Discoveries can take unpredictable paths! In a review presented in the current issue of the Journal of Innate Immunity, Meijer et al. [1] describe the rationale for evaluating roscovitine for the treatment of cystic fibrosis (CF) patients chronically infected with Pseudomonas aeruginosa. Roscovitine is a low-molecular-weight inhibitor of cyclin-dependent kinase (CDK) discovered over 20 years ago during studies on the regulation of cell division using a rather unlikely research subject, starfish oocytes [2]. Marine invertebrate oocytes and eggs have been used for decades to investigate cellular and molecular mechanisms involved in the regulation of the cell division cycle as these cells have a natural break at specific stages in the cell cycle [3]. In parallel to Xenopus oocytes and genetic models such as yeast and Drosophila, they have allowed the identification of the CDK1/cyclin B protein kinase as the major regulator in G2/M cell cycle transition. In fact, this discovery eventually led to the awarding of the Nobel Prize in Physiology or Medicine to Paul Nurse, Tim Hunt, and Leland H. Hartwell in 2001. CDK1/cyclin B is the archetype of CDKs that were found not only to be major players in all steps of the cell cycle but also to display other functions including a role in the central nervous system (CDK5/p25). Given that active CDK1/cyclin B is present in huge quantities in starfish M-phase oocytes, the group of Laurent Meijer at the CNRS (France) developed an affinity chromatography purification method combined with a simple kinase assay to search for potential pharmacological inhibitors, and this led to the discovery of roscovitine in Roscoff [4]. This molecule has been used extensively as a pharmacological tool to investigate the cell cycle, synchronize cells, and trigger cell cycle arrest or apoptosis, in order to evaluate its effects on numerous diseases ranging from cancers, viral infections, or neurodegeneration to polycystic kidney disease.

While originally roscovitine was believed to exert its effects mainly on proliferating cells, Adriano Rossi and his colleagues (University of Edinburgh) reported that roscovitine also affected neutrophils, key cells in innate immunity deprived of proliferative capacities [5–10]. They discovered that roscovitine could trigger apoptosis of neutrophils, thereby favoring their phagocytosis by macrophages to promote the resolution of inflammation [11]. Notably, this activity was due to the inhibition of CDK7 and CDK9 involved in the regulation of RNA transcription [12]. These observations opened novel perspectives in the field of anti-inflammatory drugs and especially in chronic lung inflammation like that observed in CF patients.

CF is indeed a complex genetic disease [13] that involves the mutation of the chloride channel CFTR gene, where the deletion of F508 is the most frequent mutation.
This mutation leads to an incorrectly folded protein which is not appropriately localized to the plasma membrane channel, and this ultimately results in the manifestation of CF syndrome including thickening of the mucus within the airways and sustained chronic lung disease due to colonization by P. aeruginosa and sustained neutrophil-dominated inflammation [14].

As previously documented, the CFTR mutation seems to directly affect the function of cells from innate immunity. In neutrophils, the mutation in CFTR has been shown to affect phagosome formation [15]. In macrophages, the seminal discovery by Deborah J. Nelson (University of Chicago) of a high intraphagolysosomal pH within alveolar macrophages when CFTR was deficient [16] was the start of another unexpected finding. This high intraphagolysosomal pH prevents lysosomal proteases and lipases from digesting phagocytosed bacteria, and this may explain the high rate of bacterial infections in the lungs of CF patients. Roscovitine was tested for its potential effect on intraphagolysosomal pH and surprisingly restored the acidic ability of phagolysosomes to destroy bacteria, ultimately improving the bactericidal activity of CFTR-deficient alveolar macrophages [17]. However, this effect was not due to any correcting effect of roscovitine on F508del-CFTR since identical results were obtained in the complete absence of CFTR (cftr−/− macrophages). Again, the effects of roscovitine were not associated with kinase inhibitory action as illustrated by the fact that the kinase-inactive roscovitine metabolite is active while the kinase-active (S)-isomer of roscovitine displays no intraphagolysosomal pH-correcting ability.

The final unexpected twist that brought roscovitine in the CF field came from the discovery by the group of Frédéric Beck (University of Poitiers) that roscovitine was displaying some corrector effect on F508del-CFTR [18]. Correcting mutated CFTR trafficking and intracellular localization constitutes one of the promising approaches to address CF treatment [19]. Very unpredictably, the correcting effect of roscovitine was found to be independent of its effects on protein kinases!

Safety studies of roscovitine have been carried out in healthy volunteers and cancer patients. Given the fact that roscovitine displays anti-inflammatory and analgesic properties and given the oral availability roscovitine, its distribution in the lung and its short-half life, it was tempting to repurpose this drug candidate for an evaluation in chronically infected CF patients [20]. Although roscovitine has many weaknesses, we can expect that the results of this initial clinical trial with this ‘first-in-class’ drug (at least as far as CF is concerned) will provide promising results that can provide orientation for either further optimization of roscovitine administration or the development of alternative, roscovitine-derived drug candidates.

It is ironic that over 30 years of work on the fundamental aspects of the marine invertebrate cell cycle and on kinase inhibition have ultimately led to a drug candidate acting on processes unrelated to cell division through a kinase-independent pathway on inflammatory cells that do not have any proliferation capacities! As a complex disease, CF might benefit from multifactorial therapeutic approaches, and it seems that roscovitine is endowed with multifaceted therapeutic power, which it might be useful to explore in other conditions where innate immunity is disturbed.

References


