

A comparative analysis on the efficiency of different carriers for trypsin immobilization

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Trypsin is a widely used enzyme for protein hydrolysis and can be used to improve functional and nutritional properties of foods. Its immobilization on solid carriers can offer several advantages over the free enzyme including easy handling, recovery from the reaction medium, reuse and operation in continuous reactors.

This work compares different carriers, namely zeolyte NaY, spent grains and magnetic polysiloxane-polyvinyl alcohol composite (POS-PVA), as potential candidates for trypsin immobilization. Covalent attachment to the carriers was tested using glutaraldehyde or glycidol. The efficiency of immobilization and activity, operation and storage stability of free and immobilized enzyme on the supports were evaluated.

Table 1: Immobilized protein and activity retention achieved with the different carriers

Carrier	Type of enzyme	Immobilized protein (%)	Activity (U/g carrier)	Retention of activity (%)
Zeolite	Crude	46.8±0.9	35.6±1.5	66.8
Zeolite	Purified	70.0±4.1	52.6±1.9	63.5
POS-PVA	Crude	30.3±1.3	3.27±0.27	13.7
POS-PVA	Purified	41.6±7.7	37.6±5.6	73.7
Glyoxyl spent grain	Crude	68.7±1.0	8.25±0.96	10.5
Glyoxyl spent grain	Purified	69.8±3.9	44.3±2.2	46.0

When a crude enzyme as used, the amount of immobilized protein achieved was high (up to 70 %) but the activity retention was low for all carriers except for trypsin crosslinked on zeolites, where it was satisfactory (Table 1). However, when a more purified enzyme from bovine pancreas was used with glyoxyl-spent grain or POS-PVA with glutaraldehyde, the activity retention was of 46 % and 73 % against 11 % and 9 % with crude enzyme.

The operational stability obtained was always close to 100 % while the storage stability of these immobilized enzymes was always above 80 %.

Thus it can be stated that trypsin was successfully immobilized on spent grains by multipoint covalent attachment using glycidol and on POS-PVA functionalized with glutaraldehyde. The immobilized trypsin with the highest activity was however achieved with covalent binding through glutaraldehyde to silanized zeolite followed by crosslinking with glutaraldehyde, probably due to a positive effect of the zeolite on the enzyme activity.