

Supplemental Data

Smith-Magenis Syndrome Results in Disruption of *CLOCK*

Gene Transcription and Reveals an Integral Role for

RAI1 in the Maintenance of Circadian Rhythmicity

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Table S1. Primers for Mutagenesis of *CLOCK* Constructs

Primer	Sequence	Restriction Site Introduced	Constructs
p53	gcactttagtgactcttcttcagaaaggtaccgctcaaatatcaacaaaa	<i>KpnI</i>	<i>CLOCK</i> ^{Del1} and <i>CLOCK</i> ^{Del6}
p53R	ttttgtgatattgagcggtagccttctgaaagaagagtcactaaagtc	<i>KpnI</i>	<i>CLOCK</i> ^{Del1} and <i>CLOCK</i> ^{Del6}
p800	ccattaatgcacagccttcccatattcagagctcccaccagagtg	<i>SacI</i>	<i>CLOCK</i> ^{Del2}
p800R	ccactctggtggggagctctgaatatggggaaggctgtgcattaatgg	<i>SacI</i>	<i>CLOCK</i> ^{Del2}
p798	cacataaaaaactgagggttaaggtaccgagatttcccattgactcca	<i>KpnI</i>	<i>CLOCK</i> ^{Del4}
p798R	tggagtcaatgggaaatctcggtagccttaccctcagtttttatctg	<i>KpnI</i>	<i>CLOCK</i> ^{Del4}
p802	atatcaacaaaaaaagactttagaacattaatttggtaccttagcttttctaatttaatac	<i>KpnI</i>	<i>CLOCK</i> ^{Del5} and <i>CLOCK</i> ^{Del3}
p802R	gtattaaattagaaaacgtaaaggtaccaaatatgttctaaagtctttttgttgat	<i>KpnI</i>	<i>CLOCK</i> ^{Del5} and <i>CLOCK</i> ^{Del3}

Supplemental Methods

Site directed mutagenesis (QuikChange II Kit, Stratagene, Inc.) was used according to manufacturer instructions with specific primers to create either a *SacI* or a *KpnI* restriction enzyme site within the appropriate *CLOCK*^{Luc} construct (bp 1-721). A *KpnI* site was introduced at base pair 375 in the *CLOCK*^{Luc} sequence, and the resulting plasmid was digested with *KpnI*. The remaining *CLOCK*^{Luc} plasmid was ligated back together to create *CLOCK*^{Del1} (bp 375-721), and the fragment that was removed was ligated into a new pGL3 vector to create *CLOCK*^{Del6} (bp 1-374). A *SacI* site was introduced into *CLOCK*^{Del1} at base pair 533. Digestion and ligation of the resulting construct created *CLOCK*^{Del2} (bp 375-533). *CLOCK*^{Del4} (bp 423-533) and *CLOCK*^{Del5} (bp 483-533) were created by introducing a *KpnI* site in plasmid *CLOCK*^{Del2} at positions 423 and 483, respectively, and performing the digestion and ligation procedure. Finally, the fragment removed from *CLOCK*^{Del2} to create *CLOCK*^{Del5} was ligated into a new pGL3 vector to create *CLOCK*^{Del3} (bp 375-483).