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Ring-Opening Cross-Metathesis (ROCM) as a Novel Tool for the Ligation of Peptides

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Abstract: The development of ring-opening cross-metathesis (ROCM) as a novel tool for the site-specific ligation of peptide units is reported. The resulting structural units at the site of ligation resulting from ROCM resemble proline as well as other known β-turn stabilising structural units. ROCM under mild reaction conditions between a variety of peptides bearing a cyclic olefin with amino acids or peptides results in high yields. The peptidic cross-partners for metathesis are equipped with double bonds via the *N* and the *C* terminus and the side chain, respectively, to allow the synthesis of linear as well as non-linear and branched peptides. The ligation in this manner succeeds with low catalyst loadings, with no need for any excess of one reaction partner and with a high compatibility with a wide range of functional groups. Furthermore, the stereochemical outcome of the ROCM can easily be controlled by using a Hoveyda-type chiral catalyst. Fluorescence labelling of peptides is possible in the same manner when using a cyclic olefin equipped with a fluorescence marker.

Keywords: bioorganic chemistry · diastereoselectivity · ligation techniques · metathesis · peptidomimetics

Introduction

Numerous techniques have been developed to synthesise large peptide fragments that have led to the elucidation of gene function and structure-function relationships, making possible the discovery of novel biology and new therapeutic agents. The segmental approach that ligates building blocks in peptide synthesis without using any coupling reagents is a superior strategy to the stepwise approach.[1] As a result it has become more common during the last 15 years.

An alternative strategy in the synthesis of peptide segments involves appending two peptide building blocks to the amine and carboxyl group of a secondary structural unit via the *C* and *N* termini of these fragments, by using known coupling reagents. The use of these non-peptidic structural units results in a stabilisation of β-turn regions and bioactive conformations, and therefore many peptide analogues containing such structures exhibit higher biological activity.[2]

Both ligation techniques require chemoselectivity, avoiding reaction with the manifold functional groups of amino acids. Ideally, ligation should be carried out stereoselectively under mild conditions with low catalyst loadings in high yields. As both substrates (for example, two peptide building blocks) are expensive biomolecules, there should be no need for any excess of one reaction partner. Furthermore, the ligation technique should allow the synthesis of a linear as well as a non-linear and a branched peptide.

Olefin metathesis is one of the most versatile tools for C–C-bond formation and meets all the above demands.[3] We were therefore interested in applying this classic organometallic reaction to the biochemical field of ligation and derivatization of peptides. We wished to exploit the advantages of the atom-economical ring-opening cross-metathesis (ROCM) for the segmental ligation of peptide building blocks. The resulting structural unit from ROCM should in principal be able to mimic the role of β-turn stabilising systems, such as proline, leading to a ligation approach towards β-turn mimetics.

Results and Discussion

Recently, two interesting ligation techniques were reported. One takes advantage of the Huisgen azide cycloaddition,[4] the other of the Diels–Alder reaction.[5] Both techniques offer an advantageous new opportunity for the ligation of proteins and peptides. They proceed under mild conditions

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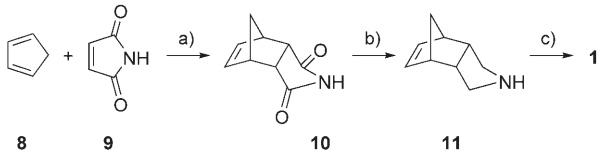
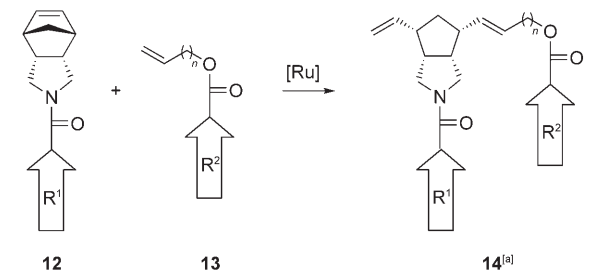
with high selectivity and are compatible with most functional groups found in proteins.

Herein, we present the development of a new concept for the efficient ligation of peptides by ROCM. Cyclic amines, such as **1**, equipped with a peptide segment, undergo ROCM with different metathesis cross-partners **2**, **3** or **4**, leading to ligation of the peptide segments (Scheme 1). Conceptually, the ligation can be achieved in a linear (**5**), nonlinear (**6**) or branched (**7**) manner by using cross-partners of types A–C. Instead of expendable separations, a homogeneous and stereochemically well-defined product should be achieved by using chiral Hoveyda-type metathesis catalysts and perhydrogenation of the resulting double bonds in **5**, **6** and **7**. Cross-partners **2** and **3** can be easily prepared by amide bond formation with the peptide *N* terminus in **2** and esterification with the peptide *C* terminus in **3**, respectively. Cross-partner **4** is a peptide containing allyl-glycine as one amino acid. Amide bond formation of amine **11**, accessible by sequential Diels–Alder reaction of cyclobutadiene **8** and maleimide **9** and reduction, leads to the cyclo-olefin **1** (Scheme 2).

To optimise the reaction conditions, we first coupled the cyclic amine **11** with Fmoc-protected glycine (Fmoc = 9-fluorenylmethoxycarbonyl) and used the glycine-ester **13a** as the coupling partner for metathesis (Scheme 3). As shown in Table 1, a solution of a 1:1 mixture of **12a** and **13a** in either dichloromethane or 1,2-dichlorethane was stirred at different temperatures by using 1 mol% of the metathesis catalysts [Ru-**1**] to [Ru-**4**]. The conversion was monitored by HPLC-ESI. A histogram of the relative product distribution of **14a**, versus olefin **12a** and peptide **13a** is shown in Figure 1. After chromatography, the product **14** was isolated as a mixture of *E* and *Z* isomers as well as a diastereoisomeric mixture.



Scheme 2. Synthesis of cyclo-olefin **1**. a) Diethyl ether, 23 °C, 2 h, 96%; b) $LiAlH_4$ (4.1 equiv), THF, 48 h, reflux, 84%; c) general procedure: 0.83 to 1.0 equiv amino acid or peptide (Fmoc-protected), 1.0 to 2.0 equiv TBTU, dichloromethane/DMF, 0 °C/0.5 h, 23 °C/1 h.

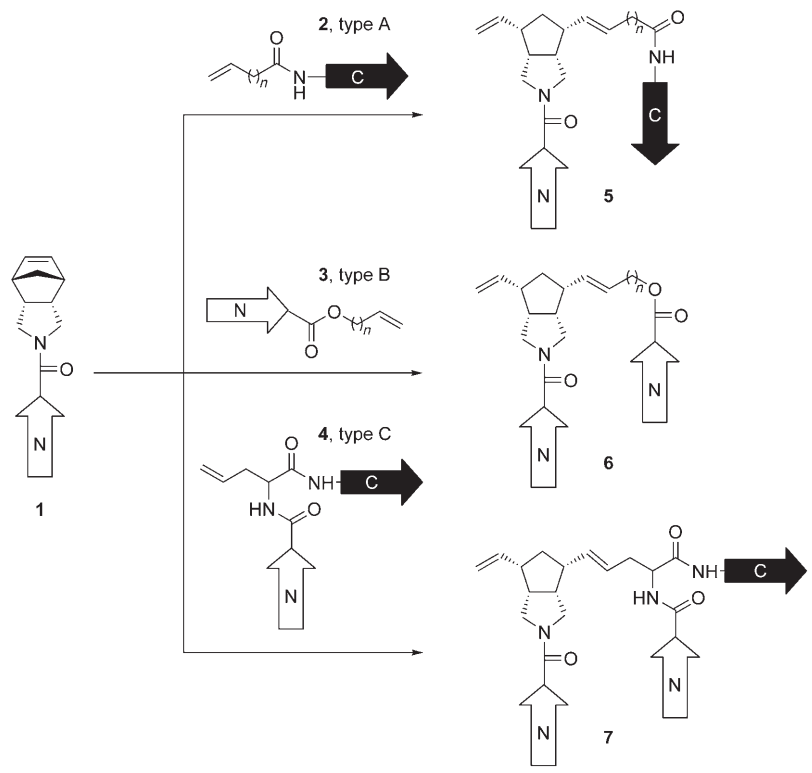


Scheme 3. [Ru]-catalyzed ROCMs of **12** and **13**. [a] Isolated as a mixture of *E* and *Z* isomers as well as a diastereoisomeric mixture.

We found the first-generation Hoveyda catalyst [Ru-**3**] to be superior to the others, as the more active second-generation catalysts [Ru-**2**] and [Ru-**4**] led to significant polymerisation as well as rapid consumption of the starting material **12a** (Figure 1).

To demonstrate the compatibility of the ROCM method with various functional groups, we investigated the ligation of peptides **12a–k** with **13a–c** (Table 2). In accordance with our optimized reaction conditions, the cross-partners were stirred in dichlormethane (0.1 M) for 17 h at 40 °C by using generally 1–5 mol% of the metathesis catalyst [Ru-**3**]. Selected examples are given in Table 2.

As expected for the aforementioned example (entry 1) and its homologues (entries 2 and 3), we were able to isolate the coupled product **14** in high

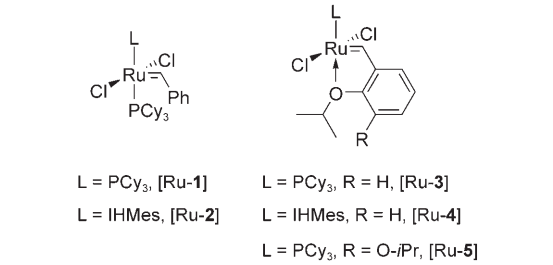


Scheme 1. General concept of peptide ligation by means of ROCM.

Table 1. ROCMs of **12a** (R^1 = Fmoc-Gly-) and **13a** (R^2 = Fmoc-Gly-).

Entry	Catalyst (1 mol %)	T [°C]	Solvent	c [mol L^{-1}]
1	[Ru-**1**][a]	40	dichloromethane	0.01
2	[Ru-**2**]	40	dichloromethane	0.01
3	[Ru-**2**]	60	DCE	0.01
4	[Ru-**3**]	40	dichloromethane	0.1
5	[Ru-**3**]	40	dichloromethane	0.01
6	[Ru-**3**]	60	DCE	0.1
7	[Ru-**3**]	60	DCE	0.01
8	[Ru-**4**]	40	dichloromethane	0.01
9	[Ru-**4**]	60	DCE	0.01

[a] 2 mol % of [Ru-**1**] was used.



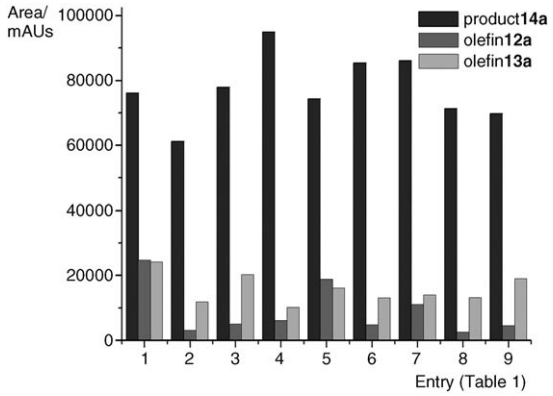


Figure 1. Results for the [Ru]-catalyzed ROCMs of **12a** and **13a**.

yields following sequential purification by silica gel and Sephadex chromatography. The UV chromatographs of the crude product **14b** (entry 2) as well as for the product after each purification step are shown in Figure 2 as an example. The presence of two peaks A and B in the product (retention time 6.5–8.0 min) represents the *E*/*Z* isomers of product **14b**. This was supported by perhydrogenation of pure **14b**, the product of which displayed a single peak by HPLC-ESI

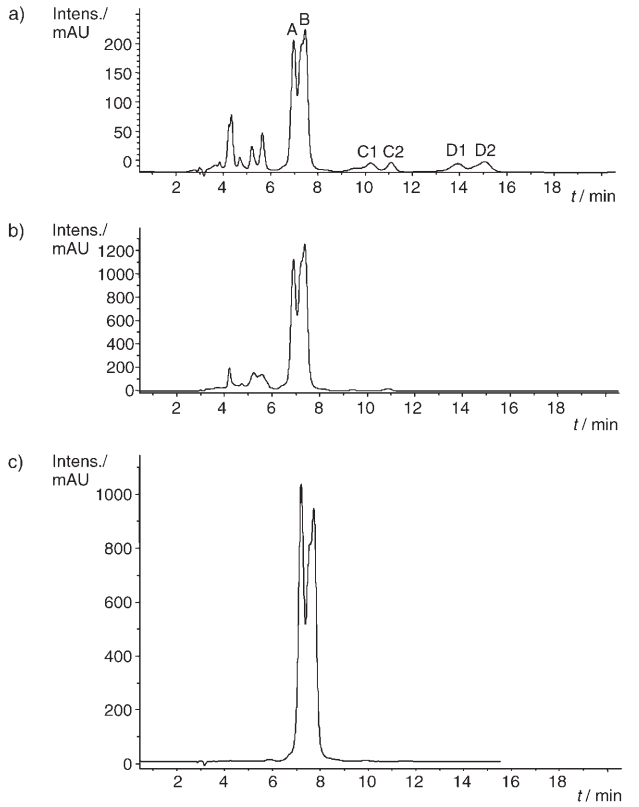


Figure 2. UV chromatograph of **14b**. a) crude product, b) after column chromatography, c) after Sephadex chromatography.

Table 2. Results for the peptide ligation by means of ROCM.

Entry	**12**	R^1	**13**[a]	R^2	Catalyst (mol %)	Product	**14**/cdp[b]	Yield [%]	E/Z[c]
1	**12a**	Fmoc-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (2)	**14a**	80:20	80	78:22
2	**12b**	Fmoc-Phe-	**13a**	Fmoc-Gly-	[Ru-**3**] (2)	**14b**	85:10[d]	74	63:37
3	**12c**	Fmoc-Val-	**13a**	Fmoc-Gly-	[Ru-**3**] (2)	**14c**	83:11[d]	82	65:35
4[e]	**12d**	Fmoc-Gly-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**2**] (1)	**14d**		35	
5[e]	**12d**	Fmoc-Gly-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**4**] (1)	**14d**		51	
6	**12d**	Fmoc-Gly-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (1)	**14d**	93:7	75	48:52
7[f]	**12d**	Fmoc-Gly-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**5**] (1)	**14d**		80	
8	**12e**	Fmoc-Phe-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (1)	**14e**	91:9	83	64:36
9	**12f**	Fmoc-Asn-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (1)	**14f**	77:23	37	67:33
10	**12g**	Fmoc-Lys(Boc)-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (5)	**14g**	90:10	80	64:36
11	**12h**	Fmoc-Cys(Trt)-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (1)	**14h**	>95:<5	83	64:36
12	**12i**	Fmoc-Cys-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**3**] (5)	**14i**	–	<5	–
13[g]	**12i**	Fmoc-Cys-Gly-Gly-	**13a**	Fmoc-Gly-	[Ru-**4**] (5)	**14i**		30	
14	**12e**	Fmoc-Phe-Gly-Gly-	**13b**	Fmoc-Leu-Ala-Pro-	[Ru-**3**] (2)	**14k**		82	
15	**12k**	Fmoc-Leu-Ala-Pro-	**13c**	Fmoc-Cys(Trt)-Gly-Gly-	[Ru-**3**] (2)	**14l**		66	
16	**12k**	Fmoc-Leu-Ala-Pro-	**13b**	Fmoc-Leu-Ala-Pro-	[Ru-**3**] (2)	**14m**		74	

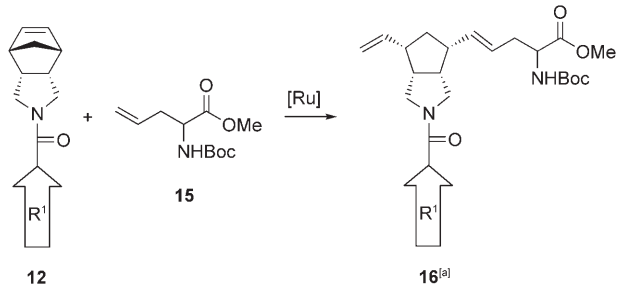
[a] Cross-partner. [b] Determined by HPLC-ESI, cdp: crossed-dimer product. [c] Determined by ^{1}H NMR spectroscopy. [d] 5–6 % double-crossed product (dcp). [e] 0.01 M, 1 h. [f] 6 h. [g] 2 equiv cross-partner, 1,2-dichlorethane, 70 °C, 0.012 M, 1 h.

analysis. This fulfilled our desire for access to a stereochemically well-defined product by simple perhydrogenation. Peaks C1 and C2 (retention time 10–12 min) correspond to the double crossed product (dcp), and peaks at 13–16 min (D1 and D2) to the crossed dimer product (cdp), as judged by their mass spectral analysis. The appearance of a shoulder in product peak B led us to attempt HPLC methods to revolve these peaks. By using more polar eluents, four peaks were displayed, representing the diasteroisomers in addition to the *E*/*Z* isomers. We did not try to separate the diastereomeric product mixture but instead investigated the influence of chiral metathesis catalysts.

To probe the reaction further, peptides with a Gly-Gly spacer were synthesized and used as ROCM substrates (entries 4–14). Glycine is one of the most common amino acids present in native β-turn regions because of the increased flexibility that accommodates the conformation of the peptide. Additionally, we observed that lower catalyst loadings (1 instead of 2 mol%) could be employed for substrates bearing a Gly-Gly spacer (comparing entries 1/6 and 2/8). We attribute this to the decreased interactions between the potentially coordinating Fmoc groups and the reaction centre during ROCM. In these Gly-Gly substrates, [Ru-**3**] displayed again the best reactivity, as demonstrated by comparison of entries 4 to 6. However, [Ru-**5**],[6] a Blechert-group catalyst gave a higher yield still (80%) of ligated peptide **14d**, in a dramatically reduced reaction time (entry 7). A starting material containing asparagine (**12f**, entry 9) resulted in the formation of large amounts of dimers and polymeric side products as observed as an insoluble material in the reaction flask. Nevertheless, 37% of coupling product **14f** was isolated. To investigate the compatibility of nucleophelic functional groups with ROCM, we examined the coupling reaction with peptidic cross-partners bearing either protected or unprotected SH groups. Whereas the trityl-protected peptide **12h** (entry 11) reacted as expected, the unprotected cysteine derivative **12i** (entry 12) was not converted when using [Ru-**3**]. A reasonable explanation is the deactivation or decomposition of the catalyst by the strongly nucleophilic SH group, inhibiting ROCM. By using the more active and functional-group compatible catalyst of the second generation [Ru-**4**] at optimized conditions (5 mol% catalyst, 70°C, 0.012 M, 1 h) gave more than 50% of dimers, <10% starting material and 30% of the coupling product **14i**, which is an acceptable result for olefin metathesis with these substrates. Higher yields might result in the case of bigger peptides if strongly nucleophilic functional groups are internally located in peptides.

We next prepared hexapeptides with a wide variety of amino acids (entries 14–16). The tripeptides **12e**,**k** and **13b**,**c** underwent ROCM with [Ru-**3**] giving the coupling products **14k–m** in 66–82% yields.

Next, we tried to expand the new methodology for ligation with type C cross-partners (Schemes 1 and 4). N- and C-protected allylglycine **15** was, therefore, used as a type C cross-partner model and we investigated its ROCM with cyclic olefins **12** (Table 3).



Scheme 4. [Ru]-catalyzed ROCMs of **12** and type C cross-partner **15**. [a] Isolated as a mixture of *E* and *Z* isomers as well as a diastereoisomeric mixture.

Table 3. Results for the [Ru]-catalyzed ROCMs with type C cross-partner **15**.

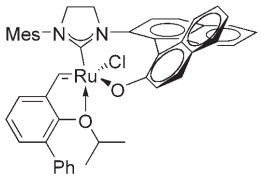
Entry	**12**	R^1	**15** [equiv][a]	Catalyst (mol%)	**16**	*c* [mol L^{-1}]	**12**/**16**/cdp[b]
1	**12a**	Fmoc-Gly-	1	[Ru-**3**] (2)	**16a**	0.1	>98:<2:0
2	**12a**	Fmoc-Gly-	2	[Ru-**4**] (1)	**16a**	0.02	>99:<1:0
3	**12a**	Fmoc-Gly-	1	[Ru-**4**] (2)	**16a**	0.1	5:56:39
4	**12d**	Fmoc-Gly-Gly-Gly-	1	[Ru-**3**] (2)	**16b**	0.1	84:11:5
5	**12d**	Fmoc-Gly-Gly-Gly-	1	[Ru-**3**] (10)	**16b**	0.1	1:72:27
6	**12d**	Fmoc-Gly-Gly-Gly-	2	[Ru-**4**] (1)	**16b**	0.02	1:59:40
7	**12d**	Fmoc-Gly-Gly-Gly-	1	[Ru-**4**] (2)	**16b**	0.1	2:60:38

[a] Cross-partner. [b] Determined by HPLC-ESI, cdp: crossed-dimer product.

In general, we found that the type C cross-partner gave a branched peptidelike product **16** (Table 3) as a mixture of *E* and *Z* isomers as well as a diastereoisomeric mixture. As the optimised reaction conditions for type B cross-partners did not lead to any significant conversion to peptides **16** (entries 1 and 4), we increased the amount of catalyst [Ru-**3**] (entry 5) which gave large amounts of dimers as detected by HPLC-ESI. Further investigation showed that the second-generation catalyst [Ru-**4**] gave the best results (entries 6 and 7) when using two equivalents of cross-partner **15**. By using these conditions, we were able to isolate 33% of the ring-opened cross-product **16b** after purification on silica gel and Sephadex. The good results of the peptide ligation with type B cross-partners as well as the observations with different spacers in the manner of fluorescence labelling (see below), indicates that higher yields of coupled products are obtainable when decreasing the steric hindrance of the substrates or by increasing the length of the side chain to the double bond.

Additionally, we investigated the ability of asymmetric metathesis catalysts to influence the stereochemical outcome of ROCM as it is important for stereoselective synthesis (of

pharmaceutical compounds). To investigate this, we analysed the stereochemical reaction outcomes of our peptide ligation reactions by HPLC-ESI when using the Hoveyda-type chiral catalyst [Ru-**6**]*.[7] This nonphosphine Ru carbene bears a bidentate styrene ether ligand and is currently one of the best asymmetric catalysts which enables reactions to be carried out under air and with undistilled solvents.



[Ru-**6**]*

In preliminary reactions (Scheme 3, Table 4, entries 1 and 3), we found that the conversion of **12** and **13** needed elevated reaction temperatures of 60 °C, probably as a result of various steric and electronic factors of [Ru-**6**]* in contrast to the achiral catalysts. Furthermore, the selectivity of [Ru-**6**]*-catalyzed ROCMs is not dependent on the solvent. While ROCM in toluene gave lower conversion and diastereoisomeric excess, ROCM carried out in 1,2-dichlorethane and THF gave better comparative results. For this, we analysed the catalytic activity of [Ru-**6**]* on a range of reactions in 1,2-dichlorethane.

Table 4. Results for the asymmetric [Ru]*-catalyst ROCMs.

Entry	**12**	R^1	**13**[a]	R^2	**14**	Catalyst/ mol %	**13**/ **14**[b]	*de* [%][b]	*E*/ *Z*[b]
1	**12b**	Fmoc-L-Phe-	**13a**	Fmoc-Gly-	**14b**	3	90:10	74	–
2	**12l**	Fmoc-D-Phe-	**13a**	Fmoc-Gly-	**14n**	3	45:55	70	87:13
3	**12c**	Fmoc-L-Val-	**13a**	Fmoc-Gly-	**14c**	2	38:62	62	84:16
4	**12m**	Fmoc-D-Val-	**13a**	Fmoc-Gly-	**14o**	2	54:46	63	84:16
5	**12k**	Fmoc-Leu-Ala-Pro-	**13b**	Fmoc-Leu-Ala-Pro-	**14m**	2	85:15	61	87:13

[a] Cross-partner. [b] Determined by HPLC-ESI.

As shown in Table 4, olefin metathesis of **12** and **13** led to products with excellent diastereoisomeric excesses (*de*) compared to the examples reported by the group of Hoveyda.[7–9] For example, the cyclic olefin **12b** bearing *N*-Fmoc-phenylalanine reacted with cross-partner **13a** in 55% yield and 70% *de* to give **14b** when using 3 mol% of [Ru-**6**]*. In contrast to Hoveyda's work, we used one instead of five equivalents of the cross-partner and lower amounts of catalyst, which might be the reason for incomplete conversions. As the ROCM stereochemical outcome using D or L starting materials (entries 1–2 and 3–4) lie within a close range, the influence of the substrate was considered to be negligible.

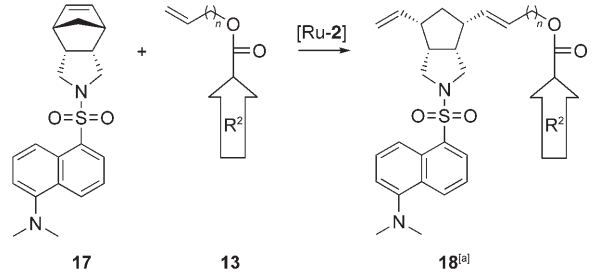
Finally, we wished to expand the concept to fluorescence labelling of peptides as it is an important application in peptide chemistry. The fluorescence-labelled cyclic olefin **17** is easily accessible from amine **11** and commercially available dansyl chloride. Sulfonamide **17** and one equivalent of the cross-partner **13** were treated with 1 mol% of the second-generation Grubbs catalyst [Ru-**2**] and stirred at 40 °C. After 30 min, the reaction was quenched with ethyl vinylsilane, and after monitoring the conversion by ^{1}H NMR analysis of the crude mixture, the product was purified by preparative TLC.

The fluorescence-labelled product **18b** was isolated in 67% yield (Table 5, entry 2) and identified by NMR spectroscopy and MS. We found the conversion of the metathesis reaction was dependant on the spacer as the conversion as well as the isolated yields of the reactions with shorter (entry 1) or longer spacers (entry 3) decreased dramatically. Nevertheless, these results show that peptides can be easily labelled by ROCM with the olefin **17** (Scheme 5).

Table 5. Results for the fluorescence labelling by means of ROCM.

Entry	**13**[a]	**18**	*n*	Conv. [%][b]	Yield [%]
1	**13d**[c]	**18a**	2	60	43
2	**13e**[c]	**18b**	4	80	67
3	**13f**[c]	**18c**	9	60	49

[a] Cross partner. [b] Determined by ^{1}H NMR spectroscopic analysis. [c] R^2 = Trt-Cys(Mmt)-Ser-Val-.



Scheme 5. [Ru]-catalyzed ROCMs of fluorescence-labelled **17** and **13**. [a] Isolated as a mixture of *E* and *Z* isomers as well as a diastereoisomeric mixture.

Conclusion

Our results demonstrate the power of ROCM in C–C-bond formation as a means of ligating smaller peptide units. By this method, the ligation can be achieved in a linear as well as in a non-linear and a branched sense, in good yields and under mild conditions, with low catalyst loadings and with no need for any excess of one reaction partner. We were able to demonstrate a high compatibility with a wide range of functionalised amino acids. The first generation Hoveyda catalyst [Ru-**3**] as well as its variants were superior to the more active catalysts of the second generation, which led to higher amounts of polymeric byproducts. The stereochemical outcome can be controlled by using the chiral catalyst [Ru-**6**]*. Relative to the published results of Hoveyda,[7,9] we

were able to obtain the ROCM products with excellent diastereomeric excesses, which should lead to a homogenous and stereochemically well-defined product after perhydrogenation.

Currently, we are investigating the peptide ligation in aqueous media, as peptides with a higher number of amino acids have a limited solubility in organic solvents. Furthermore, we are interested in the ability of the secondary structural unit at the site of ligation to stabilize or initiate β-turns.

Experimental Section

General methods and materials: Unless otherwise noted, all reactions were performed in dried glassware under air. All solvents were dried and distilled prior to use. Commercially available reagents were used as received without further purification. Column chromatography was carried out by using 60 μm silica gel from Merck. Sephadex LH-20 was supplied from Pharma Biotech. Analytical reversed-phase HPLC-(MS) was carried out on a Varian ProStar system (autosampler model 410; UV-visible-detector model 320; solvent delivery module model 210; fraction collector model 701) by using a Waters X-Terra (RP-18, 4.6*100 mm) column and on a Hewlett–Packard system series 1100 on a Waters Symmetry, RP-18, 3.9*150 mm column in combination with Bruker-Daltonics esquire 2000 (ESI), detection at 210 and 254 nm and a linear gradient of solvent B in solvent A at 0.5 mL min^{-1} flow rate. HPLC-grade solvents were delivered by Acros and Fisher Scientific. Calculated masses were obtained by using the program molecular weight calculator by Matthew Monroe Version 6.35. ^{1}H and ^{13}C NMR spectra were recorded on an AM 400 (400 MHz) and DRX 500 spectrometer (500 MHz) from Bruker. Chemical shifts are reported in ppm relative to $CHCl_3$ ($\delta = 7.27$ ppm) for ^{1}H NMR and the central resonance of $CDCl_3$ ($\delta = 77.0$ ppm) for ^{13}C NMR spectra. IR spectra were obtained on a Perkin–Elmer 881 and on a Nicolet Magna 750 Series FTIR. HRMS were obtained on a Finnigan MAT 95 SQ mass spectrometer. Optical rotations were measured on a polarimeter as solutions in a 10 cm unit cell at 589 nm. Trt = triphenylmethyl; Mmt = monomethoxytrityl.

General procedure for peptide synthesis

DCC method: The coupling partner (5-hexene-1-ol or C-protected amino acid; 1.0–1.2 equiv), 1,3-dicyclohexylcarbodiimide (DCC, 1 to 2 equiv) and 4-dimethylaminopyridine (DMAP, 0.17 equiv) were added to a solution of the Fmoc-protected amino acid (unless otherwise noted: 0.2 M in dichloromethane) at 0 °C. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. The solid was removed by filtration and the solution was transferred to a separatory funnel. It was quenched by addition of water (same volume as the organic layer). After separation, the aqueous layer was extracted with dichloromethane twice. The combined organic layers were dried over Na_2SO_4. The drying agent was removed by filtration and solvents were removed under vacuum. The crude product was purified by column chromatography and, if noted, additionally by Sephadex chromatography.

TBTU method A: The coupling partner (**11** or C-protected amino acid; 1 equiv) and *O*-benzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium tetrafluoroborate (TBTU, 1 to 2 equiv) at 0 °C were added to a solution of the Fmoc-protected amino acid (solvents mentioned in examples). The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. It was diluted with dichloromethane and washed with H_2O (2× with the same volume). The organic layer was dried over Na_2SO_4. The drying agent was removed by filtration and the solvents were removed under vacuum. The crude product was purified by column chromatography and, if noted, additionally by Sephadex chromatography.

TBTU method B: The TFA salt of the C-protected peptide (1 equiv), TBTU (1.0 to 2.0 equiv) and *N*,*N*-diisopropylethylamine (DIPEA, 2.2 equiv) at 0 °C were added to a solution of the Fmoc-protected amino acid (solvents mentioned in examples). The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. It was diluted with dichloromethane and washed with H_2O (2× with the same volume). The combined organic layers were dried over Na_2SO_4. The drying agent was removed by filtration and the solvents were removed under vacuum. The crude product was purified by column chromatography and, if noted, additionally by Sephadex chromatography.

Cyclic olefins

Imine (10): Freshly distilled cyclopentadiene (2.8 mL, 33.3 mmol, 1.01 equiv) was added to a solution of maleimide (3.2 g, 33 mmol) in diethyl ether (80 mL). The reaction mixture was stirred for 2 h at 23 °C. After this time, the solid product was separated by filtration and dried under air. Recrystallisation from diethyl ether/methanol 2:1 gave product **10** (5.2 g, 31.7 mmol, 96 %) as white crystals. M.p. 170–172 °C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.53$ (d, $J = 9.0$ Hz, 1H), 1.76 (d, $J = 9.0$ Hz, 1H), 3.24–3.44 (m, 4H), 6.21 (2H, AB-syst.), 7.62 ppm (s, 1H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta = 45.0$ (2CH), 47.4 (2CH), 52.4 (2CH_2), 134.6 (2CH), 178.5 ppm (2C_q); IR (ATR): $\tilde{\nu} = 3158$, 3063, 2991, 2962, 2954, 1753, 1718, 1699, 1354, 1167, 736 cm^{-1}; MS (EI): *m*/*z* (%): 163 (13), 98 (34), 91 (53), 66 (100); HRMS: calcd for $C_9H_9O_2N$: 163.0633; found: 163.0631 [M^+].

Amine (11): Under a nitrogen atmosphere in a thoroughly dried apparatus containing a three-necked flask, a dropping funnel and a condenser, a solution of **10** (1.63 g, 10 mmol) in THF (35 mL) was added dropwise to a suspension of $LiAlH_4$ (1.56 g, 41 mmol, 4.1 equiv) in THF (35 mL) through the dropping funnel. The reaction mixture was stirred for 48 h under reflux. After cooling down, diethyl ether (150 mL) was added and the mixture was hydrolysed by the successive addition of ethyl acetate, methanol and water until a white solid precipitated. The solid was removed by filtration, and solvents were removed under vacuum. Kugelrohrdestillation (3×10^{-2} mbar, 45 °C) of the pale-yellow crude product gave amine **11** (1.14 g, 8.4 mmol, 84 %) as a white, waxlike solid. ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 1.52$ (d, $J = 9.0$ Hz, 1H), 1.65 (d, $J = 8.8$ Hz 1H), 2.85 (dd, $J = 12.0$, 2.6 Hz, 2H), 2.96 (d, $J = 1.0$ Hz, 2H), 3.03–3.09 (m, 2H), 3.16–3.22 (m, 2H), 6.43 ppm (s, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta = 46.4$ (2CH), 48.0 (2CH), 49.6 (2CH_2), 53.1 (2CH_2), 135.9 ppm (2CH); IR (ATR): $\nu = 3221$, 3056, 2960, 2937, 2866, 1713, 1623, 1570, 1350, 735 cm^{-1}; MS (EI): *m*/*z* (%): 135 (12), 94 (18), 68 (100), 66 (13); HRMS: calcd for $C_9H_{13}N$: 135.1048; found: 135.1054 [M^+].

Fmoc-Gly-cyclo-olefin (12a): According to the general TBTU method A, amine **11** (100 mg, 0.74 mmol) in dichloromethane (2 mL) and TBTU (285 mg, 0.89 mmol, 1.2 equiv) were added to a solution of Fmoc-protected glycine (220 mg, 0.74 mmol) in DMF (3.5 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (hexanes/ethyl acetate 1:1), **12a** (142 mg, 0.34 mmol, 46 %) was obtained as a white crystalline solid. M.p. 70–72 °C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.45$ (d, $J = 8.5$ Hz, 1H), 1.58 (d, $J = 8.5$ Hz, 1H), 2.90–3.04 (m, 5H), 3.28–3.38 (m, 3H), 3.82 (d, $J = 3.2$ Hz, 2H), 4.23 (t, $J = 7.3$ Hz, 1H), 4.35 (d, $J = 7.3$ Hz, 2H), 5.81 (s, 1H), 6.19 (m, 2H), 7.31 (dt, $J = 7.5$, 1.0 Hz, 2H), 7.39 (t, $J = 7.5$ Hz, 2H), 7.60 (d, $J = 7.5$ Hz, 2H), 7.74 ppm (d, $J = 7.5$ Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta = 43.5$ (CH_2), 43.8 (CH), 45.8 (CH), 46.7 (CH), 47.1 (CH), 47.9 (CH_2), 48.2 (CH_2), 51.9 (CH_2), 67.1 (CH_2), 119.9 (CH), 125.2 (CH), 127.1 (CH), 127.7 (CH), 134.7 (CH), 136.0 (CH), 141.3 (C_q), 143.9 (C_q), 156.2 (C_q), 165.4 ppm (C_q); IR (ATR): $\tilde{\nu} = 3267$, 3062, 3037, 3028, 2966, 2944, 2870, 1713, 1627, 1532, 1451, 1246, 1032, 741 cm^{-1}; LRMS (70 eV, EI): *m*/*z* (%): 414 (41), 367 (44), 368 (100), 313 (21), 284 (42), 281 (23), 178 ppm (>100); HRMS: calcd for $C_{26}H_{26}O_3N_2$: 414.1963; found: 414.1952 [M^+].

Fmoc-Phe-cyclo-olefin (12b): According to the general DCC method, amine **11** (270 mg, 2 mmol), DCC (825.3 mg, 4 mmol, 2 equiv) and DMAP (41.5 mg, 0.34 mmol, 0.17 equiv) were added successively to a solution of Fmoc-protected phenylalanine (775 mg, 2 mmol) in dichloromethane (4 mL) at 0 °C. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. After filtration, aqueous workup and column chromatography (hexanes/ethyl acetate 3:1→1:1) **12b** (477 mg, 0.94 mmol, 47 %) was obtained as a white crystalline solid. M.p. 77–79 °C; $[\alpha]_D^{20} = +6.8°$ ($c = 0.99$ in CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta =$

1.33 (d, $J=8.6$ Hz, 0.5H), 1.39 (d, $J=8.6$ Hz, 0.5H), 1.46–1.53 (m, 1H), 2.57 (t, $J=10.0$ Hz, 0.5H), 2.65 (dd, $J=10.6$, 3.6 Hz, 1H), 2.70–2.77 (m, 0.5H), 2.79 (s, 0.5H), 2.82 (s, 0.5H), 2.87 (s, 2H), 2.90 (d, $J=6.0$ Hz, 0.5H), 2.92–2.97 (m, 1H), 2.98–3.05 (m, 1H), 3.07 (dd, $J=11.0$, 2.0 Hz, 0.5H), 3.14 (dd, $J=13.0$, 9.4 Hz, 0.5H), 3.28 (dd, $J=12.6$, 3.0 Hz, 0.5H), 3.34 (dd, $J=12.6$, 9.2 Hz, 0.5H), 3.54 (t, $J=10.0$ Hz, 0.5H), 4.15- 4.22 (m, 1H), 4.24–4.40 (m, 2H), 4.55 (dd, $J=7.4$, 6.6 Hz, 0.5H), 4.59 (dd, $J=8.0$, 6.0 Hz, 0.5H), 5.64 (dd, $J=5.0$, 2.6 Hz, 0.5H), 6.02 (dd, $J=5.0$, 2.0 Hz, 0.5H), 6.07–6.10 (m, 0.5H), 6.11–6.15 (m, 0.5H), 7.17 (t, $J=7.6$ Hz, 2H), 7.22–7.33 (m, 5H), 7.37–7.42 (m, 2H), 7.53–7.60 (m, 2H), 7.73–7.80 ppm (m, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=39.2$ (CH_2), 40.0 (CH_2), 43.5 (CH), 44.0 (CH), 45.6 (CH), 46.0 (CH), 46.4 (CH), 46.4 (CH), 46.5 (CH), 46.6 (CH), 47.2 (CH_2), 47.9 (CH_2), 48.5 (CH_2), 48.8 (CH_2), 49.2 (CH_2), 51.8 (CH_2), 52.0 (CH_2), 53.6 (CH), 53.8 (CH), 67.1 (CH_2), 120.0 (CH), 125.2 (CH), 125.3 (CH), 127.0 (CH), 127.1 (CH), 127.8 (CH), 128.5 (CH), 128.6 (CH), 129.5 (CH), 129.7 (CH), 135.2 (CH), 135.4 (CH), 135.5 (CH), 135.8 (CH), 136.3 (C_q), 136.4 (C_q), 141.4 (C_q), 144.0 (C_q), 155.7 (C_q), 155.8 (C_q), 168.7 (C_q), 169.1 ppm (C_q); IR (ATR): $\tilde{\nu}=3260$, 3062, 2964, 2870, 1715, 1628, 1533, 1451, 1247, 742 cm^{-1}; MS (EI): m/z (%): 504 (2), 191 (14), 179 (96), 178 (100), 120 (29), 69 (36); HRMS: calcd for $C_{33}H_{32}O_3N_2$: 504.2413; found: 504.2417 [M^+].

Fmoc-Val-cyclo-olefin (12c): According to the general TBTU method A, amine **11** (100 mg, 0.74 mmol) in dichloromethane (2 mL) and TBTU (285 mg, 0.89 mmol, 1.2 equiv) were added to a solution of Fmoc-protected valine (251 mg, 0.74 mmol) in DMF (3.5 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (hexanes/ethyl acetate 2:1→3:2), **12c** (178 mg, 0.39 mmol, 53 %) was obtained as a white crystalline solid. $[\alpha]_D^{20}=+1.2°$ ($c=0.49$ in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): $\delta=0.89$–0.96 (m, 6H), 1.36–1.58 (m 2H), 1.90–2.00 (dq, $J=6.7$, 3.3 Hz, 1H), 2.91–2.96 (m, 4H), 3.14–3.38 (m, 2H), 3.39–3.43 (m, 2H), 4.16–4.22 (m, 2H), 4.30–4.39 (m, 2H), 5.52 (m, 1H), 6.17 (m, 2H), 7.31 (m, 2H), 7.40 (m, 2H), 7.59 (m, 2H), 7.77 ppm (dd, $J=7.5$, 2.5 Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=17.6$ (CH_3), 17.6 (CH_3), 18.5 (CH_3), 18.6 (CH_3), 31.1 (CH), 31.3 (CH), 43.5 (CH), 43.8 (CH), 45.7 (CH), 45.8 (CH), 46.6 (CH), 46.6 (CH), 46.7 (CH), 47.1 (CH), 47.9 (CH), 48.1 (CH_2), 48.7 (CH_2), 49.7 (CH_2), 49.8 (CH_2), 51.7 (CH_2), 52.0 (CH_2), 57.4 (CH), 57.5 (CH), 67.0 (CH_2), 76.7 (CH_2), 119.9 (CH), 125.2 (CH), 127.0 (CH), 127.7 (CH), 135.1 (CH), 135.4 (CH), 136.0 (CH), 141.3 (C_q), 144.0 (C_q), 156.2 (C_q), 165.4 ppm (C_q); IR (ATR): $\tilde{\nu}=3260$, 3062, 3038, 2964, 2938, 2871, 1714, 1628, 1450, 1238, 1031, 740 cm^{-1}; LRMS (70 eV, EI): m/z (%): 456 (3), 390 (3), 294 (4), 260 (4), 179 (91), 178 (100), 165 (34); HRMS: calcd for $C_{29}H_{32}O_3N_2$: 456.2413; found: 456.2420 [M^+].

TFA·Gly-Gly-cyclo-olefin (19): Aqueous NaOH (10 mL, 1 N) and $(Boc)_2O$ (Boc = *tert*-butoxycarbonyl, 2.4 g, 11 mmol, 1.1 equiv) were added to a solution of Gly-Gly (1.32 g, 10 mmol) in dioxane and water (2:1, 30 mL) at 0 °C. The reaction mixture was stirred for 1 h at 23 °C. The dioxane was removed under reduced pressure. The aqueous layer was diluted with ethyl acetate (20 mL) and the pH was adjusted to pH 2–3 by using $KHSO_4$. The layers were separated and the aqueous layer was extracted with ethyl acetate (5×20 mL). The combined organic layers were washed with water (50 mL) and dried over Na_2SO_4. Removing the drying agent by filtration and the solvent under reduced pressure delivered a crude product (1.5 g, 6.5 mmol, 65 %). Amine **11** (1.14 g, 8.45 mmol, 1.3 equiv) in dichloromethane (26 mL), DCC (1.61 g, 7.8 mmol, 1.2 equiv) in dichloromethane (2 mL) and DMAP (140 mg, 1.1 mmol, 0.17 equiv) were added to a solution of the crude product in THF (26 mL) at 0 °C, according to the general DCC method. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. Removing the drying agent and solvent delivered a crude product (1.82 g, 5.2 mmol, 80 %) which was dissolved in dichloromethane (15 mL). To the solution was added TFA (21 mL) at 0 °C and the reaction mixture was stirred for 1 h at 23 °C. After removal of the solvent and column chromatography (dichloromethane/MeOH 10:1), **19** (0.98 g, 2.7 mmol, 52 %) was obtained as a white crystalline solid. ^{1}H NMR (400 MHz, D_2O): $\delta=1.44$ (q, $J=9.0$ Hz, 2H), 3.00 (m, 3H), 3.03 (m, 1H), 3.18 (dd, $J=11.6$, 2.5 Hz, 1H), 3.24 (d, $J=4.8$ Hz, 2H), 3.41 (dd, $J=11.6$, 9.2 Hz, 1H), 3.86 (s, 2H), 3.94 (d, $J=7.6$ Hz, 2H), 6.23 ppm (m, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=40.8$ (CH_2), 42.1 (CH_2), 43.6 (CH), 45.6 (CH), 47.0 (CH), 47.1 (CH), 48.8 (CH_2), 48.9 (CH_2), 51.5 (CH_2), 115.4 (C_q), 118.3 (C_q), 135.7 (CH), 136.0 (CH), 163.3 (C_q), 163.6 (C_q), 167.6 (C_q), 167.8 ppm (C_q); IR (ATR): $\tilde{\nu}=3284$, 3090, 3062, 2966, 2936, 2873, 2654, 1677, 1638, 1541, 1464, 1203, 721 cm^{-1}; ESIMS: m/z: calcd for $C_{13}H_{20}O_2N_3$: 250.16; found: 249.9 [M^++H−$C_2O_2F_3$], 271.9 [M^++Na], 192.9 [M^++H−Gly], 136.0 [M^++H−2Gly].

Fmoc-Gly-Gly-Gly-cyclo-olefin (12d): According to the general TBTU method B, TFA salt **19** (150 mg, 0.41 mmol) in DMF (1 mL), TBTU (145.9 mg, 0.45 mmol, 1.1 equiv) and DIPEA (158 μL, 0.91 mmol, 2.2 equiv) were added to a solution of Fmoc-protected glycine (122.7 mg, 0.41 mmol) in DMF (2.5 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (dichloromethane/MeOH 50:1→25:1), **12d** (179.3 mg, 0.34 mmol, 82 %) was obtained as a white crystalline solid. M.p. 103–105 °C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta=1.39$ (d, $J=8.3$ Hz, 1H), 1.53 (d, $J=8.3$ Hz, 1H), 2.75–3.0 (m, 5H), 3.10–3.23 (m, 3H), 3.77 (s, 2H), 3.90 (d, $J=4.8$ Hz, 2H), 3.97 (d, $J=5.2$ Hz, 2H), 4.18 (t, $J=7.0$ Hz, 1H), 4.36 (d, $J=7.0$ Hz, 2H), 5.96 (s, 1H), 6.10 (s, 1H), 7.28 (d, $J=7.5$ Hz, 2H), 7.37 (t, $J=7.5$ Hz, 2H), 7.58 (d, $J=7.5$ Hz, 2H), 7.73 ppm (d, $J=7.5$ Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=42.0$ (CH_2), 42.7 (CH_2), 43.7 (CH), 44.36 (CH_2), 45.8 (CH), 46.6 (CH), 47.0 (CH), 48.0 (CH_2), 48.3 (CH_2), 51.9 (CH_2), 53.4 (CH_2), 67.3 (CH_2), 119.9 (CH), 125.1 (CH), 127.1 (CH), 127.7 (CH), 134.8 (CH), 136.0 (CH), 141.2 (C_q), 143.8 (C_q), 156.9 (C_q), 165.5 (C_q), 169.0 (C_q), 170.0 ppm (C_q); IR (ATR): $\tilde{\nu}=3301$, 3065, 2968, 2942, 2872, 1674, 1642, 1531, 1451, 1203, 741 cm^{-1}; ESIMS: m/z: calcd for $C_{30}H_{33}O_5N_4$: 529.24; found: 529.0 [M^++H], 551.0 [M^++Na], 307.0 [M^++H−Fmoc], 192.9 [M^++H−Fmoc-Gly−Gly], 135.9 [M^++H−FmocGly−2Gly].

Fmoc-Phe-Gly-Gly-cyclo-olefin (12e): According to the general TBTU method B, TFA salt **19** (36.3 mg, 0.1 mmol) in DMF (0.25 mL), TBTU (32.1 mg, 0.1 mmol, 1 equiv) and DIPEA (38.3 μL, 0.22 mmol, 2.2 equiv) were added to a solution of Fmoc-protected phenylalanine (38.7 mg, 0.1 mmol) in DMF (0.4 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (dichloromethane/MeOH 40:1), **12e** (48.3 mg, 0.078 mmol, 78 %) was obtained as a white crystalline solid. $[\alpha]_D^{20}=10.5°$ ($c=2.3$ in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): $\delta=1.43$ (d, $J=8.6$ Hz, 1H), 1.55 (d, $J=8.6$ Hz, 1H), 2.84–3.36 (m, 10H), 3.74–3.91 (m, 3H), 3.92–4.05 (m, 1H), 4.17 (t, $J=6.9$ Hz, 1H), 4.28–4.53 (m, 3H), 5.44 (s, 1H), 6.15 (m, 2H), 6.59 (s, 1H), 6.87 (s, 1H), 7.12–7.31 (m, 7H), 7.39 (t, $J=7.4$ Hz, 2H), 7.52 (t, $J=7.4$ Hz, 2H), 7.75 ppm (d, $J=7.5$ Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=38.5$ (CH_2), 42.1 (CH_2), 42.8 (CH_2), 43.8 (CH), 45.8 (CH), 46.6 (CH), 47.1 (CH), 47.9 (CH_2), 48.3 (CH_2), 51.9 (CH_2), 56.1 (CH), 67.1 (CH_2), 119.9 (CH), 125.1 (CH), 127.0 (CH), 127.1 (CH), 127.7 (CH), 128.7 (CH), 129.3 (CH), 134.8 (CH), 136.0 (CH), 136.5 (C_q), 141.3 (C_q), 143.8 (C_q), 143.8 (C_q), 156.0 (C_q), 165.0 (C_q), 168.1 (C_q), 171.3 ppm (C_q); IR (ATR): $\tilde{\nu}=3297$, 3063, 3028, 2965, 2942, 2870, 1635, 1529, 1451, 1248, 741 cm^{-1}; ESIMS: m/z: calcd for $C_{37}H_{39}O_5N_4$: 619.28; found: 619.0 [M^++H], 641.0 [M^++Na], 393.0 [M^++H−Fmoc], 249.9 [M^++H−FmocPhe], 192.9 [M^++H−FmocPhe−Gly].

Fmoc-Asn-Gly-Gly-cyclo-olefin (12f): According to the general TBTU method B, TFA salt **19** (36.3 mg, 0.1 mmol) in DMF (0.25 mL), TBTU (32.1 mg, 0.1 mmol, 1 equiv) and DIPEA (38.3 μL, 0.22 mmol, 2.2 equiv) were added to a solution of Fmoc-protected asparagine (35.4 mg, 0.1 mmol) in dioxane/DMF 3:1 (1 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (dichloromethane/MeOH 15:1), **12f** (17.8 mg, 0.03 mmol, 30 %) was obtained as a white crystalline solid. $[\alpha]_D^{20}=+9.3°$ ($c=0.9$ in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): $\delta=1.40$ (d, $J=8.3$ Hz, 1H), 1.53 (d, $J=8.3$ Hz, 1H), 2.65 (dd, $J=15.2$, 5.5 Hz, 1H), 2.75–3.0 (m, 5H), 3.09–3.32 (m, 3H), 3.79 (d, $J=3.2$ Hz, 2H), 3.98 (d, $J=4.8$ Hz, 2H), 4.18 (t, $J=7.0$ Hz, 1H), 4.36 (d, $J=7.0$ Hz, 2H), 4.62 (q, $J=5.3$ Hz, 1H), 6.11 (s, 2H), 6.15 (s, 1H), 6.60 (s, 1H), 6.67 (s, 1H), 7.29 (d, $J=7.5$ Hz, 2H), 7.37 (t, $J=7.5$ Hz, 2H), 7.41 (s, 1H), 7.58 (d, $J=7.5$ Hz, 2H), 7.73 ppm (d, $J=7.5$ Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=37.4$ (CH_2), 42.0 (CH_2), 43.1 (CH_2), 43.8 (CH), 45.8 (CH), 46.6 (CH), 46.6 (CH), 47.1 (CH), 48.0 (CH_2), 48.3 (CH_2), 51.9 (CH_2), 53.4 (CH_2), 67.3 (CH_2), 119.9 (CH), 125.2 (CH), 127.1 (CH), 127.7 (CH), 134.9 (CH),

136.0 (CH), 141.2 (C_q), 143.7 (C_q), 143.8 (C_q), 156.3 (C_q), 165.7 (C_q), 169.0 (C_q), 171.7 (C_q), 173.5 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3306, 3063, 2966, 2944, 2872, 1665, 1639, 1531, 1451, 1256, 741 cm^{-1}; ESIMS: *m/z*: calcd for $C_{32}H_{36}O_6N_5$: 586.26; found: 586.0 [M^++H], 608.0 [M^++Na].

Fmoc-Lys(Boc)-Gly-Gly-cyclo-olefin (12g): According to the general TBTU method B, TFA salt **19** (36.3 mg, 0.1 mmol) in DMF (0.2 mL), TBTU (32.1 mg, 0.1 mmol, 1 equiv) and DIPEA (38.3 μL, 0.22 mmol, 2.2 equiv) were added to a solution of Fmoc-protected (Boc)-lysine (46.9 mg, 0.1 mmol) in DMF (0.3 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (dichloromethane/MeOH 50:1→25:1), **12e** (52.8 mg, 0.075 mmol, 75 %) was obtained as a white crystalline solid. M.p. 94–96 °C; $[\alpha]_D^{20}$ = ~8.8° (*c* = 0.89 in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.32–1.52 (m, 14H), 1.55 (d, *J* = 8.5 Hz, 1H), 1.62–1.91 (m, 2H), 2.82–3.01 (m, 5H), 3.02–3.28 (m, 2H), 3.29–3.34 (m, 2H), 3.80 (d, *J* = 3.0 Hz, 2H), 3.97 (m, 2H), 4.19 (m, 2H), 4.38 (m, 2H), 4.88 (s, 1H), 5.67 (s, 1H), 6.13 (m, 2H), 6.98 (s, 1H), 7.05 (s, 1H), 7.39 (t, *J* = 7.5 Hz, 2H), 7.58 (t, *J* = 7.5 Hz, 2H), 7.64 (m, 2H), 7.75 ppm (d, *J* = 7.5 Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 22.5 (CH_2), 28.5 (CH_3), 29.6 (CH_2), 31.9 (CH_2), 39.9 (CH_2), 42.0 (CH_2), 42.7 (CH_2), 43.8 (CH), 45.8 (CH), 46.6 (CH), 47.1 (CH), 47.9 (CH_2), 48.3 (CH_2), 51.9 (CH_2), 53.4 (C_q), 55.0 (CH), 67.1 (CH_2), 79.1 (C_q), 119.9 (CH), 125.1 (CH), 127.1 (CH), 127.7 (CH), 134.8 (CH), 136.0 (CH), 141.3 (C_q), 143.8 (C_q), 143.8 (C_q), 156.2 (C_q), 165.1 (C_q), 168.4 (C_q), 172.2 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3306, 3064, 2969, 2938, 2869, 1702, 1640, 1526, 1451, 1249, 741 cm^{-1}; ESIMS: *m/z*: calcd for $C_{39}H_{49}O_7N_5Na$: 722.36; found: 722.0 [M^++Na], 600.0.

Fmoc-Cys(Trt)-Gly-Gly-cyclo-olefin (12h): According to the general TBTU method B, TFA salt **19** (36.3 mg, 0.1 mmol) in DMF (0.25 mL), TBTU (32.1 mg, 0.1 mmol, 1 equiv) and DIPEA (38.3 μL, 0.22 mmol, 2.2 equiv) were added to a solution of Fmoc-protected (Trt)-cystein (58.6 mg, 0.1 mmol) in dichloromethane (0.4 mL) at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After aqueous workup and column chromatography (dichloromethane/MeOH 40:1), **12e** (54.9 mg, 0.067 mmol, 67 %) was obtained as a white crystalline solid. M.p. 125–127 °C; $[\alpha]_D^{20}$ = +1.5° (*c* = 0.90 in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.42 (d, *J* = 8.5 Hz, 1H), 1.54 (m, 1H), 2.68 (m, 2H), 2.82–2.99 (m, 5H), 3.15–3.30 (m, 3H), 3.74–3.85 (m, 3H), 3.86–3.92 (m, 2H), 4.19 (t, *J* = 6.7 Hz, 1H), 4.38 (d, *J* = 6.7 Hz, 2H), 5.18 (t, *J* = 6.6 Hz, 1H), 6.13 (m, 2H), 6.62 (s, 1H), 6.93 (s, 1H), 7.12–7.21 (m, 3H), 7.21–7.32 (m, 8H), 7.34–7.43 (m, 8H), 7.57 (t, *J* = 7.0 Hz, 2H), 7.74 ppm (t, *J* = 7.0 Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 33.8 (CH_2), 38.6 (CH), 42.0 (CH_2), 42.7 (CH_2), 43.8 (CH), 45.8 (CH), 46.6 (CH), 47.1 (CH), 47.8 (CH_2), 48.2 (CH_2), 51.9 (CH_2), 53.4 (CH_2), 53.4 (CH), 67.1 (C_q), 67.3 (CH_2), 119.9 (CH), 125.1 (CH), 125.2 (CH), 126.9 (CH), 127.1 (CH), 127.7 (CH), 128.1 (CH), 129.6 (CH), 134.8 (CH), 136.0 (CH), 141.2 (C_q), 143.7 (C_q), 143.8 (C_q), 144.3 (C_q), 156.0 (C_q), 165.0 (C_q), 168.0 (C_q), 170.3 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3291, 3058, 3032, 2966, 2942, 2870, 1714, 1632, 1521, 1445, 1246, 741 cm^{-1}; ESIMS: *m/z*: calcd for $C_{50}H_{48}O_5N_4SNa$: 839.34; found: 638.9 [M^++Na].

Fmoc-Cys-Gly-Gly-cyclo-olefin (12i): TFA (21 μL, 0.274 mmol, 5 equiv) was added to a solution of 12 h (45.3 mg, 0.055 mmol) and triethylsilane (44 μL, 0.274 mmol, 5 equiv) in dichloromethane (0.2 mL) at 0 °C. The reaction mixture was stirred for 2 h at 23 °C. After chromatography on silica gel (dichloromethane/MeOH 25:1) and Sephadex, **12i** (28.9 mg, 0.051 mmol, 92 %) was obtained as a white crystalline solid. $[\alpha]_D^{20}$ = 5.2° (*c* = 1.45 in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.40 (d, *J* = 8.5 Hz, 1H), 1.54 (m, 1H), 1.54 (m, 1H), 2.75–3.10 (m, 7H), 3.15–3.35 (m, 3H), 3.82 (d, *J* = 2.6 Hz, 2H), 4.02 (s, 2H), 4.20 (t, *J* = 6.9 Hz, 1H), 4.35–4.55 (m, 3H), 6.10 (m, 2H), 7.19 (s, 1H), 7.29 (m, 2H), 7.39 (t, *J* = 7.5 Hz, 2H), 7.58 (m, 2H), 7.74 ppm (d, *J* = 7.5 Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 27.2 (CH_2), 42.0 (CH_2), 43.0 (CH_2), 43.8 (CH), 45.9 (CH), 46.7 (CH), 47.2 (CH), 48.0 (CH_2), 48.4 (CH_2), 52.0 (CH_2), 53.4 (CH_2), 56.3 (CH), 67.3 (CH_2), 120.0 (CH), 125.2 (CH), 127.2 (CH), 127.8 (CH), 134.9 (CH), 136.1 (CH), 141.4 (C_q), 143.8 (C_q), 143.8 (C_q), 165.2 (C_q), 168.4 (C_q), 170.3 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3298, 3064, 2965, 2935, 2871, 1717, 1636, 1529, 1451, 1248, 742 cm^{-1}; ESIMS: *m/z*: calcd for $C_{31}H_{34}O_5N_4SNa$: 597.22; found: 597.0 [M^++Na].

Fmoc-Leu-Ala-Pro-cyclo-olefin (12k): Leu-Ala-Pro-OH (500 mg, 1.67 mmol) was dissolved in aq Na_2CO_3 and diluted with dioxane (2.5 mL). Under rapid stirring Fmoc-Cl (432 mg, 1.67 mmol) was added in portions at 0 °C. The reaction mixture was stirred for 4 h at 0 °C and additionally for 8 h at 23 °C. The mixture was diluted with water (100 mL) and extracted with *tert*-butyl methyl ether (MTB, 3×17 mL). The combined organic layers were cooled to 0 °C and the pH was adjusted to pH 3–4 by using a concentrated solution of HCl. A solid product precipitated and was separated by filtration. After removal of the solvent under reduced pressure, a product (623 mg, 1.2 mmol, 72 %) was obtained as a white solid. To a solution of this crude product (51.8 mg, 0.38 mmol) in dichloromethane (7 mL) were added amine **11** (200 mg, 0.38 mmol), DCC (95 mg, 0.46 mmol, 1.2 equiv) and DMAP (8 mg, 0.07 mmol, 0.17 equiv) at 0 °C, according to the general DCC method. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. Removing the drying agent and solvent delivered a crude product (1.82 g, 5.2 mmol, 80 %) which was dissolved in dichloromethane (15 mL). To the solution was added TFA (21 mL) at 0 °C and the reaction mixture was stirred for 1 h at 23 °C. After removal of the solvent and column chromatography (dichloromethane/MeOH 80:1→20:1), **12k** (152 mg, 0.24 mmol, 62 %) was obtained as a white solid. $[\alpha]_D^{20}$ = 43.5° (*c* = 0.78 in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 0.92 (d, *J* = 7.0 Hz, 6H), 1.20–2.2 (m, 9H), 2.75–3.4 (m, 8H), 3.65–3.71 (m, 2H), 4.21 (m, 2H), 4.38 (m, 2H), 4.68 (m, 1H), 5.18 (m, 1H), 6.13 (m, 2H), 6.83 (m, 1H), 7.30 (tq, *J* = 7.5, 1.1 Hz, 2H), 7.39 (m, 2H), 7.59 (m, 2H), 7.75 ppm (d, *J* = 7.5 Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 18.0 (CH_3), 18.1 (CH_3), 21.8 (CH_3), 23.1 (CH_3), 24.7 (CH_2), 24.9 (CH_2), 28.0 (C_q), 28.2 (C_q), 42.1 (CH_2), 43.7 (CH), 43.9 (CH), 46.0 (CH), 46.3 (CH), 46.3 (CH), 46.6 (CH), 46.6 (CH), 47.0 (CH), 47.1 (CH_2), 47.2 (CH), 48.3 (CH_2), 48.3 (CH_2), 48.6 (CH_2), 48.8 (CH_2), 51.8 (CH_2), 51.9 (CH_2), 53.4 (CH), 57.8 (CH), 57.9 (CH), 67.0 (CH_2), 119.9 (CH), 125.1 (CH), 125.2 (CH), 127.1 (CH), 127.7 (CH), 134.8 (CH), 135.2 (CH), 135.9 (CH), 136.2 (CH), 141.3 (C_q), 143.7 (C_q), 144.0 (C_q), 156.0 (C_q), 168.8 (C_q), 168.9 (C_q), 170.3 (C_q), 171.3 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3281, 3063, 2962, 2871, 1719, 1633, 1537, 1450, 1245, 741 cm^{-1}; ESIMS: *m/z*: calcd for $C_{38}H_{46}O_5N_4Na$: 661.34; found: 661.0 [M^++Na].

Dansyl-cyclo-olefin (17): A solution of $NaHCO_3$ (58.8 mg, 0.70 mmol, 2 equiv) and **11** (47.3 mg, 0.35 mmol) in water (2.7 mL) were added to a solution of dansylchloride (94.4 mg, 0.35 mmol) in acetonitrile (7 mL). The reaction mixture was stirred for 1.5 h at 23 °C. The solvents were removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1), **17** (75.2 mg, 0.2 mmol, 58 %) was obtained as a light-green solid. ^{1}H NMR (500 MHz, $CDCl_3$): δ = 1.34 (d, *J* = 8.4 Hz, 1H), 1.47 (dt, *J* = 8.4 Hz, *J* = 1.6 Hz, 1H), 2.80 (m, 2H), 2.81–2.95 (m, 13H), 3.27–3.41 (m, 2H), 5.87 (AB-syst., 2H), 7.18 (dd, *J* = 7.2 Hz, 1.0 Hz, 1H), 7.43–7.57 (m, 2H), 8.16 (dd, *J* = 7.2 Hz, 1.0 Hz, 1H), 8.46 (d, *J* = 9.0 Hz, 1H), 8.54 ppm (dd, *J* = 9.0 Hz, 1.0 Hz, 1H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 45.5 (CH_3), 45.7 (CH_3), 46.2 (CH), 49.8 (CH_2), 52.2 (CH_2), 115.2 (CH), 120.4 (CH), 123.2 (CH), 127.7 (CH), 130.0 (CH), 130.2 (C_q), 130.5 (CH), 130.7 (C_q), 133.8 (C_q), 135.5 (CH), 151.5 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3058, 2962, 2940, 2942, 2867, 2832, 2788, 1588, 1573, 1519, 1477, 1454, 1334, 1320, 1162, 1145, 793 cm^{-1}; MS (EI): *m/z* (%): 368 (16), 171 (100), 170 (24), 154 (6), 69 (7); HRMS: calcd for $C_{21}H_{24}N_2O_2S$: 368.1559; found: 368.1561 [M^+].

Cross-partners for ROCM

Fmoc-Gly-hexenol (13a): According to the general DCC method, 5-hexen-1-ol (117.8 μL, 0.93 mmol; 1.11 equiv), DCC (208 mg, 1 mmol, 1.2 equiv) and DMAP (10.3 mg, 0.08 mmol, 0.1 equiv) were added successively to a solution of Fmoc-protected glycine (250 mg, 0.84 mmol) in THF (3 mL) at 0 °C. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. After filtration, aqueous workup and column chromatography (hexanes/ethyl acetate 5:1), **13a** (255 mg, 0.69 mmol, 80 %) was obtained as a white waxlike solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.46 (m, 2H), 1.67 (m, 2H), 2.08 (tq, *J* = 7.0, 1.3 Hz, 2H), 4.00 (d, *J* = 5.5 Hz, 2H), 4.18 (t, *J* = 6.7 Hz, 2H), 4.24 (t, *J* = 7.0 Hz, 1H), 4.40 (d, *J* = 7.0 Hz, 2H), 4.93–5.06 (m, 2H), 5.28 (t, *J* = 4.5 Hz, 1H), 5.78 (m, 1H), 7.32 (dt, *J* = 7.5, 1.2 Hz, 2H), 7.41 (t, *J* = 7.5 Hz, 2H), 7.60 (d, *J* = 7.5 Hz, 2H), 7.76 ppm (d, *J* = 7.5 Hz, 2H);

^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 25.0 (CH_2), 27.9 (CH_2), 33.1 (CH_2), 42.8 (CH_2), 47.1 (CH), 65.5 (CH_2), 67.2 (CH_2), 115.0 (CH_2), 120.0 (CH), 125.1 (CH), 127.1 (CH), 127.7 (CH), 138.1 (CH), 141.3 (C_q), 143.8 (C_q), 156.2 (C_q), 170.1 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3417, 3339, 3066, 3041, 3018, 2937, 2858, 1749, 1727, 1532, 1451, 1195, 1052, 741 cm^{-1}; LRMS (70 eV, EI): *m*/*z* (%): 379 (100), 252 (20), 250 (5), 178 (>100); HRMS: calcd for $C_{23}H_{25}O_4N$: 379.1784; found: 379.1789 [M^+].

Fmoc-Leu-Ala-Pro-hexenol (13b): According to the general DCC method, 5-hexen-1-ol (50.8 µL, 0.38 mmol; 1.11 equiv), DCC (158.2 mg, 0.76 mmol, 2 equiv) and DMAP (7.9 mg, 0.07 mmol, 0.17 equiv) were added successively to a solution of Fmoc-Leu-Ala-Pro-OH (200 mg, 0.38 mmol) in dichloromethane (2 mL) at 0 °C. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. After filtration, aqueous workup and column chromatography (hexanes/ethyl acetate 2:1), **13b** (117 mg, 0.19 mmol, 50%) was obtained as a white waxlike solid. $[\alpha]_D^{20}$ = ~58.1° (*c* = 1.1 in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 0.92 (d, *J* = 5.6 Hz, 6H), 1.20–1.70 (m, 10H), 1.92–2.12 (m, 5H), 2.18–2.29 (m, 1H), 3.58–3.71 (m, 2H), 4.03–4.17 (m, 2H), 4.18–4.26 (m, 2H), 4.33–4.42 (m, 2H), 4.51 (m, 1H), 4.70 (m, 1H), 4.91–5.03 (m, 2H), 5.21 (d, *J* = 7.0 Hz, 1H), 5.77 (m, 1H), 6.81 (d, *J* = 6.3 Hz, 1H), 7.33 (t, *J* = 7.5 Hz, 2H), 7.39 (t, *J* = 7.5 Hz, 2H), 7.59 (m, 2H), 7.76 ppm (d, *J* = 7.5 Hz, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 18.0 (CH_3), 21.8 (CH_3), 23.1 (CH_3), 24.7 (CH), 24.9 (CH_2), 25.1 (CH_2), 28.0 (CH_2), 29.0 (CH_2), 33.2 (CH_2), 42.1 (CH_2), 46.8 (CH_2), 46.9 (CH), 53.5 (CH), 58.9 (CH), 65.2 (CH_2), 67.0 (CH_2), 114.9 (CH_2), 119.9 (CH), 120.0 (CH), 125.1 (CH), 127.1 (CH), 127.7 (CH), 138.2 (CH), 141.3 (C_q), 143.7 (C_q), 143.9 (C_q), 156.0 (C_q), 170.7 (C_q), 171.4 (C_q), 171.8 ppm (C_q); IR (ATR) $\tilde{\nu}$ = 3282, 3068, 2955, 2935, 2870, 1744, 1719, 1638, 1541, 1451, 1241, 741 cm^{-1}; ESIMS: *m*/*z*: calcd for $C_{35}H_{45}O_6N_3Na$: 626.32; found: 626.0 [M^++Na].

Fmoc-Cys(Trt)-Gly-Gly-hexenol (13c): Aqueous NaOH (10 mL, 1 N) and $(Boc)_2O$ (2.4 g, 11 mmol, 1.1 equiv) were added to a solution of GlyGly (1.32 g, 10 mmol) in dioxane and water (2:1, 30 mL) at 0 °C. The reaction mixture was stirred for 1 h at 23 °C. The dioxane was removed under reduced pressure. The aqueous layer was diluted with ethyl acetate (20 mL) and the pH was adjusted to pH 2–3 by using $KHSO_4$. The layers were separated and the aqueous layer was extracted with ethyl acetate (5 × 20 mL). The combined organic layers were washed with water (50 mL) and dried over Na_2SO_4. Removing the drying agent by filtration and the solvent under reduced pressure delivered a crude product (1.5 g, 6.5 mmol, 65%). 5-Hexen-1-ol (1 mL, 8.45 mmol, 1.3 equiv), DCC (1.61 g, 7.8 mmol, 1.2 equiv) in dichloromethane (10 mL) and DMAP (140 mg, 1.1 mmol, 0.17 equiv) were added to a solution of the crude product in THF (26 mL) at 0 °C, according to the general DCC method. The reaction mixture was stirred for 2 h at 0 °C and additionally for 12 h at 23 °C. Removal of the drying agent and solvent delivered a crude product (1.92 g, 6.1 mmol, 72%). A portion of the crude product (650 mg, 2.07 mmol) was dissolved in dichloromethane (6 mL). To this solution was added TFA (8 mL) at 0 °C and the reaction mixture was stirred for 15 min at 0 °C and additionally for 45 min at 23 °C. After removing of the volatile compounds and column chromatography (dichloromethane/MeOH 25:1→10:1), **20** (0.66 g, 2 mmol, 97%) was obtained as a yellow oil. A portion of this oil (200 mg, 0.61 mmol) was dissolved in dichloromethane and DMF (2:1, 3 mL). Fmoc-Cys(Trt) (356.7 mg, 0.61 mmol), TBTU (234.7 mg, 0.73 mmol, 1.2 equiv) and DIPEA (234 µL, 1.34 mmol, 2.2 equiv) were added to this solution at 0 °C. The reaction mixture was stirred for 0.5 h at 0 °C and additionally for 1 h at 23 °C. After filtration, aqueous workup and column chromatography (hexanes/ethyl acetate 1:1), **13c** (248 mg, 0.31 mmol, 50%) was obtained as a white crystalline solid. $[\alpha]_D^{20}$ = +2.3° (*c* = 1.0 in CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.42 (m, 2H), 1.60 (m, 2H), 2.06 (tq, *J* = 6.4, 1.3 Hz, 2H), 2.64–2.73 (m, 2H), 3.65 (q, *J* = 6.4 Hz, 1H), 3.80–3.99 (m, 4H), 4.07 (t, *J* = 6.7 Hz, 2H), 4.18 (t, *J* = Hz, 1H), 4.41 (m, 2H), 4.91–5.03 (m, 2H), 5.77 (m, 1H), 6.36 (t, *J* = 5.8 Hz, 1H), 6.82 (m, 1H), 7.20 (tt, *J* = 7.2, 1.3 Hz, 3H), 7.23–7.32 (m, 8H), 7.39 (m, 8H), 7.57 (m, 2H), 7.75 ppm (m, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 25.0 (CH_2), 27.9 (CH_2), 33.2 (CH_2), 33.4 (CH_2), 38.6 (CH), 40.9 (CH_2), 43.0 (CH_2), 47.1 (CH), 53.4 (C_q), 54.3 (CH), 65.4 (CH_2), 67.0 (C_q), 67.5 (CH_2), 115.0 (CH_2), 120.0 (CH), 124.9 (CH), 125.0 (CH), 127.1 (CH), 127.8 (CH), 127.8 (CH), 128.1 (CH), 129.5 (CH), 138.2 (CH), 141.3 (C_q), 143.6 (C_q), 144.2 (C_q), 156.2 (C_q), 168.6 (C_q), 169.5 (C_q), 170.4 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3306, 3062, 3032, 3019, 2973, 2935, 2859, 1701, 1648, 1529, 1446, 1245, 1207, 741 cm^{-1}; ESI-MS: *m*/*z*: calcd for $C_{47}H_{47}O_6N_3SNa$: 804.32; found: 803.9 [M^++Na].

(*N*-Boc)-allylglycine (15): Thionylchloride (0.83 mL, 11.3 mmol, 1.3 equiv) was added dropwise to a suspension of allylglycine (1 g, 8.69 mmol) in methanol (80 mL) at 0 °C. The reaction mixture was stirred for 12 h at 23 °C. After removal of the volatile compounds under reduced pressure, the crude product was dissolved in acetonitrile (33 mL). DMAP (0.2 g, 1.74 mmol, 0.2 equiv), $(Boc)_2O$ (2.01 g, 9.56 mmol, 1.1 equiv) and NEt_3 (1.5 mL) were added to this solution. The reaction mixture was stirred for 2 h at 23 °C and was then washed sequentially with $NaHCO_3$ solution, water and NaCl solution. After removal of the solvent under vacuum and silica gel chromatography (hexanes/MTB 6:1), **15** (0.62 g, 2.6 mmol, 30%) was obtained as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ = 1.43 (s, 9H), 2.50 (m, 2H), 3.79 (s, 3H), 4.37 (q, *J* = 7.4 Hz, 1H), 5.02 (d, *J* = 7.4 Hz, 1H), 5.16 (m, 2H), 5.67 ppm (m, 1H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 28.3 (CH_3), 28.5 (CH_3), 36.8 (CH_2), 52.2 (CH_3), 52.9 (CH), 79.9 (C_q), 119.1 (CH_2), 132.3 (CH), 155.2 (C_q), 172.6 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3439, 3359, 3080, 3005, 2979, 2955, 2933, 2848, 1747, 1716, 1504, 1438, 1367, 1163 cm^{-1}; LRMS (70 eV, EI): *m*/*z* (%): 189 (80), 148 (74), 146 (32), 81 (47), 69 (100), 57 (71); HRMS: calcd for $C_8H_{15}O_4N$: 189.1001; found: 189.1001 [M^+−C_3H_4].

ROCMs

General procedure for ROCM: The cyclic olefin, the cross-partner for metathesis (1 equiv) and the Ru catalyst (1 to 5 mol%) were placed in a screw-top HPLC vial (1.5 mL) and dissolved in dichloromethane (or 1,2-dichlorethane, DCE) to get a 0.1 M solution. The reaction mixture was stirred for the mentioned time at 40 °C (70 °C, respectively). The conversion was monitored by HPLC. The solvents were removed under vacuum and the crude product was purified by column chromatography and, if noted, additionally by Sephadex chromatography.

Metathesis product (14a): According to the general procedure for ROCM, **12a** (10.36 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.3 mg, 0.5 µmol, 2 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40 °C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14a** (15.88 mg, 0.02 mmol, 80%) was obtained as a white crystalline solid. M.p. 65–67 °C; IR (ATR): $\tilde{\nu}$ = 3293, 3065, 3039, 2953, 2934, 2872, 1721, 1639, 1534, 1451, 1244, 742 cm^{-1}; ESIMS: *m*/*z*: calcd for $C_{49}H_{52}O_7N_3$: 794.37; found: 794.2 [M^++H], 816.2 [M^++Na], 572.2 [M^++H−Fmoc], 515.2 [M^++H−FmocGly], 293.2 [M^++H−FmocGly−Fmoc], 350.2 [M^++H−2Fmoc].

Metathesis product (14b): According to the general procedure for ROCM, **12b** (12.62 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.3 mg, 0.5 µmol, 2 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40 °C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14b** (16.3 mg, 0.019 mmol, 74%) was obtained as a white crystalline solid. $[\alpha]_D^{20}$ = 12.0° (*c* = 0.82 in CH_2Cl_2); IR (ATR): $\tilde{\nu}$ = 3315, 3065, 3039, 2927, 2855, 1717, 1631, 1527, 1451, 1248, 741 cm^{-1}; ESIMS: *m*/*z*: calcd for $C_{56}H_{58}O_7N_3$: 884.42; found: 884.3 [M^++H], 906.3 [M^++Na], 662.3 [M^++H−Fmoc], 515.2 [M^++H−FmocPhe], 293.2 [M^++H−FmocPhe−Fmoc], 440.2 [M^++H−2Fmoc].

Metathesis product (14c): According to the general procedure for ROCM, **12c** (11.42 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.3 mg, 0.5 µmol, 2 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40 °C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14c** (17.2 mg, 0.021 mmol, 82%) was obtained as a white crystalline solid. $[\alpha]_D^{20}$ = ~4.2° (*c* = 0.86 in CH_2Cl_2); IR (ATR): $\tilde{\nu}$ = 3321, 3066, 3040, 2958, 2935, 2872, 1719, 1630, 1527, 1450, 1196, 741 cm^{-1}; ESIMS: *m*/*z*: calcd for $C_{52}H_{58}O_7N_3$: 836.42; found: 836.3 [M^++H], 858.3 [M^++Na], 614.3 [M^++H-Fmoc], 515.2 [M^++H-Fmoc-Val], 293.2 [M^++H−FmocPhe−Fmoc], 640.3 [M^++Na−Fmoc].

Metathesis product (14d): According to the general procedure for ROCM, **12d** (13.2 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.15 mg, 0.25 µmol, 1 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40 °C. The solvent was removed under

reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14d** (17.0 mg, 0.019 mmol, 75%) was obtained as a white crystalline solid. M.p. 95–97°C; $[\alpha]_D^{20}=\sim 0.1°$ ($c=0.75$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3313, 3066, 3041, 3017, 2937, 2891, 1724, 1642, 1529, 1451, 1250, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{53}H_{58}O_9N_5$: 908.42; found: 908.2 [M^++H], 930.2 [M^++Na], 686.2 [M^++H−Fmoc], 572.2 [M^++H−FmocGly−Gly], 515.2 [M^++H−FmocGly−2Gly], 293.2 [M^++H−FmocGly−2Gly−Fmoc], 708.2 [M^++Na−Fmoc], 684.2 [M^++Na−2Fmoc].

Metathesis product (14e): According to the general procedure for ROCM, **12e** (15.5 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.15 mg, 0.25 μmol, 1 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14e** (20.7 mg, 0.021 mmol, 83%) was obtained as a white crystalline solid. M.p. 75°C; $[\alpha]_D^{20}=\sim 6.3°$ ($c=1.0$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3303, 3065, 3039, 3000, 2941, 2893, 1723, 1640, 1528, 1451, 1250, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{60}H_{64}O_9N_5$: 998.47; found: 998.2 [M^++H], 1020.2 [M^++Na], 572.2 [M^++H−FmocPhe−Gly], 515.2 [M^++H−FmocPhe−2Gly], 293.2 [M^++H−FmocPhe−2Gly−Fmoc], 798.2 [M^++Na−Fmoc].

Metathesis product (14f): According to the general procedure for ROCM, **12f** (14.6 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.15 mg, 0.25 μmol, 1 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14f** (8.9 mg, 0.009 mmol, 37%) was obtained as a white crystalline solid. M.p. 87–89°C); $[\alpha]_D^{20}=2.9°$ ($c=0.45$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3321, 3065, 3041, 3017, 2933, 2855, 1720, 1669, 1643, 1531, 1451, 1252, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{55}H_{61}O_{10}N_6$: 965.44; found: 965.2 [M^++H], 987.2 [M^++Na], 629.3 [M^++H−FmocAsn], 572.2 [M^++H−FmocAsn−Gly], 515.2 [M^++H−FmocPhe−2Gly], 765.2 [M^++Na−Fmoc].

Metathesis product (14g): According to the general procedure for ROCM, **12g** (17.5 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.75 mg, 1.25 μmol, 5 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14g** (21.6 mg, 0.02 mmol, 80%) was obtained as a white crystalline solid. M.p. 63–65°C; $[\alpha]_D^{20}=\sim 4.7°$ ($c=1.1$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3316, 3066, 3040, 2972, 2937, 2864, 1710, 1642, 1524, 1451, 1270, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{62}H_{76}O_{11}N_6$: 1080.55; found: 1079.2 [M^++H], 1101.2 [M^++Na], 979.3 [M^++H−Boc], 757.2 [M^++H−Boc−Fmoc], 572.2 [M^++H−Boc−FmocLys−Gly], 515.2 [M^++H−Boc−FmocLys−2Gly], 293.2 [M^++H−Boc−FmocLys−Gly−2Fmoc], 779.2 [M^++Na−Boc−Fmoc].

Metathesis product (14h): According to the general procedure for ROCM, **12h** (20.4 mg, 0.025 mmol), **13a** (9.49 mg, 0.025 mmol) and [Ru-**3**] (0.15 mg, 0.25 μmol, 1 mol%) were dissolved in dichloromethane (0.25 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14h** (24.7 mg, 0.021 mmol, 83%) was obtained as a white crystalline solid. M.p. 84–86°C; $[\alpha]_D^{20}=\sim 0.2°$ ($c=1.2$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3301, 3063, 3018, 2942, 2895, 1723, 1638, 1523, 1450, 1249, 742\ cm^{-1}$; ESIMS: m/z: calcd for $C_{73}H_{73}O_9N_5SNa$: 1218.50; found: 1218.2 [M^++Na], 975.1 [M^++Na−Trt], 753.1 [M^++Na−Trt−Fmoc].

Metathesis product (14i): To a solution of **12i** (50 mg, 0.087 mmol) in DCE (7.25 mL) was added **13a** (66 mg, 0.147 mmol, 2 equiv) and [Ru-**4**] (2.7 mg, 4.35 μmol, 5 mol%) in a thoroughly dried flask, and the resulting mixture was stirred for 1 h at 70°C under a nitrogen atmosphere. After this time, the solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 100:1→25:1) and Sephadex, **14i** (24.9 mg, 0.026 mmol, 30%) was obtained as a white crystalline solid. M.p. 96–99°C; $[\alpha]_D^{20}=\sim 3.9°$ ($c=0.9$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3308, 3067, 3041, 2940, 2893, 1721, 1640, 1527, 1451, 1248, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{54}H_{59}O_9N_5SNa$: 976.39; found: 976.2 [M^++Na], 754.2 [M^++Na−Fmoc], 515.2 [M^++H−FmocCys−2Gly], 293.2 [M^++H−FmocCys−2Gly−Fmoc].

Metathesis product (14k): According to the general procedure for ROCM, **12e** (9.3 mg, 0.015 mmol), **13b** (9.1 mg, 0.015 mmol) and [Ru-**3**] (0.18 mg, 0.3 μmol, 2 mol%) were dissolved in dichloromethane (0.15 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14k** (15.1 mg, 0.012 mmol, 82%) was obtained as a white crystalline solid. M.p. 91–93°C; $[\alpha]_D^{20}=\sim 31.2°$ ($c=0.76$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3293, 3066, 2953, 2932, 2871, 1720, 1639, 1534, 1451, 1246), 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{72}H_{83}O_{11}N_7Na$: 1244.61; found: 1244.3 [M^++Na], 1022.3 [M^++Na−Fmoc], 816.2 [M^++H−FmocLeuAla], 390.2 [M^++H−FmocLeuAla−FmocPhe−Gly], 333.1 [M^++H−FmocLeuAla−FmocPhe−2Gly].

Metathesis product (14l): According to the general procedure for ROCM, **12k** (9.6 mg, 0.015 mmol), **13c** (11.7 mg, 0.015 mmol) and [Ru-**3**] (0.18 mg, 0.3 μmol, 2 mol%) were dissolved in dichloromethane (0.15 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14l** (14 mg, 0.01 mmol, 66%) was obtained as a white crystalline solid. M.p. 105–106°C; $[\alpha]_D^{20}=\sim 14.0°$ ($c=0.7$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3294, 3064, 3018, 2953, 2935, 2870, 1720, 1653, 1637, 1532, 1449, 1246, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{85}H_{93}O_{11}N_7SNa$: 1442.66; found: 1442.3 [M^++Na], 1199.2 [M^++Na−Trt], 1199.2 [M^++H−FmocLeuAla], 977.2 [M^++Na−Trt−Fmoc], 772.2 [M^++H−FmocLeuAla−Trt].

Metathesis product (14m): According to the general procedure for ROCM, **12k** (9.6 mg, 0.015 mmol), **13b** (9.1 mg, 0.015 mmol) and [Ru-**3**] (0.18 mg, 0.3 μmol, 2 mol%) were dissolved in dichloromethane (0.15 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **14m** (13.9 mg, 0.011 mmol, 74%) was obtained as a white crystalline solid. M.p. 110–112°C; $[\alpha]_D^{20}=\sim 45.2°$ ($c=0.7$ in CH_2Cl_2); IR (ATR): $\tilde{\nu}=3287, 3066, 2954, 2871, 1721, 1639, 1537, 1451, 1243, 741\ cm^{-1}$; ESIMS: m/z: calcd for $C_{73}H_{91}O_{11}N_7Na$: 1264.67; found: 1264.3 [M^++Na], 1042.3 [M^++Na−Fmoc], 836.3 [M^++H-FmocLeuAla], 739.2 [M^++H-FmocLeuAlaPro], 430.2 [M^++H-FmocLeuAla-FmocLeuAla], 333.1 [M^++H-FmocLeuAlaPro-FmocLeuAla].

Metathesis product (16b): According to the general procedure for ROCM, **12d** (26.4 mg, 0.05 mmol), **15** (22.9 mg, 0.1 mmol, 2 equiv) and [Ru-**4**] (0.03 mg, 0.5 μmol, 1 mol%) were dissolved in dichloromethane (0.15 mL) and stirred for 17 h at 40°C. The solvent was removed under reduced pressure. After chromatography on silica gel (dichloromethane/MeOH 50:1→25:1) and Sephadex, **16b** (12.5 mg, 0.017 mmol, 33%) was obtained as a white crystalline solid. IR (ATR): $\tilde{\nu}=3309, 3069, 2974, 2950, 2884, 1709, 1642, 1525, 1451, 1249, 742\ cm^{-1}$; ESIMS: m/z: calcd for $C_{41}H_{51}O_9N_5Na$: 780.36; found: 780.1 [M^++Na], 724.1 [M^++Na−*t*Bu], 680.1 [M^++Na−Boc], 658.2 [M^++H−Boc], 458.1 [M^++Na−Boc−Fmoc], 436.2 [M^++H−Boc−Fmoc], 322.1 [M^++H−Boc−FmocGly−Gly], 265.1 [M^++H−Boc−FmocGly−2Gly].

Metathesis product (18a): In a thoroughly dried flask, **13d** (19.3 μg, 22 μmol) and a solution of [Ru-**2**] (0.19 mg, 0.22 μmol, 1 mol%) in dichloromethane (0.1 mL) were added to a solution of **17** (8.1 mg, 22 μmol) in dichloromethane (0.4 mL) and stirred for 0.5 h at 40°C under a nitrogen atmosphere. The solvent was removed under reduced pressure. After preparative TLC (dichloromethane/MeOH 40:1), **18a** (11.6 mg, 9.5 μmol, 43%) was obtained as a white crystalline solid. ^{1}H NMR (500 MHz, $CDCl_3$): $\delta=0.75–0.95$ (m, 6H), 1.27–1.50 (m, 1H), 1.50–1.70 (m, 3H), 1.90–2.10 (m, 2H), 2.27–2.40 (m, 1H), 2.50–2.65 (m, 1H), 2.65–2.7 (m, 3H), 2.85–2.90 (m, 7H), 2.95–3.00 (m, 1H), 3.00–3.25 (m, 4H), 3.35–3.45 (m, 1H), 3.70 (s, 3H), 3.75–3.95 (m, 2H), 4.05–4.15 (m, 2H), 4.25–4.35 (m, 1H), 4.85–5.05 (m, 2H), 5.10–5.25 (m, 1H), 5.25–5.35 (m, 1H), 5.60–5.85 (m, 1H), 6.55–6.70 (m, 2H), 6.90–6.95 (m, 1H), 7.10–7.40 (m, 28H), 7.50–7.60 (m, 2H), 8.20–8.25 (m, 1H), 8.40–8.60 ppm (m, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): $\delta=14.2$ (CH_1), 18.1 (CH_3), 19.2 (CH), 25.9 (CH_2), 28.6 (CH_2), 29.8 (CH_2), 30.6 (CH), 32.1 (CH_2), 33.0 (CH_2), 35.0 (CH_2), 36.9 (CH_2), 45.5 (CH_3), 49.4 (CH_2), 49.5 (CH_2), 54.1 (CH), 55.2 (CH_3), 55.3 (CH), 56.4 (CH), 62.4 (CH_2), 65.3 (CH_2), 66.4 (C_q), 71.3 (C_q), 113.3

(CH), 115.0 (CH_2), 115.2 (CH), 115.6 (CH_2), 115.7 (CH_2), 120.2 (CH), 120.2 (CH), 123.2 (CH), 126.7 (CH), 126.8 (CH), 127.2 (CH), 127.3 (CH), 127.9 (CH), 127.9 (CH), 128.0 (CH), 128.0 (CH), 128.0 (CH), 129.0 (CH), 129.3 (CH), 129.4 (CH), 129.5 (CH), 130.6 (CH), 131.8 (CH), 144.9 (C_q), 145.8 (C_q), 146.9 (C_q), 147.2 (C_q), 158.2 (C_q), 170.5 (C_q), 171.7 (C_q), 174.0 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3493, 3343, 3083, 3057, 3031, 2958, 2931, 2871, 2849, 2791, 1738, 1653, 1607, 1588, 1575, 1508, 1251, 1182, 1161, 1144, 1033, 744, 706 cm^{-1}; FABMS: *m/z* (%) = [norm 1245]: 1245 (100) $[M]^+$, 970 (55), [norm 243]: 273 (90), 243 (100), 147 (23), 73 (46).

Metathesis product (18b): In a thoroughly dried flask, **13e** (19.9 μg, 22 μmol) and a solution of [Ru-**2**] (0.19 mg, 0.22 μmol, 1 mol%) in dichloromethane (0.1 mL) were added to a solution of **17** (8.1 mg, 22 μmmol) in dichloromethane (0.4 mL) and stirred for 0.5 h at 40 °C under nitrogen atmosphere. The solvent was removed under reduced pressure. After preparative TLC (dichloromethane/MeOH 40:1), **18b** (18.8 mg, 14.7 μmol, 67%) was obtained as a white crystalline solid. ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.75–0.95 (m, 6H), 1.27–1.5 (m, 3H), 1.50–1.70 (m, 5H), 1.85–1.90 (m, 1H), 1.90–2.10 (m, 2H), 2.27–2.40 (m, 1H), 2.50–2.65 (m, 1H), 2.65–2.7 (m, 3H), 2.85–2.90 (m, 7H), 2.95–3.00 (m, 1H), 3.00–3.25 (m, 4H), 3.35–3.45 (m, 1H), 3.70 (s, 3H), 3.75–3.95 (m, 2H), 4.05–4.15 (m, 2H), 4.25–4.35 (m, 1H), 4.85–5.05 (m, 2H), 5.10–5.25 (m, 1H), 5.25–5.35 (m, 1H), 5.60–5.85 (m, 1H), 6.55–6.70 (m, 2H), 6.90–6.95 (m, 1H), 7.10–7.4 (m, 28H), 7.50–7.60 (m, 2H), 8.20–8.25 (m, 1H), 8.40–8.60 ppm (m, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 18.1 (CH_3), 19.2 (CH), 25.2 (CH_2), 25.7 (CH_2), 25.9 (CH_2), 27.3 (CH_2), 28.0 (CH_2), 28.1 (CH_2), 28.2 (CH_2), 29.8 (CH_2), 30.6 (CH), 32.1 (CH_2), 33.3 (CH_2), 35.0 (CH_2), 36.5 (CH_2), 36.9 (CH_2), 39.4 (CH), 44.7 (CH), 45.5 (CH_3), 45.8 (CH), 46.0 (CH), 46.1 (CH), 46.3 (CH), 46.5 (CH), 46.7 (CH), 49.4 (CH_2), 49.5 (CH_2), 54.1 (CH), 55.2 (CH_3), 56.3 (CH), 57.8 (CH), 62.4 (CH_2), 65.3 (CH_2), 66.4 (C_q), 71.3 (C_q), 113.3 (CH), 115.0 (CH_2), 115.2 (CH), 115.6 (CH_2), 115.7 (CH_2), 120.2 (CH), 120.2 (CH), 123.2 (CH), 126.7 (CH), 126.8 (CH), 127.9 (CH), 127.9 (CH), 128.0 (CH), 128.1 (CH), 128.1 (CH), 129.0 (CH), 129.1 (CH), 129.4 (CH), 129.5 (CH), 130.0 (CH), 130.3 (CH), 130.5 (CH), 130.6 (CH), 130.8 (CH), 130.8 (CH), 130.9 (CH), 131.0 (CH), 133.0 (C_q), 136.6 (C_q), 138.0 (CH), 138.1 (CH), 138.3 (CH), 144.7 (C_q), 144.9 (C_q), 145.6 (C_q), 151.7 (C_q), 158.2 (C_q), 170.5 (C_q), 171.7 (C_q), 174.0 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3493, 33.43, 3083, 3057, 3031, 2958, 2931, 2871, 2849, 2791, 1738, 1653, 1607, 1588, 1575, 1508, 1251, 1182, 1161, 1144, 1033, 744, 706 cm^{-1}; FABMS: *m/z* (%) = [norm 1273]: 1273 $[M]^+$ (100), 998 (40), 952 (10), 905 (18), [norm 243]: 273 (90), 243 (100), 154 (10).

Metathesis product (18c): In a thoroughly dried flask, **13f** (21.4 μg, 22 μmol) and a solution of [Ru-**2**] (0.19 mg, 0.22 μmol, 1 mol%) in dichloromethane (0.1 mL) were added to a solution of **17** (8.1 mg, 22 μmmol) in dichloromethane (0.4 mL) and stirred for 0.5 h at 40 °C under nitrogen atmosphere. The solvent was removed under reduced pressure. After preparative TLC (dichloromethane/MeOH 40:1), **18c** (15.3 mg, 10.8 μmol, 49%) was obtained as a white crystalline solid. ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.75–0.95 (m, 6H), 1.27–1.50 (m, 12H), 1.50–1.70 (m, 7H), 1.85–1.90 (m, 1H), 1.90–2.10 (m, 2H), 2.27–2.40 (m, 1H), 2.50–2.65 (m, 1H), 2.65–2.70 (m, 3H), 2.85–2.90 (m, 7H), 2.95–3.00 (m, 1H), 3.00–3.25 (m, 4H), 3.35–3.45 (m, 1H), 3.70 (s, 3H), 3.75–3.95 (m, 2H), 4.05–4.15 (m, 2H), 4.25–4.35 (m, 1H), 4.85–5.05 (m, 2H), 5.10–5.25 (m, 1H), 5.25–5.35 (m, 1H), 5.60–5.85 (m, 1H), 6.55–6.70 (m, 2H), 6.90–6.95 (m, 1H), 7.10–7.40 (m, 28H), 7.50–7.60 (m, 2H), 8.20–8.25 (m, 1H), 8.40–8.60 ppm (m, 2H); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ = 18.1 (CH_3), 19.2 (CH), 25.9 (CH_2), 28.6 (CH_2), 29.2 (CH_2), 29.5 (CH_2), 29.8 (CH), 30.6 (CH), 32.1 (CH_2), 33.3 (CH_2), 35.0 (CH_2), 36.9 (CH_2), 39.4 (CH), 44.7 (CH_2), 45.5 (CH_3), 45.8 (CH), 46.1 (CH), 46.3 (CH), 46.7 (CH), 49.4 (CH_2), 49.5 (CH_2), 54.1 (CH), 55.2 (CH_3), 56.3 (CH), 56.3 (CH), 57.8 (CH), 62.4 (CH_2), 65.3 (CH_2), 66.4 (C_q), 71.3 (C_q), 113.3 (CH), 115.0 (CH_2), 115.2 (CH), 115.6 (CH_2), 115.7 (CH_2), 120.2 (CH), 120.2 (CH), 123.2 (CH), 126.6 (CH), 126.8 (CH), 127.2 (CH), 127.3 (CH), 127.8 (CH), 127.9 (CH), 128.0 (CH), 128.1 (CH), 129.0 (CH), 129.3 (CH), 129.4 (CH), 129.5 (CH), 130.6 (CH), 131.0 (CH), 131.0 (CH), 144.9 (C_q), 145.6 (C_q), 146.9 (C_q), 158.2 (C_q), 170.5 (C_q), 171.7 (C_q), 174.0 ppm (C_q); IR (ATR): $\tilde{\nu}$ = 3493, 33.43, 3083, 3057, 3031, 2958, 2931, 2871, 2849, 2791, 1738, 1653, 1607, 1588, 1575, 1508, 1251, 1182, 1161, 1144, 1033, 744, 706 cm^{-1}; FABMS: *m/z* (%) = [norm 1344]: 1344 $[M]^+$ (100), 1069 (60), 997 (23), [norm 243]: 273 (82), 243 (100), 55 (20).

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[1] J. P. Tam, Y. Qitao, M. Zhenwei, *Biopolymers* **1999**, *51*, 311; J. P. Tam, X. Jiaxi, K. D. Eom, *Biopolymers* **2001**, *60*, 194; P. E. Dawson, S. B. H. Kent, *Annu. Rev. Biochem.* **2000**, *69*, 923; G. T. Hermanson, *BioconjugateTechniques*, AcademicPress, SanDiego **1996**; J. W. Bode, R. M. Fox, K. D. Baucom, *Angew. Chem.* **2006**, *118*, 1270; *Angew. Chem. Int. Ed.* **2006**, *45*, 1248.

[2] R. M. Freidinger, D. F. Veber, D. S. Perlow, B. J. R. , R. Sapersetin, *Science* **1980**, *210*, 656; T. Higashijima, T. Miyazawa, M. Kawai, U. Nagai, *Biopolymers* **1986**, *25*, 2295.

[3] K. C. Nicolaou, P. G. Bulger, D. Sarlah, *Angew. Chem.* **2005**, *117*, 4564; *Angew. Chem. Int. Ed.* **2005**, *44*, 4490; R. H. Grubbs, *Tetrahedron* **2004**, *60*, 7117; R. H. Grubbs, *Handbook of Metathesis*, Wiley-VCH, Weinheim, **2004**; S. J. Connon, S. Blechert, *Angew. Chem.* **2003**, *115*, 1944; *Angew. Chem. Int. Ed.* **2003**, *42*, 1900; A. Fürstner, *Angew. Chem.* **2000**, *112*, 3012; *Angew. Chem. Int. Ed.* **2000**, *39*, 3140.

[4] H. C. Kolb, K. B. Sharpless, *Drug Discovery Today* **2004**, *9*, 1128.

[5] A. D. d. Araujo, J. M. Palomo, J. Cramer, M. Köhn, H. Schröder, R. Wacker, C. Niemeyer, K. Alexandrov, H. Waldmann, *Angew. Chem.* **2006**, *118*, 302; *Angew. Chem. Int. Ed.* **2006**, *45*, 296.

[6] M. Zaja, S. J. Connon, A. M. Dunne, M. Rivard, N. Buschmann, J. Jiricek, S. Blechert, *Tetrahedron* **2003**, *59*, 6545.

[7] J. J. Van Veldhuizen, D. G. Gillingham, S. B. Garber, O. Kataoka, A. H. Hoveyda, *J. Am. Chem. Soc.* **2003**, *125*, 12502.

[8] J. J. Van Veldhuizen, S. B. Garber, J. S. Kingsbury, A. H. Hoveyda, *J. Am. Chem. Soc.* **2002**, *124*, 4954.

[9] J. J. Van Veldhuizen, J. E. Campbell, R. E. Giudici, A. H. Hoveyda, *J. Am. Chem. Soc.* **2005**, *127*, 6877.

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