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GC–MS studies on the six naphthoyl-substituted 1-n-pentyl-indoles: JWH-018 and five regioisomeric equivalents

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ABSTRACT

The GC–MS properties of the synthetic cannabinoid drug of abuse 3-(1-naphthoyl)-1-pentylindole (JWH-018) and all 5 of its' regioisomeric 1-naphthoyl substituted 1-n-pentylindoles are compared in this report. These compounds have the 1-naphthoyl-group attached at each of the possible substituent positions of the indole ring. The six compounds have the same elemental composition $C_{24}H_{23}NO$ and the same substituents attached to the indole ring. The electron ionization mass spectra showed equivalent regioisomeric major fragment ions resulting from cleavage of the groups attached to the central indole nucleus. The characteristic $(M-17)^+$ fragment ion at *m/z* 324 resulting from the loss of an OH group was significant in the EI-MS of 3-, 4-, 5- and 6-(1-naphthoyl)-1-pentylindole. Fragment ions occurred at *m/z* 127 and 155 for the naphthyl and naphthoyl cations common to all six regioisomeric substances. Indole containing fragments produced the cations at *m/z* 284, 270, 214 and 186. The unique fragment at *m/z* 141 observed in the 1,2- and 1,7-isomers resulted from a rearrangement involving the two indole substituents to yield the $C_{10}H_7CH_2^+$ cation. The major points of EI-MS differentiation of the synthetic cannabinoid JWH-018 from the other five isomers are the high relative abundance of both the *m/z* 144 ion and the *m/z* 324 ion in the JWH-018 spectrum.

GC separations on a capillary column containing a trifluoropropyl methyl polysiloxane (Rtx-200) stationary phase provided excellent resolution of these six compounds. The elution order appears related to the relative distance between the two indole substituents with the lowest retention associated with minimum distance between the groups attached to the indole nucleus.

1. Introduction

The driving force for the continuing development of new designer drugs is often changes in the legal status for a previous generation of analogs. The legal control of designer synthetic cannabinoids of the 1-alkyl-3-acylindole variety included only 5 compounds in the initial group placed under federal control in the U. S. in early 2011 [1]. The indole derivative 3-(1-naphthoyl)-1-pentylindole, JWH-018, was one of the five compounds included in this first group of controlled compounds [1]. Designer exploration of these compounds has produced dozens of new molecules [2–5] in recent years. These next generation compounds have resulted from structural modifications at every region of the original molecule [6] including the alkyl side chain [7,8], the acyl moiety [9,10] and the central indole ring itself [11,12]. Clandestine drug development often migrates toward those designer drugs whose chemical make-up place them just outside the boundary described by the language of the most recent laws. Designer drug development can be viewed as essentially the inverse of ethical pharmaceutical drug development since often the first pharmacological data is from human subjects obtained in hospital emergency rooms.

Drug substances from natural sources such as plant biosynthetic pathways generally occur as a single isomer due to enzymatically controlled processes. Issues of regioisomerism are common in synthetic drug products due to the availability of a wide variety of synthetic precursor chemicals [2,4,5]. These totally synthetic compounds can appear in a number of regioisomeric and isobaric forms and require unique analytical methods for differentiation [13,14]. The development of an isomer specific analytical method often requires information on all the possible isomers.

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Mass spectrometry is the primary method of confirmation of structure for forensic identification and MS based techniques are the required method of analysis by some regulatory organizations. Regioisomeric and isobaric substances are a significant challenge for many analytical techniques used to identify specific substances. Those substances yielding regioisomeric forms of the major mass spectral fragment ions can present additional challenges for forensic analysis. Differentiation among these substances is extremely important when some of these molecules are legally controlled drugs of abuse or controlled precursor substances. Isomer specific forensic analytical methods are of central importance in those drug categories produced by totally synthetic methods.

Many of the 3-acyl-1-alkylindole compounds act as full agonists at both the CB_1 and CB_2 cannabinoid biological receptors [15–17]. JWH-018 has affinity for the cannabinoid brain (CB_1) receptor five times greater than that of THC and has been shown to produce psychoactive effects in animals similar to those of THC [17]. The structures for the regioisomeric indole compounds in this study are shown in Fig. 1. These six naphthoyl substituted 1-pentylindoles have the 1-naphthoyl group attached at each of the possible ring substituent positions of the indole ring. Thus, this specific set of compounds represents those substances yielding regioisomeric forms of the major mass spectral fragment ions.

2. Experimental

2.1. Instrumentation

The GC–MS system consisted of an Agilent Technologies (Santa Clara, CA) 7890A gas chromatograph and an Agilent 7683B auto injector coupled with a 5975C VL Agilent mass selective detector. The GC injector was maintained at 300 °C and the transfer line at 325 °C. The mass spectral scan rate was 2.86 scans/s and the mass spectrometer was tuned using the autotune mode. The GC was operated in splitless mode with a helium (99.999%) flow rate of 0.7 mL/min and the column head pressure was 10 psi. The MS was operated in the electron ionization (EI) mode using an ionization voltage of 70 eV and a source temperature of 230 °C. The GC studies were performed on a 30 m × 0.25 mm i.d. column coated with 0.5 μm 100% trifluoropropyl methyl polysiloxane (Rtx-200) purchased from Restek Corporation (Bellefonte, PA). The separations were obtained using a temperature program consisting of an initial hold at 80 °C for 1.0 min, ramped up to 300 °C at a rate of 30 °C/min, held at 300 °C for 0.5 min then ramped to 340 °C at a rate of 5 °C/min and held at 340 °C for 25.0 min. Samples were dissolved and diluted in high-performance liquid chromatography grade acetonitrile (Fisher Scientific, Fairlawn, NJ) and introduced via the auto injector using an injection volume of 1 μL.

2.2. Synthetic methods

2.2.1. 1-Pentylindole

A mixture of sodium hydride (NaH) in mineral oil and a solution of indole in dimethylformamide were stirred under dry nitrogen for 30 min. 1-Bromopentane was added to the indole/NaH mixture and heated for 1 h then cooled and the mixture poured into water and extracted with methylene chloride. The combined methylene chloride extracts were washed with water, dried and evaporated to yield the desired 1-pentylindole product.

2.2.2. 3-(1-Naphthoyl)-1-pentylindole

A solution of 1-n-pentylindole was dissolved in methylene chloride under nitrogen and added to a three-neck flask. The reaction mixture was cooled and 1.0 M dimethylaluminum chloride in hexane was added via a syringe/septum. A solution of 1-naphthoyl chloride in dry methylene chloride was added over a period of 5 min to the reaction mixture and then stirred over an ice bath under nitrogen. The reaction was quenched by addition of 1 N HCl and the resulting solution extracted with methylene chloride. The combined methylene chloride extracts were washed with water, saturated sodium bicarbonate and dried over potassium sulfate to yield the product, 1-n-pentyl-3-(1-naphthoyl)-indole.

2.2.3. 1-Pentylindole aldehydes

Indoles 3-, 4-, 5- and 7-carboxaldehydes were purchased from Alfa Aesar Chemical Company, Ward Hill, MA. Indoles 2- and

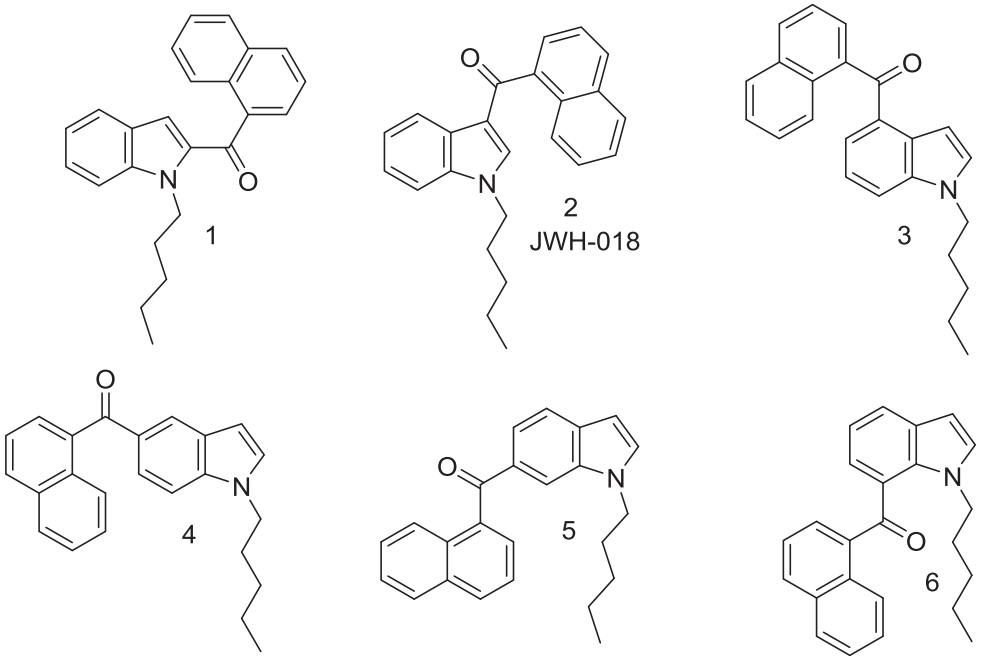

Fig. 1. Structures of the regioisomeric 2-, 3-, 4-, 5-, 6-, and 7-(1-naphthoyl)-1-pentylindoles in this study.