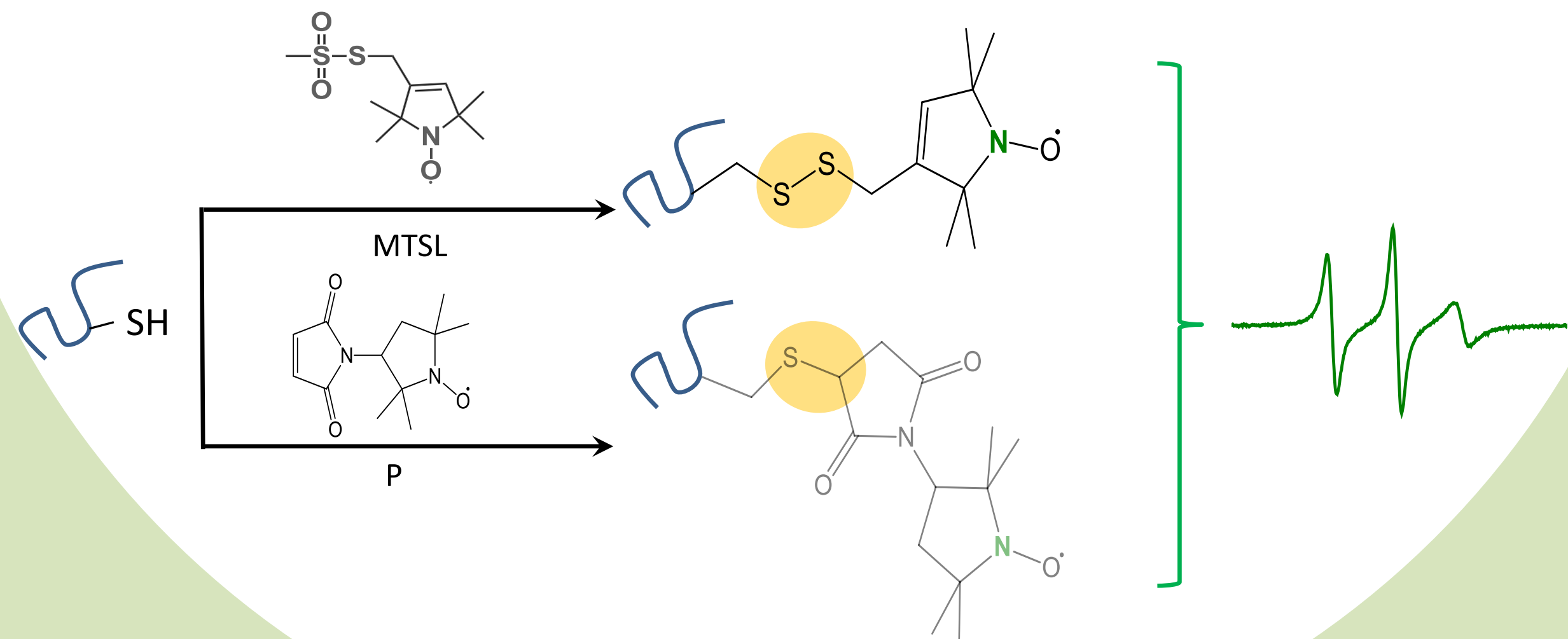


Site Directed Spin Labeling Approaches

①

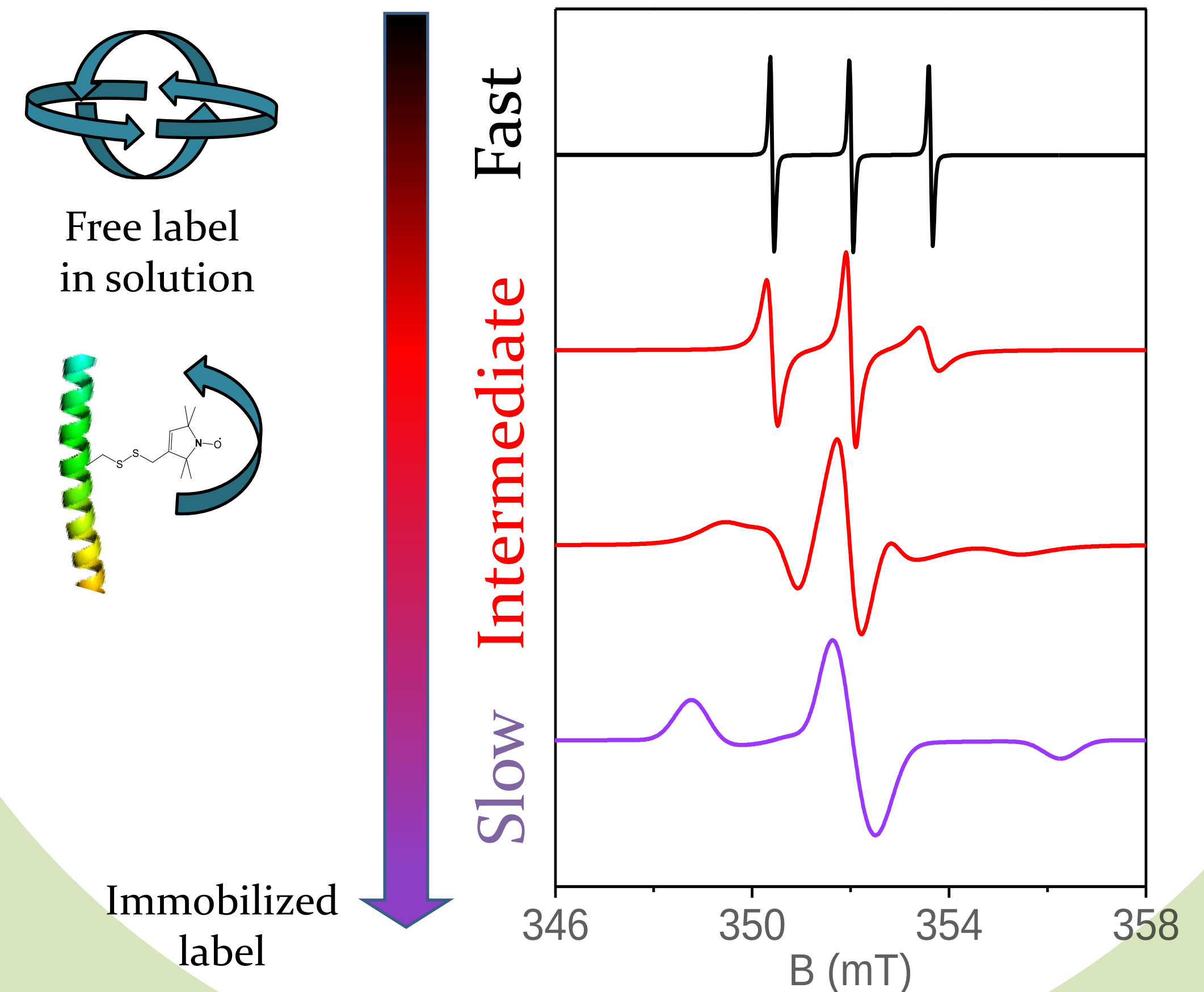
A powerful technique to study dynamic and structural properties of proteins is SDSL. It is based on **the insertion of a paramagnetic probe**, a nitroxide, onto a protein which enables its analysis using electron paramagnetic resonance spectroscopy (EPR).

Continuous waves (cw) EPR can provide information on the environment of the labels under various conditions.

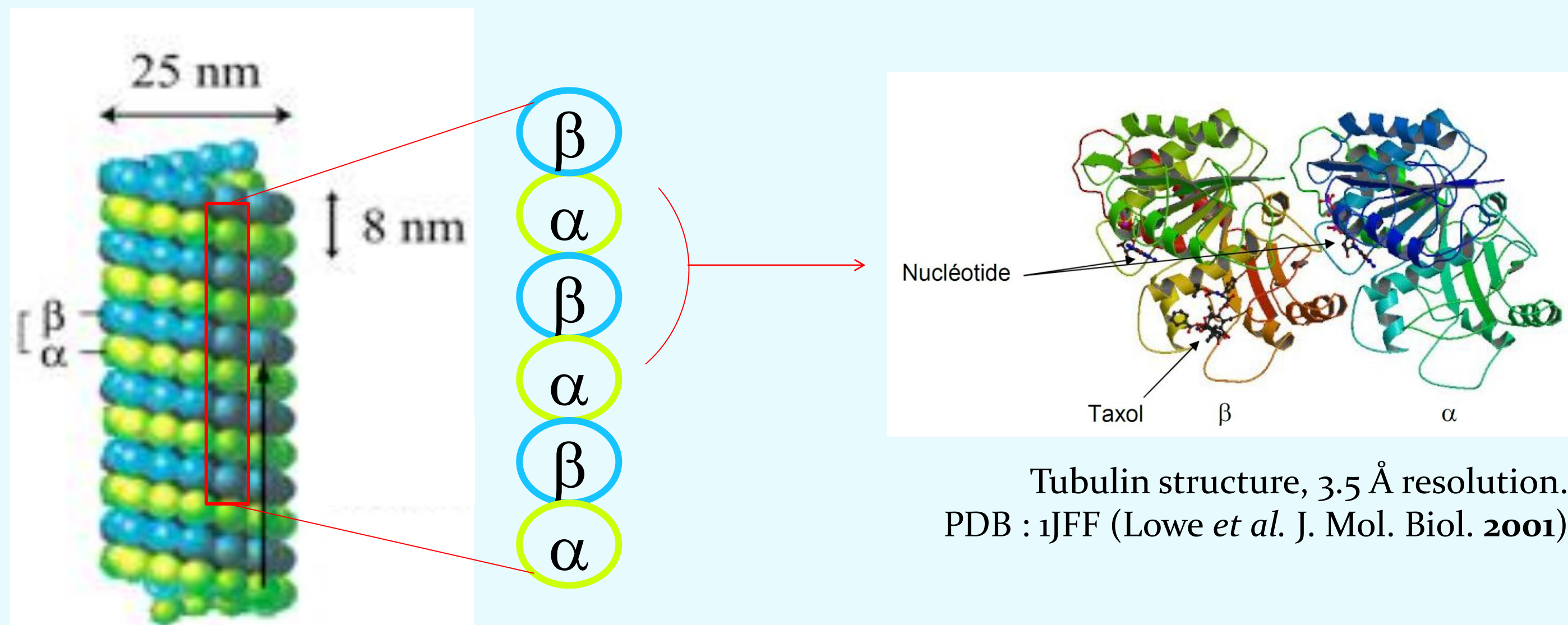


Site Directed Spin Labeling Approaches

②



Microtubules cytoskeleton component



- Tubular polymer composed of protofilaments of $\alpha\beta$ -tubulin heterodimers
- Dynamic structure regulated by *MTs Associated Protein (MAP)*

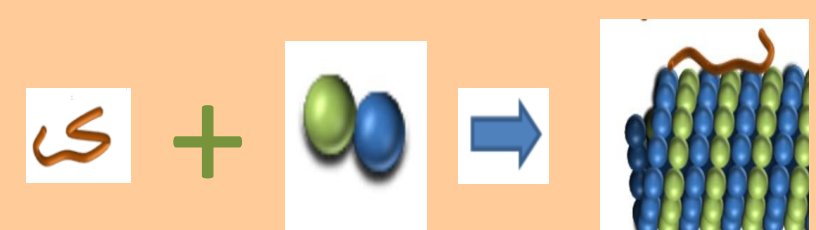
Tau is a MAP found in nerve cells

Function

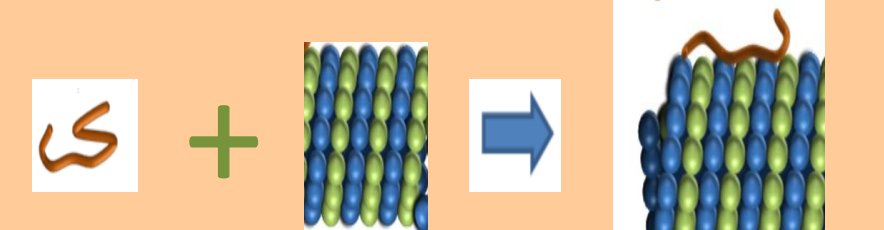
- Stabilize MTs and promote their assembly
- In numerous neurodegenerative diseases called Tauopathies (Alzheimer disease)

Structure

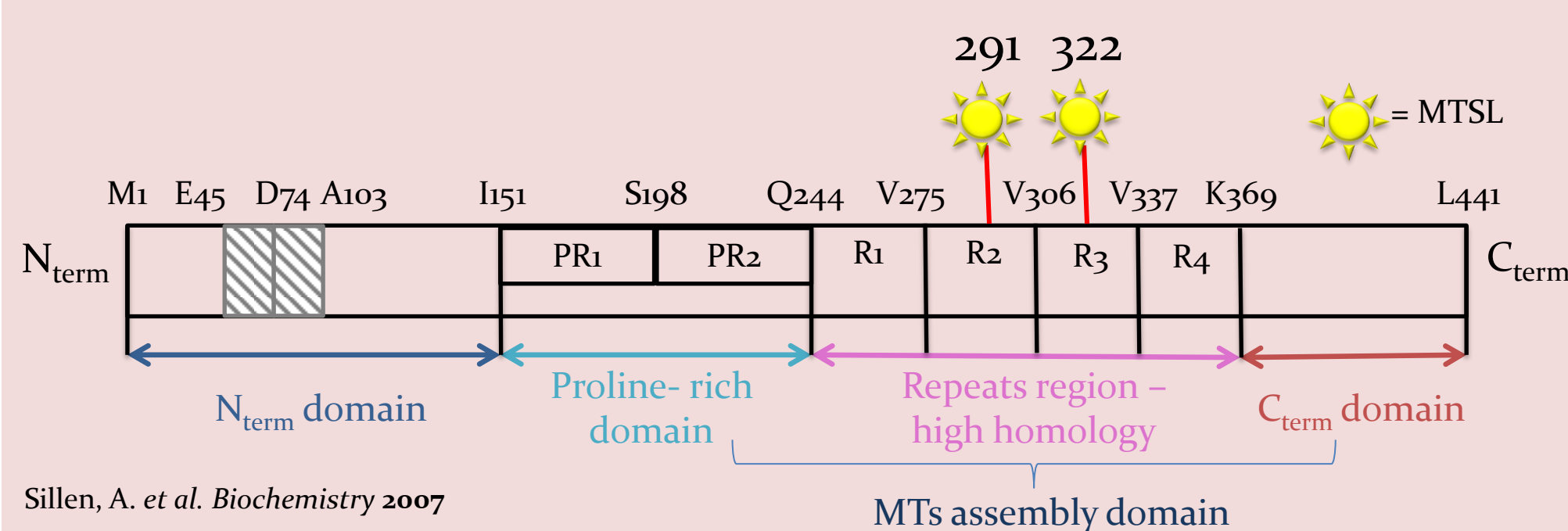
- Very long Intrinsically Disordered Protein (IDP)
- Imperfect Repeats Region
- Tau remains extended and dynamics upon binding
- Where / how does Tau bind to MTs ?



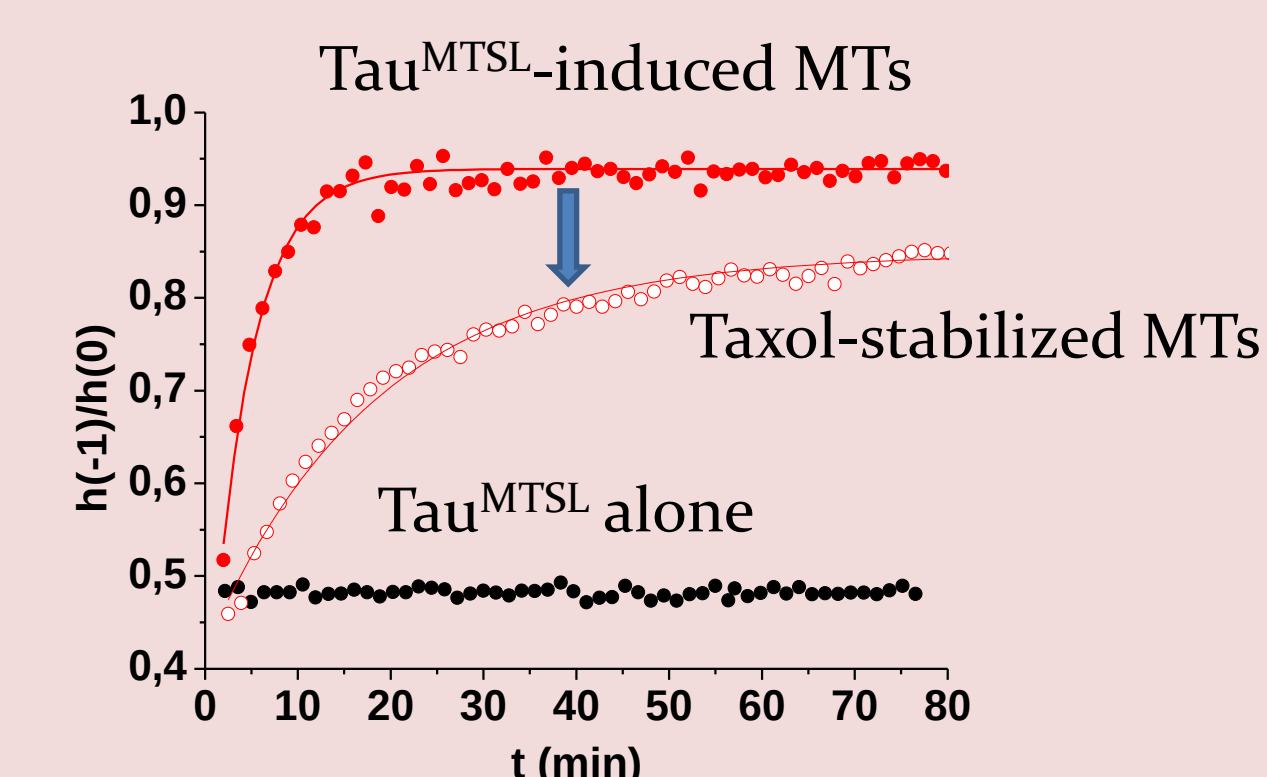
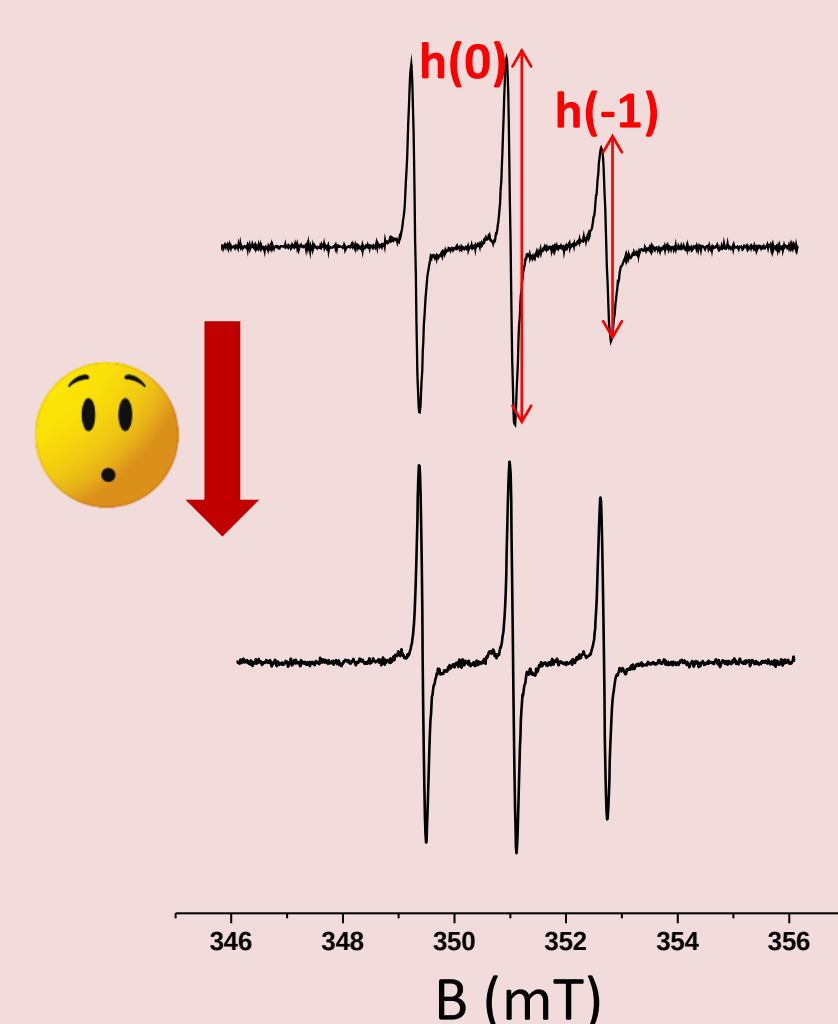
Objective: to study Tau/MTs interaction



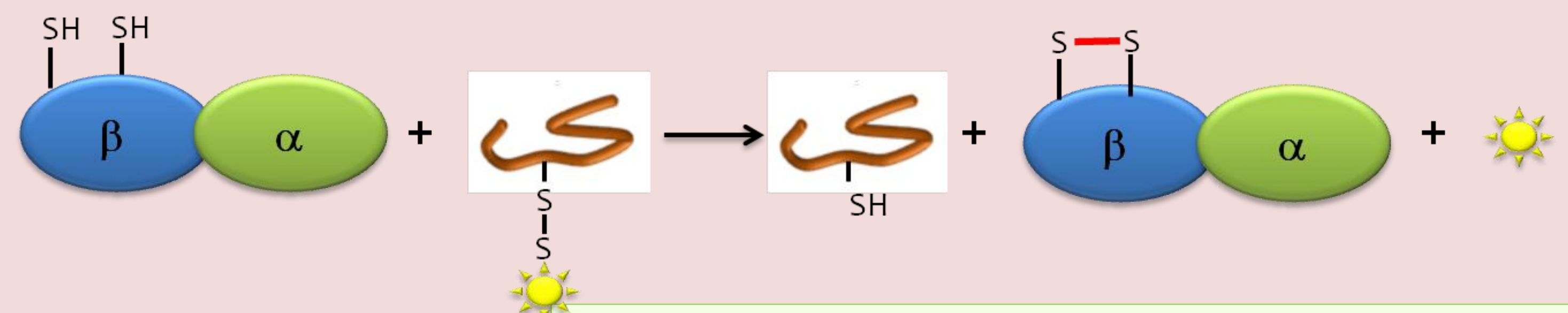
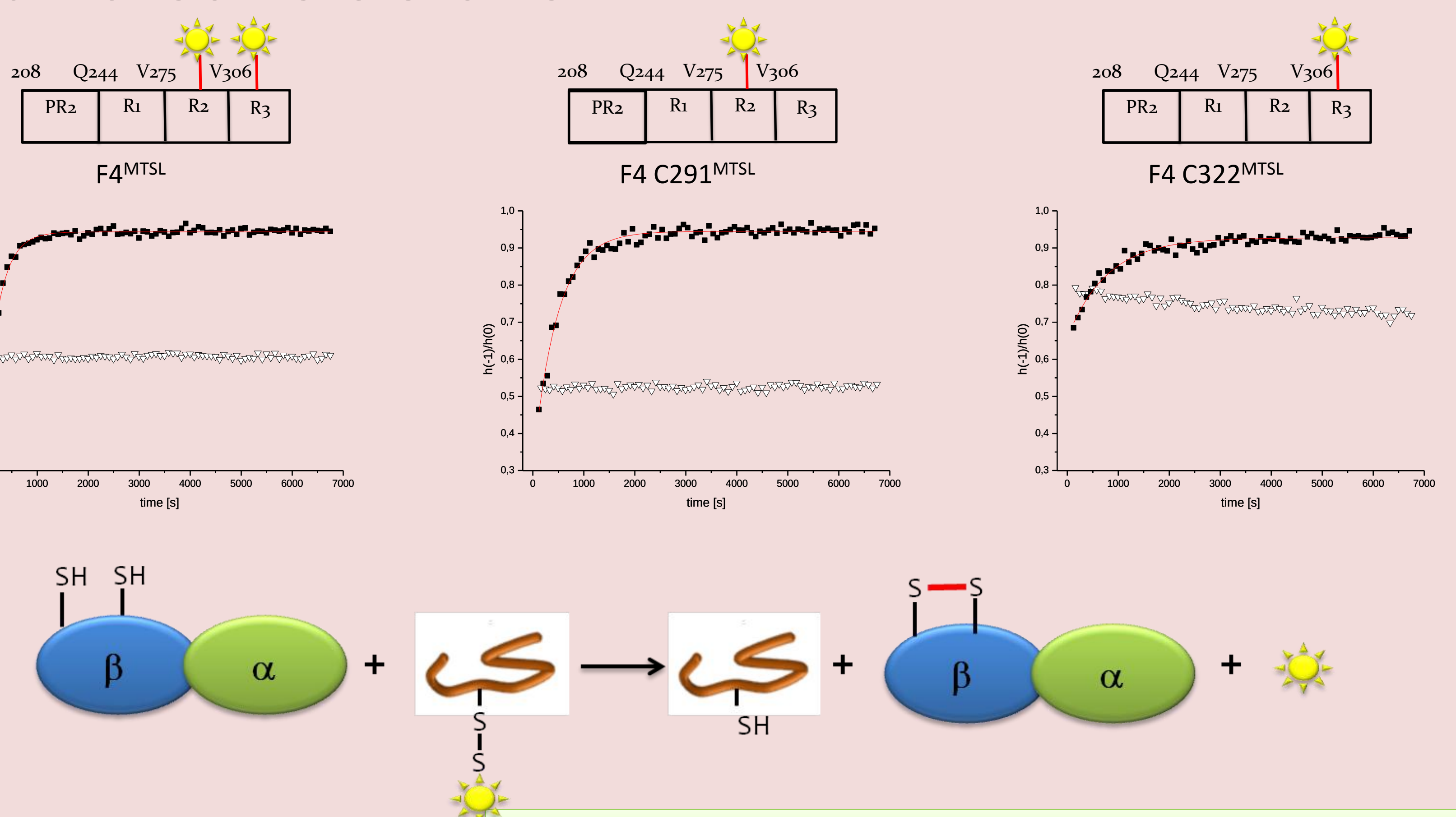
Results and conclusions



Sillen, A. et al. Biochemistry 2007

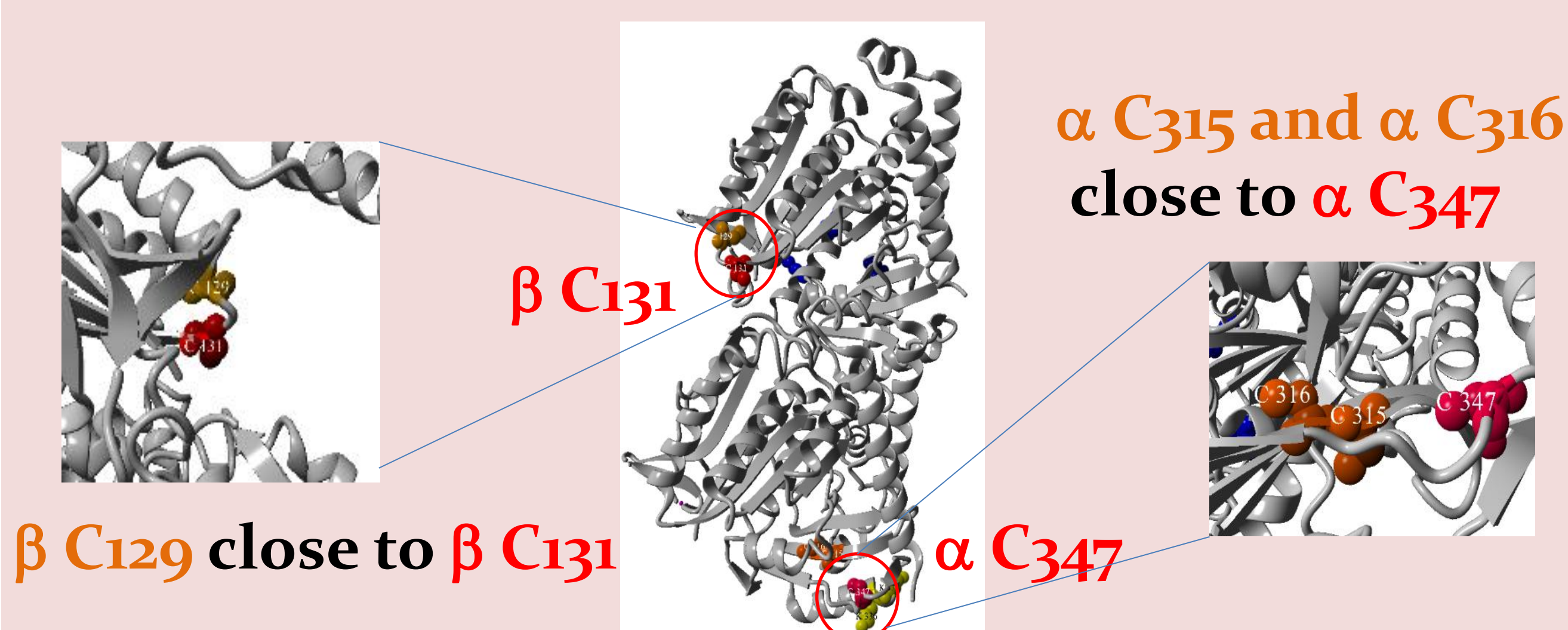


Slower kinetics when MTs are preformed

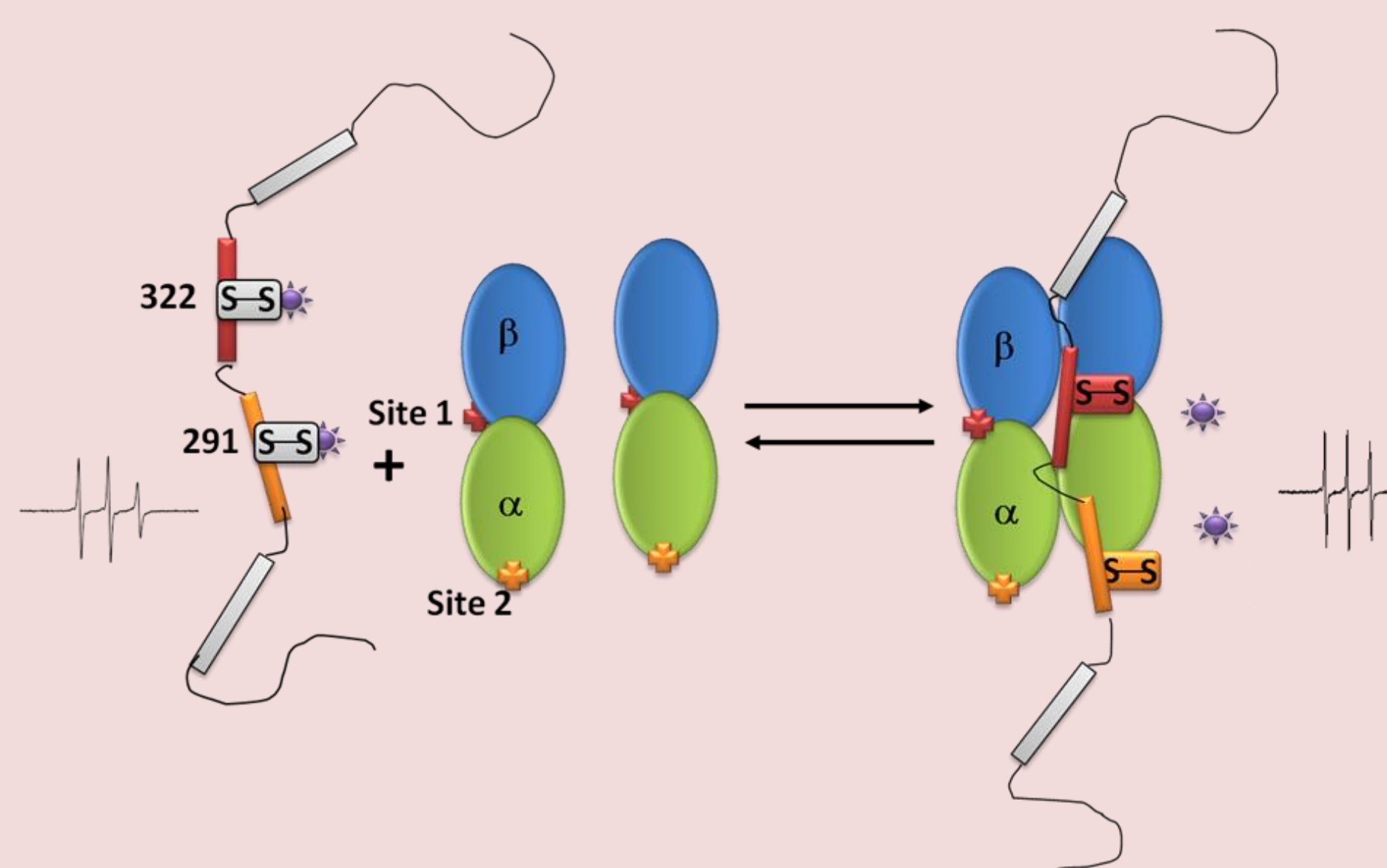


Unexpected release of the label upon interaction !!

Tau is able to perform a thiol / disulfide exchange with MTs



α -C347 and β -C131 are good candidates for Tau binding sites on MTs



- ∞ Ability of Tau to exchange a disulfide bridge with MTs
- ∞ Unusual use of SDSL-EPR spectroscopy approach
- ∞ Localization of 2 binding sites