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Vegetables containing phytochemicals with potential anti-obesity properties: a review

David J. Williams
Agri-Science Queensland

David Edwards
Agri-Science Queensland

Ingrid Hamernig
University of Queensland

Le Jian
Department of Health, Western Australia

Anthony P. James
Curtin University

See next page for additional authors

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Abstract

The incidence of obesity is rising worldwide at an alarming rate and is becoming a major public health concern with incalculable social and economic costs. Studies have exposed the relationship between the adiposity, inflammation and the development of other metabolic disorders, so dietary factors that influence some or all of these are of interest. Dietary phytochemicals appear to be able to target different stages of the adipocyte (fat cell) lifecycle. For example, several classes of polyphenols have been implicated in suppressing the growth of adipose tissue through modifying the adipocyte lifecycle. Many dietary phytochemicals also have strong anti-inflammatory activity, but the amount present in plants varies and may be affected by processing. In this review we summarise the likely mechanisms of action of plant phytochemicals. We highlight the major vegetable sources of polyphenols, including those with possible synergistic attributes, discuss the variation in polyphenol levels and their distribution in cultivars and outline the effects of food processing. The identification and characterisation of the anti-obesogenic properties of phytochemicals in vegetables, as well as an appreciation of the effect of cooking on phytochemical content provide significant new information supporting dietary guidelines that encourage vegetable consumption for the prevention and management of lifestyle related disease.

Keywords

obesity, properties, vegetables, containing, phytochemicals, potential, anti, review

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Authors

David J. Williams, David Edwards, Ingrid Hamernig, Le Jian, Anthony P. James, Stuart Keith Dr Stuart Keith Johnson, and Linda Tapsell

Review

Vegetables containing phytochemicals with potential anti-obesity properties: a review

David J. Williams[a]*, David Edwards[a], Ingrid Hamernig[b], Le Jian[c], Anthony P. James[d], Stuart K. Johnson[d], Linda C Tapsell[e]

[a] *Agri-Science Queensland, Department of Agriculture, Fisheries and Forestry (DAFF), PO Box 156, Archerfield BC, Queensland, 4108, Australia*

[b] *Institute of Molecular Biosciences, The University of Queensland, St. Lucia, Queensland, 4072, Australia*

[c] *Department of Health, 189 Royal Street, East Perth, Western Australia, 6004, Australia*

[d] *School of Public Health, Curtin Health Innovation Research Institute, Curtin University, GPO Box U1987, Perth, Western Australia, 6845, Australia*

[e] *Smart Foods Centre, School of Health Sciences, University of Wollongong NSW 2522 , Australia*

* **Corresponding author**. Tel.: +61 7 32766041 fax: +61 7 32166591 *E-mail address*: david.williams@daff.qld.gov.au

ABSTRACT

The incidence of obesity is rising worldwide at an alarming rate and is becoming a major public health concern with incalculable social and economic costs. Studies have exposed the relationship between the adiposity, inflammation and the development of other metabolic disorders, so dietary factors that influence some or all of these are of interest. Dietary phytochemicals appear to be able to target different stages of the adipocyte (fat cell) lifecycle. For example, several classes of polyphenols have been implicated in suppressing the growth of adipose tissue through modifying the adipocyte lifecycle. Many dietary phytochemicals also have strong anti-inflammatory activity, but the amount present in plants varies and may be affected by processing. In this review we summarise the likely mechanisms of action of plant phytochemicals. We highlight the major vegetable sources of polyphenols, including those with possible synergistic attributes, discuss the variation in polyphenol levels and their distribution in cultivars and outline the effects of food processing. The identification and characterisation of the anti-obesogenic properties of phytochemicals in vegetables, as well as an appreciation of the effect of cooking on phytochemical content provides significant new information supporting dietary guidelines that encourage vegetable consumption for the prevention and management of lifestyle related disease.

Keywords:

Adipocyte lifecycle

Anti-obesity phytochemicals

Polyphenols

Carotenoids

Organosulphurs

Cooking

1. Introduction

In 1998 obesity was defined as a “phenotypic manifestation of abnormal or excessive fat accumulation that alters health and increases mortality” (World Health Organisation, 1998). The WHO report stated that obesity had reached epidemic proportions worldwide. Since then its incidence has continued to rise at an alarming rate in both developed and developing countries and is becoming a major public health concern with incalculable social costs (Popkin, Kim, Rusev, Du, & Zizza, 2006; Popkin, 2009). There is a strong association between obesity and chronic diseases such as diabetes, cardiovascular diseases, hypertension, osteoarthritis, some cancers and inflammation-based pathologies which suggests that the obese are likely to have a disproportionate use of the health care system (Marinou, Tousoulis, Antonopoulos, & Stefanadi, 2010; Piper, 2011; Singla, Bardoloi, & Parkash, 2010). In order to maintain quality of life for the population and decrease the economic burden on the health system, more powerful dietary strategies to help reduce this cluster of diseases are urgently required.

While the strategy of reducing dietary fat content combined with increased physical activity has been shown to be effective in preventing obesity (Astrup, 2001; World Health Organisation, 2007), numerous studies have shown that this simple message is being ignored and alternative strategies are being sought (Kruger, Galuska, Serdula, & Jones, 2004; Stern et al., 1995; Wadden, 1993). Obesity is characterised at the cellular level by an increase in the number and size of adipocytes (fat storage cells) that have differentiated from pre-adipocytes in the adipose tissue (Furuyashiki et al., 2004). This transition from undifferentiated pre-adipocytes into mature adipocytes constitutes the adipocyte life cycle, and hence treatments that regulate both the size and number of adipocytes may provide a valuable adjunct to reduced dietary energy in combating obesity. The relationship between adiposity and inflammation is also being gradually unravelled with the recognition that adipocytes also produce inflammatory cytokines, suggesting that obesity induces an inflammatory state which may lead to further disease progression (Grundy, 2012).

With this in mind considerable interest has been aroused worldwide in the potential of dietary phytochemicals to help counteract obesity (Park & Kim, 2011; Rayalam, Della-Fera, & Baile, 2008; Santos, Rogero, & Bastos, 2010). Cell culture and animal model studies have indicated the anti-obesity effects occur through modification of the adipocyte life cycle. Polyphenols are a class of phytochemicals that are likely candidates as anti-obesity agents as several studies have suggested they can modulate the adipocyte life cycle (Rayalam et al., 2008; Yun, 2010). The strongest evidence is for this effect comes from: phenolic acid derivatives such as chlorogenic acid (Camire, Kubow, & Donnelly, 2009, Pan Lai, & Ho, 2010); the flavonols e.g. quercetin (Yun, 2010); and flavones such as luteolin (Rayalam et al., 2008). These classes of polyphenols (Figure 1) are widely distributed in plants and therefore are consumed regularly as part of the human diet.

Anti-obesity mechanisms of phytochemicals appear to involve mediation of complex and interconnected cell signalling pathways, therefore the combination of multiple phytochemicals may give rise to synergistic and enhanced anti-obesity effects. Synergistic interactions with combinations of phytochemicals have previously been investigated for the treatment of some cancers (Chan, Fong, Soprano, Holmes, & Heverling, 2003; Hermalswarya & Doble, 2006; Suganuma et al., 1999). However, such synergistic interactions among dietary bioactives acting on adipocytes have received only limited attention (Adams & Cory, 1998; Yang, Della-Fera, Hausman, & Baile, 2007). So far these studies have been encouraging with results indicating an enhanced induction of apoptosis and suppression of adipogenesis by phytochemcals used in combination. Phytochemical combinations that included polyphenols such as stilbene, resveratrol, genistein and naringenin have proven the most effective (Baile et al., 2011; Nelson-Dooley, Della-Fera, Hamrick, & Baile, 2005). Results from such studies suggest that anti-obesity effects could be achieved by consuming lower levels of phytochemicals but in specific combinations.

Vegetables provide a major dietary source for phytochemicals with potential anti-obesity properties, with the types and levels varying markedly between species and even cultivar (Nuutila, Puupponen-Pimia, Aarni, & Oksman-Caldentey, 2003; Singh, Upadhyay, Prasad, Bahadur, & Rai, 2007). In addition climatic, agronomic and harvest conditions also significantly influence the levels of these phytochemicals in vegetables (Naczk & Shahidi, 2006; Tiwari & Cummins, 2011).

Post-harvest operations, including food processing have a major influence on the levels of phytochemicals in vegetables and vegetable products. Conventional (thermal), non-thermal (e.g. high pressure, ultrasound, irradiation), domestic (e.g. washing, peeling, cutting) and industrial (canning, drying) processing are widely reported to degrade phytochemicals (Rawson, Koidis, Rai, Tuohy, & Brunton, 2010; Volden, Bengtsson, & Wicklund, 2009). Heat treatment is the most common method for processing vegetables because of its inactivation of pathogenic and spoilage microorganisms and endogenous enzymes leading to improved quality and shelf-life (Rawson et al., 2011). To retain phytochemicals during the various cooking treatments on offer, the food processor must optimise all steps in order to restrict their degradation.

With this backdrop, the aims of this review are to summarise the proposed mechanisms of action of phytochemicals on obesity related pathways, highlight the vegetable sources of phytochemicals; and discuss the influence of different cultivars and distribution within the vegetable source as well as the impact of cooking on the levels of these phytochemicals.

2. Mechanisms of action of phytochemicals on adiposity

The plausible mechanisms of action of certain vegetable phytochemicals include: (a) reducing adipose tissue mass by inhibiting the proliferation of precursor cells; (b) increasing the rate of apoptosis during the adipocyte lifecycle (Rayalam et al., 2008; Yun, 2010) and (c) the inhibition of dietary triglyceride absorption via reduction in pancreatic lipase formation (Birari & Bhutani, 2007). Obesity has been associated with a chronic inflammatory status (Pan et al. 2010) and the strong anti-inflammatory activity may be one of the mechanisms of action for counteracting the negative physiological effect of the obesogenic state.

The development and maintenance of obesity involves many complex molecular mechanisms and interconnected cell signalling pathways and to discuss recent advances in this topic is well beyond the scope of this review. Therefore the current review is restricted to a summary of proposed mechanisms of action of the major vegetable phytochemicals.

2.1 Polyphenols

Polyphenols are a class of phytochemicals widespread in vegetables that have demonstrated one or more potential anti-obesity effects. Dietary polyphenols may suppress growth of adipose tissue by modulating adipocyte metabolism (Badimon, Vilahur, & Padro, 2010; Mulvihill & Huff, 2010). Unfortunately to date, the effects of polyphenols on human adipocytes have not been studied systematically; most studies having been conducted on murine cell lines such as 3T3-L1 and in the tissues of laboratory animals (Hsu & Yen, 2006; Morikawa, Ikeda, Nonaka, & Suzuki, 2007).

Polyphenols, including their functional derivatives, esters and glycosides, have one or more phenol groups with one hydroxyl – substituted aromatic ring (Dey & Harborne, 1989). According to their structure and the type and number of structural elements binding to the rings, polyphenols are grouped into different classes (Figure 1). The classes of polyphenols for which there is most evidence of potential anti-obesity properties are: (1) the simple phenolic acids such as chlorogenic, coumaric, gallic and caffeic acids and (2) the flavonoid sub-classes, flavonols e.g. quercetin, kaempferol, myricetin and isorhamnetin and the flavones e.g. luteolin and apigenin.

2.1.1 Phenolic acids: chlorogenic acid and related compounds

Naturally occurring phenolic acids contain two distinguishing constitutive carbon frameworks: hydroxycinnamic and hydroxybenzoic structures. Although the basic skeleton remains the same, the numbers and positions of the hydroxyl groups on the aromatic ring create the variety (Figure 1). Common hydroxycinnamic acid derivatives are coumaric, caffeic and ferulic acids which frequently occur in foods as simple esters with quinic acid or glucose. The most widely occurring of these is chlorogenic acid. Unlike hydroxycinnamates, hydroxybenzoic acid derivatives are mainly present as glycosides. The most common forms in plant foods are *p*-hydroxybenzoic and vanillic acids (Herrmann, 1989).

Hsu and Yen (2006) investigated the inhibitory effect of dietary phenolic acids on mouse pre-adipocytes. Chlorogenic and coumaric acids caused significant inhibition of cell growth as well as enhancing apoptosis. Gallic acid while not affecting the adipocyte cell cycle did increase the number of apoptotic cells. A recent study (Son, Rico, Nam, & Kang, 2010) evaluated the effects of feeding ferulic acid on lipid metabolism of mice. This dietary phenolic acid suppressed the weight gain due to the high fat diet and inhibited fatty acid biosynthesis.

2.1.2 Flavonoids: flavonols, flavones and anthocyanins

Flavonoids are a class of polyphenols that are widely distributed in vegetables and can be further differentiated into sub-classes according to their structure. They share the common skeleton of diphenylpropanes (C_6-C_3-C_6). The main difference between the two important sub-classes, i.e. the flavonols and the flavones is the presence of a hydroxyl group at C_3 in flavonols (Figure 1). They both usually occur in plants as glycosides.

2.1.2.1 Flavonols: quercetin and related compounds

Quercetin is a dietary flavonol found in vegetables, for which there is most evidence for its potential anti-obesity effects. It has been shown to inhibit adipogenesis (Strobel et al., 2005) and to induce apoptosis in mouse pre-adipocytes (Fang et al, 2008; Hsu & Yen, 2006; Kuppusamy & Das, 1992). Kaempferol has also exhibited these potential anti-obesity properties but to a lesser extent (Fang, Gao, & Zhu, 2008). A recent study (Ahn, Lee, Kim, Park, & Ha, 2008) provided useful insights into the molecular mechanisms by which quercetin influences the regulation of fat cell differentiation and apoptosis.

Park, Yang, and Amabati (2008) exposed human adipocytes to quercetin in combination with the isoflavone, genistein and the stilbene, reservatrol to human adipocytes. The combined treatments caused enhanced inhibition of lipid accumulation in maturing human adipocytes, far greater than the responses to individual compounds.

Several studies have revealed that quercetin provides some protective effects against obesity-related inflammation (Al-Fayez, Cai, Tunstall, Steward, & Gesher, 2006; Chuang, Martinez, & Xie, 2010). Quercetin was demonstrated to attenuate markers of inflammation, macrophages and insulin resistance in human adipocytes and reduce circulating markers of inflammation in animal models.

2.1.2.2 Flavones: luteolin and apigenin

Park, Kim, and Kim (2009) demonstrated an anti-adipogenic effect of the flavone, luteolin on murine 3T3-L1 pre-adipocytes mediated through decreased lipid accumulation and inhibition of differentiation. An earlier study (Kuppusamy & Das, 1992) had shown that the addition of a similar flavone, apigenin induced lipolysis in rat adipocytes.

2.1.2.3 Anthocyanins

Another flavonoid sub-class with potential for anti-obesity-related effects is the anthocyanins, responsible for the red, blue and purple colours in vegetables (Clifford, 2000). *In situ*, anthocyanins are stabilised by the formation of complexes with other flavonoids and their degradation is prevented by glycosylation and esterification with various organic acids and phenolic acids. In a 2008 study, Tsuda showed that anthocyanins possess significant anti-inflammatory properties in obese adipose tissues.

Another possible anti-obesity mechanism associated with anthocyanins was reported by Sasaki et al. (2007). They observed that cyanidin, the most common anthocyanin in foods, reduced blood glucose levels as well as down regulating inflammatory protein cytokines such as monocyte chemoattractant protein-1 (MCP-1) in the adipose tissue of mice. Recent studies have demonstrated that an increase in expression of these inflammatory molecules in adipose tissue contributes to the development of insulin resistance (Kamei et al., 2006; Sartipy & Loskutoff, 2003).

2.2 Carotenoids

Carotenoids, though not polyphenols, but rather a sub-class of terpenoids, have been reported to possess anti-obesity and anti-inflammatory abilities (Gonzalez-Castejon & Rodriguez-Casado, 2011). Carotenoids are classified into hydrocarbons (carotenes) and their oxygenated derivatives (xanthophylls). They are responsible for the yellow, orange and red colour of many vegetables. α-Carotene is one of the most abundant carotenoids in the diet and can be converted in the body to an active form of vitamin A. ß-Carotene inhibits inflammatory gene expression in lipolysaccharide-stimulated macrophages. Possible anti-obesity roles for both these carotenes have been postulated based on the finding that the plasma of overweight and obese children had significantly lower levels of α-carotene and ß-carotene when compared to healthy weight children (Burrows, Warren, Colyvas, Garg, & Collins, 2009).

2.3 Organosulphurs

Several investigations have noted that organosulphur compounds isolated from Allium vegetables have induced apoptosis of human tumour cells (Nishikawa, Yamada, Hattori, Fukada, & Fujino, 2002; Siegers, Steffen, Robke, & Pentz, 1999). One of the principal constituents believed largely responsible for these effects is allicin. Allicin is not present as such in the intact vegetable but is produced in the presence of the enzyme, allinase during cutting or crushing. Allicin is chemically unstable and rapidly breaks down to give the unsaturated disulphide, ajoene.

To evaluate the efficacy of these compounds to induce apoptosis of adipocytes, Elkayam et al. (2003) fed pure allicin to rats with fructose-induced hyperlipidemia, hyperinsulinemia and hypertension. They observed that of the three feeding regimes only those given pure allicin exhibited no weight gain which indicated to the authors that allicin may have benefits in controlling weight in humans. In a similar study, Yang, Della-Fera, Nelson-Dooley, and Baile (2006) noted that the application of pure ajoene induced apoptosis in 3T3-L1 adipoytes. These findings lead them to conclude that ajoene can regulate fat cell numbers through the induction of apoptosis and therefore may be useful as a new therapeutic agent to combat obesity.

Another class of organosulphurs receiving attention as anti-obesity agents are the glucosinolate hydrolysis products (Gonzalez-Castejon & Rodriguez-Casado, 2011). Glucosinolates are sulphur containing natural plant products found in Brassica vegetables. They occur in the plant in conjunction with the hydrolytic enzyme, myrosinase and in intact tissues, the enzyme is stored separately from the glucosinolates. When tissue damage occurs (food preparation, chewing or pest attack) glucosinolates are hydrolysed by the myrosinases to a range of breakdown products such as isothiocyanates, nitriles, and indoles (Williams, Critchley, Pun, Nottingham, & O’Hare, 2008). It is these breakdown products that are biologically active most notably the isothiocyanates and indoles and not the precursor glucosinolates. The type of product that is formed depends on pH, structure of glucosinolate side chain and the presence or absence of supplementary specifier proteins such as epithiospecifier proteins (ESPs) (Williams et al., 2008). The isothiocyanate, sulphoraphane (produced by the action of myrosinase on the glucosinolate, glucoraphanin) has received much attention for its perceived anti-carcinogenic activity and possible anti-obesogenic effects. To date, investigations that aim to evaluate possible anti-obesogenic properties of these compounds have focussed on their anti-inflammatory attributes, particularly the isothiocyanate, sulphoraphane and indole-3-carbinol. Sulphoraphane has been found to decrease the production of inflammatory signalling molecules in cultured macrophages (Heiss, Herhaus, Klimo, Bartsch, & Gerhauser, 2001). Data is also accumulating that suggests sulphoraphane suppresses the activation of lipopoly-saccharide-induced transcription factors involved in inflammation and cancer (Woo & Kwon, 2007). Similar to sulphoraphane, a role for indole-3-carbinol in controlling inflammation is emerging as outlined by Cho et al. (2008). Glucosinolate breakdown products have well-known anti-carcinogenesis properties which includes blocking the cell cycle and promoting apoptosis (reviewed in Cartea & Velasco, 2008). Whether these attributes apply to the adipocyte lifecycle could prove to be an exciting area of future research.

With the exception of this observational study, most of the evidence supporting the effects of dietary phytochemicals on obesity comes from mechanistic studies using cell lines or animal models. This type of research provides help to generate hypotheses for studies in humans and adds to the plausibility of guidance to move towards more plant based diets for the prevention of chronic lifestyle related disease. Indeed, the targeting of several points in the adipocyte lifecycle by dietary phytochemicals has been proposed as a potentially effective obesity treatment approach (Badimon et al., 2010). Direct evidence of effects from clinical trials is required however, to confirm the anti-obesogenic effects of diets high in phytochemicals.

3. Major vegetable sources of anti-obesogenic phytochemicals

To move forward with this research, accurate and reproducible methods for isolating and determining the amounts of these compounds are required. The diverse chemical natures of phytochemical species complicate the extraction and hydrolysis steps required for their determination. Although numerous extraction methods for phytochemcials have been described in the literature, a common feature is that their validation is performed using only one plant material type and for only one specific class of phytochemicals. Likewise, Nuutila, Kammiovirta, and Oksman-Caldentey (2002) suggested that the hydrolysis conditions needed to be optimised separately for each plant species under investigation. Some of the discrepancies in the literature between levels of individual polyphenols from the same vegetable source could in part be attributed to the differing extraction and hydrolysis protocols used (Tables 1-3).

3.1 Vegetable sources of phenolic acids: chlorogenic acid and related compounds

Potato and sweetpotato are rich in chlorogenic acids that constitute up to 90% of the total phenolic content of these vegetables (Table 1). Tuber size and variety was reported to have little effect on chlorogenic acid content of potatoes, with smaller varieties only showing insignificantly lower values than larger ones

(Friedman, 1997; Table 1). Different sweetpotato cultivars grown under the same conditions have been reported to contain as many as five chlorogenic acid isomers (Takenaka, Nanayama, Isobe, & Murata, 2006). Chlorogenic acids in orange-fleshed sweetpotato were highest in leaves, followed by the peel then whole root (including peel) and then flesh (Table 1; Truong, McFeeters, Thompson, Dean, & Shofran, 2007). In sixteen sweetpotato cultivars (Harrison et al., 2008), wide variations in chlorogenic acid content were found between the periderm (skin layer), cortex (layer under periderm) and stele (the remaining edible portion) (Table 1). Padda and Picha (2007) also found high chlorogenic acid in young immature leaves and in small sweetpotato roots.

Other good dietary sources of chlorogenic acids are lettuce and carrot (Mattila & Hellstrom, 2007). Carrot also has been reported to have high levels of the phenolic acid, *p*-hydroxybenzoic acid (Mattila & Kumpulainen, 2002). Ferracane et al. (2008) isolated significant quantities of chlorogenic acid isomers from globe artichoke with 5-*O*-caffeoylquinic and 1,5-dicaffeoylquinic acids being the predominant forms.

3.2 Vegetable sources of flavonols: quercetin and related compounds

Quercetin is the major dietary flavonoid of the flavonol class found in vegetables. Other phytochemicals found in vegetables that are structurally close to quercetin include kaempferol, myricetin and isorhamnetin (Figure 1). These phytochemicals are present in vegetables in both free or in bound forms linked to carbohydrate moieties (Olsson, Gustavsson, & Vagen, 2010).

Onions, an important crop of the Allium family, contain high amounts of quercetin, with levels varying between cultivar but also within the different layers of the onion bulb. Studies on red onions showed the dry skin fraction contained 3 times the level of quercetin than the outer fleshy layer and the inner edible portion (Gennaro et al., 2002; Table 2). The outer layers of onions were shown to contain up to 10 times the levels of quercetin and kaempferol than the inner layers (onion variety not specified), with low levels of myricetin present but only in the inner leaves (Chu, Chang, & Hsu, 2000). It should be noted that these authors presented the flavonol values on an as-is basis (FW), therefore the drier skin and outer layers give rise to enhanced values when compared to the moist inner layers.

Lettuce (Crozier, Jensen, Lean, & MacDonald, 1997a), broccoli (El-Gharras, 2009) and curly kale (Olsen, Aaby, & Borge, 2010) also have high quercetin contents. Kale was also a significant source of kaempferol (Hollman & Arts, 2000, Olsen et al., 2010). Capsicum also contained appreciable levels of quercetin (Table 2). Sweetpotato leaves are rich in quercetin and myricetin (Chu et al., 2000) (see Table 2). Rutin, a common glycoside of quercetin has been found in large amounts (0.03-0.06% FW) in asparagus shoots (Wang et al., 2003).

3.3 Vegetable sources of flavones: luteolin and apigenin

Flavones are much less common than flavonols in vegetables (Gonzalez-Castejohn & Rodriguez-Casado, 2011). Natural flavones consist of glycosides of luteolin and apigenin, which are corresponding flavones to the flavonols, quercetin and kaempferol, respectively (Figure 1). The only significant vegetable sources of flavones identified to date are parsley and celery (Crozier, Lean, MacDonald, & Black, 1997b; Harnly et al., 2006; Meyer, Bolarinwa, Wolfram, & Linseisen, 2006; Table 3). Highly variable levels of luteolin and apigenin were found in different varieties of celery (Crozier et al., 1997b). The authors suggested that this variation may have been related to different light regimes during growth or different storage/transport conditions. Celery leaf is a significant source of luteolin and apigenin, while the stalk and heart contain much lower levels (Hollman & Arts, 2000; Crozier et al., 1997b; Table 3).

Relatively high luteolin and apigenin levels have also been reported in globe artichoke (Azzini et al., 2007) (Table 3). Sun et al. (2007) detected significant amounts of luteolin in capsicum confirming the earlier measurements of Hollman and Arts (2000).

3.4 Vegetable sources of organosulphurs

Many plants belonging to the Allium group such as garlic, onion, shallot and leek are rich sources of organosulphurs, including allicin, its precursor, allin and other thiosulphinates (Sahu, 2002). Allicin, the main bioactive ingredient is highly unstable and easily breaks down to ajoene, vinyldithins, diallyl trisulphide or other sulphides in air or water (Miron et al., 2002). It is these molecules that are responsible for the characteristic aroma and flavour of Allium vegetables (Sahu, 2002). However this instability makes comparison of allicin content between vegetables difficult with one group of researchers even suggesting that this compound cannot be detected in most biological samples (Itakura et al., 2001).

Using gas chromatography (GC) these authors measured the allicin content (determined as vinyldithin after allicin decomposed in the injection port) of a range of vegetables. Allicin was the predominant thiosulpinate constituent in garlic but was absent in the samples of onion and leeks. The erroneous picture of thiosulphinate composition given by GC had been recognised earlier by Block, Naganathan, Putman, and Zhao in 1992. After stating that high performance liquid chromatography (HPLC) offered a more reliable measure of thiosulphinates in plants they identified and quantified these molecules in a broad range of vegetables. They found that garlic possessed by far the highest levels followed by the onion varieties. The major thiosulphinate found in the garlic samples was allicin present to the extent of 0.3% FW. These findings were in direct contrast to those given by Cheng (2006). Also using HPLC this study found that onion possessed the highest allicin content with 0.168 mg/g DW followed by garlic (0.051 mg/g DW) with chives and leeks having no detectable amounts.

From a dietary point of view glucosinates are restricted to plants of the Brassica species. Several of these species are widely consumed by humans as cooked or salad vegetables such as cabbage, Brussels sprouts, cauliflower, broccoli, turnip, radish and watercress or condiments including horseradish and white mustard. Actually, more than 130 glucosinolates have been identified; however most species contain a limited number (generally less than twelve) (Halkier & Gershenzon, 2006). Previous studies have found that the composition and content of glucosinolates vary widely, not just between the various Brassica species but also the different developmental stages and tissues within a given species (Porter, Morton, Kiddle, Doughty, & Wallsgrove, 1991; Koroleva et al., 2000). Broccoli (both the sprouts and mature vegetables) are rich-sources of glucoraphanin (Fahey, Zhang, & Talalay, 1997). However it is worth noting that several studies have reported that broccoli possesses epithiospecifier proteins (ESPs) particularly in the early stages of plant development (Matusheski, Juvik, & Jeffery, 2004; Williams et al., 2008). These ESPs redirect the glucosinolate breakdown to the non-bioactive nitriles at the expense of isothiocyanate formation (reviewed by Kissen, Rossiter, & Bones, 2009).

3.5 Vegetable sources of multiple phytochemicals

Complex mechanisms are involved in regulating adipose tissue development by dietary phytochemicals, it follows that exposure of adipocytes to multiple vegetable phytochemicals found in a high vegetable diet could result in enhanced or even synergistic effects. There are several vegetables that contain appreciable levels of a range of these compounds. The presence of two natural plant pigments with anti-obesity mechanisms is also noted in these same vegetables, and recent studies (reviewed by Tiwari & Cummins, 2011) have indicated that dark coloured cultivars of vegetables possess higher levels of phytochemicals when compared to light coloured varieties.

3.5.1 Orange-fleshed sweetpotato

The commonly consumed edible portions of sweetpotato contain higher levels of chlorogenic acids than most potato varieties (Table 1). Chlorogenic acids were approximately 2-fold higher in orange-fleshed varieties compared to those with cream-coloured flesh (Rautenbach, Faber, Laurie, & Laurie, 2010). Padda and Picha (2008) further noted that total chlorogenic acid content of the purple-fleshed variety contained the highest levels, while one white-fleshed variety had nearly twice the content of the other white- and orange-fleshed varieties (Table 1).

Some sweetpotato varieties have also been reported to be rich in ß-carotene; particularly those with dark yellow or orange flesh (Rautenbach et al., 2010). These authors observed that the two orange-fleshed varieties tested had a high ß-carotene content of >14 mg/100g (fresh weight) while no ß-carotene was detected in the cream-fleshed varieties. An earlier study (Teow et al., 2007) had reported a ß-carotene range of 1.2-22.6 mg/100g (fresh weight) for orange-fleshed varieties and 0.5-5.7 mg/100g for purple-fleshed sweetpotatoes.

3.5.2 Red-leafed lettuce

Another good source of chlorogenic acids are the red-leafed varieties of lettuce. Chlorogenic acid levels in this plant were distributed across the coloured tissues, with red tissue containing approximately three times the level of green tissue and approximately eight times that of the white midrib tissue (Ferreres, Gil, Castaner, & Tomas-Barberan, 1997; Table 1).

Red-leafed lettuce also possesses substantial levels of quercetin (significantly higher than green-leafed varieties) (Table 2). Quercetin levels varied across the types of tissue similarly to the distribution of phenolic acids (Ferreres et al. 1997;

Table 2). Crozier et al. (1997b) also reported the outer leaves of red-leafed lettuce contained 2 – 10 times the quercetin content of inner leaves (Table 2).

Ferreres et al. (1997) identified and quantified substantial amounts of the anthocyanin, cyanidin-3-malonylglucoside from red lettuce leaf tissues. This compound had been previously isolated and identified from red onions (Ferreres, Gil, & Tomas-Barberan, 1996).

3.5.3 Red onions

In a study that focussed on the growth inhibitory effect of flavonoids on 3T3-L1 pre-adipocytes, the authors reported that quercetin had by far the highest inhibitory effect of the six flavonoids tested (Hsu & Yen, 2006). The richest sources of dietary quercetin are red and yellow onions, while white onions contain lower levels (Crozier et al., 1997b; Harnly et al., 2006; Slimestad, Fossen, & Vagen, 2007; Table 2). Furthermore Shon, Choi, Kahng, Nam, and Sung (2004) noted that both the total phenolic content and flavonoid content were high among red onions when compared to other coloured cultivars.

A recent study (Gorinstein et al., 2008) also indicated that red onions contain significant amounts of anthocyanins (46.02 mg of cyanidin-3-glucoside/100g dry weight) especially when compared to white onions (2.83 mg of cyanidin-3-glucoside/100g dry weight).

Several authors have indicated that some onion varieties contain appreciable quantities of thiosulphinates (Block et al., 1992; Cheng, 2006). Pertinent to this review Block et al. (1992) observed that yellow onion samples had higher levels (0.35 mole%) than red onions (0.20 mole%).

3.5.4 Red capsicum

Antioxidant compounds and their antioxidant activity in four different coloured (green, yellow, orange and red) capsicum was the focus of a 1997 study (Sun et al., 2007; Table 2). They reported that red capsicum had significantly higher total phenolics content than the other coloured varieties. The quercetin content was significantly higher than either green or yellow capsicums but similar to the orange variety. The same authors also identified significant amounts of luteolin in red capsicum confirming the earlier report by Hollman and Arts (2000). However these findings contrasted with those of Arabbi, Genovese, and Lajolo (2004) which suggested that green capsicum contained higher levels of luteolin than yellow and red capsicum (Table 3). Furthermore Sun et al. (2007) stated that red capsicum contained the highest levels of the pigmented carotenoids (ß-carotene and capsanthin). This high content of β-carotene in red capsicum was confirmed by Stahl and Sies (2005) but the authors noted that the levels were inferior to carrot but superior to most other fruits and vegetables. This study also stated that red capsicum to be a rich source of the carotenoid lycopene (Stahl & Sies, 2005). This carotenoid as well as being a powerful antioxidant has been suggested to have considerable anti-obesity action (Agarwal & Rao, 2000).

3.5.5 Red curly kale

Kale is reputed among vegetables to have one of the highest antioxidant capacities together with high concentrations of phenolic acids, flavonoids, carotenoids and glucosinolates (deAzevedo & Rodriguez, 2005; Podsedek, 2007). Due to the perceived health benefits associated with anthocyanins, red/purple coloured varieties are gaining popularity (Olsen et al., 2010). These authors characterised and quantified the polyphenols in the edible leaves of the red curly kale variety Redbor. They reported that this variety was a rich source of phenolic acids (mainly *p*-coumaric, ferulic, synapic and caffeic acids), flavonols (particularly quercetin and kaempferol) and the anthocyanin cyanidin. Previous studies on the flavonol content of the green varieties (Justesen, Knuthsen, & Leth 1998; Olsen, Aaby, & Borge, 2009) reported significantly lower levels of quercetin and kaempferol than those reported for the red/purple varieties by Olsen et al. (2010).

In a recent study on fresh and heat treated curly kale cultivars, the authors found that the raw green samples possessed double the amount of glucosinolates when compared to the red (Olsen, Grimmer, Aaly, Saha, & Borge, 2012). Interestingly, on heating the red cultivar the reduction in total glucosinolates was only 15% compared to 40% exhibited by the green samples. In fact the authors observed that the red cultivar was consistently more resistant to the losses of phytochemicals (polyphenols and even vitamin C) on heating than the green.

4. Effect of cooking methods on phytochemical levels

Data on phytochemical intake from vegetable consumption are often derived from analysis of raw material. However, many vegetables are cooked in a wide variety of ways before consumption. The magnitude and duration of the heating in these cooking processes has a very strong influence on the levels of phytochemicals remaining in the cooked product (Patras, Brunton, O'Donnell, & Tiwari, 2010). Surprisingly, given the recognised health benefits of polyphenols, literature data reporting the effect of cooking on vegetable polyphenols is scarce and often limited to only the total phenolics concentration (Ferracane et al., 2008). Studies that investigated the effect of cooking processes on the levels of potential anti-obesity phytochemicals will be reviewed.

As a general rule, the level of phytochemicals in vegetables decreases exponentially with increases in cooking duration and magnitude (Tiwari & Cummins, 2011). However there have been reported cases where heating aids the extractability of these phytochemicals leading to an apparent concentration increase (Howard, Wong, Perry, & Klein, 1999). Therefore, the content of available phytochemicals reported in vegetables after cooking is a net result of the combined effects of degradation and leaching during cooking and changes in phytochemical extractability during analysis.

4.1 Effect of cooking on phenolic acids: chlorogenic acid and related compounds

The chlorogenic acid content of fresh-cut potato strips was reported to decrease by 50%, 66%, 63% and 71% on steaming, boiling microwaving and frying, respectively (Tudela, Cantos, Espan, Thomas-Barberan, & Gil, 2002). Support for this observation was provided by Miglio. Chiavaro, Visconti, Fogliano, and Peregrini (2008), who reported a general decrease in phenolic acids for all the vegetables that underwent cooking. Other studies involving potato found that chlorogenic acids were absent in baked potatoes (Friedman, 1997), while frying resulted in losses of over 50% in potato and carrot (Friedman, 1997; Miglio et al., 2008). A possible exception was provided by Takenaka et al. (2006) when they reported a slight increase in the chlorogenic acid isomers (3-caffeoylquinic, 4-caffeoylquinic,3,4-decaffeoylquinic and 4,5-dicaffeoylquinic acids) on boiling sweetpotatoes. A later investigation (Truong et al., 2007) supported this finding when they stated that steam cooking resulted in slight increases in the concentration of individual phenolic acids identified in sweetpotato root tissues. However, prolonged boiling resulted in losses of chlorogenic acid of 60 – 100% (Friedman 1997; Takenaka et al., 2006; Miglio et al., 2008). It was suggested that the slight increase in chlorogenic acid could be attributed to release of bound phenolics and the inactivation of polyphenol oxidase during steaming (Truong et al., 2007). Polyphenol oxidase (PPO) is an enzyme present in many plants and on tissue damage, e.g. cutting, the enzyme catalyses the formation of a brown pigment from phenolic compounds. Artichoke is a vegetable that has a very high PPO activity when raw. On cooking (steaming, boiling and frying), this vegetable shows a significant increase in the concentration of the caffeoylquinic acid (Ferracane et al., 2008). It was suggested that the cooking treatments resulted in PPO inactivation that reduced the enzymatic degradation compared to the raw artichoke of the phenolic acids. In the same study, cooking reduced levels of flavones (e.g. apigenin) which the authors attributed to the increased thermal degradation of these compounds.

4.2 Effect of cooking on flavonols: quercetin and related compounds

Reductions of between 44-53% in the levels of the quercetin glycosides were reported during 60 min boiling of onions (Rodrigues, Perez-Gregorio, Garcia-Falcon, & Simal-Gandara, 2009). An earlier study had demonstrated that 15% of quercetin was lost on boiling onions for only 5 min (Lombard, Peffley, Geoffriau, Thompson, & Herring, 2005). Both groups of authors suggested that this reduction was due to thermal degradation of the quercetin but they could not discount leaching of the water soluble quercetin. This loss of quercetin confirmed the experiments of Crozier et al. (1997b) who reported that boiling and microwaving reduced levels by 75% and 64%, respectively. Frying onions in sunflower oil resulted in a reduction of only 21%, possibly due to the less effective extraction of the hydrophilic quercetin by the hot oil compared to hot water. These researchers also reported a similar reduction of quercetin in tomatoes after undergoing the same cooking treatments (Crozier et al., 1997b).

Quercetin and kaempferol levels in broccoli were less affected by steaming (losses of 40% and 1%, respectively) than by frying (70% and 45%, respectively) and boiling (90% and 85%, respectively) (Miglio et al., 2008). Greater diffusion of phytochemicals into an aqueous, boiling cooking medium as opposed to steaming and frying is to be expected for these water-soluble compounds and accounts for the different levels observed (Miglio et al., 2008).

Blanching edible sweetpotato leaves at 100°C for 30 sec reduced the levels of quercetin and myricetin by 25%, while a 60 sec treatment resulted in losses of 50%. Treatment for further 60 sec resulted in 80% loss of myricetin (Chu et al., 2000).

4.3 Effect of cooking on flavones: luteolin and apigenin

Blanching sweetpotato leaves at 100°C caused a 50% loss of apigenin after 30 sec and 65% loss after 120 sec (Chu et al., 2000). Steaming resulted in losses of 25% of apigenin in globe artichokes, while 35% was lost on boiling and 60% by frying (Ferracane et al., 2008). To date the authors could find no references that reported the effects of cooking on luteolin levels.

4.4 Effect of cooking on anthocyanins and carotenoids

There is little published information on the thermal stability of anthocyanins in vegetables. However, Oliveira, Amaro, Pinho, and Ferreira (2010) observed a 12% to 42% reduction of anthocyanins in cooked blueberries during progressive heating from 12º to 99ºC for 60 min. They suggested anthocyanins are naturally unstable and degradation is primarily caused by oxidation. This instability had been previously noted by Sadilova, Stintzing, and Carle (2006) in strawberry, elderberry and black carrot concentrates. These authors suggested the degradation mechanism is due to hydrolysis of the anthocyanin sugar moiety leading to formation of a phenolic non-bioactive aglycone.

Isomerisation and oxidation reactions upon heating of carotenoids have been reported (Rodriguez-Amaya, 1999). Even though carotenoids are susceptible to thermal isomerisation most of these compounds appear to be much more heat stable than anthocyanins (Leong & Oey, 2012; Nguyen, Francis, & Schwartz, 2001; Nguyen & Schwartz, 1998). Van Jaarsveld, Marais, Harmse, Nestle, and Rodriguez-Amaya (2006) studied the effect of thermal processing on ß-carotene in orange-fleshed sweetpotato and found minimal losses after boiling for 20 and 30 min. These small losses were later confirmed by Rautenbach et al. (2010) when they observed an average decrease of 9.7% on boiling sweetpotato for 12 min. These results seem contradictory to earlier studies of Hagenimana, Carey, Gichuki, Oyungand, and Imungi (1999) and K'osambo, Carey, Misra, Wilkes, and Hagenimana (1998) where decreases of 30.6% and 14-59% for total carotenoids on boiling for 30 min were reported. A study that evaluated the effects of heating i.e. 98ºC for 10 min on carotenoids in carrots and red capsicum reported no change in the content in the red capsicum but a significant decrease in the carrots. There are several studies that testify to the thermal stability of the carotenoid lycopene, mostly focussing on tomato and tomato products (Gupta, Balasubramaniam, Schwartz, & Francis, 2010; Kessy, Zhang, & Zhang, 2011; Nguyen & Schwartz, 1998).

4.5 Effect of cooking on organosulphurs

Considering the well recorded benefits of the thiosulphinates in Allium vegetables surprisingly few studies have evaluated the impact of cooking on these levels. In fact most of the cooking studies have focussed on monitoring changes in their anti-thrombotic activity, a property attributed to allicin and the thiosulphinates

(Ali, 1995; Cavagnaro, Camargo, Galmarini, & Simon, 2007; Chen, Chen, Tsa, & Jen, 2000). Boiling (15-30 min) was seen to completely inhibit anti-thrombotic activity in uncrushed garlic and Welsh onion (Ali, 1995; Chen et al., 2000) which the authors suggested was possibly due to the inactivation of the allinase before it could produce any anti-thrombotic agents. A later study confirmed that; 1) allicin and thiosulphinates were responsible for the anti-thrombotic activity and 2) the lack of anti-thrombotic activity found previously (Ali, 1995; Chen et al., 2000) in boiled samples was due to allinase inactivation thus preventing the formation of the bioactive thiosulphinates (Cavagnaro et al., 2007).

Glucosinolates and hydrolysis products can be lost following cooking which may reduce glucosinolate levels by 30-60%, depending on the method (e.g., conventional, microwave, high pressure), cooking intensity, and on the type of glucosinolate present (Ciska & Kozlowska, 2001; Vallejo, Thomas-Barberan, & Garcia, 2002). Glucosinolate breakdown products are barely detected after prolonged cooking, with the exception of several non-bioactive compounds (Macleod & Macleod, 1968). Handling, storage and cooking methods that provide minimal loss of bioactivity have yet to be determined. However, a paper by Song and Thornalley (2007) went some way to rectifying this when they outlined several cooking methods that preserved the glucosinolates as well as retaining some of the myrosinase thus allowing an increase in the conversion of glucosinolates to isothiocyanates. Their investigation involved seven major glucosinolates in broccoli, Brussels sprouts, cauliflower and cabbage and their stability under different cooking conditions. The findings of this study were confirmed recently by Aires, Carvalho, and Rosa (2012) when they reported that steaming of vegetables was the most successful in preserving glucosinolates as opposed to boiling which caused glucosinolate losses of up to 81%.

5. Conclusions

Scientific investigations using cell culture and animal model studies demonstrate that polyphenols derived from vegetables can induce lipolysis, decrease lipid accumulation and induce apoptosis in adipose tissue. These mechanisms indicate potential anti-obesity properties that lend themselves to testing in human clinical studies. In addition to possible effects on adipocytes themselves, the anti-inflammatory properties reported for some vegetable phytochemicals suggest a powerful adjunct to dietary energy restriction in obesity-related chronic disease management.

This review does not provide an exhaustive list of phytochemcials found in vegetables. As research identifies other compounds that modify the adipocyte life cycle or possess high anti-inflammatory activity, and knowledge of the human adipocyte lifecycle expands, additional vegetables may emerge as having the same anti-obesity potential. To date the richest vegetable sources of potential anti-obesity phytochemicals appear to be: the red varieties of onion, lettuce, capsicum and curly kale; and orange-fleshed varieties of sweetpotato. The cooking method influences the levels of these phytochemicals in vegetables. Boiling vegetables results in the greatest losses of water soluble phytochemicals such as the polyphenolics, through leaching, thermal degradation and oxidation. In contrast gentle stir-frying appears to result in the least losses.

The cell signalling pathways which control the initiation and development of obesity and related chronic diseases are complex and interconnected. It has been shown that appropriate combinations of dietary phytochemicals can interact positively with these pathways. Therefore promotion of the consumption of vegetables that are rich in a wide variety of potentially anti-obesity phytochemicals and which are cooked to best maintain the levels of these agents may assist in the dietary control of obesity and related chronic diseases through additive or even synergistic mechanisms.

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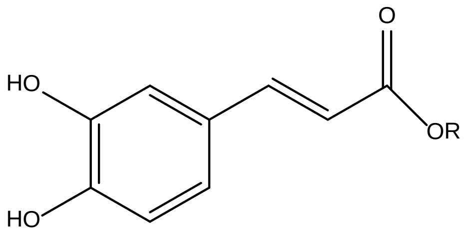
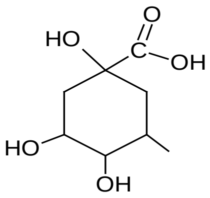
(a) Phenolic acids

Hydroxycinnamic acids

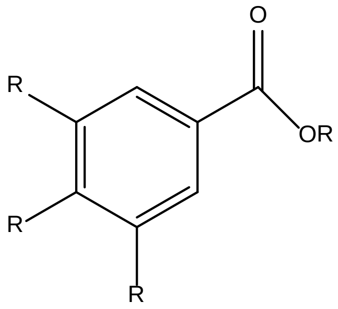


Caffeic acid R = H

Chlorogenic acid R = Quinic acid

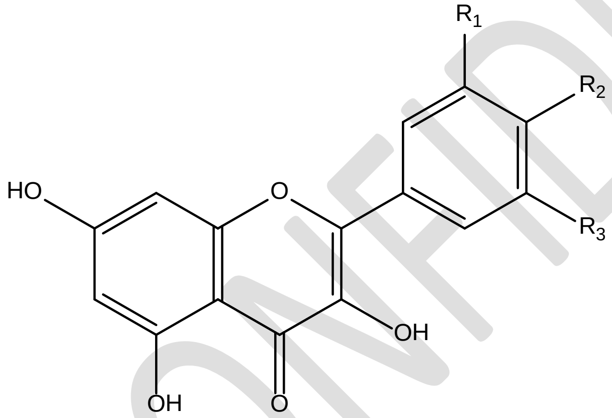


Hydroxybenzoic acids



(b) Flavonoids

Flavonols



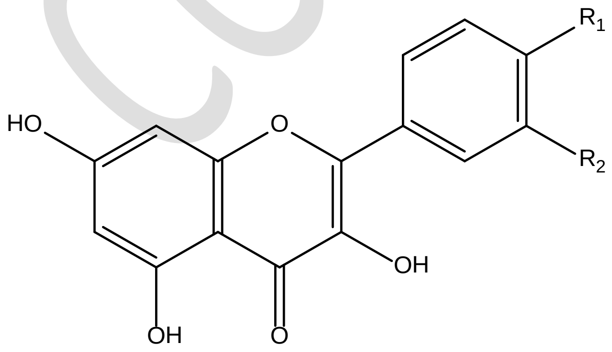
Quercetin $R_1 = H$; $R_2 = R_3 = OH$

Kaempferol $R_1 = R_3 = H$; $R_2 = OH$

Myricetin $R_1 = R_2 = R_3 = OH$

Isorhamnetin $R_1 = H$; $R_2 = OCH_3$; $R_3 = OH$

Flavones



Luteolin $R_1 = R_2 = OH$

Apigenin $R_1 = OH$; $R_2 = H$

Figure 1. Phytochemicals with reported anti-obesity effects

Table 1 Hydroxycinnamic acid content (mg/kg) of vegetables

Vegetable	Chlorogenic acid+ Caffeoylquinic acids	Reference
Carrot	150 – 260 [a]	Mattila & Hellstrom, 2007
Globe artichoke	2930	Ferracane et al. 2008
Lettuce-red leafed-red tissue	1696 [a]	Ferreres et al. 1997
Lettuce-red leafed-green tissue	570 [a]	Ferreres et al. 1997
Lettuce-red leafed-midribs (white tissue)	213 [a]	Ferreres et al. 1997
Potato-small russet	133 [a]	Friedman, 1997
Potato-large russet	142 [a]	Friedman, 1997
Potato roots	260 [a]	Friedman, 1997
Potato tubers	170 [a]	Friedman, 1997
Sweetpotato-cream fleshed	31 – 46 [a]	Rautenbach et al. 2010
Sweetpotato-orange fleshed	190 – 580 [b]	Padda & Picha, 2008
Sweetpotato-purple fleshed	1150 [b]	Padda & Picha, 2008
Sweetpotato-white fleshed	93 – 910 [b]	Padda & Picha, 2008
Sweetpotato leaves	4305 – 4630 [a]	Truong et al. 2007
Sweetpotato peel	585 – 1050 [a]	Truong et al. 2007
Sweetpotato whole root	80 – 320 [a]	Truong et al. 2007
Sweetpotato periderm	20 -1825 [b]	Harrison et al. 2008
Sweetpotato cortex	1830 – 12440 [b]	Harrison et al. 2008
Sweetpotato stele	505 – 12205 [b]	Harrison et al. 2008
Sweetpotato leaves-immature	88500 [b]	Padda & Picha, 2007
Sweetpotato root-small	10300 [b]	Padda & Picha, 2007

[a] data expressed as mg/kg fresh weight
[b] data expressed as mg/kg dry weight

Table 2 Flavonol content (mg/kg fresh weight) of vegetables

Vegetable	Quercetin	Kaempferol †	Myricetin †	Reference
Broccoli	40	40		Harnly et al. 2006
Broccoli	30 – 37	60 – 72		Hollman & Arts, 2000
Capsicum-red	34			Sun et al. 2007
Capsicum-yellow	30			Sun et al. 2007
Capsicum-orange	29			Sun et al. 2007
Capsicum-green	27			Sun et al. 2007
Kale	110 – 120	211 – 470		Hollman & Arts, 2000
Kale-red curly	467	480		Olsen et al. 2010
Lettuce-green leafed	11 – 147			Crozier et al. 1997
Lettuce-red leafed	450 – 911			Crozier et al. 1997
Lettuce-red leafed-green tissue	244			Ferreres et al. 1997
Onion-red	201			Crozier et al. 1997
Onion-red	334	11	27	USDA, 2007
Onion-red	415 – 1917			Slimestad et al. 2007
Onion-inner leaves	26	0.6	0.2	Chu et al. 2000
Onion-outer leaves	59	5	nd	Chu et al. 2000
Onion-red-dry skin	1900			Gennaro et al. 2002
Onion-red-outer fleshy layer	660			Gennaro et al. 2002
Onion-red-edible portion	600			Gennaro et al. 2002
Onion-yellow	270 - 1187			Slimestad et al. 2007
Onion-yellow	214	6	0.2	USDA, 2007
Onion-white	185 – 634			Crozier et al. 1997
Sweetpotato leaves-purple	270		156	Chu et al. 2000
Sweetpotato leaves-green	144		39	Chu et al. 2000

† blank entries indicate component levels not reported in study

Table 3 Flavone content (expressed as mg/kg fresh weight) of vegetables

Vegetable	Luteolin [†]	Apigenin [†]	Reference
Broccoli	8	nd	Harnly et al. 2006
Capsicum-green	21	nd	Arabbi et al. 2004
Capsicum-green	2		Sun et al. 2007
Capsicum-orange	7		Sun et al. 2007
Capsicum-red	11		Sun et al. 2007
Capsicum-red	5 – 11		Hollman & Arts, 2000
Capsicum-yellow	9	nd	Arabbi et al. 2004
Celery	13	46	Harnly et al. 2006
Celery leaf	200	750	Hollman & Arts, 2000
Celery stalk-white	38	97	Crozier et al. 1997
Celery heart-green	35	191	Crozier et al. 1997
Celery stalk	5 – 20	61	Hollman & Arts, 2000
Globe artichoke	75	100	Azzini et al. 2003
Celery heart-white	7	17	Hollman & Arts, 2000
Onions-inner leaves	0.2	0.04	Chu et al. 2000
Onions-outer leaves	nd	nd	Chu et al. 2000
Parsley	3	119	Meyer et al. 2006
Sweetpotato leaves-green	nd	2	Chu et al. 2000
Sweetpotato leaves-purple	4	nd	Chu et al. 2000

[†] blank entries indicate component not reported in study

nd- not detected