LIPASE-MEDIATED ROUTE TO DIASTEREO-PURE TRANEXAMIC ACID

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Diastereomerically pure tranexamic acid has been prepared *via* a diastereomeric separation of a *trans-/cis*-mixture of 1,4-cyclohexanedimethanol using lipase PS (*Pseudomonas* sp. Amano).

KEY WORDS tranexamic acid; diastereomeric separation; lipase-mediated transesterification; lipase-mediated hydrolysis; antiplasmic activity

Tranexamic acid, *trans*-4-aminomethylcyclohexanecarboxylic acid (**1**), is a clinically important compound being used as a potent antiplasmin agent.[1] Because its synthesis owes to hydrogenation of an aromatic precursor,[2] it is always accompanied by the inseparable *cis*-diastereomer which has been found to be 50 times less effective.[1a,b] We wish to report herewith a diastereoselective route to pure tranexamic acid (**1**) from a *trans-/cis*-mixture of 1,4-cyclohexanedimethanol (t/c-**2**) *via* a lipase-mediated diastereomeric separation.[3]

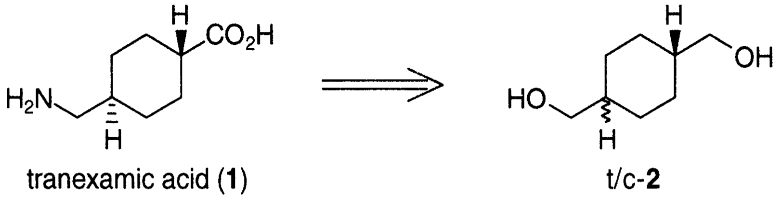

Chart 1

Recently, a base-induced equilibrium between *trans*- and *cis*-1,4-cyclohexanedimethanols (**2**) was reported to give a *trans*-enriched mixture.[4] This led us to anticipate that the commercially available *trans*/*cis*-mixture (t/c-**2**) will be a vital precursor for pure tranexamic acid (**1**) if the mixture is readily separable after the equilibrium. Thus, we first examined its separation by application of the enzymatic transesterification procedure in an organic solvent.[5] The mixture (t/c-**2**) (t/c=70:30), on reaction with vinyl acetate in *tert*-butyl methyl ether in the presence of lipase PS (*Pseudomonas* sp., Amano), furnished a readily separable mixture of the *trans*-enriched diacetate (t-**3**) (t/c=94:6) and the *cis*-enriched monoacetate (**4**) (t/c=17:83) in 74 and 10% yields, leaving the pure *cis*-diol (c-**2**) in 15% yield after 48 h.[6] The pure *trans*-diacetate (t-**3**) could be obtained in 77% yield, accompanied by the monoacetate mixture (**4**) (t/c=63:37) (13%) and the diol mixture (**2**) (t/c=48:52) (5%), by a single repetition of the same enzymatic treatment on the *trans*-enriched diol (t-**2**) obtained from *trans*-enriched diacetate (t-**3**). Overall yield of the pure *trans*-diol (t-**2**) from the starting diastereomeric diol (t/c-**2**) after a twice-repeated

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enzymatic operation was 57%. The pure *trans*-acetate (t-**3**) thus obtained afforded pure *trans*-diol (t-**2**) in quantitative yield on methanolysis in the presence of potassium carbonate at room temperature.

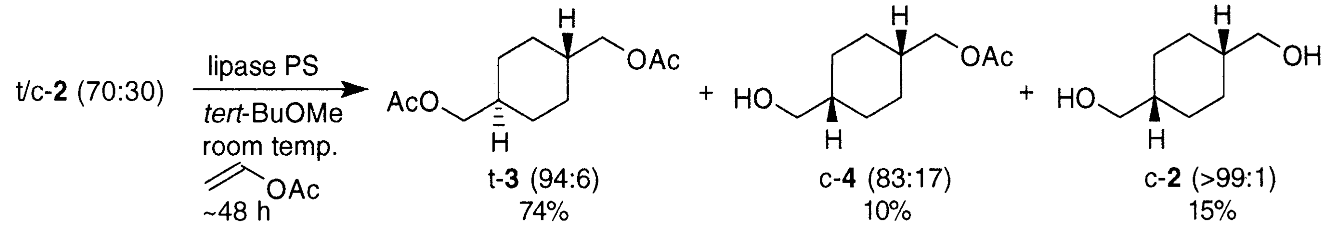

Chart 2

We next examined the lipase-mediated hydrolysis of the diacetate mixture (t/c-**3**) in aqueous condition. It is well recognized that when the same lipase is used, both the transesterification and the hydrolysis occur at the same chiral center.7) The same is true for the diastereomeric mixture (t/c-**3**) (*trans*/*cis*=70:30), which afforded the pure *trans*-diol (t-**2**) in 66% yield accompanied by 31% yield of the *cis*-enriched monoacetate (c-**4**) (*trans*/*cis*=16:84) in the presence of lipase PS in a phosphate buffer solution,8) though the process took 7 days. Thus, it was concluded that hydrolytic conditions were more appropriate for the separation of pure *trans*-diol (t-**2**).

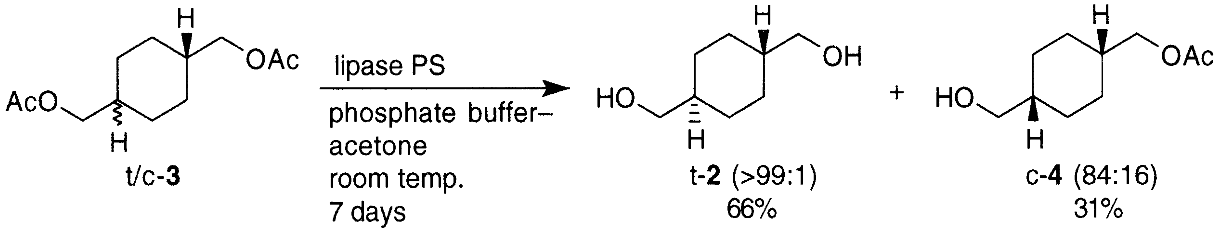

Chart 3

The pure *cis*-diol (c-**2**) as well as the mixture diol (t/c-**2**) recovered from the mixture acetates (t/c-**3**) and (t/c-**4**) could be equilibrated to the *trans*-enriched mixture (t/c=~4:1), which may be recycled. Thus, the pure *cis*-diol (c-**2**) was fused at 180 °C with sodium hydroxide (0.1 eq) for 15 min4) to furnish a mixture consisting of 82 parts of the *trans*-**2** and 18 parts of the *cis*-**2** in 92% yield after direct distillation (~165 °C/5 Torr) from a reaction flask.

Having obtained pure *trans*-1,4-cyclohexanedimethanol (t-**2**), we converted it to tranexamic acid (**1**). To minimize ditosylation, the *trans*-diol (t-**2**) was treated with 0.7 eq of *p*-toluenesulfonyl chloride to afford the monotosylate (**5**) in 47% yield (84% based on the consumed **2**) with recovery of 44% of the unchanged **2**, which was recycled. Treatment of the tosylate (**5**) with sodium azide gave the azide (**6**), in 93% yield, which on Jones oxidation afforded the carboxylic acid (**7**) in 87% yield. Finally, the azide

(**7**) was hydrogenated on 10% palladium-on-charcoal in 1% hydrochloric acid to give pure tranexamic acid (**1**) as hydrochloride[1] in 70% yield after purification by recrystallization.

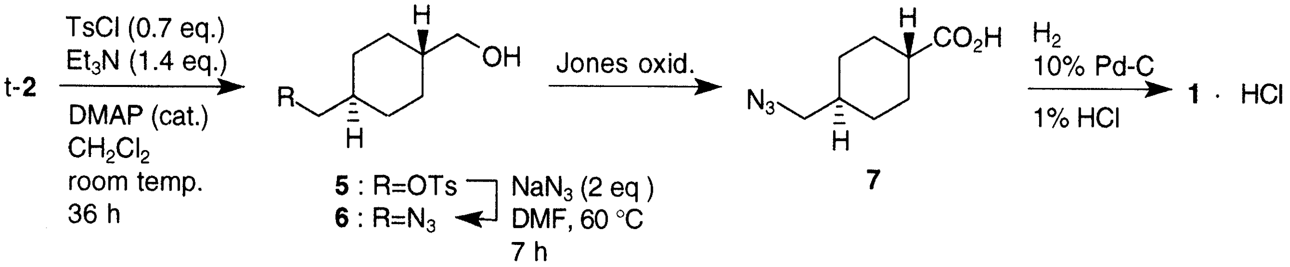

Chart 4

In conclusion, the present investigation has established a route to diastereomerically pure tranexamic acid (**1**) starting from a diastereomeric mixture of *trans*- and *cis*-1,4-cyclohexanedimethanol (t/c-**2**) formally without loss of the *cis*-component, employing a lipase-mediated separation and a base-induced equilibration.

REFERENCES AND NOTES

1) a) M. Shimizu, T. Naito, A. Okano, T. Aoyagi, *Chem. Pharm. Bull.*, **13**, 1012 (1965); b) T. Naito, A. Okano, S. Kadoya, T. Miki, M. Inaoka, R. Moroi, M. Shimizu, *ibid.*, **16**, 357, 728 (1968).

2) M. Levine, R. Sedlecky, *J. Org. Chem.*, **24**, 115 (1959).

3) A description of a lipase-mediated separation of a 1,4-cyclohexanedimethanol mixture (t/c-**2**) by acylation in an organic solvent appeared while the present investigation was going on. However, neither lipase PS nor hydrolytic conditions were employed. See S. Geresh, E. Elbaz, R. Glaser, *Tetrahedron*, **49**, 4939 (1993).

4) Y. Tatsuno, S. Yoneda, N. Okamoto, Y. Ishii, K. Kato, Japan Kokai, 2-131442 (1990).

5) Cf. A. M. Klibanov, *Acc. Chem. Res.*, **23**, 114 (1990).

6) The reaction was carried out in *tert*-butyl methyl ether (10 ml/mmol of t/c-**2**) in the presence of lipase-PS-on-Celite (10 mg/mmol of t/c-**2**) at room temperature and separated by silica gel column chromatography. The *trans*/*cis* ratio was determined by ^{1}H NMR analysis (300 MHz) of the diacetate (**3**): t-**3** exhibits $AcOC\underline{H_2}$- at δ 3.46, while the c-**3** exhibits them at δ 3.55.

7) For examples, K. Laumen, D. Breitgoff, M. P. Schneider, *J. Chem. Soc., Chem. Commun.*, **1988**, 1459; S. Takano, T. Yamane, M. Takahashi, K. Ogasawara, *Tetrahedron: Asymmetry*, **3**, 837 (1992); S. Takano, O. Yamada, H. Iida, K. Ogasawara, *Synthesis*, **1994**, 592.

8) The reaction was carried out in a mixture of 0.2 M phosphate buffer and acetone (9:1) (10 ml/mmol of **3**) in the presence of lipase PS-on-Celite (10 mg/mmol of **3**) and separated by silica gel column chromatography.

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