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Chemical characterization of synthetic cannabinoids by electrospray ionization FT-ICR mass spectrometry

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ABSTRACT

The synthetic cannabinoids (SCs) represent the most recent advent of the new psychotropic substances (NPS) and has become popularly known to mitigate the effects of the Δ^9-THC. The SCs are dissolved in organic solvents and sprayed in a dry herbal blend. However, little information is reported on active ingredients of SCs as well as the excipients or diluents added to the herbal blend. In this work, the direct infusion electrospray ionization Fourier transform ion cyclotron mass spectrometry technique (ESI-FT-ICR MS) was applied to explore the chemical composition of nine samples of herbal extract blends, where a total of 11 SCs (UR-144, JWH-073, XLR-11, JWH-250, JWH-122, AM-2201, AKB48, JWH-210, JWH-081, MAM-2201 and 5F-AKB48) were identified in the positive ionization mode, ESI(+), and other 44 chemical species (saturated and unsaturated fatty acids, sugars, flavonoids, etc.) were detected in the negative ionization mode, ESI(−). Additionally, CID experiments were performed, and fragmentation pathways were proposed to identify the connectivity of SCs. Thus, the direct infusion ESI-FT-ICR MS technique is a powerful tool in forensic chemistry that enables the rapid and unequivocal way for the determination of molecular formula, the degree of unsaturation (DBE—double bond equivalent) and exact mass (<1 ppm) of a total of 55 chemical species without the prior separation step.

1. Introduction

The *Cannabis sativa* L. plant has great therapeutic and psychotropic potentials. It contains more than 421 chemical substances, from which more than 60 were reported as cannabinoids, being the Δ^9-tetrahydrocannabinol (Δ^9-THC), the principal cannabinoid found with the highest psychoactive activity [1]. The term cannabinoid is attributed to the group of molecules composed of 21 carbon atoms present in the *C. sativa* L., and their products of transformation [2].

In the last decades, analogous compounds to Δ^9-THC were synthesized in order to explore the endocannabinoid system as having potential psychotherapeutic effects. This fact established the class of synthetic cannabinoids (SCs). The SCs represent the most recent advent of the designer drugs, which are drugs created or structurally modified to difficult their detection from conventional analytical methods, and its prohibition by current legislation, thus facilitating its illicit market [3–5]. The SCs have become popular in the trying of to mitigate the effects of Δ^9-THC, which is one of the most consumed drugs in the world [6].

In Europe, at the beginning of the 21st century, the SCs were freely commercialized, being not prescribed drugs. They were mainly named Spice and K2 and sold as incense and leaves extracts, which having or not metallic packages (Fig. 1) [5].

Currently, the SCs are synthesized in clandestine laboratories mostly in China. Afterwards, they are dissolved in organic solvents (ethanol, acetone or methanol) and sprayed in a dry herbal blend, which can also have intrinsic psychotropic effects. Among the natural products that can be mixed to the SCs, stand out: *Melissa*, *Mentha*, *Thymus*, *Damiana*, *Indian Warrior*, *Lion's Tail*, *Baybean*, *Blue Lotus*, *Vanilla*, and *Honey* [7,8].

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Fig. 1. Examples of SCs samples that are popularly known as bong basic or extremely.

The SCs produce physiologic and psychoactive effects similar to those of Δ^9-THC, but with higher intensity and for a longer time, and that might result in severe damage to human health [5,7]. However, there are still no reports in the literature on lethal doses for many of these new drugs [9].

The main clinic effects in the central nervous system caused by the SCs are convulsions, agitation, anxiety, irritability, sedation, paranoia, and psychosis. In the cardiovascular system the effects are tachycardia, arrhythmia, chest pain, myocardial infarction and arterial hypertension. In the gastrointestinal system, the symptoms include nausea, and vomits. Besides, recreational use might result in an acute kidney damage, hypokalemia, hyperglycemia, dilation of the pupil, hyperthermia, abstinence, and dependence [6].

These substances are partial agonists of the cannabinoids receptors CB1 and CB2. The CB1 receptors are responsible for changes in humor, perception and hearing and visual cognitions. The CB2 receptors stimulate the spontaneous movement of the immune cells and the release of immunomodulatory substances [10].

The SCs have mostly affinity to CB1 receptors than Δ^9-THC, and some also have higher affinity to CB2, such as JWH-015 and JWH-133. On the other hand, the JWH-018 is five times more potent than the Δ^9-THC. Moreover, some cannabinoids which are present in marijuana, such as cannabidiol, have the antagonistic effect to the Δ^9-THC, and are responsible for reducing its psychoactive effects. This fact justifies the higher toxicity, generally registered for SCs users [6,10].

The SCs are liposoluble. There are more than 400 different SCs and they can be classified into six groups, according to the chemical structures of their molecules [6,10,11]. The main principal classes of SCs are: (i) classical cannabinoids (HU-210, AM-906, O-584 and O-1315)—they have chemical structure similar to that of Δ^9-THC, however, their psychotropic effects are higher (from 100 to 800 times) [6,11]; (ii) non-classical (CP-47,497-C8, CP-55,940, CP-55,244) [11]; (iii) hybrids (AM-4030)—a combination of classic and non-classic SCs [11]; (iv) aminoalkylindoles (AAI), in which the SCs can be subdivided in naphthoylindoles (JWH-018, JWH-073, JWH-398, JWH-122, JWH-210, JWH-081 and JWH-200), phenylacetylindoles (JWH-250 and JWH-251); naphthylmethylindoles and benzoylindoles (AM-694 and RSC-4); and cyclopropoylindoles (UR-144, XRL-11, AB-005) [11–13]; (v) eicosanoids: include compounds as anandamide and its analogous [11]; and (vi) other classes of SCs such as diarylpyrazoles, naphthoypyrroles, naphthylmethylindenes and indazole 3-carbonyl derivates (THJ-2201) [11,13]. The chemical structures of the main SCs classes are shown in Table 1 [11].

Because the SCs have a high risk for the health of the people due to their potent psychotropic effects and considering the growth of the number of users, several analytical methods [14–26] are being employed for SCs detection. They are Fourier transform infrared spectroscopy [15], capillary electrophoresis (CE) [15], liquid chromatography–mass spectrometry (LC–MS) [15–17,20], gas chromatography–mass spectrometry (GC–MS) [19,20,25], GC with flame ionization detector (GC-FID) [20], high performance liquid chromatography (HPLC) [21], thin layer chromatography (TLC) [21], matrix-assisted laser desorption ionization mass spectrometry (MALDI-MS) [22], flowing atmospheric-pressure afterglow ion source for mass spectrometry (FAPA-MS) [27], direct analysis in real time mass spectrometry (DART-MS) [23], paper spray mass spectrometry (PS-MS) [26], and nuclear magnetic resonance (NMR) spectroscopy [15,24]. Among the main SCs identified are: JWH-018 [16–22], JWH-073 [18–22], JWH-081 [21,22], JWH-200 [18,21], JWH-210 [21,22], AM-2201 [21,24], JWH-015 [23], JWH-019 [21], JWH-122 [27], AM-694 [20], JWH-019 [22], JWH-250 [20,21], RCS-8 [21], UR-144 [25], and XLR-11 [25]. Additionally, the Duquenois–Levine color test, which is used to identify Δ^9-THC, provides negative results for the SCs [13].

Most of the analytical methods employed to identify and quantify the SCs use hyphenated techniques, which is the combination of separation and detection methods [16–21]. It is due to the high complexity of the organic matrix, which is composed of natural products. Besides, a great number of SCs active ingredients can be present [15].

Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) combined with a direct infusion electrospray ionization (ESI) provides a molecular detailed view of complex mixtures, especially for polar compounds, due to its unsurpassed ultra-high resolution and accuracy. The high accuracy mass measurements enabling assign molecular formula ($C_cH_hN_nO_o$),