

QUANTIFICATION OF LUPEOL IN SELECTED JUICY CULTIVARS OF MANGO (*MANGIFERA INDICA* L.) POPULARLY GROWN IN TELANGANA REGION

Soujanya B*, Kiran K. Adapa,² Sreedhar M,³ Aparna K,³ and Ravinder Reddy K⁴

^{1&4}Sri Konda Laxman Telangana State Horticultural University, Rajendranagar 500030.

²Fruit Research Station, Sangareddy, 502 001 India.

³MFPI-Quality Control Laboratory, Rajendranagar, Hyderabad 500030

Email: battulasoujanya2@gmail.com

Received-04.09.2017, Revised-23.09.2017

Abstract : The mango (*Mangifera indica* L.) is a juicy stone fruit (drupe) and also one of the most important climacteric tropical fruits in the world. Numerous phytochemicals are present in mango peel and pulp, such as triterpene, lupeol which is under basic research for its potential biological effects. Present investigation about "Quantification of lupeol in selected juicy cultivars of mango (*Mangifera indica* L.) Popularly grown in Telangana region" quantified by High performance Liquid Chromatography (HPLC) method. Experiment was designated with two factorial completely randomized design and executed with the objectives of estimation of lupeol in selected juicy varieties and estimation of lupeol in selected juicy varieties during storage at ambient conditions. Among the varieties significantly chinnarasam recorded highest amount of lupeol (67.24±8.77 µg/100g). While lowest amount of lupeol was recorded in Pandurivari Mamidi (8.45±0.10 µg/100g). Among the storage days significantly highest amount of lupeol was recorded in 4th day of storage (38.63±15.93 µg/100g). While 8th and 12th day of storage were similar amount of lupeol content 29.73±5.93 µg/100g 29.53±5.94 µg/100g respectively. Lupeol content varies among the cultivars and storage days. Showed maximum amount of lupeol content at its 4th day of storage.

Keywords: HPLC (High performance Liquid Chromatography), Juicy cultivars, Lupeol, Mango, Triterpene

INTRODUCTION

The mango (*Mangifera indica* L.) is a juicy stone fruit (drupe) and also one of the most important climacteric tropical fruits in the world. It belongs to the family *Anacardiaceae* (2n=40). Mangoes are native to South Asia (Morton and Julia Frances, 1987; Kostermans A.J.H.G and Bompard, J.M, 1993). Mango is the choicest fruit among all fruits and also has great potential for export. The principal mango producing states are Uttar Pradesh, Karnataka, Andhra Pradesh, Telangana, Bihar, West Bengal and Gujarat.

Fresh mango contains a variety of nutrients but only vitamin C and folate are in significant amounts of the Daily Value as 44% and 11%, respectively (Nutrient profile for mango from USDA SR-21). Numerous phytochemicals are present in mango peel and pulp, such as the triterpene, lupeol which is under basic research for its potential biological effects. (Chaturvedi *et al.*, 2008).

Mango peel pigments under study include carotenoids, such as the provitamin A compound, beta-carotene, lutein and alpha-carotene (Berardini *et al.*, 2005; Gouado *et al.* 2007) and polyphenols, such as quercetin, kaempferol, gallic acid, caffeic acid, catechins and tannins (Mahattanatawee, 2006; Singh, U.P. 2004). Lupeol is a pharmacologically active triterpenoid. It has several potential medicinal properties. Lupeol contains 30 carbon and its molecular weight is 426.7174 g/mol, melting point 215-216°C. It is found in all vegetables, fruits and medicinal plants. Lupeol has a

complex pharmacology, display anti protozoal, anti-microbial, anti-inflammatory, antitumor and chemo preventive properties. (Rahman and Saleem, 2011). Lupeol has a potential to act as anti-inflammatory, anti-microbial, anti protozoal, antiproliferative, anti-invasive, anti angiogenic and cholesterol lowering agent. The scientists attribute the anti-carcinogenic property of mango to a chemical compound called "lupeol" found in the fruit. (Yogeshwar Shukla, 1988).

Based on the strength of germplasm available at fruit research station (FRS), Sangareddy, an experiment has been planned to estimate lupeol levels in 3 juicy cultivars viz. Navaneetham, Chinnarasam, Pandurivari Mamidi by using HPLC (1260) with reverse phase C-18 column, by using methanol : acetonitrile (30:70%) solvent as mobile phase and C18 column as stationary phase and DAD (Diode Array Detector) at 210 nm, flow rate of 1ml/min at MFPI-Quality Control Laboratory, PJTSAU, Rajendranagar, Hyderabad, India. With the objectives of estimation of lupeol content in 3 juicy cultivars and estimation of lupeol in juicy cultivars during 3 storage days (4th, 8th and 12th day) at ambient conditions. An experiment was designed with two factorial completely randomized design and analysis was performed in triplicates of samples.

METHODOLOGY

Mango cultivars Navaneetham, Chinnarasam and Pandurivari Mamidi are commercially grown in Telangana state of India. Fruits were collected from

*Corresponding Author

the Fruit Research Station, Sangareddy, India. Cultivars were harvested at fully matured stage and subjected to ethylene treatment kept for storage at ambient conditions up to 12 days.

Pharmaceutical grade lupeol was purchased from Sigma Aldrich (Mumbai, India) and all other solvents (Methanol and Acetonitrile) were used for estimation of lupeol are HPLC grade and purchased from Merck Ltd., Mumbai, India).

Stock solution of 1 mg/ml of lupeol was prepared in methanol:acetonitrile (3:7 V/V). Six working solutions of respective compounds were prepared by dilution. 0.5 ppm, 1 ppm, 5 ppm, 10 ppm, 15 ppm and 20 ppm were prepared by the stock solution.

Three juicy cultivars of mango fruits were subjected to pulp extraction and extracted pulp was kept for drying in solar drier for 45 days at 60°C. The extraction efficiency of target compound was optimized by using solvent mixture of methanol and acetonitrile (3:7 V/V) was selected to extract the

lupeol content in pulp of mango (250 mg) powder. Mango pulp powder (250 mg) was extracted through 10 ml of methanol:acetonitrile (3:7 V/V) and filtered through wattman No. 1 filter paper and vortex 5 min after this kept for overnight at room temperature. Next day solution was again subjected to vortex for 5 min and finally filtered through 0.45 µm membrane filter and taken into small vials in HPLC for the quantification of lupeol content in pulp of mango.

The lupeol content was estimated by HPLC method in dried mango powder samples were analysed at periodical intervals of 4th, 8th and 12th day of storage. Two fruits per cultivar, in duplicate, were manually separated into peel, pulp and seed kernel. Pulp was cut into small pieces and grounded into fine pulp dried in solar drier at 60°C for analysis. Mango samples were compared with the standard peak time in HPLC isocratic method. Finally lupeol (µg/100g) amount was calculated on the basis of following formula given by Anyakora *et al.* (2008).

$$\frac{\text{Standard peak area}}{\text{Sample Peak area}} \times \text{X Conc of Std} = \frac{\text{Volume of dil X 100}}{\text{Injection Volume X Wt of sample}}$$

Dil = dilution, Wt = weight, Conc = Concentration, Std = standard

The design adopted was completely randomized design with 2 factors (storage days and varieties). All the analysis was performed in triplicate of samples and the results were presented as mean and standard deviation. Data was processed at the Computer Centre, Hyderabad, using (SAS version 9.1, Statistical Analysis System Institute, Inc. C).

RESULT AND DISCUSSION

The data pertaining to lupeol content in dried mango pulp of three mango cultivars (juicy cultivars) as influenced by the storage days at ambient conditions is presented in the table 1 and fig. 1

It is indicated from the data that, as the storage days increased there was a significant decrease in lupeol levels from 4th day to 12th day of storage except Pandurivari Mamidi showed increased amount of lupeol from 4th to 8th day (8.20±0.03 to 8.71±0.10 µg/100g). Among three juicy cultivars highest amount of lupeol was recorded in Chinnarasam (67.24±8.77 µg/100g) followed by Navaneetham (22.21±4.21 µg/100g), lowest was noticed in Pandurivari Mamidi (8.45±0.10 µg/100g). Among the storage days highest amount of lupeol was recorded on 4th day of storage (38.63±15.93 µg/100g) and lowest was recorded on 12th day of storage (29.53±5.94 µg/100g). The interaction between 3 cultivars (juicy cultivars) and 3 storage days (4th, 8th and 12th day of storage) showed significant variation (table 3.1). Significantly highest amount of lupeol was noticed in Chinnarasam (102.33±0.02 µg/100g) on 4th day of storage

Table 1. Lupeol content in dried mango powder (µg/100g) as influenced by storage days (4th, 8th and 12th day) at ambient conditions in 3 juicy mango cultivars.

followed by same cultivar on 8th and 12th day of storage (49.72±0.01 µg/100g and 49.66±0.01 µg/100g). While lowest was recorded in Navaneetham (5.37±0.01 µg/100g) on 4th day of storage but on 8th and 12th day it was increased to 30.76±0.01 µg/100g to 30.49±0.09 µg/100g.

Few chromatograms are presented in fig. 2 to 4 showing the lupeol content in samples of 3 mango cultivars. The results are in agreement with Jyotshna *et al.* (2015) estimated mangiferin and lupeol content in 4 mango cultivars and reported that highest amount of lupeol was found in Dashehari (1082 µg/100g) as compared to Bombay green (505 µg/100g), Langra (167 µg/100g) and Chausa (65 µg/100g) in pulp and peel during storage period. Similar study was reported by Saratha *et al.* (2011).

CONCLUSION

Mango juicy varieties (3 commercial/popular cultivars) were studied for lupeol content, positive results have been noticed regarding the lupeol content in the selected mango cultivars (juicy cultivars), however there was lot of variation among the cultivars and lupeol content ranged from 8.45±0.10 µg/100g (Pandurivari Mamidi) to 67.24±8.77 µg/100g (Chinnarasam). Among the storage days it varied from 29.53±5.94 µg/100g (on 12th day) to 38.63±15.93 µg/100g (4th day). As evident from the study promising levels of lupeol content (µg/100g) was noticed among the cultivars and there is lot of variation in the content level of lupeol among cultivars and storage days.

Varieties	Storage days			
	4 th day	8 th day	12 th day	Mean
Navaneetham	5.37±0.01	30.76±0.01	30.49±0.09	22.21±4.21
Chinnarasam	102.33±0.02	49.72±0.01	49.66±0.01	67.24±8.77
Pandurivari Mamidi	8.20±0.03	8.71±0.10	8.44±0.04	8.45±0.10
	38.63±15.93	29.73±5.93	29.53±5.94	
Factors	SEm±		CD at 5%	
Factors (A)	0.055		0.164	
Storage days (B)	0.055		0.164	
A x B	0.096		0.284	

Note: All the values are expressed as mean ± SD. Values with similar superscripts are statistically similar at 5% level.

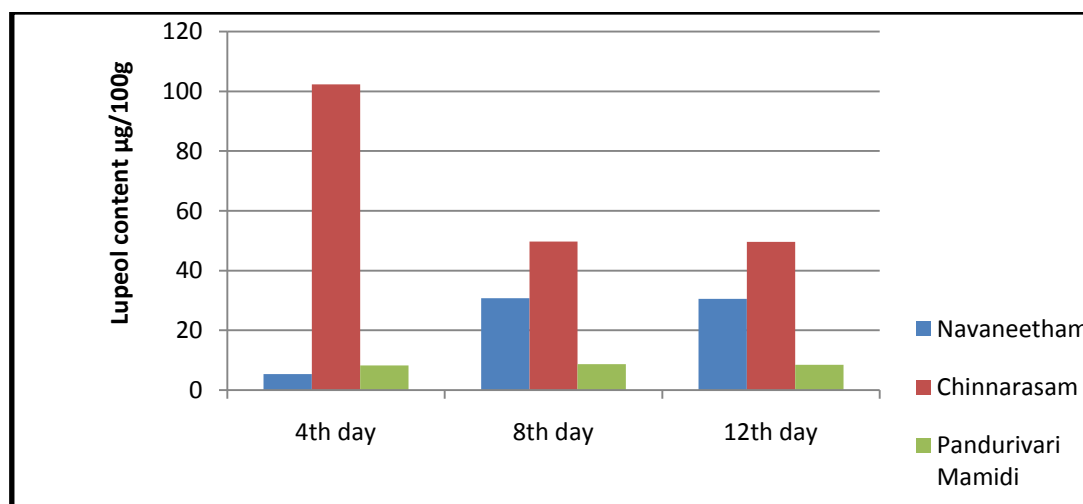
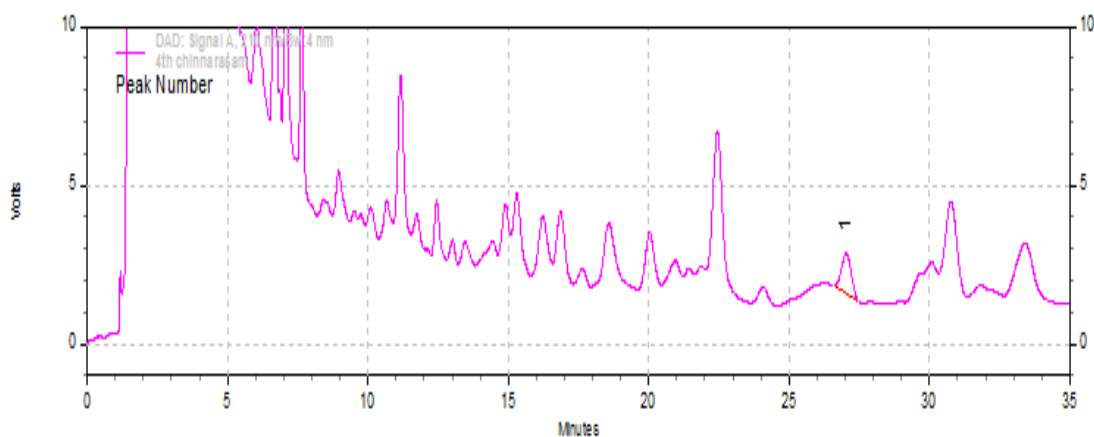


Figure 1. Lupeol content (µg/100g) in dried mango powder as influenced by storage days (4th, 8th and 12th day) at ambient conditions in 3 mango juicy cultivars.

Area % Report of Lupeol content in mango 4th day chinnarasam

Data File: D:\Lupeol\4th chinnarasam.rslt\4th chinnarasam.dat
 Method: C:\Method\Lupeol 260420161.met
 Acquired: 1/6/2017 6:20:58 PM (GMT +05:30)
 Printed: 3/14/2017 12:21:38 PM (GMT +05:30)



Name	Retention time	Area	Area %
4 th day Chinnarasam	27.04	62486	100.00

Figure 2. Chromatogram of lupeol in variety of mango Chinnarasam on 4th day obtained by HPLC Agilent 1260

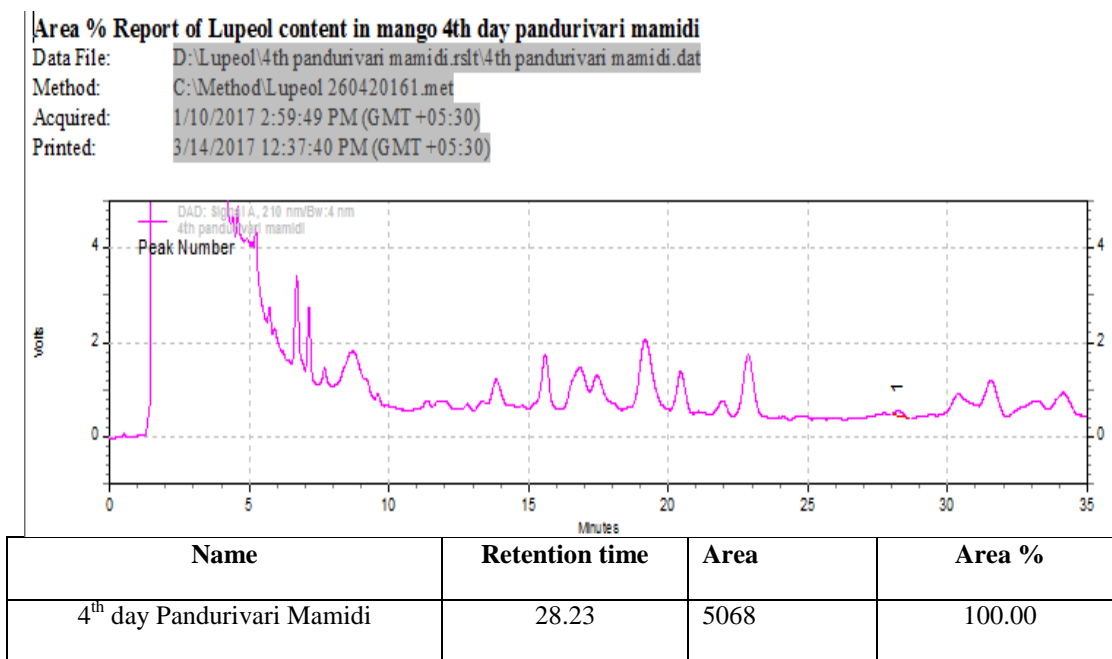


Figure 3. Chromatogram of lupeol in variety of mango Pandurivari Mamidi on 4th day obtained by HPLC Agilent 1260.

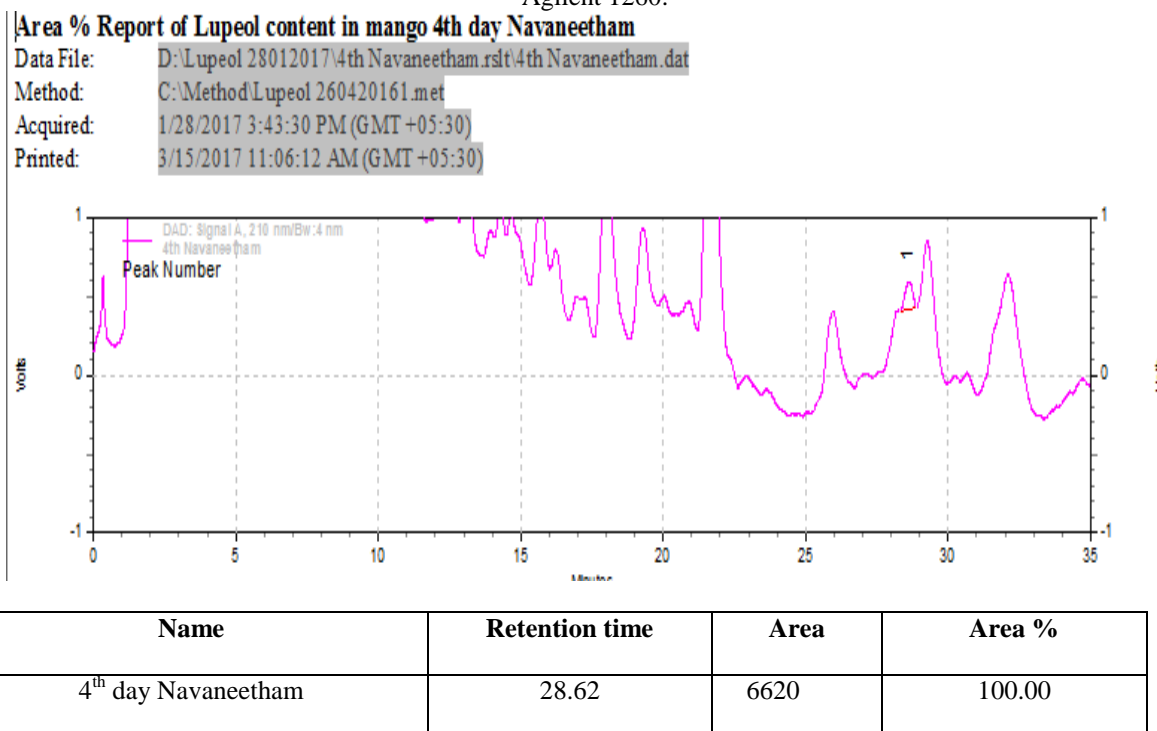


Figure 4. Chromatogram of lupeol in variety of mango Navaneetham on 4th day obtained by HPLC Agilent 1260.

ACKNOWLEDGEMENT

The authors wish to thank the Fruit research station (FRS, Sangareddy, India) and MFPI-Quality Control

Laboratory at Rajendranagar, Hyderabad, India for continuous encouragement and providing necessary facilities. Authors also acknowledge Sri Konda Laxman Telangana State Horticultural University

(SKLTSHU) for supporting this study. We are thankful to the Head of the Fruit Research Station and Quality Control Laboratory for this keen interest in the study.

REFERENCES

Nutrient profile for mango from USDA SR-21. *Nutritiondata.com*. Retrieved 31 January 2016.

Anyakora, C., Ibukam, A., Teddy, E. and Francis, O. (2008). African Journal of Pharmacy and Pharmacology. 2 (2).

Berardini, N., Fezer, R., Conrad, J., Beifuss, U., Carle, R. and Chieber, A. (2005). "Screening of mango (*Mangifera indica* L.) cultivars for their contents of flavonol O – and xanthone C-glycosides, anthocyanins, and pectin". *J Agric Food Chem.* 53(5): 1563–70.

Chaturvedi, P.K., Bhui, K. and Shukla, Y. (2008). "Lupeol: connotations for chemoprevention". *CancerLett.* 263(1):113.

Gouado, I., Schweigert, F.J., Ejoh, R.A., Tchouanguap, M.F. and Camp, J.V. (2007). "Systemic levels of carotenoids from mangoes and papaya consumed in three forms (juice, fresh and dry slice)". *Eur J Clin Nutr.* 61(10): 1180

Jyotshna., Srivastava, P., Bharti, K. and Karuna, S. (2015). Uni dimensional double development HPTLC – densitometry method for simultaneous analysis of (*Mangifera indica* L.) pulp and peel during storage. *Food Chemistry.* 176: 91-98.

Kostermans, A.J.H.G. and Bompard, J.M. (1993). The Mangoes: Their Botany, Nomenclature, Horticulture and Utilization. *Academic Press.*

Mahammad, S. (2009). Lupeol a novel anti-inflammatory and anti-cancer dietary triterpene. *Cancer Letters.* 285: 109-115.

Mahattanatawee, K., Manthey, J.A., Luzio, G., Talcott, S.T., Goodner, K. Baldwin, E.A. (2006). "Total antioxidant activity and fiber content of select Florida-grown tropical fruits". *J Agric Food Chem.* 54 (19): 7355–63.

Morton. and Julia Frances. (1987). "Mango". *Fruits of Warm Climates.* NewCROP, New Crop Resource Online Program, Center for New Crops & Plant Products, Purdue University. pp. 221–239.

Rahman, S. and Saleem, M. (2011). Beneficial health effects of lupeol triterpene; A review of preclinical studies. *Life Sciences.* 88(7-8): 285-93.

Rincon and Keer (2010). Influence of osmotic dehydration, ripeness and frozen storage on physicochemical properties of mango. *Journal of food processing and preservation.* 34 (5): 887-903.

Saratha, V., Iyyam, S. P. and Subramanian, S. (2011). Isolation and characterization of lupeol, a triterpenoid from *Calotropis gigantean* latex. *Interantional Journal of Pharmaceutical Sciences Review and Research.* 10 (2): 54-57.

Shukla, H. K. (1988). Pre and post harvest physiology of mango fruits (*Mangifera indica* L.) cv. Dashehari. Ph. D Thesis, Kanpur University. Kanpur.

Singh, U.P., Singh, D.P. Singh, M. (2004). "Characterization of phenolic compounds in some Indian mango cultivars". *Int J Food Sci Nutr.* 55 (2): 163–9.

