*Available online at* www.ijpab.com

DOI: http://dx.doi.org/10.18782/2320-7051.5730

**ISSN: 2320 – 7051**

*Int. J. Pure App. Biosci.* **5 (4):** 2141-2146 (2017)

*Research Article*

International Journal of Pure & Applied Bioscience

OPEN ACCESS

# Quantification of Lupeol in Selected Commercial Coloured Cultivars of Mango (*Mangifera indica* L.) Cultivated in Telangana Region

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Received: 8.08.2017 | Revised: 19.08.2017 | Accepted: 20.08.2017

**ABSTRACT**

*Mango (Mangifera indica L.) is one of the most important climacteric tropical fruits of the world and it is designated as "King of fruits", "National fruit of India and Philippines" and New research work about the luepol content in mango fruit (180µg/100g average), it is very potential triterpene which acts against many diseases especially cancer for this purpose a large scope in the pharmaceutical industries. Present study was first time taken up to investigate "Quantification of lupeol in selected commercial coloured cultivars of mango (Mangifera indica L.) Cultivated in Telanagana region" by High Performance Liquid Chromatography method using completely randomize design with 2 factors and 4 treatments. The results indicated that significantly highest lupeol content (47.26±12.09 µg/100g) than Vanraj (28.86±2.09 µg/100g). Among the storage days significantly highest lupeol content recorded on 12$^{th}$ day of storage (66.60±12.33µg/100g) while lowest was noticed on 4$^{th}$ day of storage (10.44±3.26 µg/100g).*

***Key words:*** *High Performance Liquid chromatography (HPLC), Coloured cultivars, Lupeol, Mango, Quantification, Triterpene*

## INTRODUCTION

The mango (*Mangifera indica* L.) is a juicy stone fruit (drupe) and is now cultivated in most frost free tropical and warmer sub tropical climates, almost half of the world's mangoes are cultivated in India alone, with the largest source being China[6] and currently ranked 5$^{th}$ in total world production among the major fruit crops[4]. It belongs to the family *Anacardiaceae* (2n=40) and originated in South East Asia[8,13]. It is the national fruit of India, Pakistan, and the Philippines, and the national tree of Bangladesh (Retrieved 16 November 2013). It is most commercially grown fruit crop of the country. It is also called as "King of fruits". Over 1000 mango varieties are available worldwide, only a few are grown on commercial scales and traded. The principal mango producing states are Uttar Pradesh, Karnataka, Andhra Pradesh, Telangana, Bihar, West Bengal and Gujarat.

**Cite this article:** Soujanya, B., Kumar, A.K., Sreedhar, M., Aparna, K. and Reddy, K.R., Quantification of Lupeol in Selected Commercial Coloured Cultivars of Mango (*Mangifera indica* L.) Cultivated in Telangana Region, *Int. J. Pure App. Biosci.* **5(4):** 2141-2146 (2017). doi: http://dx.doi.org/10.18782/2320-7051.5730

Mangoes can be considered as a good source of dietary antioxidants, such as ascorbic acid, carotenoids and phenolic compounds. Mango is naturally rich in fiber (1.6g/100g), antioxidant vitamins A (54µg/100g) and C (36.4 mg/100g) and vitamin B6 (0.119mg/100g) [9,15,18]. Mango peel pigments under study include carotenoids, such as the provitamin a compound, beta-carotene, lutein and alpha-carotene[2,4] and polyphenols, such as quercetin, kaempferol, gallic acid, caffeic acid, catechins and tannins[11,17].

Numerous phytochemicals are present in mango peel and pulp, such as the triterpene, lupeol which is under basic research for its potential biological effects[3]. There is a growing interest in natural triterpenoids, also known as phytosterols, due to their wide spectrum of biological activities. 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[14]. Lupeol content in grape, olive, mango has effects on cardiovascular diseases and inflammation[5]. Mango fruit also contains substances called triterpene and lupeol, which inhibit skin and colon cancer.

The objectives of present research work is to estimate the lupeol content in selected coloured cultivars of mango and estimate the lupeol content in coloured cultivars at 3 storage days ($4^{th}$, $8^{th}$ and $12^{th}$ day) at ambient conditions by HPLC-1260 (High Performance Liquid Chromatography) method using completely randomize design with 2 factors and 4 treatments.

## MATERIAL AND METHODS

Coloured cultivars of mango (Suvarnarekha and Vanraj) collected from the Fruit Research Station, Sangareddy, India harvested at fully matured stage and subjected to ethylene treatment kept for storage at ambient conditions up to 12 days.

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. Two coloured 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. Dried pulp was grounded into fine powder it was used for the estimation of lupeol. The extraction efficiency of target compound was optimized by using solvent mixture of methanol and acetonitrile (3:7 V/V). 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[16].

Estimation of lupeol content by HPLC (1260 series) technique 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/1min at MFPI-Quality Control Laboratory, PJTSAU, Rajendranagar, Hyderabad, India. Lupeol standard showed its peak at 27.5 to 28.5 min of HPLC isocratic method. Finally lupeol (µg/100g) amount was calculated on the basis of following formula given by Anyakora *et al*[1].

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

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

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). The design adopted was completely randomized design with 2 factors (storage days and varieties). All the analysis was performed in 4 replicates 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).

## RESULTS AND DISCUSSION

The data pertaing to lupeol content in dried mango pulp of 2 mango cultivars (coloured cultivars) as influenced by the storage days at ambient conditions is presented in the table 1 and fig. 1. The results indicated that among the 2 cultivars (coloured cultivars) significantly high amount of lupeol was recorded in Suvarnarekha (47.26±12.09 µg/100g) and lowest amount was recorded in Vanraj (28.86±2.09 µg/100g).

Significant differences were noted in relation to storage days ($4^{th}$, $8^{th}$ and $12^{th}$ day of storage), the highest amount of lupeol was noticed on $12^{th}$ day of storage (66.60±12.33 µg/100g) followed by $8^{th}$ day of storage (37.14±1.30 µg/100g) and lowest was noticed on $4^{th}$ day of storage (10.44±3.26 µg/100g). Lupeol content was observed maximum at fully ripened stage when stored at ambient conditions.

The interaction between 2 cultivars (coloured cultivars) and 3 storage days ($4^{th}$, $8^{th}$ and $12^{th}$ day of storage) showed significant variation (table 1). Significantly highest amount of luepol was recorded in Suvarnarekha (99.38±0.10 µg/100g) on $12^{th}$ day of storage followed by same cultivar on $8^{th}$ day of storage (40.59±0.01µg/100g) and lowest was also recorded in Suvarnarekha (1.82±0.05µg/100g) on $4^{th}$ day of storage. All chromatograms are presented in fig. 2 to 4 showing the lupeol content in samples of few mango cultivars at ambient conditions. The results are in aggrement with Jyotshna *et al.*[7] estimated mangiferin and lupeol content in 4 mango cultivars and reported that highest amount of lupeol was found in Dashehari (1082 µg/100g) in peel and pulp during storage period.Lupeol content was varying among the cultivars and storage days. Similar study was reported by Saratha *et al*[16].

**Table 1: Lupeol content in dried mango powder (µg/100g) as influenced by storage days ($4^{th}$, $8^{th}$ and $12^{th}$ day) at ambient conditions in 2 coloured mango cultivars**

| **Varieties** | **Storage days** | | | |
|---|---|---|---|---|
| | **$4^{th}$ day** | **$8^{th}$ day** | **$12^{th}$ day** | **Mean** |
| Vanraj | 19.06±0.01 | 33.70±0.06 | 33.83±0.03 | 28.86±2.09 |
| Suvarnarekha | 1.82±0.05 | 40.59±0.01 | 99.38±0.10 | 47.26±12.09 |
| | 10.44±3.26 | 37.14±1.30 | 66.60±12.33 | |
| **Factors** | **SEm±** | | **CD at 5%** | |
| Varieties (A) | 0.047 | | 0.140 | |
| Storage days (B) | 0.058 | | 0.172 | |
| A x B | 0.082 | | 0.243 | |

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

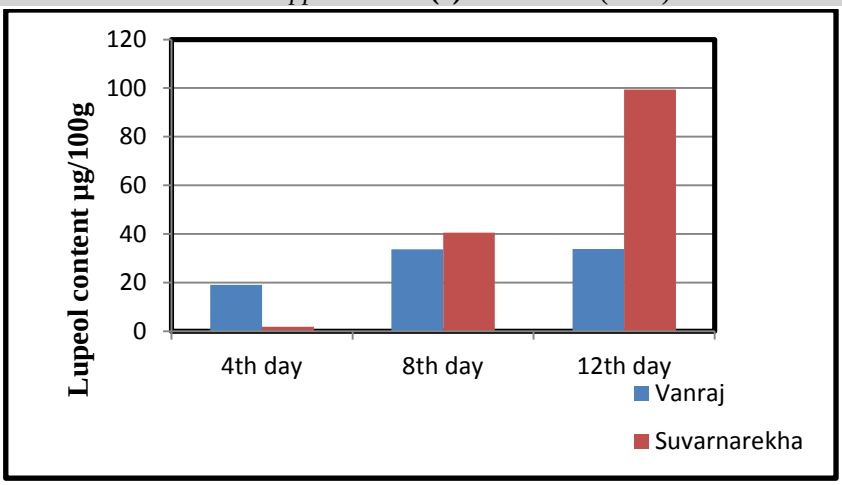
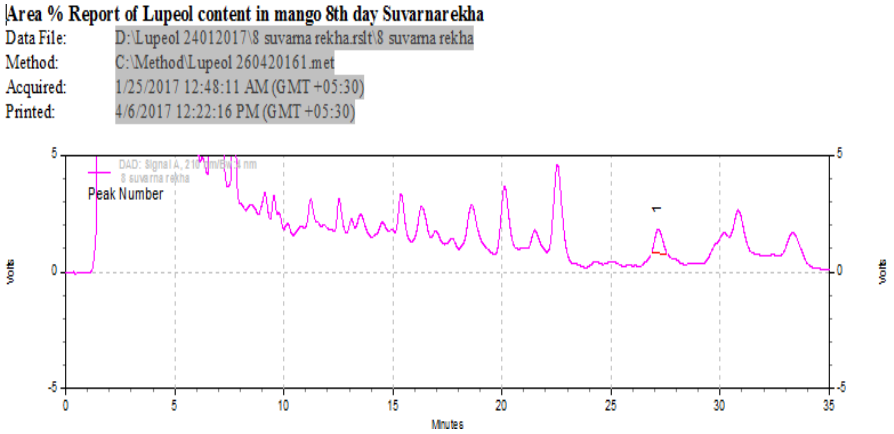
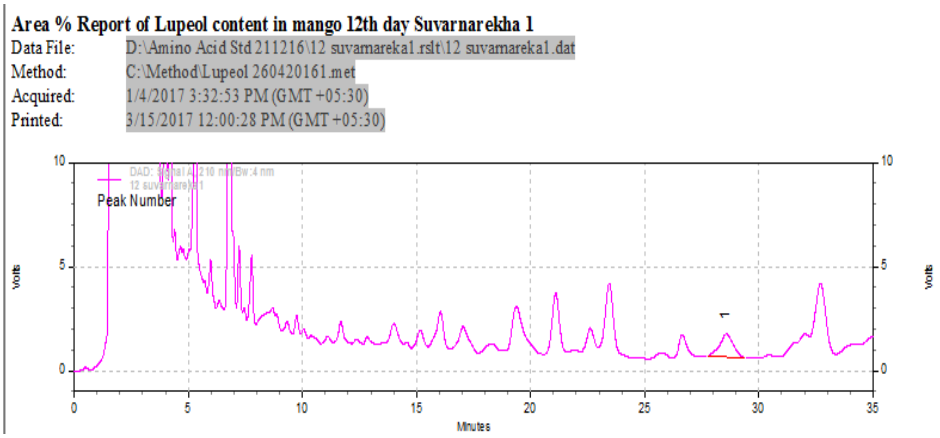



**Fig. 1: Lupeol content (µg/100g) in dried mango powder as influenced by storage days (4th, 8th and 12th day) at ambient conditions in 3 mango coloured cultivars**



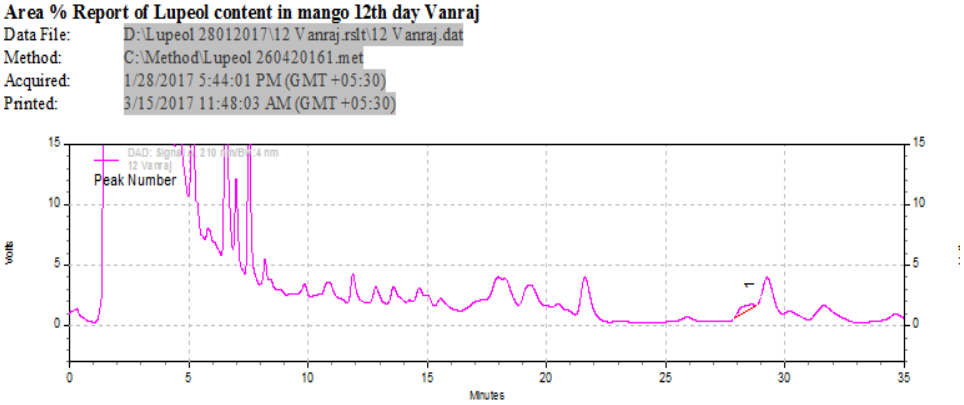

| Name | Retention time | Area | Area % |
|---|---|---|---|
| 8th day Suvarnarekha | 27.16 | 49456 | 100.00 |

**Fig. 2: Chromatogram of lupeol in variety of mango Suvarnarekha on 8th day of storage obtained by HPLC Agilent 1260**




| Name | Retention time | Area | Area % |
|---|---|---|---|
| 12th day Suvarnarekha | 28.57 | 60501 | 100.00 |

**Fig. 3: Chromatogram of lupeol in variety of mango Suvarnarekha on 12th day obtained by HPLC Agilent 1260**




| Name | Retention time | Area | Area % |
|---|---|---|---|
| 12th day Vanraj | 28.15 | 41431 | 100.00 |

**Fig. 4: Chromatogram of lupeol in variety of mango Vanraj on 12th day obtained by HPLC Agilent 1260**

## CONCLUSION

Mango varieties (2 commercial/popular culitvars) were studied for lupeol content, positive results have been noticed regarding the lupeol content in the selected mango cultivars (coloured cultivars), however there was lot of variation among the culitvars and lupeol content ranged from 47.26±12.09 µg/100g (Suvarnarekha) to 28.86±2.09 µg/100g (Vanraj). Among the storage days it varied from 66.60±12.33 µg/100g to 10.44±3.26 µg/100g (12th day and 4th day respectively). There is a huge scope on lupeol quantification from available sources of plants which can be helpful for the preparation of many pharmaceuticals and fight against different diseases especially against cancer. There is a huge availability of mango varieties and it is essential to quantify useful triterpenes (lupeol) from all available cultivars of mango.

## 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.

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