Fentanyl analytics in a case of fatal misuse of transdermal fentanyl

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Abstract

A 54-year old male (90 kg) was found in his bedroom with a fentanyl transdermal patch on his penis. With low vital reactions, the dormant man was immediately brought to hospital, where he died shortly. Systematic toxicological analysis confirmed a fatal poisoning with fentanyl. The fentanyl concentration in blood was 35.1 µg/L (LC-MS-MS). The presented case demonstrated the possibility of fentanyl misuse with a transdermal patch at a place of possibel high resorption and fatal outcome. Death due to the application of fentanyl transdermal system which was patched on the penis has not been previously reported.

Introduction

Fentanyl (**F**) is an opioid analgesic, first synthesized in the late 1950s with an analgesic potency of about 200 times higher than that of morphine. Today, **F** is extensively used for anesthesia and analgesia.

Fentanyl transdermal patches (e.g. *Fentanyl Sandoz Transdermales Pflaster* (by Sandoz Pharmaceuticals, Ismaning, Germany), *Fentanyl-CT Matrixpflaster* (by CT Arzneimittel, Berlin, Germany), *Fentanyl STADA Matrixplaster* (by STADA Arzneimittel AG, Bad Vibel, Germany) or *Durogesic Fentanyl Transdermal System* (by Janssen Pharmaceutica Products L.P., Titusville, NJ, USA)) are used in chronic pain management. Durogesic patches work by releasing **F** into subcutaneous fats, which then slowly release the drug into the blood stream over 72 hours, allowing for long lasting relief from pain. In the past few years, the patches have gone generic and are available for lower costs. Durogesic is manufactured in five patch sizes, e.g.: 12 µg/h, 25 µg/h, 50 µg/h, 75 µg/h, and 100 µg/h [1]. Dosage is based on the size of the patch. In general, the rate of absorption is dependent on a number of factors, among those, body temperature, skin type and placement of the patch can have major effects [2]. The different delivery systems used by different producers also affect individual rates of absorption. Each patch contains 100-fold more drug than what is stated on the label in order to create the gradient required to deliver the stated amount. This means a patch that contains enough fentanyl could provide a potentially lethal dose.

The plasma protein binding of **F** is 80-85 %. Fentanyl is mainly metabolised via CYP3A4 [3]. The volume of distribution of **F** is 3-8 L/kg, the elimination half-life approx. 7 h (range 3 -12 h). The chemical structure of **F** is given in Fig. 1.

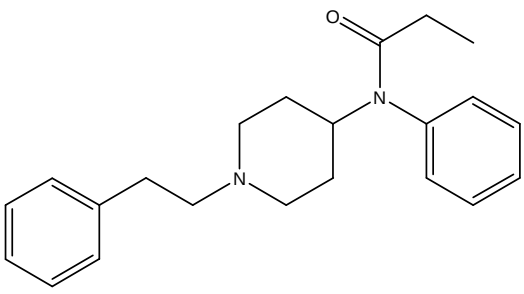

Figure 1: Chemical structure of fentanyl (N-(1-(2-phenylethyl)-4-piperidinyl)-N-phenyl-propanamide)

The therapeutic concentration range of **F** is 1-2 µg/L and the toxic concentration range 2 - 20 µg/L, respectively [4]. Adverse effects such as nausea, confusion, vomiting, somnolence, sweating, and hypoventilation have been reported [1]. Quantification of **F** based on different chromatographic methods, e.g. HPLC-MS [5], LC-MS-MS [6, 7] or GC-MS [8] is possible.

Case history

A doctor prescipted fentanyl patches to a 54-year-old male (90 kg) who was suffering from chronic back pain. He recieved analgetics for a long period of time in addition to the **F** transdermal system. On the evening before his death, the man placed the transdermal system (75 µg **F**/h) on his penis, went to bed and slept throughout the next day. In the following evening, while the patient was still lying in bed, his wife called the emergency doctor. With low vital reactions, the dormant man was immediately brought to hospital, where he died shortly. Signs of intoxication were found at the autopsy.

Materials and methods

Standards and reagents

Sodium hydroxide (p.a.), ammonium acetate (p.a.) were obtained from Merck (Darmstadt, Germany), n-butylchloride (p.a.) was from Fluka (Neu-Ulm, Germany), methanol (LC-MS-Chromasolv) and formic acid (>98%, p.a.) were obtained from Riedel de Haen (Seelze, Germany). Fentanyl citrate was from Sigma (Seelze, Germany) and fentanyl-D5 citrate (purity 99 %) was from Cerillilant (Round Rock, TX, USA).

Extraction procedure

A total of 500 µL blood, 100 µL sodium hydroxide solution (0.1 N) and 400 µL n-butylchloride (which contained the internal standard, fentanyl-D5, c=20 ng/mL) was mixed in a 1.5-ml polypropylene cup for 5 min and then centrifuged for 2 min at 16,000 x g. The upper layer (200 µL) was transferred into a 1.5-ml polypropylene cup and evaporated to dryness under a nitrogen stream (30°C). The dry residue was reconstituted in 100 µL methanol/1% formic acid (25/75, v/v) mixture and transferred into a vial for analysis by LC-MS-MS.

Instrumentation

The HPLC was equipped with a binary pump (LC-10 ADVp), a system controller (SCL-10 A), a solvent degasser (DGU-14 A), an autosampler (SIL-10 ADVp), an oven (CTO-10 ASVp) and an UV-Detector (SPD-6 A) all from Shimadzu, Duisburg, Germany. An analytical Luna 5u C8(2) column (30x3.0 mm, 5 µm), protected by a C18 security guard column (both from Phenomenex, Aschaffenburg, Germany) was used. The oven temperature was kept at 25 °C.

The mobile phase consisted of a mixture of methanol/0.1% formic acid containing 10 mM ammonium acetate (A) and methanol/0.1% formic acid (10/90, v/v) (B). At a flow rate of 0.55 mL/min, a gradient elution was as follows: 0 – 2.0 min: 95 – 6% B linear, 2.0 – 2.5 min: 6% B, 2.5 – 2.9 min: 6 – 95% B linear, 2.9 – 3.5 min: 95% B. The injection volume was 25 µL.

The detection was performed on a triple-stage quadrupole (API 3000, Apperla, Germany) equipped with an electrospray ionisation (ESI) probe in positive polarity mode. Spray voltage was 5500 V, capillary temperature 350 °C. Nitrogen was used as the nebulizer gas and as well as the drying gas at a 8 L/min at a drying gas temperature of 350 °C. Multiple reaction monitoring (MRM) was used for quantitative analysis (Table 1). Both quadrupoles were maintained at unit resolution (0.7 unit at half height). Data aquisition and processing was done with the software Analyst™ 1.41 (Apperla).

Table 1: Masses, transitions and parameters programmed in the LC-MS-MS procedure

Compound	Q1 (m/z)	Q3 (m/z)	Dwell time (ms)	DP (V)	FP (V)	CE (V)	CXP (V)	EP (V)
fentanyl	337.2	188.1	40	41	220	33	18	10
fentanyl-D5	342.1	188.1	40	41	220	33	18	10

DP=Declustering Potential, FP=Focussing Potential, CE=Collision Energy, CXP=Cell Exit Potential

Quantitation

For quantitation, blood was spiked at five concentrations (1, 5, 10, 25, 50 µg/L). All calibration (and quality control) samples were aliquoted into 1.5-ml polypropylene cups and stored frozen at -18°C. Quantitations followed the method of internal standard. Precision and accurancy for each determined compound was measured using in-house quality control (QC) samples (spiked matrix).

Results and Discussion

The choosen conditions for the analysis allowed a fast elution of the substances within 3.5 min and a lower limit of quantification of 1 µg/L (S/N >10) for fentanyl. The upper limit of quantification is 50 µg/L.

Precision was measured using in-house quality-controll-samples. The following CV-data was obtained (interday, n=10): 6.3% (4 µg/L), 8.1% (15 µg/L) and 6.4% (30 µg/L). Correlation coefficient (r) of the calibration curve (spiked matrix) was 0.9999 (figure 2).

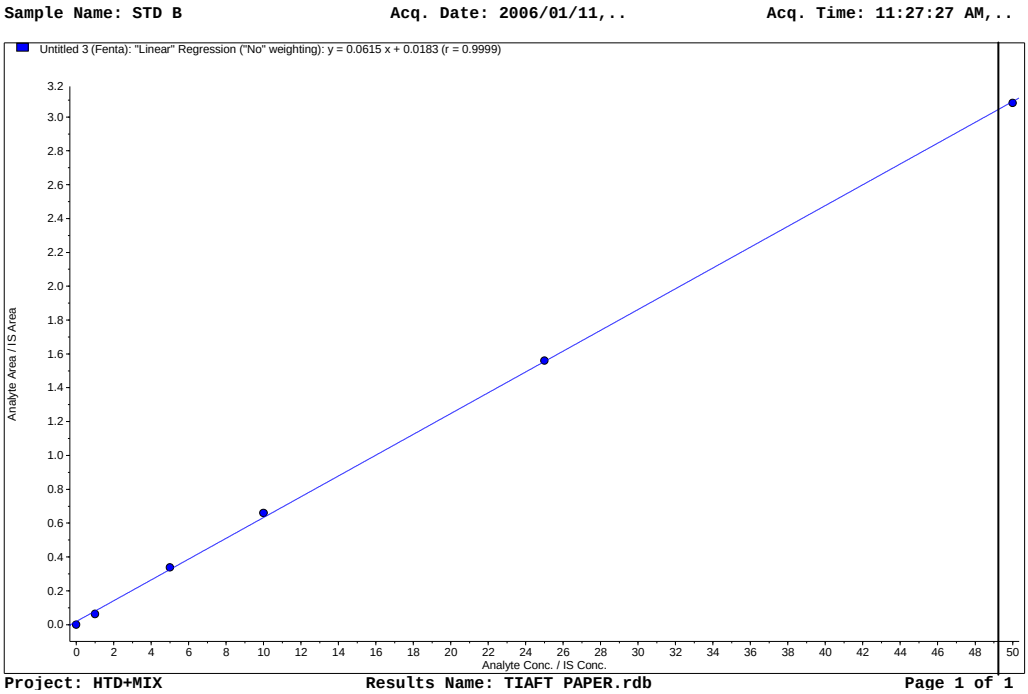

Figure 2: Calibration curve of fentanyl (LC-MS-MS)

Corresponding to the suspected fentanyl intoxication, the sample was analysed with the described analytical method. As can be seen from the chromatogram (Figure 3), **F** is clearly identified through its retention time (2.2 min) as well as through the mass transition 337.2 ->188.1 (amu). The **F** concentration in the sample was 35.1 µg/L.

It was anticipated that the placement of the patch (warm place, intensive blood circulation – or may be even more intensive due to an masturbation attempt) had a major effect on the rate of absorption which finally resulted in fatal blood concentration of 35.1 µg/L.

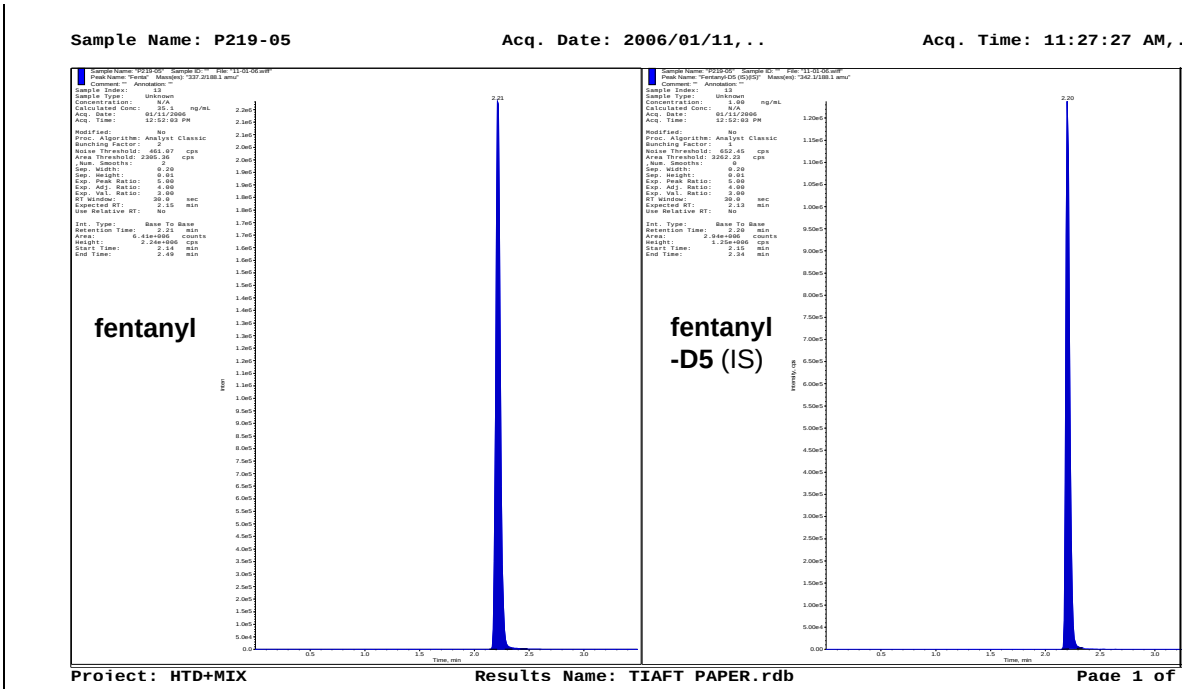

Figure 3: ESI(+)-LC-MRM-chromatogram of the patient`s sample

It is known, that the interindividual transdermal penetration rate of fentanyl transdermal varies [9]. In a study, one person showed a higher penetration rate than the average following patch application. This may indicate that human skin (e.g. area of application) and not the patch barrier was the rate-determining factor for determined individuals. In vitro experiments showed, that the placement of **F** patches in the groin region of dogs, compared with skin samples from the thoratic and neck region, may decrease the lag time to achieve analgesia perioperativelly [10].

Poisoning by fentanyl was described several times in literature [11, 12]. A variaty of routes of administration of the drug were identified: e.g. transdermal application of multiple Durogesic patches [13] intravenous injection of patch contents [14, 15], oral [16] or rectal administration [17].

In the final report, the present case was interpreted as follows: The death was caused by an overdose of fentanyl. It was hypothesised that in this case the placement of the patch had a major effect (e.g. a temperature-dependent increases of the **F** release from the system; and/or the systems was slightly damaged; and/or the skin region itself lead to a rapid release of the patch contents [1]) on the rate of absorption which finally resulted in a fatal blood concentration of 35.1 µg/L.

There were no indications of a crime or a suicidal attempt. Therefore, an accident in a self-treatment was assumed. One motivation of this unusual place of self-application of the patch could be the idea to get an extra kick of sexual fantasies and hallucinations (assuming that he rubbed his penis with the patch to release a bolus-like **F** intake during masturbation from the transdermal system).

Missing informations or actions are:

- How long was the time between (a possible) the prevoius **F** patch on the final **F** patch (cumulation)?

- Where there indications for masturbation or other sexual activities ? Was the transdermal system damaged (opening error, due to possible masturbation)? More in-vivo experiments concerning the placement of the **F** patch in “unsual” regions.

Conclusion

A case of fentanyl intoxication is reported. The blood concentration of fentanyl was 35.1 µg/L. Death due to the application of fentanyl transdermal system which was patched on the penis has not been previously reported. The presented case demonstrated fentanyl misuse with a transdermal patch at an unsual place of possible high resorption. Identification and quantification of fentanyl was carried out with a sensitive and reliable analytical method for the determination of fentanyl in human blood using LC-MS-MS. The method is based on a simple, fast liquid-liquid extraction under alkaline conditions. The time for analysis was 3.5 min.

References

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