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The Effect of Drying and Salting on the Nutrient Composition and Organoleptic Properties of *Vernonia amygdalina* Leaves

Fred Omon Joseph Oboh* and Gift Ejehiokhin Madojemu
Department of Basic Sciences, Benson Idahosa University, PMB 1100, Benin City, Nigeria

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Abstract. The preservation methods used for finding their effect on the nutrient content and organoleptic properties of *Vernonia amygdalina* leaves (bitter leaf), included oven drying at 40 °C without any pretreatment, blanching in steam prior to oven drying at 40 °C, light brining (25 g salt/L water), light dry salt treatment (25 g dry salt/kg leaves), light brine and vinegar treatment (50 g salt + 50 mL vinegar), and heavy salting (250 g/kg leaves), each for a duration of 14 days. Compared with drying alone, blanching before drying did not affect vitamin C, β-carotene, total carotene and ash content, but resulted in decrease of iron, sodium and calcium. Relative to the fresh vegetable, fermentation resulted in a decrease in the content of all the nutrients investigated except sodium and calcium which increased.

Keywords: *Vernonia amygdalina* leaves, steam blanching, drying, salting, nutrient composition

Introduction

Drying and fermentation are old and reliable food preservation methods. Pretreatment of leafy vegetables by blanching or scalding in hot water or steam makes the product tender, limits discolouration, eliminates intracellular gases responsible for oxidation reactions, kills harmful bacteria and deactivates enzymes which disintegrate the tissues (DiPersio *et al*., 2007; Kendall *et al*., 2004). Blanching and other thermal treatment of green leaves can also enhance the bioavailability of micronutrients by destroying antinutrients and by releasing the micronutrients entrapped in the plant matrix (Yang and Tson, 2006; Yadav and Sehgal, 2002). Thin-slicing facilitates better heat and chemical penetration, and water loss during drying, resulting in shorter drying time and better preservation of heat-labile nutrients.

Salting or brining of vegetables offers tremendous possibilities both for their commercial and home preservation. In the process, the salt exerts a selective action on the naturally occurring organisms to promote a desirable fermentation. Salt tolerant microorganisms use, as their nutritive material, the soluble constituents that diffuse out of the vegetable as a result of the action of the salt on vegetable tissue. These fermentative organisms bring about the production of various compounds, principally lactic acid but also acetic acid (both of which result in reduction of pH), alcohols and considerable amounts of gas. Production of sufficient amount of acid makes the medium unsuitable for the growth of food spoilage bacteria. In addition, acid and other microbial metabolites alter the flavour of the food. Substances and organisms in fermented foods can cause changes in the composition and/or activity of the gastrointestinal microbiota resulting in several health benefits (FAO, 2007; Perdigon *et al*., 1987).

Vernonia amygdalina, variously known as bitter leaf (English), oriwo (Edo), ewuro (Yoruba), shikawa (Hausa), and olubu (Igbo), is a tropical shrub, 1-3 m in height (Igile *et al*., 1995). The leaves are dark green with characteristic odour and bitter taste. The species is indigenous to tropical Africa, where it is found wild or cultivated. The leaves are eaten after crushing and washing thoroughly to remove the bitterness (Mayhew and Penny, 1988). As with other high yielding leafy vegetables, post-harvest losses may occur due to inadequate preservation.

In this study, three preservation techniques–drying without pretreatment, blanching in steam followed by drying, and salting–were employed for the preservation of *Vernonia amygdalina* leaves. The effects of the treatments on the nutrient composition and organoleptic characteristics of the vegetable were then investigated.

Materials and Methods

The leaves of *Vernonia amygdalina* were subjected to the following treatments:

Preparation and steam blanching of leaves. A known weight of thin slices of *V. amygdalina* leaves was wrapped in clean cheese cloth, tied with a string and put on the mesh in the water bath. Care was taken not to have the cloth immersed in water but deep enough to be engulfed by the steam. The lid of the water bath was replaced and the samples left to steam for 2 min.

*Author for correspondence; E-mail: fredoboh67@yahoo.co.uk

Drying of leaves. A known weight of thin-sliced *V. amygdalina* leaves was spread in a thin layer on shelves in a ventilated oven and temperature was maintained at 40 °C till the leaves were dry, hard and brittle.

Heavy salting. *V. amygdalina* leaves were gently washed to remove dirt, and the water was drained. Salt (37.5 g) and leaves (150 g) were mixed well and filled in a plastic bucket. The mixture was covered with 2 layers of muslin cloth; a pressure plate and weight were placed on it. Brine made of salt (37.5 g) and water (150 mL) was added until the pressure plate was slightly submerged. The buckets were stored in a cool, dry and shaded place for two weeks (James and Kuipers, 2003). Ambient temperature was 26.5-27.0 °C.

Light salting. ***Dry salt method.*** Washed and drained *V. amygdalina* leaves (150 g) were mixed well with dry salt (3.75 g) and filled in a plastic bucket, packing tightly. The mixture was covered with 2 layers of muslin cloth; a pressure plate and weight were placed on it. The set up was left for about 24 h until the salt drew out enough liquid from the vegetables. The bucket was stored in a cool, dry and shaded place for two weeks (James and Kuipers, 2003). Ambient temperature was 26.5-27.0 °C.

Brine method. Light brine (3.75 g salt dissolved in 150 mL water) was added to washed and drained *V. amygdalina* leaves (150 g) in layers alternately in a plastic bucket, packing tightly. The mixture was covered with 2 layers of muslin cloth; a pressure plate and weight were placed on it. The bucket was stored in a shaded part of the laboratory by the windows for two weeks (James and Kuipers, 2003). Ambient temperature was 26.5-27.0 °C.

Light salting + vinegar. Salt (7.5 g), vinegar (7.5 mL) and water (150 mL) were mixed and added to washed and drained *V. amygdalina* leaves (150 g) in layers alternately in a plastic bucket, packing tightly. The mixture was covered with 2 layers of muslin cloth; a pressure plate and weight were placed on it. The set up was stored in a shaded part of the laboratory by the windows for two weeks. Ambient temperature was 26.5-27.0 °C.

Analytical procedure. Moisture, ash and crude fibre content were determined according to AOAC (1984). pH of the deionized water extract of the vegetable was determined at an ambient temperature of 27 °C using a previously calibrated pH meter. Mineral content of the sample was determined using atomic absorption spectrophotometery for calcium and iron content; sodium ion content was determined by flame photometry (Okalebo, 1985; Novosamsky *et al*., 1983).

For the determination of β-carotene content, leaves were extracted with dimethyl sulphoxide and methanol (Minocha *et al*., 2009). Beta-carotene was determined by spectrophotometry at 436 nm after methanolic KOH saponification and extraction with heptane (Rodriguez-Amaya and Kimura, 2004).

Total carotenoid content was determined by spectrophotometry at 460 nm after KOH saponification of the methanol extract and extraction with diethyl ether. (Rodriguez-Amaya and Kimura, 2004).

Ascorbic acid of the leaf sample was determined according to the procedure given by Roe and Kuether (1943) using the absorbance of solution at 520 nm.

All determinations were done in triplicate.

Results and Discussion

Effect of treatments on organoleptic characteristics of *Vernonia amygdalina* leaves. The organoleptic characteristics of *V. amygdalina* subjected to various fermentation treatments are presented in Table 1.

The leaves of the fresh sample, were bright dark green, had a very bitter taste, a distinctive green leafy smell and were firm and slightly coarse. Preservation in light brine gave a dirty dark green coloured product, similar in taste and smell to the Nigerian native condiment *eru* prepared by the fermentation of *Citrulus lanatus* seeds (egusi). The product was slightly less firm than fresh leaves. Leaves were coarser to feel than the fresh sample. Light dry salting imparted to the leaves characteristics similar to those of the light brining product except that the leaves maintained more of the initial structure of fresh leaves. Preservation in light brine and vinegar yielded product having characteristics similar to the product of light brining except that the leaves felt smoother to touch. Also the structure of the leaves was better maintained than that of the products yielded by light salting. Heavy salting gave a highly bitter and very salty product that was slightly flaccid as compared with the fresh leaves. It was still coarse but was not as firm as the fresh produce.

From our results, salting, alone or with vinegar, apart from preserving and debittering the leaves also gave a product which had a flavour similar to that of *eru,* a Nigerian condiment. This suggests that treatment resulted in the breakdown of protein as this occurs in the preparation of this condiment from high protein content oilseed.

Fermented vegetables are preferred to the dried vegetables because they are succulent; due to their higher moisture

Table 1. Characteristics of fresh and fermented *V. amygdalina* leaves

Characteristics	Fresh	Treatments: Light salting (light brine)	Light salting (dry salt)	Light salting + Vinegar	Heavy salting
Colour	Dark Green	Darker shade of dirty dark green	Darker shade of dirty dark green	Lighter shade of dirty dark green	Dark green
Taste	Very bitter	Harsh bitterness gone; tastes like *eru*, a Nigerian condiment	Harsh bitterness gone; tastes like *eru*	Harsh bitterness gone; tastes slightly different from the product from salt treatment alone	Very bitter and salty
Odour	Fresh green leafy smell	Slight but not offensive; smells like *eru*	Slight but not offensive; smells like *eru*	Slight but not offensive; smells like *eru*	Fresh green leafy smell
Appearance and texture	Firm and slightly coarse	Slightly less firm than fresh leaves; slightly more coarse than fresh leaves	Slightly more firm than product of light brining; less coarse than fresh leaves	Retained more of the original structure of the fresh leaves than products from salt treatment alone; smooth feel	Similar to fresh leaves; slightly coarse

content they do not look dry and shrunken. Fermentation imparts better colour and flavour to the products (Battock and Ali, 1998).

Effect of treatments on pH of *Vernonia amygdalina* leaves. The acidity of foods is very important because it influences the kind of spoilage that can occur and hence the way that they are processed. Most bacteria grow readily at a pH near 7, when a food is neutral (for example at pH of 6.89, the value for the fresh leaves). On the other hand, they are more easily inhibited at lower pH of about 4 to 6 (Rose, 1976; Pyke, 1964). Low acid foods have a pH greater than 4.6 and a water activity (a_w) greater than 0.85 (Schmidt, 1983). Fig. 1 shows the pH values of *V. amygdalina* leaves subjected to different fermentation process.

V. amygdalina leaves showed a decrease in pH from 6.89 for the fresh sample to 4.30, 5.00, 5.36 and 4.50 for products of heavy salting, light brining, light salting, and light brining+ vinegar treatments, respectively. Thus the treatments resulted in fermentation of leaves by acid forming organisms, giving in each case, a product with decreased susceptibility to microbial spoilage. Fresh *V.amygdalina* leaves and those preserved by light brining and light salting, with pH values of 5.0 and 5.36, respectively, can be classified as low acid foods. Such foods are susceptible to spoilage by microorganisms, such as heat resistant, mesophilic spore forming anaerobic bacteria, *Clostridium botulinum,* (which produces a lethal water-soluble toxin) and the putrefactive anaerobe No. 3679, a *Clostridium sporogenes* type organism (Shmidt, 1983). These microbes do not grow in high acid foods, such as the products of heavy

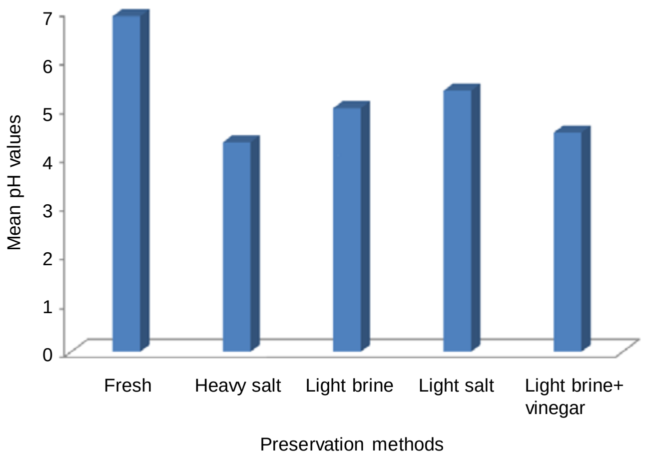

Fig. 1. pH values of fresh and fermented *V. amygdalina* leaves.

salting (pH 4.30) and light brining and vinegar treatment (pH 4.50) and therefore, should, not be a problem in these products. At low pH (i.e., high acidity) the vegetable would be preserved through prevention of the growth of these and other microorganisms that cause spoilage and food poisoning.

Effect of treatments on the nutrient composition of *Vernonia amygdalina* leaves. The β-carotene content (dry basis) of fresh leaves (0.326 ± 0.037 mg/g) was not significantly different from that of the blanched and oven dried vegetable (0.334 ± 0.036 mg/g), but was higher than the products of other treatments (Table 2). Oven drying, heavy salting, light brining, light salting and light brine and vinegar treatment resulted in heavy loss of β-carotene with retention values of only 21.78, 4.29, 9.51, 11.35 and 2.15%, respectively (Table 3). Total carotene content of fresh vegetable (1.403 ± 0.050 mg/g) was highly retained in the oven-dried and the blanched and oven-dried products (76.76 % and 98.65%, respectively). Preservation in salt resulted in serious losses (retention varied from only 1.21% for light brine and vinegar treatment to 4.13%, for light brine and light salt treatments).

Table 2 shows beta-carotene, total carotene, vitamin C, moisture and crude fibre content of fresh, dried and fermented *V. amygdalina* leaves.

Blanching with steam prior to oven drying appears to have had a protective effect on β-carotene compared with drying alone. The loss of this provitamin was, therefore, probably not due to heat denaturation but as a result of enzymatic degradation. This would explain the higher loss in the unblanched material, in which enzymes had not been inactivated by any heat treatment prior to drying. The lower value recorded for the oven dried product may also be the result of better extractability of lipids from the blanched and dried product during the determination of this compound.

Vitamin C content (dry basis) was higher in fresh leaves than in products formed from drying or fermentation. However, this vitamin was highly retained in the dried products (75.30% for oven dried and 72.81% for the blanched and oven dried products), but highly reduced in the salt-preserved products (retention was only 0.25-2.64%) (Table 3). Vitamin C is heat-labile and the decrease in the content of this substance on drying may have been due to enzymatic action during the preparation of the material for preservation and/or its destruction at the drying temperature. The severe loss of vitamin C, a water soluble vitamin, during salting was probably due to leaching into the brine and/or as a result of destruction by microbial enzyme(s).

Crude fibre showed a similar trend, the fresh samples had the highest value. Preservation in salt and in salt + vinegar resulted in a decrease in crude fibre content; only about half (46.27-52.12%) of that found in the fresh leaves was retained in the preserved products (Table 3).

V. amygdalina leaves had a moisture content of 81.00 ± 1.00%. Moisture content of oven-dried vegetable was not measured. However, oven-drying proceeded to the desired extent as it removed most of the moisture, making the product crumbly.

Table 2. Carotene, vitamin C, moisture and fibre content of *V. amygdalina* leaves subjected to different preservation treatments

Bitter leaf	β-carotene (mg/g)	Total carotene (mg/g)	Vitamin C (mg/100 g)	Moisture (%)	Crude fibre (mg/g)
Fresh	O.326 ± 0.037^{c}	1.403 ± 0.050^{b}	2131.4 ± 8.370	81.00 ± 1.00^{c}	6.44± 0.04^{b}
Oven dried	0.071 ± 0.006^{a,b}	1.077 ± 0.017^{a,c}	1604.9 ± 72.610^{a,c}	-	-
Blanched and oven dried	0.334 ± 0.036^{c}	1.384 ± 0.110 ^{c}	1551.9 ± 31.400^{a,c}	-	-
Heavy salt	0.014 ± 0.009^{a,b}	0.039 ± 0.010^{a}	15.495 ± 2.247^{a,b}	36.90 ± 1.94^{a,d}	3.23 ± 0.29^{a}
Light brine	0.031 ± 0.007^{a,d}	0.058 ± 0.014^{a,d}	56.216 ± 1.079^{a,d}	46.00 ± 3.97^{a,d}	3.04 ± 0.11^{a,d}
Light salt	0.037 ± 0.046^{a,d}	0.058 ± 0.008^{a,d}	43.603 ± 2.250^{a,d}	48.40 ± 3.50^{a,d}	3.35 ± 0.44^{a,d}
Light brine + vinegar	0.007 ± 0.001^{a,b}	0.017 ± 0.003^{a,b}	5.405 ± 1.081^{a,b}	43.80 ± 7.44^{a,b}	2.98 ± 0.15^{a,d}

t-test: ^{a} = values differ significantly compared with the fresh sample mean ($P < 0.05$); ANOVA: ^{b} = mean values differ significantly from other means within the same group ($P < 0.05$); $^{c,\,d}$ = values are not significantly different ($P < 0.05$) within the same group.

Table 3. Ash, iron, calcium and sodium content (dry basis) of fresh and preserved *V. amygdalina* leaves

Bitter leaf	Ash (mg/g)	Fe (µg/g)	Ca (µg/g)	Na (µg/g)
Fresh	3.03 ± 0.07^{c}	6.76 ± 0.57^{c}	2.79 ± 0.72	5.21 ± 0.73^{b}
Oven dried	2.28 ± 0.03^{a}	5.19 ± 1.30^{c}	1.99 ± 0.31^{a,c}	3.06 ± 1.15^{a}
Blanched and oven dried	3.43 ± 0.03^{c}	4.54 ± 0.74^{c}	1.21 ± 0.37^{a,c}	2.39 ± 0.38^{a}
Heavy salt	2.64 ± 0.04^{c}	0.46 ± 0.45^{a,d}	4.55 ± 1.72^{a}	318.00 ± 2.65^{a}
Light brine	1.47 ± 0.06^{a}	0.22 ± 0.07^{a}	2.30 ± 0.81^{b}	209.00 ± 12.29^{a,c}
Light salt	1.96 ± 0.40^{a,d}	0.60 ± 0.33^{a}	3.13 ± 0.93^{c}	295.30± 7.38^{a}
Light brine + vinegar	1.90 ± 0.27^{a,d}	0.46 ± 0.15^{a,d}	3.17 ± 2.25^{c}	181.3 ± 41.48^{a,c}

t-test: ^{a} = values differ significantly compared with the fresh sample mean ($P < 0.05$); ANOVA: ^{b} = mean values differ significantly from other means within the same group ($P < 0.05$); $^{c,\,d}$ = values are not significantly different ($P < 0.05$) within the same group.

Heavy salting, light brining, light salting and light brine with vinegar treatment reduced moisture content to 36.90 ± 1.94%, 46.00 ± 3.97%, and 48.40 ± 3.5% and 43.80 ± 7.44%, respectively. This was due to the hypertonic effect of the salt. Moisture reduction is a simple food preservation technique. This was achieved in this work by drying or salting. Salted products were further preserved by the acid produced by salt-tolerant microorganisms present in the medium and by the addition of vinegar.

Table 3 shows variation of ash, iron, calcium and sodium content of *V. amygdalina* samples subjected to different preservation treatments, and Table 4 gives percentage retention or increase of nutrients.

Ash content of fresh *V. amygdalina* leaves was similar to that of the blanched and dried product, but was higher than that of the oven dried product or the products of salting. Ash content was significantly reduced on oven-drying, but was not significantly affected when the vegetable was blanched before oven drying. Heavy salting, light salting and also preservation in light brine with vinegar resulted in significant reduction in ash content.

Iron content of dried vegetable was similar to that of fresh samples –drying did not significantly alter the content of this mineral. However, heavy salting, light salting, light brining alone or with vinegar resulted in serious depletion of iron, with only less than 10% of that present in fresh leaves being retained in the preserved vegetable (Table 3). This was probably due to leaching of this mineral into the preservation media.

Reduction of pH due to fermentation was observed in this study (Fig. 1). Teucher *et al*. (2004) observed that low molecular weight organic acids, produced during fermentation, have the potential to enhance iron absorption *via* formation of soluble ligands, while simultaneously generating low pH that optimizes the activity of endogenous phytase. These benefits may serve to mitigate, from a nutritional standpoint, the loss of iron during preservation of this vegetable in salt.

Fresh *V. amygdalina* leaves had a calcium content of 2.79 ± 0.72 μg/g. This was higher than that of the oven dried (1.99 ± 0.31 μg/g), and blanched + oven dried products (1.21 ± 0.37 μg/g) and similar to light brine preserved (2.30 ± 0.81 μg/g) products, but lower than that of the products resulting from the other salt treatments. Products of heavy salting, light salting, and preservation in light brine and vinegar had calcium content of 4.55 ± 1.72, 3.13 ± 0.93 and 3.17 ± 2.25 μg/g, respectively. The higher calcium content of the salt-preserved leaves was due to absorption of this mineral from the brine. This phenomenon was observed by Jones and Etchells (1944) for brined snap beans, green lima beans and green peas.

The sodium content of fresh and preserved products showed a distinct pattern. Compared with the fresh leaves, oven-drying, with or without blanching decreased the sodium content. Heavy salting, light brining, light salting and light brine with vinegar treatment resulted in an increase in the sodium content of products due to uptake of salt during preservation. Fermented *V. amygdalina* leaves were a better source of sodium than fresh or dried vegetables and may be used as condiment. They would be better as enhancers of the main dish than as the main ingredient because of their high salt content. They could be added in soups and salads, which may then be tasted for salt before the decision to add extra salt to the meal is made.

Compared with the fresh leaves, salt treatment resulted in the loss of about half of the crude fibre content in all cases.

Table 4. Percentage retention (or increase) of nutrients after treatments

Parameter	β-carotene	Total carotene	Vit. C	Moisture	Crude fibre	Ash	Fe	Ca	Na
	(%)								
Fresh	100	100	100	100	100	100	100	100	100
Oven dried	21.78	76.76	75.30	-	-	75.25	76.78	71.33	58.73
Blanched and oven dried	(102.45)*	98.65	72.81	-	-	(113.15)	67.16	43.37	45.87
Heavy salt	4.29	26.78	0.73	36.90	50.16	87.13	6.81	(163.08)	(6103.65)
Light brine	9.51	4.13	2.64	46.00	47.21	48.52	3.25	82.44	(4011.52)
Light salt	11.35	4.13	2.04	48.40	52.12	69.69	8.86	(112.19)	(5667.95)
Light brine + vinegar	2.15	1.21	0.25	43.80	46.27	62.71	6.81	(113.62)	(3479.85)

* = values in brackets indicate percentage increase resulting from preservation treatment.

Conclusion

Study of the effect of preservation by drying and fermentation on the nutrient composition of *Vernonia amygdalina* leaves showed that except for steam blanching prior to drying-which had a protective effect on β-carotene, total carotene and ash content-drying resulted in loss of nutrients in varying degrees relative to their contact in the fresh vegetable.

Salt treatment resulted in decrease in pH in all cases (evidence of production of acid by salt tolerant fermentative microorganisms) and also in the loss of more than half of the moisture in the vegetable. The lowering of pH and loss of moisture indicate that salt treatment gave an environment that would be unfavourable for the growth of most food spoilage organisms. However, salt treatment resulted in serious loss of β-carotene and total carotene, vitamin C, crude fibre and Fe but an increase in Na when compared with the fresh vegetable.

From the viewpoint of nutrient content, drying was a better preservation method than fermentation as it retained more nutrients. Steam blanching prior to drying was better than drying without this pre-treatment for the protection of β-carotene and total carotene.

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