ANALYSIS REPORT WYG-LNM

Report Generated
MAY 17, 2026

Analysis	**Standard Mass / Purity** (HPLC-UV) **+ Advanced HR-MS Package**
Client	Helion



Sample

Tirzepatide 50mg

BATCH 1141

SELLER Helion Peptides

RESULTS

MASS / PURITY RESULTS (HPLC-UV)

PURITY
99.70%

AMOUNT
54.83 mg

HR-MS RESULTS ADVANCED

MS PURITY
97.44%

IMPURITIES

Variants / Degradation	LOW
Process Impurities	NOT DETECTED
Contaminants	NOT DETECTED

PEPTIDE

Identity	CONFIRMED
Sequence	MATCH
Fingerprint	MATCH

COMMENT

No contaminants or process impurities detected. Identity, sequence and MS/MS fingerprint confirmed. Low level of degradation (deamidation 1.34 %, see page 3).

ANALYSIS CONDUCTED
12–15 MAY 2026
ZAGREB, CROATIA (EU)

Alex Mihaljcic
DIRECTOR

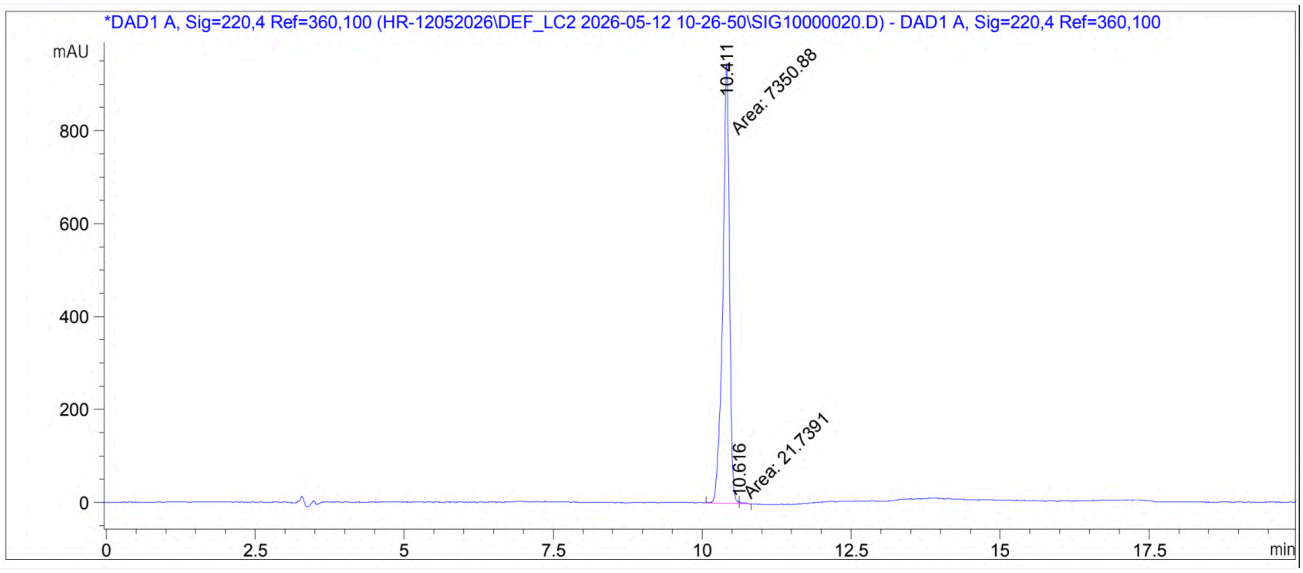
MASS / PURITY RESULTS HPLC-UV

PURITY

UV Diode-Array (DAD) at 220 nm (Reference 360 nm)

99.70%

PURITY CHROMATOGRAM — DAD1 A, SIG=220,4 REF=360,100



Purity determined by area percent method using UV diode-array detection at 220 nm. The principal peak area is expressed as a percentage of total integrated peak area.

PEPTIDE AMOUNT

UV Quantification — External Standard Method

54.83 mg

INDIVIDUAL MEASUREMENTS

TEST	MEASURED	VS LABEL
1	**55.59 mg**	111.2%
2	**54.12 mg**	108.2%
3	**54.78 mg**	109.6%
Average	**54.83 mg**	**109.7%**

Peptide content determined by comparing the sample UV peak area (220 nm) against an external reference standard of the same peptide at known concentration; the standard lot is recorded in the raw data. Three independent preparations were measured (RSD: 1.3%).

IMPURITIES HR-MS ADVANCED

MS ANALYSIS SUMMARY

MS PURITY

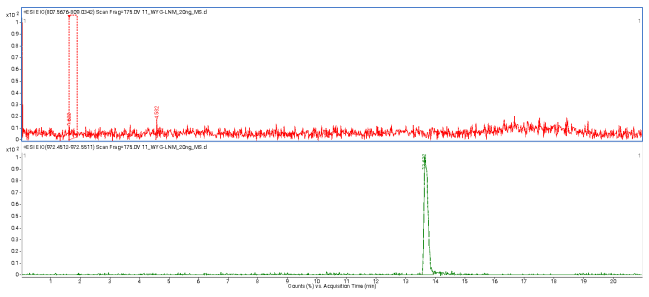
97.44%

TOTAL IMPURITIES

2.56%

BASELINE SYSTEM CONFIRMATION

Noise check: EIC at an unoccupied m/z (807.57–809.03, noise only, top) vs EIC of a principal-ion 5+ isotope (972.45–972.55, bottom)



VARIANTS / DEGRADATION

IMPURITY	MASS SHIFT	%	RISK LEVEL
Mono-oxidation	+15.99 Da	–	NOT DETECTED
Di-oxidation	+31.99 Da	–	NOT DETECTED
Methylation	+14.02 Da	–	NOT DETECTED
Deamidation	+0.98 Da	1.34%	LOW
Dehydration (water loss)	−18.01 Da	–	NOT DETECTED
Ammonia loss	−17.03 Da	–	NOT DETECTED

PROCESS IMPURITIES

IMPURITY	MASS SHIFT	%	RISK LEVEL
TFA adduct	+113.99 Da	–	NOT DETECTED
Fmoc (protecting group)	+222.07 Da	–	NOT DETECTED
tBu (protecting group)	+56.06 Da	0.01%	NOT DETECTED

OTHERS

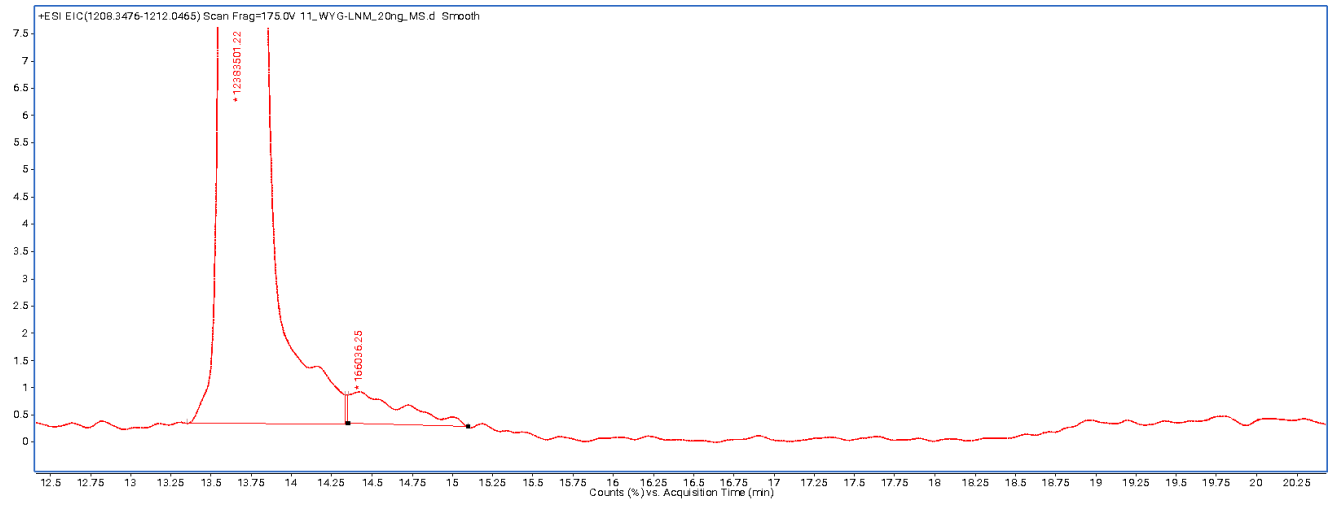
IMPURITY	MASS SHIFT	%	RISK LEVEL
Formylation	+27.99 Da	1.21%	VERY LOW

CONTAMINANTS

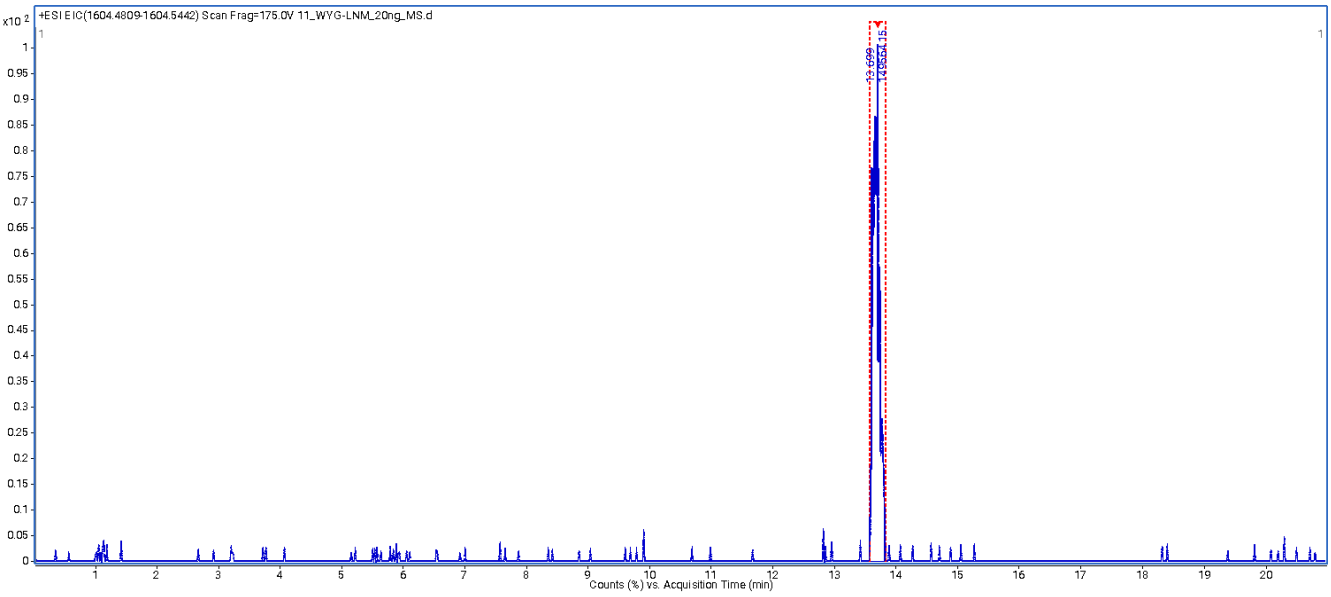
IMPURITY	RISK LEVEL
None	

High-resolution LC-MS (ESI+, Q-TOF). Each variant is identified by its exact mass shift from the principal peptide. % = extracted-ion peak area of the variant relative to the peak area of the principal peptide (same charge state); totals are computed from unrounded values, so rounded rows may not add up exactly. Rows below 0.005 % are shown as not detected. A TFA adduct signal (+113.99 Da) points to trifluoroacetate from synthesis being present as a counter-ion; when it is detected, residual TFA is checked separately.

Deamidation EIC m/z 1208.35–1212.05 (principal 4+ cluster) · later-eluting peak 14.35 min 1.34%



Formylation EIC m/z 1604.48–1604.54 (3+) · 13.70 min 1.21%

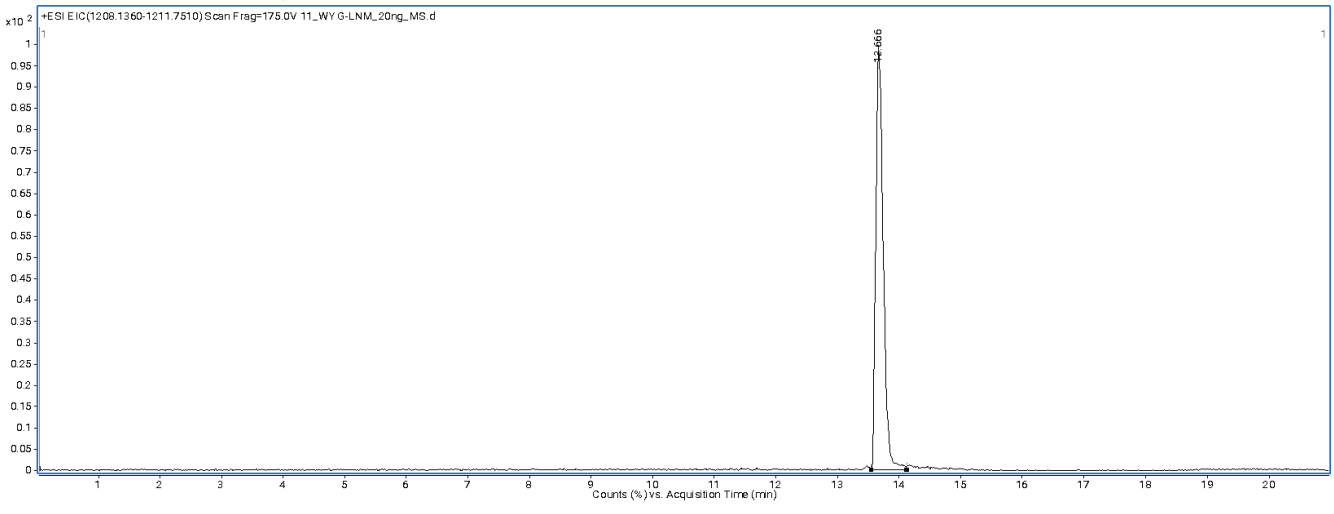


Extracted-ion chromatograms (EIC) at the m/z of each variant where a trace at the expected m/z is on file; the extracted m/z is printed in the panel header. Deamidation (+0.98 Da) cannot be resolved from the ^{13}C isotope pattern at this resolution; it is quantified from the later-eluting, chromatographically resolved peak in the EIC of the principal ion where such a peak is present, otherwise reported as an upper bound without a panel. Extracted window: in this run the most intense 4+ species is the sodium adduct [M+3H+Na]4+ (m/z 1209.88; [M+4H]4+ at 1204.14), so the principal-ion EIC covers that cluster, m/z 1208.35–1212.05. Deamidation shifts the 4+ ion by only +0.25 m/z, i.e. it falls inside the same window and is separated by retention time (14.35 vs 13.70 min), not by mass; the window is the adduct cluster of the principal peptide, not a +19 to +34 Da species.

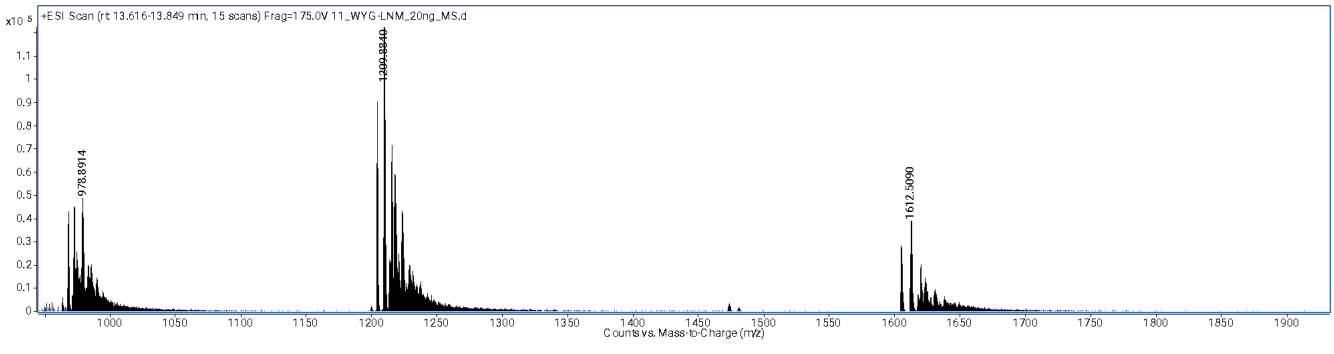
MS IDENTITY CONFIRMATION	CONFIRMED
Theoretical monoisotopic mass (from the molecular formula)	**4810.5249 Da**
Measured monoisotopic mass (deconvoluted from the charge-state envelope)	**4810.5249 Da**
Mass Error (measured − theoretical)	**+0.00 ppm**
Acceptance Limit	**≤ 5 ppm**
Molecular Formula	**C225H348N48068**

MASS SPECTRA

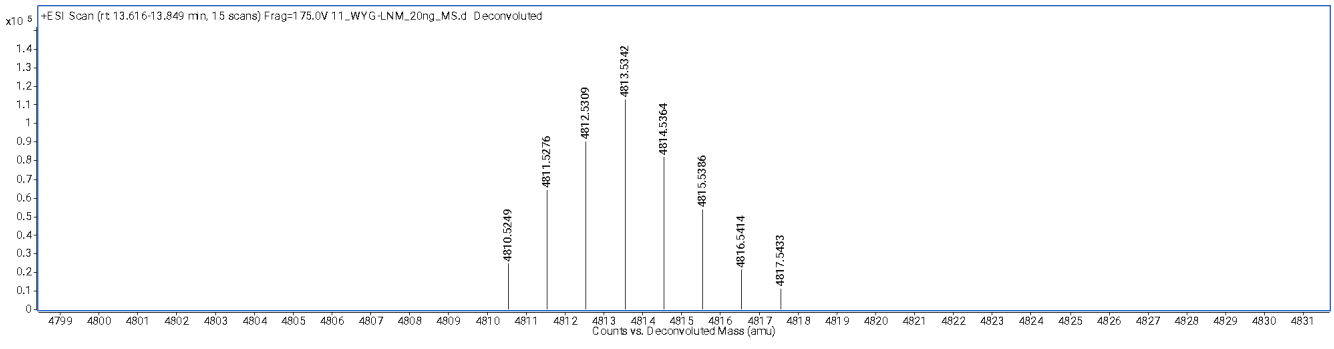
MS IDENTITY — EXTRACTED-ION CHROMATOGRAM OF THE PRINCIPAL 4+ ION CLUSTER (M/Z 1208.14–1211.75, SODIUM ADDUCT [M+3H+NA]4+, THE MOST INTENSE 4+ SPECIES IN THIS RUN)



MS IDENTITY — SPECTRUM 1: CHARGE-STATE ENVELOPE OF THE PRINCIPAL PEAK



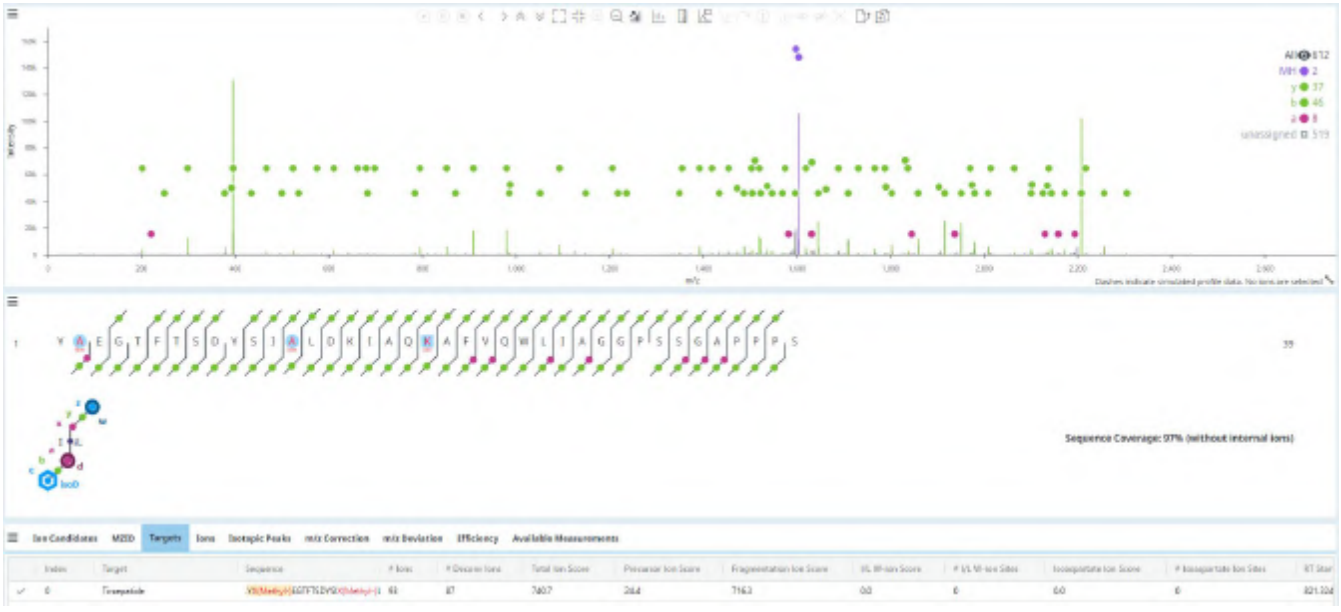
MS IDENTITY — SPECTRUM 2: DECONVOLUTED MONOISOTOPIC MASS



Identity by accurate mass (ESI+, Q-TOF): the multiply charged ions of the principal peak were deconvoluted to a neutral monoisotopic mass and compared with the monoisotopic mass calculated from the molecular formula; acceptance ≤ 5 ppm. No MS/MS fragmentation is used for this step.

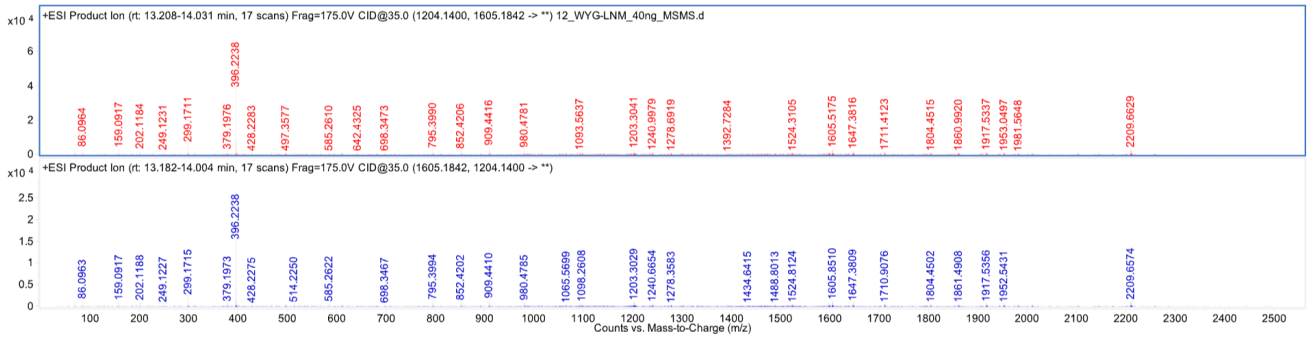
SEQUENCE ANALYSIS — MATCH

Sequence Coverage	97%
Acceptance Limit	≥ 80%
Amino Acid Match	100%



TANDEM MS FINGERPRINT — MATCH

Fingerprint Similarity	99.9%
Acceptance Limit	≥ 99.0%



Sequence: MS/MS (CID) fragment ions assigned to the expected amino-acid sequence; coverage is the share of the sequence confirmed by b-ions (from the N-terminus) and y-ions (from the C-terminus) taken together, so a residue is confirmed when it is covered from either end and the amino-acid match is checked over the full sequence (≥ 80 % coverage required). Fingerprint: the sample MS/MS spectrum compared with a working reference standard of the same peptide; ≥ 99.0 % similarity required. Not assessed when no working reference standard of the same peptide is on file.

METHODS

ANALYSIS METHODS

TEST	METHOD	HOW THE RESULT IS OBTAINED
Purity	HPLC–UV	Diode-array detection at 220 nm (reference 360 nm); principal peak area as a percentage of the total integrated peak area
Amount (net content)	HPLC–UV	Peak area at 220 nm against an external reference standard of the same peptide at known concentration; 3 independent preparations
Identity	HR–MS	Electrospray (ESI+) Q-TOF accurate mass; charge-state envelope deconvoluted to the monoisotopic mass; acceptance ≤ 5 ppm vs the formula mass
Sequence	MS/MS	Collision-induced dissociation of the principal ion; b/y fragment ions assigned to the expected sequence; ≥ 80 % coverage required
Fingerprint	MS/MS	MS/MS spectrum compared with a working reference standard of the same peptide; ≥ 99.0 % similarity required
Impurities	HR–MS	Extracted-ion chromatograms at the exact mass shift of each variant; % = variant peak area relative to the principal peptide peak area

STANDARDS, CONTROLS & LIMITS

Reference standards

Quantification uses a reference standard of the same peptide of known concentration; the lot used is recorded in the raw data.

Controls

A blank injection precedes the sample series; a baseline check is shown on the impurities page. Values below 0.005 % of the principal peak area are reported as not detected.

Scope

Results apply to the vial received and analysed. The impurity screen covers the mass shifts listed on the thresholds page; it is not an exhaustive search for every possible related substance. A detected TFA adduct indicates trifluoroacetate may be present as a counter-ion and triggers a separate check of residual TFA.

THRESHOLDS & CRITERIA

VARIANT / DEGRADATION RISK LEVELS

IMPURITY	ND	VERY LOW	LOW	MEDIUM	HIGH
Mono-oxidation +15.99 Da	= 0	< 0.5%	< 1.5%	< 3.0%	**≥ 3.0%**
Di-oxidation +31.99 Da	= 0	< 0.3%	< 0.8%	< 2.0%	**≥ 2.0%**
Methylation +14.02 Da	= 0	< 0.3%	< 0.5%	< 1.5%	**≥ 1.5%**
Deamidation +0.98 Da	= 0	< 1.0%	< 2.5%	< 5.0%	**≥ 5.0%**
Acetylation +42.01 Da	= 0	< 1.0%	< 2.5%	< 5.0%	**≥ 5.0%**
Formylation +27.99 Da	= 0	< 1.0%	< 2.5%	< 5.0%	**≥ 5.0%**
Boc (protecting group) +100.05 Da	= 0	< 1.0%	< 2.5%	< 5.0%	**≥ 5.0%**
Dehydration (water loss) −18.01 Da	= 0	—	< 0.2%	< 1.0%	**≥ 1.0%**
Ammonia loss −17.03 Da	= 0	—	< 0.2%	< 1.0%	**≥ 1.0%**

PROCESS IMPURITY THRESHOLDS

IMPURITY	ND	LOW	MEDIUM	DETECTED
TFA adduct +113.99 Da	= 0	< 0.1%	< 0.5%	**≥ 0.5%**
Fmoc (protecting group) +222.07 Da	< 0.05%	—	—	**≥ 0.05%**
tBu (protecting group) +56.06 Da	< 0.05%	—	—	**≥ 0.05%**

ACCEPTANCE CRITERIA

TEST	PASS	FAIL
Identity (Mass Error)	≤ 5 ppm	> 5 ppm
Sequence Coverage	≥ 80%	< 80%
Fingerprint Similarity	≥ 99.0%	< 99.0%
Reporting threshold (impurities)	≥ 0.005% of principal peak area is reported	below: not detected

TERMS & DISCLAIMER

OUR MISSION

A big part of our mission is harm reduction. Therefore certain assessment criteria (risk levels, acceptance limits) are set at our discretion and are disclosed in full on the "Thresholds & Criteria" page of this report, however:

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REPORT REVIEWED · NOV 08, 2026