JOC The Journal of Organic Chemistry

Note

Ring-Closing Metathesis on Commercial Scale: Synthesis of HCV Protease Inhibitor Simeprevir

Andras Horvath, Dominique Depré, Wim A. A. Vermeulen, Stijn Leopold Wuyts, Syuzanna R Harutyunyan, Gregori Binot, Jef Cuypers, Wouter Couck, and Dirk Van Den Heuvel

J. Org. Chem., **Just Accepted Manuscript** • Publication Date (Web): 05 Feb 2019

Just Accepted

ACS Publications

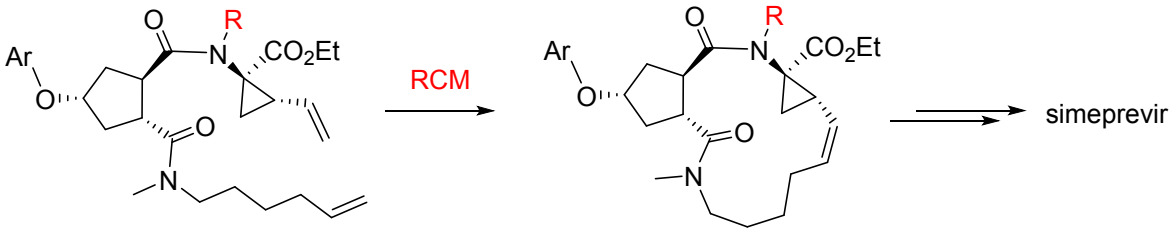
is published by the American Chemical Society. 1155 Sixteenth Street N.W., Washington, DC 20036

Ring-Closing Metathesis on Commercial Scale: Synthesis of HCV Protease Inhibitor Simeprevir®

András Horváth,* Dominique Depré,* Wim A. A. Vermeulen, Stijn L. Wuyts, Syuzanna R. Harutyunyan,† Grégori Binot,§ Jef Cuypers, Wouter Couck and Dirk Van Den Heuvel

Janssen Pharmaceutica N.V., 30 Turnhoutseweg, B-2340 Beerse, Belgium

Supporting Information Placeholder



R= H: c= 0.01 M, GH1, 47% **thermodynamic control**

R= Boc: c= 0.05 M, M2, 80% **kinetic control (SHD conditions)**

ABSTRACT: The key macrocyclization step in the synthesis of simeprevir, a hepatitis C virus (HCV) antiviral drug, was studied. *N*-Boc substitution on the diene precursor changes the site of insertion of the metathesis catalyst, and, consequently, the kinetic model of the ring closing metathesis (RCM), enabling a further increase in the macrocyclization efficiency under simulated high dilution (SHD) conditions. NMR of the inserted species of both 1st and 2nd generation RCM catalysts are reported and discussed.

Hepatitis C virus (HCV) infection afflicts approximately 150 million people worldwide, according to a recent survey.[1] Significant research efforts in the past decade towards novel chemotherapies led to the introduction of the direct acting antivirals (DAAs) which are medications targeted at specific steps within the HCV life cycle. Simeprevir (**1**, TMC435, figure 1) is a HCV NS3/4A protease inhibitor discovered by Medivir and Tibotec, and developed and launched by Janssen Pharmaceutica in 2013. Its key novel structural features are the cyclopentane core unit – reducing the peptidic character of simeprevir relative to compounds with a central hydroxyproline unit – and the 14-membered macrocycle, which define its unique binding properties.[2,3] It has activity in the low nanomolar range, clean side-effect profile and excellent pharmacokinetic properties, making it possible to achieve low once-daily therapeutic doses with high levels of sustained virologic response in a broad patient population.

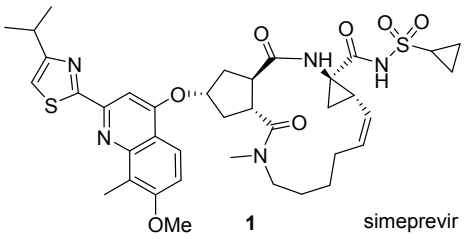
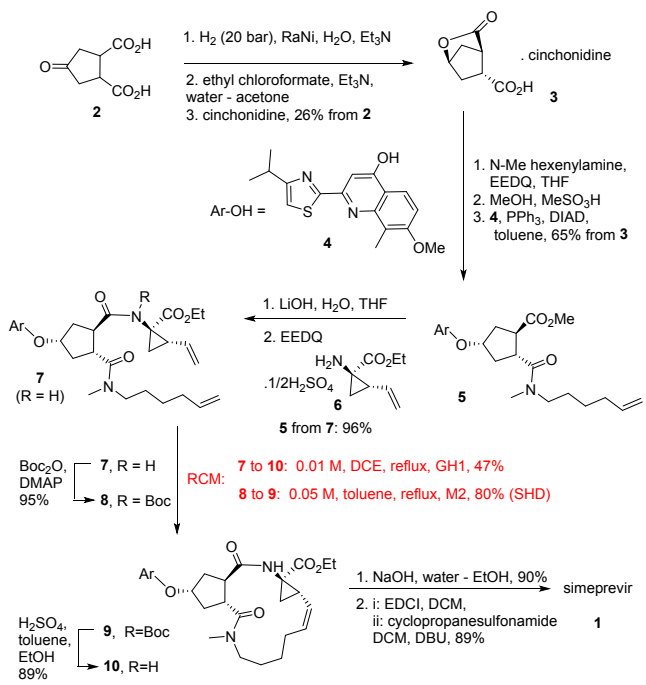


Figure 1. Structure of simeprevir

Our synthesis of simeprevir is depicted in scheme 1.[4] *trans*-Cyclopentanone-3,4-dicarboxylic acid **2**[5] was hydrogenated over Raney Ni as its triethylamine salt. The resulting hydroxy-diacid was cyclized to the corresponding lactone using ethyl chloroformate in water-acetone, which was resolved and isolated as the highly crystalline cinchonidine salt **3** in 26 % yield and 97% enantiomeric purity from **2**. Amide coupling of **3** with *N*-methylhexenylamine[6] using N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ), ring opening of the lactone by methanolysis and coupling of the secondary alcohol with the quinoline **4**[7] by a Mitsunobu reaction afforded the crystalline intermediate **5** in 65 % yield from **3**. Hydrolysis of the ester function of **5** and amidation with the aminocyclopropane **6**[8] gave the key diene intermediate **7** which was cyclized by Ring-Closing Metathesis (RCM) to the 14-membered macrocycle **10**. Hydrolysis of the ester function in **10**, activation of the carboxylic function with 1-ethyl-3-(3-dimethyl-aminopropyl) carbodiimide (EDCI) and coupling with cyclopropylsulfonamide gave simeprevir. Much of our focus was directed towards the RCM step, which was marred by the known problem of low volume efficiency, high catalyst load and extreme sensitivity to starting material quality, thus jeopardizing robustness. We wish to report here our results in preliminary form to highlight important observations on our substrate that radically depart from the results previously published in the recent literature on the process development of ring-closing metathesis.[9]



Scheme 1. The large scale synthesis of simeprevir

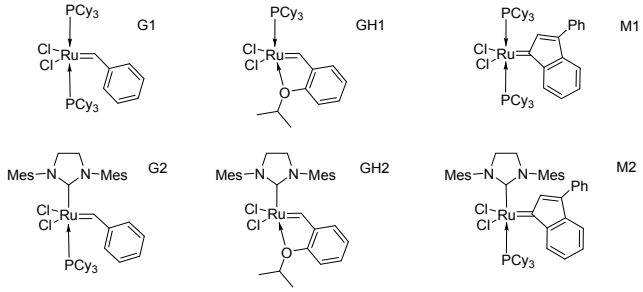
In our initial experiments based on the medicinal chemistry procedure, diene **7** was cyclized with the GH1 catalysts in refluxing 1,2-dichloroethane (DCE). The yield, as expected, was highly dependent on substrate concentration. The relationship between yield and concentration was studied under batch conditions (all materials added at the beginning of the reaction) and is presented in Figure 2. Conversions were above 95 % using 2.5 mol% of catalyst at all concentrations higher than 0.1 M,[10] but yields were low at concentrations practical for large scale application. A screen of five other commercially available Ru metathesis catalysts did not offer any improvement (see Table 1). The other 1st generation catalysts G1 and M1 produced a large amount of side products mainly due to the known epimerization reaction[11] at the vinylic carbon center on the cyclopropane. The 2nd generation catalysts G2, GH2 and M2 gave incomplete conversions with diene **7**. Notably however, no cyclopropane epimerization was seen with any of the 2nd generation catalysts.

At this point in development we chose to optimize the RCM of diene **7** using the GH1 catalyst. Disappointingly, we observed varying amounts of epimerization (8 to 25 %) in our first batches using GH1 catalysts, depending on the purity of the diene starting material.[12] This phenomenon was assumed to be due to the presence of residual bases coming from the upstream steps. Indeed, it has been reported that the presence of Brønsted[9a] or Lewis bases[11] in the reaction mixtures retard the RCM reaction and allow the competing epimerization pathway to become dominant. Reasoning that protonation of any residual base would reduce epimerization, we solved this problem by addition of 10 mol% of methanesulfonic acid (found to be the best from a range of acids tested) directly to the metathesis reaction mixture. This resulted in the stabilization of the levels of epimerized byproduct at 2-3 % and allowed for a significant (8-10 %) improvement in the average RCM yields of the large scale batches, while allowing the catalyst load to be kept at 2.5 mol%.

However, at the 0.01 M concentration - chosen as a compromise between yield and throughput - the reaction still produced large amounts of polymers (10-18 %) with high molecular mass, as shown by gel permeation chromatography (GPC) analysis. No crystallization method was found to remove these polymers and intensive purification, a combination of charcoal treatment and chromatography, was required to obtain the macrocycle in sufficient purity. Recently, we developed an efficient method to remove high-molecular-weight polymers by organic solvent nanofiltration,[13] which greatly simplified the product isolation and purification after the RCM step. However, the high dilution issue still called for further development work.

Table 1. RCM catalyst screening

	c (M)	G1	GH1	M1	G2	GH2	M2
7 to **10**, %	0.1	2	17	12	8	12	12
	0.01		71		41		43
8 to **9**, %	0.1	<2	4	5	55	45	58
	0.01		23		78		84



In situ yields of the RCM reaction of **7** and **8** under batch conditions using commercially available catalysts. G1, GH1 and M1 (2.5 mol%) were tested in DCE at reflux, G2, GH2 and M2 (1.5 mol%) were tested in toluene at reflux. Toluene and DCE respectively were selected as best solvents from a screening, for the shown catalyst-substrate combinations.

Increasing the efficiency of macrocyclizations has been studied extensively in the literature, by reaction engineering methods, such as Ziegler's simulated high dilution (SHD) method,[14] using a continuous stirred tank reactor (CSTR) rather than a batch reactor, or removing the reaction product by distillation,[15] templating[16] or encapsulating[17] agents and substrate tailoring.[18] Applicability of one or another method (or combinations thereof) is a function of the structure of the substrate and the kinetic model of the reaction.

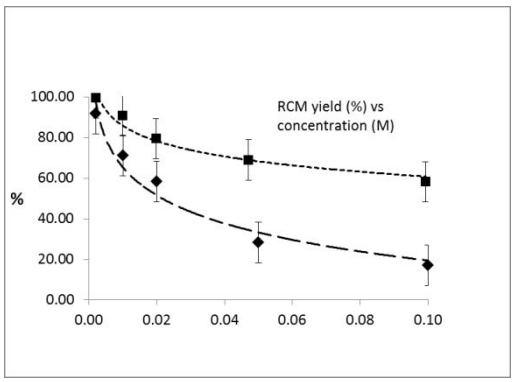


Figure 2. *In situ* yield *vs* concentration profile of the macrocyclization of **7** to **10** with GH1 (♦) and **8** to **9** with M2 (■),

as a function of the initial concentration of the dienes (2.5 mol% catalyst, batch conditions).

To learn more about the kinetic model, we next probed the reversibility of the reactions of the metathesis manifold under our reaction conditions. Treatment of macrocycle **10** with GH1 at different start concentrations led to slow conversion of the macrocycle into polymers (as shown by GPC analysis), showing that the ring opening metathesis polymerization (ROMP) reaction of the metathesis manifold was active under these conditions. Treating **10** with GH1 under 3 bar ethylene pressure resulted in > 90 % conversion to the diene **7** in six hours, whereas the similar ethenolysis of reaction mixtures containing the cyclic monomer **10** and polymers resulted in complete ethenolysis of the cyclic monomer **10** and substantial depolymerization of the polymeric species. For the acyclic diene metathesis (ADMET) and the ROMP reactions of the manifold, complete reversibility could not be shown - probably due to catalyst poisoning by the polymeric species. However, the above observations are consistent with thermodynamic control. This excludes the possibility of improving the yield for the reaction of diene **7** with GH1 by applying SHD conditions, since under thermodynamic control, the yield is determined by the final concentration alone.

Thus, we decided to address the problem of high dilution by substrate modification. Weiler *et al.* synthesized a series of 14-membered macrocyclic amides by RCM and found that the cyclization of the *N*-Boc derivatives of the amide dienes gave consistently higher macrocycle yields than the parent secondary amides.[19] This effect was later studied and exploited by Boehringer-Ingelheim researchers for the synthesis of some 15-membered macrocyclic NS3 protease inhibitors.[20,21]

We thus prepared the *N*-Boc-modified substrate **8** by a standard DMAP-catalyzed reaction of the parent diene amide **7** with Boc_2O. The cyclization of **8** to **9** was compared to that of **7** to **10** with the same six commercial catalysts (Table 1). With the *N*-Boc modified substrate **8**, the 1st-generation catalysts failed to give the expected improvement in macrocyclization efficiency, giving sluggish and incomplete reactions. However, the 2nd generation catalysts performed significantly better with diene **8** vs. **7** and allowed for an increase in the effective molarity[22] (EM), as well as reaction rate. Reaction time under the best conditions for **8** (M2 catalyst in refluxing toluene) was 0.5 h, whereas under the best conditions for **7** (GH1 in refluxing DCE) the cyclization took 6 h to complete. Importantly, there was no cyclopropane epimerization seen on **8** with any of the catalysts studied and the main side reaction was formation of dimers, rather than higher polymers (as measured by GPC). The yield-concentration profiles for the cyclization of **7** to **10** and of **8** to **9** are compared in Figure 2.

Surprisingly, we found that the RCM of **8** with the M2 catalyst proceeds under kinetic control, as neither the ethenolysis nor the ROMP of **9** gave measurable conversions with 1.5 mol% catalyst in refluxing toluene in separate experiments (catalyst initiation was ensured by the addition of 1 mol% of diene **8**). These results contrast with those obtained with the RCM of **7** with GH1 (thermodynamically controlled as explained above). For the synthesis of ciluprevir (BILN-2061), kinetics of the RCM changed in the opposite way: the reaction switched from kinetic control to thermodynamic control upon *N*-Boc-substitution.[20]

We want to stress that the RCM of **8** vs **7** is unique and markedly different from that of the analogous 15-membered proline-based dienes.[20,21] A significantly lower cyclization efficiency in the cyclization of the dienes in our study is seen due to the additional *trans*-element introduced by the cyclopentane core of simeprevir, as well as due to the somewhat higher ring tension of the 14- *vs* 15-membered cycles.[23]

The cyclization of *N*-Boc diene **8** at room temperature is quite slow and consequently, we could observe and compare the species of the insertion of both G1 and G2 by NMR for both dienes. To the best of our knowledge, this is the first literature report on the site of initiation of dienes with a 2nd generation catalyst for a productive RCM reaction. Previously reported NMR studies with 2nd generation catalysts were performed with "trapping olefins" or chelating type substrates designed to avoid cyclization and minimize polymerization.[24]

NMR analysis showed that insertion of the 1st generation catalyst occurs preferentially at the vinylcyclopropane double bond with the unsubstituted diene (as observed in other, related substrates),[20] whereas there is exclusive insertion at the hexenyl side for the Boc-substituted diene (Figure 3). In marked contrast, the 2nd generation catalyst inserts only at the hexenyl double bond for both substrates. This clearly explains why the cyclopropane epimerization only occurs with the 1st generation catalyst on **7**, and was not seen in the other substrate-catalyst combinations. It is also remarkable and unexpected that the generally more reactive 2nd generation catalysts are not capable of inserting in the double bond of the macrocycle, which explains the switch to a kinetically controlled reaction for the **8**-G2 or **8**-M2 cases. On the other hand, GH1 inserts readily in the double bond of **10**, resulting in thermodynamic control for the cyclization of **7** with GH1.

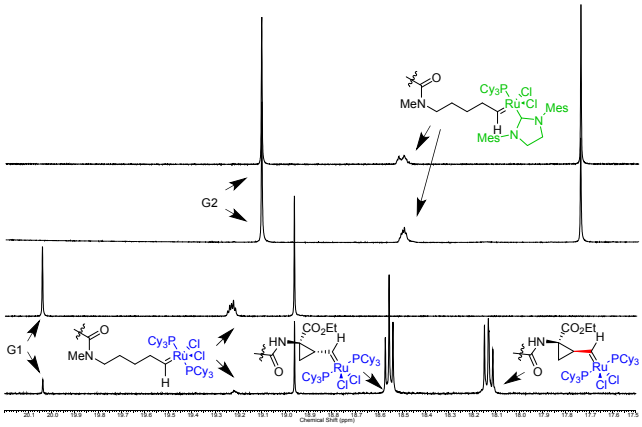


Figure 3. NMR observation of the species of G1/G2 insertion at the diene double bonds of **7** and **8**. From *bottom* to *top*: **7** + G1, **8** + G1, **7** + G2, **8** + G2 respectively (only substructures represented, see more details in the supporting material).

We took advantage of this feature by applying the SHD conditions. Thus, by adding a concentrated solution of **8** to the bulk of the refluxing toluene containing the catalyst M2 over 2 hours (end concentration 0.05 M) we obtained **9** in 80 % in situ yield (compared to 70 % under batch conditions). Importantly, only dimeric species, and no higher polymers[25] were formed as byproducts in this reaction and thus a simple crystallization proved adequate to remove all oligomers and obtain the desired macrocycle in high purity, after the cleavage of the Boc group.

Although we added two steps to the synthesis (Boc introduction and removal), this was justified by the dramatically improved overall yield and the elimination of the chromatography. In terms of the industrially relevant space-time-yield parameter (STY) this represents a 15-fold increase in productivity over the original procedure (Table 2), rendering the new method viable for use in commercial scale manufacturing.

Table 2. Comparison of RCM procedures described

	STY, kg m^{-3} h^{-1}	Isolation after RCM
7 to **10** (batch)	0.86	chromatography
8 to **9** (SHD)	14	crystallization

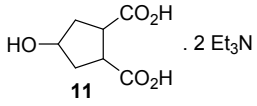
In summary, we have studied the macrocyclization by RCM, a key step in the simeprevir synthesis. We have shown that substrate modification (substitution on the amide *N*) leads to a change in the insertion site of the catalyst and is also changing the kinetic model of the RCM. The cyclization of the dienes in this study is slow, and we could characterize and report here the NMR spectra of the inserted species with a 2nd generation RCM catalyst for the first time in the literature. By combined application of substrate modification, together with the simulated high dilution technique, a significant improvement of the cyclization efficiency has been achieved.

The scope of the *N*-substituent in the RCM of amide-containing dienes has been little explored, and we are currently obtaining data suggesting the Boc effect can be further optimized. These results will be reported in a full paper.

EXPERIMENTAL SECTION

General – Analytical methods and equipment. Melting points were measured with an automated Büchi B-545 instrument. Optical rotations were measured with a Perkin-Elmer 341 polarimeter at the sodium D-line emission wavelength. Reverse-phase UPLC analyses were performed on Waters Acquity system with BDH C18 columns, using aqueous 10 mM ammonium acetate buffer – acetonitrile gradient. Enantiomeric purity of compound **3** was measured by capillary electrophoresis on a Beckman Coulter P/ACE MDQ instrument, using borax buffer with β-cyclodextrin as chiral selector. High resolution mass spectrometric analysis was performed on a Waters Q TOF Ultima, Bruker Maxis instrument using high performance liquid chromatography electrospray ionization mass spectrometry (HPLC ESI MS). The molecular weight of the drug substance, was established by HPLC ESI MS and was further supported by exact mass measurements. NMR experiments were recorded at 300 K on a Bruker Avance 600 MHz spectrometer equipped with a 5 mm CPTXI z-gradient high-resolution cryoprobe running Topspin 2.1 software. 1D ^{1}H, $^{13}C\{^{1}H\}$ and 2D COSY, NOESY/ROESY, HSQC and HMBC spectra were collected using standard Bruker pulse programs. Chemical shifts are reported in parts per million (δ, ppm) downfield from tetramethylsilane (TMS), using for ^{1}H the residual solvent resonance of the deuterated DMSO or chloroform as internal reference at respectively 2.50 ppm and 7.27 ppm and using for ^{13}C the centerline of the carbon resonance due to the deuterated DMSO or chloroform as internal reference at respectively 39.51 ppm and 77.00 ppm. Proton-Proton scalar couplings are reported in Hertz (Hz). Multiplicities are reported by the following abbreviations: s = singlet; br. s. = broad singlet; d = doublet; dd = doublet of doublet; ddd = doublet of doublet of doublet; dt = doublet of triplet; dquin = doublet of quintet; q = quadruplet; quin = quintet; spt = septet; t = triplet; td = triplet of doublet; tt = triplet of triplet; m = multiplet.

(9*R*)-9-hydroxycinchonan-1-ium 3-oxo-2-oxabicyclo[2.2.1]heptane-5-carboxylate 3. 1.67 kg (9.68 mol) of **2**[5] was placed in the 16 L autoclave followed by 9 L of de-ionized water, 2.68 L (19.36 mol) of triethylamine and 335 g (20 w/w%) of Raney-Ni catalyst. The autoclave was closed, purged three times with nitrogen and three times with hydrogen and the pressure was adjusted to 80 bar. Stirring and heating were started. As soon as the temperature reached 120°C, the pressure was re-adjusted to 80 bar and the reaction was stirred for 24 h. The autoclave was cooled to 60°C and full conversion was confirmed after sampling, the autoclave was cooled to room temperature, vented and unloaded. The crude solution of **11** was filtered over filtration aid, the filtrate was collected, and the filter was washed twice with 4 L of water. This step was repeated and further scaled up at external suppliers to ensure the manufacturing of the required quantities.

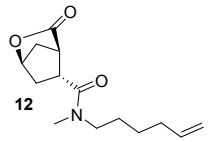


11

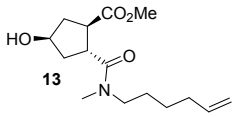
500 kg (689 mol) of a 24 w/w% aqueous solution of salt **11** was concentrated under vacuum keeping the temperature below 80°C. The oily residue was azeotropically dried by addition of 17 kg (168 mol) of triethylamine and 364 L of MeTHF and concentration under atmospheric pressure. This cycle was repeated once before the residue was cooled to 40°C and redissolved in 700 L of acetone; the level of residual water being between 0.3 and 1 w/w%. The solution was further diluted with 1360 L of acetone and 90 kg (889 mol) of triethylamine and cooled to around -5°C. 84 kg (774 mol) of ethyl chloroformate was added keeping the temperature below 5°C. After complete addition, the reaction mixture was stirred further for 3 hours at around 0°C before the triethylammonium chloride byproduct was discarded by centrifugation. The resulting filtrate (solution of crude lactone in acetone – MeTHF) was then added to a solution of 108 kg (367 mol) of cinchonidine in ethanol – isopropanol (2 x 665 L) kept at 60°C. After 1 hour at 60°C, the reaction mixture was cooled to 22°C over 4 hours then refluxed for 3 hours and again cooled to 22°C over 4 hours. After 20 hours at 22°C, the suspension was filtered and the cake (crude salt **3**) was washed with 63 L of ethanol then recrystallized from ethanol – water (452 l + 18 L, reflux to 22°C), centrifugated, washed with 40 L of ethanol and dried under vacuum. 59 kg (19% overall yield from **2**) of salt **3** is usually obtained as a white solid from the process. Enantiomeric ratio of the acid in the salt (capillary electrophoresis): 98 : 2. mp: 222.0-222.6°C. $[\alpha]^{D}$: -31.1 (1%, DMF). HRMS (ESI-TOF) *m/z*: $[M - H]^{-}$ Calcd for $C_7H_7O_4$ 155.0344; Found 155.0356. ^{1}H NMR (600 MHz, DMSO-d_6) δ ppm 8.85 (d, *J*=4.2 Hz, 1 H), 8.31 (d, *J*=7.9 Hz, 1 H), 8.03 (dd, *J*=8.5, 0.9 Hz, 1 H), 7.74 (ddd, *J*=8.4, 6.9, 1.3 Hz, 1 H), 7.61 (ddd, *J*=8.4, 6.9, 1.3 Hz, 1 H), 7.58 (d, *J*=4.2 Hz, 1 H), 5.85 (ddd, *J*=17.4, 10.2, 7.6 Hz, 2 H), 5.44 (d, *J*=6.0 Hz, 1 H), 5.01 (dt, *J*=15.5, 1.5 Hz, 1 H), 4.98 - 5.01 (m, 1 H), 4.94 (dt, *J*=10.5, 1.4 Hz, 1 H), 3.33 (t, *J*=14.0 Hz, 9 H), 3.18 (q, *J*=7.6 Hz, 1 H), 3.00 (s, 1 H), 2.97 (dd, *J*=13.4, 10.4 Hz, 1 H), 2.73 (dd, *J*=8.9, 4.3 Hz, 1 H), 2.56 (td, *J*=12.0, 4.9 Hz, 2 H), 2.30 (t, *J*=12.0 Hz, 1 H), 2.13 (ddd, *J*=13.6, 4.5, 1.9 Hz, 1 H), 2.09 (dquin, *J*=11.0,

1.9, 1.9, 1.9, 1.9 Hz, 1 H), 2.02 (ddd, *J*=13.6, 9.1, 1.9 Hz, 1 H), 1.76 - 1.81 (m, 2 H), 1.63 - 1.76 (m, 3 H), 1.51 (t, *J*=11.0 Hz, 1 H). $^{13}C\{^{1}H\}$ NMR (150 MHz, DMSO-d_6) δ ppm 176.9, 174.2, 150.1, 149.8, 147.9, 141.8, 129.7, 128.9, 126.3, 125.8, 124.0, 119.1, 114.6, 80.4, 70.0 (br. s., 1 C), 60.5, 55.3, 45.5, 41.9, 39.9, 38.9, 37.7, 33.1, 27.3, 26.7, 23.0 (br. s.).

Methyl (1*R*,2*R*,4*S*)-2-(hex-5-en-1-yl(methyl)carbamoyl)-4-((2-(4-isopropylthiazol-2-yl)-7-methoxy-8-methylquinolin-4-yl)oxy)cyclopentane-1-carboxylate 5. A mixture of 12.38 kg (50 mol) of N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ), 20.5 kg (45.5 moles) of **3** and 5.41 L (55 mol) of N-methyl-5-hexen-1-amine[6] in 228 L THF was stirred and heated to a gentle reflux (caution: foam formation due to CO_2 release). Upon full conversion, typically after 72 h, the mixture was cooled to room temperature and the precipitate was filtered off and washed with 91 L of toluene. The combined mother liquor and wash solvent were transferred into a reactor and stirred at room temperature before subsequent addition of 45 L of water and 10.83 L of HCl (aq. 35 wt%) over 30 minutes while keeping the temperature below 25°C. An extra portion of 11.38 L of water was added and the pH was checked to be around pH 1-2. After phase separation the organic layer was washed with 28.21 L of water and dried with 20 kg of anhydrous magnesium sulfate. After filtration, the solution of the lactone amide **12** was concentrated under reduced pressure at 75°C until an oil was obtained. The mixture was cooled to 60°C and 91 L of methanol and 0.19 L (0.06 equiv) of methanesulfonic acid were added. The pH was checked and adjusted to 2 by adding extra methanesulfonic acid if necessary. The reaction mixture was heated and stirred at reflux for 20 hours then cooled to room temperature. 250 g sodium hydrogen carbonate was added, and the pH was adjusted to pH ≥7 with sodium hydrogen carbonate, if necessary, before filtering off the sodium salts. To the mother liquor, 318 L of toluene was added and the mixture was heated with stirring until the vapor temperature reached 110°C to distill off methanol. After cooling to room temperature, the concentration of desired methyl ester **13** was assayed to calculate the required amounts of the reagents for the next step (typical yield: 82 % from **3**).



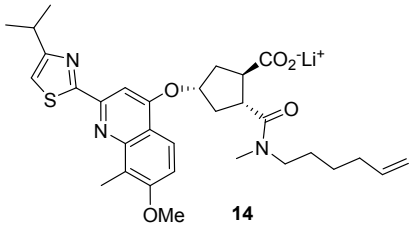
12



13

To a reactor charged with 10.58 kg (37.3 mol) **13** in toluene solution were added 9.97 kg (38.0 mol) of triphenylphosphine and 11.82 kg (37.3 mol) of **4**[7] and upon stirring, the mixture was refluxed to remove water azeotropically before cooling to -2°C. 7.48 L (38.0 mol) of diisopropyl azadicarboxylate (DIAD) was dosed over approximately one hour while maintaining a maximum temperature of 0°C. After complete addition, the reaction mixture was stirred another hour at 0°C before addition of 0.15 L water and further stirring for another hour at 0°C to allow triphosphine oxide to precipitate out from the reaction mixture. Triphenylphosphine oxide and salts were then filtered off and the filter cake was washed with 20.65 L of toluene. The mother liquor containing **5** in toluene and wash solvent were brought together into a reactor and upon stirring, the mixture was concentrated under reduced pressure (max. temperature 110°C) before addition of 107 L of 1-butanol and further concentration under reduced pressure (max. temperature 110°C). The residue was then cooled to 60°C, 107 L of isopropanol was added and the solution was refluxed for at least 15 minutes before cooling to 47°C. The solution was seeded with 0.11 kg of **5** and stirred for two hours at 47°C before cooling to 0°C over eight hours. The slurry was stirred for eight hours at 0°C before the solid **5** was isolated via centrifugation or filtration. Washing was carried out with 12.4 L of cooled isopropanol. The wet solid was dried under vacuum for 16 hours at 70°C and delivered 15.36 kg (71% yield) of **5**. mp: 128.2-129.8°C. $[\alpha]^{D}$: -8.2 (1%, MeOH). HRMS (ESI-TOF) *m*/*z*: $[M + H]^+$ Calcd for $C_{32}H_{42}N_3O_5S$ 580.2845; Found 580.2842. NMR: two rotamers are observed at 300 K in DMSO-d_6 in a 55/45 ratio. ^1H NMR (400 MHz, DMSO-d_6) δ ppm 8.04 (d, *J*=9.1 Hz, 1 H), 8.03 (d, *J*=8.8 Hz, 1 H), 7.37 - 7.50 (m, 3 H), 5.81 (m, *J*=17.0, 10.3, 6.6, 6.6 Hz, 1 H), 5.68 (m, *J*=17.1, 10.3, 6.7, 6.5 Hz, 1 H), 5.33 (s, 1 H), 4.76 - 5.09 (m, 2 H), 3.96 (s, 3 H), 3.63 - 3.71 (m, 1 H), 3.62 (s, 3 H), 3.41 - 3.51 (m, 1 H), 3.37 (dd, *J*=13.8, 7.6 Hz, 2 H), 3.25 (td, *J*=13.6, 7.1 Hz, 2 H), 3.14 (ddd, *J*=13.7, 6.8, 6.7 Hz, 2 H), 2.98 (s, 3 H), 2.79 (s, 3 H), 2.70 - 2.78 (m, 1 H), 2.58 (s, 3 H), 2.34 (d, *J*=1.3 Hz, 1 H), 2.18 - 2.31 (m, 1 H), 2.07 (q, *J*=6.9 Hz, 1 H), 1.92 (q, *J*=6.8 Hz, 1 H), 1.80 (t, *J*=12.1 Hz, 1 H), 1.53 (ddd, *J*=19.0, 7.3, 7.2 Hz, 1 H), 1.35 - 1.43 (m, 2 H), 1.34 (d, *J*=6.8 Hz, 6 H), 1.19 (quin, *J*=7.5 Hz, 1 H). $^{13}C\{^{1}H\}$ NMR (100 MHz, DMSO-d_6) δ ppm 174.1, 174.0, 171.9, 168.7, 168.7, 164.2, 160.6, 160.6, 158.0, 158.0, 151.2, 151.2, 147.9, 147.8, 138.5, 138.4, 120.6, 120.5 (br. s.), 120.0, 120.0, 116.2, 116.1, 115.5, 114.9, 114.6, 112.9, 112.8, 95.4 (br. s.), 95.4 (br. s.), 78.3, 78.2, 56.1, 51.7, 51.6, 48.6, 46.7, 45.2, 44.9, 42.6, 42.2, 36.8, 36.2, 35.7 (br. s.), 35.7 (br. s.), 34.6, 33.1, 32.9, 32.8, 30.4, 27.6, 25.9, 25.2, 25.1, 22.3, 9.8.

Ethyl (1*R*,2*S*)-2-ethenyl-1-({[(1*R*,2*R*,4*R*)-2-[hex-5-en-1-yl(methyl)carbamoyl]-4-({7-methoxy-8-methyl-2-[4-(propan-2-yl)-1,3-thiazol-2-yl]quinolin-4-yl}oxy)cyclopentyl]carbonyl}amino)cyclopropanecarboxylate 7. 1 L of water and 46.2 g (1.1 mol) of lithium hydroxide monohydrate were poured into a reactor and stirred for 15 minutes before addition of 580 g (1 mol) of **5** and 4 L of THF. The reaction mixture was stirred at room temperature for 24 hours to ensure complete hydrolysis of the ester into the salt **14**.



14

To the obtained solution of salt **14** in THF/water were added 225 g (1.1 mol) of **6**[8] and 284 g (1.15 mol) of EEDQ. The reaction mixture was stirred at room temperature for 24 hours before 13.8 g (0.1 mol) of sodium dihydrogenophosphate monohydrate was added and stirring was prolonged under the same conditions for 24 hours. 2 L of toluene, 1 L of water, and 1.6 mole of HCl (aq.) were added to the reactor and after stirring for at least 15 min at room temperature, the layers were allowed to settle. After separation of the layers, the aqueous layer was discarded and the organic one was washed with water. The THF and toluene from the organic layer were evaporated under vacuum and replaced with 1.2 L of dichloromethane (DCM). The resulting solution of **7** in DCM was stored at room temperature prior further use. Assay-yield (quantitative HPLC):

96% from **5**. A sample of **7** was isolated as an amorphous foamy solid for characterization and reference purpose. This transformation was further scaled up to around 35 kg (50 mol) for plant production. mp: 64.4-67.2°C. $[\alpha]^D$: -26.4 (1%, dichloromethane). HRMS (ESI-TOF) *m/z*: $[M + H]^+$ Calcd for $C_{39}H_{51}N_4O_6S$ 703.3524; Found 703.3517. NMR: Two major rotamers were observed at 300 K in chloroform-*d* in a 60/40 ratio. ^{1}H NMR (600 MHz, chloroform-*d*) δ ppm 8.03 (d, *J*=9.1 Hz, 1 H), 8.01 (d, *J*=9.1 Hz, 1 H), 7.48 (s, 1 H), 7.47 (s, 1 H), 7.22 (d, *J*=9.1 Hz, 1 H), 7.01 (s, 1 H), 6.99 (s, 1 H), 6.89 (s, 1 H), 5.65 - 5.82 (m, 2 H), 5.30 - 5.34 (m, 1 H), 5.27 (d, *J*=17.4 Hz, 1 H), 5.11 (d, *J*=10.6 Hz, 1 H), 5.02 (dd, *J*=17.0, 1.5 Hz, 1 H), 4.98 (d, *J*=10.2 Hz, 1 H), 4.95 (dd, *J*=17.4, 1.5 Hz, 1 H), 4.89 (d, *J*=10.2 Hz, 1 H), 4.04 - 4.17 (m, 2 H), 3.97 (s, 3 H), 3.72 (ddd, *J*=12.1, 10.2, 7.2 Hz, 1 H), 3.66 (ddd, *J*=11.5, 10.4, 7.2 Hz, 1 H), 3.46 (ddd, *J*=13.6, 8.3, 6.4 Hz, 1 H), 3.28 - 3.35 (m, 2 H), 3.22 (spt, *J*=10.0 Hz, 1 H), 3.19 (tt, *J*=7.2, 3.4 Hz, 1 H), 3.02 (s, 3 H), 2.95 (s, 3 H), 2.76 - 2.86 (m, 1 H), 2.69 (s, 3 H), 2.47 - 2.56 (m, 1 H), 2.34 (dd, *J*=14.2, 7.0 Hz, 1 H), 2.05 - 2.13 (m, 2 H), 1.97 - 2.04 (m, 2 H), 1.83 - 1.88 (m, 3 H), 1.55 - 1.68 (m, 2 H), 1.47 - 1.54 (m, 3 H), 1.45 (ddd, *J*=9.3, 5.7, 3.2 Hz, 3 H), 1.38 (d, *J*=6.8 Hz, 6 H), 1.29 - 1.36 (m, 2 H), 1.20 (t, *J*=7.2 Hz, 3 H). ^{13}C{^{1}H} NMR (150 MHz, chloroform-*d*) δ ppm 174.5, 174.3, 173.1, 172.9, 169.93, 169.90, 169.87, 164.9, 160.8, 160.7, 158.1, 151.77, 151.75, 148.7, 138.3, 137.9, 133.7, 121.8, 120.4, 117.74, 117.67, 116.88, 116.85, 115.2, 114.7, 114.13, 114.11 (br. s.), 112.3, 95.8, 77.69 (br. s.), 77.66, 61.12, 61.11, 56.2, 49.7, 48.2, 46.1 (d, *J*=12.1 Hz), 45.6, 45.4, 40.03, 40.00, 37.8, 37.2, 35.23, 35.20, 34.7, 33.7, 33.6, 33.4, 33.32, 33.30, 31.1, 27.9, 26.4, 25.94, 25.89, 23.2, 23.0, 22.51, 22.48, 22.44, 22.42, 14.3 (br. s.), 14.2, 9.8.

Ethyl (2*R*,3a*R*,10*Z*,11a*S*,12a*R*,14a*R*)-2-({7-methoxy-8-methyl-2-[4-(propan-2-yl)-1,3-thiazol-2-yl]quinolin-4-yl}oxy)-5-methyl-4,14-dioxo-2,3,3a,4,5,6,7,8,9,11a,12,13,14,14a-tetradecahydrocyclopenta[c]cyclopropa[g][1,6]diazacyclotetradecine-12a(1*H*)-carboxylate 10 from 7. A well stirred solution of 35.15 kg (50 mol) of diene **7** and 480 g (5 mol) of methanesulfonic acid in 1,2-dichloroethane (5500 L) was refluxed under nitrogen before addition of 750 g (1.25 mol) of GH1. After 6 hours of reflux, the conversion was checked and reached >95%. The reaction mixture was cooled to room temperature and the catalyst was deactivated by addition of 1164 g (7.7 mol) of 2-mercaptonicotinic acid and 627 ml (15 mol) of triethylamine to neutralize the methanesulfonic acid. After a washing with 250 L water, the organic layer was collected and the in-situ yield was determined at 74 %. Solvent was switched to 1-butanol, from which 10 crystallized out after seeding (20.6 kg obtained). Isolated yield: 60.9%. mp: 219.8-221.2°C. $[\alpha]^D$: +21.8 (1%, CH_2Cl_2). HRMS (ESI-TOF) *m/z*: $[M + H]^+$ Calcd for $C_{37}H_{47}N_4O_6S$ 675.3216; Found 675.3210. ^{1}H NMR (600 MHz, chloroform-*d*) δ ppm 8.02 (d, *J*=9.4 Hz, 1 H), 7.48 (s, 1 H), 7.21 (d, *J*=9.1 Hz, 1 H), 7.18 (s, 1 H), 7.00 (s, 1 H), 5.57 - 5.69 (m, 1 H), 5.31 (dd, *J*=10.6, 6.0 Hz, 1 H), 5.20 (dd, *J*=10.6, 6.0 Hz, 1 H), 4.65 (td, *J*=12.8, 4.5 Hz, 1 H), 4.12 (ddd, *J*=14.4, 11.0, 7.2 Hz, 1 H), 4.06 (ddd, *J*=14.3, 10.7, 7.2 Hz, 1 H), 3.97 (s, 3 H), 3.84 (td, *J*=11.4, 7.4 Hz, 1 H), 3.36 (q, *J*=10.2 Hz, 1 H), 3.19 (spt, *J*=6.8 Hz, 1 H), 3.05 (s, 3 H), 2.84 (ddd, *J*=14.7, 9.1, 6.4 Hz, 1 H), 2.69 (s, 3 H), 2.63 (td, *J*=12.8, 6.8 Hz, 2 H), 2.38 (ddd, *J*=23.0, 12.3, 6.2 Hz, 1 H), 2.29 (dd, *J*=14.5, 7.4 Hz, 1 H), 2.05 (ddd, *J*=14.0, 11.0, 3.4 Hz, 1 H), 2.00 (q, *J*=7.9 Hz, 1 H), 1.94 (dd, *J*=7.9, 5.3 Hz, 1 H), 1.80 - 1.88 (m, 1 H), 1.75 (t, *J*=12.5 Hz, 1 H), 1.53 - 1.64 (m, 1 H), 1.38 (d, *J*=7.2 Hz, 7 H), 1.24 - 1.33 (m, 1 H), 1.16 - 1.22 (m, 4 H). ^{13}C{^{1}H} NMR (150 MHz, chloroform-*d*) δ ppm 174.8, 174.0, 169.9, 169.2, 164.9, 160.8, 158.1, 151.8, 148.6, 135.6, 125.0, 121.8, 120.3, 116.9, 114.1, 112.3, 95.8, 77.4, 60.8, 56.2, 47.3, 45.7, 44.6, 41.9, 36.5, 33.7, 32.4, 31.1, 28.3, 26.2, 24.9, 24.6, 22.5, 22.4, 22.3, 14.4, 9.8.

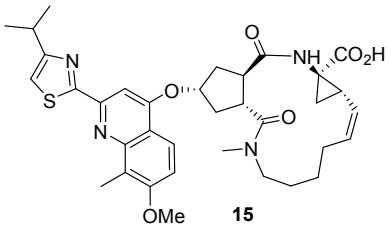
Ethyl (1*R*,2*S*)-1-[(*tert*-butoxycarbonyl){[(1*R*,2*R*,4*S*)-2-[hex-5-en-1-yl(methyl) carbamoyl]-4-({7-methoxy-8-methyl-2-[4-(propan-2-yl)-1,3-thiazol-2-yl]quinolin-4-yl}oxy)cyclopentyl]carbonyl}amino]-2-ethenylcyclopropanecarboxylate 8. A solution of 2.6 kg (3.7 mol) of **7** in 30 L of DCM was subject of a solvent switch to a volume of 18.5 L toluene. To the resulting solution, 23 g of 4-dimethylaminopyridine was added at room temperature before a first portion of 970 g (19.7 mmol) of di-*tert*-butyl dicarbonate was added and the reaction mixture was heated to 55°C under an inert atmosphere. The reaction was stirred overnight before HPLC analysis. In case of incomplete conversion an extra portion of di-*tert*-butyl dicarbonate (650 g) was added and the reaction mixture was stirred for another 4 hours before checking the conversion. Upon complete conversion of the starting material, the mixture was cooled to room temperature and washed with 5.6 L of a 0.35 wt% HCl solution. After removal of the water layer, the organic layer was dried using 370 g magnesium sulfate. The solution was concentrated to a final total weight of 8.04 kg and used as such in the next step. A sample of **8** was isolated and purified for characterization and reference material purpose. Assayed yield: 2.8 kg 8 (95 %). mp: 35.0-38.3°C. $[\alpha]^D$: +15.6 (1%, CH_2Cl_2). HRMS (ESI-TOF) *m/z*: $[M + H]^+$ Calcd for $C_{44}H_{59}N_4O_8S$ 803.4054; Found 803.4052. NMR: four rotamers were observed at 300 K in chloroform-*d* in an approximately 40/30/20/15 ratio. ^{1}H NMR (600 MHz, chloroform-*d*) δ ppm 8.10 (d, *J*=9.1 Hz, 1 H), 8.09 (d, *J*=9.1 Hz, 1 H), 7.46 (s, 1 H), 7.23 (d, *J*=9.4 Hz, 1 H), 7.22 (d, *J*=9.1 Hz, 1 H), 7.01 (s, 1 H), 5.85 - 5.97 (m, 1 H), 5.80 (ddt, *J*=17.0, 10.2, 6.8, 6.8 Hz, 1 H), 5.74 (ddt, *J*=17.0, 10.2, 6.8, 6.8 Hz, 1 H), 5.22 - 5.33 (m, 2 H), 5.17 (t, *J*=11.0 Hz, 1 H), 5.03 (d, *J*=17.0 Hz, 1 H), 4.98 (d, *J*=9.8 Hz, 1 H), 4.97 (dd, *J*=17.2, 1.7 Hz, 1 H), 4.91 (d, *J*=10.2 Hz, 1 H), 4.63 (q, *J*=9.4 Hz, 1 H), 4.04 - 4.21 (m, 2 H), 3.98 (s, 3 H), 3.68 (quin, *J*=9.8 Hz, 1 H), 3.62 (quin, *J*=8.8 Hz, 1 H), 3.25 - 3.56 (m, 2 H), 3.18 (spt, *J*=6.8 Hz, 1 H), 3.08 (br. s., 3 H), 3.07 (br. s., 3 H), 2.92 (s, 3 H), 2.71 - 2.83 (m, 1 H), 2.69 (s, 3 H), 2.67 (br. s., 1 H), 2.59 (d, *J*=7.9 Hz, 1 H), 2.21 - 2.30 (m, 1 H), 2.16 (d, *J*=7.6 Hz, 2 H), 2.07 - 2.14 (m, 1 H), 2.03 (d, *J*=6.4 Hz, 1 H), 1.86 - 1.97 (m, 1 H), 1.65 - 1.72 (m, 1 H), 1.56 - 1.62 (m, 1 H), 1.46 - 1.51 (m, 1 H), 1.45 (br. s., 4 H), 1.42 (br. s., 1 H), 1.40 (s, 5 H), 1.38 (d, *J*=6.8 Hz, 6 H), 1.30 - 1.36 (m, 1 H), 1.15 - 1.24 (m, 3 H). ^{13}C{^{1}H} NMR (150 MHz, chloroform-*d*) δ ppm 177.7 (br. s.), 177.5 (br. s.), 177.44 (br. s.), 177.38 (br. s.), 172.9 (br. s.), 172.8 (br. s.), 172.7 (br. s.), 172.4 (br. s.), 170.0 (br. s.), 169.94 (br. s.), 169.90 (br. s.), 169.5 (br. s.), 169.4 (br. s.), 169.3 (br. s.), 164.8, 161.2 (br. s.), 161.12 (br. s.), 161.08 (br. s.), 158.0, 152.3 (br. s.), 152.2 (br. s.), 151.6 (br. s.), 148.6, 138.5 (br. s.), 138.4 (br. s.), 138.04 (br. s.), 138.00 (br. s.), 133.6, 133.1 (br. s.), 121.5 (br. s.), 120.7 (br. s.), 120.6 (br. s.), 118.0 (br. s.), 117.8 (br. s.), 116.8, 115.0 (br. s.), 114.9 (br. s.), 114.6 (br. s.), 114.5 (br. s.), 114.0, 112.1, 95.6 (br. s.), 83.5, 83.42, 77.84 (br. s.), 77.79 (br. s.), 77.7, 61.24, 61.18 (br. s.), 56.1, 49.8 (br. s.), 48.6 (br. s.), 48.0 (br. s.), 48.0 (br. s.), 47.90, 47.85, 47.8 (br. s.), 44.4 (br. s.), 44.11, 44.06, 43.4 (br. s.), 42.2 (br. s.), 41.7 (br. s.), 37.3 (br. s.), 37.2 (br. s.),

37.1 (br. s.), 37.04 (br. s.), 36.99 (br. s.), 36.9, 36.8, 36.7 (br. s.), 36.5 (br. s.), 36.2, 35.2, 33.9 (br. s.), 33.8 (br. s.), 33.32 (br. s.), 33.27 (br. s.), 31.0, 28.1, 28.0 (br. s.), 27.8 (br. s.), 27.7 (br. s.), 26.5 (br. s.), 25.9 (br. s.), 25.8, 25.13 (br. s.), 25.06 (br. s.), 25.0 (br. s.), 24.9 (br. s.), 22.4 (br. s.), 14.2 (br. s.), 9.8.

13-*tert*-butyl 12a-ethyl (2*S*,3a*R*,10*Z*,11a*S*,12a*R*,14a*R*)-2-({7-methoxy-8-methyl-2-[4-(propan-2-yl)-1,3-thiazol-2-yl]quinolin-4-yl}oxy)-5-methyl-4,14-dioxo-1,2,3,3a,4,5,6,7,8,9,11a,12,14,14a-tetradecahydrocyclopenta[c]cyclopropa[g][1,6]diazacyclotetradecine-12a,13-dicarboxylate 9. 81.4 L of toluene was poured in a reactor, washed with 7.5 L of a 0.3 wt% aqueous HCl solution and subsequently azeotropically dried. This solvent is referred to as washed toluene. A solution of 50 g Neolyst M2 (1.5 mol%) in 4.63 L washed toluene was prepared. 64 L of washed toluene was introduced in a reactor and refluxed. After 30 min of reflux, the solution of M2 catalyst in toluene was dosed over a 3.5 h period. 15 min after starting this dosed addition, the concentrated solution of **8** was dosed in parallel over a period of 3 h. After complete addition of the two solutions, complete conversion (>99%) was reached (LC analysis). The reaction mixture was then cooled to room temperature and used as such in the next step. A sample of **9** was isolated and purified for characterization and reference material purpose. Typical yield (assay by HPLC): 80%. mp: 132.3-134.1°C. $[\alpha]^D$: +9.8 (1%, DMF). HRMS (ESI-TOF) *m/z*: $[M + H]^+$ Calcd for $C_{42}H_{55}N_4O_8S$ 775.3741; Found 775.3743. NMR: two major rotamers were observed at 300 K in chloroform-*d* in a 87/13 ratio. ^{1}H NMR (600 MHz, chloroform-*d*) δ ppm 8.06 (d, *J*=9.1 Hz, 1 H), 7.42 - 7.49 (m, 1 H), 7.22 (d, *J*=9.1 Hz, 1 H), 7.01 (s, 1 H), 5.52 - 5.71 (m, 2 H), 5.35 (q, *J*=6.2 Hz, 1 H), 4.60 (td, *J*=13.6, 3.0 Hz, 1 H), 4.37 (td, *J*=11.0, 7.6 Hz, 1 H), 4.16 - 4.27 (m, 1 H), 4.07 - 4.14 (m, 1 H), 4.07 (m, *J*=11.7, 11.7, 6.8 Hz, 1 H), 3.97 (s, 3 H), 3.88 (td, *J*=13.0, 3.8 Hz, 1 H), 3.78 (td, *J*=11.0, 9.1 Hz, 1 H), 3.49 (ddd, *J*=12.8, 11.3, 7.6 Hz, 1 H), 3.19 (quin, *J*=6.8 Hz, 1 H), 2.99 (s, 3 H), 2.95 (dd, *J*=14.4, 7.6 Hz, 1 H), 2.86 (s, 3 H), 2.86 (dt, *J*=13.8, 7.1 Hz, 8 H), 2.78 (m, *J*=13.6, 7.6, 6.0 Hz, 1 H), 2.74 (dd, *J*=14.0, 8.7 Hz, 1 H), 2.70 (s, 3 H), 2.56 (m, *J*=13.6, 3.0, 3.0 Hz, 1 H), 2.41 - 2.50 (m, 1 H), 2.38 (q, *J*=9.8 Hz, 1 H), 2.17 (ddd, *J*=13.6, 12.5, 7.6 Hz, 1 H), 1.97 (td, *J*=13.2, 4.9 Hz, 1 H), 1.81 - 1.88 (m, 2 H), 1.80 (dd, *J*=9.3, 5.5 Hz, 1 H), 1.73 (dd, *J*=10.2, 5.7 Hz, 1 H), 1.69 - 1.73 (m, 1 H), 1.43 (s, 9 H), 1.40 (br. s., 2 H), 1.38 (d, *J*=6.8 Hz, 6 H), 1.21 (t, *J*=7.2 Hz, 3 H) ^{13}C{^{1}H} NMR (150 MHz, chloroform-*d*) δ ppm 181.8, 179.2, 172.3, 172.2, 170.4, 170.0, 169.91, 169.86 (br. s.), 164.8, 161.04, 161.00 (br. s.), 158.0, 151.9, 151.72, 151.66, 148.6, 131.8, 131.7 (br. s.), 125.3, 124.7, 121.7, 121.6, 120.5, 120.4 (br. s.), 116.8, 116.7 (br. s.), 114.1, 112.1, 95.8, 83.3, 82.7, 77.8, 77.5, 61.1, 61.0, 60.3, 56.1, 50.2, 49.3, 48.0, 47.0, 45.0, 44.6, 43.6, 35.9, 34.7, 33.0, 32.1, 32.0, 31.8, 31.0, 27.9, 27.8, 27.7, 27.2, 27.0, 26.2, 25.9, 25.5, 25.1, 22.42, 22.35, 14.2, 14.1, 9.8.

10 from 9. 3.7 L ethanol and 450 ml concentrated sulfuric acid were added to the solution of **9** from the previous step before heating to 80°C for 6 hours. After complete conversion (>98%, confirmed by LC) was reached, the mixture was cooled to room temperature before adding a solution of 470 g sodium bicarbonate in 7.4 L water. After vigorously stirring and settling of the layers, the water layer was removed and the organic one was further washed with 3.7 L of fresh water and azeotropically dried. 1.7 kg of **10** was then isolated by crystallization after a solvent switch to 1-butanol. Isolated yield: 89% (68% overall yield from **7**).

(2*R*,3a*R*,10*Z*,11a*S*,12a*R*,14a*R*)-2-({7-methoxy-8-methyl-2-[4-(propan-2-yl)-1,3-thiazol-2-yl]quinolin-4-yl}oxy)-5-methyl-4,14-dioxo-2,3,3a,4,5,6,7,8,9,11a,12,13,14,14a-tetradecahydrocyclopenta[c]cyclopropa[g][1,6]diazacyclotetradecine-12a(1*H*)-carboxylic acid 15. A reactor was charged with 5 L ethanol and 0.5 L water and stirred before 675 g (1 mol) of **10** and 96 g (1.2 mol) of 50 w% NaOH solution were added. The mixture was refluxed for 10 hours before cooling to 60°C and addition of 11.4 ml (0.2 mol) acetic acid and 0.2 L water. Subsequently, 3.5 L water was added, maintaining the temperature at 60°C. The ethanol was evaporated under vacuum from the reaction mixture, 3 L of water was added to the reactor and the temperature was adjusted to 25°C. 7 L of dichloromethane, 68.6 ml (1.2 mol) of acetic acid and 0.2 L of water were then successively added and the reaction mixture was stirred for 2 hours. The layers were then allowed to settle and the organic layer was separated. The aqueous layer was extracted with 1 L of dichloromethane before it was discarded. The organic layers were combined and transferred in a reactor containing 0.16 L methanol. 65 g silica gel was added before stirring the mixture for at least one hour at room temperature. After filtration of the silica gel, the filtrate was concentrated under atmospheric pressure and before addition of 1 L 2-butanone and 0.045 L water. This mixture was heated to 40°C before it was seeded with a small amount of the desired carboxylic acid and stirred for at least two hours at 40°C. Part of the solvent was evaporated under atmospheric pressure until a temperature of 75°C was reached. The removed solvent was then replaced with an equal volume of 2-butanone and 0.055 L water and the reflux was maintained for at least 30 minutes before the mixture was gradually cooled to 0°C and kept at this temperature for at least another 4 hours. 582 g (90% yield) of **15** was obtained after filtration, wash with 2-butanone/water and drying under vacuum. This transformation was scaled up to multi-kg scale in the manufacturing plant. mp: 239.0-239.7°C. $[\alpha]^D$: +56.9 (1%, DMF). HRMS (ESI-TOF) *m/z*: $[M + H]^+$ Calcd for $C_{35}H_{43}N_4O_6S$ 647.2903; Found 647.2906. NMR: two rotamers were observed at 300 K in DMSO-d_6 in a 92/8 ratio; multiplets of minor rotamer not reported. ^{1}H NMR (600 MHz, DMSO-d_6) δ ppm 11.30 - 13.38 (m, 1 H), 8.02 (d, *J*=9.4 Hz, 1 H), 7.89 (s, 1 H), 7.44 (d, *J*=0.8 Hz, 2 H), 7.45 (d, *J*=9.1 Hz, 2 H), 7.44 (s, 2 H), 5.57 (td, *J*=9.8, 6.4 Hz, 1 H), 5.32 - 5.36 (m, 1 H), 5.32 (dd, *J*=10.6, 9.4 Hz, 1 H), 4.28 (ddd, *J*=13.6, 9.8, 4.5 Hz, 1 H), 3.96 (s, 2 H), 3.34 (d, *J*=5.7 Hz, 7 H), 3.27 - 3.33 (m, 5 H), 3.14 (spt, *J*=7.0 Hz, 1 H), 2.99 (s, 3 H), 2.82 (dt, *J*=13.8, 7.1 Hz, 1 H), 2.57 - 2.59 (m, 3 H), 2.55 (t, *J*=5.3 Hz, 1 H), 2.30 (m, *J*=6.0 Hz, 2 H), 2.24 (sxt, *J*=6.4 Hz, 3 H), 2.14 (dd, *J*=13.4, 6.6 Hz, 2 H), 2.03 (q, *J*=9.1 Hz, 2 H), 1.74 (ddd, *J*=13.5, 10.5, 2.8 Hz, 3 H), 1.70 (m, *J*=4.2 Hz, 3 H), 1.65 (m, *J*=5.3 Hz, 3 H), 1.53 (m, *J*=6.0 Hz, 3 H), 1.48 (dd, *J*=8.3, 4.9 Hz, 1 H), 1.38 (dd, *J*=9.4, 4.9 Hz, 1 H), 1.33 (d, *J*=6.8 Hz, 6 H), 1.27 (m, *J*=5.7 Hz, 2 H), 1.18 - 1.24 (m, 2 H). ^{13}C{^{1}H} NMR (150 MHz, DMSO-d_6) δ ppm 173.8, 173.0, 171.1, 168.7, 164.4, 160.7, 158.1, 151.3, 147.9, 132.4, 126.6, 120.4, 120.2, 116.1, 115.6, 112.9, 95.5, 78.4, 56.2, 47.1, 45.6, 45.1, 40.7, 35.4, 34.6, 34.1, 30.5, 28.0, 27.5, 26.2, 24.5, 22.40 (br. s., 1 C), 22.37 (br. s., 1 C), 22.2, 9.9.

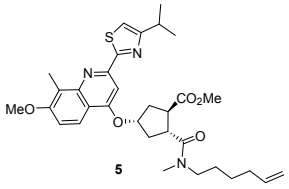


15

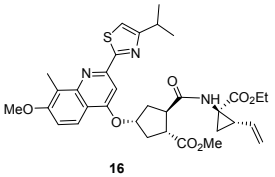
1 (Simeprevir). A reactor was charged with 5 L dichloromethane and 647 g (1 mol) **15** and the mixture obtained was stirred at room temperature. 230 g (1.2 mol) EDCI was added and stirring was continued for at least one hour before addition of 5 L water to the reactor. After 15 min stirring, the layers were allowed to settle then separated. The aqueous layer was discarded and the organic layer (solution of activated species in dichloromethane) was azeotropically dried during at least five hours using an inverse water separator, then cooled to room temperature. 151.5 g (1.25 mol) cyclopropanesulfonamide and 187 ml (1.25 mol) of DBU were added to the dried solution and stirring was maintained at room temperature for 24 hours. 2.5 L dichloromethane and a phosphate buffer solution (premixed solution of 12 mol phosphoric acid (aq.), 8 mol sodium hydroxide (aq.) and 6 L purified water) were added to the reactor. The biphasic mixture was stirred for at least 30 min before the layers were separated. The aqueous layer was discarded and the organic one was washed with 2.5 L water then dried by azeotropic distillation. 1.5 L isopropanol was added and the mixture was refluxed before cooling to 45°C and seeding. The suspension was stirred for another 12 hours at 45°C before addition of another 1 L of isopropanol and reflux again. Part of the solvent was distilled off under atmospheric pressure and the volume of the distillate was compensated with the same volume of isopropanol until a reflux temperature of minimum 81 °C was reached. The mixture was cooled gradually to room temperature over a period of 12 hours and the suspension was stirred for another 12 h at room temperature. 667 g (89% yield) of **1** was isolated after filtration, wash with isopropanol and drying. This transformation was scaled up to multi-kg scale in the manufacturing plant. mp: 238.0-239.1°C. $[\alpha]^D$: +30.8 (1%, CH_2Cl_2). HRMS (ESI-TOF) *m/z*: $[M + H]^+$ Calcd for $C_{38}H_{48}N_5O_7S_2$ 750.2995; Found 750.2977. NMR: two major rotamers were observed at 300 K in DMSO-d_6 in a 93/7 ratio; multiplets of minor rotamer are not reported. ^{1}H NMR (600 MHz, DMSO-d_6) δ ppm 11.44 (s, 1 H), 8.68 (s, 1 H), 8.03 (d, *J*=9.4 Hz, 1 H), 7.47 (d, *J*=9.4 Hz, 1 H), 7.45 (br. s, 2 H), 5.54 (td, *J*=10.1, 6.6 Hz, 1 H), 5.39 (td, *J*=6.8, 3.8 Hz, 1 H), 5.22 (*t*, J=10.5 Hz, 1 H), 4.39 (td, *J*=12.9, 2.5 Hz, 1 H), 3.96 (s, 3 H), 3.38 (td, *J*=11.5, 8.3 Hz, 1 H), 3.25 (ddd, *J*=12.8, 11.3, 6.8 Hz, 1 H), 3.14 (spt, *J*=6.8 Hz, 1 H), 2.99 (s, 3 H), 2.90 - 2.96 (m, 1 H), 2.87 (m, *J*=13.2, 7.6, 7.6 Hz, 1 H), 2.58 (s, 3 H), 2.55 (dt, *J*=13.3, 3.4 Hz, 1 H), 2.41 - 2.48 (m, 1 H), 2.29 (dd, *J*=13.6, 6.8 Hz, 1 H), 2.09 - 2.15 (m, 2 H), 1.77 - 1.83 (m, 1 H), 1.72 (td, *J*=12.7, 3.8 Hz, 1 H), 1.57 - 1.63 (m, 2 H), 1.46 (dd, *J*=8.5, 5.1 Hz, 1 H), 1.34 (d, *J*=6.8 Hz, 3 H), 1.33 (d, *J*=6.8 Hz, 3 H), 1.28 - 1.32 (m, 1 H), 1.14 - 1.25 (m, 2 H), 0.99 - 1.11 (m, 4 H). ^{13}C{^{1}H} NMR (150 MHz, DMSO-d_6) δ ppm 178.6, 172.1, 169.8, 168.6, 164.3, 160.7, 158.0, 151.3, 147.8, 131.4, 125.3, 120.3, 120.2, 116.0, 115.6, 112.8, 95.5, 78.5, 56.1, 47.6, 46.3, 43.1, 42.9, 35.1, 34.6, 33.4, 32.3, 30.6, 30.4, 26.9, 25.5, 23.7, 22.4, 22.3, 22.0, 9.9, 5.7, 5.6.

Procedures for NMR studies of RCM insertion species. To study the RCM reactions of **7** / **8** with G1 / G2 by NMR, 0.05 mmol of the diene was dissolved in 700 µl of deuterated dichloromethane and 0.025 mmol of catalyst was added. ^{1}H NMR spectra was recorded over time with the first spectrum typically recorded within 5 minutes after catalyst addition.

Correct identification of inserted species and thus insertion side of the catalyst was validated by performing the same reactions with so-called "half-substrates" **5** and **16**,[26] i.e. compounds that comprise only the hexenyl or vinylcyclopropane part of **7** and **8** (structures and respective spectra are shown in the supporting information). These compounds cannot undergo RCM. In case Ru-catalyzed dimerization is slow the inserted species tends to accumulate to a detectable level.



5



16

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website.
Copies of NMR spectra (pdf).

AUTHOR INFORMATION

Corresponding Authors

* E-mail: ahorvath@its.jnj.com
* E-mail: ddepre@its.jnj.com

Current Addresses

† Department of Organic Chemistry and Molecular Inorganic Chemistry, Stratingh Institute, University of Groningen, Nijenborgh 4, 9747 AG, Groningen, The Netherlands (E-mail: s.harutyunyan@rug.nl).
§ Syngenta Crop Protection Monthey SA, Case Postale 400, 1870 Monthey 1, Switzerland (E-mail: gregori.binot@syngenta.com).

Author Contributions

The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

REFERENCES

(1) Edlin, B.R.; Eckhardt, B.J.; Shu, M.A.; Holmberg, S.D.; Swan, T. Toward a More Accurate Estimate of the Prevalence of Hepatitis C in the United States *Hepatology* **2015**, *62(5)*, 1353-1363.

(2) Cummings, M. D.; Lindberg J; Lin, T-I.; de Kock, H.; Lenz, O.; Lilja, E.; Fellander, S.; Baraznenok, V.; Nystrom, S.; Nilsson, M.; Vrang, L.; Edlund, M.; Rosenquist, A.; Samuelsson, B.; Raboisson, P.; Simmen, S. Induced-fit Binding of the Macrocyclic Noncovalent Inhibitor TMC435 to its HCV NS3/NS4A Protease Target *Angew. Chem. Int. Ed*. **2010**, *49(9)*, 1652-1655.

(3) Rosenquist, A.; Samuelsson, B.; Johansson, P.-O.; Cummings, M.D.; Lenz, O.; Raboisson, P.; Simmen, K.; Vendeville, S.; de Kock, H.; Nilsson, M.; Horvath, A.; Kalmeijer, R.; de la Rosa, G.; Beumont-Mauviel, M. Discovery and Development of Simeprevir (TMC435), a HCV NS3/4A Protease Inhibitor *J. Med. Chem*. **2014**, *57(5)*, 1673-1693

(4) All the steps showed in the scheme were optimized using traditional chemical development tools and were efficient and easily scalable.

(5) Kurosawa, K; Obara, H. Further Study of Syntheses of dl-2,3-Dicarboxycyclopentaneacetic Acids *Bull. Chem. Soc. Jpn*. **1966**, *39(3)*, 525-530

(6) Synthesis of NMHA: Raboisson, P.; De Kock, H.; Dorange, I.; Vendeville, S.; Surleraux, D.; Tahri, A.; Simonnet, Y.; Van De Vreken, W.; Hu, L.; Simmen, K.; Nilsson, K.; Samuelsson, B.; Rosenquist, A.; Wallberg, H. WO 2007014921.

(7) Synthesis of **4**: Antonov, D.; Nilsson, K.; Raboisson, P.; Rosenquist, A.; Samuelsson, B.; Vendeville, S. WO 2008096002.

(8) Synthesis of **6**: Asuma, Y.; Suzuki, T.; Takehara, J.; Hidaka, T.; Asada, K.; Miyake, R.; Dekishima, Y.; Kawabata, H. US 9061991 B2.

(9) (a) Nicola, T.; Brenner, M.; Donsbach, K.; Kreye, P. First Scale-Up to Production Scale of a Ring Closing Metathesis Reaction Forming a 15-Membered Macrocycle as a Precursor of an Active Pharmaceutical Ingredient *Org. Proc. Res. Dev.* **2005**, *9(4)*, 513–515. See also references 11 and 20. (b) for general books and reviews on olefin metathesis, see *Handbook of Metathesis. Volume 2: Applications in Organic Synthesis*; Grubbs, R.H; O'Realy, D.J. Eds; Wiley-VCH: Weinheim, 2015; *Olefin Metathesis: Theory and Practice*; Grela, K. Ed; Wiley: Hoboken, NJ, 2014; Monfette, S.; Fogg, D.E. Equilibrium Ring-Closing Metathesis *Chem. Rev.* **2009**, *109*, 3783–3816.

(10) Concentrations higher than 0.1 M gave incomplete conversions using 2.5 mol% of catalyst, probably due to catalyst poisoning.

(11) Zeng, X.; Wei, X.; Farina, V.; Napolitano, E.; Xu, Y.; Zhang, L.; Haddad, N.; Yee, N. K.; Grinberg, N.; Shen, S.; Senanayake, C. H. Epimerization Reaction of a Substituted Vinylcyclopropane Catalyzed by Ruthenium Carbenes: Mechanistic Analysis *J. Org. Chem*. **2006**, *71(23)*, 8864-8875.

(12) Dienes **7** and **8** are glassy semisolids which we could not purify by crystallization.

(13) Sereewatthanawut, I.; Lim, F. W.; Bhole, Y. S.; Ormerod, D.; Horvath, A.; Boam, A. T.; Livingston, A. G. Demonstration of Molecular Purification in Polar Aprotic Solvents by Organic Solvent Nanofiltration *Org. Proc. Res. Dev*. **2010**, *14(3)*, 600-611.

(14) Also called pseudo-high dilution or Ziegler's infinite dilution technique: Ziegler, K. Ring-closure reactions *Ber. Dtsch. Chem. Ges*. **1934**, *67A*, 139-149. For a more recent review on macrocyclizations, including comments on SHD, see Martí-Centelles, V.; Pandey,M.D.; Burguete, M.I.; Luis, S.V. Macrocyclization Reactions: The Importance of Conformational, Configurational, and Template-Induced Preorganization *Chem. Rev.* **2015**, *115*, 8736−8834.

(15) (a) Monfette, S.; Eyholzer, M.; Roberge, D. M.; Fogg, D. E. Getting Ring-Closing Metathesis off the Bench: Reaction-Reactor Matching Transforms Metathesis Efficiency in the Assembly of Large Rings *Chem. Eur. J*. **2010**, *16*, 11720-11725; (b) Sytniczuk, A.; Dabrowski, M.; Banach, L.; Urban, M.; Czarnocka-Sniadala, S.; Milewski, M.; Kajetanowicz, A.; Grela, K. At Long Last: Olefin Metathesis Macrocyclization at High Concentration *J. Am. Chem. Soc.* **2018**, *140*, 8895-8901.

(16) (a) Evans, N. H.; Allinson, E. S. H.; Lankshear, M. D.; Ng, K. Y.; Cowley, A. R.; Serpell, C. J.; Santos, S. M.; Costa, P. J.; Felix, V.; Beer, P. D. Anion Templated Assembly of [2]Catenanes Capable of Chloride Anion Recognition in Aqueous Solvent Media *RSC Adv.* **2011**, *1*, 995-1003. (b) Chase, P. A.; Lutz, M.; Spek, A. L.; van Klink, G. P. M.; van Koten, G. Ring Closing Metathesis Employing Organometallic Substrates and the Temple Synthesis of Macrocyclic Compounds *J. Mol. Catal. A* **2006**, *254*, 2-19.

(17) Pentzer, E. B.; Gadzikwa, T.; Nguyen, S. T. Substrate Encapsulation: An Efficient Strategy for the RCM Synthesis of Unsaturated ε-Lactones *Org. Lett*. **2008**, *10*, 5613-5615.

(18) For a review, see Blankenstein, J.; Zhu, J. Conformation-Directed Macrocyclization Reactions *Eur. J. Org. Chem.* **2005**, *(10)*, 1949-1964.

(19) Goldring, W. P. D.; Hodder, A. S.; Weiler, L. Synthesis of Macrocyclic Lactams and Lactones via Ring-Closing Olefin Metathesis *Tetrahedron Lett.* **1998**, *39(28)*, 4955-4958.

(20) Shu, C.; Zeng, X.; Hao, M. H.; Wei, X.; Yee, N. K.; Busacca, C. A.; Han, Z.; Farina, V.; Senanayake, C. H. RCM Macrocyclization Made Practical: An Efficient Synthesis of HCV Protease Inhibitor BILN 2061 *Org. Lett*. **2008**, *10(6)*, 1303-1306.

(21) Wei, X.; Shu, C.; Haddad, N.; Zeng, X.; Patel, N. D.; Tan, Z.; Liu, J.; Lee, H.; Shen, S.; Campbell, S.; Varsolona, R. J.; Busacca, C. A.; Hossain, A.; Yee, N. K.; Senanayake, C. H. A Highly Convergent and Efficient Synthesis of a Macrocyclic Hepatitis C Virus Protease Inhibitor BI 201302 *Org. Lett*. **2013**, *15*, 1016-1019.

(22) Kirby, A.J. in *Advances in Physical Organic Chemistry* (1980); London: Academic Press. pp. 183–278.

(23) Galli, C.; Mandolini, L. The Role of Ring Strain on the Ease of Ring Closure of Bifunctional Chain Molecules *Eur. J. Org. Chem.* **2000**, *(18)*, 3117-3125.

(24) (a) Anderson, D.R.; Hickstein, D.D.; O'Leary, D.J.; Grubbs, R.H. Model Compounds of Ruthenium-Alkene Intermediates in Olefin Metathesis Reactions *J. Am. Chem. Soc*. **2006**, *128*, 8386-8387; (b) van der Eide, E.F.; Romero, P.E.; Piers, W.E. Generation and Spectroscopic Characterization of Ruthenacyclobutane and Ruthenium Olefin Carbene Intermediates Relevant to Ring Closing Metathesis Catalysis *J. Am. Chem. Soc.* **2008**, *130*, 4485-4491.

(25) Oligomers were not intermediates in our metathesis reactions, possibly due to the significantly heavier functionalization of the substrates in this study compared to the dienes reported by Fogg *et al.*: Conrad, J.C.; Eelman, M.D.; Duarte Silva, J.A.; Monfette, S.; Parnas, H.H.; Snelgrove, J.L.; Fogg, D.E. Oligomers as Intermediates in Ring-Closing Metathesis *J. Am. Chem. Soc.* **2007**, *129*, 1024-1025.

(26) Synthesis of **16** is described in Ormerod, D.J.; Depré, D.P.M.; Horvath, A. WO 2011113859.