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EVALUATION OF ANTIMICROBIAL AND ANTIOXIDANT POTENTIAL OF TEA AND CAROB EXTRACTS IN BUFFALO BUTTER

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ABSTRACT

The present study was carried out to evaluate the antimicrobial and antioxidant effects of black tea extract (BTE), green tea extract (GTE) and carob extract (CRE) in buffalo butter during storage at 4±1ºC for 12 weeks and compare to synthetic phenolic compound butylated hydroxytoluene, (BHT). The antibacterial effect and minimal inhibitory concentration (MIC) of extract against *Listeria monocytogenes* were studied. The effectiveness of these extracts (0.025 – 0.05%) in butter made from pasteurized cream were evaluated by incubating (10^5 cfu/g) of *Listeria monocytogenes.* The GTE had completely inhibited (P≤0.05) the viability growth of *Listeria monocytogenes* during storage at 4±1ºC (10 – 12 weeks) . During storage of butter made from unpasteurized cream, the population of lipolytic bacteria and mould&yeast markedly reduced (P≤0.05) by addition GTE (0.025 – 0.05%) compared to other treatments. Also, BHT (0.025%) and GTE had similar inhibitory effect on *Staph. aureus. E. coli* and total counts. The BTE and GTE had a most antioxidant effect on decreasing the acid value (AV), peroxide value (PV) and thiobarbituric acid value (TBA). While CRE had less effect. Organoleptically, butter with added BTE, GTE and CRE gained the highest acceptance until 8 weeks compared with BHT treatment and control sample. This research has demonstrated that the use of BTE and GTE as a natural antimicrobial and antioxidant is a promising means to provide additional safety and improve the quality of buffalo butter during storage.

Keywords: antimicrobial - antioxidant - green tea - black tea - carob extracts - buffalo butter

INTRODUCTION

Modern consumers require high-quality foods that are preservative free for expanding the shelf-life and remising safe. The microbial growth of food-born pathogens and lipid oxidation are a major problems in butter during storage (Farag et al., 1991. Hassan, 1996, Puravankara et al., 2000 and Abd El-Gawad, 2007). Concerning the commercial synthenic phenolic compounds in the food system such as butylated hydroxyanisol (BHA) and butylated hydroxyl toluene (BHT) and their negative health effect (Willimas et al., 1990). For that reasons, natural compounds as antimicrobial and antioxidants are widely used against by food-borne microorganisms or to retard the development of rancidity and extent the shelf-life of many foods (Elgallyar et al., 2001, Sandak et al., 2001, Ozcan and Ayar, 2003, Emara and Abdel-Kader, 2004, Negi and Jayapakasha, 2004, Theivendran et al., 2006, Almajano et al., 2007, Friedman, 2007 and Beverlya et al., 2008).

In Egypt many investigators could isolated *Listeria monocytogenes* from dairy products especially butter (El-Price, 1999 and Ibrahim, 2003). Many chemical antimicrobial that have inhibitory activity against *Listeria monocytogenes* in food have been investigated (Zaika and Kim, 1993 Wang and Johnson, 1997, Beverlya and Janes, 2006 and Arslan and Ozdemir, 2008). Therefore, efforts have focused on the evaluation of natural antimicrobial agents against *Listeria monocytogenes* and control growth food-borne and spoilage microorganisms in food dairy systems have been published (Payne et al., 1989, Farias et al., 2000, Eswaranandam et al., 2004, Al-Hindi and Abd El-Ghani, 2005, and Beverlya et al., 2008). In this concern, the antimicrobial properties of polyphenols tea extract (flavonoids) were decumend over the decade (Kim and Fung, 2004a, Kim et al., 2004b). Other potential benefits from these compounds include reducing cardiovascular disease, preventing cancer, averting dental caries and acting as antioxidant (Mckay and Blumberg, 2002, Rietvield and Viseman, 2003 and Kyle et al., 2007). These health benefits are attributed to the high concentration of polyphenolic (flavonoids) found in tea.

However, there have now been numerous reports on the role of polyphenols and flavonoids-rich extracts tea (black and green) on physico-chemical of milk such as heat stability of milk (O'Connell and Fox, 1999a,b), the rennet coagulation time (Gad et al., 2005), the development of cooked flavour (Colahan-Sederstrom and Peterson, 2005) and Control Maillard browning (Schamberger and Labuza, 2007).

Carob pods have also been shown content lost of polyphenols which makes utilized as functional foods or food ingredient for their antimicrobial and antioxidantitive activities (Heber, 2002, Kumazawa et al., 2002 and Yasin and Ibrahim, 2004).

Although tea and carob available in Egypt, yet no work had undertaken to study the antimicrobial and antioxidant effects of these extracts in face major problems of microbial and lipid oxidation of butter during storage. Therefore, the present investigation is aimed to evaluation the effectiveness of natural antimicrobial and antioxidants extracted from tea (black or green) and carob pods compared with synthetic phenolic component (BHT) to choose the best for prevention buffalo butter against food borne pathogens and lipid oxidation during storage.

MATERIALS AND METHODS

Fresh buffalo's milk was obtained from the herd of Faculty of Agric. Cairo Univ. Milk was separated into cream (61% fat) and skim milk. The resultant cream was divided into 2 equal batch. The first batch was pasteurized at 80ºC for 10 min. the other batch was unpasteurized. All cream was holded for 24h at 4ºC then churned to obtained unsalted butter. Fine black tea (Twings ™) and china green tea were purchased from local market (R. Twining and Company limited, London, England, product of people's repuplic of China) respectivel3y. Butylated hydroxyl toluene (BHT) was obtained from Sigma Chemical Company (St. Lowis, M.O.).

The black tea and green tea extracts (BTE and GTE) were prepared from a filtered infusion of black and green tea powder (45g) in boiling water for 10 min) and concentrated, freeze dried (Snijders Scientific& Tilburg, Holland) according to Rusak et al., 2008)

The carob extract (CRE), was prepared from carob pods powder according to the scheme suggested by Kumazawa et al., (2002).

All powdered extracts were kept in a hermetically closed glass vessel at 4±1ºC until used.

Pure strain of *Listeria monocytogenes* type I was provided by Hungarian National Collection of medical bacterial (OKL, Budapest, Hungary). The stock culture was maintained at 4ºC on tryptose soy agar (Difco) slants were transferred monthly. Intermediate culture was prepared by inoculating stock culture into tryptose soy agar and incubated at 35-37ºC for 24h.

The antimicrobial activity of BTE, GTE and CRE was tested against *Listeria monocytogenes* was tested by the agar well diffusion method as described by Lyon and Glatz (1993).

The plates of tryptose soy agar were incoluted with active culture of *Listeria monocytogenes* (approximately 10^5 cfu/ml). the BLE, GTE and CRE with different concentration (0.25, 0.5, 75 and 1%) were added into the wells (0.1 ml). Triplicate samples were then incubated at 35-37ºC for 24 hr. A control sample consisted of bacterial which contained no antimicrobial. The minimum inhibitory concentration (MIC) was reported as the lowest concentrations of BLE, GTE and CRE capable of inhibiting the complete of growth of *Listeria monocytogenes* tested observed after 48h incubation.

Butter made from pasteurized cream was malted at 40ºC in a water bath, divided into 8 equal portions and packed into sterilized glass jars (250 gr). The first jar was sarved as control, BHT (0.025%) was added to second jar. Freeze-dried black tea extract (BTC) was added to the 3rd and 4th jars at level of 0.025% and 0.05% (BTE_1, BTE_2) respectively. Green tea and carob extracts (GTE, CRE) were added to the next jars at the same manner as BTE. Each jar was in inculated with 1 ml active culture of *Listeria monocytogenes* (10^5 cfu/g). All culture was thoroughly mixed with above extracts at 40ºC for 10 min, stored at 4±1ºC for 12 weeks and tested for the ability of BTE, GTE and CRE in killing or inhibiting the growth of *Listeria monocytogenes* every 2 weeks.

Butter made from unpasteurized cream was subsequently treated under the same condition as for butter made from pasteurized cream and analysed for microbiological and chemical analysis every 2 weeks.

Butter samples were microbiologically analysed for *Listeria monocytogenes, Staphylococcus aureus,* total coliforms, total count, lipolytic bacteria and mould&yeast as mentioned in American Public Health Association, (APHA, 1992).

The total phenolic content of BTE, GTE and CRE was determined by the Folin-Cioealteu colorimetric method at 725 nm according to Singleton et al., (1999). Butter samples were evaluated for acid value (mg KOH/g) and peroxide value (meq/kg) as described method mentioned in IDF, (1969) and the Association of Official Analytical Chemists (2000) respectively.

Thiobarbituric acid (TBA) value was determined as O.D. at 532nm according to Keeney, (1971).

The organoleptic properties of butter was assessed by 10 staff members of the Dairy Science Technology Department, Fac. of Agric Cairo Univ.

Experiment was carried out in triplicate and each analysis in duplicate. The results were analysed statistically using Analysis System version 8.0 (SAS, 2000) software package. Analysis of variance was performed by ANOVA procedures. Significant differences between means from triplicate analysis at ($P<0.05$) were determined by Duncan's Multiple Range test.

RESULTS AND DISCUSSION

The total phenolic content (TPC) of freeze-dried BTE, GTE and CRE expressed as gallic acid equivalent is present in Table (1). It is clear that GTE had higher TPC than those of BTE and CRE. These results are in agreement with found by **Yen** and **Chem (1995)**, Manzocco et al., (1998) and Kumazawa et al., (2002). Green tea was a richer source of phenolic than black and white tea and antimicrobial activity and antioxidative capacity of tea extracts correlated with the total polyphenol content (Roeding-Penman and Gordon, 1997, Friedman et al., 2006, Friedman, 2007 and Rusak et al., 2008).

Table (1). Total phenolic content (TPC) of black, green tea and carob extracts.

Type of extract	Concentration (mg/g)
BTE	110.74
GTE	198.58
CRE	102.07

BTE: Black tea extract
GTE: Green tea extract
CRE: Carob extract

Antimicrobial inhibition zones from tea and carob extracts (BTE, GTE and CRE) against *Listeria monocytogenes* are show in Table (2). Zones of inhibition for *Listeria monocytogenes* exposed BTE, GTE and CRE between 4 and 21 mm. On other hand, it is evident that the antimicrobial activity of these extracts was associated with its' concentration phenolic compounds (Kim and Fung, 2004). The water tea extracts (BTE, GTE) are more effective at controlling the growth of *Listeria monocytogenes* compared to CRE. This attributed to differ widely in their content of flavonoids (polyphenolic catechins pulps the afflavins). This results reforces researches that find green and black tea extracts have an antimicrobial impact (Raccaro et al., 2004, Kim et al., 2007).

Table (2). Diameter inhibition zones (mm) of black, green tea and carob extracts against the tested strain of *Listeria monocytognes*.

Type of extract	Concentration %	Diameter of inhibition zones (mm)
BTE	0.25	8
	0.50	8
	0.70	11
	1.00	16
GTE	0.25	16
	0.50	18
	0.70	18
	1.00	21
CRE	0.25	4
	0.50	8
	0.70	10
	1.00	14

Fig. (1) showed that the antimicrobial activity of synthetic and natural phenolic compounds (BHT and BTE, GTE, CRE) were more significantly effective (P≤0.05) against inculated *Listeria monocytogenes* of buffalo butter during storage periods compared to control sample. This effective was correlationship between concentration of extracts. The order of sensitivity of *Listeria monocytogenes* to synthetic and neutral phenolic compounds were GTE_2>BHT>GTE_1 >BTE_2>BTE_1>CRE_2>CRE_1>control sample. The effectiveness of green tea extracts attributed to high total phenolic content compared to other extracts (Table 1).

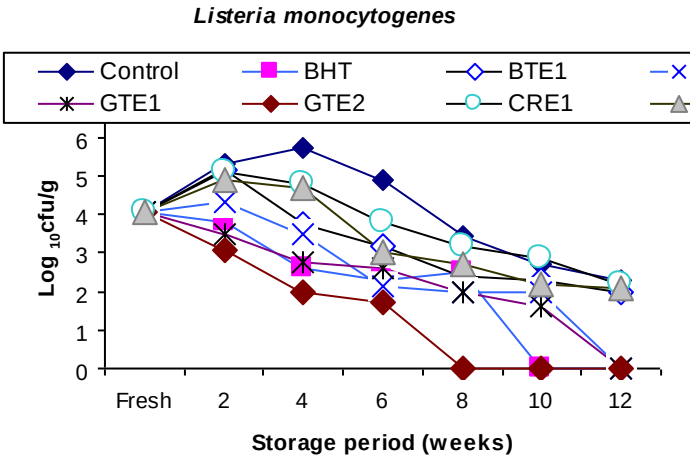

Fig (1). Inhibitory effect of natural antimicrobial (BTE, GTE and CRE against inoculated *Listria monocytogenes* on buffalo butter stored at 4±1ºC.

The antibacterial action of these phenolic compounds in mediated through their reaction with cell membrance inactivation of essential enzymes and destruction or functional inactivation of gentic materials (Prindle and Wright, 1977, Surak and Singh, 1980). However, Theivendran et al., (2006) indicated that the combination of nisin with grape seed extract and green tea extract might have contributed to the enhanced inhibitory effect of phenolic constituents in these extracts against *Listeria monocytogenes.* The mode of action of nisin involves pore formation in the cytoplamic membrance, which leads to rapid removal of free amino acid, ATP, and cations from the cell.

The effect of synthetic and natural phenolic compounds against *Staph. aureus*, *E. coli* and total counts in buffalo butter during storage is present in Fig (2,3). It could be cleared that tea extracts especially GTE and BHT exhibited the highest antimicrobial activity ($P \leq 0.05$) against *Staph.aureus, E. coli* and total count during the storage period compared to CRE. Negi and Jayaprakasha, (2001) reported the higher resistance of Gram-negative bacteria with some plant extract. The reason for the higher sensitivity of Gram-positive bacteria, relative to Gram-negative could be ascribed to the differences in their cell wall composition (Negi and Jayaprakasha, 2004). While, Kim and Fung, (2004) demonstrated that water soluble arrowroot tea extract has he potential to be used directly on food in order to prevent microbial growth of both Gram-negative and Gram-positive bacterial. However, El-Shawaf and Gomaa (2000) observed that chloroformic extract of orange peel had high inhibition activity on *Staph. aureus, Pesudomonas fluorescence, Listeria monocytogenes* and *Salmonella sp.* While inhibition activity was low on *Aspergillus flavus* and *Esherichia coli* and non-pathogenic microorganisms in buffalo's ghee and buffalo's butter.

The results from Fig. (4) show that the antimicrobial activity of natural extracts (tea and carob) ranged ($P \leq 0.05$) from inactivation to complete inhibition on lipolytic bacteria and mold&yeast during storage of buffalo butter compared with control sample. These results confirmed with data of acid value, peroxide value and thiobarbituric acid tested (Tables, 3, 4, 5). However, GTE (GTE_1&GTE_2) had advantages over other extracts because it successfully completely inhibited the growth of lipolytic bacteria and mould&yeast. Eissa et al., (2008) observed that the apple slices pretreated with cold carob extract have the highest inhibition mould&yeast after 4weeks storage at 4ºC. Generally, natural antimicrobial added to butter by different microorganisms especially when hygienic measures are inadequate.

Effect of BTE, GTE and CRE at different concentration on stability of buffalo butter are given in Tables (3,4,5). Periodic determination were made for acid value (AV), peroxide value PV and thiobarbituric acid (TBA).

The changes in the AV during storage of buffalo butter at 4±1ºC are presented in Table (3).The AV of all butter samples increased ($P \geq 0.05$) progressively with storage period. However, control sample markdly increased most during storage compared to extracts added butter sample. This increase may due to the present lipase raw milk and lipase release by microorganisms (Casrberg, 1992).While naturally polyphenols of BTE, GTE and CRE inhibited the hydrolytic rancidity. Ozcan and Ayar (2003) showed that the propolis extracts retarded hydrolytic rancidity in butter which related

to the presence of flavonoids in the propolis. Also, Yasin and Ibrahim(2004) observed that the lowest acid values were found in cake lipids containing 7.5% carob powder.

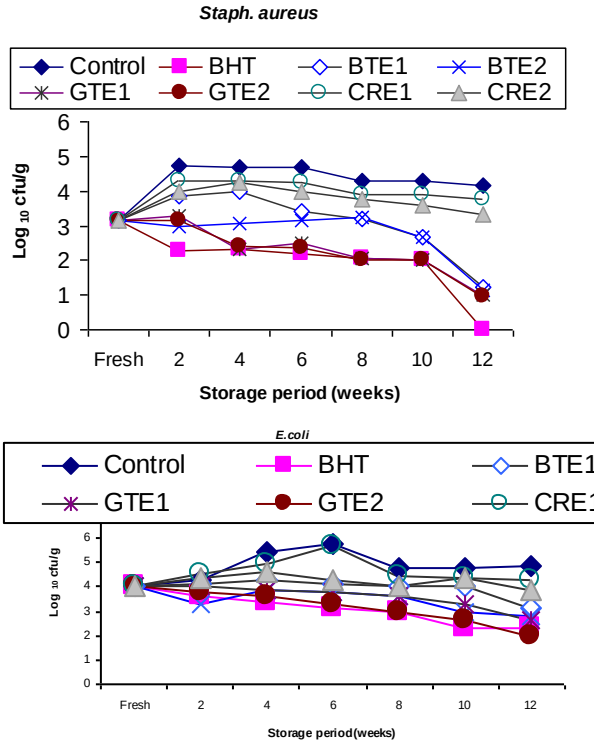

Fig (2). Effect of natural antimicrobial (BTE, GTE and CRE) on viability growth of pathogenic bacteria *Stap. aureus* and *E.coli* on buffalo butter stored at 4±1ºC.

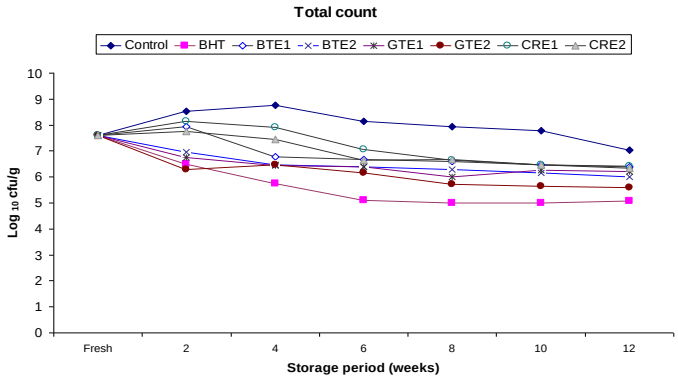

Fig (3). Log number of total count in buffalo butter made with natural antimicrobial (BTE, GTE and CRE) during storage at 4±1ºC.

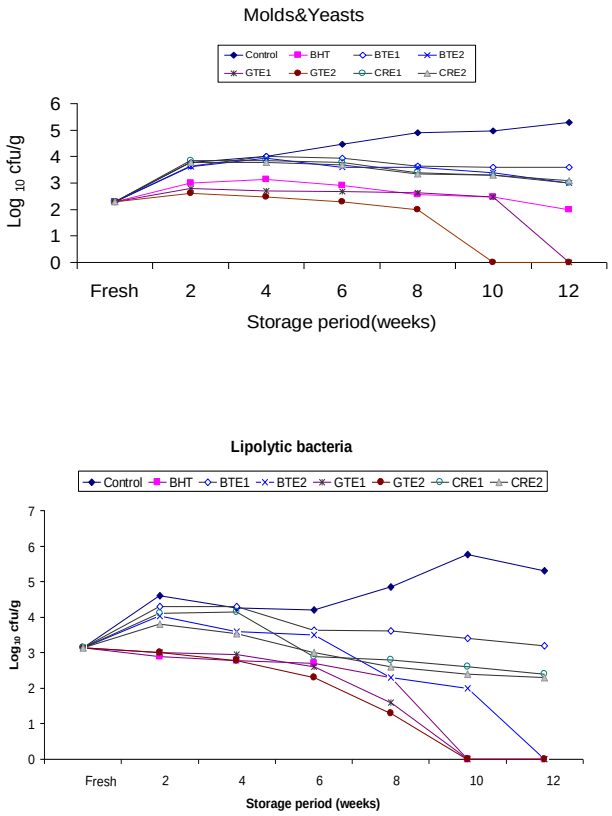

Fig (4). Log number of Lipolytic count and mould&yeast in buffalo butter made with natural antimicrobial (BTE, GTE and CRE) during storage at 4±1ºC.

Table (3). Effect of natural antioxidant on the acid values (meg KOH/g) of buffalo butter during storage at 4 ±1ºC

Storage Period (weeks)	Control	Treatments							Mean	S.E
		BHT	BTE		GTE		CRE			
		0.025%	0.025%	0.050%	0.025%	0.050%	0.025%	0.050%		
Fresh	0.620	0.620	0.620	0.620	0.620	0.620	0.620	0.620	0.620^{g}	0.0011
2	0.890	0.803	0.853	0.721	0.696	0.702	0.785	0.774	0.799^{f}	0.0185
4	1.035	0.957	0.997	0.946	0.897	0.856	1.029	0.918	0.963^{e}	0.0168
6	1.679	1.260	1.399	1.219	1.285	1.091	1.400	1.308	1.358^{d}	0.0436
8	1.765	1.418	1.780	1.395	1.639	1.267	1.726	1.495	1.567^{c}	0.0469
10	1.982	1.669	1.939	1.744	1.830	1.335	1.955	1.690	1.779^{b}	0.0508
12	2.131	1.802	2.080	1.984	1.934	1.531	2.122	2.000	1.952^{a}	0.0514
Mean	1.443^{a}	1.218^{b}	1.381^{a}	1.233^{b}	1.282^{b}	1.057^{c}	1.377^{a}	1.258^{b}		
S.E	0.1519	0.1174	0.1480	0.1329	0.1397	0.0897	0.1505	0.1346		

Different superscript (a,b,c,....) at the same raw and column are significantly different (P<0.5).

S.E:Standard Error.

From the obvious Table (3) , the AV in both level of GTE were low AV contents compared to control. The reduction rate of the mean AV was recorded 26.74 and 15.59% of buffalo butter treated with 0.05% GTE BHT respectivelly compared to control sample. The potent action of GTE is probably due to the presence of high amount of phenolic acids and other the related compounds (Majchrzak et al., 2007, and Rusak et al., 2008).

The peroxide value is a good index for the quality of a fat. It is clear from the data (Table 4) that the PV of all treatment increased (P≥0.05) with storage period. Also, the rate of increasing PV control butter was highest compared to other treatments (Amr, 1991). The rate of developing PV during storage was affected with the concentration of extracts added to butter which decreased with increasing the added level. El-Shawaf and Gomaa (2000) showed that the peroxide value of buffalo ghee during storage (60ºCfor 60 day) was affected with the concentration of citrus peel oil extracts depending on the number of active compounds.

Table (4). Effect of natural antioxidant on the peroxide values (meq/Kg)of buffalo butter during storage at 4±1º C.

Storage Period (weeks)	Control	Treatments							Mean	S.E
		BHT	BTE		GTE		CRE			
		0.025%	0.025%	0.050%	0.025%	0.050%	0.025%	0.050%		
Fresh	0.392	0.392	0.392	0.392	0.392	0.392	0.392	0.392	0.392^{f}	0.0011
2	0.876	0.846	0.475	0.426	0.523	0.509	0.598	0.516	0.649^{e}	0.0412
4	0.965	0.806	0.841	0.808	0.706	0.802	0.893	0.752	0.840^{d}	0.0244
6	1.064	0.980	1.098	0.959	0.754	0.944	1.130	1.015	0.997^{c}	0.0270
8	1.288	1.095	1.296	1.132	1.101	0.992	1.274	1.186	1.255^{b}	0.0312
10	1.472	1.139	1.314	1.142	1.163	1.009	1.314	1.289	1.239^{b}	0.0404
12	1.942	1.825	1.366	1.258	1.236	1.032	1.578	1.434	1.544^{a}	0.0739
Mean	1.142	1.012	0.961	0.884	0.839	0.811	1.025	0.941		
S.E	0.1307	0.1308	0.1010	0.0901	0.0859	0.0676	0.1091	0.1060		

Different superscript (a,b,c,....) at the same raw and column are significantly different (P<0.5).
S.E:Standard Error.

The decrease of PV for buffalo butter treated with natural antioxidants al level 0.05% was followed in the order of GTE >BTE>CRE>BHT. This attributed to the potent redical scavenging antioxidant capacity of catchins(especially flavonoids group) in green tea is due to a high number of OH group in their structure (Muzolf et al., 2008). Majchrzak et al., (2007) indicated that green teas provide approximately twice the total catechins quantity found in black teas. Naz et al., (2008) studied the effect of flavonoids on the stability of frying oil. All antioxiadants effectively reduced the oxidation rate in the oil, as detected by decrease in PVs. The order of antioxidative activity was gallic acid>quercetin>cyanidin>pelargonidin.The highest antioxidant activity of gallic acid is because of presence of an additional hydroxyl group. However, the phenolic isolated from mango seed kernel help to extent the stability ghee against autoxidation (Puravankara et al., 2000).

Sadak et al., (2001) reported that the addition of fenugreek seed extractas natural antioxidant to cotterseed oil delayed the peroxidant and

improvedits frying during heating. In addition to their antioxative properties, some flavonoids act as metal chelating agents and inhibit the superoxide-driven Fenton reaction, which is an important source of active oxygen radical (Giese, 1996).

The TBA test has been widely used as an objective measuring of secondary oxidation products of fat. It relates to the level of molonaldehyde formed oxidations of lipids and affected the butter quality which responsible for the development of rancid oder and off-flavour of butter (Allen and Hamiltm, 1983).

The changes in TBA values during storage of buffalo butter are presented in Table (5). The TBA values for all treatments increased gradually through the storage periods. The control samples had the highest TBA values among other treatments. However, results showed that reverse relationship between concentration of BTE, GTE and CRE used and the TBA values during storage. El-Shawaf and Gomaa (2000) observed that TBA was decreased with increase the concentrations of citrus peel oil extracts during storage period of ghee.

Table (5). TBA- values of buffalo butter treated with natural antioxidants (BTE, GTE and CRE) during storage at 4±1º C.

Storage Period (weeks)	Control	Treatments							Mean	S.E
		BHT	BTE		GTE		CRE			
		0.025%	0.025%	0.050%	0.025%	0.050%	0.025%	0.050%		
Fresh	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008[g]	0.000
2	0.009	0.009	0.017	0.017	0.015	0.011	0.018	0.14	0.013[f]	0.0008
4	0.68	0.54	0.039	0.025	0.031	0.025	0.032	0.028	0.043[c]	0.0039
6	0.79	0.073	0.057	0.046	0.047	0.043	0.064	0.054	0.061[d]	0.0033
8	0.106	0.093	0.074	0.068	0.064	0.058	0.074	0.060	0.079[c]	0.0042
10	0.125	0.108	0.094	0.075	0.097	0.075	0.106	0.089	0.099[b]	0.0039
12	0.141	0.128	0.124	0.110	0.100	0.094	0.132	0.127	0.123[a]	0.0037
Mean	0.078[a]	0.067[b]	0.059[c]	0.049[d]	0.050[d]	0.045[c]	0.062[c]	0.054[d]		0.0023
S.E	0.136	0.0121	0.0107	0.0095	0.0091	0.0084	0.0118	0.0110		

Different superscript (a,b,c,....) at the same raw and column are significantly different (P<0.5).
S.E:Standard

From the above results it could be indicated that the GTE was the most effective as a natural antioxidant compared to synthetic phenolic compound (BHT). Recent findings demonstrated that green tea exhibit stronger antioxidant activity than those of black tea. The inhibitory effect of antioxidants has been attributed to their donation of electrons of hydrogen atoms from phenolic hydroxyl group to fat or oil containing free radical which do not initiate nor propagate further oxidation of fat (Muzolf et al., 2008 and Rusak 2008). Kumazawa et al., (2002) showed the polyphenols of carob extract had weaker antioxidant activity in the DPPH free radical than those other polypenol compounds (quercetin and gallic acid).

Data presented in Table (6) show that buffalo butter treated with BTE, GTE and CRE (0.025 and 0.05%) had gained highly acceptant with no detectable defect up till 8 weeks compared to control and BHT samples. However, slightly acid taste appeared accompind with putried flavour after taste which was easy detected in butter after 8 weeks of storage.

Table (6). Organoleptic properties of buffalo butter made with natural antioxidant during storage at 4±1ºC.

Treatments	Concentration%	Storage period(weeks)	Acceptability	Appearance of decfect
Control	0.000	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	++	Slight acid
		10	+	Slight putrification
		12	+	Strong putrification, oxidized flavour
BHT	0.025	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	++	Slight acid
		10	+	Slight putrification
		12	+	Metalic flavour
GTE	0.025	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	+++	No detect
		10	+++	No detect
		12	++	Slight acid
	0.050	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	+++	No detect
		10	+++	No detect
		12	++	Slight acid
BTE	0.025	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	+++	No detect
		10	++	Slight acid+slight putrification
		12	+	Strong putrification
	0.050	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	+++	No detect
		10	++	Slight acid
		12	+	Slight putrification
CRT	0.025	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	+++	No detect
		10	++	Strong putrification
		12	+	Strong putrification
	0.050	2	+++	No detect
		4	+++	No detect
		6	+++	No detect
		8	+++	No detect
		10	+++	No detect
		12	++	Slight putrification

From the foregoing results it could be concluded that the natural extracts especially green tea extracts can be used as a new food preservatives because they have no healthy hazard effects (chemical preservatives) and are an effective means of improving safety and extending shelf-life of butter during storage without cause adverse effects on quality. Thefore, it be recommended for potential commercial application. Furthermore, studies are needed to identify the active antimicrobial and antioxidant compounds (phenolic) of above extracts to facilitate their use in dairy products.

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تقيم إمكانية استخدام مستخلصات الشاي والخروب كمضادات للميكروبات والأكسدة في صناعة الزبد الجاموسى

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يهدف هذا البحث إلى تقيم إمكانية استخدام المستخلصات المائية للشاي بنوعية (الأسود والأخضر) والخروب كمضادات للميكروبات والأكسدة للزبد الجاموسى مع مقارنتها بالمركب الفينولى الصناعي هيدوكسى بيوتالات التلوين BHT (٠,٠٢٥%)

في هذة الدراسة استخدمت طريقة الانتشار في أطباق الاجار لمعرفة اقل تأثير مثبط لمستخلصات الشاى بنوعية (الأسود – الأخضر) والخروب للميكروب المرضى *Listeia monocytogenes* وكان مستخلص الشاى الأخضر أكثر فعالية في معدل التثبيط لهذا الميكروب.

كما درس التأثير المثبط لنمو الميكروب المرضى *Listeia monocytogenes* لهذة المستخلصات (٠,٠٢٥ – ٠,٠٥%) في الزبد الجاموسى الملقح بهذا الميكروب (١٠ ° خلية/جرام) والمصنع من قشدة مبسترة كل أسبوعين من فترات التخزين (١٢ أسبوع) علي ٤ ±٥١م فكان مستخلص الشاي الأخضر أكثر فعالية في منع نمو هذا الميكروب بالمقارنة بالمعاملات الأخرى كما درست الخواص الميكروبيولوجية والكيمائية والحسية كل أسبوعين للزبد المخزن على ٤ ±٥١م لمدة ١٢ أسبوع والمصنع من قشدة غير مبسترة والمضاف الية المستخلصات السابقة بتركيز ٠,٠٢٥ – ٠,٠٥% وأوضحت النتائج إلى ما يلي:

١- استخدام مستخلص الشاي الأخضر بتركيز ٠,٠٥% كان لة تأثير معنوي واضح في تثبيط البكتريا المحللة للدهن والفطريات والخمائر.

٢- ظهر تأثير مشابة للمركب الصناعي BHT (٠,٠٢٥%) ومستخلص الشاي بتركيز ٠,٠٥% في تثبيط كلا من *Stap. Aureus* و *E. coli* والعدد البكتيري الكلى خلال فترات التخزين.

٣- انخفاض قيم رقم الحموضة (Acid value) ورقم البيروكسيد (PA) رقم حمض الثيوباربيوتريك (TBA) باستخدام هذة المستخلصات ويزداد معدل الانخفاض بزيادة تركيزها وكانت مستخلصات الشاي بنوعية (الأسود والأخضر) أكثر فعالية بالمقارنة بمستخلص الخروب.

٤- لم يحدث تغير في طعم الزبد المضاف لة هذة المستخلصات حتى ٨ أسابيع من التخزين.

ومما سبق توصى هذة الدراسة إلى استخدام مستخلصات الشاي والخروب وخاصة الشاي الأخضر كمواد حافظة طبيعية لاحتوائها على مركبات فينولية (الفلافونيدات) إلى الزبد بدلا من المركبات الفينولية الصناعية ذات التأثيرات الصحية السلبية بهدف تحسين قوة الحفظ وتجنب حالات التسمم الغذائي عن طريق الميكروبات المرضية وحمايتها من التزنخ الاوكسيدى أثناء التخزين.