Enzyme-catalysed Baeyer–Villiger Oxidations of Some Substituted Bicyclo[3.2.0]heptanones

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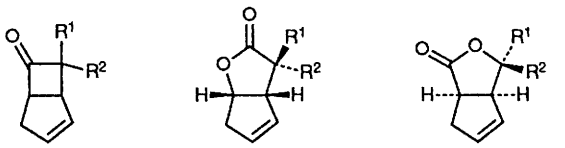
The ketones **1** and **4** are oxidized to γ-lactones **2** and **5** with low enantioselectivity by both *Acinetobacter* sp. NCIB 9871 and *Pseudomonas* sp. NCIB 9872; in contrast the ketones **7** and **10** are biotransformed to products **8/9** and **11** respectively of very high optical purity.

There is considerable current interest in the oxidation of cyclic ketones to lactones using monooxygenase enzymes as catalysts.[1] The enzymes can be used as (partially) purified protein, in which case cofactor recycling must be organized, or as part of a whole cell system.[2] Recently Furstoss reported that the racemic ketone **1** is oxidized by the bacterium TD63 to give equimolar quantities of the lactones **2** and **3** in states of high optical purity.[3] In this paper we show that the readily available organisms *Pseudomonas* sp. NCIB 9872 and *Acinetobacter* sp. NCIB 9871 promote oxidation of the ketone **1** and also selected derivatives of compound **1** with quite different results.

Oxidation of the ketone **1** using the *Pseudomonas* sp. gave the (1*S*,5*R*)-lactone **2** of low optical purity {23% enantiomeric excess (e.e.); the assessments of the ratio of enantiomers, in this case and in the other instances that are quoted below, were made by NMR spectroscopy using the shift reagent tris-[3-(heptafluoropropylhydroxymethylene)-(+)-camphorato]europium(III)} in 75% yield; the lactone **2** was contaminated with a small amount (5%) of the lactone **3**. Similarly oxidation of the ketone **1** with the *Acinetobacter* sp. gave the lactones **2** and **3**, in the ratio 9 : 1, in a modest yield (41%). The regioselectivity of these enzyme-catalysed oxidation reactions is very similar to that observed for the peracetic acid oxidation of the ketone **1** which furnishes the lactones **2** and **3** in the ratio 13 : 1.

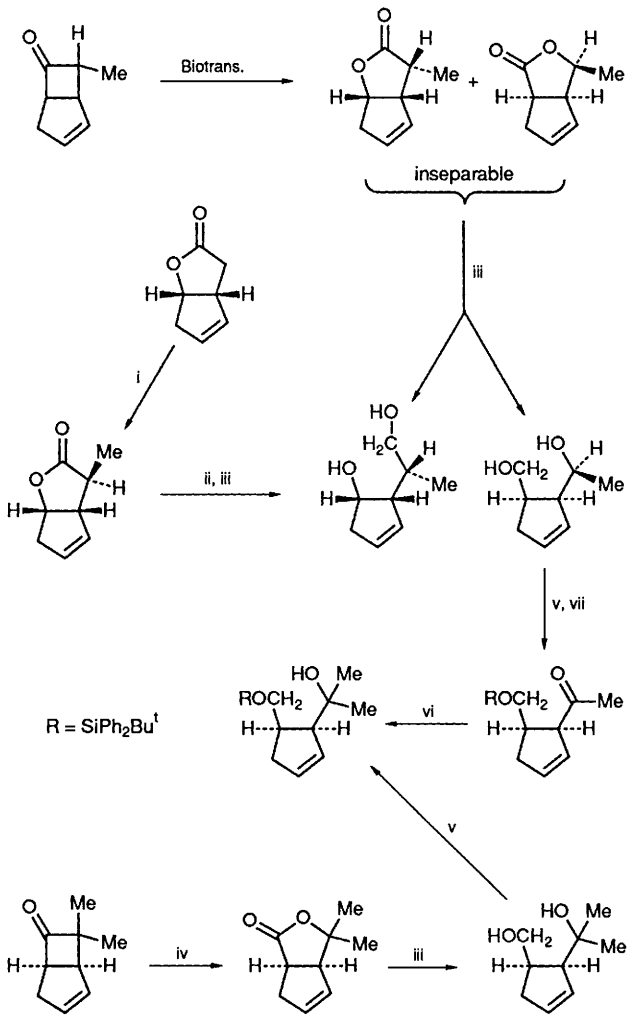
7,7-Dimethylbicyclo[3.2.0]hept-2-en-6-one **4**[4] is oxidized by peracetic acid and by the *Acinetobacter* sp. to give exclusively the lactone **5**. The enantioselectivity of the enzyme-catalysed oxidation was low, the (1*R*,5*S*)-product† being obtained in 63% yield (29% e.e.). The *Pseudomonas* sp. catalysed oxidation of the ketone **4** gave the lactones **5** and **6** in the ratio 20 : 1 (41% yield). More interestingly oxidation of 6-*endo*-methylbicycloheptenone **7**[5] using either the *Pseudomonas* sp. or the *Acinetobacter* sp. produced optically pure (e.e. >96%) lactones **8** and **9** in equal quantities (combined yields 50–55%). The absolute configurations of the compounds **8** and **9** were determined by correlation with compounds of known structure (Scheme 1).

Despite the partial purification of only a single chromosomal DNA-coded Baeyer–Villiger type monooxygenase from both *Pseudomonas* sp. NCIB 9872[6] and *Acinetobacter* sp. NCIB 9871,[7,8] these results suggest that there might be two active monooxygenase enzymes in both the whole cell systems, one with a proclivity to produce the 2-oxabicycloocten-3-one and the second protein which produces the 3-oxabicycloocten-2-one. The first enzyme preferentially biotransforms the (5*S*)-ketone while the second enzyme accepts the (5*R*)-ketone as the preferred substrate. Neither enzyme is strictly enantioselective and if the two modes of ring expansion are not energetically equivalent then both enantiomers of the

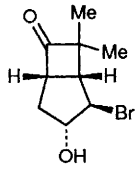


1; $R^1 = R^2 = H$ **2**; $R^1 = R^2 = H$ **3**; $R^1 = R^2 = H$
4; $R^1 = R^2 = Me$ **6**; $R^1 = R^2 = Me$ **5**; $R^1 = R^2 = Me$
7; $R^1 = H$; $R^2 = Me$ **8**; $R^1 = H$; $R^2 = Me$ **9**; $R^1 = H$; $R^2 = Me$

† The absolute configuration of the lactone **5** was determined by oxidation of the (1*R*,5*S*)-ketone **4**[4] using peracetic acid.



Scheme 1 *Reagents and conditions*: i, $(Me_3Si)_2NLi$, tetrahydrofuran (THF), −65 °C, 0.5 h then MeI, −65 to −20 °C, 1.5 h; ii, $(Me_3Si)_2NLi$, THF, −65 °C, 0.5 h then MeOH, −65 to −20 °C, 1.5 h; iii, $LiAlH_4$, ether, 0.5 h; iv, H_2O_2, HOAc, 0 °C, 48 h; v, Ph_2Bu^tSiCl, imidazole, CH_2Cl_2, 0.5 h; vi, MeLi, Et_2O, 0.5 h; vii, Swern oxidation



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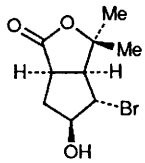

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ketone are carried through by one of the proteins to provide an enantiomerically enriched mixture of a single lactone. Further evidence to support this suggestion is currently being sought.

It is not surprising that better enantioselectivity in this system can be achieved by appending additional substituents to the bicyclic system. For example the racemic bromohydrin **10**[9] is converted by the *Acinetobacter* sp. over 7 h into equal quantities of the (1*R*,5*S*)-ketone **10** (e.e. 85%) and the lactone **11** (>98% e.e.) in 61% yield. It seems the enzyme producing the 3-oxabicycloalkan-2-one is unable to accommodate the less-favoured (5*S*)-ketone in this case.

Obviously biotransformations leading to homochiral compounds of the types **8**, **9** and **11** possessing three or four contiguous asymmetric carbon centres has important implications in synthetic organic chemistry. The simple experimental procedure required to perform these biotransformations is also noteworthy. Thus, 6 l of the appropriate basal medium[6] was innoculated with *Pseudomonas* NCIB 9872 or *Acinetobacter* NCIB 9871. The microorganism was grown until the suspension had an optical density of 0.95; the cells were then harvested by centrifugation (3800 rpm at 30 °C for 10 min) and resuspended in phosphate buffer (350 ml, pH 7.2). The substrate (*ca.* 200 mg) was added to the cell suspension (350 ml) with shaking. After complete disappearance of the starting material (GC analysis using 1 ml aliquots of solution after centrifugation), the cells were spun down by centrifugation (3800 rpm for 30 min) and the supernatant liquid was extracted with dichloromethane or ethyl acetate. After drying, evaporation of the solvent yielded the product.

We thank the SERC/DTI (Biotransformations LINK Scheme) for a research assistantship (to A. J. C.).

Received, 27th June 1990; Com. 0/02900I

References

1 *Biotransformations in Preparative Organic Chemistry: the Use of Isolated Enzymes and Whole Cell Systems in Synthesis*, H. G. Davies, R. H. Green, D. R. Kelly and S. M. Roberts, Academic Press, London, 1989, pp. 166–169.
2 B. P. Branchaud and C. T. Walsh, *J. Am. Chem. Soc.*, 1985, **107**, 2153; M. J. Taschner and D. J. Black, *J. Am. Chem. Soc.*, 1988, **110**, 6892; O. Abril, C. C. Ryerson, C. T. Walsh and G. M. Whitesides, *Bioorg. Chem.*, 1989, **17**, 41.
3 V. Alphand, A. Archelas and R. Furstoss, *Tetrahedron Lett.*, 1989, **30**, 3663.
4 S. Butt, H. G. Davies, M. J. Dawson, G. C. Lawrence, J. Leaver, S. M. Roberts, M. K. Turner, B. J. Wakefield, W. F. Wall and J. A. Winders, *J. Chem. Soc., Perkin Trans. 1*, 1987, 903.
5 M. Rey, S. M. Roberts, A. Dieffenbacher and A. S. Drieding, *Helv. Chim. Acta*, 1970, **53**, 417.
6 M. Griffin and P. W. Trudgill, *Eur. J. Biochem.*, 1976, **63**, 199.
7 N. A. Donoghue, D. B. Norris and P. W. Trudgill, *Eur. J. Biochem.*, 1976, **63**, 175; Y.-C. J. Chen, O. P. Peoples and C. T. Walsh, *J. Bacteriol.*, 1988, **170**, 781.
8 Y-C. J. Chen, O. P. Peoples and C. T. Walsh, *J. Bacteriol.*, 1988, **170**, 781.
9 Z. Grudzinski and S. M. Roberts, *J. Chem. Soc., Perkin Trans, 1*, 1975, 1767.