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Review

Tannins in Foods: Nutritional Implications and Processing Effects of Hydrothermal Techniques on Underutilized Hard-to-Cook Legume Seeds－A Review

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ABSTRACT: Tannins, water-soluble phenolic compounds, have been reported to have the ability to form complexes with nutritionally important nutrients such as protein and mineral elements thereby making them unavailable for absorption and utilization. Toxicity of tannin has been demonstrated in experimental animals although no deleterious effect of ingestion of legume tannin on human physiology has been reported. This report highlights the processing effects of soaking and hydrothermal techniques on some underutilised hard-to-cook legume crops and the importance of tannin in legume nutrition. Soaking and hydrothermal processing reduce the tannin content of processed legume seeds and hence improve the availability of protein and mineral elements. In view of the recent findings of the health benefits, classification of tannin which is traditionally regarded as an antinutrient should be reconsidered. Provision of these information will enhance knowledge of legume nutrition and economic utility. Increasing consumption of underutilized nutritionally important legume seeds, it is hoped, will alleviate the problem of protein energy malnutrition in many developing nations.

Keywords: hydrothermal techniques, legume seeds, nutritional implication, tannin

INTRODUCTION

In their natural form, tannins are water-soluble phenolic compounds with molecular weights in the range of 300 to 500 and have the ability to precipitate gelatin, alkaloids, and proteins (Mena et al., 2015). Tannins are secondary metabolites widely distributed in plants: they are polymeric phenolic substances with astringency properties (Agrawal et al., 2012; Lattanzio et al., 2012; Bone and Mills, 2013; Tamokou et al., 2017). Humans consume a number of foods containing considerable amounts of dietary tannins. Tannins are found in a huge variety of plants, including legume seeds, cider, cereals, cacao, peas, some leafy and green vegetables, coffee, tea, and nuts (Lochab et al., 2014; Suvanto et al., 2017; Fraga-Corral et al., 2020). In general, tannins can be classified into one of two classes on the basis of their structural features: hydrolyzable and condensed tannins (Hatew et al., 2016; Bee et al., 2017). In addition to other antinutritional components, such as phytic acid, hemagglutinin, saponin, goitrogen, and trypsin inhibitors, tannins are present in legume seeds at varying concentrations. These antinutritional components interfere with the digestive process and prevent the efficient utilization of nutrients (Luo and Xie, 2013; Sathya and Siddhuraju, 2015; Wu et al., 2016).

Legumes are the good source of nutritionally important nutrients such as mineral elements and dietary protein (Ojo et al., 2016; Ojo et al., 2017a; Ojo et al., 2018). Proteins from plants sources are preferable to animal sources because, unlike animal proteins, plant proteins are not usually associated with the occurrence of cardiovascular diseases. Legume seeds are good alternatives to animal protein. In recent years, there has been an increase in demand and consumption for plant protein, which has stimulated the search for more protein-rich plants (Adebowale et al., 2005; Ojo, 2015; Septiana and Analuddin, 2019). However, overdependence on common legumes has resulted in a sharp increase in prices (Ojo, 2015; Ojo et al., 2017a). Many legumes remain underutilized in South-West Nigeria, where they are planted mainly for subsistence purposes. Some of these underutilized legumes include *Cajanus cajan*, *Cassia hirsuta*, *Canavalia ensiformis*, *Mallotus subulatus*, *Vigna racemosa*, *Sphenostylis sterocarpa*, and *Vigna subterranean*. In many areas of the world, particularly in Africa, many underexploited legumes are regarded as security crops for fallow farmland in prepara-

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tion for a new planning season (Nwosu, 2013; George et al., 2020). They are also cultivated as alternative food crops that are consumed during dry seasons, when other food crops are out of stock.

The phenomenon of prolonged cooking times and the presence of antinutritional components are prominent challenges facing the use of these underutilized legumes. The problem of prolonged cooking in legume seeds was reported to be alleviated by hydrothermal processing techniques (Ojo, 2018a). In this study, the nutritional implications of tannins in foods, as well as the effects of soaking at varying hydration levels followed by hydrothermal processing, are reported. Information on the effect of hydrothermal techniques on the tannin content of underutilized legume species before and after processing will enhance the current knowledge of legume processing and nutrition and foster economic utility.

HEALTH IMPLICATION OF TANNINS IN DIETS

Traditionally, tannins are considered to have antinutritional properties (Ojo, 2018b). However, recent evidence has shown that the consumption of tannins can have health benefits. The effects of tannin on human and animal biology vary considerably and depend on the composition of the diet and dietary patterns. Tannins have the ability to form complexes with carbohydrates, proteins, and certain mineral ions in foods (Kunyanga et al., 2011). The formation of such complexes depends on a requirement for suitable conditions such as pH, temperature, and concentration. Tannins have greater tendency to form complexes with proteins than carbohydrates and other food polymers because of the strong hydrogen bond affinity of the carboxyl oxygen of the peptide group. Complexes formed by tannins and proteins have been reported to be responsible for growth depression, low protein digestibility, decreased availability of amino acid, and increased fecal nitrogen (Waghorn, 2008; Woodward et al., 2009; Dijkstra et al., 2013; Grosse Brinkhaus et al., 2016).

Although the consumption of legume seeds has been reported to confer health benefits, a number of reports have described the harmful effects of tannic acid on the gastrointestinal tissues (Maphosa and Jideani, 2017; George et al., 2020). The toxicity of orally ingested tannins is relatively low: the rectal toxicity of tannic acid is approximately twice its oral toxicity (George et al., 2020; Hassan et al., 2020). The ingestion of tannic acid has been reported to cause hardening of the gastrointestinal mucosa, which results in a reduction in the gastrointestinal absorption of nutrients. Sorghum, which contains tannins, has been implicated in the occurrence of esophageal cancer (Wexler, 2014). It is important to consider that most of the toxicity studies of tannins in experimental animals are conducted using commercially available samples of tannins or tannic acid. There was no evidence of toxicity in sheep and cattle fed diets containing tannin (Wen et al., 2002). It is therefore extremely difficult to extrapolate if tannins in legume seeds used in feeds will have similar toxicities upon ingestion. There are conflicting reports. Recently, tannins have been reported to be useful in the control of zoonotic pathogens, such as *Salmonella*, in monogastric animals (Hassan et al., 2020). There have been contrasting reports about the relationship between condensed tannins and bioactivities in animals. It has been reported that condensed tannins from forage legumes fed to ruminants resulted in improved growth, milk production, and fertility, and reduced methane emissions and ammonia volatilization from the lung and the urine (Mueller-Harvey et al., 2019). Forage legumes containing condensed tannins have been reported to be beneficial in combating the effect of gastrointestinal parasitic nematodes (Hoste et al., 2012; Mueller-Harvey et al., 2019). Tannins bind excess plant proteins in ruminant, causing them to precipitate them out of the rumen and thus preventing the creation of the stable form that is characteristic of pasture bloat (MacAdam and Villalba, 2015).

Conversely, tannic acid used to be administered for the treatment of diarrhea and the dressing of skin burn. A precipitant of proteins affecting the colloidal stability of beer, tannic acid removes some metals (such as Al, Zn, Pb, and Fe) and some polyphenols when they are bound to proteins. Tannic acid has been reported to be effective against staling, flavor, and light instability (Wexler, 2014). It has been reported that tannins in *Lotus corniculatus* seeds bind excess plant proteins in the rumen of cattle and sheep, preventing bloating. Therefore, it can be included as 50% or more in the forage mixture without the risk of bloating. Moreover, tannins in *L. corniculatus* seeds have been reported to reduce the enteric methane production of dairy cows (Woodward et al., 2004). As a result of the low pH in the abomasum, tannins release proteins, which are then digested and absorbed in the small intestine (Wen et al., 2002; MacAdam and Villalba, 2015). In humans, no evidence of toxicity arising from the consumption of excessive amounts of legume tannins has ever been reported. Excess tannins may have been attributed to the increased incidence of tumorigenic diseases or carcinogenesis in some areas of the world; however, the minimum amount of tannins in the human diet that can cause physiological disorder is not known. There is a dearth of information on the effects of legume tannins on the incidence of carcinogenesis. The acetonic extract of some processed legumes, such as field bean and pigeon pea, have been reported to have antioxidant and antidiabetic properties (Kunyanga et al., 2011). Polyphenols, including tannins, have been reported to be some of the major antioxidants in our daily diets (Montes-Ávila et al., 2018). Legume tannins are astringent and decrease the palatability of forage, leading to low feed intake, poor nutrient digestibility, and poor production performance in livestock. The growth suppression and protein digestibility effects of bean tannin may be related to their astringent taste, which decreases feed consumption, and their ability to form stable and insoluble complexes with dietary proteins (Bone and Mills, 2013; Wang et al., 2015; Tamokou et al., 2017; Gutierrez and Torres, 2019). Moreover, tannic acid has non-food uses in the manufacture of rubber and inks, and as a fixative of dyes.

INFLUENCE OF SOAKING ON TANNIN CONTENT OF LEGUME SEEDS

The seeds of some underutilized hard-to-cook legumes were soaked in distilled water at various hydration levels (0, 10, 25, 50, 75, and 100%) for a period of 24 h at an ambient temperature of 23°C to 28°C (Ojo, 2015; Ojo et al., 2017a; Ojo et al., 2018). The changes in the concentrations of tannins in the seeds before and after soaking were determined (Table 1). For each of the legume seeds, there was a significant difference ($P<0.05$) in the concentration of tannins at the different hydration levels. The lowest percentage reduction in tannin content after soaking (0.61%) was observed for *C. hirsuta* at the 10% hydration level. For *V. racemosa*, percentage reduction of 3.21% was recorded at 10% hydration level while 15.53% was recorded for both 75% and 100% hydration levels. The raw sample of *S. sterocarpa* contained 39.88 mg/g tannin. This quantity was reduced by 4.11, 7.07, 13.87, and 14.29% when the seed was soaked at the 10, 25, 50, and 100% hydration levels, respectively. As presented in Table 1, the percentage reduction for other legumes was in the range of 3.81% to 14.52% for *M. subulatus* (white species), 0.61% to 8.73% for *C. hirsuta*, and 3.53% to 13.79% for *C. ensiformis* (Ojo et al., 2017a; Ojo et al., 2018; Ojo, 2018b). In a study on kidney beans, soaking, boiling, and pressure cooking yielded a significant decrease in tannin content (Khattab and Arntfield, 2009).

The percentage reduction in tannin content of the legumes studied was similar to, but comparatively lower than, those of a previous report on *Mucuna flagellipes*, which ranged from 58.4% at 6 h of soaking to 74.9% at 24 h of soaking (Udensi et al., 2008). The smaller reduction might be due to varietal differences. The reduction in tannin content of some beans by soaking in different solutions has also been reported (Vijayakumari et al., 2007). Pinto beans with a high tannin content exhibited the greatest reduction in tannin content after soaking, whereas cranberry beans with low tannin content lost less tannins during soaking.

The reduction in the tannin content may be due to leaching of the polyphenols into the soaking water (Wu et al., 2016; Ojo et al., 2018). Tannins are polyphenols and polyphenolic compounds are mostly water-soluble in nature and mostly located in the seed coat. Therefore, it can be inferred that a pre-processing method, such as soaking, can be used to reduce the level of some water-soluble/leachable antinutritional factors, such as tannins (Ojo et al., 2017a; Ojo et al., 2018; Ojo, 2018b).

Table 1. Concentration of tannins in legume seeds before and after soaking at various hydration levels (unit: mg/g)

Legume sample	Hydration level					
	0	10	25	50	75	100
Mallotus subulatus 1 (white variety)	28.10±1.22^f	27.03±0.10^e (3.81)	26.07±0.14^d (7.22)	25.32±0.21^c (9.89)	24.28±0.07^b (13.59)	24.02±0.11^a (14.52)
Cassia hirsuta	62.31±0.37^f	61.93±0.51^e (0.61)	61.37±0.42^d (1.51)	60.17±0.22^c (3.43)	59.48±0.60^b (4.54)	56.87±0.50^a (8.73)
Canavalia ensiformis	27.47±0.44^f	26.50±0.12^e (3.53)	26.37±0.21^d (4.00)	26.12±0.03^c (4.91)	24.70±0.10^b (10.08)	23.68±0.11^a (13.79)
Vigna subterranean 1 (mottled colored)	31.58±0.31^f	30.20±0.13^e (4.36)	28.88±0.24^d (8.55)	26.72±0.17^c (15.39)	25.87±0.25^b (18.08)	25.41±0.10^a (19.54)
Vigna racemosa	28.97±0.32^e	28.04±0.10^d (3.21)	27.16±0.02^c (6.25)	26.55±0.13^b (8.35)	24.47±0.24^a (15.53)	24.47±0.24^a (15.53)
Mallotus subulatus 2 (brown variety)	38.87±0.41^f	36.42±0.21^e (6.30)	35.26±0.32^d (9.29)	34.87±0.24^c (10.29)	34.23±0.10^b (11.94)	33.08±0.12^a (14.89)
Vigna subterranean 2 (cream colored)	42.59±0.53^e	40.08±0.12^d (5.89)	38.14±0.10^c (10.45)	37.52±0.30^b (11.90)	37.34±0.21^a (12.33)	37.34±0.21^a (12.33)
Sphenostylis sterocarpa	39.88±0.24^e	38.24±0.21^d (4.11)	37.06±0.10^c (7.07)	34.35±0.22^b (13.87)	34.18±0.30^a (14.29)	34.18±0.20^a (14.29)

Values are presented as mean±SD (n=3) on dry basis and values in parentheses represent the percentage loss.
Different letters (a-f) in the same row indicate significant differences ($P<0.05$).
Data from the article of Ojo (2015), Ojo et al. (2017a), and Ojo et al. (2018).

IMPACT OF HYDROTHERMAL TECHNIQUES ON TANNIN CONTENT OF UNDERUTILIZED LEGUME SEEDS

Four hydrothermal techniques [atmospheric boiling (AB), atmospheric steaming (AS), pressure boiling (PB), and pressure steaming] were employed to process the seeds of some underutilized hard-to-cook legume seeds (Ojo, 2015; Ojo et al., 2017a; Ojo et al., 2018). The effects of hydrothermal processing techniques on the levels of tannins in the seeds are presented in Table 2. The hydrothermal techniques caused a reduction in the concentration of tannins in the legume seeds. All the hydrothermal processing methods (i.e., boiling and steaming at atmospheric pressure, as well as boiling and steaming at elevated pressure) had significant effects ($P<0.05$) at varying percentages on the level of tannins in the legume seeds.

The lowest value of tannins (6.85 mg/g) was recorded for the white variety of *M. subulatus*, which was boiled at normal atmospheric pressure, whereas the highest value (8.15 mg/g) was recorded after steaming at elevated pressure. The same percentage reduction, of 78.46%, was observed for *C. hirsuta* using conventional boiling and PB (Ojo et al., 2018). For *C. ensiformis*, the two processing methods, i.e., AB and AS, resulted in the reduction in tannins of 64.69% and 61.41%, respectively, whereas boiling and steaming at high pressure resulted in a reduction of 63.49% and 61.27%, respectively (Ojo et al., 2018). For the other legumes investigated, the mottled colored variety *V. subterranean*, *V. racemosa*, *M. subulatus*, cream-colored variety *V. subterranean*, and *S. sterocarpa*, there were similarities in the trend in tannin reduction when each of the four hydrothermal processing techniques was used (Ojo, 2015; Ojo et al., 2017a; Ojo et al., 2017b).

As recorded in Table 2, pressure processing, both boiling and steaming, resulted in relatively smaller losses of tannin than the atmospheric processing of boiling and steaming. This observation is true for all the legumes studied, except for *C. hirsuta*, in which the percentage loss of 78.46% was reported for both boiling processing methods (Ojo, 2018b). Pressure processing causes relatively lower losses because of the shorter processing times. These results agree with the study of Xu and Chang (2009) on green pea, yellow pea, and chickpea. In 2016, Fabbri and Crosby also reported that cooking improved the digestibility of beans that contained high concentrations of antinutrients, including tannins. In another study on asparagus beans (*Vigna sesquipedalis*), the tannin content was reduced by 49% after boiling (Nzewi and Cemaluk, 2011). The tannin content of varieties of *Phaseolus vulgaris* in Tanzania was investigated after conventional cooking and the percentage of tannin reduction was in the range of 20% to 81% (Mamiro et al., 2017). Percentage reductions of 65.8, 65.8, 74.3, and 74.3% were recorded after regular boiling of *M. flagellipes* for 30, 45, 60, and 90 min, respectively (Udensi et al., 2008). Similarly, the thermal treatment of *C. cajan* seeds resulted in a 47.06% reduction in tannins (Iorgyer et al., 2009).

Table 2. Tannin content of legume seeds as influenced by hydrothermal processing techniques (unit: mg/g)

Legume sample	Processing condition				
	Raw dried sample	Atmospheric boiling	Atmospheric steaming	Pressure boiling	Pressure steaming
Mallotus subulatus 1 (white variety)	28.10±0.22^{e}	6.85±0.32^{a} (75.62)	7.84±0.40^{c} (72.10)	7.57±0.05^{b} (73.06)	8.15±0.13^{c} (70.99)
Cassia hirsuta	62.31±0.37^{d}	13.42±0.33^{a} (78.46)	15.38±0.32^{b} (75.32)	13.42±0.61^{a} (78.46)	16.67±0.80^{c} (73.25)
Canavalia ensiformis	27.47±0.44^{d}	9.70±0.41^{a} (64.69)	10.60±0.34^{c} (61.41)	10.03±0.20^{b} (63.49)	10.64±0.32^{c} (61.27)
Vignasubterranean 1 (mottled colored)	31.58±0.31^{d}	11.49±0.24^{a} (63.62)	12.82±0.31^{b} (59.40)	12.31±0.57^{b} (61.02)	13.19±0.60^{c} (58.23)
Vigna racemosa	28.97±0.32^{c}	9.02±0.13^{a} (68.86)	10.15±0.21^{b} (64.96)	9.38±0.34^{a} (67.62)	10.13±0.42^{c} (65.03)
Mallotus subulatus 2 (brown variety)	38.87±0.41^{c}	9.20±0.18^{a} (76.33)	10.74±0.52^{b} (72.37)	9.61±0.29^{a} (75.28)	10.77±0.61^{b} (72.29)
Vigna subterranean 2 (cream colored)	42.59±0.53	14.57±0.57^{a} (65.79)	16.13±0.60^{b} (62.13)	15.20±0.70^{a} (64.34)	16.16±0.42^{b} (62.06)
Sphenostylis sterocarpa	39.88±0.24^{d}	10.07±0.15^{a} (74.75)	11.76±0.23^{b} (70.51)	11.01±0.40^{b} (72.39)	11.93±0.37^{c} (70.09)

Values are the mean±standard deviation (n=3) on dry basis and values in parentheses represent the percentage loss.
Different letters (a-e) in the same row represent significant differences ($P<0.05$).
Data from the article of Ojo (2015), Ojo et al. (2017a), and Ojo et al. (2018).

CONCLUSION

The processing of these underutilized legume seeds must be performed such that the tannins will be reduced to a safe level/concentration. Although there is no standardized safe limit for legume tannins, it is well established that tannins form complexes with nutritionally important compounds, such as proteins and some mineral elements, making them unavailable for absorption. Soaking and hydrothermal techniques reduce tannin concentrations in legume seeds and hence improve the bioavailability of protein and mineral elements. Although the toxicity of synthetic/commercially available tannins has been demonstrated in experimental animals, legume seed tannins have not been reported to cause any known toxicities or physiological disorders upon ingestion. Considering the recent evidence showing the health benefits, the classification of tannins that have been hitherto regarded as an antinutrient should be reconsidered. Therefore, the provision of information on the importance of tannins in legume nutrition is anticipated to foster economic utility, encourage cultivation, and prevent the imminent extinction of these lesser known legume food crops. Increasing the consumption of nutritionally important underutilized legume seeds will alleviate the problem of protein energy malnutrition in developing nations.

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AUTHOR DISCLOSURE STATEMENT

The author declares no conflicts of interest.

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