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Article

Challanges and Opportunities in Sustainable Ethanol Production

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RESUMO

Saccharomyces cerevisiae desempenha papel central na produção de bioetanol devido à sua elevada eficiência na conversão de açúcares fermentescíveis em etanol. Durante o processo fermentativo industrial, grandes quantidades de biomassa de levedura são geradas, e parte desse excedente pode ser reutilizada por meio da fermentação endógena, contribuindo para maior sustentabilidade e eficiência no uso de recursos nos sistemas de produção de etanol. Este estudo descreve as características de linhagens industriais selecionadas e de panificação de *S. cerevisiae*, explora o conceito e a aplicabilidade da fermentação endógena na produção sustentável de etanol e avalia os efeitos do estresse térmico no desempenho das leveduras. Foram realizados experimentos de fermentação utilizando as linhagens Fleischmann® e Pedra-2 cultivadas em caldo de cana-de-açúcar sob diferentes condições fermentativas. Os resultados indicam que as usinas podem empregar tanto linhagens industriais selecionadas quanto leveduras de panificação, dependendo das estratégias operacionais e da disponibilidade local. Além disso, a reutilização da biomassa excedente por meio da fermentação endógena apresenta potenciais vantagens econômicas e ambientais, ao aumentar o rendimento de etanol e reduzir resíduos. No entanto, o desempenho das leveduras pode ser negativamente afetado por fatores de estresse, especialmente temperaturas elevadas, que podem reduzir a eficiência fermentativa e a viabilidade celular, evidenciando a importância do monitoramento e do controle adequado do processo para manter uma produção de bioetanol estável e sustentável.
Palavras-chave: *Saccharomyces cerevisiae*; fermentação endógena; fatores de estresse; viabilidade celular.

ABSTRACT

Saccharomyces cerevisiae plays a central role in bioethanol production due to its high efficiency in converting fermentable sugars into ethanol. During the industrial fermentation process, large amounts of yeast biomass are generated, and part of this surplus can be reused through endogenous fermentation, contributing to greater sustainability and resource efficiency in ethanol production systems. This study describes the characteristics of selected industrial and bakery strains of *S. cerevisiae*, explores the concept and applicability of endogenous fermentation in sustainable ethanol production, and evaluates the effects of heat stress on yeast performance. Fermentation experiments were conducted using Fleischmann® and Pedra-2 strains cultivated in sugarcane juice under different fermentation conditions. The results indicate that ethanol plants may employ either selected industrial strains or bakery yeasts depending on operational strategies and local availability. Furthermore, the reuse of surplus yeast biomass through endogenous fermentation presents potential economic and environmental advantages by increasing ethanol yield and reducing waste. However, yeast performance can be negatively affected by stress factors, particularly elevated temperatures, which may reduce fermentation efficiency and cellular viability, highlighting the importance of careful monitoring and process control to maintain stable and sustainable bioethanol production.
Keywords: *Saccharomyces cerevisiae*; endogenous fermentation; stress factors; cell viability.

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Introduction

The creation of the Programa Nacional do Álcool – PROALCOOL brought profound transformations to the Brazilian sugar and ethanol sector, significantly influencing the technological and productive organization of this agroindustrial chain. Implemented in the 1970s as a strategic response to the global oil crisis, the program stimulated the replacement of fossil fuels with renewable alternatives and promoted the large-scale expansion of ethanol production in Brazil. As a result, substantial investments were directed toward research, infrastructure, and technological innovation in the sugar energy industry. These investments contributed to improvements in agricultural management, genetic selection of sugarcane varieties, mechanization of harvesting, and the modernization of industrial fermentation systems, strengthening the technological basis of ethanol production in the country (Cursi et al. 2022).

The continuous technological development of the sector also resulted in significant improvements in industrial fermentation processes. Advances in process control, yeast management, and contamination prevention increased the efficiency and stability of ethanol production systems. These technological innovations occurred simultaneously with the expansion of sugarcane cultivation areas and the establishment of new industrial plants, consolidating Brazil as one of the world's leading producers of bioethanol. In this context, the success of fuel ethanol is strongly associated with the global demand for cleaner and renewable energy sources, which contribute to the integration of economic development with environmental sustainability and the reduction of greenhouse gas emissions (Vandenberghe et al. 2022).

In Brazil, sugarcane is the main raw material used for bioethanol production and plays a central role in the national renewable energy matrix. This crop belongs to the group of C4 plants, characterized by high photosynthetic efficiency and an elevated capacity for carbon fixation, allowing efficient biomass accumulation under tropical climatic conditions. Sugarcane also presents desirable agronomic characteristics such as high productivity, significant culm yield, and high concentrations of sucrose in the stalks, which is the primary substrate used during alcoholic fermentation. The sucrose present in sugarcane is easily assimilated by yeasts during fermentation, especially by strains of the genus *Saccharomyces*, which are selected for their high fermentative capacity, metabolic efficiency, and tolerance to ethanol accumulation. These characteristics make *Saccharomyces cerevisiae* the most widely used microorganism in industrial ethanol production processes (Santos et al. 2018; Huang et al. 2020; Parapouli et al. 2020; Naghshbandi et al. 2019).

Certainly, the fermentative route is considered the most efficient pathway for ethanol production because it combines relatively low operational costs with high productivity and scalability. This process is widely applied in the bioethanol industry, particularly in sugarcane-based systems, due to the efficiency with which microorganisms convert fermentable sugars into ethanol. In industrial fermentation, the metabolic capacity of microorganisms allows the rapid transformation of substrates such as sucrose, glucose, and fructose into ethanol and carbon dioxide, making this route economically viable for large-scale production. In addition to its economic advantages, the fermentative pathway also presents environmental benefits, since it relies on renewable raw materials and contributes to the reduction of dependence on fossil fuels. These characteristics reinforce the relevance of fermentation technologies within the global context of renewable energy and bioeconomy development (Shenbagamuthuraman et al. 2022).

The ethanol production process involves several operational stages designed to ensure high fermentation efficiency and process stability. Initially, fermentation vats are supplied with the appropriate substrate, followed by the inoculation of selected yeasts with high bioconversion capacity responsible for converting sugars into ethanol through alcoholic fermentation. After the fermentation stage, yeast cells are separated from the fermented broth by centrifugation, allowing the recovery of viable biomass that can be reused in subsequent fermentation cycles. This recovered biomass is subjected to an acid treatment aimed at minimizing microbial

contamination before being returned to the fermentation tanks. However, only part of this biomass is reused, since excess yeast is continuously generated during the process. Some studies suggest the reuse of this residual yeast biomass as an additive in animal feed due to its high nutritional value and protein content, while another portion is still discarded, highlighting the need for strategies that promote better valorization of this byproduct within ethanol production systems (Nasiri et al. 2019; Rachwał et al. 2020).

The fermentative environment is extremely hostile to yeasts (Walker & Basso 2020), which are intensely exposed to variations in pH, contaminating agents and other stress factors, which intersperse and cause changes in the metabolism and physiology of cells, influencing the formation of undesirable metabolites and fermentative efficiency (Coertjens et al. 2023). The ability of these microorganisms to deal with these challenges is essential for the efficiency and success of the industrial process, especially on a larger scale such as the production of biofuels (Cavelius et al. 2023; Vickram et al., 2023). Batistote and Santos (2020), report that *S. cerevisiae* yeasts are excellent raw material converters and can be used to produce secondary compounds, since they have distinct metabolic routes that are activated due to the intensity and synergism of stress factors, triggering several intracellular responses, including the activation of signaling pathways. According to Lin et al. (2022), the function of signaling pathways is to recognize changes occurring in the environment and transmit signals from the cell surface to the nucleus.

Among the response mechanisms, there is the production of reserve carbohydrates, a yeast resource used to meet the cells' need for nutrients in adverse conditions, and is also a protective mechanism (Elbakush & Güven 2021). According to these authors, yeasts have as a tool the ability to store intracellular carbohydrates for its maintenance in the environment due to the scarcity of substrate in the fermentation medium. In which carbohydrates are used as food for yeast, in the process of endogenous fermentation, a procedure in which cells convert the reserve carbohydrates present in their intracellular environment into energy to maintain the integrity of the cell, also acting in the cells' self-protection, the main carbohydrates produced are trehalose and glycogen.

According to Sharma et al. (2023), trehalose is a non-reducing disaccharide composed of two D-glucose residues joined by a bond (α-1,1), and its function is to prevent the denaturation of proteins, as it acts as an osmoregulatory of the cell membrane, this is because in extreme conditions in the presence of high concentrations of ethanol, the cell membrane undergoes changes in relation to intracellular and extracellular flow (Eleutherio et al. 2015; Auesukaree 2017). Glycogen is a polymer that contains glucose molecules linked by glycosidic bonds (α-1,4) and, at the branching sites, glycosidic (α-1,6) (Gangoiti et al. 2020). The formation of glycogen allows the accumulation of glucose in cells without increasing osmotic pressure (Betlej et al. 2020), providing the yeast with conditions to maintain itself in the face of a shortage of food in the external fermentation.

Despite the technological advances achieved in the Brazilian sugar energy sector, important challenges remain regarding the optimization of fermentation performance under industrial stress conditions. Although several studies have described the physiological responses of *S. cerevisiae* to isolated stress factors, there is still limited understanding of the comparative behavior of industrial and commercial strains under combined thermal and ethanolic stress conditions that closely resemble industrial fermentation environments. This gap is particularly relevant because strain-specific differences directly affect cell viability, substrate conversion efficiency, ethanol yield, and reserve metabolite accumulation, all of which have direct implications for process stability and operational costs. Reduced fermentative performance may increase production costs through lower ethanol productivity, higher yeast replacement rates, and losses associated with process interruptions or reduced biomass reutilization. Therefore, understanding the physiological and metabolic responses of different strains under stressful conditions is essential not only from a microbiological perspective, but also for improving the economic sustainability and efficiency of sugarcane-based bioethanol production systems.

The sugar and alcohol sector has increasingly sought sustainable production processes from both economic and environmental perspectives, making research aimed at improving process efficiency highly relevant. In this context, the valorization of by-products generated during ethanol production represents an important strategy to enhance sustainability and productivity. It is estimated that, for every liter of ethanol produced, approximately 30 g of surplus yeast biomass are generated, which is often considered a residual by-product (O Presente Rural, 2019). Thus, this study aimed to evaluate the physiological performance, reserve carbohydrate production, and stress tolerance of different *Saccharomyces cerevisiae* strains under thermal and ethanolic stress conditions, in order to identify strains with greater fermentative resilience and potential to improve process efficiency and economic sustainability in sugarcane-based ethanol production.

Methodology

Location of study development

The study was developed in the Laboratório de Biotecnologia, Bioquímica e Biotransformação do Centro de Estudos em Recursos Naturais – CERNA, in Universidade Estadual do Mato Grosso do Sul-Dourados/MS.

Main characteristics of baker's and selected yeast strains and types of fermentation

Initially, a preliminary bibliographic survey was conducted to identify the main characteristics of baker's yeast and selected industrial yeast strains commonly used in ethanol production systems. Subsequently, a broad exploratory, quantitative, and descriptive search was performed in the Google Scholar database without time restriction, considering only open-access articles, technical reports, and scientific documents relevant to the study topic.

The search strategy included combinations of the keywords: "*Saccharomyces cerevisiae*", "ethanol fermentation", "baker's yeast", "industrial yeast", "thermal stress", "ethanol stress", and "fermentative performance". The retrieved studies were screened based on title, abstract, and methodological relevance. Duplicate and non-relevant studies were excluded. The selected studies were organized according to strain characteristics, fermentation conditions, stress tolerance, biomass production, and by-product generation. Data were compiled and descriptively analyzed using Excel 2019® software to support the experimental design and the development of the study flowchart (Figure 1). This scientometric approach provided the theoretical basis for selecting the strains and defining the fermentative conditions adopted in this research.

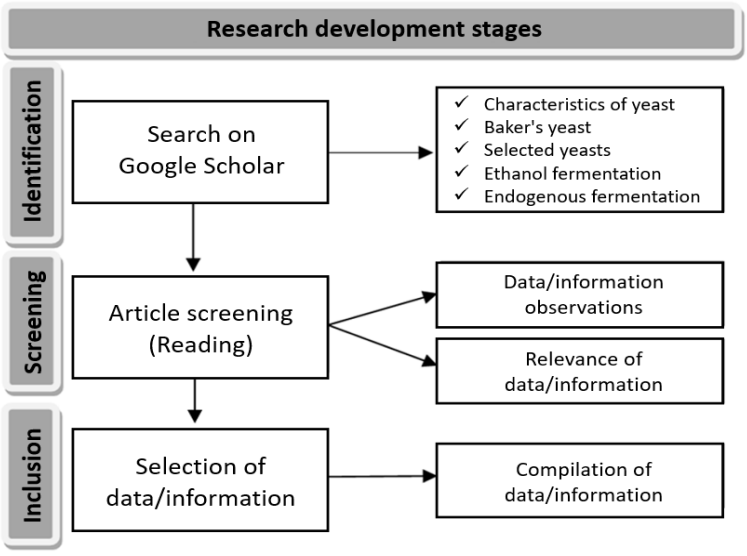


Figure 1. Flowchart of the steps adopted to develop this research. Source: Authors (2026).

Microorganisms used

The yeasts used in this study were *Saccharomyces cerevisiae* Fleischmann® from bakery products purchased in local stores and the selected strain Pedra-2 purchased from the company LNF Biotecnologia Aplicada, located in Bento Gonçalves – RS, Brazil.

Pre-inoculum and fermentative conditions

A pre-inoculum was prepared using 0.10 g of lyophilized yeast from each strain, suspended in 1.0 mL of sterile saline solution (0.85%) and homogenized. Subsequently, aliquots were inoculated onto previously prepared Petri dishes containing Sabouraud Agar medium and incubated at 30 °C for 48 h to ensure cell activation and purity. After incubation, isolated colonies were collected using a sterile platinum loop and transferred into sterile test tubes containing 20 mL of sugarcane juice adjusted to 22 °Brix and pH 5.0. The fermentations were carried out under orbital shaking at 200 rpm at three temperatures (30, 35, and 40 °C). Samples were collected after 8 and 24 h of fermentation for biomass growth and cell viability analyses. All experiments were performed in triplicate for each treatment condition.

Cell growth by biomass

Cell growth was estimated by spectrophotometric readings at 570 nm using a UV–Vis spectrophotometer, based on a previously established calibration curve according to Batistote et al. (2010). Biomass concentration was expressed as $mg \cdot mL^{-1}$. Measurements were performed in triplicate for each sample.

Cell viability

Cell viability was determined by methylene blue staining followed by cell counting in a Neubauer chamber, according to the methodology described by Lee et al. (1981). Viable and non-viable cells were quantified, and viability was expressed as percentage of live cells relative to total counted cells. All analyses were performed in triplicate.

Data analysis

The experimental data were expressed as mean ± standard deviation. Descriptive analyses were performed using Excel 2019® software in order to organize, compile, and interpret the results related to biomass growth and cell viability under the different fermentative conditions evaluated. All experiments were conducted in triplicate, and the results were presented comparatively to allow evaluation of the behavior of different Saccharomyces cerevisiae strains under the different fermentation temperatures and times.

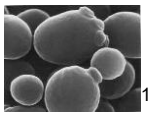
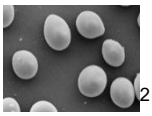
Results and Discussion

Important differences were observed between baker's and selected Saccharomyces cerevisiae strains regarding fermentative behavior, persistence in the process, and operational performance. Baker's yeast showed greater budding potential during the initial fermentation stages, which may favor rapid biomass establishment and early occupation of the fermentative medium. In contrast, the selected strain Pedra-2 showed greater permanence and dominance throughout the fermentative process, indicating higher robustness and better adaptation to industrial conditions. These differences are particularly relevant because cell persistence and physiological stability directly influence process continuity, ethanol yield, and the frequency of biomass replacement during production cycles.

From an economic and operational perspective, an important balance was observed between acquisition cost and fermentative efficiency. Although selected strains have a substantially higher purchase cost

(approximately R$ 92.00 kg^{-1}) compared with baker's yeast (R$ 21.00 kg^{-1}), their greater tolerance to thermal and ethanolic stress, longer permanence in the fermentative medium, and higher ethanol productivity may compensate for the initial investment. In addition, fermentation rate, estimated by carbon dioxide release, is an important parameter to assess strain performance. The selected strain reached approximately 8.5 g CO_2 after 3 h of fermentation, whereas baker's yeast reached around 7.9 g CO_2 under similar conditions. Although this difference may appear relatively small in short-term assays, even modest gains in fermentative speed and stability can significantly improve ethanol productivity, process consistency, and industrial efficiency when scaled up. These quantitative and operational differences reinforce the relevance of strain selection in industrial bioethanol production (Table 1).

Table 1. Fermentative characteristics related to yeasts Fleischmann® and Pedra-2.

Characteristics	Baker's yeast	Selected yeasts
Colonies	 1	 2
Budding	[2]More efficient in the first hours of fermentation	[2]Low budding at the beginning of the fermentation process
Permanence and dominance	[2]Low predominance in the last hours of fermentation	[2]More dominant in the last hours of fermentation
Ethanol yield	[3]Medium	[3]High
Quantity used (%)	[3]80	[3]20
Value (R$ kg^{-1})	[3]21.00	[3]92.00
Fermentation speed 3h (g de CO_2)	[3]7.90	[3]8.5
Other features	[3]Susceptible to temperature fluctuations and high levels of ethanol in the medium	[3]High tolerance to temperature fluctuations, tolerance to high ethanol content in the medium

Source: Adapted by the authors of [1]Bernstein & Bernstein (2016); [2]Fermentec (2024); [3]Latino Americana Biotecnologia Aplicada (LFN, 2024).

These observations are consistent with previous studies demonstrating that baker's and selected Saccharomyces cerevisiae strains play complementary roles in industrial bioethanol production. According to the literature, baker's yeast generally exhibits high budding rates during the initial stages of fermentation, which favors rapid biomass establishment and contributes to the maintenance of cell concentration at the beginning of the process. However, its lower persistence and reduced tolerance to industrial stress conditions may limit its performance over prolonged fermentation cycles. In contrast, selected industrial strains present important technological advantages, including higher ethanol yield, greater cell viability, increased tolerance to ethanol accumulation and temperature fluctuations, and improved resistance to multiple stress factors commonly found in industrial plants. These characteristics allow selected strains to adapt more efficiently to the fermentative environment, becoming more robust and maintaining higher productivity throughout the process (Favaro et al., 2019; Adebami et al., 2022; Zhou et al., 2021).

Yeasts play a fundamental role in bioethanol production and must present key physiological characteristics, such as tolerance to elevated temperatures, resistance to high ethanol concentrations, the ability to withstand pH fluctuations, and, above all, the capacity to maximize ethanol production in the shortest possible time. Therefore, process monitoring and the evaluation of yeast physiological responses are essential to ensure

fermentative stability and industrial efficiency. In this context, the strategic selection of yeast strains, including the use of mixed inoculum approaches, represents an important tool to optimize biomass establishment, process performance, and operational reliability, contributing to the competitiveness and sustainability of the sugar–energy sector (Moreira et al., 2015; Olsson et al., 2022; Bernstein & Bernstein, 2016; Fermentec, 2024; Latino Americana Biotecnologia Aplicada – LNF, 2024).

S. cerevisiae have a versatile metabolism, which makes this microorganism a precious pearl for biotechnological processes, because even in the scarcity or abundance of nutrients it is capable of metabolizing compounds. Its metabolism allows constant adjustments, as the difference between ethanolic fermentation and endogenous fermentation is directly related to how the yeast metabolizes the substrate. In ethanolic fermentation, fermentable sugars from the external environment are used, while endogenous fermentation is a yeast response to the scarcity of sugars in the environment, using the degradation of reserve carbohydrates present in the intracellular environment as an escape. These metabolic conditions are fundamental in terms of stability for the industrial process and understanding these yeast metabolic processes in the face of changes in nutrients is essential to maintain cellular integrity and maintenance of metabolite (Figure 2).

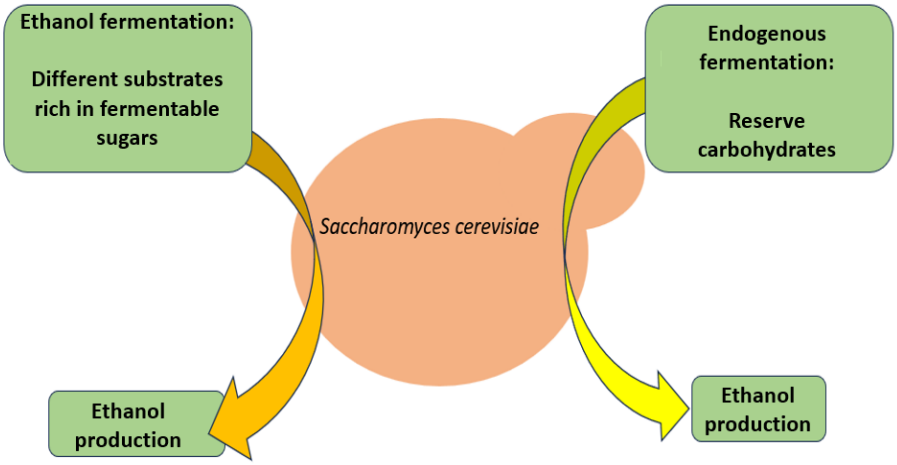


Figure 2. Types of fermentation: Ethanolic and endogenous. Source: Authors (2026).

In fermentation processes, temperature and time parameters are important factors to be considered between the two fermentation options. The ideal temperature for ethanolic fermentation is between 30 °C to 33 °C with a time between 8 hours to 10 hours, while for endogenous fermentation the temperature is higher, between 35 °C to 40 °C, with a longer time between 10 to 20 hours of process. The pH variation can be considered, as it presents a wide range between the fermentations analyzed. Regarding ethanol production, in ethanolic fermentation at the end of the process values around 8.5% of ethanol ($v.v^{-1}$) are recovered, whereas in endogenous fermentation 150 L of ethanol can be obtained per tons of yeast (Table 2). These data suggest that endogenous fermentation may be viable to increase ethanol production.

The metabolic processes of ethanolic and endogenous fermentation, used by yeasts to produce ethanol, in the face of the adverse conditions of the production process, show how versatile these microorganisms are with a high substrate conversion. *S. cerevisiae* is excellent in their ability to transform and produce numerous value-added molecules, as well as other molecules that can be explored. The metabolic efficiency of yeasts is important, as it can ensure fermentative. efficiency and the production of functional molecules for cell maintenance (Betlej et al. 2020; Mascarenhas et al. 2022).

Among the adverse conditions of the process are the stress factors in the fermentative environment that act effectively on yeast metabolism, causing loss of vitality and cell viability, as well as the production of metabolites. Adverse environmental conditions such as constant and extensive thermal and osmotic stress can

lead to low metabolite production and even cell death (Figure 3). Possibly the adjustment in metabolism enables yeast resistance to adverse disturbances in the environment, aiming to preserve cellular integrity and promote the production of metabolites, keeping the process stable.

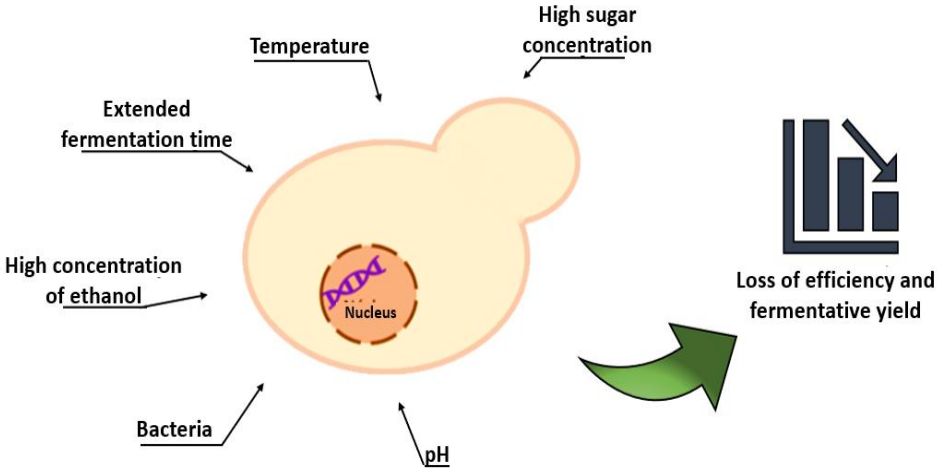


Figure 3. Stress factors that interfere with the fermentative efficiency and yield of *Saccharomyces cerevisiae*. Source: Authors (2026).

Obtaining maximum yields from an ethanol fermentation remains a major technical challenge, as stress factors cause negative impacts on the process. During fermentation, yeasts are intensely exposed to selective pressures such as high concentrations of ethanol (Lairón-Peris et al. 2021), osmotic stress, thermal instability, pH, contamination, prolonged fermentation time and others, which intersect and cause changes in the metabolism and physiology of cells, influencing the formation of undesirable metabolites and fermentative efficiency (Coertjens et al. 2023).

A relevant example occurs in the presence of high concentrations of ethanol in the fermentation medium. Although ethanol is a metabolite naturally produced by yeast, when it reaches high levels in the environment it causes damage to the cell membrane of the yeast itself. This stress factor, in conjunction with high temperatures, known as heat stress, triggers a response in yeast cells, resulting in the production and accumulation of trehalose and glycogen in the intracellular medium. These substances perform a fundamental role in stabilizing the plasma membrane of cells, as highlighted by Ribeiro et al. (2022) and Salas-Navarrete et al. (2023).

Faced with this challenging context, the sugar and alcohol industries adopt strategies to overcome the diversity of the fermentation. A common practice is to use a yeast mix to start the process. This mix typically consists of adding a baker's yeast strain, characterized by its high budding capacity, combined with selected strains that demonstrate greater adaptation to the industrial environment. The selected yeasts stand out for presenting a more robust profile, exhibiting greater resistance to the various stress factors associated with the process.

In this study, it was observed that biomass production and cell viability, under the influence of thermal stress and prolonged fermentation time, showed variations between Fleischmann® baker's yeast and Pedra-2 selected yeast strain (Table 3). Growth differences between yeasts were evident both in relation to temperature and fermentation time. When the yeasts were cultivated at a temperature of 30 °C for 8 hours of fermentation, Fleischmann yeast presented a biomass of 6.86 $mg.mL^{-1}$, with a viability of 92.33%, while Pedra-2 exhibited 8.10 $mg.mL^{-1}$ of biomass and viability around 97%. After 24 hours, under these same conditions, biomass values increased to 10.63 $mg.mL^{-1}$ and 12.76 $mg.mL^{-1}$ for Fleischmann and Pedra-2, respectively. However, viability showed a drop, reaching 81.43% for Fleischmann® and 82.32% for Pedra-2. When subjecting these

yeasts to a temperature of 40 °C, it was observed that there was a reduction in the analyzed parameters. After 8 hours of fermentation, biomass values were 7.53 mg.mL^{-1} for Fleischmann® and 9.23 mg.mL^{-1} for Pedra-2, with a slight drop in viability, which was, on average, 72.67% and 75% respectively. After 24 hours, biomass values increased to 9.28 mg.mL^{-1} and 10.62 mg.mL^{-1}, for Fleischmann and Pedra-2, respectively. However, there was a significant drop in viability, reaching 56% for Fleischmann® and 60% for Pedra-2. The data obtained indicate that, under conditions of higher temperatures and longer fermentation times, the yeasts analyzed showed a lower tolerance to thermal stress, resulting in a marked loss of viability and cellular development of the yeasts in question.

Table 3. Cell growth and viability of the Fleischmann® and Pedra-2 strains during 8 and 24 hours of fermentation at temperatures 30, 35 and 40°C.

		Fleischmann®		Pedra-2	
Temperature (°C)	**Time (h)**	**Biomass (mg.mL^{-1})**	**Viability (%)**	**Biomass (mg.mL^{-1})**	**Viability (%)**
30	8	6.86 ± 1.25	92.33 ± 3.79	8.10 ± 1.97	97.00 ± 3.06
	24	9.93 ± 1.08	81.43 ± 5.86	11.55 ± 1.68	82.32 ± 4.93
35	8	7.56 ± 0.64	88.36 ± 1.15	9.76 ± 1.27	91.34 ± 3.06
	24	9.41 ± 1.41	79.70 ± 1.58	11.86 ± 1.52	80.00 ± 6.92
40	8	7.53 ± 0.76	72.67 ± 4.51	9.23 ± 1.45	75.00 ± 2.00
	24	9.28 ± 0.55	56.00 ± 2.65	10.62 ± 1.72	60.05 ± 2.65

Source: Prepared by the authors.

Temperature and time parameters are important factors to be considered in fermentation processes, as they can cause stress to yeasts. When there is no interference from stress factors, it is expected that the rate of living yeast cells will be greater than 90%. However, it is important to highlight that the presence of stress factors can significantly affect yeast viability. In such conditions, the live cell index can reach 70% or less (Batistote & Santos 2020). Industrial yeast strains S. cerevisiae used in fermentation in ethanol plants exhibit an optimal growth range ranging from 29 to 34 °C (Lip et al. 2020). According to Caspeta & Nielsen (2015), higher temperatures have the potential to negatively impact yeast cell development. García et al. (2016), cell viability is intrinsically linked to the ability of yeast to adapt to the fermentation environment. This process triggers a transcriptional response that, in turn, affects the ability of these yeasts to confront stress conditions, such as temperature variations.

The success of fermentation is intrinsically linked to the ability of yeast to face the various stress factors that arise during this process. Thus, the ability that yeasts acquire to tolerate different types of stress, such as thermal stress, emerges as one of the main characteristics to ensure the integrity and viability of cells (Attfield 2023). Furthermore, the viability rate is an essential parameter to be monitored, as it supports the effective density of cells in the process, as highlighted Coertjens et al. (2022). In this sense, the importance of monitoring procedures for yeast viability in the fermentation process stands out. The ideal temperature conditions, the complex relationship between cell viability and environmental adaptation, are considered essential elements to optimize the performance of yeasts and, consequently, the fermentation process itself. Controlling these factors can boost the sustainability and efficiency of the ethanol production process in a sustainable way.

Final Considerations

This study demonstrated that the fermentative performance of baker's and selected Saccharomyces cerevisiae strains differs substantially under the evaluated thermal conditions, directly influencing biomass growth, cell viability, and process stability. The results showed that baker's yeast favored rapid initial biomass establishment due to its higher budding capacity, whereas the selected strain Pedra-2 showed greater persistence, higher tolerance to thermal stress, and better fermentative stability over time. These findings reinforce that strain-specific physiological behavior is a decisive factor for maintaining industrial fermentation efficiency, especially under stressful process conditions.

From an operational and economic perspective, the results indicate that although selected strains have higher acquisition costs, their greater robustness, stability, and fermentative efficiency may compensate for the initial investment by reducing biomass replacement frequency and improving ethanol productivity throughout industrial cycles. In addition, the comparative analysis of fermentation rates and strain behavior highlights the importance of mixed inoculum strategies as a viable approach to balance rapid biomass establishment and long-term process performance. Thus, the main contribution of this study lies in integrating physiological performance, fermentative behavior, and operational implications to support more efficient strain selection in sugarcane-based ethanol production systems.

Furthermore, this study reinforces the relevance of endogenous fermentation as a complementary strategy for the valorization of surplus yeast biomass generated during industrial ethanol production. The literature indicates that reserve carbohydrate metabolism enables yeasts to maintain cellular integrity under adverse conditions and may allow additional ethanol recovery from residual biomass. In this context, the present study contributes by highlighting the relationship between stress tolerance, metabolic adaptability, and the potential for improving process sustainability. Therefore, the findings provide practical and scientific support for optimizing industrial fermentation systems, reducing waste generation, and improving the technical, economic, and environmental sustainability of large-scale bioethanol production.

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