Received: 01 October 2023, Accepted: 19 October 2023

Original Research

Journal of Advanced Veterinary Research
(2023) Volume 13, Issue 10, 2023-2028

Study of Antimicrobial Activity of *Abutilon pannosum* Choline Chloride Based Extraction as Meat Preservatives

Nagwa T. Elsharawy*

Department of Food Hygiene, Faculty of Veterinary Medicine, New Valley University, Egypt.

Department of Sport Health, College of Sport Science, University of Jeddah, Jeddah, Saudi Arabia.

***Correspondence**
Corresponding author: Nagwa T. Elsharawy
E-mail address: dr.nagwa2004@yahoo.com

Abstract

Meat industry preservation methods to extend the shelf life and maintain the microbial safety of meat products. There is big shortage in research that conducting examination of the antimicrobial activity of *Abutilon pannosum* choline chloride based extracts (APCCBE). This study aimed to assess the efficacy of APCCBE in inhibiting the growth of common meat pathogens, investigated the potential impact of APCCBE treatment on the meat products shelf life. The polar fraction of *Abutilon pannosum* leaves fine powder was obtained via a solid-liquid extraction, GC-MS analysis, Antibacterial activity of fresh APCCBE was performed against four strains of microorganisms: *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis* using Muller Hinton and six antibiotics discs (50 mg/mL) as follows; "Cefoxitin (FOX), Cephalothin (KF), Cotrimoxazole (TS), Gentamicin (GM), Augmentin (AUG) and Ampicillin (AP). Determination of the storage period of meatballs using packages containing APCCBE were each 100g of meatballs were wrapped by each package film concentration, stored in dark and cool conditions and compared with meatballs wrapped by uncoated PE film package. the study has declared that the APCCBE possess efficient antimicrobial effects against the test bacterial strains. These findings provide valuable knowledge in pathogenic bacteria treatment and food preservation, in addition to its ability in extension the shelf life of the chilled meatballs to about 2 weeks instead of 5 days only in case of control meatballs before and after cooking. Further research is needed to achieve the best application and usage of the extractions of APCCBE.

KEYWORDS
Bacillus subtilis, *E. coli*, NADES, *S. aureus*, Velvet leaf.

INTRODUCTION

The preservation of meat and meat products is of paramount importance to ensure their safety and quality throughout the supply chain, from production and processing to consumption. Meat is a valuable source of protein and essential nutrients, making it a staple in diets worldwide. However, its intrinsic characteristics, including high moisture content and nutrient-rich composition, make it highly susceptible to microbial spoilage and the growth of foodborne pathogens. As a result, the meat industry relies on a range of preservation methods to extend the shelf life and maintain the microbial safety of meat products (Elsharawy *et al*., 2018; Abo hashem *et al*., 2022; Elsharawy & Alzahrani, 2022; Maria, 2023).

Traditionally, chemical synthetic preservatives, such as sodium nitrite, sodium erythorbate, and various chemical antimicrobials, have been employed to inhibit the growth of spoilage microorganisms and pathogens in meat products. While these additives have been effective in many cases, concerns about their safety, potential health risks, and environmental impact have prompted the food industry to seek natural and eco-friendly alternatives for meat preservation. In this context, plant-based compounds have gained considerable attention for their potential antimicrobial properties and their capacity to serve as safe and sustainable preservatives (Allhyani *et al*., 2021; Novais *et al*., 2022).

Natural antimicrobial compounds derived from plants, herbs, and spices have been used in food preservation for centuries. These natural agents are often rich in bioactive compounds with inhibitory effects on microorganisms, making them attractive candidates for replacing or supplementing synthetic preservatives. The appeal of natural antimicrobials lies in their perceived safety, low environmental impact, and alignment with consumer demands for clean label and minimally processed foods (Barcenilla *et al*., 2022; Choe, 2022).

Among the vast array of potential natural antimicrobials, choline-based compounds have emerged as promising candidates for various food applications, including meat preservation. Choline is an essential nutrient for humans and animals, playing a crucial role in cell membrane integrity, neurotransmission, and lipid metabolism. Choline chloride, a water-soluble salt of choline, is commonly used as a dietary supplement and in various industrial applications, including the pharmaceutical and food industries (Verbrugghe *et al*., 2021; Zannou *et al*., 2021).

Choline chloride has attracted significant attention for its potential role as a natural meat preservative due to its antimicrobial properties. Research has suggested that choline chloride may inhibit the growth of spoilage microorganisms and foodborne pathogens in meat products, thereby extending their shelf life and enhancing their safety. This antimicrobial activity is of particular interest in the context of meat preservation, where the prevention of microbial spoilage and pathogenic contamination is essential to ensure product quality and consumer health (Yu *et al*., 2023).

Abutilon pannosum, a plant with a history of traditional me-

ISSN: 2090-6277/2090-6269/

dicinal use and known antimicrobial properties. *Abutilon pannosum*, commonly known as "velvet leaf" or "Chinese lantern," belongs to the Malvaceae family and is widely distributed in various regions. The leaves of this plant have been recognized for their potential to yield choline chloride, making it an intriguing natural source for meat preservation applications. While the antimicrobial potential of choline chloride and its extraction from *Abutilon pannosum* are of significant interest, comprehensive research into the effectiveness of this natural compound as a meat preservative is still evolving. Previous studies have shown promise, but there remains a need for systematic investigations that explore the antimicrobial activity of choline chloride extracted from *Abutilon pannosum* against a range of relevant microorganisms encountered in meat products (Sánchez-Ortega *et al*., 2014; Al kamaly *et al*., 2023).

There is big shortage in research that conducting a rigorous examination of the antimicrobial activity of *Abutilon pannosum* choline chloride based extracts. This study aimed to assess the efficacy of *Abutilon pannosum* choline chloride based extracts in inhibiting the growth of common meat pathogens. Additionally, the study investigated the potential impact of *Abutilon pannosum* choline chloride based extracts treatment on the shelf life of meat products.

MATERIALS AND METHODS

Preparation of extracts

The plant was extracted according to Fazeli-nasab *et al*. (2019). The leaves were reduced to a fine powder after being cleaned with distilled water and were dried at room temperature in the dark. The polar fraction of *Abutilon pannosum* leaves was obtained via a solid-liquid extraction. The natural deep eutectic solvent (NADES) was prepared by mixing choline chloride (ChCl), which acts as a hydrogen bond acceptor (HBA) using a magnetic stirrer (IKA® C-MAG HS 7, USA) at 80°C with the speed scale set at No. 3. The Beaker glass was covered with Parafilm and then allowed to stand for approximately 1 h or until the mixture turned into a clear liquid solution, after which it was used immediately. ChCl was bought from Xi'an Rongsheng Biotechnology Co., Ltd. (Xi'an, China). Preparation of NADES extract The NADES extraction process was performed using an ultrasonicator (Krisbow, China) with power 35 W that provide frequency 42.000 Hz. The powder samples were added to vials and then mixed with distal water. NADES was then added to the vial. The mixtures were transferred to centrifuge tubes and then centrifuged (Hettich Universal 320 Centrifuge, Germany) for 10 min at 3.283 g to separate the NADES liquid extract from the waste. The liquid layer solution was then filtered using 0.45 µm Whatman micropore filter paper. The filtrate was collected and stored in a refrigerator at 20°C until further analysis (Yuniarti *et al*., 2019).

GC-MS analysis

An initial column temperature of 35°C was used with a hold duration of about 3 minutes. The temperature was programmed to rise by 8°C every minute, peaking at 280°C. Injecting about one liter of the sample into the port caused it to evaporate right away and descend the column. Helium was employed as the carrier gas to move the sample down the column once it was inserted into the port during the operation, moving at a rate of 1 ml/min. The MS spectrum was recorded at 70 eV. After the columns were separated, the components were identified and given a second round of examination by FID. In order to ascertain the names, molecular weights, and other characteristics of the compounds, their spectra were compared to those of recognized compounds in the NIST MS 2.0 structural library.

Antibacterial Activity

Antibacterial activity of fresh *Abutilon pannosum* choline chloride based extracts was performed against four strains of microorganisms: *Staphylococcus aureus* (ATCC25923), *Escherichia coli* (NCTC9001), *Bacillus subtilis* (ATCC14028) by the well-diffusion technique using Muller Hinton and six antibiotics discs (50 mg/mL) as follows; "Cefoxitin (FOX), Cephalothin (KF), Cotrimoxazole (TS), Gentamicin (GM), Augmentin (AUG) and Ampicillin (AP)" [Mast Group/ MASTRING-S], compared with different plant leaves extractions discs. Three plates were prepared from the same extract and then incubated at 37°C/24 h., then each plate's halo clear zone was measured with a ruler to estimate the inhibition zone by millimeters and calculate the average of the three readings (CLSI, 2018).

Determination of the storage period of meatballs using packages containing Abutilon pannosum choline chloride based extracts.

Minced meatballs (about 300 g) were purchased, transferred into a sterile glass container under sanitized conditions, and packaging by polyethylene (PE) films that previously prepared by different concentration of *Abutilon pannosum* choline chloride based extracts at 10 µg/ml concentration which sprayed on the PE surface "10×150 cm"), and was left to dry at room temperature (26°C). Each 100 g of meatballs were wrapped by each package film concentration, stored in dark and cool conditions (4°C) and compared with meatballs wrapped by uncoated PE film package. The shelf life of meatballs has been observed and examined daily by checking the sensory characters of the meatballs until the spoilage evidence became clear (Yilmaz and Demirci, 2010).

Sensory evaluation of the meat samples

About 500g from each treatment were formed into small meatball. The samples were prepared then given in two forms (raw & cooked) to panel members (n=10) who were not trained in the sensory analysis of meat. Five characteristics point were given to panelists as follow; 1: very poor; 2: poor; 3: good; 4: very good, and 5: excellent. Panelists were considered the above points for evaluation of color, flavor, taste and consistency of the meatball samples (Huss, 1995; Das *et al*., 2012).

Statistical analysis

The statistical program, SPSS version 16 for window, was used for the determination of means, standard error, and analysis of variance (ANOVA) using the one-way (mean at the significance level of ($P<0.05$). Statistical significance was tested at the 5% level of significance in this study (SPSS 16, 2007).

RESULTS

Gas chromatography of Abutilon pannosum choline chloride based extracts

Gas chromatography and Mass spectroscopy were used to analyze the bioactive compounds of *Abutilon pannosum* choline chloride based extracts. The active compounds were shown with their retention time, molecular formula, molecular weight, and

beak area in Table 1 and Figure 1. The plant extract showed the presence of multiple phytochemical compounds which has an antimicrobial effect such as; Tristrimethylsilyl Ether, Dihydroxyvitamin D3, Eicosatrienoic Acid, Aspidospermidine, Cyclopropaneoctanoic Acid, Pseudosolasodine Diacetate, Pentadecanoic Acid, Isopropyl Palmitate, Acetamide, Dodecenoic Acid, Octadecatrienoic Acid, Oleic Acid, Heptatriacotanol, Hexadecenoic Acid, Panaxydol, Eicosatetraenoic Acid, Dodecatetraenal, Acetic Acid, Benzofuranacetic Acid, Cyclopropaxime, Ethanimidothioic Acid, Ropanediol, Ethanimidothioic Acid, Ofuranosyl, Hexopyranoside, Diethyl Mercaptal-Pentaacetate, Methyl 6-Oxoheptanoate from higher to lowest bioactive compounds respectively.

The bioactive analysis of *Abutilon pannosum* extract declared that *Abutilon pannosum* contained more than 25 bioactive compounds in different molecular weight and different concentrations. Almost of these bioactive compounds having antimicrobial effect against different microorganisms.

Comparison between the antimicrobial activity of Abutilon pannosum choline chloride based extracts

Table 3 declared the in vitro inhibitory effect of *Abutilon pannosum* choline chloride based extracts in comparison with commercial antibiotics as follows; the most effective extraction against all tested microorganisms detected *Abutilon pannosum* choline chloride based extracts between 4.2-3.4 cm, while lower results detected in *Abutilon pannosum* water based extracts (1.0-0.8) cm as follow; the highest inhibition effect were against *E. coli* were 4.2±1.40 cm and 1.5±0.0 cm for *Abutilon pannosum* choline chloride based extracts and *Abutilon pannosum* water based extracts respectively, followed by 3.4±1.40 cm and 1.0±0.0 cm against *Bacillus subtilis* for *Abutilon pannosum* choline chlo-

Figure 1. GC analysis peaks of *Abutilon pannosum* choline chloride based extraction (cm).

Table 1. Phytochemical constituents identified in *Abutilon pannosum* choline chloride based extract by GC-MS analysis.

Peak no.	RT	Compound Name	Area%	MF	Molecular Formula	Molecular weight
1	16.23	Methyl 6-oxoheptanoate	7.94	698	$C_8H_{14}O_3$	158
2	16.29	diethyl mercaptal-pentaacetate	1.76	646	$C_{20}H_{32}O_{10}S_2$	496
3	16.35	ulofuranosyl hexopyranoside	4.03	672	$C_{18}H_{32}O_{16}$	504
4	16.65	Ethanimidothioic acid	0.35	742	$C_7H_{13}N_3O_3S$	219
5	16.71	Propanediol	0.46	599	$C_{17}H_{36}O_3$	288
6	19.15	Ethanimidothioic acid	0.2	705	$C_7H_{13}N_3O_3S$	219
7	20.1	Cyclopropaxime	0.4	654	$C_{15}H_{25}NO$	235
8	20.66	benzofuran acetic acids	0.17	671	$C_{16}H_{20}O_4$	276
9	21.55	Acetic acid	0.52	599	$C_{16}H_{26}O_2$	250
10	21.55	Dodecatetraenal	0.52	654	$C_{15}H_{22}O$	218
11	21.63	Eicosatetraenoic acid	0.38	639	$C_{21}H_{34}O_2$	318
12	21.96	Panaxydol	0.3	659	$C_{17}H_{24}O_2$	260
13	23.07	Hexadecenoic acid	0.37	675	$C_{18}H_{34}O_2$	282
14	25.27	Heptatriacotanol	1.64	702	$C_{37}H_{76}O$	536
15	25.34	Oleic Acid	0.24	639	$C_{18}H_{34}O_2$	282
16	26.99	Octadecatrienoic Acid	0.33	646	$C_{23}H_{32}O$	324
17	27.07	Dodecenoic acid	0.46	657	$C_{13}H_{24}O_3$	228
18	27.12	Acetamide	0.49	690	$C_{11}H_{18}N_2O_2$	210
19	29.01	Isopropyl palmitate	5.84	733	$C_{19}H_{38}O_2$	298
20	29.13	Pentadecanoic acid	0.86	724	$C_{20}H_{323}O_{10}S_2$	496
21	30.45	Pseudosolasodine diacetate	0.69	658	$C_{31}H_{49}NO_4$	499
22	30.58	Eicosatetraenoic acid	0.58	723	$C_{20}H_{34}O_2$	306
23	30.75	Cyclopropaneoctanoic acid	0.2	738	$C_{22}H_{38}O_2$	334
24	30.95	Aspidospermidine	0.32	770	$C_{22}H_{30}N_2O_3$	370
25	32.57	Eicosatrienoic acid	0.33	788	$C_{20}H_{34}O_2$	306
26	34.14	Dihydroxyvitamin D3	0.35	724	$C_{30}H_{52}O_3Si$	724
27	35.88	Tristrimethylsilyl ether	0.41	668	$C_{14}H_{17}NO_9$	343

ride based extracts and *Abutilon pannosum* water based extracts respectively, the lowest inhibition effects against *S. aureus* were 3.0±1.60 cm and 0.8±0.0 cm for *Abutilon pannosum* choline chloride based extracts and *Abutilon pannosum* water based extracts respectively. The obtained results declared a higher inhibition effect against tested Gram-negative microorganisms than Gram-positive tested species.

Determination of the storage period of meatballs using packages containing Abutilon pannosum choline chloride based extracts

Table 3 and Fig. 2 declared the addition of the extracts extended the shelf life of the chilled meatballs to about 2 weeks instead of 5 days only in case of control meatballs before and after cooking as following; all physical characters were excellent during first and second days of storage, pH were ranged from 5.85 to 5.90 on raw and cooked samples while from 4rd day the quality become gradually decrease to very good in all parameters before and after cooking, pH were 5.91–5.97 in raw and cooked samples. Almost all parameters remained very good during the 6th day of storage except the odor and consistency which become 'good' in raw meatballs samples, the pH ranged from 5.95 and 5.97 in raw and cooked samples during the 8th day of chilling the quality decrease to good in almost parameters except the odor and consistency which become bad in raw meatballs samples. Spoilage appeared from the 10th day of storage as all parameters were bad quality except the color of raw meat which was good, pH range from 5.98 –5.99 in raw and cooked samples. The deterioration signs become very clear on the 12th day as all parameters were bad while the odor and consistency of raw samples were very bad in raw meatballs samples, pH were 6.2 and 6.3 in raw and cooked samples. The complete spoilage recoded on the 14th day of chilling the meatballs samples, pH was 6.5 and 6.6 in raw and cooked meatballs samples.

DISCUSSION

The bioactive analysis of *Abutilon pannosum* extract declared that it contained more than 25 bioactive compounds in different molecular weight and different concentrations. Most of these bioactive compounds had antimicrobial effect against different microorganisms. The antimicrobial activity of *Abutilon pannosum* choline chloride based extracts declared a higher inhibition effect against tested Gram-negative microorganisms than Gram-positive tested species although the effect of the extracts were more effective than almost all conventional antibiotics.

Similar results were found by Aadesariya *et al*. (2018). while Tiwari *et al*. (2016) detected different results by isolation of 13 biomolecules only. The mass spectrometer analyzes the compounds eluted at different times to identify the nature and structure of the compounds. The large compound fragments into small

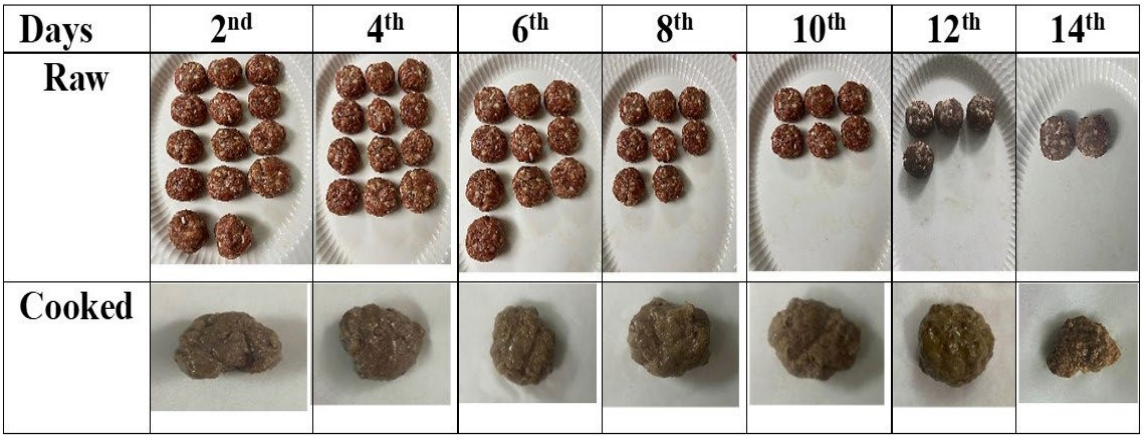

Figure 2. Sensory changes of meat along the preservation chilling periods after addition of *Abutilon pannosum* choline chloride extraction.

Table 2. Antimicrobial activity of *Abutilon pannosum* choline chloride based extracts and *Abutilon pannosum* water based extracts (cm).

Microorganisms	Choline chloride	Water extract	FOX	KF	GM	TS	AUG	AP
E. coli	4.2±1.40	1.5±0.0	3	2.5	2.5	2.6	2.5	2
Bacillus subtilis	3.4±1.40	1.0±0.0	3.4	2.5	2	2.5	2.5	2
S. aureus	3.0±1.60	0.8±0.0	3	0	3.5	3.5	3.5	4.5

Table 3. Sensory Evaluation of minced meat balls shelf life along the preservation chilling periods after addition of *Abutilon pannosum* choline chloride extraction Samples.

Days	Raw				Cooked			
	Color	Odor	Consistency	pH	Color	Odor	Consistency	pH
0	5	5	5	5.85	5	5	5	5.88
2	5	5	5	5.88	5	5	5	5.9
4	4	4	4	5.91	4	4	4	5.94
6	4	3	3	5.93	4	4	4	5.95
8	3	2	2	5.95	3	3	3	5.97
10	3	2	2	5.98	2	2	2	5.99
12	2	1	1	6.2	2	2	2	6.3
14	1	1	1	6.5	1	1	1	6.6

compounds giving rise to appearance of peaks at different m/z ratios. These mass spectra are fingerprint of those compounds which can be identified from the data library. Some of these isolated compounds are reported to possess significant medicinal applications. Further investigation may lead to isolation of bio-active compounds, their structural elucidation and screening of pharmacological activity which will be helpful in drug development. Only two compounds gamma-sitosterol and lupeol has been identified from hexane extract of A. indicum leaves through GCMS analysis (Girija *et al*., 2014).

Kamel *et al*. (2017) has done quantitative estimation of lupeol from A. indicum extract. Aadesariya *et al*. (2017) performed GC–MS chromatogram of leaves of *Abutilon pannosum* showed 9 peaks phytochemical compounds as following: 9,12-Octadecadienoic (45.05%), 9-Octadecenoic acid (33.18%), Hexadecanoic acid (12.12%), Octadecanoic acid (5.88%), Octadecatrienoic acid (2.62%), 9,12,15-Eicosanoic acid (0.60%), 9-Hexadecenoic acid (0.22%), 11-Eicosenoic acid (0.16%), Methyl tetradecanoate (0.10%), Octanoic acid (0.04%) and Tridecanoic acid (0.03%) in hexane extract of *A. pannosum* and 9,12-Octadecadienoic (48.50%)>9-Octadecenoic acid (32.02%)>Hexadecanoic acid (11.48%)>Octadecanoic acid (6.20%)>9,12,15-Octadecatrienoic acid (0.81%)>Eicosanoic acid (0.56%)>9-Hexadecenoic acid (0.19%)>11-Eicosenoic acid (0.18%) and Methyl tetradecanoate (0.06%) in hexane extract of *G. tenax*.

Survase *et al*. (2013) carried out the antibacterial activity of the plant *Abutilon pannosum* against both Gram positive and negative microorganisms. The results exhibit antimicrobial activity against the Gram positive organisms such as *B. subtilis*, *S. aureus* and gram negative organisms *E. coli*, *P. aerugenosa* with a maximum diameter of zone of inhibition ranging from 23.3 mm accompanied by ≥19.4 and 21.4, 20.0 mm, respectively. It produced a comparable activity similar to the standard antibiotics taken for the study.

Al-Ghamdi (2022) determined the phytochemical constituents and in vitro antimicrobial activity of *Abutilon pannosum* in Al-Baha region, Saudi Arabia. The phytochemical constituents and antimicrobial activity against Gram-positive bacteria (*Staphylococcus aureus*, Bacillus cereus), Gram-negative bacteria (*Escherichia coli*, *Pseudomonas aeruginosa*). Results showed that the extracts had saponins, coumarin, alkaloids, tannins, flavonoids, and steroids. GC/MS analysis of ethanol extract revealed 32 compounds, the most important of which were 9-Octadecenoic acid (Z)-, methyl ester, and methyl 10-trans,12-cis-octadecadienoate, while chloroform extract revealed 36 bioactive compounds, the most important of which was phytol, and aqueous extract revealed 43 bioactive compounds, the most important of which was benzyldiethyl (2,6-xylylcarbamoylmethyl) ammonium benzoate. Ethanol extract's antimicrobial activity increased with concentration, from inactive at 25-50mg/mL to moderately active at 100-200mg/mL to active at 300mg/mL. At all concentrations, the extract was most effective against pathogenic microorganisms, which may be used in food and pharmaceutical industry.

Determination of the storage period of meatballs using packages containing *Abutilon pannosum* choline chloride based extracts revealed that the addition of the extracts extended the shelf life of the chilled meatballs to about 2 weeks instead of 5 days only in case of control meatballs before and after cooking. We failed to receive to similar studies using *Abutilon pannosum* choline chloride based extracts to compared our results with it.

CONCLUSION

Abutilon pannosum choline chloride based extracts possess efficient antimicrobial effects against the test bacterial strains. These findings provide valuable knowledge in pathogenic bacteria treatment and food preservation, in addition to its ability in extension the shelf life of the chilled meatballs to about 2 weeks instead of 5 days only in case of control meatballs before and after cooking. Further research is needed to achieve the best application and usage of the extractions of *Abutilon pannosum* choline chloride based extracts.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

Aadesariya, M., Ram, V., Dave, P., 2017. Soxhtherm Extraction, Isolation and Identification of Fatty Acids Present in the Hexane Extract of *Abutilon pannosum* and Grewia Tenax Using Gas Chromatography-Mass Spectrometry. International Journal of Advanced Research in Chemical Science 4, 26-34.

Aadesariya, M., Ram, V., Dave, P., 2018. Phytochemical Analysis of leaves Extract of *Abutilon pannosum*inn-Butanolfor its Bioactive Components through Gas Chromatography-Mass Spectrometry (GC-MS). Journal of Engineering 8, 11-21.

Abo hashem, M.E., Enany, M.E., Ahmed, A.M, Huda, E.I., Elsharawy, N.T. 2022. Estimation of potential bacteriological hazards and their genetic virulence determinants in beef meals provided to intensive care patients. Food Research 6, 286-293.

Al kamaly. O., Alanazi, A., Conte, R., Imtara, H., 2023. Phytochemical composition and insight into antibacterial potential of *Origanum vulgare* essential oil from Saudi Arabia using *In Vitro* and In Silico Approaches. Processes. 11, 650-657.

Al-Ghamdi, A., 2022. Phytochemical Screening, Chemical Composition, and Antimicrobial Activities of *Abutilon pannosum* (Forst.f.) Schlecht. Collected from Shada Mountain, Al- Baha Region, Saudi Arabia. Egypt. J. Microbiol. 57, 35-47.

Allhyani, A.A., Baeshen, M.N., Elsharawy, N.T., 2021. Molecular Identification of Newly Isolated Foodborne Bacterial Strains from Chevon Meat. Bioscience Biotechnology Research Communications. 14, 1083-92.

Barcenilla, C., Ducic, M., Lopez, M., Prieto, M., Alvarez-Ord, A., 2022. Review Application of lactic acid bacteria for the biopreservation of meat products: A systematic review. Meat Science .183, 108661-6.

Choe, D., 2022. Microbe-derived antimicrobial red pigments for color formation and microbial growth control in sausage: A mini-review. Korean J. Food Preserv. 29, 837-851.

CLSI (Clinical and Laboratory Standards Institute), 2018. Document Vet08. Performance Standards for Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria Isolated from Animals, Approved Standard, Fourth Edition. CLSI, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898, USA.

Das, A.K., Rajkumar, V., Verma, A.K., Swarup, D. 2012. Moringa oleiferia leaves extract: a natural antioxidant for retarding lipid peroxidation in cooked goat meat patties. International Journal of Food Science and Technology. 47, 585-591.

Elsharawy, N.T., Alzahrani, H.A. 2022. Serological identification of *Escherichia coli* multidrug-resistance genes isolated originated from retailed Chicken meat. Bioscience Research. 19, 2082-2087.

Elsharawy, N.T., Ahmad, A.M., Abdelrahman, H.A., 2018. Quality assessment of nutritional value and safety of different meat. Journal of Food: Microbiology, Safety and Hygiene, 3, 2059-2476.

Fazeli-Nasab, B, Sayyed, Z., Sobhanizadeh, A. 2021. In Silico Molecular Docking Analysis of αPinene: An Antioxidant and Anticancer Drug Obtained from *Myrtus communis*. International Journal of Cancer Management. 14, e89116.

Girija, S., Duraipandiyan, V., Kuppusamy, P.S., Gajendran, H., Rajagopal, R. 2014. Chromatographic characterization and GC-MS evaluation of the bioactive constituents with antimicrobial potential from the pigmented ink of Loligoduvauceli. Int. Sch. Res. Not. 20, 1–7.

Huss, H.H., 1995. Quality and quality changes in fresh fish. FAO. FAO Fisheries Technical Paper No. 348.

Kamel, A.I., Elfedawy, M.G., Elshamy, M.M., Abdel-Mogib, M., 2017. New Physiologically Active Kaempferol Glucoside from *Abutilon pannosum*. Int. J. Sci. Eng. Appl., 6, 78-87.

Maria, F., 2023. Meat Consumption, Sustainability and Alternatives: An Overview of Motives and Barriers. Foods. 12, 2144-2150.

Novais, C., Molina, A., Abreu, R., Santo-Buelga, C., Ferreira, I., Pereira, C., Barros, L., 2022. Natural Food Colorants and Preservatives: A Review, a Demand, and a Challenge. J. Agric. Food Chem. 70, 2789–2805.

Sánchez-Ortega, I., García-Almendárez, B.E., Santos-López, E.M., Amaro-Reyes, A., Barboza-Corona, J.E., Regalado, C., 2014. Antimicrobial edible films and coatings for meat and meat products preservation. The Scientific World Journal 2014, 248935.

Survase, S., Sarwade, B., Chavan, D., 2013. Antibacterial activity of various extracts of *Abutilon pannosum* (Forst.f.) Schlecht. leaves. African

Journal of Plant Science 7, 128-130.

Tiwari, S., Mishra, S., Misra, D., Upadhyay, D., 2016. Identification of New Bioactive Compounds from Fruit of *Abutilon indicum* through GCMS Analysis. Biological Forum – An International Journal. 8, 548-554.

Verbrugghe, A., Rankovic, A., Armstrong, S., Santarossa, A., Kirby, G., Bakovic, M., 2021. Serum Lipid, Amino Acid and Acylcarnitine Profiles of Obese Cats Supplemented with Dietary Choline and Fed to Maintenance Energy Requirements. Animal. 11, 2196-2202.

Yilmaz, I., Demirci, M., 2010. Effect of different packaging methods and storage temperature on microbiological and physicochemical quality characteristics of meatball. Food Sci Technol Int. 16, 259-65.

Yu, W., Guo, J.,Liu, Y., Xue, X., Wang, X., Wei, L., Ma, J., 2023. Potential Impact of Combined Inhibition by Bacteriocins and Chemical Substances of Foodborne Pathogenic and Spoilage Bacteria: A Review. Foods. 12, 3128-3132.

Yuniarti, E., Saputri, F., Mun'im, A., 2019. Application of the natural deep eutectic solvent choline chloride-sorbitol to extract chlorogenic acid and caffeine from green coffee beans (*Coffea canephora*). J. Appl. Pharm. Sci. 9, 82-90.

Zannou, O., Pashazadeh, H., Ghellam, M., Ibrahim, S., Koca, I., 2021. Extraction of Anthocyanins from Borage (*Echium amoenum*) Flowers Using Choline Chloride and a Glycerol-Based, Deep Eutectic Solvent: Optimization, Antioxidant Activity, and In Vitro Bioavailability. Molecules 27, 134-140.