ORGANIC LETTERS

2006
Vol. 8, No. 17
3651−3654

Probing Selectivity in Recognition-Mediated Dynamic Covalent Processes

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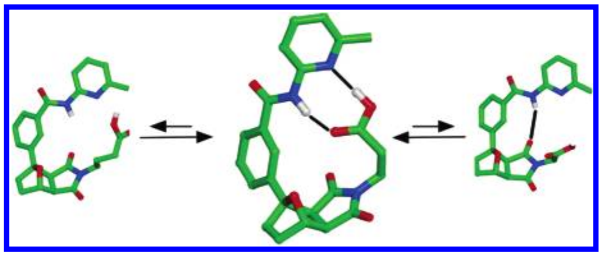
Received May 12, 2006

ABSTRACT



Two simple recognition-mediated dynamic Diels–Alder systems are used to probe the role of kinetics and thermodynamics in determining the equilibrium position in exchanging libraries and the time taken to reach that equilibrium. The selectivity expressed by recognition-driven dynamic processes is demonstrated to be less than the free-energy difference between the components as a result of compensatory effects arising from the extent of conversion to products within the library.

In contrast to combinatorial chemistry, which allows the efficient and rapid syntheses of thousands of molecules through kinetically controlled, irreversible processes, the construction of dynamic combinatorial libraries—a nascent field[1] in supramolecular chemistry—relies on the formation of structures that are generated by thermodynamically controlled reversible covalent bonds. Conceptually, two approaches may be considered depending on whether a receptor or a substrate acts as a template for the assembly of the other partner: "casting" consists of the receptor-induced assembly of a substrate that fits the receptor. Conversely, "molding" relies on the ability of the substrates to induce the assembly of a receptor that binds or fits the substrate optimally. Because all covalent bonds in the system form under thermodynamic control, there is an element of proofreading and repair of incorrect steric and electronic fits with the template. A range of reversible chemical transformations[2] (e.g., imine formation, transacylation, olefin metathesis, or (retro) Diels−Alder reactions) have been employed in the construction of dynamic selection systems. In most examples, several components which will ultimately form the receptor are mixed with the template and the system is allowed to reach equilibrium. The process, in theory, generates the most thermodynamically stable complex.

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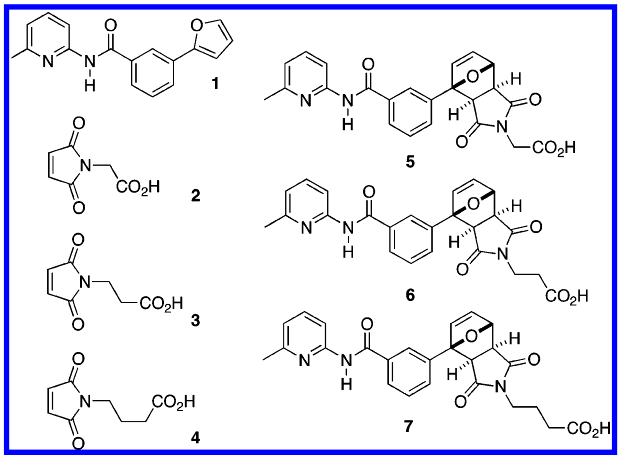
10.1021/ol061177m CCC: $33.50

Considerable effort[3] has been directed at understanding the limits of this selection process. Because the entire process is under thermodynamic control, one might expect that the energy difference between the most stable and the next most stable species in the equilibrating mixture will determine the observed selectivity.

Here, we demonstrate using a very simple model system, based on a reversible Diels−Alder reaction, that compensation effects exist in dynamic systems which serve to limit selectivity under certain conditions. Additionally, we demonstrate that kinetic effects are also a side effect of these recognition-mediated selection processes, and these may affect the time taken to reach equilibrium.



Previously, we described[4] the recognition-mediated reaction between the diene **1** and the dienophile **3**. In polar solvents, such as DMSO, this reaction[5] does not proceed in the forward direction to any significant extent as a result of the unfavorable loss of conjugation in the diene upon reaction. In nonpolar solvents, such as $CHCl_3$, the reaction proceeds rapidly to around 50% conversion. The interaction between the amidopyridine ring and the carboxylic acid in the product **6** is responsible for this difference in behavior—the formation of two hydrogen bonds between these groups in $CHCl_3$ stabilizes the product ground state significantly rendering the reaction favorable in terms of free energy. These hydrogen bonds can also serve to preorganize the reagents prior to reaction, thus imparting a strong influence on the kinetics of the reaction. Kinetic studies on the conversion of **1** and **3**−**6** revealed that, although the reaction was accelerated around 8-fold, the dominant effect was the stabilization of the product ground state. We realized, by extending our studies to encompass the related dienophiles **2** and **4**, that these systems present an opportunity to assess quantitatively the role of kinetic and thermodynamic effects within a small dynamic library of cycloadducts. In particular, we wished to explore whether the differences in stability between the cycloadducts are expressed fully in an exchanging library and whether recognition-induced rate acceleration plays any role in the equilibration processes of the library.

Initially, we determined the equilibrium positions for each of the reactions that form the cycloadducts **5**−**7** by measuring the kinetics of the reactions by ^1H NMR spectroscopy in $CDCl_3$ at 50 °C. These rate data were fitted to appropriate kinetic models, and forward and reverse rate constants were extracted (see Supporting Information). For comparison purposes, the control reaction described previously[6] was used as a model of the situation in which recognition has no influence on the reaction.

The equilibrium constants for each of the reactions and the corresponding relative free energies are shown in Figure 1. Clearly, all three systems exhibit a greater extent of reaction than the control (Figure 1, **NR**), with cycloadduct **6** being the most stable. The two-carbon spacer in **6** is exactly the correct length to permit the formation of two stabilizing hydrogen bonds between the carboxylic acid and amidopyridine. Molecular mechanics calculations indicate that, in the case of **5**, the one-carbon spacer is too short to permit the formation of two hydrogen bonds and that, in **7**, the three-carbon spacer is too long. Clearly, the recognition present in these systems is responsible for the relative stabilities of the products.

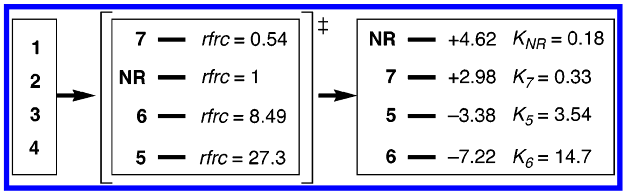


Figure 1. Relative rate constants, energies, and equilibrium constants for the conversion of diene **1** and malelimides **2**−**4** to cycloadducts **5**−**7**, respectively, in $CDCl_3$ at 50 °C. Relative energies are in kilojoules per mole. **NR** represents a control reaction where there is no molecular recognition present. *rfrc* represents the relative forward rate constant for the formation of each cycloadduct and is expressed as $k_{NR}(\text{forward}) = 1$.

Examination (Figure 1) of the relative rate constants for the forward reaction, i.e., the reaction of the diene and dienophile to form the appropriate cycloadduct, reveals a significant kinetic effect. Reducing the tether length from

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(3) (a) Saur, I.; Scopelliti, R.; Severin, K. *Chem.*−*Eur. J.* **2006**, *12*, 1058−1066. (b) Buryak, A.; Severin, K. *Angew. Chem., Int. Ed.* **2005**, *44*, 7935−7938. (c) Severin, K. *Chem.*−*Eur. J.* **2004**, *10*, 2565−2580. (d) Vial, L.; Sanders, J. K. M.; Otto, S. *New J. Chem.* **2005**, *29*, 1001−1003. (e) Corbett, P. T.; Sanders, J. K. M.; Otto, S. *J. Am. Chem. Soc.* **2005**, *127*, 9390−9392. (f) Corbett, P. T.; Tong, L. H.; Sanders, J. K. M.; Otto, S. *J. Am. Chem. Soc.* **2005**, *127*, 8902−8903.

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(5) The reaction medium is important in Diels−Alder reactions generally. (See, for example: Grieco, P. A.; Kauffman, M. D. *J. Org. Chem.* **1999**, *64*, 6041−6048.) The influence of solvent polarity on the recognition effects exposed in this study is potentially a means of fine-tuning the selectivity of these dynamic reactions, and these effects are currently under investigation in our laboratory.

(6) Given the uniformly low conversions in the absence of significant recognition-mediated stabilization of the product ground state, the extent of the control reaction is difficult to measure. To minimize errors, the value of the free energy and the rate constant used to derive the relative rate data in Figure 1 are averages from data derived from reactions between 2-phenylfuran and three different maleimides which bear no recognition sites. (See ref 4.)

three carbons to one carbon should result in an increase in rate on purely entropic grounds[7] as the number of rotors in the system is decreased. The fact that the observed rate enhancement is maximal for the reaction between **1** and **2** suggests that the single-carbon spacer allows the easiest access to the Diels−Alder transition state. However, the single-carbon spacer does not provide sufficient flexibility to permit the retention of two stabilizing hydrogen bonds in the cycloadduct **5**. Therefore, although the forward reaction proceeds faster, the product **5** is less stable ensuring that the reverse reaction is also fast. The overall outcome is a relatively low K_{eq}. When the spacer in the dienophile is increased to two carbons in **3**, the transition state stabilization is diminished, but the increased flexibility in the spacer ensures the retention of two stabilizing hydrogen bonds in the cycloadduct **6**. Therefore, although the forward reaction proceeds somewhat slower, the product is much more stable ensuring that the reverse reaction is also slow. The overall outcome is that $K_6 > K_5$. The three-carbon spacer present in **4** (and, hence, **7**) is too long to permit either transition state stabilization or retention of hydrogen bonds in the cycloadduct.

Having established the basic thermodynamic parameters for these systems, we next studied a reaction in which the maleimides **2** and **3** compete for the diene **1**. We expected that the recognition-mediated rate acceleration observed in the reaction between **1** and **2** should result in **5** being the kinetic product, whereas the greater stability of **6** should result in it being the thermodynamic product of the reaction. This expectation was confirmed (Figure 2) by the results of a kinetic experiment performed in $CDCl_3$ at 50 °C at starting concentrations of [**1**] = [**2**] = [**3**] = 25 mM. Although **5** is, indeed, formed faster than **6**, the greater stability of **6** ensures that, after 180 min, it becomes the major cycloadduct in solution. Simulation of the experimental data using the kinetic parameters derived from our initial experiments affords a pleasingly good fit (solid lines, Figure 2) to the experimental data. At equilibrium (t = 40 h), the observed selectivity (ratio [**6**]/[**5**]) is 3.55 in favor of **6**.

Given the success of the three-component system, we next studied the system in which all four components, the diene **1** and the three maleimides **2**−**4**, were present. Kinetic experiments were performed in a manner identical to that described above in $CDCl_3$ at 50 °C at starting concentrations of [**1**] = [**2**] = [**3**] = [**4**] = 25 mM. In this situation, cycloadduct **5** is once again formed most rapidly, but after 3 h, **6**, as before, becomes the dominant species in solution. In this system, at equilibrium (t = 40 h), the observed selectivities are: [**6**]:[**5**] = 3.49, [**6**]:[**7**] = 35.7, and [**5**]:[**7**] = 10.2.

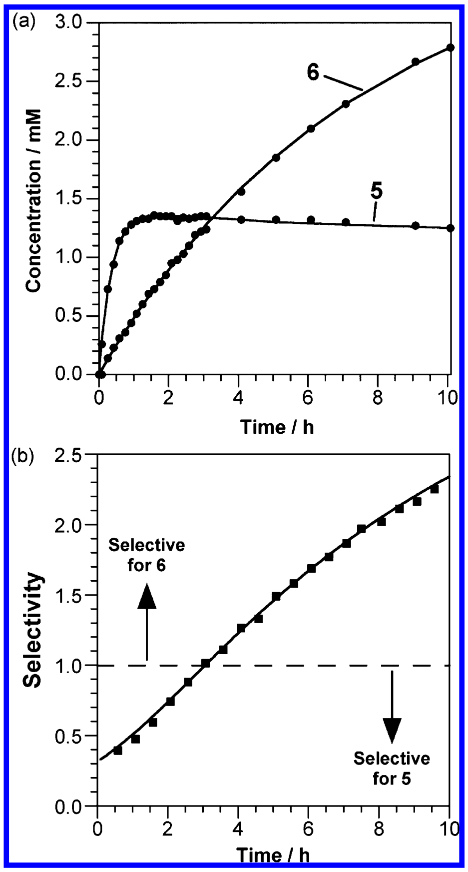

Figure 2. Progress of the three-component dynamic system as a function of time. Experimental conditions: [**1**] = [**2**] = [**3**] = 25 mM; $CDCl_3$; 50 °C. (a) Experimental concentrations of **5** and **6** as a function of time. Experimental points are the filled circles, and the solid lines represent the simulated responses. (b) Selectivity of [**6**]/[**5**] as a function of time. Experimental points are the filled squares and the solid line represents the simulated response.

The selectivities observed in both the three-component and the four-component systems are below those expected purely on the basis of the free-energy differences between the isolated systems. This effect is to be expected because the expression for the final ratio of **6**/**5**, for example, depends not only on K_5 and K_6 but also on the equilibrium concentrations of **2** and **3**. On the basis of this information, we calculated the response of both the two-component and the three-component systems to differing starting concentrations of diene **1**. The results are shown in Figure 3a.

These response curves suggest that maximum selectivity is achieved only at low conversions—at infinitely low conversion, [**6**]/[**5**] ≈ K_6/K_5. Derivation of the equation for the equilibrium constant for this competitive process reveals the reason for the observed drop in selectivity. The overall equilibrium constant K_{eq} depends not only on the K_6/K_5 ratio but also on the ratio of the remaining starting materials remaining at equilibrium, i.e., on [**3**]/[**2**]. This dependence on the equilibrium concentrations of the starting materials results in a compensatory effect in the final position of the equilibrium. At equilibrium, a system with high selectivity

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[7] The association of the diene and the dienophile renders the reaction between them pseudointramolecular. Rate enhancement is therefore expected on purely entropic grounds. See: Page, M. I.; Jencks, W. P. *Proc. Natl. Acad. Sci. U.S.A.* **1971**, *68*, 1678−1683. The calculated rotor increments on moving from **2** to **3** (23 eu) and from **3** to **4** (20 eu) are in the range expected from this model. An alternative view is that, with suitable spacer design, the association of the diene and the dienophile results in a high population of near attack conformations (NACs) for the reaction between them. See: Lightstone, F. C.; Bruice, T. C. *Acc. Chem. Res.* **1999**, *32*, 127−136. In this scenario, it is likely that the [**1·2**] complex has the highest proportion of NACs and is, therefore, accelerated most strongly.

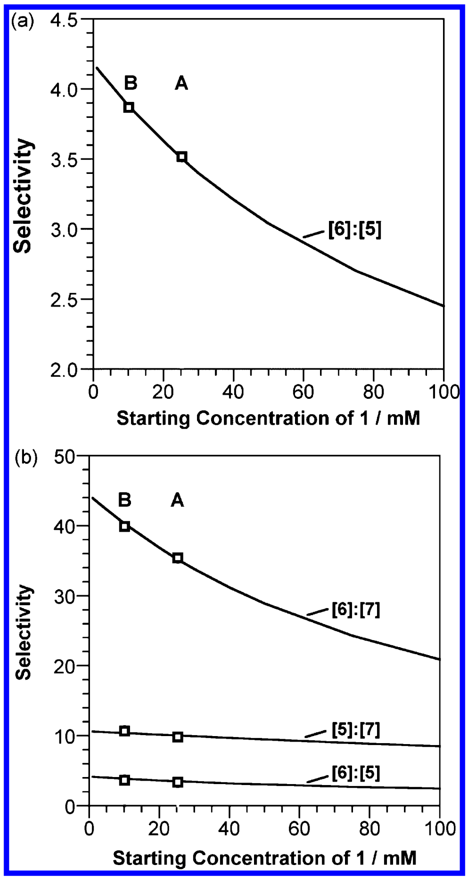

Figure 3. (a) Selectivity in the three-component system as a function of the starting concentration of diene **1**. The simulated response is the solid line. Experimental points (open squares): **A**, [**1**] = [**2**] = [**3**] = 25 mM, and **B**, [**1**] = 10 mM, [**2**] = [**3**] = 25 mM. (b) Selectivity in the four-component system as a function of the starting concentration of diene **1**. The simulated response is the solid line. Experimental points (open squares): **A**, [**1**] = [**2**] = [**3**] = [**4**] = 25 mM, and **B**, [**1**] = 10 mM, [**2**] = [**3**] = [**4**] = 25 mM. All experiments were performed at 50 °C in $CDCl_3$.

should have almost no starting material left which gives rise to the favored product. However, the concentration of the starting material which gives rise to the disfavored product will still be high at equilibrium.

Therefore, in the example here, the higher the K_6/K_5 value, the lower the [**3**]/[**2**] ratio and the bigger the compensation effect at higher conversions. This effect is exacerbated when the concentration of the common component is higher than that of the starting materials. Severin[3c] and Sanders[8] have described similar findings based on simulations of larger libraries with different topologies. In summary, the higher the selectivity for a given product, the more strongly that selectivity will depend on the starting concentrations of the components. This effect can be observed in the results for our three-component system where compound **7** is disfavored strongly. The selectivity of this product with respect to **6**, the strongly favored component, depends (Figure 3) significantly on the starting concentration of the diene **1**.

These effects can be demonstrated experimentally. We repeated the selection experiments for both systems with starting concentrations of the diene **1** of only 10 mM. Under these conditions, the overall conversion within the system will undoubtedly be lower. For the three-component system at equilibrium (t = 40 h), the observed selectivity is: [**6**]/[**5**]) = 3.83. For the four-component system at equilibrium (t = 40 h), the observed selectivities are: [**6**]/[**5**] = 3.82, [**6**]/[**7**] = 40.5, and [**5**]/[**7**] = 10.6. All of these selectivities are higher than those observed when the starting diene concentration is 25 mM. Satisfyingly, the experimentally determined values of the selectivities obtained in these experiments match well (open squares, Figure 3b) with our predictions.

Although the two dynamic systems reported here are very small in terms of the numbers of components and are limited in terms of structural diversity, they afford some important insights into the design and implementation of more complex systems. At the level of thermodynamics, one can expect less selectivity than the absolute energy differences between isolated library components. This diminished expression of free-energy differences is dependent on the starting conditions and may result in the selection of more than one structure by the dynamic library if the energy differences between target components are relatively small. At a kinetic level, preorganization of the precursor reagents is an inevitable consequence of the molecular recognition, which drives the selection process. This preorganization *must* have an effect on how fast the dynamic system reaches equilibrium. In certain circumstances, this may significantly lengthen the time taken to reach the final equilibrium position to an unacceptable level. These observations should help instruct the design of dynamic selection systems generally.

Acknowledgment. We thank the Engineering and Physical Sciences Research Council and the University of St. Andrews for financial support.

Supporting Information Available: Details of the extraction of kinetic and thermodynamic parameters by kinetic simulation and fitting. This material is available free of charge via the Internet at http://pubs.acs.org.

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[8] (a) Corbett, P. T.; Otto, S.; Sanders, J. K. M. *Org. Lett.* **2004**, *6*, 1825−1827. (b) Corbett, P. T.; Otto, S.; Sanders, J. K. M. *Chem.−Eur. J.* **2004**, *10*, 3139−3143.