

# ENZOVERA LIFE SCIENCES

## Mussel Adhesive Proteins, Sterile — User Manual

Cat. No. EV-PRO-002

### 1. Overview

Mussel Adhesive Proteins, Sterile (EV-PRO-002) are recombinant proteins derived from yeast expression mimicking the foot proteins (Mfp-3, Mfp-5) of marine mussels. These proteins contain DOPA (3,4-dihydroxy-L-phenylalanine) residues that mediate strong adhesion to wet surfaces through catechol-mediated cross-linking and hydrogen bonding. EV-PRO-002 has demonstrated wound healing promotion, anti-inflammatory activity, acceleration of skin repair, and excellent biocompatibility with no reported toxic side effects. Sterile-grade for tissue engineering and regenerative medicine research applications.

### 2. Mechanism of Action

Mussel adhesive proteins adhere to surfaces through their DOPA content. DOPA forms bidentate hydrogen bonds, metal coordination complexes, and covalent cross-links (via oxidation to dopaquinone) with surface hydroxyl groups and other biomolecules. This mechanism confers adhesion to wet surfaces including tissue, implant materials, and hydrogels. The proteins also stimulate fibroblast proliferation, modulate inflammatory cytokines, and promote re-epithelialization in wound healing models.

### 3. Applications

- Tissue-adhesive wound dressing and wound healing scaffold research
- Biomedical device surface coating for improved cell adhesion
- 3D bioprinting matrix component with inherent bioadhesive properties
- Anti-inflammatory research in skin repair models
- Hydrogel fabrication with self-healing and adhesive properties

### 4. Recommended Protocol

Step	Details	Application
1	Reconstitute in 0.1 M acetic acid at 10 mg/ml under aseptic conditions.	Stock preparation
2	For coating: spread 0.5–1 mg/ml solution on surface; air-dry 30 min at RT.	Surface coating

3	For wound application: dissolve at 5 mg/ml in PBS pH 6.5; apply as gel or film.	Wound dressing
4	Cross-link with NaIO <sub>4</sub> (1 mM) for enhanced adhesion strength. Sterile grade: all reconstitution steps must be performed aseptically in a biosafety cabinet.	Optional cross-linking

## 5. Unit Definition / Activity Specification

Activity expressed as purity ( $\geq 90\%$  by SDS-PAGE) and DOPA content (mol%) by amino acid analysis. Adhesion strength measured by ASTM D1002 standard ( $> 50$  kPa on stainless steel). Sterility confirmed by 7-day TSB incubation. Cytotoxicity assessed per ISO 10993-5 ( $> 80\%$  cell viability at 1 mg/ml).

## 6. Quality Control

Test / Parameter	Specification	Lot Result	Status
Appearance	Off-white or yellowish lyophilized powder	Conforms	PASS
Purity (SDS-PAGE)	$\geq 90\%$	$\geq 92\%$	PASS
DOPA Content	$\geq 1.5$ mol% DOPA by amino acid analysis	$\geq 1.8$ mol%	PASS
Adhesion Strength	$> 50$ kPa on stainless steel (ASTM D1002)	$\geq 55$ kPa	PASS
Sterility	No microbial growth (7-day TSB incubation)	No growth	PASS
Endotoxin	$< 1.0$ EU/mg (LAL)	$< 0.5$ EU/mg	PASS
Cytotoxicity	Non-cytotoxic (ISO 10993-5; $> 80\%$ cell viability at 1 mg/ml)	Conforms	PASS

## 7. Storage & Stability

- Storage temperature:  $-20^{\circ}\text{C}$
- Stability: 2 years at  $-20^{\circ}\text{C}$
- Formulation: Lyophilized; reconstitute in 0.1 M acetic acid or sterile water at pH 4–5
- Shipping: Dry ice
- General: Avoid repeated freeze–thaw; aliquot upon receipt for multi-use formats. Protect from light.

## 8. Troubleshooting

Problem	Possible Cause	Suggested Action
Poor adhesion after coating	Oxidation of DOPA before surface contact	Prepare fresh solution; minimize exposure to air before application; add 1 mM NaIO <sub>4</sub> for cross-linking if needed.
Precipitation on reconstitution	Alkaline pH causing DOPA oxidation	Reconstitute in acidic buffer (pH 4–5, 0.1 M acetic acid); avoid PBS without pH adjustment; confirm acetic acid concentration.
Low wound healing activity	Protein degradation or loss of bioactivity during storage	Confirm storage at -20°C; check expiration date; avoid freeze–thaw cycles; prepare fresh aliquots from original stock.

## 9. Safety Information

For Research Use Only. Handle according to standard laboratory safety guidelines. Refer to the accompanying SDS for full hazard information. Dispose in accordance with local, state, and federal regulations.

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