

## **Bacteriological, physicochemical and histological assessment of marketed beef burger in the Egyptian market**

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### **1. Abstract**

This study aimed to assess the safety and quality of the beef burger sold in the Egyptian market. Hundred samples of marketed beef burger were collected from different retail markets in Cairo and Giza governorates. Samples subjected to sensory, physicochemical, bacteriological as well as histological examinations. The findings revealed that examined samples have low scores of sensory attributes. Moreover, chemical examinations of samples showed very low protein (8.80%), high moisture (66.12%), and high fat (20.45%) content. Deterioration criteria of samples indicated that the mean pH value was  $5.60 \pm 0.05$ , high TBA value ( $0.66 \pm 0.02$  mg malonaldehyde/kg), and high TVBN value ( $18.74 \pm 0.32$  mg % sample) but their values not exceeded the permissible limit by ESS 1688/2005. Evaluation of cooking characteristics of examined samples indicated that the mean values of cooking loss, moisture retention, fat retention, diameter reduction, shrinkage and water holding capacity percentages were 23.66, 31.50, 83.52, 17.31, 19.76 and 75.15% respectively. Furthermore, bacteriological examination of samples indicated high bacterial load. The APC, psychrotrophic, Staphylococci, coliforms and fecal coliform counts ( $\log_{10}$  CFU/g) were 7.69, 6.09, 5.43, 3.84 and 2.25. The incidence of *Staph. aureus*, Salmonellae and *E. coli* in the marketed beef burger were 40%, 0 % and 60% respectively where all the isolated *E. coli* was belonging to the serovar O114:K90. The histological details of marketed beef burger illustrated that the skeletal muscles were the predominant structure which loss its organizational structure as signs for autolysis. Different tissues were also observed as wall of blood vessels, connective tissue, bone, cartilage, adipose tissue, and tissues of plant origin. Therefore, strict inspection on processing meat plants should be applied to improve the product safety and quality of this product.

**Key words:** Beef burger, Histological. Physico-chemical, Sensory, Bacterial quality

### **2. Introduction**

Consumption of meat products is an integral part of our daily diet. Meat is liked by a wide range of consumers as a result of its pleasant taste and flavor. Additionally,

Meat is a valuable source of essential nutrients such as high quality protein, vitamins (B<sub>12</sub>), minerals, essential amino acids and essential fatty acids [1]. Globally,

world consumption of meat products, especially by children and young people, has increased over the last years as they characterized by rapid preparation as fast foods, easily handled, stored, and low prices in comparison with raw meat. However, many researchers reported that consumption of meat was correlated with many diseases like liver, heart, lung, and urinary tract diseases [2].

Firstly, meat products were manufactured to produce more palatable products from low quality meat parts. They were processed from cuts of meat that contain high levels of connective tissue or fat and may be manufactured from fat and meat trimmings [3]. Meat products are mainly produced by mixing meat with salt, water, fat, spices and other ingredients [4]. The quality of raw materials and additives used in the production of meat products mainly reflected on the quality of the processed final meat products [5]. Among these products, beef burger which is a popular, delicious and nutritious meat

product for consumers especially children and young people. Generally, beef burger is a formed meat product (disc-like shape or balls) produced by mixing minced meat with fat, spices and other additives [6]. Burger was originally made of beef, but chicken and mutton burgers have become popular in recent years. Although, beef burger is regarded as a good media for growth of microorganisms due to its high moisture, nitrogen, mineral, carbohydrates as well as encouraging pH values that resulted in rapid spoilage, food borne illness and economic losses [7]. Consequently, bacteriological examination is a valuable method to evaluate the safety as well as the quality of meat products. Beef burger can be subjected for contamination during processing step from the working tables, knives as well as hands and clothes of workers. Also, it can be contaminated from the surrounding environment during transportation, distribution, handling, and preparation. These sources of contamination decrease

the product quality or even make it unfit for human consumption [8, 9]. In this regards, *Salmonellae*, *E. coli*, *Pseudomonas*, and *Staph. aureus* are the most commonly isolated microorganisms from meat and meat products that cause food borne infection and intoxication [10].

Beef burger and other meat products are also subjected for adulteration by some producers that cause food borne illness and health problems. Moreover, falsification of these products breaks the trust of consumers towards the meat industry. Adulteration of meat products with low quality raw materials yields highly perishable products that make the producers adding high amount of nitrites to extend their shelf life. Although, many researchers reported that overconsumption of such products which contain high levels of nitrite may cause cancer and methaemoglobinaemia [11]. Moreover, processors cover the flavor problems by adding monosodium glutamate (MSG) that acts as flavor enhancer [12] While, many

studies stated that MSG had genotoxic and toxic effects on human if consumed at high levels [13]. Furthermore, starch is added in different meat products as meat stabilizer if added at a rate not more than 5% and acts as a meat extender if used with a rate up to 10% [14]. While, many producers usually add it to 25% to increase the total weight and to reduce the cost of the products, which affect the technological characteristics of the product resulted in dry texture and bad binding [15]. On the other hand, histological techniques have been used since 1960 to assess the quality of meat products and also used for identification and differentiation of ingredients added in meat products [16]. Moreover, beef burger is stored under freezing conditions which expose this product for moisture loss; oxidative changes as well as protein and lipid decomposition [17].

International and national regulations of food industry make its effort to guarantee the quality and safety of the

food by monitoring its composition and the quality characteristics of foods includes: bacteriological, chemical, physical, and sensory properties [18]. Therefore, the target of the present study is the evaluation for the safety and quality of beef burger sold in some retail markets in Cairo and Giza governorates to raise the consumer awareness about dangers resulted from consumption of such commercially produced products such as food borne infection and intoxication beside their low nutritional value.

### **3. Materials and Methods**

#### **3.1. Samples collection**

Hundred commercially produced beef burger samples were randomly collected from different supermarkets in Giza and Cairo governorates. Each sample was represented by 1 package (~500g each). In a cooling ice box, samples were immediately transferred to the Department of Food Hygiene, Faculty of Veterinary Medicine, Cairo University in order to investigate their quality parameters in terms of sensory, chemical, physicochemical,

bacteriological, and histological examinations.

#### **3.2. Investigations**

##### **3.2.1. Sensory evaluation**

Raw beef burger samples were assessed for appearance, color, odor, consistency, comminution, binding, forming, fringe formation and overall acceptability as the scheme described by Larmond et al. [19]. Each sample was coded randomly, thawed completely and evaluated by twenty-seven highly trained panelists. The panelists were trained well in order to describe every sample using a numerical-score value from 1 to 9 according to their quality with 1 being low or dislike and 9 being extremely like. The beef burger patties were then cooked in a preheated electrical grill for a total of five minutes, 2.5 minutes on each side to 70°C core temperature before being coded and evaluated by the same panelists for the following parameters (appearance, color, flavor, tenderness, juiciness, binding,

forming and overall acceptability). The cooked burger samples were cooled and reweighted to determine the cooking loss percentage.

### **3.2.2. Chemical examinations**

Samples were ground into uniform mass using a meat mincer three times and mixed well, then analyzed as follows:

#### **3.2.2.1. Proximate chemical analysis:**

The technique recommended by AOAC [20] was applied to measure the moisture, protein, and fat contents of beef burger after processing. Ten grams of each sample were dried in a hot air oven at 100 °C to determine moisture content. In accordance with Kjeldahl's method, protein content was measured. Petroleum ether was used in soxhlet apparatus to measure the fat content.

#### **3.2.2.2. Deterioration criteria**

pH values were measured by mixing five grams of prepared beef burger samples with 20 ml distilled water [21], then measured with a pH meter, using a probe

electrode that was first standardized with two buffers (7.0 and 4.0). The thiobarbituric acid value (TBA, mg malonaldehyde/kg of sample) was determined by the technique mentioned by Du and Ahn [22]. Furthermore, using macro-Kjeldahl distillation, Total Volatile Base Nitrogen (TVBN, mg% sample) was measured according to the method carried out by Kearsley et al. [23].

#### **3.2.2.3. Physicochemical Characteristics**

Beef burger samples were cooked as mentioned in sensory examination then moisture retention was measured by the method described by Murphy et al [24], fat retention as well as diameter reduction was performed as the method stated by Serdaroğlu and Değirmencioğlu [25], water holding capacity was measured according to Hongprabhas and Barbut [26].

#### **3.2.3. Bacteriological Examinations:**

The aerobic plate count ( $\text{Log}_{10}$  CFU/g) was counted by using spreading technique [27], psychrotrophic bacterial count was counted as mentioned by Hitchins et al. [28],

enumeration of coliforms and isolation of suspected *E. coli* according to FAO [29], isolation of Salmonellae [29] and identification of suspected colonies [30], enumeration of *S. aureus* [31] as well as identification of suspected *S. aureus* colonies as described by Banchroft [32] to evaluate the hygienic conditions applied during processing, handling and preparation of beef burger.

#### **3.2.4. Histological examination:**

Duplicate blocks from all the examined samples were firstly fixed in formalin 10% for twenty four hours, and then washed under running water overnight. In addition to dehydrating the fixed samples in a chain of increasing ethanol concentration, Xylene cleaning followed by embedding in paraffin for 24 hours at 56°C in a hot air oven then blocks were sectioned and stained with Haematoxyline and Eosine according to the technique mentioned by Banchroft et al [33].

#### **3.2.5. Statistical Analysis:**

SPSS statistics 23.0 for windows was used to analyze the collected data in three replicates. Results are showed as mean± Standard error (SD).

### **4. Results and discussion**

#### **4.1. Sensory evaluation of commercially produced beef burger**

Sensory quality evaluations are important for grading, controlling and scoring raw materials and finished products for the investigation of the factors influencing their flavor and odor [34]. Moreover, sensory attributes of meat and meat products are broadly considered to be the most significant determinant factor of consumer preference, with special consideration to appearance, flavor, and texture [35]. Results of sensory evaluation of commercially produced raw beef burger were presented in **figure (1)**. The results indicated low sensory panel scores for appearance, color, forming, binding, consistency, odor and comminution (below 4). Consequently, the overall acceptability

for these samples was low as they scored (3.42). Concerning the sensory analysis of cooked beef burger that showed in **figure (2)**, it was clear that the overall acceptability was obviously low (3.36) probably due to the marked decrease in all the investigated sensory attributes especially flavor, tenderness and juiciness. These results were not copy with E.S.S. (1688/2005) [36] which stated that the final products must have no abnormal odor or taste. Generally, low panel scores for all attributes of examined beef burger indicate low quality raw materials, bad hygienic conditions during processing and storage. The low flavor score may be due to the lipid oxidation [37] and protein degradation [38] during frozen storage and during cooking process. The results obtained were consistent with Ramarathnam and Rubin [39] who found that the overall acceptance of meat products depends mainly on their flavor (taste and odor). The photographs of marketed raw beef burger (**Photo 1**) showed varieties of abnormal colors ranged

from brown to grey, loss of marbling and deviations from the normal shape of burger (discs) with fringe formation which indicated technical problems during the forming step, as well as fine comminution of the product which will affect both consistency of the raw product as well as tenderness of the cooked product. In this regard, the color of meat and meat products is a fundamental physical property, and it is commonly used as a grading indicator for composition, processing, quality, and formulations, due to its correlation with chemical, physical, and sensory characteristics [40]. Moreover, color is the first perception that makes the consumer purchases the product. As a result, it is used to determine whether a food is accepted or rejected [41]. On the other hand, Sánchez et al. [42] added that the distribution and variations of color as well as marbling appearance are also responsible for stable and attractive color.

#### **4.2. Chemical analysis of marketed beef burger samples:**

Proximate chemical analysis of raw market samples (**Table 1**) indicated that the mean value of protein content in beef burger was low while moisture and ether extractable fat content were high. The mean values were 66.12, 8.80 and 20.45% for moisture, protein and ether extractable fat in beef burger respectively. The unacceptably higher moisture and fat content with low protein contents may be due to the use of high percentage of non-meat ingredients in the burger formulation. The obtained protein and moisture results are far away from the E.S.S. regulations stated by (1688/2005) [36] where the protein content is about 15%, moisture content is about 60% while the fat content of marketed samples was slightly high with respect to that stated by E.S.S (1688/2005) [36] where the fat content is about 20%. These results were in harmony with those achieved by Edris et al. [5] who found that moisture, protein, and fat concentrations in examined beef burger were 61.28, 15.22, and 19.80% respectively. The low protein content in the

examined marketed beef burger can be attributed to the use of trimmings, as well as substitution of cheaper non meat components for meat proteins, since meat proteins are relatively more expensive than non-meat components [43]. On the other hand, high fat content in the examined samples may be attributed to addition of external foreign fat or improper formulation [44].

Deterioration criteria of marketed beef burger were showed in table (1) indicated that the mean pH, Thiobarbituric acid (TBA) (mg malomaldehyde %) and Total Volatile Base Nitrogen (TVBN) (mg %) levels were 5.60, 0.66 and 18.74 respectively. These results are presented in the upper border of the permissible limit stated by E.S.S. (1688/2005) [36]. The pH value of examined beef burger were similar to results obtained by Edris et al. [5] which was 5.97 and slightly lower than results recorded with El-Fakhrany et al. [45] who found pH value were 6.10. While, the values of TBA presented by Edris et al. [5]



were lower than our results (0.11 mg malonaldehyde/kg). A high TBA value can be attributed to high lipid content which may be prone to oxidation during bad freezing storage condition, or using low quality fat or oil in beef burger formulation. On the other hand, TVBN values obtained by Edris et al. [5] were lower than the obtained results (10.15 mg %). The high TVBN in marketed samples may be attributed to protein breakdown during frozen storage period or using low quality protein in the commercially produced beef burger or may be due to high aerobic bacterial count.

Evaluation of cooking characteristics of marketed beef burger (Table 1) indicated that the mean values of cooking loss, moisture retention, fat retention, diameter reduction, shrinkage and water holding capacity percentages were 23.66, 31.50, 83.52, 17.31, 19.76 and 75.15% respectively.

#### **4.3. Bacteriological examination of marketed beef burger**

The mean value of APC, psychrotrophic, presumptive Staphylococci, coliforms and Enteropathogenic *E. coli* counts ( $\log_{10}$  CFU/g) for marketed beef burger (Table 2) were 7.69, 6.09, 5.43, 3.84 and 2.25 respectively. these results were higher than those resulted showed by Dawson and Gartner [46] who found that mean value of APC, psychrotrophic, staphylococci, coliforms and fecal coliform count ( $\log_{10}$  CFU/g) were (4.47, 4.25, 1.01, 1.28). The incidence of *S. aureus*, *Salmonellae* and *E. coli* in marketed beef burger were 40%, 0% and 60% respectively where all the isolated *E. coli* was belonging to the serovar O<sub>114</sub>:K<sub>90</sub>. All these results are not in accordance with ESS 1688/2005 [36]. However, Shaltout et al. [47] found that the incidence of *E. coli* in examined beef burger samples from different places were 5.7% and can isolate O<sub>55</sub>:K<sub>59</sub>, O<sub>125</sub>:K<sub>59</sub>, O<sub>55</sub>:K<sub>59</sub>, O<sub>125</sub>:K<sub>59</sub>, O<sub>126</sub>:K<sub>71</sub>, O<sub>55</sub>:K<sub>59</sub>, O<sub>125</sub>:K<sub>59</sub>, O<sub>111</sub>:K<sub>58</sub> and O<sub>126</sub>:K<sub>7</sub>. Also salmonellae failed to be isolated by Shaltout et al. [47]. The high

bacterial count for all examined microorganisms indicates poor hygiene during processing, distribution, cross contamination or due to using contaminated water during slaughtering and evisceration [48].

#### **4.4. Histological examination of marketed beef burger**

The histological details of marketed beef burger illustrated in photo (2) indicated that skeletal muscle (A, B, C) was the predominant structure and these muscle fibers loss its organizational structure as signs for autolysis. Different tissues were observed as wall of blood vessels (D, E, F) and surrounding connective tissue (G), cartilage (H), adipose tissue (I, J, K), bone (L, M,N), and tissue of plant origin (O, P) were detected. These results agree with those detected by Mohamed et al. [49] and Malak et al. [50] who found low amounts of muscle, high amount of fat and connective tissue as well as presence of bone and cartilage. These finding can be attributed to the using of low quality of used raw

materials as well as addition of mechanically deboned meat (MDM) [51] which constitutes a public health hazards leading to food borne infection and intoxication beside the physical and chemical hazards.

#### **5. Conclusion**

The investigation made on the marketed beef burger samples revealed that high proportion of the product suffer from sensory and technological problems such as appearance, color, binding, forming and overall acceptability. Moreover, Most of the investigated marketed beef burger samples were higher than the permissible limit stated with the Egyptian Standard Specifications especially with the nutritional attributes (very low protein and high moisture and fat content) as well as very high bacteriological load. The cooking characteristics of marketed beef burger samples showing a pronounced decrease of water holding capacity, moisture retention and fat retention with subsequent diameter reduction and shrinkage. The histological

analysis of the marketed beef burger samples revealed a presence of many abnormal foreign tissues such as bones, cartilages and blood vessels. Consequently, In order to improve the safety and quality of meat products, it is necessary to raise consumer awareness and enforce strict hygienic regulations on meat processing plants. Beef burgers should also be produced from high quality raw materials and be handled in a sanitary manner during slaughtering, processing, packaging, and distribution.

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**7. Conflict of interest:** No conflict of interest of any authors could be mentioned

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**Table 1:** Chemical analysis of marketed beef burger (n=100)

	<b>Mean</b>	<b>SE±</b>	<b>Maximum</b>	<b>Minimum</b>
<b>Chemical analysis</b>				
<b>Moisture %</b>	66.12	0.43	70.80	61.00
<b>Protein %</b>	8.80	0.25	15.29	2.88
<b>Fat %</b>	20.45	0.43	26.50	13.29
<b>Deterioration criteria</b>				
<b>pH</b>	5.60	0.05	6.50	5.10
<b>TVBN</b>	18.74	0.32	27.00	11.37
<b>TBA</b>	0.66	0.02	1.31	0.35
<b>Physicochemical characteristics</b>				
<b>WHC</b>	75.15	0.98	91.35	63.95
<b>Shrinkage</b>	19.76	0.90	36.74	9.57
<b>Diameter reduction</b>	17.31	0.51	29.47	9.76
<b>Moisture retention</b>	31.50	0.39	36.17	25.87
<b>Fat retention</b>	83.52	2.29	95.09	76.00
<b>Cooking los</b>	23.66	3.22	42.49	10.49

**n:** Number of examined samples

**SE:** Standard error

**TVBN:** Total Volatile Base Nitrogen (TVBN, mg% sample)

**TBA:** Thiobarbituric acid value (TBA, mg malonaldehyde/kg of sample)

**Table 2:** Bacterial count ( $\log_{10}$  CFU/g) of raw marketed beef burger (n=100)

<b>Log<sub>10</sub>CFU/g</b>	<b>Mean</b>	<b>SE±</b>	<b>Maximum</b>	<b>Minimum</b>
<b>APC</b>	7.69	0.06	12.30	6.00
<b>Psychrotrophes</b>	6.09	0.08	7.25	5.00
<b>Staphylococci</b>	5.43	0.06	6.18	5.00
<b>Coliforms</b>	3.84	0.05	4.32	3.32
<b><i>E. Coli</i></b>	2.25	0.02	2.45	2.04

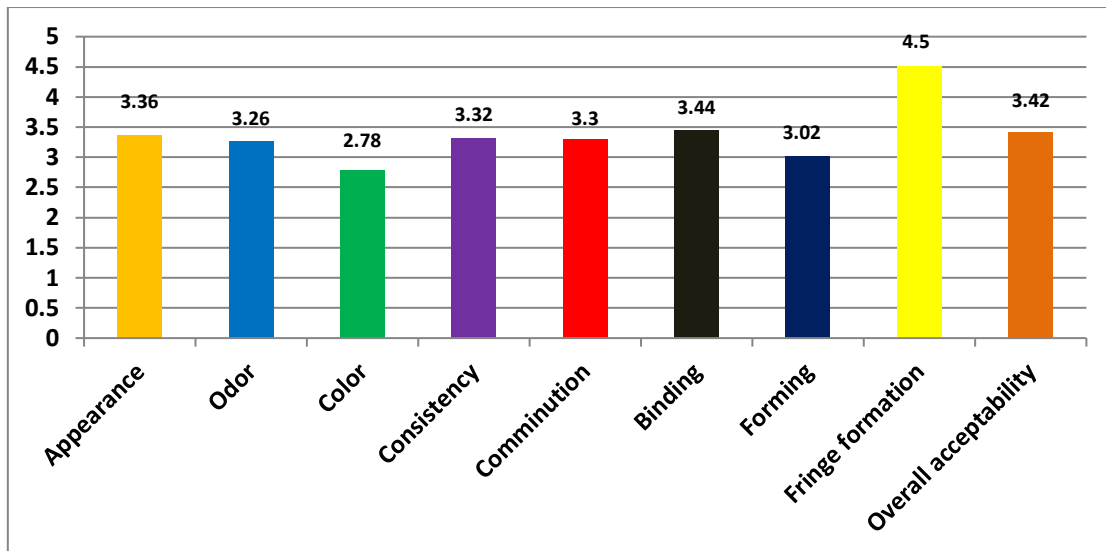
**n:** number of examined samples

**SE:** Standard error

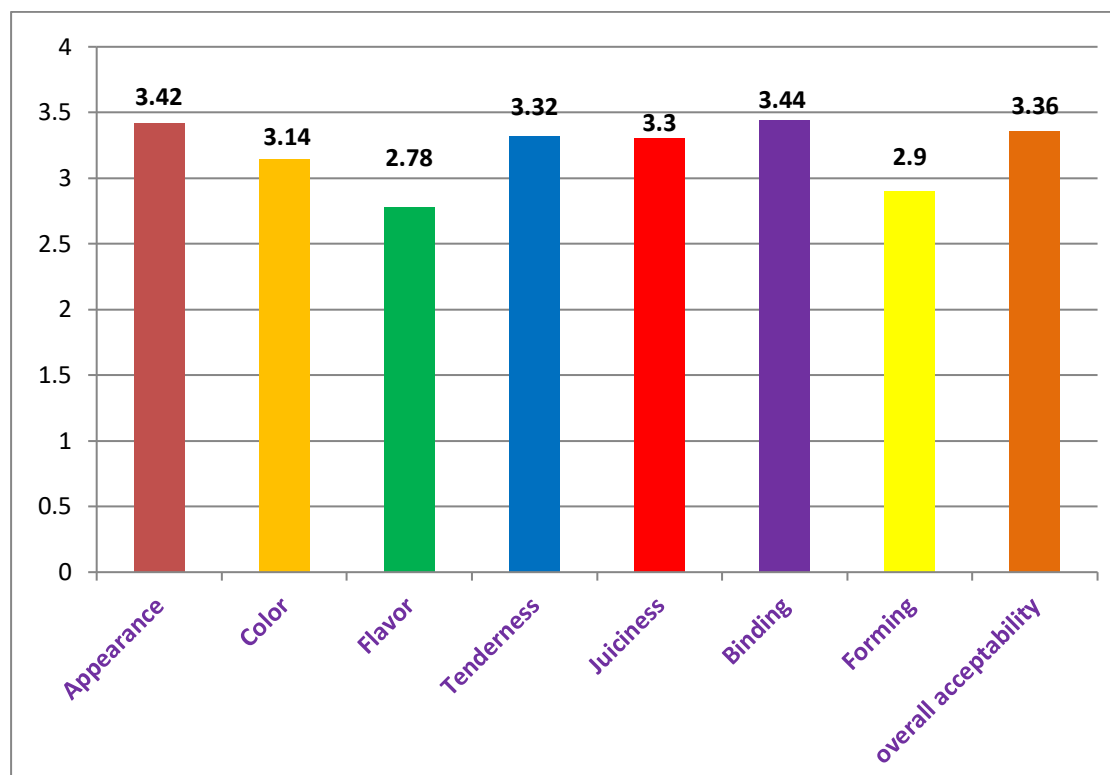
**APC:** Aerobic plate count

**Table 3:** Incidence of pathogens in raw marketed beef burger (n=100)

<b><i>Organisms</i></b>	<b><i>No.</i></b>	<b><i>%</i></b>
<b><i>S. aureus</i></b>	40	40%
<b>Salmonella</b>	0	0
<b><i>E. coli</i></b> <i>E. coli</i> O <sub>114</sub> :K <sub>90</sub>	60	60%



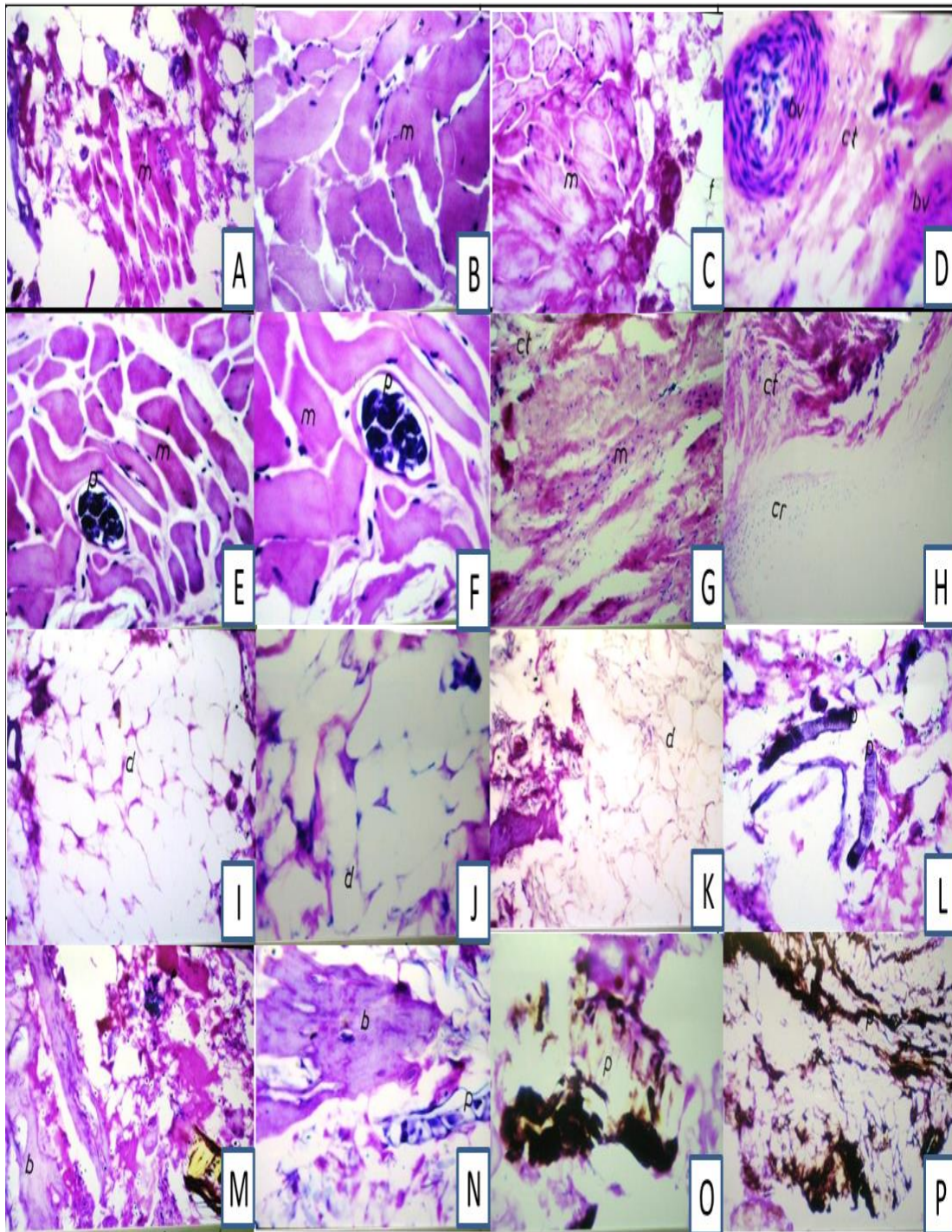
**Fig .1: Sensory panel scores for raw marketed beef burger**



**Fig. 2: Sensory panel scores for cooked marketed beef burger**



**Photo (1) Examined beef burger samples in raw condition showed varieties of abnormal colors ranged from brown to grey, loss of marbling and deviations from the normal shape of burger (discs) with fringe formation.**



**Photo (2):** Histological sections in commercially produced beef burger samples stained with H&E (40 X) (A to P) showed parts of skeletal muscles (m), connective tissue (ct), nerve (n), wall of blood (bv), adipose tissue (d), bone (b), cartilage (cr) and tissue of the plant origin (p).