Certificate of Analysis

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Analytical services

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Inovační 122
252 41 Zlatníky-Hodkovice
Czechia
www.liquilabs.cz

Verify Results Online

Sample Identification

Sample Name	TB-500 10 mg
Batch Number	BT102610
Date Published	2026-09-23 05:59

Results for LYO-0932

Peptides	Result	Unit	Uncertainty	Acceptable Range
Thymosin Beta 4 (TB-500) Assay Peptide Screening 0.1% TFA	7.72	*mg*	[± 0.04]	
Thymosin Beta 4 (TB-500) Purity Peptide Screening 0.1% TFA	99.0	*%*	[± 0.5]	
Thymosin Beta 4 (TB-500) Identification by Spectrum (FTIR) Peptide Screening 0.1% TFA	979		[± 5]	
Thymosin Beta 4 (TB-500) Identification by RT Peptide Screening 0.1% TFA	0.999		[± 0.005]	

Microbiology	Result	Unit	Uncertainty	Acceptable Range
Bacterial Endotoxin Chromgenic USP<85>/ Eur. Ph. 2.6.14. Bacterial Endotoxin Chromgenic Test	0.278	*EU/mg*	[± 0.006]	

Mass Spectrometry	Result	Unit	Uncertainty	Acceptable Range
Molecular Ion Mass Identification (MS Deconvolution) Mass Spectrometry Identity	4963	*Da*	[± 1]	

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LIQUILABS Analytical services	Method Specification	
Determination of identity, content and purity of Thymosin Beta 4		
Document number TB4_008_2026	*Superseded document* -	*Number of pages* 4

1. Content Assesment

1.1. Instrumentation

Module	Name	Serial Number
System Controller	Shimadzu CBM-40 Lite	L221226351398
Degassing Unit	Shimadzu DGU-403	NA
Pump	Shimadzu LC-40B XR	L22146350580
Autosampler	Shimadzu SIL-40C XR	L22216351622
Colum Thermostat	Shimadzu CTO-40S	L22236351602
PDA Detector	Shimadzu SPD-M40	L22276352808
SQ MS Detector	Shimadzu LCMS-2050	O12476200760

1.2. Chromatographic conditions

Chromatographic conditions	
Eluent A	0.1% DFA in Water (HPLC, Gradient Grade)
Eluent B	0.1% DFAin Acetonitrile (HPLC, Gradient Grade)
Flow rate	0.8 mL/min
Program	Gradient elution
Injection volume	2 µL
Colum Temperature	60°C
Column	Waters XSelect CSH C18, 100x2.1mm 2.5µm
Detection wavelength	225nm

Gradient Program		
Time [min]	A [%]	B [%]
1	98	2
13.99	35	65
14	5	95
15	5	95
15.01	98	2
19	end	

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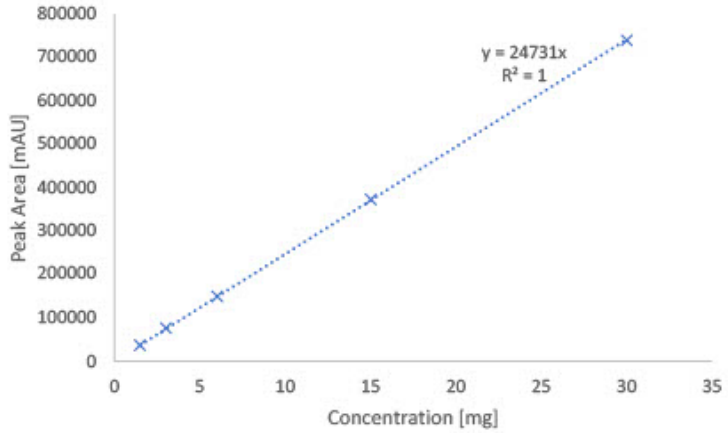
1.3. Sample preparation

Whole amount of container was dissolved in 2mL of water (LCMS Grade). 100 µL of sample was transferred to HPLC vial and diluted by 900 µL water (LCMS Grade) and submitted for analysis.

1.4. Calibration curve

Calibration curve detail	
Quantitative method	External Standard
Calibration Type	Linear
Number of calibration points	5
Force through Zero	Enabled
Weighting Method	None



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2. Purity assessment

2.1 Instrumentation

Module	Name	Serial Number
System Controller	Shimadzu CBM-40 Lite	L221226351398
Degassing Unit	Shimadzu DGU-403	NA
Pump	Shimadzu LC-40B XR	L22146350580
Autosampler	Shimadzu SIL-40C XR	L22216351622
Colum Thermostat	Shimadzu CTO-40S	L22236351602
PDA Detector	Shimadzu SPD-M40	L22276352808
SQ MS Detector	Shimadzu LCMS-2050	O12476200760

2.2 Chromatographic conditions

Chromatographic conditions	
Eluent A	0.1% DFA in Water (HPLC, Gradient Grade)
Eluent B	0.1% DFAin Acetonitrile (HPLC, Gradient Grade)
Flow rate	0.8 mL/min
Program	Gradient elution
Injection volume	2 µL
Colum Temperature	60°C
Column	Waters XSelect CSH C18, 100x2.1mm 2.5µm
Detection wavelength	225nm

Gradient Program		
Time [min]	A [%]	B [%]
1	98	2
13.99	35	65
14	5	95
15	5	95
15.01	98	2
19	end	

2.3 Purity assesment

Purity of compound assesed by area normalization method, comparing area of each peak to sum of area of all peaks detected at wavelenght of 225 nm.

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3. Identity Assessment

3.1 Instrumentation

Module	Name	Serial Number
System Controller	Shimadzu CBM-40 Lite	L221226351398
Degassing Unit	Shimadzu DGU-403	NA
Pump	Shimadzu LC-40B XR	L22146350580
Autosampler	Shimadzu SIL-40C XR	L22216351622
Colum Thermostat	Shimadzu CTO-40S	L22236351602
PDA Detector	Shimadzu SPD-M40	L22276352808
SQ MS Detector	Shimadzu LCMS-2050	O12476200760

3.2 Chromatographic conditions

Chromatographic conditions	
Eluent A	0.1% DFA in Water (HPLC, Gradient Grade)
Eluent B	0.1% DFAin Acetonitrile (HPLC, Gradient Grade)
Flow rate	0.8 mL/min
Program	Gradient elution
Injection volume	2 µL
Colum Temperature	60°C
Column	Waters XSelect CSH C18, 100x2.1mm 2.5µm
Detection	MS 220-2000 Da

Gradient Program		
Time [min]	A [%]	B [%]
1	98	2
13.99	35	65
14	5	95
15	5	95
15.01	98	2
19	end	

3.3 Molecular Ion Mass evaluation

Molecular ion mass was determined by deconvolution of multiply charged ESI-MS spectra to calculate the average neutral (zero-charge) molecular mass by equation:

$$M(neutral) = (z_i((mz_i) - H)) - ME$$

Where:
mz_i - Measured mass of charged particle
z_i - charge
H - proton mass (1.0076 Da)
ME – mass error

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Analysis Report

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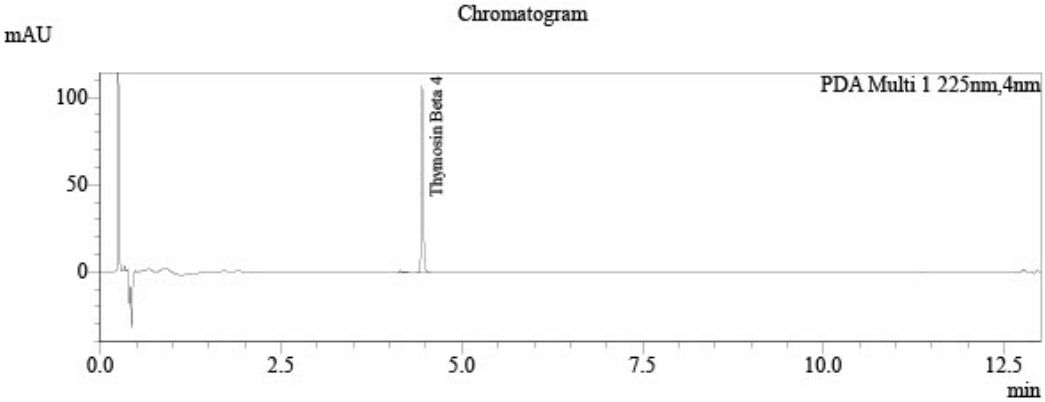
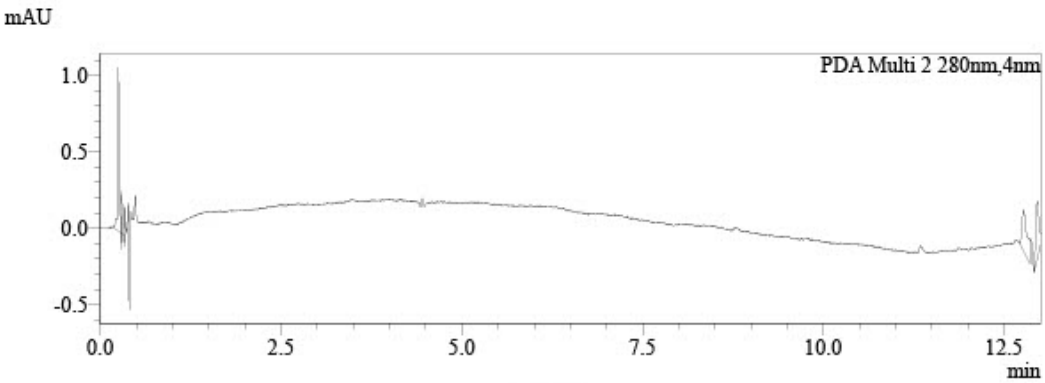
Analysis of quantity of active ingredient by UHPLC with MS detection

Sample Information

Injection Volume	: 2
Data File	: LYO-0932__015.lcd
Method File	: Peptide Screening_DFA_Group C.lcm
Date Acquired	: 9/18/2026 10:41:04 PM

Chromatogram





Peak Table

PDA Ch1 225nm

Peak#	Name	Ret. Time	Area	Area%	Conc.	Unit
1		4.149	1756	0.903	0.000	
2		4.205	133	0.068	0.000	
3	Thymosin Beta 4	4.453	192519	99.028	7.720	mg
Total			194408	100.000		

Peak Table

PDA Ch2 280nm

Peak#	Name	Ret. Time	Area	Area%	Conc.	Unit
1		0.253	1669	24.719	0.000	
2		12.768	1304	19.307	0.000	
3		12.957	1186	17.556	0.000	
4		14.230	2595	38.419	0.000	

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Peak#	Name	Ret. Time	Area	Area%	Conc.	Unit
Total			6754	100.000		

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Analysis Report

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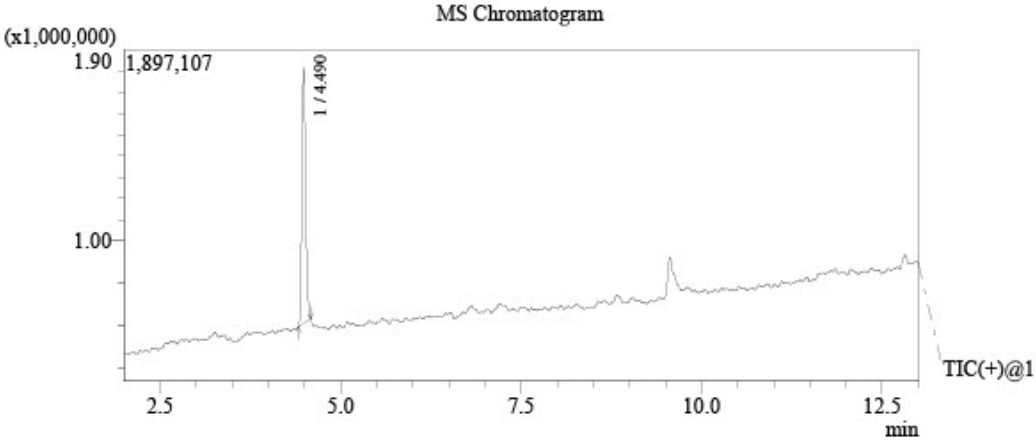
Identification of active ingredient by UHPLC Mass Spectrometr

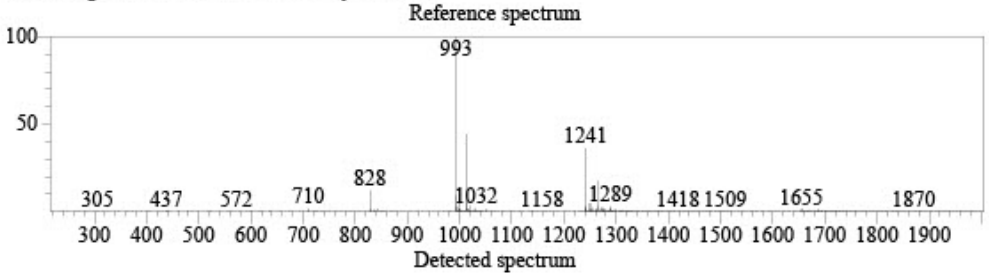
MS Chromatogram



Library Search

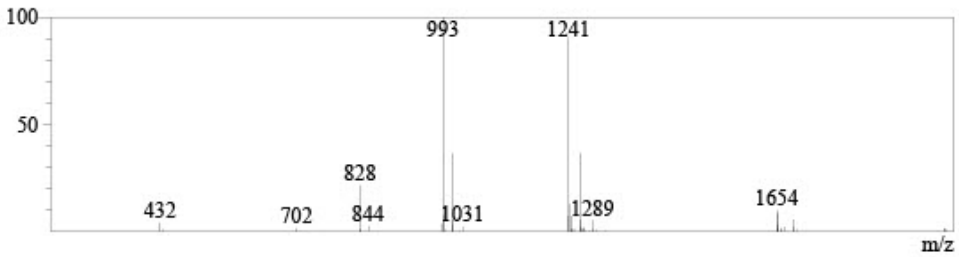
Base Peak: 993(329500)
Scan Range: 220.00 - 2000.00 Porality: Positive

Reference spectrum



Detected spectrum

Library: Peptide Screening_DFA.lib
Formula: C212H350N56O78S CAS: 77591-33-4 Mol.Weight: 4963(0!=)
Compound Name: Thymosin Beta 4



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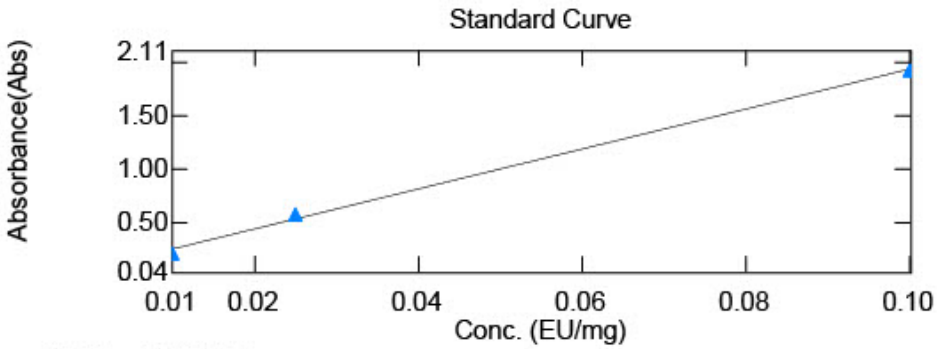
Endotoxin Determination Report

File Information
Filename: C:\UVVis-Data\Data\File_260918-2_154452.vqud
Date/Time: 09/18/2026 04:54:59 PM

Instrument Information
Instrument Type: UV-1900 Series
Model (S/N): UV-1900i (A12536253123)



y = 18.7714 x + 0.0637890
r2 = 0.99801

	Sample Name	Conc	Raw_WL545.0	Dilution Rate
1	LYO-0926	0.022	0.0847	20.00
2	LYO-0927	0.022	0.0840	20.00
3	LYO-0628	3.762	1.8295	40.00
4	LYO-0929	0.146	0.2004	20.00
5	LYO-0930	0.031	0.0928	20.00
6	LYO-0931	3.089	1.5134	40.00
7	LYO-0932	2.149	1.0724	40.00
8	LYO-0933	0.428	0.4655	20.00
9	LYO-0934	0.251	0.2992	20.00
10	LYO-0935	0.037	0.0986	20.00
11	LYO-0936	0.171	0.2243	20.00
12	LYO-0937	0.036	0.0974	20.00
13	LYO-0938	1.105	0.5824	40.00
14	LYO-0939	3.203	1.5670	40.00
15	LYO-0940	2.518	1.2455	40.00
16	LYO-0941	2.266	1.1272	40.00
17	LYO-0942	0.079	0.1380	20.00
18	LYO-0943	0.034	0.0959	20.00
19	LYO-0944	0.031	0.0928	20.00
20	LYO-0945	0.022	0.0849	20.00

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	Sample Name	Conc	Raw_WL545.0	Dilution Rate
21	LYO-0946	0.042	0.1031	20.00
22	LYO-0947	0.106	0.1637	20.00
23	LYO-0948	0.019	0.0820	20.00
24	LYO-0949	0.013	0.0763	20.00
25	LYO-0950	0.079	0.1382	20.00
26	LYO-0951	0.087	0.1458	20.00
27	LYO-0952	0.045	0.1057	20.00
28	LYO-0953	0.045	0.1062	20.00
29	LYO-0954	0.036	0.0976	20.00
30	LYO-0966	3.815	1.8541	40.00
31	LYO-0967	0.057	0.1169	20.00
32	LYO-0955	20.436	1.2626	320.00
33	LYO-0956	23.200	1.1525	400.00
34	LYO-0957	1.027	0.5456	40.00
35	LYO-0958	3.753	1.8249	40.00
36	LYO-0959	4.137	2.0051	40.00
37	LYO-0960	0.014	0.0770	20.00
38	LYO-0961	0.013	0.0757	20.00
39	LYO-0962	0.046	0.1069	20.00
40	LYO-0963	0.059	0.0748	100.00

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	Sample Name	Conc	Raw_WL545.0	Dilution Rate
41	LYO-0964	0.132	0.0885	100.00
42	LYO-0965	0.274	0.1153	100.00

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Responsibles



Mr. Ján Galbavý
CEO

Analysis results relate only to the samples tested.
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