EUROPEAN ACADEMIC RESEARCH
Vol. VIII, Issue 9/ December 2020

Impact Factor: 3.4546 (UIF)
DRJI Value: 5.9 (B+)

ISSN 2286-4822
www.euacademic.org

Evaluation of Phytotoxicity and FTIR analysis of extract and fractions of *Hertia intermedia*

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Abstract

In this research study, the extract and fractions of Hertia intermedia were evaluated for phytoxicity and FTIR analysis. Phytotoxicity assay and FTIR studies were the techniques used in this research. HFHI, AFHI and BFHI showed low phytotoxicity 13%, 5% and

20% at 100 µg/mL. HFHI and AFHI exhibited moderate activity 37% and 39% at 1000 µg/mL while BFHI showed significant phytotoxicity 100% at 1000 µg/mL. The compounds present in Methanol Extract of Hertia intermedia (MEHI) with their matched spectra with those available in the libraries are D-Erythrose, Hydroxyethyl-b-cyclodextrin, Opium powder in KBR, Hydroxyethyl cellulose, Chondroitin Sulphate, 1,2,6-Trihydroxyhexane, Dextrin, Stachyose. 4H2O, Corn Syrup Solid and 1,5-Pentanediol.

The compounds present in n-Hexane Fraction of Hertia intermedia (HFHI) with their matched spectra with those available in the libraries are D-Erythrose, Carboxymethyl cellulose, Sucrose in KBR, 1,3-Propanediol, Corn Syrup Solid, Hydroxyethyl cellulose, Opium powder in KBR, Hydroxyethyl cellulose, Chondroitin Sulphate, 1,2,6-Trihydroxyhexane. The compounds present in Fraction of Hertia intermedia (AFHI) with their matched spectra with those available in the libraries are Adonitol, 2,5-O-Methylene-D-Mannitol, trans-1,2-Cyclohexanediol, cis-4-Methylcyclohexanol, Tetrahydro-4H-pyran-4-ol, 2,5-Dichloro-p-xylene, Chenodeoxycholic acid, Mannito, b-D-Glucose pentacetate and D-Allose. The compounds present in Butanol Fraction of Hertia intermedia (BFHI) with their matched spectra with those available in the libraries are Butyl sulfoxide, 1,2-Pentanediol, 2-Methyl-1-Pentanol, Tributyl phosphate, Hydroxyethyl-b-cyclodextrin, 4-Methyl-1-Pentanol, Amygdain, 1,5-Pentanediol, 3, Cyclopentyl-1-propanol and 4-Ethylbenzyl alcohol.

Keywords: Phytotoxicity, FTIR, MEHI, HFHI, AFHI, BFHI, *Hertia intermedia*

INTRODUCTION

Natural products are gaining significance due to drug resistance and side effects of chemically originated drugs [1]. The most significant plant derived drugs are morphine, Taxol, Digitoxigenin, vincristine, camptotecin, atropine, podophyllum, artemesininaspirine, allicin, pilocarpine, curumin, capscicine, codeine, lotoxingitxigenin, vinblastine and tubocurarine [2]. The native herbalists and experienced

elders of the society are the traditional doctors from whom people seek treatment and advices [3]. Tribe Senecioneae, family Compositae consists of important genus Hertia which comprises of 12 species, present in south west Asia and north and south of Africa [4]. Othonnopsis intermedia is the alternative name of *Hertia intermedia* which is found in Balochistan for instance in Chaman, Quetta, Khanozai. Hertia intermedia are tiny shrubs with flowers of yellow colour [5]. The medicinal plant, Hertia intermedia is traditionally used as a painkiller in Pakistan. The phytochemicals are known as hertidins [6]. The phytochemical which are investigated are terpenoids, tannins, flavonoids, coumarins, and steroids [7]. In this study, phytotoxicity and FTIR analysis of extract and fractions of *Hertia intermedia* are examined.

MATERIALS AND METHODS

Collection and Identification of plant

The medicinal plant, *Hertia intermedia* was collected from Yaro, Pishin and identified by a taxonomist Prof. Dr. Rasool Bakhsh Tareen, Department of Botany, University of Balochistan, Quetta, Pakistan. Under the co-supervision of Prof. Dr. Muhammad Anwar Panezai, the extraction and fractionation of Hertia intermedia was carried in the Institute of Biochemistry, University of Balochistan, Quetta.

Extraction and Fractionation

The plant, *Hertia intermedia* was shade dried for the period of month, grinded into powdered and then 3kg was soaked in 10 litres of methanol for 7 days. Filtered and then methanol was vaporized with the help of rotary evaporator. The crude extract was 104gm which was further fractionated with n-hexane and aqueous to obtain n-hexane fraction 6.5 gm and aqueous fraction 35 gm. Aqueous fraction was further fractionated with butanol to produce butanol fraction 3 gm [8, 9].

Phytotoxicity assay

In phytotoxicity assay, for the preparation of medium, various constituents were mixed in 1000 mL distilled water and then by addition of KOH pellets, the pH was adjusted 6.0-7.0. In 1.5 mL

methanol, 30 mg extract and fractions were dissolved to form stock solutions. The stock solutions were diluted to 10, 100 and 1000 µg/mL. Under sterile condition, the solvents were evaporated overnight. 20 mL medium and 10 plants containing a rosette of two fronds of Lemna minor were added to each flask. Reference plant growth inhibitor (Paraquate) and medium were supplemented in other flasks as positive and negative controls. For 7 days, in growth cabinet, all flasks were kept. At the end of the incubation period, the number of fronds per flask was counted and recorded [10]. With the help of following formula the percentage of growth regulation was determined:

$$\text{Growth regulation (\%)} = \frac{100 - \text{number of fronds in test samples}}{\text{Number of fronds in negative control}} \times 100$$

FTIR analysis

The extracts and fractions of plant were dried for the analysis of FTIR. In FTIR analysis, the extract and fractions of plant with the concentration of 10 mg were encapsulated in the pellet of 100 mg of KBr, for the preparation of disc of translucent sample which was then loaded in FTIR Spectroscope (Shimadzu, IR Affinity1, Japan) [11].

RESULTS AND DISCUSSION

In this research study, the methanol extract and its fractions such as n-hexane, aqueous and butanol fractions were examined for invitro phytotoxicity and FTIR analysis. HFHI, AFHI and BFHI showed low phytotoxicity 13%, 5% and 20% at 100 µg/mL. HFHI and AFHI exhibited moderate activity 37% and 39% at 1000 µg/mL while BFHI showed significant phytotoxicity 100% at 1000 µg/mL. The phytotoxicity activities of extract and fractions of *Hertia intermedia* are shown in table 1.

Table 1 Phytotoxicity of extract and fractions of *Hertia intermedia*

Extact/Fractions	% Growth Inhibition		
	10 µg/mL	100 µg/mL	1000 µg/mL
MEHI	0	0	0
HFHI	0	13	37
AFHI	0	5	39
BFHI	0	20	100

The compounds present in Methanol Extract of *Hertia intermedia* (MEHI) with their matched spectra with those available in the libraries are D-Erythrose, Hydroxyethyl-b-cyclodextrin, Opium powder in KBR, Hydroxyethyl cellulose, Chondroitin Sulphate, 1,2,6-Trihydroxyhexane, Dextrin, Stachyose. 4H2O, Corn Syrup Solid and 1,5-Pentanediol and are shown in figure 1 and 2.

The compounds present in n-Hexane Fraction of *Hertia intermedia* (HFHI) with their matched spectra with those available in the libraries are D-Erythrose, Carboxymethyl cellulose, Sucrose in KBR, 1,3-Propanediol, Corn Syrup Solid, Hydroxyethyl cellulose, Opium powder in KBR, Hydroxyethyl cellulose, Chondroitin Sulphate, 1,2,6-Trihydroxyhexane and are shown in figure 3 and 4.

The compounds present in Fraction of *Hertia intermedia* (AFHI) with their matched spectra with those available in the libraries are Adonitol, 2,5-O-Methylene-D-Mannitol, trans-1,2-Cyclohexanediol, cis-4-Methylcyclohexanol, Tetrahydro-4H-pyran-4-ol, 2,5-Dichloro-p-xylene, Chenodeoxycholic acid, Mannito, b-D-Glucose pentacetate and D-Allose and are shown in figure 5 and 6.

The compounds present in Butanol Fraction of *Hertia intermedia* (BFHI) with their matched spectra with those available in the libraries are Butyl sulfoxide, 1,2-Pentanediol, 2-Methyl-1-Pentanol, Tributyl phosphate, Hydroxyethyl-b-cyclodextrin, 4-Methyl-1-Pentanol, Amygdain, 1,5-Pentanediol, 3, Cyclopentyl-1-propanol and 4-Ethylbenzyl alcohol and are shown in figure 7 and 8.

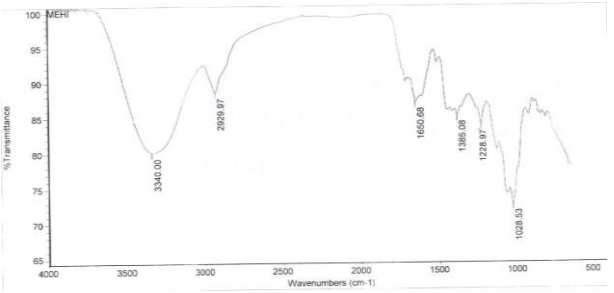

Figure 1: FT-IR Spectra of Methanol Extract of *Hertia intermedia* (MEHI)

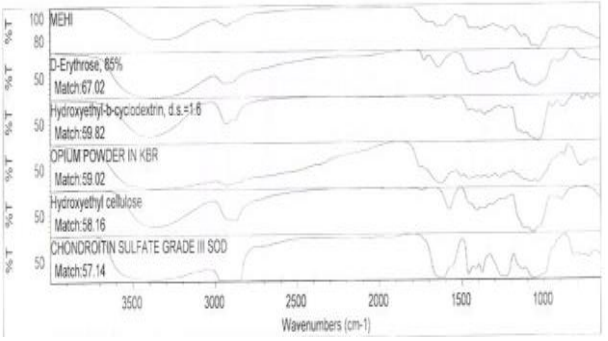

Figure 2: FT-IR Spectra of Methanol Extract of *Hertia intermedia* (MEHI) and their matched spectra with those available in the libraries

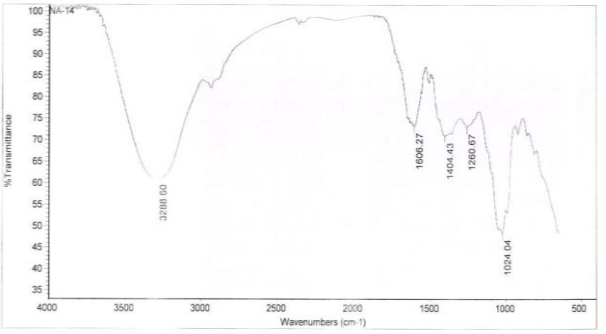

Figure 3: FT-IR Spectra of n-Hexane Fraction of *Hertia intermedia* (HFHI)

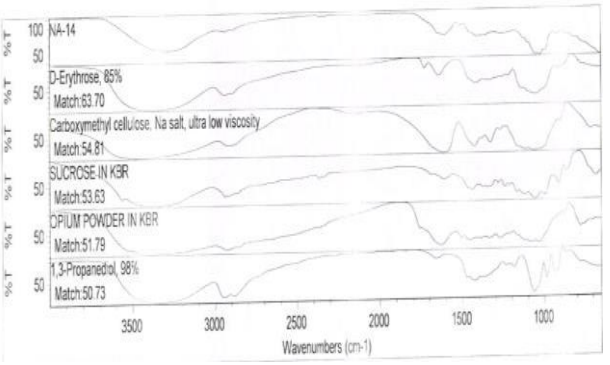

Figure 4: FT-IR Spectra of n-Hexane fraction of *Hertia intermedia* (HFHI) and their matched spectra with those available in the libraries

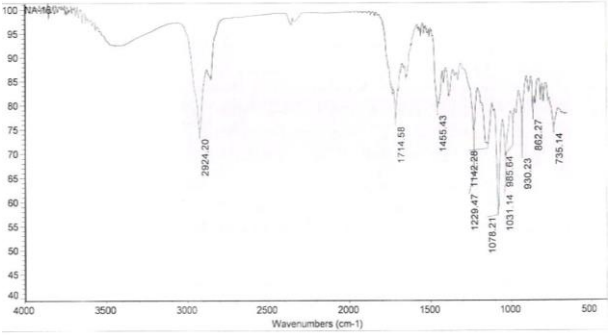

Figure 5: FT-IR Spectra of Aqueous Fraction of *Hertia intermedia* (AFHI)

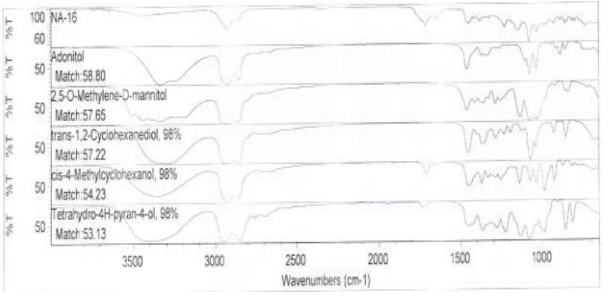

Figure 6: FT-IR Spectra of Aqueous fraction of *Hertia intermedia* (AFHI) and their matched spectra with those available in the libraries

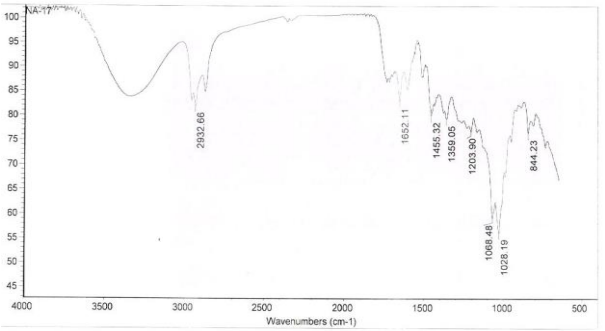

Figure 7: FT-IR Spectra of Butanol Fraction of *Hertia intermedia* (BFHI)

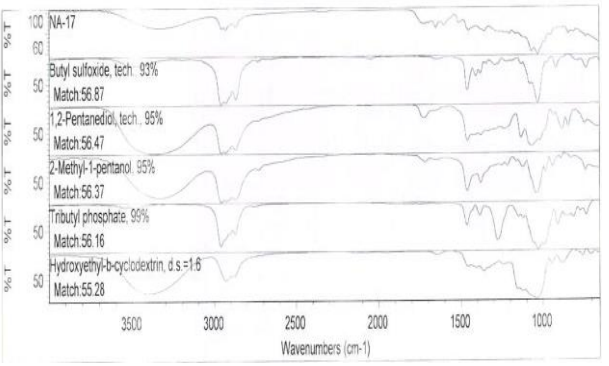

Figure 8: FT-IR Spectra of Butanol fraction of *Hertia intermedia* (BFHI) and their matched spectra with those available in the libraries

CONCLUSION

In this research study, we conclude that HFHI, AFHI and BFHI showed low phytotoxicity 13%, 5% and 20% at 100 µg/mL. HFHI and AFHI exhibited moderate activity 37% and 39% at 1000 µg/mL while BFHI showed significant phytotoxicity 100% at 1000 µg/mL. FTIR studies showed The compounds present in Methanol Extract of *Hertia intermedia* (MEHI) with their matched spectra with those available in the libraries are D-Erythrose, Hydroxyethyl-b-cyclodextrin, Opium powder in KBR, Hydroxyethyl cellulose, Chondroitin Sulphate, 1,2,6-Trihydroxyhexane, Dextrin, Stachyose. 4H2O, Corn Syrup Solid and 1,5-Pentanediol.

The compounds present in n-Hexane Fraction of *Hertia intermedia* (HFHI) with their matched spectra with those available in

the libraries are D-Erythrose, Carboxymethyl cellulose, Sucrose in KBR, 1,3-Propanediol, Corn Syrup Solid, Hydroxyethyl cellulose, Opium powder in KBR, Hydroxyethyl cellulose, Chondroitin Sulphate, 1,2,6-Trihydroxyhexane. The compounds present in Fraction of *Hertia intermedia* (AFHI) with their matched spectra with those available in the libraries are Adonitol, 2,5-O-Methylene-D-Mannitol, trans-1,2-Cyclohexanediol, cis-4-Methylcyclohexanol, Tetrahydro-4H-pyran-4-ol, 2,5-Dichloro-p-xylene, Chenodeoxycholic acid, Mannito, b-D-Glucose pentacetate and D-Allose. The compounds present in Butanol Fraction of *Hertia intermedia* (BFHI) with their matched spectra with those available in the libraries are Butyl sulfoxide, 1,2-Pentanediol, 2-Methyl-1-Pentanol, Tributyl phosphate, Hydroxyethyl-b-cyclodextrin, 4-Methyl-1-Pentanol, Amygdain, 1,5-Pentanediol, 3, Cyclopentyl-1-propanol and 4-Ethylbenzyl alcohol.

Acknowledgement
The authors are thankful to Hussain Ebrahim Jamal (HEJ), Research Institute of Chemistry, University of Karachi, Karachi, Pakistan, for providing phytotoxicity studies and Institute of Biochemistry, University of Balochistan, Quetta, Pakistan for providing lab facilities for extraction and fractionation and FTIR studies.

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