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CASE REPORT

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Fatal injections of heroin. Interpretation of toxicological findings in multiple specimens

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Abstract We report two fatalities due to injection of heroin. The first case was witnessed but during the early phase of the police investigation the question was raised whether the injection was self-administered. Multiple samples were collected from different sites and analysed in order to establish drug distribution and to determine the site of injection. Fresh injection marks were found in both antecubital fossae but histological examination failed to settle which one was the last. However, toxicological analysis of the tissues at the injection sites indicated that the injection in the right arm was the last one. This was consistent with the suspicion that the victim was given the injection by another person although probably in agreement with the deceased. In the second case, a similar toxicological procedure was used. This fatality was not witnessed, however ample evidence indicated that it was an isolated event in a former intravenous heroin addict and there was only one fresh injection mark. Even in this case, the concentration of morphine was much higher in the tissue sample from the injection mark than in any of the blood samples.

Key words Heroin · Morphine · Fatal · Blood-concentration · Disposition · Injection

Introduction

Although fatal outcome due to injection of heroin is widely recognized and documented [1–3], the postmortal blood concentrations of morphine in such deaths show large interindividual variation [3–8]. It is therefore difficult to prove that death is caused by an acute overdose taking only the blood concentration into account. The extensive variation in concentrations found in deaths attributed to heroin intoxication may be explained by differences in samples used (e.g. central vs peripheral blood) [7, 9–11], differences in tolerance [5, 12, 13] and the time interval between injection and death [5, 6, 11, 14–16]. The simple fact that an intentional overdose, when injected, may exceed the fatal dose severalfold will also contribute to the wide interindividual variation in postmortem blood concentrations. Furthermore, the victims may be influenced by other drugs and a negative correlation between morphine concentration and alcohol concentration has been shown in both unconscious and deceased drug addicts, indicating a potentiating effect of alcohol on heroin toxicity [12, 17].

Whereas multiple specimens have been collected from autopsy cases to investigate the possible postmortem redistribution and degradation of various compounds, we have been unable to find any reports where this regimen has been employed to determine the route of administration. The objective of this study was to find out whether or not toxicological analysis of multiple specimens could be of any support for determining the injection site.

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Case reports

Case 1

A 28-year-old man was brought to the emergency unit at a hospital but showed no vital reactions on admittance. The deceased had purchased heroin and visited a friend to inject it. Since he was inexperienced in injecting, a friend took him to the kitchen in order to help inject the drug. Although three witnesses were present in the adjacent living room, nobody was able to say who gave the injection. After the injection, he joined his friends in the living room and sat down in an armchair but half a minute later, his lips turned blue, he started to blink rapidly, then closed his eyes and eventually bent forward and became lifeless. A faint pulse was recorded at first, but it soon disappeared and no breathing was noted. After 5–10 min of dispute his friends finally decided to call for an ambulance which arrived 3–4 min later. There were some disagreement about the past history of the deceased. One of his friends

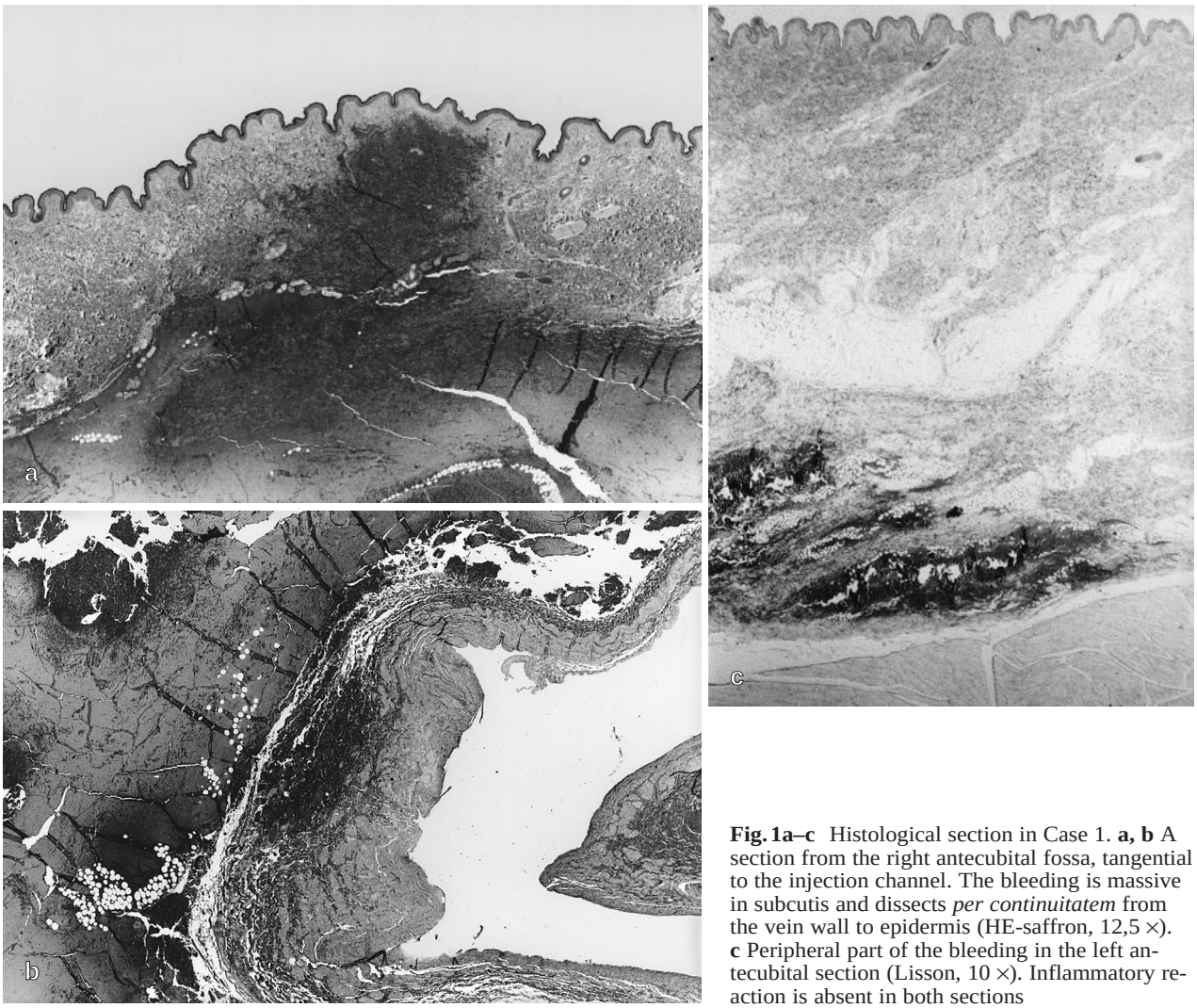

Fig. 1a–c Histological section in Case 1. **a, b** A section from the right antecubital fossa, tangential to the injection channel. The bleeding is massive in subcutis and dissects *per continuitatem* from the vein wall to epidermis (HE-saffron, 12,5 ×). **c** Peripheral part of the bleeding in the left antecubital section (Lisson, 10 ×). Inflammatory reaction is absent in both sections

claimed that the victim had abused pharmaceutical drugs for some years and occasionally injected amphetamine (but never heroin), whereas others stated he was inexperienced with drugs apart from alcohol and benzodiazepines. His mother claimed he was intentionally killed by his friends since he had many enemies, but the witnesses considered the fatality an accident as did the police.

The deceased was 183 cm in height and weighed 89 kg and was strongly built. Fresh injection marks were discovered at the antecubital fossae of both arms. On preparation of the subcutaneous tissue, large dark red haemorrhages were found measuring $2.5 \times 2 \times 0.8$ cm and $5 \times 5 \times 3$ cm on the right and left side, respectively. There were no older injection marks or scarring at the antecubital areas or forearms. The fresh injection marks constituted the only recent injuries found at the external examination of the body. Apart from pulmonary edema, the autopsy disclosed no pathology findings. There was no congestion of the inner organs suggesting that the death was rapid and not preceeded by cardiac failure. The liver showed no macroscopic fatty changes. The heart had a normal appearance with homogenous, normal brownish red myocardium and smooth valves. Specimens, approx. $4 \times 3 \times 2$ cm, were collected from the tissues surrounding the injection marks and hemisected into two identical sections, one for toxicology and one for histology. The histological specimens were fixed in 10% buffered formalin, subjected to conventional dehydration and paraffin embedded. Upon microscopical examination, extensive bleedings were seen in the dermis and subcutis. The bleedings had a similar appearance on both sides, suggesting fresh injuries without accumulation of granulocytes, macrophages or other inflammatory cells (Fig. 1). No clear fibrin deposits were found as assessed by PTAH staining. Immunohistochemistry against human fibrinogen (DAKO, Stockholm, Sweden) showed no positive material in any of the specimens and Fe staining was negative. Thus, it was not possible to histologically determine which injection mark was the last one.

Toxicological findings

Blood samples from both arms and legs were collected from the veins at the site where they enter the torso. Care was taken to avoid contamination of the peripheral specimens by central blood. Heart blood was collected from the right atrium. A $4 \times 3 \times 1$ cm tissue sample was collected from the injection sites at both antecubital fossae. Potassium fluoride was added to the blood samples to a concentration of 1%.

Ethanol was detected at a concentration of 0.18 g/dL in femoral blood and 0.25 g/dL in urine. Routine blood screening for pharmaceutical drugs was negative. Amphetamine, cannabis, cocaine, cocaine metablites and anabolic steroids were not detected in urine. Morphine and codeine were found in all samples collected (Table 1). Both of these substances were analysed using a modification of the GC-MS method described by Schuberth and Schuberth [18]. The heroin metabolite 6-acetylmorphine was detected in all specimens except in the tissue sample from the left antecubital fossa. In

Table 1 Blood and tissue concentrations of free morphine, 6-acetyl-morphine and codeine in Cases 1 and 2

Case 1

Sampling site	Morphine	6-Acetyl-morphine	Codeine
Right brachial vein	0.13	trace	0.01
Left brachial vein	0.07	trace	0.01
Right femoral vein	0.06	trace	0.01
Left femoral vein	0.07	trace	0.01
Right heart blood	0.11	trace	0.11
Tissue right arm	1.90	0.03	0.20
Tissue left arm	0.07	n.d.	0.01

Case 2

Sampling site	Morphine	6-Acetyl-morphine	Codeine
Right brachial vein	0.24	0.02	0.01
Left brachial vein	0.20	0.01	0.01
Femoral blood	0.13	0.009	0.007
Right heart blood	0.27	trace	0.01
Left heart blood	0.30	trace	0.01
Tissue right arm	0.87	n.d.	0.05
Tissue left arm	0.15	n.d.	0.01

All values are μg/g wet tissue. Trace = positive detection, but below the cut off level of 0.005 μg/g; n.d. = not detected

five of the samples, 6-acetylmorphine was detected in a concentration close to, but below the detection limit of 0.005 μg/g.

Case 2

A 23-year-old woman was found dead in her apartment, sitting on a chair in the kitchen and leaning against the breakfast table. A syringe was still in her right antecubital fossa and fell out as her boy friend tried to awaken her. The previous day, she had purchased and obviously injected heroin. She was a former heroin addict but had not taken drugs for 18 months, except for two isolated occasions when she had injected heroin. Over the last month she had occasionally abused pentazocin and propoxyphene. Presumably, she had hidden the heroin from her boy friend and injected it after he had fallen asleep, after midnight. He found her at 0800 hours and along with the syringe, a package of sugar and a teaspoon were found on the breakfast table. The boy friend called an ambulance but the paramedics concluded she had been dead for hours.

The deceased was 165 cm in height and weighed 55 kg. There were no signs of malnutrition. A tiny injection mark was found in the right antecubital fossa. A fresh, small bleeding was present at the outside of the median cubital vein, as well as in the thin, subcutaneous fat (Fig. 2). No other injection marks were seen and dissection of the subcutaneous tissues of the extremities revealed no additional fresh bleedings. A moderate lung edema was noted and the inner organs were markedly congested. No foreign material was seen in the bowel. Chronic inflammation, compatible with persistent hepatitis C, was observed in the liver. There were no signs of ischemic or hypoxic damage to the heart, liver, kidneys or brain. Tissue samples from both antecubital fossae were collected for toxicological analysis, including the injection mark on the right side. The bleeding in the subcutaneous tissue and around the injected vessel was too minute to allow for both histological and toxicological sampling.

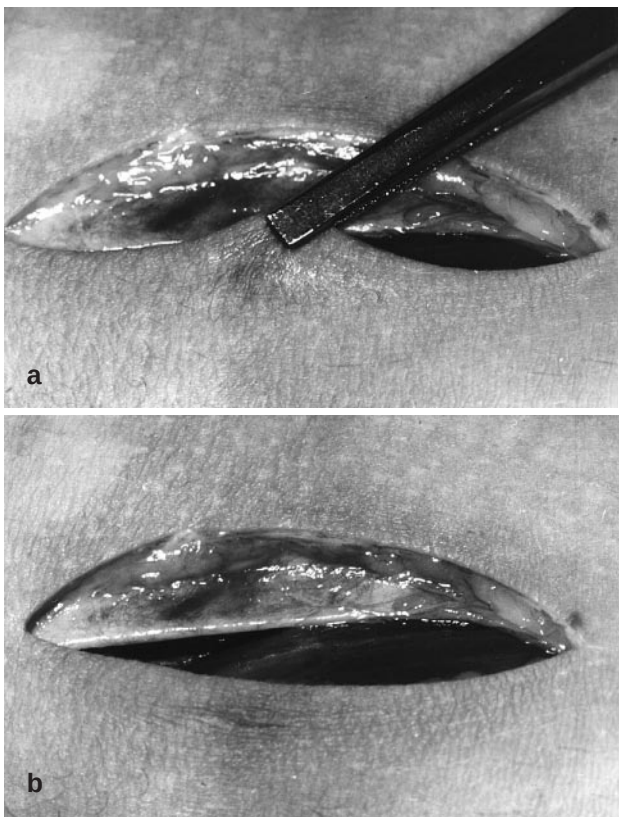

Fig. 2 a, b A close-up view of the injection mark in Case 2. Note the discrete and diffuse bleeding in the subcutis (which in this case is very thin) and the more distinct bleeding in the vessel wall. The entrance in the skin is concealed within the dark area below the cut edge (original magnification 2 ×)

Toxicological findings

Samples for toxicological analyses were collected and handled as described for case 1. Ethanol was not detected in any of the samples. Routine screening for pharmaceutical drugs was also negative. Morphine and codeine were found in all blood and tissue samples (Table 1), whereas 6-acetyl-morphine was only found in the blood samples.

Discussion

Due to gradual leakage of fluid from the injection site, the concentration of the drug injected would be expected to stay higher in the vein blood in the extremity into which the fatal injection was given, compared with other peripheral blood specimens, provided that the death ensued quickly. However, in both cases the blood distribution of morphine and codeine did not leave any clue regarding the site of the fatal injections.

The results of the tissue analyses turned out to be more helpful. The impressive difference in morphine concentrations between the injection sites indicated that the fatal injection was given into the right arm of both victims. In case 1, the fact that the deceased was right-handed and showed equally large easily accessible superficial veins on both arms, suggested that the injection in the right arm

was assisted. The absence of old injection marks and scars along with negative Fe-stain was further consistent with the statements of his friends that he was not an intravenous drug abuser. In case 2, only one injection mark was found. Toxicology showed a similar pattern to that of case 1 with the highest morphine and codeine concentrations detected in the tissue sample from the right arm. The scarce subcutaneous fatty tissue did not allow for histological examination in this case. In both cases, the blood morphine levels were of the same magnitude as the concentrations reported in published fatal cases, although at the lower end of the distribution range for case 1 [3, 6, 10, 19, 20]. With a blood alcohol concentration of 0.18 g/dL, however, this case fits the curve of combination intoxications presented by Ruttenber et al. [17].

Different measures have been employed to establish the time between administration and death. Thus, morphological changes in the lungs [21] and at the injection sites [22] in fatal and non-fatal overdose cases have been investigated. Furthermore, the impact of the morphine-6-glucuronide and morphine-3-glucuronide to free morphine ratios has been explored by some authors [6, 11, 15, 23]. Although some of these reports also include analytical data from different sampling sites, this information has not been used for the determination of the injection site. The toxicology results of both cases reported here seem to indicate that blood concentrations of morphine equilibrate rapidly within the circulatory system after a fatal injection, displaying similar levels irrespective of the collection site. A similar uniformity of the blood levels was also found for codeine. According to these results, it seems that determination of the injection site cannot be deduced from a regimen of collecting different blood samples, unless a torniquet is still in place (in which case this question should easily be solved anyway).

The distribution of codeine, with the highest concentration in the tissue specimen from the right antecubutal fossa, is compatible with the presumption that codeine, as an additive, was injected along with heroin in both cases. However, the elevated codeine concentration in the heart blood in case 1 is somewhat puzzling. It might be explained by a postmortem redistribution [7, 9–11, 24], although this phenomenon should also be expected for morphine and 6-acetyl-morphine. A separate intake at an earlier date or a chronic abuse constitute other possible explanations. Bailey and Shaw [25] reported detection of codeine in the myocardium in four cases, where the drug was not found in heart blood.

In case 2, no 6-acetyl-morphine could be detected in the tissue sample from the right arm. The reason for this remains uncertain. There is a difference between the two cases in that the amount of bleeding at the injection site was quite different. Thus, it is possible that the degradation of 6-acetyl-morphine [5, 26] might be more rapid in the skin and fatty tissue than in the blood (including haemorrhages).

This raises the question as to the properties of control samples used. Perhaps a control sample with a similar sized, time-matched bleeding should be looked for rather than an anatomically corresponding sampling site. In both of these cases, care was taken to include the same proportions of skin, fatty tissue and bleeding. However, in case 2, no other fresh subcutaneous bleedings were found to be used for comparison.

Collection of tissue specimens from injection marks and of control tissue samples has been recommended to establish the injection site [27], but we have failed to find any reports where analysis of such material has been used for this purpose except for insulin poisonings, where the objective has been to restricted to qualitative detection of exogenous insulin [28,29]. Although Goldenberger et al. [7] presented tissue concentrations of morphine, 6-acetylmorphine and heroin in tissue from one injection mark, they did not comment on this further and the appearance of the injection site, as to extent and age of the bleeding, was not reported.

Drug-addicts, known to have injected drugs more than once in the last day, may provide pit-falls, since the elimination of morphine and related compounds from injections prior to the fatal one may be slow in the subcutaneous tissue. Furthermore, in some cases injections may be very accurate leading to almost no bleeding at all and thus leaving no substantial direct contribution to the local tissue drug concentration. However, as long as the needle is not still in place, a leakage of blood with a high concentration of the drug will probably always take place for a short period through the perforation(s) of the vessel into the extravascular space.

In conclusion, the fairly simple procedure of a combination of toxicological and microscopical examination of injection marks may provide additional information of assistance for the interpretation of intravenous drug intoxications.

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