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Impact of Aqueous *Hibiscus sabdariffa* Extract on Lipid Profile in Women with Subclinical Hypothyroidism: A Study in Al-Nasiriyah City

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ABSTRACT

Subclinical hypothyroidism (SHT) is one of the common thyroid dysfunctions. This disorder is marked by elevated serum thyrotropin (TSH) levels with normal triiodothyronine (T3) and thyroxin (T4). It is often associated with dyslipidemia, particularly elevated total cholesterol (TC) and low-density lipoprotein (LDL), which increase the risk of cardiovascular disease. *Hibiscus sabdariffa* L. (sour tea) contains anthocyanins and polyphenols known for their lipid-lowering and antioxidant properties. This study was aimed to investigate the effect of aqueous extract of *H. sabdariffa* (20 g/200 ml) on metabolism of lipid in women that suffering from SHT. Fifty participants were enrolled: 10 healthy women (Group 1), and 40 women with SHT, who were subdivided into a control group (Group 2, n = 25) and a treatment group (Group 3, n = 15). The two group (group 2 & 3) took two cups of this extract daily for 6 weeks. Indices of thyroid function and lipid metabolism were measured across the study period and after it. Study results showed a significant lowering in TC, triglyceride (TG), and LDL levels and a simultaneous increase in high-density lipoprotein (HDL) and thyroid hormone parameters in the treatment group compared to the control group. The effects of extracting *H. sabdariffa* in lowering lipid level were likely due to its effects on modulating lipid metabolism and its features in enhancing antioxidant activity. For these results, daily consumption of this extract may offer a natural and supportive approach to the management of dyslipidemia in women with SHT.

Keywords: Cardioprotective properties, Dyslipidemia, *Hibiscus sabdariffa*, Lipid metabolism, Subclinical hypothyroidism (SHT)

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INTRODUCTION

Most subclinical thyroid-related dysfunctions are diagnosed through laboratory assessment of serum thyrotropin (TSH). In these conditions, free triiodothyronine (T3) and thyroxin (T4) concentrations remain physiological, despite deviations in TSH. These disorders are commonly classified into subclinical hyperthyroidism and subclinical hypothyroidism (SHT). Subclinical hyperthyroidism is defined as suppressed TSH, with normal thyroid hormone levels. SHT is defined as elevated TSH with normal T3 and T4 levels [1, 2].

Age, ethnicity, geographic distribution, and iodine consumption are among the environmental factors that influence the prevalence of subclinical thyroid disorders. Epidemiological data suggest that SHT affects approximately 4% to 10% of the population, whereas the occurrence of subclinical hyperthyroidism is reported in about 1% to 2% of individuals [3]. Thyroid hormones are crucial for metabolic processes, particularly those related to lipid homeostasis. They affect the hepatic cholesterol synthesis pathway by activating HMG-CoA reductase, the primary driver of this process. Additionally, they regulate lipoprotein lipases, the enzyme responsible for release free fatty acids and glycerol from triglycerides in chylomicrons [4, 5]. Alterations in lipid profiles-particularly elevated total cholesterol (TC) and low-density lipoprotein cholesterol (LDL-C)-are frequently observed in patients with SHT [6].

Dyslipidemia is a metabolic condition characterized by imbalances in blood lipid concentrations and is strongly linked to an increased cardiovascular risk. This condition is generally characterized by high TC, triglycerides (TG), and LDL-C, as well as low levels of high-density lipoprotein cholesterol (HDL-C). Dyslipidemia is the result of long-standing and deleterious dietary, lifestyle, and genetic factors [7, 8]. In the Iraqi context, thyroid dysfunctions-particularly hypothyroid conditions—are increasingly recognized as public health concerns, with a higher incidence among females and individuals in middle age groups [9-11]. In comparison, data from the United States reveal that thyroid disease prevalence reaches approximately 8.1% among non-Hispanic white populations [12].

Nutritional strategies have been shown to effectively reduce cardiovascular risk factors. Diets that are primarily composed of fruits, vegetables, whole grains, low-fat dairy products and unsaturated fats and limited in trans fats, saturated fats, and refined carbohydrates are strongly related to favorable cardiovascular events [13]. In recent years, the role of nutraceuticals-especially those containing high levels of antioxidant phytochemicals-has gained considerable attention for their efficacy in improving lipid profiles and managing dyslipidemia [14, 15].

Among the pharmacological supplements that have been investigated, *Hibiscus* L. (*H. sabdariffa* L.), commonly known as

sour tea, stands out as a promising candidate for regulating lipid levels. This plant belongs to the Malvaceae family and contains a wide spectrum of bioactive compounds, such as mucilaginous, pectin, anthocyanins, polyphenols, rosinic acid, and citric acid, all of which are believed to contribute to lipid reduction and cardioprotection [16–21].

Anthocyanins are among the components that have received the most attention from researchers due to their multiple physiological effects, particularly their potential role in regulating blood pressure and improving lipid metabolism [22, 23].

Based on these data, this study aimed to evaluate the effectiveness of *Hibiscus* extract in improving lipid parameters in individuals with subclinical hyperthyroidism (SHT). By focusing on dyslipidemia, which is often associated with thyroid dysfunction, the study seeks to investigate the possibility of adopting *Hibiscus* as a complementary therapeutic option.

MATERIALS AND METHODS

Study Participants

The study included fifty women, divided into two groups: ten healthy women and forty cases diagnosed with subclinical hyperthyroidism (SHT). Examinations were conducted at the Endocrinology and Oncology Center in Nasiriyah. Participants' ages ranged from 25 to 55 years, and their average body weight was approximately 98 kg.

Preparing *H. sabdariffa* Extract

To create an aqueous extract of *H. sabdariffa*, it started with 2000 grams of dried calyces, sourced from a local store in Nasiriyah. These vibrant calyces were meticulously milled to a fine powder using a lab grinder, setting the stage for the extraction process. Then, a liter of distilled water heated to a near boiling point, around 90 °C, in a clean, sanitized container. Next, 200 grams of the powdered plant material introduced carefully to the hot water, adhering to a 1:10 weight-to-volume ratio to ensure optimal extraction of those precious phytochemicals [24]. There they mingled, steeping together for a full 30 minutes, allowing the flavors and nutrients to infuse deeply into the water. After this gentle steeping period, the mixture filtered, ensuring that any remnants of plant matter were carefully removed, leaving behind only the rich extract. The final step, vibrant liquid transferred into a sterilized glass bottle, which was then tucked away in the refrigerator to maintain its freshness. To capture the full essence and benefits of this creation, the extract must use within 24 to 48 hours, a small window that promised the best results and preserved its bioactivity.

Study Design and Dose

The study duration was from February 2023 to March 2024. During this period, questionnaires were distributed to the female patients at the Oncology and Endocrinology Center, and individuals interested in participating in the study were identified. Their consent was obtained after they were informed about the study and its objectives. Continuous medical examinations were conducted throughout the 6-week study period to ensure that no complications or disruptions in their health status occurred. This study adhered to the ethical rules based on the Declaration of Helsinki and the guidelines of the World Medical Association to ensure the protection and rights of all participants, both patients and healthy volunteers.

The patient group was divided into two subgroups, resulting in 3 groups as follows:

- Group 1 (G1) = 10 healthy women
- Group 2 (G2) = 25 women with SHT who takes Levothyroxine medication
- Group 3 (G3) = 15 women with SHT who consumed *H. sabdariffa* extract drink + takes Levothyroxine medication.

Two cups of the extract were consumed daily by the participants in group 3 (1 cup in the morning, and 1 cup in the evening) for a duration of 6 weeks.

Data and Laboratory Analysis

Data collection was conducted twice: at the beginning of the study period and at the end of the six-week period. The measurements taken included weight and age. A 3 ml blood sample was drawn from all participants and placed in a centrifuge to obtain the serum, which was then used for analysing the levels of TSH, T_3, T_4, and fT_4 and the lipid profile. The study relied on the Friedewald equation to estimate the concentration of cholesterol associated with low-density lipoprotein [25].

Chemical analysis of the basic components of *H. sabdariffa* powder was conducted at the Organic Chemistry Laboratory/ College of Pharmacy/ University of Thi-Qar.

Statistical Analysis

All the results are expressed as the means ± S.D. and percentages. The data were statistically analysed via the SPSS program via T-test and ANOVA test. P-value of less than 0.05 was considered statistically significant.

RESULTS

Table 1 shows the proximate composition and functional properties of *H. sabdariffa* powder, which were analysed to determine its nutritional and bioactive components. The results revealed that the moisture content was 10.47%**,** the ash content was 11.64%**,** the crude lipid content was 1.05%**,** the crude fiber content was 1.19%**,** and the protein content was 4.17%**.** Additionally, the functional properties of the powder were evaluated, revealing a total phenolic content of 2.1%**,** antioxidant activity of 9.2%**,** total flavonoid content of 3.89%**,** and total anthocyanin content of 71.5%**.** These findings highlight the nutritional and antioxidant potential of *H. sabdariffa* powder, making it a valuable ingredient in food and pharmaceutical applications.

The mean levels of thyroid hormones in the healthy group were as follows: TSH = 2.56 ± 1.77, T4 = 93.1 ± 6.23, T3 = 1.77 ± 0.49, and fT4 = 23.3 ± 8.42. In the hypothyroid group, the hormone levels were significantly different. The mean levels were as follows: TSH = 7.22 ± 4.64, T4 = 88.29 ± 5.31, T3 = 1.18 ± 0.11, and fT4 = 5.15 ± 1.96. Elevated TSH levels and decreased T4 and fT4 levels reflect the characteristics of hypothyroidism, where the thyroid gland is underactive. Table 2 summarizes the previously mentioned findings.

Table 1 Basic components and functional properties of dried *H. sabdariffa* flower powder

Basic components				
Moisture (%)	Ash (%)	Crude lipid (%)	Crude fiber (%)	Protein (%)
10.47	11.64	1.05	1.19	4.17
Functional properties				
Total phenolic content (%)	Antioxidant activity (%)	Total flavonoids (%)	Total anthocyanin content (%)	
2.1	9.2	3.89	71.5	

Table 2 Level of thyroid hormones in healthy people and patients with hypothyroidism before the study

Groups	Parameters			
	TSH (mU/l)	T4 (nmol/l)	T3 (nmol/l)	fT4 (UI/l)
Healthy women (N=10)	2.56 * ± 1.77	93.1 * ± 6.23	1.77 * ± 0.49	23.3 * ± 8.42
Subclinical hypothyroidism women (N=40)	7.22 ± 4.64	88.29 ± 5.31	1.18 ± 0.11	5.15 ± 1.96

Data are expressed as mean ± SD, N= number. *Represents significance at P<0.05.

The data presented in Table 3 show the lipid profile parameters (TC, TG, HDL, and LDL) in healthy women and women with SHT. In the healthy group, the mean levels were as follows: TC = 134.4 ± 11.52, TG = 108.1 ± 8.23, HDL = 57.2 ± 6.069, and LDL = 73.7 ± 8.08, which are within the normal reference ranges. In contrast, women with SHT presented significantly higher levels of TC (198.9 ± 8.811), TG (210.475 ± 8.59), and LDL (125.45 ± 4.72), and lower HDL levels (35.975 ± 4.035) compared to the healthy group.

Table 3 Level of lipid profiles in healthy people and patients with hypothyroidism before the study

Groups	Parameters			
	TC (mg/dl)	TG (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
Healthy women (N=10)	134.4 * ± 11.52	108.1 * ± 8.23	57.2 * ± 6.069	73.7 * ± 8.08
Subclinical hypothyroidism women (N=40)	198.9 ± 8.811	210.475 ± 8.59	35.975 ± 4.035	125.45 ± 4.72

Data are expressed as mean ± SD, N= number, TC= Total Cholesterol; TG= Triglyceride; HDL= High Density Lipoprotein; LDL= Low Density Lipoprotein. *Represents significance at P<0.05.

Table 4 presents the mean values and standard deviations of thyroid-related hormones TSH, T4, T3, and fT4 in three groups (G1, G2, and G3). Group 2 exhibited the highest mean TSH level (6.86 ± 2.92 mU/l), significantly higher than both G1 and G3, suggesting potential subclinical or overt hypothyroidism within this group. Correspondingly, G2 showed the lowest levels of T4 (85.76 ± 4.71 nmol/L), T3 (1.17 ± 0.11 nmol/L), and fT4 (22.24 ± 1.96 UI/l), which further supports the likelihood of thyroid hypofunction. In contrast, Group 1 had the lowest TSH (3.48 ± 1.25 mU/l) and the highest levels of T4, T3, and fT4, indicating normal or potentially hyperfunctioning thyroid activity. Group 3 presented intermediate values across all parameters, with hormone levels significantly different from both G1 and G2, possibly representing a subclinical or borderline thyroid state. These findings suggest gradation in thyroid function among the groups, G3 representing an intermediate state, which indicates a potential beneficial effect of the herbal drink on thyroid function.

Table 4 Level of thyroid hormones for the three groups after the end of the study period

Groups	Parameters			
	TSH (mU/l)	T4 (nmol/l)	T3 (nmol/l)	fT4 (UI/l)
G1 (n=10)	3.48 c ± 1.25	91.1 a ± 5.86	1.96 a ± 0.44	27.3 a ± 5.52
G2 (n=25)	6.86 a ± 2.92	85.76 c ± 4.71	1.17 c ± 0.11	22.24 c ± 1.96
G3 (n=15)	5.21 b ± 1.28	88.6 b ± 3.76	1.34 b ± 0.13	24.2 b ± 2.37

Data are expressed as mean ± SD; Differences in letters a, b, c represent significant differences at P<0.05.

Table 5 shows the lipid profile parameters across the three study groups. In group 1, the mean lipid levels were within normal ranges, with TC = 127.4 ± 11.37, TG = 108.1 ± 8.23, HDL = 57.2 ± 6.07, and LDL = 68.5 ± 7.53. In contrast, group 2 presented significantly altered lipid levels, characterized by elevated TC (195.96 ± 8.77), TG (208.84 ± 9.37), and LDL (125.96 ± 5.06), along with a lower HDL level (36.4 ± 4.23), reflecting the dyslipidemia commonly associated with SHT. However, in group 3, there was a noticeable improvement in lipid parameters compared with those in group 2, with a reduction in TC (183 ± 8.94), TG (175.73 ± 12.31), and LDL (109.47 ± 5.98), along with an increase in HDL (48.27 ± 5.05).

Table 5 Level of lipid profiles for the three groups after the end of the study period

Groups	Parameters			
	TC (mg/dl)	TG (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
G1 (n=10)	127.4 a ± 11.37	108.1 a ± 8.23	57.2 a ± 6.07	68.5 a ± 7.53
G2 (n=25)	195.96 c ± 8.77	208.84 c ± 9.37	36.4 c ± 4.23	125.96 c ± 5.06
G3 (n=15)	183 b ± 8.94	175.73 b ± 12.31	48.27 b ± 5.05	109.47 b ± 5.98

Data are expressed as mean ± SD; Differences in letters a, b, c represent significant differences at P<0.05.

DISCUSSION

This study focused on the effect of consuming *H. sabdariffa* extract at a dose of 10 g/100 ml twice daily on lipid levels in women with subclinical hyperthyroidism, compared with healthy women. The results indicated that *Hibiscus* may effectively contribute to improving lipid metabolism in this group of patients. *H. sabdariffa* has been used in traditional medicine since ancient times due to its multiple pharmacological properties. Studies have shown that it has diuretic, hypotensive, antimicrobial, antioxidant, hepatoprotective, and anti-inflammatory effects. This is due to its richness in active plant compounds, such as phenolic and organic acids, flavonoids, and anthocyanins, in addition to other important compounds such as citric, malic, citric, hydroxycitric, tartaric, and ascorbic acids. [26].

The chemical composition and functional properties of *Hibiscus* powder contribute to its health benefits. Its moderate water content, low fat content, and high fiber content not only help improve storage stability but also facilitate digestion. Despite its limited protein content, the powder can be used as a supplemental protein source in the diet. On a functional level, the abundance of phenolic compounds and anthocyanins imparts significant antioxidant and heart-protective effects through the neutralization

of free radicals and reduction of oxidative stress. The results of this study are in agreement with previous literature on the composition of *H. sabdariffa* [27-29].

While research on the modulatory action of *H. sabdariffa* in thyroid hormone regulation is in its initial stages, some observations reveal a possible modulatory action. The structural resemblance between some phenolic and flavonoid compounds and thyroid hormones allows these compounds to become bound with intracellular receptors and modulate the feedback suppression of hormone release. This action could stimulate the production of thyroxin via the hypothalamic-pituitary-thyroid axis, resulting in lowered levels of TSH in individuals who consume *H. sabdariffa* [30-32].

Beyond its potential endocrine effects, *H. sabdariffa* has been shown to positively influence lipid metabolism, making it a promising candidate for managing obesity and metabolic dysregulation. Its polyphenolic content acts on different targets of metabolism, including enzymes, transcription factors, and signaling pathways, resulting in reduced inflammation, improved energy utilization, and lipid balance [33]. Different preclinical and clinical investigations support the hypolipidemic activity of *H. sabdariffa*. In animal models, administration of aqueous extract has led to significant reductions in serum triglycerides, total cholesterol, and LDL, even on high-fat diets, while HDL levels often remained stable [34-36]. Such lipid lowering is mainly attributed to antioxidant activity that prevents LDL oxidation, an early process in atherogenesis.

Mechanistically, *H. sabdariffa* supports cholesterol homeostasis by promoting HDL-mediated reverse cholesterol transport, enhancing hepatic LDL receptor activity, and inhibiting cholesterol absorption. Additional effects include inhibition of HMG-CoA reductase and fatty acid synthase, possibly through activation of AMP-activated protein kinase (AMPK) and down regulation of lipogenic transcription factors [37-39].

Anthocyanins and protocatechuic acid are among the most studied active compounds in relation to *Hibiscus*'s health effects. Additionally, hydroxycitric acid (HCA), particularly its (-)-HCA counterpart, may play a role in reducing triglyceride and cholesterol formation by inhibiting ATP-citrate lyase, a key enzyme in the lipid synthesis pathway [40]. The stamens (calyces) of the plant are highly concentrated in anthocyanins, which account for much of its cardiovascular health effects, such as regulating lipid levels and blood pressure. Along with anthocyanins, the stamens contain polyphenols and *Hibiscus* acid, which give them antioxidant properties that help protect low-density lipoprotein (LDL) from oxidation, thereby reducing the risk of atherosclerosis, a contributing factor to heart disease. [41].

CONCLUSION

In Nasiriyah, subclinical hyperthyroidism is common among women and is clearly associated with lipid disorders and an increased risk of heart disease. By monitoring daily consumption of 20g per 200ml *Hibiscus* extract, which contains high concentrations of anthocyanins and polyphenols, it was shown that this drink may help improve blood lipid profiles and reduce the risk of atherosclerosis. This natural lipid-lowering activity of *Hibiscus* suggests its potential as a supportive option for treating thyroid-related lipid problems. However, further studies are needed to further understand its mechanisms and determine its potential for clinical use.

Declarations

Ethics Approval and Consent to Participate

The study was registered in the records of the College of Pharmacy, University of Thi-Qar, under the number [IQR20230781N23], after all participants provided their informed consent to participate.

Consent for Publication

Not applicable.

Availability of Data

All data extracted or analyzed during the research period are included in this article.

Competing Interests

All authors confirm the absence of any competing interests.

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Author´s Contributions

AH was responsible for sample collection and preparation, as well as for chemical analysis of the main components of the aqueous extract of *H. sabdariffa*. Meanwhile, WS and RK interviewed study participants, distributed research information forms, obtained informed consents, and assessed their health status. WS also drew blood samples and performed the necessary biochemical analyses. WS was also responsible for study design, statistical analysis, and manuscript organization. After completing the manuscript, all researchers reviewed and approved the final version, and all authors acknowledged reading and approved it for publication.

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