The goal of physiology has always been to understand how organisms function. In a far-reaching field that encompasses aspects of biology, chemistry, and physics, the three mainstays of the physical sciences, terms such as homeostasis, or Claude Bernard’s concept of the ‘milieu interior,’ have often been used to define the balance amongst many diverse processes that yield the net outcome determining health or disease, life or death. In our drive to understand these processes, we have taken a reductionist approach to increasingly smaller and more detailed avenues of inquiry. These avenues have led to fantastic discoveries and the development of entire disciplines, including genomics, transcriptomics, proteomics, and most recently (or, in the mind of some, a return to) metabolomics. Investigations in these realms have increased our understanding of the detailed components that comprise physiological processes, and undoubtedly will continue to do so for a long time. However, there is a growing recognition that reintegration is needed to truly understand the “milieu interior.” In this issue of *Cardiovascular Research*, Willems et al. [1] have taken such a step forward with regard to our understanding of the ubiquitously important molecule, adenosine, in the heart.

The purine metabolome itself is complex, and continued exploration still yields surprises. Willems et al. have chosen to reintegrate receptor-mediated actions of adenosine with questions regarding the metabolic regulation of endogenous adenosine concentrations. From the metabolic vantage, it is interesting that in this endeavor they begin with adenosine deaminase (ADA). To some investigators, this may seem counterintuitive. ADA has often been regarded as a cytosolic enzyme of questionable importance to the purine metabolome. In considering purines as molecules involved in autocrine (and paracrine) signalling, effective interstitial, rather than intracellular, concentrations of the nucleoside are the primary focus. However, it is now recognized that extracellular ADA plays an important role in regulating concentrations of adenosine relevant to cellular signalling. Cardiac fibroblasts express the enzymes involved in the cAMP–adenosine pathway [2]. Although ADA is not encoded with a signal sequence that would lead to secretion or membrane insertion, it manages to be presented into the interstitial compartment (through as yet unknown mechanisms), and is a critical enzyme in the interstitial cAMP–adenosine pathway. In that “milieu,” evidence suggests that ADA is a critical player in regulating endogenous adenosine-mediated responses [3].

Investigators have often focused on pharmacological manipulation of ADA to influence cardiac purine metabolism. The most commonly used approach has been to inhibit ADA. The presumption in this approach is that ADA inhibition will have a singular effect: increase available endogenous adenosine to exert cardioprotection via its receptors. The results of these studies have not always been consistent with this premise. When erythro-2-(2-hydroxy-3-nonyl)adenine (EHNA) was used to inhibit ADA, the effects were beneficial or ameliorative [4–7]. In contrast, deoxycoformycin (dCF), which inhibits both ADA1 and ADA2 isozymes, led to ineffective cardiac protection, or even injury [8–11]. Many explanations are possible for these disparate results, including the differential actions of EHNA and dCF on the ADA isozymes, or the lack of recognition of ADA as a potential allosteric modulator of adenosine receptor function. Both of these are considered by
Willems et al. [1]. They employ a transgenic mouse genetically deficient in ADA1 (mice do not express ADA2) and test for potential allosteric effects of this deficiency on A1 adenosine receptor sensitivity.

Because the other central focus of the transgenic approach applied by Willems et al. is to bring A1 receptor-deficient mice into the design, they do not focus on adenosine-mediated actions alone. Oxyradicals must be considered amongst the various molecules associated with interstitial purine metabolism. Excessive oxyradical production can cause damage through direct oxidative processes or can serve as a stimulus for cell signalling, as in the case of proinflammatory cytokine expression associated with NF-κB activation [12]. Adenosine deaminase is responsible for the first step in degrading endogenously produced interstitial adenosine in the heart. Further metabolism would rapidly shuttle cardiac adenosine into the xanthine oxidase pathway with subsequent generation of oxyradicals, which can contribute to deterioration of cardiac function through a number of mechanisms [13]. Willems et al. provide a nicely balanced treatment of this perspective, as well.

By combining a uniquely viable ADA-deficient mouse line with an adenosine subtype receptor-deficient line, Willems et al. present some very interesting data that must be considered from both metabolic control and cell signaling perspectives, effectively integrating these two arenas. It is clearly not the end of the story, but it is a worthwhile beginning.

References


