Hypothalamic Inflammation and Glioses as Aetiopathogenetic Factor in High Fat Diet Induced Obesity and Various Therapeutic Options to Resolve it

Kulvinder Kochar Kaur, MD1; Gautam Allahbadia, MD (Obgyn & Gynaec), DN2; Mandeep Singh, MD, DM (Neurology)3

1Scientific Director, Dr. Kulvinder Kaur Centre for Human Reproduction, 721,G.T.B Nagar, Jalandhar 144001, Punjab, India
2Scientific Director, Rotunda-A Centre for Human Reproduction, 672, Kalpak Garden, Perry Cross Road, Near Otter’s Club, Bandra (W) 400040, Mumbai, India
3Consultant Neurologist, Swami Satyanand Hospital, Near Nawi Kachehri, Baradri, Ladowali road, Jalandhar, Punjab, India

ABSTRACT

We reviewed the literature relating to hypothalamic inflammation (HI); gliosis in relation to high-fat diet (HFD) and that how this could be reversed with various types of therapies. We searched PubMed articles with the MeSH terms “hypothalamic inflammation”, “gliosis”, “HFD”, “obesity”, and “treatments” used. During HFD intake, we found that the ventromedial hypothalamus (VMH) astrocytes uses fatty acids (FA’s) to generate ketone bodies which are then exported to neurons where they produce excess adenosine triphosphate (ATP) and reactive oxygen species (ROS), which overrides CD36 mediated FA sensing and role of astrocyte-derived ketone bodies in reducing calorie intake in diet resistant but not diet-induced obese strains was emphasized. The further role of HAM-RS2—a special starch, resolvins, abscisic acid, KBH1, unsaturated fatty acid receptor targeting GPR120/GPR40. Hepatic clock genes were effective in tackling obesity. We found that in rodents hypothalamic inflammation and glioses were found to occur immediately with HFD consumption before any significant weight gain. Sensitivity or resistance to diet-induced obesity in rodents also correlates with the presence or absence of hypothalamic inflammation and reactive glioses. Further functional interventions with the increase or decrease inflammation in neurons and glia alter diet associated weight gain. Various human magnetic resonance imaging (MRI) studies have found glioses and disrupted connectivity in obese humans. Various factors which can be used to tackle obesity like HAM-RS2—a special starch, resolvins, abscisic acid, KBH1, unsaturated fatty acid receptors, GPR120 and GPR40 are some of the explored routes by which these pathways may be explored to prevent the further extension of the HFD and one may get newer answers for arresting obesity development.

KEY WORDS: Hypothalamic inflammation; Glioses; Obesity; Resolvins; Abscisic acid; KBH1; Hepatic clock; Astrocytes; Ketone bodies.

INTRODUCTION

In our previous articles on trying to elaborate aetiopathogenetic factors in obesity we discussed various aspects of factors causing obesity, including microRNA’s, role of brown adipose tissue (BAT) metabolism, fibroblast growth factor 21 (FBG21) and in reviewing nutrient metabolism, special stress on fatty acid metabolism was emphasized and how hypothalamic inflammation (HI) precedes the development of obesity in high fat diet (HFD).1-8 We further highlighted the role of gliosis with HI in this article along with various ways of considering therapeutic action to counter these changes as an effective way of tackling obesity.

HYPOTHALAMIC INFLAMMATION (HI) AND GLIOSIS

A growing literature in rodent models suggests that obesity is associated with inflammation of and injury to hypothalamic areas critical to the control of energy balance and glucose imbalance.9-13 Histologically, this response is characterized by gliosis, the proliferation, and activation of glial cells induced by the response to central nervous system (CNS) injury. Microscopically, gliosis means infiltration of microglia and astrocytes and astrocytic proliferation including the increased density of astrocyte processes on the cell bodies of neurons.

Feeding HFD to rodents triggers inflammation and gliosis in the arcuate nucleus located in the mediobasal hypothalamus (MBH), even before obesity occurs9,12 and eventually reduces proopiomelanocortin cell number.9 Such changes are associated with both obesity and impaired glucose homeostasis in rodents13-15 and they offer an explanation for obesity-associated resistance of hypothalamic neurons to humoral signals like leptin and insulin.16,17 Even though evidence exists from animal studies, the significance of this hypothalamic glioses in humans had largely been unknown (Figure 1).16

A core concept of current research is that glioses can be detected in humans using magnetic resonance imaging (MRI) by assessing for increased signal brightness on a T2 weighted image.9,21 Clinically the visual identification of increased T2 signal intensity is used to detect CNS insults like stroke and multiple sclerosis (MS),20,21 but quantitative techniques can detect more subtle alterations in CNS tissue characteristics.11,22 One prior retrospective study on humans utilized clinical MRI examination and found a positive association between body mass index (BMI) and ratio on the T2 signal in the MBH as compared to the amygdale.9 Thus Schur et al in recent studies utilized a quantitative MRI technique to measure T2 relaxation time in the MBH, employing a dedicated sequence not typically utilized in clinical imaging protocols. Using a similar sequence they found longer MBH T2 relaxation times in diet-induced obese (DIO) mice compared to chow-fed controls.11,22 Thus using 2 separate studies they sought evidence for MBH gliosis in human studies. In study 1, an in vivo MRI study it was hypothesized that MBH gliosis when present would be associated with obesity and insulin resistance. In study 2, a postmortem study of human brain tissue hypothesized that T2 relaxation time would be related to immunohistochemistry (IHC) measures of astrocytes in the MBH.

Schur et al examined 67 patients undergoing a fasting blood draw and MRI. Cases with radiologic evidence of MBH gliosis [n=22] were identified as the upper tertile of left MBH T2 relaxation time and were compared to controls [n=23] from the lowest tertile. Besides a separate postmortem study brain slices [n=10] through the MBH was imaged by MRI and stained for glial fibrillary acidic protein (GFAP). In all the participants, longer T2 relaxation time in the left MBH was associated with

Figure 1: Model Depicting the Hypothalamic Response to an HFD in Animals Predisposed to DIO

A) AgRP and POMC Neurons are Integral Components of Energy Balance Neurocircuitry Located in the ARC Nucleus, Situated Adjacent to Third Cerebral Ventricle (3V) Along the Floor of the Hypothalamus. The Activity of these Neurons is Sensitive to Input from Circulating Hormones (e.g., Leptin and Insulin) and Nutrients, and it Plays an Important Role to Establish the Defended Level of Body Weight. B) Recent Evidence Suggests that During HFD Feeding these Neurons may be Injured Via Unknown Mechanisms, and that this Injury Triggers Activation of Local Glial Cell Populations (Astrocytes and Microglia). This Neuron Injury and Reactive Glioses Can, in Turn, Impair Homeostatic Control of Body Fat Stores, Leading to Increased Body Weight.91
higher BMI ($p<0.01$). As compared to the control cases, the participants had longer T2 relaxation times in the right MBH ($p<0.05$) as well as higher BMI ($p<0.05$), fasting insulin concentrations ($p<0.01$) and HOMAIR values ($p<0.01$) adjusted for sex and age. Elevations in insulin HOMA IR were also independent of BMI. In the postmortem study, GFAP staining intensity was positively associated with MBH T2 relaxation time ($p<0.05$) validating an MRI based method for the detection of MBH gliosis in humans. Hence they concluded that these findings links hypothalamic gliosis to insulin resistance in humans and suggests that the link is independent of the level of adiposity.23

**ASTROCYTES AND HYPOTHALAMIC GLOISES**

Data from recent studies suggests that neuronal inflammation may be a downstream event during DIO, with recruitment and activation of hypothalamic glial cells being more proximal response to HFD exposure.9,11,24-26 This gliosis process involves accumulation and multiplication of activated microglia and astrocytes in the region of MBH 9,11,24-26 Various studies have implicated microglia in the development of diet-induced inflammatory signals along with metabolic dysfunction,14,28 but a similar role for astrocytes is not clear. Buckman demonstrated a modest role of astrocytic inflammation to caloric intake on the first day of HFD feeding but there was no analysis of susceptibility to DIO.29 There are abundant astrocytes throughout the CNS and involved in many fundamental processes like synaptic transmission, neurovascular coupling, and blood-brain barrier maintenance.30 Additionally, astrocytes participate in CNS immune responses, when they take an activated phenotype having raised GFAP expression and release of proinflammatory cytokines which can enhance neurotoxicity and neurodegenerative disease progression.30-32 Hence, astrocytes have the basic requirement of energy homeostasis regulation in health and disease. MBH astrocytes modulate feeding behavior on pharmacological activation33,34 and show dynamic responses to circulating signals of nutrient availability like insulin and leptin.35-38 Also, MBH astrocytes become activated with obesity and HFD feeding in rodents and humans,3,23 which raises the possibility that astrocyte inflammation disrupts the hypothalamic regulation of energy balance and promotes DIO.

The group of Thaler et al developed a mouse model with an inducible astrocytic specific deletion of IKKβ with the use of tamoxifen. With this approach, they showed that decreasing astrocytic signaling protects mice from HFD induced HI and decreases susceptibility to DIO and glucose tolerance. The results highlight the importance of non-neuronal cells in obesity pathogenesis and suggest the possibility of newer target for therapy.59

**ROLE OF HAM-RS2**

Diets high in fibers may lower obesity risk and its morbidities.40,41 Zero point four percent decreases in body weight is reached by consumption of most dietary fibers for 4 weeks.42 The amount of weight loss was proportional to the physical and chemical properties (i.e., solubility, viscosity, and fermentability) of each type of fiber. Mechanisms by which weight loss is caused by fibers are changes in gut motility, prevention of absorption of nutrients and decrease in the total caloric input, which are associated with the physicochemical properties.51,43 Various fibers which can be fermented are under scrutiny, as the metabolites obtained after fermentation by bacteria in the gastrointestinal tract (GIT) may influence body weight. Short-chain fatty acids (SCFA) are produced from these fibers in the distal intestine which stimulates the release of GLP1 and peptide YY (PYY), which acts in synergy with leptin to induce satiety, which further regulates through the CNS.44-46 Although, SCFAs' are produced by fiber fermentation there is an inconsistent relation between GLP1 and PYY on satiety and food intake in humans. After eating a standard breakfast in the morning immediately following 3 days of consecutive intake of a barley kernel based bread with resistant starch, fasting plasma GLP1, and postprandial PYY concentrations were increased in healthy adults.47 But there was no change in appetite sensations like in satiety, hunger, desire to eat.47 Also, overweight women who had taken an enzyme hydrolyzed arabinofuran from flax at breakfast did not show a postprandial subjective satiety although there was an improvement of GLP1 and PYY, which corresponded to increased subjective satiety following maltodextrin intake in healthy humans.48 But there was no suppression of energy intake in spite of these changes. In a recent trial, there was improvement in PYY which corresponded with satiety along with 14% decrease in food intake in healthy adults who took 15 g unripe banana flour rich in resident starch for 6 weeks.49 Why there are differences in satiety peptides and satiety responses relates to fermentability patterns, nature, amount and duration of fiber intake along with gut microbe composition of individuals? Besides the blood, gut peptide concentrations may be very low to be able to cross the blood-brain barrier or the individual might be having a hypothalamic resistance which can occur from a HFD even in lieu of obesity.50

High amylose maize-resistant starch type 2 (HAM-RS2) comprises an insoluble, nonviscous fermentable fiber, which has been shown to improve glucose homeostasis,51 or those with metabolic syndrome.52-54 Yet many of trials of long duration did not report or show improvements in blood concentration of gut peptides, satiety responses or changes in food intake. The beneficial effects of HAM-RS2’s may be on glucose metabolism by increasing SCFA in blood to alter FFA and glycerol release from adipocytes and increased fat oxidation55 by affecting bile acid metabolism,56 or changing the gut microbiota profile.57 Earlier trials reported that the effects of HAM-RS2 on glucose homeostasis in healthy individuals or those with MS 15-16 Thus Maziaiz et al tried to determine the impact of daily consumption of 30 g HAM-RS2 incorporated into muffins for 6 weeks on glucose homeostasis in normoglycaemic healthy overweight adults at risk of developing glycemic abnormalities. They used a randomized control, placebo arm, a double-blind design with 18 overweight healthy adults consuming either muffins enriched with 30 g
HAM-RS2 (n=11) or 0 g HAM-RS2 (control; n=7) daily for 6 weeks. Both HAM-RS2 and control muffins were similar in total calories and available carbohydrates. They found at baseline total PYY concentrations were significantly higher 120’ following the consumption of study muffins in the HAM-RS2 group than control group (p=0.043). Within the HAM-RS2, the area under curve (AUC) glucose (p=0.028), AUC leptin (p=0.022), and postprandial 120’ leptin (p=0.028) decreased independently of changes in body composition or overall energy intake at the end of 6 weeks. Fasting total PYY increased (p=0.033) in the HAM-RS2 group but changes in insulin or total GLP 1 were not observed. Mean overall change in subjective satiety score did not correlate with mean AUC biomarker changes which suggested that the satiety peptides did not elicit a satiation response or change in overall total caloric intake. The metabolic response from HAM-RS2 occurred despite the habitual intake of a moderate to HFD (mean range 34.5% to 39.4%) of total calories. Thus, they concluded that consuming 30 g HAM-RS2/day x 6 weeks can improve glucose homeostasis, lower leptin concentrations and increase fasting PYY in healthy overweight adults without impacting body composition and may help in the prevention of chronic disease. However, between groups, differences in biomarkers were not seen and warrants future research before making specific recommendations.50

ROLE OF FGF21

It was believed that FGF21 was the hormone associated in response to nutrient restriction in lieu of increase during fasting, starvation and a ketogenic diet.59-62 But, recent data suggested that the decreased protein intake is the primary regulator of FGF21 during those interventions and FGF21 deficient mice failed to exhibit the increases in food intake and increases in energy expenditure and reductions in growth observed in wild-type mice consuming low protein diet (LPD).50 The mechanisms linking reduced protein intake to increased hepatic FGF21 expression and secretion are not clear but there is an implication of amino acid GCN2 sensor.63,64 GCN phosphorylates eIF2α in response to depletion of cellular aminoacids65 which lead to inhibition of general protein synthesis while increasing translation of transcription factors such as activation transcription factor 4 (ATF4). This links amino acid availability to metabolism specifically in the liver.66,67 The FGF21 promoter contains amino acid response elements (AARE) and both depletion of amino acid’s and activation of this Eif2α/ATF4 pathway increases FGF21.68,69-70 Thus, it was shown that hepatic Eif2α phosphorylation is induced by multiple settings of dietary protein restriction and that low protein (LP) induces increases in FGF21 and Eif2α phosphorylation are blunted in GCN2 deficient mice.53 Since FGF21 is required for metabolic and behavioral responses to protein restriction and GCN contributes to the increase of FGF21 in this setting, Laggar et al hypothesized that GCN2 deficient mice would fail to respond to reduced protein intake and thereby recapitulate this phenotype of FGF21 KO mice. They demonstrated that there is a persistent and essential role of FGF21 in the metabolic response to protein restriction. FGF21 KO mice were fully resistant to LP inducing changes in food intake, EE, body weight gain and metabolic gene expression for 6 months. GCN2KO mice recapitulate phenotype but LP induced effects on FI, EE, body weight begin to appear after 14 days on diet. They showed that this delayed emergence of LP induced metabolic effects in GCN2 KO mice coincides with a delayed but progressive increase of hepatic FGF21 concentration over time. Thus, they concluded that the data indicated that FGF21 is essential for the metabolic response to protein restriction but then GCN2 is only transiently required for LP induced FGF21.71

ROLE OF ASTROCYTES AND KETONE BODIES

The mechanisms by which HFD leads to obesity development are still poorly understood. Body weight regulation involves 2 mechanisms namely hunger and satiety. The brains actions regarding regulating these are influenced by nutrients, hormones, peptides and other related signaling molecules which cross the BBB and change the activity of particular metabolic sensing neurons which are dispersed in various anatomical sites in the brain. The mature human brain weighs only 2-3% of total body weight.72 Neurons store very little energy and hence are dependent upon the continuous exogenous supply of glucose as the primary metabolic substrate for most of brains energy requirements.73,74 Recently, importance of glia, of which mainly astrocytes have gained importance for giving this metabolic support to neurons.75,76 Since astrocytic foot processes directly impinge on brain microvessels they are the first cells which nutrients face on entering the brain.77,78 Astrocytes have important metabolic functions including neuronal transmission,79-82 glycogen storage and lactate production for neuronal metabolism during increased activity of neurons.83,84 In the ventromedial hypothalamus (VMH), ventromedial nuclei (VMN) and arcuate nuclei (ARC) astrocytes also produce ketone bodies from free fatty acids (FFA).85,86 Although, lactate production, occurs as a continuous process84,87 the ketone production from astrocytes occurs mainly if blood FA levels increase secondary to dietary intake.58,59 Recently, role of local production of ketones by VMH astrocytes as regulators of food intake during intake of HFD has been highlighted. Also, novel hypothesis by which astrocytes can regulate FA turnover in the VMH and mechanism by which astrocyte produced ketone override normal neuronal FA sensing to regulate feeding is described.

Le Foll et al tried to understand mechanisms of long-term and excessive HFD intake in obesity development. VMH is a major site involved in the regulation of glucose and energy homeostasis where metabolic sensing neurons integrate metabolic signals from the periphery. FA modulates, VMH neuronal activity through the use of FA translocator/CD36 which plays a critical role in the regulation of energy and glucose homeostasis. During LFD intake FA are oxidized by VMH astrocytes to fuel their ongoing metabolic needs. But HFD intake causes VMH astrocytes to use FA to generate ketone bodies. Thus they postulated that these astrocyte-derived ketone bodies are exported to neurons where they produce excess ATP and ROS, which over-
rides CD36 mediated FA sensing and acts as a signal to decrease short-term food intake. On a HFD, VMH astrocytes produced ketones reduced elevated calorie intake to LFD levels after 3 days in rats genetically predisposed to resist (DR) DIO but not leptin resistant DIO rats. This gives a suggestion that while VMH ketone production on a HFD can contribute to protection from obesity, the inherent leptin resistance overrides this inhibitory action of ketone bodies on food intake. Thus, astrocytes and neurons form a tight metabolic unit that is able to monitor circulating nutrients to alter FI and energy homeostasis.84

ROLE OF RESOLVINS

Infectious agents, trauma or chemical stimulus cause an acute inflammation and its prompt resolution is needed to prevent chronicity and undesired tissue damage which could lead to an unrestrained response to the original harmful stimulus.85 Lipoxins,86 resolvins,87,90 protectins88 are families of endogenously produced lipid-derived substances which act in the resolution phase of acute inflammatory processes.89 Resolvin D2 (RvD2) is one of the members of the resolvins family is produced from the ω-3-polyunsaturated fatty acid docosahexaenoic acid (DHA) as a result of a series of reactions which get catalyzed by lipoxigenases.91 The anti-inflammatory and pro-resolution effects of RvD2 are mediated at least in part by the pertussis sensitive G protein-coupled receptor (GPCR), GPR18, by signaling mechanism yet to be fully elucidated.95,96 Though most studies have explored the role of resolvins in acute inflammatory conditions, a recent study had provided evidence that both RvD2 and resolin D1 (RvD1) can modulate the chronic inflammatory process that takes place in the adipose tissue of obese subjects.97 Also, treatment with 17 hydroxy docosahexaenoic acid (17HDHA), a precursor of Rv D2, reduced inflammation and corrected IR in obese diabetic rodents.98 Obesity is one of the most prevalent diseases worldwide. Saturated fatty acids present in the diet induce an inflammatory response in the hypothalamus leads to dysfunctional regulation of caloric intake and energy expenditure,98-102 which plays an important role in the genesis and perpetuation of obesity.98 Number of pharmacological and genetic approaches used to decrease obesity linked H1 leads to reversal of the obesity phenotype in animal models9,103-105 increase the content of ω3 fatty acids can decrease obesity linked H1, increase POMC neuron-specific neurogenesis and attenuate the obese phenotype.106-107 As ω3 fatty acids are precursors of RvD2, Pascal et al examined the activity of this system in the hypothalamus of obese rodents. Male Swiss mice were fed either chow or a HFD. RvD2 receptor and synthetic enzymes were evaluated by real-time PCR and immunofluorescence. RvD2 was determined by mass spectrophotometry. Both dietary and pharmacological approaches were used to modulate the RvD2 system in the hypothalamus and metabolic consequences were determined. All enzymes involved in the synthesis of RvD2 were detected in the hypothalamus and were modulated in response to the consumption of dietary saturated fats leads to a reduction of hypothalamic RvD2. GPR18, the receptor for RvD2 which was detected in POMC and NPY neurons was also modulated by dietary fats. The substitution of saturated by polyunsaturated fats in the diet leads to increased hypothalamic RvD2 which was accompanied by decreased body mass and improved glucose tolerance. The ICV treatment with docosahexaenoic acid led to increased expression of the RvD2 synthetic enzymes increased expression of anti-inflammatory cytokines and improved metabolic phenotypes. ICV treatment with RvD2 caused decreased adiposity, improved glucose tolerance and increased hypothalamic expression of anti-inflammatory cytokines. Thus, they concluded RvD2 is produced in the hypothalamus and its receptor and synthetic enzymes are modulated by dietary fats. The improved metabolic outcomes of RvD2 make this substance an attractive approach to treat obesity.108

ROLE OF PUFA RECEPTORS GPR120 AND GPR40

Intake of large quantities of dietary fats is one of the most important environmental factors leads to obesity.108-110 Long chain saturated fatty acids trigger inflammation through the activation of toll-like receptor 4 and the induction of endoplasmic reticulum stress (ER).111-113 The low-intensity inflammation generated in this context can act both systemically and on selected anatomical regions to affect insulin and leptin action,114 insulin production,115,116 lipid metabolism117 and a number of other parameters related to whole body energy homeostasis. Since metabolic inflammation plays a part in the pathogenesis of insulin and leptin resistance, it has been hypothesized that means by which inflammation is attenuated could be beneficial for obesity, T2DM.118-120 Genetic and pharmacological approaches aimed at reducing inflammation have produced encouraging outcomes in various experimental models.116,119 Recently, in a clinical trial salsalate was used to target the inhibitor of kappa kinase (IKK) which leads to a marked decrease in glycated hemoglobin levels in patients with T2DM.121 Polyunsaturated fatty acid (PUFA) receptors GPR120 and GPR40 have been found to be attractive targets for the treatment of IR,122-125 Activation of GPR120 by PUFA or synthetic ligands engages an atypical signaling system which suppresses the metabolic inflammation in obesity and T2DM.126 A recent study has reported the beneficial effect of a synthetic agonist of GPR120 in improving glucose intolerance and hepatic steatosis in an animal model of DIO.127 Additionally, a lot of studies have shown the influence of targeting GPR40 systemically in T2DM.128 A new anti-inflammatory mechanism was described by Oh et al regarding the action of PUFA s through GPR120. On ligand binding, GPR120 recruits β arrestin-2 leads to internalization of the receptor/regulatory protein complex. The internalized β arrestin 2 binds to TAB1 and inhibits its binding to TAK1 which is a point of convergence for TNFα and TLR4 signal transduction and its inhibition impairs the progression of the signal towards JNK and IKK activation which leads to inhibition of inflammation.129 Beneficial effects of a small molecule which acts as a specific agent for GPR120 was also shown by Oh’s group.127 Obese mice treated with this molecule showed improved glucose tolerance and decreased hepatic steatosis...
which is accompanied by a decrease of metabolically inflammatory phenotype which shows GPR120 is an attractive potential target for treatment of obesity-associated metabolic disorders. A beneficial effect of GPR 40 activation has also been seen.125,129,130 GPR40 is expressed in pancreatic β cells on activation of PUFA’s it increases glucose-induced insulin secretion.125 Also, GPR40 is expressed in intestinal L&K cells, induces GLP1 and GIP secretion which provides another stimulus for insulin secretion.131 For mechanism of action of GPR120 it is shown that the induction of Ca2+ mobilization and activation of CREB may play important roles in some of the effects of this pathway.132 Hence, the potential therapeutic usefulness of agonists for GPR40 is considered important.128 Therefore, Dragano et al evaluated the expression and potential role of hypothalamic GPR120 and GPR 40 as targets for treatment of obesity. Male Swiss rats (6 weeks old), were fed with a high-fat diet (HFD, 60% of kcal from fat) for 4 weeks. This was followed by stereotactic surgery to place an indwelling cannula into the right lateral ventricle. Intracerebroventricular (ICV) cannulated mice were treated twice a day for 6 days with 2.0 µl saline or GPR40 and GPR120 agonists, GW9508, TUG 905 (2 µL,10 mM). Food intake and body mass were measured during the treatment period. At the end of the experiment, the hypothalamus was collected for real-time PCR analysis. It was shown that both receptors are expressed in the hypothalamus, GPR120 is primarily present in microglia while GPR 40 was expressed in neurons. Upon ICV treatment GW9508, a nonspecific agonist for both receptors, decreased energy efficiency and the expression of inflammatory genes in the hypothalamus. Reducing GPR120 hypothalamic expression using a lentivirus-based approach resulted in the loss of the anti-inflammatory effect of GW9508 and increased energy efficiency. ICV treatment with the GPR120 and GPR40 specific agonists TUG 1197 and TUG 905 respectively, resulted in milder effects than those produced by GW9508. Thus, it was concluded that GPR120 and GPR40 act in concert in the hypothalamus to reduce energy efficiency and regulate the inflammation associated with obesity. The combined activation of both receptors in the hypothalamus results in better metabolic outcomes than isolated activation of either receptor alone.133

ROLE OF ABSCICIC ACID

The thiazolidinediones (TZD) belong to a family of synthetic insulin sensitizers molecules; however, there are adverse effects of some of them. Hence, other compounds having similar properties but fewer side effects are needed. The phytohormone abscisic acid (ABA) was found in mammalian cells over 25 yrs back.134 Various studies have proposed it to be a universal signaling molecule.135,136 Structurally, ABA is very similar to TZD’s. ABA can improve glucose tolerance,137 decreases the levels of TNFα and reduce the adipocyte cell size in an in vivo model of obesity induced by HFD.138 Further in human and pancreatic cell lines (RIN m and INS2 cells), ABA can increase glucose-stimulated insulin secretion.139 This effect can be suppressed using pertussis toxin and PKA inhibitors.140 Dietary ABA further stimulates granulocyte function and macrophage infiltration in the adipose tissue.141 In mammalian cells the lanthionine synthetase C Like protein 2 (LANCL2) shows high homology with the ABA receptor in plants, the Arabidopsis GCP2. Blocking the expression of endogenous LANCL2 in granulocyte cells can stop ABA induction of Ca2+ response while overexpression of LANCL2 increases the ABA-mediated effects.142 Because of its role in the mediation of ABA effects, LANCL2 has been proposed as therapeutic targets for the treatment of inflammatory disease and DM.143

Also, ABA shows some molecular structural similarities to retinoic acid (RA). RA has useful effects in cognition, improving memory deficits in rodent models of Alzheimer’s disease. But both clinical and animal model data show an association between RA administration and the symptoms of depression.144 ABA given chronically has been shown to be a useful antidepressant as shown by increased sucrose intake, increased swimming in the forced swim test and decreased expression of CRH and RARα mRNA in the rat hypothalamus in control rats with no reported side effects.145 Thus Sanchez–Sarua et al tried to show if dietary ABA could improve cognitive defects resulting from a HFD induced neuroinflammation. HFD increases the levels of neuroinflammation markers in the brain146 and may be a link between obesity and degenerative disorders via IR.147 Also HFD has been shown to cause memory loss by increasing inflammatory markers in the hippocampus.148 Male Wistar rats were fed with standard diet or HFD, with or without ABA (20 ng/ml) in drinking water for 12 weeks. After 11 weeks of treatment they compared the behavior of 4 groups using 2 memory paradigms; the novel object recognition (NOR) and the T maze. Also, they measured ABA levels in the blood and cerebellum of all 4 groups using HPLC. The microglia proliferation using IHC was also analyzed. It was demonstrated that ABA administered in drinking water improved glucose tolerance and cognitive performance and decreased the levels of inflammatory markers in the hypothalamic areas. Their results confirmed a therapeutic potential of this phytohormone in the peripheral metabolic alterations. The data also strongly suggest that the potential beneficial effects of ABA in disorders of neuroinflammatory etiology, not shown earlier.149

HEPATIC CLOCK AND OBESITY

To adapt to the physiology of 24 hour rhythm of day and night most species have evolved endogenous circadian clocks in response to piles of earth rotation around its axis.150,151 These clocks are based on transcriptional-translational feedback loops built from a set of clock gene proteins which includes the 2 transcriptional factors circadian locomotor output cycles kaput (CLOCK) and brain and muscle ARNT like 1 (BMAL1 or ARNTL) which in synergy drive rhythmic expressions of three period and two cryptochrome genes through binding to E box enhancer motif BMAL 1 and CLOCK regulate other E box controlled genes in a tissue-specific manner; thus, translating the circadian clock
rhythm into physiologically meaningful signals.152,153 All body tissues have clocks which are synchronized by a master pacemaker located in the hypothalamic SCN154,155 which is controlled by the external light rhythm. Peripheral clocks and SCN controlled sleep-wake and food intake rhythms regulate the expression of many metabolically relevant genes.156 Peripheral clocks not only respond to SCN signaling but also get reset by the timing of food intake. Because of mistimed feeding rhythms which occur commonly in modern industrialized societies can promote internal clock desynchrony and the development of metabolic disorders.157-162 In liver transcriptomic analyses have shown more than 3000 rhythm transcripts163 and chromatin immunoprecipitation/DNA sequencing experiments showed more than 2000 DNA binding sites for BMAL1 in the murine liver.164 Circadian regulation has been shown for several metabolic processes like xenobiotic detoxification,165,166 mitochondrial function and lipid and glucose metabolism.163,167,168 Mice with hepatocyte-specific abrogation of clock function through deletion of BMAL1 display impaired glucose homeostasis but normal body weight regulation.162 Relatively, mice carrying a dominant negative mutation in the gene encoding the BMAL partner CLOCK ∆19 partially restored liver clock function (MT-LR) showed normal mitochondrial function and lipid and glucose metabolism.163,167,168 Mice with hepatocyte-specific abrogation of clock function through deletion of BMAL1 display impaired glucose homeostasis but normal body weight regulation.162 Relatively, mice carrying a dominant negative mutation in the gene encoding the BMAL partner CLOCK ∆19 are overweight and under HFD conditions develop symptoms of the metabolic syndrome.166 This obesogenic phenotype is associated with dysregulated feeding rhythms and overeating during the normal rest phase. Also mistimed feeding is associated with the development of obesity in mice and humans.170-172 In wild-type mice restricting access to an HFD to the night-time improves clock gene rhythms with normalized weight regulation.170,173 Appetite control and energy expenditure control are centrally controlled and therefore become difficult targets for clinical intervention.174-176 Metabolic feedback signals from the periphery like leptin and ghrelin, liver-derived factors such as FGF21,177 and ketogenic bodies,178 reach the brain and modulate neuronal circuits to adjust energy metabolism.179 This bottom-up communication from peripheral metabolic tissue to control regulatory circuits is impaired during obesity.114,180 Thus Meyer–Kovac investigated metabolic parameters of wild-type (WT) and CLOCK∆19 mutant mice (MT) under ad libitum and night time-restricted HFD feeding. Liver circadian clock function was partially rescued by hydrodynamic tail vein delivery of WT clock DNA vectors in mutant mice and transcriptional, metabolic, endocrine and behavioral rhythms studied. They found night time-restricted feeding restored food intake but not body weight regulation in MT mice under HFD, suggesting clock dependent metabolic dysregulation downstream of circadian appetite control. Liver directed clock gene therapy partially restored liver circadian oscillation function and transcriptional regulation without affecting centrally controlled circadian behavior. Under HFD, MT mice with partially restored liver clock function (MT-LR) showed normalized body weight gain, rescued 24 h food intake rhythms and WT like energy expenditure. This was associated with decreased night time leptin and daytime ghrelin levels, decreased hepatic lipid accumulation and improved glucose tolerance. Transcriptomic analyses revealed the hepatic clock rescue in MT mice affecting a range of metabolic pathways. Thus they concluded that liver clock gene therapy improves resistance against HFD induced metabolic impairments in mice with circadian clock disruption. Restoring or stabilizing liver clock function might be a promising target for therapeutic interventions in obesity and metabolic disorders.181

ROLE OF KBH1

KBH1 is a novel herbal mixture which consists of Chinese lizards (Saururus chinensis), turmeric (Curcuma longa L.) and Chinese sonega (Polygona tetumfolia). Traditionally, in Korea and other countries like China, these herbal medicines have been used for different anti-inflammatory effects, antioxidative effects, neuroprotective effects and the prevention of hypercholesterolemia.182-188 Saururus chinensis has been used in folk medicine for the treatment of various inflammatory diseases, gonorrhea, and edema in Korea180 and exhibits antiasthmatic and anti-inflammatory activities.190 Also, previous biological studies of S. chinensis have established its effects in metabolic diseases including hyperlipidemia, hyperglycemia, neuroprotective and hepatoprotective effects.191-193 Curcuma longa has been used in traditional medicine in China, Korea, India for ages as the main ingredient in prescriptions like Xia oyaoo-san for mental disorders.194 Also it has been used for the treatment of blood stasis in traditional Korean medicine.195 C. longa is the main ingredient in Gambigyeong sinhwah, which exerts its antiobesity effects through lipid accumulation and adipose PPARα activation198 and prevents high-fat diet-induced hyperlipidemia as the main ingredient in Artemesia Jacquemontii.199 Polygona tetumfolia has been used as a traditional Chinese medicine for the treatment of anxiety and dementia200,201 along with the preventive effect of this on behavioral disorders and inflammatory diseases.202,203 Lee et al studied the synergistic effect of the herbal mixture of S. chinensis, C. longa and P. tetumfolia (KBH1) in obesity and its possible molecular mechanism in obesity-induced hepatic steatosis and leptin resistance in the hypothalamus. They used Hep G2 cells, primary neuronal cells and an HFD induced obesity rat model to determine the effect of KBH1 in vitro and in vivo on hepatic steatosis and leptin resistance accompanied by obesity. To identify the alleviation effect on lipid accumulation, Hep G2 cells stimulated by FFA were stained with Oilred O, in addition, immunoblotting and qPCR were done to determine the effects of KBH1 on the activation of proteins and nuclear enzymes in Hep G2 cells and the steatotic liver of HFD induced obesity rats. For studying the effect of KBH1 on leptin resistance of hypothalamus and its possible molecular mechanisms they examined the effects of KBH1 on the activation of the leptin resistance related protein in primary cultured cortical neuron cells and the hypothalamus of an HFD induced obesity rat model. In addition, they used HPLC analysis to identify the standard compound of KBH1. KBH1 besides suppressing lipid deposition in Hep G2 cells exposed to FFA, downregulated major factors in lipogenesis and upregulated major factors in lipolysis. Similarly, in an HFD induced obesity model, KBH1 improved hepatic steatosis by alleviating the effects on lipogenic genes and kinases. Additionally, KBH1 markedly improved the leptin-mediated signals impaired by obesity or FFA in the obesity model and primary cultured cortical neuron cells. In addition, KBH1 was analyzed to include 6 standard compounds using HPLG analysis, among
these compounds, onjisaponin B, and curcumin potently suppressed the levels of triglycerides. Thus, they concluded that KBH1 inhibits alleviating effects by improving hepatic steatosis and leptin resistance by upregulating the activation of AMPK and suppressing the expression of PPARγ. These findings show the potential of KBH1 as a functional food supplement or preventive agent in the treatment of obesity.201

ROLE OF AUTOPHAGY DYSFUNCTION IN OBESITY

In animals, chronic intake of HFD causes DIO which leads to insulin and leptin resistance in hypothalamic neurons.114,205,206 Increased inflammation in the hypothalamus was identified to mediate the development of obesity and the pathways which include IKKB/NFκB pathway101,207 and upstream inputs such as MYD88,208,209 endoplasmic reticulum stress101,207,208 and JNK signaling.102,210-212 Chronic inflammatory stimuli can also lead to neuronal apoptosis which is important for the anorexigenic response.213-214 Recently, neuroimaging studies revealed that dysfunctional and neuronal loss were associated with obesity in the hypothalamus of humans and rodents.215-217 Besides having effects on food intake and energy expenditure HI seems to impair systemic glucose metabolism. Genetic and pharmacological modulation of the ER stress and inflammatory pathways in the hypothalamus affected liver gluconeogenesis.208,218-219 Inflammatory inhibition of TLR4 or TNFα signaling in the hypothalamus impaired improved insulin signal transduction in the liver and reduced hepatic glucose production.208 These studies suggest that HI plays a role in weight gain and systemic dysfunction of glycemic control. Meng and Cai have shown that neuronal autophagy is compromised under conditions of chronic excess fatty acids in the diet. In chow feeding mice, the site-specific inhibition of ATG7 in the MBH lead to autophagy inhibition, impairment of hypothalamic control of energy balance, obesity and HI through IκB activation. In HFD, these metabolic changes got increased along with the progression of insulin and leptin resistance.209 Normally autophagy is a homeostatic process that occurs in all eukaryotic cells and is needed for degrading damaged proteins as well as organelles. It also sequesters the cytoplasmic components in the double membrane vesicles known as autophagosomes.220 These autophagosomes thus fuse with lysosomes where the damaged proteins and organelles are degraded by lysosomal proteases and recycled.221-223 If this autophagy is impaired, it may cause inflammation suggesting that autophagy helps in inhibition of inflammatory response220,224,225 Portovedo hypothesized that obesity may lead to impairment in hypothalamic autophagy in mice. They examined the hypothalamic distribution and content of autophagic proteins in animals with obesity induced by 8 or 16 weeks HFD to induced obesity and in response to ICV injection of palmitic acid. They showed that chronic exposure to an HFD leads to an increased expression of inflammatory markers and downregulation of autophagic proteins. In obese mice autophagic induction leads to the downregulation of proteins like JNK and Bax which are involved in stress pathways. In neuron cell lines palmitate has a direct effect on autophagy even without inflammation activity. Thus, understanding the cellular and molecular basis of autophagy is important in finding new diagnostic and therapeutic targets for obesity.226

CONCLUSIONS

In this review, we have discussed how HI and glories precede the development of obesity and gradually is associated with loss of anorexigenic POMC neurons tilting the balance towards higher orexigenic AgRP:POMC neuron ratio. During HFD feeding these neurons may be injured by an unknown mechanism and that this injury triggers activation of local glial cell populations (astrocyte and microglia). The neuron injury and reactive glories can, in turn, impair homeostatic control of body fat stores leading to increased body weight. Further, we discussed the role of HAM-RS2, abscisic acid, KBH1, polyunsaturated fatty acid receptors GPR120 and GPR40 as potential targets for therapeutic interventions in preventing this HI and obesity and future targets for obesity treatment.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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