Evaluation of Latex Agglutination Inhibition Reaction Test for Urinary Methamphetamine

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ABSTRACT

A latex agglutination inhibition reaction test for a screening method of methamphetamine in urine has been reported by Aoki et al. An evaluation of this method was attempted by using gas chromatography and chemical ionization mass fragmentography. Some false negative and false positive cases were found. It seems that this method needs some modification for false negative cases and also it needs a confirmation test.

Methamphetamine abuse has been one of the serious social problems in Japan. In the past the abuse was limited to the organized crime groups called "YAKUZA" in Japan and persons concerned with it, but recently it is spreading among the general public.

Methamphetamine is usually abused by intravenous injection, and the highest methamphetamine concentration in the body is observed in urine. The confirmation of methamphetamine abuse is always performed by analyzing urinary methamphetamine. Therefore, the development of a screening method for methamphetamine in urine has been greatly desired.

A latex agglutination inhibition reaction test for urinary methamphetamine has been reported by Aoki et al¹⁾. An evaluation of this method was attempted by gas chromatography and gas chromatography-mass spectrometry^{2,3)}. Some false negative and false positive cases were found, and it seems that this method needs some modification for false negative cases and also it needs a confirmation test.

MATERIALS

Fifty four urine samples were collected from 20 methamphetamine abusers or suspected

abusers admitted to a mental hospital. One urine sample was also collected from a non-abuser, who had caught a cold and had been given a normal amount of a methamphetamine analog, methylephedrine.

After collecting urine samples, methamphetamine was screened by the latex agglutination inhibition reaction test, and the samples were kept frozen without adding any chemicals until analysis by gas chromatography and gas chromatography-mass spectrometry.

METHODS

1. Screening test

The method used was the latex agglutination inhibition reaction test for urinary methamphetamine developed by Aoki et al¹⁾. The agglutination reaction is inhibited when the urine sample contains more than 1 μ g/ml of methamphetamine.

2. Gas chromatography and gas chromatographymass spectrometry

The combined method of gas chromatography and chemical ionization mass fragmentography for screening and quantitative analysis of methamphetamine and its metabolite in biological material²⁾ was used. Methamphetamine in the sample was extracted by using Extrelut[®] column³. The sensitivity limit of methamphetamine or amphetamine was set at about 0.001 μ g/g or 0.05 μ g/g.

RESULTS

The summarized results were shown in Fig. 1. Sixteen samples out of 55 were positive for methamphetamine by the screening test. However, one sample collected from a non-methamphetamine abuser given methylephedrine was false positive for methamphetamine because methamphetamine and amphetamine were not detected by the chemical ionization mass fragmentographic method of which detection limit was at about $0.05~\mu g/g$ of methamphetamine or amphetamine. Two samples containing less than

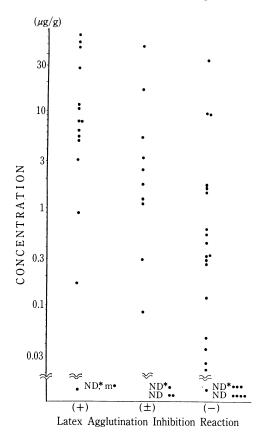


Fig. 1. Summarized results of the latex agglutination reaction test and methamphetamine concentration. ND*, m: methamphetamine concentration was less than 0.05 μ g/g and methylephedrine was given. ND*: less than 0.05 μ g/g. ND: less than 0.001 μ g/g.

 $0.2~\mu g/g$ of methamphetamine were also included

Twenty six samples out of 55 were negative for methamphetamine by the screening test. Six samples, however, contained more than 1 μ g/g of methamphetamine.

Thirteen samples out of 55 were not determined positive nor negative for methamphetamine by the screening test. Eight samples, however, contained more than 1 μ g/g of methamphetamine, and five less than 0.3 μ g/g.

Six samples were negative for methamphetamine in 26 samples of which methamphetamine concentrations were more than 1 $\mu g/g$.

DISCUSSION

The agglutination inhibition reaction test developed by Aoki et al¹⁾ is expected to be useful as a screening method for methamphetamine in urine. According to Aoki et al¹⁾, there are some problems in this method. The agglutination inhibition reaction is influenced by the urine state. For example, glucosuria or albuminuria can inhibit the reaction and it will result in false positive for methamphetamine. Urine with an extremely high or low pH level reacts nonspecifically and gives false negative results. In order to prevent these reactions, urine is diluted with 0.27 M glycine buffered saline (pH 9.6) in this method.

In 26 samples of which methamphetamine concentrations were more than 1 μ g/g, 6 samples were negative for methamphetamine. Thirteen samples, in which the methamphetamine concentrations were more than 1 μ g/g in eight and less than 0.3 μ g/g in five, were not determined positive nor negative. Since this method is a screening test, it seems that this method needs some modifications for false negative and undetermined cases.

This screening test has cross reactivities to methamphetamine analogs. One sample collected from a non-methamphetamine abuser given a normal amount of methylephedrine was positive for methamphetamine though neither methamphetamine nor amphetamine was detected by the chemical ionization mass fragmentographic method. Since this screening test sometimes gives false results, in which an over the counter drug is mistaken for methamphetamine, a confirmation test is needed.

REFERENCES

- Aoki, K. and Kuroiwa, Y. 1985. A screening method for urinary methamphetamine-latex agglutination inhibition reaction test. Forensic Sci. Int. 27: 49-56.
- Une, I., Yashiki, M., Nishiyama, Y., Kojima, T. and Yamawaki, S. 1981. The combined method of gas chromatography and chemical ionization mass fragmentography for screening and quan-
- titative analysis of methamphetamine and its metabolite in biological material (in Japanese with abstract in English). Proc. Jpn. Soc. Med. Mass Spectrom. 6: 125—128.
- Une, I., Yashiki, M., Yamauchi, J. and Kojima, T. 1983. Extraction of methamphetamine in blood and urine by Extrelut[®] column (in Japanese with abstract in English). Jpn. J. Legal Med. 37: 63-66.