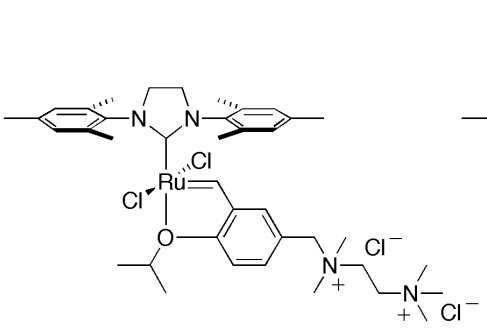
CHAPTER 6

Research Opportunities in Aqueous Olefin Metathesis

Opportunities for Catalyst Development



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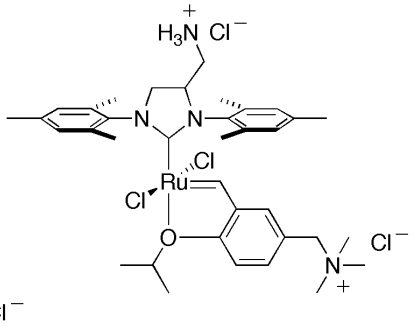

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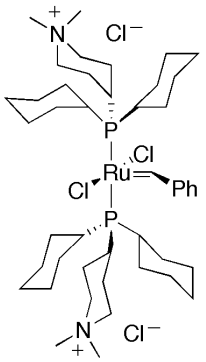

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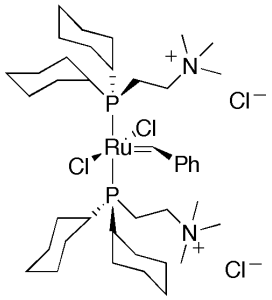

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The research presented in this thesis has established that olefin metathesis catalysts containing an *N*-heterocyclic carbene (NHC) ligand (**1** and **2**) are more stable and active in water than their bis(phosphine) counterparts (**3** and **4**). Indeed, the development of catalysts **1** and **2** represent significant progress in the ability to perform homogenous metathesis in water. As described in Chapter 5, catalysts **1** and **2** are competent at mediating ring-opening metathesis polymerization (ROMP) and ring-closing metathesis in water, yet their ability to catalyze cross-metathesis is limited. Therefore, further research is still required to generate a catalyst that fully mirrors in water the activity of olefin metathesis catalysts in organic solvents.

In all metathesis reactions, as with all catalyzed reactions, at least two processes compete: productive catalysis (reactivity) and catalyst decomposition (stability). Therefore, metathesis reactions are improved by increasing the rate of productive catalysis relative to the rate of catalyst decomposition. Two strategies are available to accomplish this objective, improve the rate of catalysis or decrease the rate of catalyst decomposition. Ideally, catalysts that are both more stable and more reactive are desired, but goals to improve one of these metrics are more realistic. Also, there is usually a trade-off between reactivity and stability in catalysis. More stable catalysts are typically less catalytically reactive and vice versa, and whether strategies that favor greater catalyst reactivity or stability are pursued depends on the targeted process. Examples of both strategies are proposed for aqueous olefin metathesis.

Improving the reactivity of water-soluble metathesis catalysts. One promising method to increase the rate of productive catalysis for olefin metathesis is to decrease the steric bulk around the metal, which allows substrates greater access to the catalytically active center.[1,2] This is the strategy employed by Grubbs and co-workers to develop catalysts **5**–**8**.[1,2] The goal of these catalysts is the facile ring-closing of geminal-disubstituted olefins to form a tetrasubstituted olefin, which is a challenging transformation in olefin metathesis (Figure 6.1).[3-8] Catalyst **8** is a particularly good catalyst for these transformations because it couples the ability to form tetrasubstituted olefins with reasonable complex stability, being more stable than catalysts **5**–**7**.[2]

Producing a water-soluble analog of catalyst **8** should be straightforward. Mixing ruthenium complex **7**[2] with isopropoxystyrene **9**[9] in the presence of copper(I)chloride should yield catalyst **10**, whose solubility properties should echo those of catalyst **1**. This

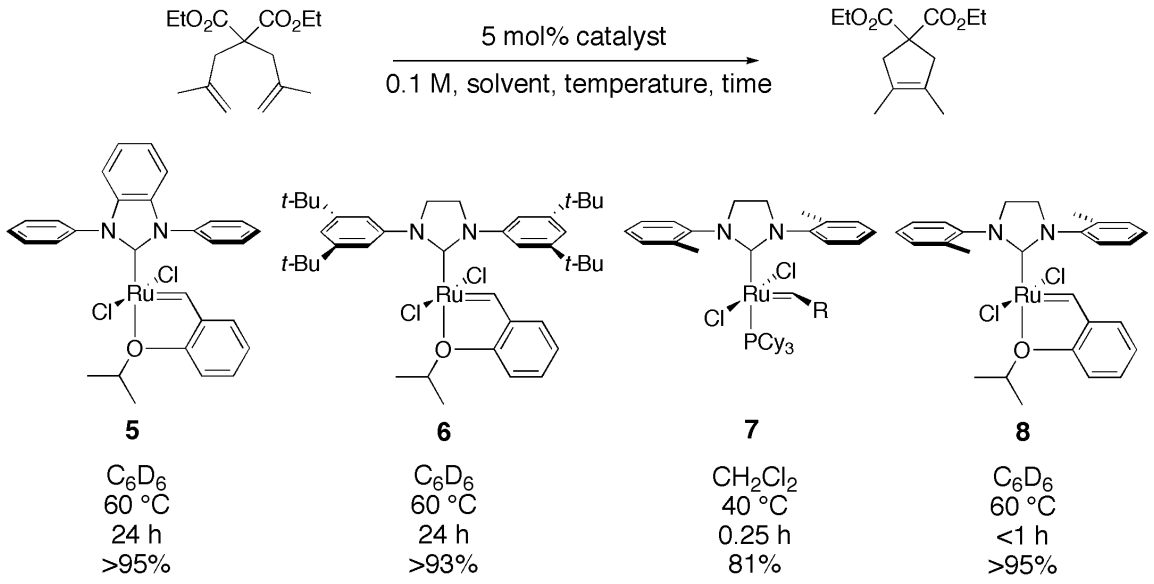
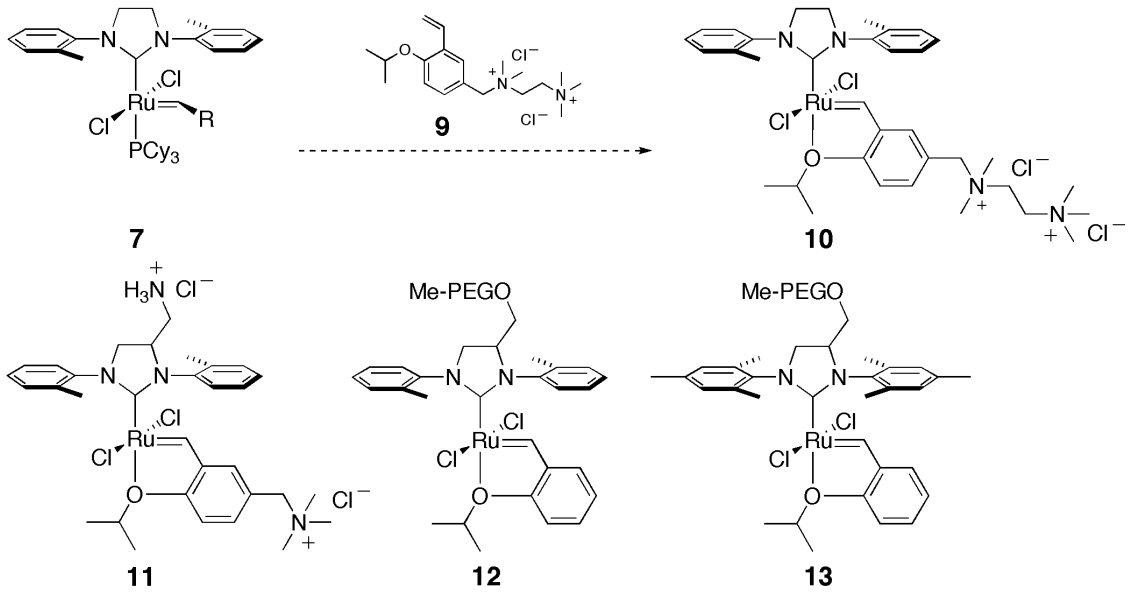


Figure 6.1. Reducing the steric bulk surrounding the ruthenium center produces more reactive metathesis catalysts, which are able to ring-close α,ω-dienes to produce tetrasubstituted olefins. The solvent, temperature, time, and conversion for the specified reaction is shown below each catalyst.

route provides a rapid method to examine this strategy for improved rates of catalysis in water. Should challenging aqueous cross-metathesis reactions with the proposed catalyst be successful, then the more-involved production of complexes **11** and **12**, analogs of water-soluble catalysts **2** and **13**,[10] may be warranted (PEG = poly(ethylene glycol)).

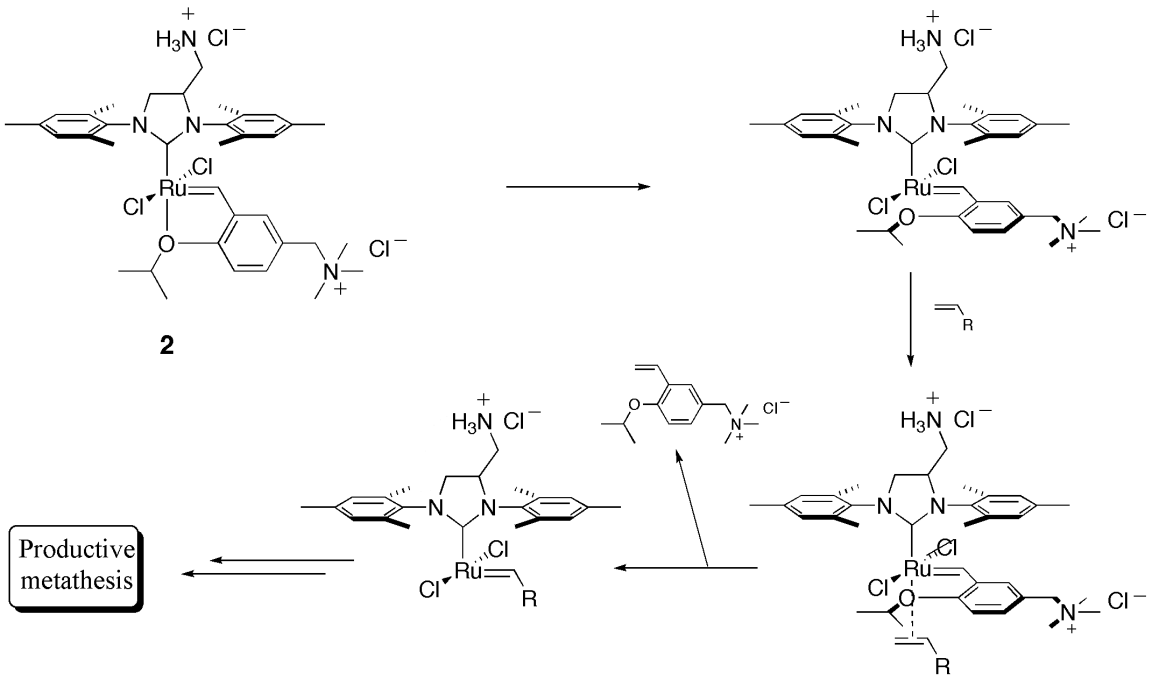


While the described method for improving catalyst reactivity can be rapidly examined, this author believes that the more promising strategy is to stabilize the catalyst against decomposition in water. Increased catalyst activity arises from increasing the ratio of the rate of productive catalysis over the rate of catalyst decomposition. The research reported in Chapters 4 and 5 shows that metathesis catalysts decompose more rapidly in water than in organic solvents. Therefore, in water, greater increases in the rate of productive catalysis are required to attain the same improvements in catalyst activity as observed for lesser rate increases in organic solvents. Also, the high stability of metathesis catalysts in organic solvents can accommodate a moderate increase in the rate of catalyst decomposition. However, for aqueous metathesis, the rate of decomposition is often similar to and even greater than the rate of productive metathesis, particularly for aqueous cross metathesis. In contrast, an increase in catalyst stability may provide aqueous metathesis reactions with sufficient time to progress to completion at a lesser rate.

Improving the stability of water-soluble metathesis catalysts. Chapters 4 and 5 reveal that the ruthenium benzylidene complexes, especially isopropoxybenzylidene complexes **1** and **2**, are highly stable toward water. However, Chapter 4 also indicates that ruthenium menthylidene and alkylidene complexes decompose rapidly in the presence of water. The limited capabilities of catalysts **1** and **2** to perform aqueous cross metathesis is further evidence for this conclusion. Therefore, strategies that increase the stability of the methylidene and alkylidene complexes may produce catalysts with increased activities in water.

The first step of productive metathesis is the dissociation of the ligand trans to the NHC.[11,12] For catalysts **1** and **2**, this ligand is the isopropoxybenzylidene's ether group. During productive metathesis, the isopropoxybenzylidene is released from the catalyst upon the reaction of the catalyst with substrate (Scheme 6.1),[13,14] and the ether group is insufficiently coordinating to stabilize the catalyst in the absence of chelation. Consequently, the methylidene and ethylidene analogs of catalysts **1** and **2** lack a ligand that can coordinate to the metal and stabilize these complexes against decomposition. Therefore, catalysts that provide such a ligand may show increased activities in water. Acknowledging the active role of phosphine in methylidene decomposition,[15,16] these catalysts should be phosphine-free.

Scheme 6.1.



A promising ligand scaffold that satisfies the stated requirements supports the catalyst with a chelating ligand that coordinates to the ether and chloride positions of

catalysts like **1** and **2** (Figure 6.2). The dissociating ligand of such catalysts remains coordinatively linked to the metal center and can serve to stabilize the methylidene and alkylidene complexes. Reported research for this type of system has produced catalysts **14**–**18** (Figure 6.2) whose dissociating ligands all chelate to the ruthenium center.[17-21]

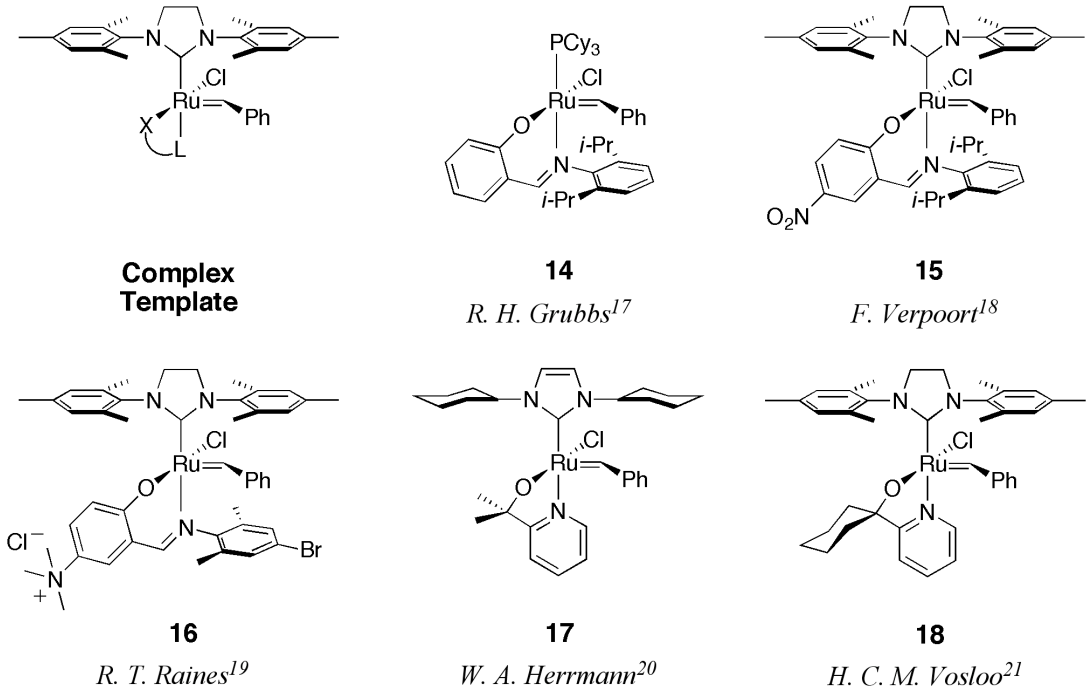


Figure 6.2. Ruthenium complexes based on the shown **Complex Template** are very stable olefin metathesis catalysts.

The first metathesis catalyst utilizing the proposed ligand scaffold was complex **14**, which employs a salicylaldimine ligand.[17] After 12 hours at 40 °C in methanol, catalyst **14** cyclizes the hydrogen chloride salt of diallylamine to the product five-membered ring in 95% conversion. This is the first example of ring-closing an α,ω-diene in a protic solvent and is the first demonstration of the potential for the proposed ligand scaffold in a polar protic environment.

More recent research has focused on incorporating chelating ligands onto ruthenium metathesis catalysts that contain an *N*-heterocyclic carbene ligand (**15**–**18**).[18-21] Catalysts reported by Verpoort and Raines include salicylaldimine ligands[18,19] while Herrmann and Vosloo describe catalysts supported by 2-pyridylcarbinols.[20,21] These catalysts are all highly stable. However, as may be expected, these catalysts are also far less reactive than catalysts containing isopropoxybenzylidene ligands and require high temperatures and/or long reaction times to mediate metathesis reactions.[18-21]

Slow dissociation of the ligand *trans* to the NHC is responsible for the poor reactivities observed with catalysts **15**–**18**.[19,20] However, the rate of catalyst initiation increases in more polar solvents,[11] and Chapter 4 shows that ruthenium complexes initiate more rapidly in the presence of water. Consequently, initiation with catalysts **15**–**18** may be sufficiently rapid in water to mediate metathesis at more moderate temperatures. Indeed, with catalyst **16**, Raines and co-workers show that catalyst activity for these systems is much higher in methanol/water mixtures than in nonpolar solvents.[19] Therefore, while the described ligand scaffold produces catalysts with poor reactivity in organic solvents, catalysts containing such ligands may strike the correct balance between stability and reactivity for aqueous metathesis.

The proposed chelating ligands are 2-pyridylcarbinols where the coordinated oxygen is presented as a phenoxide containing an ammonium salt *para* to the oxygen (Figure 6.3). 2-Pyridylphenols were chosen mainly due to salicylaldimine ligands being unstable toward water.[19] The indicated phenoxide is proposed for its increased acidity relative to alkoxides[22] and as a vehicle for incorporating a water-soluble functional group. Also, the pyridine rings of these ligands may be modified to increase complex initiation

by incorporating electron-withdrawing groups and/or steric bulk *ortho* to the nitrogen (Figure 6.3).

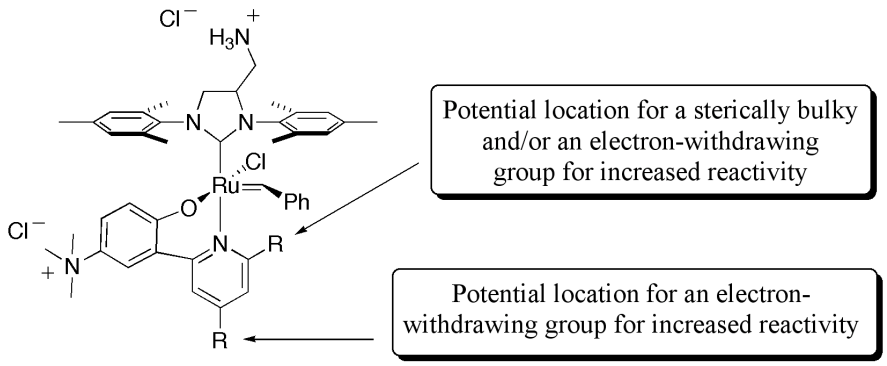
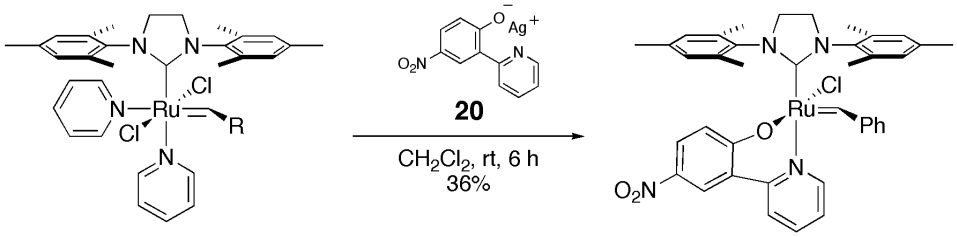


Figure 6.3. 2-Pyridylphenol supported ruthenium complexes are proposed as potentially stable water-soluble metathesis catalysts.

Results supporting this strategy. Preliminary research exploring the synthesis of a ruthenium complex containing the described ligand scaffold produced ruthenium complex **19**. Mixing the silver salt of 2-pyridylphenol **20**[23] with ruthenium bis(pyridine) complex **21**[24] in dichloromethane for three hours at room temperature gives **19** as a red solid. The ^{1}H NMR spectrum of **19** shows a single benzylidene resonance at 17.95 ppm, which is in excellent agreement with the published values for benzylidene resonances of NHC-containing catalysts supported by a 2-pyridylcarbinol.[20,21] This catalyst is very stable and is capable of ring-closing diethyl diallylmalonate in reagent grade dichloromethane, open to air to 85% conversion after 54 hours at 45 °C. To be applied to aqueous metathesis, complex **19** needs to be modified to incorporate water-soluble ionic groups.



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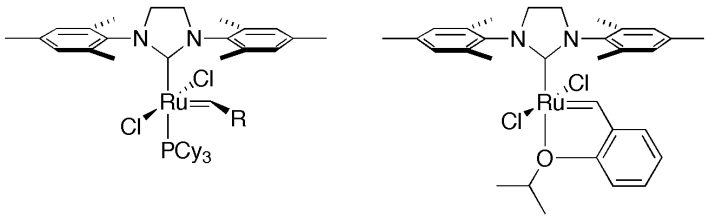
Potential Biological Applications of Aqueous Metathesis

Olefins are orthogonal to the functional groups displayed by the natural amino acids. Furthermore, techniques exist for the site-specific incorporation of unnatural amino acids displaying double bonds.[25-28] Therefore, olefin metathesis has the potential to provide a unique and useful method for the regioselective modification of proteins. However, polypeptides of biological interest are often only soluble in water, a solvent that does not dissolve commonly used, moisture-tolerant catalysts **22** and **23**. Because of their solubility and good activity in water, catalyst **1** and **2** provide the capability required to initiate an exploration of the potential for olefin metathesis in this area.



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Before venturing further, the impact of one aspect of protein research on the use of olefin metathesis in this field needs to be addressed. Solutions of proteins are usually very dilute with concentrations often ranging from nM to μM. These concentrations are much lower than the substrate concentrations employed in more traditional olefin

metathesis reactions.[2-8,29,30] The immediate implications of these low concentrations are twofold. First, metathesis reactions on proteins may require extended reaction times, which can place an increased emphasis on catalyst stability. However, this pressure on catalyst stability is moderated by the second implication of low protein concentrations, which is that water-soluble catalysts can be used in stoichiometric quantities for this application. More than that, the dilute concentrations of protein solutions even allow for the metathesis "catalyst" to be used in heavy excess for these transformations without committing exorbitant quantities of catalyst.

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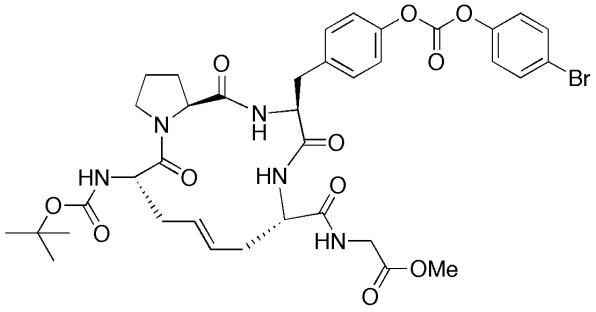
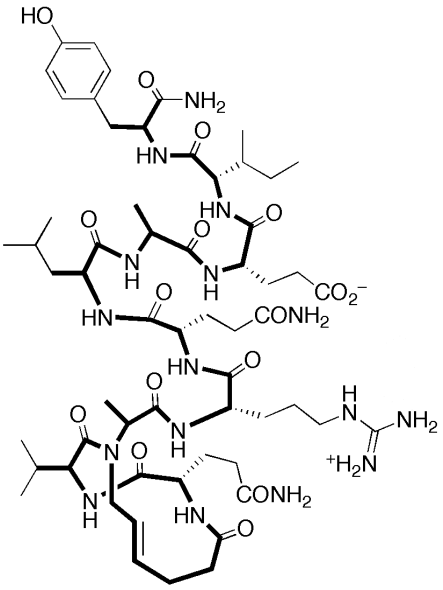


R. H. Grubbs[31]

Stabilized peptide α-helix:



P. S. Arora[38]

Figure 6.4. Olefin metathesis has been used to stabilize β-turn and α-helical secondary structures of short peptide chains.

Stabilizing protein secondary structure by olefin metathesis. Two general applications of catalysts **1** and **2** to modify protein structure will be presented. First, Chapter 1

discusses the use of olefin metathesis to stabilize two secondary structure motifs of short peptides, β-turns and α-helices (Figure 6.4).[31-38] The solubility of catalysts **1** and **2** in water allows for this application of metathesis in biology on olefin-dsplaying proteins as opposed to the model peptides used in the reported research. As these reactions can be considered examples of ring-closing metathesis, the demonstrated competency of catalysts **1** and **2** for this transformation in water makes this a particularly enticing application for the currently available catalysts.

Modifying proteins with probe molecules by olefin metathesis. Another potential application of catalysts **1** and **2** in protein modification is the use of olefin metathesis to regioselectively incorporate probes onto proteins (Figure 6.5). These probes may include chromophores for improved protein detection or molecules like biotin for simpler protein isolation.

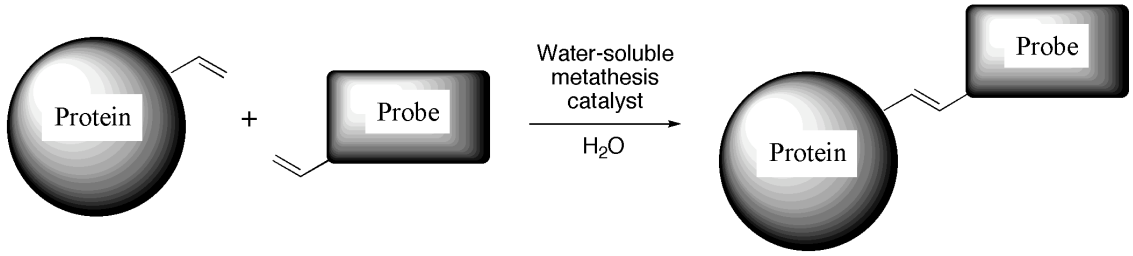


Figure 6.5. Olefin metathesis in water can potentially regioselectively modify proteins with probe molecules.

This application presents two challenges for aqueous metathesis. First, efficient modification reactions will likely require excess quantities of the probe molecule. Consequently, probe dimerization may decrease the efficiencies of the desired transformation by enabling catalyst decomposition pathways that are avoided by the stable, uninitiated isopropoxybenzylidene complex.[9] This hurdle can likely be overcome

by using probe molecules that contain olefins that participate in cross-metathesis reactions but do not homodimerize or homodimerize very slowly.[29]

The low protein concentrations commonly encountered in this area of research presents a second challenge for regioselectively incorporating probes onto proteins. The results presented in Chapter 5 reveal that isopropoxybenzylidene complexes **1** and **2** are quite stable in water. However, as already mentioned, the limited aqueous cross-metathesis activity of catalysts **1** and **2** indicates that the alkylidene complexes formed by the reaction between these catalysts with a terminal olefin are not stable in water. Successfully crossing probe molecules onto proteins requires the alkylidene complex produced by the reaction of the protein with the catalyst to persist for an extended period of time. The most direct solution to this obstacle is to develop a more stable aqueous metathesis catalyst. Even so, a different strategy may allow the use of already-developed water-soluble catalysts **1** and **2**.

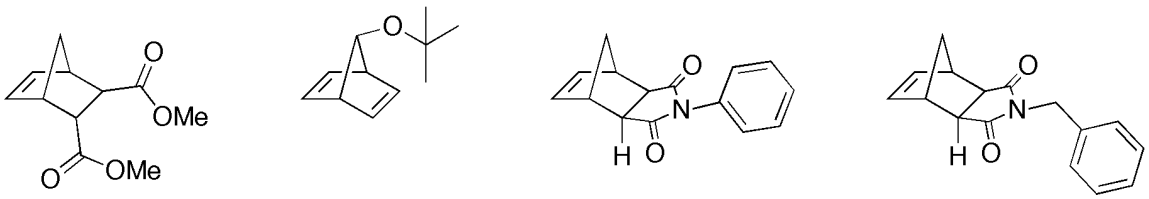
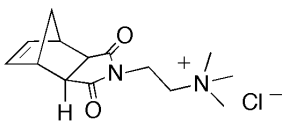


Figure 6.6. Monomers that Khosravi and co-workers have shown to chelate to ruthenium during their ROMP with catalyst **22**.[41,42]

Norbornene monomers containing esters have long been thought to coordinate to ruthenium catalysts during ROMP.[39-47] For example, Khosravi and co-workers have reported observing NMR evidence for chelating alkylidenes during the ROMP of various oxygen containing norbornene monomers (Figure 6.6).[41,42] Furthermore, Grubbs and co-workers observed that alkylidenes formed during the ROMP of *exo*-norbornene monomer

24 with catalysts **3** and **4** is stable for three months in the presence of monomer,[43] though both catalysts rapidly decompose in water in the absence of substrate.[44,45] Also, as described in Chapter 2, the alkylidene formed during the ROMP of *endo*-norbonene monomer **25** with catalyst **26** is stable for at least two days in water.[46,47] Such chelation events may serve to stabilize the alkylidene complexes formed during the modification of proteins with catalysts **1** and **2**.



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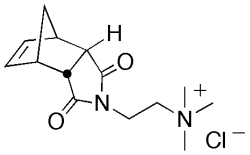

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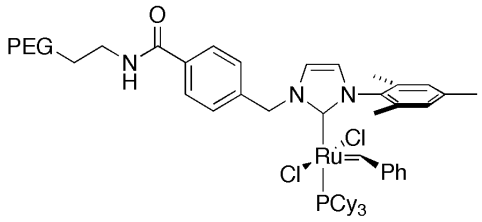

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Results that support the described strategy of protein modification. A particularly attractive probe olefin that dimerizes slowly is an acrylamide.[29,48] Preliminary research has shown that these olefins do show activity for aqueous metathesis. NMR and mass-spectral analysis reveal that catalyst **1** can cross allyl alcohol onto acylamide in water, though not catalytically. Therefore, catalysts **1** and **2** should be able to cross acrylaminde-containing probe molecules onto proteins.

Also, further evidence supporting the hypothesis that norbornenes stabilize ruthenium alkylidenes has been obtained. During the ROMP of **27** with catalyst **22** a new alkylidene resonance is observed in the ^{1}H NMR spectrum while the ^{31}P NMR only contains resonances for free tricyclohexylphosphine and uninitiated catalyst **22**. The chemical shift of this alkylidene resonance, 17.25 ppm, is consistent with similar

complexes containing a chelating alkylidene such as complex **23** (Figure 6.7(A)).[13,49-51]

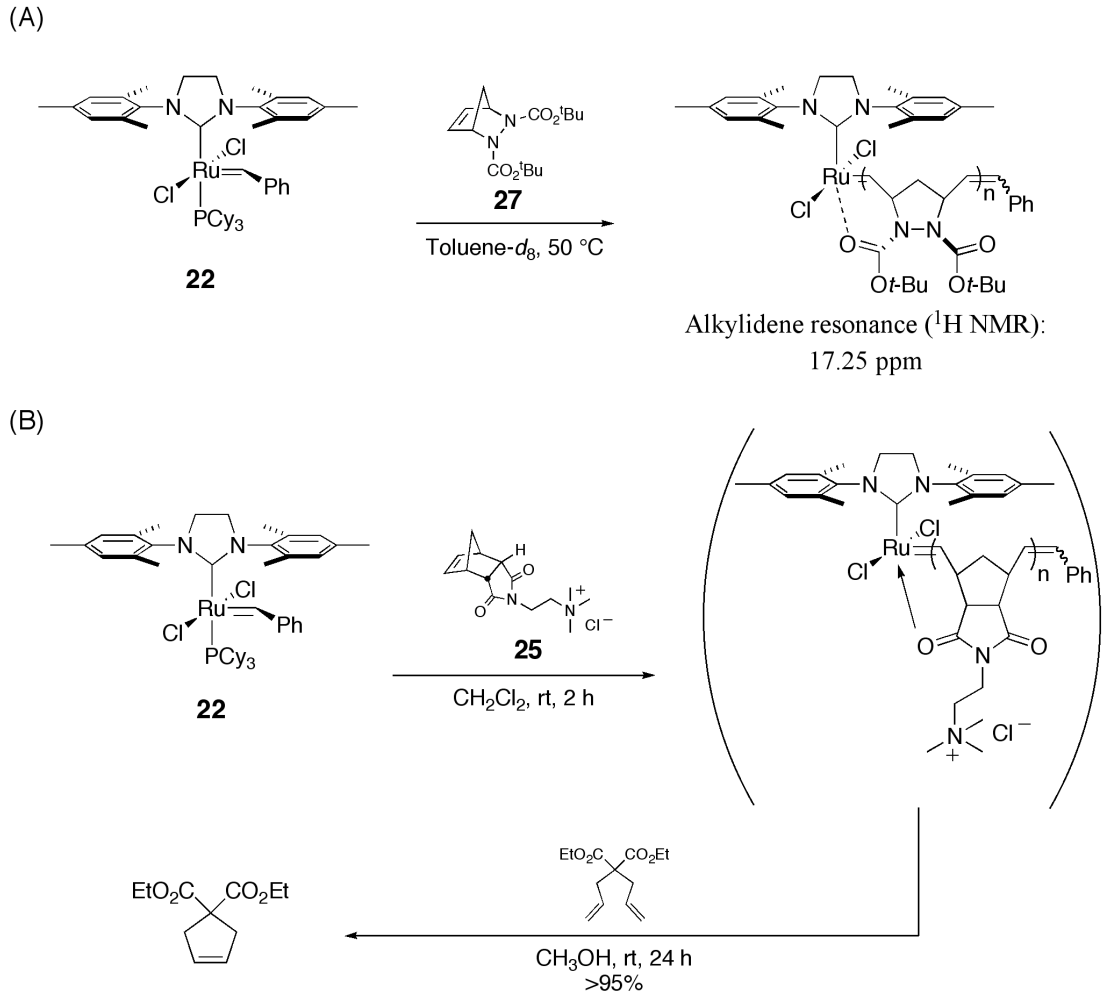


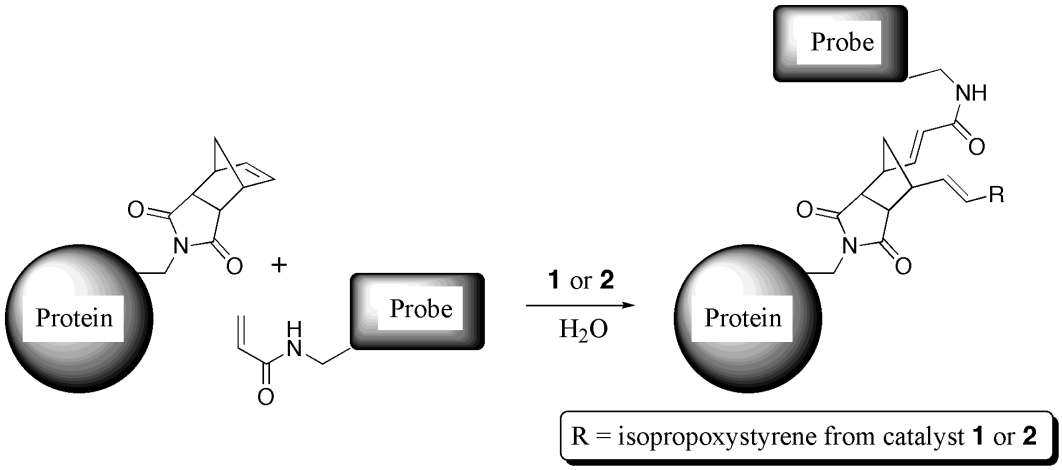
Figure 6.7. (A) NMR spectral analysis indicates that the propagating alylidene formed during the ROMP of monomer **27** with catalyst **22** is stabilized by chelation. (B) The reaction of catalyst **22** with monomer **25** produces an isolable mixture of complexes that ^{31}P NMR indicates is phosphine-free. This mixture of complexes is able to quantitatively ring-close diethyl diallylmalonate in methanol within 24 hours at room temperature.

Additionally, the reaction of catalyst **22** with monomer **25** in dichloromethane produces a stable mixture of complexes that can be isolated (Figure 6.7(B)). Interestingly, while the ^{1}H NMR spectrum of this product mixture contains four alkylidene resonances, the ^{31}P NMR spectrum indicates the absence of any species containing phosphorus. Therefore,

the produced complexes are phosphine-free and are likely stabilized by a chelating oxygen (Figure 6.7(B)). These complexes are soluble in methanol and water and are capable of quantitatively ring-closing diethyl diallylmalonate in methanol within 24 hours at room temperature. As a whole, this evidence suggests that norbornenes containing coordinating oxygens may sufficiently stabilize alkylidene complexes formed with catalyst **1** or **2** to allow for their application to the modification of proteins.

Proposed method for using catalysts 1 and 2 to incorporate probe molecules onto proteins. The complete strategy for regioselectively incorporating probe molecules onto proteins is presented in Scheme 6.2. A water-soluble olefin metathesis catalyst such as complex **1** or **2** could be used to cross an acrylamide-containing probe molecule onto a protein displaying a norbornene, which contains coordinating oxygens.[52] The choice of protein and probe molecules can be varied as desired. However, bovine serum albumin (BSA) provides a readily available platform to examine the viability of this strategy.

Scheme 6.2.



Bovine serum albumin is a heavily researched protein present in cow blood, which is available in large quantities from commercial sources.[53,54] BSA contains a single cysteine that does not participate in a disulfide bridge.[53,54] This thiol group can be used

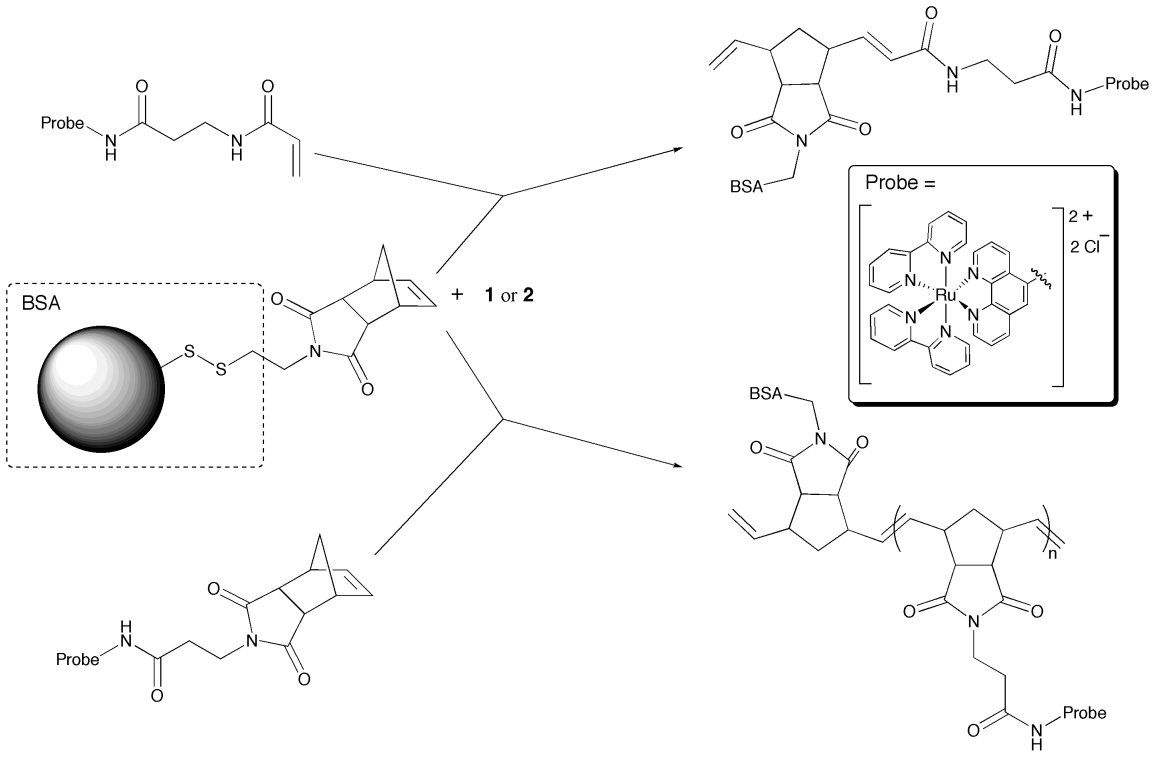


Figure 6.8. Crossing ruthenium dyes onto the protein BSA is proposed as a system for proof-of-concept research on the described strategy for employing water-soluble catalysts **1** and **2** to the regioselective modification of proteins.

to decorate BSA with various molecules by the formation of a disulfide bond. For example, Maynard and co-workers have recently used this thiol to incorporate atom transfer radical polymerization initiators onto BSA, which they employed to grow polymers from this protein.[55] The same methodology could be utilized to include the desired norbornene molecule onto BSA (Figure 6.8). Water-soluble catalysts **1** and **2** may then mediate metathesis reactions between this protein with an acrylamide-displaying

probe molecule. Acrylamide-displaying analogs of the ruthenium dyes used by Kuo and Grubbs in aqueous cross metathesis can readily serve as probe molecules for these experiments (Figure 6.8).[56] Alternatively, norbornenes can also be covalently attached to these ruthenium dyes. The propagating alkylidene produced during the ROMP of this probe norbornene can react with the norbornene-containing BSA to incorporate multiple probe molecules onto a single protein. If these proof-of-concept experiments succeed, methods for site-specifically incorporating unnatural amino acids displaying a "coordinating norbornene" need to be developed for this strategy to have practical utility for protein modification.

Summary

The development of catalysts **1** and **2** represent significant progress in the ability to perform homogenous metathesis in water. However, their limited ability to mediate aqueous cross metathesis presents an opportunity for future catalyst development. Reducing the steric bulk around the ruthenium center may produce catalysts with increased reactivity in water. Alternatively, 2-pyridylphenol ligands may be used to improve the stability of catalysts in water.

Despite their limited activity in aqueous cross metathesis, catalysts **1** and **2** might be used to modify the structure of proteins. Aqueous ring-closing reactions on proteins containing unnatural amino acids, which present carbon-carbon double bonds, may be used to stabilize such protein structural motifs as β-turns and α-helices. Also, appropriately modifying protein and probe molecules with well-chosen olefins may allow catalysts **1** and **2** to incorporate the probe molecule onto the protein, and BSA provides an excellent platform to examine this strategy of protein modification.

In conclusion, catalysts **1** and **2** provide a glimpse of the potential for olefin metathesis in water, which is a field rich in possibility. Catalysts capable of competently mediating the full range of metathesis transformations in water appear to be an attainable goal. Once developed, a variety of applications exist for such catalysts, particularly in biology. Therefore, olefin metathesis in water will surely be the subject of future research.

Acknowledgements

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Experimental

General considerations. All glove-box manipulations were performed in a N_2-filled Vacuum Atmospheres glove box (O_2 < 2.5 ppm). Otherwise reactions run under dry, degassed conditions were performed using standard Schlenk techniques under an atmosphere of dry argon using flame or oven-dried glassware. NMR spectral analysis of the products for the cross-metathesis reaction between acrylamide and allyl alcohol and the ROMP of **27** with catalyst **22** was performed on an Inova 500 (499.85 MHz for ^{1}H; 202.34 MHz for ^{31}P; 125.69 MHz for ^{13}C). All other NMR spectra were recorded on a Varian Mercury 300 (299.817 MHz for ^{1}H, 75.4 MHz for ^{13}C, and 121 MHz for ^{31}P) and reported in parts per millon (ppm) downfield from trimethylsilane as referenced to

residual protio solvent peaks. Multiplicity abbreviations used when reporting ^{1}H NMR spectra are: s = singlet, and br = broad.

Materials. All deuterated solvents were purchased from Cambridge Isotope Laboratories. Deuterated dichloromethane and deuterated toluene were dried over 4 Å molecular sieves, and deuterated methanol was dried over calcium sulfate. Deuterated methanol and deuterated dichloromethane were degassed by three freeze, pump, and thaw cycles while deuterium oxide and deuterated toluene were degassed by a generous argon sparge. Anhydrous methanol was purchased from Aldrich and degassed with a generous argon sparge. All other solvents were purchased from Fischer Scientific. Solvents were dried by passage through purification columns packed with alumina and degassed by a generous argon sparge. All commericial materials were used as obtained. The synthesis of ruthenium complexes **1** and **2** and isopropoxystyrene **9** were described in Chapter 5. Ruthenium complex **22** was a gift from Materia Inc. The syntheses of *endo*-norbornene monomer **25** and ruthenium complex **26** were described in Chapter 2. Sodium methoxide, neutral Brockman grade I alumina, acrylamide, and allyl alcohol were purchased from Aldrich. Silver nitrate was purchased from Strem. Diethyl diallylmalonate was purchased from Avocado. 2-Pyridylphenol **20** was the gift of Prof. Brian Connell. Monomer **27**,[57] and ruthenium bis(pyridine) complex **21**[24] were synthesized according to literature procedures.

Ruthenium complex 19. A flame-dried round-bottom flask, containing compound **20** (304.2 mg, 1.4 mmol), was brought into a N_2-filled glove box, charged with sodium

methoxide (79.1 mg, 1.5 mmol, 1.1 equiv), equipped with a stir bar and sealed with a septum. This flask was removed from the glove box, brought under a positive argon pressure and cooled to 0 °C. Dry, degassed methanol (5 mL) was slowly added by syringe, and the reaction mixture was stirred at 0 °C for 10 minutes, warmed to room temperature, and stirred for an additional 2.5 hours. Upon reaction completion, the volatiles were removed by rotary evaporation. The brown solid product was dissolved in water (3 mL), and silver nitrate (505.9 mg, 3.0 mmol, 2.1 equiv) was added. This reaction mixture was stirred for 10 minutes. The silver salt, which precipitates during the reaction, is isolated by vacuum filtration and generously rinsed with water, benzene, and diethyl ether in that order. The product was dried under high vacuum to obtain 389.4 mg (86%) of the silver salt as a brown solid. (Note: this salt is light sensitive.) In a N_2-filled glove box, the silver salt of 2-pyridylphenol **20** (10.2 mg, 0.032 mmol) and ruthenium bis(pyridine) complex **21** (23.4 mg, 0.032 mmol, 1.0 equiv) were weighed into a 1-dram vial. This vial was equipped with a stir bar, charged with dry, degassed deuterated dichloromethane (0.77 mL) and sealed with a septa-cap. The reaction mixture was stirred for 6 hours before removing the vial from the glove box. The product mixture was passed through a short column of neutral alumina with dichloromethane, rinsed with *n*-pentane, and dried under high vacuum to obtain 8.2 mg (36%) of complex **19** as a dark-red solid. ^{1}H NMR (CD_2Cl_2, ppm, benzylidene resonance): δ 17.95 (s).

Ring closing diethyl diallylmalonate with catalyst 19. Catalyst **19** (8.2 mg, 0.011 mmol, 0.052 equiv) was dissolved in reagent grade deuterated dichloromethane (0.7 mL) and transferred to an NMR tube. Diethyl diallylmalonate (50 μL, 0.21 mmol) was

injected by syringe, and the reaction mixture was heated to 45 °C. The reaction conversion was followed by ^{1}H NMR spectroscopy.

Stoichiometric cross metathesis of allyl alcohol and acrylamide in water with catalyst 1. In a N_2-filled glove box, a 1-dram vial was charged with catalyst **1** (5.6 mg, 0.0066 mmol, 1.1 equiv), equipped with a stir bar and sealed with a septa-cap. The vial was removed from the glove box and brought under a positive argon pressure. An aliquot (0.15 mL) of a solution of allyl alcohol (10 mL) and acrylamide (16.3 mg) in degassed deuterium oxide (2.9 mL) was added to this vial by syringe. (Actual reaction contained 0.0063 mmol of allyl alcohol and 0.012 mmol (1.9 equiv) of acrylamide.) The reaction was stirred at 30 °C for 16 hours under a positive argon pressure, and the product mixture was examined by ^{1}H NMR and mass spectral analysis. The cross-product was estimated to form in 36% conversion from the ^{1}H NMR spectrum.

ROMP of monomer 27 with catalyst 22. In a N_2-filled glove box, a screw-cap NMR tube was sealed with septa-cap, and monomer **27** was weighed into a round-bottom flask, which was then sealed with a septum. This flask and the NMR tube were brought out of the glove box, and a positive argon pressure was applied to the monomer-containing flask. Dry, degassed deuterated toluene was transferred to the monomer-containing flask using standard Schlenk techniques to produce a 0.6 M monomer solution. An aliquot (0.6 mL) of this solution was thermostated at 55 ºC for 10 minutes in the NMR spectrometer prior to the addition of an aliquot (0.10 mL) of a solution of catalyst **22** (25 mg) in dry, degassed deuterated toluene (0.25 mL). The reaction mixture was mixed by three tube

inversion and reinserted into the NMR spectrometer. The reaction progress was followed by ^{1}H NMR spectroscopy.

Complex mixture formed by the ROMP monomer 25 with catalyst 22. In a N_2-filled glove box, ruthenium complex **22** (29.6 mg, 0.035 mmol) and monomer **25** (50.2 mg, 0.18 mmol, 5.1 equiv) were weighed into a 1-dram vial. The vial was equipped with a stir bar and charged with dry, degassed dichloromethane (2 mL). The reaction mixture was allowed to stir for 2 hours before removing the vial from the glove box and isolating the product by centrifuge. Drying under high vacuum provides 54.7 mg of a brown, solid product. ^{1}H NMR (CD_3OD, ppm, benzylidene resonances): δ 18.86 (br, relative integral 1.00), 18.09 (br, relative integral 1.03), 18.02 (br, relative integral 0.39), 17.93 (br, relative integral 2.03). Mixing this solid (15 mg) with diethyl diallylmalonate (50 μL, 0.21 mmol) in dry, degassed methanol (0.6 mL) yields 95% conversion of the ring-closed product after 24 hours at room temperature.

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