Results: As expected, we show that serum-starvation increases autophagy in human SH-SY5Y neuroblastoma cell line as evidenced by increased expression of LC31/II and ATG7 and downregulation of p62 considered as key autophagy markers. Interestingly, we report that resistin inhibits starvation-induced neuronal autophagy, and this effect is exacerbated in the presence of NH4Cl, an inhibitor of autosomal degradation. Using siTLR4 approach, we also demonstrate that resistin impairs autophagy through TLR4. Furthermore, we decipher the signaling pathways involved in resistin action and demonstrate that resistin inhibits AMPK phosphorylation and increases Akt/mTOR phosphorylation contrasting with autophagy impact on these signalling pathways. Finally, we validated the impact of resistin in mice and showed, in the hypothalamus, that the inhibitory effect of resistin towards autophagy markers is completely abolished in TLR4 knockout mice.
Conclusion: Altogether, these findings reveal resisitn/TLR4 as a new regulatory pathway of neuronal autophagy and contribute to the understanding of the underlying mechanisms involved in the impairment of neuronal autophagy.
Disclosure: **Y. Benomar:** None.

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Obesity is associated with a more inflammatory phenotype of macrophages in human pancreatic islets

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Background and aims: Obesity is a major risk factor for the development of type 2 diabetes (T2D). A chronic, low-grade, sterile inflammation is present in obesity; tissue macrophages contribute to such inflammation, leading to persistent insulin resistance and β-cell failure. Changes in macrophage polarization are tightly associated to macrophage function and are involved in many diseases. In this current research, we aimed to characterize potential obesity-related changes in macrophage polarization markers in human pancreatic islets.
Materials and methods: To establish the relationship between obesity and islet macrophage markers, expression of macrophage polarization markers *ITGAX (CD11c)*, *NOS2 (iNOS)* and *IL1b* as M1 macrophage-associated genes, *CD163*, *IL10* and *TGFb* as M2 macrophage-associated genes and *CD68* as general macrophage marker was analyzed by qRT-PCR in isolated human pancreatic islets from obese (BMI >30) and non-obese (BMI <30) donors as well as in human islets following depletion of islet resident macrophages by treatment of clodronate liposomes.
Results: Islets from obese donors expressed significantly more *NOS2* (2.7-fold) than from non-obese donors, while general macrophage marker *CD68* and M2 macrophage markers *CD163* and *IL10* were not significantly changed between obese and non-obese donors. After depletion of islet resident macrophages by clodronate treatment, expression of *ITGAX*, *IL1b*, *IL10* and *CD163* was mostly deprived in human islets, indicating macrophage-dependency of these genes. In contrast, *TGFb* expression was unchanged, suggesting a macrophage-independent source in human islets. *NOS2*expression was not affected by clodronate treatment in islets from non-obese donors, but reduced in islets from obese donors, which suggests an obesity-induced iNOS expression in macrophages of human islets.
Conclusion: Our results indicate that CD11c, IL-1β, IL-10 and CD163 in human islets are associated with islet macrophages. The increased islet macrophage *NOS2* expression in obese individuals suggests that obesity is associated with a switch to an M1-like inflammatory phenotype in islet macrophages, which may further contribute to obesity-induced islet inflammation and finally β-cell failure.
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Human obesity alters circadian clock function through NF-κB activation

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Background and aims: Nutritional environment affects the transcription-translation feedback oscillators of the circadian clock that maintain metabolic homeostasis. These loops are composed of activators (BMAL1 and CLOCK) that induce the transcription of repressors, the most important being PERIOD 2 (PER2). PER2 inhibits the forward limb and this process generates a rhythm of ~24 hr. Experimental genetic models have provided evidence that many complications of obesity are mediated through metabolic inflammation induced by activation of the transcription factor NF-κB. Here, we investigated the link between clock disruption and NF-κB mediated inflammation in human obesity.
Materials and methods: Human omental adipose tissue was obtained from 5 obese and 5 non-obese age-matched patients (body mass index: ~50 *vs.* 25 kg/m^2) undergoing abdominal surgery. Using fluorescence-activated cell sorting analysis, omental preadipocytes obtained from the stromal vascular fractions were identified as PDGFRα$^+$, CD31$^-$, CD45$^-$ cells. Preadipocytes were infected with *Per2-dLuciferase*-expressing lentivirus (a rapidly-degradable form of luciferase driven by the *Per2* gene promoter), synchronized and bioluminescence was recorded continuously for 5–7 days. Binding of the p65 subunit of NF-κB and BMAL1 to *PER2* promoter was measured by chromatin immunoprecipitation (ChIP). ChIP of RNA polymerase II was also performed to explore the rhythms in nascent transcription. Additionally, ChIP were conducted on *CCL2*, a chemokine involved in the pathogenesis of type 2 diabetes. In some experiments, NF-κB inhibition was achieved either by pharmacological or gene silencing approaches, restoring NF-κB activity of obese subjects to a non-obese state.
Results: By measuring *Per2-dLuciferase* bioluminescence, we described endogenous cell-autonomous human adipose tissue oscillators, independently of food intake and nutrient signals. We found a period lengthening of *Per2-dLuciferase* oscillations in obese preadipocytes (23.33 ± 0.11 hr *vs.* 22.53 ± 0.27 hr, $p < 0.05$), reflecting an altered function of the molecular clock. ChIP assays revealed that p65 binding to *PER2* is enhanced by almost 10 times in obese preadipocytes ($p < 0.05$) while binding of BMAL1 and RNA polymerase II are significantly decreased ($p < 0.05$) consistently with the reduced *PER2* expression observed. Increased NF-κB activity was also associated with altered BMAL1 occupancy on *CCL2* promoter and a ~10 fold increased recruitment of RNA polymerase II ($p < 0.05$), consistently with increased CCL2 expression and secretion. NF-κB inhibition in obese preadipocytes restored BMAL1 binding and clock function to a non-obese level.
Conclusion: Collectively, our findings demonstrate that obesity alters clock function through NF-κB signaling in human omental adipose tissue. Our results suggest that the disruption of these oscillators in obesity leads to abnormal chemokine production and may contribute to metabolic disorders.
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Complement C3 and C4, but not their regulators or activated products, are associated with metabolic syndrome: the CODAM study

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