# TECHNOLOGY OFFER

## Recombinant Production of Horseradish Peroxidase (HRP)

Recombinant production of horseradish peroxidase (HRP) guarantees a steady supply of high-purity single-isoform HRP preparations and represents a viable alternative to traditional hairy root culture. Scalability, reproducibility and adherence to Quality by Design (QbD) concepts open up a wide range of different application areas in diagnostics and medicine.

### BACKGROUND

Horseradish peroxidase (HRP) is an industrially important enzyme with a broad field of applications in biotechnology, life sciences and medicine. Traditionally, commercial preparations of HRP are isolated from horseradish roots, which is time-consuming and dependent on environmental conditions. The respective isolate is a heterogeneous mixture of isoenzymes with plant-specific glycosylation patterns causing high batch to batch variations and immunogenicity in humans, thus limiting industrial and medical applicability. So far, alternative strategies resulted in low yields and/or decreased enzyme activity and stability. Therefore, high-yield and high-quality production of defined recombinant HRP along QbD principles is of great commercial value.

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### TECHNOLOGY

A novel, scalable process for the recombinant production of HRP from *Escherichia coli* (*E. coli*) has been established, which enables the supply of a highly pure, homogeneous single HRP isoenzyme. In fact, the respective HRP preparation competes with the plant-derived protein for enzymatic activity, stability and Reinheitszahl (Rz). The enzyme is expressed as inclusion bodies, solubilized, refolded and concentrated by 1-step chromatography to several g/L with a purity of ≥ 99 %. After dialysis, the enzyme can be lyophilized and reconstituted in water without activity loss. The recombinant, unglycosylated HRP can also be modified by PEGylation or conjugation to antibodies using glutaraldehyde, which significantly broadens the application spectrum.

### ADVANTAGES

- Non-plant derived recombinant HRP
- Scalable and highly reproducible HRP production along QbD principles
- Homogeneous single isoform with consistent biochemical properties
- Comparable Rz and catalytic activity (ABTS) to plant enzyme preparation
- No plant or host-related glycosylation (non-immunogenic)
- Conjugation to mABs and other proteins can be achieved using glutaraldehyde
- PEGylation feasible via NHS chemistry

### REFERENCE:

*M061/2019*
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### DEVELOPMENT STATUS:

Laboratory prototype

### APPLICATIONS:

Molecular targeting
Detection and quantification
Biosensor systems
Biocatalysis
Organic synthesis
Bioremediation
Cancer therapy

### KEYWORDS:

Recombinant HRP
Scalable and defined
Quality by design
Unglycosylated
Single isoform
Consistent quality
High purity
Scalable production process
Reinheitszahl
Catalytic activity

### IPR:

EP, US, JP and CN
applications filed

### OPTIONS:

License agreement

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