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Pharmacokinetics of (synthetic) cannabinoids in pigs and their relevance for clinical and forensic toxicology

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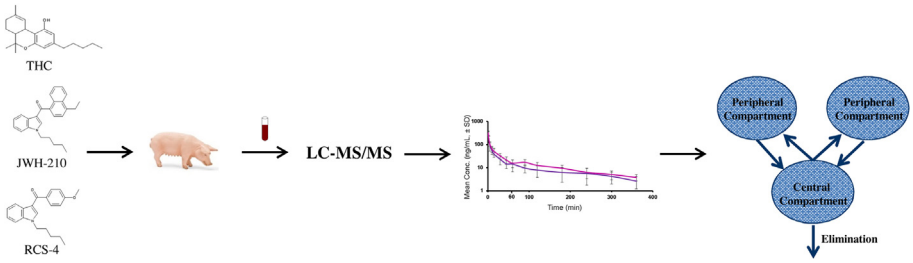
HIGHLIGHTS

- First study on the pharmacokinetics of JWH-210 and RCS-4 in pigs.
- A three-compartment model described best pharmacokinetic data of THC, JWH-210, and RCS-4.
- The allometrically upscaled THC pig model resulted in successful prediction of human exposure.
- Pigs useful for prediction of human pharmacokinetics of synthetic cannabinoids.

GRAPHICAL ABSTRACT



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ABSTRACT

Synthetic cannabinoids (SCs) are gaining increasing importance in clinical and forensic toxicology. They are consumed without any preclinical safety studies. Thus, controlled human pharmacokinetic (PK) studies are not allowed, although being relevant for interpretation of analytical results in cases of misuse or poisoning. As alternative, in a controlled animal experiment, six pigs per drug received a single intravenous dose of 200 μg/kg BW each of Δ^9-tetrahydrocannabinol (THC), 4-ethylnaphthalen-1-yl-(1-pentylindol-3-yl)methanone (JWH-210), or 2-(4-methoxyphenyl)-1-(1-pentyl-indol-3-yl)methanone (RCS-4). In addition, six pigs received a combination of the three drugs with the identical dose each. The drugs were determined in serum using LC–MS/MS. A population (pop) PK analysis revealed that a three-compartment model described best the PK data of all three cannabinoids. Central volumes of distribution were estimated at 0.29 L/kg, 0.20 L/kg, and 0.67 L/kg for THC, JWH-210, and RCS-4, respectively. Clearances were 0.042 L/min/kg, 0.048 L/min/kg, and 0.093 L/min/kg for THC, JWH-210, and RCS-4, respectively. The popPK THC pig model was upscaled to humans using allometric techniques. Comparison with published human data revealed that the concentration-time profiles could successfully be predicted. These findings indicate that pigs in conjunction with PK modeling technique may serve as a tool for prediction of human PK of SCs.

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1. Introduction

Synthetic cannabinoids (SCs) have emerged on the drugs of abuse market sold via internet as herbal mixtures or incenses, often in unknown and unexpected compositions and concentrations. Due to the fact that many of these drugs have a more potent binding strength to cannabinoid receptors than Δ^9-tetrahydrocannabinol (THC) (Huffman et al., 2003; Makriyannis et al., 2001), a consumption can lead to strong psychoactive and unpredictable toxic effects. Numerous cases of intoxications have been reported with life-threatening conditions accompanied e.g. by tachycardia, somnolence, anxiety, nausea, vomiting, seizures, hyperglycemia, hypokalemia, agitation, hallucination, and acute psychosis resulting in admission to hospital (Harris and Brown, 2013; Hermanns-Clausen et al., 2013; Kronstrand et al., 2013; Schneir and Baumbacher, 2012), or even in death (Behonick et al., 2014; Kronstrand et al., 2013; Patton et al., 2013; Schaefer et al., 2013; Shanks et al., 2012). Therefore, SCs are gaining an increasing importance in clinical and forensic toxicology. For interpretation of analytical data of impaired or poisoned persons concerning e.g. time of intake or concentration at a particular time relevant for legal reasons, pharmacokinetic (PK) data are necessary. However, respective data of controlled studies are not available as these compounds are sold and consumed without safety pharmacological tests. Only data from biotransformation studies (Grigoryev et al., 2011; Hutter et al., 2012; Kavanagh et al., 2012), single case reports (Dresen et al., 2011; Hermanns-Clausen et al., 2013; Kronstrand et al., 2013), or self-experiments have been reported so far. A patient participating in an Intramural Research Board approved research study smoked an herbal incense that contained 17 mg/g JWH-018 and 22 mg/g JWH-073 and blood samples were taken after 19, 53, 107, and 199 min. Peak whole blood JWH-018 and JWH-073 concentrations of about 5 ng/mL were detected after 19 min. After 199 min, concentrations had decreased below 1 ng/mL (Kacinko et al., 2011). In a self-experiment two subjects (one male and one female) smoked a cigarette containing 100 (volunteer one) and 150 (volunteer two) mg of the incense "Smoke". This incense was found to contain JWH-018 and the smoked dose was equal to an approximately 50 μg/kg BM dose of JWH-018. Blood samples were drawn 5, 15, and 60 min as well as 3, 12, 24, and 48 h after the consumption. Peak serum JWH-018 concentrations of about 10 ng/mL were found 5 min after administration and traces were still present after 48 h (Teske et al., 2010). In the third study, a self-experiment was conducted by oral administration of a gelatin capsule containing 5 mg of AM-2201 and serum and urine specimens were collected for 11 days. A peak serum AM-2201 concentration of 0.56 ng/mL was determined 1 h and 35 min post-ingestion and the drug remained detectable for 5 days (Hutter et al., 2013). Nevertheless, due to the limited number of sampling points and a very small collective of subjects, these studies provide only insufficient information of SCs PK and should be supplemented by systematic studies, including a larger number of individuals.

As systematic controlled human studies have not been performed, PK properties should be assessed in controlled animal studies. There is only one published controlled animal study providing PK properties of the SC WIN 55,212-2. The substance was administered as a 150 μg/kg intravenous (i.v.) dose to seven guinea pigs and plasma samples were obtained for 8 h (Valiveti et al., 2004). Experiments using small animals such as rodents are hampered by their little blood volume, not allowing for multiple blood sampling. As a consequence, a larger number of animals would be needed and complete kinetics could not be elucidated in the same animal. Pigs as a large mammalian species, however, allow for clarification of different issues in the same animal. They are suitable for extensive specimen sampling. Furthermore, pigs are closely related to the human species in terms of e.g. metabolism including cytochrome P450 (CYP) enzyme pattern (Anzenbacher et al., 1998), anatomical structures as well as physiological properties regarding e.g. cardiovascular, urogenital, and digestive systems (Bode et al., 2010; Swindle et al., 2012). In addition, they are omnivores, sensitive to a wide range of drugs and chemicals, and all routes of administration are possible using the pig (Bode et al., 2010; Svendsen, 2006; Swindle et al., 2012). Thus, alternatively to dogs and monkeys, pigs are increasingly used in preclinical toxicological testing of pharmaceuticals (Swindle et al., 2012) and they are a common model in pharmacological studies, especially to assess PK properties of substances (Mogi et al., 2012; Shimshoni et al., 2015; Sjögren et al., 2012).

SCs are closely related to THC and PK of THC has already extensively been described in the literature using different animal models (Garrett and Hunt, 1977; Leuschner et al., 1986) and also in human studies (Huestis et al., 1992; Hunt and Jones, 1980; Lindgren et al., 1981; Ohlsson et al., 1982) after different routes of administration. Recently, Brunet et al. (2006, 2010) using a small number of animals and a non-compartmental PK approach developed a pig model, which is suitable for cannabinoid PK studies after i.v. administration. They suggested that the animal data can be compared to findings of controlled human studies (Huestis, 2002; Huestis et al., 1992). Therefore, the aim of the present study was to elucidate whether domestic pigs can be used for prediction of human PK of SCs. For this purpose, we determined in a first step the concentration-time profiles after i.v. administration of the two selected SCs 4-ethylnaphthalen-1-yl-(1-pentylindol-3-yl)methanone (JWH-210) and 2-(4-methoxyphenyl)-1-(1-pentyl-indol-3-yl)methanone (RCS-4) to domestic pigs in comparison to that of THC. In a second step, we modeled the concentration-time profiles and assessed whether this model can predict published THC data in humans.

2. Materials and methods

2.1. Chemicals and reagents

The used chemicals and reagents are listed in the Electronic Supplementary material (S1).

2.2. Animals

As already described in a previous study (Schaefer et al., 2015), all experiments were performed in accordance with the German legislation on protection of animals and the National Institutes of Health Guide for the Care and Use of Laboratory Animals (permission number: 69/2013). Twenty eight domestic pigs (Swabian Hall strain; mean body weight 47.7 ± 6.4 kg) were used for the study. The animals had free access to tap water and daily standard chow. They were kept fasting a night before the experiment with free access to water.

2.3. Surgical procedures

The surgical procedures have already been described elsewhere (Schaefer et al., 2015) and are described in the Electronic Supplementary material (S2).

2.4. Study design

The study included five different groups. Pigs of the groups 1–3 (n = 6 each) received the respective drug by a single administration (200 μg/kg BW each), pigs of group 4 (n = 6) a