TECHNICAL ARTICLE

Single-crystal structure analysis of designer drugs circulating in the Japanese drug market by the synchrotron radiation X-ray diffraction

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Over the past 20 years, many designer drugs derived from controlled substances have been widely distributed as easily available psychoactive substances and have become a serious problem in Japan. In order to determine the absolute structures of four new designer drugs derived from medicines (methylphenidate and phenmetrazine) X-ray single-crystal structure analyses were performed using the BL26B1 beamline of synchrotron radiation facility SPring-8. The results show that the molecular configuration of these designer drugs (having two asymmetric carbons), which were distributed in the illegal drug market had threo-forms as found for methylphenidate and phenmetrazine.
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Key words: synchrotron XRD, designer drugs

I. INTRODUCTION

Recently, many designer drugs whose structures have been slightly modified from the structures of controlled substances have emerged in the world. According to the EMCDDA–Europol 2015 Annual Report, several analogs of methylphenidate and phenmetrazine were detected from illegal products in European countries (EMCDDA, 2015). Methylphenidate is a therapeutic medicine for the treatment of attention deficit hyperactivity disorder and narcolepsy. Additionally, phenmetrazine is a stimulant, which was previously used as an appetite suppressant. These drugs are also known as drugs of abuse. Most of their absolute structures are not clear, although they sometimes have chiral carbons in their structures. Stereoisomers can be separated using the appropriate chromatography; however, it is difficult to determine their absolute structures without reference materials. Single-crystal X-ray structure analysis should be the best method to determine the absolute structure. For rapid analysis of the absolute structure, it is important to measure the crystal without recrystallization from the test sample, because crystal structure is possibly changed by re-crystallization.

To perform accurate and rapid X-ray structure analysis from small crystals, the X-ray structure analysis techniques using the synchrotron radiation are developed in SPring-8. Using BL40XU beamline, the absolute structures of two designer drugs are determined (Hashimoto *et al*., 2015).

In this paper, we investigated the absolute structure of newly emerged four analogs of methylphenidate and phenmetrazine (having two asymmetric carbons) using the BL26B1 beamline.

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II. MATERIAL

We analyzed four designer drugs circulating in the Japanese drug market: 3-fluorophenmetrazine hydrochloride ($C_{11}H_{14}FNO\cdot HCl$) (3FPM), mephenmetrazine hydrochloride ($C_{12}H_{17}NO\cdot HCl$) (MPM), isopropylphenidate hydrochloride ($C_{16}H_{23}NO_2\cdot HCl$) (IPP), and methylnaphthidate hydrochloride ($C_{18}H_{21}NO_2\cdot HCl$) (MNP). 3FPM and MPM are the derivative compounds of phenmetrazine and IPP and MNP are the methylphenidate derivatives. Hydrochloride salts of all of them are crystalline white powders (Figure 1).

Chemical formulas were determined by accurate mass spectrometer and two-dimensional (2D) structural formulas were determined by NMR (nuclear magnetic resonance), but steric structures were not elucidated. All of these drugs have two asymmetric carbons, so all of them can occur as stereoisomers, for example, threo or erithro.

III. EQUIPMENT

A. X-ray beam and goniometer

The single-crystal X-ray diffraction (XRD) measurements were carried out by the structure measurement system installed in the SPring-8 BL26B1 beamline (Ueno *et al*., 2006).

X-rays were monochromatized by Si (111) monochromator and wavelength was 0.750 00 Å. A toroidal mirror and collimator focused beam size of 50–100 μm circular size. A single crystal of the several microns to tens of microns of the sample drug was attached to a polyimide pin. The high precision alignment of the microcrystal is brought by only the single-axis (ω-axis) rotationgoniometer. The eccentricity of the rotation axis is <1–4 μm. Control of the measurement temperature 220 K is maintained by a nitrogen stream

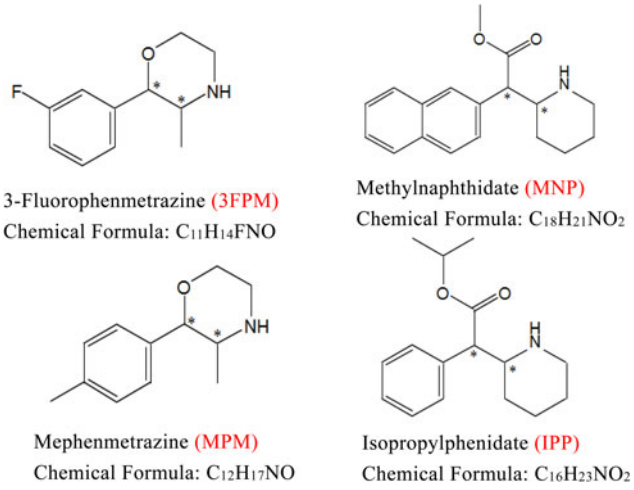

Figure 1. (Color online) Chemical formulas and 2D structures of four designer drugs.

B. 2D detector for the quick measurement

The crystal was measured by the oscillation method, and diffraction intensity data were recorded by a charge-coupled device (CCD) detector: MX225 (Rayonix) (BL26B1), whose CCD area: 225×225 mm^2

Number of pixels: 3072×3072, pixel size 73 μm.

Camera distance was 55 mm to gather high angle reflections of the organic small molecules.

Equipment is shown in Figure 2.

IV. METHOD

A. Sample pick up and X-ray measurement

Single particles from the drug powder sample were measured without recrystallization, and were picked up using a MiTeGen Micro Mount. Particle sizes are shown in Figure 3.

Because the CCD area is large, capturing of the reflections is rapid. Crystals were scanned from 0° to 180° with scan step 1°, exposure time was 1 s. Measurement times were 10 min to gather 180 diffraction images. Figure 4 shows one of the diffraction images of 3FPM. After 180 images of one crystal were measured, the orientation of crystal was changed and 180 more images were measured in order to increase the completeness. For measurement of 3FPM, 2033 reflections were collected from 360 diffraction images and completeness was

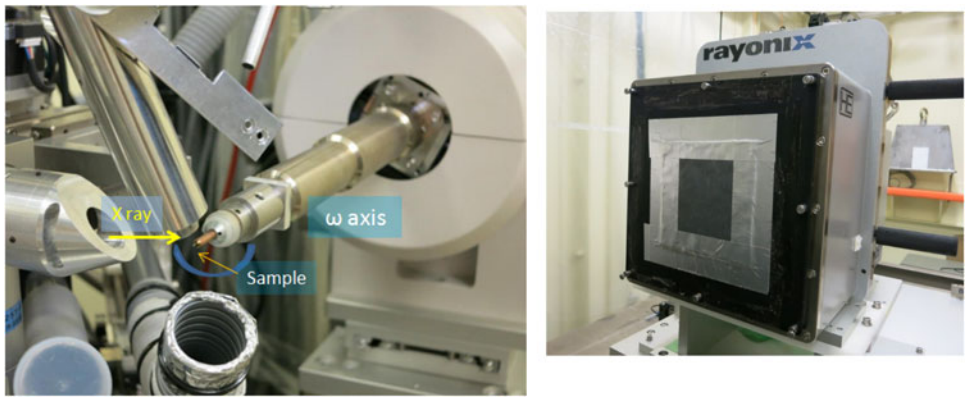

Figure 2. (Color online) Sample holder and goniometer (left side) and X-ray CCD detector (right side).

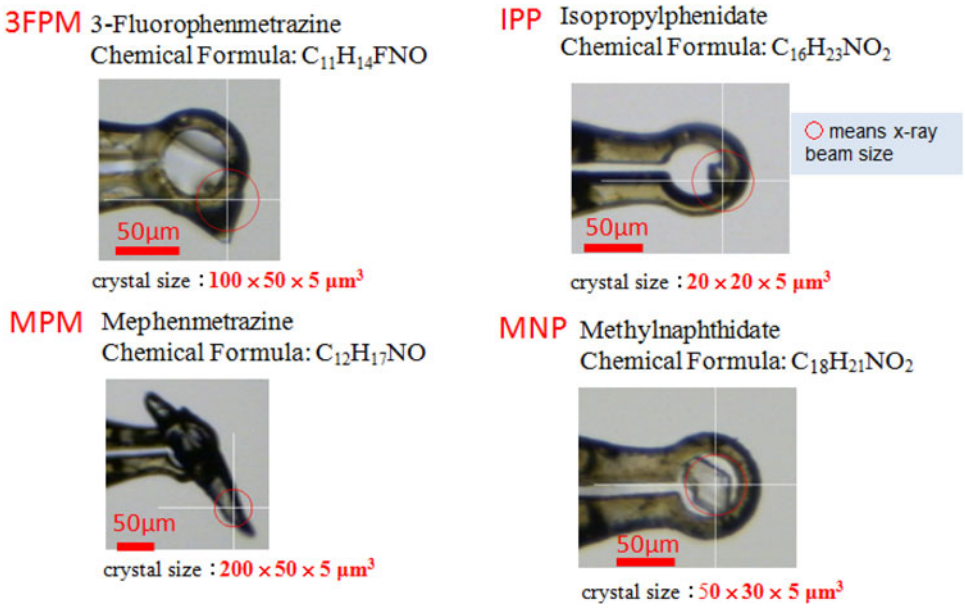

Figure 3. (Color online) Crystal powder particles of drugs in Micro Mounts. The red circle shows the X-ray beam size.

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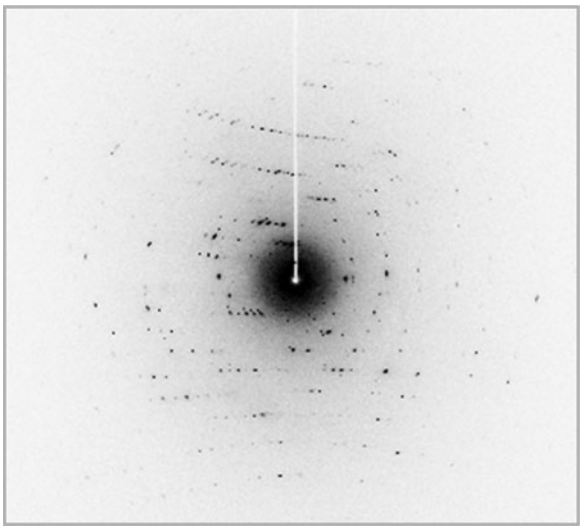

Figure 4. (Color online) One of the diffraction images of 3FPM.

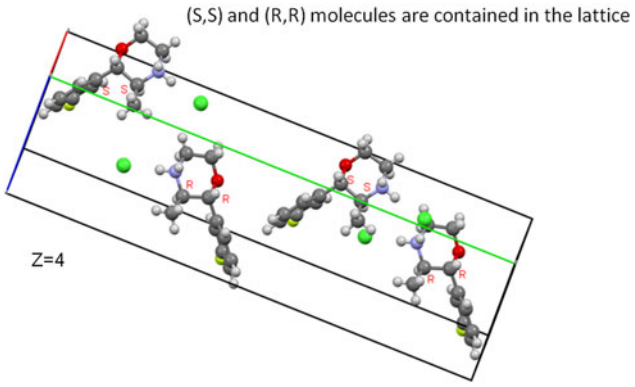

Figure 6. (Color online) Packing image of 3FPM.

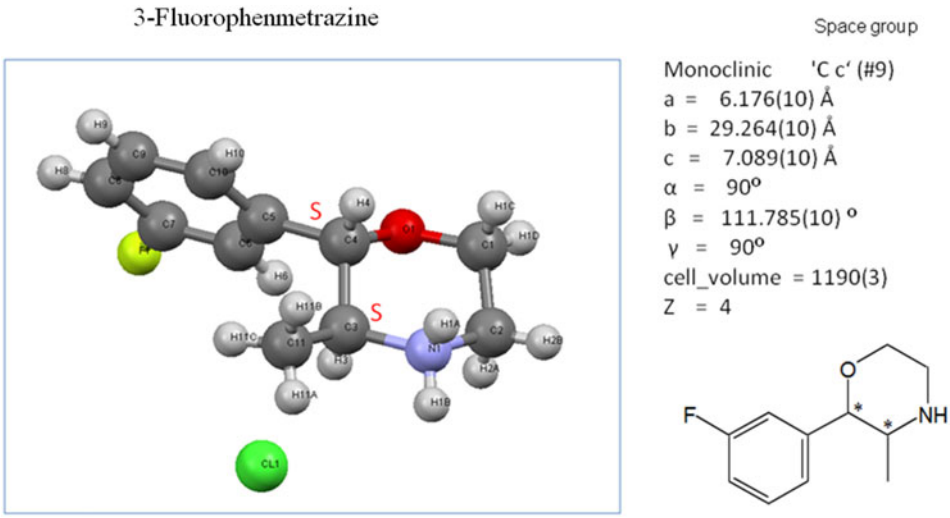

Figure 5. (Color online) Molecular structures of 3FPM.

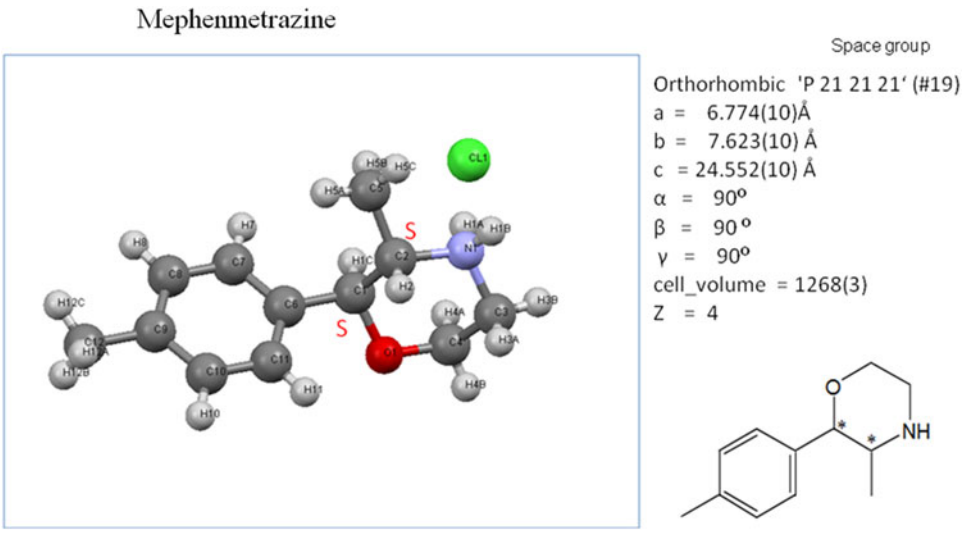

Figure 7. (Color online) Molecular structures of MPM.

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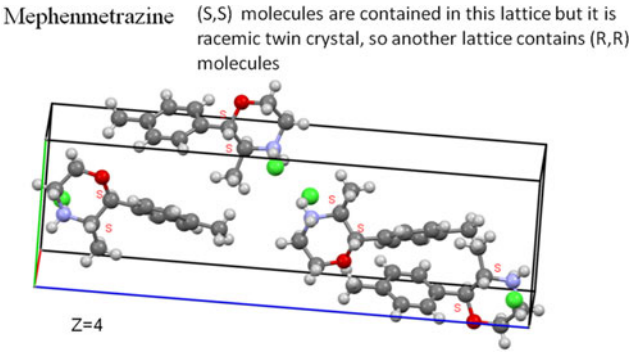

Figure 8. (Color online) Packing image of MPM.

0.986 (2θ: 2.992° - 53.708°). For the other three crystals, completenesses were as follows: IPP: 0.918 (2θ: 4.680° - 53.716°), MPM: 0.997 (2θ: 3.502 - 53.716°), MNP: 0.870 (2θ: 3.464° - 53.706°).

B. Determination of initial structure model and the refinement of structure model

1. Determination of the initial structure model

Using structure analysis software SHELXS (Sheldrick, 2008), the initial structure model was determined by direct methods. Atomic species were assigned based on the expected molecular structure. The hydrogen atoms were generated at calculated positions.

2. Refinement of structure model

Refinement software SHELXL (Sheldrick, 2008) was used for fitting the structure to measured diffraction data. Refined parameters were the positions of atoms (*XYZ*), and anisotropic displacement coefficients (U_{ij}).

V. RESULTS

A. 3-Fluorophenmetrazine hydrochloride

Figure 5 shows the molecular structure of 3FPM ($C_{11}H_{14}FNO \cdot HCl$) and Figure 6 shows the packing image. This measured structure is threo and the space group is monoclinic *Cc*. (S,S) and (R,R) molecules occur in the lattice.

B. Mephenmetrazine

Figure 7 shows the molecular structure of MPM hydrochloride ($C_{12}H_{17}NO \cdot HCl$) and Figure 8 shows the packing image. This measured structure is threo and the space group

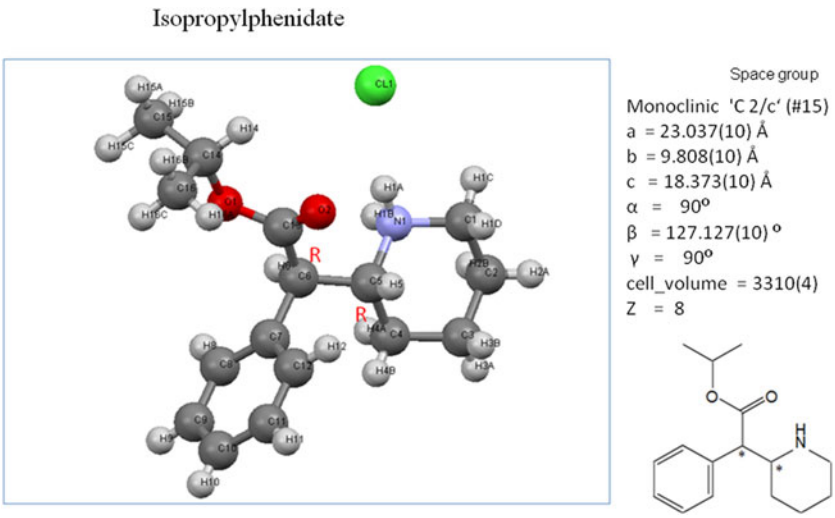

Figure 9. (Color online) Molecular structures of IPP.

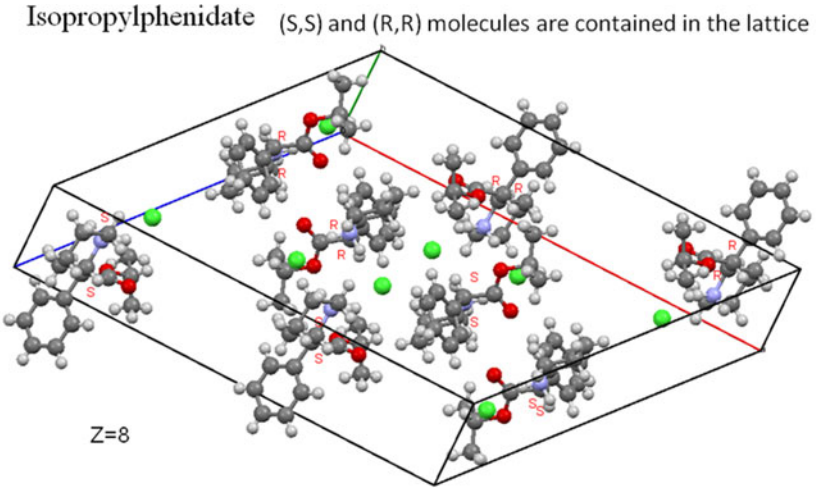

Figure 10. (Color online) Packing image of IPP.

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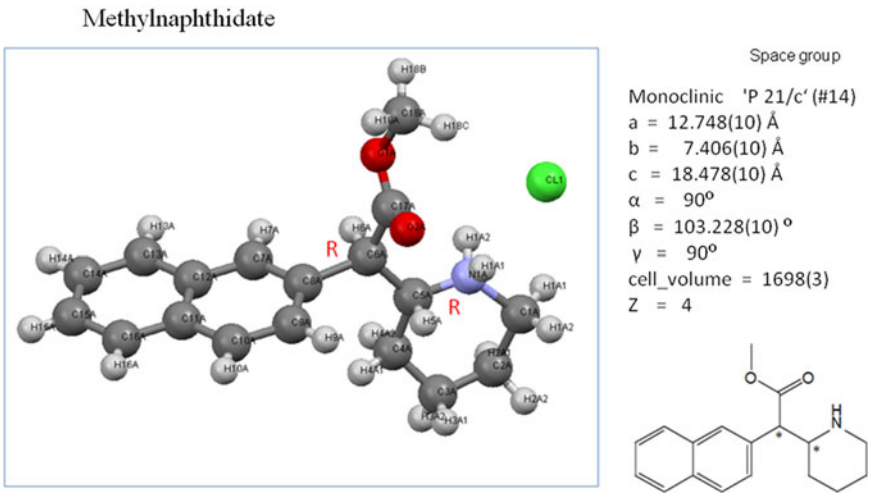

Figure 11. (Color online) Molecular structures of MNP.

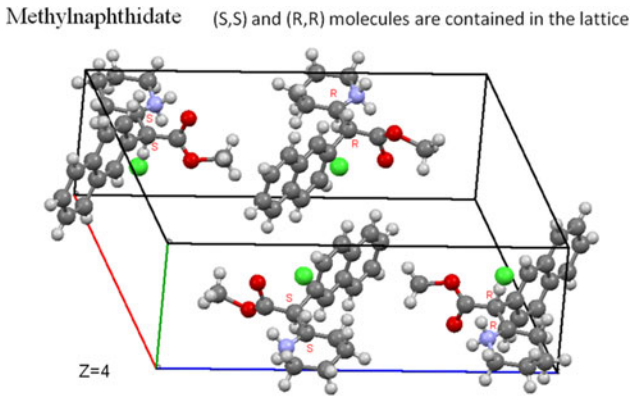

Figure 12. (Color online) Packing image of MNP.

is orthorhombic $P2_12_12_1$. Because the measured crystal was racemic twin, (S,S) and (R,R) molecule occur in the crystal.

C. Isopropylphenidate hydrochloride

Figure 9 shows the molecular structure of IPP ($C_{16}H_{23}NO_2 \cdot HCl$) and Figure 10 shows the packing image. This measured structure is threo and the space group is monoclinic $C2/c$. (S,S) and (R,R) molecules are in the lattice.

D. Methylnaphthidate

Figure 11 shows the molecular structure of MNP ($C_{18}H_{21}NO_2 \cdot HCl$) and Figure 12 shows the packing image. This measured structure is threo and the space group is monoclinic $P2_1/c$. (S,S) and (R,R) molecules are in the lattice.

Finally, the drugs for which we determined the molecular structure were hydrochlorides of:

(1) 3-Fluorophenmetrazine: threo-2-(3-fluorophenyl)-3-methylmorpholine;
(2) Methylnaphthidate: threo-methyl 2-(naphthalen-2-yl)-2-(piperidin-2-yl)acetate;
(3) Isopropylphenidate: threo-isopropyl 2-phenyl-2-(piperidin-2-yl)acetate;
(4) Mephenmetrazine: threo-3-methyl-2-(p-tolyl)morpholine.

Stereochemical structural formulas of four drugs determined by XRD and IUPAC name are shown in Figure 13.

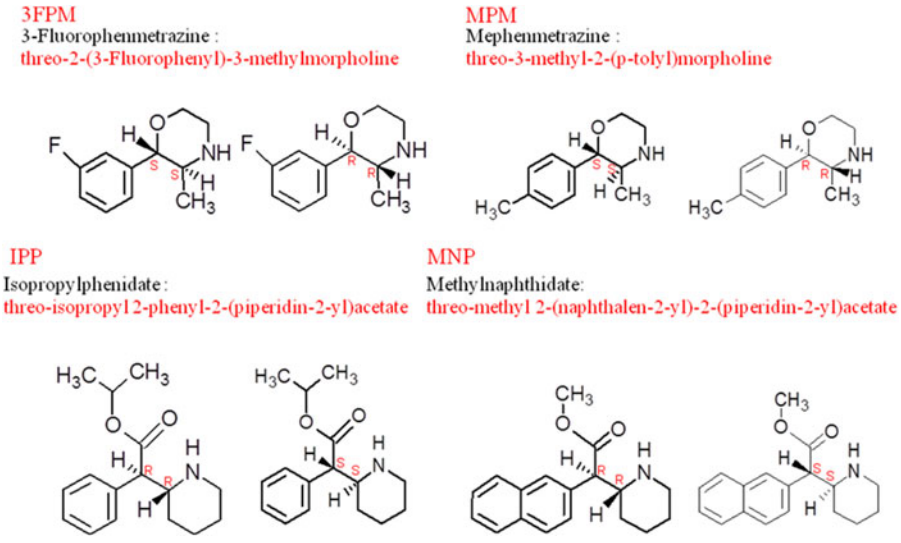

Figure 13. (Color online) Stereochemical structural formulas of four drugs and IUPAC names.

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VI. CONCLUSIONS

We successfully analyzed four designer drugs circulating in the Japanese illegal drug market. All of these drugs have two asymmetric carbon atoms, and steric structures of diastereomers of these four drugs were determined by single-crystal X-ray structure analysis.

The single-crystal X-ray structure analysis using synchrotron radiation X-ray has been proven to be the useful technique for the structure determination of designer drugs having a lot of stereoisomers.

Structural CIF files have been deposited with ICDD.

ACKNOWLEDGEMENTS

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EMCDDA (**2015**) EMCDDA – Europol 2015 Annual Report on the Implementation of Council Decision 2005/387/JHA.

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