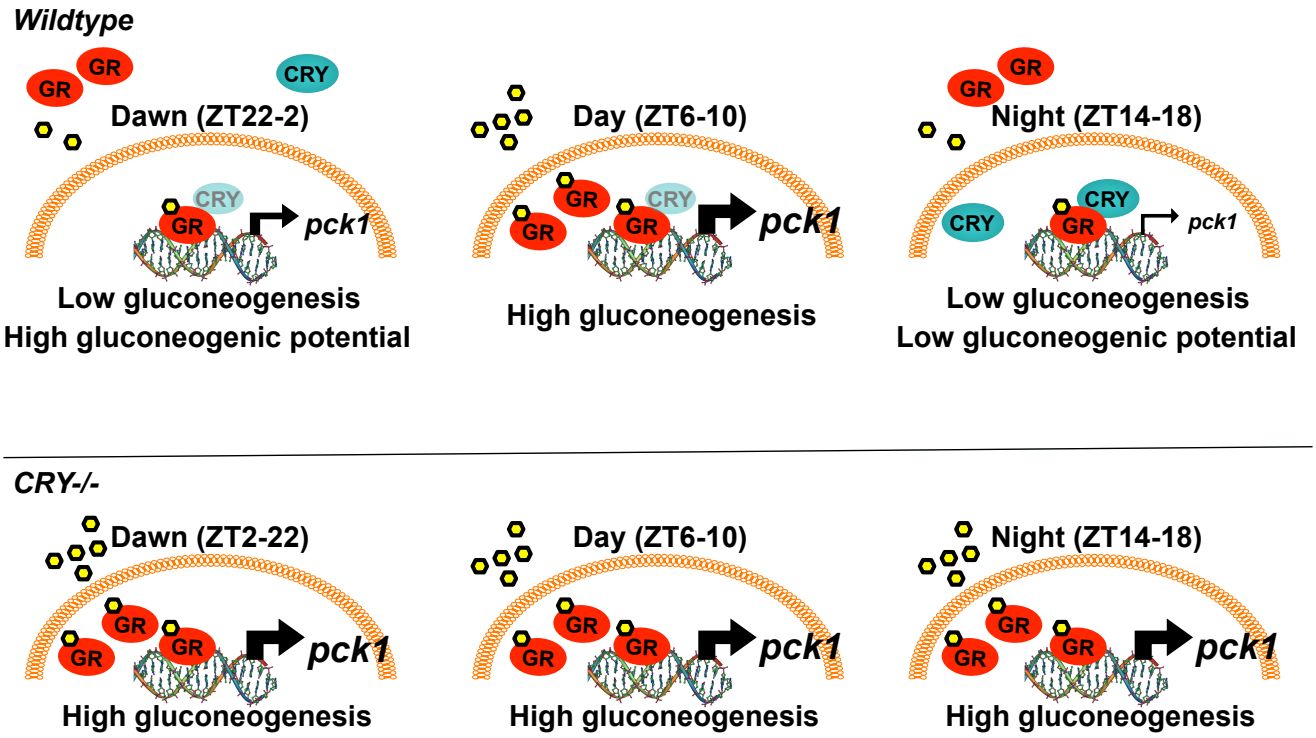

Figure S1: Model for the circadian regulation of glucocorticoid signalling and gluconeogenesis based on this work. The circadian transcriptional repressors Cry1 and Cry2 interact with and repress ligand-bound GR on chromatin. In the liver, this mechanism contributes to the diurnal regulation of gluconeogenesis via repression of *pck1*. Under normal conditions, diurnal expression of pck1 is modulated by diurnal rhythm of corticosterone as well as diurnal expression and nuclear localization of cryptochromes. Genetic loss of cryptochromes results in excessive gluconeogenesis due in part to constitutively elevated circulating corticosterone and increased GR activation of pck1, which is exacerbated by treatment with synthetic glucocorticoids.

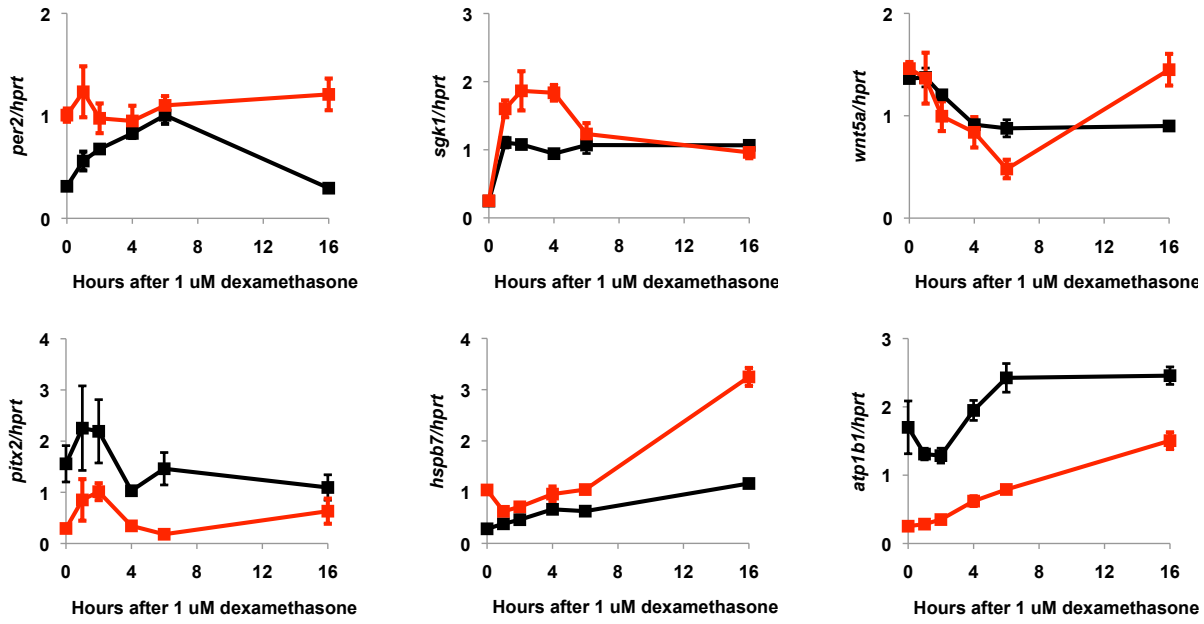

Figure S2: Loss of cryptochromes leads to a positive shift in the dynamic transcriptional response to dexamethasone. Expression of the indicated transcripts was measured in cDNA prepared from control and cry1-/-;cry2-/- MEFs by quantitative PCR following treatment with 1 mM dexamethasone for the indicated times. Data represent the mean ± s.e.m. of triplicate samples analyzed in triplicate.

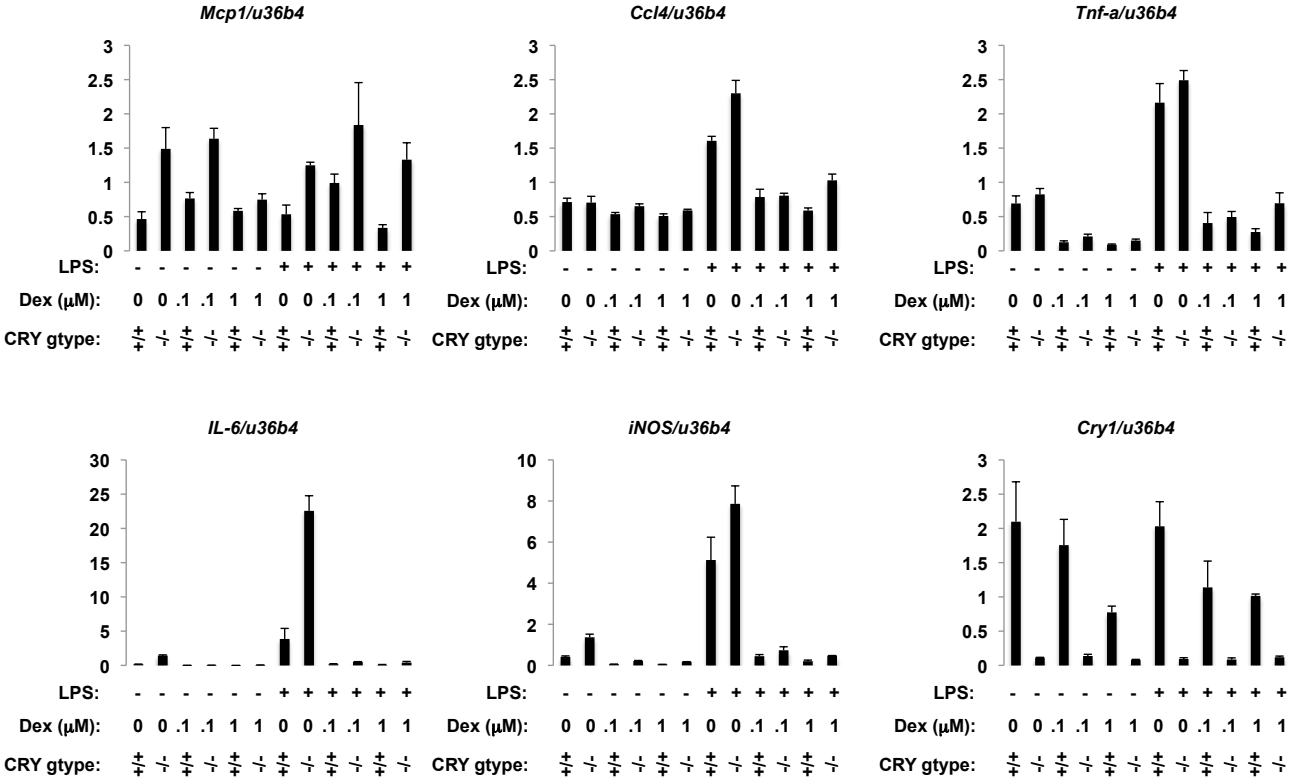

Figure S3: Cryptochromes are not required for GR-dependent repression of inflammatory genes in macrophages. Expression of indicated transcripts was measured in cDNA prepared from wildtype and cry1-/-;cry2-/- primary cultured bone marrow macrophages by quantitative PCR following treatment with vehicle or dexamethasone for 16 hours followed by LPS stimulation for 6 hours.

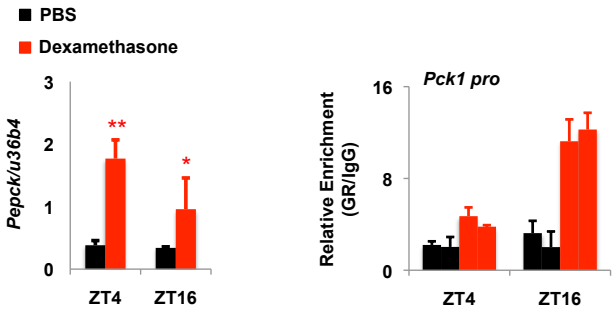

Figure S4: Induction of Pck1 is higher during the day. Left, expression of pck1 measured by quantitative PCR using cDNA prepared from wildtype mouse livers dissected at ZT4 or ZT16. * P < 0.05, **P < 0.01 relative to saline-treated. Right, recovery of chromatin containing the mouse Pck1 proximal promoter GRE from chemically cross-linked mouse liver nuclei following IP of GR.

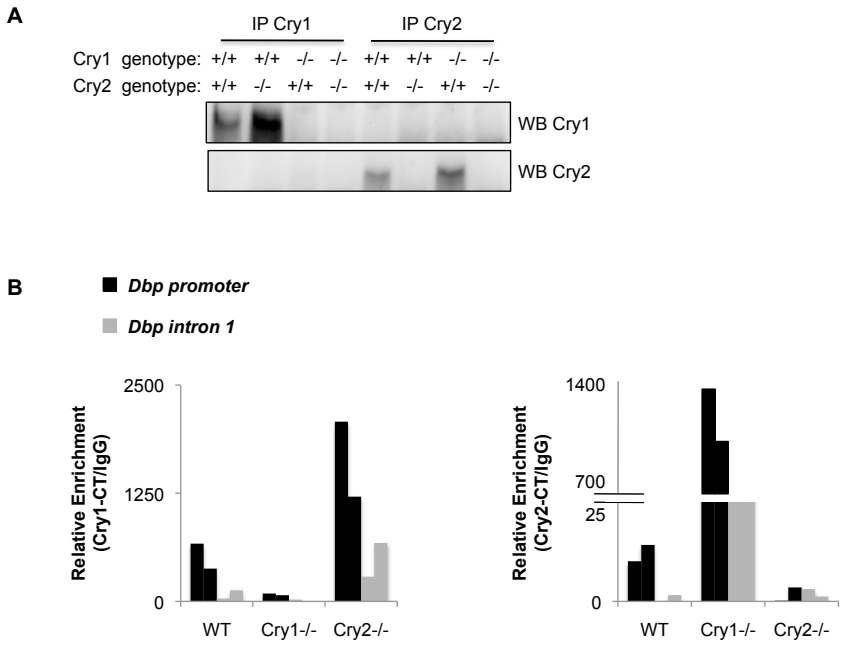

Figure S5: Validation of Cry1-CT and Cry2-CT antibodies. (A) Immunoblotting of Cry1 and Cry2 using purified anti-Cry1-CT or anti-Cry2-CT polyclonal antibodies in immunoprecipitates from mouse livers of the indicated genotypes. (B) Recovery of the Dbp promoter which has been shown to interact with Cry129 from wildtype, cry1-/- or cry2-/- mouse liver nuclei prepared at ZT2 by chromatin immunoprecipitation using anti-Cry1-CT or anti-Cry2-CT antibodies.

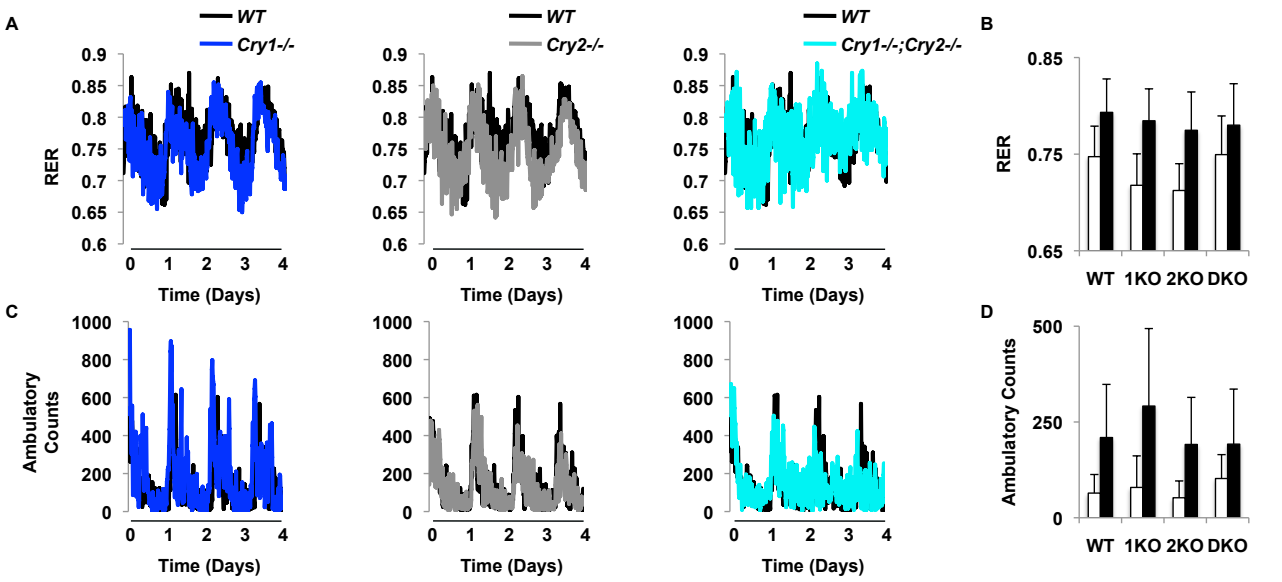

Figure S6: Genetic disruption of cryptochromes does not significantly alter behavior under light:dark conditions. (A) Continuous monitoring of respiratory exchange ratio (RER) in wildtype, cry1-/-, cry2-/- and cry1-/-;cry2-/- male mice maintained in 12 hours light:12 hours dark conditions for four days. (B) Quantitation of the light and dark phase RER from the data shown in (A). (C) Continuous monitoring of locomotor activity (beam breaks). (D) Quantitation of light and dark phase activity from the data shown in (C). Data represents the mean ± s.e.m. of 6 animals per group.

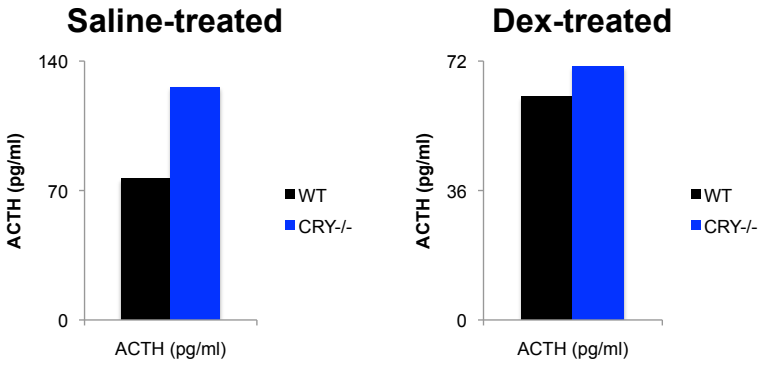

Figure S7: ACTH tends to be increased in Cry-deficient mice. Adrenocorticotropic hormone (ACTH) measured by radioimmunoassay in mouse serum collected from wildtype or CRY-/- animals following eight weeks of treatment with either saline or dexamethasone by i.p. injection every other day.