INNSPUB

International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print) 2222-5234 (Online)
http://www.innspub.net
Vol. 4, No. 9, p. 147-153, 2014

RESEARCH PAPER OPEN ACCESS

# Variation of rutin content in different parts of *Capparis spinosa* during their phenological cycles

**Moghaddasian Behnaz[1*], Eradatmand Asli Davood[2], Alaghemand Atena[1],Torabi Ali[1]**

[1]*Department of Horticulture, Faculty of Agriculture, Saveh Branch, Islamic Azad University, Saveh, Iran*

[2]*Department of Agriculture, Payame Noor University, Iran*

**Key words:** *Capparis spinosa*, medicinal use, Rutin, HPLC.

http://dx.doi.org/10.12692/ijb/4.9.147-153 Article published on May 10, 2014

**Abstract**

With an increasing focus globally on medicinal plant, *Capparis spinosa* are considered to be an important source of therapeutic compounds and therapeutic benefit of this plant is attributed to their antioxidant properties. Rutin as a natural antioxidant display a remarkable role in pharmacological activities of caper plant. The aim of this study was determine morphogenetic variation of rutin content among different parts of *C.spinosa*. Caper plants were harvested during May and August then separated into root, stem, leaf, bud, flower, fruit and seed. After drying and extracting, samples were assayed for HPLC analysis. Morphogenetic change of rutin varied during developmental cycles. Leaves contained highest amount of rutin at vegetative, floral budding and full flowering stages while maximum quantity of rutin was found in floral buds and fruit at fresh fruiting and mature fruiting stages respectively.

* **Corresponding Author:** Moghaddasian Behnaz ✉ bmoghadasian.1014@yahoo.com

## Introduction

*Capparis spinosa* L. (Capparidaceae) is a winter deciduous, spiny, woody perennial native in the southeastern Mediterranean, which was described by Theophrastus 24 centuries ago (Rhizopoulou, 1990). Consumption of capers and caperberries has a long history. Direct evidence of the consumption of *Capparis* spp. from 18,000 to 17,000 years ago was obtained by archaeological excavations from an Old World Palaeolithic site (Wadi Kubbaniya, west of Nile Valley, Upper Egypt)( Hillman, 1989).

A considerable amount of literature exists on the phytochemical constituents of caper bush, capers and caperberries (Sozzi, 2001). Different organs of the caper plant have been used as folk remedies for various diseases (Boulos, 1983, Duke, 1983, Jain and Puri, 1984, Abbas *et al.*, 1992, Husain *et al.*, 1992, Al-Said, 1993, Ghazanfar and Al- Sabahi, 1993, Ghazanfar, 1994, Bhattacharjee, 1998). *C. spinosa* is also used in phytomedicine as antifungal (Ali-Shtayeh and Abu Ghdeib, 1999), antihepatotoxic (Gadgoli and Mishra, 1995), anti-inflammatory (Ageel *et al.*,1986)chondroprotective/antidegenerative (Panico *et al.*, 2005) and antileishmania (Jacobson and Schlein, 1999).

The genus *C.spinosa* has been receiving a lot of publicity recently, because it is a source of the most important biologically active constituents namely, rutin, which possess a wide array of biological properties. Rutin has attracted considerable interest as dietary constituents and results from clinical studies indicated their possible role in protective effect against the development of diabetes (Srinivasan *et al.*, 2005), anti-inflammatory, analgesic(Pietta and Gardana, 2003), anti-mutagenic (Brindzova *et al.*, 2009), anticarcinogenic (Webster *et al.*, 1996), antifungal (Han, 2009). Increased market demand for rutin has led to several investigations in order to find valuable natural source for it. The aim of this study was to determine if there was a link between rutin content of plant materials and developmental stages during phenological cycles.

## Materials and methods

*Reagents and materials*

*Plant materials*

The Caper plant were collected from natural habitats in Tafresh of Markasi province (50̊ 01' N Lat., 34̊ 41' E Long., and 1980 m elevation), between May and August of 2011 at different stages of plant development. Collection was done once a day (11 AM) for each development stage. Details of weather condition are given in table1.

Sampling was done in this wild population by a randomized collection of 10 individuals for following phenological stages:

1-Vegetative stage: Root, stem and Leave.

2-Floral budding stage: Root, stem, Leave and bud.

3-Full flowering stage: Root, stem, Leave, bud and flower.

4-Fresh fruiting stage: Root, stem, Leave, bud, flower and fresh fruit.

5-Mature fruiting stage: Root, stem, Leave, bud, flower, fresh fruit and seed.

The plant materials were dried at an ambient room separately then grounded for 30 min and subsequently were assayed for rutin content. Air-dried plant material was mechanically ground with a laboratory mill to obtain a homogenous powder.

*Reagents and chemicals*

Methanol and acetic acid were of HPLC grade and were purchased from Merck Company. Deionized water was prepared by a Milli-Q Water Purification system. Rutin standard were purchased from Sigma Company.

*Preparation of sample solution*

An amount of 0.1-0.5 g of ground plant material was extracted with 10 ml of solution (methanol-acetic acid- water 100:2:100) for 1 hour on a shaker at laboratory temperature. 2 ml of the extract were centrifuged for 10 min at 2000 rot/min. Then solution was filtered through a micro filter with a regenerated cellulose membranes of the pore size 0.22 .The filtrate was applied for HPLC. Detection

with UV detector was carried out at 355 nm. Retention time for rutin was 6.72 min and the peak area of the sample was compared to the standard.

*Preparation of standard solutions*

Standard stock solutions of rutin were prepared in ethanol, at concentration of 1, 5, 10 and15 ppm. All sample solutions were filtered through 0.22 µm membrane filter and injected directly.

*HPLC condition*

Chromatographic analysis was carried out by using C18 column (4.6mm × 250mm) as the stationary phase and methanol: acetonitrile: water (10:10:75) containing 5% acetic acid as the mobile phase. Flow rate and injection volume were 1.0 ml/min and 10 µm respectively. The chromatographic peaks of the analytics were confirmed by comparing their retention time and UV spectra with those of the reference standards.

*Data analysis*

Data for rutin contents of plant material including whole plant, aerial parts and all tissue were subjected to ANOVA separately. Significant differences among mean values were tested with the Duncan Multiple Range Test ($P < 0.01$) by using MSTAT-C statistical software.

**Result and discussion**

The differences in rutin contents of whole plant at different development stages were found to be significant ($P < 0.01$).

**Table 1.** Weather condition in experimental months.

| Month | Air temperature | | Relative humidity | | Sunshine duration (h) (mm) | Total precipitation |
|---|---|---|---|---|---|---|
| | Min | Max | Min | Max | | |
| May | 10 | 20.6 | 32 | 63 | 221.2 | 54.5 |
| June | 16 | 28.9 | 14 | 38 | 341.1 | 5.4 |
| July | 21.2 | 32.8 | 11.2 | 28 | 321.6 | 0.0 |
| August | 21.4 | 33.1 | 13 | 30 | 332.2 | 4.5 |

Rutin content in whole plant and aerial parts didn't follow the same trend (table 2) and higher accumulation level for whole plant was observed at fresh fruiting stage(1.3%) and for aerial part it was reached at vegetative and fresh fruiting stages (1.4% & 1.3% respectively)(Fig 1).

Significant differences were also observed among different parts of caper plant with regard to rutin accumulation during phenological cycle ($P < 0.01$)(table 3).

Leaves harvested at vegetative, floral budding and full flowering stages produced the highest content of rutin (2.47%, 2.6% and 1.5% respectively). On the contrary buds and flowers were found to be superior at fresh fruiting and mature fruiting stages in terms of rutin accumulations(1.99% and 1.29% respectively)(Fig 2,3).

Rutin concentrations in tissues can vary significantly during the course of ontogenesis, which may be relevant for the utilization of medicinal plants. In the present study, the highest levels of rutin in whole plants was reached at fresh fruiting stage. Our result did not match those described for *H . triquetrifolium* whose larger amounts of rutin, was reached at flowering time (Hosni *et al*,2011). Similarly, the highest content of rutin, was determined at flowering in greenhouse-grown *Hypericum brasiliense* Choisy (Abreu *et al*, 2004).

The differences in rutin content among different parts of caper plant are in agreement with pervious results

recorded about *C. spinosa* and other plants. Highest amount of rutin was observed in the leaves of *C.spinosa* (Musallam *et all*,2011,Ramezani *et all*, 2008), buckwheat (Kalinova and Dadakova, 2009), amaranth (Kalinova *et all*, 2006). Highest level of Rutin distribution among different part didn't follow the same trent at fresh fruiting and mature fruiting stage. Floral buds and flower allocated the highest content of rutin at mentioned stages respectively.

**Table 2.** Rutin content in *C.spinosa* (% W/W).

| *C.spinosa* | | |
|---|---|---|
| Developmental stages | Aerial part | Whole plant |
| Vegetative | 1.4 | 0.9 |
| Floral budding | 1.3 | 1.1 |
| Full flowering | 1.1 | 0.8 |
| Fresh fruiting | 1.3 | 1.3 |
| Mature fruiting | 0.4 | 0.3 |

**Table 3.** Variation of rutin content in different parts of *C.spinosa* during phenological cycles (%w/w).

| | Plant organs | | | | | | |
|---|---|---|---|---|---|---|---|
| Developmental stages | Root | Stem | Leave | Bud | Flower | Fruit | Seed |
| Vegetative | 0.04 | 0.4 | 2.47 | | | | |
| Floral budding | 0.10 | 0.2 | 2.6 | 1.2 | | | |
| Full flowering | 0.2 | 0.17 | 1.5 | 1.3 | 1.3 | | |
| Fresh fruiting | 0.16 | 0.87 | 1.8 | 1.99 | 1.8 | 0.13 | |
| Mature fruiting | 0.19 | 0.21 | 0.67 | 0.10 | 1.29 | 0.07 | 0.15 |

Variation of secondary metabolites have been investigated over several decades, e.g. essential oil changes during the course of ontogenesis in *Hypericum perforatum*(Schwob *et all*,2004) and artemisinin changes during phonological cycle of *Artemisia annua*(Gupta *et all*, 2002 ). In common buckwheat the rutin content in leaves increased with the age of plant with the maxim at the stage of maturity or at the stage of full flowering in case of dry weather conditions after flowering and in stems the rutin content slightly decreased (Kalinova *et all*, 2006).Also In *Amarantha.tricolor* and *A.caudatus* rutin content in leaves increased with the plant age (Modi, 2007).

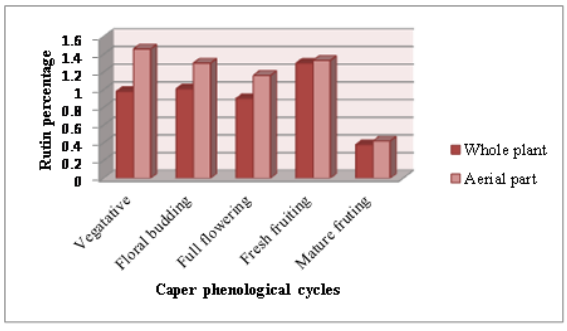


**Fig. 1.** Change of rutin content in *C.spinosa* during phenological cycles.

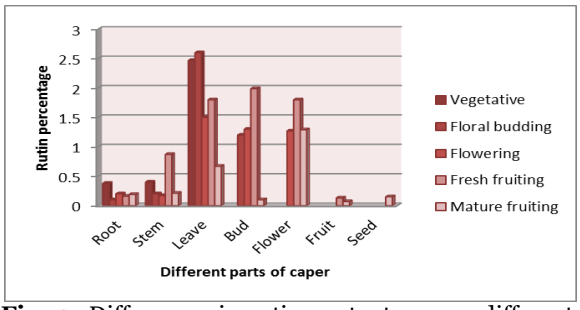


**Fig. 2.** Differences in rutin content among different parts of caper.

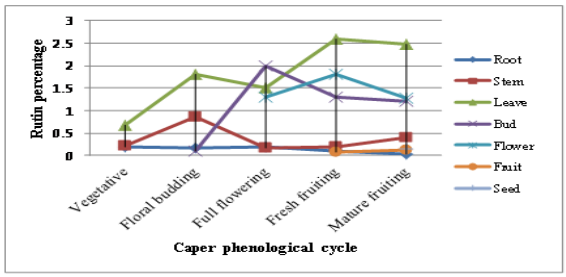


**Fig. 3.** Differences in rutin content during the developmental stages of caper plant.

Chemical concentrations vary considerably during the course of ontogenesis in a medicinal plant, not only do the concentrations of plant chemicals fluctuate through the season, but they can also be short-lived

and experience rapid turnover (Smith *et al.* 1996). Plants in wild are subjected to many of biotic/abiotic stress factors and have evolved different defence systems to avoid the damage caused by them (Carrasco *et al.* 2001).

The expression of flavonoid biosynthesis regulatory genes appears to be highly dependent on tissue type and/or response to internal or external signals such as hormones, light, microbial elicitors, UV radiation, sugars, phosphate limitation, or cold stress, which affect the signal transduction and gene expression involved in biosynthesis (Dixon and Pavia, 1995, Laura *et al.*, 2007, Leyva *et al.*, 1995, Mol *et al.*, 1996, Tsukaya *et al.*, 1991). Hormones such as cytokinin and gibberellic acid could be active a few flavonoids production enzymes (Seymour *et all*,1993) and It has been reported that sucrose and other sugars are involved in responses to many biotic and abiotic stresses, cross-talking with hormones (Gazzarrini and McCourt.

**Conclusion**

It can be concluded that there is a close relationship between rutin content with plant tissue and growth stages during phonological cycles in caper plant which offers the best harvest time and plant tissue for obtaining higher quantitative of rutin. Increased demand for finding natural source of rutin brings attention for breeding and improvement of wild caper 2003; Gibson, 2004) and modulating the expression of many genes implicated in photosynthesis, respiration, nitrogen metabolism, and defense processes (Jang *et al.*, 1997).

Morover, caper plant were collected from natural conditions, so it is not easy to separate the effects of individual factors from multifactorial influence of the environment. This would be possible in situation where the factors can be set and regulated exactly.

**References**

**Abbas JA, El-oqlah AA, Mahasneh AM.** 1992. 'Herbal plants in the traditional medicine of Bahrain'. Economic Botany. **46,** 158–163.

**Abreu IN, Porto ALM, Marsaioli AJ, Mazzafera P.** 2004. Distrubution of bioactive Substances from Hypericum brasiliense during plant growth. Plant Science **167,** 949- 954.

**Ageel AM, Parmar NS, Mossa JS, Al-yahya MA, Al-said MS, Tariq M.** 1986. 'Anti-inflammatory activity of some Saudi Arabian medicinal plants', Agents Actions. **17(3/4),** 383–384.

**Al-said MS.** 1993. Traditional medicinal plants of Saudi Arabia, Ameican Journal of Chinese Medicine. **21,** 291–298.

**Ali-shtayeh MS, Abu ghdeib SI.** 1999. Antifungal activity of plant extracts against dermatophytes. Mycoses, **42,** 665–672.

**Bhattacharjee SK.** 1998. Handbook of medicinal plants. Jaipur. India. Pointer Publ.

**Boulos L.** 1983. 'Capparaceae', in Medicinal plants of North Africa, Algonac, M I, Reference Publications. 40- 42.

**Brindzova L, Mikulasova M, Takacsova M, Mosovska S, Pattova A.** 2009. Evaluation of the mutagenicity, antimutagenicity of extracts from the oat, buckwheat and wheat bran in the salmonella/microsome assay. Journal of Food Composition. Analysis **22,** 87-90

**Carrasco AE, Serrano ML, Zapata JM, Sabater B, Martin M.** 2001. Oxidation of phenolic compounds from Aloe barbadensis by peroxidase activity: Possible involvement in defence reactions. Plant Physiology Biochem **39,** 521-527.

**Dixon RA, Pavia NL.** 1995. Stress-induced phenylpropanoid metabolism. Plant Cell. **7,** 1085-1097.

**Duke JA.** 1983. Medicinal plants of the Bible, Buffalo, New York, Conch Magazine.

**Gadgoli C, Mishra SH.** 1995.'Preliminary screening of Achillea millefolium, Cichorium intybus and Capparis spinosa for antihepatotoxic activity', Fitoterapia. **66,** 319–323.

**Gazzarrini S, McCourt P.** 2003. Cross-talk in plant hormone signalling: what Arabidopsis mutants are telling us. Annals of Botany **91,** 605-612.

**Ghazanfar SA, Al-sabahi AMA.** 1993. 'Medicinal plants of Northern and Central Oman (Arabia)', Economic Botany. **47,** 89–98.

**Ghazanfar SA.** 1994. Handbook of Arabian medicinal plants. Boca Raton. FL. CRC Press.

**Gibson SI.** 2004. Sugar and phytohormone response pathways: navigating a signalling network. Journal of Experimental Botany **55,** 253-264.

**Gupta SK, Singh P, Bajpai P, Ram G, Singh D, Gupta MN, Jain DJ, Khanuja SP, Kumar S.** 2002. Morphogenetic variation for artemisinin and volatile oil in Artemisia annua. Industrial Crops and Products **16,** 217- 224.

**Han Y.** 2009. Rutin has therapeutic effect on septic arthritis caused by Candida albicans. International. Immunopharmacology **9,** 207-211.

**Hillman GC.** 1989. 'Late Palaeolithic plant foods from Wadi Kubbaniya in Upper Egypt: dietary, diversity, infant weaning, and seasonality in a riverine environment', in Harris, D R and Hillman G C, Foraging and farming: the evolution of plant exploitation, London, Unwin Hyman. 207–239.

**Hosni K, Msaada K, Taarit MB, Marzouk B.** 2011. Phenological variations of secondary metabolites from Hypericum triquetrifolium Turra. Biochemical Systematics and Ecology **39,** 43-50.

**Husain A, Virmani OP, Popli SP, Misra LN, Gupta MM, Srivastava GN, Abraham Z, Singh AK.** 1992. Capers and caperberries 251 Dictionary of Indian medicinal plants, Lucknow, India, Central Institute of Medicinal and Aromatic Plants.

**Jacobson RL, Schlein Y.** 1999. 'Lectins and toxins in the plant diet of Phlebotomus papatasi (Diptera: Psychodidae) can kill Leishmania major promastigotes in the sandfly and in culture', Annals of Tropicals Medicine and Parasitology. **93,** 351–356

**Jain SP, Puri HS.** 1984. 'Ethnomedicinal plants of Jaunsar-Bawar hills, Uttar Pradesh, India', Journal of Ethnopharmacology. **12,** 213–222.

**Jang JC, Leon P, Zhou L, Sheen J.** 1997. Hexokinase as a sugar sensor in higher plants. Plant Cell. **9,** 5-19

**Kalinova J, Dadakova E.** 2009. Rutin and total quercetin content in Amaranth(Amaranthus SPP). Plant foods for human nurition **64,** 68-74.

**Kalinova J, Triska J, Vrchotova N. Distribution of vitamin E,** 2006. squalene, epicatechin, and rutin in common buckwheat plants (Fagopyrum esculentum Moench). Journal of Agricultural and Food Chemistry **54,** 5330-5335.

**Laura R, Franceschetti M, Ferri M, Tassoni A, Bagni N.** 2007. Resveratrol production in Vitis vinifera cell suspensions treated with several elicitors. Caryologia. **60,** 169-171.

**Leyva A, Jarillo JA, Salinas J, Martinez-Zapater JM.** 1995. Low temperature induces the accumulation of phenylalanine ammonia-lyase and chalcone synthase mRNAs of Arabidopsis thaliana in a light-dependent manner. Plant Physiology. **108,** 39-46.

**Modi TA**. 2007. Growth temperature and plant age influence on nutritional quality of Amaranthus leaves and seed germination capacity. Water SA. **33(3),** 369–378.

**Mol J, Jenkins G, Schaefer E, Weiss D, Walbot V.** 1996. Signal perception, transduction, and gene expression involved in anthocyanin biosynthesis. Critical Reviews in Plant Sciences.**15,** 525-557 .

**MusallamI, Duwayri M, Shibli R, Alali F.** 2012. Investigation of Rutin Content in Different Plant Parts of Wild Caper (Capparis spinosa L.) Populations from Jordan. Research Journal of Medicinal Plant . **6,** 27-36.

**Panico AM, Cardile TV, Garufi F, Puglia C, Bonia F, Ronsisvalle G.** 2005. 'Protective effect of Capparis spinosa on chondrocytes', Life Sciences. **77,** 2479–2488.

**Pietta P, Gardana C.** 2003. Flavonoids in Herbs. In: Flavonoids in Health and Disease, Rice-Evans, C. and L. Packer (Eds.). Marcel Dekker Inc., New York, USA., 43-49.

**Ramezani Z, Aghel N, Keyghobadi H.** 2008. Rutin from different parts of Capparis spinosa growing wild in Khuzestan/Iran. Pakistan Journal of Biological Sciences. **11(5),** 768-72.

**Rhizopoulou S.** 1990. Physiological responses of Capparis spinosa L. to drought. Journal of Plant Physiology. **136,** 341-348.

**Schwob I, Jean-Marie B, Veronique M, Josette V.** 2004. Changes in essential oil composition in Saint John's wort (Hypericum perforatum L.) aerial parts during its phenological cycle. Biochemical Systematic and Ecology **32,** 735-745.

**Seymour GB, Taylor JE, Tuker GA.** 1993. Biochemistry of fruit ripening. Chapman and hall press. 453.

**Smith R, Caswell D, Carriere A, Zielke B.** 1996. Variation in the ginsenoside content of American ginseng, Panax quinquefolius L. roots. Canadian Journal of Botany **74,** 1616-1620.

**Sozzi GO.** 2001, 'Caper bush: botany and horticulture, Horticultural Reviews. **27**, 125–188.

**Srinivasan K, Kaul Land Ramarao P.** 2005. Partial protective effect of rutin on multiple low dose streptozotocin-induced diabetes in mice. Indian Journal of Pharmacology **37,** 327-328.

**Tsukaya H, Ohshima T, Naito S, Chino M, Komeda Y.** 1991. Sugar-dependent expression of the CHS-A gene for chalcone synthase from petunia in transgenic Arabidopsis. Plant Physiology **97,** 1414-1421.

**Webster RP, Gawde MD, Bhattacharya RK.** 1996. Protective effect of rutin, a flavonol glycoside, on the carcinogen-induced DNA damage and repair enzymes in rats. Cancer Letters. **109,** 185-191.