

Storage Stability of Battered Common Carp Fillets Affected By Incorporation With Herbal Extracts

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Abstract

The work was carried out to evaluate the preservative ability of herbal extracts from (thyme, clove and mint) by their incorporation into batter mixtures of semi-fried battered common carp fish fillets on storage stability during cold storage. The common carp slices were coated with batter mixtures with/without herbal extracts, semi-fried, cooled, packed and subjected to physicochemical, microbiological and sensory evaluation during storage at 4±1°C for 24 days. The all treatments especially containing herbal extracts were had high quality and acceptability at zero time of analysis. Afterthought, the progressive storage period caused releasing ($p<0.05$) of volatile amine nitrogen and lipids deterioration, due to evolution of microbial growth, subsequently gradual descending in acceptability of all treatments. The significant differences ($p<0.05$) in values of pH, TVB-N, TMA-N, PV, TBA, TVBC and TPBC, and the scores of sensory properties were statistically assessed between treatments during cold storage. The spoilage rate in control sample (T1) followed by BHT (T2) treatment and T3 (containing 0.5% thyme extract) was faster than ($p<0.05$) the other treatments. Meanwhile all herbal extracts significantly ($p<0.05$) retarded chemical deterioration, reduced microbial evolution and kept the overall acceptability in T4, T5, T6, T7 and T8 more than the other treatments. The herbal extracts from clove, thyme and mint significantly extended the shelf life and enhanced the sensory properties of semi-fried battered common carp fillets during cold storage period, might be due to their antioxidant and antimicrobial activity. Therefore, they were useful together battering process to prolong the consumption validity and improved the acceptability of the undesirable fresh common carp fillet compared to control sample and BHT treatment.

Keywords: Common carp - herbal extracts - BHT - battering - physicochemical - microbiological - sensory quality.

Introduction

Fish meat has specified composition of elements to be qualified it a suitable food, which contributed to its contents of animal protein that necessary to supply human diet by essential amino acids, essential fatty acids such omega-3 polyunsaturated fatty acid (PUFA), vitamins, and minerals, On opposite of its dearth in connective tissues and saturated fat (EFSA, 2016; Siscovick *et al.*, 2017 and Vilavert *et al.*, 2017). Common carp (*Cyprinus carpio*) is widely cultured, and its production rapidly increasing around the world (Sterniša *et al.*, 2018). In 2017, the production of farmed *C. carpio* was 4.1 million tonnes, as value of 3.69% from globally aquaculture production (FAO, 2019). The fresh carp fishes is oftentimes not acceptable, due to its unwanted appearance, distinctive odor, and earthy or muddy taste, the abundant mucous layer in common carp may be found on skin during marketing, and the presence of intramuscular spines (Varga *et al.*, 2015 and Sterniša *et al.*, 2018). Furthermore, fish is a known highly perishable product, the quality loss and deterioration of the fish primarily start after the bringing of fish to the landing sites, due to many factors as microbial invasion and purification, autolysis, chemical oxidation, mechanical damage and environmental optimization to be encouraged the

growth of microbes (Ghaly *et al.*, 2010; Tahsin *et al.*, 2017; Odeyemi *et al.*, 2018 and Speranza *et al.*, 2021). Thus, it is necessary to promote some suitability fishery products from the carp flesh, in conformity with consumer demands to enhance its consumer acceptability (Sehgal *et al.*, 2011 and Vanitha *et al.*, 2013), as the efforts of fish processing to prepare value-added products from common carp fish, and delay or inhibit the deterioration of fish.

Amongst fish processing techniques, the biodegradable films/ or coatings recently could be enriched by natural preservatives to apply in fresh fish, which able to inhibit the microbial growth and decrease the rate of fish nutrients decomposition, and developed many ready-to-eat or/use fishery products suited with recently consumers' needs, the demand for ready-to-eat or ready-to-use fish products has led to the development of several products diverse in taste, texture and appearance. The battered and breaded products are possessed high appealing of consumers, which are commonly known coated or enrobed products (Chudasama *et al.*, 2019 and Dehghani *et al.*, 2018). A major group among them commanding high consumer appeal is the battered and breaded products. Several active edible films and coatings were applied to fresh fish fillets of rainbow trout, silver carp, grass carp, beluga sturgeon,

salmon, pike-perch, Japanese sea bass, red drum, golden pomfret, hake and other fish species (Socaciu *et al.*, 2018).

The edible coatings are one of technological methods can improve the quality of fresh, frozen, and processed poultry, meat, and seafood products, to reducing moisture loss, lipid oxidation and discoloration. It is also useful to enhance the products appearance, keeping volatile flavors, performing as carriers of food additives such as antimicrobial and antioxidant agents, and reducing oil uptake in battered and breaded products during frying (Badee *et al.*, 2013; Khan *et al.*, 2013 and Mei *et al.*, 2019). Plant extracts and essential oils are traditionally accepted by consumers, offering safe and high quality seafood with consumer acceptance, creates real challenging problems and retarding spoilage, and it could be applied in fish products (Hassoun and Çoban, 2017; Pateiro *et al.*, 2018 and Ehsani *et al.*, 2020). The natural preservatives could apply in fishery products with low temperature, such as refrigeration and edible coatings (Moawad *et al.*; 2017 and Mei *et al.*; 2019). Therefore, this investigation was aimed to offering common carp fillets in competitive high quality fish product throughout battering process incorporated with herbal extracts of thyme (*Thymus vulgaris*), mint (*Mentha sp.*), and dried clove buds (*Syzygium aromaticum*) for improving the storage stability and extend shelf life of battered common carp fish fillets during storage at 4±1°C.

Materials and methods:

Fish samples: About 30 kg from fresh common carp fish (*Cyprinus carpio*) with average weight from 5.00 to 6.00 kg, length from 45-60 cm were purchased from local market of El-Obour city, Qalyoubia Governorate, Egypt, and transported using icebox to Fish Processing and Technology Laboratory, Fish Research Station, El-Kanater El-Khiria, National Institute of Oceanography and Fisheries, Egypt. All fish samples were rewashed, beheaded, eviscerated, skin removed and filleted, then rewashed and drained till use.

Herbals materials: Three types of herbs as fresh mint (*Mentha sp.*), fresh thyme (*Thymus vulgaris*) and dried clove buds (*Syzygium aromaticum*) obtained from local market (Toukh, Qalyoubia, Egypt) were used.

Chemicals and media: Butylated hydroxy toluene (BHT), ethyl alcohol 99.9%, methanol, chloroform, sodium hydroxide, trichloroacetic acid, glacial acetic acid, picric acid, hydrochloric acid, thiobarbituric acid, sodium sulphate anhydrous, toluene, sodium poly phosphate, glycerol and sodium chloride were purchased from Techno lab for trade Co. (Cairo, Egypt). The nutrient agar medium was purchased from International Trade Association (ITA) Co., (El-Kasr El-Aini, Cairo, Egypt).

Ingredient of batter mixture: wheat flour (72% ext.), corn flour, starch, shortening oil, salt, baking

powder, skimmed milk powder, Arabic gum and eggs were purchased from local market (Toukh, Qalyoubia, Egypt).

Methods:

Herbal preparation and extraction: The herbs of thyme, mint and clove were prepared and extracted by ethanol according to the method of "solid-liquid extraction" as described by Sung-Jin *et al.* (2013) and Hermund (2016).

Technological process: The battered common carp fish fillets were produced from fillets with average weight from 800 to 900 g of each, average length from 30 to 35 cm of each and with average thickness 1.3-1.5 cm. Fish fillets were rewashed, drained, cut into slices with dimensions of 2x8x1.3-1.5 cm (width x length x highest, respectively). The batter mixture was prepared as described by Venugopal (2006) by mixing 1 kg of ingredients concluded on 45% wheat flour, 30% corn flour, 5% starch, 5% salt, 3% baking powder, 1% sodium poly phosphate, 1% glycerol, 5% shortening oil, 3% skim milk powder, 1% gum and 1% egg white, respectively with 1- 1.5 liter of cold water by electric stirring device for 5 minutes to obtain smooth dough. The fish slices and batter mixture were divided equally to 8 batches, then the tested herbal extracts and BHT were mixed with batter mixtures to obtain 8 treatments (T) as following: the 1st batch (T1) without any additives (control), the 2nd batch (T2) enriched by 0.1% BHT (w/w), the 3rd and 4th batches (T3&T4) enriched 0.5 and 1% thyme extracts, the 5th and 6th batches (T5&T6) enriched by 0.5 and 1% clove extracts and the 7th and 8th batches (T7&T8) enriched by 0.5 and 1% mint extracts, respectively. Fish slices were separately dipped in batter mixtures, then released after 15 minutes on pored stainless steel board to dispose the batter excess, deep fried at 170°C for 1 minute, cooled to room temperature, packed and stored in polyethylene bags at 4±1°C. Battered fish samples were subjected to chemical, physical, microbiological and organoleptic analysis periodically analyzed intervals 0, 3, 6, 9, 12, 15, 18, 21 and 24 days.

Analytical methods:

Physicochemical analysis: The proximate chemical analysis of the moisture, crude protein, fat and ash contents of investigated samples were determined according to the methods recommended by the AOAC (2000), total carbohydrates was calculated by difference. The pH value was carried out according to the procedure of Abdel-Aziz *et al.* (2019), the total volatile basic nitrogen (TVB-N) and trimethyl amine nitrogen (TMA-N) contents were determined according to the method described by Pearson (1976). Peroxide Value (PV) was determined using the method of Egan *et al.* (1997), and the thiobarbituric acid (TBA) value was measured

according to the method described by **Tarladgis et al. (1960)**.

Microbiological examination: The investigated fish sample were prepared for microbiological investigation to obtain the serial decimal dilutions in saline solutions (0.85%) according to **Heydari et al. (2015)**, then pour plating method was utilized for microbiological analysis of total viable bacterial count (TVBC) by using nutrient agar media (**TM 341**) incubated at $37\pm 1^\circ\text{C}$ for 48 h as described by **Heydari et al. (2015)**. Total psychrophilic bacterial count (TPBC) was determined by using commercial nutrient agar media (**TM 341**) incubated at $5\pm 1^\circ\text{C}$ for 7 days as described by **Sönmez et al. (2020)**, all results are expressed as log of the colony forming units (cfu) per gram of sample, all analyses were done in triplicate.

Sensory evaluation: The sensory evaluation of semi-fried battered common carp fillets was conducted for color, odor, taste, texture and overall acceptability a panel of judges using 9-point hedonic scale according to the procedure of **Meilgaard et al. (2007)** for all samples during storage time intervals.

All sensory properties of tested battered common carp fish slices were evaluated by 10 trained panelists from (National Institute of Oceanography and Fisheries, El-Kanater El-Khiria).

Statistical analysis: Three replicates of each trial were performed for each parameter and values expressed as the mean \pm SD. Statistical analyses were performed using Statistical package social science (SPSS) version 16.00 for Windows. Analysis of variance (ANOVA) was used and statistical significance was set at $p < 0.05$ to detect the significant effect between means during storage periods according to **Duncan' test (1955)** to separate means at a probability level of $p < 0.05$.

Results and Discussion:

The chemical composition (w/w) of fresh and semi-fried battered common carp fish fillets was shown **Table (1)**. The moisture, protein, fat, ash and total carbohydrate contents were 73.01, 13.80, 11.57, 1.00 and 0.62% of common carp flesh, respectively. These results are in agreement with those mentioned by (**Khidhir, 2011 and Abdulrahman et al., 2019**).

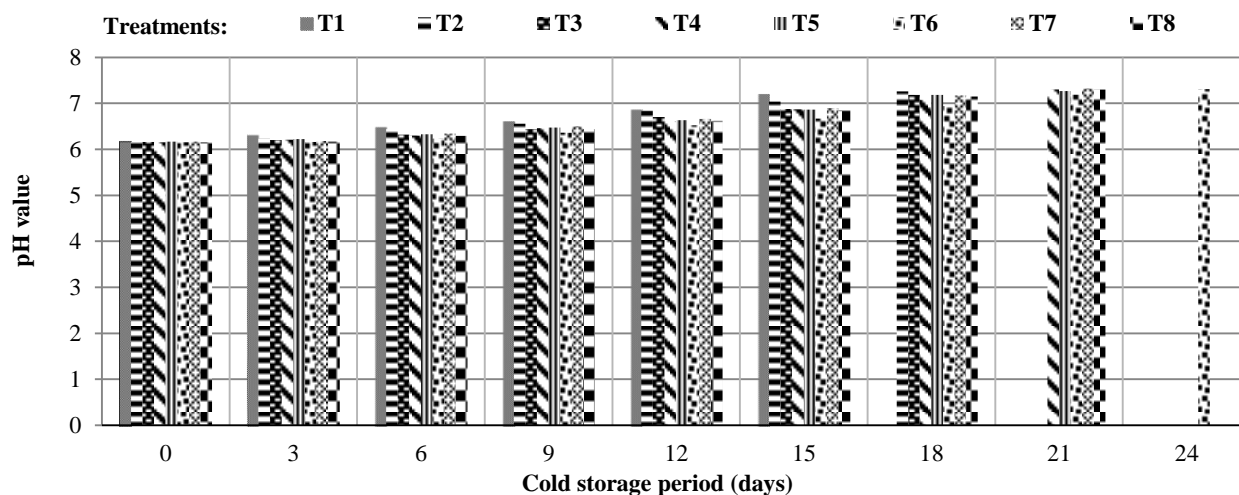
Table 1. Chemical composition of fresh and raw semi-fried battered common carp fish fillets (g/100g on wet weight bases).

Fish samples	Components (%)					Energy value Kcal/100g
	Moisture	Protein	Fat	Ash	*Total carbohydrate	
Fresh common carp flesh	73.01 \pm 1.12	13.80 \pm 3.37	11.57 \pm 3.47	1.00 \pm 0.22	0.62 \pm 0.05	161.81
Raw semi-fried battered common carp slices	62.55 \pm 1.65	17.25 \pm 1.98	14.27 \pm 4.39	1.20 \pm 0.12	4.73 \pm 0.16	216.35

Data are the mean \pm SD, n = 3 *carbohydrates percent calculated by difference.

Concerning the changes in composition of fish fillets due to battering process, the battering process was caused a greatest decreasing in moisture contents to 62.55%, in opposite of the increase of protein, fat, ash and total carbohydrate contents to 17.25, 14.27, 1.20 and 4.73%, respectively in semi-fried battered common carp fillets, compared to fresh fish meat. The increase in protein, lipid, and ash content in cooked fillets is explained by the water loss and oil uptake due to the frying process at 170°C for 1 minute (**Khanipour et al., 2014; Bandre et al., 2018; and Golgolipour et al., 2019**), the increase in carbohydrate content in battered fillets may be gained from coating materials such wheat flour, corn flour, starch and gums (**Badee et al., 2013; Hassan et al., 2013; Khanipour et al., 2014 and Elsayed et al., 2016**).

pH value: The pH value is usually a good index for quality assessment in fish and fish products (**Abdel-Aziz et al., 2019**). The changes in pH values of semi-fried battered common carp fillets affected by herbal extracts were investigated during storage at $4\pm 1^\circ\text{C}$ as presented in **Fig. (1)**. The pH values at zero time of treatments as well as T1, T2, T3, T4, T5, T6, T7 and T8 were 6.17, 6.17, 6.16, 6.15, 6.17, 6.15, 6.17 and 6.15, respectively. The gradual increment in pH values ($p < 0.05$) of all treatments was observed during cold storage period affected by treatment type. The increase in pH values may be due to the action of endogenous or microbial enzymes such as protease that cause an increase in alkaline volatile bases compounds as well as (e.g., TVB-N, TMA-N, ammonia, etc.) during prolonged storage (**Li et al., 2012; Viji et al., 2014 and Li et al., 2017**).



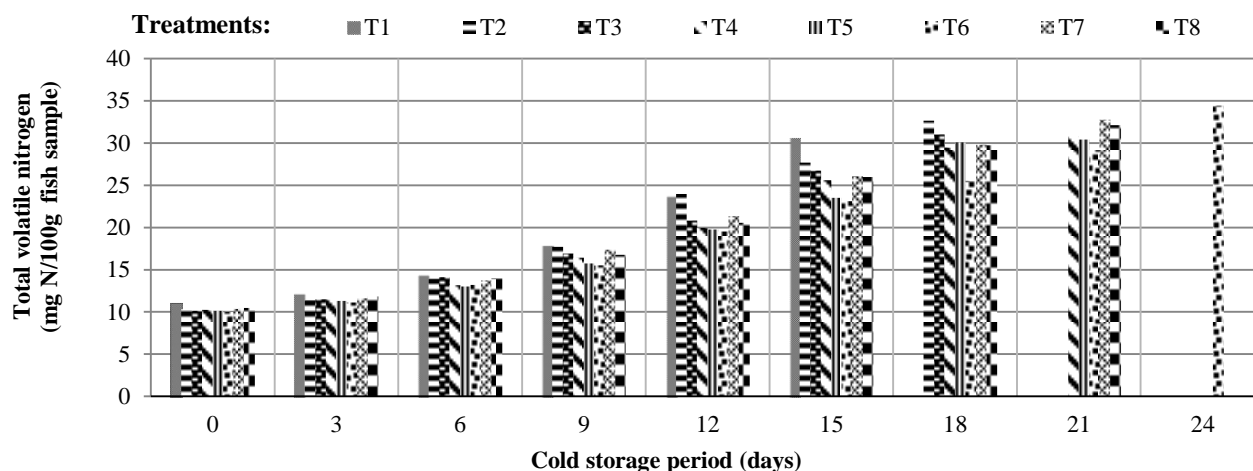
T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (1): pH values of semi-fried battered common carp slices during storage period at $4\pm 1^\circ\text{C}$.

Insomuch, the significant difference ($p < 0.05$) between pH values of treatments was assessed during cold storage, the pH values of control sample (T1) and T2 quickly increased than ($p < 0.05$) the other treatments to 7.18 and 7.26 in the 15th and 18th day of cold storage respectively, to be exceeded the value of 7.1 is an indicative for outset of fish decomposition according to Özyurt *et al.* (2009). Meanwhile, the pH values of treated samples with herbal extracts were lower than T1 and T2 treatments. It is worth mentioning that, however all herbal extracts retarded the increase of pH during cold storage compared to control sample and T2 ($p < 0.05$), but the semi-fried common carp fish fillets treated with clove extracts were exhibited the lowest values ($p < 0.05$) during cold storage, this is might cause from their antimicrobial activity due to their content of phenolic compounds. Also, the difference between treated fillets with herbal extracts could be suggested from the difference between themselves in their contents of bioactive compounds. This results are corresponded with those obtained by Hassan *et al.* (2013); Khanipour *et al.* (2014) and Abdel-Aziz *et al.* (2019).

Total volatile basic nitrogen (TVB-N): The TVB-N and TMA-N are widely used to determine the seafood quality since it has a direct relation with the

microorganism growth and enzymatic activity (Amin, 2012 and Moawad *et al.*, 2017). The changes in TVB-N contents of semi-fried battered common carp slices treated with herbal extracts during storage at $4\pm 1^\circ\text{C}$ are shown in Fig. (2). The TVB-N contents of semi-fried battered common carp treatments were 11.01, 10.31, 10.14, 10.26, 10.14, 9.98, 10.37 and 10.48 mg N/100g for T1, T2, T3, T4, T5, T6, T7 and T8, respectively at zero time of cold storage at 4°C , however the difference in initial TVB-N values between treatments was significant ($p < 0.05$), but all TVB-N values referred to high quality of raw battered treatments. Afterward, the TVB-N values of all treatments gradually increased ($p < 0.05$) with the advancement of cold storage period with different rates affected by the type of treatment, the significant differences ($p < 0.05$) in TVB-N values were detected between treatments. Therefore, the TVB-N formation showed the quickest rates ($p < 0.05$) in control sample to be 30.64 mg N/100g in the 15th day, followed by T2 and T3 to be 32.65 and 31.03 mg N/100g, respectively in the 18th day of cold storage, while the lowest rate ($p < 0.05$) of TVB-N was observed in T6 that contain 1% clove extract.



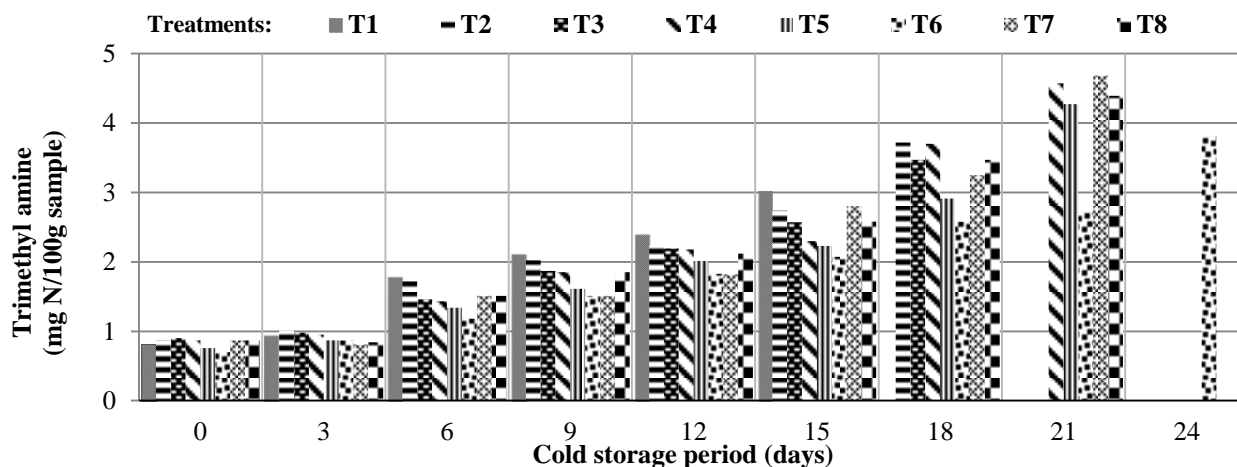
T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (2): Total volatile nitrogen values of semi-fried battered common carp slices during cold storage period at $4\pm 1^\circ\text{C}$.

According to **ESS (2019)** reported the maximum acceptable limit of TVB-N in chilled fish is 30 mg N/100g, the semi-fried battered common carp was accepted for 12 days for control sample (T1), 15 days for the T2 and T3, 18 days for T4, T5, T7 and T8, while T6 had the highest period of acceptance for 21 days during cold storage. Thus, it could be said that there was a positive effect for investigated herbal extracts and BHT as preservative additives, especially that obtained from clove extracts compared control, whereas their application caused reduction in nitrogenous volatile basic compounds of semi fried battered fish fillets and prolonged their shelf life compared to control. The elevation of TVB-N during storage attributed to the endogenous enzymatic activity or bacterial growth (**Orban et al., 2011 and Raeisi et al., 2020**), the responsibility of herbal extracts to be reduced the formation of TVB-N in treated samples associated with their antimicrobial activity resulted from phenolic compounds. The variance in this activity closely related to the differences in their contents of phenolic compounds that affected by the type and level of herbal extracts whereof the difference between treated samples appeared during cold

storage. The obtained results are agreement with those found by **Badee et al. (2013); Khanipour et al. (2014); Sarabi et al. (2017); Esmaeli et al. (2019) and Raeisi et al. (2020)**.

Trimethylamine nitrogen (TMA-N): The changes in TMA-N values of semi-fried battered common carp fish slices treated with some herbal extracts during cold storage at $4\pm 1^\circ\text{C}$ are shown in **Fig. (3)**. At the beginning of storage, the TMA-N values were 0.81, 0.87, 0.90, 0.87, 0.76, 0.70, 0.87 and 0.87 mg /100g for T1, T2, T3, T4, T5, T6, T7 and T8, respectively. Thereafter, the TMA-N contents continuously increased in all treatments during cold storage period with different rates influenced by kind and ratio of herbal extracts. The control sample was exhibited the highest increasing rate ($p<0.05$) during the cold storage period, compared to the other treatments, in contrast the lowest increasing rate was observed in T6. This increment in the TMA-N content could be attributed to the TMA-N oxide, naturally present in fish tissues, which chemically reduced to the TMA-N by either SH group existing in fish protein and to the formation of the TMA-N itself from betaine and choline which also naturally occur in fish tissues (**Rodríguez et al., 2009**).



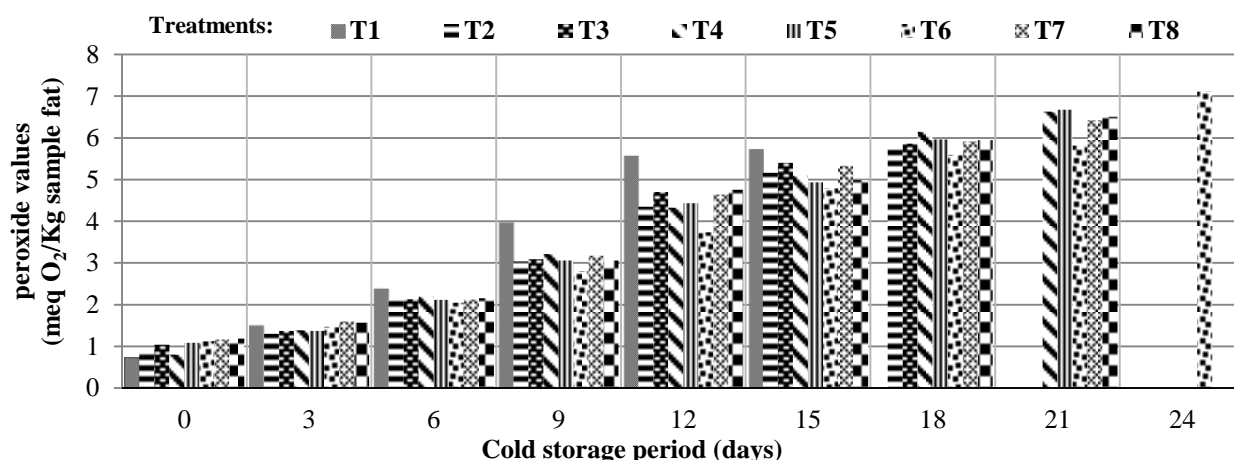
T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (3): Trimethyl amine values of semi-fried battered common carp slices during cold storage period at $4\pm 1^\circ\text{C}$.

It is worth mentioning that, the significance of differences in TMA-N values amongst all treatments were inconsistent during the wholeness of cold storage periods, and the dominant statistical trend was not significant ($p > 0.05$), the TMA-N of all treatments was remained with values below the acceptable limit of 10 mg/100 g according to **ESS (2019)**. Although, all tested herbal extracts of thyme, clove and mint were more effective to be reduced volatile amines formation in semi-fried battered common carp fillets with ratios 0.5 and 1% during cold storage compared to control sample and BHT treatment, nevertheless the best reduction obtained from treated common carp samples with 1% clove extract than the other treatments. These results are in agreement with those obtained by **Hassan *et al.* (2013)**; **Badee *et al.* (2013)**; **Sarabi *et al.* (2017)** and **Olalekan (2019)**.

Peroxide value (PV): The PV is a measure typically quantifying the primary lipid oxidation of fish

products, particularly the hydroperoxides, and it has been the most commonly employed chemical assay for evaluating the oxidative stability of fats and oils (**Chaijan, 2011 and Uçak *et al.*, 2011**). The changes in PV of semi-fried battered common carp fish slices treated with some herbal extracts during cold storage at $4\pm 1^\circ\text{C}$ are presented in **Fig. (4)**. At zero time, the peroxide values of T1, T2, T3, T4, T5, T6, T7 and T8 were 0.73, 0.90, 1.04, 0.80, 1.08, 1.12, 1.16 and 1.18 meq O_2/Kg fat, respectively with significant difference ($p < 0.05$) between treatments, but all fresh battered fish were very good fish products according to **Uçak *et al.* (2011)** they reported that the peroxide value is expected to be below 2 mmol active oxygen/kg lipids in very good material, and not exceed to 5 mmol active oxygen/kg lipids in good material. The occurrence of lipid oxidation in all treatments progressively rose ($p < 0.05$) with the progression of cold storage periods, affected by treatment type.



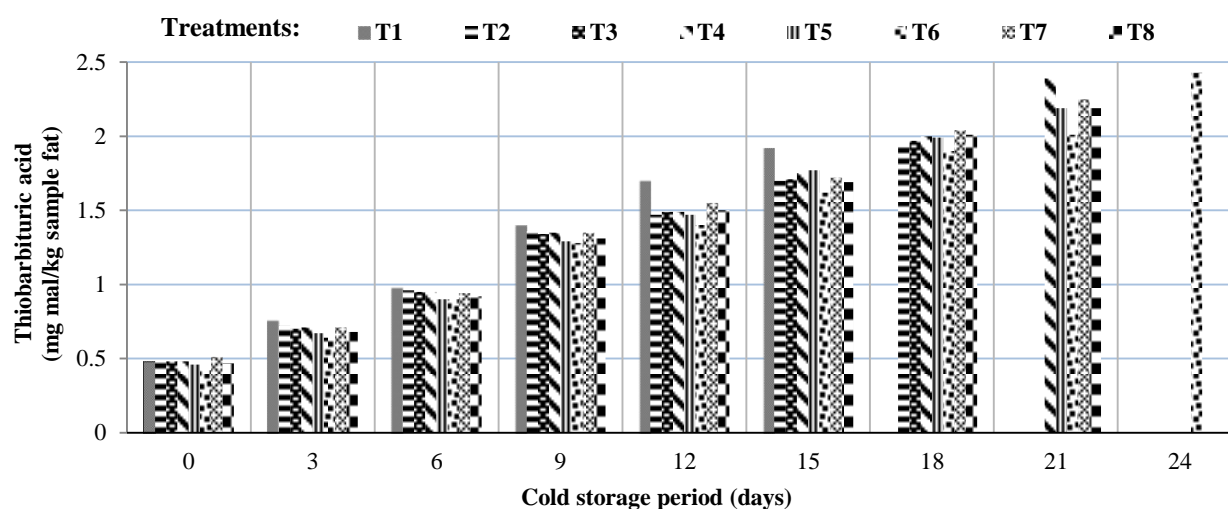
T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (4): Peroxide values of semi-fried battered common carp slices during cold storage period at $4\pm 1^\circ\text{C}$.

The highest increase rate ($p < 0.05$) of PV was found in T1 to 5.71 meq O_2/Kg after 15 days of cold storage, meanwhile the peroxide contents of the other treatments were lower ($p < 0.05$) than found in control sample. Nevertheless, the lowest raise rate of PV and the highest retarding of oxidation were obtained from treating slices by 1% clove extract compared to the other treatments, the highest extracts concentration was the lowest PV value during cold storage. The peroxide values of all treatments were below the maximum acceptable level of 10 meq O_2/kg fat during cold storage period according to (Connell, 1995). Therefore, applying clove, thyme and mint extracts in semi-fried battered common carp fillets were useful to delay lipid oxidation and prevention off-flavor during cold storage. This effect might be related to the antioxidant efficacy of

phenolic compounds in tested extracts, the difference in phenolic compounds between extracts might be responsible for the difference in PV amongst treated samples with herbal extracts. This results are in agreement with those found by Hassan *et al.* (2013); Sarabi *et al.* (2017); Esmaeli *et al.* (2019) and Ehsani *et al.* (2020).

Thiobarbituric acid value (TBA): TBA is an index widely used for assessing secondary oxidation in order to assess the degree the series lipid oxidation in fish, which is also recognized as an indicator of fish freshness (Sallam, 2007 and Alparslan *et al.*, 2014). The changes in TBA values of semi-fried battered common carp slices treated with herbal extracts during cold storage at $4 \pm 1^\circ C$ are presented in Fig. (5).



T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (5): Thiobarbituric acid values (mg mal/kg) of semi-fried battered common carp slices treatments during cold storage period at $4 \pm 1^\circ C$.

The TBA values of T1, T2, T3, T4, T5, T6, T7 and T8 were 0.48, 0.49, 0.48, 0.48, 0.46, 0.42, 0.51 and 0.47 mg mal/kg, respectively at the beginning of storage. Thereafter, the TBA values of all treatments were gradually increased ($p < 0.05$) during progressively cold storage periods, control sample presented the highest rate of increase to 1.92 mg mal/kg in the 15th day, meanwhile the lowest TBA value was 1.62 mg mal/kg for T6, the difference amongst all treated fillets was non-significant ($p > 0.05$) during storage periods. Concerning the indices of lipid oxidation such as PV and TBA, all the herbal extracts and BHT were presented acceptable efficacy ($p < 0.05$) where they could be retarded lipid oxidation in battered common carp fish fillets during cold storage, nevertheless the clove extract, and the highest concentrations of investigated herbal extracts were preserved fat quality better than the other treatments. In addition to these results, TBA values of all treatments didn't

exceed the acceptable limit of 4.5 mg mal /kg as set by (ESS, 2019). This results are similar with those finding by Hassan *et al.*, (2013); Badee *et al.* (2013); Khanipour *et al.* (2014); Sarabi *et al.* (2017); Esmaeli *et al.* (2019); Ehsani *et al.* (2020) and Raeisi *et al.* (2020).

Total viable bacterial count (TVBC): The total viable count is a traditional and helpful indicator used to assess the degree of fish freshness, and for predicting the remaining shelf life of fish (Cheng *et al.*, 2015). The changes in total viable counts (\log_{10} cfu/g) of semi-fried battered common carp fish slices treated with herbal extracts during storage at $4 \pm 1^\circ C$ are seen in Table (2). The initial TVBC of semi-fried battered common carp were 3.63, 3.58, 3.67, 3.56, 3.57, 3.65, 3.55 and 3.53 (\log_{10} cfu/g) for T1, T2, T3, T4, T5, T6, T7 and T8, respectively. Thereafter, all bacterial counts of all treatments were increased ($p < 0.05$) during cold storage period affected by treatment type, the highest rate of

increase was observed in the control sample followed by BHT treatment and reached to 6.47 and 6.49 log₁₀ cfu/g after 15 and 18 days of cold storage, respectively. While all herbal extracts particularly

obtained from clove retarded this increment during cold storage in T3, T4, T5, T6, T7 and T8, this effect may be due to the antibacterial activity contributed to the presence of bioactive compounds.

Table 2. Total viable bacterial counts as log₁₀ cfu/g of semi-fried battered common carp slices during cold storage period at 4±1°C.

Storage period (days)	Treatments							
	T1	T2	T3	T4	T5	T6	T7	T8
0	3.63 ^{Fa} ±0.03	3.58 ^{Da} ±0.04	3.67 ^{Ea} ±0.03	3.56 ^{Fa} ±0.13	3.57 ^{Fa} ±0.05	3.65 ^{Ha} ±0.12	3.55 ^{Fa} ±0.08	3.53 ^{Fa} ±0.03
3	4.70 ^{Ea} ±0.03	4.58 ^{Cab} ±0.10	4.59 ^{Dab} ±0.12	4.58 ^{Eab} ±0.11	4.43 ^{Ec} ±0.06	4.51 ^{Gbc} ±0.05	4.64 ^{Eab} ±0.02	4.59 ^{Dab} ±0.01
6	4.88 ^{Da} ±0.03	4.78 ^{Cb} ±0.02	4.79 ^{Db} ±0.07	4.78 ^{Db} ±0.01	4.75 ^{Dbc} ±0.05	4.64 ^{Gd} ±0.05	4.77 ^{DEbc} ±0.02	4.70 ^{Dcd} ±0.04
9	5.04 ^{Ca} ±0.02	4.94 ^{Cb} ±0.02	4.95 ^{CDb} ±0.07	4.94 ^{Db} ±0.01	4.91 ^{Dbc} ±0.05	4.80 ^{Fd} ±0.05	4.93 ^{Dbc} ±0.02	4.86 ^{Dcd} ±0.04
12	5.68 ^{Ba} ±0.05	5.50 ^{Bb} ±0.05	5.27 ^{BCc} ±0.13	5.27 ^{Cc} ±0.07	5.25 ^{Cc} ±0.02	5.27 ^{Ec} ±0.08	5.35 ^{Cbc} ±0.14	5.48 ^{Cb} ±0.08
15	6.47 ^{Aa} ±0.15	5.94 ^{Bb} ±0.59	5.62 ^{Bbc} ±0.20	5.42 ^{BCc} ±0.04	5.50 ^{BCbc} ±0.17	5.48 ^{Dbc} ±0.11	5.51 ^{BCbc} ±0.18	5.50 ^{Cbc} ±0.03
18	R*	6.49 ^{Aa} ±0.35	6.20 ^{Aab} ±0.52	5.61 ^{Bc} ±0.07	5.68 ^{Bc} ±0.15	5.66 ^{Cc} ±0.09	5.69 ^{Bc} ±0.16	5.85 ^{Bbc} ±0.16
21	R*	R*	R*	6.26 ^{Aa} ±0.23	6.35 ^{Aa} ±0.33	5.81 ^{Bb} ±0.02	6.36 ^{Aa} ±0.33	6.34 ^{Aa} ±0.16
24	R*	R*	R*	R*	R*	6.43 ^A ±0.06	R*	R*

T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract. Means of triplicate ± Standard deviation on wet weight basis. Mean values in the same row (small letter) or column (capital letter) with the same letter are not significantly different (at $p < 0.05$). R = rejected.

The highest reduction of total bacteria was significantly obtained from the levels of 1% of herbal extracts compared to 0.5%, especially obtained from clove extract, which retarded the excess of TVBC over than upper acceptability limit of 6 log₁₀ cfu/g for chilled fish according to **ESS (2019)** till 21 days of storage, in comparison with previous periods for the other treatments. This results are in agreement with those obtained by **Badee et al. (2013)**; **Hassan et al. (2013)**; **Ibrahim et al. (2015)**; **Raeisi et al. (2020)** and **Morachis-Valdez et al. (2021)**.

Total psychrophilic bacterial count (TPBC): The psychrotrophic bacteria are the important group of aerobic microorganisms responsible for the spoilage of fish stored in cold temperatures (**Sallam et al., 2007**), causing changes in odor, texture and flavor as a result of various metabolic compounds such as

ketones, aldehydes and biogenic amines volatile sulfides (**Özogul et al., 2013**). Therefore, the changes in TPBC counts (log₁₀ cfu/g) of semi-fried battered common carp fish slices treated with some herbal extracts were determined during refrigerated storage as shown in **Table (3)**. At zero time, the TPBC of T1, T2, T3, T4, T5, T6, T7 and T8 were 2.96, 2.65, 2.76, 2.67, 2.84, 2.91, 2.72 and 2.80 (log₁₀ cfu/g), respectively, the initial counts showed significant difference ($p < 0.05$) between control sample (T1) and the other treatments. The TPBC of all treatments significantly increased ($p < 0.05$) during cold storage period. The highest increase rate ($p < 0.05$) was observed in control sample followed by BHT treatment and reached to 6.05 and 6.22 log₁₀ cfu/g in the 15 and 18th day, respectively compared to the other treatments.

Table 3. Total psychrophilic bacterial counts (log₁₀ cfu/g) of semi-fried battered common carp slices during cold storage period at 4±1°C.

Storage period (days)	Treatments							
	T1	T2	T3	T4	T5	T6	T7	T8
0	2.96 ^{Da} ±0.12	2.65 ^{Dc} ±0.16	2.76 ^{Eabc} ±0.15	2.67 ^{Ebc} ±0.19	2.84 ^{Fabc} ±0.06	2.91 ^{Eab} ±0.13	2.72 ^{Fabc} ±0.12	2.80 ^{Eabc} ±0.04
3	3.70 ^{CDa} ±0.02	3.38 ^{Dbcd} ±0.06	3.29 ^{DEd} ±0.13	3.45 ^{DEbc} ±0.04	3.51 ^{EFb} ±0.08	3.34 ^{DEcd} ±0.11	3.42 ^{EFbcd} ±0.02	3.30 ^{Ecd} ±0.07
6	4.35 ^{BCa} ±0.02	4.28 ^{Ca} ±0.06	4.24 ^{CDa} ±0.13	4.16 ^{CDa} ±0.04	4.19 ^{DEa} ±0.08	4.11 ^{CDa} ±0.11	4.21 ^{DEa} ±0.02	4.20 ^{Da} ±0.07

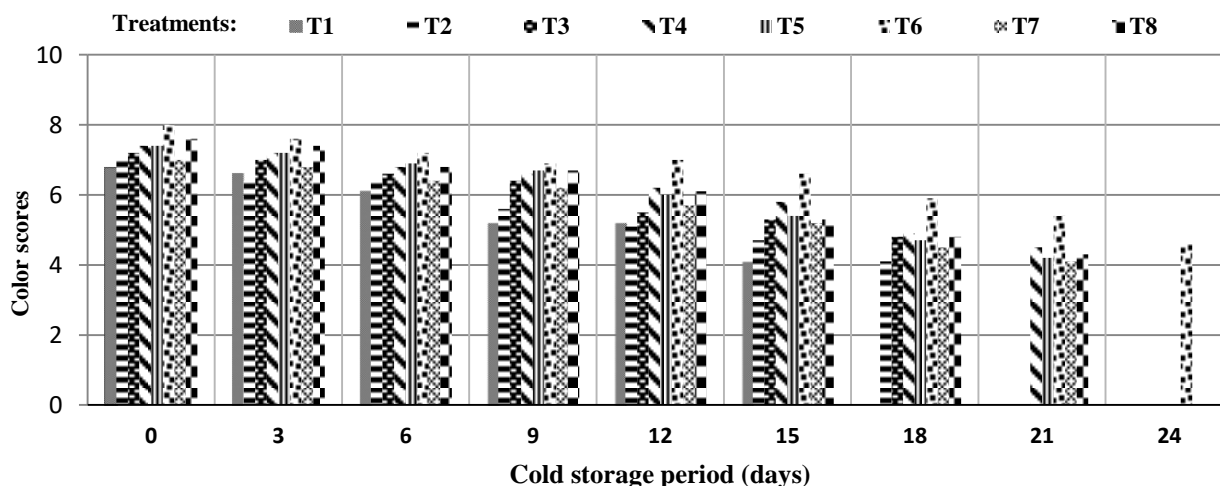
9	±0.46 4.69 ^{Ba}	±0.48 4.59 ^{Ca}	±0.59 4.53 ^{BCa}	±0.59 4.45 ^{Ca}	±0.59 4.48 ^{CDa}	±0.49 4.34 ^{CDa}	±0.58 4.59 ^{CDa}	±0.30 4.48 ^{CDa}
12	±0.10 5.18 ^{Ba}	±0.12 5.07 ^{BCa}	±1.09 4.96 ^{ABCa}	±0.89 4.95 ^{BCa}	±0.48 4.88 ^{BCDa}	±1.03 4.71 ^{BCa}	±0.84 4.95 ^{BCDa}	±0.81 4.94 ^{BCDa}
15	±0.55 6.05 ^{Aa}	±1.15 5.83 ^{ABa}	±0.58 5.38 ^{ABa}	±0.37 5.28 ^{Ba}	±0.50 5.28 ^{ABCa}	±1.04 5.11 ^{ABCa}	±0.46 5.21 ^{ABCa}	±0.51 5.17 ^{ABCa}
18	±0.84 R*	±0.02 6.22 ^{Aa}	±0.58 5.70 ^{Aa}	±0.48 5.66 ^{ABa}	±0.87 5.61 ^{ABa}	±0.49 5.56 ^{ABa}	±0.38 5.62 ^{ABa}	±0.06 5.55 ^{ABa}
21	R*	±0.02 R*	±0.13 R*	±0.16 6.17 ^{Aa}	±0.58 6.03 ^{Aa}	±0.46 5.92 ^{Aab}	±0.59 6.09 ^{Aa}	±0.49 6.00 ^{Aa}
24	R*	R*	R*	±0.17 R*	±0.28 R*	±0.47 6.10 ^A	±0.66 R*	±0.67 R*

T1: Control, T2: BHT, T3: 0.5% Thyme extract, T4: 1% Thyme extract, T5: 0.5% Clove extract, T6: 1% Clove extract, T7: 0.5% Mint extract and T8: 1% Mint extract. Means of triplicate \pm Standard deviation on wet weight basis. Mean values in the same row (small letter) or column (capital letter) with the same letter are not significantly different (at $p < 0.05$). R* = rejected.

According to the maximum acceptable limits of $6 \log_{10}$ cfu/g for chilled fish as reported by (ESS, 2019), all herbal extracts were effective in preventing the spoilage of battered common carp fish fillets than control sample and BHT treatment as shown in Table (3). The highest inhibitory effect for psychrophilic bacteria, subsequently the longest shelf life was obtained from T6 containing 1% clove extract. Although the impact of herbal extracts to be reduced bacterial growth in battered fish fillets, this is due to their antimicrobial activity. The counts of TPBC of all treatments on the final days of their analysis are more related to their TVB-N results about their acceptance limits of them. This results are in accordance with those found by Badei *et al.* (2013); Abdel-Aziz *et al.* (2019); Raesi *et al.* (2020) and Morachis-Valdez *et al.* (2021).

Sensory evaluation: The sensory evaluation is an easy, quick and efficient method for getting an idea about the quality of the product and its overall acceptance (Krokida *et al.*, 2001). Joseph (2003) reported that the acceptability of fishery products is normally based on the consumer's perception of the overall appearance based on the color, odor, texture and taste of the product. The changes in sensory

properties of semi-fried battered common carp fillets slices with herbal extracts during storage at $4 \pm 1^\circ\text{C}$ are seen in Fig. (6, 7, 8, 9 and 10). The initial scores of color, odor, texture, taste and overall acceptability of fresh semi-fried battered common carp treatments were between (6.80 to 8.00), (6.80 to 8.40), (6.90 to 7.30), (6.60 to 8.60) and (6.40 to 7.70), respectively at zero time. However, the sensory scores were referred to good fresh semi-fried battered common carp slices in accordance with physicochemical and microbiological examination at the beginning of cold storage, other than the best initial scores ($p < 0.05$) were obtained from treatments containing herbal extracts. Afterthought, the sensory properties of all treatments showed fall down trends ($p < 0.05$) due to the passing of cold storage, in relation to the deterioration advancement in all fish samples. The deterioration rate was closely associated with treatment type, the color scores of control sample were fallen down of acceptable score of (5) in the 12th day of storage, followed by BHT treatment in the 15th day. In the meantime, the T3, T4, T5, T6, T7 and T8 with herbal extracts were retained with color scores upper acceptable limit of (5) as shown in Fig. (6).

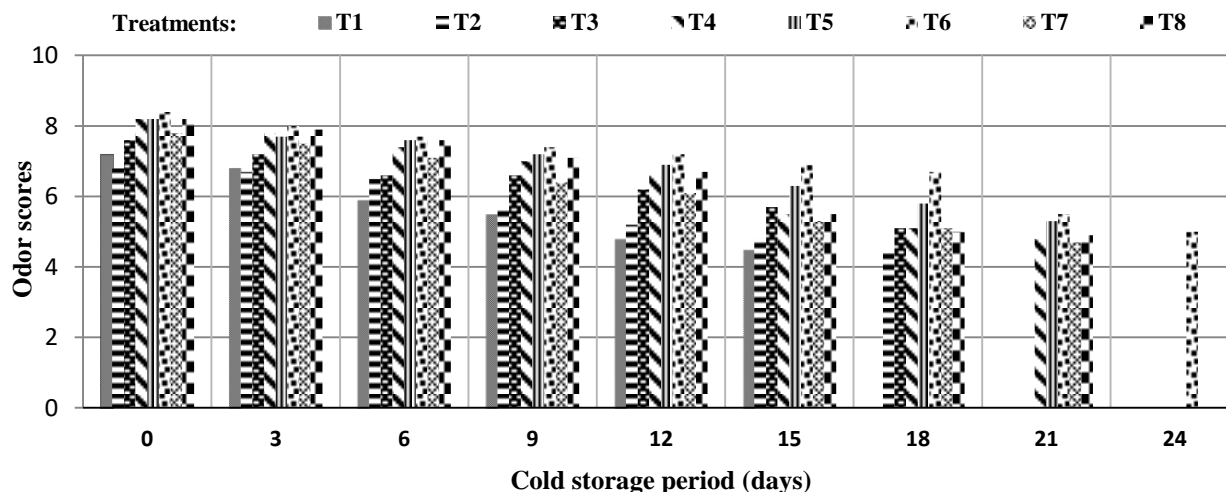


T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (6): Color scores of semi-fried battered common carp slices during storage period at 4±1°C.

With respect to the fishy odor development in fish products could be used as a sensorial index for fish spoilage, whereas the strong fishy odor was a proper sign for identification of fish spoilage during storage (Moawad *et al.*, 2017). The changes and developments of various odors in fish products during storage are closely associated with the type of

volatile compounds that formed due to spoilage (Lindsay *et al.*, 1986 and Lindsay, 1990). The greatest decline rate and the lowest scores ($p < 0.05$) was observed in T1 and T2, on contrary the herbal extracts especially obtained from clove was effectively delayed ($p < 0.05$) the deterioration of odor during cold storage as shown in Fig. (7).

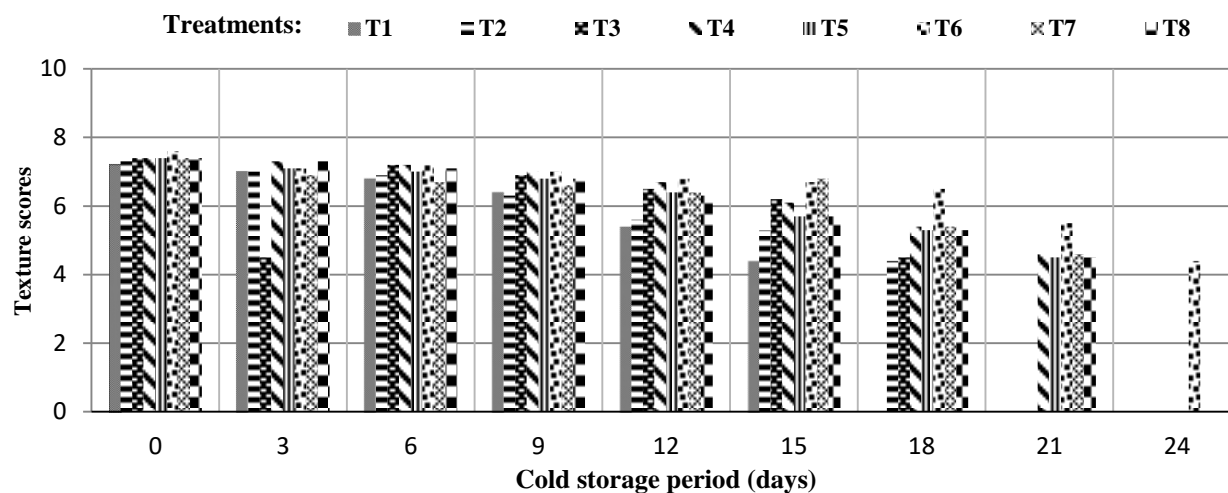


T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (7): Odor scores of semi-fried battered common carp slices during storage period at 4±1°C.

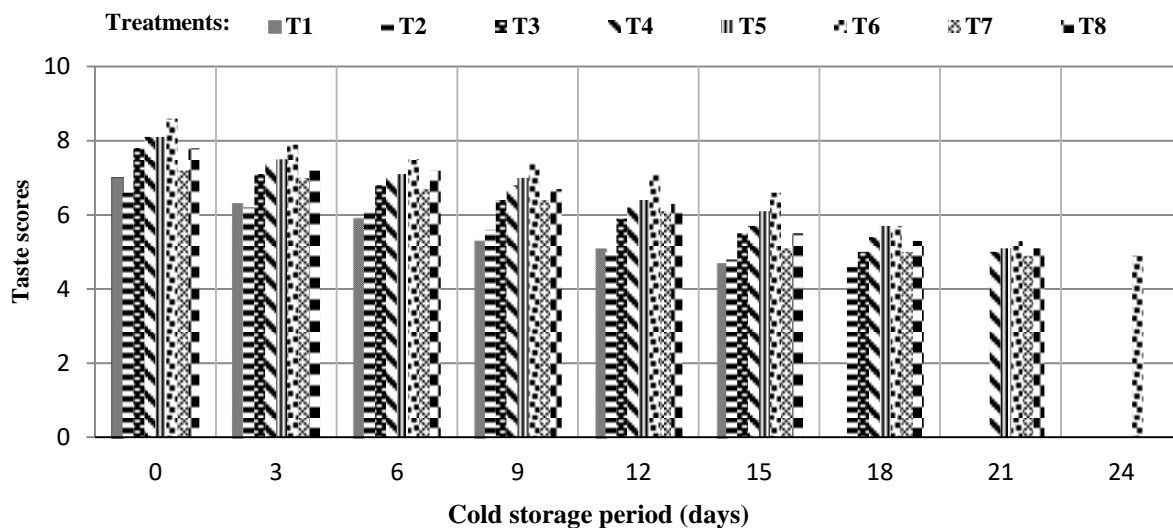
Respecting the changes in fish texture during cold storage, the texture is an important attribute contributing to the acceptability of the consumer because correct texture allows freshness and high quality (Chang *et al.*, 1993), and is directly related to the fat and water-holding capacities of the meat matrix, which in turn are influenced by the ionic strength and functional properties of the proteins (Jeyakumari *et al.*, 2016). The decline of texture scores was influenced by treatment type, to be the fastest in control sample other than treated samples. This might be correlated with the effect of herbal extracts to retarded the degradation of myofibril protein that is greatly responsible for the texture of the product. Whereas, the soft or mushy texture of fish limits the shelf life, thereby impede its

marketing. During postmortem handling and storage, the holding temperature, oxygen, endogenous or microbial proteases and moisture can result in detrimental changes in the color, odor, texture, and flavor of fish (Sriket *et al.*, 2010 and Sriket, 2014). So, the herbal extracts of thyme, clove and mint significantly prevented the softness due to protein degradation as microbial and enzymes activity, the highest level of herbal extracts (1%) was more effective than the lowest level (0.5%) to retain on the texture of battered fish fillets during cold storage. It worth mentioning that, the clove extract was exhibited efficacy more than thyme and mint extracts in retarding softening during storage as seen in Fig. (8).



T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig. (8): Texture scores of semi-fried battered common carp slices during storage period at $4\pm 1^\circ\text{C}$.



T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig.(9): Taste scores of semi-fried battered common carp slices during storage period at $4\pm 1^\circ\text{C}$.

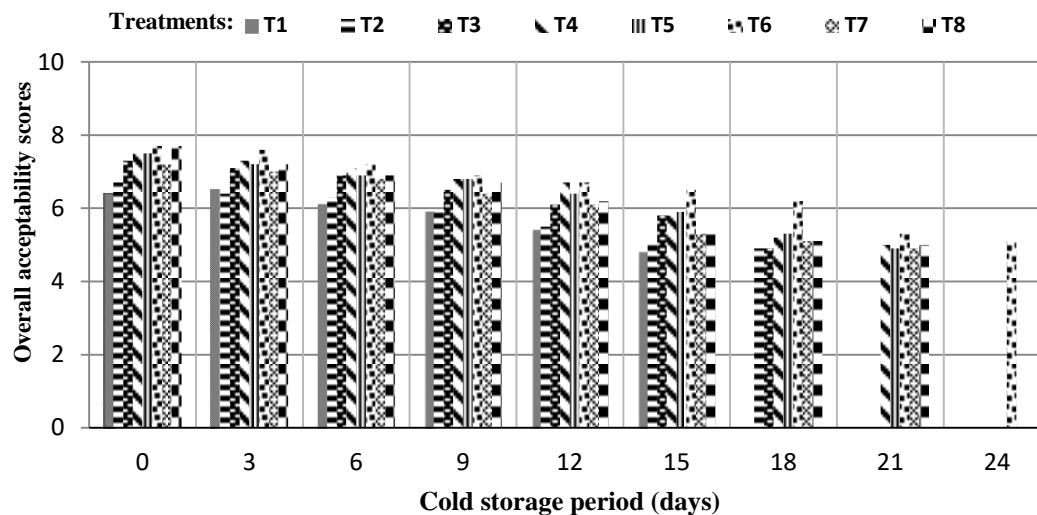
Concerning the changes of fish taste in semi-fried battered common carp fillets during cold storage are related to the chemical compounds of the food product by tasting in the mouth. Flavor is a complex concept that integrates the aroma (volatile substances), and tastes e.g. salty, sweet, metallic, sour, and bitter in the mouth (Meilgaard *et al.*, 1999). The property of taste is a part of fish flavor besides the odor, and it is affected by new compounds formation due to the deterioration of fish during storage. For example, the new products formed due to lipid oxidation/protein degradation in fish may develop unpleasant odor and taste (Toldrá and Flores, 1999 and Kilinc *et al.*, 2009). A significant difference ($p < 0.05$) was found between treatments, the T1 and T2 presented the lowest scores of taste onset of cold storage, compared to treated samples with herbal extracts. The lowest taste scores were found in control and BHT than the other

treatments. In contrast with samples containing herbal extracts had the highest scores with differences between themselves. The best scores of taste were obtained from treated samples with 1% herbal extracts, while the greatest effect was for clove extract as seen in Fig. (9).

Collectively, the overall acceptability of semi fried battered fish was affected by the changes in color, odor, taste and texture properties, to be the highest in samples treated with herbal extracts were retained by their acceptability for periods longer than control and BHT treatments, this evidence contributed to the shelf life of every treatment. The acceptable score of (< 3) was proposed to the limit of just acceptable, on this basis all battered fish fillets were accepted at their ends of storage, nevertheless, the herbal extracts particularly clove extract improved the sensory properties of battered fish fillets as presented in Figure (10).The superiority of color, odor, texture, taste and overall acceptability of

battered common carp slices was significantly obtained from herbal extracts treatments, especially those containing 1% of clove extracts, which were retained by scores upper than (5) till 21 days of cold storage compared to the other treated samples with

BHT, thyme and mint extracts. These results were corresponded with found by **Badee *et al.* (2013)**; **Hassan *et al.* (2013)**; **Elsayed *et al.* (2016)**; **Esmaeli *et al.* (2019)** and **Ehsani *et al.* (2020)**.



T1: Control, T2: BHT, T3: 0.5%Thyme extract, T4: 1%Thyme extract, T5: 0.5%Clove extract, T6: 1%Clove extract, T7: 0.5%Mint extract and T8: 1%Mint extract.

Fig.(10): Changes in overall acceptability scores semi-fried battered common carp slices during storage period at 4±1°C.

Conclusion: The obtained physicochemical, microbiological and sensorial results of semi-fried common carp fillets were obviously appeared the expected impact of incorporation batter mixture with herbal extracts. Whereas, the herbal extracts of clove, thyme and mint especially the highest concentration (1%) exhibited antioxidant and antimicrobial activity, to be reduced the values of pH, TVB-N, TMA-N, PV, TBA, TVBC and TPBC during cold storage, compared to control sample and BHT treatment. So, the fish spoilage as a result of lipid oxidation and microbial activity could be retarded by application the herbal extracts in semi-fried battered fish fillets under refrigeration, furthermore improved and developed sensory properties of fresh battered common carp fillets and during storage. Other than, the difference between tested extracts as well as applied concentrations in their preservative effect might be characterized by their antioxidant and antimicrobial activity, which closely contributed with the qualitative and quantitative effective compounds in these extracts. Finally, the defects of common carp fish flesh, further than the rapid spoilage of fish could be safely treated by battering fish fillets with suitable dough enriched by herbal extracts to give synergetic effect to delaying spoilage and improving acceptability during cold storage.

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الثبات التخزيني لشرائح أسماك المبروك العادي المغطاة والمعاملة ببعض مستخلصات الأعشاب

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إستهدفت هذه الدراسة تقييم تأثير إضافة بعض مستخلصات الأعشاب على الثبات التخزيني لشرائح أسماك المبروك العادي المغطاة خلال فترة التخزين بالتبريد على $4 \pm 1^\circ\text{C}$. وأشارت نتائج الدراسة إلى أن كل المعاملات كانت ذات جودة عالية في بداية فترة التخزين؛ ولكن بتقدم فترة التخزين حدث زيادة تدريجية ($p < 0.05$) فى محتوى النيتروجين الكلي الأميني المتطاير وزيادة أكسدة الليبيدات كنتيجة لزيادة النمو الميكروبي، متماشياً مع التناقص التدريجي للتقييم الحسي والقابلية العامة للمنتج خلال فترة التخزين بالتبريد. كما أشارت النتائج إلى وجود فروق معنوية بين المعاملات في قيم (الأس الهيدروجيني، القواعد النيتروجينية الكلية الطيارة، ثلاثي ميثيل الأمين، رقم البيروكسيد، رقم حامض الثيوباربيتوريك، العد الكلى للبكتيريا الحية والبكتيريا المحبة للبرودة)؛ علاوة على وجود فروق معنوية بين الخصائص الحسية (اللون، الرائحة، الطعم، القوام والقابلية العامة). كما أشارت النتائج إلى أن أعلى معدل للفساد ($p < 0.05$) لوحظ في كلاً من عينة الكنترول والشرائح المعاملة بمادة البيوتيل هيدروكسي تولوين و 0.5% من مستخلص الزعتر مقارنة بالمعاملات الأخرى. بينما أدى إستخدام مستخلص القرنفل (1%) إلى تأخير الفساد الكيميائي وخفض عدد البكتيريا والحفاظ على القابلية العامة بدرجة أعلى من المعاملات الأخرى. وبناءً على ذلك فإن إستخدام مستخلصات الأعشاب لكلاً من القرنفل، الزعتر والنعناع أدت إلى زيادة فترة الصلاحية لشرائح أسماك المبروك العادي المغطاة علاوة على تحسين الخصائص الحسية خلال فترة التخزين بالتبريد ويرجع ذلك إلى تأثيرها المضاد لكلاً من الأكسدة والنشاط الميكروبي. ولذلك فإن إضافة مستخلصات الأعشاب إلى مخلوط التغطية أدى إلى زيادة فترة الصلاحية وتحسين الخواص الحسية لشرائح أسماك المبروك المغطاة.

الكلمات المفتاحية: أسماك المبروك العادي - مستخلصات الأعشاب - البيوتيل هيدروكسي تولوين - التغطية - الجودة الفيزوكيميائية - الميكروبيولوجية و الحسية.