An Enzyme-Labile Safety Catch Linker for Combinatorial Synthesis on a Soluble Polymeric Support**

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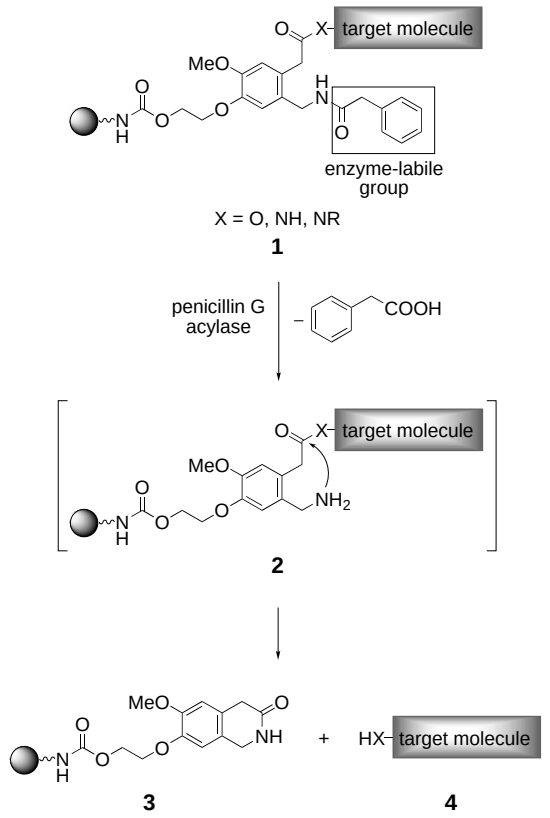
Combinatorial chemistry and parallel synthesis of compound libraries on polymeric supports are efficient methods for the generation of new substances with a predetermined profile of properties.[1] Paramount to the success of this approach is the availability of suitable polymers and widely applicable linker groups for the attachment of the desired compounds to the polymeric carrier. Recent research in this field has focused on and achieved the successful transfer of a steadily growing number of reaction types to synthesis on solid supports, and the demands on the linker groups concerning their stability under different reaction conditions and selective cleavage have risen accordingly. Therefore, new and broadly applicable linkers which are stable under a variety of reaction conditions, but which at the same time allow release of the synthesized products under the mildest conditions, are of great interest in organic synthesis and combinatorial chemistry.[2] Enzymatic methods may open up advantageous alternatives to classical chemical techniques since enzyme-catalzyed transformations often proceed under very mild conditions (pH 5–8, 25–37 °C) and with very high chemo-, regio-, and stereoselectivity.[3] We now report on the development of a new enzyme-labile safety catch linker for combinatorial chemistry[4] on a soluble polymeric support, allowing for the release of the target compounds in high yields and under neutral conditions.

In the design of the new linker groups a biocatalyzed transformation was combined with a subsequent intramolecular cyclization reaction, according to the principle of "assisted removal"[5] of the desired target molecules. To this end the linker embodies a functional group which is recognized and attacked by the biocatalyst. The enzyme liberates an intermediate which cyclizes, with release of the target compounds. Furthermore, the linker carries an additional functional group for attachment to the polymeric support. The realization of this principle is shown in Scheme 1. The linker group is attached to an amino-functionalized carrier as a urethane (→**1**). It allows for the attachment of, for example, alkyl halides, alcohols, or amines as carboxylic acid esters and amides and their subsequent conversion to product libraries. The enzyme-labile functional group, a phenylacetamide, is remote from the structures to be generated by combinatorial



Scheme 1. Principle for the development of the enzyme-labile safety catch linker.

synthesis. Thereby possible steric or electronic interactions of the biocatalyst with the synthesis products, which might lead to a reduction of the substrate tolerance of the enzyme, are minimized. The release of the target molecules proceeds according to the safety catch principle in a two-step process. In the first step, the amidase penicillin G acylase hydrolyses the phenylacetamide with complete chemo- and regioselectivity and under exceptionally mild conditions (pH 7.0, room temperature or 37 °C).[6] Subsequently benzylamine **2**, thereby generated as an activated intermediate, cyclizes to polymer-bound lactam **3** and releases the corresponding target molecule **4**.

The linker was synthesized from commercially available homovanilinic acid methyl ester **5** (Scheme 2). After alkylation of the phenolic hydroxyl group with THP-protected bromoethanol in high yield, the resulting trisubstituted aromatic compound was subjected to a completely regioselective electrophilic amidoalkylation with *N*-hydroxymethylphenylacetic acid amide.[7] Under the acidic reaction conditions the THP ether was simultaneously converted into the corresponding acetate which was then saponified. By means of this three-step sequence, linker **6** is accessible in an overall yield of 67%.

The polymeric carrier chosen for attachment to the linker was a soluble polyethyleneglycol functionalized at both termini with an amino group and with an average molecular mass of 6000 Da (POE 6000).[8] POE 6000 is soluble in numerous organic solvents but can be precipitated, filtered off, and washed after addition of diethyl ether, thereby

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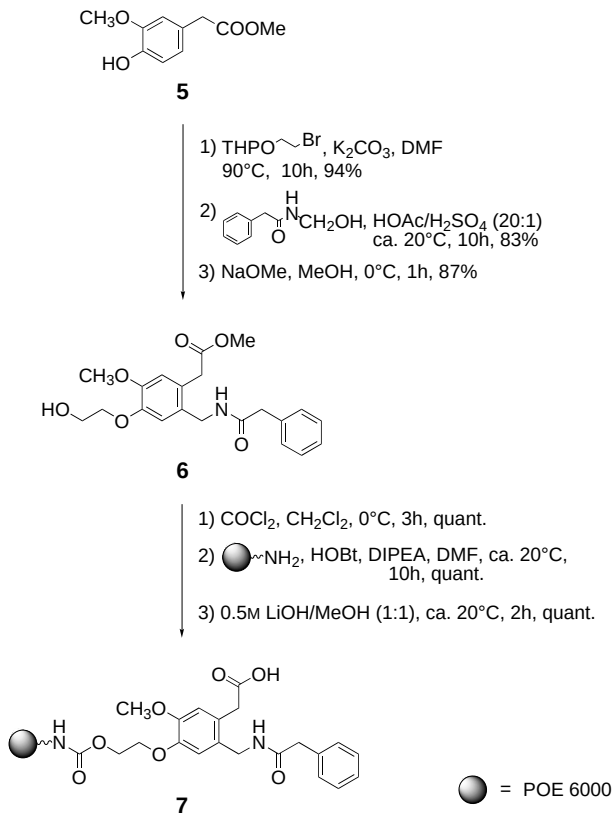
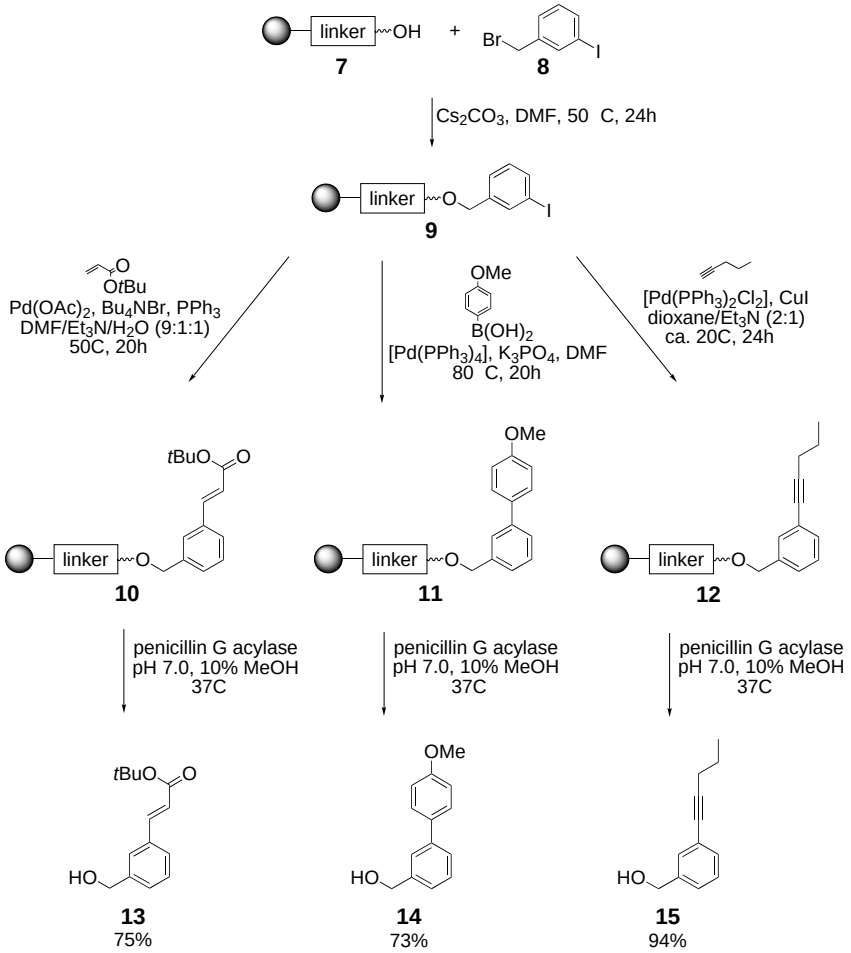


Scheme 2. Synthesis of the linker and coupling to the soluble polymer POE 6000. DIPEA=diisopropylethylamine, HOBt = 1-hydroxy-1*H*-benzotriazole, THP = tetrahydropyranyl.

facilitating the separation of surplus reagents and side products. It allows for NMR spectroscopic monitoring of the reactions and, due to its pronounced solubility in water, for biocatalyzed transformations it offers the advantage that the substrates are accessible to the biocatalyst in homogeneous phase. For the attachment of **6** to POE 6000 the primary hydroxyl group was first converted into the corresponding chloroformic acid ester with phosgene and then the aminofunctionalized polymer was acylated with this intermediate quantitatively.[9] Basic hydrolysis of the methyl ester was also quantitative and yielded functionalized polymer **7**.

The suitability of the polymer–linker conjugate **7** for combinatorial syntheses and, thereby, also the scope of its applicability were investigated for a variety of different transformations. To this end, first the carboxyl group of the linker was esterified with *m*-iodobenzylbromide **8** and the polymer-bound aryl iodide **9** generated was then further transformed by Heck[10], Suzuki[11] or Sonogashira[12] reactions (Scheme 3). NMR spectroscopic analysis revealed that these transformations proceeded quantitatively. Compounds **10**–**12** were then incubated with penicillin G acylase at pH 7 and 37 °C. After 48 hours, benzyl alcohols **13**–**15** were isolated in high yields and with a purity of > 95 % by simple extraction with diethyl ether (the phenylacetic acid remains in the aqueous phase).

In a further series of experiments Mitsunobu esterification and Diels–Alder reactions were investigated (Scheme 4). For the Mitsunobu reaction, the polymer was esterified quantitatively with 1,4-bis(hydroxymethyl)benzene. The obtained polymer-bound benzyl alcohol **16** was then treated with 4-acetamidophenol in the presence of the Mitsunobu reagent to give phenyl ether **17** in quantitative yield. By subsequent treatment of polymeric substrate **17** with penicillin G acylase, aryl benzyl ether **18** could be obtained in 81 % yield. For the Diels–Alder reaction, 4-hydroxybutyl acrylate was coupled to the linker and the polymer-bound acrylic acid ester **19** was treated with cyclopentadiene. This reaction was carried out at 60 °C at ambient pressure, and at 100 °C in a closed glass vial to investigate the pressure and temperature stability of the polymer-bound linker. According to NMR spectroscopic analysis cycloaddition product **20** was formed with an *endo*/*exo* ratio of 2.5:1 and with quantitative conversion. Subsequent enzymatic release delivered alcohol **21** in high yield and purity. The examples shown in Scheme 4 impressively prove the chemoselectivity of penicillin G acylase since the enzyme



Scheme 3. Palladium-catalyzed reactions on polymer **7** and enzyme-catalyzed release of the coupling products from the polymeric carrier.

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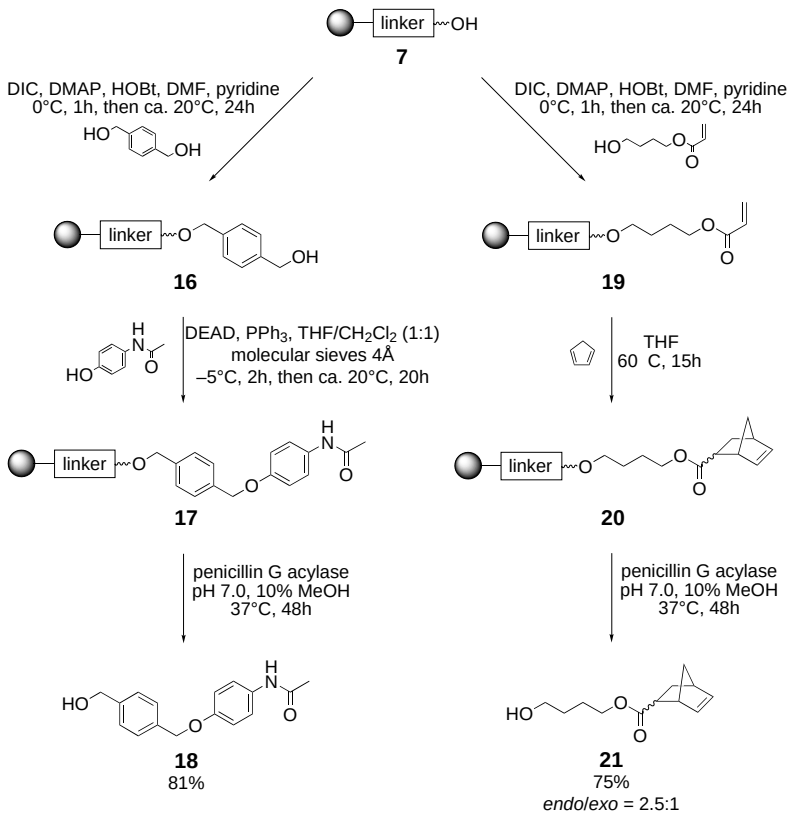

Scheme 4. Mitsunobu and Diels–Alder reactions on polymeric carrier **7** and penicillin G acylase mediated release of the target molecules. DEAD = diethylazodicarboxylate, DIC = diisopropylcarbodiimide, DMAP = 4-(dimethylamino)-pyridine.

attacks only the phenylacetamide unit and not the ester and amide groups which are also present.

In a third series of experiments we investigated whether the enzymatic method also gives access to target molecules that are coupled to the linker as amides. To this end, 4-iodoaniline **22** was coupled quantitatively to linker **7** and the polymer-fixed aryliodide **23** obtained thereby was subjected to a Suzuki reaction[11] with 4-methoxyphenyl boronic acid or a Shille reaction[13] to give enol ether **26** (Scheme 5). Upon treatment with 0.5 M hydrochloric acid, enol ether **26** yielded acetophenone **27**, thereby proving the acid stability of the linker. In order to release the coupling products, polymers **24** and **27** were incubated with penicillin G acylase at pH 7.0 and room temperature. As expected under these conditions the phenylacetamide was hydrolyzed. Due to the higher stability of amides relative to esters, however, the desired cyclization did not occur at room temperature. Upon warming of the reaction mixture to 60 °C, the expected lactam was formed and amines **25** and **28** were released from the polymer.

These results demonstrate that the new enzyme-labile safety catch linker allows for the synthesis of different classes of compounds and the executions of various reaction types. The release of the products proceeds under particularly mild conditions with pronounced selectivity and delivers the desired products in high yield and purity. The linker is stable under different conditions, even at elevated temperature. Beyond the development of a new linker system for combinatorial chemistry our results prove that enzymes, in general, are valuable reagents for transformations on polymeric supports.

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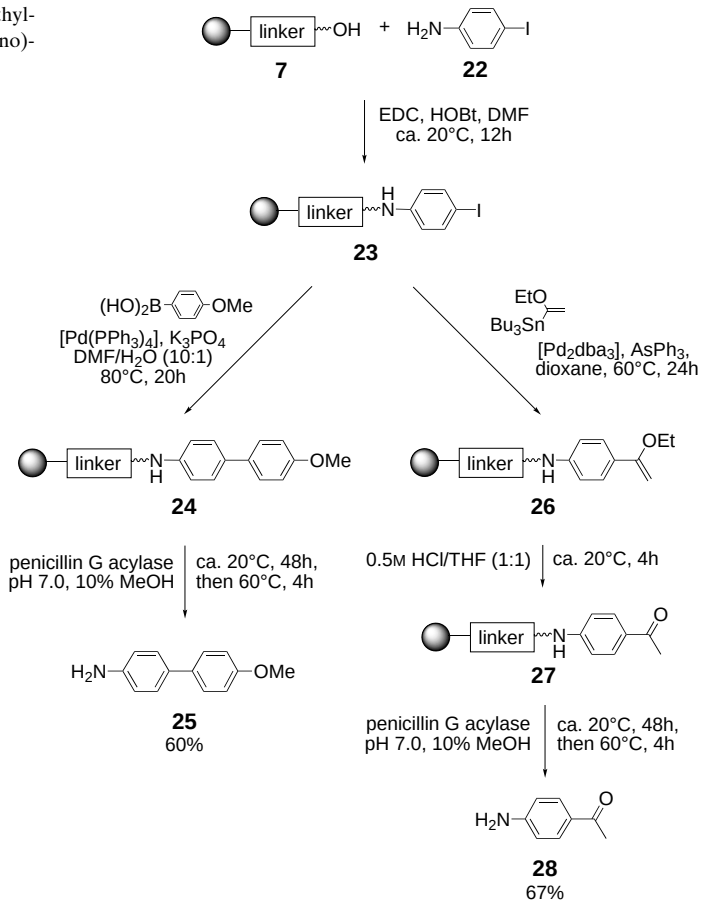

Scheme 5. Palladium-catalyzed synthesis of anilines on the polymeric carrier and their enyzmatic release. dba = *trans*,*trans*-dibenzylidenacetone, EDC = *N*′-(3-dimethylaminopropyl)-*N*-ethylcarbodiimide.

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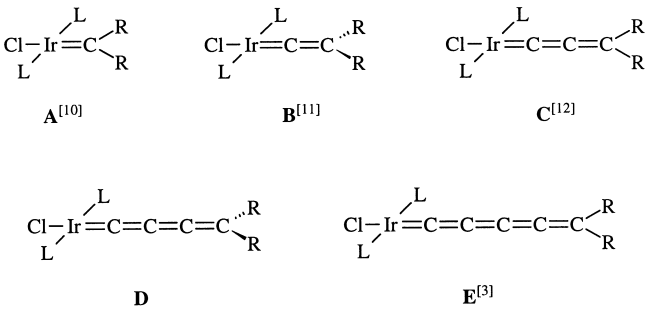
The First Structurally Characterized Metal Complex with the Molecular Unit M=C=C=C=CR_2**

Kerstin Ilg and Helmut Werner*

Dedicated to Professor Henri Brunner on the occasion of his 65th birthday

The chemistry of metallacumulenes of the general composition $[L_xM{=}C(=C)_nRR']$, which could be considered as near relatives of metal carbenes, continues to receive a great deal of attention. While species with $n = 1$ and 2 have already been extensively investigated,[1] not too much is known about the compounds with $n = 3$ and 4. In 1994, Dixneuf and co-workers reported the synthesis of the first cationic complex with $Ru{=}C{=}C{=}C{=}C{=}CPh_2$ as the building block,[2] and a few months later we described the isolation of the first neutral compound with $Ir{=}C{=}C{=}C{=}C{=}CPh_2$ as the core unit.[3] In the meantime, the field of metallahexapentaenes $[L_xM{=}C(=C)_4RR']$ has further been developed, mainly due to the work by Fischer et al.[4]

Earlier attempts to prepare *metallapentatetraenes*, that is compounds with $n = 3$ in the above-mentioned formula, date back to Lomprey and Selegue[5] and somewhat later to Bruce and co-workers.[6] The latter group, by using $[(\eta^5\text{-}C_5H_5)Ru(PPh_3)_2(thf)]PF_6$ and buta-1,3-diyne as the starting materials, generated in situ a cationic complex containing the fragment $Ru{=}C{=}C{=}C{=}CH_2$ and supported the existence of this species by trapping reactions with nucleophiles such as $NHPh_2$, PPh_3, H_2O, and imines. More recently, both the groups of Dixneuf[7] and Winter[8] reported about the in situ formation of cationic intermediates with the molecular unit $Ru{=}C{=}C{=}C{=}CHR$ and revealed that these can be converted to corresponding acylvinylidene, acylalkynyl, butenynyl, and allenylidene ruthenium derivatives. In 1999, a dinuclear cationic compound with the core fragment $[M]{=}C{=}C{=}C{=}CH[M']$ ($[M] = (\eta^5\text{-}C_5Me_5)Fe('P_2')$, $'P_2' = 1,2$-bis(diphenylphosphanyl)ethane (dppe), 1,2-bis(diisopropylphosphanyl)-ethane (dippe); $[M'] = (\eta^5\text{-}C_5Me_5)Fe(CO)_2$) was prepared by Lapinte and co-workers, using the butadiynediyl complex $[M]C{\equiv}CC{\equiv}C[M']$ as the precursor.[9] We have now succeeded with the isolation and structural characterization of the first stable neutral compound of the type $[L_xM{=}C{=}C{=}C{=}CR_2]$ with $L_xM = IrCl(P\mathit{i}Pr_3)_2$, which fills the gap in the system of the metallacumulenes **B** – **E** ($L = P\mathit{i}Pr_3$).



A[10] **B**[11] **C**[12]

D **E**[3]

$L = P\mathit{i}Pr_3$

The synthetic route for the iridium complex of type **D** follows the methodology which we used for the corresponding allenylidene derivatives *trans*-$[IrCl(=C{=}C{=}CRR')(P\mathit{i}Pr_3)_2]$.[11d, 12] Since the C_3 ligand in the compounds of type **C** was generated from propargylic alcohols $HC{\equiv}CCRR'OH$, for the preparation of a species with a homologous C_4 unit we had to find a precursor with an additional carbon atom in the C_n chain. The best choice seemed to be the ethynyl ketone $HC{\equiv}CC(O)CHPh_2$.[13] On reaction of this substrate with the dihydride **1**, the alkynyl(hydrido) complex **2** is formed which, however, is unstable in solution and isomerizes to the corresponding vinylidene derivative **3**. The existence of **2** can be confirmed by trapping the intermediate with pyridine, which results in the formation of the octahedral complex **4**. In contrast to the ^1H NMR spectrum of **3**, those of **2** and **4** display a triplet resonance in the high-field region at $\delta = -40.96$ (for **2**) and -21.68 (for **4**), the chemical shift of which is

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