Zopiclone (Zimovane™) is a cyclopentylpyrrolidine hypnotic used for the short-term treatment of insomnia. It also possesses anxiolytic, sedative, anti-convulsant and muscle-relaxant properties. Despite sharing the pharmacological actions of benzodiazepines, zopiclone acts on different receptors. Zopiclone has an oral bioavailability of approximately 80%, and is absorbed rapidly, producing peak blood concentrations within 1.5 to 2 hours. It is extensively metabolised in the liver to zopiclone-N-oxide (active) and N-desmethylzopiclone (inactive). The elimination half-life is approximately 5 hours and no accumulation has been seen after repeated dosing.

Recently there has been an increase in the number of reports of zopiclone abuse and misuse. Drug abusers are now thought to be using zopiclone as its effects are comparable to those of benzodiazepines, yet routine drug screening programmes are often unable to detect its presence. Zopiclone’s rapid onset of action, coupled with its short half-life also makes it a potential candidate for drug-facilitated crime. For these reasons it is no surprise that the number of zopiclone cases presenting to forensic toxicology laboratories is on the increase.

The detection and subsequent quantification of zopiclone is plagued with problems. It is unstable in nuclease solvents such as methanol or ethanol. It has a high molecular weight (388.6) and therapeutic concentrations are low (30-60ng/mL peak). While fluoride preservatives may assist in preserving zopiclone concentrations in post-mortem samples, prior exposure to unusual conditions, such as putrefaction may have already resulted in significant degradation. Unfortunately due to difficulties associated with zopiclone stability and analysis, routine screening procedures often overlook its presence.

Whilst the instability of zopiclone can hinder its detection, this phenomenon has the potential to be exploited. Under alkaline conditions zopiclone rapidly hydrolyses to 2-amino-5-chloropyridine (ACP). This lower molecular weight (128.6) compound is far easier to detect through routine screening. We describe HPLC-MS-MS methods for the detection of zopiclone and its main alkaline hydrolysis product, ACP in post-mortem blood and evaluate the potential value of ACP quantification as a means of establishing zopiclone overdose.

**Materials**

Zopiclone was obtained from Aventis (Nanterre, France) and ACP was obtained from Fluka (Seelze, Germany). HPLC grade methanol, acetonitrile and methyl-tert-butyl-ether (MTBE) were purchased from Rathburn Chemicals Limited (Walkerburn, Scotland). Analar grade formic acid and sodium hydroxide (40%) solution were obtained from BDH (Poole, Dorset, England). Trizma Base was obtained from Sigma (St Louis, USA). De-ionised water was prepared on site (ELG Aqua). The method was run in positive ionisation mode and set to measure precursor and product ion transitions of m/z 389.1/245.1 and 391.2/247.0 for zopiclone, which eluted after 6.8 minutes. Concentrations of both analytes could be detected at 5ng/mL.

**RESULTS**

The data show that ACP was detectable in all cases of zopiclone overdose and in all except one case of therapeutic use. The authors believe that ACP can be used reliably for routine zopiclone screening and that its use for this purpose will lead to an increase in the detection of zopiclone positive cases. The data obtained also suggest the potential quantitative use of ACP in the estimation of zopiclone concentrations. However, further research into this area is required.

**REFERENCES**