1. Product Information

Product name	**Recombinant spCas9**
Internal product code	BMC9
Catalog numbers (aliquot sizes)	540-025-GMP (2.5 mg) 540-050-GMP (5 mg)
Quality grade	**current Good Manufacturing Practice (cGMP)**
Description	Recombinant wild type Cas9 nuclease from *Streptococcus pyogenes* with nuclear localization sequence (NLS-spCas9-NLS)
Molecular weight	162 kDa
Concentration	10 mg/mL solution in buffer (frozen)
Storage buffer	25 mM Tris, 300 mM NaCl, 0.1 mM EDTA, 50% glycerol, pH 7.4 buffer solution
Storage temperature	-75 ± 15 °C
Lot. Nr.	Specified on product label
Use by date	Specified on product label
Manufacturer	Biomay AG, Ada-Lovelace-Straße 2; 1220 Vienna, Austria; www.biomay.com; info@biomay.com

2. Description

CRISPR nuclease Cas9 (spCas9 from *Streptococcus pyogenes*, uniprot Q99ZW2 (CAS9_STRP1)) with nuclear localization sequences (NLS) on the N- and C-terminus. The product has been expressed as recombinant protein in *Escherichia coli*, purified by chromatography, filtered (0.2µm) and aseptically filled as a sterile product.

3. Intended Use / Application

cGMP grade: product is a recombinant protein that has been manufactured and quality-controlled under the conditions of current Good Manufacturing Practice (GMP). It has been certified and released by a Qualified Person (QP) under EMA law (EMA directive 2001/83/EC). It was designed and is intended to be used for gene-editing of eukaryotic cells with a specific guide RNA (gRNA). It is fully compliant to be used as critical starting material for gene-editing in clinical trials (phase I-III) or for commercial use.

Biomay imposes no restrictions on clients regarding the use of the purchased Cas9, except in the field of hemoglobinopathies. In the specific area of hemoglobinopathies, Cas9 obtained from Biomay must not be used for preclinical or clinical trials (Phases I, II or III), nor for any commercial applications. Clients are required to acknowledge and accept these specific terms related to Biomay's Cas9 with the acquisition of the material.

4. Quality Control and Specifications

Parameter	Quality Control Method	Specification *cGMP grade*
Appearance	Visual inspection	Clear and colourless liquid, free of visible particles
pH-value	pH potentiometric (Ph. Eur. 2.2.3)	7.4 ± 0.3
Content (concentration)	UV spectrophotometric (UV 280 nm / Ph. Eur. 2.2.25)	9.0 - 11.0 mg/ml
Homogeneity (aggregates)	Size exclusion chromatography (HP-SEC)	≥ 95% monomer
Identity	Western blot	Positive reaction of the main band with the Cas9 specific antibody
		Migration of the main band conforms to reference
Purity	SDS-PAGE	Migration of the main band conforms to reference
		Purity: ≥ 95%
Host cell protein	E. *coli* host cell protein ELISA	≤ 350 ng/mg (≤ 0.035 % w/w)
Endotoxins	LAL test (Ph. Eur. 2.6.14 method D)	< 5 EU/mg
Residual RNase	Fluorimetric	< 5 mU/ml
Residual DNase	Fluorimetric	No DNase activity detectable
Host cell DNA	qPCR	≤ 30 ng/mg (≤ 0.003 % w/w)
Sterility	Sterility testing (Ph. Eur. 2.6.1)	Sterile
Functionality assay	RP-HPLC	70 – 150% relative EC_{50}

5. Safety information

Material is considered non-infectious, non-toxic and non-pathogenic under the conditions of the intended applications. General safety procedures should still be followed to maintain a safe working environment. Always wear appropriate personal protective equipment (PPE), including lab coats, gloves, and safety glasses, to avoid contamination and accidental exposure. Work in a clean, organized space, and handle reagents with care, avoiding direct contact. Dispose of all waste materials, including gloves and pipette tips, in designated biohazard containers, even though the commodity is non-hazardous, to prevent cross-contamination. Always wash hands after handling any biological material and before leaving the lab.