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Stereoelectronic Effects Dictate Mechanistic Dichotomy between Cu(II)-Catalyzed and Enzyme-Catalyzed Reactions of Malonic Acid Half Thioesters

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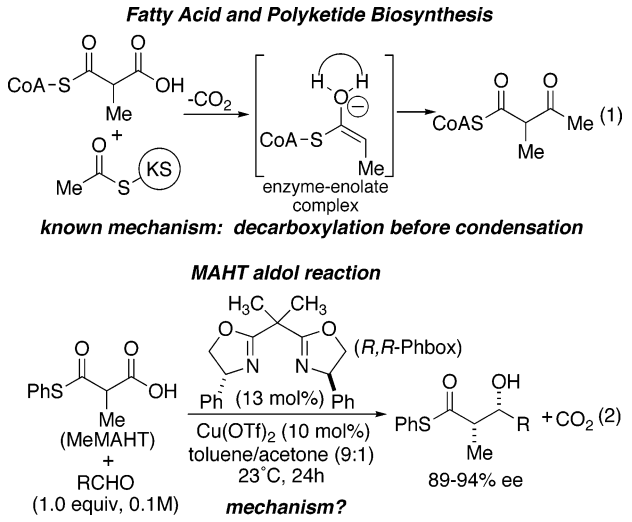
In the biosynthesis of polyketides and fatty acids,[1] enzymatic activation of malonic acid half thioesters (MAHTs) affords ester enolates that condense with thioesters resulting in a decarboxylative Claisen condensation (Scheme 1, eq 1). Inspired by this reaction, we developed a Cu(II)-catalyzed, enantioselective reaction[2] between MeMAHT and aldehydes yielding aldol products resulting from formal decarboxylation and enolate addition to the aldehydes (Scheme 1, eq 2). In principle, ester enolate equivalents can be generated from MAHTs by either decarboxylation or deprotonation, while the accepted mechanism[3] for the enzyme-catalyzed reactions involves decarboxylation to form thioester enolates followed by condensation with thioesters (Scheme 1, eq 1).

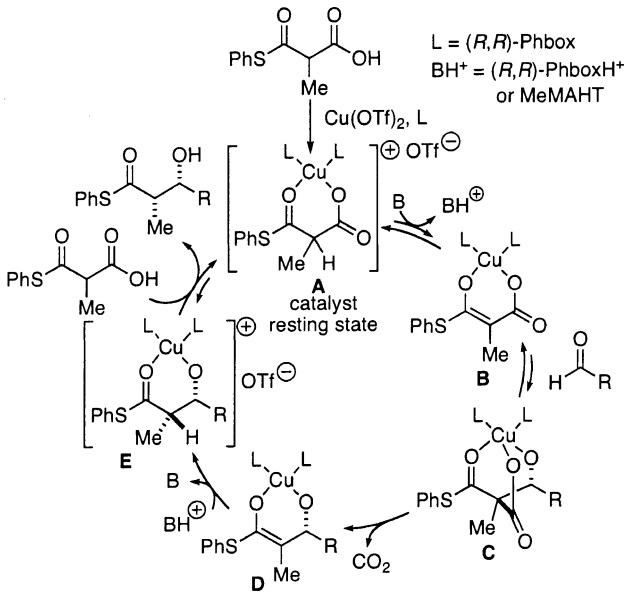
Scheme 1



Below we report evidence based on steric effects, kinetics, kinetic isotope effects (KIEs), and crossover experiments in support of a mechanism for the Cu(II)-catalyzed aldol reaction of MAHTs involving decarboxylation *after* addition to aldehydes (Scheme 2). We also provide an explanation, based on stereoelectronic effects, for the mechanistic dichotomy between Cu(II)-catalyzed and enzyme-catalyzed reactions of MAHTs.

Kinetics experiments support a transition state composed of Cu(II), (*R*,*R*)-Phbox, MeMAHT, and aldehyde. The reaction between dihydrocinnamaldehyde and a mixture of $Cu(OTf)_2$, (*R*,*R*)-Phbox, and MeMAHT in an optimized, fixed ratio of 10:13:100 was kinetically first-order in the $Cu(OTf)_2$, (*R*,*R*)-Phbox, and MeMAHT mixture taken as a whole. No reaction occurred when the (*R*,*R*)-Phbox/$Cu(OTf)_2$ ratio was equal to one, suggesting that (*R*,*R*)-Phbox serves as both a ligand and a catalytic base (Scheme 2, **A** → **B**). The reaction was first-order in aldehyde at [aldehyde] ≤ 0.2 M, but the rate decreased from that predicted by first-order kinetics at higher concentrations. These results, combined with our observations[2] that the reaction rate is highly sensitive to sterics of both the aldehyde and MAHT and that decarboxylation does not occur in the absence of aldehyde, suggest that enolization occurs by nonrate-limiting deprotonation (**A** → **B**). This precludes a mechanism mirroring the enzymatic reaction of MAHTs, involving rate-limiting decarboxylation.

Scheme 2



To further our study of MeMAHT enolization, we synthesized two isotopically labeled MeMAHTs: MeMAHT deuterated at the α-position (MeMAHT-D) and a 1:1 mixture of MeMAHT with a ^{13}C label at the carboxylate position and at the thioester position but containing no deuterium (MeMAHT-^{13}C) (see Scheme 3). We measured an enolization rate of $(5.2 \pm 0.2) \times 10^{-5}$ M/s ($t_{1/2} = 5.5 \pm 0.2$ min) by observing the rate of deuterium exchange between MeMAHT-D and MeMAHT-^{13}C under the aldol conditions. Enolization is therefore too fast to be rate-limiting under standard aldol conditions but is slow enough to be partially rate-limiting when [aldehyde] > 0.2 M. A ^1H NMR spectrum of all the reaction components exhibited two broad methyl peaks for MeMAHT when (*R*,*R*)-Phbox was used and only one peak when (±)-Phbox was used. We assign the two peaks when using (*R*,*R*)-Phbox to diastereomeric complexes formed between (*R*,*R*)-Phbox/Cu(II) and (±)-MeMAHT in rapid equilibrium with free MeMAHT[4a] indicating that the coordinated MeMAHT is not enolized in the catalyst resting state[5] (Scheme 2, **A**).

We measured an isotope effect of $k_H/k_D = 1.06 \pm 0.02$ on the rate of an aldol reaction using unlabeled MeMAHT compared to MeMAHT-D; however, a 1:1 mixture of these MAHTs produced an aldol product (Scheme 2, **E**) with a 6:1 ratio of hydrogen to

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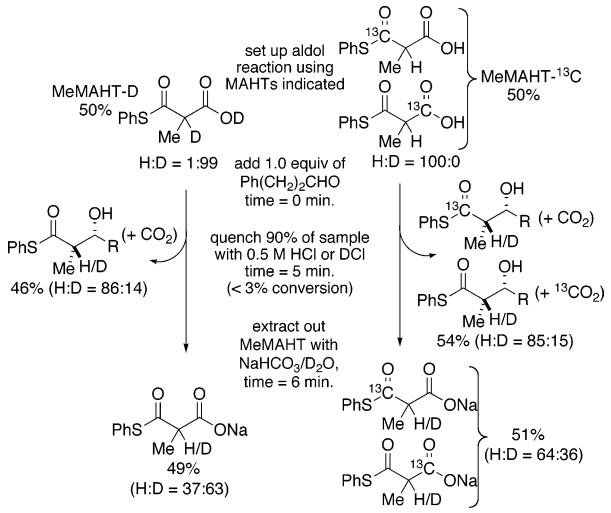
Scheme 3



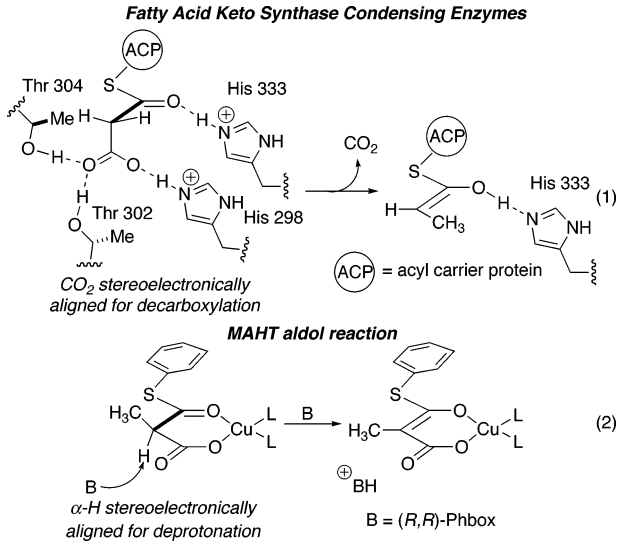
deuterium at the α-position at <3% conversion. This result indicates that the elementary step responsible for the H:D ratio in the product occurs after the rate-limiting step and is likely a protonation step late in the reaction pathway (Scheme 2, **D** → **E**).

To determine whether deprotonative enolization is essential rather than incidental to the aldol reaction, we carried out an aldol reaction with 50% MeMAHT-D and 50% MeMAHT-^{13}C. We removed 90% of the reaction mixture from the reaction vessel and isolated the product after a reaction time of 5 min (<3% conversion). The MeMAHT remaining in the reaction vessel was reisolated 1 min later (Scheme 3). If the mechanism involved decarboxylation followed by addition to the aldehyde, then the original isotope pattern in the starting materials would be largely retained in the products; however, ^{1}H NMR analysis[4b] showed that the isotopic labeling is *completely* scrambled in the product. The scrambling cannot occur after the reaction because we have observed that the product is configurationally stable to the reaction conditions, nor does complete scrambling occur prior to the reaction because the reisolated MeMAHT is not yet completely scrambled. The only possibility is that scrambling by deprotonation and reprotonation occurs during the aldol reaction itself. Moreover, the equal H/D ratios in the products indicate that the labeled MeMAHTs proceed through essentially identical intermediates (i.e., Scheme 2, **B**, **C**, and **D**) each having lost its isotopic distinction partway through the reaction. This is consistent with our proposed mechanism (Scheme 2) of deprotonative enolization, addition to the aldehyde, decarboxylation, and protonation of the β-hydroxy enolate.

Additional information about the reversibility of the elementary steps was obtained by measuring the ^{13}C KIE at the carboxylate carbon of MeMAHT-^{13}C in an aldol reaction with dihydrocinnamaldehyde. We measured a KIE of 1.020 ± 0.002 using a modification of the Singleton method[5] which is consistent with a scenario in which the MeMAHT enolate (Scheme 2, **B**) adds reversibly to the aldehyde and then decarboxylates.[6] Furthermore, MeMAHT-^{13}C reisolated at high conversion was found not to have formed a statistical mixture of isotopic isomers indicating that decarboxylation is irreversible.

Our results demonstrate that Cu(II)-catalyzed aldol reactions of MAHTs occur by a different mechanism from enzyme-catalyzed decarboxylative Claisen condensations of MAHTs. Gerlt and

Scheme 4



Holden[8] proposed that enzymes activate MAHTs by polarizing the thioester group while orienting the carboxylate orthogonally to enforce overlap between the σ orbital of the scissile C−C bond and π^* of the thioester carbonyl, which is stereoelectronically required for decarboxylation (Scheme 4, eq 1).[9] Deprotonation is prevented because if the MAHT is stereoelectronically aligned for decarboxylation, it cannot be aligned for deprotonation. We believe that in our aldol reaction, bidentate coordination of MAHT by Cu(II) orients the C-2 proton orthogonal to the π system, allowing deprotonation but not decarboxylation (Scheme 4, eq 2). The aldol reaction mechanism (Scheme 2) also explains why this reaction is compatible with protic functional groups: because the only strongly basic intermediates (i.e., Scheme 2, **D**) are generated in small concentrations late in the catalytic cycle.

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Supporting Information Available: Representative experimental procedures and characterization data. This material is available free of charge via the Internet at http://pubs.acs.org.

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(4) See Supporting Information for further details: (a) S30−S35; (b) S44−S49.
(5) We believe that the resting state catalyst binds reversibly to a second MeMAHT since the aldol reaction is inverse first order in [MeMAHT].
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(9) The reactant in Scheme 4, eq. 1 is based on a study of an MAHT modeled into the active site of KAS I from the following reference: Olsen, J. G.; Kadziola, A.; von Wettstein-Knowles, P.; Siggaard-Andersen, M.; Larsen, S. *Structure* **2001**, *9*, 233−243.

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