Total antioxidant activity of Florida's tropical fruit
First report for Trust Fund Project with Tropical Fruit Growers of South Florida
USDA/ARS Citrus & Subtropical Products Laboratory (USCSPL)
Winter Haven, FL

Kanjana Mahatanatawee
Kevin Goodner
Elizabeth Baldwin
John Manthey
Gary Luzio

Introduction

There is evidence for beneficial roles of fruit and vegetables in the human diet providing protection against cellular damage caused by exposure to high levels of free radicals. Radical scavengers have attracted special interest because they can protect the human body from free radicals that may cause many diseases, including cancer, and contribute to the aging process (Ames et al. 1993). Fruits contain many different antioxidant compounds (i.e. vitamin C and E, carotenoids and phenolic compounds), that serve as radical scavengers and it is relatively difficult to measure each antioxidant component separately. Therefore, the total antioxidant activity levels were measured directly from fruit extracts. Recently, several methods have been developed to measure total antioxidant activity, based on different reaction mechanisms. Thus, it is necessary to evaluate whether different methods can provide comparable antioxidant values for the same sample. It has been reported that the contributions of phenolic compounds to antioxidant activities were much greater than those of vitamin C and carotenoids, therefore total phenolics were also assayed. The objective of this study was to determine the radical quenching activity of tropical fruit extract and the total phenolics. Two methods were used to test the antioxidant activity of tropical fruit including one based on the evaluation of the radical scavenging capacity by using the free radical DPPH, and the other based on the peroxyl radical scavenging activity (ORAC). Since different ethnic groups prefer different maturity stages of some fruits, like mango and papaya, both ripe and "green" stages were assayed.

Materials and Methods

Twelve tropical fruit from south Florida consisting of red guava, white guava, carambola, red pitaya, white pitaya, mamey, sapodilla, lychee, longan, green mango, ripe mango, and green papaya were analyzed for their antioxidant activity and total phenolics. Fruit puree (edible portion) was extracted with methanol. Two methods were used to evaluate the antioxidant potential of fruit extracts; ORAC (oxygen radical absorbance capacity) (Talcott et al, 2003) and free radical DPPH scavenging activity (Manthey, 2004). Total phenolics were analyzed using the Folin-Ciocalteu assay and expressed in gallic acid equivalents (Sellappan et al. 2002). One-way analysis of variance and Duncan Multiple Range Test (DMRT) as conducted to identify differences among means. Statistical significance was determined at $p < 0.05$.

Results and Discussions

Florida tropical fruits had widely different total phenolics and antioxidant activity. The total phenolics, ORAC and DPPH ranged from 205.41 to 2316.71 μg gallic acid equivalent/ gram puree, 0.01 to 16.70 μmol Trolox equivalent/ gram puree, and 2.08 to 620.18 μg gallic acid equivalent/ gram puree, respectively (Table 1, Fig 1, 2 and 3). The antioxidant activities evaluated by both ORAC and DPPH showed similar trends with correlation $r = 0.85$ (Fig 4) where red guava and carambola had the highest and sapodilla and green papaya had the lowest. Differences were also observed due to variations in the antioxidant constituents of the fruit, reaction kinetics and differences in the methods of analysis. There were strong correlations ($r = 0.96$ and 0.92 assayed by ORAC and DPPH, respectively) between antioxidant activity and total phenolics (Fig 5 and 6), indicating that phenolics contributed to the antioxidant activity. The highest antioxidant capacities were observed in red guava and carambola, which also obtained a high content of total phenolics. **Guava, carambola, red dragon fruit, mamay and lychee ORAC values compared well with published ORAC values for other common fruits, including cucumber, melon, pear, tomato, carrot, apple, banana, white and red grape, pink grapefruit, kiwi, orange, plum, spinach, strawberry, red raspberry, and some blueberries (for which there is quite a range of values reported) (Table 2). The total phenolic values (antioxidant activity coming from phenolics) compared favorably to strawberry, red raspberry, blackberry, black raspberry, and blueberries (for which there is also a range of values reported).**

The most common antioxidants present in fruit are vitamins C and E, carotenoids, flavonoids, and thiol (SH) compounds. There were several reports that the contribution of phenolic compounds to antioxidant activity was much greater than those of vitamin C and carotenoids. The vitamin C content from the USDA database (http://www.nal.usda.gov) was used to estimate contribution of vitamin C to the total antioxidant activity (Table 1). The contribution of vitamin C to the total antioxidant activity by ORAC and DPPH assays were relatively low ($r = 0.42$ and 0.20 respectively, Fig 7 and 8). This suggests that the major source of antioxidant capacity of fruits may be not from vitamin C, but rather from phenolics. **For example, carambola has high total phenolic, ORAC and DPPH values, yet relatively low levels of vitamin C compared to guava. This would indicate that carambola is much more nutritious than would be assumed from vitamin C values. Likewise red guava had higher total phenolic, ORAC, and DPPH values than did white guava, but the same reported amount of vitamin C. The higher antioxidant activity of red compared to white guava and dragon fruit is likely due to the red pigments.**

The protection in the body provided against oxidative damage by fruit and vegetables has been attributed to the fact that these foods may provide an optimal mix of phytochemicals, such as natural antioxidants, fiber, and other bioactive compounds. A high intake of vitamin C may act in some situations as a pro-oxidant in the body only when free transition metals are available at the same time. Therefore, the supplementation of these natural antioxidants through a balanced diet containing adequate fruit could be much more effective than the supplementation of an individual antioxidant such as vitamin C or vitamin E.

Work is currently ongoing to determine the pectin/fiber, flavor and carotenoid content of these fruits. Although the funds supplied by the Tropical Fruit Growers of South Florida ($30,000), covered nine months of a postdoc salary, during which time the above data was collected and analyzed and other studies initiated, the USCSPL is funding the posdoc salary through October, 2005 to complete the pectin, carotenoid and flavor analyses. We also thought it would be a good idea to include ripe papaya, to compare to green papaya antioxidant and total phenolic values. Continued donation of fruit samples (ripe papaya and others, if needed) during this period would be much appreciated. A publication on the data presented here is in preparation.

References

1. Ames, B.N., Shigenaga, M.K., and Hagen, T.M.. Oxidants, antioxidants, and the degenerative diseases of aging. Proc. Natl. Acad. Sci. USA. 1993, 90, 7915-7922.
2. Talcott, S. T.; Percival, S. S.; Pittet-Moore, J.; Celoria, C. Phytochemical composition and antioxidant stavility of fortified yellow passion fruit (*Passiflora edulis*). J. Agric. Food Chem. 2003, 51, 935-941.
3. Manthey, J. A. Fractionation of orange peel phenols in ultrafiltered malasses and mass balance studies of their antioxidant levels. J. Agric. Food Chem. 2004, 52, 7586-7592.
4. Sellappan, S.; Akoh, C. C.; Krewer, G. Phenolic compounds and antioxidant capacity of Georgia-grown blueberries and black-berries. J. Agric. Food Chem. 2002, 50, 2432-2438.
5. Wang, H.; Cao G.; Prior, R. L. Total antioxidant capacity of fruits. J. Agric. Food Chem. 1996, 44, 701-705.
6. Cao, G.; Sofic, E.; Prior, R.L. Antioxidant capacity of tea and common vegetables. J. Agric. Food Chem. 1996, 44, 3426-3431.
7. Luximon-Ramma, A.; Bahorun, T.; Crozier, A. Antioxidant actions and phenolic and vitamin C contents of common Mauritian exotic fruits. J. Sci. Food Agri. 2003, 83, 496-502.
8. Prior, R. L.; Cao, G.; Martin, A.; Sofic, E.; McEwen, J.; O'Brien, C.; Lischner, N.; Ehlenfeldt, M.; Kalt, W.; Krewer, G. and Mainland C. M. Antioxidant capacity as influenced by total phenolic and anthocynin content, maturity, and variety of *Vaccinium* species. J. Agric. Food Chem. 1998, 46, 2686-2693.
9. Wang, S. Y. and Lin, H. Antioxidant activity in fruits and leaves of blackberry, raspberry, and strawberry varies with cultivar and developmental stage. J. Agric. Food Chem. 2000, 48, 140-146.

Table 1 Total phenolic, antioxidant activity by ORAC and DPPH and vitamin C content*

Fruit	Total phenolic	ORAC	DPPH	vitamin C*
	μg GA/g puree	μM TE/g puree	μg GA/g puree	mg/ 100 g
red guava	2316.71	16.70	609.26	228.30
carambola	2207.65	12.94	620.18	34.40
white guava	1589.29	9.90	298.59	228.30
red dragon	1075.76	7.59	134.14	
mamey	1010.47	6.56	247.11	14.00
lychee	770.12	5.42	103.75	71.50
white dragon	523.41	2.96	46.41	
ripe mango	508.94	2.17	123.65	27.70
green mango	505.76	1.49	167.50	27.70
sapodilla	501.76	1.36	2.08	14.70
longan	481.88	3.31	61.25	84.00
green papaya	205.41	0.01	10.36	61.80

* USDA database

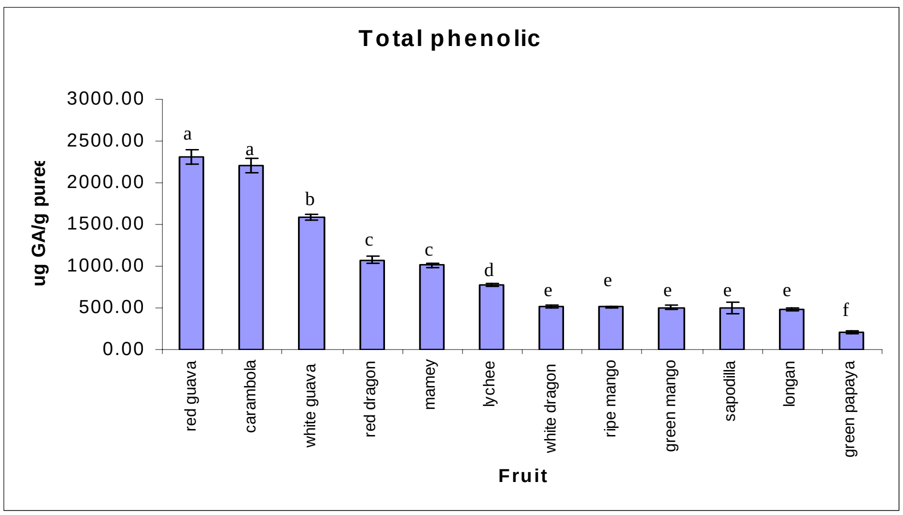

Figure 1 Rank order for total phenolic of twele fruit expressed as μg gallic acid Equivalent/ gram puree

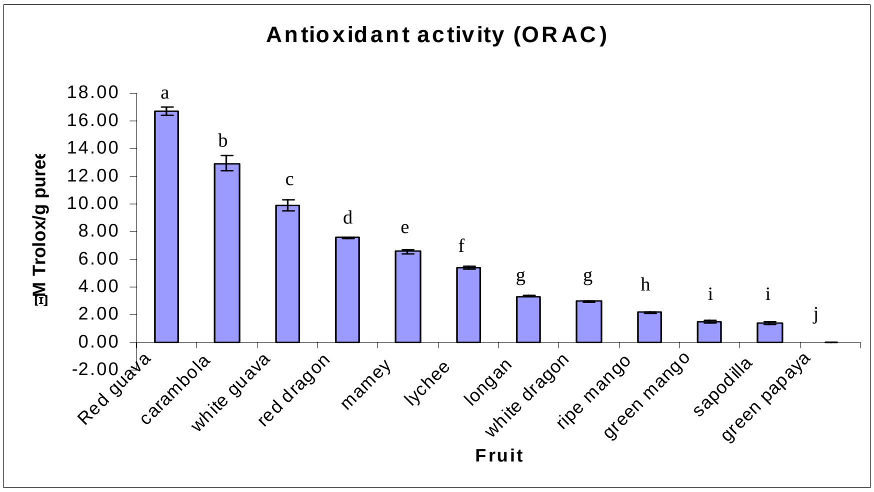

Figure 2 Rank order for antioxidant activity of twelve fruit base on the ORAC value Expressed as µmol Trolox equivalent/ gram puree

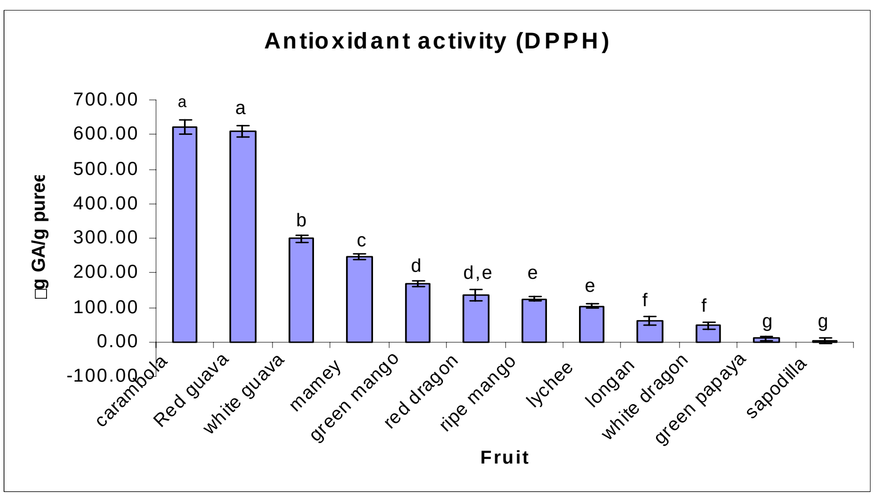

Figure 3 Rank order for antioxidant activity of twelve fruit base on the DPPH value Expressed as µg gallic acid equivalent/ gram puree

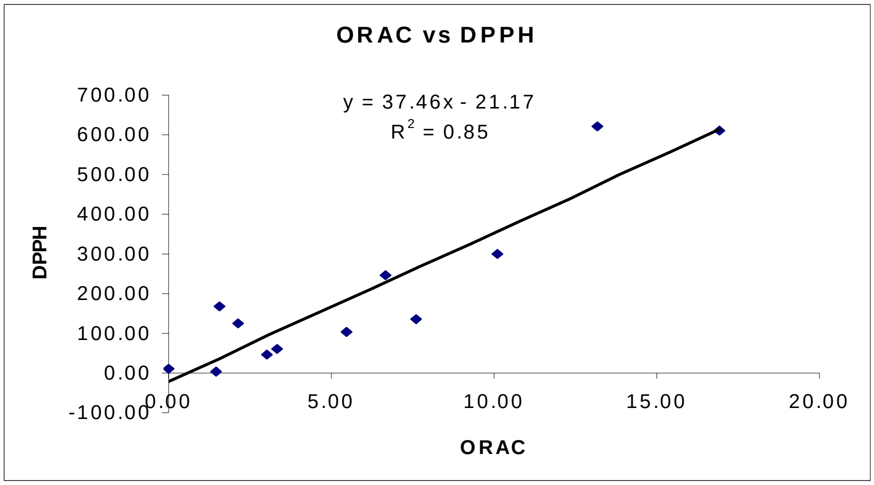

Figure 4 Correlation between ORAC and DPPH

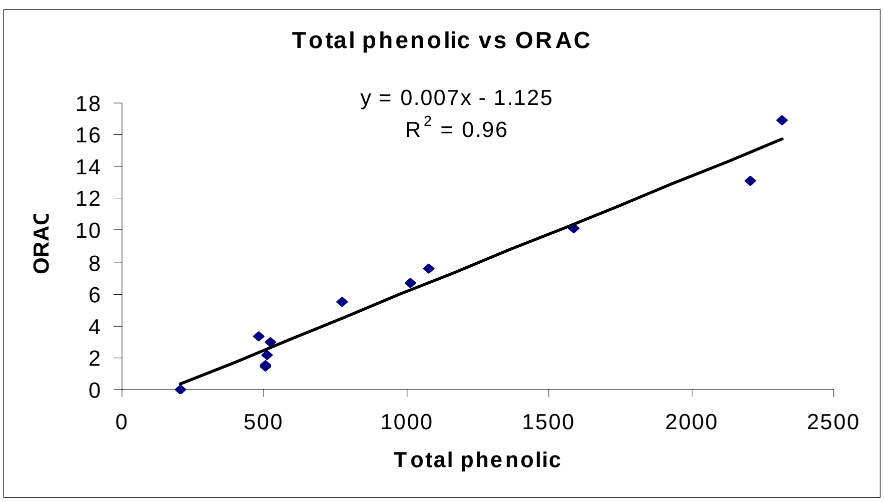

Figure 5 Correlation between total phenolic and antioxidant activity by ORAC

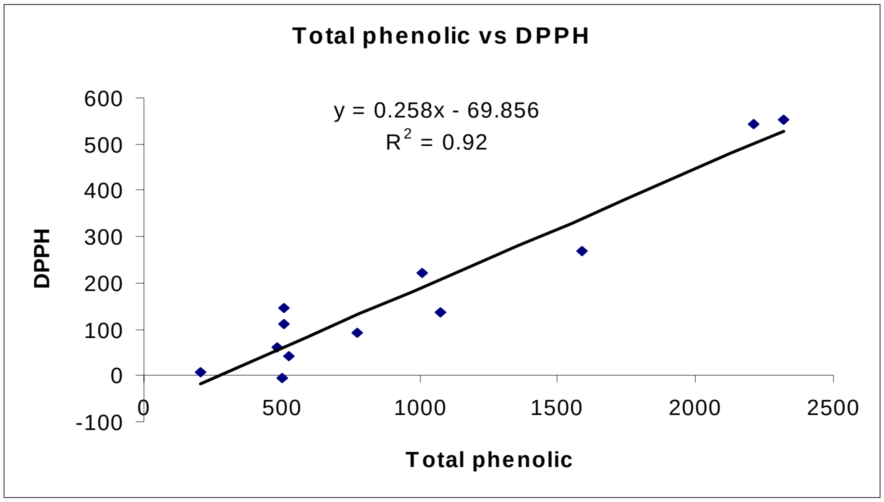

Figure 6 Correlation between total phenolic and antioxidant activity by DPPH

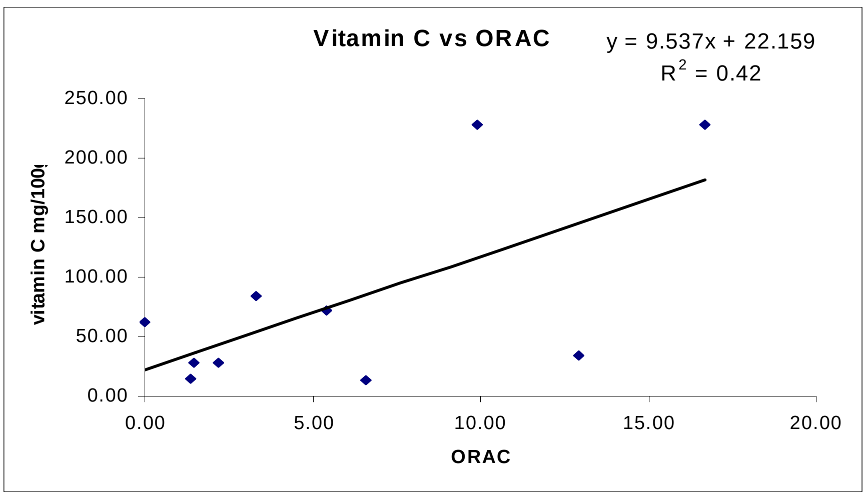

Figure 7 Correlation between vitamin C and antioxidant activity by ORAC

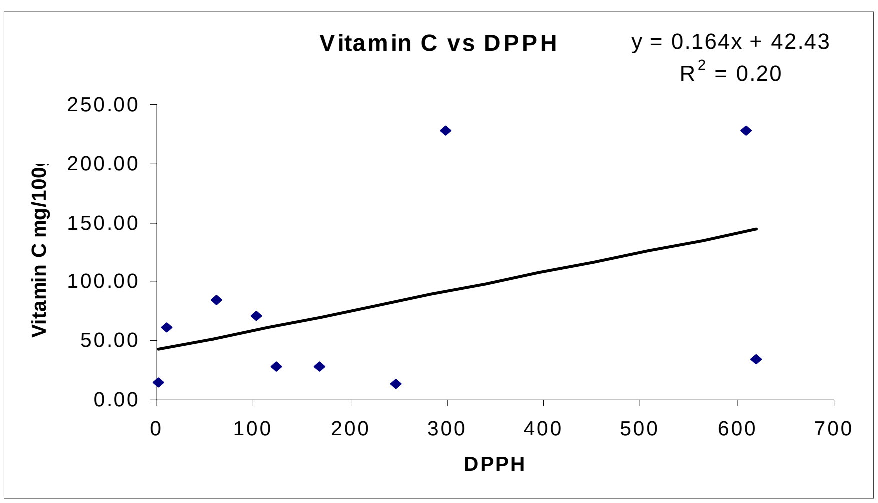

Figure 8 Correlation between vitamin C and antioxidant activity by DPPH

Table 2 Antioxidant activity (ORAC) and total phenolics of other fruits and vegetables compared to the selected tropical fruits from this study (**in bold**). Fruits are ranked from low to high ORAC values

Fruit	ORAC	Total phenolic
	μM TE/g puree	μg GA/g puree
green papaya	**0.01**	**205**
cucumber[b]	0.50	
melon[a]	0.97	
pear[a]	1.34	
sapodilla	**1.36**	**502**
green mango	**1.49**	**506**
tomato[a]	1.89	
carrot[b]	2.10	
ripe mango	**2.17**	**509**
apple[a]	2.18	
banana[a]	2.21	
white dragon	**2.96**	**523**
longan	**3.31**	**482**
grape, white[a]	4.46	
grapefruit, pink[a]	4.83	
lychee	**5.42**	**770**
kiwi fruit[a]	6.02	
mamey	**6.56**	**1010**
grape,red[a]	7.39	
orange[a]	7.50	
red dragon	**7.59**	**1076**
plum[a]	9.49	
white guava	**9.90**	**1589**
spinach[b]	12.60	
carambola	**12.94**	**2208**
strawberry[e]	14.9	1030
strawberry[a]	15.36	
red guava	**16.70**	**2317**
red raspberry[e]	18.2	2340
garlic[b]	19.40	
blackberry[e]	22.4	2260
black raspberry[e]	28.2	2670
blueberries[d]	14 - 38	1810 - 4580
carambola[c]		2099
lychee[c]		288
mango[c]		560
pink guava[c]		1264
white guava[c]		2473

Sources of data on other fruits, and from other reports on tropical fruits:
[a]Wing et al., 1996, [b]Cao et al., 1996, [c]Luximon-Ramma et al., 2003, [d]Priot et al., 1998, [e]Wang and Lin, 2000.