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Phytochemical, FTIR and NMR Analysis of Crude Extract of *Duranta plumieri* leaves

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Abstract

Duranta repens belonging to family Verbenaceae commonly known as pigeon berry and locally called `Kata mehedi` and native of West Indies, northern parts of Pakistan and central and south America. Various extracts of petroleum, chloroform, ethanol and aqueous, was prepared from the leaves based on the polarity of the solvents using cold maceration method. Phytochemical screening of these extracts revealed the presence of terpenoids and flavonoids in chloroform extract in the sample. Fourier Transform Infrared (FTIR) spectroscopy revealed the presence of various functional groups such as –OH, -COOH, -CH_2, and C=O from the IR absorption bands in the high wave region at 3456 cm^{-1} and 2939 cm^{-1} and the active compounds were identified by comparing the retrieved compounds with the standard chart. The number of protons present and the electronic state of the protons in the various compounds was analysed using H^1 NMR (Nuclear magnetic resonance). Further studies are to be done for the isolation of the active compounds that can be used to determine its therapeutic implications for the development of new drugs.

Keywords**:** *Duranta repens***,** Phytochemical, terpenioids, Fourier Transform Infrared spectroscopy, functional groups, Nuclear magnetic resonance.

INTRODUCTION

Primary health care system uses traditional medicine as the most affordable and easily assessable source of treatment. There is a paradigm shift towards use of herbal remedies or herbal based formulations. The role of medicinal plants in disease prevention and treatment has always been remarkable [1]. The ethno botany provides a rich resource for natural drug research and development [2]. In many developing countries, a large proportion of the population relies on traditional practitioners and their armamentarium of medicinal plants in order to meet health care needs. Although modern medicine may exist side-by-side with such traditional practice, herbal medicines have often maintained their popularity for historical and cultural reasons [3-4].

Duranta repens commonly known as pigeon berry and locally called `Kata mehedi` belongs to the family Verbenaceae and native of West Indies, northern parts of Pakistan and central and south America. The genus Duranta comprises about 35 species which are evergreen shrubs distributed in tropical and sub-tropical region. It was introduced to Egypt as an ornamental plant in the 1920's. An infusion of leaf and juice of the fruit are diuretic and flower is said to have stimulant properties. The slightly poisonous fruit of plant afford a medicine for the treatment of malaria, which still stand a major disease in developing countries. The fruit juice can be used as larvicidal in ponds and swamps. The fruits are also used in the treatment of intestinal worms. The leaves are used in the treatment of abscess [5]. A number of iridoid glycosides such as the durantosides I, II, III, IV and lamiide were isolated from the genus Duranta. *In vivo* antimalarial activity of the fruits of *D. repen* was also reported against Plasmodium berghei. Antioxidant and antiviral activity was reported in the ethyl acetate soluble fraction of methanol extract of *D. repens*[6]. The phytochemical contents in therapeutic plants are the mixtures intensifying the demonstration specifically or in a indirect way avert or treat illness. The maceration method is a traditional technique used for extraction mainly used by the ayurvedas to treat several diseases based on various criteria. The samples are immersed in different solvent systems and incubated for 24-72 hrs. Phytochemical screening is done to detect the primary and secondary metabolites present in the drug. FTIR and H^1 NMR are used for the identification of functional groups of the compounds present in the sample and to detect the number of protons present in the compound respectively.

MATERIALS AND METHODS

Collection of Plant material and preparation of extract

Leaves of *Duranta plumirie* were collected from Fathepur, Distt Kangra, Himachal Pradesh and were identified taxonomically by Kumar Ambrish, Scientist, Botanical Survey of India, Dehradun, having Authentication Voucher no. 118055. The leaves were cleaned with tap water and then rinsed in distilled water. Next, these were shade dried and then coarsely powdered and subjected to extraction through cold maceration. Powder of 100 gm leaves was kept in a round bottom flask and different solvents (petroleum ether, chloroform, ethanol, and water) were added successively for 48 h and then filtered with muslin cloth. The filtered extract of various solvents was concentrated using rotary evaporator and dried in hot air-oven below 50°C. The dried extracts were stored in a vacuum dessicator.

Phytochemical Screening

Different extracts were screened for phytochemical tests using standard procedures to identify the phytoconstituents (7). Various tests like carbohydrates, gums, proteins, amino acids, fats, oils, steroids, glycosides, flavanoids, alkaloids, tannins, phenolics and terpenoids were performed.

FTIR (Fourier Transform Infrared Spectroscopy) analysis of the sample

The FT-IR spectra were obtained from PerkinElmer Spectrum Version 10.03.08 using KBr disc method (ν_{max} in cm^{-1}) for the preparation of the sample. The plant extract was mixed with KBR (1:10) and made into a pellet by hydraulic press/IR press. Pellet was then inserted in sample slit and then transmittance was observed(8-9).

Proton Nuclear Magnetic Resonance (H^1 NMR)

The NMR spectra were obtained using a Bruker Avance II 400 MHz spectrometer in deuterated chloroform ($CDCl_3$) or deuterated dimethylsulfoxide (DMSO-d_6) as the solvents, and the chemical shifts(δ) were recorded in parts per million (ppm) downfield from tetramethylsilane (Me_4Si) as the internal standard. The coupling constant (*J*) was expressed in Hz. The spin multiplicities were described as singlet (s), broad singlet (brs), doublet (d), double doublet (dd), doublet of double doublet (ddd), triplet (t), quintet (quint), doublet of triplets (dt) and triplet of double doublet (tdd) etc (10-12).

RESULTS AND DISCUSSION

The phytochemical screening shows better response for the presence of terpenoids and flavonoids in chloroform extracts as listed in Table 1. As per review of literature majority of the activities are due presence of terpenoids and flavonoids in the extract. The FTIR spectroscopy was carried out to ascertain functional groups. The FT-IR spectrum of IR absorption bands in the region 3200-3540 cm^{-1} Corresponds to –OH alcohol. The observed band at 2924 and 2854 can be assigned to –CH alkene and –CH alkane. The band at 1633-1452 and 1698 can be due to aromatic ring and ketones respectively. The band at 1079 and 869 can be due to –C-O stretching and C-C bending respectively (Table 2 and Figure 1). The H^1 NMR experiment was carried out and the spectrum showed multiplet at δ 0.879-0.774 for proton of alkyl group and the other alkyl protons can be seen in a range of δ 0.985-1.139 and δ 1.601-1.253 while protons of alkene can be observed in a range of δ 2.808-1.904. The proton of alcoholic group –OH has given signal in a range of δ 3.847-3.408 while amide and aryl protons are observed at δ 5.364 and δ7.409 respectively (Table 3 Figure 2).

Table 1: Phytochemical analysis of various extracts of Duranta plumirie leaves

S. No.	Test:	Pet ether	Chloroform	Ethanol	Aqueous
1.	Carbohydrates	+	-	+	+++
2.	Gums	-	-	-	+
3.	Proteins	-	-	-	-
4.	Test for amino acids	-	-	-	-
5.	Steroids	+	+	+	+
6.	Cardiac Glycosides	+	+	+	+
7.	Anthraquinone glycosides	+	_	+	+
8.	Saponin glycoside	-	-	-	+
9.	Flavanoids	-	+++	+	+
10.	Alkaloids	-	-	-	+
11.	Tannins and phenolics	-	-	-	+
12	Organic Acids	-	-	-	+
13	Terpenoids	-	+++	++	+

Figure 1: Spectra of Fourier Transform Infrared Spectroscopy

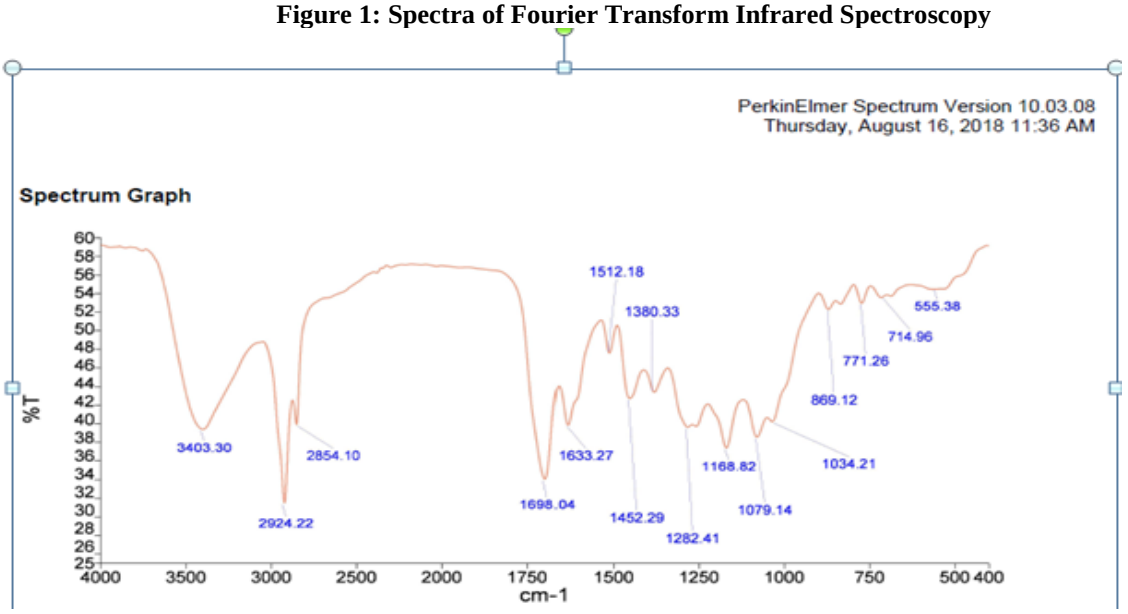

Table 2: Compound retrieved from FTIR spectrum

S. No.	Spectrum	Functional Group(Compounds)
1	3200-3540 cm^{-1}(H-bonded)	O-H(Alcohol)
2	2924 2854	-CH(sp^2)Alkene -CH(sp^3)Alkane
3	1633-1452	C-C (aromatic ring)
4	1698	$\succ$C=O(Ketones)
5	1079	-C-O stretching(Alcohol, acid, ester, ether)
6	869	C-C bending
7	1698	$\succ$C= C $\prec$

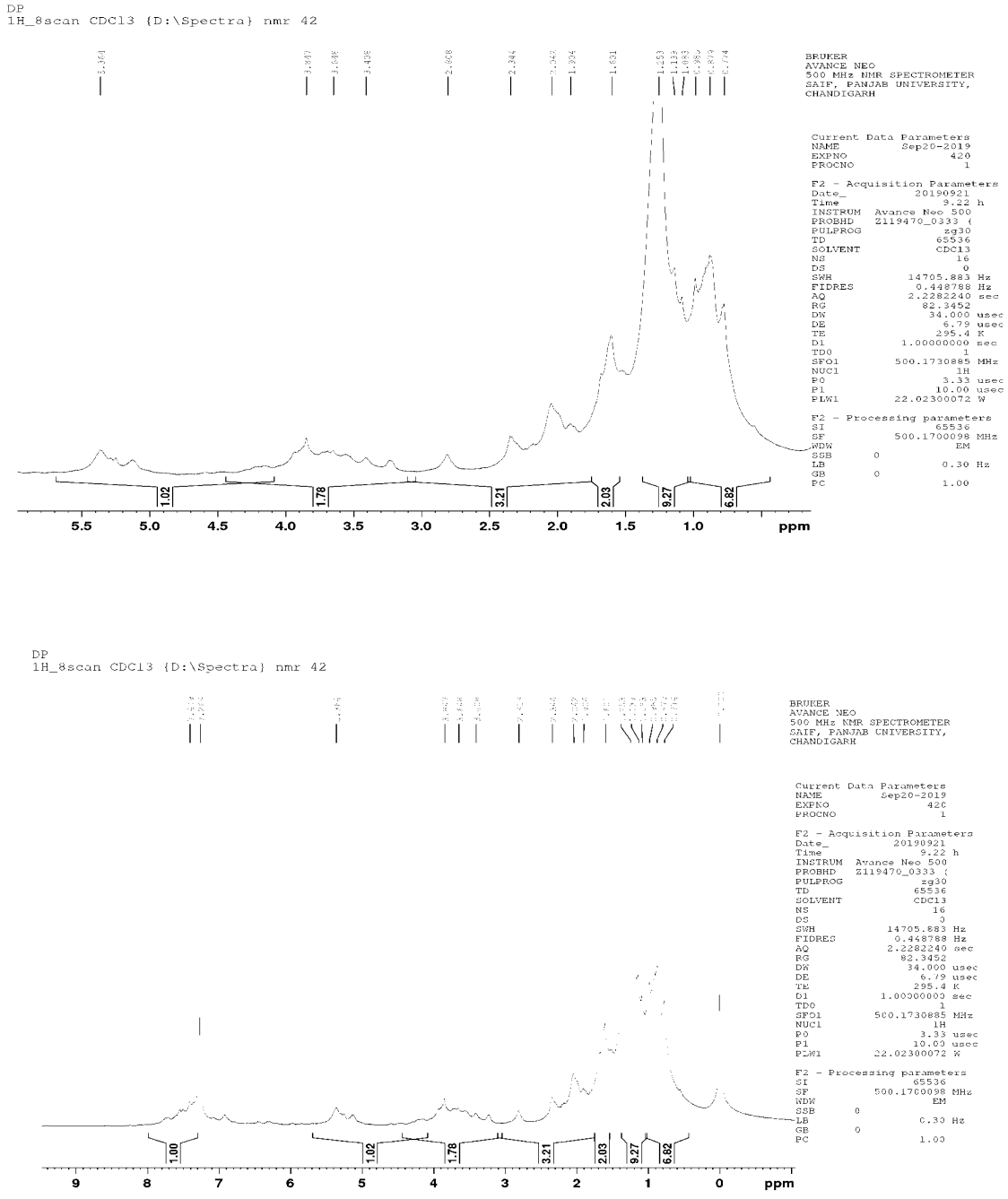

Figure 2: Proton Nuclear Magnetic Resonance (H^1- NMR)

Table 3: Compound retrieved from H^1NMR

S. No.	PPM	Compounds
1	7.409-7.264	Aryl H
2	5.364	Vinylic or amide
3	3.847-3.408	Alcohol
4	2.808-1.904	Alkene carbon
5	1.601-1.253	2H alkyl carbon
6	1.139-0.985	9H alkyl carbon
7	0.879-0.774	6H alkyl carbon

CONCLUSION

The dried powder of Duranta plumieie was extracted successively with solvents of different polarity. The chloroform extract was subjected to phytochemical screening that showed the presence of flavonoids and terpenoids. The chemical compounds were characterized on the basis of FTIR andd H1 NMR spectroscopy. The FTIR analysis of the chloroform extract of the leaves showed the presence of various functional groups such as O-H(Alcohol), CH(sp^2)Alkene, -CH(sp^3)Alkane, C-C (aromatic ring), C=O(Ketones) ,-C-O, C-C bending stretching(Alcohol, acid, ester,ether).The H^1 NMR was used to reveal the number of protons and their electronic state. Further research will be done in near future on different activities such as antibacterial, antifungal, anti-inflammatory, antipyretic for the determination of therapeutic uses of the isolated compounds.

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Conflicts of Interest

There is no conflict of interest

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