UNIVERSIDAD DE EXTREMADURA

WATERINNOVA - Development of innovative processes for the treatment and valorization of wastewater from food manufacturing processes with fertilizer production, CO_2 mitigation and food production

WATERINNOVA - Desarrollo de procesos innovadores para el tratamiento y valorización de aguas residuales de la industria agroalimentaria con producción de fertilizantes, mitigación de CO_2 y producción de alimentos

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PROGRAMA DE DOCTORADO EN MODELIZACIÓN Y EXPERIMENTACIÓN EN CIENCIA Y TECNOLOGÍA POR LA UNIVERSIDAD DE EXTREMADURA

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Resumen

Las industrias elaboradoras de queso y vino generan efluentes con una elevada carga orgánica que requiere una adecuada manipulación y almacenaje posterior. En este sentido, las estrategias enfocadas en el tratamiento de aguas residuales avanzan hacia un proceso de economía circular a través de la valorización de estos efluentes. En este trabajo se ha aplicado la patente europea EP 3447030B1 de precipitación ácida química, con agitación secuencial decreciente, en tres etapas de agitación de 1 minuto cada una. Con este tratamiento se disminuyó en un 76% de la demanda química de oxígeno disuelto. El precipitado obtenido representa hasta un 10% del volumen, sedimenta fácilmente y contiene~~n~~ alrededor de un 90% en materia seca. Estos sólidos son ricos en materia orgánica y nutrientes, lo que les abre la posibilidad de aprovechamiento agrícola. Al aplicar esta tecnología en el tratamiento de aguas residuales de queserías, se añadieron coagulantes (75 g L^{-1} de $CaCO_3$ + 1 g L^{-1} de $FeSO_4$), aumentando la eficiencia en la eliminación de parámetros indicativos de la contaminación (97% para aceites y grasas y 92%~~-~~ para fósforo total (P_{total}), 99% turbidez, 59% fenoles y 55% de la demanda química de oxígeno (DQO)). La aplicación de $Ca(OH)_2$ en el tratamiento de las aguas residuales de la industria vitivinícola permitió la eliminación del 99,9% de los fenoles. Este trabajo también se ensayó, por primera vez, la eficacia de la tecnología de precipitación química inmediata con neutralización por reacciones de captura espontánea de CO_2 atmosférico en efluentes de la industria de elaboración del vino. Se demostró que el uso de cal hidratada, 25 g L^{-1} permitió obtener reducciones del 22% de DQO, 87% de P_{total}, más del 97% de metales (zinc, cobre, hierro y manganeso), 99.9% del total de fenoles y 100% de microorganismos (*Echerichia Coli*, Coliformes Fecales y Enterococos), resultando en la neutralización espontánea del efluente en 13 días. El sobrenadante y el precipitado se utilizaron como solución nutritiva (por dilución 1:4) y corrector organomineral (dosis 1,2% p/p), respectivamente. Estos no mostraron toxicidad para los bioindicadores *Thmanocephalus platyurus, Vibrio fisheri y Pseudokirchneriella subcapitata*). La solución nutritiva y el corrector organomineral se utilizaron en la producción de Pak Choi rojo (*Brassica rapa* L. var. *chinensis* (L.) Kitam), con tasas de crecimiento superiores al control.

Palabras clave: aguas residuales de industrias agroalimentarias; precipitación química; neutralización por secuestro de CO_2 atmosférico; economía circular; reutilización agrícola; Pak Choi rojo; producción de alimentos

Resumen Extendido

En una época donde la escasez de recursos hídricos es uno de los principales problemas ambientales globales, la economía circular se presenta como una estrategia de futuro tanto desde el punto de vista económico como medioambiental. La preocupación por el medioambiente se ha generalizado y la Organización de las Naciones Unidas ha definido como objetivos de desarrollo sostenible para 2030 el reciclaje y la reutilización del agua. Esta es la mejor manera de aprovechar este recurso esencial para la vida, valorizándolo y transformándolo en un recurso adicional para la empresas de gestión aguas residuales. La industria agroalimentaria, entre ellas, la industria quesera y la vitivinícola están normalmente dispersas en el espacio, son de tamaño pequeño - mediano, y generan efluentes que, por su elevada carga orgánica y su carácter recalcitrante hacia los biotratamientos, exigen, por imperativo legal, la aplicación de sucesivos tratamientos, de elevado coste, y que precisan de mano de obra especializada. Estas exigencias tornan este tratamiento inasequible, tanto desde el punto de vista técnico como financiero impidiendo la transición hacia una economía circular.

En este sentido, los **objetivos** de este trabajo fueron (i) desarrollar tecnologías para el tratamiento de aguas residuales de la industria agroalimentaria sencillas de implementar, económicas y fáciles de aplicar *in situ*,; (ii) evaluar el impacto ambiental asociado a los sobrenadantes y precipitados obtenidos; (iii) y analizar su potencial de reutilización para la producción de alimentos.

Para lograr estos objetivos se desarrolló y patentó una tecnología de precipitación química en medio ácido (pH = 3) (patente europea EP 3447030B1) utilizando ácidos fuertes (HCl, H_2SO_4 o HNO_3) como reactivos y aplicando agitación secuencial decreciente en tres etapas de un minuto cada una: agitación a alta velocidad (1200 – 1400 rpm) d, velocidad media (700 – 800 rpm) y velocidad lenta (300 – 400 rpm). Esta tecnología permitió alcanzar una reducción significativa de materia orgánica disuelta, monitorizada a través de la evaluación de la demanda química de oxígeno (DQO disuelta). En el caso de las aguas residuales de la industria vitivinícola, se eliminó el 76% de la DQO disuelta y el 72% de la DQO total. Estas eficiencias de eliminación también permitieron pasar de una concentración de 46% de DQO disuelta en el agua cruda a solo 16% en el agua de bodega tratada. La tecnología también permitió alcanzar reducciones máximas de turbidez en torno al 90%, 60% para fenoles y 70% para el P_{total}.

Los precipitados obtenidos son fácilmente sedimentables y representan un volumen entre el 1 y el 10% del volumen de agua tratada. Los precipitados contienen alrededor del 90% de materia seca, y además son ricos en materia orgánica y nutrientes como calcio, magnesio, fósforo, cloruro, sodio y potasio, lo que les otorga la capacidad de reutilizarlos como fertilizante en agricultura.

Con el objetivo de aumentar el potencial de tratamiento de la tecnología de precipitación química con agitación secuencial decreciente para aguas residuales de queserías, se ensayaron diferentes coagulantes, en lugar de ácidos fuertes, implementando dos modos de aplicación:

1. Coagulantes individuales: $FeSO_4$, $FeCl_3$, $CaCO_3$, $Ca(OH)_2$, $Mg(OH)_2$ y NaOH.

El pH de trabajo que permitió alcanzar las mejores eficiencias se situó en el entorno de 5.0 para $FeSO_4$, 5,5 para $FeCl_3$, 6,5 para $CaCO_3$, 7.0 para $Mg(OH)_2$ y NaOH y 11.0 para $Ca(OH)_2$. Los coagulantes $Mg(OH)_2$ y $FeSO_4$, usados individualmente, no resultaron adecuados para este tipo de efluentes ya que su capacidad de eliminación de materia orgánica fue prácticamente nula. Se verificó que el coagulante que presentó mejor eficiencia global fue el CaCO3 (75 g L^{-1}), a un pH de trabajo de 7.0 (con la ventaja de no necesitar corrección para la reutilización del efluente tratado). Con este tratamiento se alcanzaron eficiencias de eliminación de turbidez (98%), P_{total} (94%) y aceites y grasas (>99%). También cabe señalar que el coagulante $Ca(OH)_2$ demostró ser el más efectivo en la remoción de fenoles, 92%.

2. Coagulantes combinados:

Las mejores concentraciones de coagulantes individuales encontradas se usaron en la combinación de coagulantes.

Secuencia 1 - $CaCO_3$, 75 g L^{-1} + $FeCl_3$ 1g L^{-1}

Secuencia 2 - $CaCO_3$ a 75 g L^{-1} + $FeSO_4$ 1 g L^{-1}

Secuencia 3 - $CaCO_3$, 75 g L^{-1} +$Ca(OH)_2$ 0,6 g L^{-1}

El pH de trabajo se situó siempre en zona neutra, 7,1, 7,5 y 7,7 para las secuencias 1, 2 y 3 respectivamente. Esta combinación de coagulantes permitió lograr un incremento en la eliminación de todos los parámetros monitorizados, siendo la combinación 2 la que permitió alcanzar reducciones del 98% para aceites y grasas y P_{total}, 99% para turbidez y 60% para fenoles. La reducción de materia orgánica alcanzó el 56%.

Existen en la literatura algunos estudios sobre la aplicación de la tecnología de precipitación química inmediata con uso de $Ca(OH)_2$, en medio básico, a efluentes de queserías reportando altas eficiencias de extracción, la neutralización del efluente mediante reacciones espontáneas con el CO_2 atmosférico así como la desinfección simultánea del efluente tratado. Sin embargo, no se hace referencia a la aplicación de esta tecnología a los efluentes de bodega. Así, además del desarrollo de pruebas con $Ca(OH)_2$, la tecnología también fue probada con las siguientes sustancias: óxido de calcio - Cal viva (CaO); cal hidratada – cal apagada [$Ca(OH)_2$] e hidróxido de calcio [$Ca(OH)_2$].

De estas tres sustancias, la cal hidratada en dosis de 25 g L^{-1} presentó los mejores resultados, reduciendo el 22% de la materia orgánica (DQO), el 87% del P_{total}, más del 97% de los metales (zinc, cobre, hierro y manganeso), 99,9% de fenoles totales y 100% de microorganismos (*Echerichia Coli*, Coliformes Fecales y Enterococos). A partir de este tratamiento se generó un precipitado con un 10% de materia orgánica y altos niveles de macronutrientes. El sobrenadante se utilizó para preparar una solución nutritiva (dilución 1:4) y el precipitado un corrector organomineral (dosis de 1,2% p/p) . Ambos residuos fueronsometidos a los bioindicadores: *Thmanocephalus platyurus* (bioensayo de mortalidad a las 24h.), *Vibrio fisheri* (inhibición de la luminiscencia a los 30 min.) y *Pseudokirchneriella subcapitata* (inhibición del crecimiento a las 72 horas), encuadrándose ambos en la clase 1- Atóxicos, compatibles con la reutilización agrícola, para la producción de alimentos.

Finalmente, se monitorizó el desarrollo de plantas de Pak Choi rojo (*Brassica rapa* L. var. *chinensis* (L.) Kitam), producidas a partir de estos valiosos subproductos, utilizados en 3 condiciones diferentes:

1- Control: Suelo ácido + riego con agua subterránea;

2- Fertirrigación: suelo ácido + riego con solución nutritiva;

3- Suelo ácido corregido con abono organomineral (precipitado) + riego con aguas subterráneas.

Las plantas de las condiciones 2 y 3 mostraron mayores tasas de crecimiento que el control.

Este estudio permitió desarrollar tratamientos innovadores, económicos y sencillos de utilizar que conducen a la producción de efluentes y precipitados con capacidad de reutilización, fomentando la economía circular y el principio de residuo cero.

Resumo

As indústrias de fabrico de queijo e vinho produzem efluentes de elevada carga orgânica e necessitam transitar para uma economia circular através da valorização dos seus efluentes. Neste trabalho, foi desenvolvida a patente europeia EP 3447030B1 de precipitação química ácida com agitação sequencial decrescente em três etapas de agitação de 1 minuto cada, alcançando-se 76%, de remoção de carência química de oxigénio dissolvido. Os precipitados representam até 10% em volume, sedimentam facilmente e contêm cerca de 90% de matéria seca. São ricos em matéria orgânica e nutrientes conferindo-lhe aptidões agrícolas. Ao aplicar esta tecnologia no tratamento de água residual de queijaria com a mistura de coagulantes, 75 g L^{-1} de $CaCO_3$ + 1 g L^{-1} $FeSO_4$, alcançaram-se incrementos significativos de eficiências de remoção (97% para óleos e gorduras e 92% fósforo total (P_{total}), 99 % para a turvação, 59% para os fenóis e 55% carência química de oxigénio, CQO). O uso de $Ca(OH)_2$ no tratamento de água residual de adega permitiu remover 99,9% de fenóis. Neste trabalho testou-se também a eficácia da tecnologia de precipitação química imediata com neutralização por reações de sequestro espontâneo do CO_2 atmosférico em efluentes de adega, pela primeira vez. Demonstrou-se que o uso de cal hidratada, 25 g L^{-1} permitiu obter remoções de 22% de CQO, 87% de P_{total}, mais de 97% de metais (zinco, cobre, ferro e manganês), 99,9% de fenóis totais e 100% de microrganismos (*Echerichia Coli*, Coliformes Fecais e Enterecocos), obtendo-se a neutralização espontânea do efluente em 13 dias. O sobrenadante e precipitado foram utilizados como solução nutritiva (por diluição de 1:4) e corretivo organomineral (dosagem 1,2% p/p). Estes não apresentaram toxicidade pela aplicação dos bioindicadores *Thmanocephalus platyurus, Vibrio fisheri e Pseudokirchneriella subcapitata*). A solução nutritiva e corretivo organominera foram usados na produção de Red Pak Choi (*Brassica rapa* L. var. *chinensis* (L.) Kitam), tendo-se obtido taxas de crescimento superiores ao controlo.

Palavras chave: água residual de indústrias agroalimentares; precipitação química; neutralização por sequestro do CO_2 atmosférico; economia circular; reutilização agrícola; Red Pack Choi; produção de alimentos

Resumo Alargado

Numa era em que a escassez de recursos hídricos é um dos principais problemas ambientais globais, a economia circular é uma premissa fundamental. A preocupação é global e a Organização das Nações Unidas definiu como objetivos para o desenvolvimento sustentável até 2030 a reciclagem e reutilização da água. Esta é a melhor forma de aproveitar este recurso essencial à vida e de o valorizar transformando-o num recurso adicional para os produtores de águas residuais. Porém, a indústria agroalimentar, nomeadamente as indústrias de fabrico de queijo e vinho são, normalmente, dispersas, de pequenas e médias dimensões e produzem efluentes que devido á sua elevada carga orgânica e à sua natureza recalcitrante exigem, por força da lei, a aplicação de sequências de tratamentos dispendiosos e com necessidade de recurso a mão de obra especializada. Tal exigência torna-se incomportável quer sob o ponto de vista técnico quer financeiro e impede-as de transitar para uma economia circular.

Neste sentido, os **objetivos** deste trabalho foram (i) desenvolver tecnologias de tratamento de águas residuais da indústria agroalimentar simples, económicas e de fácil implementação *in loco* em efluentes de queijaria e adega; (ii) avaliar o impacto ambiental associado aos sobrenadantes e precipitados obtidos; (iii) analisar o seu potencial de reaproveitamento para a produção de alimentos.

Para atingir esses objetivos foi desenvolvida e patenteada (patente europeia EP 3447030B1) a tecnologia de precipitação química em meio ácido (pH =3) utilizando como reagentes os ácidos fortes (HCl, H_2SO_4 ou HNO_3) e aplicando uma agitação sequencial decrescente em três etapas de agitação com velocidade forte (1200 – 1400 rpm) durante 1 minuto, média, velocidade média (700 – 800 rpm), durante 1 minuto e velocidade lenta (300 – 400 rpm), durante um minuto. Esta tecnologia permitiu alcançar um efeito vantajoso inesperado de redução significativa da matéria orgânica dissolvida, monitorizada pela avaliação da carência química de oxigénio (CQO dissolvida). Para o caso da água residual de adega verificou-se a remoção de 76% de CQO dissolvido e 72 % CQO total. Estas eficiências de remoção permitiram também passar de um teor de 46 % de CQO dissolvido na água bruta para apenas 16% na água tratada de adega. A tecnologia permite ainda alcançar remoções máximas de turvação na ordem dos 90%, 60% para fenóis e 70% para e P_{total}.

Os precipitados obtidos são facilmente sedimentáveis e representam um volume entre 1 e 10% do volume de água tratada. Os precipitados contêm cerca de 90% de matéria seca, sendo também ricos em matéria orgânica e em nutrientes como cálcio, magnésio, fósforo, cloreto, sódio e potássio, o que lhes confere aptidões de reutilização como fertilizante na agricultura.

Com o objetivo de incrementar as potencialidades de tratamento da tecnologia precipitação química de agitação sequencial decrescente, foram testados diferentes coagulantes, em vez dos ácidos fortes, para a água residual de queijaria em dois modos de aplicação:

1. Coagulantes individuais: $FeSO_4$, $FeCl_3$, $CaCO_3$, $Ca(OH)_2$, $Mg(OH)_2$ e NaOH.

Os pH de trabalho que melhores eficiências permitiram alcançar variou de 5 para o $FeSO_4$, 5,5 para o $FeCl_3$, 6,5 para o $CaCO_3$, 7 para $Mg(OH)_2$ e NaOH e 11 para o $Ca(OH)_2$. Os coagulantes $Mg(OH)_2$ e o $FeSO_4$, usados de forma individual, não se mostraram adequados para este tipo de efluentes por apresentarem eficácia praticamente nula na remoção de matéria orgânica. Verificou-se que o coagulante que apresentou globalmente melhor eficiência foi o uso de 75g de $CaCO_3$ por litro de água tratada, a um pH de trabalho de 7 (com a vantagem de não necessitar de correção para a reutilização do efluente tratado), principalmente na remoção de turvação (98%), P_{total} (94%) e óleos e gorduras (>99%.). De realçar ainda que o coagulante $Ca(OH)_2$ mostrou ser o mais eficaz na remoção de fenóis, 92%.

2. Coagulantes combinados:

Foram utilizadas na combinação de coagulantes as melhores concentrações de coagulantes individuais encontradas:

Sequência 1 - $CaCO_3$, 75 g L^{-1} + $FeCl_3$ 1g L^{-1}

Sequência 2 - $CaCO_3$ a 75 g L^{-1} + $FeSO_4$ 1 g L^{-1}

Sequência 3 - $CaCO_3$, 75 g L^{-1} +$Ca(OH)_2$ 0,6 g L^{-1}

O pH de trabalho situou-se sempre numa zona neutra, 7,1, 7,5 e 7,71 para as sequência 1, 2 e 3 respetivamente. Esta combinação de coagulantes permitiu alcançar incremento em todos os parâmetros monitorizados, tendo a combinação 2 permitido alcançar remoções de 98% para óleos e gorduras e P_{total}, 99 % para a turvação e 60% para os fenóis. A remoção de matéria orgânica também atingiu os 56%.

Existem na literatura alguns estudos de aplicação da tecnologia de precipitação química imediata com o uso de $Ca(OH)_2$, em meio básico, a efluentes de queijaria com a obtenção de elevadas eficiências de remoção, a neutralização do efluente através de reações espontâneas com o CO_2 atmosférico bem como, a desinfeção simultânea do efluente tratado. Porém não se encontra nenhuma referência à aplicação desta tecnologia a efluentes de adega. Assim, para além do desenvolvimento de ensaios utilizando o $Ca(OH)_2$ também se testou a tecnologia com as substâncias: óxido de cálcio - Quicklime (CaO); cal hidratada – Slaked lime [$Ca(OH)_2$] e hidróxido de cálcio [$Ca(OH)_2$].

Destes três substâncias, a cal hidratada à dosagem de 25 g L^{-1} foi a que apresentou melhores resultados, removendo 22% de matéria orgânica (CQO), 87% de P_{total}, mais de 97% de metais (zinco, cobre, ferro e manganês), 99,9% de fenóis totais e 100% de microrganismos (*Echerichia Coli*, Coliformes Fecais e Enterecocos). Proveniente deste tratamento foi ainda gerado um precipitado com 10% de matéria orgânica e elevados teores de macronutrientes. O sobrenadante foi utilizado para preparar uma solução nutritiva (diluição 1:4) e o precipitado um corretivo organomineral (dosagem de 1,2% p/p) que ao ser submetida aos bioindicadores: *Thmanocephalus platyurus* (bioensaio de mortalidade após 24h.), *Vibrio fisheri* (inibição da luminescência após 30 min.) e *Pseudokirchneriella subcapitata* (inibição do crescimento após 72h)*, ambos* apresentavam classe 1- Não tóxicos, compatíveis com reutilização agrícola, para produção de alimentos.

Por fim, foi acompanhado o desenvolvimento das plantas Red Pak Choi (*Brassica rapa* L. var. *chinensis* (L.) Kitam), produzidas a partir destes subprodutos valorizáveis, utilizados em 3 condições distintas:

1- Controlo: Solo ácido + rega com água subterrânea;
2- Fertirrega: solo ácido + rega com solução nutritiva;
3- Solo ácido corrigido com fertilizante organomineral (precipitado) + rega com água subterrânea.

As plantas das condições 2 e 3, apresentaram taxas de crescimento superiores ao controlo.

Este estudo permitiu o desenvolvimento de tratamentos inovadores, económicos e de simples aplicação que levam à produção de efluentes e precipitados com capacidade de reutilização, fomentando a economia circular e o princípio do zero desperdício.

Índice

CAPÍTULO I - INTRODUÇÃO

A escassez hídrica tem sido um dos problemas com mais expressão nos últimos anos à escala mundial, podendo trazer uma série de consequências nefastas, tais como, extinção de ecossistemas aquáticos; aumento do risco de doenças relacionadas com a água, tal como a cólera e diarreias; quebras de produção agrícola; conflitos entre países que partilhem recursos hídricos (Ogenyi, 2023; Sarkar *e* Devi 2023).

Estima-se que, a nível mundial, cerca de 56% das área agrícolas cultivadas em regime de regadio estejam localizadas em locais cujo stress hídrico é alto ou extremamente alto (Sapkota, 2019). E é precisamente o setor agrícola que consome cerca de 70-80% da água potável disponível (Prazeres *et al.,* 2017). Devido a essa escassez hídrica, atividades agroindustriais como a produção de vinho, que necessitam de água para rega das vinhas, podem estar em causa num cenário de escassez hídrica (Costa *et al.,* 2020; Bolzonella *et al.,* 2019).

Anualmente, são lançados no meio ambiente, cerca de 2212 km^3 de água/ano, de efluentes urbanos e industriais e águas de drenagem agrícola que podem ser a solução para as necessidades hídricas agrícolas, pois anualmente, a nível mundial, são necessários cerca de 1.716 km^3 de água para rega (Smol *et al.,* 2020). Desta forma, é essencial adotar estratégias sustentáveis de gestão de recursos hídricos de forma a fomentar a economia circular, nomeadamente o tratamento e reutilização de águas residuais (Rebelo *et al.,* 2020; Sapkota, 2019).

A reutilização de água residual tratada, já é encarada como uma alternativa válida e uma nova origem de água para usos múltiplos, nomeadamente a rega para fins agrícolas ou espaços verdes (como por exemplo campos de golf) e usos urbanos restritos (APA I.P., 2019). Para garantir a segurança na reutilização das águas residuais tratadas, a União Europeia publicou o Regulamento EU 2020/741 do Parlamento Europeu e do Conselho, de 25 de Maio de 2020, de caráter obrigatório e aplicável em todos os Estados-Membro a partir de 26 de Junho de 2023, que visa estabelecer os padrões mínimos de qualidade da água, bem como as respetivas disposições de monitorização e gestão de riscos no âmbito da gestão integrada da água. Em Portugal, está prevista e regulamentada a reutilização de águas residuais bem como o estabelecimento do regime jurídico de produção de água para reutilização, obtida a partir do tratamento de águas residuais, através do Decreto-Lei n.º 119/2019 de 21 de agosto, alterado pelo Decreto-Lei n.º 11/2023 de 10 de Fevereiro, ambos já contemplam grande parte das disposições instituídas no Regulamento EU 2020/741 do Parlamento Europeu e do Conselho.

A Organização das Nações Unidas (ONU) definiu na Agenda 2030 de Desenvolvimento Sustentável, 17 objetivos como forma de atenuar desigualdades e promover as práticas saudáveis e sustentáveis à escala global. Dentro desses objetivos, está a temática da água residual (incluída no objetivo 6), que visa promover o tratamento das águas residuais, a reciclagem e reutilização das mesmas como forma de minimizar danos ambientais e escassez de recursos. É também ao nível industrial que estes objetivos se devem implementar, promovendo a sustentabilidade (objetivo 9), através de uma modernização e reabilitação das indústrias, fomentando o uso de recursos, que visem minimizar os danos ambientais, fomentando assim uma economia circular.

Na Europa, as empresas da indústria agroalimentar, são consideradas as principais indústrias transformadoras, gerando um volume total de negócios de 14% (Campos-Mañas *et al.,* 2019).

Nos países Mediterrânicos, as agroindústrias de produção de vinho, azeite, café, frutas, lacticínios, entre outros, estão entre as principais atividades económicas, mas também das que representam um maior impacto ambiental ao nível do consumo de água. Esta é utilizada em grandes quantidades durante todo o processo produtivo, desde as lavagens da matéria-prima, equipamentos, superfícies e materiais, gerando produção de efluentes líquidos com elevada carga poluente, que, se descarregados sem qualquer tipo de tratamento, ou tratamento adequado, representam significativos impactos no ambiente (Amor *et al.* 2019, Smol *et al.* 2019, Campos-Mañas *et al*., 2019)

Também em Portugal, as agroindústrias do queijo e do vinho, têm forte representatividade, e como consequência, são gerados grandes volume de águas residuais.

Os efluentes gerados nas agroindústrias do queijo e do vinho apresentam-se com elevada carga orgânica, pH baixo e elevados teores de gordura, proteínas, compostos aromáticos, compostos fenólicos e sólidos totais (Caleja *et al.,* 2023; Rajagopal *et al.,* 2013). Estes efluentes podem gerar impactos ambientais e para a saúde pública graves, tais como, a poluição de massas de água superficiais e subterrâneas, formação de espumas, diminuição do oxigénio disponível, eutrofização, salinização e alteração da estrutura do solo, proliferação de vetores (Prazeres *et al.,* 2017).

Os tratamentos já existentes e eficazes para fazer face à elevada carga orgânica associada a este tipo de efluentes, necessitam de pré-tratamentos (Mader *et al.,* 2022), e tecnologias subsequentes tais como: ozonização (Lucas *et al.,* 2009), foto-Fenton (Jorge *et al.,* 2023; Mosteo *et al.,* 2006; Ormad *et al.,* 2006), sistemas de filtração e osmose inversa (Ioannou et al. 2013; Ferrarini *et al.,* 2001), zonas húmidas artificiais, (Aguilar *et al.,* 2022; Serrano *et al.,* 2011), digestão anaeróbia (Wu *et al.,* 2023; Keyser *et al*., 2003; Kalyuzhnyi *et al.,* 2001), entre outras. Porém, muitas das indústrias de fabrico de queijo e vinho são de pequenas e médias dimensões e não possuem mão de obra especializada, bem como capacidade financeira que realize *in loco* o tratamento dos seus efluentes e, por isso, são obrigadas a transportar os mesmos para unidades de tratamento distantes. Neste sentido, o objetivo deste trabalho foi o de (i) desenvolver tecnologias de tratamento de águas residuais da indústria agroalimentar simples, económicas e de fácil implementação *in loco* em efluentes de queijaria e adega; (ii) avaliar o impacto ambiental que essas águas têm; (iii) analisar o potencial de reaproveitamento dos sobrenadantes e precipitados para a produção de alimentos.

1.1. A importância e a problemática das agroindústrias

O sector agroindustrial define-se como um segmento de atividades amplas, que se iniciam no pós-colheita, com preservação e transformação de produtos agrícolas (Simão *et al*., 2022). Estima-se que 20% da superfície terrestre esteja ocupada por agroindústrias (Gaspar *et al*., 2021). Com o aumento estimado de 30% da população mundial até ao ano de 2050, existirá um inevitável aumento da procura de alimentos, e um crescente aumento das agroindústrias (Gaspar *et al*., 2021).

O setor agroindustrial é um pilar económico considerável em Portugal e na União Europeia. O sector da transformação (sector secundário), representou, no ano de 2022, um quarto da atividade económica em Portugal, como representa a figura 1. As indústrias do ramo alimentar foram em 2021 a principal atividade industrial em Portugal (INE, 2021 – Estatísticas Agrícolas). Estima-se que, 30% da população portuguesa está concentrada nos meios rurais, onde também existe uma maior concentração das agroindústrias (Gaspar *et al*., 2021).

É também no meio rural que existe uma forte cultura e tradição gastronómica, nomeadamente na região do Alentejo, as indústrias do vinho e do queijo têm grande representatividade económica e social. A indústria do vinho teve uma produção de 7,2 milhões de hectolitros produzidos no ano de 2021, representando o valor mais elevados dos últimos 5 anos. Já a indústria dos lacticínios contabilizou 2,029 milhões de litros de leite produzidos no ano de 2021, com um aumento de produção dos produtos transformados, nomeadamente o queijo de vaca (INE, 2021). Estas agroindústrias de elevada representação em Portugal estão principalmente localizadas no interior do país, nomeadamente no Alentejo (Gabinete de estratégia e estudos, 2023), com forte produção de matéria-prima e consequentemente de resíduos a elas associados.

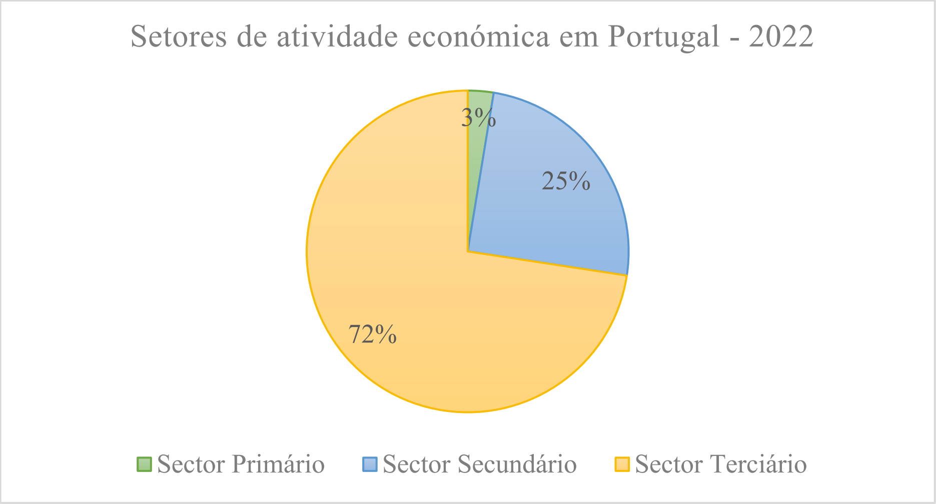


Figura 1 – Sectores de atividade económica em Portugal. Adaptado de: Pordata, 2023

Uma das maiores problemáticas associada às agroindústrias é a correta gestão dos efluentes gerados, uma vez que se esses efluentes forem descarregados no ambiente, sem tratamento ou pré-tratamento, podem ter impactos significativos (Smol *et al.,* 2020).

Tal como referido na Introdução, pode dizer-se genericamente que os efluentes das agroindústrias apresentam elevados teores de matéria orgânica (carência química (CQO) e bioquímica de oxigénio (CBO_5)) como ácidos, compostos aromáticos, proteínas, sólidos totais, óleos e gorduras (Rajagopal *et al*., 2013). Para além disso, apresentam também elevadas concentrações de nutrientes, nomeadamente, azoto e fósforo, elementos que, se não forem tratados e eliminados previamente, tornam-se num dos principais fatores para a diminuição de oxigénio nas massas de água levando a processos de eutrofização e consequentemente destruição da fauna e flora local (Prazeres *et al.,* 2017).

A melhor forma de colmatar impactos nefastos das descargas indevidas destes efluentes será a de fomentar a economia circular das indústria agroalimentares, tornando-os um produto e não num resíduo com potencialidades económicas. Por exemplo, a sua valorização para fins agrícolas permite reduzir a aquisição de nutrientes industriais, reduzir a pegada hídrica e os custos associados à aquisição de água (doce) para rega. Para além disso, permite promover o aumento da produtividade da planta, uma vez que se trata de água com mais nutrientes, fomentando ainda o incentivo à sustentabilidade empresarial, utilizando os recursos hídricos de forma consciente e eficiente (Guerra-Rodríguez *et al.,* 2020).

Para além dos efluentes líquidos gerados nas agroindústrias existe também a problemática dos resíduos sólidos decorrentes dos processos de tratamento dos efluentes. Na União Europeia, no ano de 2015, foram produzidos mais de 10 milhões de toneladas de lamas secas, sendo necessária a sua reutilização/ recuperação, uma vez que se trata de um composto rico em nutrientes, tais como, azoto e fósforo, bem como proteínas e enzinas (Guerra-Rodríguez *et al.,* 2020).

1.1.1. Indústria do queijo

Segundo a FAO (Food and Agriculture Organization, 2021), o queijo é um produto curado ou não, de textura mole, semidura, dura ou extra dura, obtido através de coagulação total ou parcial da proteína do leite, e/ou coagulado da proteína do leite e/ou derivados. É composto por vitaminas, minerais, hidratos de carbono, lípidos, cálcio e fósforo e podem ser produzidos através de leite proveniente de vaca, cabra ou ovelha (Prazeres *et al*., 2013a).

Estima-se que mundialmente são produzidas cerca de $2x10^7$ toneladas de queijo/ano, sendo os principais países produtores os Estados Unidos da América, Alemanha e França (Fox *e* McSweeney, 2017). Em Portugal, no ano de 2022, foram produzidos quase 67000 toneladas só de queijo de vaca (INE, I.P., 2023).

A indústria do queijo em Portugal é muito diversificada e com diversos dos seus produtos destacados com a Denominação de Origem Protegida (DOP), que define a qualidade e características desse produto ao meio geográfico de que é originário. Alguns dos mais conhecidos em Portugal são, queijo Serra da Estrela DOP, queijo Azeitão DOP, queijo Évora DOP, queijo São Jorge DOP e queijo Serpa DOP (Direção-Geral de Agricultura e Desenvolvimento Rural, 2023). O queijo Serpa DOP caracteriza-se por ser um queijo curado, de pasta semimole e amanteigado, apenas podendo ser produzido em área geográfica delimitada, com indicado na figura 2.

Fonte:
- Carta Administrativa Oficial de Portugal da DGT, versão 2013

DGADR
- DQRG
Abril de 2016

Figura 2 – Área geográfica de produção do queijo Serpa DOP. Fonte: Direção-Geral de Agricultura e Desenvolvimento Rural, 2023

A Indústria de Laticínios gera o maior volume de efluentes na Europa, podendo gerar em média cerca de 500 m^3 de resíduos diariamente (Rivas *et al.*, 2010, Demirel *et al.*, 2005) e, ainda, cerca de 0,2 – 10 L de efluente por litro de leite processado (Shete *e* Shinkar, 2013). Para além disso, necessita de elevados consumos de água, podendo-se utilizar volumes de água entre 4 a 15 L de água por cada litro de leite processado para a produção de queijo, gelado, manteiga ou iogurte (Pacheco *et al.* 2023).

A produção de queijo é realizada em várias etapas, algumas são comuns a todos os tipos de queijo, outras são especificas para cada tipologia (requeijão, almece, queijo fresco, seco e curado). Na figura 3 estão descritas todas as etapas desse processo. Durante o mesmo existem entradas de água e componentes que se adicionam e que são essenciais à produção de queijo, mas também existem saídas de efluentes, provenientes de perdas do processo de fabrico de queijo (ricas em soro de queijo), do processo de fabrico de requeijão (ricas em segundo soro de queijo), refrigeração, lavagens, limpeza e desinfeção de pavimentos, superfícies e materiais.

Esse efluente apresenta pH ácido de 3.92 a 6.5, CQO e CBO_5: 60 a 102 g L^{-1} e 27 a 60 g L^{-1}, respetivamente, bem como óleos e gorduras ≈ 9 g L^{-1}, turvação de 536 a 9900 NTU, sais minerais 0,46 a 10%, sólidos totais (ST) de 3 a 70 g kg^{-1}, sólidos suspensos totais (SST) de 1 a 22 g kg^{-1}, azoto amoniacal de 0,1 a 0,3 g $N\text{-}NH_4^+$ kg^{-1} , P_{total} de 0,1 a 4,9 g P L^{-1} (Pacheco *et al.*, 2023, Carvalho *et al.,* 2013). Estes efluentes, se descarregado sem tratamento terão um grave impacto no ambiente e na saúde pública, nomeadamente no consumo rápido de oxigénio nas massas de água, criação de condições anaeróbias, eutrofização e espumas, colocando em risco fauna e flora local, para além da proliferação de vetores e veiculação de doenças (Prazeres *et al.* ,2013a). Um dos principais responsáveis por estes efeitos é o soro do queijo, que está baste presente no efluente gerado, caracterizado como um leite sem gordura, ou caseína, apresenta uma coloração amarelo-esverdeado (devido à presença de vitamina B2 ou riboflavina), que eleva os teores de CQO presentes no efluente (Pacheco *et al.,* 2023; Prazeres, 2013; Carvalho *et al.*, 2013). Anualmente são gerados a nível mundial 11 milhões de toneladas de soro de leite, 4,5 são só na União Europeia, esta produção é tão elevada, uma vez que, por 1 kg de queijo produzido, são gerados 9 L de soro (Lucakova *et al.*, 2022). É por isso considerado um efluente de difícil tratamento, bastante poluente, tóxico e nefasto para o ambiente, estimando ser 60 a 80 vezes mais poluente que os efluentes domésticos (Guliyev *et al.,* 2022; Shete *e* Shinkan, 2013).

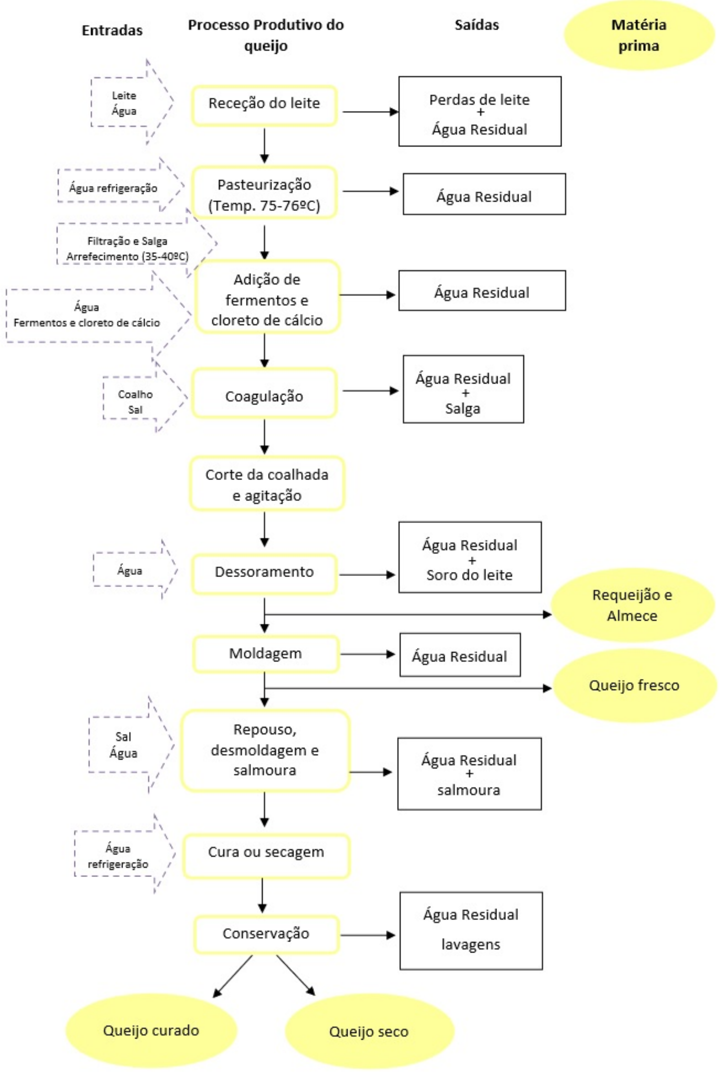


Figura 3 – Processo produtivo do fabrico de queijo. Adaptado de: Gonçalves 2019 e Prazeres 2013.

1.1.2. Indústria do vinho

A indústria do vinho tem uma elevada representação económica e social na Europa, sendo o continente que produz mais de metade do vinho no mundo, mais concretamente, 61% da produção mundial total (Bolzonella *et al,*. 2019). Dessa produção, Portugal é responsável pela produção de 7,4 milhões de hL, no ano de 2021, fazendo com que seja o 11º país com maior produção mundial de vinho, onde a região do Alentejo é uma das principais geradoras dessa economia e produção (IVV IP, 2022).

Em todo o mundo estão catalogadas mais de 4000 variedades de uvas, sendo Portugal o 2º país com maior expressão de castas indígenas e exclusivas (Vinhos do Alentejo, 2023). O Regulamento (CE) n.º 1234/2007, do Conselho, de 22 de outubro, alterado pelo Regulamento (CE) n.º 491/2009, define a classificação de castas de uvas de vinho, bem como a regras de produção no setor vinícola. No Alentejo, as principais castas de uvas de vinho são: Antão Vaz, Arinto, Roupeiro, Alfrocheiro, Alicante Bouschet, Aragonez, Cabernet Sauvignon, Castelão, Syrah, Touriga Nacional, Tinta Grossa e Trincadeira (Vinhos do Alentejo, 2023).

Os vinhos de Denominação de Origem do Alentejo apresentam 10 sub-regiões, Borba, Évora, Granja-Amareleja, Moura, Portalegre, Redondo, Reguengos e Vidigueira.

O processo de vinificação está dividido por diversas etapas, algo distintas, se se tratar do processo de vinificação de vinhos brancos ou tintos, mas têm em comum a receção da uva, desengace/esmagamento, prensagem, fermentação alcoólica e/ou fermentação malolática, transfegas, estabilização de frio, filtração e engarrafamento (Rosado, 2013; Pirra, 2007; Brito *et al.*, 2007; Rodrigues *et al.*, 2006; Vlyssides *et al.*, 2005), tal como demonstrado nas figuras 4 e 5. No decorrer do mesmo, são consumidos elevados volumes de água e produção de efluentes líquidos, uma vez que, para a produção de 1 L de vinho, podem ser consumidos 450 L de água (Costa *et al.*, 2020) e gerados 4 L de água residual (Flores *et al.*, 2019). Estes consumos são provenientes de lavagens de equipamentos (garrafas, depósitos, barricas, superfícies e linhas de transferência); lavagem de pavimento e superfícies e processos de arrefecimento (Jorge *et al.*, 2021). Para além desta água são também arrastados resíduos inerentes ao processo produtivo, tais como, películas de uva, grainhas, produtos enológicos (leveduras, ativantes, enzimas, colas, clarificantes, estabilizantes, taninos), borras, mosto, perdas de vinho, e ainda, desinfetantes, detergentes e desincrustantes. Tudo isso contribui para um efluente de pH ácido (3,5 – 6,5), de elevado teor de matéria orgânica (CQO), que pode diferir com a produção de vinho branco ou tinto (Ngwenya *et al.*, 2022), como demonstrado na figura 4 e 5, que pode ir de 2,5 a 270 g L^{-1}, P_{total} 0,04 a 0,3 g P L^{-1}, fenóis totais de 0,01 a 1,45 g Ácido gálico L^{-1}, SST 0,03 - 52 g L^{-1} (Ngwenya *et al.*, 2022), e ainda elementos como, polifenóis, álcoois, açúcares, ácidos orgânicos, taninos e ligninas (Amor *et al.*, 2019; Solís *et al.*, 2017; Arienzo *et al.*, 2009; Pirra, 2007; Malandra *et al.*, 2003).

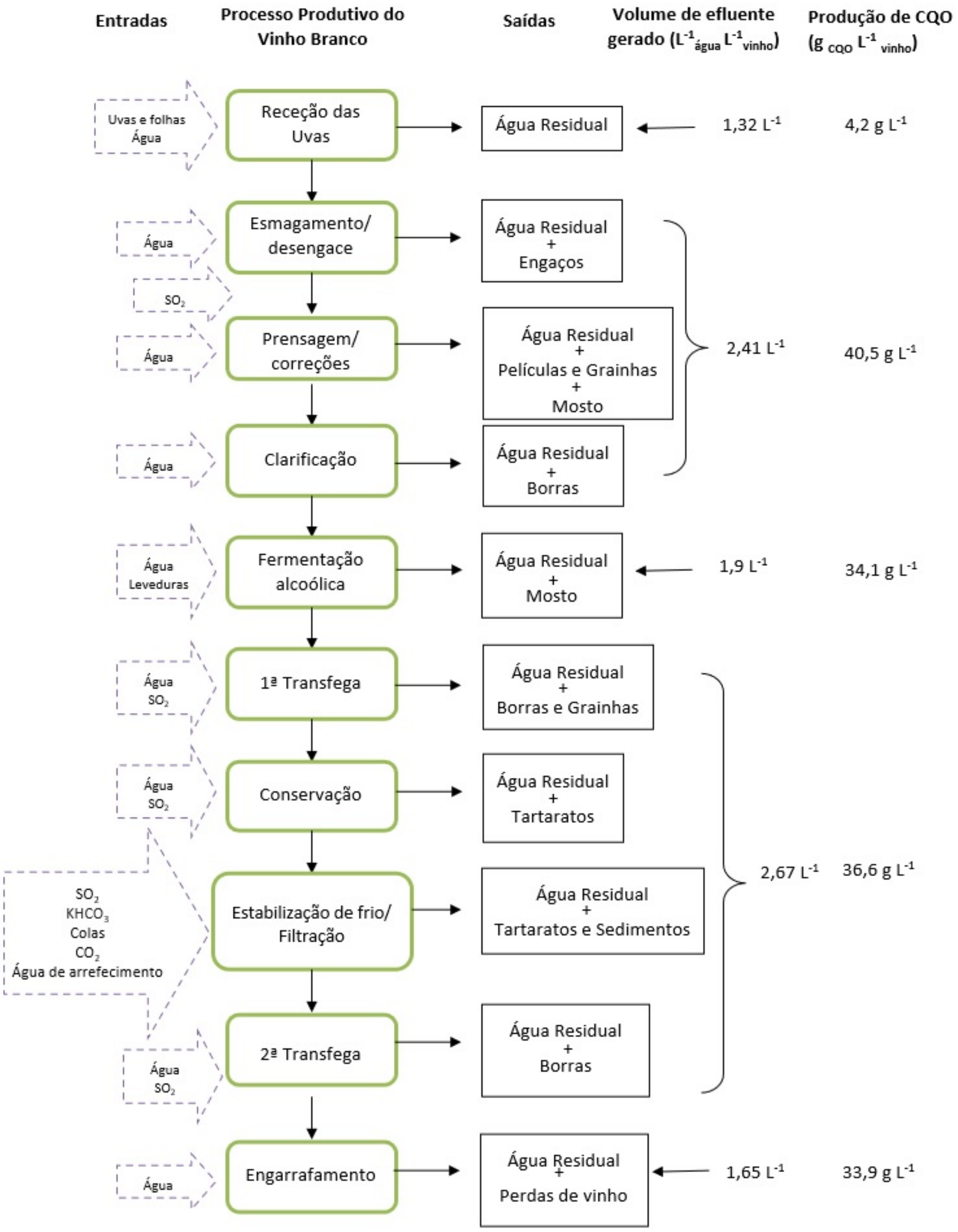


Figura 4 – Processo produtivo do vinho branco. Adaptado de: Pirra, 2007, Brito *et al.,* 2007, Rodrigues *et al.,* 2006.

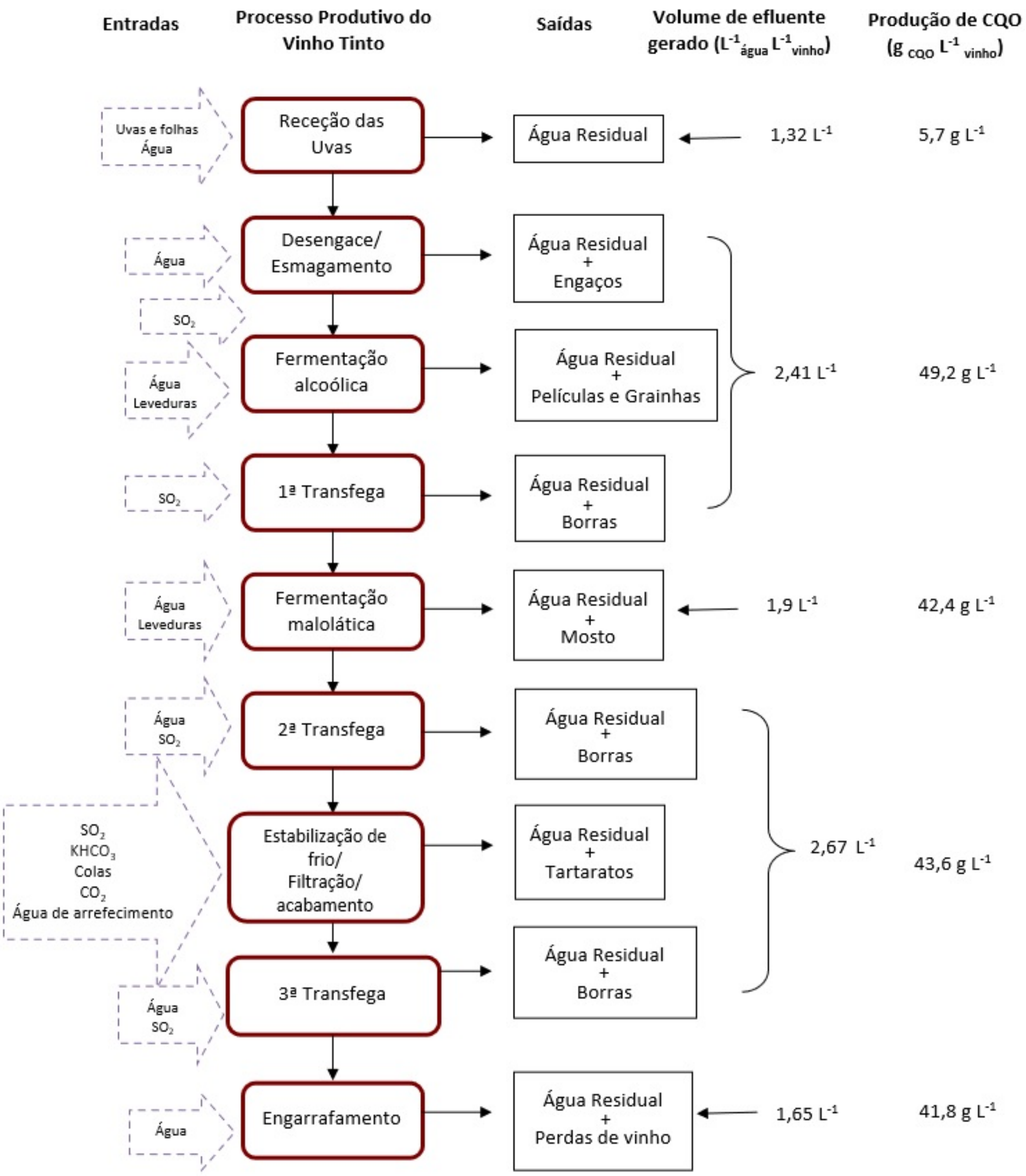


Figura 5 - Processo produtivo do vinho branco. Adaptado de: Pirra, 2007, Brito *et al.,* 2007, Rodrigues *et al.,* 2006.

1.1.3. Tecnologias para tratamento para efluentes de queijaria e adega

Existem já na literatura, inúmeras tecnologias para tratamento de efluentes de queijaria e adega, tais como:

- Digestão anaeróbia através de Biorreator anaeróbio de manta de lamas de fluxo ascendente (UASB), são bastante utilizados, uma vez que, minimizam recursos energéticos, recursos hídricos, geram poucos sedimentos. Nos efluentes de queijaria, os estudos já efetuados apontam para os tratamentos de digestão anaeróbia como os mais viáveis para a remoção das elevadas cargas orgânicas (Sampaio *et al.,* 2019, Carvalho *et al.*, 2013, Sayedm *et al.,* 1988). Este também tem sido um processo de eficácia comprovada para o efluente de adega (Ngwenya *et al.,* 2022; Keyser *et al*., 2003; Kalyuzhnyi *et al.,* 2001). No entanto, a aplicação de processos de digestão anaeróbia, apresenta como desvantagens a acidificação do pH do efluente, devido à produção de ácidos voláteis, impedindo a formação de lamas granulares e a eliminação de elevadas concentrações de CQO, comprometendo assim a sua descarga ou reutilização (Carvalho *et al.*, 2013), bem como, a necessidade de condições de estabilidade e mão de obra especializada para o seu manuseamento (Ngwenya *et al.,* 2022, Mosse *et al,.* 2012).

- Sistema de filtração por membranas, tais como, osmose inversa, nanofiltração e ultrafiltração, permitem a separação e redução da concentração de compostos orgânicos e inorgânicos de diversos efluentes, com alguma utilização no tratamento de efluentes de adega (Ioannou *et al.,* 2013; Ferrarini *et al.,* 2001) e queijaria (Yorgun *et al.,* 2008; Wenten e Khoiruddin, 2016). A principal desvantagem dos sistemas de separação por membranas, é a incrustação, devendo-se a vários fatores, nomeadamente a elevada carga orgânica, presença de sólidos , salinidade e pH ácidos (Bera *et al.,* 2021), todos estes, presentes nos efluentes de adega e queijaria.

- Processos de oxidação avançada, tais como, Fenton, foto-Fenton, fotocatálise e ozonização, têm sido processos a ganhar grande expressão, uma vez que, o oxidante químico utilizado no processo, geram radicais hidroxila, que atuam na degradação e eliminação de poluentes nas águas residuais, sendo uma ótima alternativa para efluentes como os de adega e queijaria, que apresentam um elevado teor de matéria orgânica (Jorge *et al.,* 2023; Trigueros *et al.,* 2022, Lucas *et al.,* 2009; Mosteo *et al.,* 2006; Ormad *et al.,* 2006;). A desvantagem deste processo é a difícil separação de catalisadores, pH de trabalho específico, elevada produção de lamas ricas em ferro, causando assim uma poluição secundária (Liang *et al.,* 2023).

- Zonas húmidas artificiais, são das mais utilizadas, uma vez que, pela dispersão das empresas de pequena e média dimensão, algumas delas, localizadas em áreas sensíveis, esta é a aplicação mais viável para o tratamento desses efluentes. Estas plantas têm a capacidade de eliminar componentes orgânicos e inorgânicos. Tem forte expressão nos efluentes de queijaria e adega (Aguilar *et al.,* 2022; Carvalho *et al.,* 2013; Serrano *et al.,* 2011). As suas principais vantagens são as de não agregar riscos para o ambiente na sua utilização, porém, necessita de pós-tratamentos, umas vez, o efluente de adega apresenta compostos tóxicos que inibem o processos de fitorremediação provocando o decréscimo da eficiência do de remoção de N e P bem como da fração inorgânica, tal como, Na^+, Cl^- (Aguilar *et al.,* 2022, Mader *et al.,* 2022, Flores *et al.*, 2019). Outra das desvantagens é a área considerável de implementação que este sistema carece, e, no caso dos efluentes de queijaria, a necessidade de um pré-tratamento do efluente, uma vez que estes sistemas não apresentam capacidades de remoção de óleos e gorduras (Carvalho *et al.,* 2013).

- Processos de coagulação/floculação e coagulação/floculação combinados. No processo de coagulação, os coloides em suspensão são estabilizados com através da ação do coagulante de carga oposta, as partículas destabilizadas agregam-se, fomentando a sedimentação. No processo de floculação, o floculante adsorve e liga partículas coloidais, até criar flocos, facilmente removidos por sedimentação. Uma vez que, estes processos convencionais têm sido relatados como insuficientes para a remoção de contaminantes dos efluentes, aliaram-se outras tecnologias (Jorge *et al.,* 2022), para o efluente de adega combinou-se o processo de coagulação-floculação à oxidação avançada (Jorge *et al.,* 2022). Para efluente de queijaria, foram estudados processos de coagulação/floculação usando $FeSO_4$ e $FeCl_3$ e biodegradação aeróbica (Rivas *et al.*, 2010 e 2011) que apresentam elevadas taxas de eficiência de remoção de contaminantes, no entanto, essas taxas de eficiência são melhoradas aquando da aplicação de um pré-tratamento (Rivas *et al.*, 2011).

1.2. Apresentação temática das publicações. Coerência e importância unificada da Tese.

Nas últimas décadas, a preservação do meio ambiente e o tratamento adequado das águas residuais industriais têm se tornado temas de vital importância para a sustentabilidade global. A indústria alimentar, devido à sua grande escala e diversidade, é uma das principais fontes geradoras de efluentes contaminados que podem ter um impacto significativo nos recursos hídricos e no ecossistema em geral. Entre as indústrias alimentares, a fabricação de queijo tem sido identificada como uma fonte considerável de águas residuais que requer atenção especial devido à complexidade dos seus contaminantes.

As águas residuais da fabricação de queijo são efluentes compostos por uma ampla variedade de compostos orgânicos e inorgânicos, gorduras, proteínas, lactose e bactérias, entre outros. Esses contaminantes podem afetar negativamente a qualidade da água e, se não forem tratados adequadamente, podem contribuir para a degradação do meio ambiente e colocar em risco a saúde pública. Conscientes dessa problemática, as publicações que compõem esta tese de doutorado têm se dedicado a estudar e desenvolver soluções inovadoras para o tratamento das águas residuais da fabricação de queijo (e em uma delas da adega para analisar a aplicabilidade geral do tratamento), com o objetivo de reduzir seu impacto ambiental e promover práticas sustentáveis.

Apresenta-se a seguir uma revisão temática das quatro publicações fundamentais que abordam o tratamento das águas residuais da fabricação de queijo / adega por meio de diferentes processos físico-químicos. O conjunto dessas publicações (juntamente com uma última em processo de revisão) proporciona uma visão integral e coesa da importância de encontrar soluções sustentáveis para o tratamento desse tipo de águas residuais agroalimentares.

As publicações selecionadas são as seguintes:

Cheese wastewater treatment by acid and basic precipitation: Application of H2SO4, HNO3, HCl, Ca(OH)2 and NaOH. Journal of Environmental Chemical Engineering, Volume 8, Issue 2, 103556. https://doi.org/10.1016/j.jece.2019.103556:
Esta publicação investiga a eficácia de diferentes ácidos e bases na precipitação de contaminantes presentes nas águas residuais da fabricação de queijo. O ajuste do pH por meio do uso de ácidos e bases é destacado como um método potencial para o tratamento desse tipo de efluentes. Ao explorar diferentes precipitantes químicos, o estudo estabelece as bases para futuras pesquisas na melhoria da eficiência de remoção de contaminantes e promove o desenvolvimento de soluções de tratamento mais efetivas.

Simple processes for contamination removal in cheesemaking wastewater: CaCO3, Mg(OH)2, FeSO4 and FeCl3. Journal of Environmental Chemical Engineering, Volume 8, Issue 4, 104034. https://doi.org/10.1016/j.jece.2020.104034:
Este estudo concentra-se na utilização de carbonato de cálcio, hidróxido de magnésio, sulfato de ferro e cloreto de ferro para a remoção de contaminantes presentes nas águas residuais da fabricação de queijo. A pesquisa destaca o potencial de utilizar materiais de baixo custo e fácil disponibilidade para reduzir efetivamente os contaminantes, contribuindo para uma abordagem mais sustentável e econômica para o tratamento de águas residuais.

Cheese manufacturing wastewater treatment by combined physicochemical processes for reuse and fertilizer production. Journal of Environmental Management, Volume 264, 110470.https://doi.org/10.1016/j.jece.2020.104034:
Esta publicação investiga a combinação de processos físico-químicos para o tratamento das águas residuais da fabricação de queijo. Integrando diferentes técnicas de tratamento, o estudo tem como objetivo purificar não apenas as águas residuais para possível reutilização, mas também produzir valiosos fertilizantes a partir do efluente tratado, promovendo práticas agrícolas sustentáveis e fechando o ciclo dos nutrientes.

Immediate one-step lime precipitation process for the valorization of winery wastewater to agricultural purposes. Environ Sci Pollut Res, Volume 28, 18382-18391.
https://doi.org/10.1007/s11356-020-11933-3:
Esta pesquisa explora um processo de precipitação com cal em um único passo para a valorização das águas residuais de adega para fins agrícolas. O estudo demonstra um método rápido e eficaz para tratar os efluentes de adegas, promovendo a reutilização da água na agricultura e minimizando o impacto ambiental.

A combinação de diferentes processos físico-químicos também demonstra uma abordagem abrangente para enfrentar a complexidade das águas residuais da fabricação de queijo. Os estudos que investigam a utilização de várias técnicas de tratamento enfatizam a importância de adaptar a abordagem de acordo com as características específicas das águas residuais e os objetivos desejados, como a reutilização ou a produção de subprodutos valiosos.

A reutilização das águas residuais tratadas na indústria de laticínios e na agricultura apresenta inúmeros benefícios. Em primeiro lugar, a reciclagem das águas residuais reduz a demanda por água fresca, o que pode ser especialmente relevante em regiões com escassez de água. Além disso, o uso das águas residuais tratadas na produção de queijo e na agricultura pode contribuir para uma gestão mais eficiente dos recursos hídricos e a redução dos custos associados ao abastecimento de água. A reutilização das águas residuais também pode ajudar a melhorar a sustentabilidade das operações da indústria de laticínios, reduzindo sua pegada hídrica e minimizando seu impacto ambiental.

Outro aspecto importante destacado nessas publicações é a produção de subprodutos valiosos a partir das águas residuais tratadas. Especificamente, a obtenção de fertilizantes a partir do efluente tratado apresenta um potencial significativo. Os fertilizantes derivados das águas residuais podem ser ricos em nutrientes essenciais, como nitrogênio, fósforo e potássio, que são fundamentais para o crescimento das plantas. Ao reciclar esses nutrientes e aplicar os fertilizantes nos campos agrícolas, fecha-se o ciclo dos nutrientes e reduz-se a necessidade de utilizar fertilizantes químicos sintéticos, que podem ter um impacto negativo no meio ambiente.

Além da reutilização e valorização de subprodutos, outra questão relevante abordada nessas publicações é a importância da otimização dos processos de tratamento. A eficiência dos sistemas de tratamento é fundamental para garantir que as águas residuais sejam tratadas adequadamente e que os contaminantes sejam removidos de forma eficaz. A pesquisa nesse campo busca melhorar a eficiência dos processos de precipitação, coagulação e filtração, entre outros, para obter os melhores resultados em termos de purificação do efluente.

É importante mencionar que esses estudos não se concentram apenas na aplicação dos processos físico-químicos, mas também abordam aspectos econômicos e ambientais. A viabilidade econômica dos sistemas de tratamento é um fator determinante para sua implementação em grande escala. Portanto, avaliam-se os custos associados à aplicação dos tratamentos e à produção de subprodutos, bem como os benefícios econômicos derivados da reutilização da água e da valorização de subprodutos.

Quanto aos aspectos ambientais, estudam-se as possíveis emissões de contaminantes na atmosfera e a geração de resíduos sólidos durante o processo de tratamento. A minimização dos impactos ambientais associados ao tratamento de águas residuais é fundamental para garantir a sustentabilidade global e a proteção do meio ambiente.

Em resumo, as publicações que compõem esta tese de doutorado apresentam uma visão abrangente e coesa do tratamento de águas residuais da fabricação de queijo. Desde a aplicação de processos físico-químicos para a remoção de contaminantes até a reutilização da água tratada e a produção de subprodutos valiosos, esses estudos buscam soluções integradas e sustentáveis para reduzir o impacto ambiental da indústria de laticínios. Além disso, abordam aspectos econômicos e ambientais, refletindo uma abordagem holística para o tratamento de águas residuais. Por meio da pesquisa contínua e do desenvolvimento de soluções inovadoras, esses esforços contribuem para a preservação do meio ambiente, a promoção de práticas sustentáveis e o alcance de um futuro mais resiliente e equitativo.

CAPÍTULO II - JUSTIFICAÇÃO E OBJETIVOS DA TESE

As indústrias agroalimentares de pequena e média dimensão apresentam grande expressão na região do Alentejo, dessas, a indústria do queijo e do vinho apresentam forte tradição e representação gastronómica local, com produtos de denominação de origem e denominação de origem protegida, apresentam elevadas produções, gerando assim, elevados volumes de efluentes.

Os efluentes gerados por estas agroindústrias constituem um problema ambiental e de saúde pública, devido aos conteúdos em compostos orgânicos e inorgânicos, presença de sólidos, óleos e gorduras, nutrientes e existência de condições de pH extremas, geralmente ácidos. Assim, estes efluentes podem provocar eutrofização, salinização, desoxigenação, acidificação, toxicidade e contaminação microbiana, necessitando de tecnologias complexas capazes de reduzir o nível de contaminação. Tradicionalmente estes efluentes têm sido tratados por processos biológicos e físico-químicos, como se de águas residuais domésticas se tratasse.

Os processo de tratamento até agora estudados na literatura, e mencionados no ponto 1.1.3, apesar de grandes vantagens, apresentam limitações, uma vez que muitos requerem a utilização de dois ou mais reagentes ou processos. Alguns deles apenas servem como pré-tratamento do efluente, outros utilizam equipamentos e/ou reagentes dispendiosos e de difícil aplicabilidade no local da produção do efluente. Por verificar que continua a existir uma lacuna ao nível do tratamento e reutilização destes efluentes, e verificando que a maioria destas indústrias são de pequena e média dimensão, com recursos económicos limitados e por vezes geograficamente distantes de estações de tratamento de águas residuais, o objetivo deste trabalho foi o de colmatar essa lacuna e desenvolver processos de tratamento simples e económicos e fáceis de aplicar em todo o tipo de indústria do queijo e do vinho que :

- não necessitem de pré-tratamento de modo a poderem ser aplicados às águas residuais brutas;

- apresentarem capacidade de adaptabilidade às flutuações de caudal e das propriedades físico-químicas e biológicas dos efluentes;

- não necessitam recorrer a mão de obra especializada, ou a sistemas/instalações sofisticada;

- sejam de fácil aplicação e controlo;

- possam ser aplicados à temperatura ambiente, com presença de luz e oxigénio;

- não necessitem de aquisição de reatores ou procedimentos/ infraestruturas dispendiosas;

- podem ser aplicados no local em que são produzidos;

- atinjam elevadas taxas de remoção de contaminação, nomeadamente, matéria orgânica (CQO), P_{total}, turvação e toxicidade;

- simultaneamente removam contaminação microbiológica;

- produzam um precipitado passível de ser valorizado;

- possam ser transferidos para as agroindústrias e sejam a ferramenta que permitirá a estas agroindústrias transitar facilmente para um modelo de economia circular.

CAPÍTULO III – TRATAMENTO DE ÁGUA RESIDUAL DE QUEIJARIA POR PROCESSOS DE COAGULAÇÃO

3.1. Coagulação/Floculação

Este processo é composto por duas etapas, a primeira consiste na introdução de agentes químicos coagulantes, com capacidade de destabilizar e neutralizar as cargas elétricas das partículas coloidais (cujo diâmetro é entre 0,01 e 1,0 µm), levando à produção de microflocos, fazendo com que essas partículas, e/ou matéria orgânica presentes na água, se aglomerem. A segunda etapa envolve a adição de um agente floculante, formando flocos de maior dimensão e mais densos, tornando a sua remoção mais fácil, favorecendo outras etapas à posteriori, melhorando a sedimentação, flotação e filtração (Karam *et al.*, 2021, Prazeres *et al.,* 2013, Renault *et al,.* 2009, Amuda *e* Amoo 2007). Os agentes químicos utilizados no processo de coagulação e floculação são geralmente sais de alumínio e de ferro, tais como:

Sulfato de alumínio ($Al(SO_4)_3$) é utilizado no tratamento dos efluentes a um pH ótimo entre 4,0 a 7,0, (eq. 2 e 3) (Rivas *et al.*, 2010, Alves 2007):

$$Al(SO_4)_3 + 3Ca(HCO_3)_2 \rightarrow Al(OH)_{3(s)} + 3CaSO_4 + 6CO_{2(g)} \quad (1)$$

$$Al(SO_4)_3 + 3Ca(OH)_2 \rightarrow Al(OH)_{3(s)} + 3CaSO_4 \quad (2)$$

$$Al(SO_4)_3 + 6NaOH \rightarrow 2Al(OH)_{3(s)} + 3Na_2SO_4 \quad (3)$$

$$Al(SO_4)_3 + 3Na_2CO_3 + 3H_2O \rightarrow 2Al(OH)_{3(s)} + 3Na_2SO_4 + 3CO_{2(g)} \quad (4)$$

$$Al(SO_4)_3 + 6Na_2CO_3 + 6H_2O \rightarrow 2Al(OH)_{3(s)} + 3Na_2SO_4 + 6NaHCO_3 \quad (5)$$

$$Al(SO_4)_3 + 6H_2O \rightarrow 2Al(OH)_3 + 3H_2O \quad (6)$$

Sulfato ferroso ($FeSO_4$) é usado em efluentes cujo pH seja básico, ≥ 8,5 (Alves 2007), formando hidróxidos de ferro (eq. 7 e 8), de acordo com as reações:

$$FeSO_4 + Ca(OH)_2 \leftrightarrows Fe(OH)_2\downarrow + CaSO_4 \quad (7)$$

$$4Fe(OH)_2 + O_2 + 2H_2O \leftrightarrows 4Fe(OH)_3\downarrow \quad (8)$$

Cloreto férrico ($FeCl_3$) com capacidade de clarificar a água, principalmente quando combinada com cal (eq. 10 e 11):

$FeCl_3 + 3H_2O \rightarrow Fe(OH)_3\downarrow + 3H^+ + 3Cl^-$ (9)

$2FeCl_3 + 3Ca(HCO_3)_2 \rightarrow 2Fe(OH)_{3(s)}\downarrow + 3CaCl_2 + 6CO_2$ (10)

$2FeCl_3 + 3Ca(OH)_2 \rightarrow 2Fe(OH)_{3(s)}\downarrow + 3CaCl_2$ (11)

$FeCl_3 + 3NaOH \rightarrow Fe(OH)_{3(s)}\downarrow + 3NaCl$ (12)

Sulfato férrico $Fe(SO_4)_3$ é o reagente que melhor se adapta aos diversos pH do efluentes, no intervalo de 4 a 11, permitindo a formação de precipitados, tal como demonstram as seguintes reações:

$Fe(SO_4)_3 + 3Ca(OH) \rightarrow 3CaSO_4 + 2Fe(OH)_3\downarrow$ (13)

$Fe(SO_4)_3 + 3Ca(HCO_3)_2 \rightarrow 3CaSO_4 + 2Fe(OH)_3\downarrow + 6CO_2$ (14)

$Fe(SO_4)_3 + H_2O \rightarrow Fe^{2+} + SO_4^{2-} + 2H^+$ (15)

Devido à sua capacidade de formação de flocos, fomentando a sedimentação, este processo é bastante eficaz na remoção de partículas coloidais, responsáveis pela cor e turvação do efluente, sólidos suspensos, matéria orgânica e microrganismos (Amuda *e* Aldade 2005). Para a eliminação de fósforo, os coagulantes $FeCl_3$ e $Al(SO_4)_3$, são os que, na literatura apresentam melhores remoções, uma vez, ao reagirem com o ião fosfato promovem a reação (Karam *et al.,* 2021, Prazeres 2013):

$Fe^{3+} + PO_4^{3-} \rightarrow FePO_4$ (16)

$Al^{3+} + PO_4^{3-} \rightarrow AlPO_4$ (17)

Este tem sido um processo cada vez mais utilizado em efluentes com elevada contaminação por óleos e gorduras como os efluentes de queijaria (Karam *et al.*, 2021, Zhao *et al.*, 2020). A principais vantagens na sua utilização, para além da boa capacidade de remoção de contaminação do efluente, é a facilidade de aplicação e operação, ser económico e energeticamente eficiente (Zhao *et al.*, 2020, Amuda *e* Amoo 2007). Mas também apresentam desvantagens, tais como, a formação de iões metálicos, as altas dosagens aplicadas e a sensibilidade ao pH do efluente (Zhao *et al,.* 2020), bem como a formação de lamas.

Assim, neste trabalho pretendeu-se inovar estes processos através da experimentação de condições de agitação em 3 etapas de velocidade decrescente não encontrada na literatura consultada e dois coagulantes não convencionais ($CaCO_3$ e $Mg(OH)_2$) em que os processos de coagulação ocorrem em meio básico (**Artigo 1**). A coagulação em condições de agitação sequencial decrescente de agitação foi também testada mas utilizando combinações de coagulantes (**Artigo2**).

Artigo 1 - Simple processes for contamination removal in cheesemaking wastewater: $CaCO_3$, $Mg(OH)_2$, $FeSO_4$ and $FeCl_3$

Artigo 2 - Cheese manufacturing wastewater treatment by combined physicochemical processes for reuse and fertilizer production

RESUMO

O efluente de queijaria foi recolhido no Alentejo, em Serpa, Distrito de Beja. O processo de remoção de contaminação de água residual de queijaria com os reagentes $CaCO_3$, $Mg(OH)_2$, $FeSO_4$ e $FeCl_3$ (**Artigo1**), iniciou com uma agitação 1200-1400 rpm do efluente bruto, posteriormente, seguiu-se a aplicação das várias dosagens de coagulante, de forma individual, mantendo-se a agitação por 1 minuto:

$CaCO_3$: 12,5; 25; 50; 75; 100 e 125 g L^{-1}

$Mg(OH)_2$: 12,5; 25; 50; 75; 100 e 125 g L^{-1}

$FeSO_4$: 0,1; 0,2; 0,4; 0,6; 0,8 e 1,0 g L^{-1}

$FeCl_3$: 0,1; 0,2; 0,4; 0,6; 0,8 e 1,0 g L^{-1}

Posteriormente, reduziu-se a velocidade de agitação para 700-800 rpm, durante 1 minuto, seguindo-se nova redução para 300-400 rpm, durante 1 minuto, com sedimentação de 24 horas.

Os resultados, demonstraram que o $CaCO_3$ foi o reagente com melhores eficiências de remoção de CQO (12-36%); P_{total} (47-94%); turvação (31-88%) e fenóis (44-58%). Os reagentes $Mg(OH)_2$ e $FeSO_4$, não permitiram a remoção de CQO em nenhuma das dosagens aplicadas. A aplicação de $FeCl_3$, permitiu obter remoções de matéria orgânica, mas não superiores a 27%.

Este estudo permitiu identificar o $CaCO_3$ como o melhor reagente na eliminação da contaminação da água residual de queijaria. Para incrementar as eficiências de remoção atingidas pela aplicação do $CaCO_3$ de 75 g L^{-1}, combinaram-se os coagulantes $FeSO_4$, $FeCl_3$ e $Ca(OH)_2$ nas dosagens (**Artigo 2**):

$FeSO_4$: 0,1; 0,2; 0,4; 0,6; 0,8 e 1,0 g L^{-1}

$FeCl_3$: 0,1; 0,2; 0,4; 0,6; 0,8 e 1,0 g L^{-1}

$Ca(OH)_2$ (hidratada a 200 g L^{-1}): 0,1; 0,2; 0,4; 0,6; 0,8 e 1,0 g L^{-1}

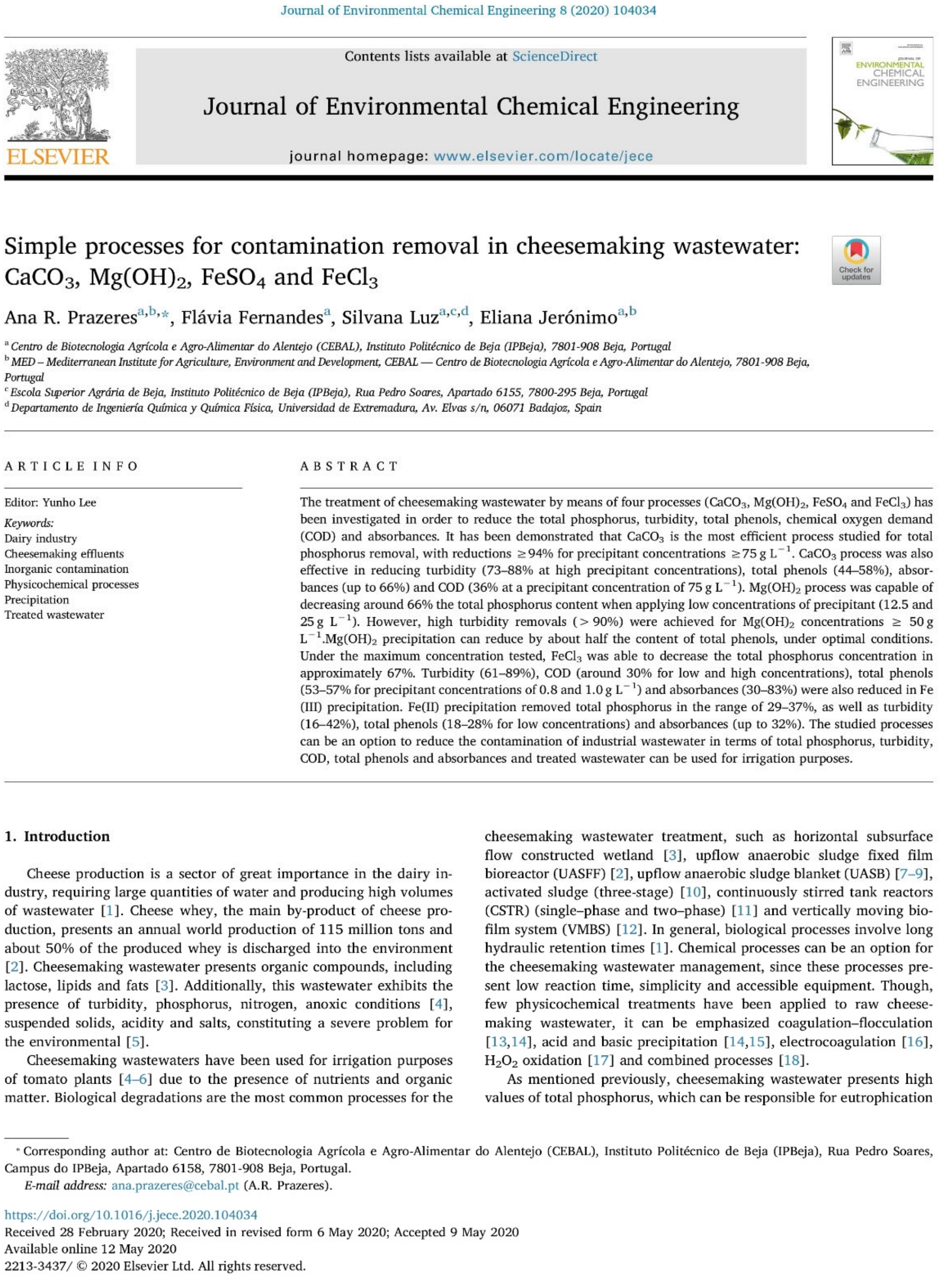
A agitação ocorreu em três etapas de velocidade decrescente já descrita, com sedimentação de 48 horas. As melhores dosagens atingidas no processo combinadas foram: $CaCO_3$ 75 g L^{-1} + $FeCl_3$ 1,0 g L^{-1}; $CaCO_3$ 75 g L^{-1} + $FeSO_4$ 1,0 g L^{-1}; $CaCO_3$ 75 g L^{-1} + $Ca(OH)_2$ 0,6 g L^{-1}. Observaram-se elevadas remoções dos parâmetros monitorizados:

P_{total} com remoção acima dos 92% e os óleos e gorduras > 97%, como se pode observar na tabela 2. O melhor tratamento foi o $CaCO_3$ 75 g L^{-1} + $FeSO_4$ 1,0 g L^{-1}, com remoções de CQO > 55%, P_{total} de 92%, turvação > 95%, sólidos suspensos totais > 50%, óleos e gorduras > 97%.

Tabela 1 – Eficiência da aplicação da coagulação a efluentes de queijaria - Percentagens de remoção em função do reagente utilizado: $CaCO_3$ + $FeCl_3$ 1,0 g L^{-1} e $CaCO_3$ + $FeSO_4$ 1,0 g L^{-1} e $CaCO_3$ + $Ca(OH)_2$ 0,6 g L^{-1}

	% Remoção					
Tratamento	CQO	Fósforo	Turvação	Fenóis	Sólidos Totais	Óleos e gorduras
$CaCO_3$ + $FeCl_3$ 1,0 g L^{-1}	14,4	98,1	59,3	98,7	31,8	98,2
$CaCO_3$ + $FeSO_4$ 1,0 g L^{-1}	55,1	92	95,7	59,1	51	97,2
$CaCO_3$ + $Ca(OH)_2$ 0,6 g L^{-1}	46,2	92,6	80,9	49	52,6	98,9

Artigo 1 - Simple processes for contamination removal in cheesemaking wastewater: $CaCO_3$, $Mg(OH)_2$, $FeSO_4$ and $FeCl_3$



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Simple processes for contamination removal in cheesemaking wastewater: $CaCO_3$, $Mg(OH)_2$, $FeSO_4$ and $FeCl_3$

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ABSTRACT

The treatment of cheesemaking wastewater by means of four processes ($CaCO_3$, $Mg(OH)_2$, $FeSO_4$ and $FeCl_3$) has been investigated in order to reduce the total phosphorus, turbidity, total phenols, chemical oxygen demand (COD) and absorbances. It has been demonstrated that $CaCO_3$ is the most efficient process studied for total phosphorus removal, with reductions $\geq 94\%$ for precipitant concentrations ≥ 75 g L^{-1}. $CaCO_3$ process was also effective in reducing turbidity (73–88% at high precipitant concentrations), total phenols (44–58%), absorbances (up to 66%) and COD (36% at a precipitant concentration of 75 g L^{-1}). $Mg(OH)_2$ process was capable of decreasing around 66% the total phosphorus content when applying low concentrations of precipitant (12.5 and 25 g L^{-1}). However, high turbidity removals (> 90%) were achieved for $Mg(OH)_2$ concentrations $\geq$ 50 g L^{-1}.$Mg(OH)_2$ precipitation can reduce by about half the content of total phenols, under optimal conditions. Under the maximum concentration tested, $FeCl_3$ was able to decrease the total phosphorus concentration in approximately 67%. Turbidity (61–89%), COD (around 30% for low and high concentrations), total phenols (53–57% for precipitant concentrations of 0.8 and 1.0 g L^{-1}) and absorbances (30–83%) were also reduced in Fe(III) precipitation. Fe(II) precipitation removed total phosphorus in the range of 29–37%, as well as turbidity (16–42%), total phenols (18–28% for low concentrations) and absorbances (up to 32%). The studied processes can be an option to reduce the contamination of industrial wastewater in terms of total phosphorus, turbidity, COD, total phenols and absorbances and treated wastewater can be used for irrigation purposes.

1. Introduction

Cheese production is a sector of great importance in the dairy industry, requiring large quantities of water and producing high volumes of wastewater [1]. Cheese whey, the main by-product of cheese production, presents an annual world production of 115 million tons and about 50% of the produced whey is discharged into the environment [2]. Cheesemaking wastewater presents organic compounds, including lactose, lipids and fats [3]. Additionally, this wastewater exhibits the presence of turbidity, phosphorus, nitrogen, anoxic conditions [4], suspended solids, acidity and salts, constituting a severe problem for the environmental [5].

Cheesemaking wastewaters have been used for irrigation purposes of tomato plants [4–6] due to the presence of nutrients and organic matter. Biological degradations are the most common processes for the cheesemaking wastewater treatment, such as horizontal subsurface flow constructed wetland [3], upflow anaerobic sludge fixed film bioreactor (UASFF) [2], upflow anaerobic sludge blanket (UASB) [7–9], activated sludge (three-stage) [10], continuously stirred tank reactors (CSTR) (single–phase and two–phase) [11] and vertically moving biofilm system (VMBS) [12]. In general, biological processes involve long hydraulic retention times [1]. Chemical processes can be an option for the cheesemaking wastewater management, since these processes present low reaction time, simplicity and accessible equipment. Though, few physicochemical treatments have been applied to raw cheesemaking wastewater, it can be emphasized coagulation–flocculation [13,14], acid and basic precipitation [14,15], electrocoagulation [16], H_2O_2 oxidation [17] and combined processes [18].

As mentioned previously, cheesemaking wastewater presents high values of total phosphorus, which can be responsible for eutrophication

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phenomenon, with social, economic and environmental impacts [19]. This phenomenon is considered as one of the leading problems [20]. Among the impacts of eutrophication, it can be stated the decrease of water quality (depth, color, transparency, oxygen content, biomass and species) and the influence in the treatment processes and recreational activities [20,21]. The relationship between nitrogen, phosphorus and carbon to promote the eutrophication is 15:1:105. Consequently, low phosphorus concentrations can be responsible for algal growth [20].

Several methods have been applied for the phosphorus removal, namely, electrodialysis [22], reverse osmosis [23], biological processes (algae [24,25], constructed wetland systems [26,27], activated sludge systems [28], anaerobic-anoxic sequencing batch reactor (SBR) [29], hydroponics [30]), adsorption [31] and chemical precipitation [32].

Calcium, magnesium and iron additions can be inexpensive and flexible alternatives for the total phosphorus reduction in wastewater. Additionally, chemical precipitation can be applied in several phases of the wastewater treatment and formed sludge can be used in agriculture [33]. Ca, Cu and Zn can be applied in the iron precipitation in order to reduce mono- and polyphosphonates by adsorption onto surfaces [34]. Fe (III) and Al (III) precipitations are responsible for 80–95% of the total phosphorus reduction [32]. $FeSO_4$ precipitation is an economic process [35] for the phosphorus removal [13]. Precipitation using calcium is a frequent process owing to the low cost and simplicity [34]. Magnesium salts have been little used for phosphorus precipitation, however, these salts allow the struvite formation, sludge stabilization and ammonia removal. Struvite (magnesium ammonium phosphate hexahydrate, $MgNH_4PO_4$. $6H_2O$) is the most important compound for wastewater plant recovery as fertilizer [34]. As a result, the present work is focused on the investigation of four innovative chemical processes using calcium, magnesium and iron ions in order to reduce phosphorus in cheesemaking wastewater. Reductions of chemical oxygen demand (COD), turbidity, total phenols and absorbances were also investigated, as well as calcium, magnesium, hydroxide, carbonates and bicarbonates behaviors.

2. Materials and methods

2.1. Chemicals

Iron (II) sulfate heptahydrate (ACS, ISO, Reag. pH Eur, $M = 278.02\ g\ mol^{-1}$) was supplied by MERCK (VWR, Portugal). Iron (III) chloride (> 97%, $M = 162.20\ g\ mol^{-1}$) was obtained from SIGMA-ALDRICH (SIGMA, Portugal). Precipitated calcium carbonate ($M = 100.09\ g\ mol^{-1}$) was supplied by José Manuel Gomes dos Santos, LDA (Portugal). Magnesium hydroxide (RFE, BP, Ph. Eur., $M = 58.33$ g mol^{-1}) was obtained from Panreac (Portugal).

2.2. Raw cheesemaking wastewater collection

The raw cheesemaking wastewater was obtained in a tank from a small factory located in the Alentejo region (Portugal). The factory has several types of cheeses, namely, sheep and goat cheeses. Samples were collected in 5 L polyethylene terephthalate (PET) containers and stored at − 20 °C and 4 °C before characterization and treatment. This factory valorizes a percentage of cheese whey for animal feed. Cheesemaking wastewater contains cheese whey (resulting from cheese production), second cheese whey (resulting from cottage cheese production) and washing water of facilities and equipment. The production of dairy effluents can reach values of approximately 9 $m^3 m^{-3}$ of processed milk.

2.3. Analytical determinations

pH was evaluated through a meter Consort C 861 (Consort nv, Belgique). Temperature and conductivity were monitored in a VWR CO 3100 H apparatus using an electrode VWR CO 11. Characteristic absorbances were measured, after appropriate dilution, in the ultraviolet and visible region using a Thermo Scientific Helios α spectrophotometer (Thermo Scientific, Germany). In the ultraviolet region, absorbances at wavelengths of 220 nm (presence of low molecular weight compounds (simple molecules) formed from complex molecules (humic acids)) and 254 nm (high molecular weight organic compounds with high degree of aromaticity, high number of double and triple bonds, and phenolic groups) were monitored, while in the visible region, absorbances at wavelengths of 410 nm (color indicator) and 600 nm (optical density of the microorganisms cultures) were evaluated [36–42]. Specific absorbances were calculated multiplying the absorbance and dilution used according to Standard Methods [36].

Turbidity was measured using a WTW Turb550 turbidimeter (WTW, Germany), comparing white light transmitted by the samples and formazine standard suspensions [36,43,44]. COD was determined by colorimetric method after a digestion step (150 ± 2 °C for 2 h) in a digester Techne Dri-block DB 200/3 (ERT Lda, Portugal) [36,44].

Total phosphorus contents were estimated after calcination at 600 °C during 2 h and digestion step with 6 M HCl solution using a heating/stirring plate. The digested samples were filtered using filters Whatman™ 1001. Total phosphorus was determined using a colorimetric method by measuring the absorbance at 470 nm after the reaction of orthophosphates with vanadate-molybdate reagent [36]. Total hardness was evaluated by volumetric complexation method and eriochrome black T indicator, while calcium hardness was monitored using the presence of calcon indicator [36]. Magnesium hardness was calculated by difference between total hardness and calcium hardness.

Phenolphthalein alkalinity and methyl orange alkalinity were evaluated using a volumetric method with a solution of hydrochloric acid and phenolphthalein indicator and indicator of methyl orange [36]. Total alkalinity was calculated through the sum of alkalinity to phenolphthalein and alkalinity to methyl orange. Folin-Ciocalteu phenol reagent was used to determine the total phenols content through a spectrophotometric method, according to Singleton and Rossi [45].

2.4. Experimental procedure

$CaCO_3$, $Mg(OH)_2$, $FeSO_4$ and $FeCl_3$ precipitations were conducted using 250 mL of raw cheesemaking wastewater. Raw wastewaters were maintained in agitation of 1200–1400 rpm before the reactions for 1 min. In the experiments, several doses of precipitant were applied: $CaCO_3$ (12.5, 25, 50, 75, 100 and 125 g L^{-1}), $Mg(OH)_2$ (12.5, 25, 50, 75, 100 and 125 g L^{-1}), $FeSO_4$ (0.1, 0.2, 0.4, 0.6, 0.8 and 1.0 g L^{-1}) and $FeCl_3$ (0.1, 0.2, 0.4, 0.6, 0.8 and 1.0 g L^{-1}). After the application of the dose, stirring was gradually stopped (700–800 rpm during 1 min followed by 300–400 rpm during 1 min). Agglomerates were formed, which precipitated during 24 h to ensure the precipitation of small and large agglomerates, forming two phases: supernatant and sludge according to Rivas et al. [13,14]. Supernatants were characterized in terms of pH, conductivity, absorbances, turbidity, COD, total phosphorus, total phenols, calcic and total hardness, alkalinity to phenolphthalein and alkalinity to methyl orange.

3. Results and discussion

3.1. Cheesemaking wastewater characterization

Cheesemaking wastewater characterization is presented in Table 1. Cheesemaking wastewater exhibited acid pH ($4.162 - 4.675$) and high conductivity value ($4.86 - 5.54\ dS\ m^{-1}$). Similar values of pH were observed by Kalyuzhnyi et al. [8], Martins et al. [46] and Rivas et al. [13]. Cheesemaking wastewater presented an organic matter content monitored by COD in the range of $4416.7 - 5250.0\ mg\ L^{-1}$, similar to that observed by Gutiérrez et al. [7] and Kalyuzhnyi et al. [8]. Calcium ($195.6 - 270.8\ mg\ L^{-1}$) and magnesium ($72.1 - 172.4\ mg\ L^{-1}$) are responsible for total hardness ($934.6 - 1347.5$ mg $CaCO_3\ L^{-1}$) and total

Table 1
Cheesemaking wastewater characterization.

Parameter	Units	Interval	Mean value
pH	Sorensen scale	4.162 − 4.675	4.520 ± 0.2055
Conductivity	dS m^{-1}	4.86 − 5.54	5.23 ± 0.277
Temperature	°C	15.1 − 17.6	16.3 ± 0.93
Specific absorbance at 220 nm	cm^{-1}	29.950 − 35.000	31.960 ± 1.917
Specific absorbance at 254 nm	cm^{-1}	7.850 − 9.800	9.020 ± 0.736
Specific absorbance at 410 nm	cm^{-1}	4.150 − 5.050	4.580 ± 0.327
Specific absorbance at 600 nm	cm^{-1}	2.650 − 3.550	2.990 ± 0.360
Phenolphthalein alkalinity	mg $CaCO_3$ L^{-1}	< DL	< DL
Total alkalinity	mg $CaCO_3$ L^{-1}	1155.4 − 1708.5	1388.9 ± 251.1
Hydroxides	mg $CaCO_3$ L^{-1}	< DL	< DL
Carbonates	mg $CaCO_3$ L^{-1}	< DL	< DL
Bicarbonates	mg $CaCO_3$ L^{-1}	1155.4 − 1708.5	1388.9 ± 251.1
Total hardness	mg $CaCO_3$ L^{-1}	934.6 − 1347.5	1054.7 ± 168.69
Calcic hardness	mg $CaCO_3$ L^{-1}	487.9 − 675.6	589.3 ± 72.20
Magnesium hardness	mg $CaCO_3$ L^{-1}	296.5 − 709.4	465.4 ± 151.30
Calcium	mg L^{-1}	195.6 − 270.8	236.2 ± 28.94
Magnesium	mg L^{-1}	72.1 − 172.4	113.1 ± 36.77
COD	mg L^{-1}	4416.7 − 5250.0	4916.7 ± 353.55
Turbidity	NTU	536.49 − 659.73	594.92 ± 49.542
Total phosphorus	mg L^{-1}	1796.1 − 4894.1	3411.8 ± 1255.64
Total phenols	mg equivalent of gallic acid L^{-1}	65.00 − 82.86	75.92 ± 6.760

Results after 5 replicates. COD, chemical oxygen demand; DL, detection limit.

alkalinity (1155.4 − 1708.5 mg $CaCO_3$ L^{-1}) was present in the form of bicarbonates. Furthermore, cheesemaking wastewater was characterized by high total phosphorus concentration in the range of 1796.1 − 4894.1 mg L^{-1}.

3.2. *Cheesemaking wastewater treatment*

3.2.1. *$CaCO_3$ addition*

Cheesemaking wastewater was treated by adding $CaCO_3$ in the range of 12.5–125 g L^{-1}. Fig. 1 presents the results obtained. The pH of the effluent maintained around 6 for $CaCO_3$ concentrations in the range of 12.5–50.0 g L^{-1}, rising to pH close to 7.0 for concentrations in the range of 75 − 125 g L^{-1} (Fig. 1A). The conductivity of treated wastewater increased compared to raw wastewater, being this situation more notorious for the maximum precipitant concentration studied (7.305 ± 0.007 dS m^{-1}) (Fig. 1A). $CaCO_3$ process was also effective in reducing characteristic absorbances (4.7–65.7%) depending on the operating conditions (Fig. 1B). As expected, $CaCO_3$ process increased the calcium content (496.4–624.3 mg L^{-1}) (Fig. 1C), being a source of calcium for the reuse of treated wastewater. Similar results were obtained for magnesium content (154.2–382.2 mg L^{-1}) (Fig. 1C) probably due to the calcium competition phenomenon by ions hydroxyl, carbonates and bicarbonates. On the other hand, increasing concentrations of precipitant generated greater sludge formation, leaving less volume of treated wastewater and consequently greater amount of magnesium. Additionally, magnesium and bicarbonates contents increased linearly ($r^2 = 0.9386$ and $r^2 = 0.9035$) with the $CaCO_3$ concentration (Fig. 1C). Carbonates and pH influence the hydroxyapatite formation, since the carbonates compete with phosphates for calcium ions, resulting in less free calcium ions. Hydroxyapatite generation can be inhibited by the presence or formation of carbonates at pH = 8.0. Nonetheless, at pH = 9.0, phosphate precipitation rate is not significantly reduced by the presence of carbonates. Moreover, carbonate and phosphate can co-precipitate when the solution presents pH in the range of 9–11, forming a precipitate with lower phosphorus concentration [34].

The removals of COD (12.2–36.3%) were only achieved for concentrations between 75 and 125 g L^{-1}, decreasing by increasing the $CaCO_3$ concentration (Fig. 1D). COD elimination may be due to the formation of insoluble species, such as calcium hydroxide and magnesium carbonate that entrain the organic particles:

$$CaCO_3 + 2H_2O \rightleftharpoons Ca(OH)_2\downarrow + H_2CO_3 \tag{1}$$

$$2CaCO_3 + 2H_2O \rightleftharpoons Ca(OH)_2\downarrow + Ca(HCO_3)_2 \tag{2}$$

$$MgCO_3 + CaCO_3 + 2H_2O \rightleftharpoons Ca(OH)_2\downarrow + Mg(HCO_3)_2 \tag{3}$$

$$Mg(OH)_2 + CaCO_3 \rightleftharpoons Ca(OH)_2\downarrow + MgCO_3\downarrow \tag{4}$$

The colloidal particles destabilization can occur when moderate to high contents of Ca (> 4 mM) were used or are present [20]. These authors mentioned that Ca complexes and neutralizes the charges present in particles, decreasing the net surface charge.

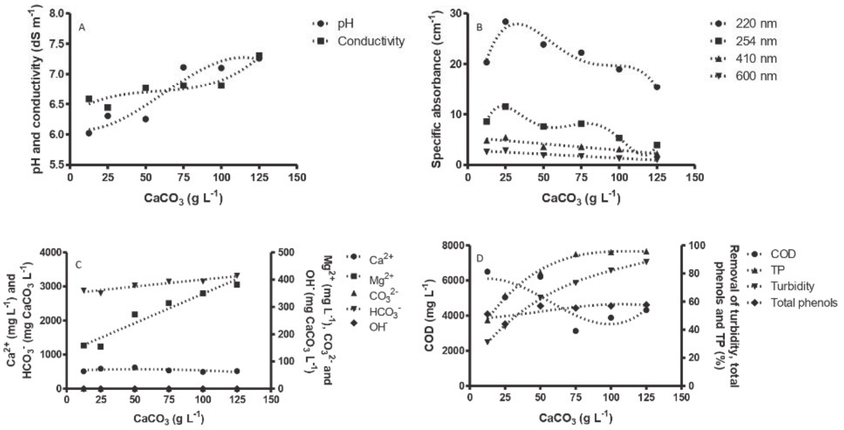

Fig. 1. Precipitation of cheesemaking wastewater by $CaCO_3$.
Effect on pH, conductivity (A), absorbances (220, 254, 410 and 600 nm) (B), calcium, magnesium, carbonates, bicarbonates, hydroxides (C), chemical oxygen demand (COD), total phosphorus (TP), turbidity and total phenols (D).

Total phosphorus removal increased (46.7–93.8%) up to the application of 75 g L^{-1} (pH = 7.112) of $CaCO_3$, remaining practically constant from this value (95.2–95.8%, pH = 7.101–7.260) (Fig. 1D). Similar behavior was obtained for turbidity removal (31.1–88.2%) (Fig. 1D). The application of $CaCO_3$ was performed with pH > 6, allowing the natural precipitation of $Ca_{10}(PO_4)_6(OH)_2$. When pH is higher than 7, $Ca_{10}(PO_4)_6(OH)_2$ is the main phosphate precipitate [20], with high removal efficiencies of phosphorus.

The addition of iron, aluminum and calcium salts can be responsible for the transformation of dissolved ortho-phosphates into particulate form (chemical precipitates of low solubility). Ortho-phosphates and particulate P can be eliminated by precipitation and flocculation [47]. The transformation of dissolved P species into particulate form can be realized through 3 mechanisms: 1) chemical precipitation with formation of metal-phosphate-hydroxo complexes of low solubility; 2) specific adsorption of phosphates onto metal-hydroxo precipitates; 3) coagulation by destabilization of P containing colloids [47]. Total phosphorus elimination can result from the interaction between calcium ions and phosphate ions to form calcium phosphates (hydroxyapatite) [34], according to the equation [48]:

$$5Ca^{2+} + 3PO_4^{3-} + H_2O \rightarrow Ca_5(PO_4)_3OH\downarrow + H^+ \quad (5)$$

The $CaCO_3$ process was able to reduce about half of the content of total phenols (44.1–57.8%) (Fig. 1D). Ni, Zn (more than 97%) and As (lower than 50%) were removed by granulated calcium carbonate as coagulant in artificially contaminated water owing to the sweep precipitation [49].

3.2.2. *$Mg(OH)_2$ addition*

$Mg(OH)_2$ precipitant was applied for the cheesemaking wastewater treatment (12.5–125 g L^{-1}). Fig. 2 reports the results obtained. The pH of the treated effluent presented values close to neutrality (6.5–6.9) (Fig. 2A). Due to the addition of ions in raw wastewater, conductivity of the treated effluent increased from 5.225 dS m^{-1} (raw wastewater) to values in the range of 6.885–7.105 dS m^{-1}. The minimum value of conductivity was obtained at a concentration of 75 g L^{-1} of precipitant (Fig. 2A). Under the same conditions, calcium exhibited the minimum value (18.8 mg L^{-1}). In this context, it can be referred that calcium removal increased from 74.5% for a $Mg(OH)_2$ precipitant concentration of 12.5 g L^{-1} to 92.0% for a $Mg(OH)_2$ precipitant concentration of 75.0 g L^{-1}. From this point, the calcium content in the treated effluent increased to value of 45.1 mg L^{-1}, representing a removal of 80.9% (Fig. 2C). As expected, the addition of $Mg(OH)_2$ increased the concentration of magnesium to values in the range of 674.2–774.5 mg L^{-1} (Fig. 2C). Thus, treated effluent can be a source of magnesium as fertilizer for agriculture. $Mg(OH)_2$ addition to raw cheesemaking wastewater was capable to remove absorbances at 220 (6.1–27.7%) and 410 (11.0–26.9%) nm (Fig. 2B). Alkalinity was present in the form of bicarbonates with concentrations in the range of 2446.0–2814.7 mg L^{-1} $CaCO_3$ (Fig. 2C).

$Mg(OH)_2$ precipitant was not efficient in removing organic matter (Fig. 2D). However, this process was capable of eliminating total phosphorus in the range of 20.5–66.5%, obtaining the highest removal at low concentrations tested (12.5 and 25 g L^{-1}) (Fig. 2D). The elimination of total phosphorus in $Mg(OH)_2$ process can be due to the formation of struvite or magnesium ammonium phosphate (MAP) according to the following reaction [19]:

$$Mg^{2+} + NH_4^{+} + PO_4^{3-} \rightleftharpoons MgNH_4PO_4\downarrow \quad (6)$$

Struvite formation occurs when wastewater presents high contents of soluble phosphorus and ammonium, low suspended solids presence, pH > 7.5 and a molecular ratio of $1(Mg^{2+}):1(NH_4^{+}):1(PO_4^{3-})$ [34].

Turbidity was removed efficiently from $Mg(OH)_2$ concentrations of 50 g L^{-1} with removals > 90% (Fig. 2D). Regarding total phenols, reductions in the range of 35.3–49.3% were obtained (Fig. 2D). Wada et al. [50] observed good results for carcinogenic phenols and aromatic amines removal through a process using tyrosinase and a cationic polymer coagulant in aqueous solution.

3.2.3. *$FeSO_4$ addition*

Cheesemaking wastewater was treated by $FeSO_4$ addition in the range of 0.1–1.0 g L^{-1} Fig. 3 illustrates the results obtained. The application of $FeSO_4$ led to the increase of pH of the treated effluent (4.705–5.745) compared to raw wastewater (4.520). pH of the treated effluent presented values around 5.7 when low concentrations (0.1 and 0.2 g L^{-1}) were applied, decreasing to values of 4.7–4.8 for concentrations of $FeSO_4$ in the range of 0.4–1.0 g L^{-1} (Fig. 3A). Opposite trend was observed for conductivity of the treated effluent, presenting

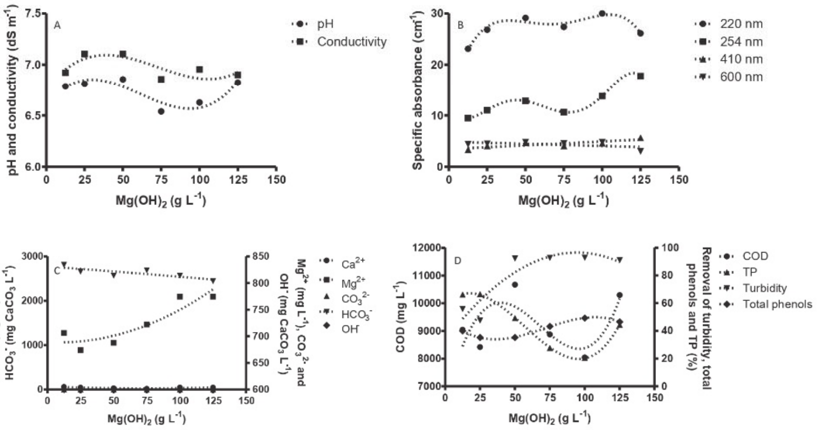

Fig. 2. Precipitation of cheesemaking wastewater by $Mg(OH)_2$.
Effect on pH, conductivity (A), absorbances (220, 254, 410 and 600 nm) (B), calcium, magnesium, carbonates, bicarbonates, hydroxides (C), chemical oxygen demand (COD), total phosphorus (TP), turbidity and total phenols (D).

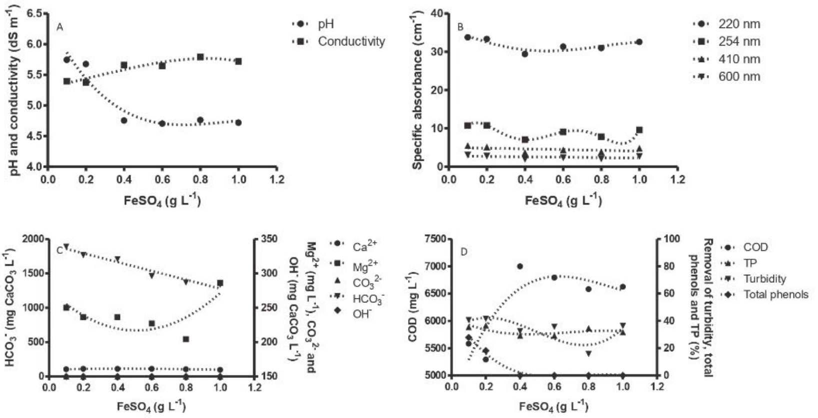

Fig. 3. Precipitation of cheesemaking wastewater by $FeSO_4$.
Effect on pH, conductivity (A), absorbances (220, 254, 410 and 600 nm) (B), calcium, magnesium, carbonates, bicarbonates, hydroxides (C), chemical oxygen demand (COD), total phosphorus (TP), turbidity and total phenols (D).

values in the range of 5.375–5.790 dS m^{-1} (Fig. 3A). In general, absorbances removal ($\leq$ 32.3 %) occurred between 0.4 and 0.8 g L^{-1} of precipitant (Fig. 3B). $FeSO_4$ addition to cheesemaking wastewater reduced the calcium content (52.2–58.6%), presenting values in the range of 97.8–112.8 mg L^{-1} (Fig. 3C). The treated effluent exhibited alkalinity in the form of bicarbonates, which presented a linear relationship ($r^2 = 0.9466$) with $FeSO_4$ concentration (Fig. 3C). Organic matter monitored by COD was not removed by $FeSO_4$ process (Fig. 3D). $FeSO_4$ can precipitate organic compounds but the oxidation of Fe(II) is necessary [35]. Thus, in this work probably did not occur the effective oxidation of Fe(II).

Nevertheless, this process removed total phosphorus in the range of 29.1–36.6%, producing effluents with total phosphorus values of 2162.7–2417.6 mg L^{-1} (Fig. 3D) According to Clark et al. [21], iron (II) process can be an attractive selection in terms of economical profits. These authors refer two mechanisms for iron (II) in a precipitation process depending on environmental circumstances:

- in the presence of oxygen, iron (II) can be oxidized to iron (III) forming strong complexes with the phosphates, namely condensed phosphates present in industrial wastewater,
- in the nonexistence of oxygen, iron (II) will not be oxidized to iron (III) forming a fine precipitate (Vivianite, $Fe_3(PO_4)_2.8H_20$).

Similar results were obtained for turbidity parameter with the highest removals ($\approx$ 41%) for low concentrations experiments of $FeSO_4$ (0.1 and 0.2 g L^{-1}) (Fig. 3D). Total phenols were also removed (18.2–27.8%) for low concentrations (0.1 and 0.2 g L^{-1}) in the $FeSO_4$ precipitation process, producing effluents with total phenols content of 56.8–64.3 mg L^{-1} (Fig. 3D). High removal efficiencies (97%) were found for phenol when Abdelwahab et al. [51] used electrocoagulation with aluminum cathode and anode at high current density, solution pH of 7, and after 2 h of reaction for the treatment of oil refinery wastewater.

3.2.4. $FeCl_3$ addition

$FeCl_3$ addition (0.1–1.0 g L^{-1}) was tested for contamination removal in cheesemaking wastewater. Fig. 4 presents the results obtained. The application of increasing $FeCl_3$ concentrations to raw wastewater decreased linearly ($r^2 = 0.996$) the pH of the treated effluent (3.883–4.768) (Fig. 4A). Opposite behavior was demonstrated by conductivity, increasing linearly ($r^2 = 0.9824$) with the precipitant concentration. This parameter increased from 5.100 dS m^{-1} at low concentrations (0.1 and 0.2 g L^{-1}) to 5.435 dS m^{-1} at high concentration (1.0 g L^{-1}) (Fig. 4A). Removals of absorbances at 220 (29.7–50.8%) and 254 (58.1–82.0%) nm increased with the precipitant concentration. Elimination of absorbances at 410 and 600 nm increased for a precipitant concentration of 0.4 g L^{-1} ($\geq$72%), maintaining practically constant from this point (Fig. 4B). Calcium content was reduced in the range of 61.8–68.2%. Thus, $FeCl_3$ process produced effluents with 75.2–90.3 mg L^{-1} of calcium content (Fig. 4C). Treated effluents presented a linear decrease of bicarbonates ($r^2 = 0.9646$) by increasing the concentration of $FeCl_3$ (Fig. 4C).

The phosphate removal in $FeCl_3$ precipitation is affected by iron concentration, final pH and initial phosphate content [32]. Phosphorus elimination occurred for $FeCl_3$ concentration $\geq$ 0.4 g L^{-1}, with removals in the range of 8.7–66.5% (Fig. 4D). The elimination of phosphorus by $FeCl_3$ process can be due to a strong interaction between PO_4^{3-} and metal ions (Fe^{3+}) [19] forming insoluble $FePO_4$ [20]. However, this system occurs through complex reactions with the formation of complexes, polymers and precipitates [32]:

$$Fe^{3+} + H_2O \rightleftharpoons Fe(OH)^{2+} + H^+ \quad (7)$$

$$Fe^{3+} + 2H_2O \rightleftharpoons Fe(OH)_2{}^+ + 2H^+ \quad (8)$$

$$2Fe^{3+} + 2H_2O \rightleftharpoons Fe_2(OH)_2{}^{4+} + 2H^+ \quad (9)$$

$$Fe^{3+} + 3H_2O \rightleftharpoons Fe(OH)_3{}^{\circ}(aq) + 3H^+ \quad (10)$$

$$Fe^{3+} + 4H_2O \rightleftharpoons Fe(OH)_4{}^- + 4H^+ \quad (11)$$

$$3Fe^{3+} + 4H_2O \rightleftharpoons Fe_3(OH)_4{}^{5+} + 4H^+ \quad (12)$$

$$H_3PO_4 \rightleftharpoons H_2PO_4{}^- + H^+ \quad (13)$$

$$H_2PO_4{}^- \rightleftharpoons HPO_4{}^{2-} + H^+ \quad (14)$$

$$HPO_4{}^{2-} \rightleftharpoons PO_4{}^{3-} + H^+ \quad (15)$$

$$Fe^{3+} + HPO_4{}^{2-} \rightleftharpoons FeHPO_4{}^+ \quad (16)$$

$$Fe^{3+} + H_2PO_4{}^- \rightleftharpoons FeH_2PO_4{}^{2+} \quad (17)$$

$$Fe_rPO_4(OH)_{3r\text{-}3}\downarrow \rightleftharpoons rFe^{3+} + PO_4{}^{3-} + (3r\text{-}3)OH\text{-} \quad (18)$$

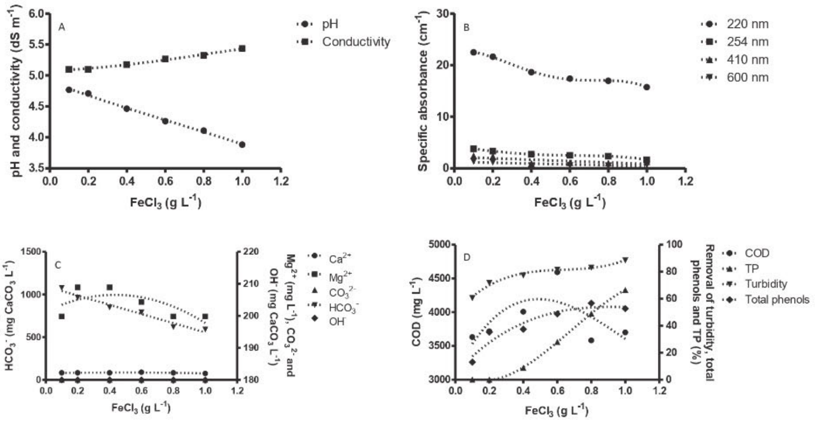

Fig. 4. Precipitation of cheesemaking wastewater by FeCl$_3$.
Effect on pH, conductivity (A), absorbances (220, 254, 410 and 600 nm) (B), calcium, magnesium, carbonates, bicarbonates, hydroxides (C), chemical oxygen demand (COD), total phosphorus (TP), turbidity and total phenols (D).

$$Fe_{1.6}H_2PO_4(OH)_{3.8}\downarrow \rightleftharpoons 1.6Fe^{3+} + H_2PO_4^{-} + 3.8OH^{-} \quad (19)$$

$$am\text{ - }FeOOH + 3H^{+} \rightleftharpoons Fe^{3+} + 2H_2O \quad (20)$$

Increasing concentrations of precipitant have caused an increase in turbidity removal (60.5–88.5%) (Fig. 4D). The formation of amorphous ferric hydroxide precipitates when applying pH > 6.5 and coagulant concentration > 0.03 mM allows the trapping and co-precipitation of colloidal impurities [20].

Similar results were observed for total phenols elimination (13.1–56.7%) (Fig. 4D). Total phenol removal can be due to the formation of $Fe(OC_6H_5)_6^{3-}$ complex according to the following reaction [52]:

$$FeCl_3 + 6C_6H_5OH \rightarrow Fe(OC_6H_5)_6^{3-}\downarrow + 6H^{+} + 3Cl^{-} \quad (21)$$

High removal efficiency of phenol can be achieved when increasing FeCl$_3$ concentration owing to the greater available surface area for adsorption through electrostatic interactions [52].

CaCO$_3$, Mg(OH)$_2$, FeCl$_3$ and FeSO$_4$ constitute effective processes to reduce the contamination of cheesemaking wastewater. Table 2 presents the costs associated with different precipitants, calculated based on market prices. CaCO$_3$ was able to reduce total phosphorus (≥ 94%) for high precipitant concentrations (≥ 75 g L^{-1}). However, this process becomes more expensive than Mg(OH)$_2$, since Mg(OH)$_2$ reduced in about 66% the total phosphorus when using low concentrations of precipitant (12.5 and 25 g L^{-1}). Similar results for the total phosphorus elimination were obtained for FeCl$_3$ process, under the maximum concentration investigated (1.0 g L^{-1}), being cheaper than Mg(OH)$_2$. The total phosphorus removal decreased for values in the range of 29–37% when FeSO$_4$ process was applied. This process is the cheapest compared to the other processes studied. CaCO$_3$, Mg(OH)$_2$, FeCl$_3$ can reduce by about half the content of total phenols and around 90% the turbidity, under optimal conditions. FeSO$_4$ was less effective in eliminating total phenols and turbidity, compared to CaCO$_3$, Mg(OH)$_2$ and FeCl$_3$. CaCO$_3$ was effective to reduce COD (36% at a precipitant concentration of 75 g L^{-1}). FeCl$_3$ presented similar COD removals (around 30% for low and high concentrations).

Table 2
Costs associated with different precipitants.

Precipitant	Concentration (g L^{-1})	Costs (€ L^{-1})
CaCO$_3$	12.5	0.118
	25	0.236
	50	0.472
	75	0.708
	100	0.943
	125	1.179
Mg(OH)$_2$	12.5	0.624
	25	1.248
	50	2.497
	75	3.745
	100	4.994
	125	6.242
FeSO$_4$	0.1	0.002
	0.2	0.005
	0.4	0.009
	0.6	0.014
	0.8	0.018
	1.0	0.023
FeCl$_3$	0.1	0.006
	0.2	0.011
	0.4	0.023
	0.6	0.034
	0.8	0.045
	1.0	0.056

4. Conclusions

Physicochemical processes with the addition of calcium, iron and magnesium may be alternatives for the reduction of contamination of cheesemaking wastewater. In this context, CaCO$_3$, Mg(OH)$_2$, FeSO$_4$ and FeCl$_3$ precipitations are simple processes for the contamination removal in cheesemaking wastewater, needing only a stirrer and a reactor. CaCO$_3$ was the most efficient process for the removal of total phosphorus (≥ 94%), followed by FeCl$_3$ (67%) and Mg(OH)$_2$ (66%). However, CaCO$_3$ has a higher cost compared with Mg(OH)$_2$ and FeCl$_3$. FeSO$_4$ process was less efficient in the removal of total phosphorus, but still able to reduce 29–37%. The latter process is the cheapest. CaCO$_3$, Mg(OH)$_2$, FeCl$_3$ processes eliminated about 50% of the total phenols. Turbidity was removed in the range of 16–93%, with the Mg(OH)$_2$ process being the most efficient in reducing this parameter. All the processes studied were able to reduce absorbances. Additionally, CaCO$_3$ and FeCl$_3$ removed organic matter monitored by COD (7–36%). Despite the high rates of contamination reduction, it is necessary to apply a complementary treatment to reduce the level of contamination. Thus, these processes can be pre-treatments of physicochemical and biological processes.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Artigo 2 - Cheese manufacturing wastewater treatment by combined physicochemical processes for reuse and fertilizer production

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Research article

Cheese manufacturing wastewater treatment by combined physicochemical processes for reuse and fertilizer production

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ABSTRACT

Making cheese manufacturing environmentally sustainable is a major concern in the integrated management of this industrial sector. This concern is mainly due to the environmental impact of the discharge of its wastewaters, carrying heavy loads of salinity, nutrients, organic matter, solids and oils and fats. These discharges must meet increasingly stringent quality requirements. Some physicochemical (e.g. coagulation-flocculation, precipitation, oxidation) and biological (e.g. aerobic and anaerobic bioreactors and wetlands) treatments have been studied to address this problem. However, these treatments involve costs that some producers cannot sustain, face difficulties with biological reactor operational stability and often fail to consistently produce effluents compatible with discharge standards. In this context, aiming at the design of a simple and economical treatment method, several precipitation processes were tested using a fixed dosage of $CaCO_3$ (75 g/L), combined with different dosages of $FeCl_3$, $FeSO_4$ or $Ca(OH)_2$. The goal of the treatment was to produce a supernatant that would be evaluated as to its suitability for discharge into natural water courses or municipal treatment systems, or for reuse applications. The generated sludge would be evaluated for possible agricultural valorization. Through the measurement of the relevant supernatant quality parameters and using statistical analysis, it was possible to choose the best dosages for each of the tested coagulants (1.0, 1.0 and 0.6 g/L for $FeCl_3$, $FeSO_4$ and $Ca(OH)_2$, respectively). Among these, the most efficient treatment was obtained with $CaCO_3$ 75 g/L + $FeSO_4$ 1.0 g/L. For this best-case scenario, the treatment led to removal yield values of 55.1% for chemical oxygen demand (COD), 92.0% for total phosphorus, 95.7% for turbidity, 59.1% for total phenols, 94.3% for nitrates, 71.0% for nitrites, 51.0% for total solids (TS) and 97.2% for oils and fats. The treatment did not produce an effluent supernatant with adequate quality for direct discharge into water courses, serving however as an efficient pretreatment for agricultural reuse. All the sludges generated in these treatments showed good potential for agricultural valorization due to their high nutrient content, along with pH and conductivity values within the acceptable ranges for soil application. Thus, this work contributes for a better integration of the cheese manufacturing industry in the overall aims of water and nutrient resources recovery in rural, agricultural areas.

1. Introduction

The main environmental impacts of the dairy industries are related to their liquid effluents since between 1 and 6 L of wastewater are generated per liter of processed milk. Solid waste and atmospheric emissions are also produced, frequently disposed of without any type of control or treatment (Henares, 2015). It is estimated that, during cheese manufacturing, milk discharges occur in the range of 0.5–4% (Omil et al., 2003), which are included in the effluent.

Wastewaters from cheese manufacturing may cause environmental

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and public health impacts (Prazeres et al., 2016b), such as salinization and chemical and microbiological contamination of soils, degradation of the quality of ground and surface waters, oxygen depletion in these, the release of strong odors, occurrence of foams and surface layers of fats, insect proliferation, and accumulation of toxic compounds in ecosystems.

The complexity of the effluents from cheese manufacturing renders their management difficult. So, detailed knowledge on their chemical composition is essential for an adequate study of potential treatments. Cheese manufacturing wastewater is characterized by a greenish yellow color due to the presence of riboflavin (vitamin B2) (de Wit, 2001). However, this wastewater may also show a whitish color. Its characteristics strongly depend on the type of milk used, and the quantity and quality of the employed wash water and cleaning agents (Carvalho et al., 2013). About 70% of the organic matter content in the effluent is lactose. In general, these effluents have acidic properties and higher contents of organic matter, oils and fats, suspended solids, nutrients and salinity when compared to other dairy industry effluents (Prazeres, 2013; Prazeres et al., 2013b). The high lactose and fat contents are the main causes of high chemical and biochemical oxygen demand values (COD and BOD, respectively). Consequently, wastewaters from cheese manufacturing cause serious problems of organic overload if discharged in a municipal sewage treatment system (Janczukowicz et al., 2008). Thus, in most cases, these wastewaters are kept in storage ponds or tanks (Luz et al., 2017) for prolonged periods, which are not adequate management options in what concerns environmental and public health protection.

Thus, the effluents from cheese manufacturing must be subjected to effective treatments to render them compatible with their intended discharge option. The latter could be into the municipal sewage network, to natural surface waters and onto soils, all covered by legal obligations in Portugal (Decree-Law 236/98; Ministério do Ambiente, 1998).

Conventional biological treatments may not be able to completely remove the pollution loads described above, but they are still the most commonly used (Tirado et al., 2018). In addition, they can be used in complement to new treatment techniques to achieve higher efficiencies, both in terms of pollutant removal and in economic terms.

Wastewaters from cheese manufacturing have also been treated by physicochemical processes, such as combined electrochemical processes (electrocoagulation followed by an electrochemical advanced oxidation process) (Tirado et al., 2018), continuous electrocoagulation (Un et al., 2014), NaOH precipitation (Prazeres et al., 2016a), $FeCl_3$ coagulation-flocculation, $Ca(OH)_2$ precipitation (Prazeres et al., 2016b), acid precipitation (H_2SO_4, HCl and HNO_3) (Prazeres et al., 2019) and H_2O_2 oxidation (Prazeres et al., 2019b). Chemical processes were shown to be an alternative to remove lactose, COD and BOD, oils and fats, total phosphorus, nitrogen, turbidity, pollutants associated to absorbance at characteristic wavelengths in the UV–visible range, total suspended solids (TSS), sulphates, calcium, magnesium and chlorides (Rivas et al., 2010, 2011; Prazeres et al., 2013b, 2016a). However, the application of $CaCO_3$ in chemical treatments of cheese manufacturing wastewaters has not been reported.

The application of some physicochemical treatments (coagulation-flocculation and precipitation) produces sludges carrying the compounds precipitated by the procedure. These sludges exhibit several features that make them adequate for application in agricultural soils. Their organic matter content can improve the physical properties of the soil including granule size, water infiltration capacity, water retention and aeration (Metcalf and Eddy, 2003). It can also contribute to soil cation exchange capacity, allowing the retention of potassium, calcium and magnesium, and increase soil microbiological diversity, helping in nitrogen, potassium and phosphorus capture by plants (Brito et al., 2009). Despite the efficiency of acid and alkaline precipitation applied to cheese manufacturing wastewaters, these techniques produce effluents with extreme pH values, requiring pH adjustment as final polishing.

In the present work, precipitation processes were studied using a combination of $CaCO_3$, keeping pH values between 7.0 and 8.0, complemented with $FeCl_3$, $FeSO_4$ or $Ca(OH)_2$, under different dosages. The application of these treatments to a real cheese manufacturing effluent was assessed, through the measurement of the most relevant quality parameters both in the resulting supernatant and in the generated sludge. Discharge and agricultural valorization options are here discussed, based on the obtained results.

2. Materials and methods

2.1. Raw cheese manufacturing wastewater

Cheese manufacturing wastewater was collected from a small facility located in the Alentejo region of Portugal. The samples were collected from the equalization tank and showed a murky appearance with high amounts of fat at the surface and in suspension. The samples were frozen within 2 h of being collected and stored at −18 °C until use. Before being used in the experiments, the samples were thawed overnight at room temperature.

2.2. Reagents and treatment procedures

In this study, a fixed dosage of calcium carbonate was used as first reagent (75 g/L), combined with iron chloride (III), iron sulfate (II) or calcium hydroxide as second reagents in different dosages of their commercial forms (0.1; 0.2; 0.4; 0.6; 0.8; 1.0 g/L). Iron chloride (III) ($FeCl_3.6H_2O$) and iron sulfate (II) ($FeSO_4.7H_2O$) were obtained from Merck Millipore, while calcium carbonate ($CaCO_3$) and calcium hydroxide ($Ca(OH)_2$) were obtained from Panreac AppliChem. Reagents $CaCO_3$, $FeCl_3$ and $FeSO_4$ were added in powder form in the experimental procedures, while reagent $Ca(OH)_2$ was added in the form of a concentrated solution in distilled water, at 200 g/L.

All other reagents were high purity pro-analysis from several suppliers.

The experiments with raw cheese manufacturing wastewater samples were carried out in Erlenmeyer flasks with a magnetic stirrer to guarantee a uniform mix. Each experiment was done in triplicate and simultaneously, under the same operational conditions. Calcium carbonate (75 g/L) was added to 200 mL of raw wastewater, and the value of pH was registered after its complete dissolution. In this period, the samples were kept under constant magnetic stirring, at close to 550 rpm. For the addition of the second reagent, the process depended on its physical state. For solid reagents, the process was as described for the first reagent. For aqueous solutions, no dissolution period was required. After adding the reagents to the raw wastewater, the mixture was maintained under constant 700–800 rpm stirring for 1 min, followed by 1 min at 300–400 rpm. During the subsequent sedimentation process, lasting 48 h, the samples were kept at room temperature without agitation, and were covered with Parafilm M® to avoid contaminations and losses by evaporation. After the settling period, the supernatants were carefully collected, excluding any surface fat layer, and stored at 4 °C. The leftover sludges, including the fat layer, were stored at room temperature.

For the best reagent combinations, further sedimentation tests were conducted in a normalized 1-L graduated cylinder filled to a height of 35 cm, using the mixtures obtained after the stirred steps of the procedure described above. The evolution in time of the height of the interface between the settled sludge blanket and the supernatant was registered for a period of 2 h to estimate the settling velocities.

2.3. Analytical methods

2.3.1. Raw and treated wastewater characterization

Sample pH was determined with a WTW InoLab apparatus (pH electrode SenTix 41). Electrical conductivity (EC) and turbidity were monitored with a VWR CO 3100 H SET conductivimeter and a WTW Turb550 turbidimeter, respectively. Absorbance at different wavelengths was measured in a Helios-Alfa UV–Visible Thermo Scientific spectrophotometer, against water. Wavelengths used are indicative of low molecular weight compounds (220 nm) as mentioned by Prazeres et al., 2019, aromatic and unsaturated compounds (254 nm) as described in APHA et al. (1999), color (410 nm) as described in Rivas et al. (2010) and optical density of suspended matter (600 nm) as described by Megharaj et al. (2003) for microorganism cultures. Chemical oxygen demand (COD) was quantified by a colorimetric method at 600 nm after hot acid digestion with potassium dichromate (150 °C for 2 h) in the presence of silver and mercury sulfates in a Hatch COD Reactor digester (APHA et al., 1999). Total phosphorus was evaluated by a colorimetric method at 470 nm after the reaction of orthophosphates with a vanadate-molybdate reagent (APHA et al., 1999). In order to determine alkalinity, samples were titrated with hydrochloric acid to a given pH end point (APHA et al., 1999), namely, 8.3 for hydroxide alkalinity using phenolphthalein as indicator and 4.5 for total alkalinity using methyl orange. The measurement of total and calcium hardness values was done to determine the concentrations of calcium and magnesium by volumetric complexation with ethylenediamine tetraacetic acid (EDTA). Total hardness was determined in the presence of eriochrome black T as indicator, while the Calcon indicator was used to determine the calcium content. The total phenolic substances content was determined by the Folin–Ciocalteu method (Makkar, 2003). Nitrates were quantified by the sodium salicylate method (Scheiner, 1974) while nitrites were determined by the Griess reaction using sulfanilamide and N-(1-Naphthyl)ethylenediamine (NED) as reactants (APHA et al., 1999). Total solids (TS) were determined directly by evaporating the water from a known volume of an unfiltered sample at 105 °C in an oven (Holelab) (APHA et al., 1999) and subsequent weighing of the residue. To determine total fixed solids (TFS), the residue remaining after evaporation was ignited at 600 °C in a muffle (Nabertherm) and the resulting ash was weighted, the loss of weight being a measure of the total volatile solids (TVS) (APHA et al., 1999). Total suspended solids (TSS) were obtained by vacuum filtration with standard filters, from the residue weight retained in the filter disk after drying at 105 °C, while total dissolved solids (TDS) were calculated by the difference between TS and TSS (APHA et al., 1999). The content of oils and fats was determined after extraction in a Sohxlet apparatus, using petroleum ether as the extracting solvent (APHA et al., 1999). The chloride content was titrated with silver nitrate in the presence of potassium chromate through Mohr's method (APHA et al., 1999).

2.3.2. Sludge characterization

The volume and weight of the settled sludges were measured after the supernatant was drawn off. Then, the sludge was spread on glass petri dishes open to the air and left to dry under ambient conditions for approximately two weeks. All subsequent analyses were done on these air-dried sludge samples. pH and conductivity values were determined in suspensions with a relation sludge: distilled water of 1:2.5 after a contact period of 1 h using a WTW InoLab apparatus (pH electrode SenTix 41) and a VWR CO 3100 H SET conductivimeter, respectively.

Dry matter was monitored by a gravimetric method by means of drying in an oven (Holelab) at 105 °C, until constant weight. Calcination at 550 °C for 2 h in a muffle (Nabertherm) was used to determine organic matter. Then, the calcined residue was digested with an HCl 3 N solution and filtered with WhatmanTM 1001 filters, and the filtrate was used to measure the concentrations of phosphorus, calcium and magnesium. P, Ca and Mg levels were evaluated by spectrophotometric and volumetric methods (Prazeres et al., 2019c,d). Chlorides were monitored in sludge extracts obtained using a 0.085 M solution of calcium nitrate in distilled water. After mixing in a horizontal agitator (Memmert) during 15 min, the sludge suspension was filtered and a standardized solution of silver nitrate (0.028 M) was used to titrate the filtrate with potassium chromate as indicator (Prazeres et al., 2013a).

2.4. Statistical analysis

The SAS System statistical software (SAS Institute Inc., Cary NC) was used to treat the experimental results. Data were analyzed using the GLM procedure of SAS, considering each analytical parameter result as main effect. The means of the results were compared in pairs using the Least Significant Difference (LSD) test for a confidence level of 95%, to determine whether they were significantly different. Letters (a to h) were assigned to each mean to identify statistically equal pairs (those with the same letter).

3. Results

3.1. Preliminary screening of chemical treatments

Table 1 presents the physicochemical characterization of the raw cheese manufacturing wastewater. It can be seen that it bears high amounts of phosphorus, COD, turbidity, phenols, total solids and oils and fats. Also, the pH value is below the acceptable range for discharge. So, treatment must be applied in order to recover this wastewater to a quality level obeying legally imposed discharge limits.

The calcium carbonate reagent was used to first adjust the pH of the cheese manufacturing wastewater to the appropriate range (between 7.0 and 8.0), before the addition of the second reagents, which act as both

Table 1
Characterization of the raw wastewater from cheese manufacturing used in the present study, and limit values for discharge in natural waters under Portuguese legislation (Decree-Law 236/98 of 1 August, annex XVIII; Ministério do Ambiente, 1998).

Parameter	Average Value	Unit	Discharge limits
pH at 25 °C	5.120 ± 0.129	Sorensen's scale	6.0–9.0
Electrical conductivity	4.88 ± 0.15	dS/m	–
Abs $_{220\ nm}$	21.767 ± 1.063	cm^{-1} (1:50 dilution)	–
Abs $_{254\ nm}$	2.400 ± 0.224	cm^{-1} (1:50 dilution)	–
Abs $_{410\ nm}$	0.350 ± 0.089	cm^{-1} (1:50 dilution)	–
Abs $_{600\ nm}$	0.242 ± 0.038	cm^{-1} (1:50 dilution)	–
Turbidity	588.07 ± 11.96	NTU	–
COD	8997 ± 507	mg O_2/L	150
Total phosphorus	3055 ± 237	mg P/L	10
Caustic alkalinity	< DL	mg $CaCO_3$/L	–
Hydroxides	< DL	mg $CaCO_3$/L	–
Carbonates	< DL	mg $CaCO_3$/L	–
Total alkalinity	1123 ± 30	mg $CaCO_3$/L	–
Bicarbonates	1123 ± 30	mg $CaCO_3$/L	–
Total hardness	394 ± 13	mg $CaCO_3$/L	–
Calcium hardness	321 ± 8	mg $CaCO_3$/L	–
Calcium	129 ± 3	mg Ca^{2+}/L	–
Magnesium	18 ± 5	mg^{2+}/L	–
Total phenols	74.5 ± 9.8	mg C_6H_5OH/L	0.5
Nitrites	0.398 ± 0.034	mg NO_2^-/L	–
Nitrates	18.052 ± 6.029	mg NO_3^-/L	50
TS	10,798 ± 711	mg/L	–
TFS	2594 ± 211	mg/L	–
TVS	8203 ± 696	mg/L	–
TSS	4210 ± 895	mg/L	60
TDS	6588 ± 964	mg/L	–
Oils and fats	34,217 ± 4862	mg/L	15
Chloride	1204 ± 49	mg Cl^-/L	–

Average values and standard deviations are given. The number of replicate measurements was 5. COD – chemical oxygen demand, TS – total solids, TFS – total fixed solids, TVS – total volatile solids, TSS – total suspended solids, TDS – total dissolved solids, DL – detection limit.

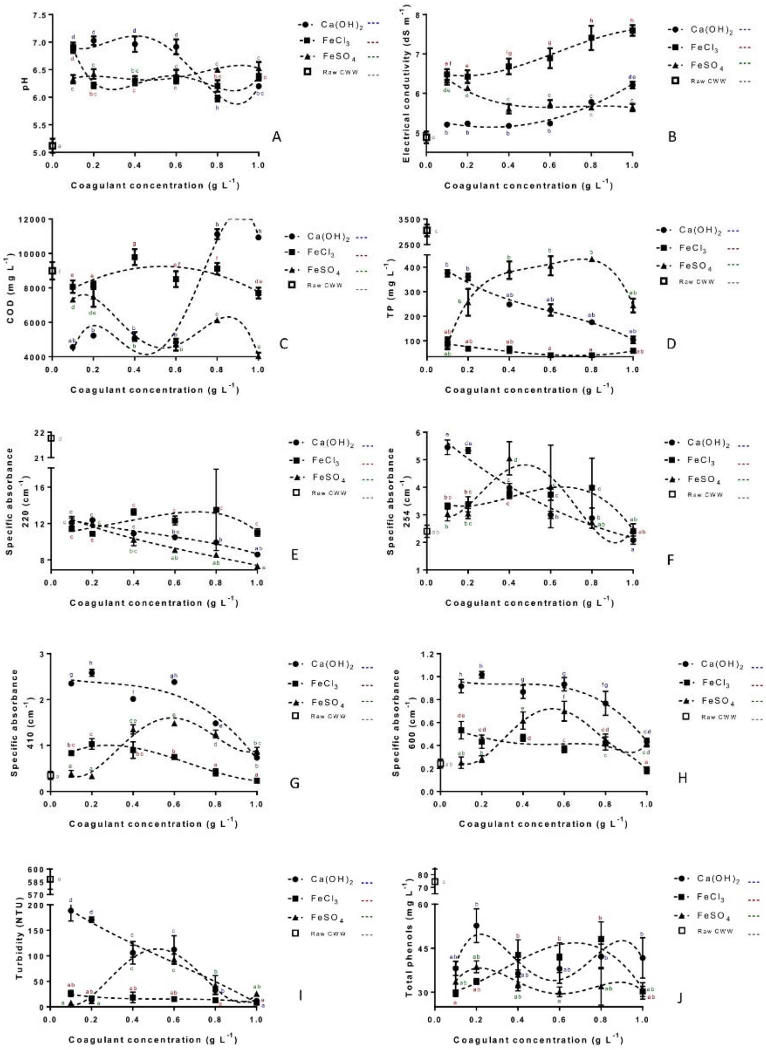


Fig. 1. Effect of the coagulant concentration on the quality parameters analyzed in the supernatants. A: pH; B: Electrical conductivity; C: COD; D: Total phosphorus; E: Specific absorbance at 220 nm; F: Specific absorbance at 254 nm; G: Specific absorbance at 410 nm; H: Specific absorbance at 600 nm; I: Turbidity; J: Total phenols. For each parameter, average and standard deviation (error bars) values are given. The number of replicate measurements was 3. Values with a different letter are significantly different ($P < 0.05$).

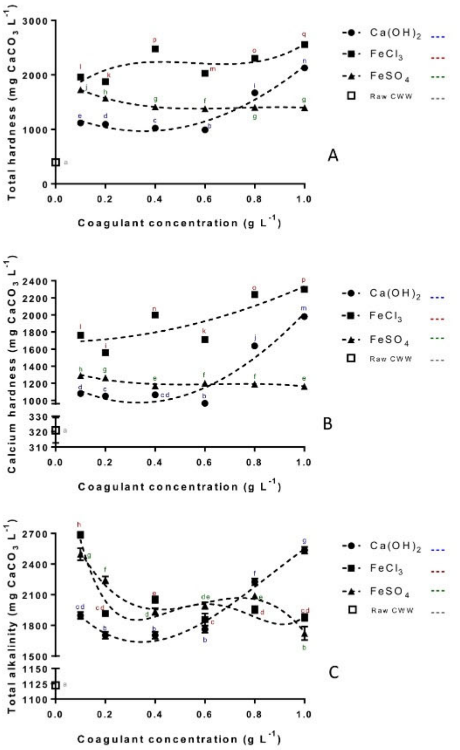


Fig. 2. Effect of the coagulant concentration on the quality parameters analyzed in the supernatants. A: Total hardness; B: Calcium hardness; C: Total alkalinity. For each parameter, average and standard deviation (error bars) values are given. The number of replicate measurements was 3. Values with a different letter are significantly different ($P < 0.05$).

precipitation agents and coagulants. These effects are expected to involve the formation of hydroxides, carbonates and phosphates of calcium, magnesium and iron (Ramalho, 1996; Eckenfelder, 2000; Metcalf and Eddy, 2003; Semerjian and Ayoub, 2003; Renou et al., 2008; Prazeres, 2013), as detailed in the following reactions:

$$CaCO_3 + 2H_2O \rightarrow Ca(OH)_2\downarrow + H_2CO_3 \quad (1)$$

$$2CaCO_3 + 2H_2O \rightarrow Ca(OH)_2\downarrow + Ca(HCO_3)_2 \quad (2)$$

$$MgCO_3 + CaCO_3 + 2H_2O \rightarrow Ca(OH)_2\downarrow + Mg(HCO_3)_2 \quad (3)$$

$$Mg(OH)_2 + CaCO_3 \rightarrow Ca(OH)_2\downarrow + MgCO_3\downarrow \quad (4)$$

$$2FeCl_3.6H_2O + 3Ca(HCO_3)_2 \leftrightharpoons 3CaCl_2 + 2Fe(OH)_3\downarrow + 6CO_2 + 12H_2O \quad (5)$$

$$2FeCl_3 + 3Ca(OH)_2 \rightarrow 3CaCl_2 + 2Fe(OH)_3\downarrow \quad (6)$$

$$Fe^{3+} + PO_4^{3-} \rightarrow FePO_4\downarrow \quad (7)$$

$$FeSO_4 + Ca(OH)_2 \rightleftharpoons Fe(OH)_2\downarrow + CaSO_4\downarrow \quad (8)$$

$$FeSO_4 + Ca(HCO_3)_2 \rightleftharpoons CaSO_4\downarrow + Fe(OH)_2\downarrow + 2CO_2 \quad (9)$$

$$4Fe(OH)_2 + O_2 + 2H_2O \rightleftharpoons 4Fe(OH)_3\downarrow \quad (10)$$

$$3Fe^{2+} + 2PO_4^{3-} \rightarrow Fe_3(PO_4)_2\downarrow \quad (11)$$

$$Ca(OH)_2 + Ca(HCO_3)_2 \rightleftharpoons 2CaCO_3\downarrow + 2H_2O \quad (12)$$

$$Ca(OH)_2 + H_2CO_3 \rightleftharpoons CaCO_3\downarrow + 2H_2O \quad (13)$$

$$5Ca^{2+} + 3PO_4^{3-} + OH^- \rightleftharpoons Ca_5(OH)(PO_4)_3\downarrow \quad (14)$$

$$3Ca(OH)_2 + 2PO_4^{3-} \rightleftharpoons Ca_3(PO_4)_2\downarrow + 6OH^- \quad (15)$$

$$4Ca(OH)_2 + 3PO_4^{3} + H_2O \rightleftharpoons Ca_4H(PO_4)_3\downarrow + 9OH \quad (16)$$

$$10Ca^{2+} + 6PO_4^{3-} + 2OH^- \rightleftharpoons Ca_{10}(OH)_2(PO_4)_6\downarrow \quad (17)$$

Results for the effects of the addition of different concentrations of coagulant on the quality parameters measured in the obtained supernatants are shown in Figs. 1 and 2.

Whatever the reagent or the added dose, there is an increase in the pH value when compared to that of the raw wastewater (Fig. 1A). All treatment tests led to pH values between 6 and 7. In general, for concentrations up to 0.6 g/L, $Ca(OH)_2$ presented a distinct behavior whereas the two iron coagulants showed very similar behavior. For higher concentrations, all coagulants led to a statistically analogous result (Fig. 1A).

For the range of concentrations tested, the electrical conductivity result was different for each reagent (Fig. 1B). While the addition of increasing concentrations of $FeCl_3$ and $Ca(OH)_2$ increased electrical conductivity, the addition of increasing concentrations of $FeSO_4$ led to a decrease in electrical conductivity (Fig. 1B).

At concentrations up to 0.6 g/L, the use of $Ca(OH)_2$ resulted in the best performance in COD removal, removing more than half of the initial load (Fig. 1C). The application of 0.8 and 1.0 g/L of $Ca(OH)_2$ produced effluents with similar, high COD values (Fig. 1C), possibly as a result of a modification in the oxidability of the organic matter in the original wastewater. Considering the use of $FeSO_4$, in general, a decrease of COD was observed with the increase of the concentration used (with the exception of 0.8 g/L), and satisfactory results were obtained, bearing in mind the high load present in this type of effluent (Fig. 1C). For the use of $FeCl_3$, there are no significant changes in the resulting COD values for concentrations in the ranges of 0.1–0.2 g/L and 0.6–1.0 g/L (Fig. 1C). This is possibly related to the nature of the organic matter in the samples, in a way which is difficult to predict. Also, the interference of chloride, remaining in solution, in the COD test could have biased the results.

The best results for total phosphorus removal were obtained when $FeCl_3$ was added, almost independently of the chosen dose, with a total phosphorus removal yield of 98% (Fig. 1D). In the case of $FeSO_4$, the best results (93–98%) were obtained with the extreme dosages (Fig. 1D). With respect to $Ca(OH)_2$, there was a direct relationship between the amount of reagent used and the amount of phosphorus removed, so to obtain higher efficiencies (89–97%) a higher amount of reagent must be added (Fig. 1D). The results suggest that the direct precipitation with trivalent iron is the most efficient total phosphorus removal mechanism. The bell-shaped curve obtained for $FeSO_4$ could have resulted from charge reversal phenomena for intermediate dosages, though it should be noted that the residual total phosphorus concentrations are all in the low range with insignificant differences.

With the addition of $Ca(OH)_2$, the best result (67–73%) for specific absorbance removal at 220 nm was obtained for concentrations of 0.6, 0.8 and 1.0 g/L (Fig. 1E). For $FeCl_3$, results from all treatment tests performed with this reagent are statistically the same (Fig. 1E) (58–66%). The best reagents for removing low molecular weight compounds absorbing at 220 nm were $FeSO_4$ for concentrations of 0.6, 0.8 and 1.0 g/L and $Ca(OH)_2$ for a concentration of 1.0 g/L (Fig. 1E).

For aromatic and unsaturated compounds absorbing at 254 nm, the best removal results (73–77%) were obtained for the concentration of 1.0 g/L irrespective of the coagulant used (Fig. 1F). This concentration led to a distinct result for $Ca(OH)_2$. In the case of $FeCl_3$, 0.1 and 0.2 g/L were equivalent and thus the lower concentration should be used. For $FeSO_4$, 0.1, 0.2 and 0.8 g/L were also alike to $FeSO_4$ 1.0 g/L, so the lower dosage is preferred.

It was observed that none of the treatments tested was effective for removing color compounds absorbing at 410 nm (Fig. 1G). Thus, for $Ca(OH)_2$, the best result was obtained for 1.0 g/L. In the case of $FeCl_3$, the best performances were obtained for concentrations of 0.8 and 1.0 g/L, with a final color value analogous to that of the raw wastewater. For the addition of $FeSO_4$, concentrations of 0.1 and 0.2 g/L were preferable, but giving values also equivalent to that of the raw wastewater.

In relation to optical density measured at 600 nm, for $Ca(OH)_2$ the concentration that led to the best results (86%) was 1.0 g/L with no other statistically alike result (Fig. 1H). In the case of $FeCl_3$, the best concentration was 1.0 g/L but which resulted in no change in comparison to raw wastewater (Fig. 1H). In relation to $FeSO_4$, better results were obtained for the lower concentrations, 0.1 and 0.2 g/L, also resulting in values alike to those of the raw wastewater (Fig. 1H).

The changes in absorbances at 254, 410 nm and 600 nm, are probably affected by changes in the light scattering nature of the aggregates produced by the treatments, particularly for those that are very small in size. Most treatments resulted in the increase of the absorbance values, with respect to that of raw wastewater. This suggests that the treatments, particularly with $Ca(OH)_2$ and $FeSO_4$ produced very small aggregates that were left behind in the supernatant, producing interference in absorbance measurements at the higher wavelengths. The highest dosages may have produced larger aggregates, resulting in the reduction of the absorbance values at 254 and 410 nm. This behavior and the differences between the coagulants are also reflected in the turbidity results (Fig. 1I), though the latter incorporate the removal of both large and small aggregates.

A significant decrease in turbidity of more than 70% was observed in all treatment tests when compared to the raw cheese manufacturing wastewater (Fig. 1I). This is expected due to the promotion of coagulation-flocculation phenomena that mostly act on suspended particles. For the addition of $Ca(OH)_2$, the best concentration was 1.0 g/L with a removal of 98%. For $FeCl_3$, all doses are effective in the removal of turbidity (96–99%). As for $FeSO_4$, concentrations of 0.1, 0.2 and 1.0 g/L are those that produced the best results (96–99%).

Similar trends were obtained for total phosphorus and turbidity, indicating that the residual total phosphorus was probably associated to small precipitate particles which did not settle out.

Good removal levels for phenolic compounds were obtained, ranging from 30% to 62% (Fig. 1J). While the removal yield was always greater than 45%, with the best dosage of 0.6 g/L more than 62% of total phenols could be removed when $FeSO_4$ was applied. In the case of the addition of $Ca(OH)_2$, the best removal yield occurred at the concentration of 0.4 g/L, reaching 49%. In the case of $FeCl_3$, a dose of 0.1 g/L is enough to obtain a 60% removal yield, while the use of higher concentrations decreases this value (except for a concentration of 1.0 g/L).

All the final values for total hardness were higher than those measured in raw wastewater (Fig. 2A). This was mainly due to the first treatment step, the addition of $CaCO_3$. Since a minimum final value of total hardness is desired, with $Ca(OH)_2$ the best result was for a concentration of 0.6 g/L. For $FeCl_3$, the best result was obtained with 0.2 g/L. Finally, for $FeSO_4$, the dose of 0.6 g/L was statistically the most effective.

For the tested coagulants, $Ca(OH)_2$, $FeCl_3$ and $FeSO_4$, the lowest residual calcium hardness values obtained were 0.6, 0.2, 0.4 and 1.0 g/L, respectively (Fig. 2B).

It should be noted that the residual hardness values, both for total and calcium hardness, incorporate the effect of the added reagents themselves and the efficiency of the precipitation process, which removes them from the final supernatant. The balance between these two effects is difficult to predict and may be at the origin, for instance, of the variable results for calcium hardness when $FeCl_3$ was employed.

The total alkalinity value was due exclusively to the residual concentration of bicarbonates (Fig. 2C). As expected, all values were higher than that of the raw wastewater, and this effect was also due to the addition of $CaCO_3$ for pH adjustment. For the addition of $Ca(OH)_2$, the best dosages were 0.2 g/L, 0.4 and 0.6 g/L. In the case of $FeCl_3$, the best dosages were 0.2, 0.6, 0.8 and 1.0 g/L. For $FeSO_4$, the concentration producing the best effect was 1.0 g/L.

To determine the best dosage to be used for each reagent, it was decided to give more importance to some quality parameters since they have a greater relevance in the assessment of the treated wastewater as suitable for discharge in natural waters. These parameters were COD, total phosphorus, turbidity and total phenols. However, a dosage was generally sought that would yield the best results in the maximum possible number of parameters.

Analyzing all the results obtained for $FeCl_3$, it was concluded that the dosage of 1.0 g/L was that which led to the minimum residual load values for more parameters, failing only for pH, electrical conductivity and calcium hardness.

In the case of $FeSO_4$, the dosage that produced the minimum values for more quality parameters was also 1.0 g/L. The failed parameters were specific absorbances at 410 nm and 600 nm, total hardness and

Table 2
Full characterization of the produced sludges from the chosen treatment tests. Value ranges appropriate for sludge application in agricultural soils are also given, compiled from several sources (APA, 2008; Brito et al., 2009; Larney and Hao, 2007; Metcalf, 2003; Soumaré et al., 2002).

Parameter/Unit		$CaCO_3$ 75 g/L + $FeCl_3$ 1.0 g/L		$CaCO_3$ 75 g/L + $FeSO_4$ 1.0 g/L		$CaCO_3$ 75 g/L + $Ca(OH)_2$ 0.6 g/L		Agriculture application ranges
pH at 25 °C	Sorensen's scale	7.150 ± 0.088	a	7.501 ± 0.048	b	7.712 ± 0.038	c	5.5–8.5
Electrical conductivity	µS/cm	1778 ± 157	b	1592 ± 258	ab	1243 ± 196	a	<3000
Wet weight	g/L	114.8416 ± 1.7459	b	100.5443 ± 4.5600	a	108.5407 ± 8.0861	ab	–
Dry weight (DW)	g/L	72.6568 ± 0.7372	b	67.1860 ± 1.4561	a	72.3496 ± 1.0052	b	–
Wet volume	mL/L	150 ± 0	a	146 ± 7	a	163 ± 7	b	–
Dry matter (DM)	g/g DW	0.9870 ± 0.0069	a	0.9958 ± 0.0009	b	0.9979 ± 0.0006	c	<0.2
Organic matter dry matter basis	g/g DM	0.0688 ± 0.0022	b	0.0383 ± 0.0062	a	0.0271 ± 0.0056	a	0.09–0.35
Organic matter dry weight basis	g/g DW	0.0679 ± 0.0021	b	0.0381 ± 0.0061	a	0.0270 ± 0.0056	a	–
Total phosphorus	mg P/g DM	54 ± 2	b	58 ± 1	c	21 ± 1	a	–
Calcium	mg Ca^{2+}/g DM	257 ± 1	b	254 ± 1	a	262 ± 0	c	–
Magnesium	mg^{2+}/g DM	5 ± 0	a	5 ± 0	a	7 ± 0	b	–
Chloride	mg Cl^-/g DM	5 ± 0	c	4 ± 0	b	2 ± 0	a	–

For each parameter, average and standard deviation values are given. The number of replicate measurements was 3. The wet weight, dry weight and wet volume values are given per L of treated wastewater sample. In each line, values with different letters are significantly different ($P < 0.05$).

calcium hardness, but also in this case the results are not unfavorable to the point of leading to the overall rejection of this dosage value.

Finally, for $Ca(OH)_2$, no single dosage was the most effective for the most important parameters so the choice had to be between 0.6 g/L and 1.0 g/L. Although the value of total phenols for the dose of 1.0 g/L did not show a great discrepancy from the best one, the COD value was excessively high. The 0.6 g/L dose led to the minimum of all the most important parameters except turbidity. Considering all the results, 0.6 g/L was chosen as the best dosage for $Ca(OH)_2$. This dosage also did not produce the minimum values of specific absorbances at 254, 410 and 600 nm but the results were not very different from the best.

To make these choices, it was not possible to apply the lowest concentration criterion to minimize costs in the purchase of reagents, since the results found for lower concentrations were not satisfactory enough to be considered useful.

So, the best dosages from this screening study were established as: $CaCO_3$ 75 g/L + $FeCl_3$ 1.0 g/L; $CaCO_3$ 75 g/L + $FeSO_4$ 1.0 g/L, and $CaCO_3$ 75 g/L + $Ca(OH)_2$ 0.6 g/L.

3.2. *Detailed examination of the products from the selected treatments*

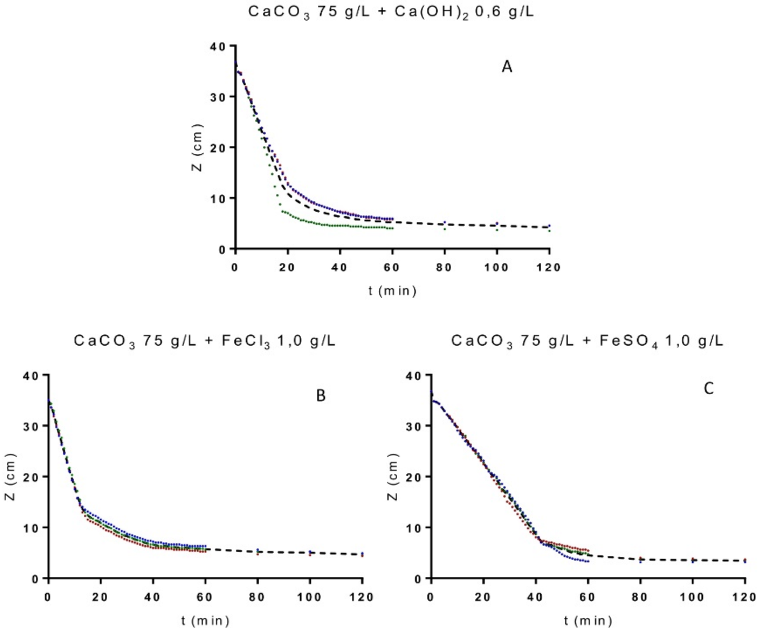
3.2.1. *Residual sludge*

Full characterization of the sludges produced in the treatment conditions selected in the screening study of section 3.1 is shown in Table 2.

Regarding pH, the treatment test that employed $FeCl_3$ resulted in the lowest value, with the highest being obtained in the $Ca(OH)_2$ test, as expected. However, for electrical conductivity, the highest result was found for the test with $FeCl_3$ and the lowest for that with $Ca(OH)_2$. This is probably because, at higher pH values, a higher fraction of inorganic matter tends to precipitate, leaving less ions in solution and thus decreasing the electrical conductivity of the wet sludge. A direct relationship could also be noted between conductivity and salinity of the generated sludge, since the test treatments associated with highest and lowest electrical conductivity also had the highest and lowest levels of chloride ions, respectively, in the sludge composition.

The wet weight, dry weight and wet volume values for the three sludges were very similar, indicating that the treatments led to similar mass recovery and sludge density. Also, the dry matter (DM) content of the air-dried sludges was very high, with all values close to 1, indicating that the two weeks of drying under ambient conditions were effective in removing most of the sludges' moisture. The organic matter fraction (both on a dry matter and a dry weight basis) was less than 7% for all treatments, reflecting the mostly inorganic nature of the precipitates. However, the assay with $FeCl_3$ led to almost twice the recovery of organic matter in the sludge, when compared with the other two tests.

In relation to macro-nutrients, high amounts of calcium are present in the sludge from all the selected treatment trials, with concentrations in the range of 250–265 mg/g DM. The assay giving the highest content was the $CaCO_3$ + $Ca(OH)_2$ process as expected since it has a double source of calcium. The high calcium concentration in all sludges is due to the high amount of calcium carbonate used in all treatment trials which precipitated along with other pollutants. In the case of total phosphorus, the obtained concentrations are also significant but much lower than those of calcium, varying in the range of 20–60 mg/g DM. The presence of phosphorus in the sludge in this concentration range can be explained by the precipitation phenomena that accumulated phosphate compounds in the sludge leaving only residual levels in the supernatant. For chloride and magnesium, low concentrations were obtained, in the same


$CaCO_3$ 75 g/L + $Ca(OH)_2$ 0,6 g/L

$CaCO_3$ 75 g/L + $FeCl_3$ 1,0 g/L

$CaCO_3$ 75 g/L + $FeSO_4$ 1,0 g/L

Fig. 3. Sedimentation curves obtained for the chosen treatment conditions. The number of replicate measurements was 3.

Table 3
Full characterization of the supernatants from the chosen treatment tests.

Parameter/Unit		$CaCO_3$ 75 g/L + $FeCl_3$ 1.0 g/L		$CaCO_3$ 75 g/L + $FeSO_4$ 1.0 g/L		$CaCO_3$ 75 g/L + $Ca(OH)_2$ 0.6 g/L	
pH at 25 °C	Sorensen' scale	6.636 ± 0.057	a	6.530 ± 0.111	a	6.951 ± 0.137	b
Electrical conductivity	dS/m	7.60 ± 0.14	c	5.64 ± 0.09	b	5.23 ± 0.05	a
Abs $_{220\ nm}$	cm^{-1} (1:50 dilution)	11.017 ± 0.404	c	7.317 ± 0.058	a	10.483 ± 0.029	b
Abs $_{254\ nm}$	cm^{-1} (1:50 dilution)	2.400 ± 0.278	a	2.500 ± 0.173	a	3.000 ± 0.132	b
Abs $_{410\ nm}$	cm^{-1} (1:50 dilution)	0.233 ± 0.029	a	0.883 ± 0.076	b	2.383 ± 0.553	c
Abs $_{600\ nm}$	cm^{-1} (1:50 dilution)	0.183 ± 0.029	a	0.417 ± 0.029	b	0.933 ± 0.058	c
Turbidity	NTU	8.12 ± 1.73	a	25.47 ± 0.53	b	112.34 ± 27.28	c
COD	mg O_2/L	7706 ± 317	c	4039 ± 210	a	4844 ± 201	b
Total phosphorus	mg P/L	59 ± 8	a	244 ± 28	b	226 ± 23	b
Total alkalinity	mg $CaCO_3$/L	1877 ± 34	b	1723 ± 67	a	1761 ± 34	a
Total hardness	mg $CaCO_3$/L	2557 ± 9	c	1398 ± 9	b	990 ± 9	a
Calcium hardness	mg $CaCO_3$/L	2299 ± 15	c	1164 ± 0	b	965 ± 9	a
Total phenols	mg C_6H_5OH/L	30.3 ± 1.7	a	30.4 ± 2.8	ab	38.0 ± 5.0	b
Nitrites	mg NO_2^-/L	0.083 ± 0.007	a	0.115 ± 0.003	b	0.159 ± 0.005	c
Nitrates	mg NO_3^-/L	0.235 ± 0.048	a	1.033 ± 0.109	b	1.746 ± 0.066	c
TS	mg/L	7360 ± 295	b	5292 ± 168	a	5120 ± 135	a
TFS	mg/L	4087 ± 62	c	3175 ± 139	b	2822 ± 108	a
TVS	mg/L	3273 ± 340	b	2117 ± 235	a	2298 ± 239	a
TSS	mg/L	60 ± 9	a	68 ± 8	ab	85 ± 15	b
TDS	mg/L	7300 ± 299	b	5223 ± 171	a	5035 ± 144	a
Oils and fats	mg/L	629 ± 152	b	949 ± 44	c	381 ± 69	a
Chloride	mg Cl^-/L	1826 ± 10	b	1078 ± 9	a	1090 ± 5	a

For each parameter, average and standard deviation values are given. The number of replicate measurements was 3. In each line, values with a different letter are significantly different ($P < 0.05$). COD – chemical oxygen demand, TS – total solids, TFS – total fixed solids, TVS – total volatile solids, TSS – total suspended solids, TDS – total dissolved solids.

order of magnitude. The higher chloride concentration observed in the $CaCO_3 + FeCl_3$ treatment was expected, due to the introduction of extra chloride.

Another important feature to deal with in precipitation-coagulation-flocculation processes is the settling properties of the sludge. The determination of the sedimentation characteristics of a heterogeneous suspension is of great importance for a correct sizing of industrial settlers. The sedimentation tests performed for each of the coagulants used showed the same phenomenon in sedimentation (Fig. 3), namely, the particles formed a blanket that settled as a coherent mass presenting a distinct interface between it and the supernatant liquid phase (type III sedimentation). As seen in Fig. 3, the sedimentation curves presented a high velocity trend in the clarification zone, low velocity in the transition zone and velocity close to 0 in the compaction zone. The highest mean settling velocity was 1.6 cm/min for $FeCl_3$, followed by 1.3 cm/min for $Ca(OH)_2$ and, least of all, 0.7 cm/min for $FeSO_4$. Also, the solids critical concentrations were calculated, corresponding to a sharp transition in settling velocity marking the onset of settled sludge thickening. The lowest critical concentration (as TS) was 10.8 g/L for $FeCl_3$, followed by 15.4 g/L for $Ca(OH)_2$ and 22.5 g/L for $FeSO_4$. There was an inverse relationship between the average settling velocity and the critical concentration. This means that the suspension from the assay with $FeSO_4$ settled slowest, but at the critical point, the concentration was highest.

3.2.2. *Supernatant*

Full characterization of the supernatants from each of the treatment conditions selected in the screening study of section 3.1 is shown in Table 3.

The treatment with $FeCl_3$ allowed removal levels of 14.4% for chemical oxygen demand, 98.1% for total phosphorus, 98.6% for turbidity, 59.3% for total phenols, 98.7% for nitrates, 79.1% for nitrites, 31.8% for total solids and 98.2% for oils and fats. The treatment with $FeSO_4$ exhibited removal levels of 55.1% for chemical oxygen demand, 92.0% for total phosphorus, 95.7% for turbidity, 59.1% for total phenols, 94.3% for nitrates, 71.0% for nitrites, 51.0% for total solids and 97.2% for oils and fats. The treatment using $Ca(OH)_2$ allowed removal levels of 46.2% for chemical oxygen demand, 92.6% for total phosphorus, 80.9% for turbidity, 49.0% for total phenols, 90.3% for nitrates, 60.1% for nitrites, 52.6% for total solids and 98.9% for oils and fats. In the wastewater treated with the combination of $CaCO_3 + FeCl_3$, the value of TDS increased because of the increased calcium and chloride contents, with respect to that of the raw wastewater. Approximate mass balances to calcium and total phosphorus using the average values of the involved parameters given in Tables 1–3 are given in Tables S1 and S2, respectively, of the supplementary material. The mass unbalances reflect analytical uncertainties and the difficulties in representative sampling of the sludge fraction. However, it can be inferred that most of the input Ca and P indeed end up in the sludge after the treatment procedures.

Analyzing the results, the best treatment was that which used $FeSO_4$, with 11 parameters presenting the statistically lowest values (pH, characteristic absorbances at 220 and 254 nm, COD, total alkalinity, total phenols, TS, TVS, TSS, TDS and chlorides).

The results for the treatments with $FeCl_3$ and $Ca(OH)_2$ were rated as equal, since both presented the statistically lowest values for 10 quality parameters (for the $FeCl_3$ tests: pH, characteristic absorbances at 220, 254, 410 and 600 nm, turbidity, total phosphorus, total phenols, nitrates, nitrites and SST; for the $Ca(OH)_2$ tests: electrical conductivity, total alkalinity, total hardness, calcium hardness, TS, TFS, TVS, TDS, oils and fats and chlorides).

To choose the best reagent, it was again decided to give more importance to some of the measured quality parameters, those which have a greater contribution to legal water quality standards, considering the discharge in natural water bodies. These parameters were COD, total phosphorus, total phenols, TSS and oils and fats. Nitrates were excluded since it was noted that the raw wastewater is already within the legal discharge limits with respect to this parameter. Taking this into consideration, it can be concluded that none of the reagents lead simultaneously to the minimum values of all the relevant parameters. While $FeCl_3$ and $FeSO_4$ provided the minimum for 3 of them, $Ca(OH)_2$ only provided the minimum for 1 parameter, excluding it immediately from the choice of the best reagent.

In conclusion, it is considered that the treatment with $FeSO_4$ is the most efficient for the wastewater used for this study, even though it leaves a higher concentration of oils and fats in the treated supernatant than the other two alternatives. If necessary, this treatment can be followed by another (e.g., flotation) to specifically remove the fatty pollutant load.

The results obtained in this study can be compared with those of a previous study (unpublished results) with the isolated addition of $CaCO_3$ to another batch of cheese manufacturing wastewater from the same producer, which resulted in removal levels of 36.3% for chemical oxygen demand, 83.8% for total phosphorus, 73.3% for turbidity and 45.5% for total phenols. Whatever the combination of reagents, the present experiments all gave higher removal results than the isolated addition of $CaCO_3$, considering the same quality parameters, except for the $FeCl_3$ process with respect to the COD parameter. This significant increase of the removal levels for the analyzed quality parameters proves the efficacy and justifies the use of the second reagent in this type of treatment, instead of using only $CaCO_3$.

4. Discussion

4.1. Potential of the produced sludges for agricultural valorization

Considering a possible agricultural valorization of the generated sludges, the quality results obtained for them in the treatment trials, given in Table 2, were compared with the appropriate ranges (APA, 2008; Brito et al., 2009; Larney and Hao, 2007; Metcalf and Eddy, 2003; Soumaré et al., 2002), which are also given in Table 2. Analyzing the pH values, all the sludges were within the appropriate values (5.5–8.5). The electrical conductivity values from all trials were also within the appropriate limit (less than 3 dS/m or 3000 μS/cm). Ensuring adequate pH and conductivity values in the sludge is of the highest importance as it prevents the degradation of soils with irreversible changes in their physical and chemical properties.

The dry matter contents of all the generated sludges are close to 1 g/g of dry sludge, reflecting the circumstance that they went through the drying process under ambient conditions for two weeks, as described in section 3.2. This value is much higher than those mentioned in the literature for wet sludges (less than 0.2 g dry matter/g of sludge). The fact that the dry matter is high does not invalidate the possibility of these sludges being used in agricultural applications, since the addition of water can be done if necessary. Moreover, the drying process, if it can be done at production scale, favors sludge stability during storage and transport.

In the case of organic matter on a dry basis, a different value was obtained for each treatment test. It is noted that the sludges from all the treatment trials had organic matter contents below the recommended range (0.09–0.35 g/g of dry matter), however these results do not exclude their possible agricultural valorization.

The concentrations of the analyzed nutrients and minerals showed different ranges among the obtained sludges. Overall the concentrations of calcium and phosphorus were significant while those of magnesium and chloride were low. There is no recommended range for the concentration of any of these compounds since the required dose depends on the needs of the recipient plant culture itself.

Overall, the sludges generated in the selected treatment trials were within the appropriate specifications, under the quality parameters considered, and their use in agricultural valorization is a hypothesis to be studied in more detail.

4.2. Disposal possibilities for the treated wastewater supernatants

Very high removal yields were obtained for almost all quality parameters in the best treatment trials (Table 3). However, to put them in context, the legal discharge limits in natural water bodies are here considered (given in Table 1), as well as the results of similar studies previously reported in the literature.

To examine the possibility of treated wastewater discharge, it is necessary to consider the legally imposed discharge limits for the treated effluent, which are very tight for discharge in natural water bodies, according to Decree-Law 236/98 of 1 August, annex XVIII (Ministério do Ambiente, 1998, Portugal) (see Table 1). Considering the characteristics of the raw wastewater in what concerns the regulated quality parameters, to guarantee legal discharge the treatment would require pollutant removal yields higher than 99.99%, which is often not possible with a single treatment step. Comparing the results obtained for the three treatment trials with the discharge limits in Table 1, it is concluded that only pH (within 6.0–9.0) and nitrates (under 50 mg/L) are within the appropriate ranges.

In the assay with $FeCl_3$ combined with $CaCO_3$, the value found for TSS was 60 mg/L, equal to the discharge limit, while the other treatments led to values of 68 and 85 mg/L, for $FeSO_4$ and $Ca(OH)_2$, respectively. Thus, a subsequent treatment capable of removing suspended solids would have to be applied in all cases, due to the possible variations of the treatment in removal efficiency and considering the standard deviation of the obtained results.

The COD emission limit value is 150 mg/L, and in this study it was possible to reduce the pollutant load from 9000 mg/L to 7710, 4040 and 4840 mg/L in the treatment trials with $FeCl_3$, $FeSO_4$ and $Ca(OH)_2$, respectively. Considering that organic matter is one of the most difficult to remove pollutants in wastewaters from cheese manufacturing (Carvalho et al., 2013), the best result obtained led to more than 50% reduction in this particular load. However, as far as the discharge limit is concerned, the treated wastewater has yet to be subjected to a post-treatment which significantly reduces the COD content, possibly a biological process.

The total phosphorus emission limit is 10 mg/L, and in this study concentrations of 59, 244 and 226 mg/L were obtained for the treatment trials with $FeCl_3$, $FeSO_4$ and $Ca(OH)_2$, respectively. Considering the value found in the raw wastewater, 3060 mg/L, removal yields were very high but not enough.

The emission limit value for total phenols is 0.5 mg/L. The introduction of $FeCl_3$, $FeSO_4$ and $Ca(OH)_2$ combined with $CaCO_3$ caused a decrease in the concentration of this pollutant from 74.5 mg/L to 30.3, 30.4, and 38.0 mg/L, respectively. Although the removal yields were in all three cases greater than 50%, these values are still far from the maximum allowable value.

In the case of oils and fats, the legally permissible emission limit value is 15 mg/L. In this study, removal yields of more than 97% were reached, reducing oils and fats from 34,200 mg/L to 629, 949 and 381 mg/L in the treatment trials with $FeCl_3$, $FeSO_4$ and $Ca(OH)_2$, respectively. Considering that the oils and fats present in this type of effluent is one of the most worrisome contaminants in water quality (Prazeres et al., 2012), the results obtained were very good. However, as far as the limit is concerned, the treated wastewater has yet to be subjected to a post-treatment which significantly reduces the content of oils and fats.

In conclusion, the tested effluent is among the most complex matrices found in dairy industry wastewaters. Thus, it is very difficult to apply only one treatment process that can reduce the concentrations of the various pollutants to the point of allowing the direct discharge into natural water bodies. It should additionally be noted that the concentration of these same pollutants can vary widely within cheese manufacturing effluents (Prazeres, 2013), whereby the application of the same treatment in the raw wastewater of a different cheese manufacturing facility will likely not lead to the same results.

No other studies were found in the literature on the treatment of wastewaters from the cheese manufacturing industry using the same combination of reagents used in this study. Thus similar studies were considered for comparison, involving the isolated use of the coagulants here tested, at different dosages, in the treatment of wastewater from dairies, all excluding the effect of the addition of $CaCO_3$.

As reported by Rivas et al. (2010), treatment trials were carried out on wastewater from cheese manufacturing with the introduction of only one reagent. After studying the effect of the addition of various concentrations of $FeCl_3$ and $FeSO_4$ from 0.1 to 2.0 g/L, it was concluded that the best results for both reagents were obtained at the concentration of 0.25 g/L. For the best condition of $FeCl_3$ addition, Rivas et al. (2010) obtained removal yields of 31.9% for COD, 71.9% for turbidity, 14.3% for total phosphorus, 20.8% for total solids and −5.5% for oils and fats. Comparing these results with those of the present study, the high potential of the treatment with $CaCO_3$ 75 g/L + $FeCl_3$ 1.0 g/L becomes evident. In the case of Rivas et al. (2010) with $FeSO_4$ addition, the best results obtained were a 42.9% removal level for COD, 96.8% for turbidity, 88.6% for total phosphorus, −16.3% for total solids and 97.2% for oils and fats. Again, there is a superior potential in the treatment of the present study with $CaCO_3$ 75 g/L + $FeCl_3$ 1.0 g/L.

In another report, Rivas et al. (2011) carried out treatment trials on wastewater from cheese manufacturing with the isolated addition of various quantities of $Ca(OH)_2$, bringing the pH of the raw effluent to values of 10.5–13.0. The best results were obtained for pH 11.0 which corresponded to an added $Ca(OH)_2$ concentration of 4.5 g/L. Compared to the $CaCO_3$ 75 g/L + $Ca(OH)_2$ 0.6 g/L test performed in the present study, the results obtained are similar, except for total solids, higher removal yields being obtained in the present study.

Thus, it can be confirmed that the treatment trials applied in this study show an improvement in relation to previously studied processes, so that their implementation as effective pre-treatments for wastewater from cheese manufacturing is a subject possibly to be studied in the future.

From the economical point of view and taking into account the price of reagents, the combination of $CaCO_3$ + $FeSO_4$ presents operating costs similar to those mentioned by Bayramoglu et al. (2006) when managing poultry slaughterhouse wastewater by electrocoagulation with an aluminum electrode.

5. Conclusions

The raw cheese manufacturing wastewater used in this study confirmed its problematic nature, posing severe environmental and public health problems due to its acidic pH and high contents in phosphorus, COD, turbidity, total phenols, total solids, oils and fats and minerals.

The treatment options examined in the present study aimed to answer the challenge to find efficient alternatives for the removal of these pollutant loads, keeping in mind the volume and characteristics of the effluent produced, the technologies available and the costs associated with their implementation and maintenance. Thus, the applied treatments produced effluents with pH values between 6 and 7. However, increases in the conductivity values were observed when compared to raw wastewater. $CaCO_3$ + $Ca(OH)_2$ showed the best results for COD removal (≥50%). Additionally, all the treatments were effective in the removal of total phosphorus, turbidity and total phenols.

Considering the statistical significance of the results and the relevance of the different parameters defining the quality of the treated wastewater, it was possible to conclude that the treatment with $CaCO_3$ 75 g/L + $FeSO_4$ 1.0 g/L was the most efficient. For this best-case scenario, the treatment allowed removal levels of 55.1% for chemical oxygen demand, 92.0% for total phosphorus, 95.7% for turbidity, 59.1% for total phenols, 94.3% for nitrates, 71.0% for nitrites, 51.0% for total solids and 97.2% for oils and fats. The results obtained are still far from producing a treated effluent quality adequate for discharge into natural water courses, but they compared very favorably with results previously reported from the application of a similar treatment strategy to the same type of effluent.

Considering the results obtained, the deigned precipitation-coagulation-flocculation process can be economical and easy to apply as a pre-treatment for a biological, anaerobic post-treatment process, since the reduction of the organic load could increase the stability of reactor operation, reduce the necessary hydraulic retention time and minimize the flotation of oils and fats. Alternatively, the treated wastewaters can be sources of organic matter and nutrients for agricultural irrigation.

Overall, the sludges generated in the selected treatment conditions were within the appropriate specifications, in what concerns the quality parameters relevant for agricultural valorization. Thus, their use as fertilizer and/or soil corrective is also a hypothesis to be studied in more detail.

Author contributions

Conceptualization, A.R. Prazeres, R.C.S. Guerreiro, S. Luz; methodology, formal analysis and investigation, R.C.S. Guerreiro, S. Luz, A.R. Prazeres, H.M. Pinheiro and E. Jerónimo; writing—original draft preparation, R.C.S. Guerreiro; writing—review and editing, A.R. Prazeres, H. M. Pinheiro and E. Jerónimo; supervision, A.R. Prazeres and H.M. Pinheiro; funding acquisition, A.R. Prazeres.

Declaration of competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jenvman.2020.110470.

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CAPÍTULO IV – TRATAMENTO DE ÁGUA RESIDUAL DE QUEIJARIA POR PROCESSO DE PRECIPITAÇÃO EM MEIO ÁCIDO

4.1. Processo de precipitação em meio ácido

Neste trabalho foi desenvolvida 1 patente, 1 artigo e 1 comunicação.

Patente 1 - Process of treatment of agroindustrial waste by reaction of acidification with addition of strong acids, European Patent application Nº EP 3447030B1

Artigo 3 - Cheese wastewater treatment by acid and basic precipitation: Application of H_2SO_4, HNO_3, HCl, $Ca(OH)_2$ and NaOH

Comunicação 1 - Aplicação de processos de precipitação ácida para o tratamento de águas residuais de adega

4.1.1. Patente 1 - Process of treatment of agroindustrial waste by reaction of acidification with addition of strong acids, European Patent application Nº EP 3447030B1

O processo patenteado (EP 3447030B1) consiste num processo de precipitação através da adição de soluções concentradas de ácidos fortes (preferencialmente, ácido sulfúrico, clorídrico ou nítrico) às águas residuais brutas (sem qualquer tratamento prévio) até à obtenção de pH inferior ou igual a 6, em condições de agitação, com três tipos de velocidade sequencial decrescente, nomeadamente, rápida, média e lenta, durante pelo menos 3 minutos no total:

- A primeira etapa consiste na aplicação dos ácidos fortes (H_2SO_4, HCl e HNO_3) ao efluente bruto, à temperatura e luz ambiente, em agitação forte (1200 – 1400 rpm) durante 1 minuto, permitindo assim a homogeneização e fomentando a reação entre o ácido e a água de modo a promover o aumento da turvação;

- Na segunda etapa a agitação é reduzida para uma velocidade média (700 – 800 rpm), durante 1 minuto;

- Na terceira etapa, reduz-se a intensidade de agitação, para uma velocidade lenta (300 – 400 rpm), por mais 1 minuto, de forma a permitir o incremento da turvação da água.

Esta agitação sequencial conduz à formação de aglomerados de partículas sedimentáveis por gravidade, em condições ambientais, durante 2 horas, formando duas fases: sobrenadante ou água tratada e precipitado ou lama. O volume de lama obtido varia ente 12,5-100 mL por cada litro de efluente tratado.

Este sistema permitiu alcançar um efeito vantajoso inesperado: reduzir significantemente a contaminação de matéria orgânica, incluindo a matéria orgânica dissolvida, monitorizada pela avaliação da carência química de oxigénio (CQO dissolvida) em aproximadamente 76% e o CQO total em cerca de 72 %, o que não aconteceu com a aplicação de outros sistemas de agitação. A formação deste precipitado de pH ácido, pode ser atribuída ao deslocamento de complexos solúveis (óxidos ou hidróxidos insolúveis), ou pela presença de compostos (químicos húmicos) de solubilidade variável (eq. 18), podendo também conter espécies metálicas anfotéricas, (eq. 19) (Riva *et al.,* 2004), explicadas nas reações:

$$M^{n+}\,(L)_m^{(m-n)-} + (m-n)H^+ \xrightarrow{H2O} M(OH)_n\downarrow + (m-n)LH \tag{18}$$

$$M^{n+}\,(OH)_m^{(m-n)-} + (m-n)H^+ \rightarrow M(OH)_n\downarrow + (m-n)H_2O \tag{19}$$

Os precipitados obtidos apresentam cerca de 90% de matéria seca, sendo também ricos em nutrientes como cálcio, magnésio, fósforo, cloreto, sódio e potássio. Estes nutrientes presentes no precipitado podem ser recuperados e reutilizados, por exemplo, como fertilizante na agricultura devido ao elevado teor em matéria orgânica e nutrientes.

4.1.2- Artigo 3 - Cheese wastewater treatment by acid and basic precipitation: Application of H_2SO_4, HNO_3, HCl, $Ca(OH)_2$ and NaOH

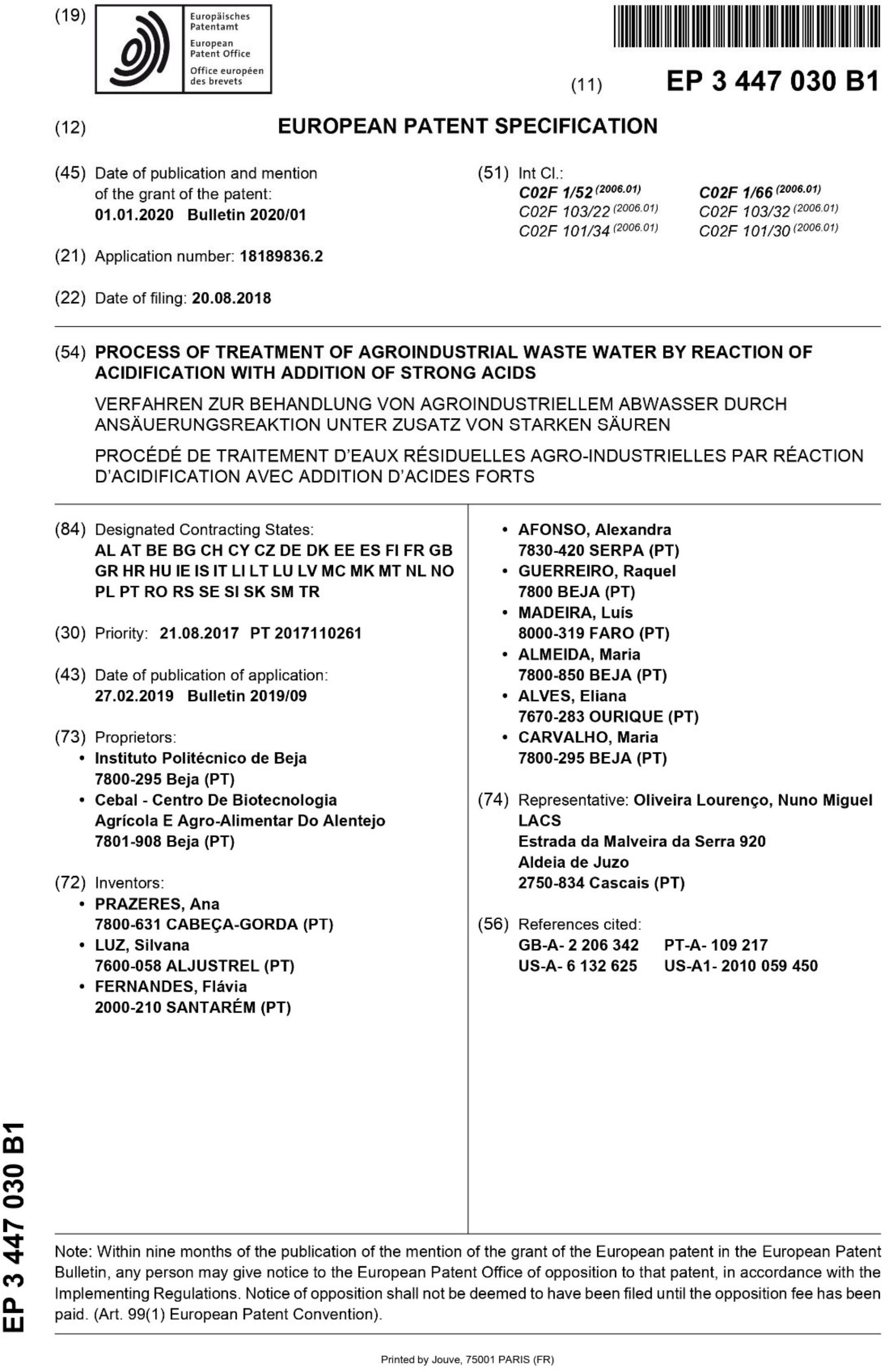
O processo de precipitação em meio ácido foi testado no efluente de queijaria, utilizando os ácidos H_2SO_4, HNO_3, HCl, aos pH 1,0; 2,0; 3,0 e 4,0, através do processo patenteado acima descrito. Este processo foi também aplicado no processo de precipitação em meio básico, com os reagentes $Ca(OH)_2$ e NaOH, aos pH de 6,0; 7,0; 8,0; 9,0; 10,0; 11,0 e 12,0.

A precipitação em meio ácido não obtiveram fracas remoções de matéria orgânica, em todos os reagentes estudados, no entanto, foi eficaz na remoção de turvação, com valores superiores a 50%, em fenóis totais, com remoções superiores a 30% e o P_{total} obteve remoções entre os 26 – 57%, em todos os reagentes. As melhores remoções da precipitação em meio ácido foram encontradas ao pH 3,0 com HCl, e remoção de 90,3% de turvação, 50% de fenóis totais, 37,2% de fósforo total e 5% de matéria orgânica.

A precipitação em meio básico, foi mais eficazes nas remoções de matéria orgânica, apresentando remoções até 44%, as remoções de turvação chegaram a atingir os 99%, o fósforo atingiu os 96% e os fenóis totais os 66%, para ambos os reagentes. A reação que obteve melhores teores de remoção de contaminação do efluente foi a precipitação em meio básico utilizando o reagente $Ca(OH)_2$ a pH 11,0, com remoção de 30% de matéria orgânica (CQO), 92,4% de P_{total}, 76% de turvação e 40% de fenóis totais.

A tecnologia da Patente 1 foi também aplicada no efluente de adega (**Comunicação 1**), com o reagente HCl, aos pH de tratamento de 1,0; 2,0; 3,0; 4,0; 5,0 e 6,0, onde foram obtidas remoções de matéria orgânica na ordem dos 32 – 37%, turvação superior a 88% e remoções superiores a 50% em fósforo e fenóis totais. Revelando uma melhor eficácia de remoção face ao efluente de queijaria.

Patente 1 - Process of treatment of agroindustrial waste by reaction of acidification with addition of strong acids. European Patent application Nº EP 3447030B1



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(54) **PROCESS OF TREATMENT OF AGROINDUSTRIAL WASTE WATER BY REACTION OF ACIDIFICATION WITH ADDITION OF STRONG ACIDS**

VERFAHREN ZUR BEHANDLUNG VON AGROINDUSTRIELLEM ABWASSER DURCH ANSÄUERUNGSREAKTION UNTER ZUSATZ VON STARKEN SÄUREN

PROCÉDÉ DE TRAITEMENT D'EAUX RÉSIDUELLES AGRO-INDUSTRIELLES PAR RÉACTION D'ACIDIFICATION AVEC ADDITION D'ACIDES FORTS

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Description

Technical domain of the invention

[0001] The present invention relates to a process for the treatment of agro-industrial waste water, preferably from slaughterhouses, olive oil presses, cheese industries or wine producing facilities by reaction of acidification with the addition of strong acids, selected from sulfuric, hydrochloric or nitric acid.

[0002] The application of this process to treatment waste water reduces the level of contamination in terms of organic matter, including dissolved organic matter, monitored by chemical oxygen demand (total and dissolved COD), chemicals such as phosphorus, calcium and magnesium, turbidity, total phenols and characteristic absorbances.

[0003] Accordingly, the present invention is in the technical field of treatment of effluents from agro-food industries.

Background of the invention

[0004] Agro-industrial wastewater treatment is a difficult task because of its level of contamination in terms of organic matter, suspended solids, oils and fats, and chemical elements. In addition, these effluents can have variable pH values and high levels of salinity.

[0005] In this regard, agro-industrial effluents can lead to serious environmental and public health problems, such as soil contamination, eutrophication phenomena, rapid depletion of oxygen in surface waters, and groundwater contamination.

[0006] The management of agro-industrial wastewater has occurred through application to the soil, mixing with other less contaminated effluents, and storage and evaporation in ponds.

[0007] Some physico-chemical processes have also been applied to reduce the level of contamination, such as: dissolved air flotation, coagulation-flocculation, basic precipitation, membrane technologies, advanced oxidation and electrochemical processes.

[0008] The article "Prazeres et al., 2011. Cheese whey wastewater treatment by acidic precipitation. Proceedings of the XIV World Water Congress, International Water Resources Association and Water and Energy Resources Secretariat of Pernambuco State, September, Porto de Galinhas-PE, Brazil." discloses a process by acid precipitation using H_2SO_4 at a pH value of 1 to 3 for treating wastewater originated out of dairy farms. These waters produce sludge through flocculent sedimentation and the removal of organic matter, suspended solids, turbidity, absorbances and chemicals is observed. In this article, optimum conditions are achieved only at extreme pH conditions (pH = 1.0) and the process is carried out in two steps: the addition of H_2SO_4 at pH values ranging from 1 to 3 under conditions similar to coagulation-flocculation, and sedimentation of the precipitate. However, the removal of the contaminants as described above by this process is not sufficiently effective, which removes only 21% of COD at conditions of pH = 1.0.

[0009] The article "Babel and Kurniawan (2004). Cr (VI) removal from synthetic wastewater using coconut shell charcoal and commercial activated carbon modified with oxidizing agents and / or chitosan. Chemosphere 54 (7), 951-967." discloses the chemical modification of the surface of coconut shell and commercial activated charcoal through the use of chitosan and / or oxidizing agents such as sulfuric acid and nitric acid. Modifications were performed as follows: washing of surfaces with ultrapure water for leaching of impurities, treatment with oxidizing agent (2% H_2SO_4 and 65% for HNO_3) in an incubator at 110°C during a large period of time (24 h for H_2SO_4 and 3h for HNO_3), cooling and drainage of the acid solution, washing with ultrapure water until pH stabilization, residual acid removal by use of a $NaHCO_3$ (2%) solution, oven drying overnight at 110°C, cooling to room temperature and desiccator storage. Also this process does not remove the relevant compounds, such as organic matter (especially dissolved organic matter), chemicals such as phosphorus, calcium and magnesium, turbidity, total phenols and characteristic absorbances.

[0010] The documents US2010/0059450, US6132625 and GB2206342 are further examples of known processes for the treatment of wastewater from agro-industries, in particular by acidification, flocculation and/or precipitation.

[0011] There is a great need to develop and implement wastewater treatment processes from agro-industries, which remove contaminants efficiently, quickly and inexpensively, especially with regard to the removal of dissolved organic matter.

[0012] The process of the present invention proposes to solve this problem by adding strong acids to the wastewaters to be treated.

Summary of the invention

[0013] The present invention relates to a process for the treatment of waste water from agro-industrial facilities, in particular from slaughterhouses, olive oil presses, cheese industries or wine producing facilities by reaction of acidification with the addition of strong acids under agitation conditions, and precipitation, according to claim 1.

[0014] The application of this process to agro-industrial wastewater reduces its level of contamination in terms of organic matter, including dissolved organic matter, monitored by chemical oxygen demand (total and dissolved COD), chemicals such as phosphorus, calcium and magnesium, turbidity, total phenols and characteristic absorbances of this type of wastewater.

[0015] In addition, the resulting sludge may be reused, for example, as a fertilizer in agriculture because of the high content of organic matter and nutrients.

General description of the invention

[0016] The present invention relates to a process for the treatment of waste water from agro-industrial facilities, in particular slaughterhouses, olive oil presses, cheese industries or wine producing facilities, by reaction of acidification processes with addition of strong acids, selected from sulfuric acid, hydrochloric acid or nitric acid.

[0017] The addition of concentrated solutions of strong acids to raw agro-industrial wastewater, i.e. without any previous treatment, achieving a pH value of 1 to 6 under stirring conditions is obtained with three types of decreasing sequential velocity, namely, fast, medium and slow, for at least 3 minutes in total under ambient conditions, leads to the formation of precipitate of particle agglomerates. After sedimentation for a minimum time of about 2 hours, the phases can be then separated.

[0018] The proposed treatment process reduces the organic load, including dissolved organic matter, and chemicals from raw agro-industrial wastewater, reducing COD, including dissolved COD, phosphorus, calcium, magnesium, turbidity, total phenols and characteristic absorbances.

[0019] During the application of this process there is also the formation of precipitates, also called sludge, with different physicochemical characteristics according to the type of water, process applied and operating conditions. These precipitates comprise recoverable or reusable compounds such as organic matter and nutrients which may be used, for example, in agriculture.

[0020] The proposed process of treatment has several benefits over conventional processes, which are listed below:

- It can be applied to raw waste water, i.e. it is not necessary to pre-treat the waste water in order to apply the process;
- It is easy to apply and control, adapting itself to the physical-chemical and biological properties of the raw waste water;
- It is applied under ambient conditions, i.e. at ambient pressure and temperature with presence of light and atmospheric air, thus not requiring manipulation of the environment in which the treatment takes place;
- It can be used as pre-treatment of physiochemical processes, such as membrane technologies, which present high costs and problems of fouling when used in raw wastewater, since it removes suspended solids;
- The reactions resulting from the addition of strong acids and agitation systems are very rapid, occurring for times less than or equal to 3 minutes, making the general process also very fast;
- It is easily implemented on an industrial scale, without the need for specialized manpower or sophisticated installations;
- The sedimentation stage is fast with a time of about 2 hours and occurs due to gravity, which reduces costs;
- The reduction of contamination in terms of total and dissolved COD, chemicals such as phosphorus, calcium and magnesium, turbidity, total phenols and characteristic absorbances is very significant and efficient;
- Treated wastewater and precipitates obtained can be used for agricultural activities or subjected to membrane processes;
- The volume and amount of moisture of the generated precipitates can be reduced by inexpensive and economical physical processes, such as by centrifugation and drying in sand filters;
- The strong acids used in this process are readily available and often used in the chemical industry.

Description of the figures

[0021]

Figure 1. Efficiency of the process of the invention applied to the treatment of wastewater from slaughterhouse, with addition of H_2SO_4 at different pH values, wherein:

Fig. 1A represents pH and conductivity,
Fig. 1B represents Ca^{2+}, Mg^{2+}, OH^-, CO_3^{2-} and HCO_3^-,
Fig. 1C represents total phenols and total phosphorus,
Fig. 1D represents absorbances at 220, 254, 410 and 600 nm,
Fig. 1E represents CQO and turbidity, and
Fig. 1F represents the volume of sludge.

Figure 2. Efficiency of the process of the invention applied to the treatment of wastewater from slaughterhouse, with addition of HCl at different pH values, wherein:

Fig. 2A represents pH and conductivity,
Fig. 2B represents Ca^{2+}, Mg^{2+}, OH^-, CO_3^{2-} and HCO_3^-,
Fig. 2C represents total phenols and total phosphorus,
Fig. 2D represents absorbances at 220, 254, 410 and 600 nm,
Fig. 2E represents CQO and turbidity, and
Fig. 2F represents the volume of sludge.

Figure 3. Efficiency of the process of the invention applied to the treatment of wastewater from slaughterhouse, with addition of HNO_3 at different pH values, wherein:

Fig. 3A represents pH and conductivity,
Fig. 3B represents Ca^{2+}, Mg^{2+}, OH^-, CO_3^{2-} and HCO_3^-,
Fig. 3C represents total phenols and total phosphorus,
Fig. 3D represents absorbances at 220, 254, 410 and 600 nm,
Fig. 3E represents CQO and turbidity, and
Fig. 3F represents the volume of sludge.

Figure 4. Efficiency of the process of the invention applied to the treatment of wastewater from wine producing facilities with addition of H_2SO_4 at different pH values, wherein:

Fig. 4A represents pH and conductivity,
Fig. 4B represents HCO_3^- and OH^-,
Fig. 4C represents total phenols and total phosphorus (TP),
Fig. 4D represents absorbances at 220, 254, 410 and 600 nm,
Fig. 4E represents CQO and turbidity, and,
Fig. 4F represents Ca^{2+} and Mg^{2+}.

Figure 5. Efficiency of the process of the invention applied to the treatment of wastewater from wine producing facilities with addition of HCl at different pH values, wherein:

Fig. 5A represents pH and conductivity,
Fig. 5B represents HCO_3^- and OH^-,
Fig. 5C represents total phenols and total phosphorus (TP),
Fig. 5D represents absorbances at 220, 254, 410 and 600 nm,
Fig. 5E represents CQO and turbidity, and
Fig. 5F represents Ca^{2+} and Mg^{2+}.

Figure 6. Efficiency of the process of the invention applied to the treatment of wastewater from wine producing facilities with addition of HNO_3 at different pH values, wherein:

Fig. 6A represents pH and conductivity,
Fig. 6B represents HCO_3^- and OH^-,
Fig. 6C represents total phenols and total phosphorus (TP),
Fig. 6D represents absorbances at 220, 254, 410 and 600 nm,
Fig. 6E represents CQO and turbidity, and
Fig. 6F represents Ca^{2+} and Mg^{2+}.

Figure 7. Characterization of the effluent treated by the process of the invention applied to the treatment of waste water from cheese industries with addition of H_2SO_4 at different pH values, wherein:

Fig. 7A represents pH and conductivity,
Fig. 7B represents CQO, total phosphorus (TP), turbidity and total phenols,
Fig. 7C represents Ca^{2+}, Mg^{2+}, CO_3^{2-}, HCO_3^- and OH^-, and
Fig. 7D represents absorbances at 220, 254, 410 and 600 nm.

Figure 8. Characterization of the effluent treated by the process of the invention applied to the treatment of waste water from cheese industries with addition of HCl at different pH values, wherein:

Fig. 8A represents pH and conductivity,
Fig. 8B represents CQO, total phosphorus (TP), turbidity and total phenols,
Fig. 8C represents Ca^{2+}, Mg^{2+}, CO_3^{2-}, HCO_3^- and OH^-, and
Fig. 8D represents absorbances at 220, 254, 410 and 600 nm.

Figure 9. Characterization of the effluent treated by the process of the invention applied to the treatment of waste water from cheese industries with addition of HNO_3 at different pH values, wherein:

Fig. 9A represents pH and conductivity,
Fig. 9B represents CQO, total phosphorus (TP), turbidity and total phenols,
Fig. 9C represents Ca^{2+}, Mg^{2+}, CO_3^{2-}, HCO_3^- and OH^-, and
Fig. 9D represents absorbances at 220, 254, 410 and 600 nm.

Figure 10. Characterization of the effluent treated by the process of the invention applied to the treatment of waste water from olive oil presses with addition of H_2SO_4 at different pH values, wherein:

Fig. 10A represents pH and conductivity,
Fig. 10B represents CQO, total phosphorus (TP), turbidity and total phenols,
Fig. 10C represents Ca^{2+}, Mg^{2+}, CO_3^{2-}, HCO_3^- and OH^-, and
Fig. 10D represents absorbances at 220, 254, 410 and 600 nm.

Figure 11. Characterization of the effluent treated by the process of the invention applied to the treatment of waste water from olive oil presses with addition of HCl at different pH values, wherein:

Fig. 11A represents pH and conductivity,
Fig. 11B represents CQO, total phosphorus (TP), turbidity and total phenols,
Fig. 11C represents Ca^{2+}, Mg^{2+}, CO_3^{2-}, HCO_3^- and OH^-, and
Fig. 11D represents absorbances at 220, 254, 410 and 600 nm.

Figure 12. Characterization of the effluent treated by the process of the invention applied to the treatment of waste water from olive oil presses with addition of HNO_3 at different pH values, wherein:

Fig. 12A represents pH and conductivity,
Fig. 12B represents CQO, total phosphorus (TP), turbidity and total phenols,
Fig. 12C represents Ca^{2+}, Mg^{2+}, CO_3^{2-}, HCO_3^- and OH^-, and
Fig. 12D represents absorbances at 220, 254, 410 and 600 nm.

Figure 13. Process of treatment of agro-industrial waste water, according to the present invention, wherein:

1. Represents a concentrated solution of strong acids, preferably sulfuric, hydrochloric or nitric acid;
2. Represents the addition of concentrated solutions of strong acids to a pH value equal or less than 6 to raw waste water from slaughterhouses, olive oil presses, cheese industries or wine producing facilities, with rapid or strong agitation (1200-1400 rpm) at ambient pressure and at room temperature in presence of light and atmospheric air producing particle agglomerates;
3. Represents mixing the waste water and concentrated solution of strong acids subjected to a rapid or strong stirring system of 1200-1400 rpm for about 1 minute, with formation of larger particle agglomerates monitored by water turbidity;
4. Represents mixing the waste water and concentrated solution of strong acids subjected to a medium velocity stirring system of 700-800 rpm for about 1 minute, with formation of larger particle agglomerates monitored by water turbidity;
5. Represents mixing the waste water and concentrated solution of strong acids subjected to a low velocity stirring system of 300-400 rpm for about 1 minute, with formation of larger particle agglomerates monitored by water turbidity;
6. Represents the sedimentation of the particle agglomerates formed, for about 2 hours, defining two phases: supernatant or treated water and precipitate or sludge;

7. Represents the separation of the phases formed in 6.

Detailed description of the invention

[0022] The present invention relates to a process for the treatment of agro-industrial waste water, preferably from slaughterhouses, olive oil presses, cheese industries or wine producing facilities by reaction of acidification with the addition of strong acids, selected from sulfuric, hydrochloric or nitric acid.

[0023] In the scope of the present invention, the term "strong acid" means "an acid which is completely ionised in aqueous solution at constant temperature and pressure, under such conditions having a concentration equal to the concentration of hydrogen ions with a dissociation constant or acidity constant pKa of less than 2.

[0024] These acids are added to the water to be treated in aqueous solution of different concentrations in order to achieve solutions with pH values of 6 or less, so that the mixing solution has a pH in the range of 1 to 6, preferably in the range of 3 to 6, more preferably in the range of 4 to 6.

[0025] The addition of any of these strong acids to the waters to be treated is carried out under agitation conditions, in particular under agitation conditions at three different speeds and in a sequential decreasing order:

(a) Initially a high-speed stirring is promoted, varying from 1200 to 1400 rpm, for about 1 minute, to allow the homogenization and reaction between the acid and the wastewater with increasing turbidity of the water;
(b) In a second step, the stirring speed is reduced to a medium speed, ranging from 700 to 800 rpm, for about 1 minute to allow increasing the turbidity of the wastewater; and finally
(c) In a third step, the stirring speed is reduced for a slow stirring velocity, ranging from 300-400 rpm, for about 1 minute to allow increasing the turbidity of the wastewater.

[0026] Thereafter, a sedimentation step is promoted for about 2 hours. After sedimentation, the phases are ready to be separated.

[0027] The application of the different stages in this process, as described above, for the treatment of waste water originated in agro-industries, reduces the level of contamination in terms of organic matter, including dissolved organic matter, monitored by chemical oxygen demand (COD total and dissolved), chemicals such as phosphorus, calcium and magnesium, turbidity, total phenols and the characteristic absorbances of this type of waste water.

[0028] Analysis of several wastewater samples treated with the process of the present invention has shown that the reduction of the level of contamination in terms of organic matter monitored by the total COD (maximum value = 97.7%) and dissolved can be achieved (maximum value = 98.3%), calcium (maximum value = 100%), magnesium (maximum value = 73%), turbidity (maximum value = 100%), total phenols maximum = 99.3%) and characteristic absorbances (220, 254, 410 and 600 nm) (maximum value = 100%).

[0029] In addition, it results in the formation of precipitates rich in organic matter and nutrients, which can be recovered and reused for example in agriculture.

[0030] In short, the process of the present invention, for application in the treatment of waste water from agro-industries, comprises the following steps:

a) Acidification, by the addition of a solution of an acid with pKa equal or less than 2, to a determined volume of waste water to be treated until a pH value of the mixing solution is of 1 to 6, being the added acid selected from sulfuric acid, hydrochloric or nitric acid, under stirring conditions, which takes place in three phases, each with a kind of decreasing speed, wherein:

- in the first stage, the stirring velocity ranges from approximately 1200 to 1400 rpm and lasts about 1 minute,
- in the second stage, the stirring velocity ranges from approximately 700 to 800 rpm and lasts for about 1 minute, and
- in the third and last agitation stage, the velocity ranges from approximately 300-400 rpm, for about 1 minute,

b) Sedimentation of the resulting solution in (a), the sludge and supernatant, during at least about 2 hours.

Examples

Example 1. Physico-chemical characteristics of raw waste water

[0031] The wastewater analysed was collected from several agro-industry facilities located in the Baixo Alentejo area of Portugal. Table 1 presents the most relevant values that characterize the waste water from slaughterhouse, olive oil presses, cheese industries or wine producing facilities, and this analysis was done in untreated water, also called raw waste water.

Table 1. Physico-chemical characteristics of raw waste water from agro-industries

Parameter	Unit	Cheese	Olive oil	Slaughter	Winery
pH	Sorensen scale	4.52±0.205	4.63±0.142	6.89±0.237	6.80±0.056
Conductivity	dS m^{-1}	5.23±0.277	2.69±0.133	2.96±0.109	1.62±0.064
Turbidity	NTU	594.9±49.54	280.1±2.63	797.8±156.19	320.8±145.06
COD	mg L^{-1}	4917±353.6	7910±330.7	4660±1040.3	2003±423.3
Dissolved COD	mg L-1	-	-	972±207.9	917±136.1
Phosphorus	mg L^{-1}	3411.8±1255.64	1049.7±84.74	676.5±90.71	210.1±24.34
Calcium	mg L^{-1}	236.2±28.94	210.6±42.55	144.9±21.33	110.8±12.86
Magnesium	mg L^{-1}	109.5±36.77	73.6±15.86	54.7±19.61	55.4±14.79
Total alkalinity	mg L^{-1} CaCO$_3$	1339.7±251.14	2039.2±344.81	884.5±96.11	700.7±48.06
Bicarbonates	mg L^{-1} CaCO$_3$	1339.7±251.14	2039.2±344.81	884.5±96.11	700.7±48.06
Total Hardness	mg L^{-1} CaCO$_3$	1039.7±168.69	970.5±470.86	586.4±120.58	504.3±49.90
Calcium hardness	mg L^{-1} CaCO$_3$	589.3±72.19	525.5±106.16	361.5±53.22	276.5±32.08
Magnesium hardness	mg L^{-1} CaCO$_3$	450.4±151.30	445.0±435.54	224.9 80.72	227.8±60.83
Total phenols	mg L^{-1}	75.9±6.76	540.4±50.42	34.8±10.19	27.8±3.01
Absorbances					
220 nm	cm^{-1}	0.639±0.0383 (1:50)	1.027±0.0994 (1:50)	0.184±0.0473 (1:50)	0.143±0.0397 (1:50)
254 nm	cm^{-1}	0.180±0.0147 (1:50)	0.366±0.0637 (1:50)	0.093±0.0315 (1:50)	0.093±0.0317 (1:50)
410 nm	cm^{-1}	0.092±0.0065 (1:50)	0.076±0.0140 (1:50)	0.047±0.0152 (1:50)	0.033±0.0161 (1:50)
600 nm	cm^{-1}	0.060±0.0072 (1:50)	0.031±0.0054 (1:50)	0.054±0.0198 (1:25)	0.067±0.0059 (1:20)
COD = chemical oxygen demand (total or dissolved)					

Example 2. Effect of the application of different agitation systems in the treatment of waste water from slaughterhouses

[0032] Analysis were performed where different agitation systems were applied to the process of treatment of wastewater from slaughterhouses. All studies were conducted under the same conditions except for the stirring conditions, namely the velocity of stirring.

[0033] Strong acid solutions were added to the waste water from slaughterers until a pH value of approximately 6 in the mixing solution was achieved under the agitation conditions described in Table 2. The analysis were conducted at room temperature and pressure and in the presence of atmospheric air and light.

[0034] After interruption of the stirring systems, sedimentation of the agglomerates of the particles was verified for a time of at least to 2 hours and finally the two phases resulting phases were separated. The processes were monitored by total and dissolved COD measurement.

[0035] Table 2 shows the process conditions with the application of 5 different stirring systems, in which:

A. Inventive Example:

[0036] The process of the present invention comprising applying three sequential stirring systems (1200-1400 rpm min^{-1}, 700-800 rpm min^{-1} and 300-400 rpm min^{-1}) for about 1 minute each;

B. Comparative Example:

[0037] A process comprising applying a conventional high velocity agitation system (1200-1400 rpm min-1) for about 3 minutes;

C. Comparative Example:

[0038] A process comprising the application of two stirring systems, the first at high velocity and the second the intermediate velocity (1200-1400 rpm min^{-1} and 700-800 rpm min^{-1}), for about 1.5 minutes each, totalling about 3 minutes of stirring.

D. Comparative Example:

[0039] A process comprising applying a conventional stirring system at an intermediate velocity (700-800 rpm min^{-1}) for about 3 minutes;

E. Comparative Example:

[0040] A process comprising applying a conventional low velocity agitation system (300-400 rpm min^{-1}) for about 3 minutes.

Table 2. Effects of the application of different agitation systems in wastewater from slaughterhouse in acidification processes with strong acids

Ex.	Veloc. (rpm)	Time (min)	COD (mgL^{-1})	COD DIS (mgL^{-1})	COD (remov. %)	COD DIS (remov.%)
A INV	1200-1400 700-800 300-400	1 1 1	250	201	93,6	79,3
B COMP	1200-1400	3	2361	472	39,7	51,4
C COMP	1200-1400 700-800	1,5 1,5	3583	650	8,5	33,1
D COMP	700-800	3	850	517	78,3	46,8
E COMP	300-400	3	583	550	85,1	43,4
COD DIS = COD dissolved Remov.% = % of COD removed						

[0041] As shown in Table 2, the application of the different agitation systems results in different degrees of efficiency of removal of contaminants in wastewater from slaughterhouses.

[0042] Thus, the system of the present invention has an unexpected advantageous effect of significantly reducing the organic matter, especially the dissolved organic matter, i.e. significant reduction of the dissolved COD by about 79% and the total COD by about 94%, which did not happen with the application of other agitation systems.

Example 3. Effect of the application of different agitation systems in the treatment of waste water from wineries

[0043] Studies were carried out in which different agitation systems were applied to the process of wastewater treatment from wineries. All the studies were carried out under the same conditions as those mentioned in Example 2, except for the origin of the waste water.

[0044] In the same way 5 different stirring systems were tested, where A, B, C, D and E correspond to systems identical to those mentioned in Example 2, i.e. A corresponds to one of the embodiments of the invention (Inv. = Inventive Example) and the remainder to Comparative Examples (COMP).

Table 3. Effects of the application of different agitation systems in wastewater from wineries in acidification processes with strong acids.

Ex.	Veloc. (rpm)	Time (min)	COD (mgL^{-1})	COD DIS (mgL^{-1})	COD (remov. %)	COD DIS (remov.%)
A INV	1200-1400 700-800 300-400	1 1 1	423	217	71,8	76,3
B COMP	1200-1400	3	350	317	76,7	65,4
C COMP	1200-1400 700-800	1,5 1,5	454	350	69,7	61,8
D COMP	700-800	3	450	450	70,0	50,9
E COMP	300-400	3	397	400	73,5	56,4

COD DIS = COD dissolved
Remov.% = % of COD removed

[0045] As in the previous example, also in the case of the application of the process of the invention to the treatment of waste water from wineries, a substantial reduction of the contamination has occurred.

[0046] It was observed that a higher amount of COD dissolved was removed with the application of three sequential stirring systems (1200-1400 rpm min^{-1}, 700-800 rpm min^{-1} and 300-400 rpm min^{-1}) according to the present invention in comparison to other agitation conventional systems, i.e. the application of three agitation systems in the acidification processes with strong acids had an unexpected effect of acting more efficiently on the dissolved COD than the other agitation systems.

Example 4. Application of acidification processes with strong acids in four raw waste waters

[0047] The proposed processes reduce the level of contamination in terms of characteristic absorbances, turbidity, COD, phosphorus, calcium, magnesium and total phenols.

[0048] Figure 1, 2 and 3 show examples of the application of these processes to raw agro-industrial wastewater from slaughterhouse under different pH conditions (values of approximately 1, 2, 3, 4, 5 and 6) and with addition of different types of acids: H_2SO_4, HCl or HNO_3.

[0049] The application of the proposed processes (example: H_2SO_4, pH = 5.0) in the raw waste water from the slaughterhouse allowed the reduction of the following contamination indicators: COD (92.5%), phosphorus (34.1%), turbidity (91.1%), total phenols (48.1%) and characteristic absorbances (69.4-80.9%).

[0050] The application of another embodiment of the present invention, for example with a pH value of the mixed solution of about 6.0 by adding HCl to the raw wastewater from slaughterhouse, allowed the following contamination indicators to be reduced: COD (79.1%), phosphorus (52.1%), calcium (20.3%), turbidity (95.9%), total phenols (69.1%) and characteristic absorbances (46.9-95.2 %).

[0051] In still another embodiment of the present invention (example: HNO_3, pH = 4.0) in the raw wastewater from slaughterhouse, the following contamination indicators were reduced: COD (94.3%), phosphorus (56.2%), calcium (33.4%), turbidity (98.8%), total phenols (92.2%) and characteristic absorbances (76.1-90.7%).

[0052] Figures 4, 5 and 6 show examples of the application of these treatment processes to raw agro-industrial wastewater from the wineries at different pH conditions (1, 2, 3, 4, 5 and 6) and with different types of acids (H_2SO_4, HCl and HNO_3) .

[0053] The application of another embodiment of the present invention (example: H_2SO_4, pH = 5.0) to the waste water from wineries allowed the reduction of COD (41.8%), phosphorus (68.1%), calcium , (9%), turbidity (93.5%), total phenols (67.8%) and characteristic absorbances (46.7-74.0%).

[0054] The application of another embodiment of the present invention (example: HCl, pH = 6.0) to the waste water from wineries allowed the reduction of COD (36.8%), phosphorus (56.0%), calcium, turbidity (90.5%), total phenols (66.8%) and characteristic absorbances (47.7-74.8%).

[0055] The application of another embodiment of the present invention (example: HNO_3, pH = 5.0) to the waste water from the wineries allowed the reduction of COD (32.6%), phosphorus (68.1%), calcium (1%), turbidity (94.4%), total phenols (71.9%) and characteristic absorbances (56.0-76.0%).

[0056] Figure 7, 8 and 9 show examples of the application of the process of the present invention to raw agro-industrial wastewater from cheese industries under different pH conditions (values of approximately 1, 2, 3 and 4) and with different types of acids (H_2SO_4, HCl and HNO_3) .

[0057] The application of another embodiment of the present invention (example: H_2SO_4, pH = 3.0) to the waste water from cheese industries allowed the reduction of phosphorus (57.0%), magnesium (14.1%), turbidity (5%), total phenols (44.1%) and characteristic absorbances (22.6-55.4%).

[0058] The application of another embodiment of the present invention (example: HCl, pH = 2.0) to the waste water from cheese industries allowed the reduction of phosphorus (40.9%), calcium (55.4%), turbidity (6%), total phenols (38.8%) and characteristic absorbances (23.8-71.7%).

[0059] The application of another embodiment of the present invention (example: HNO_3, pH = 4.0) to the waste water from cheese industries allowed the reduction of phosphorus (28.0%), calcium (17.2%), turbidity (9%), total phenols (31.9%) and characteristic absorbances (32.9-71.6%).

[0060] Figures 10, 11 and 12 show examples of the application of various embodiments of the process of the present invention to raw agro-industrial waste water from olive oil presses at different pH conditions (values of approximately 1, 2 and 3) and with addition of different types of acids (H_2SO_4, HCl and HNO_3).

[0061] The application of another embodiment of the present invention (example: H_2SO_4, pH = 1.0) to the waste water from olive oil presses allowed the reduction of phosphorus (26.9%), calcium (100%) and characteristic absorbances (10.9-94.2%).

[0062] The application of the process of the invention (example: HCl, pH = 3.0) to the waste water from olive oil presses allowed the reduction of phosphorus (22.1%), calcium (100%), turbidity (87.8%) and absorbances characteristics (20.7-47.4%).

[0063] The application of the process of the invention (example: HNO_3, pH = 3.0) to the wastewater from olive oil presses allowed a reduction of COD (23.6%), turbidity (92.8%), total phenols (14.4%) and characteristic absorbances (59.1-91.4%).

Example 5. Physiochemical properties of waste water treated by acid addition with strong acid addition

[0064] The characteristics of the treated wastewater obtained in acidification processes with the addition of strong acids were monitored through the following parameters: pH, electrical conductivity, characteristic absorbances (220, 254, 410 and 600 nm), turbidity, COD, total hardness and calcium hardness, calcium, magnesium, phosphorus, total and methyl orange alkalinity, carbonates, bicarbonates, hydroxides and total phenols.

[0065] Table 4 summarizes examples of physicochemical characteristics of treated wastewater obtained in the acidification processes with strong acids applied to raw agro-industrial waste water.

[0066] The application of strong acids to the raw effluents originated waste water treated with acidic properties. Additionally, there is a slight increase in the level of salinity monitored by the electrical conductivity in the treated wastewater, in relation to the raw waste water.

[0067] The treated wastewater had a COD content of less than 4683 mg L^{-1}, being rich in chemicals such as phosphorus, calcium and magnesium.

[0068] The alkalinity present is in the form of bicarbonates. The hardness of the water is mainly due to the presence of calcium and magnesium. On the other hand, treated wastewater presents reduced content of total phenols compared to raw wastewater.

Table 4. Physiochemical characteristics of treated waste water obtained in acidification processes with strong acids

Parameter	Unit	Cheese	Slaughter	Wineries
pH	Sorensen scale	2.99±0.045	5.48±0.000	5.30±0.066
Conductivity	dS m^{-1}	6.15±0.007	3.08±0.000	1.75±0.003
Turbidity	NTU	57.9±7.42	70.8±0.68	17.98±3.615
COD	mg L^{-1}	4683±0.00	350±47.1	1350±94.3
Dissolved COD	mg L^{-1}	-	201+17.5	217+47.1
Phosphorus	mg L^{-1}	2143.1±41.59	445.5±11.93	67.1±8.95
Calcium	mg L^{-1}	353.5±10.64	245.0±19.73	79.7±0.00

(continued)

Parameter	Unit	Cheese	Slaughter	Wineries
Magnesium	mg L^{-1}	77.5±0.00	136.6±11.97	111.2±0.00
Total alkalinity	mg $L^{-1}CaCO_3$	0.0±0.00	459.5	459.5±0.00
Bicarbonates	mg $L^{-1}CaCO_3$	0.0±0.00	459.5	459.5±0.00
Total Hardness	mg $L^{-1}CaCO_3$	1201.1±26.54	1173.4±0.00	656.5±0.00
Calcium hardness	mg $L^{-1}CaCO_3$	882.0±26.54	611.3±49.29	198.9±0.00
Magnesium hardness	mg $L^{-1}CaCO_3$	319.0±0.00	562.0±49.24	457.5±0.00
Total phenols	mg L^{-1}	39.4±0.95	0.01±0.009	7.8±1.36
Absorbances				
Abs 220 nm	cm^{-1}	0.462±0.0184 (1:50)	0.056±0.0057 (1:50)	0.691±0.0728 (1:50)
Abs 254 nm	cm^{-1}	0.093±0.0021 (1:50)	0.027±0.0021 (1:50)	0.041±0.0042 (1:50)
Abs 410 nm	cm^{-1}	0.063±0.0057 (1:50)	0.011±0.0007 (1:50)	0.0130±0.0014 (1:50)
Abs 600 nm	cm^{-1}	0.047±0.0064 (1:50)	0.257±0.0014 (1:1)	0.007±0.0007 (1:50)
COD - Chemical oxygen demand				

Example 6. Formation of precipitates with reusable and recoverable compounds

[0069] The proposed physiochemical treatment originated the formation of precipitates with different characteristics according to the type of water, applied process and operational conditions.

[0070] Table 5 shows some examples of the volume generated during the application of strong acid acidification processes. After sedimentation, 12.5 to 120 mL of precipitate per litre of effluent was obtained.

[0071] The volume of precipitate can be reduced by economical and accessible physicochemical processes, such as centrifugation and drying in sand filters, is presented in Table 5.

Table 5. Volume of precipitate obtained in acidification processes with strong acids

Parameter	Units	Volume
Cheese	mL L^{-1} effluent	25-110
Slaughter	mL L^{-1} effluent	37.5-120
Olive oil	mL L^{-1} effluent	25-62.5
Wineries	mL L^{-1} effluent	12.5-100

[0072] Table 6 shows examples of the characteristics presented by the precipitates obtained during the acidification processes with strong acids. The obtained precipitates present about 90% of dry matter and organic matter, being also rich in chemicals such as calcium, magnesium, phosphorus, chloride, sodium and potassium. These elements can be recovered and reused, as nutrients for example, in agriculture.

Table 6. Characteristics of the precipitates obtained in acidification processes with strong acids in slaughterhouse wastewater.

Parameter	Units	HCl	H_2SO_4	HNO_3
Sludge volume	mL L^{-1}	45.0±4.36	59.2±1.44	40.3±8.39
Weight of centrifuged sludge	g L^{-1}	30.6±2.67	29.6±2.16	33.2±7.26
Dry weight	g L^{-1}	3.8±0.11	3.9±0.30	3.5±0.69
Dry matter	%	91.5±0.53	91.6±0.33	91.7±0.33

(continued)

Parameter	Units	HCl	H_2SO_4	HNO_3
Organic matter (dry sample)	%	86.2±0.13	85.2±0.18	85.9±0.10
Organic matter (wet sample)	%	79.0±0.57	78.0±0.44	78.8±0.30
Calcium	g kg^{-1}	77.8±25.57	160.0±41.02	135.9±9.67
Magnesium	g kg^{-1}	605.9±106.70	287.6±60.69	339.5±91.85
Phosphorus	g kg^{-1}	<DL	0.74±0.472	0.98±0.219
Chloride	g kg^{-1}	25.1±0.02	16.8±2.08	21.0±1.04
Sodium	g kg^{-1}	1.53±0.451	2.01±0.812	2.94±0.603
Potassium	g kg^{-1}	0.28±0.065	0.38±0.153	0.50±0.077

[0073] Figure 13 represents the schematic of the physicochemical treatment of agro-industrial waste water by acidification processes with addition of strong acids and formation of precipitates with reusable and recoverable compounds.

Claims

1. Process for the treatment of wastewater from agro-industries, comprising the following steps:

    (a) An acidification step performed by the addition of an aqueous solution of acid selected from sulfuric acid, hydrochloric acid or nitric acid having a *pKa* value of less than 2, to a determined volume of wastewater to be treated until the pH value of the mixed solution achieved is of 1 to 6, and wherein the acid is added under stirring conditions, which takes place in three phases, each of said phases with a decreasing rate of velocity, as the following:

    - In the 1st phase, the stirring velocity varies from 1200 to 1400 rpm, during approximately 1 minute;
    - In the 2nd phase, the stirring velocity varies from 700 to 800 rpm, during approximately 1 minute;
    - In the 3rd phase, the stirring velocity varies from 300 to 400 rpm, during approximately 1 minute;

    b) Sedimentation of the resulting solution of (a) during at least about two hours, and
    c) Separation of the two phases formed in (b), the precipitate or sludge and the supernatant or treated water.

2. Process according to claim 1, **characterized in that** the waste water is originated by the following agro-industries: wineries, slaughterhouses, cheese factory and olive oil presses.

3. Process according to claim 2, **characterized in that** the waste water is raw waste water, without having undergone any previous treatment.

4. Process according to claim 1, **characterized in that** the sedimentation step (b) lasts for more than 2 hours.

5. A process according to any one of the preceding claims, **characterized in that** the precipitate or sludge and / or the supernatant or treated water resulting from step (c) are collected for later use through agricultural reuse.

Patentansprüche

1. Verfahren zur Abwasserbehandlung von Agroindustrien umfassend folgende Schritte:

    (a) Einen Säuerungsschritt ausgeführt durch den Zusatz von einer wässrigen Lösung von Säure ausgewählt von Schwefelsäure, Salzsäure oder Salpetersäure, die einen pka-Wert von weniger als 2 haben, zu einem bestimmten Volumen von zu behandelndem Abwasser, bis der erreichte ph-Wert der gemischten Lösung von

1 bis 6 ist, und wobei die Säure unter Rührzustände untermischt ist, welche in drei Phasen stattfindet, jede der genannten Phasen mit einer abnehmenden Geschwindigkeit, wie folgt:

- In der 1ten Phase, die Rührgeschwindigkeit ändert sich zwischen 1200 und 1400 rpm, ungefähr 1 Minute lang;
- In der 2ten Phase, die Rührgeschwindigkeit ändert sich zwischen 700 und 800 rpm, ungefähr 1 Minute lang;
- In der 3ten Phase, die Rührgeschwindigkeit ändert sich zwischen 300 und 400 rpm, ungefähr 1 Minute lang.

(b) Sedimentbildung der ergebenen Lösung von (a) während mindestens zwei Stunden, und
(c) Trennung der zwei Phasen gestaltet in (b), der Niederschlag oder Schlamm und das Überstands- oder behandelnde Wasser.

2. Verfahren gemäß Anspruch 1 **dadurch gekennzeichnet, dass** das Abwasser durch die folgenden Agroindustrien entsteht: Weinkellereien, Schlachthäuser, Käsereien und Olivenölpressen.

3. Verfahren gemäß Anspruch 2 **dadurch gekennzeichnet, dass** das Abwasser rohes Abwasser ist, ohne vorhergehende Behandlung.

4. Verfahren gemäß Anspruch 1 **dadurch gekennzeichnet, dass** der Sedimentbildungsschritt (b) länger als 2 Stunden dauert.

5. Ein Verfahren gemäß einer der vorigen Ansprüche **dadurch gekennzeichnet, dass** der Niederschlag oder Schlamm und/ oder das Überstands- oder behandeltes Wasser folgend aus Schritt (c) zur späteren Anwendung mittels landwirtschaftlicher Wiederverwendung gesammelt sind.

Revendications

1. Processus de traitement des eaux usées issues des agro-industries, comprenant les étapes suivantes :

(a) Étape d'acidification réalisée par l'addition d'une solution aqueuse d'un acide sélectionné d'un acide sulfurique, d'un acide chlorhydrique ou d'un acide nitrique, ayant une valeur pKa inférieure à 2 pour un volume déterminé d'eaux usées à traiter, jusqu'à ce que la valeur pH de solution mélangée atteinte soit comprise entre 1 et 6 et où l'acide est ajoutée dans des conditions d'agitation, qui a lieu au cours de trois phases, où chacune de ces phases ont un taux décroissant de vitesse, comme suit :

- Au cours de la le phase, la vitesse d'agitation varie entre 1200 et 1400 rpm, pendant 1 minute, approximativement ;
- Au cours de la 2e phase, la vitesse d'agitation varie entre 700 et 800 rpm, pendant 1 minute, approximativement ;
- Au cours de la 3e phase, la vitesse d'agitation varie entre 300 et 400 rpm, pendant 1 minute, approximativement ;

b) Sédimentation de la solution qui résulte de (a) pendant au moins deux heures et
c) Séparation des deux phases formées en (b), le précipité ou la boue et le surnageant ou les eaux traitées.

2. Processus selon la revendication 1 **caractérisé par le fait que** les eaux usées proviennent des agro-industries suivantes : exploitations viticoles, abattoirs, usines de fromage et pressoirs d'huile d'olive.

3. Processus selon la revendication 2 **caractérisé par le fait que** les eaux usées sont des eaux résiduaires non traitées, n'ayant subi aucun traitement préalable.

4. Processus selon la revendication 1 **caractérisé par le fait que** la phase de sédimentation (b) dure plus de 2 heures.

5. Processus selon une quelconque revendication précédente, **caractérisé par le fait que** le précipité ou la boue et/ou le surnageant ou les eaux traitées issu(s) de l'étape (c) sont collecté(e/es) pour une utilisation ultérieure à travers la valorisation agricole.

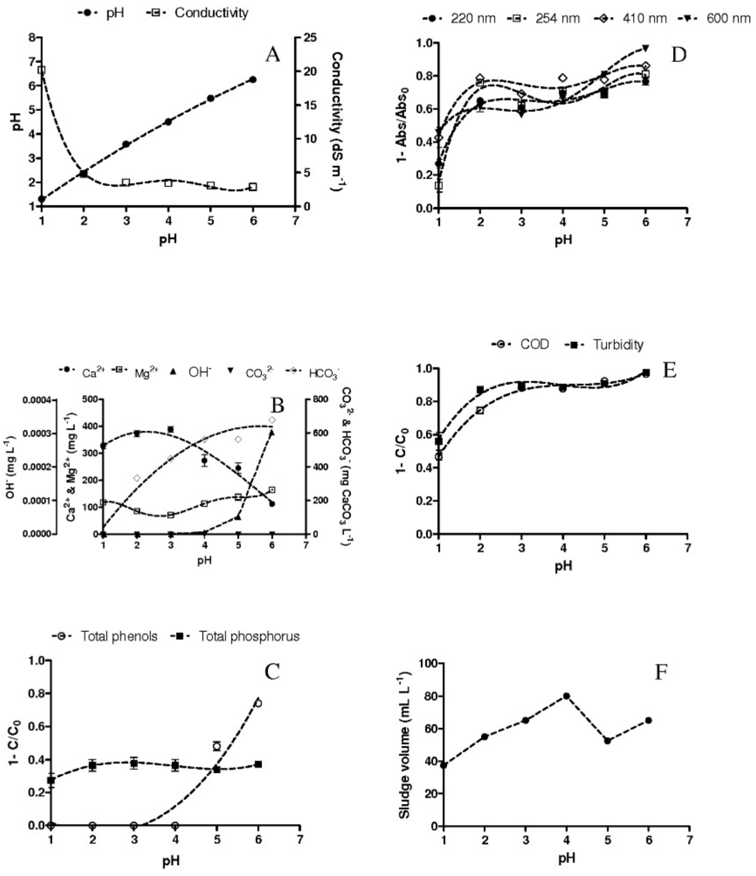
pH
Conductivity
A
pH
Conductivity (dS m^{-1})
1
2
3
4
5
6
7
8
0
5
10
15
20
25
220 nm
254 nm
410 nm
600 nm
D
1- Abs/Abs$_0$
0.0
0.2
0.4
0.6
0.8
1.0
Ca^{2+}
Mg^{2+}
OH$^-$
CO$_3$$^{2-}$
HCO$_3$$^-$
B
OH$^-$ (mg L^{-1})
0.0000
0.0001
0.0002
0.0003
0.0004
Ca^{2+} & Mg^{2+} (mg L^{-1})
0
100
200
300
400
500
CO$_3$$^{2-}$ & HCO$_3$$^-$ (mg CaCO$_3$ L^{-1})
0
200
400
600
800
COD
Turbidity
E
1- C/C$_0$
Total phenols
Total phosphorus
C
F
Sludge volume (mL L^{-1})
0
20
40
60
80
100
1
2
3
4
5
6
7
pH

Figure 1.

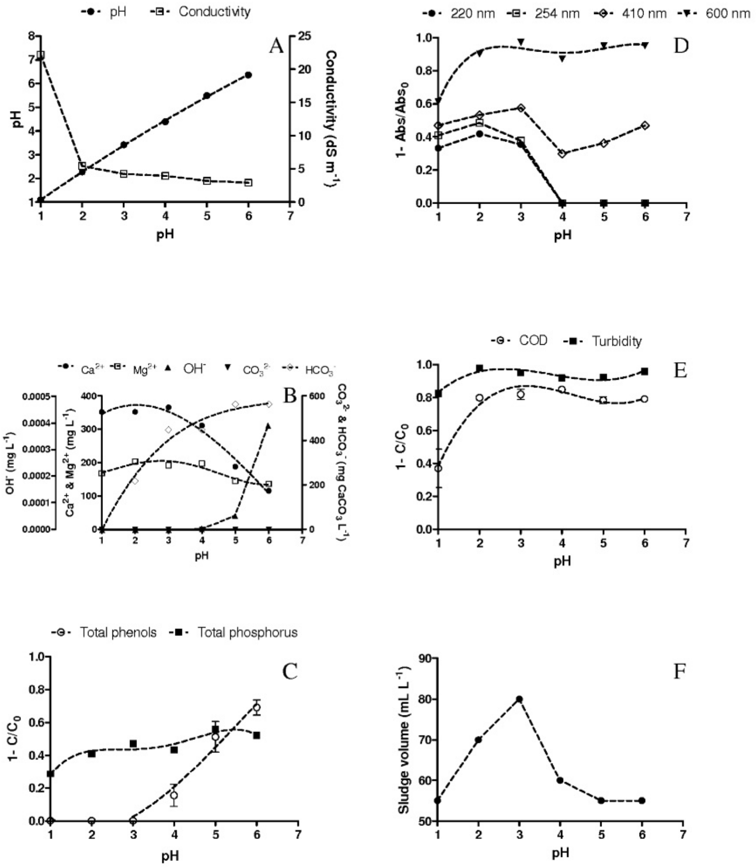
pH
Conductivity
A
pH
Conductivity (dS m^{-1})
pH
220 nm
254 nm
410 nm
600 nm
D
1- Abs/Abs$_0$
pH
Ca^{2+}
Mg^{2+}
OH$^-$
CO$_3^{2-}$
HCO$_3^-$
B
OH$^-$ (mg L^{-1})
Ca^{2+} & Mg^{2+} (mg L^{-1})
CO$_3^{2-}$ & HCO$_3^-$ (mg CaCO$_3$ L^{-1})
pH
COD
Turbidity
E
1- C/C$_0$
pH
Total phenols
Total phosphorus
C
1- C/C$_0$
pH
F
Sludge volume (mL L^{-1})
pH

Figure 2.

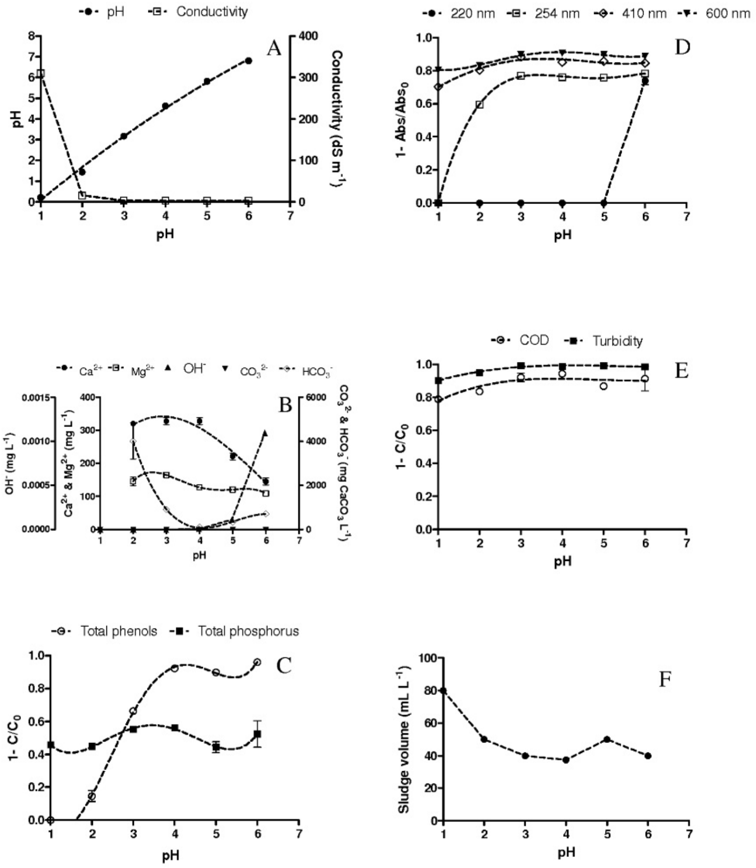
pH
Conductivity
A
Conductivity (dS m^{-1})
220 nm
254 nm
410 nm
600 nm
D
1- Abs/Abs$_0$
Ca^{2+}
Mg^{2+}
OH$^-$
CO$_3^{2-}$
HCO$_3^-$
B
OH$^-$ (mg L^{-1})
Ca^{2+} & Mg^{2+} (mg L^{-1})
CO$_3^{2-}$ & HCO$_3^-$ (mg CaCO$_3$ L^{-1})
COD
Turbidity
E
1- C/C$_0$
Total phenols
Total phosphorus
C
F
Sludge volume (mL L^{-1})

Figure 3.

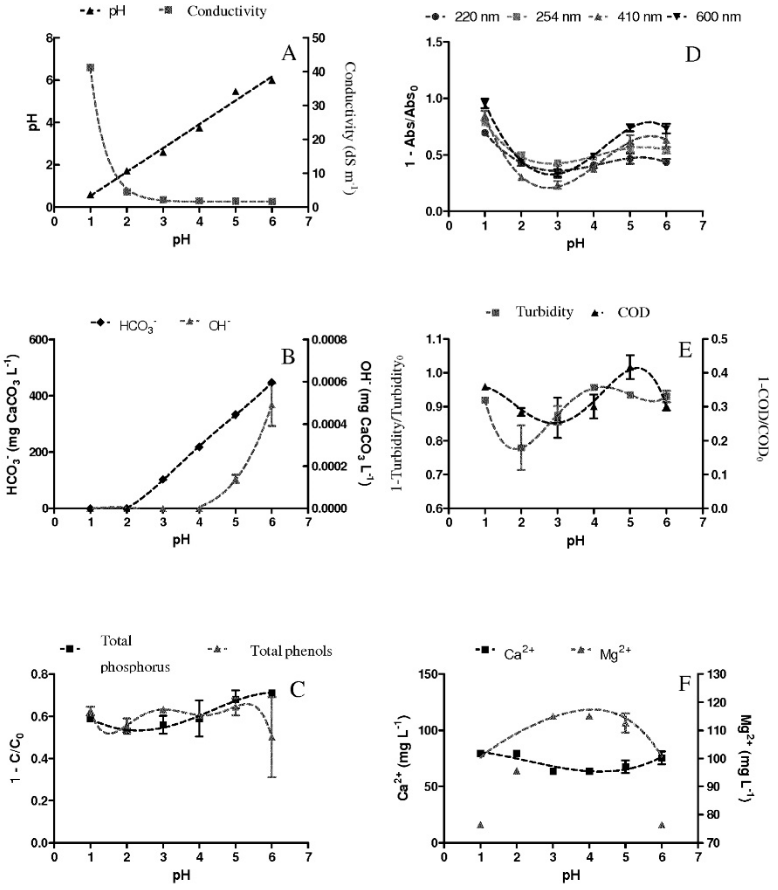
pH
Conductivity
A
pH
Conductivity (dS m⁻¹)
pH
220 nm
254 nm
410 nm
600 nm
D
1 - Abs/Abs₀
pH
HCO₃⁻
OH⁻
B
HCO₃⁻ (mg CaCO₃ L⁻¹)
OH⁻ (mg CaCO₃ L⁻¹)
pH
Turbidity
COD
E
1-Turbidity/Turbidity₀
1-COD/COD₀
pH
Total phosphorus
Total phenols
C
1 - C/C₀
pH
Ca²⁺
Mg²⁺
F
Ca²⁺ (mg L⁻¹)
Mg²⁺ (mg L⁻¹)
pH

Figure 4.

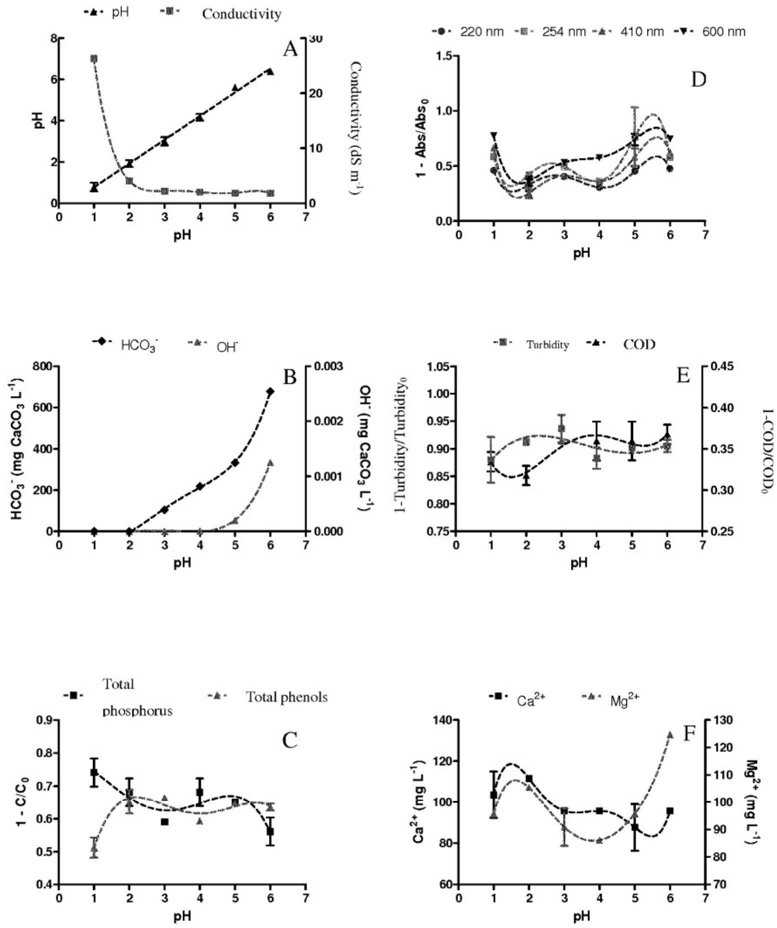
pH
Conductivity
A
pH
Conductivity (dS m-1)
0
2
4
6
8
0
10
20
30
0 1 2 3 4 5 6 7
pH
220 nm
254 nm
410 nm
600 nm
D
1 - Abs/Abs0
0.0
0.5
1.0
1.5
0 1 2 3 4 5 6 7
pH
HCO3-
OH-
B
HCO3- (mg CaCO3 L-1)
0
200
400
600
800
OH- (mg CaCO3 L-1)
0.000
0.001
0.002
0.003
0 1 2 3 4 5 6 7
pH
Turbidity
COD
E
1-Turbidity/Turbidity0
0.75
0.80
0.85
0.90
0.95
1.00
1.05
1-COD/COD0
0.25
0.30
0.35
0.40
0.45
0 1 2 3 4 5 6 7
pH
Total phosphorus
Total phenols
C
1 - C/C0
0.4
0.5
0.6
0.7
0.8
0.9
0 1 2 3 4 5 6 7
pH
Ca2+
Mg2+
F
Ca2+ (mg L-1)
60
80
100
120
140
Mg2+ (mg L-1)
70
80
90
100
110
120
130
0 1 2 3 4 5 6 7
pH

Figure 5.

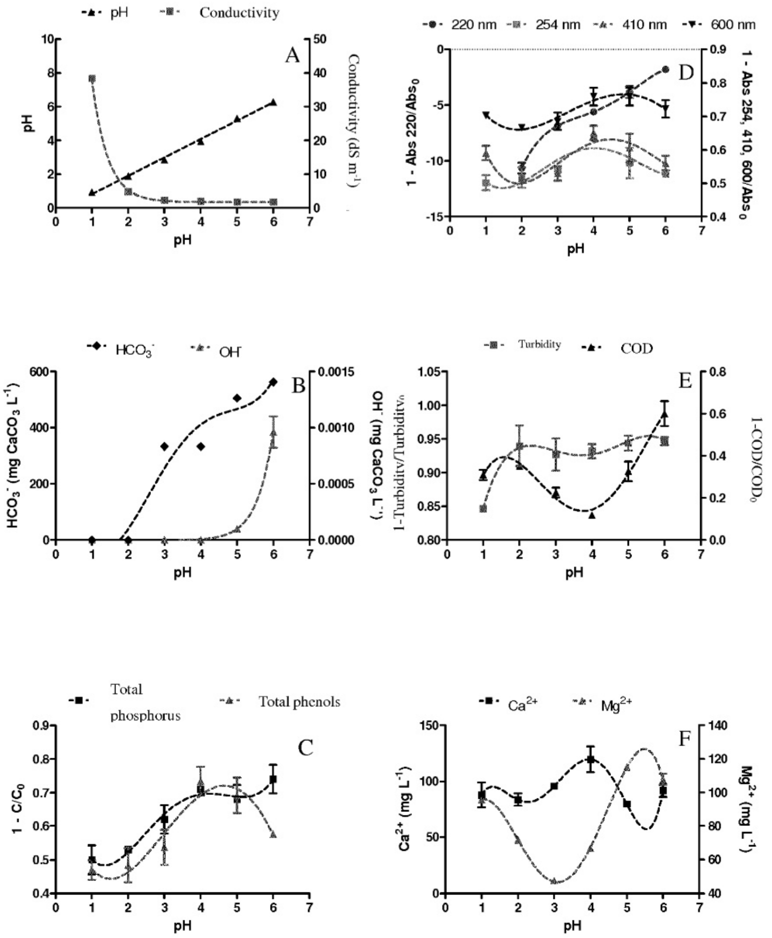
pH
Conductivity
A
pH
Conductivity (dS m^{-1})
220 nm
254 nm
410 nm
600 nm
D
1 - Abs 220/Abs$_0$
1 - Abs 254, 410, 600/Abs$_0$
HCO$_3^-$
OH$^-$
B
HCO$_3^-$ (mg CaCO$_3$ L^{-1})
OH$^-$ (mg CaCO$_3$ L^{-1})
Turbidity
COD
E
1-Turbidity/Turbidity$_0$
1-COD/COD$_0$
Total phosphorus
Total phenols
C
1 - C/C$_0$
Ca^{2+}
Mg^{2+}
F
Ca^{2+} (mg L^{-1})
Mg^{2+} (mg L^{-1})
pH

Figure 6.

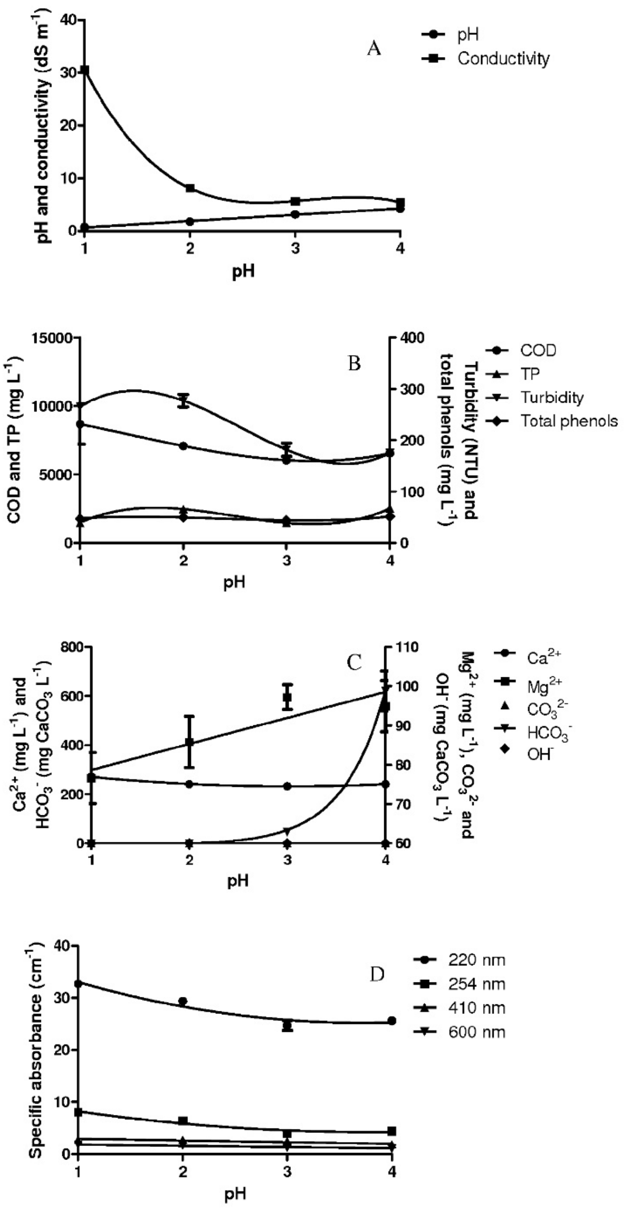
A
pH
Conductivity
pH and conductivity (dS m-1)
40
30
20
10
0
1
2
3
4
pH
B
COD
TP
Turbidity
Total phenols
COD and TP (mg L-1)
15000
10000
5000
0
Turbidity (NTU) and total phenols (mg L-1)
400
300
200
100
0
1
2
3
4
pH
C
Ca2+
Mg2+
CO32-
HCO3-
OH-
Ca2+ (mg L-1) and HCO3- (mg CaCO3 L-1)
800
600
400
200
0
Mg2+ (mg L-1), CO32- and OH- (mg CaCO3 L-1)
110
100
90
80
70
60
1
2
3
4
pH
D
220 nm
254 nm
410 nm
600 nm
Specific absorbance (cm-1)
40
30
20
10
0
1
2
3
4
pH

Figure 7.

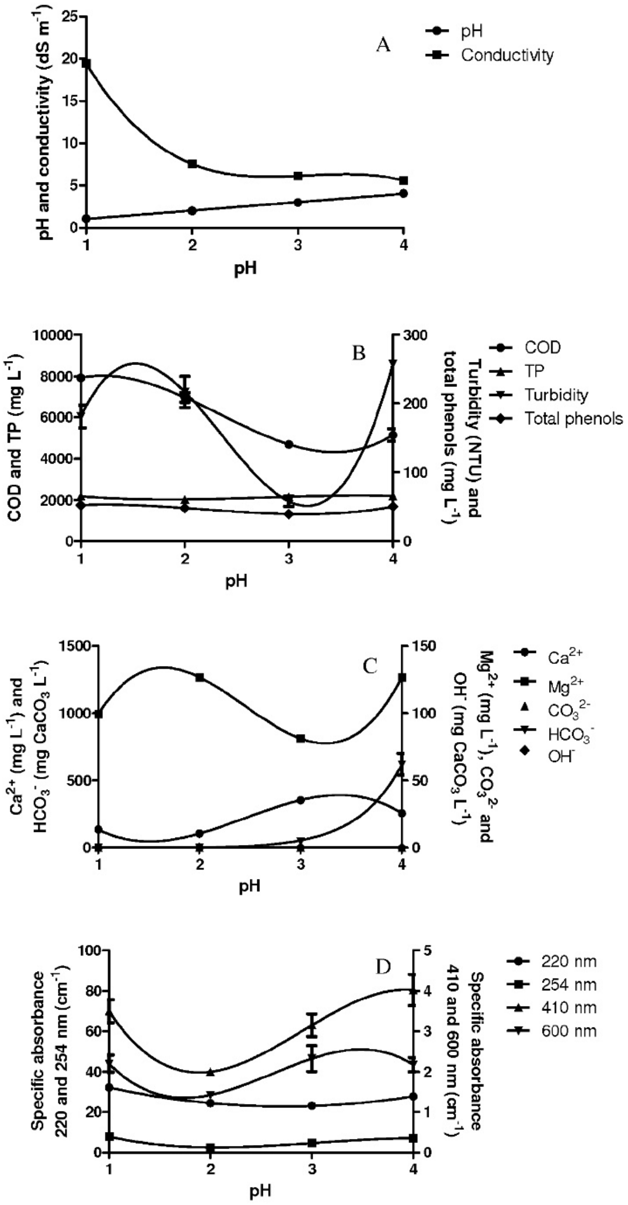
A
pH and conductivity (dS m-1)
25
20
15
10
5
0
1
2
3
4
pH
pH
Conductivity
B
COD and TP (mg L-1)
10000
8000
6000
4000
2000
0
Turbidity (NTU) and total phenols (mg L-1)
300
200
100
0
1
2
3
4
pH
COD
TP
Turbidity
Total phenols
C
Ca2+ (mg L-1) and HCO3- (mg CaCO3 L-1)
1500
1000
500
0
Mg2+ (mg L-1), CO32- and OH- (mg CaCO3 L-1)
150
100
50
0
1
2
3
4
pH
Ca2+
Mg2+
CO32-
HCO3-
OH-
D
Specific absorbance 220 and 254 nm (cm-1)
100
80
60
40
20
0
Specific absorbance 410 and 600 nm (cm-1)
5
4
3
2
1
0
1
2
3
4
pH
220 nm
254 nm
410 nm
600 nm

Figure 8

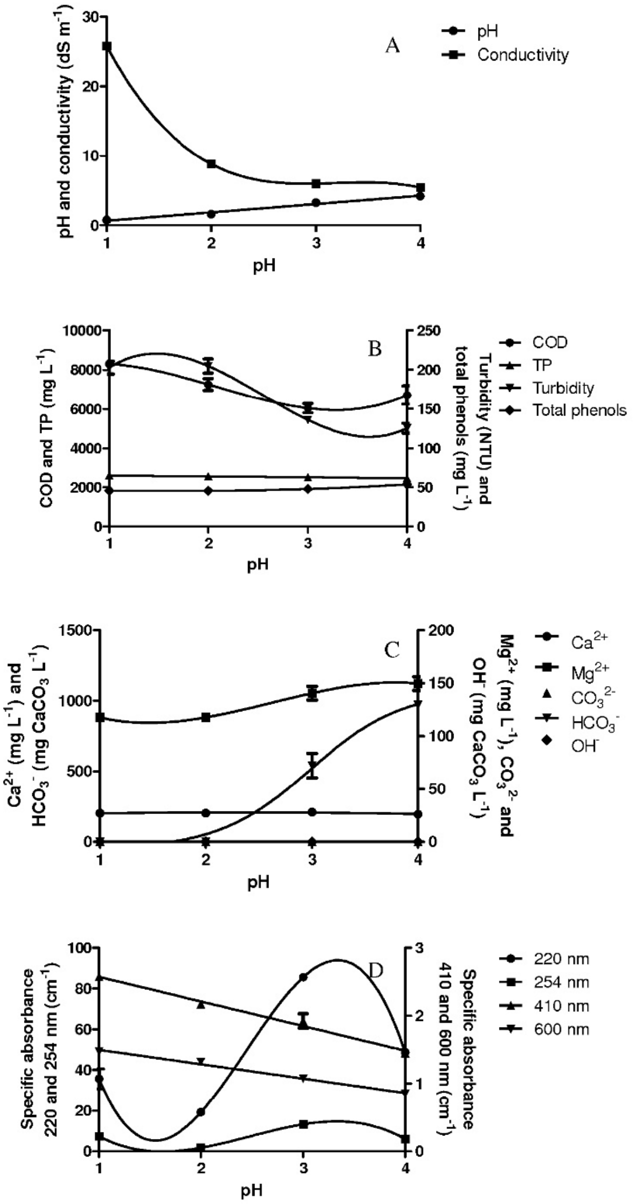
A
pH and conductivity (dS m-1)
30
20
10
0
1
2
3
4
pH
pH
Conductivity
B
COD and TP (mg L-1)
10000
8000
6000
4000
2000
0
Turbidity (NTU) and total phenols (mg L-1)
250
200
150
100
50
0
1
2
3
4
pH
COD
TP
Turbidity
Total phenols
C
Ca2+ (mg L-1) and HCO3- (mg CaCO3 L-1)
1500
1000
500
0
Mg2+ (mg L-1), CO32- and OH- (mg CaCO3 L-1)
200
150
100
50
0
1
2
3
4
pH
Ca2+
Mg2+
CO32-
HCO3-
OH-
D
Specific absorbance 220 and 254 nm (cm-1)
100
80
60
40
20
0
Specific absorbance 410 and 600 nm (cm-1)
3
2
1
0
1
2
3
4
pH
220 nm
254 nm
410 nm
600 nm

Figure 9.

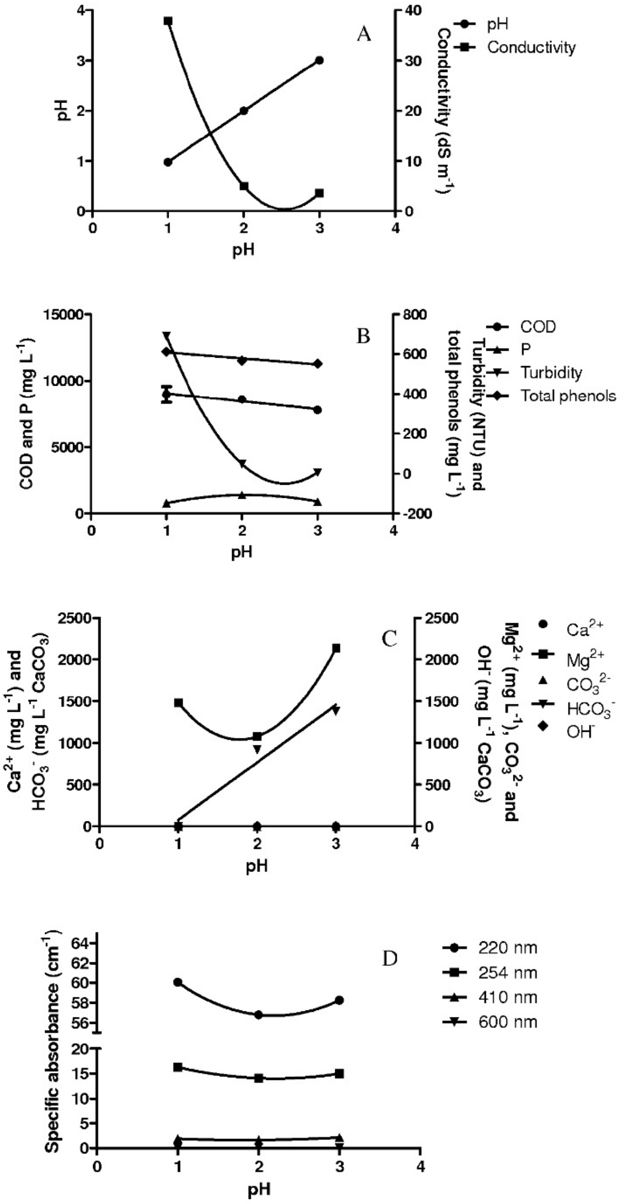
A
pH
Conductivity (dS m-1)
pH
Conductivity
0
1
2
3
4
10
20
30
40
B
COD and P (mg L-1)
Turbidity (NTU) and total phenols (mg L-1)
COD
P
Turbidity
Total phenols
0
5000
10000
15000
-200
200
400
600
800
C
Ca2+ (mg L-1) and HCO3- (mg L-1 CaCO3)
Mg2+ (mg L-1), CO32- and OH- (mg L-1 CaCO3)
Ca2+
Mg2+
CO32-
HCO3-
OH-
500
1000
1500
2000
2500
D
Specific absorbance (cm-1)
220 nm
254 nm
410 nm
600 nm
0
5
10
15
20
56
58
60
62
64

Figure 10.

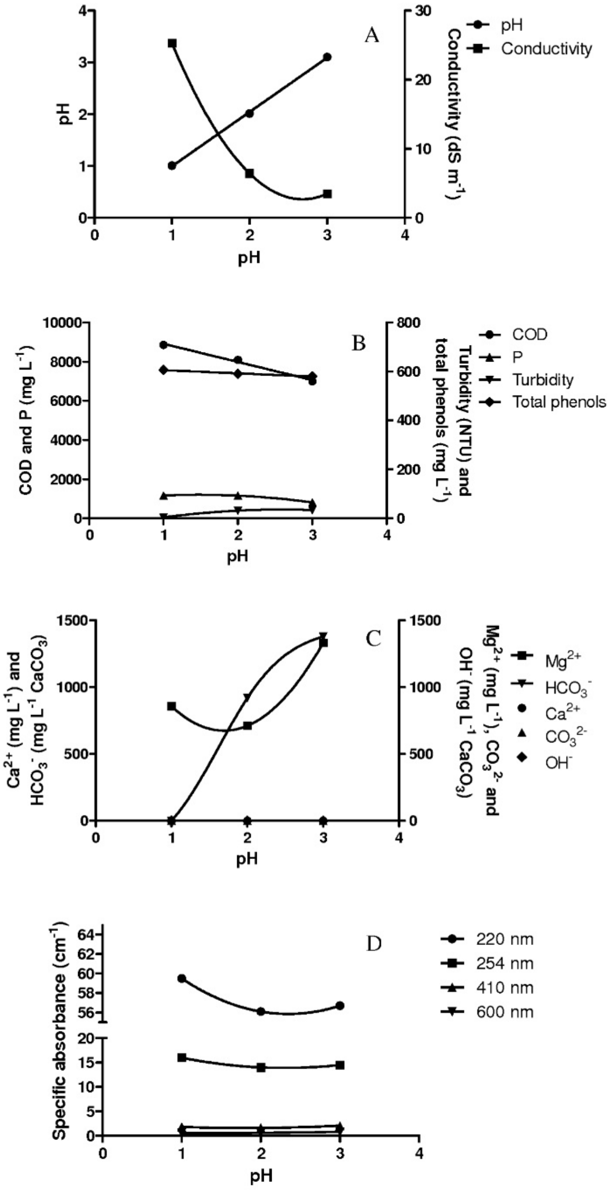
A
pH
Conductivity
pH
Conductivity (dS m-1)
0
1
2
3
4
0
10
20
30
pH
0
1
2
3
4
B
COD
P
Turbidity
Total phenols
COD and P (mg L-1)
Turbidity (NTU) and total phenols (mg L-1)
0
2000
4000
6000
8000
10000
0
200
400
600
800
pH
C
Mg2+
HCO3-
Ca2+
CO32-
OH-
Ca2+ (mg L-1) and HCO3- (mg L-1 CaCO3)
Mg2+ (mg L-1), CO32- and OH- (mg L-1 CaCO3)
0
500
1000
1500
pH
D
220 nm
254 nm
410 nm
600 nm
Specific absorbance (cm-1)
0
5
10
15
20
56
58
60
62
64
pH

Figure 11.

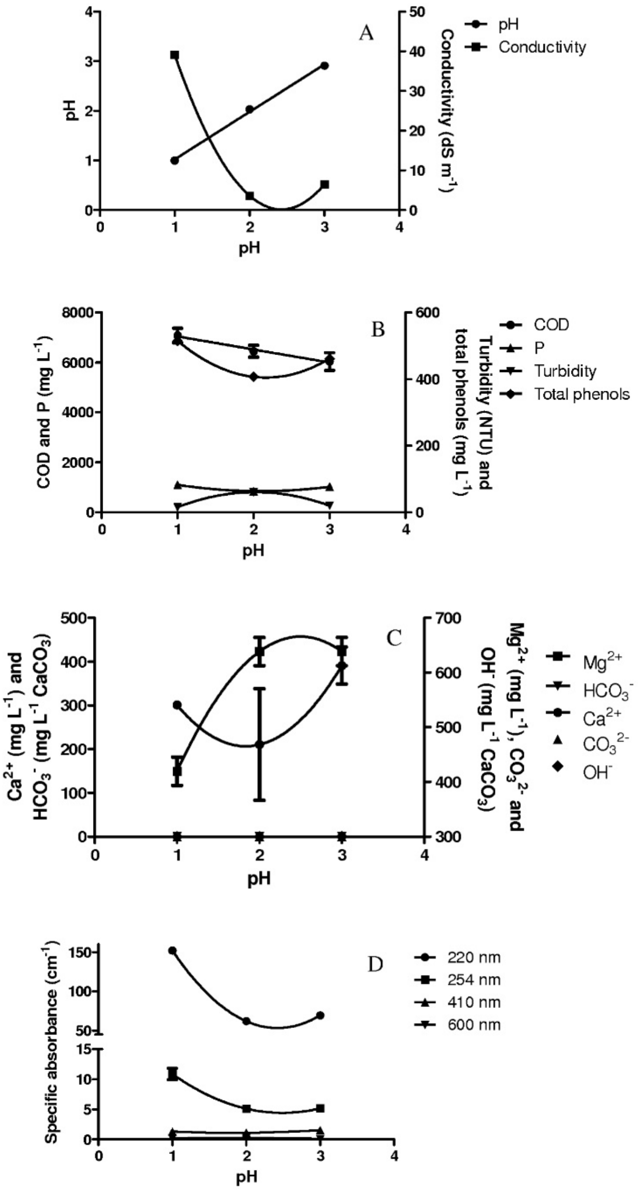
A
pH
0
1
2
3
4
Conductivity (dS m^{-1})
0
10
20
30
40
50
pH
0
1
2
3
4
pH
Conductivity
B
COD and P (mg L^{-1})
0
2000
4000
6000
8000
Turbidity (NTU) and total phenols (mg L^{-1})
0
200
400
600
pH
0
1
2
3
4
COD
P
Turbidity
Total phenols
C
Ca^{2+} (mg L^{-1}) and HCO$_3^-$ (mg L^{-1} CaCO$_3$)
0
100
200
300
400
500
Mg^{2+} (mg L^{-1}), CO$_3^{2-}$ and OH$^-$ (mg L^{-1} CaCO$_3$)
300
400
500
600
700
pH
0
1
2
3
4
Mg^{2+}
HCO$_3^-$
Ca^{2+}
CO$_3^{2-}$
OH$^-$
D
Specific absorbance (cm^{-1})
0
5
10
15
50
100
150
pH
0
1
2
3
4
220 nm
254 nm
410 nm
600 nm

Figure 12.

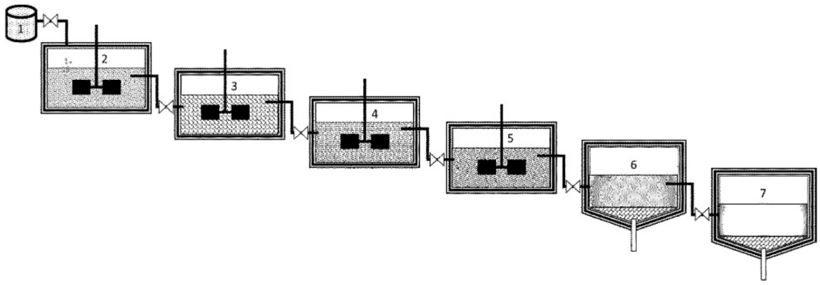

Figure 13.

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- **BABEL ; KURNIAWAN.** Cr (VI) removal from synthetic wastewater using coconut shell charcoal and commercial activated carbon modified with oxidizing agents and / or chitosan. *Chemosphere,* 2004, vol. 54 (7), 951-967 **[0009]**

Artigo 3 - Cheese wastewater treatment by acid and basic precipitation: Application of H_2SO_4, HNO_3, HCl, $Ca(OH)_2$ and NaOH

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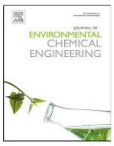

Cheese wastewater treatment by acid and basic precipitation: Application of H_2SO_4, HNO_3, HCl, $Ca(OH)_2$ and NaOH

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ABSTRACT

The performance of five innovative chemical precipitation processes (H_2SO_4, HNO_3, HCl, $Ca(OH)_2$ and NaOH) for the pretreatment of cheese wastewater was investigated.

For acid precipitation, a linear relationship between pH of the effluent and precipitation pH was found. High conductivity values were obtained when applying extreme pH conditions in acid precipitation. Alkalinity in the form of bicarbonates occurred at operating pH conditions of 3.0 and 4.0. H_2SO_4, HNO_3 and HCl were not able to effectively reduce organic matter. However, acid precipitation was revealed to be an appropriate process to reduce total phosphorus (up to 57.0 %), turbidity (up to 90.3 %) and total phenols (up to 49.9 %).

For basic precipitation ($Ca(OH)_2$ and NaOH), high pH and conductivity values in the effluent were obtained when applying extreme conditions (pH 12 and 13). In general, $Ca(OH)_2$ and NaOH processes presented alkalinity in the form of bicarbonates. These processes showed not only a reduction of chemical oxygen demand (COD) (up to 43.7 %) and total phosphorus (up to 95.6 %) but also an elimination of turbidity (up to 98.9 %) and total phenols (up to 65.5 %).

These acid and basic precipitation processes can be effective processes for the cheese whey wastewater pretreatment. Despite the rates of removal, the obtained effluents need the application of a post-treatment, such as a biological or chemical process.

1. Introduction

Utilization and disposal of cheese by-products assume a great importance [1,2]. Three types of cheese by-products can be produced during cheese production: cheese whey, second cheese whey and cheese wastewater. Cheese whey, the main component of cheese wastewater, has 85–95 % of milk volume and presents 55 % of milk nutrients [3,4]. The nutrients present in cheese whey constitute lactose (4.5–5.0 % w/v), soluble proteins (0.6–0.8 % w/v), lipids (0.4–0.5 % w/v) and mineral salts (8–10 % of dried extract) [4].

Cheese by-products are problems for the environment and public health owing to the organic, acid and saline properties and volume generated. In this sense, 9 L of cheese whey can be produced per kilogram of cheese manufactured [3]. Moreover, more than 145 million tone of cheese whey are produced per year [5]. With regard to organic properties, cheese effluents present chemical and biochemical oxygen demand (COD and BOD) values in the ranges of 0.8–102 g L^{-1} and 0.6–60 g L^{-1}, respectively. These effluents also present low pH (4.0–8.7), suspended solids (0.1–22.0 g L^{-1}), nitrogen (0.01–1.7 g L^{-1}) and total phosphorus (0.006–0.5 g L^{-1}) [6].

These effluents can not be discharged to municipal water treatment plants because of their high organic content, fats, nutrients and salts. Biological treatments have been frequently used, namely, aerobic [7–10] and anaerobic digestion [1,7,11,12]. Aerobic digestion becomes ineffective due to the costs for the application of oxygen [13]. Anaerobic digestion presents some advantages, such as the generation of biogas and the production of effluent that can be post-treated together with other wastewaters [13,14]. However, application of anaerobic digestion is limited because high biodegradability, high organic content and low bicarbonate alkalinity [1,2,13,14]. Additionally, anaerobic digestion produces sludge with problems in the sedimentation. Rapid acidification of the medium and long hydraulic retention times (HRT) are other problems that can be found in the anaerobic digestion [13].

Coagulation–flocculation with $FeSO_4$, $FeCl_3$, $Al_2(SO_4)_3$ [9,15] and

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precipitation with lime and NaOH [10,15–17] were applied to raw cheese wastewater, allowing the reduction of organic matter, nutrients, turbidity, fats and total suspended solids. However, no study reports the removal of total phenols by basic precipitation. The O_3, $O_3 + H_2O_2$, O_3 + N-150 (Fe_2O_3-MnOx), O_3 + Mn–Ce–O (70/30) [8] and Fenton [18] were also used to treat cheese wastewater, reducing organic matter. However, in these cases, a biological treatment (activated sludge) was previously applied to reduce the contamination of raw cheese wastewater. The sequence electrocoagulation + electrochemical advanced oxidation was used to treat cheese wastewater [19]. Few studies on physicochemical processes have been carried out to treat cheese wastewater. Thus, in this work, innovative physicochemical processes were applied to pretreat cheese wastewater, using H_2SO_4, HNO_3, HCl, $Ca(OH)_2$ and NaOH.

2. Materials and methods

2.1. Reagents

Hydrochloric acid (37 %, $\rho = 1.18\ g\ mL^{-1}$, $M = 36.46\ g\ mol^{-1}$) and sulphuric acid (> 95 % ,$\rho = 1.83\ g\ mL^{-1}$, $M = 98.07\ g\ mol^{-1}$) were supplied by Fisher Chemical (Vitrilab, Portugal). Nitric acid (65 %, $\rho = 1.395\ kg\ L^{-1}$, $M = 63.01\ g\ mol^{-1}$) was obtained from PanReac AppliChem (Vitrilab, Portugal). Sodium hydroxide – pellets (99.25 %, $M = 40\ g\ mol^{-1}$) were supplied by Fisher Chemical (Vitrilab, Portugal), and $Ca(OH)_2$ was obtained from MANUEL DA PIEDADE BATISTA& IRMÃO, LDA (Portugal).

2.2. Raw cheese wastewater collection

The raw cheese wastewater was collected in a tank from an enterprise located in the Alentejo region, Portugal. The enterprise produces several types of cheeses, namely, sheep and goat cheeses. Raw wastewater was stored in 5 L polyethylene terephthalate (PET) containers at −20 °C before characterization and treatment. The treated wastewaters were stored in PET containers at 4 °C.

2.3. Physicochemical characterization of raw and treated wastewater

pH was monitored using a meter Consort C 861 (Consort nv, Belgique). Temperature and conductivity were measured in a VWR CO 3100 H apparatus using an electrode VWR CO 11.

Characteristic absorbances were determined, after appropriate dilution, in the ultraviolet and visible region using a Thermo Scientific Helios α spectrophotometer (Thermo Scientific, Germany). In the ultraviolet region, absorbances at wavelengths of 220 and 254 nm were evaluated, while in the visible region, absorbances at wavelengths of 410 and 600 were measured. The absorbances at 220, 254, 410 and 600 nm indicate the presence of low molecular weight compounds (simple molecules) formed from complex molecules (humic acids); high molecular weight organic compounds with high degree of aromaticity, high number of double and triple bonds, and phenolic groups; color indicator and optical density of the microorganisms cultures, respectively, [20–26]. Specific absorbance was obtained multiplying the absorbance and dilution used according to Standard Methods [20].

Turbidity was monitored by a WTW Turb550 turbidimeter (WTW, Germany), comparing white light transmitted by the samples and formazine standard suspensions [20,27,28]. COD was evaluated by colorimetric method after a digestion step (150 ± 2 °C for 2 h) in a digester Techne Dri-block DB 200/3 (ERT Lda, Portugal) [20,27].

Total phosphorus measurements were evaluated after calcination at 600 °C during 2 h and digestion step with HCl 6 N solution using a heating/stirring plate. The digested samples were filtered using filters Whatman™ 1001. Total phosphorus was monitored through a colorimetric method by measuring the absorbance at 470 nm after the reaction of orthophosphates with vanadate-molybdate reagent [20]. Total hardness was determined by volumetric complexation method and eriochrome black T indicator, while calcium hardness was evaluated using the presence of calcon indicator [20]. Magnesium hardness was obtained by difference between total hardness and calcium hardness.

Volumetric method using a solution of known concentration of hydrochloric acid and phenolphthalein indicator and indicator of methyl orange was used to determine phenolphthalein alkalinity and methyl orange alkalinity. The sum of alkalinity to phenolphthalein and alkalinity to methyl orange corresponds to the total alkalinity.

The spectrophotometric method using the Folin-Ciocalteu phenol reagent was used to evaluate the total phenols content, according to Singleton and Rossi [29].

2.4. Experimental procedure

Experiments were carried out using 250 mL of raw wastewater. In acid (H_2SO_4, HNO_3 and HCl) and basic ($Ca(OH)_2$ and NaOH) precipitation, the precipitant was added to raw wastewater until the pH reached the desired value: 1.0, 2.0, 3.0 and 4.0 (for acid precipitation) and 6.0, 7.0, 8.0, 9.0, 10.0, 11.0, 12.0 and 13.0 (for basic precipitation). When the pH reached the desired value, stirring was gradually stopped (700–800 rpm for 1 min followed by 300–400 rpm for 1 min) and sedimentation of the formed agglomerates occurred during 24 h.

3. Results and discussion

3.1. Cheese wastewater characterization

Table 1 presents the cheese wastewater characterization. Cheese wastewater presented pH around 4.520 ± 0.2055, as observed by Kalyuzhnyi et al. [1], Gavala et al. [12], Martins et al. [18], Martins and Quinta-Ferreira [8] and Rivas et al. [9]. Additionally, cheese wastewater had high conductivity value (5.23 ± 0.277 $dS\ m^{-1}$), however, the conductivity value was lower than that obtained by Rivas et al. [9]. Alkalinity and hardness were mainly due to the presence of bicarbonates and calcium, respectively.

Cheese wastewater represents a serious problem for the environment due to the presence of organic matter, monitored by COD in the range of 4416.7–5250.0 $mg\ L^{-1}$, higher than that obtained by Fang [7]. Cheese wastewater presented solids in suspension with turbidity values of 594.92 ± 49.542 NTU, lower than that observed by Rivas et al. [9] and Rivas et al. [10]. Cheese wastewater had nutrients with a content of total phosphorus of 3411.8 ± 1255.64 $mg\ L^{-1}$, higher than that obtained by Gutiérrez et al. [11], Gavala et al. [12], Rivas et al. [9] and Rivas et al. [10].

3.2. Cheese wastewater treatment

3.2.1. Acid precipitation

3.2.1.1. *H_2SO_4 addition.* Acid precipitation was applied for the treatment of cheese wastewater using H_2SO_4 with formation of a precipitate, which may result from the existence of amphoteric metal species that precipitate in the form of oxides or hydroxides; displacement of soluble complexes by corresponding insoluble oxides or hydroxides, and existence of compounds soluble in basic medium and insoluble in acid medium, as the fraction of the humic substances [23].

Fig. 1 illustrates the results obtained for H_2SO_4 precipitation. A linear increase of the effluent pH was obtained ($r^2 = 0.9958$) according to the precipitation pH (Fig. 1A). The application of pH around 1.0 increased significantly the conductivity of the effluent (30.55 ± 0.071) (Fig. 1A). Similar results (43.1 $dS\ m^{-1}$) were obtained by Prazeres et al. [30] when treating vinasse from sugarcane with acid precipitation by H_2SO_4 15 g L^{-1} addition (pH = 1.0). Acid precipitation with H_2SO_4 addition led to reduction of absorbances characteristics (8.1–64.0 %) (Fig. 1D). At low pH, absorbance at 220 nm increased, decreasing for pH ≥ 2.0 (8.1–22.6 %). Absorbances at 254, 410 and 600 nm presented

Table 1
Cheese wastewater characterization.

Parameter	Units	Interval	Mean value ± standard deviation
pH	Sorensen scale	4.162-4.675	4.520 ± 0.2055
Conductivity	dS m^{-1}	4.86-5.54	5.23 ± 0.277
Temperature	ºC	15.1-17.6	16.3 ± 0.93
Absorbance at 220 nm	(dilution 1:25)	0.599-0.700	0.639 ± 0.0383
Absorbance at 254 nm	(dilution 1:25)	0.157-0.196	0.180 ± 0.0147
Absorbance at 410 nm	(dilution 1:25)	0.083-0.101	0.092 ± 0.0065
Absorbance at 600 nm	(dilution 1:25)	0.053-0.071	0.060 ± 0.0072
Phenolphthalein alkalinity	mg $CaCO_3$ L^{-1}	< DL	< DL
Total alkalinity	mg $CaCO_3$ L^{-1}	1155.4-1708.5	1388.9 ± 251.1
Hydroxides	mg $CaCO_3$ L^{-1}	< DL	< DL
Carbonates	mg $CaCO_3$ L^{-1}	< DL	< DL
Bicarbonates	mg $CaCO_3$ L^{-1}	1155.4-1708.5	1388.9 ± 251.1
Total hardness	mg $CaCO_3$ L^{-1}	934.6-1347.5	1054.7 ± 168.69
Calcic hardness	mg $CaCO_3$ L^{-1}	487.9-675.6	589.3 ± 72.20
Magnesium hardness	mg $CaCO_3$ L^{-1}	296.5-709.4	465.4 ± 151.30
Calcium	mg L^{-1}	195.6-270.8	236.2 ± 28.94
Magnesium	mg L^{-1}	72.1-172.4	113.1 ± 36.77
COD	mg L^{-1}	4416.7-5250.0	4916.7 ± 353.55
Turbidity	NTU	536.49-659.73	594.92 ± 49.542
Total phosphorus	mg L^{-1}	1796.1-4894.1	3411.8 ± 1255.64
Total phenols	mg equivalent of gallic acid L^{-1}	65.00-82.86	75.92 ± 6.760

Results after five replicates. COD – chemical oxygen demand. DL – detection limit.

the following reductions 10.5–55.4, 37.2–59.1 and 41.5–64.0 %. The reduction of absorbances at 410 and 600 nm improved by increasing the precipitation pH (Fig. 1D). According to Pacheco and Peralta-Zamora [31], this process allowed the precipitation of aromatic amino acids, polycyclic aromatic compounds, carbohydrates, naphthalene and derivatives.

The decrease of pH (1.0) led to an increase of calcium content (270.8 mg L^{-1}) due to the absence of carbonaceous species, maintaining practically constant in the pH range of 2-4 (233.2–240.7 mg L^{-1}) (Fig. 1C). The magnesium content decreased (14.1–32.3 %) compared with raw wastewater, varying between 76.6 and 97.2 mg L^{-1} (Fig. 1C). Prazeres et al. [30] obtained reductions of calcium, magnesium and phosphorus up to 36 % in the acid precipitation (H_2SO_4 addition) of vinasse from sugarcane. The presence of bicarbonates occurred for pH 3 and 4 (46.0–620.3 mg $CaCO_3$ L^{-1}) (Fig. 1C).

Acid precipitation with H_2SO_4 addition did not allow to eliminate organic matter, but decreased the total phosphorus content with removals of 26.0–57.0 % (Fig. 1B). Also, Prazeres et al. [30] obtained low removals of COD (12 %) when applying H_2SO_4 for vinasse treatment due to the presence of organic matter in the form of organic acids, which are soluble in acid medium. In contrast, Garg et al. [32] obtained elimination of COD when treating diluted black liquor through acid precipitation with H_2SO_4. Additionally, Prazeres et al. [33] verified removals of COD, turbidity, absorbances and total phosphorus when H_2SO_4 was used to treat cheese wastewater. Increasing precipitation pH led to an increase in the removals of turbidity (53.4–70.5 %) (Fig. 1B). Total phenols elimination in the range of 34.5–44.1 % was obtained (Fig. 1B).

3.2.1.2. HNO_3 addition. HNO_3 was added to cheese wastewater in order to reduce the organic contamination and nutrients (Fig. 2). An increase in the precipitation pH originated a linear increase in the pH of the effluent ($r^2 = 0.9836$) (Fig. 2A). Deng et al. [34] detected a decrease of the pH in the sludge by acidification with nitric acid, permitting the dissolution of heavy metals. High conductivity values were obtained when applying pH around 1.0 (25.80 ± 0.141 dS m^{-1}) (Fig. 2A). Characteristic absorbances were efficiently eliminated at 220, 254, 410 and 600 nm, according to the precipitation pH (18.2–80.6 %) (Fig. 2D). It can be referred increases in the characteristic absorbances under some operating conditions (Fig. 2D). A stable behavior was obtained for the calcium (195.6–210.6 mg L^{-1}) and magnesium (117.7–149.6 mg L^{-1}) contents, independently of the precipitation pH (Fig. 2C). As observed for precipitation with H_2SO_4 addition, the presence of bicarbonates appeared for pH 3 and 4 (540.8–971.0 mg $CaCO_3$ L^{-1}) (Fig. 2C). Organic matter was not eliminated using this acid precipitation, presenting reduction of total phosphorus (24.0–28.0 %), turbidity (65.6–78.9 %) and total phenols (31.9–42.2 %) (Fig. 2B). Deng et al. [34] also obtained reductions of Cu, Zn and Pb in sewage sludge by using nitric acid with ultrasound.

3.2.1.3. HCl addition. HCl was applied to treat cheese whey wastewater under different operating conditions (Fig. 3). As expected, pH of the effluent increased by increasing the precipitation pH ($r^2 = 0.9985$) (Fig. 3A). High conductivity value was obtained at pH 1.0 (19.46 ± 0.134 dS m^{-1}) (Fig. 3A). The highest removals of characteristic absorbances at 254, 410 and 600 nm were obtained for pH around 2.0 (Fig. 3D). Calcium content decreased when applying pH of 1 and 2 (42.7–55.4 %), compared to raw wastewater (Fig. 3C). Calcium and magnesium content presented values between 105.3–353.5 mg L^{-1} and 81.2–126.8 mg L^{-1}, respectively (Fig. 3C). The alkalinity appeared in the form of bicarbonates for pH 3.0 and 4.0 (46.0–620.3 mg $CaCO_3$ L^{-1}) (Fig. 3C). Acid precipitation was not efficient to remove organic matter due to presence of soluble organic species in acid medium, allowing the removal of total phosphorus (36.0–40.9 %), turbidity (56.7–90.3 %) and total phenols (33.4–49.9 %) (Fig. 3B) by precipitation reactions. In contrast, Rattanapan et al. [35] observed reduction of grease and oil and COD in the treatment of biodiesel wastewater with HCl and H_2SO_4.

3.2.2. Basic precipitation

3.2.2.1. $Ca(OH)_2$ addition. $Ca(OH)_2$ was added to cheese wastewater in the range pH of 6–13 (Fig. 4), remaining the effluent pH practically constant (6.098–6.951) up to pH = 11.0. From this point, the effluent pH increased up to pH = 12,719 when applying precipitation pH of 13.0 (Fig. 4A). Similar behavior was obtained for the conductivity of the treated wastewater (Fig. 4A). Characteristic absorbances were removed efficiently (39.2–95.0 %) for high precipitation pH (12.0 and 13.0) (Fig. 4D).

Calcium content was eliminated (4.5–61.8 %) at pH 6.0 and 7.0, increasing in the pH range of 8.0-13.0 (353.5–1850.3 mg L^{-1}) (Fig. 4C) due to the $Ca(OH)_2$ addition. Magnesium content increased up to pH = 7.0 (300.1 mg L^{-1}), decreasing to pH 10.0 (135.9 mg L^{-1}), maintaining practically constant from this point (145.0–154.2 mg L^{-1})

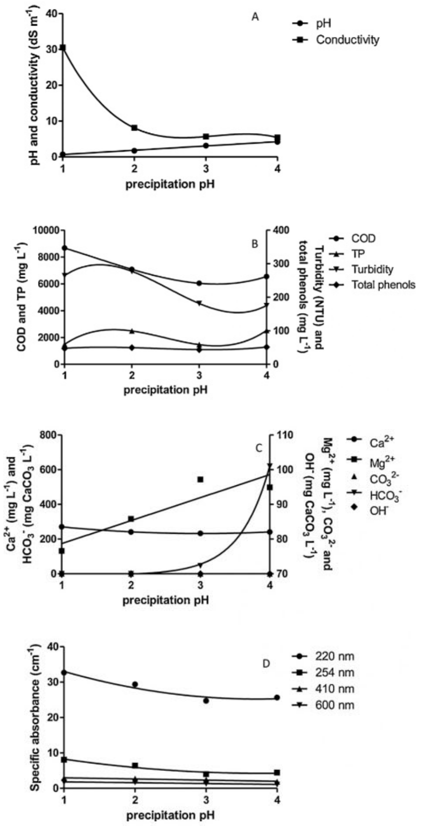

Fig. 1. H_2SO_4 precipitation for the treatment of cheese wastewater.

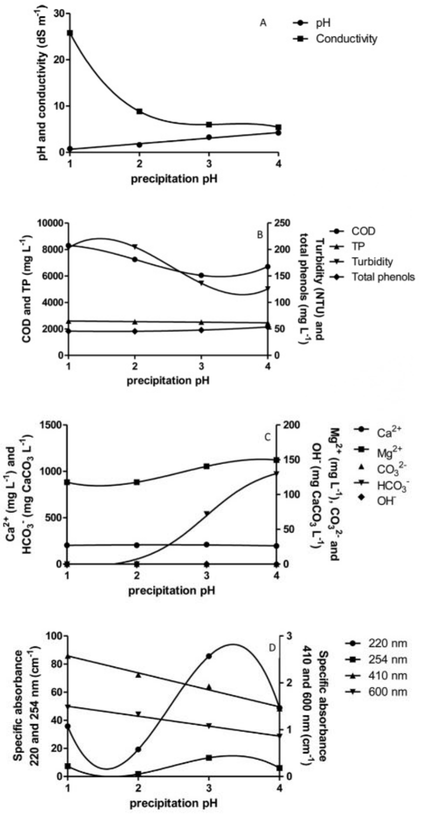

Fig. 2. HNO_3 precipitation for the treatment of cheese wastewater.

(Fig. 4C). Bicarbonates presented similar behavior. Thus, bicarbonates content increased up to pH 8–9 (2261.6 mg $CaCO_3$ L^{-1}), decreasing to 0 when pH 13 was reached (Fig. 4C).

When $Ca(OH)_2$ is added to wastewater, which presents sufficient amounts of alkalinity (bicarbonates), hardness and phosphates, the following reactions may occur [28,36–40]:

$$Ca(OH)_2 + H_2CO_3 \rightleftharpoons CaCO_3\downarrow + 2H_2O \quad (1)$$

$$Ca(OH)_2 + Ca(HCO_3)_2 \rightleftharpoons 2CaCO_3\downarrow + 2H_2O \quad (2)$$

$$Ca(OH)_2 + Mg(HCO_3)_2 \rightleftharpoons MgCO_3\downarrow + CaCO_3\downarrow + 2H_2O \quad (3)$$

$$Ca(OH)_2 + MgCO_3 \rightleftharpoons Mg(OH)_2\downarrow + CaCO_3\downarrow \quad (4)$$

$$3Ca(OH)_2 + 2PO_4{}^{3-} \rightleftharpoons Ca_3(PO_4)_2\downarrow + 6OH\text{-} \quad (5)$$

$$4Ca(OH)_2 + 3PO_4{}^{3-} + H_2O \rightleftharpoons Ca_4H(PO_4)_3\downarrow + 9OH\text{-} \quad (6)$$

$$5Ca^{2+} + 6PO_4{}^{3-} + 2OH\text{-}\!\!\rightarrow Ca_{10}(PO_4)_6(OH)_2\downarrow \quad (7)$$

The addition of $Ca(OH)_2$ allows the precipitation of calcium carbonate (reactions 1–4), magnesium carbonate and hydroxide (reactions 3 and 4) and phosphates (reactions 5–7), which trap and entrain the suspension and colloidal particles (coagulation by scanning), increasing the efficiencies of elimination of organic matter and turbidity. $Ca(OH)_2$ acts as a weighting agent, increasing the density of the particles, and consequently their fixation and the water clarification. The increase of the applied $Ca(OH)_2$ dose results in the increase of Ca^{2+} ions and inorganic carbon species ($HCO_3{}^-$), consequently increasing the possibility of precipitate formation. $Ca(OH)_2$ acts efficiently in the precipitation of phosphates, metals, microorganisms, BOD_5, COD, TSS, total phosphorus, hardness and organic compounds of high molecular weight (humic and fulvic acids) [38,39].

The highest organic matter removal (29.7 %) was obtained at pH 11.0 (Fig. 4B). High total phosphorus elimination (61.9–95.6 %) only occurred for pH ≥ 8.0 (Fig. 4B). The total phosphorus removal is very dependent on pH in $Ca(OH)_2$ precipitation, since at pH = 8.0 the present carbonates compete with the phosphate ions for the calcium ions, reducing the amount of free calcium ions, and consequently the rate of phosphate precipitation. For pH values between 9 and 11, co-precipitation of carbonate and phosphate occurs, however, the total phosphorus content in the precipitate is reduced [41]. According to Ramalho [36], lime in the pH range = 10.5–11.0 is less effective in removing total phosphorus than $FeCl_3$ and $Al_2(SO_4)_3$, obtaining only a 50 % yield and a greater volume of sludge. However, combinations of lime with $FeCl_3$ can lead to total phosphorus elimination with yields of

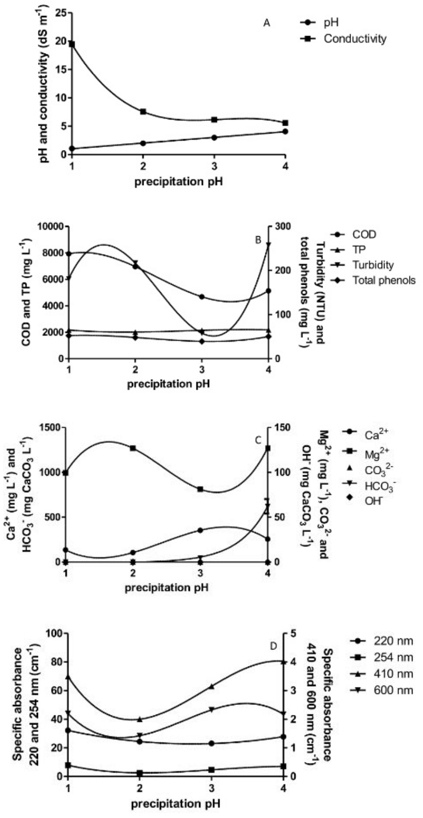

Fig. 3. HCl precipitation for the treatment of cheese wastewater.

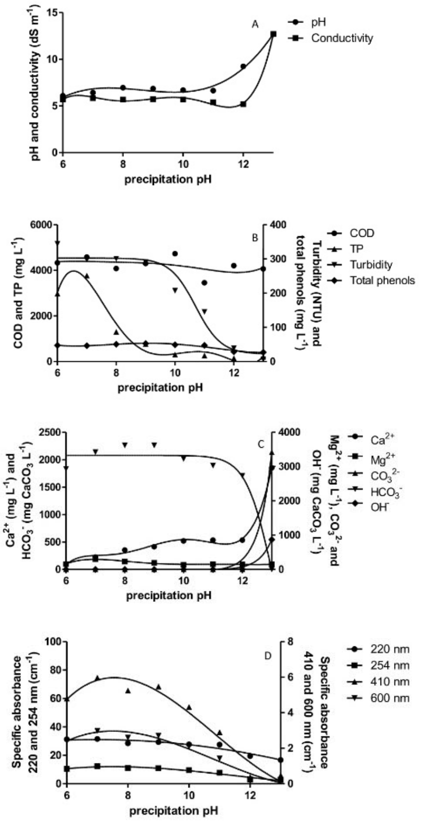

Fig. 4. $Ca(OH)_2$ precipitation for the treatment of cheese wastewater.

about 95 %.

Turbidity removal remained practically constant (42.1–51.1 %) up to pH 9.0, increasing from this point. $Ca(OH)_2$ addition was efficient in removing turbidity, reaching the maximum (93,4–98.9 %) at pH 12.0 and 13.0 (Fig. 4B). Similar results were obtained for total phenols removals with high eliminations (63.2–65.5 %) at pH 12.0 and 13.0 (Fig. 4B).

3.2.2.2. NaOH addition. Cheese wastewater was treated by NaOH addition (Fig. 5). No significant differences were obtained for the effluent pH in the precipitation pH range of 6.0–11.0, increasing significantly for high precipitation pH (Fig. 5A). Similar results were achieved for conductivity values (Fig. 5A).

High operating pH (10–13) led to an increase of low molecular weight compounds (simple molecules) formed from complex molecules (Fig. 5D). These low molecular compounds can be promoters of microorganisms due to their higher biodegradability [22,25]. In general, NaOH addition caused an increase of high molecular weight organic compounds with high degree of aromaticity, high number of double and triple bonds, and phenolic groups and color compounds (Fig. 5D). Additionally, NaOH addition allowed the removal of optical density of the microorganisms cultures, except for the highest pH (13.0) (Fig. 5D).

With respect to calcium and magnesium, removals were observed in the range of 44.3–100 % and 4.0–58.5 %, respectively (Fig. 5C). During NaOH precipitation, the formation of magnesium and calcium hydroxide and carbonates can occur (reactions 8–11) [17,38]:

$$2NaOH + Mg(HCO_3)_2 \rightleftharpoons Mg(OH)_2\downarrow + 2NaHCO_3 \quad (8)$$

$$2NaOH + Ca(HCO_3)_2 \rightleftharpoons Ca(OH)_2\downarrow + 2NaHCO_3 \quad (9)$$

$$Ca(OH)_2 + Ca(HCO_3)_2 \rightleftharpoons 2CaCO_3\downarrow + 2H_2O \quad (10)$$

$$Ca(OH)_2 + Mg(HCO_3)_2 \rightleftharpoons MgCO_3\downarrow + CaCO_3\downarrow + 2H_2O \quad (11)$$

Carbonates (459.5–4480.1 mg $CaCO_3$ L^{-1}) appeared only at elevated pH (12 and 13), while bicarbonates concentration increased up to pH 10.0 (2860.4 mg L^{-1}), reducing for a concentration of 390.6 mg L^{-1} (Fig. 5C). Organic matter (10.2–43.7 %) and total phosphorus (6.4–9.6 %) removals only occurred for pH in the range of 6.0–8.0 and 11.0–12.0, respectively (Fig. 5B). Turbidity was removed in the range of 42.0–79.4 % for pH 6.0–12.0 (Fig. 5B). Total phenols were eliminated 8.6–57.4 %, presenting high efficiencies at low pH (Fig. 5B).

Acid precipitation with H_2SO_4, HNO_3 and HCl addition was not able to remove organic compounds in cheese wastewater. However, the addition of H_2SO_4, HNO_3 and HCl to raw cheese wastewater led to the

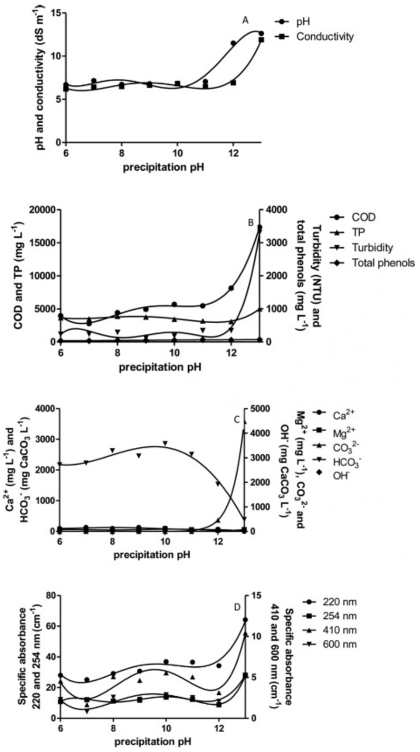

Fig. 5. NaOH precipitation for the treatment of cheese wastewater.

reduction of total phosphorus, turbidity and total phenols. Moreover, acid precipitation constitutes an inexpensive and simple process, consuming 0.4–6.3 g of acid L^{-1} of raw wastewater. Another important characteristics of acid precipitation was the formation of low sludge volume ($20-88$ mL L^{-1} of raw wastewater). A disadvantage that can occur in the acid precipitation is the presence of residual concentrations of SO_4^{2-}, NO_3- and Cl- ions in the treated effluent. However, it was verified that the application of HNO_3 to slaughterhouse wastewater reduced the NO_3- ions content in the treated effluents [42]. Basic precipitations are also cheap and simple processes. Thus, the reagent consumptions were of 0.4–3.0 g L^{-1} and 1.5–12.0 g L^{-1} for NaOH and $Ca(OH)_2$ addition, respectively. On the other hand, basic precipitation allowed the removal of COD, total phosphorus, turbidity and total phenols. NaOH addition produced $20-68$ mL sludge L^{-1} of raw wastewater, while $Ca(OH)_2$ produced $20-120$ mL sludge L^{-1} of raw wastewater. The obtained sludges are rich in organic matter and nutrients like Ca, Mg, P, Cl, Na and K [30,42].

4. Conclusions

Cheese wastewater was treated using simple and innovative chemical precipitation processes. These processes were performed with the addition of acid and basic precipitants. In the first case, H_2SO_4, HNO_3 and HCl were added to raw wastewater. $Ca(OH)_2$ and NaOH were used as basic precipitants.

Acid precipitation was capable to reduce magnesium, total phosphorus, turbidity and total phenols and absorbances at 220, 254, 410 and 600 nm. Organic matter was not eliminated by acid precipitation. HNO_3 and HCl systems allowed the removal of calcium.

Basic precipitation was effective in reducing organic matter, total phosphorus, calcium, turbidity, total phenols and absorbance at 600 nm. Magnesium was removed when NaOH was applied. This process allowed the increase of low molecular weight compounds, intensifying the biodegradability of the effluent.

The treated effluents require a post-treatment, since the COD and total phosphorus contents exceeded the limits imposed by Portuguese environmental legislation (COD = 150 mg L^{-1} and P = 10 mg L^{-1}, respectively).

Declaration of Competing Interest

The authors do not present conflicts of interest.

Acknowledgments

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Comunicação 1 - Aplicação de processos de precipitação ácida para o tratamento de águas residuais de adega

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APLICAÇÃO DE PROCESSOS DE PRECIPITAÇÃO ÁCIDA PARA O TRATAMENTO DE ÁGUAS RESIDUAIS DE ADEGA

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Resumo

Neste trabalho pretendeu-se desenvolver processos de precipitação ácida para o tratamento de efluentes de adega com vista à sua descarga ou reutilização. O efluente bruto apresentou pH próximo da neutralidade (6,8), elevados teores de matéria orgânica (carência química de oxigénio (CQO) ≈ 2003 mg L^{-1}*, carência bioquímica de oxigénio* (CBO_5) *≈ 1100 mg* L^{-1}*), fósforo total (210,1 mg* L^{-1}*), turvação (320,8 mg* L^{-1}*), e fenóis totais (27,8 mg* L^{-1}*). Aplicaram-se diferentes concentrações de precipitante ácido para atingir valores de pH entre 1-6. O processo permitiu obter remoções de CQO de 31,8-36,8%, fósforo total de 56,0-74,1%, fenóis totais de 55,6-69,4% e turvação de 88,0-93,7%. Apesar das elevadas remoções, os efluentes tratados apresentaram valores superiores ao limite de emissão para descarga.*

Palavras-chave: águas residuais industriais, contaminação, matéria orgânica, nutrientes, turvação.

Introdução

O vinho está entre as principais bebidas alcoólicas mundialmente consumidas[1], estimando-se que no ano de 2017 foram produzidos 246,7 milhões de hectolitros de vinho a nível mundial.[2] Portugal no ano de 2017 apresentou uma produção estimada de 6,6 milhões de hectolitros, revelando um aumento face ao ano de 2016[2], apresentando-se no 11.º lugar no ranking mundial[3]. Esta indústria ocupa 88% da produção total de bebidas em Portugal.[3]

O processo de vinificação inicia-se na vindima, com a colheita das uvas e encaminhamento das mesmas para a adega, este é um processo sazonal que ocorre nos meses de verão e tem uma duração aproximada de 3-4 meses[4] (dependendo do tamanho e volume de produção da adega).

Os efluentes líquidos são gerados em todas as fases do processo de vinificação, tais como, desengace e esmagamento, prensagem, clarificação, fermentação, trasfega, conservação, colagem, estabilização, filtração e engarrafamento.[1] São principalmente as lavagens de equipamentos, pavimentos, garrafas, barricas e cubas que geram maior volume de efluentes.[5] Apesar de gerar menor volume de efluente, as perdas de mosto e vinho, contribuem em grande medida para a carga orgânica do efluente bruto.[5] Estes efluentes podem atingir valores de 1-3,5 litros por cada litro de vinho produzido.[6] Quando descarregados sem serem submetidos a qualquer tipo de pré-tratamento, de forma a reduzir os seus contaminantes, os efluentes provenientes de adegas causam graves danos ambientais, ao nível do solo e águas superficiais e subterrâneas.[7]

Os efluentes provenientes de adegas apresentam elevadas quantidades de sólidos, próprios dos processos decorrentes da produção do vinho, tais como engaços, películas, grainhas, borras, resíduos e sedimentos. Assim, estes efluentes apresentam elevados teores de sólidos suspensos totais (SST=8-15000 mg L^{-1})[7,9] e nutrientes, como fósforo e azoto, com concentrações de 0,3-176 mg L^{-1} [12-15] e 4-450 mg L^{-1} [12,16], respetivamente. Adicionalmente, estes efluentes podem apresentar um pH entre 4-8[8-13], matéria orgânica elevada com valores de carência química de oxigénio (CQO) entre 120-38391 mg L^{-1} [12,14] e carência bioquímica de oxigénio (CBO_5) de 111-18360 mg L^{-1}.[13-15]

Os efluentes provenientes das adegas têm sido tratados por processos biológicos aeróbios (lamas ativadas, lagoas aeróbias, *jet-loop reactor* (JLR), *sequential batch reactor* (SBR)) e anaeróbios (digestão anaeróbia, *upflow anaerobic sludge blankets* (UASB), *anaerobic fluidized bed reactor* (AFBR), *upflow anaerobic filter* (UAF)); processos físico-químicos (precipitação química, coagulação, coagulação/floculação, eletrocoagulação, sedimentação com adição de floculantes; oxidação avançada (ozonização, radiação UV e O_3, Fenton, processo integrado de Fenton-coagulação/floculação, métodos eletroquímicos); filtração e separação por membranas (nanofiltração e osmose inversa); e processos combinados (ozonização combinada ($UV/H_2O_2/O_3$) e lamas ativadas aeróbias, oxidação combinada de solar foto-Fenton e *immobilized biological reactor* (IBR), tratamento biológico aeróbio seguido de Fenton, *sequential batch reactor* (SBR) combinado com oxidação de foto-Fenton).[17]Alguns destes tratamentos apresentam como desvantagens os elevados custos, remoções baixas e tempos curtos de vida útil das tecnologias.[17] Assim, este estudo teve como objetivo a aplicação de um precipitante ácido, através do ajuste do pH do efluente, para o tratamento de água residual de adega.

Materiais e Métodos

Recolha e caracterização do efluente bruto

O efluente estudado foi recolhido na Adega Ribafreixo Wines, situada na Vidigueira. A recolha do efluente bruto foi efetuada em Outubro de 2016, na primeira cuba da Estação de Tratamento de Águas Residuais (ETAR) da empresa, antes de sofrer qualquer tipo de pré-tratamento (Figura 1). O efluente foi armazenado em garrafões que foram posteriormente refrigerados e congelados para manutenção das propriedades do efluente. O efluente bruto apresentava uma coloração escura, odor frutado, com elevada quantidade de sólidos suspensos (Figura 2).

Figura 1: ETAR da Adega

Figura 2: Efluente Bruto

No efluente recolhido foram determinados os seguintes parâmetros: pH, condutividade, turvação, CQO, fósforo total, cálcio, magnésio, alcalinidade à fenolftaleína e alaranjado de metilo, dureza total e cálcica e absorvâncias características. Os diferentes parâmetros foram analisados segundo os métodos indicados na Tabela 1.

Tabela 1: Métodos utilizados[18]

Parâmetro	Equipamento	Método	Referência
pH	Potenciómetro Consort C 861	Potenciométrico	---
Condutividade	Condutivímetro VWR CO 3100 H	Condutivimétrico	---
Turvação	Turbidímetro WTW Turb550	Turbidimétrico	---

CQO	Placa de digestão Techne Dri-block DB 200/3 Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	19
Fósforo total	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	19
Alcalinidade à fenolftaleína	---	Volumétrico (Indicador – Fenolftaleína)	19
Alcalinidade ao alaranjado de metilo	---	Volumétrico (Indicador – Alaranjado de metilo)	19
Dureza total	---	Volumétrico (Indicador – Negro de eriocromo T)	19
Dureza cálcica	---	Volumétrico (Indicador – Calcon)	19
Fenóis totais	Espectrofotómetro Thermo Scientific Multiskan FC	Espectrofotométrico	20
Absorvâncias características			
220 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	21
254 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	19
410 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	22
600 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	23

Tratamento do efluente por adição de precipitante ácido

Neste estudo procedeu-se à aplicação de diferentes dosagens de um precipitante ácido, de forma a diminuir o pH do efluente. Em volumes de 200 mL de efluente bruto, e procedendo a agitação rápida, aplicaram-se dosagens de precipitante ácido de forma a obter pH de 6, 5, 4, 3, 2 e 1. As águas residuais tratadas foram analisadas de acordo com as metodologias indicadas na Tabela 1.

Resultados e Discussão

Caracterização do efluente bruto

O efluente proveniente da adega apresentou as seguintes características (Tabela 2): pH próximo da neutralidade (6,8), baixa condutividade (≈ 1,62 dS m^{-1}), elevados teores de matéria orgânica (CQO ≈ 2003 mg L^{-1}, CBO$_5$ ≈ 1000 mg L^{-1}), turvação ≈ 320 mg L^{-1}, fósforo total ≈ 210 mg L^{-1} e sólidos (sólidos totais (ST) ≈ 2041 mg L^{-1}, sólidos voláteis (SV) ≈ 1024 mg L^{-1}, SST ≈ 871 mg L^{-1}). Além disso, o efluente apresentou presença de cálcio e magnésio (Ca ≈ 110,8 mg L^{-1}, Mg ≈ 64,9 mg L^{-1}) e absorvâncias características (utilizando um fator de diluição de 1:50) de compostos de baixo peso molecular (0,143), compostos aromáticos e insaturados (0,093), cor (0,033) e densidade ótica celular (0,067), obtidas através dos comprimentos de onda espectrofotométricos de 220, 254, 410 e 600 nm, respetivamente.

Estes dados analíticos vão ao encontro dos descritos na bibliografia referentes às águas residuais de adega.

Tabela 2: Caracterização do efluente bruto

Parâmetro	**Unidade**	**Valor médio**
pH	escala de Sorensen	6,80 ± 0,057
Condutividade	dS m^{-1}	1,62 ± 0,064
Turvação	NTU	320,78 ± 145,063
CQO	mg L^{-1}	2003,3 ± 423,35
CBO$_5$	Mg L^{-1}	1000,0 ± 122,47
Fósforo total	mg L^{-1}	210,1 ± 24,35
Cálcio	mg L^{-1}	110,8 ± 12,86
Magnésio	mg L^{-1}	64,9 ± 21,57
Alcalinidade total	mg L^{-1} CaCO$_3$	815,6 ± 96,11
Bicarbonatos	mg L^{-1} CaCO$_3$	815,6 ± 96,11
Dureza total	mg L^{-1} CaCO$_3$	543,5 ± 82,36
Dureza cálcica	mg L^{-1} CaCO$_3$	276,5 ± 32,08
Dureza magnesiana	mg L^{-1} CaCO$_3$	267,0 ± 88,74
Fenóis totais	mg L^{-1}	25,32 ± 3,007
ST	mg L^{-1}	2041,2 ± 129,05
SV	mg L^{-1}	1024,4 ± 95,64
SST	mg L^{-1}	871,1 ± 169,78
Absorvâncias características		

220 nm	-	0,143 ± 0,0397 (1:50)[a]
254 nm	-	0,093 ± 0,0317 (1:50)[a]
410 nm	-	0,033 ± 0,0161 (1:50)[a]
600 nm	-	0,067 ± 0,0050 (1:50)[a]

[a] Fator de diluição utilizado para medir as absorvâncias características. Os dados resultaram da realização de 5 réplicas de uma amostragem

Tratamento do efluente por adição de precipitante ácido

A aplicação de diferentes dosagens do precipitante ácido levou a alterações significativas na coloração do efluente comparativamente ao efluente bruto (Figura 3). Com o abaixamento do pH (a partir do pH 3) a água foi adquirindo uma tonalidade rosada.

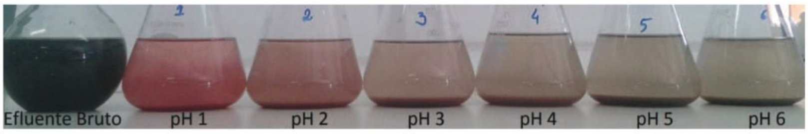

Figura 3: Efluentes bruto e tratados

A utilização de pH ≥ 2,0 permitiu obter eficiências elevadas de remoção de fenóis totais com valores superiores a 63,0% (Figura 4). O processo de precipitação mostrou-se também eficiente na remoção de fósforo total com valores de remoção superiores a 56,0%, sendo o melhor resultado obtido com pH 1, com uma remoção de 74,1% (Figura 4).

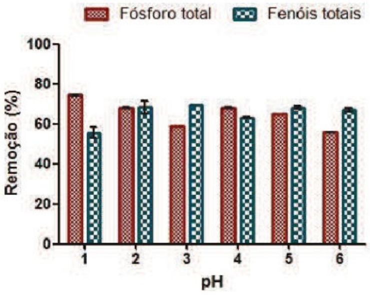

Figura 4: Efeito do pH de precipitação na remoção de fósforo total e fenóis totais

Este processo permitiu remover CQO (31,8-36,8%) e turvação (88,0-93,7%) (Figura 5). Segundo Prazeres et al. (2011), a aplicação do precipitante ácido H_2SO_4 à água residual de queijaria permitiu remoções de 12% de turvação e 21% de CQO e elevada taxa de biodegradabilidade.[24] Assim, no presente estudo foram obtidas eficiências de remoção mais elevadas relativamente à turvação e CQO.

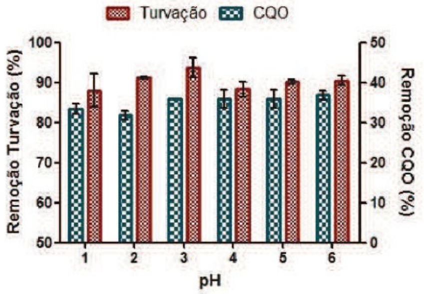

Figura 5: Efeito do pH de precipitação na remoção de turvação e CQO

Na Figura 6 é possível observar que o processo de precipitação levou à redução de compostos de baixo peso molecular (30,5-47,7%), compostos aromáticos e insaturados (36,3-76,2%), cor (25,3-66,6%) e densidade ótica celular (36,2-77,7%).

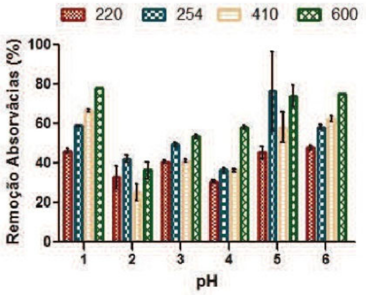

Figura 6: Efeito do pH de precipitação na remoção de absorvâncias características

A adição de um precipitante ácido leva à precipitação de compostos[24], tal como se verificou neste trabalho, com um volume de precipitado de 20 a 100 mL por litro de efluente bruto. Desta forma, ocorre uma sedimentação floculenta dos aglomerados formados pela adição do precipitante ácido, levando à remoção de matéria orgânica, SST, turvação, azoto, fósforo, proteínas totais e absorvâncias características.[24]

A formação do precipitado pode dever-se à[25]:

- Existência de espécies metálicas com propriedades anfotéricas, tal como se pode observar na equação:

$$M^{n+}(OH)_m^{(m-n)-} + (m-n)H^+ \rightarrow M(OH)_n \downarrow + (m-n)H_2O$$

- Deslocação de complexos solúveis devido à presença de óxidos ou hidróxidos insolúveis, como se representa na equação:

-

$$M^{n+}\,(L)_m^{(m-n)-} + (m-n)H^+ \xrightarrow{H_2O} M(OH)_n \downarrow + (m-n)\,LH$$

-

Presença de substâncias húmicas de diferentes solubilidades.

Conclusão

Os resultados mostram que a aplicação de um precipitante ácido no tratamento de efluentes de adega levou a remoções superiores a 50% em parâmetros como fósforo total, fenóis totais e turvação, e remoções superiores a 25% em CQO e absorvâncias características. De acordo com o Anexo XVIII do Decreto-Lei n.º 236/98 de 1 de Agosto[26], para descarga de águas residuais, os parâmetros analisados continuam com valores superiores ao limite de emissão. Desta forma, poder-se-á referir que este precipitante poderá ser utilizado como pré-tratamento que terá que ser complementado com outro tipo de tratamento. Devido à elevada taxa de biodegradabilidade que os precipitantes ácidos podem provocar no efluente tratado, sugere-se aplicação de um pós-tratamento biológico.

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CAPÍTULO V– TRATAMENTO DE ÁGUA RESIDUAL DE ADEGA POR PROCESSO DE PRECIPITAÇÃO QUÍMICA IMEDIATA EM MEIO BÁSICO E DE ETAPA ÚNICA

5.1. Processo de precipitação química imediata em meio básico e de etapa única

O processo de precipitação química imediata em meio básico e de etapa única, patenteado, Patente Nacional PT 105455/2013 para efluentes de queijaria, consiste na adição de solução de cal hidratada [$Ca(OH)_2$] de concentração de 200 g L^{-1} à água residual, em agitação (200-300 rpm), provocando a precipitação seletiva de iões da qual resulta numa abundante formação de partículas insolúveis, com capacidade de serem removidas rapidamente através de sedimentação em manto e com consequente elevação do pH (≥ 8,5). O sobrenadante resultante deste processo estabiliza naturalmente o pH por carbonatação espontânea com o CO_2 atmosférico.

A aplicação deste tratamento a águas residuais permite a remoção de moléculas orgânicas de alto peso molecular (ácidos húmicos) e moléculas aniónicas de baixo peso molecular (ácidos fúlvicos), induzindo ainda a descarbonatação do efluente (eq. 21 – 23), essa descarbonatação ocorre a pH 8,2 (Renou *et al.*, 2008, Alves 2007), com capacidade de remoção de fósforo, através da reação do $Ca(OH)_2$ com os iões fosfato, formando fosfato de cálcio insolúvel com capacidade de sedimentação (eq. 24):. Também os iões flúor têm a capacidade de precipitar sob a forma de CaF_2 (eq. 25), o amaciamento da água dá-se, devido às libertação de duas moles de hidróxido, através da reação da eq. 26:

$$Ca(OH)_2 + Ca(HCO_3)_2 \rightarrow 2CaCO_3 + 2H_2O \qquad (21)$$

$$Ca(OH)2 + Mg(HCO_3)_2 \rightarrow MgCO_3 + CaCO_3 + 2H_2O \qquad (22)$$

$$Ca(OH)2 + MgCO3 \rightarrow CaCO_3 + Mg(OH)_2 \qquad (23)$$

$$Ca(OH)_2 + HPO_4^{2-} \rightarrow CaHPO_4 + 2H_2O \qquad (24)$$

$$Ca(OH)_2 + 2HF \rightarrow CaF_2 + 2H_2O \qquad (25)$$

$$Ca(OH)_2 + CO_3^{2-} \leftrightarrows CaCO_3 + 2OH^- \qquad (26)$$

Das principais vantagens da utilização do processo de precipitação química imediata, patenteado, destaca-se: a rapidez, adaptabilidade a flutuações de caudal e concentração do efluente, baixo custo, fácil aplicação, eficácia do tratamento, com grande capacidade de desinfeção e eliminação de contaminantes e clarificação do efluente. Para além disso, dá-se o sequestro espontâneo do CO_2 atmosférico e consequentemente a contribuição para a diminuição de gases com efeito de estufa associada a tratamento de águas residuais.

O uso de soluções concentradas de cal hidratada têm sido amplamente estudadas no tratamento de efluentes industriais, tais como queijaria, vinhaça de cana de açúcar, explosivos, lixiviados de aterros sanitários e efluentes urbanos (Madeira *et al.,* 2023), porém, este processo de precipitação química básica imediata, ainda não tinha sido aplicado a efluentes vinícolas.

Este estudo pretendeu ampliar o âmbito da patente e realizou também ensaios com diferentes dosagens de óxido de cálcio (Quicklime) e hidróxido de cálcio [$Ca(OH)_2$] para além do uso de soluções concentradas (200 g L^{-1}) de cal hidratada (Slaked lime).

Os resultados obtidos foram publicados em 2 artigos e 5 comunicações orais:

Artigo 4 - Immediate one-step lime precipitation process for the valorization of winery wastewater to agricultural purposes

Comunicação 2 - Immediate one-step lime precipitation process for the valorization of winery wastewater to agricultural purposes

Comunicação 3 - Tratamento de águas residuais de adega utilizando um precipitante básico

Comunicação 4 - Redução do nível de contaminação de águas residuais de queijarias por precipitação básica

Artigo 5 - Ecotoxicological risk assessment as a decision tool for the valorisation of winery wastewater treatment byproducts to agricultural purposes

Comunicação 5 - Improving acidic soils fertility using nutrient solution and organomineral correctives obtained from winery wastewater treatment by immediate one-step lime precipitation for red pak choi production

Comunicação 6 - Restoration of soil pH for red pak choi production using organomineral fertilizer from the treatment of winery wastewater by a chemical precipitation process

RESUMO

O efluente de adega em estudo, foi recolhida no Alentejo, na Vidigueira, distrito de Beja e tratado por precipitação química imediata de uma etapa, nunca antes aplicado a efluentes vinícolas (**Artigo 4, Comunicação 2 e 3**).

Este estudo iniciou-se com a obtenção das melhores condições operatórias do processo de precipitação química imediata para efluentes vinícolas, de forma a alcançar simultaneamente a clarificação do efluente, desinfeção, remoção de matéria orgânica altamente biodegradável e remoção de fenóis. Para tal, foram estudados os seguintes reagentes, em diversas dosagens:

Óxido de cálcio - Quicklime (CaO): 5; 10; 15; 20; 25; 30; 35; 40 g L^{-1}

Cal hidratada – Slaked lime [$Ca(OH)_2$]: 5; 10; 15; 20; 25; 30; 35; 40 g L^{-1}

Hidróxido de Cálcio [$Ca(OH)_2$]: 5; 10; 15; 20; 25; 30; 35; 40 g L^{-1}

Cada dosagem foi adicionada de forma imediata, em apenas uma etapa, ao efluente bruto em agitação vigorosa, a 1000-1200 rpm à temperatura e luz ambiente, com monitorização de pH, após homogeneização, durante 2 minutos, com sedimentação durante 1 hora e posterior separação do sobrenadante e do precipitado. Este processo tem a capacidade de produzir de forma imediata, através da adição em excesso de iões cálcio, um precipitado insolúvel, com capacidade de arrastar a matéria orgânica e outros contaminantes num curto espaço de tempo (Madeira *et al.,* 2023).

De todas as dosagens aplicadas, verificou-se que, a dosagem de 25 g L^{-1} de cal hidratada (slaked lime), era a que apresentava as melhores remoções e melhores condições económicas de aplicação, com pH de reação de 12,24, condutividade de 6,11 dS m^{-1}, remoção de 21,1% de CQO e 97,6% de turvação. A dosagem de 25 g L^{-1} de cal hidratada, foi também a que obteve, de entre todas as outras dosagens, menor produção de lamas.

Esta dosagem permite ainda remover 87,1% de P_{total}, 99,9% de fenóis totais, elimina mais de 97% de todos os metais estudados (zinco, ferro, manganês e cobre) e eliminar na totalidade a os coliformes totais.

Após obtenção da melhor dosagem, o sobrenadante foi exposto ao CO_2 atmosférico, por um período de 15 dias, sendo que ao 13º dia se obteve um pH de 7,46 devido ao estabelecimento do equilíbrio CO_2(g) CO_2 (aq.). Demonstrando assim que este tratamento

pode contribuir para a remoção de gases com efeito de estufa, devido ao sequestro do CO_2 atmosférico por carbonatação espontânea. Atendendo ainda à produção anual de água residual da indústria vinícola em Portugal poderia recuperar-se 66 ton de P_{total} durante 1 ano se aplicado o processo de precipitação química imediata no tratamento destes efluentes.

O mesmo processo foi aplicado ao efluente de queijaria, com dosagens para obtenção de pH de 6,0; 7,0; 8,0; 9,0; 10,0; 11,0; 12,0 e 13,0 (**Comunicação 4**), onde também foi possível confirmar a eficiência de remoção do processo de precipitação química em meio básico numa etapa, uma vez que, se obtiveram remoções máximas de P_{total} de 96% turvação 99%, e 30% de CQO, as melhores eficiências verificaram-se nos pH superiores a 10.

Com o fim de obter uma água tratada e lamas compatíveis com a sua reutilização agrícola foi analisado o potencial de reaproveitamento dos sobrenadantes e precipitados para a produção de alimentos. Assim realizou-se o estudo do impacto ambiental desta água, avaliando o potencial de reaproveitamento do sobrenadante, através de avaliação físico-química, microbiológica e ecotoxicológica. Realizou-se também a avaliação ao potencial de reaproveitamento do precipitado como corretivo organomineral, através de avaliação físico-química, microbiológica e ecotoxicológica e, ainda, a aplicação dos sobrenadante e precipitado em produção de alimentos, na cultivar red pak choi choi (*Brassica rapa* L. var. *chinensis* (L.) Kitam), através de irrigação e correção do solo agrícola (**Artigo5, Comunicação 5 e 6).** Neste estudo, foi avaliado o impacto ecotoxicológico dos subprodutos da precipitação química imediata através dos bioindicadores: *Thmanocephalus platyurus* (bioensaio de mortalidade após 24h), *Vibrio fisheri* (inibição da luminescência após 30 min) e *Pseudokirchneriella subcapitata* (inibição do crescimento após 72h) no sobrenadante, e solo agrícola utilizado para a produção de alimentos.

O sobrenadante foi posteriormente diluído com água subterrânea, em 1:4, de forma a ajustar o pH e a condutividade para rega, convertendo-se assim em solução nutritiva.

O ensaio experimental em solo foi realizado com recurso a delineamento experimental casualizado, nos meses de maio a julho, na cultivar red pak, r=9, utilizando 1,5 kg de solo ácido, em 3 condições distintas:

4- Controlo: Solo ácido e rega com água subterrânea;

5- Fertirrigado com solução nutritiva: solo ácido fertirrigado com sobrenadante tratado e diluído (1:4);

6- Solo corrigido com fertilizante organomineral: solo ácido corrigido a pH 7,2 com precipitado seco, a uma dosagem ideal de 1,2% (p/p) e regado com água subterrânea.

Todas as condições estudadas tiveram sucesso no crescimento das plantas, porém, o solo corrigido com o corretivo organomineral obteve melhores características na altura da planta, cumprimento/ área folear. As plantas irrigadas com a solução nutritiva apresentaram o melhor resultado no diâmetro do caule. Em ambas as condições em estudo, a planta apresentou maior massa de clorofila a, b e Carotenoides face ao solo controlo, revelando um maior desenvolvimento e capacidade produtiva da planta. Demonstrando assim a eficácia da reutilização da totalidade dos elementos provenientes deste tratamento o que permitirá habilitar as adegas a serem indústrias de desperdício zero e promovendo a economia circular com valorização dos seus resíduos.

Os ensaios ecotoxicológicos permitiram-nos analisar o risco para o ecossistema aquático, pela utilização da solução nutritiva na rega e do precipitado no solo. Foi estudado o efluente bruto, revelando indicadores ecotoxicológicos letais muito tóxicos para os 3 bioindicadores, com classe 4 – toxicidade muito alta, constituindo um grave perigo para o recetor ambiental. O sobrenadante resultante da aplicação de 25 g L^{-1} de cal hidratada (slaked lime) no processo de precipitação química imediata apresentou uma toxicidade para classe 2, com menor toxicidade aguda para todos os bioindicadores utilizados. Após preparada a solução nutritiva (sobrenadante com diluição 1:4), os resultados ecotoxicológicos revelaram uma redução da toxicidade para classe 1, tornando-a assim, não tóxica. Após irrigado o solo com esta solução nutritiva, não revelou qualquer aumento de toxicidade. Revelando que esta solução não aporta risco para o ambiente ou para o alimento que está a ser produzido.

Artigo 4 - Immediate one-step lime precipitation process for the valorization of winery wastewater to agricultural purposes

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IMPACTS IN ENVIRONMENTAL TRENDS, HEALTH AND WELL BEING: A GLOBAL POLLUTION PROBLEM

Immediate one-step lime precipitation process for the valorization of winery wastewater to agricultural purposes

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Abstract
The winery wastewater (WW) of the Village of Vidigueira, in south of Portugal, presented an acid pH (4.26 ± 0.029), a high content of chemical oxygen demand (COD = 3236 ± 30.43 mg L^{-1}), turbidity (160.0 ± 7.68 NTU), total P (155.5 ± 6.36 mg L^{-1}) K (100.5 mg L^{-1}), and low level of metals: Pb, Cd, Cu, Ni, Mn, Fe, Zn. The K and the total P content could make them attractive from an agricultural point of view. However, the high turbidity biochemical oxygen demand (BOD_5 = 1266.7 ± 208.7 mg O_2 L^{-1}) fecal coliforms (45 ± 29.8 ufc/100 mL) and toxicological total phenols 13.239 ± 3.007 mg galic acid L^{-1} is the main limiting parameters. By using 5 g L^{-1} of lime solution, the best reactional conditions are achieved: pH = 12 with a minimum volume of the produced sludge's, 33.5 ± 4.92 mL L^{-1}. These reactional conditions allow reaching high removal levels of BOD_5, turbidity, total P, and total phenols: 77.9, 98.7%, 87.1%, and 99.9%, respectively. Simultaneously, the fecal coliforms and *Enterococcus* are 100% eliminated. The precipitate obtained is rich in calcium (2.7891 ± 0.4350 mg Ca kg^{-1}) organic matter (10.05 ± 0.11%), P (895.0 ± 35.4 mg P_2O_5 kg^{-1}), and K (990.0 ± 84.9 mg K_2O kg^{-1}). During carbonatation reactions, the pH (7.46) and electrical conductivity (EC) (1.805 dS m^{-1}) decreased spontaneously while the Ca^{2+}, K^+, and F^- kept high: 426.53 mg L^{-1}, 240.53 mg L^{-1}, 176.03 mg L^{-1}, respectively.

Keywords Carbonatation · Lime · Nutrient solution · Precipitation · Wine industry

Introduction

The wine industry is an industry with a great development worldwide (Petruccioli et al. 2000; Amor et al. 2019). A total of 61% of world wine production comes from the European countries, especially Mediterranean countries (Bolzonella et al. 2019) like Portugal which in 2019 produced 6.5 million hectoliters/year (IVV, 2019). The winemaking process uses water in the different steps: grape reception, destemming/crushing, pressing, clarification, fermentation, transfer, preservation, sizing, cold stabilization, filtration, and bottling, depending on the type of wine (Vlyssides et al. 2005; Brito et al. 2007). These uses of water during wine production give rise to the production of winery wastewater (WW), being estimated that for each liter of wine produced, 1–14 L of wastewater are generated (Domínguez et al. 2014; Ioannou et al. 2015). WW is characterized by high organic content, such as alcohols, sugars, organic acids, polyphenols, lignins, and tannins; and in smaller quantities, the detergents and disinfectants used in the cleaning and sanitation processes are present (Arienzo et al. 2009; Solís et al. 2017; Amor et al. 2019). Consequently, the WW represents serious environmental problems for soil, surface, and groundwater, like changes in oxygen, pH, color, temperature, turbidity, eutrophication, and addition of toxic

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Table 1 Bibliographic review of soils effects by the application of WW

WW type	WW Characteristics									Application time	Soils effects	References
	pH	EC	Na	K	Ca	SAR	Mg	Total phenols	COD			
Raw	---	---	99	240	---	---	---	0.19	13,000	30 years	Increasing inorganic species nature of the carbon present	Mosse et al. (2012)
Treated with SBR*	---	---	112	150	---	---	---	< 0.04	610	Single	Impacts on soil respiration, nitrogen cycling microbiological community	
Diluted	6.0	1.098	114.9	186.6	18.0	5.6	8.4	---	---	Single	Promoted urease and β-glucosidase's activities	Mulidzi et al. (2015)
Diluted	5.4	1.04	84.6	196.4	22.4	4.1	9.1	---	3210	Single	Suppressed phosphatase promoted urease and β-glucosidase's	Mulidzi et al. (2016)

* Concentrations in mg L^{-1}, except pH, EC (mS/cm), and SAR (adimensional)

products (Coetzee et al. 2004; Ioannou et al. 2015; Bolzonella et al. 2019). However from an agricultural point of view (Prazeres et al. 2017), the WW contains macro and micronutrients, like Ca, Mg, K, P, Cu, Fe, and water that are essential for plant growth (Jiménez-Cisneros 1995; Mosse et al. 2010; Prazeres et al. 2014). In the research literature, it is possible to find a few agricultural applications of treated WW, either as soil applications (Table 1) or as irrigation (Table 2). For example, according to Mosse et al. (2012), the application of WW in soil during a long-term period (30 years) lead to soil effects related to inorganic species and the nature of the carbon present. A single soil application (Mosse et al. 2012) showed significant impacts on soil respiration, nitrogen cycling, and soil microbiological community. Christen et al. (2010) applied WW in a land filter for pasture that was able to neutralize the pH and remove nutrient pollutants according to EPA limits. Stewart and Flinn (1984) used raw WW with a pH of 4.7 for *Eucalyptus* irrigation during 4 years obtaining good growth rates. Even the use of diluted WW (until 3 g L^{-1} COD) promoted urease and β- glucosidase's activities and suppressed phosphatase (Mulidzi et al. 2015). The effects of the use of treated WW for vine irrigation was tested by Hirzel et al. (2017) who observed no impact on the soil. However, these authors experienced a small accumulation of Na and Mg in leaves and the decreasing of Ca and K. The grapes did not show ion accumulation and no sensory was perceived in wines. The use of WW treated with SBR system with low COD (0,610 g L^{-1}) caused no significant differences in nitrogen and carbon cycling (Mosse et al. 2012). In addition, Oliveira et al. (2009) concluded that the WW, after biological treatment with air micro-bubble bioreactor (AMBB), showed some evidences highlighting the suitability to crop irrigation according to the results obtained by the bioassays with *Lepidium sativum L.* seeds. Nonetheless, the existing biological treatments require the addition of nutrients and is not adapted to the seasonality of this industry (Ioannou et al. 2015), requiring tuning treatment to be used for agricultural purposes. Consequently, it is necessary to develop an easy, cheap, and suitable process, adaptable to the seasonality and specificity of winery industry. Besides, being able to help them to achieve a circular economy by the production of agricultural reusable treated wastewater and sludge. The immediate chemical precipitation process (Prazeres et al. 2019; Madeira et al. 2020; Correia et al. 2020) consists in the use of a single reactant, lime, able to clarify, disinfect, recycle nutrients, and create conditions to capture CO_2 from the atmosphere by spontaneous reactions.

The main objectives of this paper is the development of reactional conditions of the immediate chemical precipitation process to achieve simultaneously the effluent clarifying, disinfection, high biodegradable organic matter removal, and as a novelty, the total phenols removal.

Table 2 Bibliographic review of physical chemical characterization of WW used for irrigation

WW type	Culture	pH	EC	Na	K	Ca	SAR	Mg	Total P	Total N	BOD	COD	NH4$^+$	Phenolic compounds	N03$^-$	References
Raw	eucalypts	4.7	1.560	---	---	---	---	---	---	---	---	---	---	---	---	Stewart and Flinn (1984)
Treated with AMBB[a]	Seed germination bioassays	7.7–8.8	0.75–1.050	---	---	---	27	---	0.6	2.0	---	40–200	---	0.4–0.6	---	Oliveira et al. (2009)
Raw	Pasture	5	3.4	200–933	645	---	---	---	---	---	2000	---	10.3	---	5	Christen et al. (2010)
Treated whith bioreactor + NaOH to neutralize	Vine	9.09	2.38	424.7	129.6	20.9	21	24.9	---	---	---	---	2.3	---	12.0	Hirzel et al. (2017)
Aeration ponds + filtered + K$^+$ enriched	Vine	7.48	0.428	134.5	109.0	36.1	5.7	28.9	---	---	---	---	N.D.	---	2.53	

[*] Concentrations in mg L^{-1}, except pH, EC (mS/cm), and SAR (adimensional)

[a] *AMBB*, air micro-bubble bioreactor

[b] Land filter, filtration and irrigated cropping for land treatment and effluent reuse

Materials and methods

Winery wastewater sampling

The WW under study was collected in Alentejo, south of Portugal, at the Vidigueira village. This winery features more than 100 ha of vines under integrated production. The demarcated region of Vidigueira is known for its microclimate, ideal for the production of white grape varieties, with the production of white wines being the excellence (Carrera 2019).

The sampling took place in the vintage period of 2018 at the winery wastewater treatment plant in the raw WW storage tank. The WW samples were collected in only 1 day and sent to the laboratory, in 5 L polyethylene (PET) containers, subjected to refrigeration at 4 °C or freezing at − 20 ° C with acidification, for specific metal analysis, in order to avoid alteration of properties.

Experimental set-up

Immediate one-step lime precipitation process

The immediate one-step lime precipitation process was carried out by using 1000 mL of raw winery wastewater and testing three lime substances: quicklime (CaO) (≥ 97 %, M = 56,07 g mol^{-1}, was supplied by Merck), slaked lime (Ca (OH)$_2$) (suitable for water treatment, according to NP EN 12518), and calcium hydroxide Ca (OH)$_2$ (≥ 95 %, M = 74,09 g mol^{-1}, was supplied by Panreac AppliChem) reagent grade. The lime solutions have a concentrations of 200 g L^{-1}. So different volumes of the concentrated solution (1–50 mL for hydrated lime and calcium hydroxide, and 5–40 ml L^{-1} for the calcium oxide) were used. These reagents were added according to Fig. 1a, to a graduated borosilicate glass container: the raw WW (1) was placed in a shake plate (2), with constant pH monitoring (2), under vigorous stirring (4) for 2 min followed by the sedimentation process (B) for 1 hour. After this time, the sludge (5) and supernatant (6) were separated (C). The supernatant (C) was exposed to atmospheric CO$_2$, monitored daily for pH and conductivity until pH stabilization close to the neutrality was achieved.

The immediate one-step lime precipitation process was conducted by the application of one reactant that produces immediately an abundant and insoluble precipitate able to sweep the organic matter and other contaminants from wastewater in a short time. It also had the objective of adding excess calcium ions to cause an instantaneous precipitation of calcium and magnesium salts, in order to create conditions to obtain zone settling.

This process entails the introduction of the whole lime solution at once, to the WW in a state of vigorous agitation (≈ 1000–1200 rpm) that generates an instantaneous precipitate

(Madeira et al. 2020). During all the immediate one-step lime precipitation, the pH is monitorized.

The application of the lime-based precipitant provides decarbonation of the effluent, leading to the precipitation of carbonate ions (Renou et al. 2008). The reaction mechanism is described in Eqs. (1–3). The chemical clarification of the effluent is due to the coagulation of particles, colloidal, and suspended, promoting sedimentation and sludge formation (Semerjian and Ayoub 2003). These reactions are described in Eqs. (1, 4–6):

$$Ca(OH)_2 + Ca(HCO_3)_2 \rightleftarrows 2CaCO_3\downarrow + 2H_2O \quad (1)$$

$$Ca(OH)_2 + Mg(HCO_3)_2 \rightleftarrows MgCO_3\downarrow + CaCO_3\downarrow + 2H_2O \quad (2)$$

$$Ca(OH)_2 + MgCO_3 \rightleftarrows CaCO_3\downarrow + Mg(OH)_2\downarrow \quad (3)$$

$$Ca(OH)_2 + H_2CO_3 \rightleftarrows CaCO_3\downarrow + 2H_2O \quad (4)$$

$$3Ca(OH)_2 + 2PO_4{}^{3-} \rightleftarrows Ca_3(PO_4)_2\downarrow + 6OH^- \quad (5)$$

$$4Ca(OH)_2 + 3PO_4{}^{3-} + H_2O \rightleftarrows Ca_4H(PO_4)_3\downarrow + 9OH^- \quad (6)$$

Precipitate formation occurs (due to the increase of Ca^{2+} ions and inorganic carbon $HCO_3{}^-$) with reduction of organic matter, turbidity, total phosphorus, SST, conductivity, calcium, and magnesium (Renou et al. 2008; Prazeres et al. 2013).

Atmospheric CO_2 neutralization process

For the atmospheric CO_2 neutralization process, the supernatant was separated from the sludge (Fig. 1c), because there is a decrease in pH, after this separation, according to Madeira et al. (2020). Only the supernatant from the best-studied condition was exposed to this process, for 15 days, at room temperature, with daily pH and conductivity monitorizations (Fig. 2).

Calcium and magnesium hardness (Table 4) were determined at the end of 15 days of exposure, when the pH and conductivity were already stable and there was no variation over 3 days.

Analytical determinations

Wastewater

The pH was measured by the potentiometric method using an inoLab WTW Potentiometer and a SenTix 41 electrode. The EC was measured on a METROHM 660 conductivity meter. Turbidity was analyzed on a WTW Turb550 turbidity meter. The COD parameter was determined by dichromate reflux method (APHA et al. 1998), with digester WPA HC 6016 and read on a UV-visible spectrophotometer at a wavelength of 600 nm. Total phosphorus was digested in acid medium with color development by vanadomolibdophosphoric acid colorimetry monitored on a UV-visible spectrophotometer at a wavelength of 470 nm (APHA et al. 1998). BOD5 was determined by the respirometric method through the WTW's Oxitop® gauge system (APHA et al. 1998). Calcium and magnesium were determined by volumetric complexation method using EDTA and Eriocromo T black indicators and Calcon indicators (APHA et al. 1998). Total phenols were determined by the Folin-Ciocalteu method, with gallic acid as the standard and spectrophotometrically determined on a Thermo Scientific Multiskan FC spectrophotometer (Falleh et al. 2008). Heavy metals were determined by flame atomic absorption spectrometry using Varian SpectraAA 220FS spectrometer (APHA et al. 1998). Kjeldhal nitrogen was determined by the Kjeldhal method (APHA et al. 1998). Ammonia nitrogen after sample distillation (adjusted to pH 6–7.4), was released into a Tashiro indicator boric acid solution and titrated with hydrochloric acid. The total content of metals (Pb, Cd, Zn, Fe, Ni, Mn, Cu) was determined by digesting the unfiltered sample (APHA et al. 1998) by flame atomic absorption spectrometry with the Varian device (SpectrAA 220FS).

Sludge

pH and the EC were monitored by a potentiometer with combined glass electrode (inoLab WTW) and conductivity meter

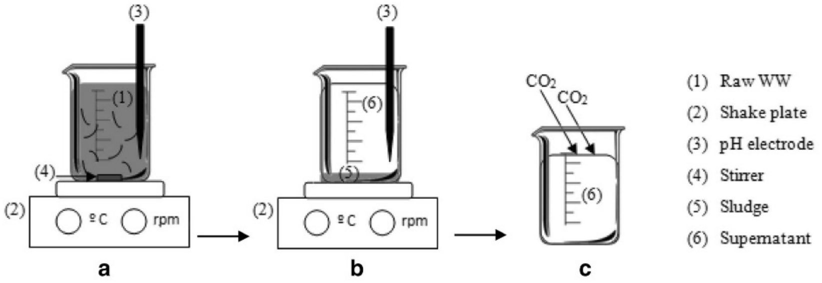

Fig. 1 Experimental set-up for immediate one-step lime precipitation

(METROHM 660), respectively, this process consists of diluting the dry mass (sludge) at 20 °C in a suspension of distilled water in the proportion of 1:10 (m/v), mixing occasionally for 1 h and after that 1 h of settlement.

Total phosphorus was monitored by colorimetry with phosphoric acid molybdovanadate (Kitson and Mellon 1944).

The determination of the Pb, Cd, Zn, Fe, Ni, Mn, Cu, Ca, and Mg was carried out by atomic absorption spectrometry after digestion with aqua regia (EN 13346 2000). The digestion of the samples was carried out using a mixture of HCl (37%) and HNO_3 (70%), in the proportion of 3:1 (v/v), at room temperature for 16 h and then, 2 h at 130 ° C, under reflux conditions. After digestion, the suspension was filtered and diluted to 100 mL with 0.5 M HNO_3. These metals were later determined by flame atomic absorption spectrometry with the Varian device (SpectrAA 220FS).

The density was obtained experimentally and determined by weighing a certain volume of sludge.

Results and discussion

Winery wastewater characterization

WW present a low conductivity of 1.243 ± 0.034 dS m^{-1}, and high turbidity of 159.77 ± 4.59 NTU and a dark brown color, with an intense fruity smell from the winemaking process, characteristic of the harvest season. As shown in Table 3, the WW pH is acid 4.26 ± 0.03 similar to that observed by Amor et al. (2019), Flores et al. (2019), in the same harvest period. WW can cause problems in the environment, discharge, due to the COD, this being 3225.56 ± 25.46 mg O_2 L^{-1}, BOD_5 1266.7 ± 208.2 mg O_2 L^{-1}, and a BOD_5/COD ratio of 0.39. Metals such as Ni, Cd, and Pb present values below the detection limit of the method. Zn, Fe, and Cu are elements with higher values of 3.150 ± 0.071 mg L^{-1}, 5.725 ± 1.591 mg L^{-1}, and 0.536 ± 0.557 mg L^{-1}, respectively. These elements are also found in studies on the same effluent (Amor et al. 2019; Guimarães et al. 2019).

Fecal coliforms and *Enterococcus* were also detected in WW, 45 ± 29.8 ufc/100 mL and 3.7×10^{-3} NMP/mL, respectively, but lower than observed by Candia-Onfray et al. (2018).

Immediate one-step lime precipitation process

The immediate one-step lime precipitation reactions was done with quicklime (CaO), slaked lime ($Ca(OH)_2$), and calcium hydroxide $Ca(OH)_2$ reagent grade, and was described in the "Experimental set-up" section. After treatment, the pH, conductivity, COD, calcium, magnesium, and sludge volume were determined.

The initial pH of the effluent is 4.26; after applying the various dosages of the three reagents tested, it was found that, in the slaked lime, there was a sudden increase in pH from 4.97 to 10.27 at doses from 2 to 3 mL; this fact can be explained by the addition of OH^- ions in solution on the effluent (Renou et al. 2008), the pH stabilized with a volume added of 35 mL reaching a pH of 12.29. In calcium hydroxide, pH stabilized after addition of 30 mL with a final pH of 12.56, and when using quicklime, a volume of 30 mL was necessitated with a final pH of 12.70, EC showed the same trend, see Fig. 3a. This increase is due to the increase in Ca^{2+} in solution, due to the excess of reagent applied, also causing the removal of organic carbon (Renou et al. 2008).

The sludge volume generated can be seen in Fig. 3b. The COD shows a decreasing evolution, according to the increase in the volume of the added precipitant (Fig. 3c).

As we can see in Fig. 3d, with the addition of 25 mL of concentrated lime to WW, it is possible to achieve maximum removal of Ca, Mg, and COD simultaneously, generating a smaller volume of sludge (Fig. 4).

The effects of lime reagents in the immediate one-step lime precipitation process and taking into account the maximum COD removal and minimum sludge volume produced slaked lime at the dosage of 25 mL L^{-1} was selected. These differences obtained may be due to the higher amounts of calcium ions in solution in the treatment with slaked lime.

This treatment benefits from reduced costs, since, according to Lee et al. (2014), 1 ton of lime costs 130€, so this treatment would cost 0.65€/m^3.

Atmospheric CO_2 neutralization

Spontaneous carbonation process allowed to lower the pH (7.46) and EC (1.805 dS m^{-1}) to values compatible with agricultural purposes (Table 4). Hence, there is no need to dilute the supernatant or adding other reagents to accomplish this process.

The neutralization process can be explained because atmospheric CO_2 dissolves in water, establishing the balance (Eq. 7), combining with water molecules, forming carbonic acid

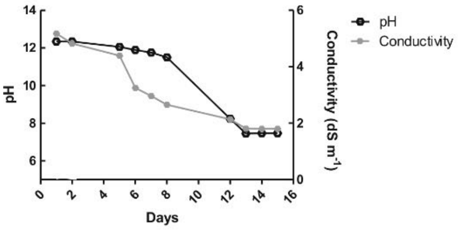

Fig. 2 Evolution of supernatant characteristics for atmospheric CO_2 neutralization process

(Eq. 8), decomposing naturally in bicarbonate and carbonate molecules, producing hydrogen ions (Eqs. 9 and 10) (Madeira et al. 2020).

$$CO_2\ (g) \rightleftarrows CO_2\ (aq) \tag{7}$$

$$CO_2\ (aq) + H_20\ (l) \rightleftarrows H_2CO_3\ (aq) \tag{8}$$

$$H_2CO_3 \rightleftarrows H^+ + HCO_3^- \tag{9}$$

$$HCO_3^- \rightleftarrows H^+ + CO_3^{2-} \tag{10}$$

According reactions Eqs. (7) and (10), one mole of CO_2 (g) is responsible for the production of 1 mol of CO_3^{2-} that react with Ca_2^+ ions existent in solution. Comparing the amount of calcium to Tables 4 and 5 is possible to conclude that for each liter for treated water is possible sequester 80.74 mg L^{-1} of the atmospheric CO_2.

Effluent characteristics

The immediate one-step lime promoted a significant increase in pH, going from acid to basic (Table 5). COD decreased 22.2%, total P 87.1%, and total phenols 99.9%. *Escherichia coli*, fecal coliforms, and *Enterococcus* were completely eliminated from water. Additionally, zinc, iron, manganese, and copper showed 98.1%, 99.5%, 97.1%, and 100% elimination, respectively. Akinwekomi et al. (2016), which used $Mg(OH)_2$/$Mg(HCO_3)_2$ and then $Ca(OH)_2$ to treat acid mine drainage, and obtained removal of 100% iron(II), manganese, copper, and nickel; aluminum and cobalt > 90%; and lead > 80%.

As we can see in the Table 5, parameters such as N-NH_4^+ (5.38 ± 0.00 mg NH_4 L^{-1}), total P (20.3 ± 8.65 mg P L^{-1}), Na (45.42 ± 14.76 mg Na^+ L^{-1}), K (240.53 ± 46.06 mg K L^{-1}), and Ca (499.76 ± 84.46 mg Ca L^{-1}) were within the parameter range, also presented in the other nutrient solutions claimed by Prazeres et al. (2017) (1.3–24.18 mg NH_4 L^{-1}, 0.6–161.92 mg P L^{-1}, 0–1042 mg Na^+ L^{-1}, 15–368.85 mg K L^{-1}). Prazeres et al. (2020) used of $Ca(OH)_2$ in the treatment of cheesemaking wastewater, with dosages of various pH, obtained removal of matter of 29.7%, for the dosage corresponding to pH 11 and 61.9–95.6% of total phosphorus. Guerreiro et al. (2020) also used $Ca(OH)_2$ in cheese manufacturing wastewater, in a working pH range of 6 and 7, where the best results obtained were 89–97% total phosphorus.

The treated WW contains nutrients that promote plants growth, replace the commercial nutrient solutions, and combat the consumption of drinking water for agriculture, the principal global consumer of fresh water (Prazeres et al. 2017).

Precipitate characteristics

The immediate one-step lime precipitation at optimal dosage, 25 ml L^{-1} of slaked lime, gave rise to a white precipitate, with a density of 1.45 ± 0.09 g m^3, with a basic pH of 12.40 ± 0.02, mainly due to basic treatment conditions used. The high pH can be used as a soil corrector, as well as a toxicity corrector (Prazeres et al. 2016). This reuse of sludge for agricultural purposes is increasingly an option adopted by several countries (Dowling et al. 2015).

Other properties of the best reaction condition of precipitate were also analyzed and are present in Table 6, such as the content of heavy metals, which, according to Decree Law no. 276/2009 and the European Council Directive 86/278/EEC, the limit values for the concentrations of heavy metals present in the studied sludge. The best reaction condition of precipitate presents values considerably lower than those allowed by law, in all metals, as well as, in the legislated values for sludge intended for application in soils of agricultural products.

Table 3 Winery wastewater characterization

Parameter	Units	Value
pH	Sorensen scale	4.26 ± 0.03
Conductivity	dS m^{-1}	1.243 ± 0.034
Total hardness	mg $CaCO_3$ L^{-1}	424.78 ± 53.1
Calcic hardness	mg $CaCO_3$ L^{-1}	18.59 ± 13.8
Magnesium hardness	mg $CaCO_3$ L^{-1}	406.19 ± 54.86
Calcium	mg L^{-1}	7.43 ± 5.52
Magnesium	mg L^{-1}	98.71 ± 13.33
COD	mg O_2 L^{-1}	3225 ± 25
BOD_5	mg O_2 L^{-1}	1266 ± 208
BOD_5/COD	-	0.39
Turbidity	NTU	159.77 ± 7.68
Ammoniacal nitrogen	mg NH_4 L^{-1}	0.72 ± 0.00
Kjeldahl nitrogen	mg N L^{-1}	22.3 ± 1.6
Total phenols	mg galic acid L^{-1}	13.239 ± 3.007
Total phosphorus	mg P L^{-1}	155.69 ± 6.53
Chlorides	mg L^{-1}	124.7 ± 1.5
Nitrates	mg NO_3^- L^{-1}	5.9 ± 0.4
Fluorides	mg L^{-1}	166.7 ± 2.3
Sulfates	mg SO_4^{2-} L^{-1}	95.4 ± 0.5
Lead	mg Pb L^{-1}	<DL
Cadmium	mg Cd L^{-1}	<DL
Zinc	mg Zn L^{-1}	3.150 ± 0.071
Iron	mg Fe L^{-1}	5.725 ± 1.591
Nickel	mg Ni L^{-1}	<DL
Manganese	mg Mn L^{-1}	0.173 ± 0.018
Copper	mg Cu L^{-1}	0.536 ± 0.557
E. Coli	ufc/100 mL	0
Fecal coliforms	ufc/100 mL	45 ± 29.8
Enterococcus	NMP/mL	$3.7x10^{-3}$

One collection of wastewater. Number of measurements = 3

DL, detection limit

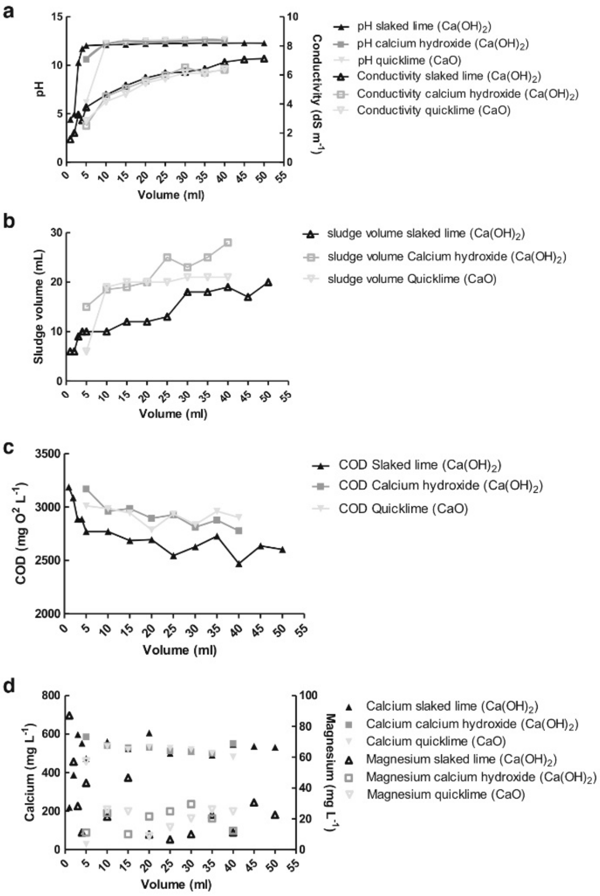

Fig. 3 Immediate one-step lime precipitation characteristics. **a** pH and conductivity. **b** Sludge volume. **c** COD. **d** Calcium and magnesium

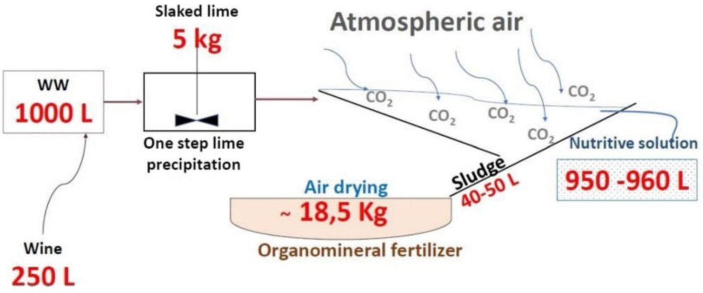

Fig. 4 Outline of the immediate one-step lime process

Table 4 Atmospheric CO_2 neutralization characteristics

Parameter	Units	Value
pH	Sorensen scale	7.46
Conductivity	$dS\ m^{-1}$	1.805
Calcic hardness	$mg\ CaCO_3\ L^{-1}$	1248.7
Magnesium hardness	$mg\ CaCO_3\ L^{-1}$	27
Calcium	$mg\ L^{-1}$	426.5
Magnesium	$mg\ L^{-1}$	6.6

Macronutrients such as total P and K have very high values, from 895.0 ± 35.4 mg P_2O_5 kg^{-1} to 990.0 ± 84.9 mg K_2O kg^{-1}. The percent organic matter is 10.05 ± 0.11. These elements are very important for this product to be valued and used in agriculture, since their presence in the soil is fundamental for the growth of plants (Carvalho et al. 2013; Mateo-Sagasta et al. 2015).

Most countries in the European Union bet mainly on the reuse of sludge for agricultural purposes. This application on the ground is legislated and must strictly comply with the directives, so as not to become a human and environmental health problem (Kelessidis and Stasinakis 2012). Worldwide, it is the countries of Europe and the USA that most use sludge from wastewater treatment plants for agricultural purposes, closing the deficit in soil organic matter (Mateo-Sagasta et al. 2015).

Table 5 Effluent characteristics

Parameter	Units	WW treated
pH	Sorensen scale	12.41 ± 0.24
Conductivity	$dS\ m^{-1}$	6.545 ± 0.615
Total hardness	$mg\ CaCO_3\ L^{-1}$	1804.97 ± 350.75
Calcic hardness	$mg\ CaCO_3\ L^{-1}$	1064.82 ± 210.74
Magnesium hardness	$mg\ CaCO_3\ L^{-1}$	740.15 ± 241.24
Calcium	$mg\ L^{-1}$	499.76 ± 84.46
Magnesium	$mg\ L^{-1}$	179.89 ± 58.63
COD	$mg\ O_2\ L^{-1}$	2508.89 ± 165.55
BOD_5	$mg\ O_2\ L^{-1}$	280 ± 0.00
BOD_5/COD	-	0.11
Turbidity	NTU	2.00 ± 0.84
Ammoniacal nitrogen	$mg\ NH_4\ L^{-1}$	5.38 ± 0.00
Kjeldahl nitrogen	$mg\ N\ L^{-1}$	7.58 ± 1.54
Total phenols	mg galic acid L^{-1}	0.006 ± 0.003
Total phosphorus	$mg\ P\ L^{-1}$	20.3 ± 8.65
Chlorides	$mg\ L^{-1}$	121.861 ± 23.78
Nitrates	$mg\ NO_3^-\ L^{-1}$	12.642 ± 2.652
Fluorides	$mg\ L^{-1}$	176.029 ± 11.143
Sulfates	$mg\ SO_4^{2-}\ L^{-1}$	56.528 ± 14.332
Sodium	$mg\ Na^+\ L^{-1}$	45.42 ± 14.76
Potassium	$mg\ K\ L^{-1}$	240.53 ± 46.06
Lead	$mg\ Pb\ L^{-1}$	<DL
Cadmium	$mg\ Cd\ L^{-1}$	<DL
Zinc	$mg\ Zn\ L^{-1}$	0.06 ± 0.004
Iron	$mg\ Fe\ L^{-1}$	0.030 ± 0.009
Nickel	$mg\ Ni\ L^{-1}$	<DL
Manganese	$mg\ Mn\ L^{-1}$	0.005 ± 0.007
Copper	$mg\ Cu\ L^{-1}$	<DL
E. Coli	CFU/100 mL	0
Fecal coliforms	CFU/100 mL	0
Enterococcus	MPN/mL	0

One collection of wastewater. Number of measurements = 3

Conclusion

The production of 1 L of wine has associated a production of 4 L of strong purplish and fruit smelling winery wastewater. The winery wastewater presents a high fluctuation in flow and composition, lack of nutrients, namely phosphorus and nitrogen; presence of recalcitrant compounds, i.e., organic acids (tartaric, malic, citric, lactic) and phenolic acids (benzoic, cinnamic) that are responsible for the incomplete biological conventional treatments. This research proposes a low-cost and an easy-to-apply immediate one-step lime precipitation process that uses a single reactant able to be recovered. Consequently, this process have potentialities to be used for all kind of winery industries, helping them to achieve a zero discharge system and a transition for a circular economy. Additionally, it contributes to the reduction of the greenhouse

Table 6 Precipitate characterization

Parameter	Units	Value
Sludge volume	$mL\ L^{-1}$	33.5 ± 4.92
Density	$g\ cm^3$	1.45 ± 0.09
pH	Sorensen scale	12.40 ± 0.02
Conductivity	$mS\ cm^{-1}$	7.25 ± 0.15
Organic matter	%	10.05 ± 0.11
Phosphorus	$mg\ P_2O_5\ kg^{-1}$	895.0 ± 35.4
Potassium	$mg\ K_2O\ kg^{-1}$	990.0 ± 84.9
Pb	$\mu g\ kg^{-1}$	0.50 ± 0.0
Cd	$\mu g\ kg^{-1}$	0.03 ± 0.0
Zn	$\mu g\ kg^{-1}$	0.12 ± 0.0
Fe	$\mu g\ kg^{-1}$	2.94 ± 0.0
Ni	$\mu g\ kg^{-1}$	0.14 ± 0.0
Mn	$\mu g\ kg^{-1}$	1.0 ± 0.0
Cu	$\mu g\ kg^{-1}$	0.05 ± 0.0
Ca	$mg\ kg^{-1}$	2.7891 ± 0.4350
Mg	$mg\ kg^{-1}$	0.0381 ± 0.0067

gases by the spontaneous carbonation reactions with CO_2 from atmosphere. The application of immediate one-step lime precipitation process during a year will allow the recovery of 66 ton of total P in Portugal. This study also allowed removal of biochemical oxygen demand and total phenols from the treated effluent and originated a nutritious precipitate, with the capacity for agricultural use.

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Authors' contributions All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by SL, JR, AA, and FC. The first draft of the manuscript was written by SL and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Data availability The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Compliance with ethical standards

Ethics approval and consent to participate Not applicable

Consent for publication All authors agreed with the content and that all gave explicit consent to submit.

Conflict of interest The authors declare that they have no conflict of interest.

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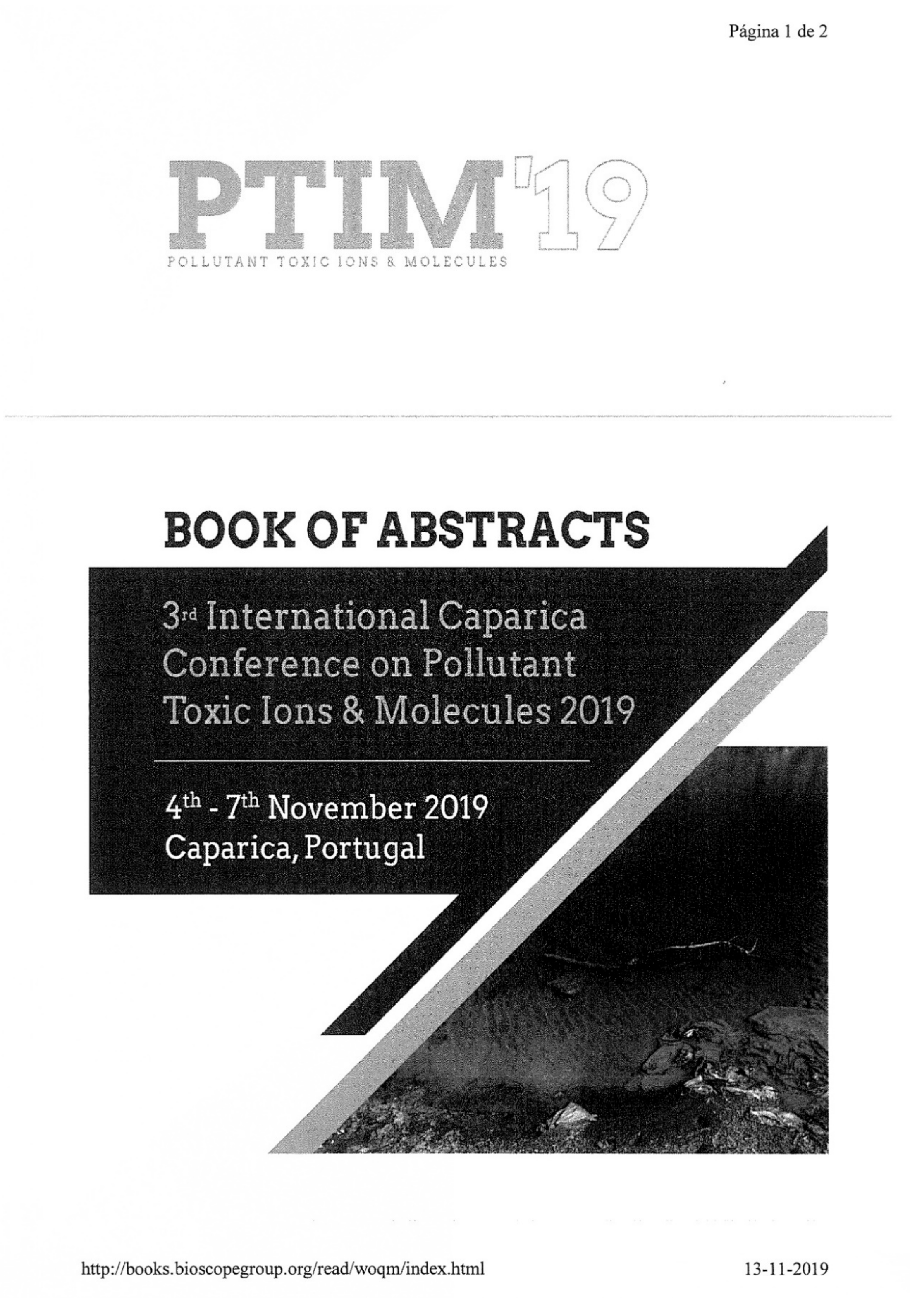
Comunicação 2 - Immediate one-step lime precipitation process for the valorization of winery wastewater to agricultural purposes



Página 1 de 2

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BOOK OF ABSTRACT

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Pollutant Toxic Ions & Molecules

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Immediate one-step lime precipitation process for the product of winery wastewater nutrient solution for agriculture purpos

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The winery industry generates, in all stages of the winemaking process, 1.0- 3.5 liters of winer
(WW) per liter of wine produced [1,2]. The high consumption of water is due to the washes ar
surfaces and equipment, refrigeration equipment, as well as the reception of grapes [3-5].
The WW were collected during the harvesting period of 2018 in the Ribafreixo winery, located
Alentejo, Portugal, which has 114 ha of vineyard with integrated production. The Ribafreixo WW
acid pH (4.26 ± 0.029); a high content of carbon oxygen demand (COD=3236 ± 30.43), turbidity (1
total phosphorus content (155.5 ± 6.36 mg L-1) and potassium (100.5 mg L-1). The metals: Pb,
Mn, Fe, Zn are present in concentration below the Portuguese irrigation rules (law decree 236/9
The immediate one-step lime precipitation process was developed by the use of one reactant (
solution at 200 gL-1), that produces immediately an abundant and insoluble precipitate able
organic matter and other contaminants from wastewater in a short time. Different precipitant c
of 1-50 mLL-1 are used to obtain the best reaction conditions (25 mL L-1 of concentered lime sol
reaction 12.24). These reactional conditions allow reaching removals of COD, turbidity and totalin
97.6% and 80.04 %, respectively. The nutritive solution was prepared using this treated Ribafre
underground wastewater in order to achieve the Electrical Conductivity (EC) according the needs
The obtained nutrient solution with EC 1.12 ± 0.053 dS m-1(law decree 236/98, annex XVI).is
(133.9 ± 12.0 mg Ca L-1), total phosphorus (11.08 ± 2.18 mg P L-1) and potassium (26.9 mgPL-1).

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Comunicação 3 - Tratamento de águas residuais de adega utilizando um precipitante básico

14º CONGRESSO DA ÁGUA

7, 8 e 9
Março 2018
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GESTÃO DOS
RECURSOS HÍDRICOS:
NOVOS
DESAFIOS

TRATAMENTO DE ÁGUAS RESIDUAIS DE ADEGA UTILIZANDO UM PRECIPITANTE BÁSICO

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Resumo

No mercado Português, a produção de vinho representa 88% da indústria de bebidas, e tem aumentado o volume de exportações nos últimos anos (Lourenço, 2017). O Alentejo foi a única região do país que aumentou o número de hectares de vinha plantada face ao ano de 1989 (Aguiar, 2016). Em 2013, o Alentejo detinha 22992 hectares de vinha, sendo o Alentejo Central, no ano de 2016, a terceira região com maior volume de produção de vinho do país (12,4%) (Aguiar, 2016).

A produção de água residual nas adegas é variável ao logo do ano, devido à sazonalidade dos processos inerentes à produção do vinho. Estes efluentes apresentam um pH ácido a neutro, elevado teor de matéria orgânica (Carência Química de Oxigénio (CQO) e Carência Bioquímica de Oxigénio (CBO_5)), Sólidos Totais (ST), Sólidos Suspensos Totais (SST) e Sólidos Voláteis (SV). Além disso, estes efluentes apresentam na sua composição fenóis totais e nutrientes como azoto e fósforo.

Como forma de minimizar os riscos ambientais, dar um novo destino à água residual e ir ao encontro das necessidades de uma indústria cada vez mais expressiva na região do Alentejo, aplicou-se um pré-tratamento utilizando um precipitante básico, doseando o reagente de forma a ajustar a água residual a pH 8, 9, 10, 11 e 12. A eficiência de remoção da turvação aumentou ligeiramente com o aumento do pH, mas a CQO teve o comportamento oposto. As remoções gerais foram de 65 - 81% de CQO, 86 - 89% de fósforo total, 76 - 82% de fenóis totais e 83 - 87% de turvação. Além disso, o processo permitiu remoções de absorvâncias características: compostos de baixo peso molecular (48 - 95%), compostos aromáticos e insaturados (60 - 69%), cor (47 - 72%) e densidade ótica celular (61 - 79%). O pH da água residual tratada variou entre 8,4 e 10,8.

Palavras-chave: Água Residual; Adega; Precipitante Básico; Matéria Orgânica; Fósforo.

Tema: Serviços de abastecimento, drenagem e tratamento de águas

1. INTRODUÇÃO

A indústria vitivinícola gera elevada quantidade de água residual, estimando-se uma produção de 1 a 3,5 litros de água residual por cada litro de vinho produzido (Montalvo et al., 2010; Domínguez et al., 2014). Esta produção deve-se principalmente às lavagens/higienizações de equipamentos e superfícies e à receção da uva, inerentes ao processo produtivo do vinho (Artiga et al., 2005; Agustina et al., 2008; Anastasiou et al., 2009; Braz et al., 2010). Na Figura 1 esquematiza-se o processo produtivo do vinho branco, onde são demonstradas algumas das etapas decorrentes do processo de vinificação, bem como o volume de efluente e a concentração de Carência Química de Oxigénio (CQO) gerados. O processo de vinificação exemplificado na Figura 1 confirma que esta é uma indústria que gera elevados volumes de efluente, podendo originar em cada fase/conjunto de fases 1,32 - 2,67 $L^{-1}_{\text{água}}\ L^{-1}_{\text{vinho}}$. O efluente gerado apresenta elevados valores de CQO, variando entre 2373 e 3891 $mg_{\text{CQO}}\ L^{-1}_{\text{vinho}}$.

Os efluentes vinícolas são maioritariamente compostos por água, açúcares, ácidos orgânicos e polifenóis, e em quantidades mais reduzidas, mas ainda assim presentes estão os detergentes e desinfetantes utilizados nos processos de limpeza e higienização (Pirra, 2005; Solís et al., 2017). Apesar de na sua maioria, os constituintes deste efluente serem naturais e resultarem da uva e do vinho, os mesmos são ambientalmente problemáticos, devido à elevada presença de matéria orgânica, ácidos orgânicos (ácido tartárico, málico, cítrico e láctico), ácidos fenólicos (ácidos benzoicos e cinâmicos) e microrganismos (especialmente leveduras e bactérias) (Pirra, 2005; Lucas e Peres, 2011; Chatzilazarou et al., 2012). Se descarregadas no ambiente sem sofrer qualquer processo de tratamento, as águas residuais provenientes da produção de vinho podem causar graves problemas ambientais, ao nível dos solos e das águas subterrâneas e superficiais, uma vez que os microrganismos que o efluente contém degradam a matéria orgânica, provocando modificações no oxigénio, pH, temperatura e turvação, e adicionando ainda produtos tóxicos (Coetzee et al., 2004; Pirra, 2005).

Os efluentes vinícolas têm sido tratados por precipitação química com agentes quelantes, sedimentação com adição de floculantes, coagulação/floculação e eletrocoagulação, obtendo-se remoções de CQO, turvação e Sólidos Suspensos Totais (SST), com valores entre 9 - 73%, 44 - 92,6% e 80 - 98%, respetivamente (Ioannou et al., 2015). Assim, neste trabalho estudou-se a aplicação de processo físico-químico por precipitação química básica para o tratamento da água residual de adega.

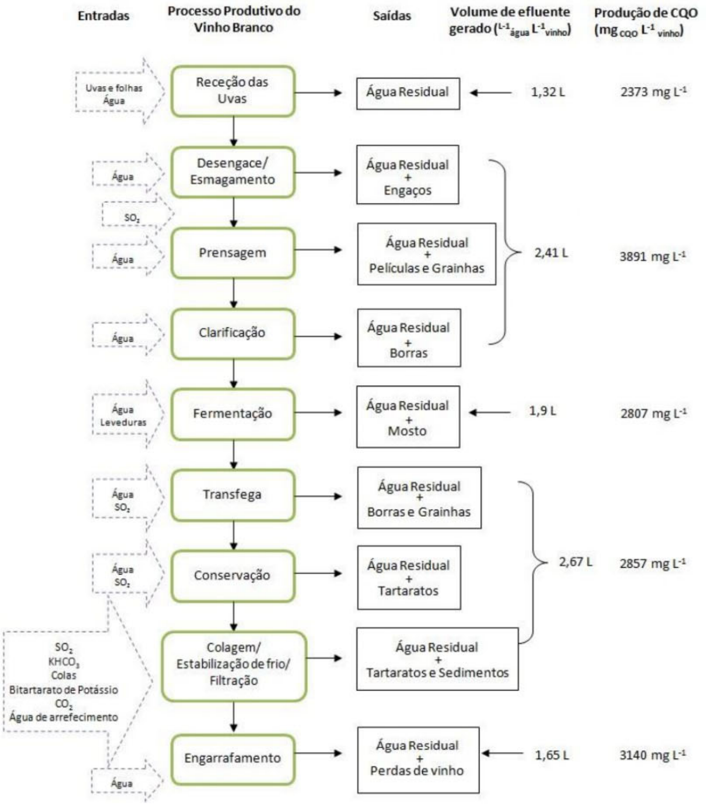

Figura 1. Descrição do processo produtivo do vinho branco

Fonte: Adaptado de Pirra, 2005; Rodrigues et al., 2006; Brito et al., 2007

2. MATERIAIS E MÉTODOS

2.1. Recolha do efluente

O efluente estudado foi recolhido no mês de Outubro de 2016 na Adega Ribafreixo Sociedade Agrícola, Lda., situada na Vidigueira, Distrito de Beja.

A recolha foi efetuada no tanque de retenção (Figura 2) da Estação de Tratamento de Águas Residuais (ETAR), antes de ser aplicado qualquer tipo de tratamento. As amostras foram armazenadas em recipientes de PVC com 5/6 litros de capacidade e mantidas a 4º C para a caracterização do efluente bruto que decorreu nos 2 a 4 dias seguintes à recolha, ou congeladas a -20 ºC para evitar alteração das propriedades.

Figura 2. Tanque de retenção da ETAR

2.2. Tratamento do efluente por precipitação básica

Neste estudo foi testado um precipitante básico (cal hidratada), através da aplicação de diversas dosagens de precipitante, de forma a aumentar o pH do efluente para 8, 9, 10, 11 e 12. As várias dosagens do precipitante foram aplicadas à temperatura ambiente e com agitação.

2.3. Parâmetros analisados e métodos

No efluente recolhido e tratado foram analisados os seguintes parâmetros: pH, condutividade, turvação, CQO, fósforo total, alcalinidade total e alcalinidade à fenolftaleína, dureza cálcica e dureza total, fenóis totais e absorvâncias características.

O pH foi medido através do método potenciométrico, utilizando um Potenciómetro Consort C 861 e um elétrodo SenTix 41. A condutividade elétrica foi medida através do método condutivimétrico num condutivímetro VWR CO 3100 H. A turvação foi analisada através do método turbidimétrico num turbidímetro WTW Turb550. O parâmetro CQO foi determinado por digestão em meio ácido, com solução de dicromato de potássio na presença de sulfato de mercúrio e sulfato de prata em placa de digestão Techne Dri-block DB 200/3 e a sua leitura foi feita num espectrofotómetro UV-visível Thermo Scientific Helios α ao comprimento de onda de 600 nm (APHA et al., 1998). O fósforo total foi digerido em meio ácido, com desenvolvimento de cor por colorimetria através do ácido vanadomolibdofosfórico e a leitura efetuada num espectrofotómetro UV-visível ao comprimento de onda de 470 nm (APHA et al., 1998).

As alcalinidades total e à fenolftaleína foram determinadas através do método volumétrico por titulação de neutralização com ácido clorídrico (APHA et al., 1998). As durezas total e cálcica foram determinadas através do método volumétrico de complexação utilizando EDTA e indicadores Negro de Eriocromo T e Calcon, respetivamente (APHA et al., 1998).

O teor em fenóis totais foi determinado segundo o método Folin-Ciocalteau, usando o ácido gálico como padrão (Falleh et al., 2008). As absorvâncias características: 220 nm (compostos de baixo peso molecular) (Kasprzyk-Hordern et al., 2006), 254 nm (compostos aromáticos e insaturados) (APHA et al., 1998), 410 nm (cor) (Rivas et al., 2005) e 600 nm (densidade ótica celular) (Megharaj et al., 2003) foram medidas com diluição apropriada num espectrofotómetro UV-visível Thermo Scientific Helios α.

3. RESULTADOS E DISCUSSÃO

3.1. Caracterização do efluente bruto

O efluente proveniente da adega apresentava uma coloração escura (vermelho escuro/castanho) com odor frutado forte. Na Tabela 1 apresentam-se as características físico-químicas do efluente bruto. O efluente bruto apresentou pH próximo da neutralidade (6,8) e baixa condutividade (1,62 dS m^{-1}), estando de acordo com os valores referidos por Ioannou et al. (2013a, 2013b) (pH 6 - 8,3) e Cusick et al. (2010) e Ioannou et al. (2013a) (condutividade elétrica de 1,3 - 3,3 dS m^{-1}). Além disso, o efluente apresentou elevados teores de matéria orgânica (CQO ≈ 2003 mg L^{-1}, Carência Bioquímica de Oxigénio (CBO_5) ≈ 1000 mg L^{-1}), de acordo com Lucas e Peres (2011) (CQO = 2000 – 20000 mg L^{-1}) e Petruccioli et al. (2000) (CBO_5 = 500 - 6900 mg L^{-1}). Também se verificou a presença de sólidos (Sólidos Totais (ST) ≈ 2041 mg L^{-1}, Sólidos Voláteis (SV) ≈ 1024 mg L^{-1} e SST ≈ 871 mg L^{-1}), tal como Lucas e Peres (2011), Ioannou et al. (2013a, 2013b) e Moreira et al. (2015), com os valores de SST, ST e SV de 500 - 1500 mg L^{-1}, 3672 - 5040 mg L^{-1} e 70 mg L^{-1}, respetivamente. A turvação foi de cerca 320 NTU, encontrando-se dentro dos valores descritos por Valderrama et al. (2012) (313 - 692 NTU) e o fósforo total foi de cerca de 210 mg L^{-1} o que está de acordo com o descrito por Montalvo et al. (2010) (250 mg L^{-1}).

Tabela 1. Caracterização do efluente bruto

Parâmetro	Unidade	Valor médio
pH	Escala de Sorensen	6,80 ± 0,057
Condutividade	dS m^{-1}	1,62 ± 0,064
Turvação	NTU	320,78 ± 145,063
CQO	mg L^{-1}	2003,3 ± 423,35
CBO_5	mg L^{-1}	1000,0 ± 122,47
Fósforo total	mg L^{-1}	210,1 ± 24,35
Cálcio	mg L^{-1}	110,8 ± 12,86
Magnésio	mg L^{-1}	64,9 ± 21,57
Alcalinidade total/Bicarbonatos	mg L^{-1} $CaCO_3$	815,6 ± 96,11
Dureza total	mg L^{-1} $CaCO_3$	543,5 ± 82,36
Dureza cálcica	mg L^{-1} $CaCO_3$	276,5 ± 32,08
Dureza magnesiana	mg L^{-1} $CaCO_3$	267,0 ± 88,74
Fenóis totais	mg L^{-1}	25,32 ± 3,007
Sólidos Totais	mg L^{-1}	2041,2 ± 129,05
Sólidos Voláteis	mg L^{-1}	1024,4 ± 95,64
Sólidos Suspensos Totais	mg L^{-1}	871,1 ± 169,78
Absorvâncias características		
220 nm	cm^{-1}	0,143 ± 0,0397 (1:50)[a]
254 nm	cm^{-1}	0,093 ± 0,0317 (1:50)[a]
410 nm	cm^{-1}	0,033 ± 0,0161 (1:50)[a]
600 nm	cm^{-1}	0,067 ± 0,0050 (1:50)[a]

[a] Fator de diluição utilizado para medir as absorvâncias características.

Os dados resultaram da realização de 5 réplicas de uma amostragem.

3.2. Tratamento do efluente por precipitação básica

Neste estudo, as águas residuais de adega foram tratadas por processos de precipitação básica a diferentes condições de pH. Os efeitos visuais foram notórios, a água clareou e apresentou menos turvação, como mostra a Figura 3.

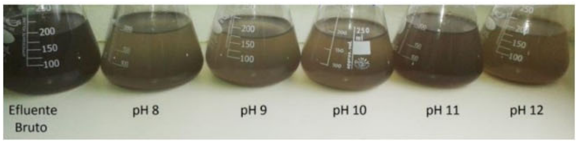

Figura 3. Efluentes Bruto e Tratados

A Figura 4 apresenta o efeito do tratamento sobre as absorvâncias características, especificamente para compostos de baixo peso molecular, compostos aromáticos e insaturados, cor e densidade ótica celular, medidas através dos comprimentos de onda 220, 254, 410 e 600 nm, respetivamente. As absorvâncias a 220, 410 e 600 nm apresentaram uma tendência de remoção semelhante. Estes parâmetros mantiveram-se constantes (com remoções superiores a 47%) entre pH 8 - 10, verificando-se aumento da eficiência de remoção para pH 11 e 12. A pH 12 foram obtidas as seguintes eficiência de remoção: 95,5% nos compostos de baixo peso molecular, 72,1% na cor e 79,2% na densidade ótica celular.

A absorvância a 254 nm referente aos compostos orgânicos de elevado peso molecular com alto grau de aromaticidade, com elevado número de duplas e triplas ligações, e grupos fenólicos não variou entre as condições operacionais estudadas (pH 8 - 12), com remoções compreendidas entre 60,1 - 69,1%. A precipitação básica tem sido geralmente utilizada para a eliminação da dureza da água, através da descarbonatação, mas também tem vindo a ser provada a sua eficiência para a remoção de moléculas orgânicas de alto peso molecular e ácidos húmicos e fúlvicos (Renou et al., 2008).

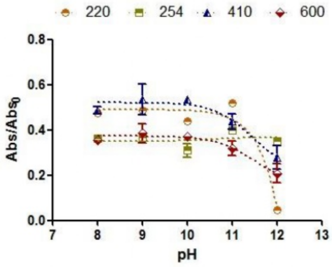

Figura 4. Efeito do tratamento nas absorvâncias características

O efeito do tratamento sobre o teor em fósforo total e fenóis totais apresenta-se na Figura 5. O fósforo total apresentou resultados semelhantes em todas as condições operacionais, com remoções entre 86,1 - 89,2%. As remoções de fenóis totais foram mais elevadas para pH 10 e 12, com valores em torno de 80%. A aplicação de processos de precipitação com cal apresenta como vantagens a não contribuição para o aumento da salinidade da água, não acarretar custos monetários muito elevados e geralmente apresenta remoções elevadas de fósforo total, SST, CQO e CBO_5 (Semerjian e Ayoub, 2003).

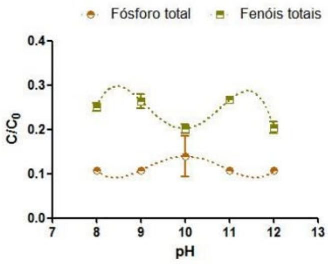

Figura 5. Efeito do tratamento no fósforo total e fenóis totais

A Figura 6 mostra o efeito do tratamento na turvação e CQO. A turvação apresentou remoções sempre superiores a 80%, verificando-se aumento da remoção para valores de pH ≥ 10. A remoção de CQO apresentou o comportamento oposto, com eficiências de remoção mais elevadas (79,2 - 80,9%) para pH mais baixos (8 - 10). O aumento do pH de precipitação diminuiu as eficiências de remoção de CQO. Boukhoubza et al. (2009) obteve 72% de remoção de CQO ao aplicar precipitação básica a pH 12 em água residual proveniente de lagares de azeite.

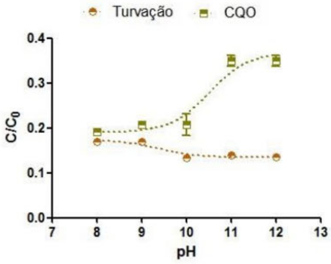

Figura 6. Efeito do tratamento na turvação e CQO

A aplicação de processos de precipitação básica com $Ca(OH)_2$ permite que ocorra na água a precipitação de fosfatos (equação 1 - 3), carbonato de cálcio (equação 4 - 7), carbonato e hidróxido de magnésio (equação 6 - 7), arrastando e precipitando as partículas coloidais em suspensão, através das seguintes reações (Semerjian e Ayoub, 2003; Renou et al., 2008; Prazeres, 2013; Prazeres et al., 2016):

$$3Ca(OH)_2 + 2PO_4^{3-} \leftrightarrow Ca_3(PO_4)_2 \downarrow + 6OH^- \quad (1)$$

$$4Ca(OH)_2 + 3PO_4^{3-} + H_2O \leftrightarrow Ca_4H(PO_4)_3 \downarrow + 9OH^- \quad (2)$$

$$10Ca^{2+} + 6PO_4^{3-} + 2OH^- \leftrightarrow Ca_{10}(PO_4)_6(OH)_2 \downarrow \quad (3)$$

$$Ca(OH)_2 + H_2CO_3 \leftrightarrow CaCO_3 \downarrow + 2H_2O \quad (4)$$

$$Ca(OH)_2 + Ca(HCO_3)_2 \leftrightarrow 2CaCO_3 \downarrow + 2H_2O \quad (5)$$

$$Ca(OH)_2 + Mg(HCO_3)_2 \leftrightarrow MgCO_3 \downarrow + CaCO_3 \downarrow + 2H_2O \quad (6)$$

$$Ca(OH)_2 + MgCO_3 \leftrightarrow Mg(OH)_2 \downarrow + CaCO_3 \downarrow \quad (7)$$

Desta forma dá-se a formação de precipitado (devido ao aumento dos iões Ca^{2+} e carbono inorgânico HCO_3^-) com redução de matéria orgânica, turvação, fósforo total, SST, condutividade, cálcio e magnésio (Renou et al., 2008; Prazeres, 2013).

4. CONCLUSÃO

A produção de efluentes na indústria vitivinícola é variável ao longo do ano (Fumi et al., 1995), devido à sazonalidade provocada pela colheita das uvas, tendo a vindima uma duração de aproximadamente 3 meses. Neste período, o volume de efluente gerado é superior ao produzido no resto do ano. O efluente produzido não poderá ser descarregado no ambiente sem sofrer um tratamento eficaz, devido à sua toxicidade.

A utilização de precipitação básica é uma opção de tratamento, devido ao seu custo reduzido e à eliminação de compostos prejudiciais ao ambiente. Apesar das elevadas remoções verificadas no processo de precipitação básica, os valores obtidos são superiores aos valores limite de emissão definidos no Decreto- Lei n.º 236/98 de 1 de Agosto (Anexo XVIII - Valores limite de emissão (VLE) na descarga de águas residuais), nomeadamente nos parâmetros CQO (383,3 mg L^{-1}) e fósforo total (22,8 mg L^{-1}). Como tal, este processo poderá funcionar como pré-tratamento, uma vez que permite uma eliminação elevada de matéria orgânica, turvação, fósforo total, fenóis totais e absorvâncias características.

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Comunicação 4 - Redução do nível de contaminação de águas residuais de queijarias por precipitação básica

Redução do nível de contaminação de águas residuais de queijarias por precipitação básica

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Resumo

As águas residuais de queijarias podem provocar graves problemas ambientais devido à sua composição físico-química, em termos de matéria orgânica com valores de cerca de 25000 mg L^{-1}, sólidos totais de aproximadamente 9000 mg L^{-1} e fósforo em torno de 3500 mg L^{-1}. Para além disso, é um efluente que apresenta um pH geralmente ácido, com valores entre 3 - 5. Assim, este trabalho teve como objetivo a aplicação de tratamento por precipitação básica às águas residuais provenientes de queijarias, de forma a reduzir o seu nível de contaminação. A água residual de queijaria bruta estudada apresentou um pH de 4,52, condutividade de 5,23 dS m^{-1}, carência química de oxigénio (CQO) de 4917 mg L^{-1}, fósforo de 3411,8 mg L^{-1}, e bicarbonatos de 1339,7 mg L^{-1} $CaCO_3$. Procedeu-se também ao estudo das absorvâncias características, utilizando um fator de diluição de 1:50, de compostos de baixo peso molecular (0,639 cm^{-1}), compostos aromáticos e insaturados (0,180 cm^{-1}), cor (0,092 cm^{-1}) e densidade ótica celular (0,060 cm^{-1}), obtidas nos comprimentos de onda espectrofotométricos a 220, 254, 410 e 600 nm, respetivamente. O tratamento consistiu na aplicação de diferentes dosagens de modo a obter um aumento de pH entre 6 e 13. A remoção máxima de CQO obteve-se a um pH de 11, com uma eficiência de aproximadamente 30%. Este processo mostrou-se eficiente na redução de fósforo, com valores de eliminação de 62 - 94%, para uma gama de pH de 8 - 13. Para as absorvâncias características obtiveram-se remoções máximas de 48, 71, 92 e 95%, respetivamente.

Palavras-chave: indústria de lacticínios, produção de queijo, desoxigenação, eutrofização, matéria orgânica

Reduction of the contamination level of cheese whey wastewater by basic precipitation

Abstract

The cheese wastewater can cause serious environmental problems due to its physicochemical composition in terms of organic matter with values of about 25000 mg L^{-1}, total solids of approximately 9000 mg L^{-1} and phosphorus around 3500 mg L^{-1}. In addition, it is an effluent that has generally an acidic pH, with values between 3 and 5. Thus, this work had the purpose of applying treatment by basic precipitation to wastewater from cheese industry, in order to reduce its level of contamination. The raw cheese wastewater studied had a pH of 4,52, conductivity of 5,23 dS m^{-1}, chemical oxygen demand (COD) of 4917 mg L^{-1}, phosphorus of 3411,8 mg L^{-1}, and bicarbonates of 1339,7 mg L^{-1} $CaCO_3$. The characteristic absorbances were also studied, using a 1:50 dilution factor, of low molecular weight compounds (0,639 cm^{-1}), aromatic and unsaturated compounds (0,180 cm^{-1}), color (0,092 cm^{-1}) and cellular optical density (0,060 cm^{-1}), obtained at the spectrophotometric wavelengths of 220, 254, 410 and 600 nm, respectively. The treatment consisted of the application of different dosages in order to obtain a pH increase between 6 and 13. The COD maximum removal was obtained at a pH of 11, with an efficiency of approximately 30%. This process proved to be efficient in reducing phosphorus, with elimination values of 62 - 94%, for a pH range of 8 - 13. For the characteristic absorbances, maximum removals of 48, 71, 92 and 95%, respectively, were obtained.

Keywords: Dairy industry, cheese production, deoxygenation, eutrophication, organic matter

1. Introdução

A produção de queijo exige elevado consumo de água, estimando-se um volume de cerca de 15 litros de água por cada litro de leite utilizado no processo de fabrico (Prazeres et al., 2018). Consequentemente, o volume de água residual produzido constitui uma questão ambiental preocupante, devido ao elevado teor de matéria orgânica, gorduras e sólidos suspensos, salinidade e pH ácido.

Este efluente é gerado durante os vários processos de produção do queijo, designadamente, na coagulação, moldagem/ prensagem, desmoldagem, embalamento e lavagem de pavimentos e equipamentos (INETI, 2001). O efluente apresenta elevado teor de óleos e gorduras, provenientes das perdas de queijo e soro, levando à formação de camadas espessas na superfície das lagoas de armazenamento. Este efeito causa condições de anaerobiose com conversão de lactose em ácido lático, provocando a libertação de fortes odores (Prazeres et al., 2016a). Essas condições são propícias ao aparecimento de vetores transmissores de doenças (Prazeres et al., 2011).

O soro do leite é um dos componentes mais problemáticos desta indústria, uma vez que é o subproduto que apresenta uma maior contaminação (Rajeshwari et al., 2000; Prazeres et al., 2012). Estima-se em média que, por cada 1 kg de queijo produzido, são necessários 10 kg de leite e gerados 9 kg de soro (Prazeres et al., 2012).

Antigamente, era prática comum os produtores de queijo fazerem a descarga dos efluentes produzidos no processo de fabrico para os terrenos ou diretamente para meios hídricos recetores, como rios, lagos, ribeiras, oceanos, etc., sem a aplicação de qualquer pré-tratamento (Prazeres et al., 2012). Contudo, o efluente proveniente da queijaria, quando descarregado no ambiente sem controlo prévio, pode provocar diversos problemas ambientais e de saúde pública (Prazeres, 2013; Prazeres et al., 2016b). Estes efluentes podem conduzir à devastação da fauna devido às gorduras suspensas, uma vez que impedem a entrada de oxigénio (condições anaeróbias). Além disso, a eutrofização dos meios recetores pode ocorrer devido à presença de fósforo e azoto (Prazeres, 2013).

Hoje em dia, a realidade é menos nociva, mas ainda longe de ser a ideal. A maioria das queijarias faz a descarga dos seus efluentes para lagoas, tanques de armazenamento, fossas ou no coletor municipal (quando cumprem com os requisitos de descarga). Para o efluente de queijaria têm vindo a ser estudados tratamentos biológicos, como a digestão anaeróbia e digestão aeróbia e processos físico-químicos, como a coagulação/floculação, precipitação e oxidação (Carvalho et al., 2013).

Com este trabalho, pretende-se estudar um processo físico-químico de precipitação básica para redução da contaminação orgânica e dos nutrientes do efluente de queijaria.

2. Material e Métodos

2.1. Água residual bruta de queijaria

A água residual, designada de efluente bruto, foi recolhida na lagoa de armazenamento de efluentes de uma queijaria instalada na região do Baixo Alentejo, que utiliza nas suas produções leite de ovelha e cabra.

O efluente apresenta uma coloração amarelada (Figura 1) devido à presença de vitamina B2, também designada de riboflavina, proveniente do soro do leite (De Wit, 2001; Prazeres et al., 2012). A existência de grandes quantidades de gordura suspensa e forte odor constituem outras propriedades deste tipo de efluente. Este é principalmente constituído por: soro e sorelho resultantes de perdas na produção, águas de lavagem com a presença de detergentes e desinfetantes que advêm da limpeza e desinfeção de equipamentos, instalações, tanques e utensílios, lubrificantes utilizados nas operações de manutenção, sólidos de leite e sal (Mendes e Mendonça, 2013; INETI, 2001).

O efluente proveniente de queijaria estudado apresenta as seguintes características (Tabela 1): um elevado teor de matéria orgânica (CQO ≈ 5000 mg L^{-1}) e nutrientes (fósforo ≈ 3400 mg L^{-1}). O pH é ácido (≈ 4,5) e a condutividade elevada (≈ 5,23 dS m^{-1}). As lagoas de armazenamento apresentam densas camadas de óleos e gorduras que estão presentes no efluente (Figura 2), contendo elevado teor de sólidos totais (≈ 9000 mg L^{-1}), principalmente na forma de sólidos dissolvidos totais (Prazeres et al., 2016a).

Figura 1. Efluente Bruto

Tabela1. Caracterização físico-química do efluente bruto proveniente de queijaria

Parâmetro	Unidade	Valor médio
pH	escala de Sorensen	4,52 ± 0,205
Condutividade	dS m^{-1}	5,23 ± 0,277
Turvação	NTU	594,9 ± 49,54
CQO	mg L^{-1}	4916,7 ± 353,55
Fósforo	mg L^{-1}	3411,8 ± 1255,64
Cálcio	mg L^{-1}	236,2 ± 28,94
Magnésio	mg L^{-1}	109,5 ± 36,77
Alcalinidade total	mg L^{-1} $CaCO_3$	1339,7 ± 251,14
Bicarbonatos	mg L^{-1} $CaCO_3$	1339,7 ± 251,14
Dureza total	mg L^{-1} $CaCO_3$	1039,7 ± 168,69
Dureza cálcica	mg L^{-1} $CaCO_3$	589,3 ± 72,20
Dureza magnesiana	mg L^{-1} $CaCO_3$	450,4 ± 151,30
Fenóis totais	mg L^{-1}	75,9 ± 6,76
	Absorvâncias características	
220 nm	cm^{-1}	0,639 ± 0,0383 (1:50)[a]
254 nm	cm^{-1}	0,180 ± 0,0147 (1:50)[a]
410 nm	cm^{-1}	0,092 ± 0,0065 (1:50)[a]
600 nm	cm^{-1}	0,060 ± 0,0072 (1:50)[a]

[a] Fator de diluição utilizado para medir as absorvâncias características.

Os dados resultaram da realização de 5 réplicas de uma amostragem.

Figura 2. Lagoa de armazenamento do efluente proveniente de queijaria

2.2. Tratamento por adição de precipitante básico

A adição de precipitantes básicos pode gerar espécies insolúveis, que retêm e arrastam partículas coloidais em suspensão, o que permite a eficaz remoção de matéria orgânica e turvação (Prazeres, 2013). Assim, neste trabalho procedeu-se à aplicação de diferentes dosagens de precipitante ao efluente bruto, de modo a obter um aumento de pH entre 6 e 13 (Figura 3). As oito condições de pH foram depois analisadas nos seguintes parâmetros: pH, condutividade, turvação, CQO, fósforo, alcalinidade à fenolftaleína e ao alaranjado de metilo, dureza total, cálcica e magnesiana, absorvâncias características (220 nm - compostos de baixo peso molecular, 254 nm - compostos aromáticos e insaturados, 410 nm - cor e 600 nm - densidade ótica celular), e fenóis totais.

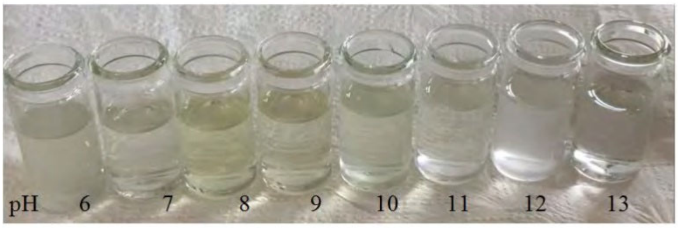

Figura 3. Efluente tratado por precipitação básica nas diversas condições operacionais

2.3. Métodos

Os equipamentos e métodos utilizados para a caracterização do efluente bruto e tratado apresentam-se na seguinte tabela.

Tabela 2. Equipamentos e métodos utilizados para a caracterização do efluente bruto e tratado

Parâmetro	Equipamento	Método	Referência
pH	Potenciómetro Consort C 861	Potenciométrico	---
Condutividade	Condutivímetro VWR CO 3100 H	Condutivimétrico	---
Turvação	Turbidímetro WTW Turb550	Turbidimétrico	---
CQO	Placa de digestão Techne Dri-block DB 200/3 Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	APHA et al., 1998
Fósforo	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	APHA et al., 1998
Alcalinidade total	---	Volumétrico	APHA et al., 1998
Dureza total	---	Volumétrico (Indicador – Negro de eriocromo T)	APHA et al., 1998
Dureza cálcica	---	Volumétrico (Indicador – Calcon)	APHA et al., 1998
Absorvâncias características			
220 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	Kasprzyk-Hordern et al., 2006
254 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	APHA et al., 1998
410 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	Rivas et al., 2005
600 nm	Espectrofotómetro Thermo Scientific Helios α	Espectrofotométrico	Megharaj et al., 2003
Fenóis totais	Espectrofotómetro Thermo Scientific Multiskan FC	Espectrofotométrico	Falleh et al., 2008

3. Resultados e Discussão

3.1. Estudo das eficiências de remoção e condições ótimas no processo de precipitação básica

A adição do precipitante básico ao efluente bruto foi realizada variando as condições de operação em relação ao parâmetro pH (6 – 13). A adição do precipitante permitiu a formação de precipitado em todas as condições de operação estudadas, obtendo-se um volume de 20 a 120 mL de precipitado por litro de efluente bruto.

Os resultados obtidos no processo estudado encontram-se na Figura 4, em relação aos seguintes parâmetros: pH e condutividade (a); cálcio e magnésio (b); hidróxidos, carbonatos e bicarbonatos (c); absorvâncias características a 220, 254, 410 e 600 nm (d); fósforo (e); CQO e turvação (f).

No efluente tratado com condições de pH mais elevado (pH 13), observou-se um aumento significativo no pH, condutividade e concentração de cálcio do efluente tratado, face as restantes concentrações. A utilização de pH elevado (pH 13) alterou também a predominância das espécies alcalinas, verificando-se a presença em grandes quantidades de hidróxidos e carbonatos, contrariamente às restantes condições (pH 6 – 12), em que predominam bicarbonatos. Verificou-se também nas condições mais extremas (pH 12 – 13), elevadas remoções de compostos de baixo peso molecular, compostos aromáticos e insaturados, cor e densidade ótica celular, obtidas nos comprimentos de onda espectrofotométricos a 220, 254, 410 e 600 nm.

No geral, um aumento do pH de precipitação conduziu a um aumento da eficiência de remoção de fósforo e turvação, obtendo-se as eficiências máximas de 96% a pH 12 e 99% a pH 13, respetivamente. Em termos de matéria orgânica (CQO) obteve-se uma redução máxima de 30% ao pH 11. Os resultados obtidos estão de acordo com trabalhos anteriores do grupo, em que a aplicação de um precipitante básico sódico permitiu obter elevadas eficiências de remoção de CQO (40%), turvação (91%), fósforo (53%) e cálcio (50%) (Prazeres et al., 2016a). Resultados análogos foram também reportados por Rivas et al., 2011 e Prazeres et al., 2011.

3.2. Caracterização do efluente obtido nas condições ótimas no processo de precipitação básica

Após o estudo das condições operacionais do processo de precipitação básica, selecionou-se o pH 12 como a condição ótima para a remoção dos parâmetros avaliados. Na Tabela 3 apresenta-se a caracterização físico-química do efluente obtido no processo de precipitação nas condições ótimas de operação. O efluente obtido apresenta pH alcalino (≈ 9,2) e elevado valor de condutividade (≈ 5,20 dS m^{-1}).

Apesar de manter um elevado teor de matéria orgânica (CQO ≈ 4200 mg L^{-1}), obteve-se reduzidas concentrações de fósforo (≈ 150 mg L^{-1}), turvação (≈ 39 NTU), fenóis totais (≈ 29 mg L^{-1}), e compostos de baixo peso molecular (≈ 0,389 cm^{-1}), compostos aromáticos e insaturados (≈ 0,061 cm^{-1}), cor (≈ 0,008 cm^{-1}) e densidade ótica celular (≈ 0,003 cm^{-1}), obtidas nos comprimentos de onda espectrofotométricos a 220, 254, 410 e 600 nm.

A alcalinidade e dureza do efluente tratado apresenta-se principalmente sob a forma de bicarbonatos e cálcio, respetivamente, com uma concentração aproximada de 1659 mg L^{-1} $CaCO_3$ e 534 mg L^{-1}, respetivamente.

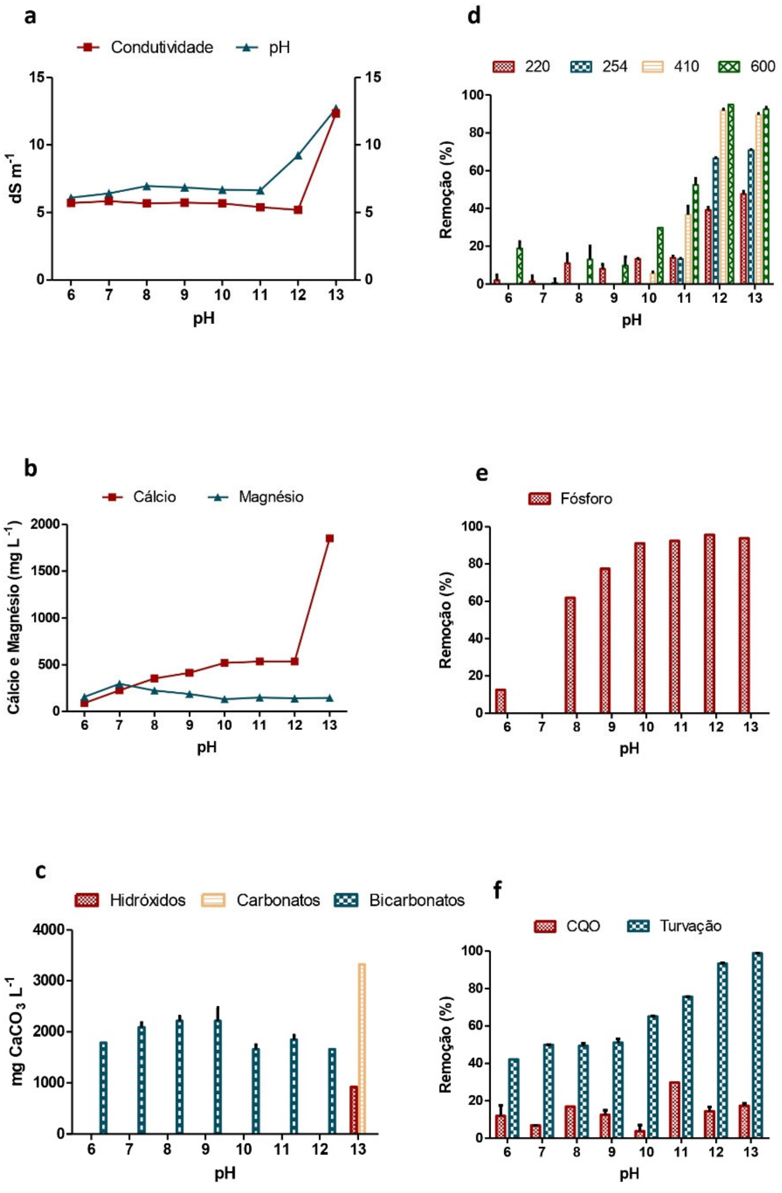

Figura 4. Resultados obtidos no processo de precipitação básica aplicado a águas residuais provenientes de queijaria

Tabela 3. Caracterização físico-química do efluente tratado por precipitação básica nas condições de operação ótimas

Parâmetro	Unidade	Valor médio
pH	escala de Sorensen	9,24 ± 0,001
Condutividade	dS m^{-1}	5,20 ± 0,035
Turvação	NTU	39,23 ± 2,905
CQO	mg L^{-1}	4208,3 ± 117,85
Fósforo	mg L^{-1}	148,7 ± 61,51
Cálcio	mg L^{-1}	534,0 ± 10,64
Magnésio	mg L^{-1}	141,4 ± 12,90
Alcalinidade total	mg L^{-1} $CaCO_3$	1659,3 ± 0,00
Bicarbonatos	mg L^{-1} $CaCO_3$	1659,3 ± 0,00
Dureza total	mg L^{-1} $CaCO_3$	1914,2 ± 76,62
Dureza cálcica	mg L^{-1} $CaCO_3$	1313,5 ± 26,54
Dureza magnesiana	mg L^{-1} $CaCO_3$	581,8 ± 53,08
Fenóis totais	mg L^{-1}	28,97 ± 1,429
	Absorvâncias características	
220 nm	cm^{-1}	0,389 ± 0,0092 (1:50)[a]
254 nm	cm^{-1}	0,061 ± 0,0007 (1:50)[a]
410 nm	cm^{-1}	0,008 ± 0,0007 (1:50)[a]
600 nm	cm^{-1}	0,003 ± 0,000 (1:50)[a]

[a] Fator de diluição utilizado para medir as absorvâncias características.

4. Conclusões

O efluente bruto proveniente de queijaria constitui um severo problema ambiental e de saúde pública, devido ao seu pH ácido e elevado teor de matéria orgânica, óleos e gorduras, sólidos suspensos, fósforo e sais.

Atualmente, a maioria das queijarias está a armazenar os seus efluentes em lagoas, tanques de armazenamento e fossas sépticas ou a descarregar no coletor municipal, o que ainda está longe se ser a solução ideal para este efluente tão problemático. Como forma de colmatar as lacunas existentes nesta área, procedeu-se ao estudo de um processo de tratamento, de forma a minimizar ou até eliminar os riscos que este apresenta no meio ambiente.

O processo de precipitação básica utilizado neste estudo não permite que o mesmo seja descarregado em meio hídrico ou no coletor municipal. No entanto, verificou-se remoções significativas em alguns parâmetros, designadamente, absorvâncias características, fósforo, fenóis totais e turvação. O efluente tratado necessita de um pós-tratamento de forma a obter valores permitidos por lei para descarga.

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Artigo 5 - Ecotoxicological risk assessment as a decision tool for the valorisation of winery wastewater treatment byproducts to agricultural purposes

ECOTOXICOLOGICAL RISK ASSESSMENT AS A DECISION TOOL FOR THE VALORIZATION OF WINERY WASTEWATER TREATMENT BYPRODUCTS TO AGRICULTURAL PURPOSES

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ABSTRACT

A simple and economical process consisting in the precipitation with slaked lime ($Ca(OH)_2$) was applied to winery wastewater. The process not only removed organic matter and other contaminants but also simultaneously captured atmospheric CO_2. In order to help the wine industry to advance to the objective of circular economy, treatment byproducts (supernatant and sludge) have been reused for agricultural purposes due to their physicochemical properties compatible with these applications. In addition to an exhaustive study of the physicochemical characteristics, the ecotoxicological impact of these by-products were also evaluated through the bioindicators: *Thmanocephalus platyurus* (mortality bioassay after 24h), *Vibrio fisheri* (luminescence inhibition after 30 min) and *Pseudokirchneriella subcapitata* (growth inhibition after 72h).

Raw winery wastewater showed high toxicity to all bioindicators, being classified as class 4 (Toxic Unit (TU); classification proposed by Personne, 1999). According values of TU of 20% for *T. platyurus* (24h), 9.17% for *V. fisheri* (30 min) and 100% for *P. subcapitata* (72h) were found. After treatment (treated WW), the toxicity significantly dropped to class 2, with values of TU of 1.34% for *T. platyurus* (24h), 2.92% for *V. fisheri* (30 min) and 3.97% for *P. subcapitata* (72h).

After the immediate one-step lime precipitation process (treated WW), the supernatant was diluted 1:4 (v/v) with groundwater (GW) to generate the so-called effluent nutrient solution (WWNS) which did not present any type of toxicity to the bioindicators used.

Accordingly, an assay was carried out to the production of red pak choi, using three different conditions: irrigation with groundwater (control); pH soil correction with organomineral fertilizer and groundwater irrigation; and irrigation with wastewater nutrient solution (WWNS). After the assay none of the soils showed any type of indirect ecotoxicity.

The pigment of the red pak choi cultivar produced in this study was evaluated through the analysis of chlorophyll a, chlorophyll b and carotenoids, showing the benefits to the healthy development of the plant.

Keywords: Wine industry; ecotoxicological assays; wastewater industry; slaked lime.

1. INTRODUCTION

The Mediterranean area is one of the most important regions in wine production worldwide (Amor et al. 2019; Oliveira et al. 2019), with the European countries representing 61% of total world production (Bolzonella et al. 2019). Portugal is the 11th wine producer in the world, with 7.4 million hL in 2021 (IVV IP, 2022), being Alentejo (one of the 14 wine regions, located in the South of Portugal) the 3rd wine producing region of the country, with almost 1.2 x 10^6 hL of wine produced in 2021/2022 (IVV IP, 2022). In the Alentejo region, the vineyards crops are irrigated, due to the meteorological conditions of the area with low precipitation and high temperatures (Costa et al. 2020).

Because of climate change and water scarcity, growth of the wine industry can be questioned (Bolzonella et al. 2019). Reuse of resources, namely water, is of paramount importance to the sustainability of this activity. During the winemaking process (from the reception of the grape to bottling), many resources are used, mainly energy and water (cleaning tanks and barrels, washing equipment and floors) (Amor et al. 2019). In the case of water, consumption can raise up to 450 L of water per liter of wine produced (Costa et al. 2020). In addition to the water used in the process, effluents generated are of major concern for the sector, with high variation in composition and flow generated throughout the natural year (Ngwenya et al. 2022). Additionally, the effluent generated contains a wide diversity of substances such as grape films and seeds, yeasts, alcohols (methanol, ethanol and glycerol), soluble acids (acetic, tartaric), tannins, lignin's, polyphenols, residues and cleaning and disinfection products (Ngwenya et al. 2022; Beer et al. 2017; Solís et al. 2017). In general, winery effluents present high values of organic matter measured as chemical oxygen demand (COD) 31.37 - 38.39 g L^{-1}, total suspended solids (TSS) 34.90 - 76.60 g L^{-1}, as well as high turbidity 319-782 NTU (Ioannou et al. 2015). Due to the seasonal character of this crop, the pH of the effluent varies throughout the year. Additionally, this effluent is characterized by low levels of nutrients such as nitrogen and phosphorus (Bolzonella et al. 2019) and high levels of sodium and potassium (Arienzo et al. 2009). Winery effluents are frequently discharged into the environment or into the sewer system without an adequate pre-treatment stage (Calheiros et al. 2018), presenting a strong environmental impact (Amor et al. 2019). If properly treated, winery wastewater can be reused for agricultural irrigation (Mosse et al. 2012). Winery wastewater treatment studies have already been presented in the specialized literature, such as, coagulation-flocculation (Braz et al. 2010; Rizzo et al. 2010); coagulation-flocculation-decantation and ozonization process (Jorge et al. 2021a) membrane processes (nanofiltration and reverse osmosis) (Ferrarini et al. 2001; Ioannou et al. 2013a); ozonation (Beltrán et al. 1999; Lucas et al. 2009a, 2009b); combination of ozone and radiation and/or hydrogen peroxide (Lucas et al. 2009 a); photo-Fenton (Mosteo et al. 2006; Ormad et al. 2006); photo-Fenton with solar radiation (Monteagudo et al. 2012); wet oxidation (Domínguez et al. 2014); anodic oxidation (Lauzurique et al. 2022; Candia-Onfray et al. 2018), anaerobic digestion (Kaira et al. 2022), combination of adsorption and thermocatalytic processes (Jorge et al. 2021b) advanced oxidation processes based on sulfate radicals (Amor et al. 2019), photocatalytic/ photolytic processes (Agustina et al. 2008) and microalgae and immobilized TiO_2/UV-A LEDs (Marchão et al. 2021). However, the application of alkaline precipitants for the treatment of wastewater in wineries still needs to be further studied. Luz et al. 2021, developed a simple and cheap treatment process for winery wastewater which consists of a single precipitation step with slaked lime ($Ca(OH)_2$) to remove organic matter and other contaminants with simultaneous atmospheric CO_2 capture. That application results in an abundant and insoluble precipitate, capable of sweeping and removing contaminants and the instantaneous combination of calcium and magnesium salts, through the addition of an excess of calcium ions (Luz et al. 2021). In addition to the process studied and to the analysis of chemical parameters, an integral evaluation of the treatment efficiency in terms of reuse or disposal is recommended. This process could help the winery industry to achieve circular economy through the reuse of generated residues (supernatant and sludge).

Application of byproducts for agricultural purposes requires a previous ecotoxicological analysis. The assessment of ecotoxicological endpoints allow a preliminary analysis of the environmental impact of these wastewaters (Mendonça et al. 2009). In this line, Sousa et al. (2019) studied the impact of vinasses wastewater obtained from the distillation of winemaking by-products on several species, such as *Aliivibrio fisheri*, *Daphnia magna* and *Zebrafish sp*. The acute results revealed a high toxicity for all species tested, indicating a strong impact in aquatic ecosystem (see Table 1).

Arienzo et al. (2009) used Garden cress (*Lepidium sativum*) and Onion (*Allium coepa*) in phytotoxicity bioassays to determine the quality of the winery wastewater (see Table 1). The authors claimed again a high toxicity, inhibiting the development and growth of the species under study correlated to high levels of COD, phenols and salinity, and low pH.

Ioannou et al. (2013b) carried out the treatment of winery wastewater by a membrane bioreactor (MBR) and Fenton's solar oxidation processes. The complete process generated an effluent without toxicity to the aquatic crustacean *D. magna*, as well as without phytotoxicity (Table 1).

In summary, the main objectives of this study were: (i) to assess the environmental impact of wastewater from wineries; (ii) analyze the potential reuse of the supernatant and precipitate in food production.

Table 1. Ecotoxicological effects of raw and treated winery wastewater

TYPE OF WASTEWATER	**BIOINDICATORS**	**BIOASSAY**	**ECOTOXICOLOGICAL ENDPOINTS (EC_{50})**	**STUDY**
Raw winery	*D. magna*	Mortality/ immobility	93.3±2% (24h)	Ioannou et al. 2013b
Winery MBR + Fenton solar oxidation process	*D. magna*	Mortality/ immobility	26.7±1.5% (24h)	
Raw vinasse	*D. magna*	Mortality/ immobility	4.8% (48 h)	Sousa et al. 2019
	Zebrafish embryos	Mortality	0.34% (96 h)	
	A. fisheri	Luminescence inhibition	7.0% (30 min)	
Raw winery	*Lepidium sativum*	Germination	2.5-15%	Arienzo et al. 2009
	Allium coepa	Germination	0.25-2%	

EC_{50} - concentration with effect in 50% of the exposed population.

2. MATERIALS AND METHODS

This article is a complement of the article of Luz et al. (2021), where a study of the best efficient processes of the immediate lime precipitation and carbonatation with atmospheric CO_2 process was reported. This work adds the description of the preparation of the nutritive solution based on the wastewater from the treated wine cellar and all the ecotoxicological characterization of the different waters, to be applied in agricultural, with correction soil properties and growth of the Red Pak Choi (*Brassica rapa L. var. chinensis (L.) Kitam*), through irrigation with diluted supernatant and incorporation of sludge into the soil.

2.1. Winery Wastewater

Winery wastewater was collected from a winery located at Vidigueira (Beja; Portugal), that has an integrated production regime, consisting of more than 100 ha of vineyards in sustainable planning, with about 10 grape varieties (Antão Vaz, Síria, Verdelho, Alvarinho, Arinto, Chenin Blanc, Aragonez, Touriga Nacional, Alicante Bouschet and Tinta Miuda).

The sampling was carried out in a single day in September 2017, at the winery effluent treatment station in the wastewater (WW) storage tank. Wastewater samples were collected in 5 L polyethylene (PET) packages, subjected to refrigeration at 4ºC or freezing at -20ºC (the specific procedure was detailed previously in Luz et al. 2021), until analysis.

2.2. Soil Characterization

Soil samples were collected from a vineyard, whose main characteristic is its acidic nature. After collection, soil samples were air-dried and sieved through a 2 mm sieve. Physicochemical characterization was carried out in triplicate. Extractable P and K were determined using the Egnere-Riehm Method (Riehm, 1958).

After air-drying, soil samples were subjected to a water leaching test at room temperature for 24 h, under constant stirring, using deionized water to form a solid-liquid solution 1:5 (w/v) ratio. The leachate was separated by centrifugation and filtered through a membrane filter of 0.45 µm pore size. Leachate was analyzed for pH, Electrical Condutivity (EC; dS m^{-1}) and ecotoxicologycal bioassays.

2.3. Metals analysis

Metal ions Pb, Cd, Zn, Fe, Ni, Mn, Cu, Ca and Mg were determined in the wastewater samples and leached from the soil, by atomic absorption spectrometry (AAS) after digestion of the samples with aqua regia, according to ISO 11466 (1995). Samples were digested with a mixture of HCl (37%) and HNO_3 (70%), in a ratio 3:1 (v/v), at room temperature for 16 h, and then at 130 ° C for 2 h, under reflux conditions.

Each suspension was then filtered and diluted to 100 mL with 0.5 mol L^{-1} HNO_3. Three independent replicates were analyzed for each sample and blanks were measured in parallel.

2.4. Ecotoxicological characterization

A battery of bioassays was performed in the following samples: Raw Wastewater (WW); Treated WW (TWW); Groundwater (GW); WW nutrient solution (WWNS) (see Table 2) assessing lethal and sub-lethal endpoints, with representative species of key taxonomic and functional groups: i) lethal effects assessed in crustaceans; and ii) two sublethal effects, light and growth inhibition assessed in bacterial and algae.

Raw WW is water directly collected from the winery's treatment plant; treated WW is the water treated through the immediate one-step lime precipitation process ((optimal dosage of 5 g L^{-1} of slaked lime (Luz et al. 2021)), and GW is the water that will be further used to dilute the TWW; WWNS is the combination TWW and GW (dilution 1:4), in order to obtain the optimum pH and EC for plant growth.

All bioassays considered have been largely used in ecotoxicological studies and, most of them, have already been standardized. The bioassays were: i) 30 min of luminescence inhibition of *V. fischeri* (ISO 11348-2, 1998); ii) 24-h mortality test with *T. platyurus* (Persoone, 1999); iii) 72-h population growth of the green microalgae *P. subcapitata* (OECD 201, 2006). All reported ecotoxicological tests fulfilled the validity requirements established by the respective guidelines.

2.4.1. *V. fischeri* bioassay

The Luminotox® bioassay was completed by using a marine bacterium *Vibrio fischeri* (NRRL B-11177). Luminescence inhibition was monitored with bacteria supplied as a dry liquid solution (Dr. Lange GmbH & Co. KG, Düsseldorf, Germany, ISO 11348-2 standard (1998) and protocol “DR LANGE luminescent bacteria test”). The solution was stored at -20 °C and rehydrated before tests. These tests were carried out with two replicates of 100% (v/v) of the samples and successive dilutions of 2% NaCl at 50, 25, 12.5, 6.25 and 3.125% (v/v) and non-toxic control of 2% NaCl solution at a fixed temperature of 15±0.5°C. In each sample, bioluminescence was measured before and after the desired incubation period (30 min.) and EC_{50} (%; the concentration of each sample that reduced 50% of bacterial luminescence) was determined. The decrease in luminescence is due to the inhibition of the bacterial luciferase enzyme by exposure to the samples.

2.4.2. *T. platyurus bioassay*

The Thamnocephalus platyurus bioassay was carried out following the protocol used in the THAMNOTOXKIT F^{TM} kit (Persoone, 1999). This test evaluates the mortality of shrimp larvae (<24h) incubated in 24-well plates, with 1.0 mL of the test solution and ten crustaceans per well. Four repetitions per treatment were competed at 25 °C for 24 hours in the absence of light, without feeding the animals during the time of the test. Concentrations of 12.5, 25, 50, 100% (v/v) were tested using the samples of supernatant. And concentrations of 25, 50, 100% (v/v) were tested using the samples (included in the test kit, also used as a non-toxic control) for the soils. The 24 hour contact with each test solution was used as the selected end point, and the EC_{50} (%) was calculated.

2.4.3. Growth inhibition of green microalgae *P. subcapitata*

The determination of the fluorospectrometric growth rate of the unicellular microalgae *P. subcapitata* was carried out for 72 hours, (protocol of OECD 201 (2006)), with the MBL Medium Woods-Holecontrol medium. At the beginning of the test, *P. subcapitata* (100 µL of an inoculum with $3\text{-}5\times10^4$ cells mL^{-1}) was exposed to water samples (900 µL) in a 24-well microplate. The test flasks were randomly incubated in an orbital shaker for 72 hours at a constant temperature of 21±2°C and with a constant light intensity (60–120 µE m^{-2} s^{-1}, equivalent to 6.000–10.000 lx). Concentrations were calculated using the microscope and Neubauer chamber.

The average rate of specific growth for a specific period was calculated as the logarithmic increase in biomass (after 72 hours) using the equation:

$$\mu_{i-j} = (\ln B_j - \ln B_i)/t_j - t_i \qquad (1)$$

Where: μ_{i-j} is the average specific growth rate from time i to j; t_i is the time for the start of the exposure period; t_j is the time for the end of the exposure period, B_i is the biomass concentration at time i, and B_j is the biomass concentration at time j.

The inhibition of algae growth was estimated as the percentage of reduction in the growth rate compared to the control:

$$\% I= [(\mu_c- \mu_t)/ \mu_c] \times 100 \qquad (2)$$

Where: % I is the mean percentage of inhibition for specific growth rate; μ_c is the mean value for the growth rate in the control, and μ_t is the mean value for the growth rate in water samples.

2.5. Red Pak Choi cultivation. Experimental design

Red Pak Choi was grown in pots with 1.5 kg of acidic soil under three different conditions: i) irrigated with groundwater (soil control); ii) correction of soil pH with organomineral fertilizer + underground irrigation; iii) irrigation with wastewater nutrient solution (WWNS). The experimental design was randomized with nine replications, and the experiment was conducted outdoors under uncontrolled temperature conditions, from May to July. The plants were placed in the tests when they had only cotyledonary leaves.

The studied acid soil had a pH of 5.3±0.1, K 152.7±4.2 mg L^{-1} K_2O fine soil, P 32.9±3.6 mg L^{-1} P_2O_5 fine soil classified as sandy loam, 0.8±0.1% organic matter, with 50.6% coarse sand, 30.1% fine sand, 8.8% silt and 10.2% clay (Table 4).

Monitoring of plant growth was through marking and measurements. Leaf biomass was sampled for determination of moisture, chlorophylls (a, b) and carotenoids.

2.6. Preparation of WW nutrient solution

WWNS was prepared from the TWW, with the addition of GW, until the ideal pH and conductivity characteristics for plants growth (*Brassica rapa L. var. chinensis (L.) Kitam*) were obtained. These vegetables are considered moderately salinity sensitive with a threshold EC of 1.3 dS m^{-1}, and the salinity of the irrigation water can compromise the vegetative growth and root development of the plant. (Ünlükara et al. 2008).

Dilution with GW was optimized to achieve the reduction of water salinity (see Table 2) as well as its toxic effect. According to Figure 1, toxicity is eliminated after dilution above 25%. Thus, the ideal condition for WWNS is the addition of 1:4 groundwater, obtaining a pH of 7.2±0.13 and EC of 1.1 ± 0.63 dS m^{-1} (Table 2).

Table 2. Characterization of WW Samples

Parameter	Units	Raw WW*	TWW*	GW	WWNS

pH	Sorensen scale	4.26±0.03	12.41±0.24	7.79±0.05	7.17±0.13
Electric Conductivity	dS m^{-1}	1.243±0.034	6.545±0.62	0.525±0.16	1.140±0.63
Total hardness	mg CaCO$_3$ L^{-1}	424.78±53.10	1804.97±350.75	319.17±5.52	345.69±23.77
Magnesium	mg Mg L^{-1}	98.71±13.33	179.89±58.63	32.01±2.07	76,75±1.86
Calcium	Mg Ca L^{-1}	7.09±6.14	426.76±84.46	75.12±5.35	134.65±8.59
COD	mg O$_2$ L^{-1}	3225±25	2509±166	0	448±48
BOD$_5$	mg O$_2$ L^{-1}	1266 ±208	280±0	11±1	17 ±2
BOD$_5$/COD	-	0.39	0.11	---	0.04
Turbidity	NTU	159.77±7.68	2.00±0.84	---	---
N- NH$_4^+$	mg NH$_4$ L^{-1}	0.72±0.00	5.38±0.00	0	0
N Kjeldhal	mg N L^{-1}	22.3±1.6	7.58±0.00	0.19±0.32	1.86±0.32
Ptotal	mg P L^{-1}	155.69±6.53	20.3±8.65	4.41±1.78	9.54±0
Phosphates	mg PO$_4$ L^{-1}	---	---	---	---
Chlorides	mg L^{-1}	124.7±1.5	121.9±23.8	23.3±0.15	78.9±5.8
Nitrates	mg NO$_3^-$ L^{-1}	5.9±0.4	12.6±2.7	86.7±0.3	21.2±0
Fluorides	mg L^{-1}	166.7±2.3	176.0±11.1	0.03±0.0	36.3±0.1
Sulphates	mg SO$_4^{2-}$ L^{-1}	95.4±0.5	56.5±14.3	34.4±0.2	52.5±0.3
Iron	mg Fe L^{-1}	5.725±1.591	0.030±0.009	---	0.030±0.001
Manganese	mg Mn L^{-1}	0.173±0.018	0.005±0.007	---	0.005±0.001
E. Coli	ufc/100mL	0	0	0	---
Fecal coliforms	ufc/100mL	45±29.8	0	0	---
Enterococus	NMP/mL	3.7×10^{-3}	0	0	---

* Luz et al. 2021 (mean±SD; n = 3; p<0.05)

2.7. Sludge

After applying the processes of immediate precipitation of lime, used in the study, the sedimentation and decantation process is carried out, where the TWW is separated from the precipitate (sludge), which, due to its characteristics, pH 12.40±0.02, EC 7.25±0.15 dS m -1, Magnesium 0.0381±0.0067 mg L^{-1} (Luz et al. 2021), was used as an agricultural corrective in the studied acid soil.

2.8. Statistics

In the *V. fischeri* bioluminescence inhibition test, the EC$_{50}$ (%) values were determined using LUMISsoft 4 Software™. The EC$_{50}$ (%) for the *T. platyurus* and *D. magna* mortality were determined using the probit analysis (Finney 1971). Data for the inhibition growth bioassay were checked for homogeneity of variance by the

Kolmogorov–Smirnov test and, when possible, subjected to one-way analysis of variance (ANOVA). Data which did not satisfy the assumption for ANOVA, were analyzed non-parametrically using Kruskal–Wallis ANOVA by ranks test. Whenever significant differences were found (*p*<0.05), a post hoc Dunnett's test was used to compare treatments with the control, for an alpha value of 0.05 (Zar 1996).

For a better interpretation of toxicity data, all measurements (EC_{50}) were converted into Toxicity Units (TU), i.e. the inverse of EC_{50} values, expressed in percent:

$$TU (\%) = 1/EC_{50} \times 100 \qquad \text{(Equation 5)}$$

With the toxicity classification system (TCS) reported by Personne et al. (2003), it is possible to classify the samples as: class 1 ($TU < 1$) exhibiting no significant toxicity; class 2 ($1 < TU < 10$) exhibiting significant toxicity; class 3 ($10 < TU < 100$) exhibiting high toxicity; and class 4 ($TU > 100$) exhibiting very high toxicity.

The results of the parameters analyzed in the remaining tests were submitted to analysis of variance (ANOVA), and their means were compared by Tukey's test, at the 0.05% level of significance, using the GraphPad.

In the ANOVA analysis of variance, it was determined that whenever the F value was greater than the critical F, there was a significant effect between treatments. Whenever F was less than the critical F, there was no statistically significant difference.

3. RESULTS

3.1. Ecotoxicological Assessment

This study presents results of the ecotoxicological analysis of several water matrices obtained from the chemical treatment applied to winery wastewaters, and the assessment of their potential use (diluted with groundwater) as irrigation water. The ecotoxicological assessment allowed to analyze the risk to the aquatic ecosystem by using the irrigation solution prepared (produced from treated winery wastewaters). The ecotoxicological results using the bioindicators *T. platyurus*, *V. fischeri* and *P. subcapitata* are summarized in Table 3. Further, the results for growth inhibition of *P. subcapitata* (a sub-lethal parameter) are shown in Figure 2.

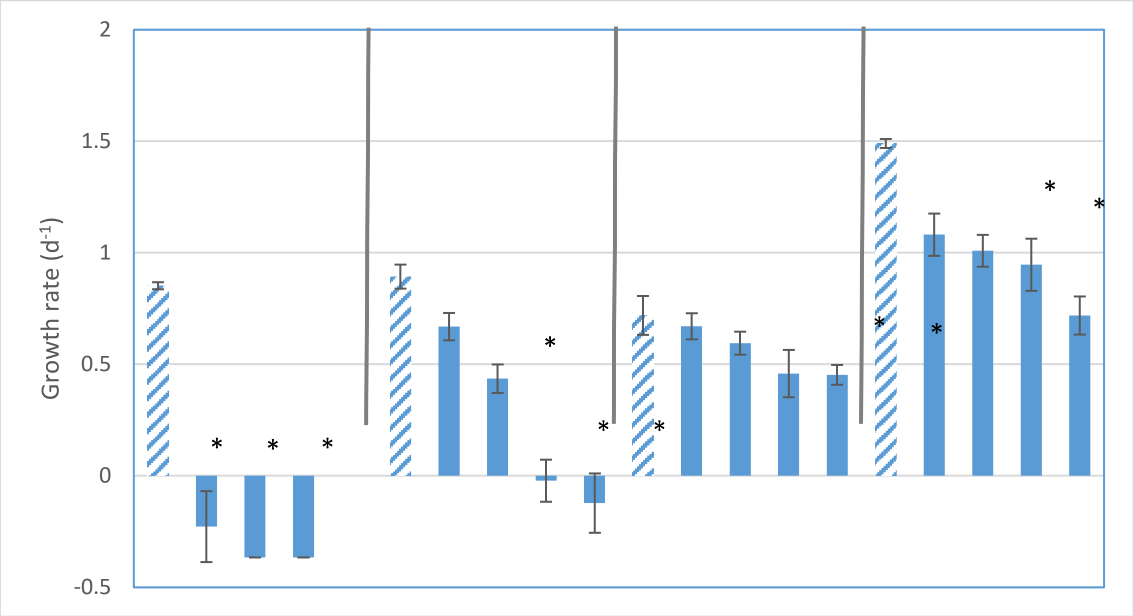
The lethal ecotoxicological results (Toxic Unit (TU); Table 3) indicated that the winery effluent was very toxic to the three bioindicators, being classified as class 4 (TU> 100; with very high toxicity), constituting a serious danger to the receiving environment. The green microalgae was the organism most sensitive to this effluent with a 72h-EC_{50} = 0.02% for growth inhibition. The toxicity detected may be correlated to: (i) the concentrations of chlorides (125 mg L^{-1}), sulphates (195 mg L^{-1}), ammonium (0.72 mg L^{-1}), and fluorides (7 mg L^{-1}) (see Table 2); and (ii) unknown organic contaminants. Authors such as, Alzahrani et al. (2013), Palma et al. (2009) and Alvarenga et al. (2009), verified the influence of concentrations of ammonia, chlorides and sulfates in the growth inhibition or mortality of aquatic organisms. The ecotoxicological effect of the one-step lime precipitation process, i.e., the capacity to reduce the wastewater ecotoxicity, was evident, resulting in a treated wastewater classified as class 2 (using the TU classification) with a lower acute toxicity for all the bioindicators used. The lime precipitation treatment led to the decrease of the concentrations of chlorides, sulphates, iron and manganese (see table 3). Despite that, ammonia concentrations are still high 5.4 mg L^{-1}, as well as sulfates concentrations (52.5 mg SO_4^{2-} L^{-1}) and fluorides (36.3 mg L^{-1}), contributing, therefore, to the remaining observed toxicity. Dilution of the treated wastewater with groundwater (1:4) resulted in a water matrix classified as class 1 (TU<1) in terms of toxicity. Although most studies conducted with wastewaters reported *V. fisheri* as the most sensitive bioindicator to ecotoxicological assays (Alzahrani et al., 2013; Latorre et al. 2007; Zgórska 2011; Palma et al. 2016), these results showed that *P. subcapitata* was in this case the most sensitive organism. This fact highlighted the need to include these two bioindicators in a battery of ecotoxicity tests to assess the environmental risk of industrial effluents, as well as in the discussion of the effectiveness of different effluent treatments.

Table 3. Values of TU (%) obtained with *V. fischeri, P. subcapitata* and the crustacean *T. platyurus* when exposed to the winery wastewater (WW), treated wastewater (TWW); groundwater (GW) and wastewater nutritive solution (WWNS). The matrices were classified into toxicity classes according to their value of toxicity units (TU).

	TU (%)			
	T. platyurus **(24h)**	***V. fisheri*** **(30 min)**	***P. subcapitata*** **(72h)**	
WW	20	9.17	100	Class 4
Treated WW	1.34	2.92	3.97	Class 2
GW	non toxic	non toxic	non toxic	Class 1
WWNS	non toxic	non toxic	non toxic	Class 1

The ecotoxicological assessment through sublethal parameters showed that the winery wastewater significantly induced growth inhibition to the microalgae *P. subcapitata* (one-way ANOVA; p<0.05), with 100% of inhibition for all the concentrations exposed (Figure 1). The results indicated that all matrices induced some growth inhibition of microalgae, less accentuated in groundwater, with growth inhibition rates always below 30% (significantly different to the control; one-way ANOVA; $F_{4,18}$=10.353, p=0.00016).

WWNS (obtained from treated wastewater diluted with groundwater) induced a growth inhibition rate of 45%, indicating toxicity for the evaluated sublethal endpoints (significantly different to the control; one-way ANOVA; $F_{4,20}$=48.423, p=0.00000). Therefore, despite the decrease in toxicity, the irrigation solution still exhibited some toxicity to microalgae. The results can be explained considering the sensitivity of the bioindicator and the toxicological endpoints used (sublethal bioassays assess different stages of the bioindicator's life cycle, being more sensitive to lower concentrations of contaminants). In fact, several authors reported toxic effects in the microalgae *P. subcapitata*, when exposed to low concentrations of organic compounds such as polycyclic aromatic hydrocarbon (PAHs; Eom et al. 2007), pharmaceuticals (Yang et al., 2008) and herbicides (Pérez et al. 2011).



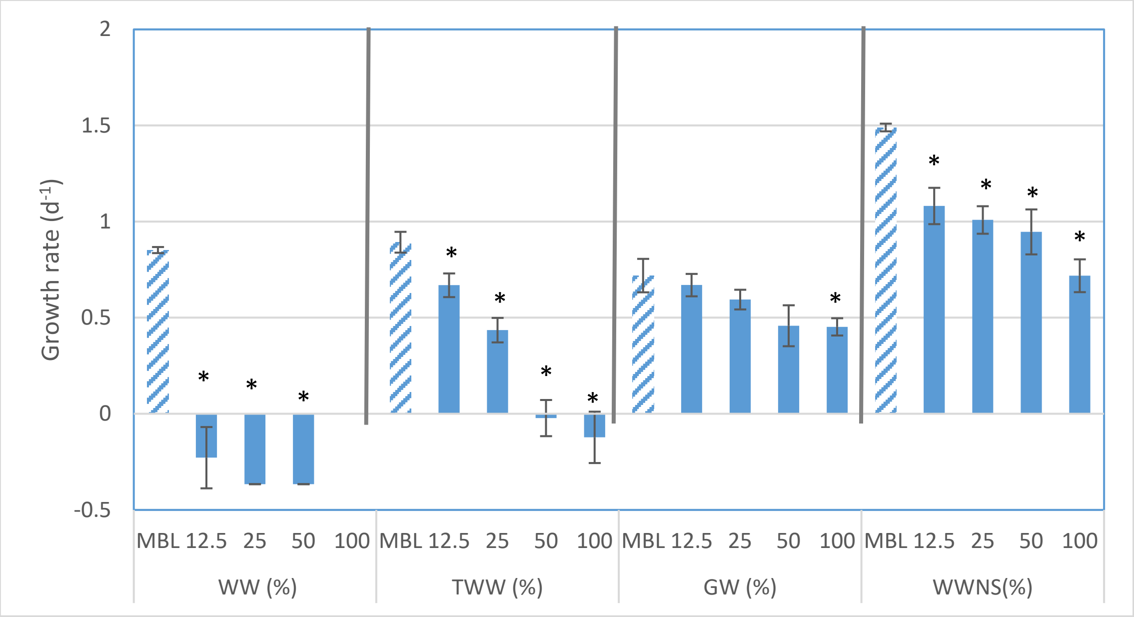


Figure 1. Growth rate (d^{-1}) of the microalgae *P. subcapitata* after 3 days exposed to the winery wastewater (WW), treated wastewater (TWW); ground water (GW) and Wastewater nutrient solution (WWNS) (mean±SD; n = 6; * p<0.05, Dunnett's test with a control (MBL).

3.2. Use of sludge amended soils in cultivars

Soil pH correction was carried out by addition of different amounts of sludge to raw soil. Figure 2 depicts the final pH achieved as a function of the percentage of added sludge. As observed with just an approximated 1.2% (w/w) of sludge the pH of the soil can be corrected to circumneutral conditions, more adequate to red pak choi growth.

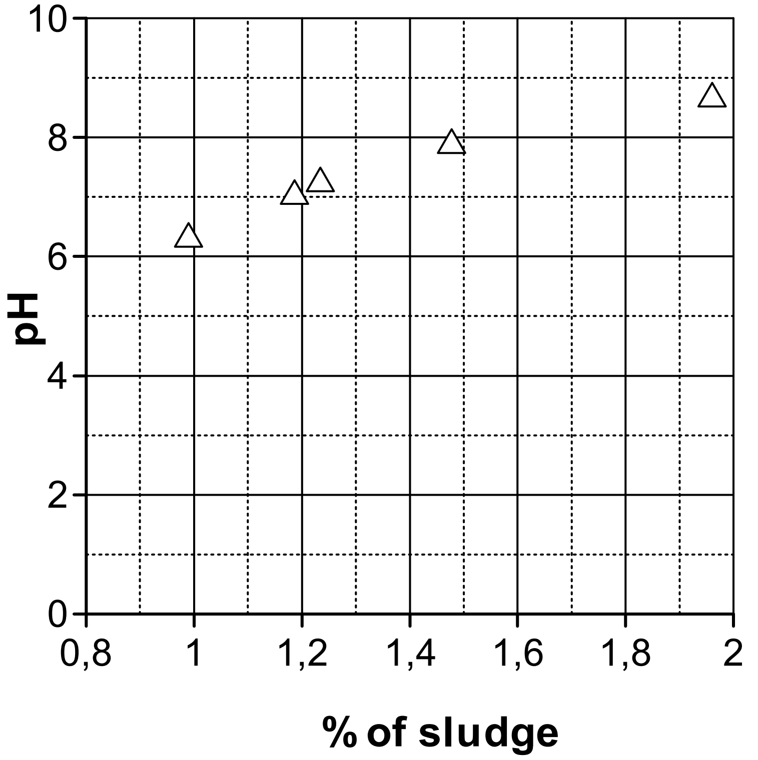


Figure 2. pH correction of acidic soil as a function of sludge percentage addition.

Table 4 compares the characterization of raw soil and corrected soil. Apart of pH, main differences are found in P content increasing from roughly 33 mg g^{-1} to 64 mg g^{-1} as P_2O_5 and K raising from 153 to 168 mg g^{-1} as K_2O. These results are sustained by the content in phosphorus and potassium of the obtained sludge after treatment, specifically 895 and 990 mg g^{-1} in P_2O_5 and K_2O, respectively (Luz et al. 2021). The critical level of nutrients differs depending on environmental conditions, soil type, plant type, method of extraction, etc. In any case, broadly speaking, P in corrected soil (14 mg g^{-1} as P) seems to be in the range of medium level content assuming that all P was available for plants while K content (139 mg g^{-1} as K) is considered slightly low if all K were extractable. The other remarkable change in concentration corresponds to Ca, which was expected given the sludge origin. Calcium deficiencies usually are found only on very acidic soils.

Table 4. Characterization of raw (acidic) and pH corrected (1.2 %) soils

	Acidic Soil		Corrected soil	
	Mean	SD	Mean	SD
pH	5.31	0.12	7.23	0.08
EC, mS cm^{-1}	0.27	-	0.40	-
% TOC	0.45	0.03	0.49	0.02
% Organic matter	0.8	0.1	0.8	-
K, mg g^{-1} K_2O	152.7	4.2	168.1	10.5
P, mg g^{-1} P_2O_5	32.9	3.6	63.7	1.7
Pb, µg g^{-1} DW	32.40	1.15	34.10	1.10
Cd, µg g^{-1} DW	2.20	0.20	2.20	0.50
Zn, mg g^{-1} DW	0.2366	0.1744	1.0197	0.5317
Fe, mg g^{-1} DW	96.647	9.1393	100.368	4.374
Ni, µg g^{-1} DW	77.30	5.00	73.90	2.00
Mn, mg g^{-1} DW	1.5116	0.0329	1.4345	0.0659
Cu, mg g^{-1} DW	0.2283	0.0118	0.2207	0.0088
Ca, mg g^{-1} DW	10.013	0.6908	102.00	20.274
Mg, mg g^{-1} DW	13.677	0.3470	15.011	0.6881

DW = Dry weight

The ecotoxicological evaluation of the different soil leachates (acid soil, soil irrigated with WWNS and corrected soil), allowed the analysis of the risk to the ecosystem after the application of the prepared irrigation solution (WWNS) and the incorporation of the precipitate as a way of correcting the pH of the soil. ground. Ecotoxicological results using the bioindicators *T. platyurus, V. fischeri* are summarized in Table 5 and the bioindicator *P. subcapitata* in Figure 3. Lethal ecotoxicological results (Toxic Unit (TU); Table 5) indicated that both irrigation and correction did not induce the increment of soil toxicity.

Table 5. Values of TU (%) obtained with *V. fischeri,* and the crustacean *T. platyurus* when exposed to the soils, acidic soil (control); irrigation with WWNS and corrected soil. The matrices were classified into toxicity classes according to their value of toxicity units (TU).

	TU (%)	
	T. Platyurus **(24h)**	***V. fisheri*** **(30 min)**

Acidic soil (Control)	non toxic	non toxic	Class 1
Irrigation with WWNS	non toxic	non toxic	Class 1
Corrected soil	non toxic	non toxic	Class 1

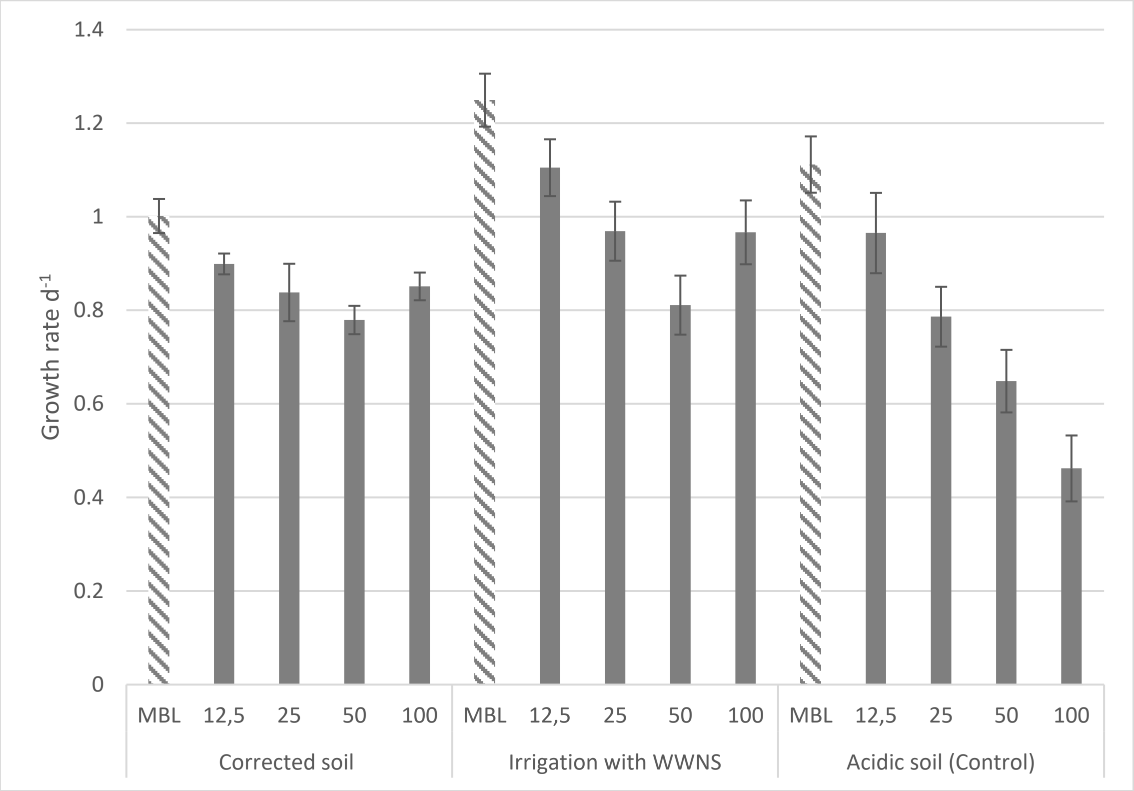


Figure 3. Growth rate (d^{-1}) of the microalgae *P. subcapitata* after 3 days exposed to the corrected soil, irrigation with WWNS and acidic soil (mean±SD; n = 6; * p<0.05, Dunnett's test with a control (MBL).

Once soils were characterized, the potential benefits of soil sludge amendment were assessed by analyzing the evolution of a control cultivar (red pak choi) under three different scenarios. Hence, red pak choi was grown in the raw acidic soil by using either groundwater or treated wastewater in the irrigation. The wastewater used was obtained after dilution (1:4) and neutralization of treated wastewater with groundwater (Luz, 2021).

Several parameters were measured to compare the effect of soil amendment and water irrigation nature on red pak choi (see Figure 4). Hence, regarding plant size a positive effect of soil correction and use of wastewater was experienced in stem diameter, plant heigh and leaf length/area while a decrease in stem heigh was obtained when the plants were cultivated under the studied systems if compared to values in acidic soil.

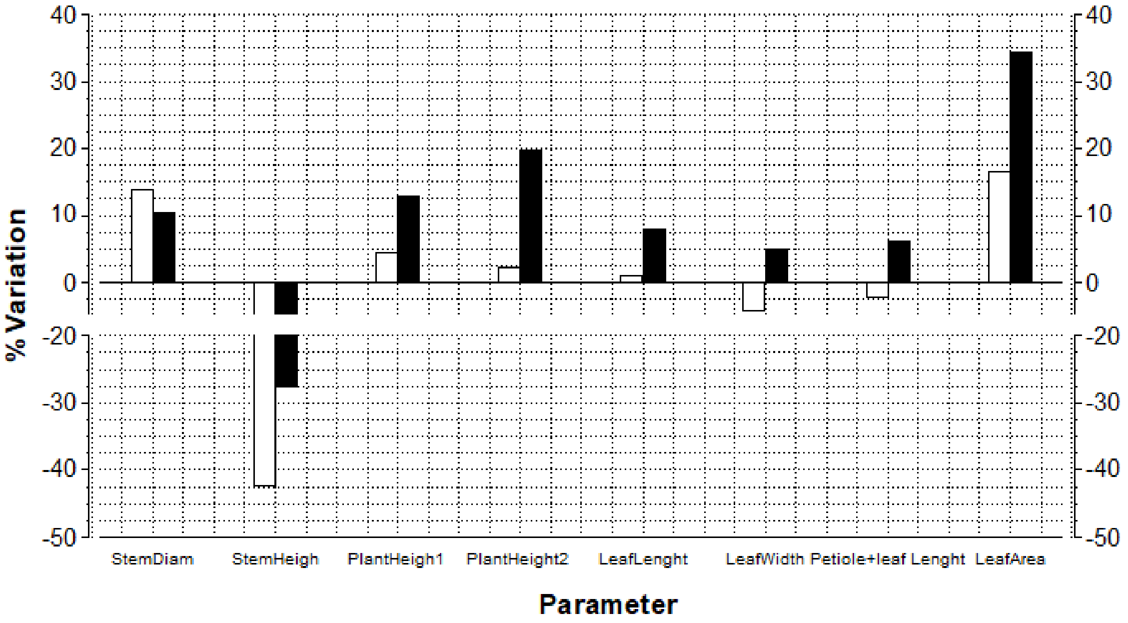


Figure 4. Use of irrigation with WWNS on acid soil (white bars) and irrigation with GW on amended soil (black bar) on red pak choi growth. Parameters compared to results obtained in uncorrected acidic soil irrigated with GW (PlantHeigh1 measured from ground to the longest leaf stretched, PlantHeigh2 measured from ground to the largest leaf without stretching).

Broadly speaking, cultivars grown in amended soil (irrigated with GW) presented better characteristics than those produced by irrigating with WWNS the uncorrected soil. Moreover, wastewater watering led to a reduction in leaf width and petiole+leaf length if compared to control experiments. The most remarkable differences in amended soil were found in plant height and leaf area with an increase of roughly 20% (PlantHeigh2) and 34%, respectively, contrasted to results obtained in the acidic soil. The only parameter negatively affected by soil amendment was stem heigh with a reduction of 28% (see Figure 5).

The higher size of plants was translated to a notable increase in the dry weight of the aerial part of the vegetables, experiencing a 23% and 42 % raise in plants grown with WWNS and corrected soil, respectively.

Contrarily to aerial data, use of corrected soil led to a decrease of root weight although an increase in length was experienced. Watering with WWNS increased the size and weight of roots, especially fresh weight with a raise of 25%.

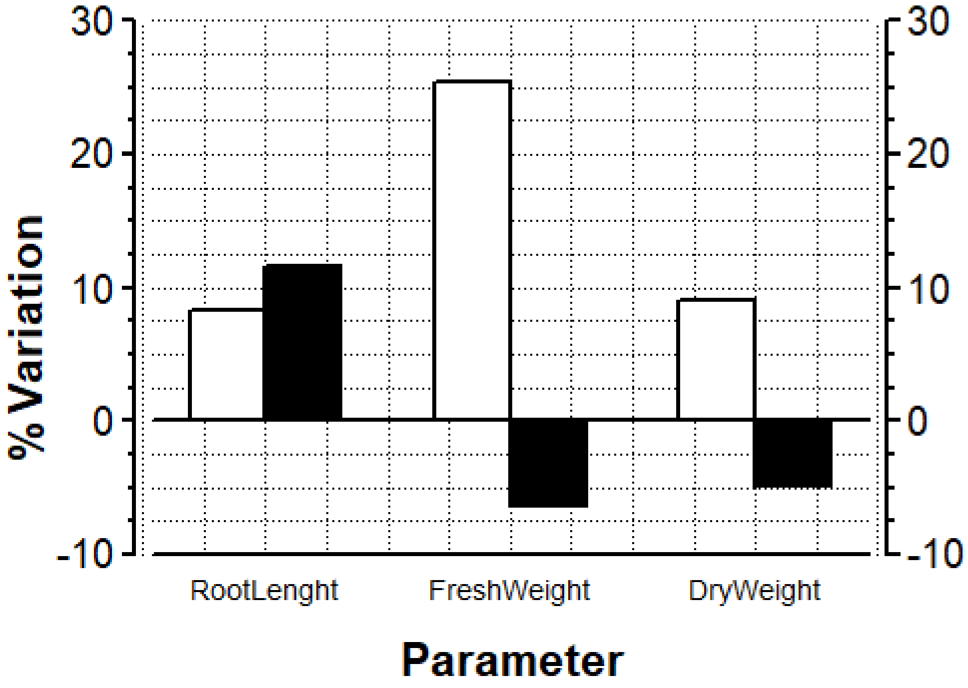


Figure 5. Use of irrigation with WWNS (white bars) or amended soil (black bar) on red pak choi growth. Root parameters compared to results obtained in uncorrected acidic soil.

3.3. Pigment content in red pak choi grown

Chlorophyll is the designation of a group of photosynthetic pigments found in the chloroplasts of plants. In green plants, chlorophyll appears in two forms: chlorophyll a and b.

This pigment, together with carotenoids, has the ability to capture solar radiation and allow plants to carry out photosynthesis, a vital function of plants, which allows the production of carbohydrates, which they use to produce essential energy for their development. and productivity.

During photosynthesis, CO_2 is consumed, and oxygen is released, hence the importance of plants for maintaining oxygen and sequestering carbon on the planet (Moreira, 2013)

As can be seen in table 6, the plants watered with WWNS, as well as those grown in the soil, where the pH correction was carried out, had higher levels of chlorophyll, which is a positive aspect, since they are plants with higher development capacity and greater productive capacity.

Nitrogen is part of chlorophyll and is directly involved in photosynthesis (Rodrigues and Coutinho, 2000).

The content in pigments (chlorophyll and carotenoids) can be an indirect tool to assess nitrogen availability in soils. Photosynthetic pigments can, therefore, detect deficiencies in nitrogen

(Balasu-Bramanian et al., 2000; Torres Netto et al., 2005). As a rule of thumb, deficiencies in N, P and K affect photosynthesis and growth (Muñoz Huerta et al., 2013), and, chlorophyll and carotenoids content is a valuable tool to reveal the shortage of these essential elements (Sanchez et al., 2018). As inferred from table xx, cultivars grown with irrigation with WWNS or in amended soils present higher levels of chlorophyll than those obtained in acidic soil. Carotenoids content does not significantly vary with the procedure of cultivation. These results suggest a positive influence of TWW and sludge amendment on NPK availability to plants.

Table 6. Pigment content in red pak choi grown under three scenarios

	Chlorophyll a mg g^{-1}		Chlorophyll b mg g^{-1}		Carotenoids mg g^{-1}	
	Mean	SD	Mean	SD	Mean	SD
Acidic soil (control)	1.55	0.265	1.06	0.262	0.24	0.040
Irrigation with WWNS	1.925	0.095	1.53	0.235	0.264	0.081
Corrected soil	1.845	0.178	1.38	0.123	0.25	0.106

4. CONCLUSION

Winery wastewater represents a serious environmental problem when not treated. Since the annual production of these tributaries is very significant, their use can be a solution for the future, offering profitability and promoting a circular economy for this industry. The raw effluent (WW) showed high toxicity, which decreased significantly after the application of the slaked lime treatment. To use it for irrigation, it was essential to meet the pH and conductivity characteristics that the plant needs, being necessary to dilute the TWW with GW, thus making it an ideal nutrient solution for the cultivar. Ecotoxicologically, WWNS did not show any toxicity. An agronomic study was then carried out using three different conditions: irrigation with groundwater (soil control); correction of soil pH with organomineral fertilizer and underground water irrigation; irrigation with WWNS, for the production of red pak choi.

All soils were analysed ecotoxicologically using indirect bioassays, and none showed toxicity. After the cultivar reached the optimal harvesting conditions, it was analysed in terms of pigmentation, through the study of chlorophylls a, b and carotenoids. All parameters presented favourable conditions for healthy plant growth.

DECLARATIONS

Consent for publication

All authors agreed with the content and that all gave explicit consent to submit.

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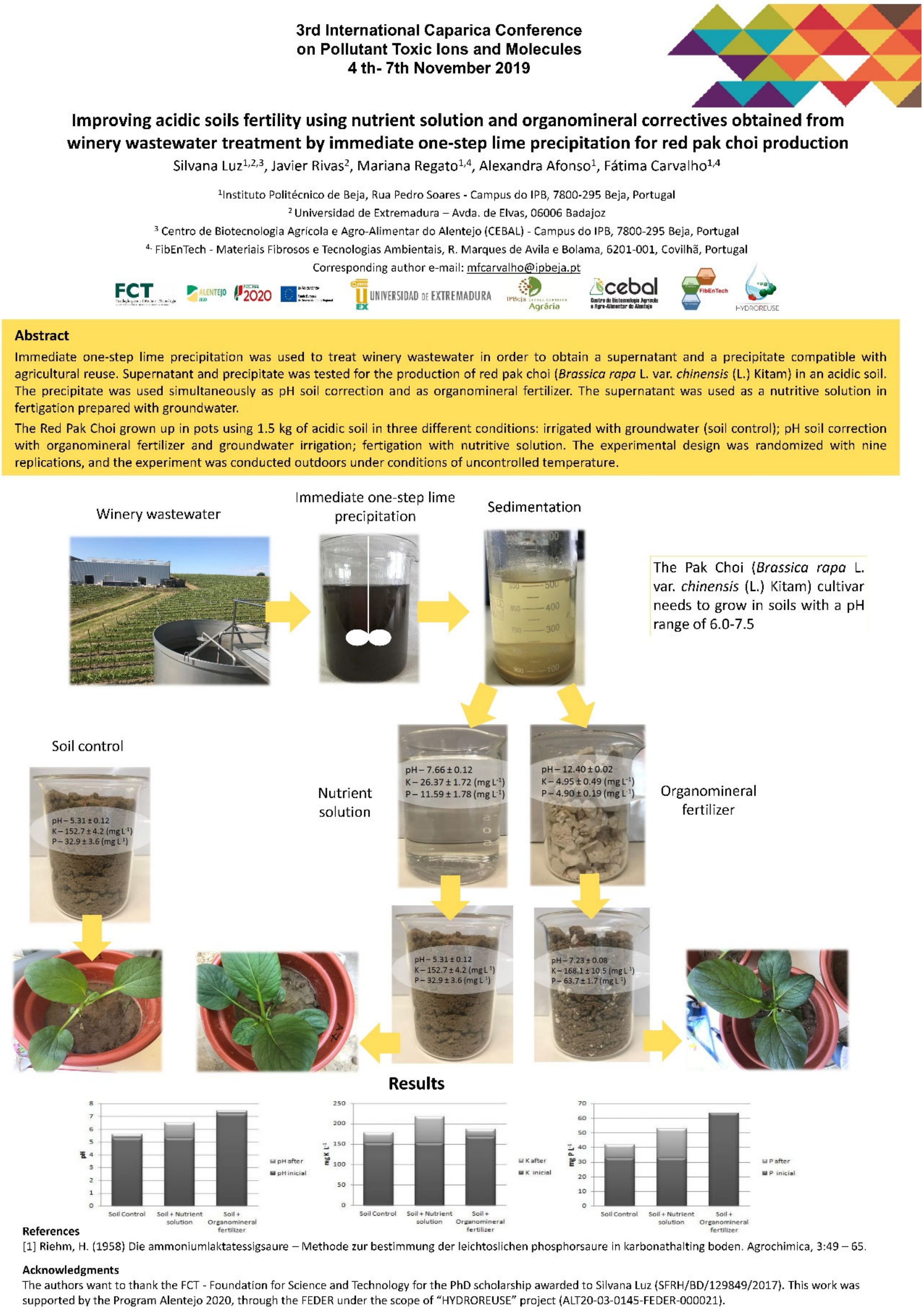
Comunicação 6 - Improving acidic soils fertility using nutrient solution and organomineral correctives obtained from winery wastewater treatment by immediate one-step lime precipitation for red pak choi production



3rd International Caparica Conference
on Pollutant Toxic Ions and Molecules
4 th- 7th November 2019

Improving acidic soils fertility using nutrient solution and organomineral correctives obtained from winery wastewater treatment by immediate one-step lime precipitation for red pak choi production

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Abstract

Immediate one-step lime precipitation was used to treat winery wastewater in order to obtain a supernatant and a precipitate compatible with agricultural reuse. Supernatant and precipitate was tested for the production of red pak choi (*Brassica rapa* L. var. *chinensis* (L.) Kitam) in an acidic soil. The precipitate was used simultaneously as pH soil correction and as organomineral fertilizer. The supernatant was used as a nutritive solution in fertigation prepared with groundwater.

The Red Pak Choi grown up in pots using 1.5 kg of acidic soil in three different conditions: irrigated with groundwater (soil control); pH soil correction with organomineral fertilizer and groundwater irrigation; fertigation with nutritive solution. The experimental design was randomized with nine replications, and the experiment was conducted outdoors under conditions of uncontrolled temperature.

The Pak Choi (*Brassica rapa* L. var. *chinensis* (L.) Kitam) cultivar needs to grow in soils with a pH range of 6.0-7.5

Results

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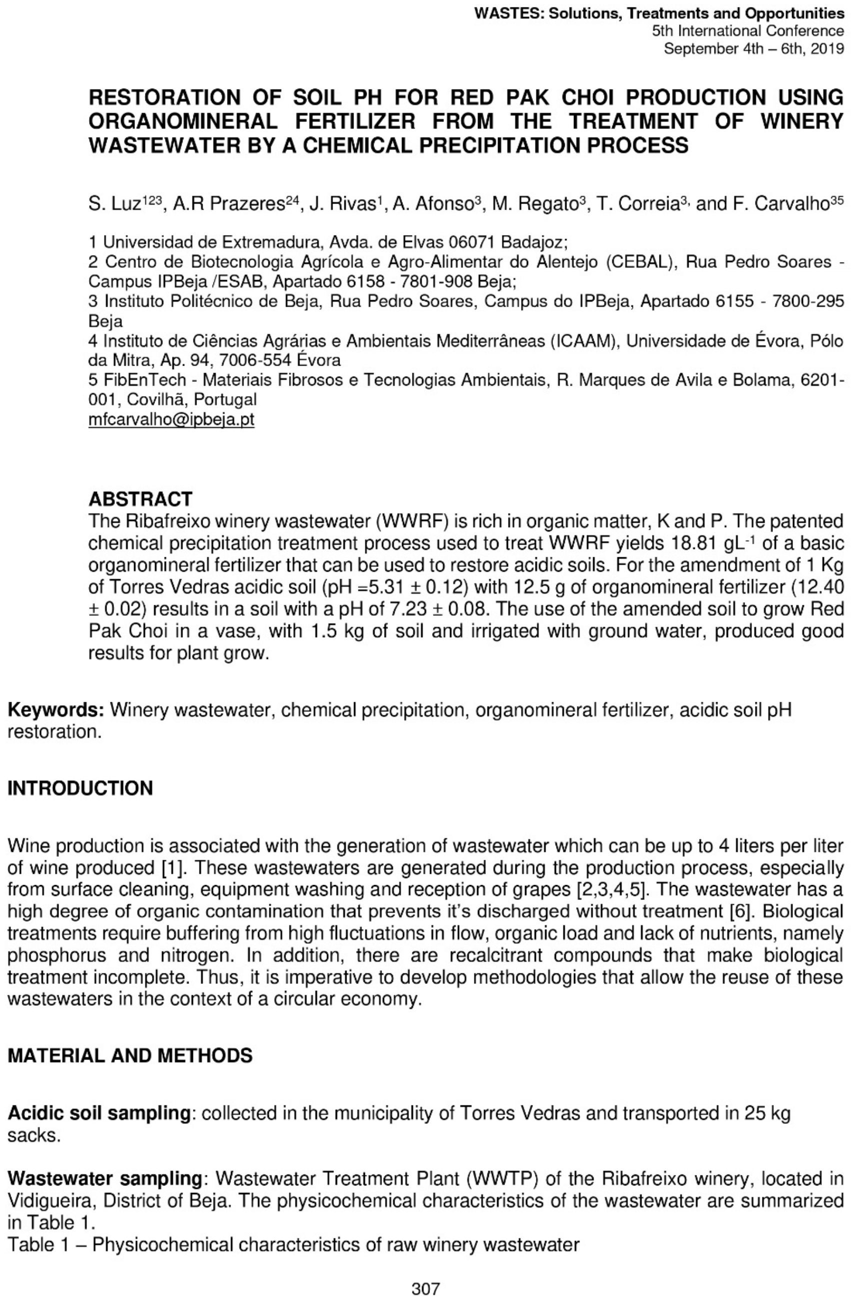
Comunicação 7 - Restoration of soil pH for red pak choi production using organomineral fertilizer from the treatment of winery wastewater by a chemical precipitation process



WASTES: Solutions, Treatments and Opportunities
5th International Conference
September 4th – 6th, 2019

RESTORATION OF SOIL PH FOR RED PAK CHOI PRODUCTION USING ORGANOMINERAL FERTILIZER FROM THE TREATMENT OF WINERY WASTEWATER BY A CHEMICAL PRECIPITATION PROCESS

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ABSTRACT

The Ribafreixo winery wastewater (WWRF) is rich in organic matter, K and P. The patented chemical precipitation treatment process used to treat WWRF yields 18.81 gL^{-1} of a basic organomineral fertilizer that can be used to restore acidic soils. For the amendment of 1 Kg of Torres Vedras acidic soil (pH =5.31 ± 0.12) with 12.5 g of organomineral fertilizer (12.40 ± 0.02) results in a soil with a pH of 7.23 ± 0.08. The use of the amended soil to grow Red Pak Choi in a vase, with 1.5 kg of soil and irrigated with ground water, produced good results for plant grow.

Keywords: Winery wastewater, chemical precipitation, organomineral fertilizer, acidic soil pH restoration.

INTRODUCTION

Wine production is associated with the generation of wastewater which can be up to 4 liters per liter of wine produced [1]. These wastewaters are generated during the production process, especially from surface cleaning, equipment washing and reception of grapes [2,3,4,5]. The wastewater has a high degree of organic contamination that prevents it's discharged without treatment [6]. Biological treatments require buffering from high fluctuations in flow, organic load and lack of nutrients, namely phosphorus and nitrogen. In addition, there are recalcitrant compounds that make biological treatment incomplete. Thus, it is imperative to develop methodologies that allow the reuse of these wastewaters in the context of a circular economy.

MATERIAL AND METHODS

Acidic soil sampling: collected in the municipality of Torres Vedras and transported in 25 kg sacks.

Wastewater sampling: Wastewater Treatment Plant (WWTP) of the Ribafreixo winery, located in Vidigueira, District of Beja. The physicochemical characteristics of the wastewater are summarized in Table 1.
Table 1 – Physicochemical characteristics of raw winery wastewater

Parameter	Unity	Winery wastewater	Parameter	Unity	Winery wastewater
pH	Sørensen scale	11.6	Magnesium	mg L^{-1}	7
Conductivity	dS m^{-1}	3.21	Potassium	mg L^{-1}	100.5
Turbidity	NTU	27.0	Sodium	mg L^{-1}	162.6
COD	mg O_2 L^{-1}	1943	Nitrates	mg L^{-1}	2.8
Total phosphorus	mg L^{-1}	235	Nitrites	mg L^{-1}	0
Calcium	mg L^{-1}	18	-	-	-

The organomineral fertilizer is obtained by air drying the precipitate from the WWRF chemical precipitation treatment process [7]. The precipitate characteristics are summarized in Table 2.

Table 2 – Characteristics of the precipitate obtained by chemical precipitation of WWRF

Precipitate characteristics	Treated
Volume	0.04 ± 0.01 LL^{-1}
Density	1.4± 0.1 kgL^{-1}
% dry matter	33.59 ± 0.09

According the data of the Table 2 (density, volume of precipitate and % dry matter), we can calculate the amount of organomineral fertilizer produced by liter of WWRF treated by chemical precipitation process, 18.81 gL^{-1}.

The physicochemical characteristics of the organomineral fertilizer are summarized in Table 3.

Table 3 – Physicochemical characteristics of organomineral fertilizer

Parameter	Unity	Treated
pH	Sørensen scale	12.40 ± 0.02
Conductivity	dS m^{-1}	*7.25* ± 0.15
Organic matter	%	9.89 ± 1.55

INSTALLATION AND EXPERIMENTAL DESIGN

Soil Correction Test: The organomineral fertilizer was wrapped and incorporated manually into the soil to obtain a homogeneous sample.
Plant growth test: The cultivars, Red Pak Choi, were planted in pots containing 1.5 kg of soil when they had two cotyledon leaves: The control test was carried out in the Torres Vedras acidic soil and the acid soil correction occurred by the addition of 18.75 g of organomineral fertilizer. The experimental design was completely randomized, with nine replicates, and the experiment was run outdoors under uncontrolled temperature conditions and irrigation was done with groundwater.

RESULTS AND DISCUSSION

The Pak Choi cultivar needs to grow in soils with a pH range of 6.0-7.5 [8]. The study of the application of the organomineral fertilizer for the amendment of acidic soil pH in the Torres Vedras area showed that it is necessary to use 12.5 g of organomineral fertilizer per 1 kg of soil. The physicochemical changes produced by the organomineral fertilizer in the amended soil are summarized in Table 4.

Table 4– Influence of the amount of organomineral fertilizer in the physical characteristics of soil

Parameter	Unity	acid soil	Restored soil
pH	Sørensen scale	5.31 ± 0.12	7.23 ± 0.08
Conductivity	$dS\ m^{-1}$	0.270 ± 0.003	0.396 ± 0.002

The use of pH amended soil in the cultivation of Red Pak Choi allowed for a favorable growth, as can be seen in Figure 1. On the day of transplantation, each cultivar had only two cotyledon leaves, one day later it began to develop the first definitive leaf and after three weeks of the transplant already had 4 definitive leaves.

Figure 1 – 1 - day 1; 2 – Day 6; 3- Day 8; 3- Day 15

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CONCLUSÃO

As agroindústrias de queijaria e adega são geograficamente dispersas, localizadas longe da rede de coletores municipais e produzem de forma sazonal consideráveis quantidades diárias de efluentes. Apresentam, por isso, pouca capacidade técnica e económica que lhes permita utilizar as tecnologias existente para tratamento destes efluentes. Consequentemente, na maioria dos casos, descarregam os seus efluentes sem tratamento prévio colocando em risco os meios recetores. O elevado teor em sais e matéria orgânica do efluente de queijaria deve-se essencialmente a perdas de soro e sorelho durante o processo de fabrico de queijo e requeijão. O elevado teor salino é um entrave quando se pretende a sua reutilização agrícola. As caraterísticas físico-químicas e o caudal do efluente de adega variam muito ao longo do ano, devido à sazonalidade da indústria, aumentando bastante o volume de produção no período da vindima (em Portugal realiza-se geralmente nos de julho a setembro). O Efluente da adega apresenta um pH ácido e elevados teores de matéria orgânica e sólidos provenientes das películas de uva, grainhas, mosto e borras resultantes do processo produtivo.

O desenvolvimento de tecnologias de baixo custo e fácil aplicação *in loco* que permita às agroindústrias transitar facilmente para uma economia circular é de crucial importância.

Como resultado deste trabalho concebeu-se uma tecnologia facilmente adaptável a pequenas e médias indústrias de produção de queijo, por utilizar reagentes baratos, fáceis de encontrar no mercado ($CaCO_3$ cal e $Ca(OH)_2$) e como opera a pH neutro, não necessita de correções adicionais de pH para a sua reutilização. Alem disso, permite alcançar elevadas eficiências de remoção para estas águas residuais como se pode verificar na Tabela 2.

Tabela 2 – Resumo da eficácia das tecnologias desenvolvidas para tratamento de efluentes de queijaria e adega.

Reagente (g L^{-1})	**pH (inicial)**	**pH (final)**	**CQO (%)**	**P_{total} (%)**	**Fenóis (%)**	**Turvação (%)**	**O&G (%)**
Coagulantes combinados – Efluente Queijaria							
$CaCO_3$ 75 g L^{-1}+ $Ca(OH)_2$ 0,6g L^{-1}	5,1	6,9	46,2	92,6	49	80,9	98,9
Precipitação básica – Efluente Adega							
$Ca(OH)_2$ 25 g L^{-1}	4,3	12,4	22,2	86,9	99,95	97,6	-

O&G - Óleos e gorduras

A Tecnologia de precipitação química básica imediata tinha já sido apontada em muitos trabalhos como sendo uma solução para o tratamento de efluentes de queijaria, matadouro, vinhaça de cana de açúcar, explosivos, lixiviados de aterros sanitários e efluentes urbanos, porém, nunca aplicação ao tratamento de efluentes de adega. Neste trabalho demonstrou-se também a sua eficácia no tratamento de efluentes de adega, incluindo a neutralização do efluente tratado a partir do CO_2 atmosférico e a produção de sobrenadantes e precipitados agronomicamente valorizáveis.

O sobrenadante e o corretivo organomineral quando aplicados como solução nutritiva e como corretivo organomineral, em solo ácido, para a produção de red pak choi apresentaram baixo impacto ambiental devido à ausência de toxicidade de acordo com os bioindicadores *Thmanocephalus platyurus, Vibrio fisheri e Pseudokirchneriella subcapitata* demonstrando o seu potencial de valorização.

Pelo acima exposto, as tecnologias desenvolvidas permitirão a modernização e a reabilitação das agroindústrias do queijo e vinho pela valorização e uso de recursos que minimizam danos ambientais e fomentam a economia circular em alinhamento com o preconizado pela ONU para a agenda 2030 de desenvolvimento sustentável.

O desenvolvimento de práticas inovadoras, sustentáveis e economicamente viáveis para o tratamento de efluentes de todo o tipo de queijarias e adegas está em alinhamento com o objetivo 6 (água potável e saneamento) e com o objetivo 9 (indústria inovação e infraestruturas).

Para além disso, a tecnologia de precipitação química imediata em meio básico e de etapa única ao fazer a captura espontânea do CO_2 atmosférico também alia tratamento de água ao combate às alterações e, nesse sentido, está alinhada com o objetivo 13 (ação climática).

Para trabalhos futuros,

Valorizar o processo desenvolvido e patenteado, EP3447030B1 através da:

- verificação da sua eficiência noutro tipo de águas residuais, incluindo as domésticas;
- aplicação do tratamento à escala piloto para a elevação do *Technology Readiness Level* (TLR);
- reutilização do efluente e precipitado obtidos em diferentes tipos de águas residuais.

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