Forensic Science International 262 (2016) 128–137

Contents lists available at ScienceDirect

Forensic Science International

journal homepage: www.elsevier.com/locate/forsciint

Post-mortem levels and tissue distribution of codeine, codeine-6-glucuronide, norcodeine, morphine and morphine glucuronides in a series of codeine-related deaths

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ARTICLE INFO

Article history:
Received 2 November 2015
Received in revised form 19 February 2016
Accepted 24 February 2016
Available online 4 March 2016

Keywords:
Forensic toxicology
Codeine
Toxicity
Post-mortem
Tissue distribution

ABSTRACT

This article presents levels and tissue distribution of codeine, codeine-6-glucuronide (C6G), norcodeine, morphine and the morphine metabolites morphine-3-glucuronide (M3G) and morphine-6-glucuronide (M6G) in post-mortem blood (peripheral and heart blood), vitreous fluid, muscle, fat and brain tissue in a series of 23 codeine-related fatalities. CYP2D6 genotype is also determined and taken into account. Quantification of codeine, C6G, norcodeine, morphine, M3G and M6G was performed with a validated solid phase extraction LC–MS method. The series comprise 19 deaths (83%) attributed to mixed drug intoxication, 4 deaths (17%) attributed to other causes of death, and no cases of unambiguous monointoxication with codeine. The typical peripheral blood concentration pattern in individual cases was C6G ≫ codeine ≫ norcodeine > morphine, and M3G > M6G > morphine. In matrices other than blood, the concentration pattern was similar, although in a less systematic fashion. Measured concentrations were generally lower in matrices other than blood, especially in brain and fat, and in particular for the glucuronides (C6G, M3G and M6G) and, to some extent, morphine. In brain tissue, the presumed active moieties morphine and M6G were both below the LLOQ (0.0080 mg/L and 0.058 mg/L, respectively) in a majority of cases. In general, there was a large variability in both measured concentrations and calculated blood/tissue concentration ratios. There was also a large variability in calculated ratios of morphine to codeine, C6G to codeine and norcodeine to codeine in all matrices, and CYP2D6 genotype was not a reliable predictor of these ratios. The different blood/tissue concentration ratios showed no systematic relationship with the post-mortem interval. No coherent degradation or formation patterns for codeine, morphine, M3G and M6G were observed upon reanalysis in peripheral blood after storage.

1. Introduction

The opiate codeine is widely used in many countries as an analgesic and cough suppressant, either alone or in combination with other drugs, and is a frequent finding in post-mortem forensic toxicology.

Codeine is mainly metabolized in the liver, although some intestinal and CNS metabolism probably occurs. The principal metabolic pathways are outlined in Fig. 1. A major part (50–70%) of a codeine dose is glucuronidated to codeine-6-glucuronide (C6G), while 10–15% is N-demethylated to norcodeine via the cytochrome P450 isoenzyme 3A4 (CYP3A4) [1]. Norcodeine is in turn glucuronidated to norcodeine-6-glucuronide (N6G), and a minor part is O-demethylated to normorphine [2,3]. Of an ingested codeine dose, 0–15% is O-demethylated to morphine by the polymorphic cytochrome P450 isoenzyme 2D6 (CYP2D6), and further glucuronidated to the inactive metabolite morphine-3-glucuronide (M3G; approximately 60% of morphine formed) and the active metabolite morphine-6-glucuronide (M6G; 5–10%

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http://dx.doi.org/10.1016/j.forsciint.2016.02.051
0379-0738/

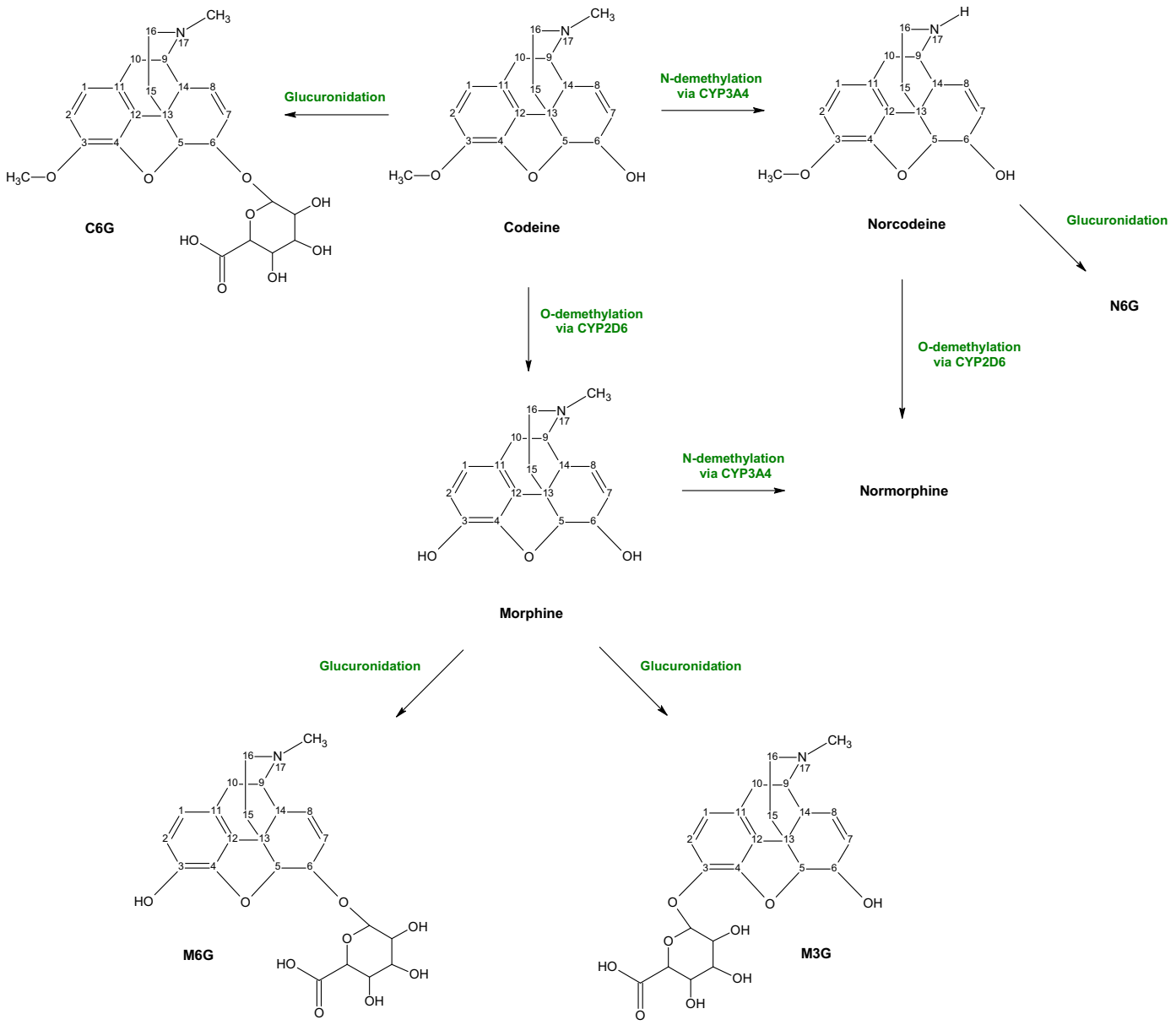

Fig. 1. Principal pathways for codeine metabolism in man. C6G: codeine-6-glucuronide; N6G: norcodeine-6-glucuronide; M3G: morphine-3-glucuronide; M6G: morphine-6-glucuronide; CYP2D6: cytochrome P450 isoenzyme 2D6; CYP3A4: cytochrome P450 isoenzyme 3A4.
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of morphine formed) [1]. A minor part of morphine is N-demethylated to normorphine [2,3]. CYP2D6 activity may be significantly influenced by genetic polymorphisms and environmental factors such as inhibitory interactions from other drugs, which results in a large and unpredictable intra- and interindividual variability in the amount of morphine produced after ingestion of codeine [4–10].

Compared to morphine and M6G, codeine and its main metabolites C6G and norcodeine have weak affinity to opioid μ-receptors [11–13]. Normorphine has about one fourth of the μ opioid receptor affinity of morphine, and is produced in small amounts [3,11]. Accordingly, the analgesic effects of codeine appear to be largely dependent on metabolic conversion to morphine by CYP2D6 [6,14–16]. Whether this also applies to the toxicity of codeine, however, remains a matter of controversy. Although unsubstantiated by receptor affinity studies [11–13], some investigators have suggested codeine, C6G and norcodeine as putative mediators of codeine toxicity [17–20].

Previous studies of codeine-related deaths have reported a limited array of codeine metabolites in biological specimens, particularly in matrices other than blood and urine. Published data derive from various case reports and series [5,21–37], of which five [23,30,31,35,37] are large autopsy series. Published post-mortem concentrations of other codeine metabolites than morphine are limited to 2 cases of fatal and severe codeine intoxication [32], and a series of 31 unspecified autopsy blood samples [38]. Furthermore, published data regarding post-mortem redistribution of codeine and its metabolites are limited and inconsistent [21,39–47]. To address these limitations in the literature, further investigations of the concentrations and tissue distribution of codeine and its metabolites in codeine-implicated deaths are warranted.

We hypothesized that CYP2D6 metabolic capacity may be reflected in the detected amounts of morphine, and possibly other metabolites, in codeine-related deaths. In a previous study we