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***κ*-opioid Receptor Gene as a Predictor of Response in a Cocaine Vaccine Clinical Trial**

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Abstract

Objectives—We examined a pharmacogenetic association between a variant in the *κ*-opioid receptor (*OPRK1*) gene and the response to treatment with a cocaine vaccine tested in a recent clinical trial. This gene has a protective allele for opiate addiction that may act by inhibiting dopamine activation associated with reinforcement.

Methods—Sixty-nine DNA samples were obtained from 114 cocaine and opioid dependent subjects who were enrolled in a 16 week Phase IIb randomized double-blind placebo-controlled trial and who received five vaccinations over the first 12 weeks. We genotyped 66 of these subjects for the rs6473797 variant of the *OPRK1* gene and we compared vaccine to placebo subjects in terms of cocaine-free urines over time.

Results—Using repeated measures analysis of variance, corrected for population structure, vaccine pharmacotherapy reduced cocaine positive urines significantly based on *OPRK1* genotype. In subjects treated with the cocaine vaccine, those who were homozygous for the protective A allele of rs6473797 had the proportion of positive urines drop from 78% to 51% on vaccine (point-wise $P < .0001$, experiment-wise $P < .005$), while the positive urines of those individuals carrying the non-protective, risk G allele dropped from 82% to 77%. Strong interactions of treatment by SNP (single nucleotide polymorphism) reflected a lower baseline and significant reduction for placebo subjects with the risk G allele ($P < 0.00001$).

Conclusions—This study indicates that a patient's *OPRK1* genotype could be used to identify a subset of individuals for which vaccine treatment may be an effective pharmacotherapy for cocaine dependence.

Keywords

Gene; vaccine; polymorphism; cocaine; treatment; opioid

Introduction

Cocaine dependence (CD) has substantial social and economic impacts on those three million abusers who are afflicted with this disease (McLellan *et al.* 2000; SAMSHA 2011).

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There are no conflicts of interest.

While CD has no FDA approved pharmacotherapy, a recent study has used a cocaine vaccine pharmacotherapy in an effort to treat cocaine dependence (Martell *et al.* 2009). This vaccine was made of an analog of cocaine, succinylcocaine conjugated to recombinant cholera toxin B-subunit protein. After multiple injections of the vaccine, antibodies against cocaine are produced. It is hoped that these anti-cocaine antibodies will block the euphoric effects of cocaine, attenuating the rewarding properties of cocaine if the patient would lapse from abstinence. In this study, the cocaine vaccine was found to significantly reduce cocaine urines among actively vaccinated patients who attained sufficient antibody levels (over 40 µg/ml), which could effectively block most street doses of cocaine (Martell *et al.* 2009). However, only 40% of the patients attained these >40 µg/ml blocking levels of antibodies, while 70% of them attained levels over 22 µg/ml, which had been sufficient to block one to two doses of smoked cocaine (Haney *et al.* 2010). This more modest blockade was considered sufficient to prevent the priming effect that often precedes a relapse to repeated use and dependence (de Wit 1996). A portion of these patients with lower antibody levels showed a reduction in cocaine use, and perhaps the identification of pharmacogenetic interactions may identify those who would reduce their cocaine use, although their antibody levels were below the optimal blocking antibody level of 40 µg/ml.

A few genetic variants have been identified as risk or protective factors for cocaine abuse, and some variants have been identified as relevant to both opiate and cocaine abuse (Haile *et al.* 2008; Haile *et al.* 2009). Among these genetic variants is the rs6473797 variant in intron 2 of the κ -opioid receptor *OPRK1* gene, an A → G transition, with the G allele having been shown to increase vulnerability of developing opioid addiction (Levrin *et al.* 2008) and of developing alcohol dependence (Xuei *et al.* 2006).

The cocaine vaccine acts pharmacokinetically by preventing cocaine from entering the brain. This pharmacokinetic action has two components. First, the vaccine slows cocaine entry into the brain, reducing the euphoria that results from the rapid rise in cocaine levels, which then quickly block the dopamine transporter (Uhl *et al.* 2002). Second, the vaccine prolongs brain levels of cocaine that may lead to more sustained aversive effects such as anxiety and paranoia, which are not dependent on a rapid rise in dopamine. These two actions of the vaccine contribute to its efficacy and could interact with abnormal dynorphin activity at variant κ -opioid receptors (KOP-r) such as the receptor encoded by the *OPRK1* rs6473797 risk G allele. KOP-r stimulation by its endogenous ligand dynorphin inhibits dopamine activity in the ventral tegmental area and its connection to the nucleus accumbens, and the risk G allele may act to enhance this inhibitory activity (Chefer *et al.* 2005). This risk allele may itself increase the expression of *OPRK1* or may be a marker for an allele that codes for a receptor that is relatively efficient at reducing the duration of the dopamine peak, as illustrated in the top part of the Figure 1. When the vaccine prolongs brain levels of cocaine, the euphoric period is extended in the G allele patients. Thus, the vaccine patients might not show any improvement in cocaine dependent patients with this variant, while the placebo patients would respond to the weekly relapse prevention cognitive behavioral therapy (CBT), that all subjects in this study received, and reduce their cocaine use from baseline (Carroll 1997; Carroll *et al.* 2004; Dutra *et al.* 2008). In contrast, lack of this risk G variant in *OPRK1*, e.g., those patients having an AA genotype, would respond to this cocaine vaccine and have a prolongation of the high dopamine levels leading to aversive actions of cocaine, as shown in the bottom half of the Figure 1. In summary, those patients with the risk G allele would do as well on placebo as those with the protective AA genotype who were vaccinated, while those with the G allele who were vaccinated would respond as poorly as those with the protective AA allele who got placebo.

Methods

Participants

One hundred and fourteen subjects, who met DSM IV criteria (APA 1994) for cocaine and opioid dependence, were enrolled and randomized into an out-patient methadone maintenance treatment program in West Haven, Connecticut between October 2003 and April 2005. The MINI (Sheehan *et al.* 1997) and the Addiction Severity Index (ASI) (McLellan *et al.* 1992) were completed on all subjects for the assessment of psychiatric and baseline characteristics. DNA samples were obtained from 71 subjects for these analyses. The *OPRK1* rs6473797 genotype was unable to be completed for five subjects, yielding a final cohort of 66 subjects. Eligibility included men or women aged 18 to 55 who had positive urine cocaine and did not have a clinically unstable chronic disease. Enrolled women had to be non-child bearing or willing to use birth control. The institutional review boards of the VA Connecticut Healthcare System, the Yale University School of Medicine, and the Baylor college of Medicine approved this study, in which all subjects gave written informed consent.

Study Design, Medications, and Counseling

The original publication provides study design details of this randomized, placebo controlled clinical trial (Martell 2009). For the current analysis, we examined treatment response during weeks one through 16 in order to extend past the 12 weeks needed to complete the full vaccination series. Differential efficacy between the placebo and active vaccine was expected to begin after week 8, when most vaccine responders should have significant IgG anti-cocaine levels (Kosten *et al.* 2002; Martell *et al.* 2005).

Methadone maintenance therapy was initiated prior to the first vaccination and the dose was stabilized by week 8 at 83 ($\pm$ 16 s.d.) mg daily. The vaccine was succinyl norcocaine covalently linked to cholera B (SNC-rCTB) and adsorbed onto aluminum hydroxide adjuvant. Subjects were randomized to receive either five vaccinations of 360 micrograms of active (SNC-rCTB) or placebo vaccine intramuscularly at 0, 2, 4, 8, and 12 weeks. All subjects participated in individual, weekly, 30–45 minute, relapse prevention cognitive behavioral therapy (CBT) sessions conducted by trained substance abuse counselors (Carroll 1997; Carroll *et al.* 2004). During these counseling sessions, urine toxicology results were reviewed with the subjects.

Outcome measures and laboratory tests

Supervised urine samples were obtained thrice weekly and tested for the presence of the cocaine metabolite benzoylecgonine using an Olympus AU 640 Emit system (Olympus America Inc., Melville, NY) with a cut-off concentration of 300 ng/ml. We obtained saliva samples for the isolation of DNA for genotyping.

Genotyping

DNA was purified as previously described (Kosten *et al.* 2012). Briefly, DNA was isolated using the Gentra Puregene Buccal Cell Kit (Qiagen, Valencia, CA) following the manufacturer's recommendations from pelleted buccal cells that were obtained by centrifugation of 10 ml Scope mouthwash that was used to rinse the subject's mouth for 60 seconds. Genotypes were determined using a 5'-fluorogenic exonuclease assay (TaqMan®, Applied Biosystems, Foster City, CA). The *OPRK1* rs6473797 genotype was determined using the TaqMan® primer-probe set (Applied Biosystems) Assay ID C_2898340_20. PCR amplification was performed using Platinum® quantitative PCR SuperMix-UDG (Invitrogen, Carlsbad, CA) on a GeneAmp® PCR system 9700. Samples were amplified at

50°C for 2 min, 95°C for 10 min, and then 50 cycles of 95°C for 15 s and 60°C for 1 min. The amplification products were analyzed using an Applied Biosystems Prism® 7900 sequence detection system and SDS 2.2 software (Applied Biosystems). A PCR assay that identifies the presence of the Y chromosome-specific SRY gene was used to confirm the subject's sex and ancestry informative makers were used to assess population structure (Kosten *et al.* 2012). TaqMan® assays were performed in duplicate by an individual unaware of the clinical status of the subjects. Excluding the SRY assay and the ten ancestry informative makers, 25 variants have been examined for pharmacogenetic association using this dataset. DNA was not available for all patients so the *OPRK1* rs6473797 genotype was obtained only for 66 patients.

Statistical Analysis

The two treatment groups were compared for baseline differences in cocaine use history and demographics using χ^2 or t-test as appropriate. As previously described (Kosten *et al.* 2012), a repeated measures analysis of variance using the number of cocaine positive urines over the total number of visits (eight) for each two-week period to compare vaccine to placebo over time and to determine if the effect of vaccine is modulated by genotype or genotype pattern using R version 2.9.1 (R_Development_Core_Team 2009). The explanatory variables were condition (whether a patient was taking vaccine or placebo), genotype group (0 = AG/GG, 1 = AA). A repeated measures ANOVA was performed using the data on all individuals who had complete data (n = 61) and unbalanced repeated measures ANOVA for all individuals (n = 66). Similar results were obtained from each analysis. The data in figure 2 is displayed as the percent cocaine-positive urines during each two-week period and the data in figure 3 is displayed as the percent cocaine-positive urines relative to the level at baseline (study week 1–2) of that particular treatment-genotype group. Effect size was calculated as a partial eta-squared statistic using a ratio of condition or SNP variance to the residual variance. There are three general levels of cutoffs for effect size: 0.01 is a small effect, 0.06 is medium, and 0.14 is large.

Population structure was determined as previously described (Kosten *et al.* 2012). All analyses were corrected for any possible confounding effects, by including the proportion of each subject from the founder populations as well as gender as covariates in the model. *P*-values were similar to those obtained when we did not correct for these covariates. Analyses were performed with the total group, then within the two genotype subgroups. Corrections for multiple testing were performed to evaluate experiment-wise significance by applying the Bonferroni correction.

Results

Baseline characteristics and retention by medication and κ -opioid receptor gene

We obtained genotypes for *OPRK1* rs6473797 on only 66 of the 114 patients enrolled in this clinical trial. These 66 patients were randomized: 33 to the vaccine and 33 to placebo. The patients included 38 with the AA and 20 with the AG and eight GG genotypes. The 66 patients were mostly Caucasian males with a mean age of 36 years and who abused cocaine for a mean of 13 years and spending \$370 in the month before entering the study. The Addiction Severity Index showed few problems except in the drug abuse area. As shown in Table 1, we found no significant baseline differences among the four treatment by genotype groups in any clinical characteristics ($P > .05$). Treatment retention showed no difference; 92% completed the full 16 week study.

Cocaine treatment outcomes and κ -opioid receptor gene

We divided the 66 patients who were genotyped for the *OPRK1* rs6473797 variant into two groups based on whether they carried the *OPRK1* rs6473797 G allele. One group consisted of those homozygous for the major frequency A allele, the AA genotype group, and the other group who consisted of carriers of the minor frequency G allele, those with either a AG or GG genotype. The cocaine positive urine rates across the 16-week clinical trial differed in the vaccine group for patients in the AA versus the AG/GG groups with both point-wise ($F = 15.3$; $df = 1,270$; $P < .0001$; Fig. 2A) and experiment-wise significance ($P < .005$), but not for those in the placebo group ($F = 3.08$ $df = 1,262$; $P > .05$; Fig. 2B). The SNP effect was 0.0618. The rate of cocaine positive urines for the patients in the vaccine group was 82% for those in the AG/GG genotype group and 78% for those in the AA genotype group during the two baseline weeks. Positive urine cocaine rates in these groups decreased to 77% for the AG/GG group and to 51% for the AA group during the last four-week period. If we included only the 61 patients who completed the study, vaccine treatment remained effective only in the AA group with both point-wise ($F = 10.75$; $df = 1,240$; $P < .002$) and experiment-wise significance ($P < .05$).

We also found a significant treatment by genotype interaction, in which cocaine positive urine rates for placebo patients in the AG/GG group were lower at baseline compared to the AG/GG vaccine treatment group (66% versus 82%), and decreased significantly during the 16-week trial from 66% to 37% with both point-wise ($F = 32.2$; $df = 2,456$; $P < .00001$) and experiment-wise significance ($P < .00001$), having an effect size of 0.064. The AA placebo group showed no significant reduction on placebo, going from 63% to 54%. In figure 3, we have displayed these results relative to the levels observed at baseline. Importantly, the vaccine AA group did not differ from the placebo AG/GG group, when relative to their baseline levels. Both patient groups improved substantially. However, the AA patients required the vaccine to improve, while the patients carrying the protective AG/GG improved with the CBT alone and started at a lower level of cocaine abuse at baseline.

Opioid Treatment Outcomes and κ -opioid receptor gene

Opioid positive urine rates within the vaccine group decreased over time in a similar manner for all the two *OPRK1* genotype groups. The mean opioid positive rate in the TT started at 40% and decreased to 16%, while within the CT/CC group started at 38% and decreased to 21% ($P = .87$). Within the placebo group the trend was similar: the opioid positive urines started at 53% in the TT group and decreased to 25%, while in the CT/CC group it started at 60% and decreased to 19% ($P = 0.06$). No significant correlation was found between the rates of opiate and cocaine positive urines ($r = -0.04$).

Discussion

Subjects with the *OPRK1* rs6473797 AA genotype, who had two copies of the allele associated with protection for opiate and alcohol dependence, reduced their cocaine positive urines, when treated with vaccine. These urine rates dropped 8-fold more with the vaccine in patients having the risk genotype compared to the protective genotype (40% versus 5% drop from baseline). However, the risk variant genotypes in the placebo group showed a greater reduction in cocaine urines than those in the placebo group with the protective variant genotypes (32% versus 6% drop from baseline). The risk genotype among the placebo group also showed lower baseline rates of cocaine abuse than the protective genotype showed [66% (AG/AA) versus 82% (AA)].

The positive response to the vaccine in the protective allele group seems readily understood, but the placebo response in the risk allele group is not obvious. Two mechanisms may

contribute to these treatment responses, both of which are dependent upon the signaling efficiency of the KOP-r variants. Dynorphin A (1–17), KOP-r's endogenous ligand, attenuates cocaine-induced conditioned place preference (CPP) (Zhang *et al.* 2004a), and synthetic KOP-r ligands block CPP (Heidbreder and Shippenberg 1994; Zhang *et al.* 2004b). These CPP actions of dynorphin A (1–17) probably are due to its actions in decreasing basal and drug (cocaine)-induced striatal dopamine levels (Claye *et al.* 1997; Spanagel *et al.* 1990) (Zhang *et al.* 2004b). Chronic “binge” cocaine induces KOP-r expression in specific dopaminergically (DA) innervated brain regions suggesting that cocaine induces dynorphin and KOP-r activity as a regulatory response to maintain homeostasis by lowering DA activity (Unterwald *et al.* 2001; Unterwald *et al.* 1994). In humans, activation of KOP-r by dynorphin A (1–13) injection raises serum prolactin levels probably by lowering DA levels (Kreek *et al.* 1999). Furthermore, KOP-r stimulation in humans is dysphoric (Dykstra *et al.* 1997; Walsh *et al.* 2001a; Walsh *et al.* 2001b). Conversely, when KOP-r is inactivated pharmacologically, it decreases pain sensitivity and reduces response to stress (McLaughlin *et al.* 2003; Simonin *et al.* 1998). We therefore propose a model in which the G risk allele codes for a KOP-r that efficiently counteracts the surge in dopamine associated with cocaine, whereas the A allele codes for a KOP-r with reduced signaling efficiency. In this model, carriers of the G risk allele will experience a surge in dopamine followed by relatively rapid decline in dopamine (Figure 1, top panel). This rapid decline in dopamine levels may produce the “crash” felt by cocaine addicts. These subjects would respond to CBT due to the aversive “crash”. When these subjects with the risk allele are treated with the cocaine vaccine, the initial increase in brain cocaine levels would be delayed, as the vaccine would reversibly bind a portion of the cocaine in the blood. Hence, subjects with the risk allele who were treated with the cocaine vaccine would not have the aversive rapid drop in dopamine that would be experienced by those with the risk allele after taking cocaine. The slower the decrease in dopamine levels would lessen the “crash”. Since the cocaine has less aversive properties, these subjects would not respond to the vaccine or to CBT. In those subjects with the AA protective genotype, the inefficient form of KOP-r would not as effectively lower dopamine levels following cocaine use as those carrying the risk allele (Figure 1, bottom panel). Since these subjects would have a slower decline in dopamine levels following the dopamine surge, they would not have as severe a “crash” as those carrying the G risk allele would. When given the cocaine vaccine, the decline in dopamine would be attenuated further producing aversive paranoid affects. Due to these aversive affects, these subjects would respond to CBT. Why the more efficient *OPRK1* G allele is a risk factor for opioid dependence and alcoholism may be that, although both opioids and alcohol raise dopamine levels, the rise may be slow. A more dramatic rise and fall may be found in individuals with the risk allele after taking opioids or alcohol and this may be more pleasurable.

The rs6473797 variant is located in the second intron of *OPRK1* and may not be functional. However, it could alter expression or splicing the gene or may be in linkage disequilibrium with other nearby variants that alter gene expression or the primary structure of the κ -opioid receptor. However, this variant has been reported in other studies to be in association with both vulnerability of developing opioid addiction (Levrant *et al.* 2008) and alcohol dependence (Xuei *et al.* 2006). Examination of CEU (Caucasian) HapMap data showed that this variant is tight linkage ($r^2 = 1.00$) with only two other common variants, rs6473798 and rs6473799, covering a block that spans 141 nucleotides of chromosome 8. Three other common variants, rs997917, rs1365098, and rs1365097 ($r^2 = .73, .67, \text{ and } .73$, respectively) are disequilibrium at a lower r^2 . These six variants span 745 nucleotides. A large insertion deletion (indel) variant (rs35566036) of 830 nucleotides located 2,000 nucleotides upstream of the *OPRK1* translation start site and 12,000 nucleotides upstream rs6473797 has lower expression when present in promoter constructs in transient transfection assays and was

found to be associated with alcohol dependence (Edenberg *et al.* 2008). In that study, the indel was found to be in linkage disequilibrium with rs6473797 ($r^2 = .70$).

Several other variants have been studied in or near *OPRK1*. In two studies, a silent variant rs1051660 (36G>T) in exon 2 of *OPRK1* was found to be associated with opiate addiction (Gerra *et al.* 2007; Yuferov *et al.* 2004). Alcohol dependence vulnerability was found to be associated with a specific haplotype containing seven variants of *OPRK1* (Zhang *et al.* 2008). SNP-SNP interactions were found to be associated with addiction susceptibility with rs16918875 and rs702764 *OPRK1* variants and the *OPRM1* A118G variant (Kumar *et al.* 2012). Hence, the rs6473797 variant we have studied may be a marker for a nearby functional variant.

In summary, cocaine addiction has a strong genetic basis of over 70%, and the KOP-r system seems central to the effects of both cocaine and opiates (Goldman *et al.* 2005). Thus, pharmacotherapy of this relapsing brain disease may be better treated using a molecular genetics approach that might include variants of *OPRK1* (Leshner 1997; Simpson *et al.* 2002; Sofuoglu and Kosten 2006; Weisner *et al.* 2003) in order to enhance therapeutic response (Laje and McMahon 2007), increase compliance (Murphy *et al.* 2003), and decrease drug toxicity (deLeon *et al.* 2006; Malhotra *et al.* 2004; Rogers *et al.* 2002). This study shows an enhanced therapeutic response based on an interaction between a cocaine blocking agent that also alters the kinetics of cocaine and potentially takes advantage of a genetic risk factor for enhanced dysphoria from an interaction with cocaine use. The cohort analyzed in this study is relatively small such that replication of these findings in a larger cohort will be necessary to validate these findings.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

- APA. Diagnostic and statistical manual of mental disorders. Fourth Edition. Washington, D.C: American Psychiatric Association; 1994.
- Carroll KM. Manual-guided psychosocial treatment. A new virtual requirement for pharmacotherapy trials? *Arch Gen Psychiatry.* 1997; 54:923–928. [PubMed: 9337772]
- Carroll KM, Fenton LR, Ball SA, Nich C, Frankforter TL, Shi J, Rounsaville BJ. Efficacy of Disulfiram and Cognitive Behavior Therapy in Cocaine-Dependent Outpatients: A Randomized Placebo-Controlled Trial. *Arch Gen Psychiatry.* 2004; 61:264–272. [PubMed: 14993114]
- Chefer VI, Czyzyk T, Bolan EA, Moron J, Pintar JE, Shippenberg TS. Endogenous kappa-opioid receptor systems regulate mesoaccumbal dopamine dynamics and vulnerability to cocaine. *J Neurosci.* 2005; 25:5029–5037. [PubMed: 15901784]
- Claye LH, Maisonneuve IM, Yu J, Ho A, Kreek MJ. Local perfusion of dynorphin A 1–17 reduces extracellular dopamine levels in the nucleus accumbens. *NIDA Res Monogr.* 1997; 174:113.
- de Wit H. Priming effects with drugs and other reinforcers. *Exp & Clin Psychopharm.* 1996; 4:5–10.

- deLeon J, Armstrong SC, Cozza KL. Clinical guidelines for psychiatrists for the use of pharmacogenetic testing for CYP450 and CYP450 2C19. *Psychosomatics*. 2006; 47:75–85. [PubMed: 16384813]
- Dutra L, Stathopoulou G, Basden SL, Leyro TM, Powers MB, Otto MW. A meta-analytic review of psychosocial interventions for substance use disorders. *Am J Psychiatry*. 2008; 165:179–187. [PubMed: 18198270]
- Dykstra LA, Preston KL, Bigelow GE. Discriminative stimulus and subjective effects of opioids with mu and kappa activity: data from laboratory animals and human subjects. *Psychopharmacology (Berl)*. 1997; 130:14–27. [PubMed: 9089845]
- Edenberg HJ, Wang J, Tian H, Pochareddy S, Xuei X, Wetherill L, Goate A, Hinrichs T, Kuperman S, Nurnberger JI Jr, Schuckit M, Tischfield JA, Foroud T. A regulatory variation in OPRK1, the gene encoding the kappa-opioid receptor, is associated with alcohol dependence. *Hum Mol Genet*. 2008; 17:1783–1789. [PubMed: 18319328]
- Gerra G, Leonardi C, Cortese E, D'Amore A, Lucchini A, Strepparola G, Serio G, Farina G, Magnelli F, Zaimovic A, Mancini A, Turci M, Manfredini M, Donnini C. Human kappa opioid receptor gene (OPRK1) polymorphism is associated with opiate addiction. *Am J Med Genet B Neuropsychiatr Genet*. 2007; 144:771–775. [PubMed: 17373729]
- Goldman D, Oroszi G, Ducci F. The genetics of addictions: uncovering the genes. *Nat Rev Genet*. 2005; 6:521–532. [PubMed: 15995696]
- Haile CN, Kosten TA, Kosten TR. Pharmacogenetic treatments for drug addiction: alcohol and opiates. *Am J Drug Alcohol Abuse*. 2008; 34:355–381. [PubMed: 18584566]
- Haile CN, Kosten TR, Kosten TA. Pharmacogenetic treatments for drug addiction: cocaine, amphetamine and methamphetamine. *Am J Drug Alcohol Abuse*. 2009; 35:161–177. [PubMed: 19462300]
- Haney M, Gunderson EW, Jiang H, Collins ED, Foltin RW. Cocaine-specific antibodies blunt the subjective effects of smoked cocaine in humans. *Biol Psychiatry*. 2010; 67:59–65. [PubMed: 19846066]
- Heidbreder CA, Shippenberg TS. U-69593 prevents cocaine sensitization by normalizing basal accumbens dopamine. *Neuroreport*. 1994; 5:1797–1800. [PubMed: 7827335]
- Kosten TR, Rosen M, Bond J, Settles M, Roberts JS, Shields J, Jack L, Fox B. Human therapeutic cocaine vaccine: safety and immunogenicity. *Vaccine*. 2002; 20:1196–1204. [PubMed: 11803082]
- Kosten TR, Wu G, Huang W, Harding MJ, Hamon SC, Lappalainen J, Nielsen DA. Pharmacogenetic Randomized Trial for Cocaine Abuse: Disulfiram and Dopamine beta-Hydroxylase. *Biol Psychiatry*. 2012 Aug 17. [Epub ahead of print].
- Kreek MJ, Schluger J, Borg L, Gunduz M, Ho A. Dynorphin A1–13 causes elevation of serum levels of prolactin through an opioid receptor mechanism in humans: gender differences and implications for modulation of dopaminergic tone in the treatment of addictions. *J Pharmacol Exp Ther*. 1999; 288:260–269. [PubMed: 9862779]
- Kumar D, Chakraborty J, Das S. Epistatic effects between variants of kappa-opioid receptor gene and A118G of mu-opioid receptor gene increase susceptibility to addiction in Indian population. *Prog Neuropsychopharmacol Biol Psychiatry*. 2012; 36:225–230. [PubMed: 22138325]
- Laje G, McMahon FJ. The pharmacogenetics of major depression: past, present, and future. *Biol Psychiatry*. 2007; 62:1205–1207. [PubMed: 17949692]
- Leshner AI. Addiction is a brain disease, and it matters. *Science*. 1997; 278:45–47. [PubMed: 9311924]
- Levrin O, Londono D, O'Hara K, Nielsen DA, Peles E, Rotrosen J, Casadonte P, Linzy S, Randesi M, Ott J, Adelson M, Kreek MJ. Genetic susceptibility to heroin addiction: a candidate gene association study. *Genes, Brain and Behavior*. 2008; 7:720–729.
- Malhotra AK, Murphy GM Jr, Kennedy JL. Pharmacogenetics of psychotropic drug response. *Am J Psychiatry*. 2004; 161:780–796. [PubMed: 15121641]
- Martell BA, Mitchell E, Poling J, Gonsai K, Kosten TR. Vaccine pharmacotherapy for the treatment of cocaine dependence. *Biol Psychiatry*. 2005; 58:158–164. [PubMed: 16038686]
- Martell BA, Orson FM, Poling J, Mitchell E, Rossen RD, Gardner T, Kosten TR. Cocaine vaccine for the treatment of cocaine dependence in methadone-maintained patients: a randomized, double-

- blind, placebo-controlled efficacy trial. *Arch Gen Psychiatry*. 2009; 66:1116–1123. [PubMed: 19805702]
- McLaughlin JP, Marton-Popovici M, Chavkin C. Kappa opioid receptor antagonism and prodynorphin gene disruption block stress-induced behavioral responses. *J Neurosci*. 2003; 23:5674–5683. [PubMed: 12843270]
- McLellan AT, Kushner H, Metzger D, Peters R, Smith I, Grissom G, Pettinati H, Argeriou M. The Fifth Edition of the Addiction Severity Index. *J Subst Abuse Treat*. 1992; 9:199–213. [PubMed: 1334156]
- McLellan AT, Lewis DC, O'Brien CP, Kleber HD. Drug dependence, a chronic medical illness: implications for treatment, insurance, and outcomes evaluation. *JAMA*. 2000; 284:1689–1695. [PubMed: 11015800]
- Murphy GM Jr, Kremer C, Rodrigues HE, Schatzberg AF. Pharmacogenetics of antidepressant medication intolerance. *Am J Psychiatry*. 2003; 160:1830–1835. [PubMed: 14514498]
- R_Development_Core_Team. R: A language and environment for statistical computing. 2.9.1 edn.. Vienna, Austria: R Foundation for Statistical Computing; 2009.
- Rogers JF, Nafziger AN, Bertino JSJ. Pharmacogenetics affects dosing, efficacy, and toxicity of cytochrome P450-metabolized drugs. *Am J Med*. 2002; 113:746–750. [PubMed: 12517365]
- SAMSHA. Results from the 2010 National Survey on Drug Use and Health (NSDUH). Rockville, MD: 2011. NSDUH Series H-41
- Sheehan DV, Lecrubier Y, Harnett-Sheehan K, Janavs J, Weiller E, Bonara LI, Keskiner A, Schinka J, Knapp E, Sheehan MF, Dunbar GC. Reliability and Validity of the MINI International Neuropsychiatric Interview (M.I.N.I.): According to the SCID-P. *European Psychiatry*. 1997; 12:232–241.
- Simonin F, Valverde O, Smadja C, Slowe S, Kitchen I, Dierich A, Le Meur M, Roques BP, Maldonado R, Kieffer BL. Disruption of the kappa-opioid receptor gene in mice enhances sensitivity to chemical visceral pain, impairs pharmacological actions of the selective kappa-agonist U-50,488H and attenuates morphine withdrawal. *EMBO J*. 1998; 17:886–897. [PubMed: 9463367]
- Simpson DD, Joe GW, Broome KM. A national 5-year follow-up of treatment outcomes for cocaine dependence. *Arch Gen Psychiatry*. 2002; 59:538–565. [PubMed: 12044196]
- Sofuoglu M, Kosten TR. Emerging pharmacological strategies in the fight against cocaine addiction. *Expert Opin Investig Drugs*. 2006; 11:91–98.
- Spanagel R, Herz A, Shippenberg TS. The effects of opioid peptides on dopamine release in the nucleus accumbens: an in vivo microdialysis study. *J Neurochem*. 1990; 55:1734–1740. [PubMed: 1976759]
- Uhl GR, Hall FS, Sora I. Cocaine, reward, movement and monoamine transporters. *Mol Psychiatry*. 2002; 7:21–26. [PubMed: 11803442]
- Unterwald EM, Kreek MJ, Cuntapay M. The frequency of cocaine administration impacts cocaine-induced receptor alterations. *Brain Res*. 2001; 900:103–109. [PubMed: 11325352]
- Unterwald EM, Rubenfeld JM, Kreek MJ. Repeated cocaine administration upregulates kappa and mu, but not delta, opioid receptors. *Neuroreport*. 1994; 5:1613–1616. [PubMed: 7819531]
- Walsh SL, Geter-Douglas B, Strain EC, Bigelow GE. Enadoline and butorphanol: evaluation of kappa-agonists on cocaine pharmacodynamics and cocaine self-administration in humans. *J Pharmacol Exp Ther*. 2001a; 299:147–158. [PubMed: 11561074]
- Walsh SL, Strain EC, Abreu ME, Bigelow GE. Enadoline, a selective kappa opioid agonist: comparison with butorphanol and hydromorphone in humans. *Psychopharmacology (Berl)*. 2001b; 157:151–162. [PubMed: 11594439]
- Weisner C, Matzger H, Kaskutas LA. How important is treatment? One-year outcomes of treated and untreated alcohol-dependent individuals. *Addiction*. 2003; 98:901–911. [PubMed: 12814496]
- Xuei X, Dick D, Flury-Wetherill L, Tian HJ, Agrawal A, Bierut L, Goate A, Bucholz K, Schuckit M, Nurnberger J Jr, Tischfield J, Kuperman S, Porjesz B, Begleiter H, Foroud T, Edenberg HJ. Association of the kappa-opioid system with alcohol dependence. *Mol Psychiatry*. 2006; 11:1016–1024. [PubMed: 16924269]

- Yuferov V, Fussell D, LaForge KS, Nielsen DA, Gordon D, Ho A, Leal SM, Ott J, Kreek MJ. Redefinition of the human kappa opioid receptor gene (OPRK1) structure and association of haplotypes with opiate addiction. *Pharmacogenetics*. 2004; 14:793–804. [PubMed: 15608558]
- Zhang H, Kranzler HR, Yang BZ, Luo X, Gelernter J. The OPRD1 and OPRK1 loci in alcohol or drug dependence: OPRD1 variation modulates substance dependence risk. *Mol Psychiatry*. 2008; 13:531–543. [PubMed: 17622222]
- Zhang Y, Butelman ER, Schlussman SD, Ho A, Kreek MJ. Effect of the endogenous kappa opioid agonist dynorphin A(1–17) on cocaine-evoked increases in striatal dopamine levels and cocaine-induced place preference in C57BL/6J mice. *Psychopharmacology (Berl)*. 2004a; 172:422–429. [PubMed: 14712335]
- Zhang Y, Butelman ER, Schlussman SD, Ho A, Kreek MJ. Effect of the kappa opioid agonist R-84760 on cocaine-induced increases in striatal dopamine levels and cocaine-induced place preference in C57BL/6J mice. *Psychopharmacology (Berl)*. 2004b; 173:146–152. [PubMed: 14712342]

Figure 1.

Model for the role of *OPRK1* rs6473797 genotype in modulating dopamine levels following cocaine use. Top panel: Effect of cocaine on dopamine levels in patients who are carriers of the efficient G risk allele. Cocaine produces a rapid rise and a rapid but aversive, decline in dopamine levels (placebo). In patients treated with the cocaine vaccine, dopamine rises more slowly due to sequestering of cocaine by the vaccine, and produces a slow decline after this rise due to a slower decrease in cocaine due to release of cocaine from the vaccine producing a less aversive “high” than found in the placebo group. Bottom panel: In subjects with the low efficiency KOR-r AA protective allele, cocaine produces the rapid increase in

dopamine, but a less rapid decline in dopamine compared to the more efficient G allele carriers. The slower decline would be less aversive. After vaccine treatment the rise in dopamine levels is slower, but more importantly, the dopamine remains elevated longer.

Figure 2.

Percentage of cocaine positive urine toxicology screens for two-week time periods across the 16 week trial for the placebo versus cocaine vaccine treatment groups. A: Treatment response of subjects in the treatment group treated with the cocaine vaccine with the AA genotype (square symbols, dashed line, N = 20) and those with AG/GG genotypes (diamond symbols, solid lines, n = 13) are displayed. B: Treatment response of subjects in the placebo group with the AA genotype (square symbols, dashed line, N = 18) and those with AG/GG genotypes (diamond symbols, solid lines, n = 15) are shown. Standard error bars are shown at each time point.

Figure 3.

Percent of cocaine positive urines relative to baseline levels for the AA and AG/GG genotype groups treated with cocaine vaccine group versus the AG/GG genotypes treated with placebo group. Treatment response of subjects relative to their levels at baseline with the AA genotype (diamond symbols, solid line, N = 20) and the AG/GG genotypes (triangle symbols, solid line, N = 13) treated with the cocaine vaccine, and those subjects with the AG/GG genotypes treated with placebo (square symbols, dashed lines, n = 15) are displayed. Standard error bars are shown at each time point.