

Further information on the two dimensional chloroquine gels utilised in the Baxter lab;

In first dimension chloroquine intercalation at 0.5µg/ml introduces $\approx +7$ supercoils from relaxed state

\therefore Chloroquine intercalation at 1µg/ml introduces upto +14 supercoils above the relaxed state

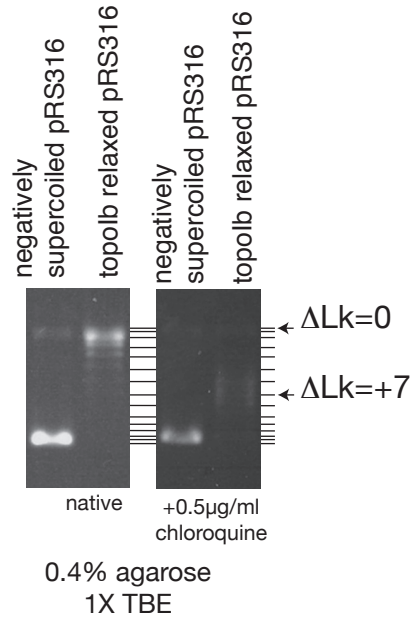
Expressed topologically.....

$$Lk = Tw + Wr$$

Following topo I (b) treatment $Lk = Lk_0 \approx Tw$ and $Wr \approx 0$

Following incubation in 0.5µg/ml chloroquine in 1xTBE $Wr \approx +7$

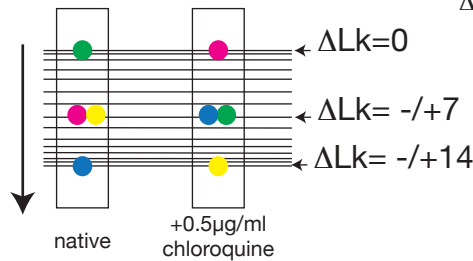
Assuming chloroquine intercalation approximately linear for intercalation between Wr 0 to 14 incubation of relaxed plasmids in 1µg/ml chloroquine 1xTBE generates $Wr @ +14$. (N.B. because chloroquine intercalation decreases with increasing Wr , theoretically Wr of relaxed plasmid in 1µg/ml chloroquine could be $\gg 7$ and ≤ 14 .)



Model of behaviour of 3 topoisomers with defined starting states resolved under the conditions described:

- $\Delta Lk = +7$ ● (yellow)
- $\Delta Lk = 0$ ● (green)
- $\Delta Lk = -7$ ● (pink)
- $\Delta Lk = -14$ ● (blue)

first dimension 0.4% agarose 1X TBE



Chloroquine lane excised incubated in 1xTBE and chloroquine at 1µg/ml before resetting in 1.2% agarose, 1X TBE, Chloroquine at 1µg/ml.

