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Original Article

The FTIR Technique Analyzed, Antioxidant [Hypochlorous Acid Scavenging and Hydroxyl Radical Scavenging] Activity of Black pepper (*Capsicum annuum* L.) Extract with Metabolite Identification

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Abstract:

Capsicum annuum L. or peppers are an important farm crop because the plant is both a vegetable and a spice. The peppers belong to the Solanaceae family. Raw peppers contain phenolic chemicals that are very effective antioxidants and which are involved in a number of defence mechanisms by plants, including vitamins A and C, acidic and neutral ones. Several species and varieties of the genus Capsicum owe their typically spicy smell to the presence of the capsaicinoids, capsaicin and dihydrocapsaicin. Peak (Wave number cm^{-1}), Functional group assignment, Type of Vibration, Bond, Area, Corr. Area and Intensity (667.37, alkyl halides, Stretch, C-Cl, 0.064, and 2.947), (873.75, Alkenes, Bending, =C–H, 0.161, and 2.864), (921.97, Alkenes, Bending, =C–H, 0.006v, and 2.459), (1026.13, alkyl halides, Stretch, C-F, 14.196, and 40.600), (1139.93, alkyl halides, Stretch, C-F, 0.381v, and 6.633), (1234.44, alkyl halides, Stretch, C-F, 0.015, and 2.346), (1317.38, alkyl halides, Stretch, C-F, 0.116, and 3.234), (1379.10, alkyl halides, Stretch, C-F, 0.026, and 3.677), (1415.75, Aromatic, Stretch, C=C, 0.223, and 4.948), (1519.91, Aromatic, Stretch, C=C, 0.065, and 2.191), (1598.99, Aromatic, Stretch, C=C, 0.085, and 3.642), (1740.72, Aldehyde, Stretch, C=O, 0.743 and 2.000), (2852.72, Alkane, Stretch, C-H, 0.244, and 2.649), (2922.16, Alkane, Stretch, C-H, 0.893, and 4.668), (3223.05, Amide, Stretch, N-H, 0.016, and 1.955), (3265.49, Amide, Stretch, N-H, 0.010, and 1.934). Hypochlorous Acid Scavenging Hydroxyl Radical Scavenging [Antioxidant activity] of pepper (Capsicum annuum L.) Crude (Methanolic extract, Ethanol fraction, Water fraction and Ascorbic acid (standard)) recorded (127.00 ± 5.69, 106.94 ± 5.10, 98.05 ± 4.71, and 215.08 ± 8.09 respectively) while recorded (167.05 ± 6.00, 180.93 ± 6.09, 200.45 ± 6.87, and 514.16 ± 18.01 respectively)

Keywords: Hypochlorous Acid Scavenging, Hydroxyl Radical Scavenging, *Capsicum annuum* L., Metabolite, FTIR Technique

Introduction:

The pepper (Capsicum annuum L.) is one of the most largely consumed vegetable fruits in the world. The pepper fruits are characterised by the presence of high amount of minerals as well as vitamin C and A. Thus, by consuming 60-80 g of fruits on a daily basis, you can have all the vitamin C that you require and a quarter of the daily needed amount of vitamin A. This horticulture product contains a number of other health promoting chemicals such as carotenoids, flavonoids and other polyphenols among others in large quantity [1-3]. There are lots of different peppers, and the only method which can be used to distinguish between them is to consider the pericarp (the outer covering of the pepper) and its colour change when it becomes ripe. This diversity also reveals itself in the variety of common names by which pepper fruits are often used in a specifically regional context. Pepper in the kitchen and on the palate The capsaicin content of pepper fruits is what defines the fruits as being either sweet or hot as applied to food and gastronomy. The spiciness of the pepper can be attributed to capsaicin, which is found only in genus Capsicum. You see, so called Scoville scale is the ranking of various pepper fruits by their pungency intensity. The family of capsaicinoids includes at least 22 primary chemicals with the phenylpropanoid alkaloid capsaicin as the chief. Hot pepper cultivars contain about 90 95 capsaicinoids of which dihydrocapsaicin and capsaicin are the sources [4, 5]. These chemicals can be separated and identified by high performance liquid chromatography with electrospray ionisation mass spectrometry (HPLC-ESI/MS), and are mainly localised in the epidermic vacuoles of the placenta and in the septum of fruits. Pharmacologically, capsaicinoids, and especially capsaicin, have been demonstrated to exert a broad range of diverse physiological and biological activities in vitro. Among them are behavior as antioxidants, stimulation of the energetic metabolism, inhibition of lipid accumulation, anti-inflammatory properties, neurone stimulation, and decreased cell death in neurodegenerative diseases [6, 7]. It was also found that capsaicin can activate p53 tumour suppressing protein by phosphorylation and that this proapoptotic effect of capsaicin is also exerted via this TRPV. Another curious circumstance in peppers is the ripening of the fruit; this is best observed when the fruit colour is altered, as in changing green to red, yellow, orange, or purple, but it may occur with any kind of pepper. It involves several changes including degradation of chlorophyll and synthesis of new carotenoids and anthocyanins, liberation of organic volatiles, synthesis of new proteins and degradation of old ones, and loosening of cell walls. In addition, there are thousands of genes associated with the pertinent transcriptome differences between immature pepper fruits (green) and ripe pepper fruits. The additional consequence of ripening fruits according to redox views is the shift in antioxidant capacity and total soluble reducing equivalents that are triggered by the alterations in reactive oxygen species (ROS) metabolism [8, 9]. The activity of significant non-enzymatic antioxidants, such as ascorbate, glutathione, carotenoids, and polyphenols has been followed during pepper fruit ripening, whereas the information on enzymatic antioxidants activity is limited. Antioxidant enzyme systems including superoxide dismutase (SOD), catalase (CAT), and ascorbate-glutathione cycle offer the primary defence against reactive oxygen species (ROS). There are few NADP dehydrogenases that act as a secondary measure to help these antioxidant enzymes. Although there is a lot of information regarding contribution of these antioxidant enzymatic systems in ripening of sweet pepper fruits, little is known about the profile of these systems in hot pepper cultivars [9-11]. Phytochemicals may have some positive health effects and it is possible that they may diminish the number of synthetic drugs needed to treat metabolic problems. It has as well been utilised to determine the range of fruit odour, preference and customer selection as a source of pigments to deepen or modify food colour. Some of the bioactive compounds that determine the antioxidant capacity of peppers include capsaicinoids, polyphenols, flavonoids and ascorbic acid. Colour and the spicy flavour, produced by capsaicinoids are the two most important parameters that are used to define quality. The increased popularity of natural compounds over synthetic compounds in the development of healthy functional foods has triggered research on the use of natural colourants. It has also been found out that the three primary pigments that are found in plants such as chlorophylls, carotenoids and anthocyanins are in reality a mixture that provides vegetables with their colour. Photosynthesis is based on light absorption by chlorophyll, a pigment that makes

plants represent their typical green color. Carotenoids are a provitamin A in the human diet, and play significant biological roles as antioxidants, and have been used as natural food colouring agents to give red, orange and yellow colour. Anthocyanins belong to flavonoid chemicals, which can be present in most parts of plants; their coloration is bluish-purple and red. Pepper is highly variable in nutrition and thus pepper varies greatly in colour, shape and taste, which make its economic significance. Colour is one of the most important features of food products. To fulfill the growing trend in spicy flavours, we are already undertaking research in a large assortment of peppers, depending on consumer demand, to match the increased demand. The peppers could be used to prepare new and useful cuisines. To find new customers and enhance the international competence of Korean pepper producer, there is a need to investigate and compare physicochemical properties and antioxidant capacities of coloured peppers. The results might be applicable in the design of new products to be sold in the foreign market [12]. Colourful peppers (e.g. orange, yellow, and purple) have also appeared within recent years, produced with an array of bioactive compounds. The paper has fulfilled its objectives, which were to examine the antioxidant capacity of black pepper (Capsicum annuum L.), and to determine the metabolites of black pepper by Fourier transform infrared spectroscopy technique.

Materials and Methods:

Plant material

The hot pepper fruits used in this study are all purchased in the local market of hillah, Iraq. Two fruits of Capsicum annuum L. were selected and before storage in a dry and dark place, they were dried in an electric drier at 35 o C. Fruits including their placenta, seeds, and the fruit wall were all extracted. To extract the dry and crushed flavour of pepper, we took 10 grammes of peppers in a Soxhlet apparatus and extracted it using 150 cc of methanol. Slowly refluxed the solution over 9 hours and filtered, cooled and dried the solution using a rotary evaporator system.

Hydroxyl radical scavenging

The assay is based on 2-deoxyribose condensation with TBA in order to quantify the decomposition product, which was slightly adapted to the procedure described by Aruoma et.al. [13]. Hydroxyl radicals could be generated in the Fenton reaction involving the reactions of Fe3+, ascorbate, EDTA, and H2O2. The reaction mixture had 1-methyl-2-ribose (2.8 mM), a KH 2 PO 4 -KOH buffer (20 mM, pH 7.4), ferric chloride (100 1M), EDTA (100 1M), hydrogen peroxide (1.0 mM), ascorbic acid (100 8M), and varying concentrations (0100 200 100 20). The combination had a total volume of 1 millilitre. mixing solution (0.5 ml) The absorbance was read against the blank solution at 532 nm following the cooling of the same. Each test was repeated six times. As a control, Mannitol, the conventional OH scavenger was used. The test solution and the blank solution were used in order to calculate the percentage of inhibition.

Hypochlorous acid scavenging

Immediately before the experiment, we prepared hypochlorous acid (HOCl) by titrating a 10 percent (v/v) solution of NaOCl with 0.6 M H 2 SO 4 to a pH of 6.2. We then determined the amount of HOCl in the solution by measurements of absorbance at 235 nm with the molar extinction coefficient of 100 M -1 cm -1. It was slightly modified and carried out as described by Pedraza-Chaverr [14] who outlined the test in detail. The scavenging activity was measured by decreasing the catalase absorbance at 404 nm. A volume of one millilitre of plant extract was put in a reaction mixture comprising of 50 millimolar phosphate buffer (pH 6.8), 7.2 micromolecular weight of catalase, 8.4 millimolar hydrochloric acid and an increasing concentration (0100 ug/ml) of the plant extract. The mixture was allowed to react at 25 o C in a period of 20 minutes after which the absorbance was read with an appropriate blank. The experiments were repeated six times. Ascorbic acid was a standard useful in the scavenging of hydrochloric acid.

Statistical analysis

Statistical analysis application SPSS (Statistical Analysis application, version 25, IBM Co., Armonk, NY, USA) carried out the experiments. I

have examined the data using Duncan test of comparison and analysis of variance (ANOVA).

Results and Discussion:

Organic chemists have long studied and tabulated the nature and position of infrared absorptions due to a great variety of chemical bonds in various chemical environments. Infrared absorption ranges are tabulated and so locating these and comparing them to the spectrum in question is straight forward. The two most significant variables in deciding the absorption region of a chemical bond are the bond order and the atomic types of the atoms connected. Conjugation and neighbouring atoms change the frequency slightly. Owing to this characteristic, identical or similar functional groups will have their molecules absorb light at approximately the same frequencies. So the tables of IR absorptions are tabulated by functional group; to be more granular, in some versions the tables are further subdivided. Peak (Wave number cm-[1]), Functional group assignment, Type of Vibration, Bond, Area, Corr. Area and Intensity (667.37, alkyl halides, Stretch, C-Cl, 0.064, and 2.947), (873.75, Alkenes, Bending, =C–H, 0.161, and 2.864), (921.97, Alkenes, Bending, =C–H, 0.006v, and 2.459), (1026.13, alkyl halides, Stretch, C-F, 14.196, and 40.600), (1139.93, alkyl halides, Stretch, C-F, 0.381v, and 6.633), (1234.44, alkyl halides, Stretch, C-F, 0.015, and 2.346), (1317.38, alkyl halides, Stretch, C-F, 0.116, and 3.234), (1379.10, alkyl halides, Stretch, C-F, 0.026, and 3.677), (1415.75, Aromatic, Stretch, C=C, 0.223, and 4.948), (1519.91, Aromatic, Stretch, C=C, 0.065, and 2.191), (1598.99, Aromatic, Stretch, C=C, 0.085, and 3.642), (1740.72, Aldehyde, Stretch, C=O, 0.743 and 2.000), (2852.72, Alkane, Stretch, C-H, 0.244, and 2.649), (2922.16, Alkane, Stretch, C-H, 0.893, and 4.668), (3223.05, Amide, Stretch, N-H, 0.016, and 1.955), (3265.49, Amide, Stretch, N-H, 0.010, and 1.934). A regular infrared spectrum appears to have two different regions. The left side might not contain too many peaks at above 2000 cm -1, but there are some very diagnostic data. The stretching frequencies of Alkane C-H molecules at slightly lower frequencies than 3000 -1 cm point to the presence or absence of saturated carbons and those at frequencies slightly higher than 3000 -1 cm point to the unsaturation. A extremely broad maximum at wavenumbers within 3100 to 3600 -1 cm indicates the presence of exchangeable protons which are usually attached to functional groups like carboxylic acid, amine or alcohol. It is also easy to determine whether there is any alkyne or nitrile functional group present or absent since there are normally no other absorptions in this 28002000 cm-1 range. On the other hand, the second part of the spectrum, less than 2000 cm-1, often contains several peaks of various intensities and most of them are not easily identifiable. In this case, it is easy to identify signals of the carbonyl group (a sharp signal at approximately 1700 cm-1) and the carbon-oxygen bond (one or two intense signals at approximately 1200 cm-1). It is also called the fingerprint region because nearly every organic compound will leave a unique pattern in this complicated lower part. Therefore, a comparison of this region to a normal spectrum is a popular means of demonstrating identity. The O-H stretching absorption peak is large and easily identifiable in an infrared spectrum with respect to alcohols and phenols. Even so we have to understand why this growth is occurring, and approximate estimates of when the peak will not be of this appearance. Start by understanding that any measurable entity of a compound is in fact an enormous collection of discrete molecules, and that each of those molecules may differ with respect to their hydrogen bond. Hence, IR absorptions at various frequencies will be observed in each of these bonds when an IR spectra is recorded. Since it is an average of all these slightly different absorptions, the IR peak will appear broadened [15, 16]. The infrared spectra of chemicals with hydroxyl groups may not show this broad signal. To prevent hydrogen bonding there are two ways; one is to dilute the sample and the other is to record the IR spectra in the gas phase. In concentrated solutions, a larger molecule may sterically inhibit hydrogen bonding and no exchange may occur. The broad O-H peak is replaced in these conditions by an abrupt signal at approximately 3600 cm-1.

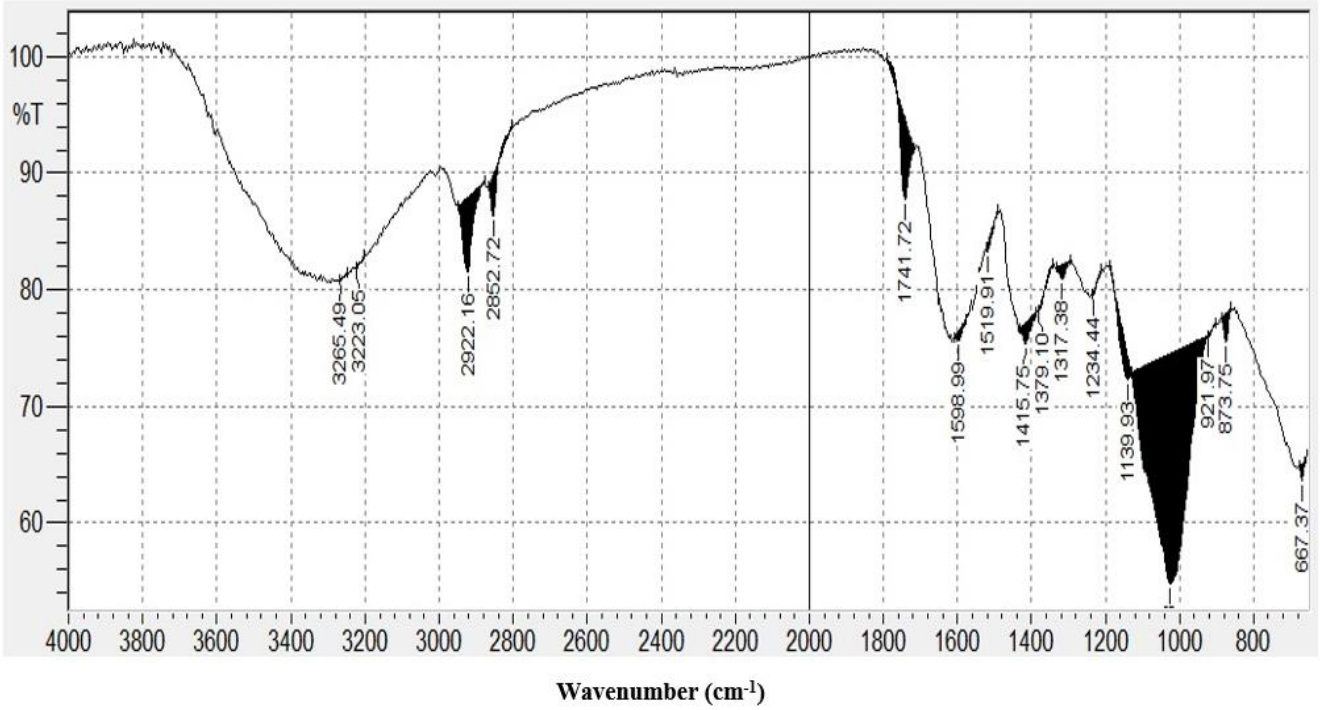

Figure 1. FTIR profile solid analysis of *Capsicum annuum* L.

Table 1. FTIR peak values of solid analysis of *Capsicum annuum* L.

No.	Peak (Wave number cm-1)	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area	Type of Intensity	Bond	Type of Vibration	Functional group assignment	Group frequency
1.	667.37	63.602	1.458	677.01	661.58	2.947	0.064	Strong	C-Cl	Stretch	alkyl halides	600–800
2.	873.75	75.464	2.486	885.33	860.25	2.864	0.161	Strong	=C–H	Bending	Alkenes	650-1000
3.	921.97	76.037	0.115	923.90	902.69	2.459	0.006	Strong	=C–H	Bending	Alkenes	650-1000
4.	1026.13	54.832	19.670	1130.29	923.90	40.600	14.196	Strong	C-F	Stretch	alkyl halides	1000-1400
5.	1139.93	72.360	1.874	1188.15	1132.21	6.633	0.381	Strong	C-F	Stretch	alkyl halides	1000-1400
6.	1234.44	79.518	0.208	1236.37	1211.30	2.346	0.015	Strong	C-F	Stretch	alkyl halides	1000-1400
7.	1317.38	80.927	1.224	1332.81	1296.16	3.234	0.116	Strong	C-F	Stretch	alkyl halides	1000-1400
8.	1379.10	78.194	0.230	1381.03	1342.46	3.677	0.026	Strong	C-F	Stretch	alkyl halides	1000-1400
9.	1415.75	75.384	1.784	1431.18	1388.75	4.948	0.223	Medium	C=C	Stretch	Aromatic	1400-1600
10.	1519.91	83.275	0.580	1521.84	1490.97	2.191	0.065	Medium	C=C	Stretch	Aromatic	1400-1600
11.	1598.99	75.718	0.635	1608.63	1577.77	3.642	0.085	Medium	C=C	Stretch	Aromatic	1400-1600
12.	1740.72	87.747	6.874	1789.94	1718.58	2.000	0.743	Strong	C=O	Stretch	Aldehyde	1720-1740
13.	2852.72	86.395	3.584	2866.22	2802.57	2.649	0.244	Strong	C-H	Stretch	Alkane	2850-3000
14.	2922.16	81.542	6.382	2947.23	2877.79	4.668	0.893	Strong	C-H	Stretch	Alkane	2850-3000
15.	3223.05	81.889	0.120	3224.98	3201.83	1.955	0.016	Bending	N-H	Stretch	Amide	3100-3500
16.	3265.49	80.746	0.173	3269.34	3248.13	1.934	0.010	Bending	N-H	Stretch	Amide	3100-3500

Hypochlorous acid scavenging and Hydroxyl radical scavenging [Antioxidant activity] of pepper (Capsicum annuum L.) Crude (methanolic extract, ethanol fraction, water fraction and ascorbic acid (standard)) recorded (127.00 +/- 5.69, 106.94 +/- 5.10, 98.05 +/- 4.71 and 215.08 +/- 8.09 respectively. The cooking of peppers is an important factor in the composition changes. The nutritional modifications are specific to cultivar because of the genetic variations in the pepper shape and physiology. The Roasting process altered the quantities of capsaicinoid chemicals in the peppers substantially. The variation in the level of capsaicinoids components among cultivars during cooking could be explained by differences in skin thickness and by physiological changes that occur during ripening. Such alterations affect the permeability of heat to the tissues of the fruit, so that boiling, Mexican peppers moderate amount of capsaicin was lost, but grilling enhanced the quantity of the capsaicin components. Cooking peppers involves heat treatment that destroys the cell walls of the peppers and can result in more difficult extraction of the various components. Green and red pepper roasted at the same temperature showed that ascorbic acid was lost to a great extent (between 8 to 80 percent). Ascorbic acid content of pepper, green pea, spinach, pumpkin, and carrot is affected by cooking. Ascorbic acid was degraded in peppers at the rate of 64.71 percent in only 30 minutes. Ascorbic acid is thermally oxidised to dehydroascorbic acid which is hydrolysed to 2, 3-diketogluconic acid. Cooked peppers lose ascorbic acid and other polymeric compounds are formed. The content of ascorbic acid in green and red pepper before and after boiling was different and it was justified by the fact that the thickness of their skins is not the same. The weaker cell membrane may have permitted the rapid leaching of ascorbic acid out of the peppers because of the ease of heat transmission. Phenolic chemicals found in roasted peppers were 1 percent to 106 percent higher in green and red stages than in fresh peppers. The increased percentage of TP in roasted peppers could be also attributed to the enhanced extractability of TP since the biosynthesis of TP does not occur after the peppers are harvested or roasted. Unlike raw peppers, cooked peppers liberate greater quantity of chemicals into the solvents and are rich in phenolic compounds [17] because of disintegration of cells. Heating of sweet peppers and chilli peppers has shown in some research to enhance its phenolic and flavonoid compounds. Microwave, boiling water and steaming increased the concentration of phenolic. It was because of this denaturation of the polyphenol oxidase enzyme by heat that contradictory reports existed that cookery methods did not alter the quantity of phenolic. Although the cooked peppers revealed lower antioxidant activity in the DPPH and ABTS assays, more antioxidant was observed in the cooked peppers due to their enhanced extractability. The synergistic feature of all the antioxidant chemicals is the antioxidant potential which varies with each type of compound. A likely Cooking Method Effect on Antioxidants is an alteration in their chemical property which influences their radical-scavenging capacity.

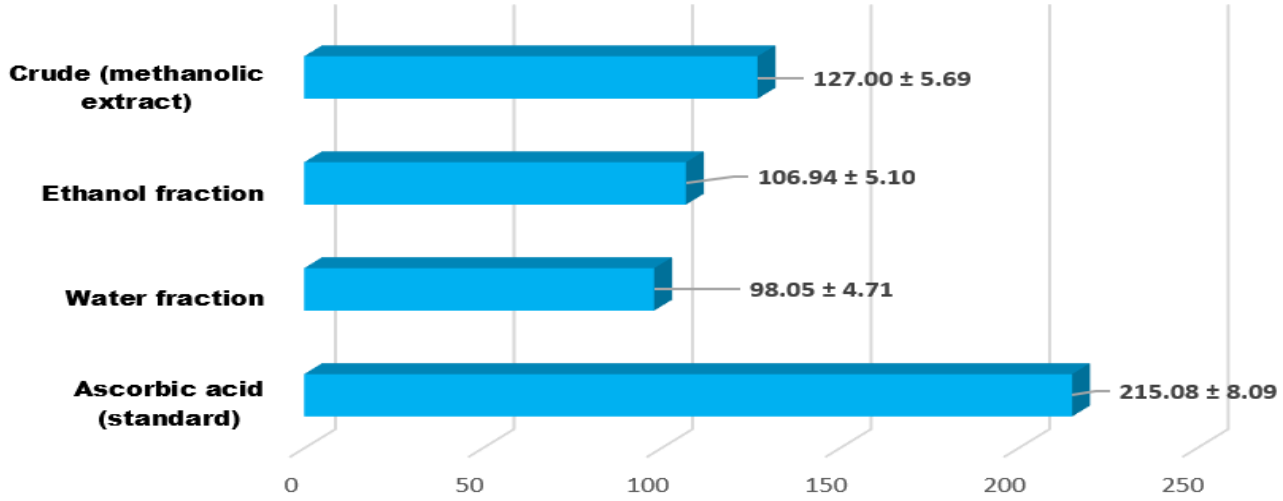

Figure 2. Hypochlorous Acid Scavenging [Antioxidant activity] of pepper (Capsicum annuum L.) Crude (Methanolic extract, Ethanol fraction, Water fraction and Ascorbic acid (standard))

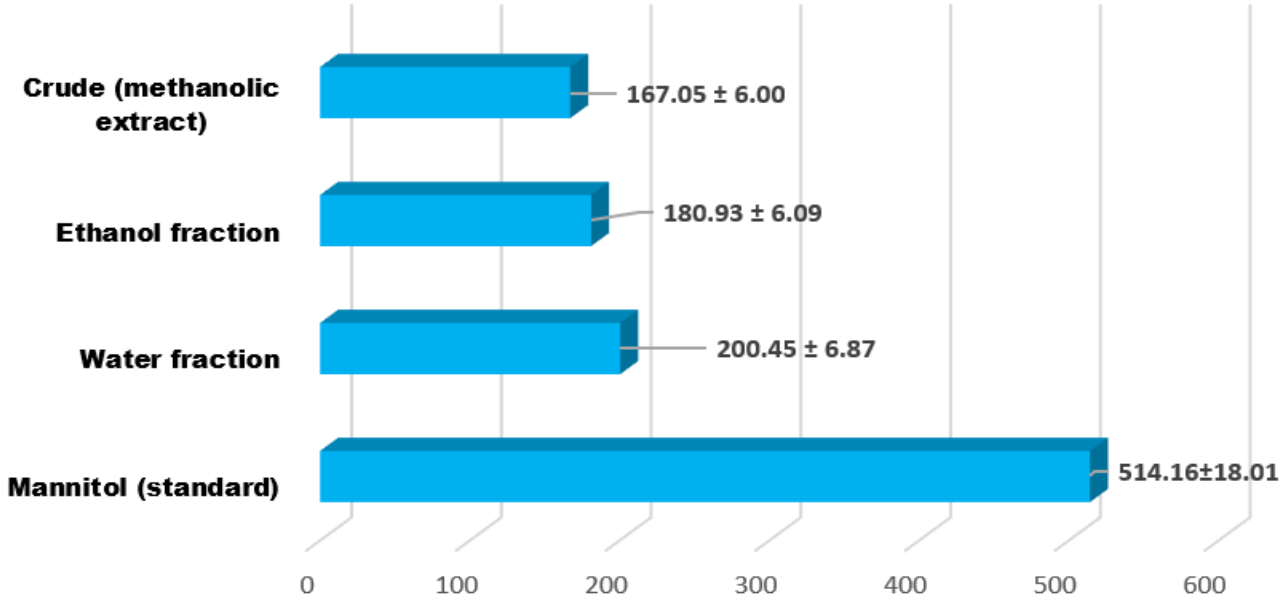

Figure 3. Hydroxyl Radical Scavenging [Antioxidant activity] of pepper (Capsicum annuum L.) Crude (Methanolic extract, Ethanol fraction, Water fraction and Mannitol (standard))

The pepper varietals in this study were selected based on the different levels of spiciness, which is determined on the Scoville scale depending on personal preferences. When grown indoors it is meant to slightly sweet pungent flavour, though just the ripe red stage is consumed. These small fruits are most often eaten in their green, cooked state. When green they are slightly spicy in flavour, but not eaten when red as they are so bitter. Fruits are mainly crimson in color with very pungent smell. An antioxidant effect of phenol and flavonoids on human nutrition and health has been found out. Phenolic chemicals and phenylpropanoids are secondary metabolites produced by plants. Other factors that affect pepper phenolic concentration are ripening time, cultivar, harvest site and year. A series of chemicals known as flavonoids has the molecular formula of C6--C3--C6. In previous pepper flavonoid studies, flavonoid aglycones, e.g., quercetin and luteolin, were identified after acid hydrolysis. Adding antioxidants to your diet will help prevent a raft of health issues, such as cancer, anaemia, diabetes and heart disease [18]. The chemicals reduce the oxidative effect on lipids due to their capacity to scavenge highly reactive oxygen free radicals. These radicals are crucial in oxidative modification of low-density lipoprotein as well as nucleic acid. The most attractive among the various antioxidant mechanisms include vitamins E, C and 8- carotene. Pepper fruits contain a lot of antioxidants. These are rich in carotenoids, xanthophylls, vitamins C and E. Peppers contain sugars and phenolic compounds, whose combination constitutes the quantity of antioxidants. The amount of antioxidants is also specific to a cultivar; e.g. spicy peppers (versus sweet peppers) are in most cases more likely to add antioxidants. Capsaicinoids are the alkaloids present in hot peppers which are specific to the Capsicum species and which possess numerous pharmacological actions. Of course, the best source of antioxidants is fresh pepper, and any technical treatment, freezing, lyophilization, boiling, blanching, etc. presupposes the loss of these components. The rate of destruction of the antioxidant also varies in reaction to alterations within the technological parameters. Freeze-drying is one of the most sophisticated procedures of drying substances. It is a enormous advantage that freeze drying can be performed at low temperatures without destroying the various heat-labile biological molecules.

The conventional methods of investigating the dietary composition of domestic herbivores are microhistological analysis of plant fragments in different samples (oesophageal extrusa, digesta and faeces) and direct measurements of plant biomass. The other option is to watch the animals graze. These monotonous operations require the training of the investigators. The estimated dietary composition is also less accessibleible to inaccuracies caused by factors like; the possibility of varied digestibility of various plant species and high proportion of unidentified fragments.

Conversely, n-alkanes, long-chain alcohols, and long-chain fatty acids which are faecal markers of plant epicuticular wax have been used to determine numerous nutritional parameters, including voluntary intake, diet composition, and digestibility. Among the numerous advantages of these markers is the fact that it has enabled the quantitative analysis of both dietary and faecal samples under the same analysis protocols, which significantly eliminates the chance of analytical error in addition to saving much analytical time. Only that these epicuticular wax constituents vary between plant species is similar between them; in other words, herbivores can employ different plants, or even different morphological parts of the same plant, to figure out what types of food they consume. The principle of their use is simply to compare the marker concentrations in a mixture of oesophageal extrudate, digesta and faeces with the marker concentrations in the ingredients, species or parts of plants that might be in the diet. Two of the largest disadvantages of the process are time and money, which happen to be the largest disadvantages of any chemical analysis as well. These historical procedures of determining n-alkanes and alcohols cannot be utilized in farms where fast decisions relating to feeding management are required. Consequently, sensitive methods of quantifying the concentration of these markers in the excrement and urine of different plant species should be invented [19]. Fourier transform infrared spectroscopy (FTIR) as a tool that mainly works in the near infrared (NIR) region offers a good prospect of studying the chemical and nutritional composition of the diet. Compared to conventional techniques, near-infrared (NIR) analyses are numerous benefits, such as having simpler interpretation, being sample-safe, quicker, and less expensive. To cap it all, they do not generate any wastes and they do not use any reagents. With the method, a broad assortment of constituents can be figured out, such as dry matter digestibility in vitro and intestinal protein digestibility simultaneously. This consists of the level of carbohydrate, protein and fibre among others. Despite these advantages, this technology also has very pronounced drawbacks that include the sheer cost of the equipment needed, time and effort to calibrate and validate, as well as the continuous adjustments that have to be made to the prediction models in order to include new kinds of matrices.

Conclusion:

The nutritional concentration of peppers and their strong antioxidant action is preserved even after roasting. The sensitive Fourier transform infrared (FTIR) technology can be utilized in determining dietary chemical and nutritional contents using a very large number of matrices. Compared to the conventional techniques, the method is user-friendly, accurate, non-hazardous to samples, quick to analyze, and cheap.

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