

**PROGRESS REPORT NO. 4**  
**U.S. Department of Energy**

**An Interim Report In the Format of a Final Report  
To Inform Environmental Management Cleanup Project Managers**

**Managing Tight Binding Receptors for  
New Separations Technologies**

Daryle H. Busch and Richard S. Givens, CoPIs  
Chemistry Department, University of Kansas, Lawrence, KS  
66045

**Project Number: 73850**  
**Grant Number DE-FG07-96ER14708**

Grant Project Officers: Roland Hirsch, Chester Miller

Project Duration: September 15, 1996 to September 14, 2003

# Table of Contents

## PROGRESS REPORT NO. 4

### U.S. Department of Energy

An Interim Report In the Format of a Final Report  
To Inform Environmental Management Cleanup Project Managers

## Managing Tight Binding Receptors for New Separations Technologies

EXECUTIVE SUMMARY .....	1
RESEARCH OBJECTIVES	
The Most Powerful Ligands--An Under Utilized Resource .....	6
A Limiting Molecular Lethargy. ....	7
Exploiting and Overcoming that Molecular Lethargy. ....	7
METHODS AND RESULTS	
Principles of Tight-binding by Ligands. ....	8
Replacing Equilibration with More Rapid Switching Processes .....	9
<i>Design and Synthesis of Generation 1 Switch-binding Ligands</i> .....	9
<i>Validation of Concept: Studies on In Situ Produced Generation 0 Ligand.</i> ...	11
<i>Proof of the Template Switch-binding Concept Using Generation 1 Ligand</i> ..	13
<i>Generation 2 Ligands-- an Untested Second Example</i> .....	18
Switch-release Studies--A replacement for Slow Equilibrium Release .....	19
<i>Photorelease of Metal Ions from Tightly Bound Adducts</i> .....	19
Slow Separations Methodology--an Extreme Case of Biomimicry .....	20
<i>Macroporous Polymers.</i> .....	21
<i>Polymer Synthesis</i> .....	21
<i>Polymer Characterization.</i> .....	22
RELEVANCE, IMPACT AND TECHNOLOGY TRANSFER	
Relevance to Critical DOE Environmental Management Problems .....	23
<i>Deactivation and Decommissioning.</i> .....	24
<i>Environmental Restoration.</i> .....	24
<i>High-level Waste.</i> .....	24
<i>MLLW/TRU and Spent Nuclear Fuel.</i> .....	25
Reducing Costs, Schedules and Risks and Improved Compliance. ....	25
Bridging the Gap between Basic Research and Timely Needs-Driven Applications. ....	25
Impact on possible Users and the Identification of Those Users .....	26
Are Larger Scale Trials Warranted?. ....	26
Improving the Capabilities of Collaborating Scientists .....	26
Advances in Scientific Understanding and Remaining Hurdles to DOE Applications. ...	27
<i>Switch-Binding.</i> .....	27
<i>Switch-Release.</i> .....	27

<i>The Slow Technology</i> .....	27
Interest Expressed by Other Government Agencies and Private Enterprizes .....	28
 PROJECT PRODUCTIVITY	
 PERSONNEL SUPPORTED	
Post Doctoral Research Associates. ....	28
Graduate Research Assistants. ....	28
 PUBLICATIONS. ....	 29
 INTERACTIONS. ....	 29
Participation/presentations .....	29
<i>Daryle H. Busch</i> .....	29
<i>Richard S. Givens</i> .....	31
Consultative and advisory Functions to Other Labs and Agencies .....	32
<i>Daryle H. Busch</i> .....	32
<i>Richard S. Givens</i> .....	33
Collaborations. ....	33
<i>Daryle H. Busch</i> .....	33
<i>Richard S. Givens</i> .....	34
 TRANSITIONS	
Daryle H. Busch. ....	34
Richard S. Givens .....	35
 PATENTS	
Daryle H. Busch. ....	35
Richard S. Givens .....	36
 FUTURE WORK	
(1) Work Leading to Completion of Project .....	36
<i>The New Slow Technololgy--the Soil Poultrice</i> .....	36
<i>Switch-Binding</i> .....	37
<i>Switch-Resease</i> .....	38
(2) Work that Follows Naturally from Having Accomplished Proposal Goals .....	38
 LITERATURE CITED .....	 42
 FEEDBACK. ....	 42
 APPENDICES .....	 42
 QUANTITIES/PACKAGING .....	 42

**PROGRESS REPORT NO. 4**  
**U.S. Department of Energy**

**An Interim Report In the Format of a Final Report  
To Inform Environmental Management Cleanup Project Managers**

**Managing Tight Binding Receptors for New Separations Technologies**

Daryle H. Busch and Richard S. Givens, CoPIs  
Chemistry Department, University of Kansas, Lawrence, KS 66045

Project Number: 73850  
Grant Number DE-FG07-96ER14708  
Grant Project Officers: Roland Hirsch, Chester Miller  
Project Duration: September 15, 1996 through September 14, 2003

**EXECUTIVE SUMMARY**

Ultra-strong Ligands Are Under Utilized Because They Are Slow. When the most challenging problems emerge it is often desirable to respond with the most powerful resources, but a seemingly perverse chemical relationship commonly prevents this in broad areas of chemical applications. When metals are present as accumulated waste or contaminants they are often manipulated by receptor molecules that are called *ligands*. The adduct that is formed is called a complex. Ligands are attached to chelating resins to take metal ions from solutions, and ligands are dissolved in solvents to separate metal ions in solvent extraction processes. In some cases, like solvent extraction, one does not want the ligand to hold on to the metal ion too tightly, but in other cases--for example trying to extract metal ions from very dilute solutions onto chelating resins, the stronger the binding the better, and that exemplifies the perversity. For ligands that achieve their great metal ion affinities through the intricacies of their molecular design--and the affinities can be enormous--big increases in metal ion affinity are accompanied by even bigger decreases in the rates

at which they engulf the metal ion. This project has two major goals that relate to this *molecular lethargy*. The first involves *molecular switches* to replace the very slow equilibration when very strong ligands bind to metal ions. The second proposes a technology to use very strong ligands in such a way that the slowness of the reaction doesn't matter because the system can be given sufficient time to equilibrate.

Replacing Classic Equilibration with Switch-Binding and Release. Macrocycles, ligands that are closed rings, form complexes much more slowly than similar linear ligands, even though both may form the same number of bonds with the metal ion. Further, fused bicyclic molecules, with cavities big enough to accept metal ions into their cage-like interiors, react still more slowly. Switch-binding does the obvious--it converts the ligand into a more constraining form as it binds to the metal ion (from linear to macrocyclic, or from macrocyclic to cage-like). The expected result, if we can figure out how to make it happen, is that the very strong cage-like ligand will bind as fast as a macrocycle, and a macrocycle will form complexes as fast as an otherwise similar linear ligand. Switch release proceeds in exactly the opposite way. A cage-like molecule is designed so that, while it is bound to a metal ion, we can "throw the switch" and convert a macrobicycle into a simple ring, or a macrocycle into a linear molecule.

Photochemical Switch-Release. The molecular switch for *switch-release* had already been identified for other purposes; it is photochemical. One must build into, for example, the macrocycle a functional group that can absorb a photon and cleave a bond, converting the ring into a linear molecule. The linear molecule has ends so it more quickly unwraps from the metal ion than the parent macrocycle. We have learned to make macrobicyclic molecules called "cryptands" that have such a switching group and we have shown that the switch works on the free ligand. Now we must show that it works when a metal ion is bound.

Templating as a Mechanism for Switch-Binding. Years ago, one of us conceived and developed the “template effect” in which a metal ion is used to control the sequence of steps that produce a molecule well suited to having certain of its atoms bound to the metal ion. It is as if the metal ion holds the components in close proximity so they can stitch together. We proposed that linear molecules can be designed that will wrap around metal ions at the velocity of linear ligands, but with reactive groups so placed that they react with themselves and rapidly form macrocycles during the course of metal ion binding.

First Affirmation of the Template Concept. Early on, a crude system provided strong evidence that *template switch binding* works; i.e., a certain linear molecule reacts with a metal ion at its normal high speed, but forms an inert macrocycle in the process. Encouraged by this *Generation 0* system, we persisted and designed and synthesized linear molecules whose ends react with each other to form either polymers or macrocycles. The idea is that the metal ion will act as a template and make the ligand form only the macrocycle. In very detailed studies we have proven the concept. The *Generation 1* ligand forms a macrocyclic complex with nickel(II) ion with a solution yield of, we believe, 100%. We isolated and purified 82%, but we have solution data that says there is only one product.

Kinetic Confirmation of the Switch-Binding Concept. Now all that our results to this point tell us is that the linear molecule forms the desired macrocycle. The critical issue is the velocity of the reaction. Does the macrocycle form at the velocity with which the linear molecule chelates? Or is it slower? We synthesized molecules that are very close relatives of the *Generation 1* ligand except that they cannot react with themselves; therefore they chelate, but they do not form macrocycles. We then explored in detail the kinetics of the reaction involving a *Generation 1* ligand and the reaction involving a non-cyclizing surrogate. The rates of the reactions of the two ligands

are very similar. Both reactions proceeded in two steps--like most chelation reactions. The first step is rapid (millisecond time scale) and usually involves formation of the first bond, or maybe the first two bonds. The second step is ~1000 times slower and results in the fully chelated complex. Thus our interest focused on the second step. Remarkably, Our Generation 1 ligand forms a macrocyclic complex slightly *more rapid* than the formation of the chelate by the ligand that can't cyclize. The step that involves formation of the macrocycle is either faster than, or the same as, the final chelation step. This proves the template switch-binding concept. Switch-binding is a viable concept and can ultimately be used in specific applications. At this point we propose to design and synthesize new bicyclic (cage-like) ligands called catenands that will both switch-bind and switch-release, and show that they can be used to control alkali metal ions like potassium and cesium.

The Soil Poultice--a Model for Bacterial Behavior. The second part of this proposal involves our "soil poultice". This is a technology designed after the manner in which certain microbes extract iron from the soil. The microbe secretes a powerful ligand into the soil. The ligand encounters solids containing iron and slowly binds the iron. The complex then encounters a bacterium and is selectively brought through the membrane into the organism. The genius of this process is (1) the fact that the ligand goes to the mineralized metal--since there isn't enough of the metal ion in solution for the usual equilibrations, and (2) the microbe selectively retrieves the metal complex.

Macroporous Polymer Beads as Microbe Models. We propose the same technology, but replacing the microbe with templated macroporous polymer beads that have been imprinted for the complex of a targeted metal ion; e.g., cesium, cobalt, an actinide, mercury, etc. In practice, beads that have been imprinted for a particular complex of the target metal ion are mixed with the contaminated soil, or applied as a paste to the surface to be treated. The ligand is mixed into the soil

or paste, or perhaps sprayed on. The soil poultice is kept moist for an experimentally determined period of time while the ligand diffuses about and binds the metal ion. Thereafter, the complex is recognized by the resin beads and bound selectively. Recovery of the beads then permits completion of the decontamination process.

The challenges are in the details of the chemistry. We have prepared several polymer samples imprinted for a metal complex that is a good first test case. We have removed the complex from the imprinted polymer and demonstrated rebinding. In these first studies, we find that we can achieve rebinding levels as good as have been reported in the literature. In fact, we are excited about a partially successful experiment in which we added a counter ion to the imprinted polymer, thereby adding coulombic forces to the hydrogen bonding forces usually active in these systems. In that case the polymer morphology was poor, but we still got better rebinding than has been reported for a non-covalent imprint. Clearly we must continue to explore the fundamentals in these systems, but very soon we expect to be studying systems that are immediate precursors to practical applications. Major advantages of the soil poultice concept are (1) the attacking reagent is mobile and yet retrievable; (2) the ligand can be very potent because the ligand can be slow in this slow technology; (3) retrievability can be designed for the specific application; and (4) high levels of selectivity can be expected. Long term, one can envision the products of this concept becoming common consumer products.

**PROGRESS REPORT NO. 4**  
**U.S. Department of Energy**

**An Interim Report In the Format of a Final Report**  
**To Inform Environmental Management Cleanup Project Managers**

**Managing Tight Binding Receptors for New Separations Technologies**

Daryle H. Busch and Richard S. Givens, CoPIs  
Chemistry Department, University of Kansas, Lawrence, KS 66045

Project Number: 73850  
Grant Number DE-FG07-96ER14708  
Grant Project Officers: Roland Hirsch, Chester Miller  
Project Duration: September 15, 1996 through September 14, 2003

**RESEARCH OBJECTIVES**

**The Most Powerful Ligands--An Under Utilized Resource of Extreme Promise**

With the advent of supramolecular chemistry<sup>1</sup> and the growth of nanotechnology<sup>2</sup>, it has become increasingly clear that molecular species and their complex aggregates can be designed and synthesized to achieve increasingly complicated and demanding goals. The present program is an example of this kind of scientific endeavor. The overall purpose of the chemistry described herein is to provide the foundations for the invention of ways in which the most powerful known kinds of ligands, or receptors, can be made useful in such applications as analytical chemistry, separations science and environmental remediation and restoration. *For our purposes, a ligand is a molecular entity with the ability to fasten itself to some range of other molecular entities, forming an assembly, called a complex or coordination entity, within which the original entities remain recognizable.* This program constitutes an investigation into the basic science of coordination chemistry, or supramolecular chemistry, a field that deals with all inter-molecular interactions. The subject cross-cuts all traditional chemical and bordering fields: analytical, bio-, inorganic, organic and physical chemistry, biology, and materials science.

The ultimate powerful ligands can capture their complements, for example, metal ions, in the most competitive of circumstances. For example ultra tight-binding ligands can remove metal ions from mineralized sites, take them away from lesser ligands, and capture metal ions from extremely dilute solutions. In these and other related circumstances ordinary ligands, such as those used in well known separations technologies, are completely ineffective. Just these kinds

of challenges arise in environmental restoration and waste management and they, and many other opportunities for applications, provide compelling arguments for finding ways to apply ultra tight-binding ligands to the management of the metallic elements under many conditions. However, there are major hurdles that must be overcome to do so for otherwise these most interesting substances would already be in heavy use. It must be emphasized that this is a basic science program and that its value lies in the vast array of applications this science underlies.

### **A Limiting Molecular Lethargy**

Historically, the applications of ultra tight-binding ligands has been limited by the slow rates at which their equilibria are established. It is a fundamental fact that the equilibrium constants for the binding of any kind of receptee (e.g., a metal ion) to its complementary receptor (i.e., ligand) vary monotonically with the rates at which the receptee is liberated from the receptee/receptor complex. Consequently, ultra tight-binding ligands, whose equilibrium constants for binding exceed the ordinary by millions or billions of times, will release their complement at least that many millions or billions of times slower. The slowness may be even more lethargic since the rate of binding is also retarded and the equilibrium constant equals the rate of binding divided by the rate of release of the receptee from the receptor. A common result of these very slow processes is that undesirable, more rapid chemical processes may happen and tie up the reactants before they can form the target complex. For example, in the presence of iron(III), an exceptionally strong ligand may react instantly with protons from water, producing hydroxide ion, followed immediately by precipitation of insoluble, relatively unreactive hydrous ferric oxide.

### **Exploiting and Overcoming that Molecular Lethargy**

In order to make best use of tight-binding receptors it is, therefore, necessary to either accommodate any specific methodology to these slow kinetic processes or to find means of accelerating the formation and dissociation rates associated with complexation. This program has aimed at both targets with the goal of producing a legacy of new principles and general methodologies that will provide the foundations for many important applications. This document is organized in terms of those goals. In each case we are specifically concerned with the proof of concept, generalization of the concept, and definition of general methodologies. These methodologies will then become tools for numerous specific applications. The three concept areas are:

(1) *Overcome the Molecular Lethargy by Switch binding of templating ligands*--accelerate the rate of binding by templating a change in structure of the ligand in accompaniment to its binding to the metal. The structural change is from a rapidly reacting, looser binding structure in the free state to a more slowly reacting, more strongly binding structure in the target metal complex.

(2) *Overcome the Molecular Lethargy by Switch release of photo reactive ligands*--accelerate the rate of release of a metal ion from a tight-binding ligand by incorporating

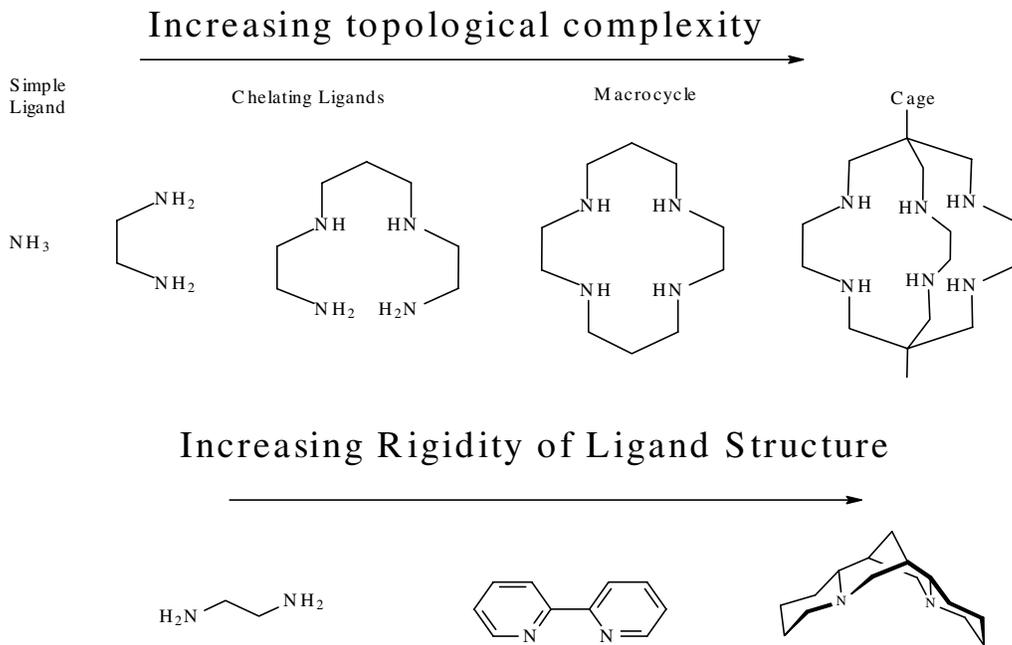
photo release, and possibly other environmentally clean release features, into the ligand structures. Again, a ligand structure change is involved, this time starting with a complex containing a tight-binding ligand structure and ending with a rapidly released, more weakly binding ligand.

(3) *Profit from the Molecular Lethargy Through A slow technology based on imprinted polymers* (e.g., a soil poultice)--design a separations methodology based on the dual selectivity of ligand/metal ion interaction combined with host/guest interaction between the metal-ligand complex and an imprinted polymer. This *soil poultice* is well adapted to slow kinetics of reaction between unmodified tight-binding receptor/receptee pairs.

## METHODS AND RESULTS

### Principles of Tight-binding by Ligands

*Complementarity* and *ligand constraints* determine the stabilities of metal complexes.<sup>4a</sup> Complementarity implies a consonance between the partners that join to form the complex. Complementarity is required for bond type, geometry, and size. For example, in the case of bond type, highly polarizable heavy transition metal ions select in favor of heavy polarizable donor atoms like sulfur or phosphorus over oxygen or nitrogen. In contrast, hard alkali and alkaline earth ions select in favor of the less polarizable or hard oxygen and nitrogen donors. In the case of shape, metal ions whose bonds are directed toward the corners of a square plane are not complementary to ligands that arrange their donor atoms in a tetrahedral fashion. Size--big metal ions require big macrocycles while small metal ions bind most strongly to small macrocyclic ligands. Given good complementarity, the constraints built into the structure of the ligand determine just how strong its complexes will be. This is illustrated in **Figure 1** where the top part shows ligands of increasing topological constraint



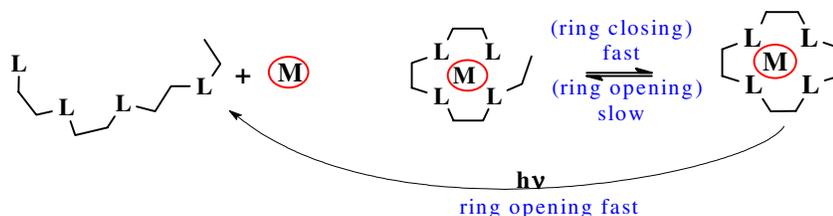
**Figure 1. Constraints that determine the affinities of ligands for a complementary metal ion.**

(simple ligand --> chelate --> macrocycle --> cage) and the bottom shows ligands of decreasing flexibility and increasing rigidity. In both cases, the strength of binding increases from left to right. These principles are applied in our selection and design of ligands for the proposed studies. Again, it is appropriate to emphasize the need for ligands of maximum binding affinity since their function is to react where lesser ligands are dysfunctional. The discussion to follow will first present the results of our switch-binding and switch-release studies and then, our progress toward the soil poultice.

Equilibrium Binding and Release --  
Slow Processes for Macrocycles and Cages



The Much Faster Alternative -- Switch Binding and Release



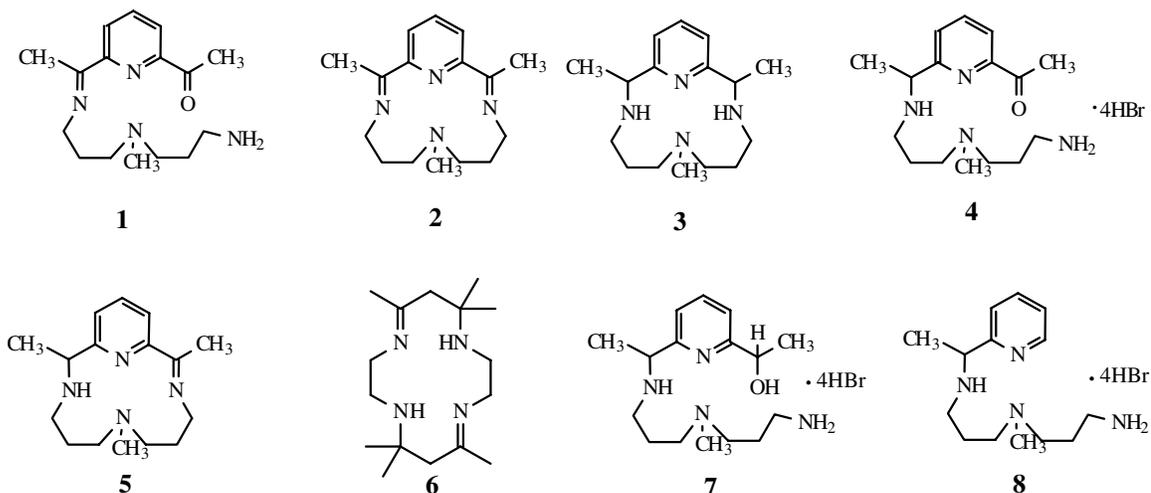
**Figure 2.** Concept of complex formation by switch-binding and -release compared to the familiar equilibration of ligand and metal ion.

**Replacing Equilibration with More Rapid Switching Processes**

**Figure 2** illustrates the contrast between complex formation and dissociation by traditional equilibrium methods with the seminal concept of switch binding and release. The key difference is the change in structure of the ligand that accompanies the complexation and decomplexation processes. In switch binding, a rapid-binding linear tetradentate ligand is changed into a macrocyclic ligand in the complex, and in switch release, a lethargic macrocyclic ligand is changed into a linear ligand to facilitate quick release of the metal ion. It must be emphasized at this point that appropriate examples of ligands for switch-binding were unknown at the time this program began and that only a few examples of switch-release ligands had been reported.

## Switch-Binding Studies--A Replacement for Slow Equilibrium Binding

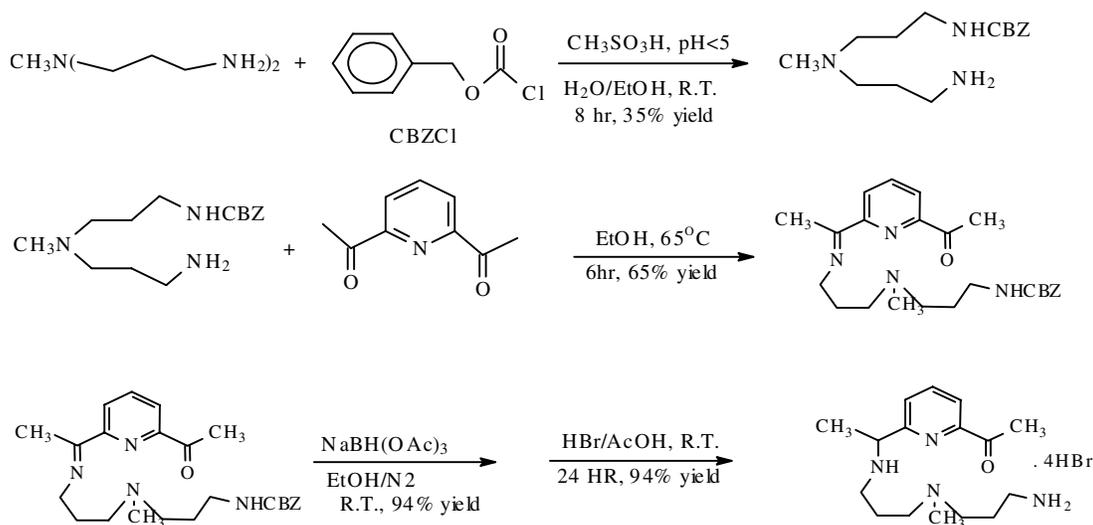
**Design and Synthesis of Generation 1 Switch-Binding Ligands.** In setting out to establish a new principle within so highly developed a field as coordination chemistry and its near neighbor, the broad realm of organic chemistry, it is essential to select systems that (1) have reasonable



probabilities of success, and (2) that are attainable with reasonable effort, and many factors determine both considerations. In early studies to be described below, it was proposed that the linear tetradentate ligand of **Structure 1** is an intermediate in the synthetic template reaction that yields the nickel(II) complex of **Structure 2**. Hydrogenation of the imine bonds yields the complex having both imine groups reduced, **Structure 3**, and the monoimine derivative. Because of this background,<sup>5</sup> the molecule of hypothetical linear tetradentate molecule having **Structure 4** was chosen for the initial investigations and is denoted here as the *Generation 1 ligand*. The fact that the macrocycle expected to form from it, **Structure 5**, has been well characterized in its nickel(II) complex was a major factor in the decision. Further, nickel(II) complexes were considered to be ideal for these studies because they often form in synthetic template reactions with a minimum of side reactions, such as redox processes. Although synthesis of the corresponding tetradentate monomethine complex with Ni(II) was previously reported, the literature preparation did not give a significant yield in our hands and, further, led to a mixture of products with varying numbers of azomethine linkages. Consequently a more traditional, alternative synthesis was designed, involving the initial protection

of one end the diamine (**Scheme 1**).<sup>6</sup> This ligand was successfully synthesized in reliable but modest overall yield and then used for further studies. Detailed studies of complexation between this Generation 1 ligand and nickel(II) are described in a section to follow. In order to have at hand data acquired under identical conditions on the kinetics of binding of a typical macrocycle, the ligand of **Structure 6** was studied. Also, to provide appropriate comparisons and for practical purposes explained below, the related linear tetradentate ligands having **Structures 7** and **8** were critical to this program. These new compounds were also synthesized using very similar procedures to those

developed to produce the Generation 1 ligand.



Scheme 1

**Validation of Concept: Studies on *In Situ* Produced Generation 0 Ligand, L<sup>1</sup> (Structure 1).** The design and development of a reliable synthetic source of Generation 1 ligands took some time, and prudence suggested that, prior to achievement of that goal, we should proceed with the development of the measurements that would eventually permit us to use our hard-won and very special ligands to prove the basic switch-binding concept. Further we saw the possibility of early validation of the concept through manipulation of the nickel(II) templating conditions used in the synthesis of a well known Schiff base macrocycle ligand. Unsaturated polyaza macrocycles containing first row transition metal templates have been prominent in the advancement of macrocyclic chemistry for nearly four decades<sup>7</sup> In particular, the evolution of cyclic imines produced by condensation of di-, tri-, tetra- and pentaamines with 2,6-diformyl (DFP) and -diacetylpyridine (DAP) has been extensive and well documented in the literature<sup>8</sup> In typical syntheses, the versatile pyridine precursors are held to the template ion by the nitrogen donor atom, enabling nucleophilic attack at the carbonyl carbons by the primary diamine moieties to complete the macrocycle. Among the most representative examples in this class of macrocycles is the tetramine ligand, L<sup>2</sup> (**Structure 2**), obtained by an *in situ* reaction of DAP with 3,3'-diamino-*N*-methylpropylamine in the presence of metals such as nickel(II) and copper(II)<sup>5</sup>. Despite the considerable number of reported preparations of L<sup>2</sup>, the mechanistic understanding of macrocycle formation by this *in situ* route was speculative and entirely qualitative. We set out to quantitate the understanding of this system.

Initially the ternary system consisting of NiCl<sub>2</sub>·6H<sub>2</sub>O, DAP, and 3,3'-diamino-*N*-methylpropylamine was studied spectroscopically in ethanol under conditions typical of preparative experiments<sup>9</sup>. The multiplicity of concurrent processes that occur upon mixing the three components simultaneously is clearly displayed by the complicated time-dependent behavior of the corresponding UV-vis spectral traces (**Figure 3a**). As expected, the data are distinctly

uncharacteristic of simple kinetic systems, since at least three ligands are present to compete for the nickel(II) ion--and two of those molecules will react with each other to give a variety of products.

For years, conventional wisdom associated with the syntheses of Schiff base complexes by template reactions has involved a period of pre-equilibration of the ligand components prior to addition of the metal ion<sup>9</sup>. During this time interval, amine and carbonyl compounds condense, decreasing the number of ligand species in solution. This lead was followed through spectroscopic studies in which ethanolic equimolar solutions of DAP and the triamine were equilibrated prior to mixing with the nickel(II) solution. The successful simplification of the system is evidenced from the visible spectral overlays in **Figure 3b**. NMR and IR studies<sup>10</sup> on the pre-equilibrated solution over a range of mixing periods (15 min to 4 h) supported the view that the equilibrium in **Scheme 2** is a dominant factor in this reaction.

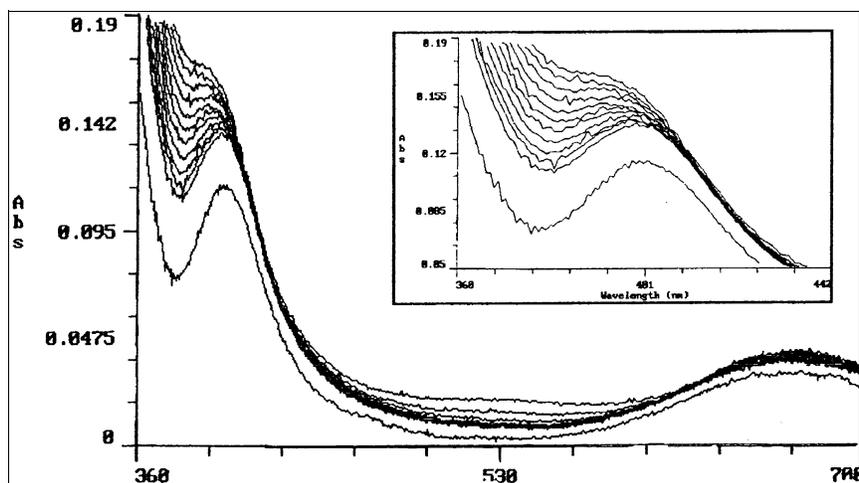


Figure 3a) Visible spectral changes accompanying the reaction of nickel(II) with equimolar amounts of DAP and 3,3'-diamino-*N*-methyldipropylamine in ethanol at 25°C.

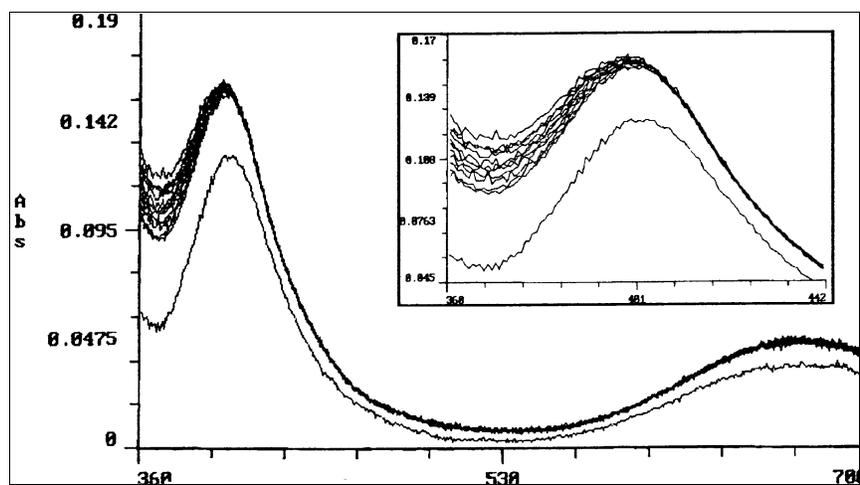
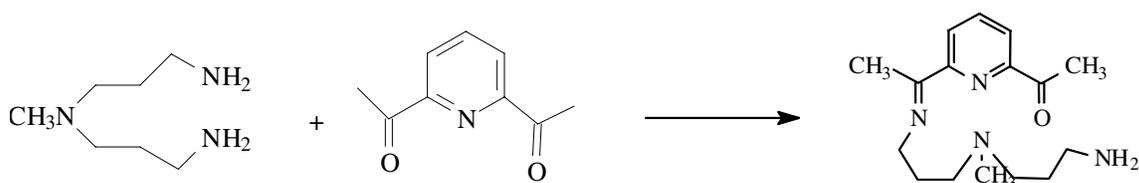


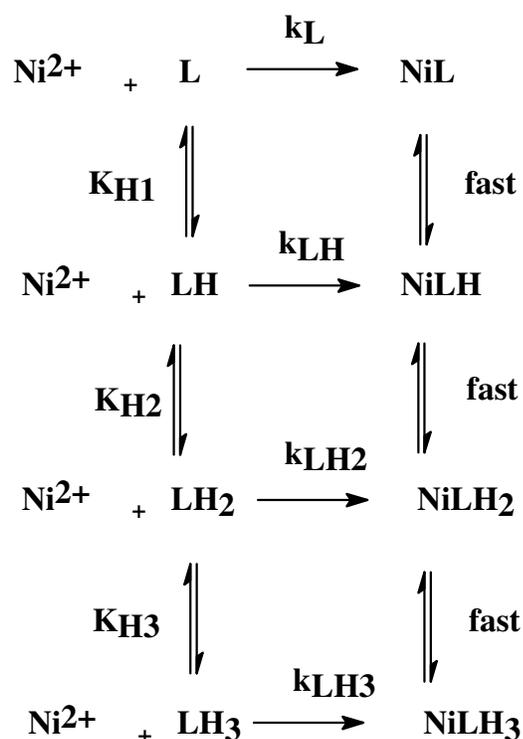
Figure 3b) Visible spectral changes accompanying the reaction of nickel(II) with a pre-equilibrated solution of equimolar DAP and 3,3'-diamino-*N*-methyldipropylamine, L<sup>1</sup>, in ethanol at 25°C.



**Scheme 2** Pre-equilibration of DAP and 3,3'-diamino-*N*-methylpropylamine in ethanol.

The rate behavior during  $[\text{NiL}^{2+}]^{2+}$  complex formation between the “aged” DAP/triamine solution and nickel(II) can be seen in the spectrophotometric scans recorded as a function of time (**Figure 3 b**). The spectral overlays display a monotonic increase in absorbance over time, obeying a single exponential function, suggesting that a single product dominates the reaction. Most decisively, the final spectrum is quantitatively identical to that of an authentic sample of  $[\text{NiL}^{2+}]^{2+}$  ( $\lambda_{\text{max}} = 398$ ,  $\epsilon = 57$ ). The positive ion FAB-MS taken on solutions following complexation experiments shows, a single nickel complex with  $m/z = 330$ , corresponding to the nickel(II) complex with  $\text{L}^2$  (**Structure 2**).

Kinetic measurements with nickel(II) as the templating ion and equimolar amounts of equilibrated DAP and triamine [neat in ethanol with  $\text{I} = 0.1 \text{ M}$  ( $\text{KNO}_3$ ) at  $25.0 \pm 0.1^\circ\text{C}$ ] showed that the rate of  $[\text{NiL}^{2+}]^{2+}$  formation is first order with respect to the concentration of the ligand mixture on two time scales both fast ( $k_1 = 137(\pm 7)\text{M}^{-1}\text{s}^{-1}$ ; by stopped flow spectroscopy) and slow ( $k_2 = 0.17(\pm 0.01)\text{M}^{-1}\text{s}^{-1}$ ; by traditional UV-vis spectroscopy). This 2-step binding process is typical of the complexation behavior of polydentate chelating agents. The more rapid rate typically represents fast formation of the first bond, or two, between the ligand and the metal ion, and the second slow step completes the polydentate chelation. In all cases the rate processes conform to single exponential equations, representing monophasic, pseudo first order reactions. Therefore the only intermediate is that formed in the “ $k_1$ ” step. Further, since this reaction pattern is characteristic of many chelation processes and since the macrocycle is formed without the appearance of any second intermediate, it can be concluded that the second step, not only involves the formation of the remaining bonds between the original linear ligand, but also ring closure to form the macrocycle. Since others have shown that macrocycles commonly bind with  $k_2$  values from hundreds to thousands of times slower, the relative rapid value of  $k_2 = 0.17 \text{ M}^{-1}\text{s}^{-1}$  strongly supports the view that this reaction proceeds at an accelerated rate because of switch binding. We have added an important example by studying the binding to nickel(II) of the well known macrocycle having structure 6,  $\text{L}^6$ ;  $k_1 = 170 \text{ M}^{-1}\text{s}^{-1}$ , very similar to that observed for our *Generation 0* switch-binding ligand ( $137 \text{ M}^{-1}\text{s}^{-1}$ ). But for the preformed macrocycle  $k_2 = 1.1(\pm 0.1) \times 10^{-3} \text{ M}^{-1}\text{s}^{-1}$  is much slower than the second step for the switch-binding ligand. These relationships greatly encouraged our on-going development of other ligands whose structures would remove all doubt about the benefits of switch-binding if they show the same behavior. Because of the uncertainties with regard to the species in solution, this study is considered supportive, but not a definitive proof of the concept of template switch-binding.



**Proof of the Template Switch-Binding Concept Using the Generation 1 Ligand.** New systems produce new challenges and the first major challenge confronted after the successful synthesis

of the Generation 1 ligands was proving precisely what species exist in its aqueous solutions under various pH conditions. The ligand protonation equilibria and competing complex formation steps for a tetradentate ligand having three ionizable protons are summarized in **Figure 4**. Because in the absence of the templating metal ion, the ligand  $L^4$  reacts with itself to form a large number of linear and cyclic oligomers, which differ with pH, it is almost impossible to determine the  $pK_a$  values for  $L^4$  directly. Therefore ligands  $L^7$  and  $L^8$  were designed to simulate the  $L^4$  (i.e., have very nearly the same  $pK_a$  values) without the frustrating ability to react with themselves. The  $pK_a$  values were determined for  $L^7$  and  $L^8$  and the average of their values were used as approximate values of  $pK_a$  for  $L^4$ . The constants were then used to calculate the speciation of the solutions of the hypothetical free ligand in order to provide foundations for deducing which levels of

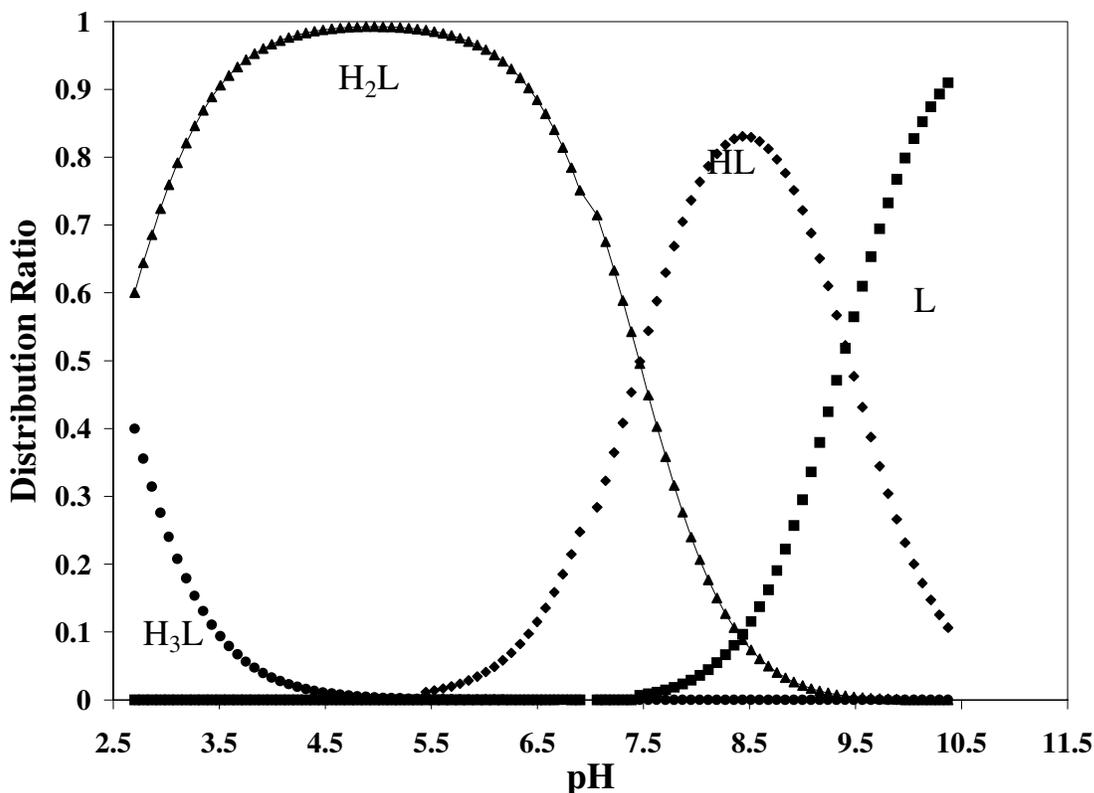
protonated ligands are important in the reaction with the nickel(II) ion. In turn this facilitates precise interpretation of data on the kinetics of the binding reaction.

**Figure 4.** Multiple equilibria in complexation process

**Table 1.** Protonation constants obtained at 25.0(±0.1)°C and I = 0.1 M (KNO<sub>3</sub>) for  $L^4$ ,  $L^7$ , and  $L^8$  together with literature values for related macrocyclic ligands.

Ligand	log $K_1$	log $K_2$	log $K_3$	log $K_4$
$L^4$ <sup>(1)</sup>	9.40	7.23	2.84	2.14
$L^7$	9.44(6)	7.07(4)	3.15(7)	2.28(1)
$L^8$	9.37(1)	7.38(1)	2.53(1)	<2.2
py[14]aneN <sub>4</sub> <sup>(2),(3)</sup>	9.92(1)	8.56(3)	4.66(4)	<1
<i>N</i> -Mepy[14]aneN <sub>4</sub> <sup>(2),(4)</sup>	9.74(4)	8.67(5)	4.67(6)	<1

<sup>(1)</sup>Estimated values; <sup>(2)</sup>Costa, J.; Delgado, R. *Inorg. Chem.* **1993**, 32, 5257.; <sup>(3)</sup>py[14]aneN<sub>4</sub> = 3,7,11,17-tetraazabicyclo[11.3.1]heptadeca-1(17),13,15-triene; <sup>(4)</sup>*N*-Mepy[14]aneN<sub>4</sub> = 7-methyl-3,7,11,17-tetraazabicyclo[11.3.1]heptadeca-1(17),13,15-triene.

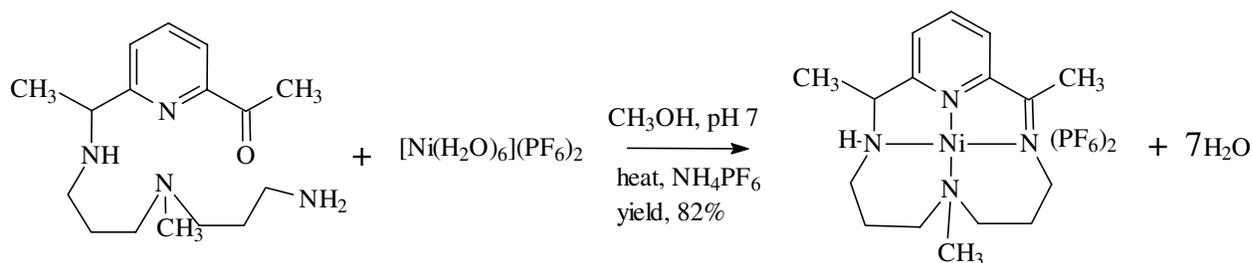


**Figure 5** Speciation diagram for  $L^8$  as a function of pH.

From the speciation curves (**Figure 5**) it is clear that the doubly protonated ligand is the dominant species present below pH 7, while the monoprotonated species is prevalent in moderately basic media. These facts, when combined with the results of rate studies, show that the monoprotonated species is the main reactant because  $LH^+$  reacts with the divalent metal ion almost a thousand times faster than does  $LH_2^{2+}$ . These speciation results are useful in the interpretation of the results of kinetic studies.

The next critical experiments were conducted to establish the nature of the products and the extent to which the desired macrocyclic complex is formed in solution under conditions suitable for mechanistic studies. **Figure 6** represents the ring closure reaction and the conditions used to prove that the main product of the reaction of  $L^4$  with nickel(II) in solution is the macrocyclic complex  $Ni(L^5)^{2+}$ . In the reaction between stoichiometric amounts of  $L^4$  and nickel(II), an absorption peak increases steadily at 468 nm ( $\epsilon = 88$ ) as expected for the formation of the macrocyclic complex. That species is identified in solution by FAB mass spectrometry (in water/NBA) that shows a nickel(II) complex with  $m/z = 331$ , again as expected for  $Ni(L^5)^{2+}$ . The addition of ammonium hexafluorophosphate to the equilibrated solution led to isolation of  $[Ni(L^5)](PF_6)_2$  in 82% yield. The composition of that product agrees. Anal. Calcd. For  $[Ni(C_{16}N_4H_{26})](PF_6)_2$ : C, 30.94; H, 3.87; N, 9.02. Found: C, 31.05; H, 3.95; N, 8.87. Infrared spectrum also agrees. Indeed the ring closure

occurs upon complex formation and this appears to be the only process occurring in the solution.



**Figure 6.** Stoichiometric formation of the macrocyclic complex in solution from the switch-binding ligand and metal salt. Yield of isolated and purified product, 82%.

The ultimate evaluation of Generation 1 ligands awaited kinetic studies to show if this ligand, which does indeed form a very stable macrocyclic complex, forms that complex significantly more rapidly than the macrocycle itself could bind. The kinetics of ligand binding were studied at selected pH values and over a range of ligand concentrations at fixed metal ion concentration, with the ligand always in ten-fold or greater excess. The rate constants for the kinetics of formation of the macrocyclic complex,  $\text{Ni}(\text{L}^5)^{2+}$ , and for its surrogate,  $\text{Ni}(\text{L}^8)^{2+}$ , are given in **Tables 2-5**. The data for the surrogate are critically important because that linear tetradentate ligand cannot ring-close to form a macrocycle in the complex. A basic question is “will the kinetics distinguish between the two ligands?” If the proximity effects associated with a nicely complementary ligand and metal ion work favorably, the ring closing step could be so fast that the behavior of the two systems could be quite similar. In any event, rate processes should be observed in two time domains, as has been pointed out above for other systems. This is indeed the case.

**Table 2** Values of  $k_{1, \text{obs}}$  ( $\text{s}^{-1}$ ) for the complexation of  $\text{L}^4$  to nickel(II) (1.0 mM) from 0.003 to 4.8 s, at varying levels of pH and  $\text{L}^4$  concentration.

$[\text{L}^4]$ , M	pH 3.8	pH 5.5	pH 7.2	pH 8.5
0.01	0.0151 ( $\pm 0.0006$ )	0.0184 ( $\pm 0.0016$ )	2.22 ( $\pm 0.21$ )	2.36 ( $\pm 0.06$ )
0.025	0.0356 ( $\pm 0.0018$ )	0.0396 ( $\pm 0.0041$ )	5.12 ( $\pm 0.43$ )	5.67 ( $\pm 0.39$ )
0.05	0.0792 ( $\pm 0.0030$ )	0.0910 ( $\pm 0.0015$ )	9.87 ( $\pm 0.86$ )	11.56 ( $\pm 0.69$ )
0.075	0.1124 ( $\pm 0.0025$ )	0.1128 ( $\pm 0.0148$ )	15.42 ( $\pm 1.23$ )	17.54 ( $\pm 0.79$ )
0.1	0.1532 ( $\pm 0.0064$ )	0.2137 ( $\pm 0.0177$ )	21.37 ( $\pm 1.96$ )	22.37 ( $\pm 1.97$ )

**Tables 2 and 3** give the constants for the rapid reaction (.003 to 4.8sec) and **Tables 4 and 5** give those for the slower followup reaction (4.8 to 522 sec). The reported rate constants are averages of triplicate measurements and are derived from fitting spectral absorbance changes over time with algorithms that account for changes at all wavelengths examined (from 300 to 700 nm); numbers in parentheses are standard deviation values.

**Table 3** Values of  $k_{1,obs}$  ( $s^{-1}$ ) for the complexation of  $L^8$  to nickel(II) (1.0 mM) from 0.003 to 4.8 s, at varying levels of pH and  $L^8$  concentration.

$[L^8], M$	pH 3.8	pH 5.5	pH 7.2	pH 8.5
	$k_{2,obs} \times 10^3, s^{-1}$			
0.01	0.0155 ( $\pm 0.0010$ )	0.0202 ( $\pm 0.0019$ )	2.39 ( $\pm 0.20$ )	6.40 ( $\pm 0.61$ )
0.025	0.0509 ( $\pm 0.0020$ )	0.0542 ( $\pm 0.0037$ )	8.20 ( $\pm 0.73$ )	14.8 ( $\pm 1.2$ )
0.05	0.0945 ( $\pm 0.0029$ )	0.101 ( $\pm 0.011$ )	11.79 ( $\pm 0.96$ )	19.9 ( $\pm 1.0$ )
0.075	0.154 ( $\pm 0.006$ )	0.151 ( $\pm 0.015$ )	15.3 ( $\pm 1.3$ )	27.8 ( $\pm 1.9$ )
0.1	0.196 ( $\pm 0.007$ )	0.224 ( $\pm 0.018$ )	21.3 ( $\pm 1.8$ )	36.9 ( $\pm 2.5$ )

**Table 4** Values of  $k_{2,obs}$  ( $s^{-1}$ )<sup>(1)</sup> for the complexation of  $L^4$  to nickel(II) (1.0 mM) from 4.8 s to 522 s, at varying levels of pH and  $L^4$  concentration.<sup>(2)</sup>

$[L^4], M$	pH 3.8	pH 5.5	pH 7.2	pH 8.5
	$k_{2,obs} \times 10^3, s^{-1}$			
0.01	2.01 ( $\pm 0.20$ )	3.94 ( $\pm 0.35$ )	16.71 ( $\pm 1.21$ )	19.15 ( $\pm 1.96$ )
0.025	4.77 ( $\pm 0.17$ )	7.75 ( $\pm 0.77$ )	16.55 ( $\pm 1.43$ )	22.80 ( $\pm 1.95$ )
0.05	9.72 ( $\pm 0.17$ )	9.16 ( $\pm 0.95$ )	16.92 ( $\pm 0.96$ )	20.55 ( $\pm 2.41$ )
0.075	15.23 ( $\pm 0.34$ )	14.46 ( $\pm 0.62$ )	17.83 ( $\pm 1.23$ )	21.09 ( $\pm 2.51$ )
0.1	18.33 ( $\pm 0.40$ )	21.37 ( $\pm 1.78$ )	16.21 ( $\pm 1.16$ )	21.41 ( $\pm 2.42$ )

**Table 5** Values of  $k_{2,obs}$  ( $s^{-1}$ ) for the complexation of  $L^8$  to nickel(II) (1.0 mM) from 4.8 to 522 s,

$[L^8], M$	pH 3.8	pH 5.5	pH 7.2	pH 8.5
	$k_{2,obs} \times 10^3, s^{-1}$			
0.01	2.44 ( $\pm 0.18$ )	3.98 ( $\pm 0.25$ )	49.5 ( $\pm 4.2$ )	87.5 ( $\pm 3.1$ )
0.025	6.28 ( $\pm 0.70$ )	8.56 ( $\pm 0.83$ )	47.9 ( $\pm 3.4$ )	91.9 ( $\pm 8.0$ )

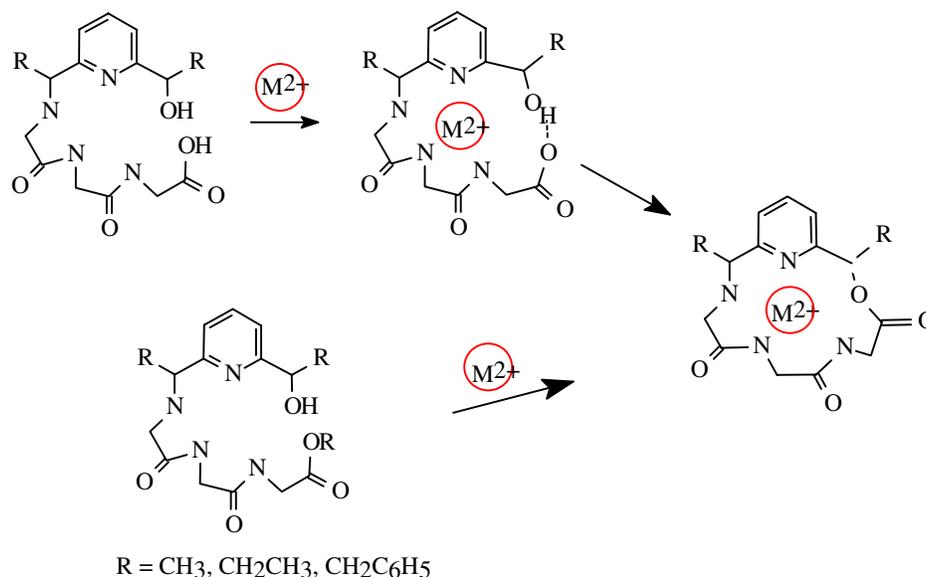
0.05	8.58 ( $\pm 0.43$ )	17.2 ( $\pm 1.4$ )	51.7 ( $\pm 2.7$ )	85.8 ( $\pm 4.5$ )
0.075	16.0 ( $\pm 1.2$ )	22.0 ( $\pm 1.0$ )	48.2 ( $\pm 3.9$ )	95.3 ( $\pm 8.1$ )
0.1	19.0 ( $\pm 1.0$ )	26.3 ( $\pm 1.3$ )	48.2 ( $\pm 3.5$ )	93.5 ( $\pm 8.6$ )

The observation of the immediate color change to bright yellow upon mixing the solutions indicates the initial metal to ligand complexation within the 0.003 to 4.8 s time frame,  $k_{1\text{obs}}$ . For both  $L^4$  and  $L^8$  the rate constants show a first order dependence on the concentration of free ligand and a strong pH dependence, but the most striking result is that the rate constants are very similar numerically. Since this first rate determined process involves formation of only the first, and possibly second, metal-ligand bond(s), it is not surprising that the two very similar linear tetradentate ligands react at essentially the same rates. The behavior of the rate constants associated with the second rate process is more revealing. The slower growth of peaks in the absorption spectra measures the binding of the remaining ligating atoms within the coordination sphere, and is rate determining for the assembly of the final complex,  $[\text{NiL}^8]^{2+}$ , from the reaction of  $L^8$  with nickel(II), and  $[\text{NiL}^5]^{2+}$  from the reaction of  $L^4$  with nickel(II). This process occurs on a slower scale, from 4.8 to 522 s. Again, the rate constants are strongly pH dependent, but they are dependent on the concentration of ligand only at low pH. This pH sensitivity of the ligand dependence is quantitatively consistent with sequential reactions in which the first step is a preequilibrium for the second step ( $k_{2\text{obs}}$ ), for the scenarios in which the first step is retarded by acid. This is totally consistent with the expected chemistry and supports the interpretations given here. From this and the earlier studies on this project that proved the formation of the macrocyclic complex,  $[\text{NiL}^5]^{2+}$ , from the reaction of the ligand  $L^4$  with nickel(II) ion, we conclude the macrocycle formation proceeds at a rate that is characteristic of a linear tetradentate ligand even though a macrocyclic complex is the product. That is, switch-binding has been successful; this new kind of process has greatly accelerated the binding of the ligand in the form of a macrocycle. This is proof of concept for Objective 1 of this program.

Extensive data treatment quantitated the results and interest focused on the second process, since the first, which is quite fast for both ligand types  $L^8$  and  $L^4$ , only involves initial bond formation and probably is not highly sensitive to structure. Analysis in terms of the rapid process as a preequilibrium, gives values for  $k_2$  of  $3.1(\pm 0.2) \times 10^{-2} \text{s}^{-1}$  for  $L^8$  and  $6.1(\pm 0.5) \times 10^{-2} \text{s}^{-1}$  for  $L^4$ . Thus the kinetic behaviors of the two systems are totally parallel and this can only mean that the actual macrocyclization step is kinetically inconsequential. That is, the first and second stages of binding of the linear chelate reactants are rate determining and the ring closure to form the macrocycle  $L^5$  from  $L^4$  is too fast to be measured by these techniques. *Indeed, we have proof of concept; tight-binding macrocyclic complexes can be formed at the relatively more rapid rates characteristic of linear chelating agents.*

**Generation 2 Ligands--an untested second example.** Examples of a second family of

ligands that should be capable of switch-binding have been prepared and are expected to undergo metal ion template directed ring closures to form macrocyclic complexes as shown in **Figure 7**. The use of lactone formation as the macrocyclization step provides a more easily controlled system than that based on Generation 1 ligands (Schiff base formation). This is a subject for future research.



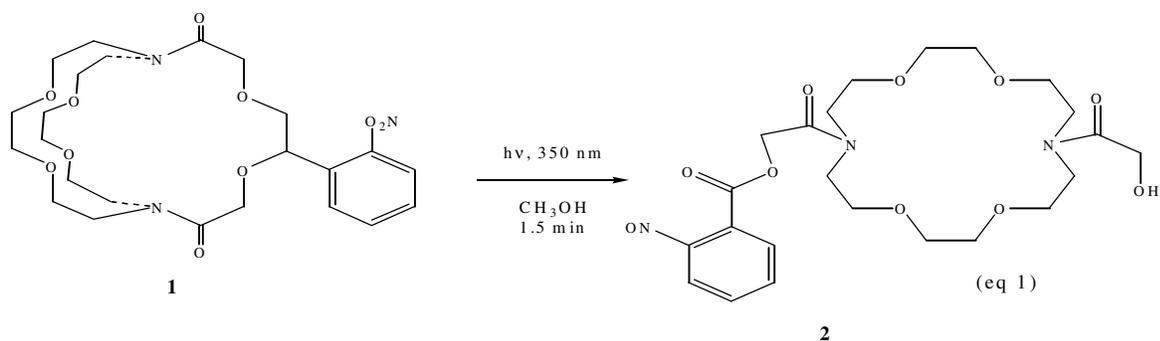
**Figure 7**

### Switch-Release Studies--A Replacement for Slow Equilibrium Release

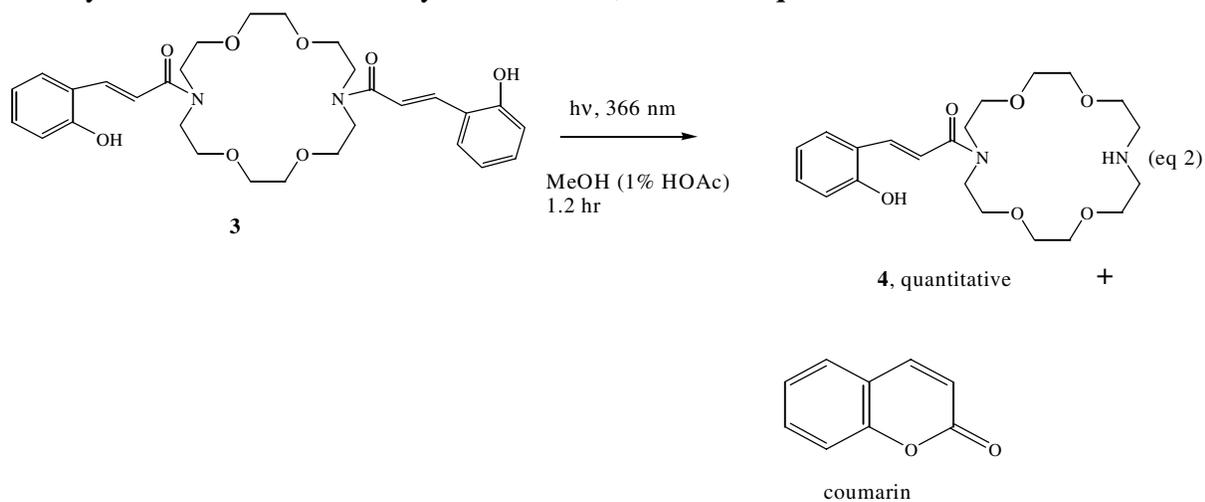
The photorelease of such 'caged' reactants as phosphates, ATP, and amino acids has been well demonstrated and applied to such important tasks as mapping neuronal pathways in mammalian brain slices. Two general reactions are particularly appropriate to the design of modified ligands to facilitate very rapid cleavage of ligand linkages, thereby providing highly accelerated release of the targeted species. These involve, respectively, benzoyl and *o*-nitrophenyl substituents attached to benzylic carbon atoms that are adjacent to heteroatoms, especially oxygen. **Equation 1** shows how the photoreaction could convert a macrobicyclic cryptate ligand (known to release metal ions very slowly) into a much more labile lariat crown ether.

**Photorelease of Metal Ions from Tightly Bound Adducts.** A cryptand capable of rapid release of tightly bound ions by photolytic fragmentation of one of the bridging rings has been synthesized. The synthesis of an *o*-nitrobenzyl substituted cryptand<sup>11</sup> has been achieved and the preliminary photochemistry examined.<sup>12</sup> Thus, cryptand **1** was obtained by modifications of the reaction sequence shown in our original proposal (X = H, a = b = 1) in an overall yield of 19% from

*o*-nitrobenzaldehyde. The cryptand diamide **1** in **equation 1** showed a  $\lambda_{\max} = 265$  nm,  $\epsilon = 5,800$ , which tails to 400 nm. The yield of two of the steps can be further optimized improving the yield to 38%. Upon photolysis, **1** was converted by ring-opening to the weaker binding macrocyclic ligand **2** in quantitative yield (**equation 1**) following the typical photochemical sequence of *o*-nitrobenzyl derivatives outlined in the original proposal. Further studies will determine the photoefficiency for this transformation, the effects of different chelated ions on the nature of the reaction products, and on the dissociation constants.



Exploratory work on cinnamoyl macrocycles has also been undertaken. As a prototype, we have synthesized the di-cinnamoyl substituted **3**, shown in **equation 2**.



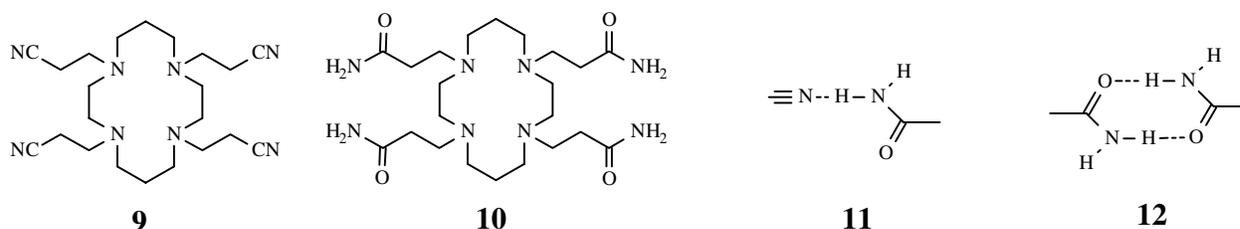
Reaction of two equivalents of *o*-hydroxycinnamic acid with 1,4,10,13-tetraoxo-1,16-diazacyclooctadecane in THF:CH<sub>2</sub>Cl<sub>2</sub> (1:1) with triethylamine and benzotriazole-1-yl-oxy-tris-(dimethylamino) phosphonium hexafluorophosphate (BOP, Castro's reagent) gave **3** in 57% yield (not optimized,  $\lambda_{\max} 325$  nm,  $\epsilon = 30,300$  tailing to 360 nm). Irradiation of **3** at 366 nm in methanol containing 1% HOAc gave an immediate blue fluorescent solution due to the formation of coumarin ( $\lambda_{\max} 225$  nm). To obtain complete conversion to the free macrocycle and coumarin, the irradiation was allowed to continue overnight.

### Slow Separations Methodology--an Extreme Case of Biomimicry.

The research reported in this section is based on the belief the behavior of microbes might well be copied in treating certain difficult cases of soil contamination. Microbes use some of the strongest binding receptors known today to extract iron from the soil.<sup>13</sup> The ligand is excreted through the cell membrane of the microorganism; it diffuses into the soil and slowly dissolves iron from the abundant oxide; and the complex then diffuses back into the cell. The points of immediate interest are the fact that the ligand diffuses away from its parent organism, binds to the metal, and then returns. This and work with macroporous templated polymers<sup>14</sup> provides a remarkable possibility that inspired these investigations.

This design proposed that a macroporous polymer be templated for the complex formed between a particular target ion and a tight-binding ligand, and that the polymer be used as a moderately selective host for the receptor/target ion adduct. One might then coat the polymer on magnetic beads, as has been done by others,<sup>15</sup> mix the particles into the soil and irrigate with water containing dissolved receptor (ligand). The ligand would then diffuse to the pollutant and slowly bind it. Subsequently, the adduct would diffuse to the template-polymer clad magnetic particles, where it would be recognized and bound. The free ligand would not be recognized by the polymer particles, and only its complex with the target ion would be bound. Recovery of the particles would then complete the decontamination process.

**Macroporous Polymers.** For proof of concept, i.e., the feasibility of recapturing the entire metal complex for which the macroporous polymer had been templated, we chose the nickel(II) ion



because of the certainty of its complex chemistry and because nickel(II) is a reasonable substitute for cobalt(II), a metal with a substantial history of contamination problems. The ligand systems are cyclam derivatives having functionalized pendant arms (**Structures 9** and **10**).<sup>16</sup> Complexes of ligands **9** and **10** were selected to provide a comparison in which **9** can only interact with the polymer via a single 2-center 1-H hydrogen bond (**Structure 11**) whereas Complex **6** can interact by forming more stable 4-center 2-H hydrogen bonds (**Structure 12**).

**Polymer synthesis.** It has been demonstrated that macroporous polymers based on hydrogen bonded amide groups show better recognition properties than those with the more commonly used carboxyl groups.<sup>17</sup> Accordingly, our polymer samples were prepared with acrylamide as the main component. To produce highly crosslinked polymers with mesopores (20-500Å diameter), ethylene glycol dimethacrylate, EGDMA, was used as a two point crosslinking monomer for all samples

except **P4**, which used a four-point crosslinker. Polymers **P1** and **P2a** were imprinted with the perchlorate salts of complexes **9** and **10**, respectively. Polymer **2b** used the vinylsulfonate salt of complex **10**, to add electrostatic attractions to the rebinding site. A control polymer (**P3**), with no imprint but active monomer, was synthesized to provide randomly arranged amide groups in randomly sized cavities. Acetonitrile was used as the sole porogen for polymers **P1**, **P2a** and **P3**, but for **P2b** a 1:1 mixture of acetonitrile to methanol was used because this complex is insufficiently soluble in acetonitrile. Stabilization of the noncovalent guest-host interactions requires polymer formation at as low a temperature as possible, and a temperature of 4 °C was used. Irradiation times for all cases were 24 hours.

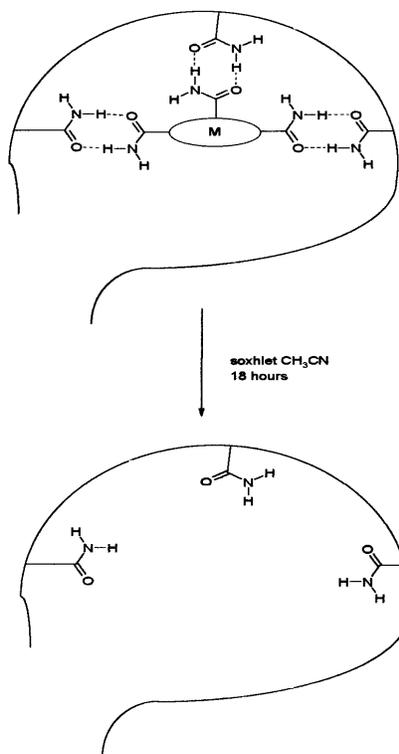
**Polymer characterization.** The new polymers were characterized by infrared spectroscopy, polymer swelling, and surface area/porosity measurements (**Table 6**). For **P1**, **P2a**, and **P4** the imprint was removed from the polymer by soxhlet extraction with acetonitrile (**Figure 8**). The color of the complex was observed to transfer into the acetonitrile solution and the soluble product could be isolated by concentration of this solution. The products displayed the same infrared spectra as the starting imprints. Taking account of the fact that **P2b** is also an ion exchange resin, the colored complex was removed by repeated extraction with ammonium chloride.

The pore size and the surface area of polymer samples were measured by gas desorption (N<sub>2</sub>) using the BET isotherm. Swelling (ml/ml) was measured by submerging the ground, dried and extracted polymer in acetonitrile for 24 hours. Even though the monomer (acrylamide), porogen (acetonitrile), and crosslinker (ethylene glycol dimethacrylate) were the same in polymers **P1**, **P2a**, **P2b** and **P3**, a large variation was observed in polymer morphology (**Table 6**). The measurements of surface area and pore size distribution indicated we were not uniformly successful in producing macroporous polymers in all cases. **P2a** and **P3** gave surface areas of slightly greater than 200m<sup>2</sup>g<sup>-1</sup>, in the range expected for macroporous resins (50-1000m<sup>2</sup>g<sup>-1</sup>). However under identical conditions the polymers **P1** and **P2b** have both much reduced surface area and less swelling, with surface areas ~10m<sup>2</sup>g<sup>-1</sup> at the lower end of the range for macroporous materials.

**Table 6.** Morphology of Polymer Samples

Polymer	Surface Area, m <sup>2</sup> g <sup>-1</sup>	Swellability	Pore volume, ml.g <sup>-1</sup>
<b>P1</b>	16	1.23	0.049
<b>P2a</b>	216	1.83	0.606
<b>P2b</b>	8	1.21	0.024
<b>P3</b>	204	1.46	0.396
<b>P4</b>	1	1.63	0.006

The pore volumes are in accord with the surface areas, being much greater for polymers **P2a** and **P3**, suggesting facile access by diffusion into the imprinted sites for these two samples. The attempt to get a higher degree of crosslinking with the tetrafunctional reagent, pentaerythritol tetraacrylate, **P4**, gave an inferior product having a high degree of swelling, low pore volume and low surface area. The sample was discarded.



**Figure 8**

**Complex Rebinding Studies.** The binding of the nickel(II) tetramide complex **10** with polymers **P1**, **P2a**, **P2b**, and **P3** was evaluated in the final step in this first proof of concept study. These experiments were carried out by atomic absorption on acetonitrile solutions of complex **10**. Sample **P2a** displays good properties for a macroporous polymer and rebinding of **10** into **P2a** led to the filling of 21% of the theoretically available sites. This is most encouraging because this number falls at the top end of the range observed for receptees held by hydrogen bonding (15-20% rebinding).<sup>18</sup> Comparison of the results of this best example of rebinding (**P2a**) with that of the polymer having randomly generated sites (**P3**) provides information about selectivity; the templated polymer (**P2a**) bound over 2&1/2 times as much complex as did its non-templated counterpart. This bodes well for the success of this entirely new methodology in important applications.

Further, we hold high hopes for greatly increasing the rebinding capacities of these macroporous resins by the simple process of templating the counter ion into the selective site, thereby introducing electrostatics as a second force favoring the rebinding process. Polymer **P2b** was designed for this purpose. Although we have not yet prepared an optimized version of **P2b**, the rebinding of complex **10** amounted to 25% of the available sites, actually exceeding the rebinding to our nicely macroporous baseline polymer **P2a**. The results of our rebinding experiments with **P1** dramatize the enhancement in binding that is provided by the presence of counter ions at the receptor sites in **P2b**; the two polymers showed similar surface areas, swellabilities, and pore volumes. The result for **P1** (2%) is barely 1/10 that for **P2a**. This failure to rebind is a fair measure of the importance of the macroporous structure; it is remarkable that a **P2b**, which is not macroporous, could rebind to 25% of the sites. Clearly this high level of rebinding in a poorly macroporous sample suggests that the optimized polymer having counter ions will have spectacular rebinding properties.

## RELEVANCE, IMPACT AND TECHNOLOGY TRANSFER

### Relevance to Critical DOE Environmental Management Problems

There are many ways in which this research has relevance and offers potential for impact

and technology transfer to environmental management problems of DOE<sup>19,20</sup>. Because the research is basic science, its value to EMSP relates to long term problems for which existing technologies do not necessarily offer definitive solutions. Our program on switch-binding and switch-release of metal ions involves fundamental inorganic chemistry and organic chemistry with the former focused on leading edge coordination chemistry and latter on advanced studies of caging and photo-release of targeted chemical species. The imprinted polymer developments associated with our soil poultice program will contribute to both Chemical Sciences and Material Sciences. Many ultimate applications of this new science will be of value in the area of biological and environmental research; they have great potential for environmental remediation.

It is a stated objective of EMSP<sup>19</sup> to “..provide scientific knowledge that will revolutionize technology and cleanup approaches to significantly reduce future costs, schedules and risks. ” The program described here will provide foundations for the development of a number of new methodologies, any one of which could revolutionize separations technology. Turning to the EMSP Science Needs by Focus Area<sup>20</sup>, the proposed research is enhancing fundamental science and, in so doing, it is building underlying foundations for many of the specific needs of EMSP.

Switch-binding and switch-release will make available the most strongly binding ligands for processes in which their particular advantages have rarely been used because they react too slowly or they simply cannot be controlled. Examples are:

**Deactivation and Decommissioning.** “Decontamination of metal surfaces”; the great affinities of ultra tight-binding ligands make them attractive for this purpose. In *Separations Chemistry*--”focus is on ligand design and ion exchange.” Here we offer some of the most advanced concepts in ligand design in all of chemistry.

**Environmental Restoration.** “Improvements in treatment technologies for DNAPLs (including mercury).” Clearly new switch-binding/release ligands containing sulfur donors would offer much promise in this area. *Analytical Chemistry and Instrumentation*: “molecular recognition based” --calls for special compounds for use in detection and monitoring of targeted species. Our new methods of switch binding and release should open access to compounds sufficiently robust for this application.

**High-Level Waste.** “Development of improved separation agents and processes to remove cesium, strontium, technetium and transuranics from supernatant solutions. ” These application are in precise alignment with ultimate goals of this project. *Separations Chemistry*: “..and removal of alkali cations.” Alkali cation removal is a specific target of this program.

The proposed *slow technology* represented by our *soil poultice* offers an entirely new approach to

critical separation needs. Examples are:

**Deactivation and Decommissioning.** “Decontamination of metal surfaces (using environmentally benign aqueous-based biopolymer solutions).” The soil poultice could be applied to the surface, kept moist and then mechanically removed after the contaminants had been adsorbed. *Separations Chemistry*: Ultimate applications of the soil poultice concepts should include “innovative separation techniques for highly toxic radioisotopes such as plutonium”.

**Environmental Restoration.** “Fundamental improvements to soil cleanup and segregation technologies in order to limit the volume of soil that must be excavated and stored in permitted waste facilities.” The soil poultice is ideal for this scenario.

**High-Level Waste.** *Actinide chemistry*: “..developing templated ion exchange resins for selective complexation of actinide ions for aqueous solutions.” Our work with the basic science of the templated resins will contribute to this goal. *Separations Chemistry*: “..development of anion binding ligands, improved ion exchange resins and...” Our soil poultice concept envisions an improved resin that has the potential for high selectivity and extremely strong binding.

**MLLW/TRU and also Spent Nuclear Fuel.** *Separations Chemistry*: “Investment is being made to advance sol-gel chemistry and molecular imprinting techniques to create hydrophilic metal oxide-based materials for tailored recognition/separation of toxic metals.” Our soil poultice gives promise of a companion technology to that envisioned here.

## **Reducing Costs, Schedules and Risks and Improved Compliance**

At the basic research level it is easy to make claims but difficult to provide substantive arguments for progress in these areas. New technologies that will simplify and accelerate decontamination of soils and surfaces are expected to be beneficial with regard to all three: reduced costs, improved schedule reliability, and reduced risks. The list provided in the preceding section contains many possible examples of advantages of this kind.

## **Bridging the Gap between Basic Research and Timely Needs-Driven Applications**

The soil poultice is squarely planted in the gap since the broad fundamental science has been done by polymer scientists first demonstrating the function of potential guests as templates in the formation of macroporous polymers. Further, the coordination chemistry to be utilized is well established. However, it is necessary to develop the basic science of designing templated macroporous polymers that will strongly re-bind metal complexes, in the field. In this context we

are very excited about our leads that suggest adding coulombic attractions to hydrogen bonding interactions may produce very strong affinities and therefore good rebinding to the polymer bead of the complex to be formed when the selective ligand is released to sequester contaminants in the soil.

Our program on making extremely powerful ligands useful for separations science by replacing lethargic spontaneous equilibration with switching processes is approaching a similar position in the gap between basic research and applications. Earlier work by others on switch-release of a metal ion by photochemical processes has been repeated and extended in our labs, but there remain some specific challenges before the methodologies can be extended to a broad range of ligand types; e.g., sulfur-containing ligands for mercury recovery. Switch-binding has been taken a giant step forward with proof of the underlying concept in our laboratories. However, there is more to be learned in these cases before successful applications will be made.

### **Impact on Possible Users and the Identification of Those Users**

The greatest value of the proposed studies is in the fundamental scientific relationships they will reveal and clarify. These involve a number of basic research fields. The binding and release relationships among receptor/receptee complexes are fundamental coordination chemistry, or supramolecular chemistry. The reason many very powerful ligands are not used in separations and other sequestration processes is because the equilibrium processes that lead to formation and dissociation of their complexes are simply too slow. We have shown emphatically that one can accomplish much more rapid binding and release by replacing the equilibrium process with *switching mechanism* on the molecular scale. Today people often write and speak about using molecular switches for such applications as computer components and other electronic devices. Here we use switching to make fundamental chemistry more useful. It is our expectation that others will make use of our innovations in many fundamental scientific areas and, ultimately, in application areas that one would not anticipate at this point time.

As we pursue our vectors in these areas, the principles will be generalized sufficiently to apply them to alkali, alkaline earth, and RECRA metals in our laboratories. Immediate plans call for combining the switches for binding and release in a single ligand suitable for alkali metal binding. This should be a prototype for applications and we will be seeking partners for those developments in, for example, actinide removal. Ultimately, we expect these principles to be applied to the binding and release of metal ions and other guest molecules under demanding conditions, for example, capturing contaminants in very dilute media and removing contaminating metal ions from mineralized contaminants. Such applications are probably a few years in the future.

### **Are Larger Scale Trials Warranted?**

The answer to this questions is simply “not yet”. The basic science is proceeding well, but it takes a while to move it to real applications. Realistically, this is the only way new tools are made available to treat the most difficult problems.

## Improving the Capabilities of Collaborating Scientists

This program has brought together an organic photochemist and an inorganic coordination chemist, and the impact on the partners has been highly beneficial. Joint research group meetings are symptomatic in the way they have added broader ranging perspectives on the challenging problems of this project with good results.

## Advances in Scientific Understanding and Remaining Hurdles to DOE Applications

**Switch-Binding.** Templating as a “switch binding” process uses a principle first enunciated and demonstrated by one of us for an entirely new purpose that differs from the goals here.<sup>21</sup> The original purpose of template reactions was simply to control the structural outcome of molecule synthesis. Remarkably, we now find that this same template effect can accelerate the rate of binding of a ligand to a metal ion by orders of magnitude, bringing those processes into rate ranges that make many applications possible. The main hurdle before us is generalizing this new phenomenon to a level where we can design ligands to control molecular species of specific interest to DOE. We have shown that the concept works with the best metal ion for this kind of study, nickel(II), whereas plutonium(IV), mercury(II), cesium(I), strontium(II), or cobalt(II) would be of greater interest to DOE. Further, we have only shown that it works with a single kind of ligand and with a single kind of ring-closing reaction. The immediate challenge is to expand the toolbox to a useable level; *generation 2* ligands and some of their variations will be most helpful in that regard. The second hurdle is associated with the mode of delivery of the switch-binding ligand. In all probability it will be as the functional group on a resin or as a component in a solvent extraction system. A final hurdle will emerge when a target species is selected because the templating ligand must be designed specifically for it.

**Switch-Release.** The "switch release" objective, which photolytically releases metal ions from photo reactive ligands, extends the new science of photo-released metal ions, a development that may be viewed as overdue. We have confirmed early literature studies on this important result and raised many new questions, including: (1) What will be the effects of the metal ion on the efficiency and, possibly, the nature of the photochemical reaction? (2) What will control the rate of release of the metal ion during the photochemical reaction? (3) Will it be possible to discover/design systems in which the switch release and switch binding can be used repetitively with the same ligand samples?

**The Slow Technology.** The *soil poultice* undergoing development in this work requires the synthesis and design of new, highly effective compounds and materials. This takes an idea from nature--how certain single-cell organisms extract metal ions from nature--and designs a slow technology that may facilitate the use of very tight binding ligands in the retrieval of metal ions from many kinds of environmental contaminations. We use imprinted polymers as the corpus of the soil poultice, and imprint them to recognize and bind a certain metal complex. The metal complex is

formed in the vicinity of a physically retrievable polymer particle by releasing the strong ligand in solution in the vicinity of metal ion contaminated site. The ligand, working at its own pace takes the metal ion from its mineralized, or surface adsorbed, form and the resulting complex is subsequently recognized and captured selectively by the macroporous polymer. The seminal aspect of these studies is the goal of having the imprinted polymer selectively recognize and bind the *metal complex* and, ideally, not the free ligand or the complex of the same ligand with other metal ions.

Because of limited resources in the first funding period for this project, the studies on this aspect of the project have been very limited. We have designed a few ligands and, with only a small number of polymer samples we have shown that macroporous polymers can be imprinted for their metal complexes, giving rebinding equivalent to or better than that commonly observed for other systems. In these first studies only hydrogen bonding was used to hold the guest complex in the polymer receptor, but a single result, in which the polymer morphology was marginal, showed that incorporation of a counter ion into the binding site may provide a profound increase in re-binding ability. Remaining hurdles that must precede designing and developing practical applications are: (1) optimizing polymer morphology; (2) demonstrating the full cycle for the present example in which the free ligand is used to retrieve metal ions from precipitates, or mineral sources; and (3) evaluating the selectivity and affinity of re-binding in the optimized system.

### **Interest Expressed by other Government Agencies or Private Enterprises**

Exposure of these results has so far been limited to theses and presentations at scientific and DOE meetings so that relatively little opportunity has arisen for such interactions and no significant business interests have been expressed.

## **PROJECT PRODUCTIVITY**

Recalling that this project involves basic research and research to span the gap between that basic research and applications, it is fair to say the project accomplished more than can necessarily be expected of a 3 year investigation with limited resources. In fact because of those limitations equal emphasis could not be placed on all three objectives and because Switch-Binding and Switch-Release are connected in principle, the slow technology received the least emphasis. During the project period we achieved a proof of concept on an entirely new principle--template controlled switch-binding, a landmark achievement for any basic science program. Further, our studies on photochemical switch-release confirmed previously known research and extended it to systems of interest to this program. The original plan to work equally on all three objectives was revised so that attention was given to the third objective, the soil poultice, for only a little over one year from among the three years of support.

## **PERSONNEL SUPPORTED**

### **Post Doctoral Research Associates**

Dr. Steven Archibald  
Dr. Andrew Danby  
Dr. Andreas Jung

### **Graduate Research Assistants**

Dr. Andreas Jung  
Erick Eduardo Honores  
Anne K. McCasland

## **PUBLICATIONS**

Anne McCasland, Ph.D. Dissertation, "Accelerating the Binding Rate for Powerful Ligands by Switch-binding," University of Kansas, 1999.

Erick Honores, MS Thesis, "Synthesis and Photochemistry of a New Conjugate Pair of Macromolecules for the Facile Release of Metal Cations," University of Kansas, 2000.

## **INTERACTIONS**

### **Participation/Presentations at Meetings, Workshops, Conferences**

**Daryle H. Busch.** January 30, 1996, Elena Rybak-Akimova, Alexander G. Kolchinski, John G. O'Brien and Daryle H. Busch, "Dynamics of Cobalt(ii) Dioxygen Carriers," International Macrocyclic Meeting, New Zealand.

August, 1996, "Octahedral Molecular Templates: Nickel(II) Complexes of Schiff Base Ligands with Terminal Functional Groups," Andrew L. Vance, Nathaniel W. Alcock, Daryle H. Busch, Joseph A. Heppert, International Conference on Coordination Chemistry, Vancouver, Canada.

June 12, 1996, "Lacunar Transition Metal Dioxygen Carriers--Good and Getting Better," Daryle H. Busch, University of Kansas, 4th Quinquennial D.H.Busch Reunion Symposium, Central Regional Meeting, ACS, Dayton, OH.

1996ACS Central Regional Meeting, Dayton, Ohio; papers joint with students:

"Preparation and Properties of a New Family of Lacunar Cyclidene Oxygen Carriers," D. H. Busch, A. G. Kolchinski, B. Korybut-Daszkiwicz, N. W. Alcock, H. J. Clase

"Synthesis and Characterization of New Lacunar Dioxygen Carriers Derived from an Extremely Bulky Tetraaza(14) Annulene," John G. O'Brien, Daryle H. Busch

"Kinetic and Thermodynamic Characterization of the Reactions of Iron(II) Cyclidene

Complexes with Carbon Monoxide,” M. Buchalova, P. R. Warburton, Daryle H. Busch

“Kinetics of Dioxygen Binding to a Vacant Site in Cobalt(II) Complexes,” E. V. Rybak-Akimova, M. Masarwa, W. Otto, K. Marek, D. H. Busch

Jan. 28-30, 1997, DOE Efficient Separations Processes Annual Technical Meeting, Gaithersburg, MD, panel member.

April 1-3, 1997, DOE Efficient Separations Processes, Annual Review of Research projects, Gaithersburg, MD, panel member.

Apr. 11-17, 1997, 213th National ACS Meeting, San Francisco, CA, papers joint with students: S.S.Siltchenko and D.H. Busch, “Synthesis and Immobilization of Functionalized Long Chain Molecules on a Mica surface”;

A.G.Kolchinski, N. W. Alcock, R. A. Roesner and D. H. Busch, “Molecular Stapling--Preparation of a [3]-Rotaxane by Oxidative Dimerization of 2-(9-Anthracenylmethylamino)-ethanethiol Threaded through a Crown Ether;

E.V. Rybak-Akimova, K. Kuczera, and D.H. Busch, “Molecular Modeling (CHARMm) Studies of Ligand Binding within the Cavity of Unbridged Cyclidenes.”

Aug. 1-2, 1997, Plenary Lecture, International Coordination Chemistry Conference, Seoul, Korea, “Optimized Transition Metal Dioxygen Carriers.”

Aug. 3- 8, 1997, Plenary Lecture, International Macrocyclic Meeting, Seoul, Korea, “Ligand Design for Enhanced Molecular Organization.”

September 1, 1998, invited lecture at International Conference on Coordination Chemistry, Florence, Italy, “Coordination Chemistry, Catalysis, and Supramolecular Materials for the Millennium”.

August 25, 1998, lecture of honored guest, National American Chemical Society Meeting, Boston, MA, “Coordination Chemistry, Catalysis, and Supramolecular Materials for the Millennium”.

September 1, 1998, invited lecture at International Conference on Coordination Chemistry, Florence, Italy, “Coordination Chemistry, Catalysis, and Supramolecular Materials for the Millennium”.

March 22, 1999, National ACS Meeting, Anaheim, CA, “Cross-bridged Macrocyclic Ligands for New Oxidation Catalysts.”

November 22, 1999, member of review panel, Alternate Technologies for Cesium Removal from Nuclear Waste, Augusta, GA.

January 5-8, 2000, Newport Beach, CA, Served on National Academy of Science Committee on Nuclear Waste Management--the Cesium Problem.

February 2, 2000, Irvine, CA, Served on National Academy of Science Committee on Nuclear Waste Management--the Cesium Problem.

April 24-27, 2000, Atlanta, GA, presented poster and participated in Department of Energy Environmental Science Program Meeting.

July 7, 2000, St. Andrews, Scotland, plenary lecture at the 2000 International Macrocyclic Meeting, "Special Ligands for Special Purposes--the Wedding of Organic and Inorganic Coordination Chemistry."

September 18, 2000, Kusatsu City, Japan, Plenary Lecture at the International New Horizons in Coordination Chemistry Conference on "Special Ligands for Special Purposes."

February 13-15, 2001, Augusta, GA, serve on NRC Committee to evaluate "Research and Development on a Salt Processing Alternative for high-Level waste at the Savannah River Site."

March 26-27, 2001, Washington, D.C., serve on NRC Committee to evaluate "Research and Development on a Salt Processing Alternative for high-Level waste at the Savannah River Site."

April 15-22, 2001, Havana, Cuba, IV International Congress on chemistry and XIII Caribbean Conference on Chemistry and Chemical Engineering, plenary lecture, "Special Ligands for Special Purposes--Applications of Coordination Chemistry."

**Richard S. Givens.** Attended DOE sponsored workshop on "Characterization, Monitoring and Modeling fo the Valdose Zone: Flow and Transport", University of Arizona, Tuscon, AR (5/17 - 19/1999).

Attended DOE sponsored workshop: "Environmental Management Science Program Workshop", Chicago, IL (7/27 - 30/1998).

Chair, Gordon Conference on Organic Photochemistry (1999-2001)

Chair, 2<sup>nd</sup> International Symposium on Caged Compounds and Photoremovable Protecting Groups, PacifiChem 2000 (1999-2000).

American Chemical Society meeting, San Diego, CA April 2,3 2001; "Photostitches: Design,

Synthesis, and investigation of New Protein Cross Linking Agents” Richard S. Givens, George T. Timberlake, J. F. W. Weber, P. G. Conrad II, S. Amslinger, R. Herpel (presented twice Monday evening 4/2 and Tuesday evening 4/3).

American Chemical Society meeting, Washington, DC Aug. 24, 2000; “Bifunctional Photoremoveable Protecting Group: Modifications Directed Toward Controlling Peptide Properties and Structure” Peter G. Conrad, R. S. Givens

Pacificchem2000; December 18, 2000; co-organizer, plenary lecture “*p*-Hydroxyphenacyl Phototriggers: In a Class by Itself” and a poster, “Phototriggers as Protein Crosslinking Agents.”

ACS National Meeting; August 20, 2000; Washington, D.C. “New Chromophores for Phototriggers; Extending the *p*-Hydroxyphenacyl Cage for Glutamate and GABA.”

Emporia State University: October 31, 2000; recruiting seminar “How to Tame a Neurotransmitter: Phototriggers for L-Glutamate, GABA, and Bradykinin”

Los Alamos National Laboratory, April 10, 2000; “Photoremoveable Protecting Groups in Chemistry and Biology”

San Jose, CR; February 2-8, 2000; Seminar to Chemistry faculty as part of the KU-KSU-CR Symposium

Marietta College, February 10, 2000; Acceptance speech, honorary D. Sc. from Marietta College, “Evolution and Science Education; Was Darwin Right?”

Marietta College Chemistry Department: Research seminar to the Chemistry Department, Marietta College the following day; “A BIRD in a Gilded Cage”.

Central Midwest ACS Regional Meeting, Cincinnati, Ohio; Invited symposium speaker, May 17-19, 2000 “How to Tame a Neurotransmitter: Phototriggers for L-Glutamate, GABA, and Bradykinin”

An invited seminar at the Universidad Complutense, Madrid, Spain on May 30, 2000;

Phototriggers, Switches and Stitches: “Photoremoveable Protecting Groups in Chemistry and Biology”

InterAmerican Photochemical Society annual meeting, Clearwater, FL. Jan. 3-7, 2000; “A BIRD in a Gilded Cage”; Richard S. Givens\*, Peter Conrad II, Jorg Weber, Andreas Jung, Sabine Amslinger, Leroy Chimilio, and George Orosz.

### **Consultative and Advisory Functions to Other Labs and Agencies**

**Daryle H. Busch.** Oak Ridge National Laboratory, Chemistry Division Review Committee, 1991-1996, chair 1993 & 1995.

Chemistry Division Annual Review, Argonne National Laboratory, 1993-5.

Served on both committees to resolve the question about which process will be used to remove cesium-137 in the course of tank waste reprocessing at the DOE Savannah River Site.

Efficient Separations and Processing Integrated Program, DOE, served on project evaluation panel from its beginning until 1996.

Brookhaven National Laboratory, Chemistry Division Review Committee (earlier)

Consultant to Savannah River Laboratory for approximately 20 years ending with termination of Dupont as prime contractor at the site.

Consultant and/or collaborative research leading to patents with the following companies: 3M, Monsanto, Procter & Gamble, Air Products & Chemicals, and Dupont. Also consulted with Praxair, Beaunit Fibers, Allyn & Bacon, Inc., and Chemical Abstracts Service.

Served on research project review panels for NIH, NSF, DOE ESMP, and USAF Systems Development Command.

**Richard S. Givens.** NSF Graduate Research Fellowship Program, (2001-).

Review Board Chemistry Panel Member, NRC-NAS Associateship Program (1987- 1996; Chair of the Chemistry Panel 1991- 1996; Chair of the Review Board, 1992-1996).

Member, Associateship Programs Advisory Committee, NAS-NRC (1991-1996).

Member, International Scientific Committee, International Symposia on Luminescence Spectrometry in Biomedical Analysis (1992-1996).

### **Collaborations**

The present project is the result of a collaboration between an organic photochemist and an inorganic coordination chemist. The program involves no additional collaborations. However, both Co-PIs have substantial records of collaboration.

**Daryle H. Busch.** October 1992 -- September 1998, Director of NSF EPSCoR programs involving from 11 to 22 other faculty members.

October 1998--present, Co-PI with Professor Bala Subramaniam on a joint project exploring “Homogeneous Catalysis in Dense Phase Carbon Dioxide”, that has been sponsored by NSF.

March 1990 -- present, collaborate with chemists at Procter & Gamble Co., on “Durable Oxidation Catalysts for Consumer Applications.”

Collaborative research with three other industrial companies have led to patents; see below.

**Richard S. Givens.** Several collaborations have resulted from the work on phototriggers (caged compounds). Early collaborations were directed toward the release of neurotransmitters and ATP; more recent collaborations have been with the use of new phototriggers and with photostitching crosslinking agents:

L-Glutamate and GABA - collaborations with Dr. Larry Katz (Duke) and Dr. Karl Kandler (U. Of Pittsburgh).

ATP - collabation with Dr. Klaus Fendler (Max Planck Institute for Biochemistry, Frankfurt, Germany).

Bradykinin - collaborations with Dr. Stanley A. Thayer (University of Minnesota); Dr. Philip Haydon (Iowa State University); Dr. George Orosz (Budapest, Hungary).

p-Hydroxyphenacyl bromide/enzyme reactivity - collaborations with Dr. Hagan Bayley (Texas A. and M., College Station, TX); with Dr. Jakob Wirz (University of Basil, Switzerland).

p-Hydroxyphenacyl oligopeptide derivatives - collaboration with Dr. Thomas Kiefhalber; (University of Basil, Switzerland); Dr. S. I. Chan, (California Institute of Technology).

Photocrosslinking agents - Dr. George Timberlake (University of Kansas Medical Center).

## TRANSITIONS

### Daryle H. Busch

Under sponsorship of the 3M Company we developed chemical systems for a superior “carbon-less copy paper” technology. Previously 3M had used their own technology that gave the familiar purple print on the copy page as, for example, used by gasoline vendors. The color was considered inferior and by investigation of basic chemical properties and synthesis of new compounds within their technology we quickly provided them with an alternative, but related, chemical system that gave an excellent deep blue imprint. Unfortunately, the patents ran out on a less expensive technology controlled by competitors and 3M chose to use that technology. This was

something like a decade ago and personnel have all turned over. Dr. David Whitcomb, now with Kodak in Rochester, would be best informed on the topic. 3M spun off the business which was subsequently purchased by Kodak.

In collaboration with Air Products & Chemicals, Inc., we developed superior new soluble chemical dioxygen carriers with potential for separating oxygen from the nitrogen and other gases in the air, especially on small scales. Patents issued but the product is not in production. This work is relatively old and the patents have expired.

Procter & Gamble personnel asked if it would be possible to design homogeneous catalysts that could selectively promote the bleaching of stains from soiled fabrics under ordinary laundry conditions. We responded by developing, in collaboration with them, new molecular designs that are remarkably effective. Tons have been manufactured and the product has undergone consumer tests. Drs. David Kitko and Chris Perkins, both with P&G in Cincinnati, are best informed on this program. The program is still alive and well.

### **Richard S. Givens**

Work on our phototriggerable collagen crosslinking compounds has attracted the attention of researchers at four companies, including Dow and 3M. The principal contacts are Dr. William Kueper and Dr. Philip Athey, respectively. It is anticipated that these two individuals will be asking for a presentation of the work on site in the near future.

Our current work on photorelease of biologically active substrates has been the basis for commercial speciality materials from Molecular Probes, Inc. We did not patent the technology for the phototriggers for release of ATP, glutamate and gaba and have chosen to share our expertise and our experimental procedures with chemists at Molecular Probes and with other researchers when requested. Our contact at Molecular Probes has been Dr. Kyle Gee.

## **PATENTS**

### **Daryle H. Busch**

1. D. P. Riley, D. H. Busch, and X. Zhang, (Monsanto Co., University of Kansas), "Peroxydinitrite Decomposition Catalyst and Therapeutic Use," Patent Publication under the Patent Cooperation Treaty (PCT), WO 98-US5567, March 26, **1998**. (US Priority 97-43394, April 1, 1997; International Application Number IPC: A61K031-435.
2. D.H.Busch, S.R.Collinson, T.J.Hubin, "Catalysts and Methods for Catalytic Oxidation," International Patent Publication under the Patent Cooperation Treaty (PCT), WO 98/39098, September 11, **1998**. (filing date March 6, 1998; International Application Number: PCT/IB98/00302)

3. D.H.Busch, S.R.Collinson, T.J.Hubin, R. Labeque, B.K.Williams, J. P. Johnston, D. Kitko, J. Burckett-St.Laurent, C.M.Perkins, "Bleach Compositions", International Patent Publication under the Patent Cooperation Treaty (PCT), WO 98/39406, September 11, **1998**. (filing date March 6, 1998; International Application Number: PCT/IB98/00300)
4. P. Richard Warburton and Daryle H. Busch, "Sensor for Carbon Monoxide", U. S. Patent 5,250,171, October 5, **1993**.
5. Loren D. Albin, David R. Boston, Derek R. Callaby, Jacqueline M. Furlong, Robert J. Lokken, Roger A. Mader, David B. Olson, Wayne O. Otteson, Norman P. Sweeny, and Daryle H. Busch, "Monosubstituted Dithiooxamide Compounds and Their Use", U.S. Patent 5,124,308, June 23, **1992**.
6. Nusrallah Jubran, Daryle H. Busch and other inventors, "Monosubstituted Dithiooxamide Compounds and Their Use", European Patent 90312531.8, June 26, **1991**.
7. Daryle H. Busch, "Salts of Cationic-Metal Dry Cave Complexes," U.S. Patent 4,888,032, **1989**.
8. John A. T. Norman, Dorai Ramprasad and Daryle H. Busch, "Pillared Cobalt Complexes for Oxygen Separation," U.S. Patent 4,735,634, April 5, **1988**.
9. Dorai Ramprasad and Daryle H. Busch, "Lacunar Cobalt Complexes for Oxygen Separation," U.S. Patent 4,680,037, July 14, **1987**.
10. Daryle H. Busch and Wade H. Jordan, "Voltaic Cells," U.S. Patent 3,545,022, December 8, 1970; Ger. Offen., DE1935941, 13 August **1970**.

### **Richard S. Givens**

1. Richard S. Givens, George T. Timberlake, Peter G. Conrad II, "Photo-triggerable Collagen Crosslinking Compounds for Wound Closure." Disclosure submitted Mar. 20, 2001. Pending
2. Richard S. Givens, Robert G. Carlson, Kasturi Srinivasachar and Osborne S. Wong, Takeru Higuchi, "Substituted 2,3-Naphthalenedicarboxaldehydes." U.S. Patent. Issued: Feb. 2, 1988 Patent No. 4,723,022 (US; Also Australian, New Zealand, European Convention).
3. Robert G. Carlson, Richard S. Givens, and Osborne S. Wong "Fluorogenic 2,1,3-Benzoxadiazoles and Fluorogenic Amine/Thiol Assays Therewith." Issued: Sept. 6, 1988 Patent No. 4,769,467 (US only).

### **FUTURE WORK**

That which remains to be done is best considered in two categories: (1) work leading to effective completion of the proposed project, as described, and (2) work that follows naturally from having accomplished the proposal goals.

### **(1) Work Leading to Completion of Project**

**The New Slow Technology--the Soil Poultice.** The very limited time and resources spent on this component of the project have provided exceptionally promising results and this effort must be continued at higher priority. Techniques will be perfected to assure preparation of macroporous polymers of reproducible morphology and properties. The combination of hydrogen bonding and electrostatic attraction due to imprinting of the counter ion into the polymer promises to give rebinding yields far in excess of those that have been observed in our labs or elsewhere for hydrogen bonding alone, perhaps approaching those involving covalent bonds. Multiple samples will be prepared and evaluated to establish reliable, critical rebinding characteristics and then efforts will move on to demonstration of the “poultice” principle.

In previous experiments and those proposed above, rebinding to a macroporous polymer is accomplished with the pre-prepared metal complex. The “poultice” principle requires the free ligand to first seek out the metal ion, complex with it, and then the complex must find the polymer and bind to it. These experiments will be carried out on small samples in the laboratory and will proceed in stages: (1) ligand will be exposed to dissolved metal ions in the presence of the macroporous terminal host; (2) the dissolved ligand will seek the metal ion in fresh precipitates, e.g., hydrous oxides, in the presence of the macroporous terminal host; and (3) ligand will be exposed to less available metal ions; e.g., dried hydrous oxides, metal ion bound to resin beads, metal ion adsorbed on soil samples, mineral forms. It is anticipated that during the course of experiments of this kind, continued improvements will be made in the design of the macroporous polymer host and in the imprinting complex, especially in its external nature to improve selectivity and affinity of binding.

**Switch-Binding.** Progress has been greatest in this part of the program. The basic hypothesis supposes that ring-closure in specially designed ligands could be templated by their target metal ions so that the binding rate would be that of a topologically unconstrained (relatively) linear ligand while the product would be a macrocycle and would be endowed with its characteristic kinetic and thermodynamic stability. In this manner the inherent lethargy of really strong ligands can be overcome. A makeshift, *Generation 0*, system strongly supported this hypothesis and encouraged

us to complete the demanding design and synthesis of a ligand to provide a valid test of the hypothesis. When its reaction kinetics were examined in detail, the *Generation 1* ligand provided the proof of concept that was sought. Indeed extremely tight-binding ligands can be made by a relatively labile pathway using switch-binding. Now it is necessary to broaden the base of this conclusion to provide adequate tools to direct this principle to more important targets, for example,

mineralized cobalt, mercury, or actinide derivatives or alkali and alkaline earth ions adsorbed on soils. To recognize the need for this requisite basic research, it is necessary to recall the fact that highly idealized examples were designed in order to prove the concept. The Schiff base reaction was chosen for the templated ring closure of the linear precursor ligand into the desired macrocyclic product ligand because that reaction has been so well used in synthetic chemical reactions. On the other hand, the Schiff base reaction is far from ideal for separations studies because it is difficult to control prior to reaction, rapidly entering into equilibrium processes in the absence of the targeted templating ion. Further, to prevent these processes it is necessary to operate at lower pH values that are practical in many applications. Our *Generation 2* ligands were designed with those limitations in mind. Conversion of a dangling ester function into a lactone or amide can be expected to proceed rapidly under some conditions, but the variety and fragility of products associated with the Schiff base reaction are not expected. In this case, experimentation is required to learn how neighboring group effects might accelerate ester exchange to form macrocyclic lactones or nucleophilic attack on the ester by a neighboring amine to form macrocyclic amides. It is necessary to explore a broad enough realm of reaction chemistry to provide the tools to design systems appropriate to the binding of important target ions. Even in those early stages of development of this new foundation for technologies, interesting systems will be investigated, for example cryptates that form by switch-binding around such alkali metal ions as potassium and cesium. Further, it is our determination to build ligands in the near future that are capable of both switch-binding and switch-release.

**Switch-release.** Early work will determine the efficiency of the photoreactions for the first switch-release ligands made in our labs, and the effect of metal cations on the efficiencies and photochemistry of these substrates. Here too, it is necessary to expand the tool-box of reactions available for future design of ligand molecules specifically for challenging metal ions. Additional examples incorporating the *o*-nitrobenzyl substituted macrocycle will be synthesized and the photochemistry examined. Work is planned that will incorporate two independent photoremovable protecting groups in the same cryptand to provide the opportunity to selectively break either of two bonds. As mentioned in the preceding section, photo-release units will be built into ligands using *Generation 2* switch-binding chemistry; cryptates are envisioned as good target molecules for these studies. We are also very interested in the possibility of recyclization of switch-released ligand products by relatively simple chemical reactions. Further, there are exploratory photochemical issues that need to be explored, for example, photochemical cleavage of carbon-nitrogen and carbon-sulfur bonds.

## **(2) Work That Follows Naturally from Having Accomplished the Proposal Goals**

The eventual consequences of these combined studies are the opening of strong possibilities for important applications and new technologies, or modified and improved technologies. Presumably, we and others would soon be writing proposals to EMSP offering to design systems aimed at specific problems identified by DOE. The application of the most powerful ligands to the sequestration of metal ion contaminants from soils and dilute solutions is of great potential

value. Among the environmental pollution scenarios that are particularly troublesome to the standard separation and purification methodologies are (a) those involving extremely dilute streams of radioactive or RCRA elements and (b) those in which such substances are absorbed in soils. In both cases, tight-binding, adequately selective, receptors offer the possibility of effective new technologies. For case (a), large binding constants are essential because of the effect of high dilution; i.e., the moderate affinities of the receptors used in traditional solvent extraction or ion exchange methodologies may not assure binding. In the case of pollutants in soils, chemical binding to soil components or the presence of mineral forms can offer strong competition to any but the strongest of competing receptors. Fixing a template-selective ligand to a resin bed may facilitate removal of contaminant ions from solutions. The beauty of using the most strongly binding ligands in a variety of applications, lies in the fact that the metal ion may be firmly chelated until the demand for release is appropriate. At this point, photo release can be applied and the metal ion can be liberated for whatever role is intended. Along with other possibilities, this could be a part of a separations process, an event in a chemical switching sequence (e.g., signaling the detection of light), or the initiation of a metal ion catalyzed process. Thus the possible benefits of the proposed chemistry are wide ranging. For the systems of interest in this project, the generality of applications seems clear, the challenge is in molecular design and synthesis, and that is why the basic research must be extended to provide a reasonably broad range of tools as a part of the legacy of this quest.

The soil poultice is particularly fascinating because of the many conditions under which such a scheme would prove beneficial. For soil contaminated with radioactive cesium, that ion will be bound selectively over sodium and potassium, but a ligand selective for cesium could sequester the cesium to a complex for which polymer beads have been templated. The cesium complex would then be selectively bound to the beads and recovery of the beads would complete the decontamination process. Similarly, many surface treatments can be envisioned. For example, assume the walls and floors of a structure are contaminated with mercury. A paste would be prepared containing a ligand having a selective affinity for mercury and a solid polymer imprinted for the ligand complex of mercury. Upon standing, the ligand would diffuse to the surface and bind and solubilize the mercury. The complex would then diffuse about the solution until captured by the polymer, whence it would bind selectively. The recovered paste would then contain the mercury contaminant. Metal ion contaminants in pools of water could be removed by immersing mesh bags of beads of polymer imprinted for the metal complex, accompanied by inoculation of the pool with the ligand designed for the metal ion(s) in question. It follows that when we have demonstrated the proposed technology adequately, then proposals will be constructed for specific problems both within and outside of DOE.

#### LITERATURE CITED

1. For example, issue number 7 of *Chem.rev.*, 1999, 99, is devoted to Nanostructures.

2. Comprehensive Supramolecular Chemistry, Vols. 1-11, J-M. Lehn, Chairman of Editorial Board, Elsevier Science Ltd., Oxford, UK.
3. (a) Office of Science Financial Assistance Program Notice 99-06, *Environmental management Science Program: Research Related to Subsurface Contamination/Vadose Zone Issues*, U.S. Department of energy; (b) "EMSP Science Needs by Focus Area", [http://EMSP.em.doe.gov/focus\\_area.htm/](http://EMSP.em.doe.gov/focus_area.htm/).
4. (a) Daryle H. Busch, "Ligand Design for Enhanced Molecular Organization--Selectivity and Specific Sequencing in Multiple Receptor Ligands, and Orderly Molecular Entanglements," in *Transition Metal Ions in Supramolecular Chemistry*. ed. Luigi Fabbrizzi, Kluwer, pp. 55-79, 1994; (b) Daryle H. Busch, "The Compleat Coordination Chemistry -- What a Difference A Century Makes", *Werner Centennial Volume*, ACS Symposium Series 565, pp. 148-164, 1994; (c) D. H. Busch, *Chem.Rev.*, 93, 847-860, (1993); (d) Daryle H. Busch, "Metals and Enzymes -- Multiple Juxtapositional Fixedness," *Chem. Eng. News*, p. 9, June 29, 1970.
5. N.W. Alcock, P. Moore, H.A.A. Omar, *J. Chem. Soc., Dalton Trans.*, **1986**, 985; J. L. Karn, D.H.Busch, *Inorg. Chem.*, **1969**, 8, 1149; E.K. Barefield, F.V. Lovecchio, N.E.Tokel, E. Ochiai, D.H. Busch, *Inorg. Chem.*, **1972**, 11, 283.
6. M. Bodanszky, Y.S. Klausner, M.A.Ondetti, *Peptide Synthesis*, New York, John Wiley & Sons, **1989**; M. Bidabszky, *Peptide Chemistry, : A Practical Textbook*, New York, Springer-Verlag, **1988**; G.J. Atwell, W.A. Denny, *Synthesis*, **1984**, 1032.
7. S.M.Nelson, F.S. Esho, M.G.B. Drew, P. Bird, *Chem. Commun*, **1979**, 1035; D.A. House, N.F. Curtis, *J. Am. Chem. Soc.*, **1962**, 84, 3248; H. Adams, DN.A. Bailer, D.E. Fenton, R.J. Goed, R. Moody, E.O. Rodriguez deBarbarin, *J. Chem. Soc., Dalton Trans.*, **1987**, 207; S. Ryan, H. Adams, D.E. Fenton, M. Becker, S. Schindler, *Inorg. Chem.*, **1998**, 37, 2134.
8. J. L. Karn, D.H. Busch, *Nature*, **1966**, 211, 160; S.M. Nelson, *Pure Appl. Chem.*, **1980**, 52, 2461; K.P. Balakrishnan, H.A.A. Omar, P. Moore, N.W. Alcock, G.A. Pike, *J. Chem. Soc., Dalton Trans.*, **1990**, 2965; C. Cairns, S.G. McFall, S.M. Nelson, M.G.B. Drdew, *J. Chem. Soc., Dalton Trans.*, **1979**, 446; N.W. Alcock, R.G.Kingston, P. Moore, C. Pierpoint, *J. Chem. Soc., Dalton Trans.*, **1984**, 1937; D. Chem, A.E.Martell, *Tetrahedron Lett.*, **1991**, 47, 6895.
9. R.H. Prince, D.A. Stotter, P.R. Woolley, *In. Chim. Acta.*, **1974**, 9, 51.
10. A. K. McCasland, "Synthetic and Kinetic Studies towards the Acceleration of

- Macrocyclic Metal Complex Formation, ” Ph.D. Dissertation, The University of Kansas, August 27, 1999.
11. E.E. Honores, “Synthesis and Photochemistry of a New Conjugate Pair of Macromolecules for the Facile Release of Metal Cations ”, MS Thesis, The University of Kansas, Jan 28, 2000.
  12. R. Warmuth, E. Grell, J-M. Lehn, J.W. Bais, G. Quinkert, *Helv. Chem. Acta*, **1991**, *74*, 671 – 681.
  13. J.R. Telford, K. N. Raymond, In *Supramolecular Chemistry*; Gokel G. , Ed.Pergamon; Oxford, **1996**; Vol 1, p245; J.R. Telford, K. N. Raymond. In *Bioinorganic Chemistry, An Inorganic Perspective of Life*, Kluwer Academic Press, Dordecht, The Netherlands, **1995**, Vol. 459, p25.
  14. For reviews see: (a) Steinke J.; Sherrington D. C. and Dunkin I. R. *Advances in Polymer Science: Synthesis and Photosynthesis* **1995**, *123*, 81(Editor: Anglioni L.) (c) Sherrington D. C. *J,Chem. Soc., Chem. Commun.* **1998**, 2275. (d) Muldoon M. T. and Stanker L. H. *Chem. and Ind.* **1996**, *6*, 204. (e) Mosbach K. *Trends in Bio. Sci.* **1994**, *19*, 9. (f) Vlatakis G.; Andersson L. I.; Muller R. and Mosbach K. *Nature* **1993**, *361*, 645.
  15. G.F. Vandergrift G. F. US Patent 5468456, **1995**.
  16. K. P. Wainwright,. *J. Chem. Soc., Dalton Trans.* **1980**, 2117; G.M. Freeman, E. K. Barefield, D.G. van der Vee, *Inorg. Chem.*, **1984**, *23*, 3092; K.P. Wainwright, *J. Chem. Soc., Dalton Trans.*, **1983**, 1149.
  17. D.R. Lide, *CRC Handbook of Chemistry and Physics*; CRC Press: Boca Raton, **1994**; C. Yu, K. Mosbach, *J. Org. Chem.* **1997**, *12*, 4057.
  18. G. Wulff, *Angew. Chem. Int. Ed Engl.* **1995**, *34*, 1812; B. Sellergren, K.J. Shea, *J. Chromatogr.* **1993**, *635*, 31.
  19. Office of Science Financial Assistance Program Notice 99-06, *Environmental management Science Program: Research Related to Subsurface Contamination/Vadose Zone Issues*, U.S. Department of energy.
  20. “EMSP Science Needs by Focus Area ”, [http://EMSP.em.doe.gov/focus\\_area.htm/](http://EMSP.em.doe.gov/focus_area.htm/).
  21. M. C. Thompson and D.H. Busch, *J. Am. Chem. Soc.*, **1964**, *86*, 3651; J. D. Curry

and D. H. Busch, *J. Am. Chem. Soc.*, **1964**, 86, 592; G. A. Melson and D. H. Busch, *J. Am. Chem. Soc.*, **1964**, 86, 4834; T. J. Hubin, D. H. Busch, "Template routes to interlocked molecular structures and orderly molecular entanglements, " *Coord. Chem. Rev.*, **2000**, 200-202, 5-52; T. J. Hubin, A. G. Kolchinski, A. L. Vance, D. H. Busch, "Template Control of Supramolecular Architecture, " in *Advances in Supramolecular Chemistry*, Vol. 5, Ed. G. W. Gokel, JAI Press, **1999**, pp. 237-357; D. H. Busch, A. L. Vance, A. G. Kolchinski, "Molecular Template Effect: Historical View, Principles, and Perspectives, " in *Comprehensive Supramolecular Chemistry*, Vol. 9, pp 1-42, J-P. Sauvage and M. W. Hosseini, Ed., **1996**.

### **FEEDBACK**

None

### **APPENDICES**

None.

### **QUANTITIES/PACKAGING**

Report to be submitted in Word Perfect 6.1 format via e-mail to emsp@osti.gov. File will use the project Grant Number as its name, ER14708.wpd. A copy will be e-mailed to "Twitchell, Kara L" <twitchkl@id.doe.gov> to keep the Idaho office informed, and the Executive Summary will be e-mailed to "Hirsch, Roland" <Roland.Hirsch@science.doe.gov> to keep the Office of Science informed.