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### Sugar beet pulp as an economical alternative for yellow corn in feeding dairy cows: its effect on blood chemistry, milk productivity and quality

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#### ABSTRACT

This work investigated the efficacy of partial replacement of imported yellow corn in the diets of Holstein Friesian dairy cows with dried sugar beet pulp on blood biochemical parameters, milk productivity and quality. Fifteen multiparous, clinically healthy lactating Holstein Friesian cows (with average body weight:  $682 \pm 25$  kg; average milk yield:  $30.4 \pm 3.0$  kg/d at 90 days postpartum) were distributed into three groups (5 for each group). A control group (G1) received a standard yellow corn-based diet, while the experimental groups (G2 and G3) received the same diets but with 30% and 50% of yellow corn were replaced by dried sugar beet pulp, respectively. The trial lasted 60 days. Blood samples were analyzed for thyroid hormones, lipid profiles, oxidative stress markers and liver enzymes. Milk samples were assessed for organoleptic properties, physicochemical composition, oxidative state and microbial quality. Results showed that both serum thyroxine (T4) and triiodothyronine (T3) levels were significantly ( $P < 0.05$ ) increased by elevating the sugar beet pulp percentage in diets. Liver enzyme activities (Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST)) were significantly ( $P < 0.05$ ) lowered with increasing sugar beet pulp proportion in the diet. The lipid profile parameters were improved but non-significantly with sugar beet pulp inclusion, showing lower each of total cholesterol, triglycerides, low-density lipoprotein cholesterol (LDL), as well as showing higher high-density lipoprotein cholesterol (HDL). However, there were no significant ( $P < 0.05$ ) effects on the oxidative stress markers. Milk yield increased significantly ( $P < 0.05$ ) with elevating the dried sugar beet pulp percentages in rations. Additionally, milk fat and protein percentages were improved significantly ( $P < 0.05$ ). No detrimental

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effects were observed on milk organoleptic properties. No significant ( $P < 0.05$ ) changes were found in milk titratable acidity, lactose %, solid nonfat % or ash content. Milk oxidative state was unaffected by dried sugar beet pulp inclusion as no significant ( $P < 0.05$ ) changes were obtained in the thiobarbituric acid reactive substances. Moreover, no adverse effects were observed on the microbial quality of milk by sugar beet pulp inclusion in diets as no significant ( $P < 0.05$ ) changes were found in total bacterial counts, total coliform counts, total staphylococcus counts, as well as total mould and yeast counts. In conclusion, dried sugar beet pulps can replace up to 50% of yellow corn in dairy cow rations without adverse effects on the cow's health, however enhancing milk yield and quality.

## INTRODUCTION

Holstein Friesian cows, renowned for their exceptional milk production, have gained popularity among Egyptian dairy farms and within the dairy industry (**Sahwan et al. 2024**). Effectively managing the nutrient needs of dairy animals is essential for achieving optimal performance and high-quality milk production. Feeding practices and strategies significantly influence milk composition and characteristics. Additionally, the use of diverse feed sources and efficient feeding management is widely recognized as a key approach to promoting sustainable animal health (**Ponnampalam et al. 2024**). Egyptian consumers and producers have become increasingly conscious of the type, composition, quality, nutritional value and safety of the consumed milk. Moreover, milk prices are typically influenced by its compositional quality, prompting Egyptian dairy farms to focus on producing high-quality milk while enhancing its overall quality and safety (**Sahwan et al. 2024**).

The rising of cereals cost is driving producers to explore alternative feed materials (**Pinotti et al. 2021**). A pressing challenge that needs addressing is the rising demand for cereals, which is partly responsible for the recent increase in price volatility. By 2050, global crops demand will be elevated by about 100-110% (**Tilman et al. 2011**), which concern about security of food causing more pressure on the industry of livestock. Therefore, the main goal in modern management of dairy cows is maximizing the energy intake (**Heydari et al. 2021**).

Replacing corn with cost-effective, high-

quality feed materials not only aligns with the physiological needs of animal nutrition but also reduces reliance on more expensive concentrate feeds (**Özkan and Deniz, 2023**). Additionally, utilizing agro-industrial by-products as cattle feed is an effective approach to waste management (**Goyal and Kaur, 2022**).

The agri-food industry generates substantial by-products and wastes that are characterized by high economic and nutritional values (**Mohsen et al. 2021**). Many of these by-product feeds are rich in fiber and low in starch, making them suitable for partially replacing both forage and concentrate in livestock diets (**Malekhhahi et al. 2023**). In Egypt, animal nutrition systems are predominantly dependent on imported grains. Nevertheless, the expansion of the sugar beet industry is gradually increasing the availability of sugar beet pulp (SBP), presenting a valuable local alternative (**Habeeb et al. 2017**). Efforts have focused on identifying alternatives to enhance the energy density of rations. From a sustainable production standpoint, SBP offers potential as a fermentable, non-forage fiber source to reduce feed costs while supporting cow health and improving milk quality (**Keim et al. 2022**).

The addition of SBP to ruminant feed may help to resolve the competitive conflict that directly exists between the livestock and human on feeding systems without sacrificing the quality of feed or the productivity of animals (**Abo-Zeid et al. 2017**). The ration produced from SBP is a highly palatable feed with high energy content, containing easily digestible fiber that benefits ruminants by maintaining

rumen health and promoting acetate production (**Habeeb, 2024**). It is worth noting that SBP has high levels of energy and crude protein similar to those of corn grains. In addition, it is rich in fermentable fiber with high water-holding capacity, which has the ability to alter the digestive tract physical and chemical characteristics, thereby affecting the feed intake as well as the animal growth (**Badaras et al. 2022**).

Beet pulp is mainly made up of neutral detergent fiber (NDF), which is highly digestible, with a digestibility of 85% (**NRC, 2001**). Neutral detergent fiber (NDF) can be utilized as an energy source in ruminant diets. It is unique due to its high cation exchange capacity, which helps in maintaining pH levels and stabilize the rumen environment. Furthermore, beet pulp NDF is low in lignin. Replacing forage with beet pulp in lactating cow diets has varying effects on feed intake and increased milk yield (**Malekhhahi et al. 2023**). Incorporating dietary byproducts with highly digestible NDF can serve as a substitute for corn, which is high in digestible starch (**Heydari et al. 2021**).

Nutritional strategies that affect milk components involve providing sufficient amount of rumen-degradable protein as well as adequate neutral detergent fiber (NDF) in the rations. Nutrition and diet changes can significantly influence concentrations of fat and protein. Fat in particular, is highly sensitive to changes in diet (**Tyasi et al. 2015**). It is well-established that the composition of structural carbohydrates (fiber) influences short-chain fatty acids production as acetate, propionate, and butyrate, as well as the methane gas formation in the rumen. As a result, these factors can determine the milk productivity, the fat content and the taste (**Ponnampalam et al. 2024**). The highly digestible dietary fiber fraction contributes to an increased acetate-to-propionate ratio. Higher acetate levels help greater content of fat and milk yield in cattle (**Goyal and Kaur, 2022**).

Moreover, fiber which is the main component of SBP, serves a mechanical function in promoting chewing, gastrointestinal peristalsis,

and healthy digestion. Chewing is crucial for quickly breaking down feed particles, increasing their surface area for ruminal microorganisms to attach, stimulating saliva production, and maintaining ruminal pH balance. Peristalsis facilitates rumen mixing, a vital process for efficient feed digestion. This ensures a more uniform environment and regulates the retention or passage of particles from the rumen (**Fustini et al. 2017**).

Generally, the nutrition of dairy cows significantly influences milk composition, quality and yield. Any changes to their diet should be carefully planned and assessed to understand their effects on milk components in dairy production (**Tyasi et al. 2015**). Quality of milk as well as its safety are essential to meet requirements of consumer. The composition of milk as fat content, protein percentages and lactose levels, can serve as valuable diagnostic tools for management of herd, particularly for monitoring lactating animals health (**Duguma, 2022**). High-quality milk is crucial both economically and for health. It increases farmers' earnings while enabling industrial institutions and organizations to produce superior dairy products. Consumers increasingly prefer milk products that are hygienic and nutritionally high-quality, driven by rising demand for healthier options (**Baran and Adıgüzel, 2020**).

Therefore, this study aimed to investigate the efficacy of partial replacing of imported yellow corn with dried SBP byproducts in Holstein dairy cows' diets. The study assessed their impacts on the cows' health status, some blood biochemical parameters, as well as evaluating changes in milk yield, organoleptic properties, physicochemical composition, oxidative state and microbiological quality.

## MATERIALS and METHODS

### Ethics Guidelines of the Animal Research:

All cows received humane care. The experimental procedures were conducted in compliance with the guidelines approved by the Institutional Animal Care and Use Committee (Agricultural Research Centre-IACUC, Ethical approval number ARC- AHRI-13-25).

## Experimental Design:

### Experimental Animals

The experiment was conducted at a private Holstein dairy farm in El-Ismailia Governorate, Egypt. A total of fifteen multiparous lactating Holstein cows, with an average body weight of  $682 \pm 25$  kg and an average daily milk yield of  $30.4 \pm 3.0$  kg, were selected for the study. The trial started 90 days postpartum and lasted for 60 days. The cows were randomly assigned to three equal treatment groups, each consisting of five cows.

### Experimental Rations

The first group (G1), which served as the control group, was fed the basal ration with Argentina yellow corn (**Table 1**), while the second (G2) and third (G3) groups were fed the basal ration with partially replacing 30% and 50% of yellow corn by Egyptian dried SBP, respectively

Table 1. Dietary ingredients and estimated nutritive values for all the experimental rations of Holstein dairy cows during the experimental period.

Dietary Ingredients	G1	G2	G3
		% as Fed Basis	
Soybean meal	8.793	8.793	8.793
Yellow corn grains	17.885	12.520	8.943
Dry sugar beet pulp	0.000	5.365	8.942
Wheat bran	7.452	7.452	7.452
Alfalfa hay	14.904	14.904	14.904
Corn silage	35.770	35.770	35.770
Molasses	2.981	2.981	2.981
Megalac	0.835	0.835	0.835
Full-fat soybean	1.192	1.192	1.192
Premix	0.179	0.179	0.179
Sodium bicarbonate	0.536	0.536	0.536
Common salt	0.238	0.238	0.238
Limestone	0.075	0.075	0.075
Selenium	0.015	0.015	0.015
Chelated Mn	0.006	0.006	0.006
Chelated Zn	0.015	0.015	0.015
Life yeast	0.030	0.030	0.030
Nitro-tox	0.030	0.030	0.030
Calibrin-Z	0.149	0.149	0.149
Water	8.942	8.942	8.942
Total	100.000	100.000	100.000
Estimated nutritive values			
ME (Mcal/kg DM)	2.39	2.38	2.37
NEL (Mcal/kg DM)	1.50	1.49	1.49
CP (g/kg DM)	163.0	163.4	163.9
RUP (g/kg DM)	64.70	66.30	68.00
RDP (g/kg DM)	98.30	97.00	95.90

ME: metabolizable energy. NEL: net energy for lactation. CP: crude protein. RUP: rumen undegradable protein. RDP: rumen degradable protein. DM: Dry Matter.

## Biochemical Studies:

### Blood sampling:

On the last day of the experiment, approximately 3 hours after morning feeding, blood samples were collected from the jugular vein of each cow. Approximately 10 mL of blood was drawn and transferred into vacuum-sealed serum separation tubes (SST) containing gel for serum separation. The blood samples were allowed to clot at room temperature for 30 minutes and then centrifuged at 3000 rpm for 15 minutes. The resulting serum was carefully aliquoted into sterile tubes and stored at  $-20^{\circ}\text{C}$  for subsequent biochemical analyses.

### Thyroid hormones Estimation:

Serum concentrations of triiodothyronine (T3) and thyroxine (T4) were measured using commercial enzyme immunoassay kits from Bio-Diagnostics Ltd., (UK), following the manufacturer's instructions

### Liver Function Enzymes Estimation:

Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were assayed by the method of Huang et al. (2006).

### Lipid profile Estimation:

The lipid profile was determined by using enzymatic colorimetric kits from different manufacturers; the total cholesterol concentrations (TC) (Cat. No. 304710050, ELITech Diagnostic, France), triglycerides (TG) (Cat. No. 303113050, ELITech Diagnostic, France), high-density lipoprotein cholesterol (HDL) (Cat. No. 0599, Stanbio Laboratory, USA), low-density lipoprotein cholesterol (LDL-C) (lot no. 990610, QCA Co., Spain) following the methodologies of Tietz (1995).

### Oxidative Stress Markers Estimation:

The activities of antioxidant enzymes were measured using established methods: superoxide dismutase (SOD) and catalase (CAT) activities were determined following the protocol described by Weydert and Cullen (2010). While, malondialdehyde (MDA) levels were measured using the method outlined by Awang Daud et al. (2022).

### Milk Productivity Determination:

Cows were mechanically milked every 8 hours. After each milking process, milk of each cow was weighed separately. Milk yield of each cow was automatically recorded by digital milk meter, which is fixed in every milking unit within the parlor. The data were transferred by network to computer system, which computed milk yield average of each cow in day.

### Milk Quality Determination:

At the experiment end, the collected raw milk samples of the three groups were immediately transported under strict aseptic conditions in a chilled, insulated icebox maintained at  $4^{\circ}\text{C}$  to the food hygiene laboratories of the Animal Health Research Institute for further analysis.

### Milk Organoleptic Examinations:

A testing group consisting of seven panelists of staff members of food hygiene department, Animal Health Research Institute, Agriculture Research Center, evaluated the organoleptic properties of milk samples collected from the three groups. All samples were heated to  $73^{\circ}\text{C}$  with 10 min holding time and cooled to  $4^{\circ}\text{C}$ . Then, 30 mL of each sample was poured into single-use plastic disposable cup. Each sample was evaluated as 5 (excellent), 4 (good), 3 (moderate), 2 (bad), and 1 (very bad) on a 5-points hedonistic value, including texture, color, odour, taste and overall acceptability as previously performed by Kim et al. (2022).

### Milk Physicochemical Analysis:

Each milk sample fat content was determined by using Gerber's method (Vašková and Bučková, 2015), while the protein percentage was determined by using Kjeldahl according to the method of Association of Official Analytical Chemistry no. 991.20 AOAC, 2006 and ISO 8968-1:2014. Additionally, the determination of the titratable acidity, lactose contents, ash contents (mineral contents) and the solid not fat (SNF) contents in milk samples were performed according to the method of Association of Official Analytical Chemistry (AOAC, 2007).

### Milk Oxidative State Determination:

Lipid oxidation of milk samples were determined by malondialdehyde (MDA) quantifi-

cation using the 2-thiobarbituric acid reactive substances (TBARS) method. Results were expressed as  $\mu\text{g}$  of MDA per ml of milk (Papastergiadis et al. 2012).

### Milk Microbiological Evaluation:

All samples were prepared according to the technique recommended by ISO, 7218 (2007); 10-fold serial dilutions of the milk samples up to  $10^{-6}$  have been made using sterilized buffered peptone water. The total bacterial counts (TBC) of all milk samples were estimated using the method of ISO, 4833-2 (2013). While, the total coliform counts (TCC) were per-

formed by the technique of ISO, 4832 (2006). Additionally, the total Staphylococcus counts (TSC) were performed according to the method of ISO, 6888-1 (2021). Moreover, the total mould and yeast counts were performed by the technique of ISO, 21527-1 (2008).

### Statistical analyses:

The data were analyzed using one-way ANOVA using SPSS version 19 (SPSS Inc., Chicago, IL, USA) statistical package. The variances between groups were determined by using Duncan's post-hoc test. A p-value less than 0.05 was considered statistically significant.

## RESULTS

Table 2. Effects of partially replacing yellow corn with dried sugar beet pulp in Holstein dairy cows' rations on serum biochemical parameters.

Biochemical analysis	Groups			Significance
	G1	G2	G3	
Triiodothyronine (T3) ng/dL	1.03 $\pm$ 0.010 <sup>b</sup>	1.07 $\pm$ 0.018 <sup>b</sup>	1.43 $\pm$ 0.11 <sup>a</sup>	**
Thyroxine (T4) $\mu\text{g/dL}$	34.98 $\pm$ 0.028 <sup>b</sup>	36.58 $\pm$ 0.029 <sup>a</sup>	37.3 $\pm$ 0.018 <sup>a</sup>	**
Alanine aminotransferase (ALT) U/L	39.43 $\pm$ 0.37 <sup>a</sup>	36.00 $\pm$ 0.71 <sup>b</sup>	31.87 $\pm$ 0.11 <sup>c</sup>	**
Aspartate aminotransferase (AST) U/L	113.7 $\pm$ 0.20 <sup>a</sup>	110.4 $\pm$ 0.34 <sup>b</sup>	106.0 $\pm$ 0.70 <sup>c</sup>	**
Total cholesterol (TC) mg/dL	160 $\pm$ 0.93 <sup>a</sup>	158 $\pm$ 0.71 <sup>a</sup>	156 $\pm$ 0.80 <sup>a</sup>	NS
triglycerides (TG) mg/dL	33.68 $\pm$ 0.55 <sup>a</sup>	33.7 $\pm$ 0.64 <sup>a</sup>	32.2 $\pm$ 0.71 <sup>a</sup>	NS
High-density lipoprotein cholesterol (HDL) mg/dL	92.0 $\pm$ 0.54 <sup>a</sup>	92.3 $\pm$ 0.28 <sup>a</sup>	93.4 $\pm$ 0.28 <sup>a</sup>	NS
Low density lipoprotein cholesterol (LDL-C) mg/dL	11.0 $\pm$ 0.35 <sup>a</sup>	10.8 $\pm$ 0.13 <sup>a</sup>	10.9 $\pm$ 0.46 <sup>a</sup>	NS
Superoxide dismutase (SOD) U/mL	67.89 $\pm$ 0.839 <sup>a</sup>	70.25 $\pm$ 0.639 <sup>a</sup>	69.39 $\pm$ 1.152 <sup>a</sup>	NS
Catalase (CAT) U/mL	46.50 $\pm$ 0.50 <sup>a</sup>	46.86 $\pm$ 0.39 <sup>a</sup>	47.18 $\pm$ 0.48 <sup>a</sup>	NS
Malondialdehyde (MDA) nmol/mL	13.34 $\pm$ 0.265 <sup>a</sup>	13.40 $\pm$ 0.367 <sup>a</sup>	14.12 $\pm$ 0.338 <sup>a</sup>	NS

G1=control group, Holstein dairy cows fed on yellow corn based-diet. G2= Holstein dairy cows fed on 30% dried sugar beet pulp based-diet. G3= Holstein dairy cows fed on 50% dried sugar beet pulp based-diet. Means within the same row with different superscripts a, b and c are significantly different ( $P < 0.05$ ). NS: not significant, \*\*: significant

Table 3. Effects of partially replacing yellow corn with dried sugar beet pulp in Holstein dairy cows' diets on the milk organoleptic properties.

Sensory Attributes	Groups			Significance
	G1	G2	G3	
Texture	4.60±0.245 <sup>a</sup>	4.80±0.20 <sup>a</sup>	5.00±0.00 <sup>a</sup>	NS
Colour	5.00±0.00 <sup>a</sup>	4.80±0.20 <sup>a</sup>	4.60±0.25 <sup>a</sup>	NS
Odour	4.80±0.20 <sup>a</sup>	4.60±0.25 <sup>a</sup>	4.80±0.20 <sup>a</sup>	NS
Taste	5.00±0.00 <sup>a</sup>	5.00±0.00 <sup>a</sup>	5.00±0.00 <sup>a</sup>	NS
Overall Acceptability	19.40±0.40 <sup>a</sup>	19.20±0.58 <sup>a</sup>	19.40±0.40 <sup>a</sup>	NS

G1=control group, Holstein dairy cows fed on yellow corn based-diet. G2= Holstein dairy cows fed on 30% dried sugar beet pulp based-diet. G3= Holstein dairy cows fed on 50% dried sugar beet pulp based-diet. Means within the same row with different superscripts are significantly different (P< 0.05). NS: not significant

Table 4. Effects of partially replacing yellow corn with dried sugar beet pulp in Holstein dairy cows' diets on the milk physicochemical properties.

Physicochemical Parameters	Groups			Significance
	G1	G2	G3	
Milk yield (kg/d)	38.9± 0.29 <sup>c</sup>	43.3± 0.49 <sup>b</sup>	45.8± 0.25 <sup>a</sup>	**
Milk fat (%)	3.25± 0.038 <sup>c</sup>	3.90± 0.034 <sup>b</sup>	4.21± 0.069 <sup>a</sup>	**
Milk protein (%)	2.95± 0.028 <sup>c</sup>	3.08±0.026 <sup>b</sup>	3.16± 0.024 <sup>a</sup>	**
Titrate acidity (%)	0.156±0.007 <sup>a</sup>	0.162±0.005 <sup>a</sup>	0.168±0.010 <sup>a</sup>	NS
Lactose (%)	3.24±0.146 <sup>a</sup>	3.41±0.096 <sup>a</sup>	3.74±0.284 <sup>a</sup>	NS
Solids nonfat (%)	8.39±0.031 <sup>a</sup>	8.66±0.038 <sup>a</sup>	9.43±0.34 <sup>a</sup>	NS
Ash (%)	0.54±0.058 <sup>a</sup>	0.61±0.061 <sup>a</sup>	0.70±0.055 <sup>a</sup>	NS
TBARS (nmol/mL)	0.244±0.026 <sup>a</sup>	0.252±0.038 <sup>a</sup>	0.219±0.045 <sup>a</sup>	NS

G1=control group, Holstein dairy cows fed on yellow corn based-diet. G2= Holstein dairy cows fed on 30% dried sugar beet pulp based-diet. G3= Holstein dairy cows fed on 50% dried sugar beet pulp based-diet. Means within the same row with different superscripts are significantly different (P< 0.05). NS: not significant. \*\*: significant. TBARS: Thio Barbeturic Acid Reactive Substances

Table 5. Effects of partially replacing yellow corn with dried sugar beet pulp in Holstein dairy cows' diets on the milk microbiological quality.

Microbiological analysis	Groups			Significance
	G1	G2	G3	
Total Bacterial Counts (CFU/ml)	$3.6 \times 10^4 \pm 9.2 \times 10^3^a$	$2.6 \times 10^4 \pm 5.1 \times 10^3^a$	$3.4 \times 10^4 \pm 8.4 \times 10^3^a$	NS
Total Coliform Counts (CFU/ml)	$5.4 \times 10^1 \pm 9.27^a$	$4.8 \times 10^1 \pm 12.24^a$	$5.0 \times 10^1 \pm 8.60^a$	NS
Total Staphylococcal Counts (CFU/ml)	$8.18 \times 10^1 \pm 4.61^a$	$7.36 \times 10^1 \pm 4.87^a$	$8.38 \times 10^1 \pm 4.03^a$	NS
Total Mould and yeast Counts (CFU/ml)	$1.8 \times 10^1 \pm 3.56^a$	$2.2 \times 10^1 \pm 3.74^a$	$2.0 \times 10^1 \pm 4.47^a$	NS

G1=control group, Holstein dairy cows fed on yellow corn based-diet. G2= Holstein dairy cows fed on 30% dried sugar beet pulp based-diet. G3= Holstein dairy cows fed on 50% dried sugar beet pulp based-diet. Means within the same row with different superscripts are significantly different ( $P < 0.05$ ). NS: not significant. CFU: colony forming units.

## DISCUSSION

### Biochemical studies:

Employing dried SBP as a cost-effective substitute for yellow corn in dairy Holstein Friesian cows' diet may affect thyroid hormones, oxidative stress biomarkers and chemical blood metabolites. So, blood biochemistry is significantly estimated to evaluate the cows health status (Tufarelli et al. 2023).

### Thyroid hormones:

The thyroid hormone levels are presented in **table 2**. The serum T4 (thyroxine) concentrations were significantly ( $P < 0.05$ ) higher in Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP when compared with those fed yellow corn-based diets. These findings were consistent with those of **Abo-Zeid et al. (2017)** who stated that upon increasing the dietary SBP levels, T4 concentrations were quadratically elevated, as the relationship between the dietary SBP levels and T4 concentrations is not linear but follows a curved pattern, often described by a second-degree equation and this implies that as SBP levels increase, T4 concentrations rise to a certain point and then may plateau or decline, forming a curve rather than a straight line. Additionally, **Suliman and Mohamed (2024)** reported that T4 were laniary increased by adding SBP. The elevation of thyroid hormone levels were associated with increased blood

cholesterol levels (**Harper et al. 2005**).

While the serum T3 (triiodothyronine) concentrations (**table 2**) were significantly ( $P < 0.05$ ) higher in Holstein Friesian dairy cows fed rations with 50% corn replaced by dried SBP when compared with those fed yellow corn-based diets. These results agreed with **Suliman and Mohamed (2024)** who reported that T3 was laniary increased by adding SBP. However, the results disagreed with **Khaliel et al. (2022)** who found a significant decrease in plasma T3 level in the SBP treated groups than yellow corn groups.

The development of the mammary glands and the maintenance of efficient lactation are significantly influenced by thyroid hormones. They work either directly by attaching themselves to the mammary gland's thyroid hormone receptors or indirectly by enhancing the effects of other hormones like glucocorticoids and prolactin. Additionally, the differentiation of mammary epithelial cells depends on the activation of progesterone and estrogen receptors, which are modulated by thyroid hormones (**Slepicka et al. 2021**). Moreover, Thyroid hormones affect the production of milk and direct the supply of nutrients to the mammary gland (**Sánchez et al. 2024**).

Thyroid hormones (T3 and T4) are indicators of metabolic activity. Because sugar beet



pulp is moderately fermentable and high in fiber, it can enhance rumen health and energy balance, which in turn affects T3 and T4 levels indirectly. The SBP improves metabolic efficiency and food consumption, so it may raise T3 and T4 concentrations. Numerous metabolic processes, including the metabolism of lipids, proteins and carbohydrates, are impacted by thyroid hormones, which increases basal energy demand (Botella-Carretero et al. 2006).

#### Liver enzymes:

The liver enzymes results are presented in **table 2**. Introducing dried SBP into the rations led to a markedly low liver enzyme activity (AST and ALT). The values of ALT and AST levels were highest in the serum of Holstein Friesian dairy cows fed yellow corn-based diets, followed by those fed rations with 30% and 50% corn replaced by dried SBP, exhibiting a significant decline ( $P < 0.05$ ).

These results may indicate that dry SBP-containing meals were safe, digested, and metabolized appropriately, with no negative effects noted, because the readings were within the usual reference range (Abdel-Azim et al. 2011; Khaliel et al. 2022). These results agreed with the previous work of Ali et al. (2019) and Mohsen et al. (2021), according to their findings, the control group had increased ALT and AST activity, while both dried SBP groups showed lower values. However, the results disagreed with the previous work of Khaliel et al. (2022) who found that buffaloes fed the studied rations containing dried SBP did not exhibit any significant difference in serum concentrations of ALT or AST. Therefore, incorporating SBP into dairy cows' diets may boost nutrient intake without having an adverse effect on rumen health, thereby improving performance.

#### Lipid profile

The lipid profile analysis (**table 2**) revealed non-significant ( $P < 0.05$ ) improvements in Holstein Friesian dairy cows' blood lipid parameters when dried sugar beet pulp (SBP) substituted 30% and 50% of corn in their diet, compared to animals receiving conventional

yellow corn-based feed. Specifically, the results showed that the total cholesterol (TC), triglycerides (TG) and low-density lipoprotein cholesterol (LDL) were slightly lower with dried SBP feeding, while high-density lipoprotein cholesterol (HDL) was higher.

These results agreed with that of Belibaskis and Tsirgogianni (1996) who found that replacing an equivalent amount of yellow corn with dried SBP in the feed of Holstein cows did not significantly change the blood levels of TG or TC. Additionally, Mohsen et al. (2021) stated that dried SBP did not significantly affect the levels of HDL, LDL, TG or TC, as these measures were nearly identical across the different groups. However, the results disagreed with that of Harper et al. (2005) and Khaliel et al. (2022) who documented that elevated thyroid hormone concentrations correlate with increased blood cholesterol levels, as evidenced by their findings. Additionally, according to Abo-Zeid et al. (2017) sugar beet pulp (SBP) contains minimal fat content alongside substantial digestible fiber, which beneficially influences lipid metabolism. The observed reductions in serum triglycerides (TG) and total cholesterol (TC) represent a significant advantage of SBP supplementation, as it enhances lipogenesis while simultaneously suppressing lipolysis processes.

#### Oxidative Stress Markers:

Regarding results showed in **table 2**, the oxidative stress markers as Superoxide dismutase (SOD), Catalase (CAT) and Malondialdehyde (MDA) showed non-significant ( $P < 0.05$ ) changes in Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP when compared with those fed yellow corn-based diets. These non-significant differences may be due to a balanced antioxidant response, as dried SBP likely provided comparable nutritional and metabolic effects to yellow corn, thereby maintaining oxidative equilibrium in the cows. The efficient utilization of SBP as an alternative energy source may have helped sustain stable oxidative stress markers. Consequently, substituting 30% to 50% of yellow corn with dried SBP did not induce oxidative stress in

Holstein Friesian dairy cows. These findings indicate that SBP can serve as a viable alternative to imported yellow corn without adversely impacting antioxidant enzyme activity or oxidative stress levels (Raederstorff, 2009).

The obtained results disagreed with the previous work of Guo et al. (2013) who reported that in dairy cows, using pelleted SBP instead of ground corn may improve antioxidant status, increase fibre digestion and lower the incidence of subacute ruminal acidosis which acts as a stress factor.

Oxidative stress occurs when there is an imbalance between antioxidants and pro-oxidants, with the latter being predominant. This imbalance is a critical factor that disrupts molecular signalling pathways and enzyme functions, ultimately causing tissue damage (Sies et al. 2017). Reactive oxygen species (ROS), which cause oxidative stress, are produced during both normal physiological functions such as cellular metabolism and during disease states. These ROS originate from internal cellular components including mitochondria, plasma membranes, endoplasmic reticulum, and peroxisomes structures where enzymatic reactions and spontaneous oxidation of diverse compounds occur (Moldovan and Moldovan, 2004). Additionally, external factors such as chronic stress, strenuous exercise, infections, allergens, and environmental pollutants can further promote ROS production (Jakubczyk et al. 2020). In dairy cows, oxidative stress plays a role in increasing their vulnerability to diseases (Khan et al. 2024). Superoxide dismutase (SOD) is a crucial enzyme that neutralizes superoxide radicals by converting them into oxygen and hydrogen peroxide, serving as the primary defence against excessive ROS (Pisoschi and Pop, 2015). The antioxidant defence system also includes CAT, which further detoxifies hydrogen peroxide into water and oxygen, preventing cellular damage. Additionally, MDA, a byproduct of polyunsaturated fatty acid peroxidation, is widely recognized as a biomarker for oxidative stress assessment (Halliwell and Whiteman, 2004). SBP contains abundant fermentable fiber that encourages the proliferation of beneficial intestinal microorganisms, particularly

Lactobacillus species. This improvement in gut microbiota composition facilitates increased production of short-chain fatty acids (SCFAs), which deliver systemic anti-inflammatory and antioxidant benefits. These SCFAs play a crucial role in modulating immune responses, thereby diminishing inflammation-triggered oxidative stress (Nogal et al. 2021).

#### Milk productivity:

The lactation performance represented as milk yield of the Holstein Friesian dairy cows are presented in Table 4. Milk yield was significantly ( $P < 0.05$ ) higher in cows fed dried SBP based diets by 30% (increased by 4.4 kg/d) and 50% (increased by 6.9 kg/d), when compared with those fed yellow corn based diets. Similar findings were observed earlier by Naderi et al. (2016), Wang et al. (2022) and Malekhhahi et al. (2023) who reported that dietary SBP inclusion improved the milk yield. However the results were not in consistent with Guo et al. (2013) and Heydari et al. (2021) who reported that the substitution of SBP for corn had no effect on milk yield.

Akasha et al. (1987) proposed the idea that the pituitary had an indirect role in the positive impact of thyroxine on milk secretion. It was thought that the hyperthyroid condition caused an increased release of the lactogenic hormone. Removing the thyroid gland reduced milk production, while giving cows a dried thyroid gland or thyroxine increased milk production (Matamoros et al. 2003). However, when thyroidectomized or unoperated normal cows were in the declining time of lactation and fed moderate amounts of dried thyroid tissues, their milk output clearly rose. Longer lactation is also associated with increased thyroid activity, which raises the levels of the biologically active thyroid hormone T3 (Capuco et al. 2008).

From another point of view, production of milk is strongly influenced by both the amount and the nutritional quality of the consumed feed. Nutritional impacts can affect milk yield in the short term through daily nutrient intake, as well as in the long term through carry-over effects related to changes in the cows' body

condition (Ponnampalam et al. 2024). The milk yield improvement for cows fed SBP - based rations can be explained by an elevation in the dry matter intake and that greater nutrients intake and dry matter digestibility, which represent a high energy and protein utilization efficiency (Malekkhahi et al. 2023).

The main component of the fiber in sugar beet remains cellulose, which is very easily digested. In order to sustain perfect rumen conditions and boost milk production, sugar beet is a very acceptable diet for ruminants when supplemented with a suitable quantity of degradable protein (Habeeb, 2024). Additionally, concentration of total volatile fatty acids increases with the elevation of dietary SBP inclusion. That high concentration of total volatile fatty acids was an energy utilization efficiency indicator, which suggested that the inclusion of dietary sugar beet increased the energy utilization efficiency (Wang et al. 2022). Moreover, the increase in fermentable substrates by increasing inclusion of dietary SBP promoted the proliferation of ruminal microbes (Heydari et al. 2021).

#### Milk quality parameters: Organoleptic characteristics

The organoleptic assessment of milk is utilized to evaluate its quality and identify potential defects, aiding in enhancing the marketability and consumer acceptance (Doaa et al. 2021).

Concerning the statistical evaluation of the organoleptic characteristics of milk samples (table 3), the results revealed no detrimental effects of replacing yellow corn with dried SBP in diets on organoleptic properties of milk, and all samples had normal sensory parameters. There were no significant ( $p < 0.05$ ) differences observed in texture, color, odour, taste and overall acceptability of milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP compared to those fed yellow corn-based diets. Similarly, several previous studies investigated the effect of different products inclusion in lactating cows rations on organoleptic characteristics as Güler et al. (2017), Sharma et al.

(2022) and Salman et al. (2024).

Milk sensory quality is essential, significantly influencing dairy product expectations, consumer choices, purchasing decisions, preferences and overall acceptance. It is significantly affected by milk fundamental compositional properties. Milk fat was a major factor influencing these sensory attributes, higher fat content enhancing the distinctive milk products aroma, flavor, taste and texture qualities (Su et al. 2022).

#### Physicochemical properties of milk

Alterations in raw milk physicochemical properties can greatly affect dairy products quality derived from it (Kazeminia et al. 2023). Upon analysis of milk samples, the results (Table 4) revealed that fat contents significantly increased ( $p < 0.05$ ) in samples of Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP compared to those fed yellow corn-based diets. According to the Egyptian standards (ES 154-1: 2005), the minimum percentage of fat in raw cow milk should not be less than 3%. Milk from all groups was within the permissible limits.

Several previous studies have investigated the impact of substituting corn with SBP or beet-related materials in rations of dairy cows on fat content of milk. Van Knegsel et al. (2014) observed increased milk fat content when grain was replaced by SBP. However, Naderi et al. (2016) reported that replacing 24% of corn with SBP led to lowering in milk fat content. Similarly, Malekkhahi et al. (2023) reported a reduction in milk fat concentration when beet-based diets were fed. However, other studies, such as those by Alamouti et al. (2014) and Dann et al. (2014), found that there were no significant impacts on milk fat content with dietary SBP inclusion. Heydari et al. (2021) also observed no changes in the milk fat content when 24% of corn was replaced with SBP dairy cows' rations. Furthermore, Wang et al. (2022) noted that increasing the dietary beet inclusion up to 45% did not affect milk fat percentage. These findings highlight the variability in responses to

SBP supplementation in dairy cow diets.

Dried SBP supplies neutral detergent fiber (NDF), which acts as a higher physically effective fiber due to its lower density and larger particle size. This promotes increase in rumination (cud-chewing) and salivation, which enhances rumen function and ultimately boosts milk fat content (**Malekkhahi et al. 2023**).

Falling neutral detergent fiber increases the risk of lowering fat content of milk, inconsistent feed intake, poor body condition, acidosis and lameness in cows (**Tyasi et al. 2015**).

Additionally, incorporating SBP increases the level of non-forage fiber, which, when digested in rumen, enhances volatile fatty acids production as acetate and butyrate. These acids are precursors for milk fat synthesis, helping to maintain milk fat levels. Butyrate supplies energy to rumen wall, where most of it is converted to beta-hydroxybutyrate. Half of the milk fat is approximately synthesized from acetate and beta-hydroxybutyrate in the udder (**Dixon and Ernst, 2001**). Another source of milk fat is derived from the fatty acids circulating in the blood, which may come from body fat mobilization, dietary absorption or fat metabolized in the liver (**Tyasi et al. 2015**).

While regarding results of milk protein contents in **Table 4**, there were significant ( $p < 0.05$ ) increases in protein contents of milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP compared to those fed yellow corn-based diets. Similarly, **Naderi et al. (2016)** observed increased milk protein levels when 12% and 24% of corn was substituted with SBP. **Kahyani et al. (2019)** noted a tendency for higher milk protein yield with SBP inclusion, potentially due to improved particle size, palatability, nutrient digestibility and nitrogen utilization efficiency. **Wang et al. (2022)** found linear increase in milk protein percentage by beet replacement up to 45%. Additionally, **Malekkhahi et al. (2023)** found that SBP-based diets resulted in greater milk protein concentrations compared to corn-based diets.

Milk protein is a key quality indicator, reflecting the cow status and serving as a herd

energy gauge. milk protein content is affected by energy availability to rumen microbes that synthesize microbial protein (**Khastayeva et al. 2021**). Beet pulp may enhance energy density which is needed in synthesis of microbial protein, leading to increase in levels of milk protein (**Nichols et al. 2019**). Dietary protein is converted into microbial protein by rumen microbes, providing cows the essential amino acids which are absorbed by mammary glands and utilized for milk proteins production (**Tyasi et al. 2015**).

While concerning titratable acidity values in milk samples (**Table 4**), results revealed that there were no significant ( $p < 0.05$ ) differences between milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP compared to those fed yellow corn-based diets. The obtained values agreed to some extent with that found by **Tan et al. (2020)** who reported the cow milk titratable acidity as 0.16%. While, lower values were reported by **Göncü et al. (2017)** who found the titratable acidity 0.142 %, and **Baran and Adıgüzel (2020)** who reported the raw milk total acidity values as 0.120%. However, higher values were found by **Li et al. (2022)** who reported the titratable acidity as 0.18%.

Milk acidity is a crucial parameter for assessing milk quality on dairy farms (**Baran and Adıgüzel, 2020**). It reflects the concentration of all acids present in the milk, including both dissociated and undissociated acids (**Fauziah et al. 2020**). It serves as key indicator for changes in the milk chemical properties and freshness. It is closely linked to flavor and taste of milk as well as dairy products (**Li et al. 2022**). Any increase in titratable acidity primarily results from the rising concentration of lactic acid, which occurs due to lactose breakdown by dominant spoilage lactic acid bacteria under conditions of low oxygen, low temperature and acidic conditions (**Tan et al. 2020**).

While according to lactose contents in examined milk samples (**table 4**), the results revealed slightly increase during the study period in milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP. However, no significant ( $P < 0.05$ )

differences were observed for lactose levels when compared to those fed yellow corn-based diets. Similarly, **Wang et al. (2022)** found that milk lactose percentage were not affected with the increasing inclusion of dietary SBP up to 45%. Unlikely, **Malekhhahi et al. (2023)** reported that significant increase was observed for milk lactose in cows fed with SBP compared with other treatments.

The primary milk carbohydrate is lactose, though other carbohydrates of small amounts are also present (**Kazeminia et al. 2023**). The levels of lactose and minerals do not consistently respond to dietary changes. Additionally, various non-nutritional factors, such as genetics, milk production level, stage of lactation, season, diseases, facilities, the cow's age and environment can influence milk composition (**Tyasi et al. 2015**).

While concerning the milk solid nonfat contents (**table 4**), results showed slight increasing during the experiment period. However, there were no significant ( $P < 0.05$ ) differences between all cows. **The Egyptian standards (ES 154-1: 2005)** stated that the allowed percentage of solids nonfat in raw milk should not be less than 8.25%. Concerning the results, milk of all cows either fed dried SBP based diets or fed corn based-diets were within the permissible limits. Comparatively similar values were obtained by **Beykaya et al. (2017)** and **Göncü et al. (2017)** who found the non-fat dry matter percentages of milk samples 7.33-9.80%, and 7.77-8.97%, respectively. Additionally, **Baran and Adigüzel (2020)** and **Tan et al. (2020)** determined that the mean values of solid nonfat contents of collected milk samples were 9.32% and 8.32%, respectively.

The solids non-fat (SNF) in milk includes all solids like lactose, minerals, and vitamins. They have crucial role for milk's flavor and nutritional value for consumers, as well as in the yield of dairy products for producers (**Baran and Adigüzel, 2020**). The milk SNF content can be affected by different factors as seasonal changes, location or other environmental conditions (**Kazeminia et al. 2023**).

Concerning the ash contents of milk sam-

ples (**table 4**), results showed no significant differences ( $P < 0.05$ ) between milk of cows fed dried SBP based diets and milk of cows fed corn based diets. Similar values were reported by **Gwandu et al. (2018)** who found the mean ash contents 0.7 %. While lower values were reported by **Adamu et al. (2020)** which were between 0.40% -0.43%. Ash contents of milk are the reflection of the mineral composition of milk (**Adamu et al. 2020**).

#### Oxidative state of milk:

Regarding the TBARS levels in milk samples which determined fat oxidation (**table 4**), the inclusion of dried SBP in diets showed no significant ( $P < 0.05$ ) changes from cows fed dried SBP based diets when compared to those fed yellow corn diets. Similarly, several previous studies investigated the effect of dietary inclusion of different byproducts on milk oxidative state of dairy cows as **Santos et al. (2016)** and **Durman et al. (2022)**.

Milk is a highly complex fat matrix, with its fatty acid profile consisting primarily of monounsaturated fatty acids, followed by a smaller proportion of polyunsaturated fatty acids because ruminants can not synthesize them "de novo". Their presence in milk is closely linked to dietary polyunsaturated fatty acids and amount absorbed in intestine. Due to the abundance of unsaturated fatty acids, milk undergoes various chemical processes, such as oxidation, which leads to the formation of volatile compounds like saturated aldehydes. These compounds can alter the composition of milk, potentially causing off-flavor aromas, which is a significant concern for consumers (**Oancea et al. 2022**).

#### Microbiological quality of milk:

Milk samples of all cows were examined for their microbiological quality. The results in **table 5** revealed that the total bacterial counts were not significantly ( $P < 0.05$ ) different in milk of Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP compared to those fed yellow corn-based diets indicating no effect of dried SBP feeding on bacterial counts. The obtained values were lower than those reported by **Baran**

and Adıgüzel (2020), Djobo et al. (2021), Elbadry et al. (2021) and Galaby et al. (2021). However, nearly similar results were reported by Das et al. (2015) and Tan et al. (2020).

Every dairy operation should aim to produce the maximum amount of milk with the highest quality possible. Milk as a rich nutrient fluid, provides an ideal environment for many microorganisms to thrive. The milk microbiological content is crucial factor for determining its quality, as microbial contamination is difficult to be avoided completely through the specific production steps. Contamination of raw milk may be originated from various sources as air, feed, soil or milking equipment (Ahmed et al. 2022). Milk is vulnerable to be contaminated by different microorganisms, which can affect its nutritional value, pose potential public health risks (Kazeminia et al. 2023), as well as its potential to cause regional economic losses (Baran and Adıgüzel, 2020).

Related to the total coliform counts of examined milk samples (table 5), the results showed no significant ( $P < 0.05$ ) differences were found between milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP and those fed yellow corn-based diets. These findings were similar to those of Tan et al. (2020). While, lower results were reported by Das et al. (2015). However, higher results were reported by Hasan et al. (2015), Gwandu et al. (2018), Djobo et al. (2021), Elbadry et al. (2021) and Ahmed et al. (2022).

Coliforms are microorganisms that are gram-negative, aerobic or facultative anaerobic, non-spore-forming rods capable of fermenting lactose to produce acid and gas at temperatures between 32-35°C (Wanjala et al. 2018). The detection of coliform bacteria is particularly important as they serve as indicators of fecal contamination and suggest the potential presence of other gastrointestinal pathogens (Ahmed et al. 2022).

Regarding the total staphylococcal count (TSC), the results in table 5 displayed that the TSC of milk samples were not significantly ( $P$

$< 0.05$ ) affected in milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP when compared with those fed yellow corn-based diets. Relatively similar results were reported by Hasan et al. (2015). While, higher results were reported by Elbadry et al. (2021) and Ahmed et al. (2022).

Staphylococcus species, belonging to the Micrococcaceae family, are gram-positive, non-motile, non-spore-forming, facultative anaerobic bacteria with a diameter of 0.5-1.5  $\mu\text{m}$  and a cocci shape. The genus contains 28 species and 32 subspecies. Contamination by staphylococci is especially concerning because many strains can produce thermostable enterotoxins, which can lead to foodborne intoxication in consumers (Baran and Adıgüzel, 2020).

According to the results of total mould and yeast counts of examined milk samples (table 5), there were no significant ( $P < 0.05$ ) differences between milk from Holstein Friesian dairy cows fed rations with 30% and 50% corn replaced by dried SBP and those fed yellow corn-based diets. Similar results reported by Tan et al. (2020). While, higher results were reported by Baran and Adıgüzel (2020), Galaby et al. (2021), Elbadry et al. (2021) and Ahmed et al. (2022).

The formation of fungi in raw milk can occur during milking, transportation, storage, and other pre-treatment processes, and is influenced by the physiological state of animals, air quality and production conditions. These microorganisms can lead to the deterioration of raw milk as well as pose a public health risk due to mycotoxins they produce, which are secondary metabolites of fungi (Baran and Adıgüzel, 2020). The potential danger of foodborne moulds and possibly yeasts lies in their capability to produce toxic byproducts called mycotoxins, which can harm human or animal health. Although the organisms that produce these toxins may not survive during milk preparation, the preformed toxins can remain, as most mycotoxins are not destroyed by heat. Additionally, some foodborne moulds and yeasts can lead to infections or allergic reac-

tions (El-Kholy et al. 2016).

### Conclusions and Recommendations

**I**n conclusion, sugar beet pulp can replace up to 50% of yellow corn in dairy cow rations without adverse effects on health or milk quality. It improves milk yield, fat and protein content, offering a cost-effective and sustainable alternative for dairy farmers.

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