Inhibitors and activator of the P-glycoprotein (P-gp) efflux pump modulate the accumulation of daptomycin in THP-1 macrophages and its intracellular activity towards *S. aureus*

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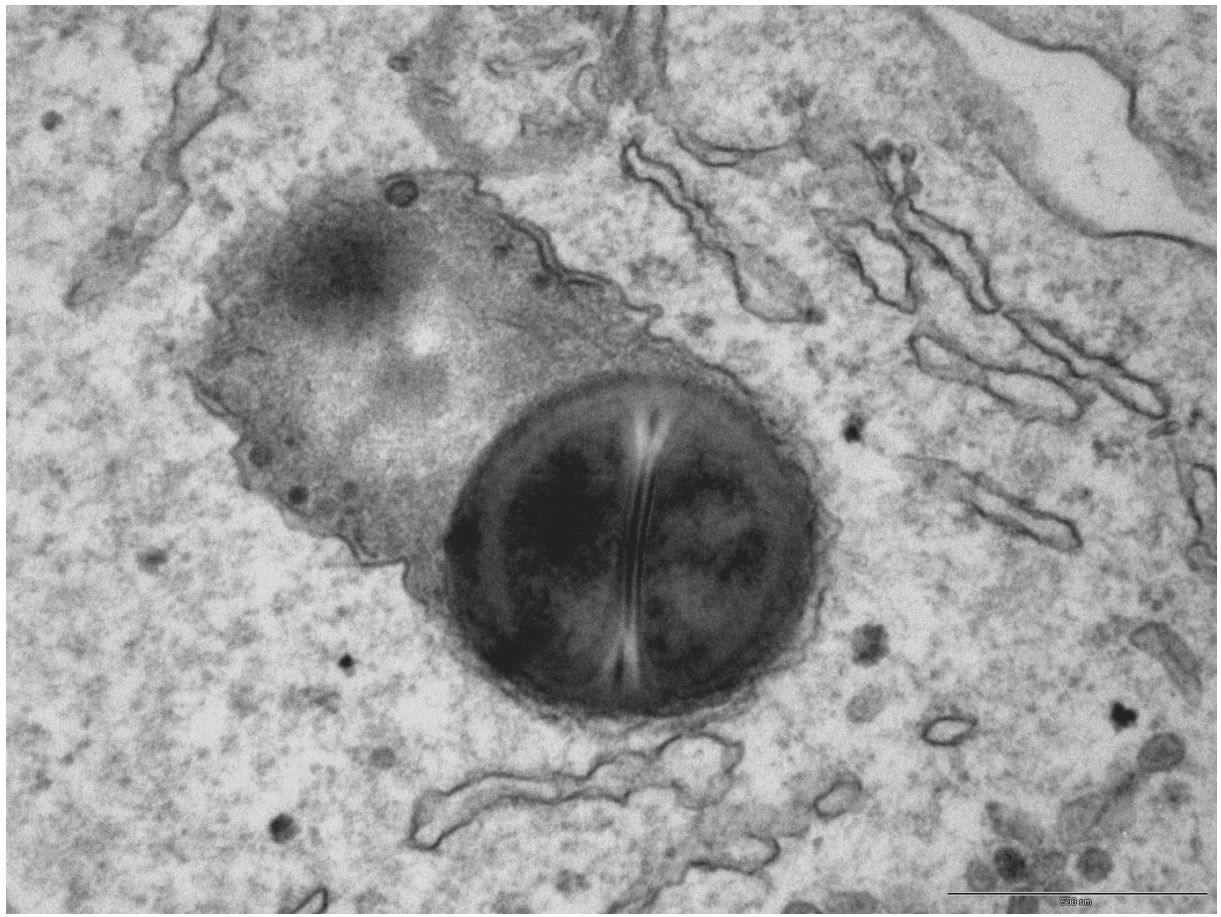
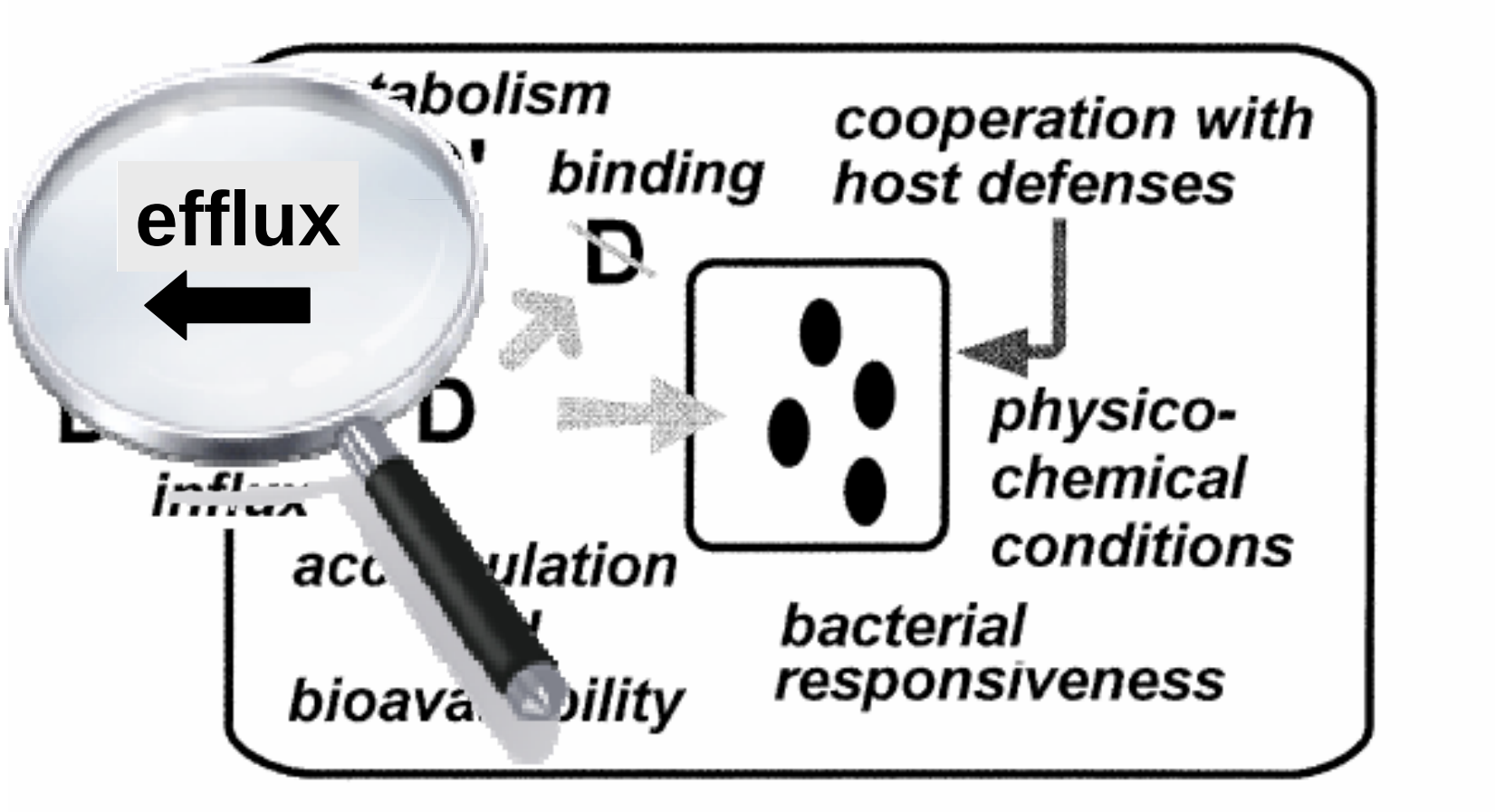
Intracellular *S. aureus* is a reality …

and you need antibiotics to fight them

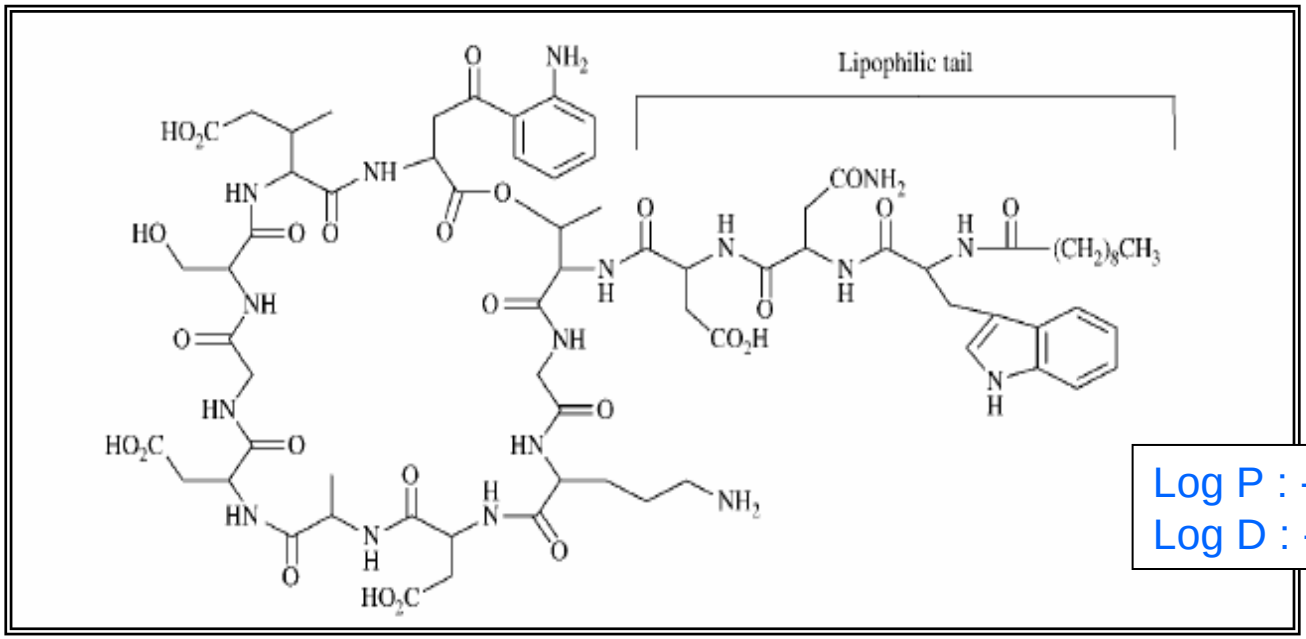




Eukaryotic efflux transporters can modulate the cellular concentration and the intracellular activity of antibiotics

Carryn et al., Infect Dis Clin N Am, 2003

Daptomycin

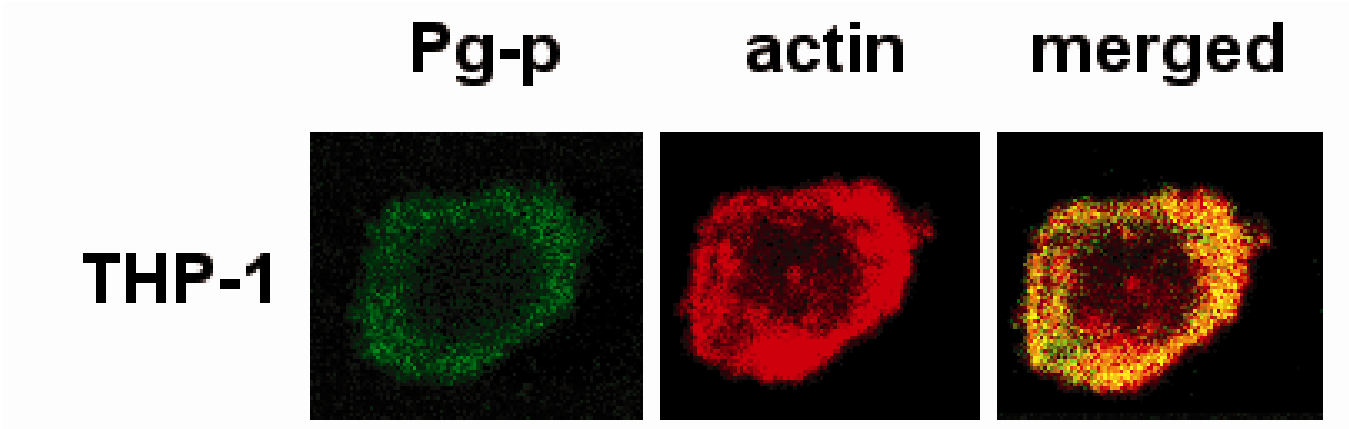


Log P : -4.1 ± 0.3
Log D : -9.6 (pH 7)

- Cyclic lipopeptide with rapid and extensive bactericidal effect towards *S. aureus*
- Activity against intraphagocytic *S. aureus* (P829, ECCMID 2007)

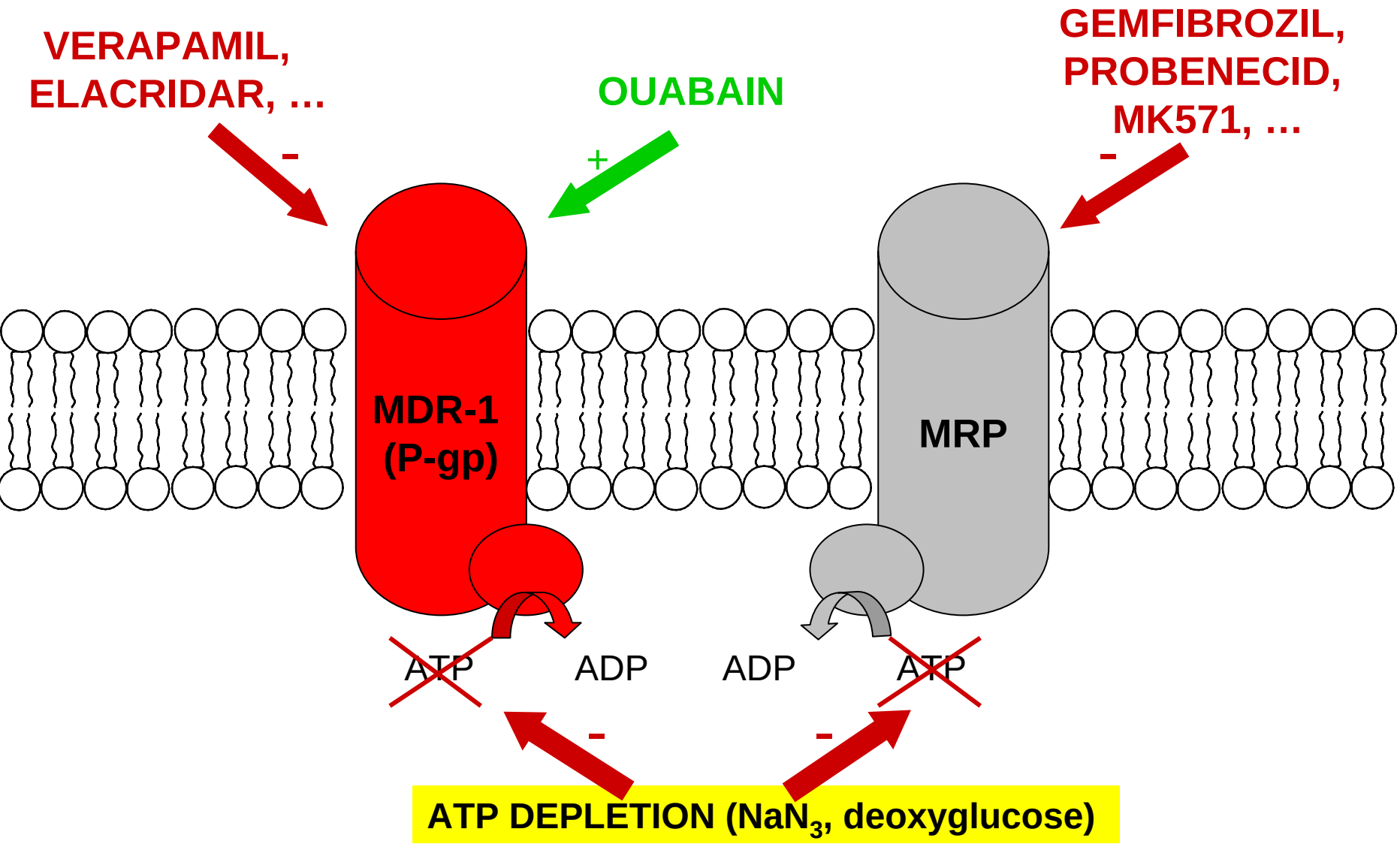
Context and aims of the study

- P-gp (MDR1; belonging to the ABC transporters superfamily) is a major drug efflux system in eucaryotic cells, and is expressed in macrophages…
- DAP is potentially active against intracellular *S. aureus*…
- Could DAP be a substrate of P-gp ?

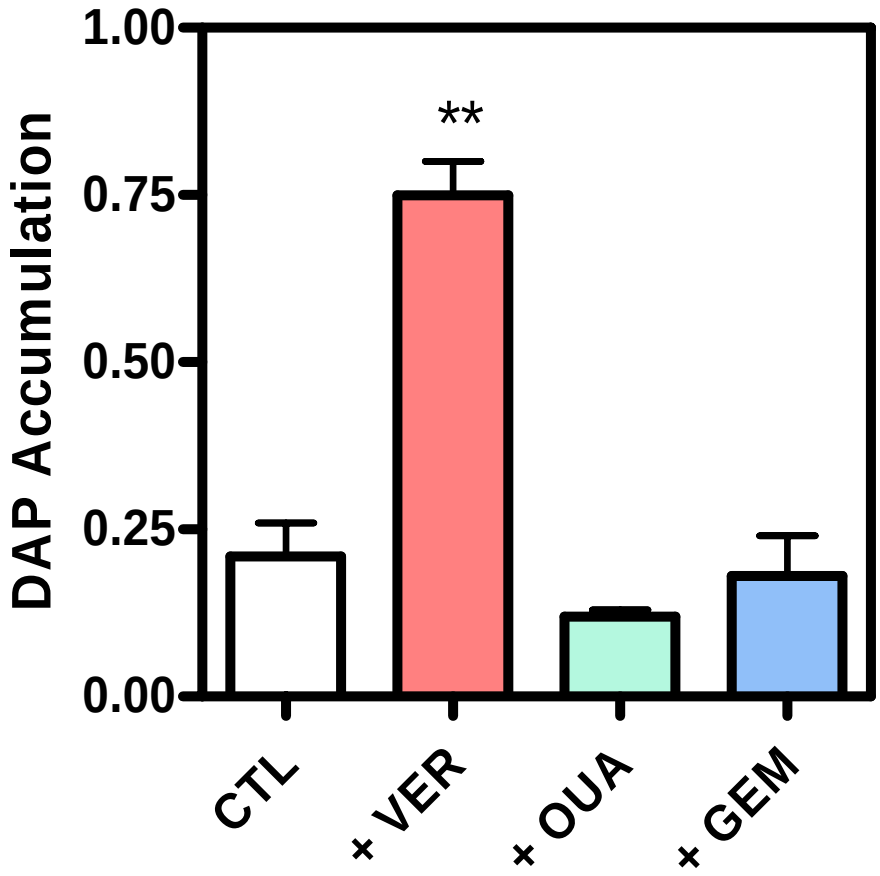


Lemaire *et al*., submitted

How to modulate ABC-transporters ?



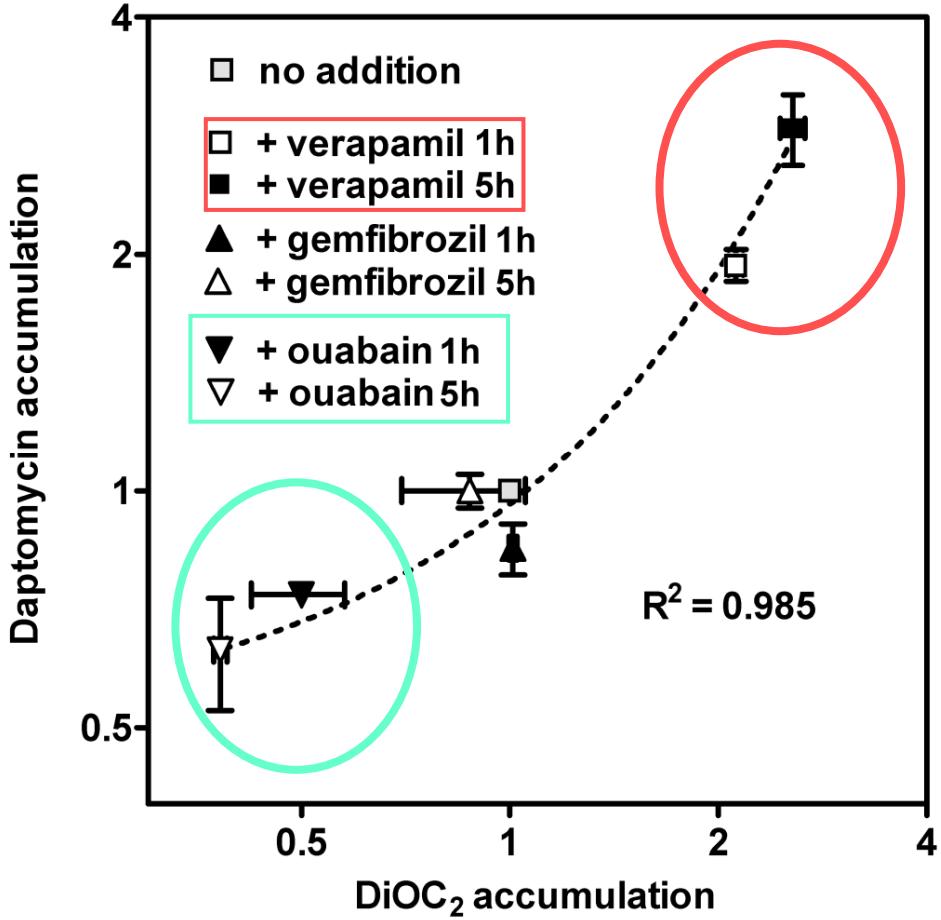
Modulation of the DAP accumulation by P-gp and MRP inhibitors



Accumulation of DAP :

- ↗ with verapamil
- ↘ with ouabain
- No effect of gemfibrozil

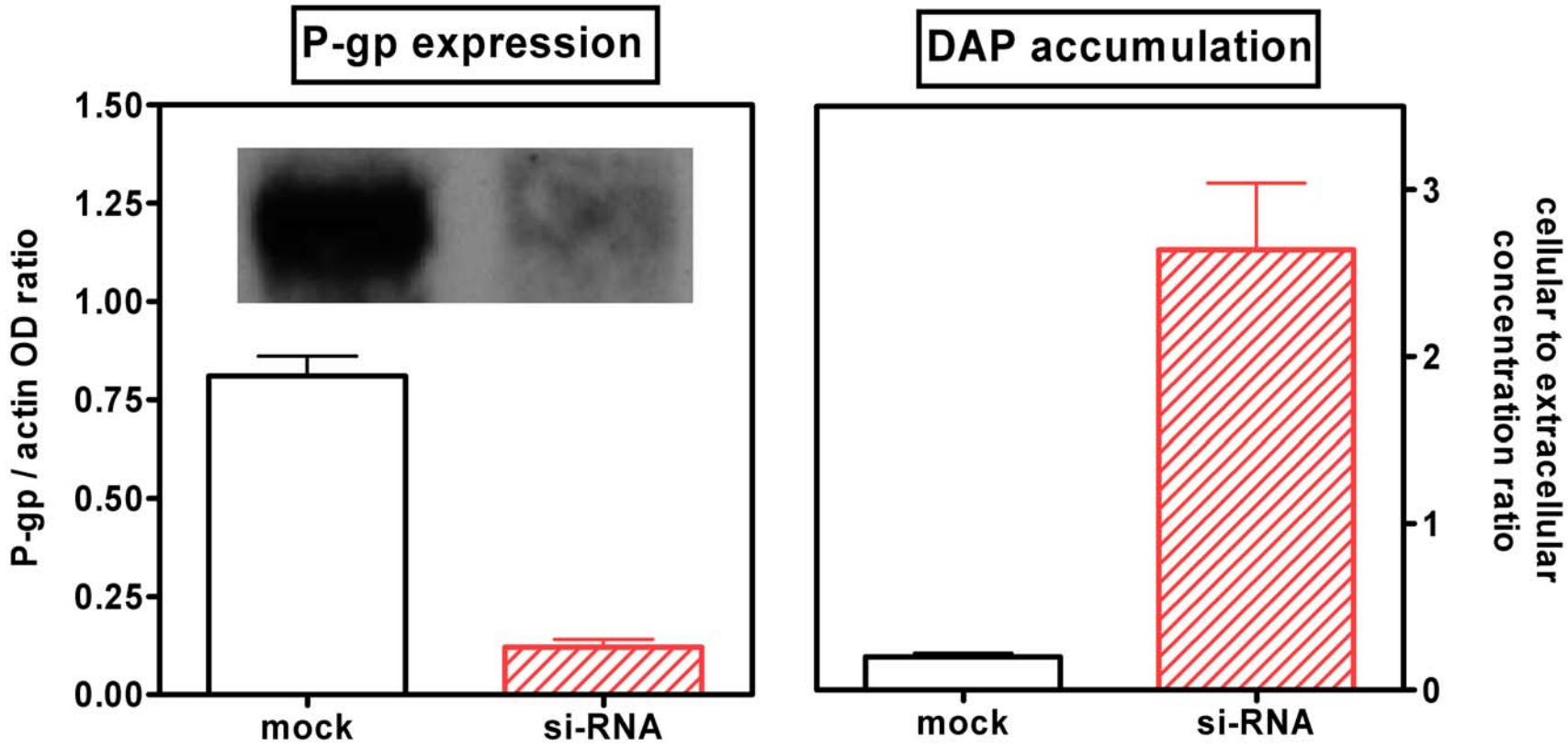
Correlation of DAP and $DiOC_2$ * accumulation



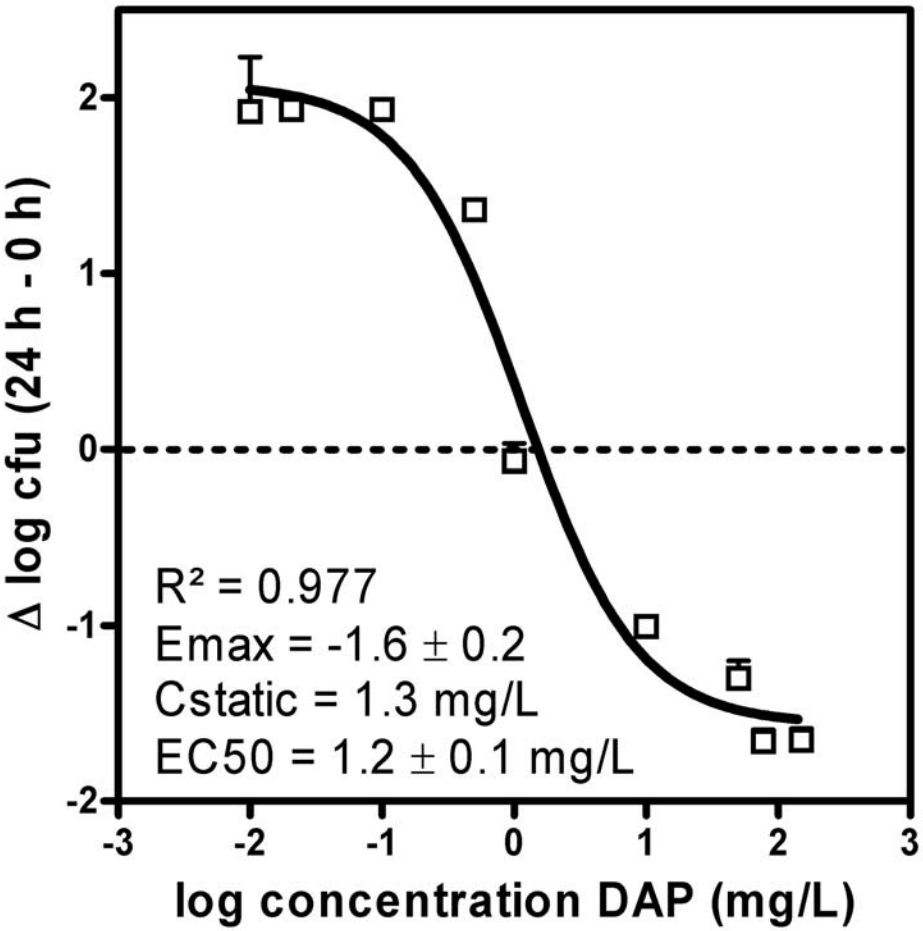
* 3-ethyl-2-[5-(3-ethyl-2(3H)-benzoxazolylidene)-1,3-pentadienyl]-iodide ($DiOC_2$), a model substrate of P-gp

Accumulation of DAP and $DiOC_2$ are influenced in parallel by P-gp modulators (verapamil & ouabain)

Silencing of *mdr1* by si-RNA and modulation of DAP accumulation



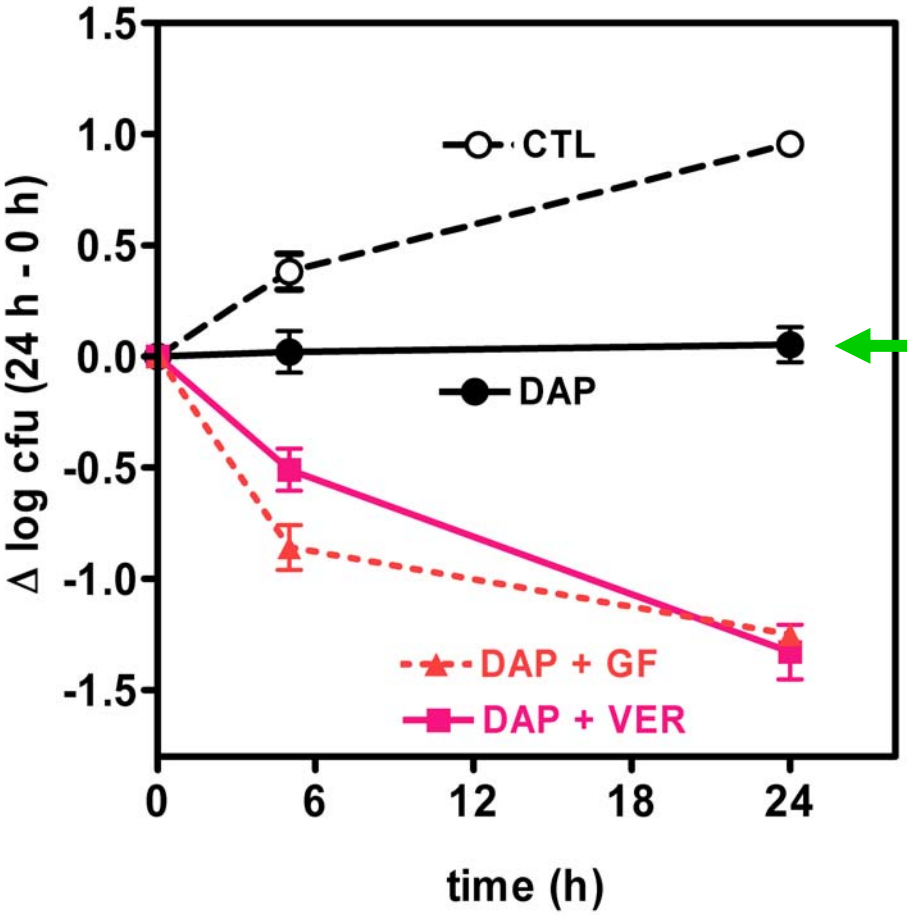
Assessing DAP intraphagocytic activity *



DAP is active against intraphagocytic MSSA

model: see Barcia-Macay et al., AAC 2006; 50:841-51.

Modulation of DAP intracellular activity by P-gp inhibitors



static concentration (1 mg/L)

Intracellular activity is increased by co-incubation with verapamil or elacridar

(no effect of these inhibitors on bacterial growth when given alone)

Conclusions

- DAP is a substrate of P-gp efflux pump, causing suboptimal accumulation and decreased activity of DAP towards phagocytized *S. aureus* in THP-1 macrophages
- Yet, DAP structure and biophysical properties (Log D) are quite remote from those of other typical P-gp antibiotic substrates (e.g., azithromycin *), suggesting the existence of different recognition sites and/or DAP complexation with lipids
- The role of P-gp in the handling of DAP *in vivo* will need to be critically assessed.

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* Seral et al. AAC 2003; 47:1047-51

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