Obesity-induced Lymph Node Dysregulation - A TEM Analysis

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INTRODUCTION

Obesity is widespread and implicated in the development of metabolic diseases such as type II diabetes, CVD, and cancer. Excess adiposity has been demonstrated to induce a chronic low grade inflammatory state, which is linked to disease susceptibility. Previous studies demonstrate adipose tissue in mice consuming a high fat diet (HFD) have higher rates of macrophage (MO) infiltration. Resulting pro-inflammation occurs not only locally but also in the lymph nodes (LN) embedded in these fat deposits[1]. These areas, central to immune cell priming and maintaining homeostasis, link the innate and adaptive immune responses[2]. In our earlier investigation we hypothesized MO infiltration resulted in increased fibrosis in LNs and was associated with increased release of inflammatory cytokines IL-1β, CXCL8, and TNF by activated MOs [1]. These molecules are associated with increased fibrosis driving microarchitectural changes and subsequent dysfunction in several tissue types[3].

HYPOTHESIS

We hypothesized that the disruption of the LN conduit system by fibrosis would affect immune cell populations, activity, and the vital cell to cell cross talk that drives the immune response.

METHODS

Subjects: Male C57 BL6 mice were fed standard rodent chow control (18% fat 33% protein 49% carbohydrate) or high fat diet (HFD) (western) (21% milk fat 34%sucrose, 45% kcal from fat, Harlan Teklad, Madison, WI) .
Termination: Occurred at 8 and >13 weeks. Visceral and subcutaneous lymph nodes and fat were collected.
Outcome Measures: Changes in lymph node architecture were examine with H& E and transmission electron microscopy. Cell numbers were quantified by flow cytometry.

RESULTS

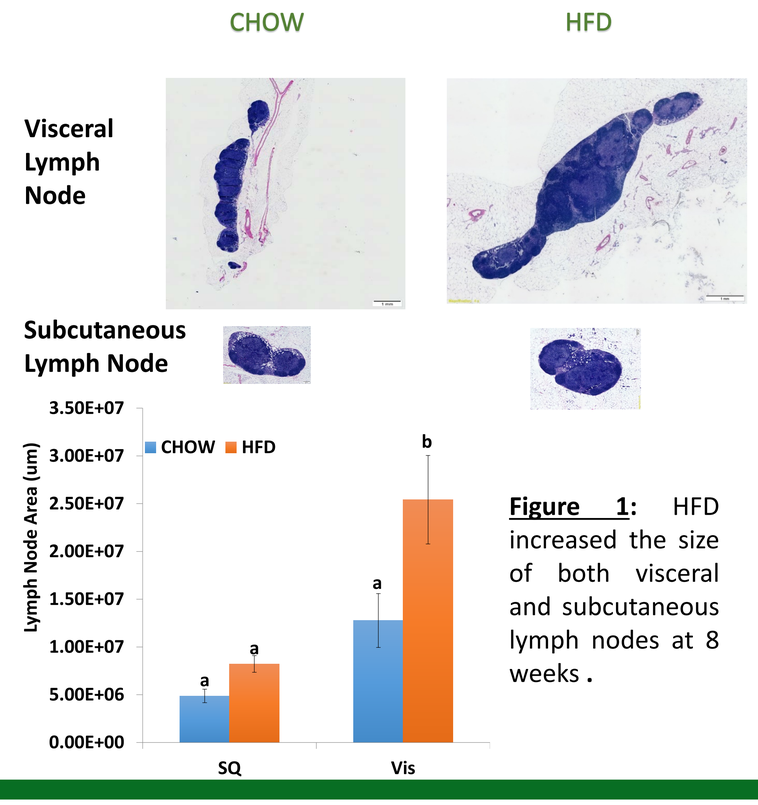


Figure 1: HFD increased the size of both visceral and subcutaneous lymph nodes at 8 weeks .

RESULTS

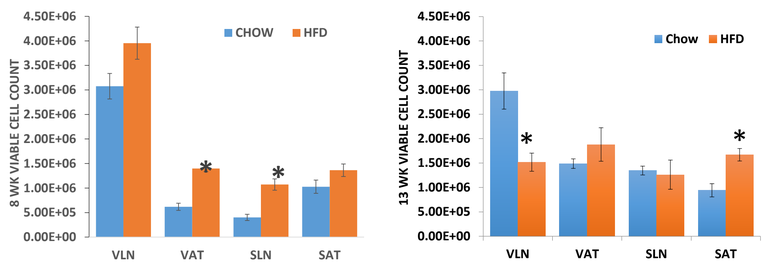


Figure 2 – **Immune cell number A.) 8 weeks –**HFD increased viable cells in VLN, visceral adipose tissue and SLN. **B.) 13 weeks –** HFD significantly decreased viable cells in VLN, but increased cell number in subcutaneous adipose tissue. (*p≤ 0.05, compared with respective control).

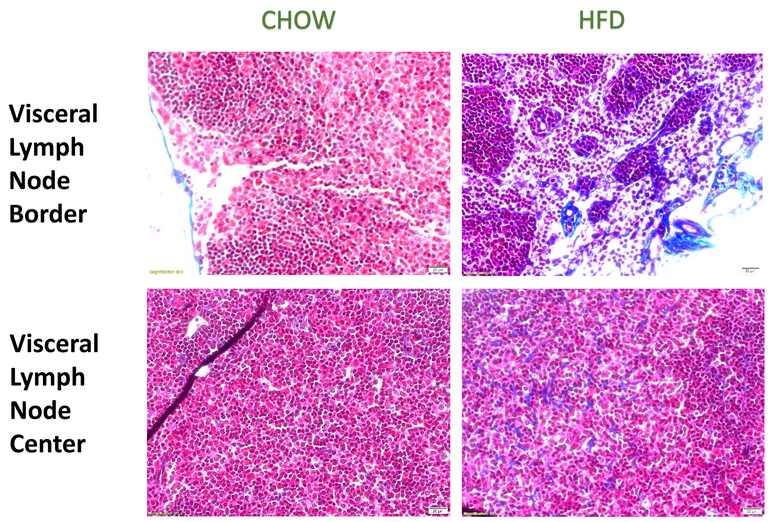
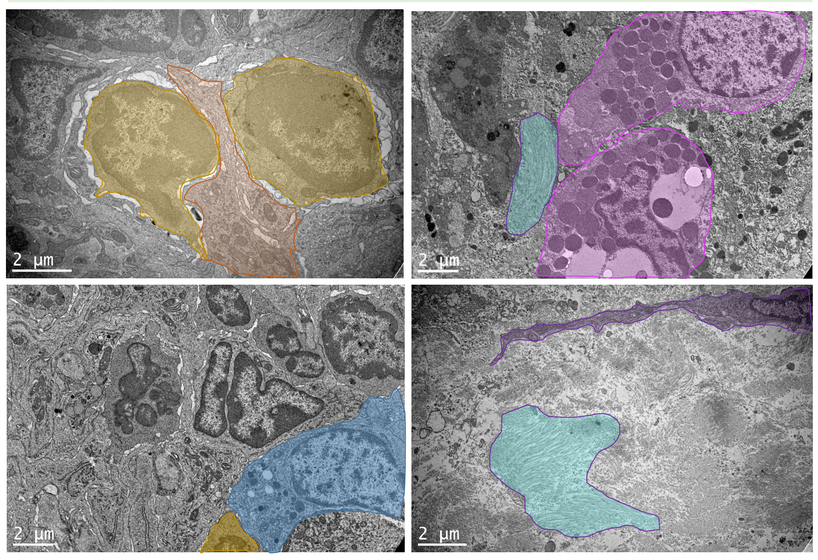


Figure 3: 13 weeks of HFD induced significant fibrosis in visceral lymph node. Fibrosis first appear in subcapsular space subsequently spreading to the paracortical T cell zone and medulla. (*p≤ 0.05, compared with respective control).

RESULTS

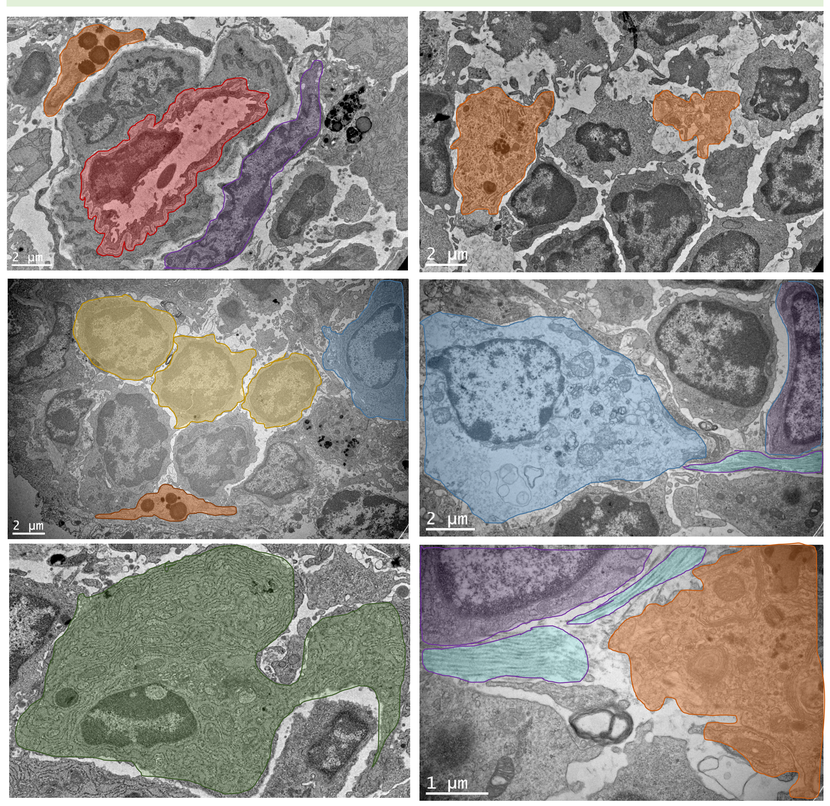
Figure 4- Subcutaneous Lymph Node



Figures 4 & 5 Key:

DC, Plasma cell, Collagen, T cell, MO, FRC, Mast cell, HEV

Figure 5 - Visceral Lymph Node



EXPERIMENTAL RESULTS

HFD LN microarchitecture was distinctly different from CHOW. CHOW SQLNs exhibited a pattern of closely packed lymphocytes interacting with APCs, including activated dendritic cells (DC), and MOs engaging in pseudopodia as evidenced by irregular MO shape. Collagen deposition in CHOW SQLNs was minimal. HFD SQLNs displayed active fibroblast reticular cells (FRCs) leaving large amounts of collagen creating physical barriers, which we postulate prevent lymphocyte and APC interaction. In addition, mast cells containing histamine granules were apparent in HFD SQLNs only. CHOW VLNs showed active phagocytic DCs and MOs interacting with lymphocytes. Plasma B cells were present, implicating a functioning conduit system. FRCs were inactive and collagen deposition is minimal, including surrounding area of the high endothelial venule (HEV). The HFD VLNs contained immature DCs, immobile MOs and highly active FRCs.

CONCLUSIONS

Based on our findings mechanisms by which fibrosis interferes with cellular communication in the LNs is elucidated. Further exploration into the implications of fibrosis on HEVs and cell trafficking may enable us to comprehend the roles obesity and inflammation play in compromising conduit system integrity. Taken together this data can help support the development of preventative measures to reduce susceptibility to immune dysregulation associated with obesity.

REFERENCES

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