Benha Veterinary Medical Journal 47 (2024) 130-134

Official Journal Issued by Faculty of Veterinary Medicine

Benha Veterinary Medical Journal

Journal homepage: https://bvmj.journals.ekb.eg/

Since 1990

Original Paper

Effect of different marinades on the production of polycyclic aromatic hydrocarbons in beef steaks

Shereen H. A. Abd El- Fatah[1], El Shater M. A[2], Wessam Hassan[3] and Hemmat M. Ibrahim[1]

[1] *Food Hygiene Department, Faculty of Veterinary Medicine, Benha University.*

[2] *Animal Health Research Institute, Agriculture Research Centre, Doki, Giza.*

[3] *Plant Pathology Research Institute, Agriculture Research Centre, Giza.*

ARTICLE INFO

Keywords

PAH4

Charcoal Grilled beef

Lemon,

Marinated

Thyme oil

Received 17/10/2024
Accepted 04/11/2024
Available On-Line 31/12/2024

ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) are of public health significance due to their potential to cause cancer, genetic abnormalities, and congenital deformities. This investigation aimed to evaluate the effect of different marinades in reducing PAH levels in charcoal grilled beef steak. Sixty samples were collected from the market of Cairo Governorate, Egypt, ten samples were control samples and fifty samples were treated with different marinade. The fifty samples were divided into five groups: group1: ten samples were marinated with lemon juice, group 2: ten samples were marinated with thyme oil, group 3: ten samples were marinated with sugar, water, cinnamon, onion, turmeric, coriander and salt, group 4: ten samples were marinated with spices, onion, cooking oil, turmeric, sugar, cinnamon, coriander salt and water, group 5: ten samples were marinated with sugar, onion, water, salt, turmeric, cinnamon, coriander, thyme oil and lemon juice.

Each group was analyzed to determine level of Benzo[a]pyrene, Benzo[b]fluoranthene, Benz[a]anthracene, Chrysene polycyclic aromatic hydrocarbons then compared to control samples to determine the effect of each marinade on PAH4.

The results indicated that the addition of lemon juice and replacement cooking oil with thyme oil had the significant effect in reducing PAH4 in the treated beef steak samples.

1. INTRODUCTION

The main goal of food preparation is to provide safe food with minimal hazardous elements. Research indicates that human exposure to polycyclic aromatic hydrocarbons (PAHs), mostly originates from food, particularly meat, which is a significant source of dietary protein, so It is important to be aware of the sources of PAHs in order to minimize exposure and reduce potential health risks. Inadequate culinary knowledge can lead to the production of PAHs in cooked meat dishes, which are human carcinogens and pose a risk of cancer and related disorders.

Polycyclic aromatic hydrocarbons (PAHs) may be generated during high-temperature processes such as drying, toasting, roasting, or frying (Dutta et al., 2022). Nevertheless, PAHs can be produced through incomplete combustion of thermal agents, fat falling onto flames during grilling, or pyrolysis of organic materials, releasing free radicals responsible for PAH production (Dutta et al., 2022).

Exposure to PAHs may result in a 30-50% rise in breast cancer incidence, oxidative DNA damage, and adverse effects on neuro-differentiation (Singh et al., 2016). Furthermore, PAHs may induce cardiopulmonary and cardiovascular illnesses by generating reactive oxygen species, leading to oxidative stress and inflammatory reactions (Singh et al., 2016). The European Food Safety Authority (EFSA, 2008) assessed the PAH4 (BaA, BaF, BbP, and Chr) as indications of carcinogenicity. The maximum residue levels for PAH4 and B[a]P are 12 µg/kg and 2 µg/kg in beef meat (EC 2011).

Marination is a prevalent culinary technique that involves submerging meat in a liquid mixture, known as a marinade, prior to cooking. This amalgamation often contains acids (such as lemon juice or vinegar), oils, spices, and herbs. Marination is often used to enhance the taste and tenderness of meat by decomposing muscle fibers (Edikou et al., 2018). However, marination minimizes cooking time, since marinated meat cooks rapidly, hence reducing its exposure to higher heat. This method may reduce the formation of harmful compounds and prevent moisture loss while cooking, especially in grilling or frying techniques. Understanding the role of marinades in minimizing the production of polycyclic aromatic hydrocarbons (PAHs) is crucial for promoting safe cooking practices and mitigating the health risks associated with the consuming of grilled or barbecued meats (Büyükkurt et al., 2020).

Research suggests that the formation of PAHs may be reduced by specific components of marinades (Viegas et al., 2014). he generation of PAHs during high-temperature heating may be reduced by ingredients with antioxidant properties, such as garlic, onion, and herbs (e.g., rosemary and thyme) (Viegas et al., 2014). The chemical reactions responsible for PAHs are inhibited by acidic components, including lemon juice, vinegar, and yogurt, which create an acidic environment (Nor et al., 2022). Oil-based marinades provide a protective layer on the meat's surface, which

* Correspondence to: shereenabdelmonem20@gmail.com

reduces direct contact with heat and flames, thereby reducing the formation of PAHs (Sampaio et al., 2021).

Thyme (*Thymus vulgaris L*.) is a commercially significant plant that is widely recognized for its essential oil aimed at food flavor and significant health benefits (Zengin and Baysal, 2015). Thyme extracts are appropriate as sources of synthetic antioxidants in the food industry due to their antioxidant properties (Wang et al., 2019). The phenolic compounds present in thyme essential oil are suggested that they are responsible for the inhibitory effects against PAHs that are produced during heating (Wang et al., 2019).

This study aimed to assess the effect of different marinades on the production of carcinogenic Polycyclic Aromatic Hydrocarbons (PAHs) to enhance the safety of meat prepared using traditional charcoal grilling methods.

2. MATERIAL AND METHODS

The protocol of this work was approved by the Scientific Research Ethics Committee of the Faculty of Veterinary Medicine, Benha University, with ethical approval number (BUFVTM 38-09-23).

1. Sampling technique (Farhadian et al*., 2012; Sampaio* et al*., 2021):*

hadian et al*., 2012; Sampaio* et al*., 2021):*

Sixty samples of beef steak, each weighing 50 grams, were collected from the same beef cut from a market in the Cairo Governorate of Egypt. The collected samples were securely enclosed in a plastic bag and placed in a dry ice box without any delay. The samples were divided into 10 control samples and 50 treated samples. Control samples were immersed in sterile distilled water for 15 minutes, then drained for 5 minutes, and then grilled over charcoal. The first marinade group, the samples were soaked in a lemon juice solution for 15 minutes, followed by a 5-minute draining period, and then grilled on charcoal. The second group, the samples were soaked in thyme oil for 15 minutes, followed by a 5-minute draining period before grilling on charcoal. The third group, the samples were soaked in marinade 3, which consisted of sugar, onion, water, salt, turmeric, cinnamon, and coriander for 4 hours at 4°C, followed by a 5-minute draining period before grilling on charcoal. The fourth group, the samples were soaked in marinade 4, which consisted of a mixture of spices (cumin, paprika, chili powder and black pepper), sugar, water, turmeric, onion, oil, cinnamon, coriander, and salt for 4 hours at 4°C, followed by a 5-minute draining period before grilling on charcoal. The fifth group, the samples were soaked in marinade 3 with the addition of lemon juice and thyme oil for 4 hours at 4°C, followed by a 5-minute draining period before grilling on charcoal. The samples were transferred to the Pesticide Residue Department of the Central Pesticide Laboratory at the Agricultural Research Center in Giza to detect polycyclic aromatic hydrocarbons, compare them with the accepted standard limits, and evaluate their suitability for human consumption.

2. Chemicals and Reagents:

Using reference standards from Sigma-Aldrich, a 1000 μg/mL stock solution of four polycyclic aromatic hydrocarbons (PAHs) was prepared with a purity that surpassed 95%. The PAHs include chrysene, benzo(a)pyrene (surrogate standard), benz (a)anthracene and benzo(b)fluoranthene.

3. Apparatus:

Fifty ml tubes with screw caps composed of Polytetrafluoroethylene or polyethylene and fifteen ml tubes containing one gram of magnesium sulfate were acquired for sample extraction. The apparatus comprises a centrifuge with a maximum operational speed of 4000 rpm (Heraeus Labofuge 400), a vortex mixer, and automated pipettes (Hirschmann Laborgerate) designed for volumes from 10 μL to 100 μL and from 100 μL to 1000 μL. Furthermore, a 10 mL solvent dispenser (Hirschmann Laborgerate) for acetonitrile is supplied. The glassware was washed with soap and water, cleaned with acetone, and dried at 90°C before use.

4. Sample Extraction (Guide, 1998).

The validation method must consider the context of suitability for purpose and cost-benefit standards. Approximately 10 grams of meat sample were measured in a 50 ml Teflon centrifuge tube. Subsequently, 50 μl of a 10 μg/mL pyrened10 solution, acting as a surrogate standard at a concentration of 50 μg/kg, was included. Each set of six repetitions was then augmented with 20, 100, and 500 μl of a 1 μg/ml spiking solution to attain final concentrations of 2, 10, and 50 μg/kg, respectively. Ten ml of acetonitrile were used for extraction, agitated for two minutes, amalgamated with Agilent QuEChERS, agitated for one minute, and centrifuged at 4000 revolutions per minute for five minutes. Aliquots of the resulting supernatant were transferred to a Teflon tube containing $MgSO_4$, vortexed for 30 seconds, and then centrifuged at 4000 rpm for two minutes. Four ml of the acetonitrile layer was thereafter placed into a 50 ml flask and evaporated to near dryness.

5. Clean up by Packed Solid Phase Extraction "SPE" (Khorshid et al*., 2015):*

All meat extracts were processed using a packed solid phase cleaning cartridge that was prepared in-house. A glass wool was placed into a ten ml syringe. Subsequently, one gram of 20% deactivated silica gel and 0.2 gram of $MgSO_4$ were measured and treated with five ml of an n-hexane/dichloromethane combination in a 3:2 ratio. The sample extract was then introduced into the cartridge using ten ml of the eluent (n-hexane/dichloromethane). Consolidate fractions in a 50 ml flask, then evaporate using a rotary evaporator at 40°C until virtually desiccated. The residue was diluted in two ml of toluene and then analyzed by GC-MS.

6. GC-MSD Conditions (Khorshid et al*., 2015):*

The Agilent 6890N series gas-chromatography instrument, used with the 5975 series mass selective detector and equipped with an Agilent GC Column model J&W HP-5ms Ultra Inert (30 m length, 0.25 mm internal diameter, 0.25 μm film thickness), was employed for the qualitative and quantitative analysis of PAHs. Helium gas functioned as the carrier gas, while the column was maintained at a constant flow rate of 1.3 mL/min.

The back injector line was maintained at a temperature of 260 degrees Celsius. The injection volume employed was 1.0 μL in splitless mode. The column temperature was initially set at 90°C for 2 minutes, subsequently raised to 180°C at a rate of 15°C/min and maintained for 15 minutes. The temperature subsequently increased to 250°C at a rate of 10°C/min, maintained for two minutes, and then raised to 290°C at the same rate, where it was held for 10 minutes. The mass spectrometer operated in ionization mode, and spectra were acquired within a mass range of 45–450 m/z.

4. RESULTS

Table (1) indicated that Chrysene, Benzo(a)pyrene, Benzo[b]fluoranthene, and Benzo(a)anthracene were detected in both the control and juice of lemon marinated charcoal grilled beef steak samples. The results indicated that the mean concentration values of PAH4 for the control and lemon juice marinated samples were 57.98 ± 7.07 and 5.01 ± 0.97 µg/kg. The maximum level for PAH4 in beef products is 12 µg/kg, as stipulated by Commission Regulation (EC, 2011).

Table (2) demonstrates the presence of benzo(a)anthracene, chrysene, benzo(a)pyrene and benzo[b]fluoranthene, in charcoal-grilled beef samples marinated with thyme oil, relative to control samples. The results demonstrated that the average concentration values of PAH4 in control and thyme oil marinated samples were 57.98 ± 7.07 and 10.19 ± 3.40 µg/kg, respectively.

The results presented in table (3) demonstrate the presence of chrysene, benzo(a)anthracene, benzo[b]fluoranthene, and benzo(a)pyrene in samples of charcoal-grilled beef steak from groups 3, 4, and 5, in comparison to the control samples. The results indicated that the mean concentration values of PAH4 for the control, marinade 3, 4, and 5 were 57.98 ± 7.07, 21.30 ± 4.99, 12.75 ± 6.59, and 4.38 ± 0.90 µg/kg, respectively.

Table 1 Concentrations of identified polycyclic aromatic hydrocarbons residues (µg/kg) in examined charcoal grilled beef steak samples marinated with lemon juice (*n= 10)

PAH Compound (Abbreviation)	Control	Lemon juice margination
Chrysene (Chr)	19.25±5.34 (11-27.6)*	0.45±0.28 (0.1-0.78)*
Benz[a]anthracene (BaA)	13.92±4.26 (6.05-19.07)*	1.26±0.43 (1.01-2.11)*
Benzo[b]fluoranthene (BbF)	9.19±4.52 (UDL-15.3)*	0.66±0.40 (0.036-1.06)*
Benzo[a]pyrene (BaP)	24.80±12.29 (6.21-22.90)*	2.64±0.90 (1.08-3.41)*
PAH4	57.98±7.07 (46.46-68.18)*	5.01±0.97 (3.57-5.65)*

*Mean ± SD with range, n=number of samples, UDL= under detection limit

Table 2 Concentrations of identified polycyclic aromatic hydrocarbons residues (µg/kg) in examined charcoal grilled beef steak samples with thyme oil (*n= 10).

PAH Compound (Abbreviation)	Control	Thyme oil
Chrysene (Chr)	19.25±5.34 (11-27.6)*	0.89±0.37 (0.42-1.63)*
Benz[a]anthracene (BaA)	13.92±4.26 (6.05-19.07)*	3.34±1.41 (1.069-5.27)
Benzo[b]fluoranthene (BbF)	9.19±4.52 (UDL-15.3)*	UDL
Benzo[a]pyrene (BaP)	24.80±12.29 (6.21-22.90)*	5.97±3.02 (1.69-9.36)*
PAH4	57.98±7.07 (46.46-68.18)*	10.19±3.40 (3.79-15.05)*

*Mean ± SD with range, n=number of samples, UDL= under detection limit

Table 3 Concentrations of identified polycyclic aromatic hydrocarbons residues (µg/kg) in examined charcoal grilled beef steak samples with various marinades. (*n= 10)

PAH Compound (Abbreviation)	Control	marinade 3	marinade 4	marinade 5
Chrysene (Chr)	19.25±5.34 (11-27.6)*	3.10±1.63 (1.036-5.6)*	1.27±0.88 (UDL-2.69)*	0.34±0.21 (UDL-0.7)*
Benz[a]anthracene (BaA)	13.92±4.26 (6.05-19.07)*	6.95±2.12 (2.45-10.78)*	4.27±2.32 (1.025-7.47)*	1.06±0.25 (0.71-1.09)*
Benzo[b]fluoranthene (BbF)	9.19±4.52 (UDL-15.3)*	4.11±2.77 (1.01-7.5)*	0.43±0.70 (UDL-2.01)*	UDL
Benzo[a]pyrene (BaP)	24.80±12.29 (6.21-22.90)*	7.14±3.65 (11.97-3.98)*	6.78±4.04 (1.87-15.2)*	2.99±0.90 (1.06-4.09)*
PAH4	57.98±7.07 (46.46-68.18)*	21.30±4.99 (13.364-29.07)*	12.75± 6.59 (6.77-23.59)*	4.38±0.90 (2.35-5.13)*

*Mean ± SD with range, n=number of samples, UDL= under detection limit

4. DISCUSSION

When several kinds of marinades were applied to charcoal beef steak, the decrease of BaP and PAH4 was variable. This was due to the fact that different marinades had distinct effects. Onions, garlic (Janoszka, 2011), and coriander (Yu et al., 2023) are natural antioxidants that have been utilized extensively to regulate the levels of polycyclic aromatic hydrocarbons (PAHs) in grilled meat products. These antioxidants have the potential to limit the development of PAHs in meat (Lee et al., 2020).

Following these findings, Yunita et al., 2023 revealed that lemon marinade led to decreased levels of carcinogen PAH4 when compared to control samples. This finding is in line with the findings of this investigation. Since lemon (*Citrus limon L*) is a citrus fruit, and a natural antioxidant, it can inhibit the creation of PAH carcinogens in grilled meat (Nor et al., 2022).

The procedure of marinating with lemons has the potential to lower the levels of carcinogens. Lemon juice includes vitamin C and vitamin E, both of which protect body cells from harm caused by free radicals (Lee et al., 2020). Additionally, lemon juice contains phenol, which reduces the incidence of fat oxidation by neutralizing reactive oxygen, which may induce the creation of PAHs (Wang et al., 2019).

In this respect, Hussein et al. (2023) revealed a decrease in the mean concentration levels of BaP and PAH4 following the application of a thyme oil marinade. The control samples had concentrations of 4.87±3.36 and 8.57±6.32, respectively, while the thyme oil marinade had concentrations of 17.07±8.33 and 32.67±22.73µg/kg. The results were similar to what was found in our study. The mean concentration levels of BaP and PAH4 in the control and marinade 2 group were 19.25±5.34, 57.98±7.07, 5.97±3.02, and 10.19±3.40 µg/kg, respectively. The antioxidant effect of thyme oil on roasted beef steak over charcoal could potentially account for this improvement.

An investigation conducted by Hussein et al. (2023) indicated that the incorporation of lemon into the basic marinade resulted in a more efficient reduction of the carcinogenic PAHs in charcoal beef steak. The beef steak was tenderized by the marinade components, which included sugar, water, onions, turmeric, lemongrass, salt, garlic, coriander, and cinnamon. This made the protein simpler to digest and allowed it to cook more quickly. Additionally, the marinade prevented the loss of moisture and prevented the beef steak from drying out and becoming rough. Therefore, our findings were in agreement with the findings that were published by Farhadian et al. (2012) about the reduction of cooking time.

As indicated by Reinik et al., (2007), the variation in PAH levels seen in grilled meats may be attributed to the different amounts of fat that are present in raw meat. During the process of grilling, a greater amount of fat is discharged onto the hot coals. As a consequence of the high temperatures, PAHs are produced, which are subsequently transported to the surface of the grill by smoke. In marinades, oils are not only utilized to enhance moisture but also to avoid sticking and to assist in the distribution of flavors.

According to Farhadian et al. (2012), the addition of cooking oils prior to grilling results in different amounts of PAHs depending on the kind of oil it is. Therefore, substituting cooking oil with thyme oil, which has an antioxidant effect, may reduce carcinogenic polycyclic aromatic hydrocarbons. This is demonstrated by the findings of marinade 4 samples, in which the amount of polycyclic aromatic hydrocarbons found in the meat was greater than that found in the marinade5.

5. CONCULOSION

This study showed that five groups of marinades were applied to the beef steak before charcoal grill, and the fifth group, which used the traditional marinade with thyme oil and lemon juice, was the most effective marinade in reducing the content of carcinogenic substances BaP and PAH4 to the permissible limits as set by the European Commission.

It is recommended that charcoal-grilled beef steaks be marinated before cooked especially with antioxidant ingredient to reduce the risks of PAHs to human health.

CONFLICT OF INTEREST

The authors certify that they possess no conflicts of interest with the present data.

6. REFERENCES

1. Dutta,K., Shityakov S., Zhu, W.and Khalifa, I., 2022. High-risk meat and fish cooking methods of polycyclic aromatic hydrocarbons formation and its avoidance strategies. Food Control, 142(109253):253-261.
2. EC (European Commission), 2011. Commission regulation (EU) No 835/2011 of 19 August 2011, amending regulation (EC) No 1881/2006 as regards maximum levels for polycyclic aromatic hydrocarbons in foodstuffs. Off. J. Eur. Union 215. 4-8.
3. Edikou, K. U. S.; Osseyi, E. G.; Karo, S. D. and Dossou, J. J., 2018. Formation of Some Chemical Contaminants in Main Forms of Cooked Chicken Meat Products: A Review Journal of Poultry Science and Technology, 6,3: 31 38.
4. EFSA (European Food Safety Authority) (2008). Polycyclic aromatic hydrocarbons food - scientific opinion of the panel on contaminants in the food. EFSA Journal, 724: 1–114.
5. Hussein, E.; Edris, A. M. and Kirrella, G. A. K., 2023. Determination of Polycyclic Aromatic Hydrocarbon in Charcoal Beef Steak and Inhibitory Profile of Thyme Oil, Lactic Acid Bacteria and Marinating on their Existence. Pakistan. Journal Zoology, 56,6:1-8.
6. Farhadian, A.; Jinap, S.; Faridah, A. and Zaidul, I.S., 2012. Effects of marinating on the formation of polycyclic aromatic hydrocarbons (benzo [a] pyrene, benzo [b] fluoranthene and fluoranthene) in grilled beef meat. Food Control Journal, 28: 420-425.
7. Guide, E., 1998. The Fitness for Purpose of Analytical Methods, English Edition 1.0.
8. Janoszka, B., 2011. HPLC-fluorescence analysis of polycyclic aromatic hydrocarbons (PAHs) in pork meat and its gravy fried without additives and in the presence of onion and garlic. Food. Chemistry, 126: 1344–1353
9. Khorshid, M.; Souaya, E. R.; Hamzawy, A. H. and Mohammed, M. N., 2015. Method Followed by Solid Phase Extraction Method for Gas Chromatographic Mass Spectrometric Determination of Polycyclic Aromatic Hydrocarbons in Fish. International Journal of Analytical Chemistry, 2015:7-15.
10. Büyükkurt, K. Ö.; Dinçer E. A.; Çam, İ. B.; Candal, C. and Erbaş, M., 2020.The influence of cooking methods and some marinades on polycyclic aromatic hydrocarbon formation in beef meat. Polycyclic Aromatic Compounds. 40(2):195-205
11. Lee, S. Y.; Yim, D. G.; Kim, O. Y.; Kang, H. J.; Kim, H. S.; Jang, A. and Hur, S. J., 2020. Overview of the e ect of natural products on reduction of potential carcinogenic substances in meat products. Trends in Food Science and Technology, 99: 568–579.
12. Nor, H. K. A.; Jinap, S.; Sanny, M.; Ainaatul, A. I.; Sukor, R. and Jambari, N. N., 2022. Effects of Honey-Spices Marination on Polycyclic Aromatic Hydrocarbons and Heterocyclic Amines Formation in Gas-Grilled Beef Satay. Journal Polycyclic Aromatic Compounds, 42(4): 1620–1648.
13. Paw, M.; Begum, T.; Gogoi, R.; Pandey, S. K. and Lal, M., 2020. Chemical Composition of Citrus Limon L. Burmf Peel Essential Oil from North East India. Journal of Essential Oil Bearing Plants, 23,3: 337-344.
14. Reinik, M.; Tamme, T.; Roasto, M.; Juhkam, K.; Tenno, T. and Kiis, A., 2007. Polycyclic aromatic hydrocarbons (PAHs) in meat products and estimated PAH intake by children and the general population in Estonia. Food Additives & Contaminants Part A, 24:429-443.
15. Sampaio, G. R.; Guizellini, G. M.; da Silva, S. A.; de Almeida, A. P.; Pinaffi-Langley, A. C. C.; Rogero, M. M.; de Camargo, A. C. and Torres, E. A. F. S., 2021. Polycyclic Aromatic Hydrocarbons in Foods: Biological Effects, Legislation, Occurrence, Analytical Methods, and Strategies to Reduce Their Formation. International Journal of Molecular Sciences, 22,11:6010-6021.
16. Singh, L.; Varshney, J. G. and Agarwal, T., 2016. Polycyclic aromatic hydrocarbons formation and occurrence in processed food. Food Chemistry, 199:768–781.
17. Viegas, O.; Novo, P.; Pinto, E.; Pinho, O. and Ferreira, V.O., 2012. Effect of charcoal types and grilling conditions on formation of heterocyclic aromatic amines and polycyclic aromatic hydrocarbons (PAHs) in grilled muscle foods. Food and Chemical Toxicology. 50 ,6: 2128-2134.
18. Wang, C.; Xie, Y.; Wang, H.; Bai, Y.; Dai, C.; Li, C. and Zhou, G., 2019. Phenolic compounds in beer inhibit formation of polycyclic aromatic hydrocarbons from charcoal-grilled chicken wings. Food Chemistry, 294: 578–586

19. Yu, Y.; Cheng, Y.; Wang, C.; Huang, S.; Lei, Y.; Huang, M., and Zhang, X., 2023. Inhibitory effect of coriander (Coriandrum sativum L.) extract marinades on the formation of polycycli Marination c aromatic hydrocarbons in roasted duck wings. Food Science and Human Wellness, 12,4: 1128–1135.
20. Yunita, C. N.; Radiati, L. E. and Rosyidi, D., 2023. Effect of Lemon Marination on Polycyclic Aromatic Hydrocarbons (PAH) and Quality of Chicken Satay. Journal Penelitian Pendidikan IPA, 9.6: 4723–4730
21. Zengin, H. and Baysal, A.H., 2015. Antioxidant and antimicrobial activities of thyme and clove essential oils and application in minced beef. Journal of Food Processing and Preservation, 39: 1261–1271.