



P-ISSN: 2349-8528
E-ISSN: 2321-4902
IJCS 2019; SP6: 197-203

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(Special Issue -6)
3rd National Conference
On

**PROMOTING & REINVIGORATING AGRI-HORTI,
TECHNOLOGICAL INNOVATIONS
[PRAGATI-2019]
(14-15 December, 2019)**

**Evaluation of chemo types diversity among
Mucuna pruriens germplasm based on L-Dopa
content of seeds and L-Dopa yield/ha**

Isha Thakur and Jai Kumar

Abstract

To screen out chemo types diversity in *Mucuna pruriens* germplasm, a systematic research trial was conducted at AICRP (M&APs) farm, Birsa Agricultural University, Ranchi. Seeds of different *Mucuna pruriens* germplasm collected from three different places were grown in the experimental field under uniform cultural conditions in Randomized Block Design with twenty four treatments and four replications. Traits like L-Dopa content (%) of seeds and L-Dopa yield/ha were recorded for different *Mucuna pruriens* germplasm. L-Dopa content in seeds was recorded maximum in germplasm IIHR MP₃ (4.44%) followed by DMAPR MP₆ (4.28%) and DMAPR MP₃ (3.98%) while minimum value of it was recorded for Ranchi MP₁ (2.83%). L-Dopa yield per ha was recorded maximum in germplasm IIHR MP₁ (221.91Kg) followed by IIHR MP₇ (193.84Kg) and DMAPR MP₅ (180.61Kg).

Keywords: *Mucuna pruriens*, germplasm, traits, L-Dopa

Introduction

Evaluation is beneficial to reveal potentially useful variability for further use in genetic enhancement of crops. Through germplasm evaluation, estimation of yield based on pattern of response of genotypes, agronomic treatments across environments and reliable guidance for selecting the best genotypes may be done. Until a collection has been properly evaluated and its attributes become known to breeders, it has little practical use. The germplasm exploration and collection have resulted in accumulation of enormous genetic diversity of crop plants in the gene banks. Concerted efforts need to be made for its characterization, evaluation and identification of trait specific accessions especially from unexplored/exotic germplasm using field Phenotyping coupled with modern genomic tools to trace the underlying gene. Therefore, to find out the chemo types diversity in *Mucuna pruriens* germplasm on the basis of quantitative traits, a systematic research trial was undertaken on it at Birsa Agricultural University, Ranchi.

Genus *Mucuna* belongs to the family Fabaceae, sub family Papilionaceae includes approximately 150 species of annual and perennial legumes. It is widespread in tropical and sub-tropical regions of the world. Its Hindi name is Kewach and English name is Velvet bean. L-Dopa (3, 4-hydroxyphenylalanine) is a basic non protein, toxic amino acid present in the seeds of *Mucuna*. High amount of L-Dopa in *Mucuna* seeds is major impediment to exploit it as food or feed. Although L-Dopa is pharmacologically active ingredient and the drug which is used to treat the Parkinson's disease in human beings.

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Material and Methods

Seeds of different *Mucuna pruriens* germplasm collected from 3 different places were grown in the experimental field under same uniform cultural conditions in Randomized Block Design with twenty four treatments and four replications at 60cm × 60cm spacing level. Area under each treatment was 3.6m × 3m=10.8m² with twelve plants per plot. Coarse powder of the dried seeds of *Mucuna pruriens* was extracted with methanol using a soxhlet apparatus. The methanol extract thus obtained was dried under reduced pressure at a temperature not exceeding 40 °C. About 350 µg of the extract was diluted to 25 ml with 0.1 M hydrochloric acid. Further, 5 ml of this solution was diluted to 100 ml with 0.1 M hydrochloric acid and sonicated for about 60 min. The solution was then filtered and used for estimation. The following chromatographic conditions were used to quantify the L-dopa. Stationary phase: Eurosphare, C18, 250×4.0mm, column oven temperature: 30 °C, mobile phase: methanol: 0.5% v/v of acetic acid (70:30, v/v), detection wavelength: 284 nm, flow rate: 1.2 ml/min, injection volume: 20 µl. The percentage of L-Dopa present in methanolic extract of

Mucuna pruriens was calculated by comparison of the areas measured for the standard and sample solution [7]. L-Dopa yield/ha: It was calculated by the formula- Seed yield/ha×% of L-Dopa in seeds.

Results and Discussion

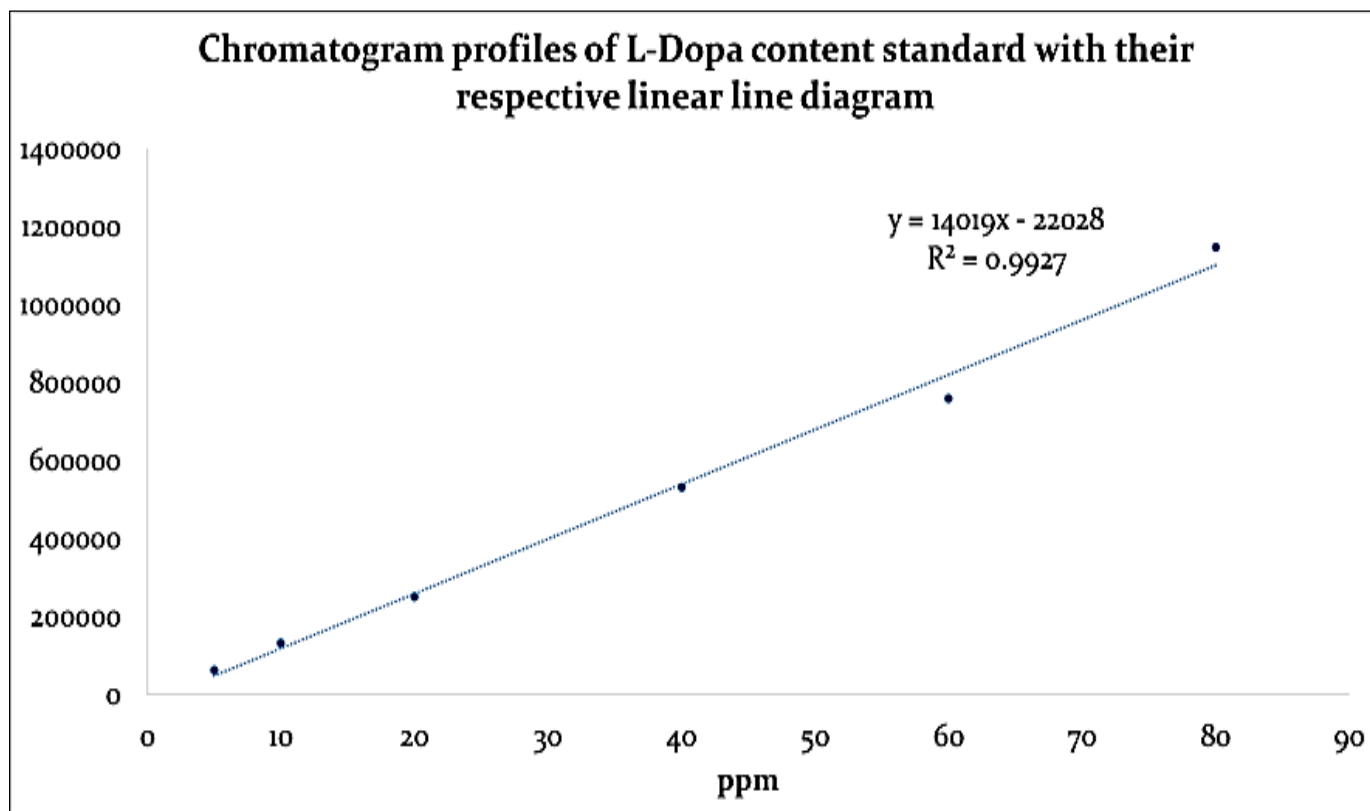
Highly significant differences were recorded in L-Dopa content in seeds (%) and maximum L-Dopa content in seeds (%) was recorded in IIHR MP3 (4.44%). Minimum L-Dopa content in seeds (%) was recorded in Ranchi MP1 (2.83%). The descending order of L-Dopa content in seeds (%) was as followed: IIHR MP₃ (4.44)>DMAPR MP₆ (4.28)>DMAPR MP₃ (3.98)>IIHR MP₁ (3.94)>IIHR MP₂ (3.77)>DMAPR MP₅ (3.72)>Ranchi MP₅ (3.68)>IIHR MP₈ (3.56)>DMAPR MP₈ (3.54)>IIHR MP₇ (3.48)>DMAPR MP₄ (3.43)>Ranchi MP₃ (3.40)>Ranchi MP₆ (3.35)>Ranchi MP₈ (3.32)>IIHR MP₄ (3.26)>DMAPR MP₇ (3.25)=Ranchi MP₂ (3.25)>DMAPR MP₂ (3.23) = Ranchi MP₄ (3.23)>IIHR MP₅ (3.06)>IIHR MP₆ (2.99)>Ranchi MP₇ (2.98)>DMAPR MP₁ (2.93)>Ranchi MP₁ (2.83).

Table 1: L-Dopa content in seeds (%) and L-Dopa yield/ha (Kg) in *Mucuna pruriens* germplasm

Treatments	Particulars	L-Dopa content in seeds (%)	L-Dopa yield/ha (Kg)
T ₁	IIHR MP ₁	3.94 ^{bc}	221.91 ^a
T ₂	IIHR MP ₂	3.77 ^c	175.47 ^{bc}
T ₃	IIHR MP ₃	4.44 ^a	7.61 ^g
T ₄	IIHR MP ₄	3.26 ^{ef}	115.98 ^{de}
T ₅	IIHR MP ₅	3.06 ^f	142.06 ^{cd}
T ₆	IIHR MP ₆	2.99 ^{fg}	104.11 ^{de}
T ₇	IIHR MP ₇	3.48 ^{de}	193.84 ^{ab}
T ₈	IIHR MP ₈	3.56 ^d	168.59 ^{bc}
T ₉	DMAPR MP ₁	2.93 ^h	93.48 ^{de}
T ₁₀	DMAPR MP ₂	3.23 ^{ef}	13.50 ^f
T ₁₁	DMAPR MP ₃	3.98 ^b	168.60 ^{bc}
T ₁₂	DMAPR MP ₄	3.43 ^{de}	115.96 ^{de}
T ₁₃	DMAPR MP ₅	3.72 ^{cd}	180.61 ^b
T ₁₄	DMAPR MP ₆	4.28 ^{ab}	118.45 ^{cd}
T ₁₅	DMAPR MP ₇	3.25 ^{ef}	15.07 ^f
T ₁₆	DMAPR MP ₈	3.54 ^{de}	125.06 ^{cd}
T ₁₇	Ranchi MP ₁	2.83 ^h	107.35 ^{de}
T ₁₈	Ranchi MP ₂	3.25 ^{ef}	111.97 ^{de}
T ₁₉	Ranchi MP ₃	3.40 ^{de}	116.30 ^d
T ₂₀	Ranchi MP ₄	3.23 ^{ef}	99.16 ^{de}
T ₂₁	Ranchi MP ₅	3.68 ^{cd}	146.95 ^c
T ₂₂	Ranchi MP ₆	3.35 ^e	30.83 ^e
T ₂₃	Ranchi MP ₇	2.98 ^g	142.62 ^{cd}
T ₂₄	Ranchi MP ₈	3.32 ^{ef}	132.36 ^{cd}
Grand mean		3.45	118.67
S.E. (m)		0.068	11.08
C.D. 5%		0.193	31.32
C.D. 1%		0.251	40.99
C.V. (%)		4.019	18.67

High variability was observed in L-Dopa yield/ha (Kg) among different *Mucuna pruriens* germplasm. Maximum L-Dopa yield/ha was recorded in germplasm IIHR MP₁ (221.91Kg) which was *at par* with IIHR MP₇ (193.84Kg). Minimum L-Dopa yield/ha was recorded in germplasm IIHR MP₃ (7.61Kg). Mohan and Janardhanan (1995) reported L-DOPA

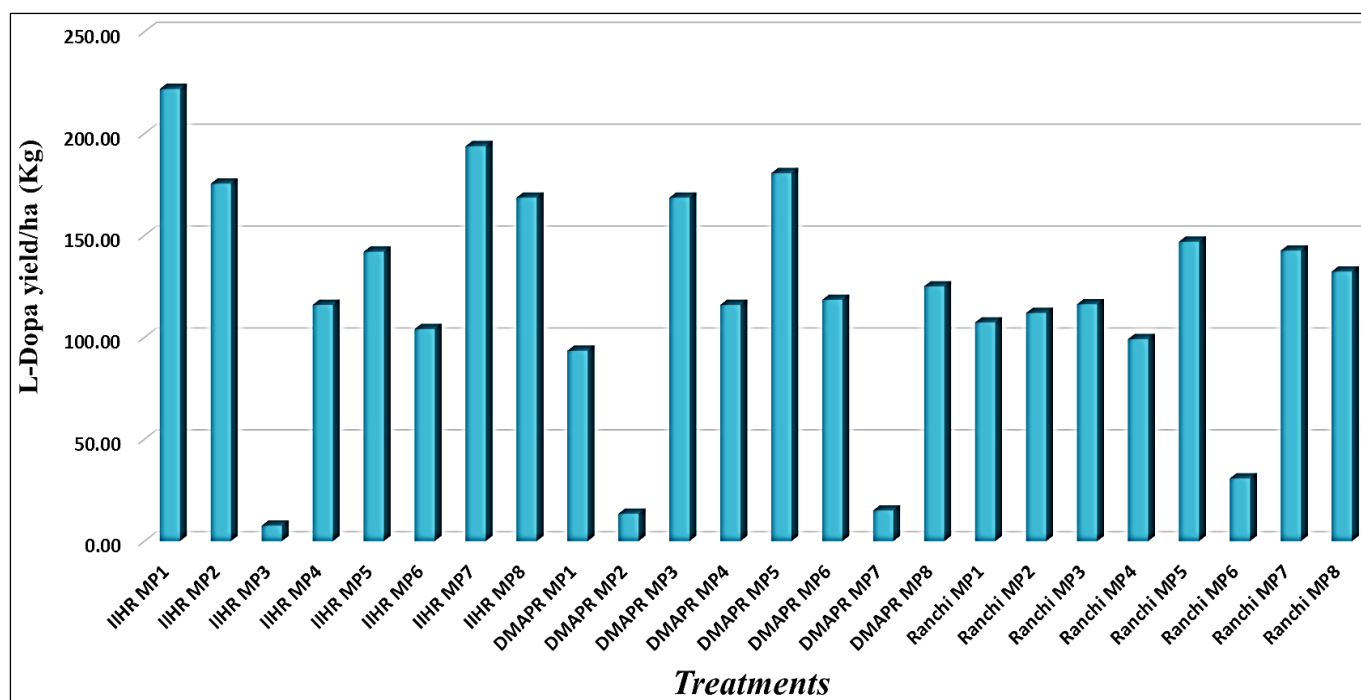
content was 6.86% in black seed coat and 6.08% in white seed coat of *Mucuna utilis* whereas lowest 4.24% in *Mucuna monosperma* [4]. Velvet Bean grown in wild has L-DOPA content 4.96% but the presence of trichomes on the pods make harvesting difficult [1]. The dehulled seeds contain more L-Dopa content (4-5%) than the wholeseeds (3.6-4.7%).



Graph 1: Regression line of L-Dopa Standard at different concentration level

L-Dopa varies from 3.6% to 4.2% in three accessions of *Mucuna* bean collected from India [5]. L-Dopa varies from 2.2% to 7.2% in study of 36 *Mucuna* accessions from Africa, America and India [2]. L-Dopa in six *Mucuna* species was highest (6.5%) in *Mucuna pruriens* and lowest (4.0%) in *Mucuna cochinchinensis* [8]. L-Dopa content varied from 6.61 per cent in IIHR MP₀₂ to 4.11 per cent in IIHR MP₁₇ in

evaluation of seed samples of 13 genotypes of *Mucuna utilis* [3]. L-Dopa was lowest 3.29% in accession IC385841 collected from Jharkhand and highest 5.44% in accession IC551549 collected from Chhattisgarh in the study of 38 germplasm collected from different geographical locations of India [6].



Graph 2: L-Dopa yield/ha (Kg) of different *Mucuna pruriens* germplasm

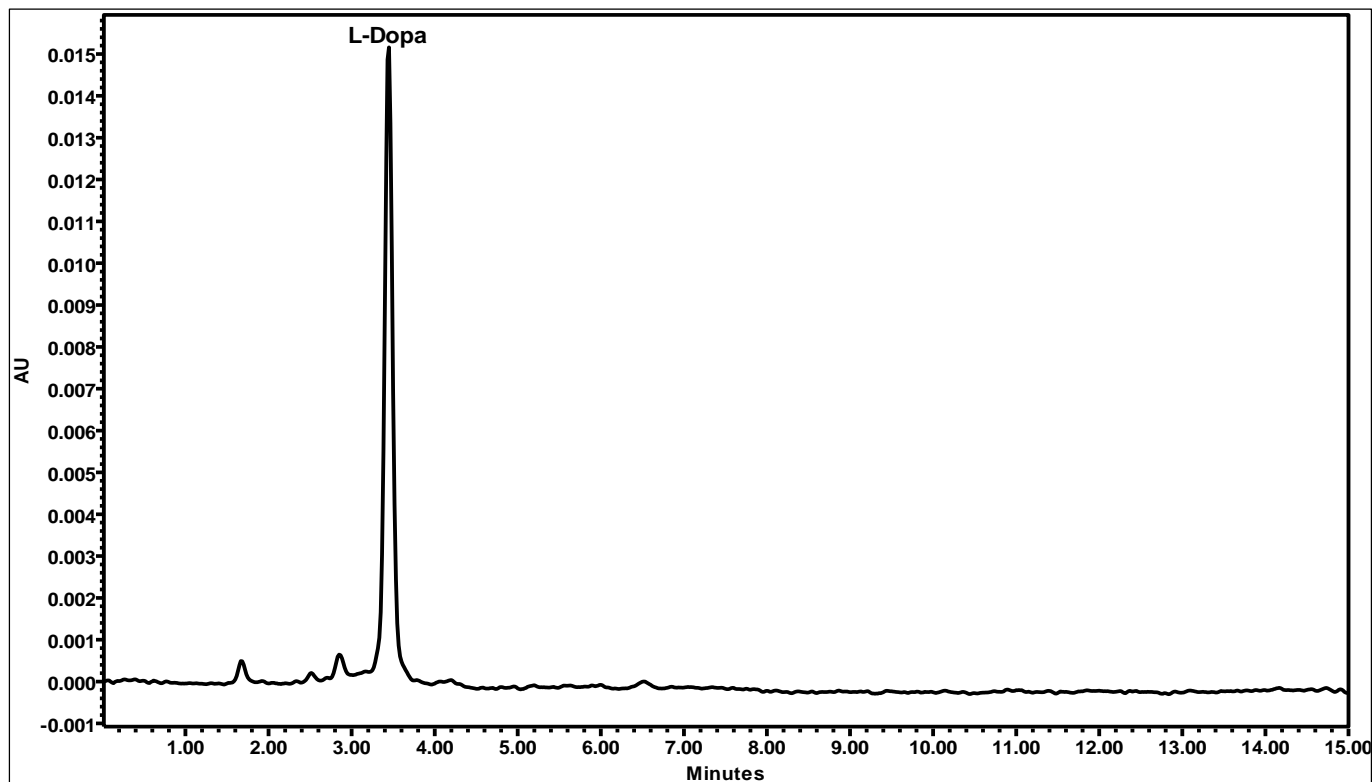


Fig 1: Chromatogram generated by HPLC of IIHR MP₃

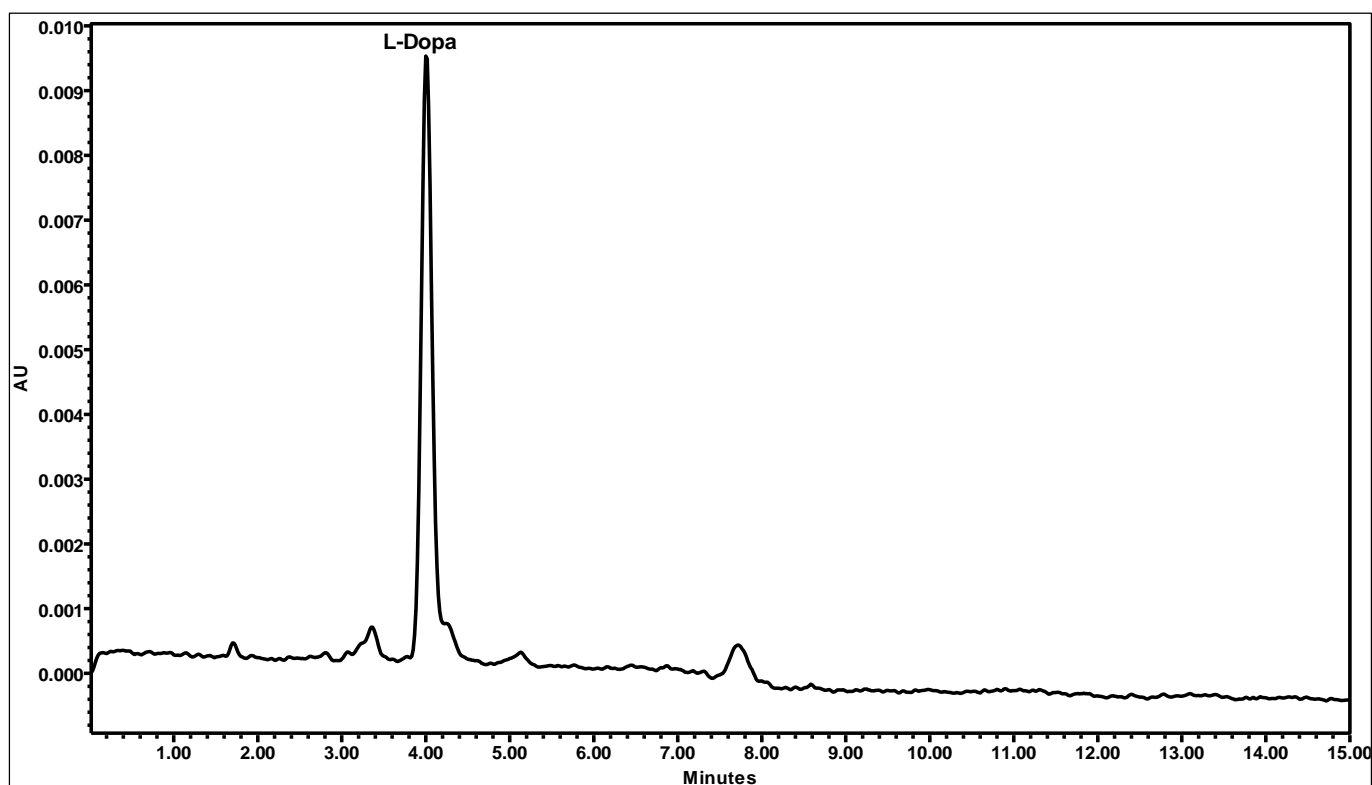


Fig 2: Chromatogram generated by HPLC of IIHR MP₆

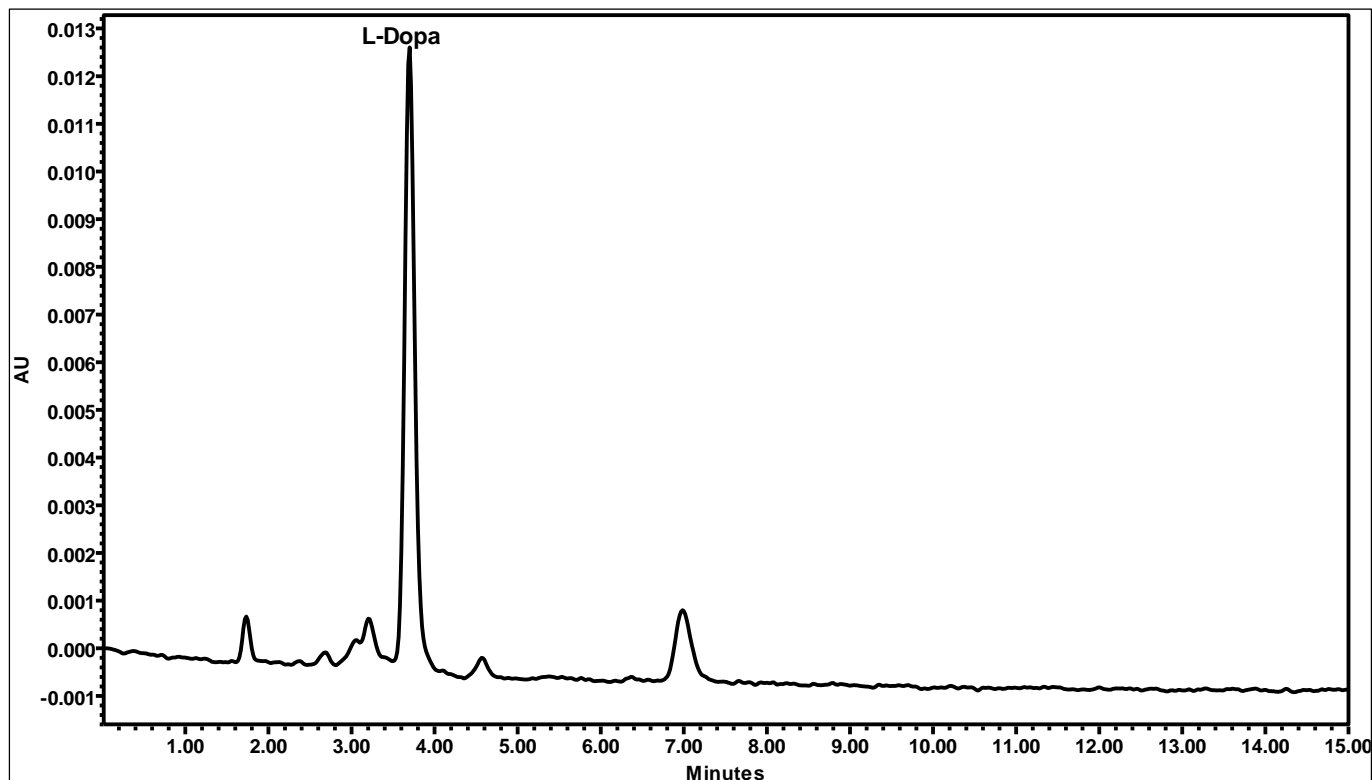


Fig 3: Chromatogram generated by HPLC of DMAPR MP₆

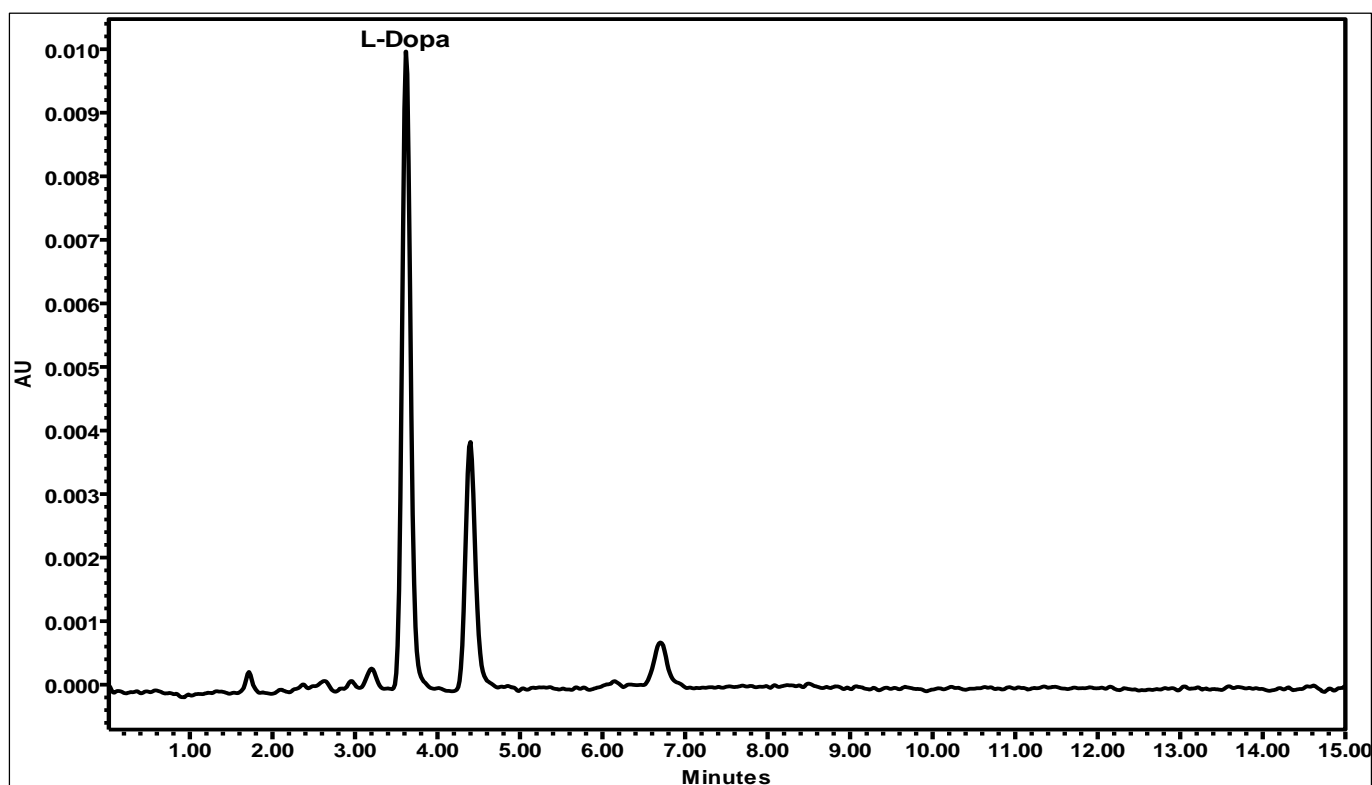


Fig 4: Chromatogram generated by HPLC of DMAPR MP₁

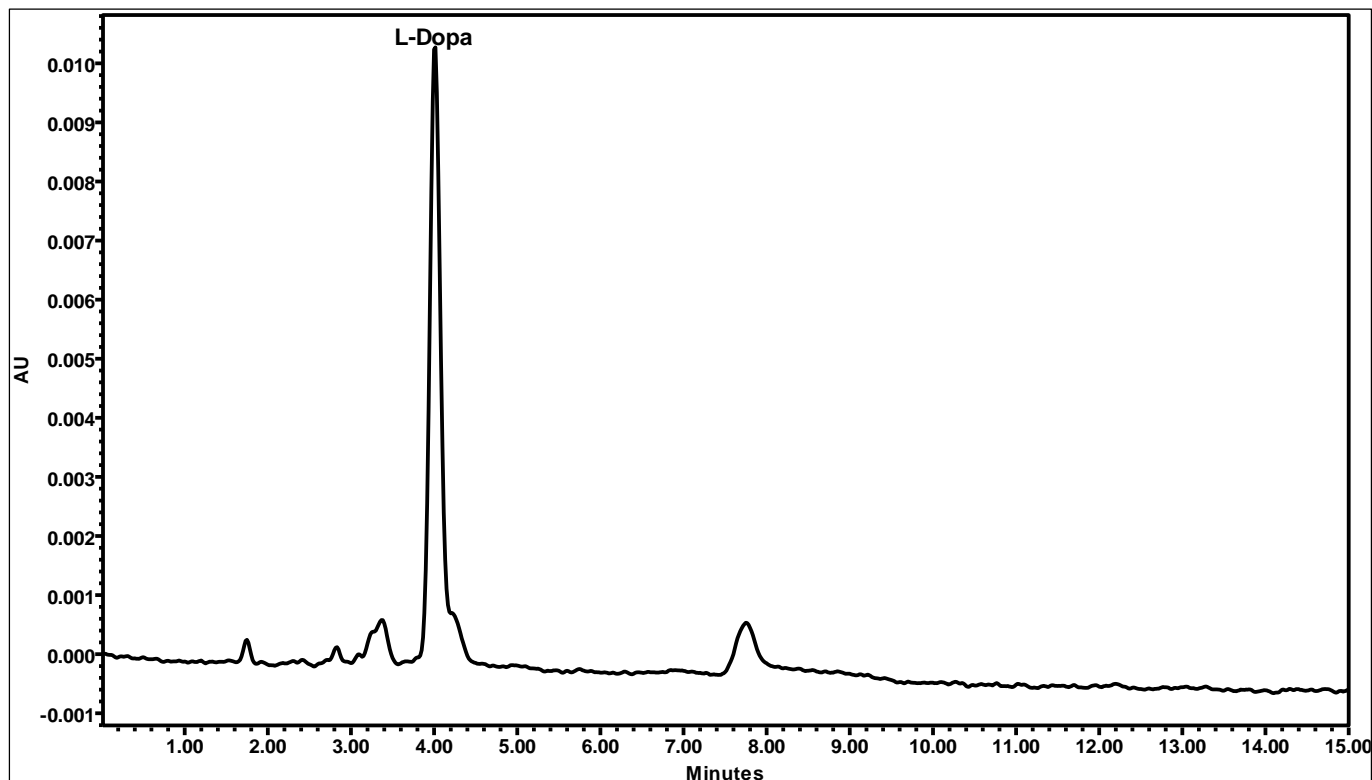


Fig 5: Chromatogram generated by HPLC of Rancho MP₅

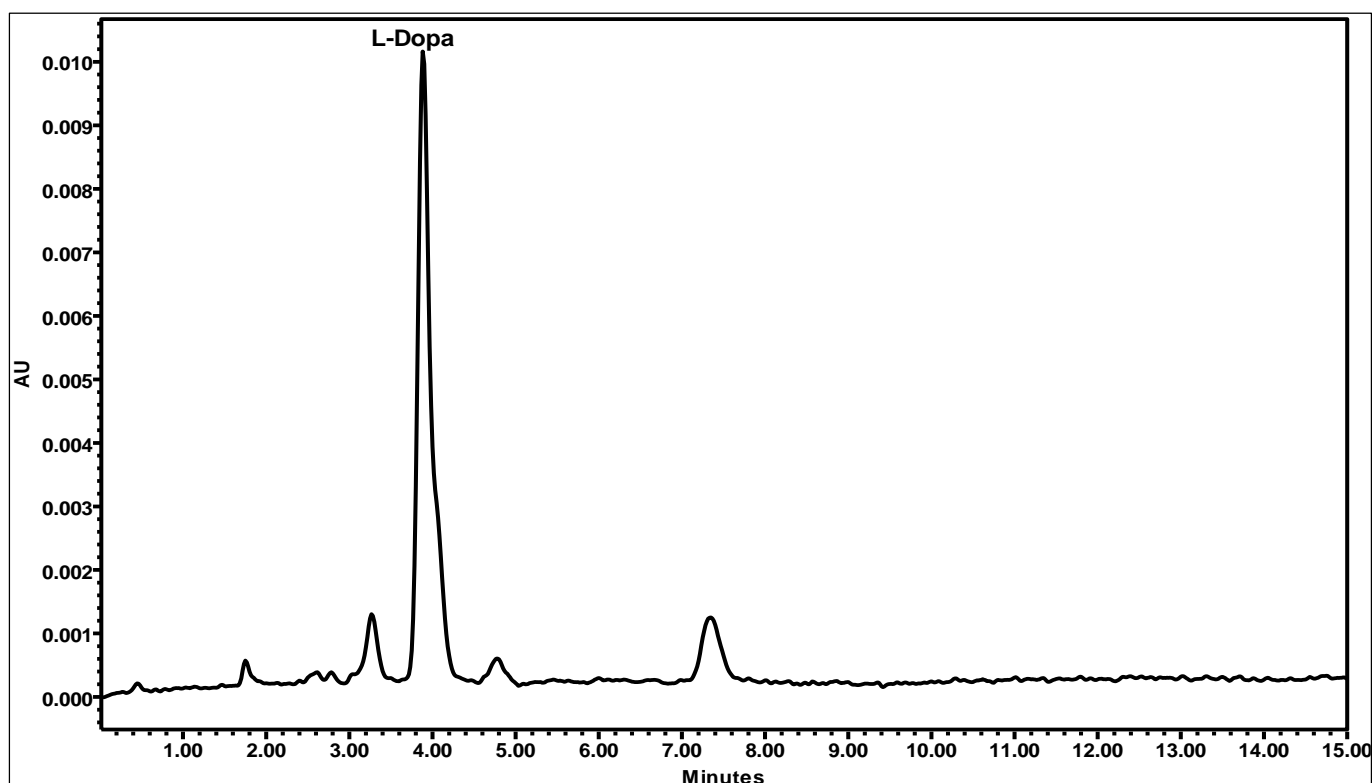


Fig 6: Chromatogram generated by HPLC of Rancho MP₁

Conclusion

L-Dopa content in seeds was recorded maximum in germplasm IIHR MP₃ (4.44%) followed by DMAPR MP₆ (4.28%) and DMAPR MP₃ (3.98%) while minimum was recorded in Rancho MP₁ (2.83%). L-Dopa yield per ha was recorded maximum in germplasm IIHR MP₁ (221.91Kg) followed by IIHR MP₇ (193.84Kg) and DMAPR MP₅ (180.61Kg) while minimum was recorded in IIHR MP₃ (7.61Kg). On the basis of L-Dopa content in seeds (%); three germplasm namely IIHR MP₃, DMAPR MP₆ & DMAPR MP₃

may be selected as superior germplasm because they produced maximum 4.44, 4.28 & 3.98% of L-Dopa content in seeds (%) which was significantly superior to rest of the germplasm.

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