LETTER TO THE EDITOR

Possible Role of ABCB1 in Lysosomal Accumulation of Azithromycin in COVID-19 Therapy

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Dear Editor,

The antiviral use of azithromycin in coronavirus disease 2019 (COVID-19) was recently reported by Damle *et al*.[1] Its combination with hydroxychloroquine did not aim at preventing bacterial superinfection as often believed, but at benefiting from their common lysosomotropic properties which buffer the acidic conditions (pH 4–5) of the endolysosomal lumen where severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) transits following its angiotensin-converting enzyme 2 (ACE-2) receptor-mediated endocytosis.[2] These two powerful cationic and amphiphilic drugs increase up to neutrality the intravesicular pH causing disorders in lysosomal functions such as enzyme inhibitions involved in the virus replication cycle.[3] We recently hypothesized that the adenosine triphosphate (ATP)-binding cassette subfamily B member 1 (ABCB1) (P-glycoprotein) could be involved in this reported synergistic effect.[4] ABCB1 is expressed both on plasma cellular and lysosome/endosome membranes of the lung epithelial cells. Therefore, the substrate transport is directed from the cell cytosol to the extracellular fluids at the level of the plasma membrane and to the lumen of lysosomes, respectively. Thus, the intracellular ABCB1 transport could result in increasing the endolysosomal accumulation of ABCB1 substrates. Damle *et al*. reported that azithromycin is not a sensitive substrate but an inhibitor and that hydroxychloroquine is both a substrate and an inhibitor of ABCB1. On the contrary, several *in vitro, in vivo*, and clinical studies clearly suggest that azithromycin is an ABCB1 substrate, whereas hydroxychloroquine was never characterized as a substrate or an inhibitor with usual *in vitro* transport assays but assumed to be a moderate inhibitor in two pharmacokinetics which have shown a slight increase of the oral bioavailability of two ABCB1 substrates, digoxin and nelfinavir, when coadministered with hydroxychloroquine.

These discrepant points of view did not challenge Damle's message that "P-gp [P-glycoprotein] efflux would not be expected to be rate-limiting" for the lung distribution because the neutral forms of both drugs have extensive volumes of distribution. Nevertheless, we could raise the question of the role of ABCB1 at the endolysosomal level vs. the substrate/inhibitor status of these drugs. On the one hand, ABCB1 can more extensively enhance the substrate azithromycin trapping as expected by the sole diffusion mechanism.[5] On the other hand, the possible ABCB1 inhibition by hydroxychloroquine could limit this effect. These dual effects on ABCB1 could be further assessed using *in vitro* cellular models to better clarify the possible interplay between the two drugs and ABCB1.

Thus, we recommend considering ABCB1 at the endolysosomal network for a better understanding of the mechanism of action of azithromycin when combined with hydroxychloroquine. Since the clinical benefit of this drug combination remains uncertain, further experimental investigations are now needed to determine the role of the endolysosomal ABCB1 in COVID-19 therapy.

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