



# Natural abundance kinetic isotope effects: expt. vs theory.

HENRY RZEPA

READ REVIEWS

WRITE A REVIEW

CORRESPONDENCE:

[h.rzepa@imperial.ac.uk](mailto:h.rzepa@imperial.ac.uk)

DATE RECEIVED:

June 10, 2015

DOI:

10.15200/winn.143386.64236

ARCHIVED:

June 09, 2015

CITATION:

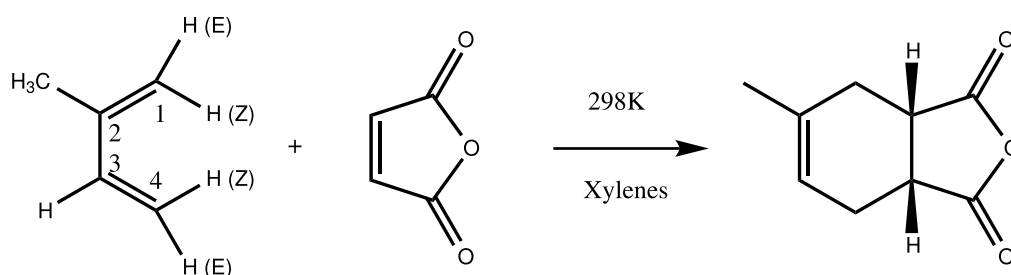
Henry Rzepa, Natural abundance kinetic isotope effects: expt. vs theory., *The Winnower* 2:e143386.64236, 2015, DOI: 10.15200/winn.143386.64236

© Rzepa This article is distributed under the terms of the [Creative Commons Attribution 4.0 International License](#), which permits unrestricted use, distribution, and redistribution in any medium, provided that the original author and source are credited.



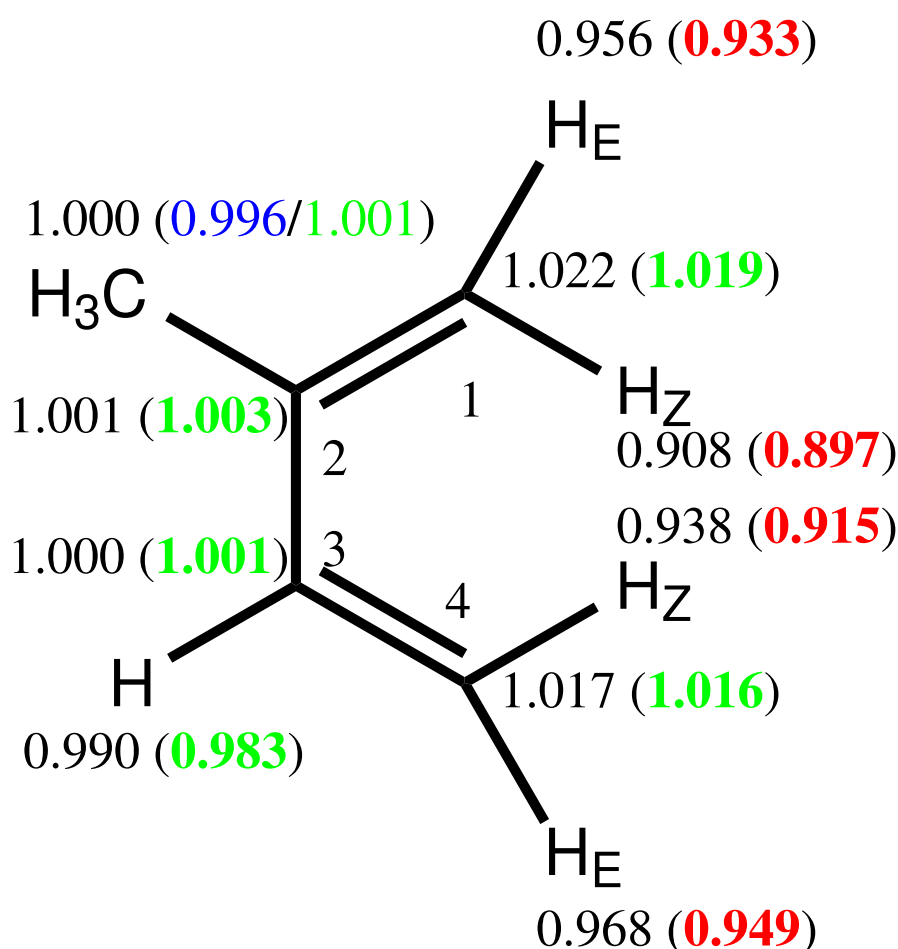
My PhD thesis involved determining kinetic isotope effects (KIE) for aromatic electrophilic substitution reactions in an effort to learn more about the nature of the transition states involved.[1] I learnt relatively little, mostly because a transition state geometry is defined by 3N-6 variables (N = number of atoms) and its force constants by even more and you get only one or two measured KIE per reaction; a rather under-defined problem in terms of data! So I decided to spend a PostDoc learning how to invert the problem by computing the anticipated isotope effects using quantum mechanics and then comparing the predictions with measured KIE.[2] Although such computation allows access to ALL possible isotope effects, the problem is still under-defined because of the lack of measured KIE to compare the predictions with. In 1995 Dan Singleton and Allen Thomas reported an elegant strategy to this very problem by proposing a remarkably simple method for obtaining KIE using natural isotopic abundances.[3] It allows isotope effects to be measured for **all** the positions in one of the reactant molecules by running the reaction close to completion and then recovering unreacted reactant and measuring the changes in its isotope abundances using NMR. The method has since been widely applied[4],[5] and improved.[6] Here I explore how measured and calculated KIE can be reconciled.

The original example uses the Diels-Alder cycloaddition as an example, with the 2-methylbutadiene component being subjected to the isotopic abundance. Although comparison with calculation on related systems was done at the time[7] the computational methods in use then did not allow effects such as solvation to be included. I thought it might be worth re-investigating this specific reaction using more modern methodology ( $\omega$ B97XD/Def2-TZVPP/SCRF=xylene), giving an opportunity for testing one key assumption made by Singleton and Allen, viz the use of an internal isotope reference for a site where the KIE is assumed to be exactly 1.000 (the 2-methyl group in this instance). This assumption made me recollect my post on how methyl groups might not be **entirely passive** by rotating (methyl "flags") in the Diels-Alder reaction between *cis*-butene and 1,4-dimethylbutadiene. I had concluded that post by remarking that *Rotating methyl groups should be looked at more often as harbingers of interesting effects*, which in this context may mean that such rotations may not be entirely isotope agnostic.



To start, I note that the *endo* (closed shell, *i.e.* non-biradical; the wavefunction is STABLE to open shell solutions) transition state obtained for this reaction[8],[9] has a computed dipole moment of 6.1D, just verging into the region where solvation starts to make an impact. Perhaps the most important conclusion drawn from Singleton and Allen's original article[2] was that the presence of an apparently innocuous 2-methyl substituent is sufficient to render the reaction asynchronous, which means that one C-C bond forms faster than the other. They drew this conclusion from observing that the inverse deuterium isotope effect was larger at C1 than C4, the difference being well outside of their estimated errors. The calculations indicate that the two bonds have predicted lengths of 2.197 (to C1) and 2.294Å (to C4) at the transition state. This means that an asynchronicity as small as  $\Delta 0.1\text{\AA}$  can be picked up in measured isotope effects!

The calculated activation free energy is 19.2 kcal/mol (0.044M), which is entirely reasonable for a reaction occurring slowly at room temperature. The barrier for the *exo* isomer is 21.0 kcal/mol, 1.8 kcal/mol higher in free energy.<sup>†</sup> The measured isotope effects are shown below with the predicted values in brackets. The colour code is green=within the estimated experimental error, red=outside the error.



The following observations can be made:

1. The internal isotope reference assumed as 1.000 is reasonable for carbon, but the "rotating methyl groups" resulting from hyper conjugation between the C-H groups and the  $\pi$  system do have a small effect resulting in a predicted KIE of 0.996 rather than the assumed 1.000. This will impact upon all the other measured values to some extent.
2. All the predicted<sup>†</sup>  $^{13}\text{C}$  isotope effects agree with experiment within the error estimated for the latter. The calculation also has its errors, of which the most obvious is that harmonic frequencies are used rather than the more correct anharmonic values.
3. The  $^2\text{H}$  isotope effects show more deviation. This might be a combination of the assumption that the internal Me reference has no isotope effect coupled with the use of harmonic frequencies for the calculation.
4. Although the  $^2\text{H}$  values differ somewhat beyond the experimental error, the E/Z effects are well reproduced by calculation. The inverse isotope effect for the (Z) configuration is significantly larger in magnitude than for the (E) form, as was indeed noted by Singleton and Thomas.
5. So too is the asymmetry induced by the methyl group. The inverse isotope effects are greater for the more completely formed bond (to C1) than for the lagging bond (to C4). They are indeed a very sensitive measure of reaction synchronicity.

The pretty good agreement between calculation and experiment provides considerable reassurance that the calculated properties of transition states can indeed be subjected to *reality checks using experiment*. Indeed, it takes little more than a day to compute a complete set of KIEs, far less than it takes to measure them. One could easily argue that such a computation should accompany measured KIE whenever possible.

<sup>†</sup>This gives me an opportunity to extol the virtues of effective RDM (research data management). The two DOIs for the data include files containing the full coordinates and force constant matrices for both reactant and TS. Using these, one can obtain frequencies for any isotopic substitution you might wish to make in <1 second each, and hence isotope effects not computed here. One option might be to *e.g.* invert the reactant from the 2-methylbutadiene to the maleic anhydride and hence compute the isotope effects expected on this species (not reported in the original article) or to monitor instead the product.<sup>[10]</sup>

<sup>†</sup>The KIE have only subtle small differences to the *endo* isomer; too small to assign the stereochemistry with certainty.

## REFERENCES

1. B.C. Challis, and H.S. Rzepa, "The mechanism of diazo-coupling to indoles and the effect of steric hindrance on the rate-limiting step", *J. Chem. Soc., Perkin Trans. 2* pp. 1209, 1975.  
<http://dx.doi.org/10.1039/p29750001209>
2. M.J.S. Dewar, S. Olivella, and H.S. Rzepa, "Ground states of molecules. 49. MINDO/3 study of the retro-Diels-Alder reaction of cyclohexene", *J. Am. Chem. Soc.*, vol. 100, pp. 5650-5659, 1978.  
<http://dx.doi.org/10.1021/ja00486a013>
3. D.A. Singleton, and A.A. Thomas, "High-Precision Simultaneous Determination of Multiple Small Kinetic Isotope Effects at Natural Abundance", *J. Am. Chem. Soc.*, vol. 117, pp. 9357-9358, 1995.  
<http://dx.doi.org/10.1021/ja00141a030>
4. Y. Wu, R.P. Singh, and L. Deng, "Asymmetric Olefin Isomerization of Butenolides via Proton Transfer Catalysis by an Organic Molecule", *J. Am. Chem. Soc.*, vol. 133, pp. 12458-12461, 2011.  
<http://dx.doi.org/10.1021/ja205674x>
5. J. Chan, A.R. Lewis, M. Gilbert, M. Karwaski, and A.J. Bennet, "A direct NMR method for the measurement of competitive kinetic isotope effects", *Nature Chemical Biology*, vol. 6, pp. 405-407, 2010. <http://dx.doi.org/10.1038/nchembio.352>
6. J.W. Storer, L. Raimondi, and K.N. Houk, "Theoretical Secondary Kinetic Isotope Effects and the Interpretation of Transition State Geometries. 2. The Diels-Alder Reaction Transition State Geometry", *J. Am. Chem. Soc.*, vol. 116, pp. 9675-9683, 1994.

<http://dx.doi.org/10.1021/ja00100a037>

7. Henry S Rzepa., "C 9 H 10 O 3", 2015. <http://dx.doi.org/10.14469/ch/191299>
8. Henry S Rzepa., "C 9 H 10 O 3", 2015. <http://dx.doi.org/10.14469/ch/191301>
9. D.E. Frantz, D.A. Singleton, and J.P. Snyder, " 13 C Kinetic Isotope Effects for the Addition of Lithium Dibutylcuprate to Cyclohexenone. Reductive Elimination Is Rate-Determining ", *J. Am. Chem. Soc.*, vol. 119, pp. 3383-3384, 1997. <http://dx.doi.org/10.1021/ja9636348>