IJCIM

International Journal of Complementary and Internal Medicine

RESEARCH ARTICLE

Effect of a High Fructose Diet on Brown Adipose Tissue Development in Adult Obese NIDDM Rats

Orien L Tulp

College of Medicine and Graduate Studies, University of Science Arts and Technology, Montserrat

Corresponding Author: Orien L Tulp. College of Medicine and Graduate Studies, University of Science Arts and Technology, Montserrat, British West Indies. MSR1110. E-mail: o.tulp@usat.edu

Received: September 13, 2023 **Published:** September 29, 2023

Citation: Orien LT. Effect of a High Fructose Diet on Brown Adipose Tissue Development in Adult Obese NIDDM Rats. Int J Complement Intern Med. 2023;5(1):224–235. DOI: 10.58349/IJCIM.1.5.2023.00136

Abstract

In a recent article, the merits of brown adipose tissue as an efficient potential energy buffer that when deficient, may be a contributor to excess fat accretion and in the development of obesity. As such, it offers to be a potential target for therapies that could be developed in its treatment. To determine the effects of dietary fructose consumption on development and cellularity of brown adipose tissue in NIDDM, groups of lean and obese SHR/Ntul//-*cp* rats demonstrating insulin resistance (IR) were fed diets containing 54% (w/w) carbohydrate as cornstarch (CS diet) or equal parts CS plus fructose (CSF diet) plus essential proteins fats, vitamins, minerals, and dietary fiber from one to nine months of age. The SHR/Ntul//-*cp* rat is a congenic animal model in which the only genetic difference between the phenotypes is the epigenetic expression of the obese (-*cp*) trait, where it is accompanied by the development of chronic IR and non-insulin dependent diabetes (NIDDM) soon after weaning in the obese phenotype. Weight gain of obese >> lean and was greater when fed the CSF than the CS diet in both phenotypes with either diet; Interscapular brown adipose tissue (IBAT) mass of obese >> Lean and was similar in both phenotypes. IBAT Mass : Body weight of Obese >> lean, and trended greater in lean CSF vs CS but less in Obese CSF vs CS. IBAT cell size of obese >> lean, and the CS vs CSF diet was similar in lean but decreased in the obese phenotype. IBAT cell number of obese >>> lean, while IBAT cell number of lean CSF > lean CS, but IBAT cell number of obese was similar with both diets. Lipoprotein lipase activity (LPL) of lean >> obese and trended to be greater with the CSF than the CS diet in both phenotypes, and IBAT tissue lipid content of obese >> lean with a trend toward CSF > CS in both phenotypes. These results indicate that IBAT development occurs via hyperplasia and hypertrophy in the Obese phenotype of this strain, and that long term consumption of the high Fructose diet enhances IBAT cellularity in the lean phenotype while the IBAT cellularity was maximally enhanced by both diets in the obese phenotype.

These results further indicate that the impact of long-term consumption of a high fructose diet impacts the development of IBAT mass and cellularity differentially in the lean and obese+NIDDM phenotype, likely at least in part due to contributions of longstanding IR in the obese+NIDDM animals. Because IR is known to impede cellular glucose uptake in isolated brown adipocytes as an essential process in the expression of cellular thermogenic responses, any potential mechanisms to override or bypass the process or to decrease the magnitude of IR would be presumed to exert a beneficial effect. Since fructose may enter tissues independently of insulin actions, it may enhance the potential for expression of IBAT development and the expression of non-shivering thermogenesis in response to alterations in diet and environment, but overall fructose consumption was found to be neither remarkably beneficial nor ameliorative in resolving critical parameters of brown adipose tissue expression as contributors to the development of obesity in the obese phenotype present study.

Keywords: Obesity, Brown Fat, NIDDM, Insulin Resistance, Rat

Synopsis

Brown adipose tissue of insulin resistant obese-diabetic (NIDDM) rats increased in mass and cellularity compared to their lean littermates when fed zero fructose- vs high fructose diets until 9 months of age. The increases in the lean phenotype fed the high fructose diet were consistent with additional hyperplasia, while the increases in IBAT in the obese phenotype were due to hyperplasia and hypertrophy, yielding a higher tissue lipid content and a greater cell lipid content. In contrast, the high fructose diet resulted in increases in brown adipocyte number in the lean but not in further increases in the obese phenotype. IBAT tissue lipid content was greater and IBAT LPL activity lower in obese than lean littermates, despite similar feeding and environmental environments. These data confirm differential carbohydrate-linked processes of brown adipocyte hyperplasia and hypertrophy development in the lean and obese phenotypes of this strain.

Introduction

The current trend in the global magnitude and prevalence of obesity is rapidly embarking on epidemic proportions.[1] Not only is the incidence on an upswing, the onset now occurs decades earlier in the lifespan of those who become affected, with some as early as early childhood or adolescence.[2] To date, many therapeutic approaches have been developed, but none are best suited for the clinical management of all cases due to the multiplicity of physiologic mechanisms they may individually or in combination result in the progressive development of obesity and its disordered elements of metabolism. In a recent Medscape article, the possible roles of brown fat in the progression and treatment were discussed, with an eye toward developing options that could elevate the activity of brown adipose tissue mediated energy expenditure in humans, as an adjunct in dietary and pharmacologic management of obesity and its numerous pathophysiologic sequela.[3] While diet and life style have always been the cornerstone of weight management treatments, specific dietary constituents including non-insulin dependent carbohydrate sources have been relegated to artificial sweeteners and received little attention as primary dietary constituents.[4]

Fructose is a unique monosaccharide, in that as a keto sugar it can obtain entry to peripheral tissues via GLUT 1 and GLUT 5 glucose transporters without the contributions of insulin actions, and therefor is often presumed to offer a metabolic and glycolytic advantage in conditions of insulin resistance and glucose intolerance.[5,6] Most peripheral tissues typically require insulin-linked GLUT4 transporters for cellular glucose uptake, a metabolic disadvantage in insulin resistance states.[7] Thus any carbohydrate moiety that could evade the insulin dependent pathway could offer a metabolic advantage in a variety of physiologic conditions. Once a metabolizable carbohydrate moiety enters the cell cytosol however it can undergo oxidation and support lipogenic activities including generation of acetyl CoA for mitochondrial oxidation and for *de novo* lipogenesis.[6] Thus, it is of interest to determine if fructose may be able to affect the growth and development of brown adipose tissue in a manner comparable to that of glucose or sucrose. The observation of brown adipose tissue has been known to occur in man and animals from cadaveric dissections for hundreds of years, but only during the recent half century have the physiologic contributions to metabolism and the thermogenic biochemical pathways in response to cold and nutritional factors been elucidated.[3,7-13.]

Brown adipose tissue differs from white adipose tissue in that the fundamental function of brown adipocytes is their ability to serve as a heat producing tissue in response to changes in diet and environment, while the primary function of white adipose tissue is energy deposition and as a reserve energy source during periods of energy deprivation or increased energy need.[14-16]

While fructose as a constituent of fruits and vegetables has always likely been a constituent of human and animal diets, where it typically occurs along with a plethora of other micronutrients, fibers and antioxidant phenolic compounds, in current society it has become a major dietary constituent often in the form of high fructose corn syrup (HFSC).[4] Since the evolution of HFCS in current Westernized diets, the per capita fructose intake has seen a 4 to 5-fold increase in human consumption in recent decades, and now appears to be approaching the upper limits of safe ingestion.[4,5,17]

Like excess glucose, fructose can undergo nonenzymatic dose related glycation reactions with plasma proteins via a Maillard reaction, rendering the glycated metabolites less physiologically active than when in their natural state following the glycation reaction.[17]Once formed, the glycated proteins remain in circulation for the duration of the lifespan of the respective modified proteins. Thus, clinical assessment of the magnitude of glycated proteins in the form of hemoglobin A_{1c} or serum fructosamine can serve as valuable clinical markers for the extent of glycemic control and are highly useful parameters in the clinical management of diabetic and glucose intolerant states of metabolism. The glycation may impact any available amino nitrogen group, whether it be in plasma proteins or erythrocytes, or in exposed plasma membrane proteins. Thus, despite the observation that fructose may undergo cellular uptake independently of insulin or GLUT4 glucose transporters, glucose remains the favored carbohydrate substrate as an energy source for most aspects of cellular oxidation and metabolism.[6,17.]

The cellular morphology of the two types of adipose tissues also differs significantly, as in white adipose tissue the adipocytes typically have a single large lipid droplet encompassing up to 90% of the cellular volume and a flattened nucleus located in a thin cytoplasmic ring surrounding the lipid droplet.[9,14,15] In contrast, in brown adipocytes, they typically contain multiple small lipid locules embedded in a mitochondria-rich cytoplasm throughout the extranuclear region of the cell, which has a spherical, centrally located and well-defined nucleus. The size of the lipid droplets of white adipocytes vs. the smaller lipid locules found in brown adipocytes plays a significant role in the efficiency of lipid mobilization, in that the smaller locules provide a greater enzymatically active surface area to mass ratio than the larger lipid droplets, thereby enabling the brown adipocytes to adjust to changes in nutritional or environmental conditions more rapidly and with greater efficiency of lipid mobilization as changes in metabolic requirements arise.[6,17,18] Insulin serves as a stimulator of lipogenesis in adipose tissues, enabling cellular lipid stores to increase, while catecholamines such as epinephrine or norepinephrine can act as a stimuli for lipid mobilization in adipocytes and glycogen mobilization in liver. Thus, the nutritional and environmental stimuli that modulate the activities of the two hormones is a key regulator of cellular activity in both types of adipocytes. Both white and brown adipocytes are now considered to have endocrine functions, via the release of hormonally active compounds that contribute to appetite and feeding behavior, including satiation.[18,19.]

In previous studies with genetically lean and obese rats, the obese phenotypes characteristically demonstrate decreases in the process of non-shivering thermogenesis in response to alterations in diet and environment.[20,21] In addition, the obese phenotypes of both the LA/Ntul//-*cp* and SHR/Ntul//-*cp* rats which both share the same epigenetic trait for obesity (the -*cp* trait) demonstrate increased IBAT mass, and an apparent 'unbrowning' of their brown adipose tissue depots as they enter adulthood.[21] The loss of the deep brown coloration towards an intermediate, less color intense presentation is associated with insulin resistance, and is also indicative of decreases in thermogenic activity. In addition, anti-thermogenic agents are also associated in increases in the lipid volume of the typical lipid locules found in brown adipose tissue.[15] Morphologically, the isolated brown adipocytes contain larger lipid locules and greater cellular lipid content following thermogenic inactivating agents including insulin resistance.[14,15.]

Rothwell and Stock were among the first investigators to demonstrate the existence of diet induced thermogenesis (DIT) in cafeteria fed rats.[22,23] This observation occurred in animals that had been induced to overeat via the offering of palatable high calorie diets containing a variety of human food items for several weeks, and subsequently the authors were also able to demonstrate thermogenic activity in humans in anatomic regions where brown fat depots are known to be located.[22,23] Himms-Hagen and others have further confirmed the phenomena of DIT in addition to elucidating the physiologic and biochemical mechanisms through which it occurs.[11-13] More recently, it has been demonstrated to consist of both sympathetic and thyroidal hormonal components which complement each other respectively during acute vs longer acting expressions of the thermic activity.[18,24,25] Only a few studies have examined the changes in adipocyte cellularity in BAT depots however, as the traditional

methods established by Hirsch and Gallian for determining adipocyte cell numbers in WAT could not be readily applied to brown adipose tissue due to the multilocularity of the lipid content, as the classic osmium tetroxide methods focus on the fixation of the lipid droplets, and the multiple locules of lipid in the brown adipocytes proved difficult to correlate with the actual cell number.[26-28] Tulp et al established a double fixation modification of the Hirsch and Gallian technique that enabled a more accurate determination of the number of intact brown adipocytes.[27,28] The purpose of the present study was to determine the characteristics of brown adipose tissue cellularity in lean and obese+NIDDM rats fed isoenergetic diets where the carbohydrate source constituted of 54% (w/w) cornstarch or equal parts cornstarch and fructose. In addition to 20% protein, 16% fat plus essential vitamins minerals and non-nutritive fibers, with the presumption that the high fructose dietary regimen would exert a lower glycemic index due to the lower glycemic index of fructose (G.I. = 32 vs 100 for glucose), thus diluting the glycemic index of cornstarch, and exert a fructose-sparing effect on post prandial plasma insulin dependent actions, and which if extended over a long duration, may reduce the magnitude of insulin resistance in the obese+NIDDM animals.[29,30]

Methods

Groups of make lean and congenic obese SHR/Ntul//-*cp* rats originally obtained from the congenic breeding colony at NIH (n=11-12 rats/group) were fed an isoenergetic diet from ~1.5 until 9 months of age. Initial body weights of the lean and obese animals were 90.35±5.9 vs 102.65 grams± 8 respectively (p=n.s.). The diet consisted of (w/w) 54% CHO as cooked cornstarch (CS diet) or equal parts CS and fructose (CSF diet) plus 20% protein (equal pats casein and lactalbumin) 16 %fat (as equal parts lard, corn oil, beef tallow and coconut oil) plus essential vitamins, minerals, (AIN vitamin /mineral mix), and non-nutritive fiber fed *ad libitum*, from ~1.5 until 9 months of age.[30] All animals experienced the same environmental conditions, including being housed in plexiglass shoebox cages in littermate pairs (I lean plus 1 obese) , 20-22°C room temperature and 50% relative humidity with a reverse light cycle (light 2000-0800 hrs.). Live body weights were obtained initially and periodically throughout the study. At the end of the study, rats were sacrificed by cervical dislocation with a small animal guillotine, truncal bloods collected for later study and the interscapular brown fat depots carefully dissected free of white adipose tissue, weighed to the nearest 0.1 mg to determine fat pad mass, and measures of brown adipocyte size and number determined in representative sections of the tissue as described previously.[27,28] Briefly, the tissue aliquots were fixed in 10% buffered formalin for 24-48 hours, post-fixed with 4% osmium tetroxide for an additional 48-72 hours, washed and sieved with Nitex filters to remove extraneous debris, and counted in a Coulter Model B particle counter.[27,28] Cell and tissue lipid content were determined gravimetrically in weighed aliquots of tissue with the method of Dole and Meinertz as performed in our laboratory and expressed a micrograms of lipid per cell and as a percent lipid in the tissue fragment.[27,28] Measures of lipoprotein lipase activity per gram of tissue were determined via the method of Shirai and Jackson[31]and expressed as uMols of FFA released per gram of tissue per hour at physiologic temperatures (37°C). Data were analyzed via standard descriptive and statistical methods including ANOVA and trend analysis.[32,33] This study was approved by the Institutional Ethics, Animal Care and Use Committee (IEACUC) of the University of Science Arts and Technology, Montserrat.

Results

The effect of feeding a high-fructose (CSF) vs a zero-fructose cornstarch (CS) based diet on body weight gain is depicted in Figure 1 and indicates that weight gain in the obese phenotype far exceeded that in their lean littermates when 9 months of age. In addition, the effects of the high fructose diet (the CSF diet) resulted in a trend toward only modestly greater weight gain in the obese phenotype than when fed the isoenergetic CS diet and house water, *ad libitum*, while the differences in weight gain in the lean phenotype were not significant. The differences in weight gain are significant in that the starting weights of the lean and obese animals were not significantly different prior to introduction of the semisynthetic diets.(Starting weight lean = CS 90.0 ± 6.0; lean CSF 89.9±5.8; Obese CS = 98.0±9.0; Obese CSF 107.3±8.0 grams BW; ANOVA P = n.s.).

The effects of diet and phenotype on brown adipose tissue mass are depicted in Figure 2 and indicate that the IBAT mass of the obese phenotype far exceeded the IBAT mass of their lean littermates, both in absolute terms of grams per depot and as a proportion to final body weight. The differences in IBAT mass and IBAT to Body Weight appear virtually irrespective of the diet consumed although the IBAT Mass to body weight ratios of the CSF obese animals tended to be modestly lower likely due to the modestly lower final body weights of obese CS fed animals (Obese CS = 820±32 g vs Obese CSF = 865 ± 36 g final body weights) combined with a similarity in IBAT mass in the two

obese+NIDDM groups, thereby resulting in a modest decrease in the IBAT:BW ratio in the obese CSF group.

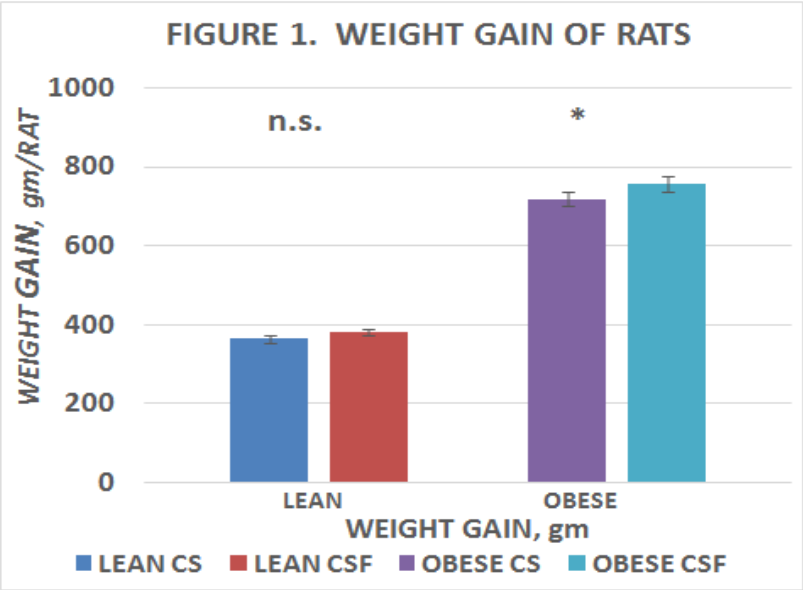

Figure 1. Effect of dietary CHO type on weight gain of rats. Data are mean ± 1 SEM, n=11-12 rats/group. ANOVA P=<0.001 for phenotype; p = ns for diet in lean CS vs CSF; p = <0.05* (trend) for diet in obese cs vs obese CSF. CS = cornstarch; CSF = cornstarch+fructose. All diets and house water were fed *ad libitum*.

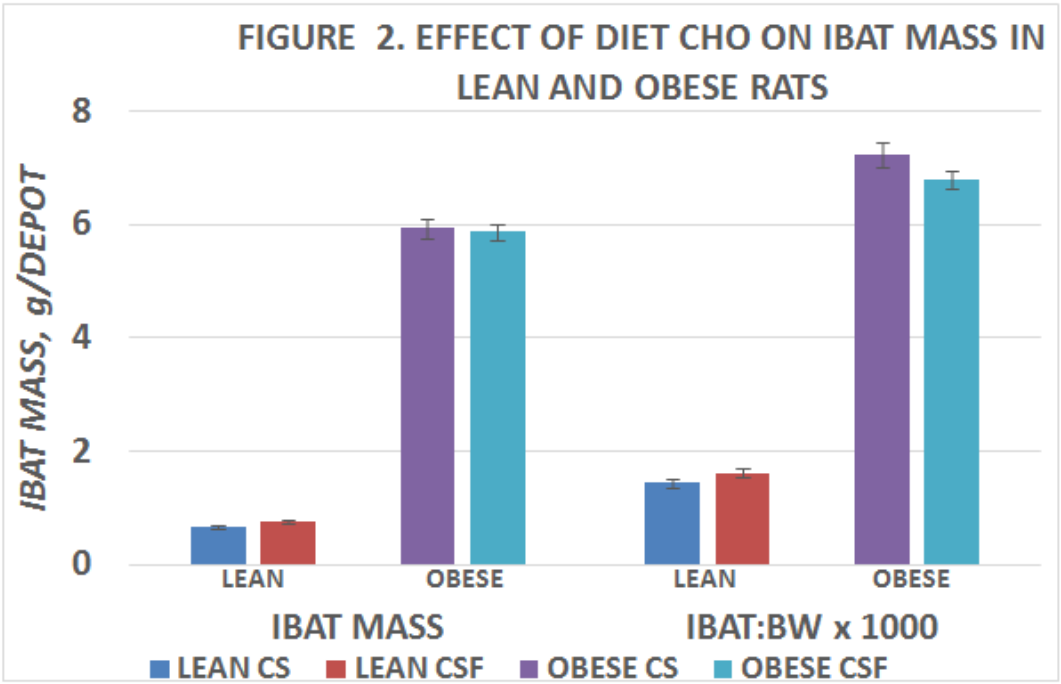

Figure 2. Effect of diet and phenotype in IBAT mass in lean and obese rats. Data are mean ± 1 SEM, n=11-12 rats/group. ANOVA P=<0.001 for phenotype; p = ns for diet lean CS vs CSF; p = <0.05* (trend) for diet in obese cs vs obese CSF.

The effects of diet and phenotype on IBAT adipocyte lipid content are depicted in Figure 3 and indicate that adipocyte cellular lipid content of obese was significantly greater than in their lean littermates, irrespective of the diet consumed. In addition, the diet was without effect on cell lipid content in the lean phenotype (Lean CS = Lean CSF) while in the obese phenotype, the likely insulin-sparing effects of the lower glycemic index of the CSF diet may have resulted in less cellular lipid content accumulation in the obese phenotype (p = <0.05).

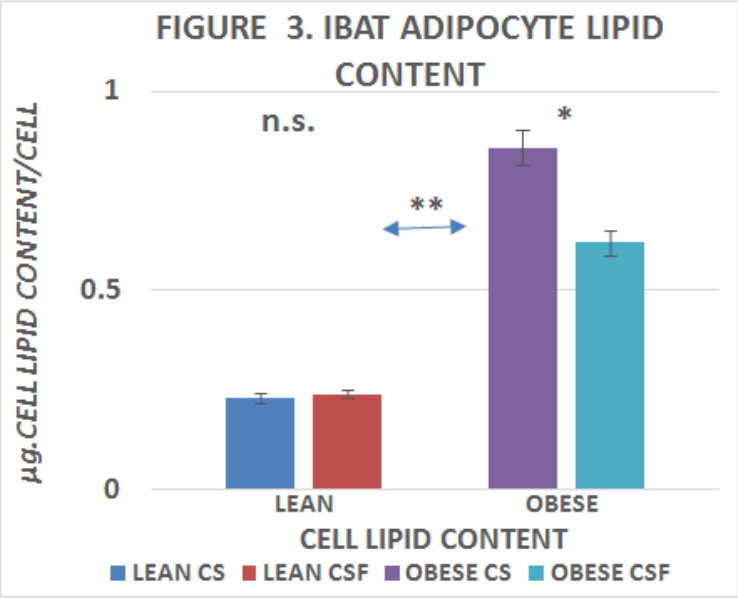

Figure 3. Effect of diet and phenotype on brown adipocyte lipid content. Data are mean ± 1 SEM N=11-12 rats / group. ANOVA phenotype p = < 0.01; CS vs CSF n.s in lean, P = < 0.05 in obese CS vs CSF.

The effects of diet and phenotype on brown adipocyte number are depicted in Figure 4 and indicate that the IBAT cell number of obese was significantly greater than occurred in their lean littermates while consuming the same respective CS or CSF diets. In contrast, the IBAT cell number of the lean was greater when fed the CSF than the CS diet, while final adipocyte numbers per IBAT depot were similar in both obese CS and CSF groups.

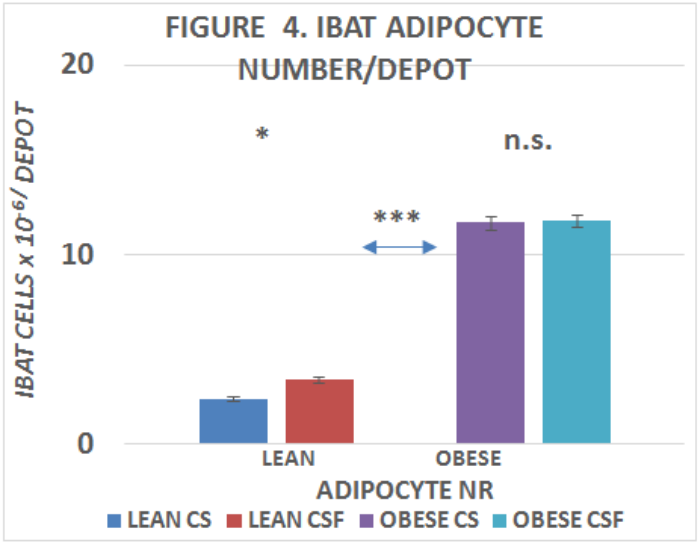

Figure 4. Effects of diet and phenotype on IBAT cell number per depot in lean and obese rats. Data are mean ± 1 SEM, n- 11-12 rats / group. ANOVA p = < 0.001 for phenotype (lean vs obese). P = < 0.05 for diet in lean (CSF vs CS; p = n.s. for obese CS vs CSF.

The effects of diet and phenotype are tissue lipoprotein lipase activity are depicted in Figure 5 and indicate that LPL activity of the lean phenotype is greatly increased compared to their lean littermates. In addition, the effects of the lower glycemic index CSF diet resulted in increases in LPL activity in both phenotypes, consistent with a decrease in possible inhibitory insulinogenic actions in both phenotypes when fed the high fructose dietary regimen.

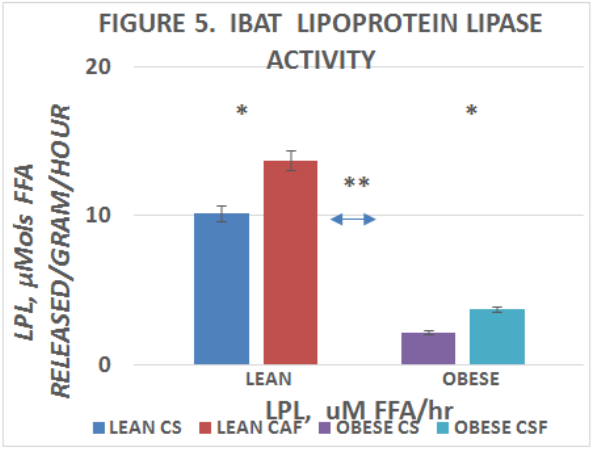

Figure 5. Effects of diet and phenotype on IBAT lipoprotein lipase activity in lean and obese rats. Data are mean ± 1 SEM, n= 6 rats/group. ANOVA p= 0.01 for phenotype; p = < 0.05 for diet in both phenotypes. FFA = free fatty acids.

The effects of diet and phenotype on IBAT tissue lipid content are depicted in Figure 6, and indicates that tissue lipid content, expressed as percent lipid (w/w) is greater in the obese than the lean phenotype. In addition, the effects of diet resulted in a modest trend (CSF > CS) in both phenotypes, corresponding with the LPL activity in the same tissues.

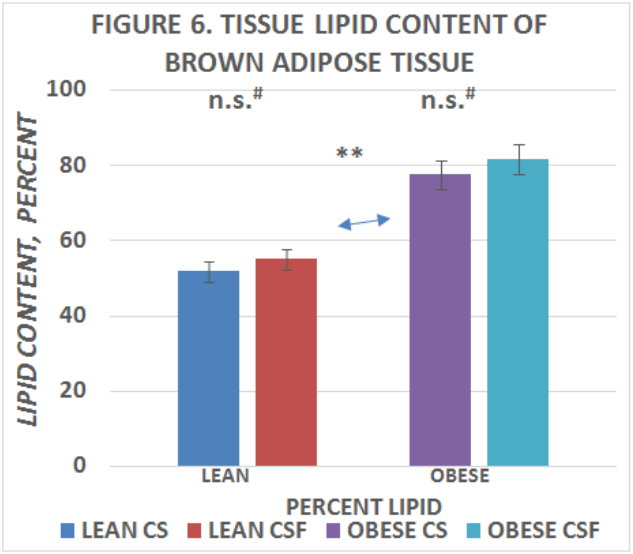

Figure 6. Effects of diet and phenotype on IBAT tissue lipid content in lean and obese rats. Data re mean ± 1 SEM, N = 10-12 animals / group. ANOVA p = < 0.01 for phenotype (lean vs obese; p = n.s. for CS vs CSF diet with modest trend for diet effects (CS vs CSF; p=<0.10[#])

Discussion

The effects of this study indicate that final weight gain and Interscapular brown adipose tissue mass and cellularity are significantly greater in the obese than the lean phenotype, and the effects of a high fructose diet resulted in only modest increases in body weight gain, and in differential effects on IBAT cellularity and cell lipid content. The CSF diet resulted in greater IBAT cell number in the lean phenotype, but the CS vs CSF diets were without further effect in the already increased cell number in the obese phenotype. Brown adipocyte proliferation tends to increase chronologically during early, pre-adult growth stages, and the increases in cell number have been reported to remain constant thereafter and remain well into adulthood. Thus, the significantly greater cell number in the obese phenotype of this study are likely to remain present, albeit likely less thermogenically active in adult life stages as suggested by the greater cellular and depot lipid content. These observations are consistent with those of Tanuma et al from cadaveric dissections, where the histology from older individuals reflected a similar observation.[8,9] In contrast, the CSF diet resulted in modestly greater brown adipocyte numbers per IBAT depot in the lean phenotype, possibly due to fructose-induced hyperphagia during their post weaning development while consuming the fructose diet. Fructose is sweeter than cornstarch, but it is not clear if the greater sweetness may have been perceived more favorably in the animals, as it often is in humans. In an earlier study in the LA/Ntul//*-cp* strain, isocaloric feeding of partially hydrolyzed cornstarch was preferred over sucrose when added to the drinking water.[34] Hyperphagia during postweaning growth has been associated brown adipocyte hyperplasia in other rodent models, and may reflect a compensatory mechanism in an attempt to expend the excess calories when caloric intake exceeds requirements, or when macronutrient imbalances indicate a physiologic need to expend the excess calories in order to preserve body composition and homeostasis in the lean phenotypes, but which physiologic mechanism may fail in the obese phenotype.[28,34-37]

In other studies, the obese phenotype consistently demonstrates a lower rate of resting and norepinephrine-simulated thermogenic energy expenditure, in addition to an economy of energy metabolism due to a significantly greater efficiency of protein turnover, a major constituent of the energy costs of intermediary metabolism.[36-38] Protein synthesis, at 4 kcal/peptide bond formed, remains one of the energetically most expensive metabolic processes of mammalian species, and thus the relative economy in the rates if protein synthesis and protein degradation afforded in insulin resistance represent a strategic physiologic measure to improve the efficiency of intermediary metabolism, similar to that which occurs in starvation or undernutrition to prolong life in an otherwise healthy animal. In the pre-obese phenotype of the LA/Ntul//*-cp* rat, which shares the same epigenetic trait for the expression of obesity (the *-cp* trait) the rate of protein turnover in the obese phenotype were found to be decreased by an average of 50% prior to the development of frank obesity, without decreasing energy intake, thereby representing an energetic and metabolic advantage on the initiation of the process of excess fat accretion.[21,37] While it was not possible to determine if the same phenomenon was present in the present study, the likelihood clearly exists as both strains of the corpulent rat express the same epigenetic trait for fat accretion in the presence of insulin resistance from an early age.[20,37]

The observation of differences in IBAT lipoprotein lipase activity are a new finding in the obese of this strain. LPL is hormonally controlled in large part by actions of insulin and catecholamines including epinephrine and norepinephrine: Insulin acts to stimulate lipogenesis, and LPL acts to mobilize the stored triglycerides as free fatty acids during conditions of energy needs.[7,17,39-43] Catecholamines in contrast, facilitate the mobilization of metabolic fuels including glucose and free fatty acids, both of which are essential for the molecular expression of thermogenic activity in brown fat. The differences in greater LPL activity of brown adipose tissue of the present study are consistent with the marked differences in IBAT tissue lipid content, and the previously reported incidence and magnitude of insulin resistance in the obese phenotype of this strain.

Insulin resistance, in concert with glucocorticoid dysregulation are a hallmark of obesity and NIDDM, and collectively exert negative influences on the intracellular formation, mobilization, intracellular transport, and transmembrane actions regarding the insulin-dependent GLUT4 glucose transporters. In contrast, fructose enters calls via GLUT 1 or GLUT 5 transporters, thereby bypassing the insulinogenic effects.[5,6] Once internalized, however, both glucose and fructose can impinge on glycolytic and lipogenic pathways, with fructose essentially resulting in a glucose-sparing effect. In high dietary doses of fructose however, fructose may overburden lipogenic pathways in addition to contributing to alterations in renal dynamics, and adversely impacting on pyrimidine metabolism resulting in the formation of excess uric acid, with its attendant adverse effects on the development and progression of the pathophysiologic symptoms of gout and cardiovascular disease.[44-47] Thus, the consumption of excess fructose whether in the form of free sugar, as a hydrolytic digestive product of sucrose, or via excess consumption of high fructose corn syrups poses a potential dose-related risk not only to renal and cardiovascular parameters, but may also impact on brown adipose tissue dynamics, especially when consumed in excess during early post weaning life in the rat. Observations during dissections revealed a high incidence of renal calculi of unknown composition in the obese+NIDDM animals fed the CSF diet, consistent with the published effects of excess fructose consumption in clinical observations.[44-47.]

In conclusion, the consumption of a high fructose diet for up to 9 months of age in a congenic, insulin-resistant, diabetic (NIDDM) animal model, resulted in significant increases in the mass and cellularity of brown adipose tissue, with only moderate effects on brown adipocyte hyperplasia in the similarly fed lean phenotype. The differences in IBAT lipid content were consistent with diet and phenotype differences in IBAT lipoprotein lipase activity in the lean vs the obese animals. Thus, the effects of a high fructose diet over an extended duration were neither beneficial nor ameliorative overall in parameters of weight gain in the obese+NIDDM animals. In addition, the fructose diet was found to modestly enhance brown adipocyte hyperplasia but not brown adipocyte hypertrophy in the lean phenotype, while the epigenetic expression of the -*cp* trait for obesity resulted in large increases in IBAT mass, tissue lipid content, brown adipocyte cellularity, and in the apparent 'unbrowning' of the brown adipose tissue in the insulin resistant obese phenotype regardless of the dietary CHO source provided.

Acknowledgements

The author acknowledges the generous Institutional support of the University of Science Arts and Technology, Montserrat for the development of this manuscript. There are no conflicts of interest in the conduct and development of this manuscript or the research leading up to its completion.

References

1. World Health Organization. Obesity and Overweight. 2021.
2. CDC, Prevalence of Childhood Obesity in the United States. Division of Nutrition, Physical Activity, and Obesity, National Center for Chronic Disease Prevention and Health Promotion, CDC report. 2022.
3. Davenport L. Unlocking the Secrets of Brown Fat. Medscape**.** News & Perspective Drugs & Diseases CME & Education Academy Video Decision Point Perspective . Medscape Diabetes & Endocrinology. 2023. Pp 1-4.
4. Bray GA. Energy and Fructose from Beverages Sweetened with Sugar or High-Fructose Corn Syrup Pose a Health Risk for Some People. *Adv Nutr*. 2013;4:220–225.
5. Douard V, Ferraris RP. Regulation of the Fructose Transporter GLUT5 in Health and Disease. *Am J Physiol Endocrinol Metab*. 2008;295: E227–E237.
6. Granner DK, Mayes PA, Murray RK, et al. Harpers Illustrated Biochemistry, McGraw-Hill Pubs. 2012.
7. Gessner K. Historie Animalium: Lib I De Quadrupedibus viviparis. 1554.
8. Tanuma Y, Tamamoto M, Ito T, et al. The occurrence of brown adipose tissue in perirenal fat in Japanese. *Arch Histol Jpn*. 1975;38(1):43-70.
9. Tanuma Y, M Ohata M, Ito T. Possible function of human brown adipose tissue as suggested by observation on perirenal brown fats from necropsy cases of variable age groups. *Arch Histol Jpn*. 1976;39(2):117-45.
10. Cannon B, Nedergaard J. Brown Adipose Tissue: Function and Physiological Significance. *Physiol Rev*. 2004; 84:277–359.
11. Himms Hagen J.Brown adipose tissue thermogenesis, energy balance, and obesity. *Can J Biochem Cell Biol*. 1984;62(7):610-617.
12. Himms Hagen J. Thermogenesis in brown adipose tissue as an energy buffer. Implications for obesity. *N Engl J Med*. 1984;311(24):1549-1558.
13. Bukowiecki LJ, Collet AJ, Follea N, et al. Brown adipose tissue hyperplasia: a fundamental mechanism of adaptation to cold and hyperphagia. *Am J Physiol*. 1982;242: E353-E359.
14. Tulp OL, D Root, R Frink. Pharmacologic-induced alterations in brown adipose tissue morphology. *Anat Rec*. 1981;199(3):259-260A.

15. Tulp OL, D Root, R Frink. “The effect of anti-hypertensive drug treatment on brown adipocyte diameter and locule distribution in rats.” *Comp Biochem Physiol*. 1984;79C(2):317-320.
16. Tulp OL, D Root, R Frink. “The effect of anti-hypertensive drug treatment on brown adipocyte diameter and locule distribution in rats.” *Comp Biochem Physiol*. 1984;79C(2):317-320.
17. Dills WL. Protein fructosylation: fructose and the Maillard reaction. *Am J Clin Nutr*. 1993;58(5 Suppl):779S-787S.
18. Danforth E. Hormonal control of thermogenesis. *Life Sci*.1981; 28:1821–1827.
19. Rahman MS, Einstein GP, Tulp OL. Autonomic, Immunological and Endocrine influences on adipose tissue as an organ. *Adv Obes Weight Manag Control*. 2021:11(2):48-58.
20. Tulp OL. Characteristics of thermogenesis, obesity, and longevity in the LA/Ntul//-cp rat. *ILAR J*. 1990;32(3):32-38.
21. Tulp OL. Does insulin resistance contribute to the ‘unbrowning’ of brown adipose tissue? *Academia Biology*. 2023;1:1–6.
22. Rothwell Stock Rothwell NJ, Stock MJ.A role for brown adipose tissue in diet-induced thermogenesis. *Nature*. 1979;281(5726):31-35.
23. Brooks SL, Rothwell NJ, Stock MJ. Increased proton conductance pathway in brown adipose tissue mitochondria of rats exhibiting diet-induced thermogenesis. *Nature*. 1980;286(5770):274-276.
24. Tulp Orien L. Revisiting the relative contributions of sympathetic and thyroidal actions to adaptive thermogenesis in man and animals. *African Journal of Internal Medicine*. 2023;11(4):pp. 001-007.
25. Tulp OL. Estimation of the Sympathetic and Thyroidal Partitions to Diet Induced Thermogenesis in the Rat. In: New Advances in Medicine and Medical Science. 2023;3(1),53-66.
26. Hirsch J, Gallian E. Methods for the determination of adipose cell size in man and animals. *J Lipid Res*.1968;9(1):110-119.
27. Tulp OL, Gambert S, Horton ES. Adipose tissue development, growth, and food consumption in protein malnourished rats. *J Lipid Res*. 1979;20(1):47-54.
28. Tulp OL. The development of brown adipose tissue during experimental over-nutrition in rats. *Int J Obesity*. 1981;5(6):579-591, and The Adipose Tissue Issue, John Libbey & Company, London, pp. 579-591. 1981.
29. Michaelis OE, Carswell N, Ellwood KG. Metabolic characteristics of the LA/N-Corpulent and SHR/N-corpulent rat strains. In: New Models of Genetically Obese Rats for Studies in Diabetes, Heart Disease, and Complications of Obesity. NIH publication, Division of Research Services, Veterinary Resources Branch, National Institutes of Health, Bethesda, MD pp. 13-15. 1988.
30. Michaelis OE, Ellwood KC, Tulp OL, et al. Effect of feeding sucrose or starch diets on parameters of glucose tolerance in the LA/N-corpulent rat. *Nutr*
31. Shirai, K, Jackson RL. Lipoprotein lipase-catalyzed hydrolysis of p-nitrophenyl butyrate. Interfacial activation by phospholipid vesicles. *J Biol Chem*. 1982;257(3):1253-1258.
32. Ott L. Multiple comparisons. In: An introduction to statistical methods and data analysis. 3rd Ed. PWS-Kent Publishers, Boston MA. 1988. Pp 437-496.
33. Page EB. Ordered hypothesis for multiple treatments: A significance test for linear ranks. *J Amer Stat Assn*. 1963;58(301):216-230.
34. Tulp OL, Carlin C, Tulp OL. “Carbohydrate overnutrition induces increased adiposity in rats”. *Obesity Update*. 1993.p.1-2.
35. Tulp OL. Effect of delayed luminal carbohydrate uptake via α-glucosidase inhibition on plasma glycemic parameters and lipid profiles in adult wistar fatty rats. *Gastroenterol Hepatol Open Access* 2022;13(5):168–173.
36. Tulp OL, ES Horton. The effect of prolonged experimental protein malnutrition and of refeeding on growth and adipose tissue development in the rat. *J Nutr*. 1981;111(7):1145-1156.
37. Biometry paper Tulp OL. Biometry, Adiposity and Mechanism of Protein Sparing Growth in Congenic Preobese LA/Ntul//-cp rats. *British Journal of Healthcare and Medical Research*. 2023;10(3):364-374.
38. Shirai, K, Jackson RL. Lipoprotein lipase-catalyzed hydrolysis of p-nitrophenyl butyrate. Interfacial activation by phospholipid vesicles. *J Biol Chem*. 1982;257(3):1253-1258.
39. Carter SU, Okamoto K. Effects of insulin and glucocorticoids on glucose transporters in rat adipocytes. *Am J Physiol*.1987;252(4):E-441-E-453.
40. Huang HL,Young AA, Koda JE. Hyperamylinemia, hyperinsulinemia, and insulin resistance in genetically obese LA/N-*cp* rats. *Hypertension*. 1992;19 (Suppl 1):1101-1109.
41. Tulp OL. Differential depot-specific parameters of adipose tissue cellularity and in preobese congenic LA/Ntul//-*cp* rats. *AOWMC*. 2023;13(4):81-85.
42. Tulp OL, Awan AR, Einstein GP. Enhanced Caloric Efficiency Contributes to Adiposity in LA/N//-*cp* Corpulent Rats. *Experimental Biology*. 2021;35.
43. Tulp OL, Einstein GP, Rizvi AAA. Can Caffeine and Norepinephrine Activate your Brown fat? An Experimental Study on Groups of Lean and Obese LA/Ntul//-*cp* Rats. Ch 14 In: Current Overview on Pharmaceutical Science, Vol 8, (Ed: S.A. Rizvi), 2023;pp.162-180.
44. Anderson ME, Tulp OL. The Effects of High Dietary Fructose Consumption on the Development of Gout (Review). 2023.
45. Legeza B, Marcolongo P, Gamberucci A, et al. Fructose, Glucocorticoids and Adipose Tissue: Implications for the Metabolic Syndrome. *Nutrients*. 2017;9(5):426.

46. Vos MB, Kimmons JE, Gillespie C, et al. Dietary Fructose Consumption Among US Children and Adults: The Third National Health and Nutrition Examination Survey. *Medscape J Med*. 2008;10(7):160.
47. Stanhope KL, Havel PJ, Fructose consumption: Recent results and their potential implications. *Ann N Y Acad Sci*. 2010;1190:15–24.