Nelson H. Knudsen and Chih-Hao Lee

IL-21 and IRF4: A Complex Partnership in Immune and Metabolic Regulation

Diabetes 2014;63:1838–1840 | DOI: 10.2337/db14-0273

COMMENTARY

Adipose tissue is comprised of a community of different immune cells that contributes to regulation of energy storage and release by adipocytes. The population of various immune cells is dynamic, undergoing phenotypic and compositional changes in response to physiologic (e.g., fasting vs. feeding) and pathologic (e.g., lean vs. obese) stimuli. Cumulative evidence suggests that a regulatory or anti-inflammatory immune phenotype promotes metabolic homeostasis, whereas a proinflammatory response is associated with metabolic dysregulation (1). Accordingly, lean, healthy white adipose tissue (WAT) is home to a population of alternatively activated macrophages (M2s) as well as immune cells that mediate M2 polarization, such as eosinophils, $CD4^+$ T-helper type (Th) 2 cells, and regulatory T cells (Tregs) (1). During obesity, WAT is infiltrated by additional classically activated macrophages (M1s), neutrophils, mast cells, and $CD8^+$ T cells that release proinflammatory cytokines, thereby sustaining metabolic inflammation, or meta-inflammation (2). Although the changes in immune repertoires associated with different metabolic states are well characterized, mechanisms underlying the switches in immune phenotypes remain unclear.

Due to their roles in skewing immune cell responses, multiple cytokines have been suggested to play a role in the development of meta-inflammation in obesity. The first studies to recognize that obesity is associated with inflammation identified tumor necrosis factor-α (TNFα) as a mediator of WAT insulin resistance. TNFα produced by infiltrating M1s triggers the activation of Jun NH_2-terminal kinase and inhibitor of κB kinase β causing antagonistic phosphorylation of insulin receptor substrates (3). Other proinflammatory cytokines produced upon inflammasome activation, namely interleukin (IL)-1β and IL-18, induce adipose inflammation and suppress insulin response (4). In contrast, Th2 cytokines, including IL-4 and IL-13 released primarily by resident eosinophils and innate lymphoid cells, activate M2 polarization and maintain WAT homeostasis (5). The anti-inflammatory cytokine IL-10 has also been shown to enhance adipose insulin sensitivity (6). Both M2s and Tregs produce IL-10. The discovery of a unique population of adipose tissue resident Tregs that are enriched in IL-10 has spurred interest into their physiologic function in metabolic regulation (7,8). Notably, obesity greatly suppresses the number of Tregs in WAT.

In this issue, Fabrizi et al. (9) demonstrate that mice lacking Il-21 are protected against high-fat diet–induced metabolic dysfunction. Il-21 is produced by Th17 cells, which in turn promotes expansion of Th17 cells and inhibits induction of Tregs (10). Mice lacking Il-21 (Il-21 KO) are resistant to high-fat diet–induced weight gain and display improved glucose and insulin tolerance. Not surprisingly, the Treg and M2 populations are increased and meta-inflammation is suppressed in WAT of Il-21 KO mice.

Several cell types are capable of transducing Il-21 signaling through expression of Il-21 receptor (*Il-21R*). Fabrizi et al. not only show that immune cells within WAT, but also adipocytes express *Il-21R*. High-fat diets increase mRNA levels of *Il-21* by immune cells and *Il-21R* by adipocytes, suggesting that Il-21 may act on adipocytes in a paracrine manner. Consistent with this model, mice lacking Il-21 have smaller adipocytes accompanied by increased expression of transcriptional regulators of oxidative metabolism (*Nrf1* and *Errα*) and fasting responses (forkhead box class O1 [*FoxO1*] and interferon regulatory factor 4 [*Irf4*]). Irf4 has been shown to control WAT lipolysis (11). During fasting, *Irf4* expression is induced by FoxO1, allowing for Irf4 to drive transcription of multiple lipolytic genes, including *Pnpla2* (adipose triglyceride lipase) and *Lipe* (hormone-sensitive lipase) (11). Adipocyte-specific *Irf4* knockout mice gain more weight, have larger adipocytes, and exhibit defective adaptive responses to prolonged fasting and cold exposure, conditions that require functional lipolysis.

Department of Genetics and Complex Diseases, Division of Biological Sciences, Harvard School of Public Health, Boston, MA

Corresponding author: Chih-Hao Lee, clee@hsph.harvard.edu.

See accompanying article, p. 2086.

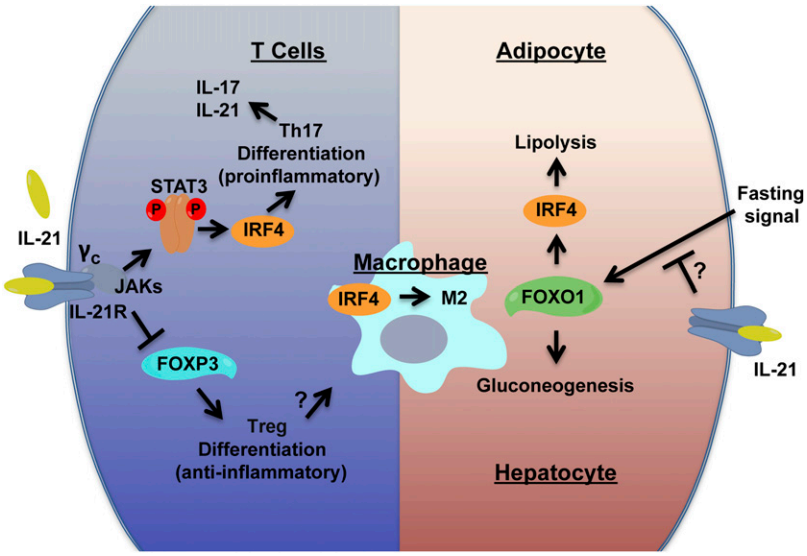

Figure 1—Roles of IL-21 and IRF4 in immune and metabolic regulation. The biological effect of IL-21 is mediated by the IL-21R/γ chain (γc)-STAT3 signaling pathway, which induces the expression of IRF4 in T cells. IRF4 is a multifunctional transcription factor. It is required for IL-21–mediated Th17 cell differentiation. In addition, IRF4 skews macrophages toward the M2 phenotype and promotes lipolysis in adipocytes during fasting. Il-21 deficiency in mice leads to a mixed Irf4 phenotype: a loss of function in T cells (increased Tregs) and gain of function in adipocytes and macrophages (increased lipolysis and M2 activation, respectively). A potential mechanism for the metabolic effect in adipocytes is that Il-21 interferes with the fasting signal that activates the FoxO1-Irf4 axis. A similar link to FoxO1 in the liver could be responsible for the enhanced gluconeogenesis in the liver of Il-21 KO mice. P, phosphorylation.

Fabrizi et al. demonstrate that Il-21 KO mice have constitutively high expression of Irf4 in both the fed and fasted states. There is also an induction of *Pnpla2* and *Lipe* expression compared with wild-type mice, which remain elevated even in the fed state. In addition, Il-21 suppresses isoproterenol-mediated upregulation of *Irf4* and its target genes when directly applied to differentiated 3T3-L1 adipocytes. Consistent with a defect in suppressing lipolysis, Il-21 KO mice have a higher circulating level of fasting free fatty acids.

The increased lipolysis could be a potential explanation for the smaller adipocytes and reduced weight gain found in Il-21 KO mice. However, uncontrolled lipolytic activity can lead to ectopic fat accumulation in tissues, such as the liver, as well as systemic insulin resistance caused by lipotoxicity. On the contrary, Il-21 deficiency improves insulin sensitivity and ameliorates hepatic steatosis in mice fed a high-fat diet. Fabrizi et al. did not directly address how Il-21 KO mice handle excessive free fatty acids. Interestingly, adipose resident Tregs appear to be capable of taking up lipids. The authors suggest that resident M2s and Tregs, both of which use fats as a primary energy source, may help clear fatty acids released by adipocytes.

Fabrizi et al. (9) demonstrate the ability of Il-21 to modulate metabolic homeostasis through inhibition of adipose tissue Tregs and Irf4-mediated lipolysis. However, these findings also raise several unanswered questions. It has been shown that through signal transducer and activator of transcription-3 (STAT3) activation, Il-21 induces *Irf4* expression to promote Th17 cell differentiation (12), suggesting that Irf4 is a downstream effector of Il-21 signaling (Fig. 1). It appears that Il-21 KO mice exhibit phenotypes of Irf4 loss of function in T cells and gain of function in adipocytes and potentially in macrophages, in which Irf4 is thought to regulate M2 polarization (13). Furthermore, despite improved insulin sensitivity, Il-21 KO mice have increased WAT lipolysis and hepatic glucose production. A potential unifying mechanism for the metabolic effects could be that Il-21 interferes with FoxO1 activity in adipocytes and the liver (and thus gluconeogenic gene expression) in a manner similar to that with Foxp3 in Tregs. Future studies using tissue-specific Il-21R KO models in combination with *Irf4* knockout/knockdown approaches will help tease apart the seemingly complex Il-21/Irf4 regulatory network, which will also provide more specific therapeutic targets to control the progression of meta-inflammation and its associated pathologies.

Funding. N.H.K. is supported by National Institutes of Health Interdisciplinary Training in Genes and the Environment grant T32-ES-016645. C.-H.L. is supported by American Diabetes Association grant 1-14-BS-122 and National Institutes of Health grant R01-DK-075046.

Duality of Interest. No potential conflicts of interest relevant to this article were reported.

References

1. Odegaard JI, Chawla A. Pleiotropic actions of insulin resistance and inflammation in metabolic homeostasis. Science 2013;339:172–177

2. Osborn O, Olefsky JM. The cellular and signaling networks linking the immune system and metabolism in disease. Nat Med 2012;18:363–374
3. Hotamisligil GS. Inflammation and metabolic disorders. Nature 2006;444:860–867
4. Wen H, Ting JPY, O'Neill LAJ. A role for the NLRP3 inflammasome in metabolic diseases—did Warburg miss inflammation? Nat Immunol 2012;13:352–357
5. Bhargava P, Lee CH. Role and function of macrophages in the metabolic syndrome. Biochem J 2012;442:253–262
6. Bhargava P, Li C, Stanya KJ, et al. Immunomodulatory glycan LNFPIII alleviates hepatosteatosis and insulin resistance through direct and indirect control of metabolic pathways. Nat Med 2012;18:1665–1672
7. Feuerer M, Herrero L, Cipolletta D, et al. Lean, but not obese, fat is enriched for a unique population of regulatory T cells that affect metabolic parameters. Nat Med 2009;15:930–939
8. Cipolletta D, Feuerer M, Li A, et al. PPAR-γ is a major driver of the accumulation and phenotype of adipose tissue Treg cells. Nature 2012;486:549–553
9. Fabrizi M, Marchetti V, Mavilio M, et al. IL-21 is a major negative regulator of IRF4-dependent lipolysis affecting Tregs in adipose tissue and systemic insulin sensitivity. Diabetes 2014;63:2086–2096
10. Liu SM, King C. IL-21-producing Th cells in immunity and autoimmunity. J Immunol 2013;191:3501–3506
11. Eguchi J, Wang X, Yu S, et al. Transcriptional control of adipose lipid handling by IRF4. Cell Metab 2011;13:249–259
12. Huber M, Brüstle A, Reinhard K, et al. IRF4 is essential for IL-21-mediated induction, amplification, and stabilization of the Th17 phenotype. Proc Natl Acad Sci U S A 2008;105:20846–20851
13. Eguchi J, Kong X, Tenta M, Wang X, Kang S, Rosen ED. Interferon regulatory factor 4 regulates obesity-induced inflammation through regulation of adipose tissue macrophage polarization. Diabetes 2013;62:3394–3403