

**GC-MS ANALYSIS OF CHLOROFORM EXTRACT OF
*CROTON BONPLANDIANUM*****KALAIVANI.M.KUPPUSWAMY¹, BHAVANA JONNALAGADDA²
AND SUMATHY AROCKIASAMY*¹**¹*Department of Biomedical Sciences, Sri Ramachandra University*²*Department of Biotechnology, S.R.M. Arts and Science College***ABSTRACT**

Croton bonplandianum belongs to Euphorbiaceae family has been used to cure liver diseases, swelling of the body, cure against ring worms and skin diseases. This study was aimed to identify the chemical compounds present in the chloroform leaf extract of *Croton bonplandianum* by GC - MS analysis. The analysis revealed the presence of sixteen phytochemicals which includes mainly long-chain fatty acids, fatty acid methyl esters and alcoholic compounds. The major compounds were 2, 4-bis (1, 1-dimethylethyl) - phenol (12.09%), 9-Eicosene (9.83%), 7-hexadecene (9.47), 1-Nonadecene (8.25%) and n-Nonadecanol-1(8.25%) identified. These compounds possess antibacterial, anti-tubercular and cytotoxic activities.

KEY WORDS: *Croton bonplandianum*, GC-MS analysis, phytochemicals, biological activity.**SUMATHY AROCKIASAMY**

Department of Biomedical Sciences, Sri Ramachandra University

INTRODUCTION

Herbal plant products have long been used to treat various ailments due to their therapeutic potential components present in them. According to WHO in 2008, 80% of the world population depends on traditional medicine for their health care¹. Medicinal plants as sources of bioactive compounds continue to play crucial role in the treatment of various diseases. Plants are considered as a reservoir of secondary metabolites with good biological properties. Hence exploring the chemical constituents of plants are utmost important not only for the discovery of novel compounds but for disclosing the new avenue in knowing the economic phytochemicals in synthesizing the new compounds and for emphasizing the importance of folklore remedies². Meanwhile, plants also synthesize toxic substances to defend against pathogens. Hence a thorough testing of every herbal product is compulsory in order to make it suitable for mankind usage. *C. bonplandianum*, an exotic weed belongs to Euphorbiaceae family is rich in secondary metabolites such as alkaloids, flavonoids, phenols, terpenoids etc and its leaves have the ability to control blood pressure, treatment of skin diseases, acts as an antiseptic and antidote³. It possesses a high anti-microbial and anti-fungal activity and also used by ethnic groups to reduce headache⁴. The seeds have the efficacy to cure jaundice, acute constipation, abdominal dropsy and internal abscesses⁵. The leaf extract has been proved to have wound healing effect and external application has shown to cure the ringworm infection. The seed of *C. bonplandianum* contains diterpenes, phorbol ester, including 12-orthotrideconeolyl-phorbol-13-acetate (TPA) and myristoyl phorbol acetate (MPA)⁶.

MATERIALS AND METHODS

Collection and Processing of Plant Material

The plant was collected from Chennai, Tamilnadu, India and authenticated, (No:PARC/2011/1021) by Dr. Jayaraman, Director, Plant Anatomy Research Center and stored in the Department of Biomedical Sciences, Sri Ramachandra University, India.

Preparation of Powder and Extract

The leaves were shade dried and grinded into a coarse powder. 100g of the coarsely powdered leaf material was extracted with 500 ml of chloroform at room temperature for 72 hours. The extracts were filtered through Whatmann filter paper No: 1 and evaporated to dryness and stored at -20°C until use.

GC-MS Study

GC-MS analysis was carried out on GC-MS-5975C [AGILENT] under the following conditions. DB-5ms Agilent (30.0mx0.25mmx0.25µm) was used. Helium was used as the carrier gas (99.9995% purity) at a constant flow rate of 1.51 ml/min and an injection volume of 2 µl was employed in a split mode. The injector temperature was maintained at 240°C and the column temperature was programmed to 70°C (isothermal for 2 min) with increasing temperature of 10°C/min to 300°C (isothermal for 9 min). Ion source temperature of 200°C and interface temperature of 240°C were maintained. The mass spectra were obtained through ionization energy of 70 eV in the EI mode. Total GC-MS running time was 30 min. The organic compounds were identified by comparison of mass spectra with the inbuilt libraries (NIST - 11).

Identification of components

The components present in the extract was identified based on the spectrum obtained using the data base of National Institute of Standard and Technology (NIST) having 62,000 patterns. The name of the compound, retention time, molecular formula and structure were determined. The area percentage of each component was calculated by comparing its average peak area to the total areas. The spectrum of the unknown compound was compared with the spectrum of the known compound stored in the NIST library.

RESULTS AND DISCUSSION

GC-MS analysis of the *C. bonplandianum* reveals the presence of 16 compounds

(Figure 1) of which 10 compounds were found to have biological activities. These compounds were confirmed based on their peak area, retention time, molecular formula and molecular weight (MW) and are represented in the table 1 and figure 1. The compounds identified possess many biological activities such as 1-Dodecanol (4.25%)⁷, 2, 4-bis(1,1-dimethylethyl)-phenol (12.09%)⁸, 7-hexadecene (9.47%)⁹ and n-Tetracosanol-1(5.67%)¹⁰ exhibited good anti-bacterial activity. 1-Tetradecene (7.38%) and 1-Nonadecene (8.25%) are long-chain fatty acids showed anti-tuberculosis activity¹¹ and

anti-fungal activity respectively¹². Further, 5-Octadecene was found to have strong sexual attraction in lactating mothers (9.47%)¹³. 9-Eicosene (9.83%) a long-chain fatty acid and n-Nonadecanol-1(8.25%), an alcoholic compound has potent anti-microbial¹⁴ and cytotoxic properties¹⁵ respectively. Methyl stearate (0.76%) is reported to have anti-diarrheal¹⁶ and cytotoxic and anti-proliferative¹⁷ activities. Based on the phytochemicals present with reported potential biological activities in earlier studies, this plant could be used as an important ingredient in the folklore medicine.

Figure 1
GC-MS analysis of chloroform leaf extract of *C.bonplandianum*

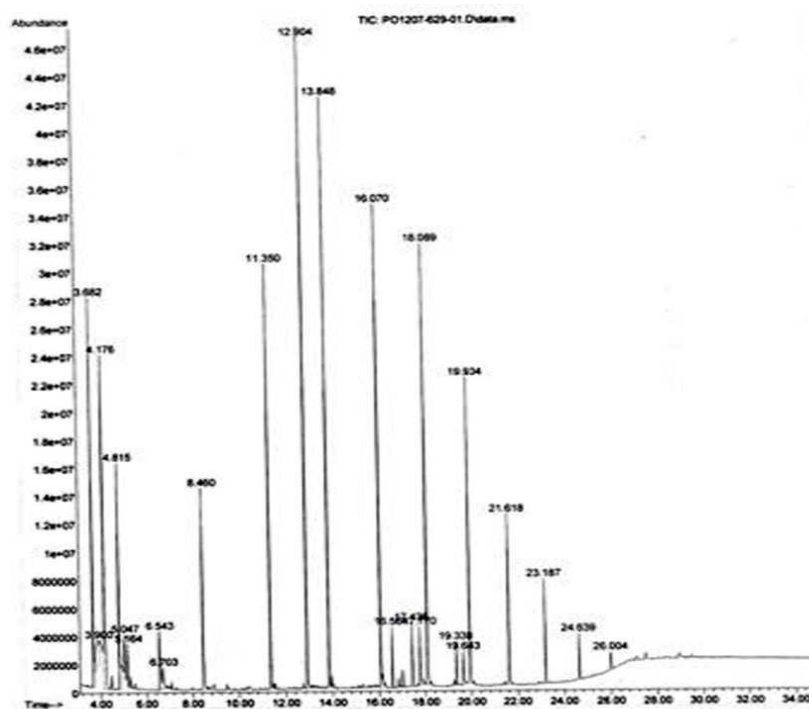


Table 1
GC-MS analysis of chloroform leaf extract of *C.bonplandianum*

S.No	Compound Name	Retention Time	Molecular Weight	Formulae	Area%
1	1-Dodecene	8.460	56	C ₁₂ H ₂₄	4.25
2	1- Dodecanol	8.460	70	C ₁₂ H ₂₆ O	4.25
3	1-Tetradecene	11.350	69	C ₁₄ H ₂₈	7.38
4	2,4-bis(1,1-dimethylethyl)-phenol	12.904	206	C ₁₄ H ₂₂ O	12.09
5	7-Hexadecene, (Z)-	13.848	224	C ₁₆ H ₃₂	9.47
6	5-Octadecene, (E)-	13.848	252	C ₁₈ H ₃₆	9.47
7	9-Eicosene, (E)-	16.070	280	C ₂₀ H ₄₀	9.83
8	1-Nonadecene	18.089	266	C ₁₉ H ₃₈	8.25
9	n-Nonadecanol-1	18.089	284	C ₁₉ H ₄₀ O	8.25
10	Methyl stearate	19.338	298	C ₁₉ H ₃₈ O ₂	0.76
11	n-Tetracosanol-1	19.934	354	C ₂₄ H ₅₀ O	5.67

Table 2
GC-MS analysis showed phytochemical compounds, their nature and their reported biological activities of chloroform leaf extract of *C. bonplandianum*

S.No	Peak area %	Retention time	Compound Name	Compound Nature	Activity
1	4.25	8.460	1-Dodecanol	long-chain alcohol	Anti-bacterial activity ⁸
2	7.38	11.350	1-Tetradecene	long-chain fatty acid	Anti-tuberculosis activity ¹²
3	12.09	12.904	2,4-bis(1,1-dimethylethyl)-phenol	Alcoholic compound	Anti-bacterial activity ⁹
4	9.47	13.848	7-hexadecene	long-chain fatty acid	Anti-bacterial activity ¹⁰
5	9.47	13.848	5-Octadecene	long-chain fatty acid	Stronger sexual characters ¹⁴
6	9.83	16.070	9-Eicosene	long-chain fatty acid	Anti-microbial and cytotoxic properties ^{15,16}
7	8.25	18.089	n-Nonadecanol-1	Alcoholic compound	Anti-microbial and cytotoxic properties ¹⁵
8	5.67	19.934	n-Tetracosanol-1	Alcoholic compound	Anti-bacterial activity ¹¹
9	8.25	18.089	1-Nonadecene	long-chain fatty acid	Anti-fungal activity ¹³
10	0.76	19.338	Methyl stearate	Fatty acid methyl esters	Anti-diarrheal ¹⁷ and antiproliferative ¹⁸ activity

CONCLUSION

In this study, GC-MS analysis explored the presence of phytochemicals in the leaves of *C. bonplandianum* and justifies the medicinal usage of this plant in folklore medicine. Based on the literature survey we believe that this is the first report of GC-MS analysis of leaf extract of this plant. Hence, further studies are insisted to evaluate its bioactivity and toxicity profile through *in-vitro* and *in-vivo* models.

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CONFLICT OF INTEREST

None of us declare the conflict of interest.

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