GC-MS ANALYSIS OF BIOACTIVE COMPOUNDS OF *MORINDA TINCTORIA* FRUIT

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ABSTRACT

The aim of the study was carried out for identification of bioactive compounds from the ethanolic extract of *Morinda Tinctoria* fruits by Gas chromatography. GC-MS is the technique to identify the compounds by comparison of mass spectra obtained from the sample with that taken from available standards values. 8 compounds were identified and they are reported as of Bicyclo [4.1.0] heptane, 7-pentynyl (10.095%), 1-Hexyl-2-nitrocyclohexane (29.189%), 1-Pentanol, 4-amino (1.140%), 2,2,3,3,4,4-Hexamethyltetrahydrofuran (1.576%), Cyclohexyl methyl formate (19.325%), (S)-(+)-2-Amino-3-methyl-1-butanol (8.400%), 2-Propanol, 1-methoxy (3.523%), 3-Decanynoic acid (25.506%) and o-Methylisourea hydrogen sulfate (1.246%) .

Key words: *Morinda Tinctoria,* GC-MS analysis and Anti-oxidant.

INTRODUCTION

Plants are the rich sources of the traditional medicines in several countries and they are used several types of medicinal systems such as, unani, siddha and ayurveda. The medicinal plants are used as healing as well as curing of several human diseases because of presence of phytochemical constituents. Phytochemical constituents present naturally in plants, leaves and roots. (1) Plants produce bioactive molecules are compounds having pharmacological and toxicological effects. Bioactive compounds are nothing but plant secondary metabolites. To detect Bioactive compounds in plant must perform extraction methods or separating techniques and recovering technique's taking measurement. (2) The modern methods describing identification and quantification of active constituents in plant material may useful for standardization of herbal and its formulations. GC-MS is the technique to identify bioactive constituents of alcohols, acids, esters, ethers, long chain fatty acids etc. (3)

Morinda Tinctoria Roxb belonging to the family Rubiaceae that grows wild and distributes thrroughtout southeast asia. It is commercially known as Nunna and is indigenous to tropical countries. M. tinctoria is considered as a folk medicine. In the traditional system of medicine, leaves and roots are used as an astringent, anti-inflammatory, analgesic and anti-diabetic. It is also used in deobstrent and to relieve pain in gout. (4) The ashes of M. tinctoria leaves are reported to to act as biosorbents in controlling ammonia pollution in waste waters. (5) The major components identified in the Nunna plant which include octoaniacis, vitamin C, terpenoids, scopoletin, flavones, glycosides, linoleic acid, morindone, alizarin and rubiadin. (6)

EXPERIMENTAL:

Collection and preparation of sample:

The fruits *of Morinda Tinctoria* were collected from the S.V.University , Tirupati, Andhra Pradesh. They are washed thoroughly with distilled water, cut into small pieces, air dried and grounded using motor and pestle.

Extraction method:

Coarsely powdered material extracted with ethanol for 48 hours and filtered through Whatman filter paper (N0. 1). The filtrate was air dried in an oven for 3 days and extract was subjected to GC-MS analysis.

GC-MS analysis:

The Clarus 680 GC was used in the analysis employed a fused silica column, packed with Elite-5MS and components are separated. The injector temperature was set at 260° C during chromatographic run. The 1 µL of extract sample injected into the instrument the oven temperature as follows: 60°C (2 min), followed by 300°C at the rate of 10°C min^{-1} and 300°C, where it has held for 6 min. The carrier gas helium at a constant flow of 1mL/min.

Identification of components:

The spectrum of compounds was compared database of spectrum of known components stored in GC-MS NIST library. The name, molecular weight, peak and structure of components of the test material ascertained.

RESULTS AND DISCUSSION:

The results obtained to GC-MS analysis led to the identification of number of compounds from GC fractions of methanolic extracts of *Morinda Tinctoria* fruits. These compounds were identified by mass spectroscopy attached with GC. The results of the present study tabulated in Table 1. Table 2 describes phytoconstituents and biological activities identified in *Morinda Tinctoria* fruit. The compound prediction is based on the GC-MS National Institute Standard and Technology Database. Table 2 listed major phytoconstituent's and biological activities obtained through GC-MS study of *Morinda Tinctoria* fruits. The results revealed presence of Bicyclo [4.1.0] heptane, 7-pentynyl (10.095%), 1-Hexyl-2-nitrocyclohexane (29.189%), 1-Pentanol, 4-amino (1.140%), 2,2,3,3,4,4-Hexamethyltetrahydrofuran (1.576%), Cyclohexyl methyl formate (19.325%), (S)-(+)-2-Amino-3-methyl-1-butanol (8.400%), 2-Propanol, 1-methoxy- (3.523%), o-Methylisourea hydrogen sulfate (1.246%). This type of analysis is the step to understand nature of medicinal fruits and type of study helpful for further studies.

Qualitative Report

File: C:\TURBOMASS\2021.PRO\Data\M-T-S2-(23ES-0701)-.raw
Acquired: 24-Aug-23 01:22:38 AM Printed: 25-Aug-23 10:29 AM
Description:
GC/MS Method: GC: METHOD-1.mth MS: METHOD-1.EXP Page 1 of 1
Sample ID: M-T-S2-(23ES-0701)- Vial Number: 16

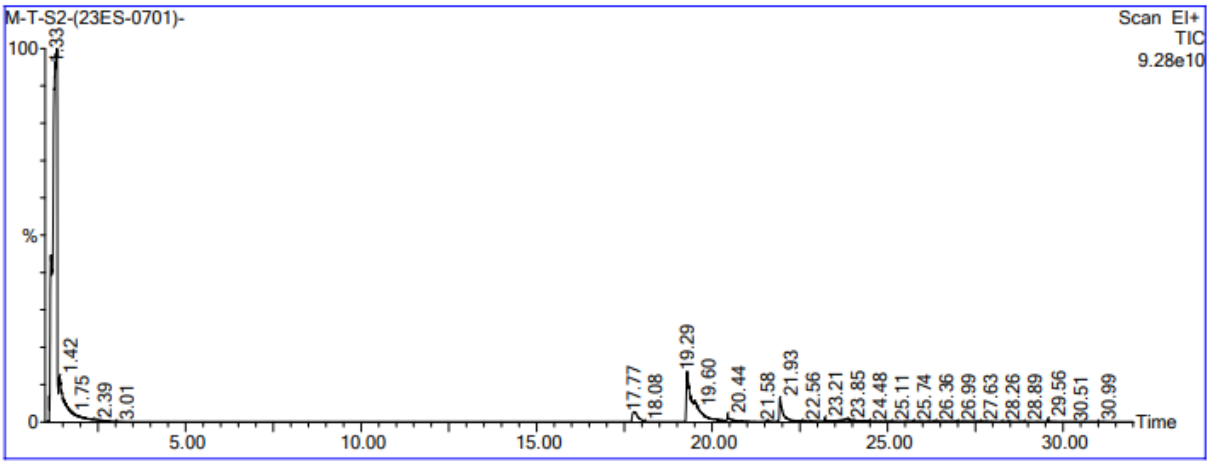

Fig 1: GC-MS chromatogram of ethanolic extract of *Morinda Tinctoria*

Table 1: Components detected in GC-MS with retention time, height, area (%), scan and norm (%).

#	RT	Scan	Height	Area	Area %	Norm %
1	1.329	66	31,465,027,584	1,321,313,152.0	25.506	87.38
2	1.394	79	4,027,804,160	64,565,292.0	1.246	4.27
3	1.419	84	4,971,852,800	182,526,960.0	3.523	12.07
4	1.469	94	2,107,089,792	59,048,380.0	1.140	3.91
5	17.765	3352	2,482,460,672	435,170,496.0	8.400	28.78
6	19.291	3657	12,517,719,040	1,512,110,080.0	29.189	100.00
7	19.516	3702	5,063,286,784	1,001,131,520.0	19.325	66.21
8	20.441	3887	1,665,140,480	81,648,240.0	1.576	5.40
9	21.937	4186	5,823,838,720	522,984,832.0	10.095	34.59

Table 2: Components detected in ethanolic extract of *MorindaTinctoria* fruit

MW: Molecular weight RT: Retention Time

S.no	Compound	Molecular formula	MW	RT	Area %
1	Bicyclo [4.1.0] heptane, 7-pentynyl	$C_{12}H_{22}$	166	21.937	10.095
2	1-Hexyl-2-nitrocyclohexane	$C_{12}H_{23}O_{2N}$	213	19.291	29.189
3	1-Pentanol, 4-amino	$C_5H_{13}ON$	103	1.469	1.140
4	2,2,3,3,4,4-Hexamethyltetrahydrofuran	$C_{10}H_{20}O$	156	20.441	1.576
5	Cyclohexyl methyl formate	$C_8H_{14}O_2$	142	19.516	19.325

6	(S)-(+)-2-Amino-3-methyl-1-butanol	C_5H_{13ON}	103	17.765	8.400
7	2-Propanol, 1-methoxy-	$C_4H_{10}O_2$	90	1.419	3.523
8	o-Methylisourea hydrogen sulfate	$C_2H_6ON_2$	74	1.394	1.246
9	3-Decanynoic acid	$C_{10}H_{16}O_2$	168	1.329	25.506

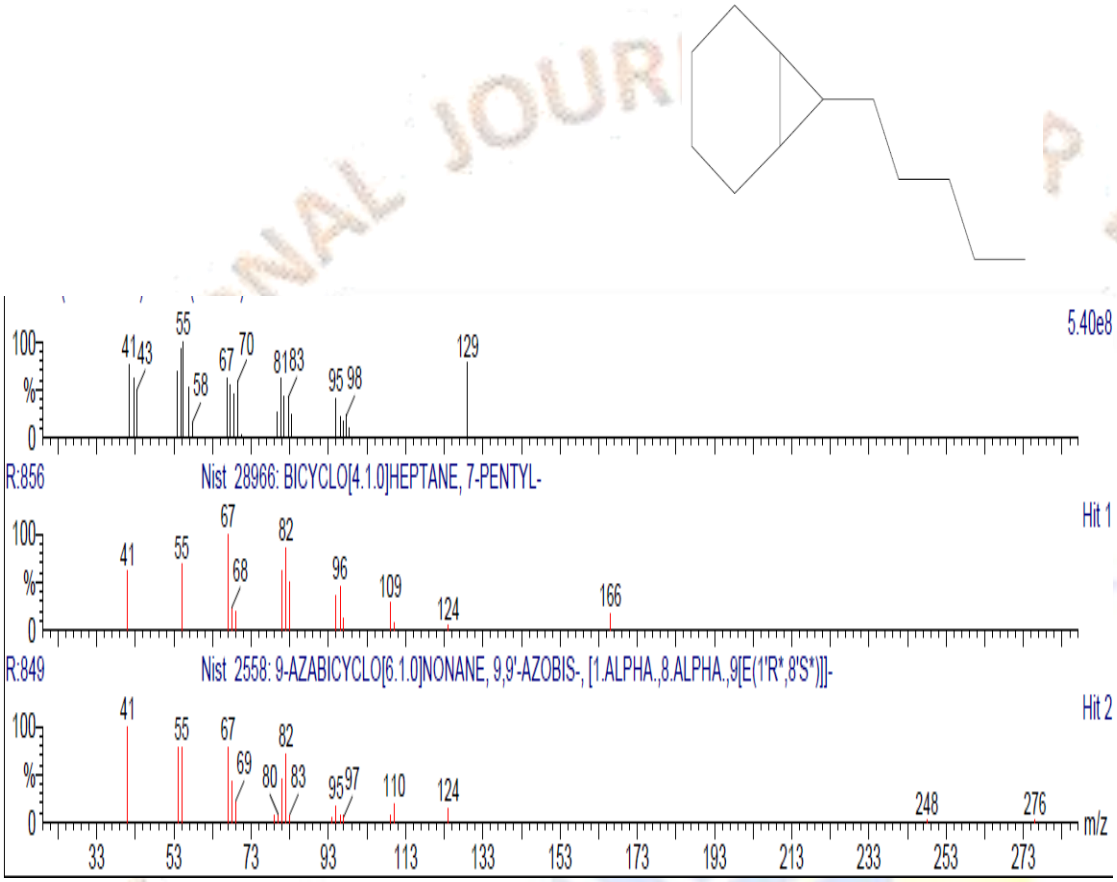

Fig 2: Mass spectrum of Bicyclo [4.1.0] heptane, 7-pentynyl. (RT: 21.937)

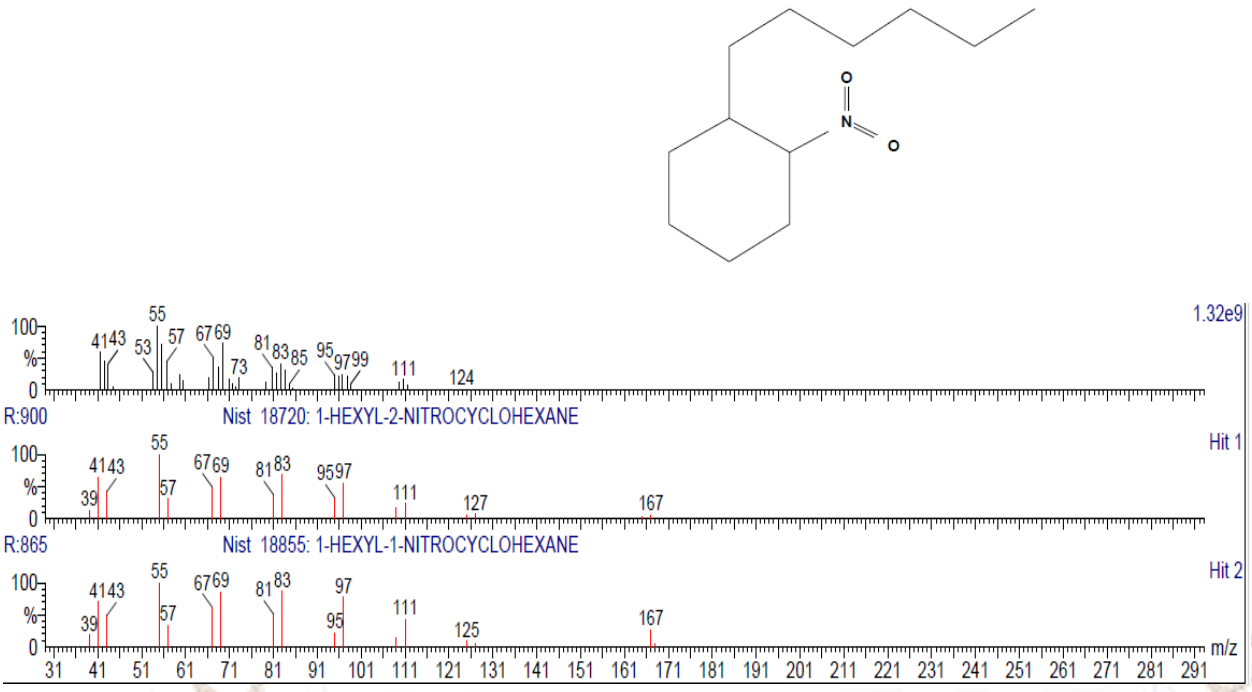

Fig 3: Mass spectrum of 1-Hexyl-2-nitrocyclohexane (RT: 19.291)

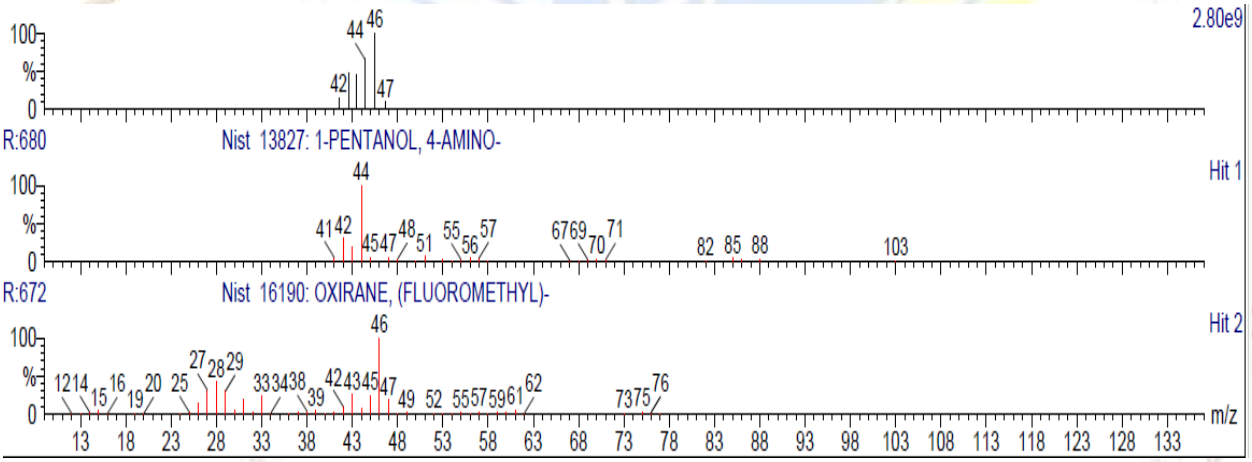

Fig 4: Mass spectrum of 1-Pentanol, 4-amino (RT: 1.469)

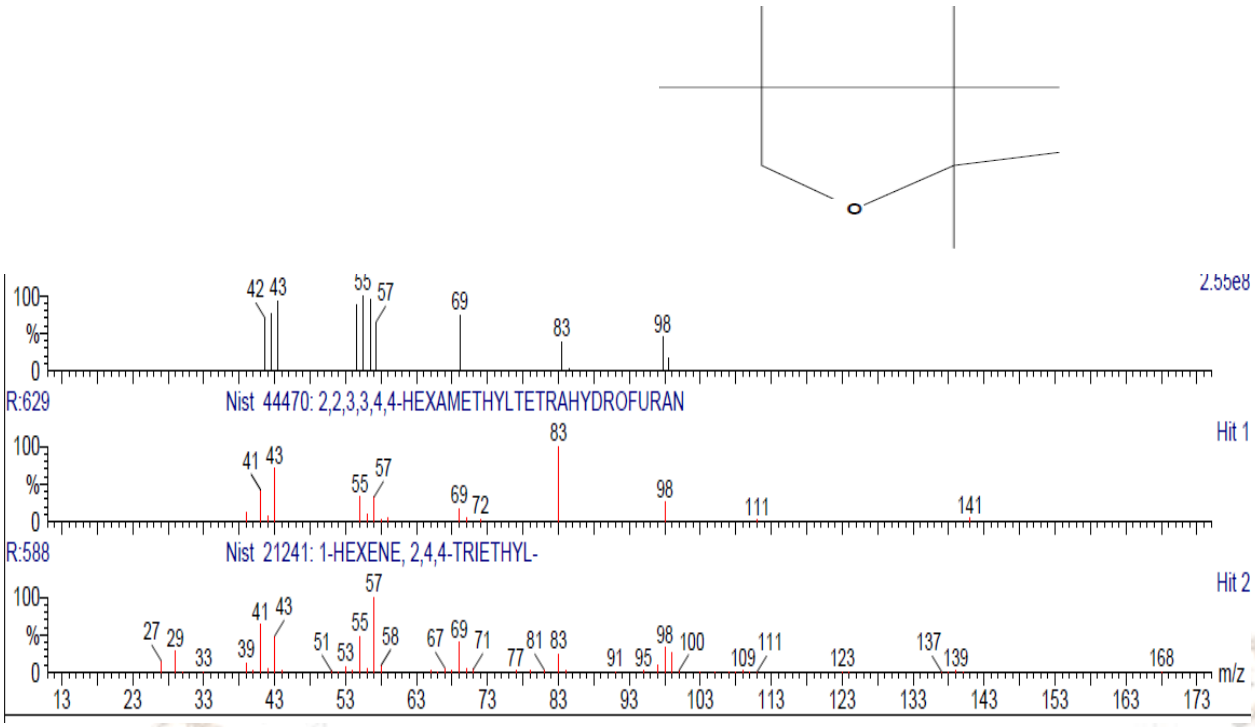

Fig 5: Mass spectrum of 2,2,3,3,4,4-Hexamethyltetrahydrofuran (RT: 20.441)

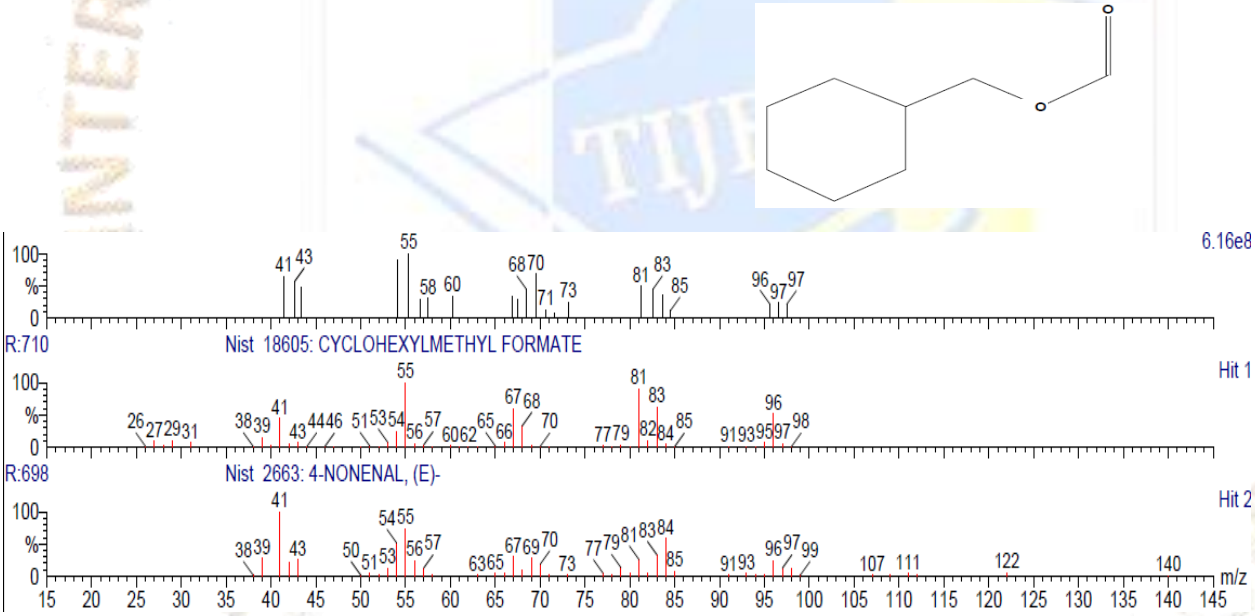

Fig 6: Mass spectrum of Cyclohexyl methyl formate (RT: 19.516)

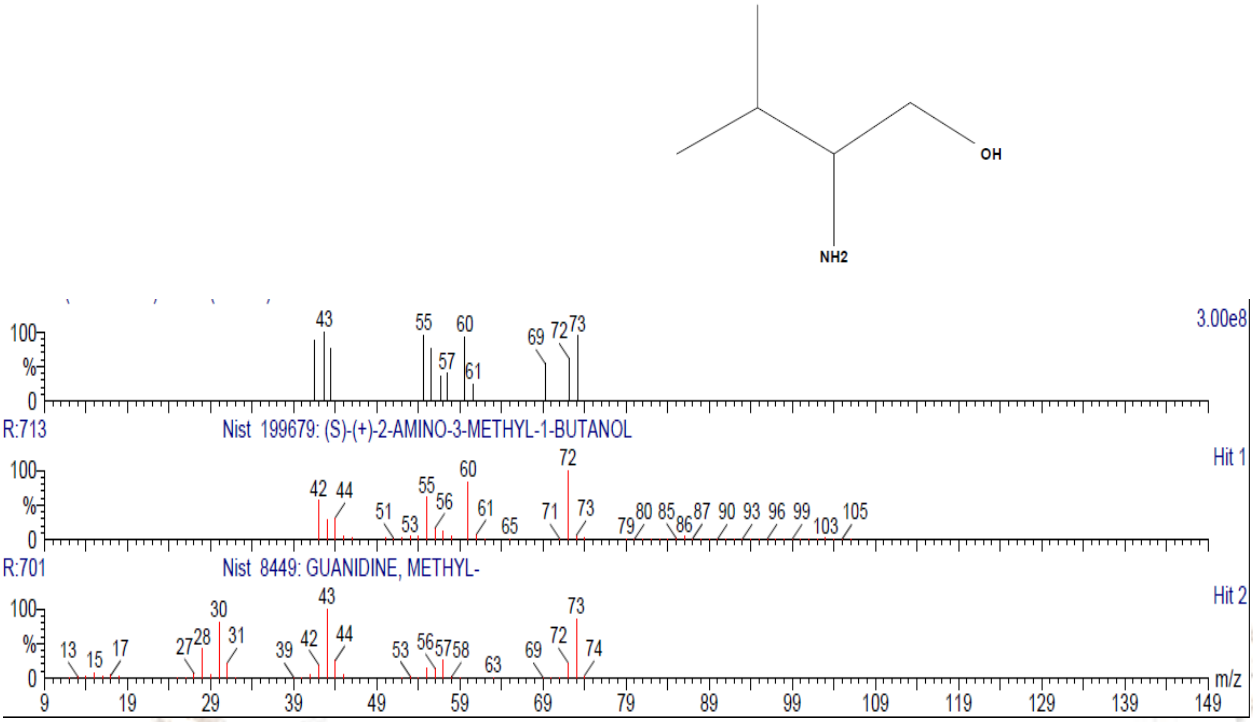

Fig 7: Mass spectrum of (S)-(+)-2-Amino-3-methyl-1-butanol (RT: 17.765)

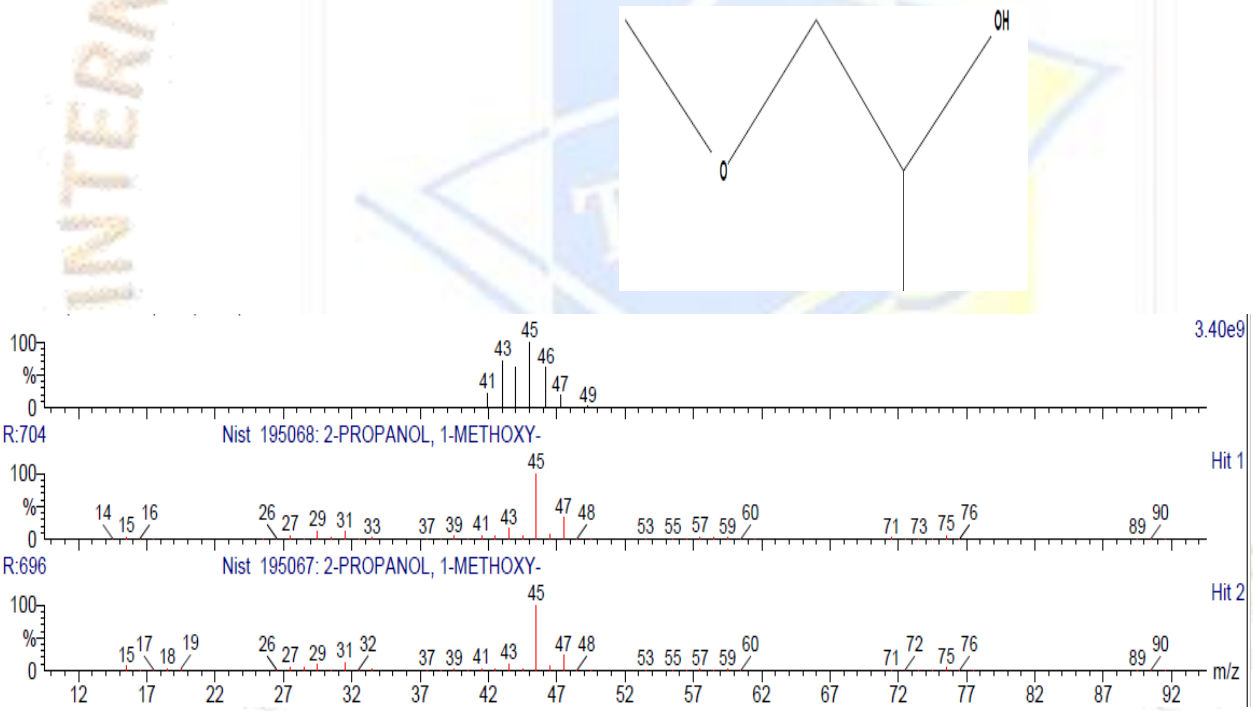

Fig 8: Mass spectrum of 2-Propanol, 1-methoxy- (RT: 1.419)

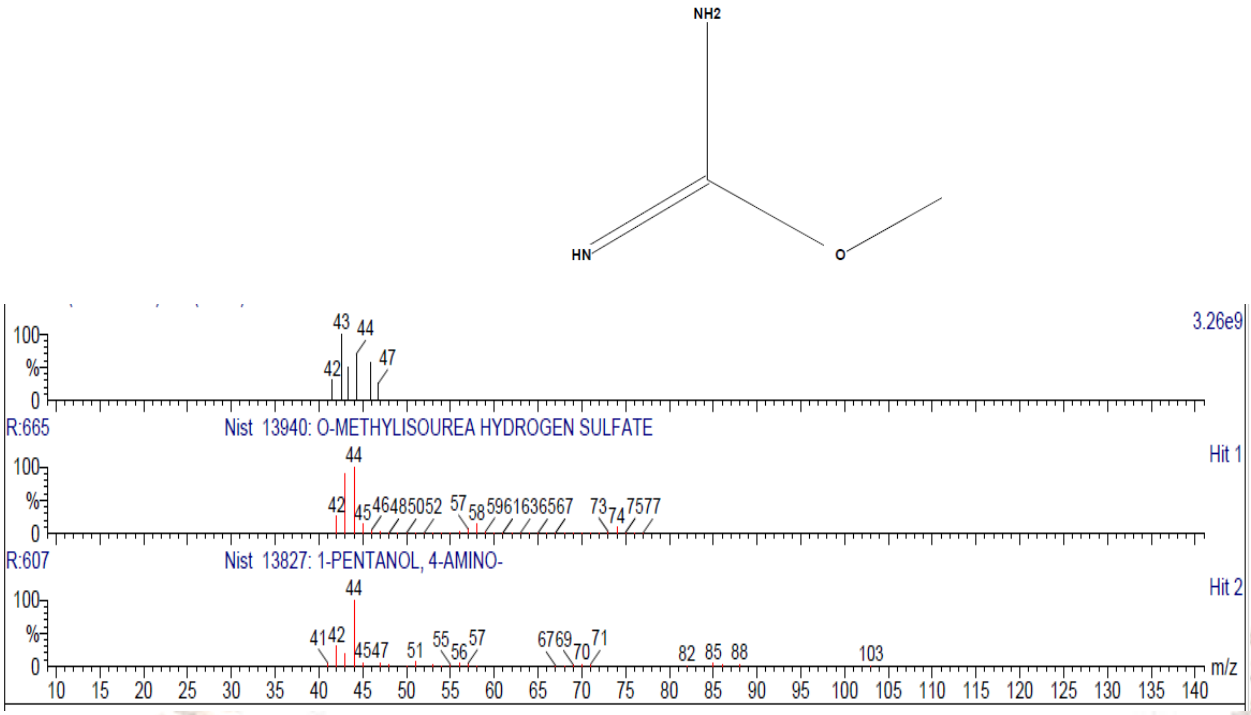

Fig 9: Mass spectrum of o-Methylisourea hydrogen sulfate (RT: 1.394)

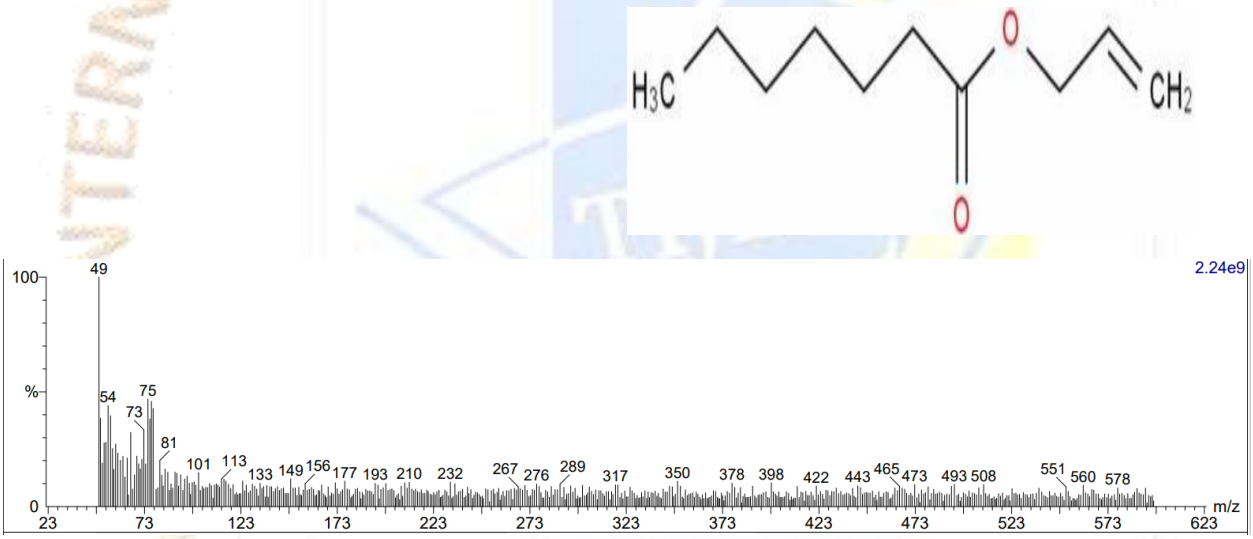

Fig 10: Mass spectrum of 3-Decanynoic acid (RT: 1.329)

Table 3: activity of phytocompounds showed in GC-MS analysis

S.no	Constituent	Activity
1	o-Methylisourea hydrogen sulfate	Anti-oxidant (7)
2	1-Hexyl-2-nitrocyclohexane	Anti-inflammatory (8) Larvicidal (9) Anti-diabetic (10)
3	1-Pentanol, 4-amino	Anti-oxidant (11)

CONCLUSION:

In this study, the GC-MS analysis of the ethanolic extract of Morinda Tinctoria fruit showed the presence of Eight compounds. In terms of the percentage amounts of phytoconstituents are Bicyclo [4.1.0] heptane,7-pentynyl (10.095%), 1-Hexyl-2-nitrocyclohexane (29.189%), 1-Pentanol, 4-amino (1.140%), 2,2,3,3,4,4-Hexamethyltetrahydrofuran (%), Cyclohexyl methyl formate (%), (S)-(+)-2-Amino-3-methyl-1-butanol (1.576%), 2-Propanol, 1-methoxy- (3.523%), 3-Decanynoic acid (25.506%) and o-Methylisourea hydrogen sulfate (1.246%). These compounds were shown to have antioxidant, antidiabetic, larvicidal and anti-inflammatory. Thus, this type of GC-Ms analysis is the first step of analysis of the nature of active principles in medicinal plants and type of study will lead to further studies.

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