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Evaluation of the Antioxidant and Antimicrobial Activities of Milk Thistle as A natural Additive and Health-enhancer

El-Shaimaa A. Rawash*, Sayed Rashad and Ghadir A. El-Chaghaby

Regional Center for Food and Feed, Agricultural Research Center, Giza, Egypt.

Abstract

MILK THISTLE (*Silybum marianum*) has been widely recognized for its bioactive properties, owing to its rich content of phytochemicals. This study presents a comprehensive analysis of milk thistle extract, focusing on its antioxidant and antimicrobial properties, as well as its phytochemical composition. Using Gas Chromatography-Mass Spectrometry, we identified a diverse array of phytochemicals in milk thistle extract. These included fatty acids (oleic acid, linoleic acid, cis-vaccenic acid), flavonoids (silybin A, silybin B and isosilybin A), and other phenolic compounds (phytol acetate and iso flavone derivative). The antioxidant activity of the milk thistle extract was substantial, with a total antioxidant activity of 21400 mg ascorbic acid equivalent/ kg. The total phenolic content was 10898.75 mg Gallic acid equivalent/kg, and the total flavonoid content was 4116 mg quercetin equivalent /kg. The extract also exhibited high free radical scavenging, ferric-reducing power, and hydrogen peroxide inhibition activities. The pigments contents of the extract were 0.039 and 1.45 mg/g for chlorophyll and carotenoids, respectively. Antimicrobial tests revealed the extract's ability to inhibit several Gram-positive and Gram-negative bacteria, such as *Escherichia coli, Klebsiella pneumoniae, and Staphylococcus aureus*, as well as fungi like *Candida albicans* and *Aspergillus* species. These findings underscore the potential of milk thistle extract as a natural antioxidant and antimicrobial agent. Given the rising concerns over chemical additives, the milk thistle extract could offer a promising alternative to food and feed additives to promote health and prevent diseases.

Keywords: Milk Thistle**,** Gas Chromatography-Mass Spectrometry, Antioxidant Properties, Antimicrobial Properties.

Introduction

Milk thistle (*Silybum marianum* (L.) Gaertn), a member of the *Asteraceae* family, has garnered significant attention for its diverse bioactive properties. Milk thistle seeds contain proteins, free fatty acids, silybonol, betaine, fixed oil, and apigenin. The bioactive components of milk thistle, primarily flavonolignans especially are responsible for its bioactivities. Milk thistle has been used traditionally in herbal medicine. Its protective effects were attributed to its bioactive ingredients, which are primarily flavonolignans like silybin, isosilybin, silychristin, and silydianin. These substances have potent antioxidant properties that aid in scavenging free radicals and averting cellular damage brought on by oxidative stress [1,2, 3].

Advanced analytical techniques such as Gas Chromatography-Mass Spectrometry (GC- MS) have enabled the detailed characterization of the bioactive compounds in milk thistle. (GC- MS) analysis provides a comprehensive chemical profile, identifying both major and minor constituents that contribute to the plant's therapeutic properties. These compounds include flavonoids, phenolic acids, fatty acids, and terpenoids, each with distinct biological activities [4].

In recent years, the potential of milk thistle as a source of natural antioxidants and antimicrobial agents has been explored with growing interest in its application as a food and feed additive. The rising consumer demand for natural and safe additives in food and feed products has driven research into plant-derived compounds with multifunctional properties [5]. Milk thistle, with its rich phytochemical profile, offers promising prospects in this regard.

The antioxidant properties of milk thistle are primarily attributed to its high content of phenolic

*Corresponding authors: El-Shaimaa A. Rawash, E-mail: shiameealy@gmail.com, Tel.: +201067460893
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compounds, which scavenge free radicals and inhibit lipid peroxidation. Antioxidants play a crucial role in food and feed preservation by preventing oxidative degradation of lipids and other nutrients, thereby extending shelf life and maintaining nutritional quality [6,7].

Moreover, the antimicrobial properties of milk thistle have been demonstrated against a range of pathogenic microorganisms. The presence of bioactive compounds with antimicrobial activity makes milk thistle an attractive natural alternative to synthetic preservatives and antibiotics in food and feed products. The incorporation of milk thistle extracts into food and feed can help in controlling microbial contamination, enhancing safety, and providing health benefits to consumers and livestock. The present study aims to investigate the bioactive components of milk thistle using GC-Ms analysis, to assess its antioxidant capacity through different in-vitro assays, to determine its content of phyto-pigments and to evaluate the antimicrobial activity of the extracts against a panel of pathogenic microorganisms.

By elucidating the chemical composition and biological activities of milk thistle, this research will provide valuable insights into its utility as a multifunctional food and feed additive. The findings will contribute to the development of natural, safe, and effective additives that meet the growing consumer demand for health-promoting and sustainable products.

Material and Methods

Plant Material Collection and Preparation

Milk thistle (*Silybum marianum*) seeds were purchased from the National Research Center and authenticated by a botanist. The seeds were cleaned, dried in the shade, and ground into a fine powder using a mechanical grinder. The powdered material was stored in airtight containers at room temperature until extraction.

The extract of milk thistle was prepared by mixing 50 g of the powdered milk thistle with 200 ml Ethanol (80%). The mixture was subjected to sonication for 2 hrs at room temperature. The extract was then filtered using Whatman No. 1 filter paper and concentrated under reduced pressure using a rotary evaporator. Finally, the concentrated extract was dried in a desiccator and stored at 4°C. [8].

Determination of Antioxidant Activity

DPPH Radical Scavenging Assay

The antioxidant activity of the extracts was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay. DPPH assay Scavenging activity on DPPH was assessed according to the method described by [9]. The reduction of the DPPH free radical was measured by reading the absorbance at 517 nm. Briefly several dilutions of milk thistle extract (50,100,250, and 500 (mg/L)) were prepared using methanol. A test tube containing each diluted sample was combined with 2 milliliters of 200 mg/L DPPH and left at room temperature for 20 minutes in the dark. Measurements of absorbance against methanol were made spectrophotometrically at 517 nm[10].

Ferric Reducing Antioxidant Power (FRAP) Assay

The reducing power of the extracts was determined using the FRAP assay. One milliliter of each sample solution (50,100,250, and 500 (mg/L)) was combined with phosphate buffer (2.5 ml, 0.2 mol/L, pH 6.6) to find the reducing power of milk thistle. Next, 2.5 ml of potassium ferric cyanide (10 g/L) were added. Following a 20-minute incubation period at 50 °C, 2.5 ml of 10% tri-chloro acetic acid was added, and the mixture was centrifuged for 10 minutes at 3000 rpm. The absorbance was measured at 700 nm against a blank (methanol) after the upper layer of solution (2.5 ml) was combined with distilled water (2.5 ml) and ferric chloride (0.5 ml, 1 g/L) [11]. Growing reducing power was indicated by the reaction mixture's increasing absorbance. Every test was run three times, and the mean value was determined.

Hydrogen peroxide scavenging activity (HPSA)

The scavenging activity of milk thistle extract against hydrogen peroxide was determined using a spectrophotometric method [12]. A 40 mM hydrogen peroxide solution was prepared in phosphate buffer (pH 7.4). The concentration of hydrogen peroxide was determined by measuring the absorbance at 230 nm using a UV-Vis spectrophotometer. In a series of test tubes, 1 ml of hydrogen peroxide solution was mixed with 1 ml of milk thistle extract at each of the following concentrations: 100, 200, 300, 400, and 500 mg/L. The mixtures were then incubated at room temperature for 10 minutes in the dark to prevent any photolytic degradation of hydrogen peroxide. After the incubation period, the absorbance of each reaction mixture was measured against a blank solution containing only phosphate buffer and hydrogen peroxide without the extract. Every test was run three times, and the mean value was determined.

Total Antioxidant Activity (TAA)

The total antioxidant activity (TAA) of milk thistle extracts was measured and expressed as ascorbic acid equivalents (AAE). The phosphomolybdenum technique [13] was used to evaluate the total antioxidant capacity of milk thistle extracts. The ascorbic acid reference antioxidant material was used to build a standard curve from which the results were obtained. The results were represented as mg ascorbic acid equivalent/kg of extract (mg AAE/kg)[13].

Determination of Total Phenolic Content (TPC)

The Folin–Ciocalteu method [14] was used to determine total phenolic content. A calibration curve for gallic acid was created, and the results were represented as mg of gallic acid equivalents per kg of extract (mgGAE/kg) based on the calibration curve's regression equation.

Determination of Total Flavonoid Content (TFC)

Utilizing quercetin as a standard reference material, the total flavonoid content of milk thistle was determined using the aluminium chloride assay [15]. The results were expressed as mg of quercetin equivalent/Kg of extract (mgQE/Kg).

Assessment of Pigments

Chlorophyll and Carotenoid Content

The chlorophyll and carotenoid content in the extracts were determined using spectrophotometric methods [16]. The absorbance of milk thistle extracts was measured at 663 nm and 645 nm using a UV-Vis spectrophotometer for two types of chlorophyll (a and b). The absorbance measurement of carotenoid content was done at 470 nm.

GC-Ms/Ms Analysis

The chemical composition of the extracts was analyzed using Gas Chromatography-Mass Spectrometry (GC-Ms/Ms).The analysis was performed using an Agilent 7890A GC system coupled with an Agilent 7000A Ms/Ms system. The GC had an HP-5MS capillary column (30 m × 0.25 mm, 0.25 µm film thickness). The oven temperature was programmed from 60°C to 300°C at a rate of 10°C/min, with a final hold time of 10 minutes. The injector temperature was set at 250°C, and helium was used as the carrier gas at a flow rate of 1 ml/minute MS conditions included an ion source temperature of 250°C and electron ionization energy of 70 eV. Mass spectra were recorded in the range of 50-550 m/z. The compounds were identified by comparing their mass spectra with those in the NIST 17 (National Institute of Standards and Technology) database, Wiley 275, Palisade 600 and ADAMS 2001)[17,18].

Antimicrobial Activity

The antimicrobial activity of the extracts was evaluated using the agar well diffusion method against a panel of pathogenic microorganisms, including Gram-positive bacteria (*Staphylococcus aureus* and *Bacilus Subtits*), Gram-negative bacteria (*Escherichia coli*, *Klebsiella pneumonia and Salmonella enterica*), and fungi (*Candida albicans, Asperagillus Nigar* and *Asperagillus Ochraceous).* The strains were obtained from the MicroAnalytical Center, Cairo University, Egypt. Wells of 6 mm diameter were punched into the agar and filled with 100 µl of the extract (50 mg/ml). Plates were incubated at 37°C for 24 hours for bacterial strains and 48 hours for fungal strains. The zones of inhibition were measured in millimeters [19].

Statistical Analysis

All experiments were performed in triplicate, and the results were expressed as mean ± standard deviation (SD). The data were analyzed using appropriate statistical software to determine the significance of the differences between the different concentrations.

Results and discussion

GC/MS/MS analysis results

The GC-Ms analysis of milk thistle extracts revealed a diverse array of bioactive compounds, including fatty acids, flavonoids, terpenes, and other phenolic compounds. These compounds contribute to the antioxidant, antimicrobial, and health-enhancing properties of milk thistle extract. Key antioxidants and antimicrobial compounds identified in the extract are listed in Table 1.

The compounds included fatty acids such as Oleic Acid and Linoleic Acid. These unsaturated fatty acids exhibit significant antioxidant properties by scavenging free radicals, thereby preventing oxidative damage to cells and tissues. Additionally, they possess antimicrobial effects against a wide range of bacterial and fungal species. In the present study, the total fatty acids in milk thistle extract accounted for approximately 19% of the area sum%. Oleic acid (omega-9) was identified as the predominant fatty acid, comprising 8.9% of the area sum%, followed by cis-vaccenic acid (omega-7) at 4.69%, isomyristic acid at 1.14%, stearic acid at 1.3%, and linoleic acid (omega-6). A previous study [6] demonstrated that the fatty acids present in milk thistle effectively inhibited the growth of Gram-positive bacteria, including *Staphylococcus aureus* and *Bacillus subtilis*, as well as Gram-negative bacteria such as *Escherichia coli* and *Streptococcus* spp. Furthermore, the oils exhibited fungicidal activity against *Candida tropicalis*.

The extract was also found to contain different terpenoids e.g. squalene which is considered as a natural antioxidant that prevents lipid peroxidation, supporting cellular health. It was detected in milk thistle extract (1.22%) area sum %. Besides its antioxidant effects, squalene is also known for its ability to enhance skin health and support the immune system. The diterpene alcohol, phytol was also detected in the extract. This compound has strong antioxidant properties, contributing to the overall antioxidant potential of the extract. It was found with area sum% (3.09%). Previously published works assessed the presence of phytol in leaves with a lower value (1.73%) [17].

The GC-Ms/Ms results also indicated the presence of Nerolidol, 7,8-Dihydro-α-ionone and Ethyl Linalool. These terpenes with documented antimicrobial properties, contributing to the extract's ability to inhibit microbial growth. These compounds were detected in the milk thistle extract with total area sum % (5.37%).

Major components of the silymarin complex (Silybin A, Silybin B, and Isosilybin A). These compounds are renowned for their potent antioxidant, antihyperglycemic, triglyceride lowering effect and hepato-protective effects [4]. These compounds were detected in the milk thistle extract with total area sum% (7.49%).

Flavonoids detected in milk thistle extract included: Vitexin, Iso-orientin, Dimethylfraxetin and Casticin that significantly enhance the antioxidant activity. These flavonoids contribute significantly to the TFC, bolstering the extract's overall antioxidant capacity. These compounds were detected in the milk thistle extract with a total area sum % (45%). Finally, (+)-Usnic Acid an alkaloid known for its potent antimicrobial effects was identified in the extract. Besides its effective antioxidant capabilities, protecting against oxidative stress. It also, displays significant antimicrobial activity, particularly against bacteria and fungi.

Antioxidant activity, phenolic and flavonoid contents

In the present work, the data for total antioxidant capacity, total phenolic and total flavonoid contents of milk thistle extracts are presented in Table (2). The results of this study indicate that milk thistle extracts possess significant antioxidant properties, as evidenced by the high values of total antioxidant activity, total phenolic content, and total flavonoid content. The TAA value of 21400 mgAAE/Kg demonstrates the strong antioxidant potential of milk thistle extracts, which is essential for combating oxidative stress and protecting cells from damage. This antioxidant capacity is likely due to the high levels of phenolic and flavonoid compounds present in the extracts.

The total phenolic content of 10898.75 mgGAE/Kg and the total flavonoid content of 4116 mgQE/Kg further support the high antioxidant activity of milk thistle extracts.

Phenolic compounds, including flavonoids, are known to be effective free radical scavengers and metal chelators, which contribute to their antioxidant activity [19]. The presence of these bioactive compounds in milk thistle suggests its potential application as a natural antioxidant in food and feed products.

In addition to their antioxidant properties, phenolic and flavonoid compounds have been reported to exhibit various biological activities, including antimicrobial, anti-inflammatory, and anticancer effects [20, 21]. The high levels of these compounds in milk thistle extracts may also contribute to their potential health benefits beyond antioxidant activity.

These findings were compatible with [22] who revealed that milk thistle are a potentially good source of natural antioxidants because they contains substantial amounts of bioactive compounds including silymarin, synidine and fatty acids. The substantial antioxidant properties of milk thistle suggests that it can be a potent source of natural antioxidants, which can be beneficial in food and feed applications to improve shelf life and enhance nutritional quality.

DPPH activity

The antioxidant activity of milk thistle extract was also evaluated using the DPPH assay, with varying concentrations of the extract (Figure 1). The results indicate a concentration-dependent scavenging effect on DPPH radicals, as evidenced by the decreasing percentage of DPPH remaining (%DPPH) with increasing concentrations of the extract.

At the highest concentration tested (500 ppm), the milk thistle extract demonstrated the highest scavenging activity, with a %DPPH of 93.76%. As the concentration of the extract decreased, the %DPPH also decreased accordingly: 78.99% at 250 ppm, 63.51% at 100 ppm, and 57.88% at 50 ppm.

These results suggest that milk thistle extract possesses significant antioxidant potential, effectively neutralizing DPPH radicals in a concentration-dependent manner. This is consistent with previous studies [23] that attribute the antioxidant properties of milk thistle to its active components such as flavonolignans (e.g., silymarin) and flavonoids, which are known for their ability to donate hydrogen atoms or electrons, thereby reducing free radicals and preventing oxidative damage.

The observed antioxidant activity of milk thistle extract makes it a promising candidate for further exploration in functional foods, nutraceuticals, and pharmaceutical formulations aimed at combating oxidative stress-related disorders.

Ferric reducing power activity

The ferric-reducing antioxidant power (FRAP) assay was employed and presented in Figure 2 to evaluate the ferric-reducing activity of milk thistle extract at various concentrations. The results indicate a concentration-dependent increase in FRAP values, reflecting the extract's ability to reduce ferric ions to ferrous ions.

At the lowest concentration tested (50 ppm), the milk thistle extract exhibited a FRAP value of 0.9025. As the concentration of the extract increased, so did its ferric-reducing activity: 1.9852 at 100 ppm, 2.5939 at 250 ppm, and 4.0023 at 500 ppm.

Comparing these results, it is evident that higher concentrations of milk thistle extract correspond to greater ferric-reducing potential. This relationship underscores the extract's capacity to donate electrons and reduce ferric ions, indicative of its antioxidant capability. The observed trend aligns with previous research attributing such antioxidant activity to milk thistle's constituents, including flavonolignans and flavonoids, which possess redox properties capable of reducing metal ions and scavenging free radicals [24]. Future investigations could delve deeper into elucidating the specific mechanisms by which milk thistle components interact with metal ions, as well as exploring synergistic effects with other natural antioxidants. These insights could further optimize its therapeutic and preventive potentials in various health contexts.

Hydrogen peroxide scavenging activity

The hydrogen peroxide scavenging activity (HPSA) of milk thistle extract was assessed at different concentrations, ranging from 100 ppm to 500 ppm (Figure 3). The scavenging activity increased with the concentration of the extract, demonstrating a concentration-dependent effect. Specifically, at 100 ppm, the scavenging activity was 11.75%. As the concentration increased to 200 ppm, 300 ppm, 400 ppm, and 500 ppm, the scavenging activities rose to 16.58%, 21.76%, 38.10%, and 61.29%, respectively. This trend indicates that higher concentrations of milk thistle extract result in a greater ability to neutralize hydrogen peroxide, and its potential to mitigate oxidative stress by reducing reactive oxygen species (ROS) levels. A common ROS can cause significant cellular damage if not adequately neutralized. Therefore, the results demonstrate that milk thistle extract may play a protective role against oxidative damage in biological systems. The significant scavenging activity at 500 ppm, which reached 61.29%, underscores the potent antioxidant capacity of the extract, making it a promising candidate for applications aimed at reducing oxidative stress.

These findings are consistent with the high total antioxidant activity observed in previous assays [25], which highlighted the extract's rich composition of phenolic and flavonoid compounds known for their antioxidant properties.

Pigment Content

The content of pigments (carotenoids, and chlorophylls) in milk thistle seeds is reported in Table 3. The pigment analysis of milk thistle extracts revealed the presence of both carotenoids and chlorophyll, with concentrations of 1.45 ± 0.01 mg/g for carotenoids and 0.039 ± 0.0005 mg/g for chlorophyll. These findings indicate that milk thistle extracts contain significant amounts of pigments, which are known to contribute to the plant's health benefits.

Although the concentration of chlorophyll in milk thistle extract is lower compared to carotenoids, its presence still contributes to the overall health-promoting properties of the extract. Additionally, the presence of these pigments could contribute to the overall antimicrobial activity observed in milk thistle extracts, as carotenoids and chlorophyll have been reported to exhibit antimicrobial properties against various pathogens [26]. Indeed other researchers [15] reported the presence of carotenoids (0.88mg/L) and chlorophylls (0.28mg/L) in milk thistle seeds.

Antimicrobial Properties of Milk Thistle Extracts

The antimicrobial properties of milk thistle extracts were listed in Table 3 and evaluated against various microorganisms, including Gram-negative bacteria, Gram-positive bacteria, and fungi.

The antimicrobial activity was assessed by measuring the growth inhibition in mm compared with standard antibiotics (100mg/ml): Gentamicin for Gram-negative bacteria, Ampicillin for Gram-positive bacteria, and Nystatin for fungi. The results demonstrated that milk thistle extracts exhibit antimicrobial activity against various microorganisms, although the inhibition zones were generally smaller than those produced by standard antibiotics.

For Gram-negative bacteria, milk thistle extract showed moderate activity against Escherichia coli (12 ± 1.0 mm) but less activity against *Klebsiella pneumoniae* and *Salmonella enterica* (9 ± 1.0 mm and 9 ± 0.5 mm, respectively). While the extract's inhibition zones were significantly smaller than those of Gentamicin, the observed antibacterial effects suggest potential utility as a supplementary or alternative treatment, especially considering the rising concern over antibiotic resistance.

In the case of Gram-positive bacteria, the milk thistle extract displayed notable activity against both *Staphylococcus aureus* (10.3 ± 0.6 mm) and *Bacillus subtilis* (11.2 ± 0.5 mm). Though the inhibition zones were smaller compared to those of Ampicillin, the results indicate that milk thistle extracts could serve as a valuable natural antimicrobial agent, particularly in settings where synthetic antibiotic use is restricted or undesirable [23]. This is consistent with previous studies [27,] which assessed the antibacterial activity of milk thistle.

Regarding fungi, the milk thistle extract exhibited significant activity against *Candida albicans* (14.3 ± 0.6 mm), which was the highest among the tested microorganisms. This suggests strong antifungal properties that could be exploited in the development of natural antifungal treatments. However, the activity against *Aspergillus niger* and *Aspergillus ochraceus* was relatively lower (9.0 mm), indicating a more selective antifungal effect. This result was compatible with the work done by *Dogan et al*.[6] who assessed

that milk thistle extracts are more effective against *Candida albicans* than the other two types

Overall, the antimicrobial efficacy of milk thistle extracts against various microorganisms highlights its potential as a natural antimicrobial agent. The presence of bioactive compounds such as silymarin, flavonoids, and phenolic acids in milk thistle likely contributes to its antimicrobial properties. These findings support the use of milk thistle extracts in food and feed additives to enhance safety and extend shelf life by inhibiting the growth of pathogenic microorganisms. Additionally, evaluating the synergistic effects of milk thistle extracts with conventional antibiotics could provide valuable insights into developing more effective antimicrobial therapies.

Conclusion

In conclusion, the study proved that milk thistle extracts, which are abundant in bioactive compounds such as phenolics, flavonoids, and fatty acids, possess potent antioxidant and antibacterial qualities. These extracts neutralize free radicals, decrease metal ions, and stop oxidative damage. Moreover, they exhibit strong to moderate antimicrobial activity against a range of microorganisms, including fungi and bacteria, both Gram-positive and Gram-negative. Bioactive components such as carotenoids, chlorophylls, and pigments support the extract's health benefits. These findings suggest that milk thistle extracts hold considerable promise for application in food, feed and pharmaceutical industries, where they can enhance product safety, extend shelf life, and provide health benefits. Future studies should explore the specific mechanisms of action and potential synergistic effects with other natural compounds, further optimizing milk thistle's therapeutic potential.

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No funding to declare.

Conflicts of Interest

The authors declare that there are no conflicts of interest.

Ethical of approval

This is not applicable to the present study.

TABLE 1. Active compounds in milk thistle extract detected by GCMs/Ms.

Retention time (min)	Compound name	Area sum %
11.78	Arachic acid	3.78
12.56	Phytol	3.09
12.68	Linoleic acid	2
12.8	cis-Vaccenic acid	4.69
12.84	Isophytol, acetate	0.6
12.9	cis-Z-α-Bisabolene epoxide	1.38
13.14	Nerolidol	2.09
13.3	Isosilybin A	2.52
13.6	4',6-Dimethoxyisoflavone-7-O-β-D-glucopyranoside	0.47
14.26	Linoleic acid	0.47
14.4	Casticin	40.11
14.43	Ethyl linalool	2
14.77	Stearic acid	1.3
14.9	Silybin B	1.63
15.64	Vitexin	2.05
16.03	Oleic Acid	8.91
16.17	Isoorientin	3.8
16.22	(+)-Usnic acid	0.9
16.44	2,3-Dehydrosilybin B	2.66
16.9	Dimethylfraxetin	0.41
17.4	Isomyristic acid	1.14
17.66	Levulinic acid	0.9
18.2	Isohumulone	0.8
20.39	Reserpine	1.1
21.2	Geranyl linallol	1.16
21.43	Squalene	1.22
21.65	4-(Hydroxymethyl)-2-methoxyphenol	1.6
21.89	Silydianin	3.22
21.93	7,8-Dihydro-α-ionone	4

TABLE 2. Total Antioxidant capacity, Total Flavonoid Content, and Total Phenolic Content of Milk Thistle extracts.

	Milk thistle extract
Total antioxidant activity (mg AAE/ Kg)	21400.00 ±284.91
Total content flavonoid (mg QE/Kg)	4116.00 ±63.80
Total phenolic content (mg GAE/Kg)	10898.75 ±105.29

*Results are reported as means ± SD of three replicates.

TABLE 3. Pigments in Milk Thistle extracts.

Carotenoids	Chlorophyll
1.45 ± 0.01	0.039 ± 0.0005

*Results are represented in (mg/g) measuring unit and reported as means ± SD of three replicates.

TABLE 4. Antimicrobial Activity of milk thistle extracts.

Microorganism	Milk thistle extract	Standard
Gram-negative bacteria		**Gentamicin**
Escherichia coli (ATCC:10536)	12±1.0	26.7±0.6
Klebsiella pneumonia (ATCC:10031)	9±1.0	25.0 ±1.0
Salmonella enterica (ATCC: 14028)	9±0.5	18.7±0.6
Gram-positive bacteria		**Ampicillin**
Staphylococcus aureus (ATCC:13565)	10.3±0.6	21.3±0.6
Bacilus Subtits (DSM:1088)	11.2±0.5	21.6±0.6
Fungi		Nystatin
Candida albicans(ATCC:10231)	14.3±0.6	21.0±1.0
Asperagillus Nigar(ATCC:16404)	9±0.5	19.3±0.6
Asperagillus Ochraceous (ATCC:22947)	9±0.5	22.0±1.0

***Zone of inhibition is expressed as mean ± Standard deviation (mm). 100µl of the sample was tested.**

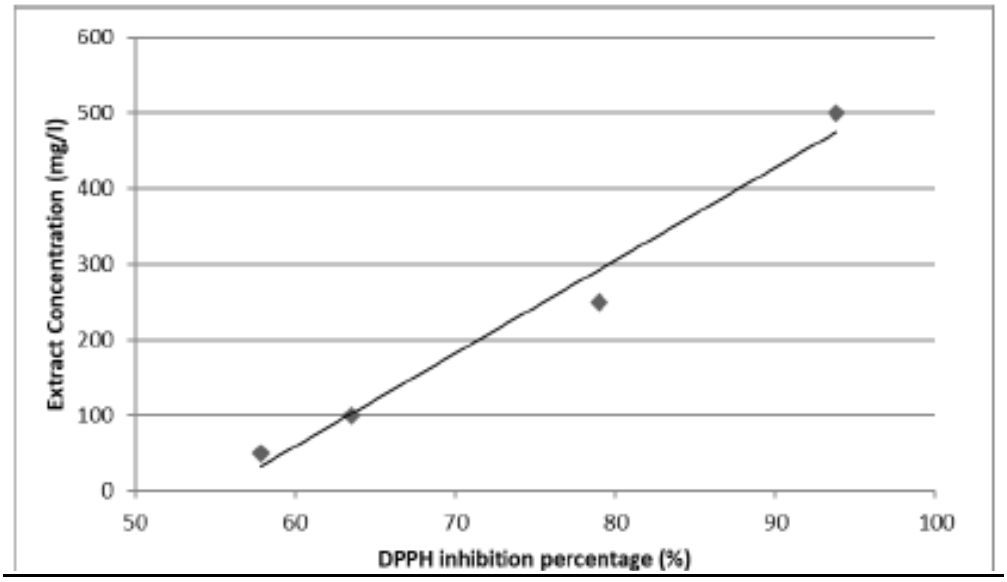

Fig.1. DPPH scavenging Activity of milk thistle extract

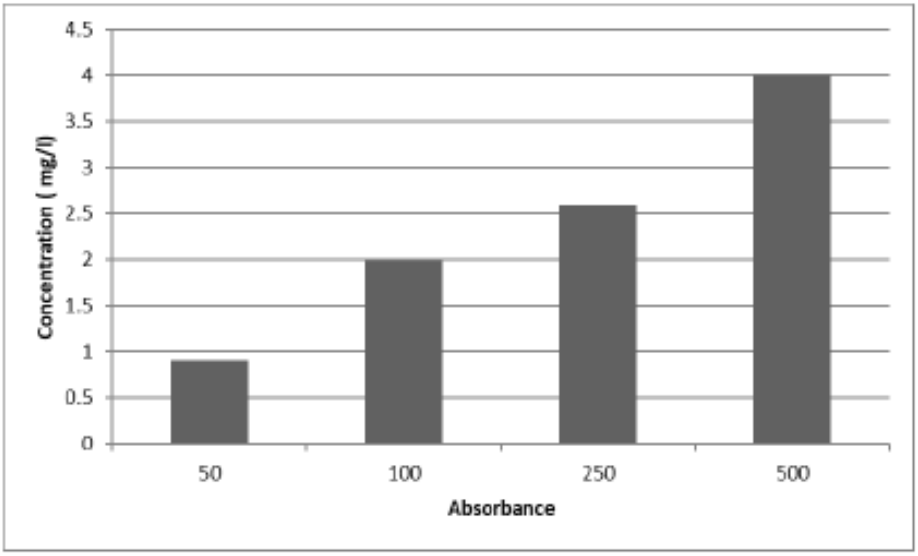

Fig.2. Ferric Reducing Power Activity of milk thistle extract

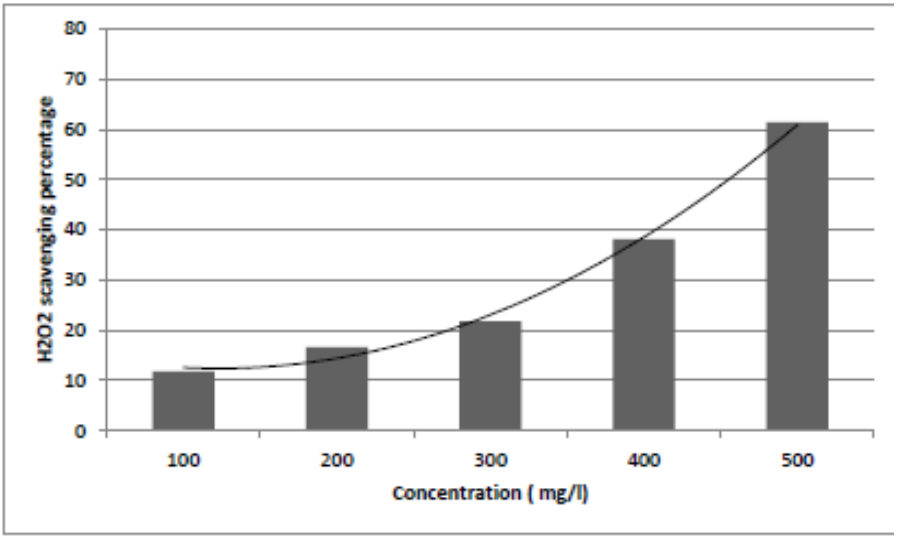

Fig. 3. Hydrogen Peroxide Scavenging Activity of milk thistle extract.

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تقييم الأنشطة المضادة للأكسدة والمضادة للميكروبات لشوك الحليب كمادة مضافة طبيعية ومحسن للصحة

الشيماء علي رواش ، سيد رشاد وغدير علي الشغبي

المركز الاقليمي للاغذيه و الاعلاف، مركز البحوث الزراعيه، الجيزه، مصر

الملخص

يُعرف نبات شوك الحليب (Silybum marianum) على نطاق واسع بخصائصه النشطة بيولوجيًا، نظرًا لمحتواه الغني بالمواد الكيميائية النباتية. تقدم هذه الدراسة تحليلاً شاملاً لمستخلص نبات الشوك الحليب، مع التركيز على خصائصه المضادة للأكسدة والمضادة للميكروبات، بالإضافة إلى تركيبه الكيميائي. باستخدام كروماتوغرافيا الغاز-قياس الطيف الكتلي، حددنا مجموعة متنوعة من المواد الكيميائية النباتية في مستخلص الشوك الحليب. وشملت هذه الأحماض الدهنية (حمض الأوليك، وحمض اللينوليك، وحمضcis-vaccenic) والفلافونويدات (سيليبين أ، وسيليبين ب، وإيزوسيليبين أ)، والمركبات الفينولية الأخرى (أسيتات فيتول ومشتقات إيزو فلافون). حيث كان نشاط مضادات الأكسدة لمستخلص الشوك الحليب كبيرًا، حيث بلغ إجمالي نشاط مضادات الأكسدة 21400 ملجم مكافئ حمض الأسكوربيك / كجم. كما وجد ان المحتوى الفينولي الكلي 10898.75 ملجم مكافئ حمض الجاليك/كجم، ومحتوى الفلافونويد الكلي 4116 ملجم مكافئ كيرسيتين/كجم. كما أظهر المستخلص أيضًا قدرة عالية على تثبيط الجذور الحرة، و اختزال ايونات الحديديك، و تثبيط نشاط بيروكسيد الهيدروجين. وكانت محتويات الصبغات للمستخلص 0.039 و 1.45 ملغم / جم للكلوروفيل والكاروتينات على التوالي. كما كشفت اختبارات مضادات الميكروبات عن قدرة المستخلص على تثبيط العديد من البكتيريا إيجابية الجرام وسالبة الجرام، وكذلك بعض انواع الفطريات. و تؤكد هذه النتائج على إمكانات مستخلص شوك الحليب كعامل طبيعي مضاد للأكسدة ومضاد للميكروبات. ونظراً للمخاوف المتزايدة بشأن المضافات الكيميائية، يمكن أن يوفر مستخلص نبات الشوك الحليب بديلاً واعداً للمضافات الغذائية والأعلاف لتعزيز الصحة والوقاية من الأمراض.

الكلمات الدالة: شوك الحليب، كروماتوغرافيا الغاز، قياس الطيف الكتلي، خصائص مضادات الأكسدة، خصائص مضادة للميكروبات.