有機りん系農薬検出キット

【キット構成】

1. NBP試薬(4-(4-ニトロベンジル)ピリジン)	1.2 ml x 1 本
2. TEP試薬(テトラエチレンペンタミン)	1.2 ml x 1 本
3. 抽出溶媒(ジエチルエーテル)	11 ml x 1 本
4. キャップ付き試験管	10 本
5. スポイト(NBP試薬、TEP試薬用)	2 本
6. スポイト(試料採取用、抽出溶媒用)	2 本

【操作手順】

1. 試料及び試薬を準備する。
2. 試料 1 ml を試験管にとる。
3. NBP試薬を 0.1 ml 加え混和する。
4. 100℃で 20分間 加熱する。
5. 冷却後、TEP試薬を 0.1ml 加え混和する。
6. 抽出溶媒 1 ml を加え混和する。
7. 静置して上層を観察する。
8. 検査の判定:薄ピンク色～赤紫色→有機りん系農薬含有

【注意事項】

- 試薬には危険物が含まれ、引火しやすいため、火気の近くでは行わないようにして下さい。
- 加熱の際は、火を使用しない器具を用いて、キャップを緩めるかあるいは外してから行って下さい。
- 抽出溶媒を加えた後は試験管のキャップを必ず閉めて下さい。

関東化学株式会社

【有機りん系農薬検出キットの検出感度】

有機りん系農薬	目視による検出感度(ppm)
Acephate	10
Bensulide	50
Butamifos	50
Cyanophos	1
Diazinon	10
Dichlofenthion	10
Dichlorvos	1
Dimethoate	1
EPN	10
Ethion	10
Ethoprophos	10
Fenitrothion	1
Iprofenfos	N.D.
Isofenphos	50
Isoxathion	1
Malathion	10
Methidathion	10
Phenthoate	1
Phosalone	10
Phosmet	1
Pyridaphenthion	10
Tetrachlorvinphos	10
Tolchophos-methyl	10
Trichlorphon	1
Vamidothion	10

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Direct colorimetric method for determination of organophosphates in human urine

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Abstract

A simple and sensitive method for determination of organophosphorus pesticides in human urine was developed by detecting the color complexes which resulted from reactions of organophosphorus pesticides and 4-(4-nitrobenzyl)pyridine (NBP) in urine. Based on studies of reaction conditions, e.g. reaction temperature and time, and reagent concentration, a colorimetric method was established. A 0.1-ml volume of NBP (45% in acetone) was added to a 1.0-ml volume of a urine sample, and the mixture was heated at 100°C for 20 min. After cooling, 0.1 ml of tetraethylenepentamine was added. The organophosphorus pesticides showed a characteristic purplish blue color and the coloring complexes which were produced were stable for several hours. Furthermore, these complexes could be determined spectrophotometrically. The detection limits were 0.10–10 μg/ml in urine. The required time for analysis was approximately 30 min for one sample. Comparing the result of the proposed method with those of the GC–MS method, the results were similar for the 12 poisoning cases studied. Thus, the proposed method is useful for detection of these pesticides in critical care practices.

Keywords: Organophosphates; Pesticides; Humans; Urine; Colorimetric test

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1. Introduction

Organophosphates are widely used for pesticides. Although in normal circumstances they are non-toxic for humans, many people die through suicidal ingestion of organophosphates every year [1]. The measurement of organophosphates in body fluids, such as urine and serum, may be important for the prompt medical management of suspected patients exposed to these compounds. A number of analytical methods based on chromatographic techniques such as paper chromatography [2], thin layer chromatography (TLC) [3,4], gas chromatography (GC) [5,6], gas chromatography–mass spectrometry (GC–MS) [7,8] and high-performance liquid chromatography [9] are used for qualitative and quantitative determination of organophosphates. They, however, are not always useful for emergency cases because of complicated laboratory instruments and time-consuming sample preparations. An accurate, simple and rapid method for detection of organophosphates in biological fluids is needed which may be helpful in critical care medicine. Our aim was to develop a simple and sensitive assay procedure for the determination of organophosphates.

4-(4-Nitrobenzyl)pyridine is used as a general chromogenic reagent for the detection of thio and nonthio organophosphates on developed paper and thin-layer chromatograms. This reagent was first used for detection of alkylating compounds such as iodomethane [10]. It was then used for the detection of organophosphates in the coloring reagent of TLC [11]. This chemical reaction was reported by Kramer and Gamson (Fig. 1) [12]. There is no report in which this reagent was used to directly react with organophosphates in biological samples.

We developed a simple and sensitive method for the detection of organophosphates in human urine. The coloring reagents were added to a urine sample and the mixture was heated. The required time for analysis was approximately 30 min for one sample. This proposed method was applied to poisoning cases and the results were compared with those of the GC–MS method and a commercial enzymatic screening kit.

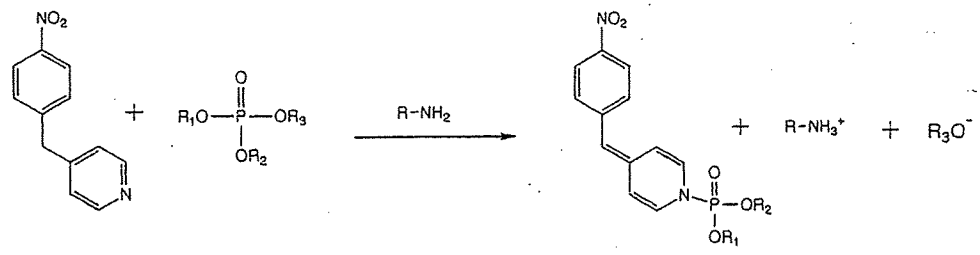


Fig. 1. Reaction scheme for the color reaction of 4-(4-nitrobenzyl)pyridine and organophosphates.

2. Material and methods

2.1. Reagents

Acephate, chlorpyrifos, cyanox, diazinon, dichlorvos (DDVP), dimethoate, disyston, edifenphos (EDDP), EPN, estox, fenitrothion (MEP), fenthion (MPP), formothion, isofenphos, isoxathion, malathion, methidathion (DMTP), monocrotophos, naled (BRP), phenthoate (PAP), parathion, prothiophos, pyridaphenthion, salithion, tetrachlorvinphos (CVMP), trichlorfon (DEP), vamidothion, 4-(4-nitrobenzyl)pyridine (NBP), tetraethylenepentamine (TEP), and other chemicals were purchased from Wako Pure Chemicals (Osaka, Japan) of analytical grade. Agri-Screen Ticket AT-10 Kit® (Wako Pure Chemicals, Osaka, Japan) was used for the assay which measured the inhibition of cholinesterase activity. Stock standard solutions (1.0 mg/ml) of organophosphates were dissolved in acetonitrile and stored at 4°C in a refrigerator. Stability of the stock solutions was analyzed by GC–MS, and they proved to be stable for several months.

Drug-free urine collected from healthy adult males was used to make urine samples containing organophosphates, and also as blank urine controls. Urine samples collected from 12 poisoning cases were kept frozen at −20°C until analyzed.

2.2. Procedure

A 0.1-ml volume of NBP (45% in acetone) was added to a sample urine (1.0 ml) and mixed for 30 s using a vortex mixer. The mixture was heated at 100°C for 20 min in a heating block (Taitec, Saitama, Japan). The mixture was then cooled to room temperature and TEP (0.1 ml) was added to the mixture. Diethyl ether (1.0 ml) was added the reaction mixture and the produced coloring complexes were extracted to the diethyl ether layer. The absorbance (λ_{520}) of the organic layer was measured using a spectrophotometer (Shimadzu, UV-200S double beam spectrophotometer).

2.3. Optimum conditions

To determine the effect of reaction temperature and time, urine samples spiked with 50 μg/ml of malathion or EPN were examined at four different temperatures (40, 60, 80, 100°C) for six different reaction times (5, 10, 15, 20, 30, 40 min). The reaction mixture was heated for 15 min at 100°C after the optimal heating time was determined.

To determine the effect of volume of color reagent used, urine samples spiked

with 50 μg/ml of malathion or EPN were examined at four different concentrations of NBP (10, 20, 30, 45%) and TEP (30, 50, 70, 100%).

Reproducibility was evaluated by examining 27 samples of organophosphates in urine spiked at 50 μg/ml on the same day in five replicates.

2.4. Gas chromatograph–mass spectrometer

The sample preparation was a slightly modified Extrelut method [8]. A 1.0-ml aliquot of urine sample was mixed with 0.5 ml of 0.01 M HCl. The mixture was applied to an Extrelut column which was pre-packed an Extrelut (2.0 g) to a glass column [180 × 1.0 mm (I.D.)] and left for 20 min. Organophosphates were eluted from the column with 4.0 ml of ethyl acetate. The eluent was evaporated to dryness in vacuo at 50°C. The residue was dissolved in 100 μl of acetonitrile. A 1-μl aliquot was injected into the GC–MS. The GC–MS used was a Hewlett Packard 5890 series II gas chromatograph–5971A mass selective detector, equipped with a 30 m × 0.25 mm (I.D.) fused-silica capillary column (Hewlett Packard, HP-5MS, film thickness 0.25 μm). The column temperature was set at 50°C for 5 min, programmed from 50 to 280°C at 15°C/min and held at 100°C for 5 min. The temperatures of the injection port and ion source were set at 250 and 280°C, respectively. Splitless injection mode was used. Helium with a flow-rate of 50 kPa was used as a carrier gas. The mass selective detector was operated in electron impact (EI) mode with 70 eV of electron energy, and a scan range from m/z 50 to 550. All data was acquired in full scan mode and selected-ion monitoring (SIM) mode.

2.5. Commercial screening kit assay

Agri-Screen Ticket AT-10 Kit® was used for the commercial screening kit which measured the inhibition of cholinesterase activity. The assayed was performed in accordance with the manufacturer's guidelines.

3. Results and discussion

3.1. Heating temperature and time

The effect of heating temperature is shown in Fig. 2A. The mixture was heated for 15 min. The absorbance of the produced coloring complexes were appreciably dependent on heating temperatures below 60°C, and were accelerated at around 100°C. The effect of heating time is shown in Fig. 2B. The mixture was heated at 100°C. The absorbance was dependent on the heating time for up to 20 min, but not beyond 20 min. The maximum absorbances of

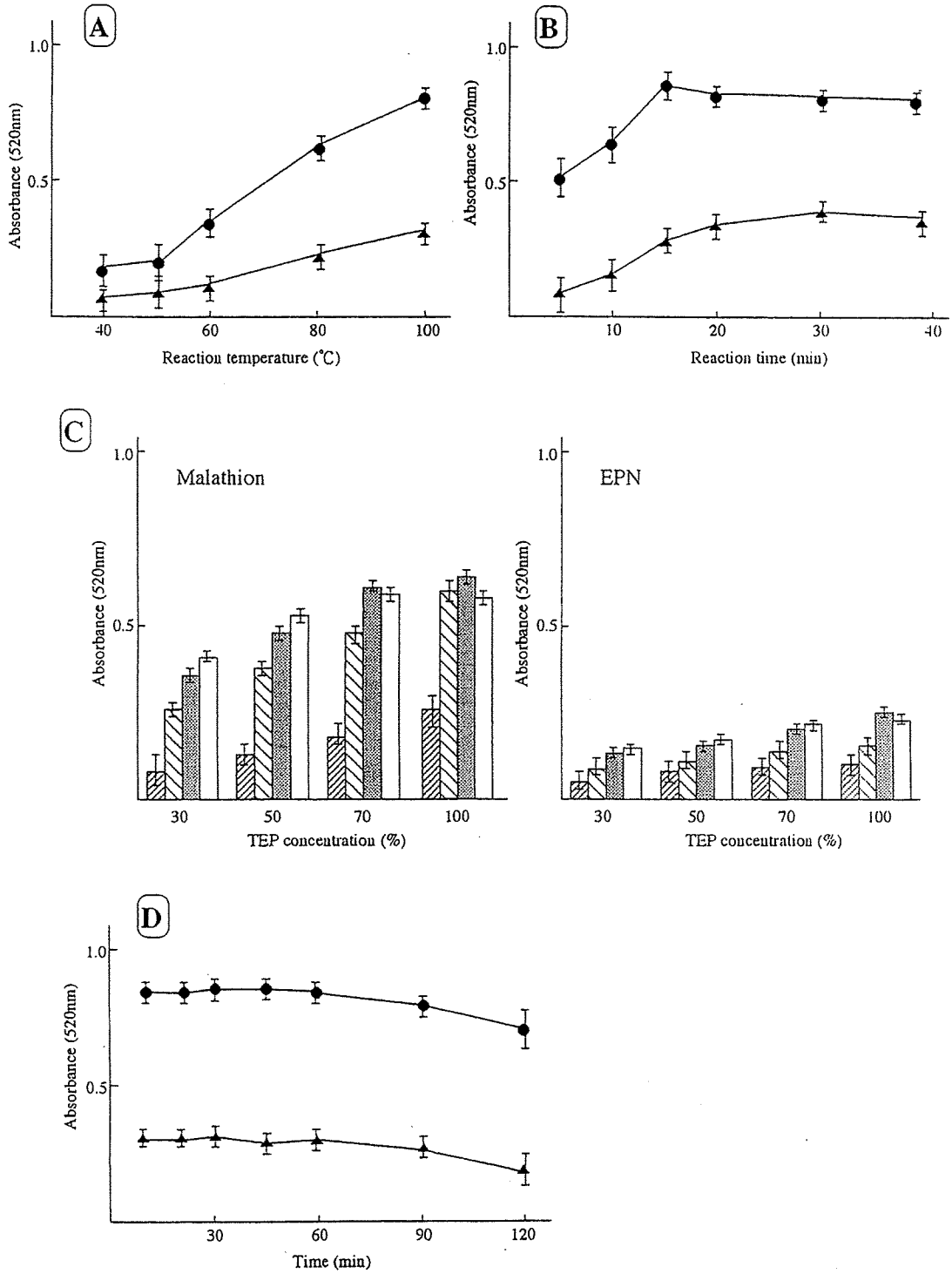


Fig. 2. The effect of reaction temperature (A), reaction time (B), reagent amounts (C) on absorbance and the stability (D) of the coloring complex in the proposed method. ●: malathion, ▲: EPN, : 10% NBP, : 20% NBP, : 30% NBP, : 45% NBP.

malathion and EPN complex were reached at 15 and 20 min, respectively. Therefore, the heating condition was adopted at 100°C for 20 min.

Sane and Kamat [13] reported that the reaction was complete after 4 min. In our study, however, the reaction required 20 min. It is possible that endogenous substances in urine slows the production of the coloring complexes.

3.2. Optimum conditions

The effects of the added amounts of NBP and TEP are shown in Fig. 2C. The absorbance was increased as the concentration of NBP increased. Maximum absorbance was obtained at a concentration of 45% NBP. Absorbance also increased as the concentration of TEP increased. Based on these results, maximum absorbance was obtained at a concentration of 70% TEP. Taking into account the effect of endogenous matrices, 45% NBP and 100% TEP were adopted in the subsequent studies.

3.3. Suitability of coloring complex

To examine the stability of the produced coloring complex, the absorbance was measured at seven different times after the reaction had completed (Fig. 2D). The adsorption spectrum of malathion is shown in Fig. 3. The same spectra were obtained for all the other tested organophosphates. The absorbance slowly decreased as time passed. The absorbance for the sample left for 120 min was

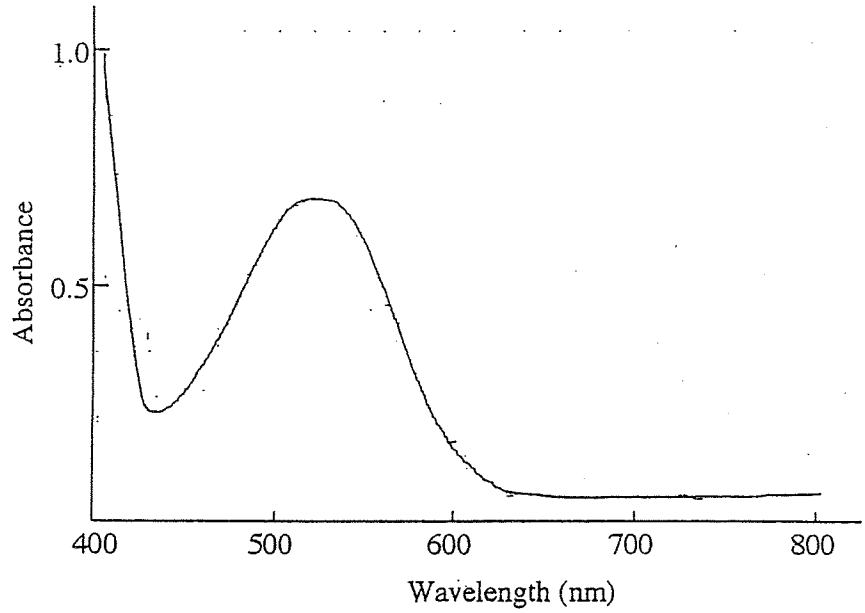


Fig. 3. The adsorption spectrum of the produced color formed with 4-(4-nitrobenzyl)pyridine and malathion.

60–80% of the maximum. It has been reported that ethylene glycol stabilizes the produced coloring complex [14]. As expected, the absorbance rose when ethylene glycol was used. The absorbance of the blank was much higher than that of acetone. For detecting low concentration of organophosphates in urine, the lower absorbance of the blank is necessary. Therefore, acetone was used as the solvent for NBP.

3.4. Urine pH

Fig. 4 shows the effect of the tested urine pH. When the pH of urine was in the vicinity of neutral, change in absorbance was not observed. Absorbance decreased rapidly when pH of urine reached 11. It is well-known that organophosphates are rapidly hydrolyzed to monoalkylphosphate and dialkylphosphate in alkaline condition. It seems that the absorbance decreased rapidly because organophosphates were hydrolyzed in the alkaline urine. If the pH of urine is not neutral, it is necessary to adjust the pH. However, in our laboratory, the pH values of more than 300 clinical urine samples were investigated, and 91% of the samples were between pH 5.0 and 7.9 [15]. Therefore, no pH conditioning was required in the urine samples.

3.5. Precision

The reproducibility for data analysis of 27 organophosphates at the concentration of 50 μg/ml are summarized in Table 1. The precision ranged from 0.30 to 4.2%, except for acephate, ESP, isofenphos, and vamidothion. Consider-

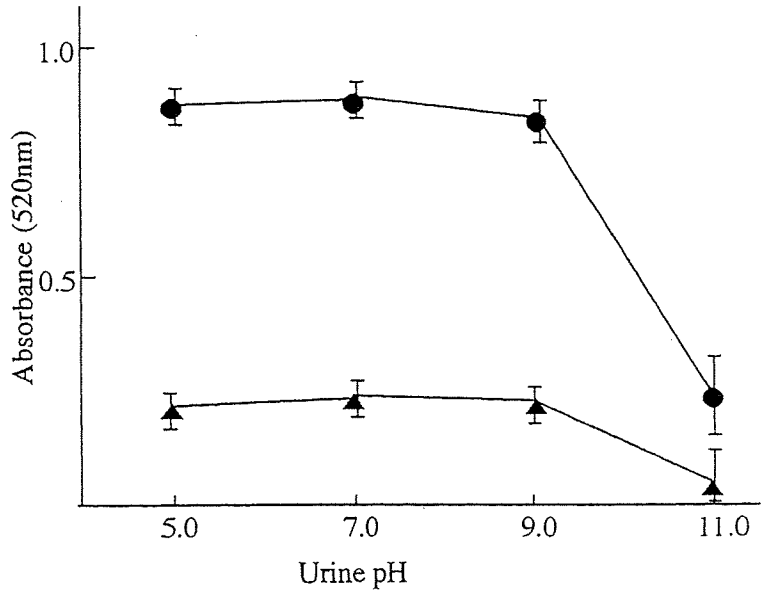


Fig. 4. The effect of urine pH on absorbance.

Table 1
Precision of 27 organophosphates for colorimetric determination

Compounds	Detection limit (μg/ml)	Molar absorptivity	CV (%) ($n = 5$)
Acephate	10	278	10.7
Chlorpyrifos	0.3	5708	1.30
Cyanox	0.3	5773	0.63
Ciazinon	0.3	3756	0.62
Dichlorvos	0.1	6236	0.30
Dimethoate	0.3	3808	1.06
Disyston	3.0	1399	1.18
Edifenphos	0.3	4637	0.50
EPN	3.0	1135	4.20
Estox	10	814	6.10
Fenitrothion	0.3	5469	1.96
Fenthion	0.3	3058	3.09
Formathion	0.5	4307	0.37
Isofenphos	10	822	18.9
Isoxathion	0.5	4814	1.15
Malathion	0.3	4019	1.06
Methidathion	0.1	6498	1.81
Monocrotophos	1.0	2059	8.23
Naled	0.3	4263	0.65
Parathion	0.5	2061	1.76
Phentoate	0.3	4138	1.13
Prothiophos	0.3	2624	1.00
Pyridaphenthion	0.3	4358	3.89
Salithion	0.3	4992	4.02
Tetrachlorvinphos	0.3	4358	3.71
Trichlorfon	0.3	3853	0.87
Vamidothion	10	550	6.17

ing the coefficients of variation for the GC–MS method, the proposed method was found to be reproducible. The precision of acephate, ESP, isofenphos, and vamidothion were 10.7, 6.1, 18.9, and 6.17%, respectively. This imprecision in the proposed method was lower because the molar absorptivity of the compounds were lower. These four compounds have a similar structure (acetamide, carboxyamide, –N–P(=S)– or –S–P(=O)–). We suggest that organophosphates which have these structure, the precision will be limited.

The detection limits of organophosphorus pesticides were 0.10–10 μg/ml in urine (Table 1). Comparing with a commercial screening kit using a cholinesterase assay, the proposed method gave higher sensitivity and selectivity. Only organophosphorus pesticides showed a positive reaction. No coloring was obtained from other agricultural chemicals.

Table 2
Results of the produced colorimetric method and the GC–MS method in 12 poisoning cases

Entry	Case (age, sex)	Colorimetric method	Commercial screening kit	GC–MS method	
					μg/ml
1	45, M	Purple	Positive	Trichlorfon	450
2	70, F	Purple	Positive	Trichlorfon	2240
3	23, M	Purple	Positive	Fenitrothion	3.7
4	62, F	Purple	Positive	Fenitrothion	65.1
5	62, F	No color change	Negative	Fenitrothion	N.D.
6	44, M	Purple	Positive	Methidathion	65.1
7	32, M	Purple	Positive	Malathion	73.3
8	50, M	Purple	Positive	Malathion	101
9	73, F	No color change	Positive	Propanil	2.7
				Carbaryl	1.9
10	83, F	No color change	Negative	Cartap	N.D.
				Dihydronereistoxin	88.9
11	63, M	No color change	Negative	Propanil	6.0
				Carbaryl	0.4
12	64, M	No color change	Negative	Paraquat	6.4

3.6. Application for poisoning cases

The proposed method was applied to 12 poisoning cases who ingested organophosphates or other agricultural chemicals. Eight of the cases had ingested organophosphates, three cases carbamates, and the other glyphosinate (Table 2).

The commercial screening kit showed a positive result for the eight cases who had ingested organophosphates, and this kit could not have selectivity for a suspected poisoning cases who ingested organophosphates or other agricultural chemicals. The proposed method gave only positive results in the cases in which organophosphates were detected by GC–MS. In four cases (entry no. 5, 10, 11 and 12), both this colorimetric method and the commercial kit were negative. No organophosphates were also detected using the GC–MS method in these four cases. The results were in good agreement with that of the GC–MS.

4. Conclusion

A simple method for qualitative detection of organophosphorus pesticides in human urine was developed by detecting the coloring which was produced by the organophosphorus pesticides and 4-(4-nitrobenzyl)pyridine (NBP). When NBP was added to 1.0 ml of a urine sample and the mixture was heated at 100°C

for 20 min, the best results were obtained. Organophosphorus pesticides showed a characteristic purplish blue color. The detection limits were 0.10–10 μg/ml in urine. Comparing with the results of the GC–MS method, the results were in good agreement for the 12 poisoning cases. Thus, the proposed method is useful for qualitative analysis of these pesticides in critical care practices.

References

[1] National Research Institute of Police Science. Annual case reports of drug and toxic poisoning in Japan. Natl Res Inst Police Sci 1989–1998;31–40.
[2] Abbott DC, Egan H. Determination of residues of organophosphorus pesticides in foods. A review. Analyst 1967;92:475–92.
[3] Tsunoda N. Simultaneous determination of organophosphorus pesticides by thin-layer chromatography. Eisei Kagaku 1986;32:447–54.
[4] Erdmann F, Brose C, Schutz H. A TLC screening program for 170 commonly used pesticides using the corrected R_f value. Int J Leg Med 1990;104:25–31.
[5] Liu J, Suzuki O, Kumazawa T, Seno H. Rapid isolation with the Sep-Pak C_{18} cartridges and wide bore capillary gas chromatography of organophosphate pesticides. Forensic Sci Int 1989;41:67–72.
[6] Miyazaki T, Yashiki M, Chikasue F, Kojima T, Hibino H. A case of death from prothiophos poisoning. Forensic Sci Int 1988;30:13–9.
[7] Suzuki O, Hattori H, Asano M. Detection of malathion in a victim by gas chromatography/negative ion chemical ionization mass spectrometry. Z Rechtsmed 1985;94:137–43.
[8] Yashiki M, Kojima T, Une I. Determination of dipterex in biological materials by gas chromatography and gas chromatography–mass spectrometry and the analysis of the distribution of dipterex in a case of fatal poisoning. Nippon Hoigaku Zasshi 1982;36:426–30.
[9] Sharma VK, Jadhav RK, Rao GJ, Saraf AK, Chandra H. High performance liquid chromatographic method for the analysis of organophosphorus and carbamate pesticides. Forensic Sci Int 1990;48:21–5.
[10] Koenigs E, Kohler K, Blindow K. Chem Ber 1925;58:B933.
[11] Watts R. 4-(*p*-Nitrobenzyl)pyridine, a new chromatographic spray reagent for the organophosphate pesticides. J Assoc Off Agric Chem 1965;48:1161–3.
[12] Kramer DN, Gamson RM. Analysis of toxic phosphorus compounds. Anal Chem 1957;29(12):21A–8A.
[13] Sane RS, Kamat SS. Simple colorimetric method foe determination of organophosphate insecticides in technical materials and formulations. J Assoc Off Agric Chem 1982;65:40–2.
[14] Suzuki M, Sano A, Takitani S. Qualitative and quantitative analysis of alkylating antitumor drugs with 4-(4-nitrobenzyl)pyridine. Iyakuhin Kenkyu 1986;17:1083–7.
[15] Namera A, Yashiki M, Okada K, Iwasaki Y, Kojima T, Ohtani M, Tsukue I. Clinical evaluation of Triage for drugs of abuse [in Japanese]. J Med Pharm Sci 1997;37:723–31.

４）規制薬物などの簡易検査（Triage）

試料：尿（乱用者の尿）

国際試薬株式会社

魚住　勝

薬毒物分析システム（Triage）

トライエージは、尿中の一定の濃度以上の乱用薬物やその代謝物を定性的に、迅速に検出できるものである。トライエージは、免疫化学的方法（ASCEND マルチイムノアッセイ法）を利用した方法で、化学的に標識した薬物（薬物抱合体）と尿中に存在する薬物が抗体結合部位を奪い合う、競合的免疫学的測定法である。

【試料】

尿（採取後、－25℃で冷凍保存）

【分析条件】

迅速競合的免疫学的測定法

【用意する試薬】

Triageキット（本測定に必要な試薬等は全てキットに含まれている）

【操作手順】

1）反応カップカバーをはずし、試料140μlを反応カップに注入する。

2）試料で反応カップ内の3種類の錠剤を完全に溶解させたのち、室温で10分間放置する。

3）きれいなピペットチップを用いて、反応カップ内の混合物を薬物検出ゾーンに移し、完全に染み込ませる。

4）キットに含まれる洗浄液3滴で薬物検出ゾーンを洗う。

5）5分以内に結果の判定を行う。

【コメント】

1）本キットにより、ベンゾジアゼピン類（BZO）、コカイン代謝物（COC）、覚醒剤（AMP）、大麻代謝物（THC）、バルビツール酸（BAR）、オピエイト（ヘロイン、モルヒネ）（OPI）、フェンシクリジン（PCP）、三環系抗うつ剤（TCA）の一斉簡易分析が可能である。

2）本検査は、予備的試験結果を得るための方法であるので、確定分析結果を得るためには、より特異性の高い方法（ガスクロマトグラフィー/質量分析法など）を使用する。

3）この試験で陰性の結果が出たとしても、カットオフ濃度より低濃度の薬物が尿検体中に存在しないとは言い切れない。

4）トライエージでの各化合物のカットオフ値を下記に示した。

化合物略号	化合物名（群）	カットオフ濃度
PCP	Phencyclidine	25 ng/ml
BZO	Benzodiazepines	300 ng/ml
COC	Cocaine (Benzoylecgonine)	300 ng/ml
AMP	Amphetamines	1,000 ng/ml
THC	11-nor-Δ9-THC-9-carbaxylic acid	50 ng/ml
OPI	Opiates	300 ng/ml
BAR	Barbiturates	300 ng/ml
TCA	Tricyclic Antidepressants	1,000 ng/ml

＊注）これらの濃度は、米国乱用薬物・精神衛生サービス管理局（SAMHSA）[前米国国立乱用薬物研究所 (NIDA)]が奨励するスクリーニング検査のカットオフ濃度を参考にしている。

・血液試料への適用

【操作手順】

1）血液 1g を試験管にはかりとる。

2）スルホサリチル酸 50mg を加え、良く撹拌する。

3）3,500 rpm で5分間、遠心分離する。

4）上清を新しい試験管に移す。

5）酢酸アンモニウム 25mg を加えて、中和する。

6）3,500 rpm で5分間、遠心分離し、上清をトライエージで検査する。

【コメント】

1）スルホサリチル酸を入れた後の遠心は、十分に行う。（回転数が不足すると、十分な量の上清が取れないことがある。）

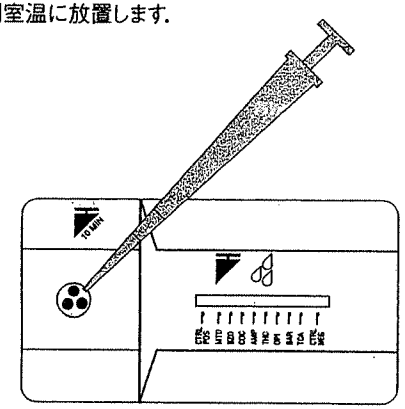
2）この手順では、THC は検出できない。

Triage 8

乱用薬物スクリーニング検査キット　トライエージ8　操作法

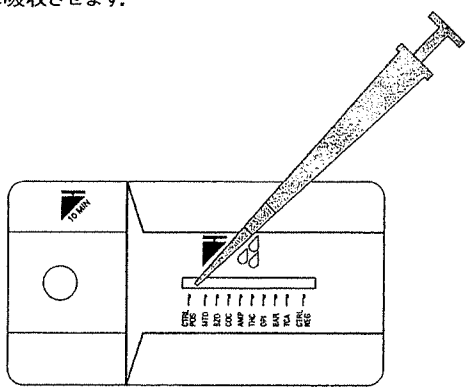
Step 1：サンプルの添加

反応槽のキャップを慎重にとりはずし，ピペットを用いてヒト尿を0.14mL 添加します.
10 分間室温に放置します.



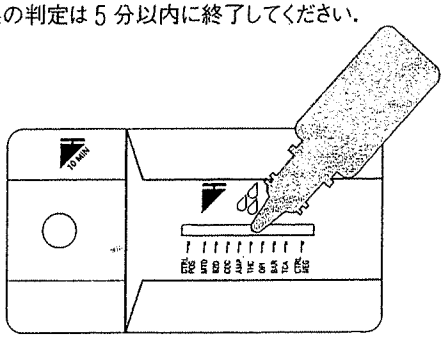
Step 2：反応液の移動

ピペットに清浄なチップをつけて，反応槽から反応液の全量を薬物検出ゾーンへ添加します.
完全に吸収させます.



Step 3：洗浄と判定

洗浄液を3滴，薬物検出ゾーンの中心へ滴下します.
完全に吸収させ，検査結果を判定します.
陰性コントロールゾーン，陽性コントロールゾーンそして薬物検出ゾーンの順に読みます.
結果の判定は 5 分以内に終了してください.



〈測定原理〉
尿中の薬物を抗原抗体
反応により検出します.

検出限界濃度		ng/mL
MTD	メサドン(合成麻薬)	300
BZO	ベンゾジアゼピン類(向精神薬)	300
COC	コカイン，ベンゾイルエクゴニン等(コカイン系麻薬)	300
AMP	アンフェタミン，メタンフェタミン(覚せい剤)	1000
THC	テトラヒドロカンナビノール(大麻)	50
OPI	モルヒネ，ヘロイン等(モルヒネ系麻薬)	300
BAR	バルビツール酸類(向精神薬)	300
TCA	三環系抗うつ剤	1000

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BIOSITE DIAGNOSTICS

(9F0011V-A)

Triage 8

乱用薬物スクリーニング検査キット **トライエージ8**

結果の記録

検体名：＿＿＿＿＿＿＿＿＿＿

採取日時：＿＿＿＿年＿＿＿月＿＿＿日〔午前・午後〕＿＿＿時＿＿＿分

メモ：

- -

- -

- -

測定者名：＿＿＿＿＿＿＿＿＿＿ ㊞ 〈日付〉＿＿＿年＿＿月＿＿日

確認者：＿＿＿＿＿＿＿＿＿＿ ㊞ 〈日付〉＿＿＿年＿＿月＿＿日

測定結果

CTRL POS	(陽性コントロール)	陽性・陰性
MTD	(メサドン)	陽性・陰性
BZO	(ベンゾジアゼピン類)	陽性・陰性
COC	(コカイン，ベンゾイルエクゴニン等)	陽性・陰性
AMP	(アンフェタミン，メタンフェタミン)	陽性・陰性
THC	(テトラヒドロカンナビノール)	陽性・陰性
OPI	(モルヒネ，ヘロイン等)	陽性・陰性
BAR	(バルビツール酸類)	陽性・陰性
TCA	(三環系抗うつ剤)	陽性・陰性
CTRL NEG	(陰性コントロール)	陽性・陰性

トライエージ８のロット番号：＿＿＿＿＿＿＿＿＿＿

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BIOSITE DIAGNOSTICS

5）アセトアミノフェンの簡易検査

試料：血清

東和科学

八十島　誠

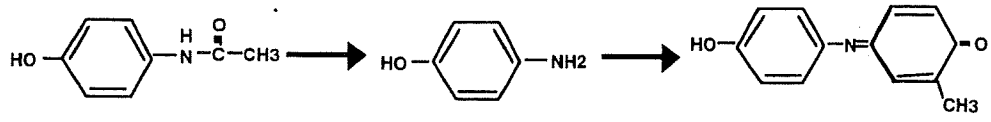
アセトアミノフェンの簡易検査（血清）

東和科学（株）八十島　誠

1. アセトアミノフェンについて
   - アセトアミノフェンは，アスピリンと並ぶ解熱鎮痛剤である．
   - 大量のアセトアミノフェンが体内に入ると，グルタチオンが枯渇し，解毒しきれないN-アセチルベンゾキノミニンがタンパクと結合して肝細胞壊死を起こす．
   - 近年では，アセトアミノフェン中毒例が多く報告されている．
   - アセトアミノフェンによる肝障害の発生とアセトアミノフェン濃度には密接な関係がある．
   - 簡便に血清中アセトアミノフェン濃度が検出できれば，肝障害の危険性を予知できる．

2. 本法の目的
   - 簡便に血清中アセトアミノフェン濃度を知る．

3. 本法の特徴
   - 本法は，インドフェノール反応を利用したものである．
   - 30分程度でおおよその血清中アセトアミノフェン濃度を知ることができる．
   - アセトアミノフェン濃度に比例して，青色が濃くなることから，目視判定が容易である．
   - 5～300 μg/ml の範囲で定量可能であった．（治療域；10～20 μg/ml）
   - 本法の反応は，以下のとおりである．



Acetaminophen　　IndophenolBlue

4. 分析方法

・血清 1g に，20%TCA（トリクロル酢酸）を 0.2ml 加えて攪拌する．
・ 3000rpm で 2 分間遠心分離し，上清をアンプルに分取する．
・濃塩酸をパスツールピペットで 1 滴加える．
・アンプルを封印し，アルミブロックにて 130℃で 20 分間加熱する．
・放冷後，1% o-クレゾール 1ml，アンモニア水 1ml を順に加える．
・攪拌し，呈色度合いからアセトアミノフェン濃度を知る．