Small molecule retention by Gram negative bacteria can be measured, and predicted based on a large-scale LC-MS screen

F. Le Goff, L. Christen, G. Bourquin, J. Hazemann, G. Pierlot, D. Ritz, G. Rueedi and O. Peter

Actelion Pharmaceutical Ltd, Allschwil, Switzerland

E-mail: francois.legoff@actelion.com

The increasing prevalence of difficult or even impossible-to-treat infections caused by multidrug resistant Gram-negative bacteria has led to a healthcare crisis, necessitating the search for new drugs with novel modes of action. Gram-negative bacteria are composed of two membranes with orthogonal properties and are equipped with powerful efflux pumps in order to prevent any xenobiotic molecules from accumulating inside the cell. Therefore, the design of drugs reaching the cytoplasm to elicit their antibacterial effect in Gram-negative bacteria has been identified as a major hurdle on the way to developing novel Gram-negative antibiotics[1].

To address the problem of cytoplasmic drug accumulation in a broader sense, we have developed a method that allows for the direct measurement of small molecule retention by Gram-negative bacteria, irrespective of antibacterial activity. The ultimate goal was to identify the structural elements that determine bacterial compound uptake and/or adherence.

A set of more than 13'000 structurally diverse compounds was tested for retention by TolC mutated *E. coli*. 49% of the compounds could be detected by MS and clearly classified. Of these, 45% were found to be retained by the bacteria, while 55% were not.

Interestingly, no single physicochemical property clearly correlates with the measured bacterial compound retention.

The screening results were then used to train a computational prediction tool based on a machine learning algorithm. It could be experimentally confirmed that this tool classifies unknown compounds correctly as retention Positive in 77.8% of cases, and as Negative in 74.4%.

References

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