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گۆڤاری کوردستانی بۆ دەستەبژێری زانستە سرووشتییەکان و بایۆمیدسن

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Ministry of Higher Education and Scientific Research
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Dear Colleagues and Researchers,

In its second issue, titled ‹Issue 8 of the Kurdistan Journal of Natural Sciences and Biomedicine,› the journal presents a collection of research articles that significantly contribute to its growing reputation as a reliable scientific platform. Clearly, the primary aim of the journal is to establish itself within the international scientific community, enhancing its credibility and adherence to rigorous scientific standards. To achieve this objective, the journal is committed to intensifying its efforts.

While academic researchers typically prioritize reputable and influential international journals, often overlooking local publications, we remain steadfast in our commitment to mitigating these barriers. Our dedication lies in fostering a more inclusive scholarly landscape, thereby offering academic institutions and researchers a clearer trajectory to support this scientific endeavor.

The measures implemented within this journal are evidently poised to pose challenges for researchers seeking to disseminate their research, particularly in limiting the number of participating researchers to a maximum of two. While such practices may not be commonplace at the international level, they underscore the accountability and responsibility expected of researchers in their work. Even within internationally recognized journals, careful oversight ensures the genuine involvement of researchers, albeit with varying degrees of success.

Hence, our foremost and paramount endeavor is to uphold academic integrity, encapsulated by two fundamental tenets. Firstly, it embodies the earnestness of communication and the unfettered pursuit and dissemination of information and findings. Secondly, it entails the meticulous accountability for all data, sources, and outcomes presented within the research. From this perspective, the caliber of research and its outcomes serve as benchmarks across various spheres, encompassing the academic persona of researchers, scientific departments, academia, and the strategic initiatives of governmental bodies.

The endeavor to promote and uphold this positive attribute within academic institutions is presently mired in a profound crisis marked by significant instability. In its absence, the economic, social, and cultural challenges facing us exhibit a linear escalation over time, further exacerbated by the proliferation of universities and institutions, particularly within the private sector.

In discerning these trends, academic journals may discern indications of diminishing advancements in progress, self-awareness, and national security. However, there appears to be a reluctance to address this predicament decisively and to undertake requisite scientific initiatives and prognostications to anticipate the challenges of future years or generations.

What is noticeable, alongside its significance for academic researchers and governmental bodies, including universities, is an aberrant surge in the production of unplanned research within academic institutions. This surge occurs without external demands from either governmental or private sectors, save for meeting specific obligations and prerequisites for attaining higher degrees or academic promotion. Regrettably, even under the most favorable circumstances, wherein this trend prevails, a majority of research endeavors across disciplines lack scientific and creative distinction. Many remain indistinguishable and merit publication solely to be consigned to oblivion thereafter. This phenomenon is exemplified by the multitude of local journals sporadically published or the plethora of master›s and doctoral theses drafted solely to fulfill the academic supervisors› expectations, devoid of broader purpose or utility. Despite the resources available during a purported ‹Golden Age,› the absence of a coherent and forward-looking plan aligned with the exigencies of the forthcoming decades has relegated these theses to gather dust on library shelves.

While this platform may not be suited for an exhaustive discussion on this matter, its significance warrants considerable attention. Scientific journals, sanctioned and overseen by the Ministry of Higher Education, assume a pivotal role in addressing this crisis and establishing robust standards to guide its progression.

We extend our sincere gratitude for the collective efforts contributed thus far. We wholeheartedly welcome any constructive suggestions and recommendations aimed at enhancing the continual development of this journal.

Best regards
Rizgar Agha
Executive Editor

Effect of Wheat Grits Level on Biogenic Amines Formation in Traditional Kurdish Fermented Food Tarsas

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Abstract:

«Biogenic amines (BA) are nitrogenous compounds produced when amino acids undergo decarboxylation or amination by microorganisms. They are found in fermented foods like sauerkraut, beer, cheese, kimchi, soy sauce, and fermented vegetables. Tarasas is a traditional fermented Kurdish dish made from turnip roots, leaves, and wheat grits. The current study aims to investigate the effect of decreasing the amount of wheat grits used in the Tarasas product on the production of biogenic amines during fermentation. The study measured pH, titratable acidity, total nitrogen, soluble nitrogen, hydrolysis degree, and determination of biogenic amines (BA), in addition to lactic acid bacteria counts, during and after fermentation of Tarasas at °٢٥C in treatments with wheat grits ratios (٪٠ ,٪٢٥ ,٪٥٠ ,٪٧٥ ,٪١٠٠) of the total weight of grits in the Tarasas product. The results were analyzed using SPSS. One-way analysis of variance and Duncan›s multiple-range test were used to test for significant differences between treatments. The results show a decrease in pH and an increase in titratable acidity (TTA). It was also observed that the degree of hydrolysis and the percentage of biogenic amines increased during the fermentation period. The growth of lactic acid bacteria (LAB) was also observed, with sharp rise levels on the ٥th day, and the growth becomes less within ١٥–٥ days of fermentation.»

Keywords: Lactic acid bacteria, pH, degree of hydrolysis, histamine, tyramine

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Introduction:

Recent trends in food security are fueling an increased interest in trace compounds that can affect human health, such as biogenic amines. Biogenic amines comprise organic bases with aliphatic (putrescine, cadaverine, spermine, and spermidine), aromatic (tyramine and 2-phenylethylamine), or heterocyclic (histamine and tryptamine) structures, which may be present in various foods (Ruiz-Capillas & Herrero, 2019). They are primarily produced by the microbial decarboxylation of amino acids, except for physiological polyamines. While biogenic amines may occur endogenously at low concentrations in non-fermented foods like fruits, vegetables, meat, milk, and fish, higher concentrations are often found in fermented foods due to contaminating microflora exhibiting amino acid decarboxylase activity (Saha, Turna, Chung & McIntyre, 2024). These compounds pose health risks, particularly to sensitive individuals, leading to symptoms such as nausea, respiratory distress, hot flashes, cold sweats, heart palpitations, headaches, red rashes, hypotension, and hypertension (Vally & Misso, 2012).

The estimation of biogenic amines, including histamine, tyramine, 2-phenylethylamine, agmatine, putrescine, and cadaverine, is crucial not only from a toxicological perspective but also as indicators of food freshness or spoilage (Alberto, Arena & De Nadra, 2004). Establishing maximum permissible levels for biogenic amines in foods is challenging due to individual responses and the presence of other amines. Proposed acceptable levels for fermented foods range from 50–100 mg/kg for histamine, 100–800 mg/kg for tyramine, and 30 mg/kg for 2-phenylethylamine (Doeun, Davaatseren & Chung, 2017). Similarly, an acceptable level of 1 g/kg for total biogenic amine content has been suggested. Some studies have reported a maximum allowable limit of 100 mg/kg of histamine (Yu et al., 2021).

Research on the types and quantities of biogenic amines in traditional foods such as Shalgam, Sunki, and Douchi has been conducted using HPLC. Understanding the correlation between grain proportions and biogenic amine production in fermented foods is essential for developing fermentation processes and ensuring food safety and quality (Gong et al., 2014).

Cereal-based fermented foods, which are staple foods in many regions including Asia, Africa, the Middle East, and parts of Europe, play a significant role in nutrition. Tarasas, a traditional fermented Kurdish food, is produced using lactic acid bacteria through spontaneous fermentation (Phiri et al., 2019). It consists of turnips, leaves, and roots mixed with cooked wheat grits. Homemade production of Tarasas involves chopping turnip roots and leaves, adding them to cooked wheat grits with salt and fermenting the mixture at 25°C for 15 days.

The current study aims to investigate the effects of adding wheat grits in different ratios on the production of biogenic amines in fermented Tarasas. By doing so, we aim to enhance the safety and nutritional profiles of fermented Tarasas while gaining insights into the complexity of biogenic amine production. **Materials and Method:**

According to the traditional Kurdish recipe, 2.0 kg of fresh turnip roots and 2.0 kg of turnip leaves were washed and cut into small sections or slices. Subsequently, 1.0 kg of wheat groats were boiled until softened, cooled at room temperature (25 °C), and then added to the mixture of turnip roots and leaf slices. Finally, 225

gm of salt was added. The treatments were placed in a clean container with a well-covered lid and stored at room temperature to start fermentation.

After 5 days of fermentation, the sample underwent phase separation, with the upper layer being removed and placed at the bottom, while the lower layer was returned to the top without agitation. The container was then sealed and left at room temperature to complete the fermentation process until it reached 15 days. At this point, the product was mixed thoroughly and became ready for further laboratory analysis.

Chemical Examination

The pH and Titratable Acidity (TTA) Determination

The pH of Tarasas treatments was measured using a pH meter (HI-2210-02 pH Bench Meter, Hanna Instruments) after calibrating the electrode meter with a fresh standard buffer at a pH of 4.0 (Daji et al., 2022). Titratable acidity was determined following the AOAC method of analysis (Lane, 1995) and expressed as a percentage of lactic acid. All measurements were conducted in triplicate.

Total Nitrogen (TA), Soluble Nitrogen (TCA-SN), and Degree of Hydrolysis

The total nitrogen was determined, and trichloroacetic acid-soluble nitrogen (TCA-SN) was determined by the Kjeldahl method as described by Tomita, Nakamura & Okada (2018). The degree of hydrolysis of nitrogen was obtained by dividing the TCA-SN result by the total nitrogen and multiplying by 100, as described by Fong et al. (2020).

Analysis of Biogenic Amines

Extraction and analysis of biogenic amine compounds in Tarasas treatments during different periods of fermentation were performed using the HPLC method (Shori & Baba, 2012).

Lactobacillus Enumeration

The enumeration of Lactobacillus in Tarasas was conducted using the standard plate-counting method. About 11 g of the sample was diluted 10-fold with 99 mL of sterile saline solution. Then, 0.1 mL of the diluted sample was inoculated onto MRS agar and M17 agar. The plates were incubated at 37 °C for 24 hours.

Statistical Analysis

Results were analyzed using the Statistical Package for the Social Sciences (SPSS, version 25, Chicago, USA). One-way analysis of variance was used to test the effect of the fermentation process on the detection of biogenic amines. Duncan's multiple range test was conducted to test the significant differences between the means of the treatments at a level of $p \leq 0.05$.

Result and Discussion:

The pH, titratable acidity, total nitrogen, soluble nitrogen, and degree of hydrolysis were assessed before the fermentation process (day zero), and subsequently after 5, 10, and 15 days for all treatments. Observations of pH and acidity changes in Tarasas during and after fermentation at 25°C indicated a pH decrease accompanied by increased acidity by the fifth day of fermentation. Additionally, acidity levels exhibited a slight rise on days 10 and 15 of fermentation (Table 1). These alterations may be attributed to microbial activity primarily responsible for converting sugars present in raw materials (such as turnip roots or grains) into various organic acids, including lactic acid, as metabolic byproducts. Consequently, this process leads to a decrease in pH and an increase in titratable acidity (Yassunaka Hata et al., 2023). Moreover, longer fermentation durations generally yield higher levels of acid production and result in lower pH levels, establishing an osmotic stress environment for microorganisms by extracting water from their cells. This phenomenon can impede the growth of numerous microorganisms, including spoilage bacteria (Park, Zhang & Kim, 2022).

Table 1: The levels of the factors affected by Lactobacillus and yoghurt in Tarasas, a Kurdish traditional food, in three periods.

Trait	Treatment (% wheat ratio)	0 day	5th day	10th day	15th day
pH	T	5.80	3.30	3.20	3.10
	T^{100}	5.80	3.20	3.20	3.10
	T^{75}	5.80	3.30	3.30	3.20
	T^{50}	5.70	3.30	3.20	3.20
	T^{25}	5.80	3.20	3.20	3.10
Treatable acidity	T^{0}	0.91	1.97	1.97	2.02
	T^{100}	0.89	1.96	1.96	2.01
	T^{75}	0.90	1.96	1.96	2.01
	T^{50}	0.93	1.96	1.96	2.00
	T^{25}	0.92	1.94	1.94	2.02
Total nitrogen %	T^{0}	1.16	1.15	1.11	1.10
	T^{100}	0.82	0.81	0.77	0.75
	T^{75}	0.57	0.56	0.54	0.53
	T^{50}	0.42	0.41	0.39	0.38
	T^{25}	0.22	0.21	0.19	0.18
Soluble nitrogen %	T^{0}	0.009	0.590	0.701	0.840
	T^{100}	0.006	0.350	0.440	0.470
	T^{75}	0.003	0.196	0.210	0.291
	T^{50}	0.001	0.115	0.125	0.174
	T^{25}	0.001	0.034	0.044	0.056
Degree of hydrolysis %	T^{0}	0.83	51.50	63.22	77.04
	T^{100}	0.77	43.70	58.07	63.05
	T^{75}	0.54	35.40	40.05	55.01
	T^{50}	0.41	27.20	32.11	46.03
	T^{25}	0.22	16.10	23.012	31.51
	T_{0}				

The results presented in Table 1 also demonstrate a variation in the total nitrogen percentage within Tarasa's product at day zero, ranging from 1.16% to 0.22% across all treatments. This slightly declined to a range of 1.10% to 0.18% by the 15th day of fermentation. This study suggests a direct relationship between total nitrogen and the proportion of wheat grits utilized, as wheat grits represent the primary nitrogen source due to their elevated amino acid content compared to turnip roots and leaves, which contain lower nitrogen levels (Wang et al., 2021).

During fermentation, a slight decrease in nitrogen was observed, attributed to microorganisms consuming nitrogen from various ingredients, notably wheat grits. Nitrogen is crucial for microbial growth and

metabolism, particularly for lactic acid bacteria (LAB) involved in Tarasa's fermentation. While wheat grits may not inherently contain abundant free nitrogen compounds, they do contain proteins that LAB can break down into peptide chains and amino acid units, serving as nitrogen sources during fermentation. Microorganisms, particularly bacteria, utilize these nitrogen-containing compounds for amino acids and other essential nutrients, thereby facilitating microbial growth and metabolic byproduct production (Thomas & Ingledew, 1990).

Moreover, the variation in nitrogen ratios in Tarasa's product may be attributed to differences in nitrogen content among its ingredients (wheat grits, turnip roots, and leaves), influenced by factors such as wheat variety, growth conditions, and processing methods. Turnip roots also contribute nitrogen through proteins, potentially at varying concentrations from wheat. The nitrogen levels in roots are influenced by factors including plant genetics, soil composition, and agricultural practices (Adamczyk et al., 2010).

Turnip leaves typically exhibit higher nitrogen content than roots, being richer in proteins and other nitrogen compounds. The nitrogen content in leaves varies depending on factors such as plant age, environmental conditions, and nutrient availability. Moreover, soluble nitrogen (SN) increased with prolonged fermentation time, as evidenced by the rise in SN-TCA levels across all Tarasas treatments. Initially, SN-TCA levels ranged from 0.009 to 0.0004 at day zero, increasing to levels between 0.88 and 0.056 by the 15th day of fermentation (Duan, 2023). Furthermore, our observations from Table 1 suggest a correlation between the decrease in SN-TCA and the reduction in wheat grits ratio in the treatments.

The increase in soluble nitrogen and heightened metabolic activity of microorganisms during Tarasas fermentation facilitate the synthesis and release of nitrogen-containing compounds, which are attributable to various microbial and biochemical processes inherent to Tarasas fermentation. Fluctuations in pH levels during fermentation can impact enzymatic activities and SN-TCA levels. Additionally, these pH fluctuations can influence the production of SN-TCA (Calsamiglia, Ferret & Devant, 2002).

The degree of hydrolysis (DH), calculated as (SN-TCA/total nitrogen) × 100, showed a significant increase with prolonged fermentation time across all treatments. At day zero, DH values ranged from 0.86% to 0.2%, escalating to 77.04% to 29.3% after 15 days of fermentation (Table 1). The primary disparities in DH observed at equivalent fermentation times stem from variations in wheat grit levels in the product. DH demonstrates a direct correlation with the level of wheat grits. Increased bacterial growth enhances metabolic activity, thereby promoting the breakdown of large proteins through the secretion of proteolytic enzymes, which convert them into peptides. An increase in the degree of decomposition reflects the success of the fermentation process (Kieliszek et al., 2021).

Biogenic amines detection:

The concentrations of biogenic amines (in mg/kg) generated with varying concentrations of wheat grits after the 5th, 10th, and 15th days of fermentation are detailed in Tables 2, 3, and 4, respectively. The estimation encompassed all biogenic amines, including histamine, agmatine, β-phenylethylamine, putrescine, cadaverine, tyramine, spermidine, and spermine.

"The objective of the current study was to assess the levels of biogenic amines in the traditional food Tarasas using varying levels of wheat grits. Biogenic amines were evaluated in the raw materials employed (turnip leaves and roots) as well as in wheat grits before mixing and fermentation. The results indicated that turnip leaves and roots contained 0.96 µg/kg of β-phenylethylamine, 0.076 µg/kg of cadaverine, and 0.89 µg/kg of histamine. In contrast, wheat grits contained 0.8 µg/kg of β-phenylethylamine, 0.086 µg/kg of cadaverine, 0.44 µg/kg of histamine, 0.8 µg/kg of putrescine, 1.04 µg/kg of agmatine, 0.66 µg/kg of tyramine, 0.92 µg/kg of spermidine, and 0.5 µg/kg of spermine

The data presented in Tables 2, 3, and 4 illustrate a consistent increase in the percentage of biogenic amines with prolonged fermentation time. For instance, the concentrations of histamine and tyramine escalated from 73.9 mg/kg and 49.3 mg/kg after 5 days of fermentation to 86.77 mg/kg and 59.3 mg/kg after 10 days, respectively, eventually reaching 107 mg/kg and 73.6 mg/kg after 15 days. These findings indicate a progressive rise in the levels of biogenic amines, including histamine, tyramine, and other identified compounds, throughout the fermentation period.

Table 2: The levels of biogenic amines (mg /kg) among the treatments on the 5th day of fermentation.

Tr.	Histamine	Agmatine	B-phenylethylamine	Putricne	Cadaverine	Tyramine	Spermidine	Spermine	The total amount of BA
T_{100}	73.90±0.01 a	68.00±0.01 a	51.20±0.01 a	54.90±0.01 a	61.50±0.01 a	49.30±0.01 a	52.10±0.01 a	49.90±0.01 a	460.8±0.01
T_{75}	64.70±0.01 b	56.57±0.01 b	43.60±0.01 b	51.60±0.01 ab	58.50±0.01 ab	44.60±0.01 b	49.90±0.01 ab	42.30±0.01 b	411.77±0.01
T_{50}	56.30±0.01 c	48.30±0.01 c	42.47±0.01 b	40.20±0.01 c	54.10±0.01 b	35.70±0.01 c	38.00±0.01 c	41.10±0.01 b	356.17±0.01
T_{25}	41.50±0.01 d	31.80±0.01 d	37.50±0.01 c	33.70±0.01 d	42.00±0.01 c	33.10±0.01 c	30.50±0.01 cd	35.60±0.01 c	285.7±0.01
T_{0}	19.60±0.01 e	26.30±0.01 e	33.30±0.01 e	29.90±0.01 e	25.70±0.01 d	22.30±0.01 d	25.70±0.01 d	28.70±0.01 d	211.5±0.01

Tr.=Treatment, T_{100}=100% wheat grits, T_{75}= 75% wheat grits, T_{50}=50% wheat grits, T_{25}=25% wheat grits, T_{0}=0% wheat grits, Values with different superscripts in a column are significantly different (P<0.05).

Table 3: The levels of biogenic amines (mg /kg) among the treatments on the 10th day of fermentation.

Tr.	Histamine	Agmatine	B-phenylethylamine	Putricne	Cadaverine	Tyramine	Spermidine	Spermine	The total amount of BA
T_{100}	86.77±0.01 a	70.480±0.01 a	75.10±0.01 a	63.50±0.01 a	91.80±0.01 a	59.30±0.01 a	69.50±0.01 a	56.70±0.01 a	573.15±0.01
T_{75}	76.90±0.01 b	63.70±0.01 b	59.50±0.01 b	59.30±0.33 b	90.10±0.01 a	51.60±0.12 b	59.90±0.12 b	49.30±0.01 b	510.3±0.01
T_{50}	73.80±0.01 bc	57.80±0.01 c	48.57±0.01 c	56.87±0.02 bc	83.90±0.01 b	47.00±0.22 c	44.90±0.11 c	44.90±0.01 c	457.74±0.01
T_{25}	61.30±0.12 c	52.60±0.01 cd	42.47±0.01 d	53.05±0.01 c	76.20±0.2 c	40.30±0.01 d	40.80±0.12 c	41.40±0.01 c	407.72±0.01
T_{0}	34.10±0.01 d	45.30±0.01 d	40.00±0.02 d	41.28±0.11 d	34.90±0.01 d	35.8±0.12 e	31.00±0.01 d	35.00±0.01 d	297.38±0.01

Tr.=Treatment, T_{100}=100% wheat grits, T_{75}= 75% wheat grits, T_{50}=50% wheat grits, T_{25}=25% wheat grits, T_{0}=0% wheat grits, Values with different superscripts in a column are significantly different (P<0.05).

Table 4: The levels of biogenic amines (mg /kg) among the treatments on the 15th day of fermentation.

Tr.	Histamine	Agmatine	B-phenylethylamine	Putricne	Cadaverine	Tyramine	Spermidine	Spermine	The total amount of BA
T_{100}	107.00±0.01 a	74.40±0.01 a	84.70±0.01 a	87.03±0.01 a	110.20±0.01 a	73.60±0.02 a	76.30±0.01 a	65.80±0.01 a	679.03±0.01
T_{75}	103.30±0.01 a	68.00±0.21 b	77.00±0.01 b	82.80±0.12 ab	107.30±0.01 a	65.10±0.12 b	64.80±0.01 b	58.60±0.01 b	626.9±0.01
T_{50}	99.00±0.01 b	64.00±0.11 bc	71.40±0.01 bc	79.50±0.12 b	102.40±0.01 ab	56.60±0.02 c	55.90±0.01 c	50.00±0.01 c	578.8±0.01
T_{25}	86.30±001 c	58.60±0.01 c	62.47±0.01 c	77.47±0.12 b	93.10±0.01 b	51.80±0.01 d	45.60±0.01 d	45.80±0.01 d	521.14±0.01
T_{0}	47.60±0.01 d	51.30±0.01 d	57.30±0.01 d	68.60±0.01 c	40.60±0.01 c	47.10±0.01 d	35.60±0.01 e	38.00±0.01 e	386.1±0.01

Tr.=Treatment, T_{100}=100% wheat grits, T_{75}= 75% wheat grits, T_{50}=50% wheat grits, T_{25}=25% wheat grits, T_{0}=0% wheat grits, Values with different superscripts in a column are significantly different ($P<0.05$).

The decrease in the percentage of wheat grits is noted to have a significant impact on the reduction in the rate of biogenic amine formation. This suggests a direct relationship between the percentage of wheat grits and biogenic amine formation, potentially due to the presence of amino acids such as histidine, tyrosine, and tryptophan in wheat. These amino acids can be decarboxylated by LAB bacteria, leading to the production of biogenic amines such as histamine, tyramine, and tryptamine (Molina-Gutierrez et al., 2022). Increased wheat grits provide more nutrients, which play a vital role in providing essential nourishment for LAB during Tarasas fermentation.

Certain strains of LAB bacteria can generate biogenic amines as by-products of metabolism during their growth and fermentation process of Tarasas (Slizewska & Chlebicz-Wojcik, 2020). Enhanced microbial activity resulting from increased nutrients from wheat grits can lead to elevated production of biogenic amines. Factors such as time, ingredient ratios, with wheat grits being particularly important, and pH can also influence the growth and metabolic activities of LAB, potentially leading to increased biogenic amine production (Li et al., 2023; Bukvicki et al., 2020).

The pH level plays a significant role in the activity of amino acid decarboxylase, with higher enzyme activity observed within the pH range of 4 to 5.5 (Zapasnik, Sokolowska & Bryla, 2022). It is important to note that the specific effects of increasing wheat grits on biogenic amine production can vary depending on the type and strain of LAB present during fermentation, the specific fermentation process used, and the overall quality and handling of the wheat grits. Furthermore, the concentrations of biogenic amines are influenced by the storage period, which increases during fermentation (Wu et al., 2022).

The allowable ratio for consuming biogenic amines varies depending on the specific amine and individual health conditions, and the country and specific regulations in place (Christensen et al., 2022). In the European Union, the allowable histamine level in fishery products is 100 mg/kg, except for certain specific products such as tuna, mackerel, and sardines which have a higher limit of 200 mg/kg (Zdziobek, Jodlowski & Strzelec, 2023). While in the United States, the allowable histamine level in finfish and shellfish is 50 mg/100g. The U.S. Food and Drug Administration (FDA) recommends that histamine levels in fish products should not exceed 50 parts per million (ppm) to minimize the risk of scombroid fish poisoning (Bintsis & Papademas, 2022).

When it comes to the potential toxicity of biogenic amines, studies have suggested that consumption of 100-800 mg/kg of tyramine and 30 mg/kg of β-phenylethylamine in foods can be considered toxic doses. Additionally, it is suggested that the upper limit for human consumption of histamine in foods should be around 100 mg/kg (Bintsis & Papademas, 2022), and Body et al. (2021) mentioned that histamine levels above 500 mg/kg or tyramine levels above 1000 mg/kg are considered toxic and dangerous for human health (Qin et al., 2022). Different countries have established upper limits of histamine in wine: 2 mg/L (Germany), 3.5 mg/L (Netherlands), 5 mg/L (Finland), 6 mg/L (Belgium), 8 mg/L (France), and 10 mg/L (Australia and Switzerland) (Akamine, Mansoldo & Vermelho, 2023). EFSA suggested that the upper limit for human consumption of histamine in fermented vegetables is 92 mg/kg, tyramine is 91 mg/kg, putrescine 549 mg/kg, cadaverine 94 mg/kg, Phenylethylamine <5 mg/kg, and suggest the upper limit of the sum of biogenic amines are 747 mg/kg. Generally, it is recommended to consume biogenic amines in moderation to avoid any potential health risks.

Detection of Lactic acid bacteria during fermentation:

The enumeration of Lactic Acid Bacteria (LAB) in Tarasas treatments was conducted using MRS and M17 media at various fermentation intervals (0, 5, 10, and 15 days) at 25°C, as depicted in Table 5. The log (CFU/g) of LAB was determined on both MRS and M17 media. Generally, the LAB counts obtained from MRS media exceeded those from M17. At the onset of fermentation, the LAB count in T100 was 6.36 log (CFU/g) on day 0, which increased to 8.96 log (CFU/g) by the 15th day. The growth of LAB on the M17 medium exhibited a count of 6.22 (CFU/g) in T100 on day 0, rising to 7.7 (CFU/g) by the 15th day of fermentation.

LAB counts significantly decreased in treatments T75–T0 during fermentation compared with treatment T100. The reduction in the percentage of wheat grits in the fermentation substrate resulted in decreased availability of nutrients for the bacteria. Consequently, the growth rate of LAB may decline, leading to a reduction in their population. This phenomenon is likely attributed to the limited availability of essential nutrients required for bacterial growth and metabolism.

Additionally, the decrease in the percentage of wheat grits may alter the overall composition of the fermentation substrate, affecting factors such as pH, moisture content, and the presence of other microorganisms. These changes can further influence the growth and activity of LAB during fermentation, consistently exhibiting the highest LAB count on both MRS and M17 media throughout the fermentation period compared to other treatments.

The initial count of Lactic Acid Bacteria (LAB) at the onset of fermentation was low, undergoing rapid proliferation during the spontaneous fermentation period between day zero and day five, followed by a slower increase thereafter. The activity of LAB significantly escalated alongside the observed decline in pH levels during various fermentation periods. The optimal pH range for Lactobacillus species typically falls between 4.0 to 7.0, though specific pH requirements may vary among different strains. Conversely, Lactococcus species prefer a pH range of 5.5 to 6.8, with optimal growth occurring around 6.5. These findings are consistent with previous studies that have identified a positive correlation between decreasing pH levels and increased LAB abundance during fermentation periods (Barcenilla et al., 2022). Furthermore, this study observed a higher increase in LAB quantity during fermentation stages (5, 10, and 15 days), which corroborates findings by Lane (1991).

It has been documented that Lactobacillus is the predominant bacteria found in fermented foods, alongside

certain strains of Lactococcus, Streptococcus, and Leuconostoc (Muyzer, 1999). Additionally, in agreement with Chan et al. (2021), the decrease in pH levels observed during the fermentation of cabbage, from 6.0 to 3.4 over 5 days, can be attributed to the activity of LAB, specifically Lactobacillus plantarum and Lactococcus lactis. LABs produce lactic acid as a metabolic byproduct during fermentation, leading to acidification of the environment. This acidic environment can positively influence the activity of hydrolytic enzymes. Many enzymes involved in hydrolysis reactions are pH-sensitive, and the acidic conditions created by LAB can optimize the activity of these enzymes.

Table 5: Log10 (CFU/g) of LAB on MRS, M17 during (0, 5, 10, 15) days of fermentation for different treatments.

Tr.	MRS media				M17 media			
	Zero-day	5th day	10th day	15th day	Zero-day	5th day	10th day	15th day
T_{100}	6.36±0.12 a	8.88±0.01 a	8.91±0.01 a	8.96±0.01 a	6.22±0.01 ab	7.56±0.01 a	7.67±0.01 a	7.70±0.01 a
T_{75}	6.29±0.03 b	8.84±0.01 ab	8.87±0.01 b	8.89±0.01 b	6.19±0.01 b	7.45±0.03 bc	7.60±0.01 b	7.66±0.01 b
T_{50}	6.15±0.01 c	8.71±0.01 bc	8.83±0.01 c	8.85±0.01 c	5.63±0.01 g	7.40±0.01 c	7.48±0.33 c	7.56±0.01 d
T_{25}	6.08±0.02 d	8.63±0.21 c	8.75±0.11 d	8.78±0.23 d	5.35±0.01 d	7.32±0.01 e	7.41±0.01 e	7.47±0.01 f
T_{0}	5.96±0.01 e	8.57±0.01 d	8.67±0.02 e	8.74±0.01 e	5.24±0.01 a	7.25±0.01 g	7.32±0.02 g	7.40±0.01 fg

Conclusion:

In conclusion, the observed increase in biogenic amines in the Tarasas product due to the presence of wheat grits can be attributed to specific compounds, such as histidine and tyrosine, which serve as precursors for biogenic amine formation. These compounds undergo enzymatic conversion by lactic acid bacteria during consumption, resulting in elevated biogenic amine levels. This elevation poses potential health risks, as high concentrations of biogenic amines have been linked to adverse effects such as allergic reactions and migraines. Therefore, it is imperative to carefully assess the impact of wheat grits on biogenic amine levels and the associated health risks before consumption.

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Clinical Characteristics of Breast Cancer in AYA, Kurd, Iraq

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Abstract:

This study investigates the clinical characteristics and outcomes of breast cancer among Adolescent and Young Adult (AYA) Kurdish women in Iraq, focusing on the interplay of genetic, environmental, and socioeconomic factors. It aims to illuminate the specific clinical patterns and decision-making processes within this demographic, highlighting the influence of cultural and healthcare infrastructure on patient care. A retrospective cohort analysis was conducted, examining medical records of AYA Kurdish women diagnosed with breast cancer between January ٢٠١٠ and December ٢٠٢٠. Data on age, tumour characteristics, treatment modalities, and comorbidities were analyzed, with statistical methods employed to identify significant trends and associations. The study found a higher prevalence of breast cancer in the older segment of the AYA group, with a notable preference for Wide Local Excision (WLE) over mastectomy. Decision-making regarding treatment was influenced by both medical professionals and familial input. A high incidence of comorbidities was observed, affecting treatment choices and outcomes. Educational level significantly correlated with the type of surgical intervention chosen. Breast cancer in AYA Kurdish women exhibits distinct clinical characteristics influenced by a complex interplay of cultural, educational, and health system factors. The findings underscore the need for culturally tailored healthcare approaches and enhanced educational and support mechanisms to improve breast cancer outcomes in this population.

Keywords: AYA Kurdish women, breast cancer, clinical characteristics, treatment decisions, comorbidities, Iraq.

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Introduction:

Cancer of the breast in adolescents and young adults (AYA) (Imperially), specifically within the Kursk population of Iraq, is a major medical and social problem. Among various adult age groups, the demographic population, in this case, refers to the 15-39-year-old individuals who clinically seek more distinct approaches compared to women above the age of 40 with breast cancer. The Kurdish region of Iraq like every other place comprises a unique mix of genetic, environmental and lifestyle factors therefore a detailed analysis of breast cancer patterns in this region has to be done(1, 2).

Breast cancer in AYAs is usually found in a later stage of disease progression and with a harder tumour type for young women than it is in older women. First, AYAs face a higher possibility of being diagnosed with high-grade tumours compared to other people similar age to them, furthermore, they are more prone to have a triple-negative breast cancer subtype that most of the time correlated with poorer prognosis. It is the specific socio-economic and cultural details of the Kurdish country such as restricted health problem sources, possible genetic predispositions and way of life stance, that might have been accepted to the disease's clinical manners and outcomes(3).

The oncological research has underlined the necessity of investigation of breast cancer in this group of people because they might have certain biological and clinical features that are different from those of older patients. The studies showed that younger patients more often have a disease with more aggressive progression and specific treatment challenges. In the case of ethnic Kurds, these difficulties are multiplied by geopolitical determinants, limited health infrastructure, and cultural barriers that hinder diagnostic measures, treatment, and psychological support(4).

This article is dedicated to the investigation of the clinical features of breast cancer whose victims are AYA group and whose medical care takes place within the Kurdish system of Iraqi healthcare. Particular attention will be given to the questions of hereditary predisposition as well as environmental and lifestyle factors involved, the disease presentation and the outcomes. Since these issues are worth mentioning thus the study aims to find all the aspects of the condition and therefore to provide a complete understanding of the whole landscape of the disease among this special population, make contributions to the global research on the quality of life in YA Canadian and present accordingly specific health strategies for the Kurds patients.

Epidemiological Trends of Breast Cancer in AYAs

Cancer diagnosed in adolescents or young adults is a rarity, but it frequently varies more aggressively compared to adults or senior patients. Research on AYA patients with breast cancer has indicated that AYA patients are more likely to be diagnosed with cancers that have a higher grade and are of larger tumour size, especially at advanced stages. Within the frame of the AYA Kurdish population, breast cancer incidence and predominant age group continue to show some uncertainties. However, local studies conducted within the region show that the number of young women with breast cancer is on the rise. Hence, a targeted assessment should be done on them(5).

Genetic and Molecular Characteristics in AYA Breast Cancer

Genetic predisposition is a matter or one of the most factors in the etiology, or development, of Breast Cancer in young adults. Breast cancer patients with BRCA mutations including BRCA1 and BRCA2 are often younger during diagnosis, which manifests as aggressive diseases in this category. Changes in the genotype of genes related to breast cancer or their interactions with the ageing of AYA's expedition may result in the peculiarity of breast cancer presentation among teens(6).

Impact of Socio-economic and Cultural Factors

The social-economic and cultural differences determine the level of breast cancer awareness, screening, and treatment access. For young patients with breast cancer in the Kurdish regions of Iraq, problems such as political-socio upheaval, shortage of economic means, and cultural norms may delay diagnosis and limit access to comprehensive cancer care resulting in worse survival rates and quality of life(7).

Clinical Outcomes and Survival Rates

The observation is that YAs with breast cancer have less obtained survival rate compared to older women who discuss more aggressive disease traits and their most important delay in diagnosis as the reasons. The literature is very scarce on the Kurdish population but according to the existing research, cultural stigma and healthcare access issues may further make things complicated for the people, reducing the rate of detection and linkage of treatment which may in turn affect the overall clinical outcome(8).

Psychosocial Aspects and Quality of Life

The mental health impact of breast cancer in AYAs is so distinct that it affects self-confidence, body image, fertility problems, and also social circle. The cultural environment of the Kurdistan region where family and social support are strongly connected with the psychosocial experiences and support needs of the young Kurdish women diagnosed with breast cancer(9).

Treatment Strategies and Therapeutic Challenges

The treatment of breast-cancer AYAs requires a special approach being the fact that the disease is quite aggressive to expose an individual to the possible impact on fertility. Chemotherapy, radiation, and surgical decisions must be scrutinized relative to the preservation of fertile potential and future hormonal balance. The difficulty of obtaining high-level oncological facilities in the Kurdish context and their low availability of fertility preservation tools are the most serious issues that may appear ineffective treatment. Research has revealed the deficient oncology services and low level of patient education as the areas to be improved to enhance the treatment outcomes in the region(10).

Breast Cancer Awareness and Screening Programs

According to studies, the younger group of diagnosed breast cancer prognosis is altered compared to other stage-of-life bearers with a diagnosis delay. Young girls now become less interested or cannot properly understand the situation which causes them to fail to prevent themselves from the disease. They often do not care about screening programs. The influence of the strong traditions of this region over the young generation is of particular importance in Kurdistan. Research revealed that you can achieve this through deliberate public awareness campaigns as well as the age-selective screening of the AYA population. The early detection will give the users a chance for a higher cure rate(11).

The Role of Lifestyle and Environmental Factors

Human behaviour and environmental influences (this could refer to diet, physical activity, and exposure to environmental toxins) have a suggestion linking to breast cancer risks and its progression. In the Kurdish region where the changes from traditional lifestyles are taking place, the establishment of the link between these factors on AYA breast cancer incidence and progression is irreplaceably important for creating the appropriate prevention strategies(12).

Material and methods:

Study Design

Objective: To investigate the clinical characteristics and outcomes of breast cancer among the AYA Kurdish population in Iraq.

Study Type: A retrospective cohort study analyzing patient records from multiple Hiwa cancer and haematology hospitals and the Cancer Control Program in Sulaimaniyah governorate-Kurdistan, Iraq.

Population and Sample

Population: AYA Kurdish females diagnosed with breast cancer.

Inclusion Criteria: Female patients, aged 15-39, of Kurdish ethnicity, diagnosed with breast cancer between January 2010 and December 2020.

Exclusion Criteria: Patients with recurrent breast cancer or with incomplete medical records.

Sample Size Calculation: Based on previous prevalence rates, with an expected margin of error and confidence level.

Data Collection

Sources: Medical records from oncology departments in Hiwa Cancer and Hematology Hospital and the Cancer Control program in Sulaimaniyah City Kurdistan, Iraq.

Data Points: Age at diagnosis, histological type, tumour size, nodal status, metastasis, treatment modalities, and survival outcomes.

Data Collection Period: January 2010 to December 2020.

Ethical Considerations

Ethics Approval: Obtain from the institutional review board of the participating hospitals.

Informed Consent: Waived due to the retrospective nature of the study, ensuring patient anonymity and data confidentiality.

Data Analysis

Statistical Methods: Descriptive statistics for demographic and clinical characteristics, Chi-square test used to find association between variables.

Software: Use statistical software like SPSS version 21

Results:

Table 1: Demographic data of AYA patients

A study of the clinical features of breast cancer in adolescent and young adult Kurdish women in Iraq demonstrates that they are somehow different from the rest of the population in terms of demography and clinical presentation. The age distribution of attendees has an age majority population of older aged AYA individuals, i.e. 52.9% of them are between 40-49 years old and the remaining 35.3% are in the 30-39 age group. Young ones (sub-20 years age group) variation minimalizes by zero four (0.4%).

In terms of residency, most of the women 68.1%) in the study were from urban zones as compared to 31.9% who were in rural areas. Marital status depicted that a great percentage of the respondents (82%) were accessible those who were termed as single individuals accounted for 15.4%, divorced 2.4% and widowed were not detectably not with the 0.2%.

The larger group of the study population by body mass index (BMI) categories was the one that was obese (42.1%), followed closely by the ones who were overweight (38.1%). Participants in the normal weight category accounted for 18.6% of the total number of individuals while underweight people corresponded to just 1.2% of the sample size.

Educationally, the hippest one was primary level which made up 35.7% of the respondents. This was followed by those who had institute-level education, university degrees, and secondary education to some extent (17.8%, 15.7%, and 14.7% respectively). Of those who participated in the poll and of whom education level was known, 15.4% were illiterate and it was also (0.7) of persons whose educational status remained unknown.

Table 1: Demographic data of AYA patients

Categories	Frequency	Percentage
Age groups/years (Option2)		
<20	**2**	**0.4**
20-29	**29**	**5.8**
30-39	**176**	**35.3**
40-49	**264**	**52.9**
Residency		
Rural	**159**	**31.9**
Urban	**340**	**68.1**
Marital status		
Divorced	**12**	**2.4**
Married	**409**	**82**
Single	**77**	**15.4**
Widowed	**1**	**0.2**
BMI		
Underweight	**6**	**1.2**
Normal weight	**93**	**18.6**
Overweight	**190**	**38.1**
Obese	**210**	**42.1**
Educational level (option2)		
illiterate	**64**	**15.4**
Institute	**74**	**17.8**
Primary	**148**	**35.7**
Secondary	**61**	**14.7**
University	**65**	**15.7**
Unknown	**3**	**0.7**

The results show that Wide Local Excision, took the charge at the rate of 43.9%, most followed by mastectomy at 39.3% and Quadrantectomy was less often applied (2,2%).

As for the surgical intervention choice, the attending surgeon was defined as the primary decision-maker in 34.5% of cases. Joint decisions made together, including family members, happened 27.3% of the time while 29.3% of the time, the relatives were the decision makers. The patient was the one who decided 5% of the cases, and in 4% of the situations, the decision self-evidently failed to identify the one who made the decision. The prevalence of participants' comorbidities is remarkable, as for almost the entire population (86.4%) this category is present, while only 13.6% of those found do not have reported comorbidities.

Table 2: Some clinical history of AYA patients

Categories	Frequency	Percentage
Surgery type		
WLE	**219**	**43.9**
Mastectomy	**196**	**39.3**
Quadrantectomy	**11**	**2.2**
Not done	**73**	**14.6**
Decision making		
Both	**136**	**27.3**
Patient	**25**	**5.0**
Relative	**146**	**29.3**
Surgeon	**172**	**34.5**
Not sure	**20**	**4.0**
History of comorbidities		
Negative	**68**	**13.6**
Comorbidities	**431**	**86.4**

Surgery Type by T-classification

On 36.8% of the diagnosed tumours less than 2 cm T-classification, MS (mastectomies) were performed while on 63.2% of cases Wide Local Excision (WLE). In cases with larger tumours (2-4 cm), 60.8% of the patients were operated on with mastectomy and 39.2% were managed with wide excision procedures. Statistically, the P-value was not significant (0.59), denoting that there was no substantial statistical difference among the surgery types adjusted against the tumour size.

However, a different result was observed, when applying another binary logic-based T-classification method to the tumour and WLE samples of 2 cm and under, with a P-value of 0.027 in favour of MS (49.3%) as compared to WLE (50.7%). A total of 74.1% of the tumours with size 3-5 cm presented the invasive form.

Surgery Type by BMI

The study demonstrates that BMI determines the preferred modality of treatment, as obese patients have more than 50% MS (46.8%) and underweight 83.3% choose mastectomies. There was little difference between groups of BMI categories, and this difference was not statistically significant ($p = 0.319$).

Decision-making for Surgery

The choice of doctors as to the type of surgery was the most significant determinant, mastectomy being the most chosen procedure in 53.6% of cases. Regarding the factor of joint or relative decision-making, the observations that led to Weak Language Engagement were higher, but the variations were not statistically significant (P=0.402).

Procedure by Educational Level of Surgery

Educational level is the other factor that affected whom respondents would rather consult for their surgical operations. The allocated WLE was obtained by the majority of college-educated patients (78.5%), whereas the number of patients who chose the mastectomy undergone by illiterate patients has been seen increased (59.4%). A highly significant difference (p≤0.001) for the patients with university education confirmed the connection between elevated education levels and the choice of WLE.

Table 3: surgery type in relation to some characteristics

Categories	Surgery type				Total	P-value
	MS		WLE			
	No.	%	No.	%		
T-classification(option1)						
< 2	7	36.8	12	63.2	19	0.59
2-4	48	60.8	31	39.2	79	
Total	55	56.1	43	43.9	98	
T-classification (option2)						
≤ 2	35	49.3	36	50.7	71	0.027
3-5	20	74.1	7	25.9	27	
Total	55	56.1	43	43.9	98	
BMI						
Underweight	5	83.3	1	16.7	6	0.319
Normal weight	37	49.3	38	50.7	75	
Overweight	73	45.3	88	54.7	161	
Obese	81	46.8	92	53.2	173	
Total	196	47.2	219	52.8	415	
Decision-making for surgery						
Both	48	42.5	65	57.5	113	0.402
Patient	11	44.0	14	56.0	25	
Relative	52	44.4	65	55.6	25	
Surgeon	82	53.6	71	46.4	153	
Unknown	3	42.9	4	57.1	7	
Total	196	47.2	219	52.8	415	
Educational level (option2)						
illiterate	38	59.4	26	40.6	64	0.034
Institute	31	41.9	43	58.1	74	0.310
Primary	78	52.7	70	47.3	148	0.096
Secondary	34	55.7	27	44.3	61	0.149
University	14	21.5	51	78.5	65	≤0.001
Unknown	1	33.3	2	66.7	3	0.628
Total	196	47.2	219	52.8	415	

Discussion:

Comparison of Surgical Choices

The study indicates a preference for Wide Local Excision (WLE) over mastectomy in the AYA Kurdish population, which contrasts with global trends where mastectomy rates vary. For instance, a study found a higher inclination towards mastectomy in Western populations, particularly in younger women with breast cancer. This discrepancy could be influenced by genetic, environmental, and cultural factors specific to the Kurdish population, suggesting a need for tailored surgical approaches(13).

Decision-making in Surgery

The decision-making process in the Kurdish study leans towards shared decision-making and surgeon influence, similar to findings in Western contexts where patient autonomy and physician recommendation play significant roles. However, the cultural context, as indicated by the influence of relatives in decision-making, reflects a unique sociocultural dynamic in the Kurdish region, underscoring the importance of cultural sensitivity in medical decision-making processes(14).

Prevalence of Comorbidities

The high prevalence of comorbidities (86.4%) in the Kurdish AYA breast cancer population aligns with global observations that comorbid conditions are common in breast cancer patients and impact treatment decisions and outcomes. This similarity stresses the universal challenge of managing breast cancer in the presence of comorbidities, although the specific types of comorbid conditions may vary based on regional health profiles(15).

Influence of Educational Level on Surgery Type

The study's observation that higher educational levels correlate with a preference for WLE over mastectomy is particularly interesting. This finding is supported by research that suggests educated patients are more likely to engage in shared decision-making and opt for breast-conserving surgery due to better awareness and understanding of the disease and treatment options. This connection highlights the role of education in health literacy and decision-making in breast cancer treatment(16).

Tumor Characteristics and Treatment Choices

The study's analysis of surgery type by T-classification revealed that smaller tumours (≤2 cm) were almost equally treated with mastectomy and WLE, a pattern that shifts with larger tumour sizes. This observation aligns with the broader trend in oncological practice where breast-conserving surgery is favoured for smaller, early-stage tumours due to comparable survival rates but better quality of life outcomes However, the higher rate of mastectomy in larger tumours in the Kurdish population may reflect a cautious approach in a resource-constrained setting or a preference shaped by local clinical practice guidelines(17).

Cultural Influences on Health Behavior

The decision-making findings highlight the significant role of relatives and surgeons, underscoring the collective nature of health decision-making in Kurdish society. This is contrasted with Western societies where individual autonomy is often prioritized. The cultural dimension in medical decision-making is crucial, as studies have shown that in collectivist cultures, family involvement in health decisions can provide emotional support and facilitate care, which is vital for patient outcomes(18).

Comorbidities and Breast Cancer Management

The high prevalence of comorbidities in Kurdish AYA patients necessitates a multifaceted approach to cancer care. Research indicates that comorbid conditions can affect the choice of cancer treatment, treatment adherence, and overall survival. In the context of the Kurdish population, where access to comprehensive healthcare might be limited, managing comorbidities presents an additional layer of complexity in breast cancer treatment, suggesting the need for integrated care models that address both cancer and comorbid conditions(19).

Educational Level and Its Impact on Treatment Decisions

study's data showing that higher educational levels are associated with a preference for less invasive surgery reflects broader global patterns where education influences health literacy and empowerment in medical decision-making. This association points to the potential benefits of enhancing educational initiatives focused on breast cancer awareness and treatment options, particularly in regions like Kurdistan where varying educational levels may impact health outcomes(20).

Conclusion

Prevalence and Surgical Choices: The study highlights that breast cancer in the AYA Kurdish population presents primarily in the older age range of this cohort, with a preference for Wide Local Excision (WLE) over mastectomy. This preference suggests a potential shift towards breast-conserving treatments, aligning with global trends for early-stage cancers, yet influenced by local clinical practices and patient preferences.

Decision-making Dynamics: Decision-making in surgical options shows a collaborative approach involving patients, relatives, and surgeons. This underscores the importance of cultural nuances in medical decision-making within the Kurdish population, where family plays a significant role, and the surgeon's influence remains paramount.

Comorbidities: The high incidence of comorbidities among the AYA Kurdish breast cancer patients emphasizes the necessity for an integrated care approach that manages both the primary cancer treatment and the accompanying health conditions, reflecting the global challenge of treating breast cancer patients with multiple health issues.

Impact of Education: The correlation between educational level and the choice of surgery type indicates that higher education levels might be associated with a greater likelihood of opting for breast-conserving surgery (WLE),

pointing to the need for improved educational programs on breast cancer awareness and treatment options in the region.

Cultural and Socioeconomic Factors: The study confirms that cultural and socioeconomic factors significantly impact the clinical characteristics of breast cancer and its management in the Kurdish AYA population. These factors contribute to delayed diagnosis, treatment choices, and potentially the overall prognosis of breast cancer patients in this demographic.

Research Implications: The findings stress the importance of conducting more focused research within the AYA Kurdish population to fully understand the unique epidemiological, clinical, and sociocultural dynamics at play. Such research should aim to develop tailored interventions that address the specific needs and circumstances of this group.

Recommendations:

Enhance Breast Cancer Awareness: Implement comprehensive awareness programs specifically tailored to the AYA Kurdish population. These programs should focus on early detection, the importance of regular screenings, and understanding the options for treatment. Special attention should be given to overcoming cultural barriers and enhancing knowledge about breast-conserving treatments.

Improve Access to Healthcare Services: Strengthen healthcare infrastructure and ensure that AYA Kurdish women have access to specialized breast cancer care, including advanced diagnostic tools, treatment facilities, and post-treatment support, regardless of their geographical location or socioeconomic status.

Culturally Sensitive Care Models: Develop and implement culturally sensitive healthcare models that consider the significant role of family and community in Kurdish society. Training healthcare professionals to understand and respect these cultural nuances can facilitate better communication, decision-making, and patient satisfaction.

Integrated Care for Comorbidities: Establish integrated care pathways that address not only breast cancer but also the common comorbid conditions found in the study population. This holistic approach should include multidisciplinary teams comprising oncologists, primary care physicians, nutritionists, psychologists, and other specialists as needed.

Educational Programs and Patient Empowerment: Create targeted educational initiatives to raise the literacy level regarding breast cancer among AYA Kurdish women. Empower patients by providing them with the information necessary to make informed decisions about their treatment options and promote the understanding of the benefits of early detection and treatment.

Research and Surveillance: Encourage and support ongoing research into the epidemiological and clinical aspects of breast cancer in the Kurdish population. Establish a surveillance system to monitor trends in breast cancer incidence, treatment outcomes, and patient survival, which can inform public health strategies and healthcare practices.

Fertility Preservation and Psychosocial Support: Address the need for fertility preservation options and psychosocial support as part of the cancer care continuum for AYA Kurdish patients. Offering counselling and support services can help patients cope with the psychological impact of breast cancer and treatment-related decisions.

Strengthen Public Health Policies: Advocate for the development of public health policies that support early detection programs, provide funding for breast cancer research in AYA populations and ensure equitable access to care across all segments of the Kurdish population.

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Utilization of Restricted feed in various time on Protein and Energy Efficiency of Female Broiler Chicks

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Abstract

Considerable research has investigated the potential benefits of reducing protein content in broiler chicken diets, yielding varied outcomes. Broiler feed intake has been restricted through various regimens, which have demonstrated reductions in feeding expenses, decreased fat deposition, enhanced feed efficiency, and lowered incidence of metabolic disorders. This study aimed to elucidate the advantages of feed restriction on the energy and protein efficiency of female broiler chicks. The experiment involved four treatments, each with five replicates, conducted on 300-day-old broiler chicks. The chick groups were randomly allocated into 20 groups of 15 chicks, with similar average body weights per pen, and subjected to four distinct treatments with five replicates each. The control group received ad libitum feeding, while the second group (T1) experienced restricted feeding for 4 hours within a 24-hour period (9 am–1 pm). The third group (T2) faced restrictions from 9 am to 5 pm, and the fourth group (T3) from 9 am to 9 pm. Water was the only freely available resource. All birds were fed a standard diet following the suggested technique (NRC, 1994). Over recent decades, significant improvements have been observed in the growth rate, feed efficiency, and meat yield of broiler chickens due to advancements in genetics, nutrition, and environmental control. The imposition of feed restriction notably enhanced weight gain at the experiment's conclusion, particularly evident in the second and third treatments. These differences were statistically significant, resulting in reduced feed consumption and meaningful improvements in food conversion efficiency, protein and energy utilization, and protein and energy conversion rates.

Keywords: Feed Removal, Protein, Energy Efficiency, Female, Broiler Chicks.

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Introduction

The global production of chicken meat is expected to exceed 125.5 million tons in 2020, demonstrating the vital role that the poultry industry plays in satisfying the increasing demand for animal protein. Over the past few decades, advancements in genetics, enhanced nutrition, and regulated environments have led to notable improvements in broiler chicken growth rate, feed efficiency, and meat yield. However, there have been conflicting findings from studies that have examined the advantages and potential of lowering the protein content of broiler chicken diets. prevent fat deposition, increase feed efficiency, and lower the incidence of metabolic disorders in broilers, restricted-feeding regimens have been implemented. The evidence of restricting feed intake can lower mortality rates, fat deposition, and early growth. However, the degree, timing, and duration of the restriction have all been linked to inconsistent findings in recent research. Genetic selection and better nutrition have largely been responsible for the substantial increases in broiler chicken growth rate, feed efficiency, and meat yield over the past few decades (Zubair and Leeson, 1996). It now only takes broiler chickens 33 days to achieve their finishing body weight of roughly 2 kg, thanks in large part to genetic advancement, better feeding, and controlled environments during the past 30 years (Wilson, 2005). In recent research, while with mixed results (Attia et al., 2018; Soares et al., 2020), the benefits and potential of lowering the protein content in broiler chicken diets have been considered. As the main source of protein in field diets, soybean meal is reduced when feed protein levels are lowered. This leads to an increase in synthetic amino acids and maize and a drop in soybean oil or another lipid source (Soares et al., 2020). However, due to the extra-calorie impacts of oils and fats, poultry performance may be negatively impacted by reducing the presence of a lipid source. to limit broiler feed intake, quantitative and qualitative restricted-feeding programs have been used (Tolkamp et al., 2005; Zhan et al., 2007). These initiatives seek to enhance feed efficiency, reduce feeding expenses and fat deposition, limit the negative impacts of fat on human health, and lessen the frequency of metabolic illnesses in birds.

According to studies Navidshad et al., (2006) and Mahmud et al., (2008), feeding restrictions have been shown to decrease early growth, fat deposition, and death rates as well as the frequency with which these health issues arise. According to recent research, feed restriction can vary in intensity, timing, and length (Navidshad et al., 2006; Khajali et al., 2007). These reports have been inconsistent. Therefore, it has been suggested that feed restriction will lessen these issues. to achieve market body weight in broiler chickens that is comparable to control groups, early feed restriction programs rely on a phenomenon called compensatory growth, sometimes referred to as catch-up growth. Compensatory growth, often referred to as catch-up growth, is unusually rapid growth relative to age. Ad libitum feeding after nutritional deprivation that has inhibited growth results in an accelerated rate of growth that surpasses the normal rate of gain. It has been hypothesized that feed limiting could help to mitigate these problems. Compensatory growth, often referred to as catch-up growth, is the strategy used in early feed restriction programs to eliminate belly and carcass fat in broiler chickens and achieve market body weights comparable to control groups. Overly rapid growth relative to age is referred to as compensatory growth, or catch-up growth. Ad libitum feeding after nutritional deprivation that has inhibited growth causes an accelerated rate of growth that exceeds the normal rate of gain. The aim of This study is effect of feed restricted in various different time on production trials, protein and energy efficiency of female broiler chicks.

Materials and Methods:

Experimental Birds:

In the Animal Science Department of the College of Agricultural Engineering Sciences at the University of Sulaimani in Iraq, a poultry farm served as the study's location. For this experiment, we utilized broiler chicks (Ross 308) that were 300 days old. The chicks were divided into 20 groups of 15 birds per pen based on their average body weight and randomly assigned to one of four treatments, each treatment having five replicates. The first group was controlled (ad libitum feeding), while the second group (T1) restricted feeding for 4 hours in 24 hours (9 am–1 pm). The third group (T2) is restricted to 8 hours (9 am–5 pm), and the fourth group (T3) is restricted to 12 hours (9 am–9 pm). Only water is provided ad libitum. Every bird was given a regular, conventional diet according to the suggested schedule (NRC, 1994).

The ingredients percentage of three types of diets include: Wheat (59.23, 65.71, and 69.1), The meal of bone, and meat, (2.5, 0.55, and 0.5), Soybean meal (30, 25, and 21.44), sunflower seed oil (4, 5, and 5), Dicalcium phosphate (2.3, 1.94, and 1.66), DL-methionine (0.2, 0.07, and 0.8), Lysine HCL (0.27, 0.22, and 0.2) Limestone (1.15, 1.16 and 1.05), salt (0.25, 0.25, and 0.25) and premix (0.1, 0.1 and 0.1) for Starter, Grower and Finisher respectively. While, the calculated contents were: Energy (2919, 3056 and 3079 Kcal/kg), protein (21.32, 19.27 and 17.8), crude fiber (3.15, 3.5 and 3.7), Calcium (0.97, 0.86 and 0.85), phosphate (0.51, 0.48 and 0.47), lysine (1.45, 1.3, and 1.22), Methionine (0.52, 0.50, and 0.48) and Methionine and cystine (0.90, 0.81, and 0.73) for Starter, Grower and Finisher respectively.

Studied Characteristics:

At the end of each week, a delicate scale was used to measure the weight of each chick. Each chick's average daily body weight gain was calculated by subtracting its average end live weight from its average initial live weight throughout the same period (usu ally weekly). A particular amount of feed was given to the chicks in each duplicate once a week. Every week, the leftovers were collected, and to find out how much feed the birds ate, we divided the amount of feed that was given to them at the start of the week by the amount that was still left at the end. It is possible to calculate the feed conversion ratio (FCR) and feed intake using the method described in (FAO, 2011).

Protein and Energy intake:

At the beginning of each week, specific ration quantities were weighed and distributed to the birds for each pan. The remaining portions of the ration from each room were weighed at the end of the week and subtracted from the original portion. The consumed diet was then multiplied by the ration's proportion of protein per age stage to determine protein consumption. Each age group's amount of metabolizable energy (i.e., 1–14 days old, 15–28 days old, 29–42 days old, and the total protein and energy consumption in one to 42 days intervals old) was multiplied by the amount of food consumed to get the weekly intake of protein and energy.

Analytical techniques for data:

The trial data was analyzed using the Excel program. There were parameter calculations for the various treatments. Using XLSTAT (2004), the data were analyzed according to Duncan (1955) and at a level of 5%.

Results and Discussion:

Table 1 shows the effect of restricted feed at various time on the weight gain (g/bird/day) of broiler chickens. The third treatment gave the highest daily weight gain of 220.35 with significant differences (P≤0.05) compared to the other treatments in 1-42 days, and the fourth treatment gave the lowest weight gain compared to the control treatment, with significant differences. As for the rearing periods, the second treatment gave the highest weight gain, with significant differences with the other treatments in the period of 29–42 days.

Table1 . Utilization of restricted feed at various time on weight gain (g/bird/day) of broiler chickens (Mean ± S.E.)

	Periods (days)			
Treatment	1-14 day	15-28 day	29-42 day	1-42 day
T1	31.77a ± 0.24	78.86a ± 2.17	102.57b ± 3.68	213.20b ± 7.54
T2	31.54a ± 0.31	76.81b ± 0.95	110.41a ± 4.21	218.76a ± 5.17
T3	30.53ab ± 0.17	78.13ab ± 1.11	111.69a ± 2.17	220.35a ± 3.28
T4	29.58b ±0.43	76.21b ± 1.36	103.18b ± 3.24	208.97c ± 6.19

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different (P<0.05).

Table 2 indicates that there are significant (P≤0.05) differences between the treatments and that the fourth treatment consumed the least amount of feed, 325.64 grams, during the rearing period of 1–42 days, while the control treatment consumed the highest amount of feed, 360.00 grams per day. As for the breeding periods, the control treatment consumed the highest amount, with significant differences from the other treatments.

According to the study's findings, restricting the birds' eating times led to decreased body weight, market weight, and feed intake during the restriction period. Several studies have also revealed findings similar to these (Navidshad et al., 2006; Khajali et al., 2007; Benyi et al., 2011). The feed-restricted birds gained weight more quickly following re-alimentation, but not quickly enough to make up for the weight they lost during the restriction period. These results are consistent with those of other research conducted by Dozier et al. (2002), Saleh et al. (2004), and Urdaneta-Rincon and Leeson (2002), which discovered that although previously restricted birds demonstrated rapid development upon resumed full feeding, they were not able to fully recover from the effect of the restriction.

Table 2. Utilization of restricted feed in various time on feed intake (g/bird/day) of broiler chickens (Mean ± S.E.)

	Periods (days)			
T.	1-14 day	15-28 day	29-42 day	1-42 day
T1	41.24a ± 1.85	111.84a ± 1.98	206.92a ± 8.43	360a ± 5.47
T2	40.97a ± 0.87	108.4b ± 1.34	201.16b ± 6.25	350.53b ± 6.12
T3	41.31a ± 0.60	106.11b ± 2.13	189.87c ± 9.17	337.۲۹c ± 7.18
T4	40.68a ± 0.93	101.05c ± 1.91	183.91d ± 4.18	325.64d ± 4.13

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different (P<0.05).

Table 3 presents the utilization of feed removal over different time periods on the feed conversion ratio (g feed intake/g body weight gain) of broiler chickens. The results indicate that there are significant differences (P≤0.05) in the feed conversion efficiency between the treatments, with the third and fourth treatments exhibiting the best results with a decrease in food conversion efficiency. Additionally, the treatments outperformed the control group for the weeks in which they were most effective.

Table 3. Utilization of restricted feed in various time on feed conversion ratio (g feed intake/g body weight gain) of broiler chickens (Mean ± S.E.)

	Periods (days)			
T.	1-14 day	15-28 day	29-42 day	1-42 day
T1	1.30ab ± 0.06	1.42a ± 0.03	2.01a ± 0.02	1.69a ± 0.05
T2	1.29b ± 0.02	1.41a ± 0.05	1.83b ±0.06	1.61b ± 0.03
T3	1.35a ± 0.05	1.36b ± 0.02	1.70c ± 0.09	1.53c ± 0.06
T4	1.37a ± 0.09	1.33b ± 0.07	1.78bc ± 0.03	1.56c ± 0.04

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different (P<0.05).

Table 4 show the effect of feed removal in various periods on protein intake (g/bird/day) of broiler chickens the first and second treatments consumed the most protein compared to the other treatments, with significant (P≤0.05) differences over the end of 1-42 days. There are considerable changes between the periods 15-28 and 29-42 days. Also, the control and second treatments consumed the most, at 21.55 and 36.83, 20.89 and 35.81, respectively.

Table 4. Utilization of restricted feed in various time on protein intake (g/bird/day)

of broiler chickens (Mean ± S.E.)

	Periods (days)			
T.	1-14 day	15-28 day	29-42 day	1-42 day
T1	8.79a ± 0.03	21.55a ± 0.31	36.83a ± 0.49	67.17a ± 0.57
T2	8.73a ± 0.05	20.89a ± 0.17	35.81a ± 0.34	65.43a ± 1.13
T3	8.81a ±0.09	20.45a ± 0.26	33.80b ± 0.71	63.06b ± 0.94
T4	8.67a ± 0.04	19.48b ± 0.24	32.74b ± 0.68	60.89c ± 0.87

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different (P<0.05).

Table 5 shows how the timing of feed withdrawal impacts broiler chickens' energy intake (Kcal/bird/day). The results showed. Over 1-42 days, the first and second treatments required significantly more energy than the other treatments (P≤0.05). There are significant differences between the durations of 15 and 28 days. Furthermore, the control treatments consumed the highest (341.78), and after 29-42 days, the control and second treatments consumed 637.11 and 619.37, respectively, compared to treatment.

Table 5. Utilization of feed restricted feed in various time on energy intake (Kcal/bird/day)

of broiler chickens (Mean ± S.E.)

	Periods (days)			
T.	1-14 day	15-28 day	29-42 day	1-42 day
T1	120.37a ± 5.14	341.78a ± 21.11	637.11a ± 95.32	1099.26a ± 90.21
T2	119.59a ± 9.21	331.27b ± 33.12	619.37a ± 108.14	1070.23a ± 102.34
T3	120.58a ± 6.44	324.27c ± 9.94	584.61b ± 76.81	1029.46a ± 78.13
T4	118.74a ± 3.17	308.81d ± 37.41	566.26b ± 102.74	993.81a ± 95.37

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different (P<0.05).

In Tables 6 and 7, the forage removal treatments gave good and significant (P≤ 0.05) results for protein conversion efficiency, and energy conversion efficiency.

The results revealed that feed removal resulted in significant differences between the treatments, that the control treatment had a significantly higher protein conversion efficiency in a period of 1-42 days, that Table 8 also had significant differences in the energy conversion efficiency, and that the control treatment had the highest efficiency during the same time period.

Table 6. Utilization of restricted feed in various time on protein conversion efficiency (g protein intake/g body weight gain) of broiler chickens (Mean ± S.E.)

Periods (days)				
T.	1-14 day	15-28 day	29-42 day	1-42 day
T1	0.28ab ± 0.01	0.28a ± 0.02	0.36a ± 0.01	0.32a ± 0.02
T2	0.27b ± 0.02	0.27a ± 0.02	0.32b ± 0.03	0.30b ± 0.01
T3	0.29a ± 0.01	0.26b ± 0.01	0.30b ± 0.01	0.29b ± 0.01
T4	0.29ab ±0.01	0.25b ± 0.01	0.32b ± 0.02	0.29b ± 0.01

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different ($P<0.05$).

Table 7. Utilization of restricted feed in various time on energy conversion efficiency (Kcal/g body weight gain) of broiler chickens (Mean ± S.E.)

Periods (days)				
T.	1-14 day	15-28 day	29-42 day	1-42 day
T1	3.79b ± 0.83	4.33a ± 0.90	6.21a ± 1.17	5.15a ± 2.14
T2	3.79b ± 0.97	4.31a ± 0.67	5.61b ± 2.36	4.89b ± 2.63
T3	3.95a ± 0.39	4.15b ± 1.93	5.23c ± 5.42	4.67b ± 4.41
T4	4.01a ± 1.01	4.05b ± 1.34	5.49b ± 3.13	4.75b ± 1.94

At the below of each table write the information as follows:

T: treatment; T1: The first group was control (ad libitum feeding); T2: restrict from feeding for 4 hours in 24 hours (9 Am – 1 Pm);; T3: restricts for 8 hours (9 Am – 5 Pm); T4: restricts for 12 hours (9 Am – 9 Pm); SEM: Standard Error of Mean; abc Means with different superscripts across the Columns significantly different ($P<0.05$).

Santoso et al. (1995) stated that the food contained 16% crude protein, which is lower than the typical 21-35% observed in diets utilized by birds without limitation or unrestricted grazing. Body weight at 43 days of re-feeding, regardless of food protein content, was faster than that of birds in the control condition with no feeding challenge. This produced birds with weights comparable to birds at 56 days of age

Benyi and Habi (1998) observed A similar impact on ultimate body weight and development rate when feeding time was reduced by two days per week. A 15% feed restriction appears to be less harmful to the bird than a 30% feed reduction in terms of quantity offered. The fact that fame through advertising governs those Despite the fact that the control birds consumed more feed than the restricted birds, the effectiveness of foraging was the same regardless of the feeding system. This runs counter to previous findings that feed restriction increased feed efficiency (Navidshad et al., 2006; Mahmud et al., 2008).

These outcomes may be attributable to the diets' reducing AA or providing enough quantities of CP. The fact that the dietary CP content was shown to have no effect on feed intake and, by extension, the weight gain of chicks is supported by the fact that the ME content was found to be constant across all diets. This finding was in strong agreement with the feed intake and weight gain outcomes observed in studies conducted by Han et al. (1992), Moran et al. (1992), Moran (1994), and Abdel-Maksoud et al. (2010). In these studies, broilers had similar outcomes when the CP content of their diet was reduced from 23% to 20% and from 20% to 17% between 0 and 3 weeks of age and 3 and 6 weeks of age, respectively. Similarly, Widyaratne and Drew (2011) found that low CP diets that are easy to digest can lead to results that are on par with high CP diets when it comes to feed intake and weight increase. Reducing the CP content of the diet from 24% to 22, 20, and 18% had no influence on feed intake, according to Cheng et al. (1997a, b).

Conclusion:

Feed restricting at different times led to an increase in weight, and the third treatment gave the highest results significant differences from the other treatments. Feed consumption, a fourth treatment, gave the lowest consumption, with significant differences from the other treatments. As for the efficiency of food conversion, the third treatment gave the best results. The treatments led to improved energy and protein consumption, as well as consumption efficiency.

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THE EFFECT OF DEEP FREEZING (30°C), ON SEED GERMINATION AND FLOWERING OF HYOSCYAMUS NIGER L.

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Abstract

This research aims to investigate the impact of deep freezing at -30°C on the germination and flowering of Hyoscyamus niger L. seeds. The study examines how seeds germinate when exposed to (-30°C) for varying durations, and explores changes in flowering patterns resulting from alterations in the duration of exposure to cold temperatures. In the experimental setup, seeds were subjected to (-30°C) for different time intervals, and their germination rates were evaluated across multiple trials. The findings indicate that reducing freezing time to 30 minutes had relatively little impact, whereas increasing it to 60 minutes resulted in significantly higher germination percentages. Seeds exposed to a freezing time of 60 minutes exhibited higher average germination rates compared to those subjected to shorter freezing durations, suggesting that prolonged freezing may enhance seed dormancy breaking. Additionally, the study examined flowering and plant regeneration post-germination, revealing that seeds exposed to longer freezing durations demonstrated superior flowering competency. These observations suggest that controlled deep freezing could serve as a beneficial pre-treatment to enhance both germination rates and flowering in Hyoscyamus niger L. The implications of this research extend to the agricultural sector, as it elucidates how seed treatments influence germination rates and flowering in a specific crop. These findings contribute to our understanding of seed dormancy strategies and cold stratification interventions, potentially advancing plant propagation techniques. The optimal germination results across all tested seed accessions were achieved when seeds were subjected to different cold stratification treatments at (-30°C) for 30 minutes.

Keywords: Dormancy, Hyoscyamus niger, Gibberellic Acid, Black henbane, Cold stratification, germination.

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INTRODUCTION

The growth of these plants has attracted significant attention, primarily due to their medicinal properties and economic value. Black henbane (Hyoscyamus niger L.) is a biennial or annual plant of the Solanaceae family, renowned for its alkaloids, particularly hyoscyamine and scopolamine, which find applications in pharmaceuticals [1]. However, the process of germination and the establishment of seedlings of Hyoscyamus niger L. from seeds present considerable challenges due to low germination rates and dormancy [2].

In various habitats the control and timing of germination emerge as critical factors determining the survival of many annual plant species. Seed germination is a multifaceted process influenced by genetic factors and environmental conditions such as temperature, photoperiod, and salinity, often resulting in elevated mortality rates [3].

An issue exacerbating the unsustainable use of medicinal plants is their ability to germinate successfully in their natural environment; however, they failed to germinate well within the controlled laboratory conditions. External factors play a pivotal role in the germination of medicinal plants [4]. Genetic variation significantly impacts germination characteristics, while environmental factors also contribute significantly to species differentiation [5]. Another important factor in the survival of the next generation is the manner of regulation of seed germination and growth. However, due to the high seed dormancy level, the germination ability of the black henbane species is low, while regulatory principles of change in many types of dormancy are preserved [6]. However, environmental stress and plant growth regulators such as cold stratification gibberellic acid (GA3), ethylene, and cytokinin, also can be used to break the dormancy of seeds [7].

There is therefore always the implication that seed dormancy has a very big influence on germination of crops and subsequent production of the given plant. Various techniques of pre-soaking treatments have been discussed in the literature for enhancing INR of seeds, seed treatment with external energy which includes mechanical scarification, chemical treatments and thermal treatments [8]. All of these, thermal treatments particularly low temperatures in cold stratification have given some positive signs in at least reducing seed dormancy and promoting growth among certain plants [9]. Another technique is the use of deep freezing, where the seeds are subjected to temperatures below freezing point: Although this practice has not been studied extensively, it has the hypothesized advantages that directly improved seed germination and the resultant formation of new plants. Speculative evidence exists about seed properties and germination processes since the response is variable, depending on the degree of freezing when seeds are deep frozen. Sometimes with deep freezing seeds or embryos may become or the capability of germination may be enhanced, Physical disruption, alteration in permeability of seed or embryo coat or certain physiological modifications within seeds or embryos may be brought about [10].

The literature presents conflicting results regarding the impact of deep freezing on seed germination. Some earlier studies suggest that deep freezing can enhance the germination rate in certain temperate species by simulating winter conditions and breaking dormancy. Conversely, other authors have reported poor germination outcomes attributed to the stressful effects of freezing, leading to cellular damage and crystallization within seed components [11]. In relation to Hyoscyamus niger L, the available literature has given minimal information about the consequences of deep freezing on seed germination, and subsequent flowering. This is especially important due to the economic value of this species as well as its medicinal benefits; that's why the effects of deep freezing on germination of its seeds and resultant plant growth should be analyzed. This gap in the literature underscores the need for studies investigating the effects of deep freezing at (-30°C) on the germination and flowering of Hyoscyamus niger L. seeds [12].

In this study, seeds of Hyoscyamus niger L. were subjected to various durations of deep freezing at (-30°C), followed by germination tests. Additionally, the study evaluated the influence of deep freezing on flowering and subsequent plant regeneration. The findings concerning germination favorability are pivotal for understanding the potential application of deep freezing as a pre-sowing treatment to enhance seed germination and optimize the cultivation of Hyoscyamus niger L.

Methodologically, this study aimed to regulate the durations of freezing exposure consistently and systematically track germination rates across multiple replicates, ensuring the reliability and replicability of the data obtained. Furthermore, the research aimed to assess various physiological changes induced by deep freezing, such as alterations in seed coat permeability and enzymatic activity. These factors are crucial for elucidating the germination responses observed in the study [13].

Further, this study discusses extending usage of deep freezing with regard to agro procedures and especially when used for growing medicinal plants. The conclusions derived from the present research may be useful for furthering the breeding practices and cultivation approaches for improving the germination rates of Hyoscyamus niger L. as well as of other medicinal plants known to exhibit similar problems [14].

Hence, the present study provides a comprehensive overview of the effects of deep freezing on seed germination and flowering in Hyoscyamus niger L. The facts and conclusions discussed in the current meticulous work aim to advance the current knowledge of seed dormancy and germination treatments and provide useful recommendations that can be used in cultivation of medicinal plant species. Future studies should delve into the molecular and biochemical processes underlying the impact of deep freezing on seed physiology, thereby maximizing its potential utilization in crop management [15].

MATERIALS AND METHODS

Henbane seeds were collected from three different locations in Iraq, Iran, and Germany. After collection, the study was carved out during the period of 2016 and 2020 in Kurdistan of Iraq and Germany.

The seeds of seven accessions of henbane were investigated in this study. The seeds of Hyoscyamus niger collected from 7 distinct regions from which one variety was provided by Göttingen botanical garden. Two were commercial products supplied from Iran. Four accessions were collected from Kurdistan Mountain in Hawraman and Penjwen area (Fig 1, 2) and (Table 1).

Figure 1: Flowering response of Hyoscyamus niger (Black henbane)

Table 1: Hyoscyamus niger Accessions used in the study

#	Country	City	Original Name	Cod
1	Iraq	Tawella	KRI Hawraman	70
2	Iraq	Hasanawa	KRI Hasanawa	71
3	Iraq	Pinjwen	KRI Pinjwen	72
4	Iraq	Daray Mar	KRI Daray Mar	73
5	Iran	Takhte	Iran Takhte	74
6	Iran	Isfahan	Iran Isfahan	75
7	Germany	Göttingen	Germany 1	76

Seeds were washed with sterilized distilled water and surface sterilized with 70% ethanol for 30 seconds then followed by 6% commercial bleach (Clorox) 5% sodium hypochlorite, for 15 min then washed three times (5min.) with sterile deionized water before the germination tests [16]. For each replicate, all treatments consisted of 3 replicates with ten seeds. The seeds were placed on two layers sterilized Whatman filter paper (No.1) in disposable sterilized plastic Petri dishes (9cm) moistened with 5ml of distilled water and kept in the dark for three days then grown under 18 hours' light and 6 hours' dark

Cold stratification: Seeds after surface sterilization was dried with sterilized filter paper and maintained at three durations of (30°C) cold stratification (10, 30 and 60 minutes stored in a deep freezer). Germination was considered complete when the radicle growth up to 2 mm in length. The experiments were continued for 30 days. The germination rate (G%) was calculated using the following formulations [17].

$$Germination\ (\%) = \frac{Number\ of\ germination\ seed}{Number\ of\ Viable\ seeds\ initiated}\ \text{X}\ 100$$

Soil characterization: Three soil types; silt, sandy, and loam plus %20 Compost were selected for their textural characteristics (Table 1). The soils samples were taken after sterilization. Experimental design: Three treatments, consisted of different soil types were examined: (silt, sandy and loam plus %20 Compost).

RESULTS

This article focuses on the effects of low temperature (-30°C) on germination and flowering of Hyoscyamus niger L. a study necessary to inform efficient plant yield in green houses. In this research work, freezing-germination and regeneration responses to freezing treatment was determined with different freezing-germination and regeneration responses to freezing treatment was determined with different freezing periods to give a holistic view on cold stress injury to seeds.

Seed Germination:

The studies reveal that germination rates are approximately 30% higher in the freezing duration of 30 minutes from the basic freezing time, especially the KRI Hawraman and Iran Takhte seeds germination reached the above 90% level. This indicates that a controlled period of freezing can perhaps render seeds non-dormant, perhaps via mi-

cro-fracturing of the seed coat, which is essential for improving moisture uptake and respiration necessary for seed germination. Also, freezing at short-term enhances only more significant metabolic processes which would enhance seed viability when thawed [18].

Consequently, no improvement in germination rates was observed with freezing durations of 0 and 10 min, suggesting that seeds need more time to develop the positive physiologic effects which freezing provides. As for prolonged freezing (60 min), germination rates under these conditions stabilized at 20%, which might be attributed to cellular damage in the wake of prolonged freezing [2]. It indicates that while freezing helps promote germination, there is an optimal point at which stress affects the germination negatively.

The level of sensitivity to freezing also depends on the type of variety grown. Varieties show a characteristic reaction on exposure to freezing. For instance, one of the varieties, Germany 4, had a lower peak at 30 minutes compared to the others, which can be a sign of the genotypic or phenotypic differences in cold tolerance. This variability puts the need for an emphasis on variety-specific responses whenever one is using freezing treatments for germination enhancement (Figure 2&3).

Flowering and Regeneration:

Consequently, the flowering and regeneration study which was conducted showed that seeds of Hyoscyamus niger L. need to undergo a procedure of initial freeze followed by a dormancy period. For the first 20 days, there was not much germination and hence, it is possible that the seeds may require some days of rest while cellular repairs and other metabolic processes within the cells can occur. The average germination rate by day 30 was also slightly higher for particular varieties including Iran Takhte and KRI Hasanawa; on average germination rate by day 40 was much higher. This delayed response further suggests that the seed requires a recovery period for the seed to be viable and go through germination process (Figure 2&3).

With even the Germany 1 showing higher germination rates at day 20 than the control, it could be seen that some varieties have faster recovery mechanisms or less dormant stages. It is similar to the trends observed concerning many plant species that undergo a dormancy period following the procedure of freezing and that require rest or stratification [20].

Mechanisms and Theories:

From what was observed, the rise in germination rates when seeds were exposed to freezing for 30 minutes is because of the disruption of physical dormancy through formation of micro cracking in the off wall of the seed coat which enables increased water uptake and exchange of gases [8]. Moreover, freezing stress can stimulate metabolic processes, and thereby improve the germination indexes after theтак next round of thawing. This process may include the activation of stress-related genes that help in the management of seed germination [21].

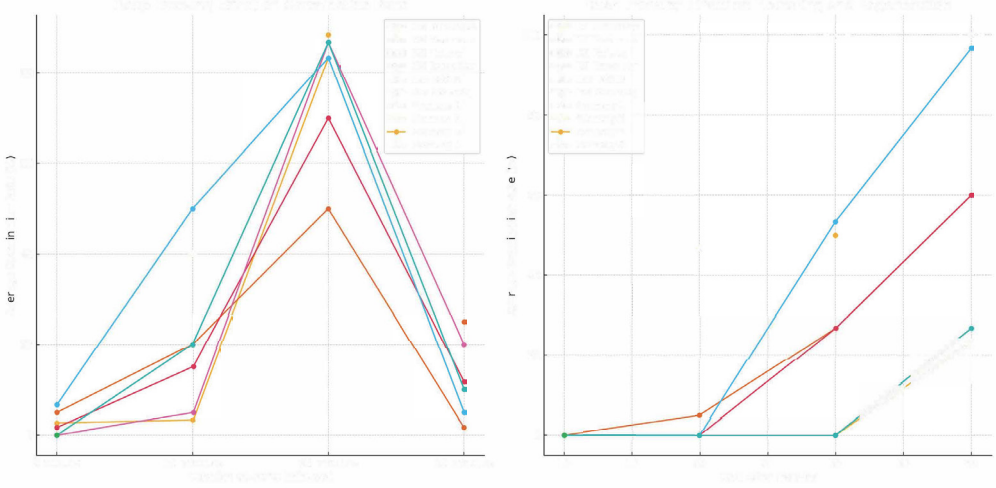

Figure 2: Effect of cold stratification at (30°C) of seed germination and flowering on Hyoscyamus niger accessions seeds.

One might assume that after freezing seeds may undergo some repair on the cellular level and an overall metabolic shift, particularly if sudden germination is a result of pick up in temperature after a period of freezing. These hormones need to be actively recycled; however, during the recovery phases, mainly gibberellins and cytokinin facilitate cell division and growth required for flowering and regeneration [22].

As per the present research, similar findings have been observed by the earlier researchers that cold stratification reduces seed dormancy and enhances the germination percentage in other plant species also [23]. The outcomes of this research can benefit guidelines on seed treatment or storage for deep freezing impacting seed germination and regeneration of Hyoscyamus niger L. seeds (Figure 2&3) [24, 25].

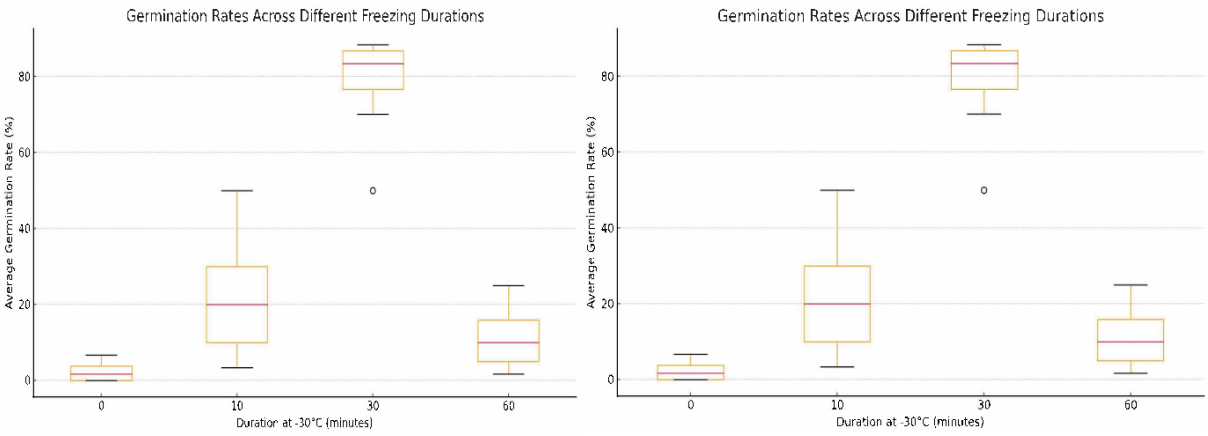

Figure 3: Germination Rates Across Different Freezing Durations, and Flowering and Regeneration Rates Across Different Days

CONCLUSION

This research conducted on the effects of deep freezing at (30°C) on the seed germination of Hyoscyamus niger L. provided vast knowledge about the enhancement of the seed's germination and the whole flowering procedure. From this study, the researcher has found facts that support the argument of a freezing time of 30 minutes since it resulted in an increased germination percentage of seeds to an average of 90% for KRI Hawraman and Iran Takhte seeds and many other different seeds. This suggests that seed dormancy can be well overcome by a freezing period that is well regulated, this may be due to the several physiological changes that favor water imbibition and metabolic rate.

However, the freezing time of zero minutes and ten minutes as well as reducing it had no impact or rather had a detrimental influence on learning and the 60-minute duration was also found to be inconsequential and time wastage. Under a long duration of frozen treatment, the germination capacity was low, as also confirmed by low germinations under subzero cell damage. In addition, on the topic of freezing, seeds were frozen and stored in the freezer then after that they were left for some time before germination on checking some of the seeds after freezing they observed very low germination rates although if they allowed the seeds to remain in freezers for 30- 40 days, more seeds germinated. This indicates that the seeds may also require time to rest and do other metabolisms like any other living organisms although it took a shorter time for water hyacinth shoots to respond.

Indeed, some of the presented results of the studies confirmed the possibility of enhancing seed germination and

plant regeneration through the method of deep controlled freezing. On a positive note, we have seen that there are ways in which freezing can be regulated/Forbidden in certain cases and thus presents new prospects for improving the practices of agriculture-enhanced germination, strong growth of plants; tangible applications for the treatment and preservation of seeds.

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THE EFFECT OF DEEP FREEZING *(-30°C)*, ON SEED GERMINATION AND FLOWERING OF *HYOSCYAMUS NIGER L*

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Abstract

This research work aims to examine the effect of deep freezing at a temperature of *(-30°C)* on the *seed germination* and flowering of *Hyoscyamus niger L.* plant. The study looks into the aspects of how seeds germinate when exposed to *(-30°C)* for different durations, as well as the flowering pattern when the period of exposure to cold temperature is altered from another. In the case of the experimental setup, the seeds were placed in (-30°C) with differing time lengths and evaluated their germination rates for numerous trials. This study shows that the decreases in freezing time to 30 minutes had relatively no impact while increasing the freezing time to 60 min allowed for significantly higher germination percentages. The average germination rate of seeds subjected to freezing time of 60 minutes tended to be above the seeds subjected to shorter exposure to freezing time hence may support the idea that extended freezing time enhances seed dormancy breaking. Furthermore, the study analyzed flowering and plant regeneration: post germination and it was established that seeds exposed to longer freezing durations had superior flowering competency. These observations conclude that controlled deep freezing can be a beneficial pre-treatment in increasing the germination rate and flowering of *Hyoscyamus niger L.* The outcome of this research could have dual effects on the agricultural field because it sheds light on how seed treatments influence germination rates and flowering of a specific crop. The findings of this study can be added to the seed dormancy strategies and cold stratification as interventions that may aid advancement of plant propagation. The best germination result for all tested seed accessions were achieved at the treatment, when the seeds were treated with different cold stratification at *(-30°C)* for 30 minutes.

Keywords: Dormancy, *Hyoscyamus niger L.*, Gibberellic Acid, Black henbane, Cold stratification, germination.

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INTRODUCTION

The growth of these plants has received a lot of attention mainly because of their curative ability and monetary worth. Black henbane *Hyoscyamus niger L.* is a biennial or annual plant of Solanaceae family, famous for its alkaloids particularly hyoscyamine and scopolamine used in pharmaceuticals [1]. The process of Germination and establishment of seedlings of *Hyoscyamus niger L.* from seeds is quite difficult due to low percentage of germination and dormancy [2].

In any of the habitats, germination control and timing are critical factors that determine the survival of many of the annual plant species. Depending on the genetic makeup of the seeds and environmental concomitants such as temperature, photoperiod, and salinity, seed germination is a challenging process in which the mortality rate is usually high [3].

One of the issues that provoke unsustainable use of medicinal plants is their ability to germinate within the local environment; however, they failed to germinate well within the controlled laboratory conditions. The external factors are very important for medicinal plants [4]. There is a genetic variation influence that affects germination characteristics. Moreover, environmental aspects are equally important in the process of derivation of species from each other [5]. Another important factor in the survival of the next generation is the manner of regulation of seed germination and growth. However, due to the high seed dormancy level, the germination ability of the black henbane species is low, while regulatory principles of change in many types of dormancy are preserved [6]. However, environmental stress and plant growth regulators such as cold stratification gibberellic acid (GA3), ethylene, and cytokinin, also can be used to break the dormancy of seeds [7].

There is always the implication that seed dormancy has a very big influence on germination of crops and the subsequent production of the given plant. Various techniques of pre-soaking treatments have been discussed in the literature for enhancing INR of seeds, seed treatment with external energy which includes mechanical scarification, chemical treatments and thermal treatments [8]. All of these, thermal treatments particularly low temperatures in cold stratification have given some positive signs in at least reducing seed dormancy and promoting growth among certain plants [9]. Another technique is the use of deep freezing, where the seeds are subjected to temperatures below freezing point: Although this practice has not been studied extensively, it has the hypothesized advantages that directly improved seed germination and the resultant formation of new plants. Speculative evidence exists about seed properties and germination processes since the response is variable, depending on the degree of freezing when seeds are deep frozen. Sometimes with deep freezing seeds or embryos may become or the capability of germination may be enhanced, Physical disruption, alteration in permeability of seed or embryo coat or certain physiological modifications within seeds or embryos may be brought about [10].

As the literature shows, the earlier works of the researchers provide conflicting results on the impact of deep freezing on the germination of seeds. For instance, the condition of deep freezing has been shown to increase the germination process rate in some of the temperate species because it is doing exactly what is intended for it, to break dormancy by mimicking the winter conditions. On the other hand, other authors have come up with poorly successful germination outcomes which were as a result of the stressing effect of freezing which leads to cell death through crystallization within the seed parts [11]. In relation to *Hyoscyamus niger L.*, the available literature has given minimal information about the consequences of deep freezing on seed germination, and subsequent flowering. This is especially important due to the economic value of this species as well as its medicinal benefits; that's why the effects of deep freezing on germination of its seeds and resultant plant growth should be analyzed. This is lacking in literature, and thus, this study seeks to establish the effects of deep freezing at (-30°C) on the germination and flowering of *Hyoscyamus niger L.* seeds [12].

For this study, seeds of *Hyoscyamus niger L.* were exposed to different durations of deep freezing at *(-30°C)* and germination tests were conducted. In addition, the study assessed the influence of deep-freezing flowering and plant regeneration that followed. The findings regarding germination favorability give the key to understanding the possibility of applying deep freezing as a pre-sowing treatment to increase seed germination and optimize the cultivation of Hyoscyamus niger L. Methodologically, this work aimed at the consistent regulation of the durations of freezing impact and the systematic tracking of the germination rates across several replicates, meaning the reliability and replicability of

the data received. Furthermore, the research also aimed at assessing the various physiological changes caused by deep freezing such as the changes in the permeability of the seed coat and changes in the enzymatic activity, both of which are imperative for explaining the germination responses captured by the study [13].

Further, this study discusses extending usage of deep freezing with regard to agro procedures and especially when used for growing medicinal plants. The conclusions derived from the present research may be useful for furthering the breeding practices and cultivation approaches for improving the germination rates of *Hyoscyamus niger L.* as well as of other medicinal plants known to exhibit similar problems [14].

Thus, the present research is focused on the synopsis of the impact of deep freezing on seed germination and flowering affecting the plant species of *Hyoscyamus niger L.* The facts and conclusions discussed in the current meticulous work aim to advance the current knowledge of seed dormancy and germination treatments and provide useful recommendations that can be used in cultivation of medicinal plant species. The molecular and biochemical processes of deep freezing on seed physiology should be investigated in subsequent studies in order to apply this treatment in the crop management to the maximum extent. [15].

1. MATERIALS AND METHODS

Henbane seeds were collected from three different locations in Iraq, Iran, and Germany. After collection, the study was The seeds of seven accessions of henbane were investigated in this study. The seeds of *Hyoscyamus niger L.* collected from 7 distinct regions from which one variety was provided by Göttingen botanical garden. Two were commercial products supplied from Iran. Four accessions were collected from Kurdistan Mountain in Hawraman and Penjwen area (Fig 1, 2) and (Table 1).

Figure 1: Flowering response of *Hyoscyamus niger L.* (Black henbane)

Table 1: *Hyoscyamus niger L.*Accessions used in the study

#	Country	City	Original Name	Cod
1	Iraq	Tawella	KRI Hawraman	70
2	Iraq	Hasanawa	KRI Hasanawa	71
3	Iraq	Pinjwen	KRI Pinjwen	72
4	Iraq	Daray Mar	KRI Daray Mar	73
5	Iran	Takhte	Iran Takhte	74
6	Iran	Isfahan	Iran Isfahan	75
7	Germany	Göttingen	Germany 1	76

Seeds were washed with sterilized distilled water and surface sterilized with 70% ethanol for 30 seconds then followed by 6% commercial bleach (Clorox) 5% sodium hypochlorite, for 15 min then washed three times (5min.) with sterile deionized water before the germination tests [16]. For each replicate, all treatments consisted of 3 replicates with ten seeds. The seeds were placed on two layers sterilized Whatman filter paper (No.1) in disposable sterilized plastic Petri dishes (9cm) moistened with 5ml of distilled water and kept in the dark for three days then grown under 18 hours' light and 6 hours' dark

Cold stratification: Seeds after surface sterilization was dried with sterilized filter paper and maintained at three durations of *(-30°C)* cold stratification (10, 30 and 60 minutes stored in a deep freezer). Germination was considered complete when the radicle growth up to 2 mm in length. The experiments were continued for 30 days. The germination rate (G%) was calculated using the following formulations [17].

$$Germination\ (\%) = \frac{Number\ of\ germination\ seed}{Number\ of\ Viable\ seeds\ initiated}\ \text{X}\ 100$$

Soil characterization: Three soil types; silt, sandy, and loam plus %20 Compost were selected for their textural characteristics (Table 1). The soils samples were taken after sterilization. Experimental design: Three treatments, consisted of different soil types were examined: (silt, sandy and loam plus %20 Compost).

RESULTS

This article focuses on the effects of low temperature *(-30°C)* on germination and flowering of *Hyoscyamus niger L.* a study necessary to inform efficient plant yield in green houses. In this research work, freezing-germination and regeneration responses to freezing treatment was determined with different freezing-germination and regeneration responses to freezing treatment was determined with different freezing periods to give a holistic view on cold stress injury to seeds.

Seed Germination:

The studies reveal that germination rates are approximately 30% higher in the freezing duration of 30 minutes from the basic freezing time, especially the KRI Hawraman and Iran Takhte seeds germination reached the above 90% level. This indicates that a controlled period of freezing can perhaps render seeds non-dormant, perhaps via micro-fracturing of the seed coat, which is essential for improving moisture uptake and respiration necessary for seed germination. Also, freezing at short-term enhances only more significant metabolic processes which would enhance seed viability when thawed [18].

Consequently, no improvement in germination rates was observed with freezing durations of 0 and 10 min, suggesting that seeds need more time to develop the positive physiologic effects which freezing provides. As for prolonged freezing (60 min), germination rates under these conditions stabilized at 20%, which might be attributed to cellular damage in the wake of prolonged freezing [2]. It indicates that while freezing helps promote germination, there is an optimal point at which stress affects the germination negatively.

The level of sensitivity to freezing also depends on the type of variety grown. Varieties show a characteristic reaction on exposure to freezing. For instance, one of the varieties, Germany 4, had a lower peak at 30 minutes compared to the others, which can be a sign of the genotypic or phenotypic differences in cold tolerance. This variability puts the need for an emphasis on variety-specific responses whenever one is using freezing treatments for germination enhancement (Figure 2&3).

Flowering and Regeneration:

Consequently, the flowering and regeneration study which was conducted showed that seeds of *Hyoscyamus niger L.* need to undergo a procedure of initial freeze followed by a dormancy period. For the first 20 days, there was not much germination and hence, it is possible that the seeds may require some days of rest while cellular repairs and other metabolic processes within the cells can occur. The average germination rate by day 30 was also slightly higher for particular varieties including Iran Takhte and KRI Hasanawa; on average germination rate by day 40 was much higher. This delayed response further suggests that the seed requires a recovery period for the seed to be viable and go through germination process (Figure 2&3).

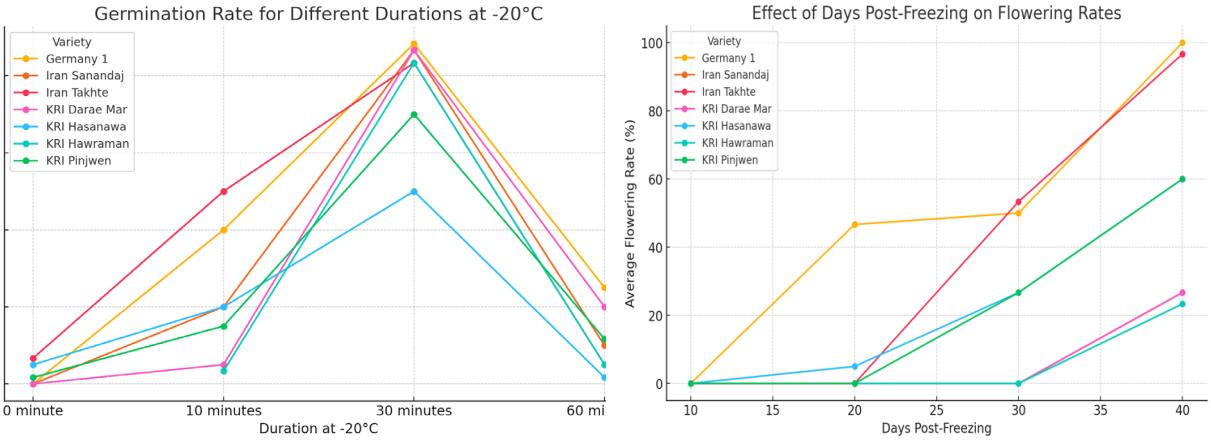

Figure 2: Effect of cold stratification at *(-30°C)* of seed germination and flowering on *Hyoscyamus niger L.* accessions seeds.

With even the Germany 1 showing higher germination rates at day 20 than the control, it could be seen that some varieties have faster recovery mechanisms or less dormant stages. It is similar to the trends observed concerning many plant species that undergo a dormancy period following the procedure of freezing and that require rest or stratification [20].

Mechanisms and Theories:

From what was observed, the rise in germination rates when seeds were exposed to freezing for 30 minutes is because of the disruption of physical dormancy through the formation of micro-cracking in the off wall of the seed coat, which enables increased water uptake and exchange of gases [8]. Moreover, freezing stress can stimulate metabolic processes, and thereby improve the germination indexes after the *next round* of thawing. This process may include the activation of stress-related genes that help in the management of seed germination [21].

It might assume that after freezing seeds may undergo some repair on the cellular level and an overall metabolic shift, particularly if sudden germination is a result of pick up in temperature after a period of freezing. These hormones need to be actively recycled; however, during the recovery phases, mainly gibberellins and cytokinin facilitate cell division and growth required for flowering and regeneration [22].

As per the present research, similar findings have been observed by the earlier researchers that cold stratification reduces seed dormancy and enhances the germination percentage in other plant species also [23]. The outcomes of this research can benefit guidelines on seed treatment or storage for deep freezing impacting seed germination and regeneration of *Hyoscyamus niger L.* seeds (Figure 2&3) [24, 25].

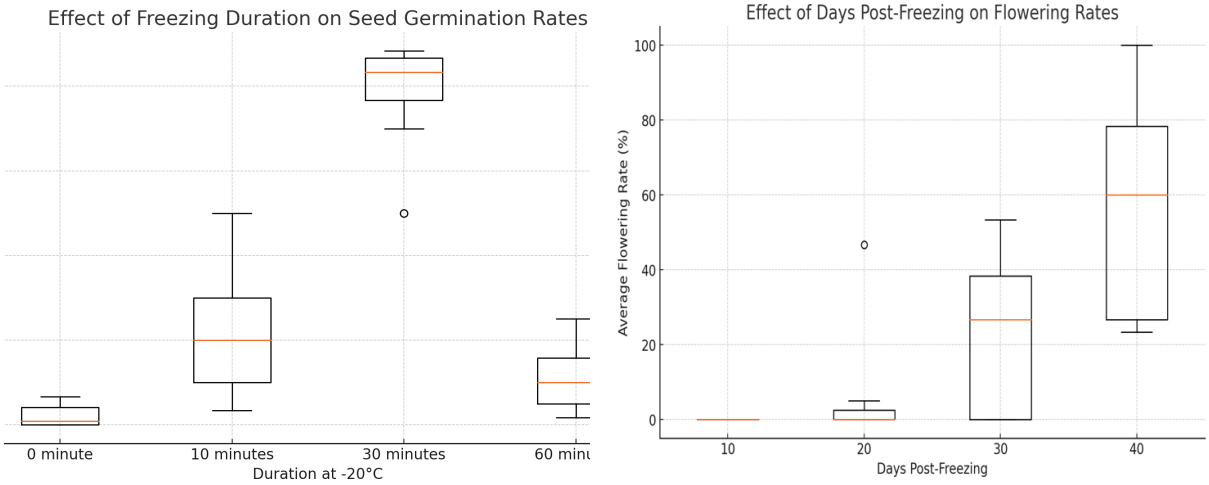

Figure 3: Germination Rates Across Different Freezing Durations, and Flowering and Regeneration Rates Across Different Days

CONCLUSION

This research conducted on the effects of deep freezing at *(-30°C)* on the seed germination of *Hyoscyamus niger L.* provided vast knowledge about the enhancement of the seed's germination and the whole flowering procedure. From this study, the researcher has found facts that support the argument of a freezing time of 30 minutes since it resulted in an increased germination percentage of seeds to an average of 90% for KRI Hawraman and Iran Takhte seeds and many other different seeds. This suggests that seed dormancy can be well overcome by a freezing period that is well regulated, this may be due to the several physiological changes that favor water imbibition and metabolic rate.

However, the freezing time of zero minutes and ten minutes as well as reducing it had no impact or rather had a detrimental influence on learning and the 60-minute duration was also found to be inconsequential and time wastage. Under a long duration of frozen treatment, the germination capacity was low, as also confirmed by low germinations under subzero cell damage. In addition, on the topic of freezing, seeds were frozen and stored in the freezer then after that they were left for some time before germination on checking some of the seeds after freezing they observed very low germination rates although if they allowed the seeds to remain in freezers for 30- 40 days, more seeds germinated. This indicates that the seeds may also require time to rest and do other metabolisms.

Indeed, some of the presented results of the studies confirmed the possibility of enhancing seed germination and plant regeneration through the method of deep controlled freezing. On a positive note, we have seen that there are ways in which freezing can be regulated/Forbidden in certain cases and thus presents new prospects for improving the practices of agriculture-enhanced germination, strong growth of plants; tangible applications for the treatment and preservation of seeds.

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Analyzing The Effective Factors Related To Electrical Energy Meter Calculation In a Transmission Field

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Abstract

This article describes both physical and non-physical factors that affect the accuracy of energy calculations within energy meter devices and Supervisory Control and Data Acquisition (SCADA) systems or any other control center that relies on energy meter devices. The aim is to obtain more reliable and accurate data and to analyze the energy loss between the energy source and destination in depth. In addition to potential measurement equipment errors, such as those stemming from transformation device errors, there are hidden factors that influence energy calculation results. These factors manifest within the electrical transmission and distribution networks, as well as in the placement of energy meter devices within substations. Furthermore, ensuring the accuracy of energy data sourced from both transmission and distribution fields, along with synchronizing calculation times for comparison with energy production data, is crucial for analyzing energy loss and identifying unknown causes.

Keywords Extra Factors Affecting Electrical Energy Calculation.

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Introduction

The study of the transmission network aims to uncover the principles behind energy meter calculations. Despite the available energy metering methods, there are unknown causes for discrepancies in recorded energy amounts between power generation stations and transmission substations. The energy amount recorded at the same line energy meter from the generator power plant and the transmission substation differs, sometimes even showing less line impedance loss than expected. Why is this the case? What is the voltage situation on the busbar when there are different voltage sources? The allowable difference in voltage for synchronizing the 132KV incoming line permission to switch on is less than 5KV; this affects the voltage on the transmission substation busbar. How do we determine the final voltage value that the energy meter device uses to calculate the energy amount accurately? Where does the power factor come into play when there are several incoming feeders on the same busbar? Does the load type affect energy metering?

These unanswered questions reveal several hidden factors that affect the energy data obtained from energy meter devices. There is a need for acceptable explanations for all these questions. This necessity drives the research, and the results suggest a review of energy meter calculation methodologies and the addition of some additional functions. This includes ensuring that the completion current circuit has an acceptable neutral point with the lowest resistance, achieved through strong earthing system connectivity to discharge current from the phase neutral point. Using C.T and V.T metering cores to connect to the energy meter is a priority to protect pure analog measurements to the energy meter devices. Line impedance can be calculated and used in a comparison equation to check the corrected energy count. In the design of the substation diagram busbar, if there are several incoming feeders, the voltage input to the energy calculator software block does not decrease as per the line impedance, but rather changes depending on the voltage value of another feeder. Synchronizing the summation time of energy counted is important to provide the total energy amount hourly.

This paper included the following subjects:

Factors Reflection

In this section, we discuss the physical and non-physical factors that affect energy calculation methods. While some of these factors have been mentioned in previous research, many have not been recognized as factors influencing energy management. Understanding the influence of these factors is essential for developing effective energy management strategies.

Research Methodology

This section provides theoretical explanations of network provocation issues and their direct influence on energy calculation equation characteristics. Additionally, it demonstrates the energy summation equation used by the control center. The theoretical methods focus on analyzing network provocation issues to accurately determine the energy calculation equation. These issues are crucial for understanding the overall energy consumption dynamics within a network system. By addressing these challenges, we can refine energy calculation equations and enhance the efficiency of the control center's energy summation process. This theoretical framework forms a solid foundation for addressing network provocation issues and optimizing energy management strategies.

Research Application

This section clarifies the energy calculation data collected directly from power generation and transmission substations over one month. It outlines the data analysis process and theoretical energy calculations performed under various scenarios. The collected field data includes information on power generation output, transmission losses, and overall energy consumption within the substation.

Results and Suggestions

In this section, we present the research results and propose potential subjects for future academic exploration. After analyzing the collected data, it is evident that further investigation is warranted into future research directions. Additionally, exploring the long-term effects contributes to a more comprehensive understanding of the topic. Overall, there are numerous opportunities for future academic exploration in this field, and I am eager to pursue these avenues in my future academic endeavors.

Recommendation

This section illustrates the influence of applying this research as a forward step in explaining energy calculation and understanding electrical networks. The control center will also be closer to understanding lost power. Detailed analog data collected during power counting inside the energy meter itself will clarify power quality, stability, and the reaction of load types in the power network.

This research offers a new perspective on this subject, aiming to review the available methods of energy calculation. It represents an effort to design a diagnostic module to identify the reasons for errors within the power network. These aspects were not addressed by existing energy meter methods. The proposed module aims to provide a more comprehensive and accurate analysis of energy usage within power networks, filling the gaps left by traditional energy meter methods. By identifying and reporting errors in the system, this module can help improve the efficiency and reliability of energy calculations, ultimately leading to more informed decision-making in energy management.

Abbreviations

KV	Kilo Volt
A	Ampere
MW	Mega Watt
MWh	Mega Watt per hour
MVar	Mega Var
MVarh	Mega Var per hour
Var	volt-ampere reactive
C.T	Current Transformer
V.T	Voltage Transformer
SCADA	Supervisory Control and Data Acquisition
EMS	Energy Management System
DMS	Distribution Management System
GPS	Global Positioning System
IEEE	Institute of Electrical and Electronics Engineers
IEC	International Electrotechnical Commission

Factors Reflection

The energy calculation software module of energy meter, designed as well as a design method of classic analog energy meter devices; is driven by using the analog input with a transformation processing from a high to a low value through current transformer C.T and voltage transformer V.T, suitably with the metering device accuracy. the principle of the energy calculation is a counted pulses generated by multiplication of Voltage, Current, and power factor values. Logically, there is no ideal value of the analog measurement could be given the pure value from source of electrical to the energy meter calculation model, consequently, the energy metering device is not able to get the pure energy value. However, the following factors are considered as a hardware error, should be taking in account for energy metering method:

Transformation errors of voltage and current, it is classified as C.T. and V. T. 1 These errors occur due to inaccuracies in the voltage and current measurements, and they are typically categorized as either C.T. errors or V.T. errors. The accuracy of these transformers is crucial for ensuring precise measurements in electrical systems. Feeding the analog measured voltage and current to the energy meter devices, should be from the metering core, as specified for the energy meter devices. Proper calibration and maintenance of C.T. and V.T. equipment is essential to minimize these errors and ensure reliable power data management.

Transformation error of the voltage and current CT and VT inside the energy meter2 itself to the small value, to be possible to use for the electronic cards of the energy meter devices, this factor is considered an accurate of the energy meter device. This factor is crucial in ensuring the precision and reliability of the energy meter readings, enabling accurate billing and monitoring of electricity consumption. By minimizing transformation errors in the voltage and current CT and VT within the energy meter, the device can provide consistent and dependable measurements, meeting the stringent requirements of modern energy management systems. As per the international standards (IEEE, IEC…), the internal transformation errors are resolved with the energy meter accuracy devices during manufacturing, and according to the application sensitivity, the designer engineering companies will choose the suitable accuracy level of the energy meter.

The obtained resistance is caused by hard wire size, termination, and loss connection (not cutting) in a wiring circuit, which connects the voltage and current circuit from the C.T and V.T to the energy meter devices, as the loss connection of the terminals; effected the current and voltage value, to arrive the real value to energy meter device. Therefore, it is crucial to ensure proper wire sizing, secure terminations, and reliable connections in the wiring circuit to maintain accurate measurements and prevent any loss in the transmission of current and voltage to the energy meter devices. Any disruptions in the connections can result in discrepancies in the readings displayed by the energy meter, impacting the overall precision and reliability of the energy consumption data recorded. Regular maintenance and monitoring of the wiring system are essential to uphold the integrity and functionality of the entire energy monitoring setup.

Earthing system connection and obtain the required resistance value from the energy meter device until the completion of the earthing circuit.

Sequence priority of analog value (current and voltage) entering, covering the pure value to the energy meter device. In this context, the sequence priority of analog values plays a crucial role in ensuring accurate and reliable measurement within the energy meter device. By establishing a clear hierarchy for the incoming current and voltage values to the energy meter devices at the first entering arrangement, the control center can effectively process and calculate these pure values into meaningful energy consumption data. This prioritization helps maintain the integrity of the measurement process, allowing the device to capture and record data in a systematic and organized manner. By giving precedence to specific analog values based on their significance and relevance, the energy meter device can generate precise energy consumption readings essential for billing, monitoring, and analysis purposes.

Time synchronization affection for energy calculation collection data between source and destination energy meter devices. This discrepancy in time synchronization can lead to inaccuracies in energy consumption calculations, po-

tentially affecting the overall reliability of the data collected by the energy meter devices. It is crucial to address and rectify these synchronization issues to ensure the precision and consistency of energy usage measurements across the system. Proper time synchronization protocols and mechanisms must be implemented to mitigate any discrepancies and maintain the integrity of the energy data collection process. Failure to address these time synchronization discrepancies can result in significant errors in energy consumption analysis and reporting. Inaccurate data may lead to financial losses, incorrect billing, and skewed energy usage patterns. By prioritizing effective time synchronization strategies, organizations can uphold the credibility and trustworthiness of their energy management systems.

The type of Load is a reason to change the power situation on the energy network as it is a reason to increase or decrease Var, affecting the Active power on the energy network. In general, different types of loads may require more or less reactive power support from the network, which can impact the overall power flow and system stability. By monitoring and managing these variations in reactive power demand, grid operators can make necessary adjustments to ensure a reliable and efficient supply of electricity to consumers. This proactive approach can help prevent potential problems such as voltage fluctuations, power outages, and equipment failures. By understanding the specific requirements of different load types, grid operators can anticipate and address any changes in power demand more effectively. Implementing smart grid technologies and advanced control systems can further enhance the stability and resilience of the energy network, ensuring reliable power delivery even under varying load conditions.

Voltage difference between source and destination, in case there will be different power supplied to the same bus bar of transmission substation. In this scenario. It is crucial to carefully monitor and regulate the voltage levels to ensure smoothly during energy calculation and to consider voltage value deferent. Failure to address voltage differences can lead to fluctuations in power supply, equipment failures, and even grid instability; therefore, there is a limitation of the voltage synchronizing, the protection system logic circuit is responsible for allowing switch-on the incoming feeder. This will help prevent any disruptions in power transmission and ensure the reliability and longevity of the equipment in the substation. It is essential to have a proactive approach to maintaining proper voltage levels to support a robust and efficient power distribution system.

Power loss cause of impedance based on the transmission line distance, type of the conductor, transformation… Impedance is a key factor determining power loss in transmission lines. The resistance and reactance of the transmission line are influenced by factors such as the length of the line, the type of conductor used, and the presence of transformers or other equipment along the line. These impedance-based losses can result in heat generation and wasted energy, making it imperative to carefully consider these factors when designing a power transmission system. Factors such as the skin effect and proximity effect can also impact impedance and contribute to power loss in transmission lines. Additionally, variations in temperature, frequency, and load conditions can further affect the impedance of the line. Proper consideration of these factors is essential to minimize power loss and ensure efficient power transmission across the grid. Additionally, implementing technologies such as power factor correction and impedance matching can help mitigate impedance-based losses and improve overall system performance. Ultimately, understanding the role of impedance in power transmission is crucial for optimizing system efficiency and reliability.

The power factor on the same busbar of the transmission substation will not same as the power factor of the generation station, in case there will be different feeding to the same busbar of the transmission substation. Practically, the power factor on the bus bar will be the average of the incoming feeder's power factors. This averaging of power factors helps to ensure a more balanced and stable power system, preventing issues such as voltage fluctuations and equipment damage. By carefully monitoring and managing power factors at various points in the transmission and generation network, operators can improve the overall efficiency and reliability of the electrical grid. This proactive approach to power factor management is essential in maintaining a smooth and reliable flow of electricity to consumers. By continuously monitoring and adjusting power factors as needed, operators can optimize the performance of the grid, reduce energy losses, and enhance system stability. Ultimately, this helps to support a more sustainable and resilient electrical infrastructure for the long term. However, the available power factor is not the same as the power of the source station, but it is changed to the average power factor; it must take in consideration.

Factors Reflection

The energy calculation software module of the energy meter, designed as well as the design method of classic analog energy meter devices, is driven by using the analog input with transformation processing from high to low values through current transformers (CT) and voltage transformers (VT), suitable for the metering device accuracy. The principle of energy calculation involves counting pulses generated by the multiplication of voltage, current, and power factor values. Logically, no ideal value of analog measurement could provide the pure value from the electrical source to the energy meter calculation model. Consequently, the energy metering device is not able to obtain the pure energy value. However, the following factors are considered as hardware errors and should be taken into account for energy metering methods:

Transformation errors of voltage and current, classified as CT and VT. These errors occur due to inaccuracies in voltage and current measurements and are typically categorized as either CT errors or VT errors. The accuracy of these transformers is crucial for ensuring precise measurements in electrical systems. Feeding the analog measured voltage and current to the energy meter devices should be from the metering core, as specified for the energy meter devices. Proper calibration and maintenance of CT and VT equipment are essential to minimize these errors and ensure reliable power data management.

Transformation errors of the voltage and current CT and VT inside the energy meter itself to small values, making it possible to use for the electronic cards of the energy meter devices. This factor is considered an accuracy factor of the energy meter device. It is crucial in ensuring the precision and reliability of the energy meter readings, enabling accurate billing and monitoring of electricity consumption. By minimizing transformation errors in the voltage and current CT and VT within the energy meter, the device can provide consistent and dependable measurements, meeting the stringent requirements of modern energy management systems. According to international standards (IEEE, IEC, etc.), internal transformation errors are resolved with energy meter accuracy devices during manufacturing. Engineering companies will choose the suitable accuracy level of the energy meter according to the application sensitivity.

The obtained resistance caused by wire size, termination, and loss connection (not cutting) in a wiring circuit connecting the voltage and current circuit from the CT and VT to the energy meter devices. Loss connection of the terminals affects the current and voltage value, resulting in the real value reaching the energy meter device. Therefore, it is crucial to ensure proper wire sizing, secure terminations, and reliable connections in the wiring circuit to maintain accurate measurements and prevent any loss in the transmission of current and voltage to the energy meter devices. Any disruptions in the connections can result in discrepancies in the readings displayed by the energy meter, impacting the overall precision and reliability of the energy consumption data recorded. Regular maintenance and monitoring of the wiring system are essential to uphold the integrity and functionality of the entire energy monitoring setup.

Earthing system connection and obtaining the required resistance value from the energy meter device until the completion of the earthing circuit.

Sequence priority of analog values (current and voltage) entering, covering the pure value to the energy meter device. In this context, the sequence priority of analog values plays a crucial role in ensuring accurate and reliable measurement within the energy meter device. By establishing a clear hierarchy for the incoming current and voltage values to the energy meter devices at the first entering arrangement, the control center can effectively process and calculate these pure values into meaningful energy consumption data. This prioritization helps maintain the integrity of the measurement process, allowing the device to capture and record data in a systematic and organized manner. By giving precedence to specific analog values based on their significance and relevance, the energy meter device can generate precise energy consumption readings essential for billing, monitoring, and analysis purposes.

Time synchronization affecting energy calculation data collection between source and destination energy meter devices. This discrepancy in time synchronization can lead to inaccuracies in energy consumption calculations, potentially affecting the overall reliability of the data collected by the energy meter devices. It is crucial to address and rectify these synchronization issues to ensure the precision and consistency of energy usage measurements across the system. Proper time synchronization protocols and mechanisms must be implemented to mitigate any discrepancies and maintain the integrity of the energy data collection process. Failure to address these time synchronization discrepancies can result in significant errors in energy consumption analysis and reporting. Inaccurate data may lead to financial losses, incorrect billing, and skewed energy usage patterns. By prioritizing effective time synchronization strategies, organizations can uphold the credibility and trustworthiness of their energy management systems.

The type of load is a reason for changing the power situation on the energy network, as it affects the reactive power (Var), which in turn affects the active power on the energy network. Different types of loads may require more or less reactive power support from the network, impacting the overall power flow and system stability. By monitoring and managing these variations in reactive power demand, grid operators can make necessary adjustments to ensure a reliable and efficient supply of electricity to consumers. This proactive approach can help prevent potential problems such as voltage fluctuations, power outages, and equipment failures. By understanding the specific requirements of different load types, grid operators can anticipate and address any changes in power demand more effectively. Implementing smart grid technologies and advanced control systems can further enhance the stability and resilience of the energy network, ensuring reliable power delivery even under varying load conditions.

Voltage difference between source and destination, in case there will be different power supplied to the same busbar of the transmission substation. It is crucial to carefully monitor and regulate the voltage levels to ensure smooth energy calculation and consider voltage value differences. Failure to address voltage differences can lead to fluctuations in power supply, equipment failures, and even grid instability. Therefore, there is a limitation of voltage synchronization, and the protection system logic circuit is responsible for allowing switch-on the incoming feeder. This will help prevent any disruptions in power transmission and ensure the reliability and longevity of the equipment in the substation. It is essential to have a proactive approach to maintaining proper voltage levels to support a robust and efficient power distribution system.

Power loss caused by impedance based on transmission line distance, type of conductor, transformation, etc. Impedance is a key factor determining power loss in transmission lines. The resistance and reactance of the transmission line are influenced by factors such as the length of the line, the type of conductor used, and the presence of transformers or other equipment along the line. These impedance-based losses can result in heat generation and wasted energy, making it imperative to carefully consider these factors when designing a power transmission system. Factors such as the skin effect and proximity effect can also impact impedance and contribute to power loss in transmission lines. Additionally, variations in temperature, frequency, and load conditions can further affect the impedance of the line. Proper consideration of these factors is essential to minimize power loss and ensure efficient power transmission across the grid. Additionally, implementing technologies such as power factor correction and impedance matching can help mitigate impedance-based losses and improve overall system performance. Ultimately, understanding the role of impedance in power transmission is crucial for optimizing system efficiency and reliability.

The power factor on the same busbar of the transmission substation will not be the same as the power factor of the generation station, in case there will be different feeding to the same busbar of the transmission substation. Practically, the power factor on the busbar will be the average of the incoming feeder's power factors. This averaging of power factors helps to ensure a more balanced and stable power system, preventing issues such as voltage fluctuations and equipment damage. By carefully monitoring and managing power factors at various points in the transmission and generation network, operators can improve the overall efficiency and reliability of the electrical grid. This proactive approach to power factor management is essential in maintaining a smooth and reliable flow of electricity to consumers. By continuously monitoring and adjusting power factors as needed, operators can optimize the performance of the grid, reduce energy losses, and enhance system stability. Ultimately, this helps to support a more sustainable and resilient electrical infrastructure for the long term. However, the available power factor is

not the same as the power of the source station, but it is changed to the average power factor; it must be taken into consideration.

Research Methodology

The type of load is a reason for changing the power situation on the energy network as it affects the active power on the energy network and may lead to an increase or decrease in Var. Different types of loads may require more or less reactive power support from the network, impacting the overall power flow and system stability. By monitoring and managing these variations in reactive power demand, grid operators can make necessary adjustments to ensure a reliable and efficient supply of electricity to consumers. This proactive approach can help prevent potential problems such as voltage fluctuations, power outages, and equipment failures. By understanding the specific requirements of different load types, grid operators can anticipate and address changes in power demand more effectively. Implementing smart grid technologies and advanced control systems can further enhance the stability and resilience of the energy network, ensuring reliable power delivery even under varying load conditions. Furthermore, by utilizing demand response programs and real-time monitoring tools, grid operators can actively manage loads to balance supply and demand more effectively. This can help reduce overall energy consumption, lower costs, and minimize environmental impacts. Additionally, promoting energy efficiency and incentivizing consumers to shift their usage to off-peak hours can help alleviate strain on the grid during periods of high demand. Overall, adapting to the changing dynamics of load requirements is essential for maintaining a reliable and sustainable energy network. Incorporating advanced forecasting techniques and predictive analytics into grid management systems can further optimize load distribution and enhance grid reliability. By leveraging data-driven insights, operators can anticipate peak demand periods, allocate resources efficiently, and deploy measures to mitigate potential grid congestion. This proactive approach not only improves system performance but also enables better integration of renewable energy sources, contributing to a more resilient and environmentally friendly energy infrastructure. By embracing innovation and continuously adapting to evolving load dynamics, the energy sector can pave the way for a more sustainable and secure future.

The energy circumstance from the production station through the transmission substation until received by the customers is affected by many situations, depending on load types, increasing and decreasing voltage, and phase angle between current and voltage, which are the basic power components, and the energy will be calculated accordingly. It can also be a reason for fluctuations in the phase angle and reactive power (Var). Since the energy calculation equation is:

$$P=V\times I\times\cos\varphi P = V \times I \times \cos \varphi P=V×I×cosφ$$

In some countries, active power is often prioritized over reactive power in network analysis. However, it is crucial to consider reactive power during network quality assessments as it plays a significant role. Reactive power arises due to consumer network conditions, serving as a vital link between power generation and consumption. Network quality enhancement remains a continuous goal, with the Control Center serving as a pivotal hub for monitoring and analyzing the power grid. Leveraging advanced technology and intricate algorithms, the Control Center ensures efficient and reliable network operations. Monitoring both active and reactive power enables the Center to detect and address potential issues proactively, averting outages and disruptions. Furthermore, the Control Center optimizes power flow to reduce losses and enhance overall performance, contributing to a stable and secure power supply while meeting the needs of both consumers and producers. As technology advances and network demands evolve, the Control Center remains at the forefront of ensuring a resilient and sustainable power infrastructure. By closely monitoring reactive power alongside active power, the Center can implement strategic measures to balance the grid, improve voltage stability, and enhance overall network efficiency. With a proactive approach to network management, the Control Center plays a vital role in maintaining a reliable power supply and meeting the growing energy requirements of modern societies.

Generally, the following situations are expected to occur in the electrical network:

The angle value between voltage and current is changeable by load type, whether it is inductive or capacitive. Then the cos φ from the equation decreases and increases accordingly. In an inductive load scenario, the angle between voltage and current tends to lag, resulting in a decrease in the power factor (cos φ). Conversely, in a capacitive load situation, the angle leads, leading to an increase in the power factor value. This dynamic relationship between load type and power factor underscores the importance of understanding the characteristics of different loads in electrical systems. In practical terms, a lower power factor in an inductive load scenario implies that more reactive power is being supplied to the system, which can result in increased energy losses and higher electricity costs. On the other hand, a capacitive load with a higher power factor tends to utilize electricity more efficiently, reducing energy wastage and improving overall system performance.

The mixed load between (Resistive, Capacitive, and Inductive) gives different network figures, changing between advantages and disadvantages. It is necessary to study very well and frequently to understand the calculation method and accordingly analyze the missed power on the networks. The Voltage value from the outgoing line of the producer plant until the customer has been clear figure to avoid any mixing reaction between the different source voltages. When working with mixed loads, it is important to carefully consider the effects of resistive, capacitive, and inductive elements on the network. Understanding the calculations and analyzing the power losses in the network is vital for ensuring efficient operation. Additionally, maintaining a consistent voltage value from the producer plant to the customer is crucial for preventing any difficulties that may arise from mixing different source voltages. It is necessary to conduct thorough studies and analyses to ensure the smooth operation of networks with mixed loads. It makes sense to have a clear diagnostic of how resistive, capacitive, and inductive elements interact within the system. By carefully managing voltage levels and power losses, we can optimize performance and prevent any potential challenges that may arise from mismatched source voltages.

The consolidation of various energy sources can complicate energy analysis, with transmission line impedance variations impacting the electrical network's energy dynamics. Energy specialists stress the importance of comprehending these intricate interrelationships among energy sources, transmission line properties, and network performance to ensure a steady and effective energy supply. By considering diverse factors affecting energy flow and consumption in the grid, professionals can make informed choices to optimize energy distribution and minimize potential disruptions. Through thorough analyses and strategic planning, industry stakeholders can collaborate to enhance the resilience and sustainability of our energy systems, benefiting society at large. Energy experts underscore those technological advancements and innovative solutions are pivotal in shaping the future of the energy industry. Integrating smart grid tech, renewable energy sources and energy storage systems can help manage supply and demand, curtail carbon emissions, and enhance system efficiency. Furthermore, promoting collaboration among industry actors, policymakers, and other stakeholders is crucial for propelling progress towards a more sustainable and resilient energy infrastructure. By embracing a comprehensive energy management approach and continuously adapting to evolving challenges, we can pave the way for a cleaner, more reliable energy future fueled by diverse and interconnected sources.

The Control Center responsible for the Energy Management System (EMS) is crucial for accurately determining energy values from production sources to distribution stations and ultimately to customers' energy meters. The Distribution Management System (DMS) complements this by providing the final energy calculations between sources and destinations. Integrating EMS and DMS enables a thorough analysis of energy flow within the system, ensuring efficient and reliable energy delivery. Data from these systems identifies improvement areas and guides future energy infrastructure expansion. This synergy enhances energy management, sustains energy supply, and enables effective responses to disruptions. Real-time data empowers operators to make informed decisions, enhancing operational efficiency and system resilience. The integration of EMS and DMS is essential for modern energy management, driving advancements towards a more reliable and intelligent energy grid.

The availability of an energy analyzer in a control center is simplified as the following equation:

Energy Data = Σ EMS data – Σ DMS data + Σ Estimated Energy Loss.

EMS coming from Energy Production Station Energy Meter Data

DMS = Σ Customer data + Σ Estimated Energy Loss.

DMS coming from Transmission Station Energy Meter Data

The method of the energy calculation software block function inside the energy meter devices did not include a detailed analysis of the mentioned reasonable factors. Furthermore, the energy management system in a control center needs those factors to obtain real data from the station's field. The absence of this crucial analysis could lead to inaccurate energy readings and mismanagement of resources, ultimately affecting the overall efficiency and reliability of the energy system. These factors must be properly accounted for in the calculation process to ensure optimal performance and informed decision-making in energy management. By incorporating a more comprehensive analysis into the energy calculation software, energy meter devices, and control centers can work together seamlessly to improve energy efficiency and sustainability. This collaboration will not only enhance the reliability of energy data but also contribute to cost savings benefits and decisions regarding the diversity and equity of power production and consumption. It is essential for energy management systems to continuously evolve and adapt to changing energy demands and technologies. By prioritizing the inclusion of crucial factors in the energy calculation process, we can pave the way for a more sustainable and efficient energy future

Research Application

This research improved in a real field theoretically and practically in the Erbil Center substation after detecting the different data of the energy meters with Khurmala Power Plant. The value of the energy data decreased and increased lower or riser than the recorded data from Khurmala Power Plants. For this reason, I studied the collected data for 1 month with several site visits and discussed with the related key persons from managers and operators within 3 months. The study consists of the following steps:

Data Resource:

The following data was collected from energy meter devices of Khurmala Power Plant and Erbil Center substation: 1- Power data recorded for single and 3 phase (Active Power MW, Reactive Power MVar, MWh, MVarh) hourly every day for 1 month. 2- Power factor recorded hourly every day for 1 month. 3- Current Voltage data recorded hourly every day for 1 month.

Data Analysis:

The energy meter device data (Voltage, Current, and Power Factor) were compared hourly with the multifunction meter data, and they matched.

The available energy meter device accounted for and recorded energy data every 3 seconds, and summed each hour; the operators recorded the energy data each hour and totaled every 24 hours. As there were no voltage, current, and power factor data recorded every 3 seconds, it was impossible to theoretically calculate the energy data every 3 seconds and compare it with measured energy data from energy meter devices; also, find the difference between measured and calculated data. The absence of real-time voltage, current, and power factor data every 3 seconds made it unfeasible to precisely compute the energy usage at that granularity. As a result, a direct comparison between the calculated theoretical energy and the actual metered data was unattainable. This limitation hindered the ability to pinpoint the exact variance between the estimated and recorded energy values on such a fine time scale. Significant challenges in accurately capturing real-time energy data at a granular level, the alternative strategies should explore to mitigate the limitations presented by the absence of instantaneous voltage, current, and power factor readings. Analyzing the gathered data retrospectively and identifying patterns in energy consumption, it became feasible to estimate the likely energy consumption trajectory at a 3-second interval, albeit with a certain margin of error in a control center software. This approach, while not providing precise real-time energy meters as energy metrics,

offered valuable insights into the general consumption patterns and fluctuations within shorter time frames.

3- There was no synchronization time system between 2 energy devices from the same feeder line in both stations on the busbar, which means the accounted energy recorded data at each hour and summation every 24 hours can be different between 2 devices for the same feeder line. This lack of synchronization could lead to discrepancies in energy readings and data, potentially confusing tracking and managing power distribution along the feeder line. Furthermore, without a reliable time system in place, discrepancies may go unnoticed, leading to inefficiencies and potential errors in monitoring and maintaining the energy grid. Addressing this synchronization issue is crucial for ensuring accurate energy accounting and efficient operation of the power system. Implementing a synchronized time system between the two energy devices on the same feeder line is essential for maintaining consistency in energy data recording and monitoring. By ensuring that both devices record and timestamp energy usage accurately, discrepancies and errors can be minimized, resulting in a more efficient and reliable power distribution system. This synchronization not only aids in accurate energy accounting but also facilitates better decision-making processes and overall system reliability. Taking proactive steps to address this issue will enhance the effectiveness and performance of the energy grid, ultimately benefiting both utility providers and consumers.

Results and Suggestions:

The following research results should be taken into consideration:

1- Current and voltage transformation errors (internal and external) of energy meter devices.

2- The resistance value caused by the available materials in a current and voltage analog input wiring circuit, to the energy meter device.

3- Earthing system design and erection loop circuit, the required earthing resistance value.

4- The analog input special metering core of CT and VT connected directly to the energy meter devices and then connected to other devices.

5- Time of summation accounted for energy data, between source and destination stations.

6- Feeding different power sources to the same busbar, caused the different voltage, phase angle, and consequently the power factor.

7- The impedance of the transmission line and primary material.

8- Network quality and load type affected the energy meter data in a destination substation.

From the result of this article, I suggest and want to follow it in my future academic work:

Developing the energy metering block software and to add: a. Transformation error correction factor, it can be through entering the C.T and V.T classification data; and the software to be able to estimate the correction for the estimated value of current and voltage value. b. The loop resistance value to be estimated in an energy calculation block function, as a reference value to the energy meter; for example, the length between C.T., and V.T. to the energy meter device, the circuit hard wire size. The software can estimate the allowable value if it is more than the device can give information. c. The neutral point of the C.T., is to be monitored as an estimated current follow depending on the phase's measured value. d. The energy meter device to be able to give the Current Voltage and Power Factor data for each accounting power circle. The software can generate those data for analysis and diagnosis. e. The line distance and related conductor data are to be entered into the device as reference data to the energy meter device, and the software to be able to estimate the impedance value, this data can be used for analysis and diagnostic.

2- The time synchronization system is important to connect through GPS to manage the daily and hourly energy summation time of energy meter devices. 3- The energy meter devices in the transmission application should have

the priority connection from the C.T. and V.T. metering core. 4- The station earthing system should be monitored and maintained with the maintenance schedule. 5- The energy metering system should be monitored as an overview of all feeders if there are different feeds to the same busbar.

Recommendation:

1- This article is helpful for the energy management system not only from the control center but also as a correction for the energy metering from the field, which is the data source for the control center. The correction of the energy meter software methods works to optimize energy data usage and efficiency at the source, ensuring that the control center receives accurate and reliable data.

2- Unless the measured data is pure, the accounting energy is not accurate. The result of this research is a step in the reform of energy management.

3- Successful energy management cannot depend on the control center's estimated data for analysis and diagnosis; instead, it needs real-time data from stations.

4- Adding analyzing and diagnostic functionality to the energy meter devices is a forward step in implementing the Network Stability System.

5- It is important to find the estimated energy loss equation depending on the transmission line impedance in the energy meter devices. Transferring this data to the control center is useful.

6- The time setting of the energy meter devices inside the substation depends on the engineering computer; without a Global Synchronization Time System, it is not easy to obtain real-time energy accounting summation.

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Design and Implementation of a Programmable Electrical Muscle Stimulator (PEMS)

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Abstract

"This research designed and constructed a Programmable Electrical Muscle Stimulator (PEMS) using an OLIMEX 328 microcontroller, an EMG shield, and controlled by a Raspberry Pi 4. The basic concept of this research is to measure the ECG and EMG of a patient while muscle stimulation is carried out. The parameters of stimulation pulses will be determined according to the ECG output. Using the Raspberry Pi 4 allows for Bluetooth remote control and programmable parameters of the PEMS device."

Keywords: EMG, OLIMEX 328, EMG shield, Raspberry Pi4, BrainBay.

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1. Introduction:

A programmable electrical muscle stimulator, in this research, is a device that uses electrical pulses to stimulate certain muscles in the body, which can be applied for treatment or as a way to improve muscle performance. With the combination of a Raspberry Pi 4 and OLIMEX 328 with an ECG and EMG shield, this device can be made even more programmable and efficient. The Raspberry Pi 4 is a powerful single-board computer that can be used for a wide range of biomedical signal processing applications. It has Wi-Fi connectivity, built-in Bluetooth, as well as several ports to communicate with various peripherals. With the right programming and software, it can be used to control a variety of devices, including an electrical muscle stimulator. Previous works of literature describe the features of EMS signals; (Azman) [1] shows that utilizing the EMS, frequency, pulse width, ramp time, duty cycle, and amplitude are among the main parameters of EMS. Frequency refers to the pulses produced per second during stimulation and is measured in Hertz. Most clinical treatments use a frequency range between 20 to 50Hz to obtain optimum results; it also varies according to the intervention, intention, or objectives at that time. Higher frequencies have proven to be more comfortable because the force response is smoothed and only gives out a tingling effect, whereas lower frequencies give out a tapping effect that distinguishes the individual pulses."

The use of modern technology in measurement and development effectively contributes to the economy with the effort and time expended by the coach and the player in achieving success (Abdulrahman) [2].

Meanwhile, (AL-IBRAHEEMI, et al) [3] show that when weightlifting players are affected by muscle weakness, it is suggested to use Electrical Stimulators to treat them for skeletal muscle impairment. EMS can provide a solution not only for the muscles but also for the affected bones.

The research by (Ahmad and Hasbullah) [4] has proven that different training programs may yield greater results when combined with EMS training.

(Achata, et al) [5] explain that atrophy refers to the decrease in the size of an organ due to the loss of protoplasmic mass. It is necessary to specify that, unlike hypoplasia, where the organ does not develop or there is arrested development. To reduce muscle fatigue, FES is triggered only when the muscle is not strong enough to move. In this situation, resulting vibrations are detected using an inertial measurement unit coupled with feature extraction and a neural classifier (Marzetti, et al) [6]."

2. Design of EMS system:

Transcutaneous Electrical Nerve Stimulation (TENS) and Neuromuscular Electrical Stimulation (NMES) are the two main types of EMS. TENS stimulates the nerves that transmit pain signals to the brain, while NMES stimulates the muscles directly.

This research worked with Neuromuscular Electrical Stimulation. The components of the smart PEMS system are the Ri4 microcontroller, OLIMEX 328 (Arduino-like board) with (EMG, ECG) shields, as shown in Fig. (1)."

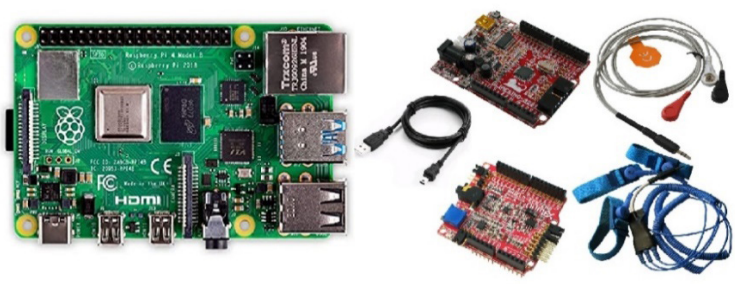

Fig. (1) PEMS devices and module.

The basic aim of the PEMS constructed in this research is to generate stimulation pulses with a desired amplitude and frequency using suitable electrodes connected to the OLIMEX 328 and EMG-ECG shields, with overall control done by the Ri4.

A C-Arduino program is written using an IDE to generate proper code for generating the desired stimulation pulses. The parameters of the stimulation pulses (amplitude, frequency, and duration) are calculated by the Ri4 single-board computer. These parameters are calculated by comparing the ECG signals with EMG signals of the person under test. Then, the parameters are sent to the OLIMEX 328 to generate the stimulation pulses, which are attached to the selected muscle by EMG electrodes.

BrainBay software [7] is used to create a Graphical User Interface (GUI) to interact with the PEMS system. The design of the GUI is shown in Fig. (2)."

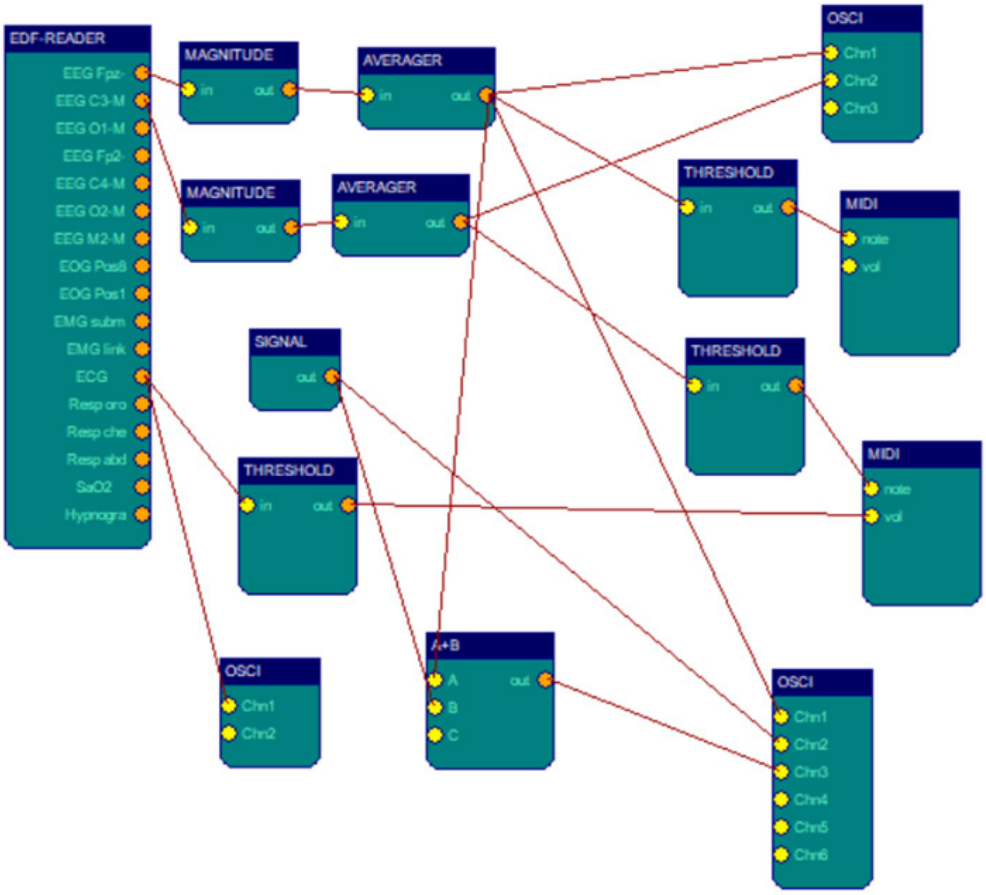

Fig. (2) BrainBay Design for Smart EMS.

3. Results and discussion:

This research designed a training course that stimulates the biceps muscles of a male under test. The frequency of these pulses varies from 10 to 30 Hz, while the amplitude ranges from 0 to 50 V, with a current not exceeding 2 mA to avoid electrical shock. During the stimulation procedure, the ECG signals of the person under test were monitored and compared with threshold values. According to the results of the comparison, the parameters of the stimulating pulses were calculated. Fig. (3) shows the first step of the stimulating procedure."

Fig. (3) PEMS signals.

The stimulation signal voltages that must be applied to the muscles using the PEMS digital therapy machine depend on the mode and intensity of the therapy being performed. The maximum voltage amplitude of the stimulation pulses depends on the mode and intensity selected for the stimulation course. The device typically delivers a series of short pulses of electrical stimulation, with each pulse lasting a few milliseconds, and the pulse frequency and width are also adjustable by the Raspberry Pi 4.

The PEMG graph can be monitored during the stimulation process, providing valuable information about the effects of the stimulation on the muscles, including the strength and timing of muscle contractions and the potential for muscle fatigue or damage. It can be used by supervisors and medical professionals to optimize therapy parameters and ensure safe and effective treatment.

It is important to take into consideration that the ECG-measured signals can be affected by EMG signals, as they may interfere with the ECG signals, especially if the patient moves during the stimulation process. Accurate and efficient software filtering processes were followed in this research to overcome this problem."

Conclusion:

This research built and tested a programmable Electrical Muscle Stimulation (PEMS) system using Raspberry Pi 4, OLIMEX 328 microcontroller, and EMG & ECG shield. According to this system, the stimulation procedure can be programmable and highly adapted to the patient's condition.

A general User Interface (GUI) was designed for this research using BrainBay software for control and monitoring purposes.

The frequency and width of the stimulation signals can be controlled by the Raspberry Pi 4 automatically according to the designed thresholds of the EMG and ECG graph of the patient or by the trainer using GUI to avoid any muscle overload, tissue damage, or any required modification or correction for the stimulation procedure.

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Section One: Writing, Submiting and Publishing Guidelines

The article is to be submitted through the Kurdistan Institute for Strategic Studies and Scientific Research (KISSR) website, and researchers must bear in mind the following instructions and guidelines regarding writing and submitting their research.The number of pages should not exceed twenty-five pages of (A4) sized paper.

1. The article must contain the following.
- Title
- Researcher name(s), Department, College, University, Country, and an official e-mail address
- Abstract in English, and two other languages, which should be placed before the bibliography (references, works cited)
- Keywords
- Article content
- Results, findings, and conclusions
- -Sources and references.

2. A cover letter must be included with each manuscript submission. It should be concise and explain why the content of the paper is significant, placing the findings in the context of existing work. It should explain why the manuscript fits the scope of the journal.

All cover letters are required to include the statements:

- We confirm that neither the manuscript nor any parts of its content are currently under consideration or published in another journal.
- All authors have approved the manuscript and agree with its submission to JKSNB .

.3 Article Writing Template

A. Mechanical Instructions

a. The title of the article should be placed at the top of the first page, be centered, and an 18-point font size with bold formatting should be used (A 16-point font size with bold formatting should be used for subheadings, a 14 -point font size for the

body, and a 10-point font size for sources, annotations, and explanations.

b. Researcher name(s), Department, College, University, Country, and an official email address should be written in size (12Indent the first line of the first paragraph by putting your cursor at the beginning of the paragraph and press the tab key once.
c. For articles written in English, font type Times New Roman should be used for the body, a 16-point font size with bold formatting for the main title, a 14-point font size for the subheadings, a 12-point font size for the body, and a 10-point font size for the sources and margins.
d. Use single line spacing between paragraphs.
e. Leave a 2 cm margin on the sides of each page.

B. Follow the Harvard Referencing System for citing and referencing sources or references,
C. The abstract should be in no more than 250 words and should be in one paragraph only. Leave a single line spacing
D. Keywords. Remember that, the keywords should be between 5-7 single words.
E. The article is presented in two different modes: Personal details are to be deleted before uploading the article to the electronic platform, and this is for the review process to run anonymouslyand for a fair review to be conducted, way from favouritism and biases.
F. When the review process is complete and the article is conditionally accepted by the reviewer(s),the editorial board contacts the researcher via their e-mail address and asks them to make the corrections suggested by the reviewer(s).Once the corrections are made, the researcher should resend the article to the journal to be checked by the reviewer(s).
G. Upon completion of the review of the article by the reviewers, the editorial board of the journal shall informs the researcher whether their research was accepted or rejected.
H. When submitting the article via electronic upload on the the journal's website, publishing costs amounting to 250,000 IQD) must be paid, and this is according to the the Ministry of Finance's letter no. on/ / 2020If this payment is not made, the article will not be forwarded to a reviewer.
I. Submission is made electronically via the journal's website or via journals@kissr.edu.iq.

4. Full experimental details must be provided so that the results can be reproduced. *Biomedicines* requires that authors publish all experimental controls and make full datasets available where possible.

Section Two: Article Review

The review process is in three stages:

The first stage: Initial (Preliminary) review:

The article is reviewed by the journal's management body in a preliminary assessment to see if the article meets the journal's requirements and that it is eligible for a revie, Here is what the process is like.

- If the language used is not proficient enough, the article will be sent to a linguist for proofreading.

The writing style is benchmarked with the one approved of by the journal to see if it is in alignment - .with the publishing instructions of the journal

-The journal staff offer formatting changes to the author if required. And, if the work is returned by the author and is still unorganized, it will rejected and will not be forwarded for a review.

- Every article or written piece will be subjected to a plagiarism detection tool or software to identify any instances of plagiarism. Authors are held responsible for the originality of their article content and the information it contains.

The Second Stage: Scientific Review

After the article passes the preliminary review stage, it is sent to two specialist reviewers in the field. If the article is rejected by one of them, the article is sent to a third reviewer, and this is because an article is only accepted with the approval of two reviewers.

The Third Stage: Approval of the Evaluation

The reviewer chooses one of three options:

1 -Rejection: a complete and final rejection of the article.

2 -Conditional Acceptance: It is accepted under the condition of making the required amendments and changes.

3- Accepting the article as it is: The journal staff might still make some minor formatting changes, but the article is not subject to a second review and further scrutiny.

i: Evaluation Guidelines for Reviewers

During evaluation, please consider the following questions

1) Does the research make a valuable contribution to current knowledge and literature, in terms of the development of a theory, generating new data, or developing new methodology?
(2 Is the research written and submitted in the light of the journal's instructions to the authors?
3) Does the research adhere to the criteria that follow?

i. Knowledge and depth of understanding of basic concepts and issues
ii. Argument and its relevance to the assignment title
iii. Analysis, including originality of examples provided
iv. Evidence and critical use of sources researched
v. Independent thought and personal evaluation of issues under discussion
vi. Accuracy and clarity of expression, grammar, and punctuation
vii. Logical organization and linking of ideas
viii. Systematic and standardized in-text citation and bibliographical references

4)The reviewer should either accept the research as submitted, demand a second evaluation after the suggested changes are made by the researcher, or, reject the research altogether.

ii.Classification Guidelines

1. An original scientific research demonstrates research results that have not previously been fully or partially published elsewhere.
2. The article under evaluation contains a comprehensive review of recent and current research in a particular area. Research in this category include questionnaires by their very nature and should contain references and critical assessments. References must be sufficient enough to allow for a good view of the subject.
3. Professional research should not be based on original research but must contribute to the application of known research results and the introduction of theoretical concepts.

iii: Final Evaluation:

Please make your final decision by ommenting A and B, and filling in Tables 1, 2 and 3.

A. **In the light of the determination of the evaluation and the final judgement of your choice, it is possible to put in place the justifications that led to that decision: (Your judgments to be written here)**

I accept the article because . . .

I reject the article because . . .

B. **In the light of the determination of the evaluation and the final judgment you have chosen, recommendations for amendments by the researcher are to be made:**

I accept the research only if the researcher makes the following changes:

1.

2.

Table (1) Procedural aspects of the research

Aspects	V.Good	Good	Fair	Accepted	Weak
Title of the article					
Research address					☐
Research references					☐
The purpose of the research Argument/ Claim					☐
Its importance and merit the study					☐
Its scientific value					☐
(Findnings align with the resarch goal(s					☐
.Hypotheses				☐	
Research approach/ Methodology			☐		
Content					☐
Conclusion					☐

Table (2) Final assessment

Classification	Evaluation Notes	Tick the box
Title of the article		
Very good	does not need to be amended	☐
Good	needs minor adjustments before publication.	☐
Acceptable	is not suitable for publication until the recommended adjustments have been made.	☐
Weak	is not suitable for publication in a scientific journal that is strict at all	☐
Others	needs a second review and further scrutiny.	☐
The Reviewer	**Signature**	**Date:**

Table (3) Personal information of the reviewer

Journal Contact

JKSNB Editorial Office

KISSR

Building No. 10, Alley 60

Gullabax 335, Shorsh Street, Opposite Shoresh Hospital

Sulaimani

Kurdistan Region, Iraq
journals@kissr.edu.iq.

Technical membaer

Mr. Tahseen Tofiq

Endnotes

- 1 It writes down on the name plate (± X%).

۲ The previous energy meter version has an internal CT and VT for decreasing analog to small value.

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