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EFFICIENCIES OF VARIOUS FORMS OF HERBALS AND SPICES IN MEAT MARINATION ON PRODUCT QUALITY AND SOME HETEROCYCLIC AROMATIC AMINE (HCA) COMPOUNDS

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***Abstract*— Conservatively, meat has been marinated to improve flavor, tenderness, and increase meat shelf life. Beneficial effects of marination on meat texture include a juicier texture and decrease of cooking loss. Another aspect of marination is the increase of yield of the raw meat, which can provide benefits to the producer and the consumer. Besides that in recent years, it was revealed that some of the herbals and spices used in marinades have potential to inhibit the occurrence of heterocyclic aromatic amines (HCA) in meat. Cooking of meat generates these heat induced toxicants HCA's which are strongly mutagenic and carcinogenic. Marinades are water-based solutions that can include salt, organic acids, oil, herbs, spices, sugar, binders, antimicrobial agents and aroma enhancers. Natural additives were increasingly demanded for meat marination since concern over the safety of synthetic additives has arisen in recent years. Herbs and spices such as thyme, garlic, ginger extract have capability to prevent oxidation and microbiological spoilage of food, possibly better than many presently used synthetic antioxidants. Also preventing the occurrence of HCA's is the another important function of some of them determined in recent years. Various forms of herbals and spices such as essential oils, organic acids, powder and extract forms are used in meat marination and efficiencies of them were detected as different.**

Keywords— **Marination, Herbal, Spices, Meat Quality, Heterocyclic aromatic amine**

I. INTRODUCTION

Marinating is a process of incorporating a liquid solution, often called a marinade, to improve flavour, texture and juiciness to muscle foods. A marinade may contain any materials added to enhance the eating quality and appearance of the finished product. These includes; salt, organic acids, water, seasoning, spices, sugar, binders, antimicrobial agents and aroma enhancers [1]. Marinade solutions may also include natural or dried ingredients, herbs and other extracts. Spices and herbs, added in marinades significantly enhance meat safety and controlled or minimized lipid oxidation [2].

At present, meat industry uses chemical additives to prevent growth of food-borne pathogens and extend the shelf life of refrigerated storage of the products. Since concern over the safety of chemical additives has arisen in recent years, consumers increasingly demand use of natural products as alternative preservatives in foods [3].

The application of extracts from fruits, herbs and plants is of increasing interest given the consumer's demand in so called "natural additives": those naturally present in plants and to which numerous effects have been ascribed [4].

Marinating can reduce the formation of some carcinogen compounds such as heterocyclic aromatic amines (HCA) which occur during the cooking process. Addition of compounds, having antioxidant properties during meat marinating was claimed to be an effective method to reduce HCA in foods [5]. Marinades can act as a barrier to avoid the direct contact of flame with the meat and substantial reductions in the concentrations of MeIQ, PhIP, Di- MeIQx, IQ, IQx, and Norharman were reported in grilled chicken, grilled beef, and deep fried lamb meat [6]. HCA are formed on the surface of meat products through the Maillard reaction due to heat treatment, which involves creatinine, free amino acids and reducing sugars (glucose and fructose, directly or via hydrolysis of sucrose) as precursors [6]. The formation of HCA is a redox reaction, and antioxidants have the potential to reduce the formation of the HCA [7]. Herbs and spices, naturally rich in phenolic compounds, present high antioxidant activity toward free radicals and may provide easy-to-use tools for reduction of HCA dietary intake, when meats are pretreated or cooked with these ingredients [8].

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II. USAGE OF ESSENTIAL OILS IN MEAT MARINATION

The demand for natural alternatives to synthetic additives increases and the replacement, in foodstuffs, of synthetic antimicrobials such as sorbate and benzoate by essential oils (EOs) is getting considerable attention [9]. The modern trends in nutrition suggest the limitation of synthetic food additives or the substitution with natural ones [3].
Over the last years, various studies have focused on use of EOs in foods in order to either extend the shelf-life or improve the food safety of products [10]. Essential oils of both plants possess numerous biological activities such as antibacterial, antifungal, antiviral and antioxidative [11,12].

The active compounds in EOs with antimicrobial properties can be divided as: terpenes, terpenoids, phenylpropenes and others. There are indications that the microbial shelf life of certain meat and fish products can be increased by treatment of the foodstuff with certain EOs, and often EO from *Origanum vulgare* or *Thymus vulgaris* has been studied in that context because they contain the antimicrobial compounds thymol and carvacrol [9].

Particular interest has been focused on the potential application of plant essential oils as safer additives for meat. Several investigations have confirmed the antimicrobial action of essential oils against foodborne pathogens and spoilage bacteria in synthetic media, food systems and real foods. Higher concentrations of essential oils are needed to achieve the same effect in foods as in synthetic media. However, these higher concentrations needed to inhibit spoilage and pathogen bacteria in food matrixes could often exceed the flavor threshold acceptable to consumers [3].

Results of the studies on the use of essential oils in meat and meat products are given in Table 1.

Table 1 Application of essential oils in meat marination

Essential oils	Meat/Meat Product Tested	Results	Reference
• Essential oils (carvacrol and thymol, added at 0.4 and 0.8% v/w)	•Chicken	• Chicken was marinated at 4°C for 24 hours. • Carvacrol and thymol controlled the growth of spoilage microorganisms in marinated chicken meat.	[10]
• Basic red wine marinades (dry red wine, table salt, black pepper, garlic powder) • *Juniperus communis* essential oil (0.125%, 0.25%, 0.5%) • *Satureja montana* essential oil (0.0625%, 0.125%, 0.25%) • *Juniperus communis* (J) and *Satura montana* (S) essential oils combination (2:1) (J0.125 + S0.0625, J0.25 + S0.125 and J0.5 + S0.25)	•Beef	• Beef was marinated at 4°C for 24 hours. • Basic red wine marinades and the ones containing each EO or their combination remarkably decreased the counts of all monitored groups comparing to saline control. • The most pronounced effect was obtained with the marinade containing EOs mixture. • For all monitored bacteria, the bactericidal effect during marination was followed by bacteriostatic effect during subsequent meat storage.	[12]
Essential oils (EOs) from; • *Oreganum compactum* (oregano) (1%) • *Cinnamomum zeylanicum* (cinnamon) (1%) • *Thymus zygis* ct. Thymol (thyme) (1%)	• Pork filet • Pork bacon • Chicken filets • Chicken skin	• The growth of yeasts and molds was inhibited by immersion of all food matrices in 1 w/w % cinnamon EO. • Use of (1 w/w % for all EO) cinnamon EO led to microbial shelf life increase of pork filet and pork bacon. • Oregano EO increased the shelf life of pork and thyme EO of pork filet. • Sensorial properties of the meat were inevitably affected when the necessary EO concentrations to extend the microbial shelf life were applied.	[9]

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• Thyme and orange essential oils (EO) (1:1)	• Chicken breast and wing	• Tumbled with a lab-scale tumbler at 20 rpm for 20 min at room temperature. • Lower purge loss (breast and wing). • Higher *a** and *b** values (wing). • Lower shear force (wing). • Positive effect on breast and wing lipid oxidation.	[13]
• Oregano oil (*Origanum heracleoticum L.*) (1.25%, 0.625% and 0.156%)	• Chicken	• All samples were stored at 4°C. Sampling was carried out every 24h during 7 days. • Enhanced the shelf-life for two days of the samples with 0.625% oregano oil and three days of the samples with 1.25% oregano oil.	[3]

III. USAGE OF SPICE AND HERBAL ADDITIVES IN MEAT MARINATION

The interest in spices and aromatic herbs has recently increased because of their ability to prevent oxidation and microbiological spoilage of food, possibly better than many currently used synthetic antioxidants [13]. The antimicrobial properties of marinades are due to lowering of the pH, lowering of the water activity and addition of certain herbs and antimicrobial food additives [9].

It is known that fruit extracts and pulps can be used as natural antioxidants in meat products. Fruits are known to contain antioxidants such as fat-soluble vitamins, carotenoids, tocopherols, flavanoids, and water-soluble vitamin C. Bioactive ingredients and dietary fiber, which are highly found in fruit products, can also be used as an antimicrobial, coloring, flavoring and thickening agent [25].

Spices and herbs are excellent sources of antioxidants. Spices are used as ingredients, typically in relatively small amounts in recipes and formulations such as spice mixes and marinating sauces to enhance food flavor. It is expected that these marinating sauces should be excellent sources of antioxidants, since their main ingredients are derived from herbs and spices. However, these sauces undergo different processing methods during their production and are often used to marinate foods for different periods of time, and are exposed to various methods of cooking. All of these factors may potentially alter the antioxidant status of sauces significantly, and consequently the amount of antioxidants available to the consumer [15].

Several studies have shown that natural antioxidants in the form of spices, berries, wine or beer can reduce the formation of HCA in beef, chicken and pork. As previous research has demonstrated, pork chops, in particular, are often marinated before barbecuing, and there exists the potential to produce marinades using antioxidant herbs and berries, thus reducing the risk of formation of HCAs during barbecuing. Most of the studies investigating the effects of different natural antioxidants have used either a pure spice or berries or an extract of these in the experiments. Furthermore, numerous investigations have focused on the ability of the active compound to reduce HCAs and have overlooked the sensory quality of the marinated meat. However, it seems important to combine the chemical analysis of the effect of the compounds with a sensory and consumer study to evaluate the gastronomic value [7].

Results of the studies on the use of spice and herbal additives in meat and meat products are given in Table 2.

Table 2 Application of spice and herbal additives in meat marination

Spice and Herbal Additives	Meat/Meat Product Tested	Results	References
• Carbernet (CAB) • Tempranillo (TEM) • Isabel (ISA) • Control (300 mL dealcoholized wine/kg meat)	• Beef	• Beef was marinated for 48 hours at 4°C. • CAB and TEM were more effective against lipid oxidation. • The lower Warner Bratzler shear force values in beef steaks marinated with ISA. • The particular phenolic composition of ISA wine and its high content in organic acids, may explain its effects against *Enterobacteriaceae* while sugars may have promoted the growth of lactic-acid bacteria in beef marinated with CAB and TEM.	[4]

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• Marinade (16 g papaya leave extract or commercial bromelain, 2.5 g salt, 2.5 g minced garlic, 2.5 g ground black pepper, 2.5 g ground cumin and 2.5 g ground white cumin) • Control (2.5 g salt, 2.5 g minced garlic, 2.5 g ground black pepper, 2.5 g ground cumin and 2.5 g ground white cumin)	• Chicken	• Marinated chicken was left in the refrigerator for overnight. • *L** value, cooking loss and shrinkage of the marinated chicken were higher than control. • *a**, *b** values, protein content, water-holding capacity, shear force, and texture profile analysis of the marinated chicken were lower than control.	[14]
• Oregano • Acerola • Dijon marinade (acerola, sumac and oregano)	• Pork chops	• All of the marinades reduced the content of MeIQx and DiMeIQx, although only with indirect heat, while PhiP was reduced using both grilling methods. • In particular the content of Harman and to a lesser extent Norhaman was very high in the Dijon-marinated chops.	[7]
• Ginger extract (GE)	• Muscovy duck breast	• Duck breasts were marinated and stored at 5°C for 14 days. • TBARS values were lower in the GE marinated samples. • GE retarded lipid oxidation. • GE enhanced the proteolysis of Muscovy duck breast muscle.	[15]
• Hibiscus extract (*Hibiscus sabdariffa*) (0.2, 0.4, 0.6, 0.8 g/100 g)	• Fried beef patties	• The concentration of MeIQx was reduced by about 50% and 40% by applying marinades containing the highest amount of extract compared to sunflower oil and control marinade, respectively.	[16]
• Table sugar (10g), brown sugar (10.3g), and honey (14.7g) with the addition of tamarind, lemon, lime, and calamansi	• Grilled chicken	• Grilled chicken was marinated for 24 h at 4°C. • Using lemon in marinades containing table sugar, concentration of DiMeIQx was significantly reduced. • Calamansi was found to reduce HCA compounds in marinades containing table sugar and brown sugar, whereas tamarind in marinades containing honey.	[6]
• Rock candy (0%, 0.5%, 1%, 5%) • Soy sauce (0%, 2%, 4%, 10%) • Rice wine (0%, 1%, 2%, 4%)	• Beef • Pork • Mutton • Chicken	• Boiling meat (98 ± 2° C) with various ingredients, including soy sauce, sugar, rice wine. • Beef had the highest content of total HCA compared with pork, mutton and chicken. • Soy sauce contributed to the formation of HCA more greatly than rock candy, soy sauce, and rice wine.	[17]
• Turmeric (0, 1, 2, 3, and 4 g/100g meat) • Torch ginger (0, 2.5, 5, 7.5 and 10 g/100g meat) • Lemongrass (0, 2.5, 5, 7.5 and 10 g/100g meat) • Curry leaves(0, 2.5, 5, 7.5 and 10 g/100g meat)	• Grilled beef (satay)	• Turmeric reduced maximum 82 ng/100g level of IQ at 4 g/100g concentration at medium doneness and 44.4 ng/100g level of it was reduced of IQ, when satay was marinated with 10 g/100g lemongrass concentration at medium doneness. • The highest level of MeIQX was reduced to 83 ng/100g of satay meat marinated in 10g/100g torch ginger at medium doneness. • Curry leaves with 10 g/100g concentration marinated beef meat was reduced the level of IQ 78.5 at medium doneness.	[18]

• Basic Marinade for all of the samples: dry red wine (300 ml/kg), honey (40 g/ kg), garlic (9 g/kg), pepper (2 g/kg), salt (5%) • Marinade 1: Basic marinade • Marinade 2: Basic marinade, thyme (4 g/ kg) (T) • Marinade 3: Basic marinade, marjoram (4 g/ kg) (M) • Marinade 4: Basic marinade, horseradish (4 g/ kg) (H) • Marinade 5: Basic marinade, T(4 g/ kg), M(4 g/ kg), H(4 g/ kg)	• Beef	• Beef was marinated at 4°C for 48 hours. • Storage at 12 days at 5°C. • Marination (dry red wine, salt, lime-tree honey, thyme, marjoram, garlic, horseradish) evidently controlled total mesophilic aerobic bacteria, lactic acid bacteria and oxidation of beef meat.	[19]
• Nanoparticle paprika oleoresin (1 and 3 g/100 mL) •	• Cooked chicken	• Marinating was carried out by tumbling the fillets under atmospheric pressure using a commercial tumbler (Inject Star, USA) with test marinades for 20 min at 10 rpm. • 3 g/100 mL paprika produced greater surface *a** and *b** values, sensory surface orange, red and colour penetration attributes. • The application of nanoparticle paprika successfully enhanced marinating performance.	[1]
• Beer and white wine (with/without alcohol) alone or mixed with herbs commonly used as meat flavoring: garlic (2.9g) , ginger (2.8g), thyme (0.25g), rosemary (0.4g), and red chili pepper (0.1g)	Beef	• Beef was marinated for 4 h at 5°C. • Before the addition of meat to the marination, wine with herbs possessed the highest scavenging activity (73.5%), followed by wine (72.5%), dealcoholized wine with herbs (53.4%), beer and herbs (41.7%), dealcoholized wine (39.6%), and beer (25.9%). • After 4h of meat marinating, a decrease in the radical-scavenging activity of all marinades was observed, although with a similar radical-scavenging profile. • All of the six marinades under the study reduced the total amount of HCA, keeping meat with good overall sensory quality. • Beer marinades were more efficient than white wine marinades, and the addition of herbs provided a superior inhibitory effect, reducing around 90% of HCA. • Herbs explained around 30% of inhibition of PhIP formation, whereas alcohol increased PhIP formation.	[8]

• Herb decoctions (the aqueous extract of rosemary, sage and thyme) (Each herb in amounts of 35kg with 30L water was used in the distillation process)	• Turkey	• The turkey meat cuts were marinated overnight in plastic boxes. • Stored for 7 days in a dark room at 4°C • Meat from the rosemary marinade had the lowest TBARS values and volatile levels, while the control samples showed the highest values throughout the marinating, cooking and storage period. • Oxidative changes in meat marinated with thyme and sage were significantly more advanced than in meat marinated with rosemary decoction. • Antioxidants contained in herb decoction, could be exploited in marinades to prevent rancidity in stored, heat-treated turkey meat products.	[22]
• Lemon (5 mL) • Greentea(5mL) • Turmeric(5mL)	• Chicken	• Chicken was marinated at 4°C for various times (0, 1, 6, 12, 24, 36 hours). • A combination of green tea, lemon and turmeric was found to be the most effective against *C. jejuni* and *S. enteritidis* (12 hours).	[23]
• Teriyaki sauce with carvacrol (CV) or thymol (TM) (0.3 and 0.5%)	Beef	• Marinated beef was incubated at 4 °C for 7 days. • 0.5% CV or TM containing teriyaki sauce inactivated all inocula without recovery within 7 days. • The pathogens relocated from the beef into the leftover marinade (3.0–3.4 log CFU/mL) were also completely inactivated. • The treatment inhibited growth of indigenous aerobic bacteria and inactivated coliform bacteria.	[24]
• Table sugar • Brown sugar • Honey	Grilled chicken	• Chicken was marinated at 4°C for 24 hours. • All types of HCA (except IQx) in samples that were marinated with table sugar were significantly higher than brown sugar; whereas those were marinated with honey had the lowest HCA concentrations. • A substantial reduction in the concentration of MeIQ, PhIP, DiMeIQx, IQ, IQx, and Norharman was achieved in chicken marinated with honey. • A correlation study indicated that adding honey into the recipe retarded the formation of most HCA (MeIQ, DiMeIQx, IQ, IQx, Norharman, and Harman), whereas table sugars enhanced the formation of all HCA except Norharman, Harman, and AαC.	[25]

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IV. USAGE OF ORGANIC ACIDS IN MEAT MARINATION

Organic acids such as citric acid, lactic acid and acetic acid have high potency and are recommended because they do not have a health risk for consumers because they are already synthesized in natural environments. Organic acids dissociate when taken into the cell and reduce the intracellular pH and contribute to the production of more reliable and quality meat products. Depending on their use, the increase in water retention capacity is shaped, the solubility of meat proteins improves and sensory properties are improved. In addition to its contribution to meat quality, its antimicrobial effect against important pathogens also supports this widespread use [26].

pH significantly influences the Maillard reaction, so the simplest and low-cost method to stabilize the pH of the meat before the heat treatment could be marinating in a solution of an organic acid [27].

Citric acid, a food acidulante, is not only often used in acid marinating to improve the water-holding capacity and tenderness of beef muscle but is also commonly used as a chelator to control the activity of pro-oxidant metals. Lactic acid is often used in the meat industry as an antimicrobial agent [2].

Results of the studies on the use of organic acids in meat and meat products are given in Table 3.

Table 3 Application of organic acids in meat marination

Organic Acids	Meat/Meat Product Tested	Results	References
• Plum juice concentrate (10°Bx-14°Bx) • Apple juice concentrate (10°Bx-14°Bx)	Chicken	• Chicken was marinated for 36 hours at 4°C. • 14°Bx apple marination had lowest thaw loss. • 10°Bx apple marination had lowest cook loss. • 14°Bx apple marination had highest general choice value as a parameter of sensory analysis.	[14]
• Lactic acid (0.5%, 1%, 1.5%) • Citric acid (0.5%, 1%, 1.5%)	Beef	• Marinated for 72 hours at 4°C. • Cooking loss was lower in samples marinated with lactic acid compared to citric acid marinated samples.	[28]
• Citrus juice (31% orange juice, 31% lemon juice, 38% distilled water)	• Shin beef	• Tenderisation of beef samples using a citrus juice marinade was observed. It could be attributed to marinade uptake by muscle proteins and also to solubilisation of collagen.	[29]
• Lactic acid • Citric acid (0.5%, 1% and 1.5%) (1:4 w/v)	•Beef	• Beef was marinated for 15 hours at 4°C. • The highest redness (a^*) value was found in the lowest concentration of lactic acid. • The lowest a^* value was in the highest concentration of citric acid. • Lactic acid had a tenderizing effect particularly at the lowest concentration and had a positive effect on overall acceptability.	[30]

V. CONCLUSION

The functionality of the marinades varies entirely according to their compositions. Since spice and herbs have different health promoting properties their investigation as herbal additives is in great expansion. Usage of various forms of spices and herbs in meat marinates were searched and revealed that they increase shelf life, inhibit bacterial growth, diminish heterocyclic amin concentration, provide variety in product aroma and flavor, to establish a nice flavor balance. As a result of the studies, it was concluded that the use of different herbal additives and spices in combinations will promise. In addition to this, usage of different herbal additives with different ratios depend on meat sensory evaluation. Therefore, various combination studies are needed. In addition, the effect of used forms of herbal and spices should be taken into consideration.

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