Germain Hansmaennel, 2010)*

Brewery	Country	Production vol. 2010 in mill. hl	Percentage of world beer production
1 AB InBev ¹⁾	Belgium	358.7	19.4 %
2 SABMiller ²⁾	United Kingdom	172.3	9.3 %
3 Heineken	Netherlands	145.9	7.9 %
4 Carlsberg	Denmark	114.0	6.2 %
5 China Resource Brewery Ltd.	China	93.3	5.1 %
6 Tsingtao Brewery Group	China	64.0	3.5 %
7 Grupo Modelo	Mexico	51.9	2.8 %
8 Yanjing	China	50.3	2.7 %
9 Molson-Coors	USA/Canada	48.7	2.6 %
10 Kirin	Japan	30.3	1.6 %
11 Efes Group	Turkey	24.2	1.3 %
12 BGI / Groupe Castel	France	23.5	1.3 %
13 Diageo (Guinness)	Ireland	22.0	1.2 %
14 Asahi	Japan	21.8	1.2 %
15 Polar	Venezuela	20.7	1.1 %
16 Grupo Schincariol	Brazil	20.0	1.1 %
17 San Miguel Corporation	Philippines	19.9	1.1 %
18 Gold Star	China	19.3	1.0 %
19 Chongqing Beer	China	17.8	1.0 %
20 Radeberger Gruppe	Germany	13.1	0.7 %
21 StarBev	Czech Republic	13.0	0.7 %
22 Grupo Mahou - San Miguel	Spain	12.5	0.7 %
23 Pearl River	China	12.1	0.7 %
24 Singha Corporation	Thailand	11.9	0.6 %
25 Petropolis	Brazil	11.0	0.6 %
26 Hite	South Korea	10.4	0.6 %
27 Saigon Beverage Corp.	Vietnam	10.1	0.5 %
28 Obolon	Ukraine	9.9	0.5 %
29 CCU	Chile	9.3	0.5 %
30 Shenzhen Kingway	China	9.2	0.5 %
31 United Brewery	India	8.8	0.5 %
32 Damm	Spain	8.5	0.5 %
33 Foster's Group	Australia	8.5	0.5 %
34 Oettinger	Germany	8.2	0.4 %
35 Oriental Brewery	South Korea	7.8	0.4 %
36 Suntory	Japan	7.5	0.4 %
37 Bitburger Braugruppe	Germany	7.4	0.4 %
38 Beer Thai (Chang)	Thailand	7.2	0.4 %
39 Sapporo	Japan	7.0	0.4 %
40 Habeco	Vietnam	6.0	0.3 %
Total		1,518.0	82.2 %
World beer production 2010		1,846.4	100.0%

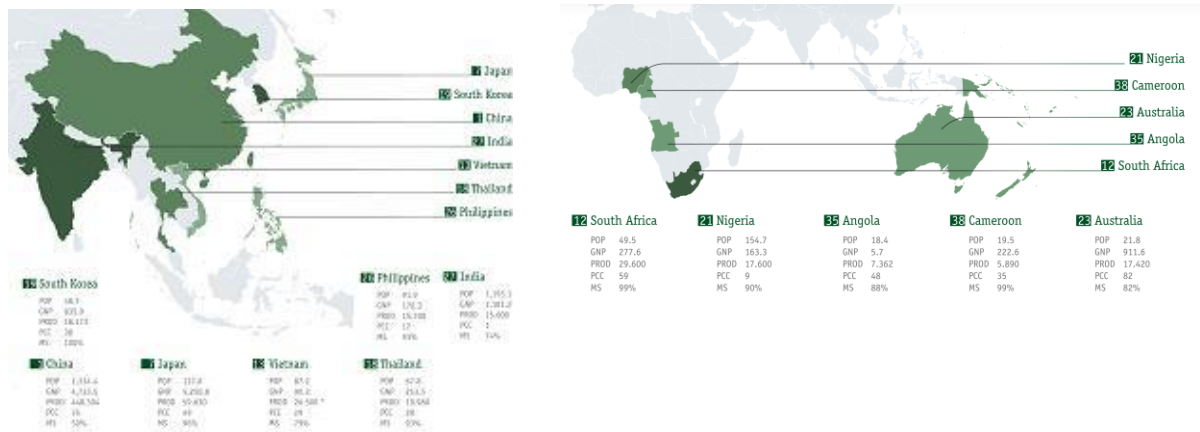


Figure 1.2 Beer production in Asia, Africa and Australasia (Bath Report 2010-11)

Europe has lost its historical primacy in the production and consumption of beer. Just 5 years ago, Europe was the first continent in the world in terms of beer production. Now, it is back in the third place after Asia and America. In the European context, in the 2010 there was a confirm of the general downward trend, that began in 2008 (-1.4% compared to 2007) and worsened in 2009 (-4.4% compared to 2008) with a production of almost 387 million hL, down by -1.2% compared to the previous year.

Germany has maintained its historical primacy in beer production and consumption till 2007. Now, it is second in the ranking of European beer producing countries, with an annual production below 100 million hL. Russia is producing about 103 million hL of beer and is the fourth largest brewing capacity in the world.

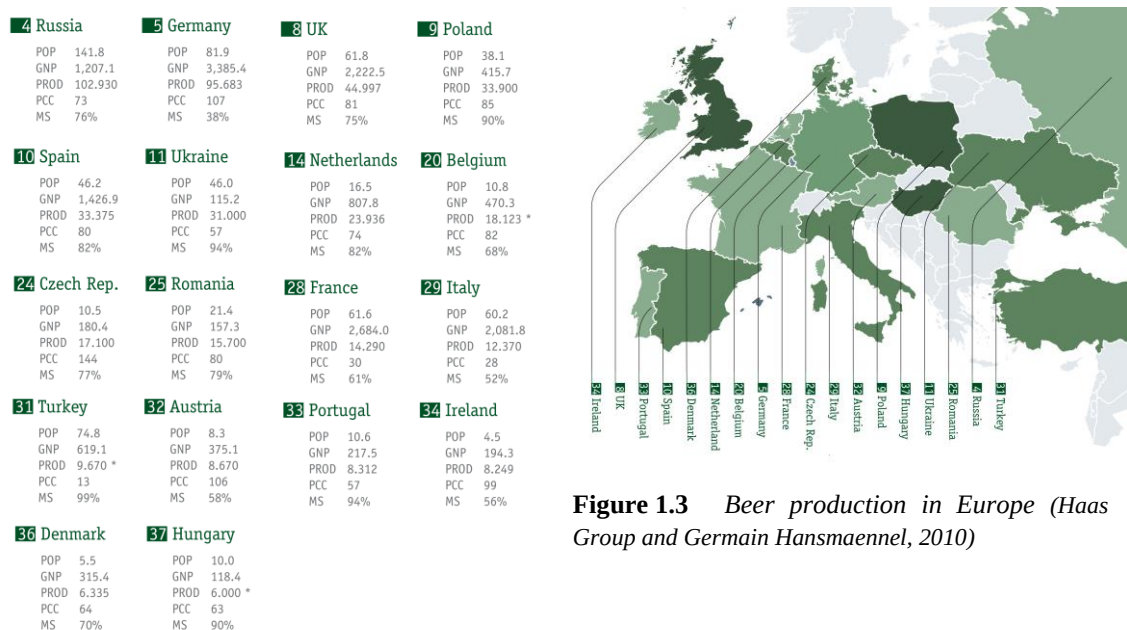


Figure 1.3 Beer production in Europe (Haas Group and Germain Hansmaennel, 2010)

In 2010, the United Kingdom produced 45 million hL of beer and occupied the eighth place in the world and the third one in the European Union. Poland (34 million hL) and Spain (33 million hL) were on the fourth and fifth places in the European Union, respectively. The emerging Ukraine produces more than 30 million hL, followed by Holland (25 million hL), Belgium (18 million hL) and finally the Czech Republic (17 million hL).

The countries that traditionally consume the greatest amounts of beer in Europe are: Czech Republic (134 L/pro capita/year), Germany (107.4 L) and Austria (106 L) (*Assobirra*, 2011). The per capita consumption of beer in France and Italy, is quite lower (29 L/yr).

Italy is at the tenth place among the European beer-producing nations. Regarding beer consumption, the Italian per capita consumption declined from 31.1 L/yr in 2007 to 29.4 L/yr in 2008 and to 28 L/yr in 2009, but slightly increased to 28.6 L/yr in 2010. About three Italian adults out of four (72.4%), that is about 36 million people, consider themselves beer drinkers, while the 79.5% of all claims to drink wine. This is in contrast with the European beer consumption: after the peak of 2007, when the European per capita beer consumption exceeded 80 L/yr, it reduced by 12.6% to 69.9 L/yr. In 2010 in Italy both beer production (+0.3% compared to 2009) and per capita consumption increased (+2.1%). By comparing the performance of the Italian brewing sector with the European one, more than 350 production facilities (16 of which being industrial plants) and more than 1,500 different brands can be accounted for. This involves an overall occupation, including related industries, of almost 150,000 people, thanks also to an increasing export, that nearly doubled in five years. The Italian brewing industry has thus shown some signs of recovery, confirming its role as a major player in the national economy: almost 13 million hL of beer products, just under 2 million hL of beer exported, 1 billion Euros spent on the purchase of goods and services, 4 billion Euros of revenue for state finances (*Assobirra*, 2011).

1.2 – BEER QUALITY PARAMETERS AND ANALYSIS

A number of scientific organizations carry out collaborative tests to specify the brewing analytical methods so as to confer them an official status. These organizations include:

- *American Society of Brewing Chemists* (ASBC),
- *Brewery Convention of Japan* (BCOJ),
- *European Brewery Convention* (EBC),
- *Institute of Brewing and Distilling* (IBD),
- *Mitteuropäische Brautechnische Analysenkommission* (Central European Brewing Technology Analysis Commission) (MEBAK).

The product quality attributes that consumers can directly perceive are the following: *flavor* (odor, taste) and *appearance* (color, haze, and foam). These are frequently assessed by sensory panels, but often also by analytical measurements, these being more precise, accurate and reproducible than panel results.

The required characteristics of a beer for sale depend upon the consumer being targeted and can be readily defined as in Fig. 1.4. However, it is product consistency and stability, that will be needed to retain brand loyalty.

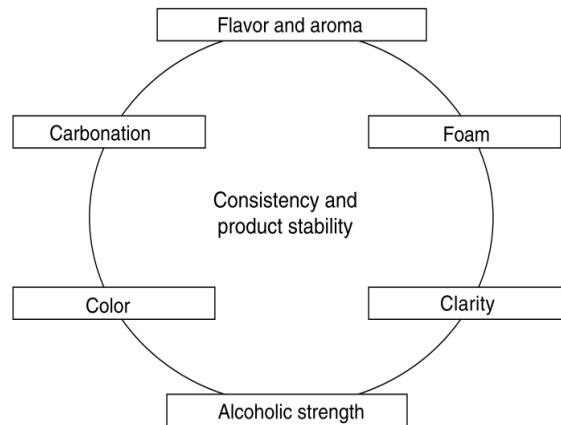


Fig 1.4. *Quality attributes of beer.*

The main beer parameters to be measured and controlled to ensure the quality (and brewhouse efficiency) targets are listed in Table 1.2.

Table 1.2. Main beer analysis methods as extracted from *Analytica (2010)*.

ANALYSIS	METHOD
Specific Gravity of beer	Pyknometer
Colour of beer	Spectrophotometric
pH of Beer	pH meter
Viscosity of Beer	Glass Capillary Viscosimeter
Haze in beer	Haze meters
β -Glucans	Enzymatic
Alcohol in beer	Distillation
Alcohol in beer	Enzymatic method
OG, Original, Real, Apparent Extract	Densimetric and distillation
Shelf-life of beer	Haze formation
Alcohol Chill haze in Beer	Test Chapon
Foam stability	Rudin Test

1.2.1 Haze

A clear packaged beer will lost its original brilliance after prolonged storage time, especially if it is kept under not optimal temperature. Beer marketing asks for more stable beers, to exert all possible options about sales and promotion without worry on stability and product.

Beer hazes are of the *biological* type or *non-biological* one. Turbidity of beer could have no effect on the sensorial quality. Infection of bright beer with either bacteria or wild yeasts will produce a biological haze due to the growth of the invading organisms, that makes the beer usually sour and unacceptable. Today, biological infection is fairly rare owing to the use of pasteurization and sterile filtration. However, after a length of time, sterile beers develop a non-biological haze. The rate of development of such hazes determines the shelf-life of bottled and canned beers. Before beer shows any haze at room temperature (20 °C), it may form a *chill haze* if cooled to 0 °C, that re-dissolves at 20 °C unless it remains, thus resulting in the so called *permanent haze*. Chill hazes are obviously more problematic with lager beers, being served at 4 °C, than with ales.

Biological haze

Biological haze is caused by microbial contamination and is avoidable with care and use of best practice procedures. Providentially, beer is an inhospitable environment for

microbial growth. It has a low pH (in the range of 3.8–4.5), ethanol is present in a range of concentrations of 3–6% (v/v), there is a limited range of nutrients, hop acids are present, the environment is anaerobic and the liquid is carbonated. Most potential contaminants originate from raw materials. Barley can contain *Fusarium* species that can release mycotoxins or cause gushing (that is, the phenomenon provoking the spontaneous formation of a great number of fine bubbles within the volume of beer that quickly ascend and flows out of the bottle, immediately after its opening).

Barley can also carry bacteria, which may supply nitrosamines and cause filtration problems. Thus, contaminants can cause flavor deterioration, turbidity and potential health problems. Most breweries pasteurize beer to ensure stability, but with good hygiene and efficient filtration the use of this expensive and potentially damaging process can be reduced. In modern brewing, non-biological haze is the key problem.

Non-biological haze

The non-biological haze isolated from beer has a complex composition:

Proteins :	57%
Polyphenolic substances:	29%
Carbohydrates:	12%
Minerals:	2%

There are three important types of non-biological haze:

- Chill haze
- Permanent haze
- Other hazes

Chill haze becomes visible when the beer is cold to below 0 °C, but it will disappear when beer is warmed to room temperature. Chill haze is formed from polypeptides and polyphenols, but unlike permanent haze (which is insoluble) it is due to a soluble complex involving non-covalent bonds (Siebert *et al.*, 1996). It will precipitate at low temperatures and form particles of 0.1–1 µm diameter. This will re-dissolve quickly if temperature rises. Chill haze can be partly removed from beer by filtration before packaging, provided that the beer is kept cold.

Permanent haze initially forms in the same manner as chill haze. However, the soluble complexes soon convert to insoluble complexes owing to the formation of covalent

bonds that will not dissolve after heating. The particle size is larger than that observed in chill haze, ranging from 1 to 10 μm diameter. The reactions are catalyzed by oxygen and metal ions and the amount of haze increases over time. The reaction is closely related to the amounts of haze-active polypeptides and polyphenols present (McMurrough *et al.*, 1992). Indeed, it has been known for years that beer proteins and polyphenols are responsible for both colloidal and foam stability. Beer contains 300–800 mg/L of raw proteins. Most of them are in the form of polypeptides with a molecular size of 5 to 100 kDa. Beer polypeptides originate primarily from barley. Much protein is broken down during malting into polypeptides and free amino nitrogen (FAN) or single amino acids by the activity of proteolytic enzymes. Only a small amount of protein is involved in beer haze: as little as 2 mg/l of protein is sufficient to cause a haze of 1 EBC (Chapon, 1994).

Polyphenols in beer originate from barley and hops. Polyphenols have been identified as having at least two roles in beer. Certain species are known to be responsible firstly for colloidal instability through binding with proteins, and secondly for flavor stability by protecting against oxidation (Whittle *et al.*, 1999). The chill haze isolated from three different breweries was between 1.4 and 8.1 mg/L, while the permanent haze accounted for 6.6–14.6 mg/L (Carrington *et al.*, 1972).

Beer contains from 100 to 300 mg/L of polyphenols (McMurrough and O'Rourke, 1997). These are also referred to as tannin, although not all polyphenols have the ability to tan animal skins. Two types are present in beer, the first group are derivatives of hydroxybenzoic and hydroxycinnamic acids, the second group are the flavanols and their derivatives (Hough *et al.*, 1982). Flavanols account for around 10% of total beer polyphenols, and this group includes the species related to colloidal instability. During mash boiling, polyphenols will precipitate with protein. After boiling more polyphenols are lost in cold break, and later in cold conditioning. Catechin, epicatechin, and complexed flavanoids are responsible for haze formation. Two dimers have been particularly associated with haze formation, that is procyanidin B3 (catechin-catechin) and prodelphinidin B3 (gallo catechin-catechin), their structures being shown in Fig. 1.5. These are referred to as proanthocyanidins (also known to brewers as anthocyanogens) and originate from both barley and hops.

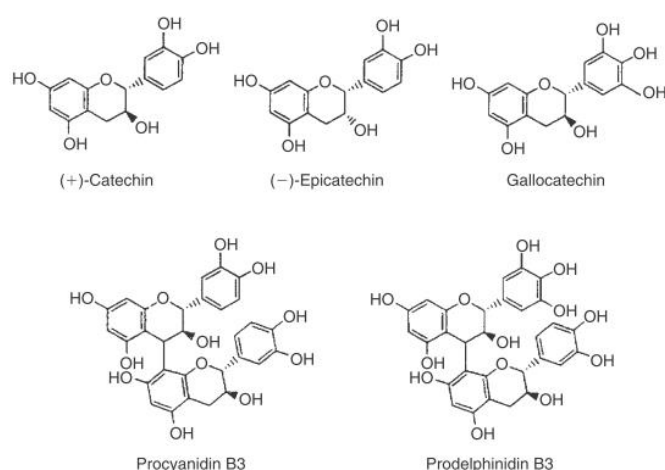


Figure 1.5 Structure of the main beer flavanol monomers and dimers (Siebert and Lynn, 1998).

The exact mechanism by which flavanoids bind to proteins and cause haze is uncertain. It has been proposed that the simple dimeric flavanols are too small to cause haze on their own and must polymerize into larger molecules (oligomers) before they are large enough to cause haze.



P and T refer to proteins and tannins, respectively

Protein binding is due to the number and position of the hydroxyl groups (-OH) on the flavanoid aromatic rings. Thus, rings with only one -OH group are almost inactive, those with two are more active, especially when they are adjacent (vicinal), rings with three groups are even more active (Siebert and Lynn, 1997). In this way, prodelphinidin B3 is more haze-active than procyanidin B3 since gallocatechin has three vicinal -OH groups, while catechin just two ones (Fig. 1.5). Gallotannins, such as tannic acid, have eight or nine gallic acid units, each with three vicinal OH groups, connected to a glucose unit (Siebert and Lynn, 1998).

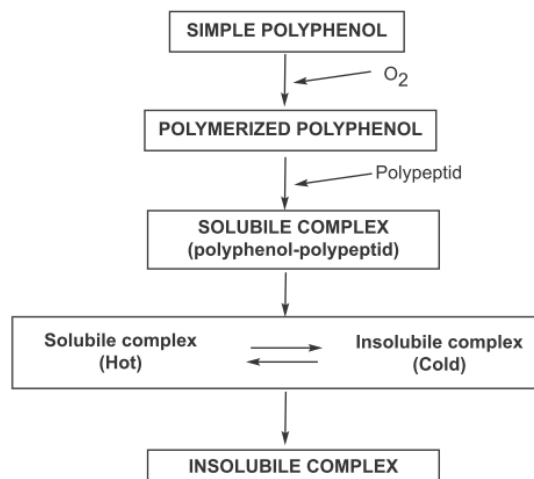


Figure 1.6 Haze formation in beer (Siebert *et al.* 1996b).

Siebert *et al.* (1996b) proposed a model for haze formation. Each polyphenol has two binding sites and each polypeptide has three binding sites. The greatest amount of haze will occur when the number of polyphenol binding sites is equal to the number of polypeptide binding sites. If there were more polypeptides than polyphenols, as in beer (Siebert *et al.*, 1996b), two polypeptides could be joined, but it would be unlikely that any further binding could occur due to the shortage of polyphenols. If there was more polyphenol than polypeptide, all the polypeptide binding sites would be filled with polyphenols, but again there would be little chance of further binding due to the shortage of protein.

The protein-polyphenol complexes are initially too small to scatter light; thus, some kind of reaction has to take place with the precursors before haze can form. Siebert (1999) proposed two pathways. Firstly, simple flavanoids polymerize to form tannins, which then combine with protein and grow to produce haze. Secondly, already complex phenolics are oxidized; then, these active phenols combine with protein and grow to produce haze (Fig. 1.6). The second pathway is supported by the finding that labeled oxygen in beer headspace was found to be incorporated in beer haze.

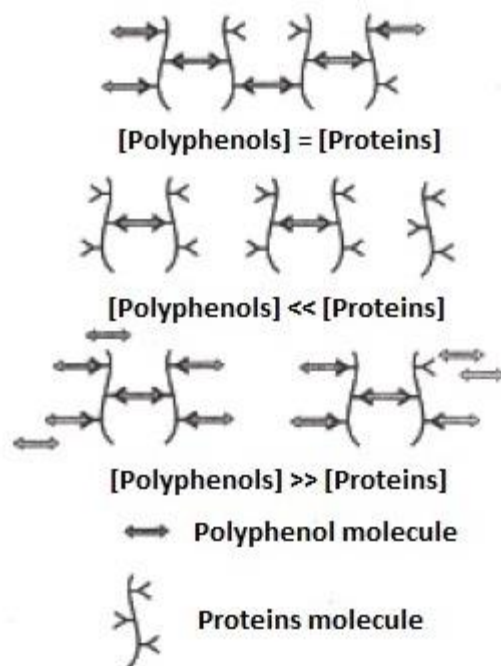


Figure 1.7 Interaction model of haze formation in beer (Siebert *et al.*, 1996a).

Raw materials are the source of haze precursors. In order to reduce the haze, some action can be taken on raw materials. Malt must be of good quality, in good condition and have a sufficient level of amylolytic enzymes. It is not certain if nitrogen modification of malt is related to protein stability. The use of adjuncts, such as syrup, rice and maize grits, dilutes haze-causing substances. Although haze-causing proteins are not significantly reduced, adjuncts derived from barley and wheat may increase the level of haze precursors. Wheat also increases the level of pentosans, but is good for foam stability. Malt with no proanthocyanidins (haze-causing polyphenols) is available. There are also some practical production issues to reduce the formation of haze in the final production: lower mash pH causes polyphenol precipitation, which is desirable as it removes haze precursors. Long run-offs extract more polyphenols, especially if the pH is allowed to rise. Excessive use of lauter rakes is as undesirable as sparging at too high a temperature. Shear damage as a result of transfer by piping and pumping, etc. should be avoided as this can contribute to haze. A good boil with agitation and turbulence is essential as it removes substances that could survive into the beer. Kettle fining removes substances that precipitate out of solution during wort cooling after boiling. Much protein is removed from wort as trub during boiling (hot break) and

during wort cooling (cold break). Apart from some loss of haze precursors in the cold break, little occurs during fermentation.

Cold conditioning is an important step of the process. Traditional brewing methods involved long maturation periods at 0-1 °C. Beer should be chilled as cold as possible without freezing to ensure precipitation of particulate matter. Filtration should be carried out at 0–2 °C after cold conditioning to allow particulates to settle. The beer should not be warm up at this stage to avoid chill haze particle dissociation and haze precursors release (Chapon, 1994).

Other hazes

These can result from substances different from proteins or polyphenols originating from raw materials, yeast, or process aids. Common constituents of such hazes are polysaccharides, such as glucans, mannans, starch and pentosans. The presence of β -glucan in beer causes filtration problems, as well as haze formation. These can be solved by using β -glucanase. Unfortunately, this practice is forbidden in some countries, like Germany in accordance with *Reinheitsgebot* law. In Italy, such a practice is allowed by the Ministerial Decree D.M. 19.11.1996 n° 68.

Haze was found to increase with increasing molecular mass and concentration of the added material. Shear stress, pH, maltose and ethanol also influenced haze formation. Haze caused by high-molecular mass β -glucans cannot be reduced by filtration or cold-conditioning.

Also oxalate, generally called *beer stone*, cause haze. It may form if the oxalic acid present in malt is not precipitated in the brew house because of low calcium levels.

Haze measurement

When light is passed through a suspension of a colored precipitate and the amount of reflection is negligible, the light absorption gives a measure of the turbidity according to Lambert's law. With white precipitates, such as beer haze, much of the light is reflected so it is measured the light reflected at a given angle to the incident light. Most of the commercial instruments in use today take measurements at 90° to the incident light. The wavelength of the incident light also varies between instruments (350-860 nm). Even if the EBC, IoB, and ASBC refer turbidity measurements to the same primary haze

standard (i.e., formazin, obtained by reacting hydrazine sulfate with hexamethylenetetramine), the scales adopted are different (Fig. 1.8):

- 10.000 ASBC Formazin Turbidity units = 145 EBC Formazin haze units
- 1 EBC haze unit = 69 ASBC haze units

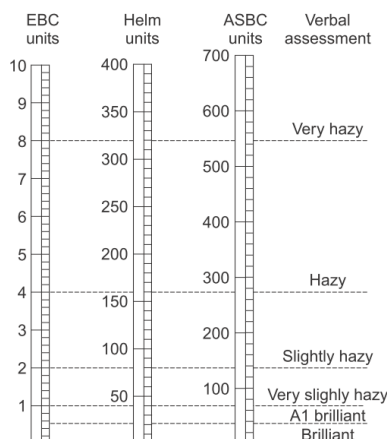


Figure 1.8 Comparison of haze scales (Analitica-EBC, 2010).

In most cases, visual assessment of beer haze correlates well with instrument readings for light scattered at 90°. However, some beers appearing bright to the eye give substantial meter readings and are said to contain *invisible* or *pseudo* haze. Such pseudo hazes are not observed with instruments using 13° forward light scattering.

Prediction assessment of haze stability in beer

A range of tests has been developed to assess the effectiveness of treatments. In general, there are two types of test available, that is the forcing or precipitative tests.

Forcing tests involve storing beer at elevated temperatures in an attempt to speed up the natural ageing process, that would occur if the beer were stored at room temperature. Thus, many months of storage can be reduced to a few days. Such tests are also known as *accelerated ageing*.

In precipitative tests, the chill haze is forced out on particular beer condition. Different brewing companies have their own preferred temperatures and incubation times with haze being measured before and after incubation. These procedures do not generally correlate with the results of actual shelf-life (Chapon, 1994), but are widely used as an indicator of beer stability or as a way of comparing different stabilizing products or dosing rates.

Colloidal shelf-life may be defined as the length of time before a beer displays a haze value of 2.5 °EBC (or 175 °ASBC) at 0 °C. There are two main types of tests: hot/cold cycling tests and precipitation tests (Lewis and Bamforth, 2007).

Hot/cold cycling tests

This test can be carried out in different way, just keeping the beer at 37 °C (1 week at this temperature is equivalent to 1 month of *normal* storage at 18 °C), or using different cycles of storage:

- at 60 °C for 2 days followed by storage at -2 °C for 1 day: (a single complete cycle is retained as equivalent to 6 weeks of *normal* storage),
- alternating 24-h cycles at 30 °C and 0 °C, a single complete hot/cold cycle approximating one month of *normal* storage.

Precipitation Tests

In the alcohol-chilling test (Chapon, 1994), ethanol is added to lower the beer temperature to -8 °C and chill haze is forced out within a total test time of 40 min. This test predicts only chill haze. Alternatively, haze-active proteins can be precipitated by the addition of tannic acid. Gallotannin can be replaced by a solution of saturated ammonium sulfate, and hence it is named as the *SASPL test*. In the first case, the amount of light scatter caused by a standard addition of tannic acid is measured. In the second, the number of milliliters of (NH₄)₂SO₄ added to cause a measurable increase in turbidity is recorded. The higher the salt needed to precipitate protein the lower the amount of precipitable protein in beer is. By using the polyvinylpyrrolidone monomer (PVP) as precipitant, tannoids can be quantified.

1.2.2 Flavor (oxidation and light)

The factors determining flavor robustness in beer are extremely complex, a myriad of chemical species combining to determine the rich complexity of beer aroma and taste. For the sake of simplicity, the first change in flavor is a perceptible decline in bitterness, and the beer may be perceived as harsh. There will also be a decline in fruity/estery and floral notes. Some beers will develop a ribes (blackcurrant buds, cat urine) aroma and most beers are claimed to develop a wet paper or cardboard character. Bready, sweet, toffee-like, honey, earthy, straw, hay, woody, winey and sherry-like are all notes that have been reported. Moreover, it is not entirely clear why displaying a pronounced age

character necessarily meet with disfavor in the marketplace. Stephenson and Bamforth (2002) also demonstrated that branding was of major significance in beer; however, when a given brand is identified there is a definite preference for the fresh version of that beer.

Achieving flavor stability is a major challenge, especially because the brewer cannot control what happens to the beer between packaging and consumption. As many as 600-700 substances contributing to beer flavor can be detected by the human taste and olfactory system, some at extremely low concentrations.

The extent to which a foodstuff, such as beer, will age in the marketplace can be described by resorting to the empirical formula given by Singh and Cadwallader (2003):

$$r_Q = \varphi(C_i, E_j)$$

where r_Q is the rate of quality deterioration, that depends on C_i , a generic compositional factor (e.g., content of reactive species, catalysts, inhibitors, pH, etc.), and E_j a generic environmental factor (e.g., temperature, light, mechanical stress).

Although such a formula makes no attempt to weight the various parameters, it is clear that a change in any one of them will impact flavor stability. Among the environmental factors, oxygen concentration is the most extensively studied. Its concentration in wort and beer increases with the partial pressure of oxygen above the liquid level, but reduces with increasing temperature and strength of the wort and beer. Oxygen is much more soluble (seven to eightfold) in organic solvents than in water. This has seldom been taken into consideration when considering oxidation in brewing systems. The oxygen concentration in a localized lipid environment (e.g. mash and trub particles) may be somewhat greater than in the bulk aqueous phase. Bamforth and Parsons (1985) firstly drew attention to the role of active oxygen species rather than ground-state oxygen in potentiating flavor damage in beer. The principle basis for the toxicity of oxygen is via its conversion to reactive free-radical forms, or reactive oxygen species (ROS), as they are often now collectively termed, because not all damaging species produced from oxygen are radicals.

On the route from barley to a beer in the customer's hand, apart from oxygen, the most dramatic impact on flavor stability is due to the temperature of beer storage. If the beer

temperature is raised by 10 °C, the rate of reactions, including those responsible for staling, increased from two- to three-fold.

Many flavor active compounds, present in uninfected beer, are capable of changing their levels during storage in the final package.

Prediction of flavor stability

Several researchers have developed forced ageing procedures. Typical regimes consist of holding beer at 60 °C for 24 h, 37 °C for 3 weeks, or 30 °C for 4 weeks prior to evaluation by tasting. These regimes are said to be good mimics for storage at 18 °C for 6 months. Another procedure involves shaking beer (to simulate transport, this factor being insufficiently considered in the context of flavor stability) and 4 days of storage at 40 °C. It is claimed that this is equivalent to storage at 20 °C for 3 to 4 months.

The main criticism to these accelerated regimes is that the flavors obtained at the higher temperatures are quite different to those developing during natural storage. As an alternative to tasting, some have advocated the chemical monitoring of species produced in forced ageing techniques. Thiobarbituric acid (TBA) has been used to measure carbonyl species produced on forced ageing, but TBA is not a very specific agent and preferentially reacts with malondialdehyde ($\text{HOCH}=\text{CH}-\text{CHO}$), that is but one of the breakdown products from unsaturated fatty acids. Another breakdown product is ethylene, which has also been cited as an indicator of staling potential. Other species are sometimes measured directly as *indices of staling*. Notable amongst these is furfural, which is not believed to directly contribute to staling per se, but it is felt that it is a good yardstick for oxidation. Others advocate the measurement of radicals by the application of Electron Spin Resonance technology (ESR). The endogenous antioxidant (EA) value is the time taken before an ESR signal is developed in an ageing test; the longer the lag, the greater the antioxidant potential of the sample.

1.2.3 Foam (stability and gushing)

Upon pouring a beer into a glass, the drinker will make judgments on the acceptability of the product, based solely on what his or her eyes are seeing. The presentation of the beer in the glass in terms of its foam head, clarity/brilliance and color are an anticipation for the perceptive drinker. Foam is perhaps one of the most appealing beer qualities. In

fact, the foam acts as an efficient gas exchange surface pitching aromas towards the drinker's olfactory sensors. Foam is also tactile to the lips and impacts mouthfeel through its stability and its structure (bubble size).

The beer foam quality is based on the interaction of various beer components. To understand the principles of beer foam physics, many authors have enucleated the following fundamental but interrelated events (Bamforth *et al*, 2008)

- Bubble formation and size
- Drainage
- Creaming (bubble rise or beading)
- Coalescence
- Disproportionation

Even if beer is supersaturated with carbon dioxide, bubbles will not form spontaneously unless nucleation occurs (Fig. 1.9), promoted by a particle, fiber or scratch in the glass or the dispense mode, be that tap or bottle. These nucleation sites should ideally be small to create smaller bubbles that create a foam appealing to the drinker (Bamforth, 2004).

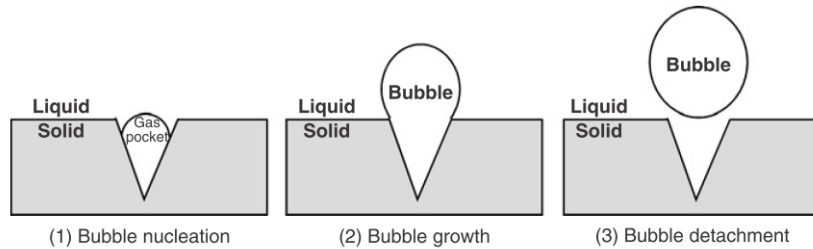


Figure 1.9 Schematic representation of the bubble formation sequence, (Ronteltap *et al*. 1991)

The factors governing the size of bubble that is generated in nucleation are described by

$$R_b = \left(\frac{3 R_m \gamma}{2 g \rho} \right)^{1/3} \quad (1.1)$$

where R_b and R_m are the radii of bubble and nucleation site (m), γ is the surface tension (N m^{-1}), ρ the beer density (kg m^{-3}), and g the acceleration due to gravity (9.8 m s^{-2})

The radius of the nucleation site is very significant, but surface tension and density are less important.

Upon its formation, foam is usually termed to be *wet*. The excess beer in the foam rapidly drains by gravity to produce a *dry* foam. In the dry foam, continued beer drainage by gravity and suction along the Plateau borders (Fig. 1.10) weakens the bubble film, eventually leading to bubble collapse.

Ronteltap *et al.* (1991) observed that the counteracting forces to drainage are viscosity of the beer, capillary effects and beer surface viscosity. The influence of beer viscosity is consistent with beer foam, this being more stable at lower temperatures. This explanation was more recently simplified to conclude that surface viscosity as opposed to bulk viscosity was most important (Bamforth, 2004). In practice, this interpretation appears to be confirmed by the observation that polysaccharides, such as β -glucans, increasing bulk viscosity have a negligible influence on beer foam stability (Lusk et al., 2001a).

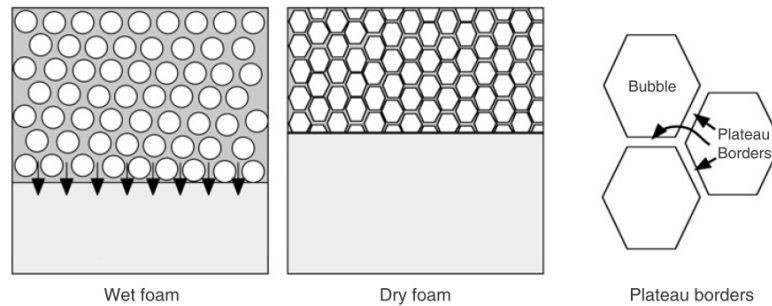


Figure 1.10 Schematic representation of foam drainage. (Prins and Marle, 1999)

Creaming or *beading* is defined as the formation of a dense foam, which should ideally be sustained for the duration of consumption (Bamforth, 2004). The nucleation activity, surface tension, beer density and CO₂ content determine the level of creaming. Accepting the central role of a nucleation site, it was found that CO₂ content was the most influential variable because the typical ranges for surface tension and density are usually not large enough to affect creaming.

$$a_n^0 = 3.11 C + 0.0962 \gamma - 218 \rho + 216 \quad (1.2)$$

where a_n^0 is the initial nucleation activity, γ and ρ are the beer surface tension and density, and C is the volumetric carbon dioxide content (vol. CO₂ /vol. beer).

Foam coalescence is defined as the merger between two bubbles caused by the rupture of the film between the bubbles to produce a larger, less stable and less appealing

bubble (Ronteltap *et al*,1991). However, if highly hydrophobic materials, such as lipid, oily snacks (i.e., crisps), lipstick, cleaning agent or dirty glasses, contact the beer, this effect can be catastrophic to beer foam stability. Commonly known as the *hydrophobic particle mechanism* (i.e., impact of lipids) or *particle spreading mechanism* (i.e., impact of detergents), such small disruptive particles, when positioned in the bubble film, rapidly initiate coalescence (Ronteltap *et al*,1991).

Disproportionation, also known as *Ostwald ripening*, is defined as the bubble fusion or foam coarsening process resulting from inter-bubble gas diffusion (Ronteltap *et al*, 1991; Bamforth, 2004). By this process, gas from smaller bubbles with higher Laplace pressure diffuse into larger bubbles with lower Laplace pressure. Thus, smaller bubbles disappear and larger bubbles become even larger, resulting in *bladdery* bubbles that are less attractive.

Disproportionation has been modeled according to the De Vries equation

$$r_t^2 = r_0^2 - \frac{4RTDS\gamma t}{P\theta} \quad (1.3)$$

where

- r_t = bubble radius at time t
- r_0 = bubble radius at the start
- R = the gas constant ($8.31 \text{ J K}^{-1} \text{ mol}^{-1}$)
- T = absolute temperature (K)
- D = gas diffusion coefficient ($\text{m}^2 \text{ s}^{-1}$)
- S = gas solubility ($\text{mol m}^{-3} \text{ Pa}^{-1}$)
- γ = surface tension
- t = time (s)
- P = pressure
- θ = film thickness between bubbles

Foam measurement

Foam measurement is problematic owing to its sensitivity to product composition (CO_2 , protein and alcohol content, etc.), cleanliness of glassware, and foaming technique used, including the diverse range of visual characters that determine foam quality. In all probability, the lack of a universally agreed assessment method derives from the necessity to measure the different components of foam quality, that is, stability, quantity, lacing, whiteness, creaminess, and strength. In addition, consumer dispense can broadly include pouring either from bottle or can packaging, or from a dispense tap, each mode having differing dynamics of foam generation. An expert assessment panel may give a ultimate verdict, but this procedure is expensive, awkward and subject to

biases based on gender, race and socio-economic background (Roza *et al.*, 2006). The most commonly used methods for commercial and research foam quality evaluation are the NIBEM (Klopper, 1973), Sigma head value (SHV), Rudin head retention (Rudin, 1997) or Ross and Clark that measure foam stability test (Ross and Clark, 1939).

Methods have been devised for measuring many foam parameters, such as foamability, foam stability, foam drainage, cling, viscoelasticity, lateral diffusion, film thickness and bubble size. Actually, most measurements concentrate on the rate of foam collapse or, inversely, the duration of head retention or foam stability.

The Institute of Brewing have adopted a modified method by Rudin (1957). As shown in Fig. 1.11, the apparatus consists of a jacketed foam tube with an inside diameter of 26-28 mm and a height of at least 350 mm, mounted over a porosity 3 glass-sintered disc. Degassed beer is added till the 10-cm mark and foamed with carbon dioxide to the 325-mm mark. As the foam collapses the time taken for the foam/beer boundary to traverse the distance between the 50-mm and 75-mm marks gives a measure of the half-life of the foam. The logarithmic rectilinear collapse of foams formed with carbon dioxide is four times faster than that for beers foamed with air or nitrogen. Thus, traces of air either in the CO₂ used for foaming or introduced into beer by pouring can cause departures from a regular logarithmic decay.

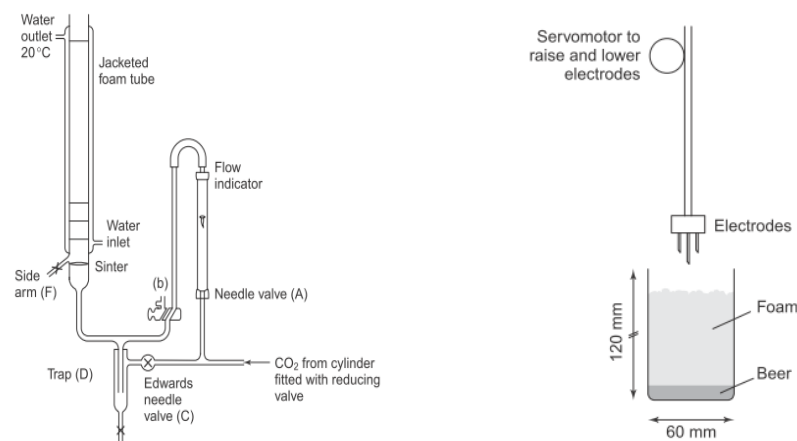


Figure 1.11 Rudin head retention apparatus and NIBEM.

The Institute of Brewing also approved the measurement of foam stability using the NIBEM meter, shown in Fig. 1.11 (Klopper, 1973). The foam is dispensed into a standard glass over which a meter head with a central electrode and four shorter needle

electrodes is mounted. When the needle electrodes are in the foam, the conductivity between the longer and shorter electrodes switches off the servomotor. As the foam collapses, the electric conductivity of the foam is reduced and the servomotor automatically lowers the electrodes until they newly contact the surface of the foam. For a beer containing >3.4 g/L CO_2 , it is measured the time for the foam to collapse over 10, 20, and 30 mm. For a beer containing <3.4 g/L CO_2 , the collapse is measured over 5, 10, and 15 mm. The IoB Analysis Committee (Sharpe, 1997) found that the precision of both the Rudin and NIBEM methods was independent of the foam stability of the sample. The repeatability and reproducibility of a new NIBEM-T meter has been improved, by protecting against air movement and allowing automatic temperature compensation. Thus, it has been accepted by the EBC Analysis Committee (Ferreira, 2003).

Glemister and Segel (1964) proposed a method to evaluate the tendency of beer foam to adhere to a glass. This simple procedure evaluates the foam through the concepts of *primary* and *secondary clings*, by pouring the beer at 5°C into a glass in such a manner that about a 2-in head of foam is produced. Such foam head is allowed to collapse for a 5-min period. The cling deposited during this time is termed the primary cling by means of suction, the level of the beer in the glass is lowered by 2% at 1-min intervals so that four or five possible rings of *secondary cling* may form (Fig. 1.12).

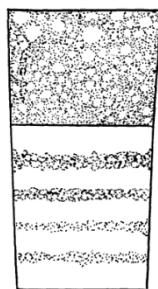


Figure 1.12 Representation of the primary and secondary cling. (Glemister and Segel, 1964)

1.2.4 Color

The color of beer is largely due to the melanoidins and caramel present in the malt and adjuncts used and caramelization that can take place during wort boiling. Minor adjustments of the color of beer can be made by the addition of caramel either to the

copper or with the primings. Other contributors to the color of beer are oxidized polyphenols, especially in the presence of trace metals, such as iron or copper. In pale beers the yellow vitamin riboflavin may significantly contribute to the color. The measurement of the color of worts, beers or caramel solutions is difficult because the absorption spectra show no maxima. As long ago as 1893, Lovibond developed a series of colored glass discs to match against the color of beer. In 1950, a new series of glass discs (2-27 EBC color units) were released and adopted by the EBC. The Standard Reference Method (SRM) was adopted in 1950 by the American Society of Brewing Chemists, which had recognized the need for an instrument-based measurement of color unburdened by the difficulties of the Lovibond system (visual comparison). Spectrophotometric measurements at one wavelength were thus adopted. Analytica-EBC chose measurements at 430 nm in a 1-cm cell to express the EBC color as

$$\text{EBC Color} = 25 A_{430} \quad (1.4)$$

Originally, the Institute of Brewing chose measurements at 530 nm, as being more suited to ales, but later shifted to 430 nm. The ASBC also take measurements at 430 nm to express the ASBC color as:

$$\text{ASBC Color} = 10 A_{430} \quad \text{in a } \frac{1}{2}\text{-in cuvette} \quad (1.5)$$

$$\text{ASBC Color} = 12.7 A_{430} \quad \text{in a 1-cm cuvette} \quad (1.6)$$

The ASBC and EBC measurements are now identical (both done at the same wavelength and in the same size cuvette), but the scaling is different. A photometer or spectrophotometer is used to measure the attenuation of light at 430 nm as it passes through a sample of beer contained in a standard 1-cm by 1-cm cuvette. The absorption is the log of the ratio of the intensity of the light beam entering the sample to the intensity leaving. This difference is multiplied by 12.7 in the SRM system and 25 in the EBC. For example, if the light intensity leaving is one hundredth the light intensity entering, the ratio is 100, the absorption (A_{430}) is 2 and the ASBC color is 25.4.

When the ASBC color for a beer or wort is larger than about 30, the log linear limit of some instruments using 1-cm cuvettes is approached. In such cases, the sample is to be diluted with deionized water. By using the Beer-Lambert law again, the ASBC color is

$$\text{ASBC Color} = 12.7 D A_{430} \quad (1.7)$$

where D is the dilution factor (D=1 for undiluted samples, D=2 for 1:1 dilution, etc.) and A_{430} the absorbance at 430 nm in 1-cm cuvette.

The EBC system of color measurement is similar to the ASBC one and the EBC color is approximately twice the ASBC color and this applies at any color depth. The agreement between ASBC color and Lovibond is fair for pale beers (10 °L~12.7 ASBC), but worsens for darker beers or worts (40 °L ~ 53.4 ASBC).

Both systems demand that the beer be free of turbidity prior to the measurement at 430 nm. In the ASBC method a second measurement is taken at 700 nm. If the absorption at this wavelength is less than 0.039 times the absorption at 430 nm, the beer is considered turbidity-free. If not, it is to be filtered or centrifuged and the reading repeated. If the ratio test is not passed after clarification; then, the beer does not have *average spectral characteristics* and, technically, is not qualified to be characterized by the ASBC method. In the EBC system the beer is required to be filtered if its turbidity is more than 1 EBC turbidity unit (equivalent to 1 FTU). No absorption measurement is made other than at 430 nm.

1.2.5 Viscosity

Dynamic viscosity is defined as the resistance to shear flow. Kinematic viscosity is a measure of the time taken by a liquid to flow through an orifice under gravity. Both are measured by an international method using the time of flow in an Ostwald viscometer at 20 °C. At this temperature the dynamic viscosity of water is 1.002 mPa s, and the kinematic viscosity is 1.00 cS (centistokes) or $1.00 \times 10^{-6} \text{ m}^2 \text{ s}^{-1}$.

The EBC method refers to a 20% (w/v) sucrose solution as an additional standard (1.945 cP), but the Institute of Brewing, use a 3.0% (w/v) solution of glycerol. Some typical values are:

worts (SG 1.030-1.100)	1.59	mPa s
lager (SG 1.007)	1.45	mPa s
stout (SG 1.009)	1.96	mPa s.

The concentration of the main components of beer, as well as that of the malt derived beer potentially influencing microfiltration efficiency, is reported in tables 1.3 and 1.4 respectively.

Sthwart (1998) reported that the viscosity of beer brewed from 24 malt samples varied considerably, ranging from 1.33 to 1.69 mPa s, with an average value of (1.45 ± 0.13)

mPa s. Both β -glucan and arabinoxylan contents in beer were positively correlated to beer viscosity ($p < 0.001$ and $p < 0.01$, respectively), the latter being significantly correlated to membrane filtration (Sthwart; 1998). Sadosky *et al.* (2002) found that arabinoxylan, β -glucans and dextrans increased the viscosity of model solutions with dextrans having the largest effect.

Table 1.3 Concentration of the main components of beer (Sadosky *et al.* 2002).

Molecular Fraction	Concentration [g/L]	Molecular weight MW [kg/kDa]
Dextrins	25-35	50-200
β -Glucans	0.07 -0.5	50-200
Pentosans	1.5-3.5	50-200
Proteoses, albumoses, peptose	0.06-0.2	50-200
Peptides	0.1-0.5	1.5-5
Amino acids	0.02-0.1	<5
Polyphenols	0.02-0.06	1-5

Table 1.4 Malt derived beer constituents potentially influencing on microfiltration efficiency (Sthwart, 1998).

Parameter	Range	Average	SD
β -Glucan (mg/L beer)	0.19-318	72	78
Arabinoxylan (mg/L beer)	0.35-97	26	26
Viscosity (mPa s)	1.33-1.69	1.45	0.13
Protein (g/L beer)	0.65-1.3	0.96	015
Polyphenol (mg/L beer)	45-106	72	18

1.3 BEER PRODUCTION AND CONVENTIONAL FINISHING OPERATIONS

Beer production consists of four chemical and biochemical process (mashing, boiling, fermentation and maturation) and three solid–liquid separations phases (wort separation, wort clarification and rough beer clarification). Finally, the clarification of rough beer and pasteurization of filtered beer represent the most important operations in the brewing process to obtain a clear, bright and microbiologically stable beer. A schematic view of the beer productions process is shown in Fig. 1.13.

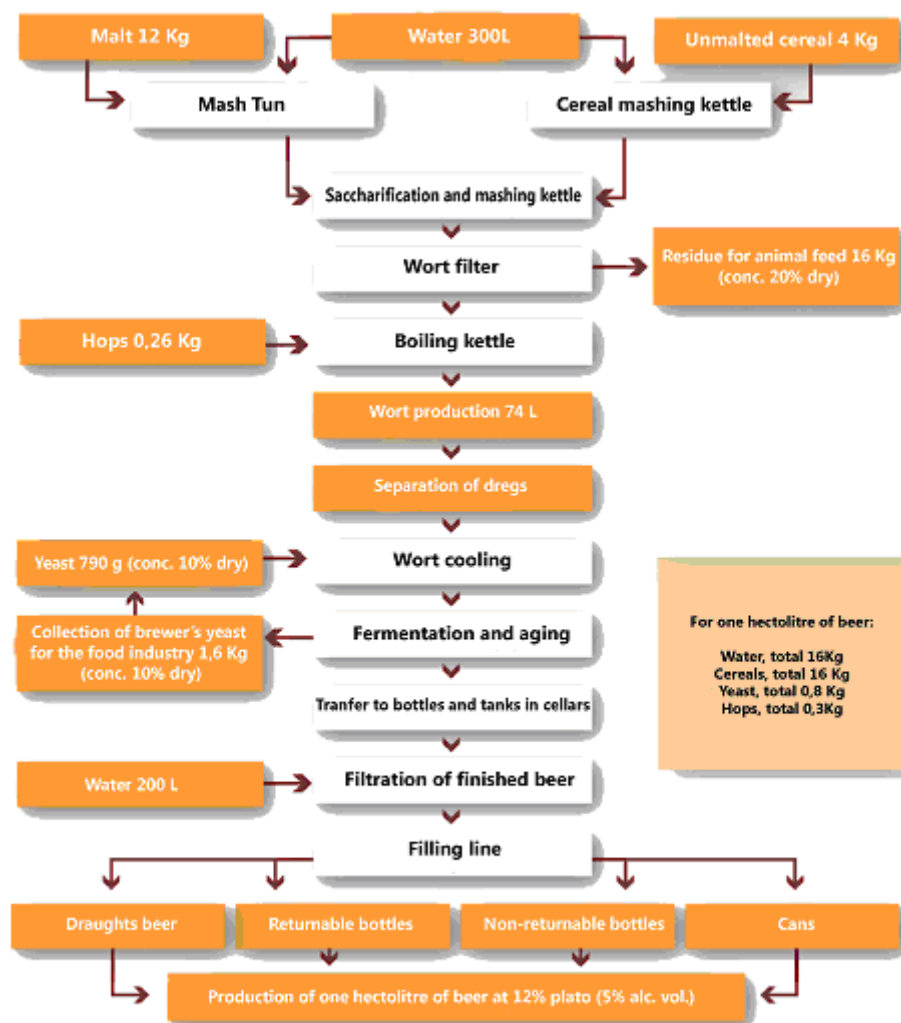


Figure 1.13 Block diagram and rough material balance of beer production (Assobirra , 2012).

Considering the 6,000-year old history of brewing (Fig. 1.14), the filtration of beer is quite a new development. Brewing beer during the 1800s was a local production. Individual villages and towns could have as many as twenty local breweries. By 1850, large cities, like Munich, Bamberg, Prague and New York, had hundreds of breweries operating. The main limiting factor for brewery growth was the short shelf-life of beer, allowing only a distribution time of 3 to 10 days before the beer showed a sharp drop in quality. This poor quality was primarily due to yeast autolysis and beer spoiling organism, and protein/polyphenol complex formation.



Figure 1.14 Egyptian Ushabti, needed to ferment beer and baking bread after the dead.

Brewers became able to produce a beer that was free from yeast and, even more important, free from spoilage bacteria, with the introduction of filtration. Beer quality became more reliable and constant as the consumer enjoyed a range of beer brewed in various regions. The first beer filter was presented at a brewing exhibition in Munich in 1880 by the German developer *Lorenz Adelbert Enzinger* (Fig.1.15).

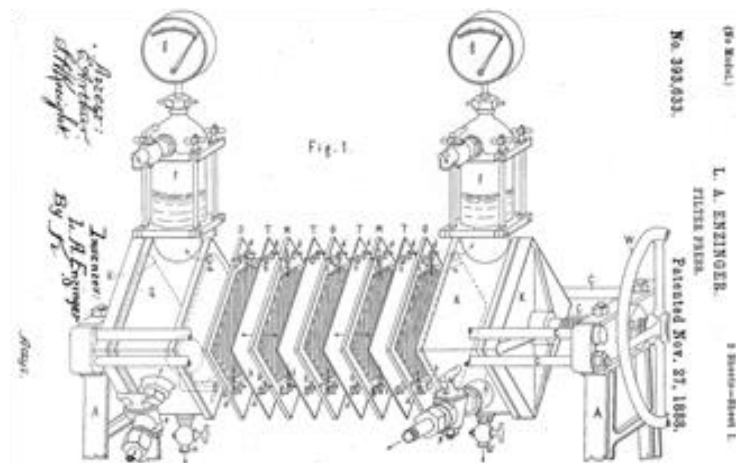


Figure 1.15 Schematic draw of the first beer filter as patented by Lorenz Adelbert Enzinger.

The filter was designed in a horizontal configuration, with individual plates having inlet and outlet channels, similar to modern plate-and-frame filters. The plate material was

black iron, while valves and meters were made of copper and brass. Filter media was made of paper leaves, which had to be changed after each filtration. The next generation of beer filters was developed early in the 1900s, again by Enzinger. This filter had a vertical design configuration aid, used brass bowls (*Enzinger Schalen*) to house pulp cakes. Pulp cake was prepared from cotton fibers mixed with asbestos. The main advantage of pulp filtration was the possibility of re-using the material for 3 - 5 filtration batches, reducing labor and downtime. Once spent, the pulp cake was removed from the brass bowls, rinsed and reprocessed in a pulp press to make new cakes. Pulp filtration was the first recycling technology used in a brewery; however, it required high water consumption and labor.

At the world exhibition in Paris in 1889, it was possible to taste filtered beers from all over the world: a global market was created. Pulp filtration dominated the brewing industry until 1930, when the first Diatomaceous Earth (DE) filter was proven in the US. Shortly after World War II, USA, English and Japanese breweries adopted the process and made DE filtration a standard unit operation for primary beer clarification. The first design for DE filters was based on a plate-and- frame filter using support sheets. Schenk later introduced the horizontal leaf-filter and Enzinger developed the candle type DE filter. Despite these three filters has been highly improved in respect to the early models, they still remain the most common platforms for primary filtration in breweries.

Today breweries use filters (and aids) also to remove proteins, polyphenols, short- and long-chain carbohydrates, and other substances that cause beer haze. These operations must comply with the haze specification of a lager beer in order to produce a clear bright beer accordingly to the *European Brewery Convention norm*, (Analytica EBC, 2010). Many of the beer products are now aimed at a global market. The centralization of brewing capacity into fewer, larger breweries according to the principles of the economy of scale reduces the production costs, but need an extended distribution networks. Many brewing companies of all sizes have long distribution chains, and the subsequent increase in the interval between production and consumption has put pressure on brewing companies to provide beers with longer shelf-lives, that can withstand temperature changes (storage and retail), flavor damage from ultraviolet light (notably retail) and agitation (transportation).

The need to achieve the required, labeled shelf-life is absolutely of fundamental importance to the company. No brand recognition or marketing expense will make up for a customer receiving a cloudy or poorly flavored beer. For the consumer, beer should be a drink with a pleasant flavor, an attractive color and clarity and should contain sufficient gas to carry the aroma and characteristic foam. The main physical properties assessed by a beer drinker (that is, color, clarity, viscosity and foam) are controlled by the filtration process.

Today, the brewer has several options for major processing operations as reported below.

1.3.1 Filtration techniques

Generally, the filtration steps fulfill two aims:

- To remove suspended materials from the green beer
- To remove potential turbidity-former compounds (stabilization).

Beer clarification is aimed at obtaining a colloidal stability. Filtration in breweries is most commonly accomplished by using filter aids. These substances, used as powders, form incompressible and highly porous filter beds, thus allowing the relatively free flow of beer. In industrial brewing process, there are usually four main clarification stages, as listed in Table 1.5:

- Primary filtration to remove solids and bulk yeast from beer.
- Trap filtration to remove DE or other process additives.
- Fine filtration to remove residual yeast cells and fine particulates that could foul a final membrane filter.
- Final membrane filtration to remove bacteria and yeast, that could spoil the packaged beer.

Table 1.5 *Main stages of industrial beer filtration.*

STAGE	DESCRIPTION
Primary filtration	0.5 - 30 µm nominal DE (0.65 µm cut-off cross-flow membranes)
Trap filtration	5 - 10 µm pleated depth cartridge
Fine filtration	1.5 µm pleated depth cartridge
Final filtration	0.65 - 0.45 µm microbial rated membrane cartridge

Primary filtration

Primary filtration is the clarification process to remove bulk yeast from fermented beer. The typical inlet turbidity range from 60 to 120 EBC, with yeast cell count of about 10^7 cells/mL.

Sedimentation alone cannot provide the level of beer clarity required for the next processing stages.

This stage helps to remove also haze forming materials, such as protein-polyphenol conglomerates and hop resins prior to downstream filtration.

Beer clarification is carried out via DE (or cross-flow filters) while, stabilization of beer is carried out using PVPP (Polyvinylpolypyrrolidone) and/or silica gel to remove polyphenols and/or protein, respectively.

DE filtration starts by pre-coating a support medium (e.g. a screen) using a coarse filter-aid suspended in water or beer. Cross-flow filtration for primary clarification often requires the use of a centrifuge in order to remove yeast and large particles and thus limit membranes fouling.

Trap filtration

This filtration is employed in breweries to ensure a bright filtrate, free from filter aids. Typical inlet haze to the trap filters should be <0.8 EBC or approximately 1 NTU. Generally, beer will have some particles in the range of 2-100 µm after DE filtration, when the average particle size 30 µm.

Depth filters are well suited to remove particulates with a broad particle size distribution. Depth cartridges, sheets or modules utilize a dense filament matrix, which can consist of inorganic materials, such as glass, cellulose, polymers, or stainless steel.

Pores throughout the filter medium may be graded or symmetric. Particles are retained on the surface of the filter, as well as throughout the depth of the medium.

Fine/final filtration

Fine and final filtrations are the preliminary steps prior to packaging. In breweries, where a three-stage system (trap, fine, and final) is employed, the fine filter serves also as the pre-filtration step to protect final membranes. It is common for smaller craft breweries to utilize depth filters, that serve as trap and fine filters for polishing effect and bio-burden reduction, without a final membrane in service.

Target microorganisms for pre- and final-filtration are yeast and the common beer spoiling bacteria detailed below (Table 1.6). Filters employed to retain these microorganisms from beer are not considered to be sterilizing-grade. Sterilizing grade specifically refers to a membrane filter cartridge that removes $>10^7$ *Brevundimonas diminuta* per cm² of filter medium, according to the FDA definition (FDA Guidance for Industry, 2004). Thus, final beer membrane filters (equipped with 0.65 to 0.45 µm rated membrane cartridges) provide a reduction of microorganisms, and not necessarily a sterile product.

Table 1.6 Main beer microbial morphology and approximate dimensions. (Nguyễn *et al* 2012)

Microrganism	Morphology and approx. dimensions
Bacteria: (<i>i.e</i> <i>Pediococcus damnosus</i>)	0.6-µm short rods, clumping
Bacteria: (<i>i.e</i> <i>Lactobacillus brevis</i>)	0.5-µm x 2-µm rods, clumping
Bacteria: (<i>i.e</i> <i>Lactobacillus lindneri</i>)	0.5-µm x 2-µm rods, mostly single cells
Yeast: (<i>i.e</i> <i>Saccharomyces sp.</i>)	~3 µm

Model organisms are often utilized to establish microbial removal ratings. For example, *Saccharomyces cerevisiae* is often utilized as a model for 0.65-µm rated membrane filters. *Serratia marcescens*, similar in size to *Escherichia coli* and *Lactobacillus sp.*, is often utilized to qualify 0.45-µm rated membrane cartridges. Alternatively, specific beer spoilage organisms may be used to qualify membrane final filter performance.

1.3.2 Kieselguhr clarification

The most common filter-aid used in breweries is kieselguhr or diatomaceous earth (DE). These materials comprise of fossils or skeletons of microscopic salt or freshwater life, known as diatoms. They die, and form deposits, often several hundred meter thick, that are mined. Crude kieselguhr contains up to 60% water. It is firstly crushed and then pre-dried. After calcination in a rotary furnace at temperature between 800 and 1200 °C, the material is size-classified to give kieselguhr of various grades (Fig. 1.16). Fig.1.17 shows a microscopic image of DE

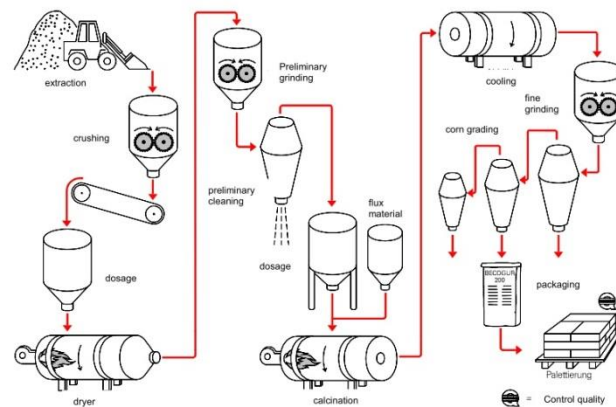


Figure 1.16 Schematic diagram of Kieselguhr production scheme. (Technical Information, www.begerow.de)

Kieselguhr clarification represents the reference standard for the filtration process of beer. It is usually selected to filter beer for the following reasons:

- Low price
- Ready availability (multiple sources)
- Excellent filtration results
- High throughput per filter area
- Good adjustability/flexibility.

The suspended solids in beer show unfavorable properties with regard to cake formation, these being fine, soft, compressible and, in part, gel-like. For this reason,

kieselguhr is used as a filter-aid, which, on the one hand, forms a layer permeable for the filtrate and, on the other hand, functions as a loose filter cake.

Nearly 99% of the beer produced today is filtered through kieselguhr, or diatomaceous earth, which cannot be recycled (Alfa Laval, 2007). Kieselguhr addition of 50 g/hL can be found in practice. The dosage generally depends on the practical experience of the master brewer. For the conventional dead-end filtration with filter-aids there is a consumption of diatomaceous earth that can be as large as 1–2 g/L of clarified beer.

At the end of the separation process, diatomaceous earth sludge has more than tripled in weight, being very rich in water and organic substances.

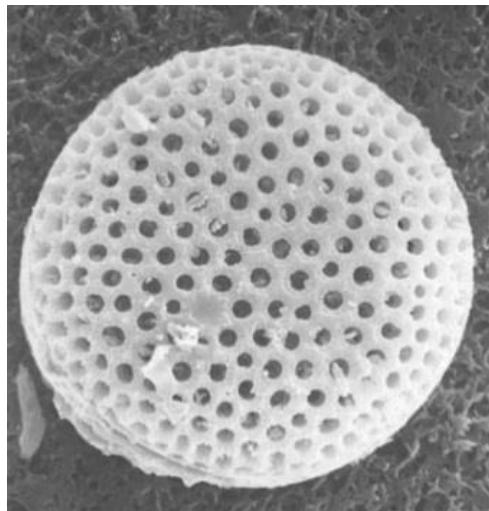


Figure 1.17 *Microscopic view of diatomaceous earth.*

Fundamentally, the method is the same for all process possibilities available:

- A one step pre-coating with perlites, cellulose, kieselguhr or a mixture of these filter-aid materials.
- Filtration with continuous filter material addition.
- Emptying of the filter.
- Removal of the filter cake.
- Cleaning and disinfection.

A consistent filter cake build-up is essential for the filtration efficacy. This is achieved by recycling a water-rich filter-aid slurry around the filter. After several minutes, the precoat is completely deposited onto the filtration surface to ensure efficient filtration of

the early part of the beer run, to guarantee the integrity of the filter throughout the run and to aid removal of the filter cake after the process cycle.

The primary pre-coating takes place with a total superficial solid loading of 1000 g/m²; the ongoing dosage lies between 50 g/hL for well-filterable beers and 300 g/hL for very poorly filterable ones. Primary pre-coat is added together with degassed water; the water is pressed off the filter with beer. The filter-aid slurry is added continuously to the flowing beer stream. Thus, the filtration surface is constantly being regenerated. In this way the filtration run time is extended, causing the process to be commercially viable. At the end of a filtration run, the beer is pressed off the filter with water. In this case, pre- and post- runs may be used to dilute stronger brewed beers.

The filtration performance is challenged by the intrinsic difficulties of beer as a product for filtration. The low temperature (0 °C) and presence of dissolved solids and alcohol result in a quite high viscosity (about 2 mPa s). Of even more significance is the nature of the suspended solids. These may be present in very high levels, perhaps up to 0.2% by volume or even higher over short periods during tank run-off. Practically, all of the suspended beer solids are compressible, this resulting in filter cakes impermeable to beer flow. Filtration may also be impaired by colloidal substances, such as glucan gels. By lowering the beer temperature, more cold trub and chill haze will form. Thus, beers emerging from maturation are to be maintained at 2 to 1 °C, through filtration. The turbidity of the beer leaving the filter must be <0.5 EBC. With good filter practice losses of color, extract, bitterness and foam potential should be minimal. Care must also be taken to avoid oxygen pick-up after maturation and during the filtration process. At the end of maturation the oxygen concentration should be <0.01 mg/L.

Three technical possibilities are available for the use of filters:

- Frame filters .
- Horizontal leaf-filters.
- Candle leaf-filters.

Plate-and-frame filter. This equipment is widely used in Europe. It consists of several vertically plates, each one covered with a cellulose filter-sheet folded at the top of the plate so that both sides of the plate are covered. Each plate alternates with a frame that will receive successively the pre-coat and beer plus body-feed. The beer passes through the kieselguhr bed, then the sheet and through the holes in the plate to be discharged

(Fig. 1.18). The sheets are washable and have a long life. After filtration, the plates are separated and the kieselguhr is dislodged from the sheets by spraying. In some plate-and-frame filters the sheet is protected by a disposable cellulosic *nappy liner*, which has a limited effect on the effective pressure difference, but prolongs the life of the sheet and aids kieselguhr disposal.

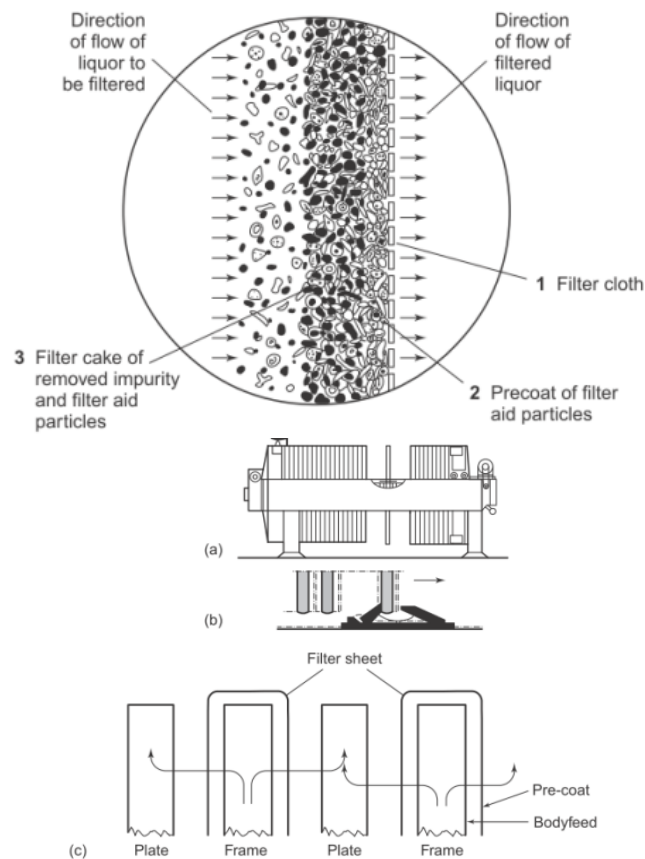


Figure 1.18: Schematic views of the plate-and-frame filters (Hough *et al*, 1982).

Leaf filters. Leaf filters (or screen filters) have a series of stainless steel leaves fitted either vertically or horizontally inside a filter body (Fig. 1.19). In a vertical leaf filter both sides of the support are coated with filter aid whereas in the horizontal leaf filter only one side is coated.

Candle filters. A candle filter is a cylindrical, vertical pressure vessel containing many filter elements (Fig. 1.20). Each element consists of a rod of Y cross-section around

which annular discs are stacked. The discs are made so that liquid can penetrate between them and then flow along the channels between the holes in the discs and the recesses of the Y-section rod. Kieselguhr powder builds up between adjacent filter discs to provide the surface area for depth filtration. Between 500 and 700 candles can be arranged in a cylindrical housing to create a very large filter.

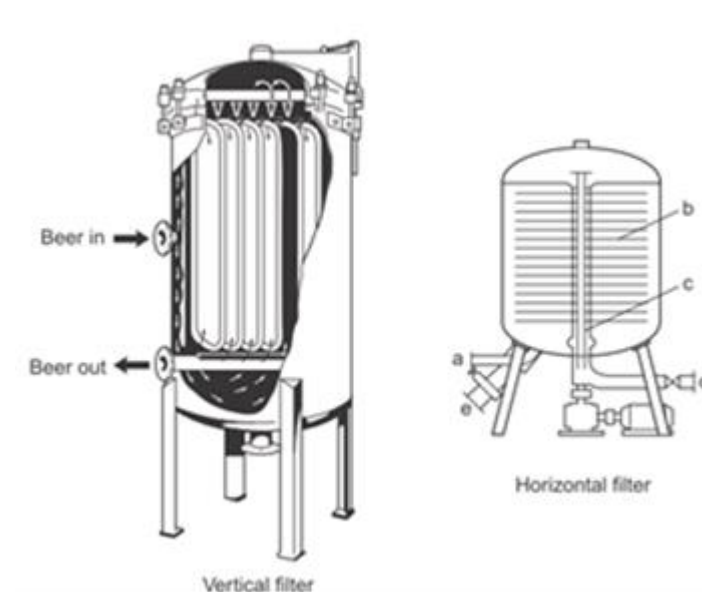


Figure 1.19: Schematic views of vertical (a) and horizontal leaf filters (b) .



Figure 1.20 Photo and schematic view of a candle filters.

The main problems associated to diatomaceous earth use

Diatomaceous earth (DE) has various advantages for filtration in brewing process. Thus it has been the standard industrial practice for more than 100 years and is continuously scrutinized from economic, environmental and technical standpoints. The World Health Organization defines the crystalline silica present in DE as a cause of lung disease (WHO, 2000). From the environmental point of view, diatomaceous earth is recovered from open-pit mines and constitutes a natural and finite resource. After use, recovery, recycling and disposal of DE after filtration are problematic due to their polluting effect. From the health perspective, DE is classified as *hazardous waste* else as such or after use it asks for ensuring safe working conditions.

The DE consumption and sludge disposal generate the main cost of the filtration process (Fillaudeau, *et al.* 2006). In Europe, the economic aspect is strengthened because its consumption is high, around 1.7 g/L of clarified beer. The disposal routes of DE sludge are into agriculture. Its recycling cost are of the order of 170 €/Mg⁻¹, but disposal costs widely vary from one brewery to another with a positive income of € 7.50/Mg up to a maximum charge of 1100 €/Mg⁻¹ of DE purchased (Knirsch *et al.*, 1999). DE regeneration offers another possibility to reduce the consumption and disposal of DE. When a horizontal leaf-filter system is employed, brewers have the possibility to add a batch recycling system, where spent DE is treated with caustic to dissolve foulants and then acid to neutralize the pH. The process only minimally affects the DE structure therefore, spent DE may be re-used for multiple filtration batches (<http://probrewer.com/resources/filtration/applications.php>)

Alternative filter-aids

Today, alternatives are currently in use to reduce DE use and its environment impact.

Perlites consist of thermally expanded volcanic glass crushed to form microscopic flat particles. These are less efficient filter-aids than DE, but are perceived as safer.

Cellulose fibers may be employed as coarse pre-coats or with the body-feed slurry. Complete replacement of DE might be achievable by using a mixture of such fibers with silica gels as body-feed slurry. This would offset disposal costs since the exhausted materials may be disposed with spent grains. The purchase price is significantly higher

than DE one, but this is compensated by the lower disposal costs (Bonnacchelli *et al.*, 1999).

Another alternative to DE is rice hull ash, a by-product of the food industry (Villar *et al.* 2004). Hulls are removed from rice during milling and are incinerated. It is possible to treat the resulting particles to minimize contaminating ions leaching into beer. About 50% replacement is recommended, even if it is unlikely to achieve the same standard of clarity with any beers.

Among the synthetic, re-generable filter-aid filtration systems proposed, it is worthy citing a synthetic polymer available in granular form with a typical particle size of 35 µm larger than that of the DE grades normally employed.

Today, used DE (Leeder *et al.* 2011) may be disposed of:

- by converting the organic fraction present in waste kieselguhr into biogas, and the inorganic residues as soil improvers;
- as a component in building bricks;
- by mixing with spent grains as cattle feed or by burning in a fluidised bed.

1.3.3 Stabilization techniques

Beer stability refers to various aspects: foam, microbiological and colloidal, and flavor stability. Microbiological stability is achieved through the removal of active yeast cells and contaminant microorganisms. Colloidal one is achieved through the removal of large particles, especially flocs formed by the coagulation of polyphenolic and proteinaceous substances. Flavor stability is achieved through retaining flavor compounds, while minimizing dissolved oxygen in clarified beer. Apart from filtration, various other treatments may be applied to enhance the shelf-life of the product and remove haze constituents. Removal of chill haze by filtration lengthens shelf-life, because chill hazes will eventually form stronger covalent cross-linkages that make the haze permanent. The two main contributors to haze instability for a given beer are dissolved oxygen during process and in pack, and initial haze. As the haze is a product of protein and polyphenols, removing or reducing the concentration of either component from beer will prevent haze formation.

The sensitive protein fraction is characterized by a significant presence of the amino functional group (-NH). Polyphenols exhibit more haze-forming potential when they form more oxidized states and polymerize into dimers, trimers, etc. Several techniques are used for this purpose. Current technologies for stabilization include PVPP, silica gels, tannic acid and papain. The longest shelf-lives are achieved with employment of more than one stabilizer.

PVPP (*polyvinylpolypyrrolidone*) is an insoluble polyamide that contains the same -NH functional group as haze-sensitive proteins. Thus, it adsorbs the polyphenolic fraction that contributes to haze. It is possible to employ PVPP (or the less cross-linked PVP) as a one-off addition to beer storage or filtration. More commonly, however, it is used and regenerated. A dedicated vessel (similar to a leaf- or candle-filter) is employed to disperse PVPP into the beer stream. Regeneration is possible with caustic.

Silica gels are sold as xerogels or hydrogels, the latter having more water in the particles. They consist of particles with a highly porous structure enabling a high adsorption area. They selectively adsorb haze-active proteins and poly-peptides. At the dosage rates employed they have no perceptible effect on beer foam (which relies on protein for its structure). Silica gels are not regenerated; they are employed in beer storage or as an addition (at 0.3 ± 0.8 g/L of beer) to the filter aid.

Tannic acid (or gallotannin) precipitates haze-forming protein. It also precipitates significant proportions of transition metal ions and lipids. These components adversely affect beer stability, the former through formation of free radicals and the latter through oxidation to stale-flavored compounds. A large dosage of tannic acid may cause a large amount of sediment, resulting in loss of high quality product.

Cold stabilization

The most effective beer treatment with respect to haze stability is the cold storage of the beer. The formation of the hydrogen bonds in protein-polyphenol complexes is not very rapid, and the effect of temperature is more important than that of time. The ideal conditions for haze formation are -1 to -2 °C for 2-3 days. Normally, the beer undergoes a period of cold stabilization for not less than seven days at 1 to 2 °C. This technique allows a reduction in the cost of other beer treatments designed to remove potential haze-forming proteins and polyphenols. At higher temperatures the breakdown rate of hydrogen bonds increases rapidly.

Pasteurization

Microbiological stabilization of beer by thermal treatment is the most common method. Alternatives are sterile filtration as described above and inclusion of a culture of brewing yeast in the pack (e.g. cask-conditioned or bottle-conditioned beers). Two processes are used in the brewery: a) flash pasteurization (Fig. 1.21) for beer before packaging, typically on its way to kegs or heat-sensitive plastic bottles, or b) tunnel pasteurization (Fig. 1.22) for can or bottled beer. The higher the temperature, the more rapidly microorganisms are destroyed. A 7 °C rise in temperature leads to a ten-fold increase in the rate of cell death.

In flash pasteurization, the beer flows through a heat exchanger, which raises the temperature typically to 72 °C. Residence times of 30-60 s at this temperature are sufficient to kill off virtually all pathogens and degradative viable microbes.

Tunnel pasteurizers are large heated chambers through which filled and sealed cans or glass bottles are conveyed over a period of minutes. Accordingly, temperatures in a tunnel pasteurizer are lower, typically 60 °C for a residence time of 10–20 min.

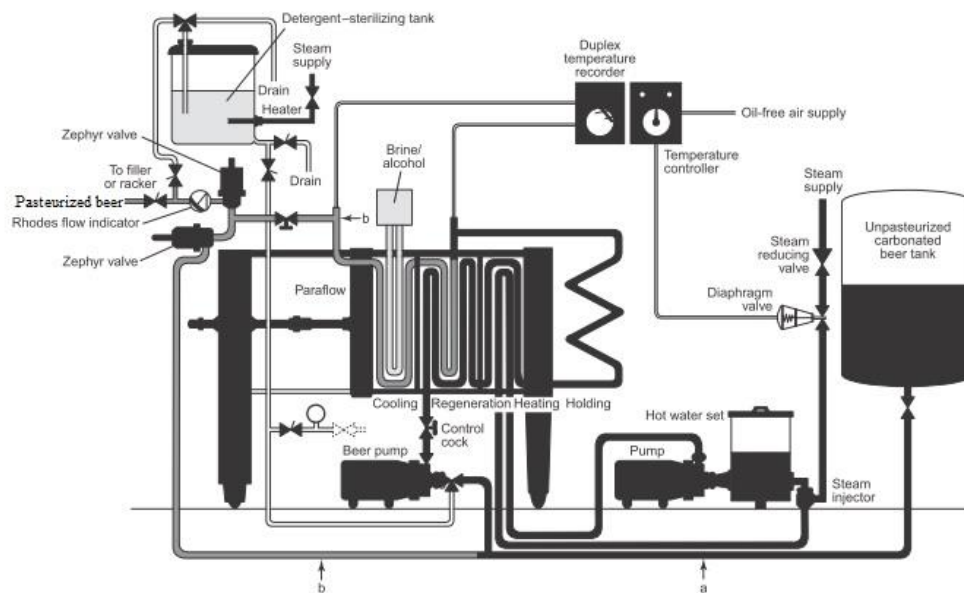


Figure 1.21 Schematic diagram of a flash pasteurizer (Briggs et al, 2004)

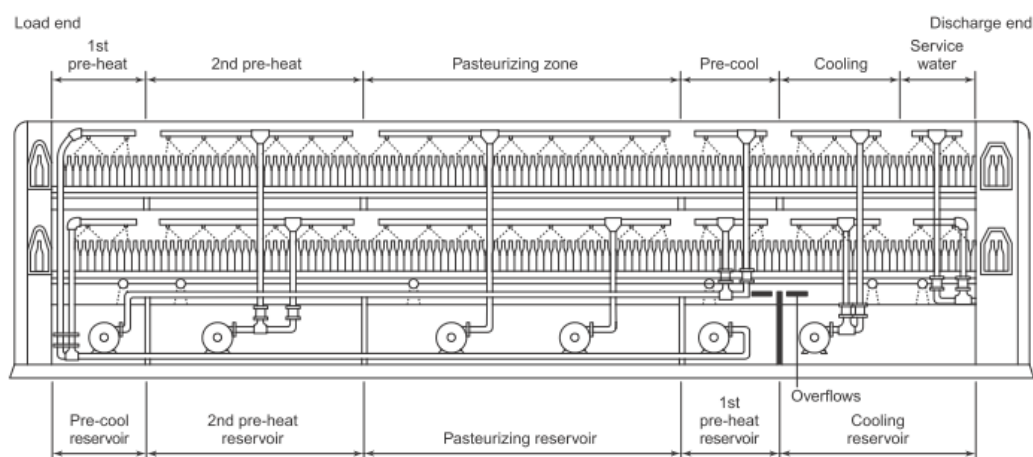


Figure 1.22 Schematic view of a tunnel pasteurizer (Hough *et al*, 1982).

Effect of pasteurization on beer quality

The main advantages of flash pasteurization are the following:

- The microbial kill-off is ensured independently of the type, morphology, and physiology of microbes or contamination count;
- Pressure fluctuations or irregularities in beer flow arising from switchover procedures or disturbances in bottling do not affect kill-off;
- The costs are the lowest comparative to all other processes.

The main disadvantages are:

- Taste stability is difficult to achieve, especially for sensitive, pale beers (e.g. Pilsener).
- High energy consumption.
- Heating of beers up to normal pasteurization temperature may denaturate some beer components (e.g. Maillard reaction).
- Some important flavor-producing components may be destroyed.

1.4 – CROSS-FLOW MICROFILTRATION MEMBRANES

A membrane can be defined as a barrier that separates two phases restricting totally or partially the transport of one or several chemical species present in each phase. Depending on the driving force and physical sizes of the separated species, membrane

processes can be classified as: microfiltration (MF), ultrafiltration (UF), reverse osmosis (RO), dialysis, electrodialysis (ED) and gas separation (GS).

Membrane processes can be operated in two major modes according to the direction of the feed stream relative to the orientation of the membrane surface: dead-end filtration and cross-flow filtration.

Figure 1.23 shows the main differences between the conventional dead-end filtration (perpendicular feed), cross-flow filtration (tangential feed).

In cross-flow filtration the product to be filtered flows tangentially over the membrane with a high flow rate and specific working pressure. The direction of filtration and the flow direction are not identical. Only part of the liquid (*permeate*) is filtered through the membrane, the accumulated non filtrate (*concentrate* or *retentate*) is circulated again to achieve a good filtration result and the concentration of separated material is thus slowly increased. A turbulent flow is created over the membrane surface to result in self-cleaning of the membrane. Furthermore, in cross-flow filtration it is possible to rinse the membrane by means of the permeate, thus increasing the volume of the retentate.

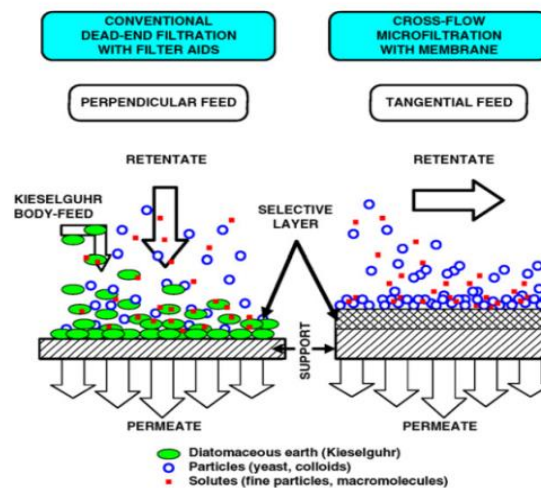


Figure 1.23 Comparison between dead-end, cross-flow and dynamic filtrations (Fillaudeau, *et al*, 2006).

The most important parameters that describe the separation performance of a membrane are its *permselectivity* and *permeability*. The latter is typically used to indicate the

capacity of a membrane for processing the permeate, whereas the former is the ability of the membrane to separate feed specific components.

1.4.1 Microfiltration membrane

Microfiltration refers to filtration processes using porous membranes to separate suspended particles with diameters between 0.1 μm and 10 μm (Fig. 1.24). Thus, microfiltration membranes fall between ultra-filtration membranes and conventional filters.

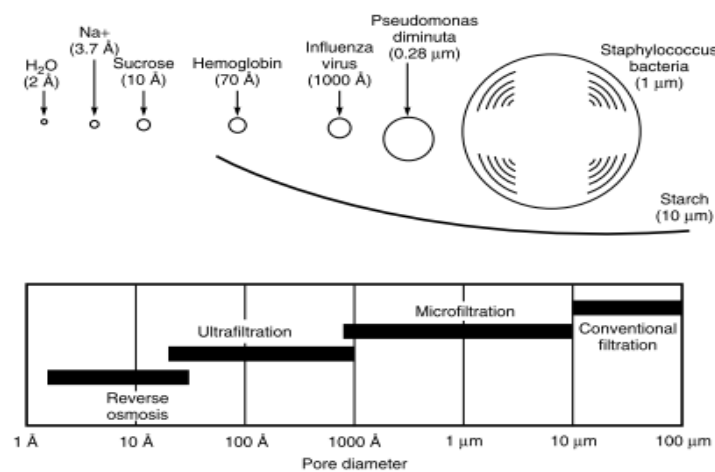


Figure 1.24 Relative size of solutes removed by each class of membrane.

The membrane configuration will obviously affect cost, ease of replacement, and efficiency of filtration. There are currently three primary configurations for the MF membranes industrially used:

- (1) plate-and-frame units
- (2) pleated cartridges
- (3) tubular/hollow-fiber modules.

Hollow-fiber membranes

The diameter of hollow fibers varies from 50 to 3,000 μm . Fibers can be made with a uniformly dense structure, but preferably they are formed by a micro-porous structure having a dense selective layer on either the outside or inside surface. The dense surface

layer can be either integral with the fiber or a separate layer coated onto the porous support fiber. Many fibers must be packed into bundles and potted into tubes to form a membrane module (Fig. 1.25). There are a few negative factors related to conventional polymeric membranes which have prevented their wide use in alcoholic beverage applications. These are: short membrane life-time, limited temperature and chemical resistance, flavor changes caused by the extraction of polymers, and compressibility of the membrane structure. The greatest single advantage of hollow-fiber modules is the ability to pack a very large membrane area into a single module.

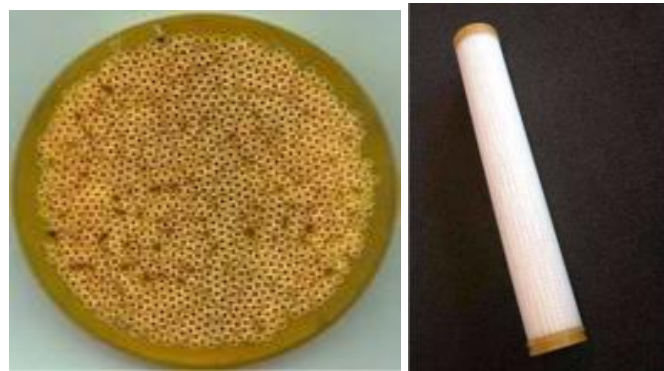


Figure 1.25 Views of hollow-fiber membrane modules

Ceramic membranes

These micro-porous membranes are made from aluminum, titanium or silica oxides. Ceramic membranes have the advantages of being chemically inert and stable at high temperatures, conditions under which polymer membranes fail. This stability makes ceramic microfiltration or ultrafiltration membranes particularly suitable for food, biotechnology and pharmaceutical applications, in which membranes require repeated steam sterilization and cleaning with aggressive solutions.

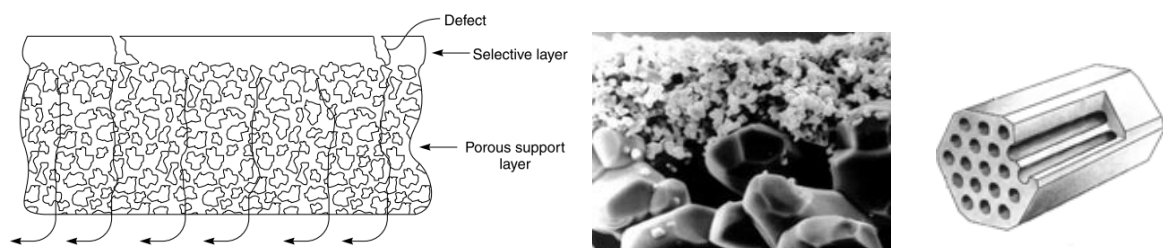


Figure 1.26 Schematic details of anisotropic ceramic membrane modules.

Ceramic membranes have an advantage over polymeric membranes regarding fouling, as they can withstand harsh cleaning methods. However, the obtained fluxes are usually significantly lower. Ceramic membranes with a small flow resistance would, therefore, be highly desirable for beer filtration (Kuiper *et al.*, 2002). The most significant advantages of a ceramic microfiltration membrane are its extraordinary thermal resistance, enabling high temperature cleaning, robustness in respect to pressure, due its anisotropic conformation (Fig. 1.26) and resistance against aggressive cleaning agents.

1.4.2 Cross-flow microfiltration in brewing processes

The CFMF has been under investigation for brewing applications over the last 25 years. Today, typically only 1~3 CFMF installations are sold yearly worldwide (Leeder *et al.*, 2011), kieselguhr filters being the mostly used worldwide in beer filtration scientific and technical literature reports three trends for future beer clarification processes:

- the reduction of Kieselguhr consumption;
- the replacement of Kieselguhr with regenerable filter-aids;
- the development of Kieselguhr-free processes (membrane filtration).

The first research applying CFMF in the brewing industry was initiated in the early 1980s (Gir & Leeder, 1992). Since then, this topic has been an area of intense research for several advantages, including the riddance of filter-aids and associated handling and disposal problems (Freeman & McKechinie, 1995), reduced beer losses (Lee, 1987), high solids handling capacity, the substitution of heat pasteurization and, consequently, a better product quality and cost savings. This process has so far found widespread use for fluid clarification or pasteurization/sterilization in the beverage, brewing, wine, cider, vinegar, fruit juice and dairy industries, biotechnology (e.g. cell separation from fermentation broth) and the treatment of oil and latex emulsions (Cheryan, 1998; Daufin *et al.*, 1998; Ho & Sirkar, 1992; Moresi & Lo Presti, 2003).

However, the CFMF operation should be able to :

- Produce a clear and bright beer with similar quality to a Kieselguhr filtered beer:
- Perform separation in a single-step without additives
- Operate at low temperature (0–2 °C)
- Achieve economic permeation fluxes.

Owing to the wide range of chemical composition and molecular size of the compounds to be retained, this operation is quite difficult to control. However, membrane processes should satisfy the same economic and qualitative criteria than conventional dead-end filtration. Beyond a low permeation flux and severe membrane fouling, their unsatisfactory separation properties are responsible for numerous negative aspects, such as inconsistent beer quality, uncertainty over productivity, large flux/quality variations among different beer brands filtered on one membrane system.

Microfiltration of beer or reclaimed beer, is generally regarded as economically feasible at flow rates of about 3,000 L/h (Leeder, 1995). The multi-channel ceramic membranes, formed using the same technology developed for making catalytic converters, are relatively inexpensive, especially when a minimum 5- year life time can be assumed compared with the typical 2-year life of polymeric membranes.

The main advantages of membrane CFMF are thus the following:

- the cross-flow filtration system can be installed as fully automatic units and all processes can be controlled and monitored so as to improve its productivity;
- any negative effect on beer quality as due to heat treatment is avoided. Moreover, no microorganisms which have been killed off remain in the beer;
- the natural fresh flavor of the beer is retained;
- good pressure stability (absence of momentary slip-leak of microorganisms from pressure surges);
- rapid developments are occurring in new membrane technologies for organic and inorganic types to achieve increased tenacity, chemical and temperature resistance, higher permeate fluxes and also a more regular pore size distribution.

However, the following disadvantages can be listed:

- membrane fouling resulting in a reduction of the permeation flux to far below the theoretical membrane capacity (steady-state fluxes are typically below 100 L h⁻¹ m⁻²);
- chemical and organoleptic analysis of beer before and after CFMF has so far confirmed that there is detectable change in the beer quality after filtration (e.g. retention of flavor-active constituents and decrease in foam stability);
- residual risks associated with the presence of small-cell beer contaminants, such as *L. lindneri*, *L. casei*, *L. coryniformis* and *L. brevisimilis*;

- For a given membrane, permeate flux may significantly vary depending on the beer to be treated (e.g., lager beer vs. dark types, such as Altbier);
- The operating costs may be twice or three times higher than those of flash pasteurization.

Actually, in brewing processes CFMF systems might play a useful role to maximize beer recovery. Loss reduction concerns two applications: the recovery of extract during wort clarification and beer recovery from tank bottoms (fermentation and maturation vessels). At present, tank bottom recovery constitutes the principal membrane application in brewing (Fillaudeau, *et al.* 2006), but MF may also be used for mash separation, clarification of rough beer and cold sterilization.

Loss reduction

The rough beer after fermentation can be rich in yeast with a very high viscosity or with a smaller number of yeast cells, but rich in proteins and polyphenols. In this case, the viscosity is close to that of clarified beer. In brief, rough beer may be clarified by using:

- a filter press (actually, this technology is no more applied because of the high oxygen uptake, contamination risk and poor quality of filtered beer);
- centrifuges and decanters (the quality of the recovered beer is uncertain and recovery rate is limited).
- cross-flow microfiltration membrane modules.

Centrifugation is expensive and may damage the permeate quality because of yeast cell degradation. Filter-presses provide a relatively low moisture solid discharge and, consequently, high extract recovery. However, no sufficient clarification of the filtrate is obtained.

The most advanced technology for extract separation asks for ceramic CFMF membrane modules. By controlling feed flow rate and pressure, it is possible to maximize permeation flux with minimum shear on yeast cells. The use of CFMF is designed to produce a permeate of acceptable quality including flavor and haze, with minimal loss of original gravity, color and bitterness, while processing a retentate of 2-4% (w/w) weight to a maximum of 20% (v/v), (Fillaudeau, *et al.* 2006). Moreover, by operating, close to 0 °C, economically permeation fluxes and hygienic beer recovery can be

achieved. Almost all the membranes installed in the breweries around the world are dedicated to the recovery of beer from fermentation and maturation tank bottoms. At present, these membrane applications have almost become industrial standards (Fig. 1.27). MF enables a retentate at 20-30% (w/w) solid content to be reached, yielding more than 50-60% of the yeast sediment to be recovered and a high quality filtrated beer with a volume reduction ratio of 2-3%.

The beer recovered from maturation tank bottom may be returned to the maturation vessel or sent for final clarification. Recovered permeate, recycled in the brewing process at a rate of 2-5%, allows beer loss and costs to be reduced. Tank bottom concentrates may be sold as livestock feed.



Figure 1.27 *Anisotropic ceramic CFMF membrane unit.*

Cold sterilization of clarified beer

The clarification of rough beer is usually followed by pasteurization to ensure the microbiological stability and conservation of beer. Currently, heat treatment is mainly performed by flash pasteurization for kegs and heat-sensitive plastic bottles or tunnel pasteurization for glass bottle. Conventional heat treatment requires water loops to heat and cool the product with high water and energy consumption. Sterile filtration appears interesting and allows the elimination of the organoleptic problems induced by heat processing (Reed, 1989, Nielsen, 1989). Potentially, cross-flow microfiltration can produce sterile beer ready to be packaged, with no negative change in beer quality and

stability. By operating at temperatures close to 0 °C, CFMF can be a truly operating alternative to pasteurization and dead-end filtration with cartridges.

Today, cold-sterile filtered-beer (draught beer or bottled beer) corresponds to a strong demand from consumers for quality and natural products and cold sterilization by CFMF membrane modules is still under trial for being industrially applied (Back, 1992). Krottenthaler *et al* (2003) reported that the technical developments of membrane filtration (membrane lifetime, running time, cleaning procedure, cost reduction), as well as market trends, reveal constant improvement. Financial aspects seem to be more and more attractive. For instance, the cost of flash-pasteurization is of the order of 0.20 € hL⁻¹, whereas membrane filtration is around 0.26 € hL⁻¹ of clarified beer (Fillaudeau, *et al* 2006).

CFMF for use in the area of cold sterile beer filtration must comply with the following specifications:

- retention of beer-spoilage organisms (bacteria and yeasts) in the cold temperature area;
- no negative changes of essential beer constituents;
- effective cleaning and sanitization of the membranes with cleaning agents (alkaline, acid, oxidative cleaning) and disinfectants (e.g. with chlorinated sanitizers);
- membrane with a narrow pore size distribution;
- easy operation;
- sufficiently long life and adequate economics.

Clarification of rough beer

Among the potential applications of CFMF, the clarification of rough beer represents a large potential market of approximately 200.000 m² surface (Fillaudeau, *et al* 2002), but till today this technology is used on a large scale only in a small number of breweries. For the membrane process, the choice of the filtration mode (dead-end, cross-flow or dynamic filtration) and membranes (chemical nature, mean pore diameter) is crucial. In conventional cross-flow filtration, high fluid velocities are generally necessary to limit the growth of cake on the membrane surface, but these high velocities require energy and also produce large pressure drops along the membrane module. The use of high transmembrane pressure difference, can limit the solute transmission into the permeate,

especially when the cake layer on the membrane is compressible, thus reducing protein transmission (Harscoat *et al*, 1999).

The first industrial plant running a CFMF unit of rough beer is located at Heineken and has a capacity of 10,000 L/h (Noordman *et al* 2001). The plant consist of 10 hollow-fiber modules X-Flow R100 (pore size: 0.45 μm ; length: 1 m; inner diameter: 1.5 mm; filter area: 9.3 m^2). The key of this process is based on a specific cleaning procedure. It combines a caustic step, an acidic step and a strong oxidative step (2 h in duration), which is successful in achieving a run time between 7 and 20 h for about 120 runs. Heineken and Norit Membrane Technology patented this procedure. Filtration is accomplished a temperature of 0 $^{\circ}\text{C}$, a feed superficial velocity of 2 m/s, and a transmembrane pressure difference up to 1.6 bar. During filtration, 10-min periods of back-flushing are applied every 2 h to remove the reversible fouling, that has built up. The flux is maintained at 100 $\text{L m}^{-2} \text{h}^{-1}$ and clarified beer fulfils the European Brewery Convention (EBC) standard in terms of turbidity (close to 0.6 EBC unit), bitterness, total extract, color, and protein content. The cost of membrane filtration for bright beer is about 0.40 € hL^{-1} . A cost comparison between the membrane and conventional process is given in Table 1.7 and Fig. 1.28.

Table 1.7 Comparison of the operating cost of different beer clarification processes. (Fillaudeau *et al*, 2006).

Operation	Membrane Process	Conventional Processing
Cold sterilization	Dead-end filtration (cartridge filter): 0.26 € hL^{-1}	Flash pasteurization: 0.20 € hL^{-1}
Clarification	Cross-flow filtration: 0.44 € hL^{-1}	Dead-end filtration with Kieselguhr: 0.43 € hL^{-1}

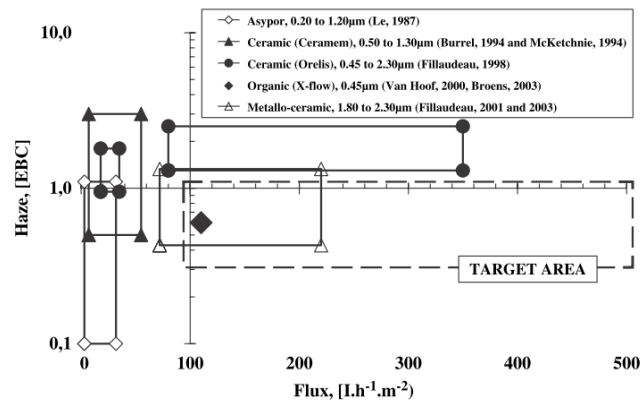


Figure 1.28 Flux versus haze in the clarification of rough beer by MF. (Fillaudeau *et al*, 2006).

1.4.3 CFMF parameters

The two main problems encountered in industrial experiments are:

- the control of fouling mechanisms and
- the enhancement of permeate quality and quantity.

Since 1995, a lot of works have indicated the economic and scientific stakes of the clarification of rough beer. Recent scientific and industrial studies have dealt with:

- i) fouling mechanisms;
- ii) the relationship between quantitative and qualitative performances;
- iii) the development of alternative membrane filtration, such as membrane structure and dynamic filtration;
- iv) industrial applications.

The most important disadvantages of using CFMF in beer clarification is the fouling and flux decline during the process, which is due to the feed viscosity and particles complexity.

Beer should be filtered at low temperatures in order to maximize the removal of chill haze. The lower the temperature, the higher the viscosity of the beer. Though macromolecules present in beer (proteins, polyphenols, polysaccharides, etc.) are much smaller than the pore size of typical MF membranes, they cause significant fouling. Güell and Davis (1996) found that protein fouling depend on protein ability to form aggregates, while Gan *et al* (1997) attributed flux decline to the internal fouling caused by carbohydrates. Also the complex interactions between fouling molecules and membrane material should be taken into account (Czekaj *et al*, 2000).

In the CFMF there are many operating parameters:

- unfiltered fluid velocity or recirculation velocity (m/s);
- specific filtration flow ($\text{L m}^{-2} \text{h}^{-1}$)
- transmembrane pressure difference (TMPD), that is the pressure difference between unfiltered and filtered fluid;
- pressure difference between the inlet and outlet section of membrane module (i.e., pressure drop).

Effect of fouling

The main reason for the current limited application of membrane separation is membrane fouling, which in beer filtration is severe and complicated, This phenomena

has caused difficulties in obtaining an economical flux, as well as good product quality. To overcome this drawback, many studies have attempted to enhance the filtration flux. During CFMF, fouling is initiated by penetration of aggregates through the membrane surface, followed by pore blocking for the first time of operation, cake formation mechanism controls the fouling phenomenon in the second time of operation and leads to the so called quasi-steady-state flux, generally the cake layer is highly compressible. The reversible fouling including gel and stationary cake layers contributes to more than 95% of the hydraulic resistance and the remainder is of the irreversible type. The in-pore fouling contributed to 5% of irreversible fouling resistance and 0.2% of total hydraulic resistance (Yazdanshenas *et al.*, 2010).

According to van der Sman *et al.*,(2012), fouling in beer CFMF implies several different mechanisms,(Fig.1.29) such as:

- gel layer formation and concentration polarization;
- cake layer formation;
- pore blocking/constriction;
- in-depth adsorption/deposition.

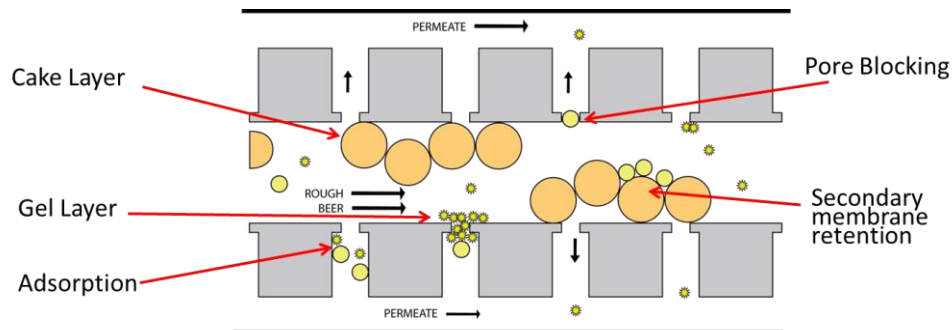


Figure 1.29 Schematic representation of different type of fouling. (van der Sman *et al.*, 2012)

The flux decline results from the superposition of various mechanisms of membrane fouling. The nature of foulants and different class and size of particle composition in beer microfiltration have been largely studied to identify a single key foulant, but probably, because of the complex nature of beer (which contains a large variety of molecular and colloidal fractions), the membrane fouling is caused by many factors, with effects changing with the operating conditions and beer used. This is probably, the main reason why membrane fouling remains a poorly understood phenomenon. Gan *et*

al (1997) was able to increase the permeation flux by over 20% by adding specific enzymes, this clearly indicating that carbohydrate species, such as β -glucans and starch molecules/particulates, affect the membrane performance. In the same study, the proteases exhibited only a small effect, this pointing out that proteins are not the major foulant. This result was slightly surprising, since microfiltered beer showed less foam stability (Head Retention Value, HRV).

Historically, the reduced efficiency of beer microfiltration has been largely attributed to β -glucans, (primarily the high molecular fractions), that increase beer viscosity by forming gels (Siebert, 1996; Leedham, 1975). Complex polysaccharides, such as β -glucans, are structural substances of malt endosperm and aleurone layer.

Beer brewed from several highly-modified malt samples had negligible β -glucan levels, whilst beer with the highest β -glucan content contained over 300 mgL⁻¹ (Table 1.4).

The average beer β -glucan content was around 72 mg/L, with a standard deviation of 88 mg/L (Table 1.40). Beer β -glucan content was highly correlated to total wort β -glucan level ($r=0.83$, $p<0.001$), (Sthwart, 1998).

More recently, beer β -glucan content was not significantly correlated to microfiltration efficiency (MFE). Sudarmana *et al* (2006) reported that the total β -glucan content did not correlate with bright beer membrane filtration and concluded that only a certain proportion of the total β -glucans content, namely the β -glucans gelling fraction was responsible for reduced permeation flux (J_v). When bright beer was dosed with small quantities of beer gelling material, membrane flux dramatically reduced. Beer gelling substances predominately contain β -glucan with an average molecular weight of 114 kDa, and smaller amounts of arabinoxylan and protein.

The effect of β -glucan molecular mass on J_v was studied by dissolving β -glucans of different molecular masses (i.e., 40, 82, 123, 137, 183, 225, and 245 kDa) in 5% (v/v) ethanol buffered to pH 4.1 with acetate (Fig. 1.30). Thus, β -glucan gels may form during the brewing process provided that β -glucans of the appropriate molecular mass is present in the wort. The study indicated that β -glucan with a low molecular mass had far less effect on J_v than large β -glucan molecules. Solutions of β -glucans with molecular masses of 40 and 82 kDa filtered efficiently, while solutions of β -glucan with an average molecular mass of 123 kDa exhibited a dramatically effect on J_v . Any further

increase in the molecular mass of β -glucans beyond 123 kDa had comparatively little effect on MFE (Fig. 1.30).

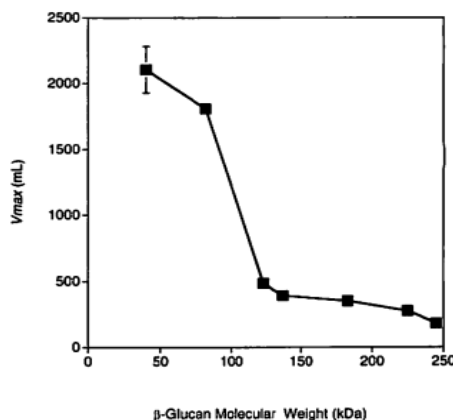


Figure 1.30 Effect of β -glucan molecular mass on microfiltration efficiency (Sthewart *et al*, 1998).

Esslinger & Narziss, (1985) and Narziss, (1993) related the poor membrane J_v to the presence of high molecular mass β -glucans. Consequently, total beer (malt or wort) β -glucans may not be the most accurate indicator of beer microfiltration performance, but rather malt β -glucan size may need to be taken into consideration. In fact, β -glucan molecules of high molecular mass had a greater influence on J_v than total β -glucan content. The beer brewed from malt samples containing the highest total β -glucan content (318 mg/L) filtered efficiently because the average molecular mass of such polysaccharides was 31 kDa. In contrast, the beer sample showing the lowest J_v value had a relatively low β -glucan content (80 mg/L), but the main fraction had a molecular mass of 220 kDa.

Sthewart *et al*, (1998) observed that J_v was more negatively correlated to the arabinoxylan content of beer ($r=-0.62$, $p<0.01$) than to the total β -glucan one ($r=-0.36$), even if beer viscosity was correlated to both contents ($r=0.68$, $p<0.05$, and $r=0.86$, $p<0.001$, respectively).

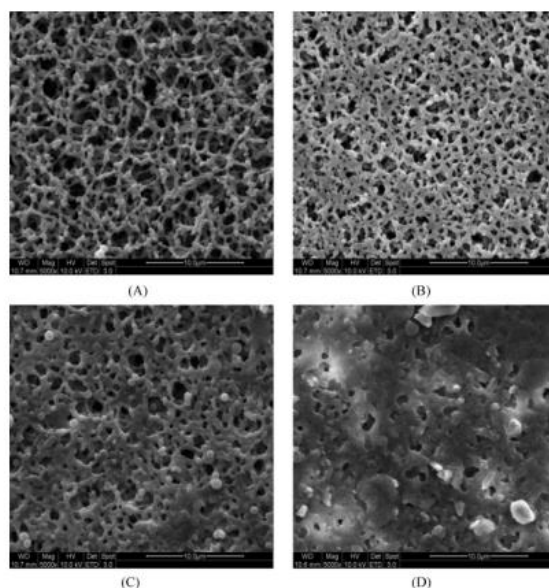


Fig. 1.31 SEM photo of 0.45- μm membrane surface (A) and SEM photos of membranes after filtration of beer containing arabinoxylan fraction with M_w of (B) 102 kDa, (C) 389 kDa, and (D) 963 kDa. (Jian Lu, 2005)

Adsorption of other macromolecules, such as proteins and polyphenols, was also been associated with reduced beer J_v (Sudarma, 1996). Gan (2001) found that pentosans was the major contributors to fouling.

Arabinoxylans are pentosans consisting of a β -linked xylan backbone substituted with arabinose. They are the major component of barley aleurone and endosperm cell walls, approximately 67% and 20%, respectively. Arabinoxylans are more water soluble than β -glucans, have the ability to form viscous solutions and as such have the potential to reduce MFE. However, there is little information regarding the effect of arabinoxylan on microfiltration performance (Fig. 1.31)

Arabinoxylan levels in beer may varied from negligible amounts to 97 mg/L when dealing with enriched wort. The average arabinoxylan content of beer (26 mg/L) is normally less than that of β -glucans (72 mg/L). Nevertheless, its negative influence on membrane filtration may have been previously underestimated. (Sthwart *et al*,1998)

Polyphenolic materials have the potential to bind protein and peptides to form larger particles, potentially capable of plugging filters. It has been suggested that dimers and trimers catechin and epicatechin can associate with proteins and peptides in beer, while proline-rich hordein fragments are suggested to have an affinity for (poly)phenolic

material. It has also been suggested that beer brewed with low proanthocyanidin malt may have improved diatomaceous earth filtration performance.

Effect of membrane pore size

Stopka (2000) studied the effect of membrane porosity of the permeation flux. Fig. 1.32 shows the time course of flux decline during beer microfiltration. The best filtration performance was achieved when using the membrane pore size of 200 nm, its steady permeation flux being approximately $17.5 \text{ L m}^{-2} \text{ h}^{-1}$ as compared to that obtained with the 500-nm membrane ($13.2 \text{ L m}^{-2} \text{ h}^{-1}$).

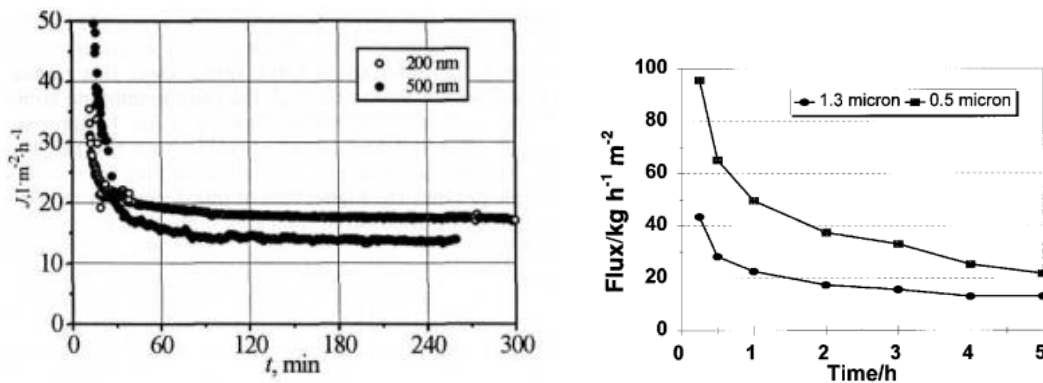


Figure 1.32 Permeation flux vs. time in microfiltration of beer through membranes of different pore size (Stopka *et al.* 2000; Gan *et al.* 1997)

The lower performance of the larger porosity membrane was explained by accounting for the accumulation of particles and foulants within the pores of the membrane. Once the feed had been discharged from the membrane module, the unit was rinsed with water before measuring the hydraulic permeation flux through the fouled membrane under the same conditions used during beer microfiltration. The steady permeate flux of the physiological solution was approximately three times higher than the steady flux observed with beer, with not a great difference between the first and the second rinsing. These results indicated strong adsorption of foulants, probably in-side the pore structure of the membrane. Quite the same value of the permeation flux after the second rinsing indicated that most foulants were strongly adsorbed, and fouling inside of the pore structure occurred. For the more opened membrane (pore size of 500 nm) the fluxes after rinsing were lower than those obtained with the 200-nm pore size membrane. This

suggested that inner blocking of larger pores occurred, this leading to a lower flux through the 500-nm membrane. On the other hand, the quality of beer recovered from surplus yeast was better for the membrane with a pore size of 500 nm.

Gan (1997) showed the effect of membrane pore size on permeation flux. The smaller pore-size membrane gave the higher fluxes (Fig. 1.32). This confirmed that one of the major mechanisms was in-depth pore plugging, as smaller pores excluded much of the material from the membrane matrix. Another important factor involved in membrane pore size is the resultant beer quality. Fig. 1.33 shows the head retention values (HRV) of the filtered beer as a function of time for 0.2- and 0.5- μm membranes. The HRV value is a quality control measure indicating foam stability and is related to the presence of head forming proteins. Clearly, the 0.5- μm pore membrane consistently transmitted such components, while the 0.2- μm pore size membrane tended to remove them. Moreover, the rate of removal increased as the membrane became fouled. A similar trend was noted with gravity (density) measurements. For the 0.2- μm membrane there was a general loss of carbohydrate molecules. It is thus obvious that product quality issues dictated the use of the 0.5- μm pore size membrane.

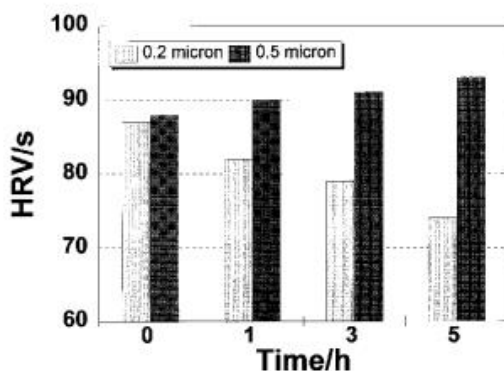


Figure 1.33 Effect of time and pore size on head retention value. (Gan et al, 1997)

Effect of TMPD

By increasing the transmembrane pressure difference, it is possible to increase the beer permeate flux, even if this increase is not always very significant.

Alicieo *et al* (2005) measured the permeate flux by setting TMPD at 1, 2, 3, and 4 bar and temperature at 6 ± 1 °C. As TMPD was increased, there was little effect on the permeate flux, but a significant reduction in beer color and turbidity, as well as other important qualitative beer parameters (Table 1.8). The steady state flux obtained with a

0.4 μm membrane was about 24.20, 27.20, 28.30, and 36.60 $\text{kg m}^{-2} \text{h}^{-1}$ for 1, 2, 3 and 4 bar respectively (Fig. 1.34).

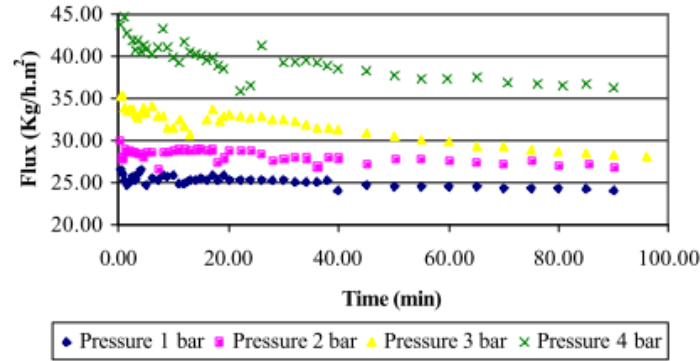


Figure 1.34 Effect of time and TMPD on the permeation flux in beer clarification using a 0,4 μm membrane module (Aliceo et al, 2005).

Table 1.8 Percentage rejection of different beer quality parameters at different TMPDs in 0.4- μm microfiltration membrane. (Aliceo et al,2005)

Parameters / Pressure (bar)	R (%)			
	1	2	3	4
Color	13.0	25.9	36.9	30.6
Bitterness	10.9	16.1	14.8	22.6
pH	0.0	1.4	-0.5	0.2
Sugar Redutor	9.7	16.2	24.3	28.6
Alcohol	-2.3	2.3	-0.6	-0.6
Primitive Extrat	3.7	9.9	10.1	9.9
Real Extrat	15.5	25.9	31.5	29.8
Aparent Extrat (°P)	28.6	43.8	53.1	50.0
Turbidity	98.2	100.0	100.0	94.3
Protein	35.4	55.7	59.0	53.2

The higher TMPD in crossflow microfiltration of beer, the more rapid flux decline and the lower the steady-state flux level become as a result of a more compact cake formation and greater in-pore fouling.

To delay the onset of fouling and maintain an efficient flux performance, it is recommended to start the operation by setting a low TMPD value and gradually increase TMPD in steps.

Effect of superficial velocity

To limit the concentration polarization phenomenon, as well as to avoid the formation of a film of organic particles on the wall of the membrane, it is necessary to increase the

feed superficial velocity to ensure a turbulent flow. The total quantity and quality of deposits and their stability within the stagnant boundary layer of the fluid is known to be dependant on the cross-flow velocity and fluid turbulent patterns. A cross-flow velocity in the range of $1\text{--}4\text{ m s}^{-1}$ is equivalent to a Reynolds number of 776 to 3,104. Thus, the effect of hydrodynamics (velocity, wall shear stress) on filtration performances remains limited.

Retentate velocity constitutes the main factor preventing cake deposition. The choice of the optimum cross-flow velocity has to be considered by accounting for these remarks (Filladeau; 1998).

A low cross-flow velocity involves weak hydrodynamic conditions and a weak cake erosion. In this case, the reversible external fouling constitutes the main hydraulic resistance, but the power consumption and negative effects on beer quality are low. By increasing the cross-flow velocity, it is possible to remove the cake at the membrane surface, thus reducing the contribution of the reversible external fouling. However, retentate velocities above the optimal values bring no significant flux enhancement, but involve high power consumption.

Flux enhancement techniques

The rapid decline in flux makes it difficult to control system operation. For this reason, microfiltration systems are often operated as constant flux systems, and the transmembrane pressure difference across the membrane is slowly increased to maintain the permeation flux as the membrane fouls. Most commonly, the feed pressure is fixed at some high value and the permeate pressure is set at a value just below the feed pressure. As the membrane is used, the decrease in permeability is counterbalanced by lowering the permeate pressure and so increasing the pressure driving force. When the permeate pressure reaches some predetermined value, the module is taken off-line and cleaned or back-flushed to restore its permeability. The membranes perform best when operated at conditions where membrane fouling is controlled by the flow of liquid across the surface. Operating at high flux or high transmembrane pressure difference leads to deposition of a thick compacted fouling layer. A large number of hydrodynamic techniques, based on fluid instabilities, have been investigated, such as co-current mode, pulsating flow, periodic stop of the transmembrane pressure, periodic back-flush or a

back-shock process, generation of Dean or Taylor vortices, introduction of turbulence promoters (baffle channel, stamped membrane) or the use of a two-phase flow (gas–liquid, liquid–solid). (Blanpain *et al*, 1999; Fillaudeau *et al*, 2007; Gan *et al*, 1997; Kuiper *et al*, 2002).

Back-flushing

The microfiltration of beer is also controlled by formation of a cake layer on the surface of the membrane and therefore hydrodynamic techniques may play a significant role in improving the performance of the operation. To increase the permeate flux, a back-flushing with permeate can be employed. Fig. 1.35 shows the effect of an applied frequency of back-flushing on the surface microfiltration membrane. It is evident that the frequency of back-flushing has to be carefully chosen, because for high frequencies the flux can decrease and eventually reach a zero value.

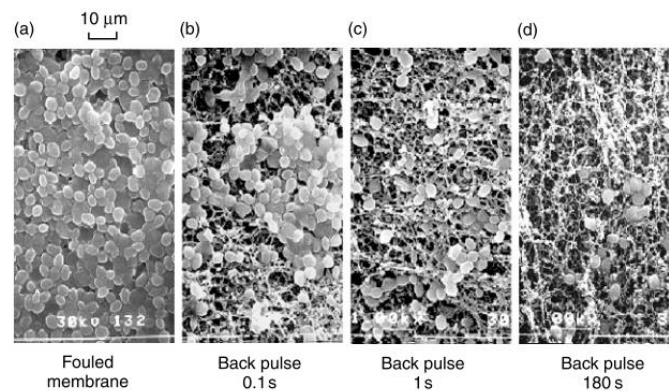


Figure 1.35 *Illustration of the efficiency of back-pulsing in removing fouling materials from the surface of microfiltration membrane (Sondhi R, Bhawe R, 2001)*

The principal aim in designing a good back-flush regime is to minimize permeate usage as back-flush media, whilst achieving the maximum pore clearance within the shortest possible time. Gan (2001) governed the back-flush by a programmed multi-stage back-pulse routine able to vary the frequency and pulse strength. The input variables included the CO₂ pressure, the duration of the pulse, duration between the end of the pulse and opening of the permeate valve and cycle frequency. These variables influence the membrane cleaning efficiency of the back-pulse, dead-time and loss of permeate. The lost permeate is the permeate recycled from the product to the feed-side and so this portion does not contribute to the net flux. Gan (2001) studied the beer clarification by

CFMF so as to improve a low permeation rate to an economically acceptable level by employing and optimizing some flux enhancement techniques.

Flux improvement through back-flush filtration with a back-flush (BF) program using the combination of CO₂ and permeate as back-flush media was adopted. When only liquid permeate was used, the amount of beer recycled from one side of the membrane to the other was excessive and the net flux was low. The backpressure was the pressure in the CO₂ line at the start of the BF routine. The duration of the period when the flux was above the moving average ranged from 60 s at the beginning of the run down to 10-20 s after 5 h of filtration. The implication is that a back-flush program should be designed to take account of this. Excessive back-flush will result in high costs through the loss of permeate and effective filtration time. A multi-stage back-flush program was designed to deliver a suitable back-flush frequency and strength with regard to the base flux level at different filtration stages. Application of the program improved the ten hour average flux by 400%, that is $22 \text{ kg m}^{-2} \text{ h}^{-1}$, as compared to the initial baseline case of standard cross-flow filtration (Gan *et al*, 1997)

1.4.4 Filtration Modelling

Several filtration models are used to describe flux decay during constant pressure filtration based on the blocking laws and cake filtration theory. These models were originally developed by Hermans & Bredee (1936) by assuming that the filtering medium consisted of capillary cylindrical pores having equal and constant diameter. Filtration models comply with the resistance models derived from Darcy's law according to which at any instant during the filtration process, the flux J ($\text{m}^3 \text{m}^{-2} \text{s}^{-1}$) is expressed as

$$J = \frac{\text{TMP}}{\eta R_T} \quad (1.7)$$

with

$$R_T = R_m + R_{\text{irr}} + R_{\text{rev}} \quad (1.8)$$

where TMPD is the transmembrane pressure difference, η the dynamic filtrate viscosity, while R_T , R_m , R_{irr} , and R_{rev} are the overall membrane resistance, intrinsic membrane resistance, and resistances of the irreversible and reversible fouling layers, respectively. In particular, R_{rev} includes concentration polarization and deposition of solids on the membrane surface (stationary cake layer), while R_{irr} is due to interaction of the membrane with the particles and aggregates in the feed stream and comprises of the blocking of the pores entrance and internal fouling inside the pores.

There are four basic mechanistic models that are generally used to describe membrane fouling, the schemes of their corresponding fouling mechanisms being illustrated in Fig. 1.35.

In the complete pore blockage (CPB) model, it is assumed that each particle arriving at the membrane seals pore opening with no superposition of particles. Thus, the flux decays as the filtrate can only pass through the unblocked pore area. The intermediate pore blockage (IPB) model is similar to the complete pore blockage model and accounts for the possibility that particles can deposit on other deposited particles that have blocked some pores. The cake filtration (CF) model presumes that a uniform, permeable cake layer of particles forms over the entire membrane surface. Finally, the standard pore blocking (SPB) or pore constriction (PC) model is based on deposition of particles onto the internal pore walls leading to a decrease in the pore diameter.

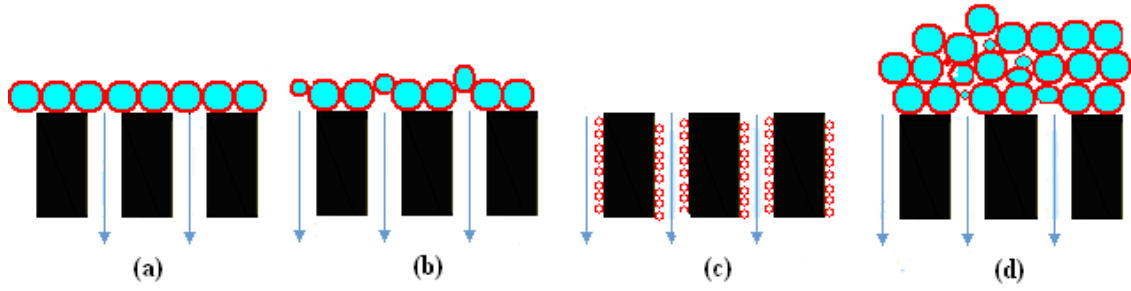


Figure 1.35 Scheme of the four basic fouling mechanisms: a) complete pore blocking (CPB); b) intermediate pore blocking (IPB); c) standard pore blocking (SPB) or pore constriction (PC); d) cake filtration (CF).

All of these models were grouped using the following relationship (Hermia, 1982):

$$\frac{d^2t}{dV^2} = k \left(\frac{dt}{dV} \right)^n \quad (1.9)$$

where t is the filtration time, V the total volume filtered, and n and k are characteristic parameters of the generic fouling model. In particular, the value of the exponent (n) allows the different fouling mechanisms to be differentiated. In the case of CPB mechanism, n is equal to 2 and k is proportional to J_{v0} (that is, the initial flux). For PC one, n equals 1.5 and k is proportional to $(J_{v0})^{0.5}$. For IPB mechanism, n is equal to 1 and k is independent of J_{v0} . Finally, for CF one, n is nil and k is proportional to $(1/J_{v0})$.

In general, the filtration tests allow the permeate volume (V) versus time (t) data to be easily collected. By resorting to the five points forward difference derivative formula or some polynomial function in restricted time intervals, it is possible to estimate numerically the first derivative (dV/dt). In particular, by referring to the five-point stencil of a point t in the grid shown in Fig. 1.36, the first derivative (dV/dt) can be approximated as:

$$\frac{dV}{dt} \approx \frac{-V(t+2h) + 8V(t+h) - 8V(t-h) + V(t-2h)}{12h} \quad (1.10)$$

Then, it is possible to estimate the volumetric permeation flux as:

$$J_v = \frac{1}{A_{m0}} \frac{dV}{dt} \quad (1.11)$$

and in sequence the derivative of t with respect to V as:

$$\frac{dt}{dV} = \frac{1}{J_v A_{m0}} \quad (1.12)$$

and the second derivate of t with respect to V as

$$\frac{d^2t}{dV^2} = -\frac{1}{J^2 A_{m0}} \frac{dJ}{dV} = -\frac{1}{J^2 A_{m0}} \frac{dJ}{dt} \frac{dt}{dV} = -\frac{1}{J^3 A_{m0}^2} \frac{dJ}{dt} \quad (1.13)$$

where A_{m0} is the initial effective surface area of the membrane module.

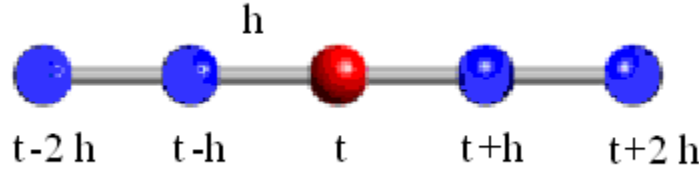


Figure 1.36 Five-point stencil of a point t in a one-dimensione grid.

By replotting the V -vs.- t data as (d^2t/dV^2) against (dt/dV) as suggested by Eq. (1.13) using log-log scales, it is possible to estimate the exponent n and infer the most probable fouling mechanism. Generally, these curves do not show a single n value for the full course of the filtration, but rather one that changes with time. When this occurs, it is possible to assess the relative importance of each mechanism at a particular point in the filtration process. From the literature, no single classical fouling model allows a good prediction of a generic filtration process.

In most cases, the characteristic curve starts with a slope sharper than that of complete pore blocking model ($n > 2.0$) owing to the irregularity and interconnectivity of the membrane pore structures. Then, the slope of the curve tends to reduce.

Alternatively, Eq. (1.9) can be rewritten in a physically more meaningful form by resorting to the unified filtration equation for CFMF developed by Field *et al*, (1995):

$$-\frac{dJ_v}{dt} = k (J_v - J^*) J_v^{2-n} \quad (1.14)$$

where k and n maintain the same meaning given above, while J^* is the so called critical flux, which should not be exceeded if fouling is to be avoided (Field *et al.*, 1995). This term is sometimes confused with the steady-state permeation flux (Stopka *et al.*, 2001).

Although these four fouling models have been developed for dead-end filtration, their use allows quite an easy identification of the likely dominant fouling mechanism, that results from the interactions among macromolecules, colloids and/or suspended

particles and membrane porous structure and leads to the permeation flux decay (Blanpain et al., 1993; Blanpain & Lalande, 1997). Nevertheless, there are significant discrepancies between the experimental flux decline data and model predictions. Moreover, the accuracy of the empirical fouling parameters n and k is strictly related to that of the numerical differentiation, which on turn depends on the smoothness of the experimental data or better on the algorithm used to smooth the raw data before being numerically differentiated. For instance, the t -vs- V data were smoothed using the Robust Loess (quadratic fit) algorithm with span of 0.2 (MATLAB® curve-fitting tools) before being numerically differentiated.

Thus, to avoid such a troublesome procedure, the data collected, that is the permeate volume (V) and volumetric permeation flux (J_v) against time (t), will be directly reconstructed by accounting not only for any of the four basic fouling mechanisms mentioned above, but also for their combined effects by means of the explicit equations relating V and J_v to time during constant pressure operation, as derived from Darcy's law by Bolton *et al.* (2006) and listed as Eqq. (1.15) – (1.34) in Table 1.10.

Table 1.10 Mathematical expressions for classical single (i.e., complete pore blocking, CPB; intermediate pore blocking, IPB; cake filtration, CF; standard pore blocking or pore constriction, PC) and combined (i.e., CF+CPB; CF+IPB; CF+PC, CPB+PC, IPB+PC) dead-end fouling models (FM), as derived from Boulton et al. (2006).

FM	Volume Equation	Eq. no.	Flux Equation	Eq. no.
CPB	$V(t) = \frac{A_{m0} J_{v0}}{k_{CPB}} (1 - e^{-k_{CPB} t})$	1.15	$J_v(t) = J_{v0} e^{-k_{CPB} t}$	1.16
IPB	$V(t) = \frac{A_{m0}}{k_{IPB}} \ln(1 + k_{IPB} J_{v0} t)$	1.17	$J_v(t) = \frac{J_{v0}}{1 + k_{IPB} J_{v0} t}$	1.18
CF	$V(t) = \frac{A_{m0}}{k_{CF} J_{v0}} (\sqrt{1 + 2 k_{CF} J_{v0}^2 t} - 1)$	1.19	$J_v(t) = \frac{J_{v0}}{\sqrt{1 + 2 k_{CF} J_{v0}^2 t}}$	1.20
PC	$V(t) = \frac{A_{m0}}{\frac{k_{PC}}{2} + \frac{1}{J_{v0} t}}$	1.21	$J_v(t) = \frac{J_{v0}}{(1 + \frac{k_{PC}}{2} J_{v0} t)^2}$	1.22
CF+CPB	$V(t) = \frac{J_{v0} A_{m0}}{k_{CPB}} \{1 - \exp[-\frac{k_{CPB}}{k_{CF} J_{v0}^2} (\sqrt{1 + 2 k_{CF} J_{v0}^2 t} - 1)]\}$	1.23	$J_v(t) = \frac{J_{v0}}{\sqrt{1 + 2 k_{CF} J_{v0}^2 t}} \exp[-\frac{k_{CPB}}{k_{CF} J_{v0}^2} (\sqrt{1 + 2 k_{CF} J_{v0}^2 t} - 1)]$	1.24
CF+IPB	$V(t) = \frac{A_{m0}}{k_{IPB}} \ln[1 + \frac{k_{IPB}}{k_{CF} J_{v0}} (\sqrt{1 + 2 k_{CF} J_{v0}^2 t} - 1)]$	1.25	$J_v(t) = \frac{J_{v0}}{(1 - \frac{k_{IPB}}{k_{CF} J_{v0}}) \sqrt{1 + 2 k_{CF} J_{v0}^2 t} + \frac{k_{IPB}}{k_{CF} J_{v0}} (1 + 2 k_{CF} J_{v0}^2 t)}$	1.26
CF+PC	$V(t) = \frac{2 A_{m0}}{k_{PC}} \{\beta \cos[\frac{2\pi}{3} - \frac{1}{3} \arccos(\alpha)] + \frac{1}{3}\}$	1.27	$J_v(t) = \frac{J_{v0}}{(k_{CF} J_{v0} \frac{V(t)}{A_{m0}} + \frac{1}{[1 - \frac{1}{2} k_{PC} \frac{V(t)}{A_{m0}}]^2})}$	1.28
	$\alpha = \frac{8}{27\beta^3} + \frac{4k_{PC}}{3\beta^3 k_{CF} J_{v0}} - \frac{4k_{PC}^2 t}{3\beta^3 k_{CF}} \quad \forall \alpha \neq -1 \text{ or } +1$	1.29		

$$\beta = \sqrt{\frac{4}{9} + \frac{4 k_{PC}}{3 k_{CF} J_{v0}} + \frac{2 k_{PC}^2 t}{3 k_{CF}}} \quad 1.30$$

$$\text{CPB+PC} \quad V(t) = \frac{J_{v0} A_{m0}}{k_{CPB}} \left[1 - \exp\left(-\frac{2 k_{CPB} t}{2 + k_{PC} J_{v0} t}\right) \right]$$

$$1.31 \quad J_v(t) = \frac{4 J_{v0}}{(2 + k_{PC} J_{v0} t)^2} \exp\left(-\frac{2 k_{CPB} t}{2 + k_{PC} J_{v0} t}\right) \quad 1.32$$

$$\text{IPB+PC} \quad V(t) = \frac{A_{m0}}{k_{IPB}} \ln\left(1 + \frac{2 k_{IPB} J_{v0} t}{2 + k_{PC} J_{v0} t}\right)$$

$$1.33 \quad J_v(t) = \frac{J_{v0}}{\left[1 + \left(\frac{k_{PC}}{2} + k_{IPB}\right) J_{v0} t\right] \left(1 + \frac{k_{PC}}{2} J_{v0} t\right)} \quad 1.34$$

Using the Darcy's model, the permeation flux can be expressed via Eq. (1.7) and the instantaneous total resistance (R_T) can be extracted from the experimental flux data as:

$$R_T = R_m + R_c = \frac{TMP}{\eta J} \quad (1.35)$$

where R_m the membrane resistance, and R_c the cake resistance.

Tracey and Davis (1994) observed that at the initial time, the total resistance versus time curves were generally concave upward suggesting fouling by pore constriction or pore blockage mechanism, while at longer filtration times, the curves were concave downward indicating fouling by cake filtration mechanism.

1.5 - ENVIRONMENTAL IMPACT OF BREWING

Even if the brewing industry has an ancient tradition, the environmental issues will force the brewing sector to demonstrate that it is performing within the constraints of product quality, process safety, economic viability, and limited environmental damage.

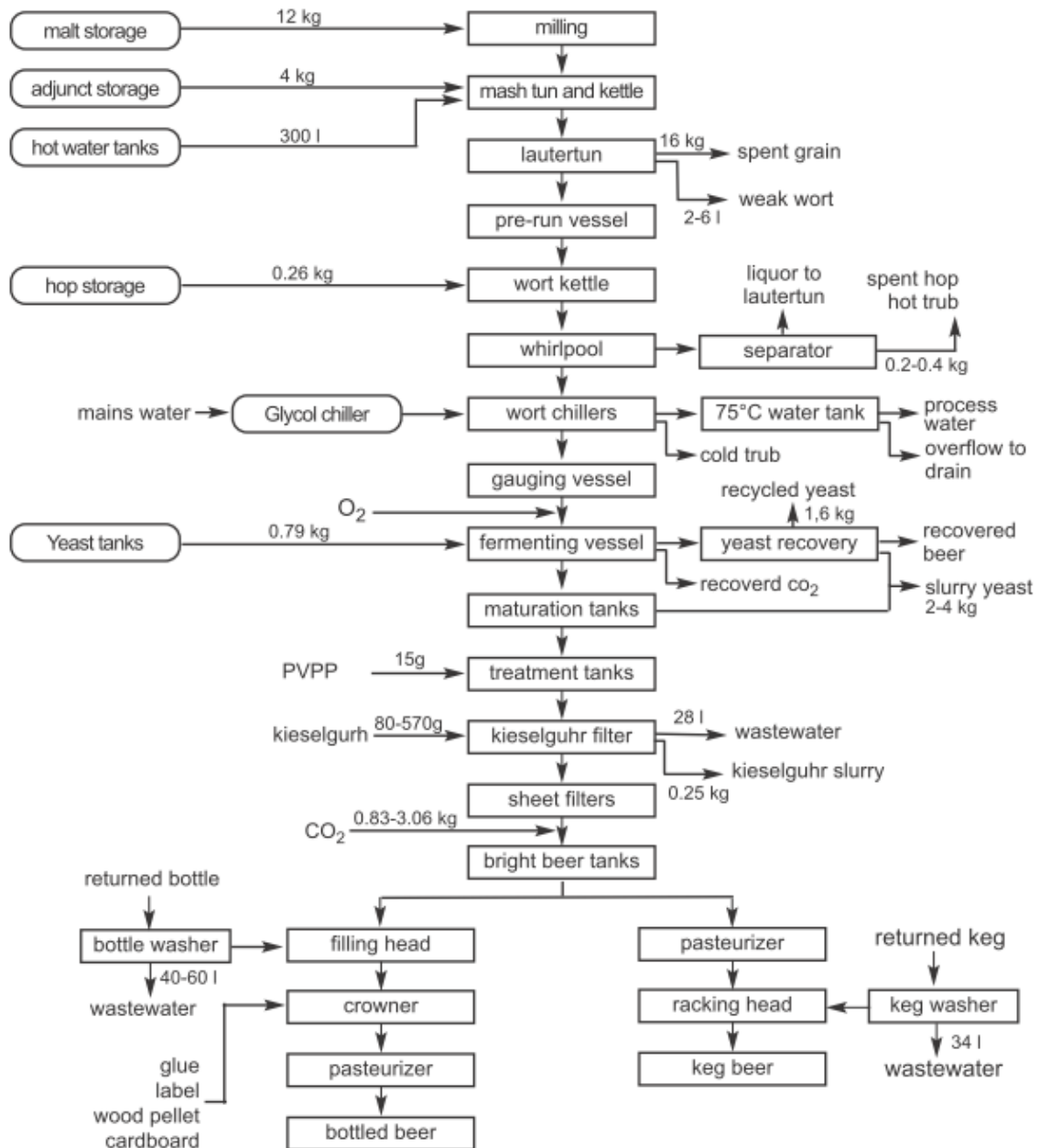


Figure 1.37 Brewery flow input-output referred to 100 L of beer produced.

Every brewery tries to keep waste disposal costs low, whereas the legislation imposed for waste disposal becomes more stringent. Thus, waste treatment or dumping costs are increasing.

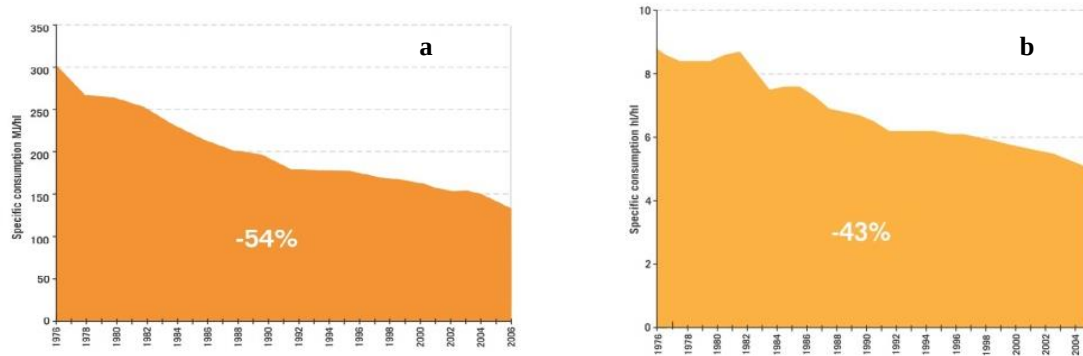


Figure 1.38 a) Delivery energy for hL of beer produced; and b) Specific water consumption (British brewery) (The British brewing industry, 2006)

The brewing industry is a large user of water per unit volume of product (Table 1.11). A lot of thermal energy is utilized in the early process steps and even more cooling energy in the later ones. Nevertheless, as shown in Fig. 1.38 by referring to a British brewery, over the last 30 years the delivered energy consumption and water consumption per hl of beer produced have fallen by 54% and 43%, respectively.

Table 1.11 Typical resource consumption data for European breweries (UNEP, 1996).

Country	Water (L/L)	Heat (MJ/hL)	Electricity (kWh/hL)
Spain	5.3 – 11.9	114 - 262	9.2 -19.7
Germany	6.6 - 8.6	153 - 244	11.0 – 16.0
United kingdom	5.9 - 11.1	155	12.5
Norway	7.4 – 10.6	209 - 232	19.2
Denmark	4.1 – 8.7	120 - 228	6.6 – 16.9

Furthermore, the position of beer as a natural product leads the brewers to pay attention to their marketing image and to take waste treatment into account.

As shown in Fig. 1.39, the main environmental issues associated with the operation phase of breweries include:

- material consumption;
- energy consumption;

- water consumption;
- wastewater ;
- solid waste and by-products;

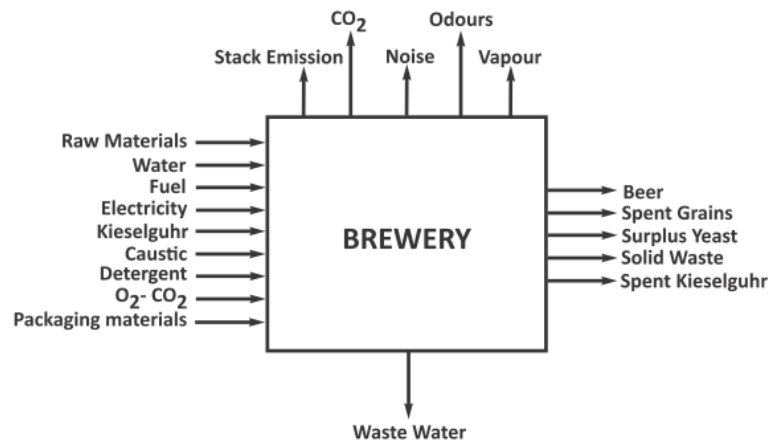


Figure 1.39 *Main resources used and main emissions generated during brewing (UNEP, 1996a)*

1.5.1 Material consumption

The consumption of raw materials depend on the type of beer produced, extract loss, and brewery global efficiency. The primary raw materials used in brewery are barley, water and hop. Barley may be substituted by several adjuncts, which are supplementary extract starch supplies, like maize, grist or rice or alternatively sugar. These are used to reduce the cost of production. Normally, about 15-17 kg of malt are needed to produce 1 hL of beer and, of this amount, not more is 30-40% adjunct. Hop can be added in the form of natural hop, hop extract or hop powder. Other auxiliary materials are needed to complete some brewery operations, like filtration and stabilization, as well as for the cleaning operation.

The main raw materials used in brewery are show in Table 1.12.

Table 1.12 *Range and average value of the specific consumption of raw materials for producing 1 hL of beer in Europe (UNEP, 1996).*

Materials	Specific consumption range	Average	Unit
Malt and cereals	15-18	-	kg/hL
Hop	0.26	-	kg/hL
Kieselguhr	80 – 570	255	g/hL
PVPP	15	-	g/hL
Carbon dioxide	0.83 – 3.06	1.83	kg/hL
Caustic	0.39 – 1.07	0,7	kg/hL

1.5.2 Energy and heat consumption

Brewery processes are intensive users of both electrical and thermal energy (Table 1.13). Thermal energy is mostly used in the form of steam in the boilers to wort boiling and water heating in the brewhouse, and in the bottling process. The refrigeration system is the largest single consumer of electrical energy, but also the brewhouse, bottling hall and wastewater treatment plant ask for large electricity demand. Electricity consumption by a well run brewery is about 8-12 kWh/hL. However, the specific energy consumption of a brewery is heavily influenced by the utility system and process design, as well differences in product recipe and packaging type, or from the input temperature of brewing water and climatic variations.

Table 1.13 *Estimated percentage of thermal and electric energy use for various brewing processes (Sorrel, 2000).*

Thermal Energy	Brewhouse	30-60%
	Packaging	20-30%
	Space Heating	<10%
	Utilities	15-20%
Electrical Energy	Refrigeration	30-40%
	Packaging	15-35%
	Compressed air	10%
	Brewhouse	5-10%
	Boiler house	5%
	Lighting	6%
	Other	10-30%

Specific heat consumption in a brewery can vary from 100 to 200 (MJ/hL), the main heat-consuming processes are:

- mashing,
- wort boiling,
- generation of hot liquor,
- CIP/Sterilisation,
- bottle/keg washing,
- pasteurization,

The heat consumption is influenced by the use of well known heat recovery and conservation techniques, like:

- heat recovery from production processes or utility systems to process or boil feed water, wort cooling, and vapor condensation from the wort vessel;
- The use of high-gravity brewing;
- Evaporation control and optimization in wort boiling;
- Insulation of steam, hot water and refrigerant pipes;
- Heating optimization in tunnel pasteurizers;

In a life-cycle assessment study on beer production in Greece (Koroneos *et al*, 2003), it was estimated that the greatest fraction (85%) of the global energy consumed in beer production system is absorbed by glass bottle production, while only 6.1% by beer production (Fig. 1.40).

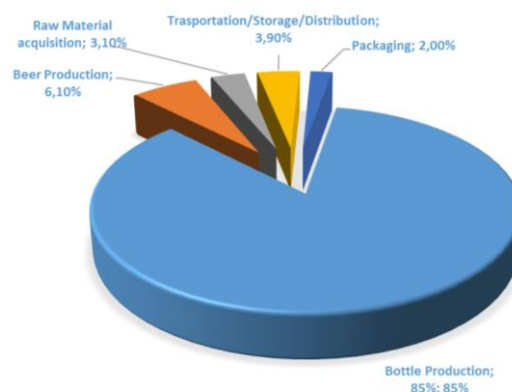


Figure 1.40 The distribution of energy in the beer production system in Greece (Koroneos *et al*,2003).

For beer that is expected to have a long shelf life, pasteurization represents one method to achieve the death of all remaining harmful bacteria before bottling, in this phase to heat the beer there are different pasteurization techniques, like tunnel or flash pasteurization. The energy requirements for the former can vary from 19 to 23 kWh per 1000 bottles (Hackensellner, 2000). Other estimates are 14.73-22.60 kJ/barrel (1 US beer barrel = 117.35 L)(Anheuser Busch, 2001). The new alternative approach to use sterile filtration should represent an energy saving approach, but some believe that these systems may require as much extra energy as they save (Galitsky C *et al*, 2003).

1.5.3 Water Consumption

High consumption of good-quality water is characteristic of beer brewing (Table 1.14). More than 90 percent of beer is water and an efficient brewery will use 4-7 L of water to produce 1 L of beer. Indeed, breweries use water for heating and cooling; cleaning packaging vessels, production machinery and process areas, like pasteurization and packaging. A bottle washer consumes more water than a can washer, also the washing water requirement is lower in case of non-returnable glass bottles than in returnable ones. Water is also lost through wort boiling and with spent grains.

Water consumption for individual process stages, within a typical brewery according to the BREF (2006) and to the publication of European Brewery Convention, EBC (1990) is reported in Table 1.14. Based on these results, the cellar with filters was marked as one of the critical points in the brewery. Higher water consumption was observed in case of bottle washing (returnable glass bottles), CIP system, and pasteurization.

Table 1.14 Specific water consumption values (L/L of beer) for different brewery processes. (Tokos 2000).

Department	BREF	EBC	
		Good practice	Best practice
Brewhouse to wort cooling & CIP	1.36 – 2.36	1.75	1.48
Fermentation and Yeast handling & CIP	0.32 – 0.53	0.09	0.05
Maturation & CIP	0.24 – 0.67	0.09	0.05
Filtration – BBT & CIP	0.31 – 1.09	0.28	0.06
Keg washing (50% of production)	0.59 – 1.63	0.34	0.17
Bottle washing (25% of production)	0.13 – 0.61	0.23	0.13
Bottle and can pasteurisation	–	0.16	0.08
CIP (Bottling)	–	0.42	0.20
Water treatment	–	0.2	0.16
Boilers	–	0.36	0.16
Evaporative cooling tower	–	0.55	0.38
Air compression & CO2	–	0.06	0.04
Total	3.7 – 4.7	4.53	2.96

From Tokos (2000), the fresh water consumption of modern breweries generally ranges from 3.7 to 4.7 L per L of beer sold. Water consumption from two to three times the above value is not unusual, particularly where the raw water temperature is high. Further data are given in Table 1.15. The average water consumed by the SAB Miller company was about 4.3 hL per hL of beer in 2010 (SAB Miller, 2011). While not all breweries operate at this level, all plants are assessed regularly, and the SAB Miller group has an internal *Water Staircase grading system*. By 2015 SAB Miller is expected to reduce its water consumption to ~ 3.5 L per L of beer (SABMiller Position Paper – Water). In 2006 the average specific water consumption in the Heineken group was 5.22 hL/hL and for a brewery the water use was broken down as follows: raw materials 1.3 hL/hL; cleaning 2.9 hL/hL; heat transfer 0.7 hL/hL; other uses 1.6 hL/hL.

Carlsberg's Environmental Report (2003-2004) provided a weighted average specific water consumption of 4.7 hl/hl; whereas in 2004 the Asahi breweries group a value of 6.3 L/L (Asahi Breweries, 2005) and in 2009 ABInBev reported a value of 4.32 L/L (ABInBev 2010)

Table 1.15 *Specific water consumption expressed as hL of beer produced by some international brewery groups.*

Brewery	Water Consumption (hL/hL)	Year
SAB Miller	4.56	2006
Heineken	5.22	2006
Carlsberg's	4.7	2004
Asahi	6.3	2005
InBev	4.32	2009

1.5.4 Wastewaters

The pollutant load of a brewery effluent is primarily composed of organic materials from process activities. Brewery processes also generate liquids, such as the weak wort (that is, the wort remaining in the lauter tun, having a low extract content), and residual beer, which the brewery should reuse rather than disposing off.

Residual beer derives from diatomaceous earth filters, process tanks, pipes, beer rejected in the packaging area, returned beer, and broken bottles in the packaging area. It equals to 1-5% of total beer production, sometimes even more.

Effluent discharge from a brewery vary from 3 to 5 m³ per m³ of sold beer, not including cooling waters (Table 1.16).

Untreated effluents typically contain suspended solids in the range of 10-60 mg/L, biochemical oxygen demand (BOD) in the range of 1,000–1,500 mg/L, chemical oxygen demand (COD) in the range 1,800-3,000 mg/L, and nitrogen in the range of 30–100 mg/L. Phosphorus can also be present at concentrations of 10–30 mg/L. Effluent pH averages about 7 for the combined effluent, but can fluctuate from 3 to 12 depending on the use of acid and alkaline cleaning agents. The average temperature of brewery effluent is about 30 °C.

Table 1.16 *Effluent indicators for efficient breweries (IFC 2007).*

Pollutants	Units	Guideline Value
pH	pH	6 – 9
BOD ₅	mg/l	25
COD	mg/l	125
Total nitrogen	mg/l	10
Total phosphorus	mg/l	2
Oil and grease	mg/l	10
Total suspended solids	mg/l	50
Temperature increase	°C	<3 ^b
Total coliform bacteria	MPN ^a / 100 ml	400
Active Ingredients / Antibiotics	To be determined on a case specific basis	
Notes: ^a MPN = Most Probable Number ^b At the edge of a scientifically established mixing zone which takes into account ambient water quality, receiving water use, potential receptors and assimilative capacity		

1.5.5 Solid Wastes and By-products

Beer production results in a variety of residues, such as spent grains, which have a commercial value and can be sold as by products to the agricultural sector, but also hot-trub (0.2-0.4% of wort volume, with a dry matter content of 15-20%) and yeast (the surplus yeast and spent yeast slurry is 2-4 kg, 10-15% dry, per hL of beer produced); spent hops; used kieselguhr, and other solid wastes associated with the process, like broken bottles that cannot be recycled to the process, and cardboard. It is very important to collect as much surplus yeast as possible to avoid high BOD levels in the wastewater, with a BOD value of 120,000-140,000 mg/l, and also yeast suspension contain 1-2% of beer which can be recovered and recycled.

Table 1.17 *By-products for 100 L of beer produced. (UNEP, 1996)*

By-Products	Amount
Spent grains	14-19 kg/hL beer
Weak wort	2-6% of wort volume
Trub	0.2-0.4% of wort volume
Yeast	2-4 kg/hL beer

Tables 1.16 and 1.17 provide examples of by-product and effluents production indicators for efficient breweries, as extracted from IFC (Anon, 2007) ; while Table 1.18 shows an estimate of the solid wastes resulting from brewing. Guideline values are indicative of good international industry practice. These guidelines are achievable under normal operating conditions in appropriately designed and operated facilities through the application of pollution prevention and control techniques.

Table 1.18 Typical solid waste generation in beer production. (UNEP, 2006)

Solid waste	kg/hL of beer
Broken glass	0.85
Treatment sludge	0.1-0.8
Labels, paper	0.29
Kieselguhr slurry, dry	0.25
Cardboard, carton	0.04
Metals	0.02
Plastic	0.5

Auxiliary materials, like kieselguhr, caustic soda and CO₂, are of concern from the safety and environmental point of view. A typical kieselghur consumption for filtering beer is about 100-300 g/hL, for caustic soda the value is 0.5-1.0 kg/hL.

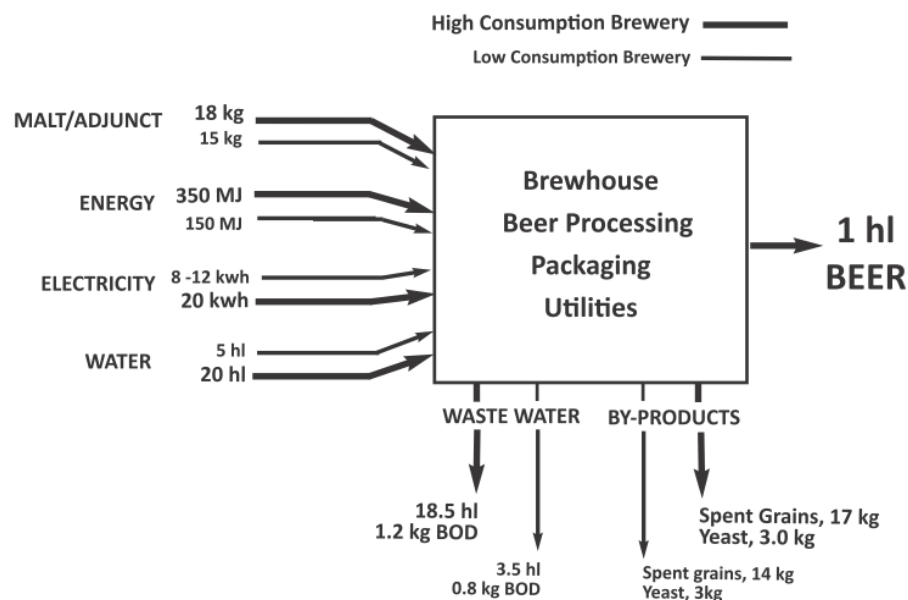


Figure 1.41 Resources used by a high- and low-consumption brewery. (UNEP, 1996).

The fermentation process generate about 3-4 kg CO₂ per hL of wort, depending on the wort strength. Many breweries recover part of this CO₂, otherwise it is vented to atmosphere. The CO₂ generated in boilers (using fuel oil – 200 MJ/hL) is around 16 kg/hL.

1.5.6 Carbon footprint and environmental product declaration

Several studies have been so far carried out to understand and verify the environmental impact generated through the entire life cycle of some lager beers in Denmark (Frees et al, 1998), Estonia (Talve, 2001), Greece (Koroneos et al, 2005), Italy (Cordella et al, 2008), and Spain (Hospido et al, 2005). Also a few environmental product declarations (EPD, 2010, 2011ab) provided reliable quantification and certification of the environmental performance of lager-type beers packed in disposable glass bottles of 33 cL (loose for sale on Ho.Re.Ca channel, in selling unit of 3 bottles for mass retail channel), steel kegs of 25 L, or Draught Master Modular 20 drums of 20 L, according to the Life Cycle Assessment methodology (ISO 14040-14044).

As an example, Table 1.19 shows the main environmental impact categories (i.e., Global Warming Potential, GWP, in 100 year perspective; acidification potential; ozone depletion potential, ground level ozone; eutrophication potential) related to a functional unit of 100 L of Tuborg® beer bottled/barrelled and delivered to final consumers (at pubs, bars, restaurants) (EPD, 2011a).

It can be noted that in agreement with Hospido et al. (2005) and Talve (1999) the most relevant environmental impact of the agricultural subsystem is the eutrophication potential of about 15 kg of PO₄⁻³_e per hL of beer, owing to the release of nitrogen and phosphorous from production and use of fertilizers.

The CO₂ equivalent emissions were greatly affected by the packaging system used. They ranged from about 106 kg/hL of beer barreled in plastic kegs (mainly made of 70.7% PET, 20% polypropylene, and 4.7% nylon) to 149 kg/hL of beer barreled in steel kegs, or to 190 kg/hL of beer bottled in glass bottles (EPD, 2011a).

Whereas the emissions due to the production (core) process represented as little as 14.8-17.7% of the overall ones, those associated to the upstream production or downstream processes were the greatest ones when including glass bottle production or steel keg distribution, respectively (Table 1.19).

Table 1.19 *Main environmental impact categories related to a functional unit of 100 L of Tuborg® beer (EPD, 2011a).*

BEER IN STEEL KEG				
Impact Categories	Entire Life Cycle	Upstream processes	Core process	Downstream Processes
Global Warming Potential (kg CO ₂ eq.)	149,05	49,52	24,35	75,18
Acidification Potential (kg SO ₂ eq.)	1,56	0,96	0,09	0,51
Ozone depletion potential (kg CFC 11 eq.)	0,00	0,00	0,00	0,00
Ground level ozone Potential (kg C ₂ H ₄ eq.)	0,54	0,50	0,01	0,03
Eutrophication potential (kg PO ₄ ⁻³ eq.)	15,58	15,58	0,00	0,00

BEER IN DM MODULAR 20 KEG				
Impact Categories	Entire Life Cycle	Upstream processes	Core process	Downstream Processes
Global Warming Potential (kg CO ₂ eq.)	105,55	56,65	17,74	31,15
Acidification Potential (kg SO ₂ eq.)	1,35	1,03	0,10	0,22
Ozone depletion potential (kg CFC 11 eq.)	0,00	0,00	0,00	0,00
Ground level ozone Potential (kg C ₂ H ₄ eq.)	0,52	0,50	0,01	0,01
Eutrophication potential (kg PO ₄ ⁻³ eq.)	15,53	15,53	0,00	0,00

BEER IN GLASS BOTTLE				
Impact Categories	Entire Life Cycle	Upstream processes	Core process	Downstream Processes
Global Warming Potential (kg CO ₂ eq.)	190,18	131,31	28,22	30,65
Acidification Potential (kg SO ₂ eq.)	1,95	1,63	0,11	0,21
Ozone depletion potential (kg CFC 11 eq.)	0,00	0,00	0,00	0,00
Ground level ozone Potential (kg C ₂ H ₄ eq.)	0,55	0,52	0,01	0,02
Eutrophication potential (kg PO ₄ ⁻³ eq.)	15,14	15,14	0,00	0,00

Another study was performed by the Climate Conservancy (2008) in cooperation with New Belgium Brewing Company to assess greenhouse gases (GHG) emitted across the full life cycle of Fat Tire® Amber Ale (FT) using the British Standard Publicly Available Specification (PAS 2050) methodology (BSI, 2008a) as detailed in the Guide to PAS 2050 (BSI, 2008b). The system boundaries of the life cycle study included acquisition and transport of raw materials, brewing operations, business travel, employee commuting, transport and storage during distribution and retail, use and disposal of waste. The carbon footprint (CF) of a 6-pack of the Amber Ale (that is, six glass bottles of 12 fluid ounce capacity each, packaged together in a paperboard carrier, equivalent to a beer volume of 6 x 12 x 29.57 = 2129.04 mL) was estimated as equal to 3,188.8 g of CO₂ equivalents (CO_{2e}). In this case, the CF was about 150 kg CO_{2e}/hL of beer bottled in glass bottled, quite smaller than the value (190 kg/hL) reported in Table 1.19 (EPD, 2011a).

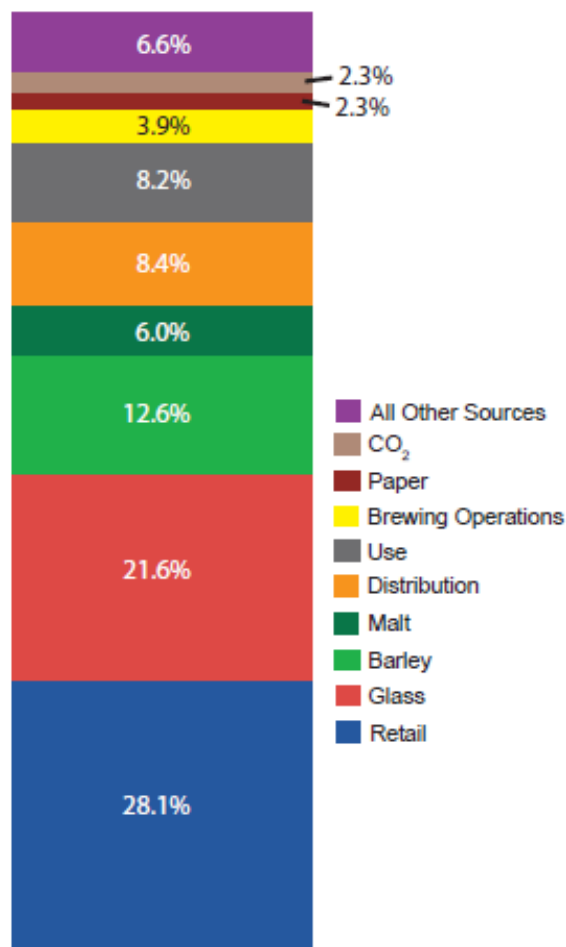


Figure 1.42 Carbon Footprint of Fat Tire® Amber Ale showing major sources of GHG emissions by percentage of total emissions (Climate Conservancy, 2008).

Of the total (GHG) emissions, New Belgium Brewing Company processing and waste disposal accounted for only 173.0 g CO_{2e}, or 5.4%. Upstream emissions during production and transportation of packaging materials and beer ingredients add up to 1,531.3 g CO_{2e}, or 48.0% of total emissions. Downstream emissions from distribution, retail, storage and disposal of waste account for the remaining 1,484.6 g CO_{2e}, or 46.6% of the total. The largest line item in the overall GHG emissions is electricity used for refrigeration at retail: 829.8 g CO_{2e}. The next largest sources are production and transportation of glass and malt (including barley): 690.0 and 593.1 g O_{2e}, respectively. These three sources alone account for 68.4% of all emissions embodied in a 6-pack of FT. The bulk of remaining emissions (25.1% of total) are accounted for by production and transportation of paper and CO₂ for carbonation, refrigeration in consumer's homes, distribution transport, and natural gas consumed during

brewing operations. As can be easily assessed by comparing the CF data (Climate Conservancy, 2008) to those reported in Table 1.19 and referred to Tuborg® beer (EPD, 2011a), it is unclear which is the effective carbon footprint of a liter of beer and its environmental impact. With regard to the comparison between the three packaging options compared in Table 1.19, beer in the novel plastic kegs turned out to cause a lower environmental load along its life cycle than beer in steel keg and in bottle due to the fact that higher emissions and higher energy consumptions were associated to the allocated glass bottles. This conclusion, as extracted from EPD (2010, 2011ab), is in line the results reported by Cordella et al. (2008), Koroneos et al. (2005), and Talve (2001), that pointed out the relevance of bottle packaging and agriculture on the overall life cycle of the beer. In the circumstances, Mata and Costa (2001) assessed the environmental impact of different reuse percentages for glass beer bottles. The advantages of the use of returnable bottles over that of non-returnable ones increase with the number of cycles performed by the returnable bottles. In the case of a 50% reuse, i.e. the same number of returnable and non-returnable bottles, the contribution of returnable bottles to global warming, acidification, photochemical ozone creation, critical air and water volume, human toxicity, energy and raw-material consumption is smaller than that of the non-returnable bottles after the second reuse. On the contrary, the contribution of returnable bottles to eutrophication, ozone depletion, solid waste, water and auxiliary material consumption is larger even after several reuse (Mata and Costa, 2001). Thus, the optimal reuse percentage should be identified by accounting not only for the environmental, but also for the economic, technological and social implications of the different alternative distributions of beer in returnable or non-returnable bottles. In any case, the information currently available might roughly suggest to consumers and producers a more responsible consumption and a more environmentally friendly production of beer through the use of draught beer (that is, the beer drawn from a plastic keg) and of reusable packaging. Further studies about the environmental impact of beer production are however needed to assess clearly the relative contribution of all phases of beer life cycle since the use of qualitative data and/or the incompleteness of certain other data does not allow any direct comparison among the environmental scores available in the literature.

CHAPTER

2

MATERIALS and METHODS

2.1 Raw Materials

Three different types of beer were used in this work. The optimal operating conditions for beer clarification via CFMF were assessed using rough beer A as such, and were confirmed using rough beers B and C, as such or after pretreatments.

Rough Beer A

Rough beer (labelled A) was produced in the laboratory-scale, in lots of 25 litres. A commercial hopped-malt extract (Pils, Brewferm, Beverlo, B) was diluted with tap water at 80 °C to a density of 1.045 kg/L. As cooled down to 20 °C and oxygenated, the resulting wort was inoculated with 11.5 g of dry ale yeast (Safale S-04, Fermentis, Marcq-en-Barœul, F). The fermentation temperature was kept constant at ~ 20 °C for about 4 days, then gradually lowered to 15 °C over the following 4 days. The phase maturation was prolonged for about 30 days. After racking, the rough beer was stored in a stainless-steel maturation vessel under a slight CO₂ overpressure at 4.0 ± 0.5 °C.

Rough Beer B

The rough beer (labelled B) was produced in 25-L lots in the pilot-scale brewery c/o CERB (Perugia, Italy). Each wort (density: 1.045 kg L⁻¹) was obtained by mashing 100% pils malt (Durst-Malz, Bruchsal-Heidelsheim, Bruchsal, D), and hopping with traditional bitter Hallertau Magnum hop pellets. Once the wort had been cooled and oxygenated, its fermentation was started by adding 11.5 g of dry yeast (Saflager W-34/70, Fermentis, Marcq-en-Barœul, F). The fermentation was carried out at ~12 °C for about 10 days, then temperature was gradually lowered to (2 – 4) °C over the following 4 days. After racking, rough beer was stored in a stainless-steel maturation vessel at 4 °C for about 30 days. After racking, the rough beer was stored in a stainless-steel maturation vessel under a slight CO₂ overpressure at 4.0 ± 0.5 °C.

Rough Beer C

The rough beer (labelled C) was produced in the industrial-scale brewery Birra Peroni Srl (Rome, Italy). The Peroni lager beer is the company's original brand, the second best selling pale lager in Italy (Assobirra, 2012), and is obtained from a decoction mash using barley malt, maize grits and hop pellets extract. This lager beer was obtained direct from the

brewery's maturation vessel, just before the filtration step and stored in a stainless-steel maturation vessel at 4 °C for no more than 30 days.

2.2 Equipment and experimental procedure

A typical temperature- and pressure-controlled bench-top CFMF plant (Fig. 2.1a) was used (Cimini & Moresi, 2013). It was equipped with ceramic tubular membrane modules (US Filter, Warrendale, PA, USA), with 6-mm inside diameter, 500-mm length, and 94.2-cm² effective membrane surface area, and porosity of 0.4, 0.8, or 1.2 µm. During their use, the water permeability at 20.0±0.1 °C for the 0.4-, 0.8-, or 1.2-µm membrane module was 521±37, 773±17, or 1716±44 L m⁻² h⁻¹ bar⁻¹ (r² =0.99), respectively. Both digital pressure transducers (Imssystem, Cagliari, Italy) and Bourdon manometers (OMET di Ceresa Srl, Pessano con Bornago, Milan, Italy), with a maximum pressure of 6 bar, were attached at the feed inlet, and retentate and permeate outlets of any MM. The process temperature was monitored by a digital temperature indicator (TI) and controlled by a thermostat (type LTD6, Grant Instrument Ltd., Cambridge, UK), this regulating automatically the flow rate of the cooling fluid (a mixture of water and ethylene glycol) through a stainless-steel plate heat exchanger (E1). Its overall heat transfer surface area was 0.36 m², while the maximum feed flow rate and pressure were 4 m³ h⁻¹ and 30 bar, respectively. The rotameter FI01 (type E5-2800/H, ASA Srl, Sesto San Giovanni, Italy) was used to measure the feed volumetric flow rate (QF) in the range of 0.1 to 1.0 m³ h⁻¹, while the rotameter FI02 (type E5-2600/H, ASA Srl, Sesto San Giovanni, Italy) allowed the permeate flow rate to be determined in the range of (2–40) L h⁻¹. The retentate flow rate was measured by the digital flowmeter transducer FI03 in the range of 0.1 to 1.8 m³ h⁻¹. The permeate flow rate was also assessed by using two technical-grade scales depending on the CFMF test performed. In particular, K1 or K2 was the type PCE-TS 150 (PCE Italia Srl, Gragnano, Lucca, Italy) or Europe 4000 AR (Gibertini, Elettronica Srl, Novate, Milan, I), its accuracy and maximum capacity being ± 20.0 or 0.01 g and 150 or 4 kg, respectively. Both scales were interfaced to a personal computer (PC) via RS-232 serial ports.

When the 25-L AISI 304 storage tank D1 had been charged with about 5 L of rough beer, the centrifugal pump G1 (type HMS, maximum volumetric flow rate of 4.20 m³ h⁻¹, head of 40 m of water and power of 0.45 kW; Lowara, Montecchio Maggiore, Italy) was switched on. To

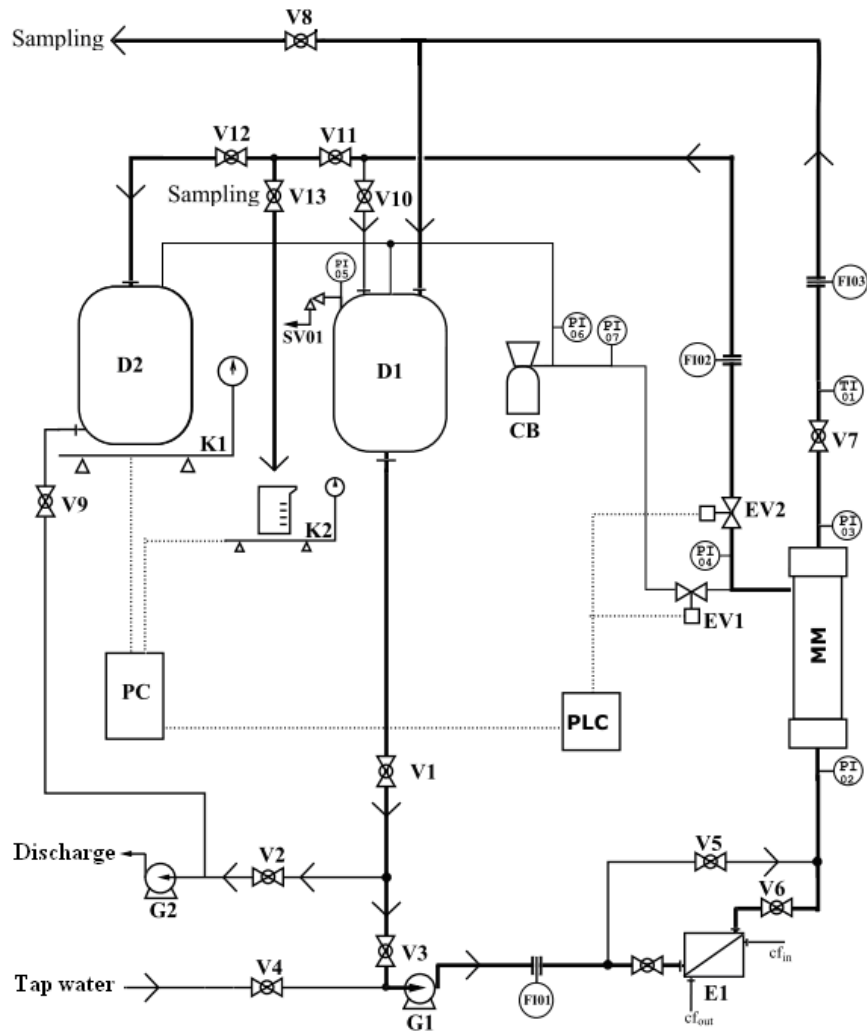
assure simultaneous setting of Q_F and TMPD, the manual ball valve (V7) was regulated while varying the frequency of the electric voltage applied to the asynchronous motor piloting G1 by means of the frequency inverter VF (type Commander SK 0.75 k, Control Techniques, Powys, UK). All the other stainless steel ball valves shown in Fig. 1a allowed the feed to be charged (V8); the retentate to be discharged (V2); the permeate to be recycled back into D1 (V10), accumulated into D2 (V11 and V12) or sampled via V13, as well as a series of other ancillary operations (such as valve, membrane module, or pump replacement) to be performed.

A 4 kg liquid CO₂ bottle (CB) at an average pressure of 200 bar was used to ensure an inert atmosphere in both tanks D1 and D2, as well as in the permeate circuit, and minimise O₂ pick-up. Moreover, by using a programmable logic controller-based process (PLC), it was possible to open or close automatically the electrovalves EV1 and EV2 and set the pressure in the permeate side of MM at a higher value than that in the retentate side for prefixed time intervals. In this way, different periodic CO₂ backflushing cycles in the membrane module used are periodically carried out at a backflush pressure difference between the permeate and retentate sides of +3 bar for 2 min. The permeate flow rates were assessed via electronic balances K1 and K2, both connected to a PC. Total recycle runs were carried out at ~10 °C by varying TMPD and v_s in the ranges of 1-5 bar, and 2-6 m/s, respectively.

Several total recycle runs were carried out at ~10 °C by setting TMPD and v_s at 3.74 bar, and 6 m s⁻¹, respectively. As the permeation flux (J_v) was approaching the quasi-steady state flux (J^*), the difference in the subsequent J_v values tended to zero. When the absolute value of such differences was smaller than a prefixed empirical value of the order of 5%, the pressure in the retentate side was manually reduced to 1 bar; then, the programmable controller automatically opened EV1 and closed EV2, each backflushing cycles differing even during a single filtration trial.

As suggested by Gan *et al.* (1999), membrane cleaning included a combined synergic caustic and oxidation cleaning, followed by acidic cleaning

Fig. 2.1a)



2.1b)

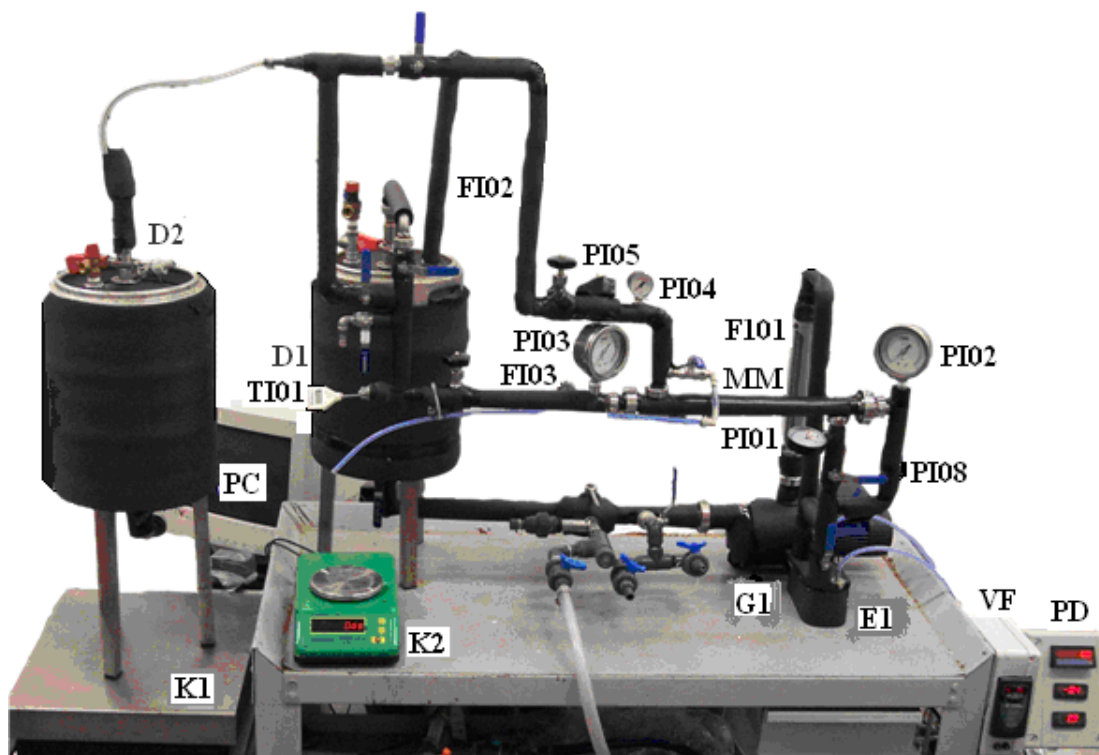


Figure 2.1 Process flow sheet (a) and photograph (b) of the bench-top membrane plant used in this work. Equipment and ancillary fluid identification items: CB, liquid CO₂ bottle; D, rough beer and permeate storage tank; cf, cooling fluid; E, plate heat exchanger; EV, electrovalve; FI, rotameter; G, centrifugal pump; K, balance; MM, CFMF membrane module; PC, personal computer; PD, control and display cabinet; PI, manometer; PLC, programmable logic controller for automatic opening or closing of the electrovalves used to perform periodic CO₂ backflushing; SV, safety valve; TI, temperature indicator; V, manual ball valve; VF, frequency inverter.

Beer pretreatment and stabilization procedure

The rough beers A-C were used as such or after pretreatments. Some samples were mashed by adding 0.15 mL of a commercial Beerzym PENTA preparation (Erbslöh Geisenheim AG, Geisenheim, Germany) per L of rough beer at 4 °C for 24 h to degrade almost all the pentosans and β -glucans present. This Enzymated beer (RBE), was used such as or centrifuged (RBEC) in 0.3-L plastic bottles using a laboratory centrifuge (Beckman mod. J2-21) at 6000 x g and less than 4 °C for 10 min.

For a good implementation of the PVPP stabilization step onto the CFMF process, a preliminary test was carried out to determine the kinetics of total phenols removal. To this end, samples (100 mL) of EC-pretreated rough beer B were poured into 100-mL jacketed beakers placed over a magnetic multistirrer (model 15, Velp Scientifica, Milan, Italy) to maintain the reaction temperature at $[(20.0 \text{ or } 1.5) \pm 0.2]$ °C by means of a cryothermostat. After seeding each sample with 0.05 g of regenerable PVPP granules (size range: 50-250 µm), several sampling (5 mL) were withdrawn at time intervals of 1 min for the first 5 min, then at time intervals of 10 min till 1 h, in order to determine the residual content of polyphenols, as reported below.

Having assessed the quick kinetics of this stabilization procedure, two different methods were used:

Firstly, the EC-pretreated rough beer B accumulated in the feed tank of the bench-top CFMF plant was *in situ* enriched with 0.3-0.5 g L⁻¹ of regenerable PVPP granules, and the resulting suspension was directly submitted to total recycle CFMF tests under the aforementioned operating conditions. Throughout the CFMF test, permeate samplings allowed the residual total phenol content, haze, and alcohol chill haze to be determined at different contact times.

Secondly, the EC-pretreated beer (B and C) was charged into 1.5-L cylindro-conical tanks, supplemented with 0.5 g L⁻¹ of regenerable PVPP, manually mixed once, and kept at (0.0 ± 0.5) °C for about 24 h. As spent PVPP-polyphenol complexes tended to sink to the bottom of each tank, two discard specimens of about 60 g each were collected at contact times of 18 and 24 h. The remaining liquids were then collected, and analyzed for haze and total phenols. After vacuum filtering through 2.7-µm Whatman filters (cat. No. 1823 047), the stabilized beer (B and C) (RBECS) was finally submitted to total recycle CFMF runs, as reported above.

2.3 Analytical Methods

The beer or permeate samples were assayed for pH, density (ρ), viscosity (η), turbidity or haze (H) at (20 and/or 0) °C, alcohol chill haze (ACH), color (C), as well as β -glucans (BG) and total phenol (TP) contents, real (RE) or original (OE) extract, and ethanol (A), in accordance with Analytica EBC (2010). According to the European Brewery Convention, the standard haze for DE-filtrated beer should be less than 0.6 EBC unit, the turbidity of 0.5 EBC unit being referred as A1-Brilliant (Analytica EBC, 2010). Some variability from batch to batch and within the same beer batch, as stored in the maturation tank, was noticeable. The main characteristics (i.e., pH, A, C, OE, RE, BG, TP) of the rough beer samples used, are summarized in Table 2.1

Tab 2.1 *main characteristics of rough beer A,B and C used in this PhD thesis.*

Sample	pH	ρ	η	H	C	BG	RE	OE	A	TP
	[-]	[kg m ⁻³]	[mPa s]	[EBC]	[EBC]	[g L ⁻¹]	[°Plato]	[°Plato]	[% v/v]	[mg/L]
A	4.2 – 4.3	1.013 –1.016	1.38 -1.59	0,72-61,9	16,1-27,7	197-154	3.0 – 4.4	10.2-14.2	3.00 -4.50	n.d
B	4.2 – 4.4	1.008 –1.009	1.38-1.41.	5 - 34	5.1-7.6	140-250	2.9 -3.8	12.6 -13.4	5.1 -5.7	176-224
C	4.2 –4.3	1.007 -1.008	1.38-1.39	3.5-25	7.5 $\pm$ 1.0	17 $\pm$ 0.1	3.4 $\pm$ 0.1	10.65 $\pm$ 0.15	4.70 $\pm$ 0.1	261-165

Table 2.2 *EBC analytical methods used*

ANALYSIS	METHOD	REF.
Specific Gravity of beer	Pyknometer	ANALYTICA EBC 9.43.1 / 2004
Colour of beer	Spectrophotometric	ANALYTICA EBC 9.6 / 2000
pH of Beer	pH meter	ANALYTICA EBC 9.35 / 2004
Viscosity of Beer	Glass Capillary Viscosimeter	ANALYTICA EBC 9.38 / 1997
Haze in beer	Haze meters	ANALYTICA EBC 9.29 /1997
β -Glucans	Enzymatic	ANALYTICA EBC 8.11.1
Alcohol in beer	Distillation	ANALYTICA EBC 9.2.1/2008
Alcohol in beer	Enzymatic method	Megazyme (K-ETOH)
OG, Original, Real, Ap. Extract	Densimetric and distillation	ANALYTICA EBC 9.4 / 2004
Shelf-life of beer	Haze formation	ANALYTICA EBC 9.30/1997
Alcohol Chill haze in Beer	Test Chapon	ANALYTICA EBC 9.41/2004
Foam stability	Rudin Test	

2.4 Statistical analysis of data

As suggested by Montgomery (2005), when dealing with complex and long-running experimental trials, one or more than one specific trial was replicated so as to estimate the mean values and standard deviations of all the dependent variables, and thus assess the error variance for all the experimental campaign. In this specific case, the average coefficient of variation in the measured permeate volume (V) and estimated permeation flux (J_v) within data population was of 7 and 9%, respectively.

As concerning the main properties of rough beer, as such or pretreated, as well as its permeate, these were measured at least three times, and their means used for further analysis.

Regression analysis of the non-linear regression models reported in table 1.10 was carried out by using the built-in nonlinear regression functions of the software Microsoft Excel (Microsoft, Redmond, CA, USA). The residual variance (s_j^2) and mean squared percentage error (MSPE_j) for any dependent variable y_j according to Eq.s (1.15) and (1.34) were estimated as follows:

$$s_j^2 = \sum_{i=1}^N \frac{(y_{j,i,calc} - y_{j,i,exp})^2}{N - p} \quad (2.1)$$

$$MSPE_j = \sqrt{\frac{\sum_{i=1}^N \left(\frac{y_{j,i,calc} - y_{j,i,exp}}{y_{j,i,exp}} \times 100 \right)^2}{N}} \quad (2.2)$$

where $y_{j,i,calc}$ and $y_{j,i,exp}$ are the i -th instantaneous calculated and experimental values of the dependent variable y_j selected (i.e., V or J_v), N is the number of the experimental data, and p ($=1$) the number of independent parameters of the CF fouling model under testing.

CHAPTER

3

RESULTS and DISCUSSIONS

3.1 Rough Beer A

Assessment the Optimal Operating Conditions for CFMF

Table 3.1 summarizes the average characteristics of the rough beers A (RB1-RB16) tested in this study, as well as those of their corresponding permeates (BP1-BP16).

Owing to the natural sedimentation of suspended matter in the green beer storage tank at 4 °C, some variability from batch to batch and within the same beer batch was noticeable, the turbidity value varying from (0.99 ± 0.06) to (61.9 ± 1.3) EBC unit. The European Brewery Convention recommends a standard haze for DE-filtrated beer of less than 0.6 EBC unit, the turbidity of 0.5 EBC unit being generally referred as A1-Brilliant (Analytica EBC, 2010). In this work, all beer permeates exhibited smaller turbidity levels at 20 °C than 0.6 EBC unit. As expected, the alcohol content (A) of micro-filtered beer was not affected by the CFMF process. On the contrary, the retention of polysaccharides and proteins by CFMF processes has been so far definitely established in the technical literature (Fillaudeau & Carrère, 2002; Gan *et al*, 2001), this being indirectly measured by the lessening in density, original and real extracts, and colour in micro-filtered beer (Table 3.1). The residual microbial density was not assessed at this stage of the experimentation, but the retention of yeast cells being quite near to 100% and residual yeast cell number of the order of 10 cell/mL (Fillaudeau *et al*, 2007).

3.1.1 Effect of TMPD and v_s on permeation flux

In agreement with the industrial CFMF application at the Heineken brewery (Noordman *et al*, 2001), the permeation flux of rough beer was initially studied after installing the 0.8- μm ceramic tubular membrane module and setting TMPD and v_s at ~ 1.96 bar and 2 m s^{-1} , respectively (Fig. 3.1). Independently of the high or low turbidity level of the rough beer sample under treatment (Table 3.1), in about half an hour J_v tended to an asymptotic value, generally called *limiting* or *quasi-steady state permeation flux* (J^*), of $20 \pm 2 \text{ L m}^{-2} \text{ h}^{-1}$. This flux depended on the fluid dynamic conditions inside the tubular membrane and membrane fouling mechanism, and agreed with the 10-h average flux of $22 \text{ kg m}^{-2} \text{ h}^{-1}$ previously achieved by Gan *et al*. (2001) and Sondhi & Bhavé (2001).

Table 3.1 Mean and standard deviations of the main characteristics (pH; density, ρ ; viscosity, η ; turbidity at 20 °C, H; colour, C; β -glucans, BG; real extract, RE; original extract, OE; alcohol content, A) of the rough beers A, as such (RB1-14) or precentrifuged (RB15-16) and their corresponding permeates (BP), assayed in this work.

Sample	ΔP	v_s	T	pH	ρ	η	H	C	BG	RE	OE	A
	[bar]	[m/s]	[°C]	[-]	[kg m ⁻³]	[mPa s]	[EBC]	[EBC]	[g m ⁻³]	[°Plato]	[°Plato]	[% v/v]
RB1	1.97	2	10	4.30 ± 0.01	1013 ± 1	1.42 ± 0.01	12.2 ± 0.7	16.1 ± 0.5	160 ± 3	3.8 ± 0.1	11.7 ± 0.1	4.00 ± 0.02
BP1				4.30 ± 0.01	1008 ± 1	1.38 ± 0.01	0.5 ± 0.04	11.7 ± 0.5	n.d	2.7 ± 0.1	10.3 ± 0.1	4.00 ± 0.02
RB2	1.97	2	10	4.20 ± 0.01	1016 ± 1	1.433 ± 0.02	1.16 ± 0.02	17.5 ± 0.5	197 ± 4	4.2 ± 0.1	10.3 ± 0.1	3.03 ± 0.02
BP2				4.20 ± 0.01	1012 ± 1	1.285 ± 0.07	0.45 ± 0.05	10.9 ± 0.5	170 ± 7	2.6 ± 0.4	8.8 ± 0.3	2.99 ± 0.02
RB3	1.97	2	10	4.20 ± 0.01	1016 ± 1	1.46 ± 0.01	1.14 ± 0.02	18.1 ± 0.5	197 ± 4	4.4 ± 0.1	10.2 ± 0.1	3.00 ± 0.02
BP3				4.20 ± 0.01	1012 ± 1	1.35 ± 0.07	0.4 ± 0.05	14.7 ± 0.5	170 ± 7	3.0 ± 0.4	8.5 ± 0.3	3.00 ± 0.02
RB4	0.97-3.97	2	10	4.20 ± 0.01	1015 ± 1	1.417 ± 0.005	0.99 ± 0.06	22.8 ± 0.5	197 ± 4	4.4 ± 0.1	10.2 ± 0.1	3.00 ± 0.02
BP4				4.20 ± 0.01	1015 ± 1	1.405 ± 0.003	0.39 ± 0.06	15.7 ± 0.5	170 ± 7	3.0 ± 0.4	8.5 ± 0.3	3.00 ± 0.02
RB5	0.97-4.73	2, 4, 6	10	4.20 ± 0.01	1015 ± 1	1.408 ± 0.003	5.89 ± 0.02	16.0 ± 0.1	197 ± 4	4.4 ± 0.1	10.2 ± 0.1	3.00 ± 0.02
BP5				4.20 ± 0.01	1014 ± 1	1.392 ± 0.01	0.40 ± 0.01	15.4 ± 0.1	170 ± 7	3.0 ± 0.4	8.5 ± 0.3	3.00 ± 0.02
RB6	0.97-4.73	2, 4, 6	10	4.20 ± 0.01	1015 ± 1	1.408 ± 0.003	14.40 ± 0.01	16.0 ± 0.1	197 ± 4	4.4 ± 0.1	10.2 ± 0.1	3.00 ± 0.02
BP6				4.20 ± 0.01	1014 ± 1	1.392 ± 0.010	0.50 ± 0.02	15.4 ± 0.1	170 ± 7	3.0 ± 0.4	8.5 ± 0.3	3.00 ± 0.02
RB7	0.97-4.73	2, 4, 6	10	4.33 ± 0.01	1012 ± 1	1.405 ± 0.002	24.8 ± 0.1	27.7 ± 0.1	194.2 ± 0.4	3.55 ± 0.01	19.4 ± 0.1	2.95 ± 0.04
BP7				4.32 ± 0.01	1011 ± 1	1.360 ± 0.003	0.37 ± 0.01	22.5 ± 0.3	154.5 ± 0.2	2.22 ± 0.01	18.0 ± 0.1	2.95 ± 0.04

RB8	3.73	6	10	4.20 ± 0.01	1016 ± 1	1.600 ± 0.001	1.40 ± 0.03	25.2 ± 0.5	197 ± 4	10.3 ± 0.1	18.8 ± 0.1	4.50 ± 0.06
BP8				4.20 ± 0.01	1015 ± 1	1.540 ± 0.004	0.50 ± 0.01	23.4 ± 0.5	170 ± 7	8.40 ± 0.04	16.9 ± 0.2	4.40 ± 0.02
RB9	3.73	6	10	4.33 ± 0.01	1016 ± 1	1.592 ± 0.007	3.69 ± 0.04	35.9 ± 0.1	194.2 ± 0.4	45.7 ± 0.1	14.2 ± 0.1	4.37 ± 0.09
BP9				4.32 ± 0.01	1013 ± 1	1.492 ± 0.002	0.44 ± 0.01	32.3 ± 0.1	154.5 ± 0.2	23.3 ± 0.1	11.7 ± 0.1	4.37 ± 0.09
RB10	3.73	6	10	4.31 ± 0.01	1012 ± 1	1.417 ± 0.001	9.6 ± 0.5	29.9 ± 0.2	194.2 ± 0.4	46.08 ± 0.04	16.0 ± 0.2	4.55 ± 0.08
BP10				4.32 ± 0.01	1010 ± 1	1.345 ± 0.004	0.44 ± 0.01	23.9 ± 0.3	154.5 ± 0.2	26.02 ± 0.07	15.9 ± 0.1	4.55 ± 0.09
RB11	3.73	6	10	4.31 ± 0.01	1012 ± 1	1.417 ± 0.001	18.0 ± 0.4	29.9 ± 0.2	194.2 ± 0.4	46.08 ± 0.04	16.0 ± 0.2	4.55 ± 0.08
BP11				4.32 ± 0.01	1010 ± 1	1.345 ± 0.004	0.45 ± 0.04	23.9 ± 0.3	154.5 ± 0.2	26.02 ± 0.07	15.9 ± 0.1	4.55 ± 0.09
RB12	3.73	6	10	4.33 ± 0.01	1013 ± 1	1.442 ± 0.002	21.3 ± 0.01	32.4 ± 0.1	194.2 ± 0.4	45.70 ± 0.01	14.2 ± 0.1	4.37 ± 0.09
BP12				4.32 ± 0.01	1010 ± 1	1.429 ± 0.001	0.45 ± 0.01	32.0 ± 0.1	154.5 ± 0.2	23.30 ± 0.01	11.7 ± 0.1	4.37 ± 0.09
RB13	3.73	6	10	4.31 ± 0.01	1012 ± 1	1.417 ± 0.001	53.8 ± 0.8	29.9 ± 0.2	194.2 ± 0.4	46.08 ± 0.04	16.0 ± 0.2	4.55 ± 0.08
BP13				4.32 ± 0.01	1010 ± 1	1.345 ± 0.004	0.47 ± 0.01	23.9 ± 0.3	154.5 ± 0.2	26.02 ± 0.08	15.9 ± 0.1	4.55 ± 0.09
RB14	3.73	6	10	4.33 ± 0.01	1016 ± 1	1.556 ± 0.001	61.9 ± 1.3	35.9 ± 0.1	194.2 ± 0.4	45.72 ± 0.05	14.2 ± 0.1	4.37 ± 0.09
BP14				4.32 ± 0.01	1013 ± 1	1.492 ± 0.002	0.45 ± 0.01	32.3 ± 0.3	154.5 ± 0.2	23.32 ± 0.01	11.7 ± 0.1	4.37 ± 0.09
RB15	3.73	6	10	4.20 ± 0.01	1016 ± 1	1.600 ± 0.007	1.00 ± 0.01	25.2 ± 0.1	194.2 ± 0.4	18.8 ± 0.1	10.3 ± 0.1	4.50 ± 0.06
BP15				4.20 ± 0.01	1015 ± 1	1.540 ± 0.002	0.40 ± 0.01	23.3 ± 0.1	154.5 ± 0.2	$21.6.9 \pm 0.2$	8.4 ± 0.1	4.40 ± 0.02
RB16	3.73	6	10	4.33 ± 0.01	1014 ± 1	1.520 ± 0.001	0.72 ± 0.02	32.4 ± 0.1	194.2 ± 0.4	45.72 ± 0.05	14.2 ± 0.1	4.37 ± 0.09
BP16				4.32 ± 0.01	1012 ± 1	1.494 ± 0.004	0.44 ± 0.03	32.0 ± 0.1	154.5 ± 0.2	23.32 ± 0.01	11.7 ± 0.1	4.37 ± 0.09

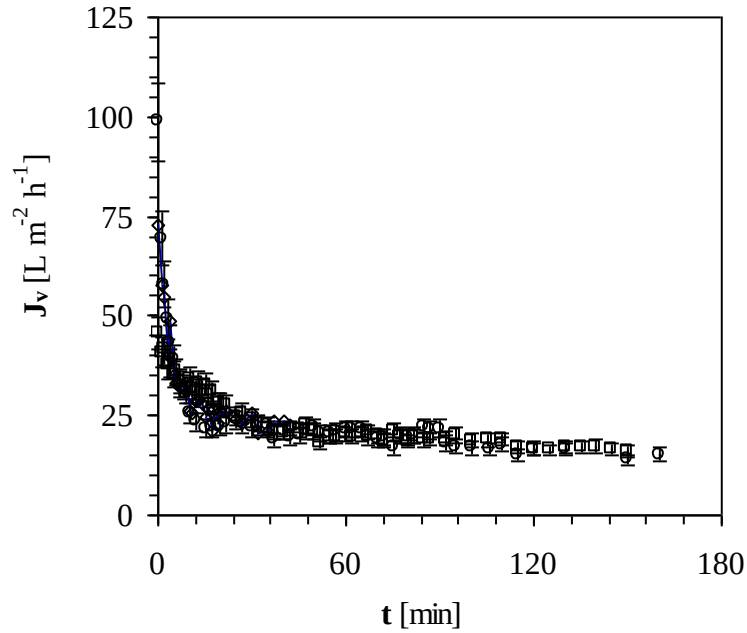


Fig. 3.1 Time course of the permeation flux (J_v) of a few rough beer A samples (RB1: $\circ$; RB2: $\square$; RB3: $\diamond$) at 10 °C when using the 0.8- μm ceramic tubular membrane module under constant TMPD (2 bar) and v_s (2 m s^{-1}). For all characteristics of rough beer samples see Table 3.1. The bar errors for J_v were $\pm 9\%$.

Fig. 3.2 shows the effect of TMPD in the interval of 0.96 to 3.97 bar at $v_s = 2 \text{ m s}^{-1}$ and $T = 10 \text{ }^\circ\text{C}$ on the permeation flux J_v of rough beer samples, the turbidity of which being equal to about 1.0 (RB4), 5.9 (RB5), or 14.4 (RB6) EBC unit. For the 0.8- μm pore membrane, J^* exhibited a significant increase at the 95% confidence level, being equal to (24 ± 2) , (31 ± 2) , (45 ± 8) , and $(68 \pm 20) \text{ L m}^{-2} \text{ h}^{-1}$ as TMPD was about 1, 2, 3, and 4 bar, respectively. This disagreed with previous observations in ceramic tubular membrane modules with nominal pore sizes of 0.4 and 0.6 μm (Alicieo *et al*, 2005), the observed increase in J^* having been insignificantly related to TMPD.

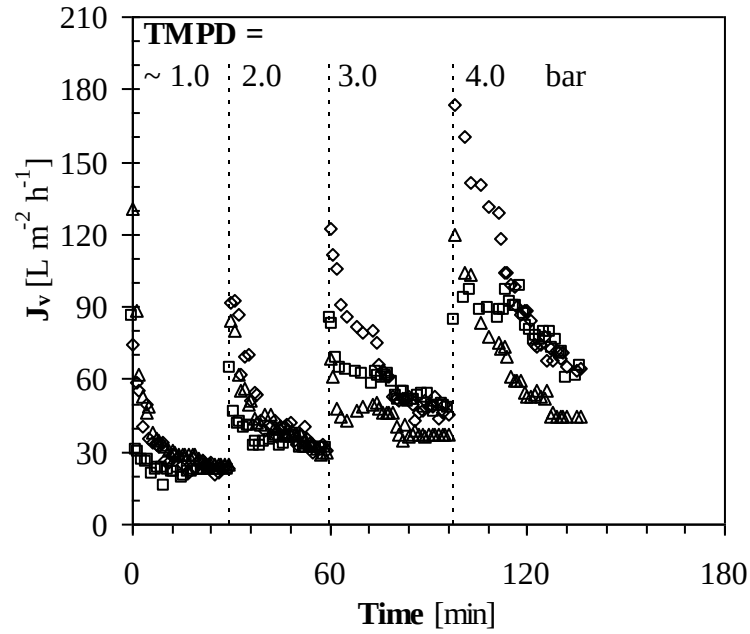


Fig. 3.2 Time course of the permeation flux (J_v) of a few rough beer A samples (RB4: $\diamond$; RB5: $\triangle$; RB6: $\square$) at 10 °C when using the 0.8- μm ceramic tubular membrane module under constant v_s (2 m s⁻¹) and TMPD increasing from about 1 to 4 bar. For all characteristics of rough beer samples see Table 3.1.

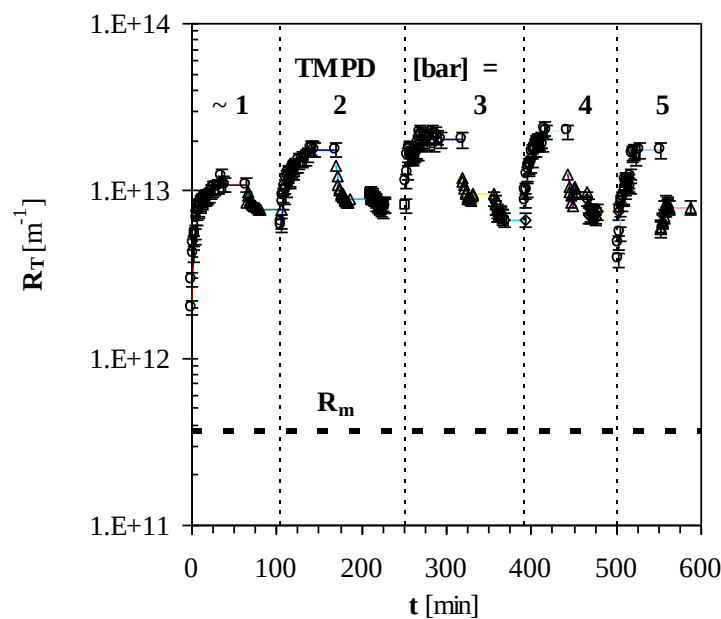
To confirm further our preliminary results, a few total recycle tests were carried out by submitting rough beer samples having turbidity of ~ 5.9 and 24.8 EBC unit to a step by step increase in TMPD from about 1 to 4 bar. Moreover, during any step the crossflow velocity (v_s) was sequentially raised from 2 to 4 and/or 6 m s⁻¹. To discriminate the fouling mechanisms, the flux decline behaviour during rough beer microfiltration can be examined by estimating the time course of the overall resistance (R_T) to filtrate flow as follows:

$$R_T = \frac{\text{TMPD}}{\eta_P J_v} \quad (3.1)$$

where η_P is the permeate viscosity, and TMPD the transmembrane pressure difference. According to Tracey & Davies (1994), the upward or downward concavity of the R_T -vs- t curve is related to internal fouling due to pore constriction or intermediate blocking or external fouling due to cake filtration. As shown in Fig. 3.3a, the total recycle CFMF test was started by setting TMPD and v_s at about 1 bar and 2 m s⁻¹, respectively. As time progressed, R_T

tended to increase till reaching a quasi-steady state value (R_T^*). A step increase in v_S under constant TMPD resulted in a fast decrease in R_T and led to a generally smaller R_T^* value. Any further increase in TMPD, while reducing again v_S to 2 m s^{-1} , replicated the aforementioned R_T trend, whatever the initial beer turbidity. For all the operating conditions examined, the ratio between any of their corresponding quasi-steady state overall membrane resistance (R_T^*) and the intrinsic membrane one ($R_m = 3.56 \times 10^{11} \text{ m}^{-1}$) was by far greater than unity. Actually, such a ratio also depended on the initial beer turbidity and varied in the range of about (19 – 64) or (40 – 200) for the low- or high- turbidity beer samples studied, respectively. In all tests, the minimum R_T^* values were associated to TMPD = (3 or 4) bar and $v_S = 6 \text{ m s}^{-1}$, these resulting in a limiting flux (J^*) of (112 ± 4) or $(140 \pm 5) \text{ L m}^{-2} \text{ h}^{-1}$, and of (59 ± 1) or $(62 \pm 1) \text{ L m}^{-2} \text{ h}^{-1}$ for the less or more turbid beer A used, respectively. Moreover, the time course of R_T was characterized by a downward concavity, this indicating generally fouling by cake filtration mechanism (Tracey & Davis, 1994). Further details about the fouling mechanisms will be reported below.

3.3a)



3.3b)

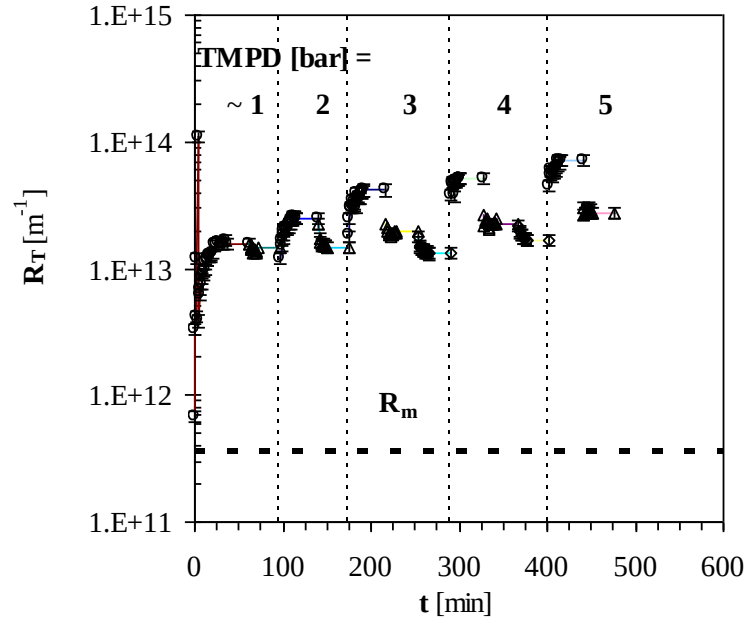
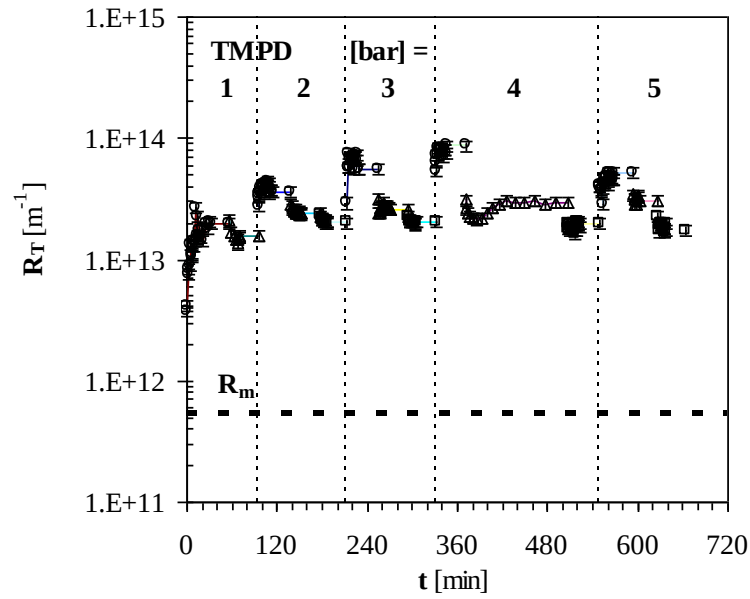


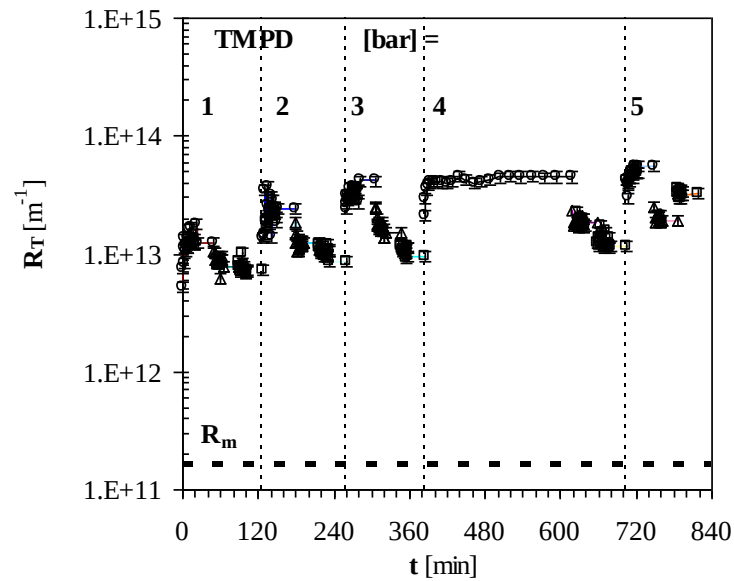
Figure 3.3 Time course of the overall membrane resistance (R_T) as resulting from a step by step increase in the transmembrane pressure difference (TMPD) and crossflow velocity (v_s) when dealing with two differently turbid rough beer A samples (a - RB5; b - RB7) and operating with the 0.8- μm ceramic tubular membrane module at 10 °C and different v_s values ($\circ$, 2 m s^{-1} ; $\triangle$, 4 m s^{-1} ; $\square$, 6 m s^{-1}). For all characteristics of rough beer samples see Table 3.1. The broken line shows the intrinsic membrane resistance (R_m) of the membrane module used, while the bar errors for R_T were $\pm 9\%$.

3.1.2 Effect of membrane pore size

The intrinsic membrane resistance of the other ceramic tubular membrane modules with nominal pore size of (0.4 or 1.2) μm was (5.28 or 1.60 $\times 10^{11}$) m^{-1} , this resulting in a smaller or a higher permeation flux, respectively. However, when using rough beers with initial turbidity of (52.2 or 16.5) EBC unit (Table 3.2), the time histories of the overall membrane resistance R_T in total recycle CFMF tests, carried out as reported previously, were similar to that observed with the 0.8- μm pore membrane module (Fig. 3.4a,b). Even in these tests, the ratio (R_T^*/R_m) was by far greater than unity, its maximum and minimum values being about (38 and 101) or (54 and 268) for the 0.4- or 1.2- μm membrane module, respectively.



3.4a)



3.4b)

Figure 3.4 Time course of the overall membrane resistance (R_T) as resulting from a step by step increase in the transmembrane pressure difference (TMPD) and crossflow velocity (v_s) when dealing with two highly turbid beer A samples (see Table 3.1) and operating with ceramic tubular membrane modules with nominal pore size of (a) 0.4- and (b) 1.2- μm at 10 °C and different v_s values ($\circ$, 2 m s^{-1} ; $\triangle$, 4 m s^{-1} ; $\square$, 6 m s^{-1}). Any broken line refers to the intrinsic membrane resistance (R_m) of the membrane module used, while the bar errors for R_T were $\pm 9\%$.

Moreover, the minimum R_T^* values were associated to TMPD = 3 or 4 bar and $v_s = 6 \text{ m s}^{-1}$, this involving a limiting flux (J^*) of (72 ± 6) or $(84 \pm 8) \text{ L m}^{-2} \text{ h}^{-1}$ and of (35 ± 1) or $(49 \pm$

2) $\text{L m}^{-2} \text{ h}^{-1}$ for the greater or smaller pore size, respectively. Of course, the limiting flux increased with membrane porosity.

As shown in Table 3.2, as the pore size was increased from 0.4 to 1.2 μm , the membrane retention for density and colour reduced.

Table 3.2 Effect of membrane porosity on the main characteristics (haze, density, viscosity, and colour at 20 °C) of the rough beer A (RB) and resulting beer permeate (BP) collected from tank D2 (Fig. 2.1a) when the bench-top laboratory plant was operating under constant temperature (10 °C), transmembrane pressure difference (3.73 bar) and crossflow velocity (6 m s^{-1}).

Pore Size [μm]	Stream	Haze [EBC unit]	Density [kg m^{-3}]	Viscosity [mPa s]	Colour [EBC unit]
0.4	RB	52.2 ± 0.1	1013 ± 1	1.49 ± 0.05	30.7 ± 0.1
	BP	0.37 ± 0.04	1009 ± 1	1.40 ± 0.07	25.0 ± 0.1
0.8	RB	10.2 ± 0.1	1015 ± 1	1.56 ± 0.04	34.2 ± 0.1
	BP	0.52 ± 0.05	1013 ± 1	1.49 ± 0.02	32.1 ± 0.3
1.2	RB	16.5 ± 0.5	1012 ± 1	1.41 ± 0.01	26.2 ± 0.1
	BP	0.45 ± 0.03	1010 ± 1	1.32 ± 0.03	24.5 ± 0.3

Clearly, the 0.4- μm pore membrane consistently retained more carbohydrate molecules and melanoidins (i.e., the compounds formed in virtue of the Maillard reactions between simple sugars with aminoacids during barley malting and wort boiling) than the 1.2- μm pore size one. Indeed, the rate of removal should increase as the membrane becomes fouled. With the smaller pore size membrane, it is expected that some of the finer particles and macromolecules are entrapped within the fouling layer, and thus this layer is ultra-filtering such compounds. This occurrence may be better grasped by measuring the retention capacity of the membrane module by accounting for the so called *brewer's point* or *excess gravity*, this reducing from (4 to 2) as the pore size was increased from (0.4 to 1.2) μm .

A similar trend was noted with colour measurements, even if with the 0.4- μm membrane there was a greater loss in colour (about 18%) than that ($\sim 6\%$) achieved with the other greater porosity membranes (Table 3.2). On the contrary, the loss in viscosity varied from (4 to 6)%. Despite all beer permeates exhibited turbidity levels < 0.6 EBC unit, the overall removal of the haze-forming poliphenol-protein complexes was significantly higher with the lower porosity membrane (Table 3.2). Moreover, the smaller pore diameter membranes should also retain most of the proteinaceous components, this yielding filtered beer with low head retention values (Gan *et al*, 2001).

Thus, even if further work is needed to assess better the complex effect of membrane porosity on filtrated beer quality, and especially on its head retention value that was

unmeasured here, the 0.8- μm pore membrane appeared to be a good compromise between a high filtration capacity and a minimum retention on beer colour and density. Moreover, this pore size is expected to reduce the initial microbial density of rough beer by a greater factor than the 1.2- μm pore membrane (Filladeau *et al*, 2007).

3.1.3 Effect of beer turbidity

During rough beer A storage at 4 °C, it was possible to withdraw a series of samples, labelled as RB8 to RB14, with turbidity values in the range of (1.4 - 61.9) EBC unit, as shown in Table 3.1. Other two samples, labelled RB15 and RB16, were centrifuged.

All these samples underwent batch CFMF using the 0.8- μm pore membrane module under constant TMPD (~ 3.7 bar), v_s (6 m/s) and T (10 °C). As shown in Figure 3.5a, the time course of the volume collected (V) varied quite a lot depending on the initial turbidity level (H) of the rough beer tested. In each test, the estimated permeation flux (J_v) tended to the quasi steady-state flux (J^*) after about (0.5 – 1) h, the flux decay being more or less drastic depending on H (Fig. 3.5b). More specifically, J^* displayed a severe reduction from about (2,880 to 130) $\text{L m}^{-2} \text{h}^{-1}$ as turbidity increased from 0 (i.e., deionised water) to 3.7 EBC unit and tended to the ultimate value of $(63 \pm 6) \text{ L m}^{-2} \text{h}^{-1}$ for $H > 21$ EBC unit (Fig. 3.6).

Such a trend was empirically described with an average standard error of 19.5% by the following regression:

$$J^* = J_{v,w} \exp(-2.77 \pm 0.11) H^{-(0.275 \pm 0.043)} \quad (r^2 = 0.85) \quad \text{for } H > 0.1 \quad (3.2)$$

where $J_{v,w}$ is the permeation flux ($2879 \pm 63 \text{ L m}^{-2} \text{h}^{-1}$) observed with deionised water ($H=0$). The centrifugation step resulted in about 2.7- to 4.1-fold increase in J^* (168 ± 8 or $259 \pm 19 \text{ L m}^{-2} \text{h}^{-1}$) provided that the haze level of rough beer A had been reduced to (1.0 or 0.7) EBC unit (Fig. 3.6). These results clearly established the effectiveness of the centrifugation step used to pretreat lager beer undergoing both the commercial Alfa Laval and Pall CFMF processes (Buttrick, 2007). It is also worth noting that by prolonging the natural sedimentation of rough beer at 4 °C it was possible to withdraw quite a clarified sample

Fig. 3.5a)

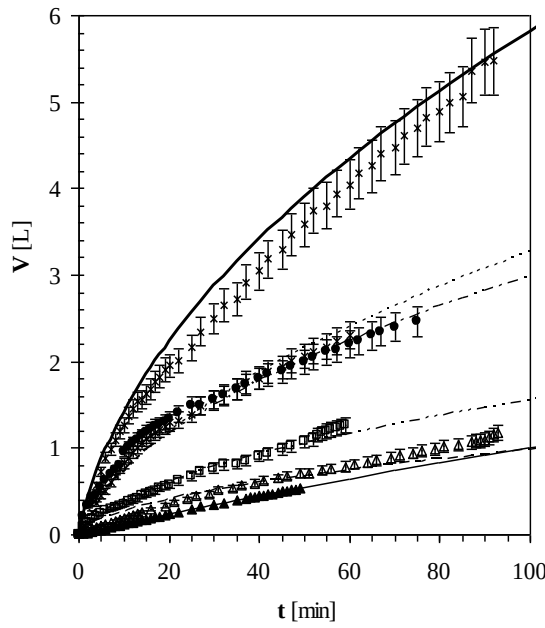


Fig. 3.5b)

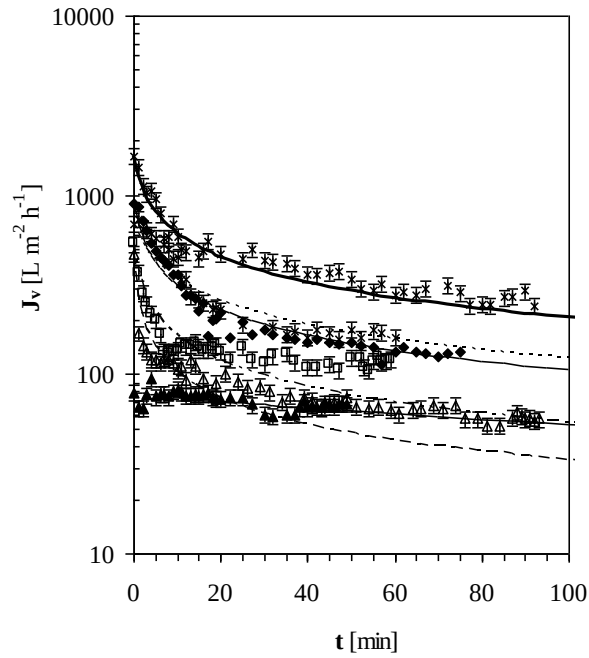


Figure 3.5 Time course of (a) the permeate volume collected (V) and (b) permeation flux (J_v) of a few differently turbid beer A samples (RB9: ●, — · — · —; RB10: □, — · — · —; RB12: △, — — —; RB13: ▲, — — —; RB15: ×, — — —; RB16: *, — — —) when operating with the 0.8- μm ceramic tubular membrane module under constant transmembrane pressure difference ($\text{TMPD} = 3.73 \text{ bar}$), crossflow velocity ($v_s = 6 \text{ m s}^{-1}$), and temperature ($T = 10 \text{ }^\circ\text{C}$). For all characteristics of rough beer samples see Table 3.1. The bar errors for J_w were $\pm 9\%$. The diverse lines representing the calculated V and J_v values were plotted using Eq. (1.19) or (1.20), respectively.

(RB8) at H of 1.40 ± 0.03 EBC unit, its CFMF processing yielding a quasi-state state permeation flux of $(130 \pm 8) \text{ L m}^{-2} \text{ h}^{-1}$ at 10°C , to some extent greater than that $(80 \text{ L m}^{-2} \text{ h}^{-1})$ claimed by the Norit process (Buttrick, 2007) at $0 - 2^\circ\text{C}$. By recovering the suspended matter with size larger than $0.5 \mu\text{m}$ (Buttrick, 2007), the centrifugation pretreatment is in all probability able to limit cake layer formation, thus enhancing the limiting permeation flux to values comparable with those $(250\text{-}500 \text{ L m}^{-2}\text{h}^{-1})$ achievable with powder filters (Buttrick, 2007; Fillaudeau *et al*, 2006).

3.1.4 Assessment of the prevailing membrane fouling mechanisms

To identify the controlling fouling mechanism during the CFMF of the aforementioned rough beer A samples, the instantaneous permeate volume-vs.-time data were firstly reconstructed by resorting to the mathematical models generally used to describe the 4 basic membrane fouling mechanisms, that is cake formation (CF), pore constriction (PC) or standard pore blocking; complete (CPB) or intermediate (IPB) pore blocking, as well as their combination, these being extracted from Bolton *et al* (2006) and listed in Table 1.10. Such a preliminary assessment was, however, unsuccessful since all the 4 basic fouling models allowed the experimental permeate volumes to be reconstructed with quite similar values of the residual variance (s_v^2) and mean squared percentage error (MSPE_v), these performance indexes being generally smaller than those characterizing the combined fouling models listed in Table 3.3. To discriminate more efficiently the 4 basic fouling models, it was decided to minimise simultaneously the time courses of V and J_v for any batch CFMF test under study.

Table 3.3 lists the empirical parameters of the aforementioned basic fouling models together with their corresponding residual variance (s_j^2) and mean squared percentage error (MSPE_j) for the two dependent variables examined. The best fitting of the experimental V and J_v data was generally obtained by resorting to the cake filtration model. This phenomenon was probably caused by the particle accumulation on the membrane surface that created a cake layer on it, this avoiding the finer aggregates to trap into the membrane pores.

By plotting k_{CF} and J_{v0} against H (Fig. 3.6), the following was pointed out.

As shown by the dash-dotted line in Fig. 3.6, J_{v0} exhibited the same double logarithmic trend previously observed for J^* and was empirically fitted by using the least squares method as:

$$J_{v0} = J_{v,w} \exp(-0.77 \pm 0.26) H^{-(0.474 \pm 0.103)} \quad (r^2 = 0.75) \quad \text{for } H > 0.1 \quad (3.3)$$

On the contrary, the cake filtration constant (k_{CF}) appeared to increase almost linearly with H in the range of (0 to 21.3) EBC unit, the coefficient of determination (r^2) of the broken line plotted in Fig. 3.5 being equal to 0.96. At H values in the range of 54-62 EBC unit, k_{CF} appeared to level off to $(200,474 \pm 18,226) \text{ s m}^{-2}$, this probably reflecting the fact that J^* and J_{v0} tended to smooth out at such turbidity levels.

According to the s_j^2 and $MSPE_j$ values shown in Table 3.3, the continuous lines in Fig. 3.5a or 3.5b, as calculated by using Eq. (1.19) or (1.20) together with the empirical parameter k_{CF} values

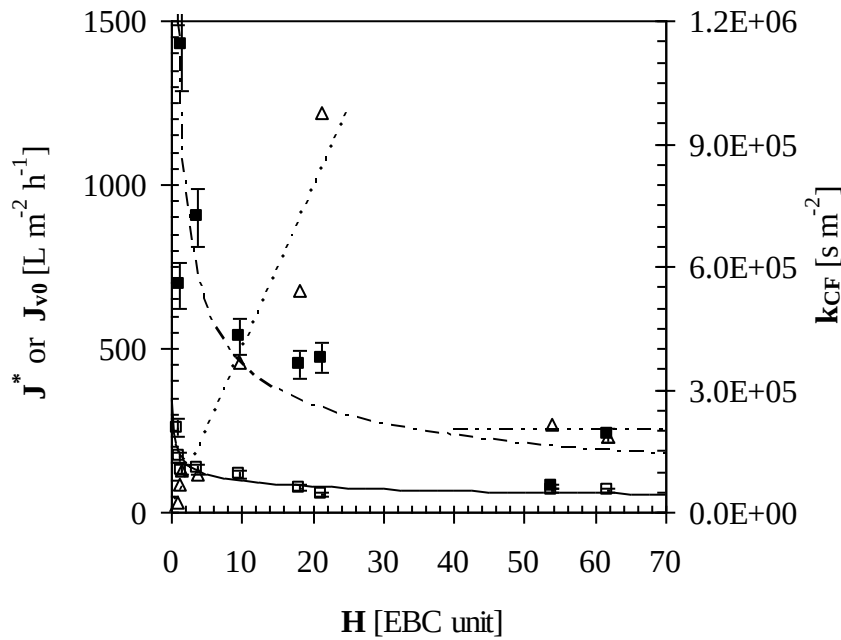


Fig. 3.6 Effect of the turbidity (H) of rough beer A samples (RB8-16) on the initial (J_{v0} : $\blacksquare$) and quasi steady-state (J^* : $\square$) permeation fluxes, and cake filtration constant (k_{CF} : $\triangle$) when operating with the $0.8\text{-}\mu\text{m}$ ceramic tubular membrane module under constant transmembrane pressure difference ($TMPD = 3.73 \text{ bar}$), crossflow velocity ($v_s = 6 \text{ m s}^{-1}$), and temperature ($T = 10 \text{ }^\circ\text{C}$). For all characteristics of rough beer samples see Table 3.1. The continuous or dash-dotted line was calculated using Eq. (3.2) or (3.3), while the broken or dash-double dotted line refers to the least squares regression line or average value of k_{CF} at the higher turbidity levels.

Table 3.3 Summary of the residual variance (s_j^2) and mean squared percentage error ($MSPE_j$) for the j -th dependent variable V or J_v , average mean squared percentage error ($MSPE_{ave}$), and empirical parameters (k_i) associated to the four basic pore blocking models (FM) described in Table 1.10 together with the number of data (N) available for any dependent variable taken into account and initial value of the experimental permeation flux (J_{v0}).

Rough beer Sample	J_{v0} [L m ⁻² h ⁻¹]	FM	N	S_V^2 [L ²]	$MSPE_V$ [%]	S_J^2 [L ² m ⁻⁴ h ⁻²]	$MSPE_J$ [%]	$MSPE_{ave}$ [%]	k_{CPB} [s ⁻¹]	k_{IPB} [m ⁻¹]	k_{PC} [m ⁻¹]	k_{CF} [s m ⁻²]
RB8	1429 ± 30	CPB	43	0.8.37	68.3	6.21 x10 ⁴	84.0	76.1	1.22x10 ⁻³	13.90	8.67	104628.4
		IPB		0.012	9.6	3980	32.6	21.1				
		PC		0.050	17.9	1.18 x10 ⁴	55.0	36.5				
		CF		0.0074	8.0	682	8.8	8.4				
RB9	900 ± 67	CPB	43	0.030	12.9	1.00 x10 ⁴	57.3	35.1	1.16 x10 ⁻³	9.53	6.55	94197.8
		IPB		0.140	8.5	1490	22.3	15.4				
		PC		0.128	9.1	4070	37.0	23.1				
		CF		0.163	15.2	2560	14.7	15.0				
RB10	536 ± 52	CPB	41	0.055	29.4	9660	73.7	51.6	1.86 x10 ⁻³	24.51	16.93	363641.6
		IPB		0.0127	15.8	3440	44.2	30.0				
		PC		0.0261	22.3	6020	58.0	40.1				
		CF		0.00233	7.4	1040	25.2	16.3				
RB11	452 ± 40	CPB	41	0.0253	33.4	5460	68.8	51.1	1.86 x10 ⁻³	29.65	20.22	542373.1
		IPB		0.006	19.6	1910	37.1	28.3				
		PC		0.0121	26.6	3340	51.4	39.0				
		CF		0.0011	8.3	529	18.0	13.2				
RB12	472 ± 43	CPB	50	0.210	67.4	5740	91.6	79.5	4.03 x10 ⁻³	49.3	35.5	974679.4
		IPB		0.0503	49.8	2610	61.1	55.5				
		PC		0.112	59.3	4220	79.7	69.5				
		CF		0.0099	32.2	743	30.6	31.4				
RB13	80 ± 10	CPB	40	1.74x10 ⁻⁵	1.8	36.1	8.6	5.2	8.78 x10 ⁻⁵	4.3	4.2	213361.7
		IPB		1.47 x10 ⁻⁵	1.8	35.7	8.5	5.1				
		PC		1.58 x10 ⁻⁵	1.8	35.9	8.5	5.2				
		CF		1.33 x10 ⁻⁵	1.7	35.5	8.4	5.1				
RB14	235 ± 21	CPB	43	0.0011	9.0	147	9.9	9.4	4.03 x10 ⁻⁴	8.7	7.3	187586.7
		IPB		3.01 x10 ⁻⁴	7.7	124	7.4	7.5				
		PC		5.53 x10 ⁻⁴	8.1	121	8.2	8.2				
		CF		2.32 x10 ⁻⁴	7.4	182	7.8	7.6				
RB15	693 ± 59	CPB	37	0.0154	12.2	3980	35.2	23.7	7.57 x10 ⁻⁴	7.2	5.4	70109.5
		IPB		0.0032	8.0	2280	16.4	12.2				
		PC		0.0055	9.2	2340	23.3	16.3				
		CF		0.0073	9.7	4350	15.9	12.8				
RB16	1654 ± 137	CPB	50	0.697	42.6	5.76 x10 ⁴	71.8	57.2	1.22 x10 ⁻³	4.9	3.4	24484.3
		IPB		0.162	34.1	1.39 x10 ⁴	33.8	34.0				
		PC		0.298	38.9	3.05 x10 ⁴	50.7	44.8				
		CF		0.063	25.8	3.99 x10 ³	13.6	19.7				

3.1.5 Validation tests under CO₂ backflushing.

Backflushing is regarded as an effective technique to reduce the fouling caused by pore-entrance blockage and in-depth particle plugging, its effectiveness depending on CO₂ pressure, pulse duration, duration between end of the pulse and opening of the permeate valve, and cycle frequency (Gan, 2001). After several trials, it was possible to regard as optimal the automatic setting of the backflush pressure difference between the permeate and retentate sides at +3 bar for not less than 2 min when the permeation flux had dropped below the quasi-steady state value. The effectiveness of the aforementioned operating conditions (TMPD = 3.73 bar, $v_s = 6 \text{ m s}^{-1}$, $T = 10 \text{ }^\circ\text{C}$) and backflush programme was further tested by filtering a few rough beer A samples with the 0.8- μm membrane module. One of these samples resulted from siphoning accurately decanted rough beer kept at 4 $^\circ\text{C}$ (RB8), while the other two ones were preliminary centrifuged (RB15 and RB16).

Fig. 3.7 shows the corresponding time course of J_v . Congruently with the cake filtration fouling mechanism, CO₂ backwashing resulted to be quite effective not only to restore the quasi-steady state (J^*) permeation flux, but also to maximise the average ($J_{v,av}$) permeation flux (Table 3.4)

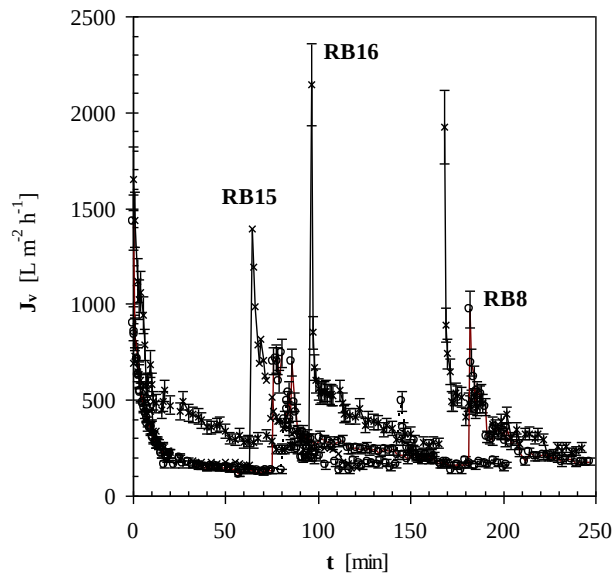


Figure 3.7 Time course of the permeation flux (J_v) of a few differently turbid beer A samples (RB8: O; RB15: X; RB16: *) when operating with the 0.8- μm ceramic tubular membrane module under constant transmembrane pressure difference (TMPD = 3.73 bar), crossflow velocity ($v_s = 6 \text{ m s}^{-1}$), temperature ($T = 10 \text{ }^\circ\text{C}$), and periodic CO₂ back-flushing. For all characteristics of rough beer samples see Table 3.1. The bar errors for J_w were $\pm 9\%$.

Table 3.4 *Effect of periodic CO₂ back-flushing on the quasi-steady state (J^*) and average ($J_{v,av}$) permeation fluxes when using rough beer A samples at low turbidity values (H). For all characteristics of rough beer samples see Table 3.1*

Rough Beer	Rough Beer Haze (20 °C) [EBC unit]	Permeate Haze (20 °C) [EBC unit]	Permeate Haze (0 °C) [EBC unit]	J^* [L m⁻² h⁻¹]	$J_{v,av}$ [L m⁻² h⁻¹]
RB8	1.40	0.50 ± 0.01	2.70 ± 0.01	159 ± 26	255 ± 35
RB15	1.00	0.40 ± 0.01	2.26 ± 0.09	164 ± 62	300 ± 60
RB16	0.72	0.44 ± 0.03	1.64 ± 0.02	257 ± 21	385 ± 33

Thus, the combined centrifugation and periodic CO₂ backflush programme resulted in 3 to 5-fold increase in the average permeation flux at 10 °C with respect to that (80 to 100 L m⁻² h⁻¹) at 0 - 2 °C claimed by the three commercial CFMF processes mentioned before (Buttrick, 2007).

3.2 Validation of the Optimal CFMF Operating Conditions using Rough Beer B and assessment of the beer stabilization using

Table 3.5 summarizes the average characteristics of the rough beers B used here (i.e., RB1 - 4), their turbidity values varying from 2.4 in RB1 to 52.4 EBC unit in RB2, as well as their corresponding permeates (i.e., P1 - 4). Owing to the natural sedimentation of suspended matter in the storage tank at 4 °C, some variability from batch to batch and within the same beer batch was noticeable. All beer permeates, except those obtained by enzymatic treatment, exhibited a turbidity slightly higher than the limiting turbidity level (< 0.6 EBC unit) suggested by the European Brewery Convention standards, even if all permeate samples appeared to be brilliantly clear. This might be explained by accounting for the fact that the aforementioned EBC threshold value results from turbidity measurements at 560 nm, while the data listed in Table 3.5 were taken at 470 nm. In fact, it is well known that long wavelengths are always less intensely scattered than short ones (Chapon, 1993). Finally, the density, colour, and alcohol degree of samples RB2 and P2 were typical of the Lite or Standard American Lager (BJCP Style Guidelines, 2008).

Table 3.5: Main characteristics (pH; density, ρ ; viscosity, η ; turbidity, H ; colour, C ; β -glucans, β -G; real extract, RE; original extract, OE; alcohol, A) of the rough beer samples, as such (RB), precentrifuged (RBC) or enzymatically treated and centrifuged (RBEC), together with the corresponding micro-filtered (P, PC, PEC) samples used in this work.

Sample	pH	ρ [kg dm ⁻³]	η [mPa s]	H [EBC]	C [EBC]	β -G [g m ⁻³]	RE [°Plato]	OE [°Plato]	A [% v/v]
RB1	4.39±0.05	1.0048±0.0004	1.41±0.01	2.4±0.1	6.9±0.1	140±4	3.5±0.2	14.4±0.1	5.1±0.1
P1	4.39±0.05	1.0048±0.0004	1.40±0.01	0.8±0.05	6.3±0.1	135±12	2.9±0.1	12.6±0.1	5.1±0.2
RB2	4.39±0.05	1.0047±0.0004	1.41±0.01	52.4±0.1	6.9±0.1	140±4	3.5±0.2	14.4±0.1	5.1±0.1
P2	4.39±0.05	1.0044±0.0005	1.39±0.01	1.1±0.05	6.3±0.1	135±12	2.9±0.1	12.6±0.1	5.1±0.2
RB3	4.39±0.05	1.0048±0.0004	1.39±0.01	18.0±0.1	6.9±0.1	140±4	3.5±0.2	14.4±0.1	5.1±0.1
P3	4.39±0.05	1.0043±0.0005	1.37±0.01	1.2±0.01	5.5±0.1	135±12	2.9±0.1	12.6±0.1	5.1±0.2
RB4	4.21±0.05	1.0050±0.0004	1.36±0.01	7.1±0.1	6.9±0.1	140±4	3.5±0.2	14.4±0.1	5.1±0.1
P4	4.21±0.05	1.0045±0.0005	1.33±0.01	0.9±0.01	6.3±0.1	135±12	2.9±0.1	12.6±0.1	5.1±0.2
RBC5	4.39±0.05	1.0043±0.0004	1.36±0.01	1.5±0.1	5.8±0.1	140±4	3.5±0.2	10.3±0.1	3.9±0.1
PC5	4.39±0.05	1.0040±0.0005	1.36±0.01	0.8±0.01	5.1±0.1	135±12	1.7±0.1	9.3±0.1	3.9±0.2
RBEC6	4.39±0.05	1.0052±0.0004	1.21±0.01	0.9±0.1	5.1±0.1	1.3±0.6	3.5±0.2	10.3±0.1	3.9±0.1
PEC6	4.39±0.05	1.0041±0.0003	1.21±0.01	0.4±0.01	4.8±0.1	0.8±0.5	1.7±0.1	9.3±0.1	3.9±0.2

In CFMF under the aforementioned constant operating parameters, that is TMPD =3.74 bar, v_s = 6 m/s and T =10 °C for up to 4 h, the permeation flux, using rough beer B, tended to a quasi steady-state value (J^*) after about 1 h, independently of the turbidity (H) of the rough beer tested in this work (Figure 3.8A). Also with rough beer B, it can be noted a drastic reduction in $J_{v,ss}$ or in the average permeation flux ($J_{v,av}$), as turbidity increased from 0 to (2 – 3) EBC unit with an asymptotical value of (91 ± 8) or (117 ± 14) L m⁻² h⁻¹ for $H > 7$ EBC unit. Both trends were empirically described with an average standard error of 4.2 or 6.2% by the following regressions:

$$J^* = J_{v,w} [1 + 0.97 (e^{-1.90 H} - 1)] \quad (3.4)$$

$$J_{v,av} = J_{v,w} [1 + 0.96 (e^{-1.59 H} - 1)] \quad (3.5)$$

where $J_{v,w}$ is the permeation flux (3,061±67 L³ m⁻² h⁻¹) with deionised water (H=0).

Figure 3.9 compares the time course of the permeation flux (J_v) at 10 °C under constant TMPD (3.76 bar), v_s (6 m/s), and periodic CO₂ backflashing when using a rough beer sample B as such or after preliminary treatments aimed at removing yeast cells and larger aggregates by centrifuging or at hydrolyzing firstly the gel forming polysaccharides and secondly get rid of the suspended solids by centrifugation.

It can be noted that the centrifugation step resulted in about (50 or 75)% increase in J^* (137 ± 13 L m⁻² h⁻¹) or $J_{v,av}$ (205 L m⁻² h⁻¹), thanks to the recovery of suspended matter

with size larger than 0.5 μm . In all probability, such a pretreatment was unable to counteract the gel layer formation, as well as the entrapping of the smaller aggregates within the membrane porous structure.

The preliminary use of the commercial enzyme preparation appeared to be capable of degrading almost all the β -glucans (Table 3.5), thus lowering their tendency to aggregate and making the smaller molecular mass fractions easier to permeate across the membrane undisturbed (Stewart *et al.* 1998). Once the enzymatically-treated beer had been centrifuged, the resulting J^* or $J_{v,av}$ values (294 ± 30) or $336 \text{ L m}^{-2} \text{ h}^{-1}$) was more than two or one and a half fold higher than that achieved when the rough beer was centrifuged and then microfiltered.

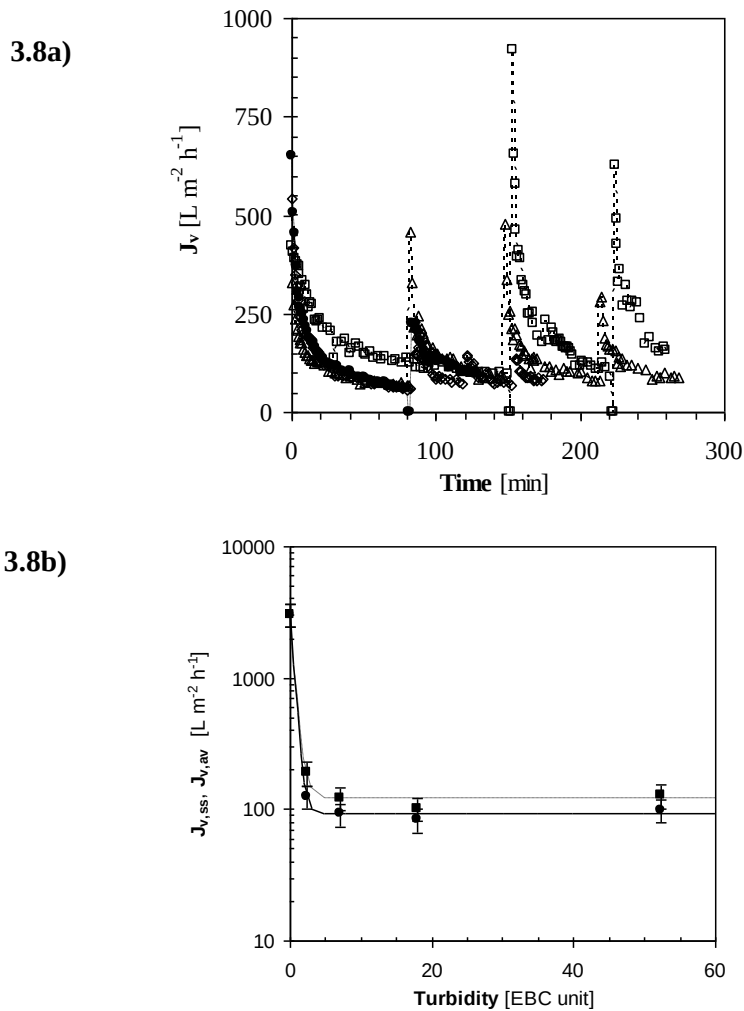


Figure 3.8: Effect of the turbidity (H) of a few rough beer B samples ($\square$, 2.4; $\triangle$, 7.1; $\diamond$, 18; $\bullet$, 52.4 EBC unit) on **a)** the time course of the permeation flux (J_v) at 10°C under constant TMPD (3.96 bar), v_s (6 m/s), and periodic CO_2 backflashing and **b)** quasi steady-state (J^*) and average ($J_{v,av}$) permeation fluxes. The continuous and broken lines were calculated using Eq.s (3.4) and (3.5), respectively.

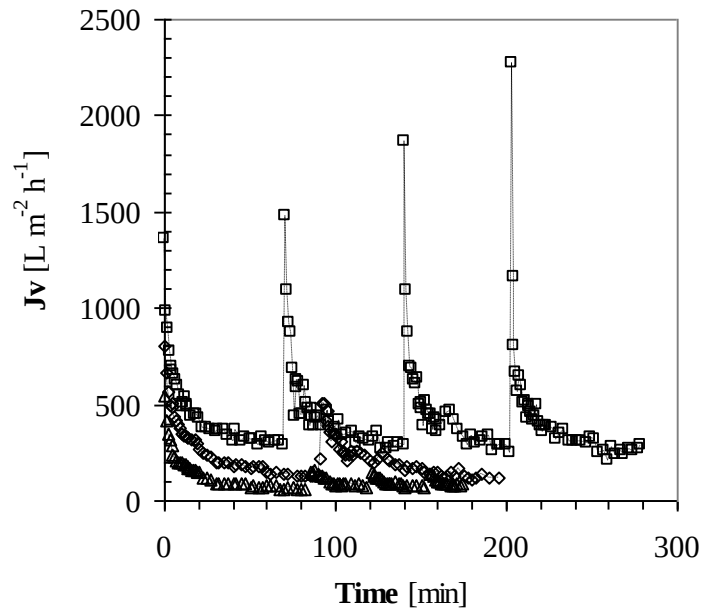
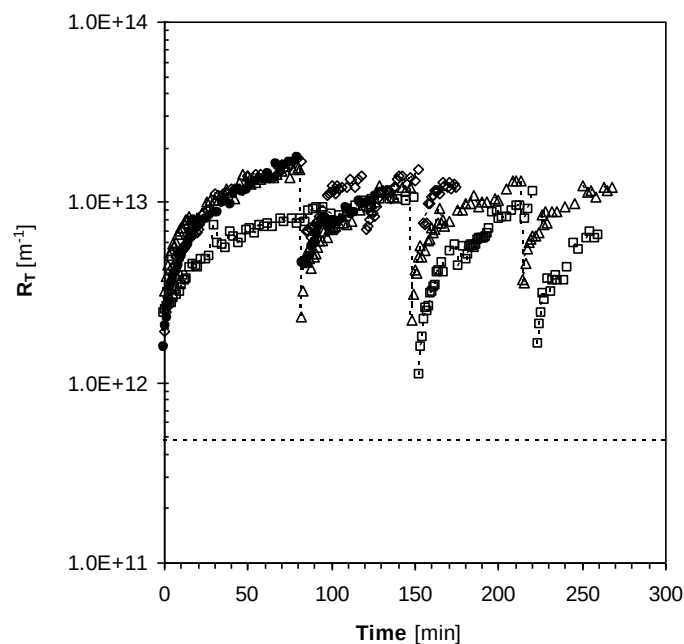


Figure 3.9: Time course of the permeation flux (J_v) of rough beer B samples as such ($\triangle$, 18 EBC unit), precentrifuged ($\diamond$, 1.5 EBC unit) or after enzymatical and centrifugal pretreatments ($\square$, 0.9 EBC unit) under constant TMPD (3.96 bar), v_s (6 m/s), temperature (10 °C), and periodic CO₂ back-flashing.

Also with beer B, to mark better the shear effect of v_s , the instantaneous permeation flux was used to estimate the corresponding overall membrane resistance (R_T) via Eq. (3.1), as show in Fig 3.10a,b

3.10a)



3.10b)

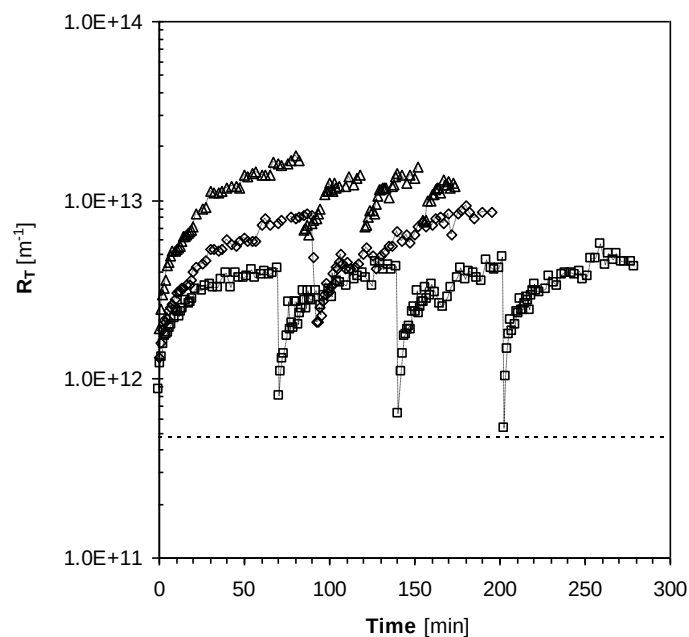


Figure 3.10: Time course of the overall hydraulic resistance (R_T) when operating at 10 °C under constant TMPD (3.74 bar), vs (6 m/s), and periodic CO₂ back-flushing, as reported in the Materials and Method section, and using: **A)** rough beer B samples of different turbidity ($\square$, 2.4; $\triangle$, 7.1; $\diamond$, 18.0; $\bullet$, 52.4 EBC unit); **B)** rough beer B samples as such ($\diamond$, 18.0 EBC unit), pre-centrifuged ($\diamond$, 1.5 EBC unit) or enzymatically pre-treated and centrifuged ($\square$, 0.9 EBC unit). Both broken lines refer to the clean membrane hydraulic resistance (R_m).

Once the rough beer B had entered the membrane module, $R_T = [(2.3 \pm 0.7) \times 10^{12} \text{ m}^{-1}]$ was found to be definitively greater than the resistance of the clean membrane $R_m = [(4.7 \pm 0.1) \times 10^{11} \text{ m}^{-1}]$. To counteract membrane fouling, the electro-valve connecting

the permeate tank to the 10-bar CO₂ cylinder (Fig. 2.1) was automatically opened to boost suddenly the pressure in the permeate side of the tubular module to +3 bar, with respect to that in the retentate side for as long as 2 min. In this way, the swift flush of CO₂ was capable of lowering R_T to $(1.1\text{--}2.3) \times 10^{12} \text{ m}^{-1}$, especially when a rough beer B of low turbidity ($H = 2.4$ EBC unit) was undergoing filtration.

Regardless of the initial turbidity of rough beer B, the time course of R_T exhibited a downward concavity typical of external fouling (i.e., cake filtration and complete pore blocking) and this explained the efficacy of CO₂ backwashing to restore the original permeation flux.

Figure 3.9b shows the effect of some rough beer B pre-treatments on R_T . Also with the rough beer B, the pre-centrifugation step not only reduced the growth rate of R_T , but also approximately halved its value from $1.67 \times 10^{13} \text{ m}^{-1}$, typical of a sample of rough beer with $H = 18$ EBC unit, to $0.88 \times 10^{13} \text{ m}^{-1}$. The enzymatic pre-treatment followed by centrifugation further reduced R_T to $\sim 0.41 \times 10^{13} \text{ m}^{-1}$. Moreover, CO₂ backflushing was able to lower R_T such as low as $(5.2\text{--}8.1) \times 10^{11} \text{ m}^{-1}$, these values being near to the resistance of clean membrane [$R_m = (4.7 \pm 0.1) \times 10^{11} \text{ m}^{-1}$].

3.2.1 Further Validation of CFMF using pretreated rough beer B

Table 3.6 lists the main characteristics (i.e., ρ , η , H at 20 or 0 °C) of a new batch of rough lager beers (B) tested and resulting permeates together with the mean and standard deviation of the quasi-steady state (J^*) permeation flux observed in total recycle tests. As an example, Fig. 3.11 compares the time course of the experimental permeate volume (V) and permeation flux (J_v) for the three cases under study. All data collected were reconstructed by using the cake formation (CF) model, this assuming the formation of a uniform, permeable cake layer of particles over the entire membrane surface. Table 3.7 lists the best fitting values of the cake filtration constant (k_{CF}), together with the residual variance (s_j^2) and mean squared percentage error ($MSPE_j$) for any dependent variable y_j , as estimated by means of Eq. (2.1 and 2.2), respectively. The continuous lines, plotted in Figs 3.11a and 3.11b, were calculated using Eq.s (1.19 and 1.20), respectively. It can be noted a remarkable agreement among the calculated and experimental time courses of V and J_v .

Table 3.6. Mean and standard deviation of the main characteristics (pH; density, ρ ; viscosity, η ; turbidity, H , at 20 and/or 0° C; color, C) of rough beer samples, precentrifuged (RBC) or submitted to enzymatic and centrifugal pretreatments (RBEC) and of corresponding (PC, PEC) micro-filtered samples, together with the quasi-steady state (J_v^*) and average ($J_{v,av}$) permeation fluxes observed in some total recycle tests carried out at $T \approx 10$ °C, $TMPD = 3.74$ bar, $v_S = 6$ m s⁻¹, and periodic CO₂ backflushing.

Sample	pH	ρ [kg/L]	η [mPa s]	$H_{20}^{\circ C}$ [EBC]	C [EBC]	$H_0^{\circ C}$ [EBC]	J_v^* [L m ⁻² h ⁻¹]	$J_{v,av}$ [L m ⁻² h ⁻¹]
RBC3	4.12	1.007±0.001	1.379±0.006	1.09±0.03	12.6±0.1	12.1±0.2		
PC3	4.12	1.006±0.001	1.370±0.002	0.52±0.00	12.1±0.1	5.1±0.1	114 ± 9	217
RBEC1	4.12	1.007±0.001	1.378±0.002	1.07±0.03	12.6±0.1	1.6±0.1*		
PEC1	4.12	1.006±0.001	1.370±0.002	0.47±0.00	12.3±0.1	0.7±0.1*	189 ± 5	357
RBEC2	4.15	1.007±0.001	1.422±0.006	1.09±0.03	12.6±0.1	5.0±0.2		
PEC2	4.15	1.006±0.001	1.415±0.002	0.64±0.00	12.1±0.1	1.7±0.2	138 ± 9	322
RBEC4	4.11	1.007±0.001	1.370±0.002	1.09±0.03	12.5±0.2	15.4±0.2		
PEC4	4.11	1.006±0.001	1.354±0.002	0.64±0.00	12.3±0.1	4.0±0.2	203 ± 42	304
RBEC5	4.09	1.007±0.001	1.378±0.001	1.09±0.03	12.6±0.1	15.3±0.1		
PEC5	4.09	1.006±0.001	1.370±0.002	0.46±0.00	12.3±0.1	4.0±0.2	217 ± 10	351
RBECs1	4.10	1.006±0.001	1.374±0.002	0.88±0.01	12.4±0.1	0.90±0.02		
PECS1	4.10	1.006±0.001	1.374±0.002	0.36±0.03	12.1±0.1	0.50±0.04	163±12	228
RBECs2	4.09	1.006±0.001	1.374±0.001	0.88±0.02	12.4±0.1	0.90±0.02		
PECS2	4.10	1.006±0.001	1.374±0.002	0.44±0.06	12.1±0.1	0.53±0.02	162±18	228

Table 3.7 Summary of lager beer CFMF data fitting using the cake filtration model: initial value of the experimental permeation flux (J_{v0}), number of data available (N), cake filtration constant (k_{CF}), residual variance (s_f^2) and mean squared percentage error ($MSPE_j$) for j expressing the dependent variable V or J_v , and average mean squared percentage error ($MSPE_{av}$). For all characteristics of rough beer samples see Table 3.

Rough Beer Sample	J_{v0} [L m ⁻² h ⁻¹]	N [-]	k_{CF} [s m ⁻²]	s_v^2 [L ²]	$MSPE_v$ [%]	s_J^2 [L ² m ⁻⁴ h ⁻²]	$MSPE_J$ [%]	$MSPE_{av}$ [%]
RBC3	2042	34	1.63 x10 ⁵	1.21 x10 ⁻²	12.7	1143	12.6	12.7
RBEC1	2816	31	6.40 x10 ⁴	2.06 x10 ⁻²	12.1	4383	17.9	15.0
RBEC2	3137	22	9.00 x10 ⁴	7.23 x10 ⁻²	23.9	59117	36.5	30.2
RBEC4	872	41	8.18 x10 ⁴	2.69 x10 ⁻³	8.9	2779	11.5	10.2
RBEC5	2830	36	4.39 x10 ⁴	1.59 x10 ⁻³	4.2	1240	7.9	6.1
RBECs1	2481	35	6.52 x 10 ⁴	6.70 x 10 ⁻³	8.3	7322	8.3	8.3
RBECs2	1893	35	5.67 x 10 ⁴	3.46 x 10 ⁻²	17.2	4533	25.9	21.5

Fig. 3.11a)

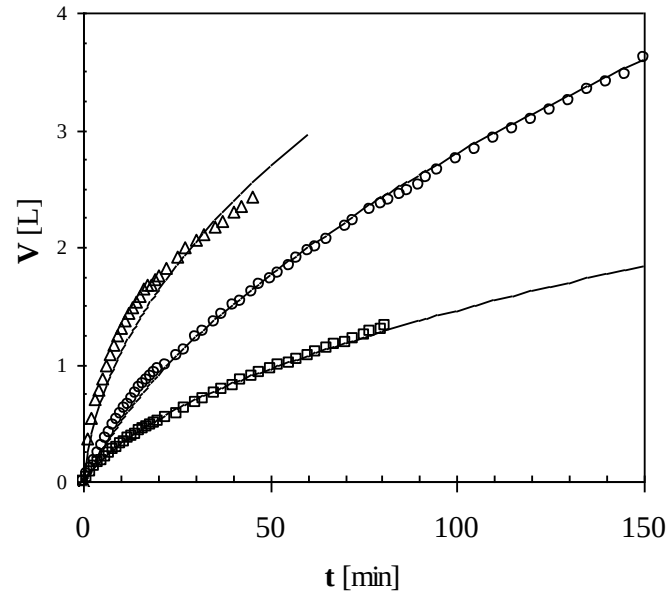


Fig. 3.11b)

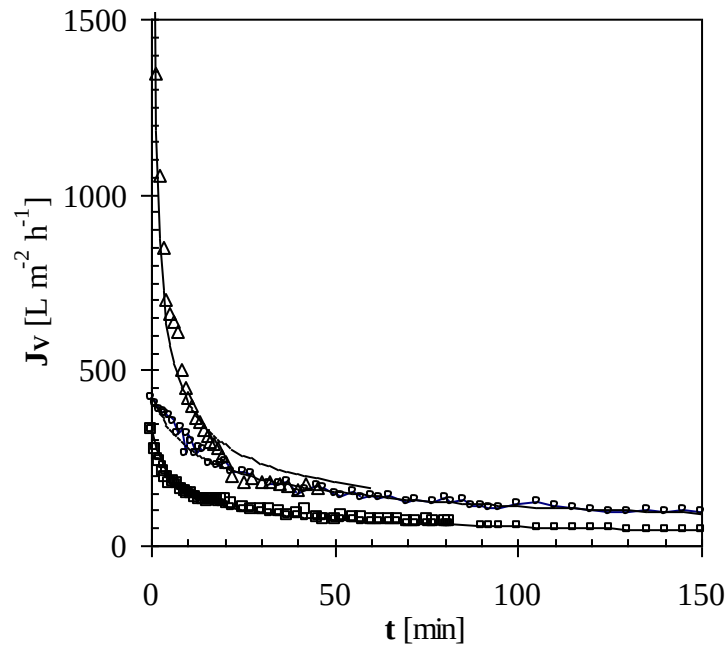


Figure 3.11 Time course of (a) the permeate volume collected (V) and (b) permeation flux (J_v) of a few rough beer samples, as such (RB3: $\square$), pre-centrifuged (RBC1: $\circ$) or pre-centrifuged and enzymatically treated (RBEC1: $\triangle$) under the CFMF conditions given in the text. For all characteristics of rough beer samples see Table 3.6. The continuous lines were plotted using Eq.s (1.19) and (1.20) with the parameters listed in Table 3.7. The average coefficient of variation for V and J_v was (5 and 9)%, respectively.

Fig. 3.12 shows the effect of the rough lager beer turbidity at 20 °C on the experimental initial permeation flux (J_{v0}) and estimated cake filtration constant (k_{CF}).

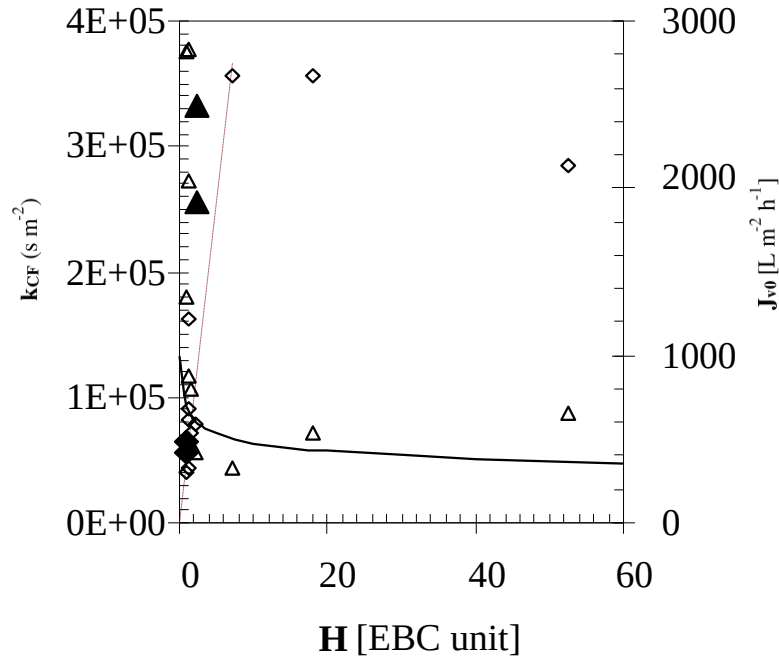


Figure 3.12 Effect of the turbidity (H) of rough beer samples on the initial permeation flux (J_{v0} : $\triangle$, $\blacktriangle$), and cake filtration constant (k_{CF} : $\diamond$, $\blacklozenge$) under the CFMF conditions given in the text. All characteristics of rough beer samples are reported in Table 3.6. In particular, open symbols refer to rough beer B samples, as such or C- and EC-pretreated, while the closed ones to EC-pretreated and stabilized beer B samples (RBECS1-2). The continuous and broken lines refer to the least squares regression lines.

As shown by the continuous line in Fig. 3.12, J_{v0} tended to reduce exponentially with H , while k_{CF} appeared to increase almost linearly with H in the range of 0 to 7 EBC unit, the coefficient of determination (r^2) of the broken line plotted in Fig. 3.12 being equal to 0.92. For H increasing from 7 to 54 EBC unit, k_{CF} ranged from $(3.6 \text{ to } 2.9) \times 10^5 \text{ s m}^{-2}$, this probably reflecting the fact that J_{v0} tended to smooth out at such high turbidities. Whatever the rough lager beer B sample under testing (Table 3.6), in about half an hour J_v tended to an asymptotic value, generally called *limiting* or *quasi-steady state permeation flux* (J^*), this increasing from (81 ± 8) to (117 ± 12) or $(205 \pm 51) \text{ L m}^{-2} \text{ h}^{-1}$ in the case of beer samples, as such or differently pretreated by centrifuging (C), or enzymatic hydrolysis and centrifuging (EC), respectively. Fig. 3.13 compares the typical time course of the experimental permeation flux (J_v) and overall membrane resistance (R_T), as estimated via Eq. (3.1), when using the aforementioned rough beer B samples and the CO_2 backwashing program.

Fig. 3.13a)

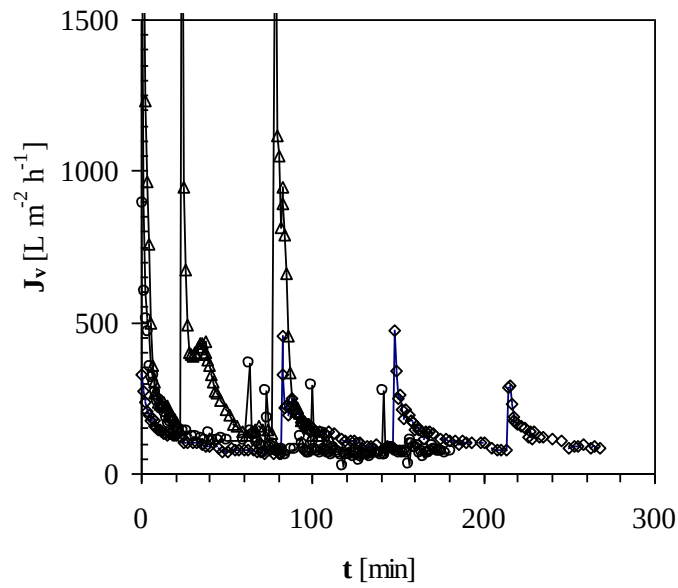


Fig. 3.13b)

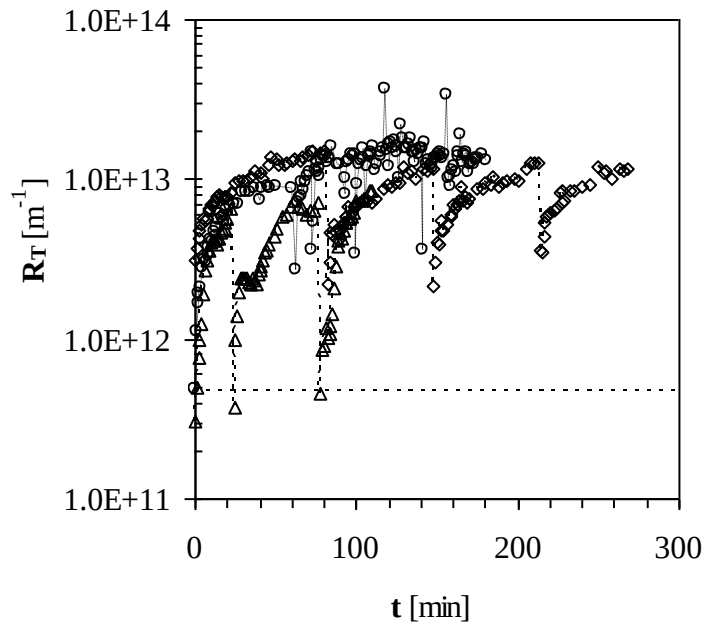


Figure 3.13 Time course of the (a) permeation flux (J_v) and (b) overall hydraulic resistance (R_T) of rough beer samples, as such (RB3: $\diamond$), precentrifuged (RBC3: $\circ$) or submitted to enzymatic and centrifugal pretreatments (RBEC2: $\triangle$) under the CFMF conditions given in the text and periodic CO_2 back-flushing.. For all characteristics of rough beer samples see Table 3.6. The broken line in Fig. 3.13b refers to the clean membrane hydraulic resistance (R_m). The average coefficient of variation for J_v and R_T was about 9%.

In the case of the cake filtration fouling mechanism, CO₂ backwashing should in principle be successful at restoring the initial permeation flux (Fig. 3.13a) and thus at maximizing the average ($J_{v,av}$) permeation flux (Table 3.6). In fact, owing to the formation of the cake layer, the overall membrane resistance (R_T) was much greater than the clean membrane resistance [$R_m = (4.7 \pm 0.1) \times 10^{11} \text{ m}^{-1}$] (Fig. 3.12b). To counteract membrane fouling, a periodic swift flush of CO₂ lowered R_T to $(2.7 - 3.6) \times 10^{12} \text{ m}^{-1}$, especially when rough beer B had been pre-centrifuged. Thus, owing to the recovery of suspended matter with size $> 0.5 \mu\text{m}$, the average value of $J_{v,av}$ for the C-pretreated rough beer amounted to $(199 \pm 17) \text{ L m}^{-2} \text{ h}^{-1}$ (Table 3.6), about two times higher than that achieved with the rough beer as such ($112 \pm 13 \text{ L m}^{-2} \text{ h}^{-1}$). Thanks to the commercial enzyme preparation, the β -glucan content of rough beer was reduced from (127 ± 4) to $(2 \pm 1) \text{ mg L}^{-1}$, this enhancing the average $J_{v,av}$ for EC-pretreated rough beer to $(332 \pm 22) \text{ L m}^{-2} \text{ h}^{-1}$ (Table 3.6).

3.2.2 Cold stabilization of beer

Kinetics of total polyphenol removal using regenerable PVPP

Despite most of the beer permeates collected during the aforementioned trials exhibited hazes at 20 °C ranging from (0.46 to 0.64) EBC unit, they still retained some soluble haze precursors responsible for post-filtration hazes. In fact, their chill haze at 0 °C increased from (0.7 to 4.0) EBC unit (Table 3.6). Thus, similarly to the great majority of DE-filtered beers, the membrane-filtered ones need to be further stabilized using polyvinylpyrrolidone (PVPP), alone or combined with selected carrageenan or silica xerogel (Rehmanji *et al*, 2005), or agarose beads (Taylor *et al*, 2006). Obviously, this post-filtration treatment would impair the sterility of beer permeates; while a pre-filtration stabilization treatment (maturation) of rough lager beer with non-regenerable PVPP would be expensive. So a good implementation of cold stabilization treatment for CFMF of beer, represents an essential requisite that is often underestimated.

Despite the measurement of all polyphenol species present in rough beer (TP) using the EBC method does not measure specifically the polyphenols involved in the beer

colloidal stability (Mélotte, 2008), the batch test depicted in Fig. 3.14 allowed the time course of residual TP content to be assessed at two different operating temperatures. From these data, it was possible to point out the fraction of polyphenolic compounds precipitated by PVPP, these generally including the low and medium molecular polyphenols, the polymers of catechin and anthocyanogens and affecting in some way the beer colloidal and taste stability (Mélotte, 2008).

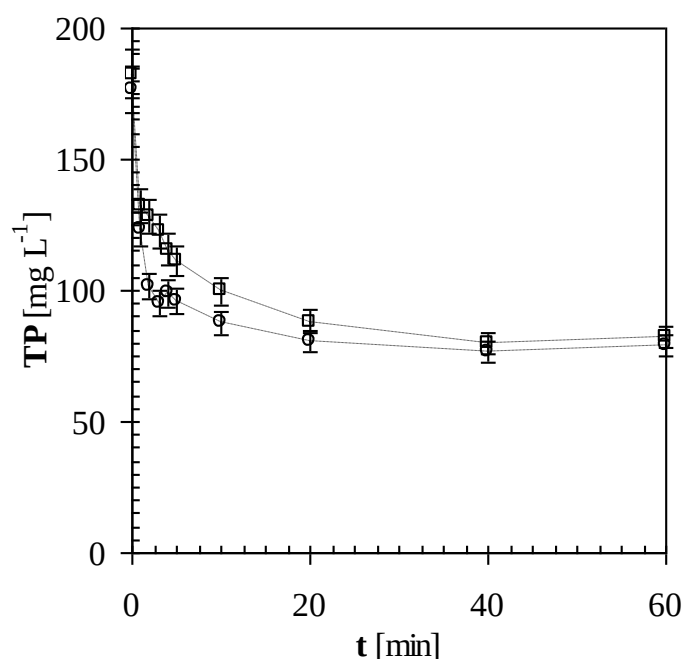


Figure 3.14 Time course of total phenol content (TP) in EC-pretreated rough beer B seeded with 0.5 g L⁻¹ PVPP at 0 (□) or 20 (○) °C.

Despite the rate of TP removal appeared to reduce with temperature, the percentage TP removal coefficient tended to almost the same equilibrium value of (45.0 ± 0.3)% at the confidence level of 95%. Since the aforementioned final TP content was approximately reached within 15-20 min, such preliminary tests suggested to combine the CFMF and stabilization processes, as reported below.

Simultaneous stabilization and crossflow microfiltration of EC-pretreated beer

To avoid supplementary filtering of PVPP stabilized beer, the EC-pretreated rough beer B was enriched with regenerable PVPP granules before being submitted to total recycle

CFMF tests. According to Meier (1993), the presence of granular or fibrous additives should prevent clogging of membrane pores and keep unaltered the above high $J_{v,av}$ values. Table 3.9 shows the time course of the TP level in permeated beer. In this case, the percentage TP removal leveled off at the $(70.0 \pm 0.1)\%$ of the initial TP concentration.

Table 3.8 Simultaneous stabilization and CFMF of EC-pretreated rough beer B: effect of contact time (t) on the mean and standard deviation of the polyphenol content (TP), turbidity (H) at (20 and 0) °C, and alcohol chill haze (ACH) at -5 °C in the beer permeate collected during total recycle CFMF tests in presence of 0.5 g L^{-1} of PVPP.

t [min]	TP [mg L ⁻¹]	$H_{20\text{ °C}}$ [EBC unit]	$H_{0\text{ °C}}$ [EBC unit]	ACH [EBC unit]
0	224.3 ± 2.9	0.46 ± 0.01	1.57 ± 0.01	28.46 ± 0.06
5	182.9 ± 3.5	0.46 ± 0.01	0.67 ± 0.03	9.40 ± 0.12
10	176.7 ± 2.9	0.46 ± 0.01	0.72 ± 0.01	5.46 ± 0.02
15	166.9 ± 0.6	0.47 ± 0.01	0.64 ± 0.01	4.43 ± 0.02
30	156.6 ± 2.3	0.46 ± 0.01	0.60 ± 0.01	3.70 ± 0.03
60	157.4 ± 2.3	0.46 ± 0.01	0.60 ± 0.01	2.23 ± 0.01

The haze at 20 °C of the resulting permeate was practically constant (0.463 ± 0.005 EBC unit), while the chill haze at 0 °C exhibited a quick reduction from about (1.6 to 0.6) EBC unit. Also the alcohol chill haze (ACH) reduced from (28.5 to 2.2) EBC unit. Such an indicative test for colloidal stability is used to estimate the probable rate of haze production and monitor the effectiveness of stabilization treatments (Mélotte, 2008). Thus, the lower the chill haze value the greater the colloidal stability is. On the contrary, the time course of J_v was indeed affected by the presence of PVPP granules. In spite of Meier's suggestions (1993), J^* and $J_{v,av}$ fell to (71 ± 3) and $(84 \pm 4) \text{ L m}^{-2} \text{ h}^{-1}$, respectively (Fig. 3.15). In the circumstances, it was undeserving to recirculate the PVPP-rich beer through the ceramic tubular membrane module, especially if $J_{v,av}$ is to be kept to $\sim 300 \text{ L m}^{-2} \text{ h}^{-1}$ to minimize CFMF operating costs.

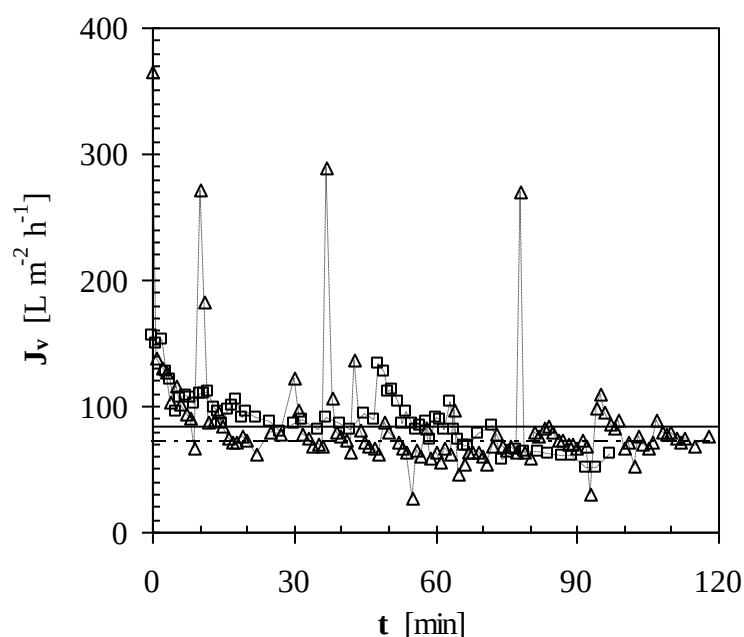


Figure 3.15 Time course of the permeation flux (J_v) of EC-pretreated rough beer samples seeded with 0.3 ($\triangle$) or 0.5 ($\square$) g L⁻¹ of regenerable PVPP under constant TMPD (3.74 bar), v_s (6 m s⁻¹), temperature (10 °C), and periodic CO₂ back-flushing. The continuous and broken lines refer to the quasi-steady state (J^*) and average permeation flux ($J_{v,av}$). The average coefficient of variation for J_v was 9%.

Separate stabilization and crossflow microfiltration of EC-pretreated beer

The process flow sheet shown in Fig. 3.16 was thus sketched out to limit the fouling contribution of yeast cells, aggregates and polysaccharides during CFMF, as well as to enhance the effectiveness of PVPP stabilization and regeneration. Once spent PVPP-polyphenol complexes had settled to the bottom of the cylindro-conical tanks, they were purged away. Table 3.9 shows the characteristics (TP, haze at 20 and 0 °C) of EC-pretreated beer samples before and after PVPP stabilization.

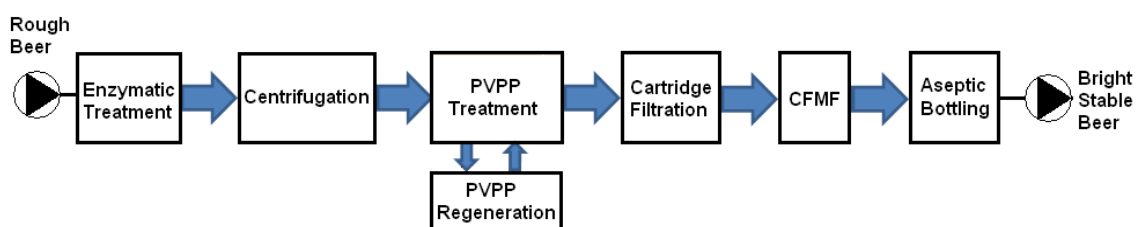


Figure 3.16 Schematic flow sheet of the novel lager beer separate PVPP stabilization and CFMF clarification process used in this work.

In both cases tested, the final TP content in stabilized beer was the $(40 \pm 1)\%$ of the initial TP one, this making the beer haze quite stable also at 0 °C. In fact, the difference in turbidity at (20 and 0) °C reduced from about 6.6 EBC unit (before stabilization) to (0.1 or 0.2) EBC unit for stabilized beer (Table 3.9).

Table 3.9 *Separate stabilization of EC-pretreated rough beer B samples seeded with 0.5 g L⁻¹ of PVPP: effect of contact time (t) on the mean and standard deviation of the polyphenol content (PF), and turbidity (H) at (20 and 0) °C.*

RBEC samples	t [h]	PF [mg L ⁻¹]	H _{20 °C} [EBC unit]	H _{0 °C} [EBC unit]
1	0	215 ± 3	1.56 ± 0.02	8.2 ± 0.3
	24	127 ± 2	0.85 ± 0.03	1.0 ± 0.1
2	0	221 ± 2	1.58 ± 0.04	8.2 ± 0.3
	24	134 ± 17	0.90 ± 0.04	1.1 ± 0.2

In the stabilized beer B recovered the total solid content, this including residual PVPP, was equal to (61 ± 15) mg L⁻¹ and generally higher than that (38 ± 7) mg L⁻¹ detected in C-pretreated beer samples. Thus, to simulate the final industrial beer filtration via the so called Filtrap cartridges (pore size: 3 µm), manufactured for instance by Filtrox AG (St Gallen, CH), such an excess of suspended solids was removed by means of 2.7-µm filters. However, such a finishing operation had a statistically insignificant effect on the turbidity at 20 or 0 °C of the EC-pretreated and stabilized beer specimens used in the total recycle CFMF tests.

The resulting filtrates were individually fed to the CFMF module operating under the aforementioned operating variables. Fig. 3.17 shows the time course of the permeation flux (J_v) and overall membrane resistance (R_T) under periodic CO₂ backwashing. In the circumstances, the quasi-steady state permeation fluxes (J^*) tended to (162 ± 11) L m⁻²h⁻¹. Such values were strictly related to the turbidity of the beer B fed to the CFMF plant (see closed ▲ symbols in Fig. 3.12). Similarly, the associated values of the cake filtration constants (k_{CF}), listed in Table 3.7 and represented by the closed ◆ symbols, scattered around the broken line in Fig. 3.12.

Fig. 3.17a)

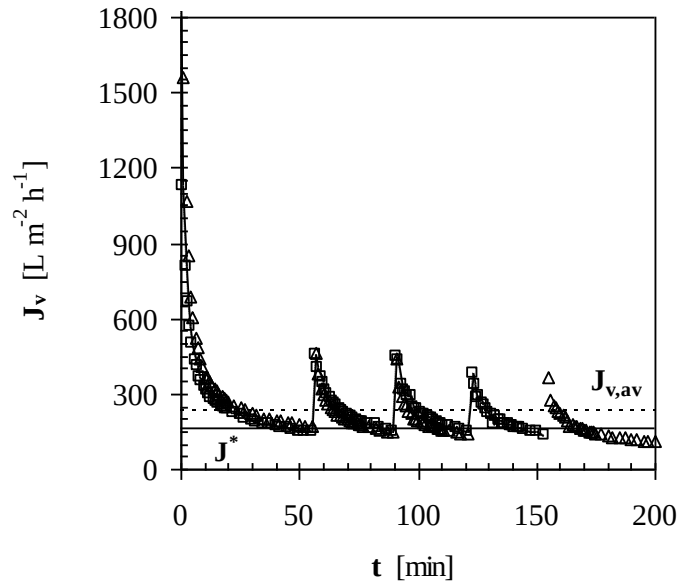


Fig. 3.17b)

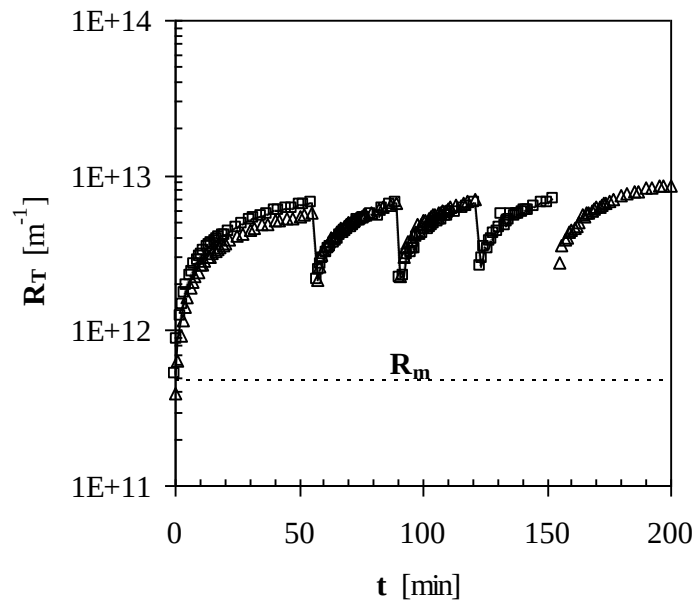


Figure 3.17 Time course of the (a) permeation flux (J_v) and (b) overall hydraulic resistance (R_T) of rough beer samples submitted to sequential enzymatic and centrifugal pretreatments and PVPP stabilization (RBECS1: $\triangle$; RBECS2: $\square$) under the CFMF conditions given in the text and periodic CO_2 back-flushing. For all characteristics of beer samples see Table 3.6. The continuous and broken lines in Fig. 3.17a refer to the quasi steady-state (J^*) and average ($J_{v,av}$) permeation fluxes, while the broken line in Fig. 3.17b refers to the clean membrane hydraulic resistance (R_m). The average coefficient of variation for J_v and R_T was about 9%.

Contrary to the previous results shown in Fig. 3.13b, the periodic CO₂ flushing lowered R_T to $(2.1 - 2.7) \times 10^{12} \text{ m}^{-1}$, that is to values equal to about 5-6 times that of the clean membrane resistance. This resulted in an average permeation flux ($J_{v,av}$) of about 229 L $\text{m}^{-2} \text{ h}^{-1}$ (Table 3.7). Owing to the enzymatic pretreatment, the β -glucan content of the rough beer samples under study reduced from (214.5 ± 0.8) to $(0.95 \pm 0.27) \text{ mg L}^{-1}$, while its final content in beer permeate further lessened to $(0.57 \pm 0.27) \text{ mg L}^{-1}$ as the result of the subsequent CFMF process.

3.3 Final validation tests for the combined stabilization and crossflow microfiltration of an industrial rough pale lager.

In this final tests, different lots of a pale lager, produced in the industrial brewery Birra Peroni (Rome, Italy), were used on the bench-top plant, shown in fig.2.1, equipped with a 0.8- μm ceramic tubular membrane module, under constant crossflow velocity (v_s) of 6 m s^{-1} , transmembrane pressure difference (TMPD) of 3.74 bar, temperature of $\sim 10^\circ\text{C}$, and periodic CO₂ backflushing. The sequential clarification, PVPP stabilization, and CFMF procedure shown in fig 3.16, was assessed using this industrial rough lager beer, as such or pre-centrifuged (C) to remove yeast cells and larger aggregates. The beer or permeate samples were assayed for pH, density, viscosity, turbidity or haze (H) at (20 and/or 0) $^\circ\text{C}$, color, as well as β -glucans and total phenol (TP) contents, real or original extract, and ethanol, in accordance with Analytica EBC (2010), as shown in Table 3.10. Generally, the beer permeates collected had the following characteristics: pH (4.3 ± 0.1); density ($1006.6 \pm 0.1 \text{ kg m}^{-3}$); viscosity ($1.399 \pm 0.001 \text{ mPa s}$); color ($6.9 \pm 0.3 \text{ EBC unit}$); β -glucans ($16.8 \pm 0.9 \text{ mg L}^{-1}$); total phenols ($85.6 \pm 2.3 \text{ mg L}^{-1}$); alcohol degree ($4.70 \pm 0.03 \% \text{ v/v}$); real extract ($3.47 \pm 0.02^\circ\text{P}$); original extract ($12.51 \pm 0.02^\circ\text{P}$).

Anaerobic microorganism, lactic bacterium and wild yeast cell counts of beer samples, collected from the retentate and permeate sides of the bench-top plant used at the end of the total recycle tests, were made in accordance with Analytica EBC (2005), and certified by CERB (Perugia, Italy).

Table 3.10 Mean and standard deviation of the total polyphenol content (TP), and turbidity (H) at (20 and 0) °C of beer samples, as such, precentrifuged, PVPP stabilized, rough filtered, and/or micro-filtered (CFMF), together with the quasi-steady state (J_v^*) and average ($J_{v,av}$) permeation fluxes observed in some total recycle tests carried out at $T \approx 10$ °C, $TMPD = 3.74$ bar, $v_s = 6$ m s⁻¹, and periodic CO₂ backflushing.

Rough beer samples	t [h]	TP [mg L ⁻¹]	H₂₀ °C [EBC unit]	H₀ °C [EBC unit]	J[*] [L m ⁻² h ⁻¹]	J_{v,av} [L m ⁻² h ⁻¹]
<i>Rough beer as such</i>						
RB1	0	176 ± 2	12.1 ± 0.1	13.3 ± 0.2		
	CFMF	162 ± 5	0.48 ± 0.01	2.32 ± 0.07	62 ± 6	86
RB2	0	185 ± 2	15.6 ± 0.1	17.5 ± 0.2		
	CFMF	179 ± 3	0.47 ± 0.01	3.62 ± 0.07	60 ± 6	89
<i>Pre-centrifuged beer</i>						
RB3	0	187±2	25.7 ± 0.1	112±6		
	CFMF	162±4	0.59 ± 0.01	1.1±0.1	129 ± 12	267
RB4	0	189±3	12.8 ± 0.1	23±6		
	CFMF	173±2	0.55 ± 0.01	1.2±0.1	154 ± 10	237
<i>Pre-centrifuged and PVPP stabilized beer</i>						
RB5	0	165 ± 1	1.40 ± 0.03	2.67 ± 0.21		
	24	89 ± 4	1.03 ± 0.12	1.23 ± 0.19		
	CFMF	nd	0.35 ± 0.02	0.45 ± 0.03	139 ± 16	183
RB6	0	199 ± 4	1.70 ± 0.04	18.58 ± 0.79		
	24	121 ± 6	7.43 ± 0.16	10.92 ± 0.68		
	CFMF	nd	0.49 ± 0.03	0.59 ± 0.32	114 ± 12	141
RB7	0	192 ± 6	1.85 ± 0.07	16.78 ± 0.05		
	24	108 ± 8	1.41 ± 0.32	2.68 ± 0.39		
	CFMF	nd	0.43 ± 0.07	0.57 ± 0.02	100 ± 10	158
<i>Pre-centrifuged, PVPP stabilized, and pre-filtered beer</i>						
RB8	0	227 ± 7	1.71 ± 0.14	17.0 ± 0.4		
	24	138 ± 6	0.76 ± 0.05	0.91 ± 0.06		
	CFMF	84±2	0.23 ± 0.02	0.31 ± 0.02	137 ± 9	338
RB9	0	263 ± 4	1.71 ± 0.14	32 ± 1		
	24	147 ± 6	0.77 ± 0.05	0.91 ± 0.08		
	CFMF	87 ± 1	0.22 ± 0.01	0.31 ± 0.06	139 ± 7	336

nd not determined.

Fig. 3.18 compares the time course of the experimental permeation flux (J_v), when using the aforementioned rough pale lager samples and CO₂ backwashing program; while Table 3.10 lists the main characteristics (i.e., total phenol content, TP; haze, H, at 20 or 0 °C) of the rough beers tested and resulting CFMF permeates together with the

mean values of the quasi-steady state (J^*) and average ($J_{v,av}$) permeation fluxes observed in total recycle tests.

Fig. 3.18a)

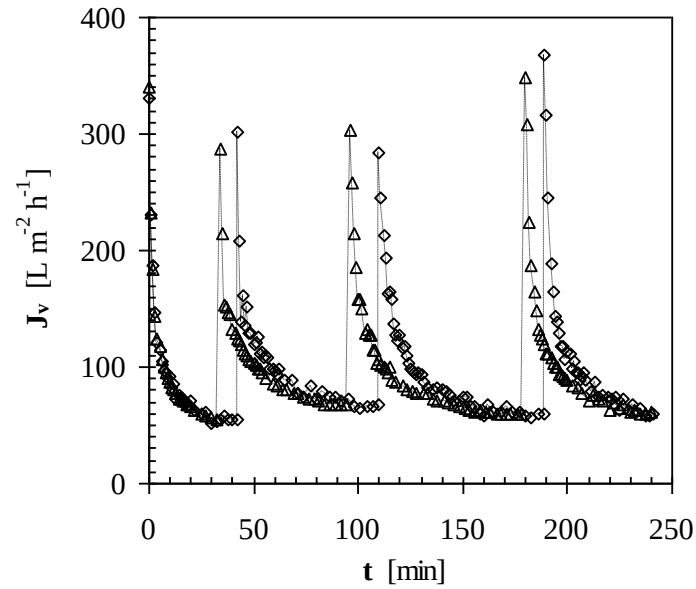


Fig. 3.18b)

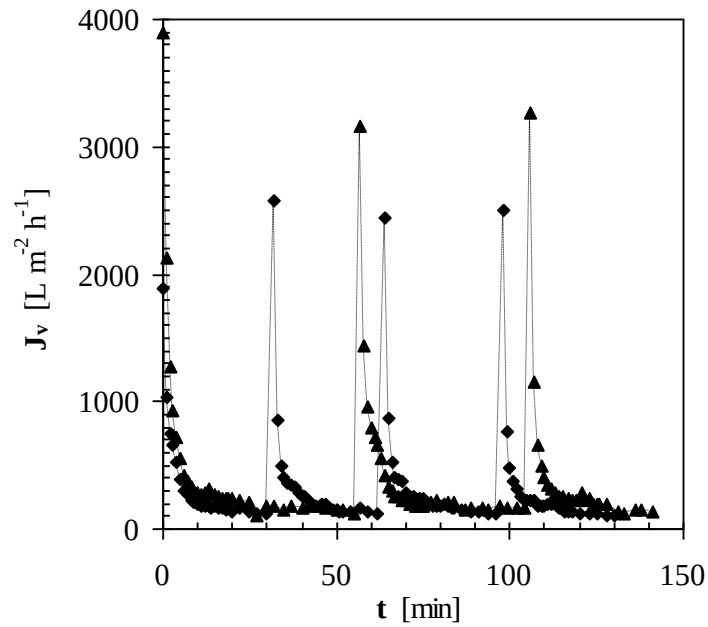


Figure 3.18 Time course of the permeation flux (J_v) of rough beer samples, (a) as such (RB1, $\triangle$; RB2, $\diamond$) or (b) precentrifuged (RB3, $\blacktriangle$; RB4; $\blacklozenge$), under the CFMF conditions given in the text and periodic CO_2 back-flushing. For all characteristics of rough beer samples see Table 1. The average coefficient of variation for J_v was about 10%.

In both cases, in about half an hour J_v tended to an asymptotic value (i.e., the limiting or quasi-steady state permeation flux, J^*). Thanks to the removal of solid particles larger than 0.5 μm by centrifuging, these being responsible for membrane pore blocking according to Norit Process Technology BV (Enschede, NL) (Buttrick, 2007), J^* for pre-centrifuged rough beer tended to $(142 \pm 18) \text{ L m}^{-2} \text{ h}^{-1}$, this about doubling that achieved with the rough beer as such $(62 \pm 6 \text{ L m}^{-2} \text{ h}^{-1})$, as shown in Table 3.10. As the result of the periodic CO_2 backflushing, the average permeation flux ($J_{v,\text{av}}$) increased from (86 ± 8) to $(252 \pm 21) \text{ L m}^{-2} \text{ h}^{-1}$, respectively.

The beer permeates collected during these trials exhibited an average haze at 20 °C of (0.54 ± 0.06) EBC unit, and thus complied with the EBC specification for a clear, bright beer (< 0.6 EBC unit). Unfortunately, they still retained some soluble haze precursors responsible for post-filtration hazes, their chill haze at 0 °C increasing from 1.1 to 3.6 EBC unit (Table 3.10).

Similarly to the great majority of DE-filtered beers, even the membrane-filtered beer had to be stabilized using for instance PVPP (Taylor et al., 2006). Since such a post-filtration treatment impairs the sterility of beer permeates, it might be replaced with a pre-filtration step of rough beer in the presence of non-regenerable PVPP, followed by centrifuging and then CFMF. The major drawback of such a combined procedure would be the high contribution of non-regenerable PVPP to the overall beer filtration costs (see below).

3.3.1 Beer stabilization using regenerable PVPP

After centrifuging, the beer was charged into four 1.5-L cylindro-conical tanks, enriched with 0.5 g L^{-1} of regenerable PVPP, and manually mixed. A blank test was also carried out in another tank. After a contact time of 18 or 24 h at $(0.0 \pm 0.5) ^\circ\text{C}$, spent PVPP-polyphenol aggregates were allowed to sink to the bottom of the tank and then removed. Stabilized beer, as such or after vacuum filtering through 2.7- μm Whatman filters (cat. No. 1823 047), was submitted to total recycle CFMF runs, as reported above.

The measurement of all polyphenol species (TP) present in rough beer via the EBC method does not appraise the polyphenols responsible for beer colloidal stability

(Mélotte, 2008). However, such a measure reveals the fraction of low and medium molecular polyphenols precipitated by PVPP, such as catechin and anthocyanogens, both of them affecting in some way the beer colloidal and taste stability (Mélotte, 2008).

Previously (§ 3.2.2), the percentage TP removal coefficient by PVPP was found to be of the order of (35 or 45) % after contact times (t) of about 4 or 15 min. To achieve a good sedimentation, the PVPP stabilization procedure was thus prolonged up to 24 h, thereafter, the spent PVPP-polyphenol complexes settled to the bottom of the cylindro-conical tanks, were withdrawn and tested for their total solid content ($1.25 \pm 0.28\%$ p/p), and then submitted to a classic PVPP regeneration process (Gopal and Rehmanji, 2000). In such a slurry, almost all yeast and suspended solids were preliminarily removed by centrifuging.

Table 3.10 shows the characteristics (TP, haze at 20 and 0 °C) of pre-centrifuged beer at the beginning (t=0 h) and end (t=24 h) of the PVPP stabilization step, this being carried out at above 0 °C for 24 h. It can be noted that such a treatment allowed the original total phenol content ($185 \pm 18 \text{ mg L}^{-1}$) of rough beer to be reduced by $(57 \pm 4)\%$. The mean haze values at 20 or 0°C of beer after CFMF tended to (0.42 ± 0.07) or (0.63 ± 0.22) EBC unit, respectively. Probably because of the fact that not all the finer PVPP particles had been removed during their settling to the bottom of the cylindro-conical tanks, the average permeation flux reduced to $(161 \pm 21) \text{ L m}^{-2} \text{ h}^{-1}$, as shown in Fig. 3.19.

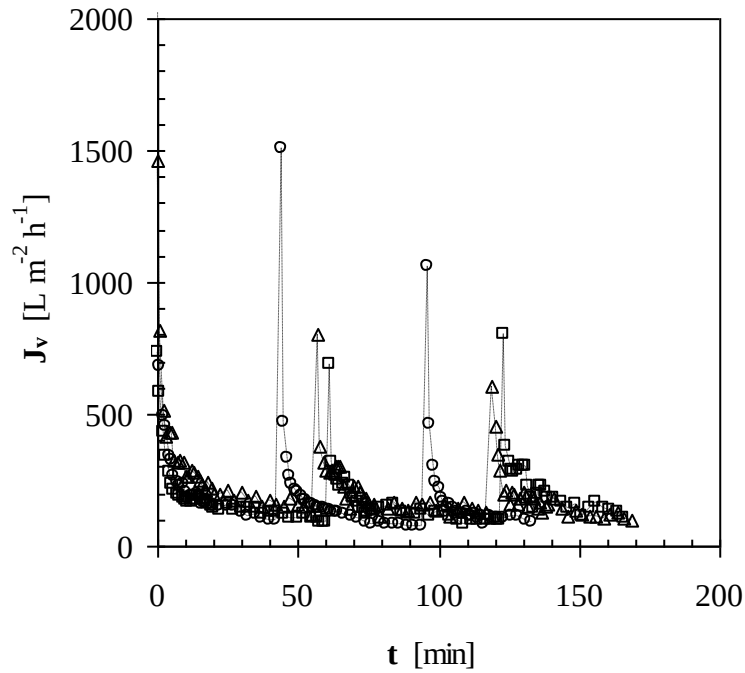


Figure 3.19 Time course of the permeation flux (J_v) of precentrifuged and PVPP stabilized rough beer samples (RB5: $\triangle$, 13.06.13; RB6: $\square$, 27.06.13; RB7: $\circ$, 22.07.13) under constant TMPD (3.74 bar), v_s (6 m s^{-1}), temperature (10°C), and periodic CO_2 back-flushing. The average coefficient of variation for J_v was 10%.

In order to simulate the final industrial beer filtration via the so called Filtrap cartridges (pore size: $3 \mu\text{m}$) (Eßlinger, 2009), such an excess of finer suspended particles was removed via a $2.7\text{-}\mu\text{m}$ filter. The resulting filtrates were then fed the CFMF unit, this being operating under the aforementioned operating variables. Fig. 3.20 shows the time course of the permeation flux (J_v) under periodic CO_2 backwashing. In the circumstances, the quasi-steady state permeation fluxes (J^*) approached $(138 \pm 8) \text{ L m}^{-2} \text{ h}^{-1}$, whereas the average permeation flux ($J_{v,\text{av}}$) re-enhanced to $(337 \pm 1) \text{ L m}^{-2} \text{ h}^{-1}$ thanks to the periodic CO_2 backflushing (Table 3.10).

In the circumstances, the quasi-steady state permeation fluxes (J^*) approached $(138 \pm 8) \text{ L m}^{-2} \text{ h}^{-1}$, whereas the average permeation flux ($J_{v,\text{av}}$) re-enhanced to $(337 \pm 1) \text{ L m}^{-2} \text{ h}^{-1}$ thanks to the periodic CO_2 backflushing

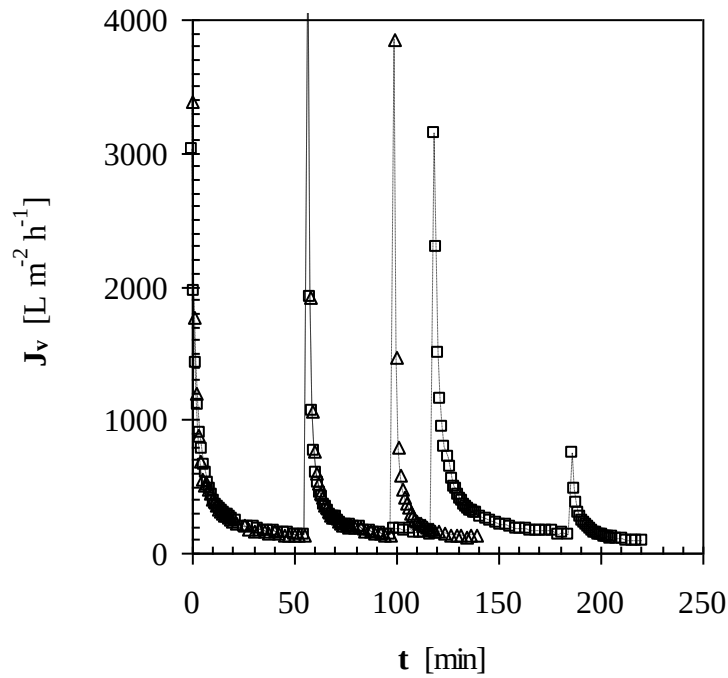


Figure 3.20 Time course of the permeation flux (J_v) of rough beer samples submitted to sequential centrifuging, PVPP stabilization, and cartridge filtration (RB8: $\triangle$, 25.02.14; RB9: $\square$, 27.02.14) under the CFMF conditions given in the text and periodic CO_2 back-flushing. For all characteristics of beer samples see Table 1. The average coefficient of variation for J_v and R_T was about 10%.

Contrary to previous testing on a malt-based rough beer containing as much as 140-250 mg L^{-1} of β -glucan (Cimini et al., 2013ab; 2014), the rough pale lager under study was prepared using about 10.8 kg of malted barley and 4.7 kg of maize grits per hL, and, thus, contained as little as $(16.8 \pm 0.9) \text{ mg of } \beta\text{-glucan L}^{-1}$. In the circumstances, the β -glucanase and pentosanase pretreatment previously used appeared to be useless. Thus, even in this case, by resorting to process flow sheet sketched in Fig. 3.16, it was possible to limit the fouling contribution of yeast cells and aggregates during beer CFMF by the sequential steps of centrifuging, PVPP stabilization and cartridge filtration. In fact, the resulting average value of the permeation flux was by far greater than that (80 to 100) $\text{L m}^{-2} \text{ h}^{-1}$ claimed at (0-2) $^{\circ}\text{C}$ by the three CFMF processes commercially available (Buttrick, 2007).

Moreover, to assess the effectiveness of the cold sterilization process, microbial (anaerobic bacteria and wild yeasts) cell retention by the 0.8- μm membrane was assessed, as shown in Table 3.11. It can be noted that their residual concentration was

less than 1 CFU mL⁻¹. Thus, the novel procedure described here was successful at yielding a chill haze-free and microbiologically stable permeate ready for aseptically packaging with no supplementary pasteurization step.

Table 3.11 Typical assessment of the microbial contamination for the beer retentate and permeate samples collected at the end of a generic CFMF total recycle test.

Microbial cell count	Ref.	Retentate	Permeate	Unit
Anaerobic microorganisms (@ 7 days)	EBC 4.3.1.4	negative	negative	UFC mL ⁻¹
Lactic Acid Bacteria	EBC 4.3.3.1	192±24	<1	UFC mL ⁻¹
Wild Yeast Count	EBC 4.2.2	<1	<1	UFC mL ⁻¹

3.4 Economic feasibility and environmental impact of the novel clarification and stabilization process

To assess the economic feasibility of the novel CFMF-based procedure sketched in Fig. 3.16 with respect to the conventional one using powder filters (DEF) (Fig. 3.21)

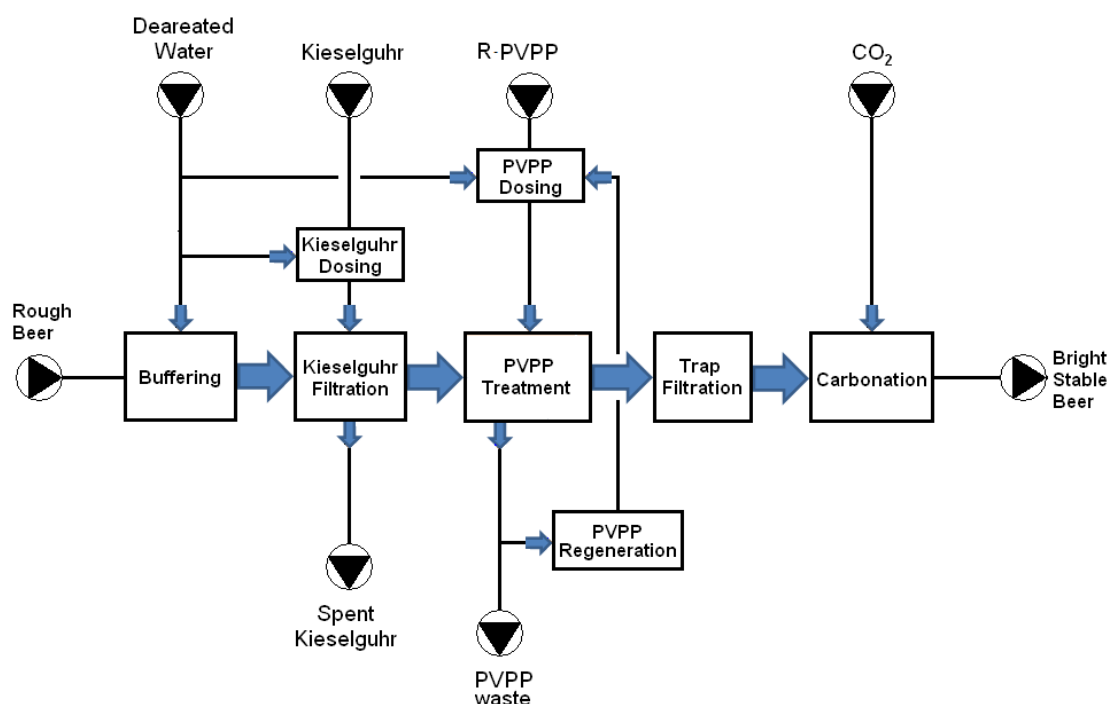


Figure 3.21 Schematic diagram of the conventional pale lager clarification and stabilisation process using kieselguhr with regenerable (R) PVPP, as applied by the industrial brewery Birra Peroni Srl (Rome, Italy).

in conjunction with single-use (SU) or regenerable (R) PVPP, a rough-grade feasibility study was carried out by referring to an industrial brewery working for 300 days per annum in three shifts per day (i.e., 20 h/day including the membrane cleaning-in-place procedure) with an overall capacity of about 2×10^6 hL of pale lager (this being of the same order of magnitude of the Birra Peroni Srl brewery from which the rough pale lager used here was obtained). In particular, a high capacity solids-ejecting centrifuge (i.e., the Brew 3001 type <http://www.alfalaval.com/solution-finder/products/brewseries/Documents/BREW%203001.pdf>) was used to feed a continuously operating CFMF unit, this being composed of several M-36P3740 modules, each one installing 36 Pall® Membralox ceramic membrane EP3740 monoliths (Pall Corporation, 2007), the main geometric characteristics of which being reported below.

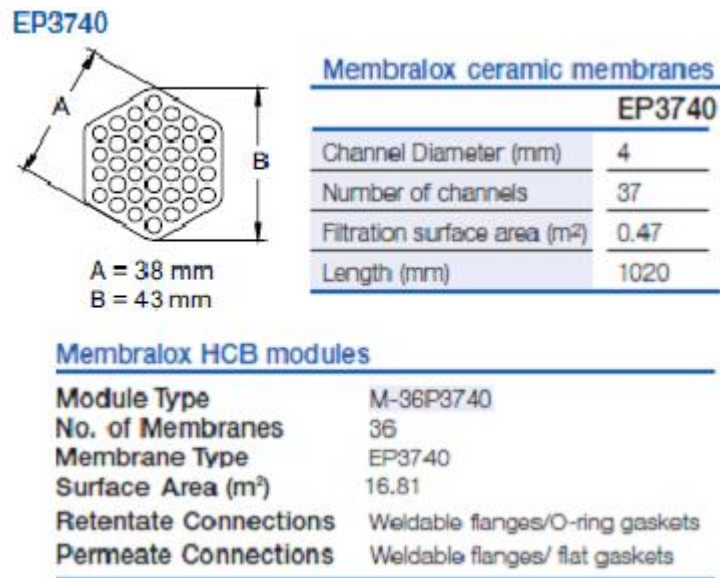


Fig. 3.21 Main geometric characteristics of chosen ceramic membrane

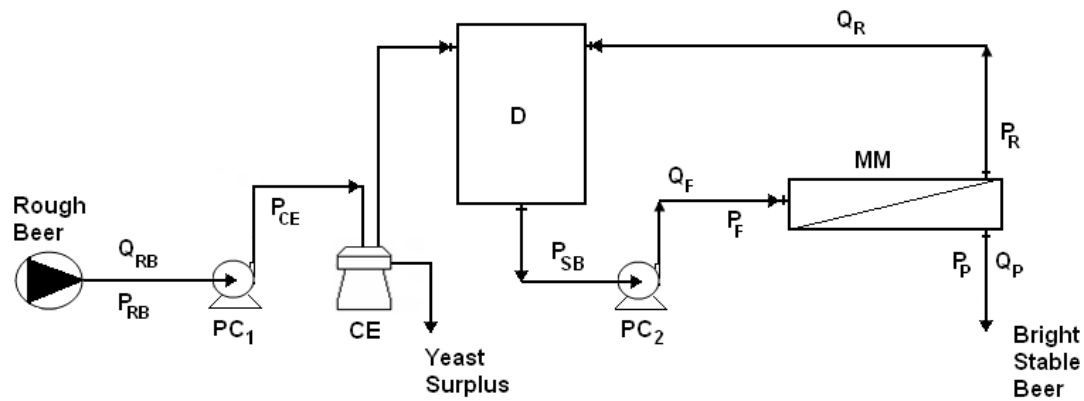


Fig. 3.22 Flow sheet of the novel lager beer clarification and stabilisation process using tubular ceramic microfiltration membranes developed in this work. Equipment, product and by-product identification items: CE, centrifuge; D, storage tank for clarified and PVPP stabilized beer; MM, CFMF membrane module; PC1, primary centrifugal pump; PC2, recirculation centrifugal pump; Q_F , Q_P , Q_R , Q_{RB} , CFMF feed, permeate, retentate and rough beer flow rates; P_{CE} , P_F , P_{RB} , P_{SB} , input CE, MM, PC1 and PC2 pressures; P_P , P_R , permeate- and retentate-side output pressures.

By assuming a design permeation flux ($J_{v,d}$) of $300 \text{ L m}^{-2} \text{ h}^{-1}$ (this being safely reduced by about 10% with respect to the average permeation flux experimentally observed here) and setting the feed superficial velocity in each channel of the membrane modules and transmembrane pressure difference (TMPD) to the aforementioned operating values, that at 6 m s^{-1} and 3.7 bar, respectively, it was possible to list in Table 3.12 the main design parameters of the CFMF unit, its flow sheet being shown in Fig. 3.22. The specific consumption yields for the main ancillary materials (*i.e.*, kieselguhr, K; kieselguhr sludges, KS; process water, PW; wastewaters, WW; single use or regenerable PVPP) and utilities (*i.e.*, electric energy, EE; live steam, S) per each hL of bright and stabilised pale lager produced were directly derived from Birra Peroni Srl (Rome, Italy), these data being also checked with the technical literature (Bock & Rögener, 2002; Leeder *et al*, 2011; Mueller & Witte, 2008).

In particular, the operating costs for the automatic cleansing of the DE-filters or ceramic membrane modules were, for the sake of simplicity, assumed as equal for both alternatives and not accounted for. Similarly, the labour and investment-related costs, except the yearly membrane replacement costs, these being of the order of 500 € m^{-2} (Mueller & Witte, 2008), were regarded of the same order of magnitude and not considered here. Moreover, the rinsing water and steam consumption to regenerate the PVPP granules was assumed as independent of the process alternative under study and equal to that registered by Birra Peroni Srl (Rome, Italy).

Table 3.12 Main design parameters of main equipment of the novel pale lager clarification and stabilisation process using tubular ceramic microfiltration membranes developed in this work. All symbols are defined in the Notation section.

Design Parameter	Value	Unit
Brewery size	2×10^6	hL yr^{-1}
Working period	300	days yr^{-1}
Filtration time including CIP	20	h day^{-1}
Rough beer throughput (Q_P)	333.3	hL h^{-1}
Centrifugal pump PC1		
Rough beer input pressure P_{RB}	1	bar
Rough beer output pressure P_{CE}	4	bar
Motor power installed N_{PC1} (overall efficiency 65%)	4.5	kW
Specific electric energy consumed	0.013	kWh hL^{-1}
Solids-ejecting centrifuge CE type BREW 3001		
Maximum volumetric flow rate	900	hL h^{-1}
Motor power installed	55	kW
Operation time	0.370	h
Specific electric energy consumed	0.061	kWh hL^{-1}
CFMF unit		
Design Permeation Flux	300	$\text{L m}^{-2} \text{h}^{-1}$
Membrane surface area (A_m) to be installed	111	m^2
Membralox M-36P3740 module: monolith no.	36	-
Geometry of monolith EP3740		
channel diameter	4	mm
channel no.	37	-
channel length	1.02	m
Overall module membrane surface area	16.81	m^2
M-36P3740 module no.	7.0	
Channel superficial velocity (v_s)	6	m s^{-1}

Input module flow rate of centrifuged beer	3615	hL h ⁻¹
Overall centrifuged beer flow rate (Q_F)	25308	hL h ⁻¹
Module pressure drop	1.2	bar
Membrane life time	10	yr
Membrane surface area replaced per annum	11.8	m ² yr ⁻¹
<i>Centrifugal pump PC2</i>		
Stabilized beer input pressure (P_{SB})	4.0	bar
Stabilized beer output pressure (P_F)	5.3	bar
Motor power installed N_{PC2} (overall efficiency 72%)	126.9	kW
Specific electric energy consumed	0.381	kWh hL ⁻¹

Finally, to assess roughly the environmental performance of the process alternatives under study, the single impact category of climate change, expressed by accounting for the six greenhouse gas (GHG) emissions in the Earth atmosphere covered by the Kyoto Protocol in CO₂ equivalent over a time-horizon of 100 years, according to the PAS 2050 procedure (BSI, 2008), was estimated. To this end, the 100-yr Global Warming Potential (GWP) for all ancillary materials and utilities used were extracted from the software SimaPro 7 vers. 7.2.2 (PRé Consultants, 2010) or from the technical literature (ISPRA, 2012; Sika Services AG, 2012), as reported in Table 3.13.

Table 3.13 Main design parameters of main equipment of the novel pale lager clarification and stabilisation process using tubular ceramic microfiltration membranes developed in this work. All symbols are defined in the Notation section.

Processing step	Specific Consumption	Alternative Process				Unitary Cost		GWP		
		DEF & R-PVPP	DEF & SU-PVPP	CFMF & R-PVPP	CFMF & SU-PVPP	Value	Ref.	Value		
Filtration	K									
	[kg hL ⁻¹]	0.112	0.112	0	0	0.700 [€ kg ⁻¹]	Leeder <i>et al.</i> (2011)	1.267 [kg CO _{2e} kg ⁻¹]		
	KS [kg hL ⁻¹]	0.336	0.336	0	0	0.170 [€ kg ⁻¹]	Fillaudeau <i>et al.</i> (2006)	0.500 [kg CO _{2e} kg ⁻¹]		
	PW [m ³ hL ⁻¹]	0.032	0.032	0.050	0.050	0.360 [€ m ⁻³]	Leeder <i>et al.</i> (2011)	0.0003 [kg CO _{2e} kg ⁻¹]		
	WW [m ³ hL ⁻¹]	0.0224	0.0224	0.035	0.035	0.050 [€ m ⁻³]	Leeder <i>et al.</i> (2011)	0.001 [kg CO _{2e} m ⁻³]		
Stabilization	EE [kWh hL ⁻¹]	0.08	0.08	0.45 ^a	0.45 ^a	0.151 [€ kWh ⁻¹]	Bortoni (2013)	0.3931 [kg CO _{2e} kWh ⁻¹]		
	S [kg hL ⁻¹]	1.80	0.000	1.80	0	0.015 [€ kg ⁻¹]	Leeder <i>et al.</i> (2011)	0.234 [kg CO _{2e} kg ⁻¹]		
	PVPP [g hL ⁻¹]	0.11	30	0.11	30	60 or 10 ^b [€ kg ⁻¹]	Morlino (2014)	2.408 [kg CO _{2e} kg ⁻¹]		
Pasteurization	S [kg hL ⁻¹]	5.8	5.8	0	0	0.015 [€ kg ⁻¹]	Leeder <i>et al.</i> (2011)	0.234 [kg CO _{2e} kg ⁻¹]		
	PW [m ³ hL ⁻¹]	0.05	0.05	0	0	0.360 [€ m ⁻³]	Leeder <i>et al.</i> (2011)	0.0003 [kg CO _{2e} kg ⁻¹]		
	EE [kWh hL ⁻¹]	0.33	0.33	0	0	0.151 [€ kWh ⁻¹]	Bortoni (2013)	0.3931 [kg CO _{2e} kWh ⁻¹]		
	A _m [m ² hL ⁻¹]	0	0	5.88x10 ⁻⁶	5.88x10 ⁻⁶	500 [€ m ⁻²]	Mueller & Witte (2008)	25.0 [kg CO _{2e} m ⁻²]		
Overall operating costs	C _o [€ hL ⁻¹]	0.35	0.62	0.12	0.39					
Overall GWP	GWP [kg CO _{2e} hL ⁻¹]	2.25	1.90	0.60	0.25					

a, related to the electric energy consumption of the centrifugation step, **b**, referred to R or SU PVPP, respectively.

The use of regenerable PVPP resulted to be beneficial whatever the filtration procedure used. Nevertheless, the use of CFMF together with regenerable PVPP reduced not only the overall beer clarification and stabilization costs (C_o) to ~36% of those associated with the powder filtration (that is from 0.35 to 0.12 € hL⁻¹); but also the overall environmental impact by ~27%, that is from (2.25 to 0.60) kg CO_{2e} hL⁻¹. The CFMF in conjunction with single use PVPP resulted in the minimum global warming potential (0.25 kg CO_{2e} hL⁻¹), but the overall operating costs increased to 0.39 € hL⁻¹.

3.4 Final procedure for a novel clarification and stabilization process using CFMF membrane

To minimise the fouling contribution of yeast cells, aggregates and polysaccharides during CFMF and maximize the effectiveness of PVPP stabilization and regeneration, the process flow sheet shown in Fig. 3.23 was outlined.

The same cylindro-conical tank D1 used to ferment the wort allows the yeast to settle to its bottom at the end of fermentation and to be collected in tank D4. By feeding the rough beer to the centrifuge C1, a yeast cream is recovered and conveyed to D4, while the centrifuged beer is fed to tank D2. A thick slurry of regenerable PVPP is then added at a rate of 0.2 to 0.5 g L⁻¹ by a proportioning pump. An inert gas (CO₂ or N₂) is also sparged to ensure thorough dispersion and minimum O₂ pick-up.

After a contact time of 24 h at ~0 °C, spent PVPP-polyphenol complexes are firstly removed after settling and accumulated in tank D6. The stabilized beer is then pumped to the CFMF module MM1 (pore size: 0.8 µm). By setting the aforementioned operating variables, it is expected a bright, stabilized permeate at an average permeation flux of ~300 L m⁻² h⁻¹, ready for aseptically packaging without an additional pasteurization step. Another CFMF system MM2, equipped with ceramic tubular membrane modules too, is used not only to recover sequentially the remaining beer from either yeast or PVPP slurry, its corresponding retentate being recirculated through tank D4 or D6, respectively; but also to regenerate used PVPP (Gopal and Rehmanji, 2000).

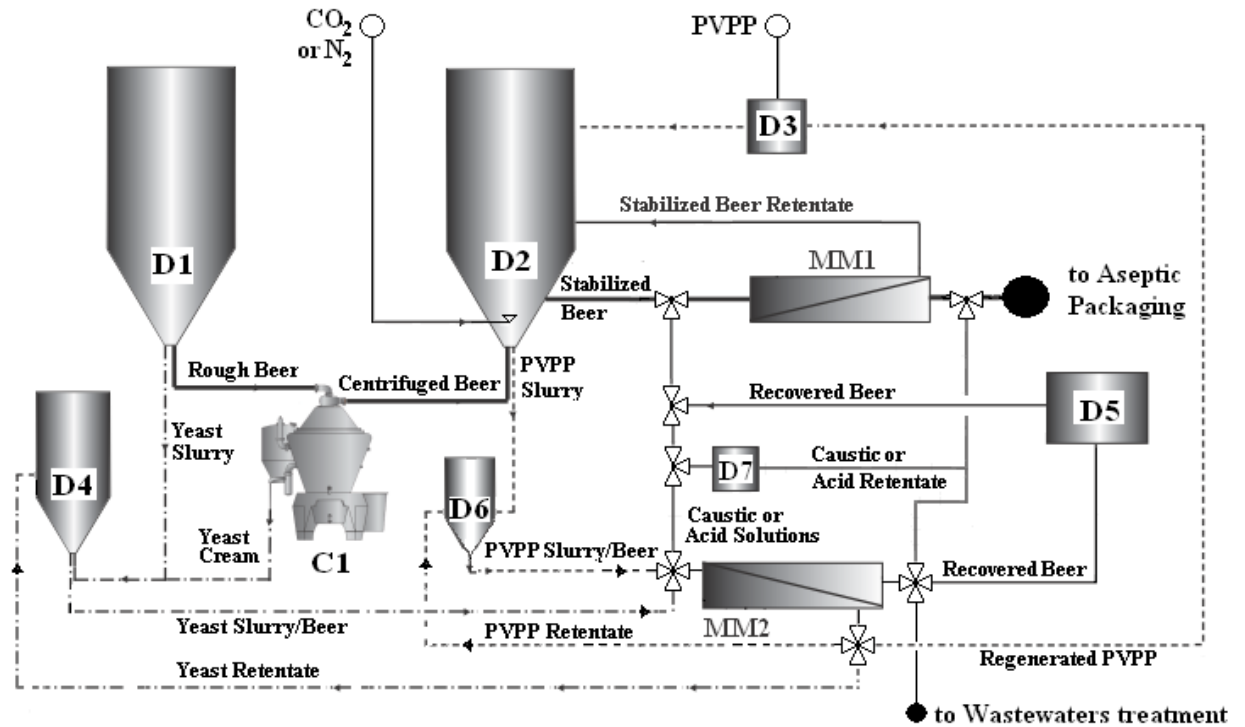


Figure 3.23 Schematic flowdheet diagram of the novel lager beer clarification and stabilisation process using tubular ceramic microfiltration membranes developed in this work. Equipment identification items: C, centrifuge; D1, fermentation and maturation uni-tank; D2, PVPP treatment tank; D3, PVPP dosing tank; D4, yeast tank; D5, recovered beer tank; D6, PVPP slurry tank; D7, tank for cleansing solutions; MM1, CFMF membrane module for beer clarification; MM2, CFMF membrane module for beer recovery from yeast or PVPP slurry or for PVPP regeneration. Product, by-product and ancillary identification lines: Beer, —; Yeast · — · —; PVPP, - -

CONCLUSIONS

By resorting to a laboratory-made green beer produced from commercial hopped-malt extracts, and an all malt lager beer, was possible to assess its crossflow microfiltration performance in a bench-top plant, appropriately designed and equipped with ceramic tubular membrane modules with nominal pore size of 0.4, 0.8 or 1.2 μm , as a function of the initial rough beer turbidity (H), feed superficial velocity (v_s) and trans-membrane pressure difference (TMPD) under constant temperature ($\sim 10\text{ }^\circ\text{C}$). and CO_2 backflushing

The cake filtration fouling mechanism was identified as the main reason for flux decline by analyzing mathematically the time course of both the experimental permeate volume and permeation flux data using Bolton *et al.*'s equations (2006). In particular, the cake filtration constant (k_{CF}) was about linearly related to H in the range of 0 to 21.3 EBC unit, whereas it levelled off in almost the same manner displayed by the initial and limiting permeation fluxes at H values as great as 54 - 62 EBC unit.

Using the optimal operating conditions and a 0.8- μm ceramic tubular membrane module, it was possible to achieve an average permeation flux ($J_{v,\text{av}}$) of $(252 \pm 21)\text{ L m}^{-2}\text{ h}^{-1}$ on condition that the industrial rough beer had been preliminary centrifuged. Yet, as the beer permeate had been cooled down to $0\text{ }^\circ\text{C}$, its chill haze was definitely greater than the recommended EBC standard value of 0.6 EBC unit. To obtain a brilliant, chill-proof and microbiologically stable beer, ready-to-be aseptically packaged, pre-centrifuged beer was firstly stabilized at $0\text{ }^\circ\text{C}$ for 24 h by seeding 0.5 g L^{-1} of regenerable PVPP, and, after purging the larger PVPP-polyphenol aggregates, fed to the CFMF unit. Whereas the permeate turbidity at $0\text{ }^\circ\text{C}$ was lowered to 0.6 EBC unit, $J_{v,\text{av}}$, reduced to $(161 \pm 21)\text{ L m}^{-2}\text{ h}^{-1}$, the finer PVPP granules probably enhancing the cake formation layer over the membrane surface. By pre-filtering the above stabilized beer, it was possible not only to re-enhance the average permeation flux to about $337\text{ L m}^{-2}\text{ h}^{-1}$ (i.e., within the range of values achievable with conventional DE-filters), but also to yield a bright and stabilized permeate ready for aseptically packaging with no supplementary pasteurization step. Such finding were further confirmed using a rough pale lager produced by the industrial brewery Birra Peroni srl (Rome, Italy)

A combined clarification and PVPP stabilisation procedure using 0.8- μm ceramic tubular membrane modules was developed and tested on a bench-top plant scale. By avoiding DE disposal problems and heat pasteurisation, and minimising beer losses, such a novel process appeared to be not only a reliable alternative to conventional powder filters in terms on the average permeation flux, but also a cost-effective one capable of mitigating the related

life cycle GHG emissions. By referring to an industrial plant capacity of 2×10^6 hL of lager beer, the estimated overall costs and global warming potential for lager beer clarification and stabilization were about one third of those associated with the current industrial DE-filtration and regenerable PVPP stabilisation procedures. It is also worthy of note that no exogenous enzymes were used to ease the CFMF clarification, this being however a direct consequence of the beer recipe used.

Further work is however still needed to check for the efficacy of the novel combined process operating at relatively higher temperatures (for instance, 10 °C, or 0-2 °C currently used during beer stabilization and filtration), as well as for the technical feasibility of beer recovery from PVPP slurry using a CFMF unit and PVPP regeneration from the resulting PVPP-rich retentate, both these phases affecting greatly beer finishing costs.

NOMENCLATURE

A	alcohol content of beer [% v/v]
A_m	effective membrane surface area [m^2]
BG	β -glucan content of beer [$g\ m^{-3}$]
C	beer colour [EBC unit]
CF	cake filtration
CFMF	crossflow microfiltration
CPB	complete pore blocking
c_{ss}	concentration of suspended solids [$mg\ L^{-1}$]
d	inside diameter of tubular membrane module [m]
FM	generic fouling model
H	beer turbidity [EBC unit]
IPB	intermediate pore blocking
i	generic i-th instantaneous value of any dependent variable y_j
J^*	quasi steady-state permeation flux [$L\ m^{-2}\ h^{-1}$]
J_v	instantaneous volumetric permeation flux [$L\ m^{-2}\ h^{-1}$]
j	generic dependent variable to be fitted by a fouling model
k	characteristic parameter of the unified filtration Eq. (3)
k_{CF}	cake filtration constant [$s\ m^{-2}$]
k_{CPB}	complete pore blocking constant [s^{-1}]
k_{IPB}	intermediate pore blocking constant [m^{-1}]
k_{PC}	pore constriction constant [m^{-1}]
L	length of tubular membrane module [mm]
L_w	membrane constant for water transport [$L\ m^{-2}\ h^{-1}\ bar^{-1}$]
$MSPE_j$	generic j-th mean squared percentage error, as defined by Eq. (29)
N	number of experimental data
n	characteristic parameter of the unified filtration Eq. (3) [dimensionless]
OE	beer original extract [$^{\circ}Plato$]
P	pressure [bar]
PC	pore constriction
p	number of independent fouling parameters
Q_F	feed volumetric flow rate [$m^3\ h^{-1}$]
r^2	coefficient of determination

R_{irr}	irreversible fouling resistance [m^{-1}]
R_m	intrinsic membrane resistance [m^{-1}]
R_{rev}	reversible fouling resistance [m^{-1}]
R_T	overall membrane resistance [m^{-1}]
R_T^*	quasi steady-state overall membrane resistance [m^{-1}]
RE	beer real extract [°Plato]
SPB	standard pore blocking
s_j^2	generic j-th residual variance, as defined by Eq. (28)
T	process temperature [°C]
t	process time [s or h]
TMPD	transmembrane pressure difference [bar]
V	cumulative volume of filtrate [L]
v_s	crossflow velocity [$m\ s^{-1}$]
y_j	generic j-th dependent variable

Greek Symbols

α	time function in cake filtration-pore constriction model, as defined by Eq. (22)
β	time function in cake filtration-pore constriction model, as defined by Eq. (23)
η	filtrate dynamic viscosity [mPa s]
ρ	density of filtrate [$kg\ L^{-1}$]

Subscripts

av	average
calc	calculated
exp	experimental
i	input
J	referred to permeation flux
o	out
V	referred to volume
W	referred to water
0	initial

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APPENDIX : Published papers, poster communications.

1. Cimini A, Marconi O, Moresi M (2013) Rough beer clarification by crossflow microfiltration in combination with enzymatic and/or centrifugal pretreatments. *Chemical Engineering Transactions* 32, 1729-1734.
2. Cimini A, Moresi M. (2013) Chiarificazione della birra con membrana ceramica tubolare. *Industria delle Bevande* 42 (245), 9-17.
3. Cimini A, Marconi O, Perretti G, Moresi M (2013) Novel procedure for lager beer clarification and stabilization using sequential enzymatic, centrifugal, regenerable pvpp and crossflow microfiltration processing, *Proc. 2013 CIGR Section VII International Technical Symposium on “Advanced food processing and quality management”*, Guangzhou, China, 3-7 November, CD ROM, contribution 128.
4. Cimini A. (2013) Novel Lager Beer Clarification and Stabilisation Process Using Ceramic Tubular Microfiltration Membranes. In *Proc.s of the 18th Workshop on the Developments on the Italian PhD Research on Food Science Technology and Biotechnology*, Università degli Studi di Padova ed Udine, Conegliano Veneto, 25-27 September, 2013, 45-50 (ISBN 978-88-97385-68-4).
5. Cimini A, Marconi O, Perretti G, Moresi M. (2013) Novel Procedure for Lager Beer Clarification and Stabilization using Sequential Enzymatic, Centrifugal, Regenerable PVPP and Crossflow Microfiltration Processing. *FABT*, DOI 10.1007/s11947-014-1306-x
6. Cimini A, Moresi M. (2013) Beer clarification using ceramic tubular membranes. *FABT*, *submitted*
7. Cimini A, Moresi M, Pallottino F, Menesatti M (2014) Development of an improved Rudin-based beer foam head retention meter using time tracking or image analysis. Poster P.24 presented at the *11th International Trends in Brewing*, Ghent, Belgium, 13-17 April, 2014.
8. Cimini A, Moresi M (2014) Beer clarification and stabilization using centrifugal, regenerable PVPP and crossflow microfiltration processing Poster P.34 presented at the *11th International Trends in Brewing*, Ghent, Belgium, 13-17 April, 2014.
9. Cimini A, Moresi M (2014) Carbon Footprint of Lager Beer. Poster P.63 presented at the *11th International Trends in Brewing*, Ghent, Belgium, 13-17 April, 2014.

