Supplementary Methods

Subjects abstained from caffeine and nicotine for 4 hours prior to the scanning session. Further subject-specific information is summarized in supplementary table 1. Genotype groups were carefully matched for age and gender (met-homozygotes: 27.8 ± 5.5 years, 7 males, 6 females; val-allele carriers: 29.3 ± 6.2, 7 males, 4 females), and no significant differences between genotype groups were found for any other demographic variable. We did not observe an effect of age on any measured variable in this study with the exception of reaction time, which increased significantly for both the control and working memory condition, but did not differ between genotypes (supplementary table 1). Reaction times were shorter in 2- than 0-back because subjects are able to prepare their motor response in advance according to the information held in working memory.

The behavioral paradigm and imaging procedures were as described previously. Each subject participated in two PET imaging sessions on a GE Advance scanner (septa retracted, 4.25 mm slice separation, 35 slices, axial FOX 15.3 cm). During the rCBF session, a version of the “n-back” working memory task was given after injection of 10 mCi H$_{2}^{15}$O per scan. For this, the numerals 1,2,3 or 4 were presented one every 1.8 seconds in a diamond-shaped array via computer for a total of 90 seconds, starting 15 seconds prior to tracer injection. Subjects held a button-box with four buttons arranged similarly to to the visual display. In the 0-back (control) condition, the button corresponding to the current number was pressed. In the 2-back (working-memory) condition, the button to press corresponded to the number seen 2 presentations before. Percentage of correct responses was used as the performance measure, and reaction times were also recorded. We did not observe an effect of age on any measured variable in this study with the exception of reaction time, which increased
significantly for both the control and working memory condition, but did not differ between genotypes (supplementary table 1).

In the other imaging session, to measure 6-FDOPA uptake, subjects were pretreated with 200 mg of Carbidopa to reduce peripheral metabolism of 6-FDOPA and increase tracer availability in the brain. Two resting rCBF scans, using the same settings as described above, were acquired in each session for coregistration. Then, 13.7 ± 3.5 (mean ± S.D.) mCi of 6-FDOPA were infused over 90 seconds, and images were acquired from the time of infusion up to 90 minutes later (25 images total).

All images were attenuation-corrected and reconstructed (32 planes, 6.5 mm FWHM). Further processing used SPM99 software (Wellcome Department of Cognitive Neurology) as described previously\textsuperscript{11}. After subtraction of background activity and registration, rCBF scans were normalized to an average template, scaled proportionally to a whole brain mean of 50 and smoothed (10 mm\textsuperscript{3} FWHM Gaussian kernel). Activations and deactivations in the 2-back relative to the 0-back task are shown in supplementary figure 3 and coordinates are given in tables 4 and 5. 6-\textsuperscript{18}F-DOPA data were aligned in-plane and registered, coregistered to the rCBF scans, and affine normalized. The kinetic rate constant K\textsubscript{i} for dopaminergic uptake was calculated voxel-by-voxel using a linear fit based on the Patlak method, using the time activity curve in an occipital reference region as the input function\textsuperscript{11}.

6-FDOPA uptake in the midbrain was quantified as the average within a template derived in normalized space from a publicly available probabilistic brain atlas (International Consortium for Brain Mapping\textsuperscript{16}).

To study the interaction of midbrain 6-FDOPA and prefrontal blood flow, both region of interest (ROI) and voxelwise approaches were used. According to the a-priori hypothesis,
a functional ROI was prepared from the areas in prefrontal cortex significantly ($P<0.01$, corrected by voxel family-wise error) activated during the working memory task as previously described. The rCBF during 0-back and 2-back, as well as the activation (2-back minus 0-back) in this region was then correlated with the $K_i$. Spearman’s $r$ was used for correlations, and the Williams-Pearson test was used to assess whether correlations differed significantly between genotype groups. To confirm these findings and search for other brain areas showing similar dependencies, midbrain 6-FDOPA was used as covariate, separately by genotype, and correlated with blood flow during 0-back across the entire brain. An F-test was used to identify any voxels correlated with midbrain dopamine; the specific dependence on genotype was then investigated using the appropriate contrasts. The resulting correlation maps assessed for significance using Gaussian random fields theory to control for multiple comparisons ($P<0.05$, corrected on the cluster level).

For post-hoc analyses, a whole striatum ROI was derived and analyzed as described for midbrain. Multiple regression was used to assess the relative contribution of prefrontal 0-back rCBF and midbrain dopamine to prefrontal activation.

Coordinates of statistically significant brain activations and correlations are reported according to the system described in the Talairach-Tournoux atlas.
Supplementary fig. 1. Relationship between dopaminergic stimulation and prefrontal cortex activity, drawn schematically after Goldman-Rakic et al.\textsuperscript{18} and Mattay et al.\textsuperscript{9} In the latter study, the differential effect of an acute increase in dopaminergic tone induced by amphetamine was used to probe the position on the u-shaped curve in the setting of acute pharmacological modulation, which led to improved function and prefrontal efficiency in val-allele carriers, but deteriorating function in met-homozygotes.

Supplementary figure 2: Task-related activations and deactivations. Effect of task - significant activations (red) and deactivations (blue), comparing the working memory (2-back) condition with its sensorimotor control (0-back). Highlighted voxels are significant at the $P<0.05$ corrected level (see supplementary tables 1 and 2).

Supplementary figure 3: Hypothetical fit of data to “inverted-u” response curve. “Inverted-u” shaped relationship between observed left DLPFC 0-back rCBF and dopamine synthesis rate, hypothetically assuming that a given rate of midbrain dopamine neuronal activity and dopamine synthesis ($K_c$) will result in twice as much prefrontal dopamine in met-homozygotes (see discussion in Chen et al. 2004); to reflect this, $K_c$s were doubled for met-homozygotes. Second-order polynomial fit curve shown. Datapoints for val-carriers shown as filled circles, met-homozygotes as empty circles.
References for supplementary section

