CO₂ as a Potential Obesogen: A Gas That Will Stick to Your Ribs

Historically considered a simple storage depot for caloric excess, white adipose tissue (WAT) is now understood to be a complex, multicellular endocrine and immune organ that regulates whole-body insulin sensitivity, inflammation, and cardiovascular function (1, 2). Adipogenesis, the process in WAT through which mesenchymal precursor cells are progressively differentiated into mature lipid-laden, insulin-responsive adipocytes, involves sequential waves of transcription factors that cooperatively remodel chromatin and reprogram gene expression (3, 4). Chief among adipogenic transcription factors is peroxisome proliferator-activated receptor (PPAR) γ , a master regulator that is both necessary and sufficient for adipogenesis (3). During obesity, excessive expansion of subcutaneous WAT, which is quantitatively the largest adipose depot in humans, leads to increases in ectopic (i.e., visceral) adipose deposition and to WAT dysfunction (infiltration of proinflammatory macrophages, abnormal adipokine and free fatty acid secretion, and insulin resistance) (5). It is generally thought that adipocyte hypertrophy (cellular lipid overload) drives these untoward effects of obesity, whereas PPAR γ -dependent adipocyte hyperplasia (i.e., recruitment of adipocytes through differentiation of precursors) may be protective (5).

Although the issue is somewhat controversial, tissue hypoxia is generally thought to be an important driver of cellular dysfunction in WAT during obesity (6). Reciprocal interactions between obstructive sleep apnea (OSA) and obesity may conspire in this regard to accelerate metabolic disarray. Obesity is a major risk factor for OSA. On the other hand, OSA is also associated with metabolic syndrome (7), perhaps in part by exacerbating WAT hypoxia and consequent dysfunction (6). In recent years, hypercapnia, another metabolic consequence of OSA and chronic lung disease, has been shown to impact the function of epithelial, immune, and other cell types through effects on cell signaling (8). Whether adipocytes are responsive to changes in local CO₂ remains unclear.

In the this issue of the *Journal*, Kikuchi and colleagues (pp. 570–580) show that elevated ambient CO₂ (i.e., 10% or 7.5% compared with 5%) dose dependently accelerates differentiation of human subcutaneous and visceral preadipocytes in culture (i.e., adipogenesis) (9). This held true under normoxia and hypoxia, under neutral (pH 7.4) and acidic (pH 7.1) extracellular conditions, and during sustained, intermittent (30 min every h, 6 h/d, for 4 d), or transient (6 continuous h/d for 4 d) hypercapnia. By contrast, hypercapnia did not impact adipocyte hypertrophy. The authors provide evidence that CO₂ drives adipogenesis through a pathway involving soluble adenylyl cyclase (sAC)-dependent production of cyclic adenosine monophosphate (cAMP). Specifically, hypercapnia-induced cAMP activates

cAMP response element binding protein (CREB), in turn promoting activation of the proadipogenic transcription factors PPAR γ and CCAAT/enhancer binding protein β . The authors also provide evidence that two additional cAMP effectors that have previously been implicated in adipogenesis, protein kinase A and exchanger protein directly activated by cAMP, are also activated by hypercapnia in a sAC-dependent manner, and also contribute to hypercapnia-induced adipogenesis. With regard to translational relevance, the authors propose that this pathway may put a deleterious positive feedback loop in motion during OSA: hypercapnia accelerates adipogenesis, increasing fat mass and hypoventilation, thereby exacerbating hypercapnia.

This interesting report adds preadipocytes to the growing list of cell types that functionally sense CO₂. sAC, in tandem with carbonic anhydrase, an enzyme that catalyzes interconversion of CO₂ and HCO₃[−], has been shown to serve as a biosensor for CO₂ in other cell types, activating protein kinase A, CREB, and other cAMP effectors (10, 11). Although the cause of its activation is ambiguous, sAC may in fact be activated in response to carbonic anhydrase-catalyzed intracellular HCO₃[−], rather than directly by CO₂ itself (10, 11). Reports that carbonic anhydrase isoforms are dramatically induced during adipocyte differentiation (12) and that carbonic anhydrase III regulates adipocyte PPAR γ and adipogenesis (13) may intriguingly suggest that differentiating preadipocytes are tuned to be either especially sensitive to or buffered against changes in local CO₂/HCO₃[−]. Given that cAMP can induce both adipogenesis and adipocyte lipolysis (14), it remains to be seen how or whether intracellular compartmentalization of hypercapnia-induced cAMP in adipocytes couples specific cAMP effectors to cellular functions, and whether this may vary in different tissue contexts.

The report by Kikuchi and colleagues has several notable strengths. It uses primary human preadipocytes from different anatomic locations rather than murine cell lines, and rigorously shows that the CO₂ effect is dependent on concentration and independent of extracellular acidosis and oxygen conditions. It also confirms the expression of sAC in human visceral and subcutaneous WAT, and convincingly ties sAC-dependent hypercapnia responses into a canonical adipogenic signaling cascade established by the field.

Key questions not addressed by the report include whether CO₂ also biases mesenchymal precursor cells toward preadipocyte commitment, and whether CO₂ impacts PPAR γ -dependent adipogenesis in brown adipose tissue (a thermogenic, energy-consuming type of adipose), and PPAR γ -dependent browning of WAT adipocytes (1, 6). Another key question is whether hypercapnia induces adipogenesis *in vivo*. Appropriately designed rodent models may allow investigators to determine not only

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whether arterial hypercapnia translates into elevated CO₂ in WAT but also whether it impacts the mass and cellular composition of WAT in different anatomic locations, adipokine secretion, and the systemic metabolic status (i.e., insulin sensitivity) of the organism. Studies showing that PPAR γ agonists increase insulin sensitivity in rodents despite increasing fat mass and body weight have revealed that adipogenesis *per se* may not be so bad after all (6), as PPAR γ appears to uncouple obesity from the metabolic syndrome. It will be interesting to determine whether hypercapnia also activates PPAR γ in other WAT cell types, in particular adipose tissue macrophages and T regulatory cells, in which PPAR γ promotes antiinflammatory, metabolically beneficial cellular programs (6). Conversely, it is interesting to consider the possibility that hypercapnia may activate PPAR γ in other tissues (e.g., kidney and bone) to induce fluid retention and bone mass loss—complications that have been linked to the use of thiazolidinedione PPAR γ agonists in humans and also associated with OSA (3, 6, 15, 16).

In closing, the report by Kikuchi and colleagues reveals adipocytes and the master regulatory transcription factor PPAR γ to be targets of the simple gas CO₂. Future work is now warranted to test whether these findings hold true *in vivo*, what the metabolic implications are for humans, and whether these signaling effects of hypercapnia translate to other cell types and functions. ■

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