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**ORIGINAL ARTICLE**

# Newborn insula gray matter volume is prospectively associated with early life adiposity gain

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**BACKGROUND:** The importance of energy homeostasis brain circuitry in the context of obesity is well established, however, the developmental ontogeny of this circuitry in humans is currently unknown. Here, we investigate the prospective association between newborn gray matter (GM) volume in the insula, a key brain region underlying energy homeostasis, and change in percent body fat accrual over the first six months of postnatal life, an outcome that represents among the most reliable infant predictors of childhood obesity risk.

**METHODS:** A total of 52 infants (29 male, 23 female, gestational age at birth = 39(1.5) weeks) were assessed using structural MRI shortly after birth (postnatal age at MRI scan = 25.9(12.2) days), and serial Dual X-Ray Absorptiometry shortly after birth (postnatal age at DXA scan 1 = 24.6(11.4) days) and at six months of age (postnatal age at DXA scan 2 = 26.7(3.3) weeks).

**RESULTS:** Insula GM volume was inversely associated with change in percent body fat from birth to six-months postnatal age and accounted for 19% of its variance ($\beta = -3.6\%$/S.D., $P = 0.001$). This association was driven by the central-posterior portion of the insula, a region of particular importance for gustation and interoception. The direction of this effect is in concordance with observations in adults, and the results remained statistically significant after adjusting for relevant covariates and potential confounding variables.

**CONCLUSIONS:** Altogether, these findings suggest an underlying neural basis of childhood obesity that precedes the influence of the postnatal environment. The identification of plausible brain-related biomarkers of childhood obesity risk that predate the influence of the postnatal obesogenic environment may contribute to an improved understanding of propensity for obesity, early identification of at-risk individuals, and intervention targets for primary prevention.

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## INTRODUCTION

Obesity currently represents among the most urgent national and global health challenges.[1] Childhood obesity is a particularly grave concern because obese children are substantially more likely to be obese as adults,[2] and to develop obesity-related diseases at earlier ages[3,4] and of greater severity.[1,5–7] Moreover, once established, obesity is extremely difficult to reverse,[8] underscoring the critical importance of primary prevention.[9]

Obesity is a complex, multi-factorial phenotype.[10] In this context, the importance of energy homeostasis (balance) systems, and the integrity of brain circuits that regulate energy homeostasis, is well established.[11] It is, however, unclear whether the observed differences in brain regions and circuitry in obese relative to normal weight individuals are a cause, consequence, or both, of the obese state. Moreover, relatively little is known about the developmental ontogeny of these brain regions and circuitry, particularly during the period of intrauterine development (when the postnatal obesogenic environment could not yet have affected this circuitry), and its prospective role in shaping propensity for childhood obesity.

The present study was conducted to determine the prospective association of intra-individual variation in newborn human brain anatomy (characterized by MR-based measures of structural brain phenotypes) with childhood obesity risk (characterized by measures of fat accretion in early infancy). Here, we focused on the insula, a paralimbic brain structure, as the primary predictor of interest for two reasons. First, among the various brain regions and circuitry implicated in energy intake, the insula represents a key component. It plays an essential, obligatory role in somatosensation, interoception, gustation, olfaction, reward/addiction and emotion.[12] The insula receives afferent signaling via taste receptors in the tongue via the rostral division of the nucleus of the solitary tract (NTS) and the ventroposterior medial nucleus of the thalamus (VPMpc) and is believed to integrate multisensory input.[13,14] It also has direct and dense reciprocal structural connections to the amygdala,[15] a brain structure responding to taste and textural coding,[16] and is thereby believed to mediate the processing of taste and reward.[17,18] Second, the insula is a relatively large structure with appreciable gray matter (GM); therefore, its size can be reliably characterized in the newborn brain. We focused on fat mass accretion in early postnatal life as the primary outcome of interest because several studies have found that growth velocity, and particularly the increase in fat mass during early infancy, is among the most reliable and valid predictors of subsequent childhood obesity risk.[19–22]

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We hypothesized that newborn insula GM volume is prospectively and negatively associated with fat mass accretion in early postnatal life. Furthermore, we hypothesized a region-specific effect of the central-posterior portion of the insula because previous studies have implicated this specific region in gustation and interoception.[23] We conducted a prospective, longitudinal study in a population-based cohort of mother-infant dyads from gestation through birth till six-months postnatal age. We conducted MR brain imaging in newborns shortly after birth to quantify insula GM volume, and whole body Dual-energy X-ray Absorptiometry (DXA) imaging shortly after birth and six-months postnatal age to quantify body composition and change in percent fat mass over the early postnatal period of life. We examined the prospective association of global as well as regional newborn insula GM volume with subsequent change in body fat percentage between birth and six-months age. Our analysis accounted for the effects of key covariates including gestational age at birth, postnatal age at assessment(s), and additional potential covariates and confounding factors including maternal pre-pregnancy body mass index (ppBMI), maternal insulin sensitivity during gestation, infant birth weight percentile, infant sex, and postnatal feeding practices.

## MATERIALS AND METHODS

### Study population

The infants evaluated in this study were born to mothers with healthy pregnancies (no major obstetric or birth complications). DXA scans were completed in 119 and 91 individuals at the newborn and six-month time point visits, respectively, with serial DXA imaging in 81. Of these 81 individuals, 54 neonates completed an MRI scan. Two individuals were removed during a blinded quality control process due to excessive movement artifact resulting in insufficient quality for tissue segmentation. Thus, the final sample size was 52 infants (29 male, 23 female). Mean gestational age at birth was 39.5(1.4) (S.D.) weeks. Neonatal T1- and T2-weighted MR brain imaging was obtained at 25.9(12.2) days after birth. Serial DXA assessments were obtained shortly after birth (24.6(11.4) days) and at six-months age (26.7(3.3) weeks). The 52 neonates included in this analysis had no significant differences in any of the key socio-demographic variables from those who were not included due to partial data collection. The demographics of the study population are provided in Table 1. The Institutional Review Board of the University of California, Irvine, approved all study procedures, and all parents provided written, informed consent.

**Table 1.** Maternal and Infant Characteristics

| | |
|---|---|
| *Maternal Characteristics* (N = *52*) | |
| Race/Ethnicity | |
| Hispanic White *N* (%) | 20 (39%) |
| Hispanic of Other Race *N* (%) | 21 (40%) |
| Non-Hispanic White *N* (%) | 4 (8%) |
| Non-Hispanic of Other Race *N* (%) | 7 (13%) |
| Pre-pregnancy BMI | |
| Mean (SD) | 28.6 (7.0) |
| *N* (Normal/Overweight/Obese) | 18/18/16 |
| *Infant Characteristics* | |
| Sex | |
| Male, *N* (%) | 29 (56%) |
| Gestational Age at Birth (weeks) | |
| Mean (SD) | 39 (1.5) |
| *N* < 37 weeks | 4 |
| Birth weight (kg) | |
| Mean (SD) | 3.35 (0.58) |
| *N* < 2.5kg | 3 |
| Feeding Status | |
| Breastfed *N* (%) | 19 (36%) |
| Mixed *N* (%) | 14 (28%) |
| Formula Fed *N* (%) | 19 (36%) |

### Dual X-Ray absorptiometry (DXA) assessments

A whole body DXA scan was obtained using a Hologic Discovery Scanner (A, QDR 4500 series, Hologic Inc., Bedford, MA, USA) in pediatric scan mode. Calibration using Hologic's anthropomorphic Spine QC Phantom was performed before each scan. During the scan, sleeping infants lay supine while swaddled in a light cotton blanket wearing only a disposable diaper. If the baby moved during the scan, a single repeat was performed once the baby had been pacified. Global body fat percentage (BF%) was defined as 100 times the ratio of global fat mass to the sum of global fat mass and global fat-free mass. BF% was residualized for gestational age at birth and postnatal age at scan. The longitudinal change (ΔBF%) was then defined as the change between newborn and six-month time points.

### MRI assessments

MRI scans were acquired during natural sleep using a 12-channel head receive coil. High-resolution anatomical scans including T1-weighted (MPRAGE, TR/TE/TI = 2400/3.16/1200 ms, Flip Angle = 8 degrees, Matrix = 256 × 256 × 160, Resolution = 1 × 1 × 1 mm, 6 m18 s) and T2-weighted (TSE, TR/TE = 3200/255 ms, Matrix = 256 × 256 × 160, Resolution = 1 × 1 × 1 mm, 4 m18 s) images were acquired. After feeding and soothing to the point of sleep, neonates were placed in a CIVCO beaded pillow (www.civco.com). The pillow covered the neonates' body and head, became rigid under vacuum, and provided a comforting swaddle, motion prevention and hearing protection when used in conjunction with standard foam earplugs. A pediatric specialist observed the neonates throughout the duration of scans, monitoring for heart rate and oxygen saturation.

### MRI volumetric analysis

Brain tissue was classified as either GM, white matter (WM), or cerebrospinal fluid using an automatic, atlas-moderated expectation maximization segmentation tool.[24] Intracranial volume (ICV) was defined as the sum of all three tissue-classes. Cortical parcellation was used to define the insula using non-linear warping of a parcellation atlas template. Insula GM volume was defined as the intersection between the GM mask and the insula region of interest (Figure 1). ICV was age-corrected in identical fashion to BF%. In separate analyses (see Results section), insula GM volume was either only corrected for gestational age and scan age independent of ICV, or additionally corrected for ICV. A simple geometric parcellation scheme was implemented by subdividing the insula along its long axis into three parcels of equal width.

### Association testing

All statistical models tested without adjusting for potentially confounding variables used Pearson correlation analyses with gestational/postnatal age-corrected ΔBF% as the outcome, and either gestational and postnatal age- corrected insula GM volume or gestational/postnatal age *and* ICV-corrected insula GM volume as the predictor. The influence of potential confounding variables was tested using multiple linear regression analysis,

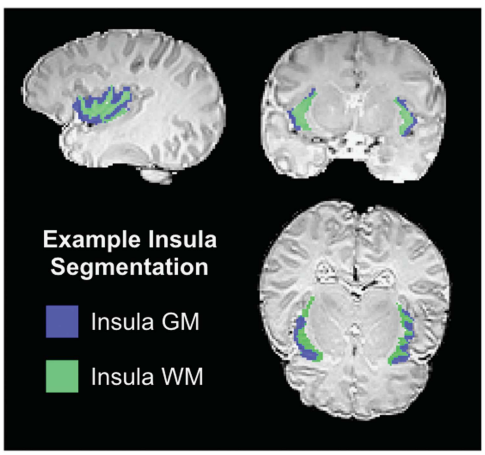


**Figure 1.** Example Insula Segmentation. Typical gray and white matter segmentations in the insula are overlaid on a newborn T2-weighted image.

with ΔBF% as the outcome and GM volume and the confounding factors (see below) as predictors.

### Receiver operating characteristic

As an indication of clinical value, Receiver operating characteristic (ROC) analysis was conducted using a threshold of 75th percentile ΔBF% as proxy categorization of participants as either at-risk or not at-risk of future obesity. The summary statistic area-under-the-curve (AUC) is reported for the ROC. ROC analysis provides a quantitative measure of the utility of bi-lateral GM insula volume as an obesity-risk screening tool by specifying a false positive rate for a given true positive rate of categorical risk identification.

### Confound variable definition

Our *a priori* selection of the covariates was based on theoretical considerations and findings from the available literature of possible associations with either only the outcome of interest (to improve model precision) or with both the outcome and primary predictor of interest (to address potential confounding). Because maternal conditions prior to or during pregnancy may concurrently influence fetal brain development and infant fat gain, and because postnatal factors may additionally influence infant fat gain, the following variables were used as potential covariates/confounds: maternal pre-pregnancy BMI, maternal insulin resistance index (HOMA-IR averaged over three trimesters), newborn birth weight percentile (birth weight corrected for gestational age at birth), infant sex (separate models for main effects and interaction), and infant feeding practices (exclusive breastfeeding, exclusive formula feeding or mixed practices). Maternal pre-pregnancy BMI was based on maternal self-report and verified using height and weight measurements taken at the first pregnancy visit (self-reported pregnancy weight was highly correlated with measured weight on the first visit ($r = 0.97$, $P < 0.001$)). Maternal insulin and glucose were measured throughout pregnancy (first, second and third trimester) using standard enzyme-linked immunosorbent assay protocols. HOMA-IR was calculated based on these measures, and the average across pregnancy was used as an indicator of maternal insulin sensitivity. Birth weight was abstracted from the medical record, and gestational age was determined by best obstetric estimate with a combination of last menstrual period and early uterine size and was confirmed by obstetric ultrasonographic biometry before 15 weeks using standard clinical criteria.[25] Infant sex was abstracted from the medical record. Infant feeding practices were assessed *via* monthly maternal interviews. A composite measure of feeding practice categorized offspring with greater than 75% of the first six months of life spent exclusively breastfeeding as breastfed, $<25\%$ of the first six months of life spent exclusively breastfeeding as formula-fed and intermediate values as mixed feeding practice.

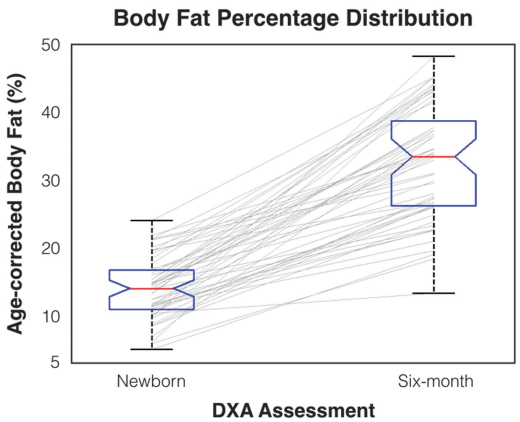


**Figure 2.** Inter-individual Variability in Body Fat Percentage Change in Early Life. Box and whisker plots depict body fat percentage distributions in newborns and six-month old infants. Red lines, whiskers, boxes, and notches are medians, extreme values, quartiles (25/75) and confidence intervals (95%) respectively. The gray lines are individual trajectories of body fat percentage change, our main outcome variable.

## RESULTS

### Early life adiposity change

Newborn and six-months DXA BF% values were, on average, 13.4 (6.0)% and 32.1(8.4)%, respectively (Figure 2). The main outcome variable, ΔBF%, was + 18.4(8.3)% (that is, an average increase of 0.80%/week).

### Global and regional brain measures

Mean ICV and whole brain GM values were 479(56) $cm^3$ and 258(31) $cm^3$, respectively. ICV and whole brain GM were significantly associated with gestational age at birth ($p_{ICV} < 10^{-3}$, $\hat{\beta}_{ICV} = 0.58$; $p_{GM} < 10^{-5}$, $\hat{\beta}_{GM} = 0.69$) and postnatal age at scan ($p_{ICV} < 10^{-3}$, $\hat{\beta}_{ICV} = 0.49$; $p_{GM} < 10^{-5}$, $\hat{\beta}_{GM} = 0.56$). Left and right insula GM volumes were on average 1378(296) $mm^3$ and 1320 (292) $mm^3$, respectively. As anticipated, left and right insula GM volumes were correlated ($R^2 = 0.44$, $P < 10^{-5}$). Insula GM volumes were associated with ICV ($\eta^2_{Left} = 31\%$, $p_{Left} < 10^{-5}$; $\eta^2_{Right} = 28\%$, $p_{Right} < 10^{-5}$) but were not associated with gestational age at birth ($p_{Left} > 0.1$; $p_{Right} > 0.1$) or postnatal age at scan ($p_{Left} > 0.1$; $p_{Right} > 0.1$).

### Associations between newborn insula gray matter volume and early life fat gain

Left/right averaged insula volume was negatively associated with ΔBF% after correcting for gestational and postnatal age at scan ($\beta_{insula} = -3.6\%$/S.D., $R^2_{insula} = 18.6\%$, $p_{insula} = 0.001$; Figure 3). The left and right insula were roughly equal contributors to this association ($\beta_{Left} = -3.4\%$/S.D., $R^2_{Left} = 16.7\%$, $p_{Left} = 0.002$; $\beta_{Right} = -3.1\%$/S.D., $R^2_{Right} = 14.4\%$, $p_{Right} = 0.005$). Because there was a tendency for a negative association between ICV and ΔBF% ($\beta_{ICV} = -2.0\%$/S.D., $R^2_{ICV} = 5.7\%$, $p_{ICV} = 0.086$), the analyses were repeated including ICV as a covariate. Although the magnitude of the effect was attenuated, the effect of average insula volume remained significant upon controlling for ICV ($\beta_{insula,ICV} = -2.4\%$/S.D., $R^2_{insula,ICV} = 8.2\%$, $p_{insula,ICV} = 0.038$). ROC analysis was performed to assess the true positive rate as a function of the false positive rate of classifying an obesity risk proxy measure ($>75$th percentile ΔBF%) using bilateral GM insula volume. The results indicate significant stratification of high − ($>75$th percentile ΔBF%) and moderate to low-percentile ($<75$th percentile ΔBF%) participants by insula GM volume (AUC = 0.74 (95% CI: 0.57 − 0.91); Figure 4).

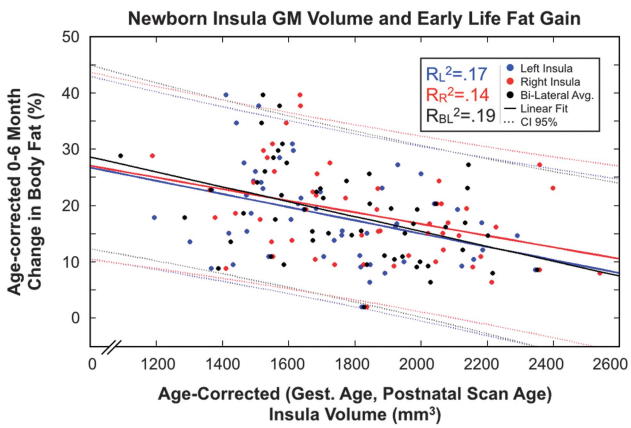


**Figure 3.** Insula Gray Matter Volume Predicts Adiposity Change In The First Six Months Of Life. Bilateral insula volume at birth is negatively associated with change in body fat percentage.

### Insula region-specific associations with infant fat gain

Regional associations between GM volume and ΔBF% (Figure 5) revealed a central-posterior pattern of significance. These findings are spatially consistent with cortical representations of gustation and interoception as defined by electrocortical stimulation,[26] magnetoencephalography[27] and functional MRI in humans.[23,28] The effect of the posterior section of the trisected parcellation was the most significant one in both left and right insula ($\beta_{Left,posterior} = -3.5\%$/S.D., $p_{Left,posterior} = 0.005$; $\beta_{Right,posterior} = -3.3\%$/S.D., $p_{Right,posterior} = 0.009$; Figure 5).

### Covariates

Bilateral insula GM volume remained significantly associated with ΔBF% ($P = 0.010$) when including the above *a priori*-specified covariates (maternal pre-pregnancy BMI, maternal insulin sensitivity during pregnancy, newborn birth weight percentile, infant sex, and infant feeding practices). In the full model, the effect size of insula GM volume predicting infant ΔBF% was larger than in the reduced model ($\beta_{insula,reduced\ model} = -3.6\%$/S.D., $\beta_{insula,full\ model} = -4.6\%$/S.D.). None of the covariates tested were significantly associated with ΔBF% in the full model. In stratified analyses, the main association between average insula volume and infant adiposity gain appeared weaker in boys ($\beta_{GM,Volume\text{-}\Delta BF\%} = 3.2\%$/S.D., $p_{GM,Volume\text{-}\Delta BF\%} = 0.045$) than in girls ($\beta_{GM,Volume\text{-}\Delta BF\%} = 4.1\%$/S.D., $p_{GM,Volume\text{-}\Delta BF\%} = 0.008$), there was no main effect of sex or interaction effect of sex and insula volume in a full model ($P > 0.1$).

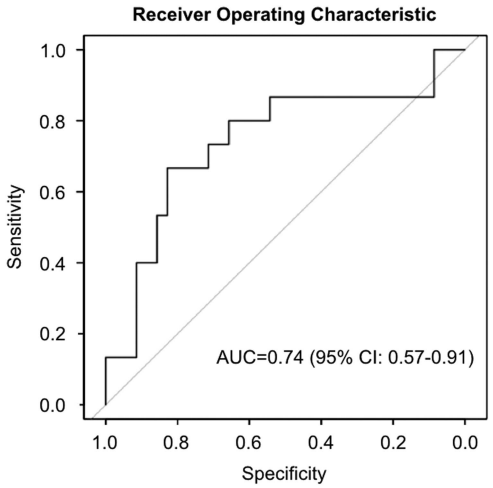


**Figure 4.** Receiver Operating Characteristic Curve. The sensitivity and specificity of correct classification (that is, plot of the true positive rate against the false positive rate) of an obesity risk proxy (>75th percentile ΔBF%) using newborn bi-lateral average insula GM volume. An AUC of 0.74 reflects the utility of insula GM volume to discriminate at-risk individuals (>75th percentile ΔBF%).

## DISCUSSION

This work represents, to the best of our knowledge, the first report prospectively relating newborn brain structure with infant fat gain over the first six months of postnatal life, a key risk factor for childhood obesity. Using a hypothesis-based and focused approach, we demonstrate a negative association between newborn insula GM volume and subsequent infant fat mass accretion. The direction of this effect is consistent with previous findings relating insula GM volume and obesity in adults, and spatially consistent with region-specificity of the insular cortex involved in gustation and interoception. Furthermore, the observed effect remained significant after controlling for key covariates. Thus, this finding, which supports our hypothesis that a smaller insula at birth is associated with greater fat gain in early life, has promising potential as a novel biomarker of childhood obesity risk.

The insula is implicated in an array of complex human processes driving feeding behavior, including somatosensation, interoception, gustation, olfaction, reward/addiction and emotion.[12] The region-specific associations identified here suggest that the central-posterior region of the insula—the region where gustatory and interoception functionality overlap[23]—is the primary driver of the observed effect. In this context, it then suggests that reduced GM volume in regions functionally responsible for sensing fullness and qualities of food intake (for example, nutritional content and viscosity) may be reflective of an attenuated ability to integrate internal (for example, stomach extension) and environmental (for example, sensing fat content) cues necessary for the normal cessation of eating, resulting in abnormal regulation of intake and homeostasis in infancy. Furthermore, one recent study examining neural responses to visual food cues demonstrated a negative association between fMRI activation in the central-posterior insula

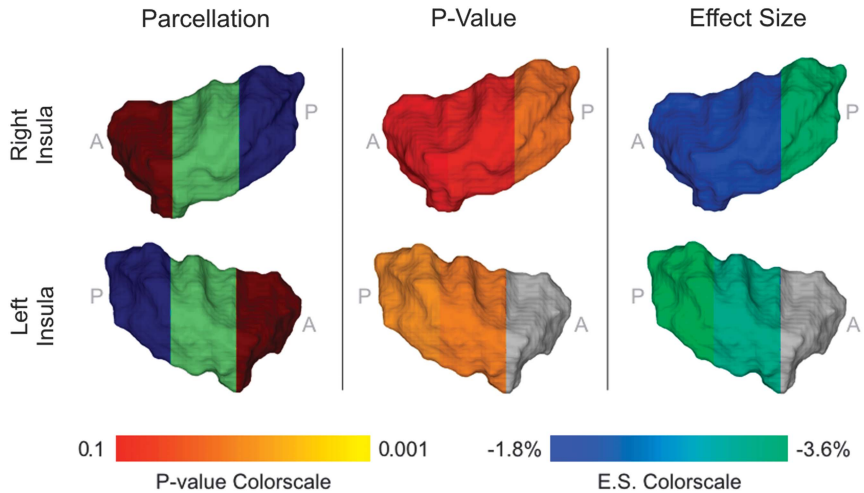


**Figure 5.** Sub-parcellation of the Insula Suggests a Central-Posterior Pattern of Association between Insula GM Volume and Infant Adiposity. The insula was geometrically parcellated using a three-region scheme. Central-posterior insula GM volume showed the strongest prediction of early life fat gain. The effect size (E.S.) illustrated here is estimated slope from the General Linear Model, ΔBF% per standard deviation in volume. Left-most row: Dark Blue = Posterior, Green = Central, Dark Red = Anterior.

and peripheral glucose levels,[29] suggesting a mechanism whereby high levels of immediately available energy decrease insula-dependence when processing food cues. In this scenario, reduced GM insula volume may limit the capacity to appropriately respond to such a signaling mechanism.

Perhaps the most salient point of this study is that insula GM volume was measured in early infancy, a time point when postnatal conditions likely have exerted only minimal influence relative to those in later infancy and childhood. This, then, raises the question of the origin(s) of its inter-individual variation at birth. Indeed, a number of prenatal conditions, including variation in maternal-placental-fetal endocrine, immune/inflammatory, metabolic, and lipid biology, have been associated with offspring obesity.[30] These same prenatal environmental conditions have the potential to 'program' the developing fetal brain,[31] which may represent one of the pathways mediating the link between the *in utero* environment and adiposity (obesity) risk. Thus, it is plausible that these environmental conditions, independently or in interaction with genotypic variation, produce a phenotype (integrity of insula circuitry) that is associated with an altered propensity for obesity.

In the broader context of obesity, the role of the insula in modulating complex feeding behaviors is well established.[11,14,32,33] This is evidenced by differential insula activation in obese relative to non-obese individuals when presented with a gastric distension,[34] food-related visual[35] and gustatory stimuli.[36] Functional activation of the insula has also been associated with ghrelin administration,[37] stomach extension,[38,39] and leptin replacement.[40] Structurally, obesity-related reductions in insula GM volume have been reported in multiple, large population-based studies.[41–44] More nuanced structural obesity-insula findings include negative associations between frontal operculum GM volume and blood leptin concentration,[45] positive associations between insula GM volume and aerobic capacity,[46] and reduced brain metabolism coupled with insula GM atrophy in obese individuals.[47] Finally, reduced insula GM in individuals identified as obese-prone relative to those categorized as obese-resistant supports the notion that reduced insula volume is itself a risk factor for future weight gain.[48]

Strengths of the study include the prospective, longitudinal research design, MRI-based measures of brain anatomy shortly after birth, and direct ascertainment of infant adiposity by serial DXA-based measures of percent body fat. As mentioned earlier, there is considerable evidence suggesting child size and growth velocity (particularly during the period of early infancy) represent among the most reliable, valid, and strongest predictors of childhood obesity risk. Increased size or rapid weight or fat gain during this phase is associated with increased infant cardiovascular risk factors,[49] increased childhood (and adult) obesity risk[19–22,50–52] and related outcomes including type 1 diabetes,[53] metabolic disorders,[54] hypertension,[55] and asthma[56] in later life. A recent study reported that change in infant fat mass during early infancy was a much stronger predictor of childhood obesity risk than weight gain alone.[19] The rationale for choosing body fat percentage, as opposed to absolute fat mass, derives from the fact that percentage, as a relative measure, is less dependent on total weight gain and growth. These findings support the critical importance of longitudinal assessments of body composition particularly during the period of early infancy and the validity of their use as outcomes in the context of the current work.

Limitations of this study include the lack of assessment of physical activity as a potential postnatal covariate, limited spatial resolution due to the constraints of imaging newborn participants and the interpretability of using early life fat gain as an indicator for future obesity risk. Early life physical activity levels (PAL) appear to be only weakly associated with adiposity at six months of age.[57] Furthermore, because the insula is thought to impact processing of feeding cues as opposed to energy expenditure, it is expected that inclusion of PAL as a confounding variable would only remove variance in ΔBF% unrelated to insula volume. Lastly, due to effectively low resolution (the newborn brain is roughly one-third the size of an adult brain) and low tissue contrast in the newborn brain, the estimates of the insula used here were limited to lobar volume and a relatively simple parcellation scheme that is not wholly reflective of fine grained insula neuroanatomy.[58] We also note that while the outcome variable used in the current study (change in infant body fat percentage in early postnatal life) represents among the most reliable and valid indicators *during this time period* of childhood obesity risk, its long-term effects on adult obesity risk likely are conditioned upon subsequent factors during childhood and beyond. Thus, future longer-term follow up studies are warranted in this regard.

An important expansion of this work beyond longer-term characterization of adiposity trajectories would include assessment of the determinants of newborn insula GM volume, including offspring genetic risk for obesity, and prenatal environmental conditions such as variation in maternal-placental-fetal endocrine, immune/inflammatory, metabolic and lipid biology. The placenta, as the interface between the fetal and maternal compartments, also represents an important target for future studies examining the influence of the intrauterine environment on brain and body composition developmental trajectories. Furthermore, future studies could benefit from imaging the newborn insula at a higher resolution[59] (allowing for reliable estimates of cortical thickness), considering appetitive characteristics as a mediator of the observed insula-fat gain association, and expanding brain predictors of adiposity change to functional and structural connectivity measures.

In conclusion, the current study demonstrates that reduced insula GM volume is *prospectively* associated with increased change in body fat percentage in the first six months of life. The magnitude of this effect is moderate to large[60] and relatively larger than that reported for physical activity[57] and genetic-adult obesity associations (using a combination of single nucleotide polymorphisms).[61] While future work is needed to establish the persistence of excess fat in individuals born with smaller insula GM volume, this work may have identified a novel biomarker that reflects a neurobehavioral foundation for future adiposity gain and obesity risk.

## CONFLICT OF INTEREST

The authors declare no conflict of interest.

## ACKNOWLEDGEMENTS

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