Methadone Pharmacogenetics

CYP2B6 Polymorphisms Determine Plasma Concentrations, Clearance, and Metabolism

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Abstract

Interindividual variability in methadone disposition remains unexplained, and methadone accidental overdose in pain therapy is a significant public health problem. Cytochrome P4502B6 (CYP2B6) is the principle determinant of clinical methadone elimination. The CYP2B6 gene is highly polymorphic, with several variant alleles. CYP2B6.6, the protein encoded by the CYP2B6*6 polymorphism, deficiently catalyzes methadone metabolism in vitro. This investigation determined the influence of CYP2B6*6, and other allelic variants encountered, on methadone concentrations, clearance, and metabolism.

Methadone

is a long-duration opioid

for acute, chronic, perioperative, neuropathic, and cancer pain

Methadone is typically a racemic mixture

R-methadone primarily confers the μ-opioid receptor activity

both enantiomers act at N-methyl-d-aspartate receptors

! In 2009, methadone accounted for only 2% of prescriptions, but 30% of prescription painkiller deaths !

Understanding methadone disposition is important for reducing adverse

Methadone is cleared by:

Hepatic cytochrome P450 (CYP)-catalyzed N-demethylation

to inactive 2-ethyl- 1,5-dimethyl-3,3-diphenylpyrrolidine (EDDP)

some urinary excretion of unchanged drug

The CYP2B6 gene in relation to Methadone.

Highly polymorphic (38 variant alleles identified)

CYP2B6*6 is the most common and clinically significant variant allele

The hypothesis:

CYP2B6*6 heterozygotes or homozygotes would have reduced metabolism and clearance.

Is to evaluate other less common genotypic variants, when encountered.

Materials and Methods

Inclusion criteria were:

18- to 50-yr-old normal healthy volunteers

good general health without remarkable medical conditions

and within 30% of ideal body weight (body mass index < 33 kg/m2)

Exclusion criteria were:

a history of hepatic or renal disease

use of prescription or nonprescription medications, herbals or foods known to be metabolized by or affect the activity of CYP2B6

Potential subjects provided a venous blood sample, and genomic DNA was isolated from peripheral blood leukocytes by using the Gentra Puregene Blood Kit

Genotyping was performed by the Genome Technology Access Center at Washington University in St. Louis by using the Fluidigm BioMark System

Genotyping results were then used to invite subject participation and create target cohorts of 20 subjects each with CYP2B6*1/*1, CYP2B6*1/*6, and CYP2B6*6/*6 genotypes

Subjects were instructed to refrain from:

alcohol for 48h before and during the study day

caffeine-containing beverages on the study day

oranges, grapefruit, or apples or their juices for 5 days before and throughout the 96-h study period

food/liquids after midnight the day before methadone administration

nonstudy medications (including over the counter and/or herbal) for 3 days before the study day, without previous approval

Table 1. Subject Demographics

CYP2B6 Genotype	Sex (M:F)	Age (yr)	Weight (kg)	Caucasian	African American	Asian	Other/Unknown
**1/*1*	9:12	28 ± 7	72 ± 14	13	1	7	
**1/*6*	14:6	28 ± 8	78 ± 11	14	3	2	1
**6/*6*	7:10	32 ± 9	71 ± 13	9	6	1	1
**1/*4*	1:0	22	68	1			
**4/*6*	1:2	29 ± 2	68 ± 15	3			
**5/*5*	2:0	28 ± 1	84 ± 0	2			
Total	34:30	29 ± 8	74 ± 13	42	10	10	2

Data and Statistical Analysis

Pharmacokinetic data were analyzed by using noncompartmental methods

Results are reported as the arithmetic mean ± SD

The primary outcome measure was methadone metabolism, measured as plasma EDDP/methadone area under the concentration– time curve (AUC0–96) ratio and EDDP formation clearance.

Secondary outcomes included methadone peak plasma concentration, exposure (plasma AUC∞)

Results

Allele frequencies are consistent with the previous reports

Plasma methadone and EDDP enantiomer concentrations are shown for oral (fig. 1) and IV (fig. 2) methadone, for the three major genotype groups (CYP2B6*1/*1, CYP2B6*1/*6, and CYP2B6*6/*6) and for *4 carriers (CYP2B6*1/*4 and CYP2B6*4/*6, shown together as CYP2B6*4/X)

Genotype influence was greater for oral than IV dosing and for S- than R-methadone

For oral methadone, average plasma exposure

FOR S-METHADONE

in CYP2B6*1/*1 was 620 ± 230 ng/ml-h

in CYP2B6*1/*6 was 734 ± 245 ng/ml-h

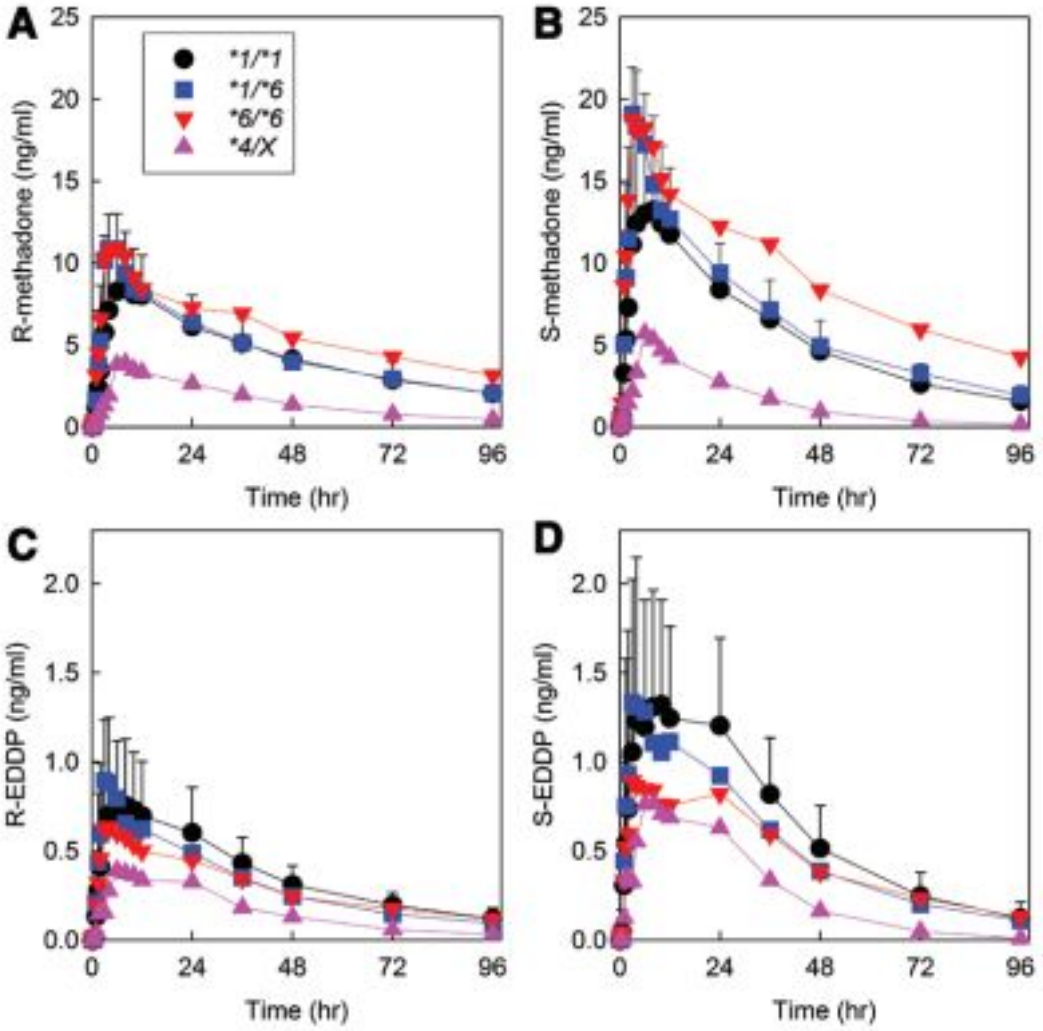
in CYP2B6*6/*6 was 1,242 ± 801 ng/ml-h

FOR R-METHADONE

in CYP2B6*1/*1 was 578 ± 205 ng/ml-h

in CYP2B6*1/*6 was 615 ± 172 ng/ml-h

in CYP2B6*6/*6 was 898 ± 507 ng/ml-h


A
B
C
D
*1/*1
*1/*6
*6/*6
*4/X
R-methadone (ng/ml)
S-methadone (ng/ml)
R-EDDP (ng/ml)
S-EDDP (ng/ml)
Time (hr)
0
24
48
72
96
0.0
0.5
1.0
1.5
2.0
5
10
15
20
25

For IV methadone

FOR S-METHADONE

in CYP2B6*1/*1 was 447 ± 85

in CYP2B6*1/*6 was 513 ± 171

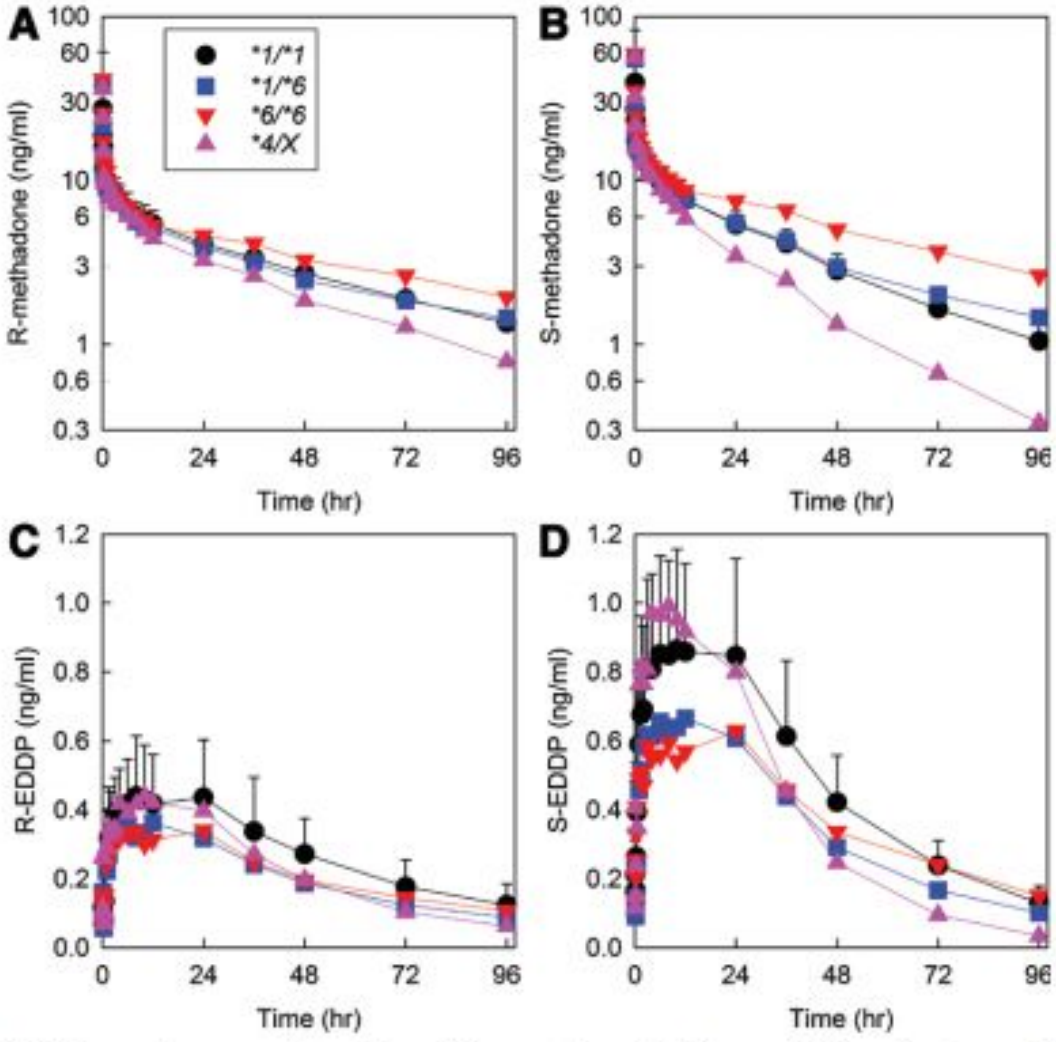
in CYP2B6*6/*6 was 801 ± 464

FOR R-METHADONE

in CYP2B6*1/*1 was 430 ± 131

in CYP2B6*1/*6 was 429 ± 135

in CYP2B6*6/*6 was 570 ± 281


A
B
C
D
*1/*1
*1/*6
*6/*6
*4/X
R-methadone (ng/ml)
S-methadone (ng/ml)
R-EDDP (ng/ml)
S-EDDP (ng/ml)
Time (hr)
100
60
30
10
6
3
1
0.6
0.3
1.2
1.0
0.8
0.4
0.2
0.0
0
24
48
72
96

Hepatic clearance (ml kg−1 min−1) was significantly less in CYP2B6*6/*6 compared with that of CYP2B6*1/*1 subjects for S-methadone (0.8 ±0.4 and 1.3±0.3) but not R-methadone (1.0 ±0.3 and 1.3±0.3)

Methadone N-demethylation was significantly less in CYP2B6*6 carriers, particularly homozygotes, and apparently greater in CYP2B6*4 carriers

S-methadone systemic clearance (ml kg−1 min−1) in CYP2B6*1/*6 and CYP2B6*6/*6 subjects (1.2 ± 0.4 and 0.96 ±0.33, respectively) was significantly less than in CYP2B6*1 homozygotes (1.5 ±0.3)

R-methadone clearances in CYP2B6*6 carriers were not significantly different from CYP2B6*1/*1 subjects

In contrast, R- and S-methadone systemic clearances (2.4 ± 0.7 and 2.7 ± 0.9) and apparent oral clearances (7.4 ± 3.8 and 8.6 ± 3.2) were numerically

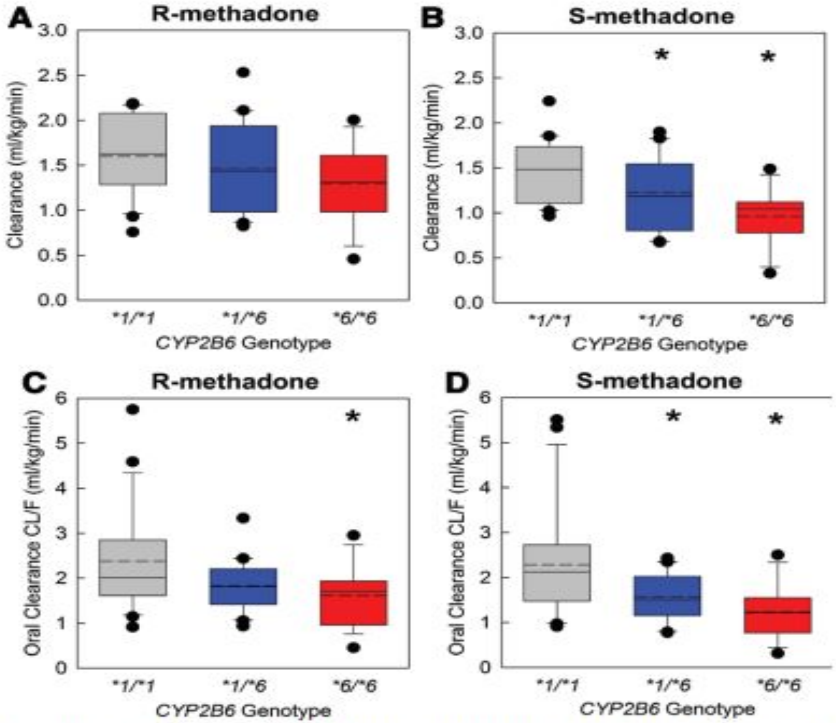


Fig. 4. Influence of *CYP2B6*6* genotype on methadone clearance. Shown are the systemic clearances for IV (*A*) R-methadone and (*B*) S-methadone and the apparent oral clearances for oral (*C*) R-methadone and (*D*) S-methadone, as *box plots* (*solid line* within the box represents the median, *dashed line* within the box represents the mean, box boundaries are the 25th and 75th percentiles, *error bars* are the 10th and 90th percentiles, and individual points are outliers). *Significantly different from wild-type (*CYP2B6*1/*1*), $P < 0.05$. CL/F = apparent oral clearance.

Conclusion

Methadone disposition was stereoselective, with greater initial exposure to S-methadone

Plasma methadone concentration change was diminished in CYP2B6*6 allele carriers and accentuated in CYP2B6*4 carriers.

CYP2B6*6 allele carriers, particularly homozygotes, had higher methadone concentrations and slower elimination, whereas CYP2B6*4 carriers had lower concentrations and faster elimination.

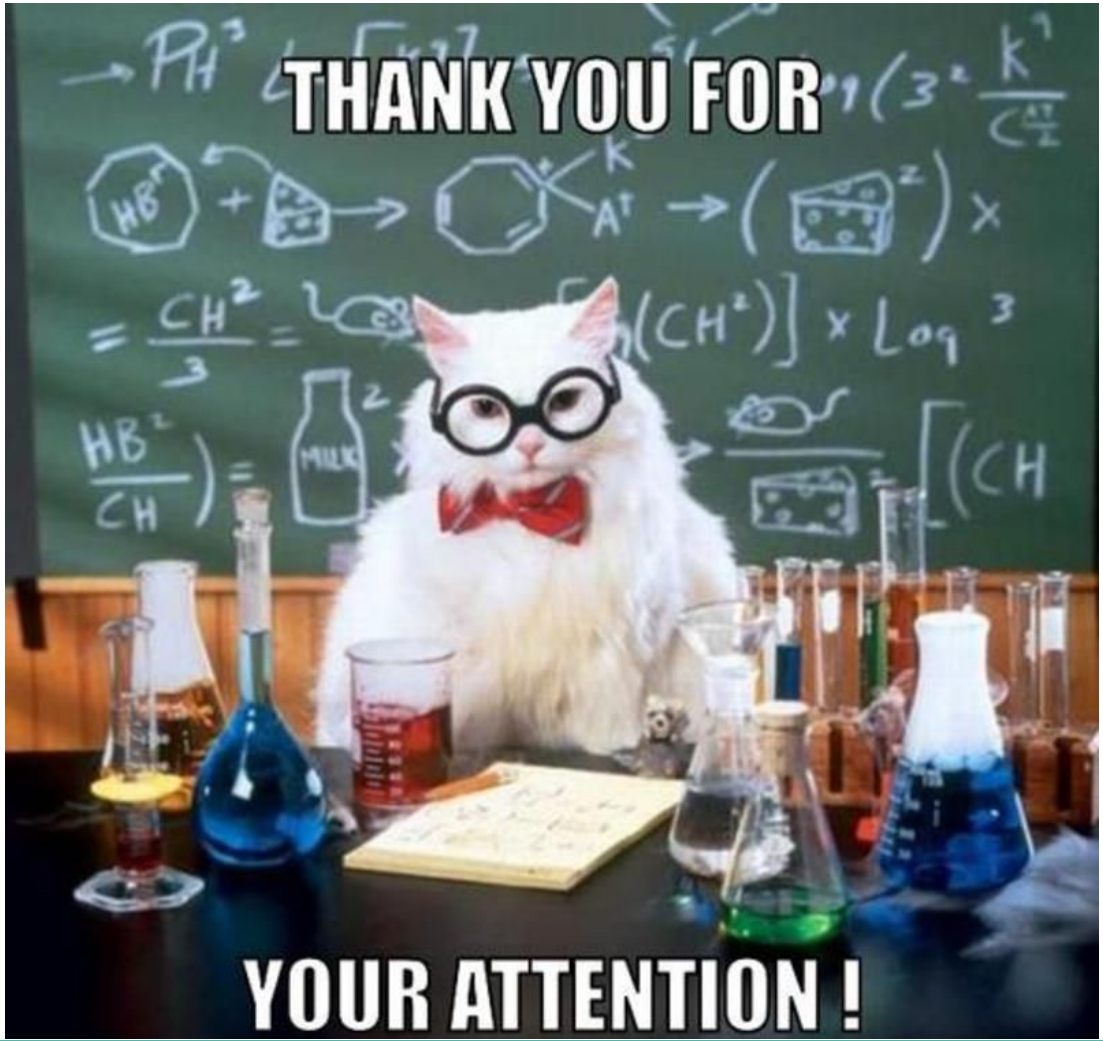
Discussion

Allelic influences on methadone concentrations were caused by differences in clearance

CYP2B6 genetic influence on methadone metabolism and clearance further highlights and reinforces CYP2B6 as the predominant CYP responsible for clinical methadone elimination.

It is now established, after recognizing CYP2B6 as a major catalyst of methadone metabolism in vitro, 28,44–47 and from numerous clinical drug interaction studies, that CYP2B6, not CYP3A4, is the principle determinant of methadone elimination.

These results provide a mechanistic understanding for interindividual variability in methadone elimination and may have clinical implications for genetically based improvements in methadone dosing, effectiveness, and


THANK YOU FOR
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