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Isolation and Characterization of *para*-hydroxy quinone from *Morinda Pubescence* leaves.

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ABSTRACT

Many species of the genus Morinda are found to be rich in secondary metabolites having a medicinal value.Morinda Pubescence (Rubiaceae) is a medicinally important plant being used in different indigenous systems of medicine such as Ayurveda, Siddha, Unani, Tibbi and Amchi. Extract of its leaves, stems and fruits are used as traditional remedy for various diseases such as in the treatment of gastropathy, dyspepsia, diarrhoea, stomach ulcer, wounds, gout, inflammation, hernia, sarcocele etc. Selective solvent extractions,steam distillation, chromatography, preparative TLC along with mixed solvent system for crystallization were the major techniques used for isolation of compounds, while structures were elucidated by integration of data from IR, UV, ^{1}H NMR and ^{13}C NMR analysis and analysized by DFT technique. The presence of para-hydroxy quinone was accounted first time from the leaves of M. Pubescence .

Key words: *Morinda Pubescence*, preparative TLC, DFT technique *para*-hydroxy quinone

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INTRODUCTION

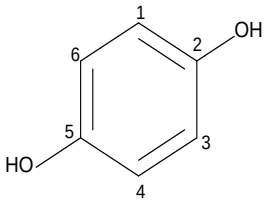
Since time immemorial, mankind has been dependent on plants for food, shelter, medicine, energy and other useful products. Indigenous medical systems totally depend on plants for their supply of drugs. As the allopathic system of medicine has proven to have other side effects, people look to herbal medicine as an alternative source of medical treatment. Therefore, pharmaceutical industries are faced with the problem of producing drugs of quality, in greater quantity, correct identification of the drugs and the identification and isolation of the active principles present in the plants. In the twenty first century, rapid progress has been made in the field of herbal medicines, and most of the countries have developed standard methods for tapping drugs from plants and have published pharmacopoeias for reference. Many species of the genus Morinda (Rubiaceae) are found to be rich in secondary metabolites having a medicinal value [1]**.** *M. pubescens* is a medicinal plant which is important in ethno medicine. Herbal medicines are reported to be used as styptic, alexeteric, digestive, carminative, febrifuge, tonic, dysentery, inflammations, boils and general debility. Hence, it is used in gastropathy, dyspepsia, diarrhoea, stomach ulcer, wounds, gout, inflammation, hernia, sarcocele etc [2,3].Some bioassay research conducted on *M.pubescens* showed a wide range spectrum of antibacterial activity[4]. The reports discloses the presence of glycoside, morindone **,** tinctomorone, damnacanthal and nordamnacanthal, ursolic acid, anthraquinones, soranjidol, ibericin, rubiadin,D-mannitol and anthragallol-2, and

3-di methyl ether[5-10]**.** Theoretical calculations are carried out with Gaussian 09 program using the Density Functional Theory (B3LYP/6-31G* level) and RHF (6-31G*) levels of the theory to predict the molecular structure and wave numbers in IR region. Taking into consideration the above facts isolation and characterization of *M. Pubescence* leaves was achieved.

MATERIALS AND METHODS

The plant material was collected from Pune, Maharashtra; India. It was authenticated at Agharkar Research Institute, Pune Maharashtra, India. Its authentication No. is AHMA-21220.

Steam volatile crude product depicted bioactive nature so bio-guided separation of it was accomplished by using preparative chromatographic technique. The *para*-hydroxy quinone was isolated by employing preparative TLC along with mixed solvent system for crystallization. The structure was assigned by modern spectral technique and assisted by DFT technique **(Fig.1, Table 1 & 2)**. Data is presented *para*-hydroxy quinone, compound 1**,** was isolated from this plant source for the first time.



Compound 1

RESULTS AND DISCUSSION

It is a white square crystalline solid. It shows its sharp melting nature at 172^0C. The LC-MS spectrum has exhibited the molecular ion peak at 110 *m/z*, suggesting the molecular formula be $C_6H_6O_2$. **GC-MS** confirms the presence of hydro quinone by 96 % similarity index.**IR** spectrum indicates characteristic broad peak at 3318 cm^{-1} for the existence of hydroxy groups. A typical indentation is observed at 3010 cm^{-1} illustrates the presence of = C-H stretching. 1514cm^{-1} &1468 cm^{-1} are displayed for aromatic stretching frequency. Presences of C-O stretching frequencies are detected at 1240 $cm^{-1}$and 1211 cm^{-1}.1096 cm^{-1},1010 cm^{-1},947cm^{-1} & 907 cm^{-1}. **^{1}H NMR** spectrum reveals a downfield singlet at δ 6.62 (s, 4H) for H 3, H 4, H 1 & H 6 identical and equivalent aromatic protons. A broad singlet is depicted at δ 3.3 for two hydroxyl protons.

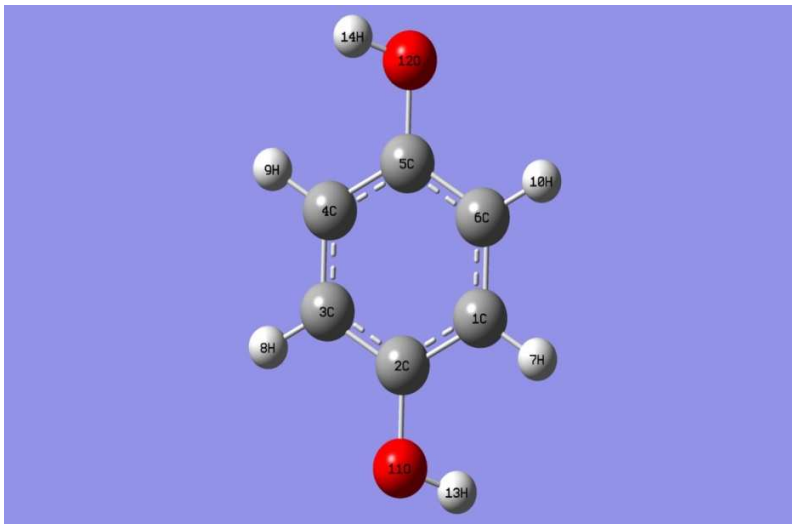


Fig.1 Parameters from DFT (B3LYP/6-31G* level) for compound 1

Table 1 - Parameters from DFT (B3LYP/6-31G* level) for compound 1

Atoms	Bond length (A^0)	Atoms	Bond angle (0)
C(1) - C (2)	1.401	C(1) - C (2) –C (3)	120
C(3) - C (2)	1.401	C(4) - C (3) –C (2)	120
C(4) - C (3)	1.401	C(5) - C (4) –C (3)	120
C(5) - C (4)	1.401	C(6) - C (1) –C (2)	120
C(5) - C (6)	1.401	H(7) - C (1) –C (2)	120
H(7) - C (1)	1.070	H(8) - C (3) –C (2)	120
H(8) - C (3)	1.070	H(9) - C (4) –C (3)	120
H(9) - C (4)	1.070	H(10) - C (6) –C (1)	120
H(10) - C (6)	1.070	O(11) - C (2) –C (1)	120
O(11) - C (2)	1.430	O(12) - C (5) –C (4)	120
O(12) - C (5)	1.430	H(13) - O(11) –C (2)	109.471
		H(14) - O (12) –C (5)	109.471

Table 2 - Dihedral angles of compound 1

Atoms	Dihedral angle (0)
H (7) – C (1) – C (2) – C (3)	180.00
H (8) – C (3) – C (2) – C (1)	-180 .00
H (9) – C (4) – C (3) – C (2)	180.00
H (10) – C (6) – C (1) – C (2)	180.00
O (11) – C (2) – C (1) – C (6)	180.00
O(12) – C (5) – C (4) – C (3)	180.00
H (13) – O (11) – C (2) – C (1)	-30.00
H (14) – O (12) – C (5) – C (4)	30.00

CONCLUSION

A bioactive molecule, *para*-hydroxy quinone has been isolated for the first time from this plant source. Due to its ubiquitous occurrence in the environment it plays an important role in drug discovery due to its chemical and pharmacological diversity.

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