

Chemical Profile of the Leaves from *Morus alba* L. Using GC-MS

José González^{1*}, Enrique Gómez², Julio C Pérez³, Max Monan⁴ and Frantz François-Haugrin⁴

¹Facultad de Educación en Ciencias Técnicas, UCP "Enrique José Varona", La Habana, Cuba

²Instituto de Farmacia y Alimentos, Universidad de La Habana, La Habana, Cuba

³Centro Nacional de Toxicología (CENATOX), Hospital Militar "Carlos J. Finlay", La Habana, Cuba

⁴ARVARNAM, Martinica, France

*Corresponding Author: José González, Facultad de Educación en Ciencias Técnicas, UCP "Enrique José Varona", La Habana, Cuba.

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Abstract

Using different extraction solvents, a preliminary chemical profile of the leaves from *Morus alba* L. that grow in Cuba was done. Chromatographic characterization after extraction with Ethyl Acetate, Hexane, Ethanol at 30%, EtOH 50% and EtOH 80% from three sections of the leaves (low, middle, high) by GCMS-QP 2010 Ultra Shimadzu were matched with NIST libraries after samples derivatization. Higher amount of chemical components was get by the extraction with hexane, while the lowest was with EtOH at 30%. Rotenone, a flavonoid that belongs to Rotenoids subgroup was detected only five times into the extracts.

Keywords: *Morus alba* L; Chemical Profile; GC-MS; Leaves; Sections; Extracts

Introduction

Morus alba L. (Moraceae) commonly known as morera or mulberry is a medicinal and nutritional plant which is recorded in Compendium of Materia Medica, a famous ancient Chinese medicinal work written by Li Shizhen in 1590. In China, the mulberry tree has been cultivated since 4 thousand years B.C., while it has been cultivated in Europe probably in early Middle Ages [1,2].

Leaves of the plant have been scrutinized to determine its chemical constituents. These are mostly polyphenolic substances, flavonoids, anthocyanins, lectins, oligosaccharides, enzymes, digestive enzyme inhibitors, stilbene glycosides, anti-bacterial substances, unsaturated fatty acids and many other physiologically active substances. Those ingredients are present in the leaves, fruits, roots and stems, however the richest source of bioactive substances are in the leaves [3,4].

Morus L. is the main source plant of sugar-mimic alkaloids, and 1-deoxynojirimycin (DNJ), a sugar-mimic alkaloid is the highest

content compound in it, which has been used for the treatment of diabetes in clinic [5,6]. The aim of this research was to determine the chemical composition of the leaves of *Morus alba* that grow in Cuba using GC-MS.

Materials and Methods

Plan sample

M. alba variety was cultivated in Havana province, in 2018. The plants were divided in three sections (low, middle and high) taking into account the cutting height of 1 m. The leaves were selected taking into consideration their size, texture, color and health. The biomass was properly dried in an oven with controlled temperature after their harvesting.

Extract preparation

Each section was extracted by maceration during 7 days with 100 mL of ethyl acetate, hexane, EtOH at 30%, EtOH at 50%, and EtOH at 80%, respectively. After each extraction, the sample was

dried at room temperature to eliminate the solvent residue. At the end of the extractive method, fifteen extracts of 20 mL were obtained after extract concentration in a rotatory evaporator at 120 rpm, at 70 °C and 500 mbar.

GC-MS analysis

For the identification of metabolites, the samples were subjected to chromatographic analysis in equipment GC/MS, brand Shimadzu QP2010, equipped with a splitter split/split less. With a BP5 (30m × 0.25 mm × 0.25microns) capillary column under the following chromatographic conditions: Helium gas carrier obtained by electron impact fragments to a power of 70 eV rate of 1.2 mL/min, 1:50 split flow and the volume of injected sample of 1 µL. Programmed oven temperature: initial temperature was 70°C with a heating ramp of 10°C/min to 300°C and remained stable at this temperature for 10 minutes. Subsequently the temperature was increased at a rate of 10°C/minute to 300°C for a total time of 78 minutes with an injector temperature 250°C and the interface temperature 300°C. The compounds were analyzed using GC/MS NIST21 and NIST107 library and having into account the results obtained after phytochemical screening according with González, *et al.* 2021 [7]. Silylation agent was N, O-bis (trimethylsilyl) trifluoroacetamide (BSTFA) CAS 25561-30-2 Lot: 0901-1 Macherey-Nagel GmbH and C. KG.

Results and Discussion

Table 1 summarize the general results of the chemical characterization after samples derivatization and chromatographic analysis by GC-MS. Hexane extract show the best chemical compound average detected, while the lowest average was detected with EtOH at 30%. The highest average among the section belongs to the middle part of the plants, while the high section exhibit the lowest average of chemical compounds detected using different solvents of extraction.

Taking into account the presence or absence of rotenone, a flavonoid that belongs to Rotenoids subgroup, this chemical compound was found out only five times among the 15 extracts analyzed in the research. In the case of ethyl acetate, rotenone was detected two times at different retention times, in the low (46.865min) and high sections (42.615min), respectively. Into hexane extract this chemical compound was detected only in low section (Rt 46.020min), while in EtOH at 50% was detected in middle section (Rt 41.720min) and in EtOH at 80% the metabolite was detected in the low section at 50.185min of retention time.

| Sections | Ethyl acetate | Hexane | EtOH 30% | EtOH 50% | EtOH 80% | Section Average |
|----------|---------------|--------|----------|----------|----------|-----------------|
| Low | 900* | 925* | 821 | 916 | 888* | 890 |
| Middle | 905 | 931 | 909 | 924* | 873 | 908.4 |
| High | 914* | 928 | 846 | 859 | 850 | 879.4 |
| Average | 906.3 | 928 | 863 | 899.6 | 870.3 | |

Table 1: General results of chemical components characterized by GC-MS in different sections of *Morus alba* L.

* Rotenone presence

Conclusions

GC-MS was used for the first time to characterize the chemical components of the leaves from *Morus alba* L. that grow in Cuba. The results of this research allow to conclude that the influence of chemical solvent and the section of the plant harvest have a big influence in the amount and kind of chemical components that researchers can get in the investigations of medicinal plants. EtOH at 30% showed the poor amount of chemical constituents and rotenone flavonoid was not detected in this extract using this chromatographic method. Further investigations will be necessary to determine the chemical composition of this medicinal plant, especially HPLC-MS and NMR spectroscopy.

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