

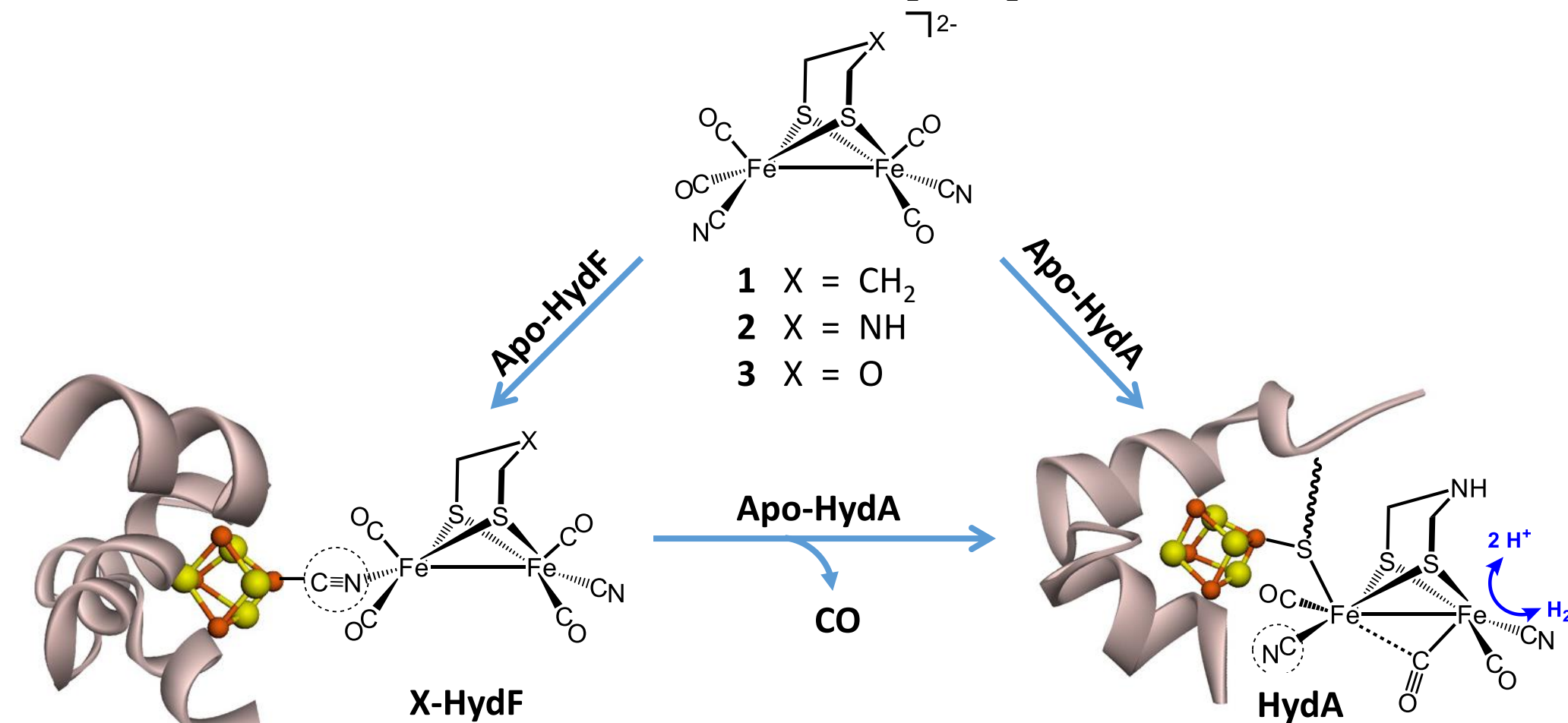
# Cyanide-Bridged Iron Complexes as Biomimetics of Tri-iron Arrangements in Maturases of the H cluster of the Diiron Hydrogenase

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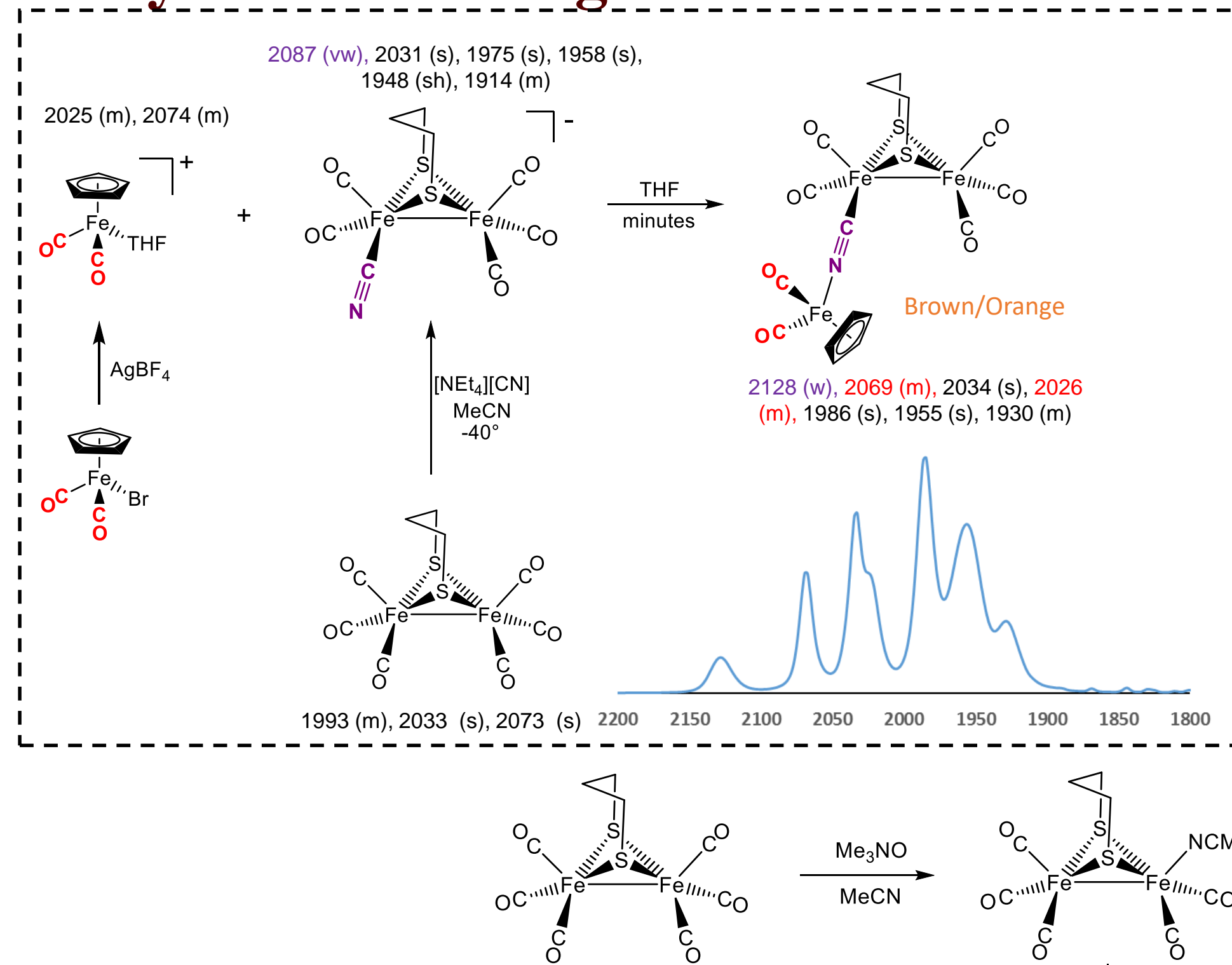
## Introduction

The discovery of cyano-iron carbonyls in the hydrogen processing enzymes, [NiFe]- and [FeFe]-hydrogenase ( $H_2$ ase) has inspired synthetic efforts by organometallic chemists to reproduce the active sites of these extraordinarily efficient  $H_2$ -producing or  $H_2$ -oxidation enzymes. The recent demonstration of the possibility of loading the apo-HydA (the [FeFe]- $H_2$ ase enzyme) and apo-HydF (a precursor protein in the maturation process) with synthetic analogues of the active site are convincing that the small molecular models are indeed the essential catalyst. Such “simple methodology of controlled metalloenzyme activation” as described by Bergrenn, et al., provided unambiguous evidence that the bridgehead atom X of the S to S linker,  $\mu$ -SCH<sub>2</sub>XCH<sub>2</sub>S, in the 2Fe-subsite is NH.



The interrogation by pulsed EPR spectroscopy of the Hyd-F protein containing the 2Fe subsite derived from the synthetic analogue yielded an unexpected conclusion. As shown above, a linear cyanide bridges the [4Fe-4S] cluster to the 2Fe subsite; furthermore, HYSCORE data found the unpaired electron of the [4Fe-4S] cluster to be strongly coupled to the C-13 nucleus of cyanide derived from the 2Fe unit prepared with labeled <sup>13</sup>CN<sup>-</sup>. The molecular interpretation of this observation is that the cyanide that anchors the 2Fe unit to the HydF carrier protein has flipped from its origin, placing the cyanide nitrogen next to the 2Fe site; another flip occurs, returning the CN<sup>-</sup> to the terminal position, as it is transferred to the apo-HydA. While examples of  $\mu$ -CN<sup>-</sup> linkage isomers are to be found in inorganic and organometallic chemistry, such maneuvers are not common.

## Synthetic Strategies for CN Isomers

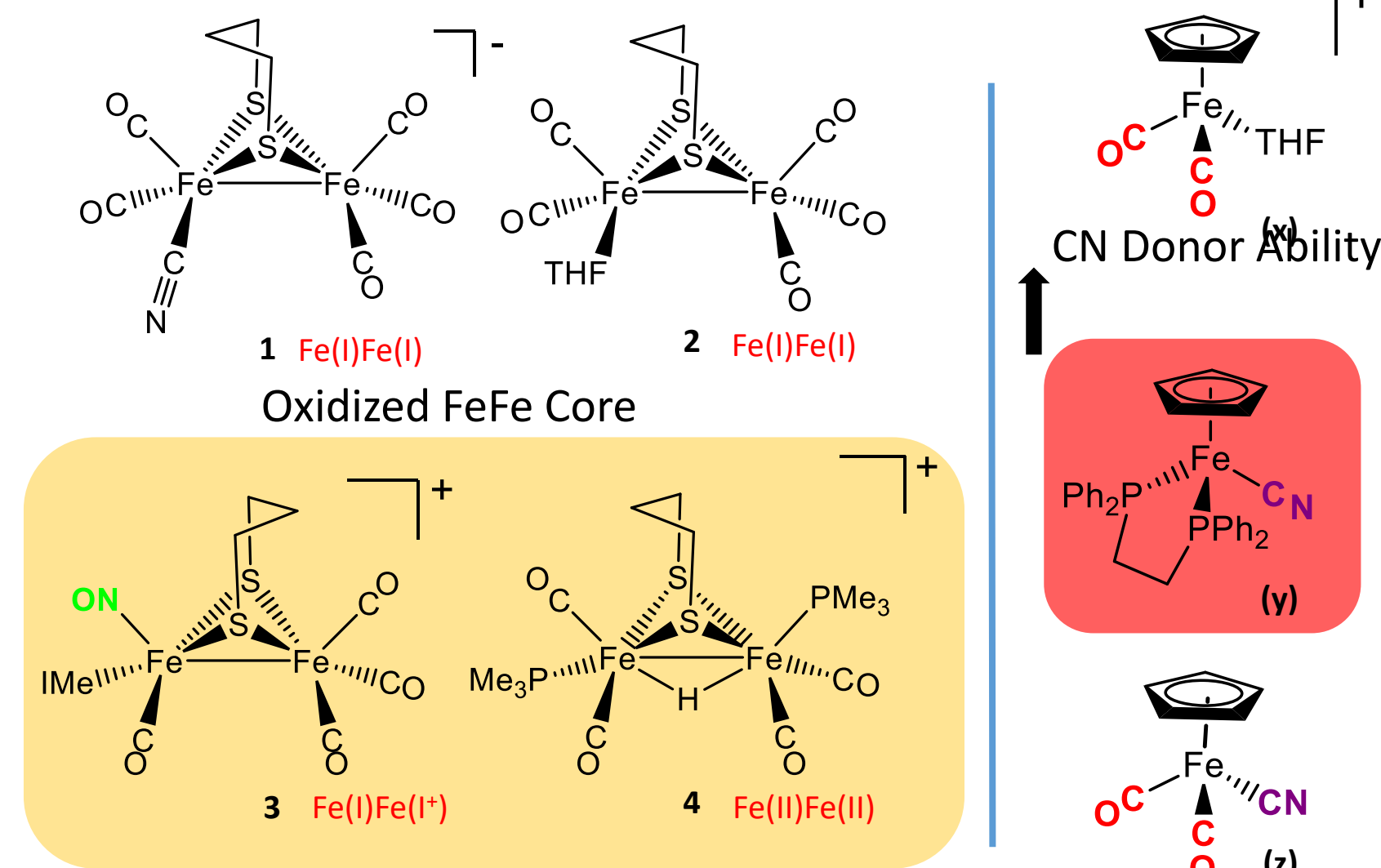


### Conclusions:

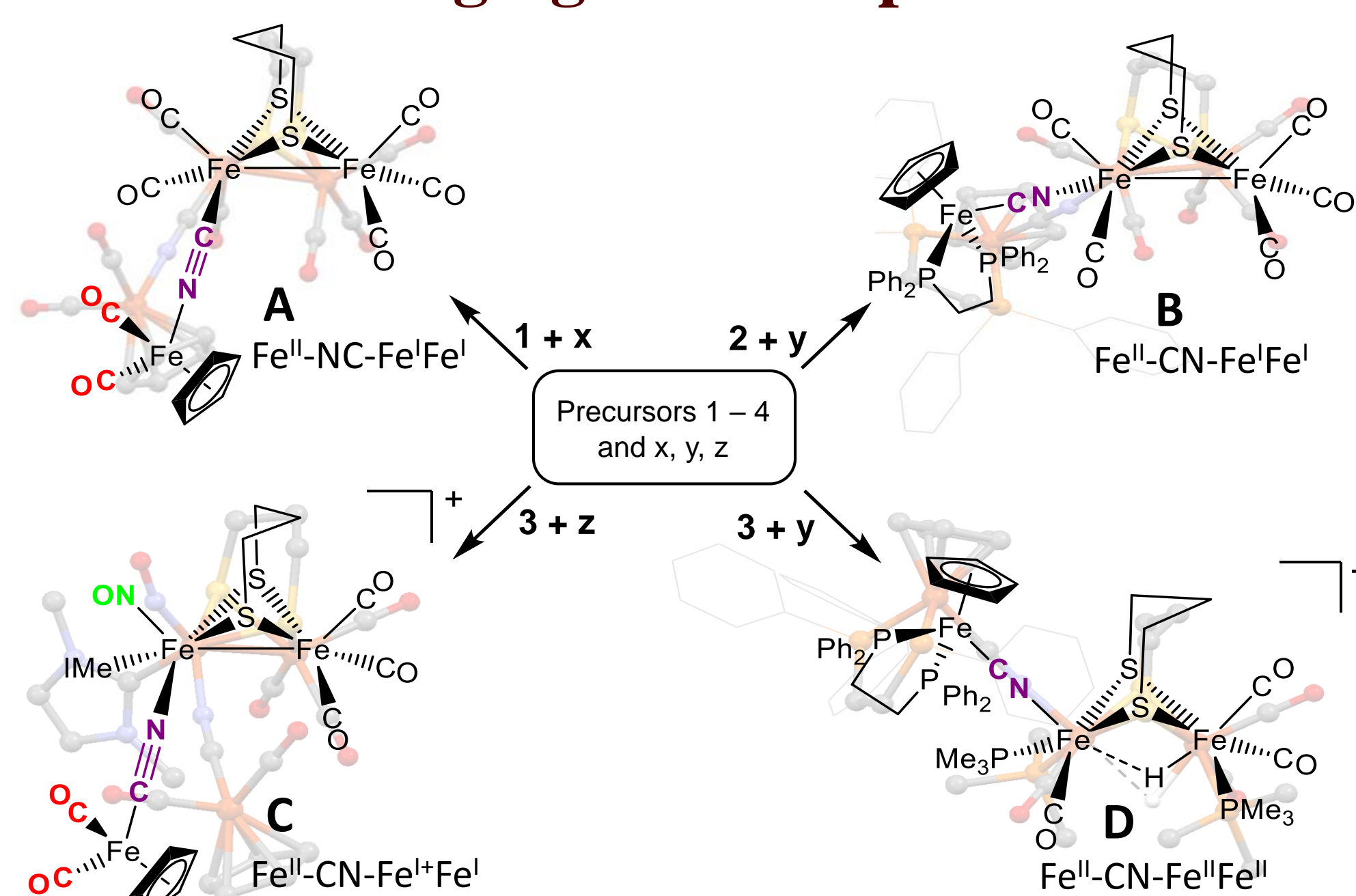
Donor ability of the cyanide on this Fe<sup>II</sup> (bound to 2 CO's) is insufficient for stable binding to Fe<sup>I</sup>. Therefore, must either increase donor ability of CpFeCN unit or increase acceptor ability of the diiron receiver unit

## Precursors

The seven compounds below were chosen as starting materials for bridging cyanide complexes **A-D**. Criteria for suitable precursors were formulated from the numerous metallo bridging cyanide studies conducted by Vahrenkamp *et al.* The precursors chosen for diiron complexes, complexes **1-4**, are noted to replicate certain features of the [FeFe]- $H_2$ ase enzyme active site, and the cyclopentadienyl iron derivatives, **x, y**, and **z**, are well known Fe(II) complexes, meant to mimic the oxidized iron in the 4Fe-4S cluster of HydF.



## Bridging CN Complexes



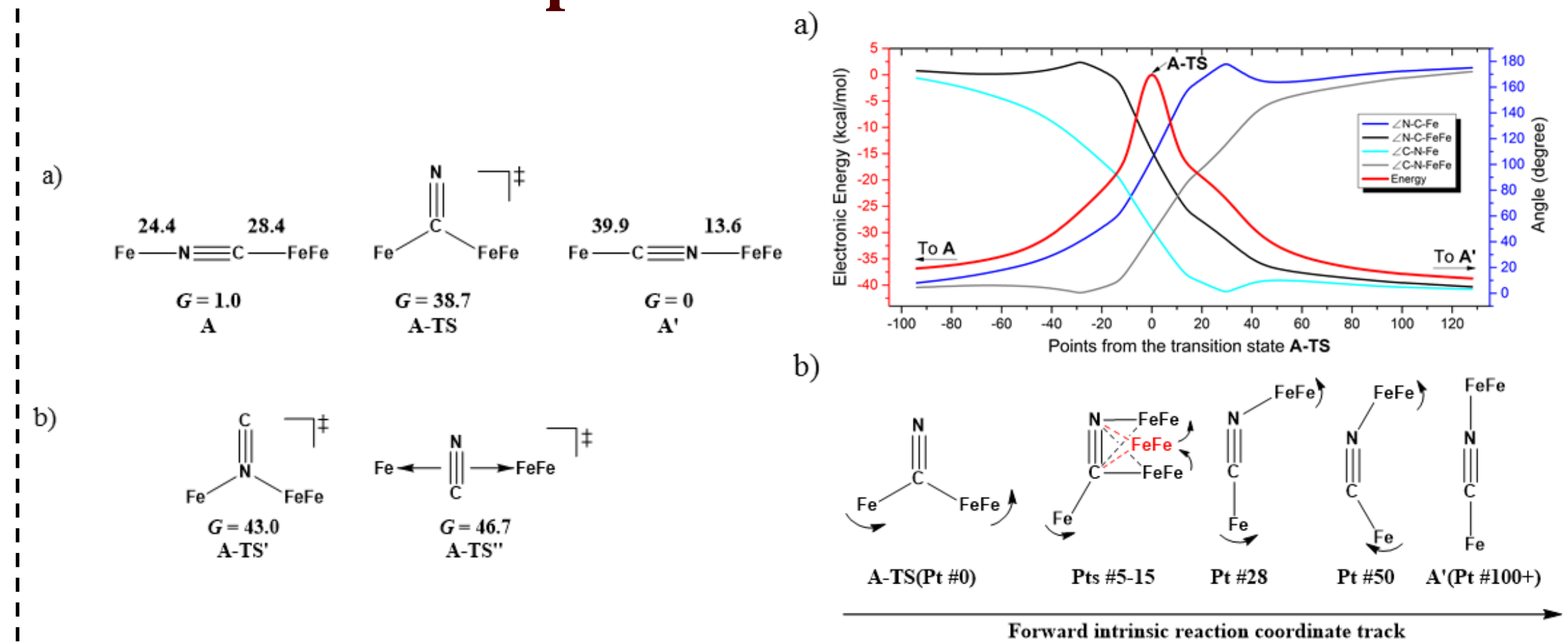
The four cyanide-bridged, three-Fe complexes displayed above were prepared, isolated, and characterized by x-ray diffraction. As noted in the table, there is a shift to higher values of the  $\nu$ (CN) in the product in contrast to the precursors, ascribed to the kinematic effect operative in bridging cyanides. There is also a gain of intensity in the  $\nu$ (CN) over the precursor consistent with the increased delocalization of  $\pi$  electron density in the Fe<sup>I</sup>-C $\equiv$ N-Fe<sup>II</sup> product. Thermal ellipsoid plots for compound **A** are shown below as well as the  $R_{int}$  for the assigned cyanide isomer (left structure below), carbon end bound to the diiron precursor, and the other possible isomer, carbon end bound to the CpFe unit (right structure below). When the carbon end of the cyanide is assigned to the CpFe unit the thermal ellipsoid is distorted and the  $R_{int}$  goes up resulting in a poor assignment for the atoms. Currently, cyanide coordination is assigned using this method as well as known Fe – C and Fe – N distances.

$R_{int} = 3.17$	$R_{int} = 3.43$
$R = 3.17$ vs. $3.43$	$R = 3.66$ vs. $3.92$
$R = 5.20$ vs. $5.38$	$R = 7.90$ vs. $7.94$

	A	B	C	D
[Fe <sup>I</sup> Fe <sup>I</sup> ] - C	1.917(4)	-	-	-
[Fe <sup>I</sup> Fe <sup>I</sup> ] - N	-	1.956 (2)	1.972 (3)	1.939 (5)
Fe <sup>II</sup> - C	-	1.876 (3)	1.900(4)	1.887(7)
Fe <sup>II</sup> - N	1.930(3)	-	-	-
C - N	1.152(4)	1.158 (4)	1.147(5)	1.172 (9)
$\nu$ (CN) cm <sup>-1</sup>	2128	2100	2154	2081

$\nu$ (CN) Values for Precursors	(cm <sup>-1</sup> )
CpFe(CO) <sub>2</sub> CN	2120
CpFe(dppe)CN	2061
( $\mu$ -pdt)[Fe(CO) <sub>3</sub> ][Fe(CO) <sub>2</sub> CN]	2087

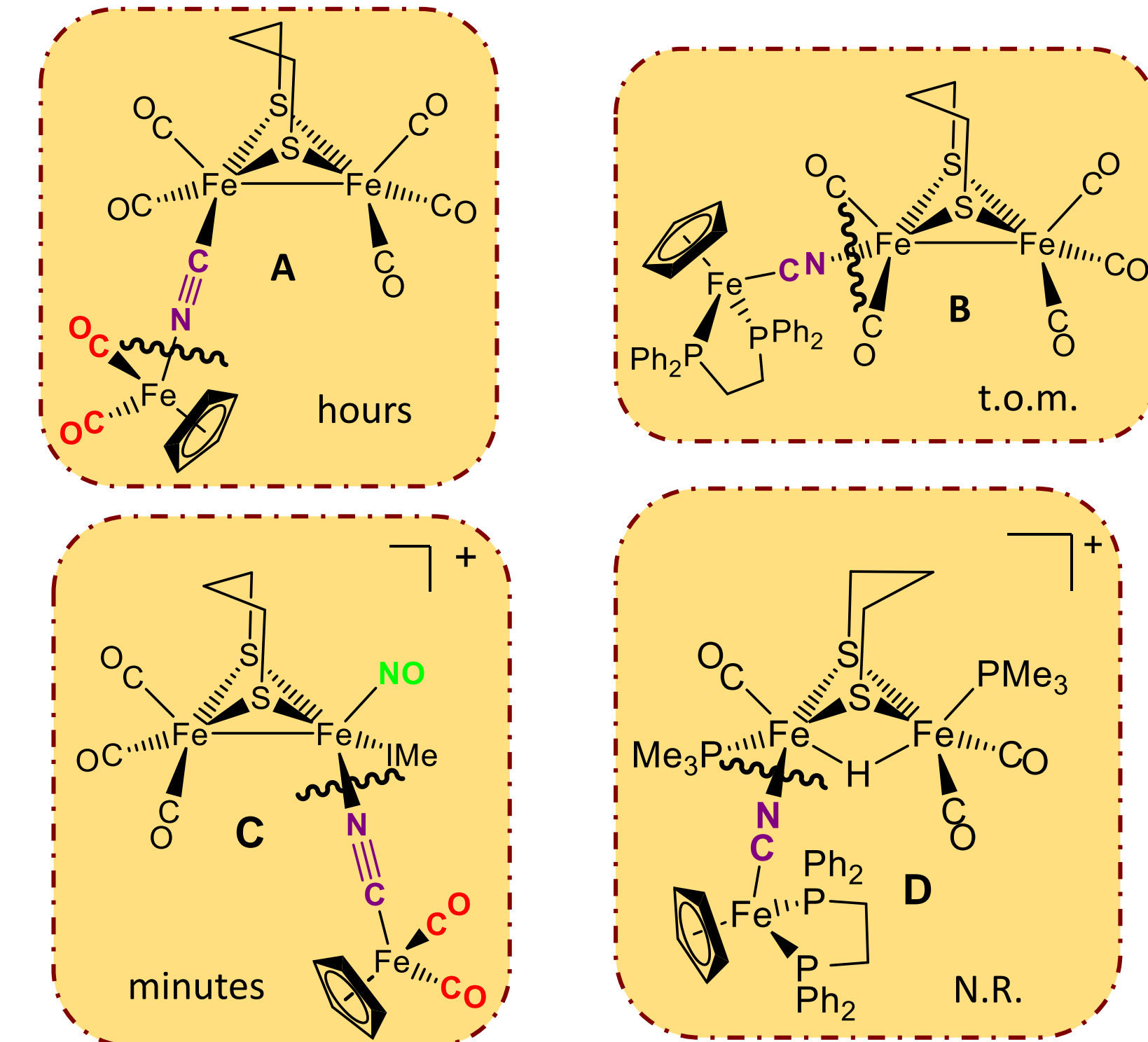
## Computational Mechanism



While there is only a 1 kcal/mol difference between isomer **A** and **A'**, the Gibbs free energy of **A-TS** connecting the two isomers was calculated to be 38.7 kcal/mol above the more stable isomer. The likely transition state connecting the isomers is associated with a wagging motion that initiates the asymmetric concomitant slide from one CN end to the other. The motion is indicative of an intramolecular transfer mechanism rather than a dissociation-association mechanism.

## Cleavage Reactions

Reactions of compounds **A-D** with nucleophiles such as CN<sup>-</sup>, PMe<sub>3</sub>, and SPh are used to model the transfer of the diiron cyanide unit from HydF to apo-HydA. Reaction of the bridging cyanide complex with nucleophiles results in cleavage of the Fe-N bond and no apparent cyanide isomerization. Complex **D** did not react.



## Conclusions

- ❖ Cyanide serves as a bridging ligand in our 3Fe constructs, consistent with proposal for the carrier maturase protein HydF
- ❖ Both linkage isomers, Fe-NC-[FeFe] and Fe-CN-[FeFe] made; orientation of CN bridge determined by the choice of reagents
- ❖ No obvious cyanide flipping was observed in synthetic routes or cleavage reactions
- ❖ Matches of electrophilicity of acceptor and nucleophilicity of donor determines stability of the bridged triiron units
- ❖ In Free World of solution chemistry, cleavage occurred in all cases at N-Fe bond.

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