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Previews

The good and bad of adipose tissue macrophage exosomes in obesity

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Adipose tissue macrophages regulate adipose tissue inflammation and systemic insulin-glucose homeostasis. In a recent study by Ying et al. (2021), M2 polarized bone marrow-derived macrophages secreted exosomes containing miR-690 that, when administered to obese mice, improved glucose-insulin homeostasis. miR-690 reduced expression of *Nadk*, which decreased inflammation and improved insulin signaling.

Obesity is a major risk factor for the development of insulin resistance that promotes the development of type 2 diabetes mellitus (T2DM). Chronic low-grade inflammation is thought to promote the alterations in insulin-glucose homeostasis associated with obesity (Saltiel and Olefsky, 2017). An important initial observation was that increased levels of inflammatory cytokines, such as tumor necrosis factor-α and interleukin-6, in adipose tissue of obese mice and humans were found to contribute to insulin resistance (Fried et al., 1998; Hotamisligil et al., 1993; Perry et al., 2015). Subsequently, it was observed that an influx of proinflammatory macrophages into adipose tissue was a significant contributor to adipose tissue and obesity-associated insulin resistance (Lumeng et al., 2007; Weisberg et al., 2003). More recently, a new mechanistic pathway involving secretion of exosomes by adipose tissue macrophages (ATMs) was found to regulate metabolic and inflammatory interactions between adipocytes and macrophages as well as distal tissues (Ying et al., 2017).

Exosomes are small (50–200 nm) extracellular vesicles surrounded by a phospholipid bilayer that carries the molecular components of one cell to another. As such, exosomes can transmit a broad range of molecular signals through exchange of lipids, proteins, and nucleic acids. For example, adipocytes have been found to secrete lipid-laden exosomes expressing the lipid droplet-associated protein perilipin1, phospholipids, neutral lipids, and free cholesterol that are taken up by ATMs and can induce differentiation of bone marrow-derived precursor cells into ATM-like cells (Flaherty et al., 2019). Recently, exosomes have been found to carry both mRNA and microRNAs (miRNAs) that can modify gene expression of the recipient cells, which has increased interest in the effects of exosomes in metabolic disorders. miRNAs are expressed first as a single strand of RNA, called pre-miRNA (or pri-miRNA), that anneals to itself and is then cleaved by Dicer, which results in double-stranded RNA (O'Brien et al., 2018). The double-stranded RNA is separated by argonaute to produce a mature miRNA-argonaute complex. The binding of the miRNA to argonaute allows it to be trafficked the endosome where it can be packaged within exosomes that make up the multi-vesicular body (MVB) (Siomi and Siomi, 2009) (Figure 1). The MVB can then fuse with the cytoplasmic membrane to release the exosomes.

In the obese state, macrophages within adipose tissue, liver, and skeletal muscle promote insulin resistance. Macrophages can be classified based upon their inflammatory phenotype; namely, in obesity the ATMs are predominantly pro-inflammatory, M1-like macrophages, whereas in lean individuals adipose tissue contains anti-inflammatory, M2-like macrophages. Interestingly, Ying et al. made the novel observation that ATMs taken from obese mice on a high-fat diet (HFD) produced exosomes that contained increased levels of miR-155. The exosomes were isolated and injected into lean mice, which increased insulin resistance and localized in adipose tissue, liver, and muscle (Ying et al., 2017). Mice with miR-155 knockout were significantly more sensitive to insulin when on an HFD, even in the absence of body weight changes.

Building upon these studies, in this issue of *Cell Metabolism*, Ying et al. treated bone marrow-derived macrophages (BMDMs) with IL-4 and IL-13 to generate M2 BMDMs and then isolated the secreted exosomes (Ying et al., 2021). Notably, injection of the secreted exosomes from M2 macrophages into obese mice improved insulin-glucose homeostasis without affecting adiposity. *In vitro* studies confirmed that the exosomes were taken up and improved insulin action in 3T3-L1 adipocytes, L6 myocytes, and isolated mouse hepatocytes. To confirm that miRNAs were causing the insulin sensitivity, they used mice with a knockout of Dicer, which eliminated all miRNAs from the exosomes produced by the M2 BMDMs, and as a result these exosomes failed to improve insulin resistance when injected in obese mice. The authors demonstrated that the M2 BMDM-derived exosomes contained high levels of miR-690 and, when the exosomes were injected, this miRNA was taken up in the relevant metabolic tissues.

To more precisely confirm the role of miR-690, Ying et al. generated an miR-690 "mimic" that was mixed with Invivofectamine and injected into obese mice. The miR-690 mimic localized in adipose tissue, liver, and skeletal muscle, where it improved insulin-glucose homeostasis. Using the TargetScan 7.2 algorithms to predict mRNA targets of miR-690, as well as several *in vitro* experiments,

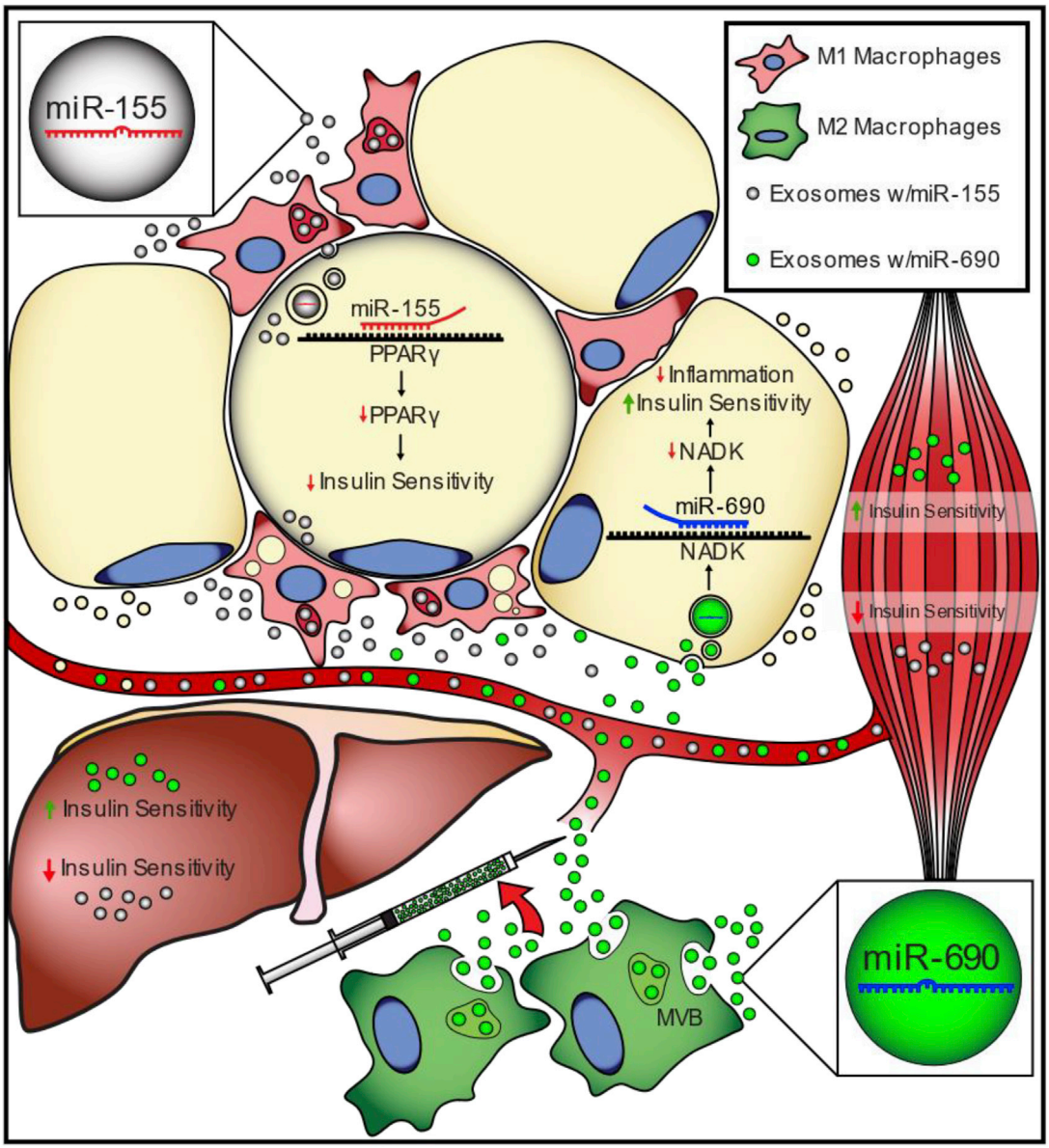

Figure 1. Exosomal microRNAs regulate insulin sensitivity in adipose, liver, and muscle
Exosomes enriched with miR-155 (gray circles) are released from pro-inflammatory M1 macrophages isolated from obese adipose tissue. Lean adipose tissue contains anti-inflammatory M2 macrophages that release exosomes enriched with miR-690 (green circles). When the exosomes containing miR-155 are injected into lean mice, they develop insulin resistance, whereas exosomes containing miR-690 from M2 bone marrow-derived macrophages injected into obese mice improve systemic insulin sensitivity. miR-155 directly targets PPARγ, leading to reduced expression and insulin action. Conversely, miR-690 directly targets and reduces expression of the NAD+ kinase (Nadk), which reduces inflammation in macrophages and increases insulin sensitivity in 3T3-L1 adipocytes and hepatocytes. The exosomes circulate systemically and can be found in adipose tissue, liver, and skeletal muscle.

the researchers demonstrated that the 3′ UTR of the gene *Nadk* is a target of miR-690. *Nadk* is a gene that encodes an NAD+ kinase. To confirm a potential role for *Nadk* in insulin action, the authors used siRNA against Nadk to reduce its expression in 3T3-L1 adipocytes and mouse hepatocytes and found that knockdown improved insulin action.

These exciting observations that macrophage exosomes regulate tissue and systemic insulin-glucose homeostasis, and specifically that miR-690 acts as an insulin sensitizer, raise several important questions. One question that arises is whether macrophages in liver, adipose tissue, and skeletal muscle all express the same miRNAs in exosomes, or if ATM exosomes and their associated miRNAs specifically circulated to liver and skeletal muscle to alter tissue specific and systemic metabolism. Additionally, are there other miRNAs in exosomes from macrophages that are yet to be identified that have important metabolic roles? How is the expression of other genes effected by miR-155 and miR-690 and do the exosome exert cell- and tissue-specific actions? Furthermore,

the mechanistic pathway for how *Nadk* regulates insulin resistance is unknown. Nadk phosphorylates NAD+ to generate NADP+, which is further reduced to NADPH for reductive anabolism. Insulin can stimulate the phosphorylation of Nadk, activating it and leading to increased production of NADP+. The Ying et al. paper did not investigate whether the knockdown of *Nadk* led to a decrease in NADP+ or increased levels of NAD+ (by preventing the conversion of NAD+ to NADP+), which will be an important avenue of future research as increased NAD+ has been shown to improve many metabolic functions. These findings are also of interest for its potential clinical implications in that Nadk, being a kinase, is an enzyme that should be easily targetable by small-molecule drugs.

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DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

Flaherty, S.E., 3rd, Grijalva, A., Xu, X., Ables, E., Nomani, A., and Ferrante, A.W., Jr. (2019). A lipase-independent pathway of lipid release and immune modulation by adipocytes. Science *363*, 989–993.

Fried, S.K., Bunkin, D.A., and Greenberg, A.S. (1998). Omental and subcutaneous adipose tissues of obese subjects release interleukin-6: depot difference and regulation by glucocorticoid. J. Clin. Endocrinol. Metab. *83*, 847–850.

Hotamisligil, G.S., Shargill, N.S., and Spiegelman, B.M. (1993). Adipose expression of tumor necrosis factor-alpha: direct role in obesity-linked insulin resistance. Science *259*, 87–91.

Lumeng, C.N., Deyoung, S.M., Bodzin, J.L., and Saltiel, A.R. (2007). Increased inflammatory properties of adipose tissue macrophages recruited during diet-induced obesity. Diabetes *56*, 16–23.

O'Brien, J., Hayder, H., Zayed, Y., and Peng, C. (2018). Overview of microRNA biogenesis, mechanisms of actions, and circulation. Front. Endocrinol. (Lausanne) *9*, 402.

Perry, R.J., Camporez, J.G., Kursawe, R., Titchenell, P.M., Zhang, D., Perry, C.J., Jurczak, M.J., Abudukadier, A., Han, M.S., Zhang, X.M., et al. (2015). Hepatic acetyl CoA links adipose tissue inflammation to hepatic insulin resistance and type 2 diabetes. Cell *160*, 745–758.

Saltiel, A.R., and Olefsky, J.M. (2017). Inflammatory mechanisms linking obesity and metabolic disease. J. Clin. Invest. *127*, 1–4.

Siomi, H., and Siomi, M.C. (2009). RISC hitches onto endosome trafficking. Nat. Cell Biol. *11*, 1049–1051.

Weisberg, S.P., McCann, D., Desai, M., Rosenbaum, M., Leibel, R.L., and Ferrante, A.W., Jr. (2003). Obesity is associated with macrophage accumulation in adipose tissue. J. Clin. Invest. *112*, 1796–1808.

Ying, W., Riopel, M., Bandyopadhyay, G., Dong, Y., Birmingham, A., Seo, J.B., Ofrecio, J.M., Wollam, J., Hernandez-Carretero, A., Fu, W., et al. (2017). Adipose tissue macrophage-derived exosomal miRNAs can modulate in vivo and in vitro insulin sensitivity. Cell *171*, 372–384.e12.

Ying, W., Gao, H., Dos Reis, F.C.G., Bandyopadhyay, G., Ofrecio, J.M., Luo, Z., Ji, Y., Jin, Z., Ly, C., and Olefsky, J.M. (2021). MiR-690, an exosomal-derived miRNA from M2-polarized macrophages, improves insulin sensitivity in obese mice. Cell Metab. *33*, this issue, 781–790.

Pancreatic β cells put the glutamine engine in reverse

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The metabolism of nutrients other than glucose influences insulin secretion by pancreatic β cells, but the mechanisms involved are incompletely understood. In this issue of *Cell Metabolism*, Zhang et al. (2020) report that reductive glutamine metabolism generates cytosolic NADPH to promote insulin secretion by β cells.

Homeostasis in mammals requires the coordination of metabolism across tissues. This involves the integration of circulating metabolites with hormonal signals that regulate nutrient utilization in specific tissues. This paradigm for whole-body metabolic regulation is best understood with respect to the use of insulin and other hormones to maintain blood glucose levels within a narrow range. A major component of this system is the release of insulin by pancreatic β cells in response to changing blood glucose levels, and defects in this process underlie metabolic diseases including diabetes and obesity. Glucose-stimulated insulin secretion (GSIS) is well studied and is initiated by glucose metabolism through glycolysis, the tricarboxylic acid (TCA) cycle, and oxidative phosphorylation to generate ATP from ADP. The resulting increased ATP/ADP ratio inhibits ATP-sensitive potassium (K_{ATP}) channels in the plasma membrane, which depolarizes the membrane to trigger voltage-gated Ca^{2+} channels to increase Ca^{2+} influx into cells and stimulate release of insulin-containing secretory granules (Figure 1) (Newgard and Matschinsky, 2011). Thus, an increased ATP/ADP ratio serves as the major metabolic output