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Research Article

Impact of Substrate Variation on Yeast Biomass Yield and Proximate Composition for Selected Protein Supplement

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ABSTRACT

The rising cost of conventional livestock feed and reliance on imported ingredients have prompted the exploration of alternative protein sources such as yeast (*Saccharomyces cerevisiae*) as a probiotic feed additive. This study aimed to evaluate the nutritional composition of yeast and determine the effects of different substrates and fermentation durations on dry matter (DM) yield to identify optimal conditions for feed application. A laboratory experiment was conducted using dry yeast cultured in three substrates dry malt extract, molasses, and sugar at concentrations of 5%, 10%, and 15% over six days, with triplicate samples. Yeast growth was assessed through DM yield, while proximate analysis measured crude protein (CP), crude fibre (CF), crude lipid (CL), and metabolizable energy (ME). Results showed that dry malt extract significantly enhanced yeast growth compared to molasses and sugar, with the highest DM yield observed at 10% concentration after six days (6.65±0.25 g), whereas 15% concentration reduced growth due to osmotic stress. Proximate analysis indicated that control yeast had the highest CP (45.65%), while 5% malt extract produced relatively high CP (38.60%) with acceptable ME (14.78 MJ/kg). Increasing substrate concentration led to decreased CP and CF but increased CL content. Overall, fermentation slightly reduced energy values compared to control. In conclusion, yeast cultured in 5% dry malt extract exhibited the most balanced nutritional profile and growth performance, indicating its strong potential as a cost-effective, sustainable protein supplement for ruminant feed formulation in Malaysia.

Keywords: Yeast, Protein, Supplement, Fermentation, Feed

INTRODUCTION

The nutritional adequacy of livestock feed is a fundamental factor influencing animal health, productivity, and overall performance in the ruminant industry (Beauchemin et al., 2006). Feed additives, particularly probiotics, have been widely recognized for their role in improving feed efficiency, enhancing gut microbial balance, and promoting animal growth (Fanelli, 2012). Among these, yeast (*Saccharomyces cerevisiae*) has gained considerable attention due to its ability to modify rumen fermentation, increase fibre digestibility, and improve feed intake, weight gain, and milk production (Quigley, 2011). Previous studies have demonstrated that probiotic supplementation can enhance crude protein availability and energy digestibility, while also serving as a potential alternative to antibiotics in livestock production systems (Georg, 2005). In addition, yeast is a nutritionally rich, single-celled microorganism that can be cultivated on various

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agro-industrial by-products, making it a cost-effective and sustainable feed ingredient (McDonald et al., 2010).

Despite these advantages, the Malaysian livestock industry continues to face challenges related to high feed costs, heavy reliance on imported feed ingredients, and limited availability of high-quality feed resources thus probiotic can act as protein potential additive (Krehbiel et al., 2003). Conventional protein sources such as soybean meal and maize are expensive and compete with human food demands, thereby increasing production costs. Although yeast has shown promise as an alternative protein source, there is limited information on optimizing its production using locally available substrates and understanding how fermentation conditions influence its nutritional quality.

Therefore, the objectives of this study were to evaluate the nutrient composition of yeast as a feed additive and to determine the effects of different substrates (dry malt extract, molasses, and sugar) and fermentation durations on dry matter yield. This study also aimed to identify the most suitable substrate concentration for maximizing yeast growth and nutritional value, thereby contributing to improved feed formulation strategies.

The significance of this research lies in its potential to provide a sustainable and cost-effective alternative protein source for ruminant feed. By utilizing locally available substrates for yeast production, this study supports efforts to reduce dependence on imported feed ingredients and lower production costs. Furthermore, the findings contribute to the advancement of animal nutrition and feed technology by offering practical insights into optimizing probiotic use in livestock production systems. Similarly, a study by McPeake et al. (2002) showed an increase of dry matter intake among the treatment group. Thus, it revealed that probiotic can provide beneficial production rate in ruminant growth.

MATERIAL AND METHODS

This study employed a completely randomized experimental design to evaluate the effects of substrate type, concentration, and fermentation duration on yeast growth and nutritional composition. Three substrates (dry malt extract, molasses, and sugar) were tested at three concentrations (5%, 10%, and 15%) over a six-day fermentation period. Each treatment was conducted in triplicate, and sampling was performed at daily intervals (day 0 to day 6) to assess changes in dry matter (DM) yield. Subsequently, proximate analysis was carried out on selected samples to determine their nutritional composition.

Commercial dry yeast (Mauri-pan brand) was procured from a local bakery supplier in Kedah, Malaysia. Substrates used for fermentation included dry malt extract, granulated sugar, and molasses. Distilled water was used for media preparation and yeast rehydration. Laboratory equipment included sterilized containers, hot plates, round-bottom flasks, filtration apparatus, drying oven (set at 56°C), analytical balance, and standard laboratory glassware. Proximate analyses were conducted using standard equipment for digestion, distillation, extraction, and drying procedures in accordance with established laboratory protocols.

Approximately 180 g of dry yeast was divided into replicates (60 g each) and rehydrated in warm distilled water (35-40°C) for 5-10 minutes until active fermentation was indicated by bubble formation. Fermentation media were prepared by dissolving 0.5 g, 1.0 g, and 1.5 g of each substrate in 10 mL distilled water to obtain 5%, 10%, and 15% concentrations, respectively. The solutions were heated for 15 minutes, cooled, and inoculated with 2 g of activated yeast. The mixtures were allowed to ferment under controlled laboratory conditions for up to six days.

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Sampling was conducted daily by harvesting one flask per replicate. The fermented mixture was processed by removing the liquid fraction, isolating the yeast biomass, and drying it in an oven at 56°C until constant weight was achieved. The dried samples were weighed to determine DM yield.

Proximate analysis was conducted on yeast samples obtained from fermentation using dry malt extract as the selected substrate. Parameters analyzed included crude protein (CP), crude fibre (CF), crude lipid (CL), and metabolizable energy (ME), following standard procedures described in AOAC methods. These analyses involved digestion, distillation, titration, extraction, and drying processes to quantify nutrient composition.

Statistical Analysis:

All data were analyzed using SPSS Statistics (Version 17.0, SPSS Inc., Chicago, IL, USA). Prior to analysis, data were tested for normality using the Shapiro–Wilk test. A one-way analysis of variance (ANOVA) was performed to assess significant differences among treatment means. Where significant effects were detected, Tukey–Kramer's Honestly Significant Difference (HSD) test was applied for multiple comparisons. Statistical significance was considered at $p<0.05$.

RESULTS

The results of this study clearly demonstrate that both substrate type and concentration, as well as fermentation duration, significantly influence the growth performance and nutritional composition of yeast (*Saccharomyces cerevisiae*). Among the substrates evaluated, dry malt extract consistently supported superior yeast growth, yielding higher dry matter (DM) production compared to molasses and sugar media. Optimal growth was observed at 10% concentration of dry malt extract, particularly at extended fermentation periods (up to six days), while higher concentration (15%) resulted in reduced growth performance, likely due to osmotic stress effects. In contrast, molasses and sugar media exhibited comparatively lower and inconsistent DM yields, indicating limited nutrient availability for sustained yeast proliferation.

Table 1 Duration of fermentation and weight of yeast produced in dry malt extract.

Fermentation duration (Days)	Dry Malt Extract		
	5 % (g)	10 % (g)	15 % (g)
1 Day	1.92±0.18 [a]	-	-
2 Day	2.01±0.33 [a]	3.83±0.08 [a]	5.4±0.54 [a]
3 Days	2.62±0.06 [b]	2.94±0.09 [a]	6.44±0.45 [a]
4 Days	2.03±0.12 [a]	4.95±0.43 [b]	6.04±0.65 [a]
5 Days	2.18±0.55 [a]	5.69±0.57 [b]	5.93±0.89 [a]
6 Days	3.11±0.04 [b]	6.65±0.25 [b]	7.72±0.95 [a]

Values were Means ± SE of mean replicates. Values followed by the same superscript(s) within column were significantly different at ($p < 0.05$).

Table 2 Duration of fermentation and weight of yeast produced in molasses

Fermentation duration (Days)	Molasses		
	5 % (g)	10 % (g)	15 % (g)
1 Day	0.23±0.06 [a]	-	-
2 Day	0.50±0.04 [a]	0.81±0.03 [a]	1.13±0.22 [a]
3 Days	0.45±0.03 [a]	0.22±0.25 [a]	1.22±0.05 [a]
4 Days	0.56±0.01 [a]	1.09±0.06 [a]	1.05±0.09 [a]
5 Days	0.23±0.09 [a]	2.88±0.10 [b]	1.05±0.09 [a]
6 Days	0.53±0.02 [a]	1.03±0.08 [a]	1.39±0.05 [a]

Table 3 Duration of fermentation and weight of yeast produced in sugar

Fermentation duration (Days)	Sugar		
	5 % (g)	10 % (g)	15 % (g)
1 Day	0.41±0.07 [a]	-	-
2 Day	2.38±0.78 [b]	0.97±0.25 [a]	2.05±0.16 [b]
3 Days	0.36±0.08 [a]	0.38±0.1 [a]	0.82±0.07 [a]
4 Days	0.14±0.07 [a]	0.27±0.07 [a]	0.34±0.06 [a]
5 Days	0.10±0.03 [a]	0.48±0.09 [a]	0.54±0.17 [a]
6 Days	0.31±0.06 [a]	0.31±0.12 [a]	0.39±0.09 [a]

Table 4 Metabolizable energy, crude protein, crude lipid and crude fibre of yeast cultured with different concentrations of dry malt extract as a selected media

	ME (MJ/Kg)	CP (%)	CL (%)	CF (%)
Control	15.6±0.02[a]	45.65±0.18[c]	0.70±0.00[a]	2.70±0.00[b]
5 %	14.78±0.00[a]	38.60±0.29[b]	0.90±0.00[a]	2.40±0.00[b]
10 %	14.57±0.00[a]	28.20±0.43[a]	0.95±0.04[a]	1.90±0.00[a]
15 %	14.57±0.00[a]	25.35±0.32[a]	1.05±0.04[b]	1.60±0.00[a]

Proximate analysis further revealed that fermentation conditions significantly affected the nutritional quality of yeast. Crude protein (CP) content was highest in control samples and remained relatively high at lower substrate concentration (5%) but decreased progressively with increasing concentration. Conversely, crude lipid (CL) content increased with higher substrate concentrations, while crude fibre (CF) content declined. Metabolizable energy (ME) values showed slight reductions in fermented samples compared to control, reflecting energy utilization during microbial metabolism.

Overall, the findings indicate that yeast cultured in dry malt extract, particularly at lower concentrations, provides a favourable balance between growth yield and nutritional quality. These results highlight the potential of yeast as a viable alternative protein source in animal feed, with substrate optimization playing a critical role in maximizing both production efficiency and feed value.

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DISCUSSION

The findings of this study demonstrate that substrate type, concentration, and fermentation duration significantly influence yeast growth and nutritional composition, supporting the initial objective of optimizing yeast as a feed additive. Dry malt extract consistently produced higher dry matter (DM) yield compared to molasses and sugar, indicating its superiority as a growth medium. Malt extracts potentially act as a key energy provider, supplementing diets with fermentable carbohydrates to living cells can be a useful approach for yeast growth in fermentation process (Biagi et al., 2007). This can be attributed to its more balanced nutrient profile, including not only carbohydrates but also proteins, vitamins, and minerals required for yeast proliferation. The relationship between certain feed and malt has been shown in earlier studies and that malt appears to be better than other media as far as growing yeast is concerned (Glen et al., 2009). In contrast, molasses and sugar primarily supply carbohydrates, which may limit yeast growth due to insufficient essential nutrients. These results align with previous studies reporting that nutrient-rich substrates enhance microbial growth and fermentation efficiency. Thus, it was presumed due to the limitation of nutrient in molasses and sugar as both supply energy and no other nutrients. Reports by USDA (2019a); USDA (2019b), suggested that both ingredients are commonly used in bakery industry and are rich in carbohydrate but little protein or fat. With limited or lack of several nutrients, yeast strains do not enter the stationary phase but opt for pathways such as pseudohyphal growth and if this situation continues, the end-point is likely to be a stationary-phase cell (Laura et al., 2022).

The observation that 10% substrate concentration yielded optimal growth, while 15% resulted in reduced performance, suggests that high substrate concentration imposes osmotic stress on yeast cells, inhibiting their metabolic activity. Attfield & Kletsas (2001) stated that yeast can produce strong fermentative activity in a medium under low osmotic stress. This finding is consistent with earlier reports indicating that excessive sugar concentrations negatively affect yeast viability and fermentation efficiency. Additionally, the reduction in crude protein (CP) and crude fibre (CF) with increasing substrate concentration, alongside an increase in crude lipid (CL), can be explained by yeast autolysis and metabolic changes during fermentation. According to Alexandre (2011), a higher concentration of protein degradation products such as amino acids has frequently been reflected as the characteristic of yeast autolysis. Similar trends have been reported in previous studies, where fermentation leads to protein degradation (Sergi, 2020) and lipid accumulation due to cellular breakdown processes. The triglycerides are cleaved representation glycerol and fatty acids (Vakhlu & Kour, 2006).

The results of this study have important implications for animal nutrition and feed technology. The identification of yeast cultured in dry malt extract, particularly at 5% concentration, as a nutritionally viable protein supplement highlights its potential as a sustainable alternative to conventional feed ingredients such as soybean meal. This is particularly relevant in regions like Malaysia, where high feed costs and reliance on imported raw materials pose significant challenges to livestock production. The use of locally produced yeast grown on agro-industrial substrates can contribute to reducing feed costs, improving feed efficiency, and enhancing overall productivity in ruminant systems. Furthermore, the probiotic properties of yeast can promote gut health and improve nutrient utilization, supporting better animal performance.

Despite the promising findings, this study has several limitations. Firstly, the experiment was conducted under controlled laboratory conditions, which may not fully represent practical farm environments. Secondly, only three substrates and limited concentration ranges were evaluated, which may restrict the generalizability of the results. Additionally, the study focused primarily on

proximate composition and DM yield, without assessing in vivo animal performance, digestibility, or long-term health effects of yeast supplementation. These factors are crucial for determining the practical applicability of yeast as a feed ingredient.

Future studies should focus on evaluating the effects of yeast supplementation on animal performance through feeding trials, including parameters such as weight gain, feed conversion efficiency, and milk production. Further research is also needed to explore a wider range of locally available substrates, including agro-industrial wastes, to enhance cost-effectiveness and sustainability. Investigations into optimizing fermentation conditions, such as pH, temperature, and aeration, could further improve yeast yield and nutritional quality. Additionally, studies examining the functional properties of yeast, including its probiotic effects and impact on rumen microbiota, would provide deeper insights into its role in animal nutrition.

CONCLUSION

This study demonstrated that substrate type, concentration, and fermentation duration significantly influence the growth performance and nutritional composition of yeast (*Saccharomyces cerevisiae*) intended for use as a feed additive. Among the substrates evaluated, dry malt extract consistently supported the highest dry matter (DM) yield compared to molasses and sugar, indicating its superiority as a fermentation medium. Optimal yeast growth was observed at 10% substrate concentration, while higher concentration (15%) resulted in reduced growth, likely due to osmotic stress. Proximate analysis revealed that yeast produced at lower substrate concentration (5%) contained relatively high crude protein (CP) and acceptable metabolizable energy (ME), while increasing substrate concentration led to reduced CP and crude fibre (CF) but increased crude lipid (CL). Overall, fermentation slightly reduced energy content compared to control samples, but yeast remained a nutritionally valuable feed component.

The findings highlight the strong potential of yeast as a cost-effective and sustainable alternative protein source in ruminant feed formulation. Specifically, yeast cultured in 5% dry malt extract provides a balanced nutritional profile suitable for improving feed quality while maintaining production efficiency. This study contributes to the field of animal science by offering practical insights into optimizing yeast production using locally available substrates, thereby supporting efforts to reduce dependence on expensive imported feed ingredients. The application of such alternative feed resources can enhance the sustainability, profitability, and resilience of livestock production systems, particularly in regions facing feed resource constraints.

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