

False positives and false negatives with a cocaine-specific field test and modification of test protocol to reduce false decision

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Abstract

The specificity of the Scott test, which is widely used in the field to detect cocaine, was investigated. Several drugs and medicines were applied to the test, and the conditions leading to false positives or false negatives were defined. The Scott test consists of three steps, each involving the addition of a certain reagent and observation of the color that consequently develops. In the first step, blue precipitates appear. In the second, these precipitates completely disappear. In the third step, blue appears again, but in the lower layer. It became clear that proper sample size is critical for correct decision, since too much heroin or dibucaine showed exactly the same color sequence as cocaine HCl and thus gave false positives, and too much cocaine HCl showed persisting precipitates in the second step, yielding a false negative. The appropriate sample size was 1 mg or smaller. Freebase (crack) cocaine gave false negatives even when the sample size was appropriate, and it could not be distinguished from a newer substance of abuse, 5-methoxy-*N,N*-diisopropyltryptamine (5-MeO-DIPT, foxy). The authors developed a new protocol to distinguish crack from 5-MeO-DIPT.

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1. Introduction

Various field tests are utilized to identify suspected illicit substance at the site of abuse or trafficking. The Scott test is widely used in the field to identify cocaine. Some kinds of recent designer drugs produce results similar to cocaine in the Scott test, possibly leading to incorrect on-site decision. In February 2004, three boys were arrested in this manner by police in Tokyo; ultimately it became clear that their substance was not cocaine but an uncontrolled drug [1].

The Scott test was developed by Scott in 1973 [2] and improved by Fasanello and Higgins, who made it applicable to crack [3]. This improved version of the Scott test is now

included in the field test manual of the United Nations [4] and is used by many Japanese law enforcement officers. Though the Scott test is widely used, few reports have examined it in detail.

The test consists of three steps, each involving the addition of a certain reagent and observation of the color that develops as a result. If a sample contains cocaine, the reactions will go as follows. In the first step, cobalt thiocyanate is added and blue precipitates appear. In the second step, hydrochloric acid is added and the blue precipitates completely disappear. In the third step, chloroform is added and blue reappears, but this time in the lower layer (Fig. 1A).

Several forensic chemists have reported a specificity problem with the Scott test. Prall described that diphenhydramine hydrochloride, chlorpromazine hydrochloride, and some other medicines showed the same color sequence as cocaine hydrochloride [5]. Ishiguro et al. reported the same

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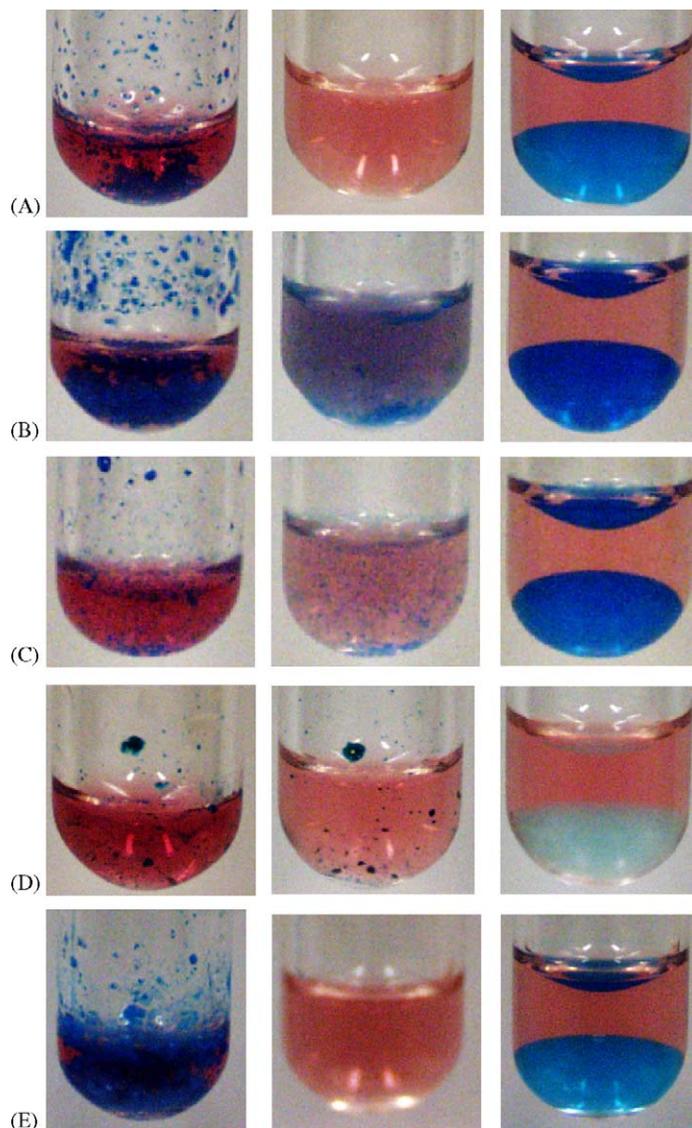


Fig. 1. Reaction sequences of drugs in Scott test: (A) cocaine HCl, 1 mg; (B) cocaine HCl, 3 mg; (C) crack, 2 mg; (D) 5-methoxy-*N,N*-diisopropyltryptamine, 1 mg; (E) heroin, 5 mg. Each photo corresponds to a reaction step in the Scott test. Left: step 1; center: step 2; right: step 3.

phenomenon about promazine hydrochloride and scopolamine [6]. Lorch reported that promethazine alone or phenacyclidine alone did not behave like cocaine in the test, but that mixing them together caused a false positive [7]. Lorch found that the combination of phenacyclidine with either promazine, dibucaine, or methapyrilene showed a false positive [8]. Grant et al. stated that tests for cocaine based on cobalt thiocyanate would continue to show an unacceptable incidence of false positives and false negatives. They suggested another field test for cocaine based on the recognition of the odor of methyl benzoate as a test product [9,10].

On the other hand, several authors have recognized the superior specificity of the Scott test for cocaine. Inoue et al.

applied this test to 105 substances and found none of them showed cocaine's color sequence [11]. Likewise, Oguri et al. applied 30 substances to the test and noted its high specificity [12]. With all these discussions, cobalt thiocyanate tests such as the Scott test are still the most popular field tests for cocaine.

The aim of our study is to clarify the conditions that cause false negatives and false positives in the Scott test and to improve the test's specificity. Some of this study's findings are expected to lead law enforcement officers to more accurate diagnoses on site. The findings are also expected to help forensic chemists obtain better analytical information in laboratories with limited equipment, where spot tests are still important [13].

2. Materials and methods

2.1. Scott reagents and test protocol

2.1.1. Original Scott reagents [2]

- Solution #1: 2% cobalt(II) thiocyanate dissolved in water and then diluted 1:1 with glycerine.
- Solution #2: concentrated hydrochloric acid.
- Solution #3: chloroform.

After being weighed, the sample powder or crystal was placed in a test tube to which 0.2 ml of solution #1 was added, and the tube was shaken. Blue precipitates then appeared (step 1). Then 0.05 ml of solution #2 was added and the tube was mechanically shaken at 1200 rpm for 2 min. The shaking machine was a Tube Mixer TRIO HM-2, a product of As One, Inc. If not all of the blue disappeared, 0.05 ml solution #2 was added and the tube was shaken again (step 2). Finally, 0.1 ml of solution #3 was added, the tube was shaken, and the color of the lower layer was observed (step 3).

2.1.2. Scott reagents applicable to crack [3]

- Solution #1: a solution consisting of 2% cobalt(II) thiocyanate in 10% acetic acid was prepared and then diluted 1:1 with glycerine.
- Solution #2: 10% hydrochloric acid [14].
- Solution #3: chloroform.

After being weighed, the sample powder or crystal was placed in a test tube, 0.2 ml of solution #1 was added, and the tube was shaken. Blue precipitates appeared (step 1). Then, 0.2 ml of solution #2 was added, and the tube was mechanically shaken at 1200 rpm for 2 min (step 2). Finally, 0.2 ml of solution #3 was added, the tube was shaken, and the color of the lower layer was observed (step 3).

2.2. Marquis test

Marquis reagent was made by adding one drop of formaldehyde solution to 1 ml of concentrated sulfuric acid. The test sample was placed in a well on a white porcelain plate, onto which two drops of Marquis reagent were added. The color that developed was noted.

2.3. Drugs and medicines

The standard cocaine HCl for quantitative analysis was Japanese Pharmacopoeia (JP) grade. Illegally traded cocaine HCl used in the Scott test experiments had been seized by Japanese police and were legally possessed by our laboratory for the purpose of research. Crack cocaine was made by dissolving powdered cocaine HCl in water, adding NaHCO₃, and heating the resultant mixture. As it cooled, crystals of this freebase formed and were filter-separated from the NaCl

solution [15]. Some of the crystals were re-crystallized from diethylether to get higher-purity crystals. The heroin HCl used had also been obtained from police seizures. Phencyclidine HCl was provided by the Japanese Ministry of Health and Welfare. 5-Methoxy-*N,N*-dimethyltryptamine and 5-methoxy-*N,N*-diisopropyltryptamine HCl were purchased on the market and identified by comparing melting point, mass spectral data, infrared spectral data, and NMR data with data from the literature [16–20].

Dibucaine HCl, lidocaine, procaine HCl, and promethazine HCl were JP grade. Chlorpromazine HCl, diphenhydramine HCl, ketamine HCl, scopolamine HBr (3H₂O), and tryptamine HCl were all laboratory grade.

2.4. Quantification of cocaine in seized samples

The purity of seized cocaine HCl and laboratory-made crack cocaine was assayed using high-performance liquid chromatography (HPLC). About 0.010 g of sample was precisely weighed, placed in a volumetric flask, to which water (for cocaine HCl) or 0.36% hydrochloric acid (for crack cocaine) was added until 20 ml was reached. The solution was diluted five-fold by water and injected into the HPLC apparatus. The chromatographic conditions were as follows: column, Zorbax extend C18 (15 cm × 4.6 mm, 3.5 μm); mobile phase, 10 mM ammonium acetate and acetonitrile (70:30); flow rate, 0.5 ml/min; oven temperature, 35 °C; wavelength for detection, 235 nm; injection volume, 5 μl.

2.5. Spectrophotometer

A Bacharach Coleman Model 35 spectrophotometer was used to obtain the absorbance data on the colored test solutions at 625 nm. A Shimadzu Model UV-2500 PC spectrophotometer was used to obtain the solution spectra. A Shimadzu Model FTIR-8200 PC infrared spectrophotometer was used to obtain the infrared spectra of crystalline chemicals by using the Nujol Mull method. A Varian Model GEMINI 2000 NMR machine (300 MHz) was used to obtain the NMR spectra.

3. Results and discussion

3.1. Scott test with potential false-positive drugs or medicines

Chemicals that had been reported in the literature at least once to have given false positives, along with some of their structural analogs, were selected for application of the Scott test. The sample weight was controlled at 1 mg. The results are shown in Table 1.

No chemical showed the same color sequence as cocaine HCl; this corresponded to the results of Inoue et al. [11] and Oguri et al. [12]. However crack cocaine gave persisting precipitates after the second reagent was

Table 1
Scott test with cocaine and potential false-positive drugs or medicines

Chemicals	Acetic acid ^a			Original ^b		
	Step 1	Step 2	Step 3	Step 1	Step 2	Step 3
Cocaine or its salt						
Cocaine HCl (JP grade)	Blue	Disappear	Blue	Blue	Disappear	Blue
Freebase cocaine A (95%) ^c	Blue	Remain	Blue	White	Remain	Blue
Freebase cocaine B (87%) ^c	Blue	Remain	Blue			
Freebase cocaine C (86%) ^c	Blue	Remain	Blue			
Chemicals yielding blue precipitate						
Chlorpromazine HCl	Blue	Remain	Blue			
Dibucaine HCl	Blue	Disappear	No	Blue	Disappear	No
Diphenhydramine HCl	Blue	Remain	Blue			
Heroin HCl	Blue	Disappear	No			
Ketamine HCl	Blue	Disappear	No			
Lidocaine	Blue	Remain	No	White	Remain	No
5-Methoxy- <i>N,N</i> -dimethyltryptamine	Blue	Disappear	No	Blue	Disappear	No
5-Methoxy- <i>N,N</i> -diisopropyltryptamine HCl ^d	Blue	Remain	Blue	Blue	Remain	Blue
Promethazine	Blue	Remain	Blue	Blue	Remain	Blue
Chemicals not yielding blue precipitate						
Phencyclidine HCl	White	Remain	Blue	Blue	Remain	Blue
Procaine HCl	No	No	No			
Scopolamine HBr	White	Disappear	No	White	Disappear	No
Tryptamine HCl	No	No	No	No	No	No

Details of reagents are described in text.

^a Improved Scott test applicable to crack.

^b Original Scott test.

^c Laboratory-made from seized cocaine HCl, and contents of freebase were quantified by HPLC.

^d So-called 'foxy'.

added (Fig. 1C). There are four chemicals that show the same color sequence as crack cocaine: chlorpromazine HCl, diphenhydramine HCl, 5-methoxy-*N,N*-diisopropyltryptamine HCl (5-MeO-DIPT, Fig. 1D), and promethazine HCl. If the complete disappearance of precipitates at the second step was considered requisite for a cocaine-positive decision, a crack cocaine sample would give a false negative. On the other hand, if the persistence of precipitate at the second step was not considered an obstacle to a positive decision, all four of the chemicals would give false positives. Thus it became clear that persisting precipitate is one cause of false decision.

Chlorpromazine and diphenhydramine were included in Inoue's and Oguri's reports as possible false-positive chemicals. However, neither of those authors found a specificity problem with those chemicals, since they did not test crack cocaine and may have neglected the significance of persisting precipitates.

3.2. Effect of sample weight

As sample weight seemed to be critical, various sample weights were used in applying the Scott test to cocaine HCl, crack cocaine, and eight other substances that could lead to false positives. The results are shown in Table 2. Cocaine HCl gave persisting precipitates when it was sampled at

weights greater than 3 mg, as shown in Fig. 1B (false negative.)

Some substances at higher sample weights showed the same color sequence as that of the normal amount of cocaine. An amount of 2 mg of dibucaine HCl or heroin HCl (Fig. 1E), and 4 mg of ketamine HCl produced such sequences (false positives.)

Thus, a sample must weigh no more than 1 mg for precise decision with the Scott test. Amounts over that will cause false negatives in the case of cocaine or false positives in the case of dibucaine, heroin, or ketamine. This is the first report of a correlation between sample weight and test results. For on-site testing, the capacity of the spoon or spatula should be measured, and law enforcement officers should be trained in the proper sampling amounts of suspected drugs.

3.3. Spectral data of final solution

Spectral data for the complex yielded in the Scott test have not been available in the literature. We took the spectral data of some substances within the range of visible wavelengths, as shown in Fig. 2. The wavelength range of maximum absorbance was 622 to 626 nm. The color tones of these complexes are so similar that they are indistinguishable from each other.

Table 2

Scott test with cocaine and potential false-positive drugs at various sample weights

Chemicals	Sample weight (mg)							
	0.1 ^a	0.2 ^a	0.5 ^a	1	2	3	4	5
Cocaine or its salt								
Cocaine HCl (JP grade)				A	A	B		
Freebase cocaine A (95%) ^b	A	A	B	B				
Freebase cocaine B (87%) ^b	B	B	B	B				
Freebase cocaine C (86%) ^b	B	B	B	B				
Chemicals yielding insoluble precipitate								
Chlorpromazine HCl	C	C	B	B				
Diphenhydramine HCl	C	C	B	B				
Lidocaine				C	B			B
5-Methoxy- <i>N,N</i> -diisopropyltryptamine HCl	C	C	C	B				
Promethazine HCl		C	B	B				
Chemicals showing color sequence like cocaine								
Dibucaine HCl				D	A			A
Heroin HCl				D	A			A
Ketamine HCl				D	D	D	A	A

Pattern of color sequence at 1st, 2nd, and 3rd steps; A: + + +, B: + - +, C: + - -, D: + + -; a certain weight of each drug was applied to the Scott test with 0.2 ml of solutions #1, #2, and #3.

^a Actual weight in the experiments was 1 mg. Multiplied volumes of Scott reagents were used and calculated to each sample weight.

^b Laboratory-made from seized cocaine HCl, and contents of freebase were quantified by HPLC.

3.4. New protocol to distinguish between crack cocaine and 5-MeO-DIPT

Distinguishing between crack cocaine and 5-MeO-DIPT by the Scott test is considered difficult, since both substances

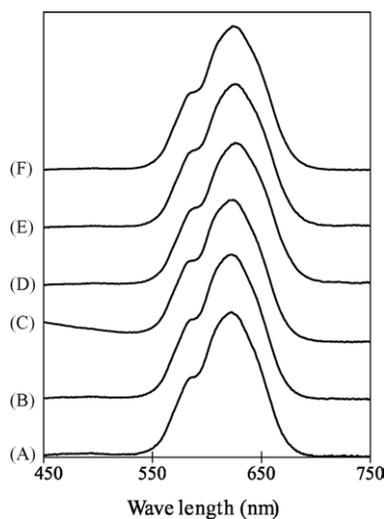


Fig. 2. Spectra of final solutions of the Scott test for some kinds of drugs. The wavelength of maximum absorbance of each mixture is shown in parentheses. (A) Cocaine HCl (622.6 nm); (B) crack (622.4 nm); (C) 5-methoxy-*N,N*-diisopropyltryptamine (624.2 nm); (D) heroin HCl (626.0 nm); (E) chlorpromazine HCl (626.0 nm); (F) diphenhydramine HCl (623.8 nm).

give the same color sequence as each other even when the sample weight is controlled. We tried to use an additional amount of solution #2, hydrochloric acid, to dissolve precipitates in a test mixture. However, excess hydrochloric acid produced a blue color in the second step, and this blue could not be extracted into the chloroform layer, as Scott reported [2].

In the case of persisting precipitate, one method of preventing 5-MeO-DIPT or other chemicals from showing a false positive is to place the supernatant of the second-step mixture into another test tube and then add chloroform to it. With this method, the blue color in the lower layer will develop only with cocaine or crack. However, this method is not very convenient for on-site use.

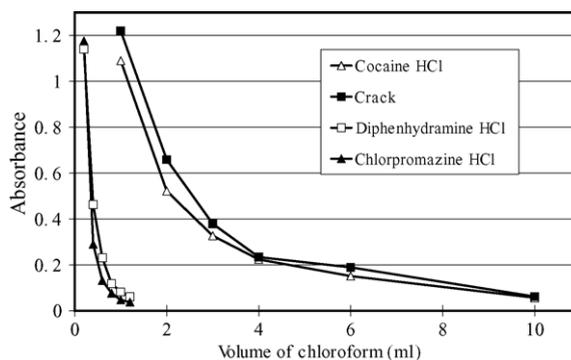


Fig. 3. Absorbance of lower layer in final reaction mixture at 625 nm.

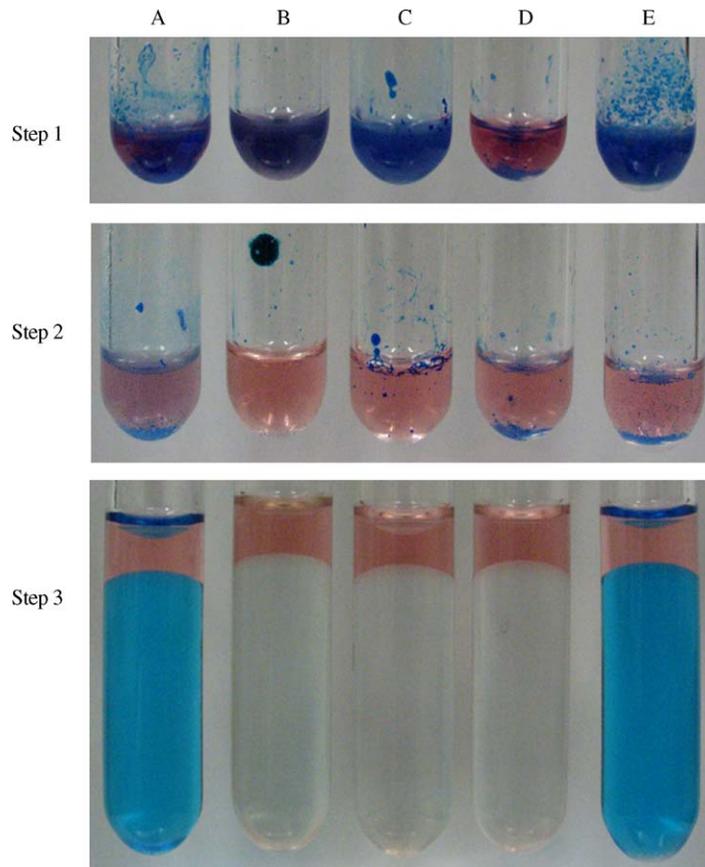


Fig. 4. Final mixture of modified protocol for persisting precipitates. (A) crack, 2 mg, (B) 5-methoxy-*N,N*-diisopropyltryptamine, 2 mg, (C) diphenhydramine HCl, 1 mg, (D) chlorpromazine HCl, 1 mg, (E) cocaine HCl, 3 mg.

On the other hand, concentration–absorbance curves suggested that crack and 5-MeO-DIPT are distinguishable from each other. Fig. 3 shows the absorbance (625 nm) of the lower layer in a final mixture of the Scott test when it was diluted stepwise. Diphenhydramine and chlorpromazine were used in this experiment because they show same color sequence as 5-MeO-DIPT and the color is stronger. The reaction solutions of diphenhydramine and chlorpromazine gave absorbance of 0.1 with 0.8 ml or 0.6 ml of chloroform, respectively. At that absorbance, the blue was so slight that it could not be seen by the naked eye. On the other hand, the third-step colors for cocaine and crack were quite deep, and about 8 ml of chloroform was needed to dilute either of them to reach an absorbance of 0.1.

Based on the strong absorbance of the cocaine–cobalt complex, a new protocol for the Scott test has been developed: If the precipitate in the second step does not disappear completely, add a larger volume of a third solution to the test mixture. If 2 ml of a third solution is added, at least 2.5 mg diphenhydramine HCl is necessary for blue to appear in the third step. On the other hand, 0.25 mg of crack or cocaine HCl will give a blue color in the same conditions. The efficacy of this increase in solution 3 is shown in Fig. 4.

3.5. Additional screening tests

Prall recommended the use of Marquis reagent as an additional test to screen out false-positive results for cocaine by chlorpromazine, diphenhydramine, doxylamine, and diphenylpyraline [5]. We applied these potential false-positive substances to the Marquis test. Dibucaine and ketamine were colorless in this test and could not be distinguished from cocaine. 5-MeO-DIPT produced a pale green immediately after the addition of Marquis reagent, consistent with a report by the US Drug Enforcement Administration [17] in which 5-MeO-DIPT is reported to show an olive green in the test. However, the color was not strong and it quickly turned into a pale brown that was somewhat indistinct from the pale brown shown by impure cocaine. (Cocaine HCl of 40–70% purity were tested.) 5-MeO-DIPT can be distinguished from cocaine by the Scott test protocol described above, and also by the Ehrlich indole test. In the Scott test, when the sample weights of dibucaine and ketamine were too high, each gave exactly the same color sequence as cocaine. The sample weight should be controlled especially carefully for those two substances.

Diphenhydramine HCl gave a brown color, while heroin, chlorpromazine, and promethazine gave a purple and lido-

caine gave a red. They were distinguishable from cocaine by the Marquis test.

3.6. Influence of mixed materials

Cocaine is sometimes mixed with various materials to increase its volume or for camouflage. These materials may cause incorrect decision in the Scott test. Starch and sucrose were each experimentally mixed with cocaine HCl and applied to the Scott test. Neither material influenced the color sequence in the test, though the solution with starch was slightly cloudy.

4. Conclusions

To maximize the specificity of the Scott test and reduce on-site false decision, the following terms would be effective.

The amount of sample tested has a definite effect on the result of the test therefore the sampling weight should be 1 mg or less.

When blue precipitate does not disappear completely in the second step, 2 ml of a third solution should be added because cocaine gives blue color even with such larger amount of third solution while any other substance tested does not.

Some substances at higher sample weights show same color sequence as that of the normal amount of cocaine. Therefore other field tests, such as the Marquis or Ehrlich indole test, should be done as additional field testing in case the sample weight is not controlled well.

When a field-test sample is suspected of being cocaine, it should be laboratory tested as soon as possible.

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