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EVIDENCE FOR A SINGLE ELECTRON TRANSFER ACTIVATION IN THE HYDRIDE TRANSFER FROM AN NADH MODEL COMPOUND TO TETRACYANOETHYLENE

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New evidence for a stepwise mechanism which requires a single electron transfer activation in the hydride transfer from an NADH model compound, 1-benzyl-1,4-dihydronicotinamide (BNAH), to tetracyanoethylene (TCNE) has been presented based on the effects of pyridine on the stoichiometry of the overall reaction, the rate constant, and the kinetic isotope effect.

There have been considerable interest and debate concerning the reaction mechanism for hydride transfer from dihydronicotinamides to substrates.[1-4] The key mechanistic question has been whether the hydride transfer between NADH model compounds and substrates occurs in a direct one-step hydride transfer[2] or in multisteps involving initial single electron transfer from NADH model compounds to substrates.[3,4] A discrepancy observed between the kinetic (k_H/k_D) and product (Y_H/Y_D) isotope effects for the reduction of some substrates by NADH model compounds has been used as strong evidence for the stepwise mechanism against the direct hydride transfer.[3] However, it has recently been shown that such a discrepancy between k_H/k_D and Y_H/Y_D cannot be used as evidence for the presence of an intermediate.[5] Thus, there is no strong evidence supporting the stepwise mechanism for hydride transfer from NADH model compounds to substrates at the present time.

We wish to report herein new evidence for a stepwise mechanism in the hydride transfer reaction from an NADH model compound, 1-benzyl-1,4-dihydronicotinamide (BNAH), to tetracyanoethylene (TCNE) based on the effects of pyridine on the stoichiometry of the overall reaction, the rate constant, and the kinetic isotope effect k_H/k_D.

An NADH model compound BNAH reduces TCNE to the radical anion $TCNE^{\overline{\cdot}}$ in acetonitrile, the concentration of which can be readily determined by its characteristic spectrum containing twelve absorption maxima between 350 and 500 nm (*e.g.*, $\lambda_{max} = 457$ nm, $\varepsilon = 5.67 \times 10^4$ mol^{-1}dm^2).[6] The stoichiometry of the reduction of TCNE by BNAH determined from the electronic spectrum is given by Eq. 1. The formation of $TCNE^{\overline{\cdot}}$ has

$$2\text{BNAH} + 3\text{TCNE} \longrightarrow 2\text{BNA}^{+} + 2\text{TCNE}^{\overline{\bullet}} + \text{TCNEH}_2 \qquad (1)$$

been considered to occur *via* a hydride transfer from BNAH to TCNE (Eq. 2), yielding TCNEH^{-} which reacts with TCNE to form $\text{TCNE}^{\overline{\bullet}}$ as well as TCNEH_2 (Eq. 3).7)

$$\text{BNAH} + \text{TCNE} \xrightarrow{k} \text{BNA}^{+} + \text{TCNEH}^{-} \quad (2) \qquad 2\text{TCNEH}^{-} + \text{TCNE} \xrightarrow{\text{fast}} 2\text{TCNE}^{\overline{\bullet}} + \text{TCNEH}_2 \quad (3)$$

The rate of formation of $\text{TCNE}^{\overline{\bullet}}$ was measured from the rise of the absorbance of the 457 nm band due to $\text{TCNE}^{\overline{\bullet}}$ by using an Union RA-103 stopped flow spectrophotometer. The reaction obeyed the pseudo-first-order kinetics in the presence of large excess TCNE relative to BNAH. The pseudo-first-order rate constant $k^{(1)}$ was proportional to the TCNE concentration and the rate constant for the hydride transfer k (Eq. 2) has been determined as $3.2 \times 10^{2}\ \text{mol}^{-1}\text{dm}^{3}\text{s}^{-1}$ at 298 K from the relation $k^{(1)} = 2k[\text{TCNE}]/3$, which is derived from Eqs. 2 and 3. The kinetic isotope effect k_H/k_D for the hydride transfer8) has also been determined as $k_H/k_D = 2.0 \pm 0.2$, which is a typical value for hydride transfer reactions of BNAH.2-4)

When pyridine was added to the BNAH-TCNE system, remarkable effects were observe on both stoichiometry and the rate constant of the hydride transfer; stoichiometry in Eq. 1 changed to Eq. 4, and the rate constant k (Eq. 2)9) increased with increasing the

$$\text{BNAH} + 2\text{TCNE} + \text{Py} \longrightarrow \text{BNA}^{+} + 2\text{TCNE}^{\overline{\bullet}} + \text{PyH}^{+} \qquad (4)$$

pyridine concentration to approach a constant value which is 1.4×10^{4} times larger than that in the absence of pyridine. Furthermore, the kinetic isotope effect k_H/k_D also changed from 2.0 ± 0.2 to 1.1 ± 0.2 by the addition of large excess pyridine ($0.59\ \text{mol dm}^{-3}$). Such a change of the kinetic isotope effect to the value close to unity strongly suggests the change of the rate-determining step from a hydrogen or hydride transfer to non hydrogen transfer, *i.e.*, electron transfer.

Thus, the effects of pyridine on all the stoichiometry, the rate constant, and the k_H/k_D values are best interpreted by the following stepwise mechanism (Scheme 1),

Scheme 1. $$\text{BNAH} + \text{TCNE} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} (\text{BNAH}^{\dot{+}}\ \text{TCNE}^{\overline{\bullet}}) \xrightarrow{k_2} \text{BNA}^{+} + \text{TCNEH}^{-} \qquad (5)$$

In the presence of pyridine $$(\text{BNAH}^{\dot{+}}\ \text{TCNE}^{\overline{\bullet}}) + \text{Py} \xrightarrow{k_3} \text{BNA}\bullet + \text{PyH}^{+} + \text{TCNE}^{\overline{\bullet}} \qquad (6)$$

$$\text{BNA}\bullet + \text{TCNE} \xrightarrow{\text{fast}} \text{BNA}^{+} + \text{TCNE}^{\overline{\bullet}} \qquad (7)$$

where electron transfer activation from BNAH to TCNE (Eq. 5) occurs to form the ion pair ($\text{BNAH}^{\dot{+}}\ \text{TCNE}^{\overline{\bullet}}$), followed by fast hydrogen transfer (or H^{+} and electron) from $\text{BNAH}^{\dot{+}}$

to $TCNE^{\cdot -}$ in the ion pair (Eq. 5). It should be emphasized that electron transfer activation must be distinguished from usual electron transfer reactions where the oxidized and reduced species diffuse apart without occurrence of the reaction in the ion pair.10)

In the presence of pyridine, proton transfer from $BNAH^{+\cdot}$ to pyridine (Eq. 6) can compete or exceed the hydrogen transfer reaction (Eq. 5), leading to the formation of $BNA\cdot$ radical which transfers an electron to TCNE much more readily than BNAH11) to yield additional $TCNE^{\cdot -}$ as required in the stoichiometry (Eq. 4).

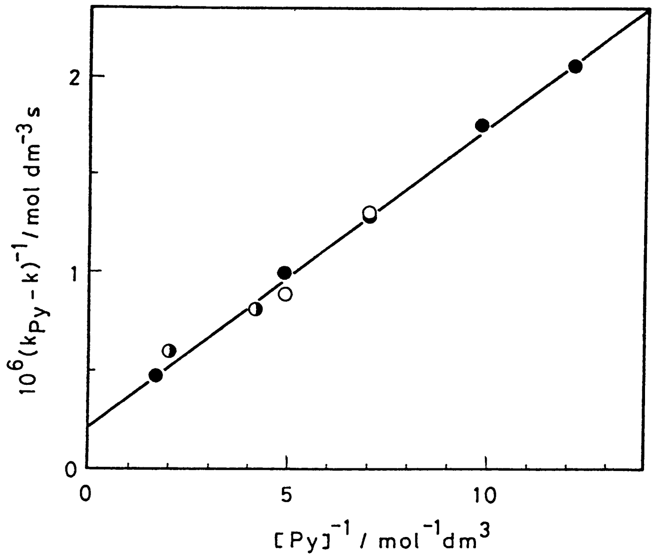

Fig. 1. A linear plot between $(k_{Py} - k)^{-1}$ and $[Py]^{-1}$ for the reaction of BNAH with TCNE in the presence of pyridine at 298 K; ○ [TCNE]: 4.97×10^{-4}, [BNAH]: 4.94×10^{-5} mol dm^{-3}; ◑ [TCNE]: 3.39×10^{-4}, [BNAH]: 2.51×10^{-5} mol dm^{-3}; ● [TCNE]: 2.04×10^{-4}, [BNAH]: 1.69×10^{-5} mol dm^{-3}; see Eq. 10.

According to Scheme 1, the rate constant of the apparent hydride transfer is expressed in terms of electron transfer rate constant k_1, hydrogen transfer rate constant k_2, and proton transfer rate constant k_3 as presented by Eqs. 8 and 9 in the absence (k) and presence of pyridine (k_{Py}), respectively. In the presence of pyridine, Eq. 9 can explain the increase of k_{Py} with increasing the pyridine concentration and k_{Py} in sufficiently large excess pyridine ($1 \gg k_{-1}/(k_2 + k_3[Py])$) corresponds to the electron transfer rate constant k_1, which accounts for the absence of the kinetic isotope effect. Since k_1 is much larger than k in the absence of pyridine, the relation $k_{-1}/k_2 \gg 1$ is derived from Eq. 8. Under such a condition, k is reduced to k_2k_1/k_{-1} where the hydrogen transfer step (k_2 in Eq. 5) is the rate-determining step, which accounts for the appreciable kinetic isotope effect $k_H/k_D = 2.0$ in the absence of pyridine. From Eqs. 8 and 9 is derived Eq. 10 by using the relation $k_{-1}/k_2 \gg 1$.

$$k = k_1/(1 + k_{-1}/k_2) \qquad (8) \qquad\qquad k_{Py} = k_1/[1 + k_{-1}/(k_2 + k_3[Py])] \qquad (9)$$

$$1/(k_{Py} - k) = k_{-1}/(k_1k_3[Py]) + 1/k_1 \qquad (10)$$

According to Eq. 10, $(k_{Py} - k)^{-1}$ is plotted against $[Py]^{-1}$ as shown in Fig. 1. The linear correlation in Fig. 1 thus confirms the validity of Eq. 10.

If one-step hydride transfer mechanism is correct, only explanation for the effects of pyridine described above is that the reaction mechanism may be changed from the one-step hydride transfer (BNAH + TCNE → BNA^+ + $TCNEH^-$) in the absence of pyridine to the electron transfer (BNAH + TCNE → $BNAH^{+\cdot}$ + $TCNE^{-\cdot}$) in the presence of pyridine. Such a change of the mechanisms, however, cannot account for the increase of the rate constant with increasing the pyridine concentration.12)

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