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Short Communication

# DART – LTQ ORBITRAP as an expedient tool for the identification of synthetic cannabinoids

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ABSTRACT

Synthetic cannabinoids as designer drugs constitute a major problem due to their rapid increase in number and the difficulties connected with their identification in complex mixtures. DART (Direct Analysis in Real Time) has emerged as an advantageous tool for the direct and rapid analysis of complex samples by mass spectrometry. Here we report on the identification of six synthetic cannabinoids originating from seized material in various matrices, employing the combination of ambient pressure ion source DART and hybrid ion trap - LTQ ORBITRAP mass spectrometer. This report also describes the sampling techniques for the provided herbal material containing the cannabinoids, either directly as plant parts or as an extract in methanol and their influence on the outcome of the analysis. The high resolution mass spectra supplied by the LTQ ORBITRAP instrument allowed for an unambiguous assignment of target compounds. The utilized instrumental coupling proved to be a convenient way for the identification of synthetic cannabinoids in real-world samples.



## 1. Introduction

Designer drugs have been the cause of growing concern since the turn of the century, prominent among them being synthetic cannabinoids and cathinone derivatives. Every year, new synthetic analogs emerge [1–3]. Recently, new herbal products containing synthetic cannabinoids appeared on the market. This substance class first emerged in herbal products in the mid-2000s as legal alternative of marihuana. In Europe, over 130 different synthetic cannabinoids have been detected in recent years, according to the European Monitoring Centre for Drugs and Drug Addiction (EMCDDA). Solely in 2014, thirty new synthetic cannabinoids have been identified and registered within the EU Early Warning System [4].

As the term 'legal highs' suggests, the novelty of these substances, alongside with the rate of emergence and structural diversity, makes their detection as well as their legal control more difficult, increasing the demand for rapid and easy-to-use analytical techniques for their detection and identification.

These substances are usually sold as herbal smoking blends, mimicking the effects of $\Delta^9$-THC, the main active component of cannabis. Many of them also show a higher potency compared to $\Delta^9$-THC, along with longer biological half-lives and higher adverse effects. They are often marketed via internet and street shops under various names, such as 'incense', K2 or 'Spice' [3,5]. The marketed drugs are prepared by spreading the solution of the cannabinoid on the surface of dried plant material and subsequent evaporation of the solvent [6,7]. The botanical composition of the carrier plants usually shows a great variability as no quality control can be expected with illicit manufacturing. So does the dosage, even within one production charge. Also, different plant parts (e.g., stem or leaf) show different content of the active ingredient because of the specific adsorption properties of the particular herbal matrix. Sometimes several different cannabinoids (or other drugs as well) can be found in one sample simultaneously [8]. Given the little amount of knowledge concerning the pharmacology and toxicology of the newly introduced compounds, they constitute a major source of potential health risk for the recipients [9,10].

Numerous methods for the analysis of cannabinoids have been developed, facilitating the identification and quantification of these substances and their metabolites in sample matrices with complex composition, as well as in body constituents [11,12]. The cannabinoids are routinely analyzed by GC–MS and LC–MS methods which are well-established and reliable but often necessitate elaborate sample pre-treatment and processing [13–16].

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The DART (Direct Analysis in Real Time) ion source allows for a direct ionization of the target compounds in various sample types under ambient conditions [17–20]. The samples can be in solid or liquid form and they can be analyzed in their native state without the necessity of further sample processing. The instrument can work with high-concentration samples as well. The samples are placed between the outlet of the ion source and the inlet of the spectrometer. Inside the ion source, the interaction between the molecules or atoms of an inert gas (typically He or N) and an electric glow discharge generates a stream of metastable atoms leaving the ion source and interacting with the sample molecules and ionizing them prior to their entrance to the mass spectrometer.

The experimental arrangement produces mostly single-charged ions of the type $[M+H]^+$ (or $[M-H]^-$ in negative-ion mode); multiple charged ions are not observed. The ionization occurs at the surface of the sample and the analysis can proceed even with high-concentration samples. Several parameters – type of gas, temperature, electrode potential, sampling mode, width of the sample gap (distance outlet-inlet) – can be varied and influence the outcome of the ionization. DART based analytical methods have been increasingly employed in bioanalysis, such as the analysis of drugs in different matrices [21] or the analysis of cannabinoids with the aid of a time-of-flight (TOF)-MS system [22].

The hybrid ion trap - LTQ ORBITRAP mass spectrometer used for the detection of the ions generated by the DART ion source combines high resolving power with outstanding mass accuracy [23,24]. This ability to discern ions with closely adjoining mass-to-charge ratios ($m/z$) permits the identification of analytes even in mixtures of high complexity. The detection of specific diagnostic fragments of a precursor ion obtained by $MS^n$ technique allows for further confirmation of the presence of a particular compound. The application of ORBITRAP technology in the area of forensic analysis has been on the rise over the last few years [25].

Centre of Excellence of Security Research originated by joint efforts of the Institute of Forensic Science and academic institutions in Slovakia. The Institute of Forensic Science in Slovakia maintains forensic expert laboratories providing services for law enforcement authorities in Slovak republic. Within this cooperation framework we received samples of seized material from 'crazy shops' in Slovakia. The aim of our study was to test the possibility to employ the DART ionization source in combination with the ORBITRAP spectrometer for the screening of synthetic cannabinoids, under different experimental conditions.

## 2. Materials and methods

### 2.1. Tested samples

The 8 samples included herbal material (leaves, stems, flowers) soaked with the drugs and solid powdered samples of pure cannabinoids were obtained from Institute of Forensic Science in Bratislava. Several types of analyte were used: plant material soaked with the synthetic cannabinoids, extracts from this plant material in methanol, and methanol solutions prepared by dissolution of powder samples of cannabinoids.

### 2.2. DART-MS analysis

For the analysis the DART – LTQ ORBITRAP combination was used. The DART-SVP ion source from IonSense was operated with the following parameters: positive ion mode, grid electrode voltage 150 V, helium gas with a flow rate 3 L/min, heater temperature 400 °C. The Thermo Scientific™ LTQ Orbitrap XL™ Hybrid Ion Trap-Orbitrap Mass Spectrometer was used in the full-scan mode ($m/z$ range 50–500). The distance between the outlet of the DART ion source and the inlet of the mass spectrometer was held at 30 mm. The sampling time was 30 s.

The solid herbal material was measured directly by holding it with tweezers between the DART ion source (can be held manually or fixed) and the mass spectrometer inlet (Fig. 2). The various types of herbal matrices (leaf, stem, and flower) were each measured separately. Extracts from the herbal material were prepared by extracting 0.1 g of the dry solid with 3 ml methanol. The mixture was vortexed for 3 min and filtered to provide a clear, slightly yellow solution. We used two kinds of liquid sample introduction: introduction by means of capillaries (Quick Strip Sample Cards) or metal grid as a sample carrier. In both cases automation is possible by means of a programmable rail. Finally, powdered samples of the pure cannabinoids were solubilised in methanol to yield solutions with different concentrations (0.5–100 ppm) and measured as described above.

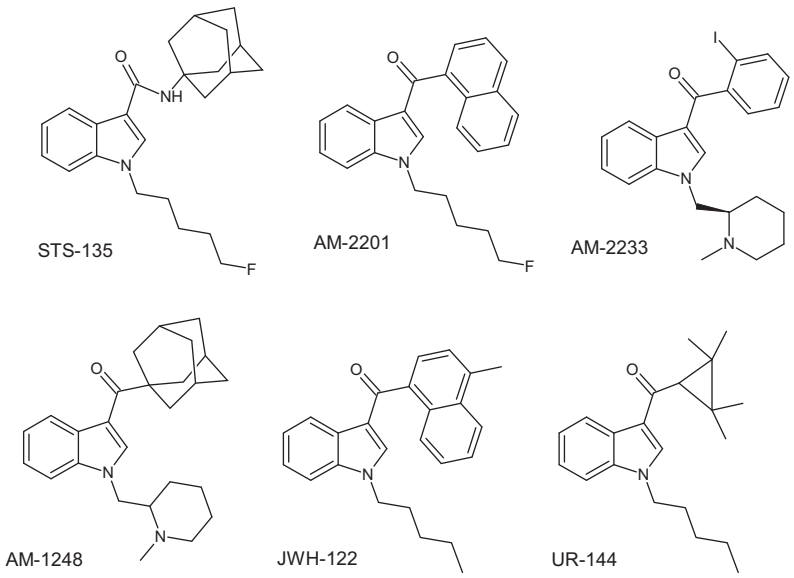


**Fig. 1.** Molecular structures of target analytes (synthetic cannabinoids).