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Natural antioxidants to reduce the lipid oxidation process of meat and meat products: A review

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Abstract

Meat is the muscle tissue of an animal which is rich in high quality protein, mineral, lipids with a small amount of carbohydrates. Due to its highly perishable nature, they are often very much susceptible to oxidation and microbial attack at any stages of processing, handling and storage. Oxidation leads to abnormal changes in meat like change in colour, texture, nutritional and organoleptic properties as well. Various synthetic antioxidants are available to retard the oxidation process in meat and meat products. Due to the carcinogenic nature of these synthetic antioxidants consumers are switching towards natural antioxidants obtained from plant sources. Natural antioxidants are those obtained from different parts of the plants like stem, leaf, bark, seeds etc. by method of extraction. Natural antioxidants can be applied by smearing or coating method in packages or it can be directly applied on meat products, help to overcome lipid and protein oxidation. Natural antioxidants also acts as preservatives in improving the storage life of meat products along with its functional or nutraceutical properties help to maintain the health of meat consumers. The natural antioxidants like grape seed extract, grape peel extract, spice extracts, rosemary extracts, clove, cinnamon extracts, tea extracts, grains, oilseeds, sage, cloudberry, beetroot, willow herb, rapeseed and pine bark, potato peel extracts are used to prevent lipid oxidation in meat and meat products. This review provides current information on natural antioxidants to reduce the lipid oxidation process of meat and meat products.

Keywords: triclosan, TCS, determination, detection, sensor

Introduction

Meat is the muscle tissue of an animal composed of protein, minerals, lipids and small quantity of carbohydrates. Due to the rich nutritional composition in meat and meat products makes it highly susceptible to quality deterioration (Devatkal *et al.*, 2012) ^[16], which often leads to chemical and microbial changes. The most common form of chemical deterioration is the oxidation of meat lipids, it is a complex process which depends on chemical composition of meat, light, oxygen access, storage temperatures and may also affected by some technical procedures followed during meat processing (Karakaya *et al.*, 2011) ^[31].

In general, oxidation is the loss of electrons or an increase in oxidation state by a molecule, atom or ion. Oxidation in lipid and protein fraction is one of the major causes of quality deterioration in meat and meat products. Oxidative deterioration in any type of meat manifest in form of discoloration, development of off-flavour and off-odour, production of potentially toxic compounds such as fatty acid peroxides, cholesterol hydroperoxide, and peroxy radicals and also nutrient and drip losses (Contini *et al.*, 2014) ^[13], consequently reducing meat products shelf life.

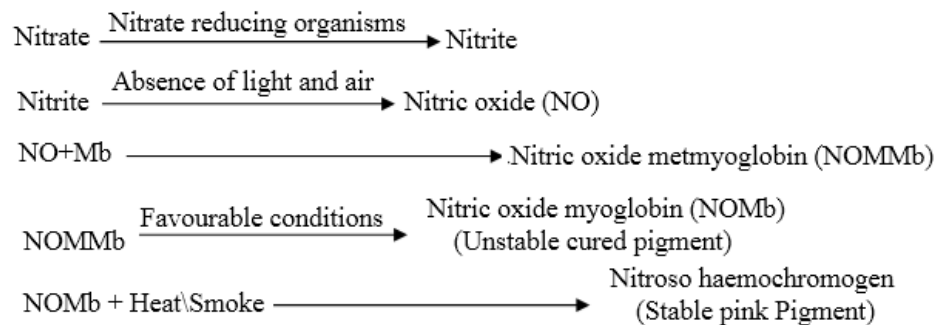
Antioxidant is any substance that present in low concentration to that of any substrate significantly reduces the oxidation of that substrate. There are two major groups of antioxidants i) Enzymatic antioxidants and ii) non enzymatic antioxidants, where enzymatic antioxidants are sub divided into primary and secondary enzymatic defences. The primary defence is composed of three important enzymes that prevent the formation of and neutralize free radical. The secondary enzymatic defence includes glutathione reductase and glucose-6-phosphate dehydrogenase. Glutathione reductase reduces glutathione (antioxidant) from its oxidized to its reduced form, and by this recycling, to continue neutralizing more free radicals (Ratnam *et al.*, 2006) ^[49]. Glucose-6-phosphate regenerates NADPH, which creates a reducing environment. These two enzymes support the primary enzymatic defence antioxidants and do not neutralize free radicals directly. The group of non-enzymatic antioxidants contains several subgroups, the main ones being: vitamins (A, E, C), enzyme cofactors (Q10),

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minerals (zinc and selenium), peptides (glutathione), phenolic acids, and nitrogen compounds (uric acid) (Carocho and Ferreira, 2013) [11].

Unsaturated lipids, fine grinding, incorporation of air, haem pigments, metal contact and high temperature during processing contribute to lipid oxidation. The typical colour of meat is due to the presence of myoglobin pigment. The hue (primary color), chroma (intensity) and the value (brightness) of meat colour are based on quantity and chemical state of myoglobin pigment. Catalase and cytochrome enzymes are also little responsible for meat colour. Myoglobin molecule



Cured flavour develops due to reaction between fatty acids and sodium nitrite resulting in the formation of benzonitrile and phenylacetone. However, nitrite has been found to be involved in the formation of nitrosamine which is supposed to be carcinogenic.

The use of antioxidants is one of the major strategies for preventing lipid and protein oxidation and may be effective in controlling and reducing the oxidation in meat and meat products. Due to the fact that synthetic antioxidant, like butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), *tert*-butylhydroquinone (TBHQ) and propyl gallate (PG) may constitute a potential health hazard for consumers (Biswas *et al.*, 2004) [5]. Nitrite is recognized as a potentially toxic compound of cured meat, including chemical toxicity, formation of carcinogens (reactions with some biogenic amines and formation of N-nitrosamines) in food or after ingestion, and reproductive and developmental toxicity (Juntachote *et al.*, 2006) [28]. Due to potential toxicological effects, interest in natural antioxidants and search on naturally occurring compounds that have antioxidant effect has been increased dramatically during the last decade. In this line, there are technological strategies that involve the application of antioxidants directly into the meat and meat products or by coating packaging materials with natural extracts to improve the oxidative stability of the products.

Natural antioxidants are mainly of plant origin which are obtained from various parts of the plant material like fruits (grapes, pomegranate, date, kinnow), vegetables (broccoli, potato, drumstick, pumpkin, curry, nettle), herbs and spices (tea, rosemary, oregano, cinnamon, sage, thyme, mint, ginger, clove) are investigated to decrease the lipid oxidation in meat and meat products and also showed preservative and antimicrobial properties in preventing the growth and activity of microorganisms (Akarpat *et al.*, 2008) [1]. The antioxidant properties is in turn affected by the method of extraction and the type of solvent used, since the extraction procedure strongly influences the composition of the extract (Brewer, 2011) [9].

Much of the work has been done on the utilization of antioxidants on meat and meat products. The main objective of this review is to emphasize the application of naturally

available antioxidants like clove oil, cinnamon, rosemary extracts and other plant extracts in reducing the oxidation of lipids in meat and meat products to prolong its quality and stability during storage prior to processing, packaging, transportation and storage.

Causes of lipid and protein oxidation

Lipids are chemically unstable and therefore prone to oxidation especially during post mortem handling and storage. Lipids are widely distributed in both the intra and extracellular spaces of meat as triglyceroids, phospholipids and sterols. Oxidation of lipids results in rancid odour, off flavor development, discolouration, loss of nutrients, decrease in storage shelf-life and accumulation of toxins which are hazardous to the human consumption point of view (Chaijan, 2008, Mapiye *et al.*, 2012) [12, 42]. Susceptibility of meat to post mortem oxidation is greatly influenced by the type of diet consumed by animals during the production phase. Exposure of meat to oxygen, light and temperature as well as preservatives and processing techniques such as chilling, freezing, additives (salt, nitrate and spices), cooking irradiation, high pressure and packaging could influence the extent of oxidation. In recent days, protein-lipid oxidation is one of the biggest economic problems in the meat industry. Zhang, Xiao, Lee and Ahn (2011) [67] reported an increase in lipid and protein oxidation in the breast muscles of birds that had been fed a dietary oxidized oil diet compared to antioxidant-supplemented and control diets.

Causes of lipid and protein oxidation in meat and meat products

Lipids are chemically unstable and therefore prone to oxidation especially during post mortem handling and storage. Lipids are widely distributed in both the intra and extracellular spaces of meat as triglyceroids, phospholipids and sterols. Oxidation of lipids results in rancid odour, off flavor development, discolouration, loss of nutrients, decrease in storage shelf-life and accumulation of toxins which are hazardous to the human consumption point of view (Chaijan, 2008, Mapiye *et al.*, 2012) [12, 42]. Susceptibility of meat to post mortem oxidation is greatly influenced by the type of diet consumed by animals during the production phase. Exposure of meat to oxygen, light and temperature as well as preservatives and processing techniques such as chilling, freezing, additives (salt, nitrate and spices), cooking irradiation, high pressure and packaging could influence the extent of oxidation. In recent days, protein-lipid oxidation is one of the biggest economic problems in the meat industry. Zhang, Xiao, Lee and Ahn (2011) [67] reported an increase in lipid and protein oxidation in the breast muscles of birds that had been fed a dietary oxidized oil diet compared to antioxidant-supplemented and control diets.

Assessment of oxidation in meat is usually done by measuring the amount of peroxide value thiobarbituric acid-reactive substances (TBARS), sulphhydryl and carbonyl group generated during the process. This analysis is carried out using spectrophotometric or chromatographic (head space gas chromatographic (GC), high-performance liquid chromatography (HPLC), liquid chromatographic mass spectrophotometer [LC-MS) and 2, 4 dinitrophenylhydrazine (DNPH)] methods. Recently, studies on protein-lipid oxidation have been conducted at a molecular level using mass spectrophotometry (MS) and liquid chromatography-tandem mass spectrophotometer (LC-MS/MS) with

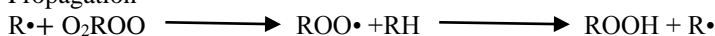
proteomic tools to better understand the mode of mechanism in relation to meat quality. Specifically, proteomic techniques have been used to identify unique oxidation sites on creatinase kinase, actin and triosephosphate isomerase in meat sample (Bernevic *et al.*, 2011) [4] and to investigate the relationship between post-mortem sarcoplasmic proteome and oxidation generation during storage and processing, as well as predictive markers that are sensitive to oxidative stress in meat sample (Promeyrata *et al.*, 2011) [47].

Oxidation of lipids is a three-step radical chain reaction which consists of initiation, propagation, and termination with the production of free radicals (Fig. 1a). Initiation reaction produces the fatty acid (alkyl) radical (R•) which in turn reacts with oxygen to form peroxy radicals (ROO•) in the propagation reaction. The peroxy radicals react with unsaturated fatty acids and form hydroperoxides

Initiation



Propagation



Termination



Fig 1a: Radical-chain of processes involved in lipid oxidation in biological systems. B: Radical chain of processes involved in protein oxidation in biological systems. (a) Chelating power of antioxidant.

Meat muscles are rich in protein content that contribute the overall quality, nutritional and sensory properties of meat products. Oxidation of protein in meat is considered as one of the innovative deterioration that indeed affects the overall physico-chemical and sensory attributes of meat. Protein oxidation occurs through a chain reaction of free radicals like oxidation of lipids in animal muscle (Lund *et al.*, 2011) [39]. Protein oxidation is described as the covalent modification of a protein induced ROS or by reacting with secondary by-products of oxidative stress (Shacter, 2000) [54], where oxidation begins with protein (PH) via ROS to form a protein carbon-centered radical (P•, reaction a) which is consequently converted into an alkylproxyl radical (POO•, reaction b) in the presence of oxygen and to an alkylperoxide (POOH, reaction c) by abstracting hydrogen atoms from another susceptible molecule.

Subsequent reactions with ROS, such as HO₂ or with reduced forms of transition metals (Mnⁿ⁺), such as Fe²⁺ or Cu⁺¹, lead to the production of alkoxy radical (PO•, reaction e and f) and its hydroxyl derivative (POH, reaction g). The oxidation of protein also occurs due to the interaction between proteins, especially the nitrogen or sulfur centers of reactive amino acid residues of protein (PH) and lipid hydroperoxide (ROOH) or secondary lipid oxidation products, such as aldehydes or reducing sugar (reaction h) (Baron, 2010; Viljanen, 2005) [3, 63]. Protein oxidation occurs through a chain reaction of free radicals like oxidation of lipids in animal muscle. The peroxy radicals (ROO•), formed during lipid oxidation, is absorbed by hydrogen atoms from protein molecules (PH) through chains of reactions summarised in Fig. 1b

In the presence of oxygen, the reaction of radicals (ROS) with muscle protein and peptides has been found to give rise to the modification of the amino acid side chain, formation of covalent intermolecular cross linked protein, protein fragmentation and aggregation (Lund *et al.*, 2011) [39]. Due to modification of amino acid side chain protein results in the formation of the thiol group, aromatic hydroxylation and

(ROOH), which later decompose to produce the volatile aromatic compounds that give meat its perceived off-flavours and rancid odour (Chaijan, 2008; Gordon, 2001) [12, 24]. The interaction of alkyl and peroxy radicals leads to the formation of non-radical products such as aldehydes, alkanes and conjugated dienes (Wsowicz *et al.*, 2004) [66]. Formation of aldehydes has been found to be directly related to the deterioration of meat colour and flavour, protein stability and functionality (Lynch *et al.*, 2001; Min & Ahn, 2005) [40]. The consequence of aldehydes has also been associated with atherosclerosis, putative mutagens and cancer formation in the body (Duthie *et al.*, 2013) [18]. The rate and extent of lipid oxidation is influenced by number of factors like iron content, distribution of unsaturated fatty acids, pH and antioxidant levels (Gattellier *et al.*, 2007) [22].

carboxyl groups (Stadtman, 1990) [59]. The formation of disulfide and dityrosine through the loss of cysteine and tyrosine residues due to cross-linkage of protein (Estevez *et al.*, 2009). According to the studies carried out by Estevez *et al.* (2009) the modification of the muscle protein is due to the denaturation and proteolysis induced change in meat quality, including texture traits, colour, aroma, flavour, water holding capacity and biological functionality. However, the strategic delivery of antioxidants from natural sources into muscle post-mortem holds a more viable option of enriching meat with health-promoting.

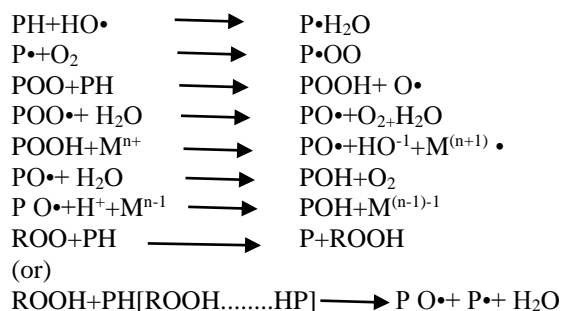


Fig 1b. Radical-chain of processes involved in lipid oxidation in biological systems (b) Reaction of antioxidants with lipid and protein at initiation stage

Source of natural antioxidants and its uses in meat and meat products

Plants are persistently the generous source to supply man with valuable bioactive substances (Tayel & El-Tras, 2012) [60] and thus different plant products are being evaluated as natural antioxidants to preserve and improve the overall quality of meat and meat products. Different parts of the plant material have been used for the extraction of antioxidants like leaves, roots, stems, fruits, seeds and bark. Some of these natural antioxidants are also available commercially and several studies have been carried out by different authors applying

commercially available natural antioxidants of plant origin to meat (Table 1).

Several studies were undertaken by researchers in preventing oxidation of meat and meat products using natural antioxidants. Mansour and Khalil (2000) [41] studied the natural antioxidative properties of potato peel, fenugreek seeds and ginger rhizome extracts on ground beef patties. Gallo *et al.* (2012) [21] studied the antioxidant properties of supercritical extracts from *Echinacea angustifolia* by mixing it with chicken meat to overcome the oxidation of lipid and protein. Kim *et al.* (2013) [33] studied the antimicrobial and anti oxidant activities of green leafy vegetable extracts and its application to meat products preservation. They observed that fatsia, a kind of green leafy have potential natural preservatives for meat products. Shah *et al.* (2009) [55] studied the effects of five spices (cinnamon bark, clove, pomegranate peel, oregano and grape seed extracts) on *Listeria monocytogens*, *Staphylococcus aureus* and *Salmonella enteric* on raw pork stored at room temperature. They found that clove was most effective in retarding lipid oxidation and

presented the highest antioxidant activities on raw pork. Siewe *et al.* (2015) [58] studied the potential application of *Sygium aromaticum* and *Cymbopogon citrates* essential oils a natural preservatives of beef patties and found that these essential oils protect beef patties against lipid oxidation and microbial growth. Rosemary (*Rosmarinus officinalis*) and hyssop (*Hyssopus officinalis*) extracts obtained from leaves and secondary branches, were used in pork meat (Fernandez-Lopez *et al.*, 2003) [20]. White peony (*Paeonia lactiflora*), red peony (*P. lactiflora*), moutan peony (*P. moutan*), sappan wood (*Caesalpinia sappan*), rehmania (*Rehmania glutinosa*), angelica, Korean (*Angelica gigas*), and rosemary (*Rosmarinus officinalis*) extracts were used in ground goat meat (Han & Rhee, 2005) [25]. Mint (*Mentha spicata L.*) leaf extract was evaluated by Kanatt *et al.* (2007) in lamb meat. Myrtle (*Myrtus communis myrtilus L.*), rosemary (*R. Officinalis L.*), nettle (*Urtica dioica*) and lemon balm (*Melissa officinalis L.*) leaf extract was investigated in beef patties (Akarpat *et al.*, 2008) [1].

Table 1: Natural antioxidants obtained from plant sources used in meat and meat products

Source	Scientific name	Parts used	Solvents used	references
Herbs used in natural antioxidant extraction				
Rosemary	<i>Rosmaricus officinalis</i>	Leaf, Secondary branches	Dimethyl sulfoxide, deionised water, acetone, exane	Fernandez-Lopez <i>et al.</i> (2003) [20]
Nettle	<i>Urtica dioica</i>	Leaf	Water	Akarpat <i>et al.</i> (2008) [1]
Mint	<i>Mentha spicata</i>	Leaf	Water	Kanatt <i>et al.</i> (2007)
Lemon balm	<i>Melissa officinalis</i>	Leaf	Water	Akarpat <i>et al.</i> (2008) [1]
Lemon grass	<i>Cymbopogon citrus</i>	Leaf	70% ethanol	Biswas <i>et al.</i> (2012)
Curry leaves	<i>Murraya koenigii</i>	Leaf	Water	Rahabah <i>et al.</i> (2011)
Green tea	<i>Camellia sinensis</i>	Leaf	Water	Rahabah <i>et al.</i> (2011)
spices used in natural antioxidant extraction				
Ginger	<i>Zingiber officinale</i>	Rhizome	90% ethanol	Mansour and Khalil (2000) [41]
Garlic	<i>Allium sativum</i>	Aerial part	Water	Tayel and El-Tras (2012) [60]
Hyssop	<i>Hyssopus officinalis</i>	Leaf and secondary branches	Dimethyl sulfoxide,	Fernandez-Lopez <i>et al.</i> (2003) [20]
Fenugreek	<i>Trigonella foenum-graecum</i>	Seed	Water	Akarpat <i>et al.</i> (2008) [1]
Cinnamon stick	<i>Cinnamomum burmannii</i>	Cortex	80% ethanol	Shan <i>et al.</i> (2009) [55]
Oregano	<i>Origanum vulgare</i>	Leaf	80% ethanol	Shan <i>et al.</i> (2009) [55]
Clove	<i>Eugenia caryophyllata</i>	Bud	80% ethanol	Shan <i>et al.</i> (2009) [55]
Onion	<i>Allium cepa L.</i>	Bulb	Water	Cao <i>et al.</i> (2013)
Fruits used in extraction of natural antioxidants				
Pomegranate Peel 80% ethanol	<i>Punica granatum</i>	Peel	70% ethanol, water,80% ethanol	Devatkal <i>et al.</i> (2010), Shan <i>et al.</i> (2009) [55], Tayel and El-Tras (2012) [60]
Grape	<i>Vitis vinifera</i> <i>Vitis vinefera</i> Var <i>Monastrell</i>	Seed pomace	80% ethanol methanol	Shan <i>et al.</i> (2009) [55] Garrido, Auqui,Marti, and Linares (2011) [23]
Kinnow	<i>Citrus reticulata</i>	Peel	Water	Devatkal <i>et al.</i> (2010),
vegetables used in extraction of natural antioxidants				
Fatsia or pepper plant	<i>Aralia elata Seem</i>	Leaf	70% ethanol	Kim, Cho, <i>et al.</i> (2013) [33], Kim, Min, <i>et al.</i> (2013) [34]
Roselle	<i>Hibiscus sabdariffa</i>	flower	Water	Karabacak and Bozkurt (2008)
Pumpkin	<i>Cucurbita moschata Duch.</i>	Leaf	70% ethanol	Kim, Cho, <i>et al.</i> (2013) [33], Kim, Min, <i>et al.</i> (2013) [34]
Chinese chives	<i>Allium tuberosum Rottler ex Spreng</i>	Leaf	70% ethanol	Kim, Cho, <i>et al.</i> (2013) [33], Kim, Min, <i>et al.</i> (2013) [34]
Bok choy	<i>Brassica campestris L. ssp. Chinensis</i>	Leaf	70% ethanol	Kim, Cho, <i>et al.</i> (2013) [33], Kim, Min, <i>et al.</i> (2013) [34]
Potato	<i>Solanum tuberosum</i>	Peel	90% ethanol	Mansour and Khalil (2000) [41]
Oilseeds used in extraction of natural antioxidants				
Sesame	<i>Perilla frutescens var. Japonica Hara</i>	Leaf	70% ethanol	Kim, Cho, <i>et al.</i> (2013) [34]
Black seeds	<i>Nigella sativa</i>	seeds	70% ethanol	Kim, Min, <i>et al.</i> (2013) [33]
Ground nut	<i>Arachis hypogaea</i>	Skin	80% ethanol	Tayel and El-Tras (2012) [60]

Spice extracts, fruit juice, tea extracts, and seed extracts and extracts from wine industry residues like grape seeds and peels are used as natural antioxidants (Brannan & Mah, 2007)

[8]. Wine industry residues are rich in phenolic compounds which are responsible for antioxidant activity. Grape peels

contain flavanols and grape seeds contain flavan-3-ol (Shirahigue *et al.* (2010) [57]).

Selani *et al.* (2011) [53] studied the antioxidant effect of wine industry residues extract (grape extracts) on lipid oxidation in raw and cooked chicken meat during frozen storage, and evaluated the parameters like pH, color, sensory properties of raw and cooked chicken meat stored at -18°C for nine months. The pH of raw and cooked samples was not affected by the addition of grape extracts and produced the satisfactory results, which did not differ from synthetic antioxidants. These findings suggest that the grape extracts are effective in retarding lipid oxidation of raw and cooked chicken meat during frozen storage.

Number of studies conducted to evaluate the potential use of grape residues as natural antioxidants in poultry meat (Brannan, 2009; Shirahigue *et al.*, 2010) [7, 57].

Plants, including herbs and spices contain phytochemicals which are potential sources of natural antioxidants, e.g. phenolic diterpenes, flavonoids, tannins and phenolic acids. These compounds have antioxidant, anti-inflammatory and anticancer activities. Spices and herbs (thyme, rosemary, sage, marjoram and black seeds) are used as natural antioxidants to prevent lipid oxidation in meat and meat products. Darwish *et al.* (2012) [15] studied the antioxidant activities of some spices and herbs on lipid oxidation in Frozen Chicken Burger.

Extraction of natural antioxidants from plant sources

Extraction method plays an important role in the recovery of overall yield quality of the essential oil obtained from the plant material. Various extraction technologies were followed for different parts of the plant materials like mechanical extraction, solvent method, supercritical fluid extraction methods, subcritical water extraction, maceration, soxhlet extraction and ultrasound assisted extraction to recover essential oils from plants. In general, plant materials are cleaned, dried and ground into a fine powder followed by solvent extraction using different solvents either separately or in combination have been used. The various methods of extraction along with the suitable solvents used are mentioned in Table. 1. The solvents like absolute ethanol, 90% ethanol, acetone, methanol, dimethyl sulfoxide, hexane and water are used (Shan *et al.*, 2014).

M. oleifera leaf extracts were prepared from dry leaves (Das *et al.*, 2012; Muthukumar *et al.*, 2012) [14, 45]. The dried leaves were powdered, sieved (No. 20) and extracted (100 g) successively with 600 mL of water in a Soxhlet extractor for 18–20 h. The extract was concentrated to dryness under reduced pressure and controlled temperature (40–50 °C).

Rababah *et al.* (2011) [48] prepared extracts from green tea leaves by mixing the powdered sample with water (1:10) and boiled for 10 min. After vacuum filtration, the filtrate was then frozen to -20 °C and freeze-dried (at b100 mTorr vacuum) to obtain the dry extract.

The dried herbs (white peony, red peony, mountain peony, sappan wood, rehmania, angelica (Korean), rosemary) were finely ground separately and then extracted thrice with 95%

ethanol (herb:ethanol, 1:4). Every time the ground herbs were stirred with ethanol on a hot plate at ~40 °C for 3 h and filtered. The combined filtrate was evaporated to dryness under vacuum (Han & Rhee, 2005) [25]. Rosemary and hyssop leaf extracts were prepared from dried powdered samples by using dimethyl sulfoxide (DMSO) (40 mg of dry weight/mL of DMSO) for 5 h at room temperature with occasional stirring, then left overnight. The extract was obtained from mixture by filtration (Fernandez-Lopez *et al.*, 2003) [20].

Khokhar and Magnusdottir (2002) [32] found water to be the best solvent for extracting tea catechins compared with 80% methanol and 70% ethanol. According to Yu, Ahmedna, and Goktepe (2005) [65], the total antioxidant activities of water and ethanol extracts of peanut skin were 3.39 and 4.10 mM Trolox Equivalent/mM of total phenolics. Turkmen *et al.* (2006) [61], reported that the antioxidant activity percentage (as measured by DPPH assay) for two different tea extracts was influenced by using different solvents such as water, acetone, N, N-dimethyl formamide, ethanol or methanol at various concentrations. Fifty percent acetone from black tea and 50% ethanol extract from mate tea had the highest antioxidant activity. Also, the results showed that solvent with different polarity had significant effect on polyphenol content and antioxidant activity.

Generally, organic solvents are very effective for extraction of antioxidants but they can affect human health if residues left in the final product which will not be acceptable for consumers. So, extra precautions are needed to remove all the traces of the extracting solvent. Water is the safest solvent, but less efficient in extracting all the antioxidants. Also, the processing methods affect the nature of the extract. Therefore, the use of safer alternative solvents and methods of extraction needs to be further examined for extraction processes. In addition, the cost-effectiveness of the extraction processes needs to be monitored to minimize the cost of natural antioxidants and ensure their wider utilization in the food industry (Vuong *et al.*, 2011) [64].

Prevention of oxidation and anti-microbial activities of natural antioxidants on meat preservation and security

Most of the natural antioxidants are obtained from plant sources such as culinary herbs, spice, vegetables as well as fruits and oilseed products. Phenolic compounds are the major contributor of the plant material that contributes to their antioxidant capacity. Phenolic compounds are thus regarded as effective sources of antioxidants to inhibit oxidation in muscle foods. Natural antioxidants can be applied either through dietary or technological strategies to reduce or prevent oxidation process in muscle food. Antioxidant compounds are usually added in moderate dosage level, since high level of oxidation may mechanically causes adverse effects through pro oxidative action (Martin and Appel, 2010) [43]. Technological strategies involve the application of antioxidants directly to the meat and meat products by coating on packaging material with plant extracts to improve the oxidative stability of the products. The dosage of antioxidant concentration, storage temperature and time to overcome lipid and protein oxidation in meat are mentioned in Table 2.

Table 2: Effect of dose concentration, storage temperature and time of natural antioxidant on lipid and protein oxidation in meat.

Natural sources	Dose in meat	Meat type	Storage temperature and time	Effect of oxidation	References
Oregano+sage leaves	2% w/v	Chicken breast and thigh	4°C, 98 h	SDL	Sampaio, Saldanha, Soares, and Torres (2012) [52]
Rosemary extracts	0.1%	Porcine liver patties	4°C, 90 days	SDP	Estevez <i>et al.</i> (2006)
Rosemary extracts	250, 500, 750 mg/kg	Porcine liver patties	-21 °C	SDL in a dose dependent manner	Doolaege <i>et al.</i> (2012) [17]
Sage extracts	0.1%	Porcine liver patties	4°C, 90 days	SDP	Estevez <i>et al.</i> (2006)
Herbal extracts (Marjoram, rosemary, sage)	0.04% v/w	Ground beef	5 °C, 41 and 48 days	SDL	Mohemeda <i>et al.</i> (2011)
Curry leaf extracts & mint leaf extracts	5ml extracts/ 500g	Pork meat	4°C, 0-12 days	SDL	Biswas <i>et al.</i> (2012)
Grape seed extracts	0.1%	Mutton slices	4 °C, 7 days	SDL	Reddy <i>et al.</i> (2013) [50]
Avocado seed extracts	50g/ 700g	Porcine patties	4 °C, 15 days	SDPL	Rodriguez <i>et al.</i> (2011) [51]
Broccoli powder extracts	1.5 & 2%	Goat meat nugget	4 °C, 4-16 days	SDL	Banerjee <i>et al.</i> (2012) [2]
Cocoa leaf extract	200mg/kg	Deboned chicken meat	4 °C, 21 days	SDL	Hassan and fan.(2005) [26]
Ginkgo biloba leaf extract	0.05% 500 ppm	Meat dumplings Meat ball	-18 °C, 180 days 4 °C, 21 days	SDL SDL	Kobus <i>et al.</i> (2010) [35] Kobus <i>et al.</i> (2014) [36]
Black current extracts	5,10 or 20g/kg	Pork patties	4 °C, 9 days	SDLP	Jia <i>et al.</i> (2012) [27]

SDL = significantly decrease lipid oxidation, SDP = significantly decrease protein oxidation, SDPL = significantly decrease lipid and protein oxidation

It was estimated that 50% of the meat spoilage and wastage occurs at the household consumption level due to poor preservative technique and facilities. Meat wastages are caused through microbial and chemical spoilage with the consequence of food borne illnesses, economic loss and food insecurity. It has been determined that different bacteria, like psychrophile, psychrotrophic, mesophile and thermophile, are able to survive under various processing conditions to cause spoilage and wastage. However, meat spoilage bacteria can be reduced by applying natural antioxidants directly into the meat products. The use of natural compounds such as organic acids and essential oils has been identified for decontamination of beef, pork and poultry products against *Salmonella*. Krishnan *et al.* (2014) [37] found a stronger antimicrobial effect of the combination of *S. aromaticum*, *Cinnomum cassia* and *O. vulgare* extracts in chicken meat than individual spices, and they attributed this to synergistic actions of each specific compounds present in the mixed spices. The presence and level of concentration of different phytochemical compounds such as phenolic, flavonoid, alkaloids, saponins, tannins, carvacrol, terpenes, and thymol among others, have been recognised as the potential source of antimicrobial activities in plant materials (Sharma *et al.*, 2012) [56]. Further study should be concentrated on the combination and application of different natural antioxidants to reduce meat spoilage and to extend the storage time, as these will greatly help to reduce financial loss, labour costs, ensure safety and ultimately improve the functional properties of the meat.

Future perspectives of edible medicinal plant antioxidants on meat

Natural anti oxidants obtained from plant sources were found to be most effective in combating the oxidation process that takes place in meat products and also its antimicrobial properties helps in overcoming the spoilage caused by many harmful microorganisms. These antioxidants obtained from plants are multifunctional in nature. The addition of

antioxidants inhibits the formation of cholesterol oxidised products, stabilizes the cholesterol levels and reduces the formation and absorption of malondialdehyde and heterocyclic amines (HCA), which have been reported as mutagenic by causing changes in DNA which may increases the risk of cancer in humans by consumption of cooked meat. By the application of this antioxidant rich extracts from medicinal plants helps to promote good health among humans and also acts as nutraceuticals or functional foods when it is applied directly over meat and meat products. In general functional foods are those which contains vitamins, minerals, dietary fibers, essential fatty acids and lignins which provides the required nutritional supplements to our body and nutraceuticals are those which contains non toxic food supplements that helps in the management, maintaining and treatment of diseases in the human body, which has medicinal stress and ROS related toxic damage. Regarding this, numerous medicinal plants, vegetables and spices have been identified to function in this capacity and their application in meat may be functional or nutraceutical. The value (Lobo *et al.*, 2010) [38]. It has been demonstrated that the consumption of meat rich in antioxidants can terminate the activity of the endogenous antioxidants against degenerative diseases linked to oxidative beneficial effect of producing meat products containing medicinal plant extracts would be to combat different health related problems related to the meat consumption over several years (Valenzuela *et al.*, 2003) [62]. The prevention of meat related disease in processed turkey and meat patties can be achieved by the incorporation of vegetable powder in the formulation (Duthie *et al.*, 2013) [18].

Conclusion

Meat and meat products are rich source of nutrients and contains high quality of protein, which is required in the human diet to carry out various functional activities. The processing of slaughtered meat products followed by its preservation and packaging prior to transportation from one place to another place is a challenging task, as these products

are rich in protein and fat are more susceptible to oxidation and microbial activities which gives rise to changes in various physical and chemical changes in meat products in relation to nutritional and organoleptic properties. Various synthetic antioxidants are available to overcome oxidation of meat products. Since, these synthetically available antioxidants are carcinogenic in nature and exhibits various health related problems, consumers are opting more for naturally available antioxidants obtained from various plant sources which are having functional and nutraceutical properties prior to consumption. The natural antioxidants not only prevent the lipid-protein oxidation but also show antimicrobial properties to retard the growth and activities of spoilage microorganisms in post slaughter carcass. They also help to improve and retain the original colour, taste, flavour other functional properties of packed meat and thus act as a preservative in extending the shelf-life of the packed meat and its products. Although, these extracts are safe on meat and meat products, further studies are needed to overcome the toxicological effects in meat and meat products as the extraction or processing conditions may alter their properties.

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