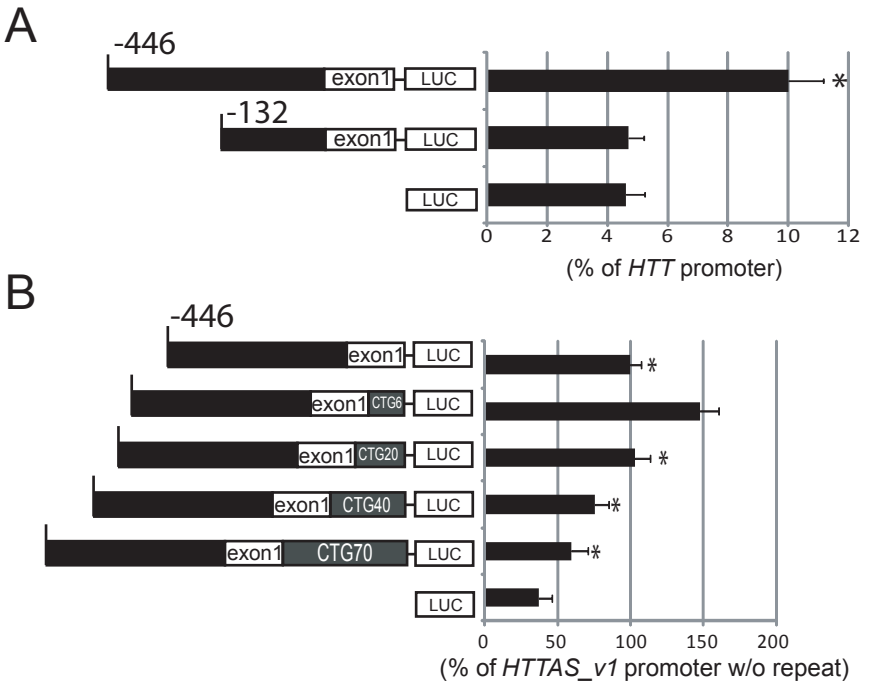
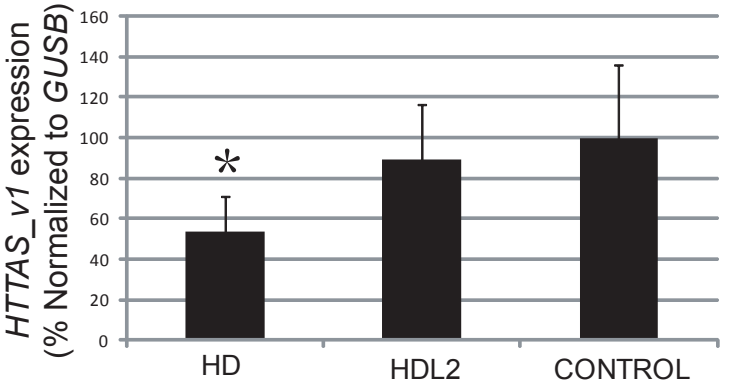
Supplemental Figure 1. HTTAS_v1 promoter activity in SHSY5Y cells.



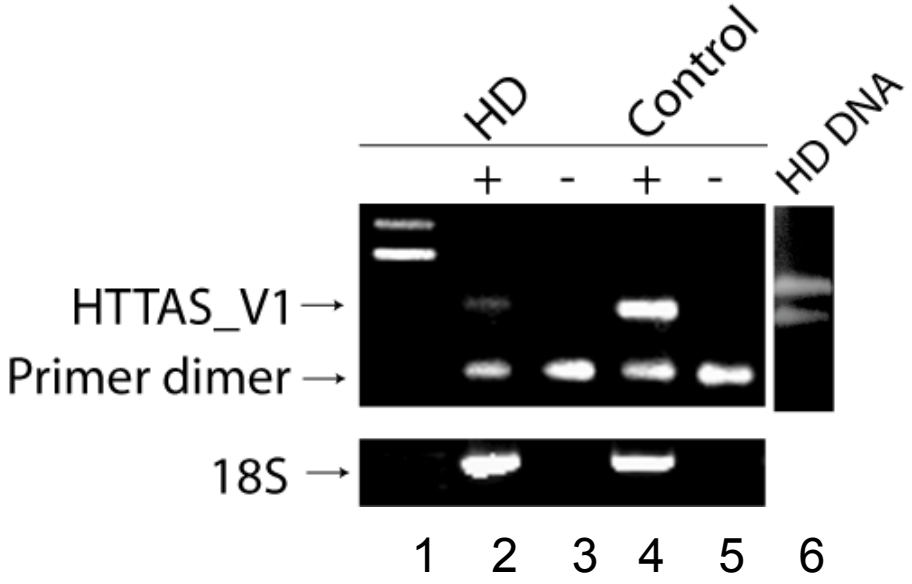
Supp Fig. 1. HTTAS_v1 promoter activity in SH-SY5Y cells (see Figure 2 for HEK293 cells). A. Promoter activity is reduced by eliminating bp -446 to -133. ANOVA n=5, F=74.55, P<0.001. Scheffe *P<0.05 -466 construct vs. -132 construct or vector. Performed three times with similar results. Error bars represent s.d. B. Promoter activity is increased by a short repeat, but is reduced by repeat expansion, in SH-SY5Y cells. ANOVA n=4, F=50.034, P<0.001. Scheffe *P<0.05 versus HTTAS1 promoter with CAG6. The experiment was performed three times in quadruplicate with similar results.

Supplemental Figure 2. HTTAS_v1 levels in HD vs HDL2 vs control cortex.



Supp Fig. 2. HD, but not HDL2, is reduced in HD frontal cortex. HTTAS_v1 expression is 50% reduced in HD frontal cortex (N = 8) compared to age matched control frontal cortex (N = 8) and available HDL2 brains (N = 4). Each sample was assayed in triplicate. ANOVA, $F=4.534$, $P<0.05$. Tukey *$P<0.05$. HD was compared to control frontal cortex in three additional experiments with independent RNA extractions, all demonstrating ~50% reduction of HTTAS_v1. Error bars represent s.d.

Supplemental Figure 3. HTTAS_v1 expression CsCl extraction.



Supp Fig 3. HTTAS_v1 allele expression in HD compared to control brain, CsCl extraction. RNA was extracted from frontal cortex of HD and control brain samples. HTTAS_v1 was detected using PCR and ethidium bromide staining as described in the text. DNA was extracted from HD tissue using standard protocols, and HD repeat length detected as described in the text. Lanes: 1—size marker. 2—HD frontal cortex. 3—HD frontal cortex, no RT. 4—control frontal cortex. 5—control frontal cortex, no RT. 6—HD genomic DNA, assayed for repeat length. Note: normal and expanded allele lengths detected in genomic DNA (lane 6), absence of HTTAS_v1 expanded allele in HD brain (second lane), qualitative reduction in HTTAS_v1 in HD vs. control, consistent with loss of expression of allele with expanded repeat.

Supplemental Figure 4. ***HTTAS_v1* is expressed from both alleles in individuals without HD.**

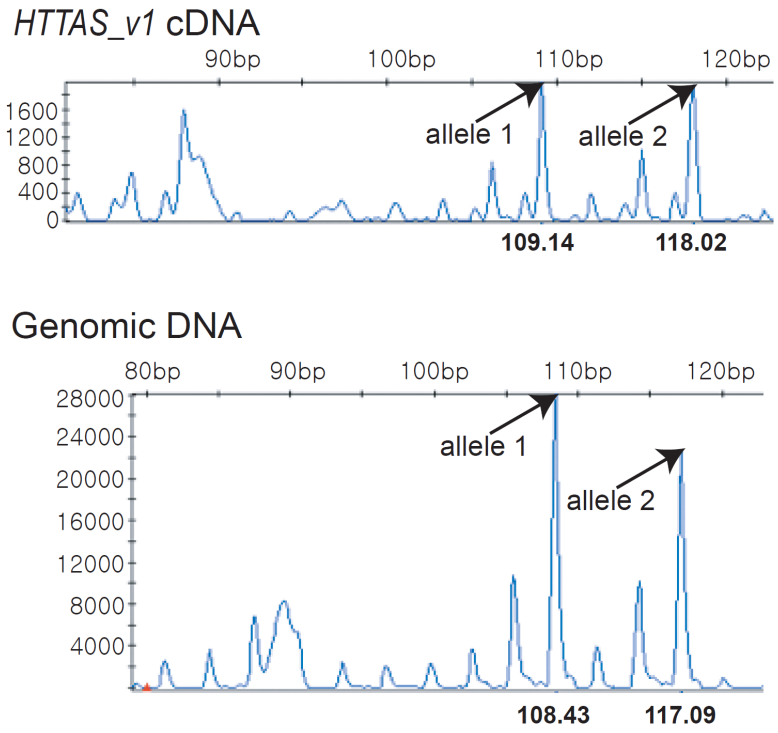
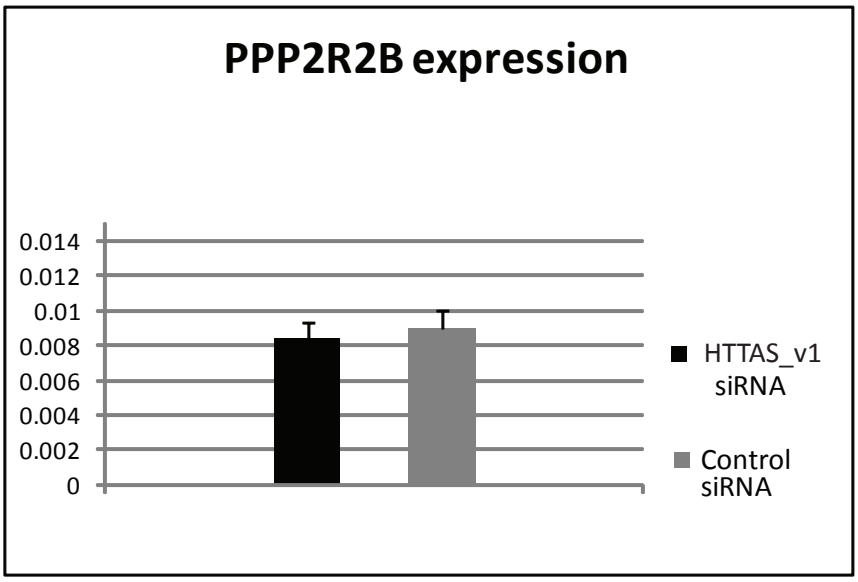
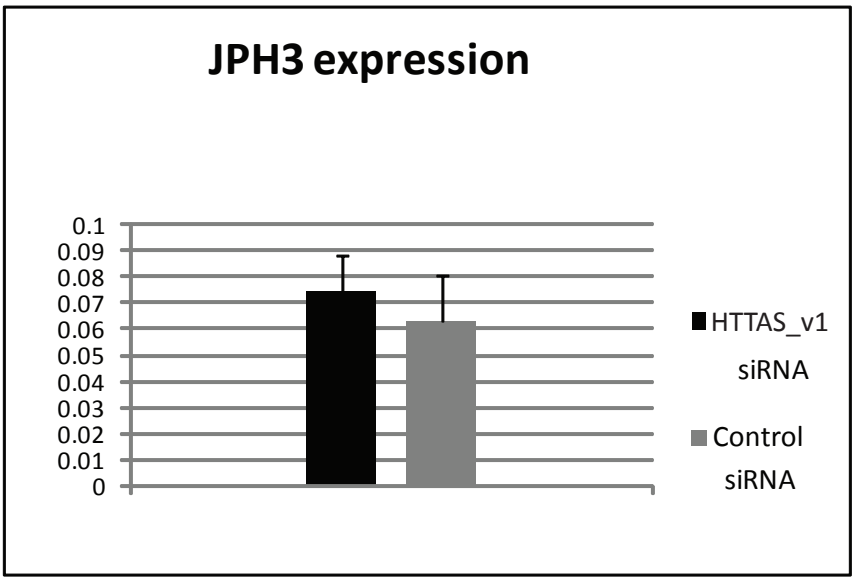


Figure 4. *HTTAS_v1* is expressed from both alleles in individuals without HD. PCR with a fluorescently labeled primer show alleles of identical length from cDNA and genomic DNA from control case 3712. The 1 bp difference in size between genomic DNA and cDNA reflects a single base pair difference in the amplicon length, from a one base difference in primer pairs. Similar findings were obtained in a second case.

Supplemental Figure 5. siRNA against HTTAS_v1 does not significantly effect PPP2R2B or JPH3 expression.





Supp Fig. 5. siRNA against *HTTAS_v1* does not significantly alter levels of *PPP2R2B* (associated with SCA12) or *JPH3* (associated with HDL2).
Transfection of pooled siRNA into HEK293 cells with assay of PPP2R2B or JPH3 by qPCR, normalized to *GUSB*. PPP2R2B: ABI probe Hs00270227_m1; vs control T = -0.813, p - .46. JPH3: ABI probe Hs00931502_m1 for exon 3_4 junction, vs. control T = 0.876, p = .43.