

Letter to the Editor

Procedures and Responsibilities in Forensic Toxicology

Sir:

Procedures for case work in forensic toxicology may be divided into three areas: the taking of relevant samples, the laboratory analysis, and the interpretation of the findings. In each of them, the forensic toxicologist has certain responsibilities. Based on practical experience and on ongoing methodological and technological innovations, the handling of these responsibilities and procedures is generally considered to be quite adequate. Yet observations of some cases in which we became involved as well as some from the literature seem to indicate that serious problems still occur and that improvements in certain areas are needed. The following cases may serve as relevant examples.

Case 1: Clandestine Amphetamine Manufacturing

The purchase from a chemical wholesaler of a bottle of phenyl acetone by an individual was reported to the authorities, and then his apartment was raided in the presence of a forensic toxicologist. A laboratory setup was found in the kitchen, suggesting that a steam distillation was being carried out. Samples from this process were taken and various chemicals found on the scene—among others a substantial quantity of unused phenyl acetone and solid ammonium formate—were seized as well. The forensic chemist assumed that the accused had been manufacturing amphetamine according to the Leuckart reaction (Fig. 1) and went to work to investigate the various seized samples from the steam distillation setup, using as identification techniques gas chromatography on SE-30, thin-layer chromatography, the Marquis reaction, the crystal test with gold chloride, gas chromatography/mass spectrometry (electron impact), and the boiling point of what was assumed to be the isolated free amphetamine base. The report of analysis read as follows:

Marquis	(+)	for amphetamine
MS (EI)		amphetamine identified
AuCl ₂		amphetamine identified
GC		amphetamine + methamphetamine
BP		231 to 235°C
TLC		no positive result

Based on this evidence it was concluded that amphetamine was present in the steam distillate samples.

The individual who had bought the phenyl acetone was a chemistry student who had had his own home laboratory for nearly eight years, ordering chemicals from wholesalers throughout this period. His explanation for the events in this case was that he had wanted to investigate if phenyl acetone could be dimerized in an aldol condensation process. He assumed that the phenyl ring may cause steric hindrance (Fig. 2). Instead of sodium acetate as buffer, which he did not have, he had chosen ammonium formate, which was at his disposal.

We were asked by the defense counsel to analyze the various samples and, after some legal battles, small amounts of them were provided by the forensic chemist who had carried out the analysis for the authorities. After careful investigation, using pure amphetamine, methamphetamine, and phenyl acetone as reference materials and taking into account the methods used in the first analysis, we made observations listed in Table 1.

Those observations indicated that in the first analysis the GC peak identified as am-

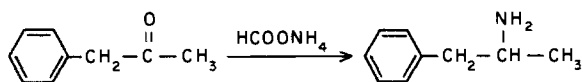


FIG. 1—Amphetamine manufactured according to the Leuckart reaction.

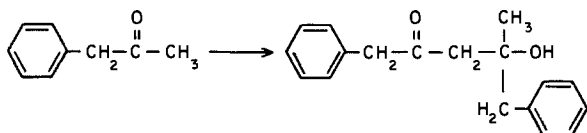


FIG. 2—Steric hindrance.

phetamine on the basis of retention time and quasi-molecular ion could well have been phenyl acetone. The retention times on SE-30 are too close to separate them, and because amphetamine gives a quasi-molecular ion of $M-1^+$ at m/z 134, electron impact mass spectrometry is of very little help here to distinguish from phenyl acetone if the other areas of the spectrum are not taken into account. Note also the relatively large differences for the boiling points of amphetamine and of the material isolated from the case samples. Further analysis then revealed that the GC peak designated earlier as being for amphetamine was in fact for over 99% unreacted phenyl acetone. In addition, it could be established that the samples contained a fair amount of the phenyl acetone dimer, apparently as a result of the aldol condensation process. Accurate quantitation was not possible because of the instability of the dimer. It may well be that the presence of the dimer gave rise to the boiling point of 231 to 235°C and to the orange coloration in the Marquis reaction. Last but not least, however, after enrichment steps, we found that the samples also contained minute amounts of amphetamine, on the order of about 0.5% of the quantity of unreacted phenyl acetone.

Apart from the apparent misidentification of amphetamine in the analysis by the prosecution, the following considerations can be made. It may be argued that the forensic chemist assigned to this case became too much involved and therefore predisposed. As a result, he may have concentrated only on those results that appeared to fit, disregarding negative indications such as the TLC experiments and the boiling point. The report of analysis also was below normal standards and the interpretation grossly inadequate. Obviously, his supervisors are also to blame for lack of control and supervision. The second important point is that there were no separate, untouched samples available for a second analysis nor for a third party in case of a conflict. This may give rise to a series of complications with regard to decomposition, contamination, inadequate handling, or even tampering with the samples.

Case 2: Cannabis in Tobacco

During their summer vacations, two Dutch students were traveling by train through Southern Europe. Also in the same train compartment were two German students. When police authorities, looking for drug smugglers, searched the four students, they found about 2 kg of hashish soles on one of the Germans and took all four into custody. The next day, the Dutch students were informed that *Cannabis* had been found in the "roll-your-own-cigarette tobacco" they had carried in three pouches with an estimated total content of 60 g.

According to the prosecution report, microscopic examination and TLC had been used. The TLC examination had been carried out using the method of Heyndrickx et al [1], which employs silica gel plates, benzene/dimethylformamide 99:1 as developing solvent, and a 1%

TABLE 1—Results of the analyses of the samples.

Test	Amphetamine	Phenyl Acetone	Case Samples
Gas chromatography retention time	2 min, 56 s \pm 4 s	2 min, 52 s \pm 3 s	2 min, 53 s \pm 4 s
Boiling point, °C	200-203	214-216	231-235
Mass spectrometry (electron impact)	M-1 ⁺ = 134	M ⁺ = 134	M ⁺ = 134
Marquis	orange \rightarrow brown	yellow	orange
TLC plus ninhydrin	positive	negative	negative
AuCl ₂	inconclusive	inconclusive	inconclusive

solution of 2,6-dichloroquinone-4-chlorimine in ethanol as visualization reagent. The microscopic examination reportedly had revealed cystolithic and silicotic hairs with the same appearance as those found in hashish and marihuana. The TLC experiments had revealed R_f values similar to those of the main cannabinoids from authentic hashish samples. Yet, to obtain a positive reaction, a rather large sample of some 5 to 10 g reportedly had to be used.

Despite these findings by the prosecution the two defendants categorically denied that they had added anything to their tobacco or that they had been aware that others had—or could have—added something to it. Although at present in many European countries possession of a small amount of hashish or marihuana is no longer considered a major criminal offense, the finding of the 2 kg of hashish soles had led the prosecution to the assumption that all four students belonged to a ring of drug smugglers. Thus, it became of crucial importance whether the tobacco did or did not contain traces of *Cannabis*.

However, an analysis by the defense was refused on procedural grounds and a request for a reexamination of the tobacco samples by the prosecution in the presence of an expert witness for the defense was refused as well.

Faced with these circumstances we could only work with pure tobacco samples and tobacco that we spiked with different amounts of hashish and marihuana. Upon microscopic examination of all drug-free tobacco samples, nonglandular single-celled silicotic and cystolithic hairs were found that looked similar, if not identical, to the nonglandular trichomes present in *Cannabis* preparations. The pure tobacco samples did not show glandular hairs of the *Cannabis* type, but such hairs had not been found in the seized samples either. When working with the TLC system it could be shown that pure tobacco samples may give at least one spot in the cannabinoid area [2]. Based on these observations it was argued in court that the initial findings by the prosecution had to be considered with doubt: glandular hairs had not been found whereas in the TLC investigation the spots reportedly observed in the cannabinoid area may have been due to endogenous tobacco compounds, especially in view of the fact that large amounts of sample (5 to 10 g) were required to get a "positive reaction."

Apart from a lack of thoroughness in the analytical work and the interpretation thereof, the major objections to this case are the refusals with regard to defense examination or a reexamination of the samples. Moreover, the arrest report was scanty and did not state the quantities of tobacco seized. There was no evidence for an adequate chain of custody of the samples. This raised suspicion that the refusals were due to the fact that the samples had been lost, misplaced, or accidentally destroyed. At the court session, however, three tobacco pouches were shown, claimed to be the exhibits. They looked full or nearly full, despite the statements by the defendants that at the time of arrest two pouches had been about half full and the third one almost empty and despite the relatively large quantities reportedly used for the prosecution analysis. A request for visual inspection of the contents of the pouches was refused.

Case 3: Detection of Curare in Embalmed Bodies

Case 3 has become known as the Jascalevich case and has received considerable publicity [3]. In 1966, a surgeon was suspected of killing patients by injecting them with tubocurarine, but the prosecution did not find reasons to file charges. In 1976, after renewed publicity and the activities of a forensic scientist who supplied an affidavit, the case was reopened. Five embalmed bodies were exhumed, and autopsies as well as chemical analysis on the remains were done by a team of experts headed by the forensic scientist. Experts for the defense were not allowed to be present at the autopsies; they received samples of tissues and embalming fluids from the experts for the prosecution more than 18 months after the exhumation and the autopsies. Techniques used for the search for curare were, among others, radioimmunoassay, high performance liquid chromatography, and mass spectrometry.

The experts for the prosecution claimed they found tubocurarine in tissues from four of the five bodies. The defense experts concentrated on the stability of tubocurarine and the distribution of the drug through the living body. Evidence was shown that tubocurarine is unstable in the presence of embalming fluids and tissue fluids and it will also decompose as a result of the influence of underground bacteria and temperature fluctuations. It was claimed, therefore, that tubocurarine could not survive in embalmed bodies buried for ten years. Moreover, studies by the defense showed that, upon injection, tubocurarine is rapidly distributed throughout the body: after 10 min some 40% is found in the muscle and 3% in the liver. Evidence was found for the presence of tubocurarine in the liver of only one body but it could not be detected in the muscle tissue of that body, which seems inconsistent with its distribution characteristics.

The question arises as to whether the forensic scientist who played a role in having the case reopened may have become too involved in the matter. It would have been better if he had not headed the expert team for the prosecution in view of his predisposition. The fact that defense experts were not allowed at the autopsies must also be considered an error and an infringement of the rights of the defendant, especially because evidence or circumstances found at an autopsy may be irreproducible and lost almost immediately, thus making adequate reexamination impossible. Furthermore, the handling of the samples was incorrect: no untouched samples were available for the defense, thus raising the same questions on such issues as decomposition and contamination as were raised in Case 1.

Discussion

Though one might argue that these cases can be considered as solitary incidents, not representative of the state of the art of our profession, it should be remembered that most case work is never reexamined by a second expert and, thus, that many things may go undetected. Of the cases we reexamined over recent years, some 15 to 20% could be considered as having questionable procedures or deficiencies of some sort. The major areas of concern appear to be the following:

1. The involvement of the forensic toxicologist. One of the basic principles for a forensic toxicologist is to perform his duties impartially. Yet, if he gets too involved with certain aspects of a case he may no longer be impartial in other areas. For example, with regard to clandestine drug manufacturing, it is often necessary to assign a forensic toxicologist to the initial investigations as an "analytical detective" to develop suppositions as to what might be taking place [4]. However, such a task may easily lead to predisposition, and so it is recommended that a second forensic toxicologist be assigned to the analysis of evidence in such cases.
2. The use of inadequate techniques and methods. Because of rapid developments and innovations, it is quite difficult for the individual toxicologist to stay current in the field.

Nevertheless, he should always try to assess such concerns as what a certain technique can do for him, what it can prove or disprove, or what the chances for interference are.

3. The interpretation of the findings. Occasionally, one might jump to conclusions that are not warranted; impartiality is required in all observations and *all* findings should be in agreement with one another.

4. Inadequate reports. Police reports describing the start of a case are often unsatisfactory in that exhibits relevant for the toxicologist are not adequately described (weight, size, identification marks, and so on). The descriptions of the handling of the exhibits and of the chain of custody are also critical. The reports of analyses and laboratory notebooks also leave much to be desired: too often they are scanty and do not adequately describe the analytical findings or document a conclusion.

5. Inadequate sampling procedures. All three cases suffered from inadequate sampling procedures in that no untouched samples were available for examination by the defense. This, in our opinion, must be considered a severe violation of the rights of a defendant. It is therefore recommended that samples taken from a scene for forensic toxicological analysis be subdivided and sealed in three equal parts. One part can be used by the prosecution, the second part reserved for the defense, and the third part remain available for third-part investigation if conflicts arise or if otherwise necessary. Such an approach, already standard in various countries for traffic-alcohol investigations and drug control, obviously requires extra effort to implement. Yet, for the cause of justice, this is a necessity, not only with regard to the defendant but also to protect our own profession against charges of improper handling or storage, of contaminating samples, of wasting invaluable pieces of evidence, or of tampering with the samples.

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