

Chapter 2

Developing Fruit Cultivars with Enhanced Health Properties

Michael J. Wargovich, Jay Morris, Vondina Moseley,
Rebecca Weber, and David H. Byrne

Abstract One hypothesis to account for the dramatic increase of inflammatory driven diseases, such as cancer, cardiovascular disease, obesity, diabetes, and others, across the world is the coincidental displacement of fruits and vegetables in the diet with processed foods as populations in the developing world rapidly acculturate to a more affluent lifestyle. Fruits are rich sources of antioxidant and anti-inflammatory natural compounds that offset many of the biological events leading to the development of the above-mentioned chronic diseases. In this review, potentially cancer-protective phytochemicals in fruits are reviewed to describe the research approaches, the range of chemistry and mechanisms seen in the study of the health benefits of fruit phytochemicals. Furthermore, given the rapid increase in research, public's interest in the health benefits of food, and the government's and food industry's efforts to develop and promote healthy foods, fruit breeders have begun to investigate the feasibility of developing health-enhanced fruit cultivars. Thus far, there appears to be ample genetic variability within fruit crops to develop cultivars with higher levels of plant phytochemicals, such as total phenolics, anthocyanins, and antioxidant activity. Nevertheless, selecting breeding targets is elusive as there is little information on which specific phytochemical or combination of phytochemicals and the levels needed to effectively enhance the health of the consuming public.

M.J. Wargovich (✉) • J. Morris • V. Moseley • R. Weber
Department of Cellular and Molecular Pharmacology and Experimental Therapeutics,
Hollings Cancer Center, Medical University of South Carolina, 86 Jonathan Lucas Street,
Charleston, SC 29424, USA
e-mail: wargovic@musc.edu; morrisjl@musc.edu; browv@musc.edu; webe@musc.edu

D.H. Byrne
Department of Horticultural Sciences, Texas A&M University,
College Station, TX 77843-2133, USA
e-mail: dbyrne@tamu.edu

Keywords Phytochemicals • Cancer • Cardio vascular disease • Obesity • Diabetes • Antioxidants • Phenolics • Anthocyanins • Carotenoids • Chronic diseases • Anti-inflammation

1 Introduction

Chronic diseases are on the rise in the developing world. At the core of risk for diseases, such as cancer, heart disease, neurological disorders, obesity, and diabetes, is uncontrolled chronic inflammation deep in the cells of the body. While inflammation is a natural process of healing damage to the body, the genetic and biochemical machinery underpinning inflammation is often corrupted, resulting in the prevalent chronic diseases we recognize today.

One recognized factor in the development of chronic disease is poor nutrition. And in an inverse way, the climb from undeveloped to developed nation status makes us come full circle from inadequate nutrition to super-adequate nutrition, both states that could be characterized as “poor.” To explain this conundrum, it is possible that populations may reach a state of affluence, where they displace the fruit and vegetable portion of the diet with super-caloric foods, devoid of natural phytochemicals, that may have helped to offset chronic disease risk. With these natural guardians against oxidative damage and inflammation, affluent societies are now afflicted with epidemics of chronic inflammatory-driven diseases.

Fruits and vegetables have always been considered a foundation of a healthy lifestyle and a healthy diet. Unfortunately, despite the solid research, government and health agency recommendations, and a population that is growing increasingly old, the public health message to eat more fruits and vegetables has fallen on deaf ears. In the USA, most Americans do not come close to the recommended consumption of five to nine servings of fruits and vegetables per day (Pollack and Perez 2008; Wells and Buzby 2008) and this aversion begins in the teen and preteen years, a time when chronic disease risk may be set (Nanney et al. 2007; Cade et al. 2006).

The intent of this chapter is to review the evidence for fruit consumption and health benefits with an emphasis on cancer and evaluate the potential of developing fruit cultivars with enhanced levels of beneficial phytochemicals as an approach to increase the consumption of these useful compounds.

2 Phytochemicals and Cancer

The USA and many developed countries are experiencing an epidemic of diseases which may have chronic, unresolved inflammation as their common etiology (Beaglehole et al. 2007). Clearly, the impact of diet is seminal in establishing protection early in life from chronic disease, and the loss of dietary protectants, by circumstance or will, may now factor into the epidemic facing all societies. In the

Table 2.1 Human evidence for cancer prevention: Fruit consumption

Site of cancer	Types of study	Finding	Reference
Oropharyngeal	2 ECO, 1CO, 35 CC	Probably preventive	AICR (2007)
Esophagus	7 ECO, 4 CO, 36 CC	Probably preventive	AICR (2007)
Lung	7 ECO, 25 CO, 32 CC	Convincingly preventive	AICR (2007)
Stomach	23 ECO, 16 CO, 51 CC	Probably preventive	AICR (2007)
Pancreas	8 ECO, 6 CO, 6 CC	Not plausible	AICR (2007)
Liver	1 CO, 5 CC	Not plausible	AICR (2007)
Prostate	3ECO, 28 CO, 18 CC	Inconsistent	Lewis et al. (2009)
Breast	8 CO, 2CC	Inconsistent	Vainio and Weiderpass (2006)

Abbreviations: *ECO* ecological studies, *CO* cohort studies, *CC* case–control studies

last 40 years, a wealth of epidemiological data, gleaned from over 150 ecological, cohort, and case–control studies, has supported the notion that persistent dietary exposure to fruits and vegetables are salutary for health. While overall evidence is suggestive of protection, evidence for reduction in risk for only a few of the major cancers is considerable enough to be called protective. It should not be concluded that phytochemicals from frequent fruit and vegetable consumption are ineffective for other cancers, rather that there is at present insufficient data to warrant a conclusive protective effect. Table 2.1 lists some of the common sites of cancer and summarizes the available data regarding cancer protection. The conclusions are drawn by an expert panel commissioned by the World Cancer Research Fund/American Institute for Cancer Research in its updated review published in 2007.

2.1 *Phytochemicals in Fruits*

In the last 15 years of research, much of the protective effects for consumption of plant foods have been ascribed to the constituent phytochemicals resident in them (Newman and Cragg 2007). In all fruits and vegetables, the major classes of phytochemicals consist broadly of carotenoids, flavonoids, isoflavonoids, and phenolic acids (Pan et al. 2008). Plant phenolics represent a structurally diverse superclass of compounds possessing one or more aromatic rings, one or more hydroxyl groups, and additional moieties covering over 8,000 unique chemicals (Huang et al. 2010). The flavonoids represent over 4,000 compounds and are an extension of the phenolic group, but have at least two aromatic rings with a variety of additional structural elements. It is this class of natural compounds that have generated so much interest in the cancer prevention research and represents many of the active compounds in fruits. Many of the bioactive agents identified from medicinal herbs and spices are members of this class of phytochemicals. Flavonoids can be further subdivided into flavones, flavonols, flavonones, isoflavones, and anthocyanidins. The latter category is of intense interest. The anthocyanidins broadly account for the red-to-purple

pigmenting of many commonly consumed fruits, especially in grapes, plums, cherries, and berries. In many tree fruits, the presence of anthocyanidins in most cultivars is typically concentrated in the skin, although most of these, such as apples, peaches, plums, and kiwis, have genotypes that contain anthocyanins in the flesh as well (Vizzotto et al. 2007; Voltz et al. 2009; Jaeger and Harker 2005).

2.2 *Fruit Phytochemicals: Evidence for Health Benefits*

Taken as whole, the production of fruits and vegetables has been robust with most of the growth in production in vegetables, rather than in fruit. Exports of fruit have grown, especially those from developing countries, and the industry has diversified, ensuring (at least in some developed countries) not only a year-round supply of fresh fruit, but oversupply has led to the marketing of specialized fruits, such as those organically grown. This for a large part has been due to the public perception that organically grown is better for health maintenance (WHO 2005).

Often, the first type of evidence for health benefits of fruit consumption is drawn from *epidemiological* studies. Three types of studies are often conducted: those at the ecological level (comparing types of fruit and quantities across populations), the cohort level (comparing fruit consumption within a population that has been followed for some time), and the case–control level (comparing fruit consumption in those with and without disease). Among tumor types, risk for cancers of the oral cavity, esophagus, and colorectum seems to be less when high amount of fruits and vegetables are in the diet. The evidence for protection is less than certain for cancers of the stomach, lung, breast, and prostate (Key 2011). The overall risk for cancer has been examined in four large and well-conducted prospective studies. In two cohort studies, the Nurse's Health Study and the Health Professionals' Follow-up study, conducted by Harvard, no significant reduction in overall risk was noted, although there was a trend to protection (Hung et al. 2004). These studies were supported by the Japanese Public Health Center prospective study while the European-based EPIC study found a significant reduction in cancer risk for consumption of fruits and vegetables (Takachi et al. 2008; Buchner et al. 2011). The US-based NIH-AARP Diet and Health Study found mixed results with more protection noted for vegetable consumption than fruits (George et al. 2009). These types of studies are notoriously difficult to conduct and to interpret, and it may well be that certain types of cancers are more amenable to prevention by specific fruits or specific vegetables based upon their unique phytochemical signatures. Examples include some of the unique phytochemicals in green tea and the phytoestrogenic compounds in soy.

Basic research into the potential mechanisms by which fruits or vegetables prevent cancer has unveiled an incredible variety of ways in which the cancer process can be interrupted. How does the process of identifying potential benefits of a particular fruit or vegetable begin? The customary protocol for this type of research originates with epidemiology. When a consumption pattern is associated with reduced risk for cancer, the usual first step is to extract the fruit or vegetable in organic

solvents or by supercritical CO₂ for testing in *in vitro* assays to test whether the extracts have cytotoxicity toward human cancer cells. Ideally, these assays detect whether the parent extract kills tumor cells in a dose- and time-related manner. Also, ideally, it should include testing on normal human cells from the same organ, but there are many limitations as these are not available from human cells for many common sites of tumorigenesis. The next step is a process of discovery and employs the concept of structure–activity-guided fractionization. Essentially, the plant extract is further purified leading to identity of specific classes or individual compounds for which the most robust anticancer activity is noted. Thus, a specific fruit can be extracted into specific flavonoid fractions, yielding a specific chemical identified through mass spectrometry. The identified chemical may be the best of the extracted agents that shows robust cytotoxicity as well as other important anticancer features, such as being anti-inflammatory, antiangiogenic, proapoptotic, or activating genes involved in cell regulation (Table 2.2). Often, cell culture studies are used to probe potential mechanisms by which phytochemicals prevent cancer growth or expansion.

After gathering data from *in vitro* systems, the next step is to evaluate the candidate-preventive phytochemicals *in vivo* in relevant *animal models* (Table 2.3) that replicate human cancer. Animal models for cancer are usually developed in mice or rats, and can be carcinogen initiated or initiated by altering key genes that have been associated with common human cancers. Typically, animal carcinogenesis assays provide the phytochemical orally either mixed into rodent diet or given in the drinking water. Sometimes, it is necessary to intragastrically intubate the animal with the test agent. In a preventive protocol, the animals are introduced to the test phytochemical prior to or during the time of “initiation” while in a therapeutic protocol, the test agent is administered after the tumorigenic process has advanced. End points typically involve the measure of incidence of cancer in the animals, the tumor burden and severity, as well as the measure of biological markers. One of the newest approaches to the testing of the anticancer capacity of a given phytochemical is to see if it may work additively or synergistically to aid and abet conventional cancer treatment. An added benefit would be to observe an increased therapeutic index while offsetting or reducing the incidence of off-target toxicity, commonly referred to as the side effects of cancer therapy.

3 Phytochemicals and Other Chronic Diseases

The previous section examined the evidence, phytochemicals, mechanisms, and the experimental approaches involved to determine the effect of fruit phytochemicals on the development of cancer. For all diseases, the experimental approaches of epidemiological studies combined with *in vitro* animal models and human clinical trials are used to identify major risk factors and potential control strategies. Because there is increasing evidence that aberrant inflammation lies at the molecular core of processes involved in more than just cancer, it is possible that fruit consumption will have collateral benefits for prevention of heart disease, obesity, diabetes, Alzheimer’s disease, and other neurodegenerative diseases.

Table 2.2 Mechanisms of tumor growth inhibition

Mechanism	Key mediators	Mechanism of action	Outcome	Fruits	References
Cell adhesion molecules	VEGF	Decreased	Decreased angiogenesis	Black raspberries	Liu et al. (2005a, b)
	ICAM-1, VCAM	Reduce upregulation induced by TNF α	Decreased cell migration	Blueberries and cranberries	Youdim et al. (2002)
		Decreased	Could be due to inactivation of NF κ B pathway	Cranberry	Ruel and Couillard (2007)
	MMPs	Expression of 2 and 9 inhibited	Decreased cell migration	Cranberries, raspberries, blackberries, blueberries, muscadine grapes	Neto (2007), Tate et al. (2004), Matchett et al. (2005)
	GAGs	Decreases 9 Decreases in sulfation	Reduce chronic inflammation	Resveratrol Blueberries	Woo et al. (2004) Neto (2007), Tovar et al. (1998)
Proinflammatory	MCP-1	Reduce upregulation		Blueberries and cranberries	Youdim et al. (2002), Neto (2007)
	MAPK/ERK	Increases ERK activation	Increased neurogenesis	Blueberry	Shukitt-Hale et al. (2008)
	IGF-1	Activates		Blueberry	Shukitt-Hale et al. (2008)
	TNF α	Reduced expression	Decreased inflammation	Black raspberries	Montrose et al. (2011), Bodet et al. (2006)
	CRP	Decreased expression	Decreased inflammation	Cranberries Raspberries	Rao and Snyder (2010), Bodet et al. (2006)
	IL-1 β	Reduced expression	Decreased inflammation	Cranberries	Montrose et al. (2011)
	NF κ B	Reduced expression via increased I κ B expression	Decreased inflammation	Black raspberries Black raspberries	Montrose et al. (2011)
		Reduced expression	Decreased inflammation	Berry fruits	Shukitt-Hale et al. (2008)
		Reduced expression	Decreased inflammation	Apple oligogalactan	Liu et al. (2010)
		Activation of pathways	Suppress inflammatory cascade	Resveratrol	Leiro et al. (2005)

COX2	Reduced expression Reduced expression Inhibit COX1/2	Decreased inflammation	Black raspberries Raspberries Blueberries and strawberries	Mallery et al. (2008) Rao and Snyder (2010) Shukitt-Hale et al. (2008)
JAK/STAT	Activation of pathways	Suppresses inflammatory cascade	Resveratrol	Wung et al. (2005)
PKC	Increase Inhibits	Reduced MMP9	Fruit phenolics, berry fruits Resveratrol	Shukitt-Hale et al. (2008) Woo et al. (2004)
JNK	Inhibits	Reduced MMP9	Resveratrol	Woo et al. (2004)
IL-8	Reduce upregulation	Decreased inflammation	Blueberries, cranberries	Bodet et al. (2006), Neto (2007)
IL-6	Reduced	Reduced	Reduced	Bodet et al. (2006)
PGE ₂	Reduced	Decreased inflammation	Cranberries	Montrose et al. (2011)
Arachadonic acid	Reduced expression Suppress pathway	Decreased inflammation	Black raspberries Fruit phenolics	Shukitt-Hale et al. (2008)
Apoptotic Oxidative stress	Reduce apoptosis Decreases ROS formation Decreased expression Reduces H2O2 Upregulation of glutathione synthesis	Protection against oxidative damage Reduced necrosis Decrease oxidative stress and reduced DNA damage	Berry fruits Blueberries and cranberries Berry juice blend Raspberries Cranberry Fruit phenolics	Neto (2007) Jensen et al. (2008) Rao and Snyder (2010) Neto (2007) Shukitt-Hale et al. (2008), Weisel et al. (2006)
Peroxide				
Glutathione				
			Berry fruits, raspberries	

(continued)

Table 2.2 (continued)

Mechanism	Key mediators	Mechanism of action	Outcome	Fruits	References
	Reactive oxygen species	Reduced	Decrease oxidative stress	Blueberries and strawberries, concord grape	Neto (2007), Shukitt-Hale et al. (2008, 2006)
		Reduced	Reactive oxygen species absorbed	Plums	Yang and Gallaher (2005)
		Reduced	Oxidative damage	Apples	Gerhauser (2008)
		Scavenge	Reducing neuronal age-related deficits	Blueberries	Shukitt-Hale et al. (2008, 2006), Joseph et al. (1999)
		Scavenge	Antioxidant radicals	Prunes (plums)	Stacewicz-Sapuntzakis et al. (2001)
Platelet effects	TXA ₂	Reduced	Lipid peroxidation	Apple extracts	Fini et al. (2011)
		Inhibits platelet aggregation, calcium mobilization, hydrogen peroxide formation, and TXA ₂ production induced from collagen and arachadonic acid	Inhibits platelet activation	Pomegranate	Mattiello et al. (2009)
Lipoprotein effects		Inhibiting oxidation of circulating lipoproteins	Slowed CIMP progression for individuals at risk for CHD	Pomegranate	Davidson et al. (2009)
		Protect against lipid oxidation	Protection against oxidative damage	Strawberries and blueberries	Shukitt-Hale et al. (2008), Neto (2007)
		Inhibits lipid peroxidase	Protection against oxidative damage	Berry juice blend	Jensen et al. (2008)
		Prevention of obesity-related colon cancer	Protection against oxidative damage	Apple juice	Koch et al. (2009)

Table 2.3 Effect of fruit phytochemicals in animal models of cancer

Organ	Animal model	Phytochemical	Result	Reference
Breast	7,12 DMBA rats	Grape seed extract	Reduction in tumor multiplicity	Kim et al. (2004), Mehta and Lansky (2004)
Skin	DMBA, TPA mouse	Pomegranate seed oil	Tumor reduction	Adhami et al. (2009), Hora et al. (2003), Afaq et al. (2005), Jang et al. (1997), Zhao et al. (1999)
		Pomegranate seed oil (anthocyanins)	Chemopreventive	
		Resveratrol		
Esophagus	UV-induced mouse	Grape seed powder		Aziz et al. (2005)
	NMBA F344 rats	Resveratrol	Chemopreventive	
		Resveratrol	Inhibits tumor multiplicity	
Colon	NMBA mice	Acai, strawberries, wolfberry, noni		Li et al. (2002), Stoner et al. (2010)
		Black raspberries	Limit cancer development	
		Black raspberries	Inhibit carcinogenesis	
Colon	AOM rat	Black raspberries		Harris et al. (2001), Lala et al. (2006), Gosse et al. (2005), Kohno et al. (2004)
	AOM, DMBA rat	Bilberry	Reduced ACF	
		Chokeberry	Promotes apoptosis	
Colon	1,2 DMH F344 rats	Grape	Chemopreventive	Durak et al. (2005)
		Apple procyanidins		
		Pomegranates		
Colon	APC Min mice	Grape seed extract	Decreased ACF	Sengottuvelan et al. (2006), Barth et al. (2005)
	1,2 DMH F344 rats	Resveratrol	Reduced colon tumors	
		Cloudy apple juice (procyanidins, pectin)	Decreased proliferation, ACF, DNA damage	
Cheek	7,12 DMBA hamster	Black raspberries	Inhibit carcinogenesis	Duncan et al. (2009), Rajakangas et al. (2008)
		White currants (anthocyanins)		
		Black raspberries		
Prostate	TRAMP model	Grape seed extract	Cell cycle arrest	Casto et al. 2002
Lung	B(a)P and NTCU mice	Tomatoes (lycopene)	Chemopreventive	Raina et al. (2007), Konijeti et al. (2010)
		Pomegranate	Chemopreventive	

Carcinogen key: *AOM* azoxymethane, *DMBA* dimethylbenzanthracene, *TPA* tetradecanoylphorbol acetate, *DMH* dimethylhydrazine, *NMBA* nitrosomethyl benzylamine, *B(a)P* benzo(a)pyrene, *NTCU* *n*-nitroso-tris-chloroethylurea

3.1 Tunneling Down: An Example of a Phytochemical Class with Promise for Prevention of Disease: Anthocyanins

With the large array of fruits and the added numbers of beneficial phytochemicals they contain, determining which whole fruit, compound, or extract is the most beneficial for a given modality can be exhausting. We have already discussed epidemiological evidence as well as some basic research involving benefits from fruits. Here, we focus on the class of fruits rich in anthocyanins, a class of phytochemicals which have been heavily studied, and the results in disease prevention have been promising (Hung et al. 2004; Neto 2007; Shukitt-Hale et al. 2008; Rao and Snyder 2010; Kim et al. 2004; Afaq et al. 2005; Jang et al. 1997; Lala et al. 2006; Larsson et al. 2008; Pan et al. 2008; Johnson 2007; Renaud and de Lorgeril 1992; Chou et al. 2001; Freedman et al. 2001; Sautebin et al. 2004; Ilbey et al. 2009; Kim et al. 2008). Anthocyanins are primarily responsible for the red, blue, and purple colors of fruits and over 400 individual compounds have been identified (Mazza and Miniati 1993). The average daily intake of anthocyanins is estimated to be 12.5 mg/day/person in the USA (NHANES 2001–2002). The amount and type of anthocyanin vary for different fruits, but for our purposes we focus on total anthocyanins. For instance, red grapes have 42.7 mg while concord grapes have 192 mg of total aglycone anthocyanins (mg/100 g fresh wt) (Wu et al. 1993). For berries like black, blue, cran, and raspberries, the total aglycone anthocyanin levels are 353, 529, 133, and 116 mg (mg/100 g fresh wt), respectively (Wu et al. 2006). Pomegranate juice has 429.9 mg/l total anthocyanins (Orak 2009), whereas the acai berry has been shown to contain 3.1919 mg/g dry wt total anthocyanins (Schauss et al. 2006). Despite these varying anthocyanin levels beneficial disease-preventative properties have been reported in all of these fruits.

Grapes and other small fruits are the most commonly known anthocyanin-rich fruits. From red and concord grapes to wine to blueberries and raspberries, most people have consumed one or more of these in their diet. For instance, the “French Paradox,” first mentioned in 1992, is related to relatively low risk of cardiovascular disease (CVD) in the French despite a diet rich in saturated fats (a risk factor component of CVD (Renaud and de Lorgeril 1992). Years later, evidence still supports that moderate consumption of red wine (one to two drinks per day) contributes beneficial cardiovascular effects in most populations (Lippi et al. 2010). Is this true for wine’s predecessors, the grape? Yes. Grapes have beneficial preventative properties too (Table 2.4). Despite the lack of alcohol (an active component in wine), compounds from red and concord grapes displayed numerous preventative effects. Extracts from grapes have been shown to improve cardiovascular health through reduction in cellular oxidation (Bertelli and Das 2009; Rice-Evans et al. 1996) by enhancing nitric oxide release (Freedman et al. 2001) and inhibiting some cholesterol intake (Leifert and Abeywardena 2008). In addition to the heart, grapes have been implicated in improving motor and memory function as well as improving mood (Shukitt-Hale et al. 2006; Krikorian et al. 2010). These are just a few of the human health-related benefits of grape consumption.

Table 2.4 Disease prevention by anthocyanin-rich fruits

Selected fruit	Selected diseases	Preventative properties	Reference
Grape	Cardiovascular disease (CVD)	Antioxidation	Bertelli and Das (2009)
Red	Brain degeneration (CVD)	Inhibits cholesterol uptake and 5-LOX activity	Leifert and Abeywardena (2008)
Concord	Dementia	Protects against decrease in synaptic protein function	Sun et al. (1999)
		Reduces LDL oxidation	Rice-Evans et al. (1996)
		Endothelial function improvement	Chou et al. (2001)
		Enhances nitric oxide release	Freedman et al. (2001)
		Increases dopamine release and motor function	Shukitt-Hale et al. (2006)
		Improves memory function	Krikorian et al. (2010)
Berries	Age-related cognitive decrease	Increase in working and short-term memory	Shukitt-Hale et al. (2009)
Blackberry	Endotoxic shock	Reduced iNOS and COX activity	Sautebin et al. (2004)
Blueberry	Diabetes	Insulin-like active principles and protection against glucose toxicity	Martineau et al. (2006)
Cranberry	Urinary tract infections	Bacterial antiadhesion	Gupta et al. (2007), Howell (2007)
Pomegranate juice	Prostate cancer	Decreases PSA doubling time, decreases cell proliferation, and increases apoptosis	Pantuck et al. (2006)
	Renal tubular cell injury	Reduces oxalate crystal formation	Ilbey et al. (2009)
	Osteoarthritis	Decrease in cell proliferation and inflammatory cells in synovial fluid	Hadipour-Jahromy and Mozaffari-Kermani (2010)
Muscadine grape	Microbial infection	Antimicrobial activity	Kim et al. (2008, 2010)
	Prostate cancer	Chemopreventative agent	God et al. (2007), Hudson et al. (2007)
Acai	Inflammation	Antioxidant	Schauss et al. (2006)

Much like grapes, berries are rich in anthocyanins (Mazza and Miniati 1993; Wu et al. 2006). Almost everyone is familiar with the effects cranberries have on urinary tract infections. This is due to the bacterial antiadhesion properties found within the anthocyanin profile of cranberries (Gupta et al. 2007; Howell 2007). Other berries, like blueberries, have disease prevention properties that differ from cranberries. Blueberries exhibit antidiabetic properties in in vitro assay, such as insulin-like

active properties, to protect against toxicity from glucose (Martineau et al. 2006). These two are not the only berries with anthocyanin-mediated health benefits. Blackberries have been reported to increase working and short-term memories which both play roles in age-related cognitive impairment (Shukitt-Hale et al. 2009). These berries can also reduce harmful effects from endotoxic shock (Sautebin et al. 2004). These are numerous other berries with high anthocyanin levels and reported human health benefits.

The historical-, clinical-, and media-driven reports of the health benefits of grapes and berries has led to the emergence of pomegranates, another fruit high in anthocyanins, as another fruit promoted for its health benefits (Wu et al. 2006). This promotion has led to a tripling of pomegranate plantings in California from 2002 to 2007 (USDA 2007). Research has shown that pomegranates are beneficial to prostate cancer prevention (Pantuck et al. 2006), can slow the symptoms of osteoarthritis (Hadipour-Jahromy and Mozaffari-Kermani 2010), and can reduce oxalate crystal formation in renal cells (Ilbey et al. 2009). Much like the grapes and berries, the pomegranate juice offers an easy enjoyable delivery system for the humans to ingest healthy anthocyanin compounds.

Muscadine grapes are common in the southeastern USA due to their ability to handle the humid summers and warmer winters (Olien 1990). They are red to purple in color like other grapes; however, they have higher antioxidant capacity than table grapes. This is due to a different anthocyanin profile, one similar to blackberries and raspberries (Rommel and Wrolstad 1993). Muscadine extracts and powders have an effect against microbial infection (Kim et al. 2008, 2010) and are potential chemopreventative agents in prostate cancer (God et al. 2007; Hudson et al. 2007).

Grapes and berries are not the only anthocyanin-rich fruits around; they are just the most well-known and, for the most part, well-studied. Other temperate fruit crops, such as apples, peach, plum, kiwi, and others, although typically do not have red flesh, have the potential to develop red-fleshed cultivars. In addition, exotic crops, such as acai berry, are starting to gain notoriety as a superfruit. The first research on acai focused on the remarkable antioxidant potential of acai berries and their impact of inflammation reduction (Schauss et al. 2006). Current research is focused on studying the health benefits of acai in animal models (Stoner et al. 2010; de Souza et al. 2010). As the beneficial effects with animal models become well-documented, hopefully the research will expand to human trials. Other fruits of interest are the pitanga (*Eugenia uniflora* L.) which has long been utilized in traditional Brazilian medicine to treat diarrhea (Brandelli et al. 2009). The more understanding of traditional medicine from plants to practice yields even more fruits with health benefits. Researchers making inroads into western Africa, Colombia, and other countries expand the knowledge of anthocyanin-rich fruits.

From this brief highlight of anthocyanin-rich fruits, it can be concluded that their health impact is widespread and varied. Previous sections have focused on specific cellular processes and epidemiological evidence. Here, we have shown how one class of bioactive compounds and fruits rich in anthocyanins are a cornerstone in understanding how specific dietary compounds can impact a myriad of maladies from heart disease and cancer to microbial infections. Research continues to show that fruit and vegetable consumption is beneficial to improved health. This is due to,

in part, anthocyanins and the increased protection they provide along with other bioactive compounds in fruits.

4 Genetic Variation Within Fruit Crops

4.1 Trend in Fruit Breeding

Fruit breeders need to anticipate the future as the cultivars they begin to develop now will not enter production for at least 10 years and frequently longer. Their objectives need to reflect the desires of the market (Byrne 2005). The previous section of this chapter has asserted that fruit phytochemicals affect the health of the people that consume them. Most of the studies have dealt with one cultivar and/or focused on a few chemical components of the phytochemicals available in the fruit. Thus, it has been clearly shown that there are differences among crops and that there is strong evidence that phytochemicals from these crops have protective properties against various chronic diseases, such as cancer and cardiovascular disease.

This information has been widely publicized and has created a proliferation of superfruits which are touted for their high level of antioxidants. These would include fruits, such as blueberries, pomegranates, cranberries, plums, acai, and others. This marketing approach has been effective in promoting the increased consumption of blueberries and pomegranates. The consumer makes the connection between food and health as the vast majority of consumers surveyed indicate that they take health into account when choosing food to purchase. This heightened awareness of the health benefits of food has increased the food industry's efforts in the development of foods with health benefits (Sloan 2006, 2008; Dillard and German 2000).

Since the 1990s, the US Government has been working toward convincing people to consume three to four portions or two cups of fruit a day, but still the average fruit consumption is only about half this recommendation (Pollack and Perez 2008; Wells and Buzby 2008). This presents an opportunity to fruit breeders. Since the amount of fruit consumed has not increased, the other approach would be to enhance the health benefits of the fruits that are consumed. As it has been seen with the health-oriented marketing of superfruits (i.e., pomegranate, blueberries), it is possible to increase the consumption of specific fruits by touting their high antioxidant capacity. The next step of this process would be to develop health-enhanced cultivars with a better phytochemical mix for a given crop.

4.2 Phytochemical Profiles Among Crops

The phytochemical profile of various crops and even their parts (peel versus flesh) also differs dramatically (Table 2.5). In apples, peaches, and plums, the peel is 6–9% of the fruit fresh weight, but because it contains from two to about five times the concentration of phenolics than the flesh, the peel is an important source of phenolics.

Table 2.5 Comparative phytochemical profile (% of total for each chemical group) of apple, peach, plum, and blueberry cultivars

Chemical group	Apple	Apple	Apple	Peach	Peach	Plum	Plum	Plum	Blueberry
	Fruit	Flesh	Peel	Flesh	Peel	Flesh	Peel	Fruit	
Procyanidins	42	53–56	38–60	50–67	40–59	71	56	0	
Hydroxycinnamic acids	29	39–40	8–10	30–46	22–36	27	12	30	
Flavanols	21	0–2	18–42	2	4–7	0	11	15	
Dihydrochalcones	7	4–6	7–12	0	0	0	0	0	
Anthocyanins	0	0	0–10	1–2	14–17	7	21	55	
Reference	Lata et al. (2009)	Khanizadeh et al. (2008), Tsao et al. (2003)	Lata et al. (2009), Khanizadeh et al. (2008), Tsao et al. (2003)	Tomas-Barberan et al. (2001)	Tomas-Barberan et al. (2001)	Tomas-Barberan et al. (2001)	Tomas-Barberan et al. (2001)	Zheng and Wang (2003)	

Procyanidins (catechin, epicatechin, procyanidin B1, procyanidin B2, other procyanidins), hydroxycinnamic acid (chlorogenic acid, neochlorogenic acid, *p*-coumaroyl quinic, caffeic acid, related compounds), flavanols (quercetin-3-rutinoside, quercetin-3-rhamnoside, other quercetin derivatives, myricetin, kaempferol), dihydrochalcones (phloretin-3-xyloglucoside, phloridzin), anthocyanins (cyanidin 3-glucoside, cyanidin 3-rutinoside, and glycosides of delphinidin, petunidin, malvidin, and others)

The peel can commonly contain 20–40% of the total phenolics and a major portion of the antioxidant capacity of these large fruited crops (Cevallos-Casals et al. 2006; Drogoudi et al. 2008; Lata et al. 2009; Khanizadeh et al. 2008; Tomas-Barberan et al. 2001). A similar situation exists in small fruits (blueberry, blackberry, raspberry) as seen in the negative correlation between fruit size and total phenolics and antioxidant activity. Although this effect is significant, when the data is adjusted for size, there is still abundant genetic variability for the total phenolic content in the flesh (Connor et al. 2002b, c, 2005a, b).

Among the cultivars of apple, peaches, plums, and blueberries surveyed, the predominance of the various chemical groups varies. All of these fruits have hydroxycinnamic acids as a predominant phenolic among their phytochemical mix. Apple, peach, and plum tend to be high in procyanidins and low in anthocyanins, whereas blueberries are the reverse. Apple is the only fruit of these that contain dihydrochalcones. Thus, the mix of phytochemicals within each crop varies from others which emphasizes the importance of the recommendation of eating a diversity of fruits to maintain good health.

This observation can be taken one step further to look at the composition of the specific compounds within each subclass in each crop. For example, the anthocyanins found in peach are mainly cyanidin 3-glucoside and cyanidin 3-rutinoside (Tomas-Barberan et al. 2001), whereas blueberries contain various forms (mainly 3-galactoside, 3-glucoside, and 3-arabinoside) of delphinidin, petunidin, cyanidin, and malvidin (Zheng and Wang 2003). This is frequently the situation within other classes of phytochemicals between the various crops.

The development of health-enhanced fruit cultivars requires that there is genetic variation for the trait within the crop with which the breeder is working. From a breeding perspective, the next step is to determine if the crop has the genetic variability needed to develop health-enhanced cultivars. Although there are hundreds of phytochemicals found in fruits, most of the literature is focused on the antioxidant bioactivity and the concentration of total phenolics and anthocyanins of fruit crops.

4.3 Antioxidants

The consumption of high levels of antioxidants is promoted as being beneficial to one's long-term health by reducing general oxidative stress within the body. Consequently, there has been interest in exploring the levels of antioxidants in fruits both among crops and more recently among cultivars and breeding materials within a crop (Tables 2.6–2.8). These studies focus on a few classes of compounds with the most frequent being vitamin C, carotenoids, total phenolics, and anthocyanins with a couple of studies looking at the levels of various phenolic compounds among cultivars.

Correlation studies among these various phytochemicals and antioxidant activity have consistently shown that among a range of crops total phenolics and, in berries such as blueberries and blackberries, anthocyanins are well-correlated with antioxidant activity, whereas carotenoids and vitamin C contribute little to the antioxidant

Table 2.6 Antioxidant activity among cultivars within selected fruit crops

Crop	Genotypes	Number	Range of AOA μ g Trolox/100 g FW	Reference
Peach/ nectarine	California cultivars	20	46–1,006 (flesh) (DPPH) 230–1,789 (peel) (DPPH)	Gil et al. (2002)
	Red-fleshed peaches	8	440–1,784 (DPPH)	Cevallos-Casals et al. (2006)
	White-fleshed peaches	4	540–1,096 (DPPH)	Vizzotto et al. (2007)
	Yellow-fleshed peaches	6	437–1,128 (DPPH)	Vizzotto et al. (2007)
	Red-fleshed peaches	9	2,787–13,505 (DPPH)	Vizzotto et al. (2007)
	Segregating progeny	218	227–630 (DPPH)	Cantín et al. (2009)
	California cultivars	20	350–2,250 (DPPH)	Byrne et al. (2009)
	California cultivars and breeding selections	45	1,311–6,471 (DPPH)	Vizzotto et al. (2007)
	Red-flesh plums	14	1,254–3,244 (DPPH)	Cevallos-Casals et al. (2006)
	California cultivars	5	205–518 (flesh) (DPPH) 701–1,314 (peel) (DPPH)	Gil et al. (2002)
Blueberries	California cultivars	6	2,300–8,600 (DPPH)	Byrne et al. (2009)
	High-bush cultivars	6	1,700–3,701 (ORAC)	Prior et al. (1998)
	Rabbiteye cultivars	4	1,390–2,550 (ORAC)	Prior et al. (1998)
	V ashei, rabbiteye cultivar and selections	4	11,100–13,000 (ORAC)	Moyer et al. (2002)
	High-bush cultivars and selections	15	1,900–9,600 (ORAC)	Moyer et al. (2002)
	High-bush cultivars	80	332–582 (ORAC)	Kalt et al. (2001)
	Low-bush cultivars	135	515–901 (ORAC)	Kalt et al. (2001)
	High-bush cultivars	4	379–549 (DPPH)	Giovanelli and Buratti (2009)
			2,130–2,640 (FRAP)	
	High-bush, low-bush cultivars	39		Giongo et al. (2006)
	High-bush and hybrid, rabbiteye cultivars	87	46–311 (ORAC)	Elhenfeldt et al. (2001)
	Breeding materials	52	500–6,300 (MeLO)	Connor et al. (2002b)
	High-bush cultivars	9	2,500–4,300 (MeLO)	Connor et al. (2002a)
	High-bush cultivars	11	2,000–7,900 (FRAP)	Beccaro et al. (2006)

(continued)

Table 2.6 (continued)

Crop	Genotypes	Number	Range of AOA μ g Trolox/100 g FW	Reference
Apples	High-bush cultivars	19	2,780–5,060 (FRAP)	Remberg et al. (2007)
	Cider cultivars and selection	8	Peel: 175–452 (FRAP, ASCE) Flesh: 32–125 (FRAP, ASCE)	Khanizadeh et al. (2008)
	Cultivars	6	Fruit: 335–739 (ABTS)	Vieira et al. (2009)
	Cultivars	11	Peel: 1225–4145 (ABTS) Peel: 1004–3878 (DPPH) Peel: 521–1161 (FRAP) Flesh: 380–961 (ABTS) Flesh: 346–891 (DPPH) Flesh: 140–262 (FRAP)	Vieira et al. (2011)

2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) and 1,1-diphenyl-2-picrylhydrazyl (DPPH) measure the scavenging of free radicals, ORAC measures the oxygen radical absorption capacity using a biologically relevant radical source, FRAP measures the ferric reducing power, MeLO measures the inhibition of peroxyl radical-induced oxidation of linoleic acid. ASCE, measured in ascorbic acid equivalents instead of Trolox equivalents

capacity of the fruit (Vizzotto et al. 2007; Cevallos-Casals et al. 2006; Kalt et al. 2001; Prior et al. 1998; Giovannelli and Buratti 2009; Connor et al. 2002a; Henriquez et al. 2009; Beccaro et al. 2006; Lee et al. 2003).

Antioxidant activity among genotypes has been reported with peach, plum, apple, and blueberry (Table 2.6) using various in vitro methods on phenolic extracts of the fruit. The most commonly used assays are the aqueous-based assays, such as 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) and 1,1-diphenyl-2-picrylhydrazyl (DPPH) which measure the scavenging of free radicals, ORAC which measures the oxygen radical absorption capacity using a biologically relevant radical source, and FRAP which measures the ferric reducing power of the extract. Less frequently, MeLO which measures the inhibition of peroxyl radical-induced oxidation of linoleic acid is used. Several studies with fruit crops have shown that these various methods were correlated among themselves (Thaipong et al. 2006; Connor et al. 2002a, b) and correlated similarly with total phenolics and other phytochemical components being studied (Vieira et al. 2011; Wojdylo et al. 2008).

For apple, peach, plum, and blueberry, there is a wide range in measured antioxidant capacity irrespective of the methodology used (Table 2.6), total phenolics (Table 2.7), and anthocyanins (Table 2.8). Although among commercial cultivars the differences were significant, in some studies that examined breeding materials and other noncommercial germplasm, the range of antioxidant capacity, total phenolics, and/or anthocyanins measured were greatly enlarged (Vizzotto et al. 2007; Cevallos-Casals et al. 2006; Moyer et al. 2002; Conner et al. 2002b). Thus, it is clear that there is variation among genotypes within crops. Currently, there are genotypes within the commercial cultivar mix that have higher levels of antioxidants that could

Table 2.7 Total phenolics among cultivars within selected fruit crops

Crop	Genotypes	Number	Range of phenolics mg/100 g FW	Reference
Peach/ nectarine	California cultivars	20	14–111 (CGA)	Gil et al. (2002)
	Processing cultivars	8	48–80 (CGA)	Chang et al. (2000)
	Red-fleshed peaches	8	100–448 (CGA)	Cevallos-Casals et al. (2006)
	White-, yellow-, red-fleshed peaches	19	137–1,260 (CGA)	Vizzotto et al. (2007)
	Yellow peach, nectarines, white nectarine	13	37–73 (GAE)	Vaio et al. (2008)
	Commercial cultivars	11	14–50 (GAE)	Taravini et al. (2008)
Japanese plum	Segregating progeny	218	13–71 (GAE)	Cantín et al. (2009)
	California cultivars and breeding selections	45	182–898 (CGA)	Vizzotto et al. (2007)
	Red-flesh plums	14	298–563 (CGA)	Cevallos-Casals et al. (2006)
Blueberries	California cultivars	5	42–109 (CGA)	Gil et al. (2002)
	High-bush cultivars	6	181–391 (GAE)	Prior et al. (1998)
	Rabbiteye cultivars	4	230–457 (GAE)	Prior et al. (1998)
	V ashei, rabbiteye cultivar and selections	4	717–961 (GAE)	Moyer et al. (2002)
	High-bush cultivars and selections	15	171–868 (GAE)	Moyer et al. (2002)
	High-bush cultivars	80	165–216 (GAE)	Kalt et al. (2001)
	Low-bush cultivars	135	346–412 (GAE)	Kalt et al. (2001)
	Store bought blueberries	5	292–672 (CGA)	Cevallos-Casals et al. (2003)
	High-bush cultivars	4	251–310 (GAE)	Giovanella and Buratti (2009)
	High-bush, low-bush cultivars	39	187–495 (catechin)	Giongo et al. (2006)
	High-bush and hybrid, rabbiteye cultivars	87	25–199 (GAE)	Ehlenfeldt et al. (2003)
	Breeding materials	52	150–945 (CGA)	Connor et al. (2002b)
	High-bush cultivars	9	401–604 (CGA)	Connor et al. (2002a)
	High-bush cultivars	11	166–459 (GAE)	Beccaro et al. (2006)
Apples	Cultivars	5	170–212 (GAE)	Henriquez et al. (2009)
	Cider cultivars and selection	8	Peel: 101–214 (GAE) Flesh: 23–52 (GAE)	Khanizadeh et al. (2008)
	Cultivars	10	Flesh: 37–90 (HPLC, epicatechin)	McGhie et al. (2005)
	Cultivars	56	Peel: 48–235 (GAE)	Lata et al. (2005)
	Cultivars	6	105–270 (GAE)	Vieira et al. (2009)

(continued)

Table 2.7 (continued)

Crop	Genotypes	Number	Range of phenolics mg/100 g FW	Reference
	Cultivars	11	Peel: 304–713 (GAE) Flesh: 128–212 (GAE)	Vieira et al. (2011)
	Cultivars	8	102–235 (GAE)	Tsao et al. (2003)
	Cultivars	4	Peel: 309–589 (GAE) Flesh: 75–103 (GAE) Fruit: 119–159 (GAE)	Wolfe et al. (2003)

Total phenolics expressed as equivalents of chlorogenic acid (CGA), gallic acid (GAE), catechin, or epicatechin

Table 2.8 Total anthocyanins among cultivars within selected fruit crops

Crop	Genotypes	Number	Range of phenolics mg C3G/100 g FW	Reference
Peach/ nectarine	California cultivars	20	Flesh: 0–23	Tomas-Barberan et al. (2001)
			Peel: 34–273	
	Red-fleshed peaches	8	1–36	Cevallos-Casals et al. (2006)
	White-, yellow-, red-fleshed peaches	19	1–266	Vizzotto et al. (2007)
Japanese plum	Segregating progeny	218	0.1–31	Cantín et al. (2009)
	California cultivars	20	0.5–7	Byrne et al. (2009)
	California cultivars and breeding selections	45	2–611	Vizzotto et al. (2007)
	Red-flesh plums	14	25–175	Cevallos-Casals et al. (2006)
Blueberries	California cultivars	5	Flesh: 0–28 (C3R) Peel: 129–1,615 (C3R)	Tomas-Barberan et al. (2001)
	California cultivars	6	15–105	Byrne et al. (2009)
	High-bush cultivars	6	93–235	Prior et al. (1998)
	Rabbiteye cultivars	4	61–187	Prior et al. (1998)
	V ashei, rabbiteye cultivar and selections	4	242–515	Moyer et al. (2002)
	High-bush cultivars and selections	15	73–430	Moyer et al. (2002)
	High-bush cultivars	80	93–148	Kalt et al. (2001)
	Low-bush cultivars	135	127–210	Kalt et al. (2001)
	Store bought blueberries	5	138–385	Cevallos-Casals and Cisneros- Zevallos (2003)
	High-bush cultivars	4	92–129	Giovanella and Buratti (2009)
	High-bush, low-bush cultivars	39	95–445	Giongo et al. (2006)

(continued)

Table 2.8 (continued)

Crop	Genotypes	Number	Range of phenolics mg C3G/100 g FW	Reference
Apples	High-bush and hybrid, rabbiteye cultivars	87	89–331	Ehlenfeldt et al. (2003)
	Breeding materials	52	1–428	Connor et al. (2002b)
	High-bush cultivars	9	105–236	Connor et al. (2002a)
	High-bush cultivars	11	30–231	Beccaro et al. (2006)
	Cider cultivars and selection	8	Peel: 0–29	Khanizadeh et al. (2008)
	Cultivars	10	Flesh: 0 Fruit: 0–3.7	McGhie et al. (2005)
	Cultivars	56	Peel: 1–56	Lata et al. (2005)
	Cultivars	6	Peel: 5–42 (C3Gal)	Vieira et al. (2009)
	Cultivars	11	Peel: 27–117 (C3Gal)	Vieira et al. (2011)
	Cultivars	8	Peel: 4–21	Tsao et al. (2003)
	Cultivars	4	Peel: 2–27	Wolfe et al. (2003)

Anthocyanins measured as equivalents of cyanidin 3-glucoside (C3G), except for plums in Tomas-Barberan et al. 2001, who used equivalents of cyanidin 3-rutinoside (C3R), and on apples in Vieira et al. 2009, 2011, who used cyanidin 3-galactoside (C3Gal)

be promoted as such and this type of marketing has already been initiated. Furthermore, in the case of peaches, plums, and blueberries, there are also genotypes outside the commercial mix of cultivars that have even higher levels of antioxidants than commercial germplasm indicating the possibility of increasing the levels even more.

Beyond examining the variation in general antioxidant activity or levels of the major classes of antioxidants (total phenolics and anthocyanins), there have been studies examining the ability of genotypes to inhibit proliferation of cancer cells, inhibition of LDL oxidation and other bioactivities in strawberries (Meyers et al. 2003), apples (Yoshizawa et al. 2005; Wolfe et al. 2003; Thompson et al. 2009), blueberries (Yi et al. 2005), peaches, plums (Chang et al. 2000; Byrne et al. 2009), and other fruits. These studies have shown that, as was seen with antioxidant activity and the levels of phytochemicals, genotypes within a crop differed in their bioactivity toward cancer growth or CVD development as measured by various *in vitro* assays. Another crucial observation is that these various bioactivities are not consistently correlated with antioxidant activity, total phenolics, or total anthocyanin content (Byrne et al. 2009; Sun et al. 2002; Liu 2004; Liu 2003; Meyers et al. 2003). This does not indicate that antioxidant activity is not important in preventing these chronic diseases, but rather that there are other mechanisms by which these diseases are regulated and that the phytochemicals within a fruit work both additively and synergistically to affect disease development (Liu et al. 2005a, b).

5 Breeding for Enhanced Phytochemical Levels

Many of the publications that report variation in antioxidants or bioactivities among genotypes within a crop mention that breeding for enhanced health properties is a goal of the breeding program (Vizzotto et al. 2007; Cantín et al. 2009; Connor et al. 2002a, 2005b, c; Vorsa and Polashock 2005; McDougall et al. 2007; Moyer et al. 2002; Kappel 2008; Khanizadeh et al. 2009). Nevertheless, it is not clear how much work is ongoing in the breeding of health-enhanced fruits as a breeder always has many competing objectives to balance. For a cultivar to be successful, it must be productive for the growers and produce high-quality fruit or it will not sell well. Both these traits are complex and are in turn divided into dozens of well-defined traits that the breeder selects for or against. One thing that is clear from various surveys is that whatever health-enhanced cultivar released also has to taste good (Sloan 2008; Byrne 2005).

5.1 Breeding Studies

As discussed previously, there have been a multiplicity of studies that have examined the genotypic variation of antioxidant activity and the level of phytochemicals in fruits of which some examined differences among years (Lata et al. 2005, 2008, 2009; Wojdylo et al. 2008) and between locations (McGhie et al. 2005; Prior et al. 1998; Connor et al. 2002b, c, 2005b, d). In general, although the cultivar effect was large, the antioxidant activity and phytochemical concentrations seen among cultivars frequently varied from year to year and among locations presumably due to differences in climatic, cultural, edaphic, or some other condition.

Breeding studies with blueberry (Connor et al. 2002a) and red raspberry (Connor et al. 2005a, c) estimated the narrow-sense heritability as moderate for antioxidant activity (0.43 and 0.54 for blackberry and red raspberry, respectively) and total phenolic content (0.46 and 0.48 for blackberry and red raspberry, respectively) and moderate to high for total anthocyanin content (0.56 and 0.74 for blackberry and red raspberry, respectively). In red raspberries, the narrow-sense heritability estimates varied from 0.45 to 0.78 for individual anthocyanins. The anthocyanin with the highest concentration (cyanidin 3-sophoroside) had a heritability of 0.56. These moderate to high heritabilities indicate that good progress can be expected in the breeding of blueberry and red raspberry for higher antioxidants (Connor et al. 2005c). In these crops, the year accounted for little of the variance, whereas the importance of the genotype \times year effect differed between the crops with only blueberry having a significant interaction effect.

In peach, a study with 15 progenies done over 3 years indicated that the cross variation explained ~20, ~34, and ~16% of the phenotypic variation seen for antioxidant activity, total phenolics, and total anthocyanins, respectively. In this study, the variation due to the year or the cross \times year effects was not significant (Cantín et al. 2009). This study used commercial germplasm which is limited in the amount

of antioxidant activity, total phenolics, and total anthocyanins as compared to the breeding germplasm available (Tables 2.6–2.8) and it is likely that the genetic component for these traits would be higher if this high antioxidant/phytochemical material was used in the breeding. Although this would facilitate rapid progress in boosting the antioxidant/phytochemical levels of peaches, further analysis would be needed as these materials are lacking in many important commercial traits.

A novel approach to improve the effective anthocyanin levels in fruit was described in cranberry, where the proportion of specific anthocyanins vary with the species. In the cultivated cranberry (*Vaccinium macrocarpon* Ait.), the major antioxidants are galactosides and arabinosides versus glucosides of cyanidin and peonidin as is found in the related species *V. oxycoccus* L. This is important as the glucoside form is more bioavailable than the galactoside and arabinoside forms. Thus, it was shown that it was possible to dramatically increase the proportion of the more bioavailable glucoside form using interspecific hybridization (Vorsa and Palashock 2005).

5.2 *Breeding for Higher Anthocyanins in Tree Fruits*

Berries, such as blueberries, blackberries, and red raspberries, have been touted for their high anthocyanin contents and breeding work indicates that in blueberries and red raspberries the total anthocyanin content is moderately to highly heritable (Connor et al. 2002a, 2005c). In contrast, the commercial cultivars of tree fruits, such as apples, peaches, and kiwi among others, generally have little anthocyanin in the flesh of the fruit and what they have is concentrated in the skin (Table 2.8). Nevertheless, there are variants of these fruit that have red flesh (Cevallos-Casals et al. 2006; Vizzotto et al. 2007; Volz et al. 2009; Jaeger and Harker 2005). In fact, there are red-flesh peaches and plums that have anthocyanin levels equal to or even greater than those reported for commercial blueberry cultivars (Cevallos-Casals et al. 2006; Vizzotto et al. 2007; Byrne et al. 2009). In peach and apple and probably in other normally white-, yellow-, or green-fleshed fruit species, there appear to be one or two major genes that allow the development of anthocyanins in the flesh (Sekido et al. 2010; Werner et al. 1997; Volz et al. 2009). As is seen in the work with peaches and plums, the red-fleshed genotypes vary widely in the total anthocyanins in the fruit (Cevallos-Casals et al. 2006; Vizzotto et al. 2007). Thus, once converted into a red-fleshed genotype, further selection would need to be done to optimize the anthocyanin content as well as multiple other traits essential for commercial success. Currently, there are traditional, advanced selections and newly released red-fleshed peach and nectarine cultivars in Asia, North America, and Europe (Byrne et al. 2009; Pascal, personal communication; Ma, personal communication), red-fleshed commercial cultivars of Japanese plum (Vizzotto et al. 2007), red-fleshed kiwis developed in New Zealand (Jaeger and Harker 2005), and work toward the development of red-fleshed apples in Japan and New Zealand (Sekido et al. 2010; Volz et al. 2009).

5.3 *Breeding Targets: An Assessment*

Multiple breeding programs have explored the levels of phytochemicals, antioxidant activity, and other bioactivities among the genotypes that comprise their breeding germplasm (Tables 2.6–2.8). These, combined with a few breeding studies, clearly indicate that there is sufficient genetic variability to develop cultivars with increased levels of antioxidant activity, total phenolics, and anthocyanins.

Epidemiological studies have indicated that low fruit and vegetable consumption is a risk factor for both cancer and CVD (Chong et al. 2010; Danaei et al. 2005). In the case of CVD, evidence supports the assertion that fruits with higher total phenolics reduce the risk of CVD more than low-phenolic fruits (Chong et al. 2010). Unfortunately, in spite of the thousands of studies which identify extracts or specific compounds that affect the development of chronic diseases, it is not clear which chemicals nor what levels of these chemicals should be the target of breeding programs. In part, this is because the bulk of the work has been done in cell culture model systems which serve to identify potentially useful chemicals and study their mechanisms of action but, due to bioavailability and other issues, not to establish the effective levels in animal model systems or for use in humans. Even the work with small animal models, although better than a cell culture protocol, does not necessarily translate well to a human system (Finley 2005). Furthermore, there are potential synergistic interactions among various phytochemicals which make the situation more complex (Liu 2004; Milde et al. 2007) and consequently more difficult to select a breeding target.

It has been frequently asserted that the consumption of higher levels of antioxidants is good for one's health and many products are sold using this claim. Nevertheless, there is not definitive proof to confirm that supplemental antioxidant consumption reduces the development of chronic disease (Amiot 2009). Thus, more research is needed to identify target phytochemicals and the levels needed to have a beneficial effect on long-term health and the development of chronic diseases. These studies need to compare cultivars with varying levels of phytochemicals as well as specific individual or combination of phytochemicals in animal model and human clinical trials to identify the key targets for the development of truly health-enhanced cultivars of fruit.

References

- Adhami, V. M., Khan, N., and Mukhtar, H. (2009) Cancer chemoprevention by pomegranate: laboratory and clinical evidence. *Nutr Cancer* 61, 811–815.
- Afaq, F., Saleem, M., Krueger, C. G., Reed, J. D., and Mukhtar, H. (2005) Anthocyanin- and hydrolyzable tannin-rich pomegranate fruit extract modulates MAPK and NF-kappaB pathways and inhibits skin tumorigenesis in CD-1 mice. *Int J Cancer* 113, 423–433.
- American Institute for Cancer Research., and World Cancer Research Fund. (2007) Food, nutrition, physical activity and the prevention of cancer : a global perspective : a project of World Cancer Research Fund International, American Institute for Cancer Research, Washington, D.C.

- Amiot, M. J. (2009) Fruit, vegetables, phytochemicals and human health: Past and future. *Acta Hort.* 817, 61–69.
- Aziz, M. H., Afaq, F., and Ahmad, N. (2005) Prevention of ultraviolet-B radiation damage by resveratrol in mouse skin is mediated via modulation in survivin. *Photochem Photobiol* 81, 25–31.
- Barth, S. W., Fahndrich, C., Bub, A., Dietrich, H., Watzl, B., Will, F., Briviba, K., and Rechkemmer, G. (2005) Cloudy apple juice decreases DNA damage, hyperproliferation and aberrant crypt foci development in the distal colon of DMH-initiated rats. *Carcinogenesis* 26, 1414–1421.
- Beaglehole, R., Ebrahim, S., Reddy, S., Voute, J., and Leeder, S. (2007) Prevention of chronic diseases: a call to action. *Lancet* 370, 2152–2157.
- Beccaro, G., Mellano, M., Botta, R., Chiabrando, V. and Bounous, G. (2006) Phenolic and anthocyanin content and antioxidant activity in fruits of bilberry (*Vaccinium myrtillus* L.) and of highbush blueberry (*V. corymbosum* L.) cultivars in north western Italy. *Acta Hort.* 715, 553–557.
- Bertelli, A. A., and Das, D. K. (2009) Grapes, wines, resveratrol, and heart health. *J Cardiovasc Pharmacol* 54, 468–476.
- Bodet, C., Chandad, F., and Grenier, D. (2006) Anti-inflammatory activity of a high-molecular-weight cranberry fraction on macrophages stimulated by lipopolysaccharides from periodontopathogens. *J Dent Res* 85, 235–239.
- Brandelli, C., Giordani, R., De Carli, G., and Tasca, T. (2009) Indigenous traditional medicine: in vitro anti-giardial activity of plants used in the treatment of diarrhea. *Parasitology Research* 104, 1345–1349.
- Buchner, F. L., Bueno-de-Mesquita, H. B., Ros, M. M., Kampman, E., Egevad, L., Overvad, K., Tjønneland, A., Roswall, N., Clavel-Chapelon, F., Boutron-Ruault, M. C., Touillaud, M., Kaaks, R., Chang-Claude, J., Boeing, H., Weikert, S., Trichopoulou, A., Naska, A., Benetou, V., Palli, D., Sieri, S., Vineis, P., Tumino, R., Panico, S., van Duijnhoven, F. J., Peeters, P. H., van Gils, C. H., Lund, E., Gram, I. T., Sanchez, M. J., Jakszyn, P., Larranaga, N., Ardanaz, E., Navarro, C., Rodriguez, L., Manjer, J., Ehrnstrom, R., Hallmans, G., Ljungberg, B., Key, T. J., Allen, N. E., Khaw, K. T., Wareham, N., Slimani, N., Jenab, M., Boffetta, P., Kiemeny, L. A., and Riboli, E. (2011) Variety in vegetable and fruit consumption and risk of bladder cancer in the European Prospective Investigation into Cancer and Nutrition. *Int. J. Cancer* 128(12), 2971–2979.
- Byrne, D. H. (2005) Trends in stone fruit cultivar development. *HortTechnology*, 15(3), 494–500.
- Byrne, D. H., Noratto, G., Cisneros Zevallos, L., Porter, W. and Vizzotto, M. (2009) Health benefits of peaches and plums. *Acta Hort.* 841, 267–274.
- Cade, J. E., Frear, L., and Greenwood, D. C. (2006) Assessment of diet in young children with an emphasis on fruit and vegetable intake: using CADET--Child and Diet Evaluation Tool, *Public Health Nutr* 9, 501–508.
- Cantín, C.M., Moreno, M.A. and Gogorcena Y (2009) Evaluation of the antioxidant capacity, phenolic compounds and vitamin C content of different peach and nectarine [*Prunus persica* (L.) Batsch] breeding progenies. *J Agric Food Chem* 57, 4586–4592.
- Casto, B. C., Kresty, L. A., Kraly, C. L., Pearl, D. K., Knobloch, T. J., Schut, H. A., Stoner, G. D., Mallery, S. R., and Weghorst, C. M. (2002) Chemoprevention of oral cancer by black raspberries. *Anticancer Res* 22, 4005–4015.
- Cevallos-Casals, B., Byrne, D., Okie, W. R. and Cisneros-Zevallos, L. (2006) Selecting new peach and plum genotypes rich in phenolic compounds and enhanced functional properties. *Food Chem.* 96, 273–280.
- Cevallos-Casals, B. and Cisneros-Zevallos, L. (2003) Stoichiometric and kinetic studies of phenolic antioxidants from Andean purple corn and red-fleshed sweetpotato. *J Agric. Food Chem.* 51, 3313–3319.
- Chang, S., Tan, C., Frankel, E. N. and Barrett, D. M. (2000) Low-density lipoprotein antioxidant activity of phenolic compounds and polyphenol oxidase activity in selected clingstone peach cultivars. *J. Agric. Food Chem.* 48, 147–151.
- Chen, T., Hwang, H., Rose, M. E., Nines, R. G., and Stoner, G. D. (2006) Chemopreventive properties of black raspberries in N-nitrosomethylbenzylamine-induced rat esophageal tumorigenesis: down-regulation of cyclooxygenase-2, inducible nitric oxide synthase, and c-Jun. *Cancer Res* 66, 2853–2859.

- Chong, M., Macdonald, R. and Lovegrove, J. (2010) Fruit polyphenols and CVD risk: a review of human intervention studies. *Brit. J. Nutrition*. 104, s28–s39.
- Chou, E. J., Keevil, J. G., Aeschlimann, S., Wiebe, D. A., Folts, J. D., and Stein, J. H. (2001) Effect of ingestion of purple grape juice on endothelial function in patients with coronary heart disease. *Am J Cardiol* 88, 553–555.
- Connor, A. M., Luby, J. J. and Tong, C. (2002a) Variation and heritability estimates for antioxidant activity, total phenolic content, and anthocyanin content in blueberry progenies. *J. Amer. Soc. Hort. Sci.* 127,82–88.
- Connor, A. M., Luby, J. J., Tong, C., Finn, C. E. and Hancock, J. F. (2002b) Genotypic and environmental variation in antioxidant activity, total phenolic content, and anthocyanin content among blueberry cultivars. *J Amer. Soc. Hort. Sci.* 127:89–97.
- Connor, A. M., Stephens, M. J., Hall, H. K., and Alspach, P. A. (2005a) Variation and heritabilities of antioxidant activity and total phenolic content estimated from a red raspberry factorial experiment. *J. Amer. Soc. Hort. Sci.* 130, 403–411.
- Connor, A. M., Finn, C. E., and Alspach, P. E. (2005b) Genotypic and environmental variation in antioxidant activity and total phenolic content among blackberry and hybridberry cultivars. *J. Amer. Soc. Hort. Sci.* 130, 527–533.
- Connor, A. M., McGhie, T. K., Stephens, M. J., Hall, H. K., and Alspach, P. A. (2005c) Variation and heritability estimates of anthocyanins and their relationship to antioxidant activity in a red raspberry factorial mating design. *J. Amer. Soc. Hort. Sci.* 130, 535–542.
- Connor, A. M., Finn, C. E., McGhie, T. K., and Alspach, P. A. (2005d) Genetic and environmental variation in anthocyanins and their relationship to antioxidant activity in blackberry and hybridberry cultivars. *J. Amer. Soc. Hort. Sci.* 130, 680–687.
- Danaei, G., Vander Hoom, S., Lopez, A., Murray, C., Ezzati, M., and the Comparative Risk Assessment collaborating group (Cancers). (2005) Causes of cancer in the world: comparative risk assessment of nine behavioural and environmental risk factors. *Lancet* 366, 1784–1793.
- Davidson, M. H., Maki, K. C., Dicklin, M. R., Feinstein, S. B., Witchger, M., Bell, M., McGuire, D. K., Provost, J. C., Liker, H., and Aviram, M. (2009) Effects of consumption of pomegranate juice on carotid intima-media thickness in men and women at moderate risk for coronary heart disease. *Am J Cardiol* 104, 936–942.
- de Souza, M. O., Silva, M., Silva, M. E., Oliveira Rde, P., and Pedrosa, M. L. (2010) Diet supplementation with acai (*Euterpe oleracea* Mart.) pulp improves biomarkers of oxidative stress and the serum lipid profile in rats. *Nutrition* 26, 804–810.
- Dillard, C. J. and German, J. B. (2000) Phytochemicals: nutraceuticals and human health. *J. Sci. Food Agric.* 80, 1744–1756.
- Drogoudi, P., Michailidis, Z., and Pantelidis, G. (2008) Peel and flesh antioxidant content and harvest quality characteristics of seven apple cultivars. *Scientia Hort.* 115, 149–153.
- Duncan, F. J., Martin, J. R., Wulff, B. C., Stoner, G. D., Tober, K. L., Oberyszyn, T. M., Kusewitt, D. F., and Van Buskirk, A. M. (2009) Topical treatment with black raspberry extract reduces cutaneous UVB-induced carcinogenesis and inflammation. *Cancer Prev Res (Phila)* 2, 665–672.
- Durak, I., Cetin, R., Devrim, E., and Erguder, I. B. (2005) Effects of black grape extract on activities of DNA turn-over enzymes in cancerous and non cancerous human colon tissues. *Life Sci* 76, 2995–3000.
- Fini, L., Piazzzi, G., Daoud, Y., Selgrad, M., Maegawa, S., Garcia, M., Fogliano, V., Romano, M., Graziani, G., Vitaglione, P., Carmack, S.W., Gasbarrini, A., Genta, R.M., Issa, J.P., Boland, C.R., and Ricciardiello, L. (2011) Chemoprevention of intestinal polyps in ApcMin/+ mice fed western or balanced diets by drinking Annurca apple polyphenol extract. *Cancer Prev Res (Phila)*. 2011 Mar 7. [Epub ahead of print] PubMed PMID: 21383028.
- Finley, J. W. (2005) Bioactive compounds and designer plant foods: The need for clear guidelines to evaluate potential benefits to human health. *Chronica Horticulturae* 45(3), 6–11.
- Freedman, J. E., Parker, C., 3rd, Li, L., Perlman, J. A., Frei, B., Ivanov, V., Deak, L. R., Iafrati, M. D., and Folts, J. D. (2001) Select flavonoids and whole juice from purple grapes inhibit platelet function and enhance nitric oxide release. *Circulation* 103, 2792–2798.

- George, S. M., Park, Y., Leitzmann, M. F., Freedman, N. D., Dowling, E. C., Reedy, J., Schatzkin, A., Hollenbeck, A., and Subar, A. F. (2009) Fruit and vegetable intake and risk of cancer: a prospective cohort study. *Am J Clin Nutr* 89, 347–353.
- Gerhauser, C. (2008) Cancer chemopreventive potential of apples, apple juice, and apple components. *Planta Med* 74(13), 1608–24.
- Gil, M. I., Tomas-Barberan, F. A., Hess-Pierce, B., and Kader, A. A. (2002) Antioxidant capacities, phenolic compounds, carotenoids, and vitamin C contents of nectarine, peach, and plum cultivars from California. *J. Agric. Food Chem* 50, 4976–4982.
- Giongo, L., Ieri, F., Vrhovsek, U., Grisenti, M., Mattivi, F. and Eccher, M. (2006) Characterization of *Vaccinium* cultivars: Horticultural and antioxidant profile. *Acta Hort.* 715, 147–151.
- Giovanelli, G. and Buratti, S. (2009) Comparison of polyphenolic composition and antioxidant activity of wild Italian blubberies and some cultivated varieties. *Food Chem.* 112, 903–908.
- God, J. M., Tate, P., and Larcom, L. L. (2007) Anticancer effects of four varieties of muscadine grape. *J Med Food* 10, 54–59.
- Gosse, F., Guyot, S., Roussi, S., Lobstein, A., Fischer, B., Seiler, N., and Raul, F. (2005) Chemopreventive properties of apple procyanidins on human colon cancer-derived metastatic SW620 cells and in a rat model of colon carcinogenesis. *Carcinogenesis* 26, 1291–1295.
- Gupta, K., Chou, M. Y., Howell, A., Wobbe, C., Grady, R., and Stapleton, A. E. (2007) Cranberry products inhibit adherence of p-fimbriated *Escherichia coli* to primary cultured bladder and vaginal epithelial cells. *J Urol* 177, 2357–2360.
- Hadipour-Jahromy, M., and Mozaffari-Kermani, R. (2010) Chondroprotective effects of pomegranate juice on monoiodoacetate-induced osteoarthritis of the knee joint of mice. *Phytother Res* 24, 182–185.
- Harris, G. K., Gupta, A., Nines, R. G., Kresty, L. A., Habib, S. G., Frankel, W. L., LaPerle, K., Gallaher, D. D., Schwartz, S. J., and Stoner, G. D. (2001) Effects of lyophilized black raspberries on azoxymethane-induced colon cancer and 8-hydroxy-2'-deoxyguanosine levels in the Fischer 344 rat. *Nutr Cancer* 40, 125–133.
- Henriquez, C., Almonacid, S., Escobar, B., Chiffelle, I., Gómez, M. and Speisky, H. (2009) Antioxidant content and activity in different structures of five apple cultivars grown in Chile. *Acta Hort* 841, 275–280.
- Hora, J. J., Maydew, E. R., Lansky, E. P., and Dwivedi, C. (2003) Chemopreventive effects of pomegranate seed oil on skin tumor development in CD1 mice. *J Med Food* 6, 157–161.
- Howell, A. B. (2007) Bioactive compounds in cranberries and their role in prevention of urinary tract infections. *Molecular Nutrition & Food Research* 51, 732–737.
- Huang, W. Y., Cai, Y. Z., and Zhang, Y. (2010) Natural phenolic compounds from medicinal herbs and dietary plants: potential use for cancer prevention. *Nutr Cancer* 62, 1–20.
- Hudson, T. S., Hartle, D. K., Hursting, S. D., Nunez, N. P., Wang, T. T. Y., Young, H. A., Arany, P., and Green, J. E. (2007) Inhibition of prostate cancer growth by muscadine grape skin extract and resveratrol through distinct mechanisms. *Cancer Research* 67, 8396–8405.
- Hung, H. C., Joshipura, K. J., Jiang, R., Hu, F. B., Hunter, D., Smith-Warner, S. A., Colditz, G. A., Rosner, B., Spiegelman, D., and Willett, W. C. (2004) Fruit and vegetable intake and risk of major chronic disease. *J Natl Cancer Inst* 96, 1577–1584.
- Ilbey, Y. O., Ozbek, E., Simsek, A., Cekmen, M., Somay, A., and Tasci, A. I. (2009) Effects of pomegranate juice on hyperoxaluria-induced oxidative stress in the rat kidneys. *Ren Fail* 31, 522–531.
- Jaeger, S. and Harker, F. (2005) Consumer evaluation of novel kiwifruit: willingness-to-pay. *J. Sci. Food Agric.* 85, 2519–2526.
- Jang, M., Cai, L., Udeani, G. O., Slowing, K. V., Thomas, C. F., Beecher, C. W., Fong, H. H., Farnsworth, N. R., Kinghorn, A. D., Mehta, R. G., Moon, R. C., and Pezzuto, J. M. (1997) Cancer chemopreventive activity of resveratrol, a natural product derived from grapes. *Science* 275, 218–220.
- Jensen, G. S., Wu, X., Patterson, K. M., Barnes, J., Carter, S. G., Scherwitz, L., Beaman, R., Endres, J. R., and Schauss, A. G. (2008) In vitro and in vivo antioxidant and anti-inflammatory

- capacities of an antioxidant-rich fruit and berry juice blend. Results of a pilot and randomized, double-blinded, placebo-controlled, crossover study. *J Agric Food Chem* 56, 8326–8333.
- Johnson, I. T. (2007) Phytochemicals and cancer, *Proceedings of the Nutrition Society*. 66, 207–215.
- Joseph, J. A., Shukitt-Hale, B., Denisova, N. A., Bielinski, D., Martin, A., McEwen, J. J., and Bickford, P. C. (1999) Reversals of age-related declines in neuronal signal transduction, cognitive, and motor behavioral deficits with blueberry, spinach, or strawberry dietary supplementation. *J Neurosci* 19, 8114–8121.
- Kalt, W., Ryan, D., Duy, J., Prior, R., Ehlenfeldt, M. and Vander Kloet, S. (2001) Interspecific variation in anthocyanins, phenolics, and antioxidant capacity among genotypes of highbush and lowbush blueberries (*Vaccinium* Section *cyanococcus* spp.) *J. Agric. Food Chem.* 49, 4761–4767.
- Kappel, F. (2008) Breeding cherries in the ‘New World’. *Acta Hort.* 795, 59–69.
- Key, T. J. (2011) Fruit and vegetables and cancer risk. *Br J Cancer*. 104(1), 6–11.
- Khanizadeh, S., Tsao, R., Rekika, D., Yang, R., Charles, M. T., and Rupasinghe, H. P. V. (2008) Polyphenol composition and total antioxidant capacity of selected apple genotypes for processing. *J. Food Comp. Anal.* 21, 396–401.
- Khanizadeh, S., Tsao, R., Rekika, D., Yang, R., Charles, M. T., and Rupasinghe, H. P. V. (2009) Advances in fruit breeding in Eastern Canada – Role of phytochemicals in designing specialty fruits. *Acta Hort* 814, 205–207.
- Khan, N., Afaq, F., Kweon, M. H., Kim, K., and Mukhtar, H. (2007) Oral consumption of pomegranate fruit extract inhibits growth and progression of primary lung tumors in mice. *Cancer Res* 67, 3475–3482.
- Kim, H., Hall, P., Smith, M., Kirk, M., Prasain, J. K., Barnes, S., and Grubbs, C. (2004) Chemoprevention by grape seed extract and genistein in carcinogen-induced mammary cancer in rats is diet dependent. *J Nutr* 134, 3445S–3452S.
- Kim, T. J., Weng, W. L., Silva, J. L., Jung, Y. S., and Marshall, D. (2010) Identification of natural antimicrobial substances in red muscadine juice against *Cronobacter sakazakii*. *J Food Sci* 75, M150–154.
- Kim, T. J., Weng, W. L., Stojanovic, J., Lu, Y., Jung, Y. S., and Silva, J. L. (2008) Antimicrobial effect of water-soluble muscadine seed extracts on *Escherichia coli* O157:H. *J Food Prot* 71, 1465–1468.
- Koch, T.C., Briviba, K., Watzl, B., Fährndrich, C., Bub, A., Rechkemmer, G., Barth, S.W. (2009) Prevention of colon carcinogenesis by apple juice in vivo: impact of juice constituents and obesity. *Mol Nutr Food Res.* 53(10), 1289–302.
- Kohno, H., Suzuki, R., Yasui, Y., Hosokawa, M., Miyashita, K., and Tanaka, T. (2004) Pomegranate seed oil rich in conjugated linolenic acid suppresses chemically induced colon carcinogenesis in rats. *Cancer Sci* 95, 481–486.
- Konijeti, R., Henning, S., Moro, A., Sheikh, A., Elashoff, D., Shapiro, A., Ku, M., Said, J. W., Heber, D., Cohen, P., and Aronson, W. J. (2010) Chemoprevention of prostate cancer with lycopene in the TRAMP model. *Prostate* 70, 1547–1554.
- Krikorian, R., Nash, T. A., Shidler, M. D., Shukitt-Hale, B., and Joseph, J. A. (2010) Concord grape juice supplementation improves memory function in older adults with mild cognitive impairment. *Br J Nutr* 103, 730–734.
- Lala, G., Malik, M., Zhao, C., He, J., Kwon, Y., Giusti, M. M., and Magnuson, B. A. (2006) Anthocyanin-rich extracts inhibit multiple biomarkers of colon cancer in rats. *Nutr Cancer* 54, 84–93.
- Larsson, S. C., Andersson, S. O., Johansson, J.E., and Wolk, A. (2008) Fruit and Vegetable Consumption and Risk of Bladder Cancer: A Prospective Cohort Study. *Cancer Epidemiol Biomarkers Prev* 17, 2519–2522.
- Lata, B., Przeradzka, M., and Binkowska, M. (2005) Great differences in antioxidant properties exist between 56 apple cultivars and vegetation seasons. *J. Agric. Food Chem* 53, 8970–8978.
- Lata, B. (2008) Apple peel antioxidant status in relation to genotype, storage type and time. *Scientia Hort* 117, 45–52.
- Lata, B., Trampczynska, A., and Paczesna, J. (2009) Cultivar variation in apple peel and whole fruit phenolic composition. *Scientia Hort* 121, 176–181.

- Lee, K. W., Kim, Y. J., Kim, D., Lee, H. J., and Chang, Y. L. (2003) Major phenolics in apple and their contribution to the total antioxidant capacity. *J. Agric. Food Chem.* 51, 6516–6520.
- Leifert, W. R., and Abeywardena, M. Y. (2008) Grape seed and red wine polyphenol extracts inhibit cellular cholesterol uptake, cell proliferation, and 5-lipoxygenase activity. *Nutr Res* 28, 842–850.
- Leiro, J., Arranz, J. A., Fraiz, N., Sanmartin, M. L., Quezada, E., and Orallo, F. (2005) Effect of cis-resveratrol on genes involved in nuclear factor kappa B signaling. *Int Immunopharmacol* 5, 393–406.
- Lewis, J. E., Soler-Vila, H., Clark, P. E., Kresty, L. A., Allen, G. O., and Hu, J. J. (2009) Intake of plant foods and associated nutrients in prostate cancer risk. *Nutr Cancer* 61, 216–224.
- Li, Z. G., Hong, T., Shimada, Y., Komoto, I., Kawabe, A., Ding, Y., Kaganai, J., Hashimoto, Y., and Imamura, M. (2002) Suppression of N-nitrosomethylbenzylamine (NMBA)-induced esophageal tumorigenesis in F344 rats by resveratrol. *Carcinogenesis* 23, 1531–1536.
- Lippi, G., Franchini, M., Favaloro, E. J., and Targher, G. (2010) Moderate Red Wine Consumption and Cardiovascular Disease Risk: Beyond the “French Paradox”. *Semin Thromb Hemost* 31, 059,070.
- Liu, L., Li, Y.H., Niu, Y.B., Sun, Y., Guo, Z.J., Li, Q., Li, C., Feng, J., Cao, S.S., Mei, Q.B. (2010) An apple oligogalactan prevents against inflammation and carcinogenesis by targeting LPS/TLR4/NF- κ B pathway in a mouse model of colitis-associated colon cancer. *Carcinogenesis* 31(10), 1822–32.
- Liu, R. H. (2003) Health benefits of fruits and vegetables are from additive and synergistic combinations of phytochemicals. *Am.J. Clin. Nutr.* 78 (Suppl.), 517s–520s.
- Liu, R. H. (2004) Potential synergy of phytochemicals in cancer prevention: mechanism of action. *J. Nutr.* 134, 3479S–3485S.
- Liu, R. H., Liu, J. and Chen, B. (2005) Apples prevent mammary tumors in rats. *J. Agric. Food Chem.* 53, 2341–2343.
- Liu, Z., Schwimer, J., Liu, D., Greenway, F. L., Anthony, C. T., and Woltering, E. A. (2005) Black raspberry extract and fractions contain angiogenesis inhibitors, *J Agric Food Chem* 53, 3909–3915.
- Mallery, S. R., Zwick, J. C., Pei, P., Tong, M., Larsen, P. E., Shumway, B. S., Lu, B., Fields, H. W., Mumper, R. J., and Stoner, G. D. (2008) Topical application of a bioadhesive black raspberry gel modulates gene expression and reduces cyclooxygenase 2 protein in human premalignant oral lesions. *Cancer Res* 68, 4945–4957.
- Martineau, L. C., Couture, A., Spoor, D., Benhaddou-Andaloussi, A., Harris, C., Meddah, B., Leduc, C., Burt, A., Vuong, T., Mai Le, P., Prentki, M., Bennett, S. A., Arnason, J. T., and Haddad, P. S. (2006) Anti-diabetic properties of the Canadian lowbush blueberry *Vaccinium angustifolium* Ait. *Phytomedicine* 13, 612–623.
- Matchett, M. D., MacKinnon, S. L., Sweeney, M. I., Gottschall-Pass, K. T., and Hurta, R. A. (2005) Blueberry flavonoids inhibit matrix metalloproteinase activity in DU145 human prostate cancer cells. *Biochem Cell Biol* 83, 637–643.
- Mattiello, T., Trifiro, E., Jotti, G. S., and Pulcinelli, F. M. (2009) Effects of pomegranate juice and extract polyphenols on platelet function. *J Med Food* 12, 334–339.
- Mazza, G., and Miniati, E. (1993) Anthocyanins in fruits, vegetables and grains, CRC Press Inc., Boca Raton.
- McDougall, G., Dobson, P., Shpiro, F. Smith, P., Stewart, D. and Fyffe, S. (2007) Assessing bio-availability of soft fruit polyphenols in vitro. *Acta Hort.* 744, 135–148.
- McGhie, T., Hunt, M., and Barnet, L. (2005) Cultivar and growing region determine the antioxidant polyphenolic concentration and composition of apples grown in New Zealand. *J. Agric. Food Chem.* 53, 3065–3070.
- Mehta, R., and Lansky, E. P. (2004) Breast cancer chemopreventive properties of pomegranate (*Punica granatum*) fruit extracts in a mouse mammary organ culture. *Eur J Cancer Prev* 13, 345–348.
- Meyers, K. J., Watkins, C., Pritts, M. and Lu, R. H. (2003) Antioxidant and antiproliferative activities of strawberries. *J Agric. Food Chem.* 51, 6887–6892.

- Milde, J., Eistner, and Graßmann, J. (2007) Synergistic effects of phenolics and carotenoids on human low-density lipoprotein oxidation, *Mol. Nutr. Food Res.* 51, 956–961.
- Montrose, D. C., Horelik, N. A., Madigan, J. P., Stoner, G. D., Wang, L. S., Bruno, R. S., Park, H. J., Giardina, C., and Rosenberg, D. W. (2011) Anti-inflammatory effects of freeze-dried black raspberry powder in ulcerative colitis. *Carcinogenesis* 32(3), 343–50.
- Moyer, R., Hummer, K., Finn, C., Frei, B. and Wrolstad, R. (2002) Anthocyanins, phenolics, and antioxidant capacity in diverse small fruits: *Vaccinium*, *Rubus*, and *Ribes*. *J. Agric. Food Chem.* 50:519–525.
- Nanney, M. S., Schermbeck, R., and Haire-Joshu, D. (2007) Examination of the adherence to the “5 A Day the Color Way” campaign among parents and their preschool children. *J Cancer Educ* 22, 177–180.
- Neto, C. C. (2007) Cranberry and blueberry: evidence for protective effects against cancer and vascular diseases. *Mol Nutr Food Res* 51, 652–664.
- Newman, D. J., and Cragg, G. M. (2007) Natural products as sources of new drugs over the last 25 years. *J Nat Prod* 70, 461–477.
- Olien, W. C. (1990) Muscadine: A classic southeastern fruit. *Hortscience* 25, 726–831.
- Orak, H. H. (2009) Evaluation of antioxidant activity, colour and some nutritional characteristics of pomegranate (*Punica granatum* L.) juice and its sour concentrate processed by conventional evaporation. *International Journal of Food Sciences and Nutrition* 60, 1–11.
- Pan, M. H., Ghai, G., and Ho, C. T. (2008) Food bioactives, apoptosis, and cancer. *Mol Nutr Food Res* 52, 43–52.
- Pantuck, A. J., Leppert, J. T., Zomorodian, N., Aronson, W., Hong, J., Barnard, R. J., Seeram, N., Liker, H., Wang, H., Elashoff, R., Heber, D., Aviram, M., Ignarro, L., and Belldgrun, A. (2006) Phase II study of pomegranate juice for men with rising prostate-specific antigen following surgery or radiation for prostate cancer. *Clin Cancer Res* 12, 4018–4026.
- Pollack, S. and A. Perez. 2008. Fruit and Tree Nuts Situation and Outlook Yearbook 2008. Market and Trade Economics Division, Economic Research Service, U. S. Department of Agriculture, October 2008, FTS-2008.
- Prior, R., Cao, G., Martin, A., Sofic, E., McEwen, J., O’Brien, C., Lischer, N., Ehlenfeldt, M., Kalt, W., Krewer, G., and Mainland, C. M. (1998) Antioxidant capacity as influenced by total phenolic and anthocyanin content, maturity, and variety of *Vaccinium* species. *J. Agric. Food Chem.* 46, 2886–2693.
- Raina, K., Singh, R. P., Agarwal, R., and Agarwal, C. (2007) Oral grape seed extract inhibits prostate tumor growth and progression in TRAMP mice. *Cancer Res* 67, 5976–5982.
- Rajakangas, J., Misikangas, M., Paivarinta, E., and Mutanen, M. (2008) Chemoprevention by white currant is mediated by the reduction of nuclear beta-catenin and NF-kappaB levels in Min mice adenomas. *Eur J Nutr* 47, 115–122.
- Rao, A. V., and Snyder, D. M. (2010) Raspberries and human health: a review, *J Agric Food Chem* 58, 3871–3883.
- Remberg, S., Mage, F., Haffner, K. and Blomhoff, R. (2007) Highbush blueberries *Vaccinium corymbosum* L., raspberries *Rubus idaeus* L. and black currants *Ribes nigrum* L. - influence of cultivars on antioxidant activity and other quality parameters. *Acta Hort.* 744, 259–265.
- Renaud, S., and de Lorgeril, M. (1992) Wine, alcohol, platelets, and the French paradox for coronary heart disease. *The Lancet* 339, 1523–1526.
- Rice-Evans, C. A., Miller, N. J., and Paganga, G. (1996) Structure-antioxidant activity relationships of flavonoids and phenolic acids. *Free Radic Biol Med* 20, 933–956.
- Rommel, A., and Wrolstad, R. E. (1993) Ellagic acid content of red raspberry juice as influenced by cultivar, processing, and environmental factors. *Journal of Agricultural and Food Chemistry* 41, 1951–1960.
- Ruel, G., and Couillard, C. (2007) Evidences of the cardioprotective potential of fruits: the case of cranberries. *Mol Nutr Food Res* 51, 692–701.
- Sautebin, L., Rossi, A., Serraino, I., Dugo, P., Di Paola, R., Mondello, L., Genovese, T., Britti, D., Peli, A., Dugo, G., Caputi, A. P., and Cuzzocrea, S. (2004) Effect of anthocyanins contained in

- a blackberry extract on the circulatory failure and multiple organ dysfunction caused by endotoxin in the rat. *Planta Med* 70, 745–752.
- Schauss, A. G., Wu, X., Prior, R. L., Ou, B., Patel, D., Huang, D., and Kababick, J. P. (2006) Phytochemical and nutrient composition of the freeze-dried Amazonian palm berry, *Euterpe oleraceae* Mart. (Acai), *Journal of Agricultural and Food Chemistry* 54, 8598–8603.
- Sekido, K., Hayashi, Y., Yamada, K., Shiratake, K., Matsumoto, S., Macjima, T., and Komatsu, H. (2010) Efficient breeding system for red-fleshed apple based on linkage with S3-RNase allele in 'Pink Pearl'. *HortScience* 45, 534–537.
- Sengottuvelan, M., Viswanathan, P., and Nalini, N. (2006) Chemopreventive effect of trans-resveratrol—a phytoalexin against colonic aberrant crypt foci and cell proliferation in 1,2-dimethylhydrazine induced colon carcinogenesis. *Carcinogenesis* 27, 1038–1046.
- Shukitt-Hale, B., Carey, A., Simon, L., Mark, D. A., and Joseph, J. A. (2006) Effects of Concord grape juice on cognitive and motor deficits in aging. *Nutrition* 22, 295–302.
- Shukitt-Hale, B., Cheng, V., and Joseph, J. A. (2009) Effects of blackberries on motor and cognitive function in aged rats. *Nutr Neurosci* 12, 135–140.
- Shukitt-Hale, B., Lau, F. C., and Joseph, J. A. (2008) Berry fruit supplementation and the aging brain. *J Agric Food Chem* 56, 636–641.
- Sloan, E. (2006) Top 10 functional food trends. *FoodTechnology* 04.06, 23–34.
- Sloan, E. (2008) The top 10 functional food trends. *FoodTechnology* 04.08, 25–35.
- Stacewicz-Sapuntzakis, M., Bowen, P.E., Hussain, E.A., Damayanti-Wood, B.I., and Farnsworth, N.R. (2001) Chemical composition and potential health effects of prunes: a functional food? *Crit Rev Food Sci Nutr* 41(4), 251–86.
- Stoner, G. D., Dombkowski, A. A., Reen, R. K., Cukovic, D., Salagrama, S., Wang, L. S., and Lechner, J. F. (2008) Carcinogen-altered genes in rat esophagus positively modulated to normal levels of expression by both black raspberries and phenylethyl isothiocyanate. *Cancer Res* 68, 6460–6467.
- Stoner, G. D., Wang, L. S., Seguin, C., Rocha, C., Stoner, K., Chiu, S., and Kinghorn, A. D. (2010) Multiple berry types prevent N-nitrosomethylbenzylamine-induced esophageal cancer in rats. *Pharm Res* 27, 1138–1145.
- Sun, J., Chu, Y. -F., Wu, X. and Liu, R. H. (2002) Antioxidant and antiproliferative activities of common fruits. *J Agric Food Chem* 50, 7449–7454.
- Sun, G. Y., Xia, J., Draczynska-Lusiak, B., Simonyi, A., and Sun, A. Y. (1999) Grape polyphenols protect neurodegenerative changes induced by chronic ethanol administration. *Neuroreport* 10, 93–96.
- Takachi, R., Inoue, M., Ishihara, J., Kurahashi, N., Iwasaki, M., Sasazuki, S., Iso, H., Tsubono, Y., and Tsugane, S. (2008) Fruit and vegetable intake and risk of total cancer and cardiovascular disease: Japan Public Health Center-Based Prospective Study. *Am J Epidemiol* 167, 59–70.
- Tate, P., God, J., Bibb, R., Lu, Q., and Larcom, L. L. (2004) Inhibition of metalloproteinase activity by fruit extracts. *Cancer Lett* 212, 153–158.
- Thaipong, K., Boonprakob, U., Crosby, K., Cisneros-Zevallos, L., and Byrne, D. H. (2006) Comparison of ABTS, DPPH, FRAP, and ORAC assays for estimating antioxidant activity from guava fruit extracts. *J. Food Composition and Analysis* 19, 669–675.
- Thompson, M., Stushnoff, C., McGinley, J., and Thompson, H. (2009) In vitro measures used to predict anticancer activity of apple cultivars and their comparison to outcomes from a rat model of experimentally induced breast cancer. *Nutrition and Cancer* 61, 510–517.
- Tomas-Barberan, F. A., Gil, M. I., Cremin, P., Waterhouse, A. L., Hess-Pierce, B., and Kader, A. A. (2001) HPLC-DAD-ESIMS analysis of phenolic compounds in nectarines, peaches, and plums. *J Agric Food Chem* 49, 4748–4760.
- Tovar, A. M., Cesar, D. C., Leta, G. C., and Mourao, P. A. (1998) Age-related changes in populations of aortic glycosaminoglycans: species with low affinity for plasma low-density lipoproteins, and not species with high affinity, are preferentially affected. *Arterioscler Thromb Vasc Biol* 18, 604–614.

- Tsao, R., Yang, R., Younf, J. C., and Zhu, H. (2003) Polyphenolic profiles in eight apple cultivars using high-performance chromatography (HPLC). *J. Agric. Food Chem.* 51, 6347–6353.
- Vainio, H., and Weiderpass, E. (2006) Fruit and vegetables in cancer prevention, *Nutr. Cancer* 54, 111–142.
- Vieira, F., Borges, G., Copetti, C., Amboni, R., Denardi, F., and Fett, R. (2009) Physico-chemical and antioxidant properties of six apple cultivars (*Malus domestica* Borkh) grown in southern Brazil. *Scientia Hort.* 122, 421–425.
- Vieira, F., Borges, G., Copetti, C., Di Pietro, P., Nunes, E., and Fett, R. (2011) Phenolic compounds and antioxidant activity of the apple flesh and peel of eleven cultivars grown in Brazil. *Scientia Hort.* 128, 261–266.
- Vizzotto, M., Cisneros, L., Okie, W. R., Ramming, D. W., and Byrne, D. H. (2007) Large variation found in the phytochemical content and antioxidant activity of peach and plum germplasm. *J. Amer. Soc. Hort. Sci.*, 132: 334–340.
- Volz, R., Oraguzie, N., Whitworth, C., How, N. Change, D., Carlisle, C., Gardiner, S., Rikkerink, E., and Lawrence, T. (2009) Breeding for red flesh colour in apple: progress and challenges. *Acta Hort.* 841:337–342.
- Vorsa, N. and Polashock, J. (2005) Alteration of anthocyanin glycosylation in cranberry through interspecific hybridization. *J. Amer. Soc. Hort. Sc.* 130, 711–715.
- Wells, H. F., and Buzby, J. C. (2008) Dietary assessment of major trends in U. S. food consumption, 1970–2005. Economic Information Bulletin No. 33 Economic Research Service, U. S. Dept. of Agriculture.
- Weisel, T., Baum, M., Eisenbrand, G., Dietrich, H., Will, F., Stockis, J. P., Kulling, S., Rufer, C., Johannes, C., and Janzowski, C. (2006) An anthocyanin/polyphenolic-rich fruit juice reduces oxidative DNA damage and increases glutathione level in healthy probands. *Biotechnol. J.* 1, 388–397.
- Werner, D. J., Creller, M. A., and Chaparro, J. X. (1997) Inheritance of blood flesh in peach. *HortScience* 33, 1243–1246.
- Wojdylo, A., Osmianski, J., and Laskowski, P. (2008) Polyphenolic compounds and antioxidant activity of new and old apple varieties. *J. Agric. Food Chem.* 56, 6520–6530.
- Wolfe, K., Wu, X., and Lu, R. H. (2003) Antioxidant activity of apple peels. *J. Agric. Food Chem.* 51, 609–614.
- Woo, J. H., Lim, J. H., Kim, Y. H., Suh, S. I., Min, D. S., Chang, J. S., Lee, Y. H., Park, J. W., and Kwon, T. K. (2004) Resveratrol inhibits phorbol myristate acetate-induced matrix metalloproteinase-9 expression by inhibiting JNK and PKC delta signal transduction. *Oncogene* 23, 1845–1853.
- World Health Organization. (2005) Fruits and Vegetables for Health: Report of a Joint FAO/WHO Workshop.
- Wu, X., Beecher, G. R., Holden, J. M., Haytowitz, D. B., Gebhardt, S. E., and Prior, R. L. (2006) Concentrations of anthocyanins in common foods in the United States and estimation of normal consumption. *Journal of Agricultural and Food Chemistry* 54, 4069–4075.
- Wung, B. S., Hsu, M. C., Wu, C. C., and Hsieh, C. W. (2005) Resveratrol suppresses IL-6-induced ICAM-1 gene expression in endothelial cells: effects on the inhibition of STAT3 phosphorylation. *Life Sci* 78, 389–397.
- Yang, Y., and Gallaher, D.D. (2005) Effect of dried plums on colon cancer risk factors in rats. *Nutr. Cancer* 53(1), 117–25.
- Yi, W., Fischer, J., Krewer, G., and Akoh, C. (2005) Phenolic compounds from blueberries can inhibit colon cancer cell proliferation and induce apoptosis. *J. Agric. Food Chem.* 53, 7320–7329.
- Yoshizawa, Y., Sakurai, K., Kawai, S., Asari, M., Soejima, J., and Murofushi, N. (2005) Comparison of antiproliferative and antioxidant properties among nineteen apple cultivars. *HortScience* 40, 5, 1204–1207.

- Youdim, K. A., McDonald, J., Kalt, W., and Joseph, J. A. (2002) Potential role of dietary flavonoids in reducing microvascular endothelium vulnerability to oxidative and inflammatory insults. *J Nutr Biochem* 13, 282–288.
- Zhao, J., Wang, J., Chen, Y., and Agarwal, R. (1999) Anti-tumor-promoting activity of a polyphenolic fraction isolated from grape seeds in the mouse skin two-stage initiation-promotion protocol and identification of procyanidin B5-3'-gallate as the most effective antioxidant constituent. *Carcinogenesis* 20, 1737–1745.
- Zheng, W. and Wang, S. (2003) Oxygen radical absorbing capacity of phenolics in blueberries, cranberries, chokeberries, and lingonberries. *J Agric Food Chem* 51, 502–509.

Fruit Breeding

Badenes, M.L.; Byrne, D.H. (Eds.)

2012, XV, 875 p. 83 illus., 41 illus. in color., Hardcover

ISBN: 978-1-4419-0762-2