ORIGINAL ARTICLE

Genetic Contribution in Low Back Pain: A Prospective Genetic Association Study

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■ **Abstract**

Objectives: Chronic pain is one of the most common reasons individuals seek medical attention. It is a major issue because of the wide interindividual variability in the analgesic response. This might be partly explained by the presence of variants in genes encoding molecules involved in pharmacodynamics and pharmacokinetics. The aim was to analyze opioid effectiveness in chronic low back pain (CLBP) relief after opioid titration, unveiling the impact of pharmacogenetics.

Methods: The study included 231 opioid-naïve patients from the Spine Unit; age 63 ± 14 years, 64% female, body mass index 29 ± 6 kg/m^2, visual analog scale pain intensity score 73 ± 16 mm. Clinical data were collected at baseline, 3 months after opioid titration, and after 2 to 4 years of follow-up concerning pain (intensity and relief), quality of life, disability, comorbidities, and drug prescription (opioid dose, rotations, and adverse events). The genotype influence of *OPRM1*, *COMT*, *UGT2B7*, *ABCB1*, *KCNJ6*, and *CYP3A5*3A* in analgesic response was analyzed by reverse-transcription polymerase chain reaction genotyping.

Results: Patients with the *COMT* G472A-AA genotype (rs4680) and *KCNJ6* A1032G-A allele (rs2070995) CLBP responded differently to opioid titration, with higher pain intensity requiring higher dosing. Furthermore, GG- genotypes of A118G (*OPRM1*, rs1799971) and A854G (*UGT2B7*, rs776746) influenced the neuropathic component. After opioid titration, CLBP intensity, neuropathic component, low back pain disability, anxiety, and depression significantly decreased, while quality of life improved.

Conclusion: Single-nucleotide polymorphisms in genes involved in pain transmission and opioid metabolism might predispose to exaggerated sensitivity and differences in the opioid analgesic effect in patients with CLBP. We encourage clinical trials for their clinical application in chronic pain management. ■

Key Words: opioids, pharmacogenetics, chronic low back pain, *COMT*, *KCNJ6*, *OPRM1*, *UGT2B7*

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INTRODUCTION

Chronic low back pain (CLBP) is a heterogeneous condition defined as back pain persisting[1] or resulting in limitation of usual activity.[2] Clinical care and research are complicated by the lack of standardized criteria for its diagnosis and the high rate of imaging studies that are completely normal. The options for nonsurgical management include opioid administration.[3] However, this is not always effective and it can be time consuming to attain the right medication and dose for a particular patient and, occasionally, hazardous for certain patients with the potential for serious damage.[4–7] These problems are especially marked in the long-term use of opioids and neuromodulators for nononcological pain,[8,9] with prescriptions rising steadily but with no clear rationale. Future studies are needed to provide valuable evidence to help in decision making regarding drug use for the treatment of CLBP.

Specific genetic traits could alter the mechanisms through which the drug affects physiology and the way it is distributed and handled in the patient's body. Pharmacogenetics has the potential to improve pain management by predicting the individual response to a specific analgesic drug before therapy initiation and, therefore, to streamline the way physicians prescribe medications to the individual. For instance, a noninvasive saliva test might one day allow clinicians to determine if a particular medication would be effective or have adverse effects, providing guidance on individualized dosages.[10]

Several genes have been involved in pain modulation after opioid treatment. Variants in the *KCNJ6* gene (A1032G and G1250A) that encodes for components of the GIRK potassium channel or in the catechol-O-methyl transferase (*COMT*) enzyme gene, responsible for the breakdown of biologically active catecholamines, such as dopamine, noradrenaline, and adrenaline, are involved in numerous physiological processes, including pain modulation.[11] In addition, the mu opioid receptor, encoded by the opioid receptor μ1 (*OPRM1*) gene, is the primary site of action for the most commonly used opioids. Therefore, it is a first-line candidate for evaluating the role of single-nucleotide polymorphisms (SNPs). Subjects carrying the variant A118G-G allele were found to have a reduced response to morphine and fentanyl treatment[12–14] and morphine-6-glucuronide,[11,14] requiring higher doses for pain relief.[15] Other gene variants like UDP-glucuronosyltransferase (*UGT2B7*) for morphine[11] and *CYP3A5*3A* for fentanyl are associated with higher promoter activity, increasing enzyme levels and metabolic drug rates.[15] The *ABCB1* gene encodes for a P-glycoprotein transporter, the inhibition of which impacts opiate-induced analgesia, especially for morphine and oxycodone, and is a major determinant of the pharmacokinetics and pharmacodynamics of several drugs.[16–18] However, use of pharmacogenetic information has been slow, due in large part to the lack of robust evidence demonstrating clinical utility.[19]

The primary goal of this study was to determine whether analysis of polymorphisms located in genes involved in opioid metabolism and analgesia could predict pain relief and influence patient response after opioid titration and in chronic use.

METHODS

Ethics

A prospective study was conducted with ambulatory patients at the Pain Unit of the Department of Health of Alicante General Hospital, Spain. Recruitment was carried out over 22 months from February 2011 to December 2013, with a follow-up of 2 to 4 years. The study was approved by the Ethics Committee of Alicante General Hospital and carried out in accordance with the Helsinki Declaration principles. All patients were following standard treatment and received information on the design and purpose of the study. Institutional review board approval and informed consent from the participants were obtained to record clinical data and genetic samples.

Patients

The study comprised 231 opioid-naïve patients with CLBP. Patients were referred to the Pain Unit from the Spine Unit for severe CLBP secondary to lumbar canal stenosis, under assessment for surgery and requiring opioid prescription.

Inclusion criteria were as follows: (1) adult (≥18 years); (2) naïve to opioid therapy (regular prescription for 3 months or longer); (3) diagnosis of severe CLBP; and (4) possessing adequate mental status to properly complete the scales and questionnaires. The exclusion criteria were lost to follow-up, patient's decision, and requirement for canal stenosis surgery, cessation of allocated medication, or non-CLBP. For the purposes of this study, participants were classified using

the ICD-9 or ICD-10 diagnostic codes according to the etiology of the pain.

Patients with neuropathic pain caused by damage to the somatosensory system[20] or nociceptive pain caused by damage to non-neural tissue or stimuli that could lead to tissue damage[21] were excluded. The diagnosis of neuropathic pain included conditions such as trigeminal neuralgia, mononeuritis, and multiple sclerosis. The diagnosis of nociceptive pain included conditions such as osteoarthritis, myalgia, myositis, carpal tunnel syndrome, and rheumatism. Subjects diagnosed with mixed pain (eg, migraine, headache, cervicalgia, nontraumatic compartment syndrome) or other conditions that may or may not be pain related (eg, restless legs syndrome, cerebrovascular disease, paraplegia) were also not included in the study.

Medical Records and Opioid Titration

All the patients were interviewed at the first visit to evaluate physical health and medical history. The following information was collected from the hospital records for each patient: demographic parameters (age, gender, ethnicity, body weight, and height), and follow-up time.

Validated scales and questionnaires were completed at each visit. All were self-administered and supported by the presence of an expert clinician. Pain intensity and relief were determined using the validated 100-mm visual analog scale (VAS; 0 = "no pain/relief" to 100 = "worst possible pain/maximum relief"). Quality of life related to health measures was assessed by the EuroQol VAS (EQ-VAS; 0 = "worst" to 100 = "best health status"). Pain intensity was determined using a Likert-based scale (descriptors: none, mild, moderate, severe, and extreme pain intensity/relief). The Hospital Anxiety and Depression Scale (HADS) was used to assess both anxiety and depression by 7 questions and scores that are categorized as normal (0 to 7), mild (8 to 10), moderate (11 to 14), or severe (15 to 21). The Oswestry Disability Index (ODI) was used to quantify disability for low back pain with 6 statements describing different potential scenarios in the patient's life. Scores are summed, then multiplied by 2 to obtain the index (%; 0 = "no disability" to 100 = "maximum disability possible" [0% to 20%: minimum functional limitation; 20% to 40%: moderate; 40% to 60%: intense; 60% to 80%: disability; above 80%: maximum functional limitation]). All questionnaires were self-administered but supported by the presence of an expert clinician.

The patients included in the study were not under regular opioid prescription at the start of the study. Upon inclusion, at the baseline visit, prescription of opioids was initiated with oral morphine or transdermal fentanyl, through a 3-month period of titration, according to the degree of patient compliance and their preferences (oral or transdermal).[22,23] Prescription was performed by 4 anesthesiologists and re-evaluated by 2 members of the research team. Rescue medication consisted of tramadol or fast-release opioids according to the intensity and frequency of pain episodes.

The optimal opioid dose was reached with a balance of 3 factors: (1) effectiveness: improved function or at least 30% reduction in pain intensity; (2) plateauing: effectiveness plateau (increasing the dose yields negligible benefit); and (3) adverse events (AEs)/complications that are manageable. With each dose increase, the patient was asked to estimate the pain intensity: a desirable response was a reduction in pain intensity (eg, VAS intensity scores from 9/10 [baseline] to 6/10 [endpoint]) and a longer duration of analgesia per dose.[24]

A follow-up visit at the Pain Unit was made 2 to 4 years after opioid titration to evaluate pain intensity (VAS), drug prescription, and AEs related to pain treatment for all the study patients.

Pharmacological Therapy and Drug Adverse Events

Physicians recorded the patients' prescribed pain therapy (opioids [dose, number of dose changes, and number of opioid rotations] and concomitant drugs [antiepileptic, antipsychotic, anxiolytic, antidepressant, or muscle relaxant]), polymedication for pain therapy (defined as $\geq$5 drugs prescribed in relation to pain), and type of pain. The total daily dose of opioids was converted to the morphine equivalent daily dose (MEDD), estimated using the equianalgesic dose.[25] Neuromodulator drugs, such as gabapentinoids (pregabalin, gabapentin) or duloxetine, could be prescribed regularly together with opioids if a neuropathic component existed.

A questionnaire with a list of the most frequent AEs selected according to the summary of the opioid product characteristics having a frequency of "very common" or "common" and a blank field to add any other AE was developed by our group to record all AEs reported by the patients.[26]

Genetic Analyses

DNA Collection and Extraction. Blood samples were collected in EDTA tubes, and DNA was isolated using the QIAamp DNA Blood Mini Kit (Qiagen, Hilden,

Germany) following the manufacturer's instructions. DNA yield was quantified with a NanoDrop spectrophotometer (Thermo Fisher Scientific, Wilmington, DE, U.S.A.). DNA was stored at −20°C until use.

Genotyping. TaqMan technology (Thermo Fisher Scientific, Pleasanton, CA, U.S.A.) was used to detect SNP variant alleles. Genotyping was carried out with a Real Time PCR Rotor Gene Q (Qiagen) system. Polymerase chain reaction (PCR) was performed in a final volume of 20 μL containing 100 ng DNA, 10 μL TaqMan Genotyping 2× Master Mix (Thermo Fisher Scientific), and 1 μL specific 20× TaqMan SNP Genotyping Assay (Thermo Fisher Scientific). All reactions were performed in duplicate, and negative controls were included in each PCR procedure. The PCR program was: 95°C for 10 minutes, followed by 40 cycles of 92°C for 15 seconds and 60°C for 1 minute.

A total of 8 SNPs located in 6 genes relevant in opioid metabolism were genotyped. The following SNPs were analyzed: A118G (rs1799971) of *OPRM1*, G472A (rs4680) of *COMT*, C3435T (rs1045642) of *ABCB1*, G211T (rs12233719) and A842G (rs7438135) of *UGT2B7*, A6986G (rs776746) of *CYP3A5*3*, and A1032G (rs2070995) and G1250A (rs6517442) of *KCNJ6*.

Statistics

The Shapiro-Wilk normality test was performed to choose a parametric or nonparametric test for comparisons. Quantitative data are presented as mean ± standard deviation (SD), and categorical variables are expressed by absolute counts and/or percentages. Relative frequencies of genotypes and alleles were calculated for each group. Observed gene frequencies were compared with expected frequencies using the chi-square goodness-of-fit test and the Hardy-Weinberg proportion. Calculation of the expected gene frequencies from respective single-allele frequencies was made according to the Hardy-Weinberg equation. In all cases, multiple testing was adjusted using the Bonferroni correction method.

For quantitative data, the *t*-test for independent samples or Mann-Whitney *U* test were used to assess differences between 2 groups; effect sizes (Cohen's *d*) and 95% confidence intervals (CIs) are also reported. Comparisons among 3 or more groups were performed with one-way analysis of variance or the Kruskal-Wallis test; effect sizes (eta squared [η^2]) are reported.

For qualitative data, the chi-square or Fisher's exact test, with Yates's continuity correction, was performed. Effect sizes for all comparisons are reported (Cramer's *V* or odds ratio [OR], respectively, together with their 95% CIs).

Analyses were carried out with GraphPad Prism version 5.02 (GraphPad Software, La Jolla, CA, U.S.A.) and R version 3.2.4 programs. $P < 0.05$ was considered statistically significant.

RESULTS

Patient Demographic and Pharmacological Data

A total of 450 candidates for opioid prescription due to CLBP were referred from the Spinal Unit with the diagnosis of lumbar canal stenosis established by the orthopedic surgeons. Patients were treated routinely at the Pain Unit in an ambulatory setting. A total of 241 patients were informed and their participation was requested in the present study, of whom 231 (51%) signed the informed consent. Over the study period, a total of 30 patients dropped out or were lost to follow-up, mainly from AEs, lack of opioid effectiveness, lumbar surgery, or death. A total of 181 subjects (63%) attended the final visit and 145 (63%) the follow-up control (Figure 1).

A summary of the demographic and pharmacological data is presented in Table 1. CLBP was more common in women 50 to 75 years of age (63% female, 63 ± 14 years, body mass index 29 ± 6 kg/m^2), with a follow-up of 2 to 3 years. The most frequent diagnosis was lumbar canal stenosis (93%), 17% of whom had a radicular component. Before titration, patients were receiving nonsteroidal anti-inflammatory drugs (9%), acetaminophen (30%), combination tramadol/acetaminophen (46%), or opioids (nonregularly prescribed) (17%; 20% morphine, 8% oxycodone). Regarding concomitant medications, 30% were receiving muscle relaxants, 15% benzodiazepines, and 25% topical treatment.

Medical Records and Opioid Titration

A summary of the medical records of the subjects included is presented in Table 2. In our CLBP population, pain intensity was mostly moderate to severe (VAS score 74 ± 17 mm) with mild anxiety (score of 8 ± 5) and depression (score of 8 ± 5); 50% had a likely or unclear neuropathic mixed component. The VAS quality

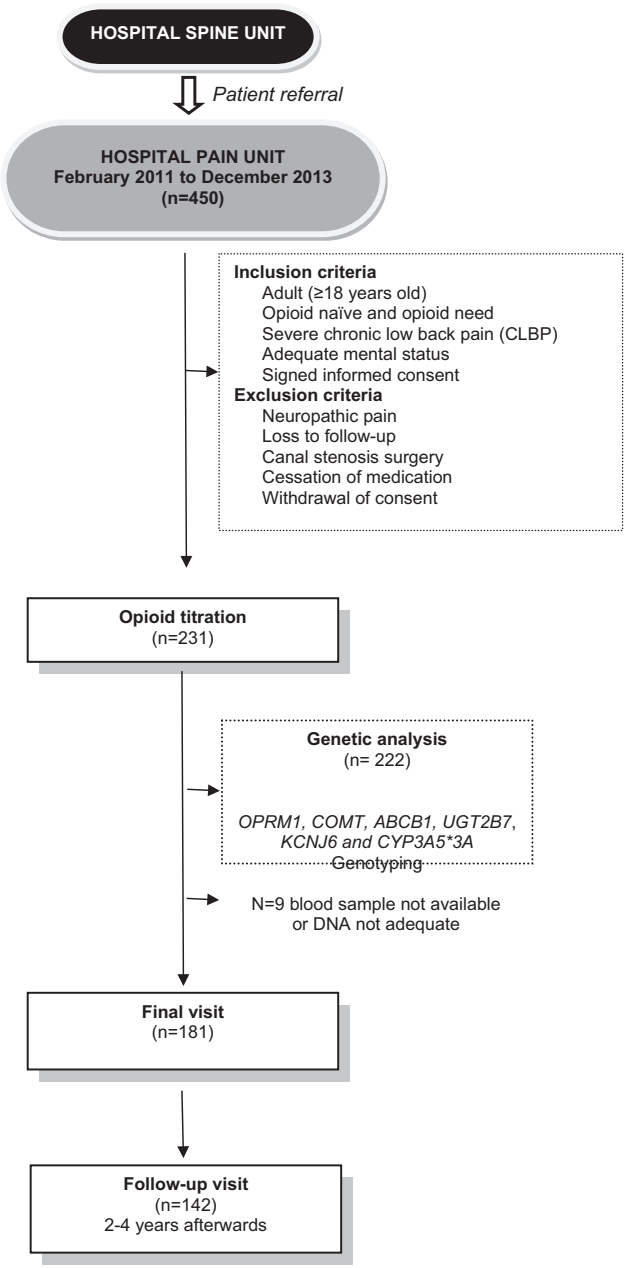

Figure 1. Study flow chart from baseline to final visit, after 3 months of opioid titration, and follow-up visit after 2 to 4 years in patients with chronic low back pain.

of life score was moderate (39 ± 19 mm) with intense disability (ODI 56%), indicating that pain was a primary problem for these patients, though they might also be experiencing significant problems in travel, personal care, social life, sexual activity, or sleep. At the first visit, fentanyl and tramadol were the most prescribed opioids (Figure 2), 89% with concomitant drugs, with pregabalin showing the most potential drug interaction.

Table 1. Pharmacological Data of Chronic Low Back Pain Patients Before and After Opioid Titration and at Follow-up Visit

	Baseline*	Final (3 months)	Follow-up (2 to 4 years)	*P* Value†	*P* Value‡
Age (years)	63 ± 14	—		—	
BMI (kg/m^2)	29 ± 6	—		—	
Gender (% females)	63	—		—	
MEDD (mg/day)	49 ± 42	55 ± 49	58 ± 74	0.157	0.585
Opioid drugs (%)	93	83	66	**0.001**	**<0.001**
Fentanyl	34	26	10	0.088	**<0.001**
Oxycodone	10	20	15	**0.004**	0.213
Morphine	14	6	5	**0.003**	1.000

*Patients were naïve to opioid prescription in pretreatment before the baseline visit when patients signed the informed consent and the physician began opioid titration. Final visit corresponds to values after 3 months of opioid titration. Follow-up visit corresponds to the visit after 2 to 4 years of opioid titration.
†*P* values correspond to the comparison of baseline vs. final visits.
‡*P* values correspond to the comparison between final and follow-up visits.
Bold *P* values means <0.05.
BMI, body mass index; MEDD, morphine equivalent daily dose.

Table 2. Clinical Data at the Beginning of the Study and 3 months After Opioid Titration in Chronic Low Back Pain

	Baseline	Final (3 months)	*P* Value
Pain intensity (VAS, range 0 to 100 mm, mean ± SD)	74 ± 17	55 ± 25	**<0.001**
Pain relief (VAS, range 0 to 100 mm)	nd	32 ± 29	nd
Quality of life (VAS, range 0 to 100 mm, mean ± SD)	39 ± 19	46 ± 23	**0.007**
Pain intensity (%)			
None	1	5	**<0.001**
Mild	3	26	
Moderate	29	42	
Severe	51	23	
Extremely severe	15	4	
Neuropathic component (PainDetect, range 0 to 38)	13 ± 7	10 ± 7	**0.002**
Likely (%)	24	14	
Unclear (%)	25	22	
Unlikely (%)	50	64	
Oswestry Disability Index (%)			
Minimal	3	8	**0.003**
Moderate	20	34	
Severe	56	39	
Crippled	20	17	
Bedbound or exaggerating	0	1	
Hospital anxiety and depression (range 0 to 21)			
Anxiety	8 ± 5	6 ± 5	**0.001**
Depression	8 ± 5	7 ± 5	**0.016**

P values of <0.05 are in boldface.
nd, not determined; SD, standard deviation; VAS, visual analog scale.

Three months after opioid titration, at the final visit, 60% of the patients (n = 122) required dose modification from the baseline visit and 58% required opioid rotation, with a final MEDD of 55 mg/day. Oxycodone use was significantly increased from the baseline visit

(10%, $P = 0.004$; OR = 0.435; 95% CI = 0.2484 to 0.7620), while morphine use was significantly reduced (9%, $P = 0.003$; OR = 2.863; 95% CI = 1.402 to 5.849). Thus, at the final visit, 66% of the patients and 62% of the physicians reported a positive increase in analgesia. Prescription produced a significant decrease in pain intensity (VAS 55 ± 25 mm, ΔVAS −21 ± 25 mm, $P < 0.001$, $d = 0.954$, $r = 0.430$) and a reduction in the neuropathic component (from 24% to 14%), which was accompanied by a significant increase in quality of life (VAS 46 ± 23 mm, $P = 0.007$, $d = 0.360$, $r = 0.177$), decreased disability (ODI 39%, moderate grade, $P = 0.003$, Crammer's $V = 0.222$), and anxiety and depression (scores of 6 ± 5, $P = 0.001$, 95% CI = 0.6883 to 2.849, $d = 0.363$, and 7 ± 5, $P = 0.034$, 95% CI = 0.09061 to 2.215, $d = 0.240$, respectively). The assessment of the global change was positive and similar between patients and clinicians.

At long-term follow-up (between 2 and 4 years) ($n = 145$, 63% of the original patients), opioid use was significantly decreased (OR = 0.408; 95% CI = 0.2498 to 0.6675, $P = 0.0004$), but fentanyl prescription was not (OR = 3.188; 95% CI = 1.738 to 5.844, $P = 0.0001$), with an increased pain intensity (VAS 61 ± 26 mm) as well as a neuropathic component (26% PainDetect positive).

Adverse Events Registration

The most frequent AEs were constipation (29%), dry mouth (27%), drowsiness (22%), sleep-related problems (20%), dizziness (18%), nervousness (17%), sexual disorder (14%), nausea (13%), depression (13%), dry skin (12%), weight gain (12%), and headache (10%) (Figure 3). Subjects with higher pain severity showed significantly greater general discomfort ($P = 0.0235$), dizziness ($P = 0.0142$), and drowsiness ($P = 0.0328$). The presence of a major neuropathic component (positive vs. inconclusive or negative) was significantly associated with increased occurrence of itching ($P = 0.0142$), sexual disorder ($P = 0.0438$), and depression ($P = 0.0038$) (Table S1).

Genetic Analyses

Allele and genotype frequencies are shown in Table 3. A total of 8 SNPs contained in 6 genes (*ORPM1*, *COMT*, *ABCB1*, *UGT2B7*, *KCNJ6*, and *CYP3A5*3A*) were genotyped by RT-PCR in 222 patients with CLBP (96%). The number of patients for genotypic analysis was the same for all the SNPs studied ($n = 222$), except for G1250A (*KCNJ6* gene) and A6986G (*CYP3A5*3* gene) ($n = 221$). Table 4 reviews their influence on pain management in the medical literature.

All the polymorphisms analyzed were in Hardy-Weinberg equilibrium (HWE), with the exception of A842G (*UGT2B7*) and A6986G (*CYP3A5*3A*) in all available samples, and C3435T (*ABCB1*) and G1250A (*KCNJ6*) in those who completed the final visit. All 4 models (dominant, recessive, codominant, overdominant, and log-additive) were tested. The lack of HWE means that genotype frequencies cannot be obtained from the allele frequencies. This is interesting in transgenerational studies, but this was not our case. Thus, we analyzed the influence of the genotype of the SNPs, except for G211T (*UGT2B7*), since all individuals were homozygous for the parent allele.

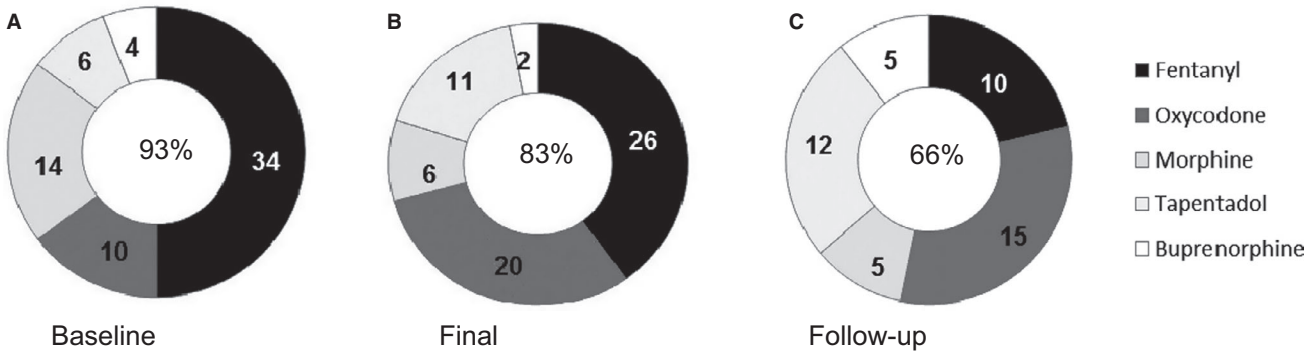

Figure 2. Total opioid prescription (% at center of circle) in patients with chronic low back pain at baseline (A), after titration procedure at final visit (B), and at follow-up visit (C).

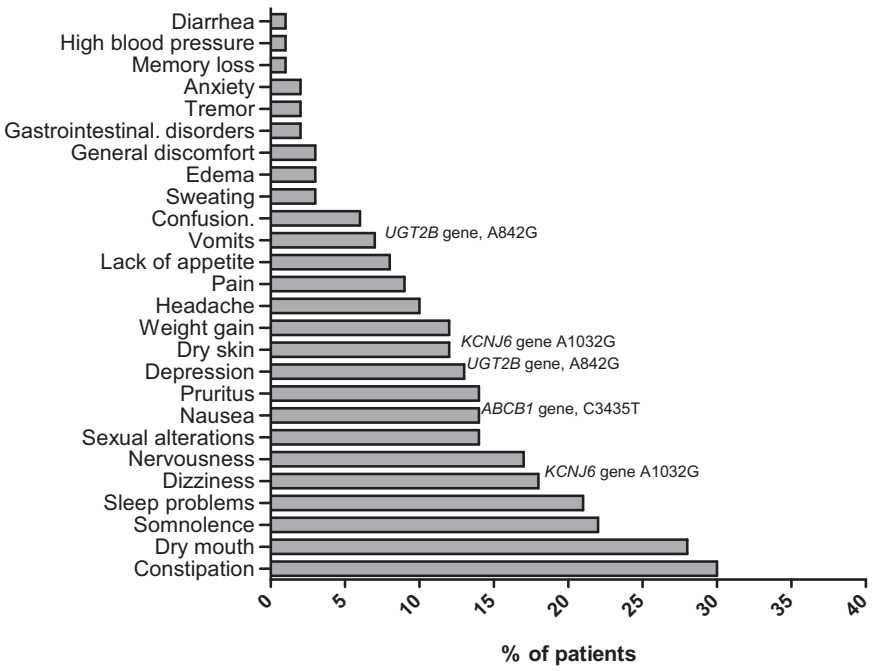

Figure 3. Distribution of adverse events and significant genotype influence in patients with chronic low back pain after titration procedure at the final visit.

Genetic Influence on Medical and Pharmacological Variables

Pain Intensity. Carriers of the *KCNJ6* gene A1032G-A allele had higher pain intensity at the final visit (A/A 54 ± 25, G/A 59 ± 25, G/G 28 ± 23 mm, $P = 0.003$, $\eta^2 = 0.063$; A/A + A/G 56 ± 25 vs. G/G 28 ± 23 mm, $P = 0.001$, 95% CI = −4.470 to −1.126, $d = 4.94$; Figure 4 and Tables S2 and S3). This higher pain intensity was maintained at the long-term visit in A1032G-A allele carriers (A/A 60 ± 27, A/G 65 ± 22, G/G 30 ± 35 mm, $P = 0.026$, $\eta^2 = 0{,}024$; A/A + A/G 62 ± 28 vs. G/G 30 ± 35 mm, $P = 0.012$, 95% CI = −57.36 to −7.411, $d = 2.864$). Similarly, differences in pain intensity at the final visit were found for the *COMT* gene, where G472A-AA individuals presented higher pain values at the final visit (G/G 57 ± 27, G/A 50 ± 25, A/A 62 ± 23 mm, $P = 0.039$, $\eta^2 = 0.035$). No differences in pain intensity at baseline or after opioid titration were observed in any of the other polymorphisms.

Neuropathic Component. Carriage of the *OPRM1* gene A118G-G allele was associated with the presence of a neuropathic component (PainDetect A/A 13%, A/G 38%, G/G 57%, $P = 0.002$, Cramer's $V = 0.233$), more disability (ODI ≥ 61 mean score, A/A 16%, A/G 24%,

Table 3. Analysis of Allele and Genotype Frequencies of Gene Polymorphisms in Chronic Low Back Pain

Gene Variants	Genotype Frequency (%)	Allele Frequency (%)	HWE *P* Value
OPRM1 A118G			
A/A	63	A 79	0.430
A/G	32	G 21	
G/G	5		
COMT G472A			
A/A	22	A 50	0.107
A/G	56	G 50	
G/G	22		
ABCB1 C3435T			
T/T	27	T 52	1.000
T/C	50	C 48	
C/C	23		
UGT2B7 G211T			
G/G	100	G 100	nd
UGT2B7 A842G			
A/A	32	A 66	**0.000**
A/G	68	G 34	
KCNJ6 A1032G			
A/A	63	A 79	0.842
A/G	33	G 21	
G/G	5		
KCNJ6 G1250A			
A/A	41	A 64	1.000
A/G	46	G 36	
G/G	13		
*CYP3A5*3A* A6986G			
A/A	1	A 1	**0.000**
G/G	99	G 99	

P values of <0.05 are in boldface.
HWE, Hardy-Weinberg equilibrium; nd, not determined; SNP, single-nucleotide polymorphism.

Table 4. Significant Influence of Single-nucleotide Polymorphisms in Chronic Low Back Pain Intensity and Opioid Consumption

Gene SNP	Clinical/Opioid Dose Influence	References
COMT G472A	Homozygosis (final): lower pain intensityMore opioid consumption	*COMT* gene variants affect enzymatic activity and opioid-induced pain relief via adrenergic pathways in chronic pain, postoperative pain, and the analgesic response[48] = *COMT* rs4680A allele carriers reported higher bone pressure pain tolerance threshold (ie, less pain) by up to 23.8% ($P < 0.015$)[49] = *COMT* haplotype rs4646312T>C/rs165722T>C/rs6269A>G/rs4633T>C/rs4818C>G/rs4680A>G as possible relevant modulators of long-term postsurgical pain outcome[50] = *COMT* rs4860 may not contribute to chronic postsurgical pain development after cesarean delivery[51]
KCNJ6 A1032G	A1032G-A allele (final and follow-up): higher pain intensityHigher opioid consumptionDizziness and dry skin	Important molecule in pain transmission ≠ *KCNJ6* A1032G and G-1250A/A1032G haplotype could serve as markers that predict increased analgesic requirements and the risk for chronic postoperative pain[40] = *KCNJ6* 1250A and *COMT* Val alleles predispose to diminished opioid-induced pain relief[52] = Associated with sensitivity to both cold and mechanical pain, susceptibility to nicotine dependence, and successful smoking cessation[53]
OPRM1 A118G	A118G-GG (baseline): less neuropathic componentInfluence on opioid consumption	It encodes an alternative isoform that affects incidence, intensity, or duration of chronic pain and the consumption of opioids[54,55] ≠ For patients carrying *OPRM1* 118-AG/GG and *COMT* 472GG or these genotypes alone, in cancer pain a significantly higher median percentage dose increase was observed (95.2% [32.8 to 345], $P = 0.0016$)[56] ≠ *OPRM1* A118G-AA patients required significantly lower opioid dose in deprescription procedure in opioid use disorder[57] ≠ A118-GG allele is associated with decreased acute postoperative pain relief after piritramide and opioid dose requirements[58,59] ≠ Homozygous for *OPRM1* A118G-A allele carriers needed significantly lower doses of morphine for pain relief (30% of differences)[58] ≠ Influences the analgesic effect of morphine for immediate acute postoperative pain in children[60,61]
UGT2B7 A842G	A842G-GG (final): more neuropathic component, depression and vomiting	Increased conjugation for buprenorphine and morphine ≠ Fentanyl sensitivity for cold pressor-induced pain was associated with the rs7439366, rs4587017, and rs1002849 SNPs of the *UGT2B7* gene. Different dose of morphine.[62] ≠ Needed significantly lower dose of morphine for pain relief. The same trend was observed for patients homozygous for ABCB1 1236T and 3435T.[58]
ABCB1 C3435T	No influence on pain intensity or opioid consumption but more nausea	*ABCB1* gene encoding the xenobiotic transporter P-gp may influence outcome of treatment with P-gp substrates ≠ Carriers of other *ABCB1* (C1236T-TT) genotypes presented a lower AUC and higher Cl, as well as a lower half-life for fentanyl in healthy volunteers[63] ≠ Lower opioid dose required for other *ABCB1* SNP (C1236T)[58] ≠ Needed significantly lower dose of morphine for pain relief[64]

=, in agreement with the literature; ≠, different from the literature; AUC, area under the curve; Pgp, P-glycoprotein, SNP, single-nucleotide polymorphism.

G/G 50%, $P = 0.044$, Cramer's $V = 0.196$), and higher anxiety (HADS scores A/A 7 ± 5, A/G 9 ± 4, G/G 13 ± 5, $P = 0.008$, $\eta^2 = 0.062$). After titration, the presence of a neuropathic component was significantly higher in the A842G-AG (*UGT2B7* gene) patients (AA 8 ± 6 vs. AG 11 ± 7, $P = 0.008$, 95% CI = −5.608 to −0.8855, $d = 0.498$; Table 5).

Opioid Dose. Patients with the *KCNJ6* gene G1250A-GG genotype required a higher baseline MEDD (27 mg/day, $P = 0.012$, $\eta^2 = 0.028$) and a higher maximum opioid dose (A/A 60.8 ± 5, A/G 69 ± 5, G/G 98 ± 16, $P = 0.008$, $\eta^2 = 0.048$), with a difference of 15.4 mg (95% CI = 11 to 54, $P = 0.008$; data not shown).

Adverse Events. Data showed that some gene variants affected the distribution of certain AEs, such as gastrointestinal and cognitive AEs. Nausea appeared more significantly with the *ABCB1* gene C3435T-C allele carriers ($P = 0.005$; Crammer's $V = 0.217$). Also, differences in prevalence among genotypes were observed for vomiting and depression in the *UGT2B* gene A842G polymorphism, and the *KCNJ6* gene A1032G for dizziness and dry skin (data not shown).

DISCUSSION

SNPs in genes involved in pain transmission and opioid metabolism might predispose to exaggerated sensitivity and differences in the opioid analgesic effect in patients with CLBP. The relevance of the *KCNJ6* A1250G (rs6517442) and *COMT* G472A (rs4680) variants in pain intensity and opioid dose requirements, together with the influence of the *OPRM1* A118G-GG

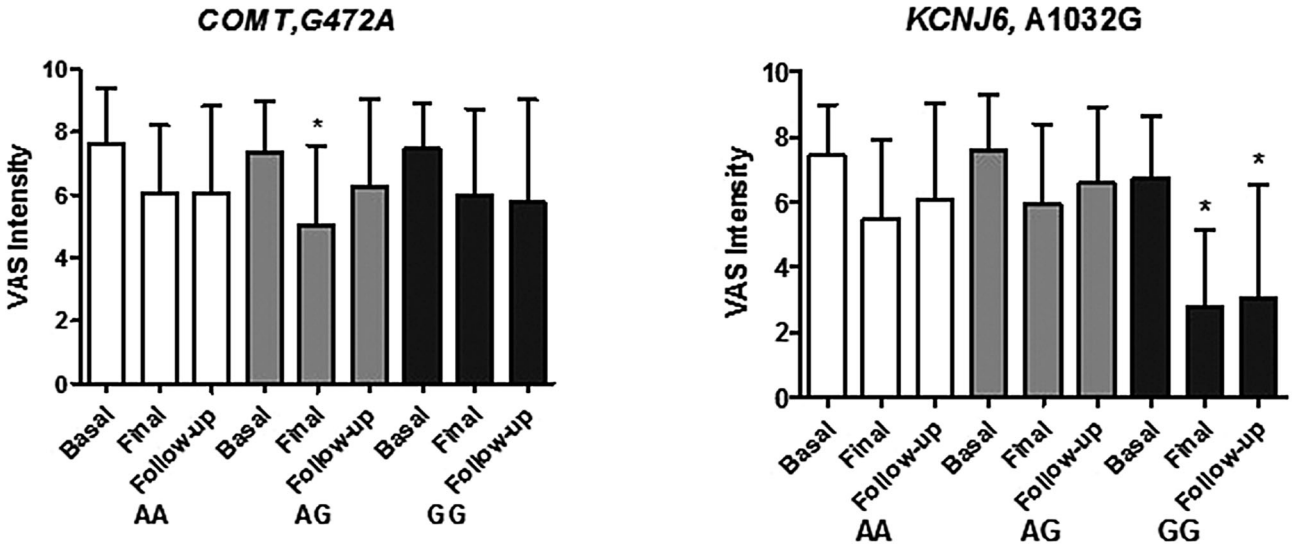

Figure 4. Analysis of pain intensity according to G472A *COMT* and A1032G *KCNJ6* genotypes in patients with chronic low back pain after titration procedure, at final visit, and at follow-up. * means $P < 0.05$ from basal visit.

(rs1799971) and *UGT2B7* A854G-GG (rs776746) genotypes in the neuropathic component, could be relevant in clinical practice. We encourage clinical trials of their clinical application in chronic pain management.

Our study population represents standard patients with CLBP from a tertiary hospital, where most patients were elderly women with moderate-severe pain and undertreated with a nonregularly prescribed analgesic prior to inclusion. Multimodal pain treatment was seen and consistent with the Spanish Drugs Agency, where the opioids oxycodone and tapentadol displaced morphine at the follow-up visit.[27–30] The drugs induced a significant reduction in pain intensity that was also accompanied by a significant iimprovement in quality of life, plus a decrease in disability, depression, and anxiety.[31,32] In addition, subjects with higher pain severity or an associated neuropathic component had more AEs. These can worsen the quality of life of the patient with chronic pain and by themselves cause more comorbidities and loss of working days, and obstruct the ability to live a healthy social life.[33]

Clinical care and research in lumbar spinal stenosis is complicated by the heterogeneity of the condition, the lack of standard criteria for diagnosis and inclusion in studies, and the high rates of anatomic stenosis on imaging studies in older people who are completely asymptomatic.[34] The natural history of spinal stenosis remains poorly understood, with studies reporting that about half of patients remain clinically stable, with a quarter worsening or improving.[35] As current therapy is insufficient, we considered that targeting patients' genetic disposition would lead to better pain management.

Table 5. Comparison of the Neuropathic Component (PainDetect Questionnaire) in Chronic Low Back Pain at the Baseline, Final and Follow-up Visits According to Single-nucleotide Polymorphism

SNP	Baseline	Final (3 months)	Follow-up (2 to 4 years)
OPRM1 A118G			
G/G	11 ± 6	13 ± 7	13 ± 8
G/A	15 ± 8	11 ± 7	14 ± 9
A/A	15 ± 10	13 ± 8	18 ± 6
P-value	**0.002**	0.174	0.231
UGT2B7 A842G			
A/A	12 ± 8	8 ± 6	13 ± 9
G/G	13 ± 7	11 ± 7	13 ± 9
P-value	0.328	**0.008**	1.000

Chi-square test was performed to compare the neuropathic component at baseline or final visits between genotypes. $P < 0.05$ is written in bold.
SNP, single-nucleotide polymorphism.

For all the potential genetic markers studied, we analyzed candidate genes recommended for clinical implementation.[36] We concluded that homozygosis for the *COMT* G472A-A and *KCNJ6* A1032G-A alleles predisposed to diminished opioid-induced pain relief, significantly higher opioid requirements, and more side effects (dizziness and dry skin). *COMT* gene variants have been reported to lead to an enzyme up to 4 times less active associated with increased opioid-induced pain relief among patients chronically treated for cancer[37] and an increased risk for the development of chronic pain disorders,[38] such as chronic postoperative pain.[39]

This is in concordance with other population pain studies that showed increased pain and postoperative analgesic requirements[40] in patients with spinal disc herniation, phantom limb pain, after traumatic limb amputation, and persistent breast pain after surgery.[41]

Current research has also indicated that *OPRM1* A118G-A allele carriers present more pain comorbidities (disability and higher anxiety) and a neuropathic component; this was also found for *UGT2B7* A842G-GG individuals. Variability in the regulatory region of the enzyme encoded by the *UGT2B7* gene has the potential to alter its expression and activity, influencing analgesic response, but the clinical significance has not yet been well defined. Results should be interpreted with caution due to the limited number of samples and possible heterogeneity between the studies. Well-designed and large-scale studies are necessary to confirm our results.[42]

We found that the C3435T-C allele (*ABCB1* gene) and A842G-A allele (*UGT2B7* gene) variants were related to fewer gastrointestinal AEs, in agreement with the literature.[43,44] However, this observation was not reproduced in a multicenter study involving patients chronically treated with morphine[45] or in other pharmacokinetic studies.[46]

Our findings should be interpreted in light of some limitations. First, the sample size was small, young adults were not included, and data were obtained from a single clinic and ethnic background. In addition, patients with neuropathic or nociceptive pain were excluded to homogenize the patient sample. This could condition the validity of our results and the extrapolation to other populations. Second, patients are very commonly prescribed several medications for multiple comorbidities, and genetics can only partially explain the variability in patient responses to analgesic drugs.[47] Furthermore, to be able to establish more robust conclusions, an analysis taking into account the type of opioid prescribed should be performed. For example, *UGT2B7* is involved in morphine and hydromorphone metabolism, so it should only be analyzed in patients taking these drugs. There have been many advances in the use of opioids in recent years; consequently, the position of morphine as the gold standard has gradually become more questioned. Third, the conclusions of this study may not remain valid in the next generation based on the HWE.

As an emerging field, pharmacogenetics confronts new challenges such as ensuring its correct standardization and correct translation to routine clinical practice.[19] Pharmacogenetics implementation will require the establishment of stable phenotype-genotype relationships through controlled clinical trials and cost-effectiveness studies.

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CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare.

Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1. Analysis of adverse events according to age, pain intensity, and neuropathic component in patients with chronic low back pain after titration procedure at final visit.

Table S2. Analysis of pain intensity at the beginning of the study (baseline), after opioid titration (final), and at the follow-up visit according to genotype in patients with chronic low back pain.

Table S3. Analysis of pain intensity after opioid titration (final visit) according to different genetic models for *COMT G472A* and *KCNJ6 A1032G* polymorphisms in patients with chronic low back pain.

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