

Valuable components of raw and ripe peels from two Indian mango varieties

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Received 8 November 2005; received in revised form 20 June 2006; accepted 22 June 2006

Abstract

Mango is one of the most important tropical fruits and India ranks first in its world production. During the processing of mango, mainly for mango pulp and preparation of amchur powder, peel is a by-product. Peel forms about 20% of the whole fruit and at present it is a waste product and its disposal has become a great problem. With a view to exploit mango peel as a source of valuable components, in the present study, proximate composition, polyphenols, carotenoids, dietary fibre contents and activities of few enzymes in raw and ripe peels of two Indian mango varieties, namely, Raspuri and Badami were determined. The polyphenol contents in these peels ranged from 55 to 110 mg/g dry peel. Dietary fibre content ranged from 45% to 78% of peel and was found at a higher level in ripe peels. Similarly, carotenoid content was higher in ripe fruit peels. Vitamins C and E contents ranged from 188 to 392 and 205 to 509 µg/g dry peel, respectively; and these were found at a higher level in ripe peels. Both raw and ripe mango peels exhibited significant amount of protease, peroxidase, polyphenol oxidase, xylanase and amylase activities.

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Keywords: Mango; Mangopeel; Bioactive compounds; Phenolics; Carotenoids; Vitamins; Enzymes; Dietary fibre

1. Introduction

Mango (*Mangifera indica* L. Anacardiaceae) is one of the most important tropical fruits in the world and currently ranked 5th in total world production among the major fruit crops (FAO, 2004). As mango is a seasonal fruit, about 20% of fruits are processed for products such as puree, nectar, leather, pickles, canned slices and chutney and these products experience worldwide popularity and have also gained increased importance in the US and European markets (Loelillet, 1994). During processing of mango, by-products such as peel and kernel are generated. Peel contributes about 15–20% of the fruit (Beerh, Raghuramaiah, Krishnamurthy, & Giridhar, 1976). As peel is not currently utilized for any commercial purposes, it is discarded as a waste and becoming a source of pollution. This waste should be treated as a specialized residue due to high levels

of residual phenolics, which may have adverse environmental impacts mainly because of the inhibition of seed germination properties of polyphenols (Negro, Tommasi, & Miceli, 2003). Therefore, industry has increasing cost for its waste treatment. While a number of investigations have been conducted on the composition and possible utilization of mango seed kernel, studies on peels are limited.

Peels are the major by-products obtained during the processing of various fruits and these were shown to be a good source of polyphenols, carotenoids and other bioactive compounds which possess various beneficial effects on human health (Larrauri, Ruperez, & Saura-calixto, 1999; Rodriguez de Sotillo, Hadley, & Holm, 1994a; Wolfe, Xianzhong, & Liu, 2003). Apple pomace has been shown to be a good source of polyphenols and exhibits strong antioxidant and anti-proliferative activity (Wolfe et al., 2003). Grape pomace, which is a wine industry by-product serves as a good source of anthocyanins, catechins, flavanoids, phenolic acids and dietary fibre (Larrauri et al., 1999; Mazza, 1995; Mazza & Miniati, 1995). Citrus peel

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is also a rich source of pectins and flavanoids such as hesperidins, eriocitrin and nobiletin (Anagnostopoulou, Kefalas, Papageorgiou, Assimopoulou, & Boskou, 2006; Coll, Coll, Laencina, & Tomas-Barberan, 1998; Li, Yu, & Ho, 2005; Mandalari et al., 2006). Banana and tomato peels are reported to be a good source of carotenoids (Baysal, Ersus, & Starmans, 2000; Subaigo, Morita, & Sawada, 1996). Aqueous extract of potato peel was shown to provide a good source of phenolic acids (Rodriguez de Sotillo, Hadley, & Holm, 1994b) and exhibited good antioxidant activity (Rodriguez de Sotillo et al., 1994a). Mango peels were found to contain polyphenols and dietary fibre (Larrauri, Ruperez, Borroto, & Saura-calixto, 1996).

Though considerable work was done with regard to enzymes like amylase and protease, among others, in mango pulp, very few reports are available with regard to enzymes in mango peel. Prabha and Patwardhan (1986), Saby John, Bhat, and Prasada Rao (2002) and Robinson, Loveys, and Chako (1993) reported the presence of polyphenol oxidase in mango peel, while Saby John et al. (2002) also reported the presence of peroxidase.

India is the major producer of mango, and peeled raw mangoes are processed for the preparation of amchur and ripe mangoes are processed for mango pulp and fruit bars. Therefore, both raw and ripe peels are available in large quantities as a by-product in mango processing industry.

In the present study, valuable bioactive compounds such as polyphenols, carotenoids, dietary fibres and enzymes in the raw and ripe peels of two Indian mango varieties, namely, Badami (Alphonso) and Raspuri were determined with the aim of exploiting the potential value of the peel.

2. Materials and methods

2.1. Materials

Raspuri and Badami mango varieties grown in CFTRI campus, Mysore, India, were used in this study. Mango varieties were harvested at maturity and peel was removed using a sharp knife and the underlying pulp removed by gently scraping with its blunt edge. To obtain the ripe peel, some fruits were allowed to ripen at room temperature and the peel was removed as described earlier. The fresh peels thus obtained were used for analysis.

Vitamin C, pyrogallol, α -amylase, pepsin, celite, α -tocopherol, bovine serum albumin, Coomassie brilliant blue G-250, catechol, *O*-dianisidine, azocasein, soluble starch, xylan, xylose, α, α' -bipyridyl, Tris, 2,5-dinitrosalicylic acid and 2,4-dinitrophenylhydrazine were obtained from Sigma Chemicals (St. Louis, MO, USA). All other chemicals and solvents were of analytical grade and obtained from Qualigens Chemicals (Mumbai, India).

2.2. Proximate composition analysis

Moisture, protein, fat, ash and crude fibre content in fresh peel were determined by AOAC methods (1998). The carbo-

hydrate estimation was done by phenol-sulphuric acid method (Dubois, Gilles, Hamilton, Robers, & Smith, 1956) after hydrolyzing the mango peel with 6N HCl (100 °C, 6 h).

2.3. Estimation of total phenolics, carotenoids, vitamin C, vitamin E and dietary fibre

Mango peel was homogenized with 80% ethyl alcohol or 80% acetone or 0.05 M sodium phosphate buffer (pH 7.5) and vortexed using a mixer. The clear solution obtained after centrifugation for 15 min at 10,000g was subjected to total polyphenol estimation using the method of Swain and Hillis (1959). Gallic acid in 80% ethanol was used as a standard. Peel (1 g) was homogenized with 40 ml of methanol containing 1 g KOH and the carotenoids were extracted using the method of Tee and Lim (1991). The total carotenoid content was estimated using two different colorimetric methods reported by Davis (1976) and Litchenthaler (1987). Vitamin C (ascorbic acid) content was determined according to the method of Omaye, Turnbull, and Sayberlich (1973). Ascorbic acid was used as a standard. Vitamin E content in mango peel was determined according to the method of Joshi and Desai (1952). α -Tocopherol was used as a standard to estimate total vitamin E content. The dietary fibre estimation was done by an enzymatic gravimetric method (Asp, Johansson, Hallmer, & Siljestrom, 1983). Peel (0.5 g) was homogenized in 20 ml of sodium phosphate buffer (0.1 M, pH 6.0) and was analyzed for soluble (SDF) and insoluble (IDF) dietary fibre. The samples were treated with thermo-stable α -amylase (Termamyl) and then digested with pepsin and pancreatin. Soluble and insoluble dietary fibre were separated by filtration. The filtrate was subjected to alcohol precipitation and filtered to obtain SDF and both the precipitates were dried overnight at 105 °C and were incinerated at 500 °C for 8 h. A control was performed following the same procedure. Total dietary fibre was then calculated as combined value of SDF and IDF.

2.4. Enzyme extraction

To 1 g of fresh peel, 0.5 g of acid washed sand was added and was ground into paste using 5 ml of 50 mM sodium phosphate buffer (pH 7.5) using mortar and pestle at 4 °C. To the resultant paste, 20 ml of buffer was added, stirred for 1 h and was centrifuged at 8000g for 15 min at 4 °C. The supernatant obtained was used to estimate the protein content and to assay peroxidase, polyphenol oxidase, xylanase, protease and amylase activities. The protein content in the mango peel extract was determined using the dye binding method of Bradford (1976). Bovine serum albumin was used as a standard.

2.5. Enzyme assays

Peroxidase, polyphenol oxidase, protease and amylase activities in mango peel extracts were determined as

described by Saby John, Bhat, and Prasada Rao (2003). Briefly, peroxidase activity was determined using 1 ml of reaction mixture containing varying quantities of appropriately diluted enzyme, 100 μ l of 8 mM *O*-dianisidine and 100 μ l of 1% H₂O₂ at its optimum pH 5.5 in 50 mM sodium acetate buffer. Polyphenol oxidase activity was assayed using 1 ml of reaction mixture containing varying quantities of appropriately diluted enzyme, 0.1 ml of 0.5 M catechol in 50 mM sodium phosphate buffer (pH 7.0). One unit of both polyphenol oxidase and peroxidase activities were defined as the amount of enzyme which produced an increase of one absorbance per minute at 460 and 420 nm, respectively. Protease activity was assayed using azocasein (25 mg/ml of 50 mM Tris–HCl buffer, pH 8.0) as substrate using 50 mM Tris–HCl buffer (pH 8.0). An increase in one absorbance per minute at 440 nm was regarded as 1 unit of activity. Amylase was assayed using 1% gelatinized soluble starch solution as substrate in 50 mM sodium acetate buffer (pH 4.6). One unit of enzyme activity was defined as one μ mole maltose equivalents released per minute. The xylanase activity was measured using 0.5% xylan as substrate in 100 mM sodium acetate buffer (pH 4.8) and the xylose released was estimated as reducing sugar by 2,5-dinitrosalicylic acid method as described by Miller (1959). Xylose was used as standard. One unit of xylanase activity was defined as the amount of enzyme required to release 1 μ mol of xylose per minute.

2.6. Statistical analysis

The experimental design was completely randomized, with three replicates. All data were expressed as mean values \pm SD. The comparison between the mean values were tested using Duncan's new multiple-range test at a level of $P \leq 0.05$ (Steel & Torrie, 1980).

3. Results and discussion

3.1. Proximate composition

The proximate composition of raw and ripe mango peels of Badami and Raspuri is shown in Table 1. The total protein content in peel ranged from 1.76% to 2.05%. The moisture content ranged from 66% to 75% and it was found to be more in ripe mango peels. The fat content ranged from 2.16% to 2.66%. The carbohydrate content in the mango peel was determined after acid hydrolysis and it ranged from 20.8% to 28.2%. The crude fibre content ranged from 3.28% to 7.40% and was found to be higher in ripe mango peels. The ash content ranged from 1.16% to 3.0%. The proximate composition of mango fruit pulp was 81% moisture, 0.6% protein, 0.4% fat, 0.4% minerals, 0.7% fibre and 16.9% carbohydrate as reported by Gopalan, Ramasastri, and Balasubramanian (1999).

3.2. Total phenolics, carotenoids, vitamin C, vitamin E and dietary fibre

Mango peels were extracted with 80% (v/v) ethyl alcohol or 80% (v/v) acetone or sodium phosphate buffer (50 mM, pH 7.5) separately and the phenolic contents in the extracts were determined. Of the three extracts, acetone extracted maximum amount of polyphenols followed by ethanol from both raw and ripe peels (Table 2). Polyphenol contents in acetone extracts of raw and ripe varied from 55 to 110 mg GAE/g dry peel and in both the varieties the total polyphenol content was found to be significantly higher in raw peels compared to that of ripe peels. Earlier, Larrauri et al. (1996) reported the total polyphenols content in aqueous methanol extract of ripe peel of Hayden variety to be 70 mg/g. These values are in the range reported in the present study. The polyphenol content

Table 1
Proximate composition (%) of mango peel

Mango variety	Protein	Carbohydrate	Crude fibre	Moisture	Fat	Ash
Raspuri raw	1.76 \pm 0.50 ^b	26.50 \pm 0.50 ^b	3.80 \pm 0.30 ^b	66.00 \pm 0.50 ^a	2.49 \pm 0.03 ^b	1.40 \pm 0.20 ^a
Raspuri ripe	2.05 \pm 0.02 ^c	28.20 \pm 0.60 ^c	5.80 \pm 0.10 ^c	72.50 \pm 0.50 ^c	2.22 \pm 0.02 ^a	1.16 \pm 0.14 ^a
Badami raw	1.45 \pm 0.11 ^a	21.52 \pm 0.12 ^a	3.28 \pm 0.14 ^a	70.25 \pm 0.25 ^b	2.16 \pm 0.06 ^a	3.00 \pm 0.20 ^b
Badami ripe	1.76 \pm 0.08 ^b	20.80 \pm 0.20 ^a	7.4 \pm 0.20 ^d	75.25 \pm 0.25 ^d	2.66 \pm 0.03 ^c	1.30 \pm 0.10 ^a

Values are expressed on dry weight basis. All data are the mean \pm SD of three replicates. Mean value followed by different letters in the same column differs significantly ($P \leq 0.05$).

Table 2
Polyphenols content (mg/g) of mango peel

Mango variety	Buffer extract	Alcohol extract	Acetone extract
Raspuri raw	29.40 \pm 0.60 ^d	73.88 \pm 0.35 ^c	109.7 \pm 0.82 ^d
Raspuri ripe	13.90 \pm 1.24 ^b	46.31 \pm 3.50 ^b	100.00 \pm 1.90 ^c
Badami raw	20.10 \pm 0.72 ^c	37.92 \pm 0.86 ^a	90.18 \pm 0.57 ^b
Badami ripe	9.84 \pm 0.88 ^a	33.31 \pm 1.20 ^a	54.67 \pm 1.50 ^a

Values are expressed on dry weight basis. All data are the mean \pm SD of three replicates. Mean value followed by different letters in the same column differ significantly ($P \leq 0.05$).

was reported in Irwin mango variety and this was higher in ripe peel compared to raw peel (Ueda, Sasaki, Utsunimiya, Inaba, & Bayashi, 2000). The content of total phenols was higher in the peel than the pulp at any stage of mango fruit development (Lakshminarayana, Subhadra, & Subramanyam, 1979; Ueda et al., 2000). The total polyphenol content in grape pomace extracts was reported to range from 68.8 to 98.3 mg GAE/g dry peel (Gulcan, Osman, Nilgun, & Zehra, 2004) which is comparable to polyphenol content in mango peels, where as in apple pomace it was reported to be 33.42 mg GAE/g dry peel (Wolfe et al., 2003) much lower than mango peels.

The carotenoid content in mango peel was estimated using two different spectrophotometric methods of Davis (1976) and Litchenthaler (1987). Results obtained using both methods are comparable (Table 3). The carotenoid content was found to be more in ripe mango peels compared to raw peels (Table 3). Modi and Reddy (1967) showed that ripe mangoes are 10 times richer in carotenoids than partially ripe ones. The present study showed that carotenoid content in mango peels was 4–8 times higher in ripe mango peels than in raw mango peels.

The vitamin C content ranged from 188 to 392 µg/g of the peel and in both varieties it was more in ripe peels compared to raw peels (Table 4). Earlier, vitamin C content in mango peel of five different varieties was reported which ranged from 190 to 2570 µg/g (Teotia, Manan, & Saxena, 1987). The vitamin E content in mango peel ranged from 205 to 509 µg/g and was higher in ripe mango peel than raw mango peels (Table 4). Recently, Burns, Fraser, and Bramley (2003) reported the presence of α-tocopherol in

mango pulp. However, no report is available with respect to vitamin E content in mango peel.

The crude fibre content of mango peel presented in Table 1 represents mainly cellulose fractions, which is a major part of insoluble dietary fibre. The dietary fibre content in mango peels of different varieties were estimated (Table 5). The total dietary fibre (TDF) content in dry peel varied from 45% to 78%. In both Raspuri and Badami, soluble and insoluble fibre contents were higher in ripe peels compared to raw peels. Among these, Badami ripe showed highest TDF content and Raspuri raw showed the lowest. Earlier, Larrauri et al. (1996) reported that the dried mango peel contained 28.1% of SDF and 43.4% IDF. In the present study, IDF/SDF ratio varied from 1.7 to 2.0. The soluble dietary fibre content in both raw and ripe mango peels are more than 35% of TDF. For health benefits, it is reported that 30–50% SDF and 50–70% insoluble dietary fibre are considered to be well-balanced proportions (Schneeman, 1987). Though in terms of health benefits, both IDF and SDF complement each other, each fraction has different physiological effect. Insoluble dietary fibre relates to both water absorption and intestinal regulation where as SDF associates with cholesterol in blood and diminishes its intestinal absorption. The characteristic feature of mango peel is that it has high content of soluble dietary fibre, which is reported to have more health beneficial effects. SDF content in apple waste was reported to be 23% of the TDF, while it was 36% in orange byproducts. However, SDF contents in wheat bran and oat bran were 6.6% and 15%, which are low (Grigelmo-Miguel & Martin Beloso, 1999).

3.3. Enzymes of mango peel

Peroxidase, polyphenol oxidase (PPO), protease and carbohydrases are involved in fruit ripening and these enzymes may be associated with mango peel. Therefore, both raw and ripe mango peels were assayed for these enzymes. PPO activity varied between 36 and 108 U/g dry peel and was higher in ripe peels. Badami ripe peel had more PPO activity, while Raspuri raw had less activity (Table 6). Earlier, presence of PPO was reported in mango peel (Prabha & Patwardhan, 1986; Robinson et al., 1993; Saby John et al., 2002). Prabha and Patwardhan (1986) reported that PPO content in ripe peel was more than that

Table 3
Total carotenoids content (µg/g) of mango peel

Mango variety	Davis method ^A	Litchenthaler method ^B
Raspuri raw	493 ± 31 ^a	547 ± 18 ^b
Raspuri ripe	3945 ± 85 ^c	3337 ± 65 ^d
Badami raw	365 ± 10 ^a	387 ± 35 ^a
Badami ripe	1400 ± 40 ^b	1520 ± 84 ^c

Values are expressed on dry weight basis. All data are the mean ± SD of three replicates. Mean value followed by different letters in the same column differ significantly ($P \leq 0.05$).

^A Davis (1976).

^B Litchenthaler (1987).

Table 4
Vitamin E and Vitamin C content (µg/gm) of mango peel

Mango variety	Vitamin E	Vitamin C
Raspuri raw	205 ± 4 ^a	188 ± 18 ^a
Raspuri ripe	308 ± 11 ^b	349 ± 11 ^c
Badami raw	337 ± 3 ^c	315 ± 10 ^b
Badami ripe	509 ± 14 ^d	392 ± 21 ^d

Values are expressed on dry weight basis. All data are the mean ± SD of three replicates. Mean value followed by different letters in the same column differs significantly ($P \leq 0.05$).

Table 5
Dietary fibre content (%) of mango peel

Mango variety	SDF	IDF	TDF
Raspuri raw	15.70 ± 0.30 ^a	28.99 ± 0.51 ^a	44.70 ± 0.71 ^a
Raspuri ripe	23.81 ± 0.91 ^c	39.99 ± 2.90 ^b	63.80 ± 3.80 ^b
Badami raw	21.42 ± 0.73 ^b	42.61 ± 0.02 ^b	64.13 ± 0.25 ^b
Badami ripe	28.06 ± 0.41 ^d	50.33 ± 0.75 ^c	78.40 ± 0.20 ^c

SDF, soluble dietary fibre; IDF, insoluble dietary fibre; TDF, total dietary fibre.

Values are expressed on dry weight basis. All data are the mean ± SD of three replicates. Mean value followed by different letters in the same column differs significantly ($P \leq 0.05$).

Table 6
Enzyme activities (U/g) of mango peels

Mango variety	Amylase	Xylanase	Protease	Peroxidase	Polyphenol oxidase	Protein (mg/g)
Raspuri raw	1.1 ± 0.08 ^a	5.9 ± 0.45 ^b	4573 ± 14 ^a	275 ± 10 ^d	36.4 ± 1.2 ^a	9.9 ± 1.1 ^b
Raspuri ripe	2.2 ± 0.13 ^b	4.3 ± 0.33 ^a	10363 ± 363 ^b	242 ± 2 ^b	74.7 ± 0.1 ^b	4.0 ± 0.8 ^a
Badami raw	0.9 ± 0.07 ^a	9.3 ± 1.12 ^c	11058 ± 381 ^c	213 ± 8 ^a	72.0 ± 4.0 ^b	12.7 ± 0.4 ^c
Badami ripe	2.8 ± 0.05 ^c	6.4 ± 0.80 ^b	11173 ± 74 ^c	260 ± 4 ^c	108.0 ± 1.0 ^c	5.2 ± 0.7 ^a

Values are expressed on dry weight basis. All data are the mean ± SD of three replicates. Mean value followed by different letters in the same column differs significantly ($P \leq 0.05$).

of raw peel. PPO has been widely studied in various fruits such as pineapple (Das, Bhat, & Gowda, 1997) and apple (Jonovitz-Klapp, Richard, & Nicolas, 1989), among others. The PPO activity was detected in the skin and flesh of Irwin mango variety and a gradual increase in the enzyme activity was reported from raw to ripe stages of mango during maturation (Ueda et al., 2000). Peroxidase activity was highest in Raspuri raw peel and lowest in Badami raw mango peel (Table 6). Increase in peroxidase activity in fruit ripening was reported in apples (Gorin & Heidema, 1976), while decrease in peroxidase activity with ripening was reported in tomato (Thomas, Jen, & Morr, 1981). However, no such trend was seen in case of peroxidase in mango peels in the present study.

Peel extracts showed good protease activity. The protease activity was found to be significantly more in ripe mango peels than in the raw mango peels. The protease activity ranged from 4573 to 11173 U/g of dry peel (Table 6). The protease activity was found to be higher in Badami ripe peel. The protease activity may be involved in protein turn over and particularly during the final stages of senescence by protein catabolism, which is responsible for increase in free amino acids and amides. Xylanase enzymes are a group of cell wall degrading enzymes. The xylanase activity ranged from 4.3 to 8.8 U/g dry peel. Xylanase activity was found to be maximum in raw mango peel than the ripe mango peel. Ali, Aurmugam, and Lazan (1995) reported the presence of xylanase activity in mango fruit extract and their activities unchanged throughout the ripening stages of mango. Amylase activity in mango peel ranged from 1.1 to 2.8 U/g dry peel. Amylases have a role in fruit ripening. It has been reported that mango fruit at the mature green stage contains some accumulated starches (Subramanian, Gouri, & Krishnamurthy, 1976), which is mobilized during ripening (Morga, Lustre, Tunac, Balagot, & Soriano, 1979). α -Amylase activity was reported to increase at least four times in activity during ripening of mango fruit (Majumdar, Modi, & Palejwala, 1981).

4. Conclusions

This study showed that mango peel is a rich source of bioactive compounds such as polyphenols, carotenoids, vitamins C and E, dietary fibres and enzymes. Consumption of mango either as whole fruit or in some processed form will increase their intake in the diet. Mango peel, by-product of mango processing industry, could be a rich

source of bioactive compounds, and enzymes such as protease, peroxidase and polyphenol oxidase. This new source will be potential as a functional food or value added ingredients in future in our dietary system. Mango peel if conveniently processed, could furnish useful products that may balance out waste treatment costs and also decrease the cost of main product. Therefore, there is a scope for the isolation of these active ingredients and also use of mango peel as an ingredient in processed food products such as bakery products, breakfast cereals, pasta products, bars and beverages.

Acknowledgements

C.M. Ajila thanks Council of Scientific and Industrial Research, New Delhi for the award of Senior Research Fellowship. We thank Dr. R.N. Tharanathan and Dr. Mahadevamma for their help in dietary fibre estimation.

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