

ENZYMES AND FOOD PROCESSING

**Edited by
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ENZYMES AND FOOD PROCESSING

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Enzymes are biological catalysts which speed up chemical reactions. They are found in all living organisms and are essential for life. In food processing, enzymes are used to improve the quality and shelf life of products.

Enzymes are classified into six main groups: oxidoreductases, transferases, hydrolases, lysozymes, isomerases, and ligases.

Enzymes are used in a wide variety of food processing applications, including the production of bread, beer, cheese, and wine.

Enzymes are also used in the production of food additives, such as flavorings and colorants.

Enzymes are used in the production of food products that are low in fat, sugar, and salt.

*An industry-university co-operation Symposium organised
under the auspices of the National College of Food Technology,
University of Reading, 31 March-2 April, 1980*

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Glossary

AP	Alkaline phosphatase
ATP	Adenosine 5'-triphosphate
AU	Anson units—of protease activity, based on the change in absorbance at 750 nm occasioned by its degradation of haemoglobin for 10 min at 25°C, pH 7.5
BML	Bovine milk lysozyme
CANP	Calcium-activated neutral proteinase
DE	Dextrose equivalent = reducing sugars content as dextrose % dry substance
DEAE-cellulose	Diethylaminoethylcellulose
DFP	Diisopropylphosphofluoridate
DH	Degree of hydrolysis
DNA	Deoxyribonucleic acid
DP	Degree of polymerisation
DS	Dry substance (wt. %)
DX	wt. % of dextrose as such
EC	Enzyme Commission classification
EDTA	Ethylenediamine tetraacetic acid
Endo-enzyme	Enzyme capable of attacking its substrate at any internal linkage
Endomysium	Intramuscular connective tissue uniting fibres into bundles
EWL	Egg-white lysozyme
Exo-enzyme	Enzyme which attacks only at the end of a chain
FAD	Flavine-adenine dinucleotide
FGM	Fat globule membrane
GMP	Good Manufacturing Practice

G6PD	Glucose-6-phosphate dehydrogenase
HCFS	High-fructose corn syrup
HFGS	High-fructose glucose syrup
HML	Human milk lysozyme
HTST	High temperature, short time (pasteurisation)
INT	2- <i>p</i> -iodophenyl-3- <i>p</i> -nitrophenyl-5-phenyltetrazolium chloride
ISSPH	Isoelectric-soluble soy protein hydrolysate
IU ₄₂₀	International unit of colour based on absorbance at 420 nm
K_i	Inhibitor constant—equilibrium constant of the reaction $E + I \rightarrow EI$
K_m	Michaelis constant—substrate concentration at which the reaction velocity is half its maximum value (when enzyme is saturated with substrate)
LPL	Lipoprotein lipase
LPO	Lactoperoxidase
LTLT	Low temperature long time (pasteurisation)
NAD	Nicotinamide-adenine dinucleotide
NADH	Reduced nicotinamide-adenine dinucleotide
NADP	Nicotinamide-adenine dinucleotide phosphate
NADPH	Reduced nicotinamide-adenine dinucleotide phosphate
PER	Protein Efficiency Ratio
Perimysium	Intramuscular connective tissue uniting fibre bundles into muscles
PMS	Phenazine methosulphate
PMSF	Phenylmethanesulphonyl fluoride, $C_6H_5CH_2SO_2F$
RNA	Ribonucleic acid
SO	Sulphydryl oxidase
SOD	Superoxide dismutase
TCA	Trichloroacetic acid
UHT	Ultra-high temperature (flash pasteurisation)
XO	Xanthine oxidase

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Introduction

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Among the material to be discussed in this first section of the 'Enzymes and Food Processing Symposium' is subject matter that can be viewed as a marriage between enzyme technology and sugar stereochemistry. In order to bring the significance of the material to be presented into proper perspective, I would like you to pretend, for a moment, that you are a researcher making a proposal on this subject to a Research Granting Agency in order to obtain financial support for your ideas. *However, the year is 1880.*

Under the 'objectives' section of your proposal, you state that you intend to attach the intangible vital force or spirit—that is, the catalyst unique to the chemistry of living organisms—to an inert substrate such as sand. Thereafter you will pass a solution of right-handed glucose (also known as starch sugar) past the 'vital force' and in the process convert it to left-handed glucose (also known as fruit sugar). The peer review committee would probably reject the proposal as sheer nonsense because the statements made were not only contrary to their experience, but also contrary to what they had been taught. Perhaps a few select people would have some feeling for what you were talking about, but commiseration would be the only form of support that they could offer.

We can now return to the year 1980, and while researchers still have the same problems with peer review committees, we can rephrase our objectives. In doing so, I would like to remind you that two of the fundamental concepts needed, which form the basis of much of the material to be presented, were initially rejected as being absurd by many people. Parenthetically the concepts and their proofs were also the products of researchers located at institutions with a mission not unlike that of the National College of Food Technology at

Weybridge. One of the persons who placed the key into the lock on the box containing the mysteries of carbohydrate stereochemistry was van't Hoff. With his idea of the tetrahedral nature of the carbon atom, van't Hoff laid the foundation of modern stereochemistry. At the time, van't Hoff was employed by the Veterinary College at Utrecht—a fact that, to the great German chemist Kolbe, was unforgivable. Kolbe accused van't Hoff of mounting his winged horse Pegasus (and Pegasus was apparently taken from the stables of the Veterinary College) and, on his journey to the stars, 'seeing the arrangement of atoms in space'. As you well know, van't Hoff received the Nobel Prize for his efforts, in spite of the initial scathing criticism. His thoughts were proved via quantum mechanical calculations in 1931, by a then-recent graduate of the Oregon State Agricultural College in Corvallis by the name of Linus Pauling.

The enzyme chemist who suffered a similar fate was Professor James B. Sumner. The notion that biological catalysts were mysterious forces attached to a colloidal carrier had carried over until about 1926. Sumner was among those who had the audacity to believe that enzymes themselves were distinct chemical entities, probably proteins, and as such could perhaps be crystallised. He succeeded in doing just that with the enzyme urease from Jack-bean meal. As one of Sumner's last students I marvel to this day at that accomplishment. The very first laboratory course in his advanced biochemistry course was to crystallise urease from Jack-bean meal. The idea then was to repeatedly recrystallise the enzyme, and demonstrate an asymptotic increase in activity with successive recrystallisations. My conclusion from this exercise was that Professor Sumner, in order to demonstrate the proteinaceous and crystallisable nature of enzymes, had made a very poor choice. This enzyme is about the most sensitive enzyme that there is. It is inactivated by traces of nearly any metal or even by shaking a solution of it. It requires excellent chemical technique to prove the relation.

For his efforts Sumner was immediately subjected to the wrath of the famous Professor Willstätter because Sumner's ideas violated the Willstätter-Träger theory. Sumner subsequently received the Nobel Prize, however. All of this came about in spite of the fact that he was employed by the New York State College of Agriculture at Cornell University, and had been told by his Professor, Folin at Harvard, that he could never amount to much as a research chemist because he had only one arm.

Because they are distinct chemical entities, enzymes can be isolated, crystallised, and immobilised on inert substrates—even on sand. If the enzyme happens to be an isomerase, then it will execute an appropriate stereochemical transformation, such as the conversion of right-handed glucose to left-handed fructose. The implications of such thinking, and the potential applications are exciting.

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Keynote: Enzymes and Food Processing

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ABSTRACT

The main unifying theme of any conference on this subject must be the use of enzymes to modify biopolymers. As these are almost invariably condensation polymers, the main chemical reaction involved is hydrolysis, which may be very slight or be taken to the stage of complete breakdown to monomers as in the preparation of crystalline glucose from starch.

There are many reasons for wishing to manipulate biopolymers. The improvement of digestibility, palatability and general attractiveness are important; so also are improvements in keeping quality, nutritional value, ease of preparation and yield of edible fraction from raw material. The increasing cost of food has also recently emphasised the economic necessity of utilising polymers formerly regarded as waste or as outside the range of normal foodstuffs, e.g. the recovery of waste protein or the utilisation of cellulose. The same reason leads to a search for cheaper starting materials for the supply of well established foodstuffs, e.g. the switch from sucrose to glucose as a source of fructose since the advent of glucose isomerase. In this economic context it should be borne in mind that enzymes may well encounter political as well as technological opposition. As food becomes more sophisticated, the subtlety of the processes involved in its preparation increases, demanding even stricter control and therefore increasing variety and purity in the catalysts concerned.

Unfortunately, enzymes are not always beneficial—for example, proteolysis in dough, autolysis in vegetables designated for freezing and rancidity in fat-containing foods all involve some degree of