In utero Sources of Skeletal Variation: the Role of Maternal Prenatal Stress

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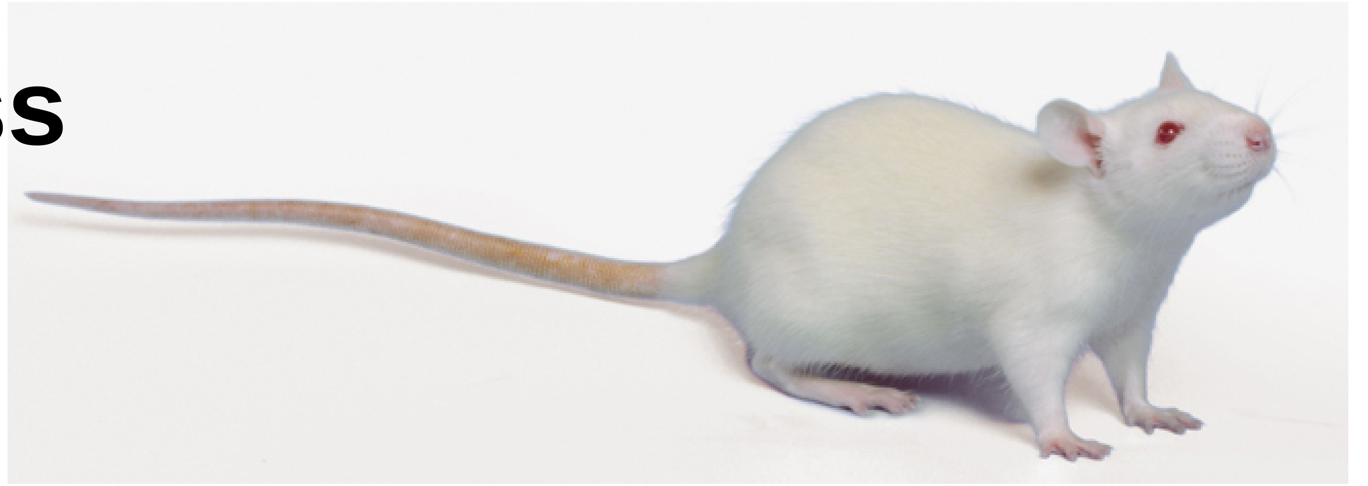


Background

Fossilized bones are the primary evidence we have of our lineage's evolutionary history. As such, the morphological variation of these bones is our main source of information about what they looked like, where and when they lived, and possibly, how selection operated on them. While many biologists focus on genetic underpinnings of skeletal shape variation, it is equally as important to understand the non-genetic factors that influence the shape and histological structure. Non-genetic influences include bone remodeling in response to stresses and strains faced during life. An increasingly recognized non-genetic source of variation is the influence of prenatal environment. Biologists are finding that *in utero* experiences can influence the phenotype long into adulthood. This study tested the hypothesis that maternal stress during pregnancy will have negative effects on offspring's skeletal development.

Materials & Methods

Twenty-eight female and seven male Wistar rats (*Rattus norvegicus*) were purchased and bred to produce 176 offspring. During pregnancy, dams were randomly divided into four groups (n=7, per group) and immobilization stress induced as follows; Group 1:immobilization stress on days 1-7 of pregnancy, Group 2: on days 8-14, Group 3: on days15-21, Group 4: left undisturbed.

STRESS SCHEDULE FOR PREGNANT DAMS					
Stress Period	Gestational Week (GW) 1	GW2	GW3	No Stress (Control)	TOTAL
Total Number of Dams	7	7	7	7	28
SACRIFICE SCHEDULE FOR OFFSPRING					
Total offspring per group	56	56	35	43	224
Number of pups sacrificed at week 4	11	11	7	5	44
Number of pups sacrificed at week 8	11	11	13	8	44
Number of pups sacrificed at week 12	11	11	8	12	44
Number of pups sacrificed at week 16	11	11	8	18	44

Maternal cortisol hormone, food intake, and weight gain were monitored during pregnancy. Pups were raised under normal laboratory conditions and sacrificed at ages: 4, 8, 12, and 16 weeks to determine the effect of prenatal stress. At necropsy, the tibia was removed and processed for histology.

Statistical analysis: Differences among groups were determined by T-test or analysis of variance. Linear regression analysis was performed to establish the relationship between stress in utero and indicators of bone development in offspring. P values ≤ 0.05 were considered significant. All analyses were performed with StataCoRP. 2009. Stata; Release 11. Statistical software. College Station, TX: StataCorp LP.

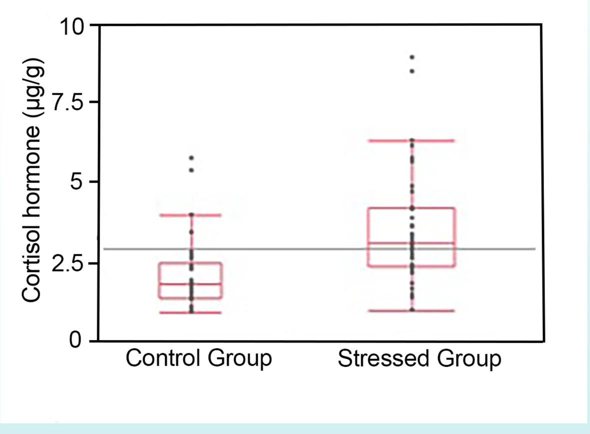
Summary of Research

1) Cortisol hormone levels in controls were lower than those of stressed animals.
2) Stressed dams consumed 12.5% less food per day compared to controls and gained less weight.
3) Offspring stressed during GW3 gained more weight postnatally than did the controls or the other stressed groups
4) GW3 offspring had a higher rate of bone formation.

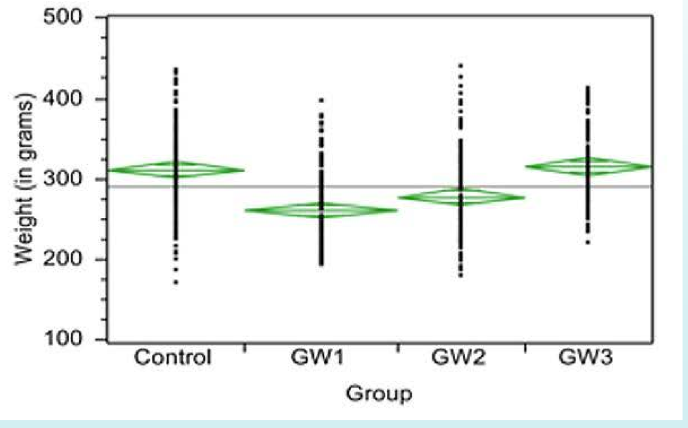
Stress during the last third of pregnancy results in increased cortisol and reduced food intake in mothers, but faster growth and higher weight gain in the offspring.

Results

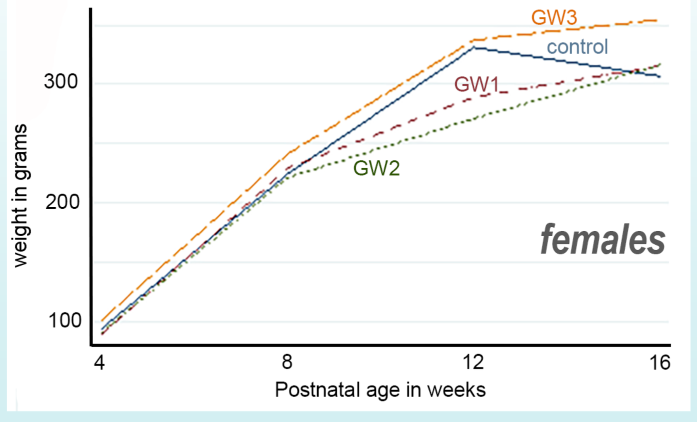
Cortisol hormone levels between Control and Stressed mothers

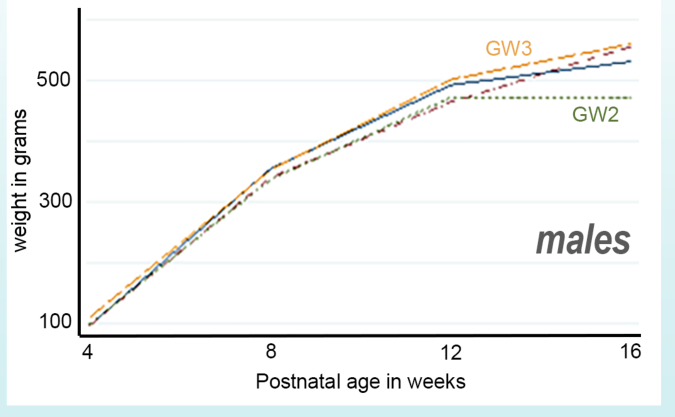


Maternal weight gain during pregnancy

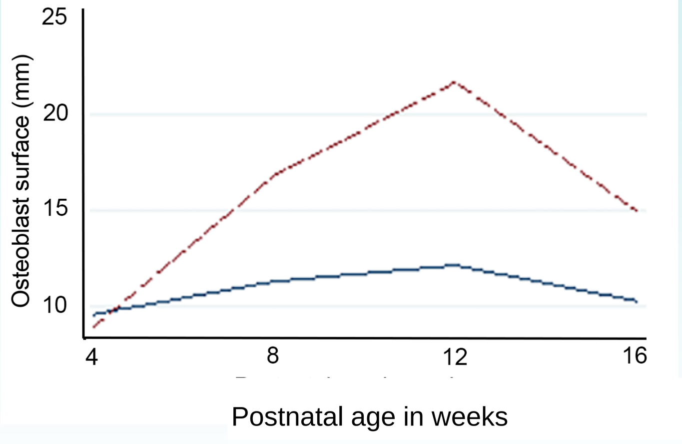


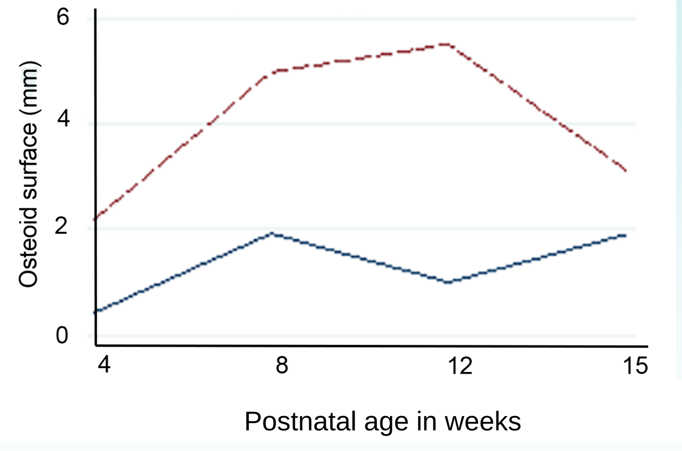
Weight (in grams) of offspring separated by sex

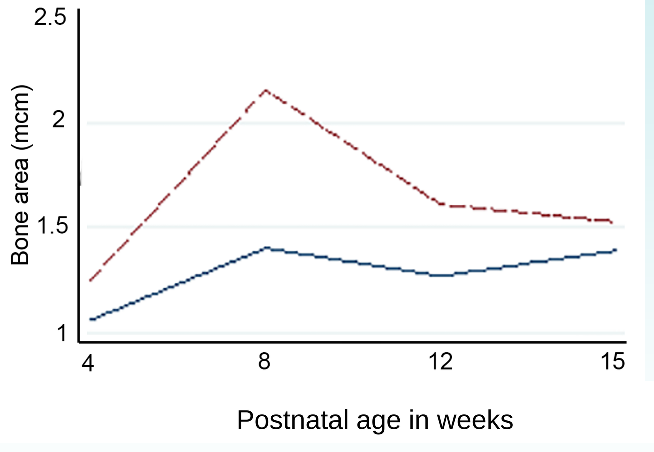


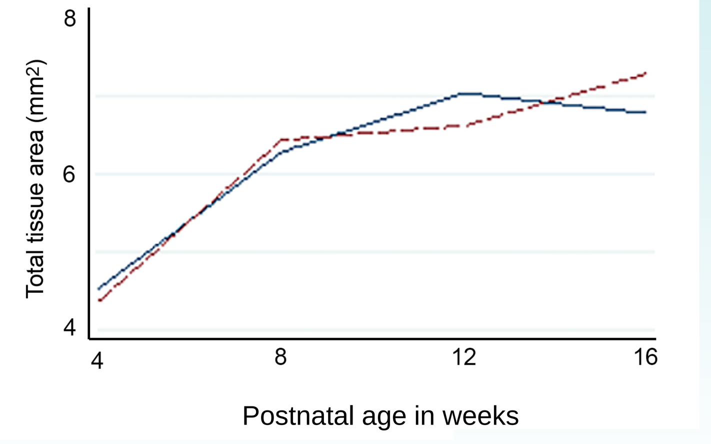


The changes in histomorphometric changes at age 4, 8, 12 and 16 weeks in GW3 offspring (dashed line) and control offspring (solid line).

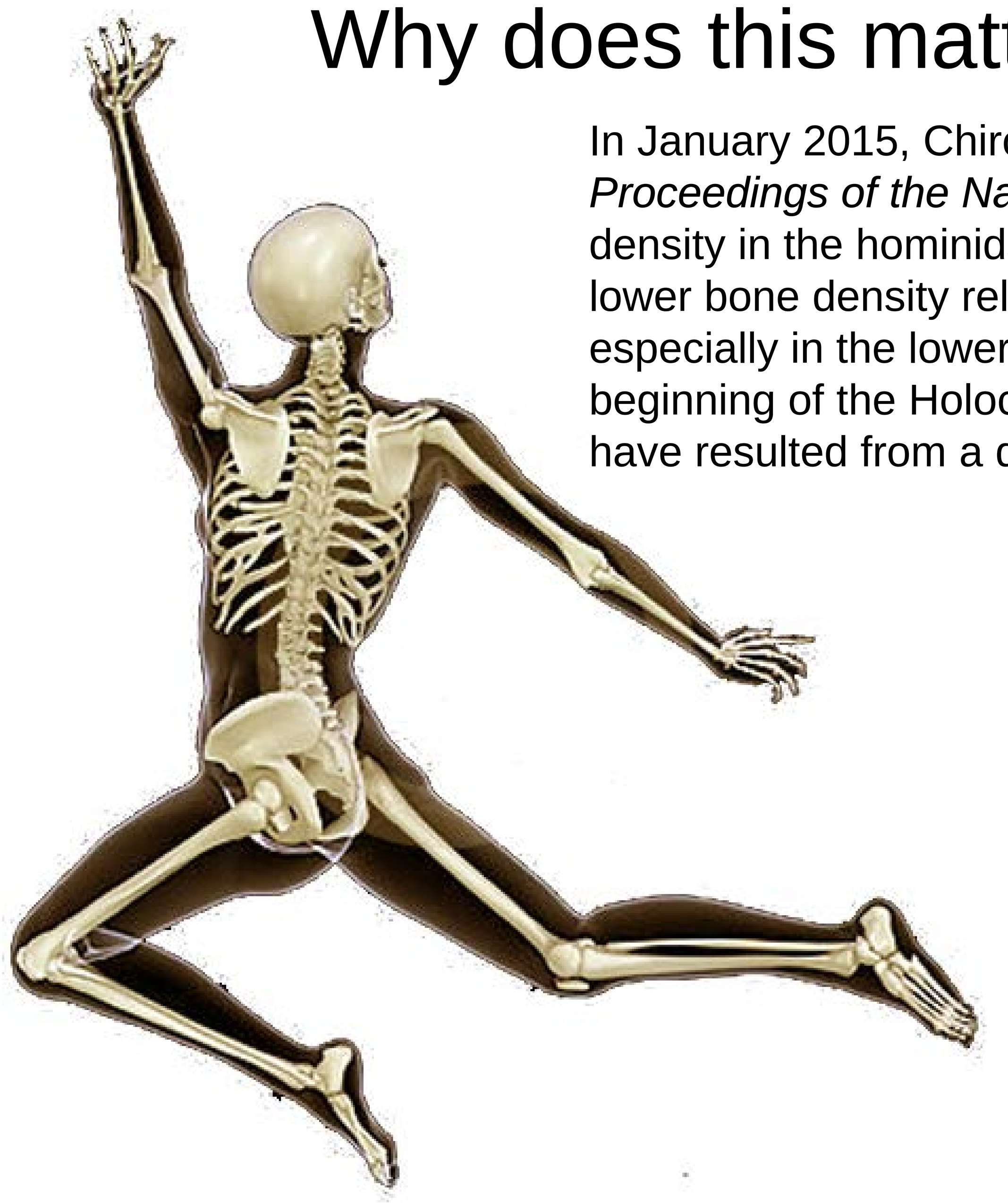








Why does this matter for paleoanthropology?



In January 2015, Chirchir and colleagues[3] published a study in the *Proceedings of the National Academy of Sciences USA* about trabecular bone density in the hominids. They found that modern humans have significantly lower bone density relative to other apes and earlier members of our Family, especially in the lower limbs. This shift in bone density occurred with the beginning of the Holocene. The authors suggest that this phenomenon may have resulted from a decrease in mobility (i.e., shorter distances walked, etc.).

Our results suggest that other factors, such a the dramatic effects that a shift to agriculture brings with it could provide plausible alternative hypotheses. For example, agricultural may reduce maternal stress.

The more we know about the etiology of the observed biology, the better our ability to discern the evolutionary history of our species.

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3. Chirchir et al. (2015). Recent origin of low trabecular bone density in modern humans. PNAS 112(2):366-371.