The *tert*-butyl group in chemistry and biology

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The unique reactivity pattern elicited by the crowded *tert*-butyl group is highlighted by summarising characteristic applications. Starting from the use of this simple hydrocarbon moiety in chemical transformations, *via* its relevance in Nature and its implication in biosynthetic and biodegradation pathways, the way through to its possible application in biocatalytic processes is described.

1. Introduction

In spite of its very simple hydrocarbon structure, the *tert*-butyl group has attracted much attention from organic chemists for many decades due to its unique properties. Indeed, this moiety exhibits a reactivity pattern that enriches the chemist's toolbox in many ways. Starting from simple aspects such as steric hindrance or stability of the associated carbocation all the way through to more complex concepts including chiral catalysis or induced conformational changes in macromolecules, *tert*-butylated compounds have made their way into many domains of organic and chemoenzymatic synthesis. Hence, the *tert*-butyl moiety plays an outstanding role in the field of catalysis science, dramatically affecting the regio- and stereoselectivity of many organic reactions such as reductions, oxidations or C–C bond formations. However, relatively few natural products bearing the *tert*-butyl substructure have been reported, and surprisingly, the implementation of the *tert*-butyl group into chemoenzymatic processes has very rarely been described.

Herein we endeavour to focus on some of the major issues concerning the application of the *tert*-butyl structure in chemical and biocatalytic transformations and to outline the prospects for its potential use in chemoenzymatic processes.

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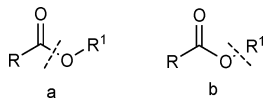
2. The *tert*-butyl moiety in protecting group chemistry

As peptide synthesis gained importance and total synthesis became the "grail" for organic chemists, protecting group technology captured major attention. The *tert*-butyl group with its unique behaviour afforded solutions for orthogonality, which is a key issue in the sequence of protection and deprotection.[1]

2.1 Protection of the carboxyl group

Classically, taking advantage of the carbonyl activity of carboxylic acid esters, hydrolysis can be achieved either under acidic conditions (leading to an equilibrium between ester and carboxylic acid) or irreversibly under basic conditions, affording a carboxylate and the corresponding alcohol (a). Furthermore, a second pattern that leaves the carboxyl carbon intact is known for certain esters (b).



The path ultimately adopted depends on the nature of the alcoholic moiety of the ester (R^1) and on the reaction conditions.

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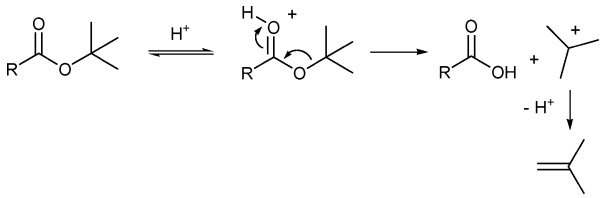
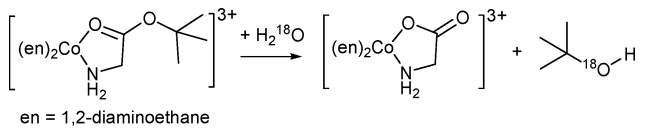
If R^1 is *tert*-butyl, acidic cleavage will proceed according to pattern (b) to give the quite stable *tert*-butyl carbocation[2] and the carboxylic acid (Scheme 1).



Scheme 1 Cleavage of the alkyl–oxygen bond in ester hydrolysis.

In contrast, in basic medium the reaction follows the classical addition–elimination path (a).[3] The more specific case of α-amino acid esters provided evidence for the differential hydrolysis of *tert*-butyl esters compared to other alkyl esters. Indeed, the studies by Wu *et al.* on the hydrolysis behaviour of the cobalt(III) complexes of esters showed that *tert*-butyl esters were prone to undergo a cleavage of the oxygen–alkyl bond, whereas the acyl-oxygen bond was broken in the other cases (Scheme 2). This hypothesis was evidenced and supported by kinetic studies and ^{18}O exchange measurements.[4,5]



Scheme 2 Cobalt-promoted hydrolysis of *tert*-butyl esters.

2.2 Protection of the hydroxyl group

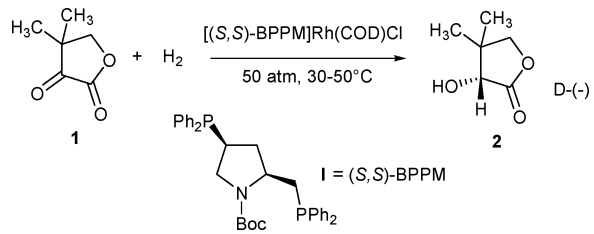
Since the *tert*-butoxy group is extremely stable to strongly basic conditions, it is one of the most versatile alcohol protecting groups. Very recently, a mild removal strategy using anhydrous $CeCl_3$ and NaI in acetonitrile has been designed and developed by Bartoli *et al.*[6] This procedure should further contribute to the popularity of the *tert*-butoxy protecting group, since it can be successfully applied to aliphatic and aromatic *tert*-butyl ethers and is compatible with a series of other protecting groups and functionalities such as ethyl esters, hydroxyl moieties, nitriles and carbonyls.

3. The *tert*-butyl group in catalysis

The biological activity of, for example, pharmaceuticals or agrochemicals, arises from the specific interaction of small molecules with chiral macromolecules as receptors, enzymes or DNA. Thus, stereospecific interplay with enantiomerically pure chiral ligands is prone to elicit an optimal biological response. Accordingly, the quest for such compounds has concentrated the efforts of the synthetic organic chemistry community. Third- and fourth-generation asymmetric synthesis[7] using either chiral reagents or chiral catalysts is considered as the most powerful tool to pursue these objectives. Thus, the development of efficient chiral catalysts allowing for high yields and high enantioselectivities has become a major task for organic chemists. With its tremendous steric bulk and the associated non-bonded repulsions it triggers, the *tert*-butyl group has been widely used in the design and the optimisation of such chiral catalysts. Indeed, the introduction of bulky groups is, with cyclisation, one of the two common methods used to force organic molecules into unusual geometries. In this section, considering the large number of catalysts bearing a *tert*-butyl substructure, we focus on some of the more recent striking examples of the use of chiral catalysts bearing this moiety. Further, some examples of achiral catalytic processes in which the *tert*-butyl group rules the regioselectivity and the intrinsic catalytic activity are outlined.

3.1 Asymmetric reductions

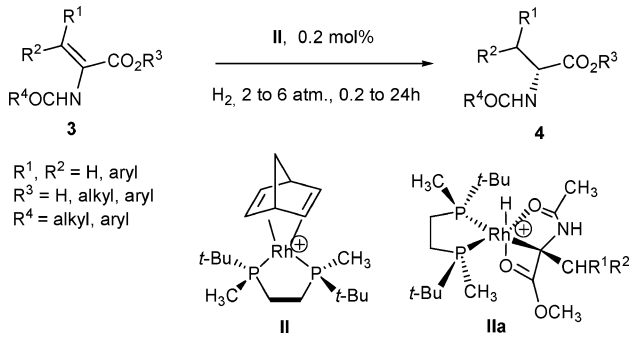
As the carbonyl group plays a pivotal role in organic synthesis, hydrogenation of the C=O bond and its isosteres (enol, imine, enamine) represent, together with the reduction of isolated C=C bonds, the most examined reaction.[8] The majority of these reactions are metal-catalysed, and their efficiency in respect of yield and enantioselectivity depends on the substrate, the metal used and the catalyst from which the chiral information is carried over to the reaction product. As early as 1985, a standardised procedure for rhodium-catalysed asymmetric reduction of ketolactone **1** using (2*S*,4*S*)-*N*-*tert*-butyloxycarbonyl-4-diphenylphosphinomethylpyrrolidine (**I**) ((*S*,*S*)-BPPM) appeared in an *Organic Syntheses* procedure (Scheme 3).[9]



Scheme 3 Asymmetric hydrogenation of ketopentoyl lactone **1**.

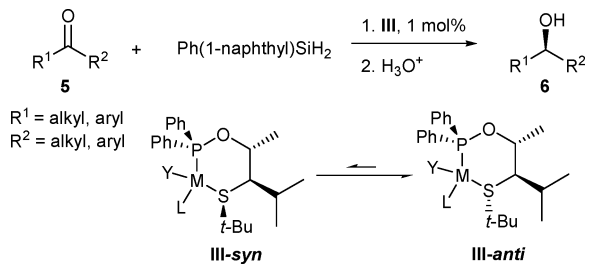
More recently, Imamoto *et al.* reported a class of tetra-alkyl *P*-chiral bis-phosphine ligands that assemble to form five-membered C_2-symmetric chelates with the metal, and thus induce high enantioselectivity in rhodium-catalysed hydrogenation reactions.[10,11] As a crucial feature, these ligands possess a bulky substituent (*tert*-butyl) and a methyl group (the smallest possible alkyl group) attached to each phosphorous atom. The hydrogenation of enamides **3** and β-ketoesters proceeded with ee-values as high as >99% and up to 89%, respectively (Scheme 4), and high catalytic activity. X-Ray structure analysis of the catalyst precursor **II** clearly showed a λ-conformation of the chelate complex with a quasi-symmetrical coordination of the diene. The pseudo-equatorial orientation of the stereo-demanding *tert*-butyl groups and the resulting pseudo-axial positioning of the two methyl groups in the thus rigidified chelate complex **II** is the clinching argument for a highly enantioselective process. Dynamic studies by means of NMR techniques have shown that the observed stereoselection actually reflects the stereochemical prevalence at the stage of the monohydride intermediate **IIa**. This observation suggests that the migratory insertion step is irreversible and that the geometry in **IIa** is stereo-determining. However, replacement of the *tert*-butyl group on the phosphorus atom by either an isopropyl or the even bulkier Et_3C- leads to lower enantioselectivities.

Evans *et al.*[12] reported a new family of mixed phosphorus/sulfur ligands lacking C_2-symmetry that are capable of



Scheme 4 *P*-Chiral bis(trialkylphosphine)s in enantioselective hydrogenation.

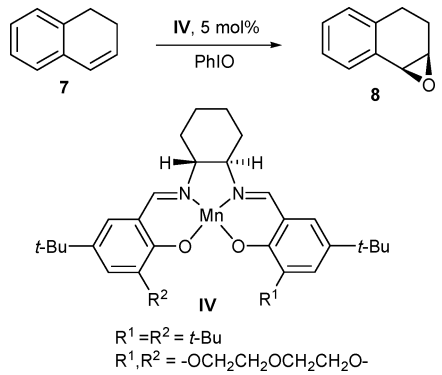
rhodium-catalysed enamide reductions and asymmetric hydrosilylation of prochiral ketones **5** (Scheme 5). The determining features of these ligands are the presence of a strong and weak donor heteroatom pair and the generation of a stereogenic centre at the sulfur atom upon coordination with the metal. The rationale behind the design of such ligands is that since the stereochemical information is located on the sulfur it should improve the stereocontrol of the reaction as compared to ligands with the chiral information located more remotely from the metal centre. The major drawback of this strategy is the low inversion barrier observed for metal-coordinated thioethers. The *tert*-butyl group introduced at the sulfur atom is forced into a pseudoaxial orientation by means of non-bonded repulsions to avoid interaction with the backbone and serves to control the equilibrium of the *anti*- and *syn*-diastereomeric chelate complexes **III-*syn*** and **III-*anti***, respectively, ensuring high enantioface selectivity. Furthermore, the strong electron-donating character of the *tert*-butyl group forces the enamide to coordinate to the sulfur with its most electron-poor carbon atom. Enantiomeric excesses ranging from 78% to 98% have been obtained for these reactions, illustrating that the *tert*-butyl group influences the stereochemical outcome of such a reaction not exclusively by means of steric hindrance.



Scheme 5 Chiral P/S-ligands in asymmetric ketone reduction.

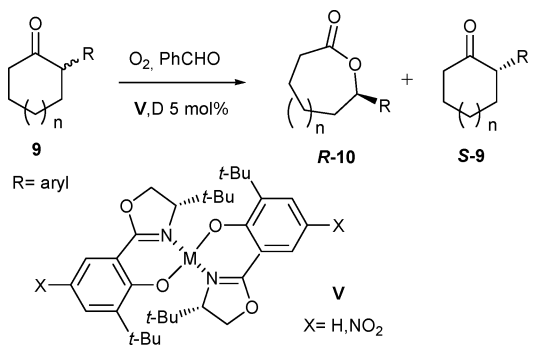
3.2 Asymmetric oxidations

The Nobel Prize Committee has paid tribute to the tremendous work done in the area of asymmetric oxidation reactions by rewarding K. Barry Sharpless with the Nobel Prize for Chemistry in 2001 (together with Noyori and Knowles; see reductions). Because optically active epoxides open a large window for follow-up chemistry leading to many biologically active chiral compounds, the asymmetric epoxidation of unfunctionalized alkenes remains of great interest. Larrow and Jacobsen have developed an efficient large-scale synthesis for a Schiff-base ligand (**IV**) bearing four *tert*-butyl groups capable of the Mn(III)-catalysed epoxidation of olefins (Scheme 6).[13] Very recently, Martinez *et al.* have bridged the two salicylindene moieties by replacement of the *tert*-butyl groups at the 3- and 3′-positions by symmetrical aliphatic polyether or benzylic diether linkers. The modest ee-values obtained sustain the essential role of the *tert*-butyl groups in the enantiodiscrimination process.[14] The *tert*-butyl groups most likely endow a preferred conformation to the catalytic complex without rigidifying it.



Scheme 6 Enantioselective epoxidation.

An asymmetric metal-catalysed Baeyer–Villiger-type oxidation, which formally corresponds to a kinetic resolution, has been reported by Bolm *et al.*[15] The 2-aryloxazoline Ni(II) complex used as the chiral catalyst has four *tert*-butyl groups that are not only essential for optimal chiral induction but also for high catalytic activity. Indeed, removal of the *tert*-butyl groups from the arenes leads to a loss of catalytic activity (yields <10%). Using 5 mol% of the chiral catalyst **V**, 2-phenylcyclohexanone **9** dissolved in water-saturated benzene in the presence of benzaldehyde as oxygen acceptor is converted to lactone (*R*)-**10** at room temperature in an oxygen atmosphere with 41% yield and 65% ee (Scheme 7). Replacement of the *tert*-butyl moiety by isopropyl on the oxazoline ring results in a drop of enantioselectivity (32% ee).



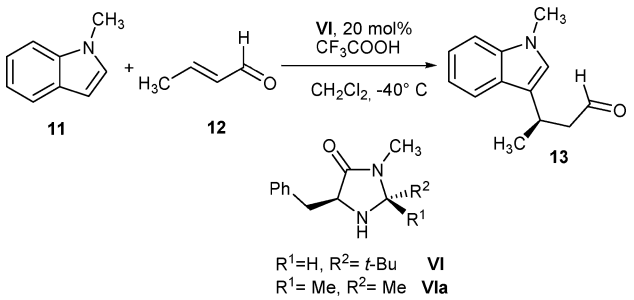
Scheme 7 Kinetic resolution of cyclic ketones by a Baeyer–Villiger-type oxidation.

3.3 Asymmetric C–C bond-forming reactions

Reactions that allow for asymmetric C–C bonding are of paramount importance for many syntheses of chiral compounds based on a retrosynthetic approach.

The mixed phosphorus/sulfur ligands **III** described above (Section 3.1) were actually originally developed by Evans *et al.* for palladium-catalysed allylic substitution.[16] Using these ligands, values ranging from 90–98% ee were obtained for the allylic substitution of 1,3-diphenylpropenyl acetates and cycloalkenyl acetates with diethyl malonate as nucleophile. Applied to heterocycles, this method allowed for the efficient enantioselective synthesis of 3-substituted piperidines and dihydrothiopyrans.

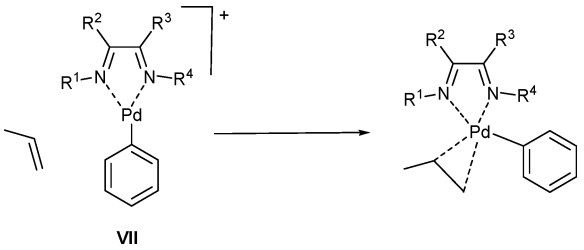
Austin and MacMillan have developed a new phenylalanine-based organocatalyst for the Friedel–Crafts alkylation of non-electron-rich heteroaromatic compounds such as indoles.[17] Since the overall rate of the iminium-catalysed reaction depends on the rate of iminium formation as well as on the rate of C–C bond formation, the rationale that guided the design of **VI** is as follows: implementation of a *tert*-butyl group provides an imidazolidinone conformation with an exposed nitrogen lone pair (responsible for rapid iminium formation) and an increased *Si*-face coverage (associated with a less hindered *Re*-face in the resulting activated iminium species), which should lead to an increased face selectivity and substrate addition rate. Thus, with this synergistic effect, both the reactivity and the enantioface discrimination should be increased. In fact, it was observed that with trifluoroacetic acid as co-catalyst and 20 mol% of **VI** in CH_2Cl_2 at −40 °C, the Friedel–Crafts alkylation of the unreactive *N*-methylindole (**11**) with various α,β-unsaturated aldehydes (such as **12**) can be achieved with yields >80% and with 85–92% ee (Scheme 8), compared to 56% ee using catalyst **VIa**.



Scheme 8 Enantioselective indole alkylations.

3.4 Miscellaneous

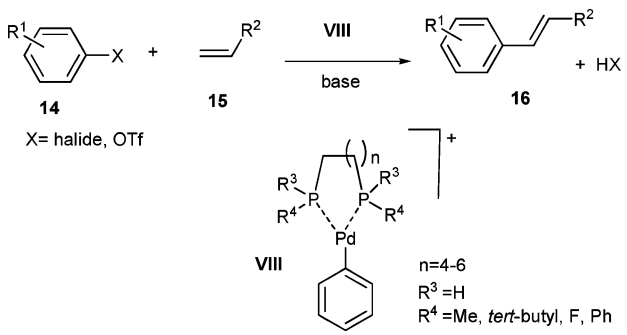
Heck arylation is usually catalysed by Pd(0) species in the presence of phosphine ligands. Nevertheless, other metals and other metal-coordinating atoms have been employed. The major issue for the catalytic entity is activity and regiochemistry in the case of unsymmetrical alkenes. Von Schenck *et al.* have studied this reaction catalysed by a palladium(II) diimine complex **VII** (Scheme 9).[18] All four positions R^1–R^4 have been systematically substituted, affecting the electronic parameters (OMe, F) as well as the steric parameters (Me, *t*-Bu). The π-coordination of propene to Pd(II) depends on the σ-donation from the π-orbital of propene and the π-back-donation from Pd to the π*-orbital of propene. Both factors affect the HOMO and LUMO energies. It was found by calculation that electron-donating groups (EDGs) such as OMe destabilize HOMOs and LUMOs, by increasing the π-coordination energy, which destabilizes the system. The effect is reversed in the case of electron withdrawing groups (EWGs)



Scheme 9 Electronic and steric ligand effects in the Heck reaction.

like F. The *tert*-butyl and methyl groups are EDGs, but their steric effect is the main factor that affects this reaction. The interaction between propene and the ligand system of the catalyst with bulky groups increases the energy of π-coordination as well as the activation energy when R^1 and R^2 are *tert*-butyl groups, thus dramatically affecting the regiochemistry.

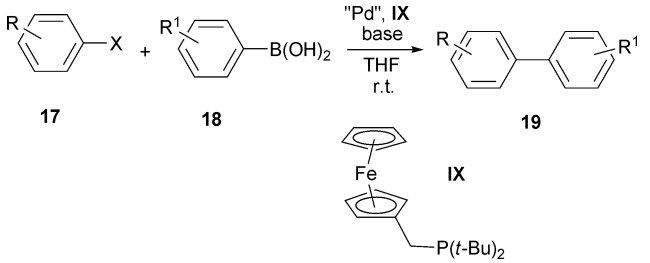
Chen *et al.* studied a similar system.[19] They reported the Heck reaction of ethene **15** with cationic phenylpalladium and phosphine ligands **VIII** (Scheme 10). Several ring sizes (4–6) and several substituents were used in this study. No linear correlation was found between π-coordination strength and the frontier molecular orbital (FMO) energies. However, EDGs increase the insertion barrier (9.3, 9.7, and 11.6 kcal mol^{-1} for R^4 = H, CH_3, and *t*-Bu, respectively), while EWGs decrease it (8.0 kcal mol^{-1} for R^4 = F). The dramatic increase observed in the case of *tert*-butyl illustrates the major role played by the steric factor for the insertion reaction.



Scheme 10 Ligand effects on migratory insertion in the Heck reaction.

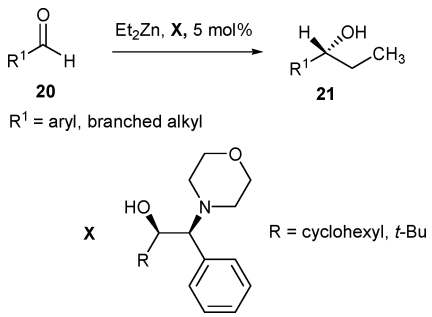
Primary dialkyl(ferrocenylmethyl)phosphines are known to be air-stable. The addition of two highly electron-donating substituents affords di-*tert*-butyl(ferrocenylmethyl)phosphine **IX**, capable of acting as a ligand in Pd-catalysed Suzuki–Miyaura couplings of organoboronic acids with aryl bromides at room temperature. Despite the electron-donating effect of the two *tert*-butyl groups, ligand **IX** described by Sliger *et al.*[20] (Scheme 11) is air-stable in the solid state, allowing for easy handling. However, this catalyst exhibits only modest Heck coupling activities.

Implementation of ferrocenophane monomers with a hydrophobic *tert*-butyl group was found to enhance the solubility in organic solvents of the polymers obtained upon ring-closing metathesis polymerization. Thus, subsequent analysis, treatment and processing of the ferrocene-containing polymers was facilitated.[21] This illustrates an example of where it is not the bulk but the hydrophobic interactions arising from the *tert*-butyl group that play the crucial role.



Scheme 11 Di-*tert*-butyl(ferrocenylmethyl)phosphine in cross-coupling reactions.

Finally, to show that the use of *tert*-butyl groups does not always improve the selectivity of catalysts, we describe one counter-example. Enantioselective addition of diethyl zinc to aldehydes **20** is a common method to test the activity and the enantioselectivity arising from the use of a new ligand. Amino alcohol **X**, a ligand that combines a "bulky" substituent on the OH-terminal side and a "flat" one on the NH-side, was shown to induce high enantioselectivity in such a reaction. Replacement of the cyclohexyl moiety by the *tert*-butyl group resulted in a decrease of enantioselectivity (Scheme 12).[22]

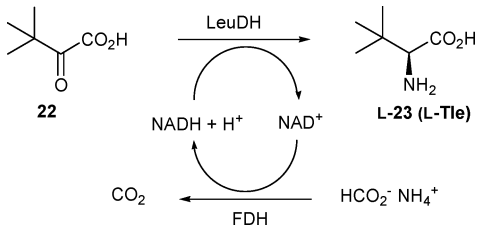


Scheme 12 Enantioselective addition of organozinc reagents to aldehydes.

3.5 Catalysts derived from *tert*-leucine (Tle)

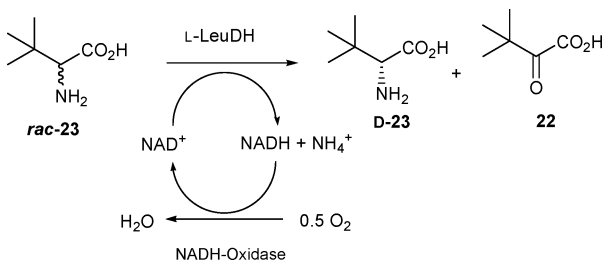
Tle is considered separately in this section. Although it is a non-coded α-amino acid, it is nevertheless one of the *tert*-butylated features found in Nature, and thus serves here as a transition between the synthetic chemistry of the *tert*-butyl moiety discussed above and its role in Nature that will be focussed on below.

Tle-based chiral catalysts have found their place in the chemist's toolbox, and many different approaches have been devised for the synthesis of enantiomerically pure Tle. However, Tle is not readily available from the chiral pool, and so it has to be obtained either from resolution of a racemic mixture or by asymmetric synthesis. Both strategies can be achieved either chemically[23,24] or enzymatically,[25–27] and some routes to enantiomerically pure D- and L-Tle are summarized in a review by Bommarius *et al.*[28] L-Tle is prepared on the tonne scale by a chemoenzymatic method developed at Degussa AG (now Evonik).[29,30] This method is based on the nicotinamide adenine dinucleotide (NADH)-dependent reductive amination of trimethylpyruvic acid catalysed by leucine dehydrogenase (LeuDH) in the presence of formate dehydrogenase (FDH) (Scheme 13). Accordingly, only a catalytic amount of cofactor is needed since the latter is regenerated by means of an irreversible FDH-catalysed reaction.



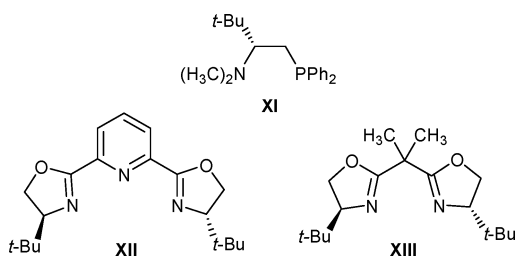
Scheme 13 Chemoenzymatic synthesis of L-Tle.

More recently, Hummel *et al.*[27] have reported on an enzymatic oxidation strategy for the synthesis of enantiomerically pure D-Tle. Resolution of the racemic mixture is based on the reverse reaction, namely the NAD^+-dependent oxidation of L-Tle catalysed by LeuDH. Since the equilibrium strongly favours the reduced product **L-23** in these reactions, they can only be used for oxidation upon development of an efficient irreversible cofactor regeneration system (Scheme 14). The authors have isolated and characterised an NADH oxidase from *Lactobacillus brevis* that is capable of NAD^+ regeneration while producing H_2O as a by-product. Thus, the equilibrium is driven to the oxidation side (compound **22**).

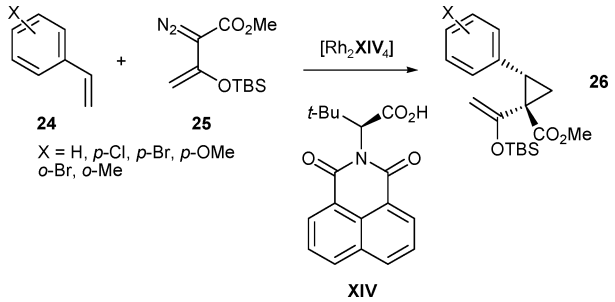


Scheme 14 Chemoenzymatic synthesis of D-Tle.

In the early 1980s Hayashi *et al.*[31,32] reported the preparation of (*R*)-*tert*-Leuphos **XI** as a ligand for Ni(II)-catalysed asymmetric Grignard cross-coupling with high enantioselectivities. Different ligands including an oxazoline substructure based on *tert*-leucinol[33] have been designed for Rh, Ir and Cu catalysis. Indeed, C_2-symmetric bis-oxazoline catalysts **XII** and **XIII** have been developed[34] for hydrosilylation and hydrogenation of arylalkylketones[35,36] and cyclopropanation reactions[37,38] with good enantioselectivities.

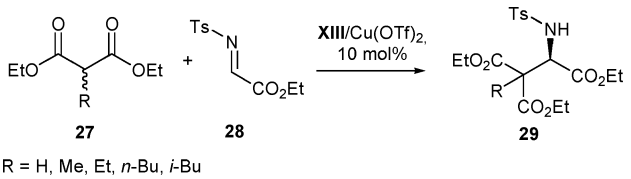


(*S*)-*N*-1,8-Naphthoyl-*tert*-leucine **XIV** has been developed for the enantioselective Rh(II)-catalysed cyclopropanation of styrene **24** with (silanyloxyvinyl)diazoacetates such as **25** with *cis*/*trans* diastereoselectivities of up to 98.5% de and enantioselectivities of 98% ee (Scheme 15).[39] The same catalyst was used for a highly enantioselective intramolecular CH insertion with alkyl diazo(trialkylsilyl) acetates.[40]



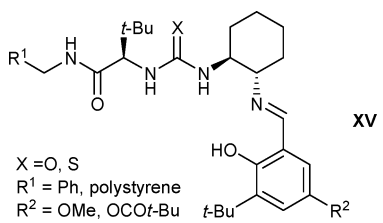
Scheme 15 Catalytic asymmetric cyclopropanation of styrene.

Stereoselective construction of all-carbon quaternary centres is still very challenging for synthetic organic chemists.[41] Marigo *et al.* developed a direct Mannich reaction of 2-substituted malonates **27** and β-ketoesters with *N*-tosyl-α-imino esters **28**. Use of the *tert*-butylbis(oxazoline) ligand **XIII** and $Cu(OTf)_2$ yielded the Mannich adducts with impressive enantiomeric excesses of up to 96% (Scheme 16).[42] The phenyl analogue of **XIII** catalyses the same reaction with only a modest 35% ee.



Scheme 16 Direct catalytic asymmetric Mannich reaction.

Jacobsen *et al.*[43] have identified Schiff bases of general structure **XV** as an efficient soluble non-metal catalysts for the asymmetric Strecker reaction. Alkyl-, cycloalkyl- and arylaldimines undergo hydrocyanation upon treatment with trimethylsilyl cyanide at −70 °C in presence of 2 mol% catalyst affording the corresponding aminonitriles in good yields and up to 97% ee. From a library of 70 different compounds, the highest ee-values clearly arose from the catalysts obtained from Tle. A continuous decrease in ee is observed upon moving from Tle to leucine *via* valine and isoleucine. Moreover, the 5-pivaloyl-substituted salicylimine moiety proved to be the best catalyst in this series. Even if the starting material is converted quantitatively with no detectable by-product, the isolated yields suffer from some losses due to work-up, which includes a column chromatography to remove the catalyst. Thus, the authors have developed a polymer-bound analogue, resulting in improved isolated yields with a marginal loss of enantioselectivity. This resin-bound catalyst allows for simple filtration work-up, and additionally it can be re-used without loss of activity or enantioselectivity.

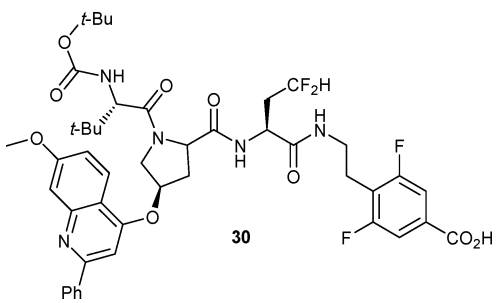


The scope of catalysts of general structure **XV** has been extended to the addition of other nucleophiles to aldimines or methylketimines. Thus, asymmetric catalytic addition of ester enolate equivalents to imines, a Mannich-type reaction, leads to protected β-amino acids – another key element in the development of stable small peptides – with high isolated yields and excellent enantioselectivities.[44] The mechanisms of these non-metal-catalysed C–C bond-forming reactions has been tackled, and it has been found that Strecker reaction follows a classical Michaelis–Menten kinetic with a first-order dependence on catalyst and the cyanide source, and saturation kinetics with respect to the substrate. Accordingly, a reversible imine–catalyst complex has to be formed prior to cyanide addition. The structure of the catalysts strongly suggests that the complex is stabilised by hydrogen bonding between the imine nitrogen and an acidic proton in the catalyst. Sustained by this type of mechanism and enforced by the amide–urea substructure in **XV**, the authors introduced the notion of enzyme-like catalysts.[45]

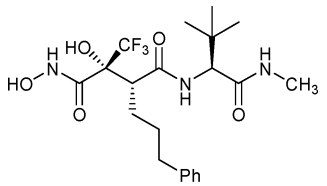
In this section, we have outlined some reactions in which the *tert*-butyl group is a key player with regard to reactivity and/or selectivity. Moreover, even though steric bulk is an essential aspect of the reactivity pattern elicited by the *tert*-butyl moiety (*e.g.*, by modifying secondary structures), its strong electron-donating character, its hydrophobic nature influencing, for example, solubility or non-bonded repulsions, and the stability of the associated carbocation-driving mechanisms, are other properties upon which this special hydrocarbon group exerts its influence.

4. The *tert*-butyl group in synthetic bioactive compounds

Among the non-proteinogenic amino acids, Tle is certainly one of the most popular in the synthesis of stable bioactive peptides, resulting in dramatically improved pharmacokinetic properties. The replacement of valine, leucine and/or isoleucine by Tle increases the stability of peptides in respect of chemical and/or enzymatic hydrolysis not only by means of shielding the carbonyl carbon by steric hindrance but also by drastically modifying the secondary structures.[46] Analogues of bioactive peptides **30** bearing the L-Tle subunit have been described as inhibitors of hepatitis C NS3 protease, which is responsible for preprotein cleavage, an essential step in the life cycle of the virus.[47–49]

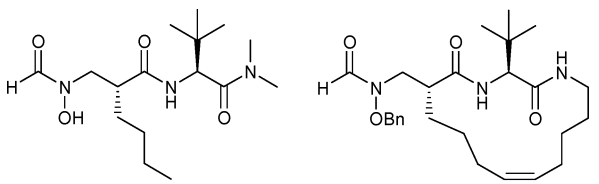


A loss in the regulation of matrix metalloproteinases (MMP), a family of Zn-dependent endopeptidases involved in the degradation of major components of the extracellular matrix, is associated with severe diseases such as cancer and arthritis. A series of stable peptidomimetic hydroxamic acid derivatives **31** with a L-Tle subunit have been developed as inhibitors of MMP-3, -6 and -9 subclasses of matrix metalloproteinases.[50,51] Selective inhibitors of MMP-3 have been conceived for the topical treatment of chronic dermal ulcers.[52]



31

As antibiotic resistance has become a major concern, the identification of new therapeutic targets to fight bacteria is of prime importance. Peptide deformylase (PD), an essential iron-containing metallo-enzyme involved in the deformylation of the *N*-formylmethionine of ribosomes, is one of the new putative targets for antibacterial chemotherapy. Both the dipeptide isostere **32**[53] and the macrocyclic dipeptide **33**[54] bearing a L-Tle moiety have been described as potential antibiotics that target the PD.



32 33

Among many other bioactive compounds containing a Tle moiety, γ-secretase inhibitors targeting Alzheimer's disease,[55] metalloproteinases responsible for protransformation of growth factor α as putative weapons to fight cancer,[56] and HIV-protease inhibitors[57] are some of the most impressive examples.

5. The *tert*-butyl group in Nature

Although natural products containing *tert*-butyl groups are considered to be rare,[58] several different substance classes such as peptides, terpenes, carbinols, esters and even a ketone bearing a *tert*-butyl group have been described in the literature.[59] As mentioned above, Tle is a non-proteinogenic amino acid that, nevertheless, occurs in both enantiomers in Nature as a feature of diverse natural products. Antimitotic peptides containing a Tle unit that interact with tubulin and display high cytotoxicity towards human cell lines have been isolated from marine sponges. The structures include hemiasterlin, hemiasterlin A, milnamide A, criamide A and B (Fig. 1).[60,61] Other polypeptides with antibiotic activity, including discodermin E, a tetradecapolypeptide bearing both a D- and L-Tle unit, have been found in marine sponge species.[62,63]

However, the *tert*-butyl group is not just found as Tle in Nature, and the gingkolides are prominent terpene representatives of *tert*-butyl-bearing natural products (Fig. 2). First isolated in 1932, the structures of the ginkgolides were elucidated in the late 1960s by the pioneering work of Nakanishi.[64] At the time, they were described as unique cage molecules bearing an unprecedented *tert*-butyl group. A trilactone sesquiterpene (bilobalide) bearing a *tert*-butyl group and closely related to the C-20 ginkgolides has been described.[65] More recently, ginkgolide derivatives have been evaluated as human platelet-activating factor receptor antagonists.[66] Related structure–activity relationships have proven the essential role of the *tert*-butyl moiety for this biological activity by means of binding to a hydrophobic pocket in the receptor.[67]

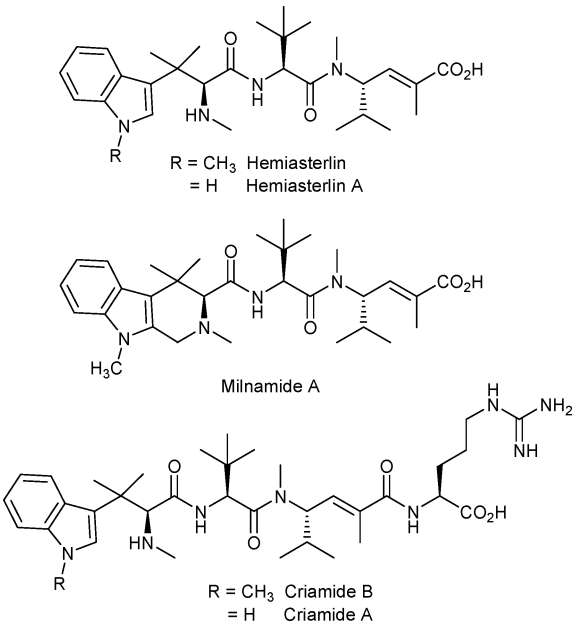


Fig. 1 Peptides bearing a Tle substructure.

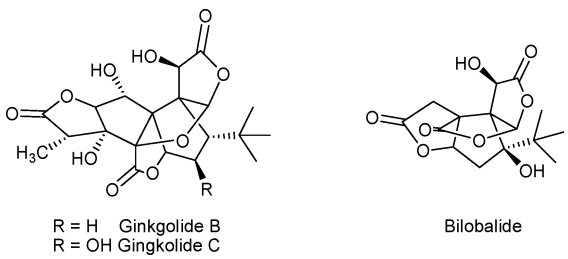


Fig. 2 Ginkgolides.

Mujumdar *et al.* have isolated the very unusual *tert*-butyl ketone coumarin swietenone (**34**) from a benzene extract of *Chloroxylon swietenia*, a tree found in India.[68] A terminal *tert*-butyl group is found in the antifungal polyketide butyrolactol A (Fig. 3).[69]

In addition to many sterols with highly branched side chains obtained from marine sponges, sterols bearing *tert*-butyl side chains, *e.g.* **35**,[70,71] have also been isolated from higher plants.[72] *tert*-Butyl ethers such as **36**[73] and bryostatin 14[74] (a pivalic acid ester like neristatin) illustrate the structural and functional diversity of *tert*-butyl-containing natural products. In total, almost 300 compounds belonging to a few structural families have been isolated from natural sources.[75,59]

5.1 Biosynthetic origin

Only a few reports dealing with the biosynthetic aspects of the *tert*-butyl group have been published. Nakanishi and Habagushi[76] have described a mevalonate pathway for the biosynthesis of ginkgolides, thus establishing their terpenoid nature. In their scheme, the *tert*-butyl group arises by cleavage of the C–C bond adjacent to a *gem*-dimethyl unit followed by methylation elicited by *S*-adenosylmethionine (SAM) (Scheme 17). Recent biosynthetic studies have led to the hypothesis that the classical mevalonate pathway is only a minor route and that a non-mevalonate pathway accounts for the synthesis of the ginkgolides.[77,78] However, the

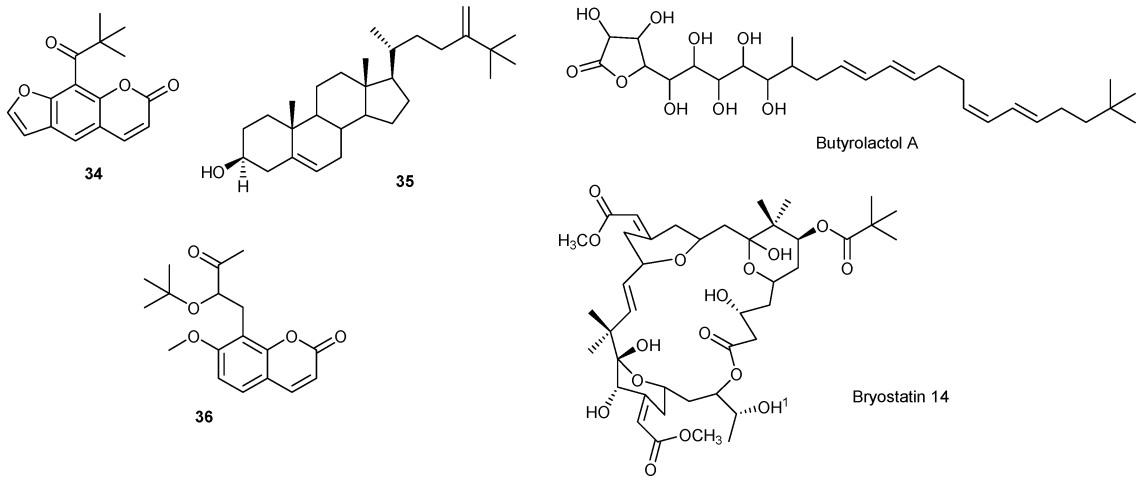
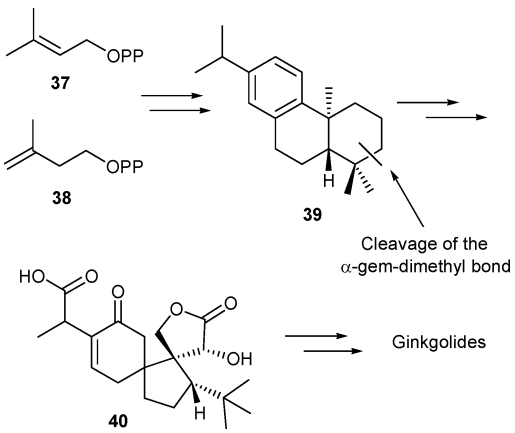


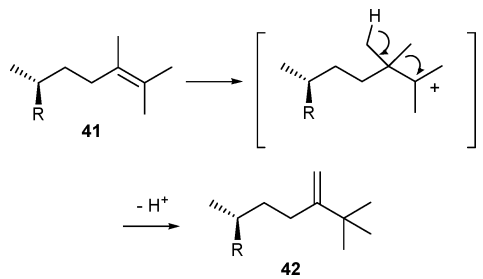
Fig. 3 Natural products with a *tert*-butyl group.



Scheme 17 Biosynthetic origin of the *tert*-butyl group in the ginkgolides.

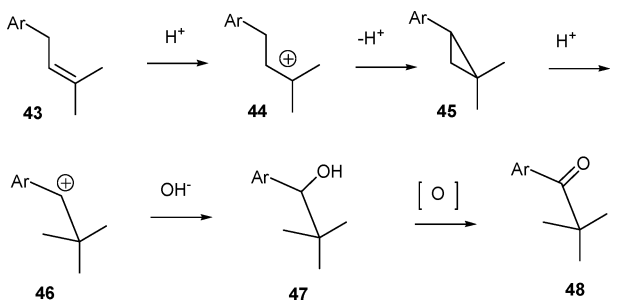
origin of the *tert*-butyl group in the ginkgolides is not further discussed.

An analogous mechanism involving SAM with subsequent hydride abstraction and methyl shift has been postulated by Giner and Djerassi[72,79] for the origin of the *tert*-butyl side chain in **35** (Scheme 18).



Scheme 18 Postulated biosynthetic pathway for *tert*-butylated sterols.

Mujumdar *et al.*[68] proposed a slightly different mechanism to explain the occurrence of the *tert*-butyl group in the coumarin derivative swietenone (**34**). Assuming an isoprene origin, they postulate a mechanism involving formation and subsequent opening of a cyclopropane ring. The driving force for this reaction is the intermediate benzylic carbocation obtained upon cyclopropane ring opening (Scheme 19).



Scheme 19 Putative biosynthetic origin of the *tert*-butyl group in swietenone (**34**).

Recently, Moore and Hertweck have rationalised the presence of a terminal *tert*-butyl group in butyrolactol A.[80] They consider that, even though it could arise from a *tert*-valeryl polyketide starter unit derived from Tle, it is more likely that it arises from an isobutyrate starter with subsequent methylation with SAM by means of a free-radical mechanism. However, this hypothesis has not yet been confirmed by experimental evidence.

5.2 The *tert*-butyl group in chemoenzymatic processes

The steric hindrance of a *tert*-butyl group can either prevent a substrate from reacting, or it can be used to induce high selectivity in a given reaction. Hence, *tert*-butyl ketones or imines, respectively, undergo only a limited series of chemical reactions but with high regioselectivity and (in the presence of chiral catalysts) with high stereoselectivity. Among such asymmetric reactions, hydrogenation,[81] the Strecker reaction, the Mannich reaction, allylation, and alkynylation[82,83] constitute the most prominent examples. *tert*-Butyl-β-oxocarboxylic acids are also prone to undergo enantioselective hydrogenation employing chlorodiisopinocamphenylborane as the reducing agent.[84] The stereochemical induction is explained by two diastereomeric bicyclic transition states **49a** and **49b** (Fig. 4). Bulky R groups (such as *tert*-Bu) lead to >98% ee and chemical yields >90%.

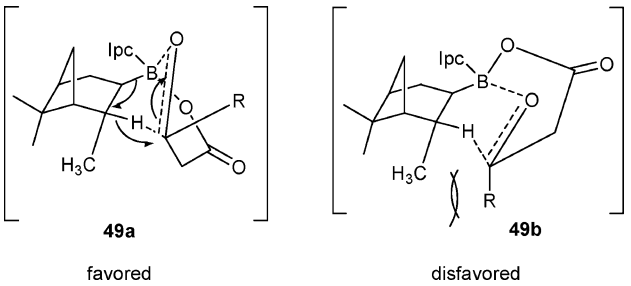


Fig. 4 Diastereomeric transition states in hydrogenation employing chlorodiisopinocamphenylborane.

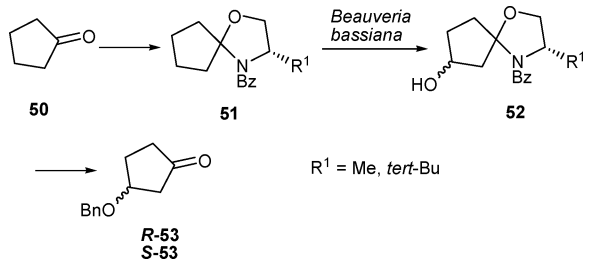
The reactivity problem resulting from bulky groups is even more striking with regard to chemoenzymatic conversions. Indeed, to accept substrates containing groups such as *tert*-butyl, enzymes need either a hydrophobic pocket to interact with it (or a space that can receive this group),[85] or to be able to interact with the substrate in such a way that the *tert*-butyl group does not severely interfere with the catalytic activity.

Esters from sterically hindered alcohols such as *tert*-butyl esters or other esters of tertiary alcohols are known to be very poor substrates for most of the known hydrolases; the few observed conversions occur with quite low reaction rates and enantioselectivity. Hotta *et al.*[86] have reported on a carboxylesterase (Est) from a hyperthermophilic archaeon capable of the hydrolysis of *tert*-butyl acetate. Since Est shows high activity at elevated temperature and high stability in water-miscible solvents, it is ideal to solubilise and react with hydrophobic substrates such as *tert*-butylated long-chain carboxylic acids.

Fatty acid esters from sterically hindered alcohols are barely accepted as lipase substrates. Yeo *et al.* have screened a series of lipases obtained from 279 bacteria strains selected on their ability to grow with *tert*-butyl octanoate as the sole carbon source. From this extensive screening they have found only one lipase from *Burkholderia* sp. YY 62 that showed 100-fold improved activity over commercial lipases.[87] Earlier, Schulz *et al.* reported on the enzymatic cleavage of *tert*-butyl esters.[88] Indeed, a serine protease from *Thermoactinomyces vulgaris*, thermitase, characterised by a high ratio of esterase/peptidase activity, is capable of *tert*-butyl ester hydrolysis of *N*-protected peptides. An analogous *tert*-butyl deprotection is observed with glycopeptides. Another enzymatic removal of the carboxyl *tert*-butyl protection has been described by Schmidt *et al.*[89] This work is based on the finding that a certain amino acid sequence, the GGG(A)X-motif, in the active site of hydrolases engenders catalytic activity towards tertiary alcohol ester hydrolysis. It is proposed that the conserved motif contributes to the formation of the oxyanion hole, and thus stabilises the oxygen anion of the carboxyl group in the tetrahedral intermediate state upon H-bonding. However, the reported activities and substrate scope are quite modest.[90]

Besides hydrolases, only few other enzymes are known to accept *tert*-butylated compounds as substrates. Among the biopolymers capable of such reactions, Zhu and Hua have evaluated the activity and the substrate range of a short-chain dehydrogenase/reductase encoded by a gene from *Sporobolomyces salmonicolor*.[91] This NADPH-dependent enzyme hydrogenates the carbonyl function of ketones, α-ketoesters and β-ketoesters. Most importantly, sterically bulky substrates such as ethyl 3,3-dimethyl-2-oxobutanoate, ethyl 4,4-dimethyl-3-oxopentanoate and 2,2-dimethyl-1-phenylpropanone, all bearing a *tert*-butylketone substructure, are reduced with remarkable specific activity and >98% ee. 4,4-Dimethyl-3-oxopentanenitrile is reduced to the corresponding (*S*)-alcohol with 71% yield and 83% ee by the fungus *Curvularia lunata*.[92]

De Raadt *et al.*[93,94] have described a biohydroxylation in position 3 of cyclopentanone using the fungus *Beauveria bassiana* ATCC 7159. For this transformation they have employed the docking/protecting concept. Thus, with R^1 = Me they obtained (*R*)-**53**. With R^1 = *tert*-butyl the enantiomeric (*S*)-**53** was gained with 50% yield and 89% ee (Scheme 20). This change in the stereochemical induction illustrates the essential role of the *tert*-butyl group in the geometry of the enzyme–substrate complex.



Scheme 20 Enantioselective biohydroxylation.

A recombinant Zn^{2+}-containing hydroxynitrile lyase from *Linum usitatissimum* catalyses the enantioselective cyanide addition to pivalaldehyde and hydroxypivalaldehyde with 89% and 74% ee, respectively.[95]

6. Prospects

In preparation for the 55th Annual Meeting of the American Philosophical Association Eastern Division, the philosopher and theologist Charles Hartshorne engaged himself in a reflection on the meaning of "synthesis" and more precisely on the meaning of "creative synthesis".[96] Accordingly, if synthesis is by definition "putting things together", then the art of synthesis is being able to draw on the experience of others to create new entities. In that sense the term "creative synthesis" is redundant. Nevertheless, creativity might also arise from the way you address the problem. With regard to the implementation of the *tert*-butyl group in biocatalytic processes, or more generally the generation of all-carbon quaternary centres, there are several ways of achieving this objective. From the synthetic chemist's point of view, one approach would be to understand the steric and electronic requirements that are common to the biocatalysts that accept bulky substrates, to model them, and then to improve them by means of mutations and subsequent expression. This "downstream" approach is illustrated by the examples given above (Section 5.2).

Another method, an "upstream" approach, would be to try to tackle the issue using the putative mechanisms by which Nature creates and catabolises the *tert*-butyl group. An important source of putative biocatalysts accepting sterically hindered substrates are metabolic enzymes, *e.g.* those involved in the biodegradation of methyl *tert*-butyl ether and ethyl *tert*-butyl ether.[97,98] Different enzyme classes like oxidases,[99] dehydrogenases,[97] or a putative esterase[100] have been described to be involved in the metabolism of

such compounds. Further, Smith *et al.* have reported experimental evidence for the involvement of an organic free radical in the biodegradation of n-hexane.[101] This should open a window to the search for putative biocatalysts capable of asymmetric C–C bond formation, and, thus, the generation of all-quaternary carbon centres by a free-radical mechanism.

The important role of the *tert*-butyl structural moiety has been highly appreciated in synthetic organic chemistry, but its relevance in biosynthesis and biocatalysis has not so far been fully recognized. It is fairly straightforward to predict that the combination of chemical, biosynthetic and metabolic knowledge with regard to bulky groups like the *tert*-butyl group will result in an increase in biocatalytic applications. This might enable the development of enzymes with a broad substrate range, resembling non-enzymatic catalysts. It might also enable synthetic organic chemists to gain access to exciting transformations such as selective C–H activation or asymmetric radical chemistry.

Considering the occurrence of the various families of natural products bearing *tert*-butyl groups, the question arises: why does Nature make these compounds? The occurrence of *tert*-butyl groups in natural products may induce specific geometries that are essential for further biotransformation, or (in the case of toxins) render biotransformation impossible to the predator; in the latter case, the *tert*-butyl structure could be viewed as a protecting group. The discovery and understanding of biopolymers capable of transforming *tert*-butylated substrates will certainly provide insights into – and might even provide answers to – this question.

References

1 P. J. Kociensky, *Protecting Groups* (3rd edn), Thieme, Stuttgart, 2004.
2 G. E. Douberly, A. M. Ricks, B. W. Ticknor, P. v. R. Schleyer and M. A. Duncan, *J. Am. Chem. Soc.*, 2007, **129**, 13782.
3 R. Brückner, *Advanced Organic Chemistry: Reaction Mechanisms*, Elsevier – Academic Press, Munich, 2001.
4 S. G. Cohen and A. Schneider, *J. Am. Chem. Soc.*, 1941, **63**, 3382.
5 Y. Wu and D. H. Busch, *J. Am. Chem. Soc.*, 1970, **92**, 3326.
6 G. Bartoli, M. Bosco, A. Carlone, M. Locatelli, E. Marcantoni, P. Melchiorre and L. Sambri, *Adv. Synth. Catal.*, 2006, **348**, 905.
7 R. A. Aitken and S. N. Kilényi, *Asymmetric Synthesis* (1st edn), Chapman & Hall, London, 1992.
8 T. Ohkuma, M. Kitamura and R. Noyori, in *New Frontiers in Asymmetric Catalysis*, ed. M. Mikami and M. Lautens, Wiley-VCH, Toronto, 2007.
9 I. Ojima, T. Kogure and Y. Yoda, *Org. Synth.*, 1985, **63**, 18.
10 I. D. Gridnev, Y. Yamanoi, N. Higashi, H. Tsuruta, M. Yatsutake and T. Imamoto, *Adv. Synth. Catal.*, 2001, **343**, 118.
11 K. V. L. Crépy and T. Imamoto, *Adv. Synth. Catal.*, 2003, **345**, 79.
12 D. A. Evans, F. E. Michael, J. S. Tedrow and K. R. Campos, *J. Am. Chem. Soc.*, 2003, **125**, 3534.
13 J. F. Larrow and E. N. Jacobsen, *J. Org. Chem.*, 1994, **59**, 1939.
14 A. Martinez, C. Hemmert, H. Gornitzka and B. Meunier, *J. Organomet. Chem.*, 2005, **690**, 2163.
15 C. Bolm, G. Schlingloff and K. Weickhardt, *Angew. Chem., Int. Ed. Engl.*, 1994, **33**, 1848.
16 D. A. Evans, K. R. Campos, J. S. Tedrow, F. E. Michael and M. R. Gagné, *J. Am. Chem. Soc.*, 2000, **122**, 7905.
17 J. F. Austin and D. W. C. MacMillan, *J. Am. Chem. Soc.*, 2002, **124**, 1172.
18 H. von Schenck, B. Åkermark and M. Svensson, *Organometallics*, 2002, **21**, 2248.
19 Q. Chen, B.-L. Lin, Y. Fu, L. Liu and Q.-X. Guo, *Res. Chem. Intermed.*, 2005, **31**, 759.
20 M. D. Sliger, G. A. Broker, S. T. Griffin, R. D. Rogers and K. H. Shaughnessy, *J. Organomet. Chem.*, 2005, **690**, 1478.
21 R. W. Heo, J-S. Park, J. T. Goodson, G. C. Claudio, M. Takenaga, T. A. Albright and T. R. Lee, *Tetrahedron*, 2004, **60**, 7225.
22 W. A. Nugent, *Org. Lett.*, 2002, **4**, 2133.
23 W. Steglich, E. Frauendorfer and F. Weygand, *Chem. Ber.*, 1971, **104**, 687.
24 S. Allenmark and B. Lamm, *Chirality*, 2001, **13**, 43.
25 N. J. Turner and J. R. Winterman, *Tetrahedron Lett.*, 1995, **36**, 1113.
26 K. Laumen, O. Ghisalba and K. Auer, *Biosci., Biotechnol., Biochem.*, 2001, **65**, 1977.
27 W. Hummel, M. Kuzu and B. Geueke, *Org. Lett.*, 2003, **5**, 3649.
28 A. S. Brommarius, M. Schwarm, K. Stingl, M. Kottenbahn, K. Huthmacher and K.-H. Drauz, *Tetrahedron: Asymmetry*, 1995, **6**, 2851.
29 A. S. Brommarius, K. Drauz, W. Hummel, M.-R- Kula and C. Wandrey, *Biocatalysis*, 1994, **10**, 37.
30 A. S. Brommarius, M. Schwarm and K. Drauz, *Chimia*, 2001, **55**, 50.
31 T. Hayashi, M. Fukushima, M. Konishi and M. Kumada, *Tetrahedron Lett.*, 1980, **21**, 79.
32 T. Hayashi, M. Konishi, M. Fukushima, K. Kanehira, T. Hioki and M. Kumada, *J. Org. Chem.*, 1983, **48**, 2195.
33 W. Behnen, C. Dauelsberg, S. Wallbaum and J. Martens, *Synth. Commun.*, 1992, **22**, 2143.
34 K. A. Lutomski, A. I. Meyers, in *Asymmetric Synthesis*, ed. J. D. Morisson, Academic Press, Orlando, 1984, vol. 3, pp. 213.
35 G. Helmchen, A. Krotz, K.-T. Ganz and D. Hansen, *Synlett*, 1991, 257.
36 H. Nishiyama, M. Kondo, T. Nakamura and K. Itoh, *Organometallics*, 1991, **10**, 500.
37 R. E. Löwenthal and S. Masamune, *Tetrahedron Lett.*, 1991, **32**, 7373.
38 D. A. Evans, K. A. Woerpel, M. M. Hinman and M. M. Faul, *J. Am. Chem. Soc.*, 1991, **113**, 726.
39 P. Müller, G. Bernardinelli, Y. F. Allenbach, M. Ferri and H. D. Flack, *Org. Lett.*, 2004, **6**, 1725.
40 P. Müller, F. Lacrampe and G. Bernardinelli, *Tetrahedron: Asymmetry*, 2003, **14**, 1503.
41 J. Christofers and A. Baro, *Adv. Synth. Catal.*, 2005, **347**, 1473.
42 M. Marigo, A. Kjærsgaard, K. Juhl, N. Gathergood and K. A. Jørgensen, *Chem. Eur. J.*, 2003, **9**, 2359.
43 M. S. Sigman, P. Vachal and E. N. Jacobsen, *Angew. Chem., Int. Ed.*, 2000, **39**, 1279.
44 A. G. Wenzel and E. N. Jacobsen, *J. Am. Chem. Soc.*, 2002, **124**, 12964.
45 P. Vachal and E. N. Jacobsen, *J. Am. Chem. Soc.*, 2002, **124**, 10012.
46 F. Formaggio, C. Baldini, V. Moretto, M. Crisma, B. Kaptein, Q. B. Broxterman and C. Toniolo, *Chem. Eur. J.*, 2005, **11**, 2395.
47 F. Orvieto, U. Koch, V. G. Matassa and E. Muraglia, *Bioorg. Med. Chem. Lett.*, 2003, **13**, 2745.
48 P.-O. Johansson, M. Bäck, I. Kvarnström, K. Jansson, L. Vrang, E. Hamelink, A. Hallberg, Å. Rosenquist and B. Samuelsson, *Bioorg. Med. Chem.*, 2006, **14**, 5136.
49 P. Örtqvist, S. D. Peterson, E. Åkerblom, T. Gossas, Y. A. Sabnis, R. Fransson, G. Lindeberg, U. H. Danielson, A. Karlén and A. Sandström, *Bioorg. Med. Chem.*, 2007, **15**, 1448.
50 I. C. Jacobson, P. G. Reddy, Z. R. Wasserman, F. D. Hardman, M. B. Covington, E. C. Arner, R. A. Copeland, C. P. Decicco and R. L. Magolda, *Bioorg. Med. Chem. Lett.*, 1998, **6**, 837.
51 M. Sani, D. Belotti, R. Giavazzi, W. Panzeri, A. Volonterio and M. Zanda, *Tetrahedron Lett.*, 2004, **45**, 1611.
52 M. J. Fray, R. P. Dickinson, J. P. Huggins and N. L. Occleston, *J. Med. Chem.*, 2003, **46**, 3514.
53 M. K. Gupta, P. Mishra, P. Prathipati and A. K. Saxena, *Bioorg. Med. Chem.*, 2002, **10**, 3713.
54 X. Hu, K. T. Nguyen, C. L. M. J. Verlinde, W. G. J. Hol and D. Pei, *J. Med. Chem.*, 2003, **46**, 3771.
55 B. Schmidt, *ChemBioChem*, 2003, **4**, 366.
56 C. L. Hinkle, M. J. Mohan, P. Lin, N. Yeung, F. Rasmussen, M. E. Milla and M. L. Moss, *Biochemistry*, 2003, **42**, 2127.
57 D. J. Kempf, L. M. Codacovi, D. W. Norback, J. J. Plattner, H. Sham, S. J. Wittenberger and C. Zhao, *Eur. Pat. Appl.* EP486948, 1992.
58 J. Falbe and M. Regitz, *Römpp-Chemie Lexikon* (9th edn), Thieme, Stuttgart, 1989, pp. 415.
59 However, from more than 200 compounds containing a *tert*-butyl group within Beilstein Crossfire (22nd November 2007) which are described as "Isolated Natural Products", a significant portion have been wrongly assigned in the data bank. Care has to be taken with respect to an overestimation of the still-limited diversity of *tert*-butyl-containing natural products. This holds also true for ref. 75.

60 C. Chevallier, A. D. Richardson, M. C. Edler, E. Hamel, M. K. Harper and C. M. Ireland, *Org. Lett.*, 2003, **5**, 3737.
61 E. Hamel, *Biopolymers*, 2002, **56**, 142.
62 S. Matsunaga, N. Fusetani and S. Konosu, *Tetrahedron Lett.*, 1985, **26**, 855.
63 G. Ryu, S. Matsunaga and N. Fusetani, *Tetrahedron Lett.*, 1994, **35**, 8251.
64 K. Nakanishi, *Pure Appl. Chem.*, 1967, **14**, 89.
65 K. Nakanishi, K. Habaguchi, Y. Nakadaira, M. C. Woods, M. Maruyama, R. T. Major, M. Alauddin, A. R. Patel, K. Weinges and W. Bähr, *J. Am. Chem. Soc.*, 1971, **93**, 3544.
66 L. Hu, Z. Chen, X. Cheng and Y. Xie, *Pure Appl. Chem.*, 1999, **71**, 1153.
67 S. B. Vogensen, K. Strɸmgaard, H. Shindou, S. Jaracz, M. Suehiro, S. Ishii, T. Shimizu and K. Nakanishi, *J. Med. Chem.*, 2003, **46**, 601.
68 R. B. Mujumdar, A. V. Rama, Rao, S. S. Rathi and K. Venkataraman, *Tetrahedron Lett.*, 1975, **16**, 867.
69 C. Kotake, T. Yamasaki, T. Moriyama, M. Shinoda, N. Komiyama, T. Furumai, M. Konishi and T. Oki, *J. Antibiot.*, 1992, **45**, 1442.
70 W.-S. Chan and W.-H. Hui, *J. Chem. Soc., Perkin Trans. 1*, 1973, 490.
71 X. Li and C. Djerassi, *Tetrahedron Lett.*, 1983, **24**, 665.
72 J.-L. Giner and C. Djerassi, *Phytochemistry*, 1991, **30**, 811.
73 A. Z. Abyshev, P. P. Denisenko, N. P. Kostuschenko, O. S. Anisimova, A. I. Ermakov and U. N. Sheiker, *Chem. Nat. Compd. (Engl. Transl.)*, 1970, **6**, 689.
74 G. R. Pettit, F. Gao, D. Sengupta, J. C. Coll, C. L. Herald, D. L. Doubek, J. M. Schmidt, J. R. Van Camp, J. J. Rudloe and R. A. Nieman, *Tetrahedron*, 1991, **47**, 3601.
75 V. M. Dembitsky, *Lipids*, 2006, **41**, 309.
76 K. Nakanishi and K. Habagushi, *J. Am. Chem. Soc.*, 1971, **93**, 3546.
77 F. Rohdich, K. Kis, A. Bacher and W. Eisenreich, *Curr. Opin. Chem. Biol.*, 2001, **5**, 535.
78 K. Strɸmgaard and K. Nakanishi, *Angew. Chem., Int. Ed.*, 2004, **43**, 1640.
79 J.-L. Giner, *Chem. Rev.*, 1993, **93**, 1735.
80 B. S. Moore and C. Hertweck, *Nat. Prod. Rep.*, 2002, **19**, 70.
81 E. J. Corey, R. K. Bakshi, S. Shibata, C.-P. Chen and V. K. Singh, *J. Am. Chem. Soc.*, 1987, **109**, 7925.
82 M. Nakamura, A. Hirai, M. Sogi and E. Nakamura, *J. Am. Chem. Soc.*, 1998, **120**, 5846.
83 B. Saito and T. Katsuki, *Synlett*, 2004, 1557.
84 Z. Wang, C. Zhao, M. E. Pierce and J. M. Fortunak, *Tetrahedron: Asymmetry*, 1999, **10**, 225.
85 D. J. Ramon and M. Yus, *Curr. Org. Chem.*, 2004, **8**, 149.
86 Y. Hotta, S. Ezaki, H. Atomi and T. Imanaka, *Appl. Environ. Microbiol.*, 2002, **68**, 3925.
87 S.-H. Yeo, T. Nihira and Y. Yamada, *J. Gen. Appl. Microbiol.*, 1998, **44**, 147.
88 M. Schultz, P. Hermann and H. Kunz, *Synlett*, 1992, 37.
89 M. Schmidt, E. Barbayianni, I. Fotakopoulou, M. Höhne, V. Constantinou-Kokotou, U. T. Bornscheuer and G. Kokotos, *J. Org. Chem.*, 2005, **70**, 3737.
90 R. Kourist, P. D. de María and U. T. Bornscheuer, *ChemBioChem*, 2008, **9**, 491.
91 D. Zhu and L. Hua, *J. Org. Chem.*, 2006, **71**, 9484.
92 J. R. Dehli and V. Gotor, *Tetrahedron: Asymmetry*, 2000, **11**, 3693.
93 A. de Raadt, B. Fetz, H. Griengl, M. F. Klingler, I. Kopper, B. Krenn, D. F. Münzer, R. G. Ott, P. Plachota, H. J. Weber, G. Braunegg, W. Mosler and R. Saf, *Eur. J. Org. Chem.*, 2000, 3835.
94 D. F. Münzer, H. Griengl, A. Moumtzi, R. Saf, T. Terzani and A. de Raadt, *Eur. J. Org. Chem.*, 2005, 793.
95 K. Trummler, J. Roos, U. Schwaneberg, F. Effenberger, S. Förster, K. Pfizenmaier and H. Wajant, *Plant Sci.*, 1998, **139**, 19.
96 C. Hartshorne, *J. Philos.*, 1955, **55**, 944.
97 N. Lopes, Ferreira, C. Malandain and F. Fayolle-Guichard, *Appl. Microbiol. Biotechnol.*, 2006, **72**, 252.
98 M. M. Häggblom, L. K. G. Youngster, P. Somsamak and H. H. Richnow, *Adv. Appl. Microbiol.*, 2007, **62**, 1.
99 G. Schmitz, D. Franke, S. Stevens, R. Takors, D. Weuster-Botz and C. Wandrey, *J. Mol. Catal. B: Enzym.*, 2000, **10**, 313.
100 C. A. Smith, K. T. O'Reilly and M. R. Hyman, *Appl. Environ. Microbiol.*, 2003, **69**, 796.
101 R. Fabus, H. Wilkes, A. Behrends, A. Armstroff, T. Fischer, A. J. Pierik and F. Widdel, *J. Bacteriol.*, 2001, **183**, 1707.