

**IMPACT OF COOKING METHODS ON THE QUALITY ATTRIBUTES
AND POLYCYCLIC AROMATIC HYDROCARBONS (PAHS) LEVELS
IN THE MUTTON SHEEK KEBAB**

BY

Dr. P.GOWTHAM B.V.Sc & A.H

I.D. NO: GVM/2018-018

Thesis submitted to the

SRI VENKATESWARA VETERINARY UNIVERSITY
in partial fulfilment of the requirements for the award of the degree of

MASTER OF VETERINARY SCIENCE
[LIVESTOCK PRODUCTS TECHNOLOGY]
(IN THE FACULTY OF VETERINARY SCIENCE)



DEPARTMENT OF LIVESTOCK PRODUCTS TECHNOLOGY
NTR COLLEGE OF VETERINARY SCIENCE, GANNAVARAM - 521102

SRI VENKATESWARA VETERINARY UNIVERSITY

TIRUPATI – 517 502. (A.P) INDIA

JUNE 2021.

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JUNE 2021.**

CERTIFICATE

Dr. P.GOWTHAM has satisfactorily prosecuted the course of research and that the thesis entitled “**IMPACT OF COOKING METHODS ON THE QUALITY ATTRIBUTES AND POLYCYCLIC AROMATIC HYDROCARBONS (PAHS) LEVELS IN THE MUTTON SHEEK KEBAB**” submitted is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination. I also certify that the thesis or part thereof has not been previously submitted by him for a degree of any University.

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Dr. B. ESWARA RAO

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CERTIFICATE

This is to certify that the thesis entitled “**IMPACT OF COOKING METHODS ON THE QUALITY ATTRIBUTES AND POLYCYCLIC AROMATIC HYDROCARBONS (PAHS) LEVELS IN THE MUTTON SHEEK KEBAB**” submitted in partial fulfilment of the requirements for the degree of “**MASTER OF VETERINARY SCIENCE**” of Sri Venkateswara Veterinary University, Tirupati, is a record of the bonafide research work carried out by **Dr. P. GOWTHAM** under our guidance and supervision. The subject of the thesis has been approved by the student’s advisory committee.

No part of the thesis has been submitted for any other degree or diploma. The published part has been fully acknowledged. All assistance and help received during the course of investigations have been duly acknowledged by the author of the thesis.

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P.GOWTHAM

DECLARATION

I **Dr. P. GOWTHAM** hereby declare that the thesis entitled “**IMPACT OF COOKING METHODS ON THE QUALITY ATTRIBUTES AND POLYCYCLIC AROMATIC HYDROCARBONS (PAHS) LEVELS IN THE MUTTON SHEEK KEBAB**” submitted to Sri Venkateswara Veterinary University, Tirupati for the degree of **MASTER OF VETERINARY SCIENCE** is the result of original research work done by me. I also declare that the materials contained in this thesis have not been published earlier.

DATE:

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ABSTRACT

In the present research work, an attempt was made to evaluate the impact of cooking methods viz., direct charcoal grilling (Control), moist cooking followed by charcoal grilling (Treatment 1) and hot air oven cooking followed by charcoal grilling (Treatment 2) on the quality attributes, levels of polycyclic aromatic compounds (PAH) and shelf -life of mutton sheek kebabs.

The T₁ formulation had significantly ($p<0.05$) higher cooking yield, moisture content, better texture profile of lower hardness and fracturability values, lower fat content and TBARS values, and higher scores for all organoleptic characteristics except flavor. In contrast, the control group received higher flavor scores. The overall mean pH of control was significantly ($p<0.05$) lower than treatment groups, and pH increased with the storage period.

T₁ had significantly ($p<0.05$) higher color parameters except for hue angle. Sensory and textural parameters decreased in all the groups towards the end of 15 days of refrigerated ($4\pm 1^{\circ}\text{C}$) storage. Control (C) had significantly ($p<0.05$) lower total plate count and yeast & mould count values than the treatment groups throughout the storage period. Psychrophiles were not detected in any of the groups during the 15 days of storage period.

The level of flouranthene, the only polycyclic aromatic hydrocarbon detected in all the groups, was below the maximum tolerance limits. Control group of kebabs had significantly ($p<0.05$) higher value of flouranthene, while the T₁ had the lowest value. The rest of the eight PAH compounds are below the level of quantification (LOQ).

Thus, the present study suggests moist cooking followed by charcoal grilling as the best and safe cooking method to prepare mutton sheek kebab.

List of Symbols and Abbreviations

PAH	Polycyclic Aromatic Hydrocarbon
Bap	Benzopyrene
HPLC	High Performance Liquid Chromatography
LC- MS	Liquid Chromatography with mass spectrometry
GC-MS	Gas Chromatography with mass spectrometry
⁰ C	Degree centigrade
mg	Milligram
μg	Microgam
g or gm	Gram
Pa	Pascal
LDPE	Low density polyethylene
Kg	Kilogram
MDA	Melonaldehyde
TBARS	Thiobarbituric Acid Reactive Substances
ml	Millilitre
Fig.	Figure
OD	Optical density
SE	Standard error
hrs	hours
mins	Minutes
%	Per cent
Cm and cms	Centimeter and centimeters
M and mts	Meter and meters
T(1)	Treatment 1
T(2)	Treatment 2
i.e.	That is
μ	Microns
N	Newtons
viz	That is
≤	Less than or equal to
≥	Greater than or equal to



Chapter – I

Introduction

CHAPTER I

INTRODUCTION

Muscle foods in addition to being a reliable source of nutrients especially protein, minerals, and vitamins, have high satiety index due its unique taste and flavor. For enhancement of taste and flavor and provide diversified products, meat is subject to several processing and cooking methods. Sectioning, grinding, restructuring, enrobing, curing and smoking, drying, fermentation, canning etc. are some of the important processing and cooking methods adopted to create wide range of value added meat products which suits to the requirements of diverse population. Liberalization, urbanization, fast growing economy and increasing disposable income are fuelling an ongoing trend toward consumption of value added animal products especially in metros, cities and smaller towns of developing countries. Even though, cultural patterns rather than income dominate meat consumption in India, the ready-to-eat meat sector is growing with consumer affluence.

Though processing and cooking of meat enhance the taste and flavor, inhibit microorganisms and improve digestibility, cooking at very high temperatures for longer duration may modify the chemical composition with a consequent changes in nutritional value and also formation of some undesirable compounds, which are found to be harmful to the consumers. Meats cooked until well/very well done and/or by high-temperature cooking techniques such as grilling, smoking or frying becomes source of mutagenic and carcinogenic compounds such as polycyclic aromatic hydrocarbons (PAHs) (Aaslyng *et al.*, 2013; Smith *et al.*, 2008). Polycyclic aromatic hydrocarbons (PAHs) are group of compounds generated by the combustion of organic matter under the intense and direct heating conditions. They are formed when the oil from meat grilled directly spills over an open fire, generating flames. PAHs may enhance the generation of DNA adducts that interfere with apoptosis provoking the risk of cancer

(Cross *et al.*, 2007). Some of the recent epidemiological studies have suggested that frequent consumption of well-done cooked meat, fish, and poultry may increase the risk of colorectal, pancreatic, breast and prostate cancer (IARC 1993; Sugimura, 2002).

Polycyclic aromatic hydrocarbons adhere to the meat surface that is produced with the flames (Farhadian *et al.*, 2012). The formation of PAHs is influenced by the type of marinating sauces, heat source, duration of cooking, fat content and the geometry of grilling (Alomirah *et al.*, 2011; Kao *et al.*, 2014; Singh *et al.*, 2016). The PAH levels are significantly affected by boiling than frying, roasting, and barbecuing (Onwukeme *et al.*, 2015). The charcoal with the wood chips used as fuel resulted in the higher benzo[*a*]pyrene (BaP) formation in barbecued food and cooking time may result in a moderate increase of PAHs (Rose *et al.*, 2015).

Ensuring the quality and safety of meat products from the production, processing, and delivery to the consumers is the responsibility of all the stakeholders involved in this entire value chain. There are certain possible interventions and innovations to prevent or reduce PAHs formation. Cooking at lower temperatures, employing techniques like baking, broiling, using slow cookers, sous-vide cooking, working with a microwave oven to cook the meat preceding to higher temperatures exposure, making use of the spices and herbs in the marinade are few ways to reduce the level of formation of PAH compounds (Lee *et al.*, 2016; El-Badry 2010; Farhadian *et al.*, 2011).

Grilled and smoked meat products represent a notable segment with everyday diet in India. Sheek Kebab, one of the celebrated grilled meat products, which are prepared with minced mutton, chevon, or chicken. Being a traditional product, it is prepared manually by hand molding, the meat mixture into oblong shapes around skewers and grilled in a charcoal oven. The manual preparation often results in rough and uneven the exterior surfaced kebab. Further, the long duration of charcoal broiling

at very high temperatures leads to case hardening of the outer surface of the product and higher cooking loss. The long duration of the charcoal broiling may also contribute to deposition of higher levels of harmful smoke substance on the product surface. Little information is available about the PAHs levels on the various meat products consumed in India (Muthukumar *et al.*, 2018). Hence, the development of innovative processes, which can enable automation in product making to a certain extent and yield products of consistent quality with low levels of harmful smoke substance and good economic return, assumes importance.

With this background, the present work has been designed to prepare mutton sheek kebabs by employing the traditional charcoal grilling procedure together with the modified methods of moist cooking and hot air oven cooking followed by charcoal grilling and assessing the quality attributes and polycyclic aromatic hydrocarbons (PAHs) levels in the products. In the modified innovative method, the kebab was prepared to employ the sausage stuffer, followed by the moist cooking and finally grilled in a charcoal oven. Similarly after cooking in hot air oven, kebab was grilled in a charcoal oven for shorter duration. This work was conducted at the Department of Livestock Products Technology, NTR College of Veterinary Science, Gannavaram in collaboration with the National Research Centre on Meat, Hyderabad with the following objectives.

1. Preparation of mutton sheek kebabs by employing traditional method i.e. Charcoal grilling and modified methods i.e. moist cooking and hot air oven method of cooking followed by shorter charcoal grilling.
2. To evaluate the organoleptic and physico-chemical properties of the products.
3. To evaluate the impact of cooking methods on the levels of PAH compounds in the sheek-kebabs.



Chapter – II

Review of Literature

CHAPTER II

REVIEW OF LITERATURE

2.1 TRADITIONAL MEAT PRODUCTS:

India is a nation of diverse ethnic groups with a large range of food habits and customs. A large variety of indigenous meat products are prepared all over India. The preparation process and taste vary from place to place. They are primarily native in origin and are referred to as traditional/indigenous meat items. Depends on the availability of raw materials in their geographical location, people prepared the traditional products with a unique taste and flavor.

Table no 1. Traditional meat products in India

Regional specific Ethnic / Traditional Meat Products
<i>Northern region:</i> Goshtaba, Tabak manss, Nate- Yakini
<i>Eastern region:</i> Meat curry, Meat dopiaza, Meat fry, Meat Briyani, Meat Pakora, Meat balls, Meat Kofta, Meat tikka, Meat Kababs
<i>Western region:</i> Vindaloo , Meat rolls, Shakudi
<i>Southern region:</i> Meat fry, Chicken curry, Biriyanies, Chettinad chicken, Dry salted meat.
<i>Himalayan region (North- Eastern Region):</i> Kargyong, Kheuri, Satchu, Suka Ko masu, Chilu, Chartayshya, Gemma, Arjia.
<i>Special Hyderabadi/ Deccani meat recipes:</i> Bheja, Pasinda, Chicken Kaccha Kofta, Mutton Kofta Haleem, Hyderabad dum briyani (chicken, Mutton), Paya, Nahari,
<i>Jammu and Kashmir:</i> Goshtaba, Nate Yakhni, Tabak manss, Rista, Aba gosh, Roganjosh, Dopyaza, Balti meat curry, Rogan josh, Kababs
<i>Punjab:</i> Tandoori meat products
<i>Uttar pradesh:</i> Dum briyani, Meat samosa, Meat pakora, Kebabs
<i>Tamil Nadu:</i> Meat curries, Chettinadu Chicken
<i>Andhra Pradesh:</i> Haleem, Gonguru mamsamu, Munagakaya mamsam, Boti Kababs

(Sudhakar *et al.*, 2010)

Some of the traditional products such as kebabs, tandoori items, koftas etc., brought to India by the Arabs, Persians, Mughals and Portuguese during the early invasion era. Over course of time, these items have become traditional and are widely

consumed across the nation.

2.2 VALUE ADDED MEAT PRODUCTS:

The proportion of value addition of meat in India is just 10-15 per cent, compared to 60 percent in developed nations (Sudhakar *et al.*, 2010). The value addition by altering the shape, texture, color of meat, incorporation of several additives and processing methods not only to provide variety of meat foods with rich flavor and unique taste to the consumers, but also improves the shelf life of perishables meat products. Further, fresh meat sale is comparatively less profitable than the value added meat products. Value added products also enable the profitable sale of secondary cuts, spent meat and tough meats, which fetch low revenue to the farmers.

Western and exotic meat products, namely sausages, salamis, hot dogs and frankfurters are also popular among Indian urban dwellers. Consumption habits of people regarding meat products with added value are also growing day by day. The value addition of meat can be accomplished through the creation of better comminuted, restructured, enrobed, marinated products and ready meals or snacks.

Based on the type of processing methods, the meat products are further classified into different types viz., chunked, restructured, emulsion based, cured and smoked products, enrobed/coated, dried, fermented and canned/retort pouched meat products.

2.3 COOKING/ PROCESSING METHODS:

Roasting: The sufficient browning of meat for better flavors and appearance is achieved by roasting over 163°C temperatures in a pan.

Broiling: It consists of direct radiant heat for cooking meat, such as the open fire of a gas flame, live coals or an electric oven. At a temperature of 176°C, broiling is carried out until the top side is just done. Broiling is a quicker way to cook meat over dry heat than roasting. For tender cuts that are at least 2.5 cm thick, broiling is applied.

Pan Broiling: Meat is positioned and heated in a cold girdle so that meat bakes slowly. Any fat accumulating in the pan is extracted so that rather than pan fry, the meat begins to pan broil.

Frying: Pan-frying and deep frying are two ways of frying. Too high temperatures lead to an undercooked inside and too low temperatures lead to a greasier product.

Moist heat: This technique is used for less tender cuts, resulting in tender meat product because of the conversion to gelatin from connective tissue.

Braising: Meat is cooked with or without the addition of water in this method; the meat is first carefully browned by broiling, pan broiling or frying on all sides. It is possible to incorporate tomatoes and fruit juices as liquids.

Curing: This refers to the changes in the meat that affects preservation, taste, color, and tenderness by the addition of curing ingredients. Principle ingredients used are sodium chloride (preservative and adds flavour), sodium nitrate or sodium nitrite (unique flavor-acts as a preservative-anti-botulinum activity-fix the red color), sugar (stabilize color and add flavor) and spices (flavor).

Smoking of Meat: Meat curing and smoking are closely interrelated and are frequently done together. Smoking has a preservative effect on meat, similar to curing. There is a development of aroma, color, oxidation protection and protective skin on emulsion type sausages.

Al-Bachir *et al.* (2010) studied the effect of gamma irradiation (2, 4 and 6 KGy) on chicken kabab and reported that gamma irradiation of dose 4, 6 KGy increased the shelf-life of chicken kebab by decreasing the microbial load and the major constituents of chicken kebab (moisture, protein and fats) remain undisturbed by irradiation.

Pandey *et al.* (2014) reported that deep fat fried shami kebab at 120°C for 5 minutes and 170°C for 2 minutes and grilling at 180 and 140°C and for 3 and 5 minutes

respectively revealed that taste, color, aroma, texture, and overall acceptability of the product were significantly at 170°C deep fat frying conditions than grilling.

The effect of cooking techniques on the nutritional value of foal meat was investigated by Dominguez *et al.* (2015) and reported that the PUFA content was slightly higher for grilled and roasted samples than for microwave samples with a lower fat content relative to fried samples.

Onwukeme *et al.* (2015) reported that the boiling had a greater impact on the levels of PAHs than other cooking techniques i.e. frying, roasting and barbecuing.

Albuquerque *et al.* (2016) recorded that when compared to deep-fat frying, the cholesterol content of baked chicken breaded nugget was two times higher.

The steam cooked nuggets (97.16 percent) had significantly higher cooking yield compared to the oven and microwave cooked chicken nuggets (92.83 percent and 88.57 percent) (Pathera *et al.* 2017).

Banerjee *et al.* (2018) prepared the chicken sheek kebab using different methods of cooking viz. grilling, smoking and roasting and reported that cooking yield and texture parameters were higher in grilled kebabs, whereas the smoked kebabs are preferred for the sensory parameters due to its characteristic color and flavor.

2.4 SHEEK KEBAB:

Kebabs are ready to eat, one of the famous convenience meat products that are sold in worldwide food outlets. A common traditional meat product in the Indian subcontinent and gulf nations, *Sheek Kebab*, originally referred to as *Shish Kebab*, literally represents a small chunk of meat roasted on a skewer. In fact, shish means a skewer or sword and kebab mean roasting (Banerjee *et al.*, 2018)

Ground or minced meat is mixed and pressed onto a skewer along with the condiments and seasonings, forming a uniform layer of the mix around the skewer, which can be cooked in an open heat source such as a grill or even broiled in a

microwave oven. The meat used mostly for kebab is traditionally lamb, but now the chicken kebabs often become increasingly common over the years, with increased consumer acceptance for chicken meat and as per regional availability, taste, and cost-effectiveness.

Sensory analysis of doner kebab prepared by using turkey meat was performed by Kayaardi *et al.* (2006) stated that turkey doner kebab would be appropriate for panel consumption after 12 days of storage and off-flavors significantly reduced kebab acceptance after that time.

Modi *et al.* (2007) optimized the level of binders (refined wheat flour, starch, and milk powder) in dehydrated chicken kebab mix and reported that the addition of 4.5% starch, 9.5% refined wheat flour and 2% milk powder was optimal. They also reported that the fried chicken kebabs prepared from mix were acceptable even after stored at ambient temperature of 27.2°C for up to 6 months.

Bhat *et al.* (2010) has effectively used black bean (10, 15 and 20%) as an extender in the chicken sheek kababs prepared from the meat of spent hens. They reported that the kebab extended with 10% black bean was optimum based on the physicochemical and sensory parameters.

Successful incorporation of green gram (10, 15 and 20%) in sheek kebab prepared from the spent chicken meat by microwave cooking was carried by Bhat and Pathak (2011) and reported that the 15% inclusion of green gram paste resulted in better sensory and physicochemical properties.

Bhat *et al.* (2013) formulated the chicken sheek kebabs with low-cost vegetable origin extenders (15% cowpea, 15% green gram, and 10% black bean) and prepared chicken sheek kebabs by microwave method of cooking. They reported that the product's sensory attributes and acceptability are similar to kebab prepared without legumes.

Ali and Kihna (2018) reported that the addition of chicken breast meat at different concentrations (10 percent, 15 percent, 20 percent, 25 percent, 30 percent) to the standardized kebab mix containing (20 percent beef, 5 percent sheep fat tail, and 75 percent sheep flank meat) resulted in decreased cholesterol and fat content, but increased protein content in kebab.

2.5 QUALITY ATTRIBUTES OF SHEEK KEBAB: (COOKING YIELD, PROXIMATE COMPOSITION, COLOR, TEXTURE AND SENSORY ATTRIBUTES)

2.5.1 COOKING YIELD

Murphy *et al.* (2001) reported that the moisture loss of oven cooked chicken patties was increased three to five times with increasing the final product temperature from 50 to 80°C. They also reported that significant differences were obtained on product yield between the patties cooked in oven at a humidity of 50 and 95% (10% higher yield in 95% humidity).

Barbanti and Pasquini (2005) studied the influence of hot air cooking on chicken breast and reported that the cooking losses at 130°C and 170°C were 13.52 and 17.26% for the duration of 4 minutes while higher cooking losses of 23.36 and 34.03% were noticed when the meat was cooked for the duration of 12 minutes at 130°C and 170°C.

Deep fat frying of circular discs (40×13 mm) of chicken kebab mix resulted in product yield in the range 85.8–86.2% and shrinkage in diameter in the range 7.8–8.3% with the storage period having a marginal effect on springiness, cohesiveness, and firmness (Modi *et al.*, 2007)

Pork patties cooked in a steam-convection oven at 190°C for 17 minutes resulted in a cooking loss of 14.4% (Danowska-oziewicz 2009).

Alfaia *et al.* (2010) investigated the effect of grilling, boiling and microwaving on the nutritional quality of the beef and reported that the cooking loss increased with

the increase in internal temperature of meat and cooking time. They also reported that the cooking loss for beef samples are in descending order for microwaving (42.71%), boiling (39.91%) and grilling (32.64%).

Bhat *et al.* (2010) conducted a study regarding the usage of the black bean as an extender in chicken sheek kebabs prepared by microwave cooking. They reported that the cooking yield raise significantly with increase in the extender level. The elevated value of cooking yield (87.90%) was observed for the kebabs extended with 20% black bean compared to the kebab prepared with 10% black bean (85.69%).

Nisar *et al.* (2010) studied the effect of cooking methods of buffalo meat patties with 5% added fat and reported that the hot air method had a lower cooking yield (85.72%) compared to the microwaving (87.80%) and pressure cooking (86.39%).

Bhat and Pathak (2011) studied the effect of green gram (*Vigna radiata*) on the quality parameters of chicken Sheek kebab prepared by microwave method. They found a cooking yield (%) of 80.48 ± 0.68 , 83.33 ± 0.79 , 85.74 ± 0.62 and 87.90 ± 0.77 for the sheek kebab prepared with 0, 10, 15 and 20% of green gram, respectively.

Sous-vide cooking of pork at 80°C revealed significantly higher weight losses than others cooked at 60°C. The cooking yield was significant affected by the cooking duration. The samples that cooked for 5 h showed lesser cooking weight losses (16.7%) than the other cooked for 12 h (20.2%) (Del Pulgar *et al.*, 2012).

Cooking losses of the lamb loins cooked by sous-vide method were affected by the cooking temperature and an increase in temperature leads to higher weight losses (20.77, 30.67, 35.22% cooking loss at 60, 70, 80°C, respectively). Similarly cooking time also significantly affected the cooking losses (20.77, 24.72, 28.78% cooking loss for 6, 12, 24 hours of cooking) (Roldan *et al.*, 2013).

Naveena *et al.* (2014) reported that the moist cooked chicken nuggets resulted in higher cooking yield (89.38%) compared to the dry cooked patties (85.98) and deep-fat fried croquettes (77.42%).

The cooking yield of hot air oven dried goat meat cubes at 60°C for 18 h (37.66%) was higher than the microwave dried goat meat cubes at 540 W for 20 minutes at (34.81%). (Nayar *et al.*, 2014)

Pandey *et al.* (2014) reported that the percent cooking yield of shami kebab was better in grilled (93.05%) as compared to the deep fat frying (91.23%) method of cooking. They also stated that the variation in the cooking yield showed no significant difference ($P < 0.05$) at deep fat frying (120°C) and grilling (180°C).

The influence of different cooking methods of chicken breasts was studied by Chumngoen *et al.* (2016) and reported that chicken breasts placed-in-bag and cooked in a water bath (19.43%) had higher cooking loss than the direct hot water cooking (18.56%).

Malav *et al.* (2016) reported a cooking yield of 90.60% for mutton patties cooked in a hot air oven at 180°C for 30 minutes.

Oz *et al.* (2017) reported the cooking loss of 35.45% in beef steaks processed by hot air oven at 200°C for 9 minutes compared to microwaving method of cooking for 4.5 minutes (38.62%).

The cooking method had a major influence on cooking loss. Smoked kebabs registered a slightly higher cooking loss (17.14 percent), while grilled kebabs (12.52 percent) showed the lowest value (Banerjee *et al.*, 2018).

Chumngoen *et al.* (2018) investigated the effects of moist (water-cooking) and dry-heat (oven-cooking) on chicken breast and reported that moist cooked chicken breast had lower cooking loss (24.43%) than oven cooking (30.31%).

Kiran *et al.* (2018) reported that the moist cooking of chicken meat incorporated with rice as binder (nuggets) resulted in higher cooking yield (74.62 %) compared to the dry cooked patties (72.94 %) and deep-fat fried croquettes (67.46%).

Głuchowski *et al.* (2019) reported that the roasted fish fillets resulted in lower cooking yield (83.8%) than the sous vide cooking (93.7%) and steam cooking (88.4%).

The cooking loss of the chicken breast fillets processed by the sous vide method of cooking was at 9.33–14.09 %, and it was lower than the conventional boiling cooking (Karpinska-Tymoszczyk *et al.*, 2020).

2.5.2 PROXIMATE COMPOSITION

The chemical composition of the doner kebabs marketed in the Tekirdag market was determined by Kayisoglu *et al.* (2003) and reported the values of moisture (61.28–51.67%), protein (19.04–26.45%), fat (16.23–14.03%), ash (1.94– 3.01%), and salt (1.28–2.08%) for the raw and cooked kebabs. Chicken kebabs had significantly higher protein (26.45%) compared to beef doners (25.01%).

Gonulalan *et al.* (2004) reported significant ($P < 0.05$) variation between the sausage (sucuk) doners and traditional doners in terms of protein and ash content. The elevated level of the ash content in the grilled and raw sausage doner (4.27% and 4.08%) was due to the ingredients used for curing which were not regularly used in the traditional doners.

The chicken doner kebab sold in Turkish restaurants showed 47.27% (38.47–52.494%) moisture, 0.99% (0.22–3.12) salt, 21.01% (14.36–28.91) fat and 26.17% (20.36–31.44) protein content (Vazgecer *et al.*, 2004).

Chicken breast cooked in a hot air oven at 130°C for 4 minutes had 70.33% moisture, 27.24% protein, fat 1.04%, and 1.37 % ash. (Barbanti and Pasquini 2005)

Modi *et al.* (2007) prepared the chicken kebab mix (dehydrated) with optimized quantities of spices (garlic, onion, pepper, cumin, green chilli, coriander leaves powder)

and binders (starch, wheat flour, and milk powder) and reported that the kebab mix had 5.4% moisture, 8.9% fat, 51.5% protein, 6.7% ash and 2.9% NaCl. The water activity value of the prepared mix gradually increased from 0.31 ± 0.013 to 0.42 ± 0.016 during 6 months of storage at ambient temperature of 27.2°C due to the concomitant rise in moisture value from 5.4 to 6.2 %.

Nemati *et al.* (2008) carried out the chemical analysis of the Bonab kebabs marketed in the Tabriz market and reported moisture (66.23– 56.94%), protein (14.3– 19.59%), fat (14.05–18.58%), salt (1.27–1.96%) and ash (2.06– 2.72%) values for the raw and cooked sample.

The influence of various cooking methods (roasting, frying, grilling, and microwave) on lipid oxidation of meat from Iberian pigs was studied by Broncano *et al.* (2009) and they indicated that cooking causes a remarkable reduction of moisture, barring the fried meat. They also added that cooking produced no changes in the lipid content of the meat because the frying process was superficial; accordingly the penetration of oil was low. Moisture and fat content of grilled, fried, microwave and roasted pork samples were 54.12, 43.32, 49.23, 46.88 and 40.49% and 25.47, 29.0, 27.69, 28.77 and 31.81%, respectively.

Pork patties cooked in a steam-convection oven at 190°C for 17 minutes resulted in 68.39 % moisture, 13.66% protein and 6.74% fat (Danowska-oziewicz 2009).

Al-Bachir *et al.* (2010) studied the impact of gamma irradiation on chemical properties of the chicken kebab and estimated the values of moisture ($67.43 \pm 1.41\%$), crude protein ($18.97 \pm 0.04\%$), crude fat ($8.57 \pm 0.95\%$), and ash content ($2.36 \pm 0.03\%$). There were significant difference in the ash contents between the non-irradiated and the samples irradiated with 6 kGy. They added that irradiation does not have an impact on the moisture, protein and fats of chicken kebab.

Kebabs prepared from the spent hen's meat using black bean as an extender

reported that all the proximate parameters except ash content were decreased significantly with the raise in extenders levels for both raw and cooked kebabs. Control sheek kebab with zero addition of black bean showed the value of 63.25% moisture, 15.62% protein, 14.10% fat and 2.19% ash while the sheek kebab with 20% addition of black gram had value of 61.36% moisture, 13.51% protein, 12.61% Fat and 2.86% Ash (Bhat *et al.* 2010)

Nisar *et al.* (2010) reported that the buffalo meat patties with 5% added fat prepared by the hot air method had lower moisture content (65.97%) compared to microwaving (67.11%) and pressure cooking (66.45%).

Bhat and Pathak (2011) reported a gradual decrease in the protein and moisture percent in chicken Sheek kebab due to the addition of green gram paste which contains lower moisture (56.65%) and protein content (16.97%) than control chicken kebab (58.92% moisture and 18.83% protein). Fat content showed a significant decline (12.73%) at the twenty percent extension level of green gram paste due to the dilution effect of the paste which is particularly low in fat content. Ash percent showed a significant increase (2.83%) due to the declining trend of moisture with an increasing level of green gram paste. Control sheek kebab with zero addition of green gram paste showed the value of 14.71% fat and 2.38% ash.

Physico-chemical, structural and textural characteristics of pork cheeks (sous-vide cooked) were affected by vacuum packing, cooking time, and temperature. Samples cooked at 80°C resulted in significantly lower moisture content (66.5%) than cooked at 60 °C (60.8%) (Del Pulgar *et al.*, 2012).

The increased cooking temperatures in lamb loins (sous-vide cooked) led to lower moisture content (66.42, 66.38 and 64.18% at 60, 70 and 80°C, respectively). However, the cooking time had no significant effect upon the moisture content of the sous-vide cooked lamb loins (Roldan *et al.*, 2013).

Naveena *et al.* (2014) reported that the moist cooked chicken nuggets had higher moisture content (67.82%) compared to the dry cooked chicken patties of moisture (66.00%), while the protein (22.66 %), fat (3.17%) and ash content (3.07%) of dry cooked chicken patties were higher than the moist cooked nuggets (22.27 %, 3.12%, 2.87%).

Nayar *et al.* (2014) reported that the hot air oven dried goat meat cubes at 60°C for 18 h had higher moisture content (8.83%) and fat (8.49%) compared to the microwave dried goat meat cubes at 540 W for 20 minutes of moisture (8.10%) and fat (5.45). They also reported protein values of 49.68 and 49.86% and ash values of 8.71 and 8.48% for hot air oven dried and microwave dried goat meat cubes, respectively.

Pandey *et al.* (2014) reported that the variation in the moisture content of shami kebab samples was not significantly affected by deep fat frying at 170°C for 2 minutes (51.88%) and 120°C for 5 minutes (53.15%), except grilling at 140°C conditions (54.67%).

Choi *et al.* (2016) reported that charcoal grilled chicken steaks had higher ash content (1.60%) than the boiled chicken steaks (1.42%).

Malav *et al.* (2016) reported that the mutton patties cooked in a hot air oven at 180°C for 30 minutes yielded products with 60.24% moisture, 17.42% Protein, 15.14 % Fat, and 2.52 % Ash.

Ali and Kihna (2018) revealed that there were significant variations in the moisture values between raw and grilled Kebab. The average moisture content was higher in raw kebab (56.85%) than grilled Kebab (48.27%). They further added that the mean fat and protein content was significantly ($p < 0.05$) increased with the addition of chicken breast meat (10%) for raw (19.15 and 21.66%) and grilled samples (26.01 and 23.86%), but there was no significant variation ($p > 0.05$) in ash content compared to the control samples with no chicken breast.

The cooking process had major impact on the moisture percentage of the chicken sheek kebabs. Smoked kebabs reported a slightly higher cooking loss (17.14 percent), while grilled kebabs (12.52 percent) showed the lowest value. No significant variation in terms of protein and fat content was reported between kebabs cooked using different methods (grilling, roasting, and smoking) (Banerjee *et al.* 2018)

Kiran *et al.* (2018) reported that the moist cooked chicken nuggets had higher moisture content (71.05%) than the dry cooked patties (67.31%). They also added that the protein (16.74%), fat (4.71%) and ash content (2.02%) of dry cooked chicken patties were higher than the moist cooked nuggets (17.06 %, 3.32%, 1.93%).

Karpinska-Tymoszczyk *et al.* (2020) reported that the moisture content of the raw chicken breast meat was 76.13%, which found to be reduced significantly ($p < 0.05$) after cooking. The highest (74.94%) and the lowest (73.04%) moisture content was recorded in sous vide cooked chicken breast fillets at 61°C and 58°C, respectively. The fat content of raw chicken breast fillets was 1.27%, reported to be increased significantly in fillets processed at 58°C (24.33) and 61°C (22.85%). The protein content of raw meat was 20.99%, but increased to 21.79–24.33%, owing to the changes in protein concentration resulting from water loss.

2. 5. 3 COLOR

Garcia-Segovia *et al.* (2007) assessed the influence of cooking method on the color and mechanical properties in the beef muscle and suggested that the cooked steaks were always lighter (higher L*) and more yellow (higher b*), while a* (red color) decreased with increasing temperature and cooking time. They reported that the L* values were significantly higher ($p < 0.05$) for the cook-vide system than the samples cooked at atmospheric or sous-vide conditions. The a* values and b* values were significantly higher ($p < 0.05$) for beef steaks cooked at sous-vide system than they cooked at the cook-vide or atmospheric pressure conditions.

The color reflectance values of the chicken kebab mix measured on the Hunter color difference meter was investigated by Modi *et al.* (2007) revealed that the Hunter L* (lightness) and b (yellowness) values marginally (P = 0.05) increased from 51.8 ± 2.14 to 53.4 ± 2.42 and 19.1 ± 0.49 to 19.5 ± 0.16, respectively, whereas the Hunter a* (redness) values increased gradually (P= 0.05) from 0.30 ± 0.244 to 1.29 ± 0.379 during the 6 months storage period. They also added that even after the changes in the color of the kebab mix product were acceptable for six months of storage period at 27 ± 2°C, despite the marked (P =0.05) changes in the Hunter a* (redness) values.

Christensen *et al.* (2011) studied the effect of sustained heat treatment from 48°C to 63°C on the color of *Longissimus dorsi* (LD) muscle of pigs. They reported that lightness (L* values) increased with rising temperatures while increasing time did not impact L* values and a* values decreased in LD from sows (P<0.05) at 63°C.

Nikmaram *et al.* (2011) reported that the braising (moist cooking) of veal muscle had higher lightness (56.6) and redness (6.89) values than the roasted samples of lightness (53.54) and redness values (4.67).

Del Pulgar *et al.* (2012) reported that the sous-vide method of cooked pork samples at 60°C showed higher L*, lower Hue angle (H*) values, and intense red color (greater a* values) than those samples cooked at 80 °C (P<0.0001). They also insisted that all the color parameters were affected by temperature.

The effect of various temperature-time combinations on the physicochemical features of the sous-vide method of cooked lamb loins was studied by Roldan *et al.* (2013) and reported that the color parameters were highly affected by cooking temperature (P=0.021 for lightness L*, P<0.001 for (redness a* and yellowness b*). Samples cooked at 60 °C showed slightly higher L* values than those cooked at 80°C or 70°C while increasing the cooking time and cooking temperature produced greater yellowness values b* (P<0.001 for both).

Nayar *et al.* (2014) reported that the hot air oven dried goat meat cubes at 60°C for 18 h had higher hue angle (63.93) compared to the microwave dried goat meat cubes at 540 W for 20 minutes (43.09). They also reported that the chroma and redness values of hot air oven dried and microwave dried goat meat cubes were (2.60 and 2.99) and (1.32 and 2.27), respectively.

Pandey *et al.* (2014) studied the effect of processing conditions on shami kebab and reported that the lightness (L*) values in all temperature conditions varied significantly ($p < 0.05$), though the grilled temperature conditions were found to be less compared to deep fat frying (DFF). At 140°C grilled condition, both L* (45.56) and yellowness (b*) (24.52) were obtained highest and with the lowest redness (a*) value (9.11). Redness (a*) in the product does not show a significant difference ($p < 0.05$) at 120°C DFF and 180°C grilled conditions. Yellowness (b*) in product differed significantly at all the processing conditions. They also added that the kebabs fried and grilled at lower temperature conditions found lighter than the high temperature processed ones.

Choi *et al.* (2016) reported that the boiled chicken steak had higher lightness values (93.43) than the oven (90.90) and grilled chicken steaks (82.21).

The mutton patties cooked in a hot air oven at 180°C for 30 min and packed in a vacuum package revealed the Redness a* value of 10.64, Yellowness (b* value) of 17.54, Hue angle of 58.92, and chroma value of 20.63 (Malav *et al.*, 2016).

Oz *et al.* (2017) reported the color values (lightness L*, redness a*, and Yellowness b*) of beef steaks processed by hot air method of cooking and microwaving were 41.41 and 41.74, 13.87 and 14.17, and 7.23 and 6.23, respectively.

The meat surface color was brighter (greater L* value - 38.13 ± 0.35) and high yellow (greater b* value - 33.94 ± 1.86) for the smoked kebabs compared to the samples

from the grilled (30.21 ± 2.07 and 26.82 ± 2.14) and roasted cooking treatment (26.29 ± 1.78 and 31.12 ± 2.11) (Banerjee *et al.*, 2018).

Chumngoen *et al.* (2018) reported that a higher lightness values was noticed in moist cooked chicken meat (81.22) than the oven cooked chicken (78.75).

Głuchowski *et al.* (2019) reported that the sous vide cooked salmon fish had higher values of redness (22.04), yellowness (28.34) and chroma (35.9) when compared to the roasted samples of redness (13.81), yellowness (28.34) and chroma (23.85).

Karpinska-Tymoszczyk *et al.* (2020) revealed no significant differences in lightness (L^*) (83.00-83.75) color values of the chicken breast fillets cooked by sous vide method. However, a significant ($p < 0.05$) decrease in redness was noted when the heating time was longer by 1 hour (3.29 and 2.44 at 55°C for 260 and 320 mins). The value of yellowness (b^*) on the surface of meat cooked for the shorter time was significantly greater in samples cooked at 58°C (13.13) than in meat processed at lower (55°C - 10.42) and higher (61°C - 11.42) temperature.

2. 5. 4 TEXTURE PROFILE ANALYSIS:

Modi *et al.* (2007) analyzed the texture quality parameters of chicken kebab and reported that large variations (183.2– 200.2 N) in firmness value due to variations in the raw material qualities or the degree of frying, etc. However, the cohesiveness and springiness values fluctuated in the limited range of 0.22–0.23 and 3.9–4.2 mm respectively, during the storage of kebab mix for 6 months. The differences in all the texture quality parameters during the storage interval were not significant ($P = 0.05$).

Del Pulgar *et al.* (2012) reported that the sous-vide method of cooking pork samples at 60°C treatment for 12 or 5 hours produced no significant effect in terms of springiness, hardness, cohesiveness or chewiness, and adhesiveness while the lower values in springiness (0.69-0.61 cm), hardness (16.3-9.5 N), cohesiveness (0.52-0.38), and chewiness (5.8-2.3N/cm) in the samples cooked at 80°C from 5 to 12 hours was

highly distinctive.

Roldan *et al.* (2013) studied the effect of different time-temperature combinations on the textural attributes of lamb loins cooked by sous-vide method and reported that the cohesiveness, chewiness, and springiness were significantly affected by temperature ($P < 0.001$, $P = 0.013$, and $P = 0.001$, and respectively) and cooking time ($P < 0.001$ for springiness & cohesiveness, $P = 0.005$ for chewiness, $P = 0.003$ for hardness, and $P = 0.004$ for gumminess & Shear force). Adhesiveness was not affected by both the cooking time and temperature.

Nayar *et al.* (2014) reported that the microwave dried goat meat cubes at 540 W for 20 minutes had higher Hardness (1567 N/cm²), Gumminess (444.51 N/cm²), and chewiness (349.19 N/cm) compared to the hot air oven dried goat meat cubes at 60°C for 18 h (Hardness - 1215.88 N/cm², Gumminess - 258.44 N/cm², and Chewiness - 119.58 N/cm).

Deep fat frying of shami kebab had enhanced higher moisture loss in the kebab affecting the viscoelastic behavior and the effect was prominent in the springiness (18.54 mm) and cohesiveness values (0.112). Gumminess behavior of the kebab samples resulted in higher energy requirement in the complete disintegration and retained a significant difference ($P < 0.05$) in both processing techniques (deep fat frying and grilling) at all temperature conditions (0.276 and 0.347 N/cm²) (Pandey *et al.*, 2014).

The mutton patties cooked in a hot air oven at 180°C for 30 min had 27.21 N/cm² Hardness, -18.43 N/g Adhesiveness, 0.37 cm/mm Springiness, 0.34 Cohesiveness, 9.29 N/cm², Gumminess, and 3.48 N/cm chewiness (Malav *et al.*, 2016)

The cooking techniques had a significant effect on the texture profile of chicken kebabs lower hardness (45.13±1.4 N/cm²), chewiness (31.97±0.51 N/cm), gumminess (19.53±0.53 N/cm²), cohesiveness (0.43±0.025), and springiness (1.63±0.05 cm) values were found in grilled kebabs, while the highest scores were reported in smoked samples

(hardness - 48.70 ± 0.74 N/cm²; chewiness - 55.12 ± 3.41 N/cm; gumminess - 32.80 ± 1.91 N/cm²; cohesiveness - 0.67 ± 0.03 and springiness - 1.68 ± 0.02 cm) (Banerjee *et al.*, 2018).

Wang *et al.* (2019) stated that the superheated steam cooking (120°C for 5 min) of pork meat had higher textural characteristics when compared to the hot air cooking (120°C for 5 min). The values for hardness, springiness and chewiness of superheated steam cooked and hot air cooked pork meat were 73.03 and 66.86 N/cm², 70.4 and 56.05 percent and 3.37 and 2.24 kg/mm, respectively.

Karpinska-Tymoszczyk *et al.* (2020) examined the effect of cooking at six combinations of the temperature and time duration (55°C (260 and 320 minutes); 58°C (140 and 200 minutes) and 61°C (90 and 150 minutes) on the texture profiles of chicken breasts. Cooking time and temperature had no significant ($P > 0.05$) influence on the hardness or gumminess. Similarly, no significant ($P > 0.05$) differences in chewiness were noted in chicken meat samples cooked for a shorter time at each tested temperature. A significant ($P < 0.05$) improvement in chewiness was observed only at a temperature of 58°C.

Suleman *et al.* (2020) compared the textural quality of lamb patties prepared by charcoal grilling, infrared grilling and superheated steam roasting. They reported that the hardness in the charcoal grilled and superheated steam roasting lamb patties increased with increasing the cooking time ($P < 0.05$) while trend was reversed in the infrared grilled cooking on patties, as the hardness was decreased at all cooking intervals. The superheated steam roasting had lower hardness values (8076.59gm) than the charcoal grilled samples (8974.35gm).

2. 5. 5 SENSORY EVALUATION

Gonulalan *et al.* (2004) reported that the sausage doners made from the sucuk dough had significantly greater scores at day 0 for flavor (9.50), texture (9.0), color

(9.00) and overall appearance (8.83).

The flavor and acceptability scores of the turkey doner kebab decreased (from day 12 onwards) with increasing TBA and peroxide values. These results showed that there was a negative relationship between the sensory properties and peroxide and malonaldehyde values. Correlations between the general acceptability and other sensory parameters showed that the least effective parameter was the color of the samples (0.47) while the chewiness of the samples was highly correlated (0.86) (Kayaardi *et al.*, 2006).

Modi *et al.* (2007) evaluated the sensory characteristics of the dehydrated chicken kebab mix and reported a descending trends of the mean sensory scores for all the quality characteristics from 0 day products (freshly prepared) (7.7–8.1) to 6 months stored (7.4–7.8) kebab mix. However, the chicken kebab had sensory acceptable score (7.4) when prepared from kebab mix even after stored for 6 months at $27 \pm 2^\circ \text{C}$.

Danowska-oziewicz (2009) reported that the intensity of meat flavor was the highest in combi oven-cooked patties (hot air + 30% steam) (8.7), whereas lower scores were given to hot air cyclojet-cooked patties and pan-fried patties (7.4 and 7.1, respectively). He also reported that the highest scores for overall acceptability were given to the combi oven-cooked patties (8.0) while the lowest value for the microwave oven cooked patties (5.0).

The influence of gamma irradiation on the sensory parameters of chicken kebab was studied by Al-Bachir *et al.* (2010) and found that the sensorial parameters (taste, texture, color, and flavor) of the chicken kebab were not influenced by the irradiation treatment (6 and 4 kGy). They also indicated that the irradiation of chicken kebab at these doses (6 and 4 kGy) does not significantly affect ($P < 0.05$) the sensory characteristics of chicken kebab and can be suitable for prolonging the shelf-life of the chicken kebab.

Bhat *et al.* (2010) conducted a study about the usage of black bean (extender) on

the quality parameters of the chicken sheek kebabs by microwave cooking and reported that the sensory characteristics viz., flavor, overall acceptability, juiciness, and texture decreased significantly ($P < 0.05$) with raise in the range of extender level. They also indicated that 10 percent black bean paste was the optimum for the formulation of extended chicken sheek kebabs.

Higher sensory scores (texture - 6.29, appearance and color - 6.60, flavor - 6.55, juiciness - 6.17, and overall acceptability - 6.29) were obtained for the hot air oven cooked beef patties than other cooking methods i.e microwaving and pressure cooking (Nisar *et al.*, 2010).

The effect of green gram (*Vigna radiata*) on the quality parameters of microwave cooked chicken Sheek kebab was studied by Bhat and Pathak (2011) and they observed a significant ($P < 0.05$) influence on overall acceptability, flavor, and texture and a steady decline in the appearance and juiciness scores of kebabs due to the extension with green gram paste.

Bhat *et al.* (2013) analyzed the quality attributes of the microwave cooked chicken sheek kebabs extended with various non-meat ingredients (legumes- cowpea, green gram and black gram) and found that the products had similar acceptability and sensory attributes as that of control sheek kebabs made without legume extenders.

Hot air oven dried goat meat cubes had higher sensory scores of overall acceptability (6.80), flavor (6.88), and texture (6.75) compared to the microwave dried cubes (values of 6.49, 6.47, and 6.50 respectively) (Nayar *et al.*, 2014).

Pandey *et al.* (2014) investigated the effect of the processing conditions on the textural properties of the shami kebab and reported that the color, texture, aroma, taste, and overall acceptability of the product were significantly different ($P < 0.05$) at 170°C deep fat fried conditions than a grilled kebab. Overall acceptability was highest for the deep fat frying conditions (8.01 at 170°C).

Malav *et al.* (2016) evaluated the sensory characteristics of the vacuum packed mutton patties stored in refrigerated storage and reported that the sensory scores were in the decreasing order for day 0 and 60 viz., general appearance (7.18 and 6.43), flavor (7.20 and 6.33), texture (7.18 and 6.47), binding (7.16 and 6.44), juiciness (7.18 and 6.44), and overall acceptability (7.24 and 6.36).

Pathera *et al.* (2016) stated that the steam cooked chicken nuggets showed significantly higher values (7.22) for the overall acceptability than an oven (6.89) and the microwave (6.75) cooked nuggets during the refrigerated storage.

Soladoye *et al.* (2017) studied the influence of different cooking methods (microwave or frying pan) in the bacon and reported that the frying pan method of cooking increased the crispiness and saltiness of bacon ($P < 0.05$) whereas the other sensory parameters were not affected ($P > 0.05$) by the cooking methods and storage interval. They also reported that the microwave cooked bacon had a lower impact on the generation of carcinogenic compounds with minimum impact on sensory characteristics.

The changes in the sensory characteristics of Kebab (20% beef, 5% sheep fat tail, and 75% meat of sheep flank) when admixed with the chicken breast meat was determined by Ali and Kihna (2018). They found significant differences ($P < 0.01$) in the color and tenderness characteristics among the products. Comparatively lower values of the sensory attributes were obtained (3.5 out of 5) for the kebab prepared with the higher levels of the chicken breast meat (30%) whereas 5% chicken breast inclusion yielded maximum value (4.8 compared to control of 4.4).

The development of typical sensory properties in chicken sheek kebabs during smoking may result in a greater ($P < 0.05$) overall acceptability of smoked kebabs over grilling and roasting methods (Banerjee *et al.*, 2018). They reported significant differences ($P < 0.05$) in appearance, texture, flavor, overall acceptability, and juiciness of kebabs cooked by different methods (grilling, roasting, and smoking).

Wang *et al.* (2019) stated that the superheated steam cooked (140°C for 10 min) pork meat had higher values for sensory attributes viz. mouth coating (6.24), color (7.33), appearance (6.88), and lower flavor values (3.42) when compared to the hot air cooked pork (140°C for 10 min) (mouth coating (5.94), color (6.04), appearance (4.89), and flavor values (4.21)).

Karpinska-Tymoszczyk *et al.* (2020) reported that the chicken breast fillets had most desirable taste, juiciness, and aroma when cooked at the lowest temperature (55°C) and the least desirable taste and aroma when cooked at 58°C. They also found that the longer cooking at the lowest (55°C) and the highest (61°C) temperature had reduced the tenderness of the chicken breast fillets assessed organoleptically and the heat treatment at 61°C for 150 min deteriorated the juiciness of the meat.

2. 6 POLYCYCLIC AROMATIC HYDROCARBONS:

2. 6. 1 PAH AND IT'S FORMATION:

Polycyclic aromatic hydrocarbons (PAHs) are a broad group of organic compounds consisting of two, three, or more fused aromatic rings. They were formed exclusively of carbon and hydrogen without hetero atoms. PAHs are produced by incomplete combustion or by pyrolysis of organic matter. Consequently, the natural along with anthropogenic sources in the environment were numerous. PAHs generally occurred in complex nature that comprised hundreds of compounds. The composition of these mixtures varied with the process of generation (EFSA 2008).

Polycyclic aromatic hydrocarbons are single or fused aromatic rings of polycyclic aromatic hydrocarbons with a pair of carbon atoms shared in their molecular rings. PAHs with six fused aromatic rings are referred to as 'tiny PAHs' and those with more than six aromatic rings are referred to as 'big PAHs'. The predominant scientific work on PAHs was carried on the small PAHs as a result of the presence of various small PAHs samples (Abdel-Shafy *et al.*, 2016).

Alomirah *et al.* (2011) explained about the three possible mechanisms for the generation of PAH compounds. The first mechanism is the pyrolysis of organic matter such as fat, carbohydrates, and protein with temperature exceeding 200°C, and PAH formation was favoured around the range of 500 to 900°C. The second mechanism is with the direct contact of lipids that dripped by having direct contact over the flame at intense heat. The third mechanism is the PAHs production by incomplete combustion of charcoal onto the surface of the food.

2.6.2 PUBLIC HEALTH SIGNIFICANCE

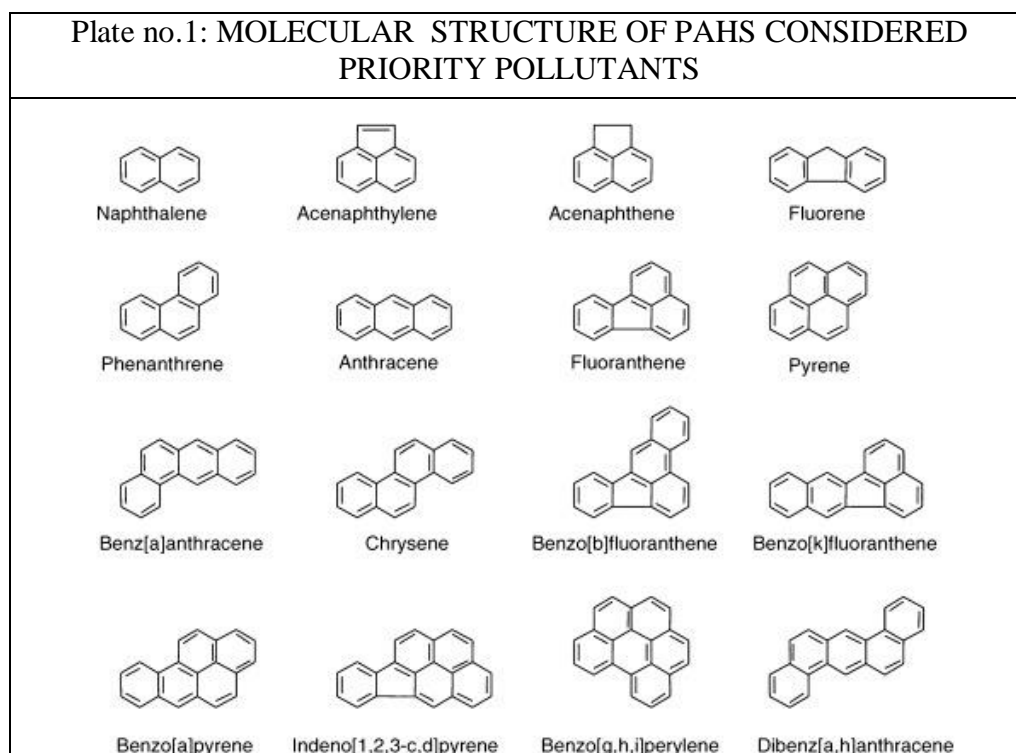
Cross *et al.* (2007) carried out a study on the intake of red and processed meat having an interrelation with the risk of cancer and reported that the PAH intake from cooked meat had been found to elevate the risks of colorectal neoplasia, squamous cell oesophageal cancer, and cancers of the lung, pancreas, and prostate.

Yoon *et al.* (2007) estimated that PAHs had Lifetime Average Daily Intake (LADI) of about 3.22×10^{-3} µg/kg/day for having carcinogenic effects. It showed higher incidences in the people, under 20 year's younger group relative to the older groups. The cancer potency of benzo(a)pyrene for estimating the risk of cancer (7.3 mg/kg/day) was 2.3×10^{-5} , which was seen to be equivalent to an eruption of tumor in the upper gastrointestinal tract of two per hundred thousand persons.

The dietary exposure of PAH in Catalonia, Spain was investigated by Marti-Cid *et al.* (2008) and they found that the towering individual PAH levels equivalent to phenanthrene (29.66 µg/kg), naphthalene (25.87 µg/kg), and fluoranthene (13.66 µg/kg), while the shortest levels were benzo[a]pyrene (1.28 µg/kg), benzo[k]fluoranthene (1.31 µg/kg) and indeno[1,2,3-c,d]pyrene (1.44 µg/kg). Significantly the inflated levels of total PAHs were recognized in meat and its products (25.56 µg/kg), oils and fats (23.48 µg/kg), and cereals (20.44 µg/kg). The dietary intake of the sum of PAHs was higher (12.0 µg / day) for an average male adult (70-kg body weight) than that observed

in the year 2000 (8.4 μ g / day).

Martorell *et al.* (2010) estimated the intake of PAHs by the population of Catalonia, Spain and reported that the elevated levels of total PAH were recognized in meat and its products (38.99 μ g/kg), accompanied by oils and fats (18.75 μ g/kg), and dairy products (7.57 μ g/kg). The highest PAH intake was contributed by the meat and its products (4.75 μ g/day).



(Source: Shukla *et al.* 2014).

Nisha *et al.* (2015) studied the effects of polycyclic aromatic hydrocarbons on health, its generation by diversified procedures including smoking, grilling, cooking, and reduction of PAH in the meat that was processed. Additionally, reported that the smoking and grilling of meat led to the high concentration of PAH rather than other cooking methods.

Chaber and cunningham (2016) analyzed the polycyclic aromatic hydrocarbons (15 PAH) levels in the smoked dried fish and found that levels were three to more than five times higher, compared to the maximum permitted level in the European

Community which implicated the high incidence of primary liver and stomach cancer reported in Nigeria.

Zhu *et al.* (2019) stated that the average carcinogenic risk of PAH exposure in the southwest China region exceeded the appropriate level of incremental cancer risk for all participants (10^{-6}), and 10.7% of urban residents and 2.1% of rural residents exceeded the extreme risk (10^{-4}). In food samples, the average concentration of PAH (16 compounds) was $12.04 \pm 8.52 \text{ ng/g}$. The average lifetime daily exposure dose of benzo(a)pyrene equivalent was $6.07 \times 10^{-6} \text{ mg/kg/day}$. This indicated that dietary PAHs might pose risk for cancer in the southwest China region.

Table No.2: List of PAH compounds and their properties

PAH compound	Abbreviation	Molecular weight (g/mol)	Chemical formula
Benzo[a]pyrene	BaP	252	$\text{C}_{20}\text{H}_{12}$
Benz[a]anthracene	BaA	228	$\text{C}_{18}\text{H}_{12}$
Benzo[b]fluoranthene	BbF	252	$\text{C}_{20}\text{H}_{12}$
Benzo[j]fluoranthene	BjF	252	$\text{C}_{20}\text{H}_{12}$
Benzo[k]fluoranthene	BkF	252	$\text{C}_{20}\text{H}_{12}$
Benzo[ghi]perylene	BghiP	276	$\text{C}_{22}\text{H}_{12}$
Chrysene	Ch	228	$\text{C}_{18}\text{H}_{12}$
Cyclopenta[cd]pyrene	CPP	226	$\text{C}_{18}\text{H}_{10}$
Dibenz[a,h]anthracene	DBahA	278	$\text{C}_{22}\text{H}_{14}$
Dibenzo[a,e]pyrene	DBaeP	302	$\text{C}_{24}\text{H}_{14}$
Dibenzo[a,h]pyrene	DBahP	302	$\text{C}_{24}\text{H}_{14}$
Dibenzo[a,i]pyrene	DBaiP	302	$\text{C}_{24}\text{H}_{14}$
Dibenzo[a,l]pyrene	DBalP	302	$\text{C}_{24}\text{H}_{14}$
Indeno[1,2,3-cd]pyrene	IP	276	$\text{C}_{22}\text{H}_{12}$
5-methylchrysene	5MeCh	242	$\text{C}_{19}\text{H}_{14}$
Benzo[c]fluorine	BcF	216	$\text{C}_{17}\text{H}_{12}$

(Source: Ledesma *et al.*, 2016).

2. 6. 3 SOURCES OF CONTAMINATION OF PAH COMPOUNDS:

PAHs were continuously emitted into the atmosphere from combustion-related

emission sources such as motor vehicles and residential/commercial fossil fuel, which could lead to significant atmospheric deposition. These compounds were eliminated from the atmosphere through dry particle deposition, gaseous air deposition, wet deposition, and atmospheric transformation. However, the residence time in the atmosphere could be dependent on the distribution of the compounds between the particle and gaseous phases within the atmosphere (Gigliotti *et al.*, 2005).

For the general population, the major routes of exposure were through the inhaled air and food, while in smokers, the contributions from smoking and food may be of a similar magnitude. Food could be contaminated by environmental PAHs existing in air, soil, or water, by food processing methods occurring in industries (e.g. smoking processes heating, and drying) and at home by the preparation of food (e.g. roasting grilling processes) (EFSA 2008).

The amount of PAH in smoked meat was influenced by the nature of wood. The lowest concentration for individual and total PAH was seen in the samples smoked with apple-tree and alder. The highest value was obtained with the samples smoked with spruce. The difference in the content of total PAH (from 47.94 till 470.91 $\mu\text{g}/\text{kg}$) and BaP (from 6.04 till 35.07 $\mu\text{g}/\text{kg}$) indicated that the preference of wood for smoking could be the critical parameter to be controlled to diminish the contamination of food products (Stumpe-Viksna *et al.*, 2008).

PAHs are volatile compounds with a high propensity to adsorb on organic matters and low solubility in water. Once released, PAHs could spread in all compartments of environments like air, water, sediments, soil, and plants, resulting in contamination of terrestrial and aquatic species. So, high levels of PAHs could be encountered in the human food supply. Raw products (animals and agricultural products) contamination occurred during growth but, also during transport, resulting from the deposit of aromatic hydrocarbons from the exhausts of engines on foodstuff

and storage (Paris *et al.*, 2018).

2. 6. 4 OCCURRENCE OF PAH COMPOUNDS IN ENVIRONMENT:

Moret *et al.* (2005) reported that the polycyclic aromatic hydrocarbons are seen in vegetable oils from canned foods and the oil from preserved vegetables generally presented PAH in high concentration, compared with oil from preserved fish. The elevated PAH concentration in the oil from a grilled mushroom sample was 11.3 ppb of BaP and 47.1 ppb of total PAH (heavy). The contamination level was high in an olive oil sample from canned tuna with a value of 1.9 ppb of BaP and 11.3 ppb of total PAH (heavy). The high PAH concentrations from vegetable oil products would be due to the contribution from contaminants in the vegetables.

Rey-Salgueiro *et al.* (2008) evaluated the direct (flame-toasting, coal-grilling or gas oven-toasting) and indirect toasting (electric oven-toasting) of sandwich bread for PAHs formation and interpreted that indirect heating resulted in no PAH generation, while up to 350 µg/kg of PAHs (total) were detected in toasted bread by wood flame (direct method). They also added that the levels of Benzo[a]pyrene ranged from 0 to 0.23µg/kg.

Roberto *et al.* (2009) determined the soil-borne PAH level in El Paso, Texas and reported that light PAHs (such as fluoranthene, pyrene, phenanthrene, and naphthalene) ranged from 80-100 percent in the analyzed samples. Heavy PAHs (such as pyrene, benzo[a]pyrene and, benz[a]anthracene) were less frequently detected. The concentration of the PAH compound was found with fluoranthene (538.5 g /kg dry soil), benzo[a]pyrene (364.6 g /kg), pyrene (397.1 g kg⁻¹), and chrysene (346.6 g /kg). The mean PAHs (16 PAHs) concentration in the soil ranged from 0.1 to 2225.5g/kg.

The concentration of PAHs in toasted bread was determined by Al-Rashdan *et al.* (2010) and concluded that the PAHs (total) were varied in the levels of 1.06–44.24 µg/kg and 3.08–278.66 µg/kg for heavy PAHs (H-PAHs) and light PAHs (L-PAHs),

respectively. B[a]P were ranged from 2.83 to 16.54 µg/kg.

Tujeta *et al.* (2011) carried out the quantitative assessment of PAHs with leafy and underground vegetables in Panipat district, Haryana. The result showed that leafy vegetables were contaminated more with PAHs than underground vegetables and the lower-molecular weight PAHs compounds were greater in its concentration than higher molecular weight PAHs in all types of vegetables.

The PAH levels in Egyptian vegetables (vegetables & fruits) were evaluated by Abou-Arab *et al.* (2014) and recorded the concentration of total PAHs in spinach (8,977 µg/kg), potatoes (6,196µg/kg), guava (2,334µg/kg), and apples (2,867µg/kg). The amount of total and carcinogenic PAHs in spinach was reduced by 4.23 and 5.73%, 54.02 and 66.03%, and 74.36 and 79.62% by washing with tap water 1%, and 2% acetic acid solution, respectively. They also found that the concentrations of total and carcinogenic PAHs were reduced with peeling (93.85 and 93.18%) and boiling of peeled potatoes (98.39 and 97.19%).

Yu *et al.* (2019) investigated wild marine fish species from the northern South China Sea for PAHs and found that the concentration of total 16 PAHs ranged from 199 to 606ng/g dry weight. PAHs were substantially higher in fish found in the Pearl River Delta than in fish from the Yachen gas fields. Planktivorous fish had significantly higher levels of PAHs relative to carnivorous and omnivorous fish. He also suggested that the pollution of PAH originated from petroleum inputs. The risk assessments of cancer and non-cancer conditions concluded that there is a minimal (2.5) risk associated with the intake of PAHs via fish consumption.

2. 6. 5 OCCURRENCE OF PAH COMPOUNDS IN MEAT PRODUCTS:

Panalaks (1976) reported presence of PAH compounds (4-7 rings) in approximately 70% of the commercial samples of charcoal broiled meat in Canada. Total and individual PAHs ranged from 0-164 ppb and 0-60 ppb respectively in the

charcoal broiled samples.

S. No.	Foodstuffs	PAH	Maximum levels (µg/kg)
1.	Dietary foods for special medical purposes intended specifically for infants	Benzo(a)pyrene	1.0
		Sum of PAH4	1.0
2.	Oils and fats (excluding cocoa butter and coconut oil) intended for direct human consumption or used as an ingredient in food	Benzo(a)pyrene	2.0
		Sum of PAH4	10.0
3.	Smoked meat and smoked meat products	Benzo(a)pyrene	5.0 until 31.8.2014 2.0 as from 1.9.2014
		Sum of PAH4	30.0 as from 1.9.2012 until 31.8.2014 12.0 as from 1.9.2014
4.	Muscle meat of smoked fish and smoked fishery products.	Benzo(a)pyrene	5.0 until 31.8.2014 2.0 as from 1.9.2014
		Sum of PAH4	30.0 as from 1.9.2012 until 31.8.2014 12.0 as from 1.9.2014
5.	Smoked sprats and canned smoked sprats; bivalve mollusks (fresh, chilled or frozen); heat treated meat and heat treated meat products sold to the final consumer	Benzo(a)pyrene	5.0
		Sum of PAH4	30.0
6.	Bivalve molluscs (smoked)	Benzo(a)pyrene	6.0
		Sum of PAH4	35.0
7.	Processed cereal-based foods and baby foods for infants and young children	Benzo(a)pyrene	1.0
		Sum of PAH4	1.0
8.	Infant formulae and follow-on formulae, including infant milk and follow-on milk	Benzo(a)pyrene	1.0
		Sum of PAH4	1.0
PAH4 = benzo(a)pyrene, benz(a)anthracene, benzo(b)fluoranthene and chrysene			

Source: Commission Regulation (EC) No 1881/2006

Table No 3. Maximum levels in regulation for polycyclic aromatic hydrocarbons in certain foodstuffs

The smoked meat products had PAH concentration (total) in the range of

2.6µg/kg (cooked ham), 29.8µg/kg (grilled pork chops), 9.3µg/kg (smoked shrimp), and 86.6µg/kg (smoked salmon). The concentrations of individual and total PAHs in the smoked chicken products ranged from non-detectable in the sliced breast (liquid smoke flavouring added) to 6.1µg/kg for Phenanthrene in barbecued wings, 4-5µg/kg in the sliced breast processed with liquid smoke and 22.4µg/kg in barbecued wings. The concentration of carcinogenic PAHs (Benzo[a]anthracene, Benzo [b] fluoranthene, Benzo[a]pyrene, Dibenzo [fl,A]anthracene, and Indeno [1,2,3-c, d] pyrene) ranged from non-detectable (0) for sliced breast (liquid smoke flavouring) to 5.5 µg/kg in barbecued wings (Gomaa *et al.*, 1993).

Zabik *et al.* (1996) reported total PAHs concentrations (ng/g wet weight) of 1.52 and 6.34 in raw trout samples and 154.4 and 270.9 in smoked trout samples harvested from Lakes Michigan and Superior, respectively.

Kazerouni *et al.* (2000) estimated the daily benzo[a]pyrene (BaP) intake and found that the levels of BaP were high in grilled/barbecued steaks (4.86ng/g), hamburgers (1.52ng/g) and grilled/barbecued chicken with skin (4.57ng/g). They also stated that grilled / barbecued meat in broiled or pan-fried meat samples had lower BaP concentrations and that it was little for non-meat products. But, the level of BaP levels was 0.5ng/g in certain cereals and greens (e.g. kale, collard greens). Significantly grilled/barbecued meat and the bread/cereal/grain contributed 21% and 29%, respectively to the average daily intake of BaP in Washington.

The smoked meat products had a concentration of PAH in the range from 24µg/kg (salami) to 64µg/kg (bacon), while fish products, the level ranged from 22µg/kg in an electric oven smoked mackerel to 1387µg/kg in smoked herring by direct smoking (Duedahl-Olesen *et al.*, 2006). The benzo[a]pyrene concentration from all samples were below the maximum permissible level of 5µg/kg for fish and meat (smoked) formulated by the European Commission. They also reported that intake of

benzo[a]pyrene from these smoked products is about 2 to 4 ng/person/day in Denmark.

Reinik *et al.* (2007) reported that 1.6 times higher PAH concentration was seen in disposable grilling unit compared to the traditional wood-burning grill. The total PAH concentrations with the highest value were found to be 16µg/kg in smoked meat and ham, 19 µg/kg in smoked sausage, and 6.5µg/kg in smoked chicken. Prevalent cooking methods like grilling and smoking in Estonia leads to the impact of overall PAH intake on meat products.

Akpambanga *et al.* (2009) reported that grilled or smoked meat and fish products prepared using a traditional system with wood fire were heavily contaminated with benzo[a]pyrene (BaP). The levels ranged from 2.4 to 31.2 µg/kg wet weight. Markedly, lower levels of contamination were found in the smoked or grilled samples by using a charcoal fire (0.7 to 2.8µg/kg wet weight).

The PAHs concentration in the popular grilled meat dishes from Malaysian ranged from 3.51 to 106ng/g (Farhadian *et al.* 2010). They further reported that the charcoal-grilled dishes showed the higher level of PAHs (132ng/g) and oven grilled dishes had a lower level (3.51ng/g).

Wretling *et al.* (2010) reported that in nine samples out of thirty-eight samples of smoked meat and six samples out of thirty nine samples of smoked fish showed high levels of BaP varied from 6.6 to 36.9 mg/kg and 8.4 to 14.4 mg/kg, respectively. The levels of BaP exceeded the European Commission (Regulation (EC) No 208/2005) of 5.0 µg/kg maximum level for meat and fish (smoked). However, the meat and fish samples, smoked by using the indirect technique, providing smoke from an external smoke generator, were known to have BaP levels, below the limit of quantification (0.3 mg/kg).

Alomirah *et al.* (2011) examined the grilled and smoked foods in Kuwait for the PAHs concentrations and reported that the meat tikka had higher mean concentrations

of benzo[a]pyrene (BaP) (2.48 mg /kg).

Chung *et al.* (2011) reported a concentration of 3.0µg/kg and 0.15 µg/kg of benzo(a)pyrene in charcoal-grilled beef and pork samples of Korea, respectively.

Silva *et al.* (2011) found the total PAHs level of 856.2 µg/kg, 780.8 µg/kg, 120.8 µg/kg and 37.9 µg/kg and fluoranthene levels of 329.2 µg/kg, 17.9 µg/kg, 82.9 µg/kg, 68.2 µg/kg in fish samples smoked by for the sawdust smoked, firewood smoked, charcoal smoked and oven-dried respectively. However, the oven-dried and smoked fishes, there was no detected level of benzo (a) pyrene.

Cho and shin (2012) examined sixty-nine smoked meat products and thirty-one smoked fish products from the Korean market for assessing the concentration of benzo[a]pyrene and reported that the levels of BaP was in the range of nil to 2.87µg/kg. The highest BaP content was found in chicken (2.87±0.49 µg/kg).

The charcoal roasted goat meat by direct heat had an average PAH content (total) of 4.77µg/kg, compared with an average of 2.13µg/kg in raw meat sample, with the benzo(a)pyrene level being 8.84% of the PAH (total). Roasted beef possessed the greatest individual PAH of Dibenz (a, h) anthracene (5.03µg/kg) and the highest total PAHs value (17.88µg/kg). (Onyango *et al.*, 2012).

The beef and salmon grilled with coconut shell charcoal had an amount of 1.36 ng/g and 0.50 ng/g of polycyclic aromatic hydrocarbons whereas the salmon and beef grilled with wood and charcoal had an amount of 4.72ng/g and 0.41ng/g, respectively (Viegas *et al.*, 2012).

Jahurul *et al.* (2013) estimated levels of individual PAHs in meat and fish products in Malaysia and found that the compound in elevated level was observed to be fluoranthene (219.74 ng/g), whereas the lowest level was benzo[a]pyrene (24.33 ng/g). Among all food products, grilled beef satay showed the highest total PAHs (66.28 ng/g). They additionally reported that the mean dietary intake of three PAHs

(benzo[a]pyrene, benzo[b]fluoranthene, and fluoranthene) in Malaysia was 297.58ng/day.

The sum of concentrations of PAHs from the smoked fish fillets was 606.1 µg/kg for the cold-smoked sample and 180.69 µg/kg for hot smoked (El-Lahamy *et al.* 2016).

Muthukumar *et al.* (2018) examined the levels of polycyclic aromatic hydrocarbons in tandoori chicken, chicken tikka, and smoked chicken marketed in the Hyderabad using GC-MS and found that none of the products indicated the presence of carcinogenic PAH compounds viz., chrysene, benzo[a]anthracene, benzo[b]fluoranthene, and benzo[a]pyrene and the levels of carcinogenic PAH compounds were under the limit of quantitation (0.01mg/kg).

Samiee *et al.* (2020) reported that the total PAH was in the levels of 8.08 to 29.55µg/kg in sausage and 10.18 to 29.85µg/kg in burger samples in Iran. The levels of acenaphthylene (13.4µg/kg) and anthracene (14.12µg/kg) were greater than the maximum level (2µg/kg) specified by European Commission (Regulation (EC) No 208/2005). The meat products containing 90 % of meat showed an elevated level of total PAHs (19.34µg / kg), while fried meat products showed the highest mean level of PAHs (23.31µg/kg). A positive and important association was noted between the product brand, cooking method and the PAH level.

2. 6. 6 IMPACT OF COOKING METHODS AND OTHER FACTORS ON THE LEVELS OF PAH COMPOUNDS:

Karl and Leinemann (1996) interpreted the impact of smoking type kilns on the PAH levels and reported that the mean benzo[a]pyrene concentration from commercial kilns (smoking) with external smoke generation was 0.1µg/kg (wet weight) while for smoked fish samples, using traditional kilns was 1.2µg/kg.

Chen and Lin (1997) reported that the charcoal grilling of duck samples with skin contained the highest amount of total PAHs, followed by charcoal grilling without

skin, roasting, smoking, steaming and liquid smoke flavoring (LSF). The total PAHs concentrations increased from 18.7 to 52.6 ppb during smoking for 0.5 to 3.0 hours.

The impact of the smoking process on the concentration of polycyclic aromatic hydrocarbons in meat products was analyzed by Ciecierska and Obiedzinski (2007) and reported that the traditional method of smoking resulted greater contamination of PAHs than the industrial process. They also found that the level of contamination was lower with interior rather than exterior for the same products.

Alomirah *et al.* (2011) looked over the grilled and smoked foods for the concentrations of polycyclic aromatic hydrocarbons and indicated a strong influence of kind of heat source, duration, the geometry of grilling, fat content, and the use of marinating sauces on PAHs formation. The highest concentrations of mean benzo[a]pyrene (BaP) (2.48 mg/kg) were found in meat tikka.

Chung *et al.* (2011) worked on finding the consequence of roasting and grilling on the concentration of PAH in pork and beef and found high levels of benzo(a)pyrene (3.0 µg/kg) in the charcoal grilled pork, while the mean benzo(a)pyrene of grilled beef (charcoal) samples are 0.15 µg/kg. They also reported that the PAH levels are dependent on the different types of heating sources and cooking techniques.

The traditional processing of dry/fermented sausage caused a greater ($p < 0.01$) total PAH levels (3237.10 µg/kg DM) comparatively with the industrial/modified procedures (1702.85 µg/kg DM). In both final products and raw materials, the light PAH compounds constitute about 99.0% of the total PAHs. This is mostly accounted by those compounds having three rings (acenaphthene–16.9%; anthracene–3.9%; phenanthrene–19.5% and fluorene–27.1%) or two (naphthalene–27.5%). The PAH compounds in the products placed in bars nearer to the smoking source/heating speeded up their transfer from the outer/surface portion to the inner part of the product (Roseiro *et al.*, 2011).

The fish samples smoked with charcoal gave the minimum level of total PAHs,

followed by the firewood method, while the sawdust usage gave the maximum level of PAHs. The level of PAHs in fish varieties that were smoked also positively correlated along with the fat content (Silva *et al.* 2011).

Gomes *et al.* (2013) examined the impact of added casing type (hog and collagen), fat (20% and 40%) and smoking procedures (both indirect and direct exposure) on the EPA priority PAHs (16) in dry sausages (fermented) produced by traditional processing and found significantly greater ($p < 0.05$) contamination levels in hog casing treated samples combining direct smoking procedures and high-fat content.

Compared to the use of beech wood chips, the use of poplar and hickory wood chips resulted in a 35-40% decrease in BaP, 15 + 1 EU-PAH, and PAH4 content for Frankfurt-type sausages, and a 36-55% decrease in mini-salamis (Hitzel *et al.*, 2013).

The generation of PAHs was associated with grilling time, temperature, fat content and the highly carcinogenic benzo[a]pyrene were reported in chicken heart grilled at 100°C (26 min), chicken breast at 84°C (30 min), chicken drumstick at 74°C (20 min), lamb steak at 88 °C (12 min), duck drumstick at 85°C (40 min), and with its level accounting to 2.4, 1.3, 4.0, 5.8 and 3.1 ng/g, respectively (Kao *et al.*, 2014).

The impact of cooking methods viz., boiling, frying, roasting and barbequing on the concentration of polycyclic aromatic hydrocarbons of chicken meat was analyzed by Onwukeme *et al.* (2015) and found that the levels of the PAHs were remarkably affected by boiling than other cooking techniques i.e. frying, roasting and barbecuing.

Rose *et al.* (2015) determined the effects of different cooking methods (roasting, frying, barbecuing, grilling, and toasting) in homemade foods for the formation of PAHs and found that the charcoal with wood chips used as fuel resulted in the higher benzo[a]pyrene (BaP) formation in barbecued food. They also reported that the beef burgers had the median and maximum BaP concentrations of 12 and 29µg/kg, respectively.

Lee *et al.* (2016) worked on finding the effects of grilling conditions on the level of PAHs (chrysene, benzo[a]anthracene, benzo[a]pyrene, and benzo[b]fluoranthene) and concluded that the smoke resulting from incomplete fat combustion dripping onto the fire was the key factor contributing to the generation of PAHs in grilling. Via treatments which removed meat drippings, brought substantial reductions in the amount of the PAHs.

The potential strategies to avoid and minimize contamination of PAHs in processed food products were examined by Singh *et al.* (2016) and found that the type of method of cooking, viz. roasting, grilling, barbecue, heating, baking, smoking, drying, ohmic-infrared cooking, etc used to have a pronounced impact on PAHs contamination. They also reported that the level of PAH depended on various factors like distance from fuel used, heat source, cooking durations, level of processing and methods.

2.6.7 METHODS/ PROCESS TO REDUCE PAH COMPOUNDS IN MEAT PRODUCTS:

The polycyclic aromatic hydrocarbons in grilled meat preparations were assessed by Fretheim *et al.* (1983) and concluded that the PAH contamination of grilled foods was mainly due to melted fat pyrolysis. Practical steps that minimize fat pyrolysis include limiting grilling to lean meat, avoiding unnecessary heating, or keeping melted fat away from the heat source. They also stated that electric grilling could be preferable to reduce PAH contamination.

The use of liquid smoke would produce smoked food with a decreased polynuclear aromatic hydrocarbon content relative to foods smoked on wood or charcoal (Lijinsky, 1991)

Gomaa *et al.* (1993) reported that the smoked meat products (turkey, chicken, pork, and beef) processed with liquid smoke flavourings had a lesser level of total PAHs

and total carcinogenic PAHs compared to those processed with natural wood smoke.

Ye *et al.* (1995) checked out the ability of *Sphingomonas paucimobilis*, (a soil bacterium) strain EPA 505 capable of make use of fluoranthene as the primary source of energy and carbon for growth, to metabolize a variety of polynuclear aromatic hydrocarbons (high molecular weight). They reported that after 16 hours of incubation with PAH (10 ppm), *S. paucimobilis* suspension of resting cell (1mg wet cells/mL) grown on fluoranthene degraded 80.0, 31.5, 72.9, 33.3, 7.8, and 12.5% of pyrene, chrysene, benz[a]anthracene, benzo[a]pyrene, dibenz[a,h]anthracene, and benzo[b]fluoranthene, respectively.

The PAH concentration is reduced to more than 50% by the use of low-density polyethylene (LDPE) layer on the smoked meat from their initial levels. PAHs (e.g., Benzo[a]anthracene BAA, Benzo[b]fluoranthene BBF, and Benzo[a]pyrene BAP) levels were reported in roasted duck after and before packaging with LDPE, in which the concentration levels of BBF (from 3.7 to 1.7 $\mu\text{g}/\text{kg}$), BAA (from 143 to 130 $\mu\text{g}/\text{kg}$), and BAP (from 3.5 to 4.9 $\mu\text{g}/\text{kg}$), were reduced after a storage interval of 24 hours (Chen and Chen, 2005).

The effect of food additives in household cooking methods on polycyclic aromatic hydrocarbons production in chicken meat was carried out by El-Badry (2010). He reported that the chicken meat treated before to thermal processing and cooking with food additives treatment (spices mixture, garlic paste, garlic paste, spices mixture, and marinades) cause a reduction in the formation of the PAHs during cooking. He also suggested avoiding direct gas grilling of meat because it induces PAHs formation and causing health hazards.

Farhadian *et al.* (2010) analyzed the Malaysian popular grilled-meat dishes (nine types) for toxic PAHs, i.e. fluoranthene, benzo(a)pyrene, and benzo(b)fluoranthene using HPLC-FD and reported that the maximum concentration of PAHs was reported in

charcoal-grilled followed by the oven grilled and flame-gas grilled dishes. Beef satay had a maximum concentration of PAH in charcoal-grilled dishes. PAHs concentrations were comparatively low when the gas-flame source was kept vertical, but no significant difference ($p < 0.5$) in PAH concentrations between chicken and beef grilled by a vertical source.

Farhadian *et al.* (2011) determined the effects on the PAH (Benzo(a)pyrene, Benzo(b)Fluoranthene, and Fluoranthene) contents by using two types of treatments (Steam cooking and Charcoal grilling), wrapping (aluminum and banana leaf) and preheating (steam and microwave) of the meat samples before charcoal grilling. The study showed that wrapping and preheating treatments on the meat samples significantly affect the levels of PAHs for charcoal-grilled meat. The carcinogenic PAHs (BbFln and BaP) were not detected (100% reduction) in the samples after aluminum wrapping or steam and microwave preheating. Important reductions in the fluoranthene level, of up to 81% and 46% in chicken and beef samples by banana leaf wrapping and steam preheating, respectively.

The smoked fish samples produced by traditional smoking, in which the food was exposed directly to smoke from a log fire, had the concentrations of PAH ranged from 0.6 to 8.4 $\mu\text{g}/\text{kg}$. Consequently, the samples of fish smoked by an external smoke generator, carried BaP under the level of quantification, i.e., 0.3 $\mu\text{g}/\text{kg}$ (Mihalca *et al.*, 2011).

Farhadian *et al.* (2012) investigated the impact of marination on the formation of polycyclic aromatic hydrocarbons (benzo[a]pyrene, fluoranthene and benzo[b]fluoranthene) in grilled beef meat and observed significant minimization (70%) of PAH in grilled beef samples treated with the marinade (acidic containing lemon juice 1.2%).

The salmon fish grilled with coconut shell charcoal had significantly lesser

amounts of PAHs than salmon grilled with ordinary wood charcoal. Continuous use of the same charcoal for barbecuing resulted in higher PAH formation due to the combustion of fat that falls during the grilling period over the charcoal (Viegas *et al.*, 2012).

The mean PAH (total) levels in the smoked fishes ranged from 250.59 to 1376.09 $\mu\text{g}/\text{kg}$ in tuna, sardine, cigar minnows, and smoke-cured mackerel smoked with sugarcane bagasse, acacia, and mangrove for 2 to 8 hours. Mutagenic toxicity equivalency factors (MEF and TEF) and risk assessment conducted using carcinogen (benzo[a]pyrene) showed immensely high risk due to the consumption of smoke-cured fish using hardwoods (mangroves and acacia). Among the three wood types, sugarcane bagasse was the relatively safest smoke-generating source for smoke-curing fish when using the conventional kiln (Essumang *et al.*, 2013).

The occurrence of polycyclic aromatic hydrocarbons in beef meat (Egypt) was examined by Abou-Arab *et al.* (2014) and observed that the concentration of PAHs was 2.611 $\mu\text{g}/\text{kg}$. Treatment of beef samples by boiling for forty minutes reduced the carcinogenic and total PAHs by 2.69% and 14.52 respectively. By using food additives (pepper, salt, bharrat, onion, rosemary and cumin) as a pre-treatment before boiling, the percentage of reduction in carcinogenic and total PAHs reached 91.32 percent and 89.79 percent, respectively.

Kpoclou *et al.* (2014) reported that shrimp smoked with acacia charcoal in chorkor kiln resulted in safe product regarding its PAH contamination (benzo[a]pyrene - 5 $\mu\text{g}/\text{kg}$ and a sum of benzo[a]pyrene, chrysene, benzo[a]anthracene and benzo[b]fluoranthene - 28 $\mu\text{g}/\text{kg}$) following a European standard of 5 and 30 $\mu\text{g}/\text{kg}$, respectively, whereas shrimp smoked with wood exceeds European Union limits.

The effect of marinating meat with black beer, non-alcoholic, Pilsner beer on the generation of polycyclic aromatic hydrocarbons in pork grilled with charcoal was

evaluated by Viegas *et al.* (2014) and found that the marinade of black beer was the most effective in reducing the formation of PAH, providing a suitable mitigation strategy.

Bansal and Kim (2015) concluded that the usage of lean meat and fish, exposing the area containing less fat for grilling, avoiding the direct food contact with flames during barbequing and cooking at lower temperature and replacement of traditional wood with grill and kiln reduce PAH formation. They also stated that the use of electric or gas meat broilers over charcoal and the use of acid-base marination like lemon juice before grilling reduced the PAH formation.

The effects of food additives in household cooking techniques on polycyclic aromatic hydrocarbons formation in small ruminants (goat, sheep) meat varieties in Punjab, Pakistan was studied by Ahmad (2017) and reported that the modified condiments type (Normal condiments + onion + garlic + ginger + clove + lemon juice) reduced the production of PAHs in the heat treated meat samples. He also stated that open flame grilling should be avoided and condiments used before cooking minimize the production of PAHs.

Wongmaneepratip and vangnai (2017) reported that the addition of an alkali and/or oil in the marinade would be significant in maximizing the concentration of PAHs in the meat of grilled variety. A higher pH (7.51) of the marinade treatment-double Base (DB) marinade treatment (water, sugar, oyster sauce, salt, garlic powder, black pepper powder, palm-oil and sodium bicarbonate) significantly increased the levels of the PAHs, especially of BbF (146.5 $\mu\text{g}/\text{kg}$) and BaP (84.6 $\mu\text{g}/\text{kg}$), which were the heavy PAHs. These are considered as (group 2B) possible carcinogens to humans and also increased the intensity of the dark color of grilled chicken owing to the maillard reaction.

2.7 SHELF-LIFE OF SHEEK KEBAB (pH, TBARS AND MICROBIAL QUALITY)

2.7.1 pH

The mean values of pH for raw ingredients, uncooked donairs (pH 5.8), cooked donairs (pH 5.9), overnight cold-stored donairs (pH 6.4) and reheated donairs (pH 6.1) indicated a slight rise in pH during cooking and storage. The lesser pH values of the raw ingredients were due to the marination effect (pH 4.5 to 5.1, mean pH 4.8) (Todd *et al.* 1986).

The Institute of Standards and Industrial Research (Iran) recommended the pH range of 5.8–6.2 for the raw kebabs (ISIRI 1998). The pH of kebabs depends on the storage temperature, time and the materials used in the marination.

Kayisoglu *et al.* (2003) reported that the chicken doners had greater pH values than beef doners. The mean pH values of cooked doners (6.03) were significantly greater than that of the raw doners (5.86).

The pH values of the cooked and raw doner kebabs and sucuk dough decreased with storage time. The pH values of the doners were remarkably different at 60, 30, and 0 days of the storage. The pH values of the cooked sucuk doners were initially 6.18 but decreased gradually to 6.00 on day 30 and 5.64 on day 60 of storage (Gonulalan *et al.*, 2004).

The average value of the pH of chicken doner kebab was 6.03 (5.44–6.28). There was a significant difference in pH of doner kebab manufactured by different Turkish restaurants (Vazgecer *et al.* 2004)

Modi *et al.* (2007) reported that the pH value of the dehydrated chicken kebab mix decreased ($P = 0.05$) from 5.8 ± 0.08 to 5.5 ± 0.12 during the 6 months storage period at ambient temperature of 27.2°C. The gradual decrease in pH was due to chemical changes and not to microbial activity.

Das *et al.* (2008) reported that the pH values in vacuum packaged goat meat patties were in increasing trend during refrigerated ($4\pm 1^{\circ}\text{C}$) storage (6.46- 6.54).

Chemical analysis of the Bonab kebabs marketed in the Tabriz market showed the pH values of 5.99–6.45. The average pH values of cooked Bonab kebab (6.45) samples were significantly greater than the raw samples (5.99) (Nemati *et al.*, 2008).

Al-Bachir *et al.* (2010) studied the impact of gamma irradiation on the chemical quality of chicken kebab and reported a pH value of 6.10 ± 0.08 . They also added that irradiation of chicken kebab (6, 4, and 2 kGy) significantly increased the pH values.

Bhat *et al.* (2010) evaluated the impact of black bean as an extender in chicken sheek kebabs and reported that pH increased significantly ($P < 0.05$) with raise in the level of extender. Maximum pH value (6.19) was noticed in the kebabs extended with 20% black bean compared to control samples (6.09).

The effect of green gram (*Vigna radiata*) on the quality parameters of chicken Sheek kebab (microwave cooked) was studied by Bhat and Pathak (2011) and they reported that pH value increased significantly with the level of extender and reached a maximum pH value (6.30) when extended with 20% green gram in comparison to control samples (6.15).

Biswas *et al.* (2012) reported that the pH values of enrobed pork patties increased significantly ($P<0.05$) during refrigerated ($4\pm 1^{\circ}\text{C}$) storage. (6.1 – 6.5)

Nayar *et al.* (2014) compared the pH values of microwave dried and hot air oven dried goat meat cubes and interpreted that the hot air oven dried meat had significantly higher pH value (6.45) than that of microwave dried meat (6.15).

Ali *et al.* (2018) determined the changes in the chemical and physical properties of Kebab (20% beef, 5% sheep fat tail, and 75% meat of sheep flank) when admixed with chicken breast meat. They observed significant difference in pH value ($P<0.05$) between the raw (5.61-5.82) and grilled sample (6.00-6.06).

Malav *et al.* (2016) reported that the mutton patties cooked in a hot air oven revealed a pH value of 6.27 at 180°C for 30 minutes.

Oz *et al.* (2017) reported the pH value of 5.62 in beef steaks processed by a hot air oven at 200°C for 9 minutes was similar to the microwaving method of cooking for 4.5 minutes (5.63).

Karpinska-Tymoszczyk *et al.* (2020) evaluated the impact of the time and temperature of chicken breast fillets (sous vide cooked) and concluded that significant ($p < 0.05$) differences in pH were observed between samples cooked at the same temperature but over different times, and meats subjected to longer cooking were characterized by a significantly lower pH ($p < 0.05$).

2.7.2 2-THIOBARBITURIC ACID REACTIVE SUBSTANCES (2-TBARS)

De Vore (1988) estimated the TBARS levels of the cooked and raw beef patties after a storage period of 0, 2, and 4 days at 4°C reported that the TBA values increased markedly after two days of storage and doubled from day 2 to day 4. TBA values for raw beef patties (0.2, 0.8 and 1.3) and cooked beef patties (0.1, 5.8 and 9.5) over 0, 2 and 4 days of storage, respectively.

Al-Ismail (2002) studied the effect of two methods of grilling (direct blue and charcoal radiation heat) on the oxidative rancidity in chicken shawarma and beef and reported that TBARS values were not significant ($P=0.05$) with the method of grilling. This is due to the elevated temperatures of grilling which resulted in the decomposition of TBA. He also added that the TBA levels of chicken shawarma were significantly ($P=0.5$) greater than beef samples.

The combined effect of cooking techniques (roasting and grilling) and chilling storage (without and with air) on cholesterol and lipid oxidation in the chicken breast was analyzed by Conchillo *et al.* (2003) and interpreted that aerobic storage enhances the TBARS values for raw samples (0.04 to 0.06 ppm), grilled samples (0.21 to 1.20

ppm) and roasted samples (0.24 to 1.62 ppm). Roasted samples only showed a significant increase in TBARS during vacuum storage. They also added that the TBARS values increased with cooking and especially with storage.

Sausage doner showed significantly ($P < 0.05$) greater TBARS values than traditional doner during storage ($-30 \pm 1^\circ\text{C}$). With storage time, the sample TBARS values tended to increase (raw and grilled sausage type of doners had 0.390 and 0.460 mg/kg malonaldehyde (MA) on day 0, but had TBARS values of 0.467 and 0.530 on day 60, respectively) (Gonulalan *et al.*, 2004).

The chemical characteristics of doner kebab prepared from turkey meat was analyzed by Kayaardi *et al.* (2006) and revealed that the TBA values increased 36% from day 1 to 9, and 100% from day 9 to 18, in which the values showed that the refrigerated storage duration had a significant effect ($P < 0.05$) on TBA values.

Modi *et al.* (2007) measured the oxidative rancidity of the chicken kebab mix (dehydrated) by the TBARS method and interpreted a gradual ($P = 0.05$) ascending trend from initial values of $2.9 \pm 0.58 - 5.3 \pm 0.44$ during storage for six months.

Das *et al.* (2008) reported that the TBARS values were in increasing trend in vacuum packaged goat meat patties refrigerated storage (0.248- 0.585 mg malonaldehyde/kg).

The effect of the several cooking methods (roasting, grilling, microwave, and frying) on the lipid oxidation of from Iberian pig meat was studied by Broncano *et al.* (2009) and reported that significantly greater lipid oxidation TBARS ($p < 0.001$) took place, in which the roasted meat (1.35 ± 0.19 mg MDA/kg) and fried meat (1.19 ± 0.14 mg MDA/kg) had the highest oxidation level followed by the microwave (1.03 ± 0.09 mg MDA/kg) and grilled samples (0.86 ± 0.11 mg MDA/kg). He also claimed that the application of heat to the roasted method over a long period of time (20 min) produced higher oxidation compared to the changes induced by the use of higher temperature

(190 C) in the grilled method over a shorter period of time (4 min).

Al-Bachir *et al.* (2010) studied the impact of gamma irradiation on the chemical quality of chicken kebab and reported that the TBA value was not affected by the dose of irradiation up to 6 kGy.

Del Pulgar *et al.* (2012) observed the physico-chemical characteristics of the pork cheeks (sous-vide cooked) and reported a significant interaction ($P= 0.007$) between the temperature and time influences the TBARS values, while the time and temperature tended to be significant ($P= 0.084$ and $P= 0.072$ respectively). It might be implied that prolonged cooking duration and temperature would lead to more lipid oxidation. He also stated that there were no major variations in TBARS between cooking unpackaged samples in boiling water and packaged samples cooked under the same conditions.

The impact of processing conditions on the physico-chemical properties of the shami kebab was studied by Pandey *et al.* (2014) and revealed that the TBA values ranged from 0.38 to 0.68 mg malonaldehyde/kg and TBARS values were slightly higher in the grilled compared to deep-fat frying conditions. He further stated that a direct correlation existed between the degree of lipid breakdown and the processing temperature.

Malav *et al.* (2016) reported that the mutton patties cooked in a hot air oven revealed a TBARS value (mg malonaldehyde/kg) of 0.17 at day 0, 0.33 at day 15, 0.47 at day 30, 0.59 at day 45, and 0.86 on day 60 of refrigerated storage.

The TBARS value in the meat products increased significantly with the advancement of storage interval. Lipid oxidation for oven-cooked chicken nuggets (0.79 mg malonaldehyde/kg) however, showed slightly higher lipid oxidation ($p < 0.05$) than steam-cooked nuggets (0.44 mg malonaldehyde/kg) and less compared to the microwave-cooked nuggets (1.12 mg malonaldehyde/kg). (Pathera *et al.*, 2016)

The TBARS value of beef steaks processed by hot air oven at 200°C for 9 minutes (3.32 µmol malonaldehyde/kg) is lower when compared to the microwaving method of cooking for 4.5 minutes (4.44 µmol malonaldehyde/kg) (Oz *et al.*, 2017).

Soladoye *et al.* (2017) investigated the impact of cooking methods (microwave or frying pan) and refrigerated storage on the formation of lipid oxidation (TBARS) in bacon and reported that frying pan produces higher levels of TBARS (P<0.001) in bacon.

Kiran *et al.* (2018) reported that hot air oven dried chicken patties had significantly lower TBARS value (0.45) than the moist cooked chicken nuggets (0.48 mg malonaldehyde/kg).

Karpinska-Tymoszczyk *et al.* (2020) evaluated the effect of the time and temperature of chicken breast fillets by sous vide cooking and concluded that the TBARS value in the raw meat was 0.29 mg malonaldehyde/kg. Thermal processing at several temperatures (55°C, 58°C and 61°C) for the shorter cooking time (260 and 90 minutes) showed the TBARS value of (0.25, 0.41, and 0.23) mg malonaldehyde/kg, respectively and induced no significant change (P>0.05), but when cooking duration was longer at the temperatures of 55°C and 61°C, (320 and 150 minutes) TBARS value increased significantly to 0.65mg malonaldehyde/kg and 0.49mg malonaldehyde/kg respectively.

2. 7. 3 MICROBIAL ANALYSIS

Todd *et al.* (1986) observed the highest microbial counts (10^7 aerobic colony counts ACC/g and 10^3 E. coli/g) occurred in the raw meat of doner kebab. Marination or addition of spices lowered the counts only slightly and cook for 1 hour or less marginally lowered and when cooked for over 1 hour resulted in ACCs less than 10^5 Cfu/g and not detectable *Escherichia coli* counts.

Kayisoglu *et al.* (2003) observed the microbiological characteristics of doner

kebab and reported the values of aerobic plate count (5.68–4.92 cfu/g), psychrotrophic bacteria (5.14-3.48 cfu/g), *coliforms* (4.79–2.88) cfu/g, and mould and yeast (4.96–3.55 cfu/g) for the raw and cooked doner kebab treatments respectively.

The microbiological safety of the shelf-stable intermediate moisture mutton kebabs prepared by employing the hurdle technology (irradiation, reduced water activity, and vacuum packing) was studied by Chawla and chander (2004), revealed that radiation treatment (2.5 kGy) resulted in the complete elimination of the inoculated 10^6 cfu/g of *Staphylococcus aureus* and *Bacillus cereus* but not with *Clostridium sporogenes*. Vacuum packaging and the water activity (a_w) of 0.85 of products eliminated the growth of all three organisms inoculated into that sample during three months of storage period at room temperature.

Gonulalan *et al.* (2004) carried out a quality analysis of doner kebab prepared from sucuk dough and reported that the microorganisms count decrease with storage time in both the raw doners and grilled counterparts, due to the low storage temperature. They also interpreted that the effects of storage on the microbial count raised with time (total *coliforms* of the raw sausage doner was log 5.16 cfu/g at the day 0 while it was 4.95 and 4.86 at the day 30 and day 60 of storage, respectively).

The mesophilic aerobic bacteria counts of chicken doner kebab were found to be in the level of 1.0×10^2 - 6.4×10^5 /g. *Bacillus cereus*, *coliforms*, and *staphylococci* counts were less than 10^2 /g for a total of 48%, 61%, and 50% of the samples, respectively. *Escherichia coli* was reported in 31% of the samples and their level ranged between 2.0×10^2 and 5.0×10^2 / g. (Vazgecer *et al.*, 2004)

Sensory and chemical analysis of doner kebab prepared from turkey meat revealed that the total the bacterial count increased exponentially with the storage time (the initial total bacterial count of the samples was 4.63 log cfu/g, and increased to 9.20 log cfu/g at the end of 24 days of storage). This raise was found to be significant

($P < 0.05$) (kayaardi *et al.* 2006).

Modi *et al.* (2007) recorded that over the six-month storage span, the dehydrated chicken kebab mix suggested low bacterial counts and the absence of coliforms, while the Standard Plate Count and spore counts fluctuated within the small range of 4.4-4.7 and 3.8-4.4 log cfu / g. *Staphylococci* counts initially were 3.3 log cfu/g which markedly ($P = 0.05$) decreased by over 2 log cycles after one month of storage and the counts then marginally ($P = 0.05$) increased to 2.0 log cfu/g after storage for 6 months.

Cholan (2008) studied the effect of fat replacers on the quality and storage stability of low-fat chicken nuggets and reported that psychrophiles were not detected in any of the treatments during refrigerated storage ($4 \pm 1^\circ\text{C}$).

Microbiological analysis of the Bonab kebabs marketed in the Tabriz market showed 6.75–3.22 cfu/g aerobic plate count, 3.36–0 cfu/g *Clostridium*, 4.53–3.39 cfu/g mold and yeast and 5.30–1.69 cfu/g *coliforms* for the raw and cooked samples, respectively (Nemati *et al.*, 2008)

Al-Bachir *et al.* (2010) studied the influence of gamma irradiation on the sensory quality, chemical, and microbial load of the chicken kebab and found that the action of irradiation on bacterial damage was immediate. On the 0th day, total counts were lowered to less than 10 cfu/g for chicken kebab irradiated at 6, 4, and 2 kGy. The shelf-life interval will be extended to a period of 3 and more than 5 months when samples are irradiated at 4 or 6 kGy.

Chandralekha (2010) studied the effect of natural (bengal gram flour) and synthetic antioxidants (BHA and BHT) in chicken meat balls and reported that the psychrophiles and yeast and moulds were not detected in any of the treatments during refrigerated storage ($4 \pm 1^\circ\text{C}$).

Nisar *et al.* (2010) reported that the beef patties with 5% added fat processed by microwave oven cooking had higher standard plate count and psychrotrophic count

(1.67 and 0.54 log cfu/g) compared to the hot air oven cooking (1.59 and 0.52 log cfu/g) and pressure cooking (0.67 and 0.19 log cfu/g).

The sous-vide method of cooking of the lamb loin dramatically reduced the microbial population even with less intense heat treatment (60 °C for 6 hours). Samples of the raw loin showed the counts for psychrotrophic and mesophilic bacteria greater than 4 log Cfu/g; whereas for *Enterobacteriaceae*, and *Coliforms*, the counts vary from 3.23 to 3.58. However, for microbial groups, the counts were not detectable or lesser than the log 1 cfu/g, except for mesophilic bacteria even after heat treatment. (Roldan *et al.*, 2013)

Naveena *et al.* (2014) who reported that higher total plate count values were observed in moist cooked nuggets (2.6 log₁₀cfu/g) than the dry cooked chicken patties (2.3 log₁₀ cfu/g).

Nayar *et al.* (2014) interpreted that the total plate count (3.64 log cfu/g) and yeast and mold count (1.80 log cfu/g) in the hot air oven dried goat meat cubes were higher when compared to the microwave dried goat meat cubes (1.45 log cfu/g) and (1.46 log cfu/g) respectively.

The total plate count values of vacuum packaged functional mutton patties during refrigerated storage were in the increasing order of 0.93 on day 0 to 4.07 (cfu/g) at day 60 (Malav *et al.* 2016).

Pathera *et al.* (2016) reported that the cooking method does not change the microbiological characteristics of chicken nuggets during storage. All the microbial counts [Standard plate count (log cfu/g), psychrotrophic count (log cfu/g), and yeast and mould count (log cfu/g)] were significantly increased during the refrigerated storage period.

Reddy *et al.* (2017) evaluated the microbiological quality of chicken meat patties incorporated with natural antioxidant extracts i.e., rosemary and green tea and reported

that no psychophiles were detected in any of the treatments and control during refrigerated ($4\pm 1^{\circ}\text{C}$) storage.



Chapter – III

Materials and Methods

CHAPTER III

MATERIALS AND METHODS

In the present investigation, an attempt was made to study the impact of cooking methods (charcoal grilling, moist cooking followed by charcoal grilling and hot air oven cooking followed by charcoal grilling) on the quality attributes and levels of polycyclic aromatic hydrocarbons (PAHs) in the mutton sheek kebab. The entire work was divided into two phases. The first phase was the preparation of mutton sheek kebab by the above three cooking methods. The products so prepared were packed in low-density polyethylene bags and stored at refrigeration temperature ($4\pm 1^{\circ}\text{C}$).

In the second phase, the prepared products – Control (C)(direct charcoal grilling), and Treatment (T1) (moist cooking followed by charcoal grilling) and Treatment (T2) (hot air oven cooking followed by charcoal grilling) were be evaluated for various physico-chemical properties, microbial and sensory attributes on 0, 5, 10 and 15 days of refrigerated storage ($4\pm 1^{\circ}\text{C}$). Further the levels of polycyclic aromatic hydrocarbons (PAH) were also estimated.

3.1 LOCATION

The study was carried out in the ICAR-National Research Centre on Meat, Hyderabad. Preliminary trails were conducted to standardize the preparation of mutton sheek kebab employing various cooking methods. A total of six trials were conducted to evaluate the impact of different cooking methods and refrigerated storage on various physico-chemical properties, microbial and sensory attributes of mutton sheek kebab.

3.2 SOURCE OF RAW MATERIAL

Boneless mutton was procured from the local retail shops at Hyderabad and the experimental abattoir of the National Research Centre on Meat, Hyderabad. The tendon and other connective tissue were removed and the meat was chilled overnight at $4\pm 1^{\circ}\text{C}$. The spices and other ingredients were purchased from the local markets of Hyderabad.

3. 3 PREPARATION OF MUTTON SEEKH KEBABS

Chilled meat was minced in a meat mincer (Scharfen, Model X70, 58413 Witten, Germany) with a 6 mm plate and preblended with common salt and sodium tripolyphosphate followed by mixing with butter, refined wheat flour (maida), spice mix, and condiments in a planetary mixer (Smaky, France) for 10 minutes.

Table 4: Formulation of Mutton Sheek kebab

S. No.	Ingredients	Proportion (%)
1.	Mutton (boneless)	75
2.	Butter	05
3.	Onion	05
4.	Refined wheat floor (Maida)	03
5.	Green chilli	2.5
6.	Garlic	02
7.	Ginger	01
8.	Coriander leaves	0.7
9.	Coriander powder	0.5
10.	Spice mix (Garam masala)	1.0
11.	Salt	1.5
12.	Red chilli	01
13.	Sodium tripolyphosphate	0.3
14.	Fenugreek leaves (Methi)	0.5
15.	Chickpea (Channa) powder	01

Three different cooking methods viz., direct charcoal grilling (Control), moist cooking followed by charcoal grilling (Treatment 1) and hot air oven cooking followed by charcoal grilling (Treatment 2) were chosen, as representative of most common domestically employed methods of cooking. For each group, duplicate batches were prepared.

Charcoal grilling was carried out in a domestic type outdoor grill (Prestige) (45.8 x 28.8 x 13.2 cm). Approximately 2 kg of charcoal was placed in the grill oven and heated up. About 60 g of mutton batter was manually molded onto stainless steel skewers and were grilled until the internal temperature reached a minimum of 80°C (for

about 10 min). The samples were turned periodically (every 2-3 min) during the cooking period. No oil was applied to the kebabs before or after grilling.

In the modified innovative method, the kebabs were prepared employing sausage stuffer (SIRMAN, Model IS V15 IDRA VERT RUOTE, Italy). The preparation involved loading the kebab batter into the sausage stuffer and then moulding batter over the skewer, which was placed in the centre of the casing. After stuffing the batter to the full length of the skewer, both end of the casing were tied and then cooked in boiling water (moist cooking) until the internal temperature reaches 80°C (for about 10 min). The moist cooked kebabs were finally grilled in a domestic type outdoor grill for a duration of 3 minutes in order to get the desired flavor and colour. This process produced a smooth surfaced and uniformly sized kebab.

Hot air oven cooking (roasting) of the kebab samples was done in an oven (Retigo Combi Oven, Model B1011, Czech Republic) at 150°C until the core temperature of the product reached 80°C (for about 15 min) and finally, the kebabs were grilled in a domestic type outdoor grill for a duration of 3 minutes to impart the desired flavor and colour.

The prepared kebabs of control, treatment 1, and treatment 2 groups were aerobically packed in low density polyethylene packs and stored at refrigerated temperature ($4 \pm 1^\circ\text{C}$) for further analysis.

3.4 PHYSICO-CHEMICAL CHARACTERISTICS

3.4.1 Proximate analysis

Moisture, protein, fat and total ash content of kebabs were determined according to AOAC (2005). The moisture content was determined by weight loss after 12 hours of drying at 100°C in a hot air oven (Memmert type, VTS Equipments, 45 ×45× 45 cm). Protein was determined by block digestion using an auto analyser (Kel Plus, Model KES6L, Pelican Equipments, Chennai) and the fat content was estimated by an ether

extraction procedure in an automatic Soxlet Apparatus (Sox Plus, Model SCS6, Pelican Equipments, Chennai).

3.4.2 pH

The pH of mutton sheek kebab formulations was estimated following the method of Trout *et al.* (1992). Five grams of sample was blended with 45 ml of distilled water using Ultra Turrax Tissue Homogenizer (IKA digital ULTRA-TURRAX, Model T- 25, Germany) for one minute. The pH was recorded by immersing the glass electrode of digital pH meter (Eutech Instrument, Cyberscan, Singapore Model) into the homogenate. The pH of the sample was measured with the pH meter, which was calibrated with buffer solutions of pH 4 and 7 as per user manual instructions to avoid errors.

3.4.3 2-Thiobarbituric acid reactive substances (2-TBARS) value

Lipid oxidation in terms of thiobarbituric acid reactive substances (TBARS) value of mutton sheek kebab was estimated as per the procedure outlined by Strange *et al.* (1977) with slight modification. A 4 gram of kebab sample was blended for 3 min with 20 ml of 20% trichloroacetic acid. The blended sample was kept for centrifugation at 5000 rpm for 15 minutes. The supernatant content was filtered through Whatman No.1 filter paper (18.5 cm diameter) and the filtrate was collected. The filtrate termed TCA extract was used in the estimation of thiobarbituric acid number (TBA).

The test solution was prepared by mixing 3 ml of 0.1% thiobarbituric acid to the 3 ml of TCA filtrate. After mixing the contents, tubes were kept in a boiling water bath (100°C) for 30 min along with blank. Blank was prepared by mixing 5 ml of 20% trichloroacetic acid with an equal amount of 0.1% thiobarbituric acid reagent and was run simultaneously to check the experimental error. After cooling the tubes, optical density (O.D) was measured in a UV- VIS spectrophotometer (SHIMADZU, UV-1700, Japan) at 532 nm. TBARS was calculated as mg malonaldehyde per kg of sample.

3.4.4 Cooking yield

After cooking, the kebab samples were cooled to room temperature and the cooking yield was calculated from differences in raw and cooked weight.

$$\text{Cooking yield (\%)} = \frac{\text{Weight of mutton sheek kebab after cooking}}{\text{Weight of raw kebab batter}} \times 100$$

3.4.5 Instrumental color

The color measurement (Hunter L*, a* and b*) was performed using a X-Rite RM200QC Imaging Spectrocolorimeter, Lovibond, China with a 8 mm aperture set for illumination D 65, 10° standard observer angle. Color measurement was done on the cut surface of kebabs from the randomly chosen spots (Hunter and Harold, 1987). The values obtained for L*, a* and b* were used to calculate the chroma (C) and hue angle (h°) according to the following equations (Del Pulgar *et al.*, 2012)

$$\text{Chroma} = (a^{*2} + b^{*2})^{1/2}$$

$$\text{Hue angle} = \text{arc tag} (b^*/ a^* \times (360^\circ/2 \times 3.14))$$

3.4.6 Instrumental texture profile analysis (TPA)

The TPA tests were performed using a texturometer (Tinius Olsen, Model H1KF, Redhill, RH1 5DZ, England) to determine hardness, cohesiveness, springiness, gumminess, and chewiness. Three representative pieces of each sample (1.5 cm³) were compressed twice to 80% of the original height. A crosshead speed of 2 mm/s was used.

3.4.7 QUANTIFICATION OF POLYAROMATIC HYDROCARBON

3.4.7.1 Extraction and clean up process: (AOAC 2007.01)

Place 15 gram homogenized kebab sample into a 50 ml centrifuge tube (Abdos - P/N: 10403) and 15 mL of chilled acetonitrile (ACN) was added, vortexed for 5 mins. Then add Quechers AOAC salts (6g Mg So4/ 1.5g Sodium Acetate – DISQUE WATERS) (Part No: 186006812) and vigorously handshake for 1-2 minutes. Centrifuge the mixture (5000 rpm for 10 minutes at 4°C) and the supernatant was collected. Then

3mL of the supernatant was added to 6 cc/200mg HLB Cartridge (Oasis) (Part No: 186008057) for *solid-phase extraction (SPE)* in a Tube Vacuum Manifolds (Phenomenex). Discard 0.8 ml initially and the next 2 ml collected in the tubes are transferred to vials (Waters 9mm, Amber, screw top vial, 12×32mm with cap & pre-slit Polytetrafluoroethylene/ Silicone septa P/N: 186000847C) and injected into the LC – MS system. (Waters Corporation, model name: XEVO TQ-S micro system, Milford, USA)

3.4.7.2 LC Conditions

The PAH analytes were separated using a ACQUITY UPLC® BEH phenyl 1.7 column of 100 mm x 2.1mm, using a 1.7 µm particle size (2.1 x 100 mm, 1.7 µm, Waters, made in Ireland). The temperature of the column oven was set at 30 °C. The mobile phases consisted of (A) containing 0.1% formic acid in water and (B) acetonitrile. The flow rate was 0.4 mL/min and the mobile phase gradient is shown in Table 1. The injection volume was 10µL. (Wu *et al.*, 2017)

Table 5. Mobile phase gradient.

	Time(min)	A%	B%
1	0.0	50	50
2	1.0	50	50
3	3.0	1	99
4	16.0	1	99
5	17.0	50	50
6	20.0	50	50

Table 6. MS source conditions

Ion Source	ESI positive
Electrospray Voltage	5500 V
Drying Gas	90 arbitrary units
Nebulizer Gas	320 arbitrary units
Source Temperature	500 °C
HSID Temperature	320 °C
Detection Mode	MRM

Table 7. MRM transitions used to detect PAH, alkylated derivatives, and photo-degradation products (Sakuma *et al.*, 2011)

Analyte	Q1	Q3 (quantifier)	Q3 (qualifier)
Flouranthene	202	150	200
Benzo(a)anthracene	228	150	226
Chrysene	228	200	226
Benzo(a)pyrene	252	224	250
Benzo(b)flouranthene	252	224	250
Benzo(k)flouranthene	252	224	250
Indeno(123cd)pyrene	276	246	274
Benz(ghi)perylene	276	248	274
Dibenz(a, h)anthracene	278	248	276

Table 8. Chan Reaction of different analytes of PAH compounds.

	Chan Reaction	Dwell (secs)	Cone Volt.	Energy	Delay (secs)	Compound
1.	202.00 > 150.00	0.040	80.0	50.0	0.025	Flouranthene
2.	202.00 > 200.00	0.040	80.0	50.0	0.025	Flouranthene
3.	228.00 > 150.00	0.040	80.0	50.0	0.025	Benzo(a) anthracene
4.	228.00 > 226.00	0.040	80.0	50.0	0.025	Benzo(a) anthracene
5.	228.00 > 200.00	0.040	80.0	20.0	0.025	Chrysene
6.	228.00 > 226.00	0.040	80.0	20.0	0.025	Chrysene
7.	252.00 > 224.00	0.040	80.0	50.0	0.025	Benzo(a)pyrene
8.	252.00 > 250.00	0.040	80.0	50.0	0.025	Benzo(a)pyrene
9.	276.00 > 246.00	0.040	80.0	50.0	0.025	Indeno(123cd pyrene)
10.	276.00 > 274.00	0.040	80.0	50.0	0.025	Indeno(123cd pyrene)
11.	276.00 > 248.00	0.040	80.0	50.0	0.025	Benz(ghi)perylene
12.	276.00 > 274.00	0.040	80.0	50.0	0.025	Benz(ghi)perylene
13.	278.00 > 248.00	0.040	80.0	50.0	0.025	Dibenz(a,h)anthracene
14.	278.00 > 276.00	0.040	80.0	50.0	0.025	Dibenz(a,h)anthracene

The equipment was calibrated using duplicates of six concentrations of mixed standard calibration solutions (0.025, 0.050, 0.100, 0.250, 0.500 and 1.000 µg/mL). The correlation coefficient of each PAH standard ranged from 0.93 to 0.97. Level of Quantification (LOQ) of the LC-MS used for the analysis ranges from 0.02 to 1ng/ mL.

3.4.8 SENSORY EVALUATION

The sensory attributes such as appearance, flavor, texture, juiciness, and overall acceptability of the products were evaluated using an 8 point descriptive scale (Keeton, 1983). In the 8 point scale, 8 corresponded to 'components characteristic of the highest quality', scores from 5 to 8 were considered acceptable and scores from 1 to 5 were considered unacceptable. The panel consisted of minimum of 6 trained and experienced members of the institute, who were familiar with the characteristics of the meat product. The kebabs were warmed in a microwave oven for 60 seconds just before sensory evaluation and coded samples were served at room temperature in separate booths. Water was served for cleansing the mouth between samples.

3.4.9 MICROBIAL ANALYSIS

The microbial quality of products was evaluated by estimating the Total plate count (TPC), Psychrophilic Bacterial Count (PBC), and Yeast and Mould counts (YMC) following the spread plating technique as per the procedure of ICMSF (1980).

3.4.9.1 PREPARATION OF SERIAL DILUTIONS

For microbiological analysis, 5g of representative kebab sample was homogenized with 45ml of 0.1 % sterile peptone water in a laboratory blender, and tenfold serial dilutions were made from each sample by using 0.1% peptone water as diluent.

3.4.9.2 TOTAL PLATE COUNT

About 15 – 20 ml of sterilized plate count agar (Himedia, Mumbai) at 44-46°C was poured into petri plates gently and the rotated well for even distribution of the agar without air bubbles and plates were allowed for some time to solidification of agar. The plates were then incubated in inverted positions at 37°C for 24 hrs for checking the sterility of the poured plates. Before inoculation of the sample, the agar plates are again incubated in inverted positions at 37°C for 24 hrs for sterility check. These sterile petri

plates were inoculated aseptically with 0.1ml sample from appropriate dilution in duplicates by the spread plate method using the sterilized L spreader. The plates were then incubated in inverted positions at 37°C for 24 hrs. Plates having 40- 300 colonies were selected and the colonies were counted. The results were expressed as log units per gram of the sample.

3.4.9.3 YEAST AND MOULD COUNT

The yeast and mould count was determined by spread plate method using Potato dextrose agar (Himedia, Mumbai). Sterile agar plates were inoculated with diluted sample (0.1ml) using sterilized L spreader was incubated at 23-25°C for 5 days. Colonies were counted and expressed as log units per gram of the sample.

3.4.9.4 PSYCHROPHILIC COUNT

Psychrophilic counts were obtained by incubating the plates through spread plate method using plate count agar. Plates inoculated with the diluted sample (0.1ml), using sterilized L spreader were incubated for 7-10 days at $7\pm 1^{\circ}\text{C}$. The colonies were counted and results were expressed as log units per gram of the sample.

3.5 STATISTICAL ANALYSIS

The data thus obtained was subjected to statistical analysis using SPSS MAC, version 20.0, SPSS Chicago (US).

Fig 1: Flowchart of mutton sheek kebab preparation:

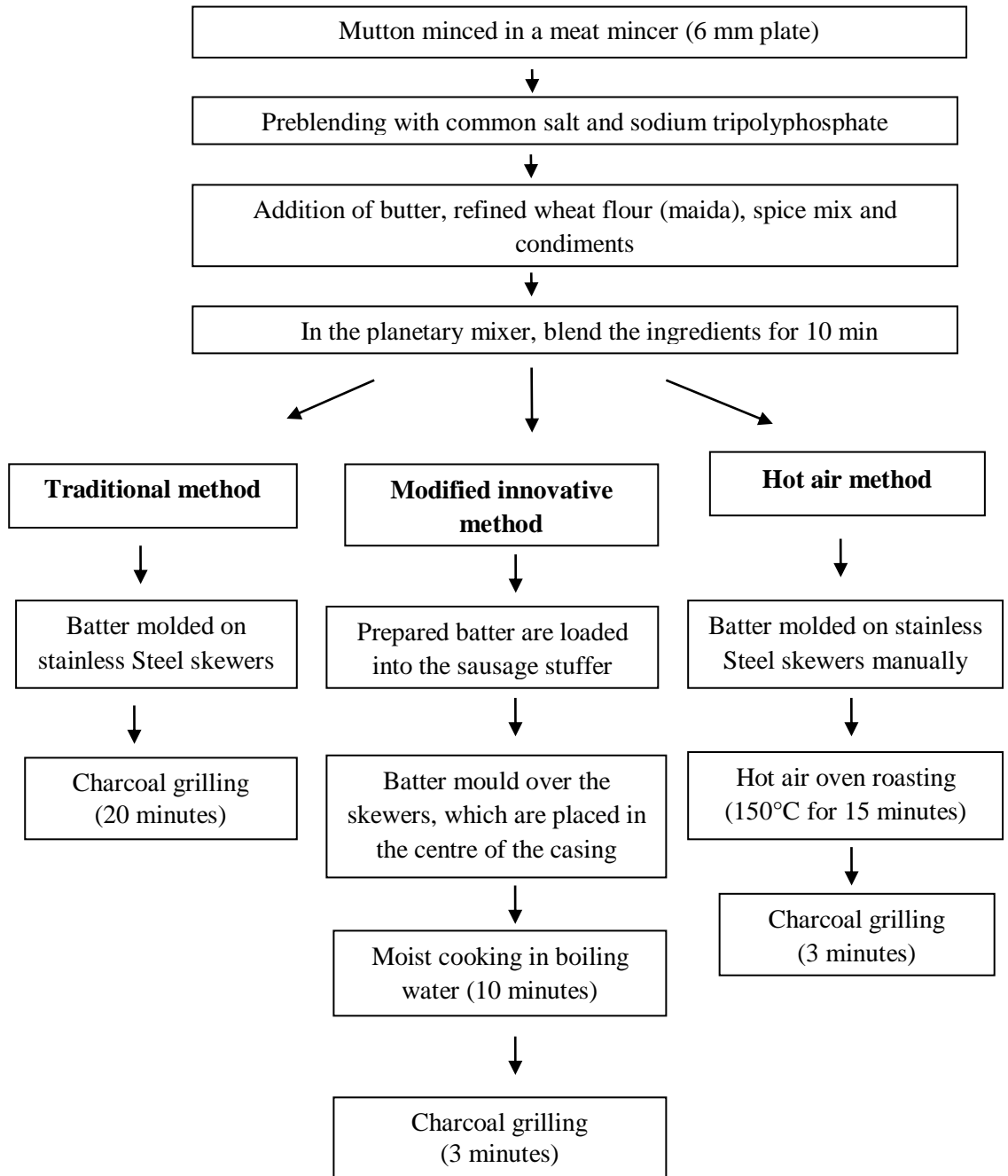




Plate no.2 Minced Mutton



Plate no.3 Batter filling process in casing along with skewer

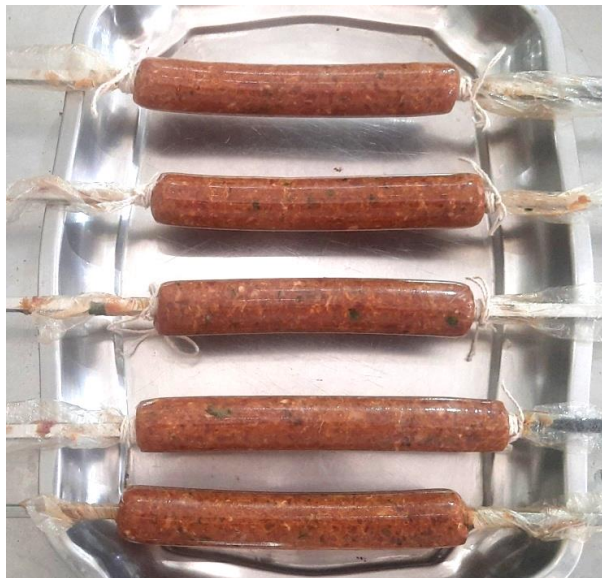


Plate no.4 Batter packed in sausage casings



Plate no.5

Moist cooking process

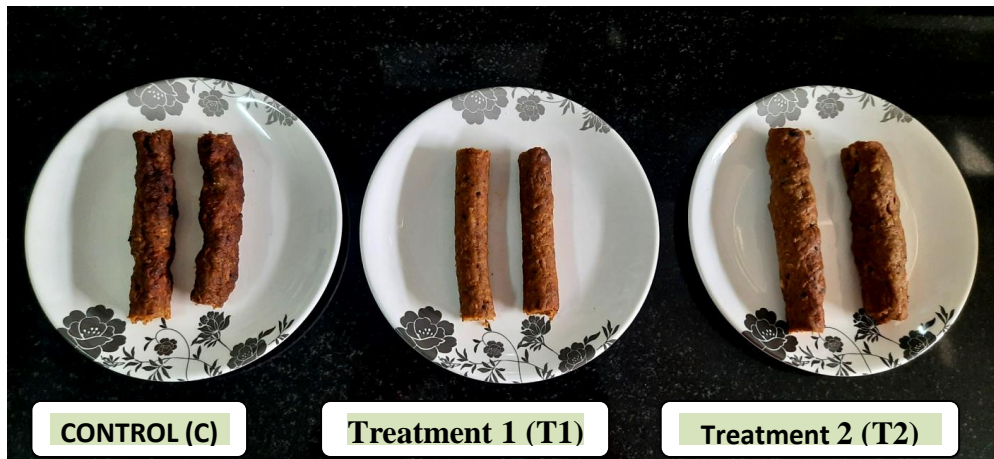


Plate no.6

Hot air method of cooking equipment (Retigo combi oven)



Plate no.7 Direct charcoal grilling (domestic type outdoor grill)



CONTROL (C)

Treatment 1 (T1)

Treatment 2 (T2)

Plate no.8 Control (C) - Direct Charcoal Grilling

Treatment (T1) - Moist Cooking followed by Charcoal Grilling

Treatment (T2) - Hot Air Oven Cooking followed by Charcoal Grilling



Plate no. 9 Liquid Chromatography Mass Spectrometer (LC-MS)



Plate no.10 Spectrocolorimeter, Lovibond



Chapter – IV

Results

CHAPTER IV

RESULTS

During the first phase of investigation, formulations and procedure for the preparation of mutton sheek kebab employing various cooking methods [direct charcoal grilling-control (C), moist cooking followed by charcoal grilling (T₁) and hot air oven cooking followed by charcoal grilling (T₂)] were standardized and several quality parameters were analyzed on the day of preparation of the products. Further, the levels of polycyclic aromatic hydrocarbons (PAH) in the prepared kebabs were also estimated.

During the second phase, kebabs prepared by different cooking methods were evaluated for several physico-chemical properties, sensory attributes and microbial quality on 0, 5, 10 and 15 days of refrigerated (4±1°C) storage. The results obtained for those parameters during phase-I and phase-II of the investigation were analyzed and presented under the following heads.

4.1 Physico-chemical properties of mutton sheek kebab on the day of preparation.

4.1.1 Cooking yield

The Mean ± SE values of cooking yield for the control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs are presented in Table 9 and the analysis of variance in Table 12.

The mean cooking yield values for the control (C), treatment 1 (T₁) and treatment 2 (T₂) group kebabs were 67.10±0.64, 78.66±0.24 and 74.69±0.33, respectively. Significant difference (P<0.05) in cooking yield was noticed among the control and treatment group kebab samples. The mutton kebab prepared by moist cooking followed by charcoal grilling (T₁) had significantly (P<0.05) higher cooking yield than the kebabs prepared by hot air oven cooking followed by charcoal grilling (T₂) and direct charcoal grilling (C).

4.1.2 Proximate analysis

4.1.2.1 Moisture content

The Mean \pm SE values of moisture content for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 9 and analysis of variance in Table 12.

The mean percent moisture values for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 57.79 \pm 0.26, 66.01 \pm 0.36 and 61.68 \pm 0.20, respectively. Significant difference (P<0.05) was noticed in moisture values among the control, T₁ and T₂ groups. The moisture content of moist cooked followed by charcoal grilled mutton sheek kebab (T₁) was significantly (P<0.05) higher than the hot air oven cooked and charcoal grilled mutton sheek kebabs (T₂) and control group.

Table 9. Effect of different cooking methods on the cooking yield, proximate composition of mutton sheek kebab (Mean \pm SE)

Parameter	Control (C)	Treatment 1 (T ₁)	Treatment 2 (T ₂)
Cooking yield (%)	67.10 \pm 0.64 ^a	78.66 \pm 0.24 ^c	74.69 \pm 0.33 ^b
Moisture (%)	57.79 \pm 0.26 ^a	66.01 \pm 0.36 ^c	61.68 \pm 0.20 ^b
Crude protein (%)	23.86 \pm 0.15 ^c	19.37 \pm 0.30 ^a	20.23 \pm 0.12 ^b
Crude fat (%)	6.32 \pm 0.07 ^c	5.73 \pm 0.02 ^a	5.96 \pm 0.03 ^b
Ash (%)	4.15 \pm 0.04 ^c	3.60 \pm 0.01 ^a	3.76 \pm 0.04 ^b

Control: Direct charcoal grilling; **Treatment -1:** Moist cooking followed by charcoal grilling; **Treatment -2:** Hot air oven cooking followed by charcoal grilling.

Note: (P<0.05); Means bearing common superscript along the rows do not differ significantly.

4.1.2.2 Per cent crude protein

The Mean \pm SE values of percent crude protein for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 9 along with analysis of variance in Table 12.

The mean percent crude protein values of control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 23.86±0.15, 19.37±0.30, and 20.23±0.12 respectively. Significant difference (P<0.05) in percent crude protein values was noticed among control (C), treatment 1 (T₁) and treatment 2 (T₂) groups. The control (C) samples showed significantly (P<0.05) higher crude protein than T₁ and T₂ treatment groups.

4.1.2.3 Per cent crude Fat

The Mean ± SE values of percent crude fat for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 9 and analysis of variance in Table 12.

The mean percent crude fat values of control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 6.32±0.07, 5.73±0.02 and 5.96±0.03, respectively. Significant difference (P<0.05) in percent crude fat was noticed among the control, T₁ and T₂ groups. The control (C) showed significantly (P<0.05) higher crude fat content than T₁ and T₂ treatment groups.

4.1.2.4 Ash content

The Mean ± SE values of ash content (%) for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 9 and analysis of variance in Table 12.

The mean ash content of control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 4.15± 0.04, 3.60±0.01 and 3.76±0.04 respectively. Significant difference (P<0.05) in ash content was noticed among the control, T₁ and T₂ groups. The control (C) showed significantly (P<0.05) higher ash content followed by T₂ and T₁ treatment groups.

Fig 2: Effect of different cooking methods on the cooking yield of mutton sheek kebab

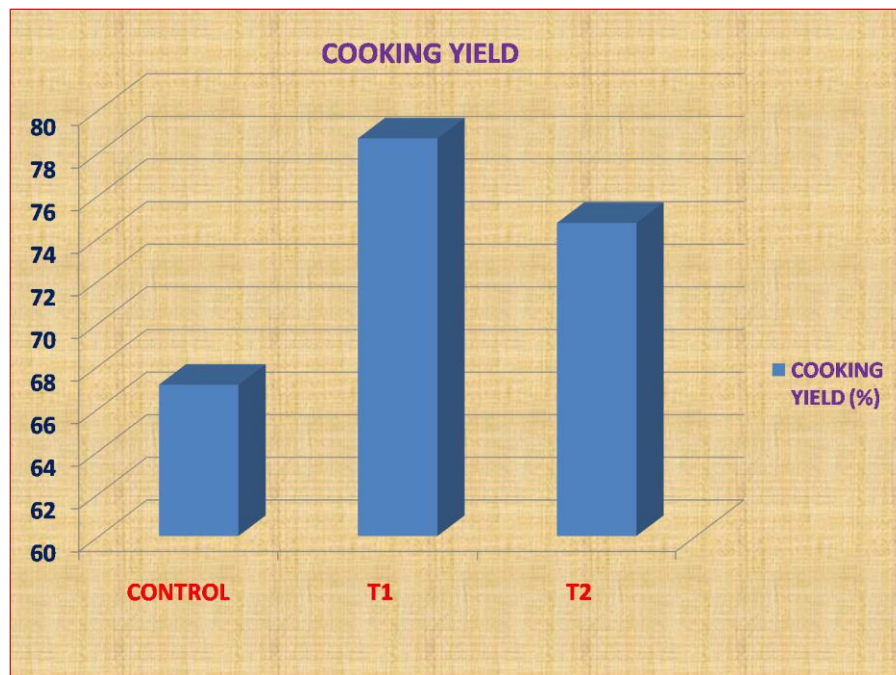
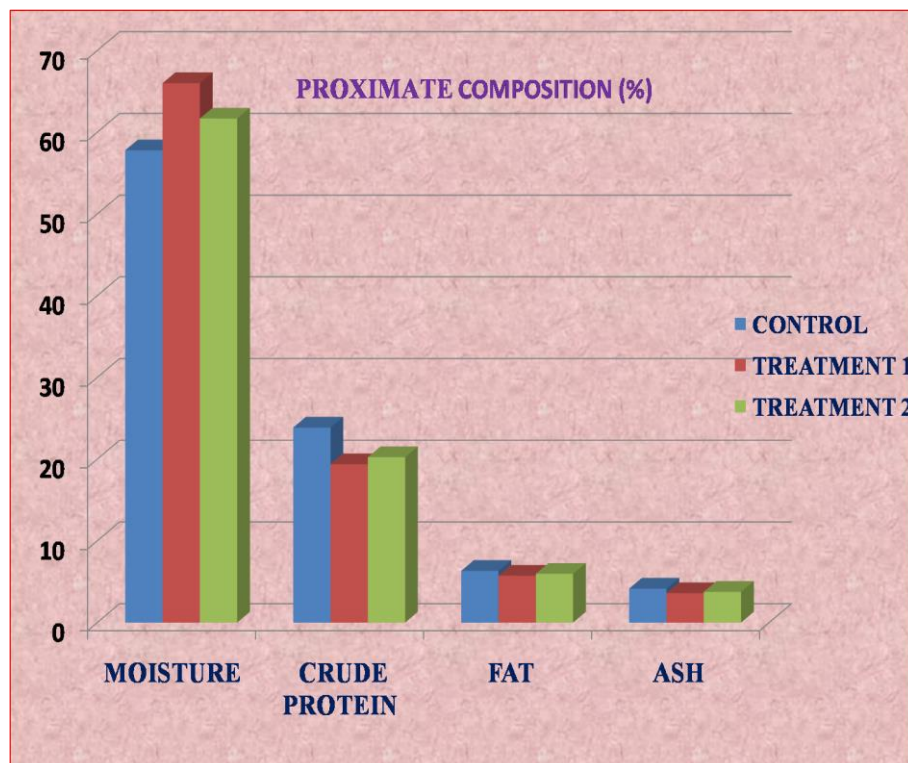


Fig 3: Effect of different cooking methods on the proximate composition of mutton sheek kebab



4.1.3 Color

4.1.3.1 Lightness (L*)

The Mean \pm SE values of lightness (L*) for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 10 and analysis of variance in Table 12.

The mean L* values for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 34.84 \pm 0.34, 37.33 \pm 0.39 and 36.33 \pm 0.85, respectively. Mutton kebabs prepared by moist cooking followed by charcoal grilling (T₁) had significantly (P<0.05) higher lightness values than T₂ and control groups.

Table 10. Effect of different cooking methods on the color parameters of mutton sheek kebab (Mean \pm SE)

Parameter	Control (C)	Treatment 1 (T ₁)	Treatment 2 (T ₂)
Lightness (L*)	34.84 \pm 0.34 ^a	37.33 \pm 0.39 ^b	36.33 \pm 0.85 ^{ab}
Redness (a*)	11.36 \pm 0.19 ^a	13.41 \pm 0.56 ^b	10.8 \pm 0.31 ^a
Yellowness (b*)	17.93 \pm 0.38 ^a	18.71 \pm 0.94 ^a	16.68 \pm 0.74 ^a
Hue angle	57.5 \pm 0.56 ^b	54.17 \pm 0.88 ^a	56.79 \pm 1.18 ^{ab}
Chroma	21.11 \pm 0.33 ^{ab}	23.03 \pm 1.03 ^b	19.98 \pm 0.67 ^a

Note: (P<0.05); Means bearing common superscript along the rows do not differ significantly. **Control:** Direct charcoal grilling, **Treatment -1:** Moist cooking followed by charcoal grilling and **Treatment -2:** Hot air oven cooking followed by charcoal grilling.

4.1.3.2 Redness (a*)

The Mean \pm SE values of redness (a*) for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 10 and analysis of variance in Table 12.

The mean a* values for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 11.36 \pm 0.19, 13.41 \pm 0.56 and 10.8 \pm 0.31, respectively. The

Redness values of T₂ group were significantly (P<0.05) higher than the rest of the formulations.

4.1.3.3 Yellowness (b*)

The Mean \pm SE values of Yellowness (b*) for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 10 and analysis of variance in Table 12.

The mean Yellowness (b*) values for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 17.93 \pm 0.38, 18.71 \pm 0.94 and 16.68 \pm 0.74, respectively. Though the T₂ group samples had numerically higher yellowness (b*) values than T₁ and Control group, there was no significant difference (P>0.05) among them.

4.1.3.4 Hue angle

The Mean \pm SE values of Hue angle for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 10 and analysis of variance in Table 12.

The mean Hue angle values for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 57.5 \pm 0.56, 54.17 \pm 0.88 and 56.79 \pm 1.18, respectively. Among the mutton kebabs, the samples prepared by moist cooking followed by charcoal grilling (T₁) had lower (P<0.05) Hue angle values followed by T₂ and control groups.

4.1.3.5 Chroma

The Mean \pm SE values of chroma for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 10 and analysis of variance in Table 12.

The mean chroma values for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups mutton sheek kebab were 21.11 \pm 0.33, 23.03 \pm 1.03 and 19.98 \pm 0.67, respectively.

Mutton kebabs prepared by hot air oven followed by charcoal grilling (T₂) had significantly lower (P<0.05) chroma values than T₁ and control samples.

4.1.4 Quantification of Polycyclic Aromatic Hydrocarbons

The Mean \pm SE values of flouranthene for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups are presented in Table 11 and analysis of variance in Table 12.

Out of 9 PAH compounds, only one PAH compound flouranthene is detected in all the kebabs formulations, while the rest of the PAH compounds are below the level of quantification.

Table 11. Effect of different cooking methods on the levels of PAH compounds in the mutton sheek kebab (Mean \pm SE)

Parameter (ng/ml)	Control (C)	Treatment 1 (T₁)	Treatment 2 (T₂)
Flouranthene	0.034 \pm 0.009 ^b	0.0237 \pm 0.002 ^a	0.0267 \pm 0.005 ^a
Benzo(a)anthracene	ND	ND	ND
Chrysene	ND	ND	ND
Benzo(a)pyrene	ND	ND	ND
Benzo(b)flouranthene	ND	ND	ND
Benzo(k)flouranthene	ND	ND	ND
Indeno(123cd)pyrene	ND	ND	ND
Benz(ghi)perylene	ND	ND	ND
Dibenz(a, h)anthracene	ND	ND	ND

Note: (P<0.05); Means bearing common superscript along the rows do not differ significantly. **Control:** Direct charcoal grilling, **Treatment -1:** Moist cooking followed by charcoal grilling and **Treatment -2:** Hot air oven cooking followed by charcoal grilling.

ND = not detected, below the LOQ.

The mean flouranthene values for control (C), treatment 1 (T₁) and treatment 2 (T₂) mutton sheek kebab groups were 0.034 \pm 0.009, 0.0237 \pm 0.002 and 0.0267 \pm 0.005 respectively. The flouranthene values of direct charcoal grilling (C) group were

Fig 4: Effect of different cooking methods on the color parameters of mutton sheek kebab

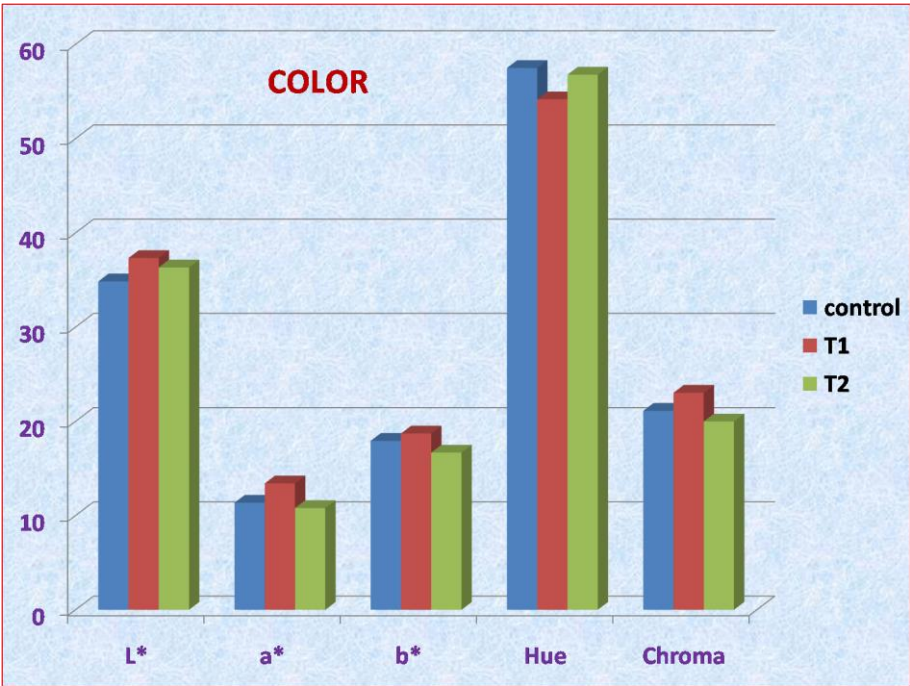
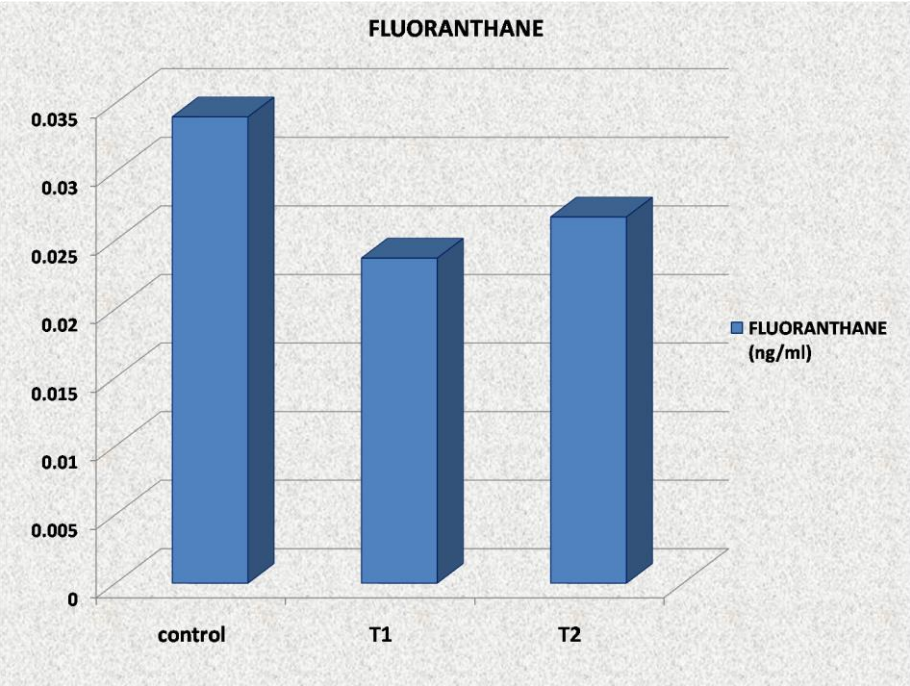


Fig 5: Effect of different cooking methods on the fluoranthane level (ng/ml) of mutton sheek kebab



significantly ($P < 0.05$) higher followed by moist cooling followed by charcoal grilling (T_1) and hot air oven cooked and grilled kebab (T_2).

Table 12: Analysis of variance (ANOVA) for physico-chemical and microbial quality attributes of mutton sheek kebab as influenced by different cooking methods on day 0.

ANOVA						
		Sum of Squares	Df	Mean Square	F	Sig.
Cooking yield	Between Groups	414.531	2	207.265	180.69	0.000*
	Within Groups	17.206	15	1.147		
	Total	431.736	17			
L*	Between Groups	18.973	2	9.487	4.783	0.025*
	Within Groups	29.753	15	1.984		
	Total	48.727	17			
a*	Between Groups	22.646	2	11.323	12.876	0.001*
	Within Groups	13.191	15	0.879		
	Total	35.838	17			
b*	Between Groups	12.625	2	6.312	1.998	0.170 ^{NS}
	Within Groups	47.389	15	3.159		
	Total	60.014	17			
Hue	Between Groups	37.618	2	18.809	3.771	0.047*
	Within Groups	74.821	15	4.988		
	Total	112.439	17			
Chroma	Between Groups	28.504	2	14.252	4.378	0.032*
	Within Groups	48.833	15	3.256		
	Total	77.337	17			
Moisture	Between Groups	203.127	2	101.563	216.09	0.000*
	Within Groups	7.050	15	0.470		
	Total	210.177	17			
Protein	Between Groups	67.955	2	33.978	129.88	0.000*
	Within Groups	3.924	15	0.262		
	Total	71.879	17			
Fat	Between Groups	1.066	2	0.533	35.981	0.000*
	Within Groups	0.222	15	0.015		
	Total	1.288	17			
Ash	Between Groups	0.964	2	0.482	53.213	0.000*
	Within Groups	0.136	15	0.009		

	Total	1.099	17			
Flouranthene	Between Groups	.000	2	.000	3.990	.041*
	Within Groups	.001	15	.000		
	Total	.001	17			

(*) - Significant, (NS) – Not significant

4.2 Shelf- life studies of mutton sheek kebab during refrigerated ($4\pm 1^{\circ}\text{C}$) storage.

4.2.1 pH

The mean \pm SE pH values for control (C), treatment 1 (T_1) and treatment 2 (T_2) group of mutton sheek kebabs during refrigerated ($4 \pm 1^{\circ}\text{C}$) storage period are presented in Table 13 and analysis of variance in Table 28 and 29.

The mean pH \pm S.E values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 6.12 ± 0.19 , 6.17 ± 0.02 , 6.22 ± 0.01 and 6.27 ± 0.01 respectively.

The mean pH \pm S.E values for treatment 1 group kebabs (T_1) on 0, 5, 10 and 15th day of refrigerated storage were 6.13 ± 0.02 , 6.17 ± 0.02 , 6.23 ± 0.01 and 6.27 ± 0.01 respectively.

The mean pH \pm S.E values for treatment 2 group kebabs (T_2) on 0, 5, 10 and 15th day of refrigerated storage were 6.20 ± 0.00 , 6.22 ± 0.01 , 6.25 ± 0.01 and 6.28 ± 0.00 respectively.

The overall mean \pm S.E values for pH of mutton sheek kebab for control (C), treatment 1 (T_1) and treatment 2 (T_2) groups were 6.19 ± 0.01 , 6.20 ± 0.01 and 6.24 ± 0.01 respectively. The overall mean \pm S.E. values for pH of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 6.15 ± 0.01 , 6.19 ± 0.01 , 6.23 ± 0.01 and 6.27 ± 0.00 respectively.

On all the storage days, control (C) group had lower pH values, compared to both T_1 and T_2 treatment groups.

Analysis of variance revealed a significant ($P < 0.05$) difference in pH value of mutton sheek kebabs in T_2 group on all storage days, while no significant difference ($P > 0.05$) was noticed in control and treatment T_1 group till day 5. Irrespective of the groups, a gradual increase in pH values was noticed, as the storage period advanced.

Spoilage was noticed in the mutton kebab prepared by moist cooking followed by charcoal grilling (T_1) on day 15 at refrigerated ($4 \pm 1^\circ\text{C}$) temperature. T_1 remains unaffected until day 10, while control and T_2 remains unaffected for the entire storage period of 15 days.

Table 13. Mean values of pH of mutton sheek kebabs influenced by different cooking methods at refrigerated (4±1°C) storage.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	6.12±0.19 ^{Aa}	6.17±0.02 ^{Aab}	6.22±0.01 ^{Abc}	6.27±0.01 ^{Ac}	6.19±0.01
T1	6.13±0.02 ^{Aa}	6.17±0.02 ^{Aab}	6.23±0.01 ^{Ab}	6.27±0.01 ^{Ac*}	6.20±0.01
T2	6.20±0.00 ^{Ba}	6.22±0.01 ^{Bb}	6.25±0.01 ^{Bc}	6.28±0.00 ^{Ad}	6.24±0.01
Overall mean of storage days	6.15±0.01	6.19±0.01	6.23±0.01	6.27±0.00	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.2 2- TBARS

The mean \pm SE 2- TBARS values for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4\pm 1^\circ\text{C}$) storage period are presented in Table 14 and analysis of variance in Table 28 and 29.

The mean \pm S.E 2- TBARS values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 0.20 ± 0.00 , 0.26 ± 0.01 , 0.31 ± 0.00 and 0.35 ± 0.00 respectively.

The mean \pm S.E 2- TBARS values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 0.19 ± 0.00 , 0.24 ± 0.00 , 0.28 ± 0.01 and 0.32 ± 0.00 respectively.

The mean \pm S.E 2- TBARS values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 0.20 ± 0.00 , 0.24 ± 0.01 , 0.30 ± 0.00 and 0.36 ± 0.01 respectively.

The overall mean \pm S.E 2- TBARS values of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 0.29 ± 0.01 , 0.26 ± 0.01 and 0.27 ± 0.01 respectively. The overall mean \pm S.E 2- TBARS values for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 0.20 ± 0.00 , 0.25 ± 0.01 , 0.30 ± 0.01 and 0.34 ± 0.01 respectively.

On all the storage days, T₁ group had lower 2-TBARS values than control (C) and T₂ groups. Analysis of variance revealed that there was no significant ($P>0.05$) difference in 2-TBARS values among the treatment and control groups and an increasing trend of TBARS values were noticed in all groups towards the end of storage period.

Spoilage was noticed in the mutton sheek kebab prepared by moist cooking followed by charcoal grilling (T₁) on day 15 at refrigerated ($4\pm 1^\circ\text{C}$) temperature.

Table 14. 2- TBARS (Mean± SE) (mg/kg malonaldehyde) values of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1 °C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	0.20±0.00 ^{Aa}	0.26±0.01 ^{Ab}	0.31±0.00 ^{Bc}	0.35±0.00 ^{Bd}	0.29±0.01
T1	0.19±0.00 ^{Aa}	0.24±0.00 ^{Ab}	0.28±0.01 ^{Ac}	0.32±0.00 ^{Ad*}	0.26±0.01
T2	0.20±0.00 ^{Aa}	0.24±0.01 ^{Ab}	0.30±0.00 ^{ABc}	0.36±0.01 ^{Bd}	0.27±0.01
Overall mean of storage days	0.20±0.00	0.25±0.01	0.30±0.00	0.34±0.01	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

Fig 6: Mean values of pH of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.

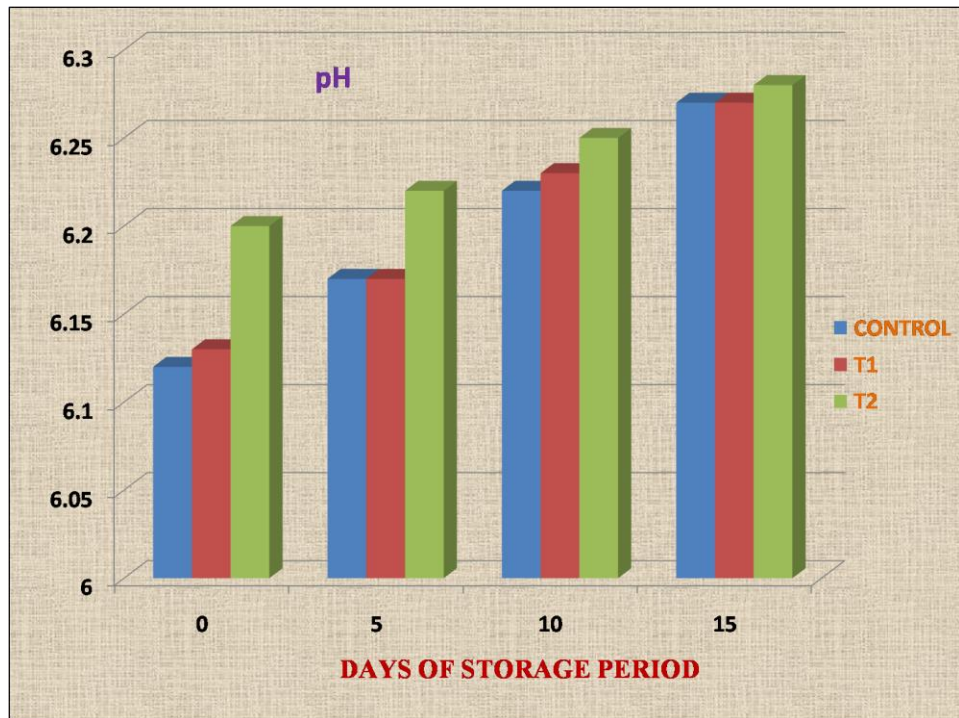
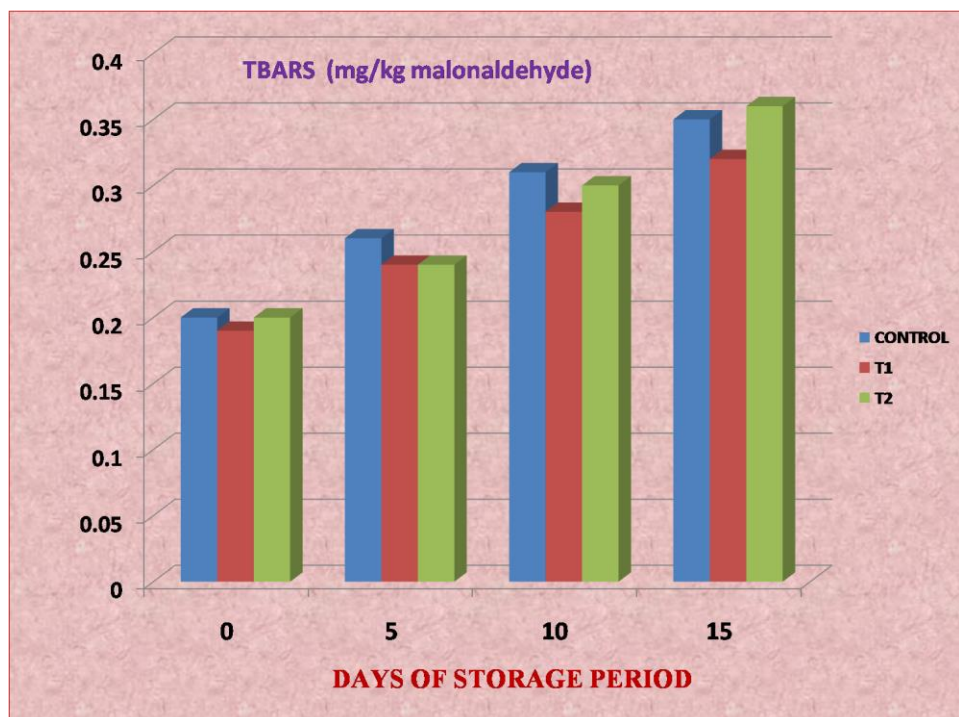


Fig 7: Mean values of TBARS (mg/kg malonaldehyde) of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.



4.2.3 Texture profile analysis:

The mean \pm S.E values of texture profile analysis of mutton sheek kebabs during refrigerated ($4\pm 1^\circ\text{C}$) storage period are given in Table 15 - 20 and analysis of variance in Table 28 and 29.

4.2.3.1 Chewiness

The mean \pm SE chewiness values for control (C), treatment 1 (T_1) and treatment 2 (T_2) group of mutton sheek kebabs during refrigerated ($4\pm 1^\circ\text{C}$) storage period are presented in Table 15 and analysis of variance in Table 28 and 29.

The mean \pm S.E chewiness values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 39.80 ± 0.68 , 39.06 ± 0.68 , 38.38 ± 0.82 and 37.96 ± 0.73 respectively.

The mean \pm S.E chewiness values for treatment 1 group kebabs (T_1) on 0, 5, 10 and 15th day of refrigerated storage were 35.02 ± 1.10 , 34.11 ± 0.98 , 33.68 ± 0.95 and 32.86 ± 1.00 respectively.

The mean \pm S.E chewiness values for treatment 2 group kebabs (T_2) on 0, 5, 10 and 15th day of refrigerated storage were 39.55 ± 0.30 , 39.11 ± 0.27 , 38.32 ± 0.26 and 37.66 ± 0.28 respectively.

The overall mean \pm S.E values for chewiness of mutton sheek kebab for control (C), treatment 1 (T_1) and treatment 2 (T_2) groups were 38.80 ± 0.37 , 33.92 ± 0.50 and 38.66 ± 0.20 respectively.

The overall mean \pm S.E. values of chewiness of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 38.13 ± 0.67 , 37.43 ± 0.69 , 36.79 ± 0.67 and 36.16 ± 0.69 respectively.

Analysis of variance revealed that there was no significant ($P>0.05$) difference in chewiness values for the control (C) and treatment T_1 group on all days of storage while T_2 showed no significant ($P>0.05$) difference until day 5.

Among the kebabs, treatment T₁ group had significantly (P<0.05) lower chewiness values followed by T₂ and control group on all the days of refrigerated storage.

Chewiness values revealed a slight decline from day 0 to 15th day of storage in the formulations.

Spoilage was noticed in the T₁ mutton sheek kebab prepared by moist cooking followed by charcoal grilling on 15th day at refrigerated ($4 \pm 1^{\circ}\text{C}$) temperature.

Table 15. Mean chewiness scores (N/cm) of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	39.80±0.68 ^{Ba}	39.06±0.68 ^{Ba}	38.38±0.82 ^{Ba}	37.96±0.73 ^{Ba}	38.80±0.37
T1	35.02±1.10 ^{Aa}	34.11±0.98 ^{Aa}	33.68±0.95 ^{Aa}	32.86±1.00 ^{Aa*}	33.92±0.50
T2	39.55±0.30 ^{Bc}	39.11±0.27 ^{Bbc}	38.32±0.26 ^{Bab}	37.66±0.28 ^{Ba}	38.66±0.20
Overall mean of storage days	38.13±0.67	37.43±0.69	36.79±0.67	36.16±0.69	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.3.2 Cohesiveness

The mean \pm SE cohesiveness values for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 16 and analysis of variance in Table 28 and 29.

The mean \pm S.E cohesiveness values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 0.47 ± 0.17 , 0.45 ± 0.01 , 0.43 ± 0.01 and 0.42 ± 0.01 respectively. The mean \pm S.E cohesiveness values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 0.42 ± 0.01 , 0.39 ± 0.00 , 0.38 ± 0.01 and 0.37 ± 0.01 respectively. The mean \pm S.E cohesiveness values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 0.39 ± 0.00 , 0.38 ± 0.01 , 0.36 ± 0.01 and 0.35 ± 0.01 respectively.

The overall mean \pm S.E cohesiveness values of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 0.44 ± 0.01 , 0.39 ± 0.01 and 0.37 ± 0.01 respectively. The overall mean \pm S.E. cohesiveness values of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 0.43 ± 0.01 , 0.41 ± 0.01 , 0.39 ± 0.01 and 0.38 ± 0.01 respectively.

Analysis of variance revealed that there was no significant ($P>0.05$) difference in cohesiveness values for the control (C) for all days of storage while both the treatments T₁ and T₂ showed the same until day 5. Irrespective of the groups, a gradual decline in cohesiveness values was noticed, as the storage period advanced. On all the storage days, T₂ group had lower cohesiveness values than control (C) and T₁ group.

Table 16. Mean of cohesiveness scores (ratio) of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	0.47±0.17 ^{Ba}	0.45±0.01 ^{Ba}	0.43±0.01 ^{Ba}	0.42±0.01 ^{Ba}	0.44±0.01
T1	0.42±0.01 ^{ABb}	0.39±0.00 ^{Aab}	0.38±0.01 ^{Aa}	0.37±0.01 ^{Aa*}	0.39±0.01
T2	0.39±0.00 ^{Ab}	0.38±0.01 ^{Aab}	0.36±0.01 ^{Aa}	0.35±0.01 ^{Aa}	0.37±0.01
Overall mean of storage days	0.43±0.01	0.41±0.01	0.39±0.01	0.38±0.01	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.3.3 Fracturability (N)

The mean \pm SE fracturability values for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs at during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 17 and analysis of variance in Table 28 and 29.

The mean \pm S.E fracturability values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 45.30 ± 0.45 , 44.39 ± 0.43 , 43.71 ± 0.44 and 42.41 ± 0.61 respectively. The mean \pm S.E fracturability values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 33.34 ± 0.78 , 32.57 ± 0.71 , 31.88 ± 0.71 and 30.92 ± 0.54 respectively. The mean \pm S.E fracturability values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 43.00 ± 0.22 , 42.00 ± 0.22 , 41.17 ± 0.34 and 40.41 ± 0.34 respectively.

The overall mean \pm S.E fracturability values of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 43.95 ± 0.32 , 32.18 ± 0.37 and 41.64 ± 0.24 respectively. The overall mean \pm S.E fracturability values for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 40.55 ± 1.29 , 39.65 ± 1.27 , 38.92 ± 1.27 and 37.91 ± 1.25 respectively.

Among the kebabs, treatment T₁ group had significantly ($P < 0.05$) lower fracturability values followed by treatment T₂ and control (C) group for all the days of refrigerated storage. No significant difference in T₁ during the entire storage period but the control showed the same until day 10. Fracturability values showed a gradual decline towards the end of storage period in all groups.

Table 17. Mean fracturability values (N) of mutton sheek kebab as influenced by different cooking method, stored at refrigeration ($4\pm 1^{\circ}\text{C}$) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	45.30 \pm 0.45 ^{Cb}	44.39 \pm 0.43 ^{Cb}	43.71 \pm 0.44 ^{Cab}	42.41 \pm 0.61 ^{Ca}	43.95\pm0.32
T1	33.34 \pm 0.78 ^{Aa}	32.57 \pm 0.71 ^{Aa}	31.88 \pm 0.71 ^{Aa}	30.92 \pm 0.54 ^{Aa*}	32.18\pm0.37
T2	43.00 \pm 0.22 ^{Bc}	42.00 \pm 0.22 ^{Bbc}	41.17 \pm 0.34 ^{Bab}	40.41 \pm 0.34 ^{Ba}	41.64\pm0.24
Overall mean of storage days	40.55\pm1.29	39.65\pm1.27	38.92\pm1.27	37.91\pm1.25	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different ($P < 0.05$). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.3.4 Gumminess

The mean \pm SE gumminess values for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 18 and analysis of variance in Table 28 and 29.

The mean \pm S.E gumminess values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 22.91 ± 0.30 , 22.45 ± 0.42 , 21.74 ± 0.41 and 21.00 ± 0.27 respectively. The mean \pm S.E gumminess values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 24.02 ± 0.53 , 23.46 ± 0.51 , 23.00 ± 0.58 and 22.46 ± 0.53 respectively. The mean \pm S.E gumminess values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 25.07 ± 0.20 , 24.45 ± 0.18 , 23.68 ± 0.19 and 23.05 ± 0.22 respectively.

The overall mean \pm S.E gumminess values of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 22.02 ± 0.22 , 23.24 ± 0.28 and 24.06 ± 0.18 respectively. The overall mean \pm S.E. gumminess values for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 24.00 ± 0.29 , 23.45 ± 0.29 , 22.80 ± 0.30 and 22.17 ± 0.29 respectively.

On all the storage days, treatment T₂ group had significantly ($P < 0.05$) higher gumminess values than control group and the same were similar with T₁ group. Irrespective of groups, the gumminess values showed a declining trend towards the end of storage period. No significant difference in gumminess values was noticed in treatment T₁ during the entire storage period while the control showed the same until day 10.

Table 18. Mean gumminess scores (N/cm²) of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	22.91±0.30 ^{Ab}	22.45±0.42 ^{Ab}	21.74±0.41 ^{Aab}	21.00±0.27 ^{Aa}	22.02±0.22
T1	24.02±0.53 ^{ABa}	23.46±0.51 ^{ABa}	23.00±0.58 ^{ABa}	22.46±0.53 ^{Ba*}	23.24±0.28
T2	25.07±0.20 ^{Bc}	24.45±0.18 ^{Bbc}	23.68±0.19 ^{Bab}	23.05±0.22 ^{Ba}	24.06±0.18
Overall mean of storage days	24.00±0.29	23.45±0.29	22.80±0.30	22.17±0.29	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

Fig 8: Mean fracturability (N) values of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.

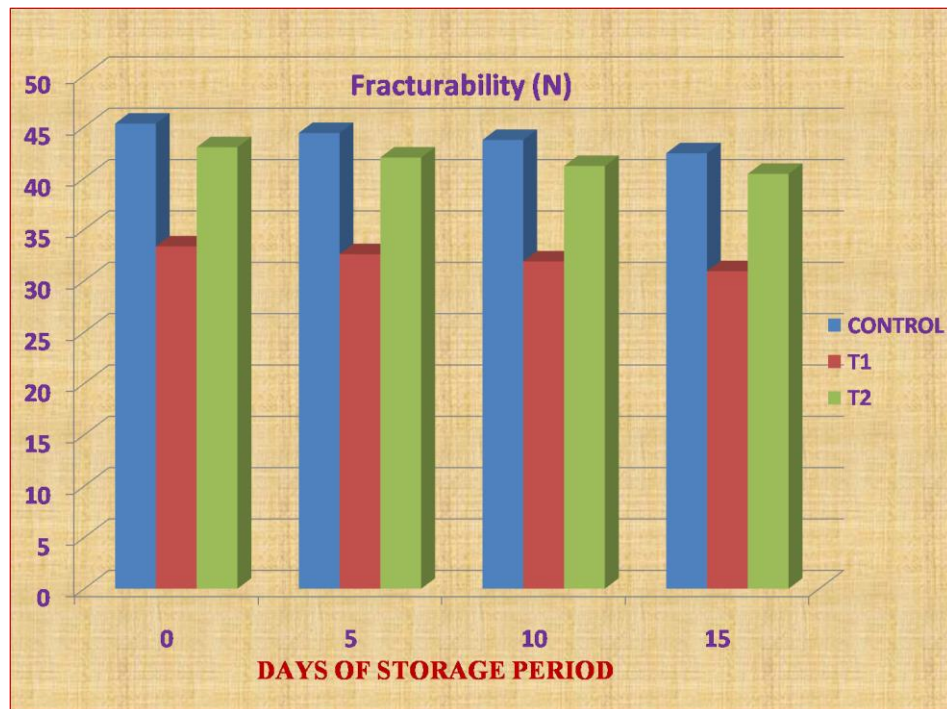
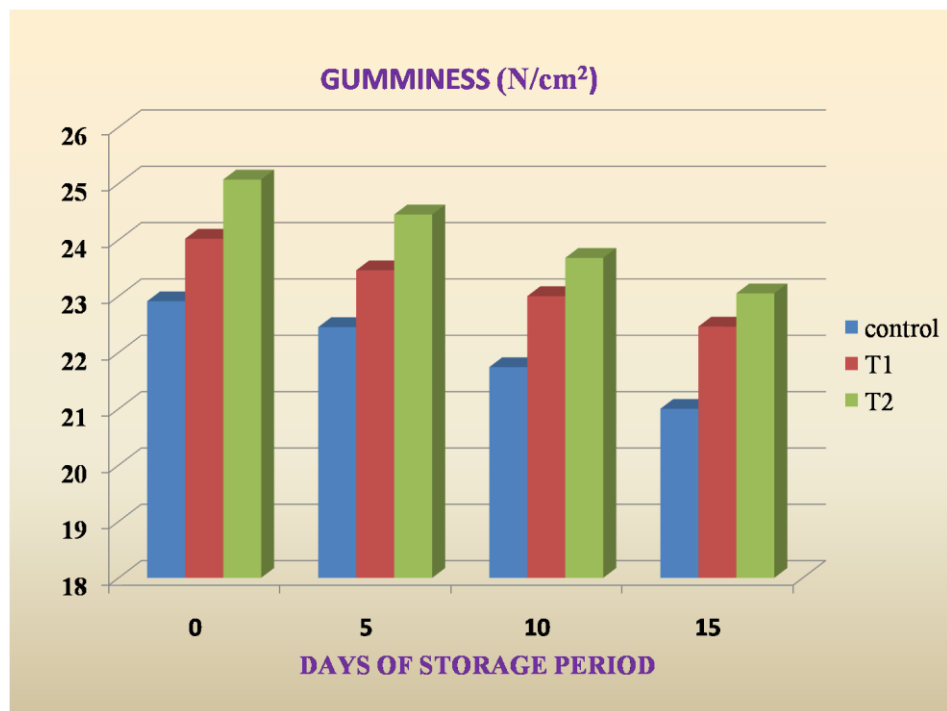


Fig 9: Mean gumminess (N/cm^2) values of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.



4.2.3.5 Hardness

The mean \pm SE hardness values for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 19 and analysis of variance in Table 28 and 29.

The mean \pm S.E hardness values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 57.20 ± 0.55 , 55.88 ± 0.62 , 55.13 ± 0.55 and 54.59 ± 0.45 respectively. The mean \pm S.E hardness values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 47.82 ± 0.43 , 47.06 ± 0.31 , 46.33 ± 0.26 and 45.67 ± 0.29 respectively. The mean \pm S.E hardness values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 51.00 ± 0.48 , 50.63 ± 0.46 , 49.90 ± 0.36 and 49.29 ± 0.36 respectively.

The overall mean \pm S.E hardness values of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 55.70 ± 0.33 , 46.72 ± 0.22 and 50.20 ± 0.24 respectively. The overall mean \pm S.E hardness values for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 52.01 ± 0.98 , 51.19 ± 0.92 , 50.45 ± 0.90 and 49.85 ± 0.91 respectively.

Among the kebabs, treatment T₁ group had significantly ($P < 0.05$) lower hardness values followed by treatment T₂ and control (C) on all the storage days.

A gradual decline in hardness values was evident as the storage period advanced in all the groups. No significant difference for hardness values in control and T₂ until day 10 while the same for T₁ until day 5.

Table 19. Mean hardness scores (N/cm²) of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	57.20±0.55 ^{Cb}	55.88±0.62 ^{Cab}	55.13±0.55 ^{Cab}	54.59±0.45 ^{Ca}	55.70±0.33
T1	47.82±0.43 ^{Ac}	47.06±0.31 ^{Abc}	46.33±0.26 ^{Aab}	45.67±0.29 ^{Aa*}	46.72±0.22
T2	51.00±0.48 ^{Bb}	50.63±0.46 ^{Bab}	49.90±0.36 ^{Bab}	49.29±0.36 ^{Ba}	50.20±0.24
Overall mean of storage days	52.01±0.98	51.19±0.92	50.45±0.90	49.85±0.91	

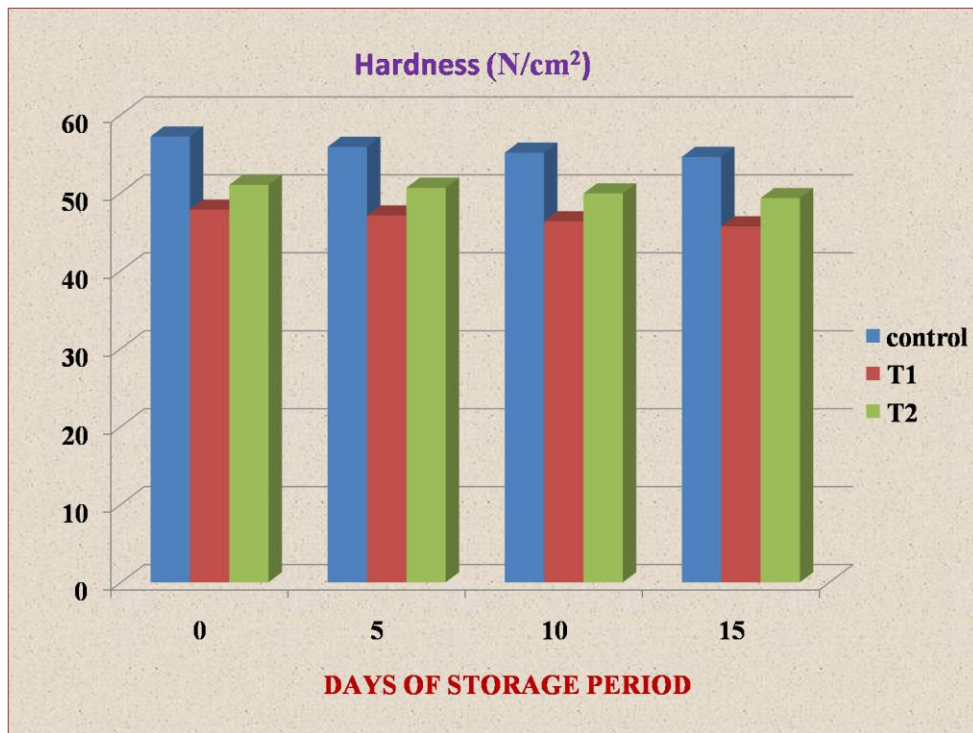
NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling.

Fig 10: Mean hardness (N/cm²) values of mutton sheek kebabs influenced by different cooking methods at refrigerated (4±1°C) storage.



4.2.3.6 Springiness

The mean \pm SE springiness values for control (C), treatment 1 (T_1) and treatment 2 (T_2) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 20 and analysis of variance in Table 28 and 29.

The mean \pm S.E springiness values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 1.53 ± 0.03 , 1.48 ± 0.03 , 1.43 ± 0.04 and 1.41 ± 0.03 respectively. The mean \pm S.E springiness values for treatment 1 group kebabs (T_1) on 0, 5, 10 and 15th day of refrigerated storage were 1.51 ± 0.04 , 1.46 ± 0.03 , 1.42 ± 0.04 and 1.36 ± 0.04 respectively. The mean \pm S.E springiness values for treatment 2 group kebabs (T_2) on 0, 5, 10 and 15th day of refrigerated storage were 1.45 ± 0.01 , 1.42 ± 0.02 , 1.36 ± 0.01 and 1.32 ± 0.01 respectively.

The overall mean \pm S.E springiness values of mutton sheek kebab for control (C), treatment 1 (T_1) and treatment 2 (T_2) groups were 1.46 ± 0.01 , 1.44 ± 0.02 and 1.39 ± 0.01 respectively. The overall mean \pm S.E. springiness values for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 1.50 ± 0.01 , 1.45 ± 0.02 , 1.41 ± 0.02 and 1.36 ± 0.02 respectively.

On all the storage days, no significant difference ($P>0.05$) noticed among the control (C), treatment T_1 and treatment T_2 groups.

Mean springiness values of mutton sheek kebab showed a declining trend in all the groups towards the end of storage life and no significant difference ($P>0.05$) in springiness values of control and treatment T_1 groups on entire storage period, but T_2 showed the same until day 5.

Table 20. Mean springiness (cm) scores of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	1.53±0.03 ^{Aa}	1.48±0.03 ^{Aa}	1.43±0.04 ^{Aa}	1.41±0.03 ^{Aa}	1.46±0.01
T1	1.51±0.04 ^{Aa}	1.46±0.03 ^{Aa}	1.42±0.04 ^{Aa}	1.36±0.04 ^{Aa*}	1.44±0.02
T2	1.45±0.01 ^{Ab}	1.42±0.02 ^{Ab}	1.36±0.01 ^{Aa}	1.32±0.01 ^{Aa}	1.39±0.01
Overall mean of storage days	1.50±0.01	1.45±0.02	1.40±0.02	1.36±0.02	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.4 SENSORY EVALUATION:

4.2.4.1 Appearance

The mean \pm SE appearance score for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^{\circ}\text{C}$) storage period are presented in Table 21 and analysis of variance in Table 28 and 29.

The mean \pm S.E appearance score of mutton sheek kebab for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 6.60 ± 0.04 , 6.41 ± 0.02 , 6.08 ± 0.02 and 5.90 ± 0.01 respectively. The mean \pm S.E appearance score values for treatment 1 group kebabs (T₁) on 0, 5, 10th day of refrigerated storage were 7.51 ± 0.04 , 7.38 ± 0.04 and 7.22 ± 0.06 respectively. The mean \pm S.E appearance score for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 6.75 ± 0.02 , 6.51 ± 0.04 , 6.26 ± 0.03 and 6.06 ± 0.02 respectively.

The overall mean \pm S.E appearance score of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 6.25 ± 0.06 , $7.36 \pm 0.05^*$ and 6.30 ± 0.06 respectively. The overall mean \pm S.E appearance score for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 6.95 ± 0.10 , 6.76 ± 0.10 , 6.52 ± 0.12 and $5.98 \pm 0.12^*$ respectively.

Analysis of variance revealed that there was a significant difference ($p < 0.05$) in appearance score in all the groups up to 15 days of storage. During the storage period, T₁ revealed a significantly ($P < 0.05$) higher appearance scores compared to control and treatment T₂ groups. The appearance score revealed a slight decline from day 0 to 15th day of storage in control and all the treatments. Spoilage was noticed in the T₁ mutton sheek kebab prepared by moist cooking followed by charcoal grilling on 15th day at refrigerated ($4 \pm 1^{\circ}\text{C}$) temperature.

Table 21. Appearance (Mean± SE) scores of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	6.60±0.04 ^{Ad}	6.41±0.02 ^{Ac}	6.08±0.02 ^{Ab}	5.90±0.01 ^{Aa}	6.25±0.06
T1	7.51±0.04 ^{Bc}	7.38±0.04 ^{Bb}	7.22±0.06 ^{Ba}	----#	7.36±0.05*
T2	6.75±0.02 ^{Ad}	6.51±0.04 ^{Ac}	6.26±0.03 ^{Ab}	6.06±0.02 ^{Aa}	6.39±0.05
Overall mean of storage days	6.95±0.10	6.76±0.10	6.52±0.12	5.98±0.12*	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (#) indicates spoiled sample. (*) indicates overall mean excluding spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.4.2 Flavor

The mean \pm SE flavor scores for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 22 and analysis of variance in Table 28 and 29.

The mean \pm S.E flavor score for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 7.10 ± 0.45 , 6.74 ± 0.12 , 6.04 ± 0.10 and 5.47 ± 0.08 respectively. The mean \pm S.E flavor score for treatment 1 group kebabs (T₁) on 0, 5, 10th day of refrigerated storage were 6.93 ± 0.04 , 6.77 ± 0.07 and 6.01 ± 0.20 respectively. The mean \pm S.E flavor score for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 6.99 ± 0.06 , 6.69 ± 0.06 , 5.87 ± 0.04 and 5.34 ± 0.07 respectively.

The overall mean \pm S.E flavor score of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 6.34 ± 0.14 , $6.56 \pm 0.22^*$ and 6.22 ± 0.14 respectively. The overall mean \pm S.E. flavor score for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 7.01 ± 0.03 , 6.73 ± 0.05 , 5.97 ± 0.07 and $5.40 \pm 0.12^*$ respectively.

Analysis of variance revealed a significant difference ($p < 0.05$) in flavor score in control and T₂ during the entire storage period, while similar in T₁ was noticed, after day 5 of refrigerated storage.

On all the storage days, control (C) group had higher flavor values followed by T₂ and T₁ groups. Irrespective of all the groups, a gradual decline in flavor scores was observed as storage period advanced.

Table 22. Flavor (Mean± SE) scores of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatment	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	7.10±0.45 ^{Bd}	6.74±0.12 ^{Ac}	6.04±0.10 ^{Ab}	5.47±0.08 ^{Aa}	6.34±0.14
T1	6.93±0.04 ^{Ab}	6.77±0.07 ^{Ab}	6.01±0.20 ^{Aa}	----#	6.56±0.22*
T2	6.99±0.06 ^{ABd}	6.69±0.06 ^{Ac}	5.87±0.04 ^{Ab}	5.34±0.07 ^{Aa}	6.22±0.14
Overall mean of storage days	7.01±0.03	6.73±0.05	5.97±0.07	5.40±0.12*	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (#) indicates spoiled sample. (*) indicates overall mean excluding spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

Fig 11: Mean appearance scores of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^{\circ}\text{C}$) storage.

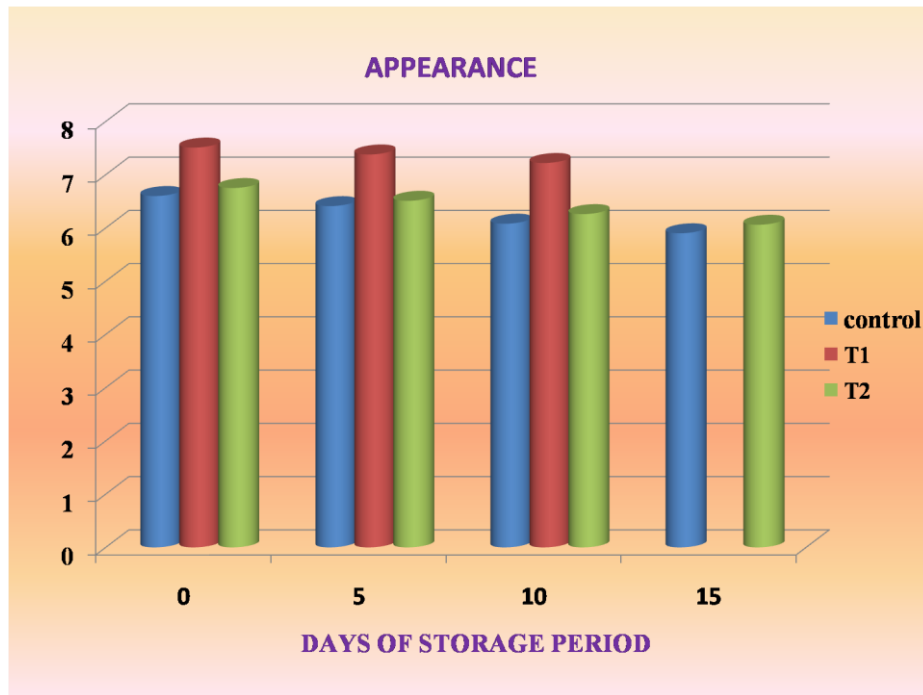
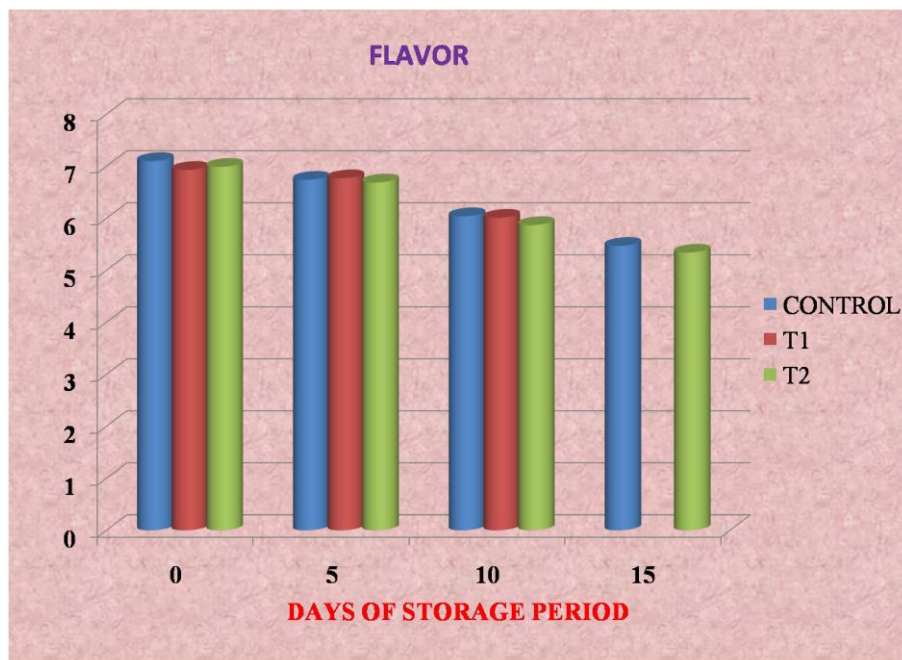


Fig 12: Mean flavor scores of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^{\circ}\text{C}$) storage.



4.2.4.3 Juiciness

The mean \pm SE juiciness score for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 23 and analysis of variance in Table 28 and 29.

The mean \pm S.E juiciness score for control group kebabs (C) in 0, 5, 10 and 15th day of refrigerated storage were 6.74 ± 0.06 , 6.39 ± 0.05 , 5.92 ± 0.17 and 5.62 ± 0.16 respectively. The mean \pm S.E juiciness score for treatment 1 group kebabs (T₁) in 0, 5 and 10th day of refrigerated storage were 7.43 ± 0.07 , 7.03 ± 0.14 and 6.30 ± 0.13 respectively. The mean \pm S.E juiciness score for treatment 2 group kebabs (T₂) in 0, 5, 10 and 15th day of refrigerated storage were 6.72 ± 0.03 , 6.37 ± 0.02 , 5.86 ± 0.10 and 5.61 ± 0.10 respectively.

The overall mean \pm S.E juiciness score of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) were 6.17 ± 0.08 , $6.92\pm 0.17^*$ and 6.14 ± 0.09 respectively. The overall mean \pm S.E. juiciness score of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 6.97 ± 0.09 , 6.60 ± 0.09 , 6.02 ± 0.09 and $5.61\pm 0.12^*$ respectively.

Among the kebabs, treatment T₁ group had significantly ($P < 0.05$) higher juiciness score than control and T₂ groups except on day 15.

Analysis of variance revealed a significant difference ($p < 0.05$) in juiciness score was noticed in both the control and T₂ until day 5, while T₁ showed significant difference from day 5 to the end of storage period. The juiciness score of mutton sheek kebabs were decreased towards the end of storage period.

Table 23. Mean juiciness scores of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatment	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	6.74±0.06 ^{Ad}	6.39±0.05 ^{Abc}	5.92±0.17 ^{Bab}	5.62±0.16 ^{Aa}	6.17±0.08
T1	7.43±0.07 ^{Bb}	7.03±0.14 ^{Bb}	6.30±0.13 ^{Aa}	-----#	6.92±0.17*
T2	6.72±0.03 ^{Ac}	6.37±0.02 ^{Ab}	5.86±0.10 ^{Ba}	5.61±0.10 ^{Aa}	6.14±0.09
Overall mean of storage days	6.97±0.09	6.60±0.09	6.02±0.09	5.61±0.12*	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (#) indicates spoiled sample. (*) indicates overall mean excluding spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.4.4 Texture

The mean \pm SE texture scores for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 24 and analysis of variance in Table 28 and 29.

The mean \pm S.E texture scores for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 6.60 ± 0.05 , 6.16 ± 0.04 , 5.21 ± 0.10 and 4.98 ± 0.02 respectively. The mean \pm S.E texture scores for treatment 1 group kebabs (T₁) on 0, 5 and 10th day of refrigerated storage were 7.22 ± 0.04 , 7.00 ± 0.08 and 6.47 ± 0.12 respectively. The mean \pm S.E texture scores for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 6.72 ± 0.05 , 6.35 ± 0.01 , 5.92 ± 0.04 and 5.40 ± 0.05 respectively.

The overall mean \pm S.E texture scores of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 5.73 ± 0.14 , $6.89\pm 0.19^*$ and 6.10 ± 0.10 respectively. The overall mean \pm S.E texture scores for mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 6.85 ± 0.07 , 6.50 ± 0.09 , 5.87 ± 0.14 and $5.19\pm 0.06^*$ respectively.

Among the kebabs, treatment T₁ group had significantly ($P<0.05$) higher texture scores followed by T₂ and control groups except on day 15 of storage period.

The texture scores of mutton sheek kebab were decreased towards the end of storage period. Significant difference ($p<0.05$) of texture score was noticed in T₂ during the entire storage period while the same noticed in control until day 10.

Table 24. Mean texture scores of mutton sheek kebab as influenced by different cooking method, stored at refrigeration ($4\pm 1^{\circ}\text{C}$) temperature.

Treatments	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	6.60 \pm 0.05 ^{Ac}	6.16 \pm 0.04 ^{Ab}	5.21 \pm 0.10 ^{Aa}	4.98 \pm 0.02 ^{Aa}	5.73\pm0.14
T1	7.22 \pm 0.04 ^{Bb}	7.00 \pm 0.08 ^{Cb}	6.47 \pm 0.12 ^{Ca}	-----#	6.89\pm0.19*
T2	6.72 \pm 0.05 ^{Ad}	6.35 \pm 0.01 ^{Bc}	5.92 \pm 0.04 ^{Bb}	5.40 \pm 0.05 ^{Aa}	6.10\pm0.10
Overall mean of storage days	6.85\pm0.07	6.50\pm0.09	5.87\pm0.14	5.19\pm0.06*	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different ($P < 0.05$). (#) indicates spoiled sample. (*) indicates overall mean excluding spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

Fig 13: Mean juiciness scores of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.

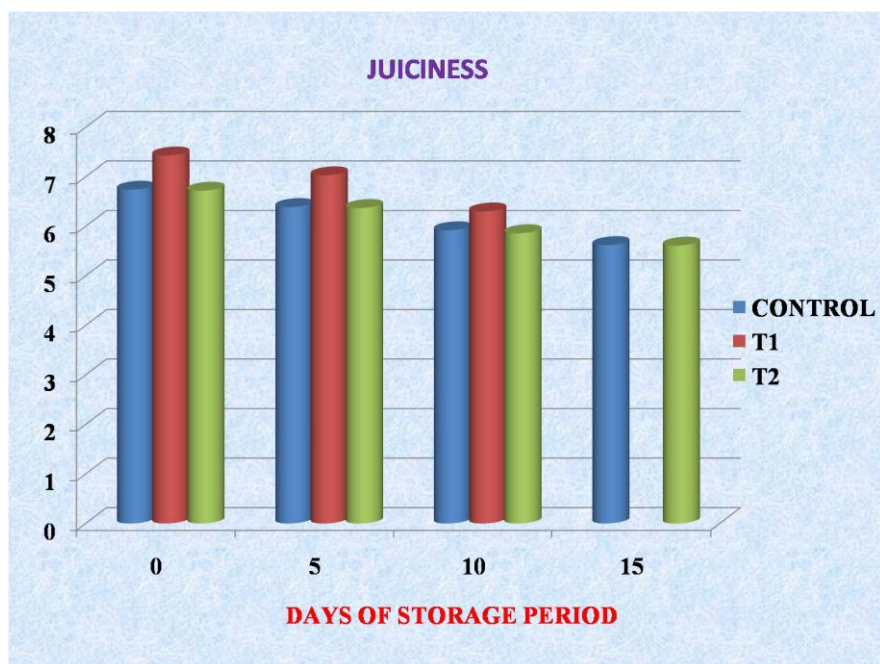
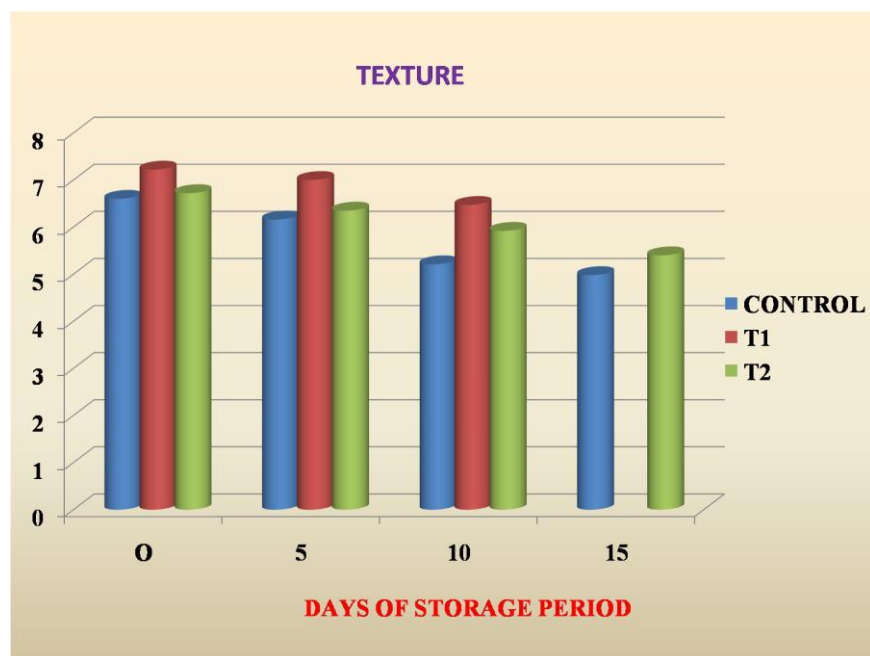


Fig 14: Mean texture scores of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.



4.2.4.5 Overall acceptability

The mean \pm SE overall acceptability scores for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 25 and analysis of variance in Table 28 and 29.

The mean \pm S.E overall acceptability scores for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 6.61 ± 0.05 , 6.19 ± 0.03 , 5.59 ± 0.12 and 5.05 ± 0.02 respectively. The mean \pm S.E overall acceptability scores for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 7.3 ± 0.04 , 6.92 ± 0.06 , 6.12 ± 0.06 and 4.91 ± 0.08 respectively. The mean \pm S.E overall acceptability scores for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 6.82 ± 0.03 , 6.37 ± 0.03 , 5.81 ± 0.07 and 5.43 ± 0.07 respectively.

The overall mean \pm S.E overall acceptability scores of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 5.86 ± 0.13 , $6.78 \pm 0.22^*$ and 6.11 ± 0.11 respectively. The overall mean \pm S.E. acceptability scores of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 6.91 ± 0.07 , 6.49 ± 0.08 , 5.84 ± 0.07 and $5.24 \pm 0.12^*$ respectively.

On all days of storage, control (C) had the lowest score for overall acceptability among all the groups and between treatments, T₁ group had significantly ($P < 0.05$) higher overall acceptability score, except on day 15 of storage period.

A gradual decline in overall acceptability scores was evident in all treatments and control group of mutton sheek kebabs as the storage period advanced. All treatments and control group are significantly ($P < 0.05$) different from each other during entire storage period.

Table 25. Mean overall acceptability scores of mutton sheek kebab as influenced by different cooking method, stored at refrigeration (4±1°C) temperature.

Treatment	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	6.61±0.05 ^{Ad}	6.19±0.03 ^{Ac}	5.59±0.12 ^{Ab}	5.05±0.02 ^{Aa}	5.86±0.13
T1	7.3±0.04 ^{Cc}	6.92±0.06 ^{Cb}	6.12±0.06 ^{Ba}	-----#	6.78±0.22*
T2	6.82±0.03 ^{Bd}	6.37±0.03 ^{Bc}	5.81±0.07 ^{ABb}	5.43±0.07 ^{Ba}	6.11±0.11
Overall mean of storage days	6.91±0.07	6.49±0.08	5.84±0.07	5.24±0.06*	

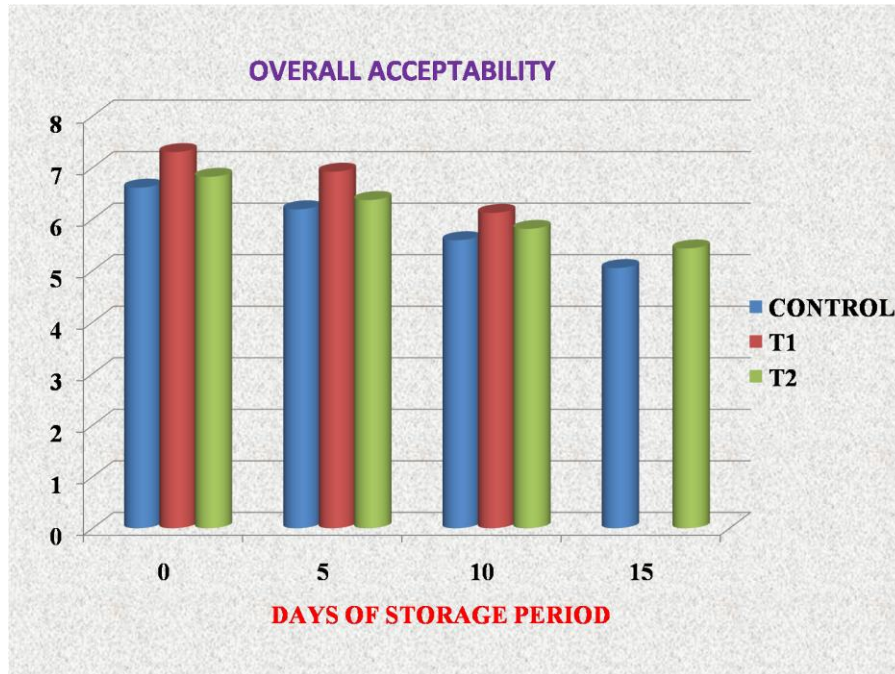
NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different (P<0.05). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

Fig 15: Mean overall acceptability scores of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^{\circ}\text{C}$) storage.



4.2.5 MICROBIAL ANALYSIS

4.2.5.1 Total plate count

The mean \pm SE total plate count values of for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 26 and analysis of variance in Table 28 and 29.

The mean \pm S.E total plate count values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 1.94 ± 0.01 , 2.21 ± 0.02 , 2.41 ± 0.01 and 2.55 ± 0.00 respectively.

The mean \pm S.E total plate count values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 2.10 ± 0.01 , 2.35 ± 0.01 , 2.53 ± 0.01 and 2.63 ± 0.00 respectively.

The mean \pm S.E total plate count values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 2.03 ± 0.00 , 2.23 ± 0.01 , 2.44 ± 0.01 and 2.57 ± 0.00 respectively.

The overall mean \pm S.E values of total plate count of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 2.28 ± 0.05 , 2.40 ± 0.42 and 2.34 ± 0.04 respectively. The overall mean \pm S.E. values of total plate count of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 2.02 ± 0.02 , 2.26 ± 0.02 , 2.46 ± 0.01 and 2.58 ± 0.01 respectively.

Among the kebabs, the total plate count values of control (c) were significantly ($P < 0.05$) lower, followed by treatment T₂ and treatment T₁ groups during the entire storage period.

Irrespective of all the treatments and control group, the total plate count values of mutton sheek kebabs were significantly ($P<0.05$) increased with increasing storage period.

4.2.5.2 Psychrophilic Counts

There was no psychrophilic bacterial growth observed in any of the treatments along with control during the entire refrigerated ($4\pm 1^{\circ}\text{C}$) storage of mutton sheek kebabs.

Table 26. Total plate count (\log_{10} Cfug of sample) (Mean \pm SE) values of mutton sheek kebab as influenced by different cooking method, stored at refrigeration ($4\pm 1^{\circ}\text{C}$) temperature

Treatment	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	1.94 \pm 0.01 ^{Aa}	2.21 \pm 0.02 ^{Ab}	2.41 \pm 0.01 ^{Ac}	2.55 \pm 0.00 ^{Ad}	2.28\pm0.05
T1	2.10 \pm 0.01 ^{Ca}	2.35 \pm 0.01 ^{Bb}	2.53 \pm 0.01 ^{Bc}	2.63 \pm 0.00 ^{Cd*}	2.40\pm0.42
T2	2.03 \pm 0.00 ^{Ba}	2.23 \pm 0.01 ^{Ab}	2.44 \pm 0.01 ^{Ac}	2.57 \pm 0.00 ^{Ad}	2.32\pm0.04
Overall mean of storage days	2.02\pm0.02	2.26\pm0.02	2.46\pm0.01	2.58\pm0.01	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different ($P < 0.05$). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling

4.2.5.3 Yeast and mould count

The mean \pm SE yeast and mould count values for control (C), treatment 1 (T₁) and treatment 2 (T₂) group of mutton sheek kebabs during refrigerated ($4 \pm 1^\circ\text{C}$) storage period are presented in Table 27 and analysis of variance in Table 28 and 29.

The mean \pm S.E yeast and mould count values for control group kebabs (C) on 0, 5, 10 and 15th day of refrigerated storage were 1.88 ± 0.01 , 2.18 ± 0.01 , 2.35 ± 0.01 and 2.43 ± 0.01 respectively. The mean \pm S.E yeast and mould count values for treatment 1 group kebabs (T₁) on 0, 5, 10 and 15th day of refrigerated storage were 2.05 ± 0.01 , 2.24 ± 0.01 , 2.42 ± 0.01 and 2.52 ± 0.01 respectively. The mean \pm S.E yeast and mould count values for treatment 2 group kebabs (T₂) on 0, 5, 10 and 15th day of refrigerated storage were 1.95 ± 0.01 , 2.18 ± 0.01 , 2.36 ± 0.01 and 2.45 ± 0.01 respectively.

The overall mean \pm S.E yeast and mould count values of mutton sheek kebab for control (C), treatment 1 (T₁) and treatment 2 (T₂) groups were 2.21 ± 0.04 , 2.31 ± 0.03 and 2.24 ± 0.04 respectively. The overall mean \pm S.E yeast and mould count values of mutton sheek kebab on 0, 5, 10 and 15 days of refrigerated storage were 1.96 ± 0.02 , 2.20 ± 0.01 , 2.38 ± 0.01 and 2.47 ± 0.01 respectively.

On all the storage days, the yeast and mould count values of mutton sheek kebabs were significantly ($P < 0.05$) increased with increasing storage period. Among the kebabs, the yeast and mould count values of control (c) are significantly ($P < 0.05$) lower, followed by treatment T₂ and treatment T₁ groups during the entire storage period.

Table.27 Yeast and mould count (\log_{10} CfU/g of sample) (Mean \pm SE) values of mutton sheek kebab as influenced by different cooking method, stored at refrigeration ($4\pm 1^{\circ}\text{C}$) temperature

Treatment	Storage period (days)				Overall mean of treatments
	0	5	10	15	
Control	1.88 \pm 0.01 ^{Aa}	2.18 \pm 0.01 ^{Ab}	2.35 \pm 0.01 ^{Ac}	2.43 \pm 0.01 ^{Ad}	2.21\pm0.04
T1	2.05 \pm 0.01 ^{Ca}	2.24 \pm 0.01 ^{Bb}	2.42 \pm 0.01 ^{Bc}	2.52 \pm 0.01 ^{Bd*}	2.31\pm0.03
T2	1.95 \pm 0.01 ^{Ba}	2.18 \pm 0.01 ^{Ab}	2.36 \pm 0.01 ^{Ac}	2.45 \pm 0.01 ^{Ad}	2.24\pm0.04
Overall mean of storage days	1.96\pm0.02	2.20\pm0.01	2.38\pm0.01	2.47\pm0.01	

NOTE: Means bearing different superscript between rows (lowercase alphabets) and between columns (Uppercase alphabets) are significantly different ($P < 0.05$). (*) indicates spoiled sample.

Control: Direct charcoal grilling,

Treatment -1: Moist cooking followed by charcoal grilling

Treatment -2: Hot air oven cooking followed by charcoal grilling.

Fig 16: Mean total plate count values ($\log_{10}\text{cfu/g}$ of sample) of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.

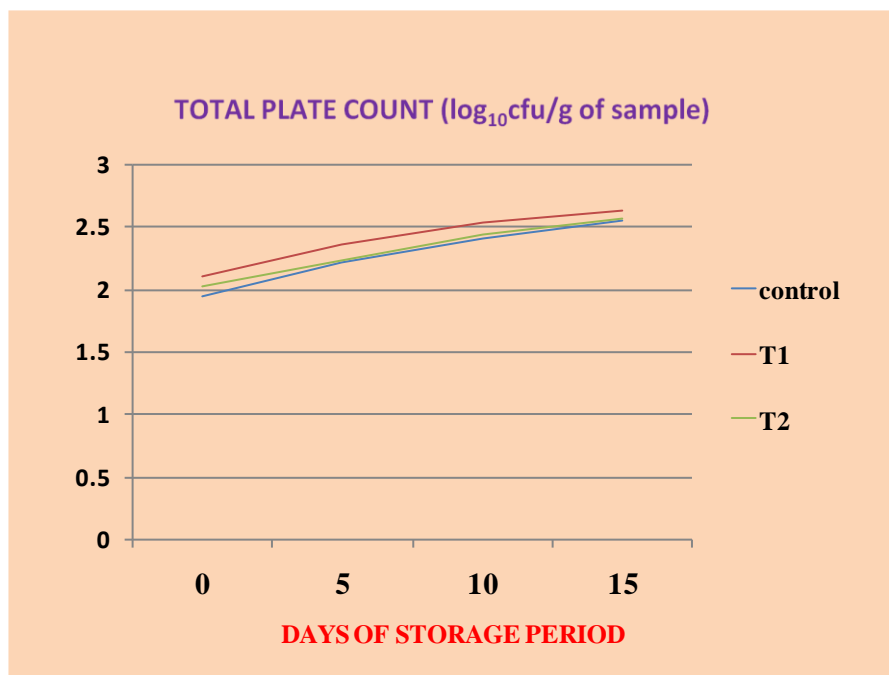


Fig 17: Mean yeast and mould count values ($\log_{10}\text{cfu/g}$ of sample) of mutton sheek kebabs influenced by different cooking methods at refrigerated ($4\pm 1^\circ\text{C}$) storage.

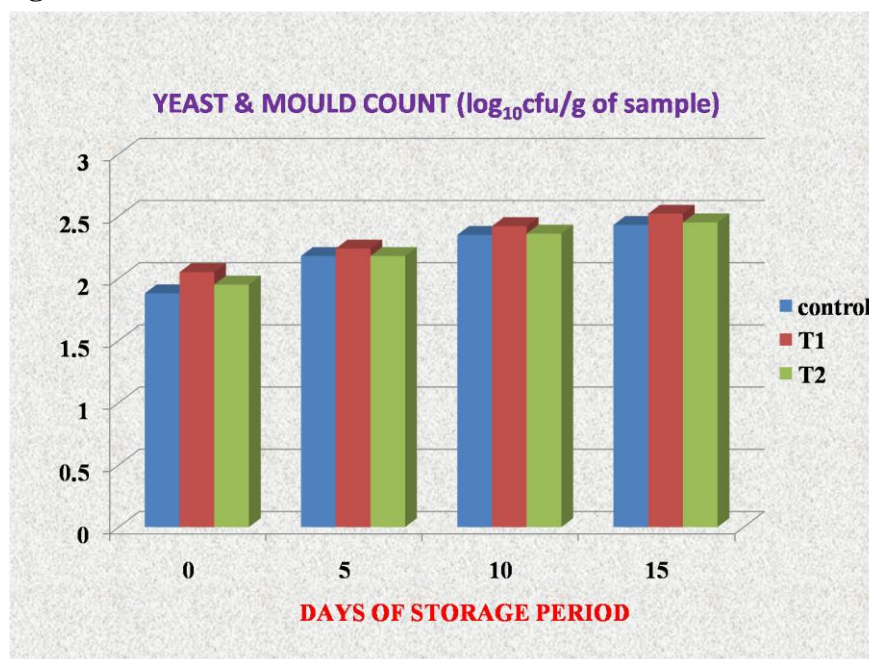


Table no. 28 Analysis of variance (ANOVA) for physico chemical and microbial parameters of mutton sheek kebab as influenced different cooking methods.

ANOVA							
	CM		Sum of Squares	df	Mean Square	F	Sig.
pH	C	Between Groups	0.074	3	0.025	17.038	0.000*
		Within Groups	0.029	20	0.001		
		Total	0.102	23			
	T	Between Groups	0.070	3	0.023	20.771	0.000*
		Within Groups	0.022	20	0.001		
		Total	0.092	23			
	H	Between Groups	0.028	3	0.009	44.255	0.000*
		Within Groups	0.004	20	0.000		
		Total	0.032	23			
TBARS	C	Between Groups	0.076	3	0.025	100.085	0.000*
		Within Groups	0.005	20	0.000		
		Total	0.081	23			
	T	Between Groups	0.052	3	0.017	57.015	0.000*
		Within Groups	0.006	20	0.000		
		Total	0.058	23			
	H	Between Groups	0.083	3	0.028	121.535	0.000*
		Within Groups	0.005	20	0.000		
		Total	0.088	23			
Appearance	C	Between Groups	1.180	3	0.393	66.377	0.000*
		Within Groups	0.119	20	0.006		
		Total	1.299	23			
	T	Between Groups	0.747	3	0.249	12.206	0.000*
		Within	0.408	20	0.020		

		Groups					
		Total	1.156	23			
	H	Between Groups	2.152	3	0.717	164.287	0.000*
		Within Groups	0.087	20	0.004		
		Total	2.239	23			
Flavour	C	Between Groups	9.610	3	3.203	63.763	0.000*
		Within Groups	1.005	20	0.050		
		Total	10.614	23			
	T	Between Groups	25.986	3	8.662	118.599	0.000*
		Within Groups	1.461	20	0.073		
		Total	27.446	23			
	H	Between Groups	10.242	3	3.414	163.000	0.000*
		Within Groups	0.419	20	0.021		
		Total	10.661	23			
Juiciness	C	Between Groups	4.400	3	1.467	15.689	0.000*
		Within Groups	1.870	20	0.093		
		Total	6.270	23			
	T	Between Groups	26.228	3	8.743	113.163	0.000*
		Within Groups	1.545	20	0.077		
		Total	27.773	23			
	H	Between Groups	4.530	3	1.510	46.869	0.000*
		Within Groups	0.644	20	0.032		
		Total	5.174	23			
Texture	C	Between Groups	10.656	3	3.552	178.514	0.000*
		Within Groups	0.398	20	0.020		
		Total	11.053	23			
	T	Between Groups	19.431	3	6.477	148.355	0.000*
		Within Groups	0.873	20	0.044		
		Total	20.304	23			

	H	Between Groups	5.793	3	1.931	147.598	0.000*
		Within Groups	0.262	20	0.013		
		Total	6.054	23			
Overall acceptability	C	Between Groups	8.426	3	2.809	106.899	0.000*
		Within Groups	0.525	20	0.026		
		Total	8.951	23			
	T	Between Groups	26.077	3	8.692	263.904	0.000*
		Within Groups	0.659	20	0.033		
		Total	26.736	23			
	H	Between Groups	6.759	3	2.253	114.981	0.000*
		Within Groups	0.392	20	0.020		
		Total	7.151	23			
Chewiness	C	Between Groups	11.725	3	3.908	1.230	0.325 ^{NS}
		Within Groups	63.546	20	3.177		
		Total	75.271	23			
	T	Between Groups	14.559	3	4.853	0.795	0.511 ^{NS}
		Within Groups	122.105	20	6.105		
		Total	136.664	23			
	H	Between Groups	12.652	3	4.217	9.044	0.001*
		Within Groups	9.326	20	0.466		
		Total	21.978	23			
Cohesiveness	C	Between Groups	0.009	3	0.003	2.474	0.091 ^{NS}
		Within Groups	0.025	20	0.001		
		Total	0.034	23			
	T	Between Groups	0.009	3	0.003	4.993	0.010*
		Within Groups	0.013	20	0.001		
		Total	0.022	23			
	H	Between Groups	0.008	3	0.003	5.825	0.005*
		Within	0.009	20	0.000		

		Groups					
		Total	0.016	23			
Fracturability	C	Between Groups	26.723	3	8.908	6.277	0.004*
		Within Groups	28.382	20	1.419		
		Total	55.104	23			
	T	Between Groups	18.992	3	6.331	2.196	0.120 ^{NS}
		Within Groups	57.657	20	2.883		
		Total	76.649	23			
	H	Between Groups	22.334	3	7.445	14.989	0.000*
		Within Groups	9.933	20	0.497		
		Total	32.267	23			
Gumminess	C	Between Groups	12.524	3	4.175	5.485	0.006*
		Within Groups	15.221	20	0.761		
		Total	27.744	23			
	T	Between Groups	8.073	3	2.691	1.554	0.232 ^{NS}
		Within Groups	34.641	20	1.732		
		Total	42.714	23			
	H	Between Groups	13.904	3	4.635	19.968	0.000*
		Within Groups	4.642	20	0.232		
		Total	18.546	23			
Hardness	C	Between Groups	23.022	3	7.674	4.313	0.017*
		Within Groups	35.588	20	1.779		
		Total	58.611	23			
	T	Between Groups	15.443	3	5.148	7.893	0.001*
		Within Groups	13.043	20	0.652		
		Total	28.487	23			
	H	Between Groups	10.459	3	3.486	3.317	0.041*
		Within Groups	21.020	20	1.051		
		Total	31.479	23			

Springiness	C	Between Groups	0.052	3	0.017	2.343	0.104 ^{NS}
		Within Groups	0.149	20	0.007		
		Total	0.201	23			
	T	Between Groups	0.068	3	0.023	2.472	0.091 ^{NS}
		Within Groups	0.185	20	0.009		
		Total	0.253	23			
	H	Between Groups	0.064	3	0.021	21.911	0.000*
		Within Groups	0.019	20	0.001		
		Total	0.083	23			
Total plate count	C	Between Groups	1.259	3	0.420	350.698	0.000*
		Within Groups	0.024	20	0.001		
		Total	1.283	23			
	T	Between Groups	0.987	3	0.329	1011.218	0.000*
		Within Groups	0.007	20	0.000		
		Total	0.993	23			
	H	Between Groups	0.994	3	0.331	633.648	0.000*
		Within Groups	0.010	20	0.001		
		Total	1.004	23			
Yeast and mould count	C	Between Groups	1.077	3	0.359	440.174	0.000*
		Within Groups	0.016	20	0.001		
		Total	1.093	23			
	T	Between Groups	0.759	3	0.253	413.593	0.000*
		Within Groups	0.012	20	0.001		
		Total	0.772	23			
	H	Between Groups	0.864	3	0.288	596.734	0.000*
		Within Groups	0.010	20	0.000		
		Total	0.874	23			

CM- Cooking method, C – Control (Direct charcoal grilling); T- Treatment 1 (Moist cooking followed by charcoal grilling), H- Treatment 2 (Hot air roasting followed by charcoal grilling). (*) Significant, (NS) - not significant.

Table no. 29 Analysis of variance (ANOVA) for physico chemical and microbial parameters of mutton sheek kebab as influenced different cooking methods at refrigeration ($4\pm 1^{\circ}\text{C}$) temperature.

ANOVA							
	days		Sum of Square	Df	Mean Square	F	Sig.
pH	0	Between Groups	0.019	2	0.010	6.344	0.010*
		Within Groups	0.023	15	0.002		
		Total	0.042	17			
	5	Between Groups	0.011	2	0.006	4.543	0.029*
		Within Groups	0.018	15	0.001		
		Total	0.029	17			
	10	Between Groups	0.004	2	0.002	3.416	0.060 ^{NS}
		Within Groups	0.008	15	0.001		
		Total	0.012	17			
	15	Between Groups	0.001	2	0.001	1.204	0.327 ^{NS}
		Within Groups	0.006	15	0.000		
		Total	0.007	17			
TBARS	0	Between Groups	0.000	2	0.000	1.717	0.213 ^{NS}
		Within Groups	0.001	15	0.000		
		Total	0.002	17			
	5	Between Groups	0.002	2	0.001	2.035	0.165 ^{NS}
		Within Groups	0.008	15	0.001		
		Total	0.010	17			
	10	Between Groups	0.003	2	0.001	8.128	0.004*
		Within Groups	0.002	15	0.000		
		Total	0.005	17			

	15	Between Groups	0.005	2	0.003	10.85	0.001*	
		Within Groups	0.004	15	0.000			
		Total	0.009	17				
Appearance	0	Between Groups	2.882	2	1.441	179.9	0.000*	
		Within Groups	0.120	15	0.008			
		Total	3.002	17				
	5	Between Groups	3.447	2	1.724	208.3	0.000*	
		Within Groups	0.124	15	0.008			
		Total	3.571	17				
	10	Between Groups	4.558	2	2.279	219.7	0.000*	
		Within Groups	0.156	15	0.010			
		Total	4.714	17				
	15	Between Groups	4.483	2	2.242	156.8	0.000*	
		Within Groups	0.214	15	0.014			
		Total	4.697	17				
	Flavor	0	Between Groups	0.104	2	0.052	3.648	0.051 ^{NS}
			Within Groups	0.214	15	0.014		
			Total	0.318	17			
5		Between Groups	0.020	2	0.010	0.211	0.812 ^{NS}	
		Within Groups	0.722	15	0.048			
		Total	0.743	17				
10		Between Groups	0.091	2	0.045	0.438	0.653 ^{NS}	
		Within Groups	1.557	15	0.104			
		Total	1.648	17				
15		Between Groups	4.923	2	2.461	94.34	0.000*	
		Within Groups	0.391	15	0.026			
		Total	5.314	17				
Juiciness		0	Between Groups	1.989	2	0.994	50.11	0.000*
			Within	0.298	15	0.020		

		Groups					
		Total	2.287	17			
	5	Between Groups	1.712	2	0.856	18.82	0.000*
		Within Groups	0.682	15	0.045		
		Total	2.395	17			
	10	Between Groups	0.678	2	0.339	2.892	0.087 ^{NS}
		Within Groups	1.758	15	0.117		
		Total	2.435	17			
	15	Between Groups	3.343	2	1.672	18.97	0.000*
		Within Groups	1.322	15	0.088		
		Total	4.665	17			
Texture	0	Between Groups	1.293	2	0.646	49.59	0.000*
		Within Groups	0.196	15	0.013		
		Total	1.488	17			
	5	Between Groups	2.326	2	1.163	73.25	0.000*
		Within Groups	0.238	15	0.016		
		Total	2.564	17			
	10	Between Groups	4.828	2	2.414	45.62	0.000*
		Within Groups	0.794	15	0.053		
		Total	5.622	17			
	15	Between Groups	0.841	2	0.421	20.66	0.000*
		Within Groups	0.305	15	0.020		
		Total	1.147	17			
Overall acceptability	0	Between Groups	1.486	2	0.743	66.05	0.000*
		Within Groups	0.169	15	0.011		
		Total	1.655	17			
	5	Between Groups	1.756	2	0.878	87.34	0.000*
		Within Groups	0.151	15	0.010		
		Total	1.907	17			

	10	Between Groups	0.849	2	0.425	9.077	0.003*
		Within Groups	0.702	15	0.047		
		Total	1.551	17			
	15	Between Groups	0.867	2	0.433	17.33	0.000*
		Within Groups	0.375	15	0.025		
		Total	1.242	17			
Chewiness	0	Between Groups	86.763	2	43.38	12.39	0.001*
		Within Groups	52.510	15	3.501		
		Total	139.27	17			
	5	Between Groups	99.276	2	49.63	16.60	0.000*
		Within Groups	44.850	15	2.990		
		Total	144.12	17			
	10	Between Groups	87.402	2	43.70	13.28	0.000*
		Within Groups	49.358	15	3.291		
		Total	136.75	17			
	15	Between Groups	98.100	2	49.05	15.24	0.000*
		Within Groups	48.260	15	3.217		
		Total	146.36	17			
Cohesiveness	0	Between Groups	0.018	2	0.009	7.499	0.006*
		Within Groups	0.018	15	0.001		
		Total	0.037	17			
Cohesiveness	5	Between Groups	0.015	2	0.007	13.01	0.001*
		Within Groups	0.009	15	0.001		
		Total	0.024	17			
	10	Between Groups	0.017	2	0.009	13.53	0.000*
		Within Groups	0.010	15	0.001		
		Total	0.027	17			
	15	Between Groups	0.016	2	0.008	12.33	0.001*
		Within	0.010	15	0.001		

		Groups					
		Total	0.025	17			
Fracturability	0	Between Groups	483.72	2	241.8	141.1	0.000*
		Within Groups	25.704	15	1.714		
		Total	509.42	17			
	5	Between Groups	468.60	2	234.3	157.9	0.000*
		Within Groups	22.254	15	1.484		
		Total	490.858	17			
	10	Between Groups	465.460	2	232.7	142.7	0.000*
		Within Groups	24.448	15	1.630		
		Total	489.908	17			
	15	Between Groups	451.92	2	225.9	143.8	0.000*
		Within Groups	23.567	15	1.571		
		Total	475.48	17			
Gumminess	0	Between Groups	13.985	2	6.992	8.488	0.003*
		Within Groups	12.357	15	0.824		
		Total	26.342	17			
	5	Between Groups	11.980	2	5.990	6.393	0.010*
		Within Groups	14.054	15	0.937		
		Total	26.034	17			
	10	Between Groups	11.715	2	5.857	5.439	0.017*
		Within Groups	16.153	15	1.077		
		Total	27.868	17			
	15	Between Groups	13.359	2	6.679	8.391	0.004*
		Within Groups	11.940	15	0.796		
		Total	25.298	17			
Hardness	0	Between Groups	273.405	2	136.703	95.63	0.000*
		Within Groups	21.442	15	1.429		

		Total	294.84	17			
	5	Between Groups	236.16 6	2	118.08 3	85.53	0.000*
		Within Groups	20.708	15	1.381		
		Total	256.87	17			
	10	Between Groups	235.02 6	2	117.51 3	117.7	0.000*
		Within Groups	14.964	15	0.998		
		Total	249.99	17			
	15	Between Groups	241.80 6	2	120.90 3	144.6	0.000*
		Within Groups	12.538	15	0.836		
		Total	254.34	17			
Springiness	0	Between Groups	0.022	2	0.011	2.033	0.165 ^{NS}
		Within Groups	0.081	15	0.005		
		Total	0.103	17			
	5	Between Groups	0.014	2	0.007	1.153	0.342 ^{NS}
		Within Groups	0.090	15	0.006		
		Total	0.103	17			
	10	Between Groups	0.019	2	0.009	1.550	0.244 ^{NS}
		Within Groups	0.090	15	0.006		
		Total	0.108	17			
	15	Between Groups	0.029	2	0.015	2.372	0.127 ^{NS}
		Within Groups	0.092	15	0.006		
		Total	0.121	17			
Total plate count	0	Between Groups	0.076	2	0.038	86.80	0.000*
		Within Groups	0.007	15	0.000		
		Total	0.083	17			
	5	Between Groups	0.067	2	0.033	21.33	0.000*
		Within Groups	0.023	15	0.002		
		Total	0.090	17			
	10	Between Groups	0.046	2	0.023	34.84	0.000*

		Within Groups	0.010	15	0.001		
		Total	0.056	17			
	15	Between Groups	0.024	2	0.012	170.8	0.000*
		Within Groups	0.001	15	0.000		
		Total	0.025	17			
Yeast and mould count	0	Between Groups	0.090	2	0.045	112.7	0.000*
		Within Groups	0.006	15	0.000		
		Total	0.096	17			
	5	Between Groups	0.010	2	0.005	5.152	0.020*
		Within Groups	0.015	15	0.001		
		Total	0.026	17			
	10	Between Groups	0.018	2	0.009	13.57	0.000*
		Within Groups	0.010	15	0.001		
		Total	0.028	17			
	15	Between Groups	0.023	2	0.011	23.30	0.000*
		Within Groups	0.007	15	0.000		
		Total	0.030	17			

(*) significant, (NS) – not significant



Chapter – V

Discussion

CHAPTER V

DISCUSSION

5.1 PHYSICO-CHEMICAL PROPERTIES OF MUTTON SHEEK KEBAB

5.1.1 COOKING YIELD

Cooking yield of meat products is the most important criteria for the meat industry to arrive the product pricing and at the same time it plays greater role in determining the acceptability of the product by the consumers.

The cooking yield of moist cooked and charcoal grilled mutton sheek kebabs (T₁) was significantly (P<0.05) higher than that of hot air oven cooked and charcoal grilled kebabs (T₂) and direct charcoal grilled (C) kebabs. The findings of the present study are in congruence with the previous studies of Naveena *et al.* (2014) and Kiran *et al.* (2018), who also reported higher cooking yield in moist cooked chicken nuggets than the oven cooked patties. Similarly Chumngoen *et al.* (2018) also reported that the moist cooking of chicken steaks resulted in higher cooking yield than oven cooking.

Presence of cellulose casing layer over kebab during moist cooking and shorter duration of charcoal grilling could have contribute to the lower cooking loss of moist cooked and charcoal grilled mutton sheek kebabs (T₁). Chumngoen *et al.* (2016) reported that higher cooking temperature and prolonged cooking time contribute to higher cooking loss in meat and meat products.

Similarly, Gluchowski *et al.* (2019), also reported an inverse correlation between cooking yield and increase in cooking temperature in fish fillets. Longer cooking duration at higher temperature could be the cause for the reduced cooking yield of mutton sheek kebab in hot air oven method and direct charcoal grilling.

5.1.2 PROXIMATE COMPOSITION

The direct charcoal grilled kebabs (C) had significantly (P<0.05) lower moisture content followed by the hot air oven cooked kebab (T₂) and moist cooked mutton sheek

kebab (T₁). In case of protein, fat and ash content, the direct grilled kebabs (C) showed significantly ($P < 0.05$) higher protein, ash and fat content, followed by hot air oven cooked (T₂) and moist cooked and charcoal grilled mutton sheek kebab (T₁).

Lower moisture content in direct charcoal grilled kebabs might be due to the prolonged high temperature (200°C) and long cooking duration (20 minutes) in charcoal grill compared to moist cooking (100°C for 10 minutes) and hot air oven cooking method (150°C for 15 minutes). The higher moisture content in moist cooked and hot air oven cooked kebab compared to grilled kebab could be due to shorter exposure of kebabs to high temperature charcoal grilling method. These results are in agreement with the Naveena *et al.* (2014) and Kiran *et al.* (2018), who also reported higher moisture content in moist cooked chicken nuggets than dry cooked patties. The moisture loss of a product depends on the mass transfer process during thermal treatment (Murphy *et al.*, 2001). Further, Roldan *et al.* (2013) reported found that increase in cooking temperature resulted in more moisture loss in sous-vide cooked lamb loins.

Among the kebabs prepared by different cooking methods, the direct grilled kebabs (C) had significantly ($P < 0.05$) higher protein and fat content compared to the hot air oven cooked kebabs (T₂) and moist cooked kebabs (T₁). The higher protein and fat content was due to the higher moisture loss in control grilled kebabs compared to other treatments groups (Karpinska-Tymoszczyk *et al.*, 2020). Significantly higher protein content in direct grilled kebabs compared to the hot air oven cooked kebabs is in congruence with the findings of Bannerjee *et al.* (2018), who employed different cooking methods for the preparation of chicken sheek kebabs. The hot air grilled kebabs (T₂) had significantly ($P < 0.05$) higher protein content compared to the moist cooked kebabs (T₁). Similarly, Naveena *et al.* (2014) and Kiran *et al.* (2018) also

reported higher protein and fat in the dry hot air oven cooked emulsion based chicken products compared to the moist cooked products. The protein and fat content of kebabs varied inversely with moisture content is similar in Naveena *et al.* (2014) and Kiran *et al.* (2018) might be the probable cause for reduced fat % in moist cooked kebabs.

The ash content of the mutton sheek kebabs are significantly ($P<0.05$) affected by different cooking methods employed in our study. Higher ash content in direct grilled kebabs compared to the moist cooked kebabs is similar with the findings of Choi *et al.* (2016), who prepared marinated chicken steaks by employing different cooking methods. The hot air grilled kebabs (T_2) had significantly ($P<0.05$) higher ash content compared to the moist cooked kebabs (T_1). Similarly, Naveena *et al.* (2014) and Kiran *et al.* (2018) also reported ash content in the dry hot air oven cooked emulsion based chicken products compared to the moist cooked products.

5.1.3 COLOR

The product with more lightness is highly preferred by the consumers. Mutton kebabs prepared by moist cooking followed by charcoal grilled (T_1) had higher lightness L^* values than the hot air oven cooking (T_2) and direct charcoal grilling (C). Higher lightness in the boiled sample might be associated with opaque-white color of denatured myofibrillar protein (Putra *et al.*, 2019).

These results are in congruence with the previous findings of Choi *et al.* (2016) who reported higher lightness values in boiled chicken steak compared to oven and grilled chicken steaks and Nikmaram *et al.* (2011) found that the lower moisture loss in braised veal resulted in higher lightness values than roasting. Similarly, Chumngoen *et al.* (2018) also reported that dry-heat cooking particularly roasting, accelerates the maillard reaction which resulted in lower lightness values for the oven cooking than the moist cooking in chicken. Direct charcoal grilled kebabs had lower lightness values due

to the formation of crust on the outer surface of kebabs (Bannerjee *et al.*, 2018)

Among the kebabs prepared by different cooking methods, moist cooked and charcoal grilled kebabs had significantly ($P < 0.05$) higher a^* and b^* values than rest of the formulations which are in agreement with Nikmaram *et al.* (2011) who reported that the braising of veal muscle reported higher redness values than roasted samples. Similarly, Głuchowski *et al.* (2019) also found higher redness and yellowness values in sous vide cooked fish than roasted fish. Increase in time-temperature combination (Głuchowski *et al.*, 2019) and protein denaturation in direct charcoal grilled kebab (200°C for 20 minutes) and hot air oven cooked kebabs (150°C for 15 minutes) tends to have lower a^* and b^* values (Karpinska-Tymoszczyk *et al.*, 2020).

Mutton kebabs prepared by moist cooking followed by charcoal grilling (T_1) had lower ($P < 0.05$) hue angle followed by hot air oven cooked kebab (T_2) and direct charcoal grilled (C) kebab which might be due to the inverse relationship between that the hue angle and redness (a^*) values is similarly reported in Del Pulgar *et al.* (2012) in sous-wide cooked pork.

The chroma values relates to the concentration of myoglobin and also to its degree of denaturation and its values are based on the redness a^* and yellowness b^* values. Among the kebabs prepared by different cooking methods, moist cooked and charcoal grilled kebabs (T_1) had significantly higher ($P < 0.05$) chroma values which are in agreement with Głuchowski *et al.* (2019) who reported that the sous wide cooked fish fillets had higher chroma values than roasted samples.

5.1.4 QUANTIFICATION OF POLYCYCLIC AROMATIC HYDROCARBONS

Polycyclic Aromatic Hydrocarbons compounds are formed during the pyrolysis of organic matter such as fat, carbohydrates, and protein with temperature exceeding 200°C, (Codex 2005 CX/FAC 05/37/34; Mcgrath *et al.*, 2003; Alomirah *et al.*, 2011).

Benzo[a]pyrene and benz[a]anthracene were observed at and above 500 °C (Mcgrath *et al.*, 2003).

The second major contributing factor for the PAH formation is the direct contact of lipids that dripped by having direct contact over the flame at intense heat. Other factors which influence PAH formation are the cooking method, time and temperature, fat content (Onwukeme *et al.*, 2015; Lee *et al.*, 2016), kind of fuel, position and distance of the product in relation to the heat source (Alomirah *et al.*, 2011; Rose *et al.*, 2015, Onwukeme *et al.*, 2015).

In our present study, out of 9 PAH compounds analyzed, only one compound flouranthene (light PAH) is detected in all the kebab while the rest of the PAH compounds are below the level of quantification. The grilling temperature and the fat content of kebabs mainly influence the levels of PAH, while majority of researchers reported PAH generation takes above 200°C.

These results are similar to the observations documented by Kao *et al.*, (2014) who reported that only fluoranthene compounds detected out of 9 PAH compounds in grilled chicken, seafoods and red meat products. Similarly Muthukumar *et al.* (2018) reported the levels of carcinogenic PAH compound were under the limit of quantitation (0.01mg/kg) in tandoori chicken, chicken tikka, and smoked chicken. Further, Onwukeme *et al.*, (2015) reported that fluoranthene detected only in roasted chicken compared to the boiled chicken.

Flouranthene compound is formed by the fusion of naphthalene and benzene unit connected by a five-membered ring, positive genotoxic, class 3 (not classifiable carcinogen) IARC classified PAH compound. The flouranthene values are significantly ($P < 0.05$) higher in direct charcoal grilled mutton sheek kebab followed by hot air oven cooked and grilled kebab (T_2) and the moist cooked and charcoal grilled kebabs (T_1). Higher flouranthene values in direct charcoal grilled mutton kebabs are due

to the higher temperature and prolonged charcoal grilling time (200°C for 10 minutes) which is similarly reported in Rose *et al.*, (2015) and Kao *et al.*, (2014). Presence of cellulose casing layer over kebab during moist cooking followed by shorter grilling time could have contribute to the lower flouranthene values of moist cooked and charcoal grilled mutton sheek kebabs (T₁). Higher fluoranthene levels in grilled chicken than the oven roasted chicken is similar to the findings of Farhadian *et al.*, (2010).

The levels of PAH compounds in the mutton sheek kebabs are influenced by cooking methods. Though the levels of PAH compounds in the kebabs are below the tolerance limit by the European regulation, moist cooking generates less PAH compounds compared to hot air oven and direct charcoal grilling and is regarded as safest among other cooking methods.

5.2 SHELF-LIFE STUDIES OF MUTTON SHEEK KEBAB DURING REFRIGERATED (4±1°C) STORAGE

5.2.1 pH

The pH values of the direct charcoal grilled (C) were significantly (P<0.05) lower than hot air oven (T₂) and moist cooked kebabs (T₁). The results of our study are in congruence with Kiran *et al.* (2018) who reported higher pH values in dry cooked patties than moist cooked chicken nuggets. Lower pH in direct charcoal grilling (C) is due to the longer cooking time is similar in Karpinska-Tymoszczyk *et al.* (2020) in sous vide cooked chicken breast fillets.

Irrespective of the groups, a gradual increase in pH values was noticed, as the storage period advanced which are in agreement with Das *et al.* (2006) in vacuum packaged goat meat patties refrigerated storage, Bhat *et al.* (2011) in chicken sheek kebabs under refrigerated storage, Malav *et al.* (2016) in vacuum packaged functional mutton patties under refrigerated storage.

The production of ammonia coupled with accumulation of microbial metabolites due to bacterial spoilage might have increased the pH during the refrigerated storage is similar in Biswas *et al.* (2012) in pork patties under refrigerated storage.

5.2.2 2-THIOBARBITURIC ACID REACTIVE SUBSTANCES

Among the kebab samples, moist cooked and charcoal grilled kebabs (T₁) had lower TBARS values followed by hot air cooked kebabs (T₂) and direct grilled control kebab groups (C) which are not in congruence with Kiran *et al.* (2018) who reported that hot air oven dried chicken patties had significantly lower TBARS value than the moist cooked nuggets. Lower TBA values in moist cooked kebabs and hot air cooked kebabs indicates reduced oxidation of lipids might be combined effect of a relatively shorter cooking time and temperature is similar in Kiran *et al.* (2018), Broncano *et al.* (2009) could have contributed to the reduced TBARS values.

There was no significant difference ($P>0.05$) in 2-TBARS values among the treatment groups (T₁ and T₂) until day 10 is similar in Naveena *et al.* (2014) who reported that similar TBARS values for the dry cooked chicken patties and moist cooked nuggets.

An increasing trend of TBARS values were noticed in all groups towards the end of storage period due to the lipid oxidation of meat. Similar increase in TBARS values during storage have been reported by Das *et al.* (2008) in vacuum packaged goat meat patties refrigerated storage, Kayaardi *et al.* (2006) in doner kebabs under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage, Malav *et al.* (2016) in vacuum packaged functional mutton patties under refrigerated storage.

5.2.3 SENSORY EVALUATION:

5.2.3.1 APPEARANCE

Among the kebabs prepared by different methods, moist cooked and charcoal grilled kebabs (T₁) scores higher appearance, which might be due to the uniform shape of the kebabs which happens due to the even distribution of batter inside the cellulose casing remains unchanged, after cooking. These results are in congruence with Kiran *et al.* (2018), who reported higher appearance score in moist cooked chicken nuggets than dry cooked patties. Similarly Choi *et al.* (2016) also reported higher appearance score in boiled chicken steaks than oven cooked steaks. Further, Bannerjee *et al.* (2018) also reported that higher appearance score in roasted chicken sheek kebabs than grilled kebabs.

Progressive decline in the appearance score is observed in all the formulations during storage period are in accordance with Kayaardi *et al.* (2006) in doner kebab made from turkey meat stored under refrigerated storage, Malav *et al.* (2016) in vacuum packed mutton patties stored under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage.

5.2.3.2 FLAVOR

The flavor scores of mutton sheek kebabs are superior in direct charcoal grilling (C) than hot air cooking (T₂) and moist cooking (T₁). The typical smoky flavor development in direct charcoal grilling is due to the longer duration of grilling compared to rest of other formulations. These results are similar to the observations made by Choi *et al.*, (2016) who reported high flavor scores in charcoal grilled chicken steaks compared to the boiled and oven cooked steaks. Similarly, Kiran *et al.* (2018) also reported higher flavor score in oven cooked chicken patties than moist cooked nuggets. Further, Chumngoen *et al.* (2018) reported lower flavor scores in moist cooked chicken breast compared to the oven cooking.

Similar findings of decrease in flavor scores during storage of mutton sheek kebabs have been documented by Kayaardi *et al.* (2006) in doner kebab made from turkey meat stored under refrigerated storage, Malav *et al.* (2016) in vacuum packed mutton patties stored under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage.

5.2.3.3 JUICINESS

Among the kebabs, moist cooked and charcoal grilled kebabs (T₁) had higher juiciness scores followed by direct charcoal grilled kebabs (C) and hot air cooked kebabs (T₂). Higher juiciness in treatment (T₁) is correlated with the higher moisture content and lower hardness scores (Bannerjee *et al.* 2018).

The results of our findings are in agreement with Bannerjee *et al.* (2018) who reported higher juiciness values in charcoal grilled chicken kebabs than oven cooked kebabs. But our results are not in accordance with Kiran *et al.* (2018) who reported higher juiciness scores in oven cooked chicken patties than moist cooked nuggets.

The juiciness score of mutton sheek kebabs were decreased towards the end of storage period which are in correlation with Kayaardi *et al.* (2006) in doner kebab made from turkey meat stored under refrigerated storage, Malav *et al.* (2016) in vacuum packed mutton patties stored under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage.

5.2.3.4 TEXTURE:

Moist cooked and charcoal grilled kebabs (T₁) had higher texture scores followed by the hot air cooked kebabs (T₂) and direct charcoal grilled kebabs (C). Higher texture scores for moist cooked kebabs is due to the uniform distribution of batter inside the cellulose casing layer while cooking and are also correlated with the higher moisture content and lower hardness scores (Bannerjee *et al.*, 2018).

The results of our study are not in agreement with findings of Bannerjee *et al.* (2018) who employed different cooking methods for the preparation of chicken sheek kebabs and found higher texture scores in charcoal grilled chicken kebabs than oven cooked kebabs. Similarly Kiran *et al.* (2018) also reported higher texture scores in oven cooked chicken patties compared to the moist cooked nuggets.

Progressive decline in texture scores of mutton sheek kebabs are observed during storage period is similar to the findings of Kayaardi *et al.* (2006) in doner kebab made from turkey meat stored under refrigerated storage, Malav *et al.* (2016) in vacuum packed mutton patties stored under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage.

5.2.3.5 OVERALL ACCEPTABILITY

Mutton sheek kebabs prepared by moist cooking are mostly preferred by the sensory panelists due to their juiciness and desired textural attributes and comparatively uniform shaped products with the higher moisture content and lower hardness scores.

Moist cooked and charcoal grilled kebabs had higher overall acceptability scores followed by the hot air cooked kebabs (T₂) and direct charcoal grilled kebabs (C). These observed results are in congruence with Kiran *et al.*, (2018) who reported higher overall acceptability scores in moist cooked chicken patties than oven cooked nuggets. Similarly, Bannerjee *et al.* (2018) also reported that the roasted chicken kebabs had higher overall acceptability scores than the grilled kebabs.

A gradual decline in overall acceptability scores is evident in all groups of mutton sheek kebabs during storage period. Similar decrease in overall acceptability have been documented by Kayaardi *et al.* (2006) in doner kebab made from turkey meat stored under refrigerated storage, Malav *et al.* (2016) in vacuum packed mutton patties stored under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage.

5.2.4 TEXTURE PROFILE ANALYSIS

5.2.4.1 CHEWINESS

Mutton sheek kebabs prepared by moist cooked and charcoal grilled kebabs (T₁) had significantly ($P < 0.05$) lower chewiness values followed by hot air oven cooked kebabs (T₂) and direct charcoal grilled kebabs (C), our findings are in accordance with Putra *et al.* (2019) reported higher chewiness values in grilled goat meat than the roasted and boiled samples.

Chewiness value indicates the number of chews required for preparing the sample for swallowing, which are correlated with the values of gumminess and springiness (Putra *et al.* 2019).

But our results are not in agreement with the findings of Bannerjee *et al.*, (2018) who reported higher chewiness values in roasted chicken kebab than grilled kebabs and Aguirre *et al.*, (2018) reported the higher chewiness values in roasted chicken than grilled samples. Similarly, Suleman *et al.* (2020) also reported higher chewiness values in roasted lamb patties than grilled samples.

5.2.4.2 COHESIVENESS

Among the kebabs prepared by different methods, direct charcoal grilled kebabs had higher cohesiveness values which are mainly due to the positive correlation with the moisture content in the product (Bannerjee *et al.* 2018).

Mutton kebabs prepared by hot air oven cooking followed by charcoal grilling (T₂) had lower cohesiveness values followed by moist cooked kebabs (T₁) and direct charcoal grilled kebabs (C) samples which are similar to the findings of Suleman *et al.* (2020) who reported higher cohesiveness values in charcoal grilled samples lamb patties than superheated steam roasting. But our results are not in congruence with the observations made by Bannerjee *et al.* (2018) who reported higher cohesiveness values in roasted chicken kebabs than grilled kebabs. Similarly, Aguirre *et al.* (2018) also

found no significant difference in cohesiveness values between the roasted and grilled chicken kebabs.

5.2.4.3 FRACTUABILITY

The mean fracturability values are significantly ($P < 0.05$) affected by cooking methods. Lower fracturability values in moist cooking followed by charcoal grilling (T_1) might be due to the correlation between the higher moisture content and lower hardness values compared to the rest of the formulations. Higher fracturability in direct charcoal grilled kebabs is related to the hardness values because of the crust formation on the outer surface during grilling (Bannerjee *et al.*, 2018 and Putra *et al.*, 2019).

5.2.4.4 HARDNESS

Hardness values of the mutton sheek kebabs are significantly ($P < 0.05$) affected by different cooking methods employed in our study. Higher grilling temperature (200°C) in direct charcoal grilled (C) kebabs leads to lower moisture content and prolonged grilling duration (20 minutes), which in turns produces more crust formation on the outer surface of kebabs, contributes to higher hardness (Bannerjee *et al.*, 2018 and Putra *et al.*, 2019).

These results are in correlation with Putra *et al.* (2019) who reported higher hardness values in grilled goat meat compared to the roasted and boiled samples. Similarly, Suleman *et al.* (2020) also found that the grilled lamb patties had higher hardness values than roasted patties. But our results are not in congruence with Bannerjee *et al.* (2018) in chicken sheek kebab reported higher hardness values in roasted kebab than grilled kebabs. Further, Aguirre *et al.* (2018) also reported higher hardness values in roasted chicken than grilled chicken.

5.2.4.5 GUMMINESS

The gumminess values indicates the degree to which the samples sticks to the mouth which mainly depends on the values of hardness and cohesiveness which might

be probable cause for the lower gumminess values of direct charcoal grilled (C) kebabs.

Direct charcoal grilled control kebabs (C) had lower gumminess values followed by moist cooked kebabs (T₁) and hot air cooked kebab (T₂) groups which are in congruence with Bannerjee *et al.*, (2018), who reported lower gumminess values in grilled chicken kebab than the roasted kebabs. But our results are not in congruence with Suleman *et al.* (2020) who reported higher gumminess values in grilled lamb patties than roasted patties. Similarly, Putra *et al.*, (2019) also reported higher gumminess values in grilled goat meat compared to the roasted and boiled samples.

5.2.4.6 SPRINGINESS

The springiness values indicates the extend to which sample returns to the original shape after pressing and are mainly correlated with the influence of moisture content in the product (Bannerjee *et al.*, 2018).

Cooking method had no significant effect ($P>0.05$) on springiness values among all the kebab groups. Similar results have been documented by Putra *et al.* (2019) who employed different cooking methods in goat meat.

The direct charcoal grilled mutton sheek kebab control (C) had higher springiness values followed by moist cooked and charcoal grilled kebabs (T₁) and hot air cooked kebab (T₂) groups which are in congruence with Bannerjee *et al.* (2018) who reported higher springiness values in grilled kebabs than the roasted kebabs. But these results were not in agreement with Suleman *et al.* (2020) who reported the higher springiness in grilled lamb patties than roasted patties. Similarly Aguirre *et al.* (2018) also reported similar springiness values were observed for both roasted and grilled chicken samples.

In our present study, the cooking methods viz., moist cooking, hot air oven cooking and direct charcoal grilling of mutton sheek kebabs significantly ($p<0.05$)

affects the textural parameters such as chewiness, cohesiveness, hardness, gumminess, fracturability while, no significant difference in springiness values are noticed among them. A slight decline in all the textural profile values is noticed during the storage period.

5.2.5 MICROBIAL ANALYSIS

Among the kebabs, the total plate count and yeast and mould count values of direct charcoal grilled kebabs control (c) were significantly ($P < 0.05$) lower, followed by hot air cooked (T_2) and moist cooked kebabs (T_1) during the entire storage period, which might be due to the higher temperature and prolonged cooking time (200°C for 20 minutes) and lower moisture content of the control kebabs which resulted in reduced microbial activity. Similar results have been documented by Naveena *et al.* (2014) who reported the higher total plate count values are observed in moist cooked nuggets than the dry cooked chicken patties.

The total plate count values of mutton sheek kebabs are significantly ($P < 0.05$) increased with increasing storage period which are in accordance with kayardi *et al.* (2006) in doner kebab under refrigerated storage, Malav *et al.* (2016) in vacuum packed mutton patties stored under refrigerated storage, Pathera *et al.* (2016) in chicken nuggets under refrigerated storage. Similarly the yeast and mould count values of mutton sheek kebabs are also significantly ($P < 0.05$) increased with increasing storage period which are in accordance with Pathera *et al.* (2016) in chicken nuggets under refrigerated storage.

For the entire 15-day storage time, no psychrophilic bacterial counts are found in mutton sheek kebabs. This might be due to the partial destruction of psychrophilic organisms by means of cooking process and temperature variation for the growth of psychrophilic bacteria. Similar results of psychrophilic counts have been documented

by Cholan (2008) in low-fat chicken nuggets and Chandralekha (2010) in refrigerated chicken meat balls incorporated with natural and synthetic antioxidants and Reddy *et al.* (2017) in refrigerated chicken patties incorporated with natural antioxidants.



Chapter – VI

Summary

CHAPTER- VI

SUMMARY

Meat plays a crucial role in human health as a rich source of well-balanced diet with high biological value protein, containing all the essential amino acids, highly bio-available minerals, vitamins, and micronutrients. Cooking makes the meat easier to chew, digest and makes it more nutritious than raw meat.

Meat and meat products undergo many chemical changes while cooking, including the formation of numerous compounds imparting desirable/undesirable characteristics to them. Cooking at high-temperature affects the nutritional composition of meat due to the destruction of amino acids and vitamins, depending on the specific cooking conditions like temperature, time, pH and moisture content. Recent modern analytical technologies and investigations have revealed that cooking can generate toxic compounds generally not present in raw meat. One such compound, polycyclic aromatic hydrocarbon (PAH) is formed mainly during grilling and smoking.

In the present investigation, mutton sheek kebabs were prepared by three types of cooking methods – direct charcoal grilling (Control), moist cooking followed by charcoal grilling (Treatment 1), hot air oven cooking followed by charcoal grilling (Treatment 2). In moist cooking, sausage stuffer was used for stuffing the batter into the cellulose casing to get the uniform-shaped kebabs. The formed kebabs were cooked by moist cooking followed by charcoal grilling.

Mutton sheek kebabs prepared by all the three methods cooking were stored under refrigerated ($4\pm 1^{\circ}\text{C}$) storage for 15 days. Several physico-chemical parameters were estimated at regular intervals of 0, 5, 10 and 15th day of refrigerated ($4\pm 1^{\circ}\text{C}$) storage.

The cooking yield was significantly higher in the moist cooked kebabs (T₁) followed by the hot air oven cooked kebabs (T₂) and the direct charcoal grilled kebabs (C). Proximate composition analysis of the mutton kebabs revealed that the moist cooked kebabs had significantly higher moisture content, whereas the direct charcoal grilled kebabs are significantly higher in protein, fat and ash content.

Mutton kebabs prepared by moist cooking followed by charcoal grilling (T₂) had significantly higher color values of lightness (L*), redness (a*) and yellowness (b*) and chroma. In contrast, direct charcoal grilled kebabs (C) had a significantly higher Hue angle.

Among the nine PAH compounds chosen for the analysis, only fluoranthene (light PAH compound) was detected and other compounds were found below the quantification level (0.02 ppb). Among the kebab samples, charcoal grilled kebabs had a significantly higher fluoranthene level followed by hot air cooked and moist cooked kebabs.

The mean pH values are lower in direct charcoal grilled kebabs (C) followed by the moist cooked kebabs (T₁) and the hot air oven cooked kebabs (T₂). There was an increasing trends of pH values observed during the storage period in all the three types of kebabs. Moist cooked and charcoal grilled kebabs (T₂) had lower TBARS values than the hot air cooked kebabs (T₂) and direct charcoal grilled kebabs (C). TBARS values were progressively increased as the storage period advanced.

Cooking methods significantly (P<0.05) affected the textural parameters of mutton sheek kebab. Texture profile analysis revealed that the moist cooked and charcoal grilled kebabs (T₁) had significantly lower hardness, fracturability, chewiness values, while higher values of gumminess and springiness compared to the hot air oven

cooked kebabs and direct charcoal grilled kebabs. During storage period, gradual decline in all the textural parameters were noticed.

The trained sensory panelist analyzed the mutton sheek kebabs during the storage period. Mutton sheek kebab prepared by moist cooking followed by charcoal grilling (T₁) was mostly preferred by the sensory panelist and ranked first in all the sensory attributes except in flavor upto 10 days of storage. Higher flavor score were noticed for the direct charcoal grilled kebabs (C). Moist cooked kebabs spoiled at 15th day of storage. Sensory attributes of all the groups showed a diminishing trend as the storage period progressed.

Microbial analysis revealed that the direct charcoal grilled kebabs (C) had significantly lower total plate count and yeast & mould count values followed than the hot air oven cooked kebabs. In contrast, the moist cooked and charcoal grilled kebabs (T₁) had significantly higher values in both total plate count and yeast and mould count. An increasing trend in microbial counts was observed during the storage period in all the three type of kebab samples. Psychrophilic bacteria counts were not detected in any of the samples during 15 days of refrigerated storage.

Based on the results of present study, it can be concluded that,

- Mutton sheek kebabs prepared by moist cooking and charcoal grilling (T₁) were exceptionally good in cooking yield, color parameters, appearance, texture, juiciness, and overall acceptability.
- The innovative method of using sausage stuffer and cellulose casing to prepare kebabs in moist cooking yields good results in uniform-shaped appearance, mostly liked by sensory panelists and simultaneously increased the cooking yield.

- Direct charcoal-grilled kebabs (C) performed well in terms of reduced pH, increased protein content which is related to its reduced moisture content, flavor, microbial quality, and shelf-life stability of the product.
- The levels of Polycyclic aromatic compounds (PAH) - undesirable chemicals generated during the cooking process were minimized in moist cooking compared to the hot air cooking and direct charcoal grilling of kebab.



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APPENDIX

ANNEXURE

National Research Centre on Meat, Chengicherla, Hyderabad
Form for sensory Evaluation of Meat Products

Panelist's Name:

Product Name:

Date:

Descriptive Scales for Sensory Attributes of Meat Products								
Sensory attributes	8	7	6	5	4	3	2	1
Appearance	Excellent	Very good	Good	Fair	Slightly poor	Moderately poor	Very poor	Extremely poor
Flavor	Extremely desirable	Very desirable	Moderately desirable	Slightly desirable	Slightly undesirable	Moderately undesirable	Very undesirable	Extremely undesirable
Juiciness	Extremely juicy	Very juicy	Moderately Juicy	Slightly juicy	Slightly dry	Moderately dry	Very dry	Extremely dry
Texture	Extremely desirable	Very desirable	Moderately Desirable	Slightly desirable	Slightly undesirable	Moderately undesirable	Very undesirable	Extremely Un desirable
Overall acceptability	Extremely palatable	Very palatable	Moderately Palatable	Slightly palatable	Slightly unpalatable	Moderately unpalatable	Very unpalatable	Extremely unpalatable

Product code	Appearance	Flavor	Juiciness	Texture	Overall acceptability
101					
102					
103					
104					

Remarks:

Signature of Panelist