ANALYSIS OF FORENSIC SAMPLES OF “ECSTASY” TABLETS SEIZED IN NOVI SAD DURING THE 2004 YEAR

Dragana M. Zgonjanin, Eva S. Lončar and Miloš M. Tasić

The paper presents results of the analysis of illicit synthetic drugs in the form of tablets distributed under the name “Ecstasy”, seized by the police in the broader area of Novi Sad 2004. A huge number of tablets has been analyzed (n=121), of various colours and with impressed symbols from the total amount of 93 seizures, which totally amounted to 1458 tablets. Regarding the number of seizures ecstasy (3,4-methylendioxy-N-methyl-amphetamine – MDMA) is dominant among all, and according to the quantity of seized tablets it is amphetamine (AP), while other amphetamine-type drugs (methamphetamine – MA, 3,4-methylendioxiamphetamine – MDA, 3,4-methylendioxi-N-ethyl-amphetamine – MDEA) have been found in rather small quantities and very rarely. Tablets mostly contain caffeine as an additive.

In the analytical procedure, the samples of tablets were subjected to liquid-liquid extraction and afterwards analyzed on the GCD (GC-EI) Hewlett-Packard instrument. The method is fast, reliable and reproducible for the analysis of amphetamine, methamphetamine, MDA, MDMA, MDEA, as well as various additives in the samples of seized tablets.

KEYWORDS: Amphetamine derivatives; Ecstasy; GCD analysis; drug abuse; forensic samples; Novi Sad

INTRODUCTION

Expansion of usage of synthetic hallucinogens in the last three decades has been observed worldwide, that has also been evident in our country. The drugs most frequently

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abused by young population are amphetamine and its derivatives methamphetamine (MA), 3,4-methylenedioxy-N-methyl-amphetamine (MDMA), 3,4-methylenedioxy-ethyl-amphetamine (MDA) and 3,4-methylenedioxy-N-ethyl-amphetamine (MDEA). Increased popularity of these drugs, especially of ecstasy or MDMA, originates from the illusion of having extremely high energy, euphoria, control of inhibition, and belief that they are “safe drugs”. Of course, this belief is false, since a huge number of intoxications, even fatalities, have been recorded.

3,4-Methylenedioxy-N-methyl-amphetamine (MDMA), the popular name “Ecstasy”, was developed by E. Merck Pharmaceutical Company in Germany in 1914, in an attempt to produce a new appetite suppressant, although it was never distributed on the market or gained general acceptance. In the 1970’s and early 1980’s the drug was investigated again as an adjunct to psychotherapy based on its reported ability to improve interpersonal communications and enhance emotional awareness (1-4). Prior to its classification as a Schedule I drug in 1985, in the USA, it enjoyed some popularity in the psychiatry due to its therapeutic characteristics (1, 4). Namely, it has been suggested that the use of MDMA, in a controlled therapeutic setting, promotes trust and confidence between patients and therapists (5). Such reputation of MDMA is due to the fact that is generally characterized as an empathy-enhancing compound (6), i.e. as an empathogen or entactogen (Greek: “en” = inside; “gen” = to produce, originates, gives; and Latin: “tactus” = touch; i.e. “to touch within oneself”) (7). During the mentioned period, recreational use of MDMA was growing (6). Evidence on the widespread abuse as a recreational drug on the college campuses has been reported (8, 9).

Since then MDMA (ADAM, ecstasy, E, X, XTC, M&M, euphoria, “rave”, “hug-drug”, disco-biscuits, white doves, love drug, “rolls” and essence) has become widely abused and extremely popular as a recreational drug at parties, raves and discos. Its users report as “positive” effects, the changes in feelings and emotions, enhanced communication, empathy and understanding, changes in cognitive or mental associations, euphoria or ecstasy, and changes in perception, including hallucinations (10). Other reported effects include a great sense of pleasure, dissociation, sexual stimulation, relaxation, increased responsiveness to intimate touch, increased self-esteem, high energy and tireless individuals (11, 12). In addition, the belief of being a “safe drug” has been widespread among the addicts.

However, MDMA shares the same properties as other amphetamines, acting on both the heart and the central nervous system, with increased release of catecholamines (including serotonin) and prevention of reuptake (13,14). There is tremendous overlap between recreational and toxic levels. In seven patients who died of MDMA toxicity, blood levels ranged from 110 ng/mL to 1260 ng/mL. Levels in five patients who survived serious bouts of toxicity were from 200 to 970 ng/mL, while the levels in five car-accident victims were from 50 to 340 ng/mL (15, 16). Experience in Europe shows that MDMA abuse may result in a diverse group of psychiatric syndromes, including toxic psychosis (panic attacks, depersonalization, depression, “flashbacks” and obsessive-compulsive symptoms) (17- 20).

Numbers of fatalities have been described in the British literature, and from the data that has been presented, it appears that the increased toxicity seen in the U.K. is, at least partly, a function of how the drug is used (16, 21, 22). Almost all of the case reports from England involve young people who develop hyperthermia, rhabdomyolysis, renal failure, and disseminated intravascular coagulation (22). Common to almost all the cases is the fact that, after taking MDMA, the victim danced for many hours in hot, poorly ventilated clubs. The acute effects after taking MDMA are serotonin syndrome associated with hy-
perthermia and mental status changes and cardiovascular instability, severe hyponatremia induced cerebral or and pulmonary edema and idiosyncratic hepatotoxicity. There is also a direct amphetamine-like effect on the cardiovascular system and hyperthermia may be related to the key role serotonin plays in the hypothalamus in thermoregulation (23, 24).

In many cases, the designer drugs result in a physiological effect much like the “parent drug” and in some cases have additional very harmful side effects. In order to successfully prosecute these underground activities, it is very important to be able to reliably determine the exact structure of the particular designer drug. Analysis of this class of compounds is carried out by the method SPME/GC-MS (25), GC/MS (26, 27), high performance liquid chromatography (HPLC) (28), infrared spectroscopy and recently, by two variants of capillary electrophoresis (CE), micellar electrokinetic capillary chromatography (MECC) and capillary zone electrophoresis (CZE) (29).

The aim of this research was to profile samples (to determine the structure) based on the GCD analysis of seized tablets, which were distributed as ecstasy (MDMA) on the “black market”, and to show diffusion of this and other similar drugs, i.e. amphetamine derivatives (MA, AP, MDA, MDEA) in the broader area of Novi Sad during 2004. Analyses have been carried out in the laboratory of the Institute of Forensic Medicine in Novi Sad.

EXPERIMENTAL

Materials and reagents

Seized samples of synthetic drugs in the form of tablets have been analyzed. Samples of tablets of various colours and impressed symbols on their surface have been distributed on the “black market” under the name of “Ecstasy” and originated from the seized material from the wide area of Novi Sad, in the period January–December 2004.

Analysis of totally 121 tablets from 93 single seizures amounting to 1458 tablets has been carried out. At seizures of several tablets, a number of samples considered as representative for that seizure was analyzed, based on the method of free selection.

All used reagents were of HPLC grade.

Preparation of samples for GCD

Samples of tablets for analysis were prepared by liquid−liquid extraction (LLE). The tablets were crushed and 10 mg of the homogeneous powder were added to 1 mL deionized water, to which first 100 µl buffer pH=10 (NH₄Cl/NH₃) have been added. The solution has been mixed for 20 s, and after addition of 1 mL of ethyl acetate, for another minute. The sample has been then centrifuged for 5 min at 3000 rpm and the organic layer has been taken off and dried over anhydrous Na₂SO₄. After filtration a sample for GCD was obtained.

Analytical method

Analyses were carried out using a HP 1800A Hewlett-Packard instrument (Palo Alto, CA, USA). GCD is an advanced gas chromatography (GC) system introduced by Hewlett-Packard in 1994. It consists of a gas chromatograph, an electron ionization (EI) detection
system for m/z up to 550 and a data system. The EI detection generates retention time, abundance and the mass spectral data, so that the obtained are comparable with those obtained with a GC – mass spectrometry (MS) instrument. The conditions of the analysis were as follows: capillary column (Hewlett-Packard HP5, cross linked 5% phenylmethyl-silicone, 30 m × 0.32 mm i.d., film thickness 0.25 μm) with helium as the carrier gas; sample volume 1μL was injected with split flow (split mode) at the injection port temperature of 250°C, temperature of detector 280°C. The column oven temperature was programmed from an initial temperature of 50°C, then at a linear increase of 20°C/min to 130°C (held for 1 min), then at 9°C/min to 230°C (held for 3.89 min).

Components were identified by comparing their mass spectra with those found in reference libraries (Wiley 275, PMW-TOX3). A list of possible compounds was generated using an automated library search routine with probability-based matching (PBM) algorithm, and each mass spectrum was visually inspected to verify the match.

RESULTS AND DISCUSSION

Active components of illicit stimulating drugs and additives that are deliberately added to increase the mass, have been determined in tablet samples seized 2004 on the broad area of Novi Sad. In total, 1458 tablets have been seized in 93 seizure actions, out of which 121 tablets have been analyzed, whereby the tablets for analysis have been chosen within one seizure by the method of free selection.

According to the visual characteristics, the tablets differed from each other according to the colour and impressed symbol (logo), however there were no different tablets within one seizure. The most frequent impressed symbols were "Mitsubishi", "Mercedes", "Adidas", "Puma", "Nike", a star, "yin-yang", "smile", "?" and rarely a diamond, heart, dolphin, bow and arrow, “Batman”, X and E. The most frequent colours were beige, pink, gray and green. Weight of tablets was in the range from 100 to 300 mg. Since these colours, the logo and the structure are identical to those that are well known from the forensic literature of the USA and Western Europe, it makes us believe, that Vojvodina is the secondary market for this type of narcotics and that originated from the illegal laboratories in the USA and Western Europe (30).

In the seized samples, amphetamine and methamphetamine MDA, MDMA and MDEA have been identified. These illegal substances are on the list of narcotics “Decision on Determination of Narcotics”, Official Gazette (31), and any possession of them is punishable. The mentioned compounds were completely separated and detected, but they were also detected simultaneously if they were present in the same sample.

None of the compounds usually encountered in illicit samples interfere with target compound (e.g., amphetamine or ecstasy). This allows the detection of additives like, for example, caffeine and some pharmaceutical preparations that are present in these synthetic mixtures (barbitone, aminophenazone, etc.). Representative profile chromatograms of two tablet samples of both amphetamine and ecstasy from different seizures are shown in Figs. 1 – 4 and tables 1 – 4.
Table 1. Retention time ($t_R$), compound and relative content (Rel. %) of profile of the tablet from Fig. 1

<table>
<thead>
<tr>
<th>$t_R$ (min)</th>
<th>Compound</th>
<th>Rel. %</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.66</td>
<td>amphetamine</td>
<td>25.55</td>
</tr>
<tr>
<td>10.37</td>
<td>N-formylamphetamine</td>
<td>4.26</td>
</tr>
<tr>
<td>14.70</td>
<td>caffeine</td>
<td>52.91</td>
</tr>
<tr>
<td>15.76</td>
<td>aminophenazone</td>
<td>11.15</td>
</tr>
</tbody>
</table>

Fig. 1. Profile of the tablet sample of amphetamine from the October seizure

Fig. 2. Profile of the tablet sample of amphetamine from the December seizure
Table 2. Retention time ($t_R$), compound and relative content (Rel. %) of profile of the tablet from Fig. 2

<table>
<thead>
<tr>
<th>$t_R$ (min)</th>
<th>Compound</th>
<th>Rel. %</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.59</td>
<td>amphetamine</td>
<td>6.70</td>
</tr>
<tr>
<td>10.44</td>
<td>barbitone</td>
<td>18.92</td>
</tr>
<tr>
<td>14.19</td>
<td>4-methylbenzaldehyde</td>
<td>2.45</td>
</tr>
<tr>
<td>14.74</td>
<td>caffeine</td>
<td>68.33</td>
</tr>
</tbody>
</table>

Fig. 3. Profile of the tablet sample of ecstasy from the February seizure

Table 3. Retention time ($t_R$), compound and relative content (Rel. %) of profile of the tablet from Fig. 3

<table>
<thead>
<tr>
<th>$t_R$ (min)</th>
<th>Compound</th>
<th>Rel. %</th>
</tr>
</thead>
<tbody>
<tr>
<td>6.33</td>
<td>methamphetamine</td>
<td>2.35</td>
</tr>
<tr>
<td>10.93</td>
<td>MDMA (Ecstasy)</td>
<td>90.15</td>
</tr>
<tr>
<td>14.73</td>
<td>caffeine</td>
<td>6.21</td>
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</table>
On the basis of results profiling (structure determination) of clandestinely manufactured drugs, it is clear (Fig. 1-4) that they contain a great variety of active components (amphetamine, MDMA, MDEA), additives (caffeine, aminophenazone) and impurities (4-methylbenzaldehyde, N-formylamphetamine) derived from their manufacture, and consequently, do not belong to a common “batch” (the same source). Because profiling serves as a tool to relate different street drug to a common sources, to determine synthetic routes for synthetic drugs and to identify impurities which accumulate during the synthetic sequence because of the lack of quality control in clandestine laboratories, and which may cause public health risks because of their inherent chemical or biological influences.

Analyses have shown, that tablets distributed under the name “Ecstasy” were not always MDMA. The most frequent seizures contained ecstasy (MDMA) tablets (70 seizures out of 93), where the number of seized tablets predominantly varied from 1 to 3, while regarding the quantity of seized amphetamine tablets was dominant (761), (Table 5).
Table 5. Quantity of seized tablets distributed as “Ecstasy”, expressed in number of seized tablets per month

<table>
<thead>
<tr>
<th></th>
<th>Amphetamine</th>
<th>Ecstasy</th>
<th>MA</th>
<th>MDA</th>
<th>MDEA</th>
<th>Ephedrine</th>
</tr>
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<tbody>
<tr>
<td>January</td>
<td>202</td>
<td>17</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>February</td>
<td>498</td>
<td>282</td>
<td>4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>March</td>
<td>4</td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>April</td>
<td>1</td>
<td>23</td>
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<td>3</td>
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<td>July</td>
<td>68</td>
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</tr>
<tr>
<td>August</td>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>September</td>
<td>2</td>
<td>13</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>October</td>
<td>31</td>
<td>46</td>
<td>2</td>
<td>10</td>
<td></td>
<td></td>
</tr>
<tr>
<td>November</td>
<td>14</td>
<td>7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>December</td>
<td>5</td>
<td>186</td>
<td>2</td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Σ</td>
<td>761</td>
<td>674</td>
<td>6</td>
<td>4</td>
<td>3</td>
<td>10</td>
</tr>
</tbody>
</table>

Other stimulative drugs (methamphetamine – MA, 3,4-methylendioxyamphetamine – MDA, 3,4-methylendioxy-N-ethyl-amphetamine – MDEA and ephedrine) have been found in small quantities and rarely.

Fig. 5. shows percentage of the structure of the totally seized drug quantity under the street name “Ecstasy” 2004. As Figs. 5. and 6. show, in the number of monthly seizures, the largest number of amphetamine tablets was seized in January (200) and in February (497), they originated from a single seizure, while in March, July and August there were no seizures of amphetamine at all. On the other hand, MDMA has been seized every month.

A comparison of calendar frequency of seizures (Fig. 6) the two most frequently consumed narcotics, amphetamine and MDMA, with the quantity (and the type) of seized drugs (Fig. 5), shows that seizures of small quantities of MDMA were carried out in the period March – September, with the exception of July (6 seizures, 68 tablets), which is partly identical with the period when the International Music Festival EXIT was held in our town. The slight growth of the seizure number and seized quantity of drugs can be noticed in October, while a significant seizure of MDMA (11 seizures, 186 tablets) took place in December. The frequency of “Ecstasy” tablets seizure per month shows that a number of seizures have been carried out at the beginning and at the end of the year, whereby larger quantity of tablets has been seized, when the weather is cold, so that young people meet mainly at parties, raves, and in the disco and consume tablets of this type as a recreational drug.

The largest part of the amphetamine quantity was with additives (only five tablets out of 761 were pure amphetamine, it means 0.66%) and various additives were frequently combined. Caffeine was always present in additives 99.34% (756 tablets) and as the only
Fig. 5. Distribution of “Ecstasy” tablets, in percentage
(numbers on the columns represent No. of tablets of a certain kind)

Fig. 6. Number of seizures of ecstasy (MDMA) and amphetamine in 2004
additive it was present in 721 tablets (94.74%), in combination with N-formylamphetamine in 28 tablets (3.68%), in combination with barbitone and 4-methylbenzaldehyde in 4 tablets (0.53%), and with N-formylamphetamine and aminophenazone in 3 tablets (0.39%).

The total quantity of seized ecstasy (674 tablets) contained additives in less cases (Fig. 7) in comparison with amphetamine.

![Fig. 7. Proportion of tablets of pure MDMA and MDMA with various types of additives](image)

Out of 674 tablets MDMA 408 (60.53%) was without additives, with caffeine as additive there were 243 tablets (36.05%), with caffeine and methamphetamine 13 tablets (1.93%), and with 3,4-methylendioxy-N-ethyl-amphetamine only 10 tablets (1.48%).

As could be expected, caffeine was the most frequent additive in the analyzed tablets. As an additive caffeine is added in order to increase the mass of tablet, as well as for the individual stimulating effect on the central nervous system. N-formylamphetamine, 4-methylbenzaldehyde and other intermediaries due to insufficiently controlled production process in clandestine laboratories, may cause undesired side effects in the form of toxicity and effect modification of the very active component, by which they increase the health risk of the addict.

Six seized tablets of methamphetamine and 4 tablets of 3,4-methylendioxyamphetamine were without additives, while all three tablets of 3,4-methylendioxy-N-ethyl-amphetamine additionally contained 3,4-methylendioxy-N-methyl-amphetamine in a small quantity.

It is interesting that in one seizure of 10 tablets ephedrine has been found in combination with other pharmaceutical preparations (paracetamol, dramamine, metorfane, chlorfeniramine). The list of narcotics in Serbia and Montenegro does not contain ephedrine and in this case it can be assumed that it was to serve as a drug substitution.

CONCLUSION

From the results of the analysis it can be concluded that the dominant street drug in the area of Novi Sad is ecstasy (3,4-methylendioxy-N-methyl-amphetamine – MDMA), which is often found pure (without additives), while the next drug is amphetamine, which is almost always combined with additives. Other synthetic amphetamine-type drugs (MA, MDA, MDEA) can be found rarely in this area.
The most frequent tablets seizures happen at the beginning and at the end of the year. Periodic frequency of seizures shows its partial connection with mass manifestations for young people and it also shows its permanent presence on the “black market”, as well as the trend of increased consumption of ecstasy (MDMA) in this area.

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АНАЛИЗА ФОРЕНЗИЧКИХ УЗОРКА ТАБЛЕТА “ЕКСТАЗИ” ЗАПЛЕЊЕНИХ У НОВОМ САДУ ТОКОМ 2004. ГОДИНЕ

Драгана М. Згоњанин, Ева С. Лончар и Милош М. Тасић

Представљени су резултати анализе незаконитих синтетских дрогоа у виду таблета дистрибуираних под називом “екстази”, заплењених од стране полиције на ширем подручју града Новог Сада у току 2004. године. Анализиран је велики број узорака таблета (n=121), разних боја и утиснутих симбола, из укупно 93 заплена, што представља 1458 таблета. По броју заплена доминантан је екстази (3,4-метилендиокси-N-метил-амфетамин – МДМА), а по количини заплењених таблета амфетамин (АП), док су друге дроге амфетаминског типа (метамфетамин – МА, 3,4-метилендиоксиамфетамин – МДА, 3,4-метилендиокси-N-етил-амфетамин – МДЕА) нађене у минорним количинама и веома ретко. Таблете углавном садрже као адитив кофеин.

У аналитичком поступку узорци таблета подвргнути су течно-течној екстракцији и потом анализирани на GCD (GC-EI) уређају. Метода је брза, поуздана и репродуктивна за анализу амфетамина, метамфетамина, МДА, МДМА, МДЕА, као и разних адитива у узорцима заплењених таблета.

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