

For reprint orders, please contact: reprints@futuremedicine.com



Genetics of pain perception, *COMT* and postoperative pain management in children

Background: Effective perioperative analgesia is lacking for children owing to interindividual variations and underdosing of opioids caused by fear of adverse effects. We investigated the role of *COMT* SNPs on postoperative pain management in children. **Methods:** One hundred and forty nine children undergoing adenotonsillectomy were enrolled. The associations of four *COMT* SNPs (rs6269, rs4633, rs4818 and rs4680) with postoperative pain were analyzed and outcome measures included maximum pain scores, need for postoperative opioid interventions and postoperative morphine requirements. **Results:** We detected an association of postoperative opioid intervention need with all four *COMT* SNPs. Minor allele carriers of *COMT* SNPs were approximately three-times more likely to require analgesic interventions than homozygotes of major alleles (p-value range: 0.0031–0.0127; odds ratio range: 2.6–3.1). In addition, significant association was detected between maximum Face, Leg, Activity, Consolability, Cry (FLACC) pain scores and three *COMT* SNPs (rs6269, rs4633 and rs4680). Haplotype 1 (ATCA: 51.3%) and Haplotype 2 (GCGG: 36.2%) are more frequent. Haplotype 2 was associated with higher odds of intravenous analgesic intervention need in postanesthesia recovery unit with an odds ratio of 2.6 (95% CI: 1.2–5.4; p-value = 0.022). **Conclusion:** *COMT* SNPs may play a significant role in interindividual variation in postoperative pain perception and postoperative morphine requirements in children.

Original submitted 16 August 2013; Revision submitted 13 December 2013

KEYWORDS: *COMT* ■ genetics ■ pain ■ pediatric pain ■ postoperative pain

Perioperative pain management in children is often inadequate with up to 50% experiencing inadequate pain control and serious side effects from narcotics [1–4]. There is a wide range of responses to analgesics administered to treat postoperative pain secondary to variability in patient age [2], gender, race (ancestry), anxiety [5], type of surgery, previous experience [2] and genetic factors. It is currently believed that up to two-thirds of the variability in morphine response and adverse effects results from genetic variation in adults and children [6,7].

Several adult pain studies have shown an association between *COMT* genetic variations and interindividual variability in pain perception [8–11]. *COMT* is the enzyme mainly responsible for the metabolism of noradrenaline, adrenaline and dopamine. *COMT* is a key regulator of pain perception, cognitive function and mood, and is a pivotal regulator of catecholamine concentrations in the pain perception pathway [12,13]. Four common *COMT* SNPs (rs6269, rs4633, rs4818 and rs4680 – Val158Met) define haplotypes predictive of lower *COMT* enzyme activity [10]. Lower *COMT* activity increases catecholamine levels, causes hyperalgesia through stimulation of β_2 -adrenergic receptors [14,15] and increases

dopaminergic transmission leading to depletion of enkephalin in the brain and upregulation of opioid receptors [16]. Catecholamines and their metabolism may play a significant role in pediatric surgical pain. Therefore, a better understanding of the *COMT* gene in children may allow for individual tailoring and optimization of analgesic therapy, including opioids.

Little literature exists on the effect of *COMT* variations on postoperative pain perception and morphine analgesia in children [17,18], and large pediatric studies with homogenous pain phenotype are lacking. We studied *COMT* candidate polymorphisms, which have been shown in adults to be significantly associated with pain perceptions and analgesia [8–11,19–24], in a homogenous sample of children undergoing tonsillectomy, to identify genetic profiles associated with increased postoperative pain and inadequate postoperative analgesia with morphine.

Methods

We performed a prospective, genotype-blinded, observational study in a cohort of children undergoing adenotonsillectomy. This study is part of a larger ongoing clinical study, Personalizing Perioperative Morphine Analgesia in

Senthilkumar
Sadhasivam*¹,
Vidya Chidambaran¹,
Vanessa A Olbrecht¹,
Hope R Esslinger¹,
Kejian Zhang²,
Xue Zhang²
& Lisa J Martin²

¹Clinical Anaesthesia & Paediatrics, Acute & Perioperative Pain Service, Department of Anaesthesia, Cincinnati Children's Hospital Medical Center, 3333 Burnet Avenue, MLC 2001, Cincinnati, OH 45229, USA

²Division of Human Genetics, Cincinnati Children's Hospital Medical Center, 3333 Burnet Avenue, Cincinnati, OH 45040, USA

*Author for correspondence:

Tel.: +1 513 636 4408

Fax: +1 513 636 7337

senthilkumar.sadhasivam@cchmc.org

Children, which is registered with clinicaltrials.gov (NCT01140724). The study was approved by the Cincinnati Children's Hospital Medical Center's institutional review board and written informed consent was obtained from parents and assent was obtained from children before enrollment.

Children scheduled for elective, outpatient adenotonsillectomy were enrolled. Children between 6 and 15 years of age and American Society of Anesthesiologists (ASA) status 1 or 2, including children with obstructive sleep apnea (OSA) and recurrent adenotonsillitis were recruited. Self-reported race by parents was used to determine ancestry. Children from European (Caucasian) and African (African-American) and other ancestries were included. Children were excluded if they were non-English speaking, had an allergy to morphine, were developmentally delayed, had liver or renal disease, or had preoperative opioid use. All children received standard and uniform perioperative care with an anesthetic consisting of an intraoperative dose of 0.2 mg/kg of morphine (children with OSA received 0.1 mg/kg of morphine). Intraoperatively, all children received prophylactic antiemetics dexamethasone (0.1 mg/kg, maximum 4 mg) and ondansetron (0.1 mg/kg, maximum 4 mg). Tonsillectomy was performed in all children using an electrocautery technique. Additional use of intra- and post-operative morphine (0.05 mg/kg/dose) was administered per discretion of the anesthesia team based on clinical need.

Analgesic outcome measures were postoperative pain scores and postoperative intravenous analgesic interventions needed to achieve comfort, postoperative morphine use, prolonged postanesthesia recovery unit (PACU) stay due to inadequate pain control and duration to achieve PACU discharge readiness. Postoperative pain scores in PACU were simultaneously measured using the Face, Leg, Activity, Cry and Consolability (FLACC) scale by nursing staff and a subjective patient report of Numerical Rating Scale (NRS) upon arrival and every 5 min for 15 min, and then every 15 min until discharge. After intravenous opioid interventions, pain scores were measured every 5 min for 15 min until pain scores were below 4 out of 10. For pain scores >4 in PACU, intravenous opioids were administered as analgesic interventions. Intravenous morphine in 0.05 mg/kg doses were administered for significant (pain scores >4 out of 10) postoperative pain. We defined prolonged PACU stay as greater than 90 min following arrival and prior to

discharge. The criteria to be met for PACU discharge were: level of consciousness arousable or awake, airway patent with adequate air exchange, core temperature $\geq 36.3^{\circ}\text{C}$, acceptable pain level, hemodynamically stable and surgical site without any bleeding or complications.

Blood was drawn in the operating room to obtain DNA for genotyping. *COMT* SNPs were analyzed by TaqMan[®] genotyping assays (Life Technologies, Applied Biosystems, CA, USA).

To test the hypothesis that *COMT* SNPs were associated with postoperative analgesia outcomes in children, statistical analyses were conducted using SAS 9.3 and JMP Genomics 5.1 (SAS Institute, NC, USA). Prior to analysis, distributional qualities of all outcomes were examined. For the *COMT* variants, allelic and genotype frequencies were examined, and Hardy-Weinberg equilibrium (HWE) was tested [25]. HWE states that both allele and genotype frequencies in a population remain constant, and thus provides an ideal baseline against which changes in a population can be analyzed [25]. To analyze dichotomous outcomes (intervention need and prolonged PACU), logistic regression was performed. For postoperative morphine use and duration for PACU discharge readiness, general linear models were used because these outcomes followed a normal distribution. The maximum NRS pain scores were not normally distributed thus were analyzed using Wilcoxon rank sum test. FLACC exhibited a zero-inflated negative binomial distribution with 27.5% of the patients experiencing no pain (score = 0). Thus, FLACC were analyzed using a zero-inflated negative binomial model to appropriately capture the distribution of this variable. Prior to evaluation of *COMT* variants, the effects of covariates (age, sex, ancestry, weight, OSA, intraoperative morphine and total morphine) were evaluated. To select the best fitting model, log likelihood, Akaike and Bayesian Information Criterion were compared, and residuals were examined. Covariates that significantly improved model fitting ($p \leq 0.05$) were retained for genetic analyses. For dichotomous outcomes, adjusted odds ratio and 95% CIs were reported; for the rest, least square means and 95% CIs were reported. To confirm stability of the estimated odds ratios, random subsampling cross-validation was applied. Haplotypes were estimated and assigned by JMP Genomics.

In this study, since the clinical outcomes correlated with each other (mean Rho: 0.43, range: 0.16–0.74), we did not perform a very conservative Bonferroni correction for multiple testing.

Instead, a correlation adjusted Bonferroni correction [101], was applied ($\alpha = 0.02$). As linkage disequilibrium was extremely high between the *COMT* SNPs ($D' = 0.99$), no additional multiple testing adjustment was performed. We also report associations reaching $\alpha = 0.05$ as nominally associated.

Results

Participants were primarily non-Hispanic Caucasian (of white or European ancestry) children with slightly more girls than boys. The majority of participants were between 7 and 11 years of age. All four *COMT* SNPs were common with minor allele frequencies ranging from 0.37 to 0.48, and all were in HWE. Demographic data and intraoperative and total morphine requirement (mean \pm standard deviation) are summarized in TABLE 1. Allelic and genotype frequencies of the four *COMT* SNPs are shown in TABLE 2.

A relationship was observed between postoperative rescue analgesic intervention need and *COMT* genotypes, with the minor allele carriers having a larger proportion of individuals requiring postoperative rescue analgesic intervention (TABLE 3). After adjusting for ancestry and intraoperative morphine, significant association of intervention need was detected with all four *COMT* SNPs (p-value range: 0.0031–0.0127; odds ratio range: 2.6–3.1, FIGURE 1). Cross-validation supported the validity and stability of these findings. Children with the CC genotype of the *COMT* SNP rs4818 compared to minor allele carriers demonstrated a trend towards

Table 1. Demographic characteristics.

Characteristic	Value (total n = 149)
Age (years), median (IQR)	8.4 (7.1–11.2)
Weight (kg), median (IQR)	36.1 (26.5–51.4)
Sex, n (%)	
Female	80 (54)
Male	69 (46)
Race, n (%)	
White	121 (81)
Non-white	28 (19)
OSA	
Yes, n (%)	68 (46)
No, n (%)	81 (54)
Intraoperative morphine requirement (mg/kg), mean \pm SD	0.19 \pm 0.06
Total morphine requirement (mg/kg), mean \pm SD	0.25 \pm 0.08

IQR: Interquartile range; OSA: Obstructive sleep apnea; SD: Standard deviation.

increased postoperative morphine requirement ($p = 0.07$; TABLE 3).

Another analgesic outcome exhibiting significant association was maximum FLACC score. One SNP (rs4633) exhibited significant association and two others (rs6269 and rs4680) exhibited nominal association (TABLE 4). NRS, duration to PACU discharge readiness and prolonged stay in PACU due to pain were not associated with the *COMT* SNPs ($p > 0.05$).

Haplotype analyses showed that Haplotype 1 (ATCA: 51.3%) and Haplotype 2 (GCGG:

Table 2. *COMT* alleles and genotypes.

SNP	Allele			Genotype			HWE test (p-value)
	Allele	Count	Frequency	Genotype	Count	Frequency	
rs6269	A	175	0.59	A/A	49	0.33	0.42
	G	123	0.41	A/G	77	0.52	
				G/G	23	0.15	
rs4633	C	142	0.48	C/C	30	0.20	0.21
	T	156	0.52	C/T	82	0.55	
				T/T	37	0.25	
rs4818	C	189	0.63	C/C	58	0.39	0.49
	G	109	0.37	C/G	73	0.49	
				G/G	18	0.12	
rs4680	A	154	0.52	A/A	38	0.26	0.56
	G	144	0.48	A/G	78	0.52	
				G/G	33	0.22	

Four *COMT* polymorphisms were tested, allelic frequency and HWE from the study cohort are reported.
HWE: Hardy–Weinberg equilibrium.

Table 3. Morphine need, pain outcomes and time in postanesthesia recovery unit stratified by *COMT* genotypes.

SNP	<i>COMT</i> genotype	n	Postoperative analgesic intervention need (% yes)	Prolonged PACU stay due to pain (% yes)	Postoperative morphine requirement, mg/kg (mean ± SD)	Maximum FLACC score, median (IQR)	Maximum NRS score, median (IQR)	Time to PACU discharge readiness, min (mean ± SD)
rs6269	AA	49	45	24	0.04 ± 0.05	2 (0–4)	6 (2–8)	104.7 ± 41.3
	GA [†]	77	70	38	0.07 ± 0.06	4 (1–6)	6 (3–8)	104.6 ± 50.6
	GG [†]	23	74	35	0.07 ± 0.06	4 (1–6)	6 (2–8)	92.6 ± 30.8
rs4633	CC [†]	30	70	33	0.07 ± 0.06	4 (1–6)	6 (2–8)	93.2 ± 28.7
	CT [†]	82	68	38	0.07 ± 0.07	4 (0–6)	6 (3–8)	107.3 ± 52.6
	TT	37	43	22	0.03 ± 0.04	2 (0–3)	6 (2–8)	100.4 ± 36.0
rs4818	CC	58	50	29	0.05 ± 0.07	2 (0–5)	6 (3–8)	104.9 ± 42.2
	CG [†]	73	71	37	0.06 ± 0.05	3 (1–5)	6 (4–8)	104.7 ± 49.9
	GG [†]	18	67	28	0.06 ± 0.05	3 (1–6)	6 (2–7)	87.9 ± 28.8
rs4680	AA	38	45	24	0.04 ± 0.05	2 (0–4)	6 (2–8)	100.1 ± 35.5
	AG [†]	78	68	37	0.07 ± 0.07	4(0–6)	5 (3–8)	107.9 ± 53.9
	GG [†]	33	70	33	0.06 ± 0.06	3 (1–6)	6 (3–9)	93.6 ± 27.7

[†]Genotypes carrying minor allele.

FLACC: Face, Leg, Activity, Cry and Consolability; IQR: Interquartile range; NRS: Numerical Rating Scale; PACU: Postanaesthesia recovery unit; SD: Standard deviation.

36.2%) are more frequent (TABLE 5). Single haplotype association tests were carried out on continuous pain outcomes (TABLE 5). Nominal association was detected between Haplotype 1 and Haplotype 2 with intravenous analgesic intervention need in PACU, with opposite effects.

This is consistent with our single SNP associations. Haplotypes 4 and 5 are associated with postoperative morphine requirement. In the single SNP association test, postoperative morphine requirement was associated with rs4818. The haplotype association could be due to segregation of rs4818 genotypes. Haplotypes 4, 5 and 6 are too rare to develop stable statistical models. Children that are homozygous for Haplotype 1 had lower incidence of intravenous analgesic intervention need in PACU. Heterozygous Haplotypes 1 and 2 and homozygous Haplotype 2 were comparable and they had higher intravenous analgesic intervention need in PACU than homozygous Haplotype 1. This is consistent with the single SNP association tests. Haplotype 2 was associated with odds of higher intravenous analgesic intervention need in PACU with an odds ratio of 2.6 (95%CI: 1.2–5.4; p-value = 0.022).

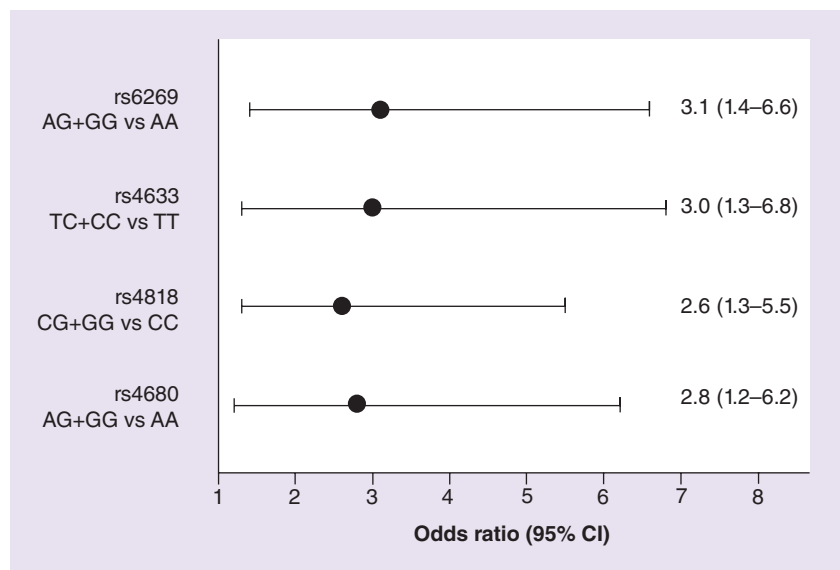


Figure 1. Relationship between postoperative intravenous rescue analgesic intervention need and *COMT* genotypes (grouped as minor and major alleles). The minor allele carriers had a larger proportion of children requiring postoperative rescue analgesic intervention (~threefold increase). After adjusting for ancestry and intraoperative morphine, significant associations of higher intervention need (odds ratio range: 2.6–3.1) were detected with all four *COMT* SNPs (p-value range: 0.0031–0.0127).

Discussion

Pain is a complex experience with sensory, emotional and cognitive components [4]. Morphine, one of the most commonly used opioids, has a narrow therapeutic index and large interpatient variability in both analgesic responses. Genetic variations in the gene coding for *COMT* have been suggested to affect clinical and experimental pain-related phenotypes [4]. Our findings indicate that *COMT* genetic variation is

associated with the need for postoperative analgesic intervention and postoperative pain in children. Although the role of *COMT* genotype with postoperative analgesia and morphine requirements has been studied in adults, this is the first study to address this issue in a population of children having homogenous surgical pain.

In this homogenous group of tonsillectomy patients, the need for postoperative morphine was significantly ($p < 0.013$) associated with four SNPs in *COMT* (rs6269, rs4633, rs4818 and rs4680). The strongest association (rs6269) yielded increased odds (3.1-fold) of postoperative intravenous opioid interventions in individuals with the copy of the G allele compared with the AA genotype. Additionally, there was significant association with the FLACC pain score and rs4633 ($p = 0.01$). Children with the CC genotype of rs4818 demonstrated a trend towards increased postoperative morphine requirement compared with other genotypes ($p = 0.07$, TABLE 3).

Our study results are consistent with a postoperative study in adults with postlumbal disc surgery [26]. Specifically, the haplotype with the protective allele from the four *COMT* SNPs in our pediatric study was associated with less analgesic interventions, which is consistent with the association between the TT genotype of the *COMT* SNP rs4633 and improved Oswestry Disability Index (ODI) was demonstrated 1 year after surgery in adult patients [26]. In a recent study of 109 Italian adults undergoing surgery, when 12 different polymorphisms of *COMT*, *OPRM1* and *UGT2B7* genes were investigated for association with postoperative morphine requirement, only *COMT* polymorphisms were predictive of morphine requirement [27]. Another recent study of postoperative cardiac patients showed that the *COMT* polymorphism Val158Met contributed to pain sensitivity [28]. These consistent postoperative pain and functional outcomes in our study children and adults suggest that patients with certain *COMT* genotypes are at higher risk for increased immediate postoperative pain.

Our pediatric study is one of the early studies looking at associations between genetic make up and postoperative pain in a homogeneous group of healthy children undergoing one type of surgery, tonsillectomy with electrocautery surgical technique with standard and uniform intraoperative and postoperative care to minimize confounding. In another recently published small study of 168 children undergoing heterogeneous surgical procedures, including abdominal and

Table 4. Postoperative pain, morphine analgesic efficacy and genetic associations with *COMT* polymorphisms.

SNP	Genotype	Postoperative analgesic intervention need		Prolonged stay in PACU due to inadequate analgesia*		Maximum FLACC pain score ⁵		Postoperative morphine requirement (mg/kg) ¹¹		Duration to PACU discharge readiness (min) [#]	
		Adjusted OR (95% CI)	p-value	Adjusted OR (95% CI)	p-value	LS mean (95% CI)	p-value	LS mean (95% CI)	p-value	LS mean (95% CI)	p-value
rs6269	AA	3.1 (1.4–6.6)	0.0031	1.7 (0.8–3.9)	0.16	3.9 (3.1–4.9)	0.04	0.09 (0.07–0.12)	0.58	107.1 (94.9–119.4)	0.39
	AG+GG					5.1 (4.4–5.8)		0.10 (0.09–0.11)		100.6 (92.1–109.1)	
rs4633	TT	3.0 (1.3–6.8)	0.0074	1.9 (0.8–4.8)	0.14	3.5 (2.6–4.6)	0.01	0.08 (0.05–0.11)	0.18	104.6 (90.4–118.9)	0.77
	TC+CC					5.0 (4.4–5.7)		0.10 (0.09–0.11)		102.1 (94.0–110.2)	
rs4818	CC	2.6 (1.3–5.5)	0.0082	1.3 (0.6–2.8)	0.45	4.4 (3.6–5.4)	0.32	0.11 (0.09–0.13)	0.07	105.3 (94.2–116.5)	0.56
	CG+GG					5.0 (4.3–5.7)		0.09 (0.08–0.11)		101.1 (92.2–110.0)	
rs4680	AA	2.8 (1.2–6.2)	0.0127	1.7 (0.7–4.0)	0.24	3.8 (2.9–5.0)	0.05	0.09 (0.06–0.12)	0.45	103.7 (89.7–117.7)	0.88
	AG+GG					5.0 (4.4–5.7)		0.10 (0.09–0.11)		102.4 (94.3–110.6)	

ORs are of minor allele carriers vs homozygous major allele carriers.

¹Adjusted for race and intraoperative morphine.

²Adjusted for race, obstructive sleep apnea, and race x obstructive sleep apnea.

³Adjusted for age, race, weight and intraoperative morphine.

⁴In patients who need postoperative morphine; adjusted for race.

⁵Adjusted for morphine by weight.

FLACC: Face, Leg, Activity, Cry and Consolability; LS: Least square; OR: Odds ratio; PACU: Postanaesthesia recovery unit.

Table 5. Postoperative pain, morphine analgesic efficacy and genetic associations with common *COMT* haplotypes.

Haplotype (frequency)	Postoperative [†] rescue analgesic intervention need		Prolonged stay in PACU due to inadequate analgesia [‡]		Maximum FLACC pain score [§]		Postoperative morphine requirement (mg/kg)		Duration to PACU discharge readiness (min) [#]	
	Adjusted OR (95% CI)	p-value	Adjusted OR (95% CI)	p-value	Effect size (95% CI)	p-value	Effect size (95% CI)	p-value	Effect size (95% CI)	p-value
A-T-C-A (51.3%)	0.6 (0.3–1.0)	0.0395	0.8 (0.5–1.4)	0.53	0.88 (0.74–1.04)	0.12	-0.00 (-0.02–0.01)	0.63	5.4 (-4.8–15.7)	0.30
G-C-G-C (36.2%)	1.8 (1.0–3.1)	0.0354	1.1 (0.6–1.8)	0.86	1.08 (0.92–1.28)	0.34	-0.01 (-0.03–0.01)	0.19	-6.9 (-17.3–3.6)	0.20
A-C-C-G (6.0%)	0.6 (0.2–1.6)	0.30	0.7 (0.2–2.0)	0.49	0.97 (0.70–1.34)	0.86	0.01 (-0.02–0.04)	0.63	2.9 (-17.3–23.0)	0.78
G-C-C-G (4.7%)	3.0 (0.5–17.2)	0.19	3.5 (0.8–15.7)	0.09	1.12 (0.80–1.56)	0.50	0.06 (0.03–0.09)	0.0003	0.4 (-24.4–25.2)	0.97
A-T-C-G (1%)	0.4 (0.0–5.1)	0.47	0.5 (0.0–6.5)	0.56	1.03 (0.50–2.13)	0.94	-0.08 (-0.15 to -0.01)	0.0303	5.0 (-45.0–55.1)	0.84
Rare (<1%)	3.0 (0.1–124.7)	0.18	7.1 (0.2–339.9)	0.32	1.61 (0.84–3.08)	0.17	0.05 (-0.02–0.13)	0.13	3.7 (-57.9–65.3)	0.91

OR is for each additional copy of the haplotype.
Effect size: mean changes for one copy change of haplotype.
[†]Adjusted for race and intraoperative morphine.
[‡]Adjusted for race, obstructive sleep apnea, and race x obstructive sleep apnea.
[§]Adjusted for age, race, weight and intraoperative morphine.
^{||}In patients who need postoperative morphine; adjusted for race.
[#]Adjusted for morphine by weight.
FLACC: Face, Leg, Activity, Cry and Consolability; OR: Odds ratio; PACU: Post-anaesthesia recovery unit.

orthopedic surgery, postoperative pain scores during mobilization were lower for GG genotype than AA and GA genotypes of the *COMT* polymorphism Val158Met [18]. Another recent trial comparing 68 Latino and non-Latino children showed a statistically significant difference in morphine's response; however, none of the 28 genetic polymorphisms examined in this study, including *COMT* gene polymorphisms, could explain the ethnicity-related differences in morphine's response, this is probably because of the limited sample size and the heterogeneous ethnicity of the study population [17]. Recently, the 158A>G variant in the *COMT* gene (rs4680), specifically the AG/GG genotype was associated with shortened length of stay and less treatment with two or more medications than the AA genotype among infants with neonatal abstinence syndrome caused by *in utero* opioid exposure, providing early insights into the mechanisms underlying neonatal opioid withdrawal [7]. This *COMT* variant has a minor allele frequency of approximately 50% in Caucasians (in our study population it was 48%) and has been associated with responses to pain and morphine dosing requirements in adults [29].

In our study, although both the pain measures of NRS and FLACC are validated measures, only FLACC exhibited an association with *COMT* genotyping. The failure to identify associations with NRS could be because the NRS is a subjective measure of pain versus FLACC, which is a relatively objective measure of pain. Clearly, additional studies should examine the relationship of NRS with FLACC to better understand the factors that may influence differential reporting of postoperative pain in children.

Conclusion & future perspective

By demonstrating an association between morphine need after surgery and *COMT* genetic variation, this study contributes to a greater understanding of the underlying genetic basis of why children vary in their sensitivity to pain and postoperative analgesia. It provides essential preliminary supportive results toward establishing associations between *COMT* genotypes and postoperative pain and opioid use related outcomes in children. It is important to note, however, that, besides *COMT*, many genes may influence morphine's effects, and morphine's pharmacokinetics. While variations at the *COMT* gene have been shown to influence pain sensitivity and postoperative morphine need in children, postoperative pain and opioid responses could be influenced by many different

genes [15,17,18,30]. Large clinical trials having adequate power for population stratification to study simultaneously multiple genes potentially affecting pain and opioid responses along with gene–gene interactions over longer postoperative course in future will contribute to the essential foundation for genotype-directed analgesic selection and dosing to maximize pain relief while improving patient safety by minimizing serious side effects.

Trial registration

This clinical trial reflects a portion of a larger study, *Personalizing Perioperative Morphine Analgesia in Children*, NCT01140724, registered at clinicaltrials.gov.

Authors' contribution

S Sadhasivam conceptualized and designed the study, designed the data collection instruments, recruited subjects, and coordinated and supervised data collection, drafted the initial manuscript, and approved the final manuscript as submitted. V Olbrecht helped with the initial draft, reviewed and revised the manuscript, and approved the final manuscript as submitted. V Chidambaran helped with recruitment, reviewed and revised the manuscript, and approved the final manuscript as submitted. HR Esslinger helped with patient recruitment, data collection and approved the final manuscript. K Zhang helped with genetic study design and analysis. X Zhang helped with data analyses, draft statistical section, tables and revision of the manuscript. LJ Martin helped with data analyses, statistical methods, revision of the manuscript and approved the final manuscript as submitted.

Financial & competing interests disclosure

This work was supported in part by USPHS Grant #UL1 RR026314 from the National Center for Research Resources, NIH and with the Place Outcomes Research Award and Translational Research Award, Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA. Additional research funding support was provided by the Department of Anesthesia, Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA. No financial support was received except for departmental salary support for the authors. This pharmacogenetic study was designed and undertaken by the authors. The sponsor of this study, the Cincinnati Children's Hospital Medical Center (CCHMC) provided funding support for the genetic analyses and supported the salary of the research team. The authors directed and had access to all the analyses and the full clinical and genetic database, wrote all drafts of the report, decided to publish the results, and attest for the accuracy and completeness of the data. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

No writing assistance was utilized in the production of this manuscript.

Ethical conduct of research

The authors state that they have obtained appropriate institutional review board approval or have followed the principles outlined in the Declaration of Helsinki for all human or animal experimental investigations. In addition, for investigations involving human subjects, informed consent has been obtained from the participants involved.

Executive summary

Interindividual differences in postoperative pain & opioid requirement are associated with common polymorphisms of COMT gene

- Genetic variants of *COMT* play a role in interindividual variations in postoperative pain perception and postoperative morphine requirements in children undergoing adenotonsillectomy.
- Minor allele carriers of *COMT* polymorphisms (rs6269, rs4633, rs4818 and rs4680) were approximately three-times more likely to require analgesic interventions than homozygotes of major alleles (odds ratio range: 2.6–3.1; p-value range: 0.0031–0.0127).
- Haplotype 2 (GCGG: 36.2%), which was the second most common haplotype of these four *COMT* SNPs was associated with higher odds of intravenous analgesic intervention need in PACU with an odds ratio of 2.6 (95% CI: 1.2–5.4; p = 0.022).

Interindividual difference in postoperative pain & postoperative analgesic requirement is caused by multiple factors

- Besides *COMT* genetic variations, other genetic and nongenetic factors, and gene–gene interactions might contribute to variations in surgical pain perception and analgesic requirement in the perioperative period.
- Large clinical studies to simultaneously study *COMT* and multiple other genes affecting pain and opioid responses will contribute to the essential foundation for genotype-directed analgesic selection and dosing to maximize pain relief while improving patient safety by minimizing serious side effects.

References

- 1 Walco GA, Cassidy RC, Schechter NL. Pain, hurt, and harm. The ethics of pain control in infants and children. *N. Engl. J. Med.* 331(8), 541–544 (1994).
- 2 Berde CB, Sethna NF. Analgesics for the treatment of pain in children. *N. Engl. J. Med.* 347(14), 1094–1103 (2002).
- 3 Beaulieu P, Cyrenne L, Mathews S, Villeneuve E, Vischoff D. Patient-controlled analgesia after spinal fusion for idiopathic scoliosis. *Int. Orthop.* 20(5), 295–299 (1996).
- 4 Duedahl TH, Hansen EH. A qualitative systematic review of morphine treatment in children with postoperative pain. *Paediatr. Anaesth.* 17(8), 756–774 (2007).

- 5 Schechter NL, Blankson V, Pachter LM, Sullivan CM, Costa L. The ouchless place: no pain, children's gain. *Pediatrics* 99(6), 890–894 (1997).
- 6 Ross JR, Rutter D, Welsh K *et al.* Clinical response to morphine in cancer patients and genetic variation in candidate genes. *Pharmacogenomics J.* 5(5), 324–336 (2005).
- 7 Wachman EM, Hayes MJ, Brown MS *et al.* Association of *OPRM1* and *COMT* single-nucleotide polymorphisms with hospital length of stay and treatment of neonatal abstinence syndrome. *JAMA* 309(17), 1821–1827 (2013).
- 8 Nackley AG, Tan KS, Fecho K, Flood P, Diatchenko L, Maixner W. Catechol-*O*-methyltransferase inhibition increases pain sensitivity through activation of both β 2- and β 3-adrenergic receptors. *Pain* 128(3), 199–208 (2007).
- 9 Diatchenko L, Nackley AG, Slade GD *et al.* Catechol-*O*-methyltransferase gene polymorphisms are associated with multiple pain-evoking stimuli. *Pain* 125(3), 216–224 (2006).
- 10 Diatchenko L, Slade GD, Nackley AG *et al.* Genetic basis for individual variations in pain perception and the development of a chronic pain condition. *Hum. Mol. Genet.* 14(1), 135–143 (2005).
- 11 Reyes-Gibby CC, Shete S, Rakvag T *et al.* Exploring joint effects of genes and the clinical efficacy of morphine for cancer pain: *OPRM1* and *COMT* gene. *Pain* 130(1–2), 25–30 (2007).
- 12 Lotta T, Vidgren J, Tilgmann C *et al.* Kinetics of human soluble and membrane-bound catechol *O*-methyltransferase: a revised mechanism and description of the thermolabile variant of the enzyme. *Biochemistry* 34(13), 4202–4210 (1995).
- 13 Zubietta JK, Heitzeg MM, Smith YR *et al.* *COMT* val158met genotype affects mu-opioid neurotransmitter responses to a pain stressor. *Science* 299(5610), 1240–1243 (2003).
- 14 Khasar SG, McCarter G, Levine JD. Epinephrine produces a β -adrenergic receptor-mediated mechanical hyperalgesia and *in vitro* sensitization of rat nociceptors. *J. Neurophysiol.* 81(3), 1104–1112 (1999).
- 15 Sadhasivam S, Chidambaran V. Pharmacogenomics of opioids and perioperative pain management. *Pharmacogenomics* 13(15), 1719–1740 (2012).
- 16 Berthele A, Platzer S, Jochim B *et al.* *COMT* Val108/158Met genotype affects the mu-opioid receptor system in the human brain: evidence from ligand-binding, G-protein activation and preproenkephalin mRNA expression. *Neuroimage* 28(1), 185–193 (2005).
- 17 Jimenez N, Anderson GD, Shen DD *et al.* Is ethnicity associated with morphine's side effects in children? Morphine pharmacokinetics, analgesic response, and side effects in children having tonsillectomy. *Paediatr. Anaesth.* 22(7), 669–675 (2012).
- 18 Mamie C, Rebsamen MC, Morris MA, Morabia A. First evidence of a polygenic susceptibility to pain in a pediatric cohort. *Anesth. Analg.* 116(1), 170–177 (2013).
- 19 Oertel BG, Schmidt R, Schneider A, Geisslinger G, Lotsch J. The mu-opioid receptor gene polymorphism 118A>G depletes alfentanil-induced analgesia and protects against respiratory depression in homozygous carriers. *Pharmacogenet. Genomics* 16(9), 625–636 (2006).
- 20 Chou WY, Wang CH, Liu PH, Liu CC, Tseng CC, Jawan B. Human opioid receptor A118G polymorphism affects intravenous patient-controlled analgesia morphine consumption after total abdominal hysterectomy. *Anesthesiology* 105(2), 334–337 (2006).
- 21 Chou WY, Yang LC, Lu HF *et al.* Association of mu-opioid receptor gene polymorphism (A118G) with variations in morphine consumption for analgesia after total knee arthroplasty. *Acta Anaesthesiol. Scand.* 50(7), 787–792 (2006).
- 22 Klepstad P, Rakvag TT, Kaasa S *et al.* The 118 A>G polymorphism in the human mu-opioid receptor gene may increase morphine requirements in patients with pain caused by malignant disease. *Acta Anaesthesiol. Scand.* 48(10), 1232–1239 (2004).
- 23 Coulbault L, Beaussier M, Verstuft C *et al.* Environmental and genetic factors associated with morphine response in the postoperative period. *Clin. Pharmacol. Ther.* 79(4), 316–324 (2006).
- 24 Collier JK, Barratt DT, Dahlen K, Loennechen MH, Somogyi AA. *ABCBI* genetic variability and methadone dosage requirements in opioid-dependent individuals. *Clin. Pharmacol. Ther.* 80(6), 682–690 (2006).
- 25 Stern C. The Hardy–Weinberg law. *Science* 97(2510), 137–138 (1943).
- 26 Dai F, Belfer I, Schwartz CE *et al.* Association of catechol-*O*-methyltransferase genetic variants with outcome in patients undergoing surgical treatment for lumbar degenerative disc disease. *Spine J.* 10(11), 949–957 (2010).
- 27 De Gregori M, Garbin G, De Gregori S *et al.* Genetic variability at *COMT* but not at *OPRM1* and *UGT2B7* loci modulates morphine analgesic response in acute postoperative pain. *Eur. J. Clin. Pharmacol.* 69(9), 1651–1658 (2013).
- 28 Ahlers SJ, Elens LL, van Gulik L *et al.* The Val158Met polymorphism of the *COMT* gene is associated with increased pain sensitivity in morphine-treated patients undergoing a painful procedure after cardiac surgery. *Br. J. Clin. Pharmacol.* 75(6), 1506–1515 (2013).
- 29 Rakvag TT, Klepstad P, Baar C *et al.* The Val158Met polymorphism of the human catechol-*O*-methyltransferase (*COMT*) gene may influence morphine requirements in cancer pain patients. *Pain* 116(1–2), 73–78 (2005).
- 30 Sadhasivam S, Krekels EH, Chidambaran V *et al.* Morphine clearance in children: does race or genetics matter? *J. Opioid Manag.* 8(4), 217–226 (2012).

Website

- 101 SISA.
www.quantitativeskills.com/sisa/