

Flavocytochrome b_2 (Fcb2) is a flavin-dependent redox enzyme that catalyzes the oxidation of L-lactate to pyruvate. The mechanism of this enzymatic reaction, in particular the lactate $C_{\alpha}-H_{\alpha}$ bond cleavage step, is still debated.

Relevant physicochemical properties of L-lactate, a substrate of Fcb2, have been calculated both in the vacuum and in an active-site model of the enzyme.

The results indicate that negative hyperconjugation effects in L-lactate are a key factor for the determination of the Fcb2-catalyzed L-lactate-to-pyruvate dehydrogenation mechanism. Indeed, these effects, responsible for the $C_{\alpha}-H_{\alpha}$ bond weakening in the isolated L-lactate molecule, are enhanced when L-lactate hydroxyl group is involved in a hydrogen bond with the active site base of the Fcb2 enzyme in the Michaelis complex.

Negative hyperconjugation effects (also known as Bolhmann effects or $n-\sigma^*$ interactions), which are also present in the substrates of other flavin-dependent enzymes, may be of general relevance in understanding the mechanism of dehydrogenation reactions catalyzed by flavoenzymes.

The picture shows the negative hyperconjugation interaction between the lone pair on the oxygen atom and the antibonding orbital $\sigma^*(C_{\alpha}-H_{\alpha})$ of the L-lactate molecule.

