UNIVERSIDADE FEDERAL DA PARAÍBA

CENTRO DE TECNOLOGIA

PROGRAMA DE PÓS-GRADUAÇÃO EM CIÊNCIA E TECNOLOGIA DE ALIMENTOS

LÍVIA DUARTE DE MEDEIROS

INFLUÊNCIA DAS LEVEDURAS AUTÓCTONES DE ENGENHOS DE CACHAÇA NA PRODUÇÃO DE HIDROMEL

JOÃO PESSOA – PB

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Dissertação apresentada ao Programa de Pós-Graduação em Ciência e Tecnologia de Alimentos, Centro de Tecnologia, Universidade Federal da Paraíba em cumprimento aos requisitos para obtenção do título de Mestre em Ciência e Tecnologia de Alimentos.

Orientador: Profº. Dr. Normando Mendes Ribeiro Filho
Coorientador: Profº. Dr. Marcelo Barbosa Muniz

JOÃO PESSOA– PB
2025

Catalogação na publicação
Seção de Catalogação e Classificação

M488i Medeiros, Livia Duarte de.
Influência das leveduras autóctones de engenhos de cachaça na produção de hidromel / Livia Duarte de Medeiros. - João Pessoa, 2025.
60 f. : il.

Orientação: Normando Mendes Ribeiro Filho.
Coorientação: Marcelo Barbosa Muniz.
Dissertação (Mestrado) - UFPB/CT.

1. Leveduras autóctones. 2. Fermentação. 3. Hidromel. I. Ribeiro Filho, Normando Mendes. II. Muniz, Marcelo Barbosa Muniz. III. Título.

UFPB/BC CDU 664(043)

Elaborado por GRACILENE BARBOSA FIGUEIREDO - CRB-15/794

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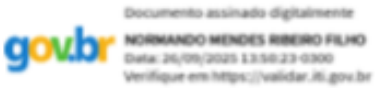
DATA:29/08/2025

BANCA EXAMINADORA



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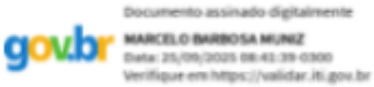

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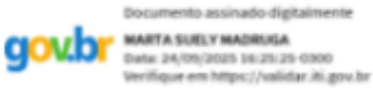

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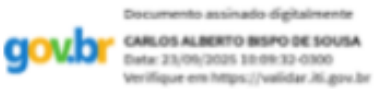

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Dedico este trabalho ao meu pai,
minha força constante e maior inspiração.

AGRADECIMENTOS

Aos meus pais e familiares, por todo apoio emocional e financeiro, e aos meus amigos, por serem abrigo em momentos difíceis. Agradeço aos colegas da pós-graduação, em especial Matheus, Rayane, Isabel, Júlia e Maria Clara, bem como aos técnicos de laboratório Mércia, Clóvis, Éllyda e Leila, pelo auxílio fundamental durante a realização desta pesquisa. Ao meu orientador, Prof. Dr. Normando Mendes Ribeiro Filho, e ao coorientador, Prof. Dr. Marcelo Barbosa Muniz, expresso minha sincera gratidão pela orientação, apoio e incentivo durante toda a realização deste trabalho. A todos os professores que contribuíram com o trabalho e/ou cederam a estrutura de seus laboratórios: Profª Drª Marta Suely Madruga, Prof. Dr. Carlos Alberto Bispo de Sousa, Prof. Dr. Flavio Luiz Honorato da Silva e Prof. Dr. Julice Dutra Lopes. Por fim, agradeço à Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) pelo apoio concedido por meio da bolsa de estudos, a qual possibilitou a dedicação integral a esta pesquisa e contribuiu de maneira significativa para o desenvolvimento deste trabalho.

Sonho que se sonha só é só um sonho,
mas sonho que se sonha junto é realidade.

Raul Seixas

RESUMO

O mel é uma substância natural de valor histórico e nutricional, amplamente apreciada por suas propriedades funcionais e sensoriais. Sua composição rica em açúcares, compostos fenólicos e voláteis permite seu uso como substrato na produção de bebidas fermentadas, como o hidromel, cuja identidade sensorial está diretamente relacionada tanto à origem do mel quanto às leveduras utilizadas no processo. Este estudo teve como objetivo avaliar o desempenho da fermentação e a composição química do hidromel produzido utilizando cepas de leveduras autóctones isoladas de fermentações de cachaça, destacando seu potencial para melhorar a qualidade sensorial e as propriedades bioativas da bebida. Considerando o potencial da biodiversidade brasileira e a tradição da produção de cachaça, foram aplicadas duas cepas autóctones (TF e J), isoladas de engenhos de cachaça, e uma linhagem comercial (C), na fermentação de hidromel elaborado com mel da região semiárida da Paraíba. As análises iniciais do mel indicaram qualidade adequada para a fermentação, conforme parâmetros físico-químicos legais. Durante o processo fermentativo, as cepas autóctones demonstraram boa adaptação, especialmente a TF, que gerou maior produção de etanol e menor doçura residual. A cepa comercial, por outro lado, apresentou fermentação limitada, resultando em uma bebida com menor teor alcoólico. Em relação aos parâmetros cinéticos, as três leveduras mostraram comportamentos bem distintos durante a fermentação: a cepa TF apresentou o melhor desempenho fermentativo, com maior crescimento, consumo de açúcar e produção de etanol e CO_2, destacando-se em quase todos os parâmetros cinéticos e de rendimento; a levedura comercial (C) teve os piores resultados, com baixa eficiência na conversão de substrato em etanol e biomassa. Além disso, foram identificados 128 compostos voláteis nos hidroméis, com diferenças na composição de acordo com a linhagem utilizada. Compostos como álcool isoamílico e acetato de isoamila contribuíram de forma expressiva para o aroma, embora não tenham se mostrado eficazes para distinguir os diferentes perfis. Em termos de compostos fenólicos totais, a cepa autóctone TF apresentou o maior teor. A capacidade antioxidante medida por ABTS e DPPH não diferiu entre as formulações, indicando resposta similar entre as leveduras nesses ensaios. No entanto, pelo método FRAP, a levedura comercial (C) destacou-se com o maior valor. Em relação ao perfil de fenólicos, ácidos orgânicos e açúcares, TF apresentou elevada concentração de hesperidina e praticamente esgotou a frutose, ao passo que J e C acumularam naringina e frutose residual. A levedura comercial (C) apresentou o maior teor de ácido cítrico, enquanto o ácido acético foi mais elevado nas formulações TF e J. Os dados obtidos destacam o potencial das leveduras autóctones na produção de hidromel com características sensoriais diferenciadas e influência significativa sobre parâmetros bioativos, abrindo caminhos para a valorização de recursos locais e para a inovação na cadeia de bebidas fermentadas.

Palavras-chave: Fermentação alcoólica; Leveduras autóctones; Hidromel; Compostos voláteis; Álcool isoamílico; Acetato de isoamila.

ABSTRACT

Honey is a natural substance of historical and nutritional value, widely appreciated for its functional and sensory properties. Its rich composition of sugars, phenolic compounds, and volatile compounds allows it to be used as a substrate in the production of fermented beverages, such as mead, whose sensory identity is directly related to both the origin of the honey and the yeasts used in the process. This study aimed to evaluate the fermentation performance and chemical composition of mead produced using indigenous yeast strains isolated from cachaça fermentations, highlighting their potential to improve the sensory quality and bioactive properties of the beverage. Considering the potential of Brazilian biodiversity and the tradition of cachaça production, two indigenous strains (TF and J), isolated from cachaça mills, and a commercial strain (C), were applied to the fermentation of mead made with honey from the semiarid region of Paraíba. Initial analyses of the honey indicated adequate quality for fermentation, meeting legal physicochemical parameters. During the fermentation process, the native strains demonstrated good adaptation, especially TF, which generated higher ethanol production and lower residual sweetness. The commercial strain, on the other hand, showed limited fermentation, resulting in a beverage with a lower alcohol content. Regarding kinetic parameters, the three yeasts displayed very distinct behaviors during fermentation: the TF strain exhibited the best fermentation performance, with greater growth, sugar consumption, and ethanol and CO_2 production, excelling in almost all kinetic and yield parameters; the commercial yeast (C) had the worst results, with low efficiency in converting substrate into ethanol and biomass. Furthermore, 128 volatile compounds were identified in the meads, with compositional differences depending on the strain used. Compounds such as isoamyl alcohol and isoamyl acetate contributed significantly to the aroma, although they were not effective in distinguishing the different profiles. In terms of total phenolic compounds, the native TF strain had the highest content. The antioxidant capacity measured by ABTS and DPPH did not differ between the formulations, indicating a similar response among the yeasts in these assays. However, using the FRAP method, commercial yeast (C) stood out with the highest value. Regarding the phenolic, organic acid, and sugar profile, TF presented a high concentration of hesperidin and virtually depleted fructose, while J and C accumulated naringin and residual fructose. Commercial yeast (C) presented the highest citric acid content, while acetic acid was higher in TF and J formulations. The data obtained highlight the potential of indigenous yeasts in the production of mead with differentiated sensory characteristics and significant influence on bioactive parameters, paving the way for the valorization of local resources and innovation in the fermented beverage chain.

Keywords: Alcoholic fermentation; Native yeasts; Mead; Volatile compounds; Isoamyl alcohol; Isoamyl acetate.

SUMÁRIO

1. INTRODUÇÃO

O mel, alimento com uso milenar, apresenta elevado valor em função de suas propriedades nutricionais e medicinais. Produzido por abelhas do gênero *Apis* a partir do néctar floral, o mel é composto principalmente por açúcares redutores, como glicose e frutose. Contudo, sua composição é ainda mais complexa, contendo pequenas quantidades de proteínas, ácidos orgânicos, minerais, vitaminas, enzimas, compostos fenólicos e substâncias voláteis, que contribuem para suas propriedades funcionais (DA SILVA *et al*., 2016). A composição química do mel é diretamente influenciada pela flora da qual o néctar é coletado, o que gera variações sensoriais notáveis entre méis de distintas origens botânicas. Dentre seus componentes, os compostos voláteis se destacam por definir o perfil aromático e podem atuar como marcadores químicos eficazes na identificação da origem floral (CASTELL *et al*., 2023; ZHU *et al.,* 2022).

Dentro do universo das bebidas fermentadas, o hidromel — elaborado a partir da fermentação de uma solução de mel e água — destaca-se como uma das mais antigas bebidas alcoólicas conhecidas, com registros que datam do período Neolítico, cerca de 7000 a.C. A ampla variedade de méis disponíveis influencia diretamente as características sensoriais do hidromel (DENG *et al*., 2023). A utilização de mel multifloral na sua produção favorece a formação de compostos como o trans-nerolidol e o acetato de feniletila, responsáveis por conferir notas florais à bebida (CHITARRINI *et al*., 2020). Em contraste, o hidromel feito com mel de melada apresenta compostos como ácido caprílico e cáprico, resultando em um sabor rançoso (CHITARRINI *et al*., 2020).

A escolha da cepa de levedura utilizada na fermentação é outro aspecto fundamental na produção de hidromel. Fatores adversos, como oscilações de temperatura, limitações nutricionais, estresse osmótico e elevação na concentração de álcool, podem comprometer o desempenho fermentativo das leveduras. Nessas circunstâncias, cepas menos adaptadas podem não conseguir conduzir a fermentação de maneira eficiente, além de favorecer a formação de subprodutos indesejáveis — denominados off-flavours — que prejudicam a qualidade sensorial da bebida (BAUER & PRETORIUS, 2000; HOHMANN & MAGER, 2003; ATTFIELD, 1997; BISSON, 1999). Uma alternativa promissora para aumentar a eficiência do processo é a reutilização de leveduras, prática já consolidada em diferentes setores da indústria de fermentação. Além de reduzir custos operacionais e impactos ambientais, essa estratégia acelera a fermentação, uma vez que encurta a fase de adaptação celular. Ademais, o uso sucessivo da mesma biomassa pode favorecer o desenvolvimento de

características adaptativas nas leveduras, tornando-as mais resistentes a condições específicas do processo, como flutuações de pH, variações térmicas e elevados teores alcoólicos (WHITE & ZANAISHEFF, 2010).

Este trabalho propõe uma abordagem inovadora ao investigar a aplicação de cepas de leveduras autóctones, isoladas de fermentações artesanais de cachaça, na elaboração de hidromel. Diferentemente dos processos tradicionais que fazem uso de leveduras comerciais do gênero *Saccharomyces cerevisia,,* este estudo analisa tanto o desempenho fermentativo quanto a composição química de hidroméis obtidos com essas leveduras nativas, evidenciando seu potencial para elevar a qualidade sensorial e incrementar as propriedades bioativas da bebida. As leveduras autóctones podem proporcionar perfis aromáticos diferenciados, favorecer uma melhor produção de metabólitos e ampliar a capacidade antioxidante do produto final. Os achados desta pesquisa podem contribuir para expandir as possibilidades de uso de leveduras nativas na indústria de bebidas, além de valorizar o potencial biotecnológico de microrganismos provenientes de fermentações tradicionais.

2. OBJETIVOS

2.1 Objetivo geral

- Avaliar o potencial de cepas de leveduras autóctones isoladas de fermentações de cachaça na melhoria da qualidade sensorial e das propriedades bioativas do hidromel elaborado com mel do bioma caatinga.

2.2 Objetivos específicos

- Determinar as características físico-químicas, voláteis, capacidade antioxidante, fenólicos totais, perfil de fenólicos, perfil de ácidos orgânicos e perfil de açúcares do mel utilizado como matéria-prima;
- Analisar e comparar a viabilidade celular das leveduras autóctones dos engenhos de cachaça e das leveduras comerciais, verificando sua adequação para a fermentação do hidromel;
- Obter hidromel utilizando leveduras autóctones de engenhos de cachaça;
- Realizar caracterização físico-química, volátil, capacidade antioxidante, fenólicos totais, perfil de fenólicos, perfil de ácidos orgânicos e perfil de açúcares dos hidroméis obtidos.

3. REVISÃO DE LITERATURA

3.1 Fermentação alcoólica: aspectos gerais

A levedura *Saccharomyces cerevisiae* é o microrganismo mais utilizado para produção de etanol. Através do processo de fermentação alcoólica ela metaboliza açúcares (hexoses) e os convertem em dióxido de carbono e etanol. A fermentação pode ser conduzida de três modos: descontínuo, descontínuo-alimentado e contínuo (BAI *et al.,* 2008; AZHAR *et al.,* 2017). No processo descontínuo a adição do inóculo e substrato ocorre apenas no início (HADIYANTO *et al.,* 2013). A fermentação acontece em sistema fechado com alta concentração de açúcares e inibidores no início e termina com alta concentração de produto (THATOI *et al.,* 2014). Os benefícios incluem a esterilização completa, não necessidade de mão-de-obra especializada e fácil controle das matérias-primas (IVANOVA *et al.,* 2011; JAIN & CHAURASIA, 2014). As desvantagens são: menor nível de automação do processo (sendo necessário maiores custos em mão-de-obra), baixo rendimento e possibilidade de inibição do crescimento celular devido à presença de altas concentrações de açúcares (CHENG *et al.,* 2009).

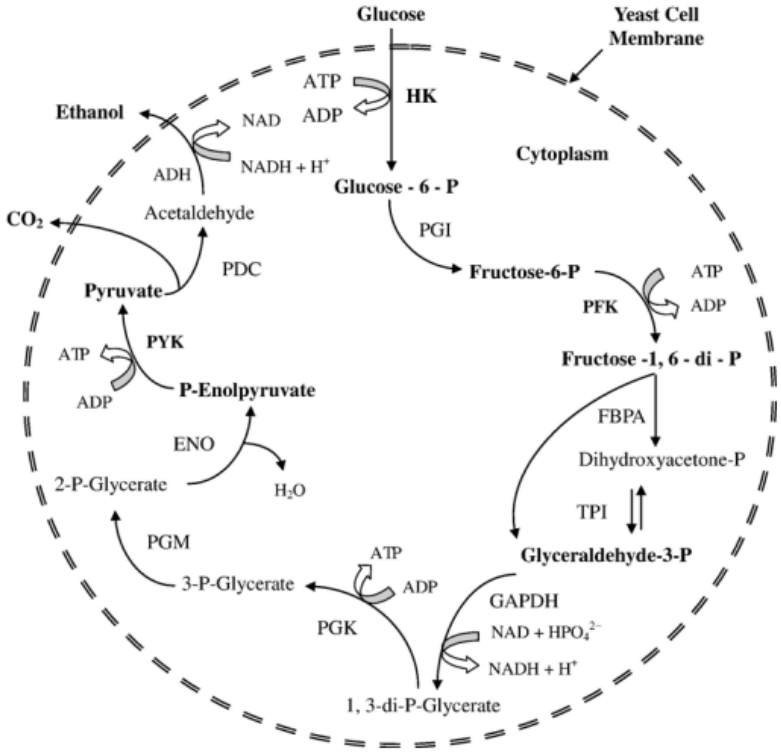
Na fermentação descontínua-alimentada o substrato é adicionado de modo gradual ou contínuo na dorna. Desse modo, a concentração de substrato se mantém controlada e permite a conversão de uma quantidade suficiente de açúcares fermentáveis em etanol (JAIN & CHAURASIA, 2014). As vantagens incluem a maior quantidade de oxigênio dissolvido no meio, maior produtividade, redução de inibição por compostos tóxicos (CHENG *et al.,* 2009). Entretanto, a produtividade é limitada pela taxa de alimentação e crescimento celular (MARGARITIS, 1987). No processo contínuo o substrato, meio de cultura e nutrientes são adicionados continuamente no biorreator contendo células ativas. O volume da dorna é mantido constante através da retirada dos produtos da fermentação (IVANOVA *et al.,* 2011). As vantagens são maior produtividade, redução do volume do biorreator e custos operacionais (JAIN & CHAURASIA, 2014). A desvantagens estão atreladas ao aumento da possibilidade de contaminação, fator que influencia na viabilidade celular e produtividade (CHANDEL *et al.,* 2007).

3.2 Bioquímica da fermentação alcoólica

A principal via metabólica do processo de produção do etanol é a glicólise, através dela uma molécula de glicose é metabolizada e duas moléculas de piruvato são produzidas (MADIGAN *et al*., 2000). Em condições anaeróbicas o piruvato é reduzido a etanol com

liberação de CO_2, conforme ilustrado na **Figura 1**. Dois ATPS produzidos na glicólise são usados para a biossíntese celular das leveduras. Sem o consumo contínuo do ATP voltado para o crescimento celular das leveduras a glicólise será interrompida devido ao acúmulo de ATP, que irá inibir a fosfofrutoquinase (PFK), uma das principais enzimas reguladoras da glicólise (BAI *et al*., 2008).

Figura 1 – Via metabólica da fermentação do etanol em *S.cerevisiae*



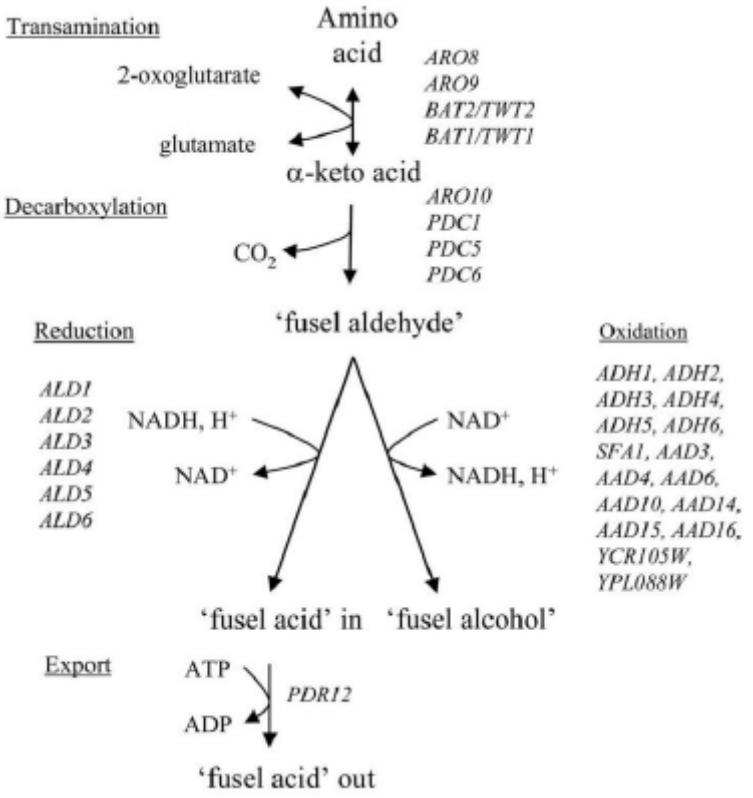
Fonte: *BAI et al., 2008*.

Figura 2 – Via de Ehrlich: Formação de álcoois superiores.

Além do etanol e do CO_2, outros subprodutos também são formados durante a fermentação alcoólica, sendo o glicerol o mais expressivo na maioria das vezes. Outros subprodutos, como os álcoois superiores — formados através da via de Ehrlich, como mostra a **Figura 2** — são produzidos em níveis mais baixos. Os álcoois superiores podem ainda reagir com o acetil-CoA através da ação das álcool acetiltransferases e formar seus ésteres correspondentes (ZHANG *et al.*, 2012). A produção desses compostos, bem como o crescimento das leveduras, acaba diminuindo o rendimento de etanol devido ao direcionamento de compostos intermediários glicolíticos para as vias metabólicas correspondentes (BAI et al., 2008; INGLEDEW, 1999).

Os subprodutos formados influenciam o sensorial das bebidas alcoólicas. A concentração desses compostos depende da composição química do mosto (BILVERSTONE

et al., 2015; WIETSTOCK *et al.*, 2015). Segundo Ribeiro-Filho *et al.* (2021) mostos com amônia-nitrogênio, fosfato inorgânico, potássio, magnésio, ferro, manganês ou mistura composta de todos os nutrientes resultou em aumento da concentração de álcoois superiores para NCYC2592, a mesma cepa aumentou a formação de ésteres de ácidos graxos quando houve suplementação com amônia-nitrogênio, fosfato inorgânico, potássio, magnésio, cobre, zinco, ferro ou manganês.



Fonte: HAZELWOOD *et al*., 2008.

3.3 Tratamento e reutilização de leveduras

O processo de fermentação alcoólica industrial do Brasil apresenta como um dos diferenciais o reciclo de mais de 90% das leveduras utilizadas (BASSO *et al*., 2008; WHEALS *et al.*, 1999). Além de diminuir impactos ambientais, esse processo também otimiza a fermentação devido ao fato do inóculo ser um creme rico em células e pobre em açúcares, fazendo com que praticamente não exista fase lag, e sim, fermentação principal desde o início da adição do mosto (LIMA *et al*., 2001). Ao final da fermentação o vinho levedurado (mosto fermentado) é enviado para uma dorna pulmão que alimenta a centrífuga que separa o vinho levedurado em duas partes. A primeira é o vinho (mistura de água, etanol, e produtos secundários) que é acondicionado na dorna volante para posterior utilização na destilaria e a segunda é um creme rico em células. O creme rico em leveduras é encaminhado

para tanques onde será realizada uma diluição com água e tratamento ácido, feito isso, poderá ser utilizado para um novo ciclo fermentativo (BNDES & CGEE, 2008; AMORIM *et al.*, 2011; VASCONCELO, 2012).

A suspensão rica em células não contém apenas leveduras, sendo assim, outros microrganismos são reciclados (SEO *et al*., 2020). A contaminação bacteriana é um dos principais problemas enfrentados pela indústria de bebidas. Segundo Lucena *et al*. (2010), bactérias ácido láticas, principalmente gênero *Lactobacillus*, são os contaminantes mais frequentes no processo de fermentação alcoólica utilizando cana-de-açúcar como substrato. Esses contaminantes competem com as leveduras pelos mesmos substratos, ocasionando a redução da viabilidade celular das leveduras, floculação do fermento, menor rendimento alcoólico do vinho e liberação de metabólitos tóxicos à levedura (principalmente ácido lático e acético), consequentemente, afetam o desempenho do processo fermentativo e a qualidade do produto final (SKINNER & LEATHERS, 2004).

Segundo Munford *et al.* (2020), o tratamento ácido antes da reutilização do fermento pode diminuir até quatro fases logs do crescimento de bactérias contaminantes. Na maioria das indústrias de bebidas alcoólicas, o pH do mosto é regulado até aproximadamente 2,5 utilizando ácido sulfúrico (podendo ser incubado por um período de 1 a 3 horas), todavia, alguns estudos indicam que o uso de tal substância pode afetar a viabilidade celular da *Saccharomyces cerevisiae* (CECCATO-ANTONINI, 2018; BASSI *et al*., 2013; DELLA-BIANCA *et al*., 2014). Bassi *et al.* (2013) verificaram o efeito do tratamento com ácido sulfúrico sob duas espécies de leveduras *Dekkera bruxellensis* e *Saccharomyces cerevisiae.* A primeira espécie apresentou um crescimento semelhante mesmo em condições de pH 2,0 por 2 horas, todavia, a *Saccharomyces cerevisiae* apresentou uma queda da viabilidade celular.

Levando em consideração que o tratamento ácido pode afetar a viabilidade celular das leveduras, a utilização de água para a recuperação do fermento (rinsagem) pode ser vista como uma alternativa simples e de baixo custo. Esta prática visa aumentar a viabilidade da população de leveduras, eliminando as células mortas e reduzindo os resíduos de fermentação. Além disso, a lavagem com água ajuda a minimizar a concentração de álcool remanescente no creme de leveduras, promovendo um ambiente mais favorável para as leveduras em reutilizações subsequentes (WHITE & ZANAISHEFF, 2010).

3.4 Hidromel: Classificação, legislação e produção

O mel pode ser utilizado na produção de diversas bebidas alcoólicas, como sidras "duras", uísques aromatizados, cachaças, cervejas e Bourbon (STAROWICZ &

GRANVOGL, 2020). Durante o processo produtivo, ele pode ser adicionado tanto na etapa de fervura quanto na fermentação (STAROWICZ & GRANVOGL, 2022), conferindo à bebida um sabor doce, suave e aveludado, além de um agradável aroma floral. Além disso, o uso do mel aumenta o teor alcoólico das bebidas, devido ao aumento de açúcares disponíveis para a fermentação (STAROWICZ & GRANVOGL, 2020).

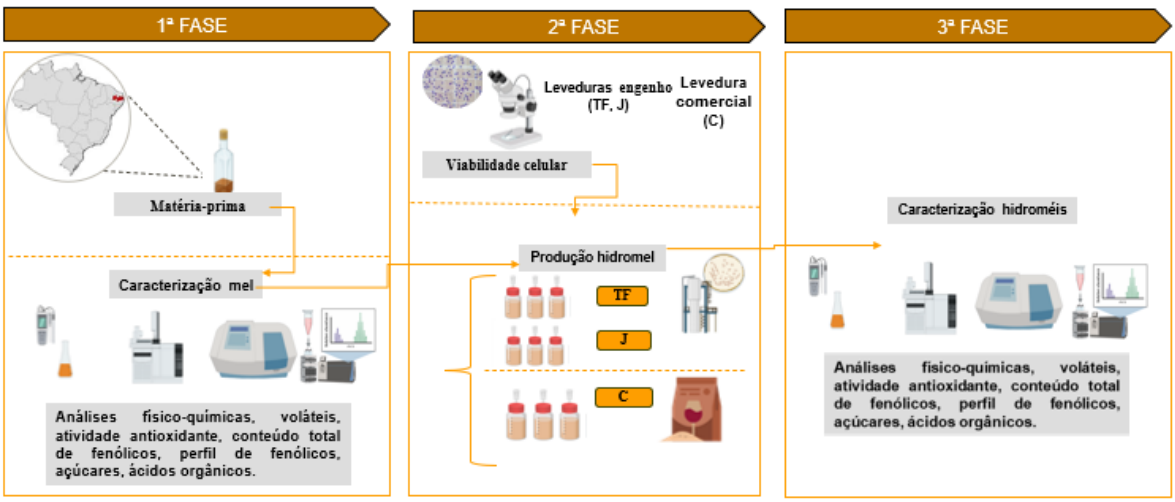
De acordo com o decreto n° 6871, o teor alcoólico do hidromel deve ser entre 4% e 14% (BRASIL, 2009). A legislação brasileira também proíbe a adição de açúcar ou ingredientes como frutas, ervas e especiarias, e classifica o hidromel como seco, se contiver até 3g/L de açúcar residual, ou suave, se exceder esse limite (BRASIL, 2012). Em muitos países, no entanto, é comum adicionar frutas, ervas e especiarias ao hidromel, resultando em classificações como pyment, feito com uvas; *cyser*, feito com maçãs; melomel, feito com frutas; e *metheglin*, feito com especiarias (MCCONNELL; SCHRAMM, 1995).

A primeira etapa do processo produtivo do hidromel é preparar o mosto, diluindo o mel em água em proporções de 1:0,5 a 1:3 (mel:água) (STAROWICZ & GRANVOGL, 2020). O mosto pode ser suplementado com sais inorgânicos (GUPTA & SHARMA, 2009) e o pH ajustado para 3-5 com ácidos orgânicos como o ácido tartárico (BRASIL, 2022). Após a preparação, a fervura opcional dura 2-4 horas. Em seguida, a levedura, geralmente *Saccharomyces cerevisiae*, é inoculada para fermentação (GUPTA & SHARMA, 2009). Após a fermentação, a bebida é engarrafada e pode ser maturada por 9 meses a 2 anos (STAROWICZ & GRANVOGL, 2020).

4. DELINEAMENTO EXPERIMENTAL

Conforme apresentado na **Figura 3,** o experimento foi dividido em três etapas. Na primeira fase, foi feita a seleção da matéria-prima na região de São Mamede (PB) e a caracterização do mel por meio de análises físico-químicas (pH, umidade, cinzas, acidez, sólidos solúveis totais, atividade de água, açúcares redutores e açúcares redutores totais), voláteis, atividade antioxidante conteúdo, total de fenólicos, perfil de fenólicos, açúcares e ácidos orgânicos. Na segunda fase, a viabilidade celular das leveduras de engenho e comerciais foi avaliada como uma pré-etapa antes de iniciar a produção do hidromel, sendo inoculadas cerca de $1{,}5x10^7$ células/mL. Na terceira fase, o hidromel produzido foi submetido a análises físico-químicas (pH, acidez, sólidos solúveis totais, teor alcoólico, açúcares redutores e açúcares redutores totais), análise de voláteis, testes de atividade antioxidante, conteúdo total de fenólicos, perfil de fenólicos, açúcares e ácidos orgânicos utilizando cromatografia líquida. As análises físico-químicas, análise de compostos voláteis, testes de antioxidante e conteúdo total de fenólicos, foram realizadas no Laboratório de Análises Químicas de Alimentos (LAQA/CT), em João Pessoa-PB. Para a determinação, açúcares, ácidos orgânicos e perfil de fenólicos foi utilizado Cromatografia Líquida de Alta Eficiência – CLAE, utilizou-se a estrutura de um laboratório parceiro do IFPE – Campus Petrolina. As demais fases foram conduzidas no Laboratório de Tecnologia de Produtos Agropecuários (LTPA/CCA), em Areia-PB.

Figura 3 – Delineamento experimental.



Fonte: Autoria própria, 2024.

5. RESULTADOS

Após o final da fermentação do hidromel, todas as cepas de leveduras avaliadas apresentaram 100% de viabilidade celular, demonstrando alta resistência aos estresses típicos de fermentações prolongadas. Na análise da produção de CO_2, a cepa TF foi a que apresentou melhor desempenho, a cepa J mostrou uma produção mais lenta e constante, sem estabilização até o final do período, enquanto a cepa C teve o pior desempenho e apresentou grande variação entre réplicas, sugerindo baixa adaptação ao meio de fermentação do hidromel. A análise físico-química do mel utilizado como matéria-prima para produção do hidromel revelou que todos os parâmetros estavam dentro dos limites estabelecidos pelas normas brasileiras e internacionais, confirmando a qualidade do mel e sua adequação para a produção da bebida.

As diferentes leveduras utilizadas na fermentação do hidromel influenciaram significativamente nas características do produto final. A levedura autóctone TF resultou em um hidromel com maior teor alcoólico (4,3%) e menor teor de açúcares residuais, indicando fermentação mais eficiente. Já a levedura comercial (C) gerou um hidromel com menor teor alcoólico (1,6%), grande quantidade de açúcares residuais e elevado e extrato seco, enquanto a levedura J apresentou desempenho intermediário em termos de teor alcoólico, mas também apresentou elevada concentração de açúcares residuais.

Em relação aos parâmetros cinéticos, as três leveduras apresentaram comportamentos distintos na fermentação. A cepa TF teve o melhor desempenho, com crescimento mais rápido, maior produção de etanol, CO_2 e biomassa, além dos maiores rendimentos e produtividades. A levedura comercial (C) mostrou baixo desempenho fermentativo e menor conversão de açúcar em etanol, apesar do maior valor de $YCO_2/EtOH$, sugerindo maior desvio de carbono para respiração. A cepa J teve desempenho intermediário. Esses resultados reforçam o vigor fermentativo da TF.

Quanto ao perfil volátil, foram identificados 128 compostos distribuídos em 12 classes químicas. Os hidroméis mostraram predominância variável de compostos voláteis em termos de concentração, maioria de ésteres no C, álcoois no TF e terpenos no J. Foram identificados 19 compostos-chave nas 3 formulações, sendo o álcool isoamílico e seu éster (acetato de isoamila) os de maior relevância em termos de impacto sensorial para as três formulações, contribuindo com aromas fermentados e frutados, respectivamente.

A análise de fenólicos totais e atividade antioxidante revelou diferenças significativas entre as formulações. A levedura autóctone TF apresentou o maior teor de compostos fenólicos totais. A capacidade antioxidante medida por ABTS e DPPH não apresentou

diferenças significativas entre os hidroméis, enquanto o teste FRAP evidenciou superioridade da levedura comercial. No perfil de compostos fenólicos, ácidos orgânicos e açúcares, os hidroméis J e C apresentaram maior teor de naringina, enquanto a cepa TF destacou-se pela presença exclusiva de hesperidina; o ácido cítrico foi mais elevado na formulação C e o acético predominou em TF e J. Em relação aos açúcares, TF consumiu praticamente toda a frutose, enquanto J e C mantiveram altos valores residuais.

5.1 Artigo 1

Mead produced using Autochthonous Yeasts selected from Cachaça productions

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ABSTRACT

This study evaluated the fermentation performance and chemical composition of mead produced using indigenous yeast strains isolated from cachaça fermentations, highlighting their potential to improve sensory quality and bioactive properties. Meads were produced with honey from Apis mellifera collected in the semiarid region of Paraíba, using three yeast strains: two indigenous (TF and J) and one commercial strain (C). Physicochemical analysis of the honey confirmed its suitability for fermentation, with all parameters within regulatory standards. The indigenous yeasts adapted well to the fermentation environment. Strain TF stood out with greater CO_2 production and more efficient sugar-to-ethanol conversion, resulting in a mead with 4.3% alcohol and lower residual sweetness. In contrast, the commercial strain showed weak fermentation performance, producing only 1.6% alcohol. Nevertheless, all strains maintained 100% cell viability at the end of fermentation, indicating resistance to stress conditions. Regarding kinetic parameters, the three yeasts exhibited distinct behaviors. TF showed the best performance, with faster growth, higher sugar consumption, and increased ethanol and CO_2 production—outperforming the others in most evaluated indicators. The commercial strain had the lowest efficiency, with limited biomass and ethanol production, while strain J demonstrated intermediate performance. A total of 128 volatile compounds, distributed across 12 chemical classes, were identified in the meads. Each yeast strain influenced the volatile profile differently: esters dominated in C, alcohols in TF, and terpenes in J. Isoamyl alcohol and isoamyl acetate were prominent across all samples (OAV $\geq$ 1), contributing to fruity and fermented notes, but their even distribution limited their use as differentiating markers. Principal Component Analysis (PCA) of key volatiles revealed differentiation among the meads, driven by compounds such as ethyl caprylate and β-cyclocitral. Regarding phenolics, TF had the highest total content. Antioxidant activity measured by ABTS and DPPH was similar across meads, but the FRAP method showed higher values for the commercial strain. Sugar and acid profiles also varied: TF concentrated hesperidin and nearly depleted fructose, while J and C accumulated naringin and residual sugars. Citric acid content was highest in the C formulation, whereas acetic acid levels were greater in TF and J. These findings underscore the value of indigenous yeasts in crafting meads with unique sensory profiles and enhanced bioactive potential, promoting the use of local microbial resources and fostering innovation in the fermented beverage sector.

Keywords: Alcoholic fermentation; Autochthonous yeast strains; volatile compound profile; Semi-arid *Apis mellifera* honey; Odor Activity Value (OAV).

1. Introduction

Honey is a natural product of significant nutritional and therapeutic value, with a history of use dating back thousands of years. Produced by bees of the *Apis* genus from floral nectar, honey is primarily composed of simple sugars such as glucose and fructose. However, its complexity extends further, as it also contains smaller amounts of proteins, organic acids, minerals, vitamins, enzymes, phenolic compounds, and volatile substances that contribute to its functional properties (Da Silva et al., 2016). The chemical composition of honey is directly influenced by the flora from which the nectar is collected, resulting in noticeable sensory differences among honeys of various botanical origins. In particular, volatile compounds are responsible for the aromatic profile and may serve as useful chemical indicators for determining floral origin (Castell et al., 2023; Zhu et al., 2022).

In the realm of fermented beverages, mead — produced by fermenting a solution of honey and water — stands out as one of the oldest forms of alcoholic beverage, with records dating back to the Neolithic period, around 7000 B.C. The diversity of available honeys directly affects the sensory profile of the final product (Deng et al., 2023). The use of multifloral honey for mead production promotes the formation of compounds such as trans-nerolidol and ethyl phenylacetate, which contribute floral notes to the beverage (Chitarrini et al., 2020). In contrast, honeydew honey tends to generate more intense flavor profiles due to the presence of fatty acids like caprylic and capric acid, which are associated with harsher or rancid notes.

Another critical factor in mead production is the yeast strain selected for fermentation. The performance of the yeast can be hindered by adverse conditions such as temperature fluctuations, nutrient deficiencies, osmotic stress, and high alcohol content. Under such conditions, poorly adapted strains may fail to complete fermentation efficiently and produce undesirable by-products — known as off-flavours — that compromise the sensory quality of the beverage (Bauer & Pretorius, 2000; Hohmann & Mager, 2003; Attfield, 1997; Bisson, 1999). One strategy with the potential to improve process efficiency is the reuse of yeast, a practice already widely adopted in other fermentation industries. This approach not only reduces production costs and environmental impact but also accelerates fermentation by shortening the yeast's lag phase. Moreover, with continued use, yeast strains may develop adaptive traits that enhance their resistance to process-specific conditions, such as pH variation, temperature shifts, and alcohol concentration (White & Zanaisheff, 2010).

This study introduces a novel approach by evaluating the use of autochthonous yeast strains, isolated from traditional cachaça fermentations, in mead production. Unlike conventional methods that rely on commercial *Saccharomyces cerevisiae*, this study evaluates the fermentation performance and chemical composition of mead produced using autochthonous yeast strains isolated from cachaça fermentations, highlighting their potential to enhance the sensory quality and bioactive properties of the beverage. These native yeasts may contribute to unique aromatic profiles, improved metabolite synthesis, and increased antioxidant activity. The results may broaden applications for native yeasts in the beverage industry and stimulate interest in the biotechnological potential of microorganisms from traditional fermentations.

2. Materials and methods

2.1 Collection of honey used as raw material for the production of mead

The honey used in mead production was collected in the municipality of São Mamede, located in the semiarid region of Paraíba, Brazil, within the Caatinga biome. The samples underwent sequential processing steps, including uncapping, to remove the wax layer that seals the honeycomb; centrifugation, to extract the honey using centrifugal force; filtration, to remove larger particles; and decantation, to separate finer particles through sedimentation. After processing, the honey was stored in sterile glass bottles until use.

2.2 Physicochemical characterization of honey used as raw material and mead

The honey used in this study was previously characterized to determine physicochemical parameters, total phenolics, antioxidant activity, and volatile compound profile (Table 1). Physicochemical characterization included analyses of pH, HMF, moisture, ash, total acidity, total soluble solids, and water activity for honey, while for mead, analyses of pH, total acidity, total soluble solids, alcohol content, and dry extract were performed, following the methodology described by the AOAC (2016). The sugar content (reducing sugars, total sugars, and sucrose) was determined by the DNS method, according to Miller (1959).

2.3 Yeast propagation

TF and J strains were propagated in YPD medium (1% yeast extract, 2% peptone, and 2% glucose) and incubated for 48 h at 30 °C.

Table 1: Physicochemical parameters, total phenolics, antioxidant activity and key aroma compounds of *Apis mellifera* honey from the Caatinga biome, used as the raw material for mead production

Parameters		Unit	Threshold	Concentration
Physicochemical	Moisture	%		17.0 ± 0.2
	Ashes	%		0.04 ± 0.02
	HMF	mg/kg		2.5 ± 0.8
	Total sugars	g/100 mL		84 ± 0
	Reducing sugars	g/100 mL		83 ± 0
	Acidity (mEq/kg)	mg/L		32.5 ± 0.9
	pH			4.58 ± 0.14
	a_w			0.6 ± 0.0
	Sucrose	g/100 mL		1.1 ± 0.1
Bioactivity	ABTS	µmol TE/100 g		163.5 ± 7.4
	FRAP	µmol TE/100 g		244.1 ± 21.7
	DPPH	µmol TE/100 g		213.54 ± 3.72
	Total Phenolics	mg Ac.Gál./100 g)		164.72 ± 3.10
Key aroma compounds	Anisole, p-methyl-	mg/L	0.025	0.03 ± 0.01
	Butanal, 3-methyl-		0.0004	0.003 ± 0.001
	β-Damascenone		0.000002	0.12 ± 0.01
	Methyleugenol		0.006	0.34 ± 0.02
	Undecanal		0.0125	0.03 ± 0.01
	Decanal		0.003	0.7 ± 0.1
	Benzeneacetaldehyde		0.0063	0.7 ± 0.1
	Octanal		0.0006	0.07 ± 0.02
	β-Cyclocitral		0.003	0.34 ± 0.02
	cis-Linalool oxide		0.1	5.8 ± 0.3
	Linalool		0.0002	0.5 ± 0.1

Data is expressed as mean ± standard deviation of analyzes performed in triplicate. HMF = hydroxymethylfurfural; a_w = water activity; TE = Trolox Equivalent.

2.4 Yeast collection

The indigenous yeasts (TF and J) used in this study were part of a yeast collection maintained at the Laboratory of Technology of Agroindustrial Products (DSER/UFPB), consisting of isolates obtained from different cachaça distilleries in the Brejo region of Paraíba, Brazil. These colonies were previously isolated, purified, and stored in an ultra-freezer at −80 °C. Before use, the strains underwent phenotypic characterization, including growth — considering their growth potential in spot plates on solidified agar culture medium — at different temperatures (4, 15, 20, 25, 30, 37 and 40 °C), utilization of various carbon sources such as glucose, fructose, galactose, xylose, mannose, rhamnose, sucrose, maltose,

lactose and melibiose, and nitrogen sources such as potassium nitrate, sodium nitrite and lysine, tolerance to inhibitory compounds including ethanol at 10, 15 and 20%, acetic acid at 2, 6 and 10%, and production of hydrogen sulfide (H2S), allowing us to assess their metabolic and adaptive potential.

2.5 Mead production and carbon dioxide loss

The mead was produced using three yeast strains including a commercial yeast (C) and two indigenous yeasts (TF, J) selected from cachaça productions, which were washed and centrifuged before reuse. Fermentation occurred in 250 mL minireactors equipped with airlocks, with an inoculation of 1.5×10^7 cells/mL in must composed of honey diluted in water with a concentration of 10 °Brix. The minireactors were weighed periodically until the end of fermentation in order to construct a carbon dioxide loss curve. Fermentations were conducted at 28 °C. Prior to inoculation, TF and J yeasts, propagated in YPD medium, were centrifuged and rinsed with water.

2.6 Cell viability

The cell viability (%) of all yeasts was evaluated at the beginning and after the end of fermentation using the Neubauer chamber and methylene blue dye (0.01%). The methylene blue solution was prepared by dissolving 0.01 g of methylene blue in 10 mL of water, then adding 2 g of sodium citrate, stirring until completely diluted. Finally, the volume of the solution was adjusted to 100 mL using distilled water (Pierce, 1970). The yeast suspension (0.5 mL) was homogenized with methylene blue solution (0.5 mL), incubated at 28°C for 5 minutes, and analyzed under a microscope. Cell viability was assessed by distinguishing between viable and non-viable cells, with the results presented as a percentage (Da Silva et al., 2023).

2.7 Kinetic parameters

Fermentation kinetics were analyzed using the specific cell growth rate (μ), substrate consumption rate (q_s), and metabolite production rate (q_p). Conversion coefficients were also considered, including yields of substrate to biomass ($Y_{X/S}$), substrate to product ($Y_{P/S}$), and product to biomass ($Y_{P/X}$). Furthermore, biomass (P_X) and product (P_P) productivities were determined. All these parameters were calculated based on the equations presented in numbers 1 to 12, as described in the supplementary material (List of equations).

2.8 Analysis of volatile compounds

The volatile compounds were extracted by HS-SPME. The fiber used was PDMS 100 µm (Gray). For honey, 10 g, 10 mL of Milli-Q water and 3 µL of internal standard (1,2 dichlorobenzene) were added in a 100 mL glass vial with a septate and screwed cap. For mead, 5 ml of mead with NaCl added until saturated and 3 µL of internal standard (1,2-dichlorobenzene) were used. The extraction will be performed in a water bath at 45 ºC. Initially, the fiber will be in equilibrium for 15 minutes and then will be exposed for 45 minutes. The volatile compounds extracted by HS-SPME will be analyzed in a gas chromatograph coupled to a mass spectral detector. The linear retention indices (RI) of the chromatographic peaks will be determined experimentally using the retention time of a series of homologous n-alkanes (C8-C20). The identification of the volatile compounds present in honey and mead will be performed by comparing the obtained spectra with those available in the National Institute of Standards and Technology (NIST) GC-MS library (version 2.0, 2008) and by verifying the linear retention indices with data from the scientific literature for columns of similar polarity.

2.9 Antioxidant activity

Antioxidant activity was evaluated using the DPPH● (2,2-diphenyl-1-picrylhydrazyl), ABTS●+ (2,2-azino-bis(3-ethylbenzothiazoline)-6-sulfonic acid), and FRAP (ferric reducing antioxidant power) methods, with measurements performed on a UV–vis spectrophotometer. For the DPPH method, the procedure described by Rufino et al. (2007) was followed, in which 0.1 mL of the sample was mixed with 2 mL of the DPPH solution, and absorbance was read at 515 nm after 30 minutes in the dark. The ABTS method was performed according to Re et al. (1999), by adding 30 µL of the sample to 3 mL of the ABTS●+ solution, with absorbance measured at 734 nm after 6 minutes of reaction. The FRAP assay was adapted from Benzie and Strain (1996), involving the reaction of 0.1 mL of the sample with 3 mL of the FRAP reagent, prepared with acetate buffer, TPTZ (2,4,6-tris(2-pyridyl)-s-triazine) solution, and ferric chloride at pH 3.6, incubated at 37 °C for 30 minutes, with absorbance read at 593 nm. Results were expressed in µmol Trolox equivalents (TE) per 100 mL for mead and per 100 g for honey.

2.10 Total phenolic compouns

Total phenolic compounds were determined by UV-vis spectrophotometric analysis. The procedure followed the methodology proposed by Biluca *et al.* (2017), with the addition

of 50 µL of the sample to 250 µL of Folin-Ciocalteu reagent, followed by the incorporation of 750 µL of 20% sodium carbonate solution. After a two-hour incubation period, absorbance was measured at 765 nm. The results were expressed as milligrams of gallic acid equivalent per 100 mL of mead and per 100 g of honey.

2.11 Profile of phenolic compounds

The extraction of phenolic compounds from honey followed the method described by Biluca et al. (2017), with some modifications. The sample was first diluted in a 2% NaCl solution (1:1) and extracted with ethyl acetate. It was then dehydrated with sodium sulfate for 15 minutes, filtered, and concentrated on a rotary evaporator at 40 °C. The dry extract was dissolved in 1 mL of methanol, microfiltered (0.20 µm), and subsequently diluted in 30% methanol. For the mead samples, only filtration with a 0.20 µm microfilter was performed. Chromatographic analysis was performed according to the protocol of Lima et al. (2024). A volume of 10 µL of each extract was injected into an HPLC-DAD system equipped with an Eclipse Plus RRHT RP-C18 ultra-high-performance column (50 × 4.6 mm, 1.8 µm; Zorbax, SC, USA). The mobile phase consisted of 0.5% phosphoric acid (solvent A) and 0.5% acidified methanol (solvent B), maintained at a flow rate of 1 mL/min and a temperature of 40 °C. The elution gradient used was: 0 min (0% B), 2 min (10% B), 13 min (26% B), 19 min (50% B), 21 min (80% B), 21.1–23.1 min (100% B), returning to 0% B in 23.2 min and maintained for an additional 3 min. Detection was performed at wavelengths of 220, 280, 320, 360, and 520 nm. Compound identification and quantification were performed by comparison with external standards, using calibration curves, retention times, and spectral similarity. Data processing was performed using OpenLAB CDS ChemStation software. All calibration curves showed coefficients of determination greater than $R^2 > 0.997$, with limits of detection (LOD) lower than 0.31 g/L and limits of quantification (LOQ) lower than 0.88 g.

2.12 Profile of sugars and organic acids

The honey sample, previously diluted at a ratio of 1 g to 9 mL of ultrapure water, and the mead samples were filtered through 0.45 µm microfilters and analyzed by HPLC for the simultaneous quantification of sugars and organic acids as described by Coelho et al. (2018). For each analysis, 10 µL of the samples were injected into an Agilent 1260 Infinity LC HPLC system (Agilent Technologies, Santa Clara, CA, USA), equipped with an Agilent Hi-Plex H column (300 × 7.7 mm, 8.0 µm). The mobile phase consisted of a 4.0 mM/L H_2SO_4 solution, maintained at a flow rate of 0.7 mL/min and at 70 °C. Organic acids were detected using the

DAD detector (model G1315D, 210 nm), while sugars were identified using the RID detector (model G1362A, 50 °C). Data processing was performed using the OpenLAB CDS ChemStation Edition™ software, using retention time and comparison with authentic standards as criteria for compound identification.

2.13 Statistical analysis

Statistical analysis of the data was performed using analysis of variance (ANOVA), followed by Tukey's multiple comparison test to evaluate relevant differences between treatments, with a significance level of 5% ($p < 0.05$). These analyses were performed using Microsoft Excel, complemented by XLSTAT®. Principal Component Analysis (PCA) was performed to explore patterns and relationships between variables, using Python in Google Colab®.

3 Results and Discussion

3.1 Cell viability

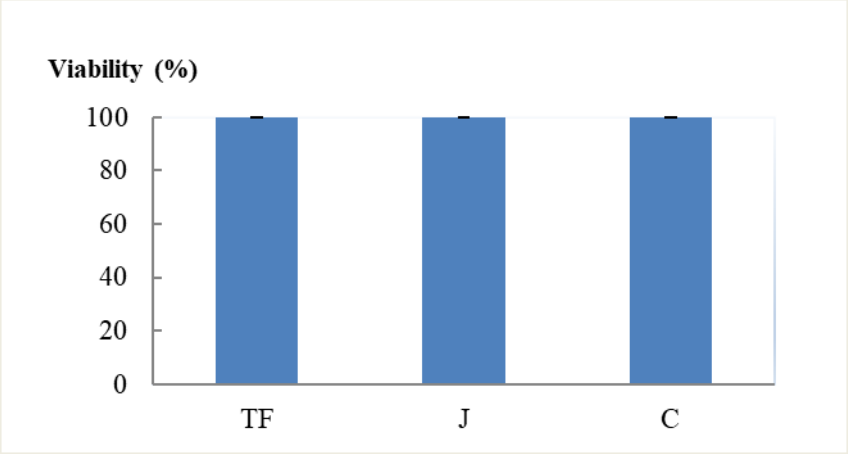

Figure 1: Cell Viability of Different Yeast Strains During Mead Fermentation After 360 Hours.

Monitoring cell viability during fermentation allows us to understand how strains respond to prolonged stress. After 360 hours of fermentation, it was observed that all experimentally evaluated strains—TF and J (autochthonous, isolated from cachaça distilleries) and C (commercial)—showed 100% viability (Figure 1), suggesting a high resistance to the stress typically associated with long-term fermentations. This finding indicates that the

autochthonous yeasts not only tolerate ethanol accumulation and nutrient limitation, but also exhibit performance comparable to the widely used commercial strain in fermentation processes. Different strains exhibit significant variations in stress resistance, with some able to maintain plasma membrane integrity and physiological intracellular pH even after exposure to organic acids and ethanol at pH 3.4, demonstrating effective adaptive mechanisms under adverse conditions (Houngbédji et al., 2019). Therefore, the data from this analysis reinforce the biotechnological potential of the autochthonous TF and J strains as viable candidates for application in industrial fermentations, such as mead production.

3.2 CO_2 Loss

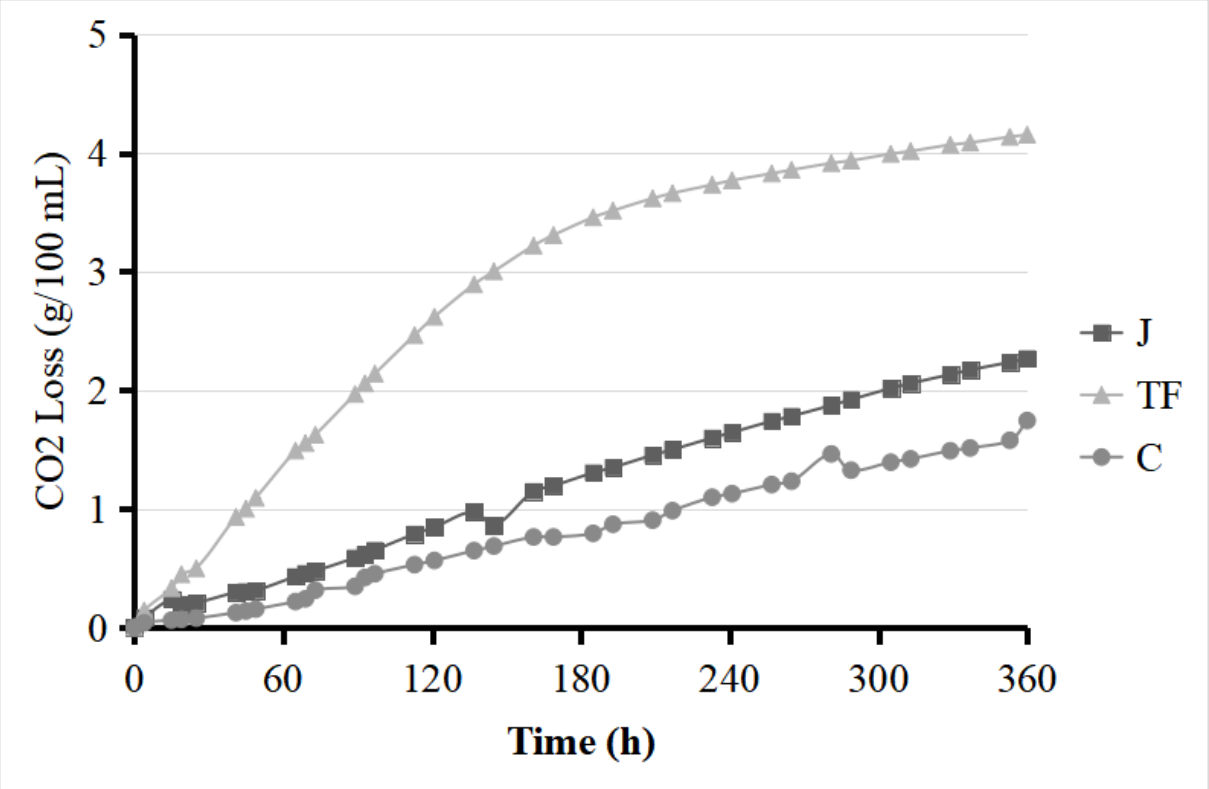

Figure 2*:* CO_2 Loss During Mead Fermentation Using Different Yeast Strains (TF, J, and C).

The carbon dioxide (CO_2) is parameter, which indicates the fermentation performance. All yeast strains evaluated generated CO_2 during mead fermentation (Figure 2). The TF strain demonstrated the best performance, with rapid CO_2 generation during 250 h (Figura 2). Moreover, when J strain was inoculated into mead must, CO_2 generated was lower than the quantity generated by TF strain; however, it was higher than the amount generate by C strain (Figure. 2). Then, the commercial strain (C) showed the lowest generation of CO_2 (Figura 2). Therefore, autochthonous yeasts isolated from cachaça productions exhibit a good adaptability to the mead must, resulting in a fermentation efficiency compared to commercial

*Saccharomyces cerevisia*e strain. The observed performances reinforce the suitability of autochthonous strains for maximizing mead production at an industrial scale, contributing to greater efficiency, versatility, and sustainability in fermentation processes (Grellet et al., 2022).

3.3 Physicochemical analysis of mead

Table 2: Physicochemical parameters of meads fermented with different yeast strains (C, TF, J).

Yeast	Total acidity (meq/L)	Total Soluble Solids (°Brix)	ABV (%)	Total sugars (%)	Reducing Sugars (%)	Sucrose (%)	Dry Extract (g/L)	pH
TF	$31.4^{b} \pm 0.0$	$3^{c} \pm 0$	$4.3^{a} \pm 0.0$	$0.60^{c} \pm 0.01$	$0.50^{c} \pm 0.02$	$0.1^{a} \pm 0.01$	$14.3^{c} \pm 0.05$	$3.60^{a} \pm 0.04$
J	$55.9^{a} \pm 1.7$	$5^{b} \pm 0$	$2.3^{b} \pm 0.0$	$5.40^{b} \pm 0.06$	$5.2^{b} \pm 0.1$	$0.2^{a} \pm 0.15$	$54.6^{b} \pm 0.21$	$3.40^{b} \pm 0.02$
C	$23.2^{c} \pm 0.57$	$6^{a} \pm 0$	$1.6^{c} \pm 0.0$	$3.3^{a} \pm 0.1$	$3.2^{a} \pm 0.04$	$0.2^{a} \pm 0.1$	$75.5^{a} \pm 0.48$	$3.4^{b} \pm 0.0$

Note: Different lowercase letters in the same column indicate significant differences between means according to Tukey's test ($p < 0.05$).

The different yeast strains significantly influenced the soluble solids, total sugars, and reducing sugars contents of the meads produced (Table 2). The mead fermented with TF yeast had the lowest values of total sugars (0.60%), reducing sugars (0.5%), sucrose (0.1%), and total soluble solids (3 °Brix), as well as a lower dry extract, evidencing greater fermentation efficiency and a more complete conversion of sugars to ethanol. In contrast, the mead produced with C yeast had a higher dry extract (75.5 g/L) and a higher total soluble solids value (6 °Brix), indicating a less complete fermentation and a sweeter and fuller-bodied profile. Yeast J showed intermediate performance in terms of dry extract, but had the highest total (5.4%) and reducing sugar (5.2%) contents. Thus, yeast selection can be strategically used according to the desired mead profile: more efficient yeasts are recommended for dry meads, while strains that leave greater amounts of residual sugars favor sweeter, denser beverages. Furthermore, these attributes directly influence the sensory acceptance and market potential of fermented products (Sottil et al., 2019; Schwarz et al., 2020).

The variation between strains also influenced total acidity and pH, parameters that play a decisive role in both product stability and sensory balance. Total acidity varied considerably between samples, with mead produced with strain J having the highest value (55.9 meq/L), followed by TF (31.4 meq/L) and C (23.2 meq/L). Despite these differences in total acidity, pH values remained similar between samples, ranging from 3.40 to 3.60. Like

sugar content, total acidity and pH are strongly correlated with the perception of sour flavor in alcoholic beverages, reinforcing the need to control these variables to achieve the desired sensory profiles in meads (Senn, Cantu, & Heymann, 2021).

Alcohol content varied according to the yeast used, reflecting differences in fermentation efficiency. TF mead had the highest value (4.3%), followed by J mead (2.3%) and C mead (1.6%). Higher values indicate greater conversion of sugars to ethanol and a drier profile, while lower values are associated with a higher amount of residual sugars and sweeter, fuller-bodied beverages. These results demonstrate that yeast choice is crucial for balancing alcohol content and can be used strategically to direct the beverage toward a drier, more alcoholic profile or a sweeter, lower-alcohol profile (Sottil et al., 2019; Schwarz et al., 2020). Furthermore, ethanol content is directly correlated with the perception of alcoholic warmth, reinforcing its importance in defining the sensory experience (Senn, Cantu, & Heymann, 2021).

3.4 Kinetic parameters

Table 3: Kinetic parameters for meads produced with different yeasts (TF, J, C).

Yeast	μ	td	q_s	q_p	$Y_{p/s}$	$Y_{p/x}$	$Y_{x/s}$	$Y_{CO2/EtOH}$	$Y_{CO2/s}$	$Y_{CO2/x}$
TF	0.03	26.3	0.15	0.06	0.38	2.2	0.17	0.98	0.37	2.1
J	0.03	22.1	0.13	0.05	0.35	1.6	0.22	1.00	0.35	1.6
C	0.05	15.4	0.24	0.04	0.18	1.1	0.16	1.06	0.20	1.2

μ: specifc microbial growth rate (h^{-1}); td: cell doubling time (h); q_s: specifc substrate consumption rate (h^{-1}); q_p: specific ethanol production rate (h^{-1}); Y: yield coefcients; $Y_{p/s}$ (g of ethanol/g of substrate); $Y_{p/x}$ (g of ethanol/g of cells); $Y_{x/s}$ (g of cells/g of substrate); $Y_{CO_2/EtOH}$ (g of CO_2/g of ethanol); $Y_{CO_2/s}$ (g of CO_2/g of substrate), $Y_{CO_2/x}$ (g of CO_2/g of cells).

Table 4: Productivity for meads produced with different yeasts (TF, J, C).

Yeast	P_x	P_p	P_{CO2}
TF	0.08	0.17	0.17
J	0.04	0.06	0.06
C	0.05	0.04	0.05

P_x: cell productivity (g/h); P_p: ethanol productivity (g/h); P_{CO2}: CO_2 productivity (g/h).

The kinetic parameters indicated that the three yeasts evaluated exhibited distinct behaviors during fermentation (Table 3, Table 4). Strain C stood out for its faster growth (higher μ and lower td) and for its higher sugar consumption rate (q_s). Strain TF, on the other hand, maintained the highest specific rates of ethanol production (q_p), as well as the highest yields of product relative to substrate ($Y_{p/s}$) and biomass ($Y_{p/x}$), and of CO_2 relative to biomass formed ($Y_{CO_2/x}$) and substrate ($Y_{CO_2/s}$). These results were also reflected in higher productivity values for biomass (P_x), ethanol (P_p), and CO_2 (P_{CO_2}), confirming its fermentative vigor.

These results, however, differ from the findings of Park et al. (2020), who observed that strains with shorter duplication time (td) and higher growth rate (μ) tend to present higher levels of intracellular ATP, which translates into greater stability and fermentative efficiency. In the present study, although strain C exhibited faster growth, it was not the most efficient in terms of ethanol production or overall productivity. Yeast C showed a higher $Y_{CO_2/EtOH}$ value. The fact that strain C presented a higher $Y_{CO_2/EtOH}$ suggests that a more significant fraction of the substrate carbon was diverted to respiratory pathways, since the predominance of respiration in *Saccharomyces cerevisiae* results in greater CO_2 release and proportionally lower ethanol synthesis (Yerushalmi & Volesky, 1981).

3.5 Volatile profile of mead

The analysis of volatile compounds in mead allows us to evaluate how different yeast strains contribute to the development of its aromatic profile, as each chemical class is associated with specific sensory descriptors. A total of 128 volatile compounds belonging to 12 chemical classes were identified in mead produced with TF, J and C yeasts: Terpenes (33 compounds), Aldehydes (6), Alcohols (25), Aromatics (7), Ketones (4), Esters (32), Ethers (4), Norisoprenoids (3), Hydrocarbons (1), Nitrogen compounds (2), Acids (10) and furans. (1).

Most of the volatile compounds identified in the mead produced with yeast C belong to the ester class (Figure 3a), corresponding to 52.6% in terms of the total concentration of volatile compounds or 37 μg/mL. On the other hand, in the mead fermented with yeast TF, a predominance of alcohols was observed (Figure 3b), representing 55.6% of the total concentration of volatile compounds or 31.7 μg/mL. Similar to what occurs in honey, terpenes were the predominant class in the mead produced with yeast J (Figure 4c), corresponding to 39.7% of the total compounds identified or 15 μg/mL.

Of the 128 volatile compounds identified, 19 were classified as key aroma compounds (OAV ≥ 1; Table 5; Table S1). Among the compounds present in all three meads (C, TF, J), 1-Butanol, 3-methyl- (OCR = 65.89%, 55.1%, and 37.4%) and 1-Butanol, 3-methyl-, acetate (OCR = 15.47%, 31.3%, and 50.2%) stood out, being considered the most relevant in terms of aromatic impact, providing fermented and fruity notes, respectively.

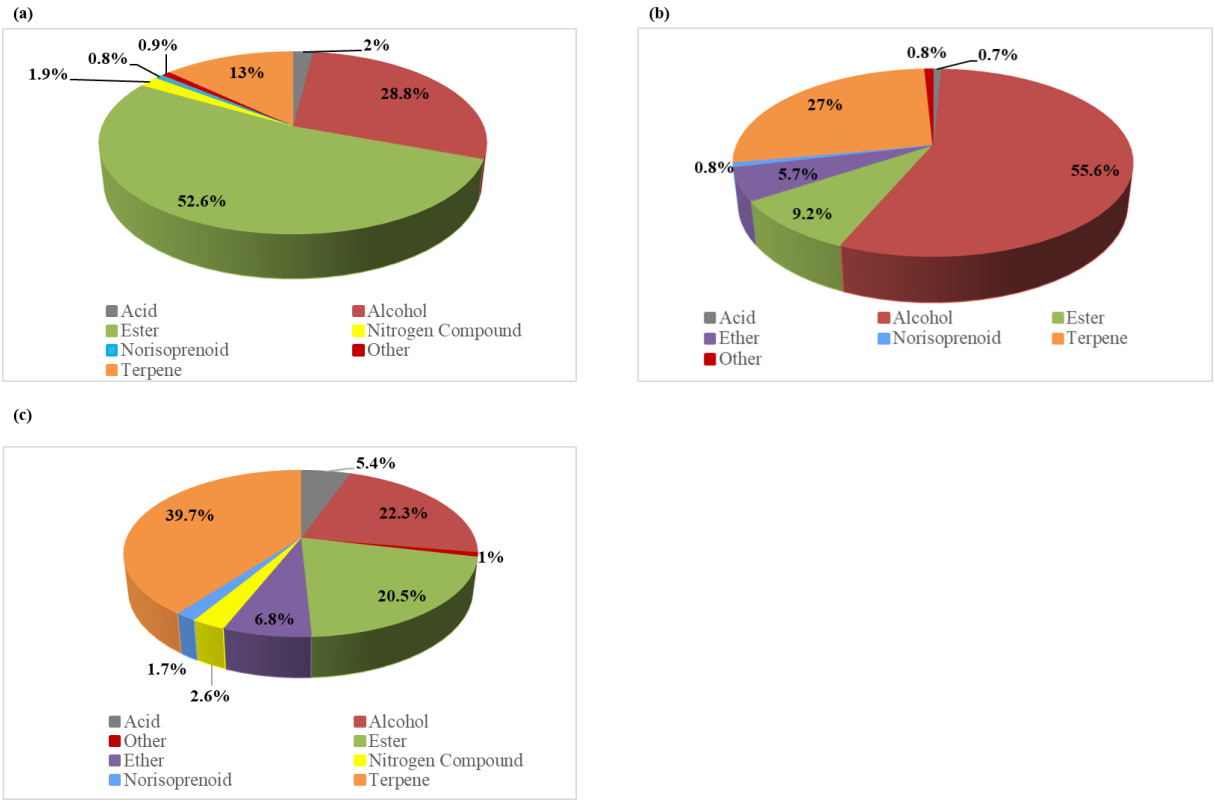

Figure 3: Distribution of the concentration of volatile compounds in mead fermented with different yeasts: (a) yeast C, (b) yeast TF and (c) yeast J.

The autochthonous yeast TF showed a greater diversity of higher alcohols than the commercial strain C, contributing to a more complex and balanced aromatic profile. It also showed a higher concentration of 1-Butanol, 3-methyl- (isoamyl alcohol) (13.6 µg/mL) compared to the commercial strain (8.8 µg/mL). This result differs from the findings of Parapouli et al. (2019), who observed a higher concentration of this compound in commercial strains, attributing this performance to high expression of genes such as BAT1, BAT2, and ARO10, involved in the Ehrlich pathway. This divergence may be related to the origin of the strains, as TF was reused from the cachaça industry, which may have made it more adapted to stressful fermentation conditions, whereas the strain analyzed by Parapouli et al. (2019) was

isolated from grapes. Additionally, the fermentation in this study occurred at 28 ± 2 °C, a higher temperature than the 18 °C used by Parapouli et al. (2019).

Alcohols are compounds widely present in fermented foods and play a crucial role in defining the aromatic profile of these products. Among them, ethanol is one of the most common, but higher alcohols, such as 1-Butanol, 3-methyl- (isoamyl alcohol), are responsible for more complex and intense aromas (Yeshurun & Sobel, 2010; De-La-Fuente-Blanco et al., 2016). According to De-La-Fuente-Blanco et al. (2016) and Rapp & Versini (1991), these compounds have a positive influence on the aroma of red wines, contributing alcoholic, floral, and fruity notes when present at concentrations below 300 µg/mL, but they may negatively affect the perception of these aromas when present at higher concentrations.

The formation of higher alcohols occurs mainly through the corresponding amino acid pathway, via the Ehrlich route, in which amino acids are transaminated, decarboxylated, and subsequently reduced to alcohols (Hazelwood et al., 2008). Yeasts such as *Saccharomyces cerevisiae* are the main microorganisms involved in this conversion during alcoholic fermentations. The intensity and diversity of alcohols produced are directly related to the microbial strain used (Parapouli et al., 2019).

Esters, in turn, are widely recognized for their sweet and fruity aromas and are considered the main contributors to pleasant sensory notes in fermented beverages (Verstrepen et al., 2003). 1-Butanol, 3-methyl-, acetate (known as isoamyl acetate) is one of the most relevant esters in the aromatic context of fermented foods and beverages. Its biosynthesis occurs through the action of alcohol acetyltransferase (AATase) enzymes, which catalyze the reaction between the precursor alcohol 1-Butanol, 3-methyl- (isoamyl alcohol) and the acetyl donor molecule acetyl-CoA (Zhang et al., 2012).

The regulation of isoamyl acetate biosynthesis is multifactorial and depends on the interaction between genetic and metabolic factors. The main enzymes involved in this process, alcohol acetyltransferases (AATases), are encoded by the ATF1 gene, whose expression levels directly influence ester production (Zhang et al., 2012). Furthermore, the availability of substrates, especially isoamyl alcohol and acetyl-CoA, regulates the reaction flux. The accumulation or scarcity of these compounds may limit or stimulate ester formation (Mitra et al., 2022).

Table 5: Key compounds in mead fermented with different yeasts (C, TF, J).

Name	Description	Threshold (µg/mL)	Concentration (µg/mL)			OAV			OCR (%)		
			C	TF	J	C	TF	J	C	TF	J
1-Butanol, 3-methyl-	Fermented	0.004	$8.827^{b} \pm 2.065$	$13.585^{a} \pm 1.895$	$5.123^{b} \pm 0.903$	2206.7	3396.4	1280.7	65.89	55.11	37.4
1-Butanol, 2-methyl-	Ethereal	0.0159	n.d	$1.741^{a} \pm 0.283$	n.d	n.d	109.5	n.d	n.d	1.8	n.d
1-Hexanol	Herbal	0.0056	$0.010^{a} \pm 0.001$	n.d	n.d	1.7	n.d	n.d	0.05	n.d	n.d
1-Heptanol	Green	0.0054	$0.030^{a} \pm 0.003$	n.d	n.d	5.6	n.d	n.d	0.17	n.d	n.d
Phenylethyl alcohol	Floral	0.14	$8.004^{b} \pm 2.138$	$14.678^{a} \pm 3.054$	$0.679^{c} \pm 0.011$	57.2	104.8	4.9	1.71	1.7	0.1
Benzeneacetaldehyde	Green	0.0063	$0.020^{b} \pm 0.007$	$0.084^{a} \pm 0.003$	$0.021^{b} \pm 0.003$	3.2	13.4	3.3	0.09	0.2	0.1
β-Cyclocitral	Tropical	0.003	n.d	n.d	$0.100^{a} \pm 0.016$	n.d	n.d	33.4	n.d	n.d	100
Methyleugenol	Spicy	0.006	$0.079^{a} \pm 0.004$	$0.044^{b} \pm 0.003$	$0.038^{b} \pm 0.005$	13.2	7.4	6.4	0.39	0.1	20
Ethyl acetate	Fruity, Ethereal	0.005	n.d	$0.722^{a} \pm 0.262$	$0.699^{a} \pm 0.126$	n.d	144.4	139.8	n.d	2.3	410
Ethyl caproate	Fruity	0.005	$0.3508^{b} \pm 0.0353$	$0.4195^{a} \pm 0.0160$	$0.0624^{c} \pm 0.0029$	70.2	83.9	12.5	2.1	1.4	0.4
Ethyl caprylate	Waxy	0.0193	n.d	$1.762^{a} \pm 0.276$	$0.081^{b} \pm 0.005$	n.d	91.3	4.2	n.d	1.5	0.1
Ethyl caprate	Waxy	0.005	$0.89^{a} \pm 0.30$	$0.34^{b} \pm 0.08$	$0.039^{b} \pm 0.003$	178.4	68.6	7.9	5.33	1.1	0.2
Ethyl α-toluate	Floral	0.1556	$0.38^{a} \pm 0.12$	n.d	$0.068^{b} \pm 0.009$	2.4	n.d	n.d	0.07	n.d	n.d
Butanoic acid, ethyl ester	Fruity	0.0009	$0.009^{b} \pm 0.003$	$0.0165^{a} \pm 0.0009$	$0.006^{b} \pm 0.002$	10	18.3	6.3	0.3	0.3	0.2
1-Butanol, 3-methyl-acetate	Fruity	0.0002	$0.078^{b} \pm 0.003$	$0.289^{a} \pm 0.022$	$0.258^{a} \pm 0.015$	518	1925.1	1719.1	15.47	31.3	50.2
1-Butanol, 2-methyl-acetate	Fruity	0.005	$0.012^{c} \pm 0.004$	$0.06^{b} \pm 0.01$	$0.089^{a} \pm 0.006$	2.5	12	17.8	0.07	0.2	50
Phenethyl acetate	Floral	0.2496	$32.65^{a} \pm 1.07$	$0.878^{c} \pm 0.168$	$6.3510^{b} \pm 0.7498$	130.8	3.5	25.4	3.91	0.1	0.7
cis-Linalool oxide	Earthy, Floral	0.1	n.d	$10.05^{a} \pm 2.28$	$9.5^{a} \pm 0.7$	n.d	100.5	94.7	n.d	1.6	280
Linalool oxide A	Floral	0.06	$8.935^{a} \pm 2.158$	$4.796^{b} \pm 0.826$	$4.148^{b} \pm 0.194$	148.9	79.9	69.1	4.45	1.3	2

Note. Adapted from The Good Scents Company (n.d.) and Van Gemert (2011). Values represent mean ± standard deviation (n = 3). Different letters in the same row indicate significant difference by Tukey's test ($p < 0.05$); nd = not detected.

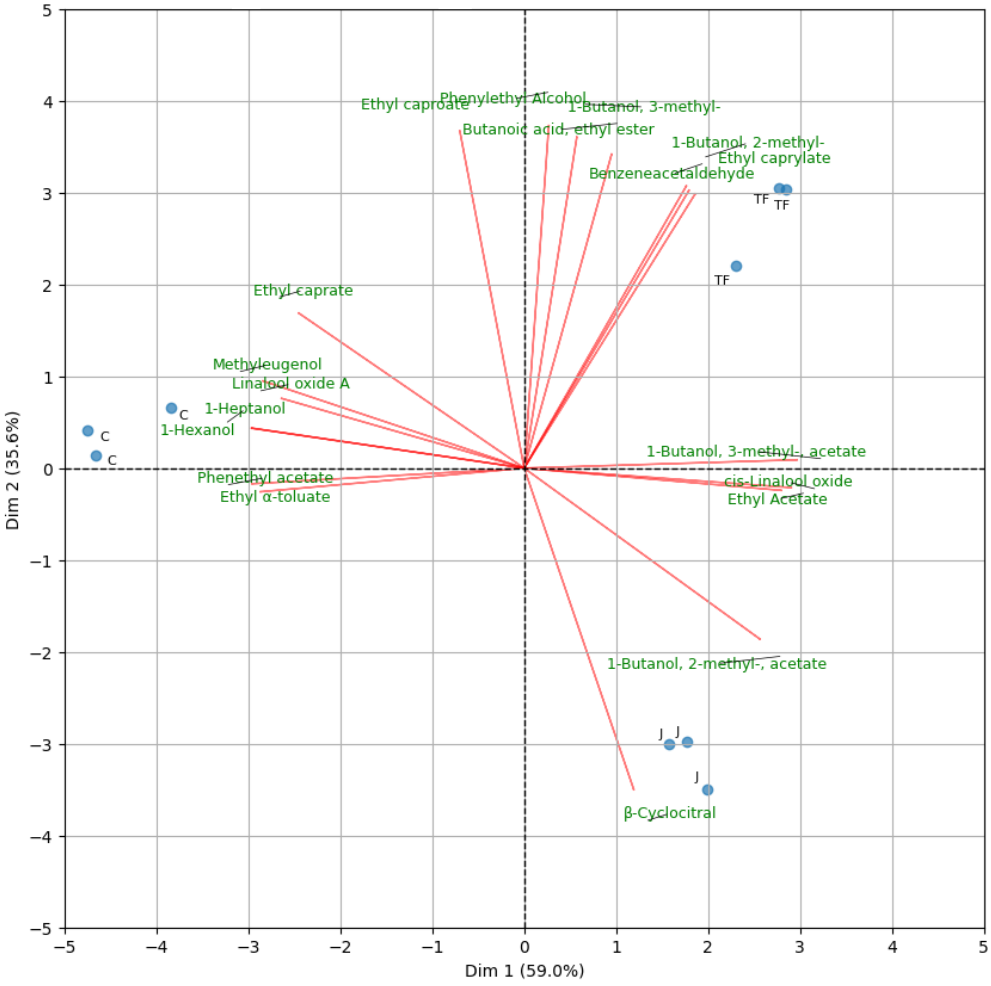

Figure 4: Key compounds in mead fermented with different yeasts (C, TF, J).

The principal component analysis (PCA) based on the key aromatic compounds identified in the meads, allowing for visualization of the contribution of these compounds to the differentiation between the samples fermented with yeasts C, TF, and J (figure 4). The relative positioning of the samples and vectors reveals differentiation patterns among the volatile profiles. The mead fermented with yeast TF is strongly associated with compounds such as benzeneacetaldehyde, ethyl caprylate, and 1-butanol, 2-methyl-, suggesting that these compounds are distinctive markers of this mead, contributing green, waxy, and ethereal notes.

On the other hand, the mead fermented with strain J showed greater proximity to compounds such as β-cyclocitral and 1-butanol, 2-methyl-, acetate, serving as distinctive markers for this formulation and contributing tropical and fruity notes. Meanwhile, the samples fermented with the commercial yeast C clustered in a region opposite to the others,

near compounds such as 1-heptanol, 1-hexanol, ethyl α-toluate, phenethyl acetate, linalool oxide A, and methyleugenol, contributing green, herbal, floral, and spicy notes.

Although some compositions present high OAV and OCR values in all samples, such as isoamyl alcohol and its corresponding ester, their balanced distribution in all formulations prevents them from acting as differentiators between meads. The differentiation patterns revealed by PCA are not necessarily associated with the sensory importance of the compounds (Braga et al., 2013). Thus, the graphs reinforce that the most relevant compounds for distinguishing formulations are not always the most impactful from a sensory point of view.

3.6 Total phenolics and antioxidant activity

The bioactive activity of meads produced with different yeasts varied significantly in the analyzed parameters, reflecting the influence of the strain type on the retention and transformation of bioactive compounds during alcoholic fermentation (table 6). Strain TF presented the highest concentration of total phenolic compounds (0.213 mg gallic acid/100 mL), followed by samples C (0.204 mg/100 mL) and J (0.19 mg/100 mL). The statistical difference observed between TF and the other samples ($p < 0.05$) indicates that the autochthonous strain TF was more efficient in preserving or releasing phenolic compounds during fermentation.

Table 6: Total phenolic content and antioxidant activity (ABTS, FRAP and DPPH assays) of mead samples fermented with different yeast strains (TF, J and C).

Yeast	Fenólicos (mg Ac.Gál./100 mL)	ABTS (µmol TE/100 mL)	FRAP (µmol TE/100 mL)	DPPH (µmol TE/100 mL)
TF	$0.213^{a} \pm 0.001$	$0.48^{a} \pm 0.08$	$1.49^{b} \pm 0.08$	$0.41^{a} \pm 0.04$
J	$0.19^{c} \pm 0.00$	$0.48^{a} \pm 0.03$	$1.3^{c} \pm 0.0$	$0.48^{a} \pm 0.03$
C	$0.204^{b} \pm 0.005$	$0.45^{a} \pm 0.04$	$1.83^{a} \pm 0.03$	$0.39^{a} \pm 0.06$

Values represent mean ± standard deviation (n = 3). Different letters in the same column indicate significant difference by Tukey's test ($p < 0.05$); TE = Trolox Equivalent.

Regarding the antioxidant capacity evaluated by the ABTS method, no statistically significant differences were observed between the samples ($p > 0.05$), indicating that the type of yeast did not influence the measured antioxidant capacity, that is, the ability to neutralize the synthetic radical ABTS•+ in the laboratory assay.

For the antioxidant capacity test using the FRAP method, which is based on the reduction of a ferric complex to ferrous, the commercial yeast presented the highest concentration (1.83µmol TE/100mL), differing significantly from the other two mead formulations ($p<0.05$) produced with autochthonous yeasts, with the TF yeast presenting the lowest performance.

The results of the antioxidant capacity test using the DPPH method, which is based on the neutralization of the stable free radical DPPH• by hydrogen or electron donating compounds, did not show any significant difference between the three formulations ($p > 0.05$), suggesting that the type of yeast did not influence the antioxidant capacity measured by this method.

The findings in this study are in agreement with those found by Grieco et al. (2019). Grieco et al. (2019) compared the increase in bioactive activity between indigenous and commercial yeasts in wines. Their results showed that the six strains analyzed significantly increased the content of total phenolic compounds and antioxidant capacity, assessed by the ABTS method.

Although strain choice positively influenced phenolic contents and antioxidant activity, the observed increase was not as significant when compared with values obtained using commercial strains. This indicates that, in addition to yeast selection, the use of complementary strategies, such as adjuncts rich in phenolic compounds (fruits, spices, herbs) and the use of wood chips for mead maturation, can be explored to enhance these parameters (Socha et al., 2019; Fortes et al., 2023).

3.7 Profile of sugars, organic acids and phenolic compounds

The differences in the metabolism of each strain were reflected in the final meads, creating unique profiles of sugars, organic acids, and phenolic compounds (Figure 5). The meads obtained with yeasts J and C presented the highest phenolic concentrations, such as naringin, 44.3 mg/L and 47.8 mg/L, respectively. This phenolic compound is a member of the flavonoid class and is very abundant in citrus fruits, but is also found in bee products originating from the flora of the Caatinga biome (Aldana-Mejía et al., 2024). In contrast, the mead fermented with strain TF did not present this compound, but did have a high concentration of hesperidin (178 mg/L).

The exclusive presence of hesperidin in mead fermented with yeast TF, related to the absence of naringin, may be associated with the metabolic transformation of structurally similar phenolic compounds. Both naringin and hesperidin belong to the flavonoid class, a

subclass of flavanones, and share the same aglycone (naringenin), differing only in the conjugated sugar residues (Madureira et al., 2023). This structural similarity suggests the possibility of enzymatic conversions during fermentation, mediated by the metabolic activity of the TF strain.

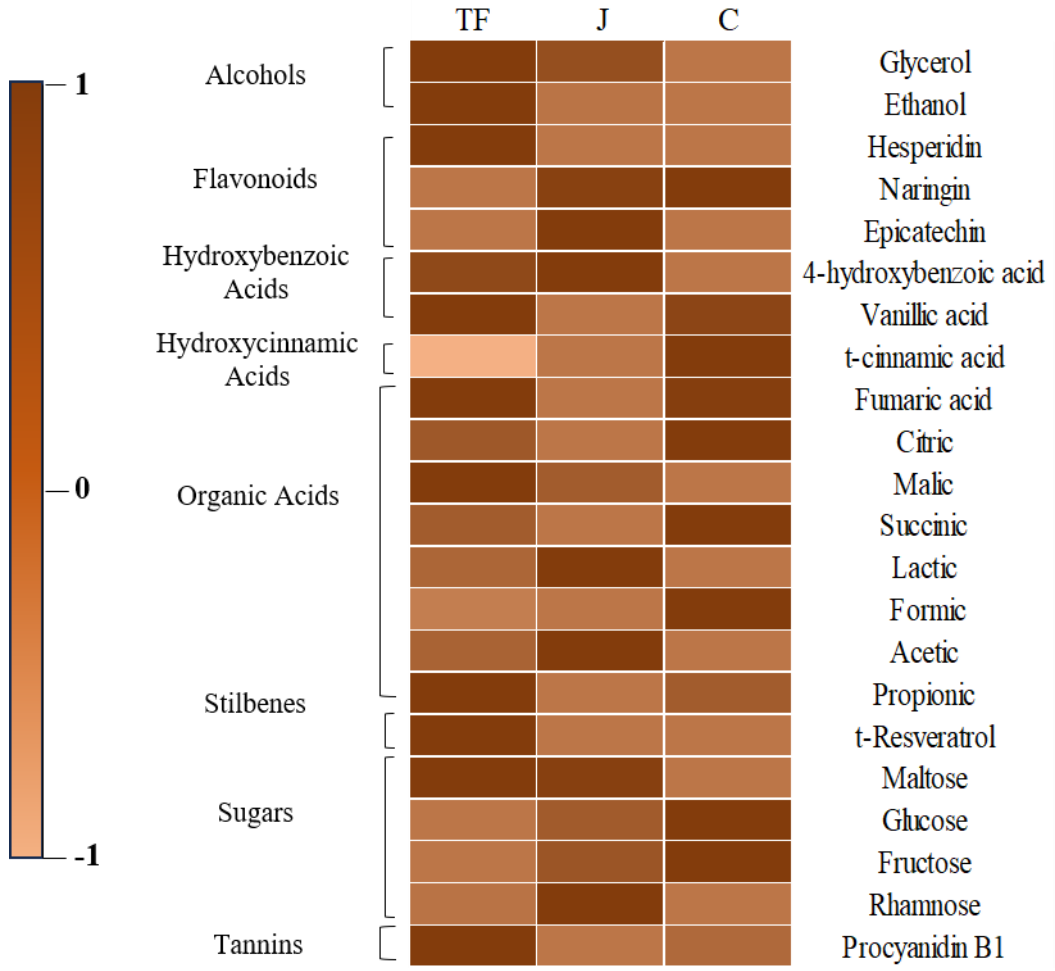

Figure 5: Heatmap showing the profile of sugars, organic acids and phenolic compounds in mead samples fermented with different yeast strains (TF, J and C).

Hesperidin has cardiovascular, neurological, and antitumor effects (Li & Schluesener, 2017). Furthermore, it has anti-inflammatory properties and effects on the regulation of lipid and glucose metabolism (Xiong et al., 2019). Naringin has protective effects against obesity, diabetes, hypertension, and oxidative stress (Alam et al., 2014). It has also been widely studied for its therapeutic properties, including antioxidant, anti-inflammatory, and antitumor activities (Chen et al., 2016; Ghanbari-Movahed et al., 2021).

Among the three meads evaluated, the formulation obtained with commercial yeast (C) had the highest citric acid content (0.6 g/L). The presence of citric acid in the meads may

be related to residual mitochondrial activity during fermentation, as this compound is a key intermediate in the Krebs cycle, which occurs in the mitochondrial matrix (Daunoraitė et al., 2024). This result may be associated with situations of low glucose availability or greater cellular stress, in which there is residual activation of mitochondrial pathways and increased respiratory flow through the Krebs cycle (Blank & Sauer, 2004). Among the organic acids evaluated, acetic acid presented the highest concentrations in the TF and J formulations (0.5 and 1.4 g/L, respectively). It is a byproduct of alcoholic fermentation by *Saccharomyces cerevisiae* that can act as a cellular stress agent, being involved in programmed cell death (Giannattasio et al., 2013). However, this risk is only associated with concentrations above 4.8 g/L. Therefore, the levels observed in the formulations analyzed remain below this limit, not posing a significant risk to yeast (Ludovico et al., 2001); (Giannattasio et al., 2005).

Residual fructose concentrations were found in small amounts in the TF mead, suggesting a more efficient metabolism of the indigenous TF yeast in assimilating this monosaccharide. In meads J and C, the higher values (13.9 and 23.9 g/L, respectively) indicate that some of the fructose was not consumed, which may be related to the strain's lower affinity for this sugar or possible incomplete fermentation. The efficiency of fructose utilization during fermentation is multifactorial, involving the specific characteristics of the yeast strain, the conditions of the fermentation environment, and the possible influence of factors such as ethanol or nutrient deficiencies (Berthels et al., 2004).

In contrast, mead C had the lowest residual maltose concentration (8.7 g/L) compared to formulations fermented with yeasts TF and J (11.2 and 11 g/L, respectively). This result may be directly related to the fact that the yeast used was baker's yeast. The greater ability of these baker's yeasts to utilize maltose is likely due to their artificial selection of duplications in the MAL genes, which are involved in maltose metabolism (Bai et al., 2022). Wild strains, such as those used in cachaça production, do not necessarily share this ability.

Among the meads evaluated, the TF formulation presented the highest glycerol concentration (2.37 g/L), followed by J (2.1 g/L) and C (1.51 g/L). Glycerol production in *S. cerevisiae* occurs in response to ethanol stress, acting as an alternative pathway for NAD^+ regeneration (Vriesekoop et al., 2009). Thus, the increased synthesis of this compound by indigenous yeasts represents a more efficient adaptive response to preserve cell viability, helping to maintain cellular integrity and functionality, contributing to better fermentation performance (Long et al., 2022).

The results of this study demonstrate that indigenous yeasts from cachaça mills play a decisive role in the characteristics of mead, influencing its chemical composition, volatile

compound profile, production of substances with functional potential, and fermentation performance (with emphasis on the TF strain). These results enable the industry to explore biodiversity and value regional ingredients and microorganism strains to obtain alcoholic beverages with unique characteristics. However, the limitations of the work performed include: use of laboratory scale, with a reduced number of strains evaluated and no inclusion of sensory analysis. Suggestions for future work: increasing the scale of the process to a pilot volume, increasing the number of native strains, performing sensory analysis with trained panels and consumers, in addition to including detailed genetic analyses of the same strains and investigating the use of mixed cultures, aiming to improve process performance. The scientific contributions of this work include expanding knowledge about the role of native yeasts in fermentation, reconciling tradition and innovation, in addition to offering the industry a sustainable and low-cost alternative for the development of beverages with high sensory potential.

4 Conclusion

The results of this study demonstrate that yeast selection is a decisive factor in mead production. The TF strain stood out for its higher fermentative efficiency, producing a mead with greater alcohol content, lower residual sugar, higher fructose consumption, and improved kinetic and phenolic profiles. In contrast, the commercial yeast (C) showed limited performance, while strain J exhibited intermediate behavior. Sensory and chemical analyses revealed that, although the predominant compounds were similar, significant differences occurred in compounds present at lower concentrations, highlighting the influence of yeast on the final character of the beverage. Therefore, indigenous yeasts, particularly TF, have great potential to produce meads with differentiated characteristics, especially when combined with regional raw materials such as Caatinga honey, enhancing regional identity, sensory differentiation, and competitiveness in the fermented beverage market.

References

Alam, M. A., Subhan, N., Rahman, M. M., Uddin, S. J., Reza, H. M., & Sarker, S. D. (2014). Effect of citrus flavonoids, naringin and naringenin, on metabolic syndrome and their mechanisms of action. *Advances in Nutrition, 5(4)*, 404–417. https://doi.org/10.3945/an.113.005603.

Aldana-Mejía, J., Ribeiro, V., Katragunta, K., Avula, B., Tatapudi, K., Bastos, J., Khan, I., Meepagala, K., & Ross, S. (2024). Chemical Characterization and Antimicrobial Activity of Green Propolis from the Brazilian Caatinga Biome. *Plants, 13*. https://doi.org/10.3390/plants13243576.

Association of Official Analytical Chemists. (2016). *Official methods of analysis* (20th ed.). AOAC International.

Attfield, P. V. (1997). Stress tolerance: The key to effective strains of industrial baker's yeast. *Nature Biotechnology, 15(13)*, 1351–1357. https://doi.org/10.1038/nbt1297-1351.

Bai, F., Han, D., Duan, S., & Wang, Q. (2022). The Ecology and Evolution of the Baker's Yeast Saccharomyces cerevisiae. *Genes, 13*. https://doi.org/10.3390/genes13020230.

Bauer, F. F., & Pretorius, I. S. (2000). Yeast stress response and fermentation efficiency: How to survive the making of wine – A review. *South African Journal of Enology and Viticulture, 21(1)*, 27–51. https://doi.org/10.21548/21-1-3557.

Benzie, I. F., & Strain, J. J. (1996). The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": the FRAP assay. Analytical Biochemistry, 239(1), 70-76. https://doi.org/10.1006/abio.1996.0292.

Biluca, F. C., de Gois, J. S., Schulz, M., Braghini, F., Gonzaga, L. V., Maltez, H. F., Rodrigues, E., Borges, D. L. G., Costa, A. C. O., & Fett, R. (2017). Phenolic compounds, antioxidant capacity and bioaccessibility of minerals of stingless bee honey (Meliponinae). *Journal of Food Composition and Analysis*, *63*, 89-97. https://doi.org/10.1016/j.jfca.2017.07.039.

Bisson, L. F. (1999). Stuck and sluggish fermentations. *American Journal of Enology and Viticulture, 50(1)*, 107–119. https://www.ajevonline.org/content/50/1/107.

Blank, L. M., & Sauer, U. (2004). TCA cycle activity in Saccharomyces cerevisiae is a function of the environmentally determined specific growth and glucose uptake rates. *Microbiology, 150(4)*, 1085–1093. https://doi.org/10.1099/mic.0.26845-0.

BOGDANOV, Stefan. *The Book of Honey: A short history of honey*. Mühletürnen: Bee Product Science, 2011.

Braga, C. M., Zielinski, A. A. F., Silva, K. M. da, de Souza, F. K. F., Pietrowski, G. de A. M., Couto, M., & Nogueira, A. (2013). Classification of juices and fermented beverages made from unripe, ripe and senescent apples based on the aromatic profile using chemometrics. *Food Chemistry, 141(2)*, 967–974. https://doi.org/10.1016/j.foodchem.2013.04.007.

Brazil. (2000). Normative Instruction No. 11, of September 27, 2000. Technical regulation on the identity and quality of honey. Brasília.

Canseco Grellet, M. A., Dantur, K. I., Perera, M. F., Ahmed, P. M., Castagnaro, A., Arroyo-Lopez, F. N., Gallego, J. B., Welin, B., & Ruiz, R. M. (2022). Genotypic and phenotypic characterization of industrial autochthonous *Saccharomyces cerevisiae* for the selection of well-adapted bioethanol-producing strains. *Fungal Biology*, *126(10),* 658–673. https://doi.org/10.1016/j.funbio.2022.08.004.

Castell, A., Arroyo-Manzanares, N., Guerrero-Núñez, Y., Campillo, N., & Viñas, P. (2023). Headspace with gas chromatography-mass spectrometry for the use of volatile organic

compound profile in botanical origin authentication of honey. *Molecules, 28(11)*, 4297. https://doi.org/10.3390/molecules28114297.

Chen, R., Qi, Q.-L., Wang, M.-T., & Li, Q.-Y. (2016). Therapeutic potential of naringin: An overview. Pharmaceutical *Biology, 54(12)*, 3203–3210. https://doi.org/10.1080/13880209.2016.1216131.

Chitarrini, G., Debiasi, L., Stuffer, M., Ueberegger, E., Zehetner, E., Jaeger, H., Robatscher, P., & Conterno, L. (2020). Volatile profile of mead fermenting blossom honey and honeydew honey with or without Ribes nigrum. *Molecules, 25(8)*, 1818. https://doi.org/10.3390/molecules25081818.

Coelho, E. M., da Silva Padilha, C. V., Miskinis, G. A., de Sá, A. G. B., Pereira, G. E., de Azevêdo, L. C., & dos Santos Lima, M. (2018). Simultaneous analysis of sugars and organic acids in wine and grape juices by HPLC: Method validation and characterization of products from northeast Brazil. Journal of Food Composition and Analysis, 66, 160-167. https://doi.org/10.1016/j.jfca.2017.12.017.

Da Silva, P. M., Gauche, C., Gonzaga, L. V., Costa, A. C. O., & Fett, R. (2016). Honey: Chemical composition, stability and authenticity. *Food Chemistry, 196*, 309–323. https://doi.org/10.1016/j.foodchem.2015.09.051.

Da Silva, J. R., Correia-Lima, L., Fernandes, G., Ribeiro-Filho, N., Madruga, M. S., Lima, M. dos S., & Muniz, M. B. (2023). Mandacaru fruit pulp (Cereus jamacaru D.C.) as an adjunct and its influence on the beer properties. *Food Chemistry, 406*, Article 135066. https://doi.org/10.1016/j.foodchem.2022.135066.

Daunoraitė, K., Castañeda-Ovando, A., Araújo, R. G., Venskutonis, P. R., & Ferreira, I. C. F. R. (2024). Integrated application of grape pomace: From the fermentation of phenolic compounds to circular economy and sustainable practices. *Journal of Cleaner Production, 448*, 140146. https://doi.org/10.1016/j.jclepro.2024.142914.

Deng, Y., Kan, H., Li, Y., Liu, Y., & Qiu, X. (2023). Analysis of Volatile Components in Rosa roxburghii Tratt. and Rosa sterilis Using Headspace–Solid-Phase Microextraction–Gas Chromatography–Mass Spectrometry. *Molecules*, *28(23)*, 7879. https://doi.org/10.3390/molecules28237879.

De-La-Fuente-Blanco, A., Sáenz-Navajas, M. P., & Ferreira, V. (2016). On the effects of higher alcohols on red wine aroma. *Food Chemistry, 210*, 107–114. https://doi.org/10.1016/j.foodchem.2016.04.021.

Dewick, P. M. (2009). *Medicinal natural products: A biosynthetic approach* (3rd ed.). John Wiley & Sons.

Escuredo, O., Míguez, M., Fernández-González, M., & Seijo, M. C. (2013). Nutritional value and antioxidant activity of honeys produced in a European Atlantic area. *Food Chemistry, 138*(2–3), 851–856. https://doi.org/10.1016/j.foodchem.2012.11.015

FAO; WHO. (2000). *The draft revised Codex standard for honey*. Codex Alimentarius Commission. Food and Agriculture Organization of the United Nations.

Finola, M. S., Lasagno, M. C., & Marioli, J. M. (2007). Microbiological and chemical characterization of honeys from central Argentina. *Food Chemistry, 100*(4), 1649–1653. https://doi.org/10.1016/j.foodchem.2005.12.046

Fisher, C., & Scott, T. R. (2007). *Food flavours: Biology and chemistry*. Royal Society of Chemistry.

Fortes, J. P., Franco, F. W., Baranzelli, J., Ugalde, G. A., Ballus, C. A., Rodrigues, E., Mazutti, M. A., Somacal, S., & Sautter, C. K. (2023). Enhancement of the Functional Properties of Mead Aged with Oak (Quercus) Chips at Different Toasting Levels. *Molecules, 28(1)*, 56. https://doi.org/10.3390/molecules28010056.

Gaweł, R., Sluyter, S., & Waters, E. (2007). The effects of ethanol and glycerol on the body and other sensory characteristics of Riesling wines. *Australian Journal of Grape and Wine Research*, 13, 38-45. https://doi.org/10.1111/J.1755-0238.2007.TB00070.X.

Ghanbari-Movahed, M., Jackson, G., Farzaei, M. H., & Bishayee, A. (2021). A systematic review of the preventive and therapeutic effects of naringin against human malignancies. *Frontiers in Pharmacology, 12*, 639840. https://doi.org/10.3389/fphar.2021.639840.

Ghazali, N., Chin, N., Othman, S., Chang, L., Baroyi, S., & Yusof, Y. (2024). Stability of physicochemical and microbiological properties of *Geniotrigona thoracica* honey after moisture reduction and long-term storage in different containers. *Journal of Food Composition and Analysis, 136*, 106767. https://doi.org/10.1016/j.jfca.2024.106767.

Giannattasio, S., Guaragnella, N., Corte Real, M., Passarella, S., & Marra, E. (2005). Acid stress adaptation protects Saccharomyces cerevisiae from acetic acid–induced programmed cell death. *Gene, 354*, 93–98. https://doi.org/10.1016/j.gene.2005.03.030.

Giannattasio, S., Guaragnella, N., Ždralević, M., & Marra, E. (2013). Molecular mechanisms of Saccharomyces cerevisiae stress adaptation and programmed cell death in response to acetic acid. *Frontiers in microbiology, 4*, 33. https://doi.org/10.3389/fmicb.2013.00033.

Grieco, F., Carluccio, M. A., & Giovinazzo, G. (2019). Autochthonous Saccharomyces cerevisiae Starter Cultures Enhance Polyphenols Content, Antioxidant Activity, and Anti-Inflammatory Response of Apulian Red Wines. *Foods, 8(10)*, 453. https://doi.org/10.3390/foods8100453.

Hallahan, T. W., & Croteau, R. (1989).Monoterpene biosynthesis: Mechanism and stereochemistry of the enzymatic cyclization of geranyl pyrophosphate to (+)-cis- and (+)-trans-sabinene hydrate. *Archives of Biochemistry and Biophysics, 269*(1), 313–326. https://doi.org/10.1016/0003-9861(89)90113-6.

Hazelwood, L. A., Daran, J. M., van Maris, A. J., Pronk, J. T., & Dickinson, J. R. (2008). The Ehrlich pathway for fusel alcohol production: a century of research on Saccharomyces cerevisiae metabolism. *Applied and Environmental Microbiology, 74(8),* 2259–2266. https://doi.org/10.1128/AEM.02625-07.

Hernández, O. M., Fraga, J. M. G., Jimenez, A. I., Jimenez, F., & Arias, J. J. (2005). Characterization of honey from the Canary Islands: Determination of the mineral content by atomic absorption spectrophotometry. *Food Chemistry, 93*(3), 449–458. https://doi.org/10.1016/j.foodchem.2004.10.036.

HOHMANN, S.; MAGER, W. H. (Ed.). Yeast stress responses. 1. ed. Berlin: Springer, 2003. v. 1. DOI: https://doi.org/10.1007/3-540-45611-2.

Houngbédji, M., Johansen, P., Padonou, S., Hounhouigan, D., Siegumfeldt, H., & Jespersen, L. (2019). Effects of intrinsic microbial stress factors on viability and physiological condition

of yeasts isolated from spontaneously fermented cereal doughs.. *International journal of food microbiology*, 304, 75-88. https://doi.org/10.1016/J.IJFOODMICRO.2019.05.018.

Jerković, I., & Kuś, P. M. (2014). Terpenes in honey: Occurrence, origin and their role as chemical biomarkers. *RSC Advances, 4*(60), 31710–31728. https://doi.org/10.1039/C4RA04791E

Kotseridis, Y., Baumes, R. L., & Skouroumounis, G. K. (1999). Quantitative determination of free and hydrolytically liberated β-damascenone in red grapes and wines using a stable isotope dilution assay. *Journal of Chromatography A, 849*(1), 245–254. https://doi.org/10.1016/S0021-9673(99)00540-3.

Li, C., & Schluesener, H. (2017). Health-promoting effects of the citrus flavanone hesperidin. *Critical Reviews in Food Science and Nutrition, 57(3)*, 613–631. https://doi.org/10.1080/10408398.2014.906382.

Lima, M. S., Dantas, B. S., Carvalho, A. J. D. B. A., Pereira, G. E., Pimentel, T. C., & Magnani, M. (2024). A novel method for ultra-fast determination of phenolics with performance comparable to UPLC/DAD: Method development and validation on analysis of seedless table grapes. Journal of Food Composition and Analysis, 134, 106511. DOI: https://doi.org/10.1016/j.jfca.2024.106511.

Long, X., Xu, Y., & Zhao, X. (2024). Response mechanism of Saccharomyces cerevisiae under benzoic acid stress in ethanol fermentation. Scientific Reports, 14. https://doi.org/10.1038/s41598-024-80484-1.

Ludovico, P., Sousa, M. J., Silva, M. T., Leão, C., & Corte-Real, M. (2001). Saccharomyces cerevisiae commits to a programmed cell death process in response to acetic acid. *Microbiology, 147(9)*, 2409–2415. https://doi.org/10.1099/00221287-147-9-2409.

Maffei, M. E., Gertsch, J., & Appendino, G. (2011). Plant volatiles: Production, function and pharmacology. *Natural Product Reports, 28*(8), 1359–1380. https://doi.org/10.1039/C1NP00021G.

Miller, G. L. (1959). Use of dinitrosalicylic acid reagent for determination of reducing sugar. Analytical Chemistry*, 31(3)*, 426–428. https://doi.org/10.1021/ac60147a030.

Mitra, M., Singh, R., Ghissing, U., Das, A., Mitra, A., & Maiti, M. (2022). Characterization of an alcohol acetyltransferase GcAAT responsible for the production of antifungal volatile esters in endophytic Geotrichum candidum PF005.. *Microbiological research, 260*, 127021 . https://doi.org/10.1016/j.micres.2022.127021.

Parapouli, M., Sfakianaki, A., Monokrousos, N., Nikas, V., Louka, N., & Hatziloukas, E. (2019). Comparative transcriptional analysis of flavor biosynthetic genes of a native Saccharomyces cerevisiae strain fermenting in its natural must environment, versus a commercial strain, and correlation of gene activities with the flavor compounds produced. *Journal of Biological Research-Thessaloniki, 26(1),* 5. https://doi.org/10.1186/s40709-019-0096-8.

Park, J., Lee, S., Lee, M., Park, K., Lee, S., Kim, J., & Kim, P. (2020). Accelerated Growth of Corynebacterium glutamicum by Up-Regulating Stress- Responsive Genes Based on Transcriptome Analysis of a Fast-Doubling Evolved Strain. *Journal of Microbiology and Biotechnology, 30*, 1420 - 1429. https://doi.org/10.4014/jmb.2006.06035.

Pereira, J. R., Campos, A. N. da R., Oliveira, F. C. de, Silva, V. R. O., David, G. F., Da Silva, J. G., Nascimento, W. W. G., Silva, M. H. L., & Denadai, Â. M. L. (2020). Physical-chemical characterization of commercial honeys from Minas Gerais, Brazil. *Food Bioscience, 36*, 100644. https://doi.org/10.1016/j.fbio.2020.100644.

Pierce, J. S. (1970). Institute of Brewing: Analysis Committee: Measurement of yeast viability. *Journal of the Institute of Brewing, 76(5)*, 442–443. https://doi.org/10.1002/j.2050-0416.1970.tb03325.x

Plotto, A., Margaría, C. A., Goodner, K. L., & Baldwin, E. A. (2008). Odour and flavour thresholds for key aroma components in an orange juice matrix: Esters and miscellaneous compounds. *Flavour and Fragrance Journal, 23*(6), 398–406. https://doi.org/10.1002/ffj.1888.

Rapp, A., & Versini, G. (1995). Influence of nitrogen compounds in grapes on aroma compounds of wines. In G. Charalambous (Ed.), *Food flavors: Generation, analysis and process influence* (Vol. 37, pp. 1659–1694). Elsevier. https://doi.org/10.1016/S0167-4501(06)80257-8.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free radical biology and medicine*, 26(9-10), 1231-1237. DOI: https://doi.org/10.1016/S0891-5849(98)00315-3.

Ribeiro, G., Villas-Bôas, J., Spinosa, W., & Prudêncio, S. (2017). Influence of freezing, pasteurization and maturation on Tiúba honey quality. *Lwt - Food Science and Technology*, *90,* 607-612. https://doi.org/10.1016/J.LWT.2017.12.072.

Rowland, C. Y., Blackman, A. J., D'Arcy, B. R., & Rintoul, G. B. (1995). Hotrienol in Australian honeys. *Journal of Agricultural and Food Chemistry, 43(3)*, 753–763. https://doi.org/10.1021/jf00051a036.

Rufino, M. S. M., Alves, R. E., Brito, E. S., Morais, S. M., Sampaio, C. G., Jimenez, J. P., & Calixto, F. (2007). Scientific Methodology: Determination of total antioxidant activity in fruits by free radical capture DPPH. Embrapa ISSN, 1679-6535.

Senn, K., Cantu, A., & Heymann, H. (2021). Characterizing the chemical and sensory profiles of traditional American meads. *Journal of Food Science, 86*(3), 1048–1057. https://doi.org/10.1111/1750-3841.15607.

Socha, R., Pająk, P., Fortuna, T., & Buksa, K. (2015). Phenolic Profile and Antioxidant Activity of Polish Meads. *International Journal of Food Properties, 18*, 2713 - 2725. https://doi.org/10.1080/10942912.2015.1004588.

Schwarz, L., Marcon, Â., Delamare, A., Agostini, F., Moura, S., & Echeverrigaray, S. (2020). Selection of low nitrogen demand yeast strains and their impact on the physicochemical and

volatile composition of mead. *Journal of Food Science and Technology, 57*, 2840-2851. https://doi.org/10.1007/s13197-020-04316-6.

Sottil, C., Salor-Torregrosa, J., Moreno-García, J., Peinado, J., Mauricio, J., Moreno, J., & García-Martínez, T. (2019). Using Torulaspora delbrueckii, Saccharomyces cerevisiae and Saccharomyces bayanus wine yeasts as starter cultures for fermentation and quality improvement of mead. *European Food Research and Technology, 245*, 2705 - 2714. https://doi.org/10.1007/s00217-019-03384-z.

Terrab, A., Recamales, Á. F., Hernanz, D., & Heredia, F. J. (2004). Characterisation of Spanish thyme honeys by their physicochemical characteristics and mineral contents. *Food Chemistry, 88*(4), 537–542. https://doi.org/10.1016/j.foodchem.2004.01.068.

The Good Scents Company. (2025). Information about fragrances and aromatic ingredients. https://www.thegoodscentscompany.com/

Tomasino, E., & Bolman, S. (2021). The potential effect of β-ionone and β-damascenone on sensory perception of Pinot Noir wine aroma. *Molecules, 26(5)*, 1288. https://doi.org/10.3390/molecules26051288

Tornuk, F., Karaman, S., Ozturk, I., Toker, O. S., Tastemur, B., Sagdic, O., Dogan, M., & Kayacier, A. (2013). Quality characterization of artisanal and retail Turkish blossom honeys: Determination of physicochemical, microbiological, bioactive properties and aroma profile. *Industrial Crops and Products, 46*, 124–131. https://doi.org/10.1016/j.indcrop.2012.12.042

Van Gemert, L. J. (2011). *Odour thresholds: Compilations of odour threshold values in air, water and other media* (2nd ed.). Oliemans Punter & Partners BV.

Verstrepen, K. J., Van Laere, S. D. M., Vanderhaegen, B. M. P., Derdelinckx, G., Dufour, J. P., Pretorius, I. S., Winderickx, J., Thevelein, J. M., & Delvaux, F. R. (2003). Expression levels of the yeast alcohol acetyltransferase genes ATF1, Lg-ATF1, and ATF2 control the formation of a broad range of volatile esters. *Applied and Environmental Microbiology, 69(9)*, 5228–5237. https://doi.org/10.1128/AEM.69.9.5228-5237.2003.

Vîjan, L., Mazilu, I., Enache, C., Enache, S., & Topală, C. (2023). Influência da origem botânica em algumas características físico-químicas e propriedades antioxidantes do mel. *Foods, 12*. https://doi.org/10.3390/foods12112134.

Villamor, R. R., & Ross, C. F. (2013). Wine matrix compounds affect perception of wine aromas. *Annual Review of Food Science and Technology, 4*, 1–20. https://doi.org/10.1146/annurev-food-030212-182707.

Vriesekoop, F., Haass, C., & Pamment, N. (2009). The role of acetaldehyde and glycerol in the adaptation to ethanol stress of Saccharomyces cerevisiae and other yeasts.. *FEMS yeast research, 9 3*, 365-71 . https://doi.org/10.1111/j.1567-1364.2009.00492.x.

Xiong, H., Wang, J., Ran, Q., Lou, G., Peng, C., Gan, Q., Hu, J., Sun, J., Yao, R., & Huang, Q. (2019). Hesperidin: A therapeutic agent for obesity. *Drug Design, Development and Therapy, 13*, 3855–3866. https://doi.org/10.2147/DDDT.S227499.

Yerushalmi, L., & Volesky, B. (1981). Experimental bioenergetics ofSaccharomyces cerevisiae in respiration and fermentation. *Biotechnology and Bioengineering, 23*. https://doi.org/10.1002/bit.260231017.

Yeshurun, Y., & Sobel, N. (2010). Multisensory integration: An inner tongue puts an outer nose in context. Nature Neuroscience, 13(2), 148–149. https://doi.org/10.1038/nn0210-148.

Yücel, Y., & Sultanoğlu, P. (2013). Characterization of honeys from Hatay Region by their physicochemical properties combined with chemometrics. *Food Bioscience, 1*, 16–25. https://doi.org/10.1016/j.fbio.2013.02.001.

Zhang, G.-Z., Chen, T., Zhou, J.-H., Wang, H.-Y., Zhang, L., Gao, H., & Yang, Y.-S. (2021). Investigation of the maturity evaluation indicator of honey in natural ripening process: The case of rape honey. *Foods, 10*(11), 2882. https://doi.org/10.3390/foods10112882.

Zhang, J., Zhang, C., Dai, L., Jian, D., Liu, Y., Guo, X., & Xiao, D. (2012). Effects of overexpression of the alcohol acetyltransferase-encoding gene ATF1 and disruption of the esterase-encoding gene IAH1 on the flavour profiles of Chinese yellow rice wine. *International Journal of Food Science and Technology, 47*, 2590-2596. https://doi.org/10.1111/J.1365-2621.2012.03140.X.

Zhu, M., Sun, J., Zhao, H., Wu, F., Xue, X., Wu, L., & Cao, W. (2022). Volatile compounds of five types of unifloral honey in Northwest China: Correlation with aroma and floral origin based on HS-SPME/GC–MS combined with chemometrics. Food Chemistry, 384, 132461. https://doi.org/10.1016/j.foodchem.2022.132461

6. CONSIDERAÇÕES FINAIS

Os resultados evidenciam que a seleção da levedura é decisiva para o sucesso da produção de hidromel, impactando fermentação, aroma e propriedades funcionais. A cepa TF destacou-se por sua maior eficiência fermentativa, maior formação de compostos chave (como álcool isoamílico e acetato de isoamila) e preservação de fenólicos e metabólitos bioativos. Cada cepa, contudo, imprimiu uma assinatura aromática própria, confirmada pela análise de componentes principais. No conjunto, esses resultados reforçam o potencial do uso de leveduras autóctones, associadas a matérias-primas locais, como uma estratégia promissora para o desenvolvimento de hidroméis com identidade própria, maior valor agregado e potencial competitivo no mercado de bebidas fermentadas.

REFERÊNCIAS

BRASIL. Agência Nacional de Vigilância Sanitária. Resolução da Diretoria Colegiada – RDC nº 740, de 9 de agosto de 2022. *Diário Oficial da União*: seção 1, Brasília, DF, 17 ago. 2022.

AMORIM, Henrique V. et al. Scientific challenges of bioethanol production in Brazil. *Applied microbiology and biotechnology*, v. 91, p. 1267-1275, 2011.

AOAC. *Association of Official Analytical Chemists*. 20. ed. Rockville, Maryland: AOAC International, 2016.

ATTFIELD, P.V. Stress tolerance: the key to effective strains of industrial baker's yeast. *Nature Biotechnology***,** v. 15, p. 1351-1357, 1997.

AZHAR, Siti Hajar Mohd et al. Yeasts in sustainable bioethanol production: A review. *Biochemistry and biophysics reports*, v. 10, p. 52-61, 2017.

BAI, F. W.; ANDERSON, W. A.; MOO-YOUNG, M. Ethanol fermentation technologies from sugar and starch feedstocks. *Biotechnology advances*, v. 26, n. 1, p. 89-105, 2008.

BASSI, Ana Paula Guarnieri et al. Effects of single and combined cell treatments based on low pH and high concentrations of ethanol on the growth and fermentation of Dekkera bruxellensis and Saccharomyces cerevisiae**.** *World Journal of Microbiology and Biotechnology*, v. 29, p. 1661-1676, 2013.

BASSO, Luiz C. et al. Yeast selection for fuel ethanol production in Brazil. *FEMS yeast research*, v. 8, n. 7, p. 1155-1163, 2008.

BAUER, F.F.; PRETORIUS, I.S. Yeast stress response and fermentation efficiency: how to survive the making of wine – a review. *South African Journal of Enology and Viticulture*, v. 21, p. 27-51, 2000.

BILVERSTONE, Terry W.; WHITE, Rod; BOULTON, Chris A. Manipulation of conditions during wort collection in production-scale fermentations to regulate volatile ester synthesis as an aid to product matching for multisite brewing. *Journal of the American Society of Brewing Chemists*, v. 73, n. 4, p. 347-353, 2015.

BISSON, L.F. Stuck and sluggish fermentations. *American Journal of Enology and Viticulture***,** v. 50, p. 107-119, 1999.

BNDES; CGEE. *Bioetanol de cana-de-açúcar*: Energia para o desenvolvimento sustentável.1 ed. Rio de Janeiro, 2008.

Brasil. Ministério da Agricultura, Pecuária e Abastecimento. *Decreto no 6.871, de 04 de junho de 2009*. Dispõe sobre a padronização, a classificação, o registro, a inspeção, a produção e a fiscalização de bebidas. Diário Oficial da República Federativa do Brasil, Brasília, DF, 5 jun. 2009.

Brasil. Ministério de Agricultura, Pecuária e Abastecimento. *Instrução normativa nº 34, de 9 de novembro de 2012*. Complementa os padrões de identidade e qualidade para as bebidas fermentadas. Diário Oficial da República Federativa do Brasil, Brasília, DF, 9 nov. 2012.

CASTELL, Ana et al. Headspace with gas chromatography-mass spectrometry for the use of volatile organic compound profile in botanical origin authentication of honey. *Molecules*, v. 28, n. 11, p. 4297, 2023.

CECCATO-ANTONINI, Sandra Regina. Conventional and nonconventional strategies for controlling bacterial contamination in fuel ethanol fermentations. *World Journal of Microbiology and Biotechnology*, v. 34, n. 6, p. 80, 2018.

CHANDEL, Anuj Kumar et al. Economics and environmental impact of bioethanol production technologies: an appraisal. *Biotechnology and Molecular Biology Reviews*, v. 2, n. 1, p. 14-32, 2007.

CHENG, Ngoh Gek et al. Production of ethanol by fed-batch fermentation. *Pertanika Journal of Science and Technology*, v. 17, n. 2, p. 399-408, 2009.

CHITARRINI, G.; DEBIASI, L.; STUFFER, M.; UEBEREGGER, E.; ZEHETNER, E.; JAEGER, H.; ROBATSCHER, P.; CONTERNO, L. Volatile profile of flower honey fermented into mead and of mead with or without Ribes nigrum. *Molecules*, v. 25, n. 8, p. 1818, 2020.

DA SILVA, Priscila Missio et al. Honey: chemical composition, stability and authenticity. *Food Chemistry*, v. 196, p. 309-323, 2016.

DELLA-BIANCA, Bianca E. et al. Physiology of the fuel ethanol strain Saccharomyces cerevisiae PE-2 at low pH indicates a context-dependent performance relevant for industrial applications. *FEMS yeast research*, v. 14, n. 8, p. 1196-1205, 2014.

DENG, Y.; KAN, H.; LI, Y.; LIU, Y.; QIU, X**.** Analysis of volatile components in *Rosa roxburghii* Tratt. and *Rosa sterilis* using headspace–solid-phase microextraction–gas chromatography–mass spectrometry. *Molecules*, v. 28, n. 23, p. 7879, 2023.

GUPTA, J.K.; SHARMA, R. Production technology and quality characteristics of mead and fruit-honey wines: A review. *Natural Product Radiance*, v. 8, n. 4, p. 345-355, 2009.

HADIYANTO, H. et al. Batch and fed-batch fermentation system on ethanol production from whey using Kluyveromyces marxianus. *International Journal of Renewable Energy Development*, v. 2, n. 3, p. 127, 2013.

HAZELWOOD, L. A.; DARAN, J. M.; VAN MARIS, A. J.; PRONK, J. T.; DICKINSON, J. R. The Ehrlich pathway for fusel alcohol production: a century of research on Saccharomyces cerevisiae metabolism. *Applied and Environmental Microbiology*, v. 74, n. 8, p. 2259–2266, 2008.

HOHMANN, S.; MAGER, W.H. Yeast stress responses. Berlin Heidelberg: Springer-Verlag, 2003.

INGLEDEW, W. M. Alcohol production by Saccharomyces cerevisiae: A yeast primer. In: LYONS, T. P.; KELSALL, D. R. *The Alcohol Textbook*, 3. ed. Nottingham: Nottingham University Press, 1999. p. 49-87.

Ivanova, V., Petrova, P., Hristov, J. Application in the Ethanol Fermentation of Immobilized Yeast Cells in Matrix of Alginate/Magnetic nanoparticles, on Chitosan-Magnetite Microparticles and Cellulose-coated Magnetic nanoparticles**.** *International Review of Chemical Engineering*, vol. 3, n. 2, p. 289-299, 2011.

JACKSON, R. S. *Wine science: principles and applications*. 4 ed. San Diego: Academic Press, 2014. 978p.

JAIN, Anjali; CHAURASIA, Satyendra P. Bioethanol production in membrane bioreactor (MBR) system: a review. *International Journal of Environmental Research and Development*, v. 4, n. 4, p. 387-394, 2014.

LANE, J. H.; EYNON, L. *Determination of reducing sugars by Fehling's solution with methylene blue indicator*, Normam Rodge, London, 8p., 1934.

LIMA U.A.; BASSO L.C.; AMORIM H.V. Produção de etanol. In: AQUARONE U. A.; BORZANI E.; SCHMIDELL W. *Biotecnologia Industrial*: processos fermentativos e enzimáticos. 1 ed. São Paulo: Editora Edgard Blucher. 2001. 616p

LUCENA, B. T.; SANTOS, B. M. dos; MOREIRA, J. L. et al. Diversity of lactic acid bacteria in the bioethanol process. *BMC Microbiology*, v. 10, p. 298, 2010.

MADIGAN *et al*. Nutrition and metabolism. *Brock Biology of Microorganisms*. 9 ed. Saddle River: Prentice Hall, 2000. 992p.

MARGARITIS, Argyrios; MERCHANT, Fahar JA. The technology of anaerobic yeast growth. In: *Yeast Biotechnology*. Dordrecht: Springer Netherlands, 1987. p. 231-276.

McCONNELL, D. S.; SCHRAMM, K. D. Mead success: ingredients, processes and techniques. *Zymurgy*, v. 4, p. 33–39,1995

.

MILLER, G. L. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Analytical Chemistry*, v. 31, n. 3, p. 426 428, 1959.

MUNFORD, A.; CHAVES, R.; SANT'ANA, A. Cinética de inativação de bactérias de deterioração da cerveja (Lactobacillus brevis, Lactobacillus casei e Pediococcus damnosus) durante a lavagem ácida da levedura cervejeira. *Food Microbiology*, v. 91, p. 103513, 2020.

NASANIT, Rujikan et al. The assessment of epiphytic yeast diversity in sugarcane phyllosphere in Thailand by culture-independent method. *Fungal biology*, v. 119, n. 12, p. 1145-1157, 2015.

PARAPOULI, Maria et al. Saccharomyces cerevisiae and its industrial applications. *AIMS microbiology*, v. 6, n. 1, p. 1, 2020.

RIBEIRO-FILHO, Normando et al. Influence of essential inorganic elements on flavour formation during yeast fermentation. *Food Chemistry*, v. 361, p. 130025, 2021.

RIVERA, Elmer Ccopa et al. Effect of temperature on sugarcane ethanol fermentation: Kinetic modeling and validation under very-high-gravity fermentation conditions. *Biochemical engineering journal*, v. 119, p. 42-51, 2017.

RYU, D & WOLF-HALL, C. Yeasts and molds. In: SALFINGER, Y & TORTORELLO, ML. *Compedium of Methods for the Microbiological Examination of Foods***.** 5. ed. Washington, D.C.: American Public Health Association, 2015. p. 287- 297.

SEO, S.; PARK, S.; JUNG, S.; RYU, C.; KIM, J. Anticontamination strategies for yeast fermentation. *Microorganisms*, v. 8, 2020.

SKINNER, Kelly A.; LEATHERS, Timothy D. Bacterial contaminants of fuel ethanol production. *Journal of Industrial Microbiology and Biotechnology*, v. 31, n. 9, p. 401-408, 2004.

STAROWICZ, M.; GRANVOGL, M. An overview of mead production and the physicochemical, toxicological, and sensory characteristics of mead with a special emphasis on flavor. *Trends in Food Science & Technology*, v. 106, p. 402-416, 2020.

STAROWICZ, M.; GRANVOGL, M. Effect of Wort Boiling on Volatiles Formation and Sensory Properties of Mead. *Molecules*, v. 27, n. 710, 2022.

STONE, H.; SIDEL, J.; OLIVER, S.; WOOLSEY, A.; SINGLETON, R. C. Sensory evaluation by quantitative descriptive analysis. *Food Technology*, v. 28, p. 24-34, 1974.

THATOI, Hrudayanath et al. Bioethanol production from tuber crops using fermentation technology: a review. *International Journal of Sustainable Energy*, v. 35, n. 5, p. 443-468, 2016.

VASCONCELO, J. N. D. Fermentação Etanólica. In: SANTOS, F.; BORÉM, A.; CALDAS, C. *Cana-de-açucar*: bioenergia, açucar e etanol. 2 ed. Viçosa: Folha de Viçosa, Ltda, 2012, cap. 16. p. 451-487.

WHEALS, Alan E. et al. Fuel ethanol after 25 years. *Trends in biotechnology*, v. 17, n. 12, p. 482-487, 1999.

WHITE, C.; ZAINASHEFF, C. *Yeast***:** The Practical Guide to Beer Fermentation. Colorado: Brewers Publications, 2010.

WIETSTOCK, P. C. et al. Uptake and release of Ca, Cu, Fe, Mg, and Zn during beer production. *Journal of the American Society of Brewing Chemists*, v. 73, n. 2, p. 179-184, 2015.

ZHANG, J.; ZHANG, C.; DAI, L.; JIAN, D.; LIU, Y.; GUO, X.; XIAO, D. Effects of overexpression of the alcohol acetyltransferase-encoding gene ATF1 and disruption of the esterase-encoding gene IAH1 on the flavour profiles of Chinese yellow rice wine. International. *Journal of Food Science and Technology*, v. 47, p. 2590-2596, 2012.

ZHU, Min; SUN, Jing; ZHAO, Haoan; WU, Fanhua; XUE, Xiaofeng; WU, Liming; CAO, Wei. Volatile compounds of five types of unifloral honey in Northwest China: correlation with aroma and floral origin based on HS-SPME/GC–MS combined with chemometrics. *Food Chemistry*, v. 384, p. 132461, 2022.

SUPPLEMENTARY MATERIAL

Table S1: Concentration of volatile compounds (µg/mL) identified in meads fermented with different strains of Saccharomyces cerevisiae (C = commercial; TF and J = autochthonous).

Name	C (µg/mL)	TF (µg/mL)	J (µg/mL)	Class
Acetic acid	$0.218^{b} \pm 0.064$	nd	$1.782^{a} \pm 0.081$	Acid
Hexanoic acid	$0.0012^{a} \pm 0.0004$	nd	nd	Acid
n-Decanoic acid	$0.038^{b} \pm 0.013$	nd	$0.078^{a} \pm 0.011$	Acid
Undecanoic acid	$0.43^{a} \pm 0.12$	nd	nd	Acid
Octanoic acid	$0.085^{a} \pm 0.028$	$0.043^{b} \pm 0.005$	nd	Acid
Benzeneacetic acid, 4-methoxy-	$0.632^{a} \pm 0.164$	$0.38^{a} \pm 0.06$	$0.082^{b} \pm 0.013$	Acid
Icosapentaenoic acid	$0.0255^{a} \pm 0.0055$	$0.011^{b} \pm 0.004$	nd	Acid
10,12-Octadecadiynoic acid	$0.002^{b} \pm 0.001$	$0.017^{a} \pm 0.002$	$0.005^{b} \pm 0.002$	Acid
Butanoic acid, 3-hydroxy-	nd	$0.0057^{a} \pm 0.0001$	nd	Acid
Hydnocarpic acid	nd	nd	$0.118^{a} \pm 0.033$	Acid
4-Penten-2-ol	$3.155^{a} \pm 0.462$	nd	$0.377^{b} \pm 0.121$	Alcohol
1-Propanol, 2-methyl-	$0.0003^{b} \pm 0.0000$	$0.09^{a} \pm 0.03$	nd	Alcohol
1-Butanol, 3-methyl-	$8.827^{b} \pm 2.065$	$13.585^{a} \pm 1.895$	$5.123^{b} \pm 0.903$	Alcohol
2-Butanol, 3-methyl-	$0.0017^{a} \pm 0.0006$	nd	nd	Alcohol
1-Hexanol	$0.010^{a} \pm 0.001$	nd	nd	Alcohol
2-Heptanol	$0.014^{a} \pm 0.002$	nd	nd	Alcohol
1-Heptanol	$0.030^{a} \pm 0.003$	nd	nd	Alcohol
3-Octen-1-ol, (E)-	$0.0213^{a} \pm 0.0008$	nd	nd	Alcohol
2-Propyl-1-pentanol	$0.205^{a} \pm 0.038$	nd	nd	Alcohol
Ethanol	nd	$1.38^{a} \pm 0.46$	$1.99^{a} \pm 0.30$	Alcohol
2-Pentanol	nd	$0.0030^{a} \pm 0.0004$	nd	Alcohol
1-Butanol, 2-methyl-	nd	$1.741^{a} \pm 0.283$	nd	Alcohol
2,3-Butanediol	nd	$0.020^{a} \pm 0.006$	$0.016^{a} \pm 0.004$	Alcohol
Methylolacetone	nd	$0.0056^{a} \pm 0.0014$	nd	Alcohol
2-Propanol, 1-(1-methylethoxy)-	nd	$0.0007^{a} \pm 0.0002$	nd	Alcohol
3-Hepten-1-ol	nd	$0.0101^{a} \pm 0.0002$	nd	Alcohol
3-Heptanol, 3-methyl-	nd	$0.001^{a} \pm 0.001$	nd	Alcohol
1-Hexanol, 2-ethyl-	nd	$0.224^{a} \pm 0.017$	$0.196^{a} \pm 0.017$	Alcohol
exo-2,7,7-trimethylbicyclo[2.2.1]heptan-2-ol	nd	$0.008^{a} \pm 0.003$	nd	Alcohol
2-Hexanol	nd	nd	$0.007^{a} \pm 0.002$	Alcohol
4-Amino-1-butanol	nd	nd	$0.006^{a} \pm 0.001$	Alcohol
1,7-Octadiene-3,6-diol, 2,6-dimethyl-	nd	nd	$0.027^{a} \pm 0.007$	Alcohol

Dihydroeugenol	nd	nd	0.0132[a] ± 0.0045	Alcohol
ent-Germacra-4(15),5,10(14)-trien-1β-ol	nd	nd	0.0200[a] ± 0.0035	Alcohol
Phenylethyl Alcohol	8.004[b] ± 2.138	14.678[a] ± 3.054	0.679[c]± 0.011	Alcohol
2-Hexenal, 2-ethyl-	0.009[a] ± 0.002	nd	nd	Aldehyde
Benzeneacetaldehyde	0.020[b] ± 0.007	0.084[a] ± 0.003	0.021[b] ± 0.003	Aldehyde
3,5-Heptadienal, 2-ethylidene-6-methyl-	0.0775[a] ± 0.0016	0.062[b] ± 0.002	nd	Aldehyde
Safranal	nd	nd	0.047[a] ± 0.011	Aldehyde
β-Cyclocitral	nd	nd	0.100[a] ± 0.016	Aldehyde
Cyclopentaneacetaldehyde, 2-formyl-3-methyl-α-methylene-	nd	nd	0.01340[a] ± 0.00002	Aldehyde
Benzyl linoleate	0.0094[a] ± 0.0003	0.007[b] ± 0.001	0.0020[c] ±0.0001	Aromatic
Acetomesitylene	nd	0.0121[a] ± 0.0004	0.016[a] ± 0.003	Aromatic
o-Cymene	nd	nd	0.036[a] ± 0.001	Aromatic
2-Methylcoumaran	0.023[a] ± 0.008	nd	nd	Aromatic
Aceteugenol	0.0012[b] ± 0.0004	0.009[a] ± 0.001	0.010[a] ± 0.003	Aromatic
Methyleugenol	0.079[a] ± 0.004	0.044[b] ± 0.003	0.038[b] ± 0.005	Aromatic
Panaxydol	0.016[a] ± 0.004	0.006[b] ± 0.002	0.0045[b] ± 0.0003	Aromatic
Dodecanoic acid, 2-phenylethyl ester	0.011[a] ± 0.003	nd	nd	Ester
2,6,10,14-Hexadecatetraen-1-ol, 3,7,11,15-tetramethyl-, acetate, (E,E,E)-	0.025[a] ± 0.008	nd	nd	Ester
Ethyl Acetate	nd	0.722[a] ± 0.262	0.699[a] ± 0.126	Ester
Butanoic acid, 2-methyl-, ethyl ester	nd	0.011[a] ± 0.002	nd	Ester
Butanoic acid, 3-methyl-, ethyl ester	nd	0.027[a] ± 0.004	nd	Ester
Ethyl caprylate	nd	1.762[a] ± 0.276	0.081[b] ± 0.005	Ester
Nonanoic acid, ethyl ester	nd	0.039[a] ± 0.013	nd	Ester
Benzenepropanoic acid, ethyl ester	nd	0.155[a] ± 0.023	0.046[b] ± 0.001	Ester
Ethyl 9-decenoate	nd	0.234[a] ± 0.071	nd	Ester
Dodecanoic acid, ethyl ester	nd	0.030[a] ± 0.009	nd	Ester
Octanoic acid, decyl ester	nd	0.102[a] ± 0.001	nd	Ester
Hexanoic acid, tridec-2-ynyl ester	nd	0.066[a] ± 0.007	nd	Ester
Ergosta-5,22-dien-3-ol, acetate, (3β,22E)-	nd	0.005[a] ± 0.002	0.0038[a] ±0.0002	Ester

Propanoic acid, 2-phenylethyl ester	nd	nd	$0.044^{a} \pm 0.014$	Ester
Hexadecanoic acid, ethyl ester	nd	nd	$0.007^{a} \pm 0.001$	Ester
(E)-Valerenyl isovalerate	nd	nd	$0.010^{a} \pm 0.001$	Ester
cis-4,7,10,13,16,19-Docosahexaenoic acid, methyl ester	nd	nd	$0.0130^{a} \pm 0.0003$	Ester
Retinol, acetate	nd	nd	$0.0113^{a} \pm 0.0025$	Ester
Butanoic acid, ethyl ester	$0.009^{b} \pm 0.003$	$0.0165^{a} \pm 0.0009$	$0.006^{b} \pm 0.002$	Ester
1-Butanol, 3-methyl-, acetate	$0.078^{b} \pm 0.003$	$0.289^{a} \pm 0.022$	$0.258^{a} \pm 0.015$	Ester
1-Butanol, 2-methyl-, acetate	$0.012^{c} \pm 0.004$	$0.06^{b} \pm 0.01$	$0.089^{a} \pm 0.006$	Ester
Ethyl caproate	$0.3508^{b} \pm 0.0353$	$0.4195^{a} \pm 0.0160$	$0.0624^{c} \pm 0.0029$	Ester
Pentanoic acid, 2-hydroxy-4-methyl-, ethyl ester	$0.029^{b} \pm 0.003$	$0.074^{a} \pm 0.010$	nd	Ester
Ethyl caprate	$0.89^{a} \pm 0.30$	$0.34^{b} \pm 0.08$	$0.039^{b} \pm 0.003$	Ester
Ethyl α-toluate	$0.38^{a} \pm 0.12$	nd	$0.068^{b} \pm 0.009$	Ester
Phenethyl acetate	$32.65^{a} \pm 1.07$	$0.878^{c} \pm 0.168$	$6.3510^{b} \pm 0.7498$	Ester
Hexanoic acid, 2-phenylethyl ester	$0.3208^{a} \pm 0.1006$	nd	$0.014^{b} \pm 0.002$	Ester
Benzenepropanoic acid, hexyl ester	$0.0115^{a} \pm 0.0028$	nd	nd	Ester
Octanoic acid, 2-phenylethyl ester	$0.07^{a} \pm 0.02$	nd	nd	Ester
Benzeneacetic acid, 2-phenylethyl ester	$1.875^{a} \pm 0.008$	nd	nd	Ester
Decanoic acid, 2-phenylethyl ester	$0.183^{a} \pm 0.009$	nd	nd	Ester
(Z)-Ethyl heptadec-9-enoate	$0.136^{a} \pm 0.045$	nd	nd	Ester
1,3-Dioxolane, 2,4,5-trimethyl-	nd	$3.15^{a} \pm 0.10$	$2.565^{a} \pm 0.667$	Ether
Diethoxymethyl acetate	nd	$0.043^{a} \pm 0.014$	nd	Ether
Pentane, 1-(1-ethoxyethoxy)-	$0.0072^{b} \pm 0.0007$	$0.06^{a} \pm 0.02$	$0.019^{b} \pm 0.005$	Ether
2-t-Butyl-5-propyl-[1,3]dioxolan-4-one	$0.215^{a} \pm 0.038$	$0.010^{b} \pm 0.002$	nd	Ether
cis-5-Isopropenyl-2-methyl-2-vinyltetrahydrofuran	$0.087^{a} \pm 0.004$	$0.069^{a} \pm 0.022$	$0.0706^{a} \pm 0.0001$	Furan
Cyclodecane	$0.0019^{a} \pm 0.0002$	nd	nd	Hidrocarboneto
2-Heptanone, 3-methyl-	nd	$0.0024^{a} \pm 0.0005$	nd	Ketone
2-Nonanone	nd	$0.0215^{a} \pm 0.0019$	nd	Ketone
2-Heptanone	$0.025^{a} \pm 0.005$	nd	nd	Ketone
Cuminone	$0.033^{a} \pm 0.002$	nd	nd	Ketone

1-Propanamine, N,2-dimethyl-	0.263[a] ± 0.016	nd	nd	Nitrogen Compound
Ethanol, 2-Nitro-	1.104[a] ± 0.115	0.143[b] ± 0.048	0.993[a] ± 0.143	Nitrogen Compound
3-Oxo-7,8-dihydro-β-ionol	nd	0.026[a] ± 0.002	0.030[a] ± 0.006	Norisoprenoid
cis-Arbusculone	0.581[a] ± 0.021	0.42[b] ± 0.08	0.536[ab] ± 0.034	Norisoprenoid
4-Methyleneisophorone	nd	nd	0.070[a] ± 0.004	Norisoprenoide
Dehydrosabinene	nd	0.015[a] ± 0.005	nd	Terpene
trans-Sabinene hydrate	nd	0.162[a] ± 0.018	0.103[b] ± 0.015	Terpene
cis-Linalool oxide	nd	10.05[a] ± 2.28	9.5[a] ± 0.7	Terpene
Myrtenyl methyl ether	nd	0.0099[a] ± 0.0006	nd	Terpene
Isothujol	nd	0.017[a] ± 0.003	nd	Terpene
2,6-Dimethyl-3,5,7-octatriene-2-ol, ,E,E-	nd	0.0012[b] ± 0.0003	0.016[a] ± 0.002	Terpene
α-Limonene diepoxide	nd	0.173[a] ± 0.014	0.045[b] ± 0.004	Terpene
1,5,5-Trimethyl-6-methylene-cyclohexene	nd	0.017[a] ± 0.004	0.021[a] ± 0.005	Terpene
Caryophyllene	nd	0.037[a] ± 0.010	nd	Terpene
Caryophyllene oxide	nd	0.020[a] ± 0.005	0.007[b] ± 0.001	Terpene
trans-Sesquisabinene hydrate	nd	0.017[a] ± 0.001	0.019[a] ± 0.002	Terpene
2-Butenal, 2-methyl-4-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	nd	0.0008[b] ± 0.0003	0.018[a] ± 0.002	Terpene
Camphenol, 6	nd	nd	0.44[a] ± 0.07	Terpene
trans-p-Menth-2,8-dien-1-ol	nd	nd	0.003[a] ± 0.001	Terpene
Cosmene	nd	nd	0.044[a] ± 0.003	Terpene
α-Terpineol	nd	nd	0.035[a] ± 0.006	Terpene
Nopol	nd	nd	0.014[a] ± 0.002	Terpene
cis-β-Farnesene	nd	nd	0.134[a] ± 0.030	Terpene
α-Curcumene	nd	nd	0.053[a] ± 0.002	Terpene
β-Guaiene	nd	nd	0.012[a] ± 0.004	Terpene
Isoeugenol methyl ether	nd	nd	0.046[a] ± 0.012	Terpene
cis-α-Bisabolene	nd	nd	0.08[a] ± 0.03	Terpene
Nerolidol	nd	nd	0.087[a] ± 0.009	Terpene
Spathulenol	nd	nd	0.0051[a] ±0.0009	Terpene
Longipinocarveol, trans-	nd	nd	0.0021[a] ±0.0007	Terpene
trans-Z-α-Bisabolene epoxide	nd	nd	0.017[a] ± 0.003	Terpene
α-Bisabolol oxide B	nd	nd	0.0131[a]±0.0006	Terpene
Aromadendrene oxide-(1)	nd	nd	0.0104[a] ±0.0015	Terpene
Santalol, E-cis,epi-β-	nd	nd	0.013[a] ± 0.003	Terpene
Artemiseole	0.005[a] ± 0.002	nd	nd	Terpene

Linalool oxide A	$8.935^{a} \pm 2.158$	$4.796^{b} \pm 0.826$	$4.148^{b} \pm 0.194$	Terpene
Nerol oxide	$0.194^{a} \pm 0.014$	$0.090^{c} \pm 0.008$	$0.131^{b} \pm 0.014$	Terpene
8-Hydroxylinalool	$0.0028^{b} \pm 0.0004$	$0.06900^{a} \pm 0.00001$	$0.091^{a} \pm 0.016$	Terpene

Note. Values represent mean ± standard deviation (n = 3). Different letters in the same row indicate significant difference by Tukey's test ($p < 0.05$); nd = not detected.

Supplement - List of equations

$$\mu = \frac{\ln X_2 - \ln X_1}{t_2 - t_1} \quad (1)$$

$$t_d = \frac{\ln(2)}{\mu} \quad (2)$$

$$q_s = \frac{S_1 - S_2}{X \cdot \Delta t} \quad (3)$$

$$q_p = \frac{P_2 - P_1}{X \cdot \Delta t} \quad (4)$$

$$P_p = \frac{\Delta P}{\Delta t} \quad (5)$$

$$P_x = \frac{\Delta X}{\Delta t} \quad (6)$$

$$Y_{p/s} = \frac{P}{S} \quad (7)$$

$$Y_{p/x} = \frac{P}{X} \quad (8)$$

$$Y_{x/s} = \frac{X}{S} \quad (9)$$

$$Y_{CO_2/EtOH} = \frac{CO_2}{EtOH} \quad (10)$$

$$Y_{CO_2/s} = \frac{CO_2}{S} \quad (11)$$

$$Y_{CO_2/x} = \frac{CO_2}{X} \quad (12)$$

Specific Growth Rate (μ, h^{-1})
Doubling Time (td)
Substrate Consumption Rate (q_s, h^{-1})
Product Formation Rate (q_p, h^{-1})
Volumetric Productivity (P_p, g/L.h)
Biomass Productivity (P_x, g/L.h)
Product Yield per Substrate ($Y_{p/s}$, g/g)
Product Yield per Biomass ($Y_{p/x}$, g/g)
Biomass Yield per Substrate ($Y_{x/s}$, g/g)
CO_2 Yield per Ethanol ($Y_{CO2/EtOH}$, g/g)
CO_2 Yield per Substrate ($Y_{CO2/s}$, g/g)
CO_2 Yield per Biomass ($Y_{CO2/x}$, g/g)