Introduction

Drug screening systems are generally dependent upon several limiting factors. In the forensic toxicological examination of samples taken under the British Road Traffic Act (RTA) the most important are as follows:

a) The biological samples available for analysis and their volumes.

In this Laboratory the samples were blood alone (2 millilitres on average) in 66 per cent of cases, urine alone (up to 25 millilitres) in 25 per cent of cases and blood and urine together in 8 per cent of cases.

b) The classes of drug under examination.

In RTA cases we are concerned with those which can impair judgement, psychomotor activity etc. in a way detrimental to driving. This commonly includes sedatives, tranquillizers, stimulants, anti-histamines, anti-convulsants, anti-depressants, hallucinogens and narcotics.

c) The parameters of identification.

Normally at least two independent (uncorrelated) techniques are required for positive identification. (e.g. TLC and GC or GC and Mass Spectrometry). For many drugs detected in blood by radio-immunoassay confirmation poses such a problem that it is the subject of a large proportion of current research in toxicological analysis.
d) The background information available.

In negative or low alcohol RTA cases where drugs analysis is requested, the following information is routinely sought in the form of a questionnaire to the officer in the case as follows:

(i) Were there any drugs in the defendant's possession? If so, what?, where are they now?, could they be submitted for identification?

(ii) What was the defendant's condition? (e.g. drowsy, asleep, agitated etc).

(iii) Did the police surgeon certify impairment?

(iv) Is the subject prescribed any drugs by his own doctor? If so, what?, when was the last dose taken?, how long has he been taking them?.

(v) Is the subject a known drug user? (or registered drug addict).

**Extraction and detection of drugs**

The general scheme for acidic and neutral drugs is shown in Figure 1. Evaporated extracts are dissolved in acetone for TLC on silica gel plates in chloroform/acetone (4:1). Spot location is by UV - short wave absorbance and by spraying with van Urk reagent (for carbamates and benzodiazepines), ferric chloride/ferricyanide reagent (for paracetamol etc) and mercuric chloride/diphenylcarbazone reagent (for barbiturates etc). The same extract is then methylated with diazomethane prior to temperature programmed GC on SP2510DA with dual NPD/FID detection. Peak identification is based on relative retention to methylated cyclopal internal standard and detector response ratio followed by GC on Apiezon L and (if necessary) GC-MS. The general scheme for basic drugs (Figure 2) involves the above mentioned aliquot of urine. TLC screening is performed on caustic silica gel plates in chloroform/methanol (4:1). Spots are located by their UV-short wave absorbance and by spraying separately with:
(i) **NQS (B - naphthaquinone sulphonate) reagent**

(ii) **dilute sulphuric acid (noting UV long wave fluorescence) followed by iodoplatinate reagent.**

This extract is then screened by GC on OV-17 with dual AFID/FID detection. This involves triple temperature isothermal screening (150, 200 & 260°C) using respectively nicotine, diphenhydramine and methaqualone as GC markers. Nowadays there are many basic drugs routinely available and an associated wide range of volatility; therefore this form of isothermal screening is preferred to temperature programmed screening because it is more discriminating and the relative retention data is more reproducible.

The general scheme for detection of benzodiazepines (as benzophenones) and morphine in urine is described in Figure 3. For benzophenones TLC is performed on caustic silica gel plates using chloroform or toluene as eluting solvent. Spots are located by their yellow colour in daylight and the colours produced with

(i) **Bratton-Marshall reagent and**

(ii) **methanolic paradimethylamino - benzaldehyde oversprayed with methanolic trichloro - acetic acid.**

The TLC system for morphine is identical to that described for basic drugs.

The details for blood benzodiazepine screening are described later under batch methods of analysis (Figure 5). Recent improvements in the gas chromatography of benzodiazepines by using OV-7 and OV-225 stationary phases instead of OV-17 alone have provided a more comprehensive screen for the members of this class of drugs plus improved resolution of drugs from co-extractives arising from the rubber septum cap of the blood container. The latter has permitted simplification of the extraction procedure and we now use a one-step 30-second "whirlymix" extraction on 100 µl blood using 100 µl butylacetate as solvent followed by direct injection of an aliquot of the latter onto the GC column.
During the period of this survey, the radio-immunoassays routinely used were those for morphine (Abuscreen), Cannabinoids, Lysergide (LSD) and diazepam.

**Batch method of analysis**

Due to the increasing number of RTA cases requiring drugs analysis, the analysis of those cases involving blood alone in batches of six was investigated. The schemes for analysis are shown in figures 4 and 5. They were found to have the advantages of being time saving (usually only two calibrations involved), more efficient in use of machine time and more convenient in that they enable comparison of case chromatograms and quick detection of spurious peaks from solvent impurities or biological co-extractives. With careful labelling of extraction and evaporation tubes and attention to such details during solvent transfers and analysis, the only disadvantage appears to be the tedious nature of the batch procedure.

**Survey of drugs detected in 1978 RTA cases**

The pattern of frequency of detection of drugs or classes of drug is shown in figure 6. The benzodiazepine class of drugs is the most prevalent being encountered in 38 per cent of cases and being in the form of diazepam in 25 per cent of cases. Sedatives and tranquillizers account for impairment in most of the cases and narcotic analgesics were significantly detected in about 13 per cent of cases. Cannabis, stimulants, anti-histamines, anti-convulsants and anti-depressants considered as a whole account for only 10 per cent of the cases. Other than anti-convulsants, these drugs and classes of drugs are mainly those which prove difficult to screen for in small blood samples. The importance of also having urine submitted in RTA cases is emphasized if we compare percentage detection of drugs in all RTA drug cases with that in such cases where only blood was submitted for analysis (figure 7). With blood alone the percentage detection of stimulants, anti-histamines and anti-depressants falls to zero. It follows that, since two thirds of all cases involved blood alone, the percentage overall detection of these classes of drugs may be a gross underestimate of their involvement in RTA cases. Since radio-immunoassays for amphetamine-type drugs and tricyclic anti-depressants became commercially available this situation has
partly been rectified, the problem now being one of having sufficient blood sample to analyse. The reasons for no significant change in percentage detection of cannabinoids is that RIA forms the sole basis for screening both blood and urine. There are also no significant differences in the percentage detections for benzodiazepines and for barbiturates. As expected the percentage detection of "no drugs" is noticeably higher in RTA cases involving blood alone. A previous survey of this nature indicated a much higher percentage "negatives" in cases involving only blood:

<table>
<thead>
<tr>
<th></th>
<th>1976 (Jan to June)</th>
<th>1978 (Jan to Sept)</th>
</tr>
</thead>
<tbody>
<tr>
<td>RTA cases involving urine</td>
<td>32</td>
<td>49</td>
</tr>
<tr>
<td>Percentage negative</td>
<td>22 ←—</td>
<td>20 ←—</td>
</tr>
<tr>
<td>RTA case involving blood alone</td>
<td>39 17</td>
<td>99 8</td>
</tr>
<tr>
<td>Percentage negative</td>
<td>39 ←—</td>
<td>28 ←—</td>
</tr>
</tbody>
</table>

The approximate one third reduction in the percentage negatives for 1978 "blood only" cases is due to improvement in analytical techniques and, in particular, to the introduction of RIA into casework. Also the much smaller difference in percentage negatives between "blood only" and "urine" cases in 1978 prompts one to question the need to analyse urine. An example of results from an RTA case involving blood and urine (table 1) shows that if blood alone had been analysed, there would have been an incomplete picture of the defendants multiple drug misuse. Indeed one can envisage a competent counsel rightly dismissing a positive RIA result for morphine as being non-specific and also assigning the amylobarbitone and quinalbarbitone to being residual from a previous night's hypnotic dose. In other words, it is the overall picture of drug taking which is important; not just the detection of a drug and reporting the case as "positive".

"Drugs and Driving" and "Drug Abuse"

Cases such as that described above have led to an investigation of the degree to which drug abuse is a factor in drugs and driving cases. The results are given in table 2. Information concerning the first two categories of drug abusers was obtained from the previously mentioned questionnaire for background information. In the third category examples of results taken as indicating drug abuse would be "positive for restricted drug(s)", "unusually
high barbiturate/methaqualone level", "strong detection of diethylpropion" etc.

The findings indicate that 35 per cent of 1978's drugs and driving cases in London and surrounding counties involved drug abuse. In all probability the actual percentage figure is even higher because blood alone was submitted in two thirds of our cases and there was therefore an incomplete picture of overall drug consumption.

The public at large probably think that most drugs and driving cases involve normal patients undergoing normal drug therapy who, having consumed alcohol socially and in moderation, are found to have their driving impaired by such a combination of drink and drug(s). Table 2 shows that this is only known to occur in 11 per cent of cases.

Correlation of "drugs and driving" and "drug abuse" trends

Statistics for the past eight years (figure 8) show some degree of correlation between increase in "drugs and driving" and "illicit drug possession". Many factors influence such statistics; for instance, the degree of police activity towards a particular type of crime. However, the increased incidence of "drugs and driving" is highlighted by the fact that it has occurred during a period in which the yearly totals for RTA cases involving alcohol have markedly decreased. This may be attributed to the more serious consequences of "drugs and driving".

The initial upsurge of "drugs and driving" cases from 1972 is probably partly due to the introduction of large RTA blood specimens coupled with an increase in expertise which led to drugs analysis being actively encouraged in RTA cases where low blood alcohol figures had been reported.
Discussion

This survey has shown that drug abuse contributes overwhelmingly to the drugs and driving cases encountered in Metropolitan and neighbouring County Constabularies. In other large cities, traffic accident studies have suggested that users (and abusers) of narcotics (1), sedative-hypnotics (2) and cannabis (3) (4) have driving records which do not differ significantly from those of age-matched members of the general population. However, the narcotics study has been criticized because of the nature of the control groups who, although not opiate addicts, may have been users of other drugs. Also the findings in relation to sedative hypnotics and cannabis are greatly disputed by studies showing a considerable over-involvement of barbiturate users among drivers in fatal accidents (5) and above-normal accident rates for users of cannabis (6). Laboratory studies of simulated driving show that all the drugs or classes of drugs examined in the present survey are capable of producing impairment (7). The fact that, in this survey, they appear mainly subject to abuse raises the question: should abusers, who are addicted to such drugs, be prevented from holding a licence to start with?

Finally, with regard to the preponderance of benzodiazepine tranquillizers (especially diazepam) in this survey, it is suggested that they be included in a class similar to the anti-histamines such that there is, by law, a printed warning of the drug's possible adverse effect upon driving etc. and the contraindication of alcohol on the outside of each prescription container. The number of court defences where it is alleged that there was no similar warning by the defendant's physician suggests that this is a problem which may be better solved by forensic pharmaceutical controls rather than by verbal recommendations left to be passed on from an overworked and sometimes forgetful physician to his ailing patient.

Reference


Figure 1

DETECTION OF ACIDIC & NEUTRAL DRUGS

10 ml URINE
OR
0.5 ml BLOOD

(ACIDIFY URINE)
EXTRACT WITH ETHER (x2)

ETHER

AQUEOUS

WASH WITH SATD. NaHCO₃

AQUEOUS

ETHER

TLC GC-AFID

BARBITURATES
METHAQUALONE
UREIDES
CHLORMETHIAZOLE
ANTICONVULSANTS ULN2001AZEPINE (OVERDOSES)
ANALGESIC/ANTIPYRETICS

Figure 2

DETECTION OF BASIC DRUGS

10 ml URINE

CONC. NH₄OH to
pH 10 - 12
EXTRACT WITH ETHER (x2)

ETHER

AQUEOUS

EVAPORATE TO
LOW BULK

TLC GC-AFID
ZONAL SCREEN ON OV-17
SCREEN 3 ISOTHERMAL TEMPS

DATA AVAILABLE FOR OVER 200 DRUGS.
NARCOTIC ANALGESICS
STIMULANTS
ANTIDEPRESSANTS
ANTIHISTAMINES
PHENOTHIAZINES
ANTIMALARIALS
LOCAL ANAESTHETICS
DETECTION OF BENZODIAZEPINES (AS BENZODIAZEPINES) AND MORPHINE

Figure 3

10 ml URINE

ADD 5ml CONC. HCL
HEAT AT 100°C FOR 30 MIN.
COOL. EXTRACT (x2) WITH ETHER.

ETHER

EVAPORATE TO DRYNESS. TAKE UP IN 20 μl MeOH

TLC FOR BENZODIAZEPINES

ORGANIC PHASE

EVAPORATE AND REDISSOLVE IN MeOH

AQUEOUS

TLC FOR MORPHINE

ADJUST TO pH 8.5 EXTRACT WITH ETHYL ACETATE/IPA (9:1)
FIGURE 4: SCHEME FOR SCREENING ACIDIC AND NEUTRAL DRUGS IN BLOOD - BATCH METHOD

1. Confirmation
2. 6 RTA bloods
3. Fresh aliquot extracted
4. GC-MS
5. Temp. Prog. GC Screen
6. Qual. \& Quant. results
7. Evaporate & redissolve in 20 μl acetone
8. Isothermal GC Screen
9. Concentrate calibration of 4 bars
10. Methylate C CH₂N₂
11. Ether extract & bicarb. wash

FIGURE 5: SCHEME FOR BLOOD BENZODIAZEPINE SCREENING - BATCH METHOD

1. Confirmation
2. 6 RTA bloods
3. Fresh aliquot extracted
4. GC-MS
5. EC-GC analysis
6. Qualitative + quantitative results
7. Evaporate hexane, dissolve in 100 μl hexane
8. Concentrate calibration for diazepam
9. Basify and extract aq. fraction & hexane
10. Extract & dryness
11. Back extract into N/4 HCl
12. 0.1 ml blood + prazepam
13. Also for cannabis + LSD
FIG 6 - FREQUENCY OF DETECTION OF DRUGS IN RTA CASES

DIAZEPAM

L  F  N  OTH

BARBITURATES

NO DRUGS

NARCOTIC ANALGESICS

MQ  CMZ  G  C

NON-BARBITURATE, NON-BENZODIAZEPINE SEDATIVES

CANNABINOIDS

STIMULANT

ANTIHISTAMINE

ANTICONVULSANT

ANTIDEPRESSANT

% DETECTION

L = LORAZEPAM
F = FLURAZEPAM
N = NITRAZEPAM
MQ = METHAQUALONE
CMZ = CHLORMETHIAZOLE
G = GLUTETHIMIDE
C = CHLORAL HYDRATE
FIG 7: A COMPARISON OF DRUG DETECTION IN "BLOOD ONLY" RTA CASES WITH THAT FOR ALL RTA DRUG CASES

- Diazepam
- Lor
- Flur
- Nitr
- Oth

- Barbiturates
- No drugs
- Narcotic analgesics

- MQ
- CMZ
- Other sedatives
- Cannabinoid

- Detection in all RTA drugs cases
- And in "blood only" RTA drugs cases

Percentage detection

0 10 20 30 40
Figure 8

YEARLY TOTALS FOR DRUGS & DRIVING CASES
ALCOHOL/DRUGS-DRIVING CASES, AND DRUGS
POSSESSION CASES

RTA DRUGS CASES

TOTAL RTA CASES ($\times 10^3$)

TOTAL DRUGS POSSESSION CASES ($\times 10^3$)

1971 72 73 74 75 76 77 78

30

20

10

300

200

100

100

5

6

7
TABLE 1

R. v. C. R. BROWNE

DEFENDANT ADMITTED:  
ASPIRIN  
GOLD INJECTION  
INDOMETHACIN  
METHADONE  

FOUND ON ANALYSIS:  

URINE  
AMYLOBARBITONE  
QUINALBARBITONE  
(PLUS HYDROXY METABOLITES)  
AMPHETAMINE  
METHYL AMPHETAMINE  
EPHEDRINE  
PROCAINE  
METHADONE  
(PLUS METABOLITE)  
MORPHINE  

BLOOD  
ZERO ALCOHOL  
AMYLOBARBITONE  
(1.1 µg/ML)  
QUINALBARBITONE (NOT MEASURABLE: Me-TBTH INTERFERED)  
MORPHINE RIA POSITIVE (28 NG/ML)  
(SAMPLE EXHAUSTED)

TABLE 2: RTA DRUGS CASES 1978 (January to September)

CASES INVOLVING "REGISTERED" OR "SELF-CONFESSED" DRUG ADDICTS  9.5%

CASES INVOLVING "KNOWN" DRUG ABUSERS  15.5%

CASES IN WHICH THE ANALYTICAL RESULTS INDICATE DRUG ABUSE  11 %

CASES WITH NORMAL THERAPEUTIC BLOOD CONCENTRATION OF DRUG PLUS BELOW-THE-LIMIT ALCOHOL  11 %