ScienceDirect

Legal Medicine

Volume 50, May 2021, 101869

Determination of AB-FUBINACA and 5F-NPB-22 in rats exposed to “*Bonsai*” via inhalation and analysis of seized product

Pinar Efeoglu Ozseker [a], Nebile Daglioglu [a], Mete Korkut Gulmen [a], Ilknur Tolunay [b], Fehiman Efeoglu [a]

Show more

https://doi.org/10.1016/j.legalmed.2021.101869

Get rights and content

Highlights

- The majority of cannabinoid users prefer to consume cannabinoids like they smoking cigarettes.
- Type of synthetic cannabinoid in “*Bonsai*” was detected.
- The dose dependent accumulation increased in the lungs of rats exposed acutely to “Bonsai”
- They are also likely to have kidney damage.
- Sensitive analytical methods and an extraction procedure were developed and validated.

- These methods can be implemented in forensic and clinical toxicology laboratories.

Abstract

Synthetic cannabinoids (SCs) are the most rapidly growing class of recreational designer drugs. Illicit drug manufacturers began to produce herbal smoking materials under a variety of brands names, e.g. “Spice, K2, Bonsai, Yucatan Fire”. They were appeared on the European market in 2008. In this study, types of SCs in the herbal product sold as “*Bonsai*” in Turkey were determined and the identification of these substances in biological samples collected from rats depending on the inhalation of different amounts of plant material were aimed.

To determine the SC species in the content of the plant product, analysis was performed via gas chromatography-mass spectrometry. Liquid-liquid extraction methods were utilized for blood and organ samples, while solid-phase extraction with β-glucuronidase enzyme treatment was applied for urine sample preparation. The relationship between the amount of burned plant and the amount of SCs accumulated in the blood, urine and organ samples of rats exposed to the plant product by inhalation was examined by liquid chromatography-tandem mass spectrometry.

AB-FUBINACA and 5F-NPB-22 were detected in the herbal product. A significant correlation was found between the amount of herbal product inhaled and the prevalence of SCs, especially in lung tissues while no SCs were detected in the blood and urine samples of rats.

There is currently no study on biological samples of individuals exposed to herbal products containing SCs by inhalation. Regarding the findings obtained in this study, the overall increase in the amounts of herbal product inhaled was demonstrated to pose a potential risk to humans.

Introduction

Synthetic cannabinoids (SCs) are chemical substances that show cannabis-like psychoactive effects by activating cannabinoid receptors known as CB1 and CB2 [1]. Their chemical structures are unlike that of tetrahydrocannabinol, which is the main psychoactive component of marijuana. However, the binding affinity of SCs to cannabinoid receptors is approximately 100–800 times higher than that of THC [1], [2].

These compounds were referred to as legal highs, herbal highs, designer drugs, or herbal incense to obtain legal status when they first appeared on the market [3], [4]. Most of these chemicals are produced in China and dissolved in solvents, such as acetone, ethanol and methanol. Then, they are sprayed on herbs, such as lemon balm, oregano, mint, damiana or veronica. After this process, the plant material is dried in the shade and packaged for sale [3], [5]. These green/brown plant materials are usually finely cut and supplied in quantities of 0.5–3 g in colorful and professionally designed packages. These packages are marked as “not for human consumption”, “incense,” and “only for aromatherapy”. It is claimed on the packaging labels that these herbs can have cannabis-like effects when consumed. These products are said to be pharmacologically active only because of the sophisticated mixture of natural elements in the material [5], [6], [7].

SCs are lipophilic substances, just as THC is. After inhalation, SCs are absorbed through the lungs, rapidly spread and accumulate in fat tissues. This condition causes a rapid decrease in the concentration of the precursor drug in the blood after exposure. Saito et al. reported the accumulation of some SCs, such as MAM-2201, in adipose tissue due to their high lipophilicity [8], [9]. They can cross the blood-brain barrier and accumulate in brain tissues [10].

Acute toxicity is a common finding and often shows a combination of serious psychiatric and medical effects, such as anxiety, agitation, psychosis, and tachycardia [11], [12]. Lip dryness, dizziness, and headache are among the noted physiological side effects [1], [13]. Other undesirable effects are sweating, chills, dystonia and difficulty breathing [14]. Serious cases of toxicity requiring emergency treatment associated with SCs include acute renal failure, seizures and myocardial infarction [15], [16]. In addition, the use of SCs may also cause death [17], [18].

Over the last few years, new generation SCs have emerged, producing cannabis-like effects in humans, such as PB-22, 5F-PB-22, XLR-11, AKB-48, UR-144, AB-PINACA and AB-FUBINACA [19]. In Turkey, AB- FUBINACA (N-[(1S)-1-(aminocarbonyl)-2-methylpropyl]-1-[(4-fluorophenyl) methyl]-1*H*-indazole-3 carboxamide) and 5F-NPB-22 (1-(5-fluorophenyl)-8-quinolinyl ester-1*H*-indazole-3 carboxylic acid) are widely available.

The majority of cannabinoid users prefer to intake these substances by smoking, as this rapidly initializes their pharmacological effects. Most of these substances evaporate without decomposition when they are smoked due to their high lipophilicity [20]. However, most studies in the literature are based on intravenous applications [21], [22].

In this study, we aimed to determine the chemical content of the herbal product sold as “*Bonsai*” in Turkey and identify the SCs in biological samples of rats exposed to the herbal product through inhalation.

Section snippets

Chemicals and reagents

AB-FUBINACA, AB-FUBINACA metabolites 2A and 2B and JWH-073-d7 were purchased from Cayman Chemicals (Ann Arbor, USA). 5F-NPB-22 was supplied by Chiron (Norway). Fig. 1 shows the chemical structures of AB-FUBINACA, AB-FUBINACA metabolites 2A and 2B, 5F-NPB-22 and JWH-073-d7.

Acetonitrile, methanol, ethyl acetate, and water of LC/MS grade (for chromatography) were obtained from Merck, Germany. Ammonium formate, potassium dihydrogen phosphate monohydrate (KH_2PO_4) and β-glucuronidase (Sigma Aldrich,…

Herbal analysis

The herbal product sold under the name “*Bonsai*” was obtained from the Adana Criminal Police Department. It was prepared by weighing approximately 50 mg of material that was transferred to a glass tube (10×160 mm), and 5 mL of methanol was added. The mixture was incubated for 72 h with a shaker. After incubation, the solution was filtered through a 0.22 µm membrane filter and injected into the GC–MS for analysis.…

Extraction of blood, urine, and organ samples

The liquid-liquid extraction method was chosen for the preparation of blood…

Method validation

For analysis, linear regression and $1/x^2$ weighting were applied and reported as the correlation coefficient R2. The linearity was assessed using calibration standards over the range of 0.5–10ng/mLfor whole blood, 1–50ng/mL for urine and 2–60ng/g for tissue samples. The R2 value for each curve was 0.99 or better. The extraction recoveries were between 72 and 103% for blood, 75 and 97% for urine and 70 and 96% for lung tissue. The retention times and ion ratios were observed to confirm the…

Discussion

SCs, which are increasingly used by young people and adults, are an important public health problem and they have recently become an analytical challenge for forensic, clinical, and workplace drug testing laboratories. However, although there are many records demonstrating the use of SC-containing herbal products, there is no systemic laboratory study on humans due to the potential toxic concerns associated with these chemicals. And also there has been very few information on the…

Conclusion

The types of SCs in seized products seem to be diversifying, and more serious health risks will be associated with their use than ever before. Therefore, the constant of monitoring of these substances remains necessary in clinical and forensic context. Chemicals sprayed on herbal products cannot be detected in systemic toxicologic analyzes. The lack of metabolic standards prevents the development of standardized methods. In this study, both extraction and sensitive analytical methods have been…

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.…

Acknowledgements

Funding: This work was supported by the Cukurova University Scientific Research Projects Commission (project no.: TF2013D5).…

Ethical approval

All procedures performed in studies involving animals were under the ethical standards of the Cukurova University Health Sciences Experimental Application and Research Center at which the studies were conducted (Ethics Committee Approval Number: 6)….

Recommended articles

References (32)

I. Vardakou *et al.*
Spice drugs as a new trend: mode of action, identification and legislation
Toxicol. Lett. (2010)

R. Lindigkeit *et al.*
Spice: a never ending story?
Forensic Sci. Int. (2009)

G. McLachlan
Taking the spice out of legal smoking mixtures
Lancet (2009)

N.E. Camp
Synthetic cannabinoids
J. Emerg. Nurs. (2011)

J.L. Poklis *et al.*
Detection and disposition of JWH-018 and JWH-073 in mice after exposure to “Magic Gold” smoke
Forensic Sci. Int. (2012)

R. Vandrey *et al.*
A survey study to characterize use of spice products (synthetic cannabinoids)
Drug Alcohol Depend. (2012)

L.M. Labay *et al.*
Synthetic cannabinoid drug use as a cause or contributory cause of death
Forensic Sci. Int. (2016)

J.B. Zawilska *et al.*

Next generation of novel psychoactive substances on the horizon – a complex problem to face

Drug Alcohol Depend. (2015)

A.H. Lichtman *et al.*

The pharmacological activity of inhalation exposure to marijuana smoke in mice

Drug Alcohol Depend. (2001)

J.M. Wiebelhaus *et al.*

Inhalation exposure to smoke from synthetic “marijuana” produces potent cannabimimetic effects in mice

Drug Alcohol Depend. (2012)

H. Kakehashi *et al.*

Effects of lipophilicity and functional groups of synthetic cannabinoids on their blood concentrations and urinary excretion

Forensic Sci. Int. (2020)

E.W. Gunderson *et al.*

A survey of synthetic cannabinoid consumption by current cannabis users

Subst. Abuse (2014)

EMCDDA 2016 Report. European Drug Report: Trends and Developments,...

UNODC

Synthetic Cannabinoids in Herbal Products

(2011)

S. Kneisel *et al.*

Analysis of synthetic cannabinoids in abstinence control: long drug detection windows in serum and implications for practitioners

Drug Test. Anal. (2014)

T. Saito *et al.*

A fatal case of MAM-2201 poisoning

Forensic Toxicol. (2013)

There are more references available in the full text version of this article.

Cited by (0)

View full text