Supporting Information

Use of cysteine-reactive crosslinkers to probe conformational flexibility of human DJ-1 demonstrates that Glu18 mutations are dimers

Janani Prahlad, David N. Hauser, Nicole M. Milkovic, Mark R. Cookson and Mark A. Wilson



Figure S1: Thermal stabilities of reduced, C106-SO₂-oxidized, and C106S DJ-1.

Differential scanning fluorimetry was used to determine the thermal stabilities of wildtype reduced DJ-1 (WT Red), wild-type Cys106-SO₂⁻ DJ-1 (WT Ox), and C106S DJ-1 (C106S). Melting temperatures (T_m) are reported as the maximum value of the first derivative of the fluorescence as a function of temperature (dF/dT; Y-axis). Oxidation to Cys106-SO₂⁻ increases the T_m of wild-type DJ-1 from 64° C to 76° C, as previously reported. The C106S mutant has a T_m value of 72° C, stabilizing the protein by ~6° C compared to wild-type.



Figure S2: Western blots of recombinant DJ-1 crosslinking by BMOE.

Recombinant wild type DJ-1 and mutants were crosslinked with BMOE and then analyzed by Western blot using antibodies specific for epitopes near the N-terminus (Abcam ab76008; Panel A) or C-terminus (Santa Crux sc-27006; Panel B) of DJ-1. Since the primary antibodies were raised in different host species, they were used simultaneously and detected on the same blot using different IR-conjugated secondary antibodies.