Determination of New Isomer of Palmitoleic Acid in Mucuna Pruriens Seed Oil

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Abstract
Palmitoleic acid is an unsaturated fatty acid and it is very important in different essential enoic acid. Most of which are not synthesized by the body and are to be supplied through edibles. By the formation of DMOX derivative of FAME, allows the determination of deposition of the double bond in fatty acid carbon chain. This work is also important to quick and reproducible methods used to screen the utility of non-conventional sources of oil for edible and industrial purposes.

For the present investigation, non-conventional source mucuna pruriens (papilionaceae) seed is used. Seeds have been reported to cure diarrhea and Parkinson’s disease. It is a natural source of L-dopa.

Keywords: FAME, DMOX derivative, palmitoleic acid

1. Introduction
Unsaturated fatty acid content has been given emphasis keeping in view the importance of essential enoic acids most of which are not synthesized by the body and are to be supplied through edibles. The work lay also importance to quick and reproducible methods used to screen the utility of non-conventional oils for edible/industrial purposes. The crude materials have been collected from the different nearby locations of the state.

Mucuna pruriens (Papilionaceae) is grown in tropics. It has flattened pods with three to six seeds in each. Seeds have been reported to cure diarrhea. Recently, the seed powder has been found effective against Parkinson’s disease. As a natural source of L-dopa, the seed formulation may possess advantage in the long term. Food perspectives of this underutilized legume has also been recently taken up.

2. Experimental
The seeds have been collected from M.P./U.P. border town of Mahoba and authenticated at the Forest Department. The oil was obtained by Soxhlet extraction of the coarsely ground seeds with hexane. The solvent was later removed by distillation.

The fatty acids were separated from the oil by refluxing with methanolic KOH at room temperature for three hours. The solvent was removed and after washing with water, the upper layer containing fatty acids was esterified using methanol/H₂SO₄, refluxing the contents for two hours. After removal of solvent, the sample was taken in hexane (fatty acid methyl ester – FAME) and injected into GC (Varian Vista 6000) with FID using fused silica column coated with CP-Sil 88 (50m × 0.25mm) (Chrompack) with H₂ as carrier gas with oven temperature - 70°C 2’, 30°C/min. to 135°C (1’), 3°C/min. to 180°C, 15°C/min. to 250°C. Spectra Physics integrator was used for data acquisition.

For location of double bond, GC – MS analysis of dimethyl oxazoline (DMOX) derivative was carried. For preparation of DMOX derivative – FAME was reacted with 2-amino-2-methyl propanol at 180°C for four hours, followed by bringing the contents to room temperature and extraction with methylene chloride. Finnigan MAT 8430 mass spectrometer connected to HP 5890 Gas Chromatograph equipped with 30m × 0.25mm capillary column (DBWAX) using He as carrier gas, on column injector, oven at 50°C for 2’, 4°C/min. up to 240°C, held for 5’. The mass spectra were acquired at 70 eV.

3. Results and Discussion
3.1 Figure 1 GLC profile of FAME (M. pruriens)
Fatty oil was obtained from the seeds in an yield of 25%. Fig. 1 shows the GC profile of the FAME. The identified acids as esters are: Peak 1 Palmitoleic (16:1), Peak 2 Oleic (18:1), Peak 3 Gadoleic (20:1, Eicosenoic) and Peak 4 Cetoleic (22:1, Docosenoic).

3.2 Figure 2 GC-MS profile of DMOX derivative of FAME (M. pruriens)
The GC/MS profile Fig. 2 shows molecular ion at m/z 307. The point of unsaturation is indicated in the MS by interruption of 14 mass spaced homologous ion series. A mass separation of 13 Da (amu) instead of 14
between two neighbouring homologous fragments, containing n-1 and n carbon atoms of the original fatty acid moiety, indicates a double bond between carbon n and n+1. The double bond located between carbons n and n-1 (ions at m/z 237 and 224) also gives abundant ions with n+2 and n-2 carbon atoms (ions at m/z 264 and 210, respectively).

4. Conclusion

The DMOX derivative thus allows determination of the position of the double bond in the fatty acid carbon chain. According to the above fragmentation pattern, the double bond is located in the 11th C (16:1 ∆11). This is a new isomer of 16:1 different from Palmitoleic which is 16:1, 9 enoic acid. It can be represented as:

![Fatty Acid Structure](image)

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References:

Fig. 1: GLC profile of FAME (M. pruriens)
Fig. 2: GC-MS profile of DMOX derivative of FAME (M. pruriens)
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