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Minerva Anestesiologica 2018 August;84(8):946-54
DOI: 10.23736/S0375-9393.18.12697-6

ORIGINAL ARTICLE

μ-opioid receptor genetic polymorphisms and duration of epidural fentanyl analgesia during early labor

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ABSTRACT

BACKGROUND: Epidural fentanyl is commonly used for initiation of early labor analgesia. The aim of this prospective study is to test the hypothesis that duration of epidural fentanyl analgesia differs in nulliparous women requesting epidural analgesia in early labor who are variant allele carriers of the *OPRM1* SNVs *118A>G* rs1799971, *IVS2+31G>A* rs9479757, and *IVS2+691G>C* rs2075572.

METHODS: Two hundred and twenty parturients who received epidural analgesia with fentanyl were included in the *118A>G* analysis, and a 196 sub-cohort was included in the *IVS2+31G>A* and *IVS2+691G>C* exploratory analysis. Peripheral blood DNA was genotyped using Taqman allele discrimination assays.

RESULTS: One hundred and seventy-four subjects (79%; 95% CI: 74-84) were homozygous for the wild type *OPRM1 118A>G* SNV (AA), and forty-six subjects (21%; 95% CI: 16-26) were heterozygous AG (N.=40) or homozygous GG (N.=6) for the variant allele. The minor allele frequency (MAF) was hence 12%. The MAFs for the *IVS2+31G>A* and *IVS2+691G>C* SNVs in the sub-cohort of 196 participants were 5% and 59% respectively. There was no significant difference in duration of epidural fentanyl analgesia for the three SNVs (161±68 and 143±51 min for wild type and allele carriers of the *118A>G* SNV respectively [P=0.08]). Similarly, no significant differences were shown with the visual analog scale scores, side effects, and satisfaction for each of the three SNVs.

CONCLUSIONS: *OPRM1* SNVs did not affect the duration of epidural fentanyl administered for early labor analgesia in nulliparous women. These results should be confirmed in patients receiving epidural opioids in other clinical settings.

(*Cite this article as:* Zgheib NK, Aouad MT, Taha SK, Nassar AH, Masri RF, Khoury MY, *et al.* μ-opioid receptor genetic polymorphisms and duration of epidural fentanyl analgesia during early labor. Minerva Anestesiol 2018;84:946-54. DOI: 10.23736/S0375-9393.18.12697-6)

KEY WORDS: Analgesia, epidural - Fentanyl - Labor, obstetric - OPRM1 protein, human.

Lipid soluble opioids, such as fentanyl, are commonly used for initiation of labor analgesia via neuraxial route. When epidural fentanyl was given as the sole analgesic without local anesthetic, large doses ranging between 150 and 200 μg provided satisfactory labor analgesia.[1, 2] In addition, Connelly and colleagues have shown that 100 μg epidural fentanyl after a lidocaine-epinephrine test dose provides a mean±SD duration of labor analgesia of 108±40 to 145±50 min.[3, 4] This inter-individual variability in dose and duration may arise from multiple factors such as ethnicity, parity, and stage of labor.[5] In addition, genetic polymorphisms in pharma-

cokinetic (*e.g.* drug metabolizing enzymes and drug transporters) and pharmacodynamic (*e.g.* μ-opioid receptors - *OPRM1*) candidate genes may play a role in opioid (including fentanyl) bioavailability and responsiveness.[5, 6]

Over the past decade, a number of pharmacogenetic studies have implicated the role of a common *OPRM1* single nucleotide variant (SNV), *118A>G* (dbSNP rs1799971), in alteration of individual response to opioid analgesia.[5-8] This polymorphism leads to a variant μ-opioid receptor in which an asparagine is substituted for aspartate at the 40th amino acid of the protein (p.Asn40Asp) which corresponds to the N-terminal region of the receptor in the extracellular space, and therefore leading to loss of a putative N-glycosylation site and reduced receptor stability.[9]

Results of previous studies assessing the possible role of *OPRM1 118A>G* in pregnant laboring population and other pain models are widely divergent. For instance, two previous studies found no difference in the duration of intrathecal fentanyl analgesia for labor pain according to *OPRM1* genotype,[10, 11] while two others demonstrated that the variant allele of *OPRM1 118A>G* was associated with higher potency manifested by lower ED50 for epidural sufentanil[12] and intrathecal fentanyl[13] for early labor analgesia.

No associations were found with two additional, though considerably less studied, synonymous SNVs [*IVS2+31G>A* (dbSNP rs9479757) and *IVS2+691G>C* (dbSNP rs2075572)] in relation to opioid use (mainly morphine) in chronic cancer and postoperative pain.[14-16] This is despite evidence that these two SNVs, though intronic, may change the affinity of the transcriptional regulatory factors and hence altering mRNA expression.[17-19]

To the best of our knowledge, the effect of *OPRM1* genetic polymorphisms on the duration of epidural fentanyl analgesia in early labor has not been previously described. Given that ethnicity plays a major role in pharmacogenetic analyses due to the variable allele frequencies and the differential effect of these polymorphisms among different populations,[11, 20-22] we conducted a prospective study to test the hypothesis that duration of epidural fentanyl analgesia differs in Middle Eastern (Lebanese) nulliparous women requesting epidural analgesia in early labor who are variant allele carriers of the *OPRM1* SNVs *118A>G* rs1799971, *IVS2+31G>A* rs9479757, and *IVS2+691G>C* rs2075572 when compared to homozygous wild types.

Materials and methods

This study was conducted with institutional review board approval and was registered at the www.clinicaltrials.gov protocol registration system (NCT02099877).

Subjects

Eligible subjects were recruited over three years between June 2013 and June 2016. Written informed consent to participate in the study was obtained shortly after admission to the maternity unit from nulliparous parturients, ASA physical status 2, in early labor (cervical dilatation 2-5 cm), gestational age ≥37 weeks, with a singleton pregnancy in vertex presentation, in spontaneous labor or with spontaneous rupture of membrane, and requesting epidural analgesia for pain relief. A reassuring fetal heart rate pattern was required for inclusion in the study. Women with severe medical or obstetric complications, contraindication to epidural analgesia, local anesthetics or fentanyl allergy, and history of chronic opioid use, substance abuse, or systemic opioid analgesia before initiation of epidural analgesia were excluded from the study.

Analgesia

All subjects received oxytocin infusion for labor augmentation. During contractions, all parturients completed a baseline assessment of pain using a visual analog scale (VAS) with a 100 mm unmarked line (0 mm=no pain and 100 mm=worst possible pain). Upon patient's request for analgesia, either the obstetric resident or attending examined the cervix to verify the extent of dilation. Then epidural analgesia was initiated with a test dose of 3 mL of 1.5% lidocaine and epinephrine 15 μg followed by a dose of fentanyl 100 μg diluted to a total volume of 10 mL with preservative-free normal saline.

At second request of analgesia, the cervix was again examined, followed by epidural administration of a mixture of 10-15 mL of bupivacaine 1 mg/mL and fentanyl 2 μg/mL. Once pain relief was achieved, participants received patient-controlled epidural analgesia (PCEA) with the same solution (bolus 5 mL; lockout 15 min, and background infusion 5 mL/h). Manual bolus injections of bupivacaine 2.5 mg/mL were given by the anesthesiologist to treat breakthrough pain.

Subjects were withdrawn from analysis if they failed analgesia (VAS >30 mm 30 min after the epidural injection of fentanyl), if the subject progressed to either vaginal or cesarean delivery before the second request for analgesia, or breach of the protocol.

Data collection

The primary outcome was duration of epidural fentanyl analgesia, defined as the interval time from initial epidural fentanyl dose to subsequent request for epidural analgesia. During this time period, patients were asked to report about the presence of pruritus (none, mild, moderate, or severe), nausea (none, mild, moderate or severe), and vomiting (yes, no). Times from fentanyl initial administration to complete cervical dilation (10 cm) and delivery were also calculated. VAS scores for pain were recorded 10, 20, and 30 min after the administration of epidural fentanyl, and at the second request for analgesia. In addition, the maternal demographic data, maximum oxytocin dose, mode of delivery, neonatal weight, and Apgar scores were recorded. One to two hours after delivery, subjects were asked about their satisfaction with labor analgesia using a 100 mm scale with 0 mm being not satisfied at all and 100 mm being very satisfied. At that point, the study ended.

DNA collection and genotyping

Venous whole blood (2 mL) was sampled into coded EDTA tubes at the time of study enrollment in the maternity unit and stored at -80 °C. Whole blood DNA was then isolated using Qiagen (Germantown, MD) DNA isolation kit according to the manufacturer's guidelines, and stored at -20°C until analysis. Genotyping of the 3 *OPRM1* SNVs (*118A>G* rs1799971; *IVS2+31G>A* rs9479757; and *IVS2+691G>C* rs2075572) was performed using 3 Taqman® allele discrimination assays on a CFX36 real-time PCR from BioRad. Ten percent of the samples were run in duplicates to check for assay reproducibility.

The anesthesiologist performing the epidural anesthesia and the research assistant enrolling patients and collecting data were unaware of subjects' genotypes at the time of clinical assessment as genotyping was conducted after delivery. The technician who performed the genotyping assays was also blinded to clinical data.

Statistical analysis

Sample size was estimated for a potential association between *OPRM1 118A>G* polymorphism and the primary outcome (duration of epidural fentanyl analgesia). Mean and (SD) for sample size calculation was based on a previous study for the duration of epidural fentanyl for labor analgesia (126±57min).[4] We assumed that 70% of subjects would be homozygous for the wild type *OPRM1 118A>G* SNV (AA) and 30% would be homozygous or heterozygous for the variant allele (AG & GG).[6, 23] In order to demonstrate a 25 min difference in the time to request additional analgesia between the two groups, a difference that is considered clinically significant, a number of 220 patients was needed for the study (2-sided, α=0.05, β=0.2). The sample size was increased to 250 to compensate for potential subject loss that may occur during the course of the study.

Note that this study was originally conceived to test the role of *OPRM1 118A>G* SNV on response to epidural analgesia during labor. Nevertheless, and at the conclusion of recruitment, we decided to include genotyping for *IVS2+31G>A* and *IVS2+691G>C* SNVs as exploratory outcomes. This was performed after securing approval from the IRB and only on subjects who consented that their samples be used for additional genetic investigations.

Genetic association analysis was performed, and data for the recessive allele model comparing wild type genotypes with variant allele carriers are presented. Allele frequencies for the three SNVs were tested for Hardy Weinberg Equilibrium (HWE) using χ^2. Continuous data are reported as median and interquartile range (IQR)

or mean and standard deviation (SD) and were analyzed using Student's *t*-test (for normally distributed variable or sample size more than 30) or Mann-Whitney U Test (for non-normally distribution and sample size less than 30). Categorical data are reported as numbers and percentages and were analyzed using the χ^2 test (sample size more than 30), or the Fisher Exact Test (sample size less than 30). The duration of epidural fentanyl analgesia was analyzed using Kaplan-Meier survival analysis with comparison between groups using log-rank test. All statistical analyses were performed using SPSS software (version 24). P<0.05 was considered statistically significant.

Results

Two hundred and fifty subjects were assessed for eligibility. Five subjects were not enrolled in the study because of cervical dilatation at analgesic request >5 cm. Twenty-five subjects were excluded from final analysis: 11 because of failed analgesia (VAS remaining >30 mm 30 min after the epidural injection of fentanyl), nine because of rapid progression to cesarean delivery before the request for additional analgesia, and five because of protocol violation (incorrect study drug administered). These 220 subjects were all genotyped for *OPRM1 118A>G* SNV. One hundred and ninety-six subjects had consented that their samples be used for additional investigations, and were hence also genotyped for the *IVS2+31G>A* and *IVS2+691G>C* SNVs (Figure 1).

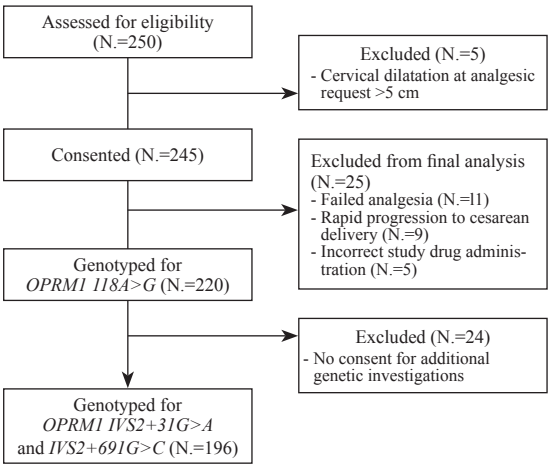

Figure 1.—Study flow diagram.

Genotyping results were 100% reproducible. Allele frequencies for the three SNVs were in Hardy Weinberg Equilibrium. As shown in Table I, one hundred and seventy-four subjects (79%; 95% CI: 74-84) were homozygous for the wild type *OPRM1 118A>G* SNV (AA), and forty-six subjects (21%; 95% CI: 16-26) were heterozygous AG (N.=40) or homozygous GG (N.=6) for the variant allele. The minor allele frequency (MAF) was hence 12%. In the sub-cohort of 196 participants, one hundred seventy-eight subjects (89%, 95% CI: 85-93) were homozygous for the *IVS2+31G>A* SNV (GG) and 18 subjects (11%; 95% CI: 7-15) were heterozygous (GA). The MAF for the *IVS2+31G>A* was hence 5%. Thirty-one subjects (15.8%; 95% CI: 11-21) were homozygous for the *IVS2+691G>C SNV (GG)* and 165 subjects (84.2%; 95% CI: 79-89) were heterozygous (GC) or homozygous (CC). The MAF for *IVS2+691G>C* SNVs was hence 59%. Of note that, and as per study design, none of the 11 subjects who were excluded from final analysis because of failed analgesia had blood sent for genotyping. We were hence unable to compare and contrast their genotyping profile. Nevertheless, there were no significant demographic differences between these excluded subjects and the analyzed cohort.

Maternal characteristics and delivery outcome data were comparable between all groups except for gestational age that was significantly lower for *IVS2+31G>A* wild type genotype when compared to allele carriers (Median [IQR]: 39.0 [1.0] *vs*. 40.0 [1.0]; P<0.05]) (Table I). There were no statistically significant differences in duration of epidural fentanyl analgesia (161±68 *vs.* 143±51 min, P=0.08 for wild type and allele carriers of the *118A>G SNV* respectively; 157±65 *vs.* 170±77 min, P=0.62 for wild type and allele carriers of the *OPRM1 IVS2+31G>A* respectively; 174±92 *vs.* 156±60 min, P=0.28 for wild type and allele carriers of the *OPRM1 IVS2+691G>C respectively* (Table II; Figure 2, 3, 4). Also, there were no statistically significant differences in VAS scores, side effects, and satisfaction scores according to genotype for each of the three SNVs (Table II).

TABLE I.—*Maternal baseline variables.*

Polymorphism (rs#)	*OPRM1 118A>G (1799971)*			*OPRM1 IVS2+31G>A (9479757)*			*OPRM1 IVS2+691G>C (2075572)*		
Genotype	AA N.=174	AG & GG N.=46	P value	GG N.=178	GA N.=18	P value	GG N.=31	GC & CC N.=165	P value
Age (years)	27 (6)	27 (5)	0.99	27 (6)	28 (3)	0.74	29 (7)	27 (6)	0.13
Body Mass Index (kg/m^2)	29 (5)	29 (4)	0.59	29 (5)	30 (5)	0.19	29 (5)	29 (5)	0.91
Gestational age (weeks)	40 (1)	39 (1)	0.79	39 (1)	40 (1)	0.04	39 (1)	40 (1)	0.73
Cervical dilation at baseline (cm)	2.5 (2.5)	3.0 (2.0)	0.09	3.0 (2.0)	2.0 (2.2)	0.14	3.0 (2.0)	3.0 (2.9)	0.71

Data are median (interquartile range).

TABLE II.—*Maternal (obstetrical, analgesia and side effects) and neonatal outcome variables.*

Polymorphism (rs#)	*OPRM1 118A>G (1799971)*			*OPRM1 IVS2+31G>A (9479757)*			*OPRM1 IVS2+691G>C (2075572)*		
	AA N.=174	AG & GG N.=46	P value	GG N.=178	GA N.=18	P value	GG N.=31	GC & CC N.=165	P value
Cervical dilation at 2nd request (cm)	4.0 (3.0)	4.5 (2.2)	0.42	4.0 (3.0)	4.0 (4.0)	0.32	5.0 (2.0)	4.0 (3.0)	0.15
Maximum oxytocin infusion (mIU/min)	10 (13)	13 (17)	0.48	11 (13)	15 (12)	0.43	10 (14)	11 (13)	0.69
Duration of epidural fentanyl analgesia (min)	161±68	143±51	0.08	157±65	170±77	0.62	174±92	156±60	0.28
Time from fentanyl administration to complete cervical dilation (min)	378±203	402±246	0.56	379±221	375±173	0.88	296±109	391±228	0.15
Time from fentanyl administration to delivery (min)	481±196	513±242	0.42	482±211	471±218	0.72	394±110	496±220	0.06
Mode of delivery (%)			0.34			0.06			0.56
Spontaneous vaginal	109 (63.0)	34 (73.9)		116 (65.5)	9 (50.0)		18 (58.1)	107 (65.2)	
Instrumental vaginal	4 (2.3)	1 (2.2)		3 (1.7)	2 (11.1)		1 (3.2)	4 (2.4)	
Cesarean section	60 (34.7)	11 (23.9)		58 (32.8)	7 (38.9)		12 (38.7)	53 (32.3)	
Neonatal weight (g)	3348 (589)	3263 (565)	0.35	3335 (585)	3428 (521)	0.22	3345 (585)	3345 (570)	0.92
Apgar score at 1 min	9 (0)	9 (1)	0.13	9 (0)	9 (1)	0.28	9 (0)	9 (1)	0.94
VAS (cm)									
At 1st analgesia request	6.0 (2.0)	6.0 (3.0)	0.23	6.0 (2.0)	6.0 (4.0)	0.65	6.0 (3.0)	6.0 (2.0)	0.70
10 min after epidural fentanyl	0.0 (1.0)	0.0 (2.0)	0.51	0.0 (1.0)	0.0 (2.0)	0.47	0.0 (1.0)	0.0 (1.0)	0.97
20 min after epidural fentanyl	0.0 (0.0)	0.0 (0.0)	0.42	0.0 (0.0)	0.0 (0.0)	0.86	0.0 (2.0)	0.0 (0.0)	0.13
30 min after epidural fentanyl	0.0 (0.0)	0.0 (0.0)	0.19	0.0 (0.0)	0.0 (0.0)	0.23	0.0 (0.0)	0.0 (0.0)	0.06
At 2nd analgesia request	5.0 (3.0)	5.0 (1.0)	0.99	5.0 (2.0)	5.0 (2.0)	0.56	5.0 (2.0)	5.0 (2.0)	0.90
Severity of nausea (%)			0.23			0.22			0.74
None	151 (86.8)	39 (84.8)		156 (87.6)	14 (77.8)		28 (90.3)	142 (86.1)	
Mild	19 (10.9)	4 (8.7)		16 (9.0)	3 (16.7)		2 (6.4)	17 (10.3)	
Moderate	3 (1.7)	1 (2.2)		4 (2.2)	0 (0.0)		1 (3.2)	3 (1.8)	
Severe	1 (0.6)	2 (4.3)		2 (1.1)	1 (5.6)		0 (0.0)	3 (1.8)	
Incidence of vomiting (%)	3 (1.7)	3 (6.5)	0.11	5 (2.8)	1 (5.6)	0.44	1 (3.2)	5 (3.0)	1.00
Severity of pruritus (%)			0.46			0.92			0.30
None	93 (53.4)	21 (45.6)		99 (55.6)	10 (55.6)		21 (67.7)	88 (53.3)	
Mild	43 (24.7)	17 (37.0)		41 (23.0)	5 (27.8)		5 (16.1)	41 (24.8)	
Moderate	20 (11.5)	4 (8.7)		20 (11.2)	1 (5.6)		4 (12.9)	17 (10.3)	
Severe	18 (10.3)	4 (8.7)		18 (10.1)	2 (11.1)		1 (3.2)	19 (11.5)	
Satisfaction with analgesia (cm)	8.0 (2.0)	8.0 (3.0)	0.72	8.0 (2.0)	8.0 (3.0)	0.40	8.0 (3.0)	8.0 (2.0)	0.65

Data are median (interquartile range), mean (±SD) or numbers (percentage).
VAS: Visual Analogue Score.

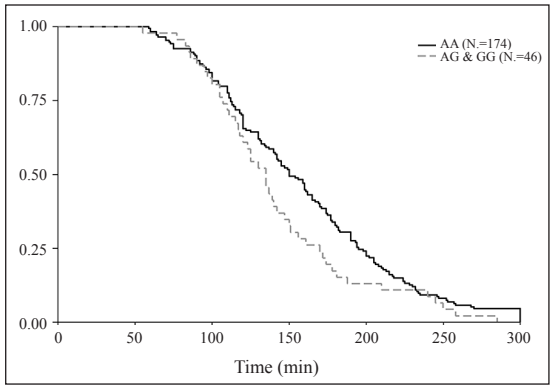

Figure 2.—Kaplan-Meier survival curves of duration of epidural fentanyl analgesia by *OPRM1 118A>G* genotype in the original cohort (N.=220) (P=0.06 by log-rank test).

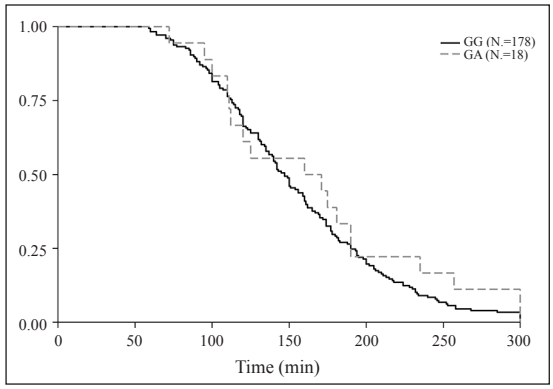

Figure 3.—Kaplan-Meier survival curves of duration of epidural fentanyl analgesia by *OPRM1 IVS2+31G>A* genotype in the sub-cohort (N.=196) (P=0.31 by log-rank test).

Discussion

This is the first pharmacogenetic study to evaluate the frequency of three *OPRM1* SNVs in a Lebanese cohort of nulliparous women, and to find no association between *OPRM1 118A>G*, *IVS2+31G>A,* and *IVS2+691G>C* polymorphisms and duration of epidural fentanyl analgesia. In addition, we identified that the genetic distribution for these three SNVs is similar to previous reports among Caucasian women,[6] but that of *OPRM1 118A>G* is slightly lower than participants from Jerusalem (MAF=16% though with overlapping CI).[11]

Previous literature showed that the *OPRM1 118A>G* SNV was associated with decreased receptor mRNA and protein expression in brain autopsy tissue.[9, 24] Also, early studies on acute pain models suggest a decreased opioid potency when administered systemically in patients carrying the *OPRM1 118A>G* allele.[15, 20, 25-33] In addition, clinical studies of systemic opioids in acute postoperative and chronic settings showed a trend towards increased systemic morphine requirements in patients carrying at least one variant allele, findings that are not in accordance with the results of our study with epidural fentanyl.[34] Thus, the reduced response to intravenous opioid such as morphine in the presence of variant allele may not exist in response to opioids administered neuraxially.

Neuraxial opioid bioavailability is an important component of analgesic duration. Due to its high lipophilicity, fentanyl injected into the epidural space is sequestered in the epidural fat, from where it rapidly returns to the systemic circulation via epidural veins. This rapid elimination may explain the lack of difference in drug response due to μ-opioid polymorphism, if it exists.[35]

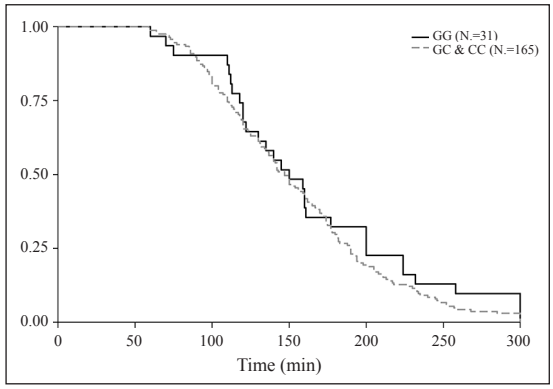

Figure 4.—Kaplan-Meier survival curves of duration of epidural fentanyl analgesia by *OPRM1 IVS2+691G>C* genotype in the sub-cohort (N.=196) (P=0.28 by log-rank test).

Our results are in agreement with previous findings in the obstetric anesthesia literature. In fact, four pharmacogenetic studies assessed the role of *OPRM1 118A>G* in neuraxial opioid labor analgesia. Ginosar *et al.*[11] studied two ethnically distinct hospital populations (Miami and Jerusalem) and found no significant effect for the *OPRM1 118A>G* SNV on analgesia duration after spinal fentanyl for labor pain. Similarly, in a North American cohort, Wong *et al.*[10] showed that the *OPRM1 118A>G* polymorphism was not

significantly associated with neither duration of intrathecal fentanyl analgesia in laboring women nor requirement for supplemental analgesia or duration of analgesia after intrathecal morphine following cesarean delivery.

As for the potency of neuraxially-administered opioids, in an Italian population, Camorcia *et al.*[12] showed that women carrying the variant *A118G* allele had a lower ED50 for epidural sufentanyl given for early labor analgesia than women homozygous for the wild type allele. Also in a Swiss cohort, Landau *et al.*[13] showed that the *A118G* allele reduces intrathecal fentanyl ED50 for labor analgesia. Additionally, parturients with the *A118G* allele requested analgesia at greater cervical dilation than those without the variant allele suggesting greater tolerance to pain. This increased analgesic effect in women carrying the variant allele was explained by a higher receptor-binding affinity of *OPRM 1* in response to neuraxial opioids.

In one study, the binding affinity and potency of β-endorphin was increased approximately three-fold in *OPRM1 118A>G* polymorphic receptors with the variant allele compared to wild type receptors. In contrast, other investigators found no difference in receptor binding of β-endorphin between mutant and wild-type receptors.[36] The *in-vitro* studies tend to suggest that the *118A>G* genetic polymorphism is more likely to affect μ-opioid function via alterations in expression, transduction systems or receptor trafficking rather than via altered receptor binding affinity.[37]

Administration of opioids at doses larger than the ED95 may mask differences in analgesic duration in the presence of a genetic polymorphism.[10] In our study, the dose of epidural fentanyl used was less than the ED95 [140 (126-153) μg],[38] but well within the ranges used elsewhere, yet no difference in the duration of analgesia was observed between subjects carrying the variant allele and subjects homozygous for the wild type allele.

It is worthy to mention that there was no difference in pain scores measured with assessment of VAS and cervical dilation at first request for analgesia for all three SNVs. In addition, and in contrast to previous reports on systemic opioids use,[6, 7] the incidence of side effects, mainly pruritus, nausea and vomiting, was comparable between the genotype groups. Moreover, it is not known why carriers of the *OPRM1 IVS2+31G>A* SNV had significantly higher gestational age. In fact, all patients were recruited at gestational age ≥37 weeks. We believe that the result is incidental and not clinically relevant especially that all other maternal and neonatal factors such as cervical dilation and neonatal weight were similar.

Limitations of the study

Despite the advantage of being conducted on a homogenous population from both cultural and ethnic stand point, this study does have limitations in being restricted to early labor and three SNVs in one candidate gene only (*OPRM1*), and the failure of adjusting for some potentially confounding materno-fetal and obstetric management factors. In addition, the study is not adequately powered to show significant differences in the secondary end points such as side effects with the three SNVs, and to draw any conclusions on the effect of *IVS2+691G>C* SNV and the rarer *IVS2+31G>A* SNV on epidural fentanyl analgesia.

As for generalizability, results are not applicable to men and non-pregnant women since pregnancy is known to induce antinociception via hormonal changes or other mechanisms modifying the opioid system.[39] Linkage disequilibrium should be kept in mind between *OPRM1 118A>G* alleles and other functional variants in addition to the possible polygenic nature of this pharmacogenetic effect to include the role of drug metabolizing enzymes such as CYP3A4. Of note, the high cesarean rate in our study suggests that many labors were probably dystocia.

Conclusions

In conclusion, *OPRM1 118A>G, IVS2+31G>A,* and *IVS2+691G>C* polymorphisms did not affect the duration of epidural fentanyl administered for early labor analgesia in nulliparous women of the Middle Eastern region. These results should be confirmed in patients receiving epidural opioids in other clinical settings.

What is known

• A number of pharmacogenetic studies revealed conflicting results concerning the potential role of *OPRM1* genetic polymorphisms in the inter-individual variability of the response to opioid analgesia.

What is new

• *OPRM1 118A>G, IVS2+31G>A,* and *IVS2+691G>C* polymorphisms did not affect the duration of epidural fentanyl administered for early labor analgesia in nulliparous women of the Middle Eastern region.

• No significant differences were shown with the visual analog scale scores, side effects, and satisfaction for each of the three polymorphisms.

• It is probably not warranted to pre-emptively genotype for these three polymorphisms in nulliparous women receiving epidural fentanyl administered for early labor analgesia

References

1. Carrie LE, O'Sullivan GM, Seegobin R. Epidural fentanyl in labour. Anaesthesia 1981;36:965–9.

2. Justins DM, Francis D, Houlton PG, Reynolds F. A controlled trial of extradural fentanyl in labour. Br J Anaesth 1982;54:409–14.

3. Connelly NR, Parker RK, Lucas T, El-Mansouri M, Komanduri V, Nayak P, *et al.* The influence of a bupivacaine and fentanyl epidural infusion after epidural fentanyl in patients allowed to ambulate in early labor. Anesth Analg 2001;93:1001–5.

4. Connelly NR, Parker RK, Pedersen T, Manikantan T, Lucas T, Serban S, *et al.* Diluent volume for epidural fentanyl and its effect on analgesia in early labor. Anesth Analg 2003;96:1799–804.

5. Wong CA. The promise of pharmacogenetics in labor analgesia…tantalizing, but not there yet. Int J Obstet Anesth 2012;21:105–8.

6. Zahari Z, Ismail R. Impact of opioid receptor Mu 1 (OPRM1) polymorphisms on pain sensitivity and clinical response to opioid analgesic therapy. Curr Pharmacogenomics Person Med 2013;11:59–75.

7. Walter C, Lötsch J. Meta-analysis of the relevance of the OPRM1 118A>G genetic variant for pain treatment. Pain 2009;146:270–5.

8. Song Z, Du B, Wang K, Shi X. Effects of OPRM1 A118G polymorphism on epidural analgesia with fentanyl during labor: a meta-analysis. Genet Test Mol Biomarkers 2013;17:743–9.

9. Huang P, Chen C, Mague SD, Blendy JA, Liu-Chen LY. A common single nucleotide polymorphism A118G of the μ opioid receptor alters its N-glycosylation and protein stability. Biochem J 2012;441:379–86.

10. Wong CA, McCarthy RJ, Blouin J, Landau R. Observational study of the effect of mu-opioid receptor genetic polymorphism on intrathecal opioid labor analgesia and post-cesarean delivery analgesia. Int J Obstet Anesth 2010;19:246–53.

11. Ginosar Y, Birnbach DJ, Shirov TT, Arheart K, Caraco Y, Davidson EM. Duration of analgesia and pruritus following intrathecal fentanyl for labour analgesia: no significant effect of A118G μ-opioid receptor polymorphism, but a marked effect of ethnically distinct hospital populations. Br J Anaesth 2013;111:433–44.

12. Camorcia M, Capogna G, Stirparo S, Berritta C, Blouin JL, Landau R. Effect of μ-opioid receptor A118G polymorphism on the ED50 of epidural sufentanil for labor analgesia. Int J Obstet Anesth 2012;21:40–4.

13. Landau R, Kern C, Columb MO, Smiley RM, Blouin JL. Genetic variability of the mu-opioid receptor influences intrathecal fentanyl analgesia requirements in laboring women. Pain 2008;139:5–14.

14. Klepstad P, Rakvåg TT, Kaasa S, Holthe M, Dale O, Borchgrevink PC, *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 2004;48:1232–9.

15. Hayashida M, Nagashima M, Satoh Y, Katoh R, Tagami M, Ide S, *et al.* Analgesic requirements after major abdominal surgery are associated with OPRM1 gene polymorphism genotype and haplotype. Pharmacogenomics 2008;9:1605–16.

16. Ross JR, Rutter D, Welsh K, Joel SP, Goller K, Wells AU, *et al.* Clinical response to morphine in cancer patients and genetic variation in candidate genes. Pharmacogenomics J 2005;5:324–36.

17. Hoehe MR, Köpke K, Wendel B, Rohde K, Flachmeier C, Kidd KK, *et al.* Sequence variability and candidate gene analysis in complex disease: association of mu opioid receptor gene variation with substance dependence. Hum Mol Genet 2000;9:2895–908.

18. Lötsch J, Geisslinger G. Relevance of frequent mu-opioid receptor polymorphisms for opioid activity in healthy volunteers. Pharmacogenomics J 2006;6:200–10.

19. Xin L, Wang ZJ. Bioinformatic analysis of the human mu opioid receptor (OPRM1) splice and polymorphic variants. AAPS PharmSci 2002;4:E23.

20. Fukuda K, Hayashida M, Ikeda K, Koukita Y, Ichinohe T, Kaneko Y. Diversity of opioid requirements for postoperative pain control following oral surgery—is it affected by polymorphism of the μ-opioid receptor? Anesth Prog 2010;57:145–9.

21. Hastie BA, Riley JL 3rd, Kaplan L, Herrera DG, Campbell CM, Virtusio K, *et al.* Ethnicity interacts with the OPRM1 gene in experimental pain sensitivity. Pain 2012;153:1610–9.

22. Hernandez-Avila CA, Covault J, Wand G, Zhang H, Gelernter J, Kranzler HR. Population-specific effects of the Asn40Asp polymorphism at the mu-opioid receptor gene (OPRM1) on HPA-axis activation. Pharmacogenet Genomics 2007;17:1031–8.

23. Gelernter J, Kranzler H, Cubells J. Genetics of two mu opioid receptor gene (OPRM1) exon I polymorphisms: population studies, and allele frequencies in alcohol- and drug-dependent subjects. Mol Psychiatry 1999;4:476–83.

24. Zhang Y, Wang D, Johnson AD, Papp AC, Sadée W. Allelic expression imbalance of human mu opioid receptor (OPRM1) caused by variant A118G. J Biol Chem 2005;280:32618–24.

25. 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 2006;105:334–7.

26. Sia AT, Lim Y, Lim EC, Goh RW, Law HY, Landau R, *et al.* A118G single nucleotide polymorphism of human mu-opioid receptor gene influences pain perception and patient-controlled intravenous morphine consumption after intrathecal morphine for postcesarean analgesia. Anesthesiology 2008;109:520–6.

27. Sia AT, Lim Y, Lim EC, Ocampo CE, Lim WY, Cheong P, *et al.* Influence of mu-opioid receptor variant on morphine use and self-rated pain following abdominal hysterectomy. J Pain 2013;14:1045–52.

28. Wu WD, Wang Y, Fang YM, Zhou HY. Polymorphism of the micro-opioid receptor gene (OPRM1 118A>G) affects fentanyl-induced analgesia during anesthesia and recovery. Mol Diagn Ther 2009;13:331–7.

29. Zhang W, Chang YZ, Kan QC, Zhang LR, Lu H, Chu QJ, *et al.* Association of human micro-opioid receptor gene polymorphism A118G with fentanyl analgesia consumption in Chinese gynaecological patients. Anaesthesia 2010;65:130–5.

30. Zhang W, Yuan JJ, Kan QC, Zhang LR, Chang YZ, Wang ZY. Study of the OPRM1 A118G genetic polymorphism associated with postoperative nausea and vomiting induced by fentanyl intravenous analgesia. Minerva Anestesiol 2011;77:33–9.

31. Chou WY, Yang LC, Lu HF, Ko JY, Wang CH, Lin SH, *et al.* Association of mu-opioid receptor gene polymorphism (A118G) with variations in morphine consumption for analgesia after total knee arthroplasty. Acta Anaesthesiol Scand 2006;50:787–92.

32. Tan EC, Lim EC, Teo YY, Lim Y, Law HY, Sia AT. Ethnicity and OPRM variant independently predict pain perception and patient-controlled analgesia usage for post-operative pain. Mol Pain 2009;5:32.

33. Henker RA, Lewis A, Dai F, Lariviere WR, Meng L, Gruen GS, *et al.* The associations between OPRM 1 and COMT genotypes and postoperative pain, opioid use, and opioid-induced sedation. Biol Res Nurs 2013;15:309–17.

34. Landau R. Pharmacogenetic influences in obstetric anaesthesia. Best Pract Res Clin Obstet Gynaecol 2010;24:277–87.

35. Ummenhofer WC, Arends RH, Shen DD, Bernards CM. Comparative spinal distribution and clearance kinetics of intrathecally administered morphine, fentanyl, alfentanil, and sufentanil. Anesthesiology 2000;92:739–53.

36. Bond C, LaForge KS, Tian M, Melia D, Zhang S, Borg L, *et al.* Single-nucleotide polymorphism in the human mu opioid receptor gene alters beta-endorphin binding and activity: possible implications for opiate addiction. Proc Natl Acad Sci USA 1998;95:9608–13.

37. Margas W, Zubkoff I, Schuler HG, Janicki PK, Ruiz-Velasco V. Modulation of Ca2+ channels by heterologously expressed wild-type and mutant human micro-opioid receptors (hMORs) containing the A118G single-nucleotide polymorphism. J Neurophysiol 2007;97:1058–67.

38. Capogna G, Camorcia M, Columb MO. Minimum analgesic doses of fentanyl and sufentanil for epidural analgesia in the first stage of labor. Anesth Analg 2003;96:1178–82.

39. Hernandez-Avila CA, Wand G, Luo X, Gelernter J, Kranzler HR. Association between the cortisol response to opioid blockade and the Asn40Asp polymorphism at the mu-opioid receptor locus (OPRM1). Am J Med Genet B Neuropsychiatr Genet 2003;118B:60–5.

Conflicts of interest.—The authors certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

Funding.—This study was funded by the American University of Beirut Faculty of Medicine (AUBFM) Medical Practice Plan (MPP) grant to Dr. Sahar M. Siddik-Sayyid.

Authors' contributions.—Nathalie K. Zgheib and Sahar M. Siddik-Sayyid designed the study and analyzed data together with Maha H. Makki; Sahar M. Siddik-Sayyid collected the clinical data; Nathalie K. Zgheib supervised the genotyping experiments in her lab; Nathalie K. Zgheib and Sahar M. Siddik-Sayyid wrote the first draft of the manuscript; Marie T. Aouad, Samar K. Taha, Anwar H. Nassar, Riwa F. Masri and Mira Y. Khoury were involved in clinical data collection and critically revised the manuscript for important intellectual content.

Article first published online: May 14, 2018. - Manuscript accepted: May 9, 2018. - Manuscript revised: April 11, 2018. - Manuscript received: January 4, 2018.