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Analysis of Inflammatory Markers in Response to Induction of Reprometabolic Syndrome by a Eucaloric High Fat Diet in Normal Weight Women

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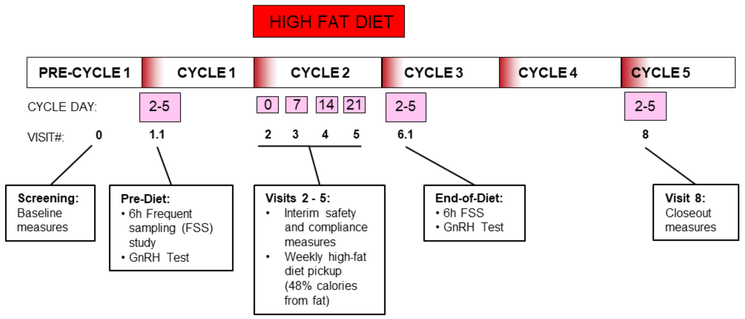
INTRODUCTION

Obesity in women is associated with a spectrum of metabolic and reproductive endocrine disorders including decreased fertility, adverse pregnancy outcomes, and relative hypogonadotropic hypogonadism, which we term Reprometabolic Syndrome.[1] We have shown that the decrease in LH and FSH levels and impaired response to GnRH observed in women with obesity can be recapitulated, in normal weight women, by administration of a one-month eucaloric high fat diet (HFD).[2]

To identify potential mediators of Reprometabolic Syndrome, since obesity is characterized by chronic inflammation, we examined the impact of the HFD on serum levels of a comprehensive panel of inflammatory markers and adipocytokines.[3]

STUDY DESIGN

One Menstrual Cycle Exposure to High Fat, Eucaloric Diet



18 healthy, eumenorrheic, normal weight (BMI 18-24.9 kg/m2) were exposed to a one menstrual cycle eucaloric, prescribed high fat diet (48% calories from fat) and underwent a 6-hour frequent (q10min) blood sampling study in the early follicular phase (days 2-5), before and after consumption of the diet.

METHODS

- Serum samples (60 - 180 min) were pooled and analyzed for a panel of inflammation markers (Table 2) using a multiplex immunoassay (Mesoscale Discovery, Rockville, MD) in the University of Colorado Human Immune Monitoring Shared Resource.
- C-reactive protein (CRP) was measured by University of Colorado Clinical Translational Core Laboratory.
- Differences between pre-diet and end of HFD were analyzed by paired t-test.

DEMOGRAPHICS

Characteristic	Mean (SD)
Age (y)	29.7 (5.9)
BMI (kg/m²)	
Pre-diet	21.6 (2)
End-of-diet	21.4 (1.8)
Height (cm)	166.7 (9.1)
Cycle length (d)	28.3 (2.2)
TSH (uU/ml)	1.84 (1.1)
HbA1c (%)	5.05 (0.2)
PRL (ng/ml)	12.6 (6.4)

TABLE 1: Participant characteristics. Data shown as mean (SD)

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RESULTS

Figure 1: Cytokine and Chemokine Levels Pre- and End-of- High-Fat Diet

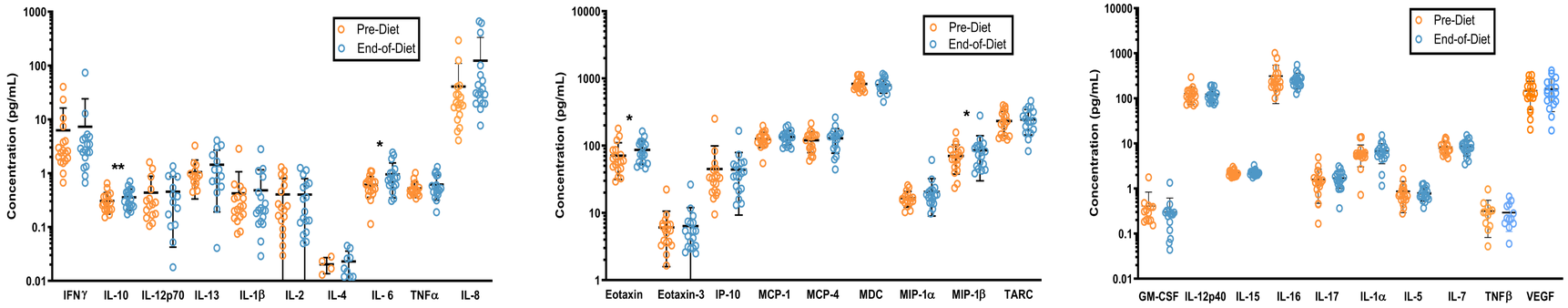

Figure 1: Pooled serum samples from the frequent blood sampling study from 18 normal weight women were analyzed for the indicated meditators of inflammation as described in methods. Horizontal line (—) denotes the mean and error bars are standard deviation. ** p = 0.04 and * p = 0.07.

TABLE 2: Changes in cytokines, interleukins, and chemokines in Normal Weight Women in response to the HFD

Serum Analyte	N	Mean Change	Confidence Interval	p value
Anti-Inflammatory				
IL-10	**18**	**0.05 pg/mL**	**(0, 0.1)**	**0.04**
IL-4	4	0.01 pg/mL	(-0.01, 0.02)	0.31
IL-13	12	0.49 pg/mL	(-0.3, 1.27)	0.2
Pro-Inflammatory				
Eotaxin	**18**	**15.4 pg/mL**	**(-1.13, 31.92)**	**0.07**
Eotaxin-3	18	0.29 pg/mL	(-0.95, 1.54)	0.63
IFN-γ	18	1.04 pg/mL	(-8.34, 10.42)	0.82
IL-12p70	14	-0.01 pg/mL	(-0.19, 0.18)	0.94
IL-1β	18	0.07 pg/mL	(-0.1, 0.25)	0.39
IL-2	16	0 pg/mL	(-0.09, 0.09)	0.98
IL-6	**18**	**0.34 pg/mL**	**(-0.03, 0.72)**	**0.07**
IL-8	18	82.51 pg/mL	(-33.16, 198.17)	0.15
IL-8 (HA)	11	140.71 pg/mL	(-73.53, 354.95)	0.17
IP-10	18	-0.8 pg/mL	(-30.79, 29.19)	0.96
MCP-1	18	8.89 pg/mL	(-6.99, 24.76)	0.25
MCP-4	18	8.41 pg/mL	(-3.46, 20.27)	0.15
MDC	18	-34.72 pg/mL	(-98.08, 28.64)	0.26
MIP-1α	16	4.37 pg/mL	(-2.42, 11.16)	0.19
MIP-1β	**18**	**14.78 pg/mL**	**(-1.39, 30.95)**	**0.07**
TARC	18	12.52 pg/mL	(-9.93, 34.96)	0.26
TNF-α	18	0.1 pg/mL	(-0.05, 0.24)	0.17
GM-CSF	12	-0.08 pg/mL	(-0.19, 0.02)	0.1
IL-12/23p40	18	-5.98 pg/mL	(-21.93, 9.97)	0.44
IL-15	18	0.06 pg/mL	(-0.12, 0.23)	0.51
IL-16	18	-50.6 pg/mL	(-154.71, 53.51)	0.32
IL-17A	18	0.12 pg/mL	(-0.35, 0.59)	0.6
IL-1α	18	0.59 pg/mL	(-0.2, 1.39)	0.13
IL-5	18	-0.08 pg/mL	(-0.3, 0.14)	0.44
IL-7	18	0.34 pg/mL	(-0.68, 1.36)	0.49
CRP	18	-0.08 mg/L	(-0.24, -0.67)	0.872
TNF-β	9	-0.04 pg/mL	(-0.28, 0.2)	0.71
VEGF	18	11.41 pg/mL	(-12.57, 35.39)	0.33

SUMMARY & CONCLUSIONS

- *Induction of Reprometabolic Syndrome in normal weight women was associated with a significant elevation in the anti-inflammatory cytokine IL-10, which may represent a counterregulatory response to the High Fat Diet.*
- *Eotaxin, IL-6 and MIP-1β exhibited similar increases in response to the HFD, which approached statistical significance (p=0.07). These cytokines have been linked to obesity, impaired gonadotropin signaling, and infertility.*
- *Macrophage Inflammatory Protein MIP-1β (CCL4) was also elevated in our prior acute studies of induced hyperinsulinemia and hyperlipidemia in normal weight women and may represent a mediator of the reported effects on the HPO axis.[3]*
- *These findings suggest that the chronic increase in multiple inflammatory markers, characteristic of obesity, is not a primary mediator of the relative hypogonadotropic hypogonadism of Reprometabolic Syndrome.*
- *The impact of circulating lipids, dietary factors and select cytokines on the hypothalamic-pituitary-gonadal axis in obesity merits further investigation.*

REFERENCES

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