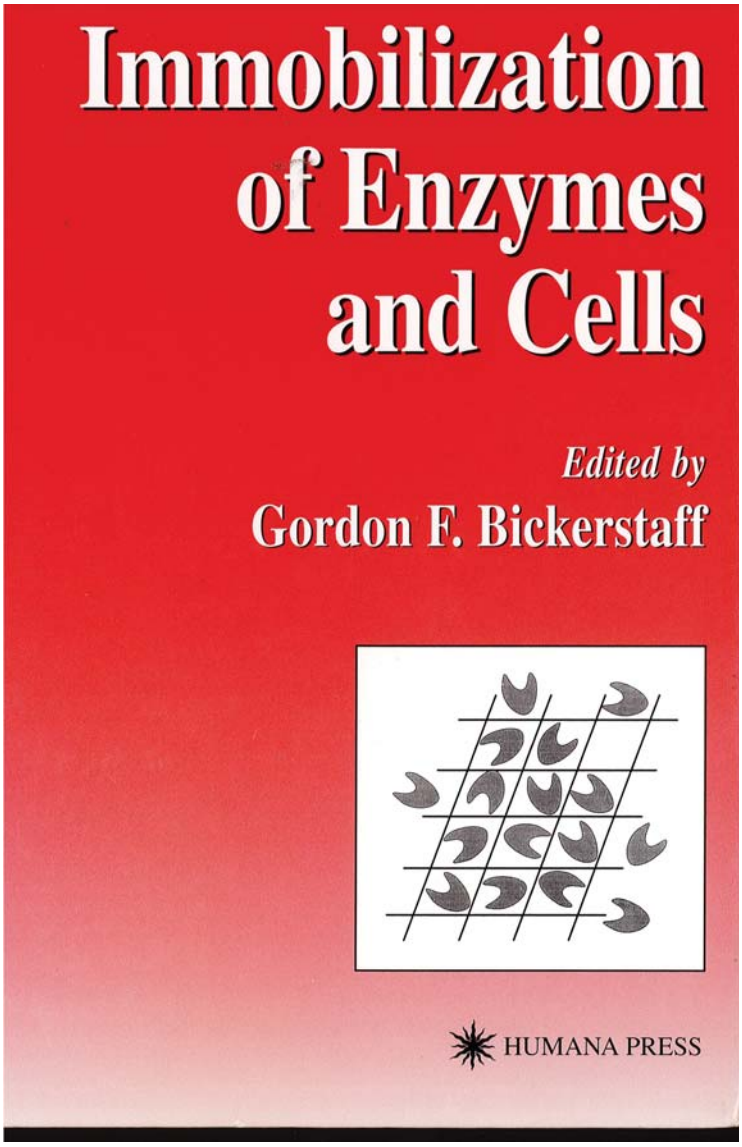

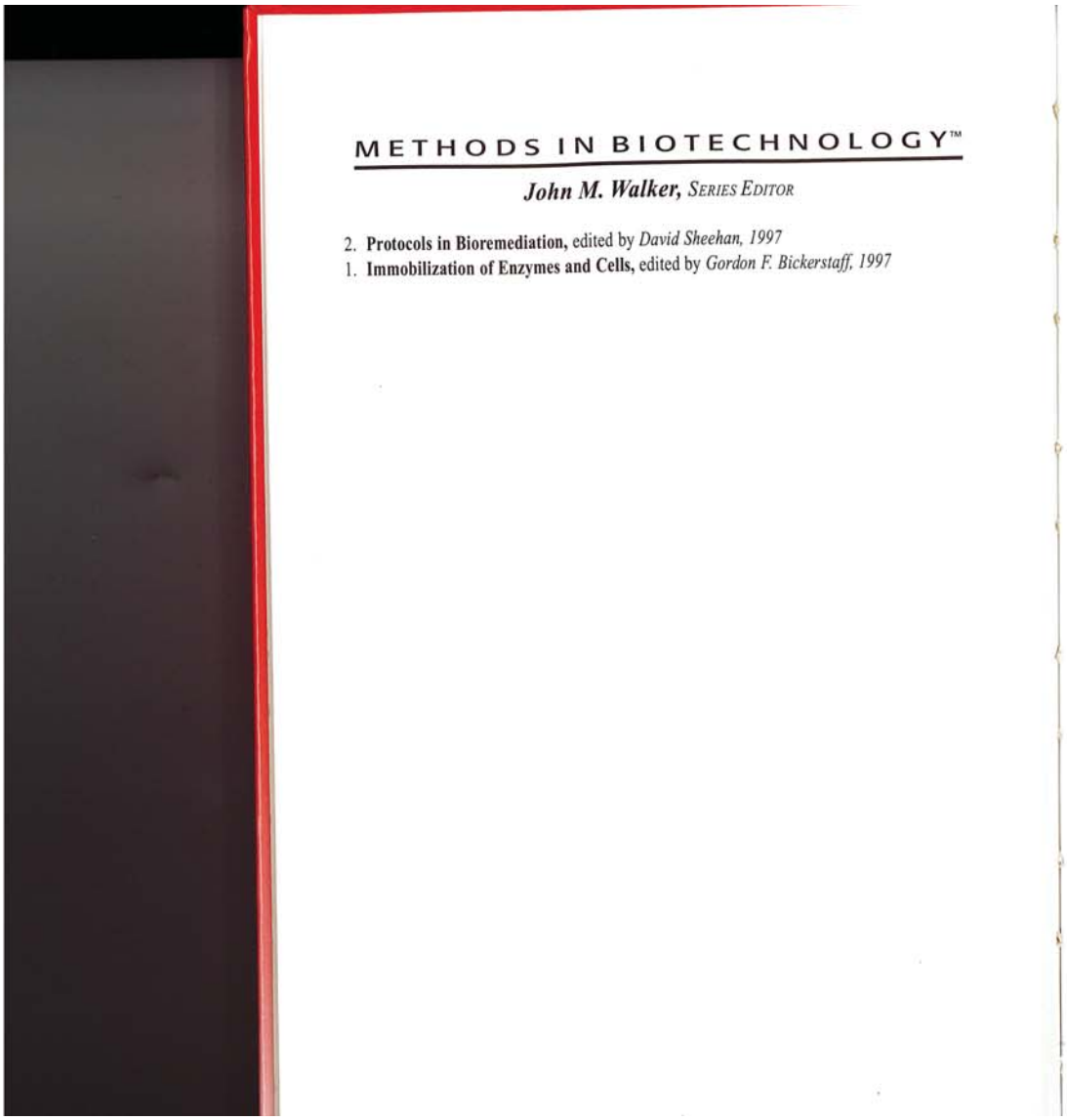

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Preface

Immobilization of enzymes, cells, ar
in the past 30 years as the advantages of ir
and utilized in analytical, biotransformatio
sequence of this explosion of technology
array of permutations for the immobilizati
pose of *Immobilization of Enzymes and C*
tool for all academic and industrial research
the use of immobilization techniques in the
provide comprehensive coverage of the va
will serve as a launch pad for potential use

One reason for the vast expanse of im
subject material to be immobilized. Biolog
and cells) have a high degree of individu
immobilization techniques have wide appl
even a few methods to cater to the great di
biological material. This is especially so v
mum system in which the immobilized bio
of efficiency, stability, and so on.

The normal situation faced by researc
more methods of immobilization to reveal t
the biological catalyst, then adapt the metho
try another method when the first approach
use or activity of the biocatalyst. This proces
it is also time-consuming, usually frustratin
successful start. *Immobilization of Enzymes*
vide a wide range of representative exampl
use by postgraduate, postdoctoral, senior
throughout academia, industry, government,
thereby enabling rapid entry into the world

All of the chapters, with the except
instructions of the materials and methods
lization procedure described. Each author
on accumulated experience and valuable

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1

Immobilization of Enzymes a

Some Practical Considerations

Gordon F. Bickerstaff

1. Introduction

The technology for immobilization of ce
the first 25 years of its existence *(1)*, but in r
not a slight decline. However, the expansio
developments that will accrue from advanc
ized enthusiasm for immobilization of enzyr
opment work has provided a bewildering ar
for immobilization. Much of the expansion
provide specific improvements for a given ap
been few detailed and comprehensive co
methods and supports. Therefore, no ideal su
lization has emerged to provide a standard fo
tion of support material and method of imn
various characteristics and required features
the properties/limitations/characteristics of t
A number of practical aspects should be co
mental work to ensure that the final immobil
fit for the planned purpose or application ar
ness *(4–6)*. This chapter does not aim to pr
but does provide some background to assist
method of immobilization.

2. Choice of Support and Principal

In solution, soluble enzyme molecules b
are readily dispersed in the solution and h

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Edited by: G. F. Bickerstaff Huma

Table 1
Fundamental Considerations in Selecting a Support and Method of Immobilization

Property	Points for consideration
Physical	Strength, noncompression of particles, available surface area, shape/form (beads/sheets/fibers), degree of porosity, pore volume, permeability, density, space for increased biomass, flow rate, and pressure drop
Chemical	Hydrophilicity (water binding by the support), inertness toward enzyme/cell, available functional groups for modification, and regeneration/reuse of support
Stability	Storage, residual enzyme activity, cell productivity, regeneration of enzyme activity, maintenance of cell viability, and mechanical stability of support material
Resistance	Bacterial/fungal attack, disruption by chemicals, pH, temperature, organic solvents, proteases, and cell defense mechanisms (proteins/cells)
Safety	Biocompatibility (invokes an immune response), toxicity of component reagents, health and safety for process workers and end-product users, specification of immobilized preparation (GRAS list requirements for FDA approval) for food, pharmaceutical, and medical applications
Economic	Availability and cost of support, chemicals, special equipment, reagents, technical skill required, environmental impact, industrial-scale chemical preparation, feasibility for scale-up, continuous processing, effective working life, reuseable support, and CRL or zero contamination (enzyme/cell-free product)
Reaction	Flow rate, enzyme/cell loading and catalytic productivity, reaction kinetics, side reactions, multiple enzyme and/or cell systems, batch, CSTR, PBR, FBR, ALR, and so on; diffusion limitations on mass transfer of cofactors, substrates, and products

CRL: calculated risk level, CSTR: continuous stirred tank reactor, PBR: packed bed reactor, FBR: fluidized bed reactor, ALR: air lift reactor.

(1). Enzyme immobilization is a technique specifically designed to greatly restrict the freedom of movement of an enzyme. Most cells are naturally immobilized one way or another, so immobilization provides a physical support for cells. The first consideration is to decide on the support material, then the main method of immobilization, taking into account the intended use and application. Some of the points to consider when making a decision are listed in Table 1, and

Table 2
Influence of Immobilization Meth on the Biotransformation of Suc to Isomaltulose by Cells of *Erwi*

Free cells	
Immobilization method	
Entrapment in calcium alginate	
Entrapment in polyacrylamide	
Adsorption to DEAE-cellulose	
Crosslinking with glutaraldehyde	
Entrapment in κ-carrageenan	
Entrapment in agar	
Adsorption to bone char	

Reprinted with permission from ref. *2*.

an indication of how different methods o activity and half-life of a cell-based biotra There are five principal methods for immo tion, covalent binding, entrapment, encapsu The relative merits of each are discussed b

2.1. Adsorption

Immobilization by adsorption (*see* Fig. 1) reversible surface interactions between enz The forces involved are mostly electrostatic and hydrogen bonding interactions, althoug nificant. These forces are very weak, but su reasonable binding. For example, it is kn chemistry that is substantially negatively charged support will enable immobilization the enzyme/cells and support is utilized so is required and little damage is normally do of immobilization. The procedure consist component(s) and a support with adsorpti tions of pH, ionic strength, and so on, for collection of the immobilized material nonbound biological components.

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