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Mini Review

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Food Processing By-Products as Natural Sources of Antioxidants: A Mini Review

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ABSTRACT

The growing interest in the replacement of synthetic food antioxidants has led to multiple investigations in the field of naturally-sourced antioxidants. The search for cost-efficient natural antioxidants has led to the exploration with raw materials of residual origin. The present review starts with an introduction of lipid oxidation and the antioxidant mechanisms, as well as the most recent research on the recovery and utilization of food processing wastes. Most studies found high levels of compounds with antioxidant activities in waste materials, encompassing a wide category of fruits and vegetables, roots and tubers, grains and seeds; a majority of these natural materials contains phenolic acids and flavonoids. Also, the levels of actives in the wastes are usually found to be higher than the actual products. Gaps in the area of by-products research as well as constraints of waste exploitation are also discussed.

KEYWORDS: Food waste; Food by-product; Natural antioxidants; Plant extracts; Antioxidant activity.

INTRODUCTION

The research and exploration of natural antioxidants has been rising in recent years. This increased attention is driven by several trends in the food industry. First of all, the reformulations of food products using healthier ingredients have imposed a negative impact on product shelf life. For example, whole grain ingredients are increasingly used instead of refined flour. Major food manufacturers have gone far beyond sliced breads to incorporate whole grains into a variety of shelf stable products. The germ in the whole grain contains high levels of naturally occurring fat composed of polyunsaturated fatty acids, which are susceptible to oxidation. Therefore, whole grain reformulation calls for additional protection from antioxidants to maintain high product quality. Omega-3 enrichment has been another continuing trend. Omega-3 fatty acids are being added to a wide range of product categories, from baked snacks to dairy products, sauce and dressing and beverages. The desire to enrich food with nutritional ingredients such as fish oils (omega-3s fatty acids) greatly increases oxidative challenges due to the oxidative instability of these ingredients. This challenge must be met by using antioxidant systems of different combinations of natural phenols, vitamins and organic acids. Moreover, increasing number of manufacturers are placing their product in clear packaging. This transparency attracts consumers, yet poses great challenges to food stability because light decomposes food. Addition of antioxidants helps to fight light-induced oxidation and protect the freshness of the product.

Synthetic antioxidants such as BHA (Butylated hydroxyanisole), BHT (Butylated hydroxytoluene), TBHQ (*tert*-Butylhydroquinone), Propyl gallate, Ethylenediaminetetraacetic acid (EDTA), etc. have been widely used in the food industry because they are more effective and less expensive than natural antioxidants. Yet, with the consumer's desire for fewer artificial ingredients, synthetic additives are being removed from many food and beverage formulations.. The clean label trend toward simpler, more recognizable ingredients is expected to direct food product development into the future.[1-3]

Several review studies looked at the potential of using agriculture by-products as antioxidant sources for food industry application.[4-6] The present review will focus on some of the mostly studied materials such as apple, exotic fruits, olive, and potato. The problem and limitation of by-product exploitation will also be addressed.

WHY WASTES AND BY-PRODUCTS?

According to the Food and Agriculture Organization (FAO), about one-third of food produced for human consumption is lost or wasted globally. FAO's statistics[7] (Figure 1) showed that even though losses on the consumer side differ between regions, all regions have major losses at production. Further analyses on food waste category showed that fruits and vegetables, along with roots and tubers represent the category that has the highest wastage rates, followed by cereals, oilseeds, dairy and meat. In 2012, 45% of fruits and vegetables, or roots and tubers are lost compared to 30% for cereal, 20% for oilseeds, and 20% for dairy.[8] Food waste is usually derived from the food production processes. At present, substantial quantities of waste/by-products are generated and are disposed at a cost to the manufacturer.

Recycling of the by-products has been supported by numerous findings that many polyphenols and other chemically active compounds are located specifically in the peels and other waste stream materials. This discovery is due in part to advancing analytical technology during the past decades that enables efficient detection of active compounds from these waste stream materials. As the search for effective and non-toxic natural compounds with antioxidant activity intensifies, it provides means for reusing the waste which is both highly beneficial and economically advantageous. Nonetheless, only a few by-product derived antioxidants have been developed successfully at a commercial scale from the vast quantities of plant residues produced by the food processing industry. The effectiveness of recovery and extraction, the marketability of resulting extracts and the practical suitability for the food are some of the most important limiting factors for the commercialization of waste agriculture products.

LIPID OXIDATION AND ANTIOXIDANT MECHANISM

Rancidity occurs when oils or oil-containing foods become oxidized. Autoxidation is the major mechanism of oil oxidation at ambient condition. Autoxidation leads to the generation of off-odors and off-flavors that render foods unacceptable. The word autoxidation denotes a free radical chain reaction that takes place in three stages: initiation, propagation and termination (Figure 2). In the initiation stage, heat, metal or light catalyze the formation of alkyl radicals in lipids. In the propagation stage, alkyl radicals react with oxygen to form peroxyl radicals, which can abstract hydrogen from new lipid molecules and form the primary oxidation product hydroperoxides. During this process, new radicals are formed that propagate the reaction chain. The hydroperoxides decompose readily to generate secondary oxidation products, many of which are responsible for the undesirable odors and flavors. Autoxidation process terminate when radicals react with each other to yield stable non-radical species. Oxidation of lipid and lipid containing foods reduces food quality, shortens shelf life, and compromise their nutrition value.

Antioxidants inhibit lipid oxidation under different mechanisms (Figure 2). Free radical scavenging is the most common mechanism; these antioxidants quench radicals in lipid or food by donating a hydrogen molecule. The resulting antioxidant molecules are more stable than the lipid radical, thus intercept further free radical reactions. Antioxidants may also slow down oxidation by chelating transition metals, which is a major catalyst for oil degradation. EDTA and citric acid are the most common chelators; flavonoids, phospholipids, polyphenols, and amino acids can also chelate metals. Light can also be a pro-oxidant. The mechanism of light-induced oxidation is very different from oxidation without the influence of light, and usually results in a rapid degradation of foods. Photosensitizers such as chlorophylls that are present in oil and food absorb visible or UV light and transfer the energy to oxygen to form singlet oxygen. Singlet oxygen can directly react with unsaturated lipids. Antioxidants such as carotenoids, tocopherols, phenolics, and amino

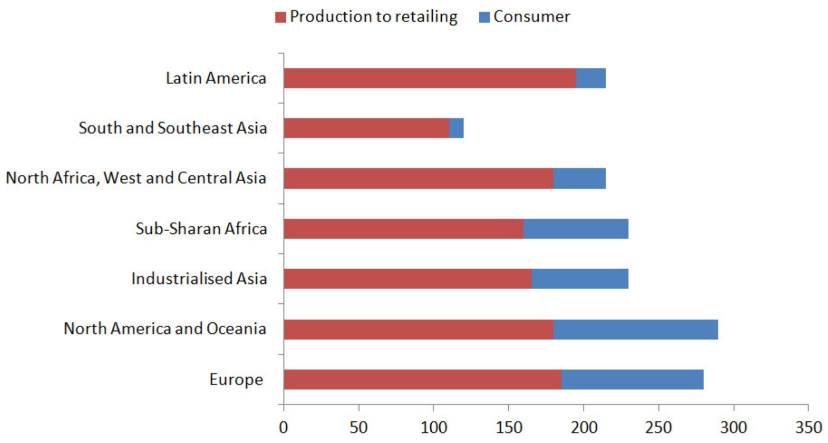


Figure 1: Food losses by region (sourced from The Food Agriculture and Organization of the United Nations (FAO)[7]).

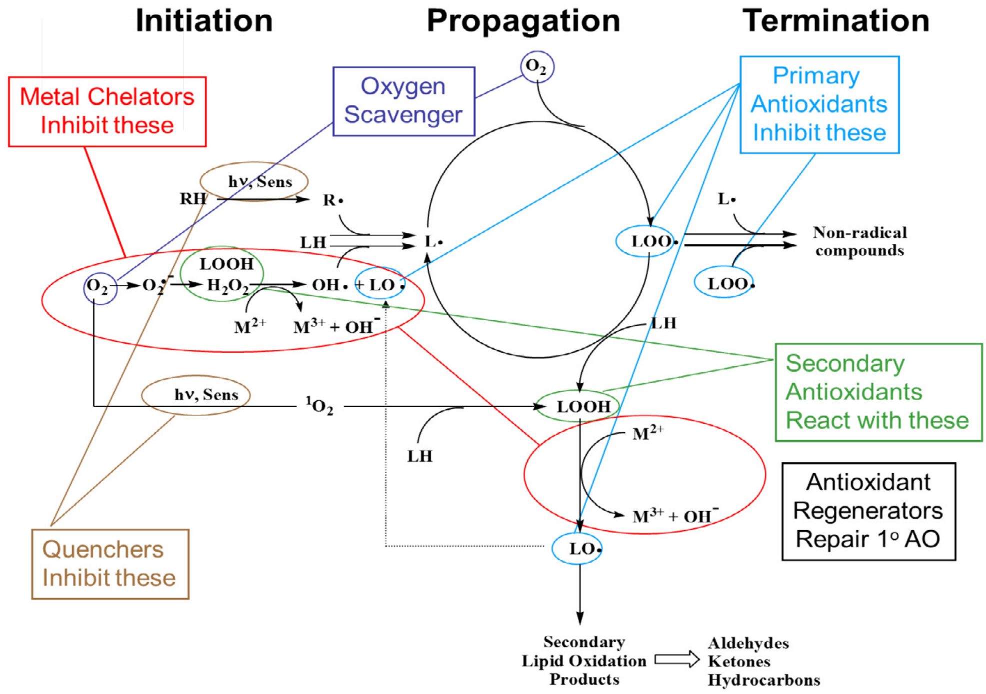


Figure 2: Autoxidation pathways and reaction points of antioxidants (adapted from Berdahl et al[9]).

acids can either quench singlet oxygen or inactivate photosensitizers, resulting in the slowing down of photoxidation reactions. Some antioxidants exhibit more than one mechanism of activity. In addition, usually more than one antioxidant is present in a complex food system. Each antioxidant may work with each other and provide synergistic effects.

EVALUATION OF ANTIOXIDANT ACTIVITY OF WASTE MATERIALS

Different methods were adopted to measure the antioxidant activity of natural materials. These include total phenolic contents, ABTS (2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid)), DPPH (2,2-diphenyl-1-picrylhydrazyl), ORAC (oxygen radical absorbance capacity), FRAP (ferric reducing antioxidant power), CUPRAC (cupric reducing antioxidant capacity), and β-carotene bleaching by LOO·. The accuracy and validity of using these assays to predict antioxidant activity of natural materials is still a disputed area[10-12]; the complexity of chemical composition of natural materials is one of the major causes for this disconnection. Moreover, many studies found conflict between the levels of phenolic compounds and antioxidant capabilities measured by ABTS, DPPH or FRAP,[13-16] indicating above assays have bias on some compounds over others. Lou et al[17] found a high correlation of total phenolic content and DPPH assays in their study of immature calamondin, while Morais et al[18] found a better correlation between FRAP assays and phenolics and flavonoids in various tropical fruits, but not as much for the DPPH assay. The authors explained that the Folin-Ciocalteu assay, which is used to measure phenolic compounds content, shared a similar mechanism as FRAP, with both evaluating the reduction potentials of antioxidant compounds, whereas DPPH evaluates radical quenching activity by transferring hydrogen to lipid radicals. Besides, Vieira et al[13] found the correlation between phenolic content and antioxidant activity measured by FRAP, ABTS and DPPH vary between peel and flesh,[13] suggesting these assays measures different compounds present in flesh and peels respectively. Nevertheless, these *in vitro* capability assays were commonly used by researchers around the world, with total phenolics, DPPH, ABTS being the most extensively used methods (Table 1).

In model or real food systems, researchers commonly use Peroxide Values (PV) and Rancimat to measure induction time or inherent stability of oil and foods, as well as Thiobarbituric acid (TBA) assay or Thiobarbituric acid reactive substances (TBARS) assay and volatile compounds to evaluate degrees of secondary oxidation.

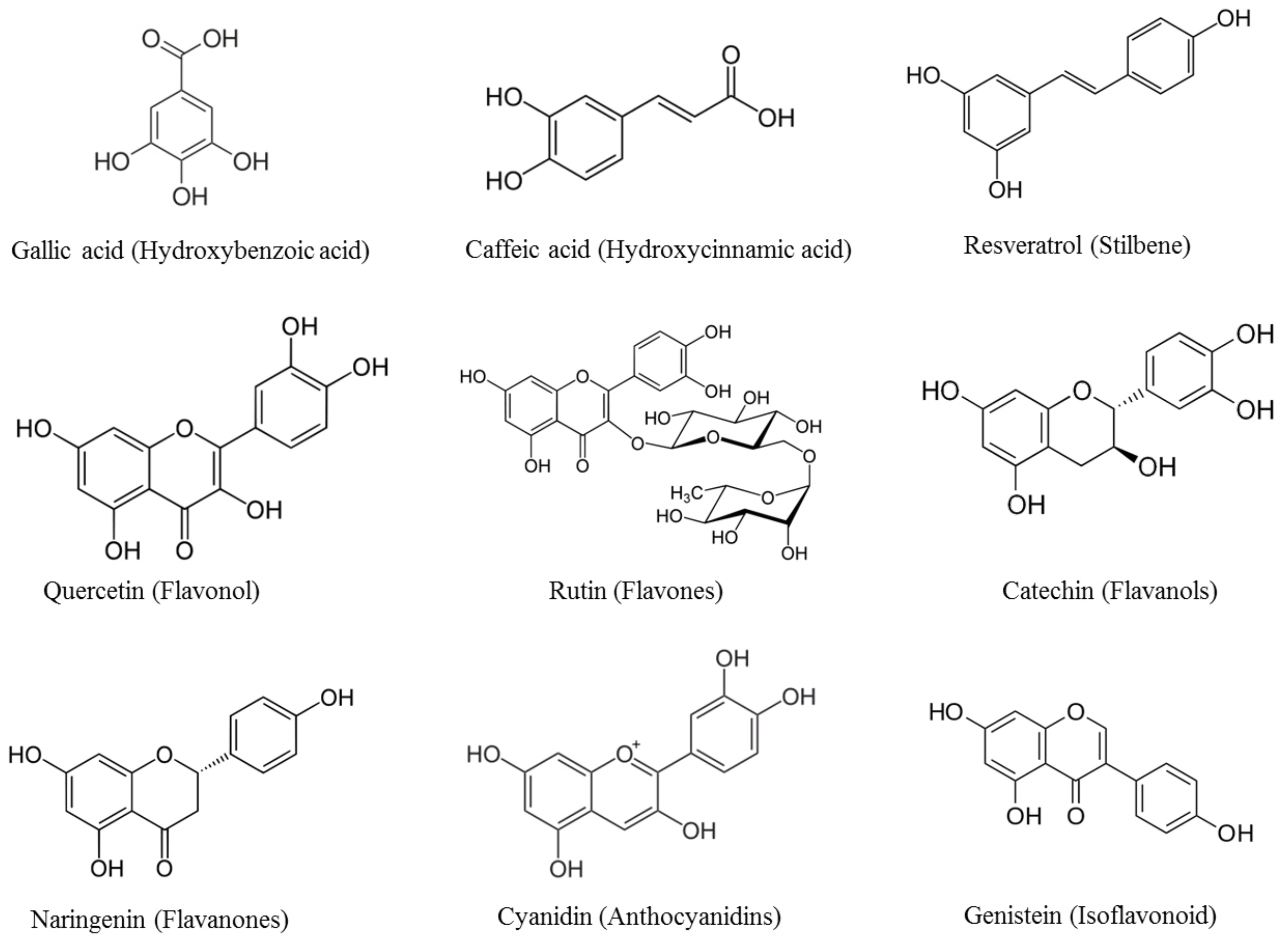
ANTIOXIDANTS FROM WASTE MATERIALS

Copious evidence shows that processing waste from agriculture materials usually contains higher amount of antioxidants compared to the flesh.[13,15,19-23] It was pointed out that the removal of peels may induce a significant loss of antioxidants.[13,22] Table 1 listed the result of an extensive literature review on the studies of food by-product. Sources, active compounds and evaluation methods were listed, divided by sector type of wastes. It is evident that phenolic compounds are the major group of antioxidants in waste products. Structures of some example compounds representative of major phenolic classes are shown in Figure 3.

Kabir et al[51] examined the water and ethanol extracts

Waste product sources	Antioxidant compounds	Evaluation methods	References
Immature calamondin	Phenolic acids, flavonoids	TPC, Flavonoids, DPPH, ORAC	Lou et al[24]
Pomegranate peels	Phenols	TPC, DPPH, stability of ghee	El-Shourbagy and El-Zahar[25]
Grape skin	Phenols		Pinelo et al[26]
Vitis Vinifera	Phenolics	total polyphenols, total flavonoids, *o*-diphenols	Casazza et al[27]
Citrus mandarin peels	Polyphenols	TPC and DPPH	Karsheva et al[28]
Passion fruit peel	Phenolics	FRAP, TPC	Nascimento et al[29]
Immature kumquat	Flavoinoids	TPC, DPPH, PRAC	Lou et al[24]
Peel from purple star apple	Phenolic acids and flavonoids	Soluble phenols and total flavonoids, DPPH, ABTS	Moo-Huchin et al[30]
By products from various tropical fruits	Phenolics	TPC, total flavonoids, FRAP, DPPH	Morais et al[18]
Wild chocke berry waste	Phenolics	TPC	Vauchel et al[31]
Apple peel, pomace, core	Phenolic acids and flavonoids	DPPH, FRAP, total phenolics	Wijngaard et al[32]
Kiwifruit peel	Phenolic acids and flavonoids	DPPH, FRAP, total phenolics	Wijngaard et al[32]
Grapefruit peel	Phenolic acids and flavonoids	DPPH, FRAP, total phenolics	Wijngaard et al[32]
Corn silk	Polyphenols and flavonoids	Total phenolic content, total flavonoid content, ABTS	Rahman and Rosli[33]
Carrot peel	β-carotene, phenols	TPC, β-carotene bleaching assay	Chantaro et al[34]
Broccoli byproducts	Glucosinolates, phenolic acids, and flavonoids	DPPH, Vitamin C	Domínguez-Perles et al[35]
Horse chestnut seeds	Quercetin and kaempferol glycosides	Total and individual flavonoids	Kapusta et al[36]
Chicory leaf residue	Phenolics	Induction time of corn, peanut, soybean oils	Lante et a[37]
Avocado seeds and peels	Catechins, epicatechins and their oligomers	ORAC and DPPH	Wang et al[38]
Olive oil waste	Hydrpxytyrosol, 3,4-dihydroxyphenylglycol, elenolic acid derivatives	ABTS, DPPH, TBARS	Rubio-Senent et al[39]
Olive oil mill wastewater	Phenols, L-Proline, tocopherols	Rancimat	Ranalli et al[40]
Olive mill residues	Phenolics	Total phenolics and individual phenolic compounds	Lesage-Meessen et al[41]
Olive mill waste	3,4-Dihydroxyphenylglycol		Rodríguez et al[42]
Olive Mill Waste Waters	Phenolics	TBARS, LOOH, DPPH, superoxide anion scavenging	Visioli et al[43]
Soybean oil deodorizer distillate	Phytosterols		Yang et al[44]
Sorghum	Phenolics, 3-Deoxyanthocyanidins, vitamin E, carotenoid	TPC, DPPH	Cardoso et al[45]
Buckwheat	Phenolics	ABTS	Serpen et al[46]
Fermented rice bran	Phenolic acids	DPPH, inhibition of peroxidase and polyphenol oxidase	Schmidt et al[47]
Wheat bran	Phenolics	ABTS	Serpen et al[46]
Barley	Phenolics	ABTS	Serpen et al[46]
Rice bran	Peptides	ORAC	Wattanasiritham et al[48]
Peanut skin	Flavanols and phenolic acids	TPC, DPPH, stability of ghee	El-Shourbagy and El-Zahar[25]
Peel from yellow cashew	Phenolic acids and flavonoids	Soluble phenols and total flavonoids, DPPH, ABTS	Moo-Huchin et al[30]
Peel from red cashew	Phenolic acids and flavonoids	Soluble phenols and total flavonoids, DPPH, ABTS	Moo-Huchin et al[30]
Brewery waste stream	Phenolic acids and flavonoids	DPPH	Barbosa-Pereira et al[49]
Brewery waste stream	Phenolic acids, flavanols and flavonols	TPC, DPPH, β-carotene bleaching assay	Barbosa-Pereira et al[50]

Table 1: Sources, active compounds and evaluation methods of food waste products, divided by sector type of wastes.



Gallic acid (Hydroxybenzoic acid)
Caffeic acid (Hydroxycinnamic acid)
Resveratrol (Stilbene)
Quercetin (Flavonol)
Rutin (Flavones)
Catechin (Flavanols)
Naringenin (Flavanones)
Cyanidin (Anthocyanidins)
Genistein (Isoflavonoid)

Figure 3: Chemical structures of compounds belonging to different phenolic classes.

of numerous agriculture wastes and by-products including underutilized fruits, fruit wastes, vegetable wastes, by-products and hulls. They measured phenolic content, as well as radical scavenging activities using DPPH assay. Most extracts exhibited potent antioxidant activities, with Chinese quince, immature pear and apple, buckwheat hull, and grape seed showing both high phenolic contents (34.5 to 43.9 mg chlorogenic acid hemihydrate equivalent/g dry sample), as well as strong DPPH activity. On the other hand, grape bunch stem and perilla pomace showed high phenolic contents but insignificant DPPH activity.

Solid residual from olive oil production is one of the most widely studied waste materials. Wastes are generated during washing, malaxation, centrifugation and filtration process (Figure 4). The significant scale of olive oil production in Mediterranean countries generates large quantities of wastes that contain skin, pulp, and kernels pieces. It was said that only 2% of the phenolic compounds are transferred to the oil while 98% remain in the cake.[52] Obied et al[53] summarized the major biophenols in Olive Mill Waste (OMW), including hydroxytyrosol, oleuropein, tyrosol, caffeic acid, vanillic acid, verbascoside, elenolic acid, *p*-courmaric acid, catechol, and ruin. Among them, hydroxytyrosol, oleuropein, caffeic acid, vanillic acid, elenolic acid, *p*-courmaric acid, and catechol were also proved to possess antimicrobial activities. Lozano-Sánchez et al[54] also concluded hydroxytyrosol, tyrosol, decarboxymethyloleuropeinaglycone, and luteolin to be the major phenolic compounds in the by-products generated during the filtration process of extra virgin olive oil. Regarding the commercialization and full utilization of the biophenols from olive mill waste, it was emphasized that the divergence in variety, season, geography and agriculture conditions can affect phenol profile.[53] Of the same importance is the storage of these chemically and microbiologically active raw materials prior to extraction.

Apple is another widely studied matrix in the fruit category. In the apple juice industry, about 75% of apple is utilized for the juice and the remaining is the by-product, apple pomace.[5] Apple pomace contains different kinds of nutrients including carbohydrate, pectin, crude fiber, proteins, vitamins, minerals, and antioxidants. Due to the large quantity of produced apple pomace, the preparation of a single product would not be economically feasible and production of alternative products should be explored.[5] A study[56] that investigated underutilized crab apples, along with other genotypes such as Honeycrisp, Red Delicious, McIntosh, and Royal Gala found that the skin extracts of some crab apple varieties had higher antioxidant activity than the more common varieties, and was effective in inhibiting oxidation of a methyl linoleate emulsion system (Table 2).

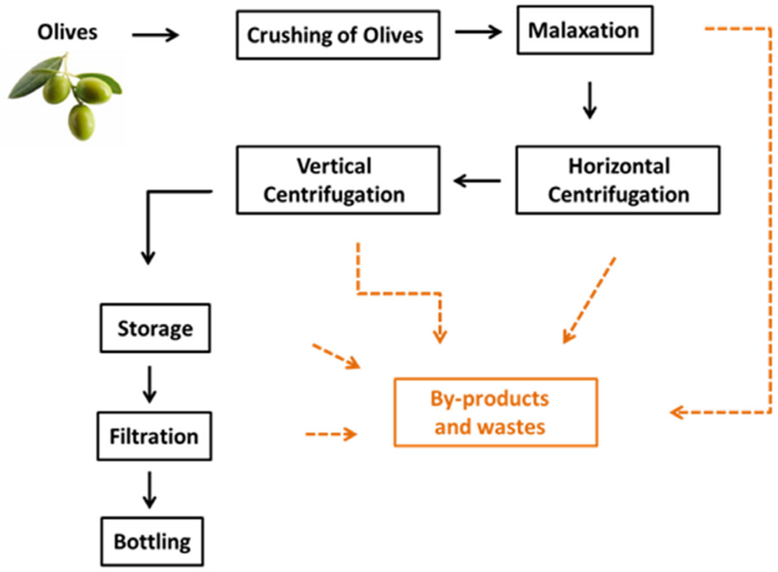


Figure 4: Olive oil production process and source of waste (adapted from Frankel et al[55]).

Apple genotype		Total phenolic by HPLC-MS/MS (mg/100 g DW)	FRAP (gTE/100 g DW)	ORAC (gTE/100 g DW)	% Inhibition of methyl linolenate oxidation
Crab apples	Crabapple – r3t4	551	1.94	13.4	94.5
	Antanovka	290	1.25	10.3	96.5
	Dolgo	642	3.28	14.2	95.7
Commercial cultivars	Red Delicious	380	2.17	12.5	91.7
	Royal Gala	484	1.74	13.5	95.7
	McIntosh	296	1.33	6.25	88.8

Table 2: Total phenolic content and antioxidant activities of crab apple cultivar and commercial cultivar (adapted from Huber et al[56]).

Various berries have high amount of wastes generated during industry manufacturing. These include raspberries, blackberries, and chokeberries, etc. Bakowska-Barczak et al[57] investigated black currant (*Ribesnigrum* L.) from five cultivars and found that the seeds have high levels of tocopherols (1143 mg/100 g of oil on average) and phytosterols (6453 mg/100 g of oil on average). Additionally, phenolic compounds can be extracted out of the residues from the oil extraction, adding further value to this by-product processing.

Avocado seed constitutes 16-26% of the total weight of the fruit, yet has been largely been underutilized.[58,59] Despite their high carbohydrate content, the seeds cannot be used to feed livestock because the polyphenols in the seeds delivers bitter taste and possible toxicity. Deepti et al[60] found a total of 283.2±5.8 mg/g Gallic Acid Equivalent (GAE) phenolic content of anaqueous methanol extract of avocado seeds. Soong and Barlow[61] measured phenolics in the seed and the pulp and found 88.2 mg/g GAE in the seed and only 1.3 mg/g GAE in the pulp. The same study showed that the ethanol/water extract of the seed has a radical scavenging capacity and a ferric reducing power 55 and 155 times higher than of the pulp, respectively. Catechin, epicatechin and their oligomers have been identified as the major phenolic compounds in the seeds[38]; other phenols such as protocatechuic acid, vanillic acid and kaempferide were also found present.[58]

Exotic fruits also produce large quantities of waste by-products, including peels, seeds and flesh. Ayala-Zavala et al[62] proposed an interesting idea of using the fruit's own by-product to provide antioxidant and antimicrobial protection to fresh-cut fruits. They pointed out that utilizing fresh-cut fruit by-product to enrich antioxidant capacity while offering antimicrobial protection to the final fresh-cut produce would fulfill consumers' requirements of naturally preserved products, as well as lead the industry to a lower-waste agribusiness. The same study also found significantly high levels of phenols and flavonoids in mango seeds and peels, when compared to by-products from mandarin, apple, papaya, and pineapple. This result is supported by Abdalla et al[63] who found the combination of both mango seed kernel extract and oil had high antioxidant potency. Ajila et al[64] also found increased radical scavenging activity when mango peel powder was added into macaroni. Macaroni stability was not evaluated as part of this study. Morais Ribeiro da Silva et al[65] compared the phenolic contents in the by-products (peel, pulp waste, and seed) generated during the pulp production process of 12 tropical fruits, with the phenolic contents of the pulp. Among the results, pineapple, Surinam cherry, sapodilla have much higher phenolic in the by-products, while acerola, mango and papaya showed the opposite results (Figure 5).

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The amount and types of active compounds in vegetables differ from fruits. A study by Wijngaard et al[32] found fruit waste and by-products generally show a higher antioxidant activity than vegetable waste and by-products.[63] Potatoes are one of the mostly consumed vegetables around the world and the peels are the major by-product of potato processing industries. Mohdaly and Smetanska[66] studied the antioxidant activity of potato peel, sesame cake and sugar beet pulp extracted using different solvents. Methanol extraction gave the highest phenolic contents, as well as the highest antioxidant activity when compared to BHA and BHT. Their activities, however, were inferior to that of TBHQ. Potato peel extracts exhibited the strongest antioxidant capacity in all assays (ABTS, DPPH, and β–carotene bleaching), followed by sugar beet pulp and sesame cake. Several other studies also evaluated potato peels. Kanatt et al[67] tested the effectiveness of potato peels to retard oxidation in radiation processed lamb meat. To be used as an antioxidant, potato peel extract would be added to the meat before irradiation. An advantage of this treatment is that the irradiation process actually increased the yield of phenolics in the potato peels. Chlorogenic acid was identified as the major antioxidant compounds in the peels, followed by gallic acid and protocatechuic acid. The addition of potato peel extracts at the concentration of 0.04% largely inhibited the formation of carbonyls and reduced Thiobarbituricacid (TBA) value during 7-day storage of the lamb meat at 0-3 °C. A similar benefit was found in seafood. Farvin et al[68] found a positive effect of potato peel extract on the stability of minced horse mackerel. They identified high amounts of protocatechuic acid in both ethanol and water extracts. Water extracts were shown to contain significantly ($p<0.001$) higher levels of gallic acids and p-hydroxy benzoic acid when compared to ethanol extracts, while the ethanol extracts showed higher levels of gentisic and caffeic acids. The differences in the amount of extracted actives may contribute to the different activity of the two extracts.

Krisch et al[4] identified two concerns limiting the by-product exploitation in their studies of eleven fruits and vegetables including apple, strawberry, pear, red beet, artichoke, asparagus, tomato, broccoli, cucumber, endive, chicory, and two minor crops including golden rod and woad. One concern is the heterogeneity and missing specification of cultivars, provenance, storage time and process conditions. The second concern is the high cost entailed due to the requirements for fast and soft drying conditions of the usually high moisture raw material. These problems are in agreement with Obied's[53] observation for the olive mill industry.

APPLICATION IN FOOD PRODUCTS

Despite the large amount of research that examined the antioxidant potentials of agriculture by-products, only a few studies tested them in real food systems. Lante et al[37] tested the ability of red chicory by-products (leaves and stems) in improving the stability of soybean oil, corn oil and peanut oils. 0.015% or 0.15% of the extracts was compared to the same concentrations of BHT. They found that the red chicory extract provided some improvement over control sample, but was not comparable to BHT. Also, the antioxidant performance was similar at low and high concentration, indicating a saturation of its antioxidant capability. El-Shourbagy et al[25] studied the stability of ghee by measuring induction time at 130 °C. Stability study at 63 °C for 21 days was also conducted using peroxide value, acid value and

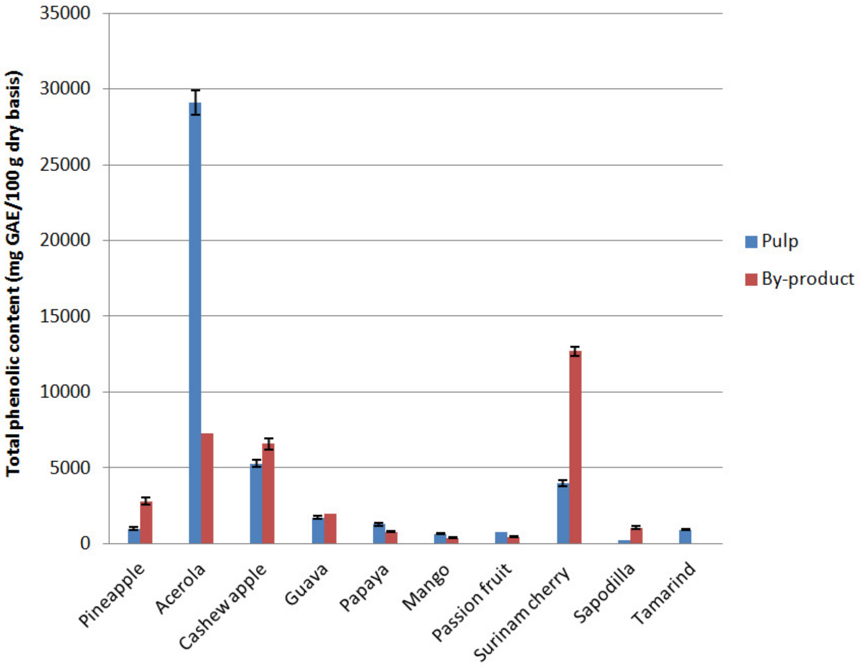

Figure 5: Total phenolic content of pulps and by-products of tropical fruits from Brazil obtained during pulp production process (adapted from Silva et al[65]).

TBA as the evaluation parameters. They found peanut skin and pomegranate peel extracts added at 200 ppm showed equal performance compared to 200 ppm BHT; olive showed comparable performance at a higher concentrations of 600 ppm. Farvin et al[68] looked at the effect of adding potato peel extracts at concentrations levels of 0.24% or 0.48% into minced horse mackerel. The meat was stored in aluminum boxes at 5 °C for 96 h. Ethanol extracts of potato peels showed superior performance than water extract, significantly reducing peroxide value, tocopherol degradation, formation of volatile secondary oxidation products such as 1-pentene-3-ol,and 2,4-heptadienal, and retarding the loss of protein. In these studies, performance of extracts in food products did not always correlate with that of *in vitro* antioxidant capability tests, which reinforces the need of evaluating activity beyond test tube scales.

CONCLUSION

Increasing evidence has pointed towards the benefit of adding value to agriculture and food processing waste which contains high levels of antioxidant compounds for the potential of inhibiting lipid oxidation and slowing down the onset of rancidity in foods. There's no doubt that these materials possess certain advantages in contrast to synthetic antioxidants or isolated natural compounds in terms of consumer acceptance and legal requirements. Nevertheless, the exploitation of waste products is yet to be a mature industry due to several obstacles. One obstacle is the inconsistency in final products caused by the variance in raw materials and processing conditions. Another obstacle is the implied high cost from the processing step of food waste materials.[5] Thus it is important to obtain valuable and high-value added products to justify the investment. Finally, results of *in vitro* antioxidants assays do not always correlate antioxidant activity in food products. Therefore, shelf life studies in food systems need to be conducted before the real antioxidant capacity can be determined.

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