

*Full Length Research Paper*

# Variation of quercetin content in different tissues of Welsh onion (*Allium fistulosum* L.)

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Accepted 3 September, 2011

Due to its proposed potential role in reducing risk of cardiovascular disease and some forms of cancer, flavonol level in plants is an important trait. In this study, nine Welsh onion cultivars were grown in 2006 in Jinan, and six cultivars were grown in 2007 in Qingdao to study the distribution of quercetin (a flavonol) among different tissues of the Welsh onion. After harvest, quercetin levels in leaves, upper pseudo stems, and inner and outer rings of lower pseudo stems were measured for each cultivar. Large variations in quercetin levels were observed among cultivars. 'Jilin Xiao Cong', a local cultivar of Northern China, had significantly higher quercetin levels in pseudo stems (about 4 times) than the other cultivars in the Jinan trial ( $P < 0.05$ ). Leaves of 'Zhangqiu Da Cong' and 'Shi Guo Yi Ben Tai' had higher quercetin levels than those of the other four cultivars in the Qingdao trial ( $P < 0.05$ ). Leaves contained significantly higher amounts of quercetin than pseudo stems ( $P < 0.05$ ). The inner core tissues of pseudo stems had significantly higher levels of quercetin than the outer rings ( $P < 0.05$ ). The results indicated that intake more leaves of Welsh onion may contribute a significant amount of flavonoids to one's diet and health benefits. The results of this experiment are useful for plant breeders who are interested in improving this trait in Welsh onion.

**Key words:** *Allium fistulosum* L., Welsh onion, flavonols, quercetin, variation, spectrophotometry.

## INTRODUCTION

Welsh onion (*Allium fistulosum* L. var. *giganteum* Makino) is an important vegetable in Asian countries, especially China, Japan and Korea. Welsh onion leaves contain high levels of quercetin, a flavonol compound with potential benefit to human health. Quercetin was reported to have protective effects in reducing the risk of cardiovascular disease and act as anti-cancer and antioxidant agents due to its antiprostanoic and anti-inflammatory responses and decreased rate of DNA degradation (Crystal et al., 2003).

Information of quercetin has been thoroughly documented in bulb onions (*A. cepa*). Flavonol levels in the edible portion of *Allium* vegetables including leeks, shallots, green onions, garlic, and onions range from less than 0.03 to 1 g/Kg of vegetables (Leighton et al., 1992). Bulb onions ranked highest in quercetin content in a survey of 28 vegetables and nine fruits (Herrmann, 1976;

Hertog and Hollman, 1996). Amounts of quercetin in bulb onions are high in the outer rings (Patil and Pike, 1995) and vary with bulb color, type, and cultivars (Leighton et al., 1992; Patil and Pike, 1995). Red and yellow colored onions have higher quercetin levels than white onions (Patil et al., 1995; Lombard et al., 2002).

Improvement of quercetin content in red bulb onions (*A. cepa*) cv. 'Red Raider' is usually manipulated through selective breeding (Crystal et al., 2003). Similarly, improvement of quercetin content in Welsh onions might be achieved through traditional breeding. However, variation of quercetin content in various tissues of Welsh onion cultivars is still lacking. The objective of this study was to determine the variation and distribution of quercetin in various tissues of Welsh onion cultivars.

## MATERIALS AND METHODS

Nine Welsh onion cultivars, provided by the Institute of Vegetables and Flowers, Shandong Academy of Agricultural Sciences, were seeded on April 12, 2006 in Jinan, China (Table 1). Seedlings were transplanted on August 9, 2006 with a spacing of 100 cm between

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**Table 1.** Cultivars used in the 2006 and 2007 trials in Jinan and Qingdao, respectively.

Jinan, 2006	Qingdao, 2007
Don Jing Xia Hei	Don Jing Xia Hei
Laizhou Da Cong	Takihikari
Gong Chuan Yi Ben Tai	Gong Chuan Yi Ben Tai
Takihikari	Tie Gan Da Cong
Zhonghua Ju Cong	Zhangqiu Da Cong
Fu Cong	Shi Guo Yi Ben Tai
Chang Bai Tiao	
Tie Gan Da Cong	
Jilin Xiao Cong	

rows and 5 cm within rows. The plants were harvested on October 26, 2006 and shipped to Qingdao Agricultural University in a cooler (5°C) for quercetin analysis. Another six Welsh onion cultivars, purchased from the Qingdao Academy of Agricultural Sciences, were seeded in the Chengyang Experiment Station of Qingdao Agricultural University (Qingdao, China) on January 18, 2007. Seedlings were transplanted on May 29, 2007 with a spacing of 100 cm between rows and 5 cm within rows, and were harvested on October 12, 2007 (Table 1). Each trial was a completely randomized block design with three blocks. Five plants per cultivar were evaluated for quercetin content within each block. For the Jinan trial, leaves of onion plants were removed and only stems were used for quercetin analysis. In the Qingdao trial, each plant was cleaned with tap water and divided into portions of leaves, upper stems, and lower stems. Lower stems were further divided into outer rings (1 to 3) and inner rings (the core tissue).

#### Quercetin analysis

Each fresh sample (about 100 g) was chopped into small pieces, immediately frozen with liquid nitrogen, and ground to a fine powder using a FM-100 high speed grinder (Tianjin, China) and quercetin was extracted from 1 g of ground Welsh onion powder in 10 ml of 80% EtOH at room temperature for 24 h (Liu et al., 2008). The solution was passed through a medium flow rate filter paper (Hangzhou Xinhuan Group, Hangzhou, China), and then 0.6 ml of the extracts were diluted 5:1 with 80% ethanol to a total of 3 ml. Absorbance (AU) readings were made at 362 nm using a UV-2550 spectrophotometer (Shimadzu, Japan) and quercetin content was calculated based on a standard curve. When the concentration was within a range of 1 to 10 mg/L, the absorbance and concentration of quercetin showed a good linear relationship ( $r = 0.9702$ ) (Liu et al., 2008). The quercetin standard with a purity of 99.0% was purchased from Beijing UE Biotech Co. Ltd. (Beijing, China).

#### Statistical analysis

Analysis of variance ( $n = 5$  plants/cultivar) was performed using general linear model (Proc GLM) of SAS (SAS Institute Inc., Cary, N.C.). Means were separated by Fisher's protected least significant difference (LSD) test ( $P < 0.05$ ).

## RESULTS

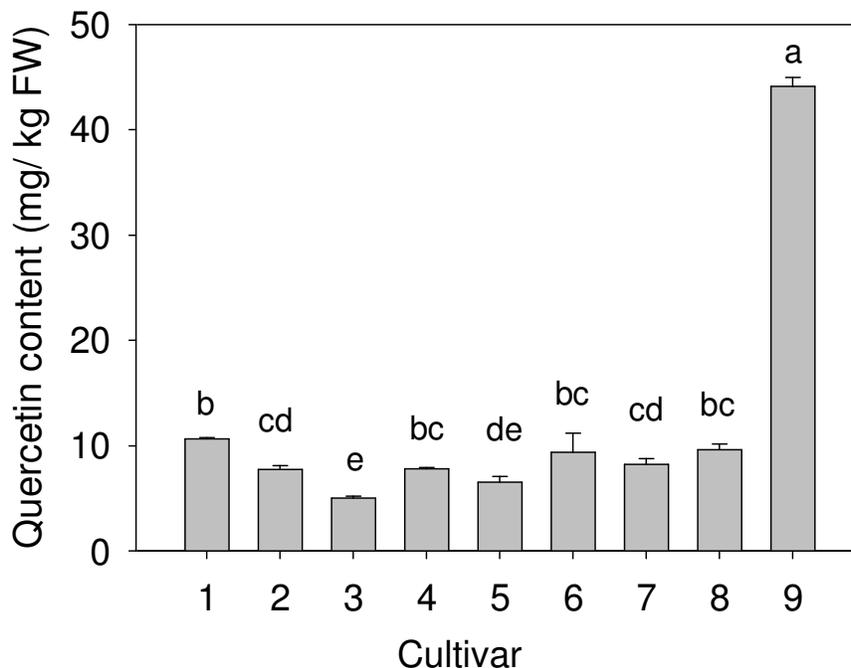
A large range of quercetin level in pseudo stems was

observed among the nine cultivars grown in 2006 in Jinan (Figure 1). Significant differences among the tested cultivars were found.

Mean quercetin levels ranged from 5.04 to 44.14 mg/Kg FW. 'Jilin Xiao Cong' had the highest amount of quercetin, followed by 'Don Jing Xia Hei', 'Fu Cong', 'Tie Gan Da Cong', 'Chang Bai Tiao' and 'Laizhou Da Cong'. 'Gong Chuan Yi Ben Tai' had the least amount of quercetin among the nine cultivars. Most of the evaluated cultivars had lower levels of quercetin (10 mg/Kg FW) than previously reported (Xu et al., 2005). The high quercetin level in 'Jilin Xiao Cong', which was at least 4 times as much as that of the other eight cultivars, and was higher than that reported by Xu et al. (2005), suggests that 'Jilin Xiao Cong' is a valuable germplasm for improvement of quercetin content in Welsh onions through breeding programs. The high level of quercetin in pseudo stems of cv. 'Jilin Xiao Cong' is possibly due to its red color. Since it has been reported that quercetin level is inheritable and can be genetically modified in bulb onion (Crystal et al., 2003), genetically modifying Welsh onion for high quercetin levels is a possibility.

Significant differences of quercetin levels in leaves were found among the six tested cultivars grown in 2007 in Qingdao ( $P < 0.05$ ) (Table 2). Mean quercetin levels ranged from 186.01 to 305.45 mg/Kg FW or 2044.07 to 3329.41 mg/Kg DW, based on a mean relative water content of 91%. Quercetin level in 'Zhangqiu Da Cong' was not significantly different from that of 'Shi Guo Yi Ben Tai', but significantly higher than the other four cultivars ( $P < 0.05$ ). 'Takihikari' and 'Don Jing Xia Hei' had relatively low levels of quercetin.

In each of the six tested cultivars, leaves contained significantly higher amounts of quercetin (about 10 times) than either the lower or upper pseudo stems ( $P < 0.05$ ) (Figure 2). This may be attributed to the fact that leaves get more sunlight than the pseudo stems, as the expression of the chalcone synthase (CHS) gene, a key enzyme in quercetin biosynthesis, is regulated by blue light (Wang and Wang, 2002). There were no significant differences in quercetin content between lower and upper



**Figure 1.** Quercetin content of pseudo stems of nine Welsh onion cultivars grown in 2006 (Jinan, China). Bars with the same letters indicate no significant difference between mean quercetin content of cultivars ( $P < 0.05$ ). Cultivars: 1= 'Don Jing Xia Hei'; 2= 'Laizhou Da Cong'; 3= 'Gong Chuan Yi Ben Tai'; 4= 'Takahikari'; 5= 'Zhonghua Ju Cong'; 6= 'Fu Cong'; 7= 'Chang Bai Tiao'; 8= 'Tie Gan Da Cong'; 9= 'Jilin Xiao Cong'. There were no statistical differences among the means shown by the same letters at 5% probability level.

**Table 2.** Mean quercetin contents of leaves in six Welsh onion cultivars grown in 2007, Qingdao.

Cultivar	Mean $\pm$ SE (mg/kg FW)*
Takahikari	186.01 $\pm$ 5.2 <sup>c</sup>
Don Jing Xia Hei	189.59 $\pm$ 9.56 <sup>c</sup>
Gong Chuan Yi Ben Tai	227.58 $\pm$ 7.09 <sup>bc</sup>
Tie Gan Da Cong	246.45 $\pm$ 10.99 <sup>b</sup>
Zhangqiu Da Cong	305.45 $\pm$ 12.6 <sup>a</sup>
Shi Guo Yi Ben Tai	268.43 $\pm$ 9.99 <sup>ab</sup>

\*Different letters within a column indicate significant differences between cultivars at the  $P < 0.05$  level.

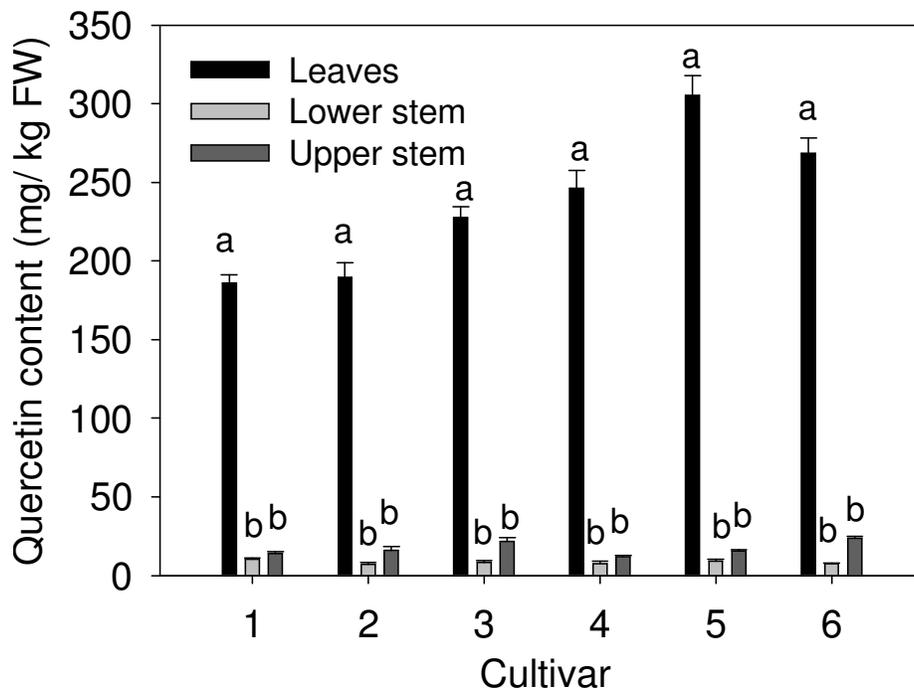
pseudo stems for each of the six tested cultivars ( $P > 0.05$ ). This result suggests that consuming leaves of Welsh onions may enhance intake of quercetin.

For pseudo stems in each of the six tested cultivars in 2007, quercetin content in the inner tissues was significantly higher than that of the outer 1 to 3 rings ( $P < 0.05$ ) (Figure 3).

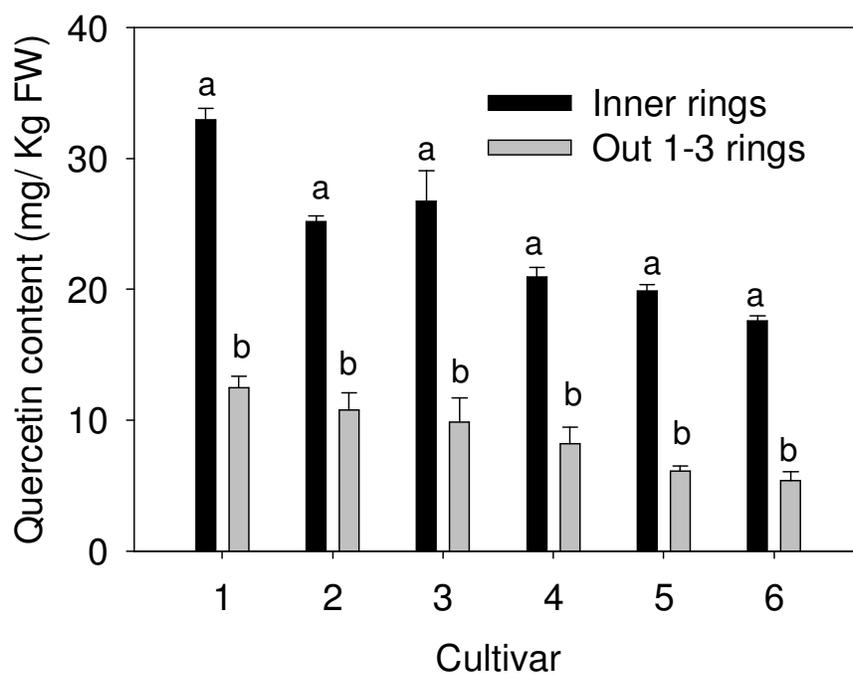
## DISCUSSION

Chinese local Welsh onion cultivars 'Laizhou Da Cong',

'Zhonghua Ju Cong', 'Chang Bai Tiao', 'Fu Cong', and 'Zhangqiu Da Cong' are widely grown in northern China, including Shandong, Hebei, Liaoning, and Henan provinces; while 'Jilin Xiao Cong' is mainly grown in Jilin province. Japanese cultivars including 'Shi Guo Yi Ben Tai', 'Don Jing Xia Hei', 'Gong Chuan Yi Ben Tai', 'Takahikari', and 'Tie Gan Da Cong' have been widely grown in eastern China, especially in Shandong province for export to Japan and Korea. Welsh onions are mainly eaten raw in Northern provinces, especially Shandong province, while in other areas of China and Japan, Welsh onions are eaten both raw and cooked.



**Figure 2.** Quercetin content in leaves and lower and upper pseudo stems of six Welsh onion cultivars grown in 2007 (Qingdao, China). Bars with the same letters indicate no significant difference between mean quercetin content of cultivars ( $P < 0.05$ ). Cultivars: 1 = 'Takahikari'; 2 = 'Don Jing Xia Hei'; 3 = 'Gong Chuan Yi Ben Tai'; 4 = 'Tie Gan Da Cong'; 5 = 'Zhangqiu Da Cong'; 6 = 'Shi Guo Yi Ben Tai'.



**Figure 3.** Quercetin content in the inner and outer rings of pseudo-stems of six Welsh onion cultivars grown in 2007 (Qingdao, China). Bars with the same letters indicate no significant differences between the inner and outer rings within a cultivar ( $P < 0.05$ ). Cultivars: 1 = 'Takahikari'; 2 = 'Don Jing Xia Hei'; 3 = 'Gong Chuan Yi Ben Tai'; 4 = 'Tie Gan Da Cong'; 5 = 'Zhangqiu Da Cong'; 6 = 'Shi Guo Yi Ben Tai'.

Unlike bulb onions which are mainly consumed for their edible bulbs, Welsh onion is mainly consumed for its notably long pseudo stem, which is composed of 3 to 4 outer rings of leaf sheath and 1 to 2 young leaves of the inner core tissue. So far, there are only few reports on quercetin in Welsh onion. Xu et al. (2005) reported that Welsh onion had 23.6 mg of quercetin per kg of fresh weight (FW), with no other flavonols detected. Meanwhile, Mian and Mohamed (2001) reported that Welsh onion leaves had a total flavonol (TF) content of 2720.5 mg/Kg of dry weight (DW), with 1497.5 mg/Kg of quercetin, 391 mg/Kg of luteolin, and 832 mg/Kg of kaempferol. Mean quercetin levels ranged from 186.01 to 305.45 mg/Kg FW or 2044.07 to 3329.41 mg/Kg DW, based on a mean relative water content of 91%. Even cannot be exactly compared due to the different units of quercetin levels reported (FW vs. DW), this result is similar to that (2720.5 mg/Kg DW) of Mian and Mohamed (2001) but is much higher than that of Xu et al. (2005). However, information on variations of these different results is not clear, but may be possibly due to the various cultivars or extraction procedures used by different authors (Mian and Mohamed 2001; Xu et al., 2005).

It was reported that outer rings of bulb onion had higher quercetin levels than inner rings (Patil and Pike 1995). However results of this experiment indicated that quercetin content in the inner tissues of Welsh onion was significantly higher than that of the outer 1 to 3 rings ( $P < 0.05$ ). The inner core of the Welsh onion pseudo stem is composed of young green leaves, which has relatively high quercetin content. While in bulb onions, the inner rings do not get as much sunlight exposure as the outer rings. More experiments are needed to elucidate the difference of quercetin content distribution between bulb onion and Welsh onion. As stems contain lower amount of quercetin than leaves, increasing quercetin content in stems of Welsh onion through breeding or biotechnology will be more important in the future.

In summary, great variation of quercetin levels existed among pseudo stems and leaves in Welsh onion cultivars. 'Jilin Xiao Cong' had significantly higher quercetin levels in pseudo stems (about 4 times) than the other eight tested cultivars in 2006 ( $P < 0.05$ ). Leaves had significantly higher amounts of quercetin than pseudo stems ( $P < 0.05$ ). The inner core portion of pseudo stems

had significantly higher levels of quercetin than the outer rings ( $P < 0.05$ ). The results of this experiment are useful for those plant breeders who are interested in the improvement of this trait.

## ACKNOWLEDGEMENTS

This research was supported by the Doctoral Research Foundation of Qingdao Agricultural University.

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