A fatal case of poisoning with ethanol and psychotropic drugs with putrefactive changes

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SUMMARY

We present a fatal case involving poisoning with paroxetine, flunitrazepam, and ethanol, with putrefactive changes. Quantitative toxicological analysis showed that the concentrations of paroxetine and 7-aminoflunitrazepam, a metabolite of flunitrazepam, in the femoral blood were 0.28 μg/ml and 0.17 μg/ml, respectively. We also detected an ethanol level of 2.90 mg/ml and an n-propanol level of 0.10 mg/ml. We concluded that the cause of death was due to the interaction of paroxetine, flunitrazepam, and ethanol. The effects of putrefactive changes should be considered during forensic toxicological evaluation.

Keywords: flunitrazepam – ethanol – paroxetine – gas chromatography mass spectrometry (GC/MS).

Otrava etanolem a psychotropními látkami u případu s hnilobnými změnami

SOUHRN

Je prezentován případ smrtelné otravy paroxetinem, flunitrazepamem a etanolem v terénu hnilobných změn. Kvantitativní toxikologická analýza vykázala, že koncentrace paroxetinu a 7-aminoflunitrazepamu (metabolitu flunitrazepamu) ve vzorku femorální krve byla 0,28 μg/ml a 0.17 μg/ml. Také byla zjištěna hladina alkoholu 2,90 mg/ml a n-propanolu 0,10 mg/ml. Usuzujeme proto, že smrt nastala v důsledku vzájemné interakce paroxetinu, flunitrazepamu a etanolu. Bylo též uvažováno o vlivu hnilobných změn na forensní toxikologické vyhodnocení.

Klíčová slova: flunitrazepam – etanol – paroxetine – plynová chromatografie s hmotnostní spektrometrií (GC/MS).

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The evaluation of toxicity due to ingestion of multiple psychotropic drugs, with or without ethanol, is an important problem in the field of forensic toxicology (1,2). Paroxetine, a selective serotonin reuptake inhibitor, has a high affinity for serotonergic uptake sites. This drug increases the concentration of serotonin in the synaptic cleft by inhibiting its re-uptake (3,4). Flunitrazepam, an *N*-methyl-2´-fluoro analogue of nitrazepam (5,6), is a central nervous system depressant that may cause drowsiness, hangover, fatigue, dizziness and ataxia (5), and additive effects may occur when ethanol is co-ingested (6). Here we report a case of death with putrefactive changes involving the toxicity of paroxetine, flunitrazepam, and ethanol.

CASE REPORT

A Japanese male in his fifties was found dead in his room in the middle of summer. He had a history of alcohol dependence. Subsequent investigation by the authorities revealed that he had been receiving therapy for depression and alcohol problems, and was taking prescribed drugs.

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The deceased was 168 cm in height and 74.5 kg in weight. Putrefactive changes were evident. The heart weighed 389 g and contained 37 ml blood without coagulum. The brain weighed 1405 g and was discolored. The left and right lungs weighed 498 g and 484 g, respectively, and were congested. Approximately 20 ml of stomach contents, which included a red-brownish fluid, were noted. Other than congestion and putrefactive changes, no notable changes in other organs were observed. A drug screening test using a Triage™ (Biosite Diagnostic Inc., San Diego, CA, USA) panel was positive for benzodiazepines. Postmortem samples of the left/right heart blood, femoral venous blood, urine, and stomach contents were collected for toxicological examination and stored at -20°C until analysis.

Toxicological analysis

Toxicological analysis was performed using a 6890N gas chromatograph combined with a 5973 MS mass spectrometer (Agilent Technologies, Santa Clara, CA, USA). Identification and quantification of each drug were performed as described (7). Chromatographic separation was performed with a fused-silica capillary column DB-5MS (30 m × 0.25 mm I.D., 0.25 μm film thickness; J&W Scientific, Folsom, CA, USA). The operating conditions for gas chromatography mass spectrometry (GC/MS) were as follows. The carrier gas was helium in constant pressure mode. The injector temperature was set at 260 °C. The oven temperature was set at an initial temperature of 60 °C for 2 min, and was programmed to then rise 20 °C/min to 300 °C with maintenance at 300 °C for 10 min. The MS system was operated in the electron-impact mode with an electron energy of 70 eV

Table 1. Concentrations of drugs (μg/ml or μg/g), ethanol (mg/ml), and n-propanol (mg/ml) in each sample and their therapeutic and fatal ranges.

Specimen	paroxetine	flunitrazepam	7-aminoflunitrazepam	ethanol	n-propanol
Left heart blood	0,08	N.D.	0,10	2,71	0,08
Right heart blood	0,24	N.D.	0,10	3,03	0,08
Femoral venous blood	0,28	N.D.	0,17	2,90	0,10
Urine	0,48	N.D.	0,41	3,17	0,02
Stomach contents	1,62	N.D.	0,77	N.A	N.A
Therapeutic range	0.031-0.062 [10]		0.005-0.015 [11]		
Fatal range	0.41< [12]		0.16< * [6,13]		

* combined total levels of flunitrazepam and 7-aminoflunitrazepam [13]
** Each figure in parentheses represents the references.
N.D.: not detected
N.A.: not analyzed

and an ion source temperature of 230 °C. The sample solution (1 μl) was injected in splitless mode. We used diazepam-d_5 as an internal standard. The retention times were fixed using the retention-time locking technique with diazepam-d_5 as the locking compound (7). Samples were prepared as described (7). Extraction was performed with a cartridge Focus column (Varian, Lake Forest, CA, USA), and acetylation was performed. The eluate was evaporated and reconstituted in 100 μl ethyl acetate, and 1 μl of this solution was injected into the GC/MS.

Quantitation of ethanol and n-propanol was performed using head-space gas chromatography as described (8,9).

Toxicological analysis identified paroxetine, 7-aminoflunitrazepam, which is a metabolite of flunitrazepam, ethanol and n-propanol. Table 1 shows the quantitation of each substance in the blood, urine, and stomach contents of the deceased, and also summarizes the fatal and therapeutic levels of these substances (6,10–13).

DISCUSSION

In the present case, although the blood concentration of paroxetine exceeded therapeutic levels by approximately 4-fold (10), this concentration was still lower than the fatal concentration for paroxetine alone (12). However, to the best of our knowledge, no reports have been published describing the effects of putrefaction on postmortem blood paroxetine levels.

We detected an ethanol level of 2.90 mg/ml and an n-propanol level of 0.10 mg/ml in the blood, and an ethanol level of 3.17 mg/ml and an n-propanol level of 0.02 mg/ml in the urine. Because n-propanol forms in the blood or urine during the process of putrefaction, we must consider postmortem formation of ethanol in the present case (14,15). However, endogenous production of ethanol may be as high as 0.15% (1.5 mg/ml) in the blood (14). The antemortem ethanol concentration can be roughly estimated by deducting 25 times the concentration of n-propanol from the concentration of ethanol in the blood or urine (15). Although we could not precisely determine the antemortem blood ethanol levels, we were able to estimate the presence of ethanol in the blood at the time of death.

We detected 7-aminoflunitrazepam, but not flunitrazepam in any sample. This may have been due to postmortem bioconversion of flunitrazepam to 7-aminoflunitrazepam (16). In addition, the presence of 7-aminoflunitrazepam is an important marker of flunitrazepam usage (6,17). Because a portion of flunitrazepam is converted to 7-aminoflunitrazepam (16), its effect can generally be estimated from the sum total of the flunitrazepam and 7-aminoflunitrazepam concentrations in the femoral blood (17).

Flunitrazepam is a central nervous system depressant (5), and the concentration that is fatal is lower when ethanol is co-ingested (6). The concentration of 7-aminoflunitrazepam plus ethanol in our case was at fatal levels (6), and additive depressive effects on central nervous system function would have occurred due to the high levels of paroxetine. We concluded that the cause of his death in this case was due to the ingestion of multiple psychotropic drugs plus ethanol.

CONCLUSION

We sometimes observe postmortem changes in daily forensic practice, and have only a limited amount of data concerning the postmortem changes or the effects of drug interactions with ethanol. The present case indicates that we should pay more attention to the effects of postmortem putrefactive changes and the toxicity of drug interactions when evaluating the cause of death.

CONFLICT OF INTEREST
The authors declare that there is no conflict of interests regarding the publication if this paper.

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XXIII. Congress of International Academy of Legal Medicine

Dva a pol roka po 22. kongrese Medzinárodnej akadémie súdneho lekárstva (IALM), ktorý sa konal na východe Európy v Istanbule, sa miesto konania 23. kongresu posunulo ešte viac na východ do Ázie na Arabský polostrov na pobrežie Perzského zálivu do najväčšieho mesta Spojených arabských emirátov Dubaja. Išlo o prvý kongres IALM organizovaný v regióne Blízkeho východu a severnej Afriky. Dubaj je významným obchodným a turistickým centrom s významným letiskom, ale je aj miestom vysnívaných dovoleniek mnohých, ktorým ponúka luxus na každom kroku a extrémne horúce počasie. To bol zrejme aj dôvod na skrátenie tradičného trojročného časového intervalu od posledného kongresu Medzinárodnej akadémie súdneho lekárstva.

Kongres bol organizovaný pod záštitou najvyšších štátnych predstaviteľov, prezidenta a viceprezidenta Spojených arabských emirátov a korunného princa Dubaja, pre nás trochu nezvykle v spolupráci s hlavným miestnym organizátorom dubajskou políciou. Miestom kongresových aktivít bolo Dubai International Convention & Exhibition Centre v komplexe Dubai World Trade Centre, ktoré sa nachádza pri stanici metra v blízkosti „mrakodrapovej aleje" a je ľahko dostupné aj taxíkom.

V rámci odborného programu kongresu od 19. do 21. januára 2015 odznelo v 13 sekciách 131 prednášok a v 13 rovnomenných sekciách bolo prezentovaných 212 vývesiek. Boli prezentované pôvodné práce a kazuistiky zo všetkých oblastí súdnolekárskej praxe a hraničných odborov. Najviac príspevkov bolo prezentovaných v sekciách „Open topics" a „Forensic pathology". Téma využitia zobrazovacích metód v postmortálnej diagnostike a expertíze je už roky na vzostupe a oslovila najmä domácich odborníkov, ktorí plánujú zaviesť tieto metódy do praxe, čo pri ekonomických možnostiach krajiny určite nebude problém. Právo rozhodnúť o forme prezentácie si vyhradili organizátori. Od slovenských autorov boli prezentované práce: „Experimental wound ballistic of ricochet projectiles: report of continuing studies" autorov: N. Moravanský, L. Juříček, V. Rekeň, P. Dohnal, P. Kováč, A. Zummerová a „Medicolegal analysis of drug-related deaths in Slovakia between 2004 and 2013" autorov: J. Šidlo, I. Šteliar. Je potešiteľné, že obidve práce boli vedeckým a organizačným výborom vybraté na ústnu prezentáciu. Pri opakovanej neúčasti českých kolegov na vrcholných súdnolekárskych podujatiach je dobrou správou, že doc. MUDr. Petr Hejna, Ph.D, MBA sa stal členom redakčnej rady novovznikajúceho časopisu „Arab Journal of Forensic Sciences & Forensic Medicine". V rámci odborného programu sa konali aj 4 obsiahle workshopy. Workshop „Forensic Antropology Society of Europe (FASE)", „International Committe of the Red Cross (ICRC)", „Technical Working Group Post-mortem Angiography Methods (TWGPAM)" a „Violence Against Women (VAW)". Kongresu sa zúčastnilo viac ako 900 účastníkov z 55 krajín.

V rámci „Gala Dinner", ktorá sa konala v Armani Pavilion v dolnej časti najvyššej budovy sveta Burj Khalifa (828 m), mali pozvaní prednášajúci a predsedovia sekcií možnosť vyviezť sa výťahom na vyhliadkovú plošinu na 124. poschodí (452 m), kde sa im naskytol skutočne fascinujúci pohľad na večerné mesto. Priamo z pavilónu mohli hostia sledovať každú polhodinu spievajúcu „Dubai Fountaine", ktorá poskytuje po každý krát úžasný zážitok kombináciou hudby, svetelných efektov a výstrekov vody v niektorých fázach až do výšky 150 metrov.

V rámci kongresu sa konala aj členská schôdza International Academy of Legal Medicine, ktorá opätovne zvolila staronového prezidenta prof. Santo Davide Ferraru ako aj staronové prezídium Akadémie a odsúhlasila niektoré zmeny stanov. Miesto a termín 24. kongresu neboli určené, nakoľko zatiaľ nikto neprejavil záujem o jeho organizáciu.

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