Editorial: "Crowning" eosinophils in adipose tissue: does location matter?

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RECEIVED APRIL 29, 2015; REVISED JUNE 22, 2015; ACCEPTED JUNE 23, 2015. DOI: 10.1189/jlb.3CE0415-178RR

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Introduction

It is well known that the incidence of obesity is increasing at an alarming rate, leading to increased prevalence of insulin resistance and metabolic disease. Excess visceral adiposity is a major risk factor for development of obesity-associated disease and directly correlates with elevated levels of inflammatory markers, including TNF-α, IL-6, and IL-1β. In addition to its role in energy storage, adipose tissue is becoming increasingly accepted as an immunologically active organ [1]. In 2003, seminal studies identified increased macrophage accumulation in obese adipose tissue as the primary source of increased TNF-α in mice fed a high-fat diet [2]. Since these initial observations, many significant findings have increased our understanding of the immunologic changes in adipose tissue during obesity [3]. Perhaps the most remarkable change is the switch from an anti-inflammatory environment, characterized by the presence of alternatively activated macrophages and regulatory T cells, toward a proinflammatory environment, rich in classically activated macrophages, cytotoxic CD8 T cells, and Th1 CD4 T cells. Importantly, many of the leukocyte changes that have been described in mouse models have been confirmed in human adipose tissue.

More recently, the surprising observation was made that eosinophils serve a protective role maintaining adipose tissue homeostasis during obesity [4]. Eosinophils were found to be the major cellular source of IL-4, which promotes macrophage-alternative activation. Systemic increases in eosinophils protected mice from high-fat diet-induced insulin resistance. Subsequent studies identified ILC2 cells as the primary source of IL-5 in adipose tissue, responsible for the increase in eosinophils [5]. Although the regulation of immune cells in adipose tissue is not entirely understood, the intersection of immune function and metabolic disease has clear, clinical relevance for human health.

In this issue of *JLB*, Bolus et al. [6] used a *Ccr2*$^{-/-}$ mouse model on a high-fat diet to study immune cell regulation in obese adipose tissue. CCR2 is a chemokine receptor known to regulate macrophage migration to sites of inflammation. Ccr2$^{-/-}$ mice were previously shown to have decreased macrophage accumulation in adipose tissue during high-fat diet-induced obesity, correlating with improved insulin and glucose homeostasis [7]. Previous work by the Hasty lab [8] reported the accumulation of a novel but unidentified myeloid cell in the adipose tissue of obese Ccr2$^{-/-}$ mice. In the current study, this cell population is confirmed to be eosinophils, validated by nuclear morphology and Siglec-F expression, with the latter being an inhibitory receptor that marks murine eosinophils. Interestingly, the increased number of eosinophils in Ccr2$^{-/-}$ mice was limited to the peritoneal cavity and adipose tissue but was not observed systemically in the bone marrow, blood, spleen, or liver. Furthering the mechanistic understanding of how eosinophils might help maintain adipose tissue homeostasis during obesity, Bolus et al. [6] created 3-dimensional renderings of immunofluorescent confocal microscopy images, which revealed that eosinophils were recruited specifically to CLS in the obese adipose tissue of Ccr2$^{-/-}$ but not wild-type mice. This raises an intriguing question: does tissue location matter for eosinophil function? Macrophages that localize near CLS are thought to induce increased inflammatory activity compared with macrophages in interstitial spaces. Given the current data by Bolus et al. [6], it seems feasible that a similar, but perhaps opposite, paradigm also exists for eosinophils (**Fig. 1**). It is noteworthy that whether the location of immune cells in adipose tissue matters is not well understood. It remains to be determined whether tissue location determines the function of the cell or vice versa.

It is not clear whether the increase in eosinophils in obese Ccr2$^{-/-}$ mice was mediated by increased recruitment during obesity or whether local proliferation was the causative factor. In addition to IL-5 discussed above, eotaxins have been reported to increase in the serum and adipose tissue of obese individuals, implicating them in the regulation of eosinophil recruitment to obese adipose tissue. Bolus et al. [6] measured increased Ccl11 and Ccl3 in obese Ccr2$^{-/-}$ adipose tissue but reduced Ccl24 expression. Therefore, it is likely that IL-5 and eotaxins regulate eosinophil recruitment to obese adipose tissue, but the precise mechanism remains to be fully delineated (Fig. 1). The increase in eosinophil recruitment to CLS in Ccr2$^{-/-}$ mice suggests that CCR2-positive cells somehow actively inhibit this process in wild-type obese mice. Whether this regulation is via cytokines or direct cell-cell contact is not known. In support of cytokine-mediated regulation, eosinophil secretion of IL-4 has previously been shown to regulate macrophage alternative activation [4]. Furthermore, the role of ILC2 cells, which are known to regulate eosinophils in adipose tissue, as discussed above, was not tested in the obese Ccr2$^{-/-}$ mice. It is noteworthy that eosinophils and

Abbreviations: CLS = crown-like structures, ILC2 = innate lymphoid type 2

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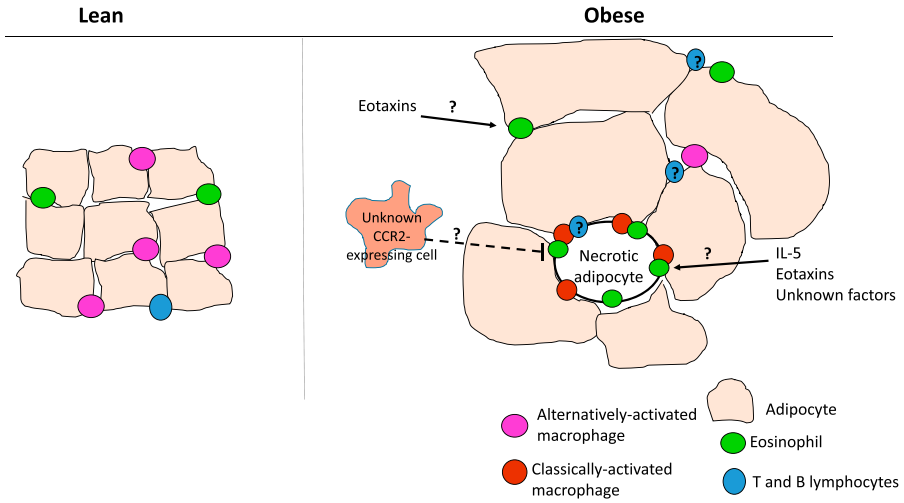

Figure 1. Regulation of eosinophils in obese adipose tissue. Increased eosinophils in adipose tissue protects against metabolic dysregulation in diet-induced obese mice. The regulation of eosinophils in obese adipose tissue is mediated by several factors, including IL-5, eotaxins, and a population of CCR2-expressing cells, although the precise regulatory mechanisms is not clear. Whether the location of eosinophils is important for their function and how this is regulated is unknown.

ILC2 cells were recently shown to be protective against obesity and even promote beiging of white adipose tissue [9]. Adipocyte beiging is a phenomenon in which white adipose tissue takes on characteristics of brown adipose tissue with increased mitochondrial uncoupling. This switch results in increased thermogenic energy expenditure and weight loss and has become a major area of interest in adipocyte biology. Interestingly, IL-33, a well-known eosinophil activator, increases adipose tissue beiging and energy expenditure and improves metabolic homeostasis in mice [9], further suggesting an important role of eosinophils in metabolism. To our knowledge, no studies have carefully tested whether increasing eosinophils in human patients confers similar protection against obesity-related disease. However, some studies suggest positive correlation of eosinophil counts with increased markers of diabetic nephropathy in patients with type 2 diabetes [10], whereas other studies report an inverse relationship with reduced eosinophils in type 2 diabetes [11]. Of note, whether allergic responses in humans impact adipose tissue biology or improved insulin action also remains unclear. Given drugs that target eosinophil-associated disorders are being tested, it may be important to study the impact of these therapies on a subgroup of patients with high body mass index and adiposity. Clinically, whether manipulation of eosinophils will impact obesity-associated comorbidities deserves careful attention.

In summary, the study by Bolus et al. [6] confirms the protective role of eosinophils in obese adipose tissue and implicates a new role of CCR2 as one of the mechanisms controlling eosinophil–macrophage homeostasis in CLS of adipose tissue. This study also raises an interesting and challenging question in immunometabolism that remains to be addressed for most immune cells in adipose tissue: does leukocyte function differ based on its specific location?

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KEY WORDS:
obesity · inflammation · crown-like structures · chemokines