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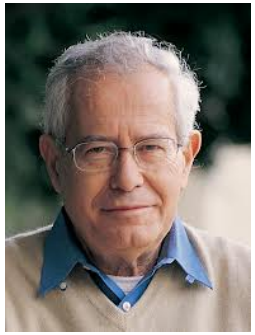
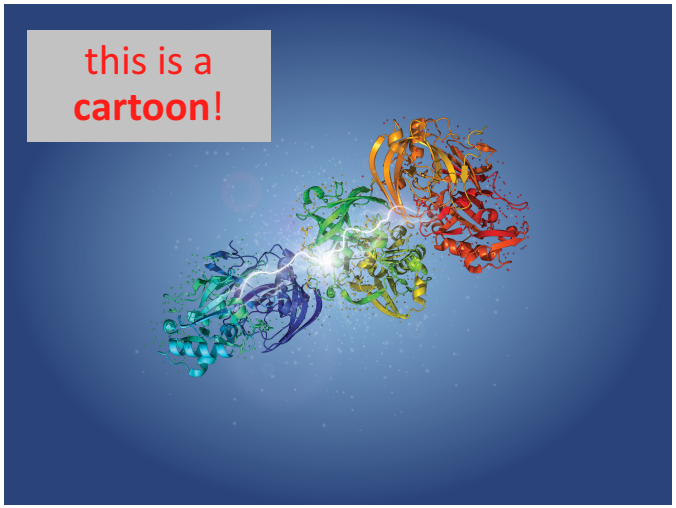
Biomolecular Electronics : Electron Transport across Proteins

with

Mordechai Sheves **&** **Israel Pecht**





Support

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Minerva Foundation, Munich
Israel Science Foundation
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Proteins are Soft Materials

→ "soft electronic biomaterials"

1st challenge in studying such electronic materials is to contact them electrically,

reproducibly, affecting them minimally.

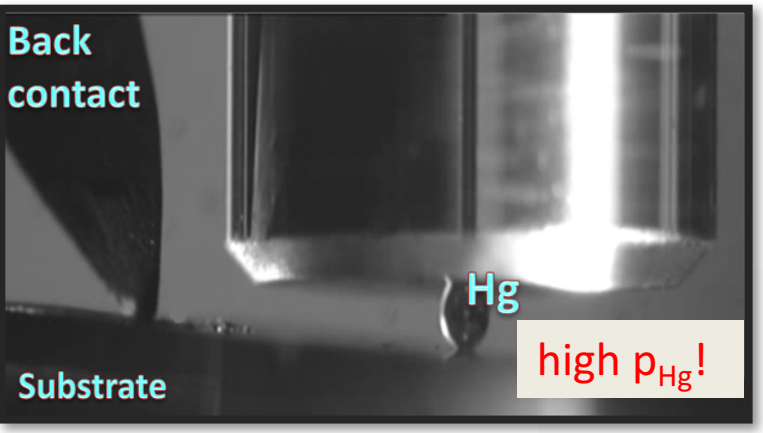
It is even harder to make contacts for

reliable current transport measurements

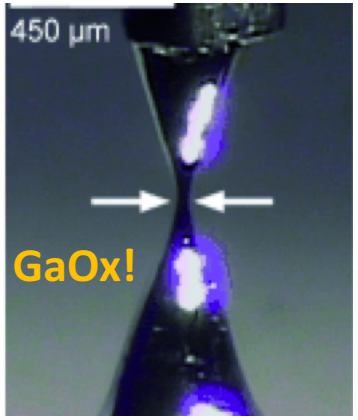
(i.e., beyond applying bias)

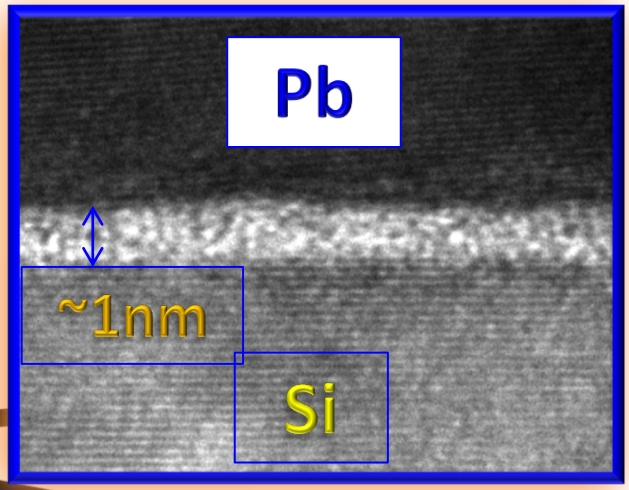
Non-destructive ***electronic*** contacts to soft matter:

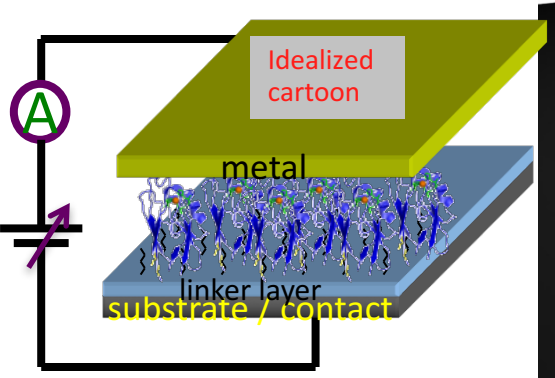
Hg or In-Ga

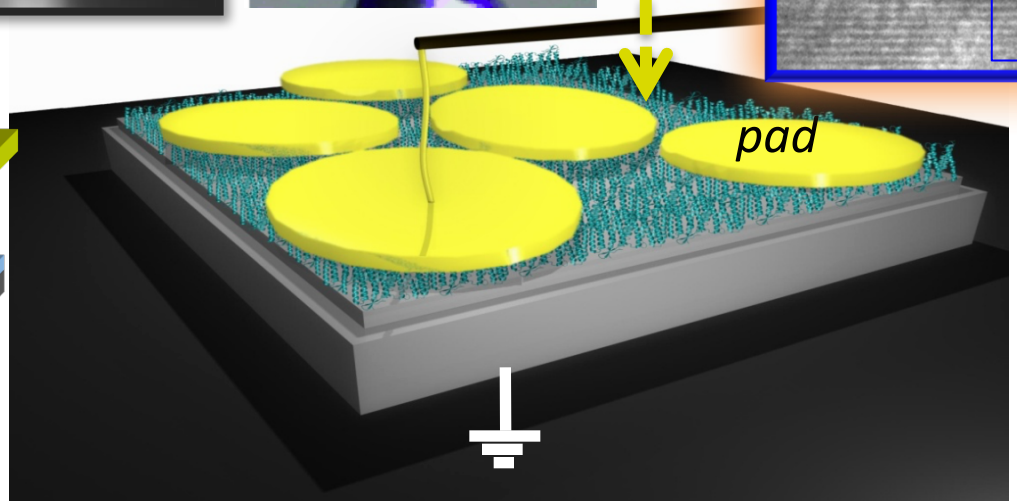


Hg drop







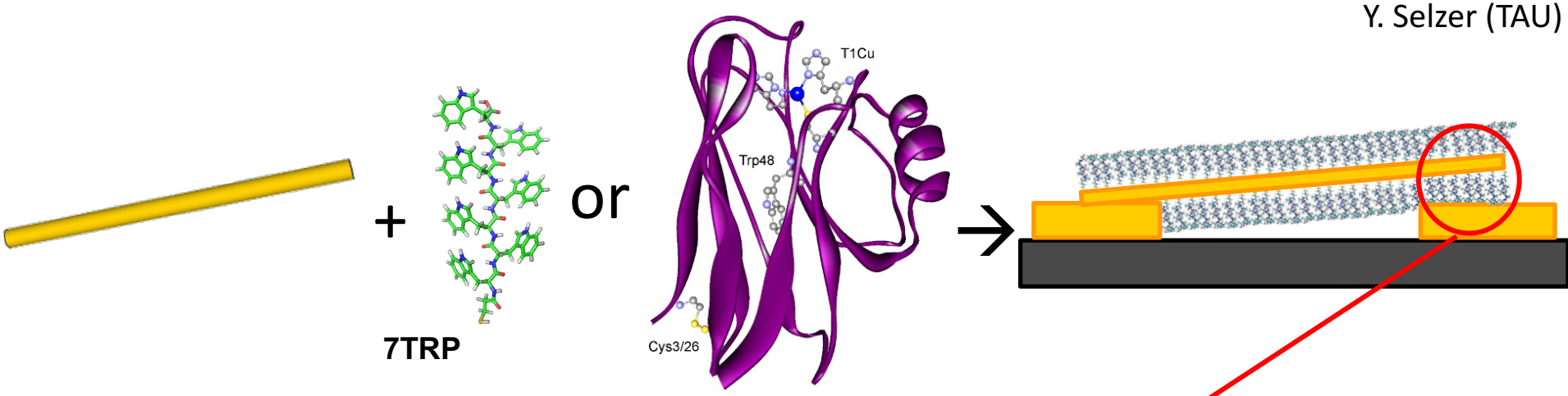


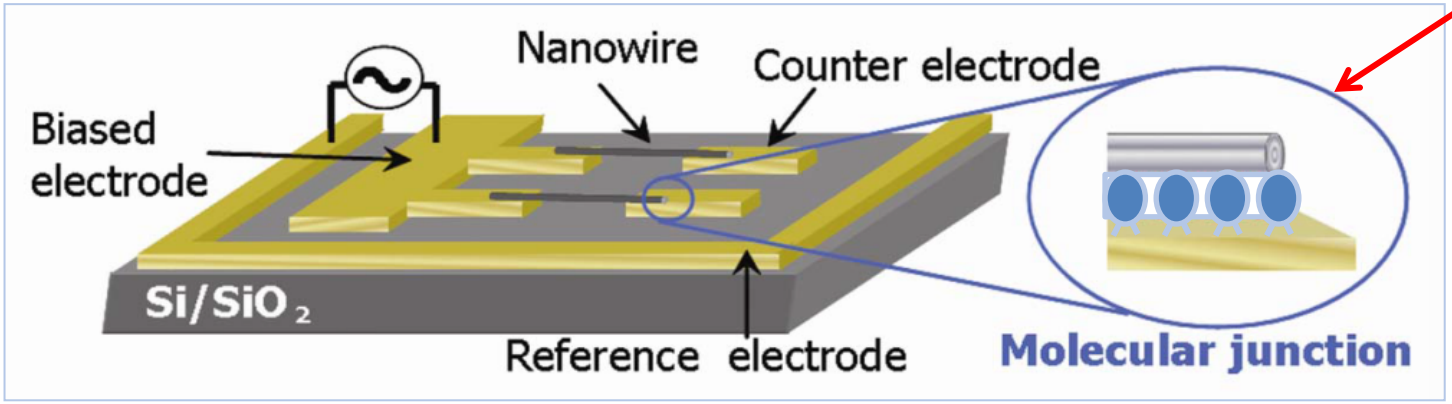
Lift-off float-on (LOFO) e.g.: Au, PEDOT-PSS

~0.2 mm^2 → ~ 10^6 – 10^7 (formally 10^{10}) proteins/contact

Dielectrophoretic assembly of nanowires
suspended nanowire technique

Y. Selzer (TAU)





> ~10% junctions "work"

Formally ~ ≥ 1000 (small) proteins / contact; ~ 0.05 μm^2

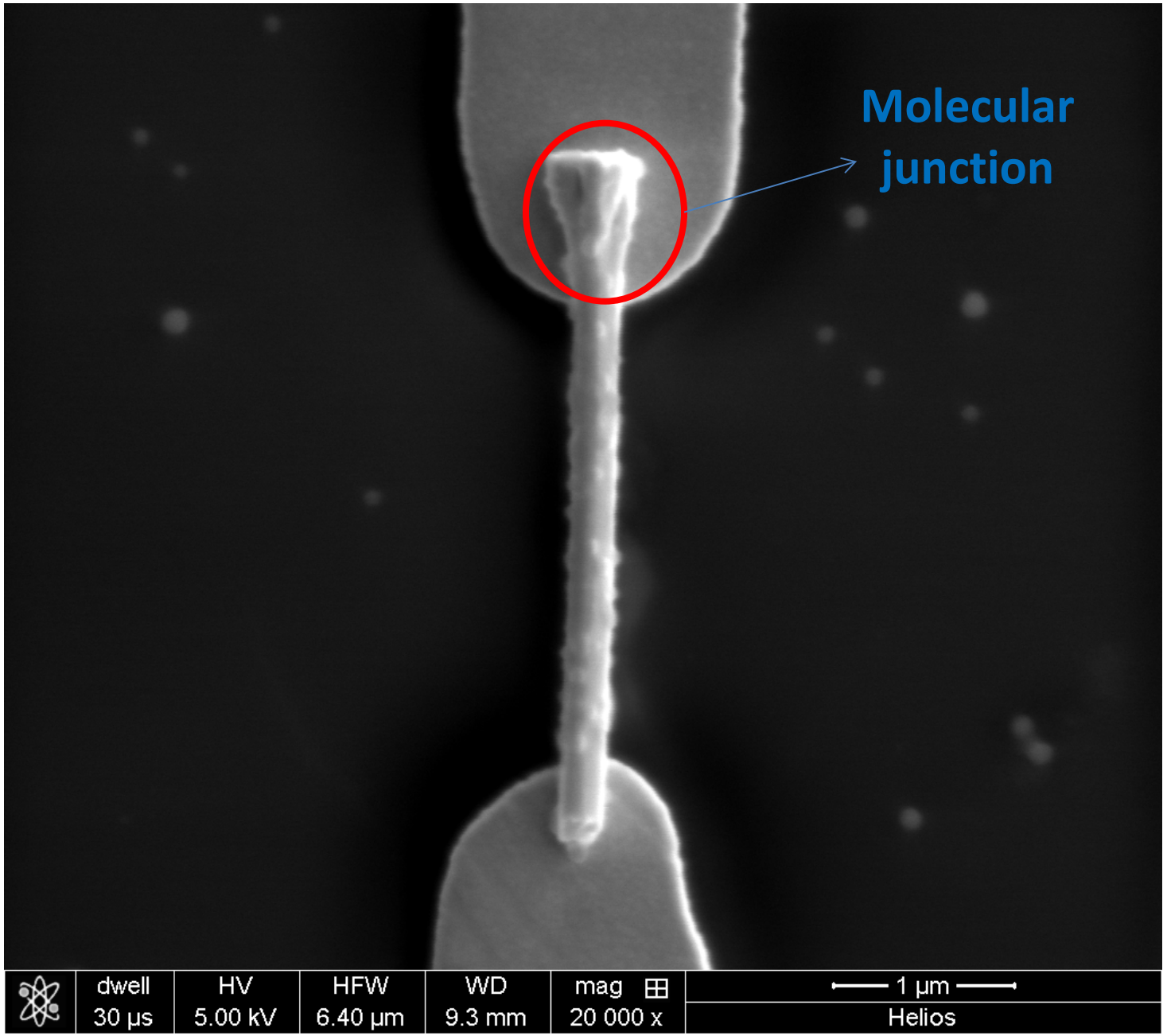
Noy, Ophir, Selzer, 2010, *Angew Chem.*
Sepunaru *et al.*, (2015) *JACS*

Yoram Selzer

&

Rani Arielly

(TAU)



We can also work with nanoscale contacts;

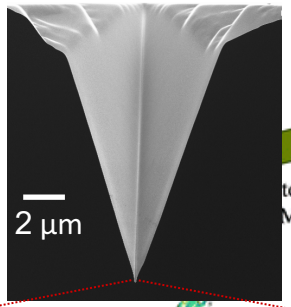
let's take a closer look at such experiment:

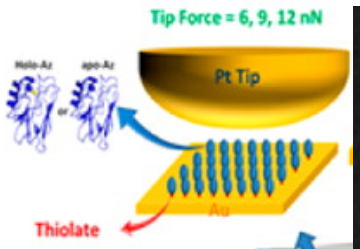
Nanoscopic

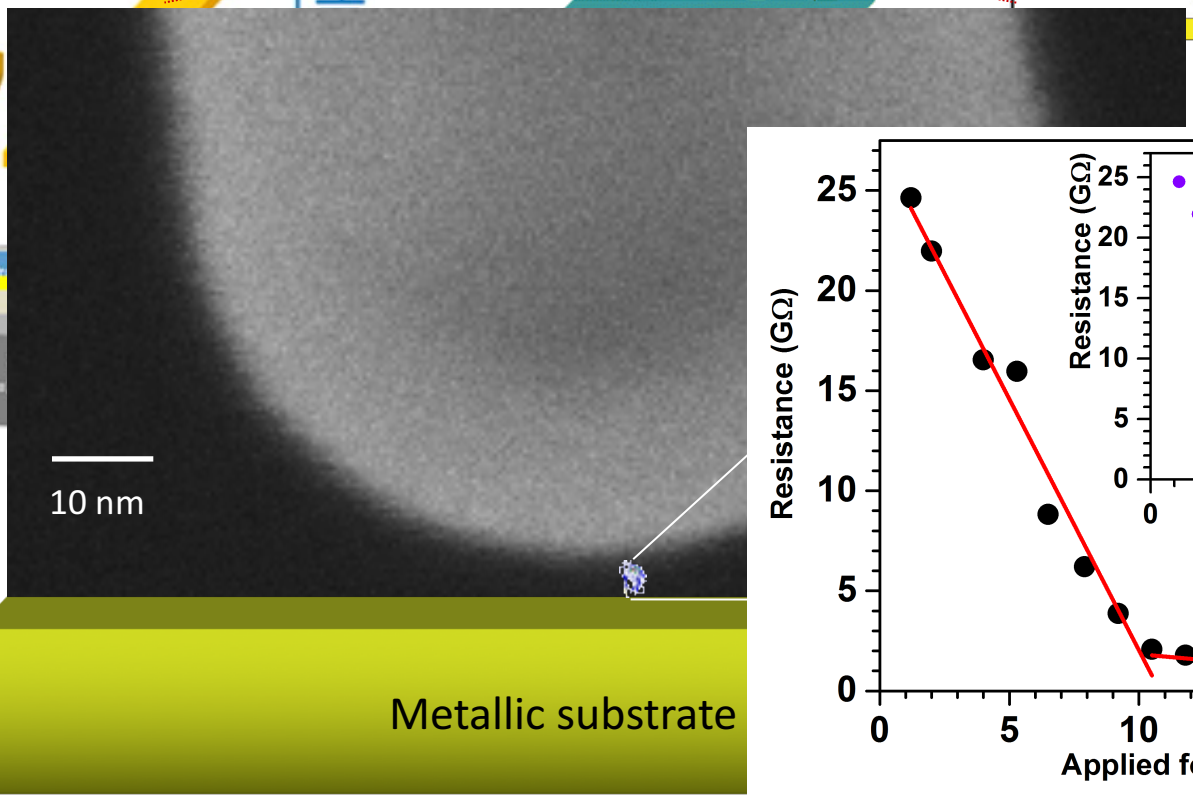
Idealized Cartoons!

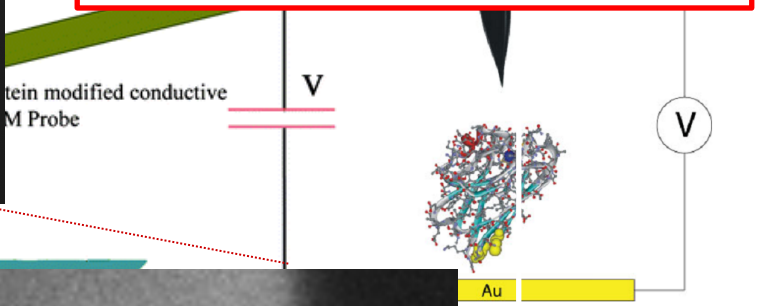
~10 –~ 50 proteins/contact ?

Geometrical contact area:
~ 300 - 500 nm^2









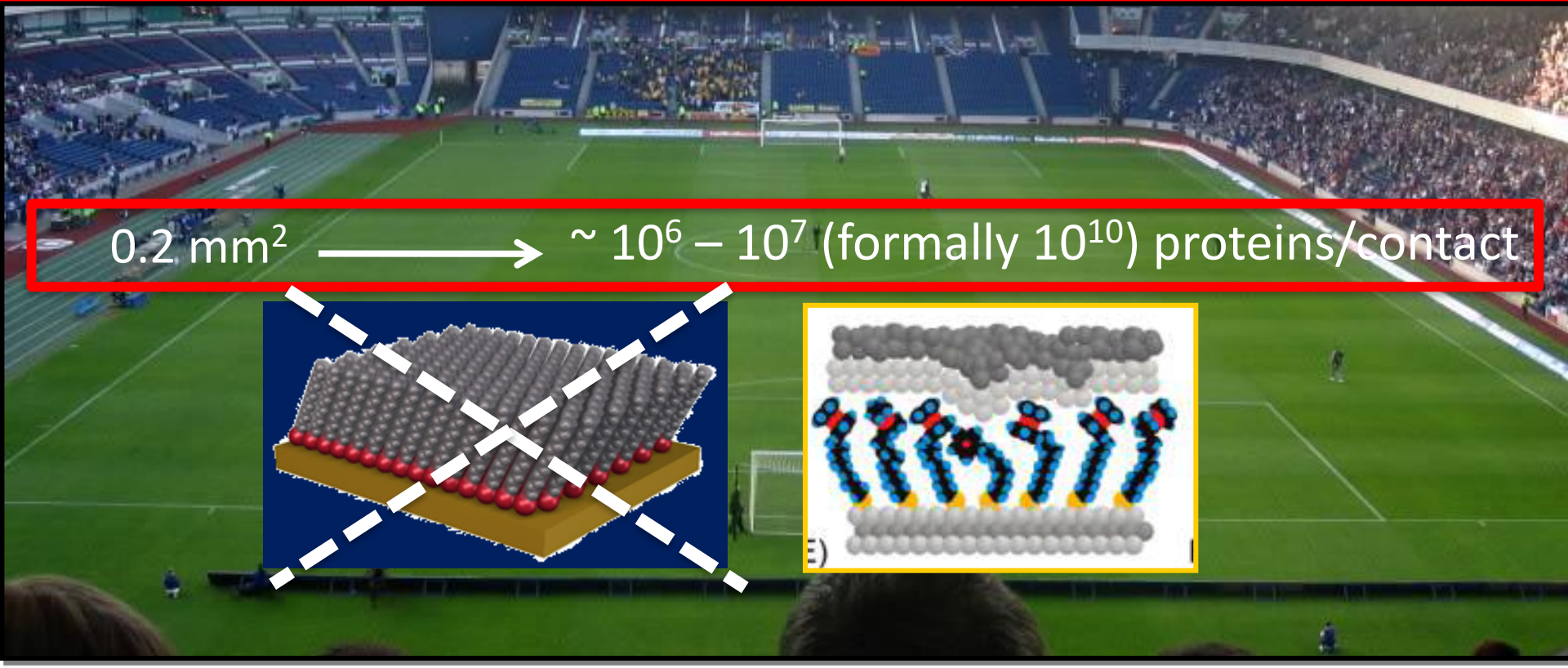
The Darker Side of Monolayer-based ML

intimate 10-20 μm^2 contact to a 10-20 nm^2 / protein monolayer?

like contacting each grass leaf (~5 cm^2) on 70×100 m^2 soccer field[*Akkerman*]

still, higher over-all currents → large measuring ability gain;

But, .. aren't you just measuring pinholes? Well, .. do the math, incl. probability, can vary top and bottom contacts, all with different roughness and still…)



Proteins (& peptides) are Soft Materials

→ "soft electronic biomaterials"

After learning how to contact them **electronically**, **reproducibly**, **reliably**, *affecting them minimally*

you find that (esp. as monolayers) they are

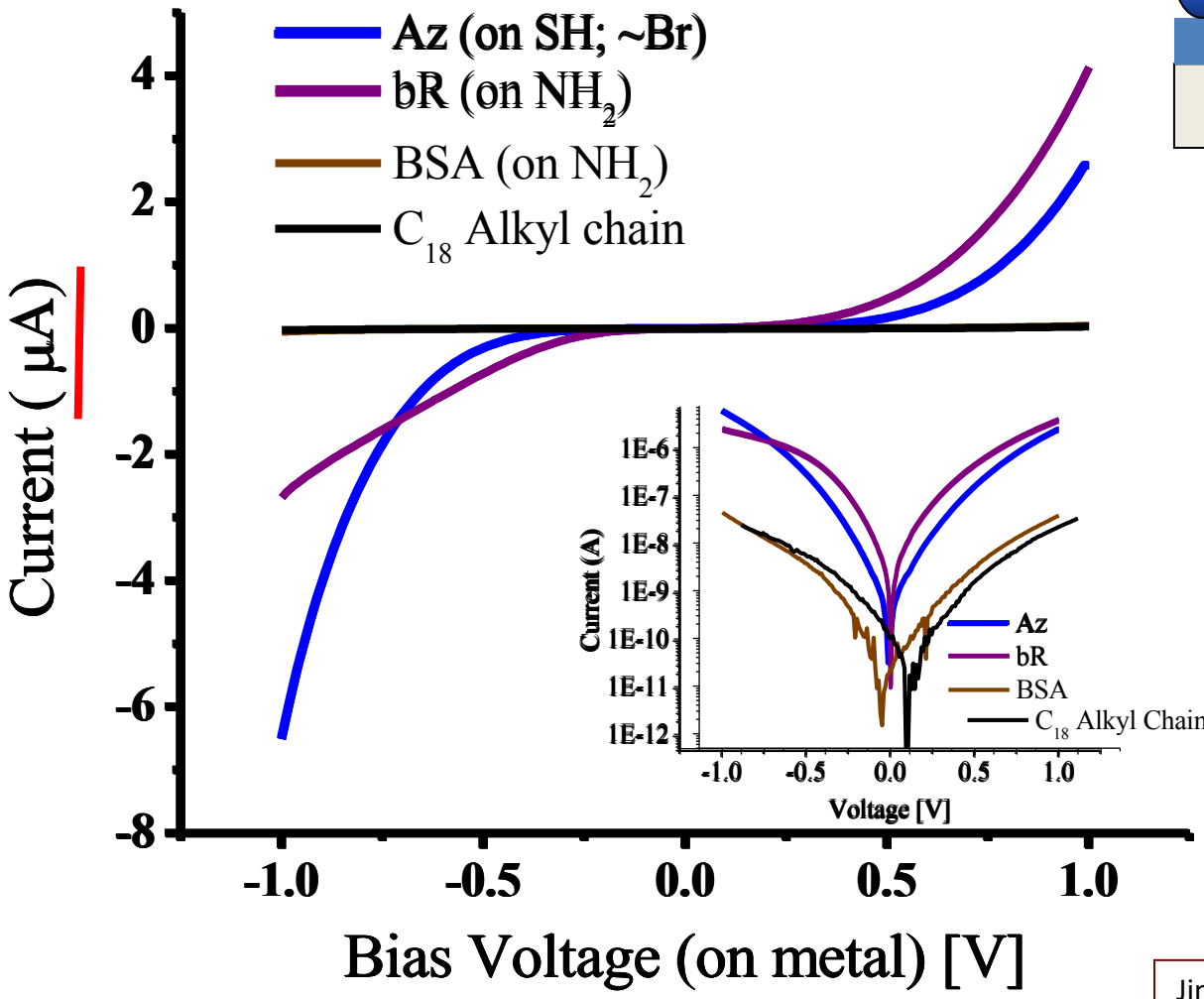
amazing (solid state-compatible) **electronic materials**

As yet ... it is not (so) clear why and how …

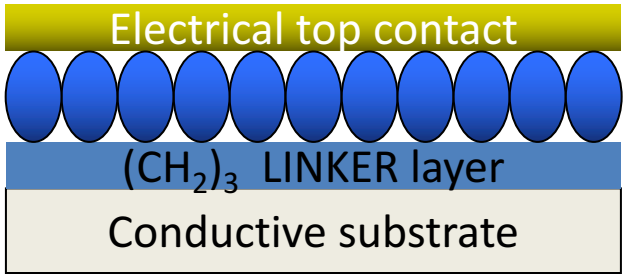
I-V characteristics

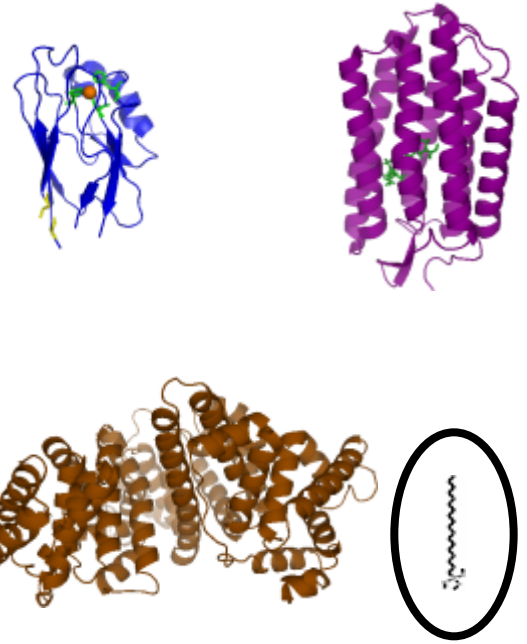
of protein junctions

Quite a few proteins "survive" *partial dehydration*



idealized cartoon





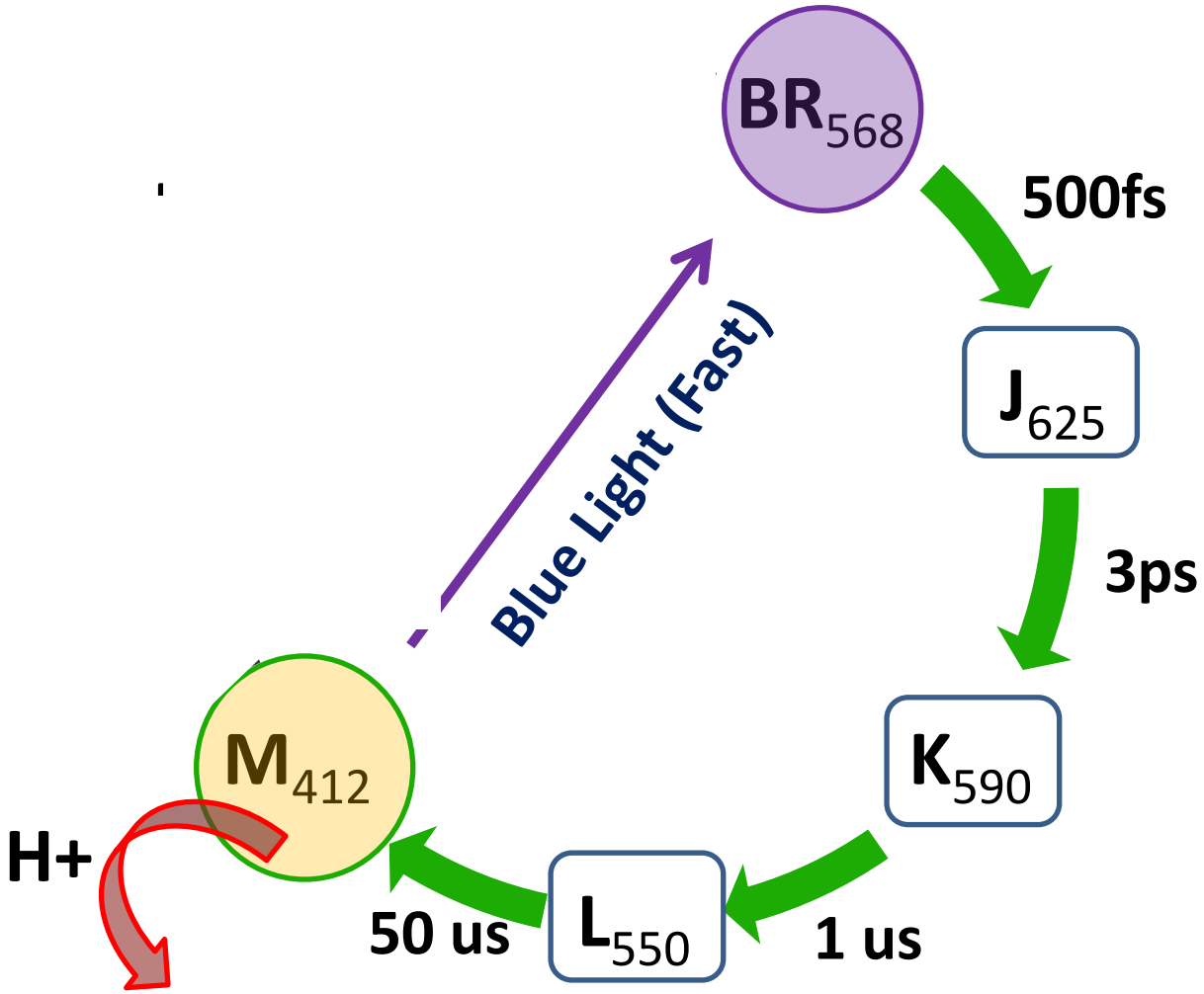
Jin et al., PNAS, 2006 …….. Ron et al, *JACS*. 2010

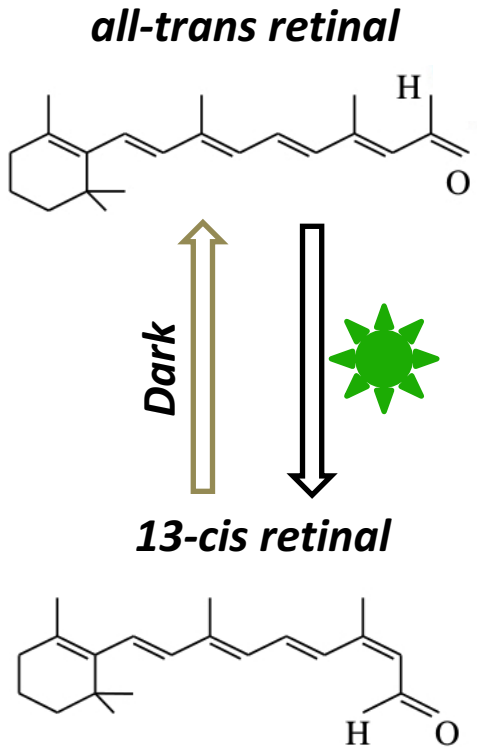
There are quite a few

Potential optoelectronic proteins

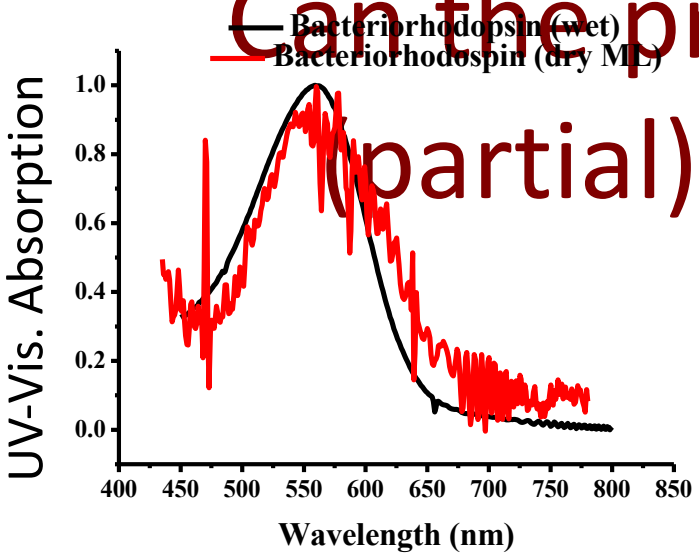
(PSI &II c.s., rhodopsins, LOV-ones [YtvA], ++)

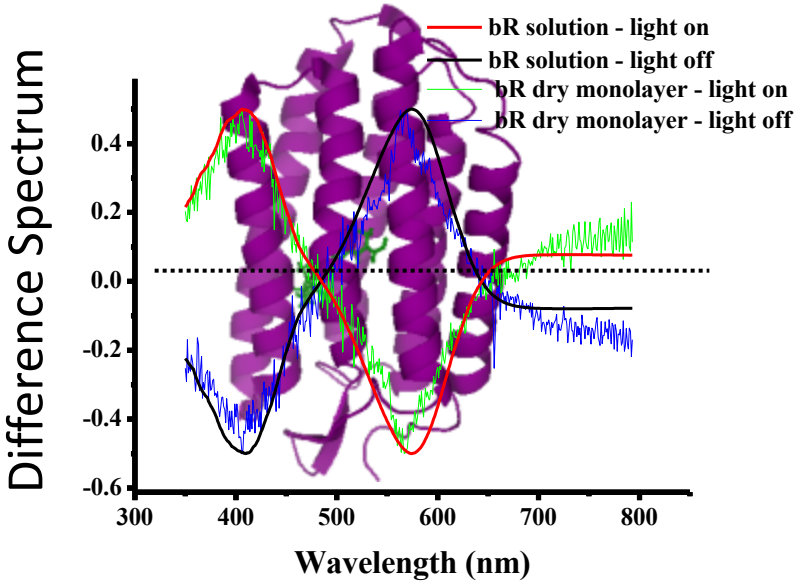
The bacteriorhodopsin PHOTOCYCLE, briefly…





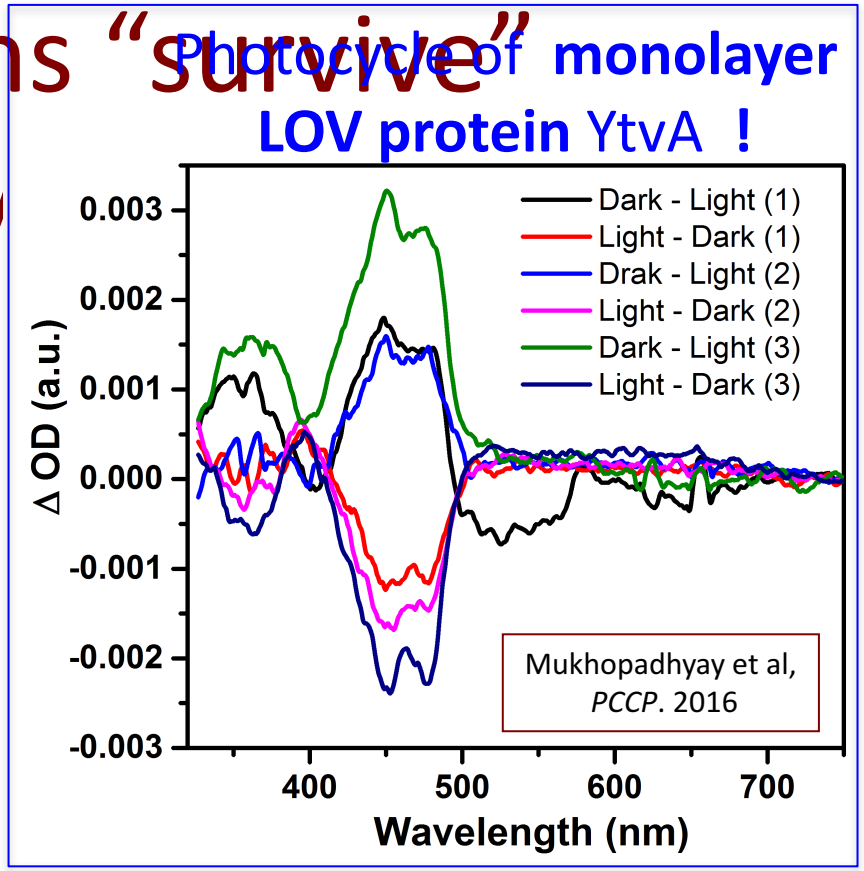
Can the proteins "survive" (partial) dehy

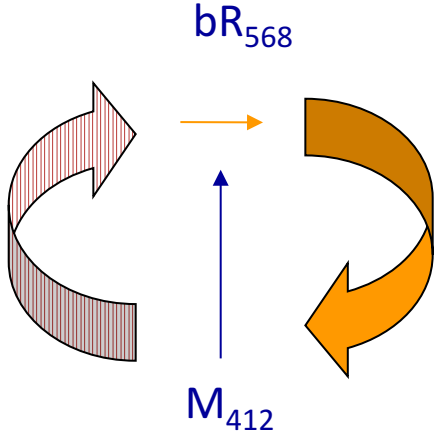




Bacteriorhodopsin (bR)

Photocycle of **monolayer LOV protein** YtvA !



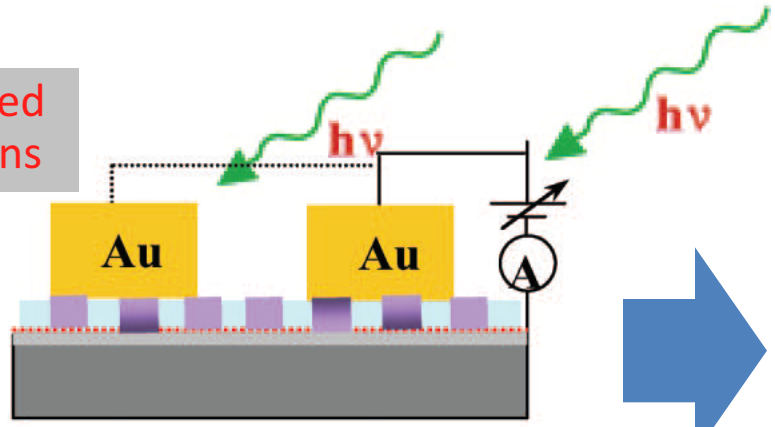


Ron et al, *JACS* 2010

Photo-conductance across Bacteriorhodopsin (bR) monolayer

Green irradiation – *Formation of metastable M-state*

Idealized cartoons



APTMS-modified Al/AlO_x
bR monolayer junctions

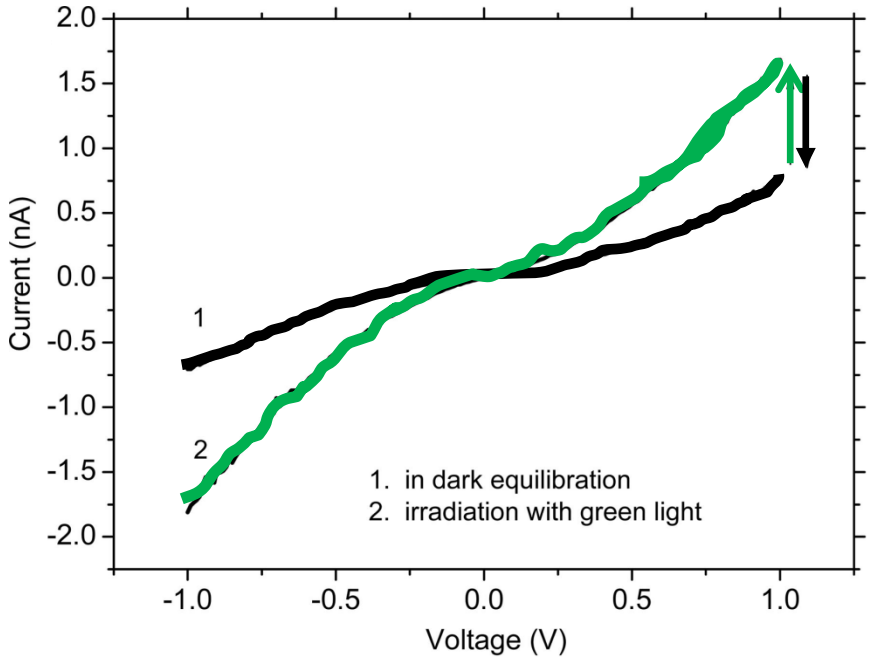


Photo-effect originates from M-state accumulation (conformation change)

Jin *et. al.* PNAS **2006** , *103*, 8603

Can the proteins “survive” (partial) dehydration ?

For Azurin we’ll show:

- @ denaturation temperature irreversible decrease in conductance *

and

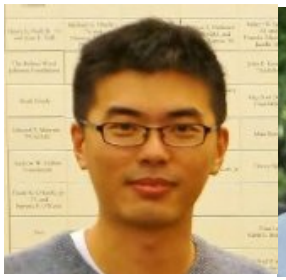
- In-situ vibrational spectroscopy shows Amide 1 & 2 of proteins in the junction**

* **L. Sepunaru** *et al*., (2011) *JACS*
** **X. Yu** *et al.,* (2015) *ACS Nano*

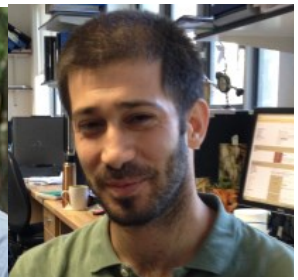
Some protein electronics **riddles**

at least some proteins

- **conduct electrons "too efficiently"**
- **show tunneling-like behaviour over "too long" distances**

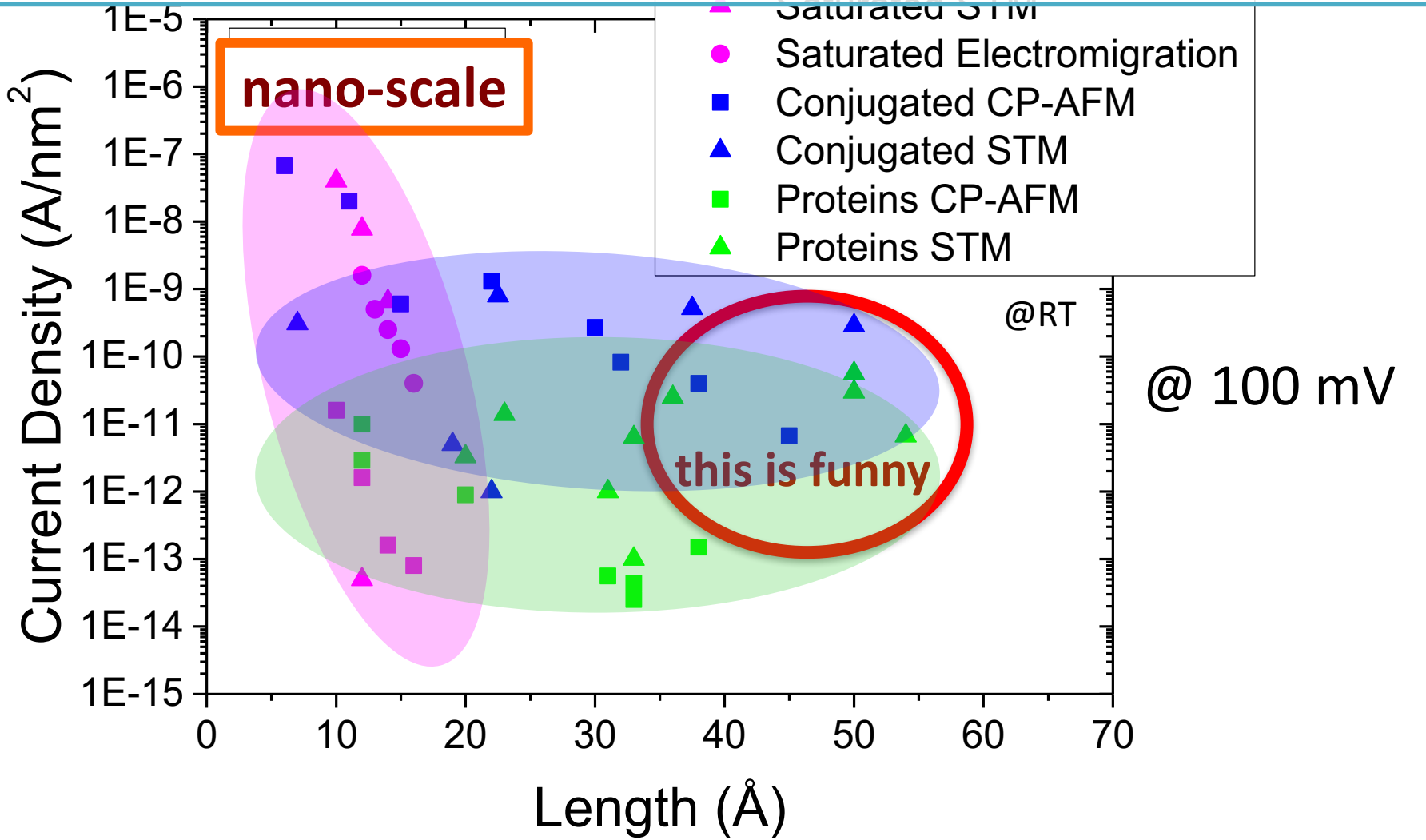

Xi Yu


Jerry Fereiro


Lior Sepunaru

Protein *vs.* conjugated & saturated molecule conduction

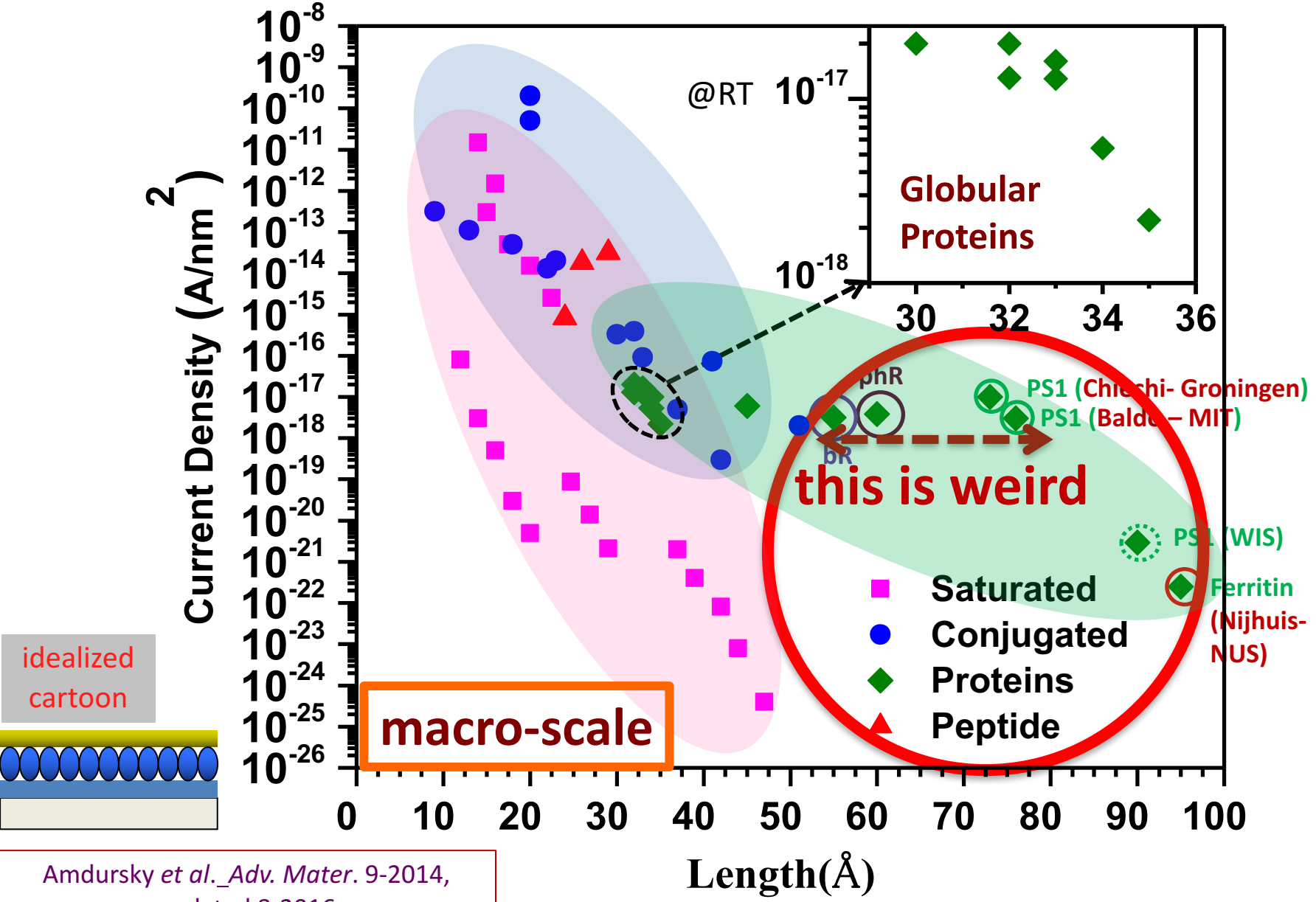
While long-range Electron Transport involving several proteins is known (respiration, photosynthesis), *across one protein …? …,* ***that's different***



Amdursky *et al.*, *Adv. Mater*. 9-2014

Protein *vs.* conjugated & saturated molecule conduction

Log(current) @ 0.1 V vs. length plot of macro-scale measurement data



Amdursky *et al._Adv. Mater*. 9-2014, updated 8-2016

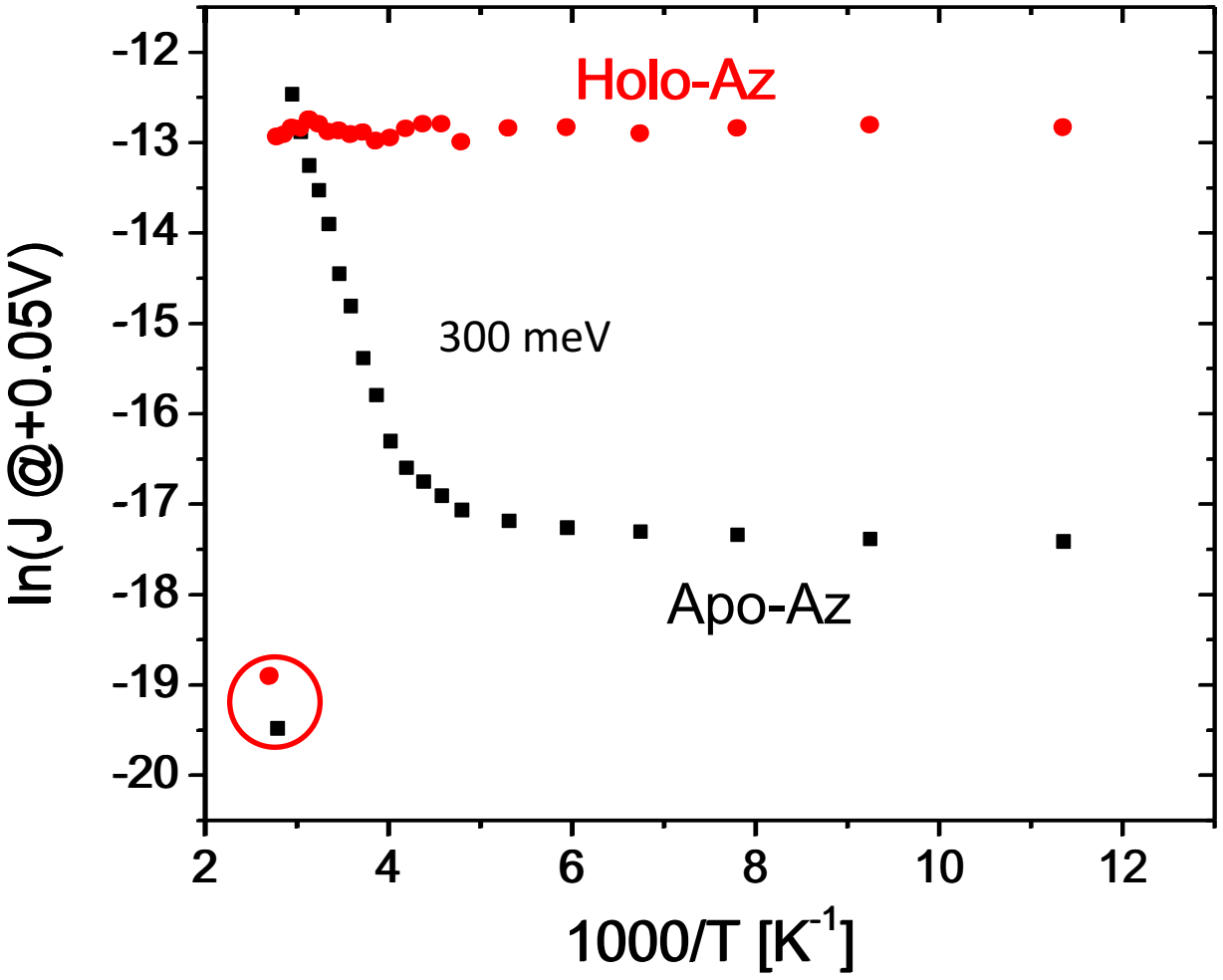
Some protein electronics **riddles**

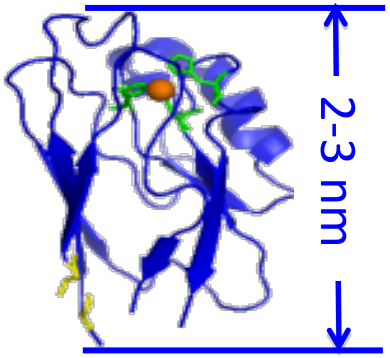
at least some proteins

- conduct electrons “too efficiently”
- **show tunneling-like behaviour over “too long” distances**

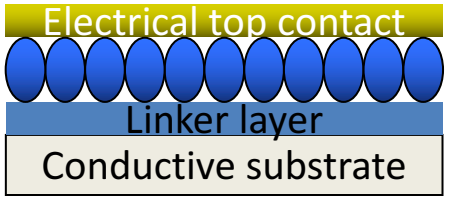
Temperature dependence of "solid state" Electron Transport, ETp, of Azurin

Cu ion removal



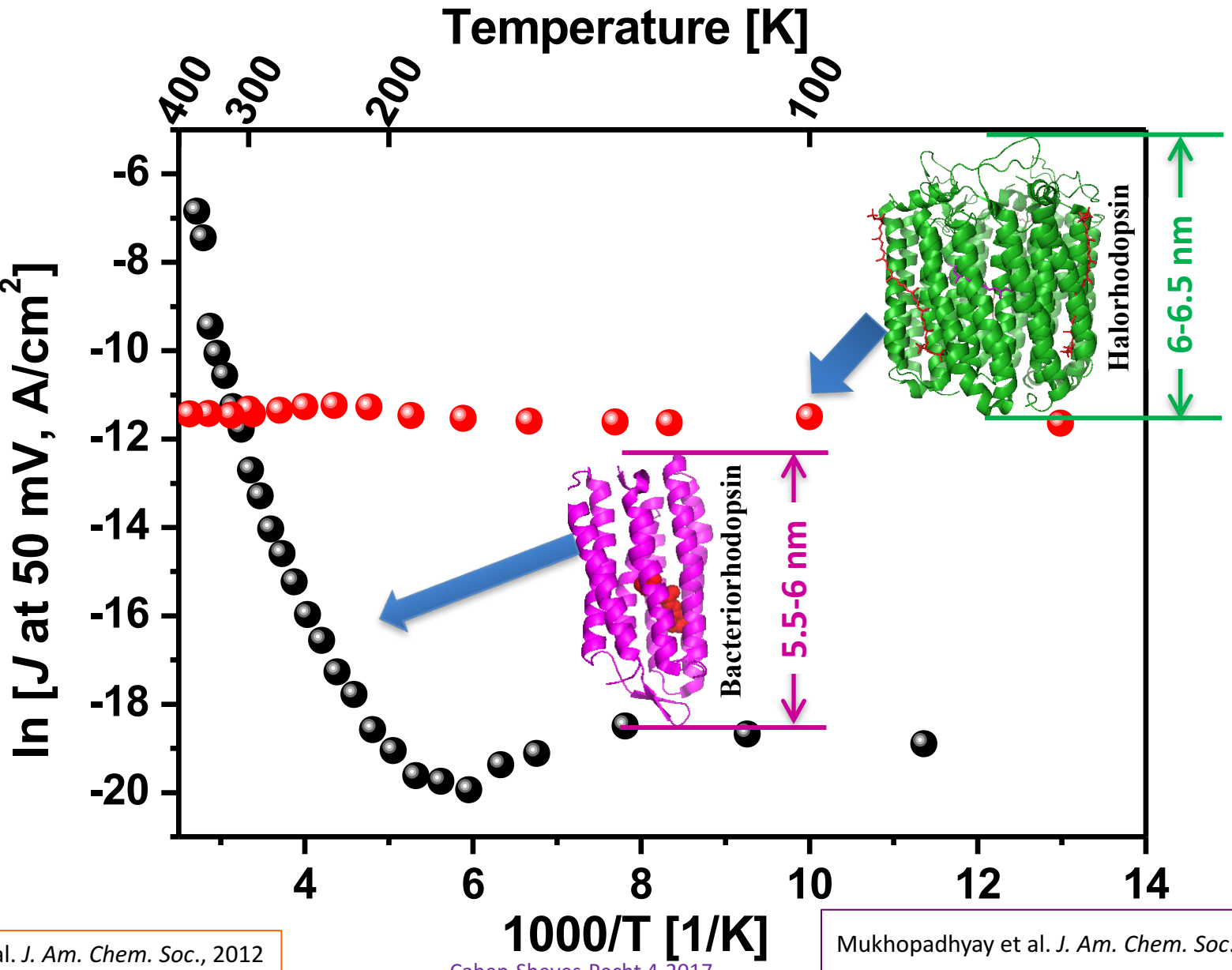


idealized cartoon



Sepunaru et al., *J. Am. Chem. Soc*. 2011

Temperature-dependent ETp of bacteriorhodopsin (bR) and Halorhodopsin (phR)

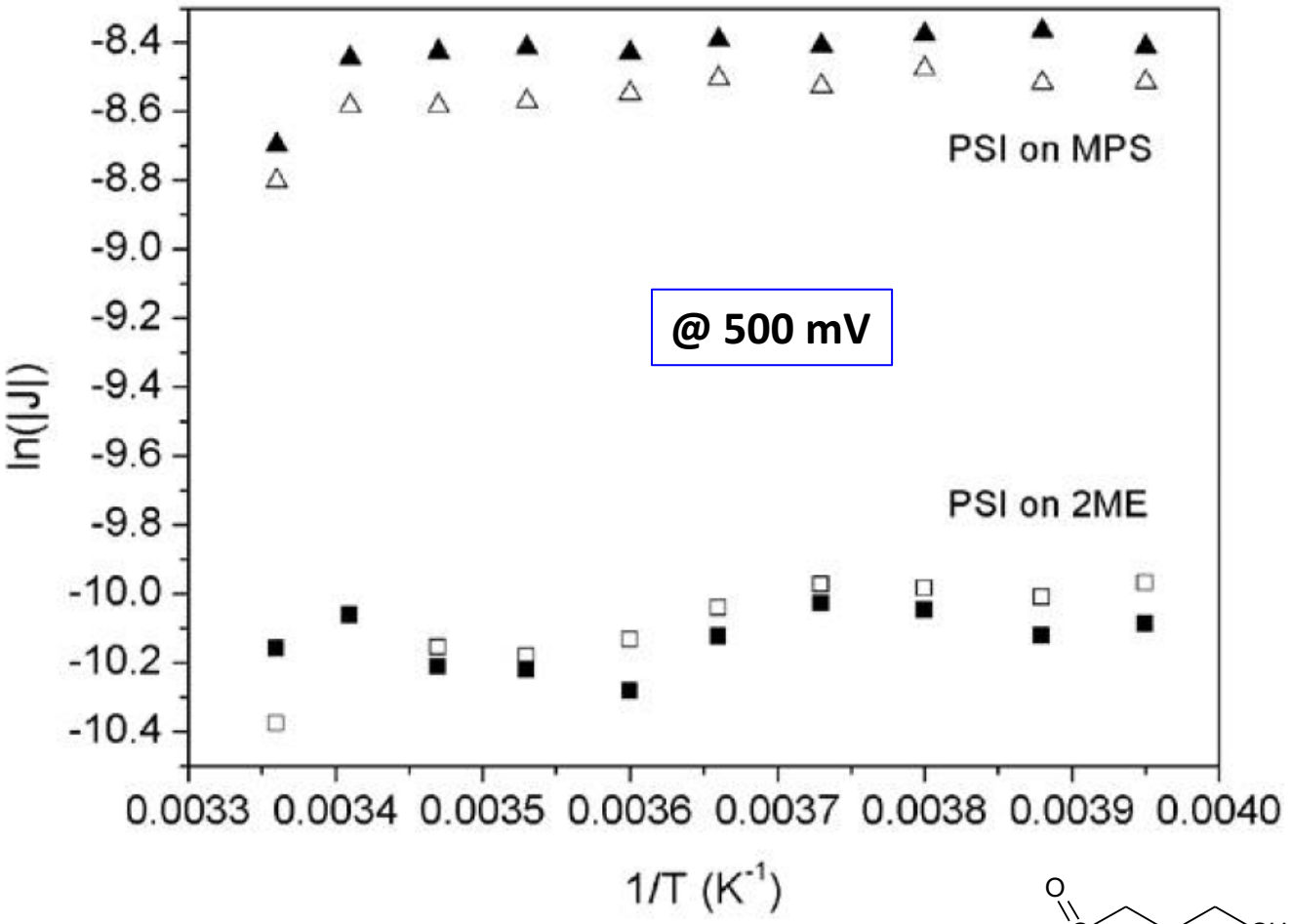


Sepunaru et al. *J. Am. Chem. Soc.*, 2012

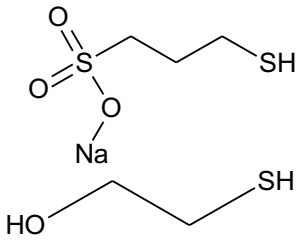
Mukhopadhyay et al. *J. Am. Chem. Soc.* 2015

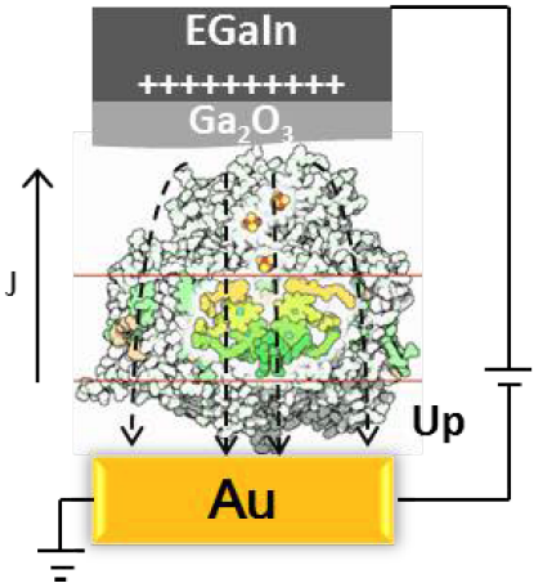
Not for publication

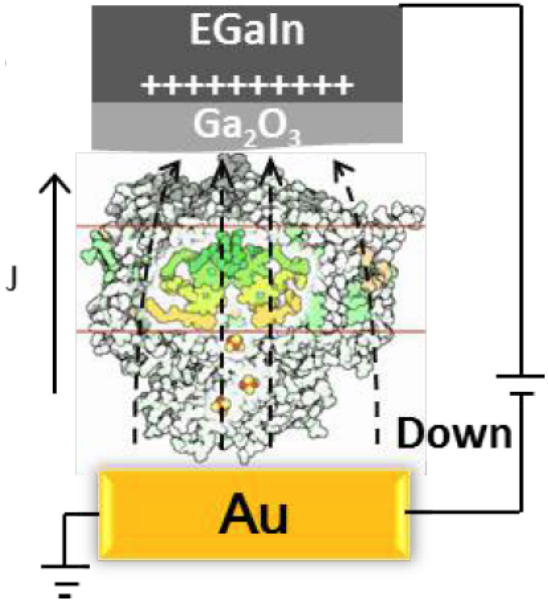
Temperature (in)dependent ETp of PS I (~ 7 nm !)



MPS = sodium 3-mercapto-1-propanesulfonate
2ME = 2-mercaptoethanol







Castañeda Ocampo, O. E. *et al.*. *J. Am. Chem. Soc.*. (2015)

Rates of Intramolecular Electron Transfer in Ru(bpy)$_2$(im)(His83)-Modified Azurin Increase below 220 K

Lars K. Skov*,‡

Department of Chemistry, University of Copenhagen DK-2100, Copenhagen, Denmark

Torbjörn Pascher,†,§ Jay R. Winkler,*,§ and Harry B. Gray*,§

Beckman Institute, California Institute of Technology, Pasadena, California 91125

Received July 28, 1997

But , ...

nihil novi sub solem

אין חדש תחת השמש

Ecclesiastes קהלת 1.9

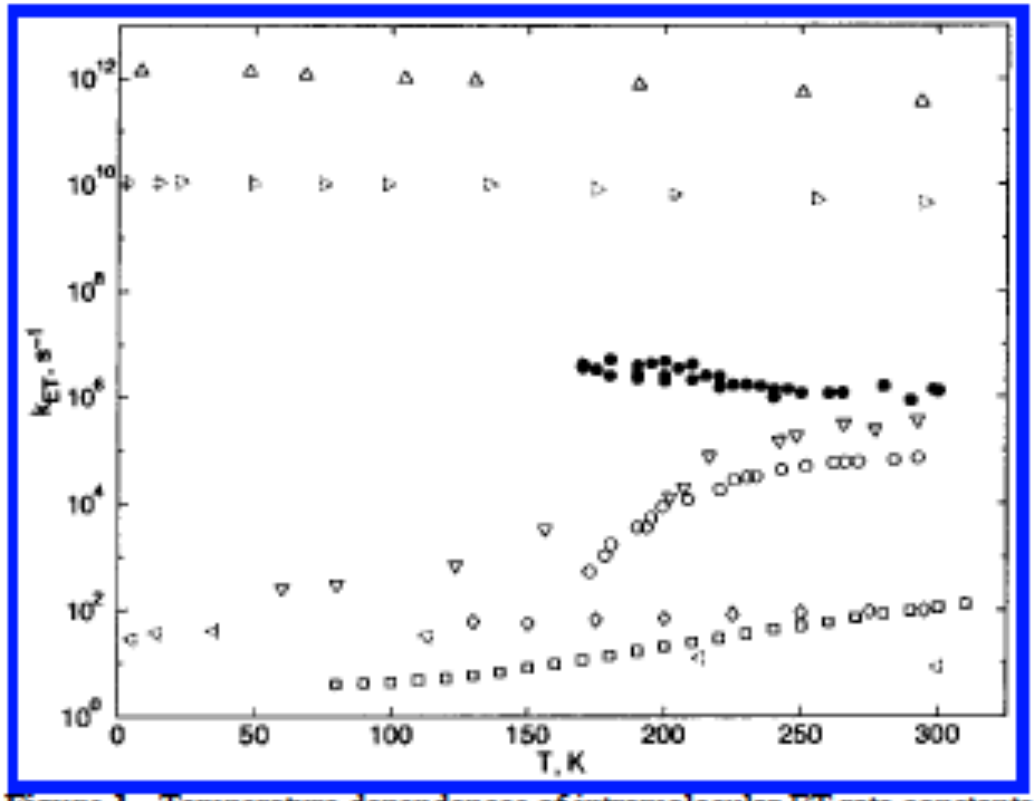
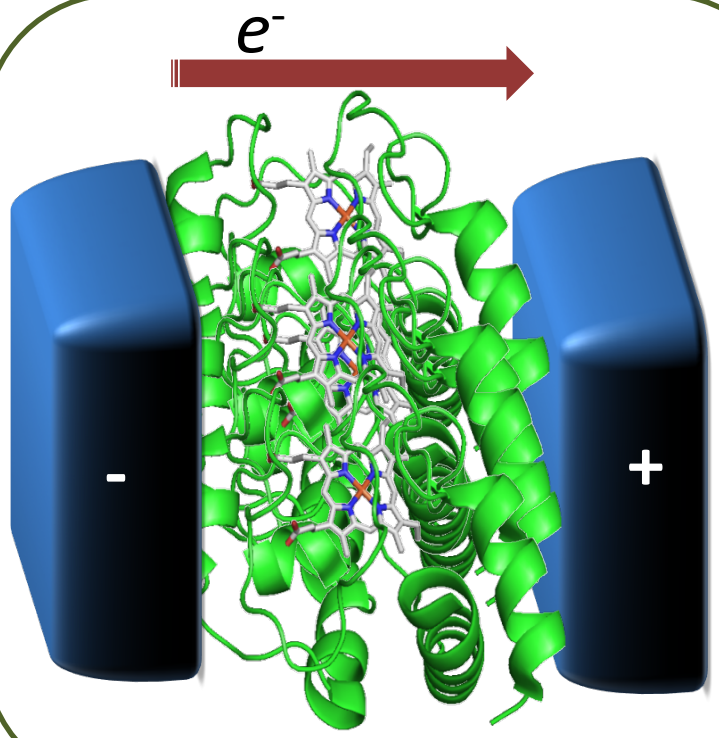


Figure 1. Temperature dependences of intramolecular ET rate constants in proteins: (△) primary charge separation in the *Rhodopseudomonas viridis* photosynthetic reaction center;[1] (triangle pointed to the right) ET from reduced bacteriopheophytin to quinone in the *Rhodopseudomonas sphaeroides* reaction center;[2] (▽) oxidation of cytochrome *c* in *Chromatium vinosum* reaction centers;[4] (triangle pointed to the left) reduced quinone to oxidized special pair ET in the *Rps. sphaeroides* reaction center;[3] (□) ET from triplet-excited Zn-porphyrin (*ZnP) to a ferriheme in a metal-substituted hybrid hemoglobin;[5] (◇) ET from a cyanoferroheme to a Mg-porphyrin radical cation in a metal-subsituted hybrid hemoglobin;[6] (○) *ZnP→Ru(NH$_3$)$_5$(His48)$^{3+}$ ET in Zn-substituted myoglobin;[8] and (●) Cu^+→Ru^{3+} ET in Ru(His83)-azurin.

Electron transfer vs. Electron transport

Cytochrome C is an Electron Transfer (ET) protein

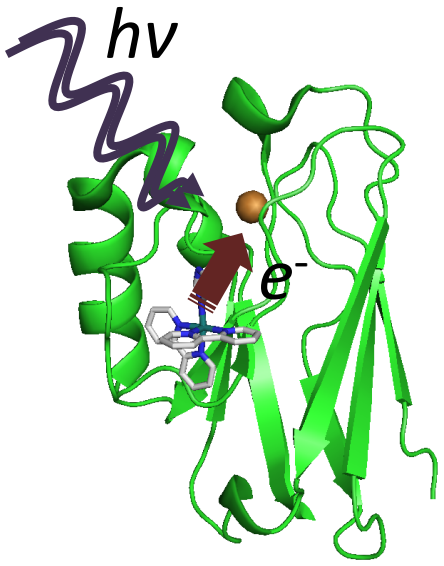
ETp



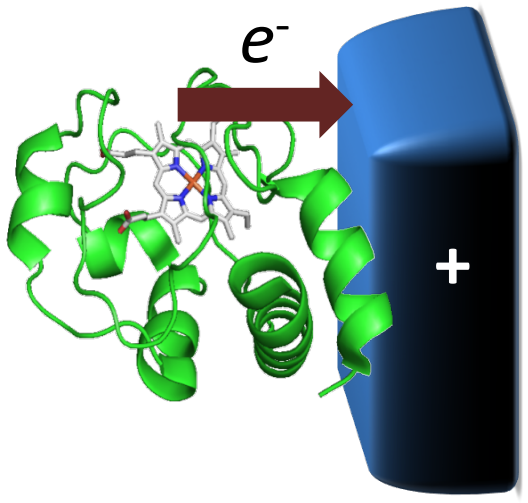
in solid state!

Biomolecular electronic junction

ET



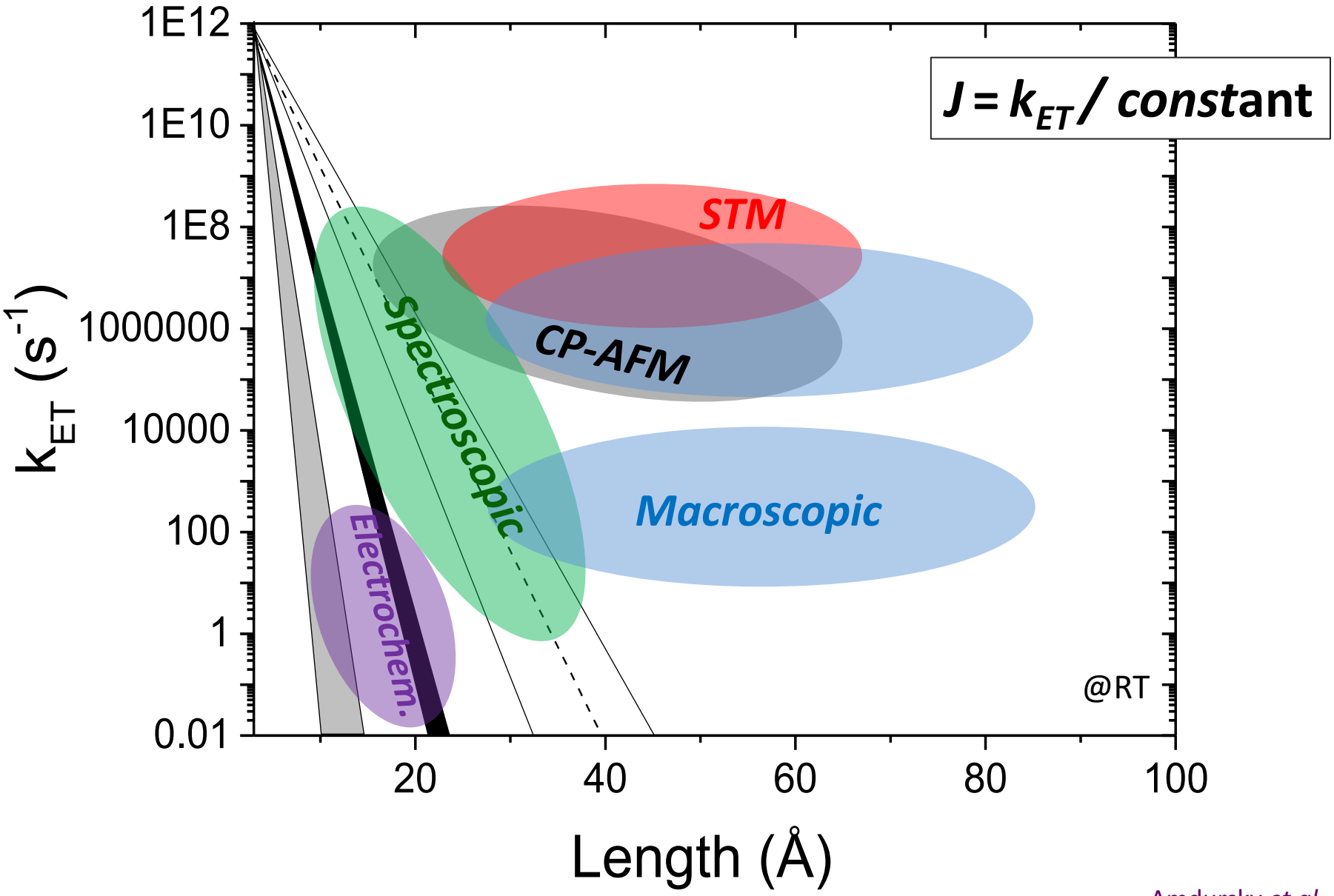
Spectroscopy



Electrochemistry

in solution

N. Amdursky et. al. (PCCP 2014)



Amdursky *et al.*,
Adv. Mater. -2014

So, what do we know that we don't understand?

Protein monolayers conduct, length-normalized @ RT, as conjugated molecules **or** (*esp. membrane proteins*) even better

Some proteins show *temperature – independent* current transport, even across 6-10 nm

➡ **How** do electrons cross proteins?

Possible Electron Transfer (ET) and ETp mechanisms

Superexchange-mediated Tunneling

Thermal fluctuations

→ degenerate D & A levels

→ Tunnel along bridge

Gray, Winkler et al.

Hopping

→Incoherent transport

→Localized charge hops

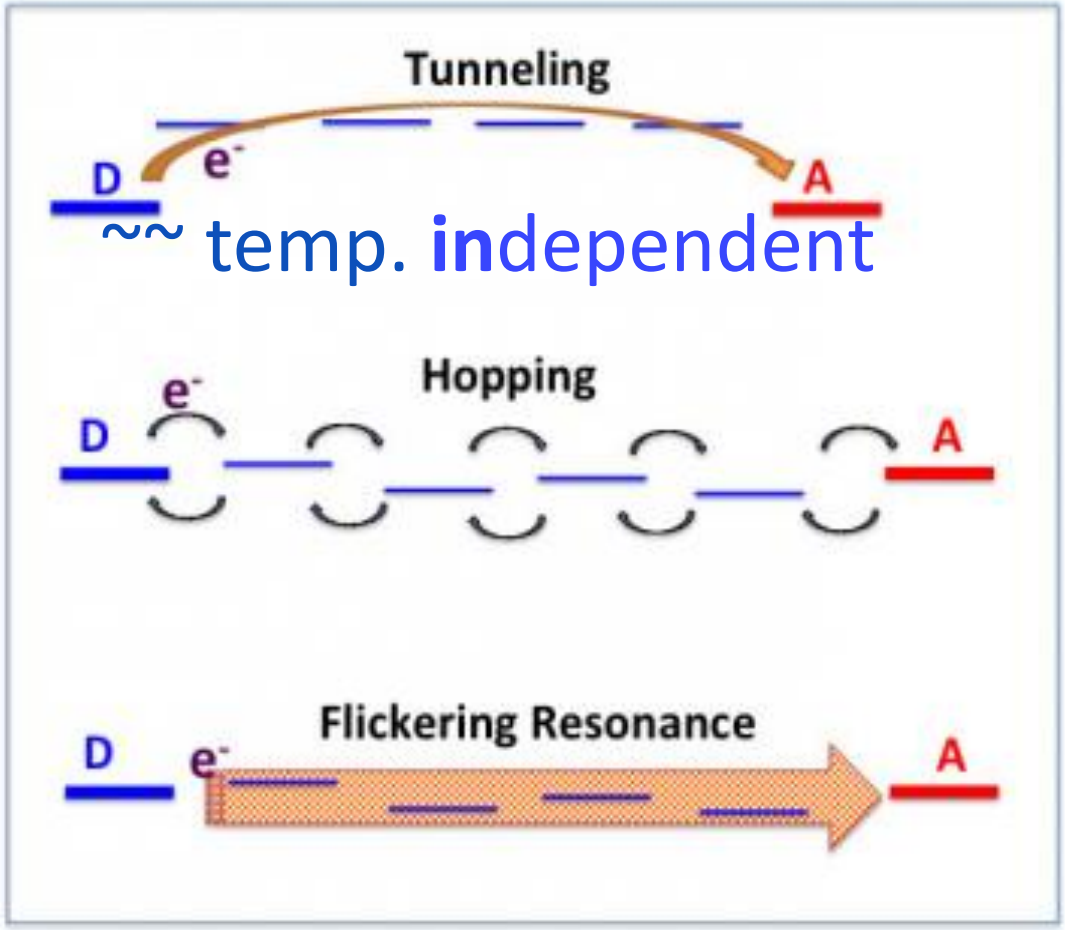
between consecutive sit

Flickering resonar

→ Redox sites in chain m

in/out of resonance

Beratan, Skourtis



from J. Blumberger, *Chem. Rev.*, 2015

Possible Electron Transfer (ET) and ETp mechanisms

Proteins (and peptides) are Soft Materials;

ergo: *structural dynamics → energetic disorder*

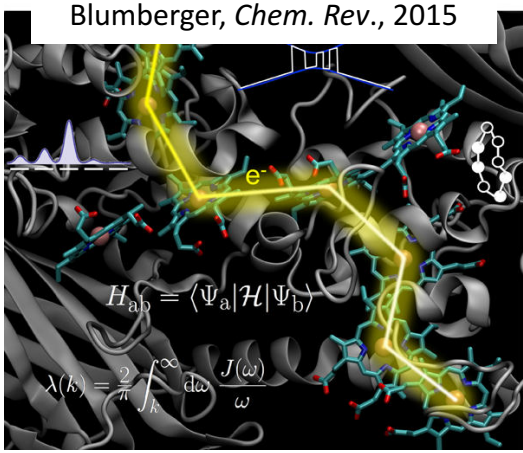
Charge Transfer in Dynamical Biosystems, or

The Treachery of (Static) Images

Beratan *et al.*, *Acc. Chem. Res.*, 2015

these are not tunnelling paths, but CARTOONS !

Quantum path for ET in Protein
Blumberger, *Chem. Rev.*, 2015



Some questions that define what we don't know

- **How** do electrons cross proteins?
- What controls electron transport in & **across** proteins?

What controls Electronic Transport **in** and **across** proteins ?

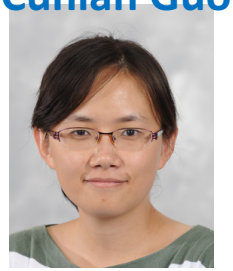
H-bonds ✔

cofactors ✔

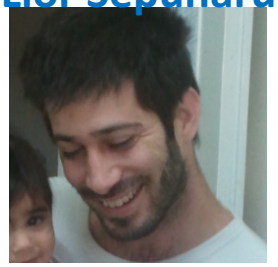
peptide backbone ?

contacts (✔)

Cunlan Guo



Lior Sepunaru

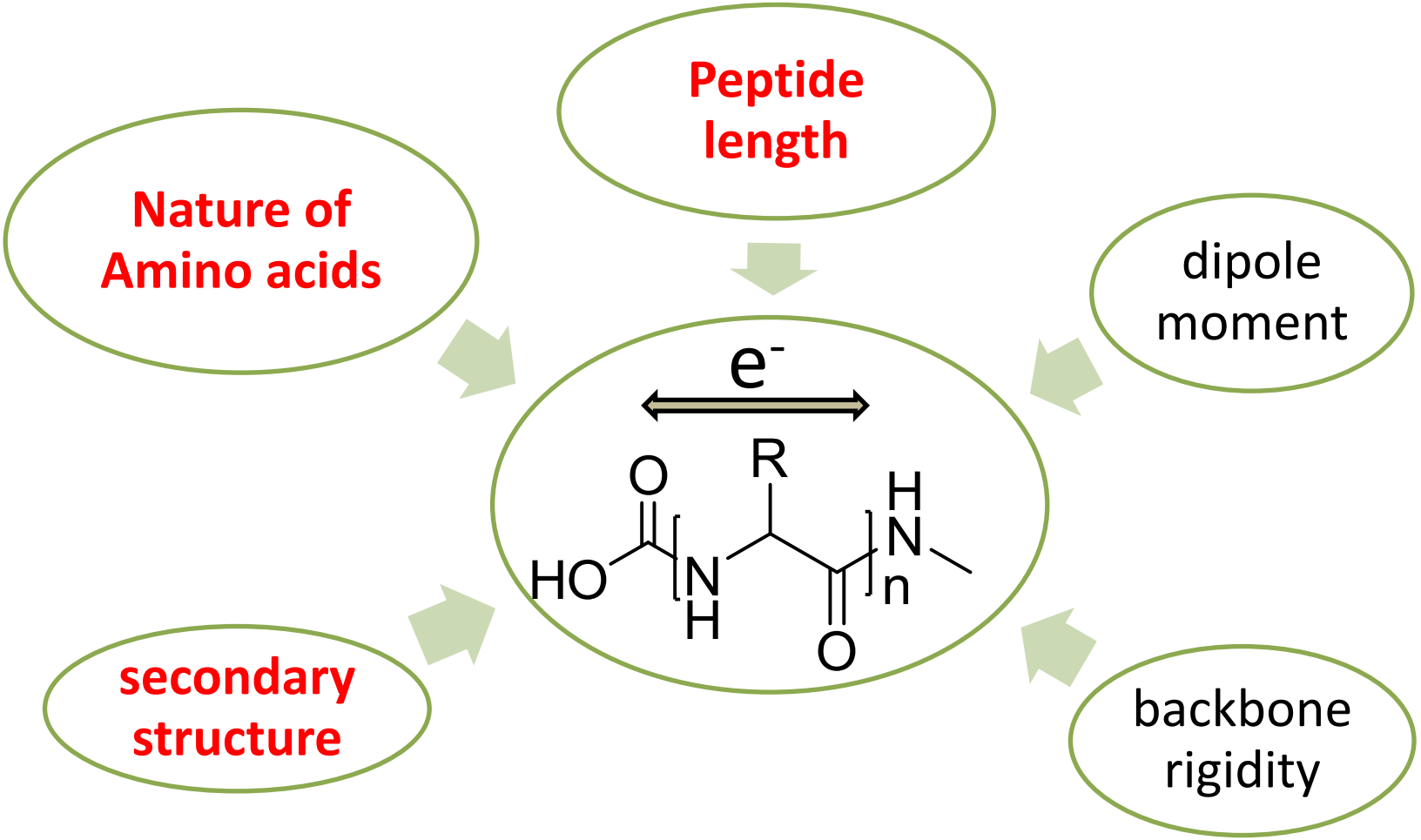


with

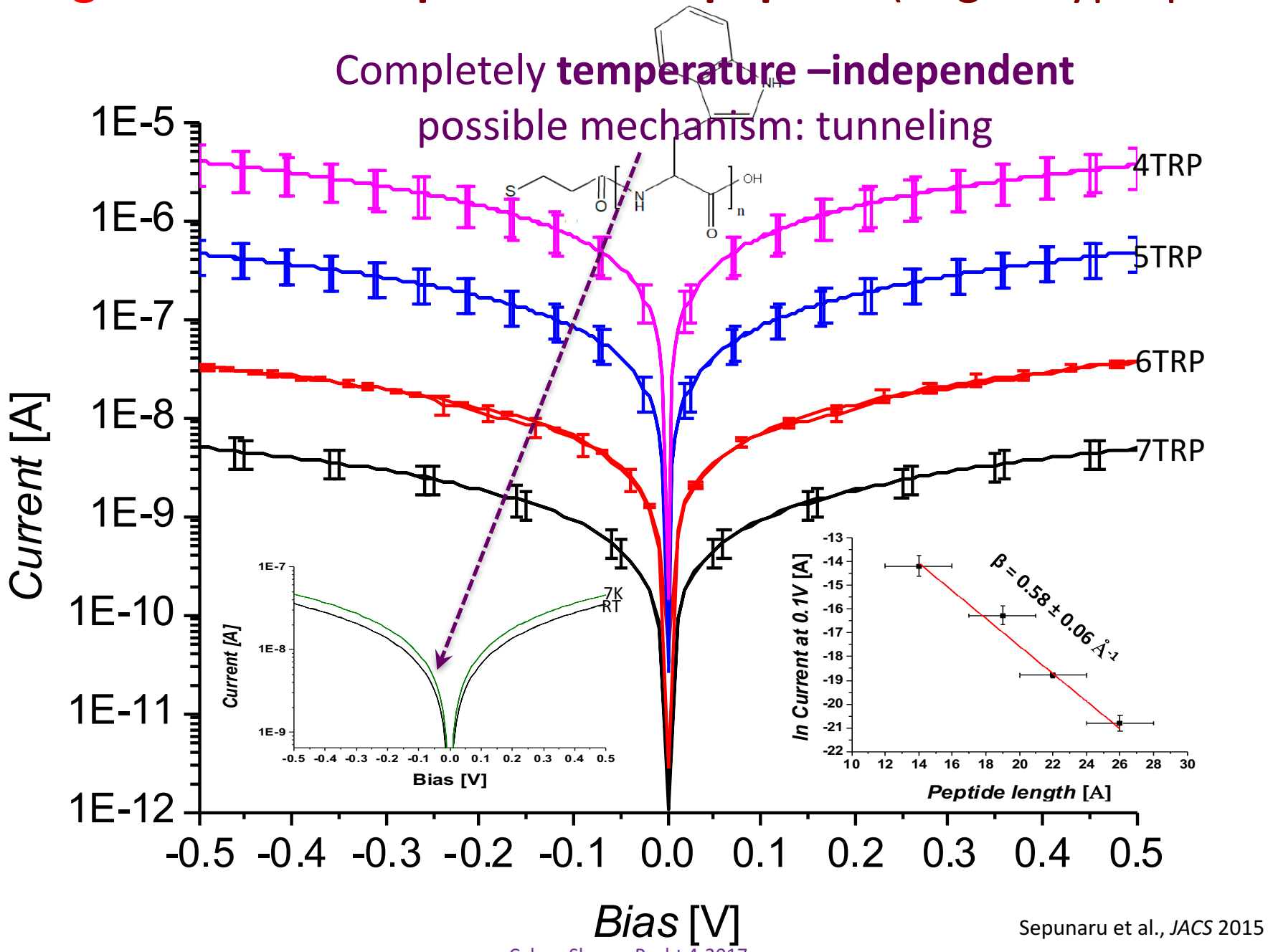
Leeor Kronik (DFT)

Koby Levy (MD)

Factors affecting peptide ET (→ ETp)

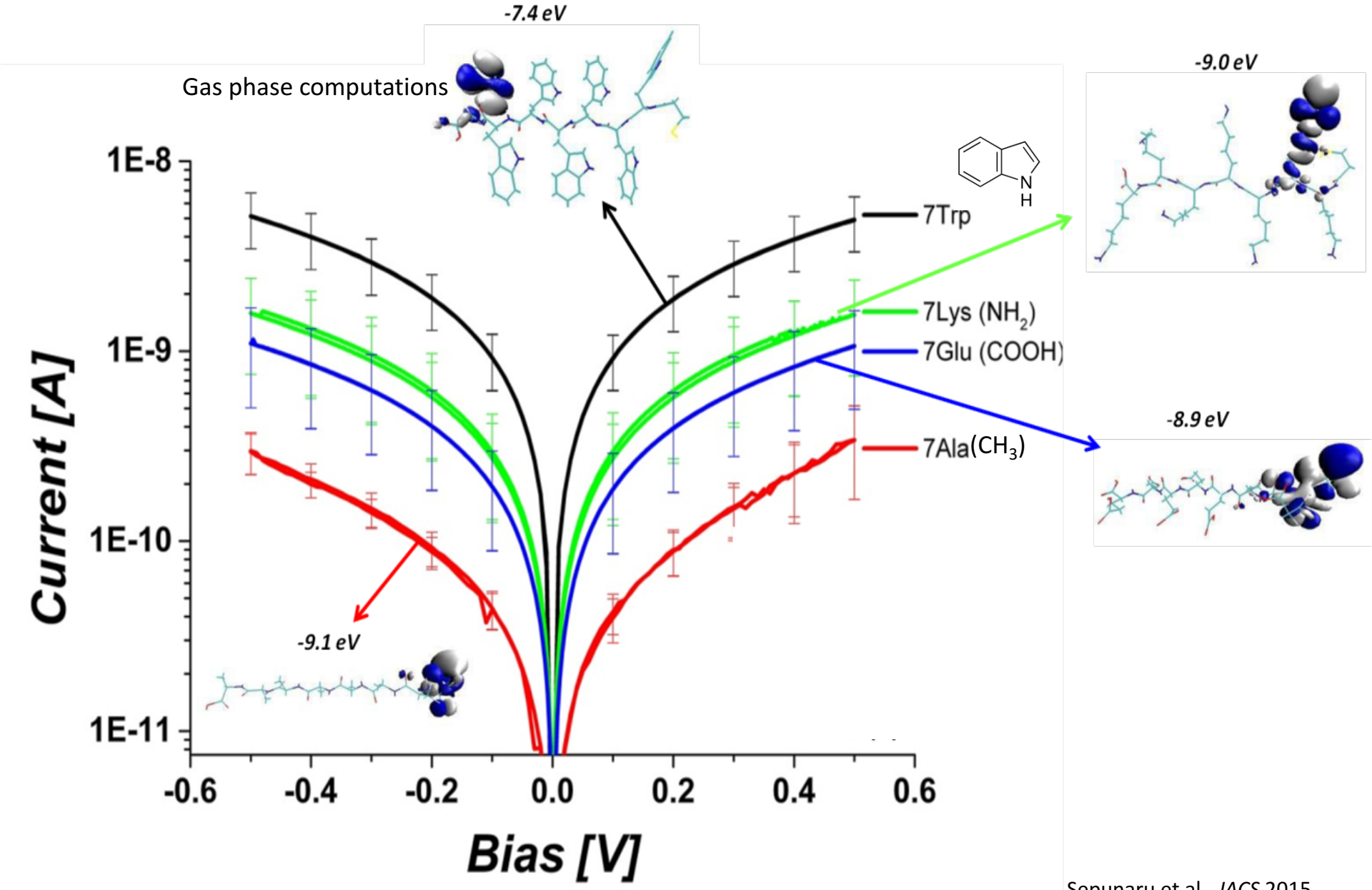


Length effect on ETp via HOMO-peptide (oligo-Tryptophan)



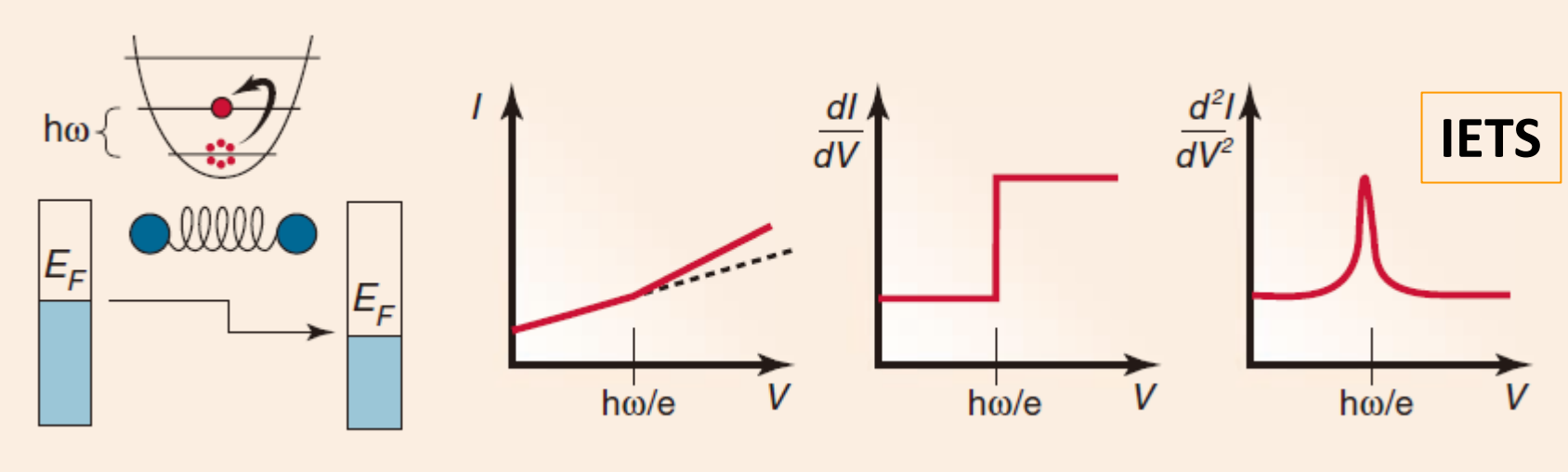
Sepunaru et al., *JACS* 2015

Strong **residue effect** on electron transport



Sepunaru et al., *JACS* 2015

Are there molecules in the junction ?



- Some tunneling electrons can **lose (or gain) energy by exciting (excited) vibrations of the molecules** between junctions.

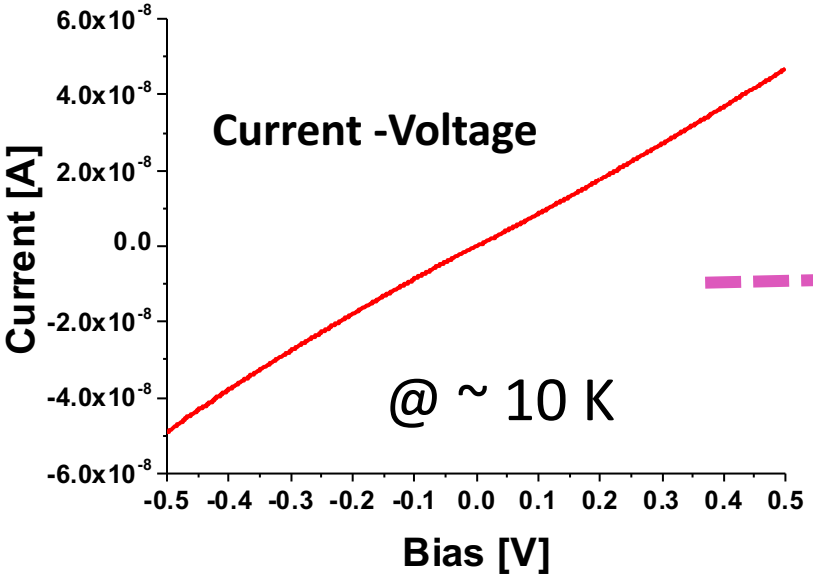
- The inelastic tunneling event is related with molecular vibrations:

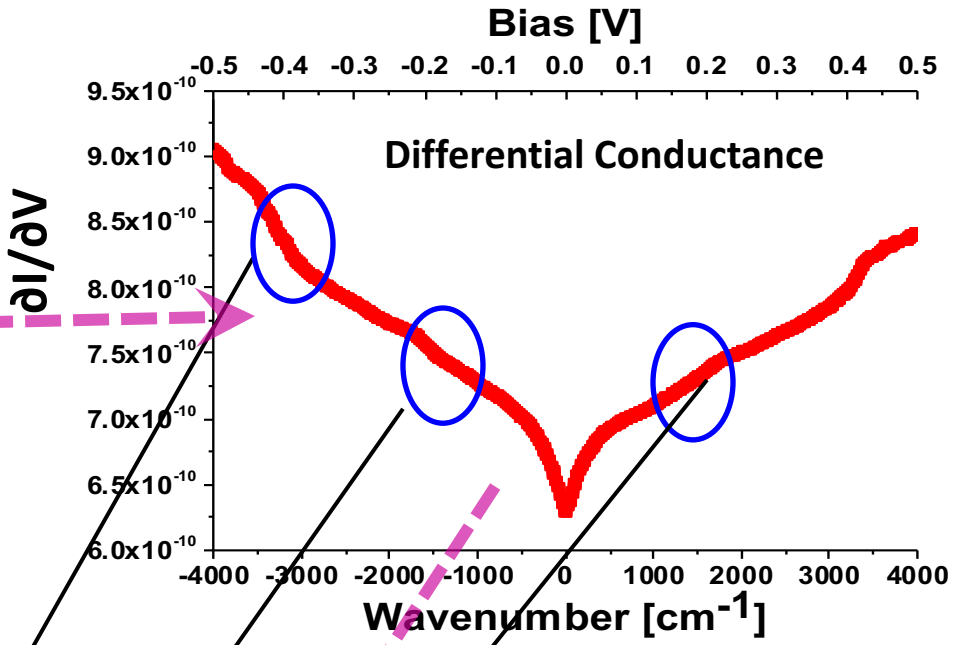
  Inelastic tunneling channel opens when $eV = \hbar\omega_{vib}$

- It can be compared with IR and Raman spectroscopy.

Jaklevic RC et. al. *PRL* 1966, Galperin M et. al. *Science* 2008

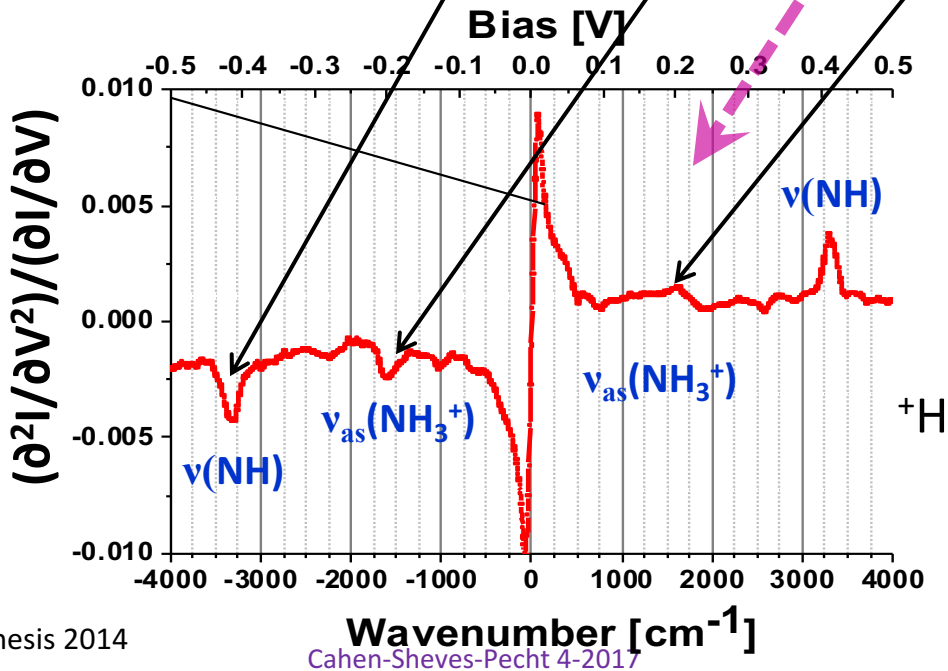
Yes, there are molecules in the junction !

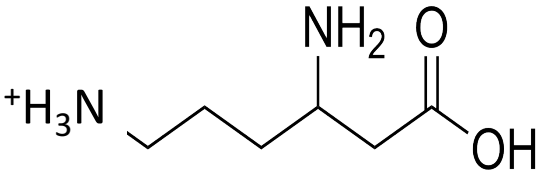




PEPTIDE

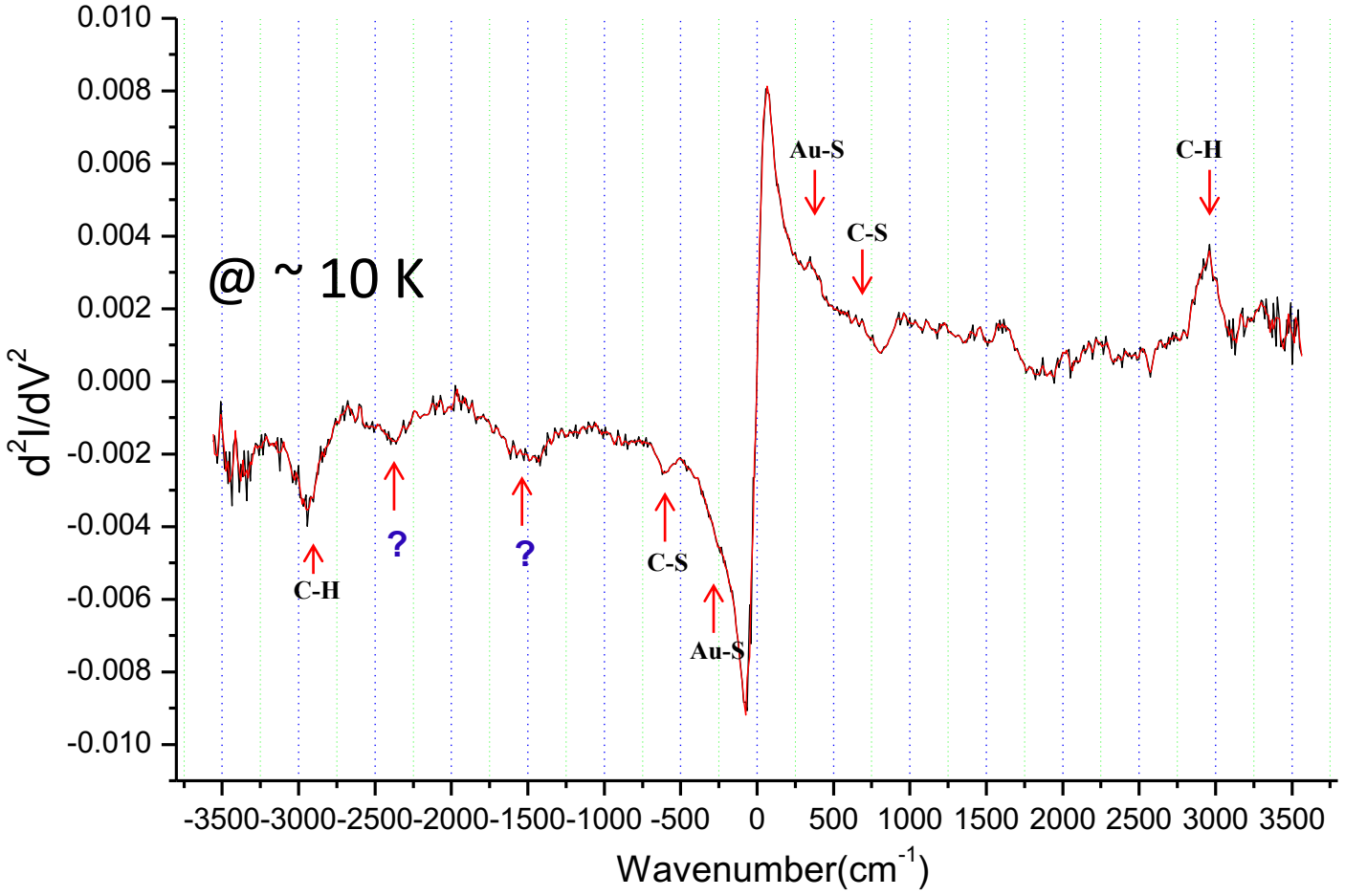
7-Lys, protonated





Sepunaru et al., *JACS* 2015; PhD thesis 2014

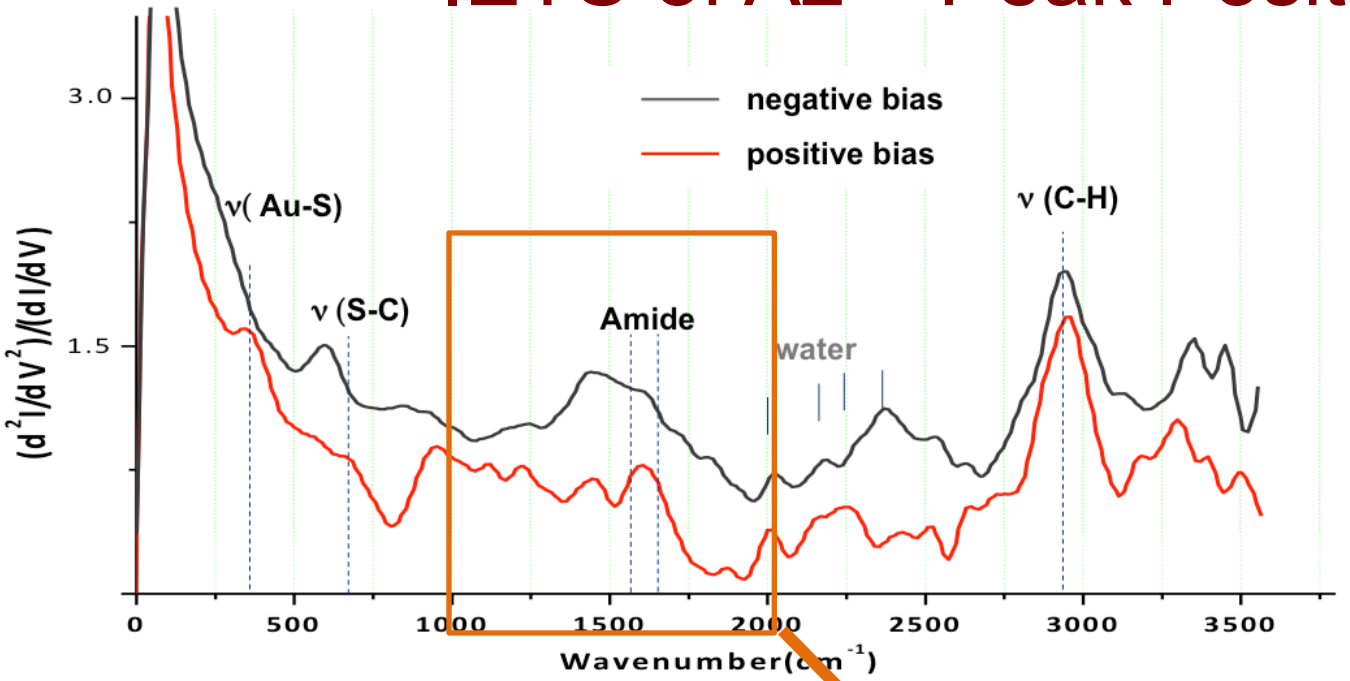
and also proteins:
IETS of Azurin on suspended nanowire



Au-S and S-C stretchings are clearly seen.
Electron injected into protein through Au-S-C.

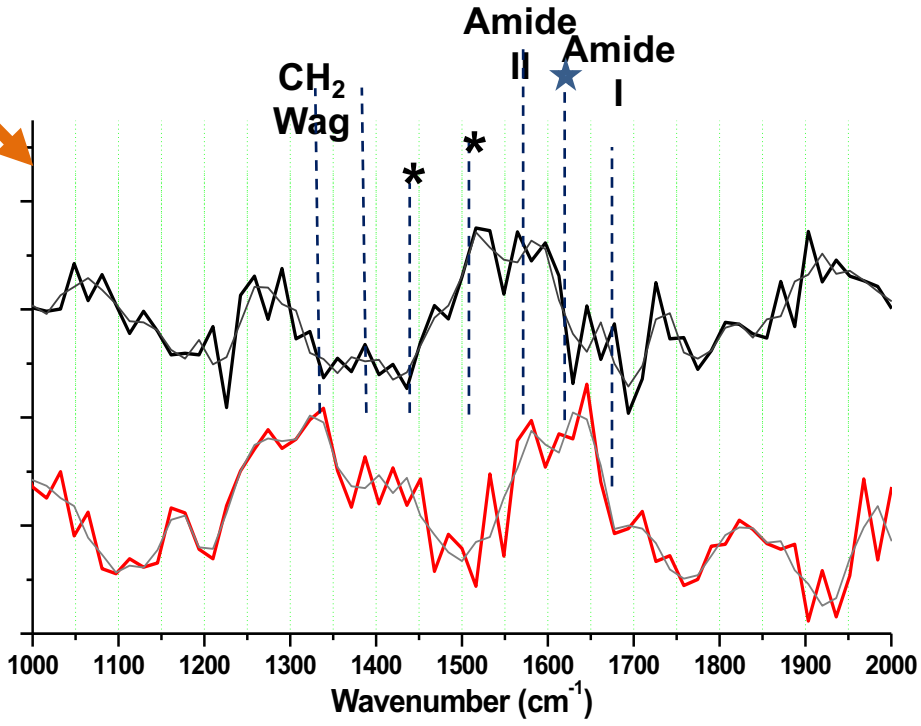
X. Yu *et al.,* (2015) *ACS Nano*

IETS of Az – Peak Positions

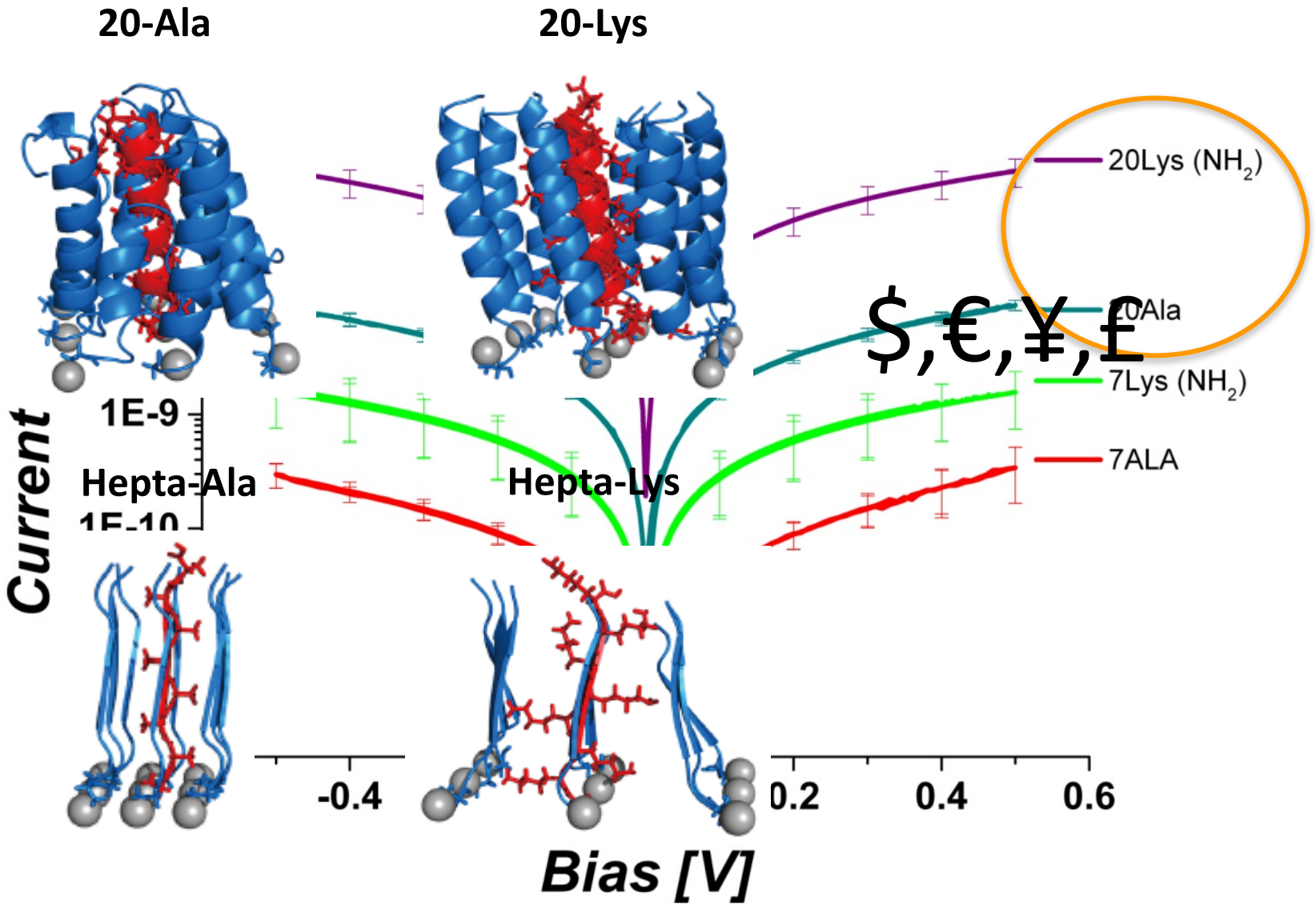


@ ~ 10 K

X. Yu *et al.,* (2015) *ACS Nano*



Adding **Secondary Structure** to the Peptides

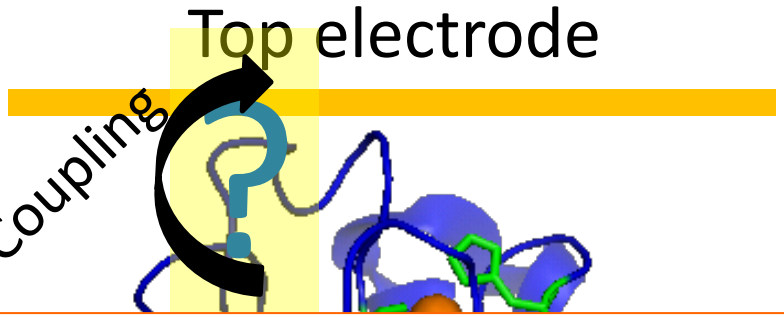


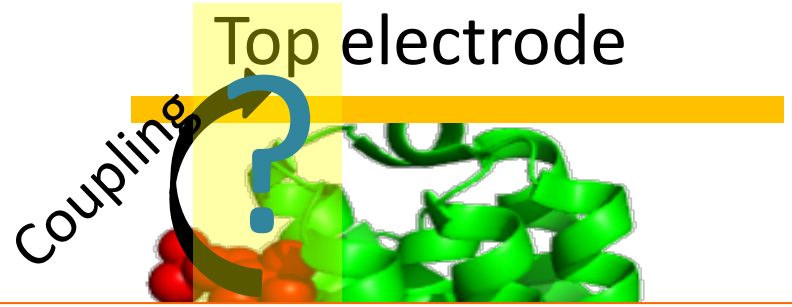
Sepunaru et al., *JACS* 2015

Some questions
that define what we don't know

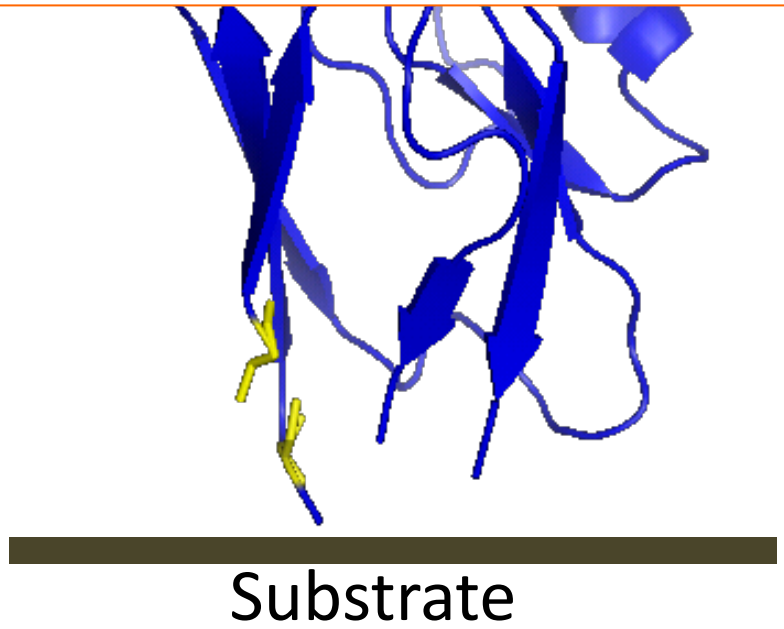
- What controls electron transport **across** proteins?
  - What controls electron transport **in** proteins?
  - How do electrons get in, and out of proteins?

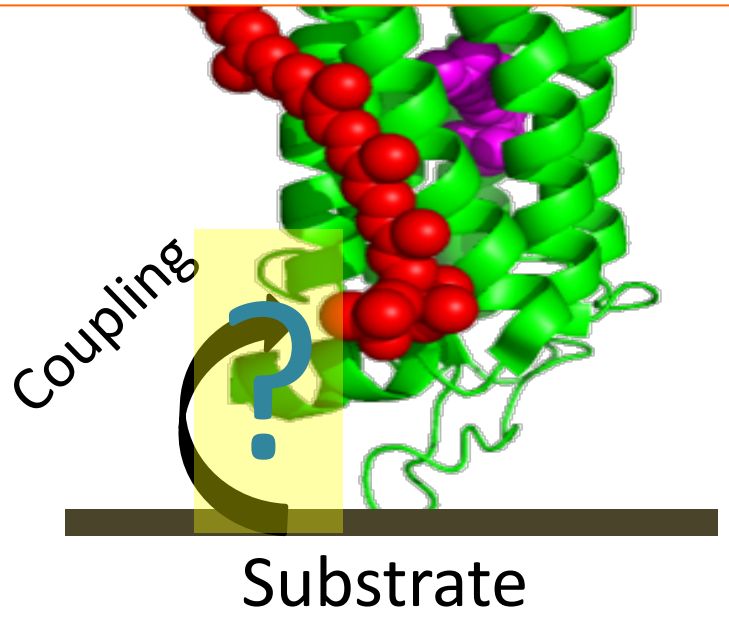
Temperature dependence is all about Electrode-Protein coupling (←→ Electrostatics)





Once the electron is IN, there seems no barrier till it EXITS





Sepunaru et al. *J. Am. Chem. Soc.*, 2011

Mukhopadhyay et al. *J. Am. Chem. Soc.* 2015

So, what do we know that we don't understand?

➡ **How** do electrons cross proteins?

Not yet for publication

Not yet for publication

Not yet for publication

Not yet for publication

Not yet for publication

Conclusions (1)

- ETp efficiency strongly affected by amino acid side chain.
  - Dominant transport mechanism – *(elastic) tunneling*
- Secondary structure– lowers barrier for ETp
- **Coupling to electrodes ≠ chemical bonding;**
  - apparently metal can screen *some* electrostatic barriers @ contacts

Conclusions (2)

→ Electron transport through a protein can be fully coherent

→ Consistent with "IN → OUT hypothesis" :
once electron enters, it can reach, without relaxation
or scattering the other electrode, *i.e.*, coherently

IF coupling is T- (in)dependent → transport is T- (in)dependent….

→ implications for Electron Transfer in and out of proteins
and its **control**

Progress of our work & understanding:

Acc. Chem. Res. **2010**, *43*, 945
Adv. Mater. **2014**, *42*, 7142
ArXiv **2017** abs/1702.05028

Thanks to

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+

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Nadav Amdursky (Technion)

Kronik group:

Sivan Refaely-Abramson

Piyush Agrawal

David Egger

Levy group:

Yulian Gavrilov,

Summary of protein electronics **riddles**

Proteins

- **conduct electrons "too efficiently"?**
- **show tunneling-like behaviour over "too long" distances ?**
- If, indeed electrode-protein coupling controls electron transport , ETp, **across** proteins, let's speculate:

  does redox entry & exit of e^--s control ET across proteins,

  while main ET(p) mechanism in protein is coherent tunneling,

  .. and redox protects protein from electron reducing power?

Future: Meet challenge of *solid state gating* (better handle on energetics);

protein-protein ETp → bacterial (Cyt) nanowires;

effects of soft electrodes (organic, lower [charge], on coupling;