# Carbon-13 Labelling Study of the Methylitaconate ⇄ α-Methyleneglutarate Model Rearrangement Reaction

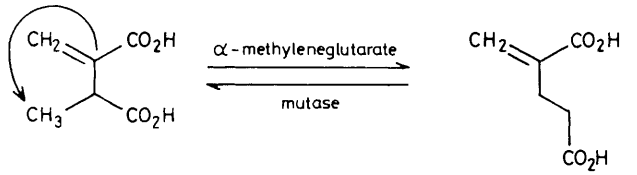
**Paul Dowd* and Roger Hershline**

*Department of Chemistry, University of Pittsburgh, Pittsburgh, PA 15260, U.S.A.*

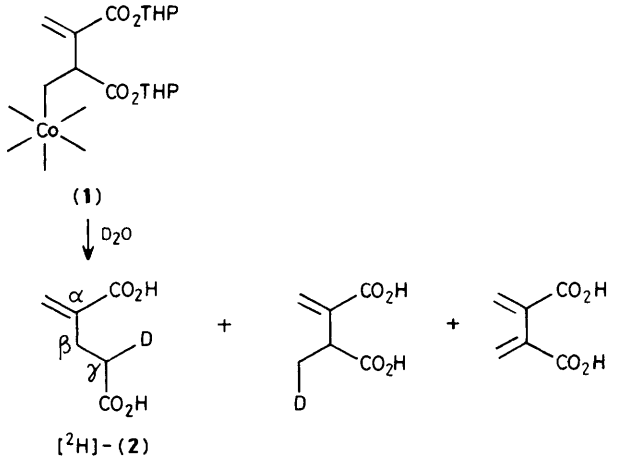
The model rearrangement mimicking the coenzyme $B_{12}$-dependent, enzyme-catalysed interconversion of α-methyleneglutaric acid with methylitaconic acid has been carried out with a carbon-13 label demonstrating beyond question that the acrylate group is the migrating group in the model as it is in the enzyme-catalysed rearrangement.

One of the most interesting of the coenzyme $B_{12}$-dependent enzyme-catalysed rearrangements[1] is the reversible carbon skeleton rearrangement of methylitaconic acid with α-methyleneglutaric acid (Scheme 1).[2] This is the key step in the metabolic breakdown of nicotinic acid by the anaerobic bacterium *Clostridium barkerii*.[2] Examination of the course of the reaction sequence using [5-$^{14}$C]nicotinic acid revealed that the *acrylate* is the migrating group in the enzymic rearrangement (Scheme 1).[2]

Several years ago, we discovered a nonenzymic model reaction† which mimics the enzyme-catalysed rearrangement.[3] When the model rearrangement (Scheme 2) was carried out in $D_2O$, deuterium was incorporated at the γ-position of the rearrangement product α-methyleneglutaric acid[4] [$^2$H]-(**2**). The conclusion was drawn[4] that the *acrylate* is probably the migrating group as it is in the enzyme-catalysed reaction. This conclusion was well founded only insofar as the rearrangement is straightforward as shown in Scheme 3(a). It

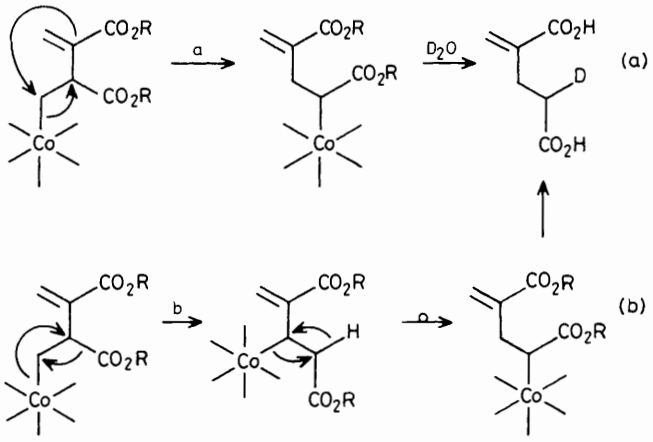



**Scheme 1**

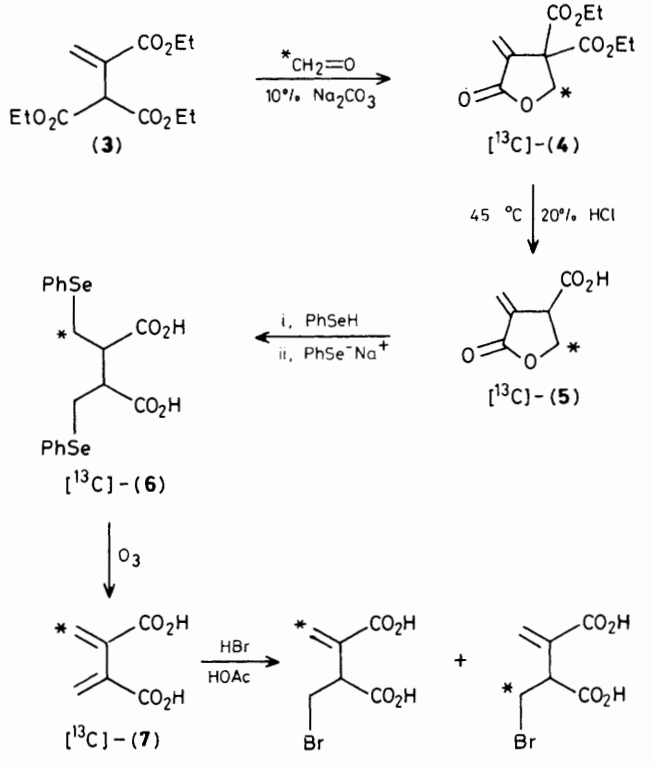



**Scheme 2.** THP = tetrahydropyran-2-yl.

† In all the model reactions described in this paper we used vitamin $B_{12}$. Accordingly, the >Co< symbol indicates the presence of the full vitamin $B_{12}$ nucleus.



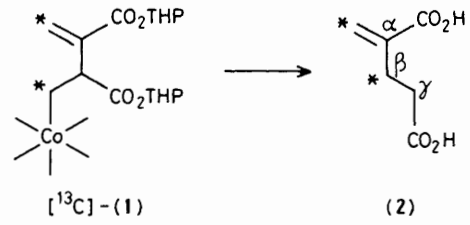
Scheme 3. R = THP.



Scheme 4

was recognised,[4] but not ruled out, that there exists another possibility. If the *carboxy group* were the migrating group, and if carboxy migration were followed by an interchange of hydrogen with cobalt then the deuterium labelling result would be the *same* as that observed [Scheme 3(b)].

Since the model rearrangement (Scheme 2) is important to our understanding of the workings of the carbon skeleton rearrangements, it is worthwhile to establish the sense of the rearrangement with finality. Accordingly, we have prepared the carbon-13 labelled model compound [$^{13}$C]-(**1**), enriched at the vinyl methylene group and at the methylene group attached to cobalt. The synthesis was carried out as outlined in Scheme 4. Thus, treatment of the triester (**3**)[5] with 90% $^{13}$C-enriched paraformaldehyde in the presence of base yielded the lactone [$^{13}$C]-(**4**). Hydrolysis and decarboxylation



gave the labelled lactone acid [$^{13}$C]-(**5**), which was opened by successive treatment with phenyl selenol and phenyl selenide. Oxidative elimination gave the dicarboxylic acid [$^{13}$C]-(**7**), hydrogen bromide addition to which then yielded a mixture of [1-$^{13}$C] and [4-$^{13}$C]-1-bromomethylitaconic acid from which the model [$^{13}$C]-(**1**) was prepared.[3] The $^{13}$C n.m.r. spectrum of the model [$^{13}$C]-(**1**) showed a vinyl carbon triplet ($J$ 159 Hz) at δ 126 and two methylene triplets ($J$ 142 Hz) at δ 27.4 and 27.0. The latter correspond to the diastereoisomeric mixture produced when the carbon–cobalt bond of [$^{13}$C]-(**1**) is formed. The chemical shift of the cobalt-bound methylene group compares favourably to that, δ 24.3, observed for the 5′-methylene group in the $^{13}$C n.m.r. spectrum of coenzyme $B_{12}$.[6]

The model [$^{13}$C]-(**1**) was allowed to decompose, in aqueous solution, in the dark, at room temperature and at pH 8.3, and then the rearrangement product α-methyleneglutaric acid (**2**) was isolated. The proton-decoupled $^{13}$C n.m.r. spectrum of (**2**) showed a vinyl methylene carbon singlet at δ 126 and a β-methylene carbon singlet at δ 28.0. No resonance was observed at δ 33.0, the position of absorption of the γ-carbon. We conclude that the *acrylate* is the exclusive migrating group in the model rearrangement as it is in the enzyme-catalysed transformation.

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