Supporting Information

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SI Materials and Methods

Sex. An interaction term between maternal cortisol in pregnancy and sex of the child was included in all our statistical models. In these regression models, sex was included as a binary variable (girls = 0, boys = 1). Therefore, the interaction term (i.e., the product of sex and cortisol) becomes zero for girls, so the main effect of cortisol in our model represents the cortisol effect among girls. The interaction effect is then interpreted as the expected change in the response variable (i.e., regional brain volume) corresponding to one unit increase in cortisol measurement among boys in addition to what is expected for girls.

Multiple Imputation. The protocol included serial saliva collection at five time points over gestation, but not all women provided five samples. Complete-case analysis (i.e., removing observations with missing values) reduces the sample size and hence the efficiency of estimates, and can thereby lead to biased estimates. To minimize this issue, we used the multiple-imputation method to generate five completed data sets. For this purpose, all the relevant explanatory variables (i.e., the covariates listed earlier) as well as the response variables (i.e., brain volumes) were included in the model. Note that including the response variables in missing data imputation does not result in a circular analysis, and the exaggeration of the importance of explanatory variables (1) but instead increases the imputation accuracy. Multiple imputation methods draw samples (in this case, five) from the conditional distribution of the target variable (i.e., the variable with missing values) given the observed values of all other relevant variables. The regression analysis was then performed on each completed data set separately, and the final estimates of regression parameters were obtained by averaging over the multiple imputations. The corresponding variance–covariance matrix for the final estimates is adjusted for variability due to imputation. The Hmisc package in R software was used to perform multiple imputations analysis.

Covariates. Because the literature on whether body mass index is associated with cortisol concentrations is inconsistent, with approximately equal numbers of studies suggesting higher body mass index in association with higher cortisol concentrations (e.g., refs. 2 and 3) or lower cortisol concentrations (e.g., refs. 4 and 5), we tested whether, in the present sample, maternal weight at any of the five gestational time points was associated with cortisol concentrations when controlling for maternal height. Because this was not the case (15 wk, $r = -0.20$, $P = 0.26$; 19 wk, $r = -0.12$, $P = 0.36$; 25 wk, $r = 0.01$, $P = 0.93$; 25 wk, $r = 0.01$, $P = 0.93$; 31 wk, $r = -0.06$, $P = 0.64$; 37 wk, $r = -0.07$, $P = 0.62$), maternal weight was not included in the statistical models as a covariate.

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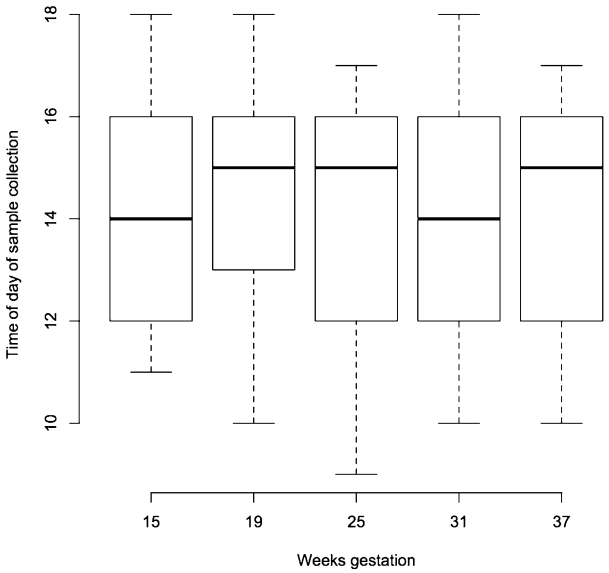

Fig. S1. Box plots show distribution of time of day of sample collection at each gestational visit.