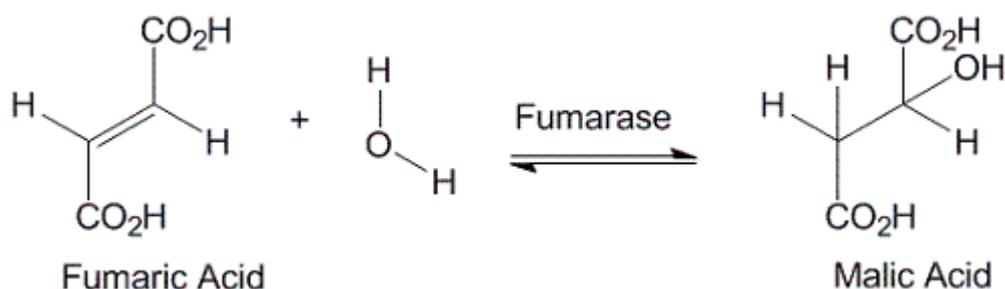


The Enzyme Catalysed Hydration of Fumaric Acid to Malic Acid

by Scott G. Stewart

Experiment Overview

This experiment involves the reaction of water with fumaric acid to form malic acid. The analogous experiment with D_2O may be followed by 1H NMR, through either *syn* or *anti* addition across the double bond to provide two possible diastereoisomers. Through the 1H NMR the coupling constant of vicinal protons in the product, the vicinal Karplus correlation diagram (pictorial representation) is used to predict which isomer is formed.



Learning Experience

Combines both NMR theory and enzymatic processes together to give the student a well rounded hands on approach to this subject matter which connects physical and biological chemistry.

Aims and Objectives

To make students aware of:

- 1H NMR techniques / theory;
- enzymatic reactions / processes;
- formation of isomers in reactions (physical chemistry); and,
- the Karplus equation.

Level of Experiment

Second year undergraduate

Keyword Descriptions of the Experiment

Domain

biological chemistry, physical chemistry

Specific Descriptors

enzymatic reactions, NMR, Karplus equation, fumarase, synthesis, isomerism

Course Context and Prerequisite Knowledge and Skills

- Biological chemistry (2nd Year)
- ^1H NMR practice and theory
- General organic chemistry or biochemistry

Time Required to Complete

Prior to Lab: 30 min (reading time)

In Laboratory: 3 h (preparation, Day 1) plus 30 min (final NMR, Day 3)

After Laboratory: 1 h

Experiment History

Commonly used towards the end of second year practical experiments for Biological Chemistry.

Comments

Requires the use of a NMR spectrometer (low frequency is sufficient) and additional molecular modelling kits.

References

1. Olsen, J. A. & Olsen, R. J. (1991). An NMR study of the stereochemistry of the fumarase-catalyzed hydration of fumaric acid. *Journal of Chemical Education*, **68**,436-437.
2. Nigh, W. G. (1976). Kinetic investigation of an enzyme catalyzed reaction. A biochemistry or physical chemistry laboratory. *Journal of Chemical Education*, **53**,668-669.