Coffee’s Melanoidins. A critical review of contemporary scientific literature

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Abstract: Melanoidins are brown pigments thermally generated during the non-enzymatic Maillard reaction and are present in a large number of baked and roasted food products (e.g., bakery products, dark beer, coffee, etc.), conferring their typical color and improving their appearance, which is usually considered, by the end-consumer, as an indicator of quality. After all, quality is in the eye of the beholder. The amount of melanoidins varies depending on the precursors’ concentration and the type of processing to which a given food product is submitted (baking time + temperature). Additionally, melanoidins have been in our diets for millennia, not only improving the organoleptic qualities of food but also exerting a great array of physiological benefits directly linked to their chemical composition, molecular conformation, and structural size. Aside from their prebiotic effects, melanoidins also display other beneficial properties, among which the most salient are their antioxidant capacity, antibacterial and chelating activities, and anticancer action. However, regardless of the plethora of in vitro experimental evidence that validates the properties mentioned above, there is still controversy about their significance for human health since many of these properties seem to be associated with high molecular weight melanoidins, which, because of their size, cannot cross the intestinal wall suggesting their action is relegated to the intestinal tract where after being fermented and fragmented are finally converted in a series of metabolic derivatives some of which manage to cross into the bloodstream while others are simply excreted through the feces. The following is a synthesis collected from the available scientific literature which aims to elucidate several aspects of melanoidins (i.e., synthesis, determination, metabolism, & biological activity) to create awareness about their importance for human health and provide information about where to find them to improve our diets.

Key words: Synthesis, fractionation, separation, antioxidant activity.

Introduction

Melanoidins are a complex mixture of different polymers (e.g., carbohydrates [polysaccharides], proteins, amino acids and chlorogenic acids)\(^1,2\), which not only make up the majority of the high molecular weight molecules present in roasted coffee but also contribute to the aroma\(^3,4\) and typical coffee coloring thanks to its ability to absorb solar radiation in oscillating quantities between 405 to 420 nm\(^2\). In addition, they are also responsible for the brown coloring of unroasted and decaffeinated beans.

Melanoidins are nitrogen-containing compounds resulting from the last stage of the non-enzymatic Maillard reaction\(^7,8,10,12-16\). Although coffee is considered its main source in the human diet, they are also produced during the processing of a series of foods such as bread, meat, malt, cocoa, and honey, among others\(^1,4,8,13\).

Their molecular structure is complex and unpredictable (Figure 1)\(^15,16-17\) to the point that in some cases most of their molecular weight (≤ 90%) is constituted by unknown materials\(^15\); however, structurally speaking, they bear a resemblance to the cereals’ arabinoxylans\(^7\), and although there is a substantial variation concerning their size, the consensus is that their molecular weight ranges between 2 to 22 kDa\(^2\).

Melanoidins’ contribution to the dry weight of a substrate is calculated by subtraction after eliminating the percentage contribution of all other known compounds\(^1,2\). Using this technique, it has been calculated that they constitute between 23 to 25% of the dry weight of roasted coffee\(^1,2,4,16,20-24\) and 29 to 30% of the dry weight extracted from the coffee drink\(^1,7,20,21\). Additionally, the observed variations within this range are linked to the species/variety, the degree and type of roast & the infusion preparation method (Figure 2)\(^25\).

Regarding the type of roasting, there is evidence that roasted coffees produced by adding sugar during the roasting process contain a higher content of melanoidins (7-10%) than those conventionally roasted\(^27\).

In addition, melanoidins concentration can also be calculated thanks to their ability to absorb ultraviolet light at 405 to 420 nm\(^2,3,10,26\) and with the help of the extinction coefficient (\(c\)) the concentration (c) of melanoidins present in a roasted coffee sample\(^1,9\) can be calculated using the Beer-Lambert equation (\(A = εcd\)).

Melanoidins have different physicochemical properties that are not shared by all of them, such as their hydrophobicity or their metal ions chelating capacity \(\text{(e.g., Fe}^{3+}, \text{Mg}^{2+}, \text{Cu}^{2+})\), the latter being attributed to their anionic nature, which manifests more strongly in the high molecular weight molecules\(^1,2,9,13,23,29\) and which seems to have its origin in the presence of chlorogenic acids coupled to its structure\(^9\).

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However, there is also experimental evidence that suggests that, in the absence of chlorogenic acids, the type of amino acid involved during its formation also plays an important role in defining its electrical charge. In addition, they also can reduce hydroperoxides in non-radical compounds through the donation of a hydrogen atom.

Synthesis

Although the mechanisms responsible for the formation of melanoidins in foods are not yet clearly elucidated,

the evidence suggests that, in coffee, there is a positive relationship between their synthesis and the degree of grain roasting; In other words, the more roasted the grain is the greater the concentration of melanoidins will be.

The high molecular weight carbohydrates (e.g., galactomannans & arabinogalactans) and proteins are considered melanoidins precursors. Currently, three hypotheses describe their synthesis: (a) they are formed due to the polymerization of unsaturated carbonyl compounds or by the repetitive accumulation of low molecular weight compounds such as furans and pyroles produced in the last stages of the non-enzymatic Maillard reaction (b) they are formed through the interaction of low molecular weight sugars with proteins (Figure 3) (c) or they are formed due to the interaction of sugar derivatives in the early stages of the Maillard reaction which are subsequently polymerized through condensations.

Based on described mechanisms in figure 3, three types of melanoidins can be identified:

a) Melanoproteins are partially brown color water-soluble polymers formed from the reaction between deoxy-sones (sugar degradation products) and proteins. Deoxisones are α-dicarbonyl groups derived from reducing sugars (i.e., glucose and fructose) which act as reactive intermediates during the Maillard reaction;

b) Conversely, some melanoidins are produced when dicarbonyl groups are condensed, during the Maillard reaction, with high molecular weight polysaccharides such as galactomannans and arabinogalactans both constituents of the cell wall.

c) However, there is also evidence of melanoidins interaction with phenolic compounds such as chlorogenic acids or their derivatives (e.g., caffeic, ferulic, coumaric, and protocatechuic acid) which act as reactive intermediates during the Maillard reaction; and their interaction with the amino groups (NH2) of amino acids and proteins results in the formation of low and high molecular weight molecules of up to 100 kDa.

In other words, melanoidins molecular weight depends on the nitrogen-containing compounds (i.e., free amino acids, peptides, oligopeptides, polypeptides or proteins) involved in their synthesis during the Maillard reaction.

Melanoidins’ amino acid composition tends to be similar and the most abundant amino acids present in their molecular conformation are glutamic acid, glutamine, glycine, alanine, aspartic acid, isoleucine, leucine, and proline.

According to Moreira et al., about 50% of the high molecular weight melanoidins present in the coffee drink correspond to arabinogalactan-protein complexes (AGPs). AGPs are complexes formed by arabinogalactan covalently bound to proteins with a high content of hydroxyproline, an amino acid.
and quinic acids) through covalent and non-covalent bonds (e.g., hydrogen, hydrophobic, and ionic bonds) resulting in melano-chlorogenic complexes1,4,13,15-17,21,33 which also have antioxidant properties6,29,34. However, although the raw grain has up to 10% of chlorogenic acids, less than 1% of them are incorporated into the molecular structure of melanoidins 2. This happens because chlorogenic acids are thermally degraded during grain roasting. In addition, the incorporation of chlorogenic acids in the melanoidin’s molecular structure is the cause of the development of dark pigments in other food matrices (e.g., sweet wine and grape syrup)2,9.

Finally, there is evidence of coffee melanoidins’ interaction with a variety of low molecular weight volatile compounds suppressing or reducing their organoleptic attributes12,23. For example, among the volatile compounds with sensory properties capable of forming complexes with melanoidins through covalent bonds, are the thiols (e.g., 2-furfurylthiol, 3-methyl-2-butentiol, 2-methyl-3-furantio, & methanethiol),2,12,23,34 the 3-mercapto 3-methyl butyl for mat,2,12,23,34 the sulfides, pyrroles, pyrazines, pyridines, and the diketones2,12 which causes the suppression of the typical sulfured and roasted odors of coffee14.

**Determination of melanoidins**

Empirically, we can say that the greater the degree of roasting, the darker the appearance of the grain will be and the higher the concentration of high molecular weight melanoidins. In contrast, to a lesser degree of roasting, the grain coloration will be clearer, and low molecular weight melanoidins will prevail35.

On the other hand, the main limitation in studying melanoidins structure is the lack of specific analytical methods for their isolation and separation into homogeneous fractions. One of the most common approaches is the isolation by molecular weights using different fractionation techniques that separate melanoidins based on their molecular weight13,16. Among these techniques, we can mention ultrafiltration, diafiltration, and dialysis. The foundation of these techniques is the separation of particles based on their molecular weights using semipermeable membranes with specific pore sizes or hollow fiber with cut-off points ranging from 3 to 100 kDa22.

Such techniques can remove low molecular weight molecules; However, modified polysaccharides and proteins with high molecular weight are retained together with melanoidins13,14,16. The result of this fractionation is the recovery of the High Molecular Weight Material (HMWM). Next, the techniques' basis used for the melanoidins isolation process is briefly described as follows:

**Ultrafiltration:** is the process through which substances are divided, separated and concentrated without them undergoing phase changes. In addition, a semipermeable membrane with defined pores is used to limit the particle size that will pass through it. Because of its semipermeable nature, applying pressure (4-8 atm) facilitates the flow of particles through the membrane with cut-offs ranging from 100, 50, 10 to 3 kDa. In other words, the size of molecules capable of passing through the membrane will depend on the membrane's average pore diameter and applied pressure.

**Diafiltration:** is a type and specialized form of ultrafiltration where the retained fraction is diluted in water and recirculated so that the concentration of soluble components is reduced, thereby increasing the concentration of insoluble compounds in the retained fraction.

**Dialysis:** is a form of molecular filtration through which molecules are separated according to their size, by using semipermeable membranes with pores of dimensions smaller than macromolecular. These pores block the passage of larger molecules but allow small molecules (e.g., solvents, salts, and small metabolites) with cut-off (molecular weights) ranging from 2, 12 or 14 kDa to diffuse through the membrane.

**Separation (isolation or purification)**

Once the HMWM has been isolated, the separation of the melanoidins from it is performed based on their Physico-chemical properties:
Solubility

The solubility of coffee melanoidins depends on the polymer size and its chemical nature. Once HMWM is obtained, polysaccharides-derived melanoidins can be separated based on their solubility. The fractionation is performed using EtOH: H₂O in different proportions. The galactomannan-rich fraction is precipitated in 40 to 50% EtOH whereas the arabinogalactan-rich fraction is precipitated in 75 to 80% EtOH. In contrast, the low carbohydrate fraction remains soluble in 75 to 80% EtOH.

The Yariv phenyl glycoside reagent is used to induce the precipitation of melanoidins containing water-soluble arabinogalactan-protein (AGP) complexes.

Electrical charge

The type of amino acid present during their synthesis determines the anionic property of melanoidins. According to their anionic behavior, coffee melanoidins can be isolated by anion exchange chromatography. When precipitation with EtOH does not allow the recovery of homogeneous fractions, combined methodologies can be applied. For instance, ion-exchange chromatography followed by purification through metal ion affinity chromatography (e.g., Cu²⁺) can be used in the first stage.

Chelating capacity

It is based on separating fractions with chelating capacity from the non-chelating ones using techniques such as Cu²⁺ affinity chromatography.

Hydrophobicity

The separation mechanism is based on hydrophobic interactions induced by salt formation between the melanoidins’ non-polar functional groups and the stationary phase ligands. The coffee melanoidins have a hydrophobic behavior due to their average molecular weight. Based on this property, melanoidins can be separated from the coffee infusion or the HMWM using gel filtration chromatography (Sephadex-LH20) followed by hydrophobic interaction chromatography (Octylsepharose) that separates them based on their surface hydrophobicity. A general scheme of melanoidin purification is shown in Figure 4.

Quantification

The content of melanoidins, in a food sample, can be measured at an absorbance ranging from 405 to 420 nm using a specific extinction coefficient to spectrophotometrically estimate the level of browning (absorbance values) about the concentration of melanoidins as a means to characterize them in coffee.

According to the Beer-Lambert equation \( A = εcλ \), there is a direct linear relationship between absorbance (A), and concentration (c). To quantify the melanoidins using this equation, the specific extinction coefficient (ε) for each substance must be known.

These coefficients have been calculated through reaction models of different sugars and amino acids, thus producing various types of melanoidins. After their evaluation, the specific extinction coefficients for each type have been reported.

Structural characterization

The characterization of the different recovered fractions is done through two-dimensional Nuclear Magnetic Resonance (NMR) as in the studies by SQHC (Simple Quantum Hetero-Nuclear Correlation).

The analysis is done by grouping the compounds according to their carbon-hydrogen correlation and the chemical displacement of H+ and C+. This technique allows the correlation between the H+ and C+ of different compound groups.

These experiments have shown the correlation between H+ ions coupled to C+4 atoms linked to hetero atoms such as oxygen and nitrogen (e.g., HC-O & HC-N) which differ from the typical bonds of proteins and carbohydrates. This allows inferring the presence of chromophores substances in the complex structure of melanoidins.

The above-described stages were applied in a study developed by Gniechwitz et al., in which hydrophobic interaction chromatography was implemented in the analysis of the fraction poor in carbohydrates and amino acids but possibly rich in chromophores and antioxidant compounds present in arabica coffee.

For this, fractionation was performed by ultrafiltration, recovering fractions with molecular weights ranging between 3 to 10, 10 to 50, 50 to 100, & > 100 kDa. However, the predominant fractions were those with molecular weights ranging between 3 to 10 & > 100 kDa which represent...
about 37 & 38% of the high molecular weight material, respectively\(^\text{16}\).

The predominant fractions were subsequently isolated with Sephadex LH-20 using aqueous NaCl (0.5 mol l\(^{-1}\)) as the mobile phase until fraction A (SepA) was eluted. Subsequently, fraction B (SepB), characterized by intense brown color, was also eluted with water. In addition, fraction B constituted 4 & 13% of fractions with a molecular weight ranging from 3 to 10 & > 100 kDa respectively\(^\text{16}\).

Monitoring was performed using a UV detector at 405 nm, and the fractions were collected at 32 min intervals. Subsequently, the recovered fractions (ie, SepA & SepB) were washed with water by ultrafiltration to remove the salt. The fractions were dried and weighed for further analysis.

Fraction B was subjected to another fractionation via hydrophobic interaction chromatography with Octyl Sepharose using aqueous NaCl as solvent (Figure 5)\(^\text{16}\). The detection was performed at 405 nm, and the fractions were collected at 20 min intervals\(^\text{16}\). However, the results indicated that a significant additional fractionation was not possible when applying this procedure (Figure 6).

In figure 6 the result obtained during the chromatographic separation, with a UV detector, of the fractions re-covered by ultrafiltration using Sephadex LH-20 is shown graphically. The molecular masses of the predominant fractions ranged from 3 to 10 kDa & > 100 kDa.

The results show the importance of the membrane’s pore size. It can be seen in figure 6.

On the other hand, the anomeric 1H/13C correlations were characterized by their low intensity and could correspond to carbohydrates present in the analyzed fraction. Finally, there were correlations of unknown compounds which do not correspond to carbohydrates or proteins.

The HSCQ-NMR analysis shows no evidence of ferulic acid.
or caffeic acid presence, so it is inferred that phenolic compounds bound to melanoidin structures were linked through a phenolic condensation reaction.

**Metabolism**

The average daily melanoids consumption is approximately 1 to 2 g\(^4\), (others have reported 0.2–2.6 g per day\(^1\)), but for those who consume ≥ 6 cups of coffee daily, the intake can reach up to 5 g reason why it is estimated that coffee's melanoids can contribute to meet the daily require - ment of dietary fiber (~ 10 g day\(^-1\))\(^3\),\(^1\). However, only the soluble fraction should be taken into account as insoluble melanoids are retained in the paper filters used in many coffee makers\(^3\)\(^1\).

However, although several authors qualify them as dietary fiber, the truth is, that they are not coffee bean polysaccharides; in consequence, they are not part of the dietary fiber per se\(^3\),\(^1\). The most accepted dietary fiber definition includes all plant polysaccharides (i.e., hemicelluloses, cellulose, lignin, oligosaccharides, polysaccharides, pectins, gums, and waxes) present in foods that are not degraded by the human’s small intestine enzymes\(^1\).

In addition, dietary fiber is divided into two fractions: soluble and insoluble. The soluble fraction has a greater water retention capacity, reduces the lipids and carbohydrates absorption in the small intestine and promotes bacterial multiplication\(^4\). In contrast, the insoluble fraction has limited water retention capacity but promotes fecal motility through the digestive tract so it effectively mitigates the effects of constipation.

Furthermore, the high molecular weight carbohydrates extracted from food in a chemical, physical or enzymatic way are also considered as dietary fiber as well as those of synthetic origin that have proven health benefits.

However, the presence of a protein fraction and phenolic compounds not only increases the complexity of melanoids structures (which are not clearly elucidated) but also means that they do not conform to the above-described definition despite the fact they show many dietary fiber characteristics\(^3\),\(^1\).

High molecular weight melanoids are not digested in the small intestine and due to their alleged inability to cross the intestinal wall, they end up being fermented by microorganisms in the large intestine (colon) being partially fragmented, and in the process, producing high concentrations of acetate and propionate.\(^4\),\(^1\),\(^8\),\(^3\) These can reach concentrations of 0.5 to 2.0 g day\(^-1\) in the case of moderate and constant drinkers respectively\(^7\),\(^1\),\(^4\),\(^3\) to then be excreted through feces.

In contrast, the evidence suggests that low molecular weight melanoids partially cross the intestinal wall (~
and, after being metabolized, end up being excreted in the urine. According to Finot and Magenat, experiences with rodents indicated the urinary excretion of low molecular weight melanoidins was 27% whereas 61 and 87% of the low and high molecular weight fractions respectively were excreted through feces.4

Biological activity

Although there is in vitro and in vivo evidence of a series of biological activities attributed to melanoidins, there is also doubt about the relevance that these could have on human health; since being molecules of variable molecular weight there is no confirmation of their ability to cross through the intestinal wall (duodenum).1 4,7,15,18,21,

Among the biological activities attributed to melanoidins we can cite its anti-caries effect by preventing Streptococcus mutans (causative agent of caries) from adhering to the surface of the teeth, thus preventing plaque formation4,7,9; In the absence of an external cell membrane 9. This effect is greater with high molecular weight melanoidins due to their ability to form complexes with cations such as Fe+3 and Mg+2 thereby damaging proteins, lipids, and DNA 35,43. Additional properties include an antibacterial effect, antioxidant capacity through the inhibition of the angiotensin-converting enzyme (ACE) which is a blood pressure regulatory factor9,8,4. The ACE is a zinc (Zn+2) dependent dipetidyl carboxypeptidase which once activated its function is to turn angiotensin I into angiotensin II, a potent vasoconstrictor8,44-46, thus causing hypertension.4

Although the molecular mechanisms through which melanoidins inhibit in vitro the action of ACE have not been elucidated, the prevailing hypothesis is based on their ability to sequester Zn+2 thus preventing its activation8. However, excessive coffee consumption should be taken with caution due to anti-nutritional effects such as acrylamide and phenolic acids as well as transition metal ions (eg, Ca+2, Fe+3, Cu+2, Mg+2, Zn+2) forming chemically inactive complexes with varying degrees of digestibility4,8,9. The ability of melanoidins to chelate Cu+2 is of particular importance since this transition metal can alter homeostasis and possibly indirectly induce intracellular oxidative stress thereby damaging proteins, lipids, and DNA.26,43. Additionally, their ability to form complexes with cations such as Fe+3 and Mg+2 gives melanoidins an antimicrobial effect7.

Chelating activity

Due to their negative charge, melanoidins are considered chelating agents due to their ability to couple with other compounds with opposite charges (cations) such as acrylamide and phenolic acids as well as transition metal ions (eg, Ca+2, Fe+3, Cu+2, Mg+2, Zn+2) forming chemically inactive complexes with varying degrees of digestibility4,8,9. The evidence suggests that at low concentrations, in vitro melanoidins exert a bacteriostatic effect on the digestive tract bacteria such as Gram-negative (i.e., Escherichia coli, Proteus mirabilis, Pseudomonas aeruginosa, and Salmonella typhymurium) and Gram-positive (Staphylococcus aureus & Bacillus cereus) at ≥ 4 mg ml⁻¹ and to 3 mg ml⁻¹ respectively, probably due to its chelating capacity1,4. However, Gram-positive bacteria are more susceptible to melanoidins’ antimicrobial activity presumably as a result of the absence of an external cell membrane4. On the other hand, the evidence suggests the existence of a positive correlation between the bacteriostatic effect and the coffee roasting degree, that is, the more roasted the coffee the greater its bacteriostatic capacity9. Also, the bacteriostatic effect is greater with high molecular weight molecules and decreases as their size are reduced1,4,46.

In contrast, at high concentrations, melanoidins exert a bactericidal effect4 attributed to their chelating capacity and to elucidate this phenomenon, three possible mechanisms have been proposed: (a) the bacteriostatic activity is due to Fe+3 ions chelation when, in solution, melanoidins are present in low concentration1,4,9; (b) in the presence of siderophores producing bacteria (i.e., molecules specialized in Fe+3 ions sequestration) then melanoidins chelate the siderophore-Fe+3 complex thus limiting the microorganism multiplication and (c) in high concentration, melanoids can subtract Mg+2 ions from the bacterial cell membrane causing it to rupture4,7,9,25.

Antioxidant activity

The ability to transfer H+ ions thus turning free radicals (eg, NO+, NO-, ONOO-, H2O2, O2·-) into stable non-reactive molecules is one of the mechanisms through which melanoidins inhibit lipids oxidation which otherwise contributes to the development of atherosclerosis in humans24,25,35 and in the case of processed foods they contribute by extending their shelf life by preventing oxidative rancidity of mono and polyunsaturated lipids1,4,9 and by preventing the proliferation of microorganisms due to their bacteriostatic and bactericidal effect1,9. For this reason, they can be used as additives in the food industry. Its antioxidant capacity is, in part, attributed to chro- nogenic acids (CAs) bound to their molecular structure through undefined bonds and which is presumably manifested by its metallo-chelator and free radical cleaning activi- ties1,4,7,9,24,25,49.

Coffee’s bioactive properties are many. For example, experiences reported by Nakayama et al.47 suggest that in rodents it’s habitual consumption helps to regulate the intestinal microflora due to its antibacterial effect (e.g., bacterios- tatic or bactericidal) that contributed to reducing Escherichia coli and Clostridium spp. populations; while concomitantly increasing the population of beneficial bacteria such as Bifidobacterium spp40,47.

Food compounds capable of promoting the multiplication of beneficial bacteria (e.g., Bifidobacterium spp., Lactobacilli) in the intestinal tract and simultaneously reducing populations of pathogenic bacteria are known as prebiotics18.

This result could be attributed to coffee melanoidins ability to regulate the colon’s microflora7,13,42 presumably because of its high molecular weight polysaccharide fraction which is similar to that observed in cereals.2 This effect has been corroborated in human trials which demonstrate that a melanoidins-rich diet increases the concentration of bifidobacteria in the participants’ feces.4

The evidence suggests that at low concentrations, in vitro melanoidins exert a bacteriostatic effect on the digestive tract bacteria such as Gram-negative (i.e., Escherichia coli, Proteus mirabilis, Pseudomonas aeruginosa, and Salmonella typhymurium) and Gram-positive (Staphylococcus aureus & Bacillus cereus) at ≥ 4 mg ml⁻¹ and to 3 mg ml⁻¹ respectively, probably due to its chelating capacity1,4. However, Gram-positive bacteria are more susceptible to melanoidins’ antimicrobial activity presumably as a result of the absence of an external cell membrane4. On the other hand, the evidence suggests the existence of a positive correlation between the bacteriostatic effect and the coffee roasting degree, that is, the more roasted the coffee the greater its bacteriostatic capacity9. Also, the bacteriostatic effect is greater with high molecular weight molecules and decreases as their size are reduced1,4,46.

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which also have the antioxidant capacity, as has been demonstrated through the study of these in cereals. Unfortunately, analyzing arabininoxylases’ antioxidant capacity in coffee is expensive and difficult to measure due to the simultaneous presence of chlorogenic acids and other phenolic compounds in the same matrix.

In addition, melanoids antioxidant activity is not limited to the lower intestinal tract (colon) or blood plasma since there is experimental evidence that shows a reduction in free radicals’ synthesis (hydroperoxides) during a simulated turkey meat gastric digestion assay. High molecular weight melanoids (> 10 kDa) isolated from an instant coffee sample were incubated (37 °C) with a turkey meat sample during an in vitro digestion test, in which they were able to reduce hydroperoxides levels when melanoids concentration reached 1.5 mg ml⁻¹. Still, its suppressive effect was even more significant when reaching 3 mg ml⁻¹.

From previous experience, it can be deduced that melanoids can inhibit the production of free radicals at the stomach level, and the intensity of the suppressive effect is linked to their concentration.

Of particular importance is the effect of the degree of roasting on the coffee drink’s antioxidant capacity. It has been shown that at a higher degree of roasting, the concentration of CAs is reduced (due to its degradation), and consequently, the coffee drink’s antioxidant capacity is lower. In contrast, the evidence shows that the higher the degree of roasting, the higher the concentration of high molecular weight melanoids⁴⁹.

However, low molecular weight melanoids have a higher antioxidant capacity than those of high molecular weight because they form complexes with a greater number of phenolic compounds while those of higher molecular weight are affected by steric hindrance that decreases their ability to react with other compounds.

Consequently, the synergistic effect of the loss of CAs and a higher concentration of high molecular weight melanoids would cause a drastic decrease in the antioxidant capacity of black or roasted coffees compared to what is observed in moderately roasted ones.

In contrast, Torrefacto coffees that normally show an intense black color are an exception to the rule because due to their higher content of melanoids they also have a greater antioxidant capacity compared to what is observed in conventionally roasted coffees.

In summary, melanoids are responsible for approximately 26 to 38% of the antioxidant capacity of coffee drinks through different mechanisms such as electron transfer and scanning of oxygen (O₂) molecules or hydroxyl radicals (HO)⁴⁹,²².

**Anticancer activity**

Melanoids can inhibit, in vitro, the metalloprotease matrices (MMP-1, MM-P2, and MM-P3) which are a class of endopeptidases associated with the development of inflammatory and degenerative diseases as well as playing an important role in tumor growth (especially at the level of the colon) and metastasis which results in possible protection against colon cancer in humans, especially when their concentrations range between 0.25 to 1.0 mg ml⁻¹.

On the other hand, the evidence suggests that the daily consumption of ~ 600 ml of coffee for five consecutive days can protect the human lymphocytes’ DNA from damage caused by free radicals⁴.

In summary, melanoids can prevent the development of colon cancer through a) the increase in intestinal motility which helps the rapid elimination of carcinogens through feces, b) through its prebiotic effect and c) by absorbing and neutralizing free radicals⁵⁹.

**Conclusions**

From a nutritional standpoint, lightly or medium roasted coffees are more convenient, for the end consumer, due to a greater concentration of melanoid-chlorogenic complexes with a stronger antioxidant effect attributed to the presence of chlorogenic acids or their monomeric constituents. However, the general public’s preference for black or well-roasted coffees demonstrate that the end consumer choice is not driven by nutritional facts, but by the product’s organoleptic properties.

This, of course, does not imply the public is well-informed about nutritional and other important issues (e.g., antioxidant properties, species/varieties, blends, roasting degree, origin) to be able to discriminate between the quality of roasted coffees since such information, in the majority of cases, is not even display in the package label.

Nevertheless, regardless of their lower antioxidant capacity, black or well-roasted coffees tend to have a higher concentration of high molecular weight melanoids which even though are not considered dietary fiber per se partially cover its daily requirement promoting gastrointestinal motility which in turn lessons constipation.

Additionally, due to their inability to cross the intestinal membrane (at the duodenum), the high molecular weight melanoids are forced into the lower intestinal tract (colon) where they exert their prebiotic, antibacterial, bacteriostatic, and anticancer effect during their transition.

Melanoids are found in many common products where their precursors undergo the non-enzymatic Maillard reaction when submitted into thermal processing. Unfortunately, due to the high incidence of metabolic syndrome, which is a collection of physiological disorders that occur concomitantly and are typically associated with impaired glucose metabolism, some of these products (e.g., bakery products, dark beer among others) can not be consumed in sufficient quantities to fulfill the daily dietary fiber requirements as in the case of individuals diagnosed with obesity-related pathologies (e.g., diabetes mellitus type II, non-alcoholic fatty liver disease, abnormal cholesterol or triglyceride levels, and hypertension).

In conclusion, coffee as a source of melanoids seems to be a good alternative for patients suffering from the above-mentioned maladies since the infusion has a glycemic index equal to zero (http://www.montignac.com), provided the beverage is not consumed with added sugar. Nevertheless, it is worth mentioning that coffee melanoids’ properties presented herein should be interpreted only as educational or informational material and never as a substitution for your physician’s advice since all health issues required medical supervision.

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