Supporting Information

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SI Results

Expression of GABA$_A$ Subunits in α2 $^{-/-}$ Mice. RT-PCR analysis of expression of other GABA$_A$ receptor subunits, including α1, α3, α4, α5, α6, β1, β2, and γ1 and γ2 in punches obtained from accumbens, indicated that expression of all of these subunits in the α2$^{-/-}$ mice did not differ significantly from WT mice (Fig S1).

Electrophysiology in Medium Spiny Neurons of α2$^{-/-}$ Mice. Deletion of the α2 subunit produced an ≈33% decrease in miniature inhibitory postsynaptic current (mIPSC) amplitude and a clear prolongation of the mIPSC decay (Fig. 2*A* and *B* and Table S1). The frequency of mIPSCs did not differ from that recorded for WT neurons (see Table S1). Thus, we confirmed that GABA$_A$ receptors containing the α2 subunit play a quantitatively important role in mediating GABA-induced currents in cell bodies of medium spiny neurons.

Conditioned Place Preference. Fig S2 shows the time spent in the vehicle-paired and cocaine-paired chambers in the post-conditioning minus the preconditioning test session. A significant chamber by treatment interaction ($F_{(1,44)} = 11.002$, $P < 0.01$) indicates a difference in time spent in the chambers according to treatment received. This preference is evident within cocaine treated groups [$t(23) = -3.055$, $P < 0.01$] but not vehicle treated [$t(23) = 1.361$, $P = 0.187$], confirming the presence of cocaine place preference. However, no effects of genotype were evident (nonsignificant genotype by treatment by chamber interaction, $F_{(1,44)} = 1.151$, $P = 0.289$; nonsignificant effect of genotype, $F_{(1,44)} = 0.234$, $P = 0.863$). Thus, deletion of the α2 subunit does not impair the ability to develop conditioned associations.

DOPAC/Dopamine Ratio in the Nucleus Accumbens of α2(H101R) Knock-In, and WT Mice Following Ro 15-4513. In accumbens, there were no main effects of genotype ($F_{(1,41)} = 0.97$, $P = 0.330$) or treatment ($F_{(1,41)} = 0.40$, $P = 0.530$), and no significant genotype by treatment interaction ($F_{(1,41)} = 2.74$, $P = 0.106$) on dopamine turnover as measured by dopamine/DOPAC ratios (Fig S3). There were also no significant differences between dopamine (no main effect of genotype, $F_{(1,41)} = 0.00$, $P = 0.979$, no main effect of treatment, $F_{(1,41)} = 0.20$, $P = 0.655$, nonsignificant genotype by treatment interaction, $F_{(1,41)} = 0.39$, $P = 0.536$) or DOPAC (no main effect of genotype, $F_{(1,41)} = 0.47$, $P = 0.496$, no main effect of treatment, $F_{(1,41)} = 1.37$, $P = 0.248$, nonsignificant genotype by treatment interaction, $F_{(1,41)} = 0.03$, $P = 0.875$) concentrations in the NAcc between vehicle and Ro 15-4513–treated WT and α2(H101R) mice (Fig S3).

SI Materials and Methods

Behavioral Studies. Cocaine hydrochloride and midazolam maleate, purchased from MacFarlan Smith and Sigma-Aldrich, respectively, were dissolved in sterile 0.9% saline. The midazolam solution was adjusted to a pH of 7 by addition of 0.1M sodium hydroxide. For i.p administration, Ro 15-4513 purchased from Sigma-Aldrich was suspended in a saline solution containing 0.2% Tween 80. Doses were corrected for the base. For intracranial infusion, Ro 15-4513 was suspended in 18% glycerol formal and 17% Solutol in saline.

Locomotor Activity. Locomotor activity was measured in circular runways (1). Each session began with a 30-min (α2$^{-/-}$ experiments) or 1-h (α2(H101R) experiments) habituation period, following which each animal was dosed with the assigned drug, and immediately returned to the runways for a further 1.5 h (2). During sensitization, mice received the assigned drug twice weekly, with intervals of 2 to 3 days.

Behavioral Sensitization to Cocaine in α2$^{-/-}$ Mice. ***Acute cocaine administration.*** Eight male WT and 8 male α2$^{-/-}$ mice were administered with saline, 3, 10, and 30 mg/kg cocaine in a Latin-square design before exposure to the circular runways.
Repeated, intermittent cocaine. Sixteen α2$^{-/-}$ and 18 WT mice were used. WT (n = 9) and α2$^{-/-}$ (n = 7) received repeated, intermittent treatment of 10-mg/kg cocaine, while the remainder were administered vehicle, over 25 sessions.

Conditioned Reinforcement in α2$^{-/-}$ Mice. Mice were trained in operant boxes for 15 × 1-h sessions with both levers retracted and the houselight off. Sweetened food pellets (20 mg; Noyes precision pellets, Formula P; Research Diets, Inc.) were delivered at random intervals (VT 120-s schedule) in conjunction with a conditioned stimulus (CS). The CS consisted of two flashing stimulus lights and a tone that commenced 5 s before food pellet delivery and continued for 5 s after. The stimulus lights were located above and to either side of the food magazine, with the tone generator directly above the magazine. The percentage of nose-pokes into the food magazine during the CS was recorded as a measure of discriminated approach.

After training, two levers were introduced into the operant boxes. Operation of one lever (counterbalanced across animals) resulted in illumination of the flashing cue lights and tone delivery for 1 s. Responses on the alternative lever had no programmed consequences. No food was delivered. The ability of the cues to act as conditioned reinforcers was assessed as higher rates of lever pressing on the cue-associated lever than on the alternative lever. At this stage, the mice were administered at 2-day intervals with cocaine (0, 3, 10 mg/kg) using a Latin-square design.

Intra-Accumbens Administration of Ro 15-4513. Eight female α2 (H101R) and 10 WT littermates were implanted stereotoxically under isofluorane anesthesia with guide cannulae (26 ga.) aimed at nucleus accumbens (coordinates AP1.34; L$^{+/-}$ 1.00; DV 3.20 in ref. 3). After 10 days recovery, the mice were sham-injected on 2 days by inserting injection cannulae (1-mm extension) into the guide cannulae, and placed in the locomotor apparatus for 120 min. Two days later, the mice were infused bilaterally with 0.5-μL vehicle, and at further 2-day intervals with Ro 15-4513 (15 and 45 nMol) and locomotor activity recorded.

Behavioral Sensitization Induced by Agonism at the Benzodiazepine Site of α2-Containing Receptors. Thirty-two WT and 32 α2 (H101R), 16 males and 16 females of each genotype, were used. Half of the male and female WT and α2(H101R) mice received repeated, intermittent Ro 15-4513 (10 mg/kg) treatment and the other half received vehicle. A total of 14 treatments were administered over a 7-week period. To establish whether behavioral sensitization was maintained after the cessation of treatment, the mice were treated with their assigned drug, vehicle or Ro 15-4513, following a 28-day drug-free period.

Dopamine Turnover in the Nucleus Accumbens Following Ro 15-4513 Treatment in WT and α2(H101R) Mice. To measure the effect of Ro 15-4513 on dopamine turnover in the accumbens, WT and α2 (H101R) mice, males and females, were dosed with vehicle or 10-mg/kg Ro 15-4513 (group size 7–12). Fifteen minutes later, mice were killed and accumbens dissected, immediately placed

on dry ice, and stored at −80 °C until assay by HPLC with electrochemical detection.

Place Preference Conditioning. All experiments were performed in a three-compartment place-conditioning apparatus (1). On day 1 (preconditioning day) mice (n = 12 per group) were placed into the apparatus and allowed to explore all compartments for a total of 20 min. Baseline preferences were recorded. On days 2 to 13, mice were injected i.p. with either cocaine (0, 10 mg/kg) or vehicle (10-mL/kg saline), immediately before placement in one compartment of the apparatus for 40 min. Conditioning days were alternated so that mice were placed in a different compartment each day, but the compartment paired with cocaine, and the days on which they were exposed to this compartment were counterbalanced. On day 14 (postconditioning day) mice were again given free access to all compartments for 20 min to determine postconditioning preferences.

Slice Preparation and Electrophysiology. Coronal slices containing the nucleus accumbens (300-μM thick) were prepared from WT and α2H101R mice [postnatal day 17 (P17) to P24] of either sex, as previously described (4). All slices were maintained at room temperature in a holding chamber filled with oxygenated extracellular solution (ECS) containing the following (in mM): 126 NaCl, 26 $NaHCO_3$, 2.95 KCl, 1.25 NaH_2PO_4, 2 $MgCl_2$, 2 $CaCl_2$, 10 glucose, (305-310 mOsm, pH 7.4). Slices were allowed to recover for a minimum of 1 h before obtaining electrophysiological recordings. Whole-cell patch-clamp recordings were obtained from visually identified neurons of the nucleus accumbens core at 35 °C, at a holding potential of −60 mV, using the ECS containing 1-μM strychnine, 2-mM kynurenic acid (Sigma-Aldrich), and 0.5-μM tetrodotoxin (TTX; Tocris Bioscience). Patch electrodes were prepared from thick-walled borosilicate glass (Garner Glass Company) and had an open-tip resistance between 3 and 5 MΩ when filled with an intracellular solution containing (in mM): 135 CsCl, 10 hepes, 10 EGTA, 1 $CaCl_2$, 2 $MgCl_2$, 2 Mg-ATP, 5 QX-314 (pH 7.2–7.3 with CsOH, 300–308 mOsm). Recordings were discarded if the series resistance changed (20% tolerance) during the course of the experiment. Currents were filtered at 2 kHz using an eight-pole low-pass Bessel filter and recorded onto digital audio tape using a DTR 1205 recorder for subsequent offline analysis.

Drug Application. Bicuculline methobromide and strychnine hydrochloride were prepared as aqueous stock solutions while Ro 15-4513 was dissolved in DMSO. Stock solutions were diluted in ECS to the desired final concentration. The final maximum DMSO concentration (0.1%) had no effect on the properties of the mIPSCs. All drugs tested were obtained from Sigma-Aldrich or Tocris Bioscience. Ro 15-4513 was allowed to infiltrate the slice for a minimum of 8 to 10 min before data acquisition.

Data Analysis. Recordings were digitized (NIDQMX interface card, National Instruments) and sampled at 10 kHz on a personal computer using the Strathclyde Electrophysiology Software, Electrophysiology Data Recorder / Whole Cell Analysis Program (WinEDR / WinWCP; J. Dempster, University of Dundee). mIPSCs were detected using an automated low amplitude (−4 pA, duration 1.5 ms) threshold detection algorithm and visually inspected for validity. Accepted events (at least 50 for each recording condition and with a rise time ≤ 1 ms) were analyzed with respect to their peak amplitude, rise time (10–90%) and their decay time course. A minimum of 50 mIPSCs were also digitally averaged by alignment at the midpoint of the rising phase and a least-squares minimization algorithm was used to determine the decay time constant of mIPSCs. The decay phase of individual mIPSCs was fitted (98–10% of the peak amplitude) by either a monoexponential [$y(t) = A.e^{(-t/\tau)}$] or biexponential [$y(t) = A_{fast}.e^{(-t/\tau fast)} + A_{slow}.e^{(-t/\tau slow)}$] function, where t is time, A is the amplitude, and τ is the decay time constant. Analysis of the SD of residuals and use of the F-test to compare goodness of fit revealed that the averaged mIPSC decay was always best fit with the sum of 2 exponential components. Thus, a weighted decay time constant (τ_w) was also calculated according to the equation: $\tau_w = \tau_1 P_1 + \tau_2 P_2$, where τ_1 and τ_2 are the decay time constants of the first and second exponential functions and P_1 and P_2 are the proportions of the synaptic current decay described by each component.

To determine the mIPSC frequency events were automatically detected using the EDR program on the basis of their rate of rise (30–50 pA ms^{-1}) and subsequently manually scrutinized to exclude spurious noise and include events that had failed to meet the trigger specifications.

All data are presented as the arithmetic mean ± SEM. When data are presented normalized, the mean value was calculated by averaging the normalized change for each cell following drug application. The statistical significance of mean data were assessed using the Student's t-test, one-way repeated measures ANOVA (RM ANOVA) as appropriate using the Sigma Stat (SPSS) software package. Statistical significance was set at $P < 0.05$. The nonparametric Kolmogorov-Smirnoff test (SPSS v12.0) was used to compare cumulative probability distributions. For a stringent comparison, statistical significance was set at $P < 0.01$ for the Kolmogorov-Smirnoff test.

Human Genetic Association Studies. ***Patients.*** For patients in this study, 699 cocaine abusers, 668 males and 31 females (mean age: 26.7 years; SD = 7.2), were ascertained (5). The study group consisted of drug users who were in treatment from August 1997 to October 1998 in one outpatient and six inpatient units located in the city of São Paulo, Brazil. Inclusion criteria were: age 18 years and older, a history of cocaine abuse, and under drug treatment at the selected centers. Individuals with another psychiatric diagnosis, such as psychosis, or a chronic physical illness, such as diabetes or other metabolic disorders, were excluded. All current cocaine users were then interviewed using a structured interview to collect data on sociodemographic characteristics, sexual behaviors, and drug-use profile. All subjects satisfied an ICD10 diagnosis of cocaine dependence (ICD-10: The ICD-10 Classification of Mental and Behavioral Disorders, World Health Organization, 1993). Blood samples were collected from all participants for genetic and other analyses. A total of 63.8% of the participants reported having smoked cocaine ("crack") over the previous month and 51.5% had snorted cocaine over the same period. The overall lifetime prevalence for heroin use in the sample was <5% (6).

Controls. For controls in this study, 866 healthy controls, 592 males, 274 females (mean age:31.7 years, SD = 9.9), were recruited from the Blood Transfusion Unit of the Hospital das Clinicas, Faculty of Medicine, University of São Paulo. Each blood donor was screened using a short questionnaire investigating contagious diseases and the use of any kind of drug. Subjects with a past history of drug abuse or with recent use of an illegal drug were excluded. During the act of donation, a short interview was conducted and subjects with a lifetime history of a psychiatric disorder requiring admission to hospital or suffering from a psychiatric condition at time of interview were excluded.

Ethics. All of the subjects included in this study gave written informed consent and this project was approved by the Ethical Committee of the Federal University of São Paulo and other relevant ethics committees.

Isolation of DNA. Genomic DNA was extracted using standard phenol/chloroform or modified salting-out method. The former was used for the preparation of the control samples and the latter for the patient samples. The concentration of extracted DNA was determined by spectrophotometric measurement. Five microliters of the extracted DNA was diluted 1:50 in 0.2× TE buffer. Ab-

sorption was measured for both blank (only 0.2× TE) and diluted DNA solutions at 260 nm using the GeneQuant (Amersham Pharmacia). An absorbance (A_{260}) of 1.0 corresponds to 50 ug of double stranded DNA per milliliter. After quantification, the DNA was diluted to a working level of 10 ng/uL.

Genotyping. All genotyping was performed under contract by Prevention Genetics (7). Allele-specific PCR assays were designed as described (8). These assays are based on competitive allele-specific PCR that allows the simultaneous amplification and detection of DNA within a closed reaction vessel. The homogeneous assay utilizes two different fluorescently labeled universal primers, two unlabeled and tailed allele-specific primers, and a common reverse primer in a single well reaction. Submicroliter PCR reactions were carried out with ArrayTape instrumentation and allele calls were generated based on the clustering of fluorescent signals (9, 10). All details of assays and primer sequences are available on request.

Statistical analysis. Genotype, haplotype, and allele frequencies were compared by using a χ^2 test, and *P*-values were assessed by using SPSS version 12.0, Haploview (11). The role of other clinical variables was tested with ANOVA or a nonparametric test as appropriate. GENECOUNTING Version 2.0 (12) and WHAP (13) were used to estimate global haplotype significance. Odds ratios and 95% CIs were derived from logistic regression.

Stratification analysis. ADMIXMAP uses ancestry informative genetic markers to model admixture in a population formed by two or more founding population (Table S4). If individual admixture is estimated, it is possible to investigate the relationship between admixture and disease risk and to control for the confounder—admixture proportions—in genetic associations studies by modeling its effect in the analysis (14). To adequately estimate admixture, a hierarchical model is required, where the distribution of individual admixture at the population level and the admixture of each individual are estimated. In this approach, the distribution of parental admixture proportions in the population, the admixture of each individual's parents, and the ancestry of the maternally and paternally derived alleles at each locus are unknown. Classical likelihood-based methods are unsuitable to apply when the majority of the data are missing and, thus, the alternative Bayesian approach is used to specify a full-probability model where all observed and missing data are treated as random variables and prior distributions are assigned when necessary. This model is then fitted using Markov chain–Monte Carlo simulation to generate the posterior distribution of all unobserved variables, given the observed genotypes and trait values. For large sample sizes and noninformative prior distributions, the posterior distributions of Bayesian analyses produce results that are asymptotically equivalent to the maximum-likelihood estimate. With the posterior probabilities for the unknown data generated, one can evaluate any null hypothesis of interest. This is performed by constructing a score test by averaging over the posterior distribution of the missing data, which for large samples is equivalent to the likelihood ration test, which can be conveniently used to estimate the effect of any variable of interest. In case-control designs, a regression model is specified for the dependence of the trait on the mean admixture proportions of both gametes, together with any other explanatory variables specified by the user (14), which included genotyping of 75 genetic variations selected for high variability between ethnic groups (SNPs and microsatellites selected for high Fst).

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Fig. S1. Expression levels of GABA(A) receptor subunits in accumbens of WT, α2$^{+/-}$ (heterozygote; HT), and α2$^{-/-}$ (knockout; KO) mice. Gene expression is shown as fold-change in expression from WT (+/– SEM). ANOVAs revealed an effect of genotype on expression of α2 [$P < 0.05$, $F(2,11) = 598$, as expected by gene dosage, ~50% expression in heterozygote and no detectable expression in the knockout mice]. Levels of expression of other subunits did not differ across genotype. Data for α6 are not presented as we were unable to detect this subunit in our accumbens samples.

Fig. S2. Changes in time spent in the vehicle- and drug-paired chambers (postconditioning minus preconditioning session), for KO and WT mice. Drug treatment was either cocaine (Coc) or its vehicle (Veh).

Fig. S3. Dopamine and DOPAC concentrations in the nucleus accumbens and prefrontal cortex, in α2(H101R) mice and their WT counterparts treated with vehicle or Ro 15-4513.

Table S1. Summary of the miniature inhibitory postsynaptic current properties of accumbens neurons of WT, α2(H101R), and $\alpha2^{-/-}$ mice

	WT (*n* = 19)	α2H101R (*n* = 11)	$\alpha2^{-/-}$ (*n* = 37)
Peak amplitude, pA	67 ± 4	70 ± 7	45 ± 3[a]
Rise time, ms	0.5 ± 0.1	0.5 ± 0.1	0.7 ± 0.1[a]
τ_w, ms	6.7 ± 0.3	6.5 ± 0.4	9.1 ± 0.5[a]
Charge transfer, fC	399 ± 30	422 ± 43	301 ± 24[a]
Frequency, Hz[b]	4 ± 0.5	3.7 ± 0.4	3.4 ± 0.4

[a] $P < 0.01$ vs. wild WT, unpaired Student's *t* test.
[b] $n = 17$ for the frequency analysis.

Table S2. Association of haplotypes with cocaine abuse in whole population and two Caucasian subsamples

Haplotype	Freq.	Case, control ratio counts	Frequencies: Case	Frequencies: Control	χ^2	*P* value
Total sample						
CTG	0.352	490.7: 913.3, 530.0: 968.0	0.349	0.354	0.06	0.8061
CTA	0.265	377.2: 1026.8, 391.0: 1107.0	0.269	0.261	0.219	0.6399
CAG	0.253	379.9: 1024.1, 354.5: 1143.5	0.271	0.237	4.418	0.0356
TTG	0.125	147.1: 1256.9, 215.2: 1282.8	0.105	0.144	10.018	0.0015
Ethnicity >70% European						
CTG	0.355	306.4: 539.6, 274.2: 513.8	0.362	0.348	0.361	0.548
CTA	0.261	237.5: 608.5, 188.5: 599.5	0.281	0.239	3.638	0.0565
CAG	0.255	209.3: 636.7, 206.6: 581.4	0.247	0.262	0.466	0.4948
TTG	0.126	90.3: 755.7, 115.6: 672.4	0.107	0.147	5.92	0.015
Ethnicity self-declared European						
CTG	0.351	348.7: 627.3, 262.5: 501.5	0.357	0.344	0.351	0.5534
CTA	0.265	269.5: 706.5, 190.8: 573.2	0.276	0.25	1.531	0.2159
CAG	0.254	252.8: 723.2, 189.5: 574.5	0.259	0.248	0.27	0.6032
TTG	0.126	101.1: 874.9, 117.8: 646.2	0.104	0.154	10.002	0.0016

Table S3. Association of SNPs with cocaine abuse

SNP	Allele	Cases %	Control %	Case number	Control numbers	χ^2	*P* value	Adjusted *P* value	Adjusted OR	95% CI
rs279871	A/G	43.6	38.3	581/751	534/862	8.123	0.0044	0.006	1.24	1.06–1.45
rs279845	A/T	43.2	38.2	586/770	549/887	7.181	0.0074	0.009	1.22	1.05–1.43
rs279836	A/T	44.9	41.2	610/750	601/859	3.91	0.048	0.060	1.16	0.99–1.34
rs279841	A/G	41.4	37.9	542/768	540/886	3.51	0.061	0.074	1.15	0.99–1.35
rs189957	A/G	50.7	46.1	680/662	668/780	5.744	0.0165	0.020	1.20	1.03–1.39
rs1442059	C/T	42.3	39.3	558/762	556/858	2.463	0.1165	0.136	1.13	0.96–1.31
rs9291283	A/G	27.3	26.3	327/871	365/1025	0.353	0.5525	0.393	1.03	0.97–1.09
rs2119767	A/T	27.7	23.9	377/985	348/1108	5.259	0.0218	0.025	1.22	1.03–1.44
rs894269	C/T	89.7	85.5	1207/139	1236/210	11.221	0.0008	0.001	1.48	1.17–1.86
rs894269	T/C	10.3	14.5	139/1207	210/1236	11.221	0.0008	0.001	0.67	0.95–0.54

P values are given for association adjusted for ethnicity, age, and sex as well as unadjusted values.

Table S4. Ancestry informative markers used stratification analyses and ancestry estimations by ADMIXMAP

Marker information				Allele frequency			Allele frequency difference, Δ			Information content, *f*		
Name	Chrom	Alleles	Genetic position, M	European	African	Native American	European/ African	European/ Native American	African/ Native American	European/ African	European/ Native American	African/ Native American
rs2814778	1	2	164.8	1.00	0.00	0.98	1.00	0.02	0.98	0.99	0.01	0.96
rs10752631	1	2	166.1	0.65	0.16	0.93	0.49	0.28	0.77	0.25	0.12	0.60
rs905595	1	2	169.8	0.76	0.19	0.00	0.57	0.76	0.19	0.32	0.61	0.11
rs2340727	1	2	169.8	0.85	0.11	0.97	0.74	0.11	0.86	0.55	0.04	0.74
rs12737539	1	2	169.8	0.64	0.05	0.43	0.59	0.21	0.38	0.39	0.04	0.20
rs2806424	1	2	171.3	0.71	0.16	0.31	0.55	0.40	0.15	0.31	0.16	0.03
rs1780349	1	2	173.9	1.00	0.53	1.00	0.47	0.00	0.47	0.31	0.00	0.31
rs723822	1	2	175.8	0.88	0.79	0.13	0.09	0.75	0.66	0.02	0.56	0.43
rs6003	1	2	212.0	0.90	0.25	1.00	0.65	0.10	0.75	0.43	0.05	0.60
rs1008984	1	2	212.7	0.88	0.31	0.28	0.57	0.61	0.03	0.34	0.38	0.00
rs1506069	1	2	245.9	0.93	0.06	0.63	0.87	0.29	0.58	0.76	0.13	0.37
rs2752	1	2	265.1	0.51	0.20	0.35	0.31	0.17	0.15	0.11	0.03	0.03
rs1861498	2	2	18.2	0.83	0.10	0.99	0.73	0.16	0.89	0.53	0.08	0.80
rs1435090	2	2	76.0	0.75	0.79	0.15	0.04	0.60	0.64	0.00	0.36	0.41
rs3287	2	2	79.3	0.81	0.32	0.73	0.49	0.08	0.41	0.24	0.01	0.17
rs1344870	3	2	41.5	0.99	0.93	0.10	0.06	0.89	0.83	0.02	0.79	0.69
rs17203	3	2	106.5	0.83	0.22	0.24	0.61	0.59	0.02	0.37	0.35	0.00
rs768324	3	2	118.4	0.90	0.80	0.10	0.11	0.81	0.70	0.02	0.65	0.49
rs1465648	3	2	119.7	0.73	0.12	0.92	0.60	0.20	0.80	0.37	0.07	0.63
D3S3045	3	9	124.2	0.28	0.26	0.00	0.06	0.10	0.10	0.14	0.66	0.68
rs938431	3	2	130.6	0.80	0.61	0.06	0.19	0.74	0.55	0.04	0.56	0.33
rs1316579	3	2	193.9	0.69	0.27	0.06	0.42	0.63	0.21	0.18	0.42	0.08
rs719776	4	2	55.6	0.89	0.13	0.86	0.76	0.03	0.73	0.57	0.00	0.53
D4S2623	4	14	114.0	0.53	0.23	0.51	0.05	0.03	0.06	0.15	0.12	0.28
rs1112828	4	2	149.0	0.79	0.95	0.12	0.16	0.67	0.82	0.05	0.45	0.68
rs1403454	4	2	163.5	0.88	1.00	0.13	0.12	0.76	0.88	0.06	0.57	0.78
rs3309	5	2	70.5	0.72	0.60	0.25	0.11	0.47	0.35	0.01	0.22	0.13
D5S1501	5	25	85.3	0.22	0.10	0.13	0.04	0.05	0.05	0.31	0.45	0.41
rs1461227	5	2	170.6	0.89	0.59	0.19	0.30	0.70	0.40	0.11	0.49	0.17
rs3340	5	2	171.3	0.81	0.93	0.33	0.12	0.48	0.60	0.03	0.24	0.39
rs2077681	6	2	7.4	0.95	0.17	0.79	0.79	0.17	0.62	0.63	0.06	0.39
rs1935946	6	2	137.2	0.58	0.19	0.02	0.39	0.56	0.17	0.16	0.38	0.08
rs2763	7	2	2.0	0.82	0.91	0.57	0.09	0.25	0.34	0.02	0.07	0.15
rs2161	7	2	110.1	0.65	0.44	0.30	0.21	0.36	0.15	0.04	0.13	0.02
rs2396676	7	2	132.4	0.79	0.13	0.58	0.66	0.21	0.45	0.44	0.05	0.23
rs2341823	7	2	148.3	0.90	0.11	0.49	0.79	0.41	0.38	0.63	0.20	0.18
rs1320892	7	2	164.6	0.77	0.89	0.10	0.12	0.67	0.78	0.02	0.45	0.61
D8S560	8	19	42.2	0.17	0.00	0.06	0.08	0.05	0.11	0.72	0.26	1.00
rs1373302	8	2	101.8	0.73	0.63	0.08	0.10	0.65	0.55	0.01	0.44	0.32
rs1808089	8	2	106.2	0.57	0.62	0.04	0.05	0.54	0.59	0.00	0.34	0.39
rs1987956	8	2	134.4	0.96	0.92	0.21	0.04	0.75	0.72	0.01	0.58	0.52
rs1980888	9	2	113.0	0.91	0.80	0.05	0.11	0.86	0.75	0.03	0.74	0.57
rs1327805	9	2	158.0	0.84	0.38	0.09	0.46	0.75	0.30	0.22	0.57	0.12
rs1594335	10	2	97.0	0.74	0.20	0.76	0.54	0.02	0.56	0.29	0.00	0.31
rs2207782	10	2	112.5	0.69	0.10	0.05	0.58	0.64	0.06	0.36	0.44	0.01
rs1891760	10	2	140.1	0.60	0.81	0.06	0.21	0.54	0.74	0.05	0.33	0.56
rs1487214	11	2	36.0	0.94	0.14	0.84	0.80	0.10	0.70	0.65	0.03	0.49
rs594689	11	2	86.5	0.56	0.89	0.84	0.33	0.28	0.05	0.14	0.09	0.01
rs1042602	11	2	112.3	0.61	1.00	0.96	0.39	0.35	0.03	0.24	0.19	0.01
rs5443	12	2	18.1	0.59	0.22	0.66	0.37	0.07	0.44	0.14	0.01	0.20
rs726391	12	2	36.7	0.77	0.05	0.40	0.73	0.38	0.35	0.55	0.15	0.18
rs2078588	13	2	74.4	0.95	0.02	0.86	0.93	0.09	0.84	0.86	0.02	0.71
D13S285	13	16	112.8	0.37	0.08	0.71	0.05	0.05	0.09	0.24	0.21	0.59
rs1153849	15	2	40.5	0.75	0.11	0.00	0.65	0.75	0.10	0.43	0.60	0.05
rs4646	15	2	46.5	0.71	0.68	0.28	0.02	0.43	0.40	0.00	0.18	0.16
rs2351254	15	2	97.1	0.93	0.40	0.21	0.53	0.72	0.19	0.32	0.54	0.04
rs764679	16	2	26.0	0.97	0.80	0.27	0.17	0.70	0.53	0.07	0.52	0.28
rs2816	17	2	19.2	0.53	0.00	0.04	0.53	0.49	0.04	0.35	0.30	0.02
rs1074075	17	2	94.0	0.85	0.09	0.94	0.76	0.08	0.85	0.58	0.02	0.72

Table S4. Cont.

Marker information				Allele frequency			Allele frequency difference, Δ			Information content, *f*		
Name	Chrom	Alleles	Genetic position, M	European	African	Native American	European/ African	European/ Native American	African/ Native American	European/ African	European/ Native American	African/ Native American
rs717962	17	2	104.6	0.78	0.88	0.19	0.10	0.59	0.69	0.02	0.34	0.48
rs1369290	18	2	106.4	0.94	0.09	1.00	0.86	0.05	0.91	0.73	0.03	0.83
rs386569	19	2	81.5	0.75	0.85	0.06	0.10	0.70	0.80	0.02	0.50	0.64
rs11467165	20	2	54.8	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
rs718092	20	2	59.7	0.85	0.25	0.18	0.60	0.67	0.06	0.37	0.45	0.01
D21S2055	21	25	40.5	0.29	0.00	0.00	0.06	0.05	0.07	0.60	1.00	0.77
rs878825	22	2	17.0	0.91	0.56	0.14	0.35	0.77	0.43	0.15	0.60	0.20
rs16383	22	2	54.0	0.78	0.23	0.08	0.55	0.70	0.15	0.30	0.50	0.04
D22S1169	22	10	60.6	0.33	0.15	0.02	0.09	0.12	0.10	0.23	0.43	0.30
rs1986586	X	2	122.6	1.00	0.43	0.00	0.57	1.00	0.43	0.40	1.00	0.28
rs2188457	X	2	141.8	0.84	0.28	0.95	0.56	0.11	0.66	0.31	0.03	0.46
rs1415878	X	2	143.8	0.84	0.99	0.06	0.16	0.77	0.93	0.08	0.60	0.87

The haplotypes identified in our sample were similar in frequency to those predicted by combining the European, African, and Asian (used as a proxy for Native Americans) haplotype estimates from HAPMAP in ≈70%, 20%, and 10% proportions.