There is some evidence that chronic diseases, such as cancer and cardiovascular disease, may occur as a result of oxidative stress. Apple peels have high concentrations of phenolic compounds and may assist in the prevention of chronic diseases. Millions of pounds of waste apple peels are generated in the production of applesauce and canned apples in New York State each year. We proposed that a valuable food ingredient could be made using the peels of these apples if they could be dried and ground to a powder without large losses of phytochemicals. Rome Beauty apple peels were treated with citric acid dips, ascorbic acid dips, and blanches before being oven-dried at 60 °C. Only blanching treatments greatly preserved the phenolic compounds, and peels blanched for 10 s had the highest total phenolic content. Rome Beauty apple peels were then blanched for 10 s and dried under various conditions (oven-dried at 40, 60, or 80 °C, air-dried, or freeze-dried). The air-dried and freeze-dried apple peels had the highest total phenolic, flavonoid, and anthocyanin contents. On a fresh weight basis, the total phenolic and flavonoid contents of these samples were similar to those of the fresh apple peels. Freeze-dried peels had a lower water activity than air-dried peels on a fresh weight basis. The optimal processing conditions for the ingredient were blanching for 10 s and freeze-drying. The process was scaled up, and the apple peel powder ingredient was characterized. The total phenolic content was 3342 ± 12 mg gallic acid equivalents/100 g dried peels, the flavonoid content was 2299 ± 52 mg catechin equivalents/100 g dried peels, and the anthocyanin content was 169.7 ± 1.6 mg cyanidin 3-glucoside equivalents/100 g dried peels. These phytochemical contents were a significantly higher than those of the fresh apple peels if calculated on a fresh weight basis (p < 0.05). The apple peel powder had a total antioxidant activity of 1251 ± 56 µmol vitamin C equivalents/g, similar to fresh Rome Beauty peels on a fresh weight basis (p > 0.05). One gram of powder had an antioxidant activity equivalent to 220 mg of vitamin C. The freeze-dried apple peels also had a strong antiproliferative effect on HepG2 liver cancer cells with a median effective dose (EC50) of 1.88 ± 0.01 mg/mL. This was lower than the EC50 exhibited by the fresh apple peels (p < 0.05). Apple peel powder may be used in a various food products to add phytochemicals and promote good health.

INTRODUCTION

The leading causes of death in the United States are cardiovascular diseases and cancer. Both types of diseases are thought to be partially the result of oxidative stress, which can lead to damage of biomolecules. It has been hypothesized that an increase in dietary antioxidants can reduce oxidative stress and prevent chronic diseases. Fruits and vegetables contain many antioxidant compounds, including phenolics, thiols, carotenoids, tocopherols, and glucosinolates, that may protect against cardiovascular diseases and cancer through a variety of mechanisms. Such findings have lead to the National Research Council (NRC) to recommend consuming five or more servings of fruits and vegetables a day.

Apples are very significant part of the diet. Hertog et al. (1) determined that apples are the third largest contributors of flavonols in the Dutch diet behind tea and onions. In Finland, along with onions, they are the top contributors (2). Twenty-two percent of the fruit phenolics consumed in the United States are from apples, making them the largest source (3). Consumption of apples has been linked to the prevention of chronic disease in a number of studies. Apple intake has been negatively associated with lung cancer incidence in at least two studies (2, 4). It has also been related to reduced cardiovascular disease. Coronary and total mortality (5), symptoms of chronic obstructive pulmonary disease (6), and risk of thrombotic stroke (7) have all been inversely associated with apple consumption.

KEYWORDS: Phenolics; flavonoids; apple; antioxidant activity; cancer; value-added
Apples are a good source of phenolic compounds (8). The total extractable phenolic content has been investigated and ranges from 110 to 357 mg/100 g fresh apple (9, 10). It is known that the concentration of total phenolic compounds is much greater in the peel of apples than in the flesh (11–13). The nature and distribution of these phytochemicals between the flesh and the peel of the apple is also different. Among others, the flesh contains catechins, procyanidins, phloridzin, phloretin glycosides, caffeic acid, and chlorogenic acid; the peel possesses all of these compounds and has additional flavonoids not found in the flesh, such as quercetin glycosides and cyanidin glycosides (11, 13–15). It has been noted in other fruits and vegetables that the peels have high phytochemical concentrations and/or high antioxidant activity (16–21).

Previously, our research group found that peeled and unpeeled apples have high antioxidant activity and inhibit the growth of human cancer cells in vitro (8). The antioxidant and antiproliferative activities of unpeeled apples were greater than that of peeled apples. This suggested that the peels alone would be even more effective. Apple peels are a waste product of applesauce and canned apple manufacture, and if they showed potential to improve health when consumed, their utilization should be investigated. Industrially, apples are peeled both by using a hot caustic method and mechanically. The National Agriculture Statistics Service (NASS) (22) reported that 267 million pounds of apples were processed in applesauce and canned apple production in New York State in 2001, and we estimated that 20 million pounds of peels were generated. The apple peels are typically used for nonvaluable purposes. They, along with the core materials, are often pressed to make juice or vinegar, pressed into a cake for livestock feed, or used as fertilizer. Sometimes they are used as a source of pectin. We definitively showed that apple peels from apple varieties used in applesauce manufacture have higher antioxidant activities and greater inhibitory effects on cancer cells than the flesh or flesh + peel of the same apples (37). The extent of these effects seemed highly dependent on apple variety. Out of the four varieties we examined (Rome Beauty, Cortland, Idared, and Golden Delicious), Rome Beauty apple peels appeared to be the best candidate to continue working with, as they had the highest total phenolic and flavonoid contents and most antiproliferative activity. The Rome Beauty apple variety is popular in New York State and is ranked sixth in production (22). It also has excellent cooking and baking characteristics and is a commonly used variety in applesauce and canned apple products.

Therefore, the object of this study was to develop a value-added ingredient from Rome Beauty apple peels. In order for the ingredient to be nonperishable and easily used, the apple peels were dried and ground to a powder. The challenge was to minimize the oxidation and destruction of the antioxidant compounds present in the peels. Pretreatments involving citric acid dips, ascorbic acid (vitamin C) dips, and blanching were evaluated for their effectiveness in preserving the phytochemical content of the peels. The best drying conditions were then determined. Total phenolic content, flavonoid content, anthocyanin content, and water activity of the dried peels were measured as markers in the evaluation of the best processing methods. The antioxidant activity and antiproliferative effects of the manufactured ingredient were quantified to better characterize it.

MATERIALS AND METHODS

Chemicals. Sodium nitrite, (+)-catechin, Folin–Ciocalteu reagent, citric acid, ascorbic acid, and α-keto-γ-methylbutyric acid (KMB) were purchased from Sigma Chemical Company (St. Louis, MO). Sodium carbonate, sodium hydroxide, ethanol, aceton, and potassium phosphate were obtained from Mallincrodt (Paris, KY). Aluminum chloride, potassium chloride, and sodium acetate were purchased from Fisher Scientific (Pittsburgh, PA) and gallic acid from ICN Biomedical Inc. (Costa Mesa, CA). 2,2’-Azobisisobutyronitrile (AABP) was obtained from Wako Chemicals (Richardson, VA). The HepG2 cells were obtained from the American Type Culture Collection (ATCC) (Rockville, MD) and the MTS-based Cell Titer 96 nonradioactive cell proliferation assay was from Promega (Madison, WI). Williams Medium E (WME) and fetal bovine serum (FBS) were purchased from Gibco Life Technologies (Grand Island, NY).

Samples. Rome Beauty apples were obtained from Red Jacket Orchards (Geneva, NY). The apples were kept in modified atmosphere storage until purchased. The apples were washed and dried before experimentation.

Determination of Pretreatment Method. Five randomly selected apples were peeled using an Apple Master apple parer. The peels were left untreated or were treated with citric acid dips, ascorbic acid dips, or boiling water dips (blanched). For the citric acid treatments, the five apple peels (approximately 75 g) were dipped for 3 min in 500 mL of 0.1, 0.2, 0.5, and 0.8% citric acid solutions and then drained. The ascorbic acid treatments involved dipping the peels in 0.1, 0.2, and 0.5% ascorbic acid for 3 min and draining. The blanching times were 1, 10, 20, and 30 s. After blanching, the peels were dipped in a cooling bath of distilled water for 10 s. The peels were placed in stainless steel wire baskets in a VWR 1325F Mechanical Convection Oven (VWR Scientific Products, Bridgeport, NJ) at 60 °C and remained there until they reached constant weight (approximately 13 h). This temperature was chosen because earlier trials showed it to be quite damaging to phenolic compounds. Thus, any benefit from the treatments should be easy to see. The dried apple peels were ground in a coffee grinder and stored at −40 °C until use. Each treatment (e.g., 0.1% citric acid) was performed in triplicate with different apple peels used each time.

Determination of Drying Method. Five randomly selected apples were peeled using an Apple Master apple parer. The apple peels were treated with the best method from above. In addition, peels from another five apples were left untreated for comparison. The apple peels were oven-dried in a mechanical convection oven at 40, 60, or 80 °C; air-dried in a fume hood, or freeze-dried. The oven-dried peels were placed in stainless steel wire baskets in a VWR 1325F Mechanical Convection Oven (VWR Scientific Products, Bridgeport, NJ) at the appropriate temperature and remained there until the peels reached a constant weight. The air-dried peels were placed in stainless steel wire baskets in a fume hood and dried to constant weight. The freeze-dried peels were dried in a Virtis 100 SRC freeze-dryer in the pilot plant in the Department of Food Science, Cornell University. The condenser temperature was −40 °C, the shelf temperature was set at 25 °C, and the vacuum was 150 µm for 72 h. The dried apple peels were ground in a coffee grinder and stored at −40 °C until use. Each drying trial was performed in triplicate.

Extraction of Dried Apple Peels. The phytochemicals of dried apple peels were extracted by a method similar to that reported previously by our laboratory (8, 23). Briefly, the dry equivalent of 25 g of apple peels was combined with 200 g of chilled 80% acetone or chilled 80% ethanol solution and homogenized for 5 min using a Virtis 45 homogenizer. The slurry was filtered through Whatman No. 1 filter paper in a Buchner funnel under vacuum. The filter cake was washed twice with 15 mL of the acetone or ethanol solution. The filtrate was recovered and evaporated.

Extraction of Fresh Apple Peels. The phytochemicals of apples were extracted by a method similar to that reported previously by our laboratory (8, 23). Briefly, 25 g of apple peels were blended with 200 g of chilled 80% acetone or chilled 80% ethanol solution in a Waring blender for 5 min. The sample was then homogenized for 3 min using a Virtis 45 homogenizer. The slurry was filtered through Whatman No. 1 filter paper in a Buchner funnel under vacuum. The solids were scraped into 150 g of 80% acetone or ethanol and homogenized again for 3 min before filtering. The filtrate was recovered and evaporated.
using a rotary evaporator at 45 °C until less than 10% of the initial volume remained. The extract was made up to 50 mL with distilled water and frozen at −40 °C until analysis. The extracts were made in triplicate.

**Determination of Total Phenolic Content.** The total phenolic contents of the apple peel samples were measured using a modified colorimetric Folin–Ciocalteu method (23, 24). Ethanol extracts were used in the analysis of apple peel treatments, while acetone extracts were used for evaluation of drying conditions and the scaled-up product. Volumes of 0.5 mL of deionized water and 0.125 mL of a known dilution of the extract were added to a test tube. Folin–Ciocalteu Reagent (0.125 mL) was added to the solution and allowed to react for 6 min. Then, 1.25 mL of 7% sodium carbonate solution was aliquoted into the test tubes, and the mixture was diluted to 3 mL with deionized water. The color developed for 90 min, and the absorbance was read at 760 nm using a MRX II DYNEX spectrophotometer (DYNEX Technologies, Inc., Chantilly, VA). The measurement was compared to a standard curve of gallic acid concentrations and expressed as mg gallic acid equivalents/100 g ± SD fresh apple peels for the triplicate extracts.

**Determination of Flavonoid Content.** The flavonoid contents of the apple peel samples were measured using a modified colorimetric method (23, 25). Ethanolic extracts were used in the analysis of apple peel treatments, while acetone extracts were used for evaluation of drying conditions and the scaled-up product. A volume of 0.25 mL of a known dilution of extract was added to a test tube containing 1.25 mL of distilled water. To the mixture, 0.075 mL of 5% sodium nitrite solution was added and allowed to stand for 5 min. Then, 0.15 mL of 10% aluminum chloride was added. After 6 min, 0.5 mL of 1 M sodium carbonate was added, and the mixture was diluted with another 0.275 mL distilled water. The absorbance of the mixture at 510 nm was measured immediately using a MRX II DYNEX spectrophotometer (DYNEX Technologies, Inc., Chantilly, VA) and compared to a standard curve of catechin concentrations. The flavonoid content was expressed as mg catechin equivalents/100 g ± SD fresh apple peels for the triplicate extracts.

**Determination of Anthocyanin Content.** Monomeric anthocyanin contents of the apple peels were measured using a spectrophotometric pH differential protocol (26, 27). The apple peel acetone extracts were mixed thoroughly with 0.025 M potassium chloride pH 1 buffer in 1:3 or 1:8 ratio of extract:buffer. The absorbance of the mixture was then measured at 515 and 700 nm against a distilled water blank. The apple peel extracts were then combined similarly with sodium acetate buffer pH 4.5, and the absorbance of these solutions was measured at the same wavelengths. The anthocyanin content was calculated as follows:

\[
\text{total monomeric anthocyanins (mg/100 g fresh peel)} = \frac{A \times \text{MW} \times 1000 (\epsilon \times C)}{A_{515} - A_{700} \times \text{MW}} - \frac{(A_{315} - A_{515}) \times 449.2}{\epsilon}
\]

where \(A\) is absorbance = \(A_{515} - A_{700}\), MW is molecular weight for cyanidin 3-glucoside = 449.2; \(\epsilon\) is the molar absorptivity of cyanidin 3-glucoside = 26,900; and \(C\) is the concentration of the buffer in mg/mL. Anthocyanin content was expressed as mg cyanidin 3-glucoside equivalents/100 g fresh apple peel for the triplicate extracts.

**Determination of Water Activity.** Water activity is the ratio of the partial pressure of water vapor in a product to the saturation vapor pressure at equilibrium. It is a measure of the availability of water to participate in chemical reactions. Water activity was measured at approximately 28 °C using the AquA-Lab CX2 Water Activity Meter (Decagon Devices, Inc., Pullman, WA). Each reading was done in triplicate.

**Quantiﬁcation of Total Antioxidant Activity.** The total antioxidant activity of the apple peels was determined using the total oxyradical scavenging (TOSC) assay (28) modiﬁed in our laboratory (8). In this assay peroxo radicals are formed from 2,2’-azobis(2,4-dimethylvaleric acid) (ABAP) and oxidize \(\alpha\)-ketol-\(\gamma\)-methylbutyric acid (KMB) to form ethylene. The ethylene produced can be measured by gas chromatographic headspace analysis. Total antioxidant activity is measured by the degree of inhibition of formation of ethylene by the apple peel acetone extracts.

The antioxidant activity was measured at 15, 30, 45, and 60 min for four different concentrations of the acetone extracts to determine the TOSC values. The area under the kinetic curve was integrated to calculate the TOSC value at each concentration as follows:

\[
\text{TOSC} = 100 - \left(\frac{\text{SA}}{\text{CA}}\right) \times 100
\]

where /SA is the integrated area from the sample reaction and /CA is the integrated area from the control reaction. The median effective dose (EC\(_{50}\)) was determined for the apple peels from the dose–response curve of apple peel concentration versus TOSC. The antioxidant activity was expressed as micromoles vitamin C equivalents for 1 g of apple peel. The TOSC values were stated as mean ± SD for triplicate samples.

**Determination of Inhibition of HepG2 Cell Proliferation.** The acetone extracts were used to measure the ability of apple peels to inhibit human liver cancer cell proliferation (8, 27). The cell cultures were exposed to various concentrations of the apple peel extracts during a 96-h growth period. The antiproliferative activity of the apple peel extracts was measured by the ability of viable cells to reduce 3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfenyl)-2H-tetrazolium (MTS) to formazan. This product absorbs light at 490 nm and the absorbance was measured using a MRX II DYNEX spectrophotometer (DYNEX Technologies, Inc., Chantilly, VA). At least three replications for each sample were used to determine the inhibitory effect on cell proliferation. The effective median dose (EC\(_{50}\)) was calculated and expressed as mg apple peels/mL ± SD.

**Statistical Analysis.** All data was reported as mean ± standard deviation of three replicates. The results were compared by analysis of variance (ANOVA) using Minitab software (Minitab, Inc., State College, PA). Pairwise multiple comparisons were done by Tukey’s significant difference test with a family error rate of 0.05. Comparisons between blanched and unblanched samples were done by paired t-tests. Comparisons between the fresh apple peels and the scaled-up dried apple peels were done by two-sample t-tests. Differences at \(p < 0.05\) were considered significant.

**RESULTS**

**Pretreatment.** Rome Beauty apple peels were treated with citric acid dips, vitamin C dips, or blanched then oven-dried at 60 °C and evaluated for total phenolic content (Figure 1). Fresh peels had more phenolics than the dried samples with 401.9 ± 11.2 mg gallic acid equivalents/100 g fresh peels (\(p < 0.05\)). The total phenolic content of the peels dipped in 0.1, 0.2, 0.5, and 0.8% citric acid solutions (137.2 ± 14.2, 104.6 ± 40.1, 102.1 ± 0.9, and 107.3 ± 20.9 mg gallic acid equivalents/100 g fresh peels, respectively) were not significantly different from each other (\(p > 0.05\)). Remarkably, the untreated dried peels
**Figure 2.** Total phenolic content of Rome Beauty apple peels dried under various conditions (mean ± SD, n = 3).

(control, **Figure 1**) had higher values than those treated with 0.2, 0.5, and 0.8% citric acid (p < 0.05). The values for those samples dipped in 0.1, 0.2, and 0.5% vitamin C solutions were similar (152.9 ± 37.1, 171.6 ± 1.0, and 218.7 ± 13.6 mg gallic acid equivalent/100 g fresh peel, respectively) (p > 0.05). There was a trend for the phenolic content to increase with vitamin C concentration, though that trend was more likely due to the measurement of the added ascorbic acid, not endogenous phenolic compounds. The total phenolic contents of the apple peels blanched for 1, 10, 20, and 30 s were 218.8 ± 9.3, 258.1 ± 45.2, 253.9 ± 24.6, and 217.1 ± 31.5 mg gallic acid equivalents/100 g fresh peels, respectively. There was no significant difference between these values (p < 0.05). The apple peels blanched for 10 s and 20 s were statistically higher than those that were untreated or subjected to citric acid or vitamin C dips (p < 0.05). The flavonoid content data mimicked the results of the total phenolic assay (data not shown). As the peels blanched for 10 s had the highest total phenolic and flavonoid contents, we chose that as the best pretreatment.

**Drying.** Rome Beauty apple peels were either blanched for 10 s or left untreated, then oven-dried at 40, 60, or 80 °C, air-dried, or freeze-dried. The peels were extracted with 80% acetone and the total phenolic contents of the dried samples were analyzed (**Figure 2**). The values for the fresh peels (500.2 ± 13.7 mg gallic acid equivalents/100 g fresh peel), untreated air-dried peels (478.0 ± 22.9 mg gallic acid equivalents/100 g fresh peel), and untreated freeze-dried peels (491.6 ± 19.9 mg gallic acid equivalents/100 g fresh peel) were similar (p > 0.05), and higher than the untreated oven-dried samples (p < 0.05). The untreated peels dried at 40, 60, and 80 °C had similar total phenolic contents of 361.3 ± 60.9, 303.4 ± 24.2, and 300.4 ± 44.4 mg gallic acid equivalents/100 g fresh peels, respectively (p > 0.05). The total phenolic contents of the air-dried blanched peels (463.7 ± 27.3 mg/gallic acid equivalents/100 g fresh peels) and freeze-dried blanched peels (460.6 ± 41.7 mg/gallic acid equivalents/100 g fresh peels) were not different from that of the fresh peels (p > 0.05). The 40, 60, and 80 °C oven-dried blanched peels had similar phenolic contents (346.0 ± 47.2, 392.3 ± 47.9, and 375.2 ± 20.0 mg gallic acid equivalents/100 g fresh apple peels, respectively) (p > 0.05) and the values were significantly lower than that of the fresh peels (p < 0.05). There was no significant difference between total phenolic contents of the untreated and blanched peels dried under any drying condition.

The flavonoid contents of the untreated and blanched peels dried under various conditions were determined and compared to the flavonoid content of fresh apple peels (**Figure 3**). The fresh peels had a flavonoid content of 306.1 ± 6.7 mg catechin equivalents/100 g. The untreated air-dried and freeze-dried peels had similar contents with 302.8 ± 23.5 and 308.0 ± 16.7 mg catechin equivalents/100 g fresh peels (p > 0.05). All untreated oven-dried peels had flavonoid contents that were significantly lower than the fresh peels and untreated freeze-dried peels (p < 0.05). The values were 233.6 ± 47.7, 172.3 ± 14.4, and 152.2 ± 28.7 mg catechin equivalents/100 g fresh peels for the untreated 40, 60, and 80 °C oven-dried peels, respectively. The fresh peels, blanched air-dried peels (286.4 ± 23.4 mg catechin equivalents/100 g fresh peels), and blanched freeze-dried peels (291.4 ± 26.1 mg catechin equivalents/100 g fresh peels) were higher in flavonoids than the blanched 40 °C oven-dried peels (218.8 ± 30.8 mg catechin equivalents/100 g fresh peels) (p < 0.05). The flavonoid contents of the blanched 60 and 80 °C oven-dried peels (257.9 ± 30.7 and 250.1 ± 14.1 mg catechin equivalents/100 g fresh peels, respectively) were similar to those of the fresh peels, blanched air-dried peels and blanched freeze-dried peels (p > 0.05). There were no significant differences between the flavonoid contents of the untreated and blanched peels under any drying condition (p > 0.05).

The anthocyanin contents of the apple peel extracts were measured (**Figure 4**). The untreated air-dried and freeze-dried peels had similar anthocyanin contents (5.84 ± 0.63 and 6.08 ± 0.96 mg cyanidin 3-glucoside equivalents/100 g fresh peels,
respectively) \((p > 0.05)\) and had higher contents than the fresh peels \((2.09 \pm 0.03 \text{ mg cyanidin 3-glucoside equivalents/100 g})\) \((p < 0.05)\). The untreated oven-dried peels had similar anthocyanin contents to the fresh peels \((p > 0.05)\). The values were \(2.09 \pm 0.03, 2.54 \pm 0.74, 0.90 \pm 0.16, 0.69 \pm 0.25 \text{ mg cyanidin 3-glucoside/100 g fresh peels for the untreated 40, 60, and 80 °C oven-dried peels, respectively.} The blanched freeze-dried peels \((6.43 \pm 1.57 \text{ mg cyanidin 3-glucoside/100 g fresh peels})\) contained more anthocyanins than the fresh peels \((p < 0.05)\) and had a similar content to the blanched air-dried peels \((5.10 \pm 0.79 \text{ mg cyanidin 3-glucoside/100 g fresh peel})\). There was no difference between the anthocyanin contents of the blanched oven-dried peels \((40, 60,\) and \(80 °C peels had 2.20 \pm 0.31, 3.61 \pm 1.27,\) and \(2.33 \pm 0.91 \text{ mg cyanidin 3-glucoside/100 g fresh peels, respectively} and the fresh peels \((p > 0.05)\). The anthocyanin contents of the untreated and blanched peels were similar at each temperature \((p > 0.05)\). The appearances of the apple peel powders showed that the blanched freeze-dried peels had the most desirable color (Figure 5).

The water activity of the apple peel powders was measured (Figure 6). They ranged from 0.12 (blanched 80 °C oven-dried peels) to 0.36 (blanched air-dried peels). There was no significant difference in water activities between the untreated and blanched peels under a given drying condition \((p > 0.05)\), except for the 80 °C oven-dried peels \((p < 0.05)\). The water activities of the peels decreased with increasing drying temperature for the oven-dried samples. The freeze-dried peels had water activities of 0.16 ± 0.01 for the untreated peels, which was similar to the blanched peels \((0.15 \pm 0.01)\) \((p > 0.05)\).

After reviewing the data, we chose to blanch the peels for 10 s and freeze-dry them to make the most stable food ingredient with high phytochemical content and desirable color. To better characterize the apple peel powder, the production was scaled up and the resulting ingredient was analyzed for phytochemical content, water activity, total antioxidant activity, and antiproliferative activity.

Scale-Up. One hundred twenty Rome Beauty apples were peeled, blanched for 10 s, and freeze-dried as described above. The resulting dried peels were ground to a 60-mesh powder in a benchtop hammermill and stored in sealed glass containers at \(-40 °C\) until analysis. The phytochemical contents of the apple peel powder ingredient and fresh apple peels are shown in Figure 7. The freeze-dried apple peels had a total phenolic content of 3342 ± 12 mg gallic acid equivalents/100 g freeze-dried peels. For easier comparison with fresh peels, the phenolic content of the dried peels on a fresh weight basis was calculated to be 580.0 ± 2.1 mg gallic acid equivalents/100 g fresh peels. This was significantly higher than the phenolic content of the fresh peels \((401.9 ± 11.2 \text{ mg gallic acid equivalents/100 g})\) on a fresh weight basis \((p < 0.05)\). The flavonoid content was 2299 ± 52 mg catechin equivalents/100 g freeze-dried peels, and was equivalent to 398.9 ± 8.9 mg catechin equivalents/100 g fresh peels. Again, on a fresh basis, this was higher than the flavonoid content of fresh apple peels \((306.1 ± 6.7 \text{ mg catechin equivalents/100 g fresh peels})\) \((p < 0.05)\). The anthocyanin content of the freeze-dried peels \((170 ± 2 \text{ mg cyanidin 3-glucoside equivalents/100 g or 29.5 ± 0.3 mg cyanidin 3-glucoside equivalents/100 g fresh peels})\) was much higher than the anthocyanin content of the fresh peels \((2.09 ± 0.02 \text{ mg cyanidin 3-glucoside/100 g})\) \((p < 0.05)\). This is an apparent 14-fold increase in the anthocyanin content of the apple peels after blanching and freeze-drying. The apple peel powder had a total antioxidant activity of 1251 ± 56 µmol vitamin C equivalents/g (217.1 ±
9.8 \( \mu \text{mol vitamin C equivalents/g fresh peels} \). Thus, 1 g of dried apple peels had the same antioxidant value as 220 mg of vitamin C. On a fresh weight basis this was similar to the antioxidant activity of fresh Rome Beauty apple peels (223.6 ± 6.5 \( \mu \text{mol vitamin C equivalents/g} \) (\( p > 0.05 \)). The freeze-dried apple peels also had a strong antiproliferative activity on HepG2 liver cancer cells (Figure 8) with a median effective dose \( EC_{50} \) of 1.88 ± 0.01 mg/mL (10.83 ± 0.03 mg fresh peels/mL). This was lower than the \( EC_{50} \) of 12.41 ± 0.36 mg/mL exhibited by the fresh apple peels (\( p < 0.05 \)). All values for the apple peel powder were significantly different from those of the fresh apple peels when compared on a dry weight basis (\( p < 0.05 \)).

**DISCUSSION**

In this study we have developed a value-added food ingredient from Rome Beauty apple peels. This variety of apple is commonly used in applesauce manufacture in New York State. We feel this could be a valuable use of the 20 million pounds of waste peels generated from applesauce and canned apple processing in this state each year. The phytochemical content, antioxidant activity, and antiproliferative activity of the fresh peels were preserved through blanching the peels for 10 s followed by freeze-drying and grinding them to a powder. This apple peel powder ingredient could be added to various food products to enhance their nutritional values.

Polyphenoloxidase is responsible for enzymatic browning of apple tissue. When the cellular structure is broken during slicing and other processes, the compartmentalized enzyme is mixed with substrate. Chlorogenic acid is thought to be the main substrate for enzymatic browning reactions. Enzymatic browning can be controlled using heat inactivation of enzymes, sulfur dioxide or ascorbic acid treatments, deaeration, storage at low temperatures, or acidification with citric or malic acids (29). We did not find citric acid or ascorbic acid treatments effective in preventing oxidation of the phenolic compounds during oven-drying of the apple peels at 60 °C. Heat inactivation by blanching the peels for 10 s was much more successful. We did not use sulfur dioxide treatments, as many people are hypersensitive to these compounds and producers of value-added products prefer to keep their labels “clean.” Nonenzymatic browning of apples can also be a problem during dehydration. Heat increases this type of browning under low moisture conditions. It is best to obtain the lowest moisture content possible with minimal heat damage to prolong the shelf life of dried apples (29). Our results support this, as the air-dried and freeze-dried apple peels retained their phenolic compounds better than the oven-dried samples.

We saw some slight, insignificant decreases in the phenolic and flavonoid contents of air-dried and freeze-dried apple peels that were blanched for 10 s, compared to the untreated peels. This was likely due to leaching of the compounds into the boiling water. This loss of phytochemicals during blanching has been noted in other produce. Broccoli that was steam-blanched or water-blanched for four minutes showed large losses of glucosinolates, with the water blanch leading to greater losses due to leaching (30). The 10 s duration of the apple peel blanch was probably not long enough to lead to great phytochemical losses. Despite the slightly lower flavonoid and phenolic contents, the appearances of the apple peel powders made from blanched peels were much more red and appealing than the powders made from unblanched apple peels. The color compounds, anthocyanins, can be degraded by glycosidases, polyphenoloxidases, and peroxidases (31). Thus, blanching likely inactivated these enzymes and preserved the color of the apple peels.

On the basis of the collected data, we believed that the blanched freeze-dried peels would be the best choice for a food ingredient. Although the freeze-dried peels did not have significantly different phenolic, flavonoid, or anthocyanin contents from the air-dried peels, they did have lower water activity. Decreased water activities could allow for greater chemical stability and increased shelf life of the apple peel powder. Apple pomace extracts dried on maltodextrin that had water activities lower than 0.3 had a much longer shelf life than those with higher water activities (31). The untreated freeze-dried peels had similar phytochemical contents and water activity to the blanched peels. However, the presence of potentially active enzymes in the untreated peels could lead to problems with the apple peel powder during storage and in food applications. Peels blanched for 10 s and freeze-dried before being ground to a powder would make the most stable product.

Unexpectedly, on a fresh weight basis the freeze-dried peels contained significantly higher phytochemical contents than the fresh peels and exhibited higher antiproliferative activity. The Rome Beauty apples used in the scale-up were purchased a few months later than the ones used in the fresh peels analysis. However, they were grown at the same orchard and in the same season. It has been shown that seasonal variations of specific phytochemicals in a particular apple cultivar appear to be small in most cases (15), and cold storage of apples does not substantially affect the flavonoid and chlorogenic acid content of apple peels (14, 32). Other factors have much larger influences on the phytochemical content of apples. The apple variety (15) and the amount of light the apples receive (33) affect some phenolic concentrations. It was determined that concentrations of catechins, chloridzin, and chlorogenic acid in the peel of Jonagold apples are independent of light exposure; however, shading decreases levels of cyanidin 3-galactoside and quercetin 3-glycosides (33). The particular orchard apples are grown in can influence flavonoid content, possibly due to differences in shading and soil nutrient availability (34). Thus, the differences between the apple peel powder ingredient and the fresh apple peels could be accounted for by the variations in the amount of light the apples received. Another explanation is that the processing conditions could release bound phenolic compounds and make them available for measurement, as over 50% of phenolics in apples are conjugated (3). Lycopene in...
tomatoes (23) and ferulic acid in corn (35) are released in this manner during heat processing.

An attempt was made in the past to create a valuable product from apple peels (36). The peels from apples were soaked, macerated, and mixed with macerated core materials that had been deseeded and destemmed. The mixture was then drum-dried and passed through a 100-mesh screen. The powder was used as a thickener in apple pies. The harsh drying conditions resulted in the loss of most of the aroma from the peels and a brown appearance. The pies made with the apple peel powder did not have higher taste ratings, probably due to the undesirable brown color of the powder.

We feel the apple peel powder ingredient we have developed could be a valuable and attractive addition to healthy food products. A small amount could greatly increase the phytochemical content and antioxidant activity of foods. Applications may include cereals, granola bars, fruit leathers, and sports bars. Inclusion of phytochemical-rich apple peels in a diet high in brown color of the powder.

We feel the apple peel powder ingredient we have developed could be a valuable and attractive addition to healthy food products. A small amount could greatly increase the phytochemical content and antioxidant activity of foods. Applications may include cereals, granola bars, fruit leathers, and sports bars. Inclusion of phytochemical-rich apple peels in a diet high in fruits, vegetables, and whole grains could assist in the prevention and management of chronic diseases.

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