

# Low-Density Lipoprotein Antioxidant Activity of Phenolic Compounds and Polyphenol Oxidase Activity in Selected Clingstone Peach Cultivars

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The antioxidant potential of eight clingstone peach cultivars was investigated by determining phenolic compounds and inhibition of low-density lipoprotein (LDL) oxidation. Cultivars low in polyphenol oxidase (PPO) were also selected to minimize enzymatic browning. Inhibition of LDL oxidation varied from 17.0 to 37.1% in peach flesh extract, from 15.2 to 49.8% in whole peach extract, and from 18.2 to 48.1% in peel extract. Total phenols were 432.8–768.1 mg/kg in flesh extract, 483.3–803.0 mg/kg in whole extract, and 910.9–1922.9 mg/kg in peel extract. The correlation coefficient between relative LDL antioxidant activity and concentration of total phenols was 0.76. Peel PPO activity was higher than flesh activity in most cultivars. The lowest PPO and specific activities were found in the Walgant cultivar, followed by Kakamas and 18-8-23. These three cultivars combine the desirable characteristics of strong antioxidant activity, low PPO activity, and lower susceptibility to browning reactions.

**Keywords:** *Peach; antioxidants; low-density lipoproteins; LDL oxidation; phenolic compounds; polyphenol oxidase*

## INTRODUCTION

Polyphenolic compounds in the diet enhance the stability of low-density lipoprotein (LDL) to oxidation, and evidence exists that LDL oxidation plays a significant role in atherosclerosis and coronary heart disease (Steinberg et al., 1989). The role of natural antioxidants in fruits and vegetables in the delay of the onset of atherogenesis and pathogenesis has received considerable attention (Steinberg et al., 1989). The phenolic compounds in red wine and grape extracts were shown to be very effective in inhibiting the oxidation of LDL in vitro (Frankel et al., 1993, 1995; Kanner et al., 1994; Teissedre et al., 1996; Meyer et al., 1997). The phenolic compounds in commercial grape juices also were shown to have an antioxidant effect on human LDL (Frankel et al., 1998).

Peaches are an important economic fruit that are utilized as processed sauce, slices, and halves. Recent studies of the antioxidant composition of clingstone peaches (Barrett and Garcia, unpublished work) revealed that phenolic compounds serve as major sources of potential antioxidants. Phenolic compounds also were cited recently as inhibitors of brown rot in clingstone peaches (Gradziel and Wang, 1993).

Conversely, some phenolic compounds are well-known as substrates for undesirable browning reactions, which are catalyzed by the enzyme polyphenol oxidase (PPO) (Luh et al., 1967). For this reason, it was previously thought that breeding peaches with high phenolic compound concentrations might be undesirable. However, in light of their potential role as antioxidants for the prevention of human heart disease and cancer, as well as their inhibitory effect on brown rot in clingstone

peaches, phenolic compounds may be viewed as positive constituents. Using high-phenolic-containing clingstone peaches for breeding may be limited if the peaches are high in PPO activity. Three components are required for undesirable browning reactions to occur: phenolic compounds, PPO, and oxygen. It may be possible to achieve the positive aspects of phenolic compounds while preventing browning by selecting cultivars that are high in phenolic compounds and low in PPO activity. The purposes of this study were to determine the role of clingstone peach phenolic compounds as antioxidants, to correlate the relative antioxidant activities with individual phenolic compounds, and to measure PPO activity and phenolic compound composition in a selected number of clingstone peach cultivars.

## MATERIALS AND METHODS

**Chemicals.** Catechol, catechin, gallic acid, rutin (quercetin 3-rutinoside), hexanal, and Folin–Ciocalteu phenol reagent were obtained from Sigma Chemical Co., St. Louis, MO. Copper sulfate, sodium carbonate, HPLC-grade acetonitrile, ammonium phosphate, potassium phosphate, sodium phosphate, and orthophosphoric acid were obtained from Fisher Scientific (Fair Lawn, NJ). Polyethylene glycol was purchased from Matheson Coleman & Bell, Norwood, OH.

**Sample Preparation.** Eight clingstone peach cultivars (Andross, Ross, Bolinha, Walgant, Halford, Kakamas, Corona, and breeding line 18-8-23) were harvested at optimum maturity on the basis of skin color from the University of California orchard in Winters, CA, during the summer of 1997. The fruit was washed with tap water and air-dried. Three extracts, that is, peel, flesh, and whole peach, were analyzed in triplicate for antioxidant activity and phenolic compound composition. The peel fraction was removed with a sharp knife and immediately frozen with liquid nitrogen, placed in a plastic freezer bag, and stored at  $-20^{\circ}\text{C}$  until evaluation. The flesh fraction consisted of a peeled wedge, representing approximately one-eighth of the fruit, which was chopped into small

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pieces, frozen in liquid nitrogen, and also stored at  $-20^{\circ}\text{C}$ . The whole fraction was prepared in the same way as the flesh fraction, but the peel was not removed.

**Extraction of Phenolic Compounds.** Phenolic compounds were extracted by blending 10 g of peach flesh fraction with 50 mL of 80% aqueous methanol (MeOH) at maximum speed in a Waring blender for 1 min. The homogenate was filtered through Whatman No.1 paper in a Büchner funnel. The remaining residue in the blender was rinsed with another 30 mL portion of 80% MeOH and filtered. The filtrate was combined, and the methanol was removed by rotary evaporation under vacuum at  $40^{\circ}\text{C}$ . The final volume was made up to 17 mL with doubly distilled water and filtered through a 45 mm polytetrafluoroethylene (PTFE) filter prior to analysis for total phenols, LDL oxidation, and phenolic compound composition by HPLC.

**Analyses of Phenolic Compounds.** Total phenol content in the peach extracts was determined according to the Folin–Ciocalteu procedure (Singleton and Rossi, 1965), and the results were expressed as gallic acid equivalents (GAE). Phenolic compound composition of peach extracts was analyzed by HPLC, using three filtered buffer systems: buffer A contained 50 mM ammonium phosphate at pH 2.6; buffer B contained 80% HPLC-grade acetonitrile and 20% buffer A; and buffer C contained 200 mM orthophosphoric acid at pH 1.5 (Lamuela-Raventos and Waterhouse, 1994). Phenolic compounds were divided into four classes by each buffer according to spectral properties and quantified with the following standard compounds: hydroxycinnamates, detected at 316 nm and expressed as chlorogenic acid equivalents; anthocyanins, detected at 520 nm and expressed as malvin equivalents; flavan-3-ols, detected at 280 nm and expressed as catechin equivalents; and flavanols, detected at 365 nm and expressed as rutin equivalents.

**Inhibition of Human LDL Oxidation.** LDL was prepared according to the procedure described previously (Frankel et al., 1992). Plasma LDL was isolated by sequential density ultracentrifugation and dialyzed overnight with deoxygenated phosphate buffer (pH 7.4) at  $4^{\circ}\text{C}$  to remove EDTA. The protein concentration in LDL was determined according to the Lowry assay, using a Sigma Protein Lowry kit. LDL was diluted with phosphate buffer to a final concentration of 1 mg of LDL protein/mL prior to oxidation analysis.

The inhibition of copper-catalyzed LDL oxidation ( $80\text{ }\mu\text{mol/L}$  copper sulfate) by peach phenolic compounds was assayed by measuring hexanal production using static headspace gas chromatography using a capillary DB-1701 column (J&W Scientific, Folsom, CA) (Frankel et al., 1992). Aqueous peach extracts were tested at concentrations of  $10\text{ }\mu\text{M}$  GAE. The percent inhibition of LDL oxidation was determined by the formula

$$\% \text{ inhibition} = [(C - S)/C] \times 100$$

where  $C$  is the hexanal produced by the control and  $S$  is the hexanal produced by Cu-catalyzed oxidation (Frankel et al., 1995). Relative percent inhibition of LDL oxidation was calculated by multiplying the percent inhibition at  $10\text{ }\mu\text{M}$  GAE by the dilution factor from the extract used in the LDL oxidation analysis and using the highest percent inhibition value as 100% (Frankel et al., 1995). All samples were run in duplicate.

**PPO Assay.** PPO activity and specific activity were assayed in the peel and flesh fractions only. Acetone powders were prepared by blending frozen peach fractions with 2 volumes of cold acetone ( $-13^{\circ}\text{C}$ ) and 5% polyethylene glycol (MW of 3000–3700) as a phenolic compound scavenger for 1.5 min and filtering under vacuum. The residues were blended again with additional cold acetone, filtered, and washed several times with cold acetone (Jen and Kahler, 1974). The acetone powders so obtained were dried overnight in a desiccator to remove residual acetone and stored at  $-20^{\circ}\text{C}$ .

For measurement of PPO activity, the acetone powders were suspended in 40 volumes (w/v) of 0.1 M sodium phosphate buffer (pH 6.2) containing 1 M NaCl and stirred for 30 min at

room temperature. The suspensions were centrifuged at  $1200g$  for 20 min and filtered through glass wool, and the filtrate was used as the source of enzyme for the assay. PPO activity was determined in triplicate by measuring the increase in absorbency at 420 nm with a spectrophotometer (Shimadzu, Ramsey, NJ). The sample contained 2.9 mL of a mixed solution of 0.1 M sodium phosphate buffer (pH 6.2), 0.3 M catechol (1:1), and 0.1 mL of enzyme extract. One unit of PPO activity was defined as the change in absorbance per minute per milliliter of acetone powder extraction. The protein content of the acetone powder was determined by using the Bradford dye-binding protein assay (Bradford, 1976). Specific activity is the activity per milligram of protein.

**Color Determination.** Eight fruits were randomly selected from each cultivar for color analysis.  $L^*$  (lightness),  $a^*$  (green to red),  $b^*$  (yellow to blue), and hue angle ( $\tan^{-1} b/a$ ) color values were measured using a Minolta CR-200 (Ramsey, NJ) color meter, calibrated with both white and orange reference tiles. Approximately 5 mm thick slices were cut from four sides of each peach, and the cut slice was evaluated immediately using three to five replicate color measurements per slice. Replicate measurements for each slice were averaged, and the four slice values were averaged for each peach.

**Statistical Analysis.** Differences in antioxidant activities and PPO activity were tested by one-way ANOVA. Means were separated by Duncan's and LSD multiple-range test. The significance level was  $P < 0.05$ .

## RESULTS AND DISCUSSION

**Total Phenols.** The levels of total phenolic compounds in the three extracts of clingstone peach cultivars varied widely (Table 1). Peach peels contained 2–2.5 times the concentration of total phenolic compounds as compared to flesh and whole extracts. The concentration of total phenols varied from 467 to 801 mg/kg in flesh extracts, from 415 to 765 mg/kg in whole extracts, and from 877 to 1896 mg/kg in peel extracts. The Halford cultivar had the greatest amount of total phenols in the whole, flesh, and peel extracts, whereas Andross and Walgant flesh and whole extracts were lowest in total phenol content. In commercially processed peach products, peels are generally removed from the peach and discarded. It may be of interest to utilize waste peach peels as a source of antioxidants. In this study, the highest amount of total phenolic compounds in the flesh, which is the primary portion of the peach consumed, was found in Halford followed by Kakamas.

**Antioxidant Activity.** The antioxidant activity of three extracts for the inhibition of LDL oxidation was determined at  $10\text{ }\mu\text{M}$  GAE (Table 1). The percentage inhibition of LDL oxidation varied from 44.3 to 74.7% in flesh extracts, from 45.2 to 72.7% in whole extracts, and from 32.2 to 85.7% in peel extracts. The Walgant cultivar had high antioxidant activity in all three extracts, whole (73.1%), flesh (72.7%), and peel (85.7%). Kakamas and breeding line 18-8-23 also had relatively high antioxidant activities in whole and flesh extracts. Halford had the lowest antioxidant activity among the cultivars in all three extracts.

Cultivars may be grouped by significant differences in percentage inhibition into high and low classes. In the whole extract, Kakamas, 18-8-23, Andross, and Walgant fit into the high class, whereas Ross, Corona, Halford, and Bolinha may be regarded as low in antioxidant activity. Walgant, 18-8-23, Bolinha, and Kakamas had significantly higher activity in the flesh extract than Andross, Ross, Corona, or Halford. Finally, antioxidant activity in the peel extract was significantly higher in Walgant, Bolinha, 18-8-23, and Corona. The

**Table 1. Total Phenolic Content, Antioxidant Activity, and PPO Activity in Clingstone Peaches**

cultivar	fraction	total phenol <sup>a</sup> (mg GAE/kg)	% inhibition 10 $\mu$ M GAE <sup>b</sup>	relative % inhibition 10 $\mu$ M GAE <sup>b</sup>	PPO activity units	specific activity (units/mg of protein)
Andross	whole	498 $\pm$ 23.1	74.7 $\pm$ 1.3 <sup>ab</sup>	35.3 $\pm$ 1.1		
Bolinha		598 $\pm$ 14.1	44.3 $\pm$ 5.8 <sup>c</sup>	24.8 $\pm$ 3.6		
Corona		597 $\pm$ 15.4	53.4 $\pm$ 5.8 <sup>c</sup>	29.9 $\pm$ 3.6		
Halford		801 $\pm$ 11.6	44.7 $\pm$ 5.8 <sup>c</sup>	33.1 $\pm$ 4.7		
Kakamas		582 $\pm$ 15.4	84.5 $\pm$ 3.4 <sup>a</sup>	48.9 $\pm$ 2.5		
Ross		545 $\pm$ 1.3	58.1 $\pm$ 3.2 <sup>bc</sup>	29.8 $\pm$ 2.0		
Walgant		467 $\pm$ 25.0	73.1 $\pm$ 16.0 <sup>ab</sup>	32.4 $\pm$ 6.7		
18-8-23		516 $\pm$ 4.5	80.3 $\pm$ 1.4 <sup>a</sup>	39.0 $\pm$ 1.2		
Andross	flesh	415 $\pm$ 5.8	63.7 $\pm$ 0.2 <sup>b</sup>	25.4 $\pm$ 0.4	13.7 $\pm$ 0.6 <sup>a</sup>	23.45
Bolinha		585 $\pm$ 5.1	70.2 $\pm$ 1.5 <sup>a</sup>	38.6 $\pm$ 1.3	10.8 $\pm$ 1.2 <sup>c</sup>	24.58
Corona		423 $\pm$ 6.4	47.8 $\pm$ 8.6 <sup>d</sup>	19.4 $\pm$ 3.3	9.50 $\pm$ 0.5 <sup>d</sup>	34.95
Halford		765 $\pm$ 2.6	45.2 $\pm$ 2.3 <sup>d</sup>	32.0 $\pm$ 1.3	4.70 $\pm$ 0.1 <sup>e</sup>	17.17
Kakamas		607 $\pm$ 1.3	67.9 $\pm$ 5.1 <sup>ab</sup>	38.6 $\pm$ 2.4	2.00 $\pm$ 0.1 <sup>fg</sup>	4.72
Ross		513 $\pm$ 5.1	56.2 $\pm$ 2.0 <sup>bc</sup>	27.3 $\pm$ 1.3	12.1 $\pm$ 0.2 <sup>b</sup>	31.16
Walgant		425 $\pm$ 1.3	72.7 $\pm$ 3.1 <sup>a</sup>	29.7 $\pm$ 0.9	1.80 $\pm$ 0.1 <sup>g</sup>	7.11
18-8-23		479 $\pm$ 8.3	70.7 $\pm$ 6.1 <sup>a</sup>	32.2 $\pm$ 3.2	2.90 $\pm$ 0.6 <sup>f</sup>	12.10
Andross	peel	1337 $\pm$ 0.0	44.8 $\pm$ 2.6 <sup>d</sup>	56.3 $\pm$ 4.0	2.90 $\pm$ 0.3 <sup>a</sup>	15.69
Bolinha		1210 $\pm$ 3.2	77.9 $\pm$ 1.6 <sup>b</sup>	91.0 $\pm$ 0.8	8.90 $\pm$ 0.8 <sup>b</sup>	39.07
Corona		877 $\pm$ 10.9	69.8 $\pm$ 6.6 <sup>c</sup>	58.8 $\pm$ 6.3	11.5 $\pm$ 0.3 <sup>a</sup>	45.71
Halford		1896 $\pm$ 5.1	32.2 $\pm$ 3.2 <sup>e</sup>	56.4 $\pm$ 6.4	5.70 $\pm$ 0.2 <sup>c</sup>	23.80
Kakamas		1263 $\pm$ 4.5	49.4 $\pm$ 6.6 <sup>d</sup>	59.8 $\pm$ 8.7	3.40 $\pm$ 0.2 <sup>d</sup>	23.26
Ross		1394 $\pm$ 3.2	47.5 $\pm$ 1.0 <sup>d</sup>	62.0 $\pm$ 2.0	11.7 $\pm$ 1.0 <sup>a</sup>	65.92
Walgant		1219 $\pm$ 7.7	85.7 $\pm$ 1.1 <sup>a</sup>	100 $\pm$ 0.0	3.20 $\pm$ 0.4 <sup>d</sup>	10.35
18-8-23		1470 $\pm$ 1.9	63.0 $\pm$ 0.9 <sup>c</sup>	87.3 $\pm$ 2.4	6.20 $\pm$ 0.2 <sup>c</sup>	34.58

<sup>a</sup> GAE, gallic acid equivalents by Folin–Ciocalteu method. <sup>b</sup> Antioxidant activities by LDL oxidation assay. Values in the same column and same fraction followed by the same letter are not significantly different at  $P < 0.05$ .

flesh extract is the most important for consumption purposes; therefore, promotion of the Walgant, 18-8-23, Bolinha, and Kakamas cultivars may be desirable.

Composites of whole, flesh, and peel extracts of all cultivars showed no significant difference in antioxidant activity. To compare antioxidant activities of the peaches, the relative inhibition of LDL oxidation was calculated on the basis of their total phenol contents by multiplying the percent inhibition values by the dilution factor used in the assay (Frankel et al., 1995). The relative inhibition of LDL oxidation in peels was thus much higher than that in the flesh and whole extracts. The whole extracts of Kakamas and 18-8-23 showed the greatest relative inhibition of LDL oxidation (48.9 and 39.0%, respectively) followed by whole Andross, Halford, and Walgant extracts (35.3, 33.1, and 32.4%, respectively). The relative inhibition of LDL oxidation in peach flesh extracts decreased in the following order: Bolinha, Kakamas, 18-8-23 and Halford > Walgant, Ross, and Andross > Corona. Walgant peel had the highest activity (100% inhibition), whereas Andross and Halford peel had the lowest inhibition (56.3 and 56.4% inhibition, respectively).

**Phenolic Compound Composition.** HPLC analysis showed that hydroxycinnamates and flavan-3-ols were significant components of peach extracts (Table 2). Hydroxycinnamates consisted primarily of neochlorogenic acid, which ranged from 11.5 to 80.9 mg/kg, and chlorogenic acid, ranging from 23.9 to 470.5 mg/kg. Chlorogenic acid was present in the highest concentration in the eight cultivars of the clingstone peaches. This result is in agreement with previous reports that chlorogenic acid was the dominant phenolic compound in clingstone peaches (Luh et al., 1967; Barrett and Garcia, unpublished work). Much higher levels of chlorogenic acid were found in the peel fraction, particularly in the Halford, Ross, and 18-8-23 cultivars. Bolinha flesh contained the highest amount of chlorogenic acid of all flesh extracts.

Malvin was identified as the main anthocyanin compound in this study. Anthocyanins are responsible for the red color in peaches; therefore, malvin was present primarily in the peel extracts and absent in all flesh extracts. The peel of Halford, Bolinha, and 18-8-23 cultivars appeared to be very green, and anthocyanins were not detected in these cultivars.

The major flavan-3-ol compounds included procyanidin B1, ranging from 9 to 279 mg/kg, and catechin, ranging from 8 to 434 mg/kg. These two compounds were also found to be more abundant in the peel, as compared to the flesh and whole extracts. The peel of cultivar 18-8-23 contained the highest level of procyanidin B1, and Halford peel had the highest level of catechin. The catechin concentration was second only to chlorogenic in the clingstone peach cultivars evaluated. Although catechin is a good antioxidant for the prevention of lipid oxidation in vitro (Teissedre et al., 1996), it is also an excellent substrate for fruit PPO (Lee et al., 1990). Chlorogenic acid is an even better PPO substrate than catechin.

Rutin and isoquercetin were the primary flavonol compounds found in clingstone peaches. As with malvin, rutin and isoquercetin were present in low levels in all peach extracts and were found mainly in the peels rather than the flesh.

The relative LDL inhibition obtained for all peach samples correlated with total phenols and individual phenolic compounds as follows:  $r = 0.76$  for gallic acid equivalents,  $r = 0.79$  for procyanidin B1,  $r = 0.85$  for rutin,  $r = 0.65$  for catechin, and  $r = 0.66$  for chlorogenic acid. No correlation was found between relative LDL inhibition and anthocyanin, isoquercetin, or neochlorogenic acid contents. In contrast, the relative LDL antioxidant activity of grape extracts correlated with anthocyanins, but not with hydroxycinnamates (Frankel et al., 1995, 1997). A higher correlation coefficient of relative LDL antioxidant activity with total phenols was found in the grape extracts ( $r = 0.89$ ) and wine ( $r =$



**Table 2. Phenolic Composition of Clingstone Peach Extracts (Milligrams per Kilogram)**

cultivar	fraction	hydroxycinnamates		anthocyanins	flavonols		flavan-3-ols	catechin
		neochlorogenic acid <sup>a</sup>	chlorogenic acid	malvin	rutin	isoquercetin <sup>b</sup>	procyanidin B1 <sup>c</sup>	
Andross	whole	19.3	34.9	0.0	0.0	0.0	13.6	10.9
Bolinha		51.1	73.8	0.0	5.6	6.3	66.8	61.1
Corona		61.6	86.1	17.8	2.6	4.9	74.7	112.9
Halford		39.3	60.9	6.1	0.0	0.0	47.8	49.7
Kakamas		36.3	56.7	0.0	0.0	0.0	83.3	89.2
Ross		48.3	61.7	0.0	0.0	0.0	28.6	25.0
Walgant		18.3	46.2	0.0	0.0	0.0	22.0	17.7
18-8-23		42.9	66.1	0.0	0.0	0.0	49.3	89.7
Andross	flesh	41.5	70.3	0.0	0.0	0.0	20.0	16.0
Bolinha		78.3	109.4	0.0	0.0	0.0	53.9	44.5
Corona		56.8	60.0	0.0	0.0	0.0	56.6	53.0
Halford		28.7	38.7	0.0	0.0	0.0	53.9	22.7
Kakamas		31.9	77.9	0.0	0.0	0.0	64.6	
Ross		37.9	34.8	0.0	0.0	0.0	19.5	17.6
Walgant		11.5	23.9	0.0	0.0	0.0	9.1	8.3
18-8-23		43.4	52.6	0.0	0.0	0.0	50.2	90.8
Andross	peel	32.0	202.7	75.6	28.9	89.6	95.5	69.2
Bolinha		29.3	164.4	45.8	26.6	54.7	225	246
Corona		40.8	200.3	34.8	40.8	78.7	87.8	77.8
Halford		66.7	470.5	0.0	27.7	90.2	270	434
Kakamas		22.5	178.8	0.0	19.9	41.7	69.6	53.1
Ross		65.6	325.1	0.0	81.3	96.9	255	177
Walgant		45.1	186.9	38.8	16.8	38.0	184	166
18-8-23		80.9	323.4	0.0	7.1	42.9	279	301

<sup>a</sup> Neochlorogenic acid as chlorogenic acid equivalents (CLE). <sup>b</sup> Isoquercetin as rutin equivalents (RUE). <sup>c</sup> Procyanidin B1 as catechin equivalents (CCE).

0.94). The antioxidant activity toward LDL oxidation thus may be widely distributed among the phytochemical phenolic constituents. Also, the amount of phenolic compounds detected by HPLC represented only half of or less than the amount of total phenols obtained from the Folin–Ciocalteu method for most of the cultivars. Antioxidant activity may be contributed by some of the phenolic compounds in peach extract that were not separated and identified by HPLC and other compounds such as ascorbic acid.

**PPO and Specific Activities.** PPO activity in extracts from acetone powders of both flesh and peel also varied widely among the different cultivars (Table 1), showing significant differences ( $P < 0.05$ ) among all of the cultivars. The Walgant cultivar had the lowest PPO activity in both flesh and peel extracts, followed by Kakamas and Halford. A similar result was reported by Kader (1980) for these cultivars. The PPO activity in the flesh extract of the eight cultivars increased in the following order: Walgant, Kakamas, 18-8-23, Halford, Corona, Bolinha, Ross, and Andross. According to Lee et al. (1990), the degree of actual browning of an individual peach cultivar correlates to its PPO activity. Peach cultivars with lower PPO activity show a lower rate of browning. Therefore, the cultivars Walgant, Kakamas, and 18-8-23 had the lowest browning potentials.

The PPO activity of the peel was higher than flesh activity for most of the cultivars (six of eight) and also varied greatly among different cultivars, with highest activity in the peel of Ross and Corona cultivars. It was not possible to predict relative flesh PPO activity of any particular cultivar from peel activity, or vice versa. Andross flesh had the highest PPO activity, whereas its peel extract showed the lowest activity. Both the flesh and peel of Ross had similar PPO activities, whereas Kakamas and Walgant exhibited the same low PPO activity in both flesh and peel.

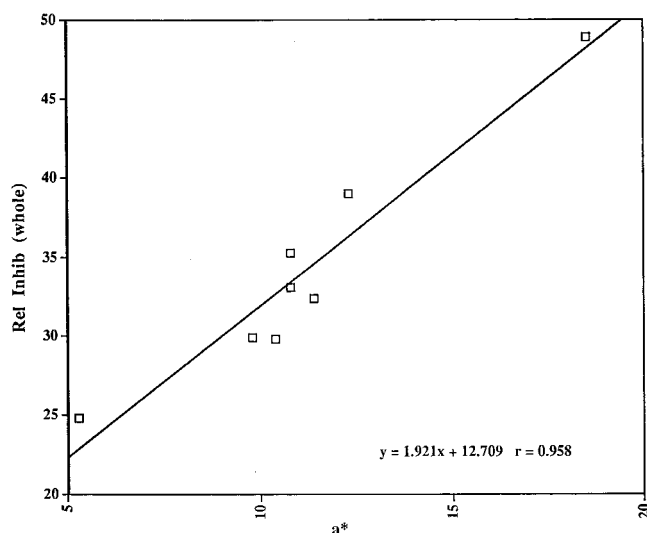
Kakamas had the lowest (4.72 units/mg of protein) and Corona the highest (34.95 units/mg of protein)

**Table 3. Color of Clingstone Peach Cultivars**

cultivar	$L^*$	$a^*$	$b^*$	hue angle
Andross	$50.9 \pm 2.7$	$10.8 \pm 4.2$	$56.7 \pm 3.4$	$79.2 \pm 3.0$
Bolinha	$67.5 \pm 2.5$	$5.3 \pm 2.5$	$56.8 \pm 4.4$	$84.7 \pm 2.4$
Corona	$66.5 \pm 1.9$	$9.8 \pm 2.2$	$54.2 \pm 4.3$	$49.8 \pm 2.4$
Halford	$64.3 \pm 3.5$	$10.8 \pm 2.8$	$52.4 \pm 5.7$	$78.3 \pm 1.9$
Kakamas	$63.1 \pm 1.6$	$18.5 \pm 2.2$	$56.0 \pm 2.7$	$71.7 \pm 2.1$
Ross	$53.4 \pm 3.3$	$10.4 \pm 2.2$	$52.1 \pm 3.8$	$78.7 \pm 2.1$
Walgant	$49.9 \pm 1.4$	$11.4 \pm 2.9$	$53.7 \pm 2.1$	$78.0 \pm 2.4$
18-8-23	$66.9 \pm 1.8$	$12.3 \pm 1.7$	$49.1 \pm 4.0$	$76.0 \pm 1.7$

specific activity of the flesh extracts (Table 1). Walgant had the lowest specific activity (10.35 units/mg of protein) of the peel extracts. Ross exhibited the highest peel specific activity (65.92 units/mg of protein) followed by Corona (45.71 units/mg of protein) and Bolinha (39.07 units/mg of protein). Interestingly, the specific activities of all the peel extracts were higher values than the activity of the flesh extracts, except for the Andross cultivar. This difference may be attributed to the acetone powder preparation procedure. Although the same amounts of flesh and peel were extracted, a much greater volume of acetone powder was obtained from the peel than from the flesh. The protein concentration in the peel acetone powder was much lower than that in the flesh powder, resulting in a higher specific activity in the peels.

**Color Determination.** There were significant differences ( $P < 0.05$ ) among the eight cultivars in all color values of the fruit flesh measured, that is,  $L^*$ ,  $a^*$ , and  $b^*$  values and hue angle (Table 3). Hue angle is an indication of the overall color value perceived by the eye, with values of red purple equal to  $0^\circ$  and yellow equal to  $90^\circ$ . The Bolinha cultivar was the lightest (highest  $L^*$ ) and most yellow (hue angle) in color, whereas Kakamas was the most red by both  $a^*$  (highest) and hue angle measurements. Walgant and Andross were the darkest (lowest  $L^*$ ), but there were no significant differences ( $P > 0.05$ ) in lightness ( $L^*$ ) among Bolinha, Corona, Halford, and 18-8-23. Andross, Halford, and Ross had similar degrees of redness ( $P < 0.05$ ) according



**Figure 1.** Correlation between peach redness ( $a^*$ ) and relative percent inhibition of LDL oxidation.

to both  $a^*$  and hue angle values. The  $b^*$  values for the cultivars investigated were relatively consistent, indicating that these cultivars had about the same degrees of yellowness.

Results of hue angle measurement were in agreement with a previous study of the cultivars Halford, Ross, Corona, Kakamas, and 18-8-23 (Barrett and Garcia, unpublished work), showing Corona to have the highest hue angle (most yellow) and Kakamas the lowest (most red).

Peach color was determined to evaluate the potential relationship to antioxidant activity for inhibition of LDL oxidation. Although there was a strong correlation ( $r = 0.96$ ) found between  $a^*$  value (redness) and percent relative inhibition in whole peach extracts (Figure 1), the anthocyanin content did not correlate well with percent relative inhibition. This high correlation may be attributed, therefore, to colored constituents other than anthocyanins.

**Conclusions.** All eight clingstone peach cultivars demonstrated good antioxidant activity in the inhibition of LDL oxidation. Significant differences were found in individual phenolic compounds and PPO activities of all cultivars. The Kakamas, Walgant, and 18-8-23 cultivars showed the highest percentages of inhibition of LDL oxidation and the lowest PPO activities. Because Corona had the lowest percentage inhibition of LDL oxidation and a relatively high PPO activity, it would not be a good selection for consumption. Kakamas and 18-8-23 were the most red and Walgant the darkest (low  $L^*$ ) of all of the cultivars investigated. A strong correlation was observed between redness ( $a^*$ ) and relative percent inhibition in whole peach extracts.

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