

## Supporting Information

### **Development of a Rapid Adeno-Associated Virus (AAV) Identity Testing Platform through Comprehensive Intact Mass Analysis of Full-length AAV Capsid Proteins**

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## Supplementary Information 1: Detailed AAV SMART Digest peptide mapping protocol

### AAV sample preparation for pepsin digestion on a KingFisher Duo prime

1. For a single digest, 4 µg (35 µL) of AAV sample was transferred to a 500 µL Eppendorf tube and brought to a final volume of 50 µL with LC-MS grade water.
2. For AAV sample denaturation and reduction, 148 µL of SMART Digest™ buffer and 2 µL of TCEP (Bond-Breaker™ solution, 0.5M) were added to the Eppendorf tube containing the AAV sample so that the final solution volume was 200 µL with a final TCEP concentration of 5 mM.
3. In a separate 500 µL tube, 15 µL of SMART Digest™ Pepsin beads and then 100 µL of SMART Digest Buffer™ were added and mixed to create a pepsin bead mixture for digestion
4. In a third 500 µL tube a pepsin bead wash solution was prepared by mixing 50 µL of SMART Digest™ Buffer with 150 µL of LC-MS grade water.
5. All 200 µL of the final AAV sample from Step 2 was transferred into well A1 of a KingFisher™ deepwell 96 plate (Thermo Fisher Scientific, Vantaa, Finland) with subsequently prepared AAV samples transferred into wells A2 and A3. 100 µL of the pepsin bead mixture prepared in Step 3 was added to well D1 of the same deepwell plate, with subsequently prepared pepsin bead mixtures transferred into wells D2 and D3. All 200 µL of the pepsin bead wash solution prepared in Step 4 was then added to well E1 of the aforementioned deepwell plate, with subsequently prepared wash solutions transferred into wells E2 and E3. Finally, a 12-tip comb for microtiter 96 deepwell plate was placed in Row B to complete plate setup for AAV pepsin digestion on a KingFisher™ Duo Prime purification system under the control of Thermo Scientific BindIt software, version 4.0(see plate design below).

### King Fisher Plate Design

| Row / Column                | 1    | 2    | 3    | 4    | 5    | 6    | 7    | 8    | 9    | 10   | 11   | 12   |
|-----------------------------|------|------|------|------|------|------|------|------|------|------|------|------|
| A: AAV Sample w/ TCEP (µL)  | 200  | 200  | 200  |      |      |      |      |      |      |      |      |      |
| B: 12-tip cone (Tips)       | Tips | Tips | Tips | Tips | Tips | Tips | Tips | Tips | Tips | Tips | Tips | Tips |
| C: (Empty)                  |      |      |      |      |      |      |      |      |      |      |      |      |
| D: Pepsin bead mixture (µL) | 100  | 100  | 100  |      |      |      |      |      |      |      |      |      |
| E: Pepsin bead wash (µL)    | 200  | 200  | 200  |      |      |      |      |      |      |      |      |      |
| F                           |      |      |      |      |      |      |      |      |      |      |      |      |
| G                           |      |      |      |      |      |      |      |      |      |      |      |      |
| H                           |      |      |      |      |      |      |      |      |      |      |      |      |

**Table S1: BPF 4.1 software parameter settings for peptide mapping analysis**

| <b>Component Detection Parameters</b>                                       |  |
|---|--|
| Absolute MS Signal Threshold (MS Noise Level * S/N Threshold)               | 250000   |
| MS Noise Level  | 1000   |
| S/N Threshold   | 250  |
| Beginning Peak Width (min)  | 0.1  |
| Typical Chromatographic Peak Width (min)                                    | 0.3  |
| Ending Peak Width (min)   | 0.75   |
| Maximum Chromatographic Peak Width (min)                                    | 0.5  |
| Use Restricted Time   | Yes  |
| Time Limits   | 5 to 115   |
| Relative MS Signal Threshold (% of base peak)                               | 1  |
| Relative Analog Threshold (% of highest peak)                               | 1  |
| Width of Gaussian Filter (represented as 1/n of chromatographic peak width) | 3  |
| Minimum Valley to be Considered as Two Chromatographic Peaks (%)            | 80.00%   |
| Minimum MS Peak Width (Da)  | 1.2  |
| Maximum MS Peak Width (Da)  | 4.2  |
| Mass Tolerance (ppm for high-res or Da for low-res)                         | 4  |
| Maximum Retention Time Shift (min)  | 1  |
| Maximum Mass (Da)   | 30000  |
| Mass Centroiding Cutoff (% from base)                                       | 15   |
| <b>Identification Parameters</b>  |  |
| Search by Full MS Only  | No   |
| Use MS/MS   | Use All MS/MS  |
| Maximum Peptide Mass  | 7000   |
| Mass Accuracy (ppm)   | 5  |
| Minimum Confidence  | 0.8  |
| Maximum Number of Modifications for a Peptide                               | 1  |
| Mass Changes for Unspecified Modifications                                  | -129 to 169  |
| Glycosylation   | Human  |
| Search for Amino Acid Substitutions   | None   |
| Enable Residue Deletion   | False  |
| Perform Disulfide Bond Search   | No   |
| Protease  | Pepsin   |
| N-Term  | FIMYWVL  |
| C-Term  | CDEFLMTWY  |
| Specificity   | High   |
| Static Modifications  | None   |
| Variable Modifications  | Acetylation (N-term), Acetylation (SideChain), Deamidation (N), Deamidation (Q), Dimethylation (K), Glycation (K), Oxidation (C), Oxidation (M,W), Phosphorylation (S,T,Y), Phospho S (S,T,Y), SUMOylation by SUMO1 (K), SUMOylation by SUMO2 (K), Ubiquitination (K), Amidation (D,E), Methylation (K,R), Formylation (S,T), Double Oxidation (M,W,C), Oxidation to hydroxy-kynurenine (W), Oxidation to kynurenine (W), Double Phosphorylation (S,T,Y), Triple Phosphorylation (S,T,Y), Phosphorylation1x_Oxidation1x (S,T,Y,M,W,C), Cysteaminylation (C), Double Deamidation (N,Q, Succinimide (N), Succinimide (D), NH3 loss_Glutarimide (Q) |



**Table S2: BPF 4.1 software parameter settings for intact mass analysis**

| <b>Component Detection Parameters</b>            |  |
|--|--|
| <b>Chromatogram Parameters</b>                   |  |
| Use Restricted Time                              | Enabled  |
| Time Limits                                      | 20.4 to 31   |
| m/z Range  | 1100 to 2100 (15,000 Res)*, 867 to 2100 (45,000 Res) |
| Chromatogram Trace Type                          | TIC  |
| Sensitivity                                      | High   |
| Rel. Intensity Threshold (%)                     | 1  |
| Source Spectra Method                            | Sliding Windows                                      |
| <b>Sliding Windows Definition</b>                |  |
| RT Range   | 20.4 to 31   |
| Target Avg Spectrum Width                        | 0.6  |
| Target Avg Spectrum Offset Scan                  | 1  |
| <b>Sliding Windows Merging Parameters</b>        |  |
| Merge Tolerance                                  | 5 ppm  |
| Merge Scheme                                     | Improved Merge Scheme                                |
| Max RT Gap                                       | 0.1 minutes  |
| Min. Number of Detected Intervals                | 6  |
| Deconvolution Algorithm                          | ReSpect™ (Isotopically Unresolved)                   |
| <b>ReSpect Main Parameters</b>                   |  |
| <b>Deconvolution Results Filter</b>              |  |
| Output Mass Range                                | Min 50000 , Max 85000                                |
| Deconvoluted Spectra Display Mode                | Isotopic Profile (new)                               |
| <b>Charge State Distribution</b>                 |  |
| Deconvolution Mass Tolerance                     | 10 ppm   |
| <b>Choice of Peak Model</b>                      |  |
| Choice of Peak Model                             | Intact Protein                                       |
| Resolution at 400 m/z                            |  |
| Raw File Specific                                |  |
| <b>ReSpect Advanced Parameters</b>               |  |
| <b>Charge State Distribution</b>                 |  |
| Model Mass Range                                 | Min 10000 Max 1000000                                |
| Charge State Range                               | 5 to 100   |
| Minimum Adjacent Charges (low & high model mass) | 4 to 6   |
| <b>Noise Parameters</b>                          |  |
| Rel. Abundance Threshold (%)                     | 0  |
| <b>Deconvolution Quality</b>                     |  |
| Quality Score Threshold                          | 0  |
| <b>Choice of Peak Model</b>                      |  |
| Target Mass                                      | 71000 Da   |
| <b>Peak Model Parameters</b>                     |  |
| Number of Peak Models                            | 1  |
| Left/Right Peak Shape                            | Left 2 Right 2                                       |
| <b>Peak Filter Parameters</b>                    |  |
| Peak Detection Minimum Significance Measure      | 1  |
| Peak Detection Quality Measure                   | 95%  |
| <b>Specialized Parameters</b>                    |  |
| Peak Model Width Factor                          | 1  |
| Intensity Threshold Scale                        | 0.01   |
| <b>Deconvolution Parameters</b>                  |  |
| Noise Compensation                               | Enabled  |
| Charge Carrier                                   | H+ (1.00727663)                                      |
| Negative Charge                                  | Disabled   |
| <b>Identification Parameters</b>                 |  |
| Sequence Matching Mass Tolerance                 | 100 ppm (15,000 Res**), 50 ppm (45,000 Res)          |
| Enable Drug-to-Antibody Ratio (Protein Only)     | Disabled   |
| Variable Modification Selected                   |  |
| <b>Multiconsensus Component Merge</b>            |  |
| Mass Tolerance                                   | 10 ppm   |
| RT Tolerance                                     | 0.1 minutes  |
| Minimum Number of Required Occurrences           | 1  |

**Table S2 Continued:**

| <b>Sequences, Modifications</b> |  |
|---------------------------------|--|
| Sequence Name                   | AAV2_VP1   |
| Static Modifications            | Acetylation (N-term)(NTerm,A,x1)   |
| Sequence Name                   | AAV2_VP2   |
| Static Modifications            | None   |
| Sequence Name                   | AAV2_VP3   |
| Static Modifications            | Acetylation (N-term)(NTerm,A,x1)   |
| Sequence Name                   | AAV2 VP3 Variant   |
| Static Modifications            | Acetylation (N-term)(NTerm,A,x1)   |
| Variable Modifications          | Acetylation (SideChain), Deamidation (N), Oxidation (M,W), Phosphorylation (S,T,Y), Double Oxidation (M,W,C), Triple Oxidation (M,W,C) Oxidation to kynurenine (W), Double Phosphorylation (S,T,Y), Triple Phosphorylation (S,T,Y), Phosphorylation1x_Oxidation1x (S,T,Y,M,W,C), Double Deamidation (N,Q), Succinimide (D), NH3 loss_Glutarimide (Q) |
| Max # Modifications             | 3  |
| Glycosylation                   | None   |

\* m/z range needed to minimize impact of high background ions in mass spectra from mobile phase

\*\* Needed to expand tolerance from 50 ppm to 100 ppm to detect oxidation on VP2 and VP1

**Table S3: BPF 4.1 software parameters for detection of components under VP3 Prime peak\***

| <b>Component Detection Parameters</b>     |  |
|---|--|
| <b>Chromatogram Parameters</b>            |  |
| Time Limits                               | 22.5 to 24.0 (45°C, 15,000 Res), 25.2 to 26.7 (25°C, 45,000 Res) |
| m/z Range                                 | 867 to 2100  |
| <b>Sliding Windows Definition</b>         |  |
| RT Range                                  | 22.5 to 24.0 (45°C, 15,000 Res), 25.2 to 26.7 (25°C, 45,000 Res) |
| Target Avg Spectrum Width                 | 0.3  |
| <b>Sliding Windows Merging Parameters</b> |  |
| Merge Tolerance                           | 20 ppm   |
| Min. Number of Detected Intervals         | 3  |
| <b>ReSpect Advanced Parameters</b>        |  |
| <b>Specialized Parameters</b>             |  |
| Intensity Threshold Scale                 | 0.005  |

\*The BPF 4.1 software parameters used for the determination of components within the VP3 Prime peak are the same as those described in Table S2 for intact mass analysis except for the parameters outlined in the table above.

**Table S4: Rapid AAV Identity Testing Method Data Processing Parameters**

| <b>MS Detection (Tab)</b>              |  |
|--|--|
| <b>Extracted Ion Chromatogram</b>      | MS Default Detection Settings  |
| <b>Detection Algorithm</b>             | Cobra  |
| <b>Parameter: Consider Void Peak</b>   |  |
| Retention Time                         | Initial  |
| Parameter Value                        | Off  |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>Parameter: Baseline Noise Range</b> |  |
| Retention Time                         | Initial  |
| Parameter Value                        | Auto   |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>Parameter: Smoothing Width</b>      |  |
| Retention Time                         | Initial  |
| Parameter Value                        | 0.500 minutes  |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>Parameter: Minimum Area</b>         |  |
| Retention Time                         | 0.000 min  |
| Parameter Value                        | 7000 Signal*min  |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>MS Component Table (Tab)</b>        |  |
| <b>Component: AAV2_VP1</b>             |  |
| Retention Time                         | 25.180   |
| Window                                 | 0.600 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1388.43359, 1545.44517, 1489.33590, 1342.91721, 1365.28302, 1412.36224 |
| MS Confirming Peak 1                   | 1388.43359   |
| MS Confirming Peak 2                   | 1545.44517   |
| MS Confirming Peak 3                   | 1489.33590   |
| MS Confirming Peak 4                   | 1342.91721   |
| MS Confirming Peak 5                   | 1365.28302   |
| MS Confirming Peak 6                   | 1412.36224   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 81854.900000   |
| <b>Component: AAV2_VP2</b>             |  |
| Retention Time                         | 26.200   |
| Window                                 | 0.600 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1415.63640, 1446.41490, 1255.53048, 1478.50577, 1279.60936, 1304.68622 |
| MS Confirming Peak 1                   | 1415.63640   |
| MS Confirming Peak 2                   | 1446.41490   |
| MS Confirming Peak 3                   | 1255.53048   |
| MS Confirming Peak 4                   | 1478.50577   |
| MS Confirming Peak 5                   | 1279.60936   |
| MS Confirming Peak 6                   | 1304.68622   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 66487.900000   |
| <b>Component: AAV2_VP3</b>             |  |
| Retention Time                         | 20.380   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1428.95544, 1500.35458, 1463.78919, 1395.74845, 1364.05468, 1538.79198 |
| MS Confirming Peak 1                   | 1428.95544   |
| MS Confirming Peak 2                   | 1500.35458   |
| MS Confirming Peak 3                   | 1463.78919   |

**Table S4 Continued:**

|                            |  |
|----------------------------|--|
| MS Confirming Peak 4       | 1395.74845   |
| MS Confirming Peak 5       | 1364.05468   |
| MS Confirming Peak 6       | 1538.79198   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 59973.780000   |
| <b>Component: AAV5_VP1</b> |  |
| Retention Time             | 24.090   |
| Window                     | 0.600 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1362.62784, 1410.41467, 1435.57674, 1317.99600, 1339.94862, 1488.69741 |
| MS Confirming Peak 1       | 1362.62784   |
| MS Confirming Peak 2       | 1410.41467   |
| MS Confirming Peak 3       | 1435.57674   |
| MS Confirming Peak 4       | 1317.99600   |
| MS Confirming Peak 5       | 1339.94862   |
| MS Confirming Peak 6       | 1488.69741   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 80335.080000   |
| <b>Component: AAV5_VP2</b> |  |
| Retention Time             | 25.160   |
| Window                     | 0.600 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1389.99731, 1519.19696, 1484.69186, 1420.19615, 1555.34834, 1451.72711 |
| MS Confirming Peak 1       | 1389.99731   |
| MS Confirming Peak 2       | 1519.19696   |
| MS Confirming Peak 3       | 1484.69186   |
| MS Confirming Peak 4       | 1420.19615   |
| MS Confirming Peak 5       | 1555.34834   |
| MS Confirming Peak 6       | 1451.72711   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 65282.690000   |
| <b>Component: AAV5_VP3</b> |  |
| Retention Time             | 14.530   |
| Window                     | 1.000 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1525.66894, 1565.78381, 1608.07850, 1451.30371, 1487.55296, 1416.76686 |
| MS Confirming Peak 1       | 1525.66894   |
| MS Confirming Peak 2       | 1565.78381   |
| MS Confirming Peak 3       | 1608.07850   |
| MS Confirming Peak 4       | 1451.30371   |
| MS Confirming Peak 5       | 1487.55296   |
| MS Confirming Peak 6       | 1416.76686   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 59462.370000   |
| <b>Component: AAV8_VP1</b> |  |
| Retention Time             | 23.330   |
| Window                     | 0.600 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1409.08000, 1459.38216, 1385.20869, 1513.38797, 1184.60525, 1339.82767 |
| MS Confirming Peak 1       | 1409.08000   |
| MS Confirming Peak 2       | 1459.38216   |
| MS Confirming Peak 3       | 1385.20869   |
| MS Confirming Peak 4       | 1513.38797   |
| MS Confirming Peak 5       | 1184.60525   |

**Table S4 Continued:**

|                            |  |
|----------------------------|--|
| MS Confirming Peak 6       | 1339.82767   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 81666.710000   |
| <b>Component: AAV8_VP2</b> |  |
| Retention Time             | 24.500   |
| Window                     | 0.600 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1386.80868, 1358.52407, 1331.39296, 1305.30768, 1416.29428, 1623.39744 |
| MS Confirming Peak 1       | 1386.80868   |
| MS Confirming Peak 2       | 1358.52407   |
| MS Confirming Peak 3       | 1331.39296   |
| MS Confirming Peak 4       | 1305.30768   |
| MS Confirming Peak 5       | 1416.29428   |
| MS Confirming Peak 6       | 1623.39744   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 66518.100000   |
| <b>Component: AAV8_VP3</b> |  |
| Retention Time             | 15.730   |
| Window                     | 1.000 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1574.80007, 1496.10855, 1534.44183, 1459.64505, 1273.44472, 1391.80234 |
| MS Confirming Peak 1       | 1574.80007   |
| MS Confirming Peak 2       | 1496.10855   |
| MS Confirming Peak 3       | 1534.44183   |
| MS Confirming Peak 4       | 1459.64505   |
| MS Confirming Peak 5       | 1273.44472   |
| MS Confirming Peak 6       | 1391.80234   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 59804.680000   |
| <b>Component: AAV9_VP1</b> |  |
| Retention Time             | 23.160   |
| Window                     | 0.600 AG   |
| Channel                    | All Channels   |
| Factor                     | 1.000000   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1378.81961, 1333.65071, 1355.86805, 1402.58411, 1534.80787, 1452.63616 |
| MS Confirming Peak 1       | 1378.81961   |
| MS Confirming Peak 2       | 1333.65071   |
| MS Confirming Peak 3       | 1355.86805   |
| MS Confirming Peak 4       | 1402.58411   |
| MS Confirming Peak 5       | 1534.80787   |
| MS Confirming Peak 6       | 1452.63616   |
| Display Window             | RW   |
| Display Time               | RT   |
| Molecular mass             | 81290.340000   |
| <b>Component: AAV9_VP2</b> |  |
| Retention Time             | 24.100   |
| Window                     | 0.600 AG   |
| Channel                    | All Channels   |
| Evaluation Type            | Area   |
| Peak Type                  | Autodetect   |
| MS Quantitation Peaks      | 1409.72987, 1162.58873, 1472.33406, 1274.28659, 1380.37065, 1352.23093 |
| MS Confirming Peak 1       | 1409.72987   |
| MS Confirming Peak 2       | 1162.58873   |
| MS Confirming Peak 3       | 1472.33406   |
| MS Confirming Peak 4       | 1274.28659   |
| MS Confirming Peak 5       | 1380.37065   |
| MS Confirming Peak 6       | 1352.23093   |
| Display Window             | RW   |

**Table S4 Continued:**

|   |  |
|---|--|
| Display Time  | RT   |
| Molecular mass  | 66209.760000   |
| <b>Component: AAV9_VP3</b>  |  |
| Retention Time  | 18.150   |
| Window  | 1.000 AG   |
| Channel   | All Channels   |
| Evaluation Type   | Area   |
| Peak Type   | Autodetect   |
| MS Quantitation Peaks   | 1358.58061, 1390.14960, 1299.55588, 1328.41535, 1271.92993, 1457.90702 |
| MS Confirming Peak 1  | 1358.58061   |
| MS Confirming Peak 2  | 1390.14960   |
| MS Confirming Peak 3  | 1299.55588   |
| MS Confirming Peak 4  | 1328.41535   |
| MS Confirming Peak 5  | 1271.92993   |
| MS Confirming Peak 6  | 1457.90702   |
| Display Window  | RW   |
| Display Time  | RT   |
| Molecular mass  | 59732.620000   |
| <b>MS Settings (Tab)</b>  |  |
| <b>MS Spectra Bunching</b>  |  |
| <i>Peak Spectrum Bunch</i>  |  |
| Width   | 1 spectra  |
| <i>Baseline Correction</i>  |  |
| No Baseline Correction  | Selected   |
| <b>MS Noise Reduction</b>   |  |
| Do not reduce noise   | Selected   |
| <b>MS Chromatogram Settings</b>   |  |
| <i>Mass Precision</i>   |  |
| Decimal Places  | 5  |
| <i>Mass Tolerance</i>   |  |
| Mass Tolerance  | 20 ppm   |
| <i>Integration</i>  |  |
| Inhibit integration for TIC channel   | Unselected   |
| <i>Smoothing</i>  |  |
| Type  | Moving Average   |
| Points  | 21   |
| <b>Composite Scoring (Tab)</b>  |  |
| <b>Composite Scoring</b>  |  |
| <i>Scoring result on checked criteria</i>   |  |
| Pass score if at least  | 1 criteria is passed   |
| Fail score if less than   | 1 criteria is passed   |
| Indeterminate score if neither passed nor failed  |  |
| <i>MS Criteria</i>  |  |
| Intact Protein Deconvolution  |  |
| Match the following rules   | All  |
| (Component.massToChargeRatio-<br>ipdComponent.mass)/component.massToChargeRatio<br>*1000000 | Is between -25 and 25  |

**Table S5: AAV PTM Monitoring Method Data Processing Parameters**

| <b>MS Detection (Tab)</b>              |  |
|--|--|
| <b>Parameter: Consider Void Peak</b>   |  |
| Retention Time                         | Initial  |
| Parameter Value                        | Off  |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>Parameter: Baseline Noise Range</b> |  |
| Retention Time                         | Initial  |
| Parameter Value                        | Auto   |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>Parameter: Smoothing Width</b>      |  |
| Retention Time                         | Initial  |
| Parameter Value                        | 0.500 minutes  |
| Injection Type                         | Any  |
| Channel                                | All Channels   |
| <b>MS Component Table (Tab)</b>        |  |
| <b>Component: AAV2_VP1</b>             |  |
| Retention Time                         | 28.030   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1342.89589, 1388.38818, 1365.27091, 1300.30104, 1187.32055, 1279.99847 |
| MS Confirming Peak 1                   | 1342.89589   |
| MS Confirming Peak 2                   | 1388.38818   |
| MS Confirming Peak 3                   | 1365.27091   |
| MS Confirming Peak 4                   | 1300.30104   |
| MS Confirming Peak 5                   | 1187.32055   |
| MS Confirming Peak 6                   | 1279.99847   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 81854.900000   |
| <b>Component: AAV2_VP1_1xOxidation</b> |  |
| Retention Time                         | 28.030   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1343.19118, 1154.15007, 1388.69979, 1187.57166, 1412.61042, 1170.61591 |
| MS Confirming Peak 1                   | 1343.19118   |
| MS Confirming Peak 2                   | 1154.15007   |
| MS Confirming Peak 3                   | 1388.69979   |
| MS Confirming Peak 4                   | 1187.57166   |
| MS Confirming Peak 5                   | 1412.61042   |
| MS Confirming Peak 6                   | 1170.61591   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 81870.890000   |
| <b>Component: AAV2_VP2</b>             |  |
| Retention Time                         | 29.340   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1446.39156, 1415.62751, 1304.68346, 1478.50307, 1209.87072, 1386.16667 |
| MS Confirming Peak 1                   | 1446.39156   |
| MS Confirming Peak 2                   | 1415.62751   |
| MS Confirming Peak 3                   | 1304.68346   |
| MS Confirming Peak 4                   | 1478.50307   |
| MS Confirming Peak 5                   | 1209.87072   |
| MS Confirming Peak 6                   | 1386.16667   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 66487.900000   |

**Table S5 Continued:**

|  |  |
|--|--|
| <b>Component: AAV2_VP2_1xOxidation</b> |  |
| Retention Time                         | 29.380   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1446.76009, 1305.02224, 1167.73752, 1358.26268, 1147.62767, 1331.10684 |
| MS Confirming Peak 1                   | 1446.76009   |
| MS Confirming Peak 2                   | 1305.02224   |
| MS Confirming Peak 3                   | 1167.73752   |
| MS Confirming Peak 4                   | 1358.26268   |
| MS Confirming Peak 5                   | 1147.62767   |
| MS Confirming Peak 6                   | 1331.10684   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 66503.900000   |
| <b>Component: AAV2_VP3</b>             |  |
| Retention Time                         | 23.100   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1364.31094, 1500.57013, 1333.96633, 1395.92511, 1463.89207, 1225.37382 |
| MS Confirming Peak 1                   | 1364.31094   |
| MS Confirming Peak 2                   | 1500.57013   |
| MS Confirming Peak 3                   | 1333.96633   |
| MS Confirming Peak 4                   | 1395.92511   |
| MS Confirming Peak 5                   | 1463.89207   |
| MS Confirming Peak 6                   | 1225.37382   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 59973.780000   |
| <b>Component: AAV2_VP3_Variant</b>     |  |
| Retention Time                         | 22.890   |
| Window                                 | 0.600 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1447.37441, 957.48841, 1380.09043, 1163.73649, 1099.11976, 973.14452   |
| MS Confirming Peak 1                   | 1447.37441   |
| MS Confirming Peak 2                   | 957.48841  |
| MS Confirming Peak 3                   | 1380.09043   |
| MS Confirming Peak 4                   | 1163.73649   |
| MS Confirming Peak 5                   | 1099.11976   |
| MS Confirming Peak 6                   | 973.14452  |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 59301.030000   |
| <b>Component: AAV2_VP3_1xOxidation</b> |  |
| Retention Time                         | 23.370   |
| Window                                 | 1.000 AG   |
| Channel                                | All Channels   |
| Evaluation Type                        | Area   |
| Peak Type                              | Autodetect   |
| MS Quantitation Peaks                  | 1277.42805, 1396.17933, 1464.22888, 1250.82261, 1305.18328, 1225.32372 |
| MS Confirming Peak 1                   | 1277.42805   |
| MS Confirming Peak 2                   | 1396.17933   |
| MS Confirming Peak 3                   | 1464.22888   |
| MS Confirming Peak 4                   | 1250.82261   |
| MS Confirming Peak 5                   | 1305.18328   |
| MS Confirming Peak 6                   | 1225.32372   |
| Display Window                         | RW   |
| Display Time                           | RT   |
| Molecular mass                         | 59989.780000   |



**Table S5 Continued:**

|   |  |
|---|--|
| <b>Component: AAV2_VP3_1xSuccinimide2</b>   |  |
| Retention Time  | 23.090   |
| Window  | 1.000 AG   |
| Channel   | All Channels   |
| Evaluation Type   | Area   |
| Peak Type   | Autodetect   |
| MS Quantitation Peaks   | 952.67401, 923.37411, 968.01959, 937.78948, 1017.18192, 1000.24990     |
| MS Confirming Peak 1  | 952.67401  |
| MS Confirming Peak 2  | 923.37411  |
| MS Confirming Peak 3  | 968.01959  |
| MS Confirming Peak 4  | 937.78948  |
| MS Confirming Peak 5  | 1017.18192   |
| MS Confirming Peak 6  | 1000.24990   |
| Display Window  | RW   |
| Display Time  | RT   |
| Molecular mass  | 59955.770000   |
| <b>Component: AAV2_VP3_2xOxidation</b>  |  |
| Retention Time  | 23.330   |
| Window  | 1.700 AG   |
| Channel   | All Channels   |
| Evaluation Type   | Area   |
| Peak Type   | Autodetect   |
| MS Quantitation Peaks   | 1133.19273, 1092.02719, 1112.21724, 1072.51992, 1539.66503, 1464.60605 |
| MS Confirming Peak 1  | 1133.19273   |
| MS Confirming Peak 2  | 1092.02719   |
| MS Confirming Peak 3  | 1112.21724   |
| MS Confirming Peak 4  | 1072.51992   |
| MS Confirming Peak 5  | 1539.66503   |
| MS Confirming Peak 6  | 1464.60605   |
| Display Window  | RW   |
| Display Time  | RT   |
| Molecular mass  | 60005.780000   |
| <b>MS Settings (Tab)</b>  |  |
| <b>MS Spectra Bunching</b>  |  |
| <i>Peak Spectrum Bunch</i>  |  |
| Width   | 1 spectra  |
| <i>Baseline Correction</i>  |  |
| No Baseline Correction  | Selected   |
| <b>MS Noise Reduction</b>   |  |
| Do not reduce noise   | Selected   |
| <b>MS Chromatogram Settings</b>   |  |
| <i>Mass Precision</i>   |  |
| Decimal Places  | 5  |
| <i>Mass Tolerance</i>   |  |
| Mass Tolerance  | 20 ppm   |
| <i>Integration</i>  |  |
| Inhibit integration for TIC channel   | Unselected   |
| <i>Smoothing</i>  |  |
| Type  | Moving Average   |
| Points  | 21   |
| <b>Composite Scoring (Tab)</b>  |  |
| <b>Composite Scoring</b>  |  |
| <i>Scoring result on checked criteria</i>   |  |
| Pass score if at least  | 1 criteria is passed   |
| Fail score if less than   | 1 criteria is passed   |
| Indeterminate score if neither passed nor failed  |  |
| <i>MS Criteria</i>  |  |
| Intact Protein Deconvolution  |  |
| Match the following rules   | All  |
| (Component.massToChargeRatio-<br>ipdComponent.mass)/component.massToChargeRatio<br>*1000000 | Is between -35 and 35  |

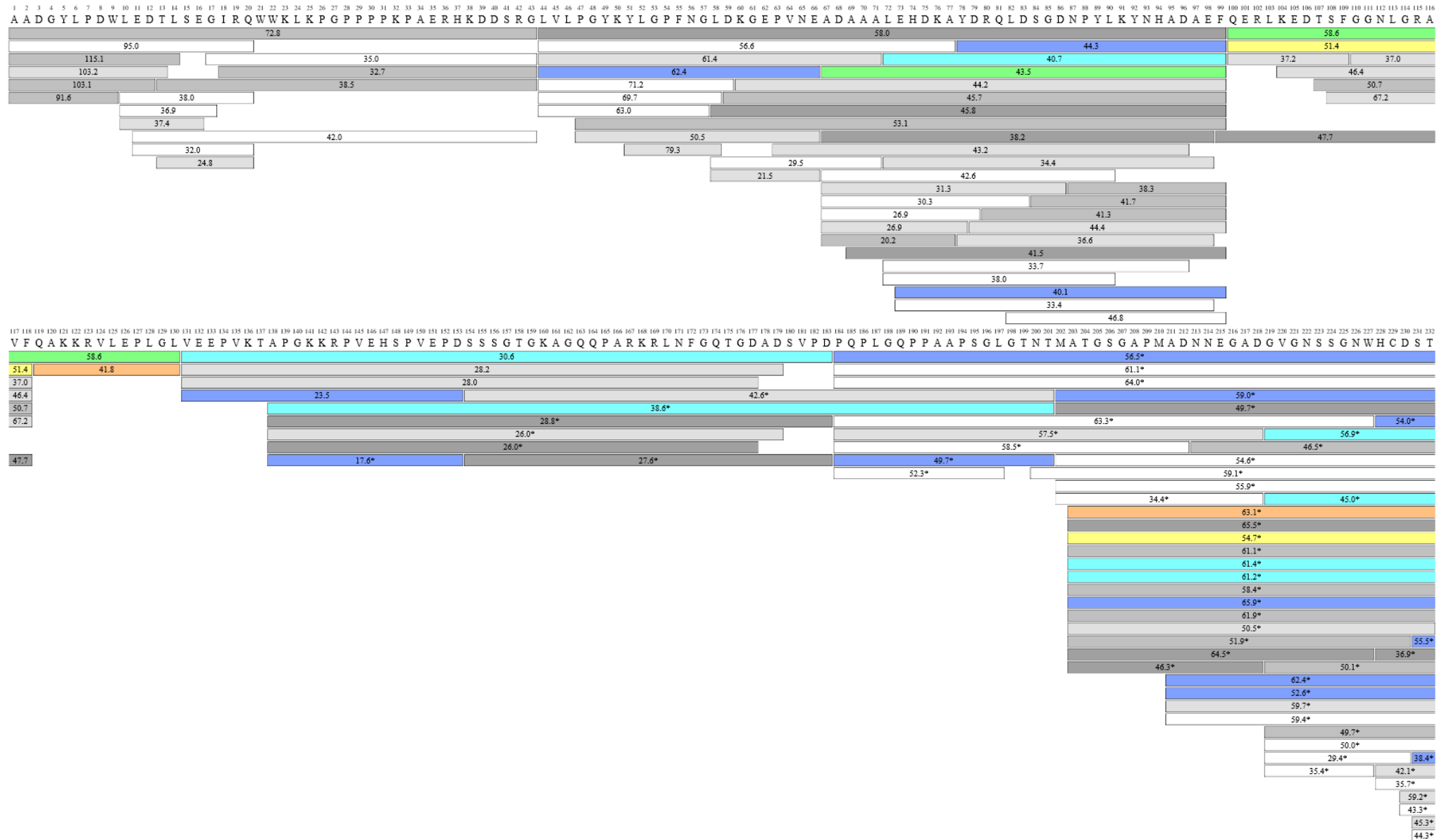
**Table S6:** Acetylated peptides illustrating the presence of the VP3 variant A211(Ac) – L735\*

| Peptide Sequence                              | Positions | Modification | Site | Delta (ppm) | Confidence Score | ID Type | RT    | M/Z      | Charge State | Mono Mass Exp. | Mono Mass Theo. |
|---|-----------|--------------|------|-------------|------------------|---------|-------|----------|--------------|----------------|-----------------|
| ADNNEGADGVGNSSGNWHCDSTWMGDRVIT                | 211-240   | Acetylation  | A211 | -0.22       | 100              | MS2     | 59.42 | 1070.445 | 3            | 3206.3047      | 3206.3054       |
| ADNNEGADGVGNSSGNWHCDSTWMGDRVITT               | 211-241   | Acetylation  | A211 | -1.2        | 100              | MS2     | 59.67 | 1103.792 | 3            | 3307.3491      | 3307.3531       |
| ADNNEGADGVGNSSGNWHCDSTWMGDRVITTSTRT           | 211-245   | Acetylation  | A211 | -0.14       | 100              | MS2     | 52.64 | 939.403  | 4            | 3752.5811      | 3752.5816       |
| ADNNEGADGVGNSSGNWHCDSTWMGDRVITTSTRT           | 211-245   | Acetylation  | A211 | -0.33       | 100              | MS2     | 52.65 | 1252.535 | 3            | 3752.5803      | 3752.5816       |
| ADNNEGADGVGNSSGNWHCDSTWMGDRVITTSTRTWALPTYNNHL | 211-255   | Acetylation  | A211 | 0.81        | 100              | MS2     | 62.45 | 1242.304 | 4            | 4962.1787      | 4962.1747       |
| ADNNEGADGVGNSSGNWHCDSTWMGDRVITTSTRTWALPTYNNHL | 211-255   | Acetylation  | A211 | 0.72        | 100              | MS2     | 62.45 | 993.843  | 5            | 4962.1782      | 4962.1747       |

\*Peptides and acetylation PTM were identified using BioPharma Finder

| Color code for peptide recovery |        |        |       |       |       |
|---------------------------------|--------|--------|-------|-------|-------|
| >50.0%                          | >20.0% | >10.0% | >5.0% | >2.0% | >1.0% |
| good                            | fair   |        |       |       | poor  |

VPI



**Figure S1: Sequence coverage map for AAV2 capsid protein peptides.** Peptides are separated on a PepMap nano RSLC C18 column after pepsin digestion. The coloured bars show the identified peptides, with the numbers in the bars reflecting the retention time. The different colours indicate the peptide recovery in the MS1 scan as illustrated in the “Colour code for peptide recovery” box.

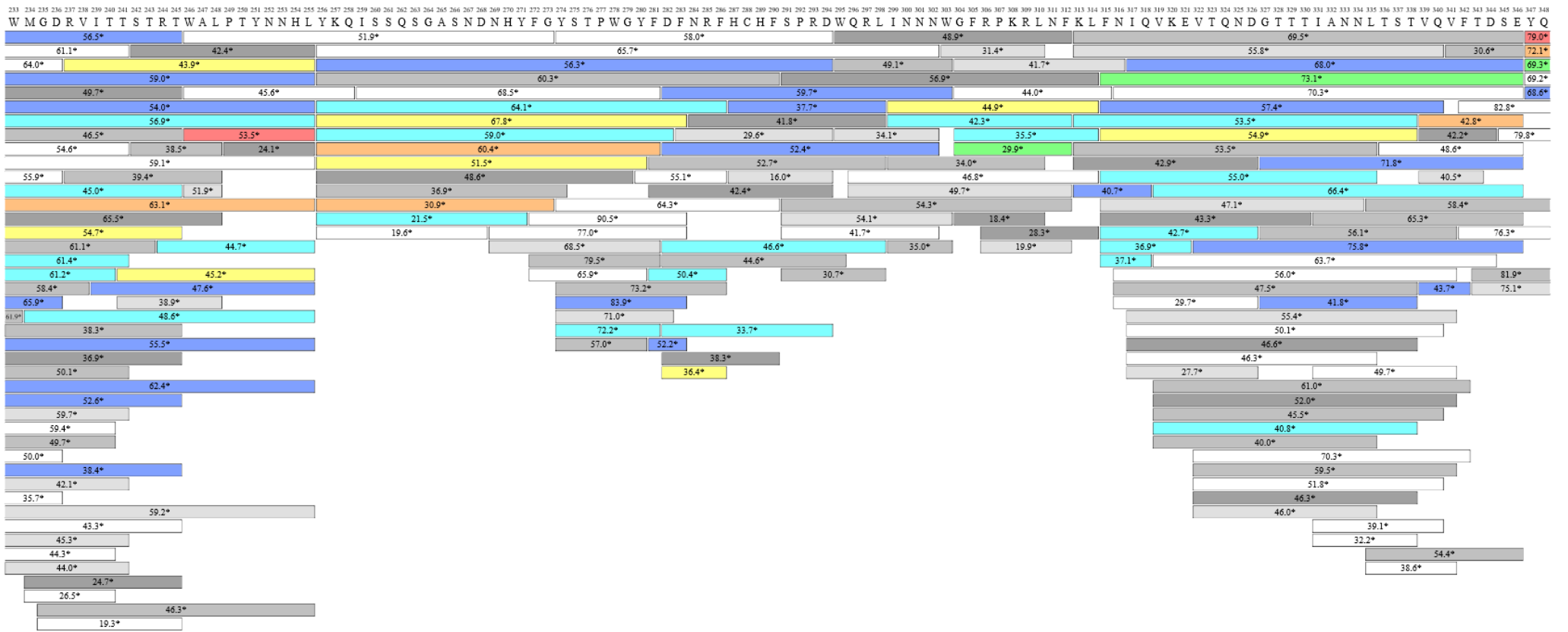


Figure S1 Continued:

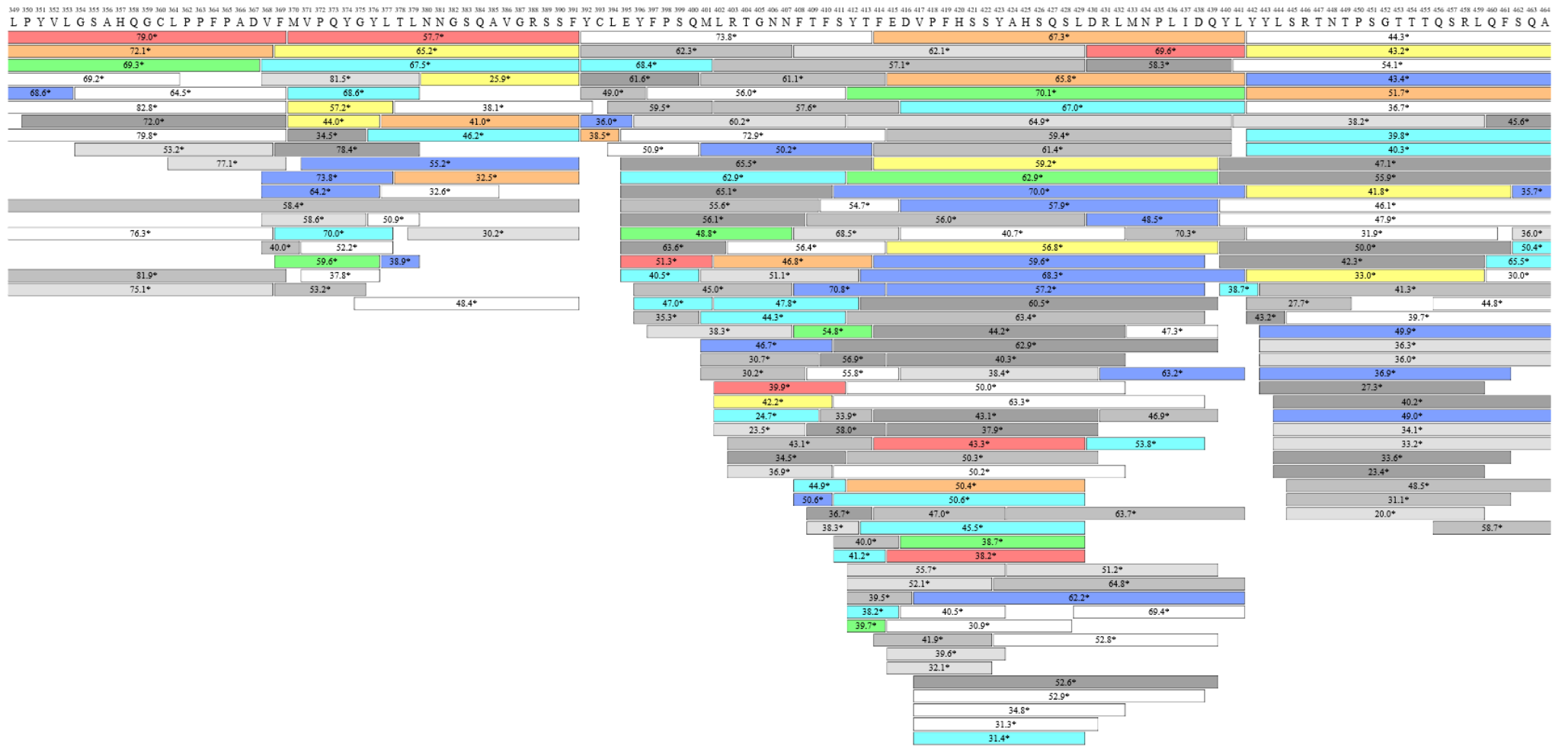


Figure S1 Continued:

465 466 467 468 469 470 471 472 473 474 475 476 477 478 479 480 481 482 483 484 485 486 487 488 489 490 491 492 493 494 495 496 497 498 499 500 501 502 503 504 505 506 507 508 509 510 511 512 513 514 515 516 517 518 519 520 521 522 523 524 525 526 527 528 529 530 531 532 533 534 535 536 537 538 539 540 541 542 543 544 545 546 547 548 549 550 551 552 553 554 555 556 557 558 559 560 561 562 563 564 565 566 567 568 569 570 571 572 573 574 575 576 577 578 579 580

G A S D I R D Q S R N W L P G P C Y R Q Q R V S K T S A D N N N S E Y S W T G A T K Y H L N G R D S L V N P G P A M A S H K D D E E K F F P Q S G V L I F G K Q G S E K T N V D I E K V M I T D E E E I R T T N P V A T E Q Y G S V S T

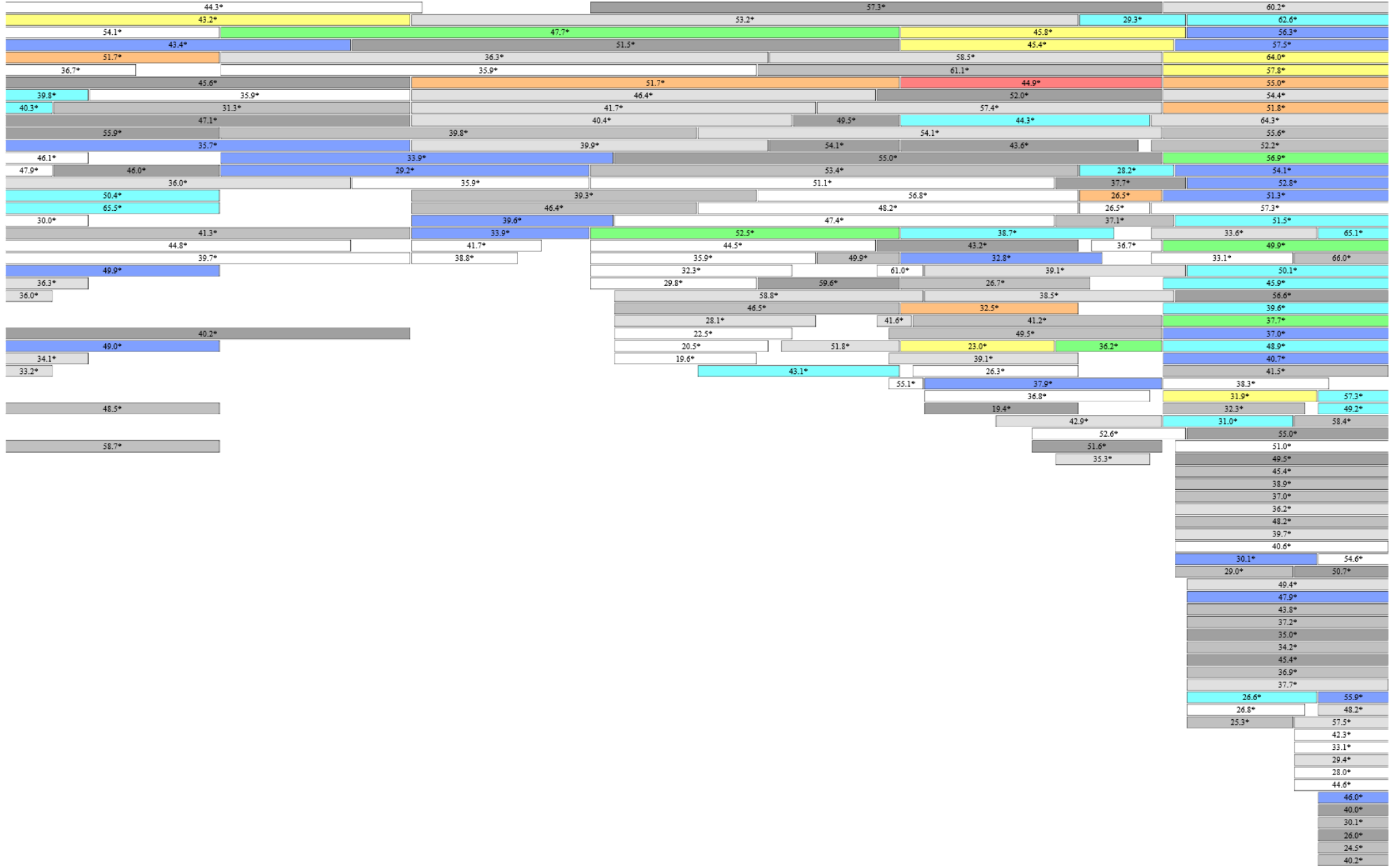


Figure S1 Continued:

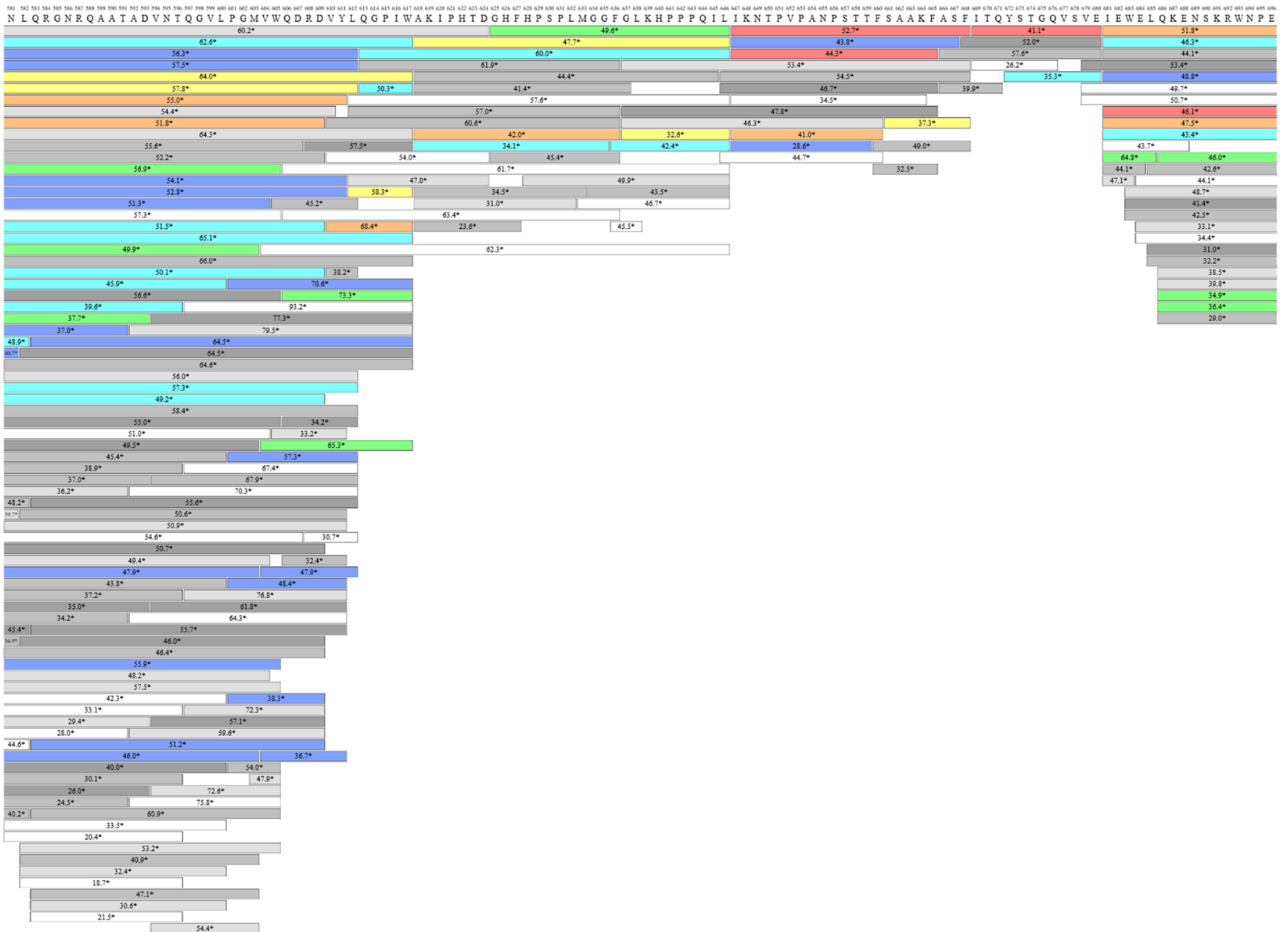


Figure S1 Continued:

697 698 699 700 701 702 703 704 705 706 707 708 709 710 711 712 713 714 715 716 717 718 719 720 721 722 723 724 725 726 727 728 729 730 731 732 733 734

I Q Y T S N Y N K S V N V D F T V D T N G V Y S E P R P I G T R Y L T R N L

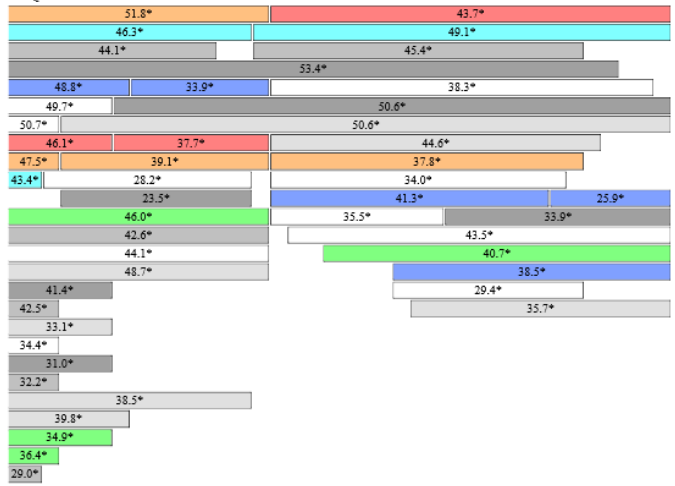
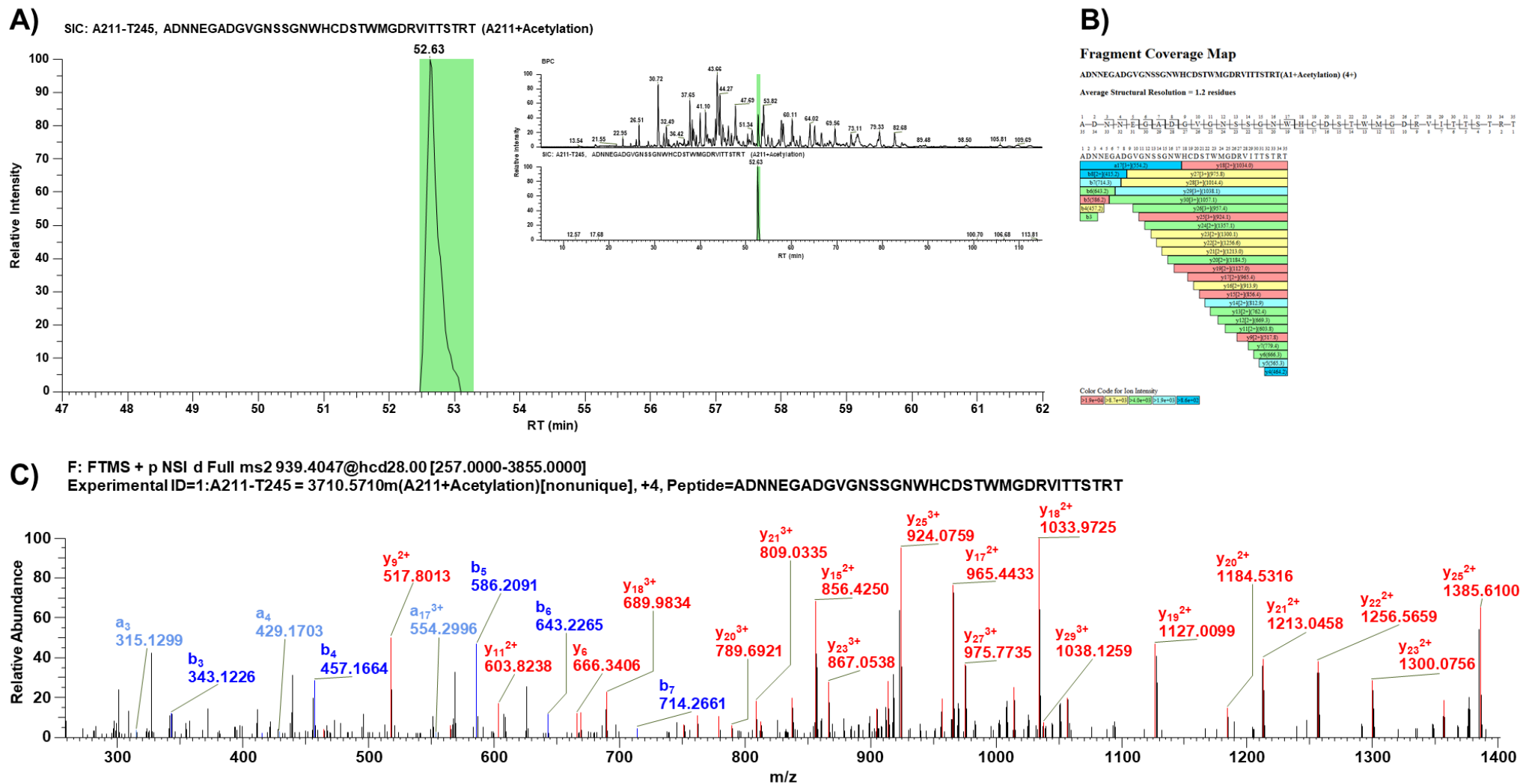
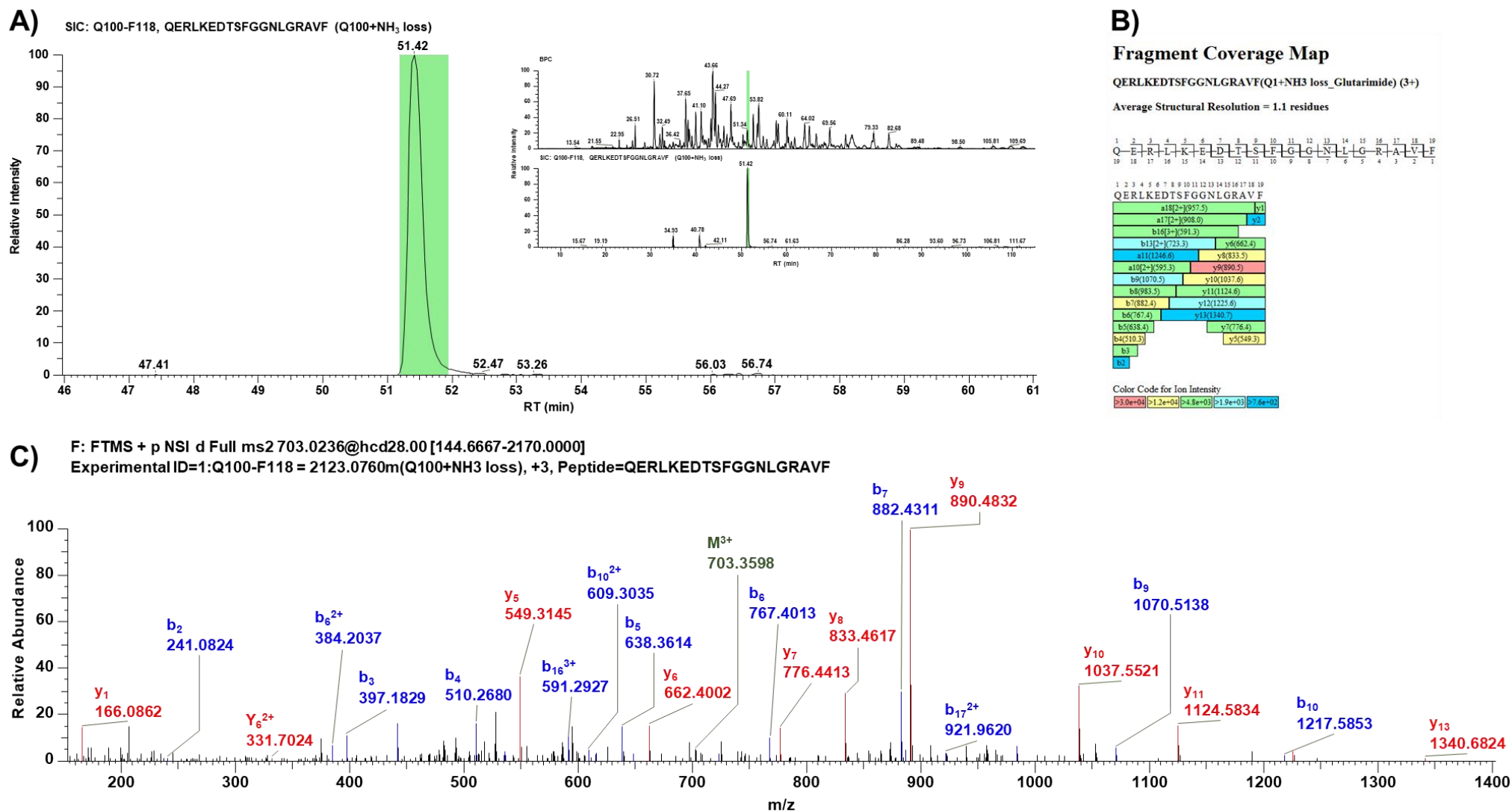


Figure S1 Continued:

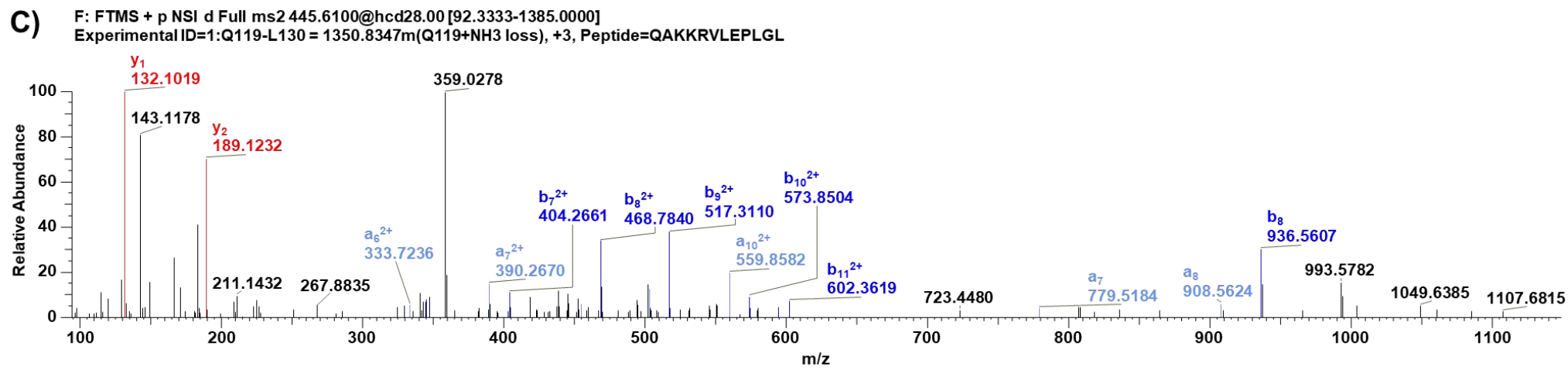
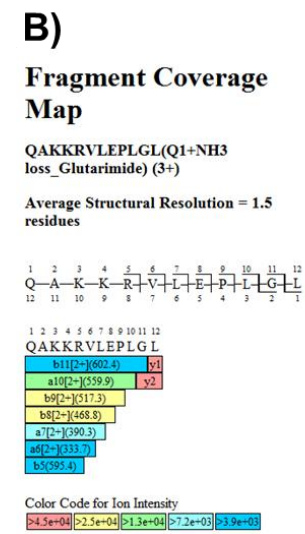
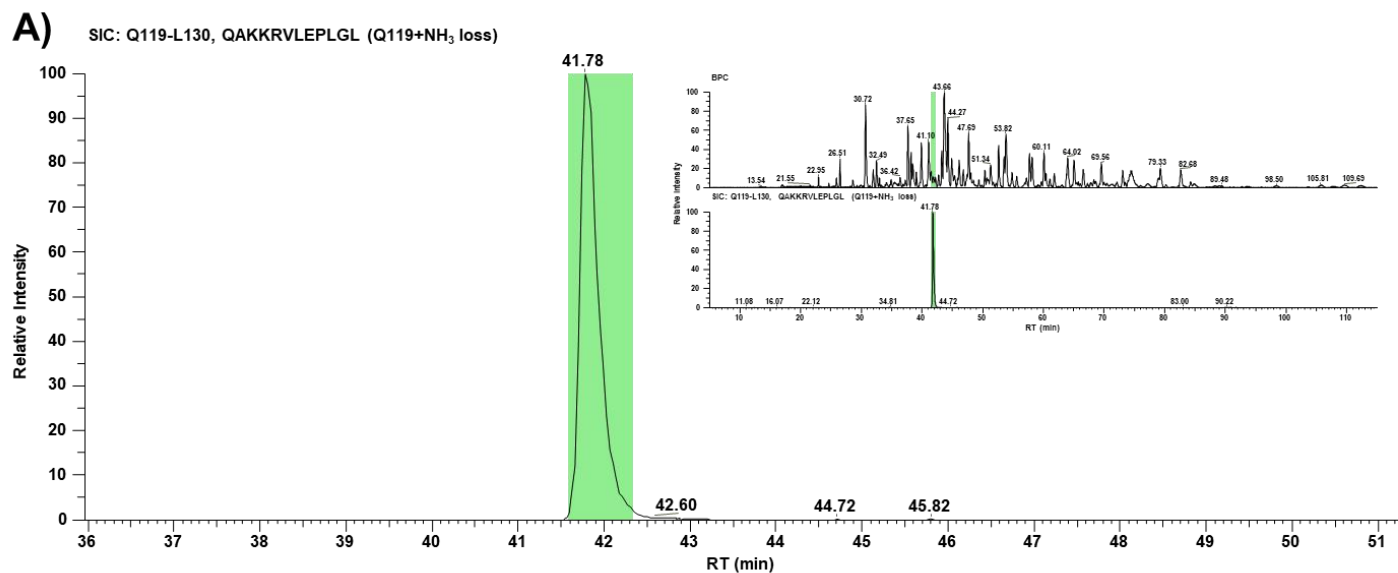




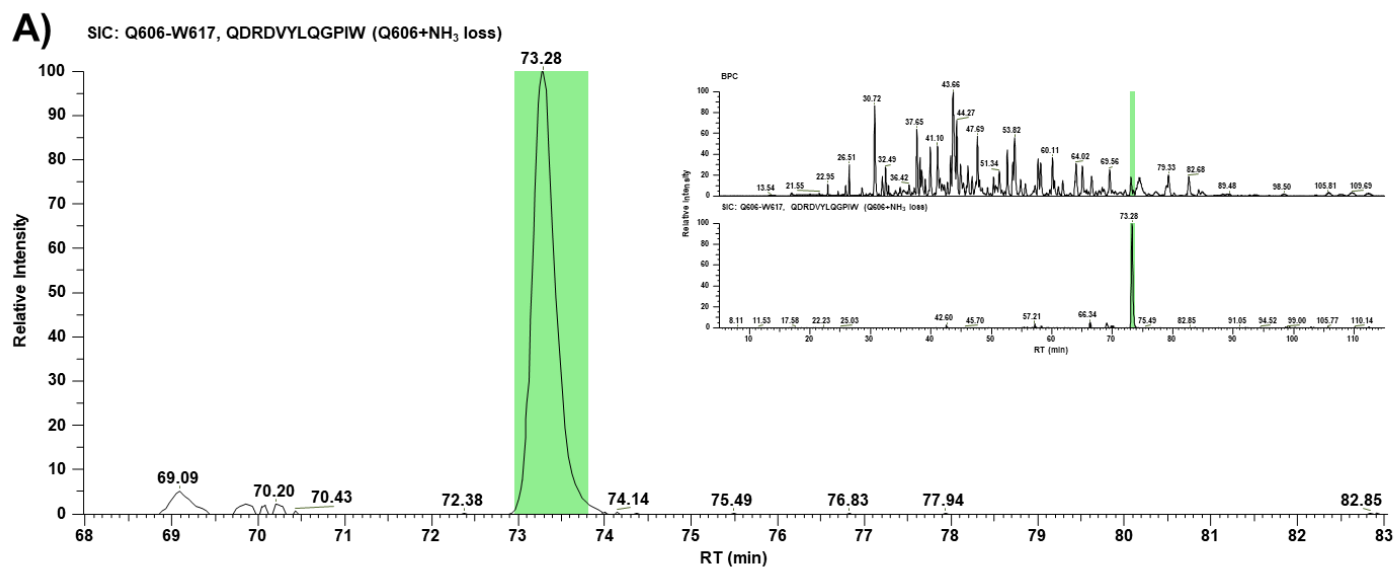
**Figure S2: Peptide *ADNNEGADGVGNSSGNWHCDSTWMGDRVITTSTRT* with acetylation at A211.** Peptide and PTM are identified using BioPharma Finder (BPF) software. Peptide is representative of the peptides detected in BPF that indicate the presence of the VP3 variant A211(Ac) – L735. A) Selected ion chromatogram (SIC) of acetylated peptide (insert shows SIC in relation to base peak chromatogram (BPC)). B) Peptide fragment coverage map. C) MS/MS spectrum with diagnostic fragment ion assignments.



**Figure S3: Peptide *QERLKEDTSFGGNLGRAVF* with NH<sub>3</sub> loss at Q100.** Peptide and PTM identified using BioPharma Finder software. A) Selected ion chromatogram (SIC) of peptide containing the NH<sub>3</sub> loss PTM (insert shows SIC in relation to base peak chromatogram (BPC)). B) Peptide fragment coverage map. C) MS/MS spectrum with diagnostic fragment ion assignments.



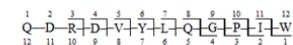
**Figure S4: Peptide QAKKRVLEPLGL with NH<sub>3</sub> loss at Q119.** Peptide and PTM identified using BioPharma Finder software. A) Selected ion chromatogram (SIC) of peptide containing the NH<sub>3</sub> loss PTM (insert shows SIC in relation to base peak chromatogram (BPC)). B) Peptide fragment coverage map. C) MS/MS spectrum with diagnostic fragment ion assignments.



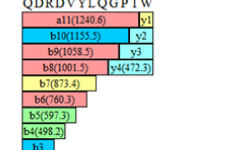
**B)**  
Fragment Coverage Map

QDRDVYLQGIW(Q1+NH<sub>3</sub> loss\_Glutarimide) (2+)

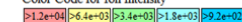
Average Structural Resolution = 1.2 residues



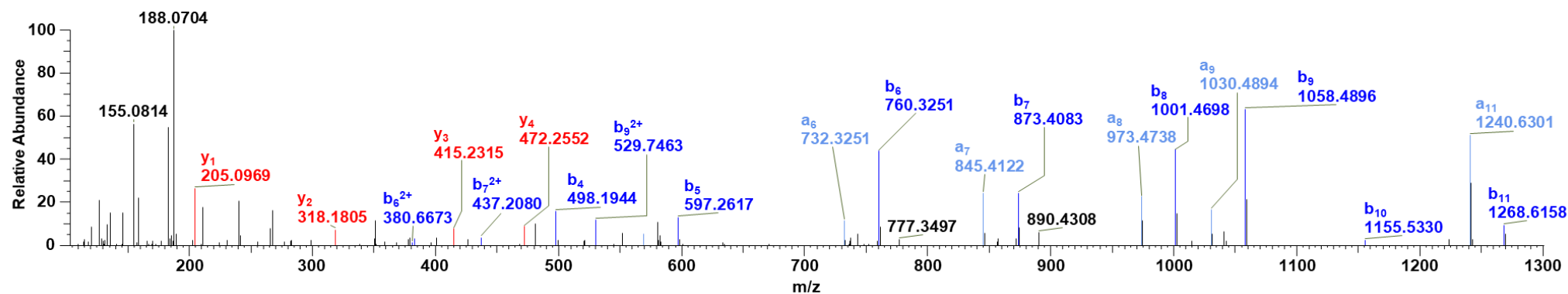
1 2 3 4 5 6 7 8 9 10 11 12  
QDRDVYLQGIW



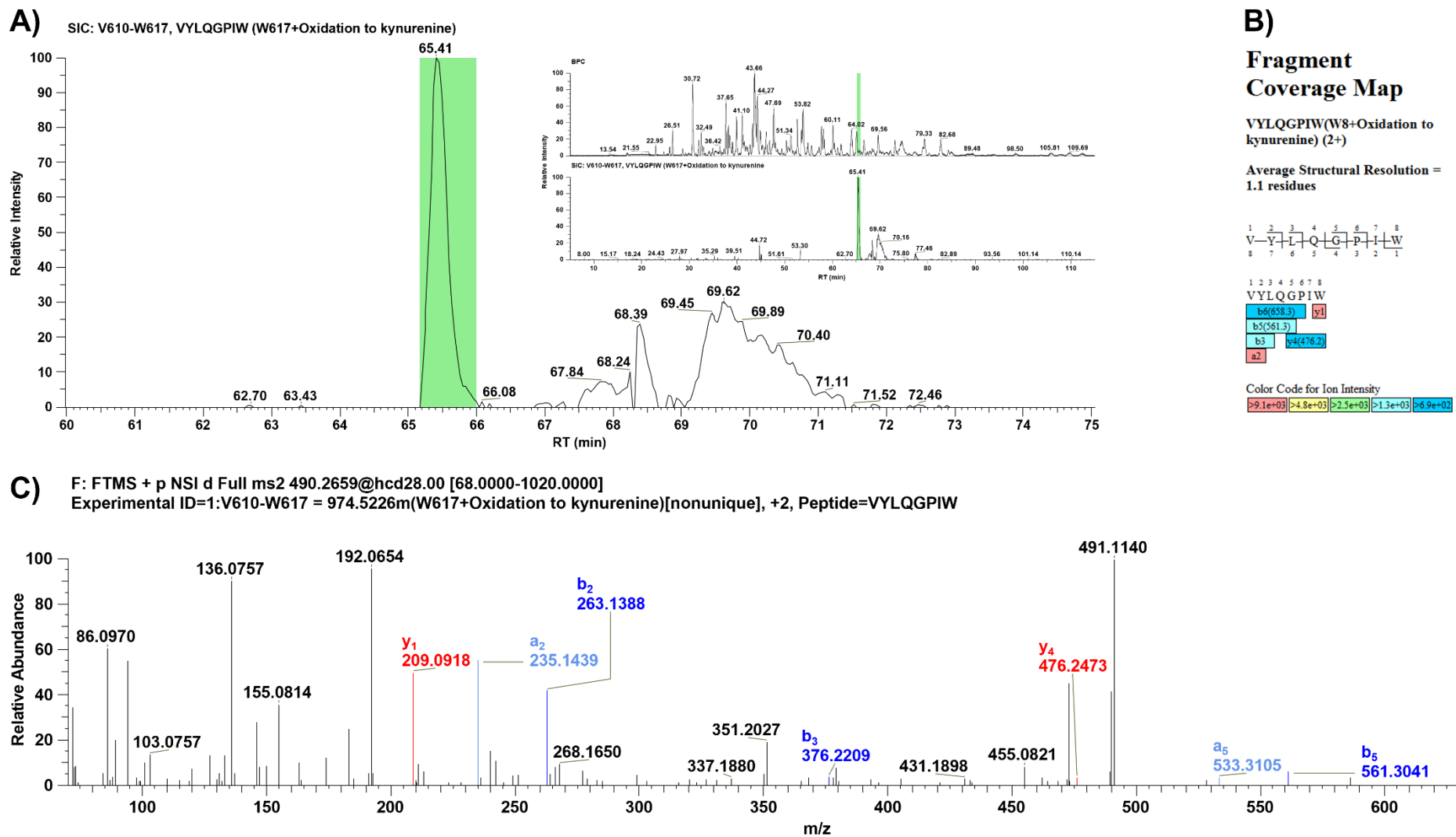
Color Code for Ion Intensity



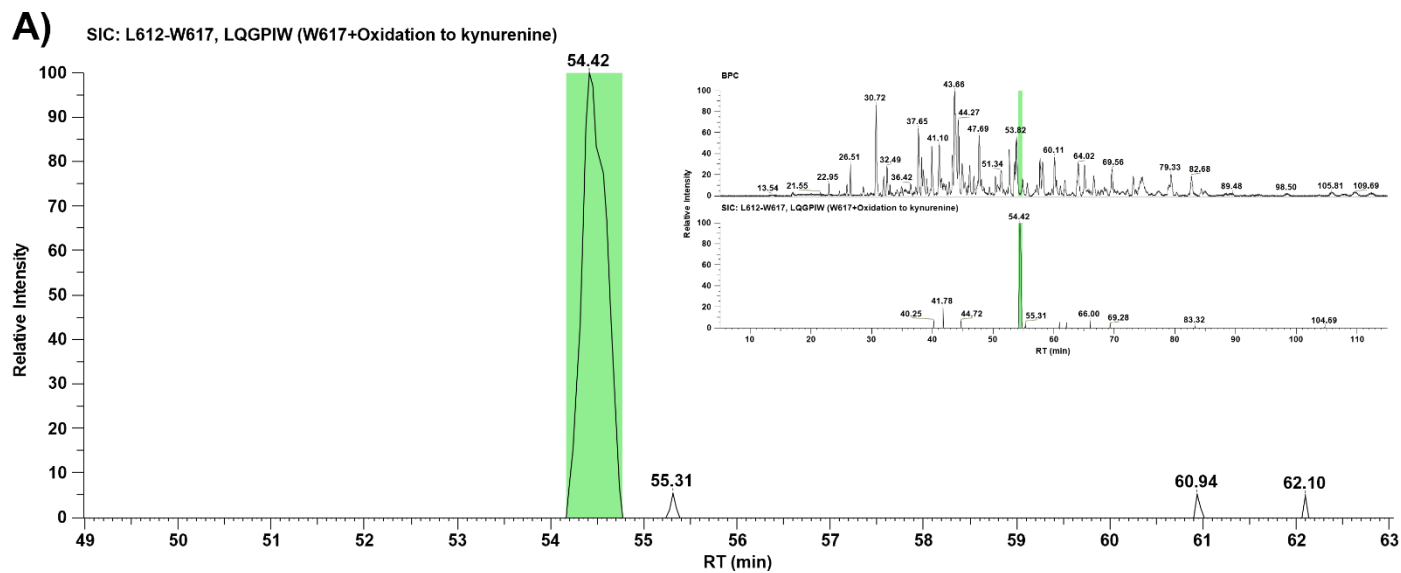
**C)** F: FTMS + p NSI d Full ms2 736.8621@hcd28.00 [101.6667-1525.0000]  
Experimental ID=1:Q606-W617 = 1488.7361m(Q606+NH<sub>3</sub> loss)[nonunique], +2, Peptide=QDRDVYLQGIW



**Figure S5: Peptide QDRDVYLQGIW with NH<sub>3</sub> loss at Q606.** Peptide and PTM identified using BioPharma Finder software. A) Selected ion chromatogram (SIC) of peptide containing the NH<sub>3</sub> loss PTM (insert shows SIC in relation to base peak chromatogram (BPC)). B) Peptide fragment coverage map. C) MS/MS spectrum with diagnostic fragment ion assignments.



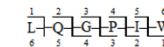
**Figure S6: Peptide VYLQGIW with oxidation to kynurenine at W617.** Peptide and PTM identified using BioPharma Finder Software. A) Selected ion chromatogram (SIC) of peptide containing the oxidation to kynurenine PTM (insert shows SIC in relation to base peak chromatogram (BPC)). B) Peptide fragment coverage map. C) MS/MS spectrum with diagnostic fragment ion assignments.



**B)**  
**Fragment Coverage Map**

LQGPIW(W6+Oxidation to kynurenine) (1+)

Average Structural Resolution = 1.0 residues

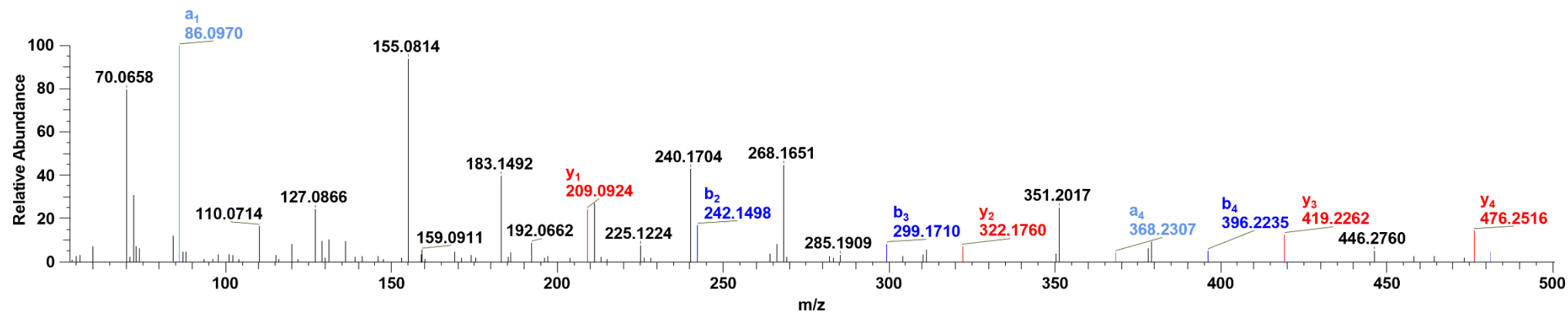


|           |           |   |   |   |   |
|-----------|-----------|---|---|---|---|
| 1         | 2         | 3 | 4 | 5 | 6 |
| L         | Q         | G | P | I | W |
| a5(481.3) | y1        |   |   |   |   |
| b4(396.2) | y2        |   |   |   |   |
| b3        | y3        |   |   |   |   |
| b2        | y4(476.3) |   |   |   |   |
| a1        |           |   |   |   |   |

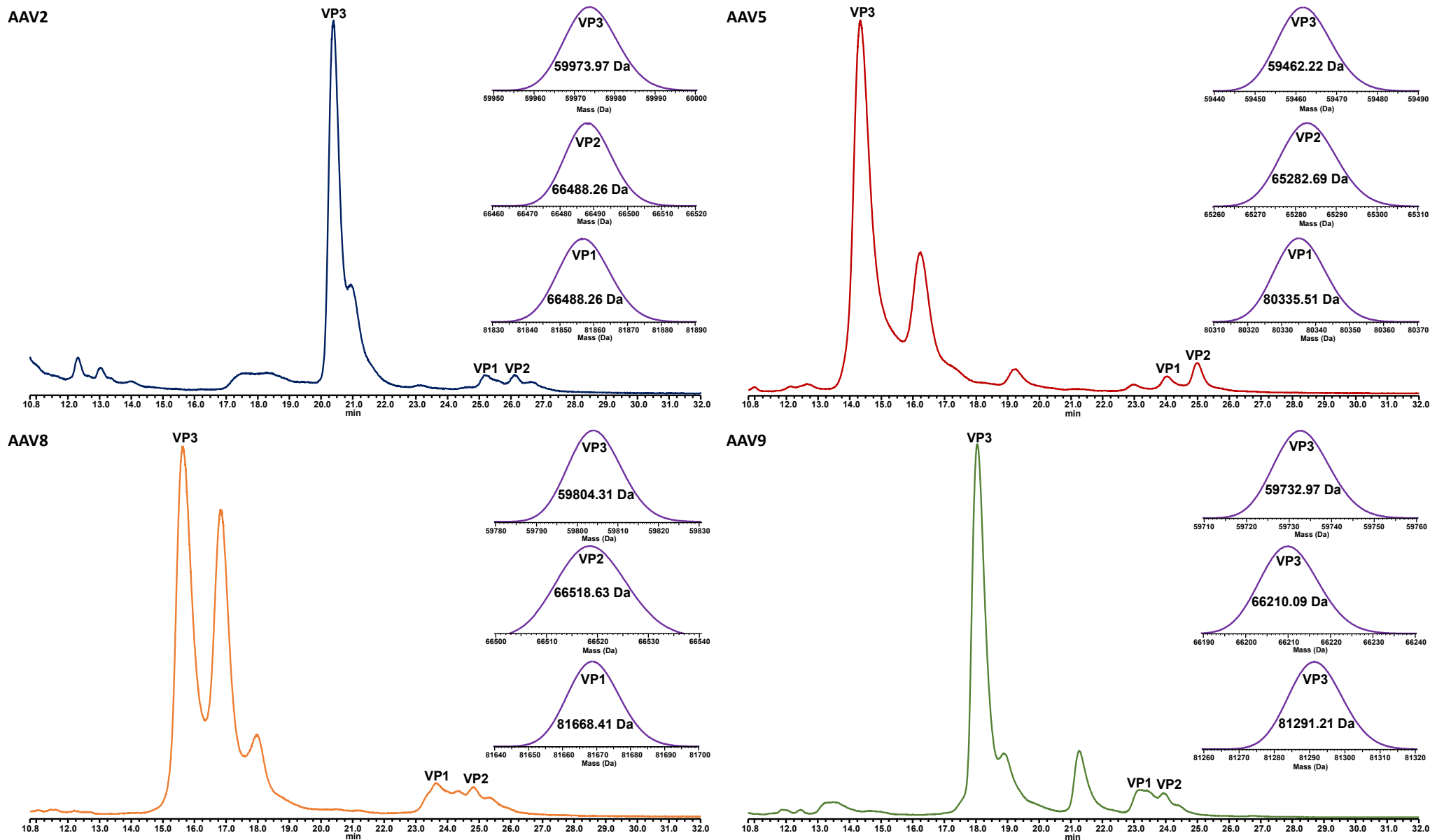
Color Code for Ion Intensity

|          |          |          |          |          |
|----------|----------|----------|----------|----------|
| >1.6e+04 | >8.9e+03 | >4.9e+03 | >2.7e+03 | >1.5e+03 |
|----------|----------|----------|----------|----------|

**C)** F: FTMS + p NSI d Full ms2 717.3935@hcd28.00 [50.0000-750.0000]  
Experimental ID=1:L612-W617 = 712.3908m(W617+Oxidation to kynurenine)[nonunique], +1, Peptide=LQGPIW



**Figure S7: Peptide LQGPIW with oxidation to kynurenine at W617.** Peptide and PTM identified using BioPharma Finder Software. A) Selected ion chromatogram (SIC) of peptide containing the oxidation to kynurenine PTM (insert shows SIC in relation to base peak chromatogram (BPC)). B) Peptide fragment coverage map. C) MS/MS spectrum with diagnostic fragment ion assignments.



**Figure S8: FLR traces of VP separations performed using baculovirus/Sf9 derived AAV2, AAV5, AAV8, and AAV9 serotypes.** Serotypes were analysed to help develop a universal processing method for serotype identification. The development of this processing method is a part of the creation of a data processing workflow capable of rapid serotype identification and PTM characterization. The peaks representing the unmodified VP proteoforms are labelled as those masses (along with their respective retention times) were used to differentiate the serotypes. Deconvoluted spectra of each VP for their respective serotypes are shown on the right side of each FLR trace.

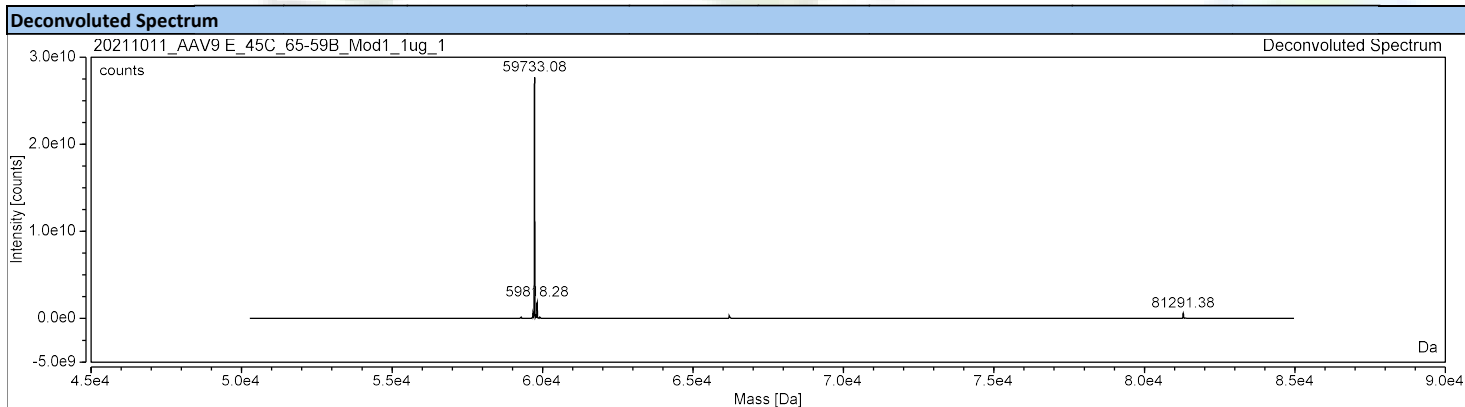
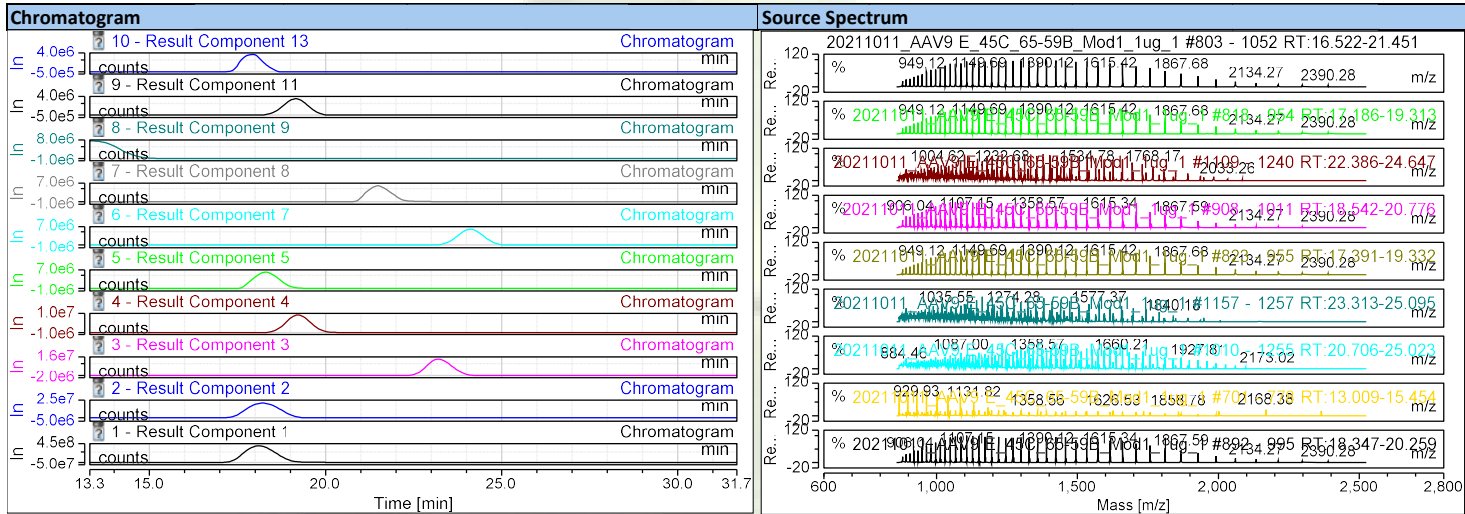
**Supplementary Report 1:**

Report of AAV rapid identity test generated in Chromeleon



## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV9 E_45C_65-59B_Mod1_1ug_1 | Injection Volume: 2.80                                       |
| Vial Number:      | R:A5                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 45.00                                 | Injection Date/Time: 12/Oct/21 14:28                         |



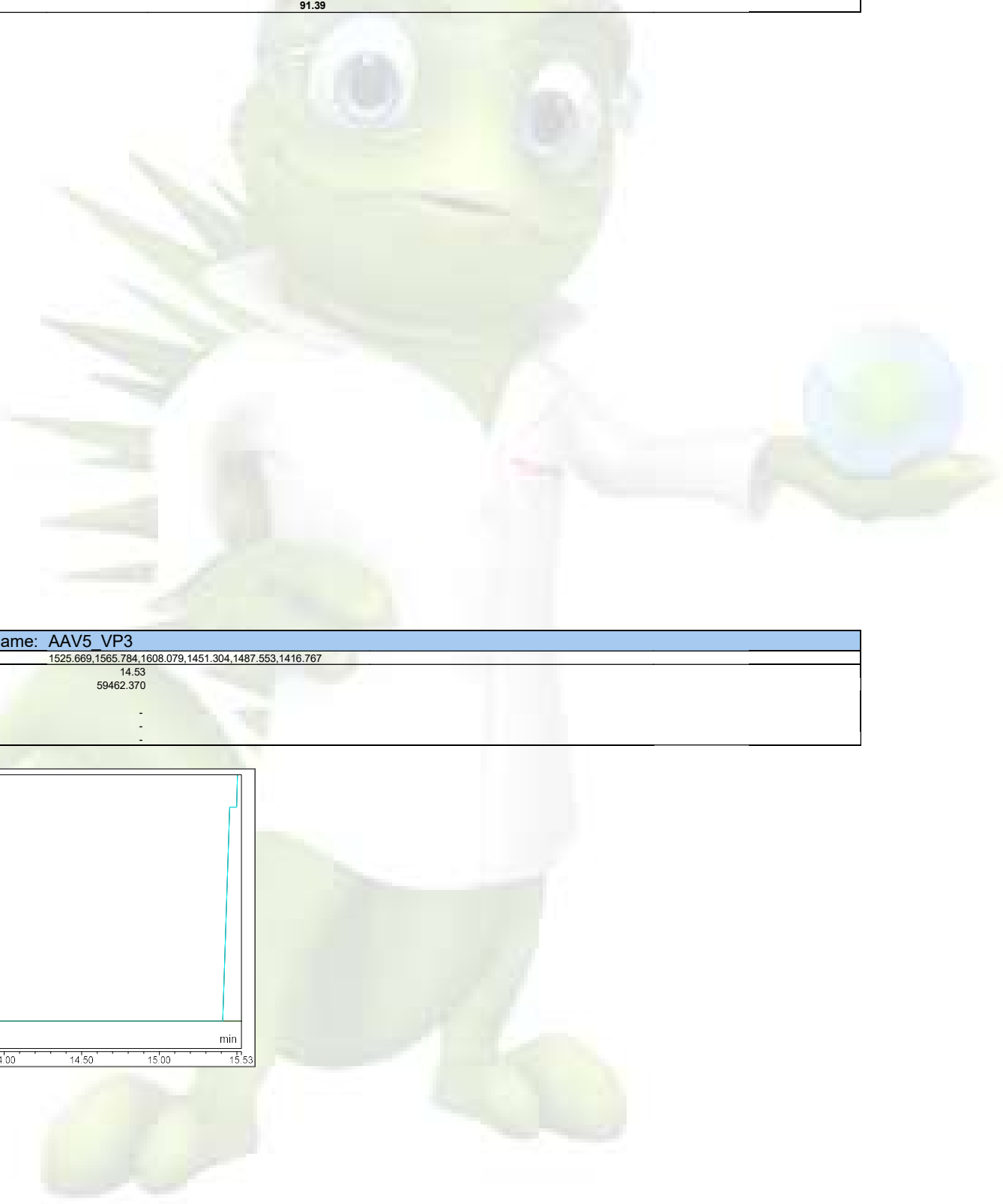
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

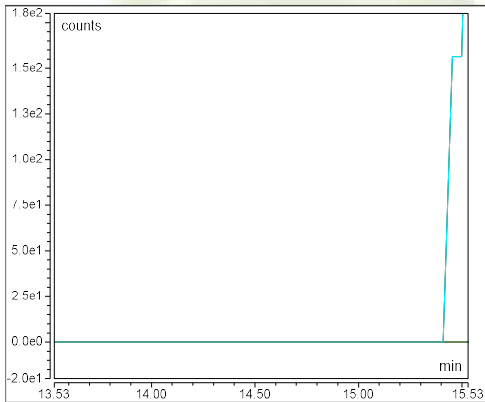
|  |   |  |
|--|---|--|
| <i>Injection Name:</i> 20211011_AAV9_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i> 2.80   |  |
| <i>Vial Number:</i> R:A5                                   | <i>Instrument Method:</i> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <i>Injection Type:</i> Unknown                             | <i>Processing Method:</i> MS Quantitative_45C_65-59B                            |  |
| <i>Run Time (min):</i> 45.00                               | <i>Injection Date/Time:</i> 12/Oct/21 14:28                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV9_VP3     | 18.129                | 5305032.777        | 88.65              | 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907 | 59732.620 | PASS                           |
| AAV9_VP1     | 23.207                | 106915.843         | 1.79               | 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636 | 81290.340 | PASS                           |
| AAV9_VP2     | 24.049                | 57302.637          | 0.96               | 1409.730,1162.589,1472.334,1274.267,1380.371,1352.231 | 66209.760 | PASS                           |
| <b>91.39</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

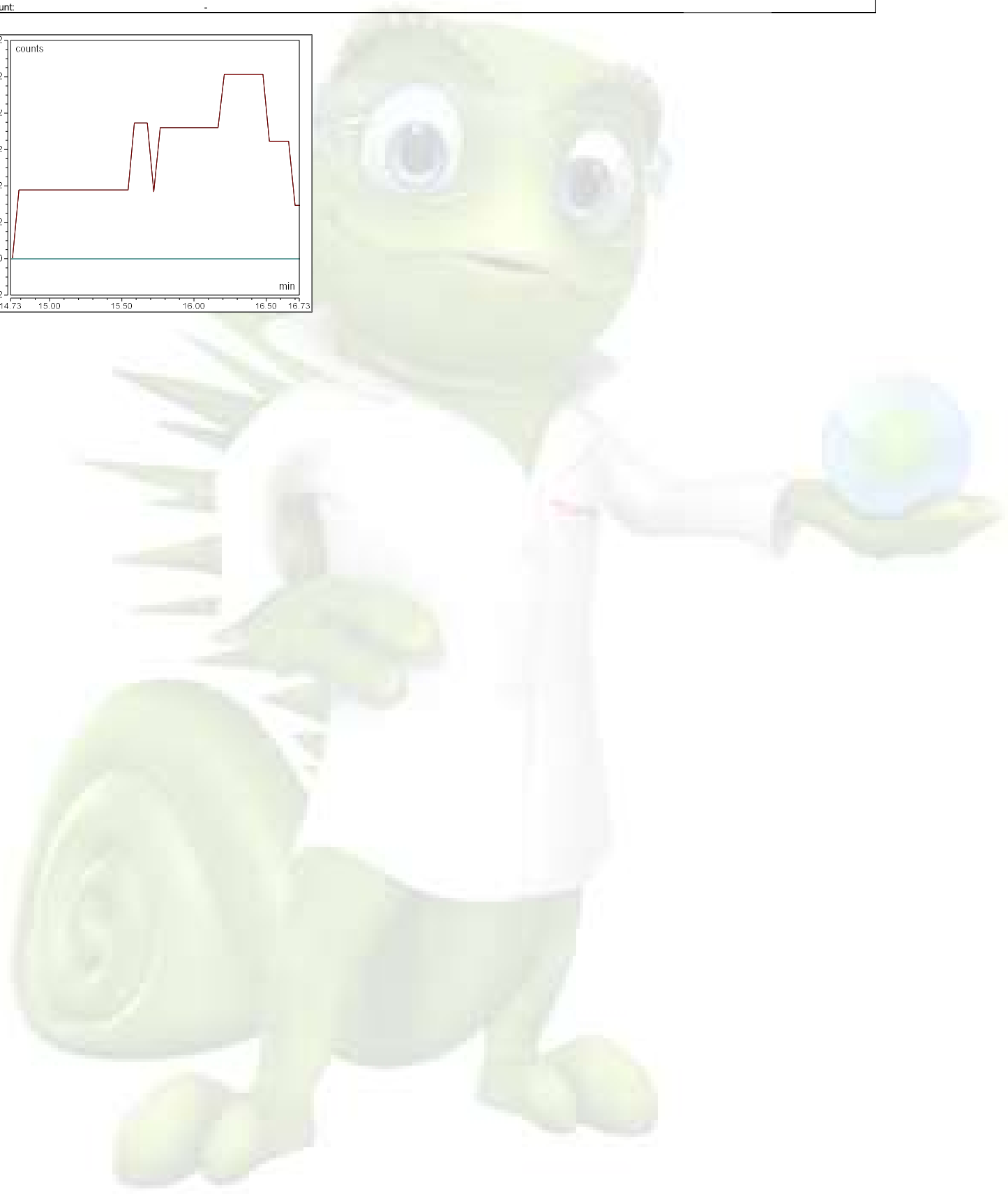
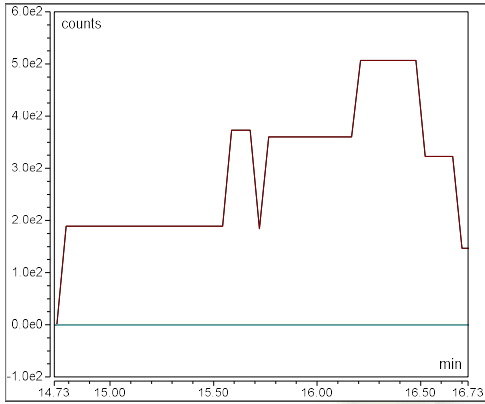
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

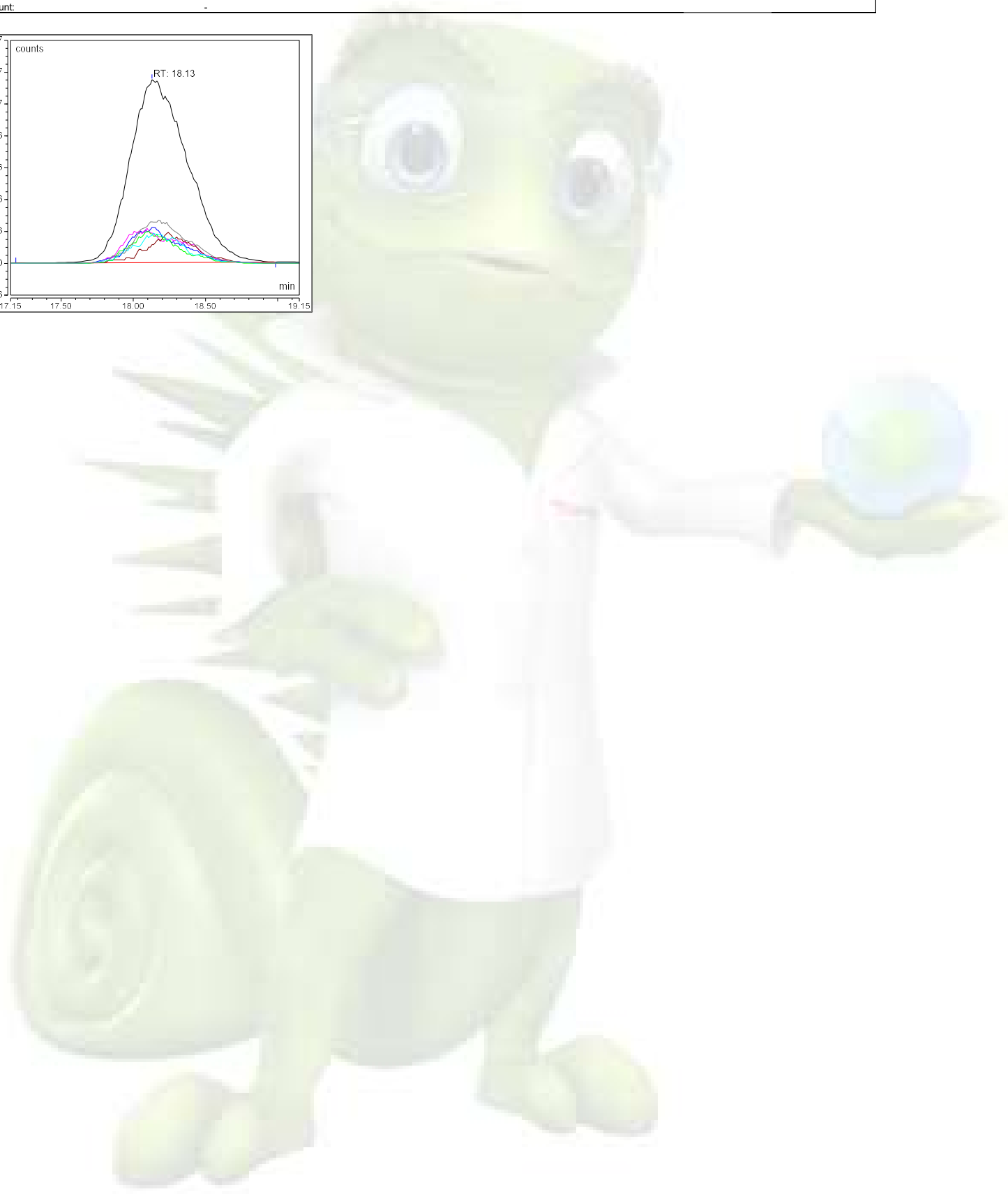
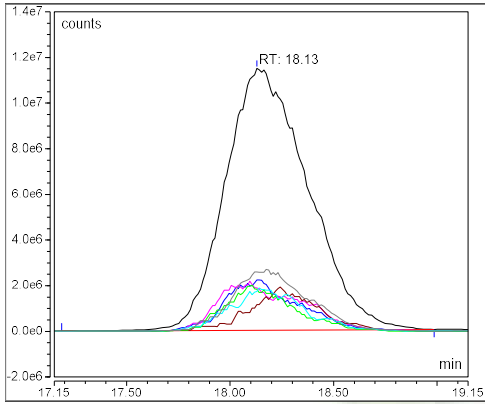
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): 5305033

Intensity (counts): 11472987

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

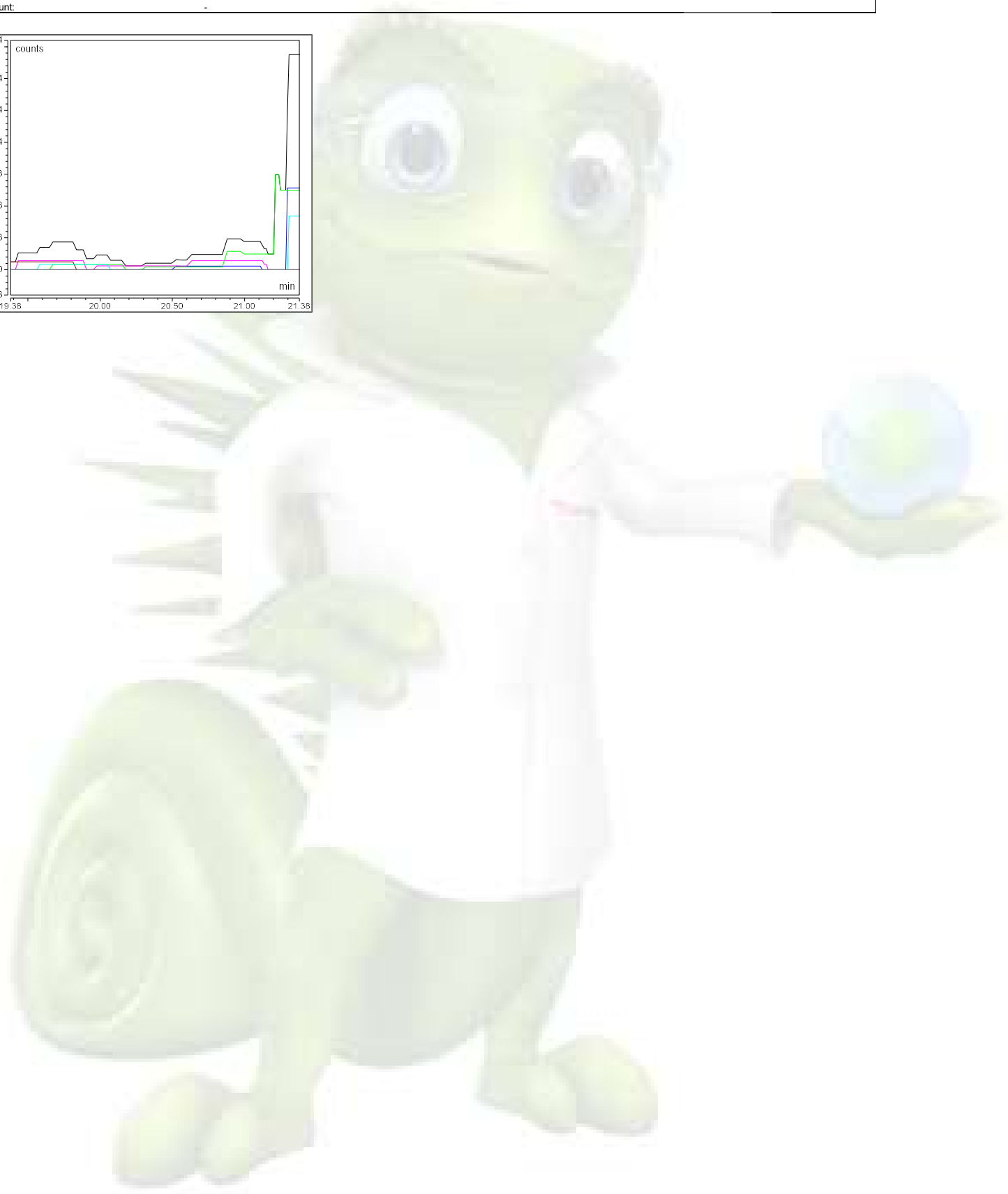
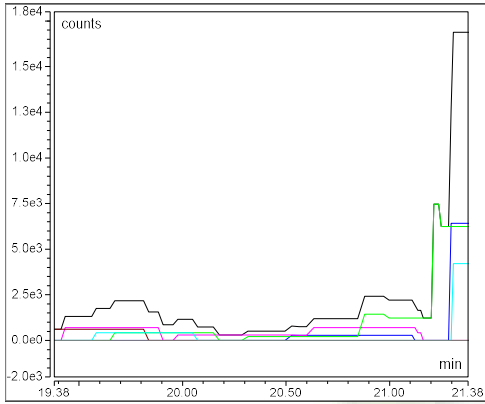
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

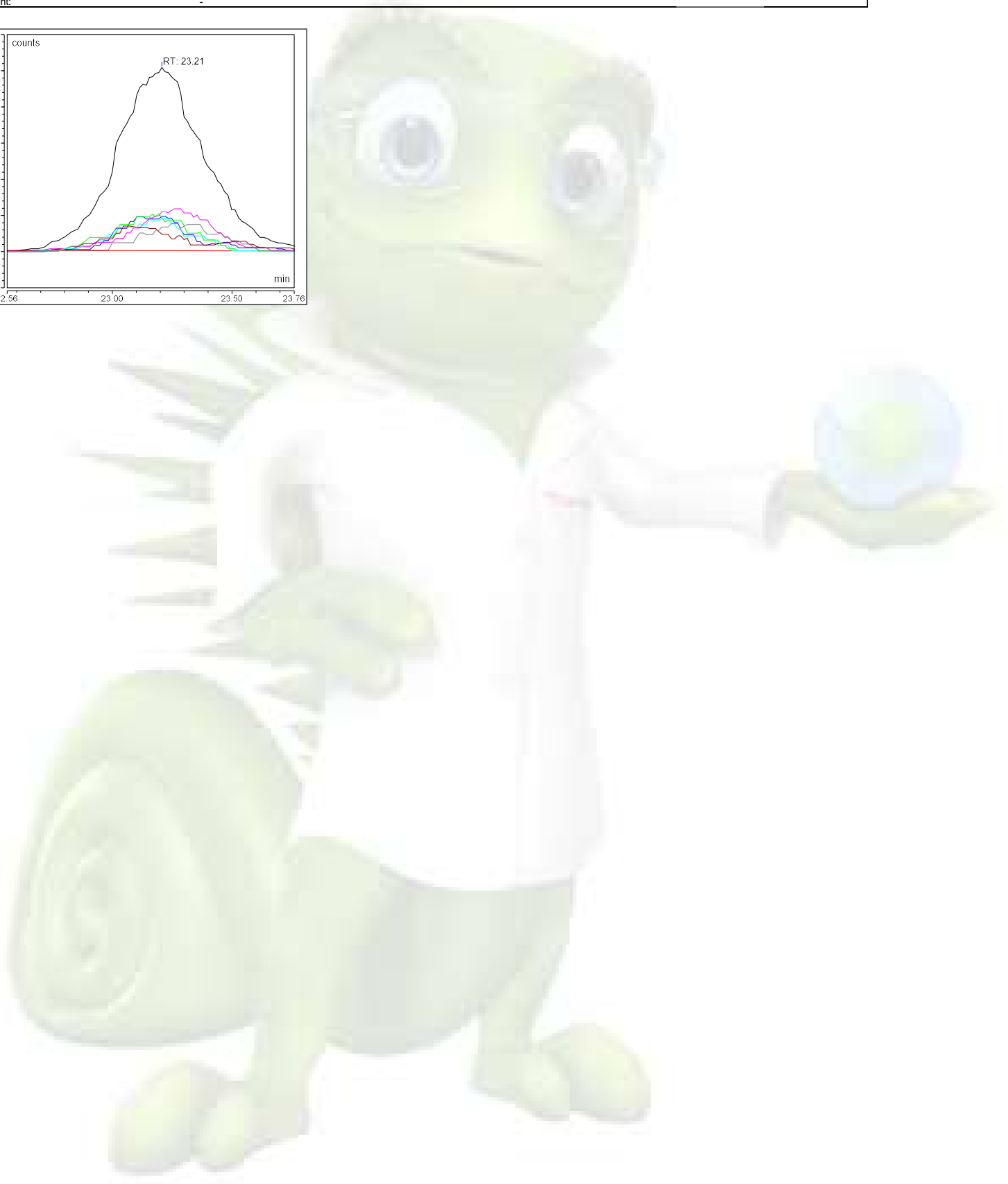
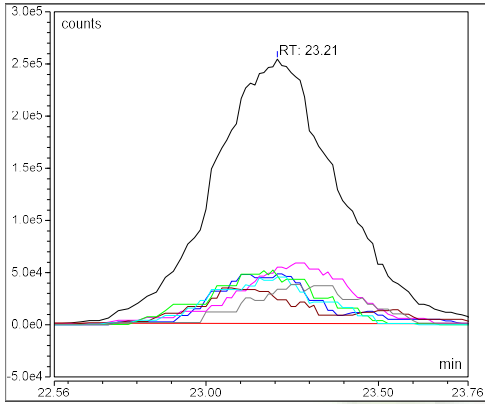
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): 106916

Intensity (counts): 253171

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

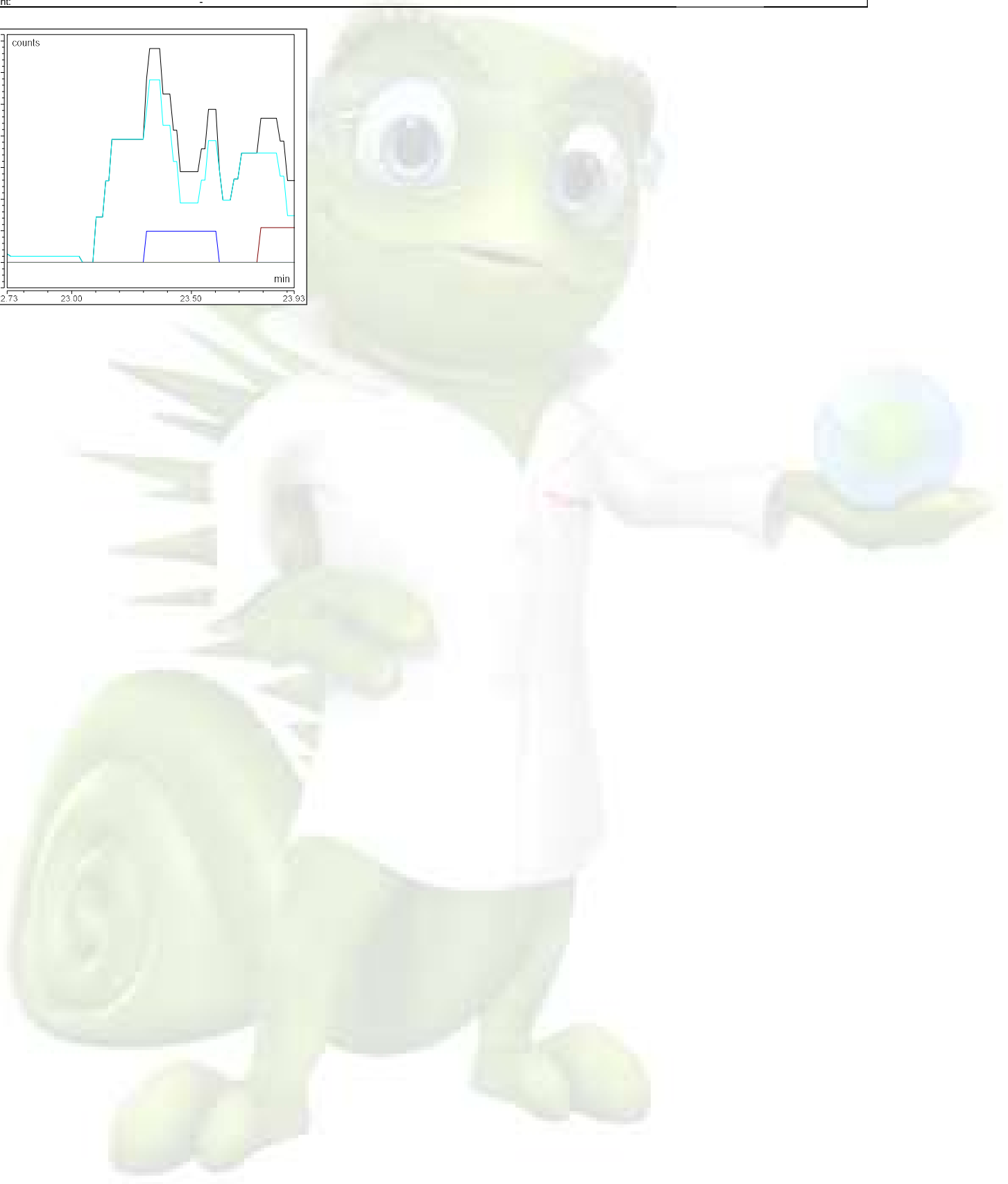
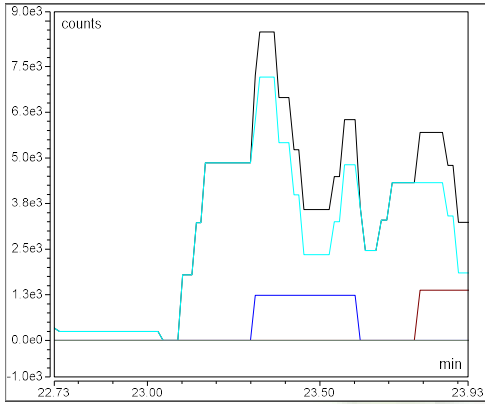
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

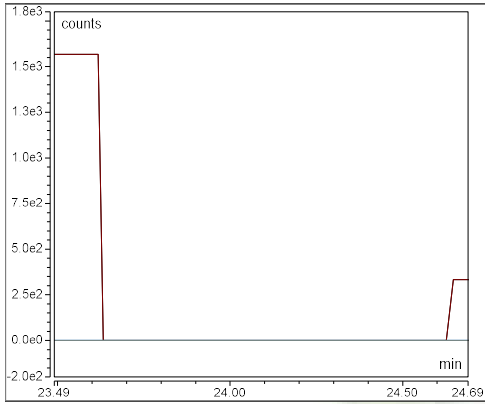
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

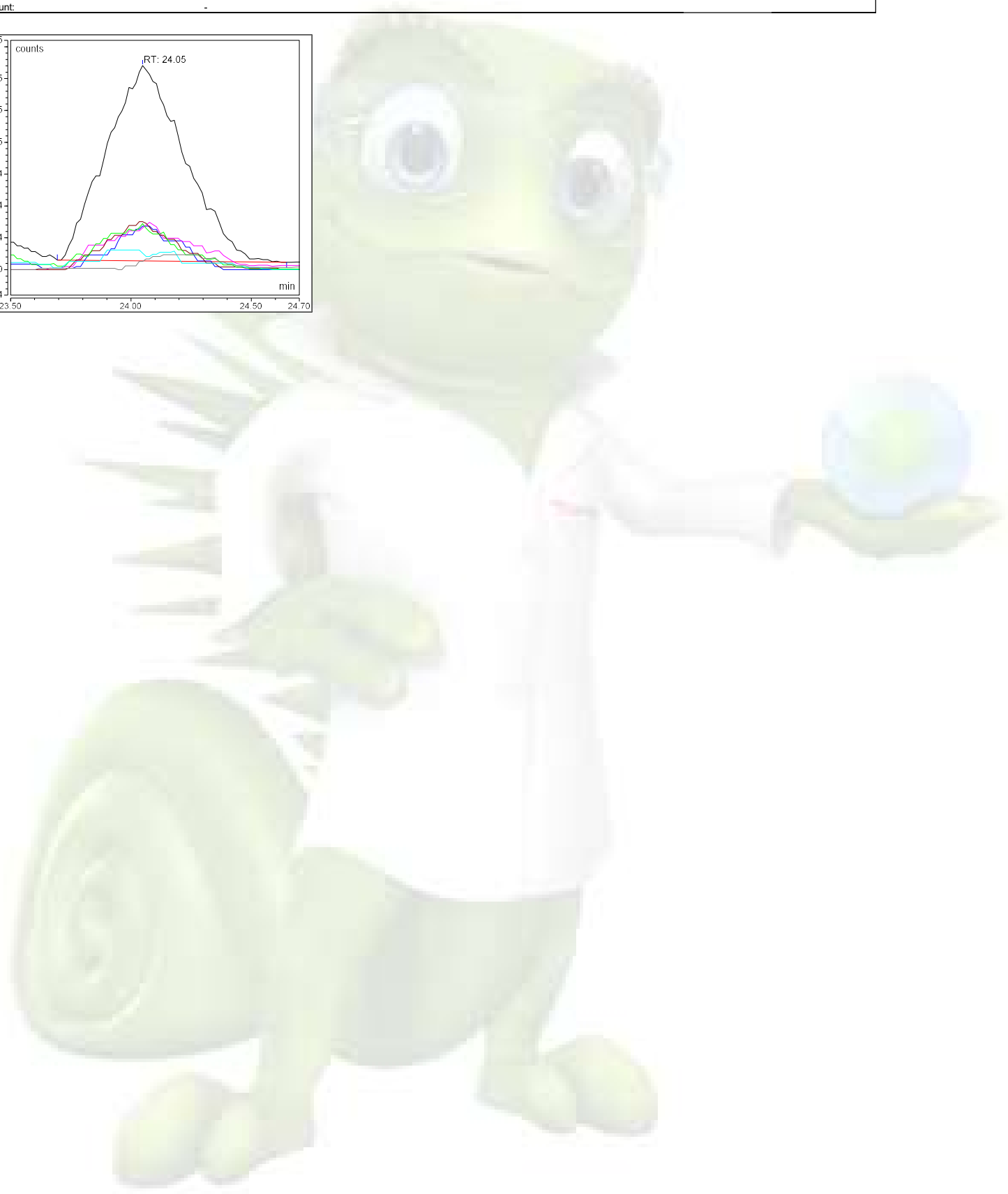
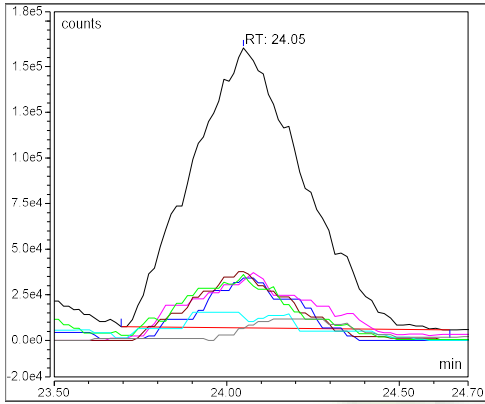
Amount: -





**Component Name: AAV9 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231 |
| Retention time (min): | 24.10   |
| Expected Mass:        | 66209.760   |
| Area (count*min):     | 57303   |
| Intensity (counts):   | 153294  |
| Amount:               | -   |



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

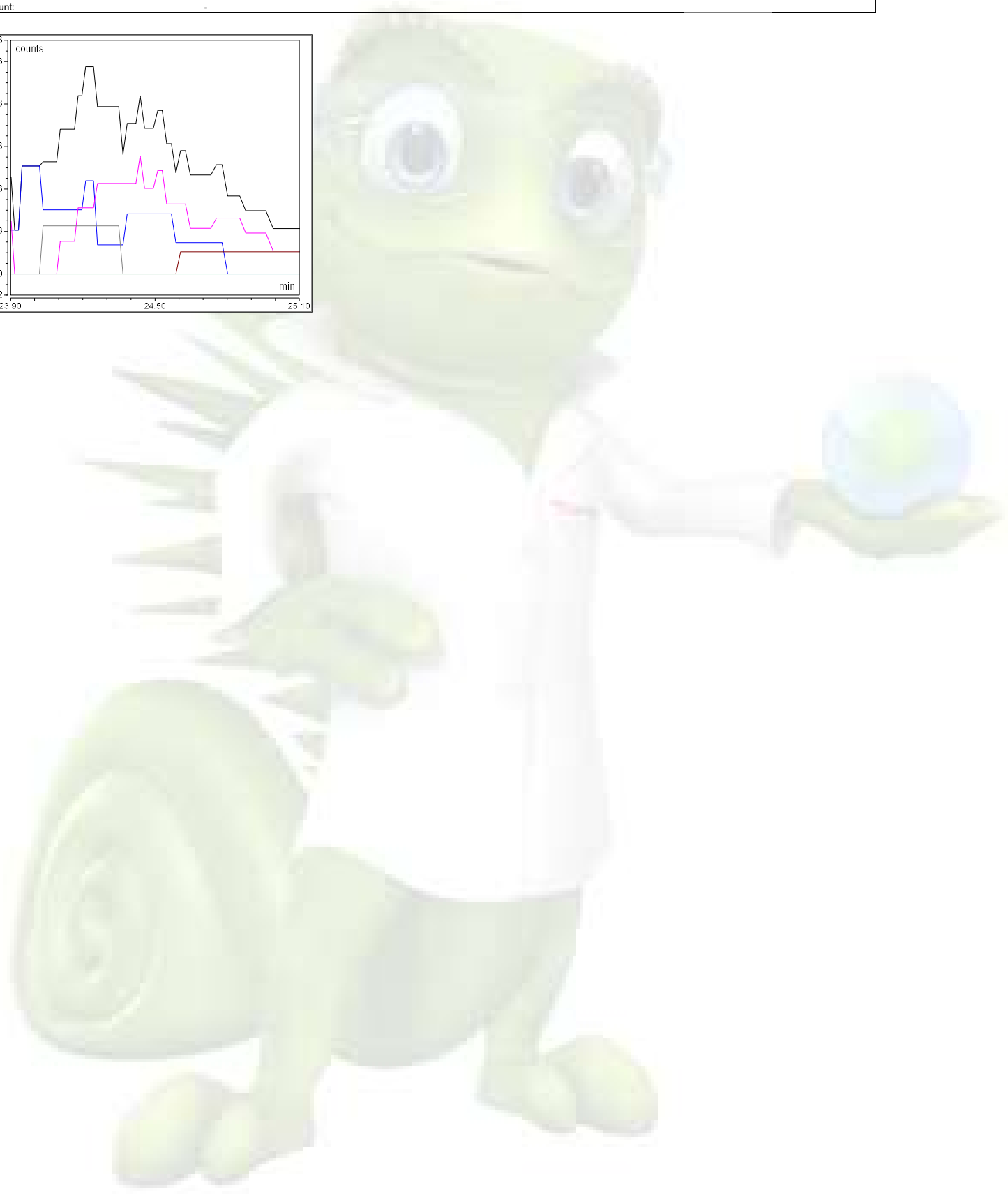
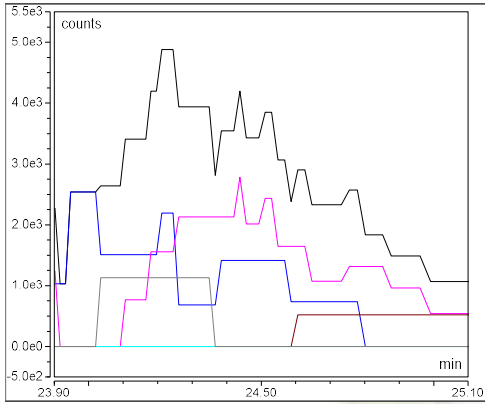
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

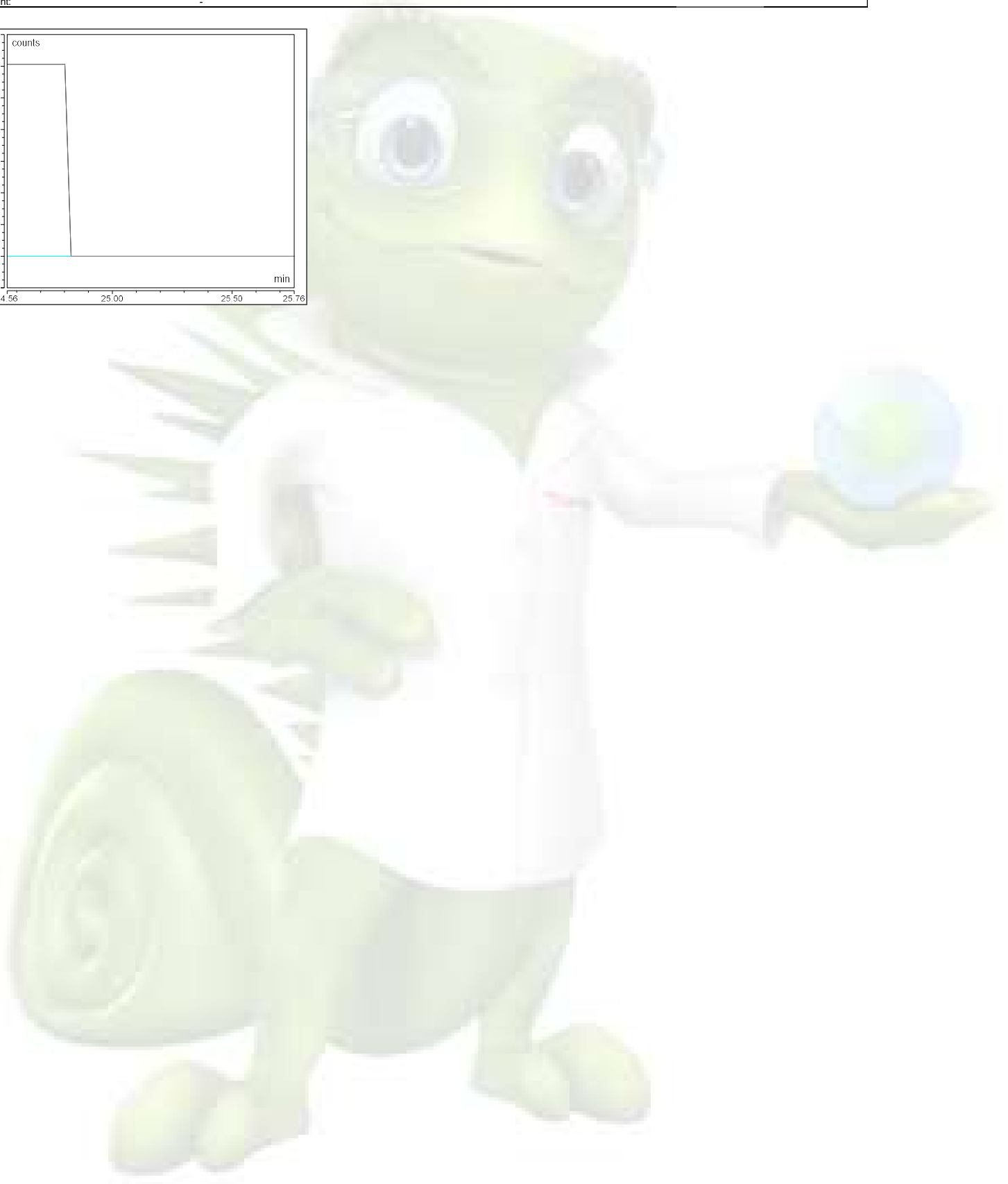
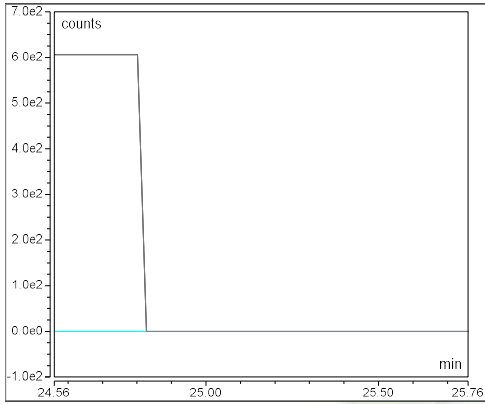
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

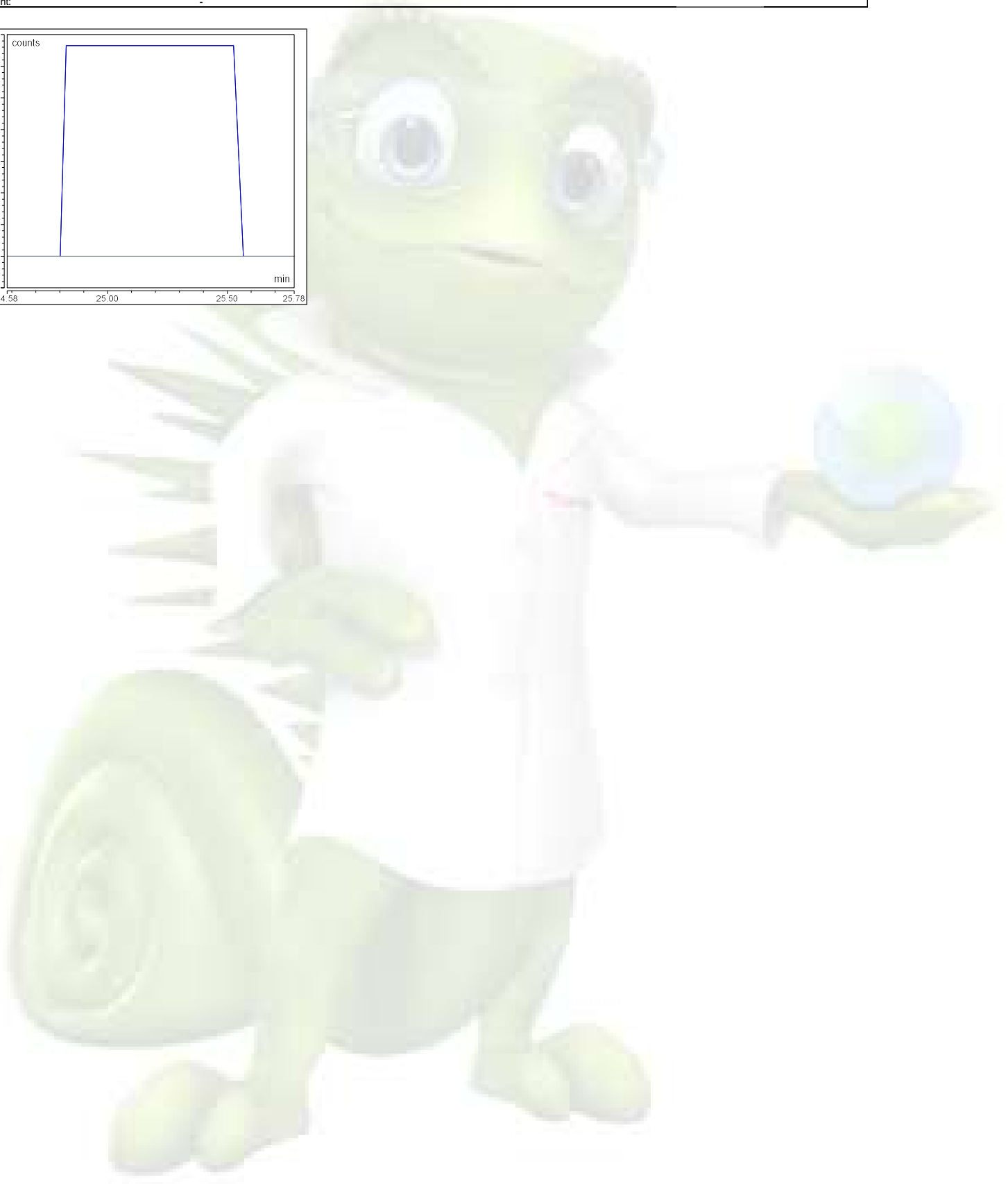
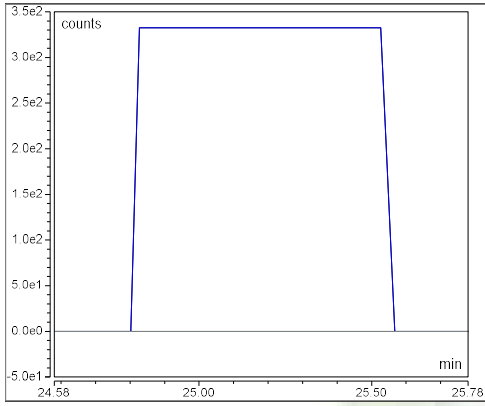
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

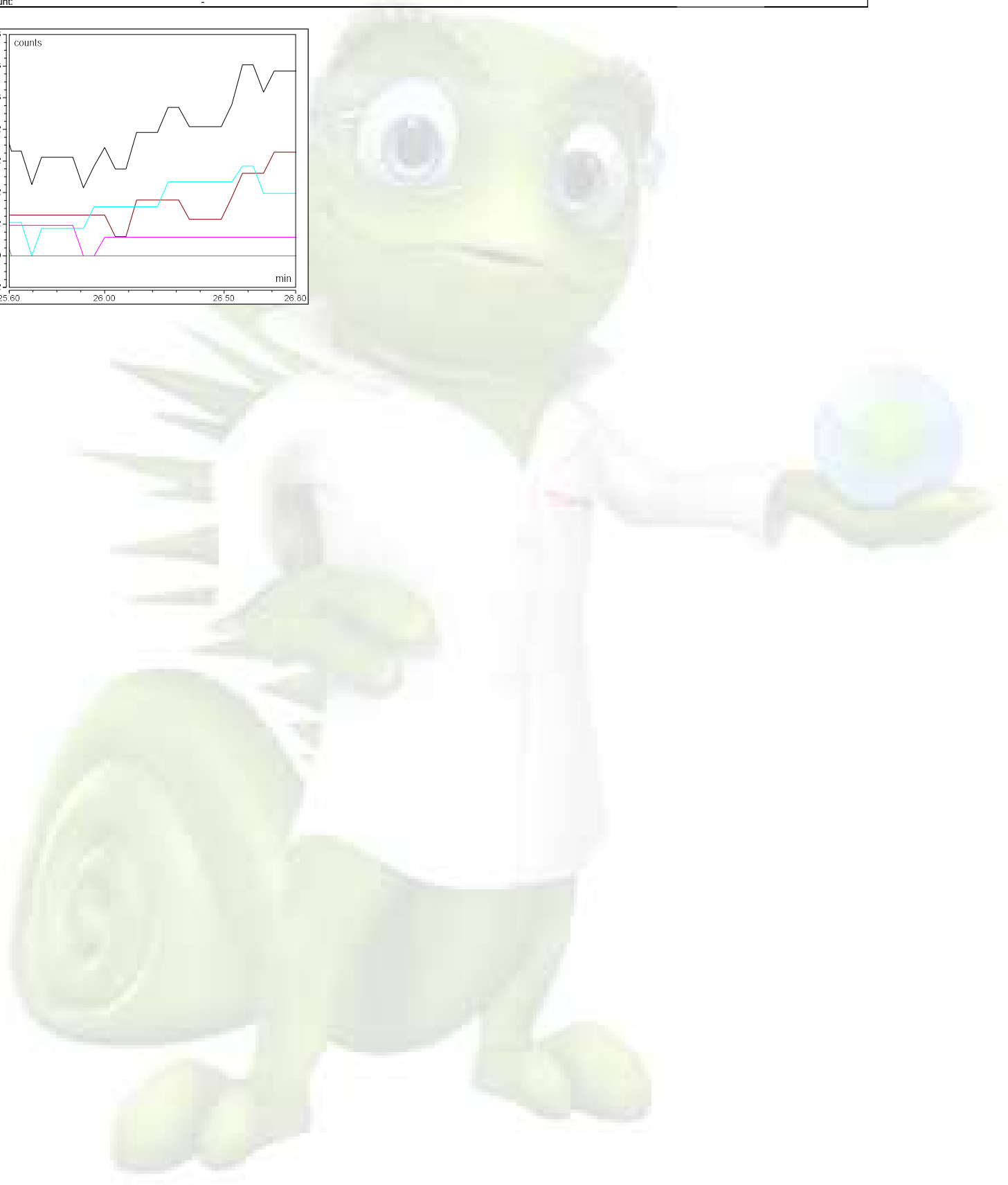
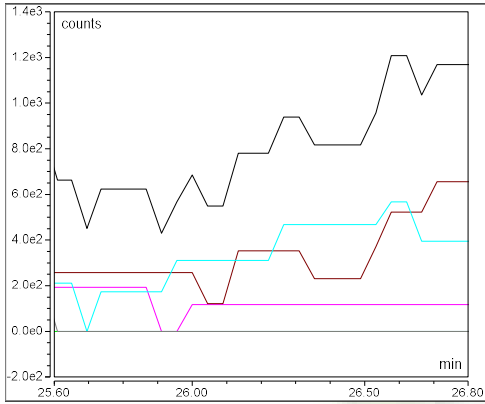
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

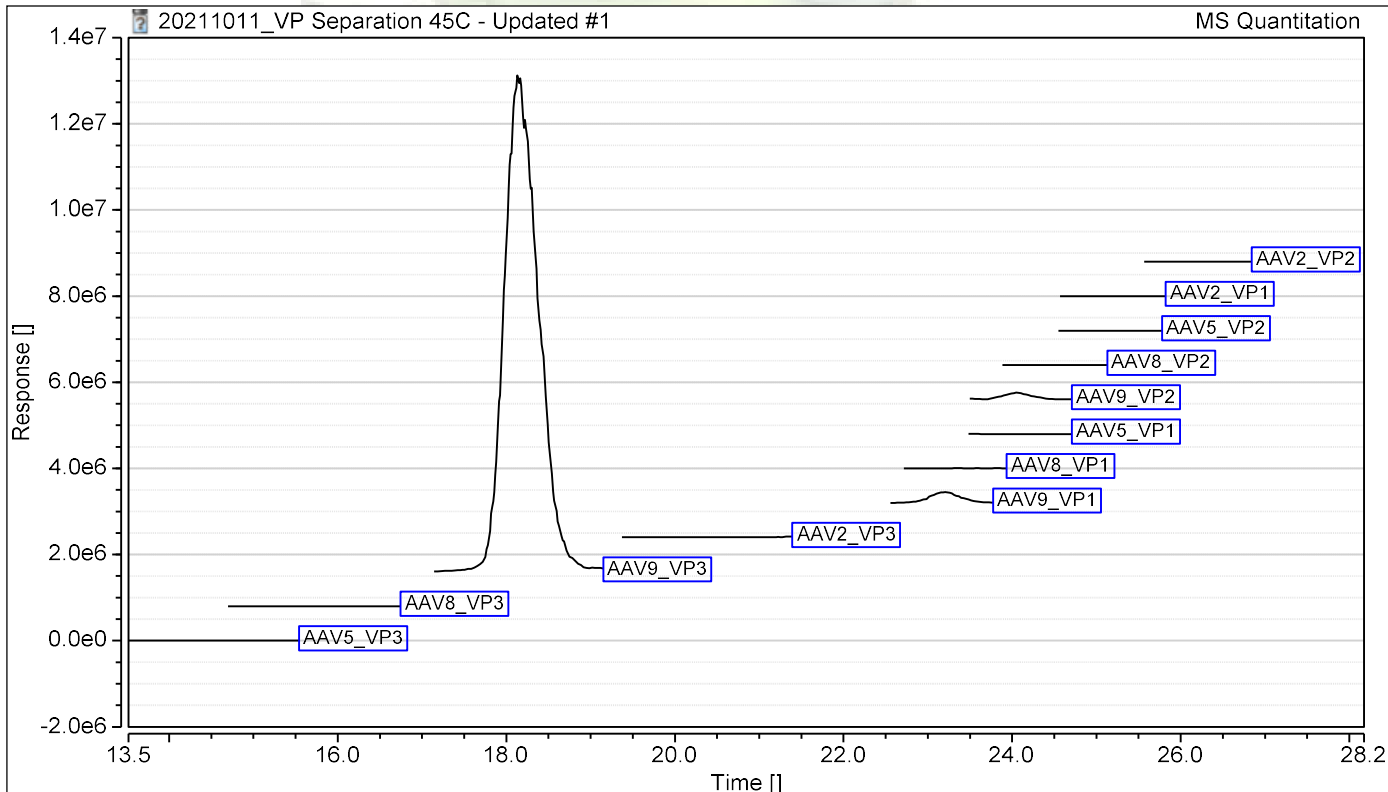
Amount: -



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV9 E_45C_65-59B_Mod1_1ug_1  | Run Time (min): 45.00   |
| Vial Number:         | R:A5   | Injection Volume: 2.80  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                         |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 12/Oct/21 14:28  | Sample Weight: 1.0000   |

### Chromatogram

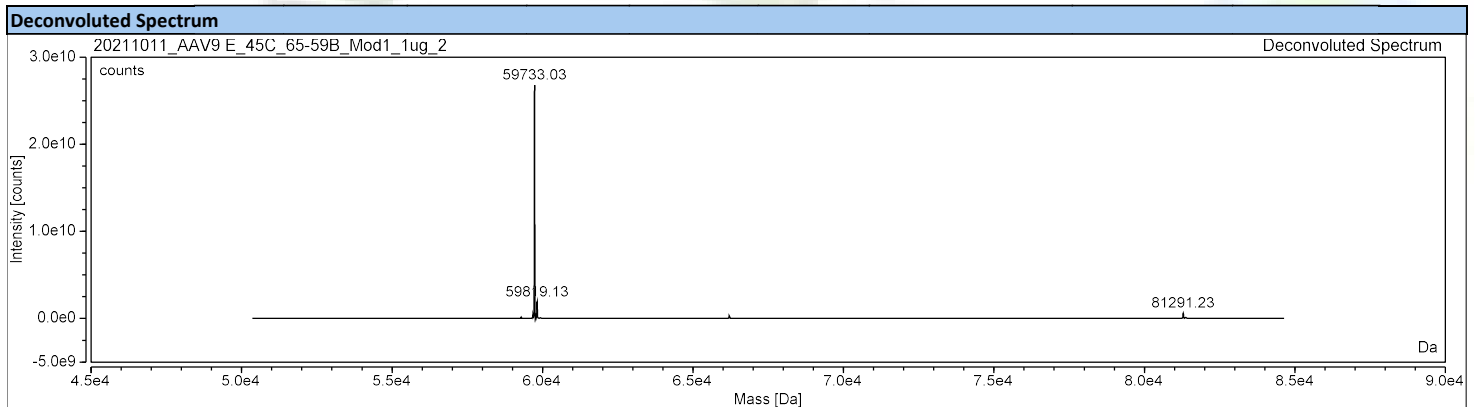
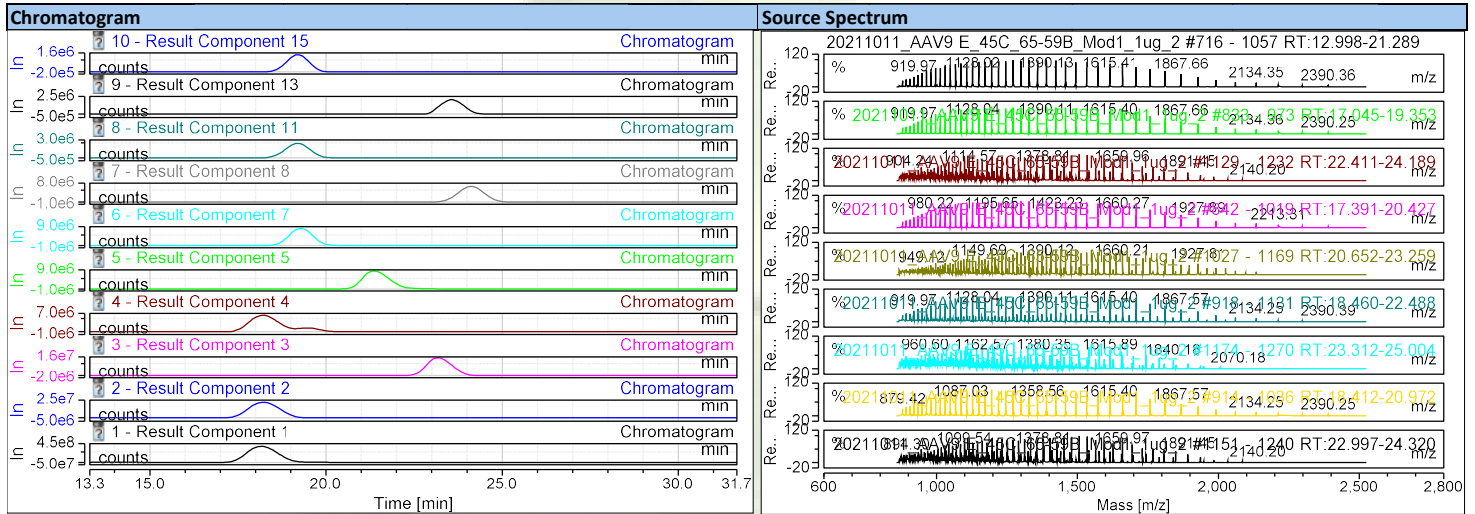


### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |                                       |   |
|-------------------|---------------------------------------|---|
| Injection Name:   | 20211011_AAV9 E_45C_65-59B_Mod1_1ug_2 | Injection Volume:                         |
| Vial Number:      | R:A5                                  | Instrument Method:                        |
| Injection Type:   | Unknown                               | Processing Method:                        |
| Run Time (min):   | 44.98                                 | Injection Date/Time:                      |
|                   |                                       | 2.80                                      |
|                   |                                       | AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
|                   |                                       | MS Quantitative_45C_65-59B                |
|                   |                                       | 12/Oct/21 15:14                           |



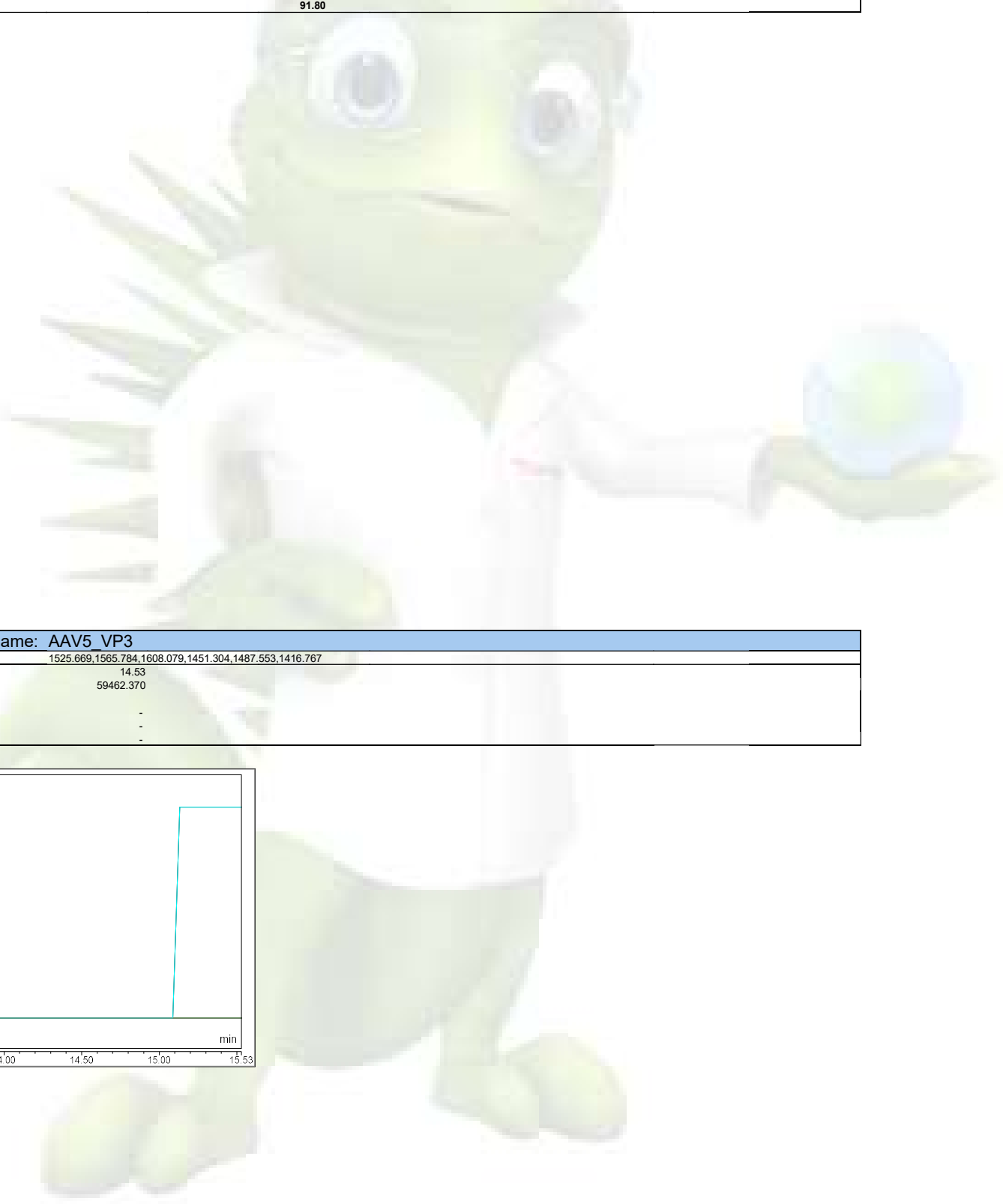
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

**Component Summary**

**Injection Details**

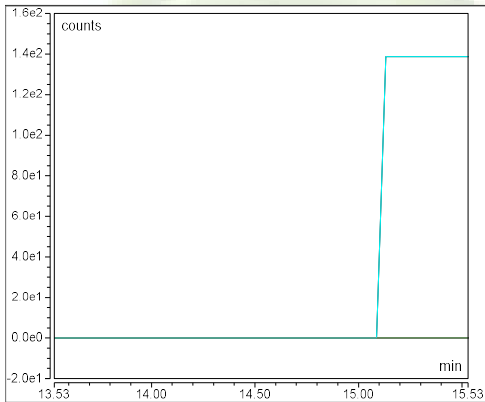
|                        |                                     |                             |   |
|------------------------|-------------------------------------|-----------------------------|---|
| <i>Injection Name:</i> | 20211011_AAV9_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i>    | 2.80  |
| <i>Vial Number:</i>    | R:A5                                | <i>Instrument Method:</i>   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |
| <i>Injection Type:</i> | Unknown                             | <i>Processing Method:</i>   | MS Quantitative_45C_65-59B                            |
| <i>Run Time (min):</i> | 44.98                               | <i>Injection Date/Time:</i> | 12/Oct/21 15:14                                       |

| Peak Name | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|-----------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV9_VP3  | 18.181                | 4513899.308        | 88.57              | 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907 | 59732.620 | PASS                           |
| AAV9_VP1  | 23.221                | 112639.998         | 2.21               | 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636 | 81290.340 | PASS                           |
| AAV9_VP2  | 24.159                | 52114.190          | 1.02               | 1409.730,1162.589,1472.334,1274.267,1380.371,1352.231 | 66209.760 | PASS                           |
|           |                       |                    | <b>91.80</b>       |   |           |                                |



**Component Name: AAV5\_VP3**

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |





**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

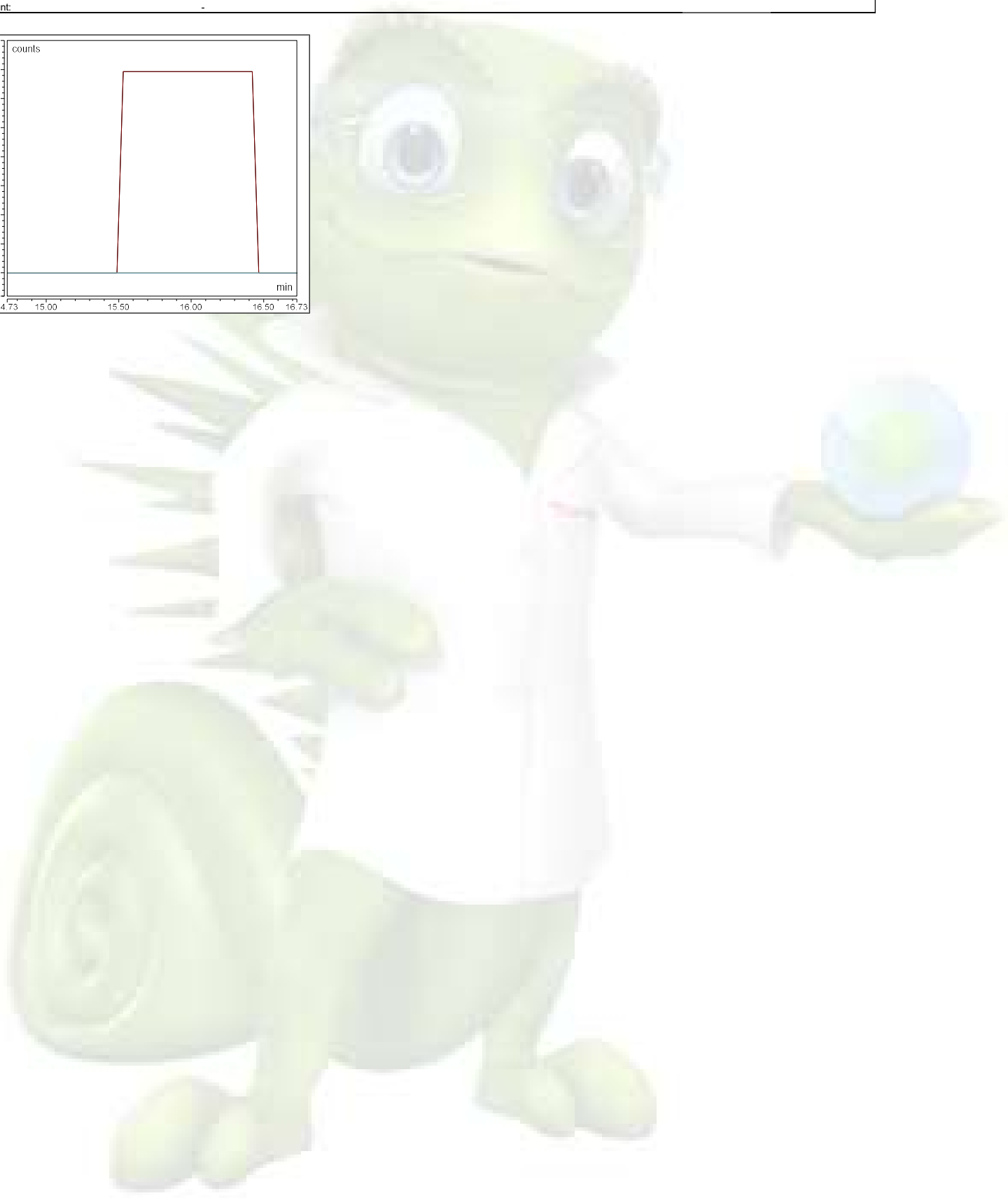
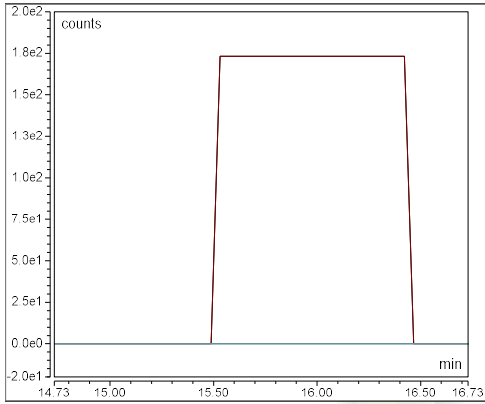
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

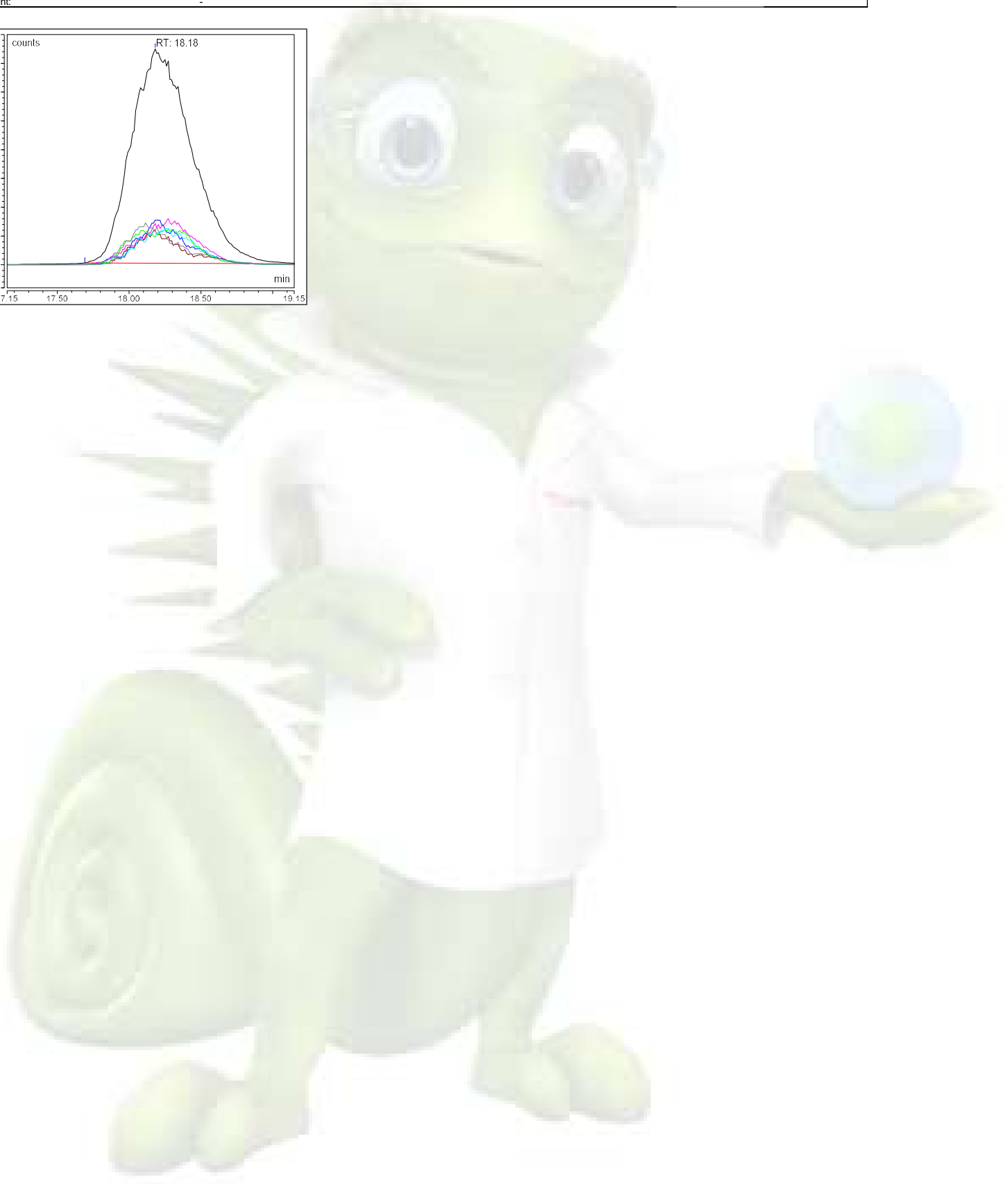
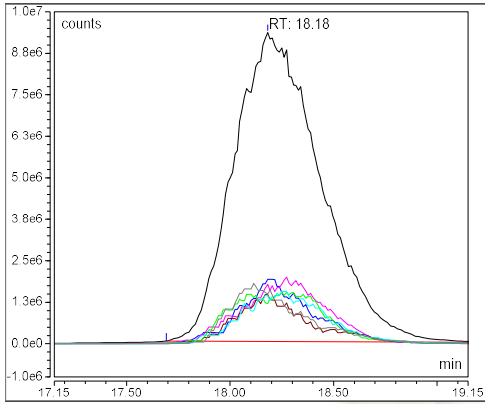
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): 4513899

Intensity (counts): 9305524

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

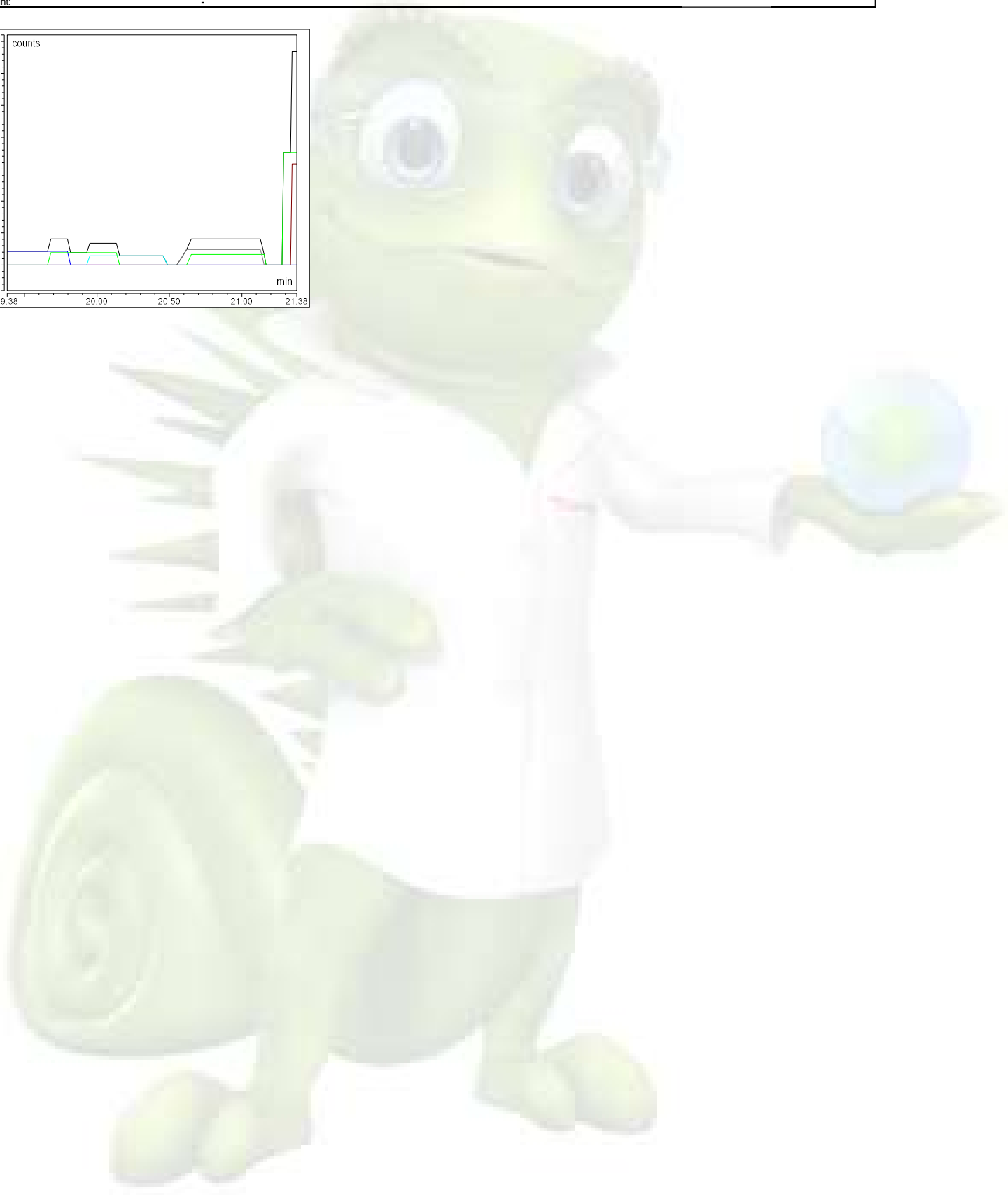
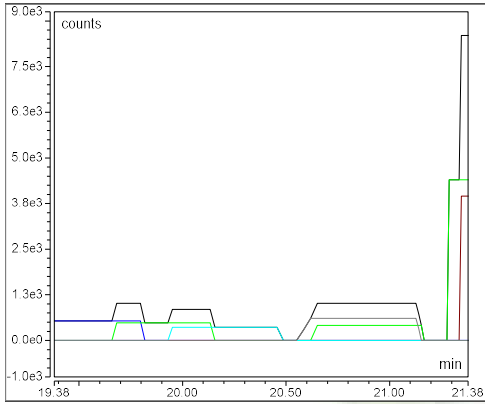
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

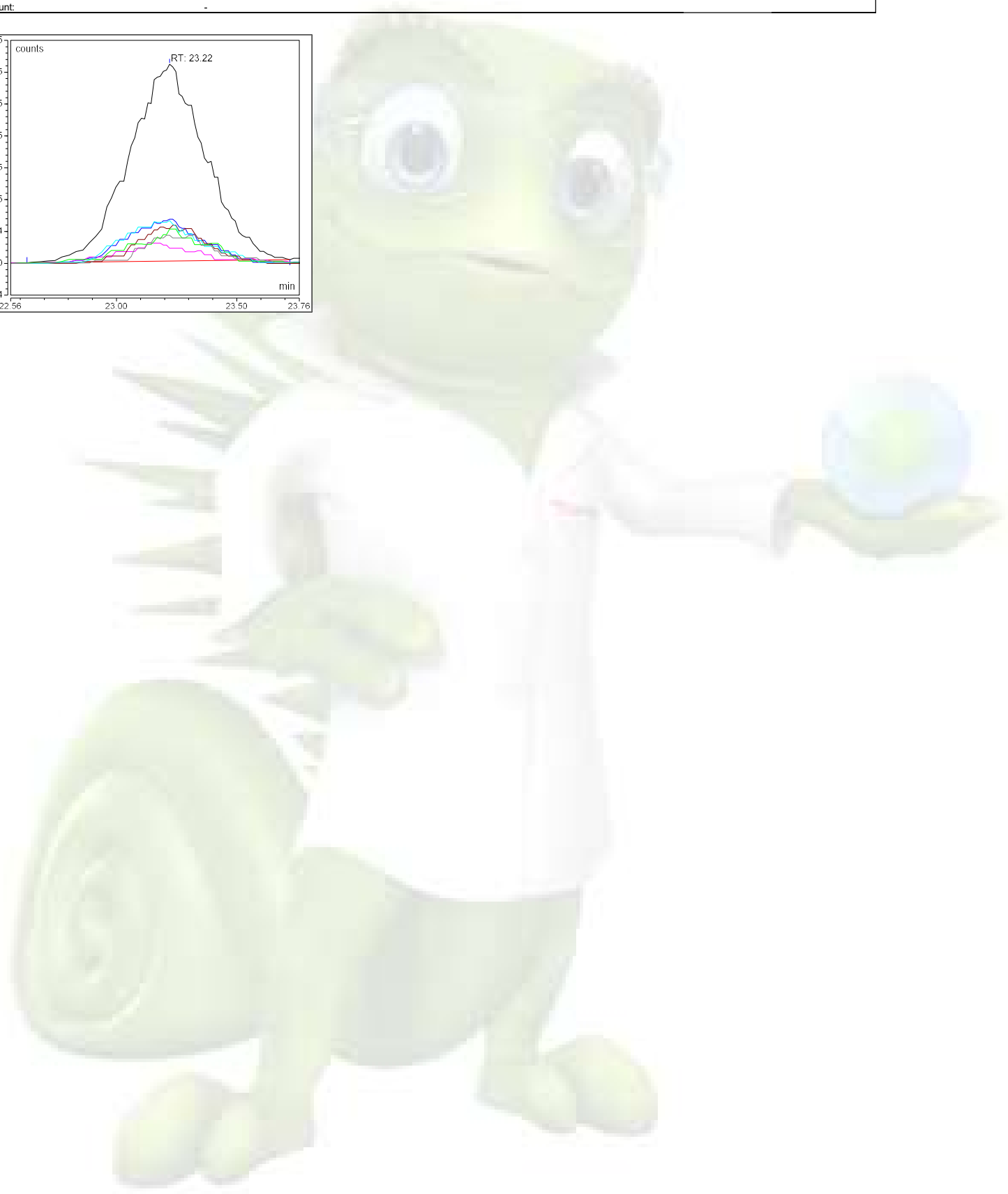
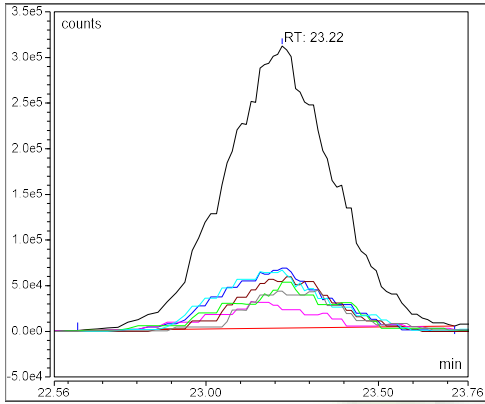
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): 112640

Intensity (counts): 308856

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

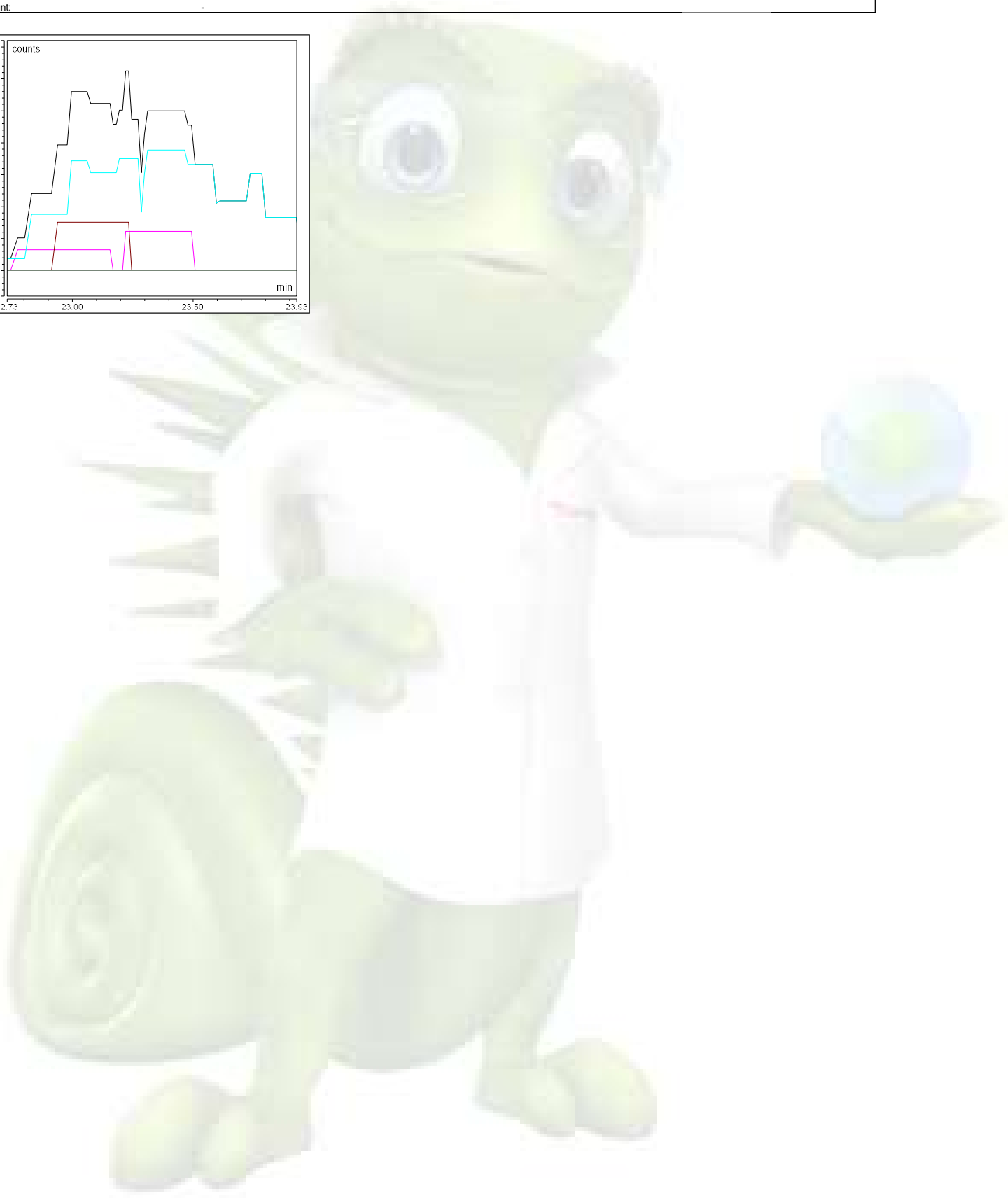
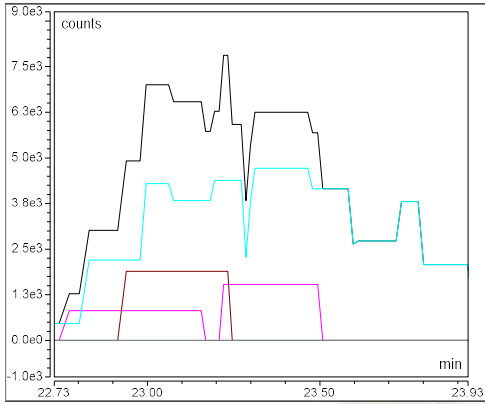
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

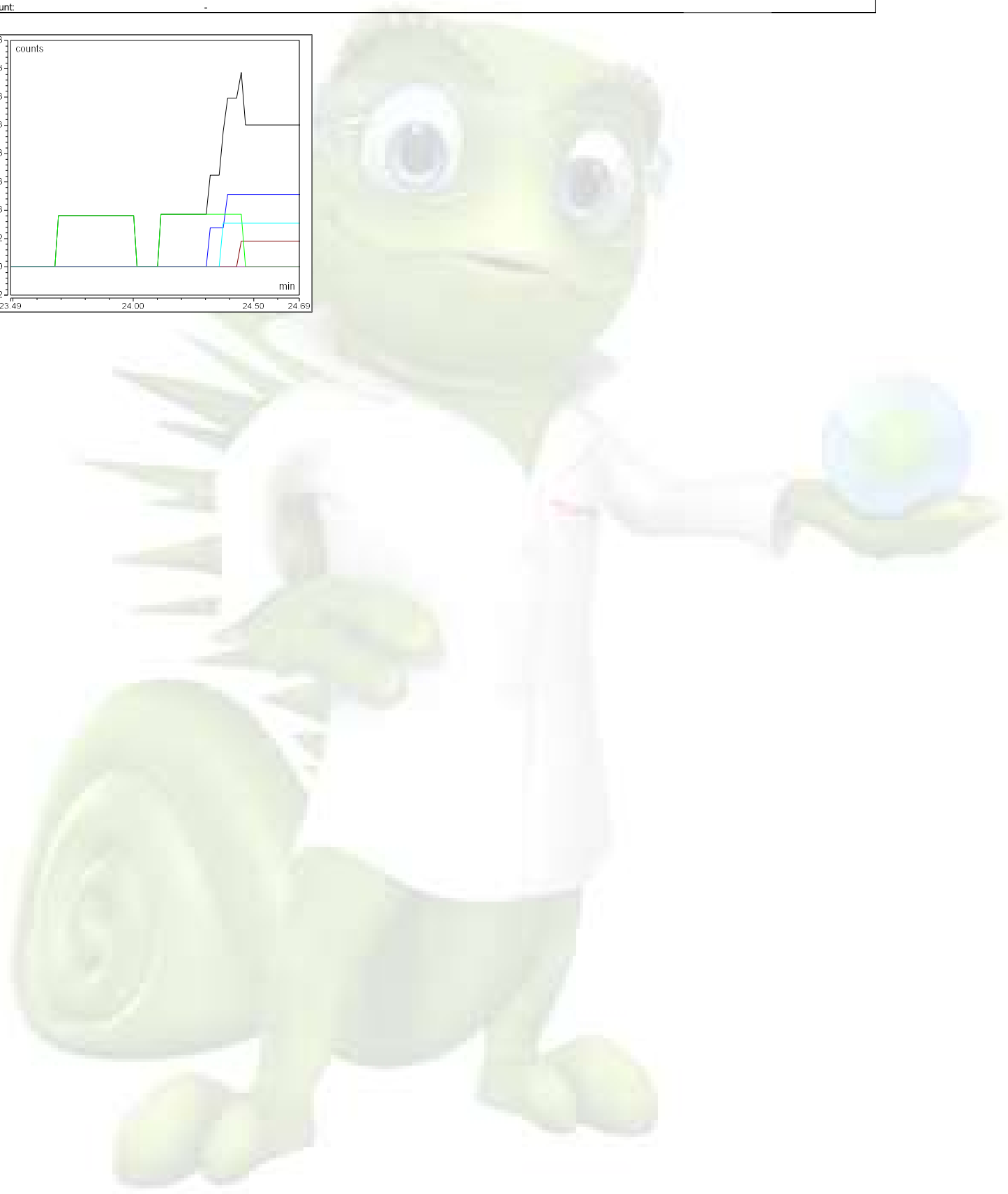
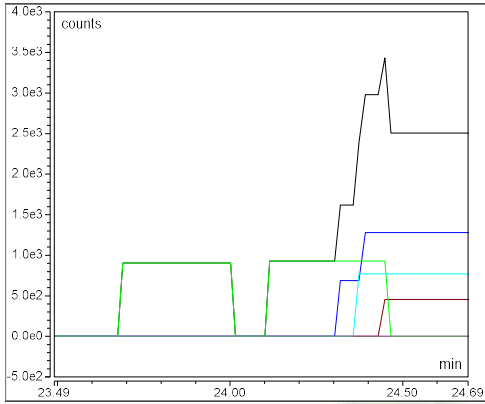
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

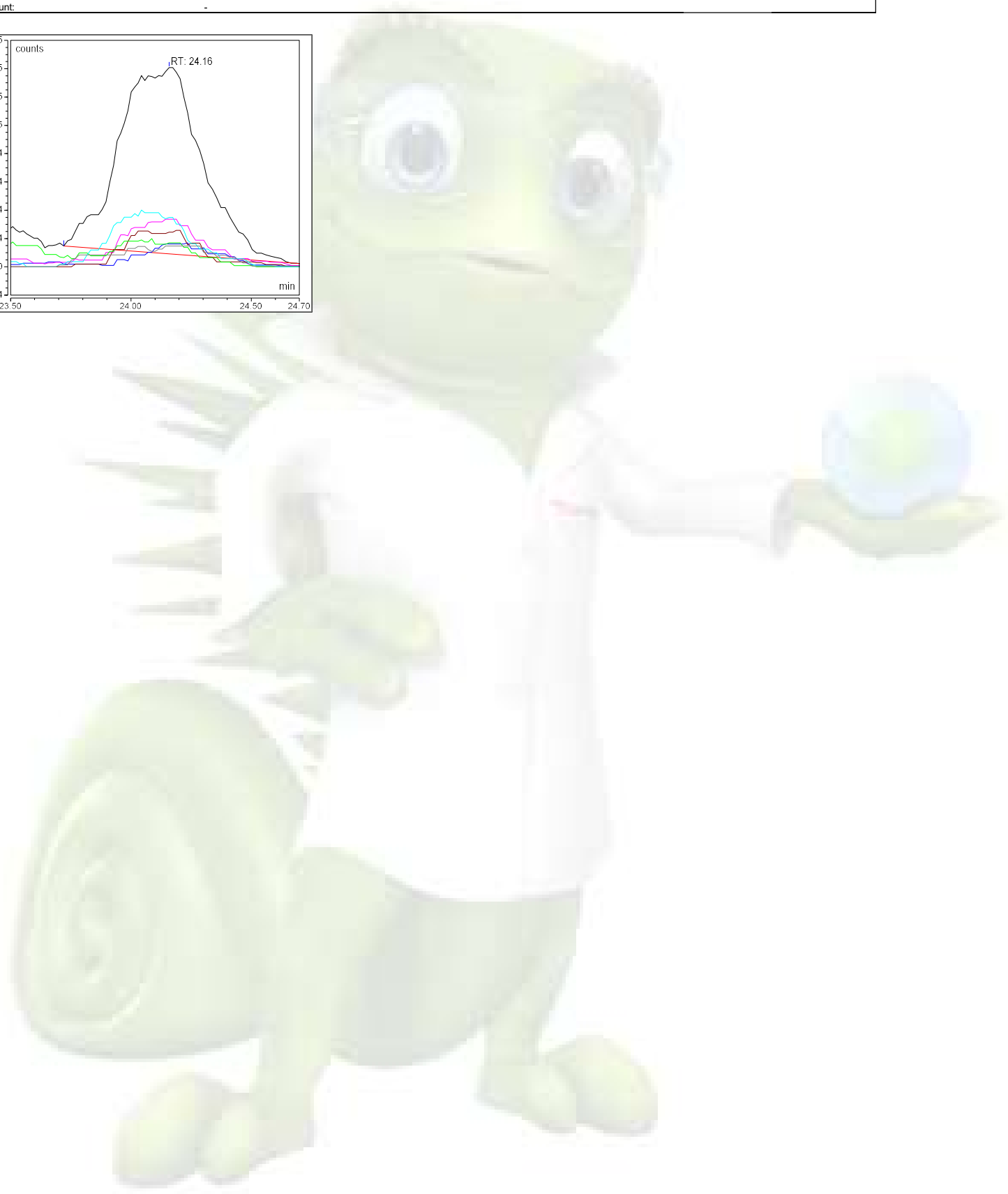
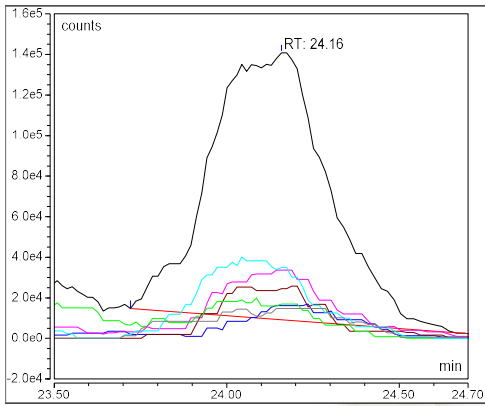
Intensity (counts): -

Amount: -



**Component Name: AAV9 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231 |
| Retention time (min): | 24.10   |
| Expected Mass:        | 66209.760   |
| Area (count*min):     | 52114   |
| Intensity (counts):   | 131634  |
| Amount:               | -   |



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

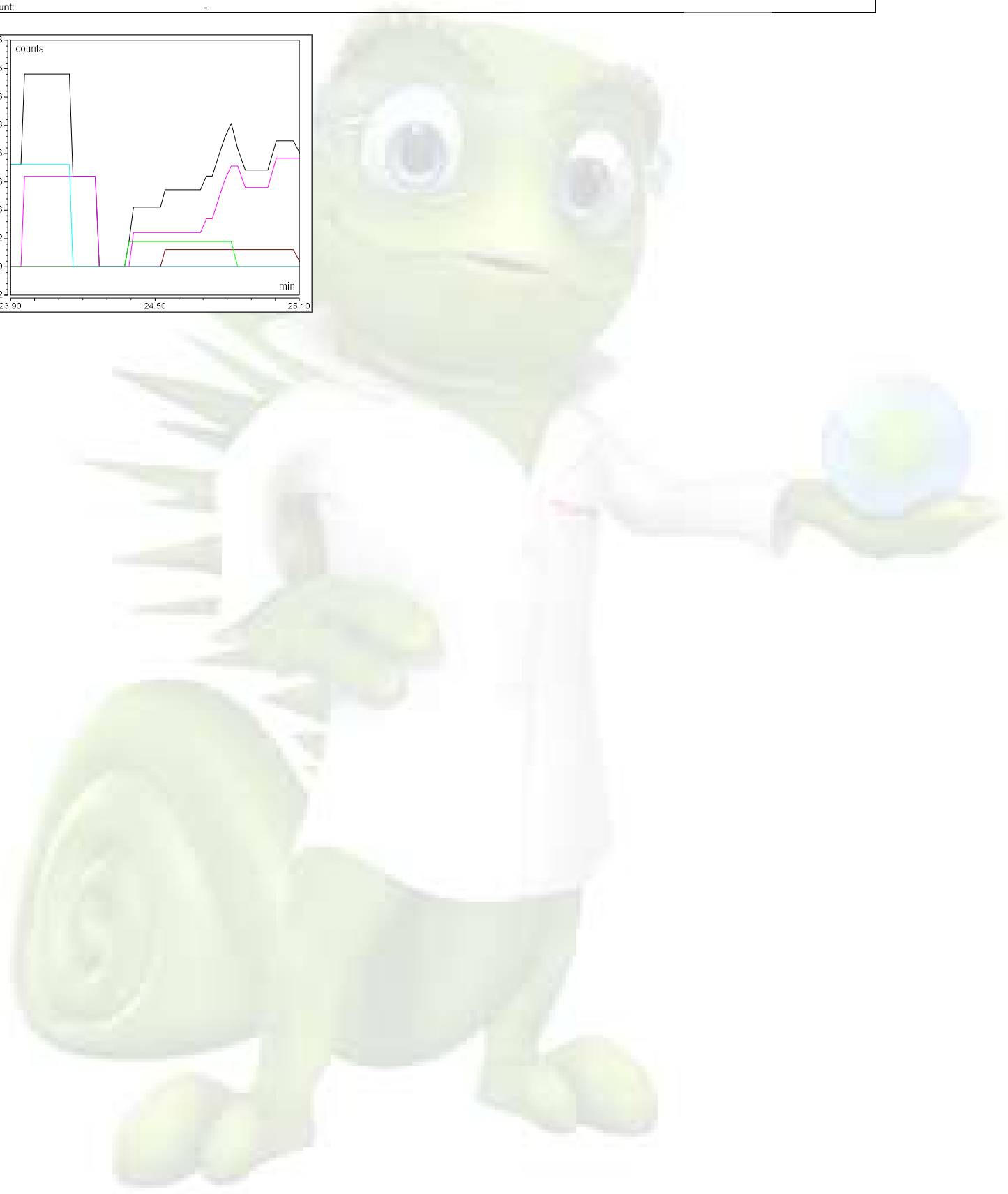
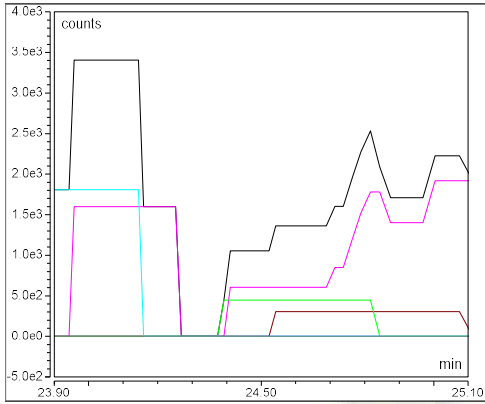
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

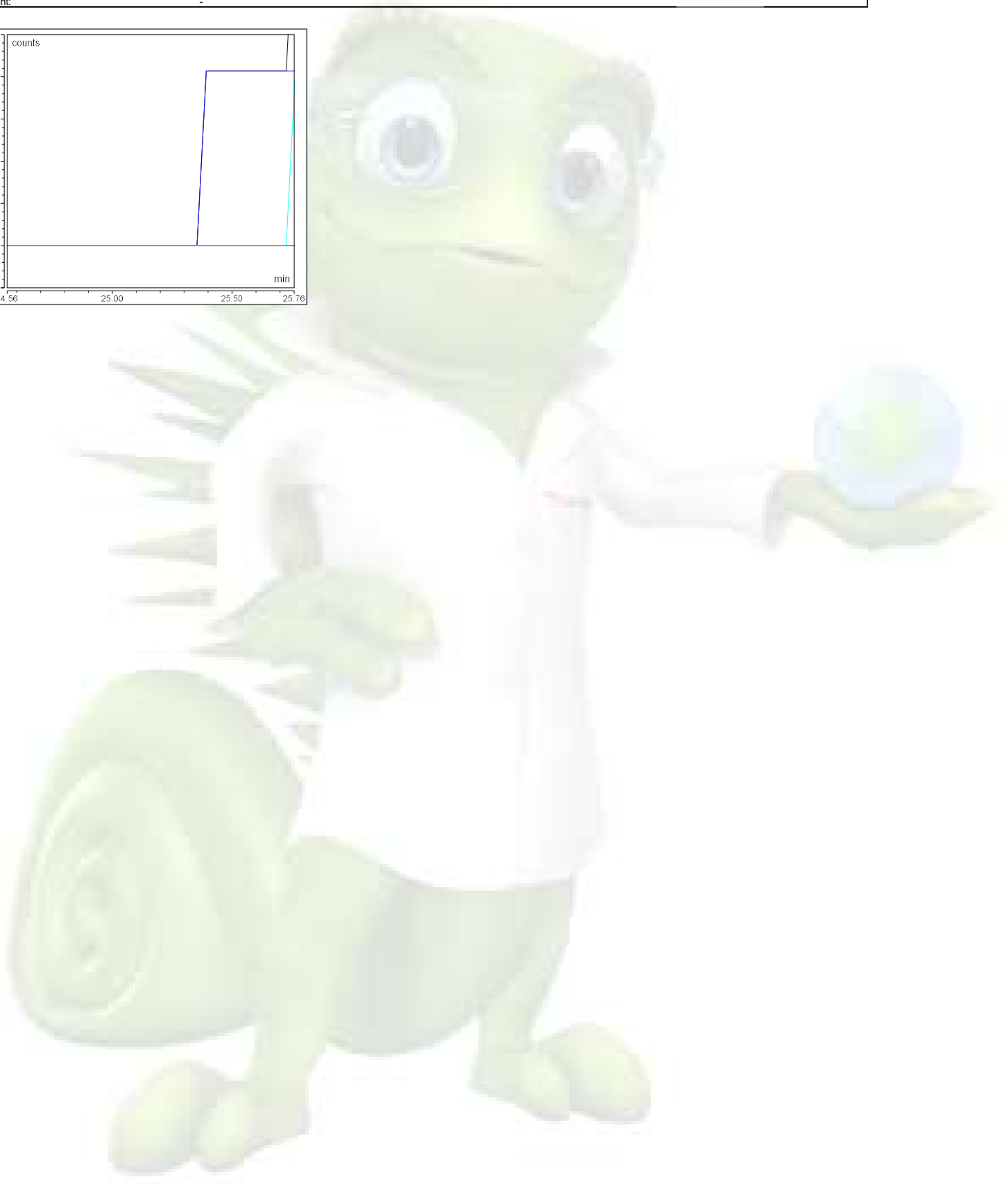
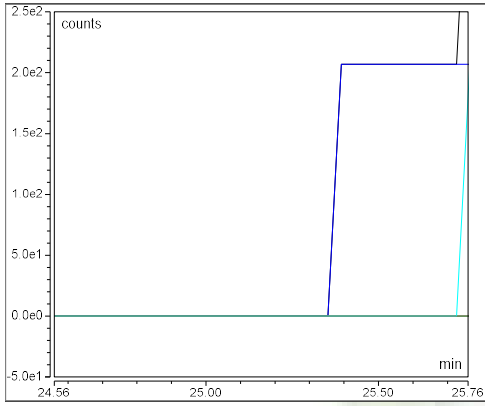
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

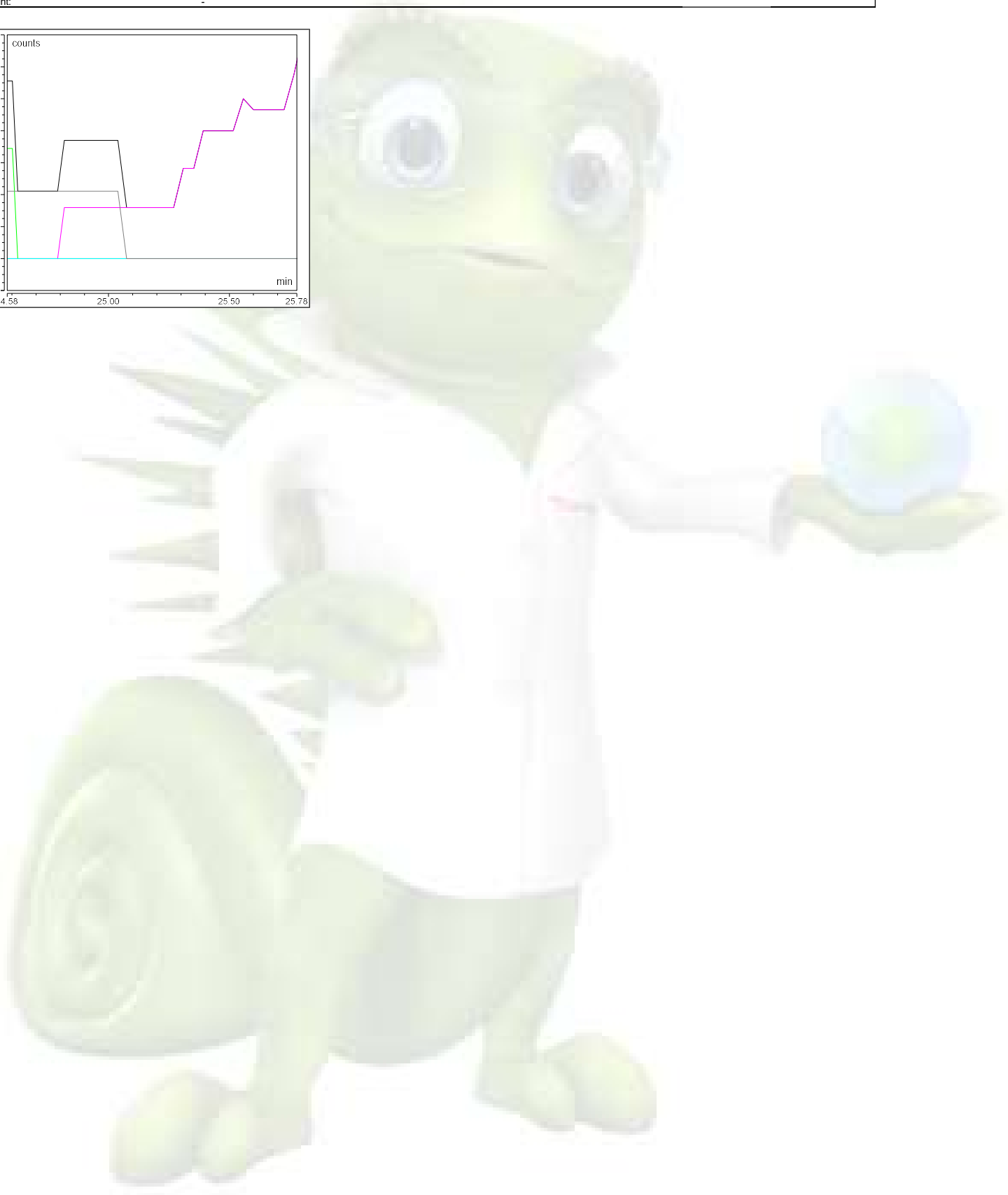
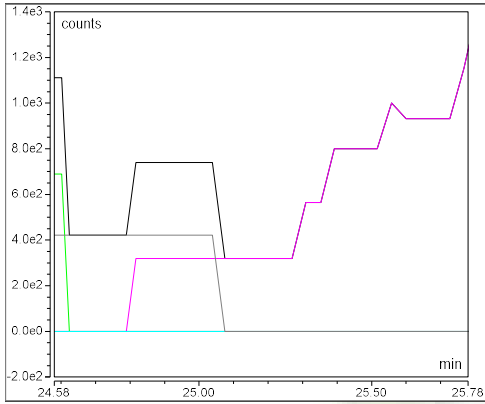
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

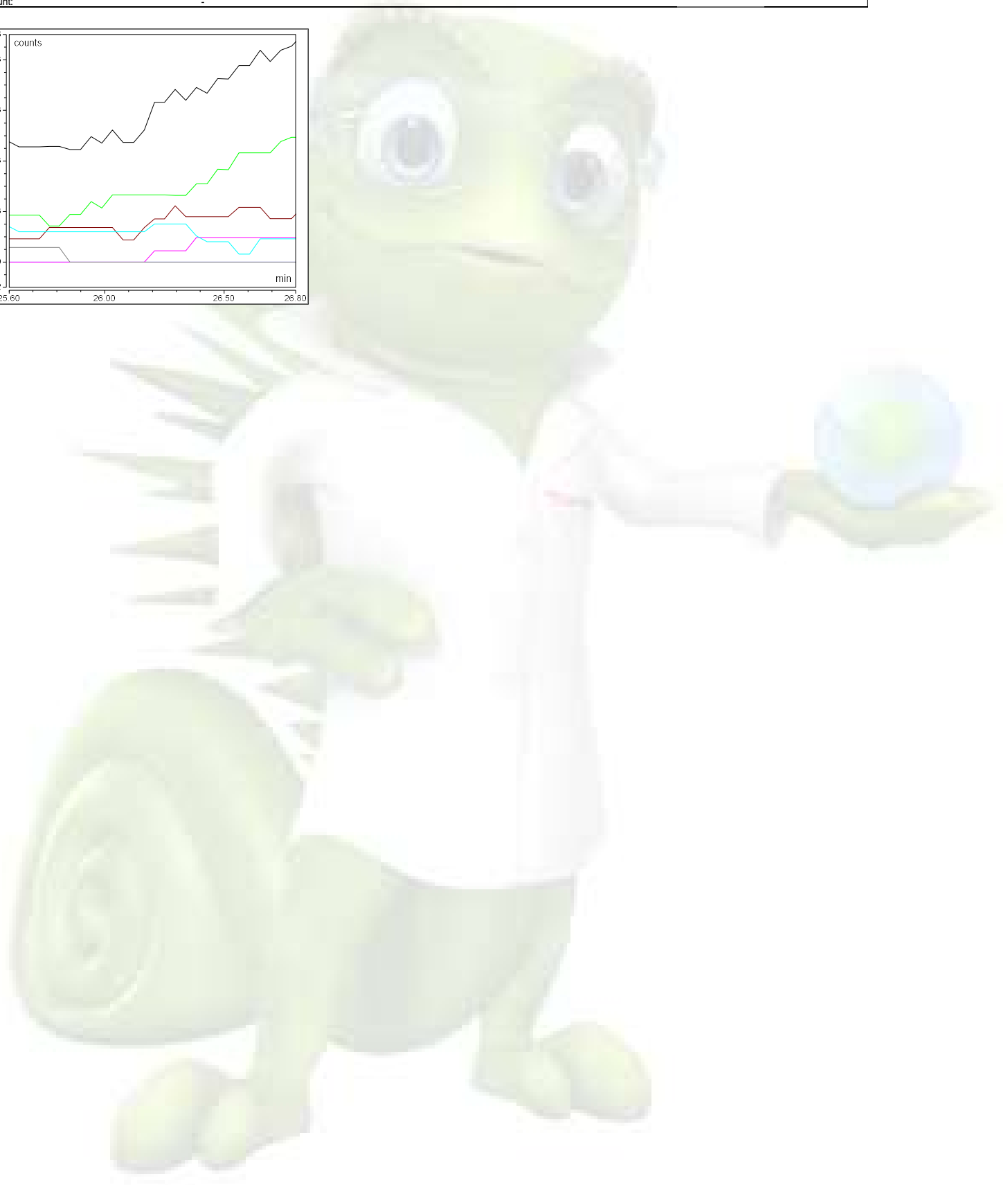
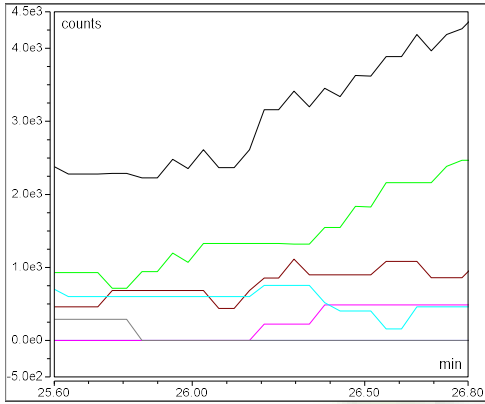
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

Amount: -

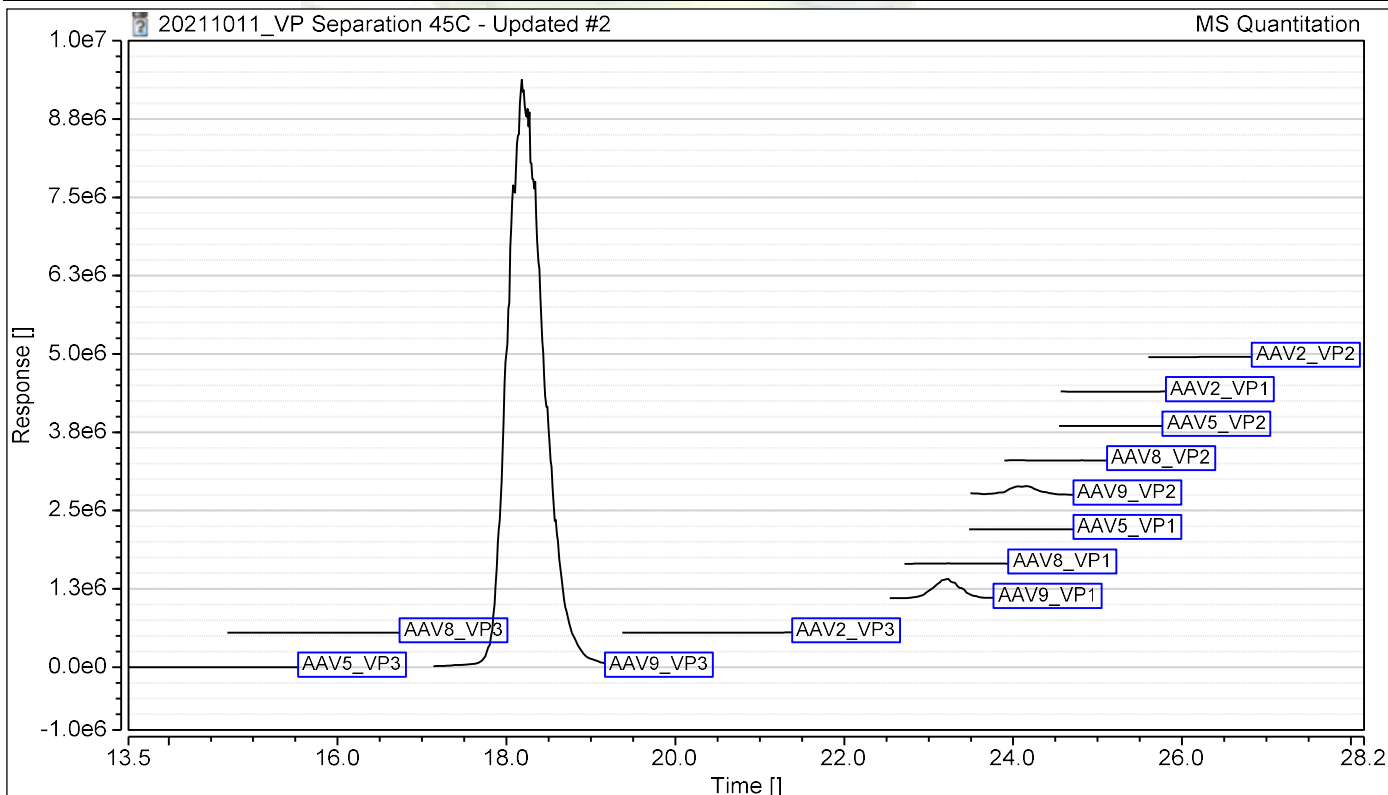


## Chromatogram and SST Results

### Injection Details

|                      |  |                   |        |
|----------------------|--|-------------------|--------|
| Injection Name:      | 20211011_AAV9 E_45C_65-59B_Mod1_1ug_2                                      | Run Time (min):   | 44.98  |
| Vial Number:         | R:A5   | Injection Volume: | 2.80   |
| Injection Type:      | Unknown  |                   |        |
| Calibration Level:   |  |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 |                   | S      |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 12/Oct/21 15:14  | Sample Weight:    | 1.0000 |

### Chromatogram

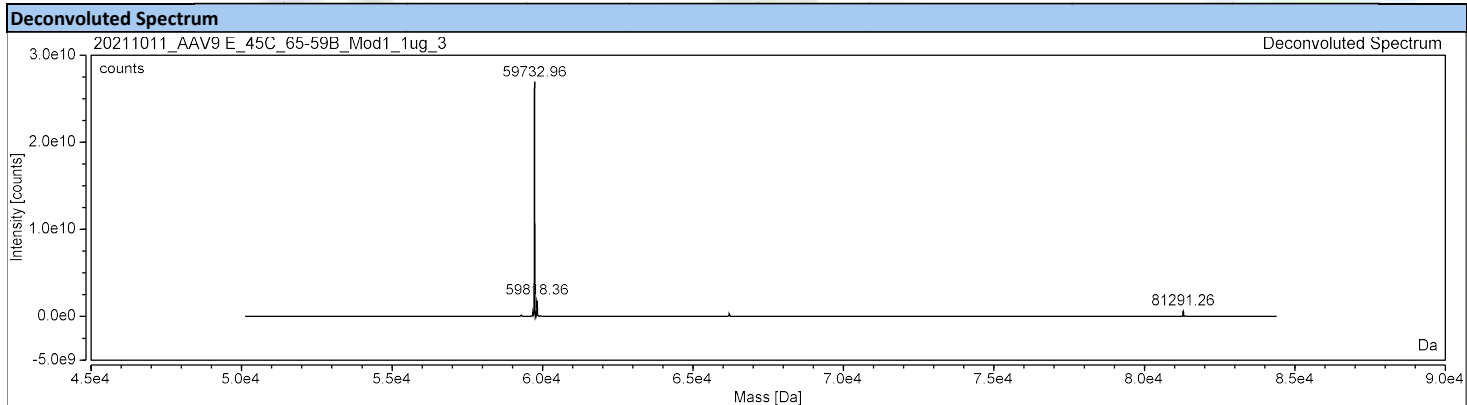
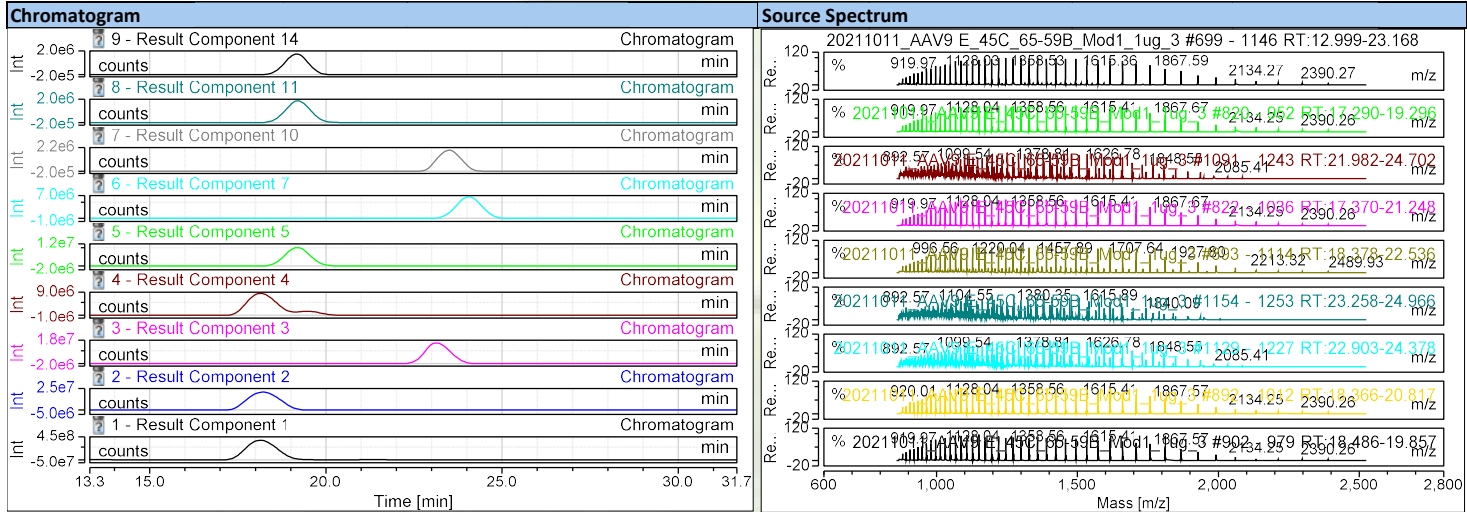


### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV9 E_45C_65-59B_Mod1_1ug_3 | Injection Volume: 2.80                                       |
| Vial Number:      | R:A5                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 45.00                                 | Injection Date/Time: 12/Oct/21 16:00                         |



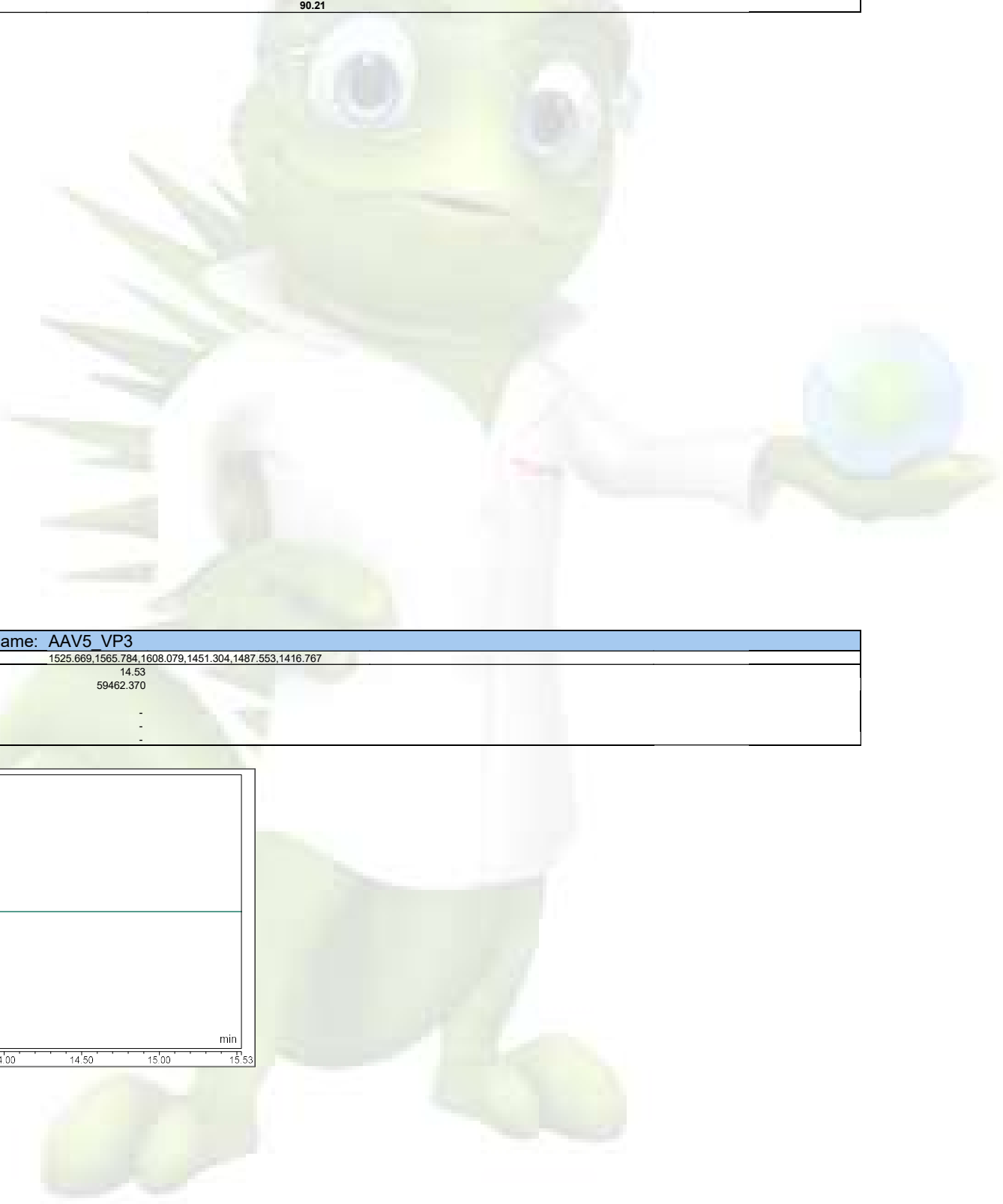
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

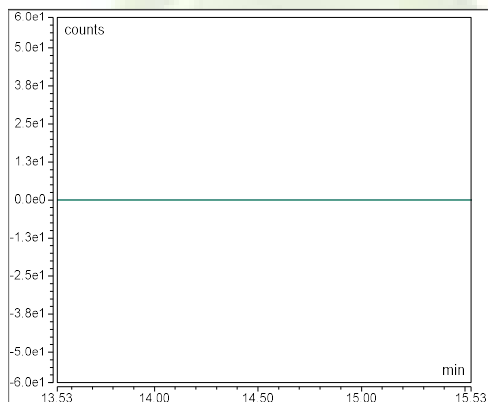
|  |   |  |
|--|---|--|
| <i>Injection Name:</i> 20211011_AAV9_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i> 2.80   |  |
| <i>Vial Number:</i> R:A5                                   | <i>Instrument Method:</i> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <i>Injection Type:</i> Unknown                             | <i>Processing Method:</i> MS Quantitative_45C_65-59B                            |  |
| <i>Run Time (min):</i> 45.00                               | <i>Injection Date/Time:</i> 12/Oct/21 16:00                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV9_VP3     | 18.149                | 4808333.069        | 87.11              | 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907 | 59732.620 | PASS                           |
| AAV9_VP1     | 23.155                | 122883.800         | 2.23               | 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636 | 81290.340 | PASS                           |
| AAV9_VP2     | 24.010                | 48701.492          | 0.88               | 1409.730,1162.589,1472.334,1274.267,1380.371,1352.231 | 66209.760 | PASS                           |
| <b>90.21</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

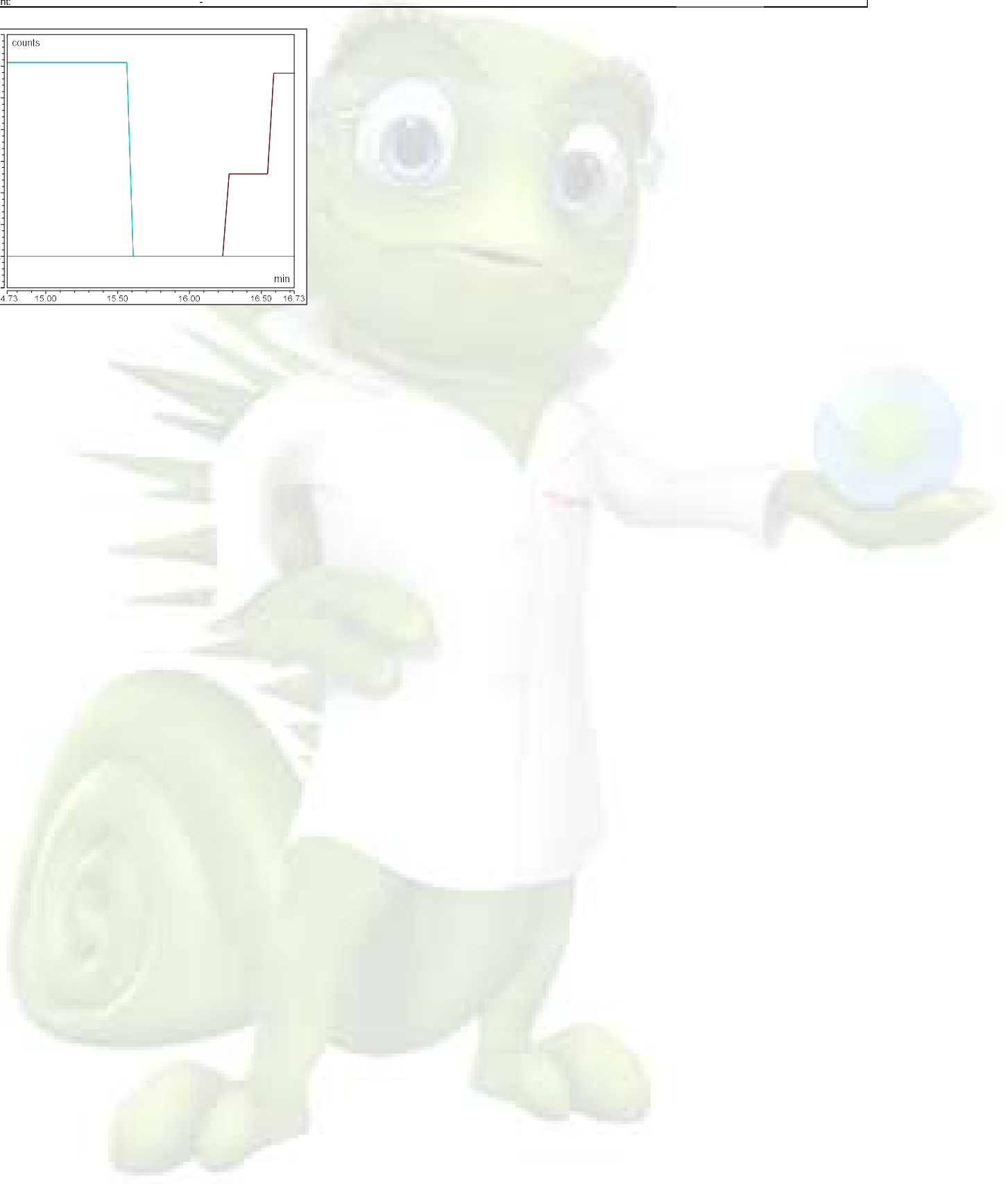
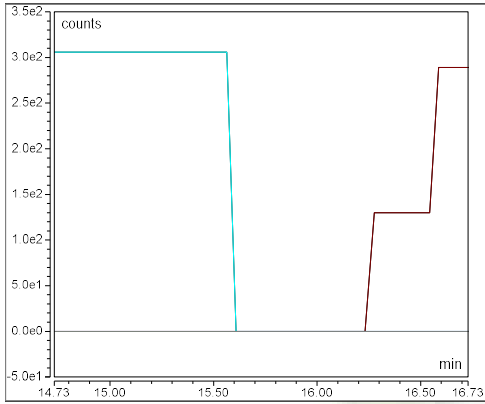
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

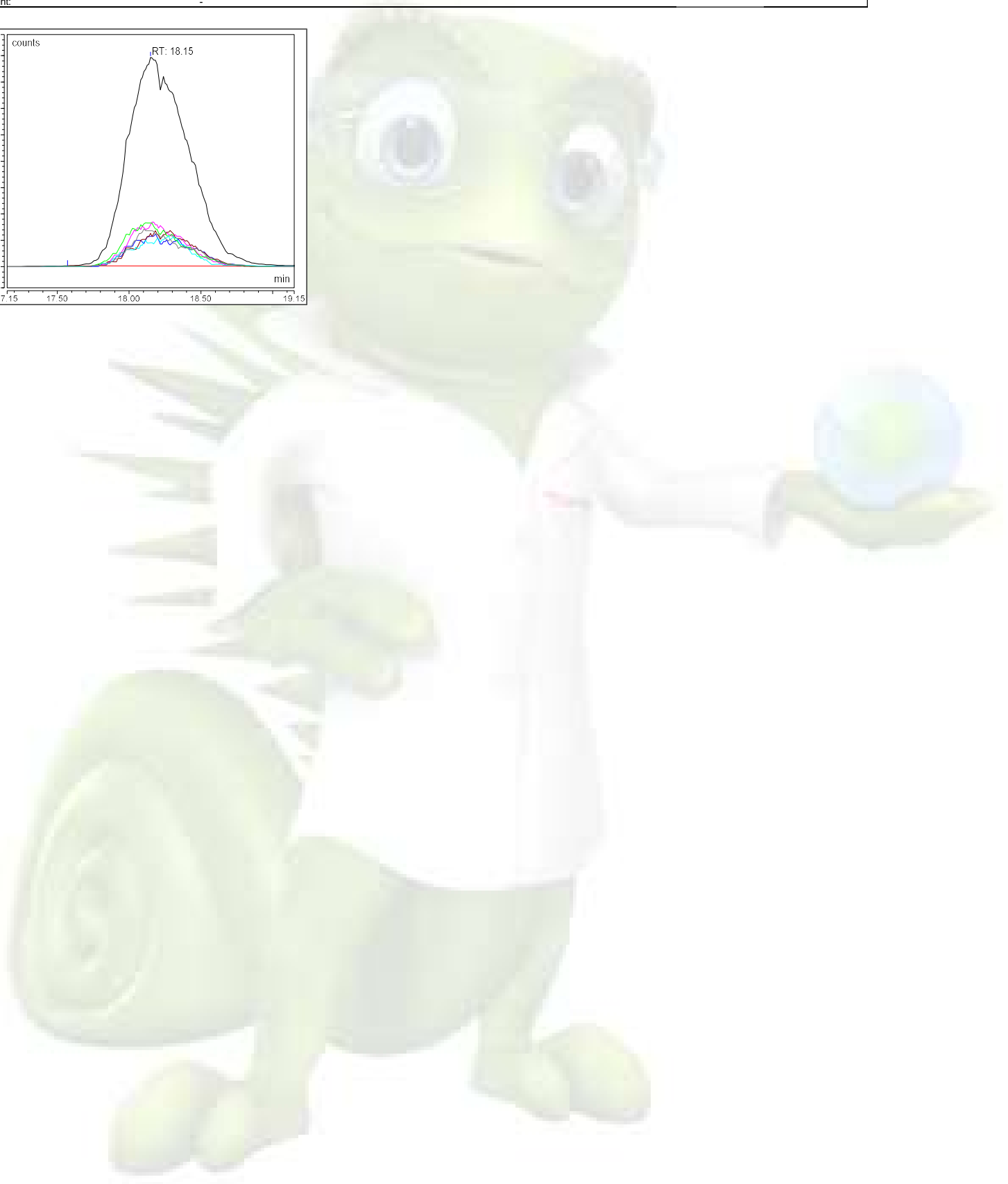
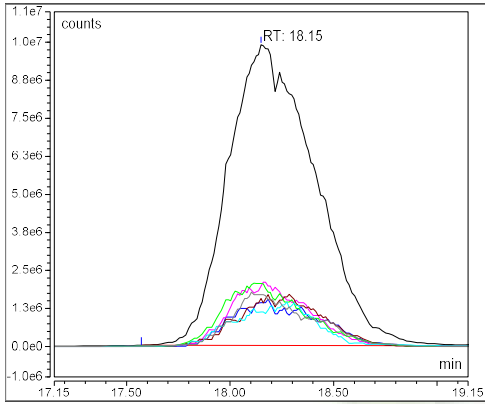
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): 4808333

Intensity (counts): 9879358

Amount: -





**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

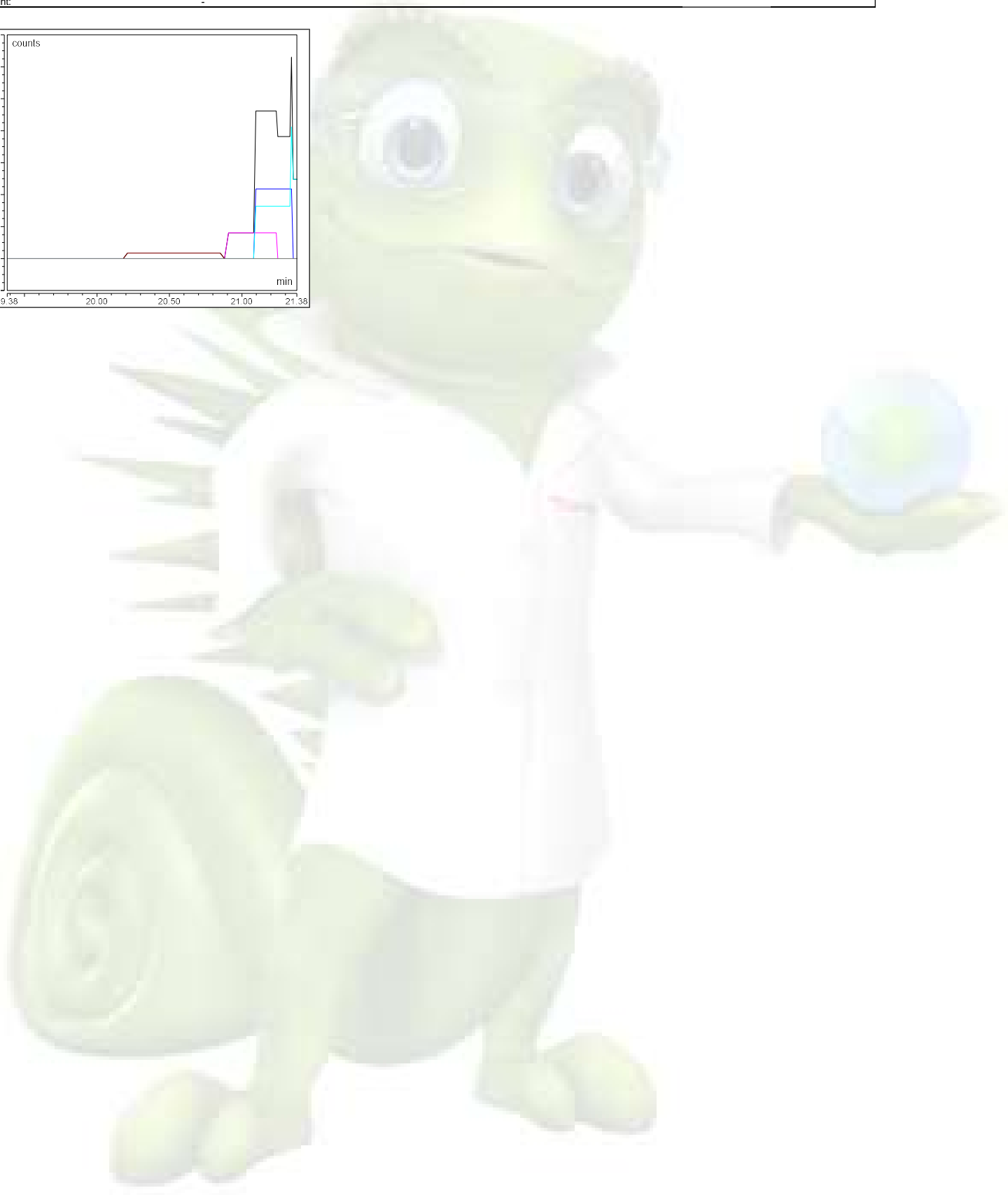
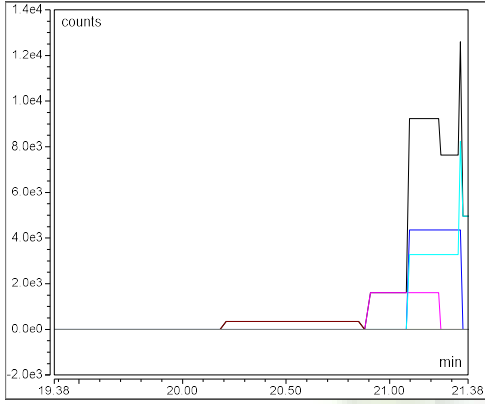
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

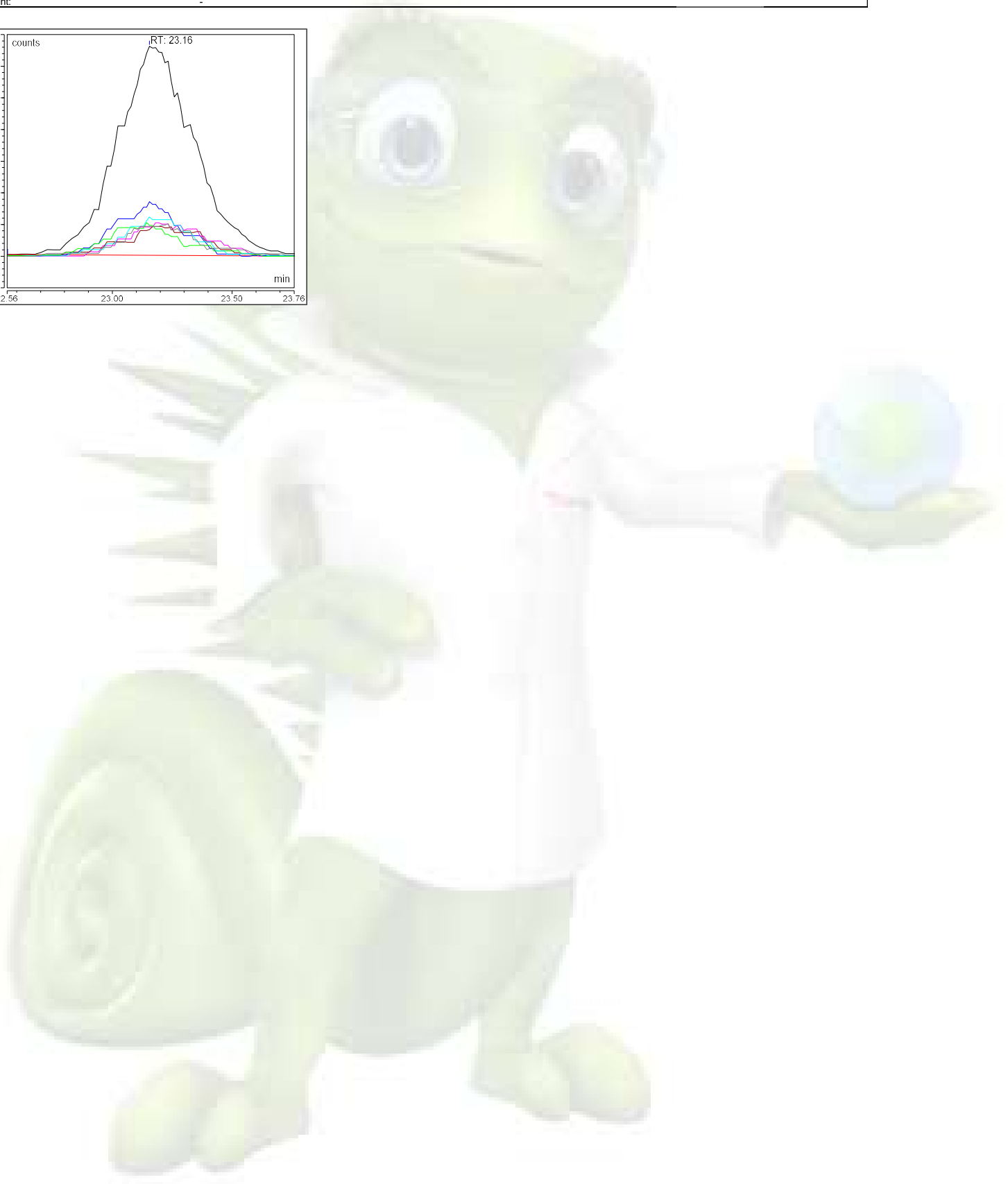
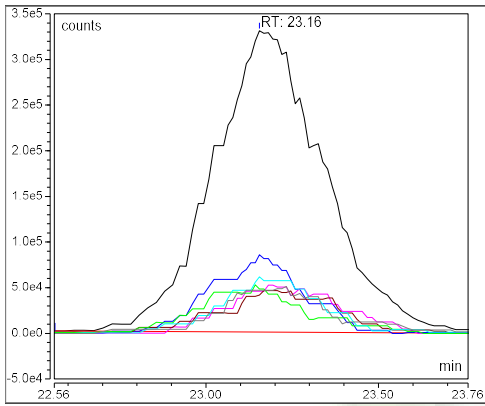
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): 122884

Intensity (counts): 330166

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

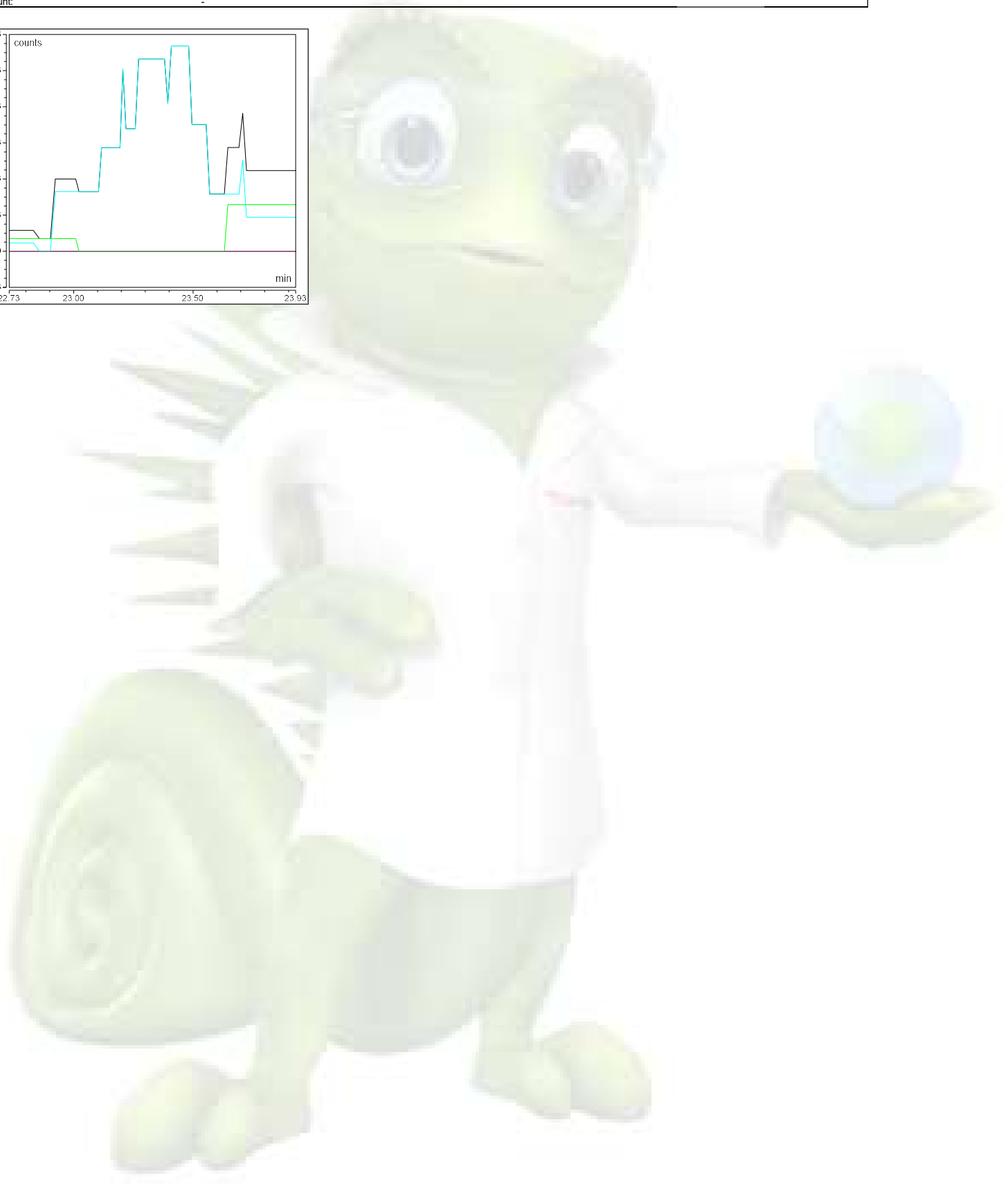
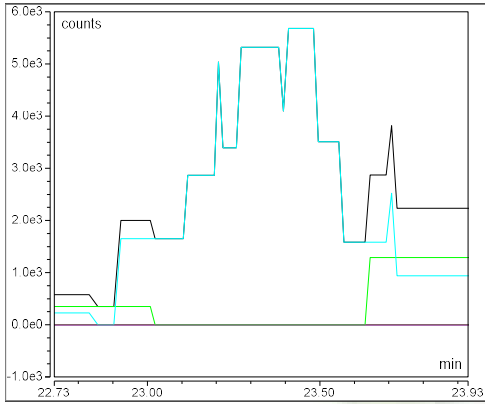
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

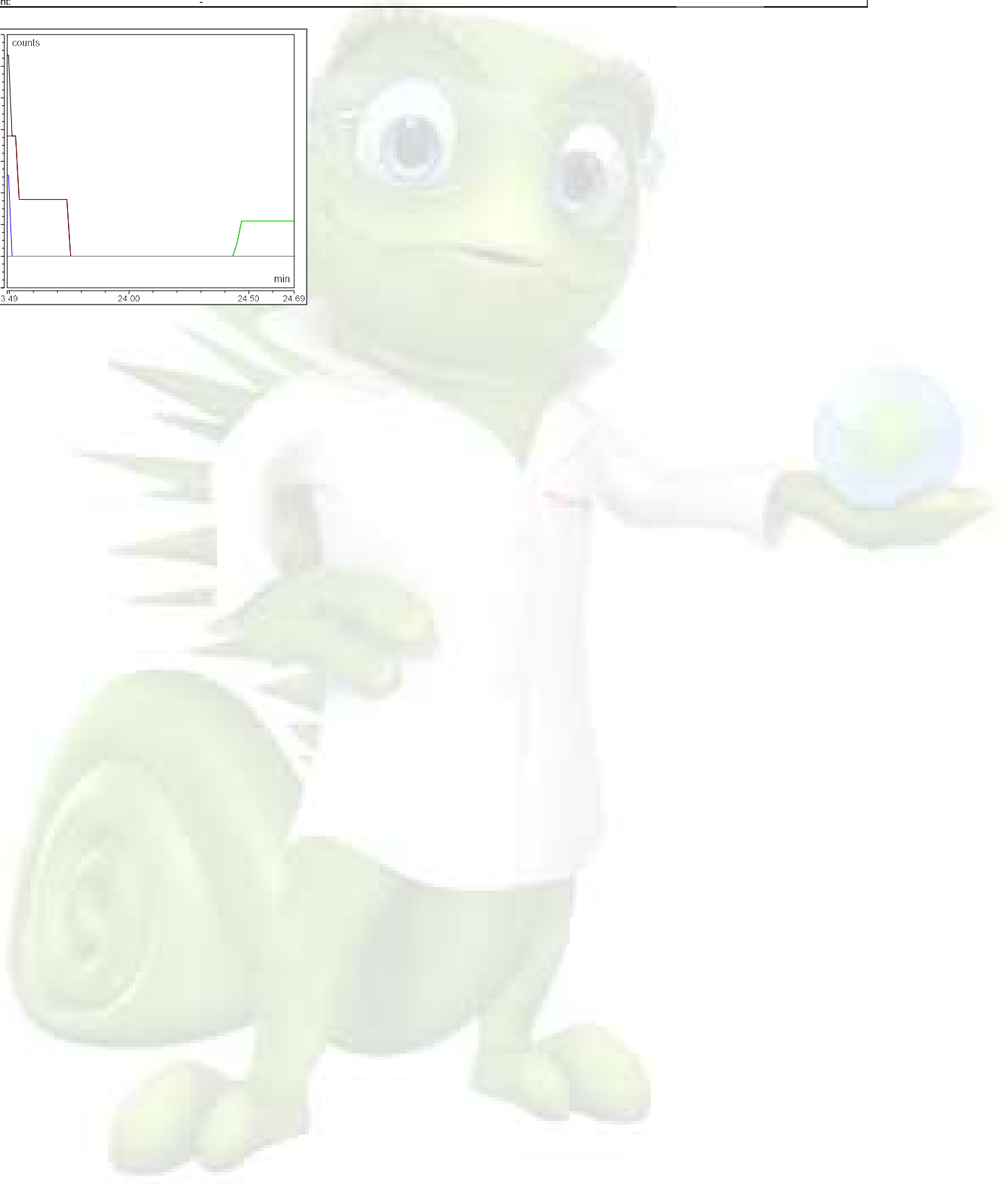
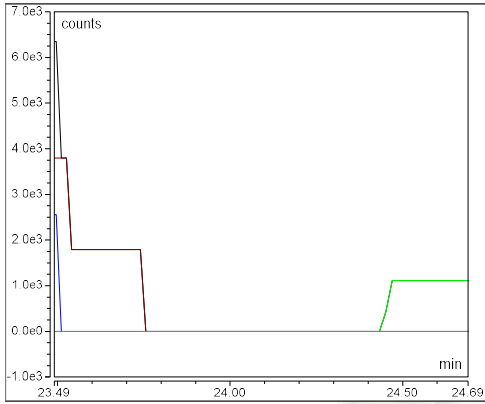
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

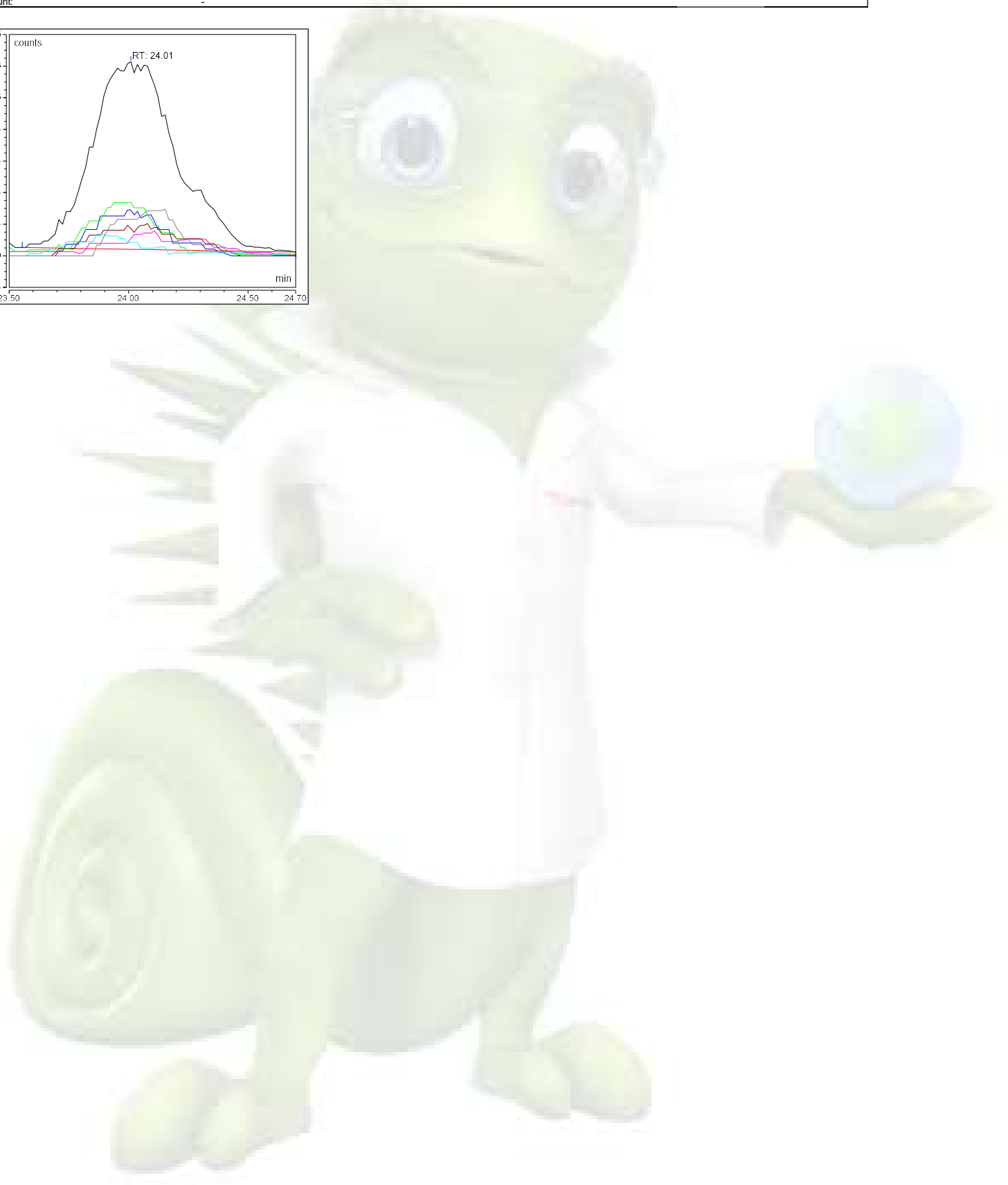
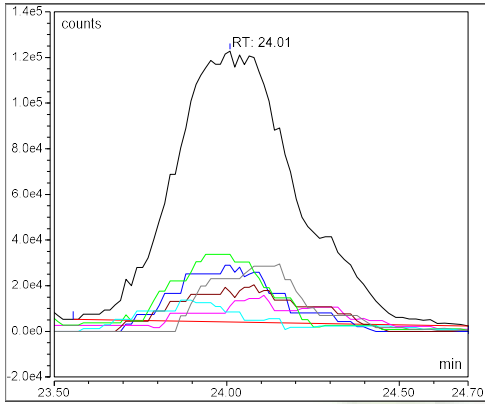
Intensity (counts): -

Amount: -



**Component Name: AAV9 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231 |
| Retention time (min): | 24.10   |
| Expected Mass:        | 66209.760   |
| Area (count*min):     | 48701   |
| Intensity (counts):   | 118626  |
| Amount:               | -   |



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

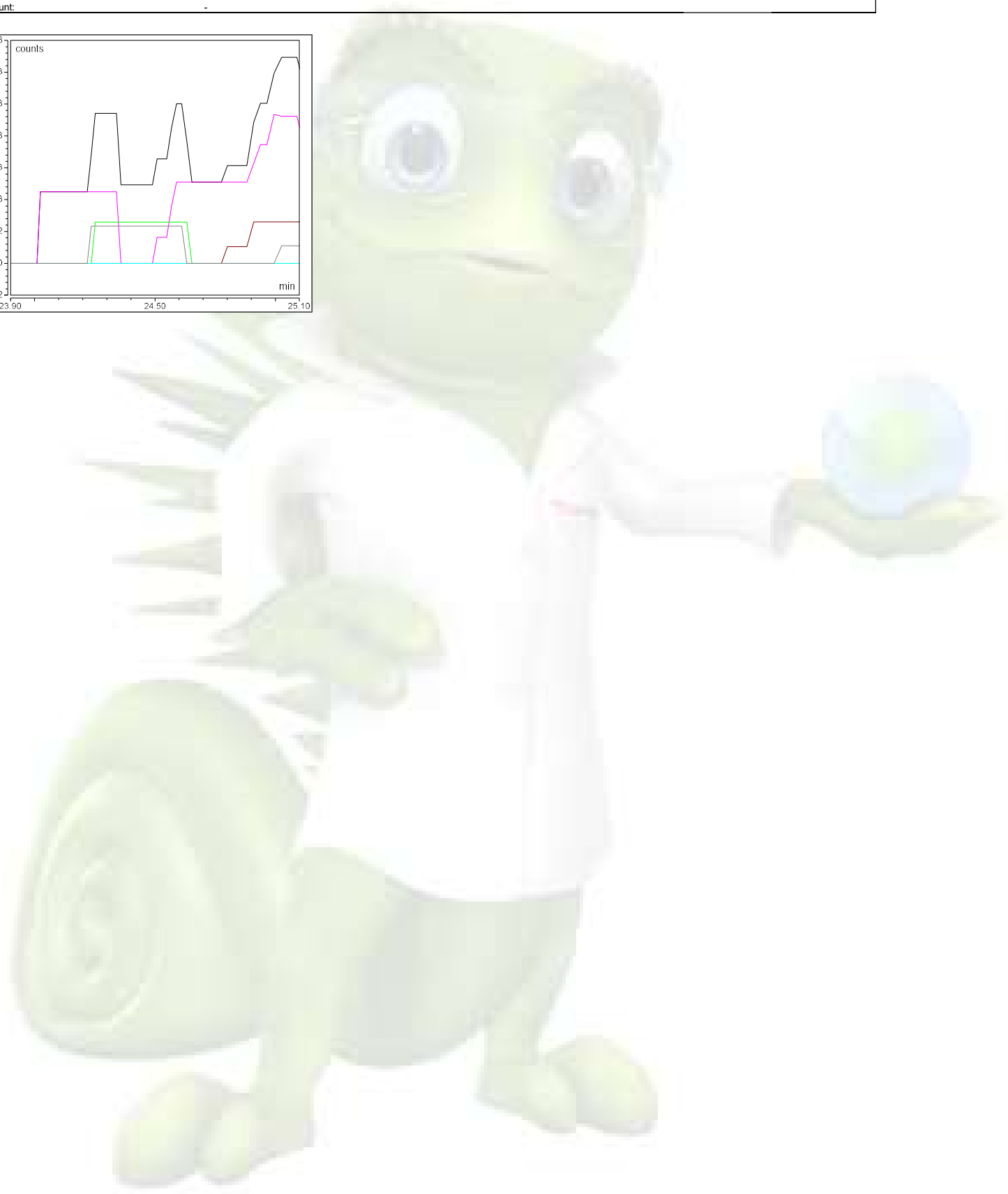
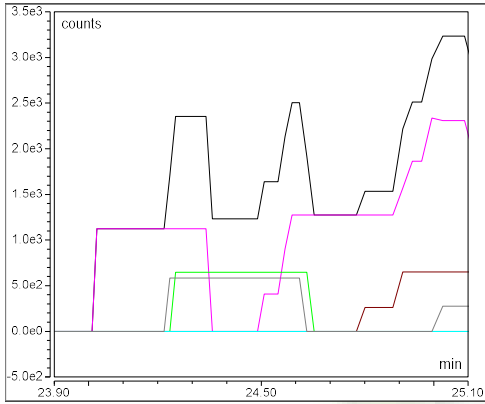
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

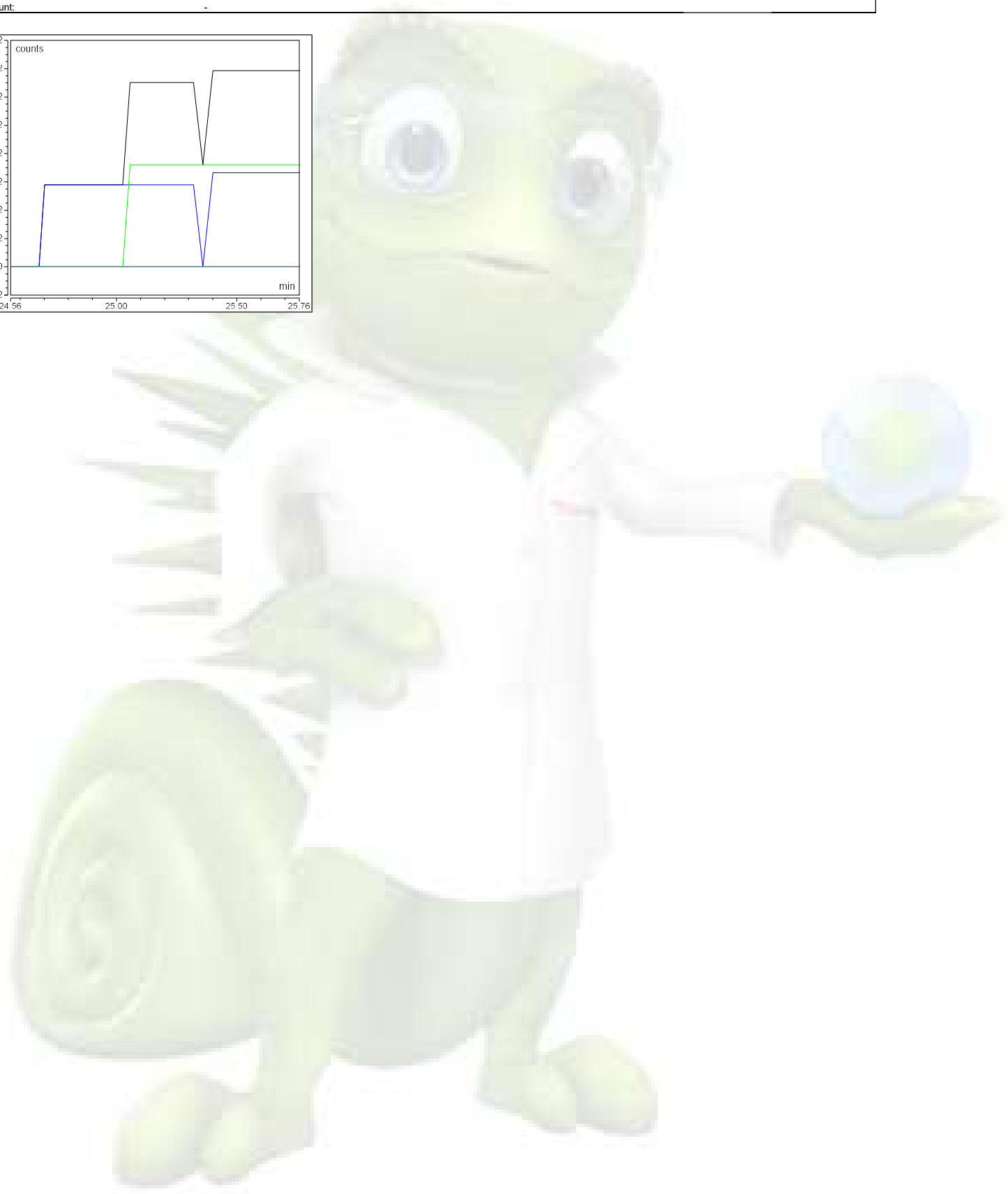
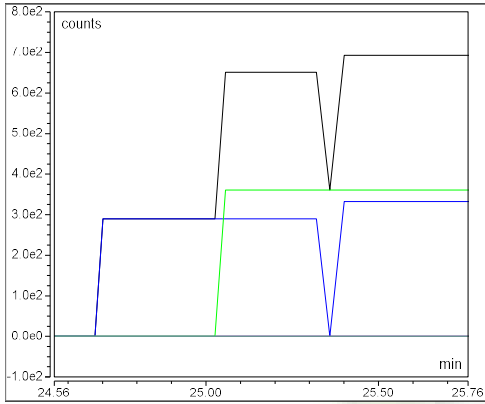
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

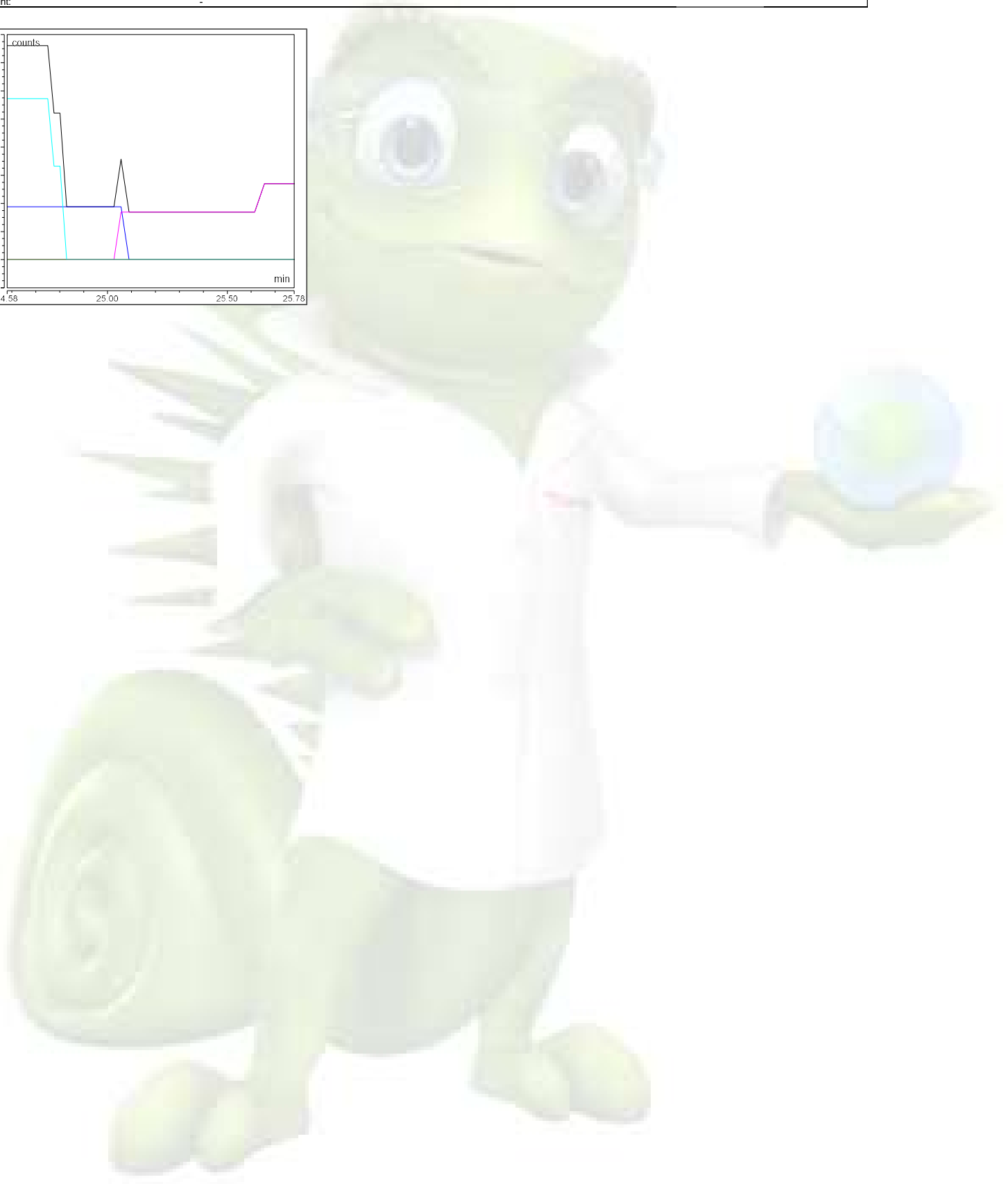
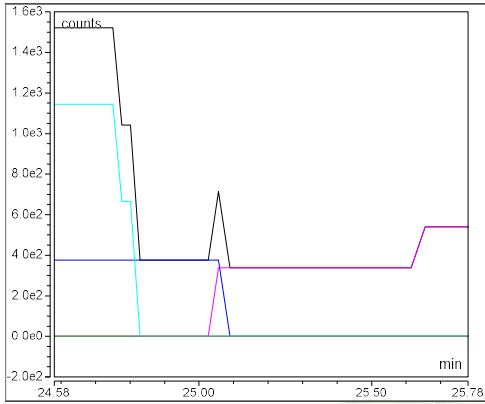
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

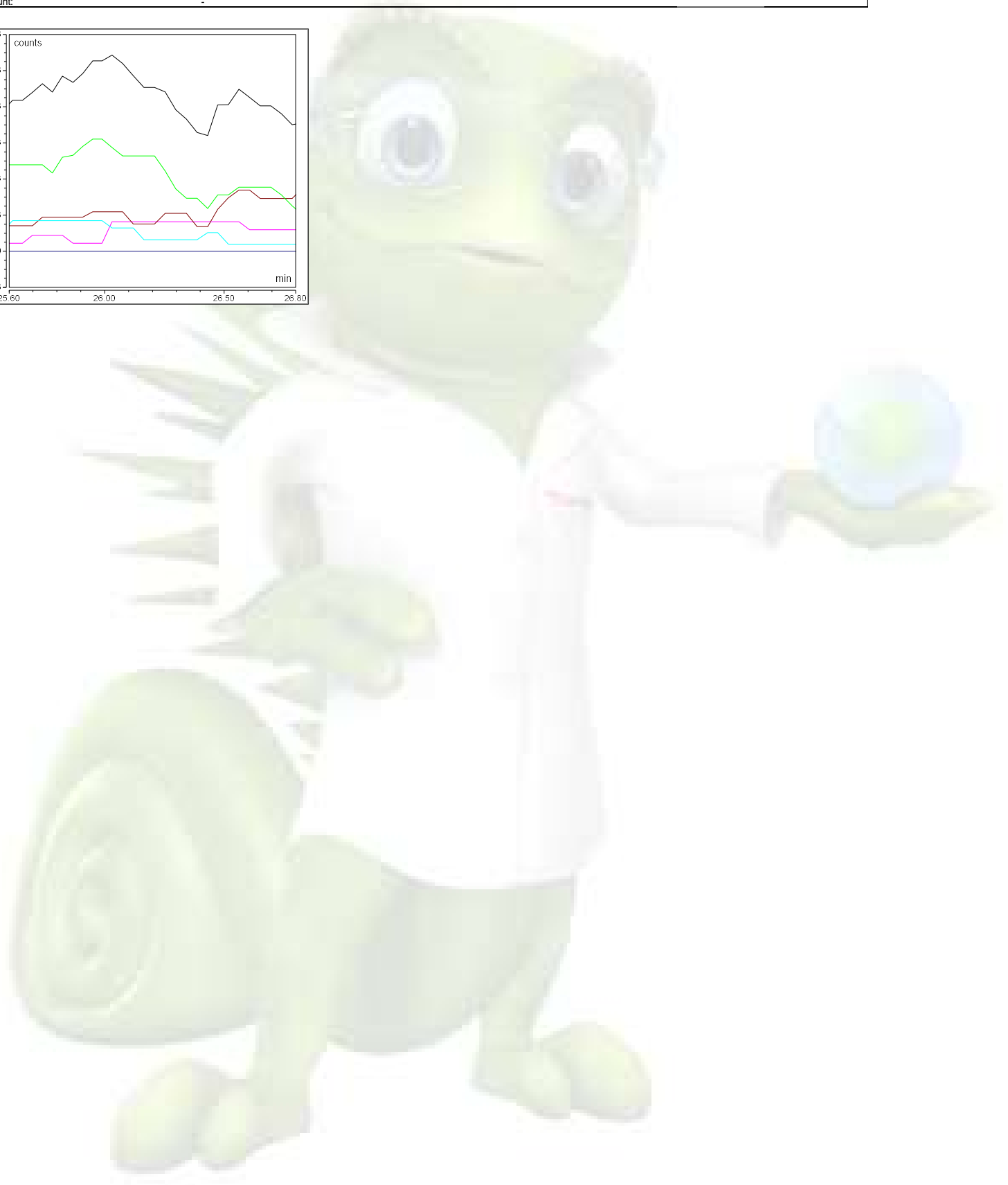
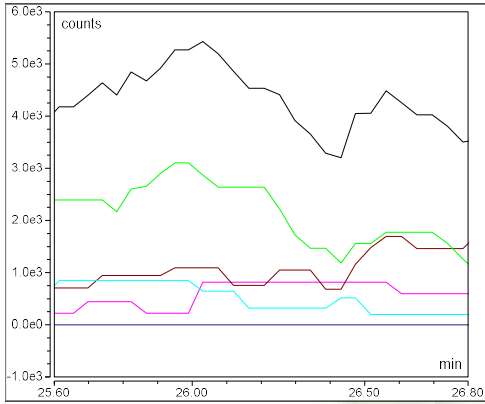
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

Amount: -

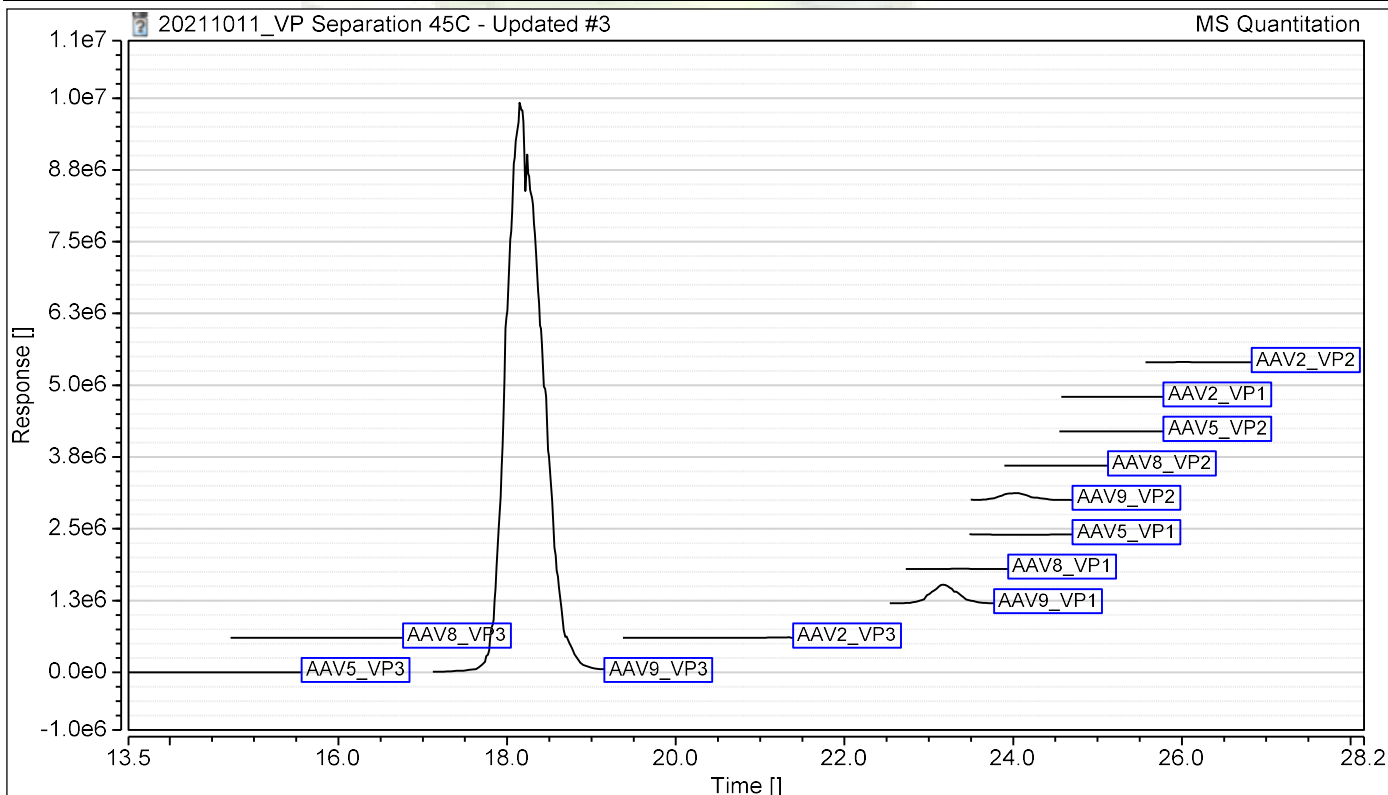


## Chromatogram and SST Results

### Injection Details

|                      |  |                   |        |
|----------------------|--|-------------------|--------|
| Injection Name:      | 20211011_AAV9 E_45C_65-59B_Mod1_1ug_3  | Run Time (min):   | 45.00  |
| Vial Number:         | R:A5   | Injection Volume: | 2.80   |
| Injection Type:      | Unknown  |                   |        |
| Calibration Level:   |  |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                   |        |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 12/Oct/21 16:00  | Sample Weight:    | 1.0000 |

### Chromatogram

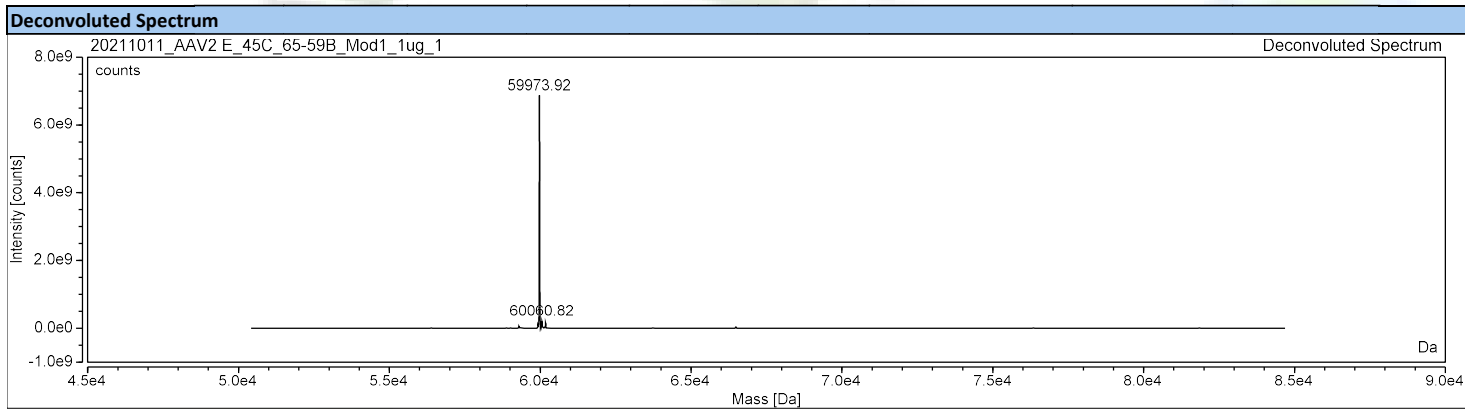
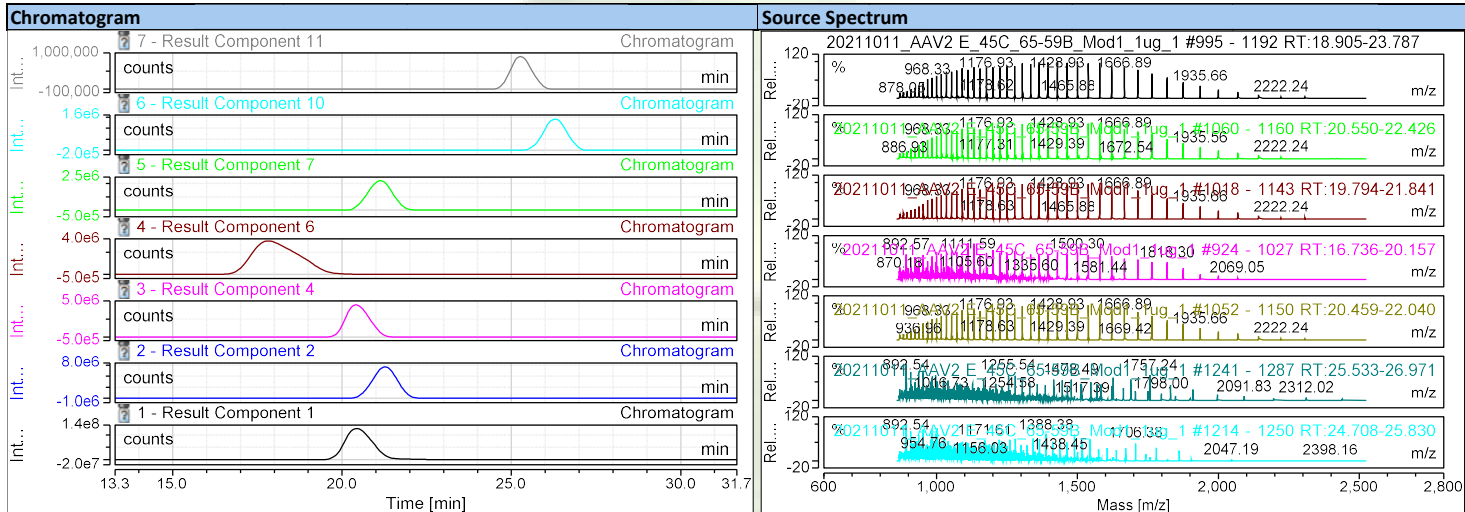


### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |                                       |                            |
|-------------------|---------------------------------------|----------------------------|
| Injection Name:   | 20211011_AAV2 E_45C_65-59B_Mod1_1ug_1 | Injection Volume:          |
| Vial Number:      | R:A1                                  | Instrument Method:         |
| Injection Type:   | Unknown                               | Processing Method:         |
| Run Time (min):   | 45.00                                 | Injection Date/Time:       |
|                   |                                       | MS Quantitative_45C_65-59B |
|                   |                                       | 12/Oct/21 21:57            |



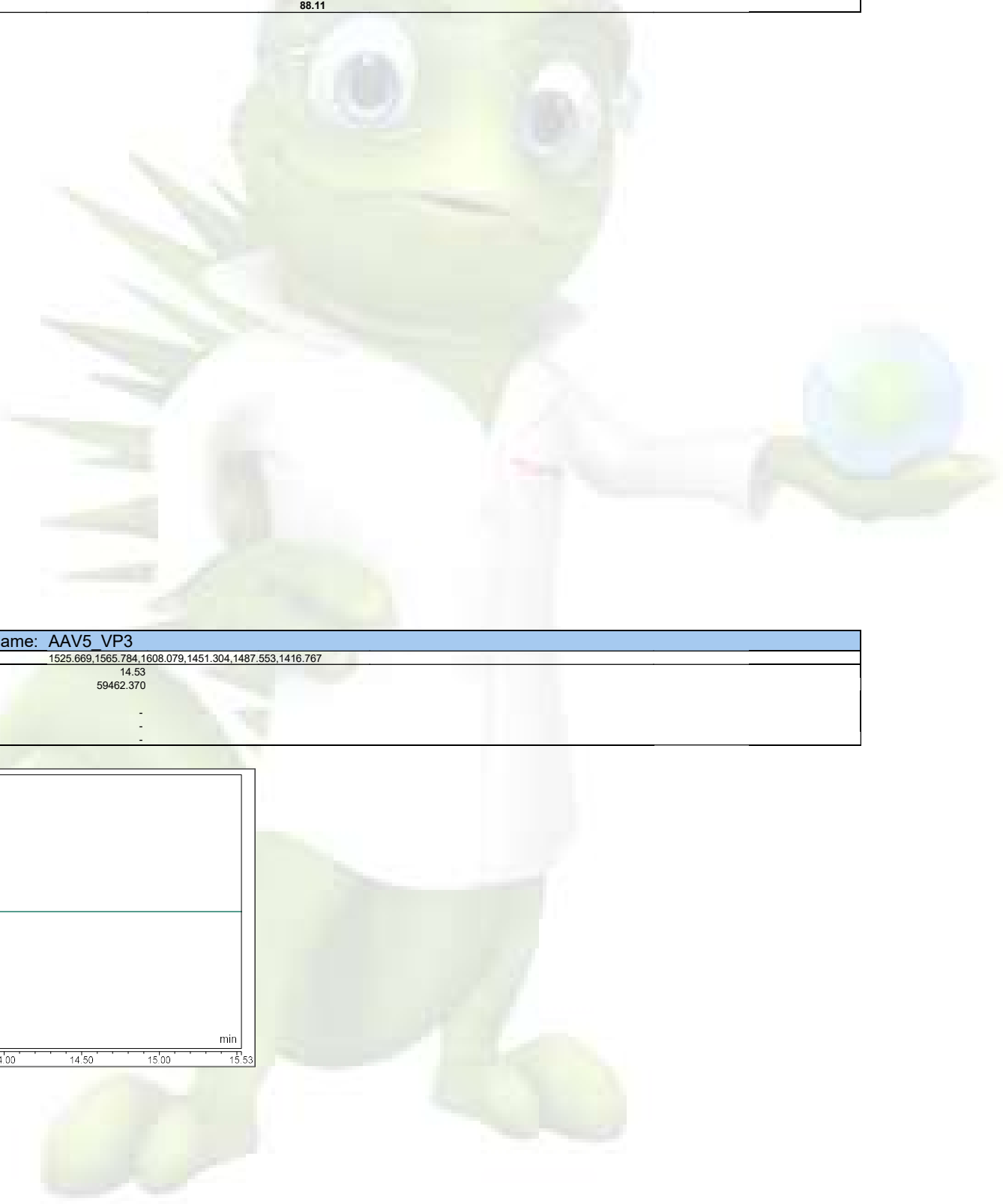
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

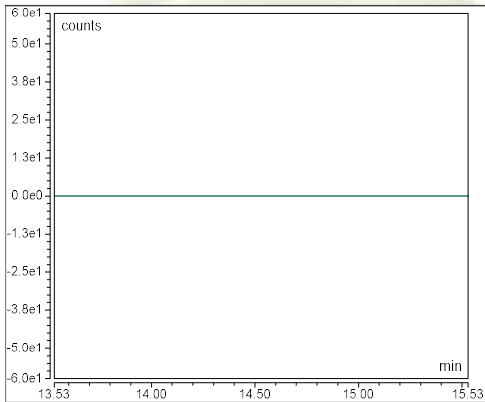
**Injection Details**

|   |  |  |
|---|--|--|
| Injection Name: 20211011_AAV2_E_45C_65-59B_Mod1_1ug | Injection Volume: 2.86   |  |
| Vial Number: R:A1                                   | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| Injection Type: Unknown                             | Processing Method: MS Quantitative_45C_65-59B                            |  |
| Run Time (min): 45.00                               | Injection Date/Time: 12/Oct/21 21:57                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3     | 20.493                | 1423308.517        | 86.33              | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 | 59973.780 | PASS                           |
| AAV2_VP1     | 25.313                | 13464.491          | 0.82               | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 | 81854.900 | PASS                           |
| AAV2_VP2     | 26.236                | 15926.651          | 0.97               | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 | 66487.900 | PASS                           |
| <b>88.11</b> |                       |                    |                    |   |           |                                |


**Component Name: AAV5\_VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| Retention time (min): | 14.53   |
| Expected Mass:        | 59462.370   |
| Area (count*min):     | -   |
| Intensity (counts):   | -   |
| Amount:               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

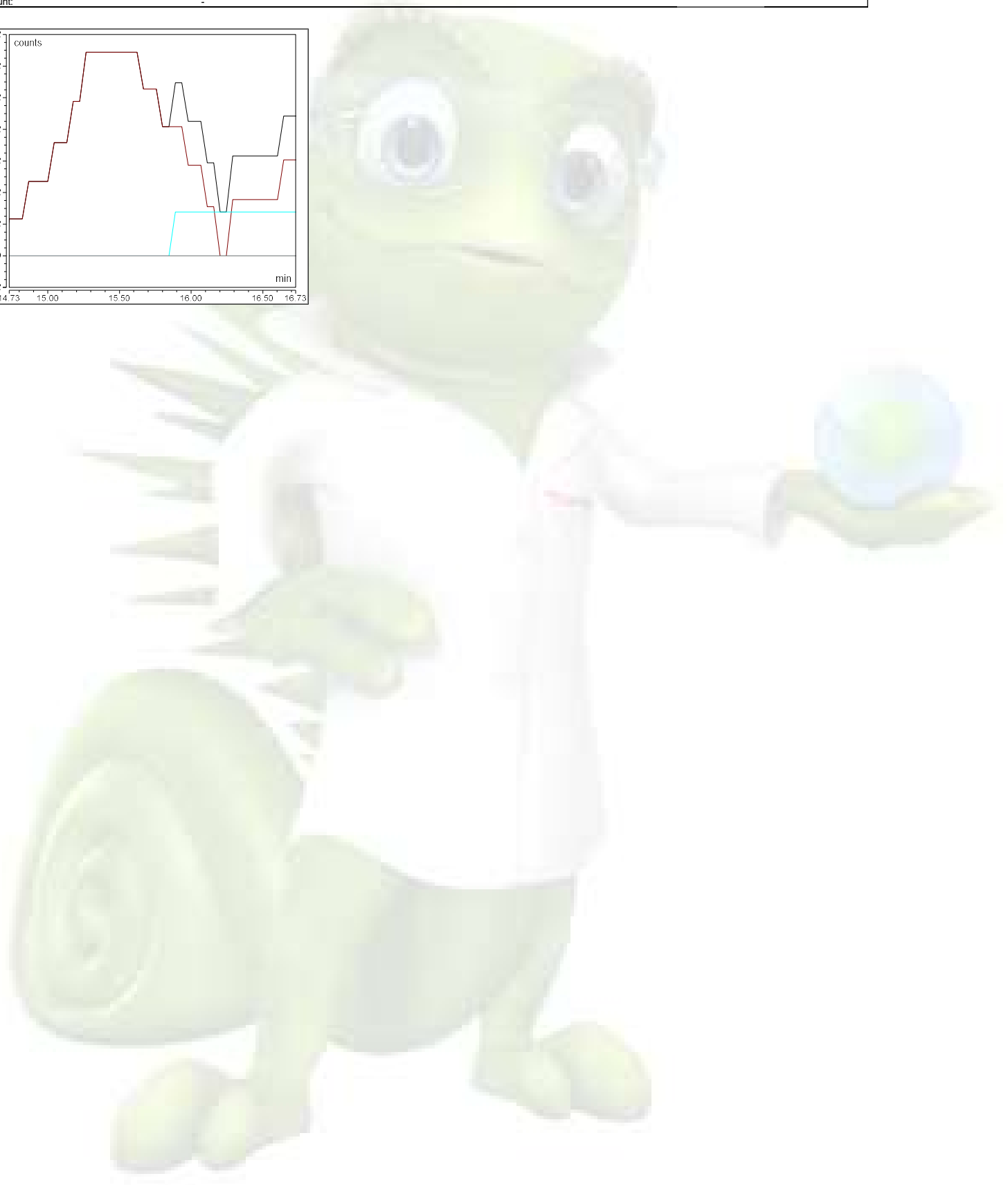
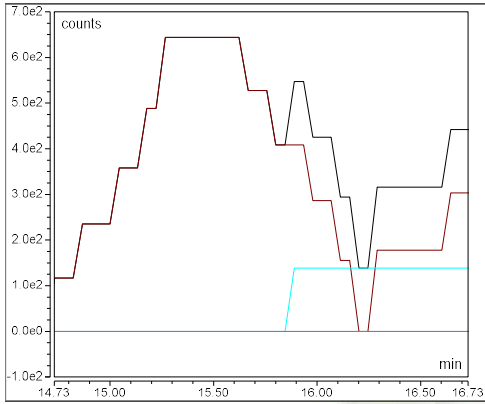
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

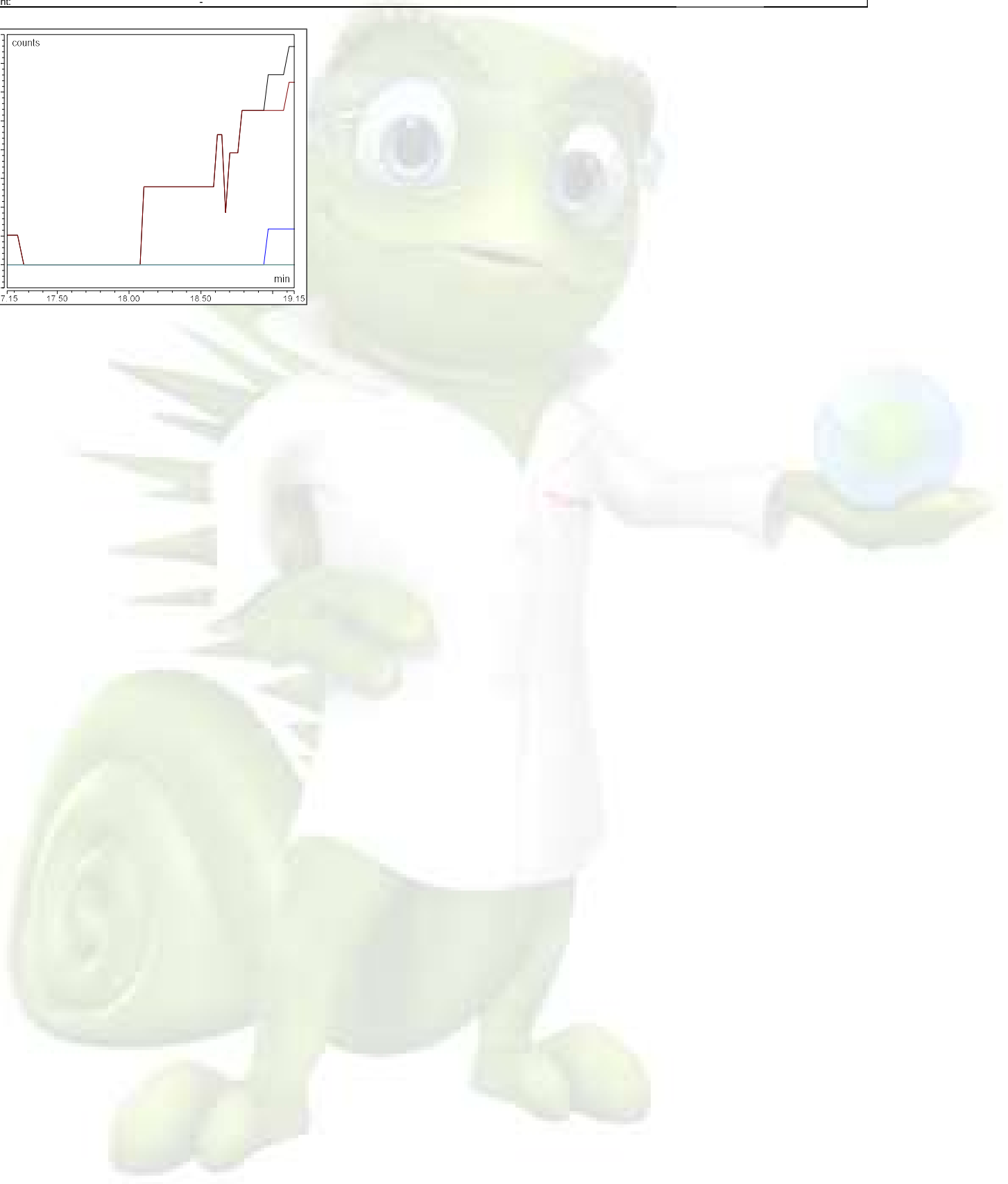
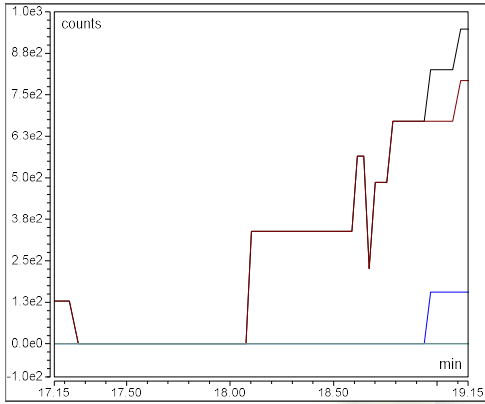
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

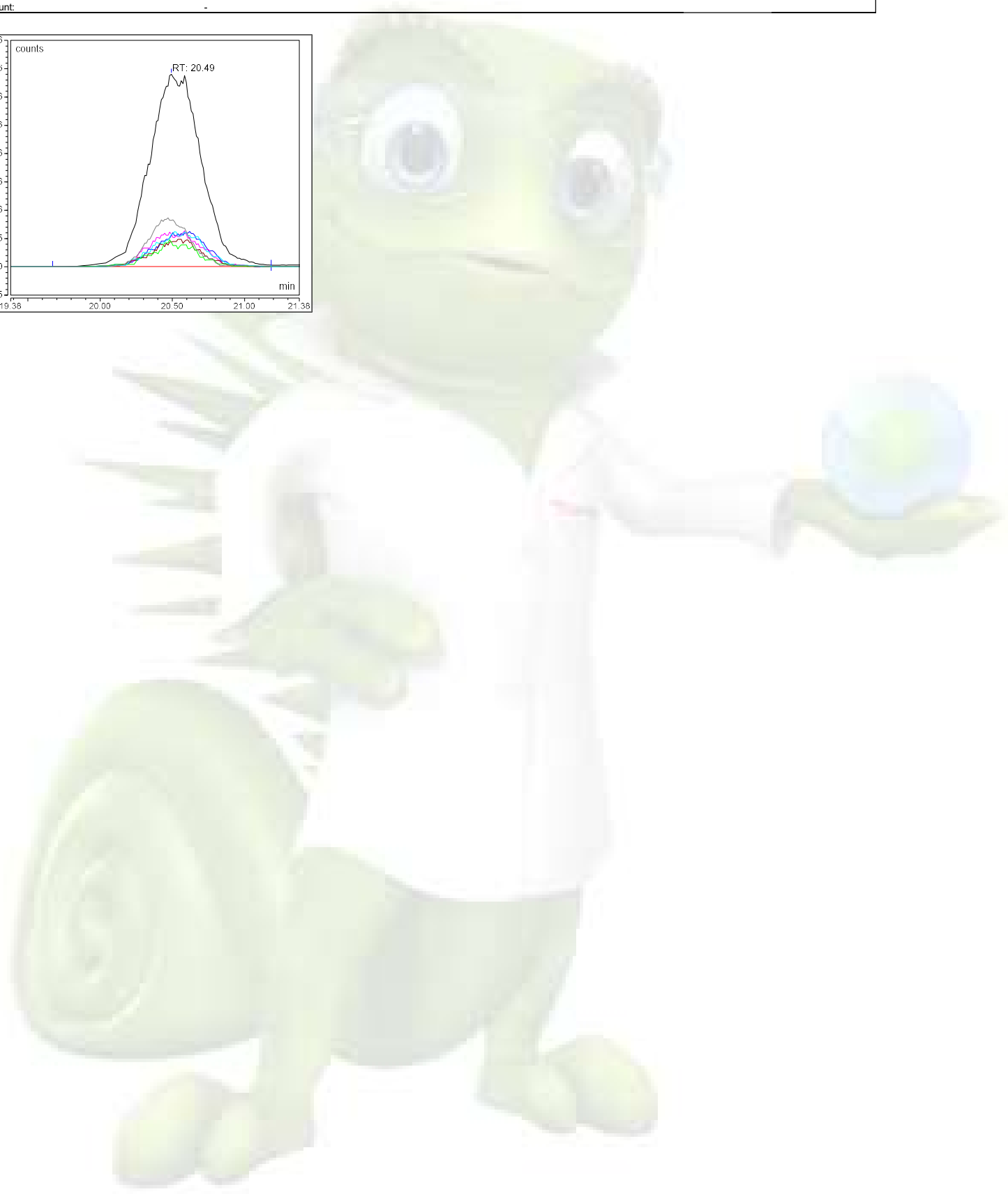
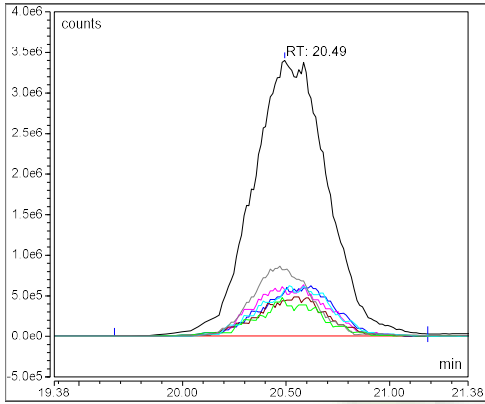
Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 |
| Retention time (min): | 20.38   |
| Expected Mass:        | 59973.780   |
| Area (count*min):     | 1423309   |
| Intensity (counts):   | 3398310   |
| Amount:               | -   |



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

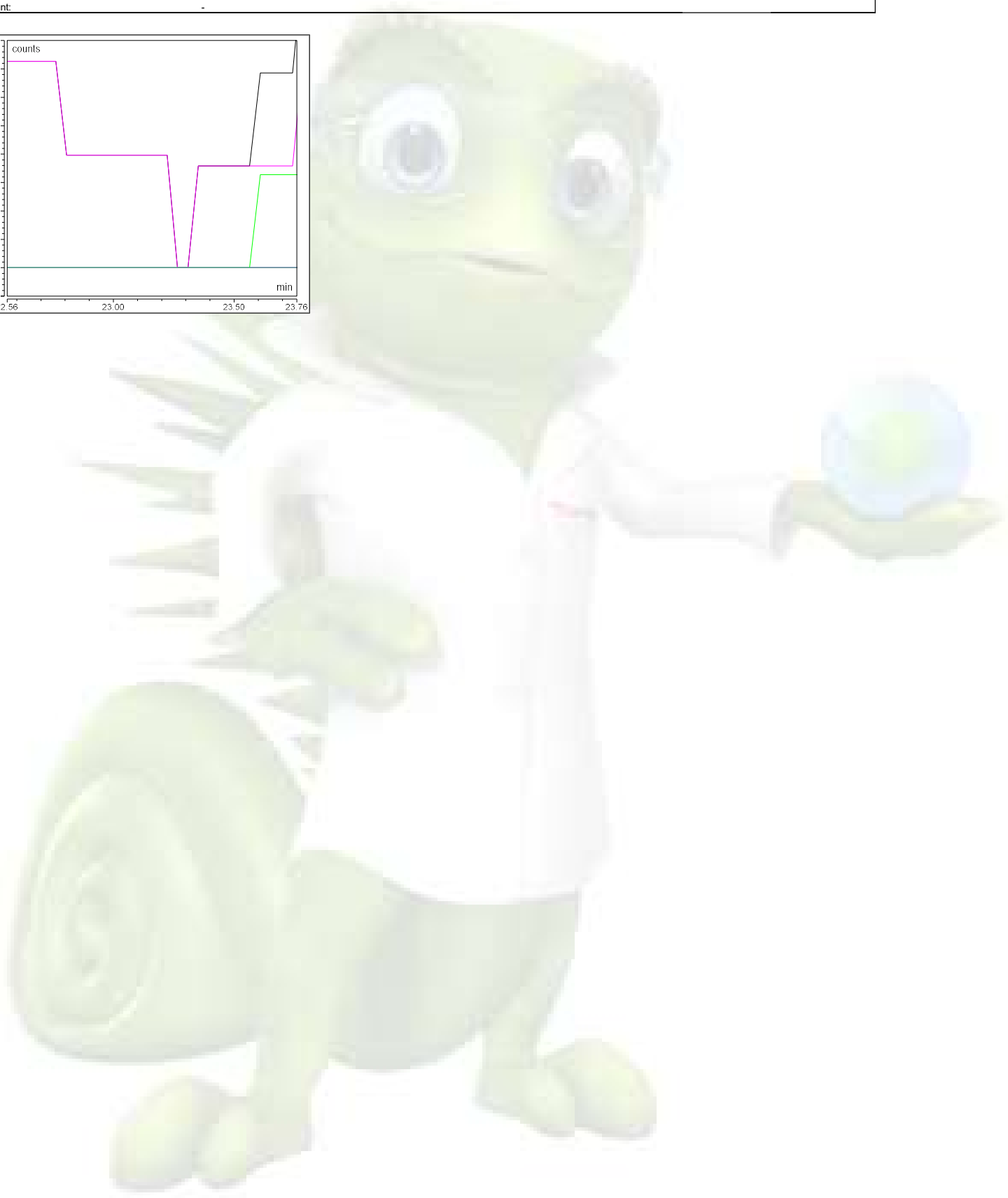
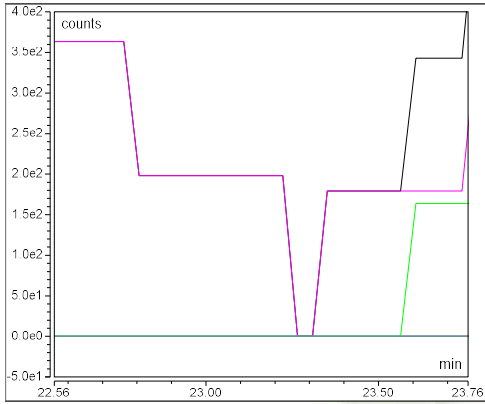
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

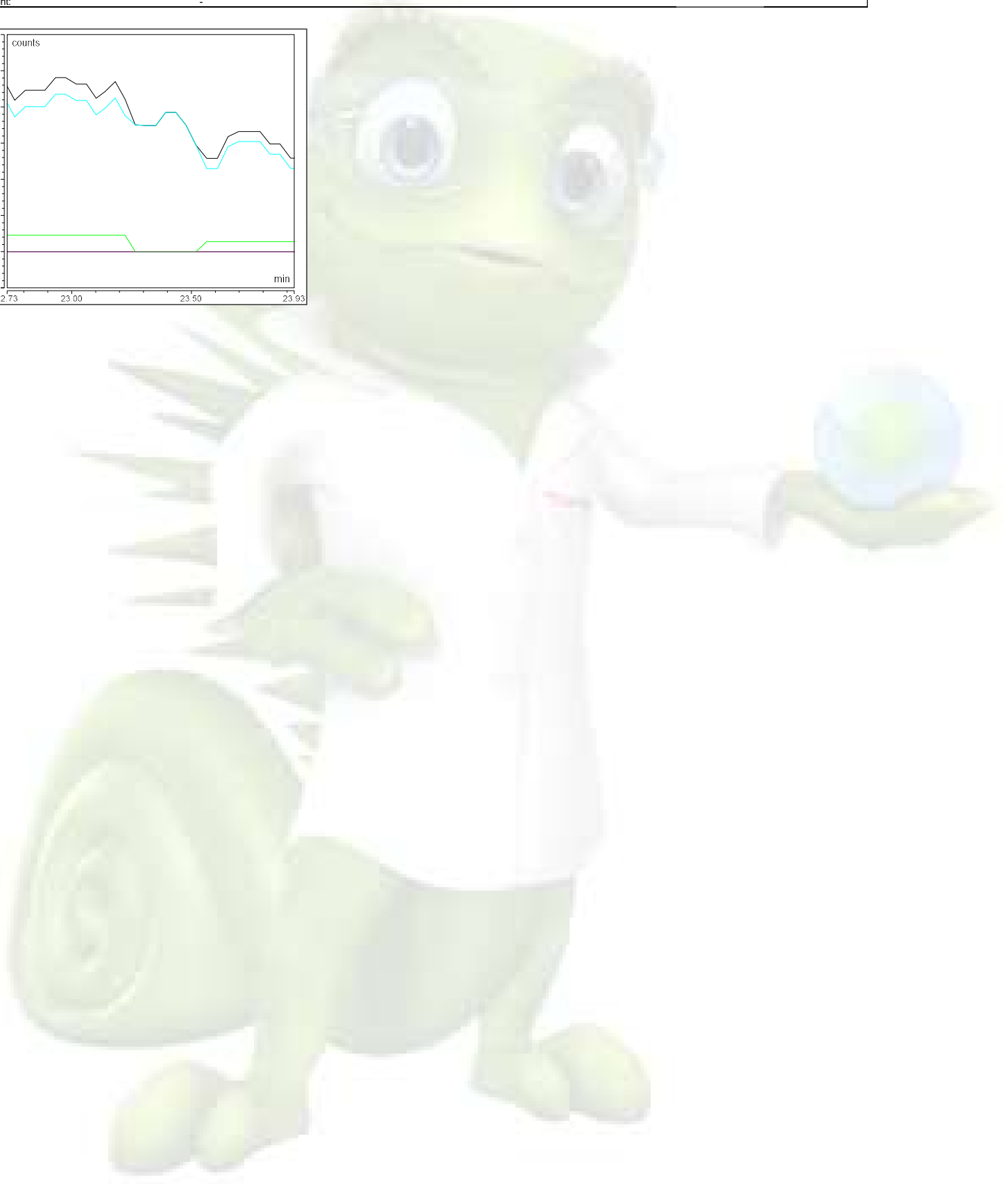
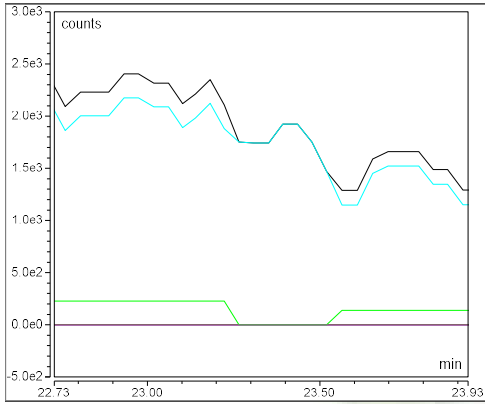
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

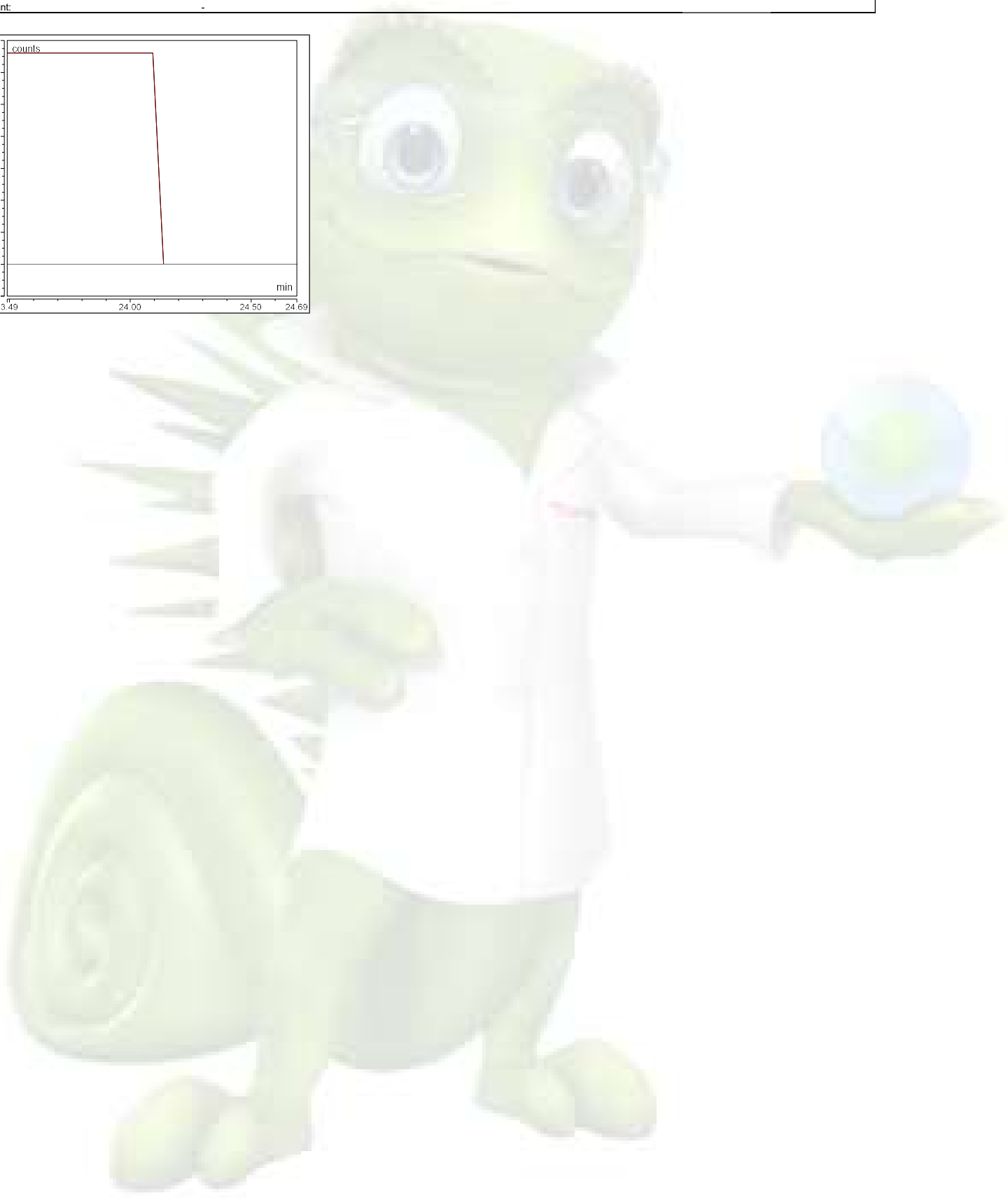
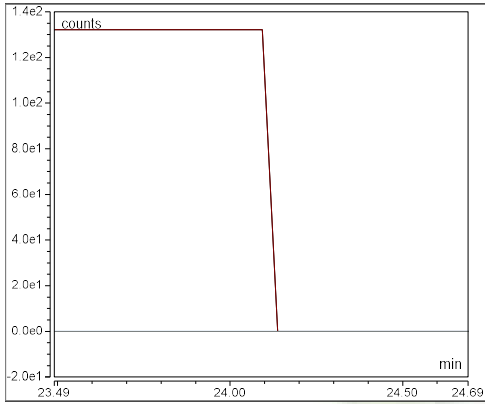
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -

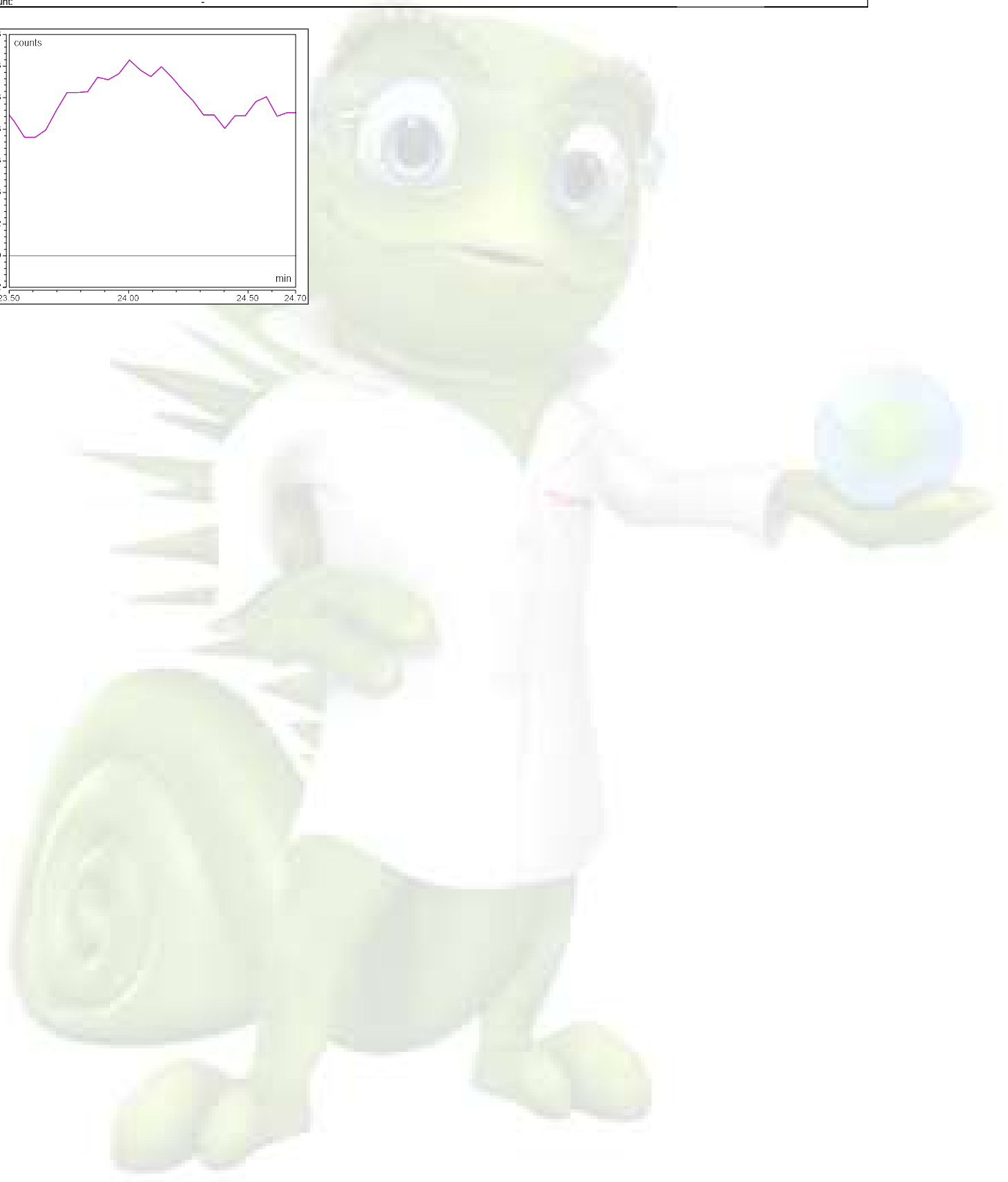
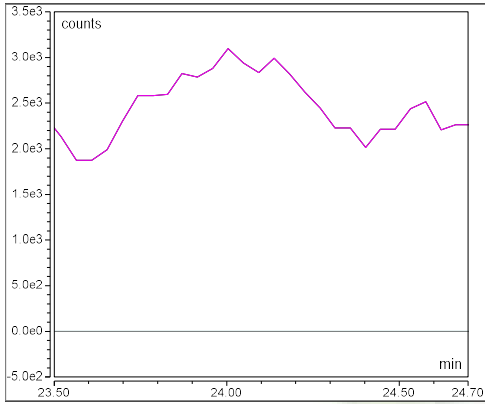


**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

Retention time (min): 24.10  
Expected Mass: 66209.760

Area (count\*min): -  
Intensity (counts): -  
Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

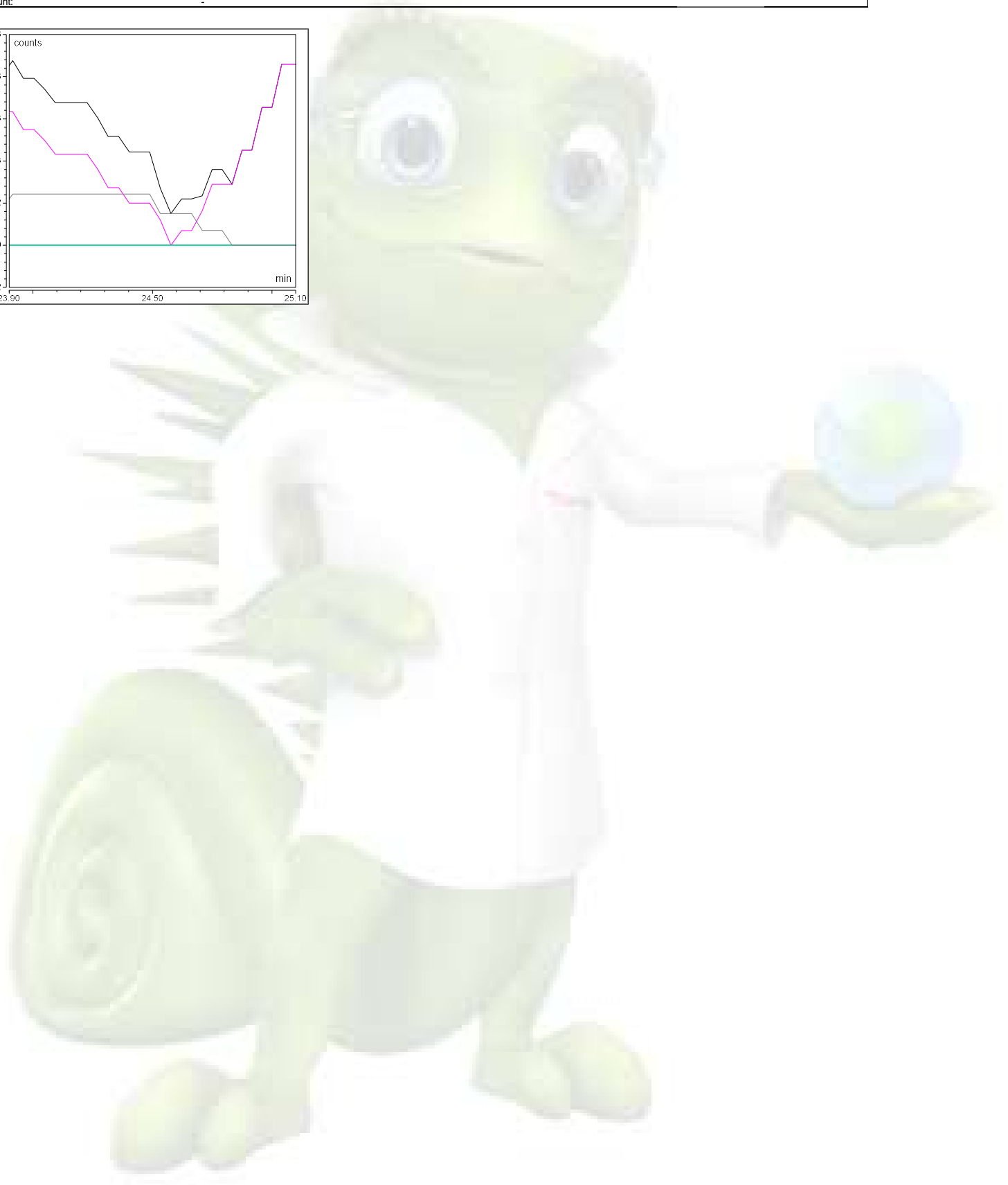
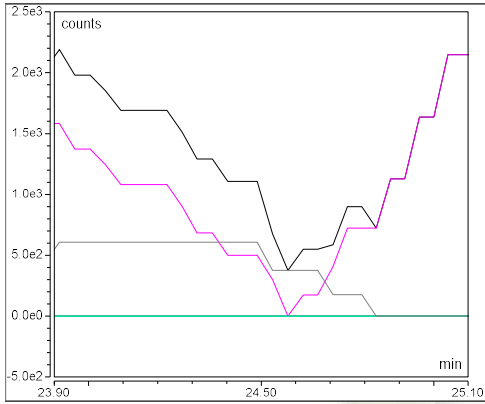
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

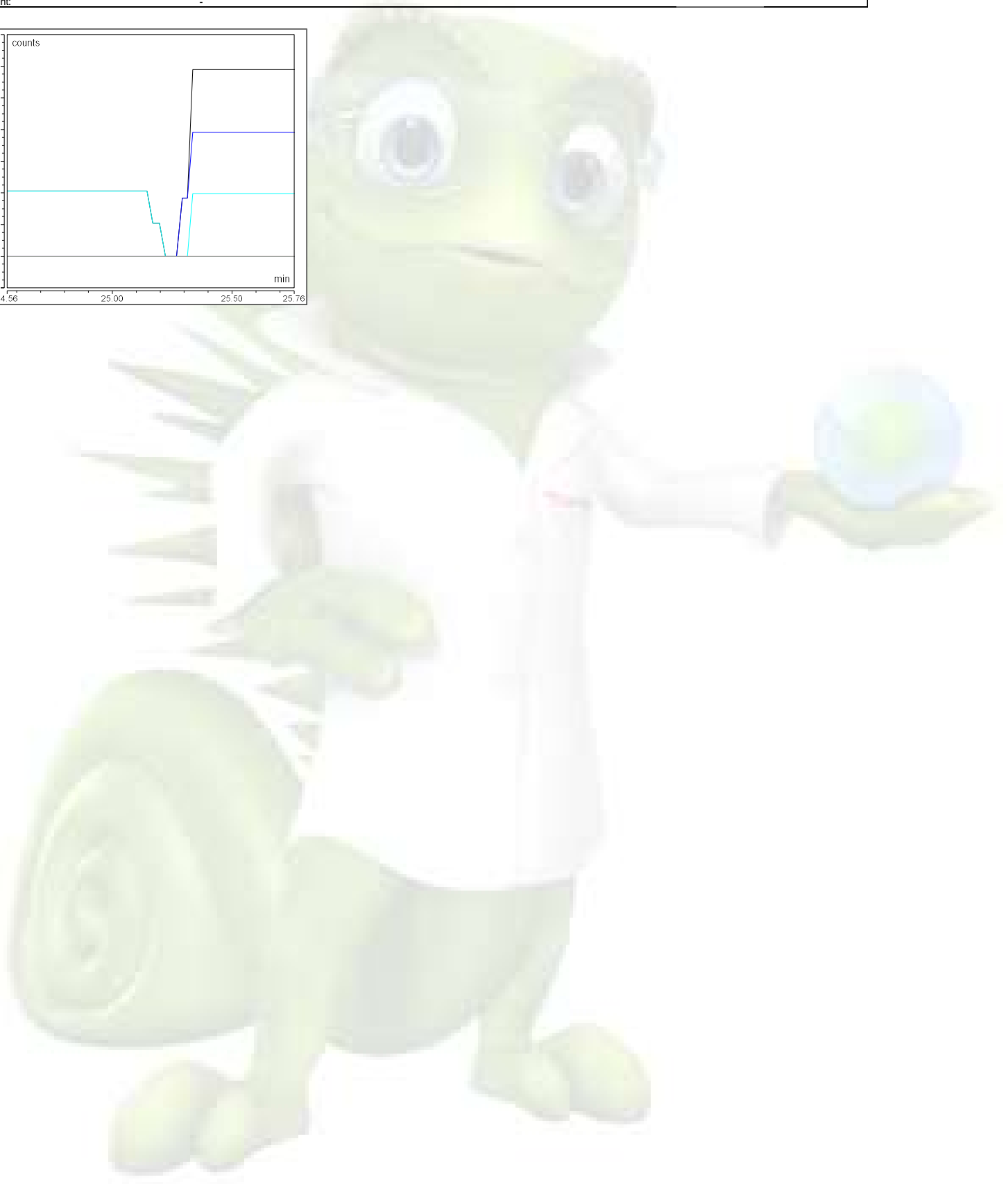
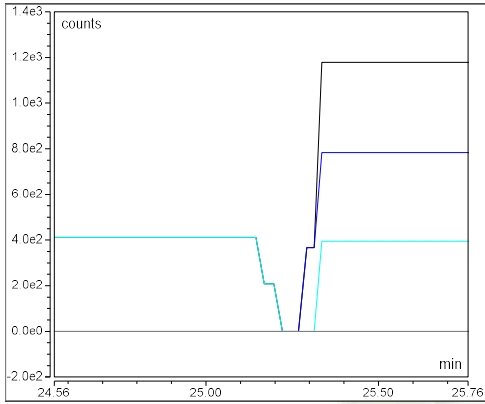
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

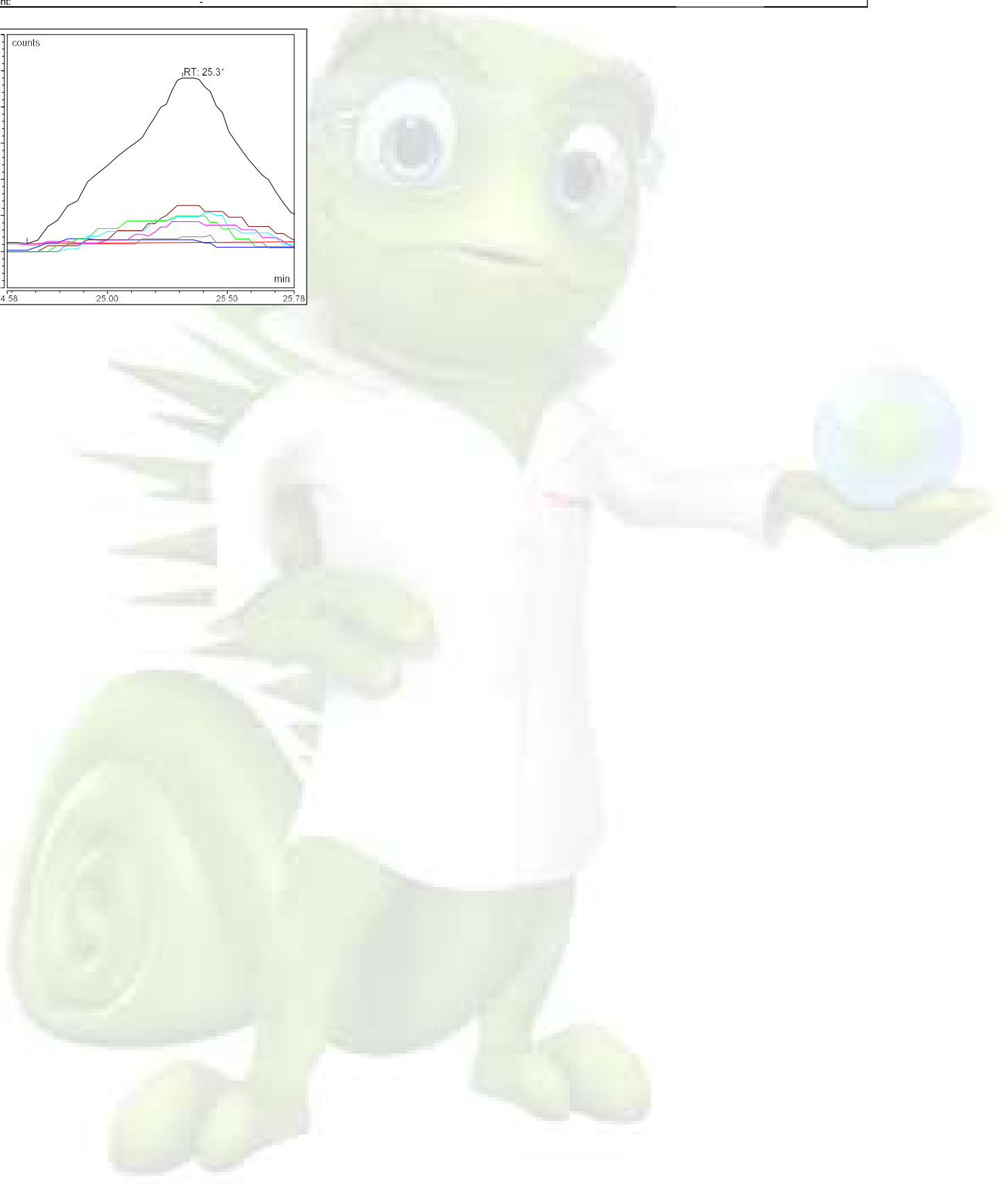
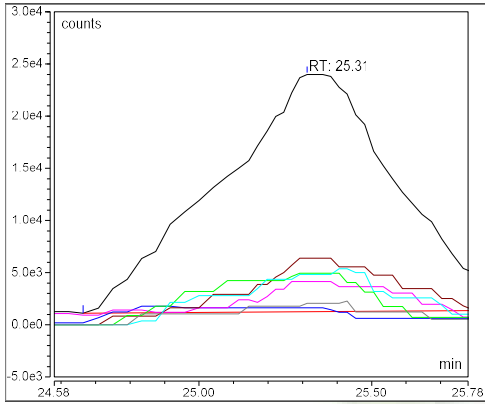
Intensity (counts): -

Amount: -



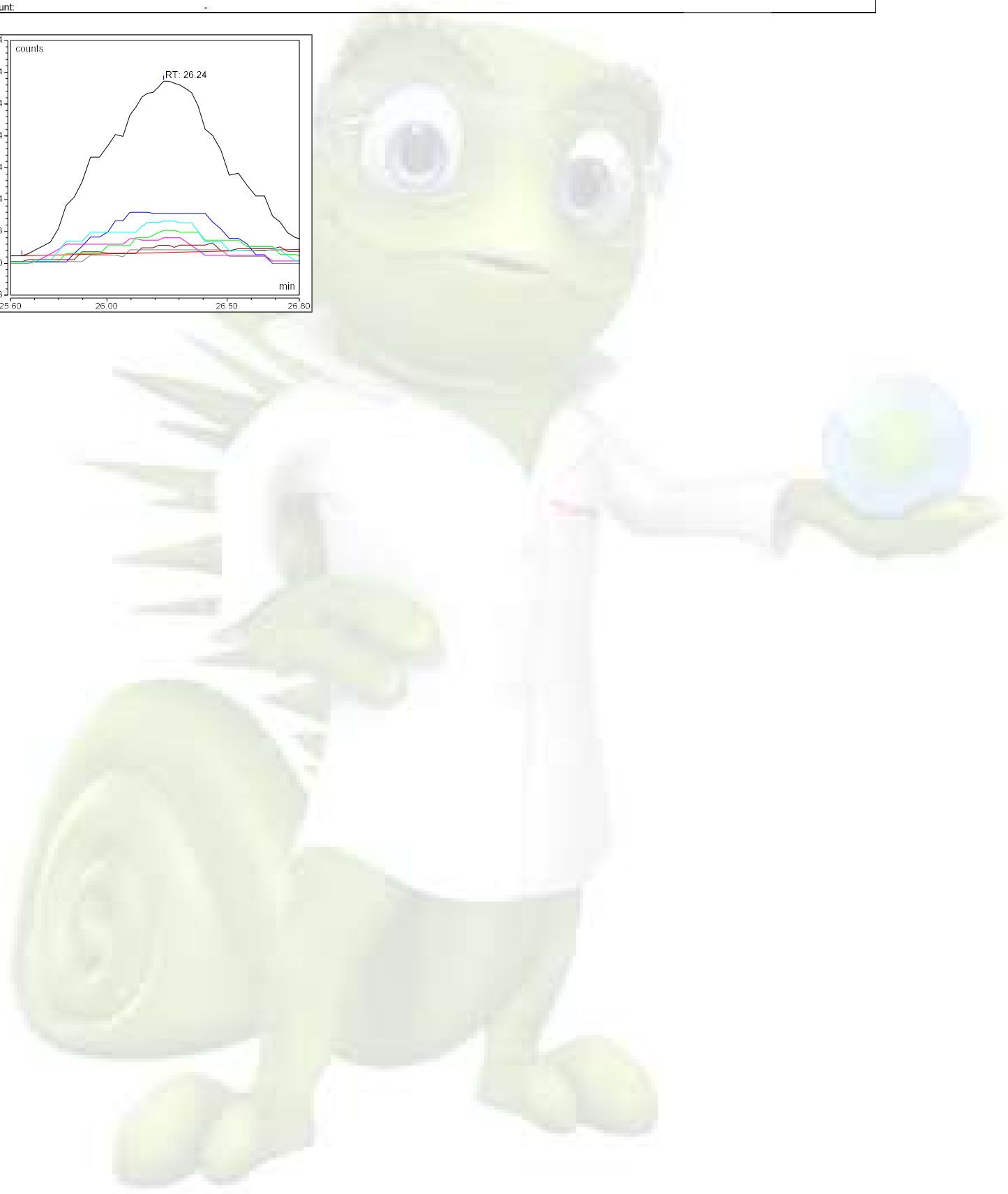
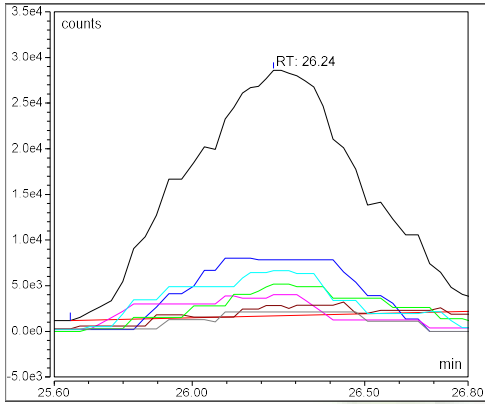
**Component Name: AAV2 VP1**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 |
| Retention time (min): | 25.18   |
| Expected Mass:        | 81854.900   |
| Area (count*min):     | 13464   |
| Intensity (counts):   | 22733   |
| Amount:               | -   |



**Component Name: AAV2 VP2**

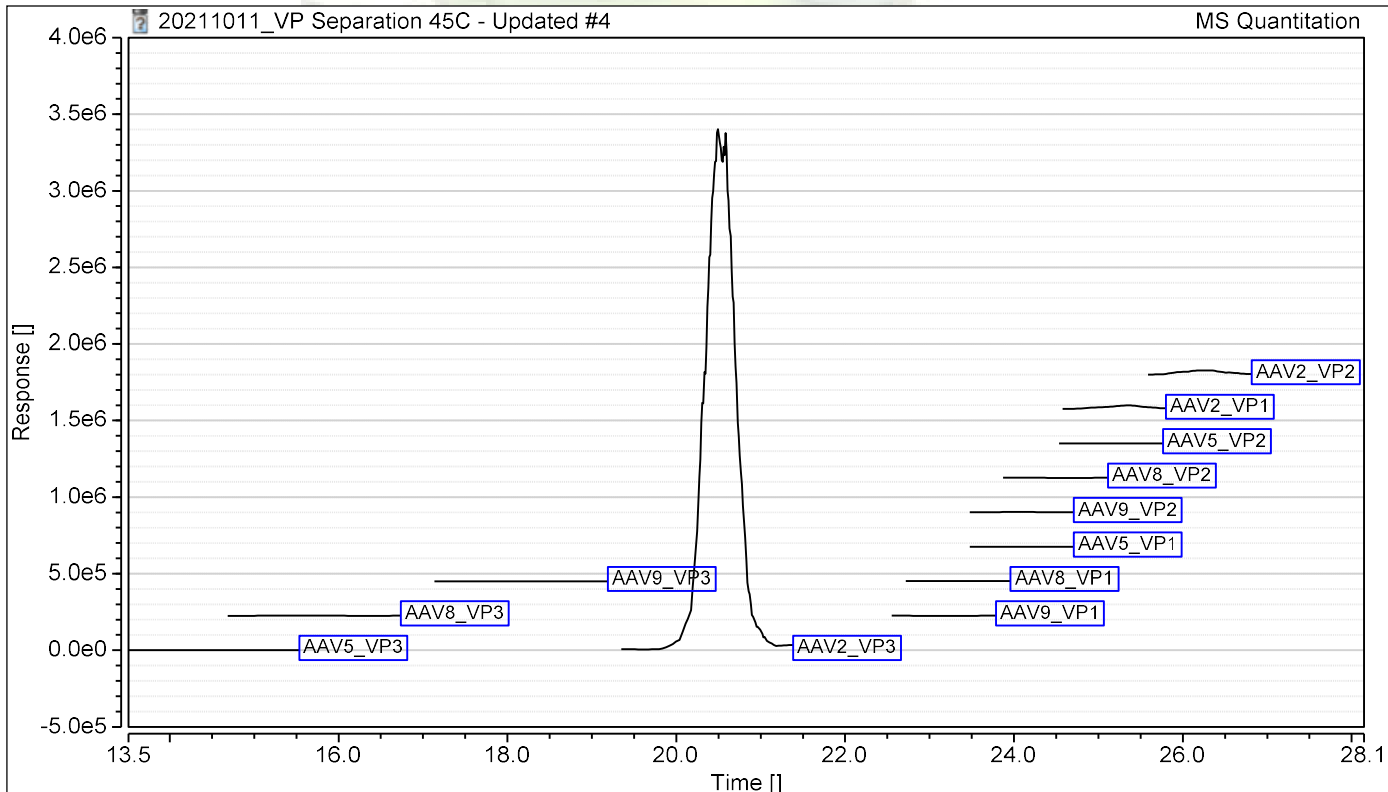
|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 |
| Retention time (min): | 26.20   |
| Expected Mass:        | 66487.900   |
| Area (count*min):     | 15927   |
| Intensity (counts):   | 26899   |
| Amount:               | -   |



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV2 E_45C_65-59B_Mod1_1ug_1                                      | Run Time (min): 45.00   |
| Vial Number:         | R:A1   | Injection Volume: 2.86  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 | S                       |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 12/Oct/21 21:57  | Sample Weight: 1.0000   |

### Chromatogram

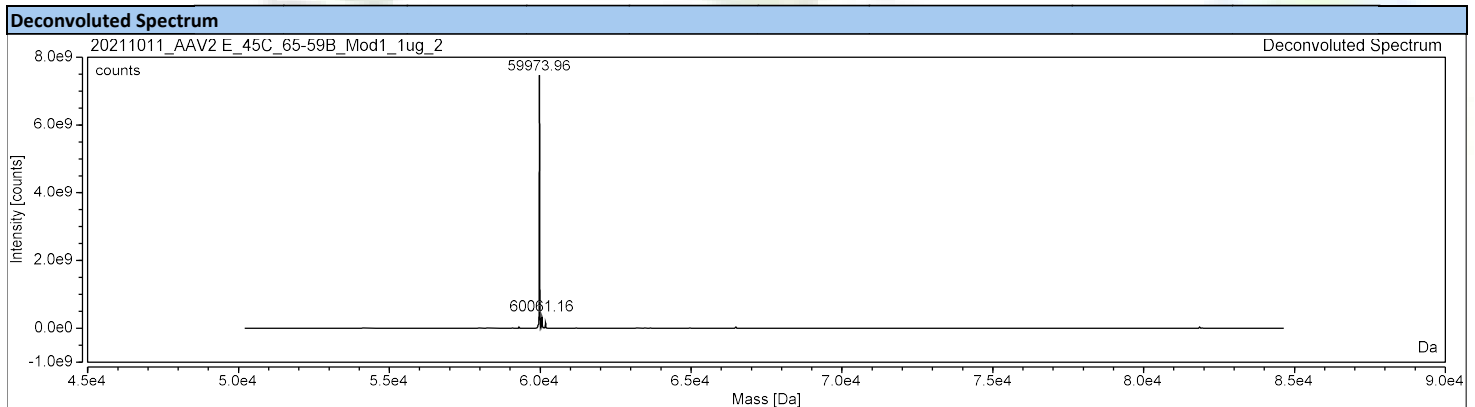
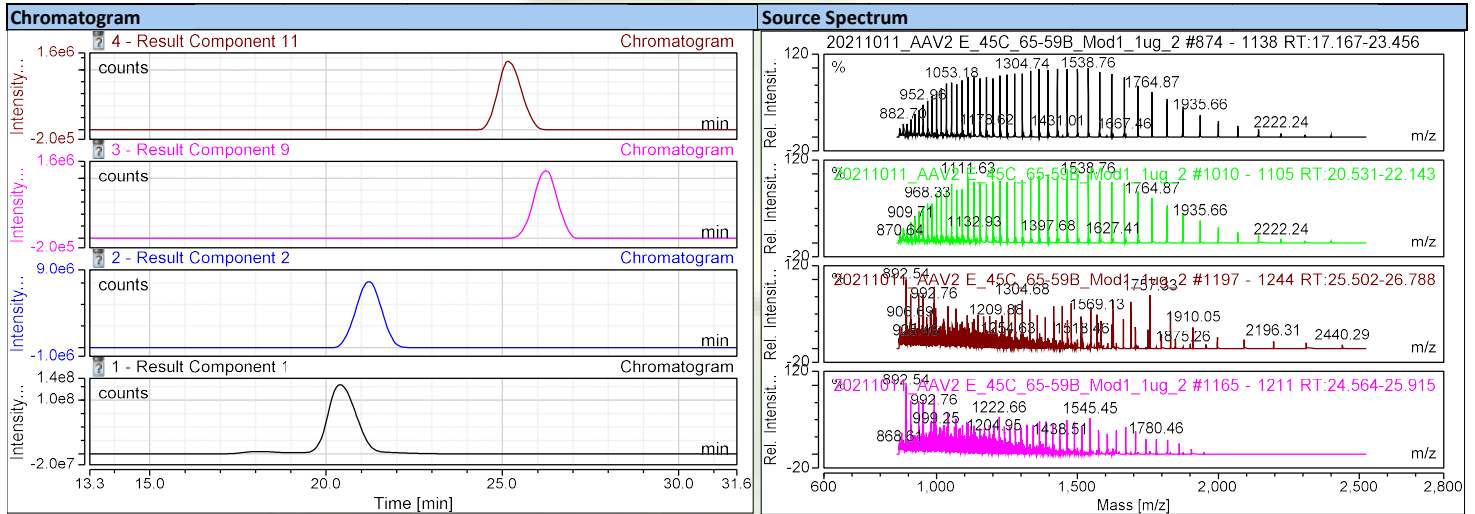


| SST Results                         |      |               |               |             |           |
|-------------------------------------|------|---------------|---------------|-------------|-----------|
| No.                                 | Name | Inj.Condition | Peak          | Test Result | Injection |
| Number of executed test cases: n.a. |      |               | Total Result: | Passed      |           |



## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV2 E_45C_65-59B_Mod1_1ug_2 | Injection Volume: 2.86                                       |
| Vial Number:      | R:A1                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 44.99                                 | Injection Date/Time: 12/Oct/21 22:43                         |



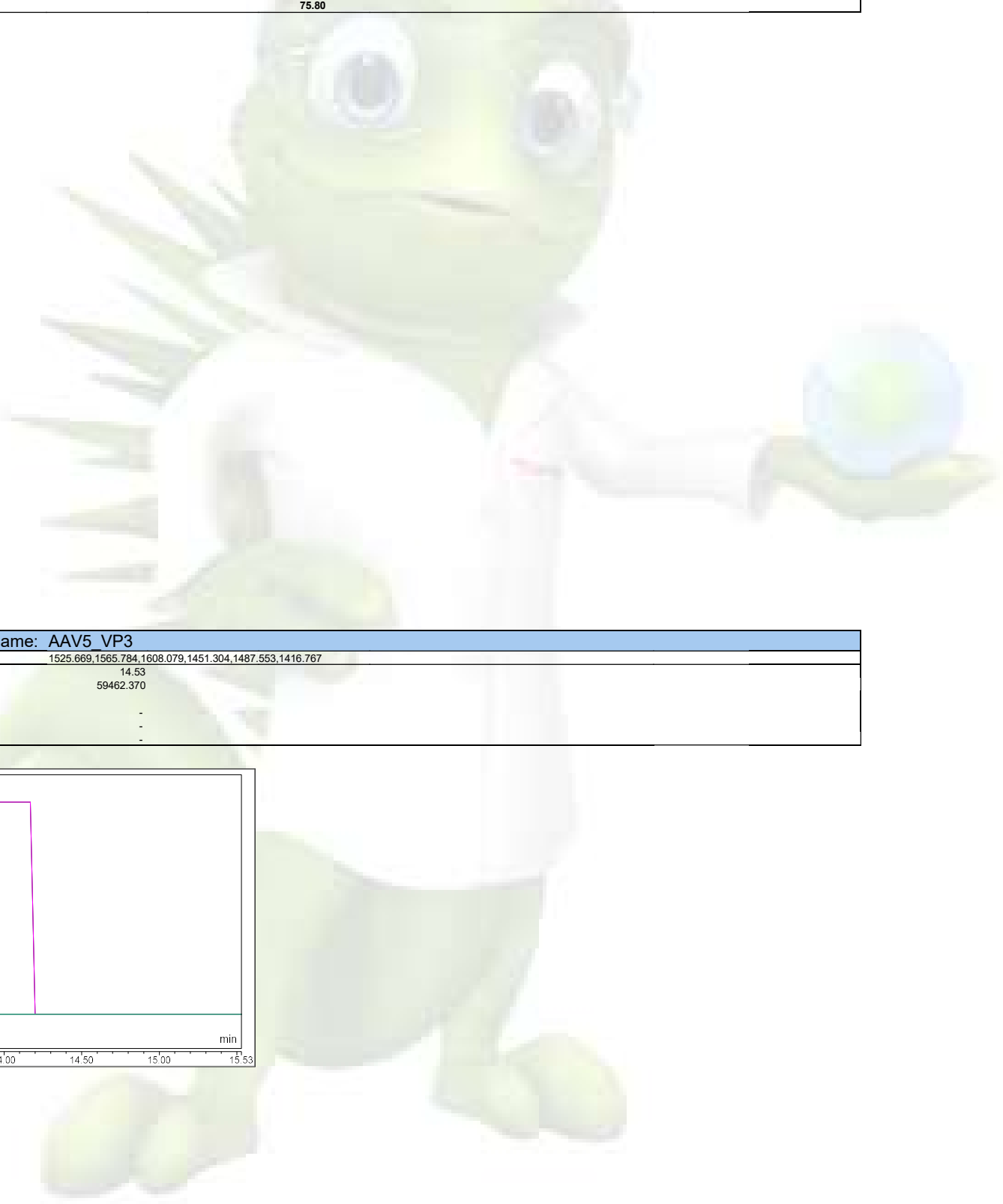
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

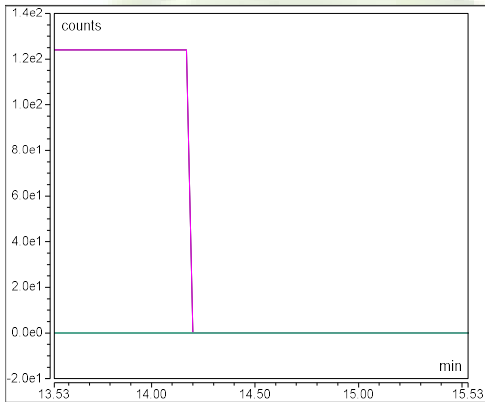
|  |   |  |
|--|---|--|
| <i>Injection Name:</i> 20211011_AAV2_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i> 2.86   |  |
| <i>Vial Number:</i> R:A1                                   | <i>Instrument Method:</i> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <i>Injection Type:</i> Unknown                             | <i>Processing Method:</i> MS Quantitative_45C_65-59B                            |  |
| <i>Run Time (min):</i> 44.99                               | <i>Injection Date/Time:</i> 12/Oct/21 22:43                                     |  |

| Peak Name | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|-----------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3  | 20.463                | 1500924.040        | 74.26              | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 | 59973.780 | PASS                           |
| AAV2_VP1  | 25.294                | 10480.860          | 0.52               | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 | 81854.900 | PASS                           |
| AAV2_VP2  | 26.228                | 20760.822          | 1.03               | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 | 66487.900 | PASS                           |
|           |                       |                    | <b>75.80</b>       |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

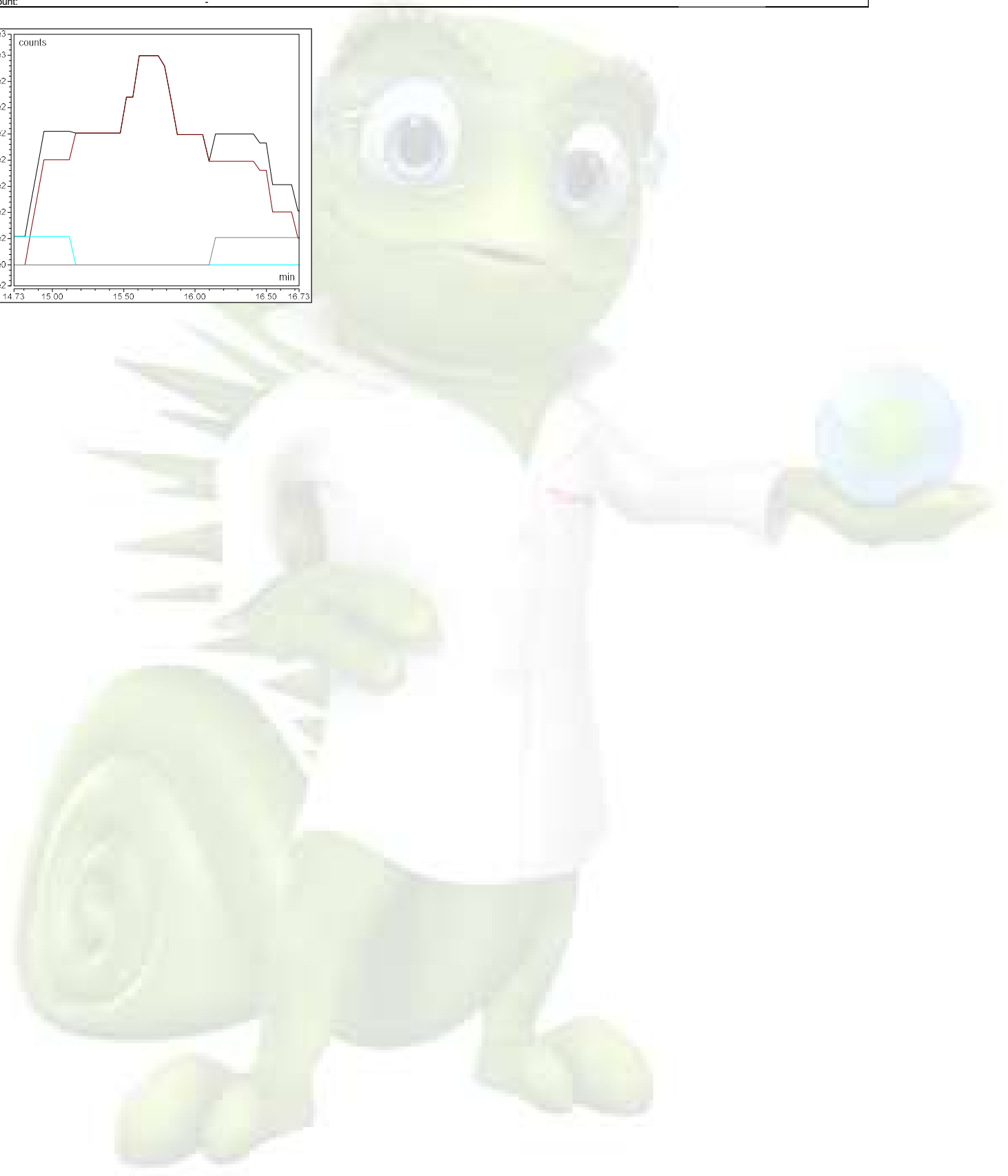
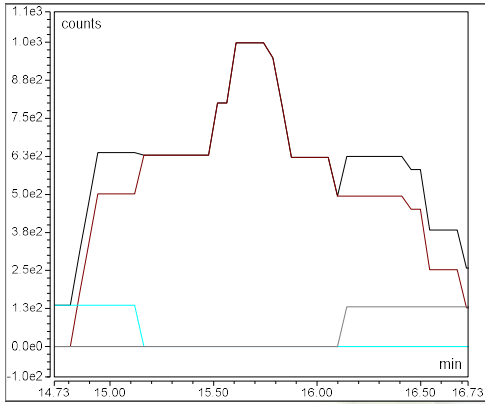
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

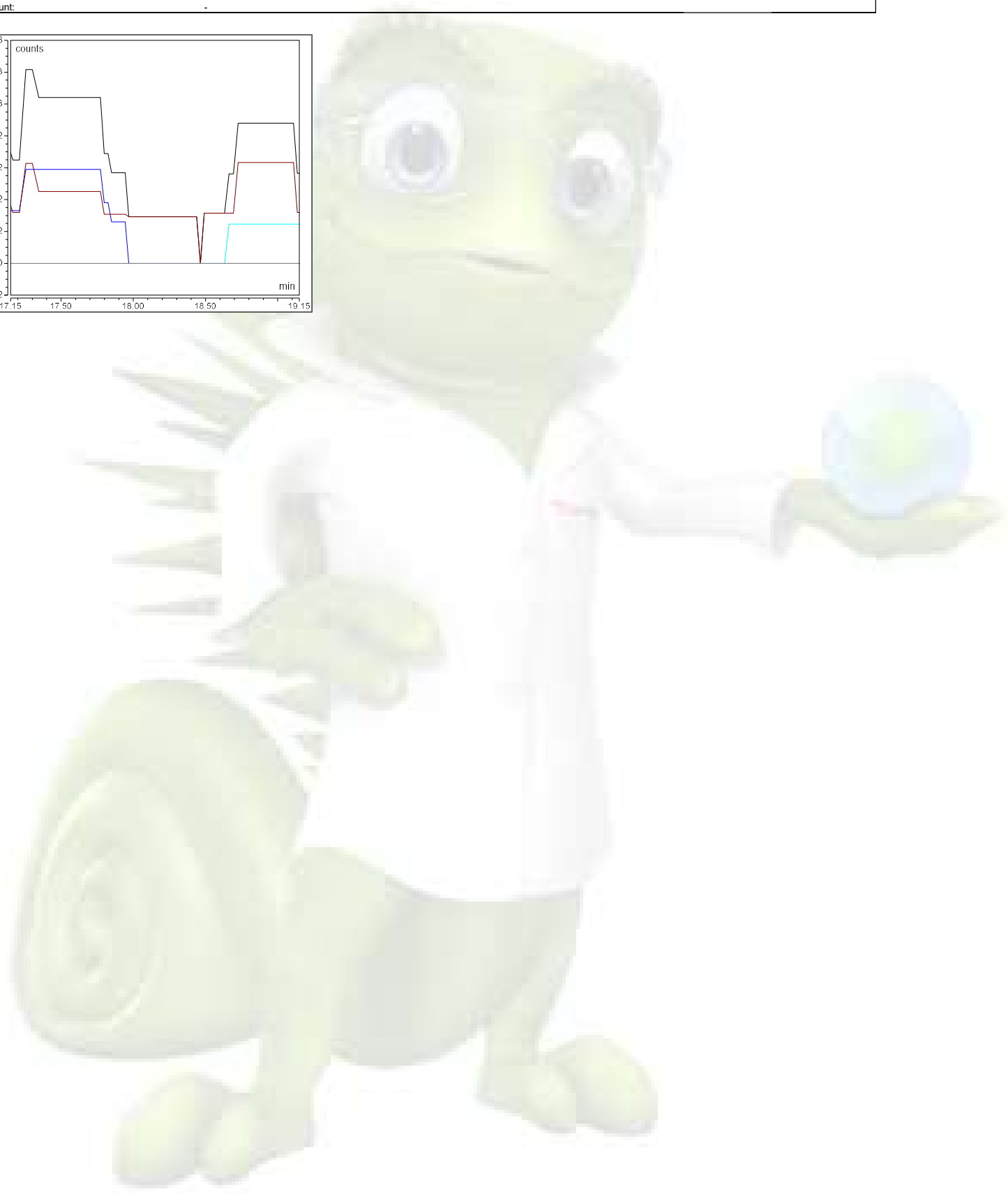
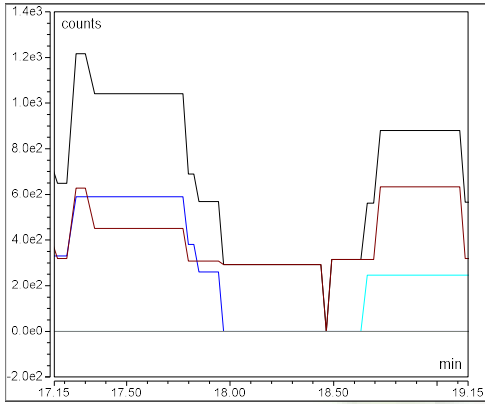
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

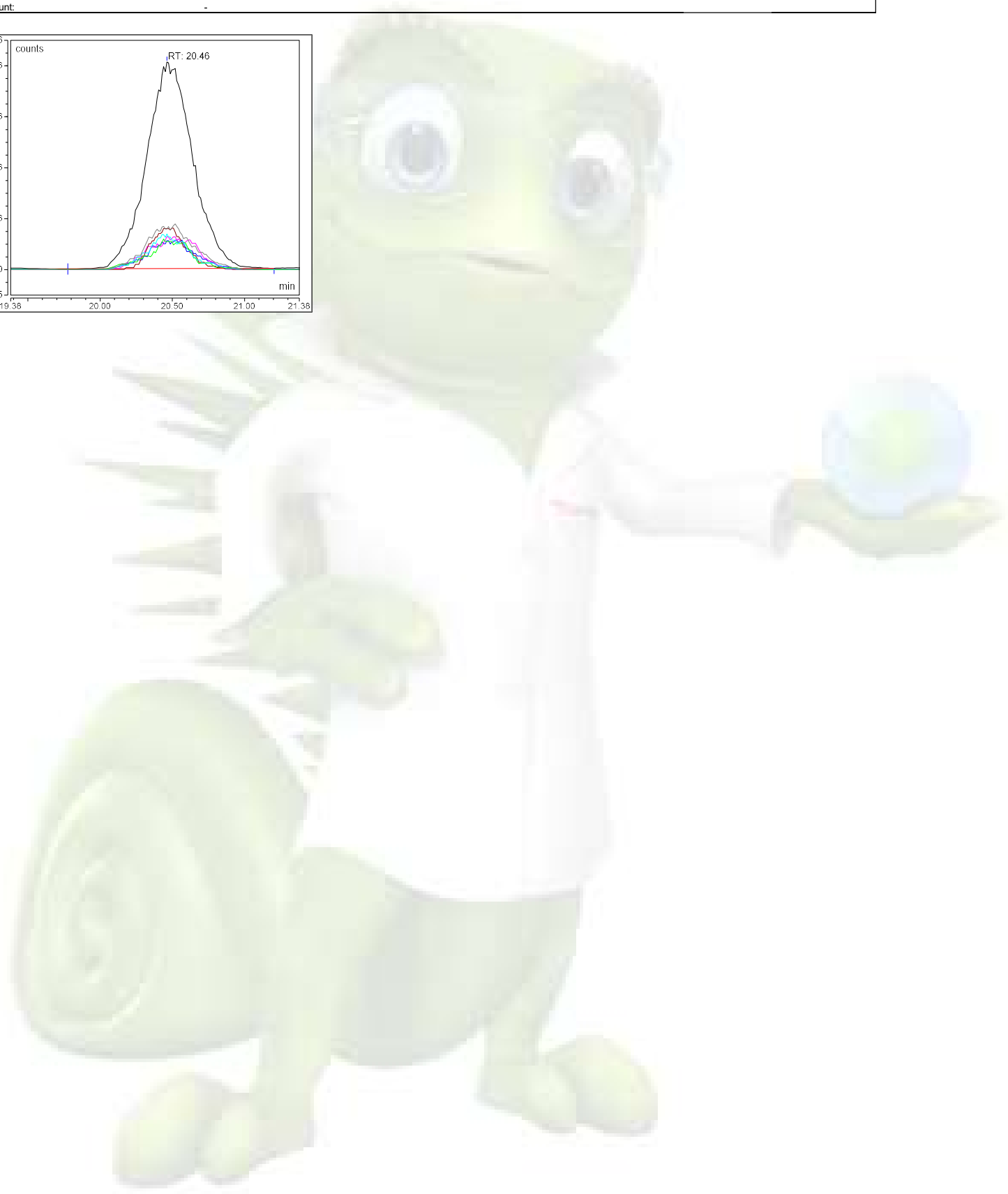
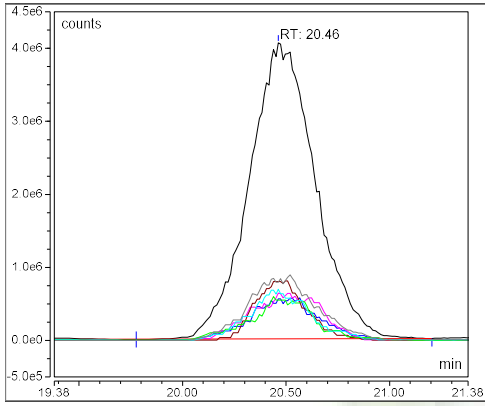
Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 |
| Retention time (min): | 20.38   |
| Expected Mass:        | 59973.780   |
| Area (count*min):     | 1500924   |
| Intensity (counts):   | 4051828   |
| Amount:               | -   |



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

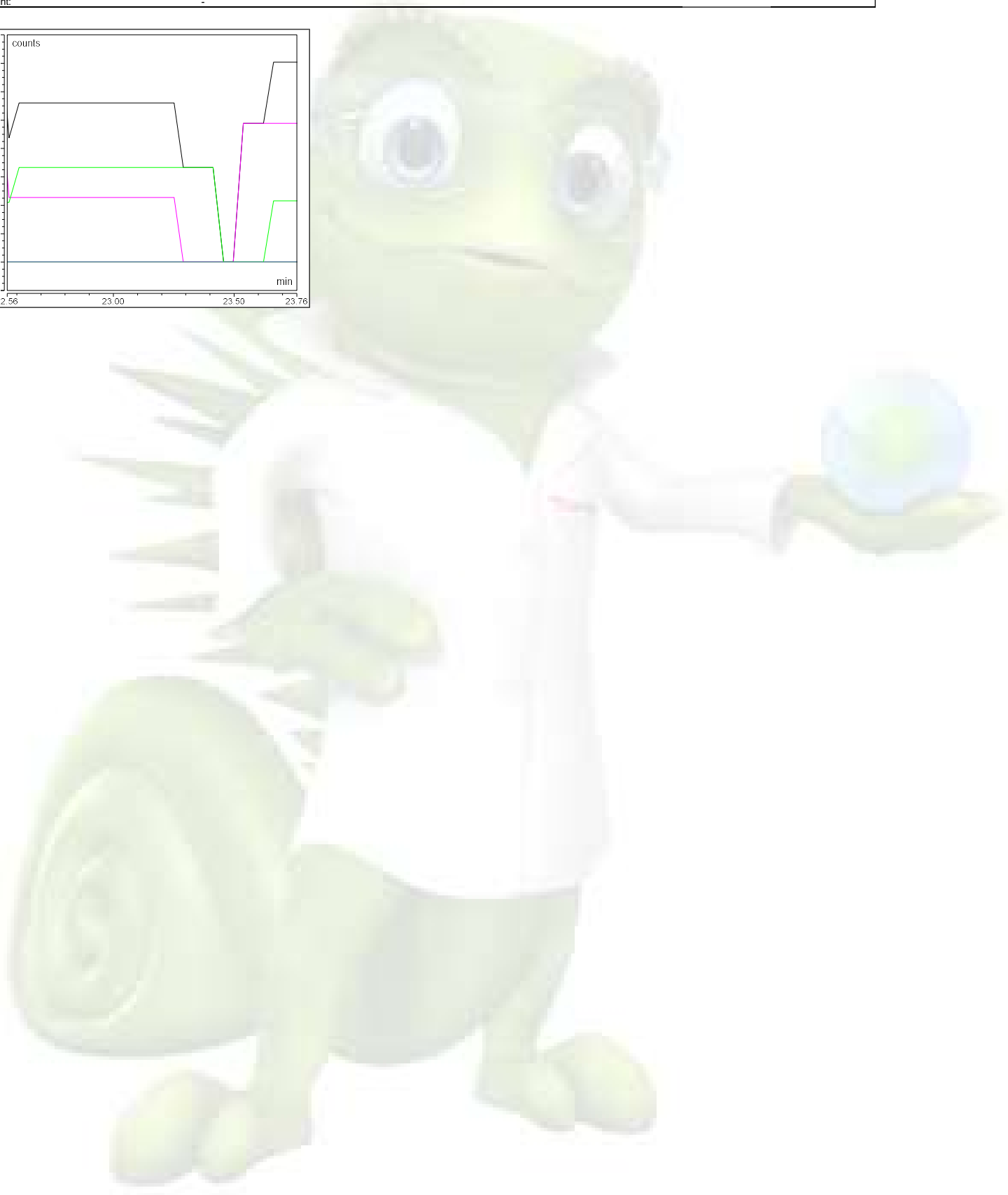
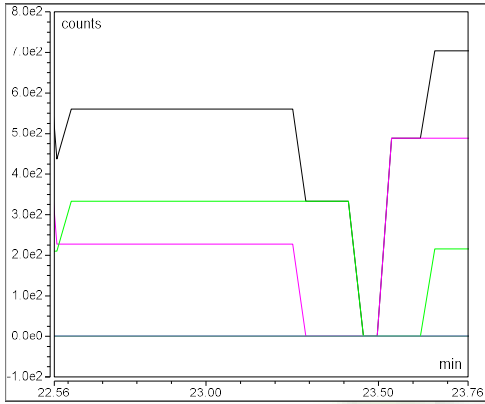
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

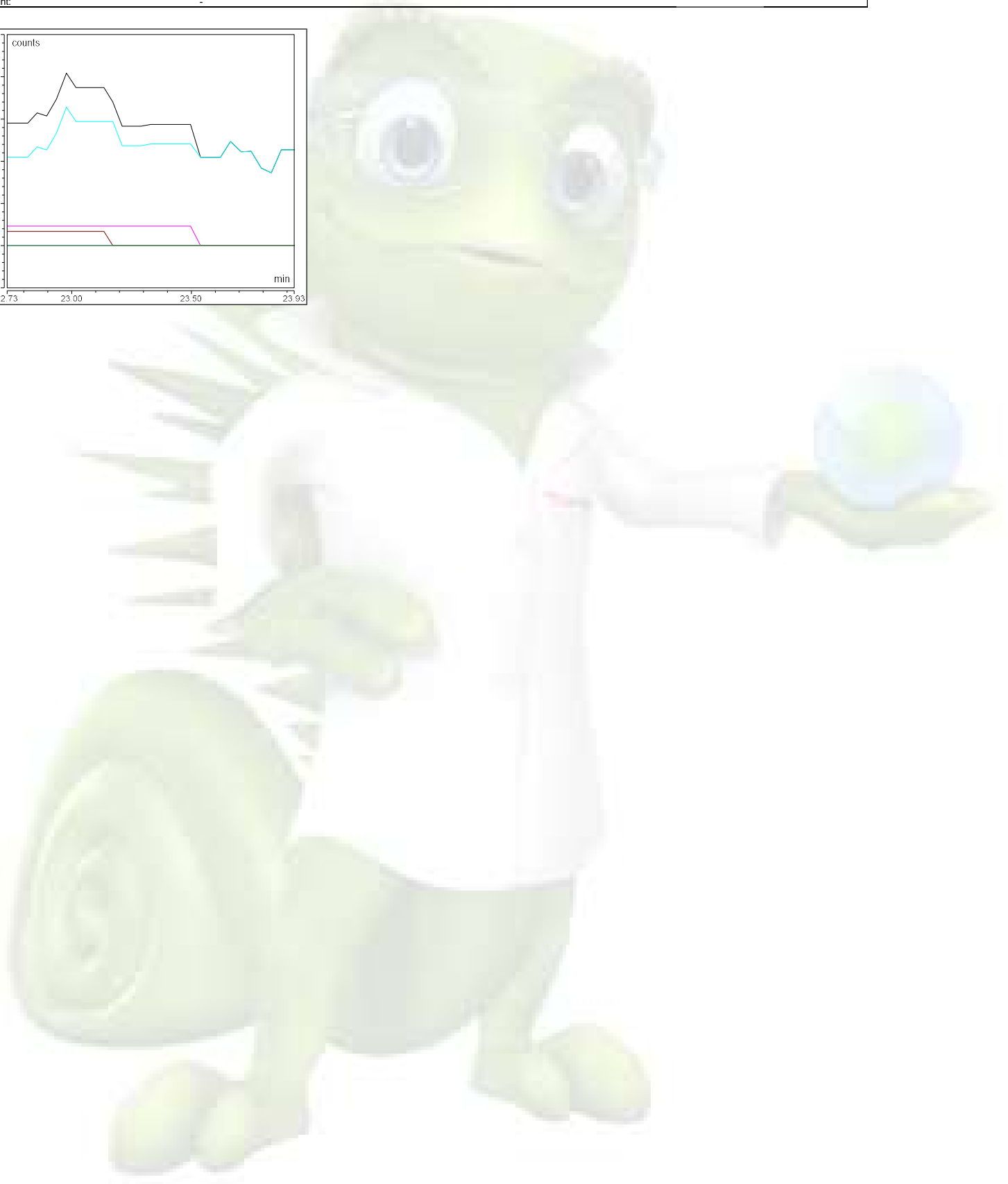
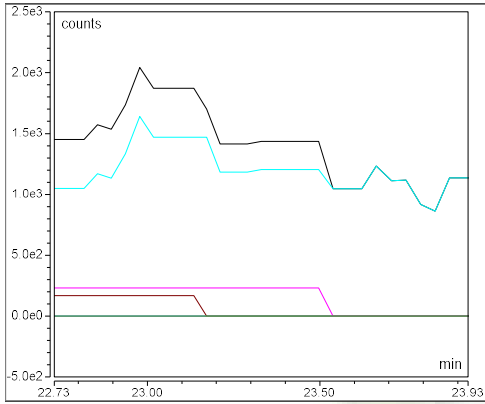
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

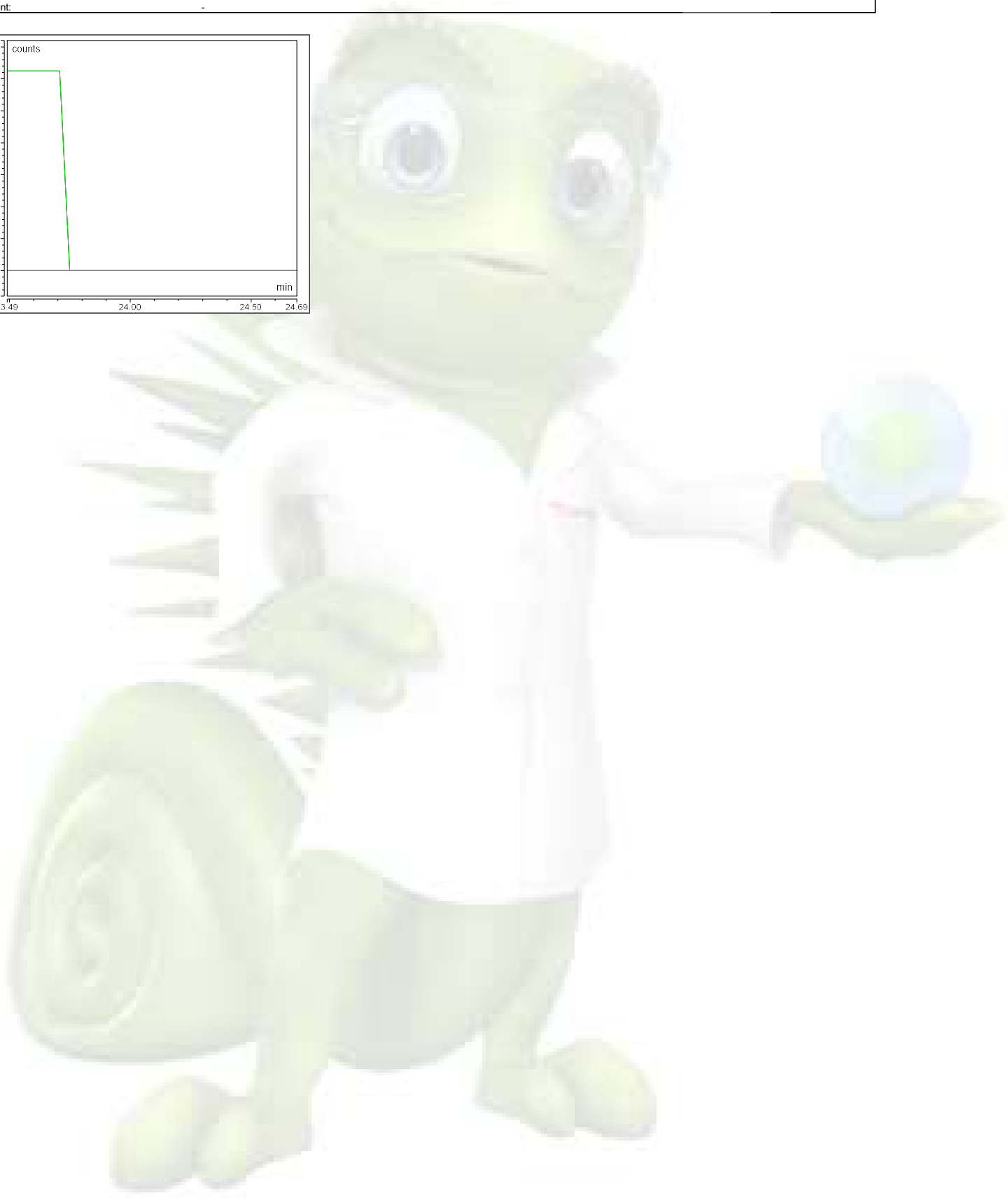
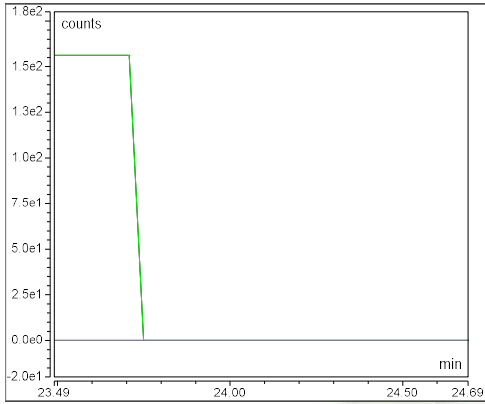
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

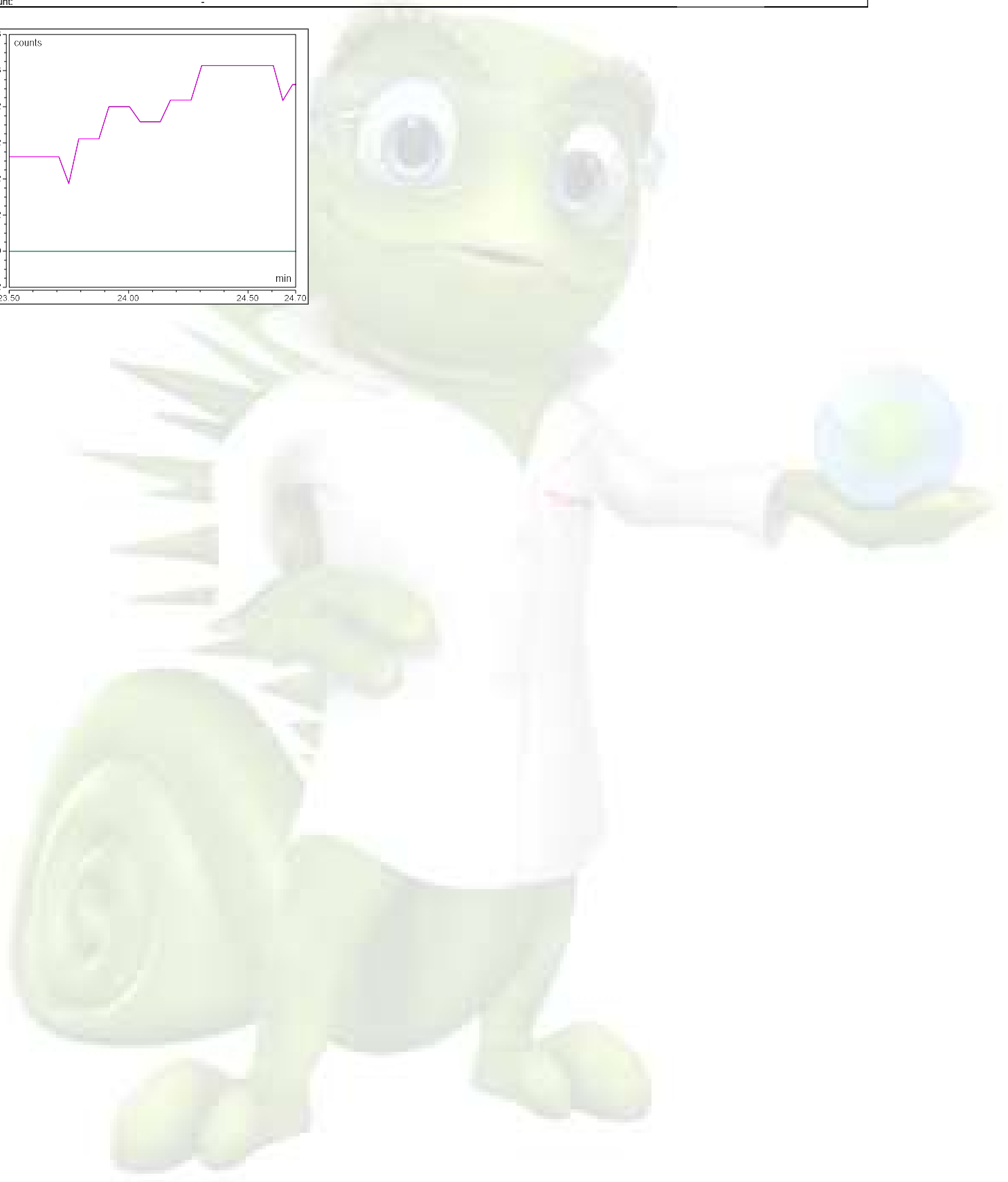
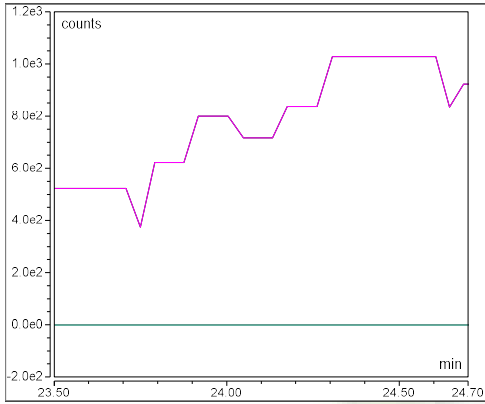
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

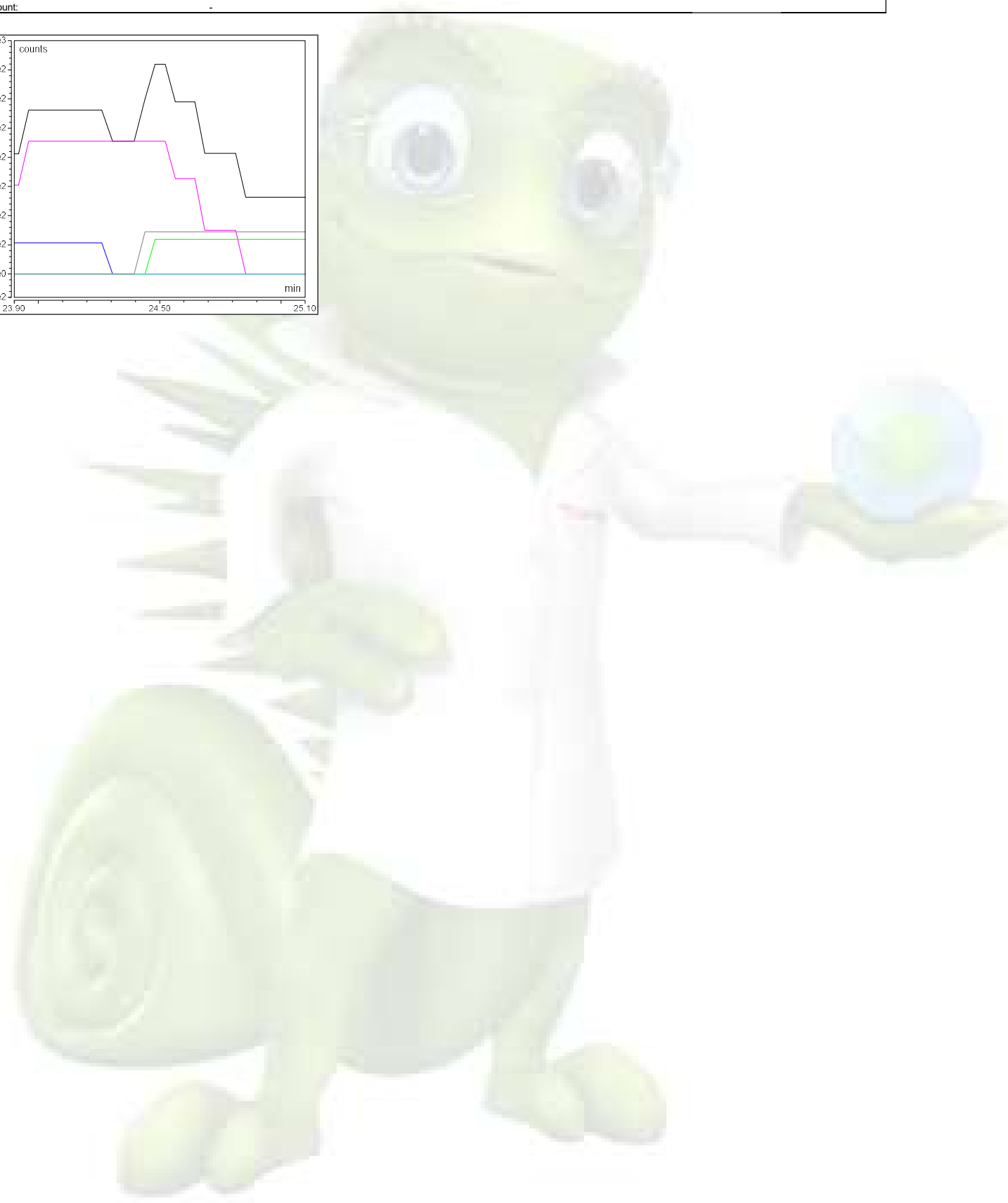
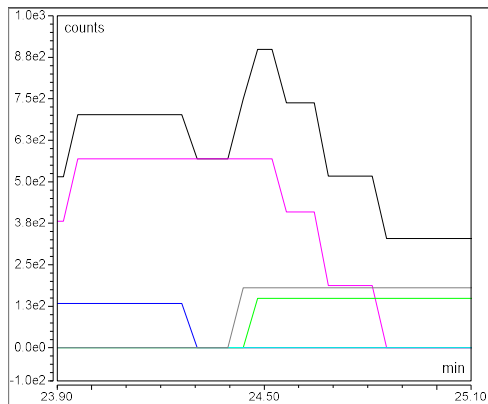
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

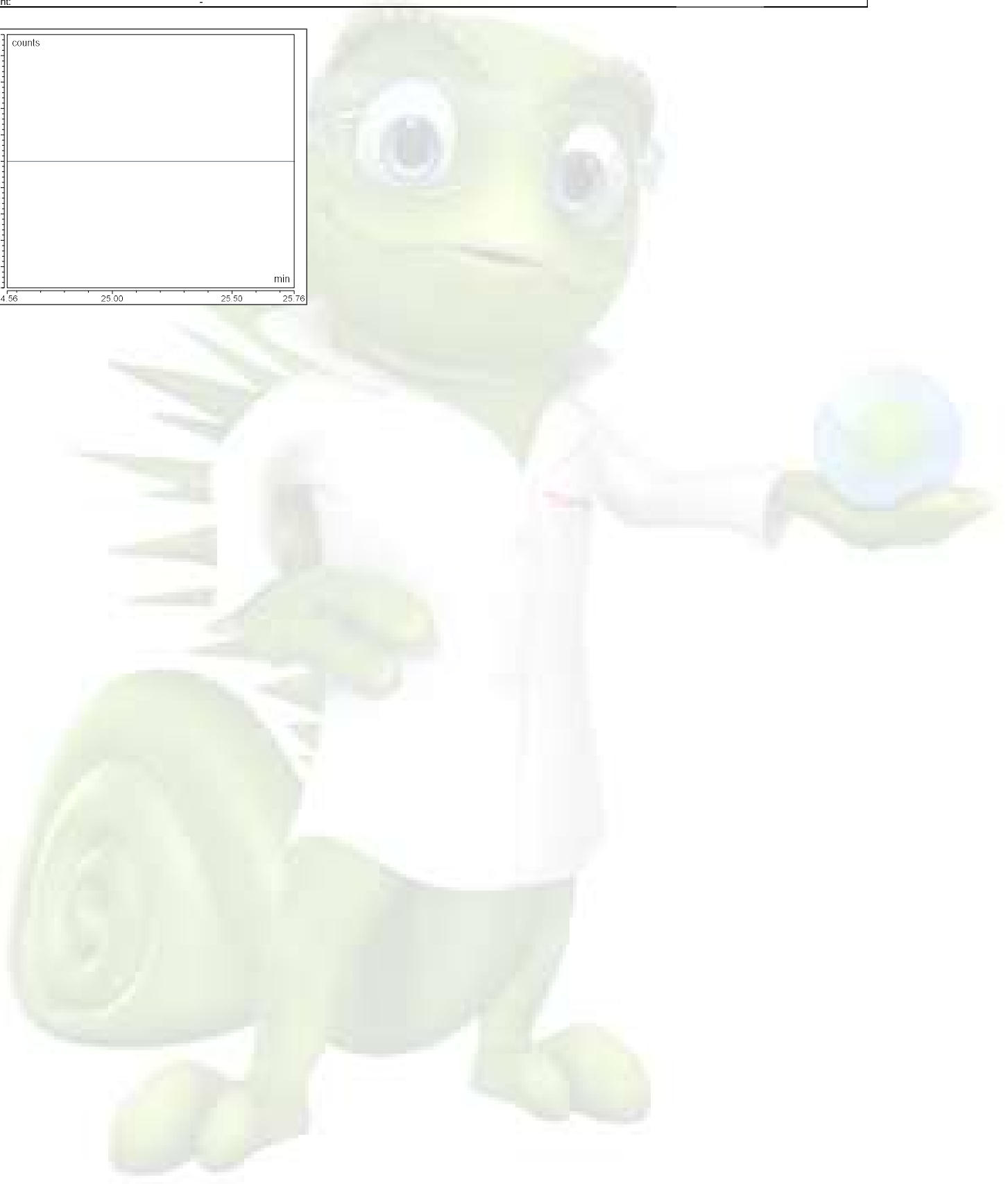
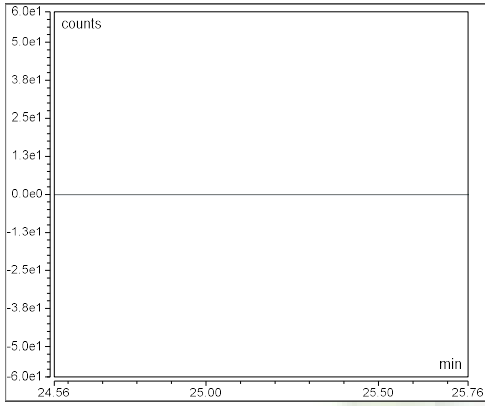
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

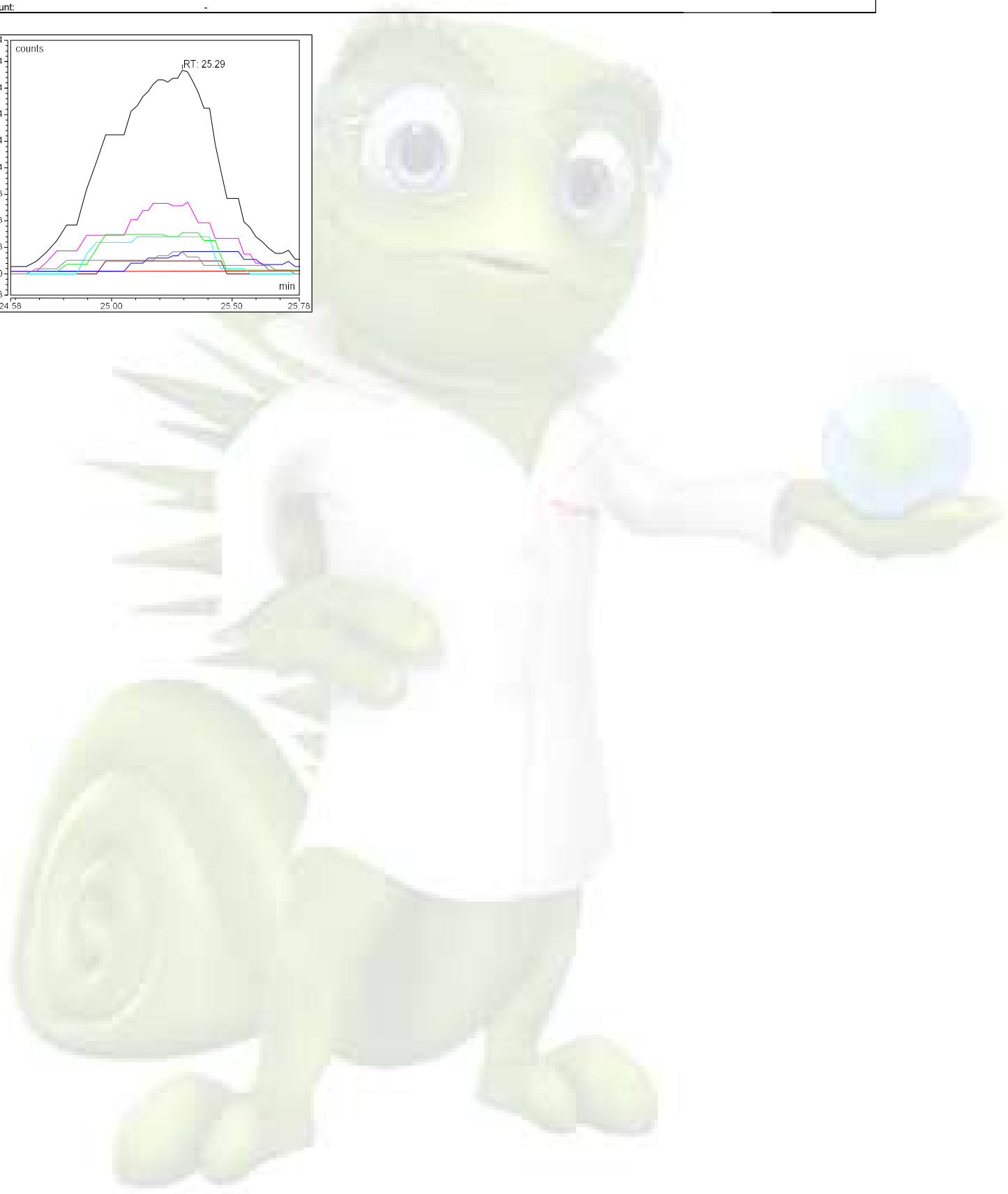
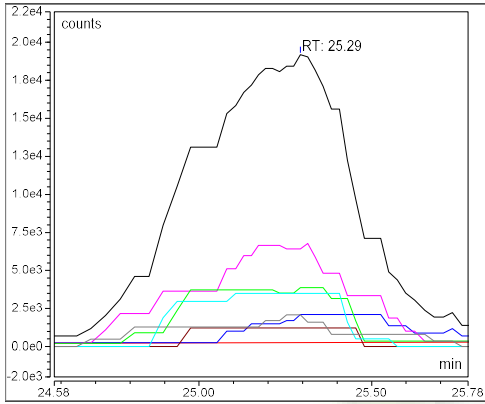
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): 10481

Intensity (counts): 18929

Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

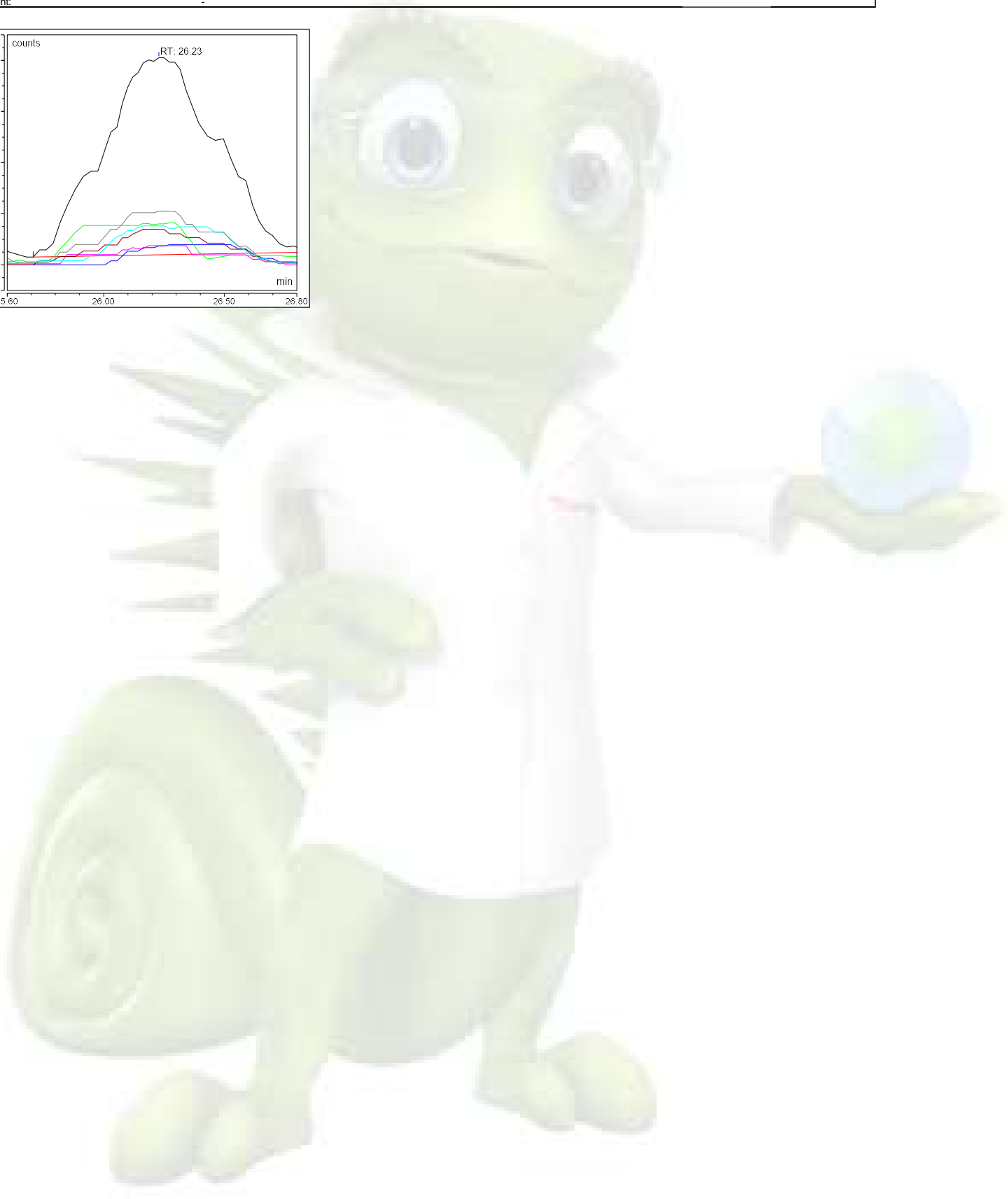
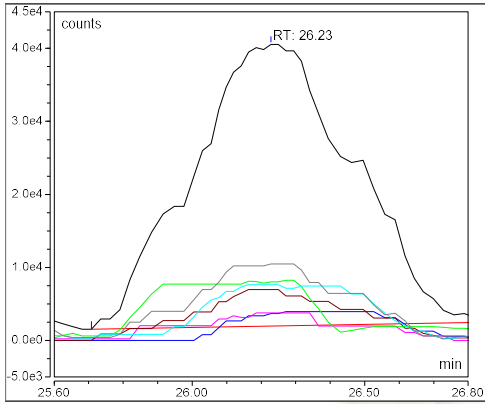
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): 20761

Intensity (counts): 38577

Amount: -

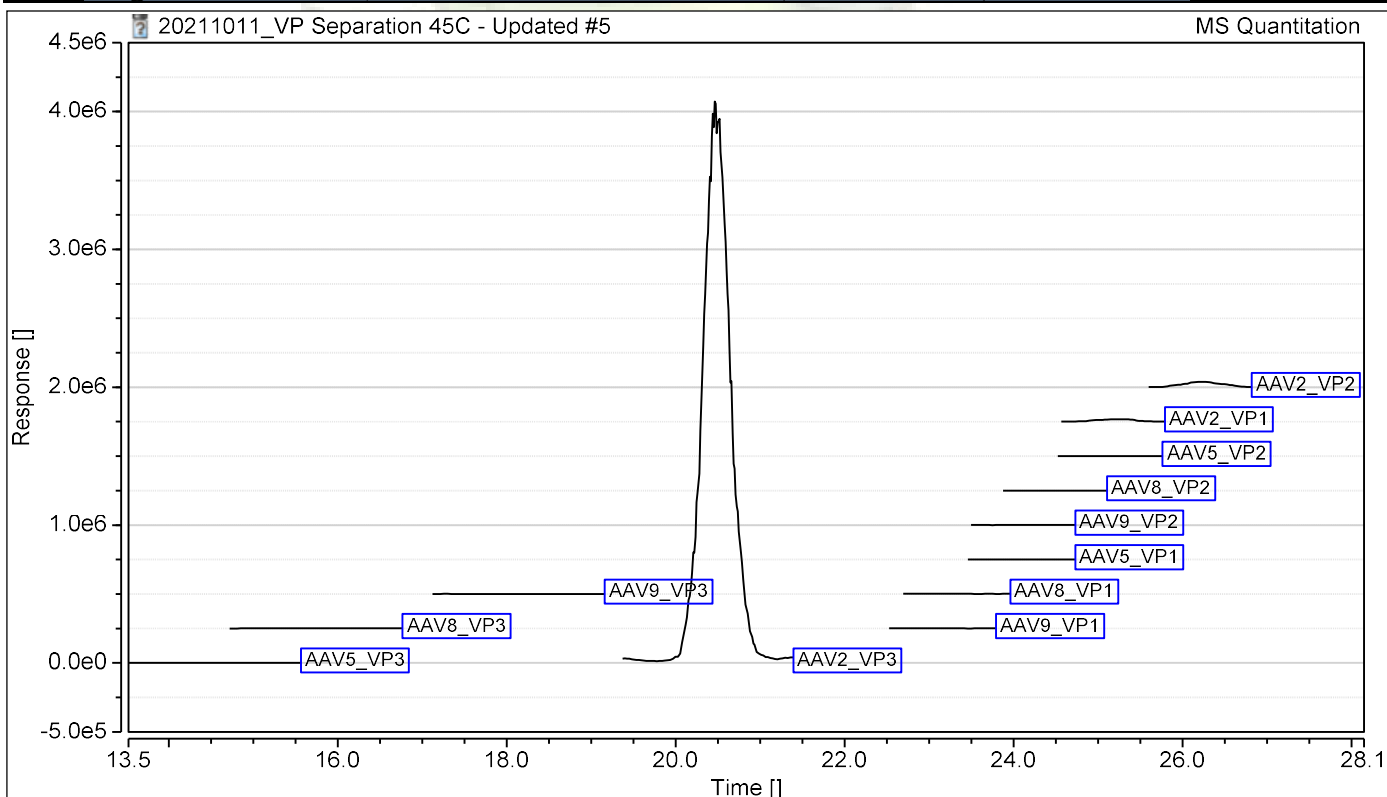


## Chromatogram and SST Results

### Injection Details

|                      |  |                   |        |
|----------------------|--|-------------------|--------|
| Injection Name:      | 20211011_AAV2 E_45C_65-59B_Mod1_1ug_2  | Run Time (min):   | 44.99  |
| Vial Number:         | R:A1   | Injection Volume: | 2.86   |
| Injection Type:      | Unknown  |                   |        |
| Calibration Level:   |  |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                   |        |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 12/Oct/21 22:43  | Sample Weight:    | 1.0000 |

### Chromatogram

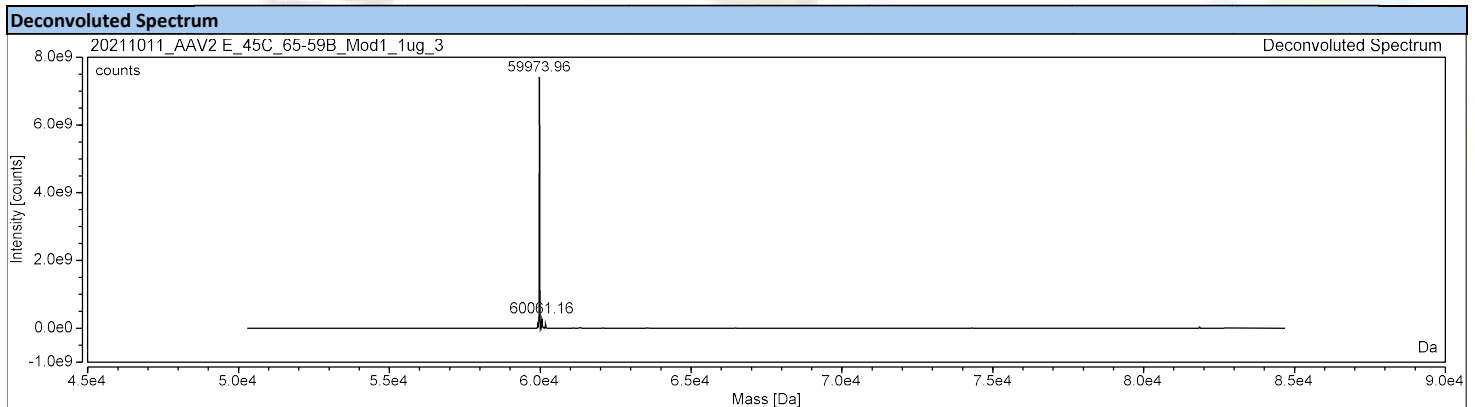
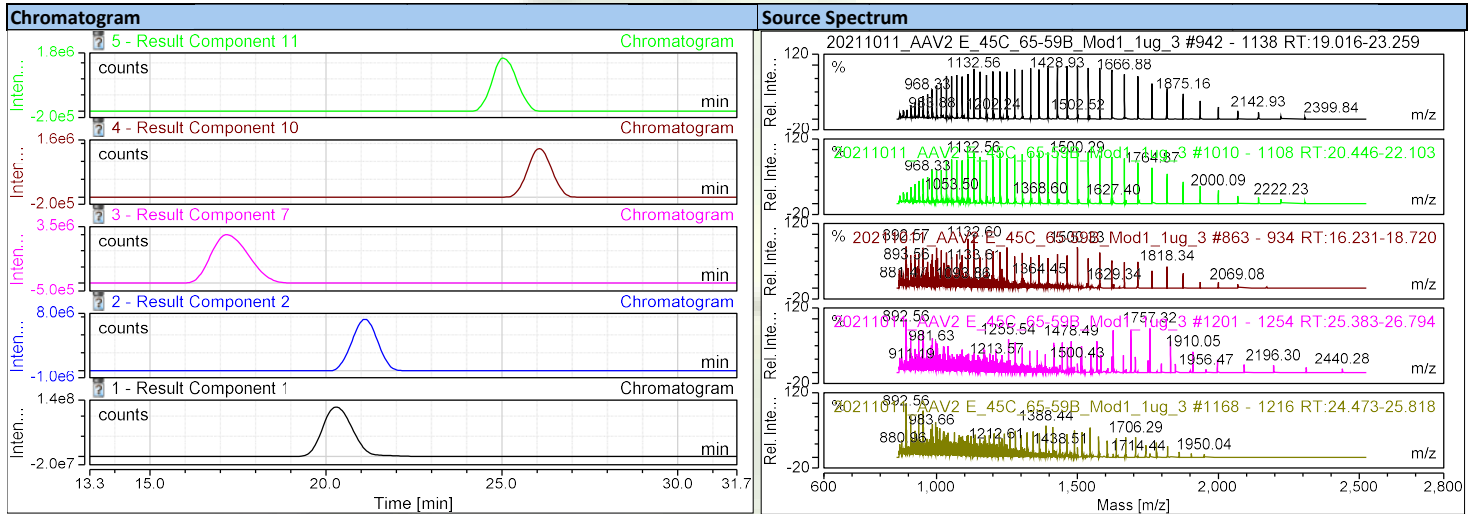


### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV2 E_45C_65-59B_Mod1_1ug_3 | Injection Volume: 2.86                                       |
| Vial Number:      | R:A1                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 45.00                                 | Injection Date/Time: 12/Oct/21 23:29                         |



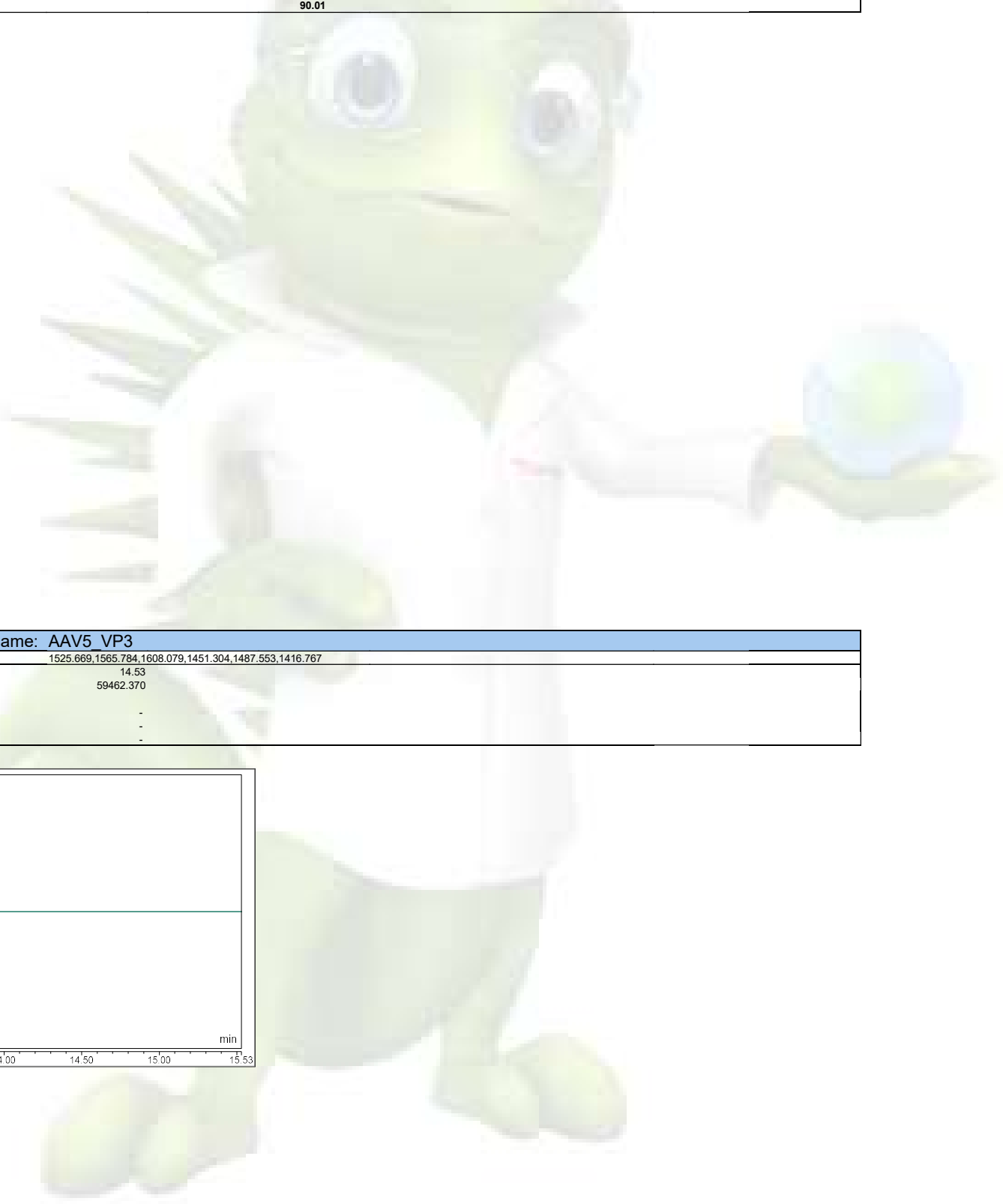
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

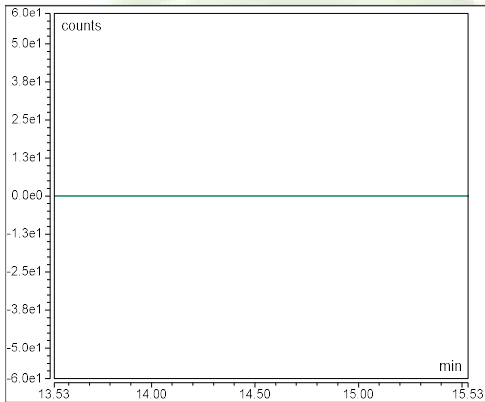
|  |   |  |
|--|---|--|
| <i>Injection Name:</i> 20211011_AAV2_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i> 2.86   |  |
| <i>Vial Number:</i> R:A1                                   | <i>Instrument Method:</i> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <i>Injection Type:</i> Unknown                             | <i>Processing Method:</i> MS Quantitative_45C_65-59B                            |  |
| <i>Run Time (min):</i> 45.00                               | <i>Injection Date/Time:</i> 12/Oct/21 23:29                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3     | 20.331                | 1247828.061        | 87.99              | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 | 59973.780 | PASS                           |
| AAV2_VP1     | 25.120                | 11832.608          | 0.83               | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 | 81854.900 | PASS                           |
| AAV2_VP2     | 26.051                | 16781.587          | 1.18               | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 | 66487.900 | PASS                           |
| <b>90.01</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |





**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

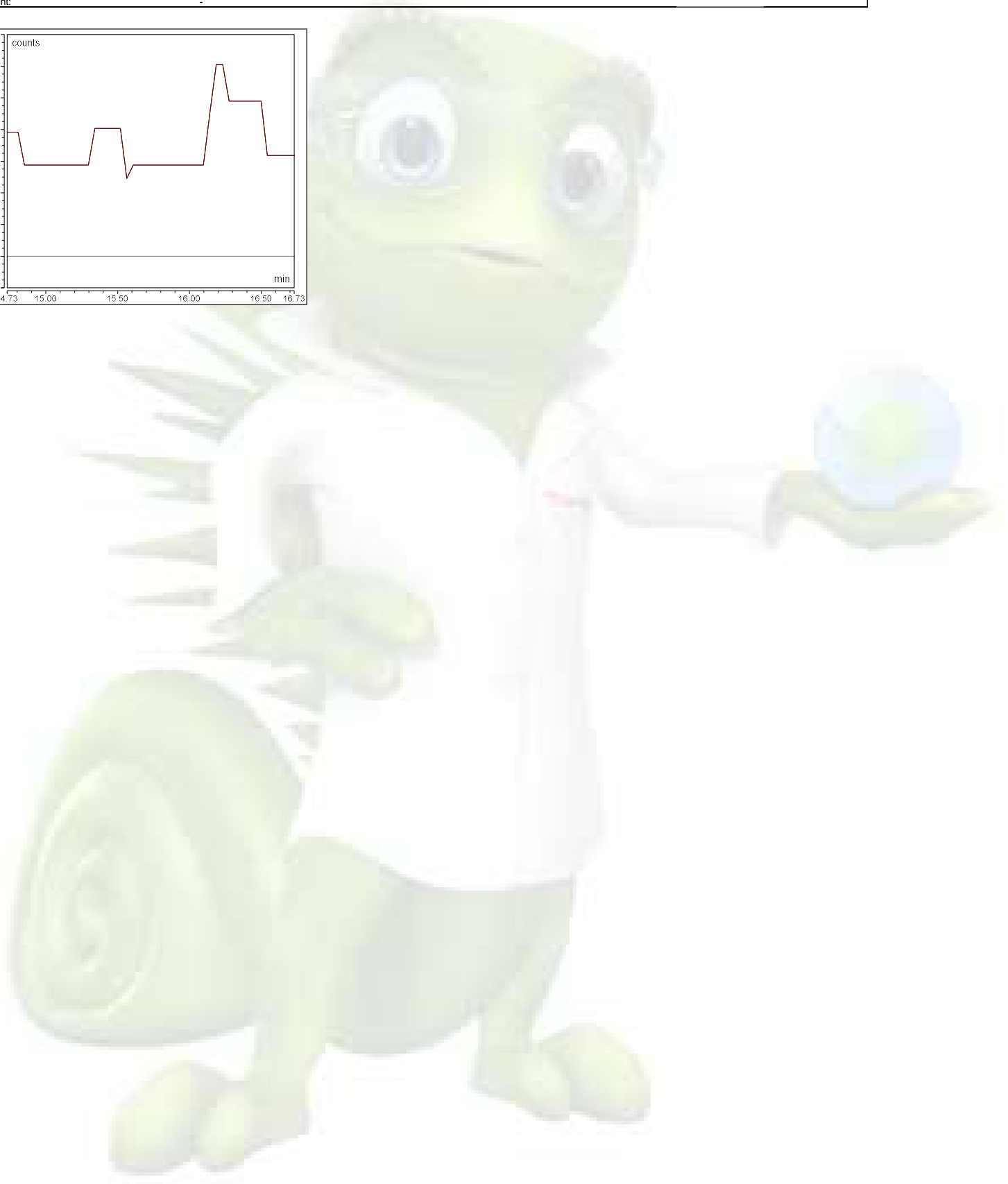
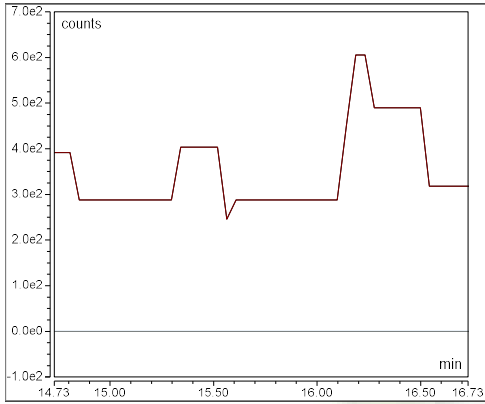
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

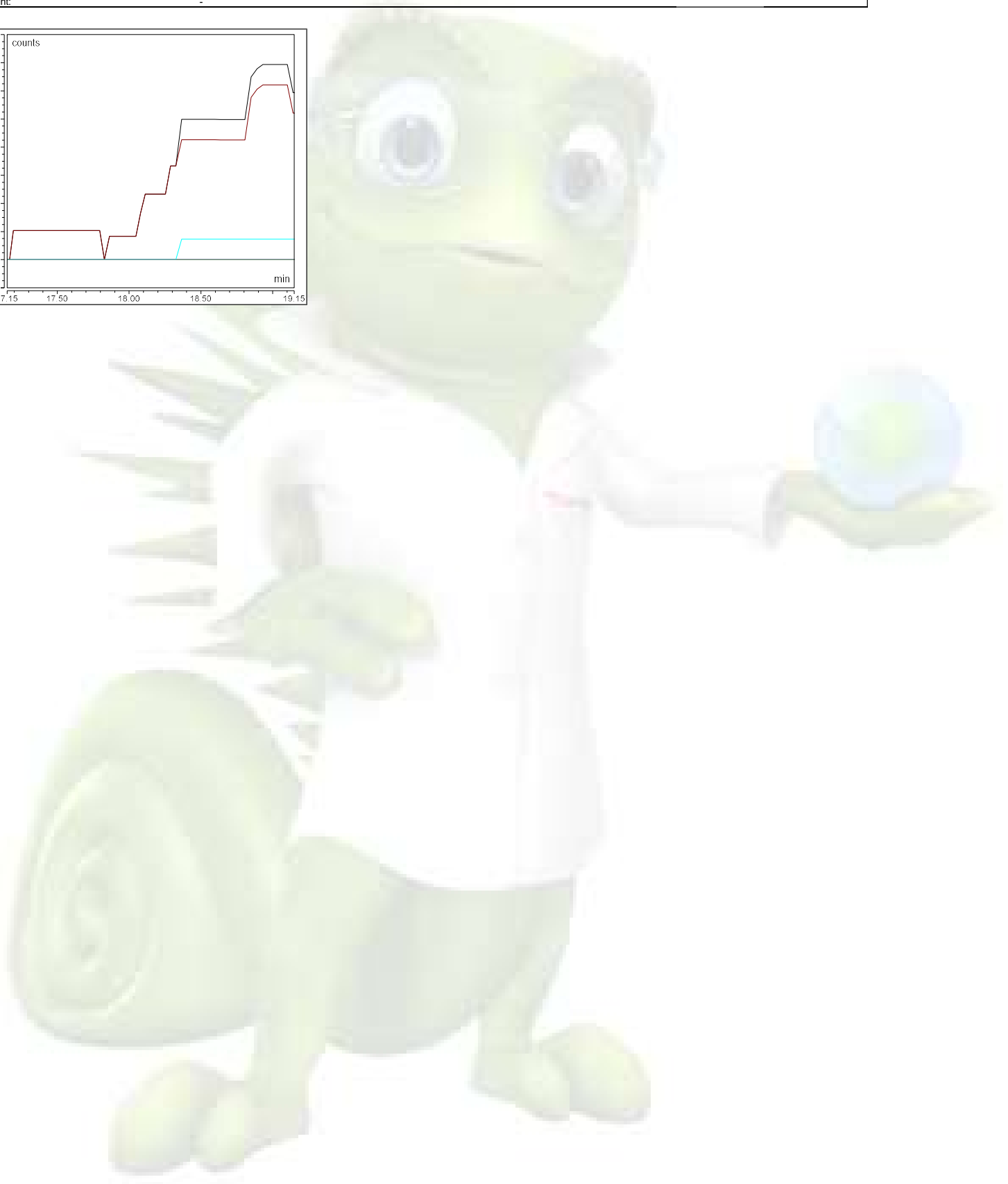
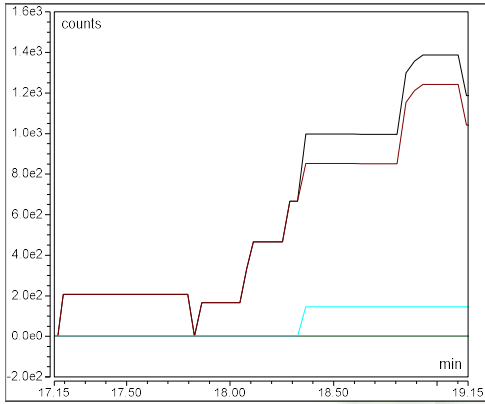
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

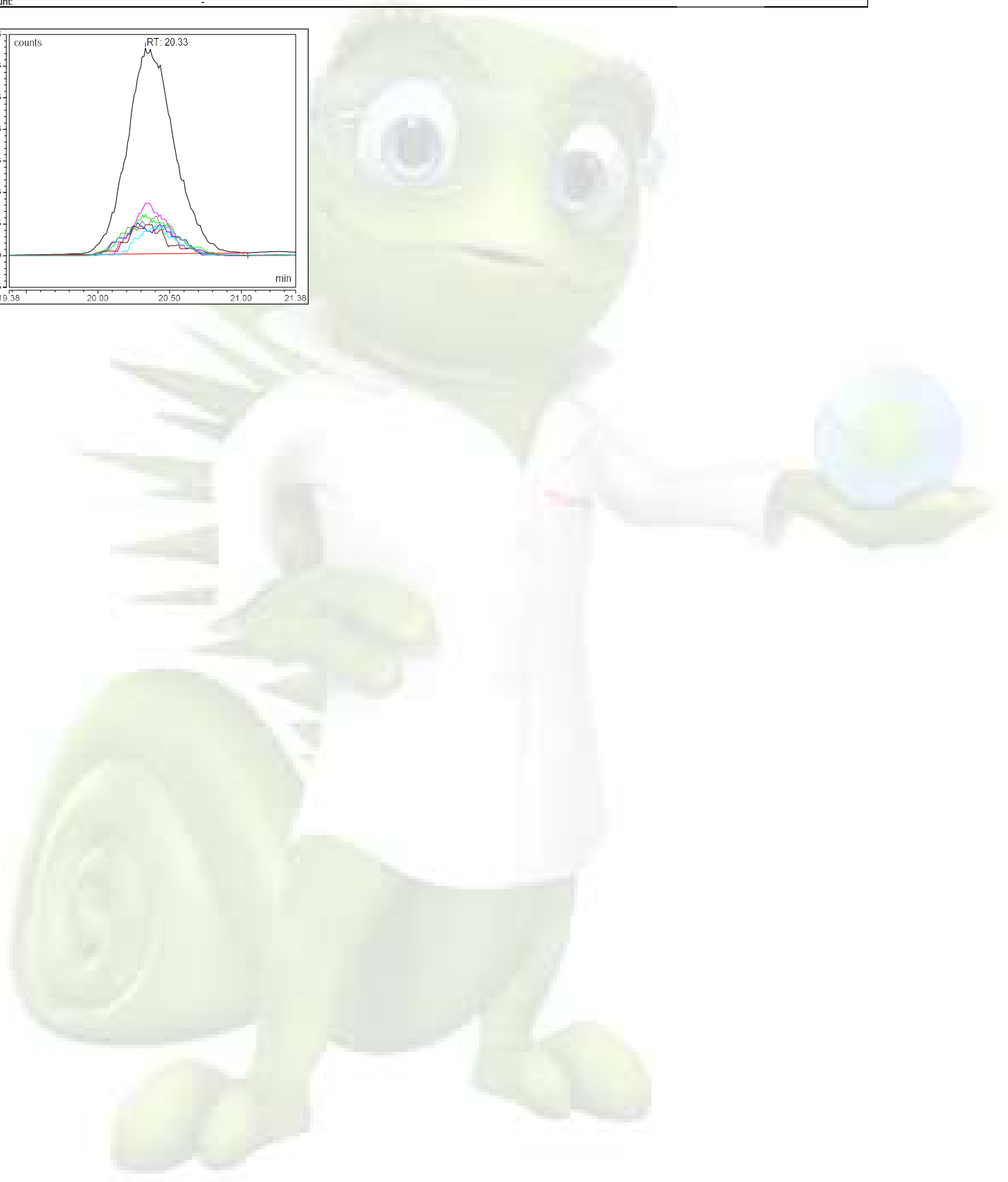
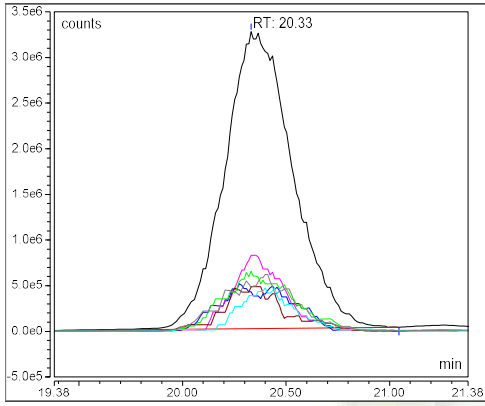
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): 1247828

Intensity (counts): 3256317

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

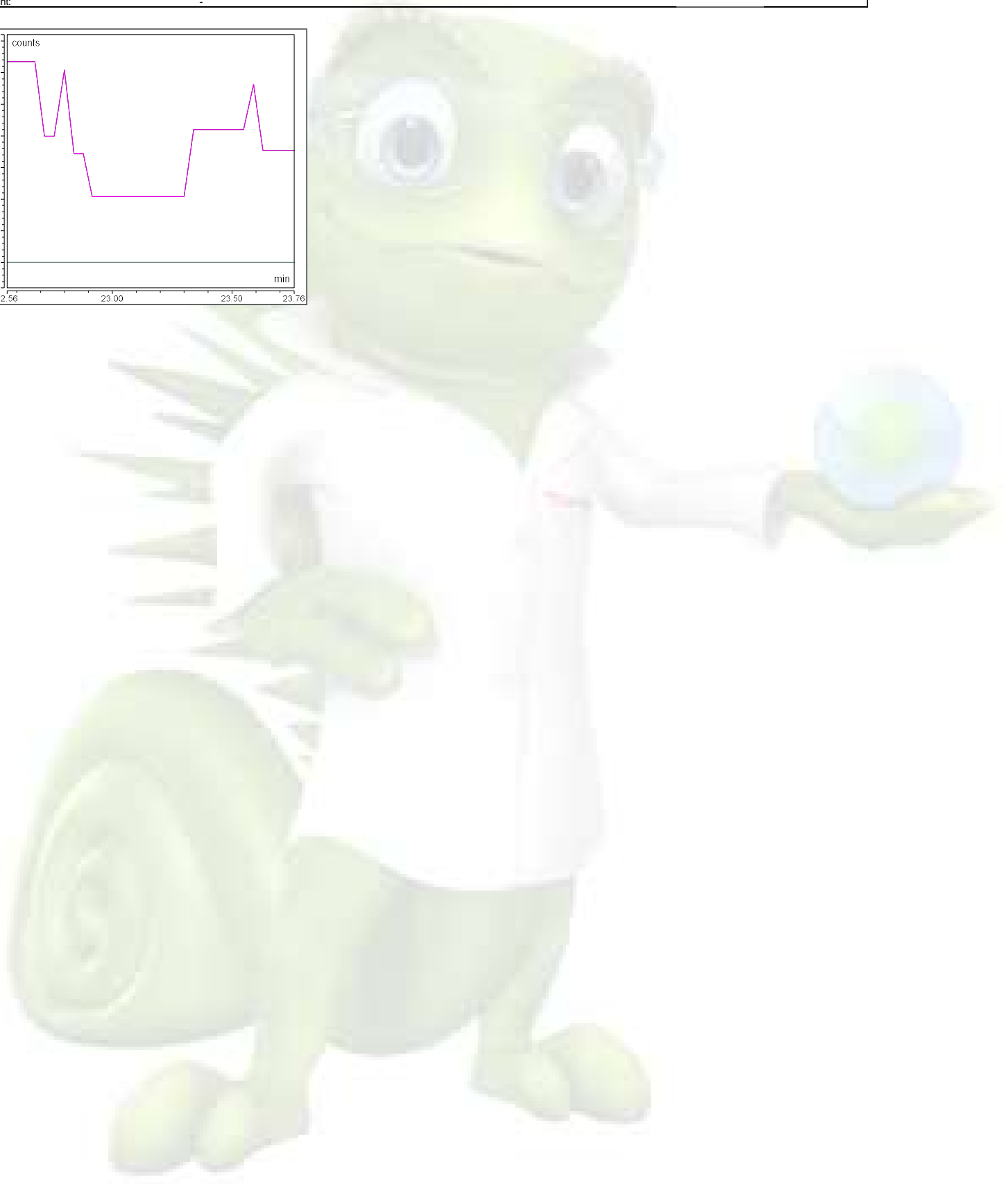
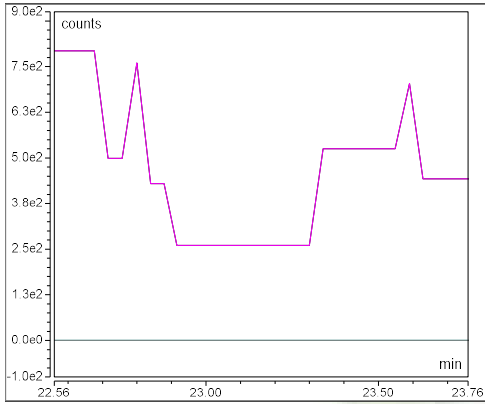
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

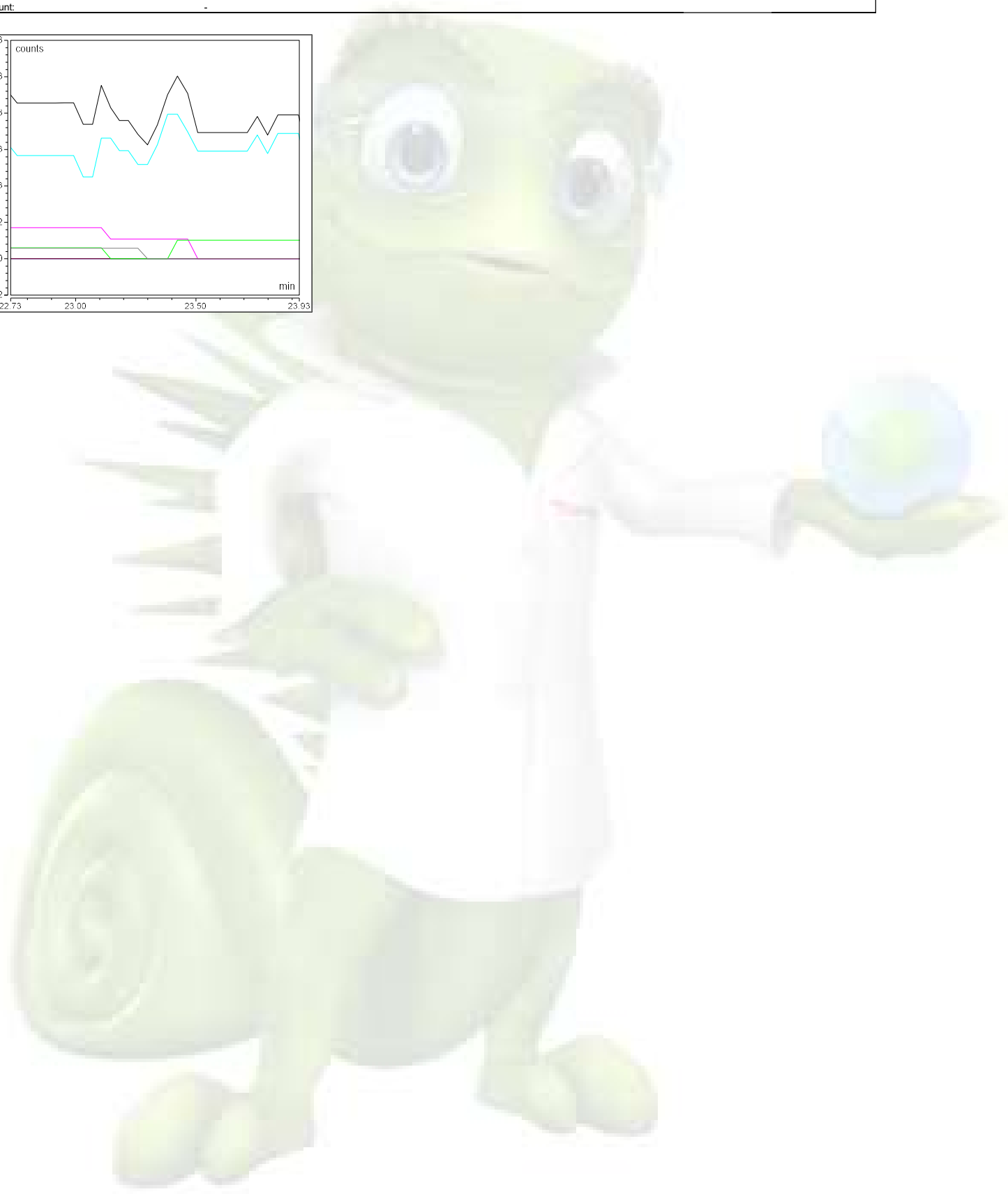
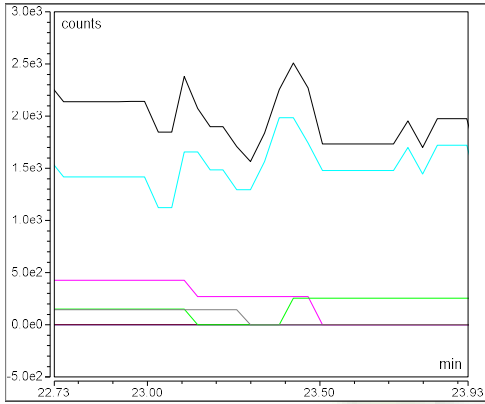
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

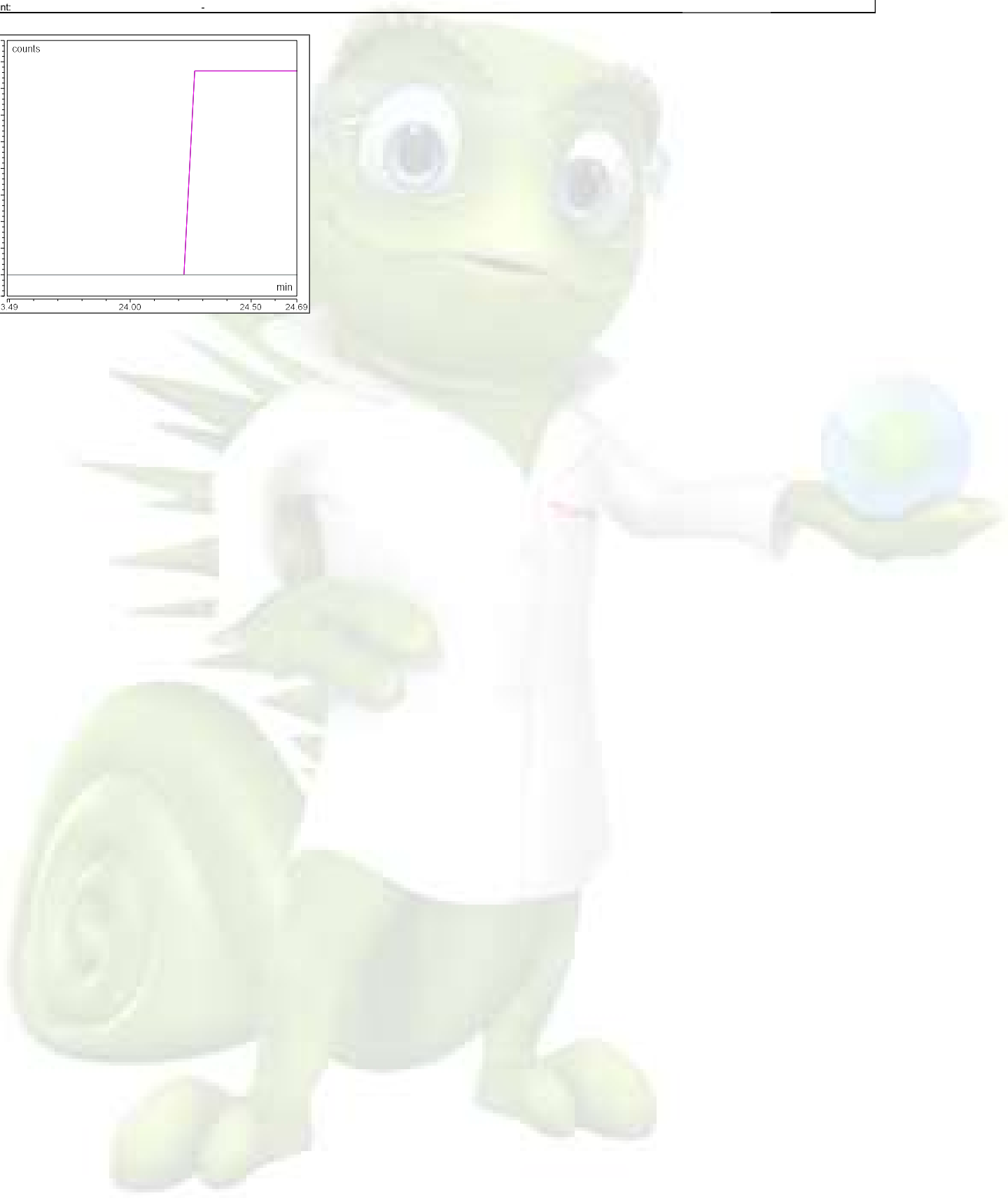
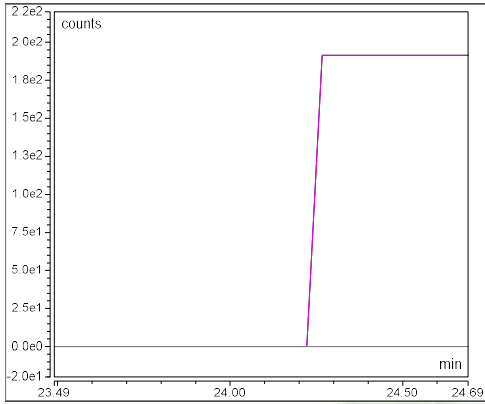
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

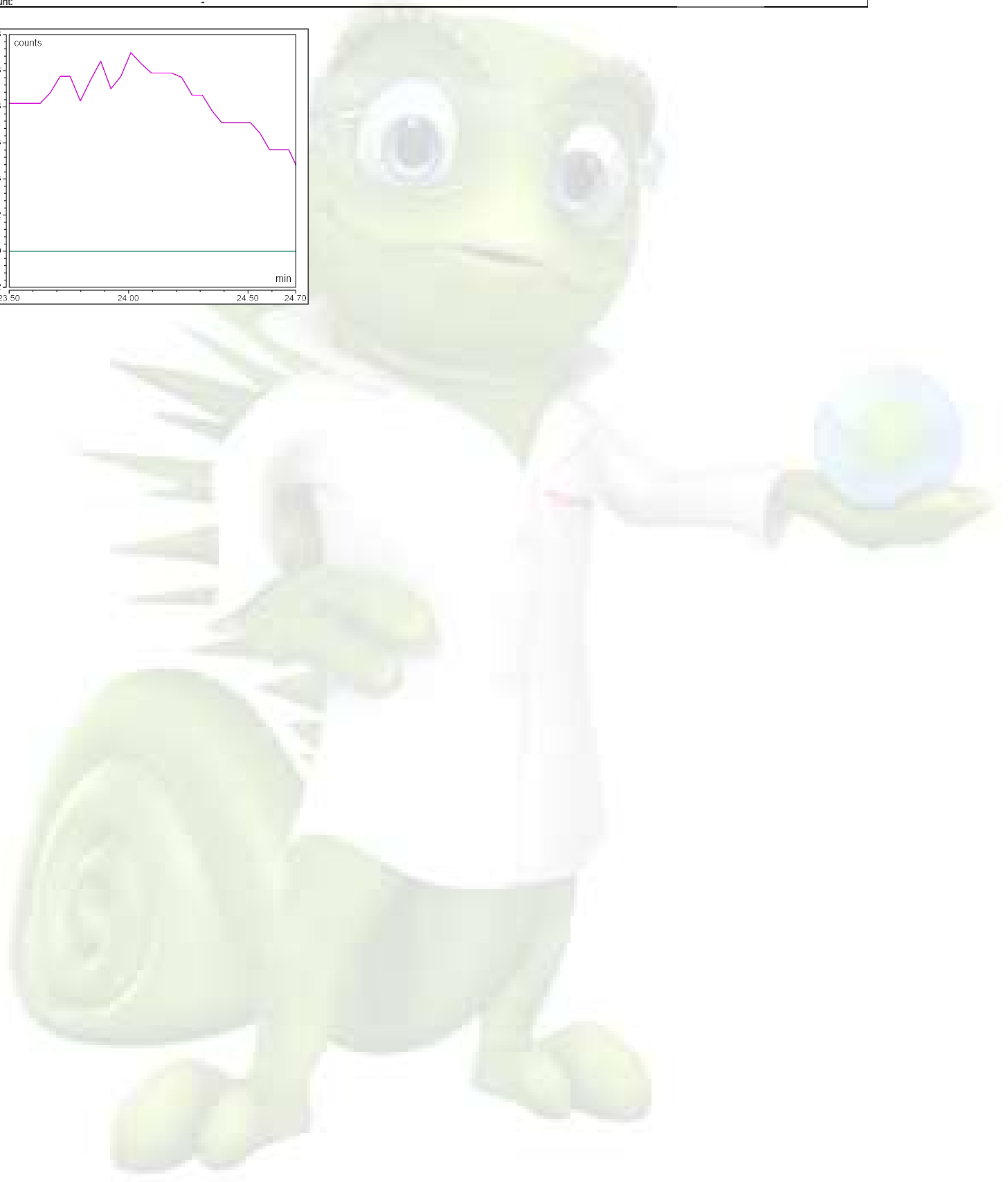
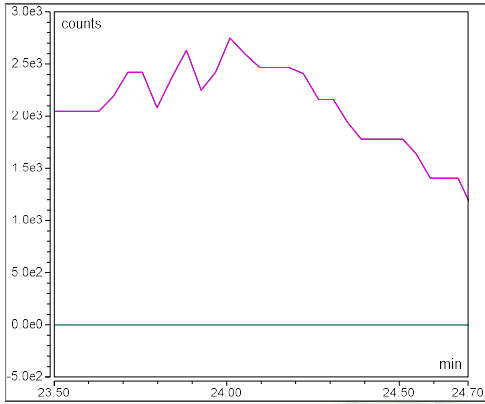
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

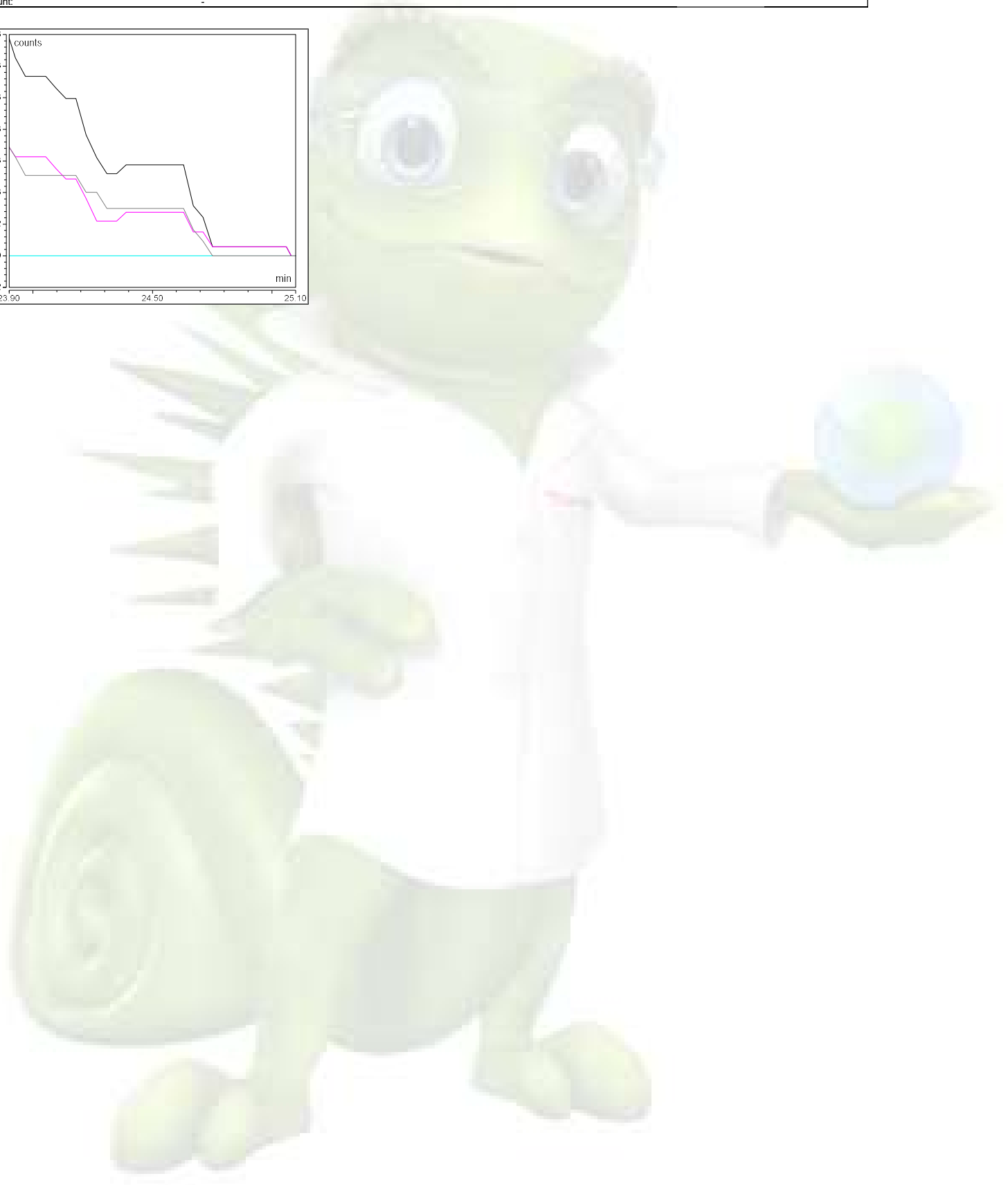
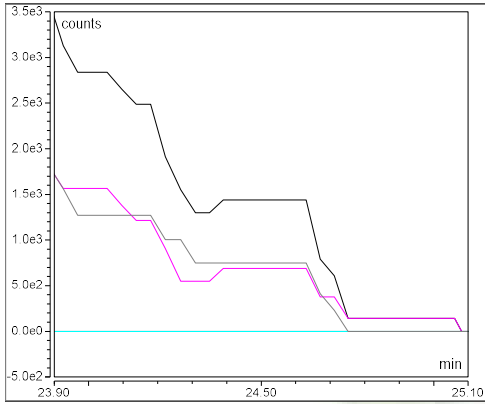
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

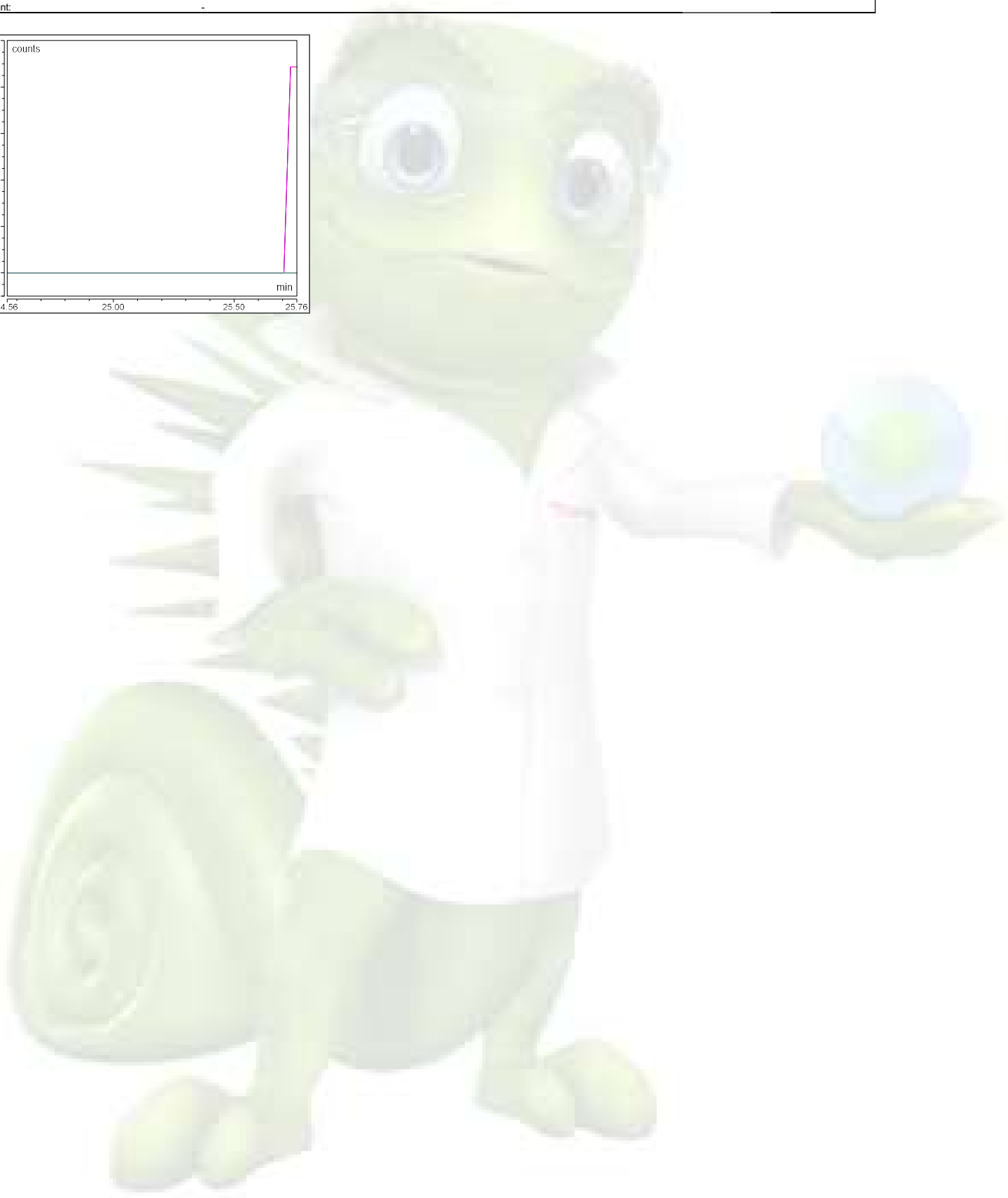
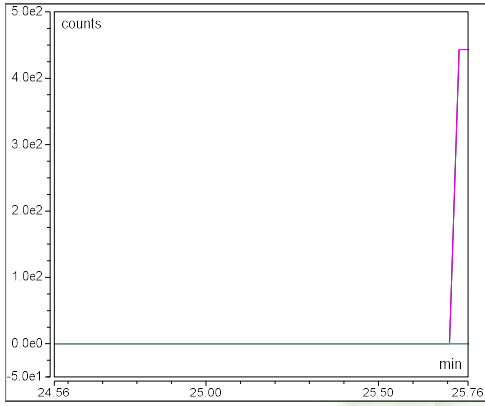
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

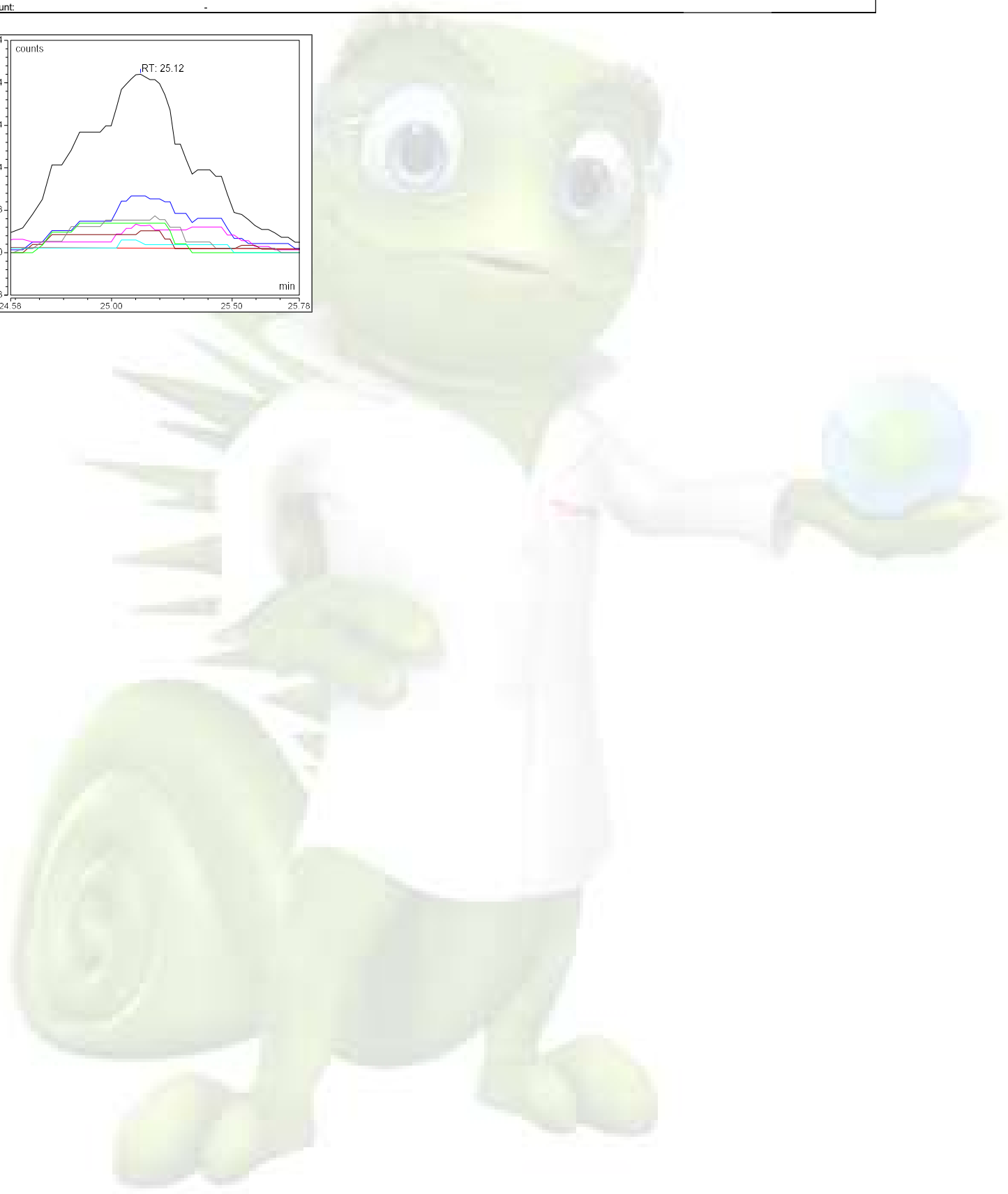
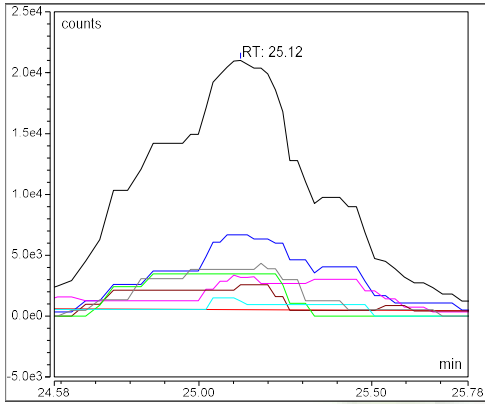
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): 11833

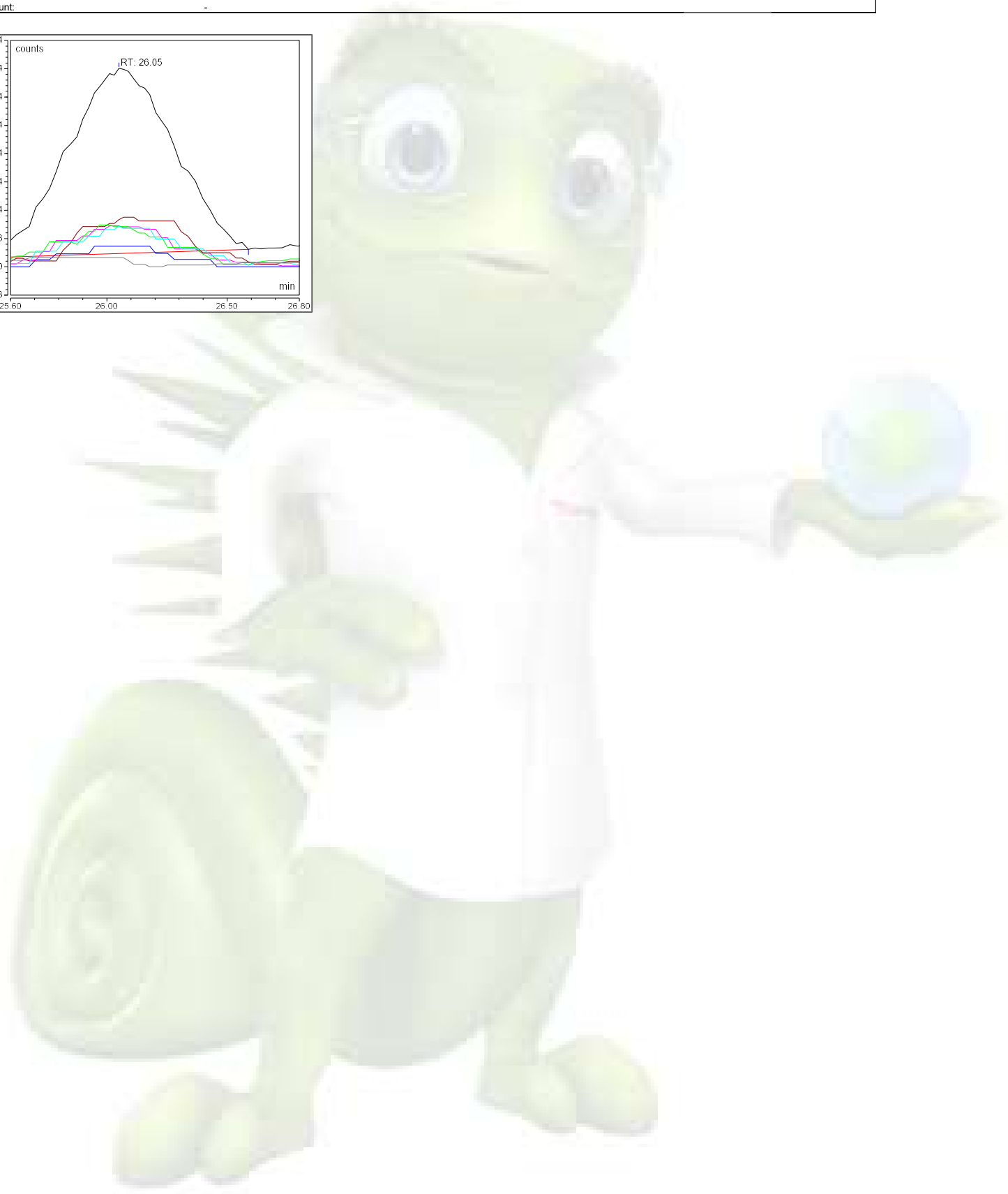
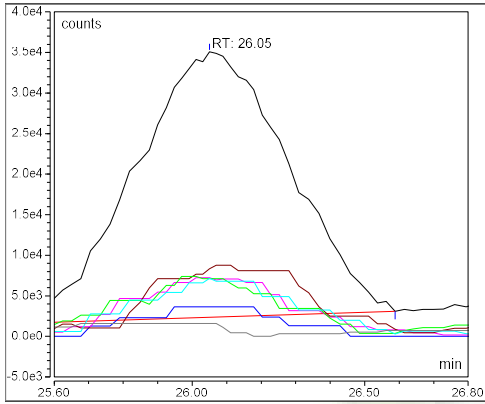
Intensity (counts): 20477

Amount: -



**Component Name: AAV2 VP2**

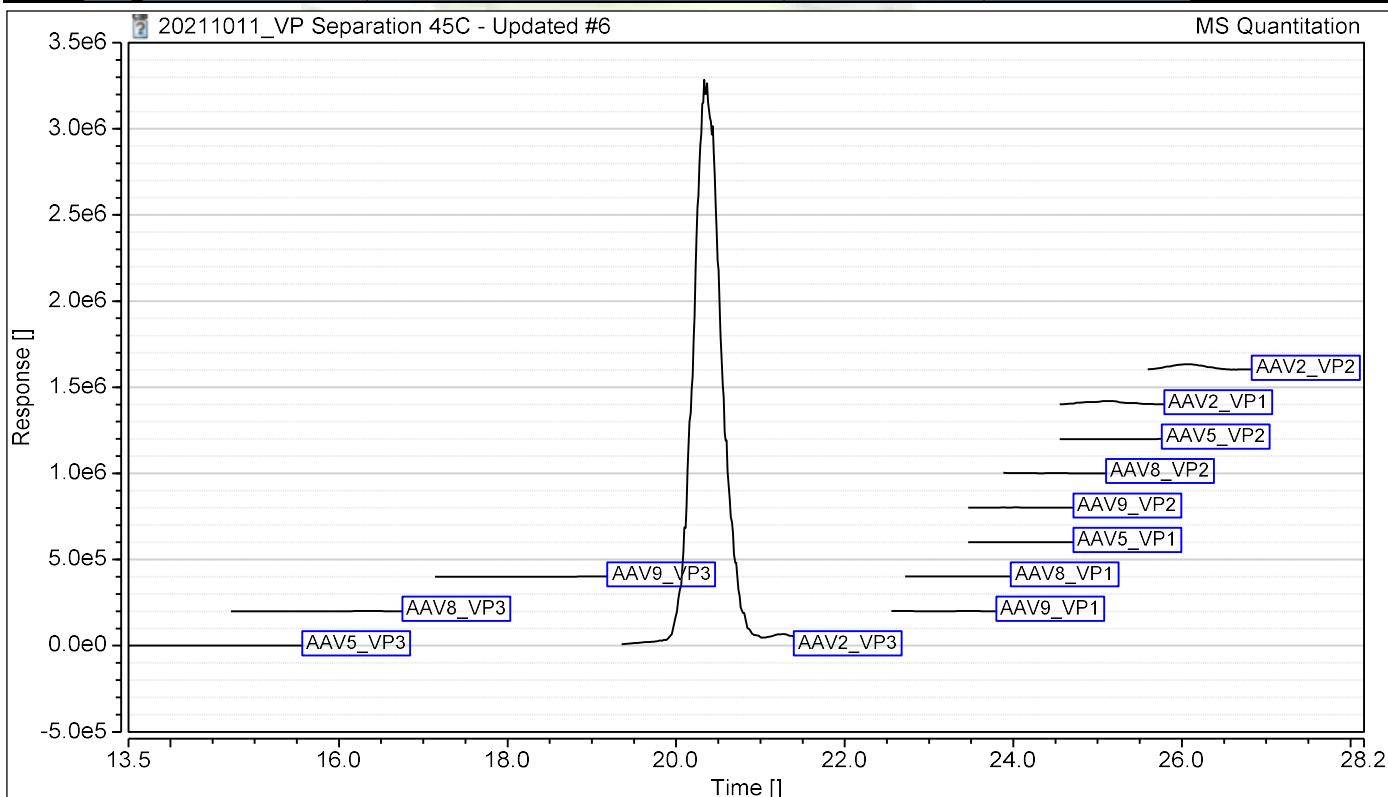
|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 |
| Retention time (min): | 26.20   |
| Expected Mass:        | 66487.900   |
| Area (count*min):     | 16782   |
| Intensity (counts):   | 32732   |
| Amount:               | -   |



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV2 E_45C_65-59B_Mod1_1ug_3                                      | Run Time (min): 45.00   |
| Vial Number:         | R:A1   | Injection Volume: 2.86  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 | S                       |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 12/Oct/21 23:29  | Sample Weight: 1.0000   |

### Chromatogram

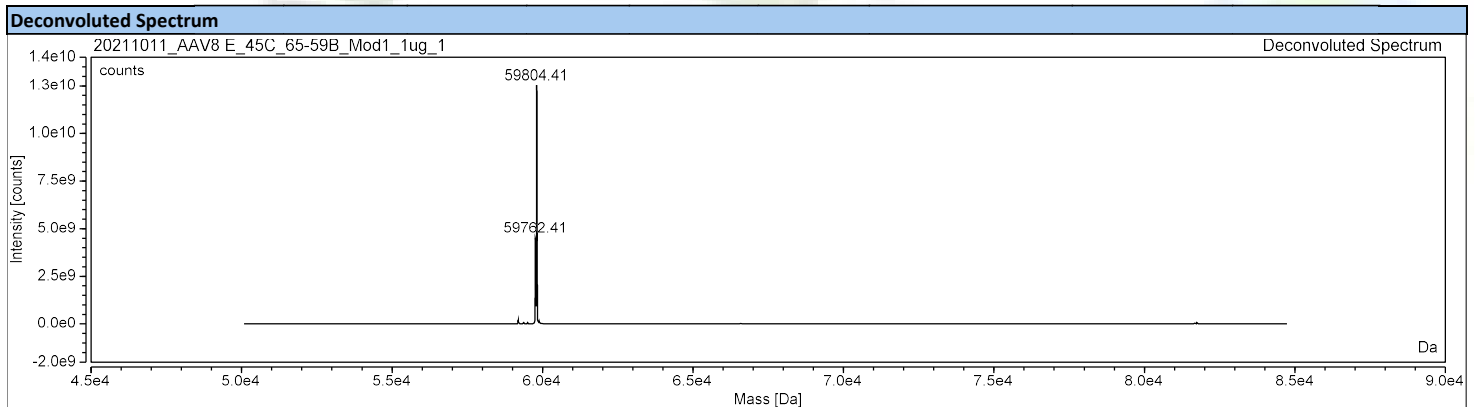
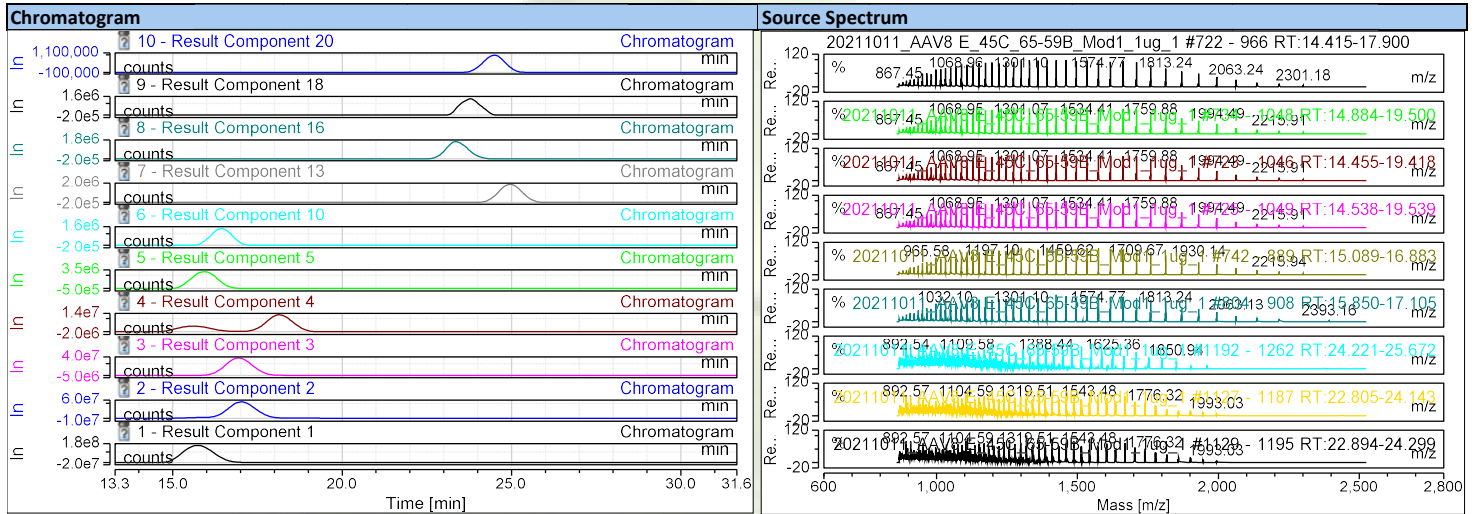


### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV8 E_45C_65-59B_Mod1_1ug_1 | Injection Volume: 6.25                                       |
| Vial Number:      | R:A3                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 45.00                                 | Injection Date/Time: 13/Oct/21 01:42                         |



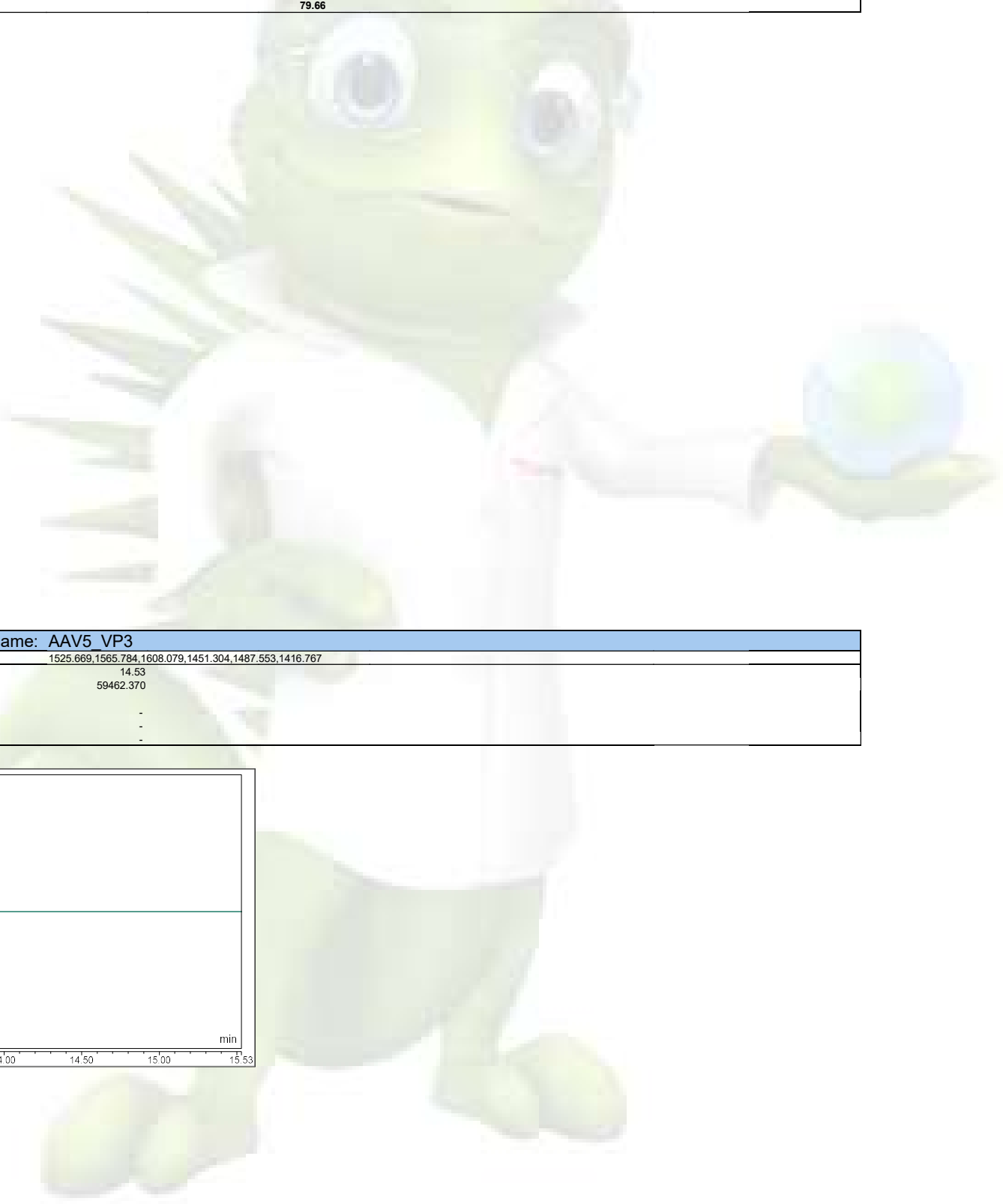
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

**Component Summary**

**Injection Details**

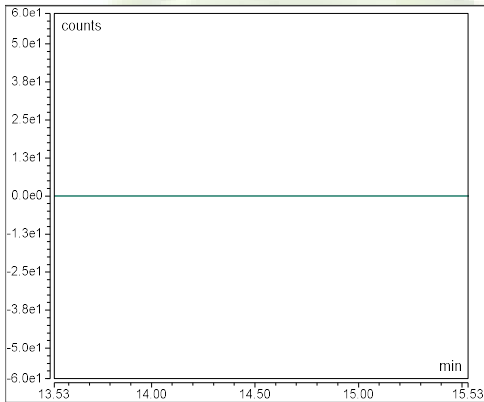
|                        |                                     |                             |   |
|------------------------|-------------------------------------|-----------------------------|---|
| <i>Injection Name:</i> | 20211011_AAV8_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i>    | 6.25  |
| <i>Vial Number:</i>    | R:A3                                | <i>Instrument Method:</i>   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |
| <i>Injection Type:</i> | Unknown                             | <i>Processing Method:</i>   | MS Quantitative_45C_65-59B                            |
| <i>Run Time (min):</i> | 45.00                               | <i>Injection Date/Time:</i> | 13/Oct/21 01:42                                       |

| Peak Name | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|-----------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV8_VP3  | 15.727                | 1734029.695        | 78.06              | 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802 | 59804.680 | PASS                           |
| AAV8_VP1  | 23.494                | 21159.537          | 0.95               | 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828 | 81666.710 | PASS                           |
| AAV8_VP2  | 24.467                | 14492.280          | 0.65               | 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397 | 66518.100 | PASS                           |
|           |                       |                    | <b>79.66</b>       |   |           |                                |



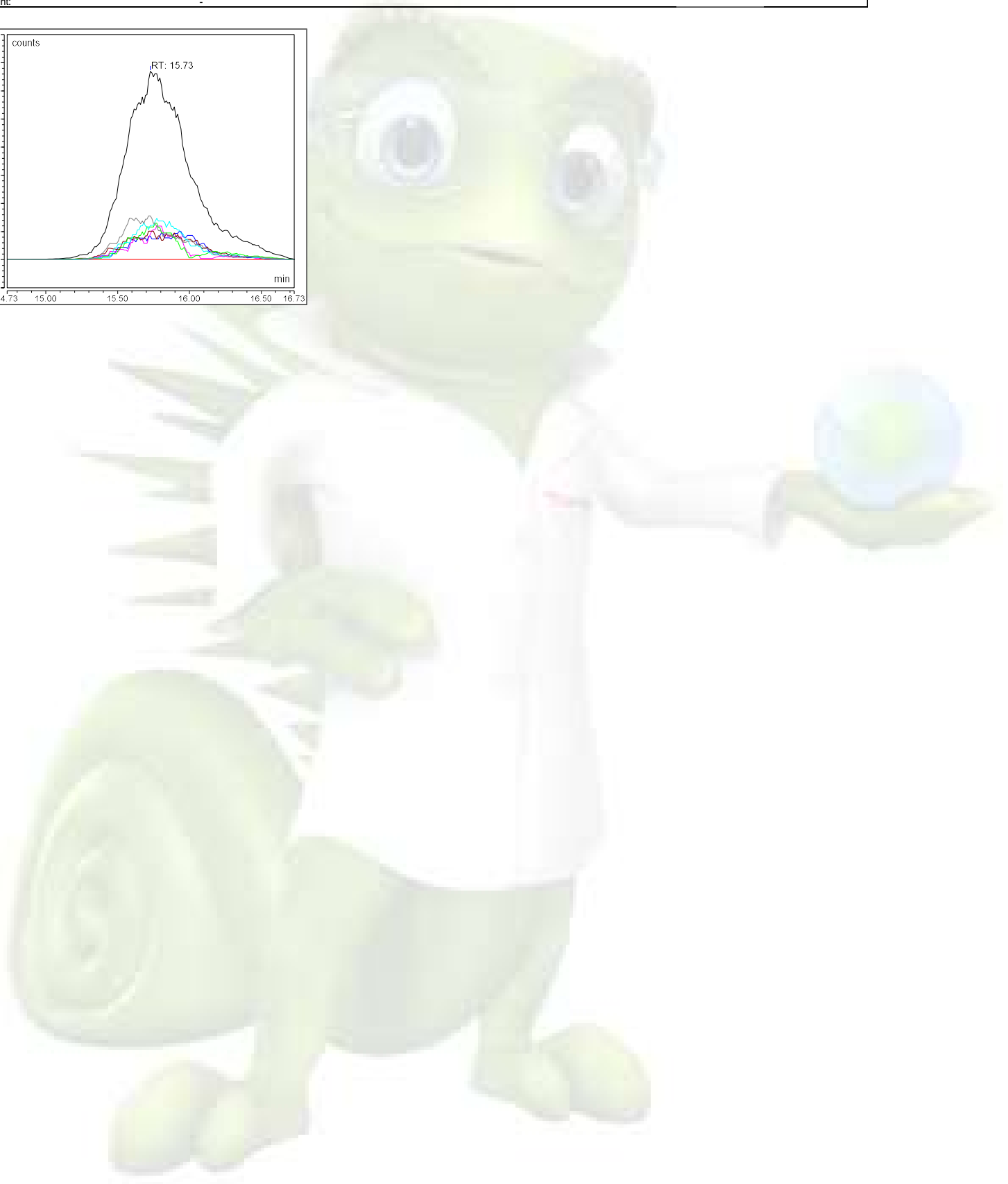
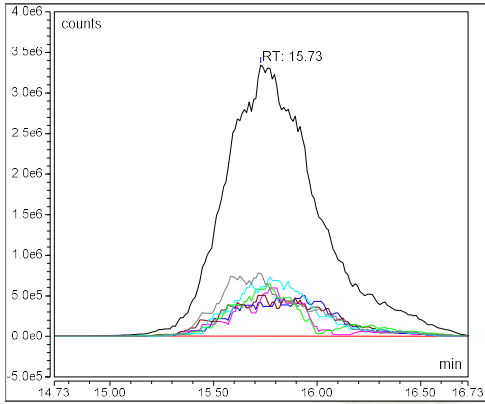
**Component Name: AAV5\_VP3**

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802 |
| Retention time (min): | 15.73   |
| Expected Mass:        | 59804.680   |
| Area (count*min):     | 1734030   |
| Intensity (counts):   | 3342550   |
| Amount:               | -   |



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

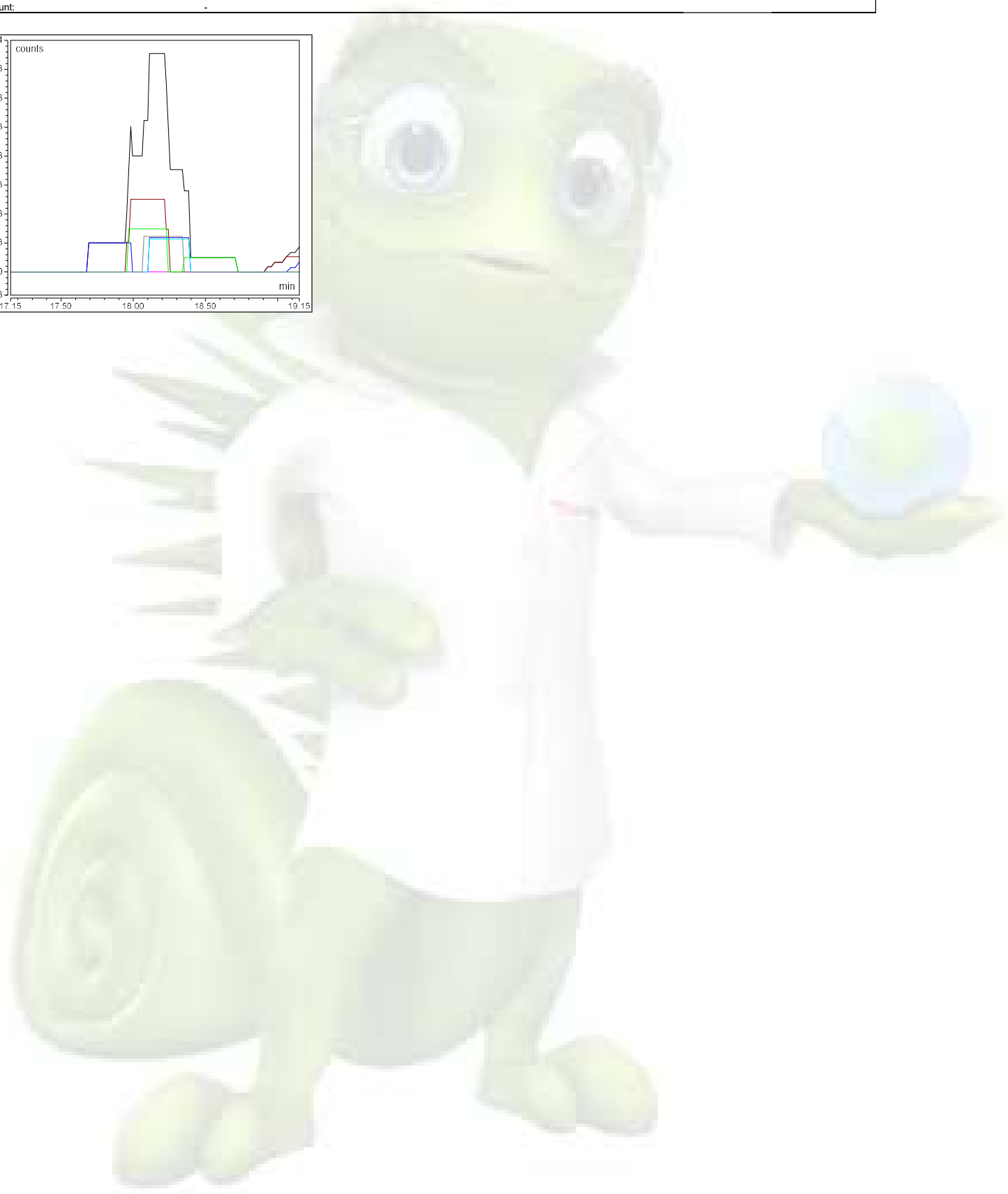
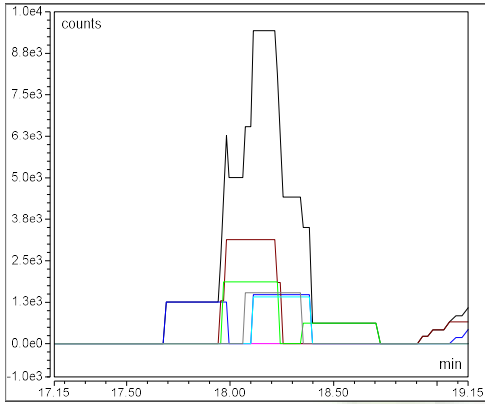
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

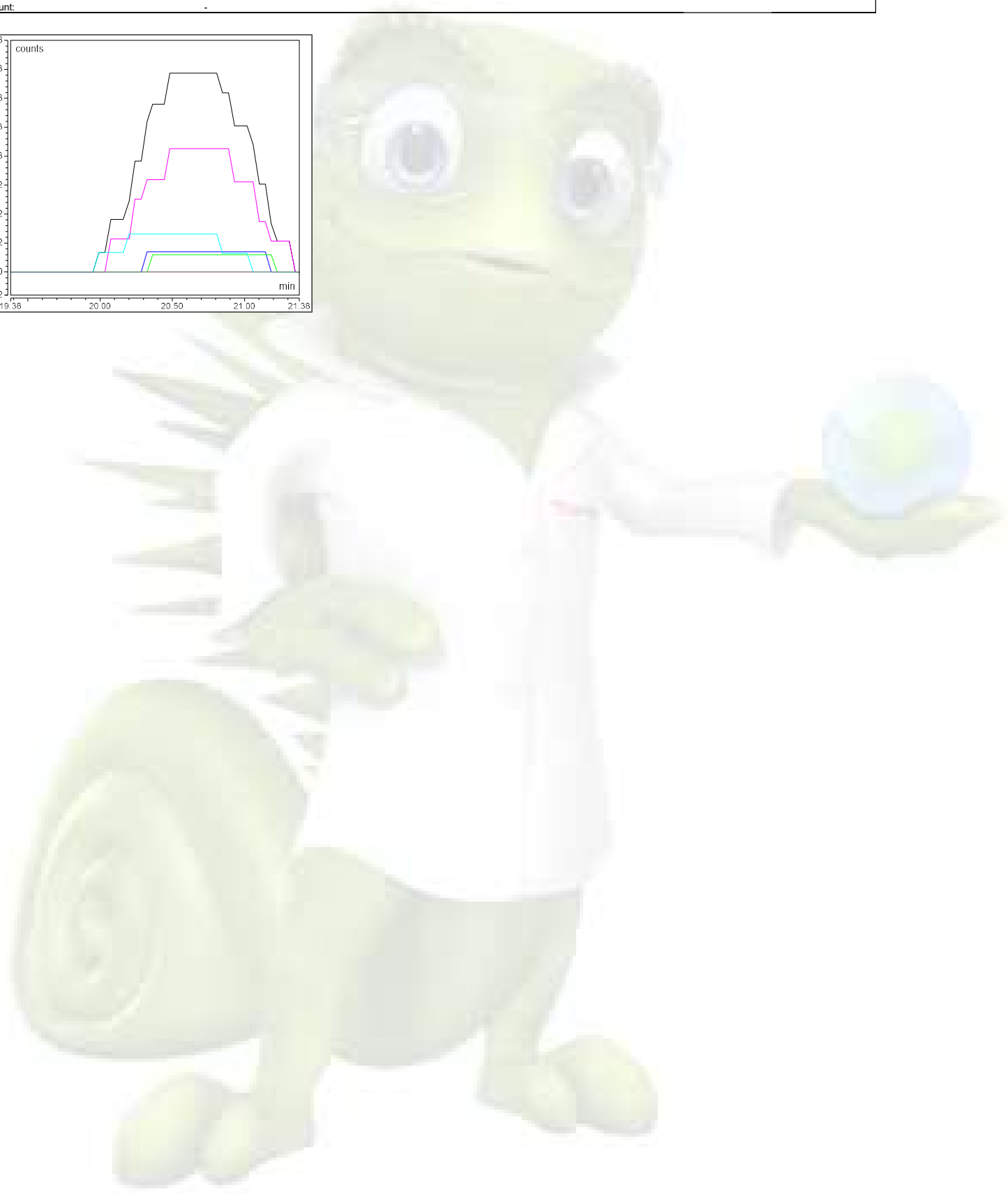
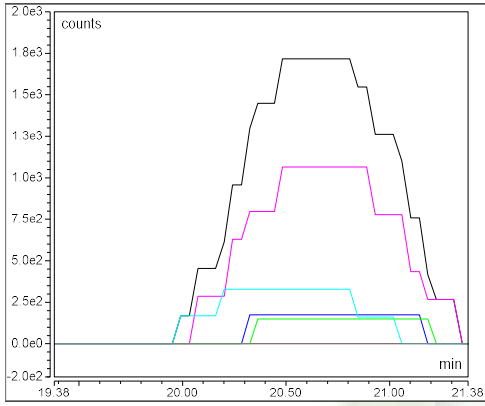
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

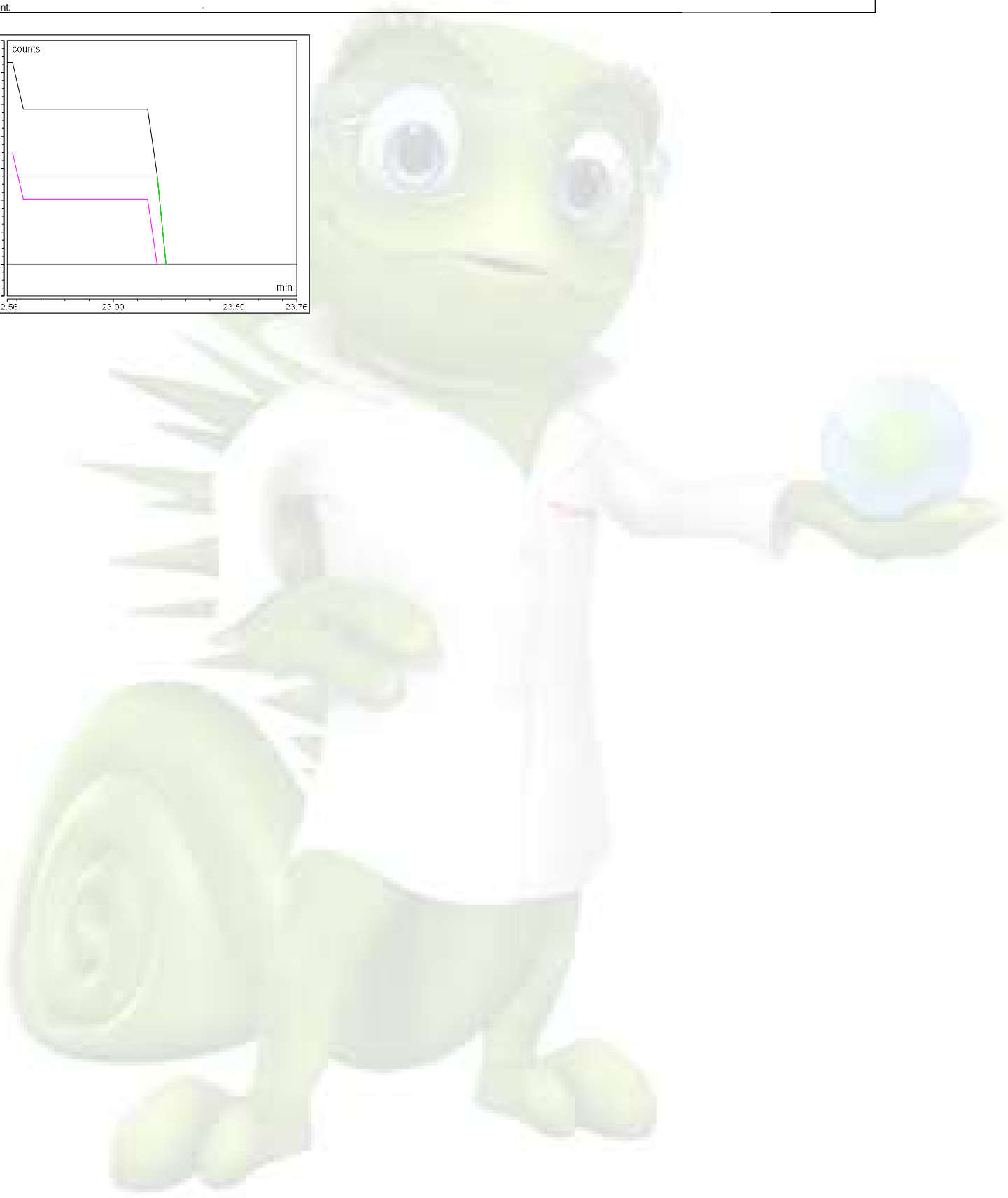
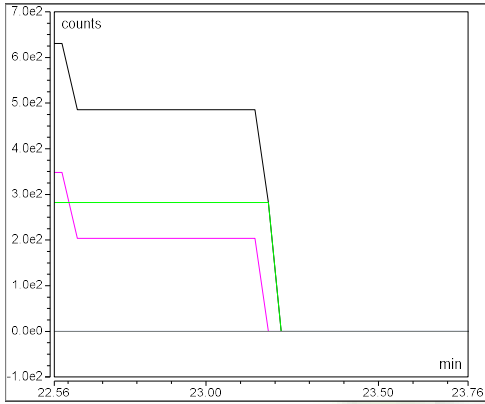
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

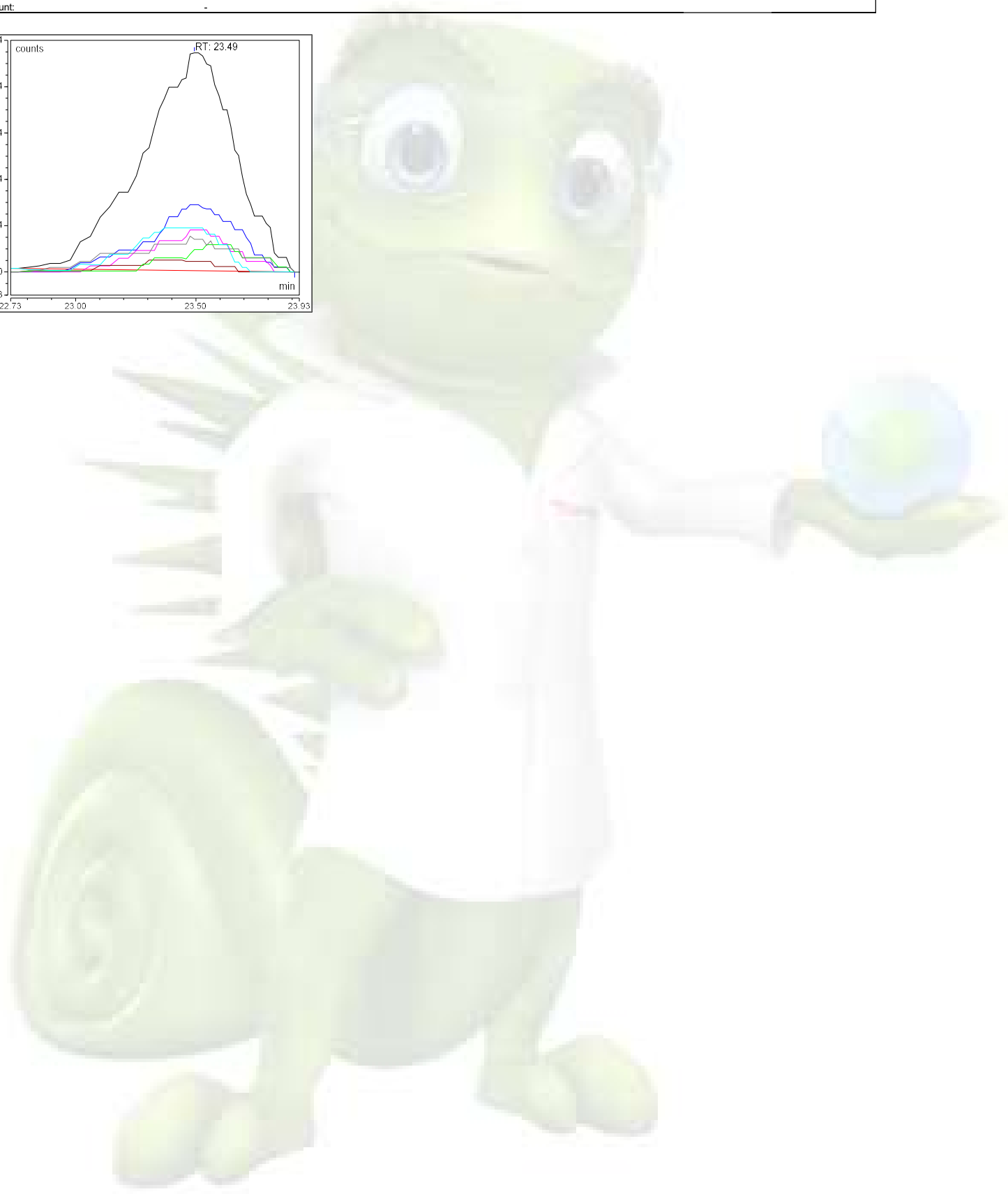
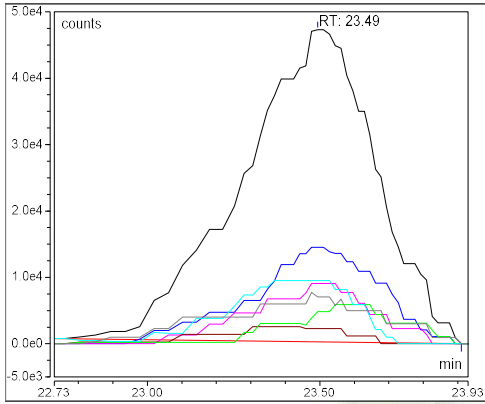
Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828 |
| Retention time (min): | 23.33   |
| Expected Mass:        | 81666.710   |
| Area (count*min):     | 21160   |
| Intensity (counts):   | 47056   |
| Amount:               | -   |



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

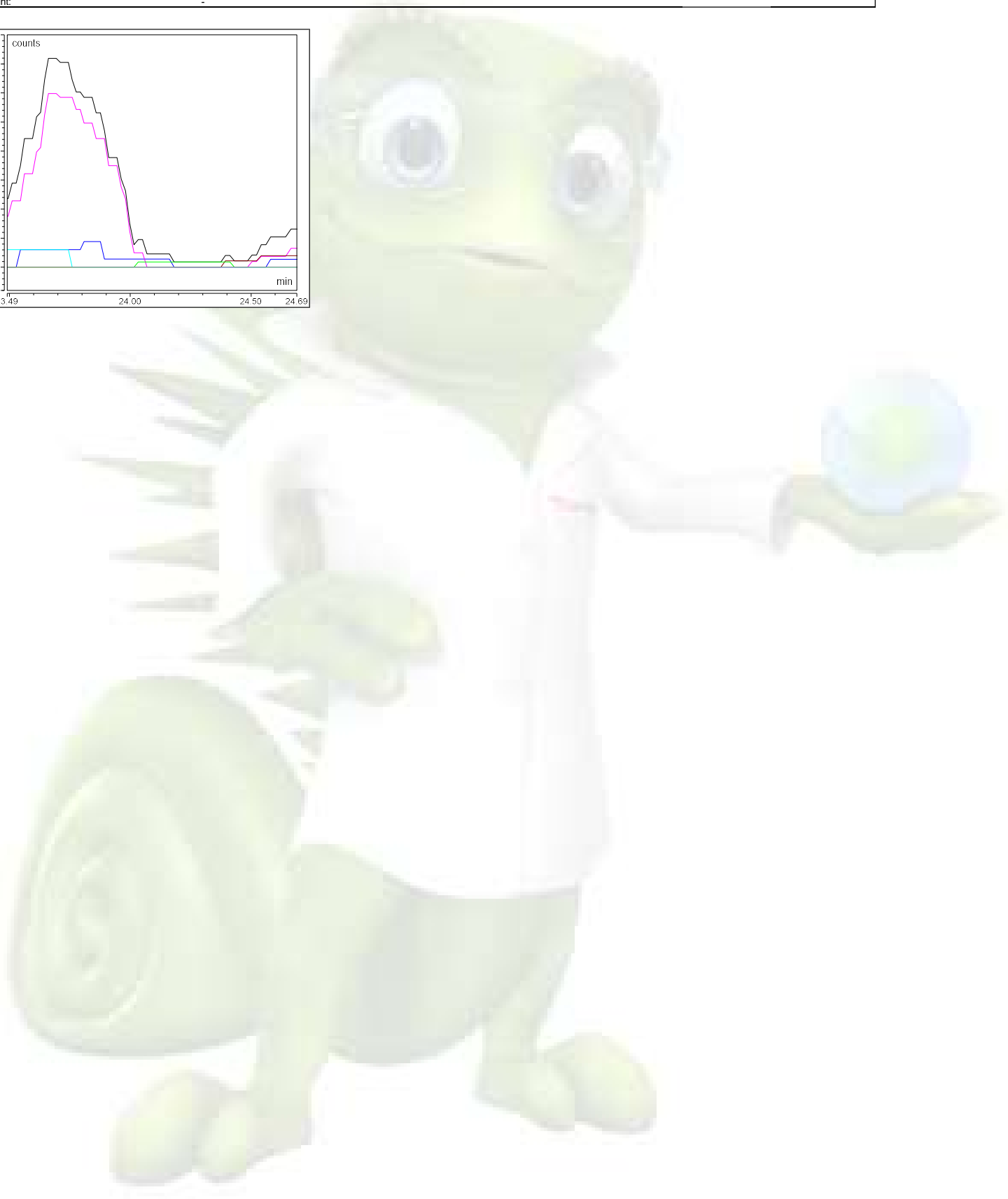
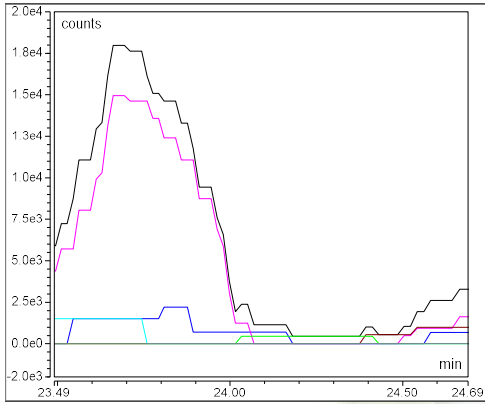
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

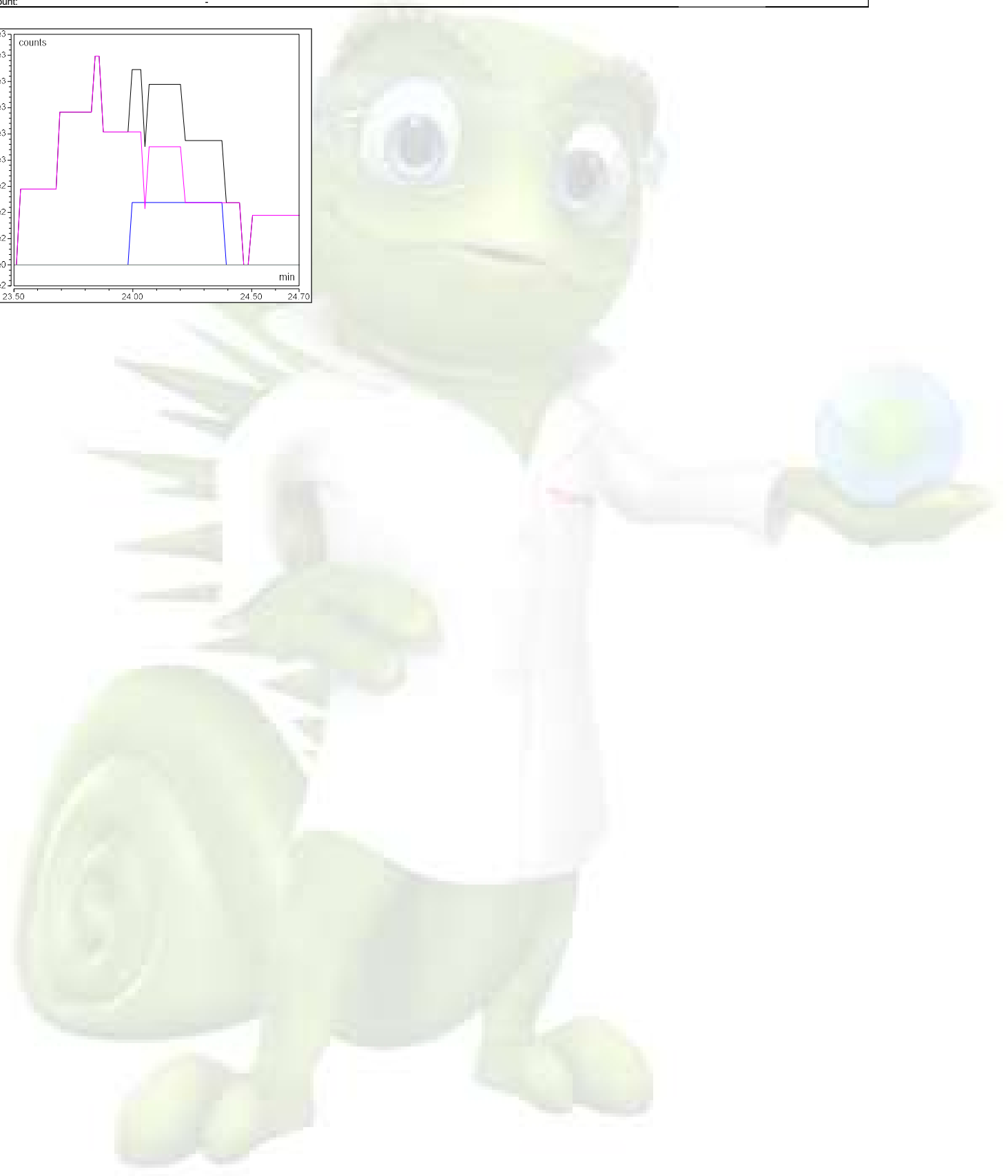
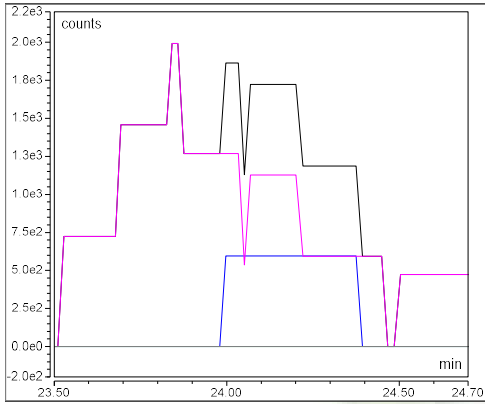
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

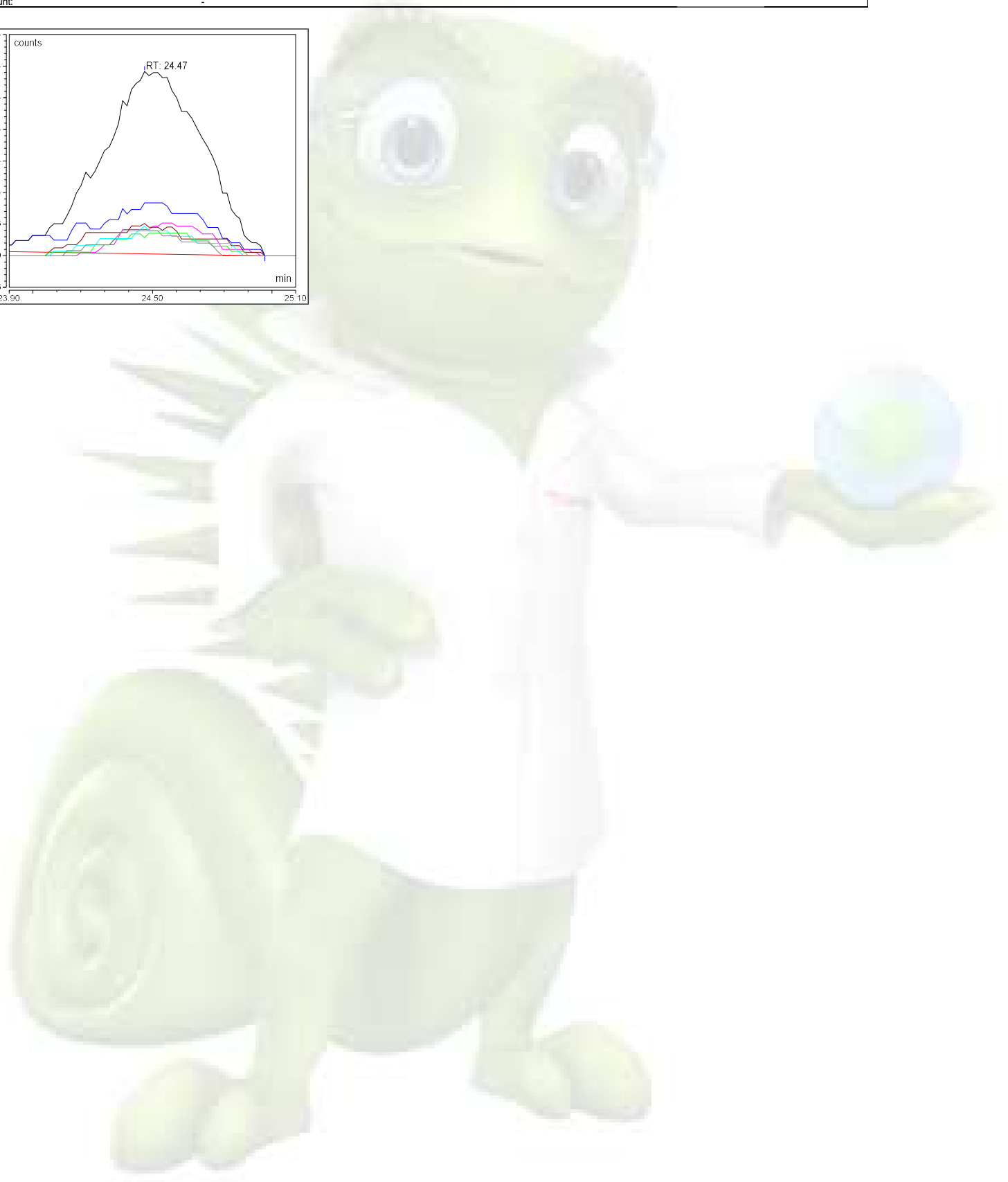
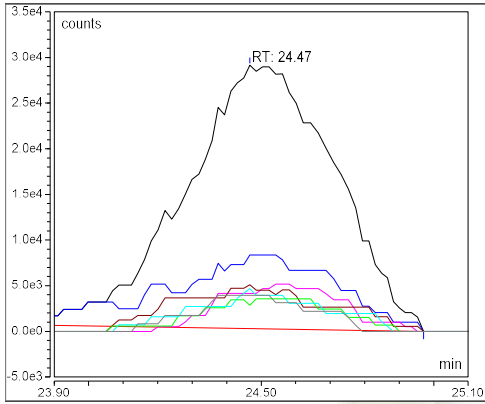
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): 14492

Intensity (counts): 28825

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

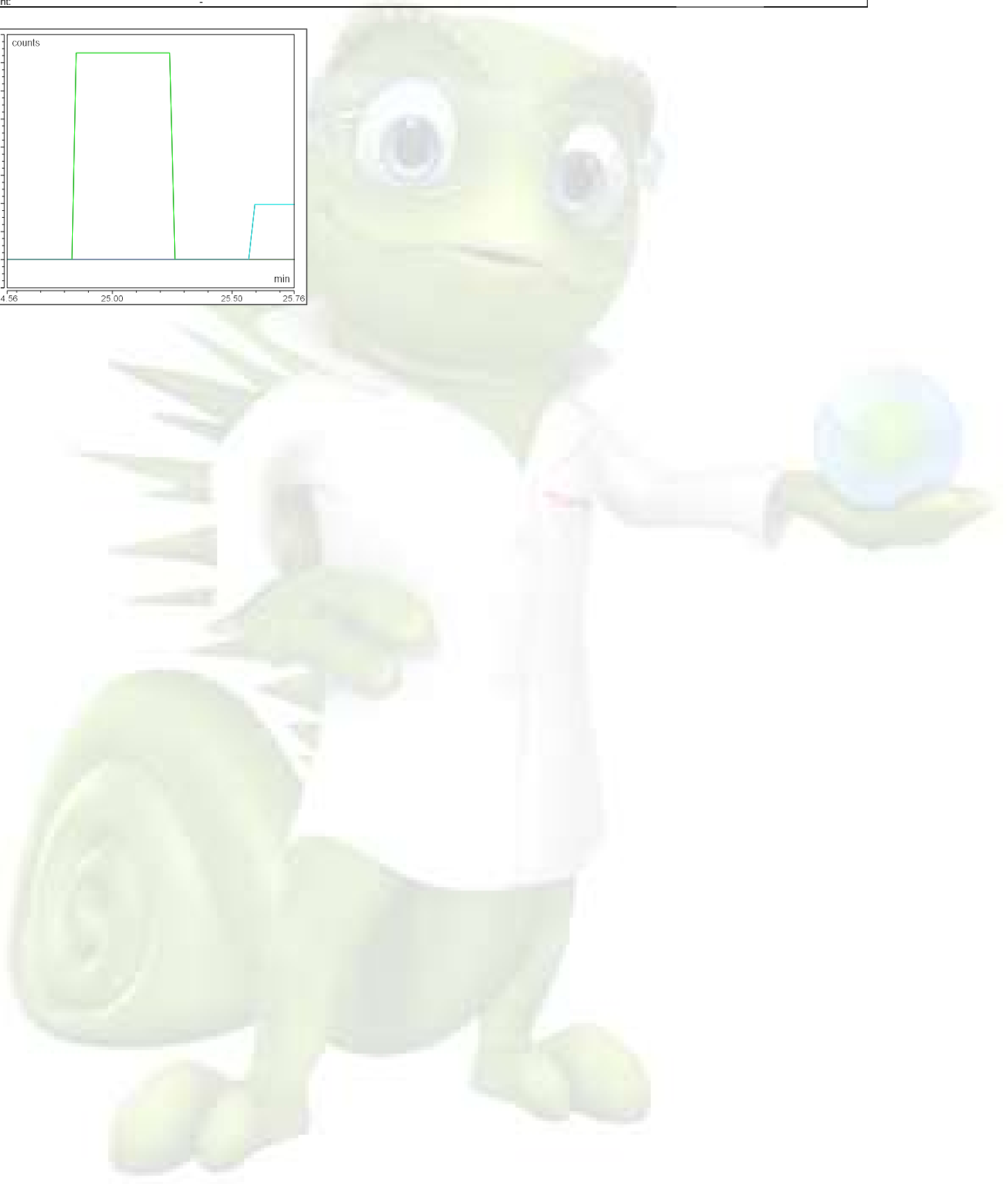
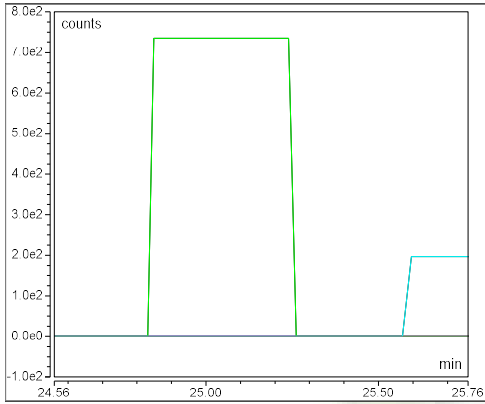
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

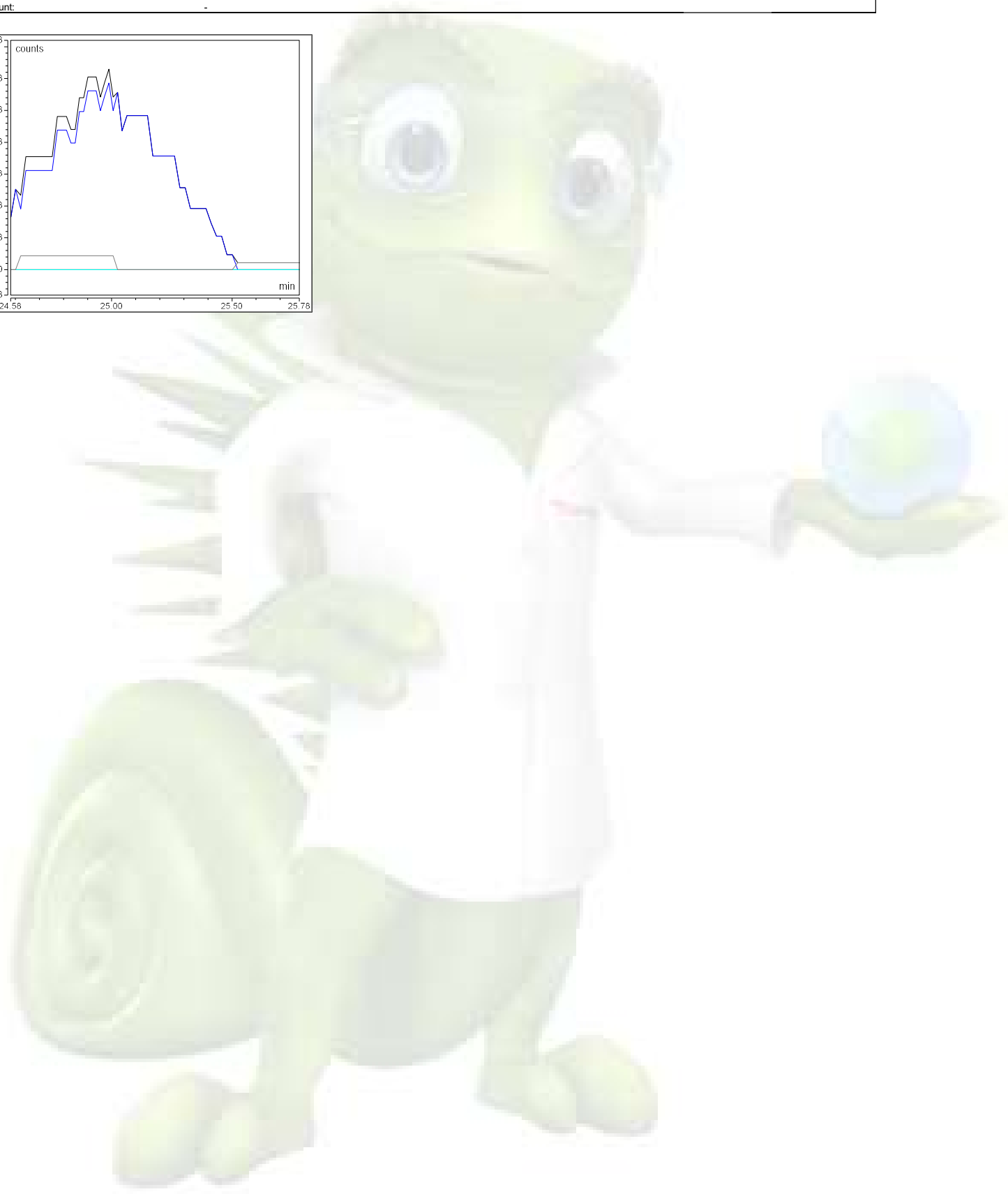
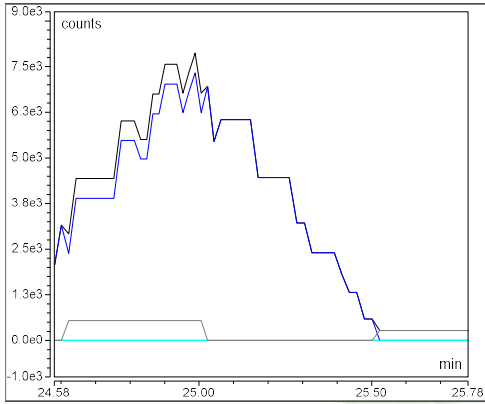
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

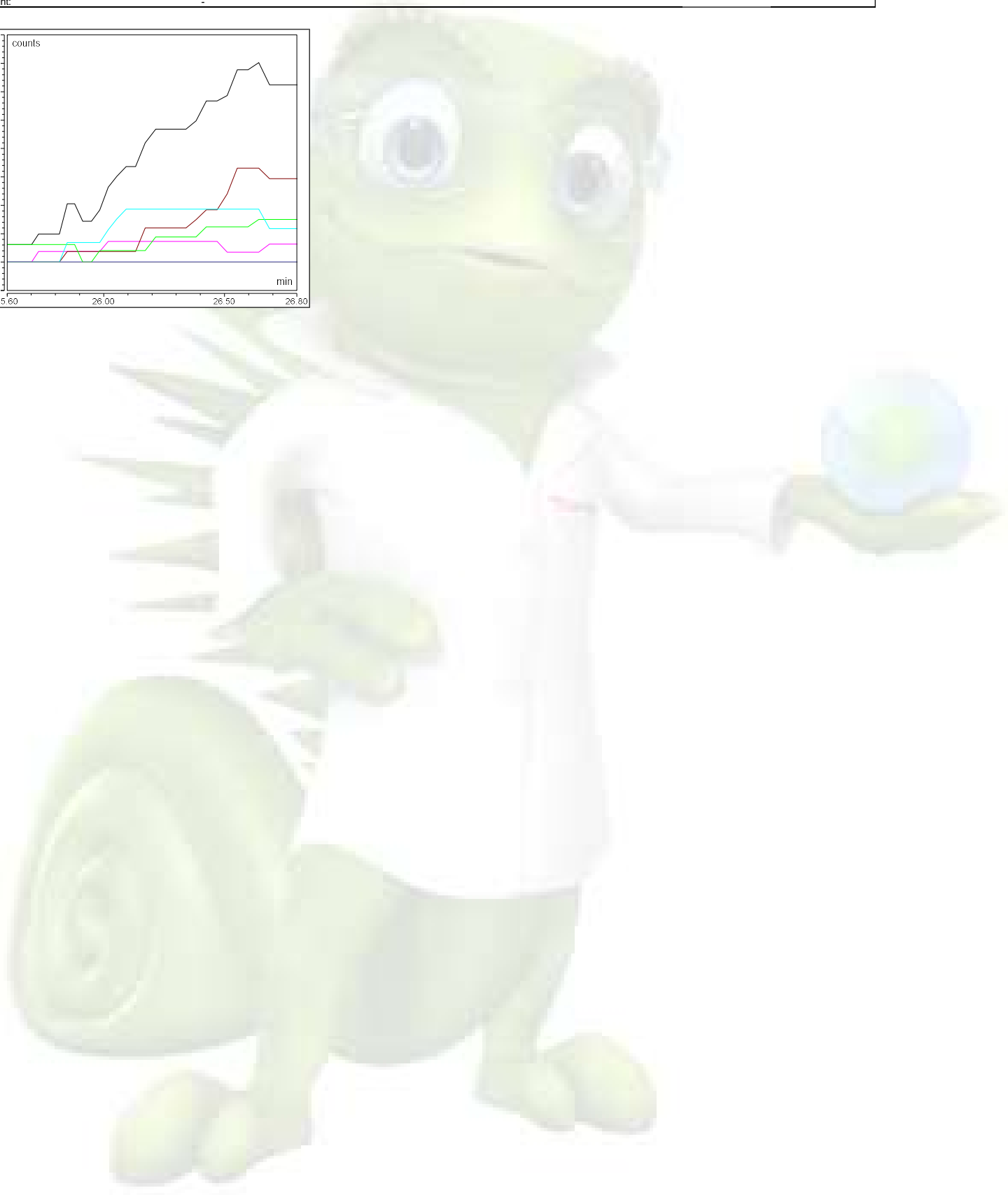
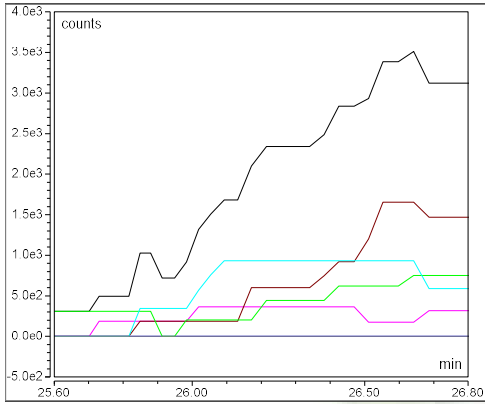
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

Amount: -

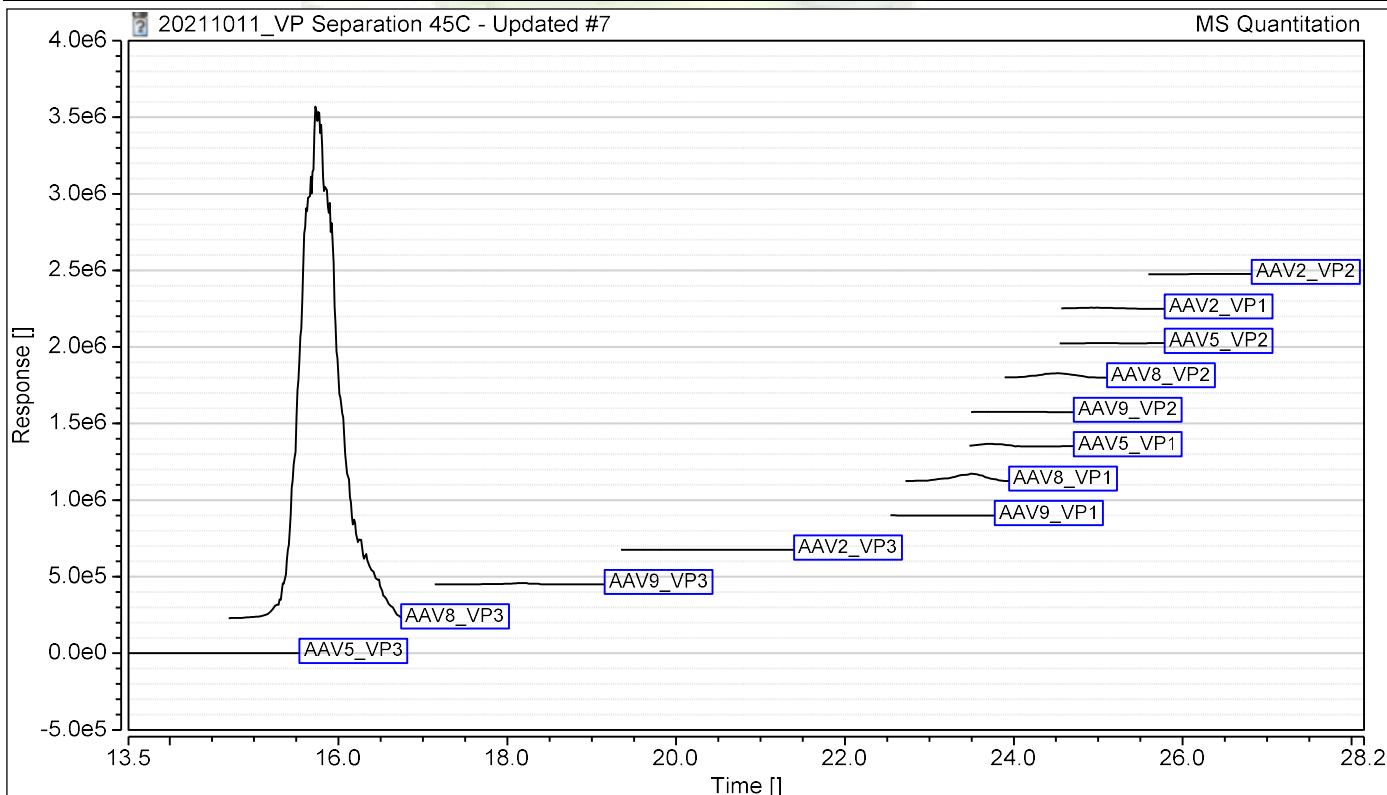


## Chromatogram and SST Results

### Injection Details

|                      |  |                   |        |
|----------------------|--|-------------------|--------|
| Injection Name:      | 20211011_AAV8 E_45C_65-59B_Mod1_1ug_1  | Run Time (min):   | 45.00  |
| Vial Number:         | R:A3   | Injection Volume: | 6.25   |
| Injection Type:      | Unknown  |                   |        |
| Calibration Level:   |  |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                   |        |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 13/Oct/21 01:42  | Sample Weight:    | 1.0000 |

### Chromatogram

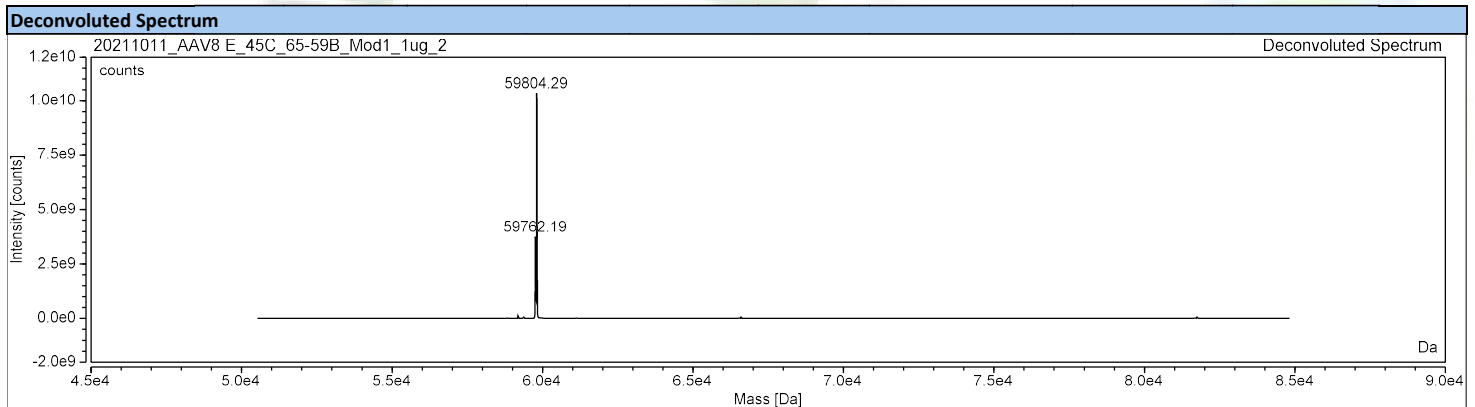
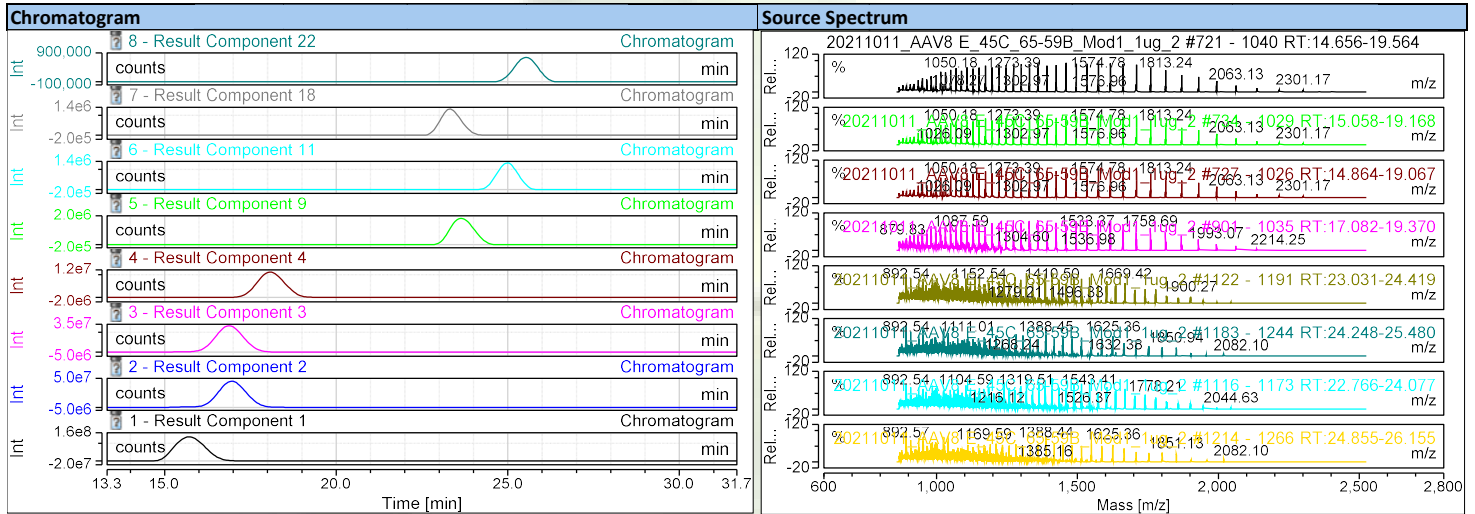


### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |                                       |                            |
|-------------------|---------------------------------------|----------------------------|
| Injection Name:   | 20211011_AAV8 E_45C_65-59B_Mod1_1ug_2 | Injection Volume:          |
| Vial Number:      | R:A3                                  | Instrument Method:         |
| Injection Type:   | Unknown                               | Processing Method:         |
| Run Time (min):   | 45.00                                 | Injection Date/Time:       |
|                   |                                       | MS Quantitative_45C_65-59B |
|                   |                                       | 13/Oct/21 02:28            |



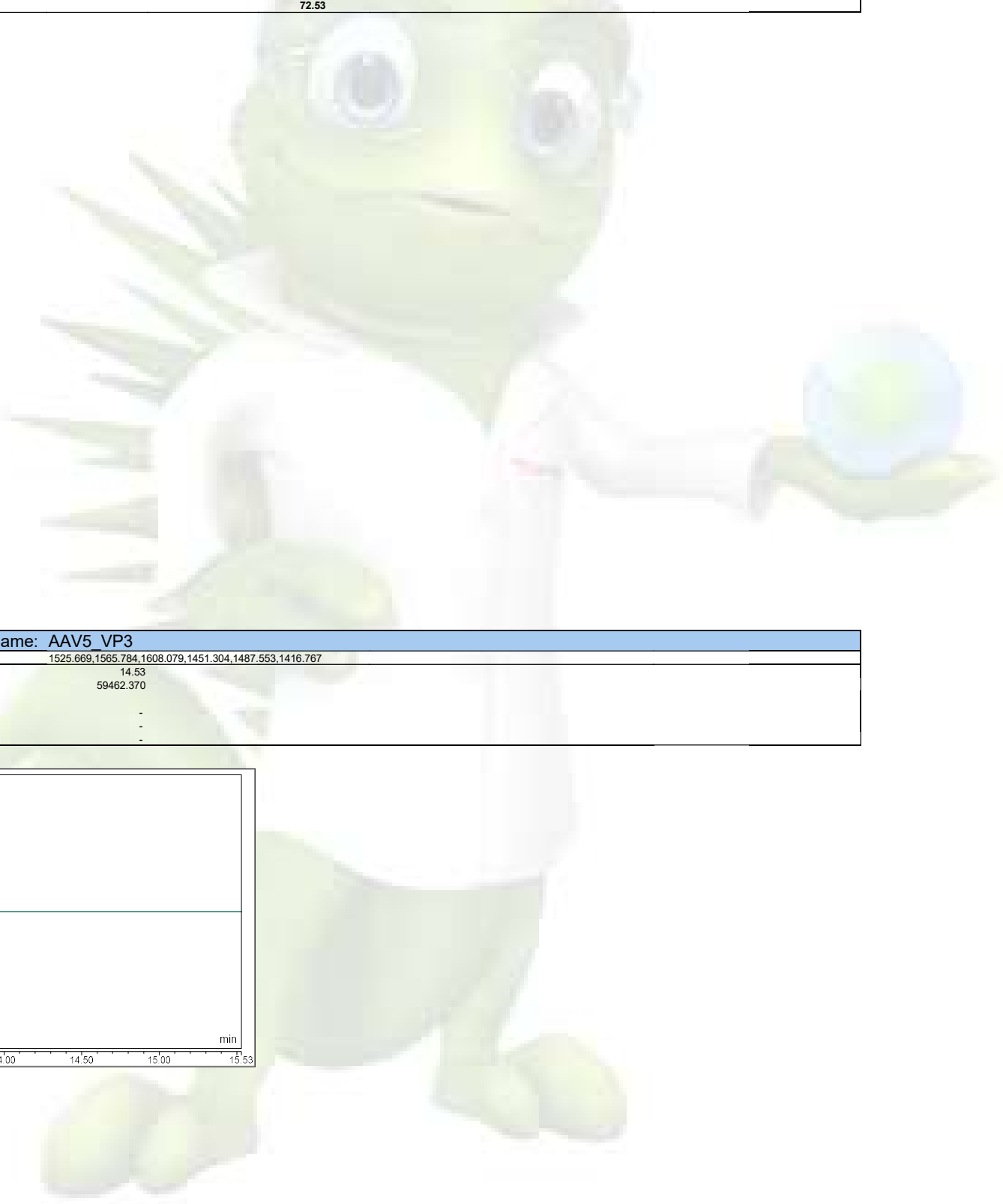
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

## Component Summary

### Injection Details

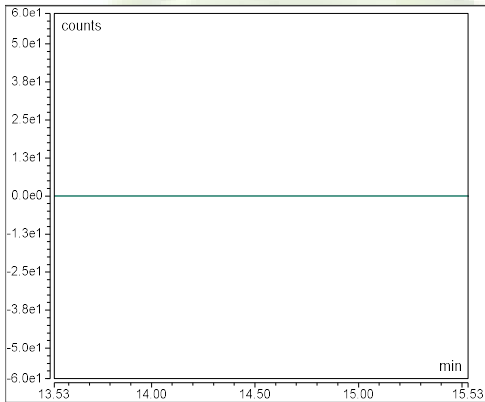
|  |   |  |
|--|---|--|
| <i>Injection Name:</i> 20211011_AAV8_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i> 6.25   |  |
| <i>Vial Number:</i> R:A3                                   | <i>Instrument Method:</i> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <i>Injection Type:</i> Unknown                             | <i>Processing Method:</i> MS Quantitative_45C_65-59B                            |  |
| <i>Run Time (min):</i> 45.00                               | <i>Injection Date/Time:</i> 13/Oct/21 02:28                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV8_VP3     | 15.698                | 1326273.881        | 71.23              | 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802 | 59804.680 | PASS                           |
| AAV8_VP1     | 23.490                | 13385.770          | 0.72               | 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828 | 81666.710 | PASS                           |
| AAV8_VP2     | 24.474                | 10871.521          | 0.58               | 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397 | 66518.100 | PASS                           |
| <b>72.53</b> |                       |                    |                    |   |           |                                |



### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

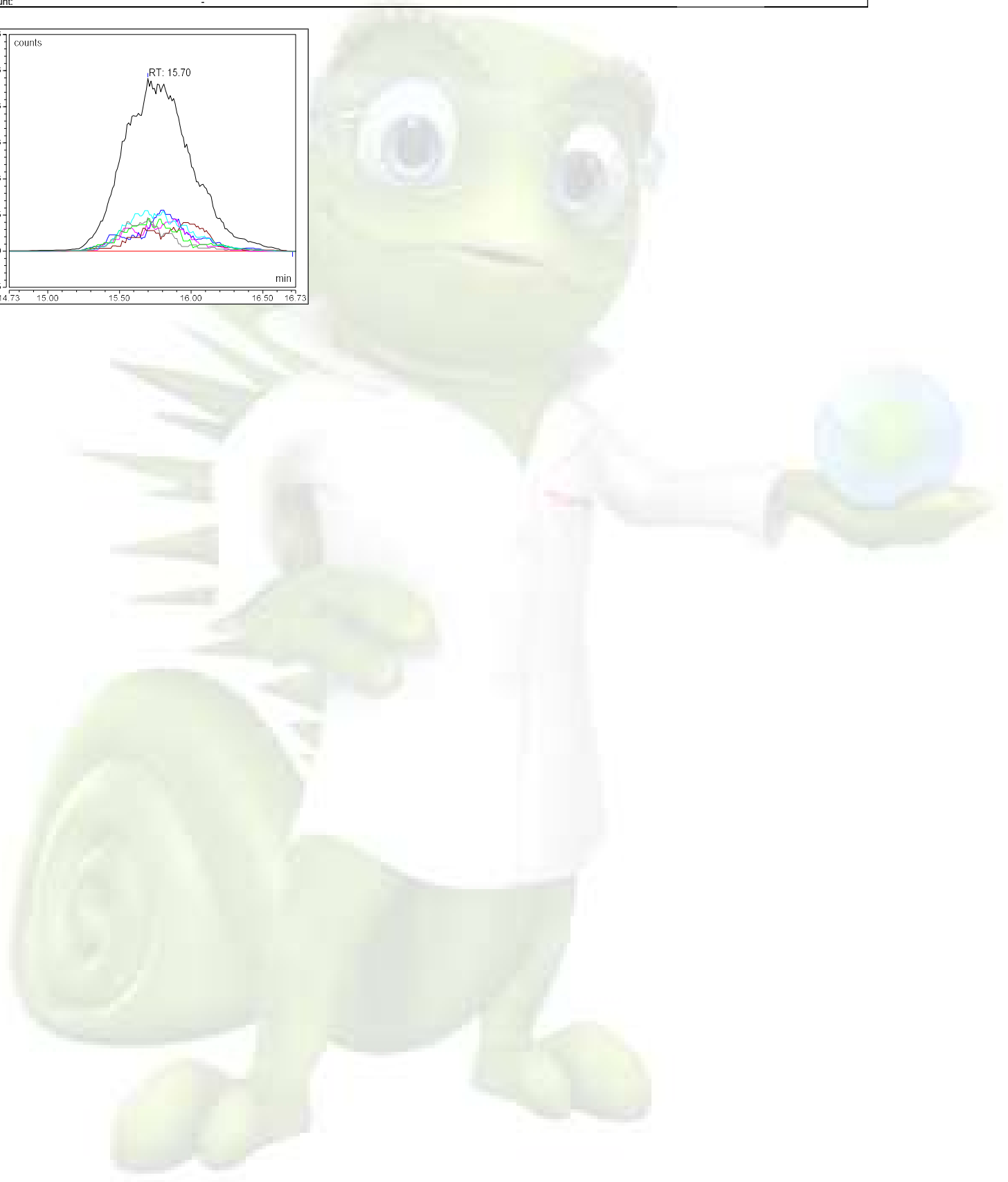
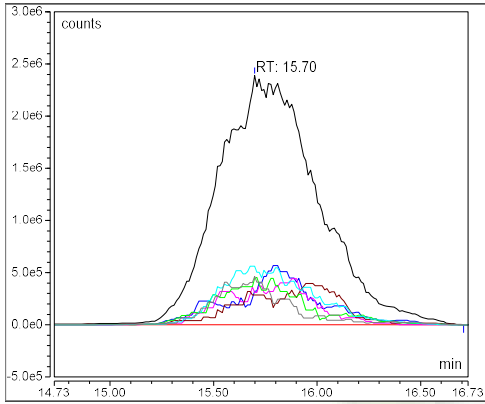
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): 1326274

Intensity (counts): 2388981

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

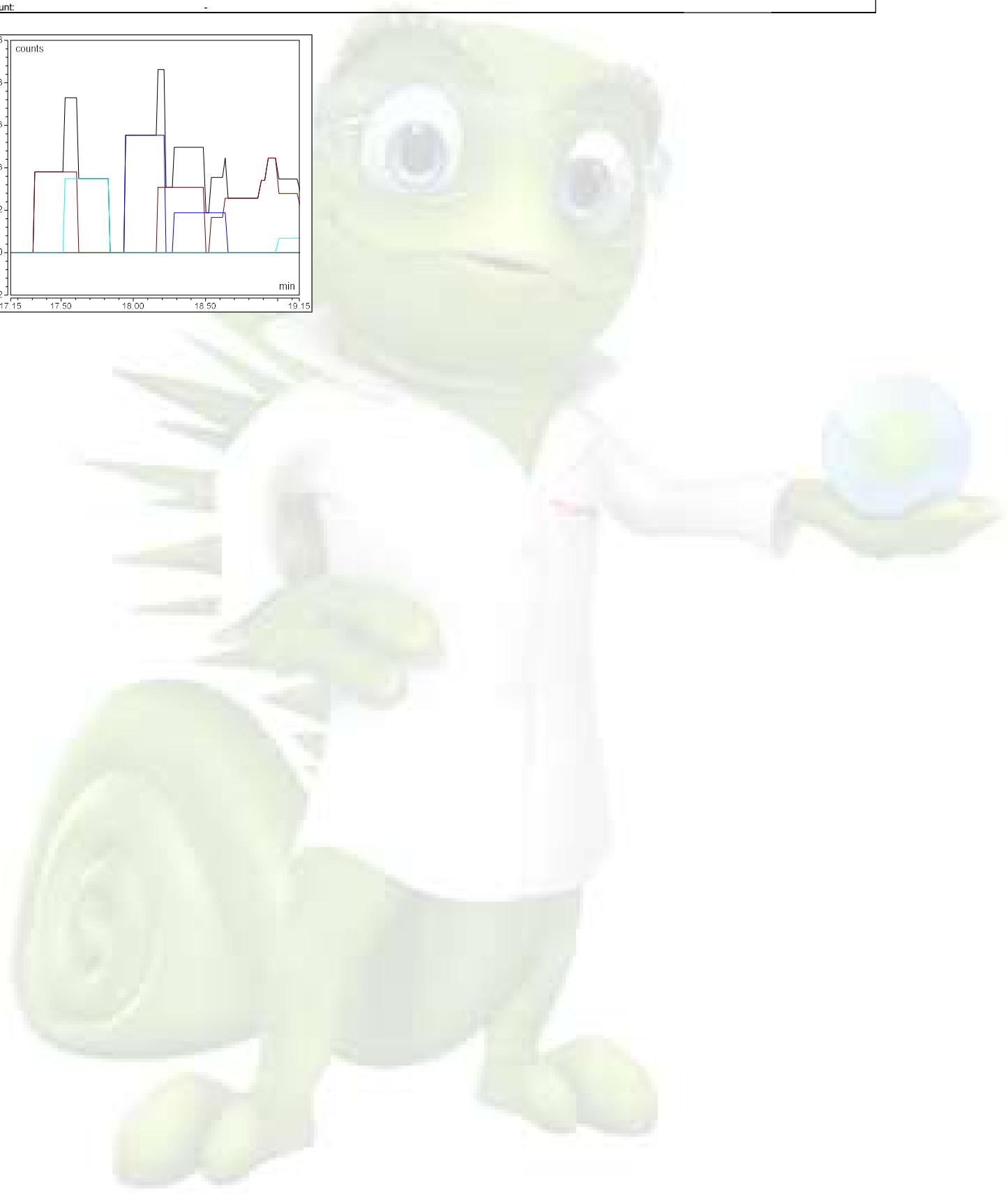
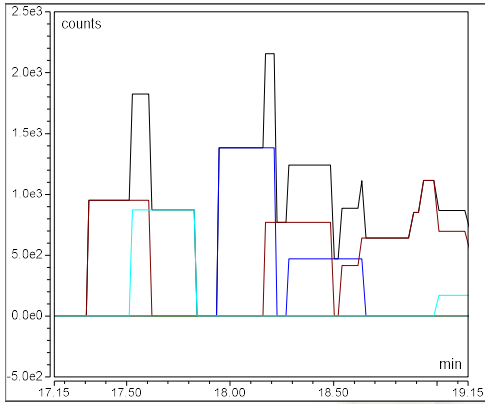
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

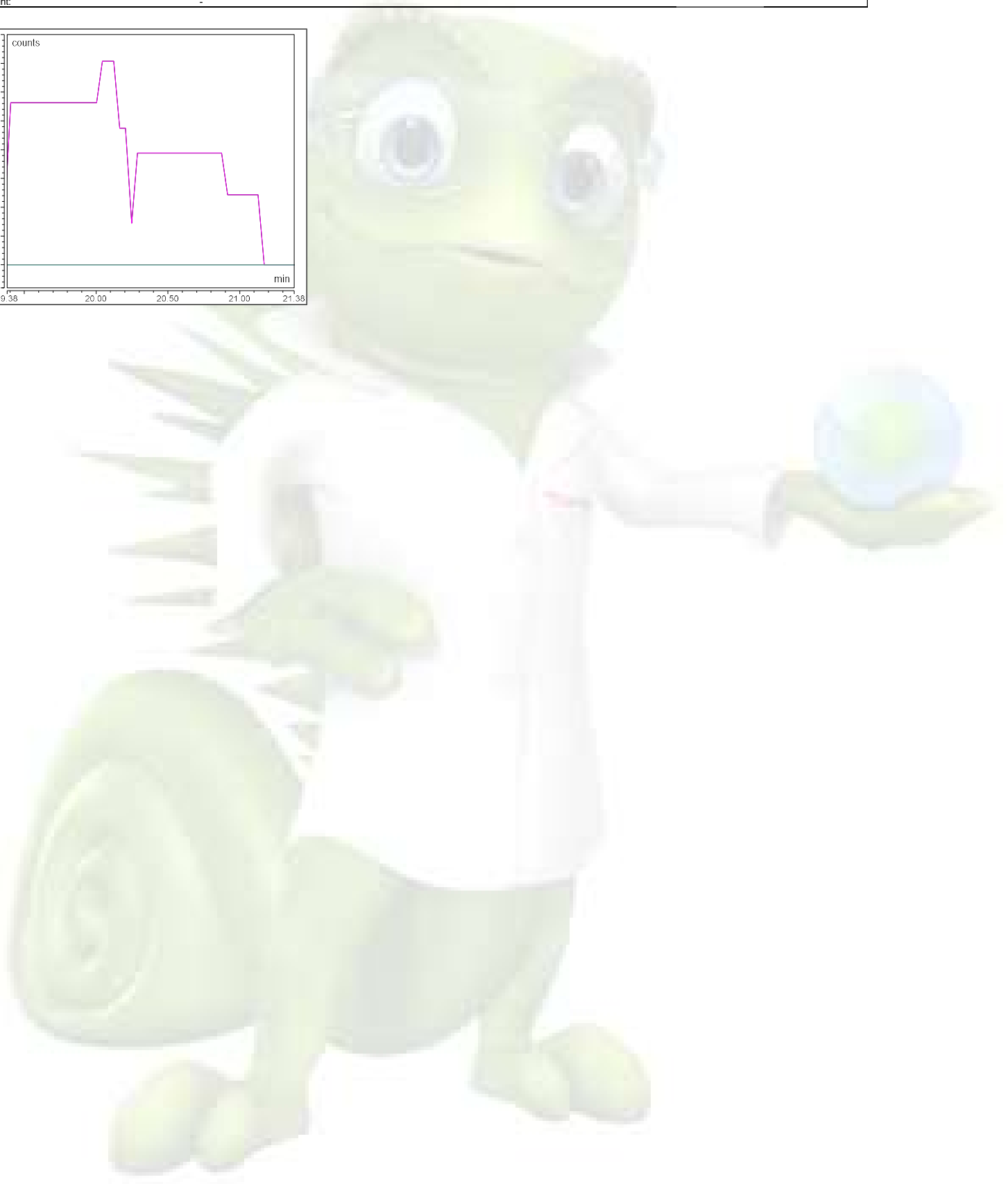
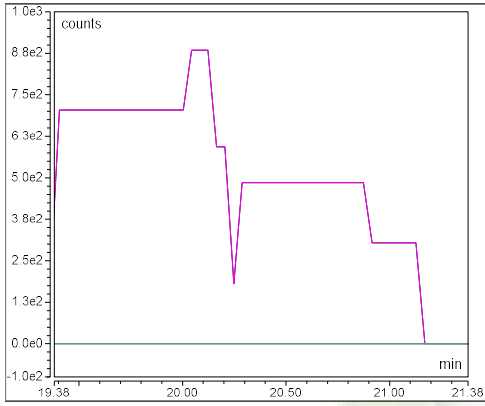
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

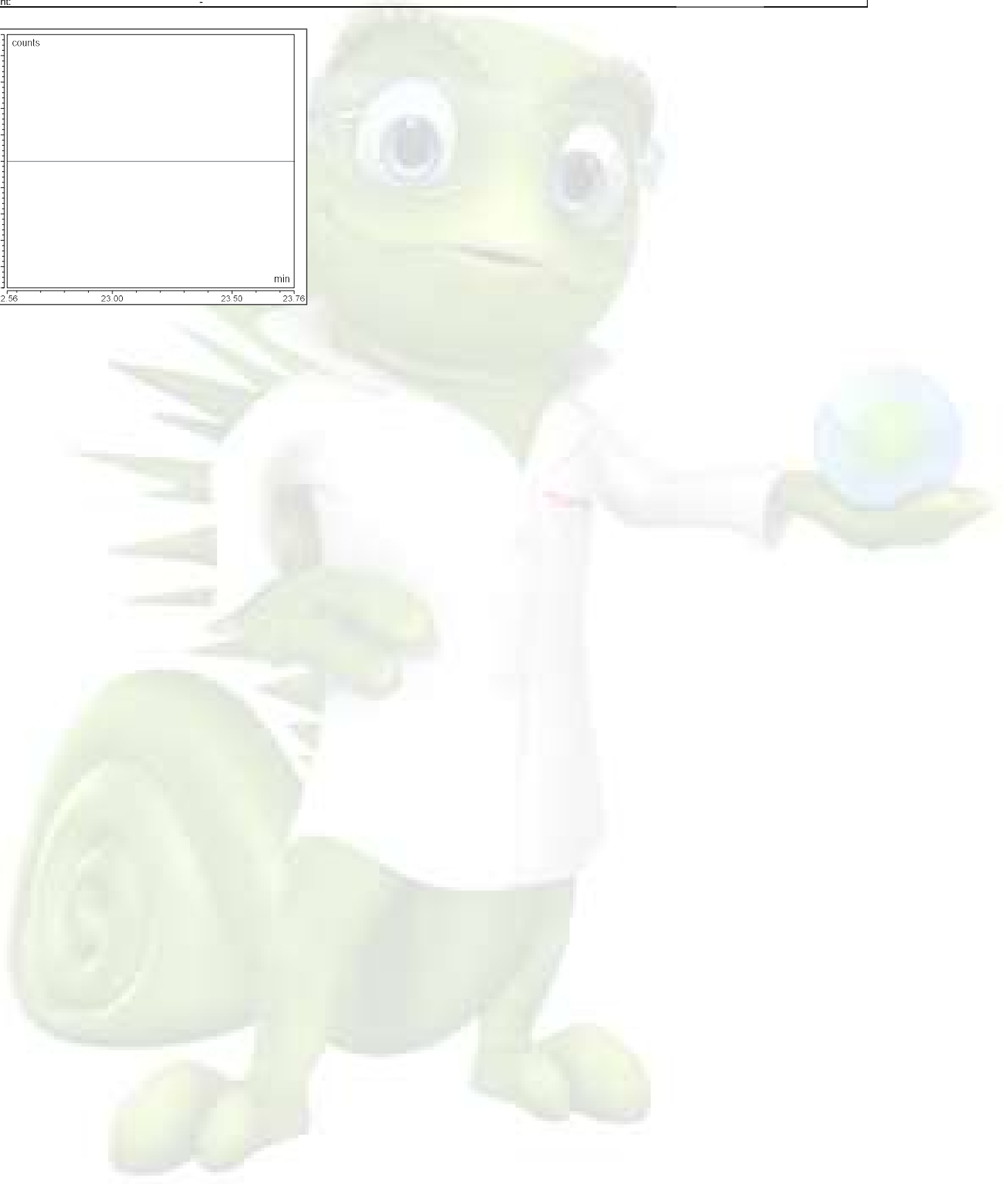
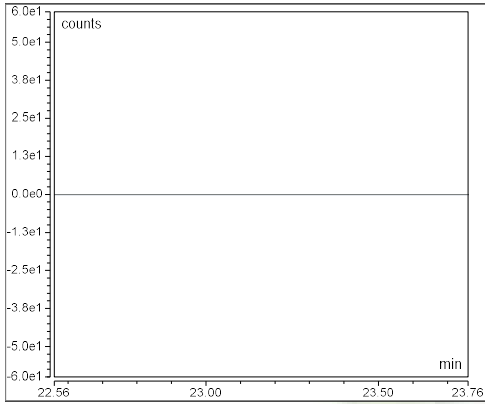
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

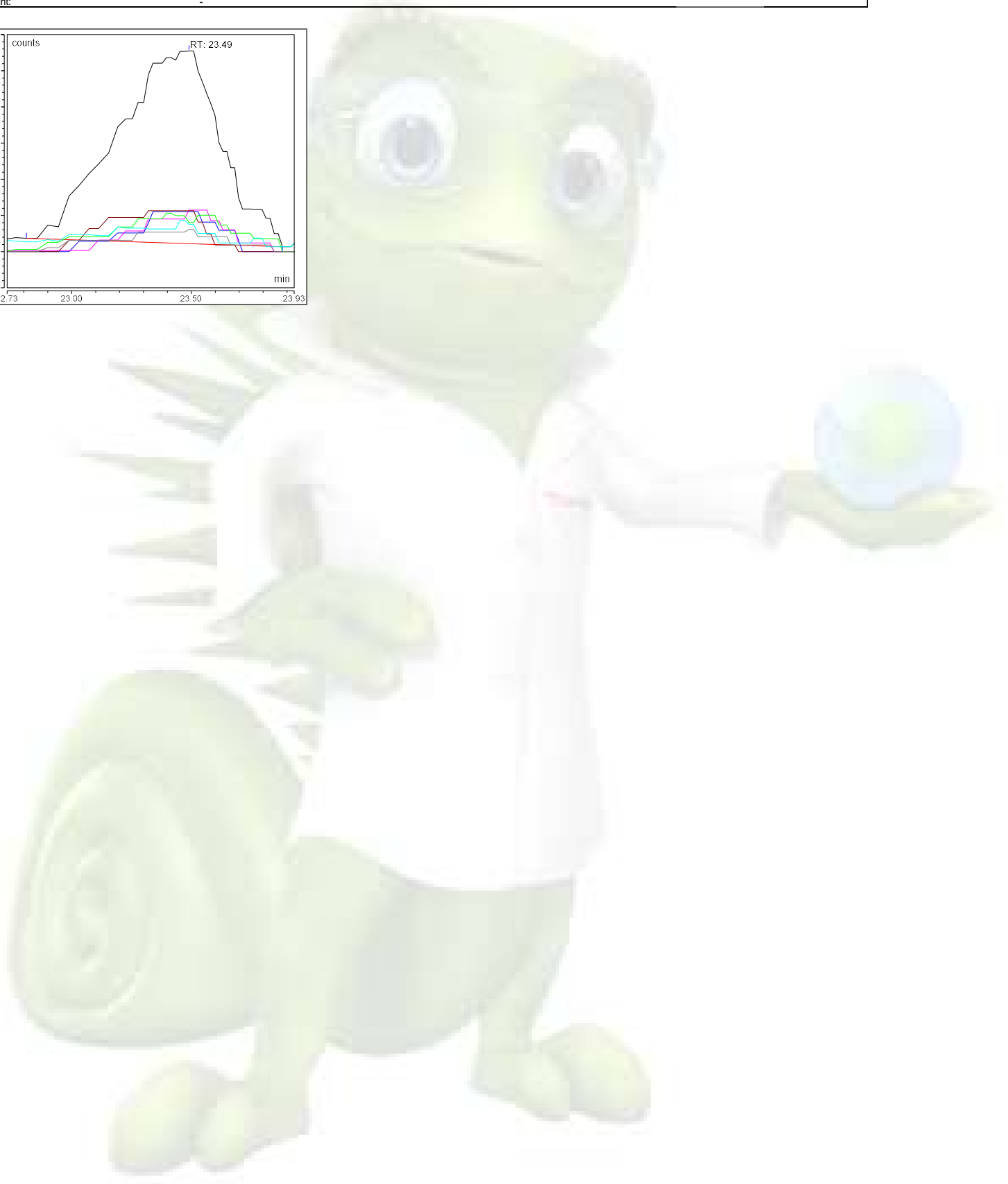
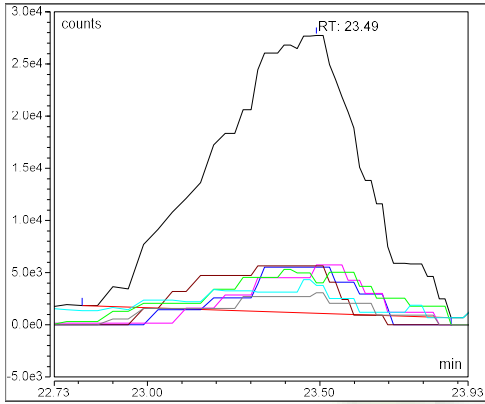
Amount: -





**Component Name: AAV8 VP1**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828 |
| Retention time (min): | 23.33   |
| Expected Mass:        | 81666.710   |
| Area (count*min):     | 13386   |
| Intensity (counts):   | 26629   |
| Amount:               | -   |



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

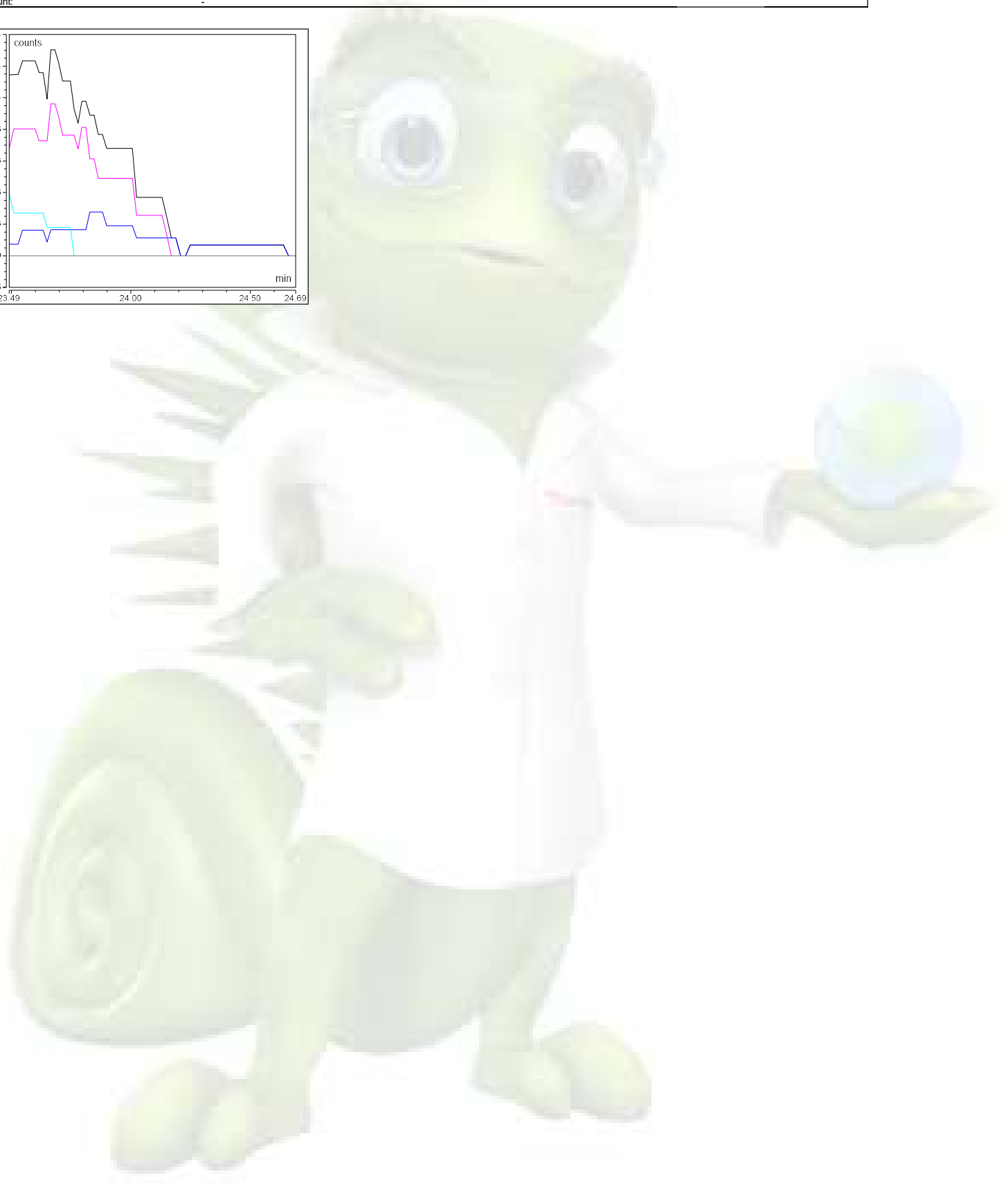
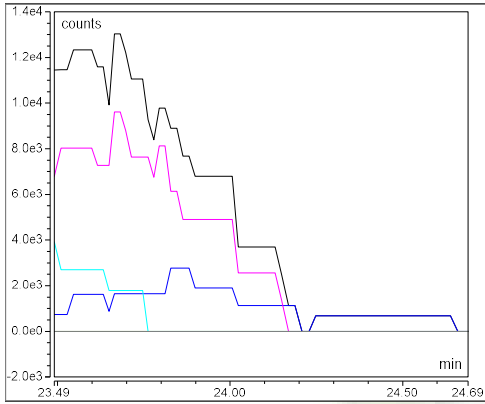
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -

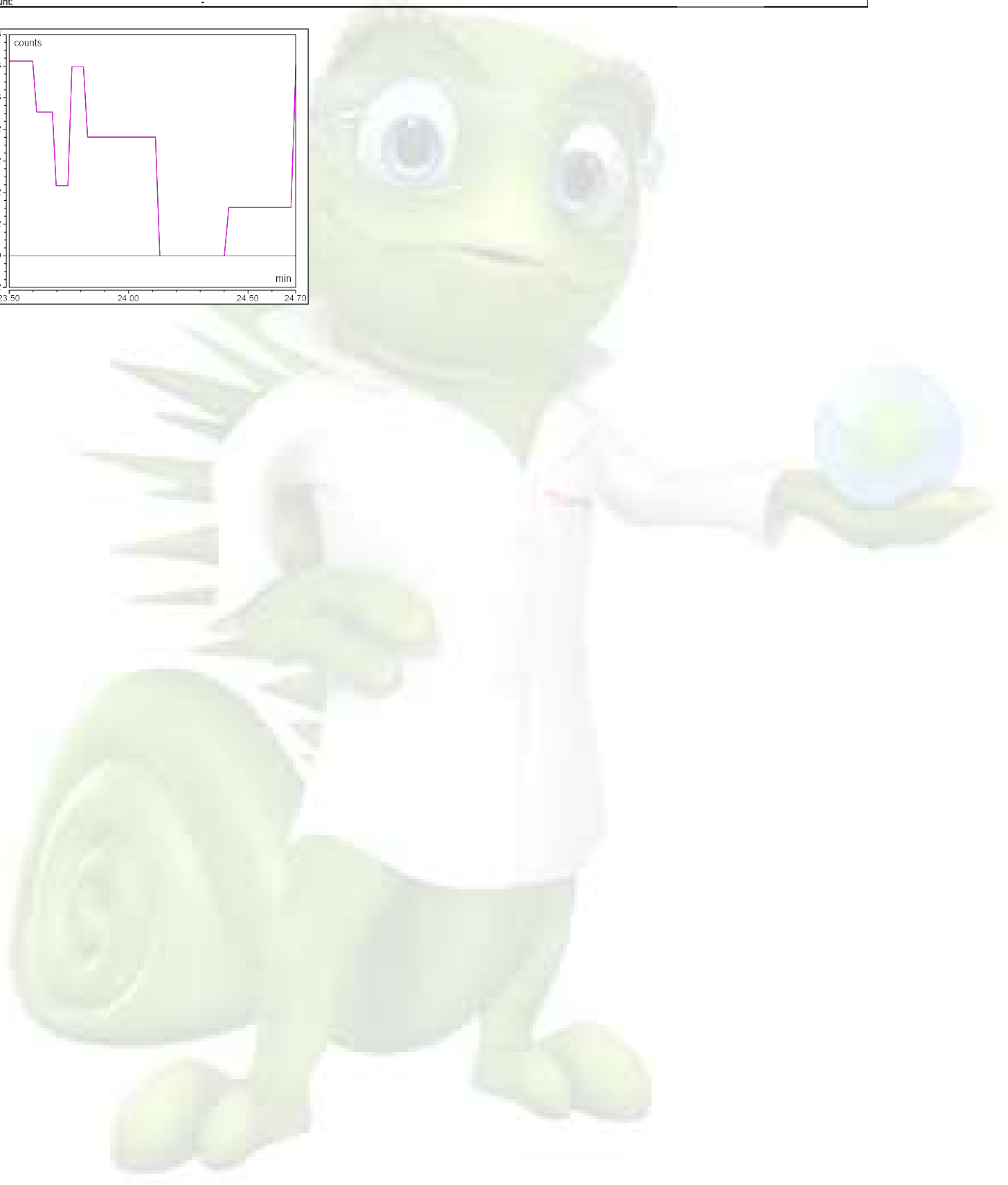
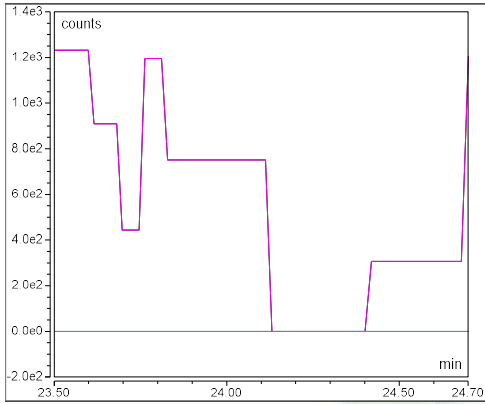


**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

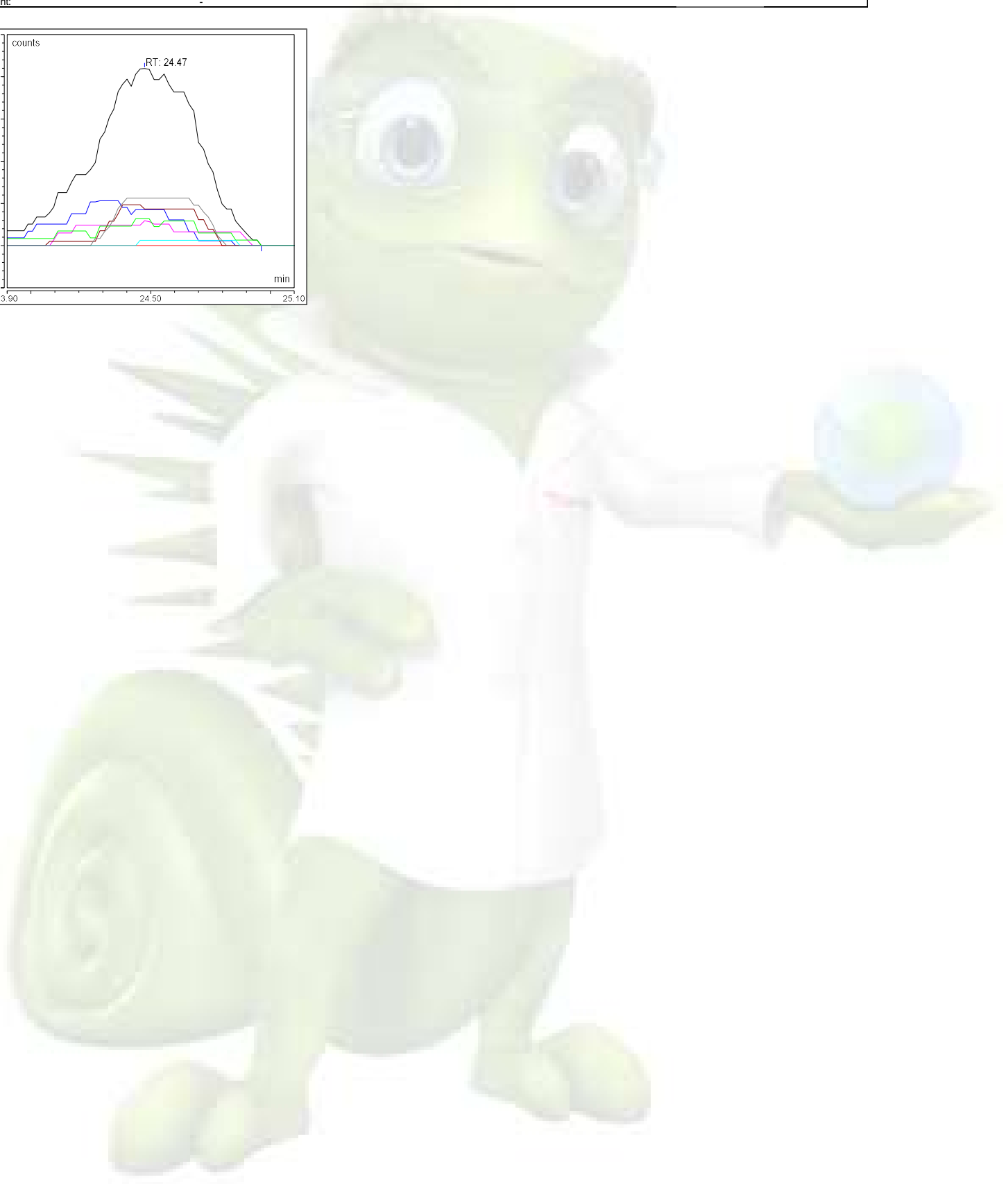
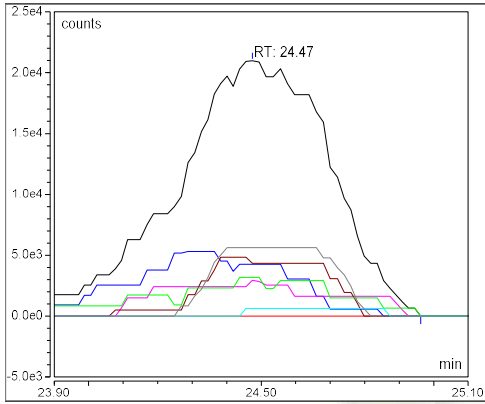
Retention time (min): 24.10  
Expected Mass: 66209.760

Area (count\*min): -  
Intensity (counts): -  
Amount: -



**Component Name: AAV8 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397 |
| Retention time (min): | 24.50   |
| Expected Mass:        | 66518.100   |
| Area (count*min):     | 10872   |
| Intensity (counts):   | 20978   |
| Amount:               | -   |



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

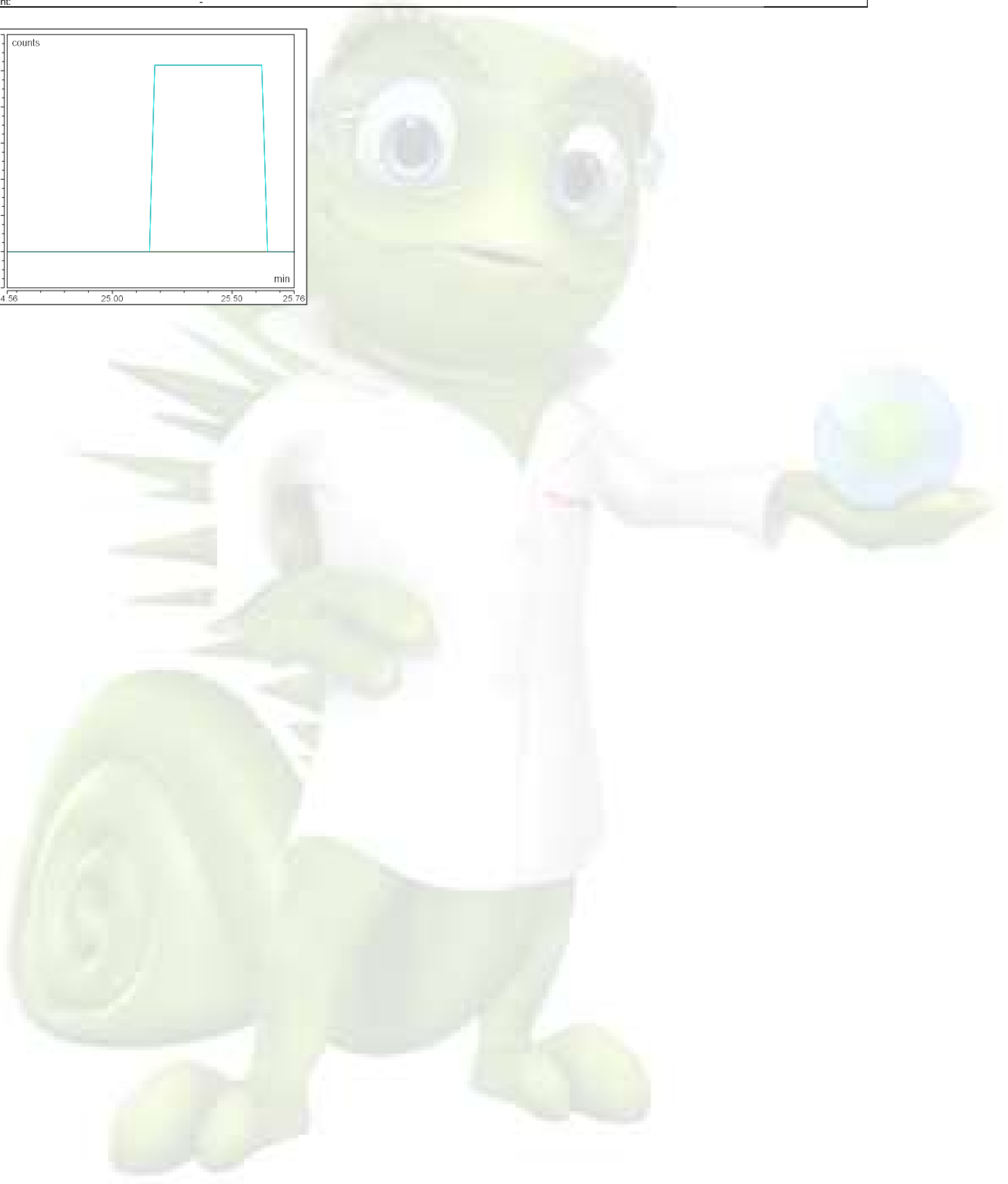
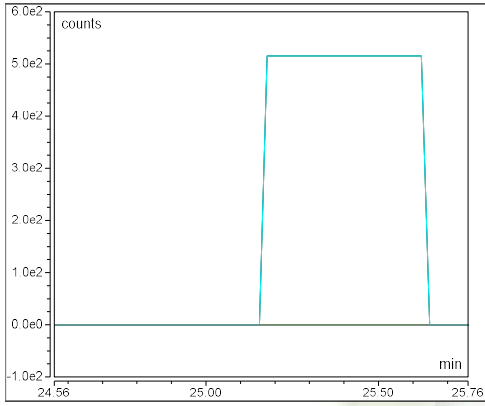
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

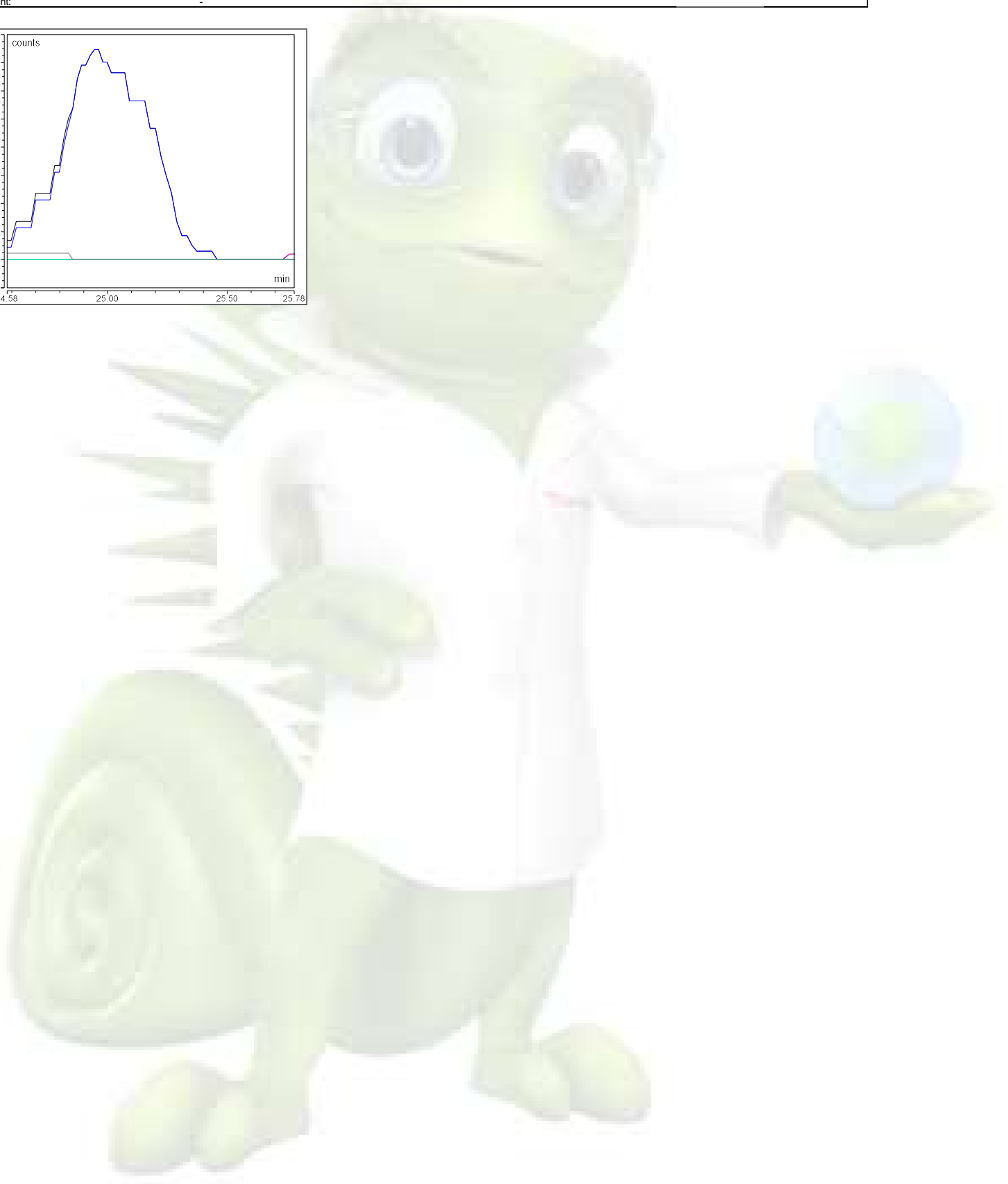
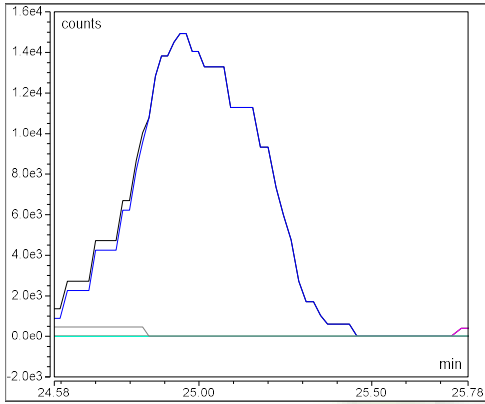
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

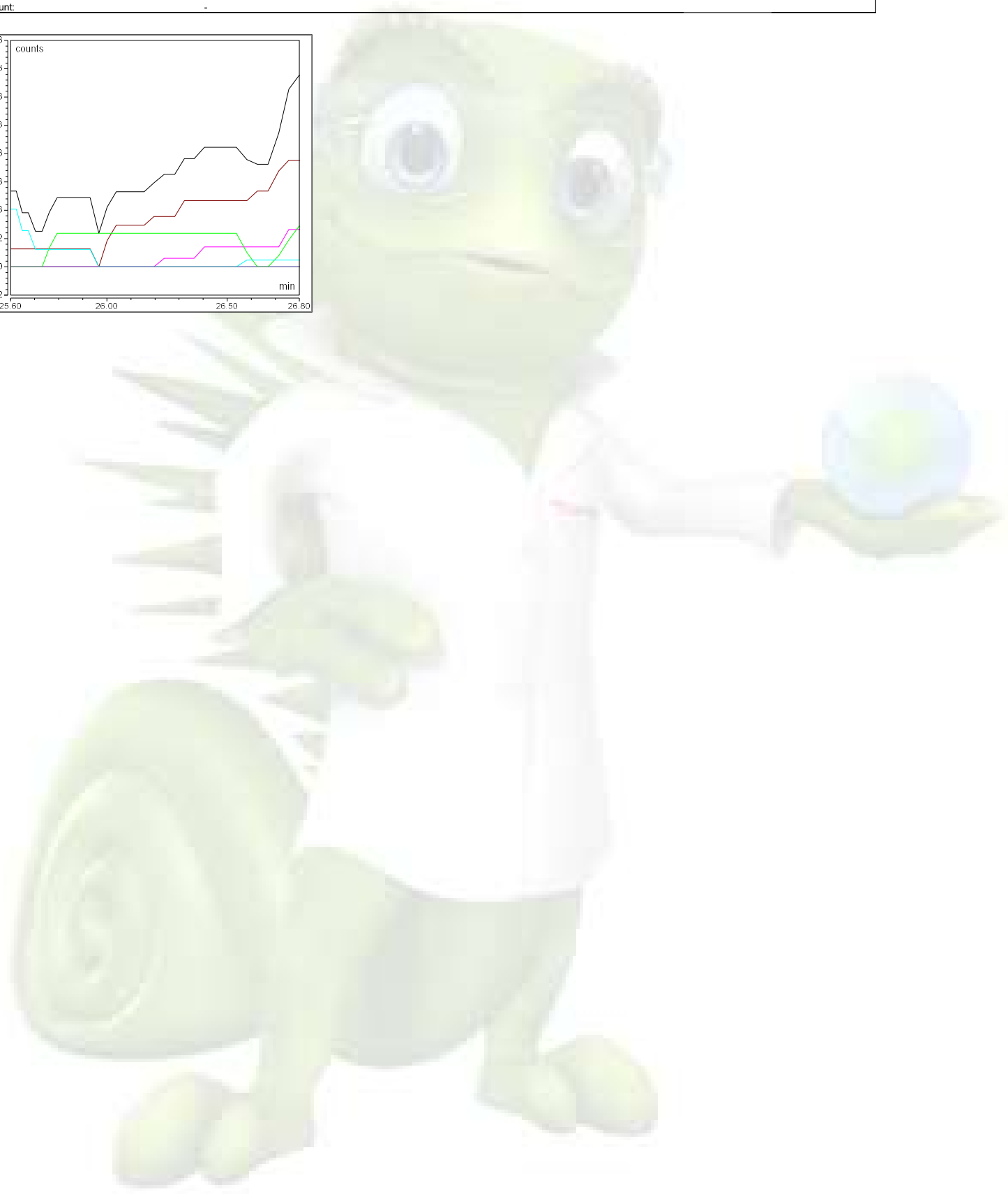
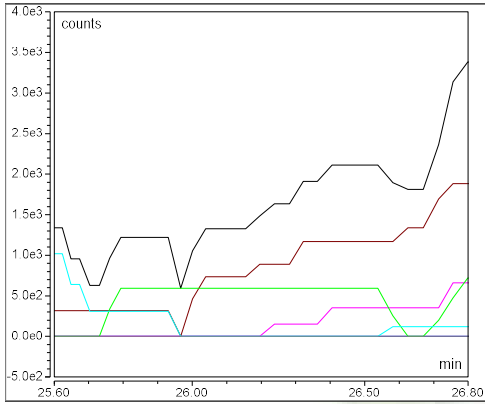
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

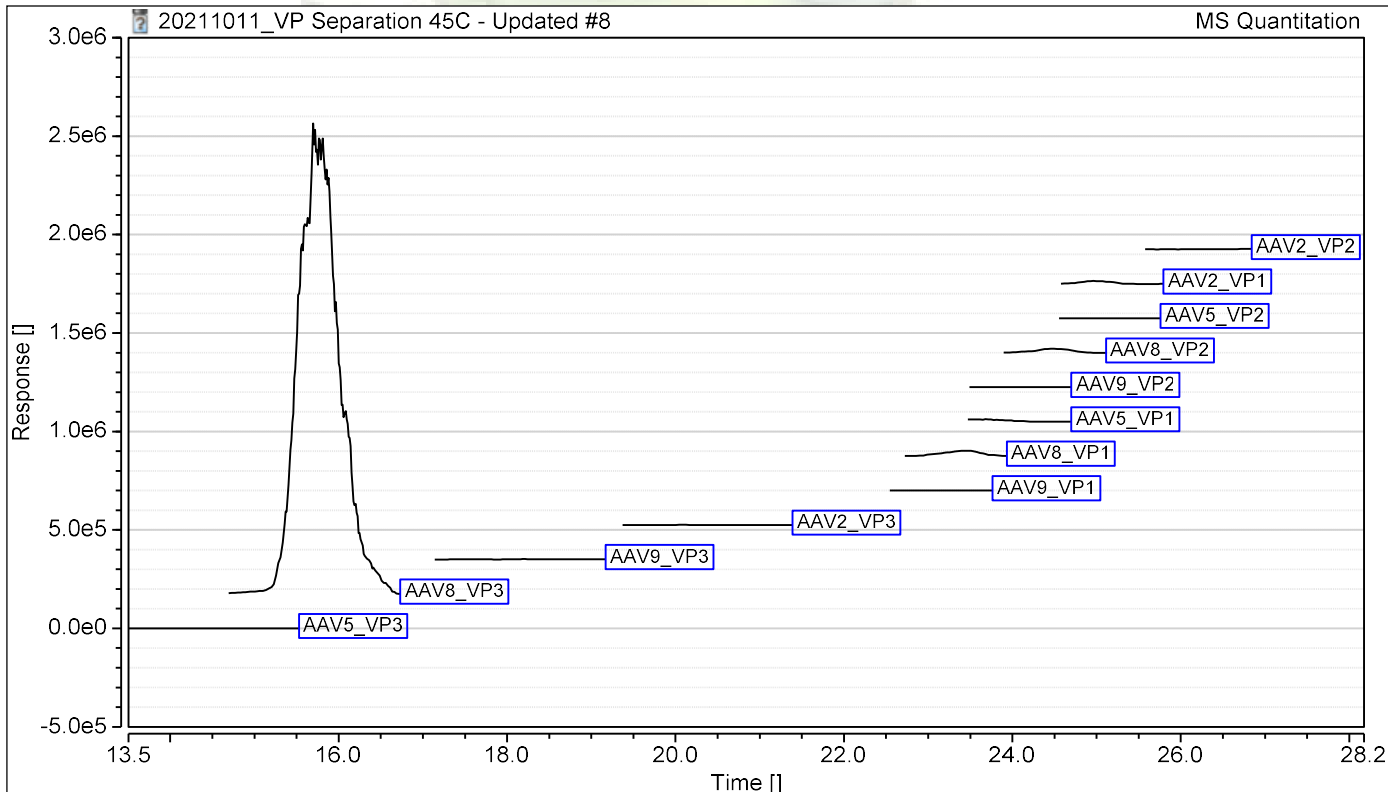
Amount: -



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV8 E_45C_65-59B_Mod1_1ug_2  | Run Time (min): 45.00   |
| Vial Number:         | R:A3   | Injection Volume: 6.25  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                         |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 13/Oct/21 02:28  | Sample Weight: 1.0000   |

### Chromatogram



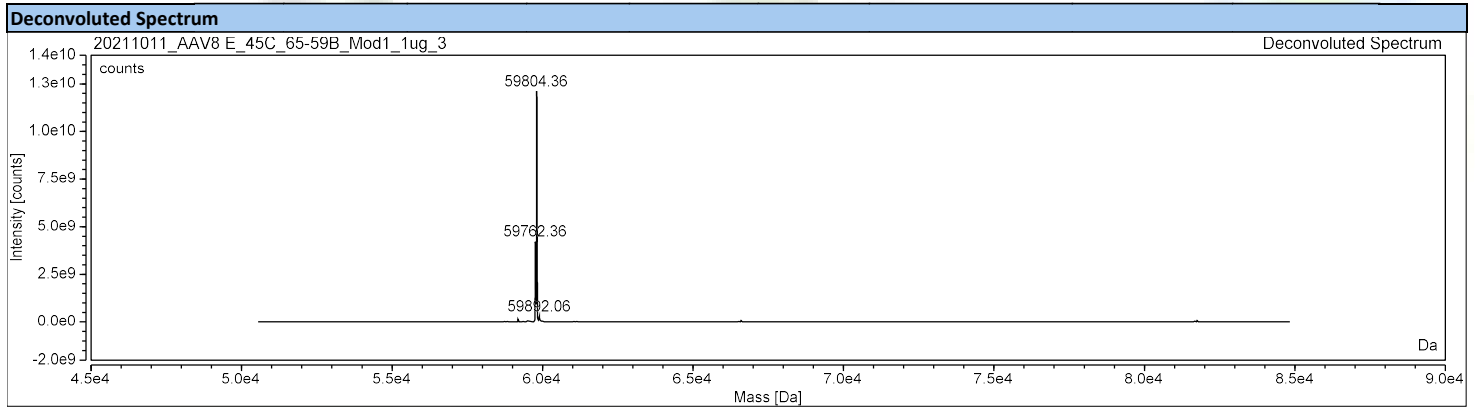
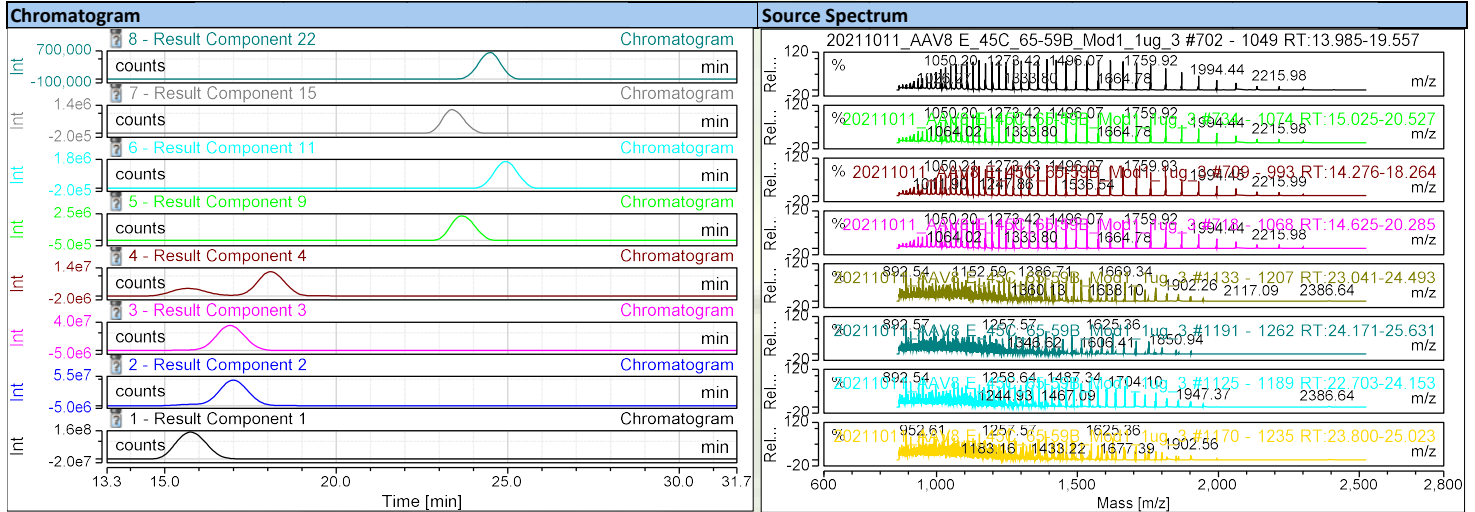
### SST Results

| No.  | Name | Inj. Condition | Peak | Test Result          | Injection     |
|--|------|----------------|------|----------------------|---------------|
| <b>Number of executed test cases: n.a.</b> |      |                |      | <b>Total Result:</b> | <b>Passed</b> |



## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV8 E_45C_65-59B_Mod1_1ug_3 | Injection Volume: 6.25                                       |
| Vial Number:      | R:A3                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 44.99                                 | Injection Date/Time: 13/Oct/21 03:14                         |



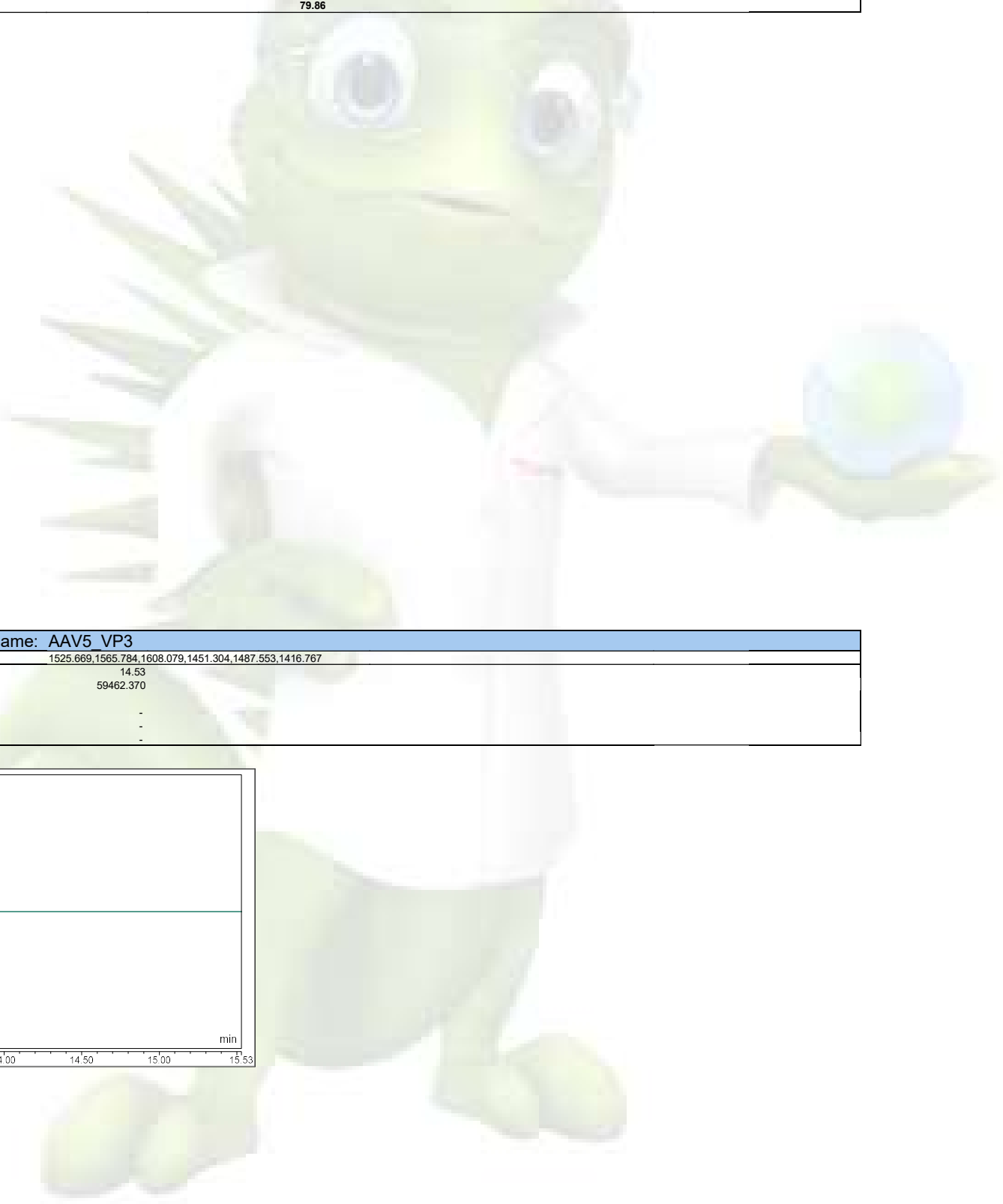
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

**Component Summary**

**Injection Details**

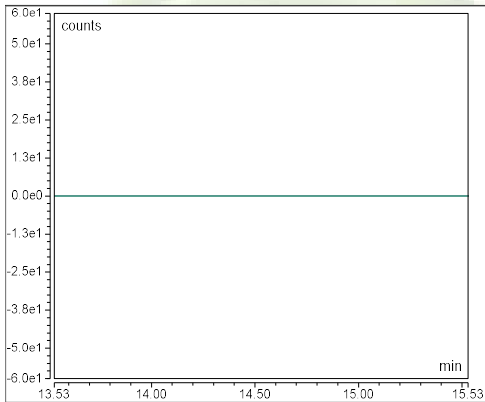
|                        |                                     |                             |   |
|------------------------|-------------------------------------|-----------------------------|---|
| <i>Injection Name:</i> | 20211011_AAV8_E_45C_65-59B_Mod1_1ug | <i>Injection Volume:</i>    | 6.25  |
| <i>Vial Number:</i>    | R:A3                                | <i>Instrument Method:</i>   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |
| <i>Injection Type:</i> | Unknown                             | <i>Processing Method:</i>   | MS Quantitative_45C_65-59B                            |
| <i>Run Time (min):</i> | 44.99                               | <i>Injection Date/Time:</i> | 13/Oct/21 03:14                                       |

| Peak Name | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|-----------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV8_VP3  | 15.758                | 1818378.111        | 79.07              | 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802 | 59804.680 | PASS                           |
| AAV8_VP1  | 23.482                | 10174.093          | 0.44               | 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828 | 81666.710 | PASS                           |
| AAV8_VP2  | 24.344                | 8116.115           | 0.35               | 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397 | 66518.100 | PASS                           |
|           |                       |                    | <b>79.86</b>       |   |           |                                |



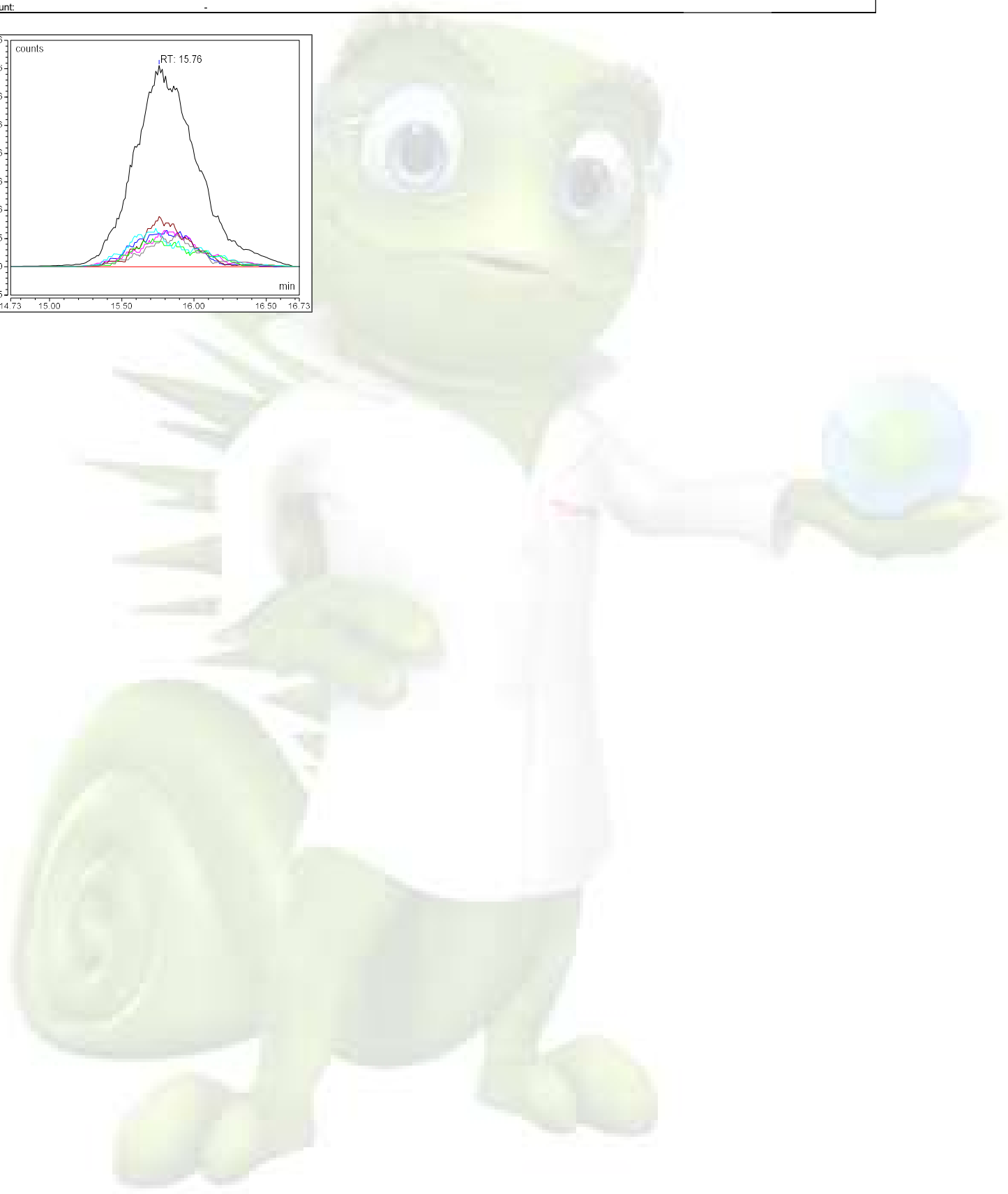
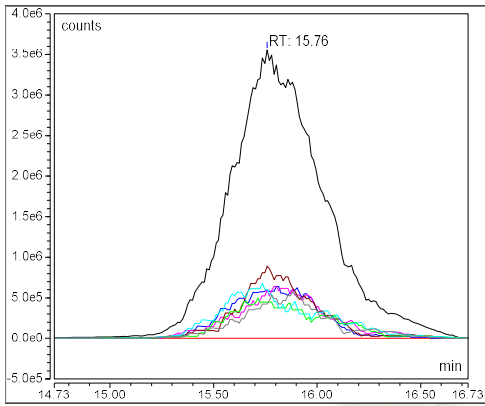
**Component Name: AAV5\_VP3**

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802 |
| Retention time (min): | 15.73   |
| Expected Mass:        | 59804.680   |
| Area (count*min):     | 1818378   |
| Intensity (counts):   | 3555275   |
| Amount:               | -   |



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

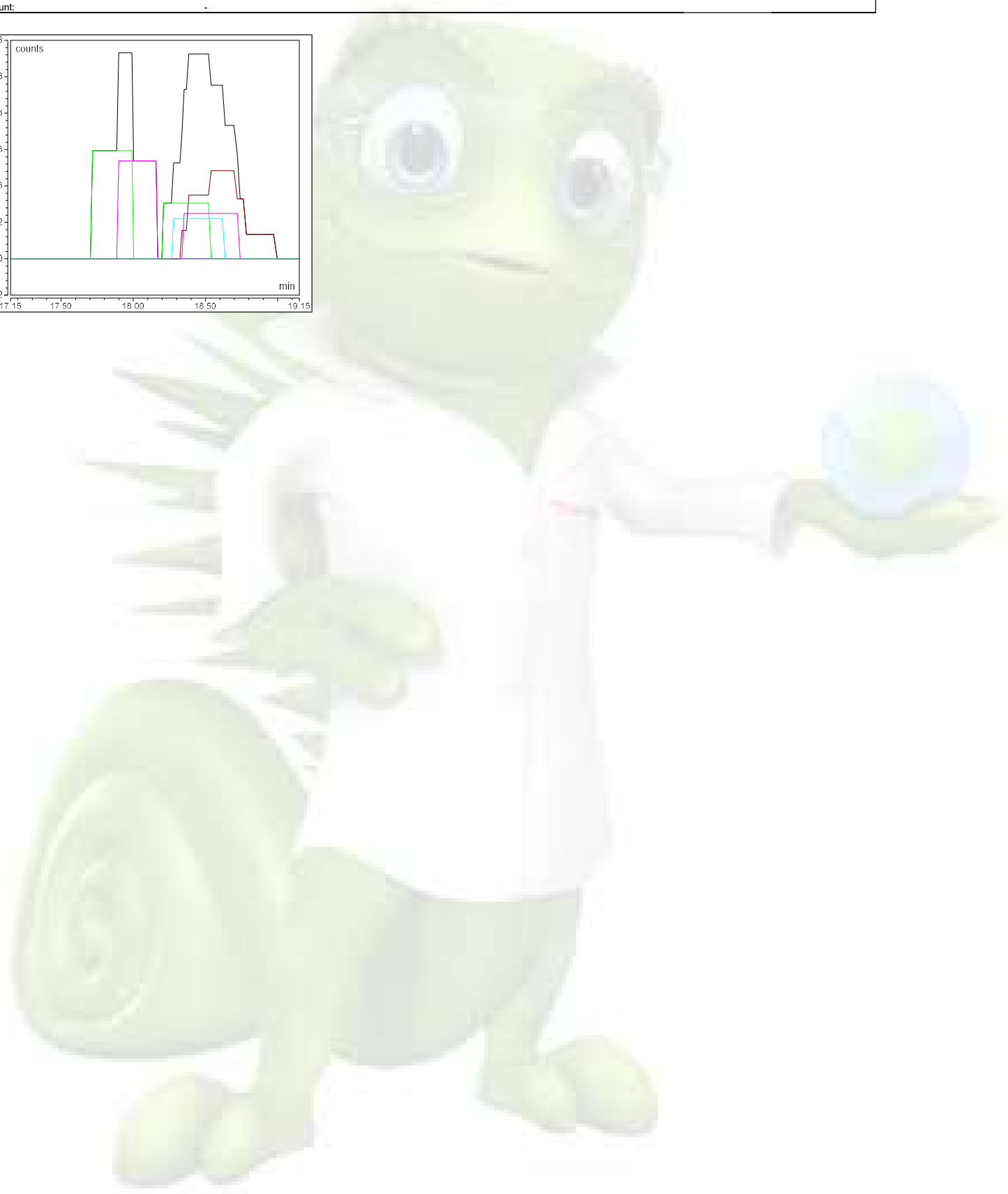
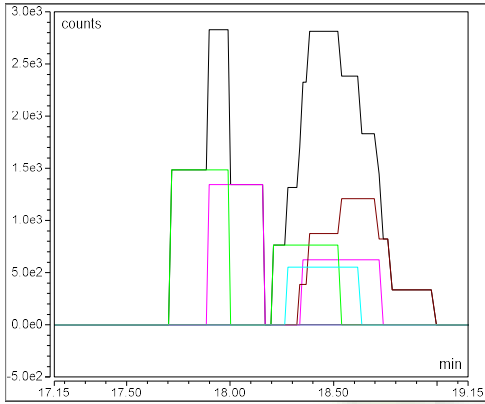
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

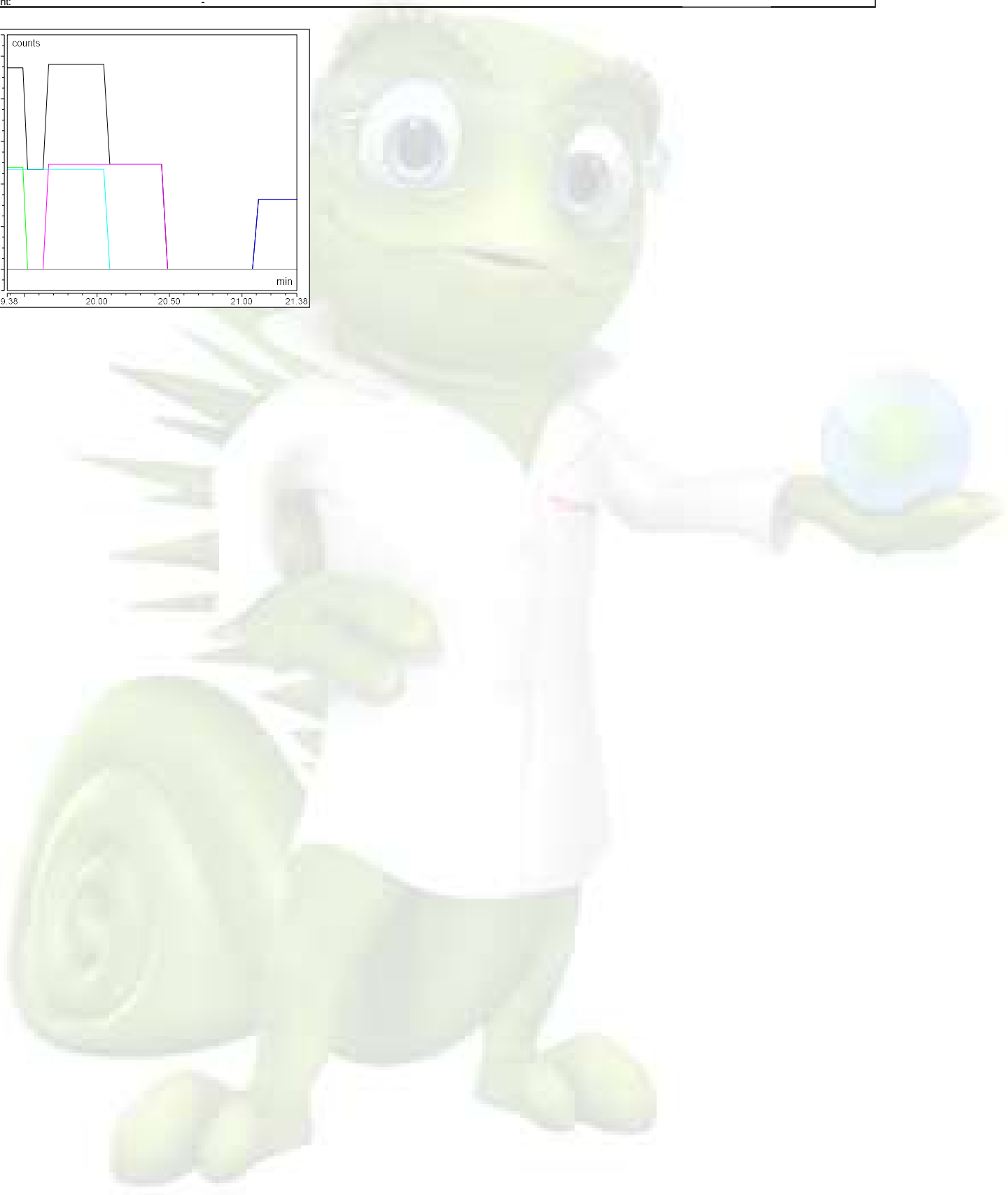
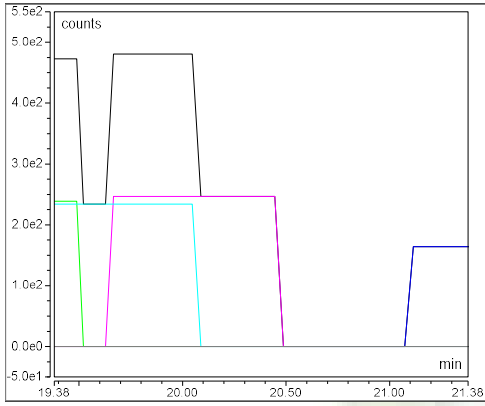
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

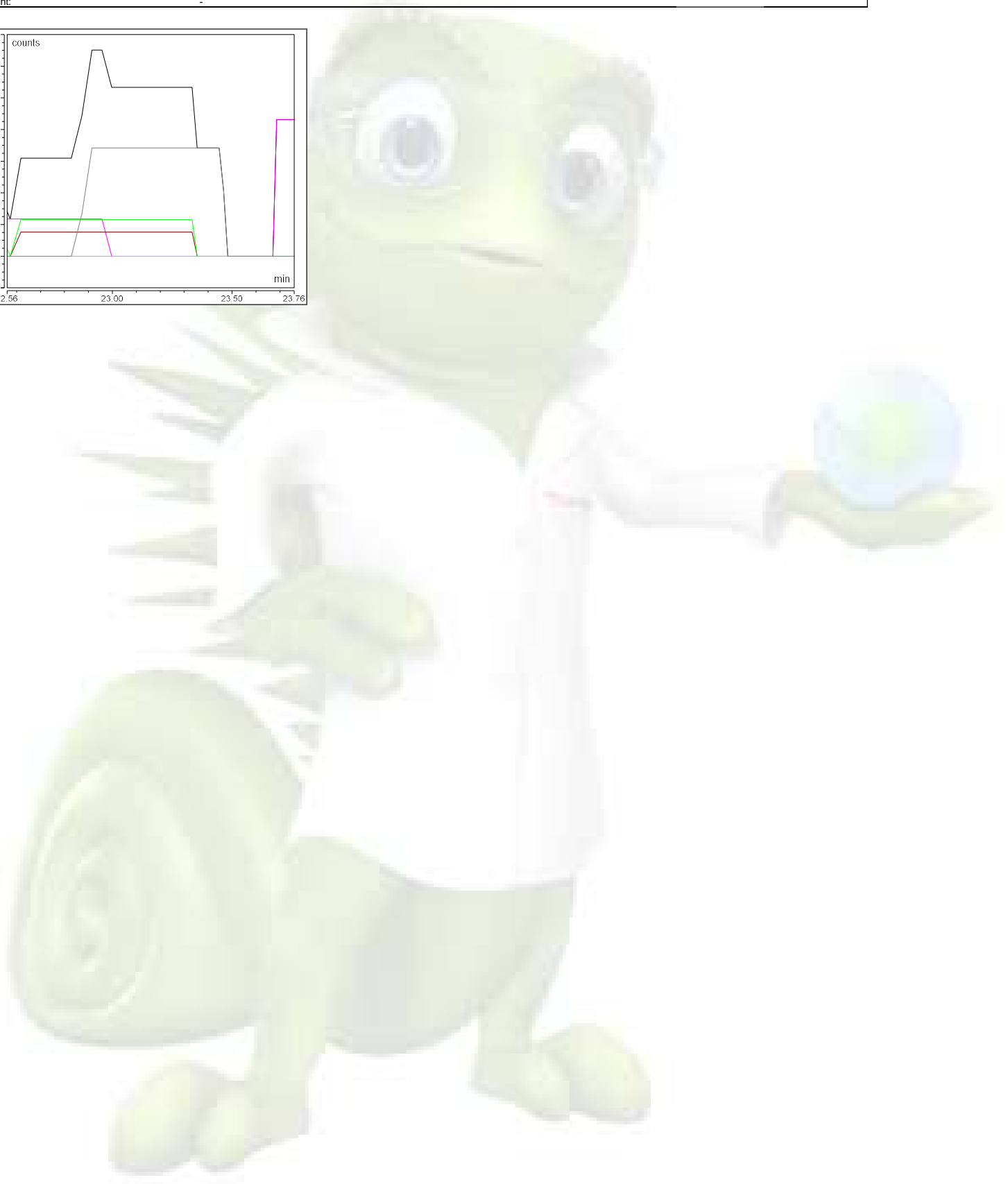
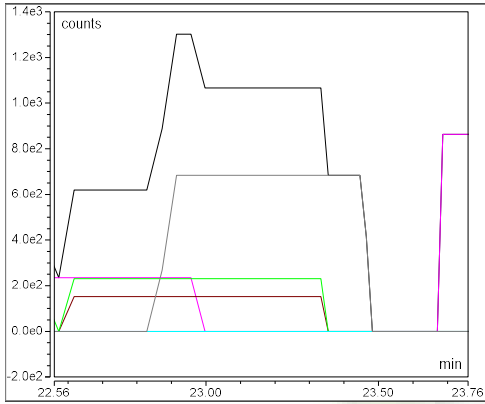
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

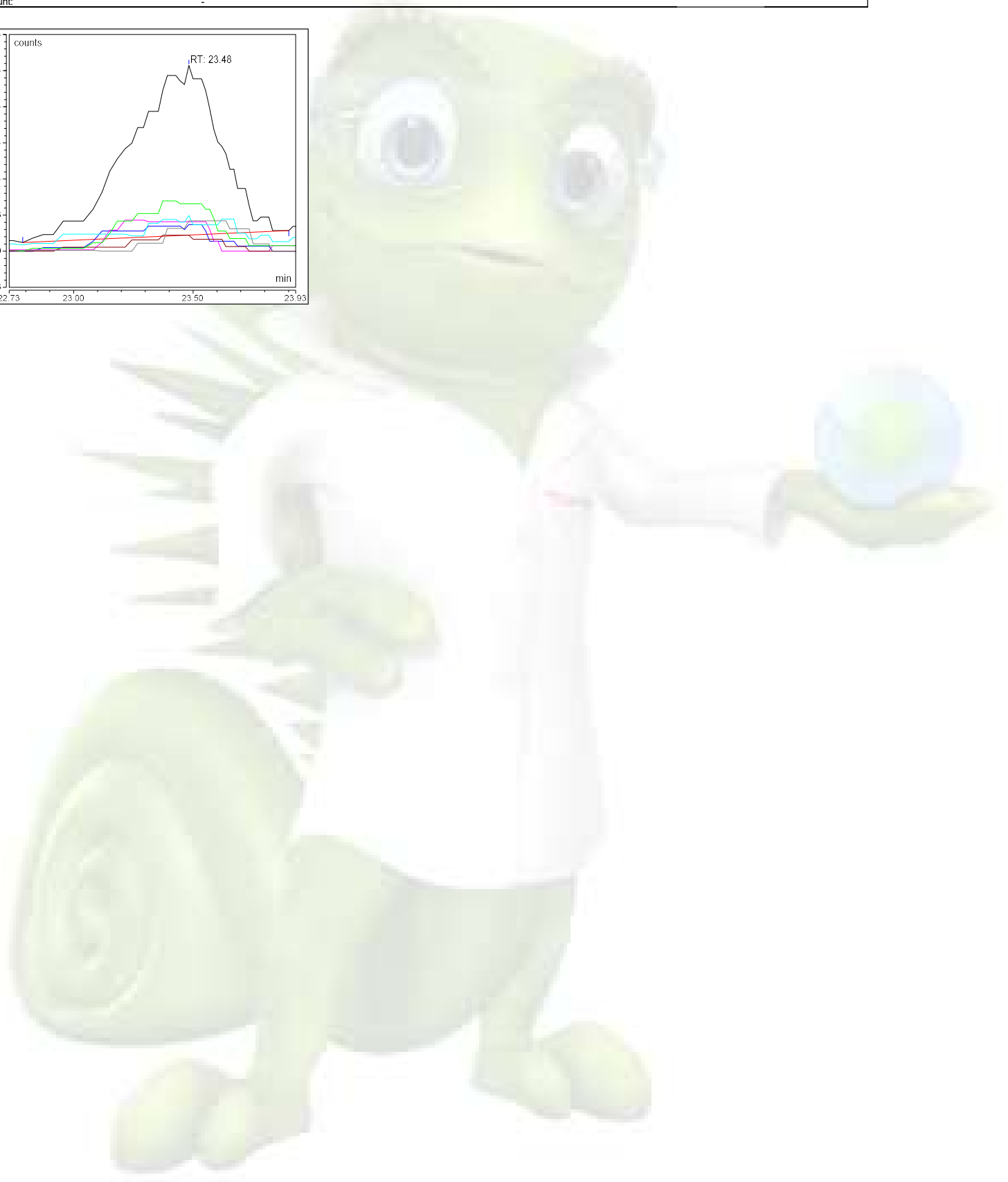
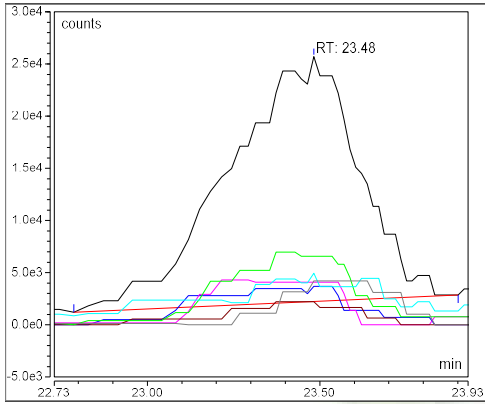
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): 10174

Intensity (counts): 23484

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

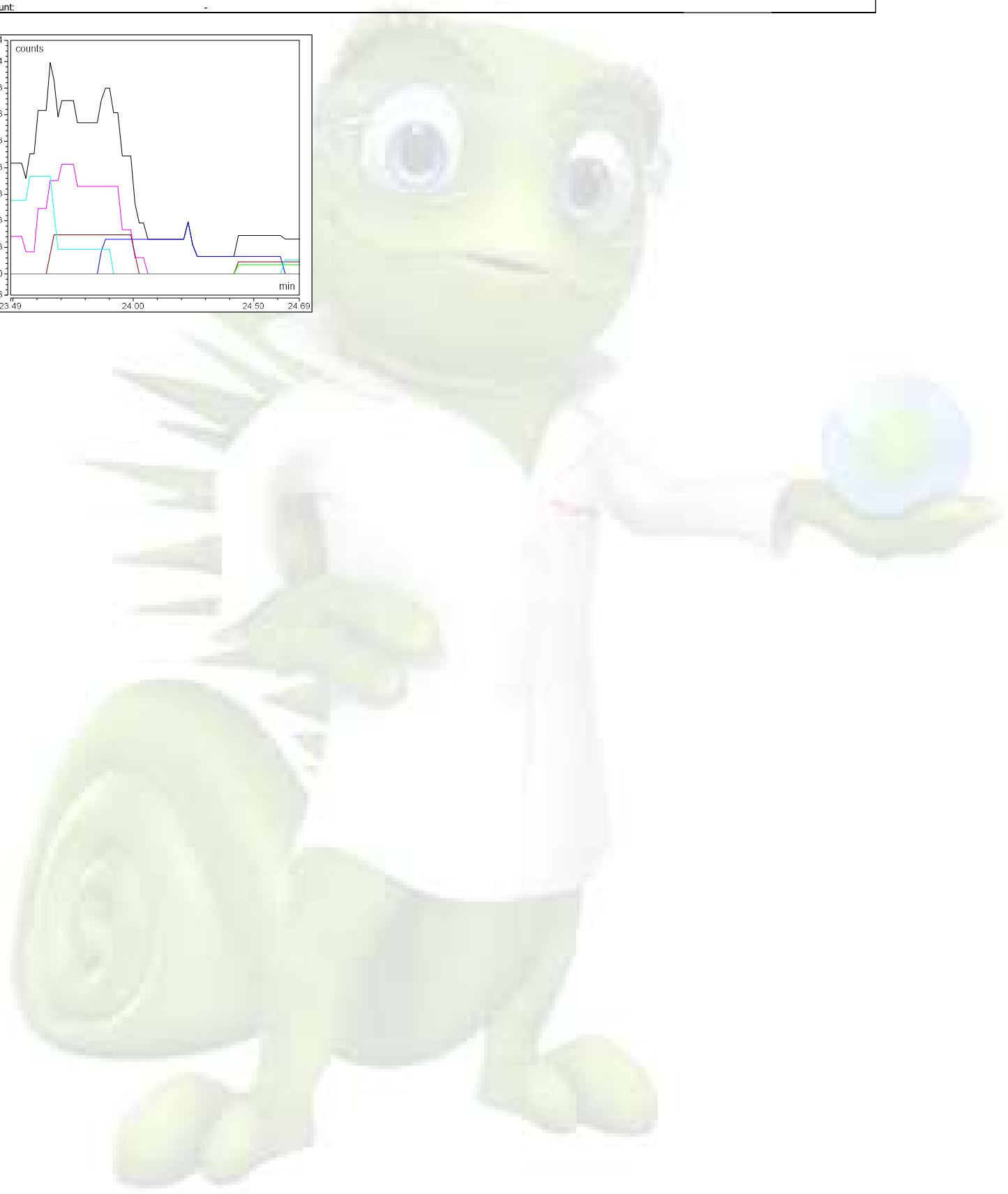
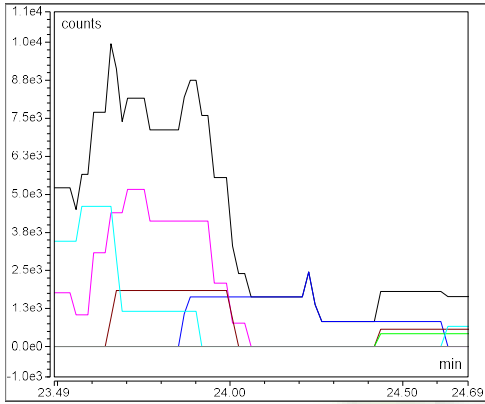
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

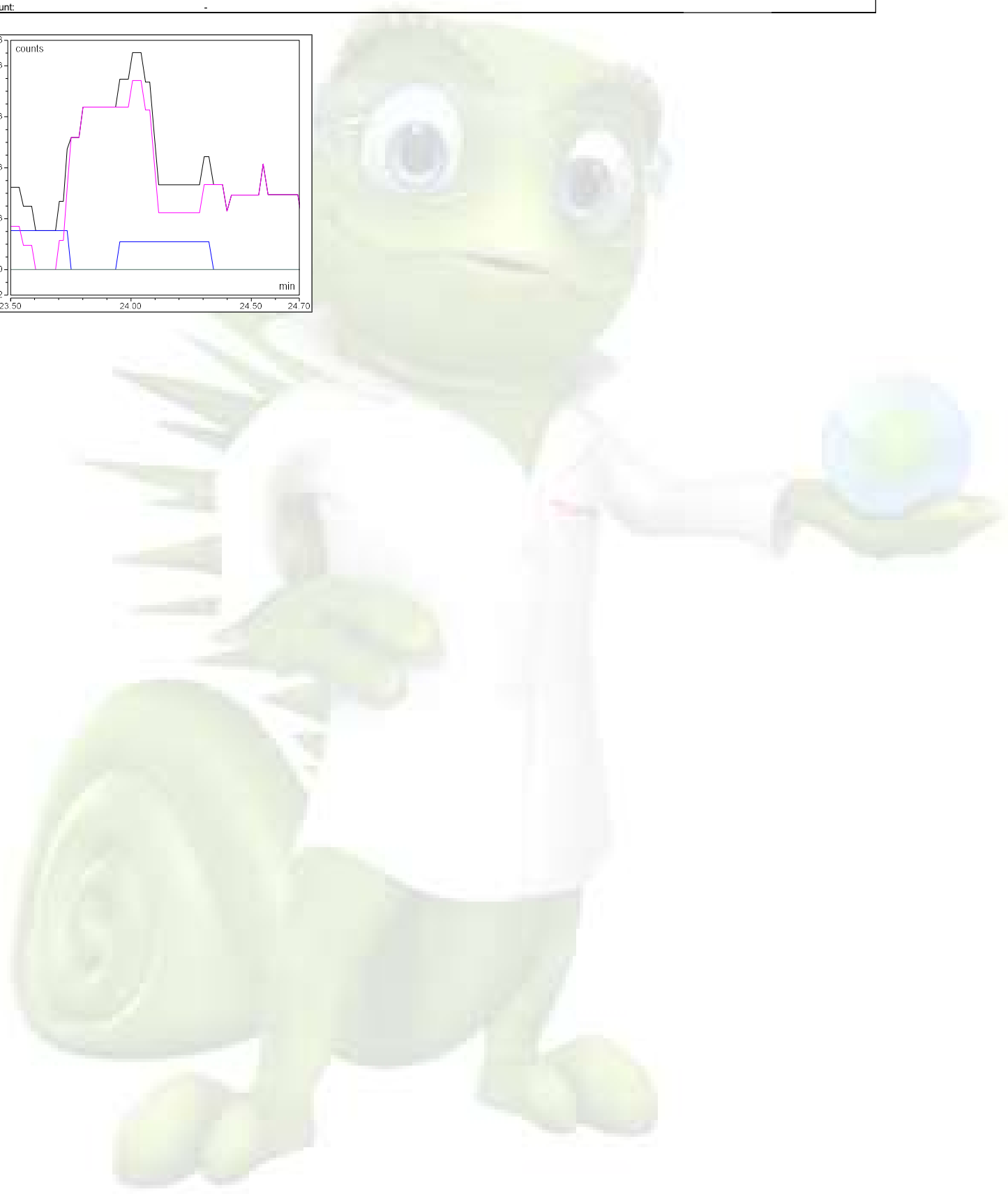
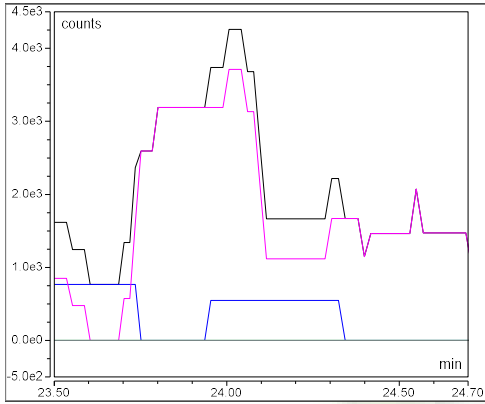
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

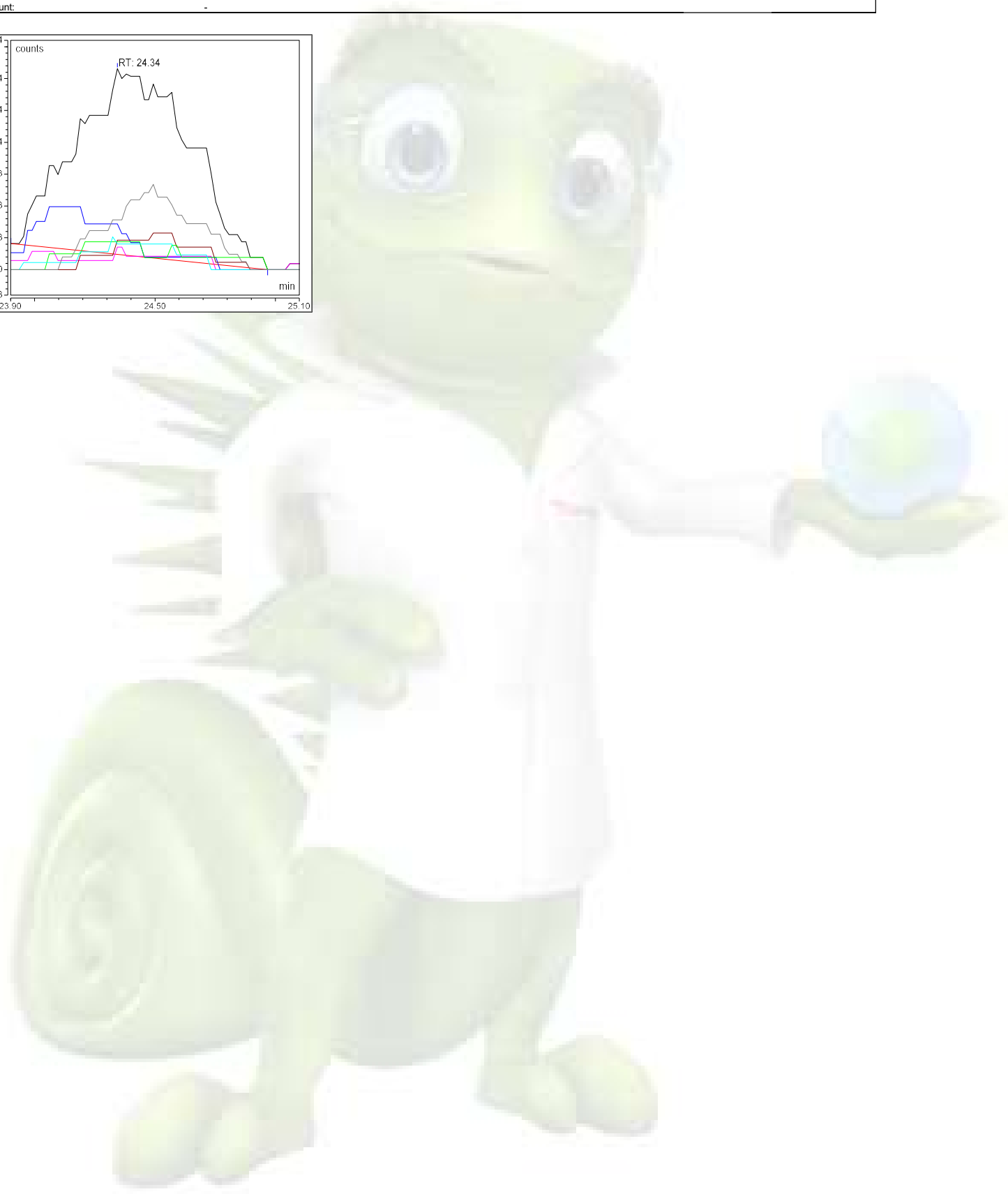
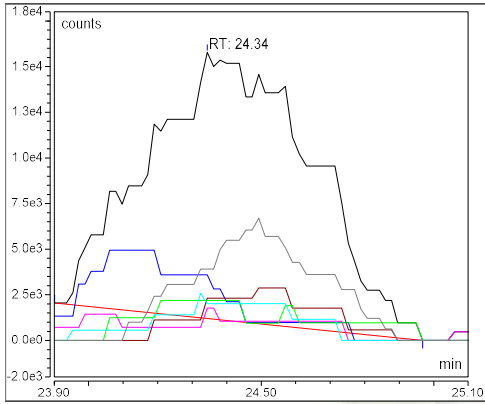
Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397 |
| Retention time (min): | 24.50   |
| Expected Mass:        | 66518.100   |
| Area (count*min):     | 8116  |
| Intensity (counts):   | 14575   |
| Amount:               | -   |



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

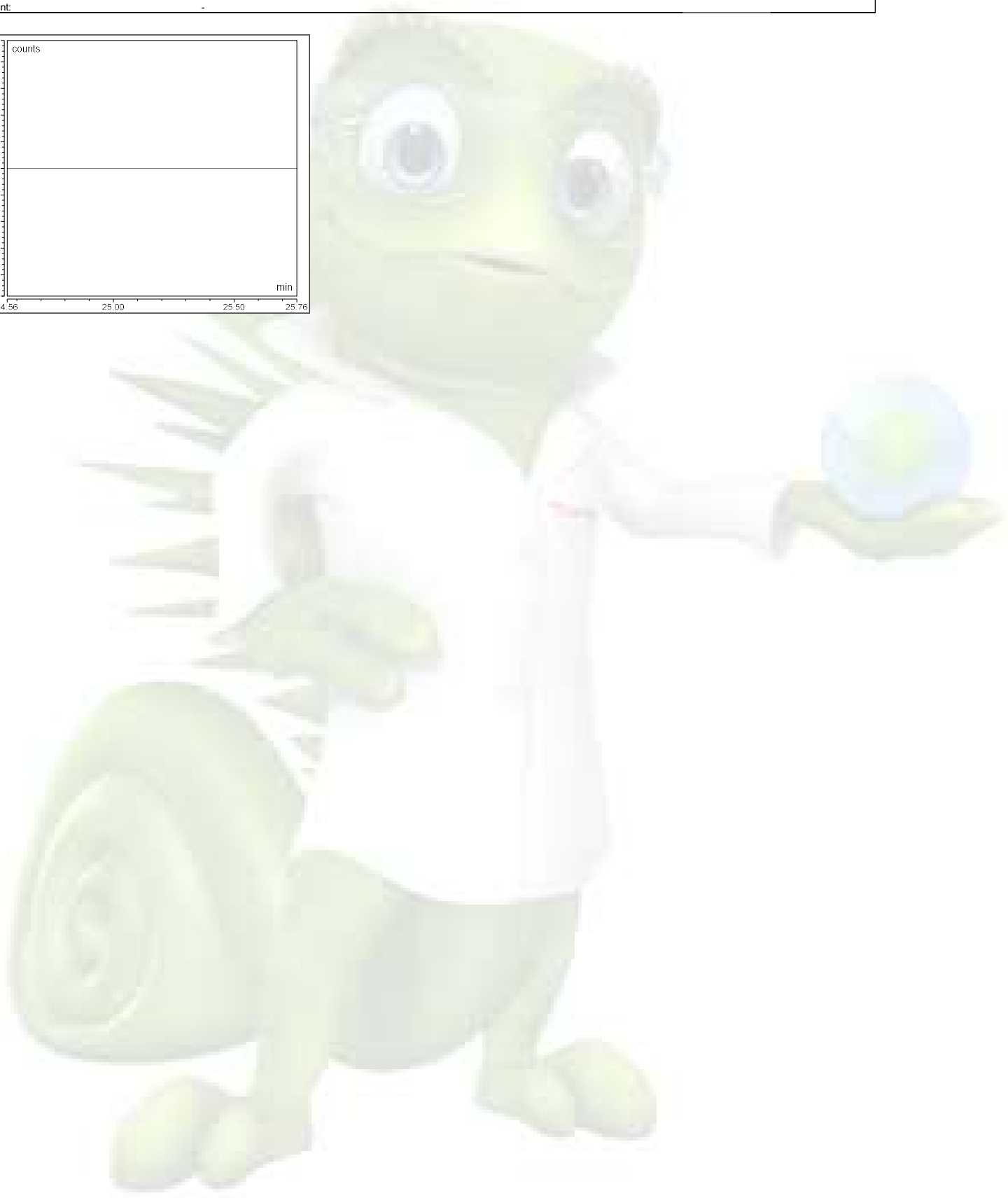
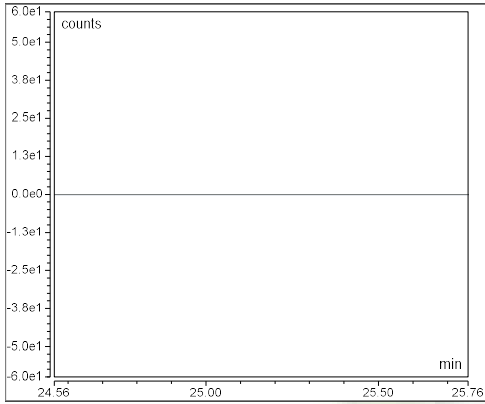
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

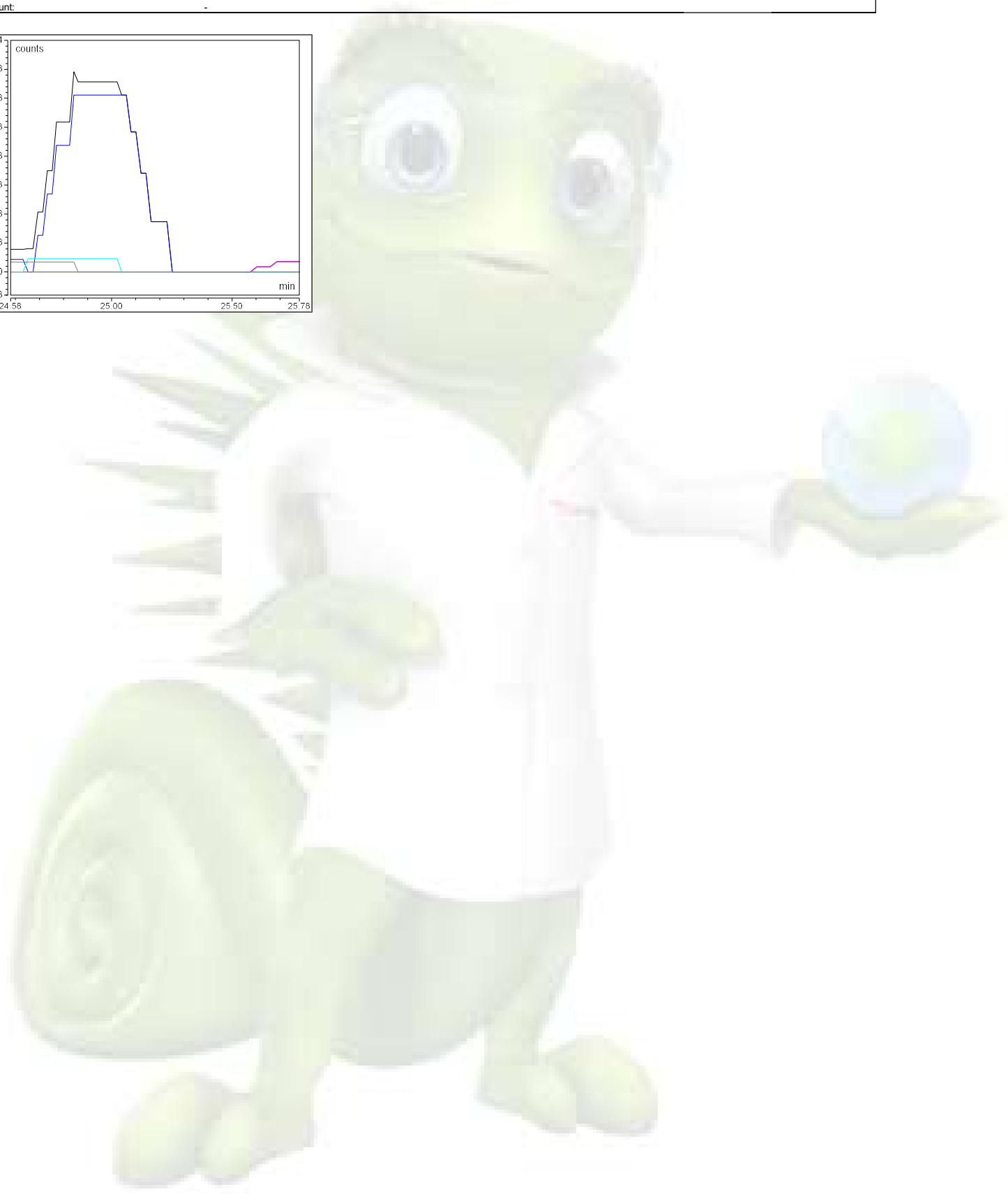
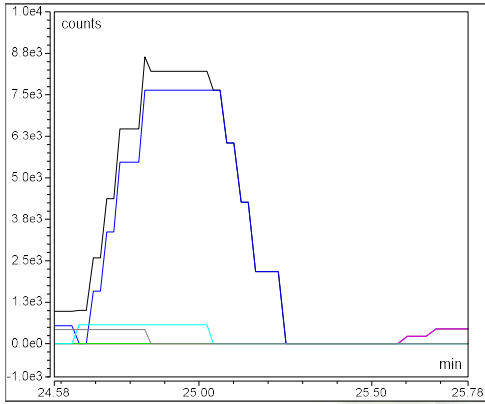
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

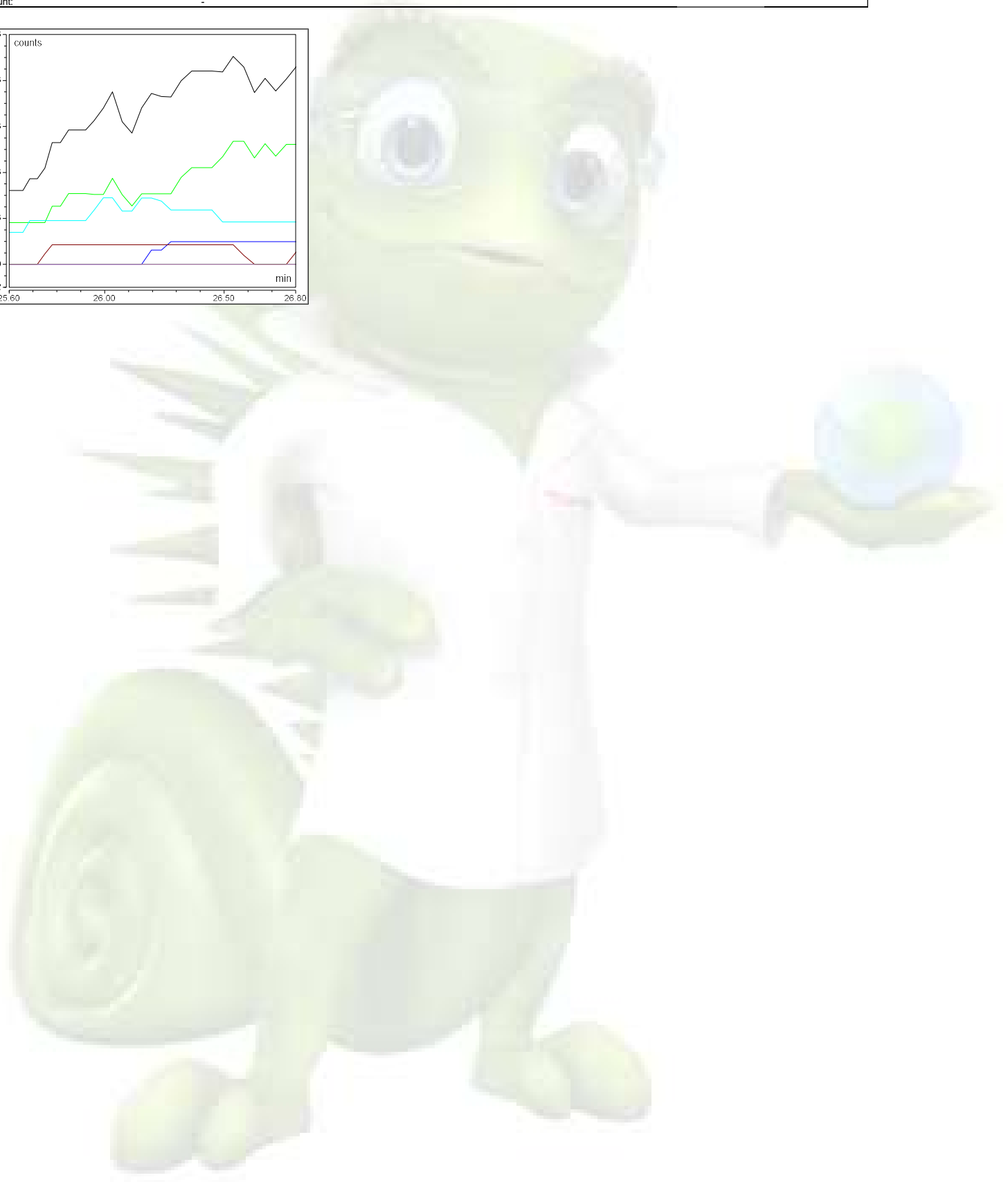
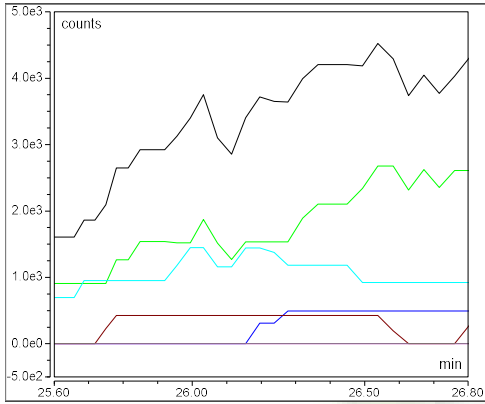
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

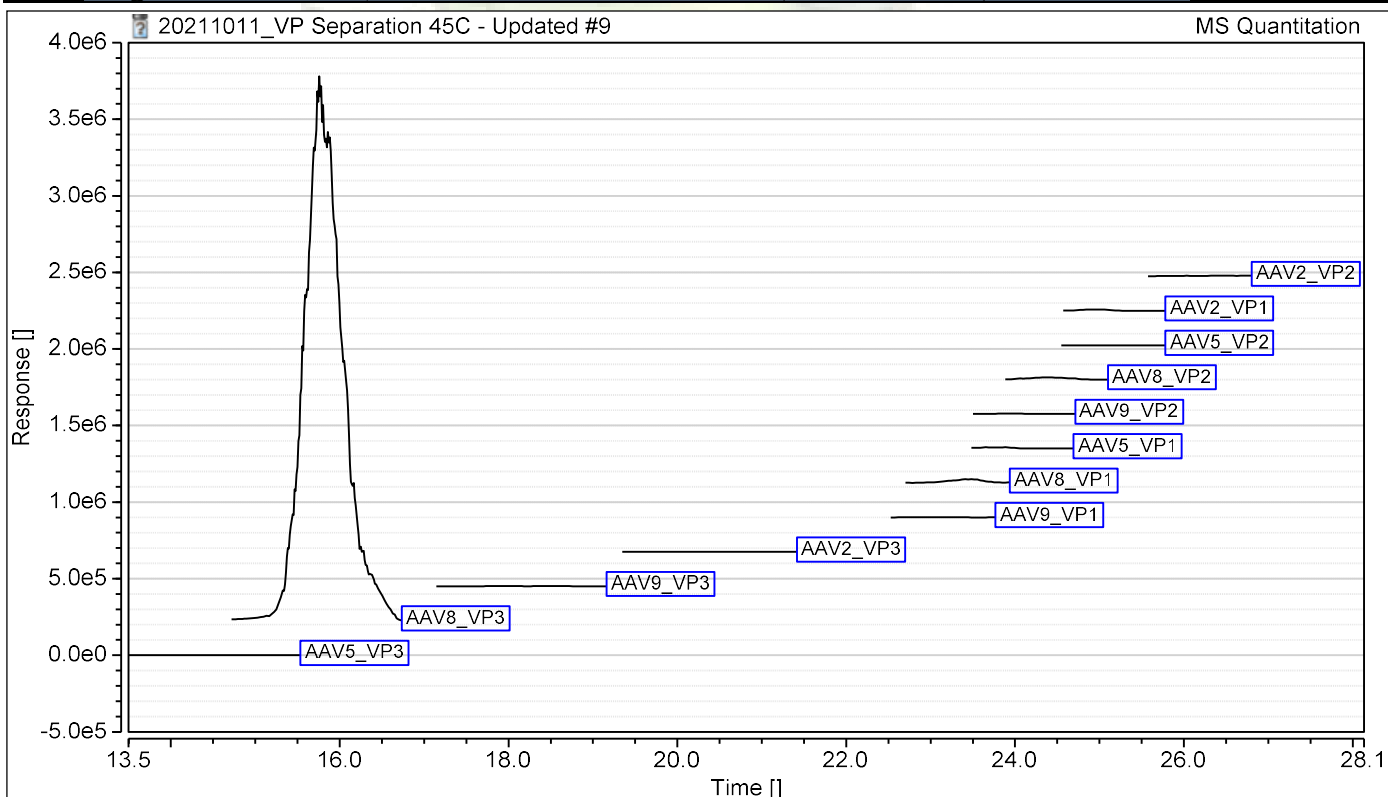
Amount: -



## Chromatogram and SST Results

| Injection Details    |  |                                |
|----------------------|--|--------------------------------|
| Injection Name:      | 20211011_AAV8 E_45C_65-59B_Mod1_1ug_3  | Run Time (min): <b>44.99</b>   |
| Vial Number:         | R:A3   | Injection Volume: <b>6.25</b>  |
| Injection Type:      | Unknown  |                                |
| Calibration Level:   |  |                                |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                                |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: <b>1.0000</b> |
| Injection Date/Time: | 13/Oct/21 03:14  | Sample Weight: <b>1.0000</b>   |

### Chromatogram

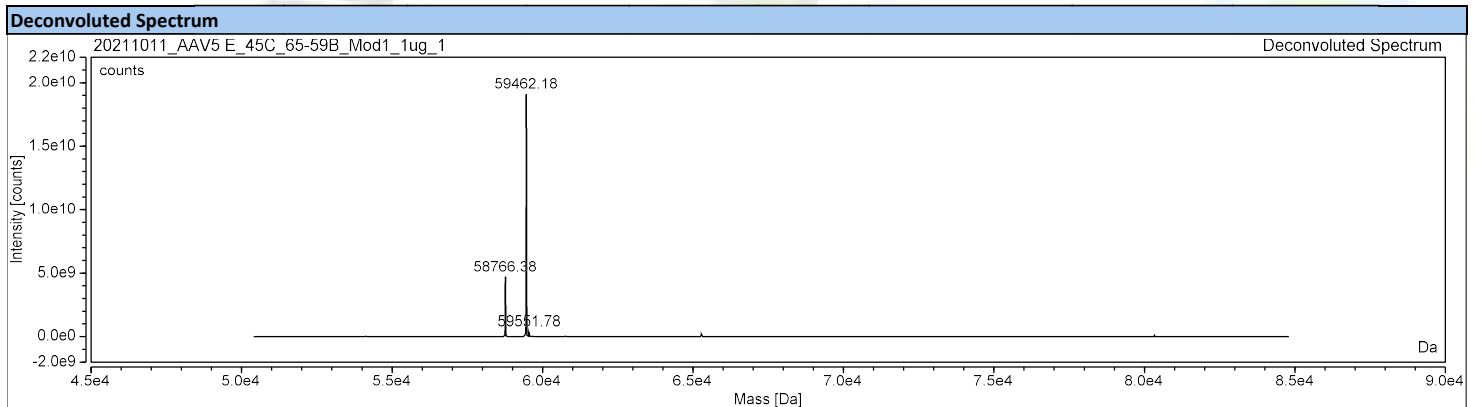
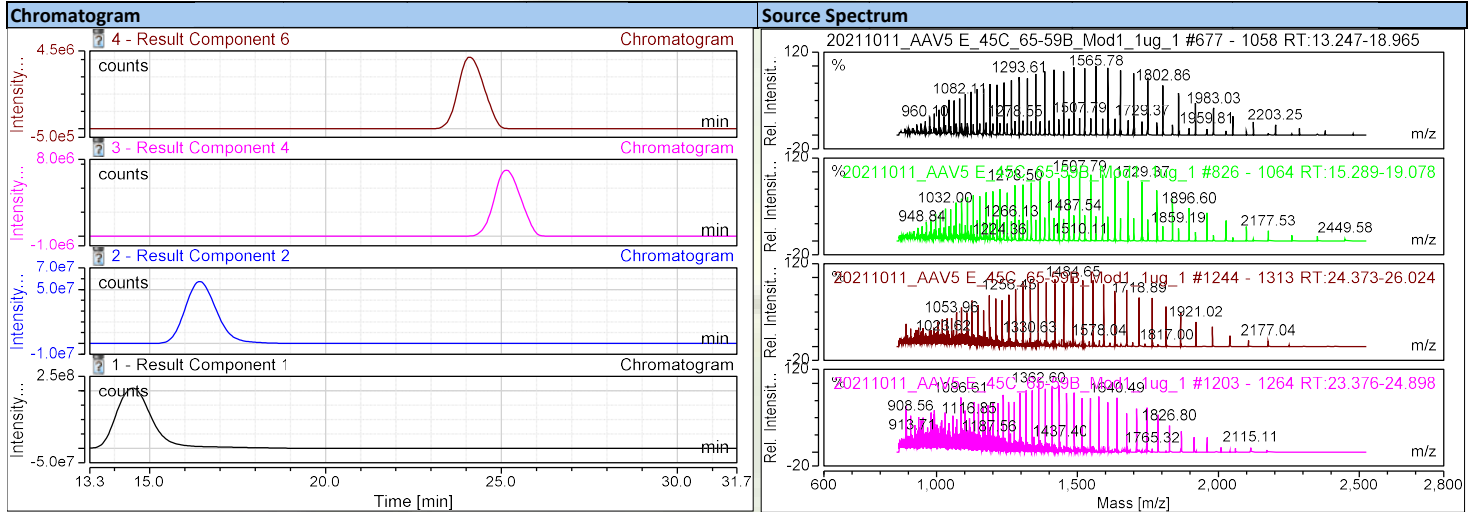


### SST Results

| No.  | Name | Inj. Condition | Peak                 | Test Result   | Injection |
|--|------|----------------|----------------------|---------------|-----------|
| <b>Number of executed test cases: n.a.</b> |      |                | <b>Total Result:</b> | <b>Passed</b> |           |

## Protein Deconvolution Summary

| Injection Details |                                       |                            |
|-------------------|---------------------------------------|----------------------------|
| Injection Name:   | 20211011_AAV5 E_45C_65-59B_Mod1_1ug_1 | Injection Volume:          |
| Vial Number:      | R:A7                                  | Instrument Method:         |
| Injection Type:   | Unknown                               | Processing Method:         |
| Run Time (min):   | 44.99                                 | Injection Date/Time:       |
|                   |                                       | MS Quantitative_45C_65-59B |
|                   |                                       | 13/Oct/21 05:26            |



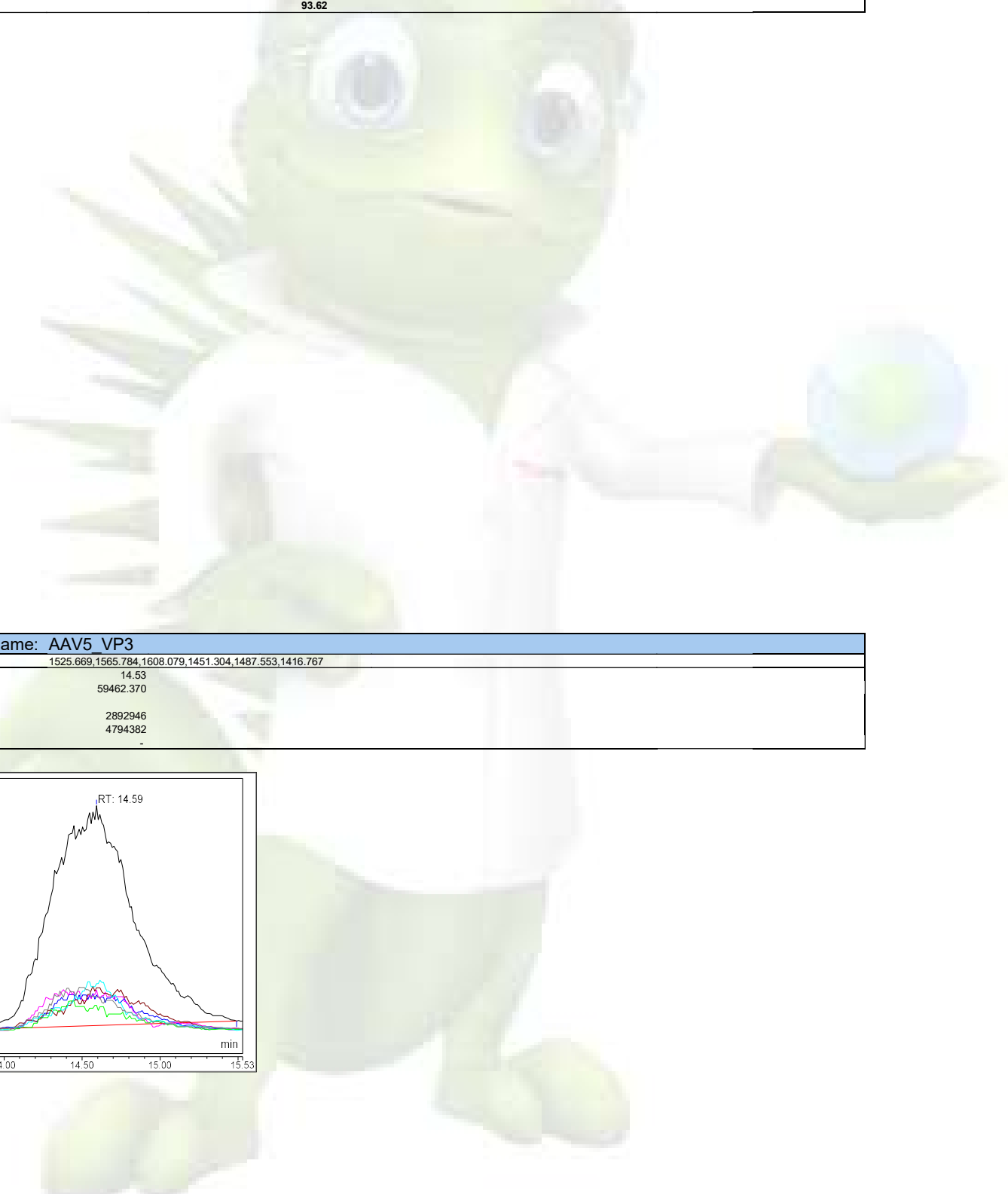
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

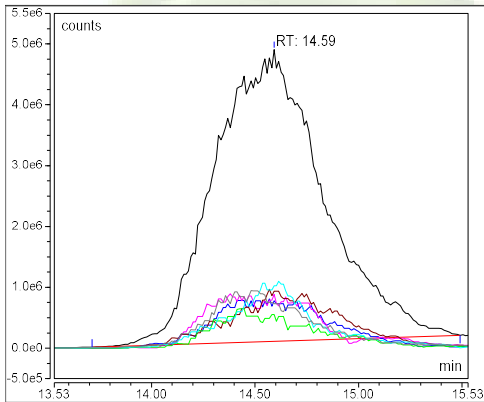
|  |   |  |
|--|---|--|
| <b>Injection Name:</b> 20211011_AAV5_E_45C_65-59B_Mod1_1ug | <b>Injection Volume:</b> 3.17   |  |
| <b>Vial Number:</b> R:A7                                   | <b>Instrument Method:</b> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <b>Injection Type:</b> Unknown                             | <b>Processing Method:</b> MS Quantitative_45C_65-59B                            |  |
| <b>Run Time (min):</b> 44.99                               | <b>Injection Date/Time:</b> 13/Oct/21 05:26                                     |  |

| Peak Name | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |  |
|-----------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|--|
| AAV5_VP3  | 14.592                | 2892946.404        | 89.95              | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 | 59462.370 | PASS                           |  |
| AAV5_VP1  | 24.157                | 33639.319          | 1.05               | 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697 | 80335.080 | PASS                           |  |
| AAV5_VP2  | 25.077                | 84281.750          | 2.62               | 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727 | 65282.690 | PASS                           |  |
|           |                       |                    | <b>93.62</b>       |   |           |                                |  |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <b>Quantitation ions:</b>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <b>Retention time (min):</b> | 14.53   |
| <b>Expected Mass:</b>        | 59462.370   |
| <b>Area (count*min):</b>     | 2892946   |
| <b>Intensity (counts):</b>   | 4794382   |
| <b>Amount:</b>               | -   |





**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

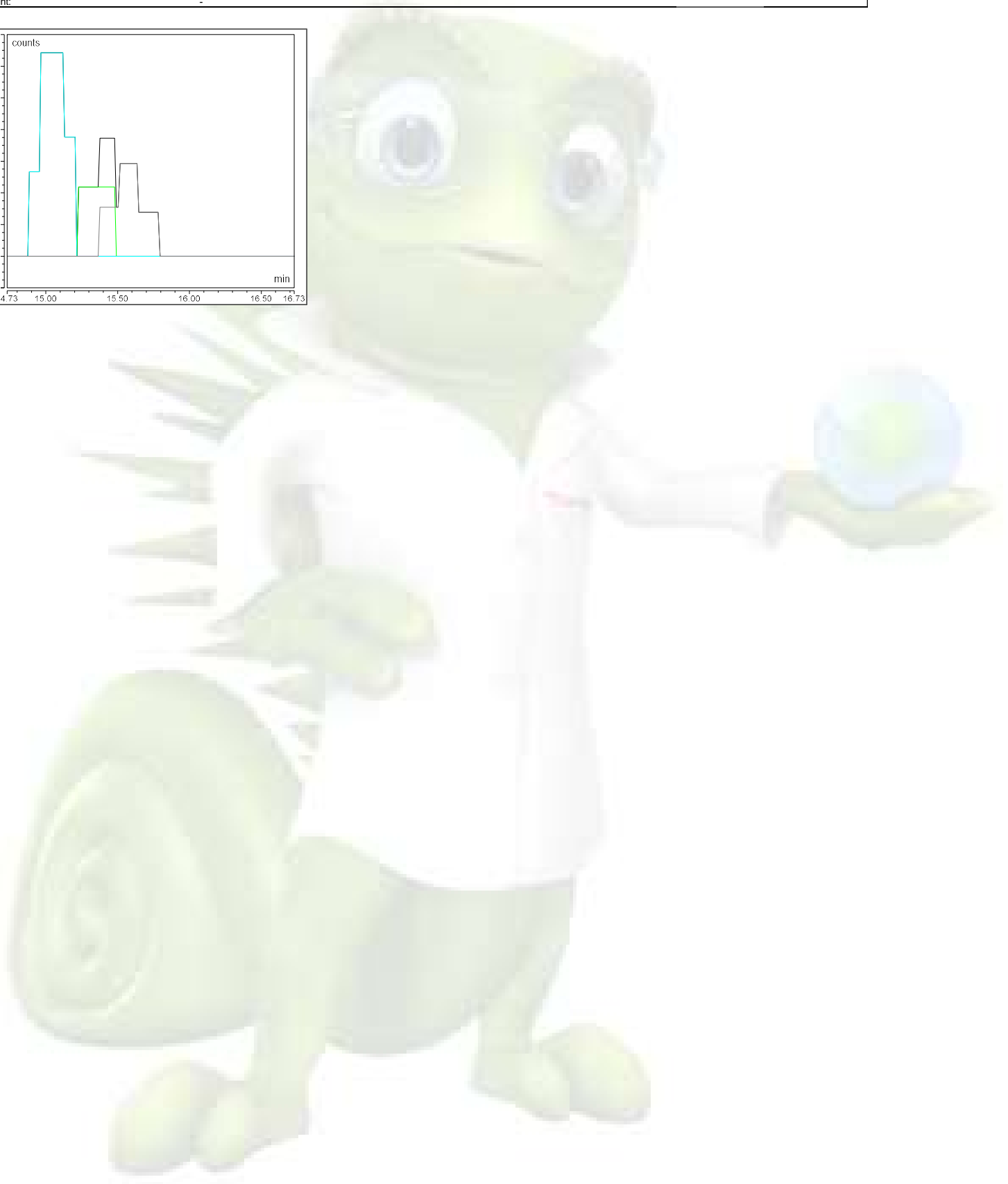
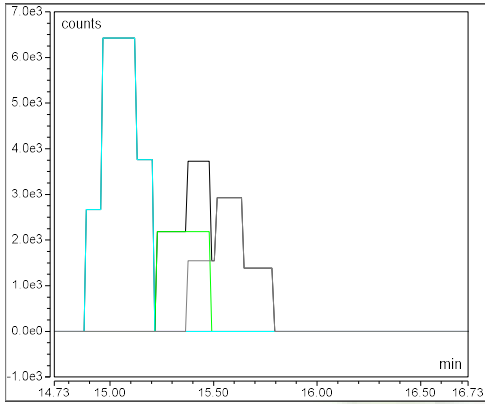
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

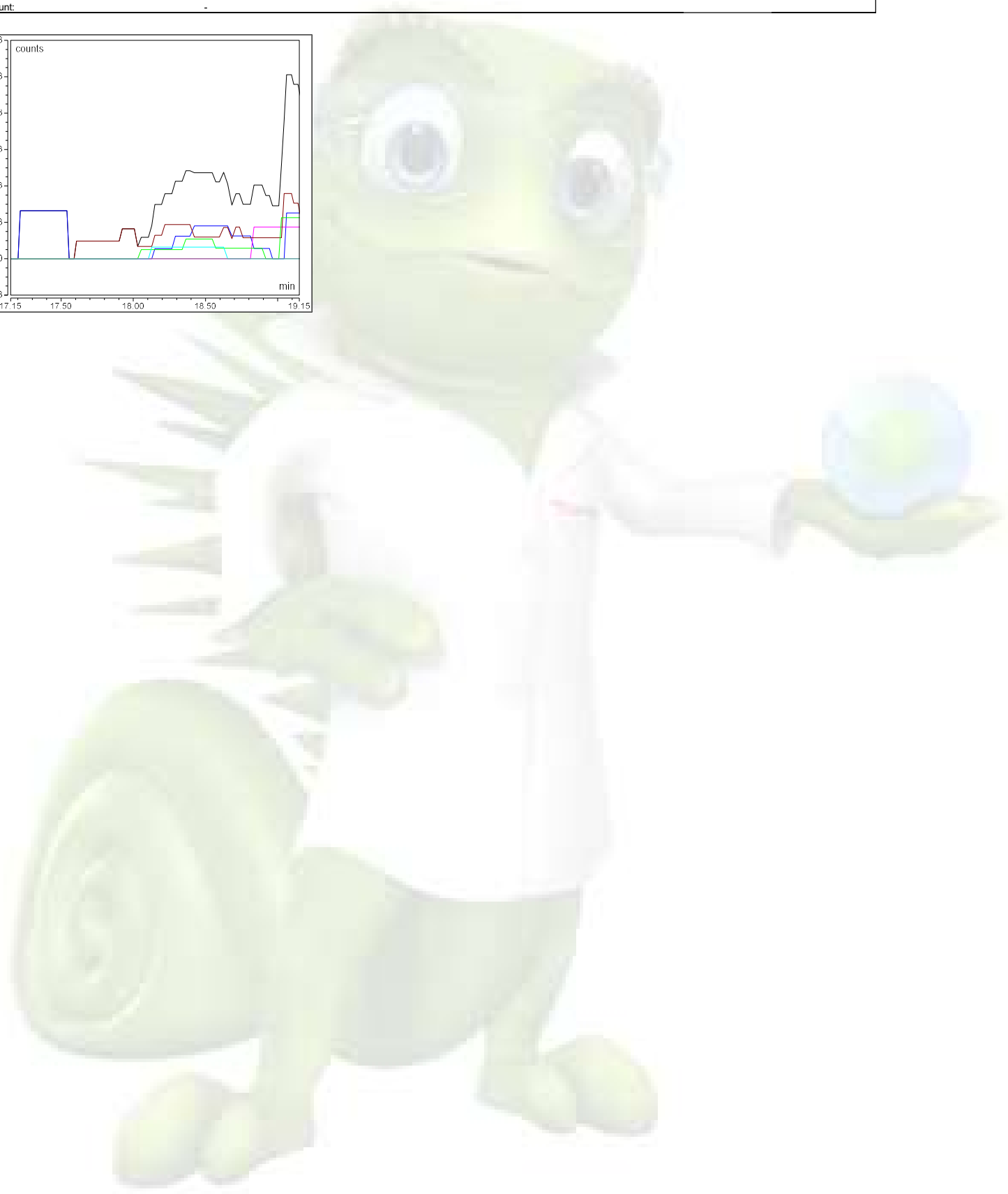
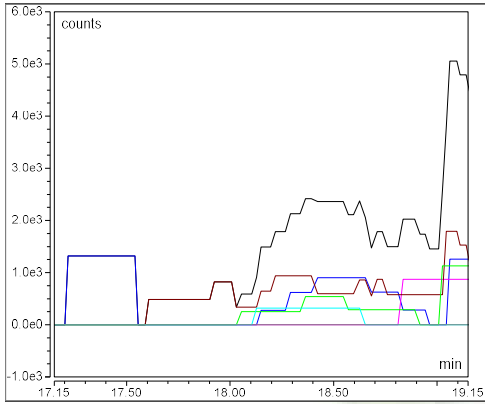
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

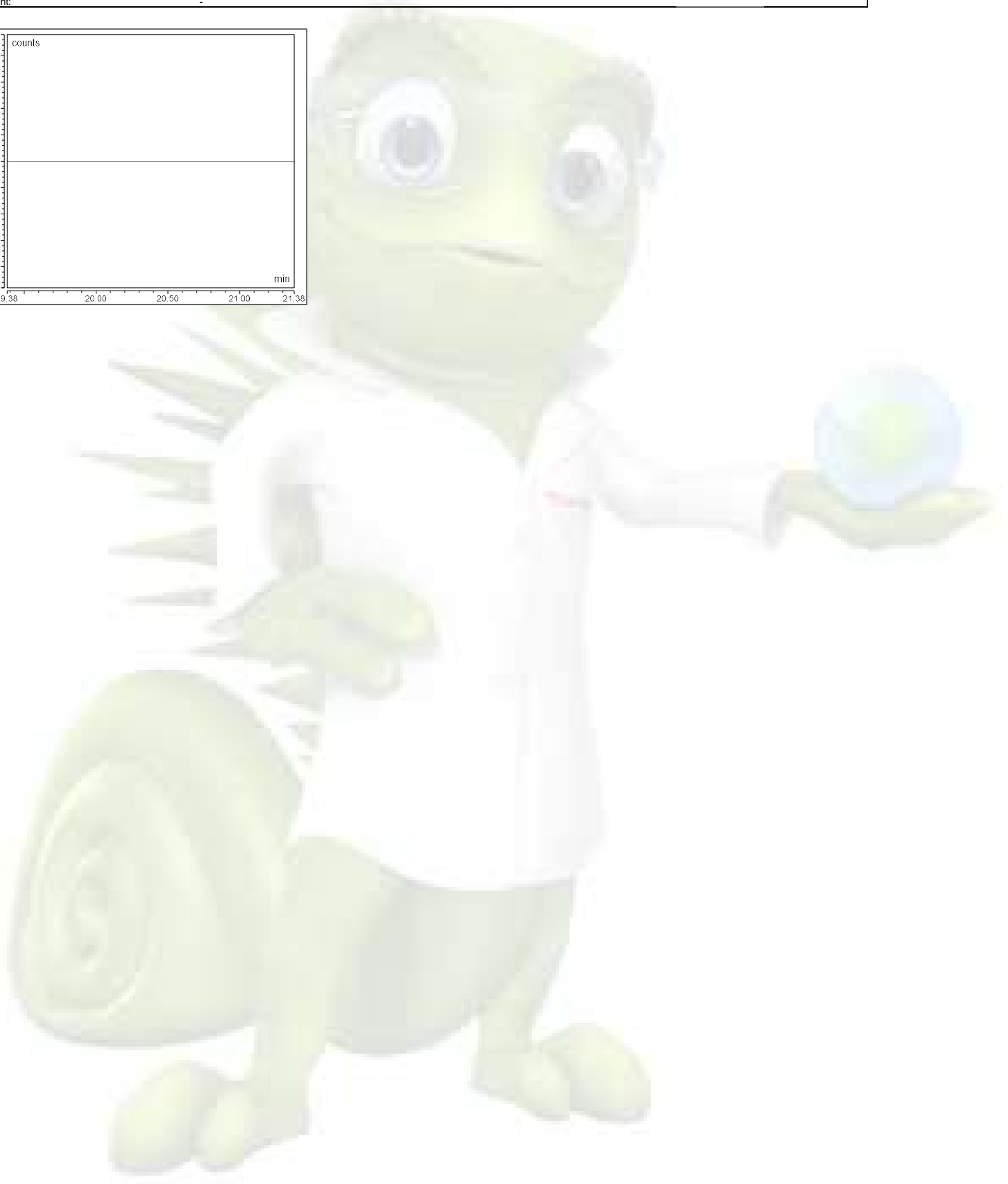
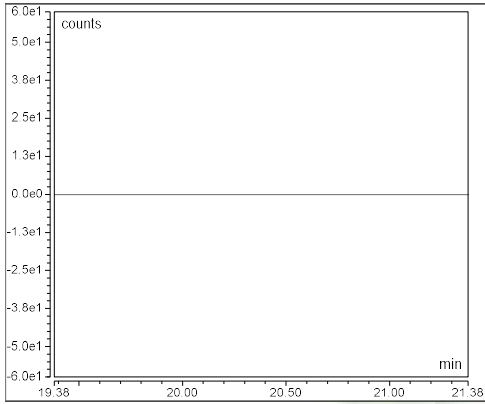
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

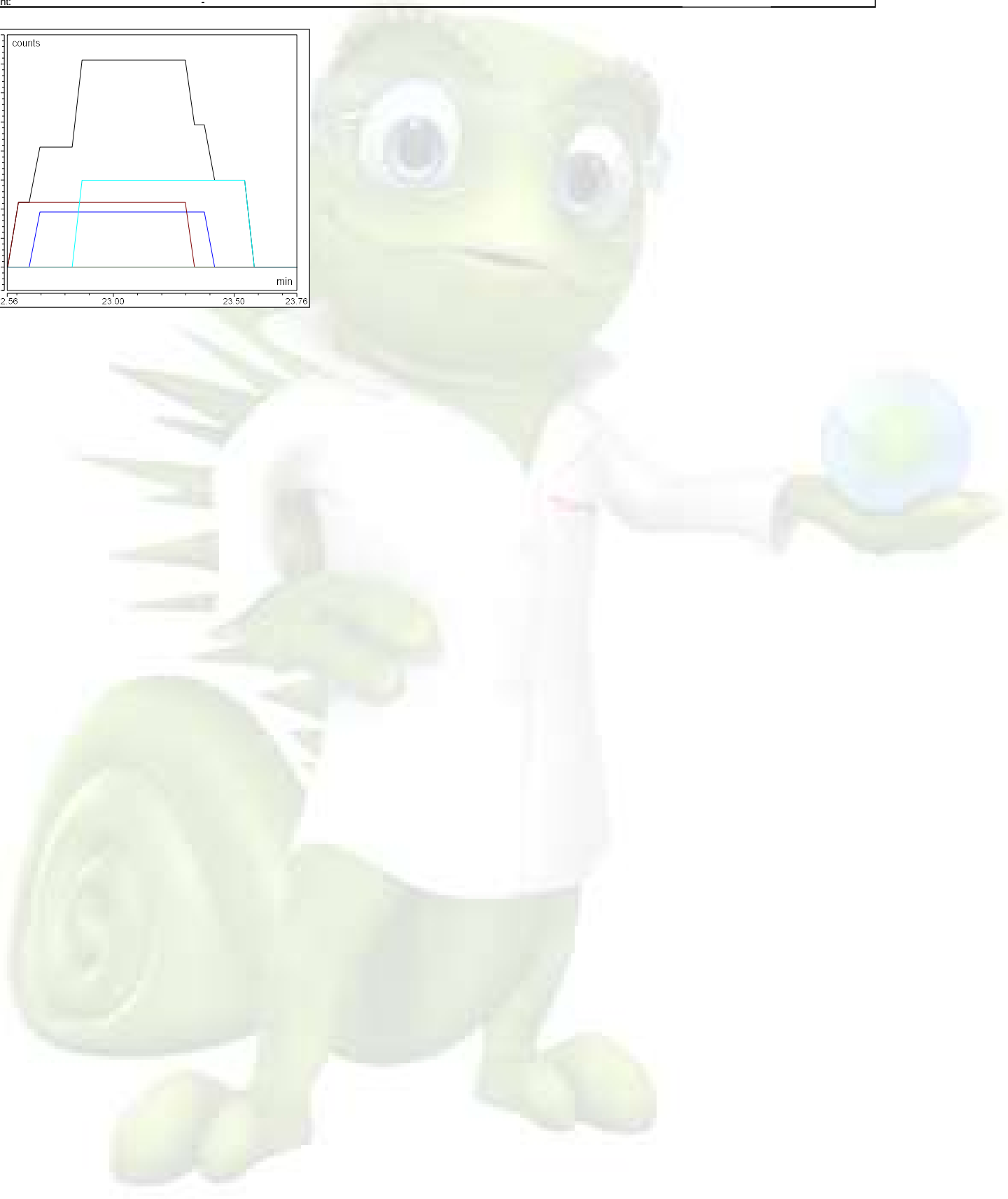
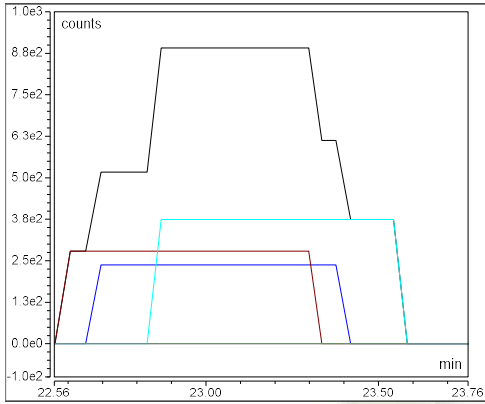
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

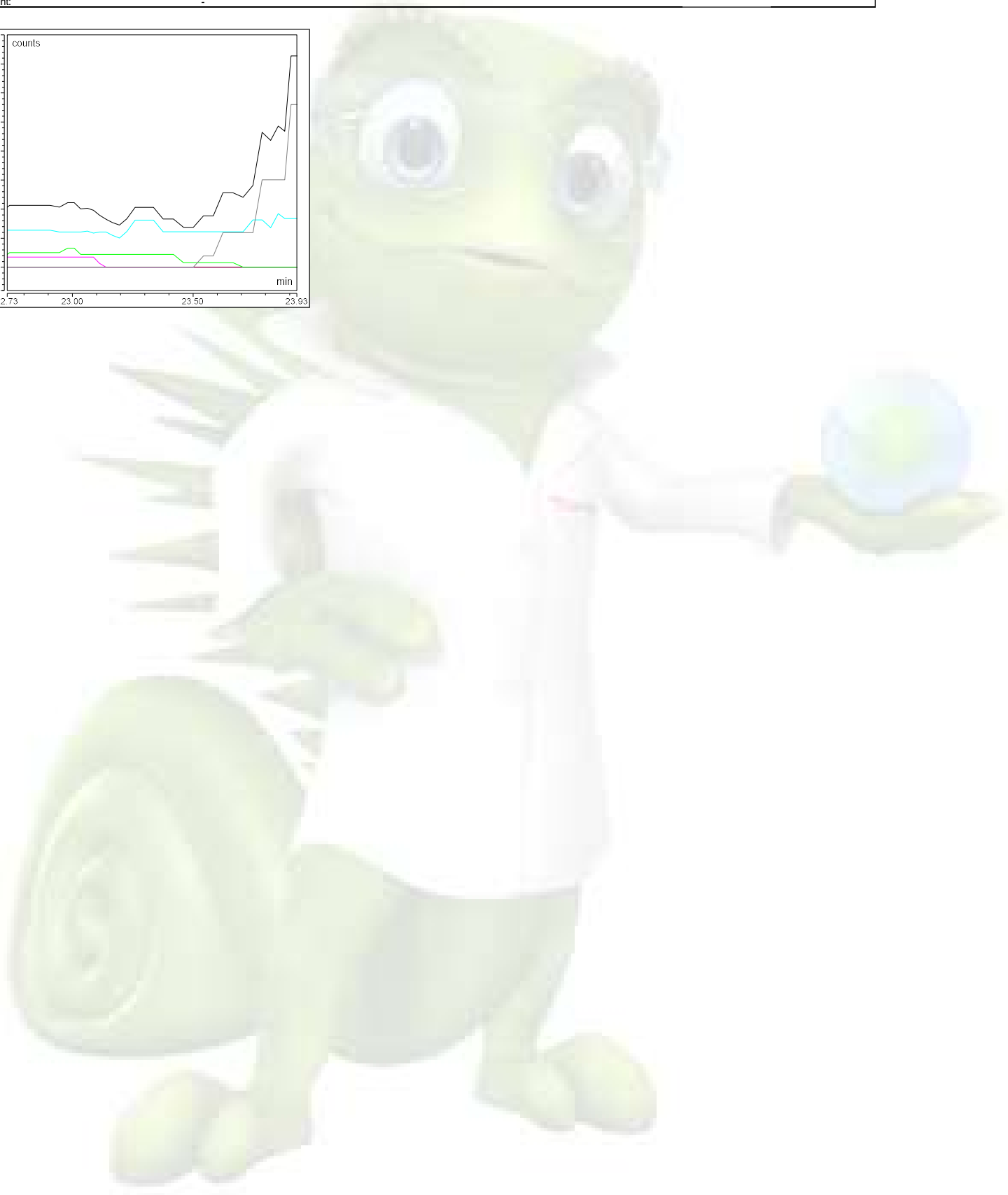
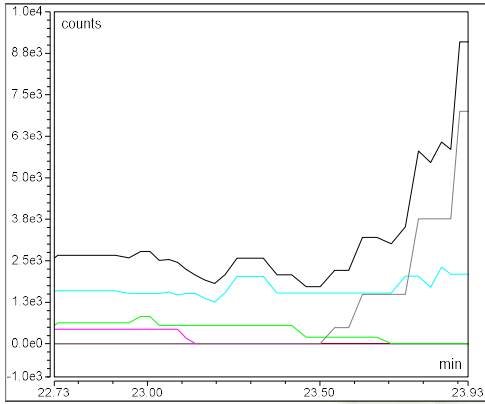
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

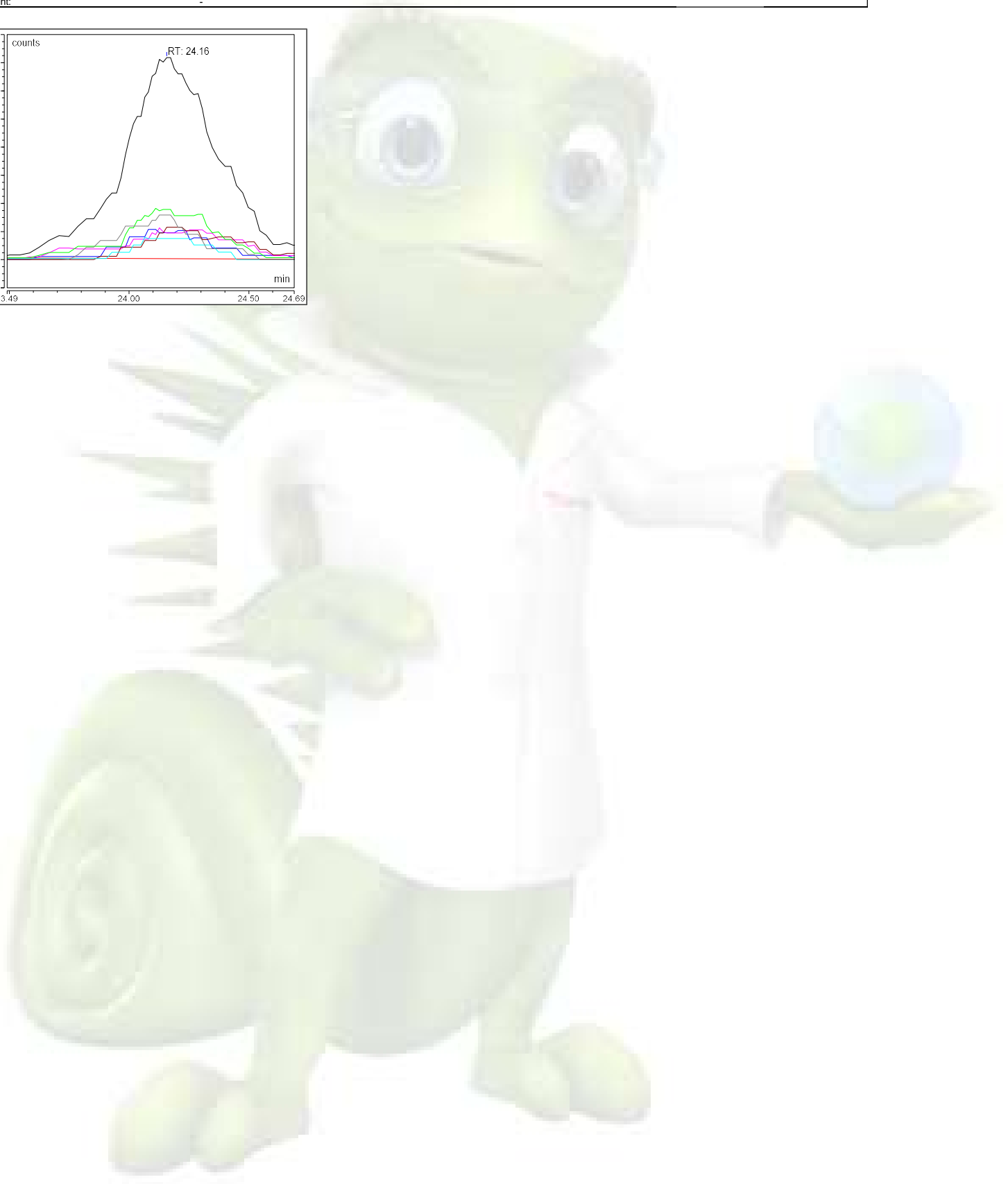
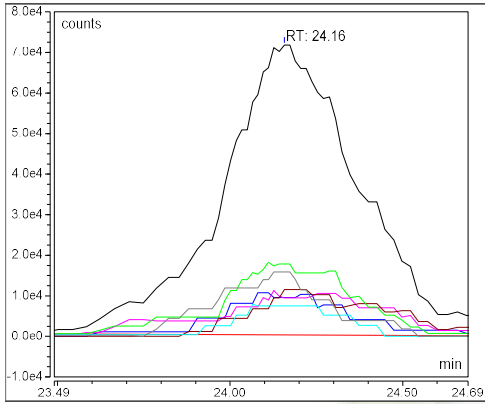
Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697 |
| Retention time (min): | 24.09   |
| Expected Mass:        | 80335.080   |
| Area (count*min):     | 33639   |
| Intensity (counts):   | 71494   |
| Amount:               | -   |



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

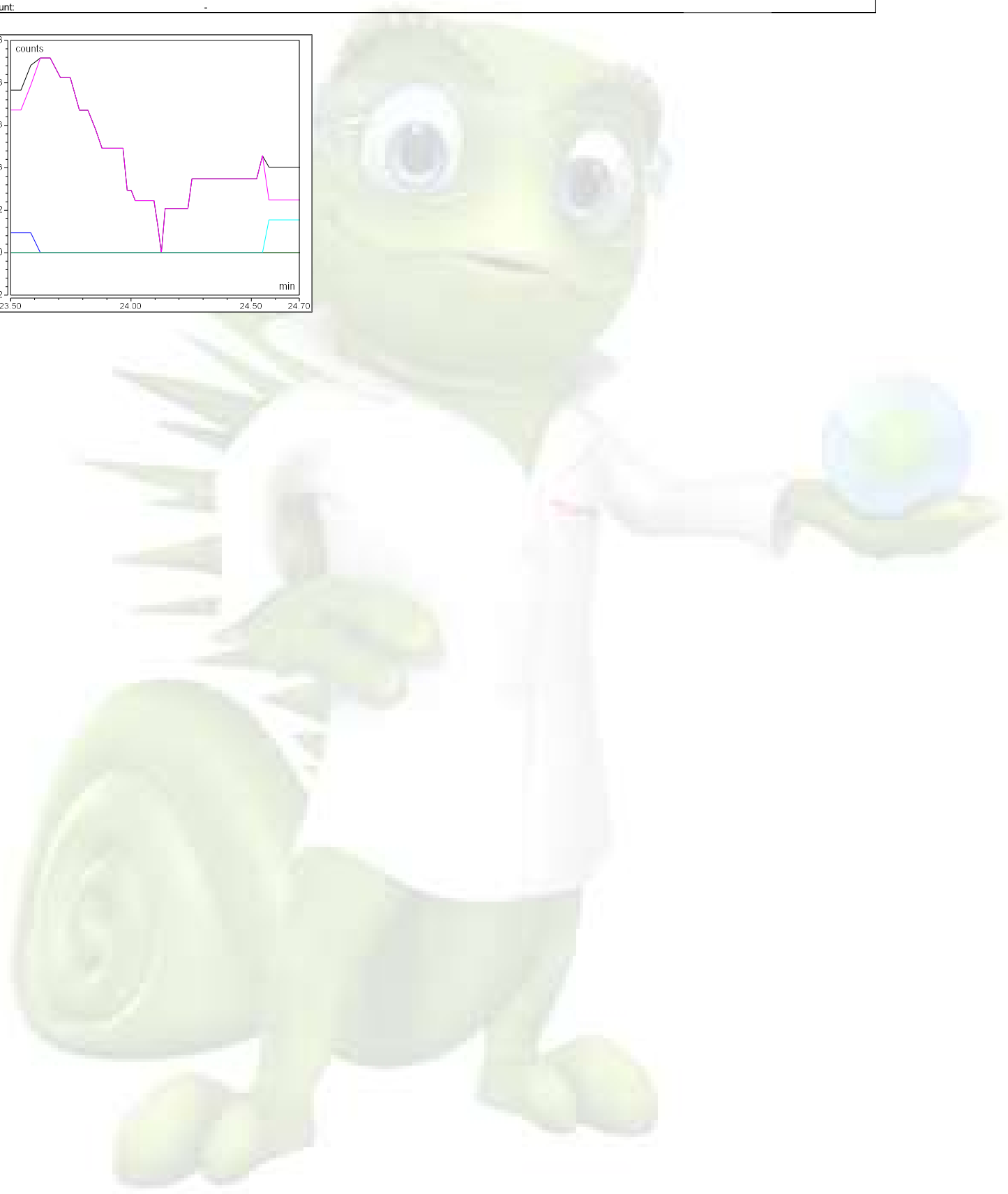
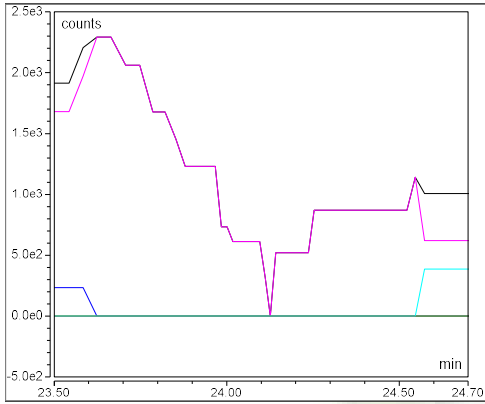
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

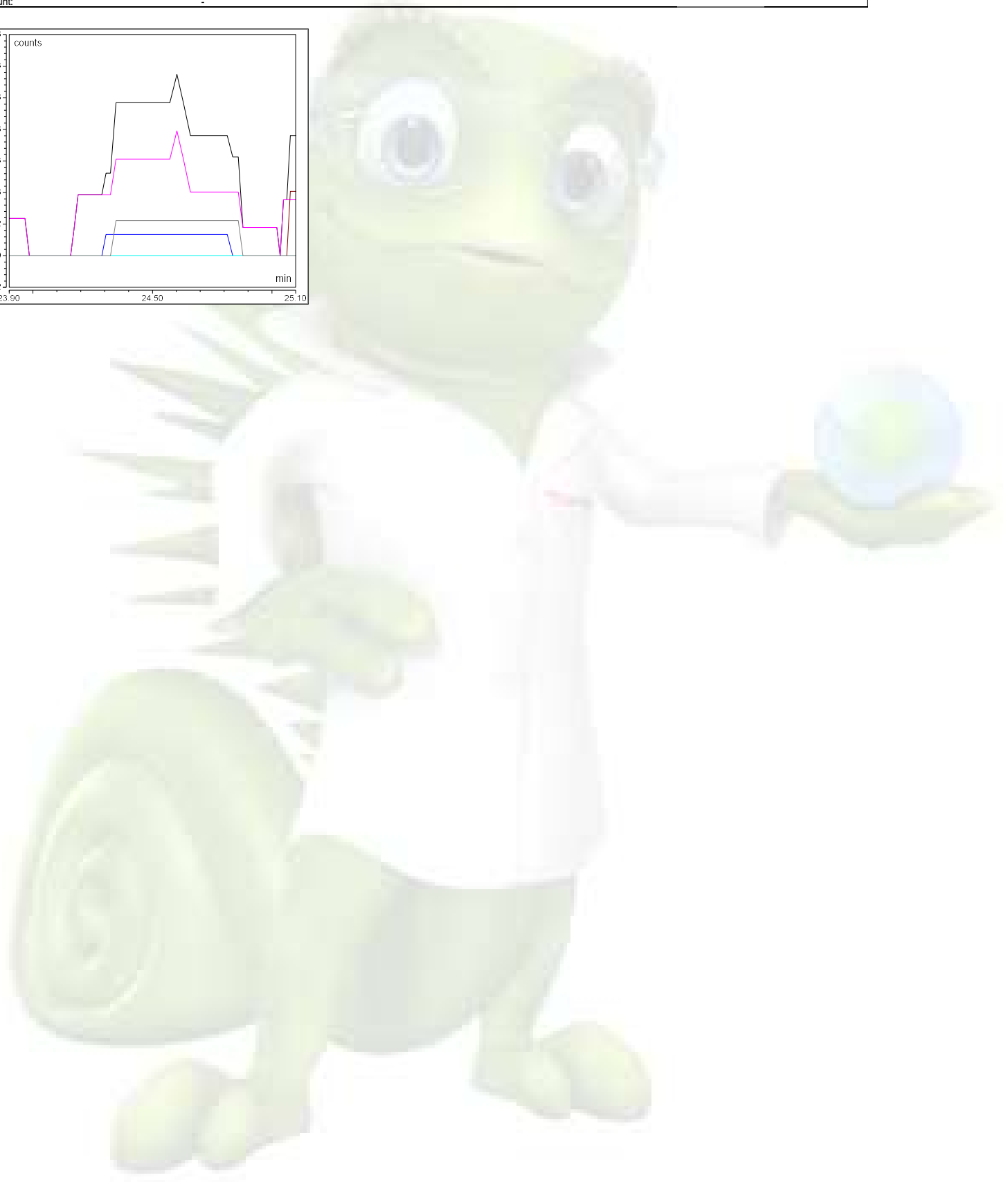
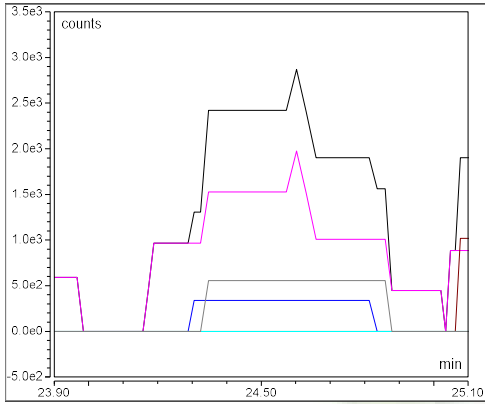
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

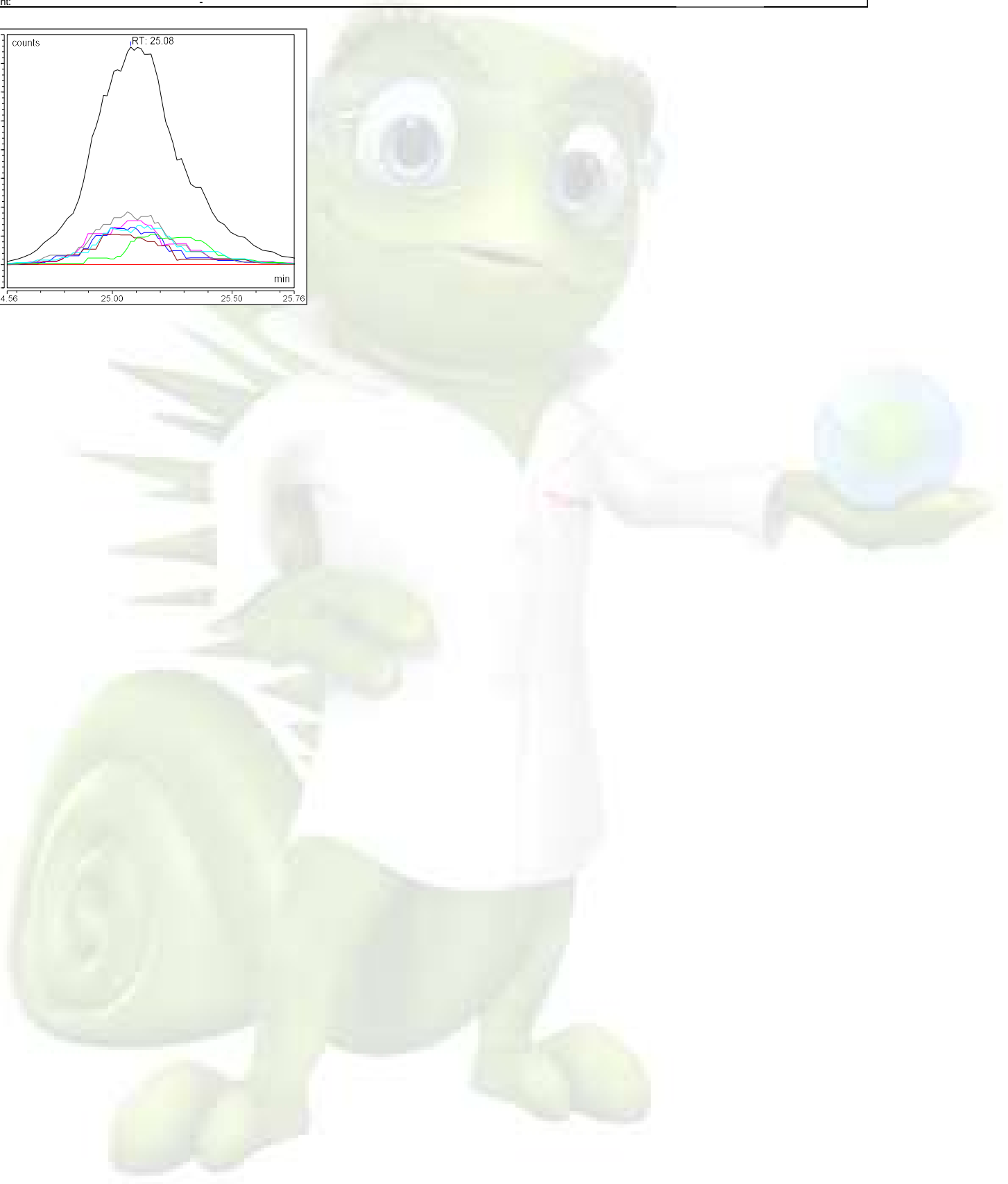
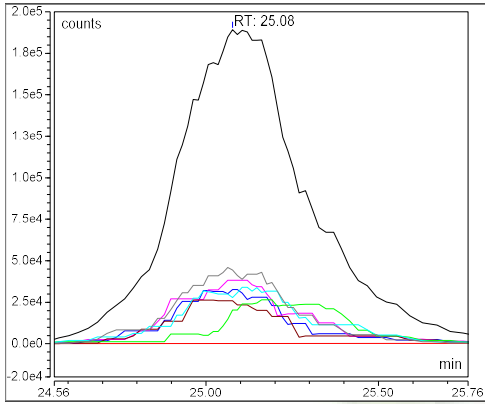
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): 84282

Intensity (counts): 189087

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

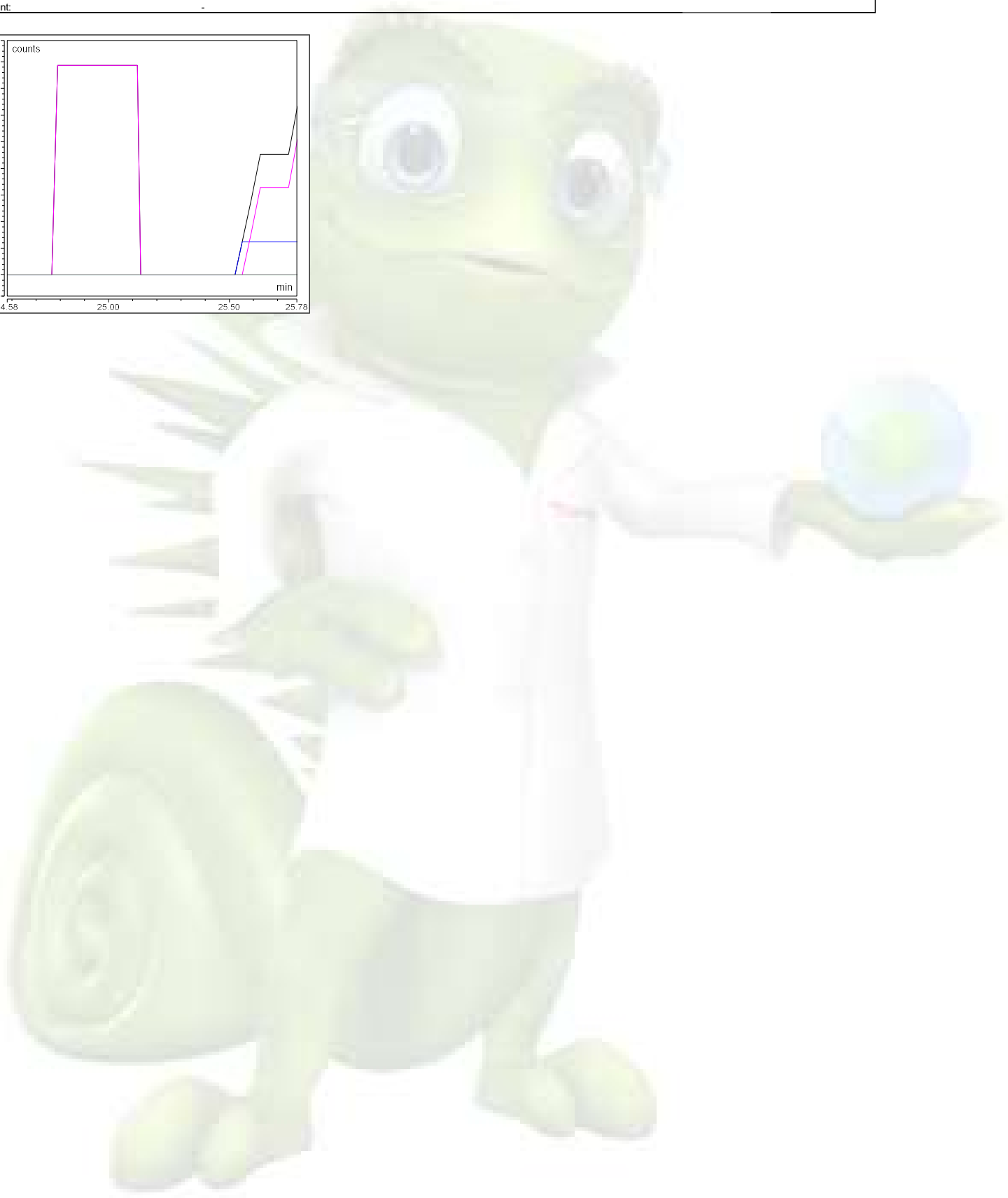
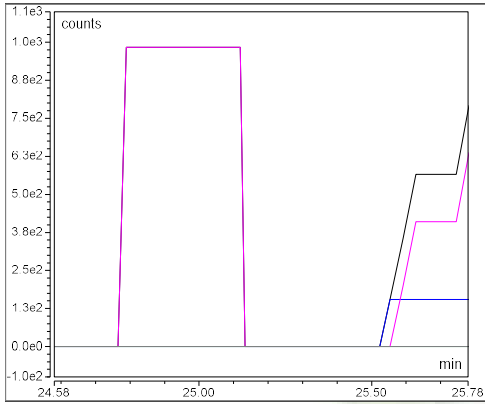
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

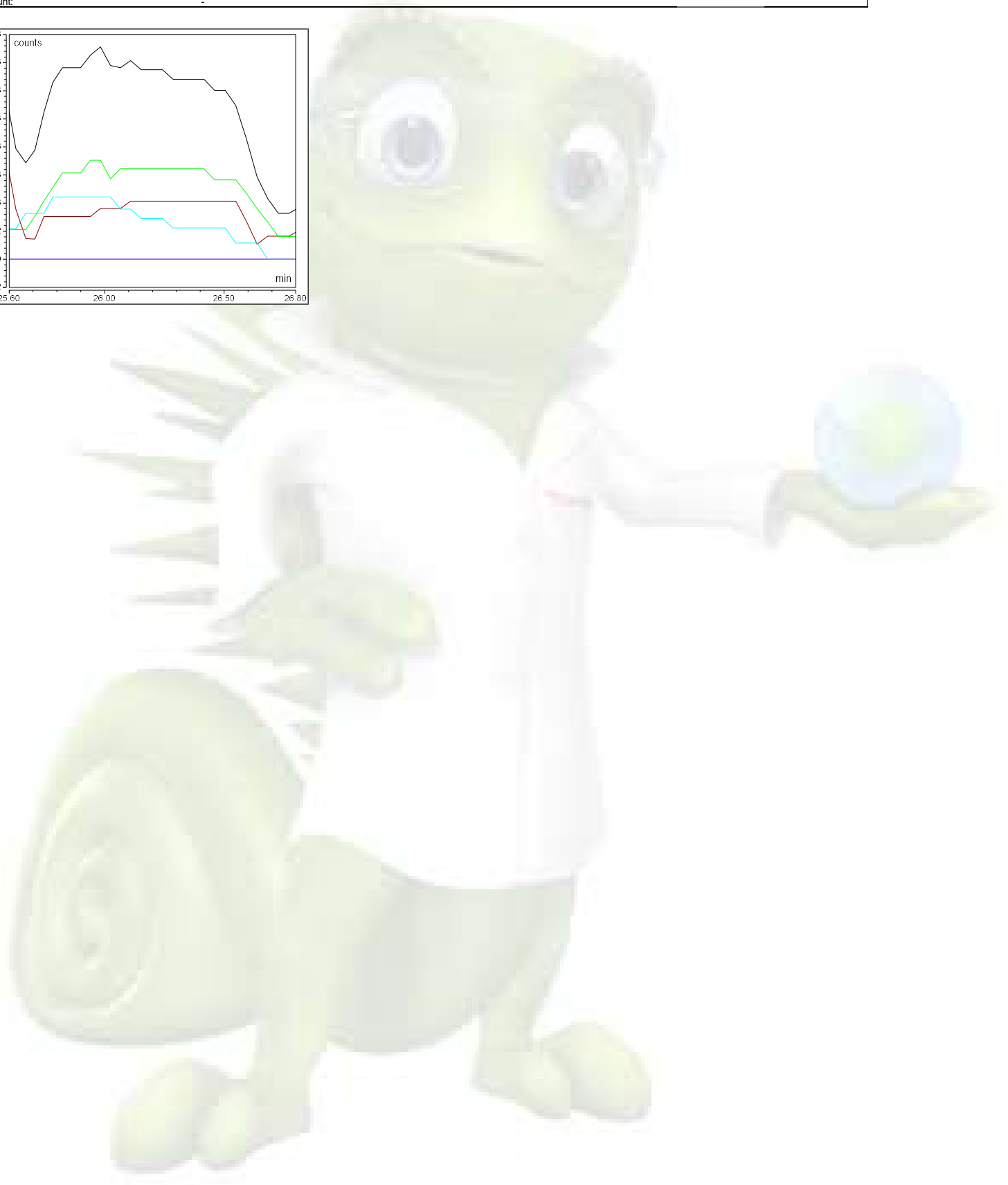
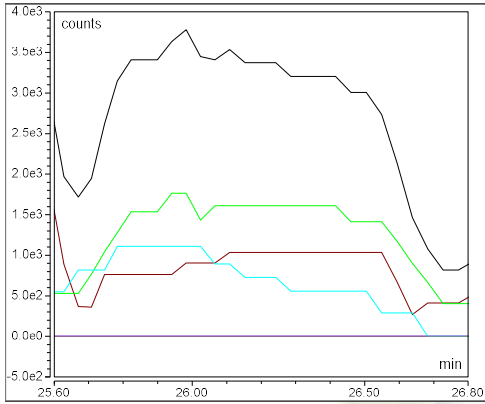
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

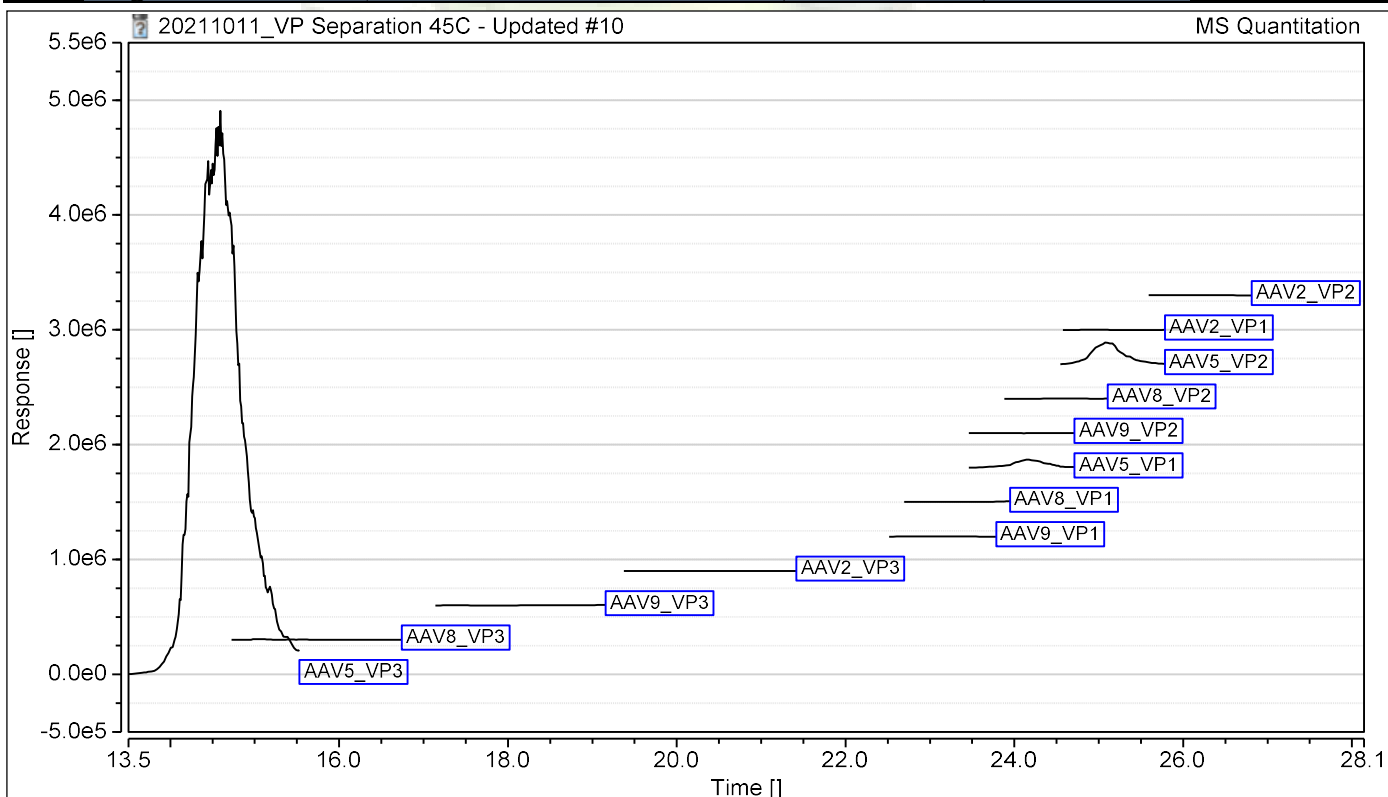
Amount: -



## Chromatogram and SST Results

| Injection Details    |  |                                |
|----------------------|--|--------------------------------|
| Injection Name:      | 20211011_AAV5 E_45C_65-59B_Mod1_1ug_1  | Run Time (min): <b>44.99</b>   |
| Vial Number:         | R:A7   | Injection Volume: <b>3.17</b>  |
| Injection Type:      | Unknown  |                                |
| Calibration Level:   |  |                                |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                                |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: <b>1.0000</b> |
| Injection Date/Time: | 13/Oct/21 05:26  | Sample Weight: <b>1.0000</b>   |

### Chromatogram

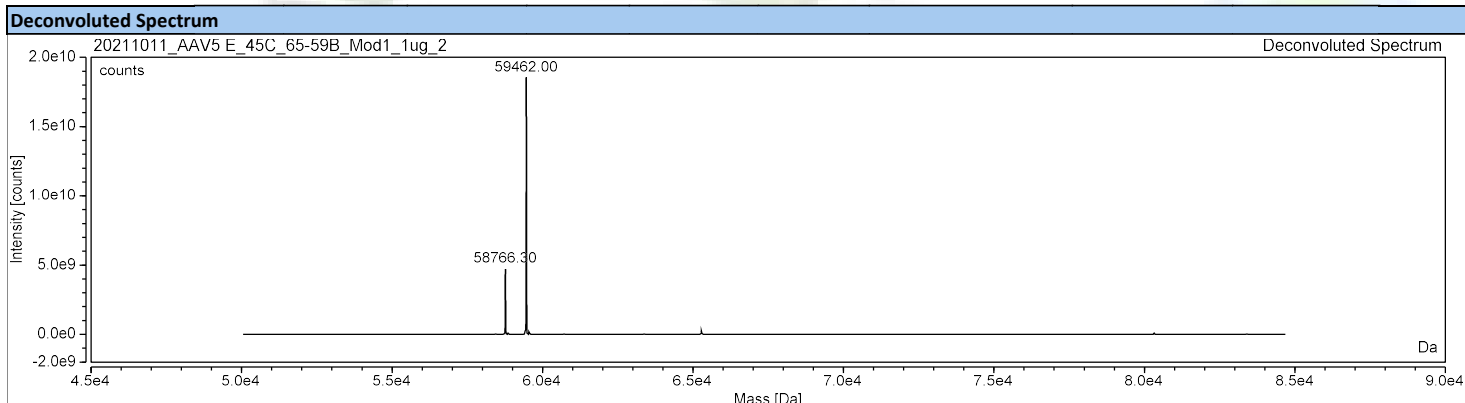
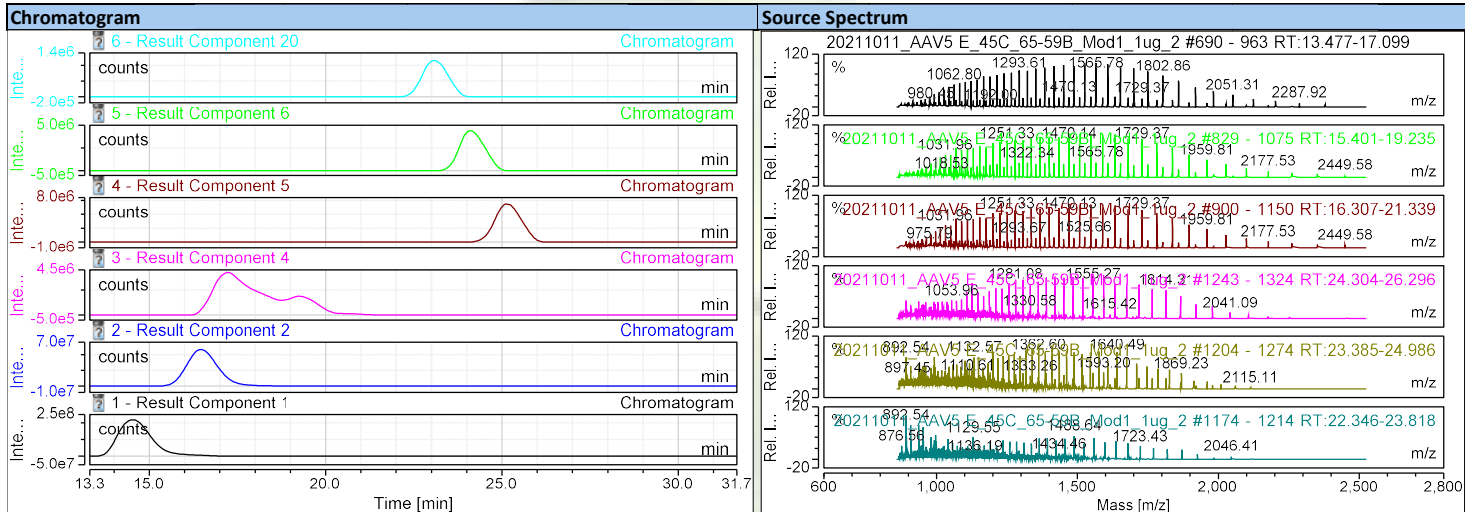


### SST Results

| No.  | Name | Inj. Condition | Peak | Test Result                 | Injection |
|--|------|----------------|------|-----------------------------|-----------|
| <b>Number of executed test cases: n.a.</b> |      |                |      | <b>Total Result: Passed</b> |           |

## Protein Deconvolution Summary

| Injection Details |                                       |                            |
|-------------------|---------------------------------------|----------------------------|
| Injection Name:   | 20211011_AAV5 E_45C_65-59B_Mod1_1ug_2 | Injection Volume:          |
| Vial Number:      | R:A7                                  | Instrument Method:         |
| Injection Type:   | Unknown                               | Processing Method:         |
| Run Time (min):   | 44.98                                 | Injection Date/Time:       |
|                   |                                       | MS Quantitative_45C_65-59B |



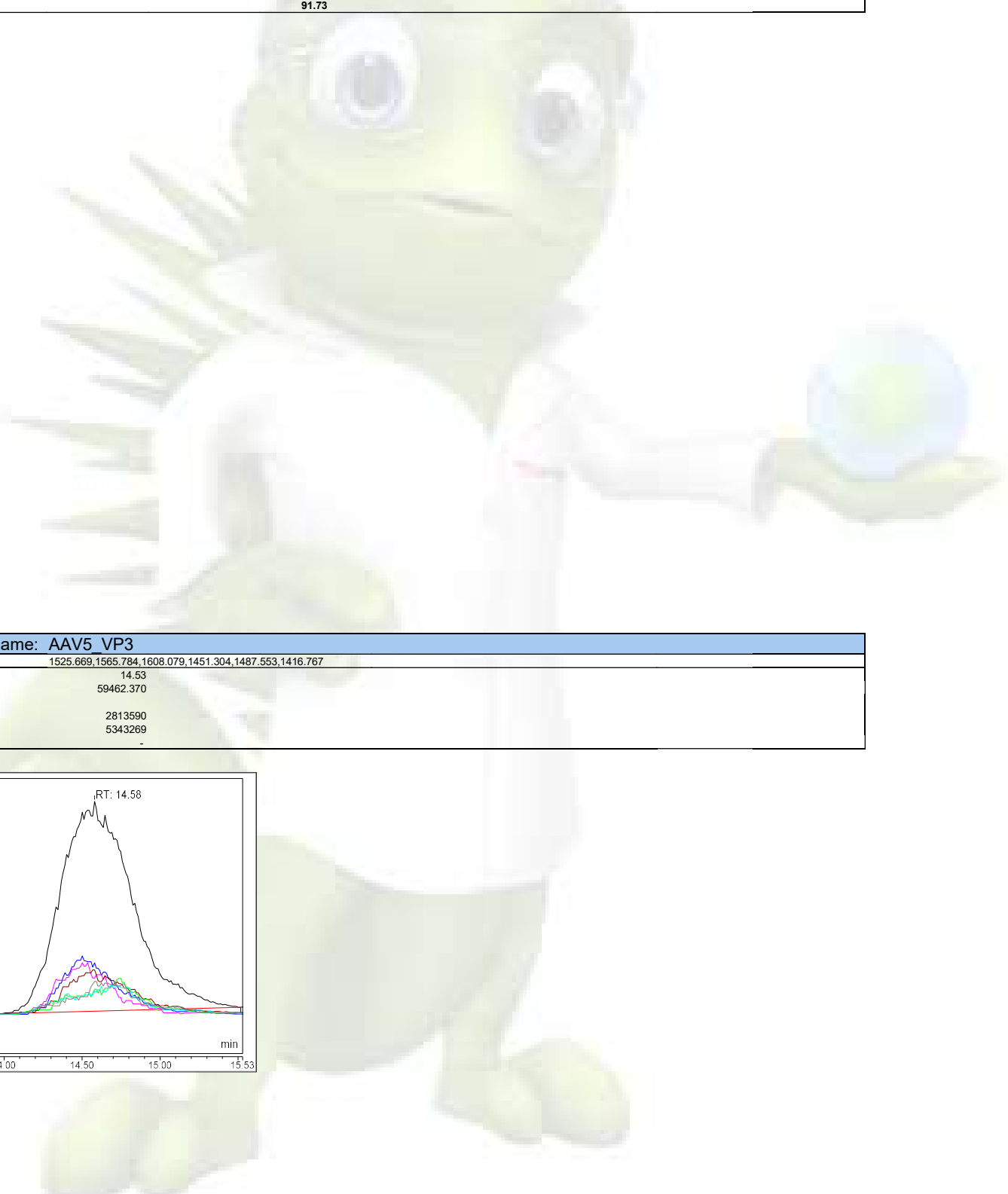
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

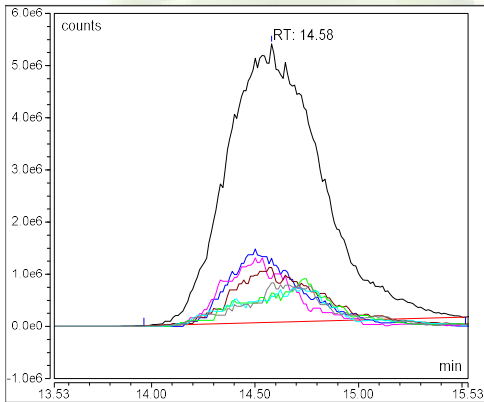
|  |   |  |
|--|---|--|
| <b>Injection Name:</b> 20211011_AAV5_E_45C_65-59B_Mod1_1ug | <b>Injection Volume:</b> 3.17   |  |
| <b>Vial Number:</b> R:A7                                   | <b>Instrument Method:</b> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <b>Injection Type:</b> Unknown                             | <b>Processing Method:</b> MS Quantitative_45C_65-59B                            |  |
| <b>Run Time (min):</b> 44.98                               | <b>Injection Date/Time:</b> 13/Oct/21 06:12                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV5_VP3     | 14.579                | 2813589.775        | 87.79              | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 | 59462.370 | PASS                           |
| AAV5_VP1     | 24.095                | 40080.572          | 1.25               | 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697 | 80335.080 | PASS                           |
| AAV5_VP2     | 25.069                | 86266.766          | 2.69               | 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727 | 65282.690 | PASS                           |
| <b>91.73</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <b>Quantitation ions:</b>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <b>Retention time (min):</b> | 14.53   |
| <b>Expected Mass:</b>        | 59462.370   |
| <b>Area (count*min):</b>     | 2813590   |
| <b>Intensity (counts):</b>   | 5343269   |
| <b>Amount:</b>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

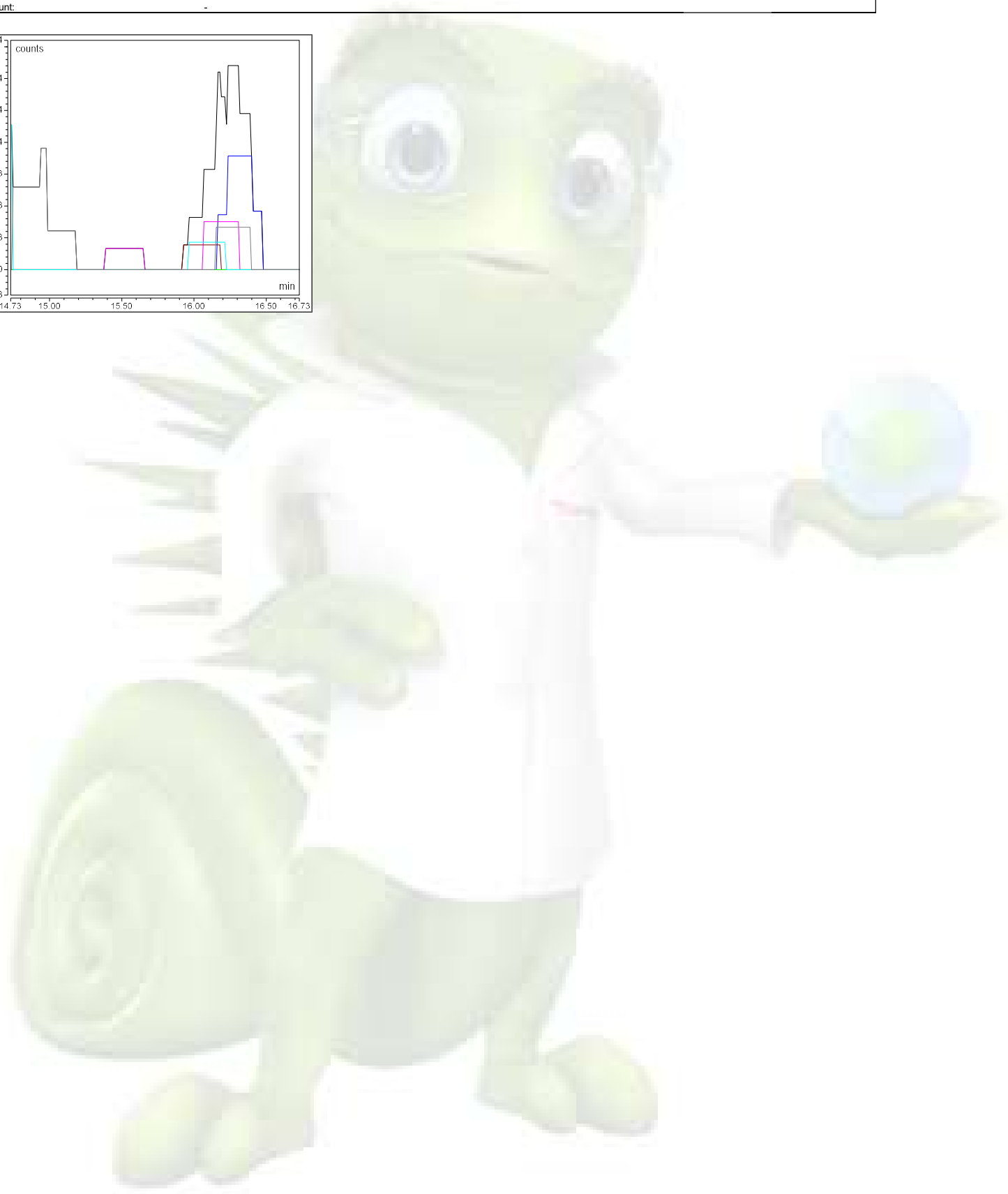
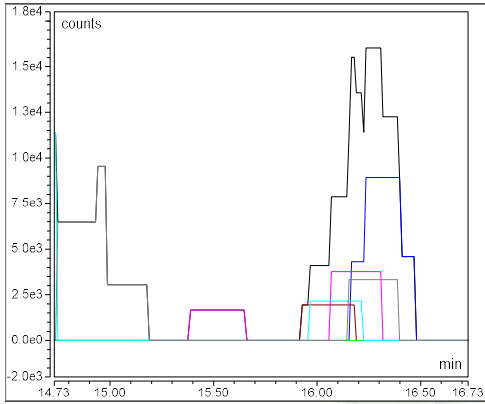
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

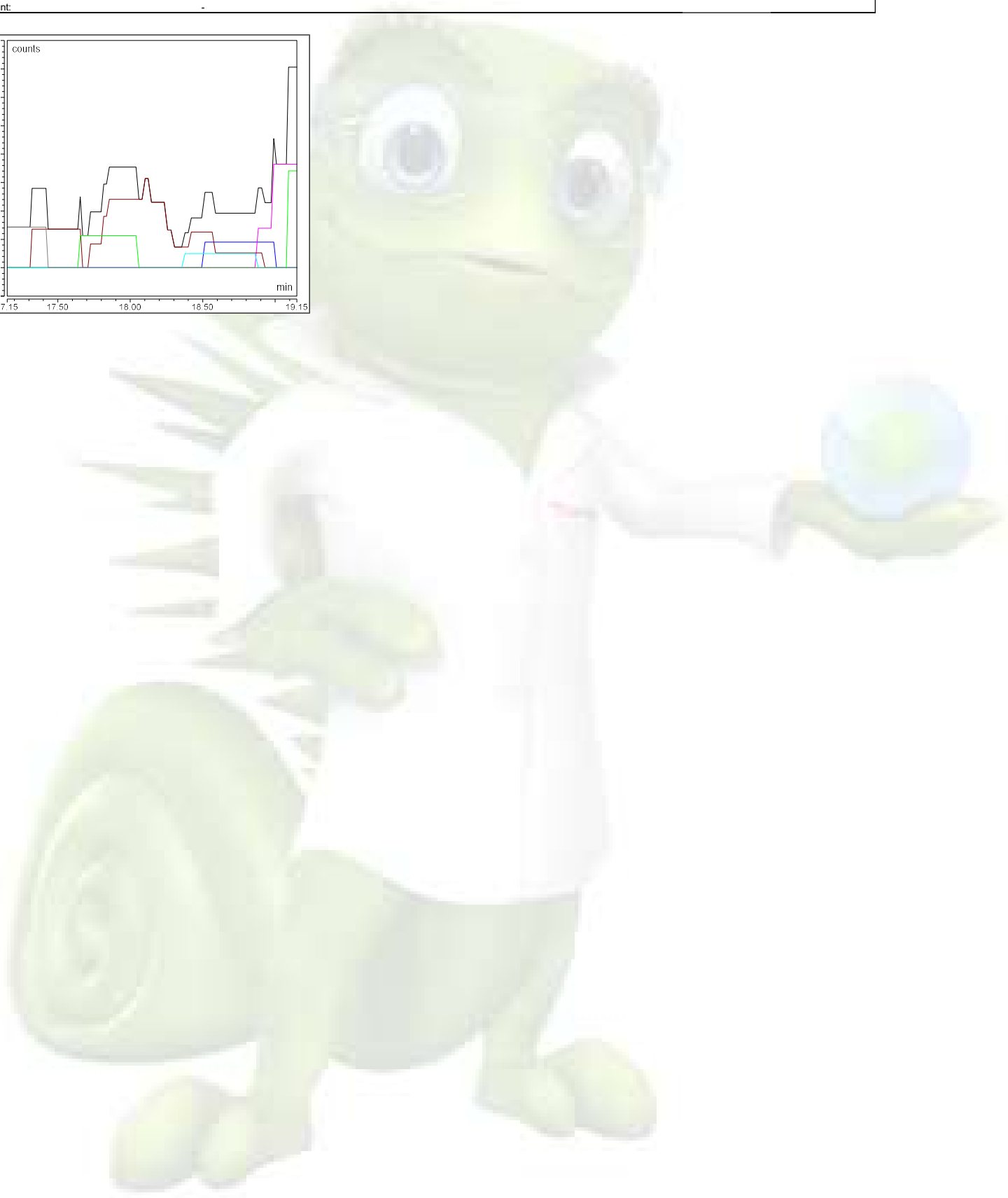
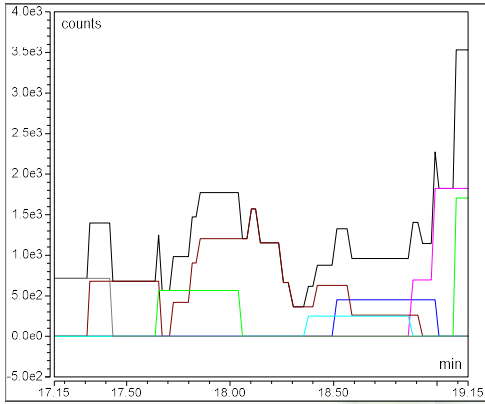
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

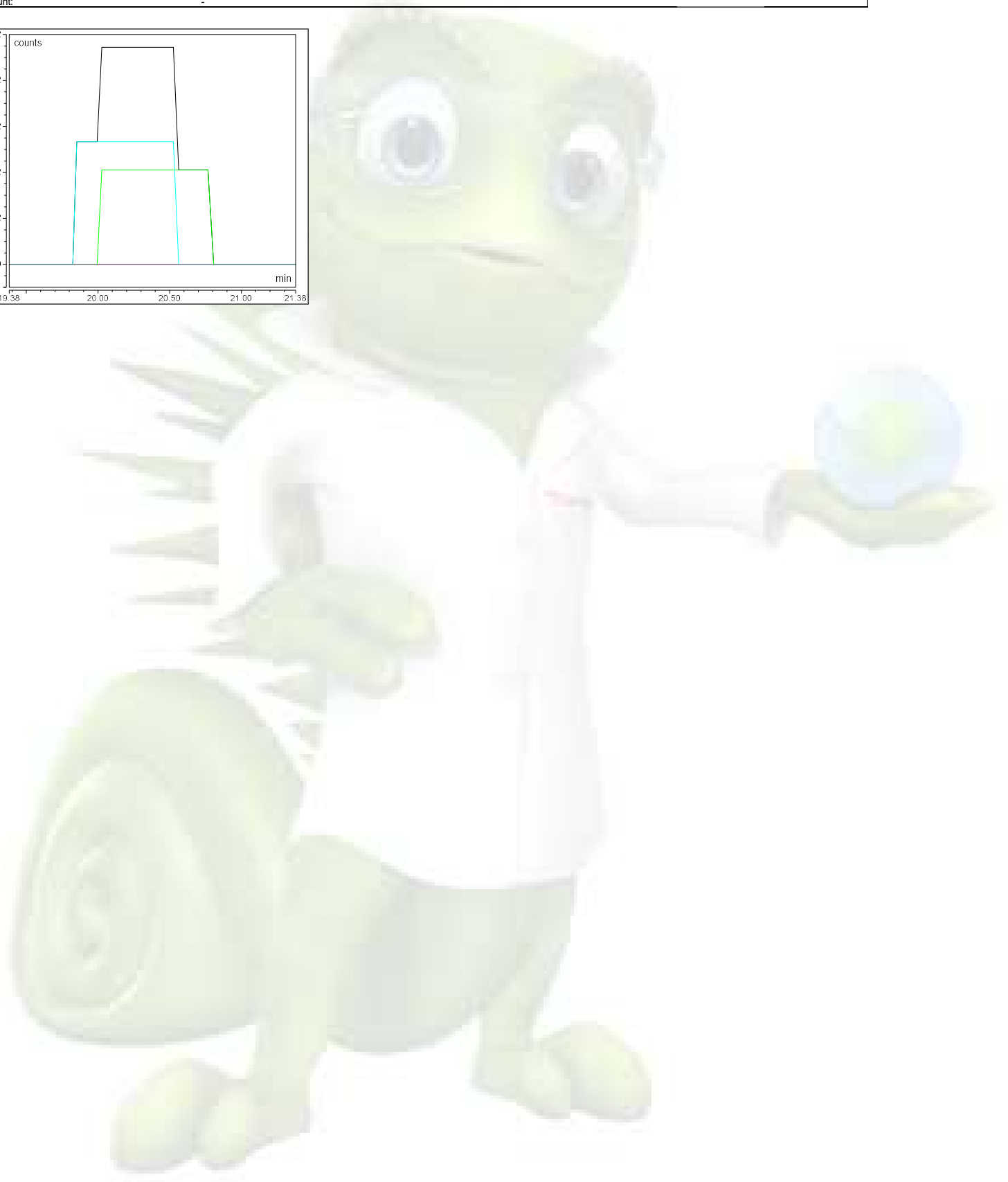
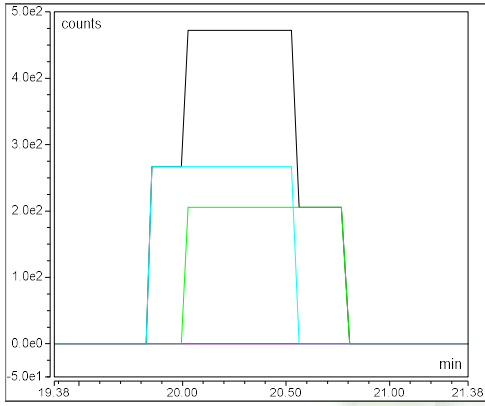
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

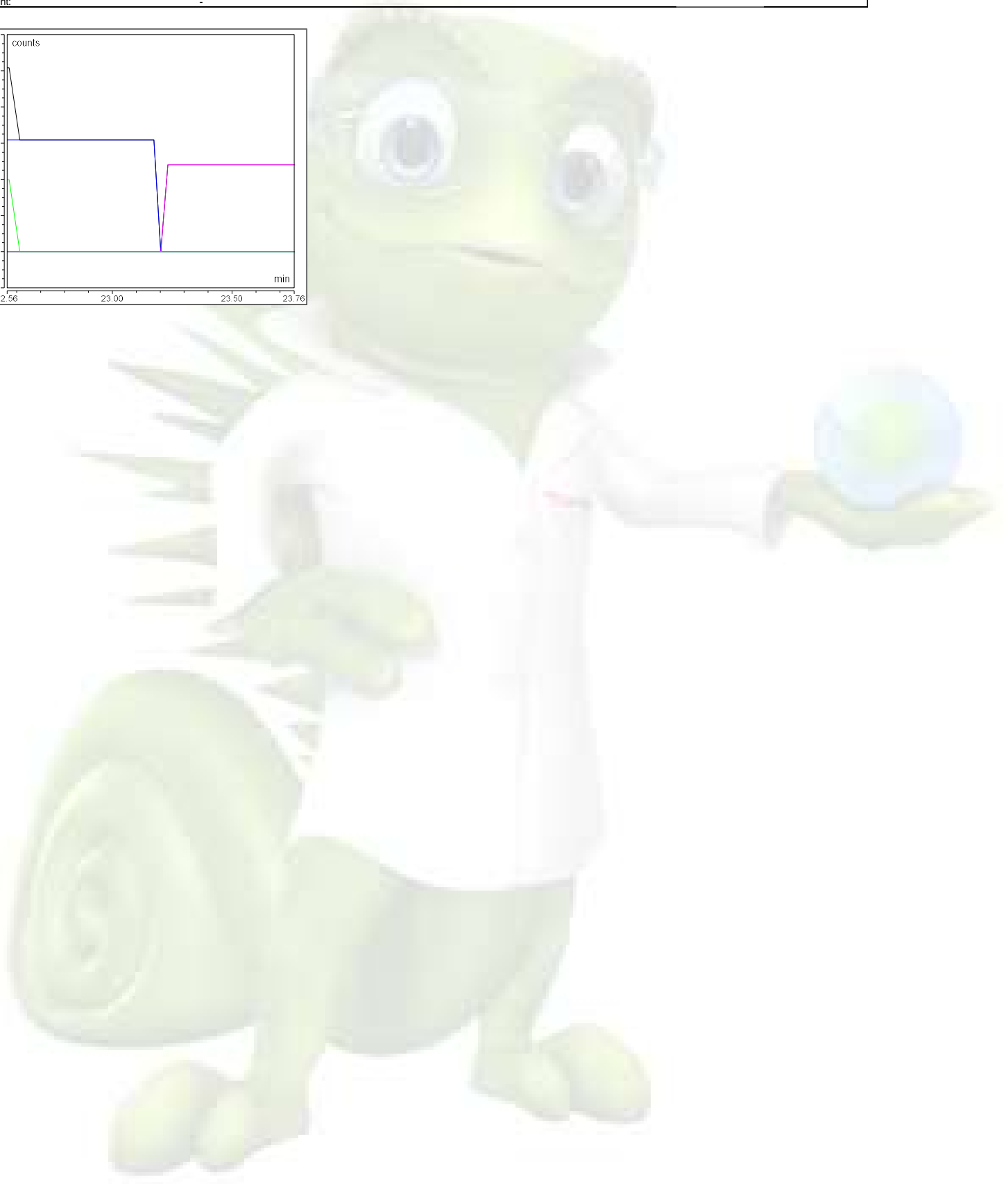
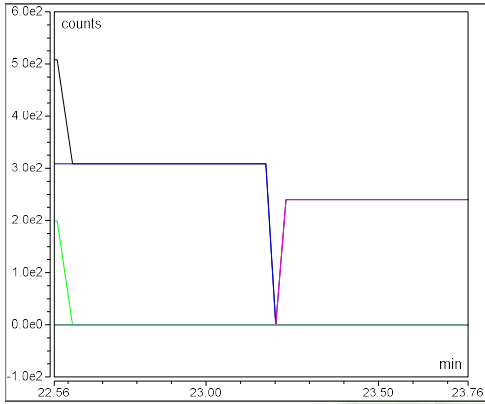
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

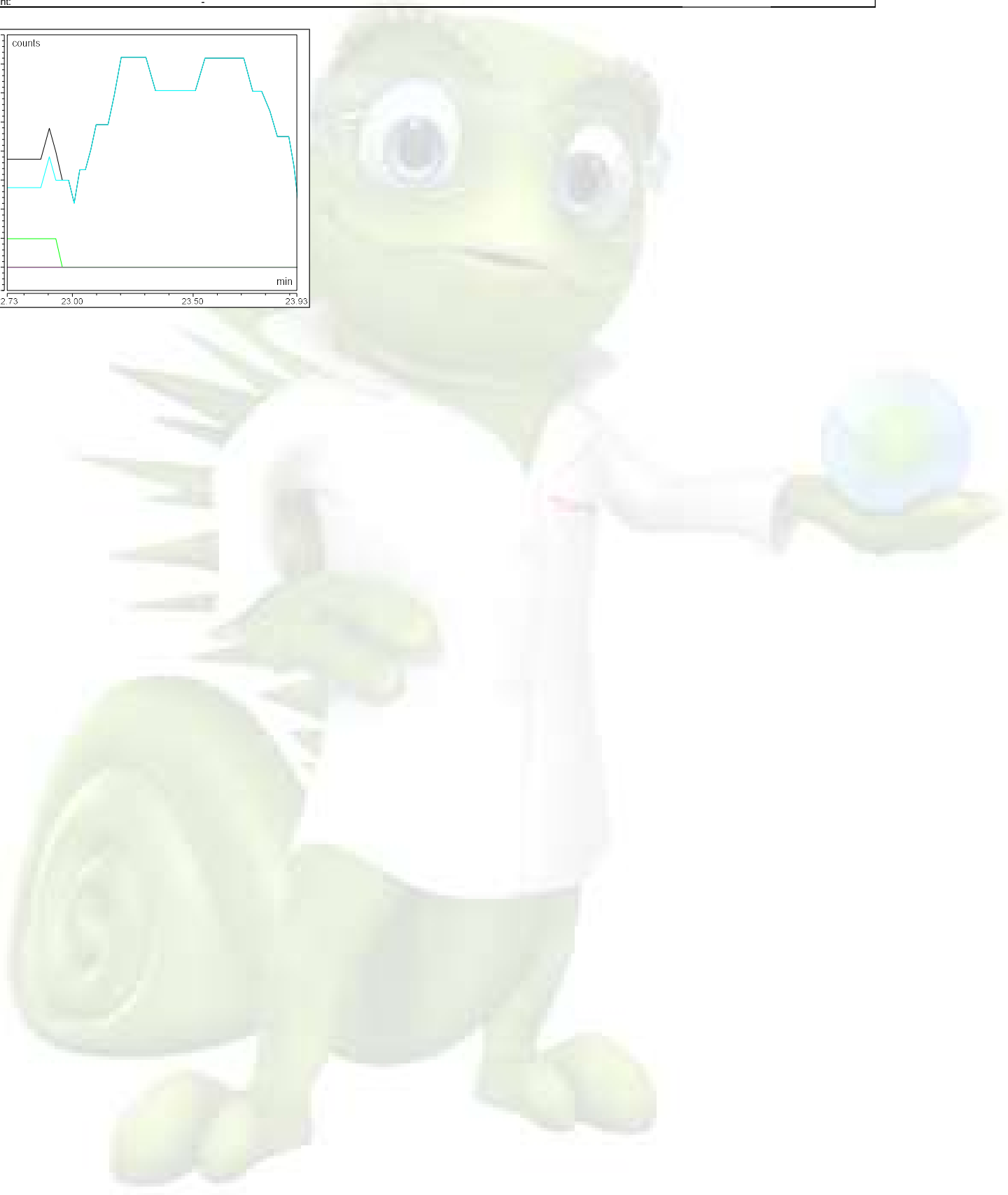
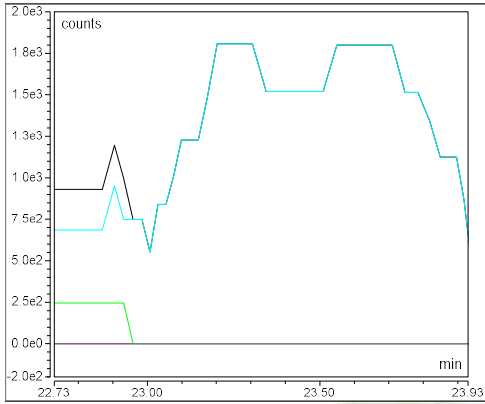
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

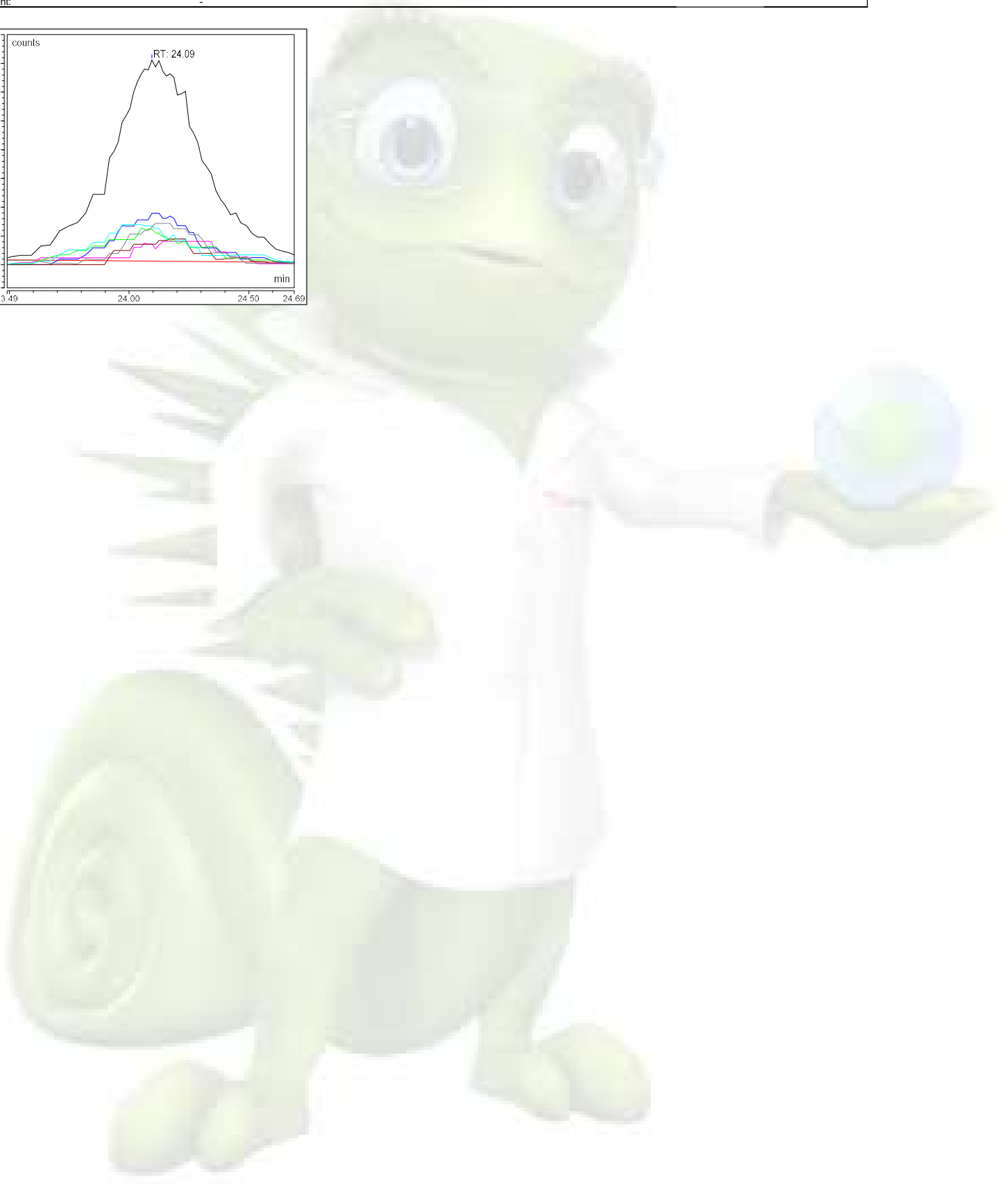
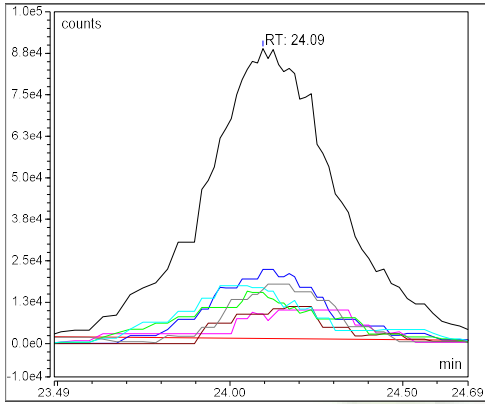
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): 40081

Intensity (counts): 87360

Amount: -



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

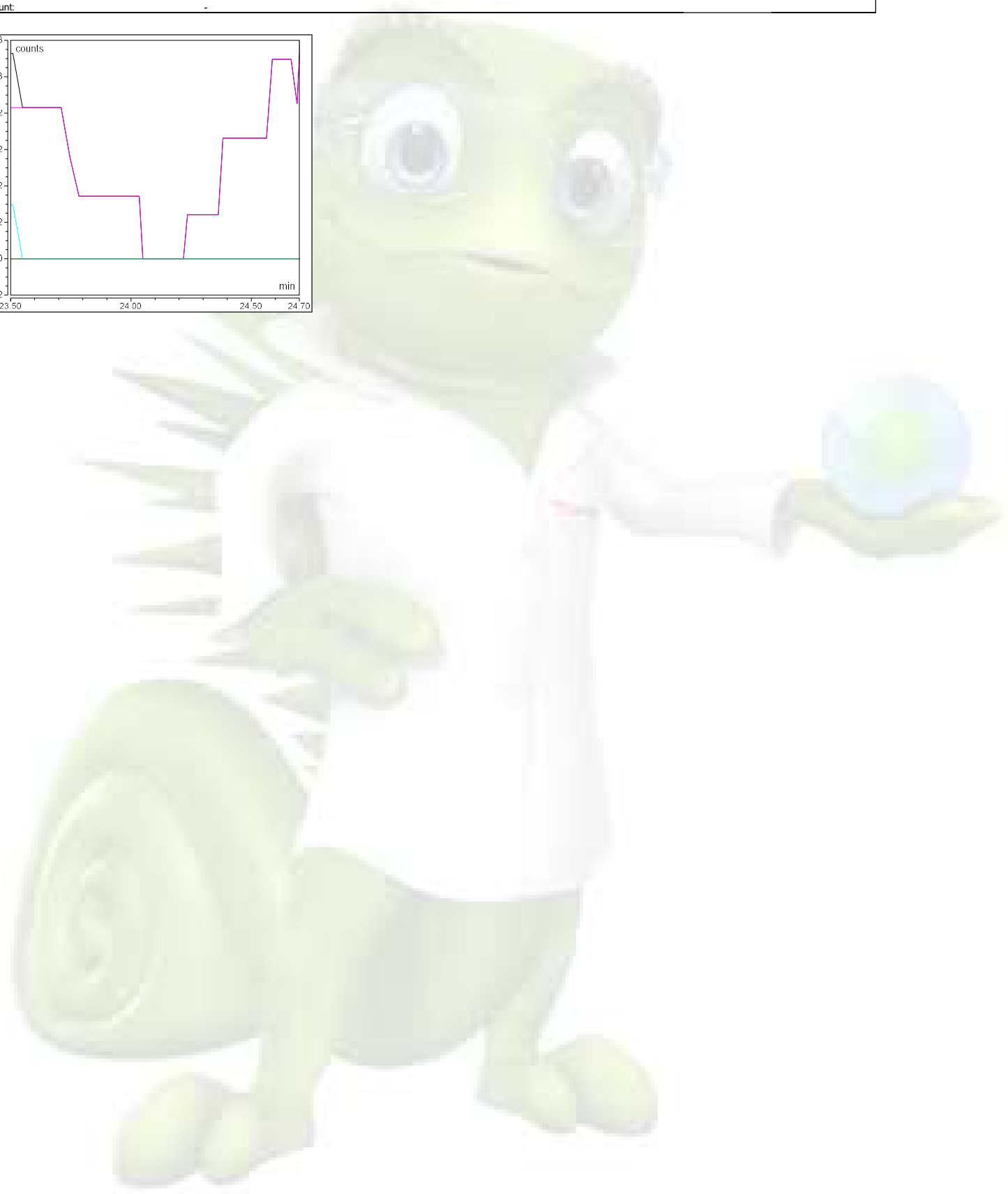
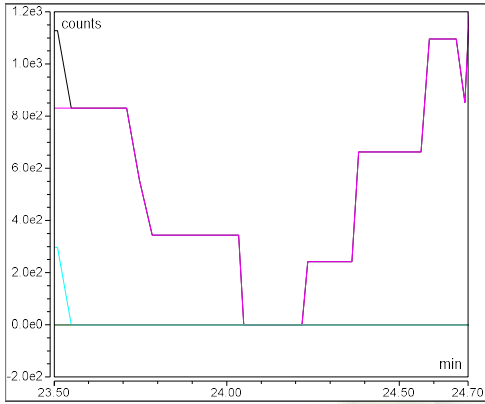
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

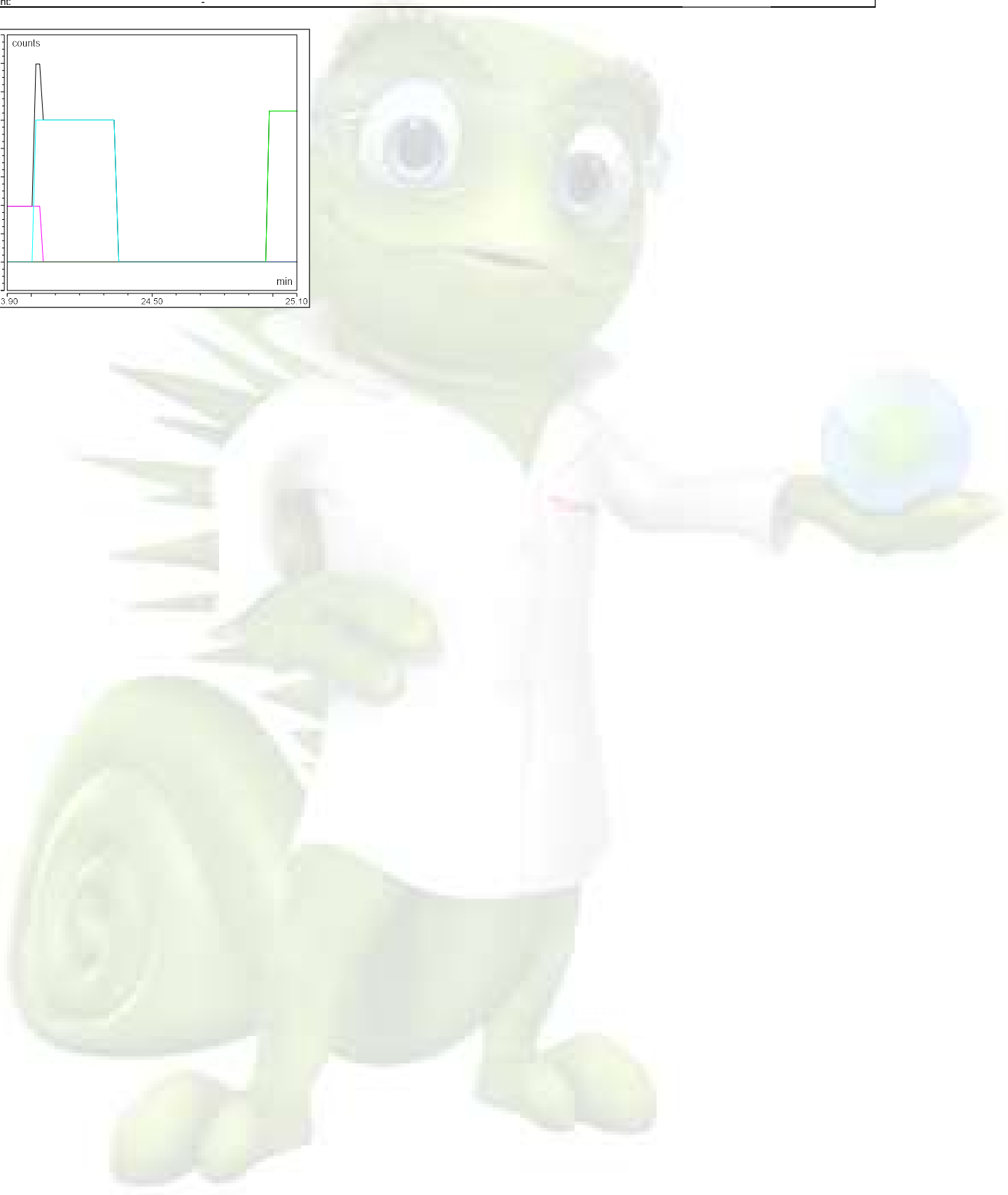
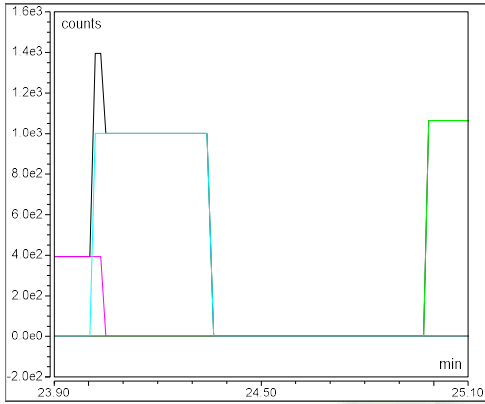
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

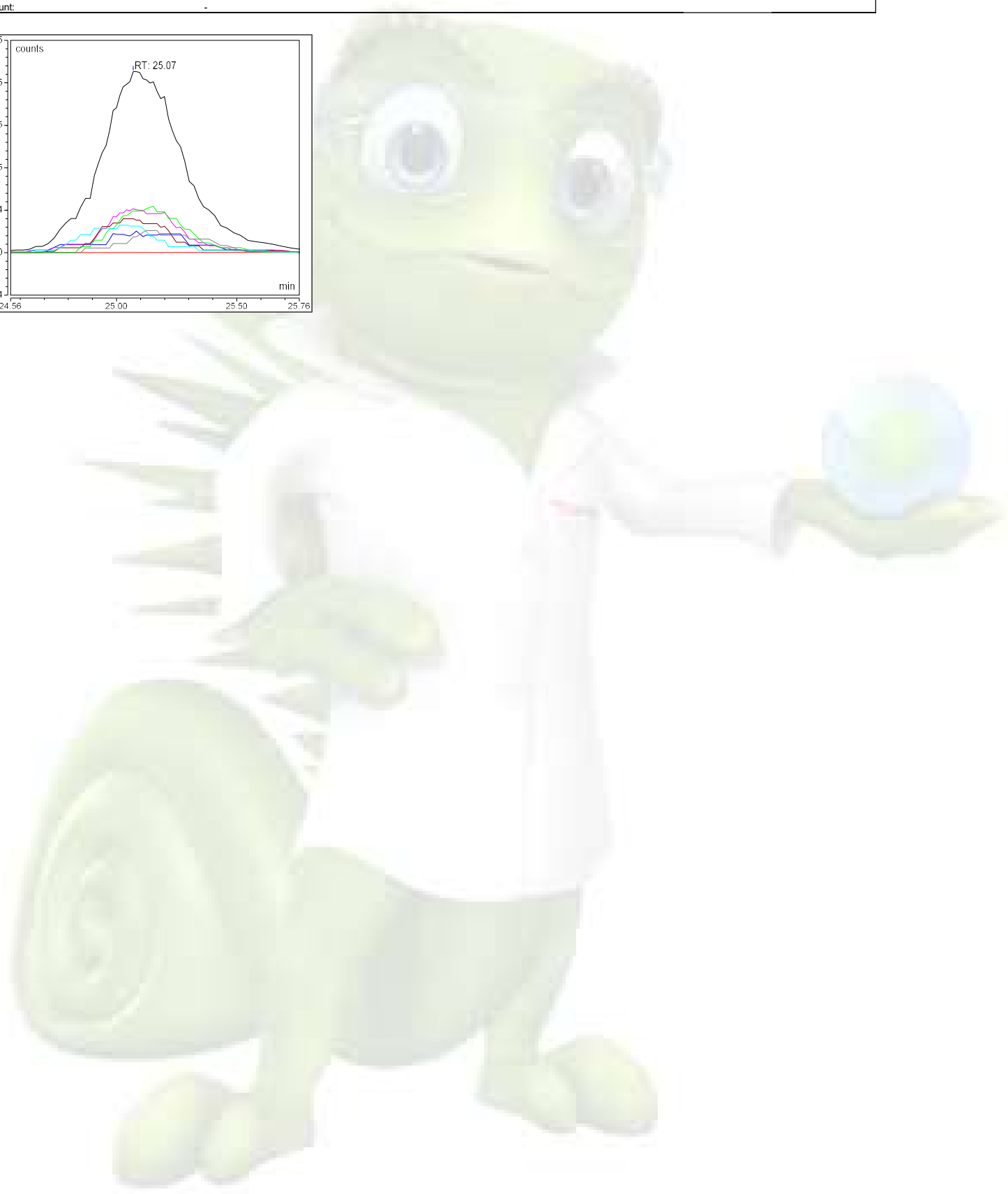
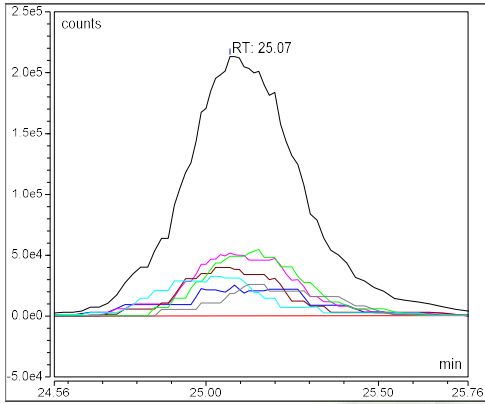
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): 86267

Intensity (counts): 213000

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

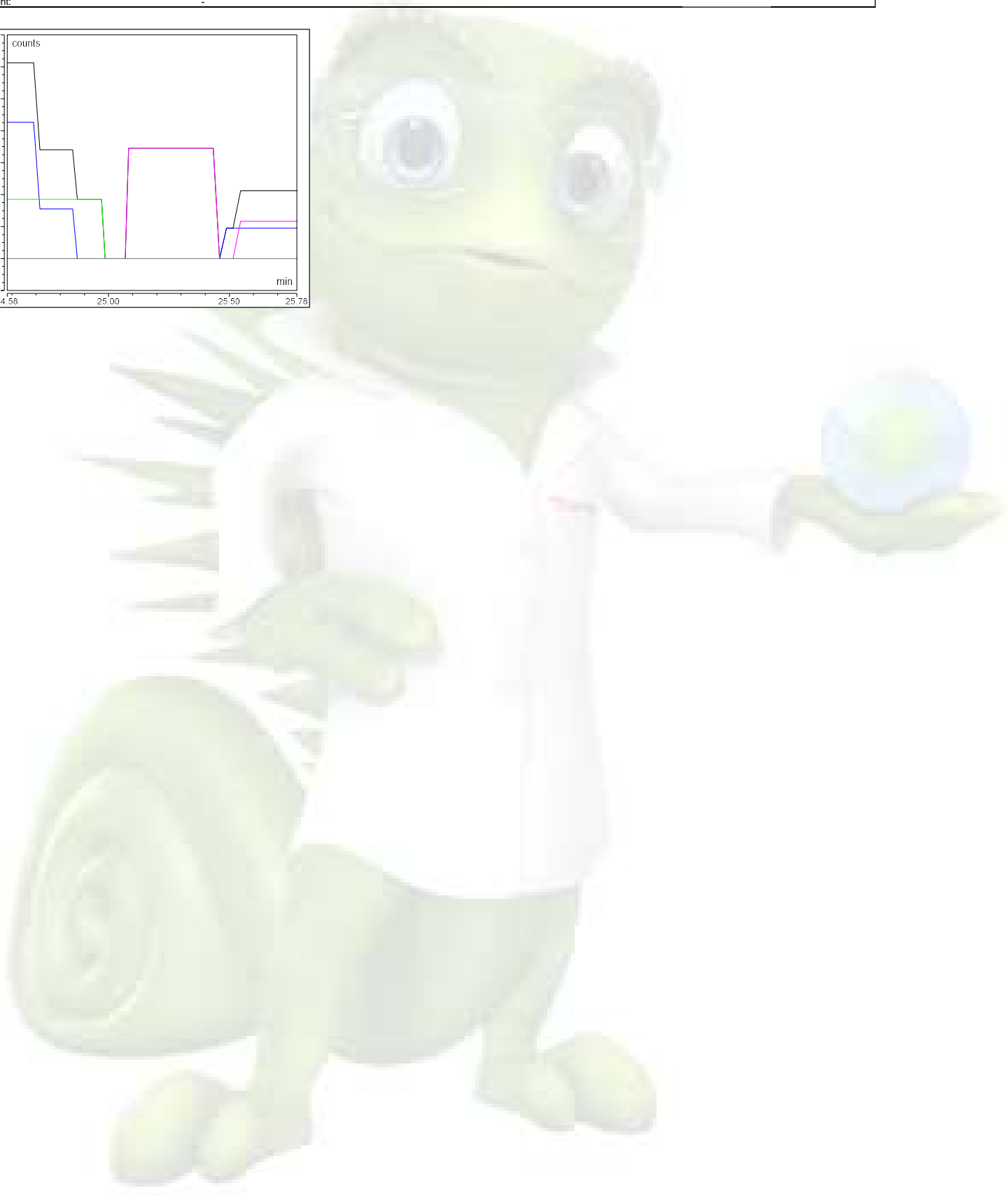
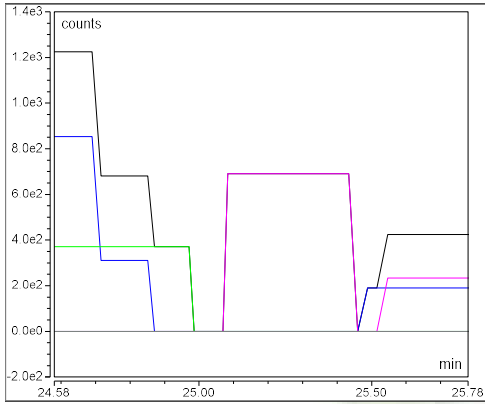
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

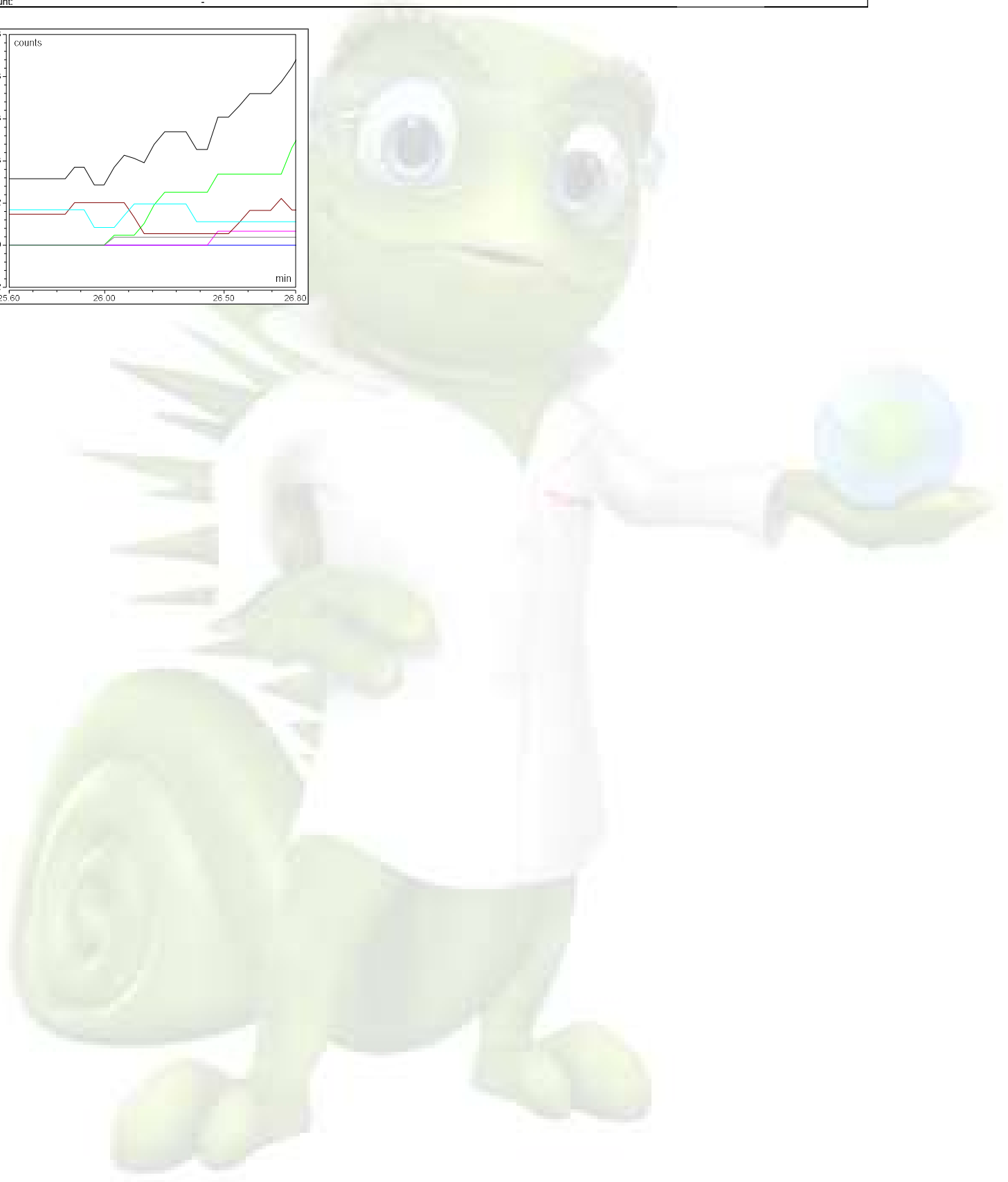
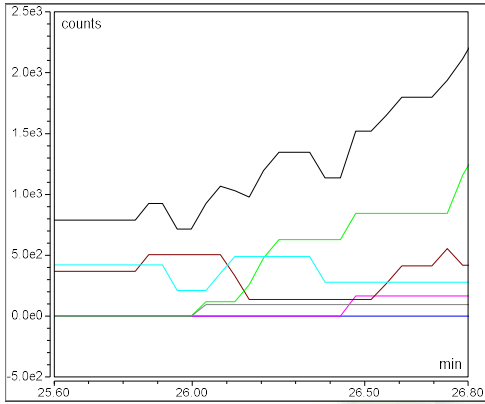
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

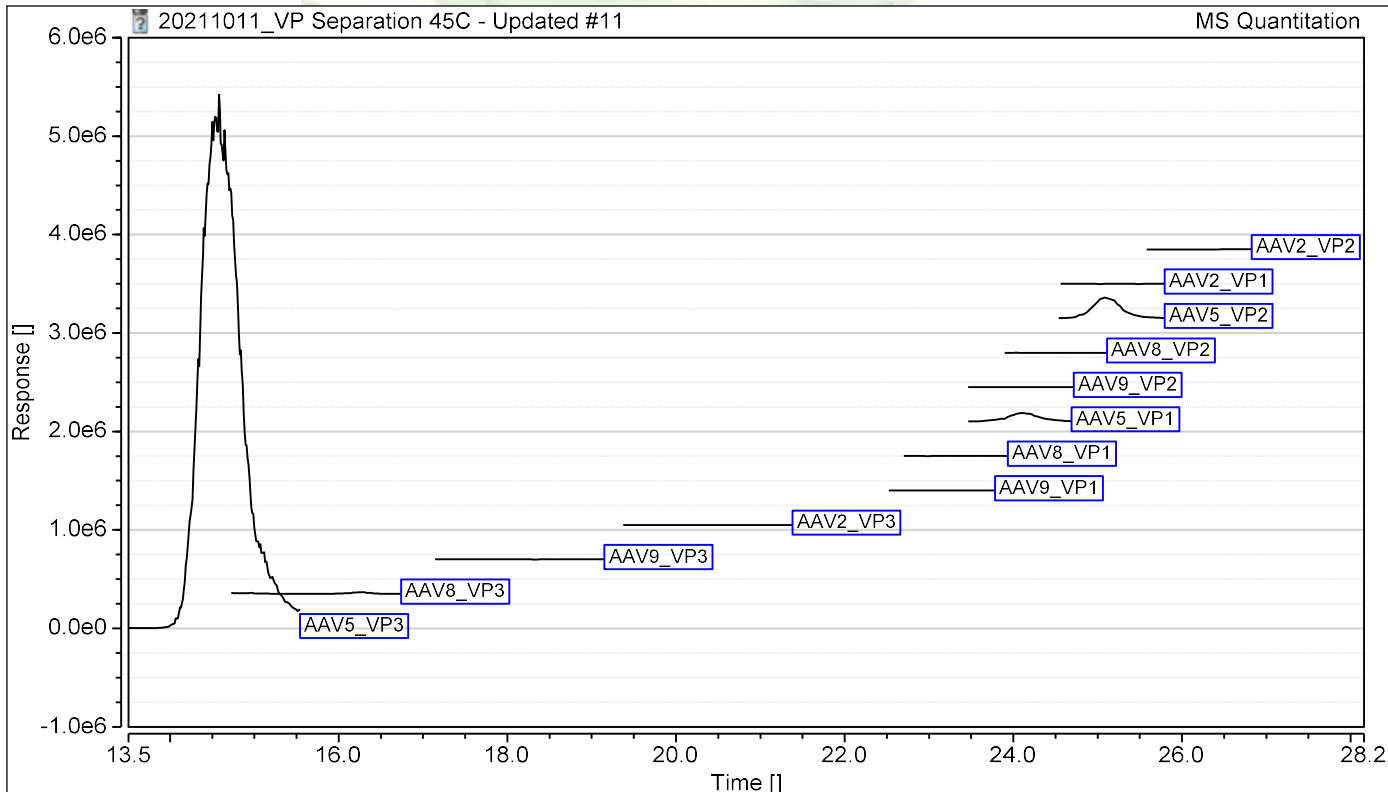
Amount: -



## Chromatogram and SST Results

| Injection Details           |   |                                       |
|-----------------------------|---|---------------------------------------|
| <i>Injection Name:</i>      | <b>20211011_AAV5 E_45C_65-59B_Mod1_1ug_2</b>  | <i>Run Time (min):</i> <b>44.98</b>   |
| <i>Vial Number:</i>         | <b>R:A7</b>   | <i>Injection Volume:</i> <b>3.17</b>  |
| <i>Injection Type:</i>      | <b>Unknown</b>  |                                       |
| <i>Calibration Level:</i>   |   |                                       |
| <i>Instrument Method:</i>   | <b>AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S</b> |                                       |
| <i>Processing Method:</i>   | <b>MS Quantitative_45C_65-59B</b>   | <i>Dilution Factor:</i> <b>1.0000</b> |
| <i>Injection Date/Time:</i> | <b>13/Oct/21 06:12</b>  | <i>Sample Weight:</i> <b>1.0000</b>   |

### Chromatogram

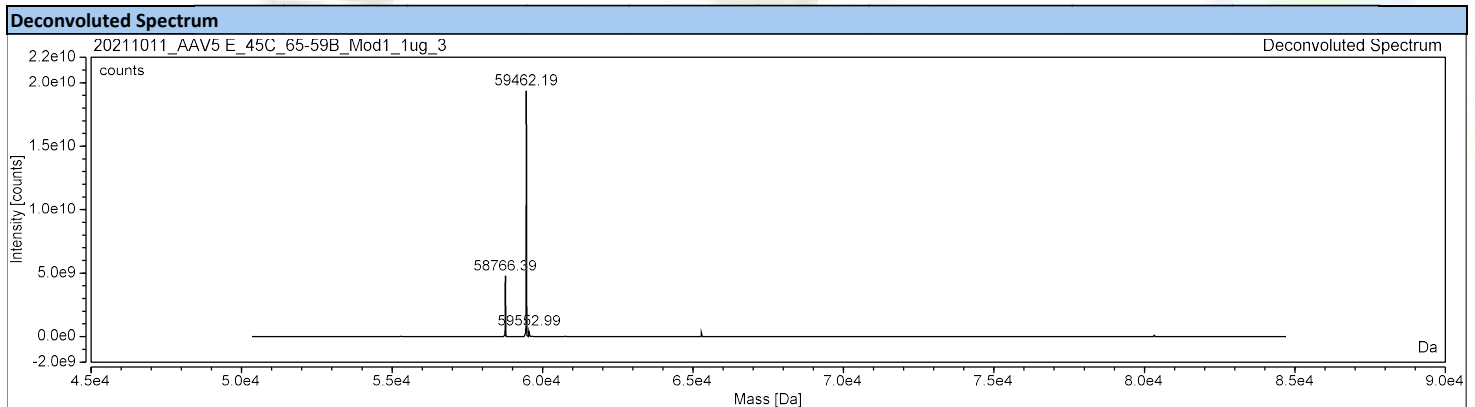
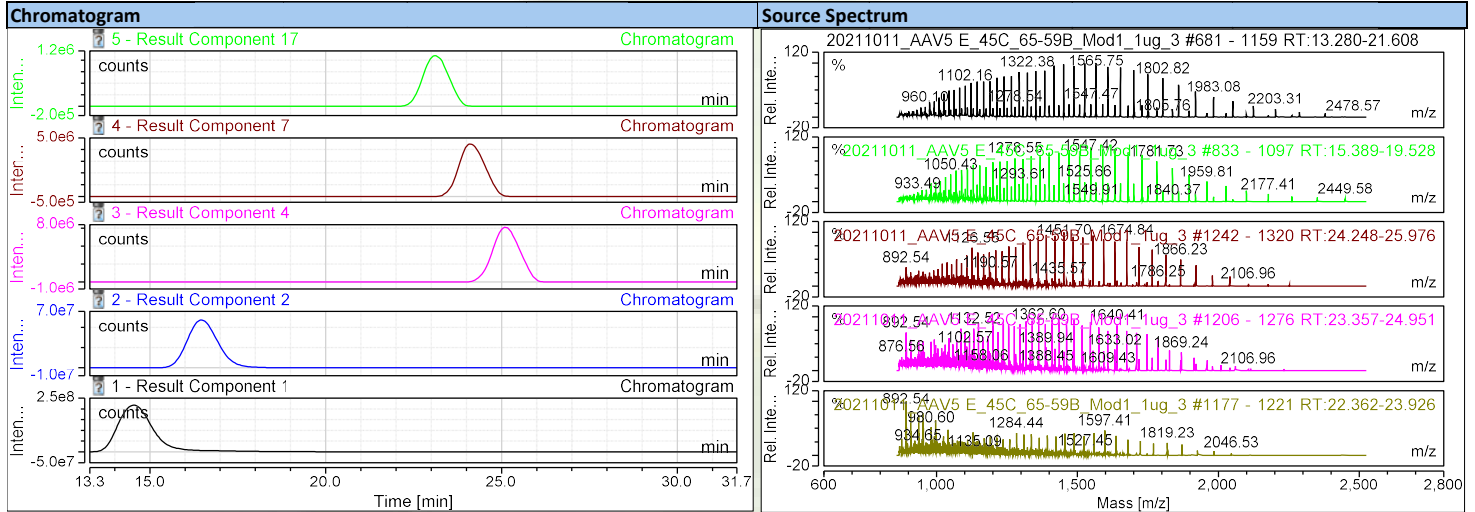


### SST Results

| No.  | Name | Inj. Condition | Peak | Test Result          | Injection     |
|--|------|----------------|------|----------------------|---------------|
| <b>Number of executed test cases: n.a.</b> |      |                |      | <b>Total Result:</b> | <b>Passed</b> |

## Protein Deconvolution Summary

| Injection Details |                                       |  |
|-------------------|---------------------------------------|--|
| Injection Name:   | 20211011_AAV5 E_45C_65-59B_Mod1_1ug_3 | Injection Volume: 3.17                                       |
| Vial Number:      | R:A7                                  | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                               | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 45.00                                 | Injection Date/Time: 13/Oct/21 06:58                         |



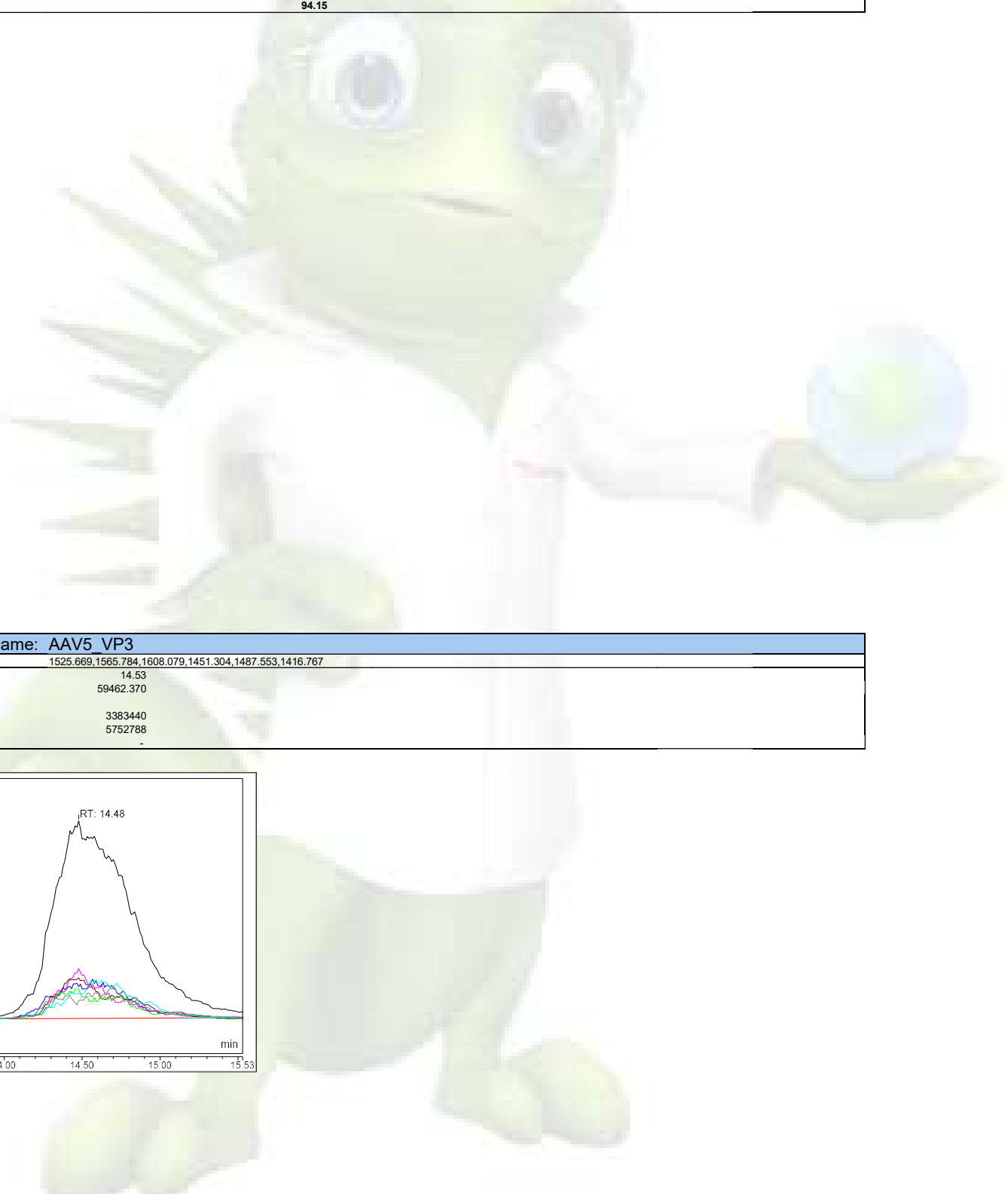
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

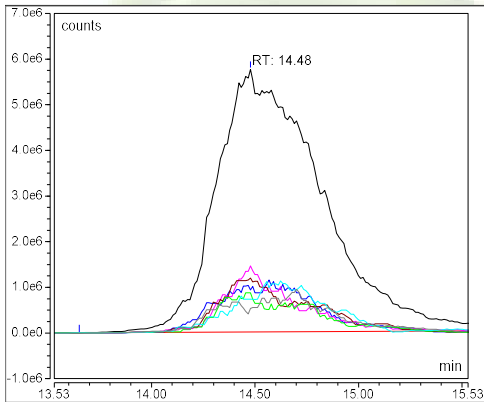
|  |   |  |
|--|---|--|
| <b>Injection Name:</b> 20211011_AAV5_E_45C_65-59B_Mod1_1ug | <b>Injection Volume:</b> 3.17   |  |
| <b>Vial Number:</b> R:A7                                   | <b>Instrument Method:</b> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <b>Injection Type:</b> Unknown                             | <b>Processing Method:</b> MS Quantitative_45C_65-59B                            |  |
| <b>Run Time (min):</b> 45.00                               | <b>Injection Date/Time:</b> 13/Oct/21 06:58                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV5_VP3     | 14.478                | 3383440.026        | 91.46              | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 | 59462.370 | PASS                           |
| AAV5_VP1     | 24.186                | 32188.637          | 0.87               | 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697 | 80335.080 | PASS                           |
| AAV5_VP2     | 25.076                | 67494.962          | 1.82               | 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727 | 65282.690 | PASS                           |
| <b>94.15</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <b>Quantitation ions:</b>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <b>Retention time (min):</b> | 14.53   |
| <b>Expected Mass:</b>        | 59462.370   |
| <b>Area (count*min):</b>     | 3383440   |
| <b>Intensity (counts):</b>   | 5752788   |
| <b>Amount:</b>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

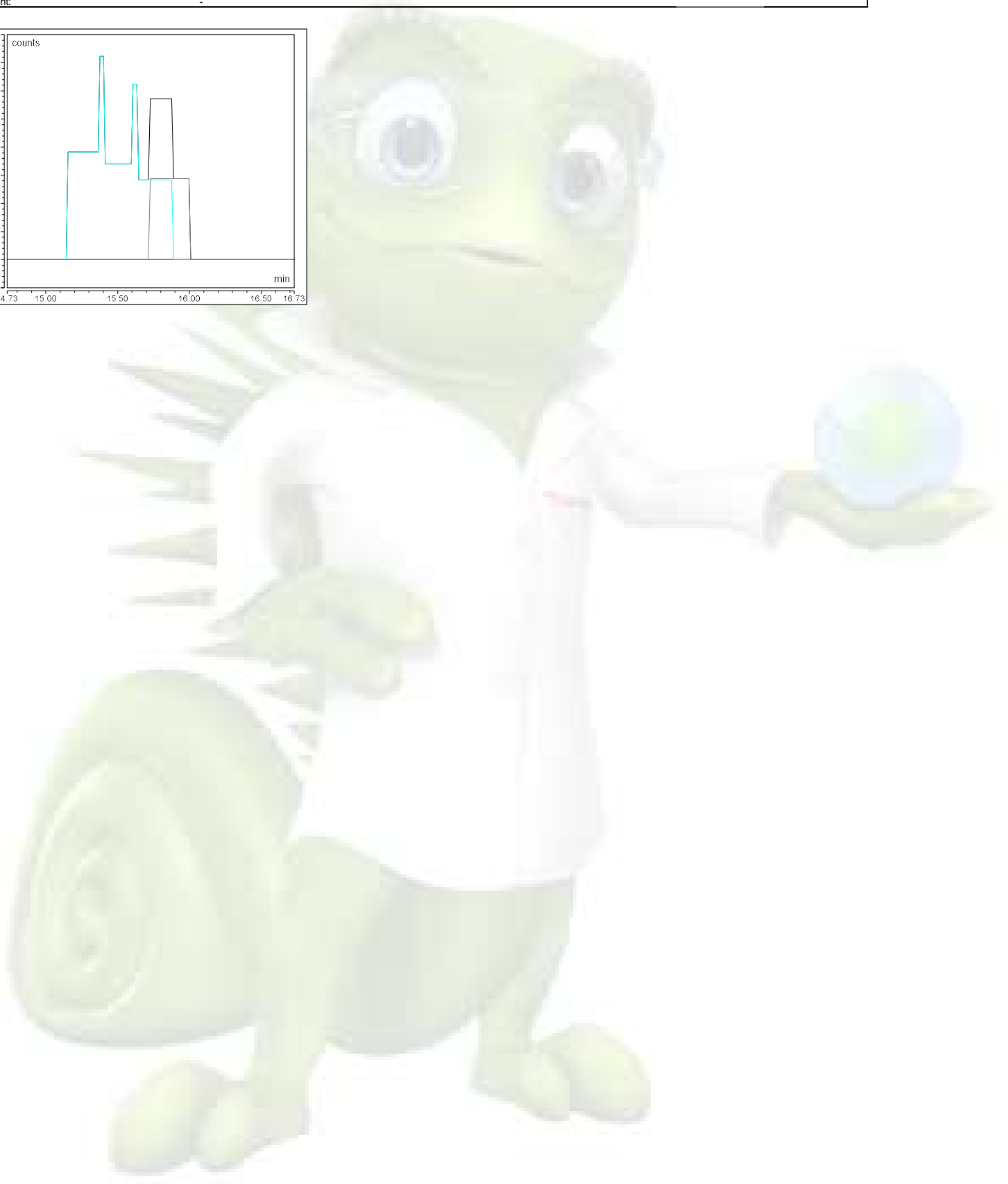
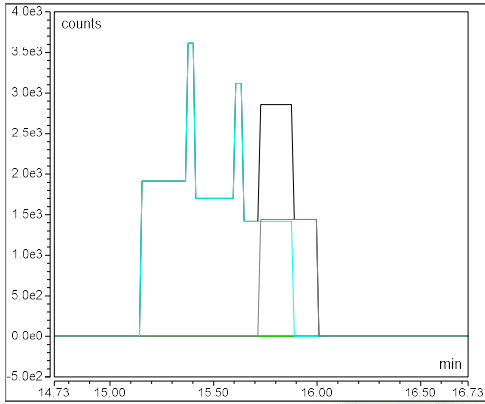
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

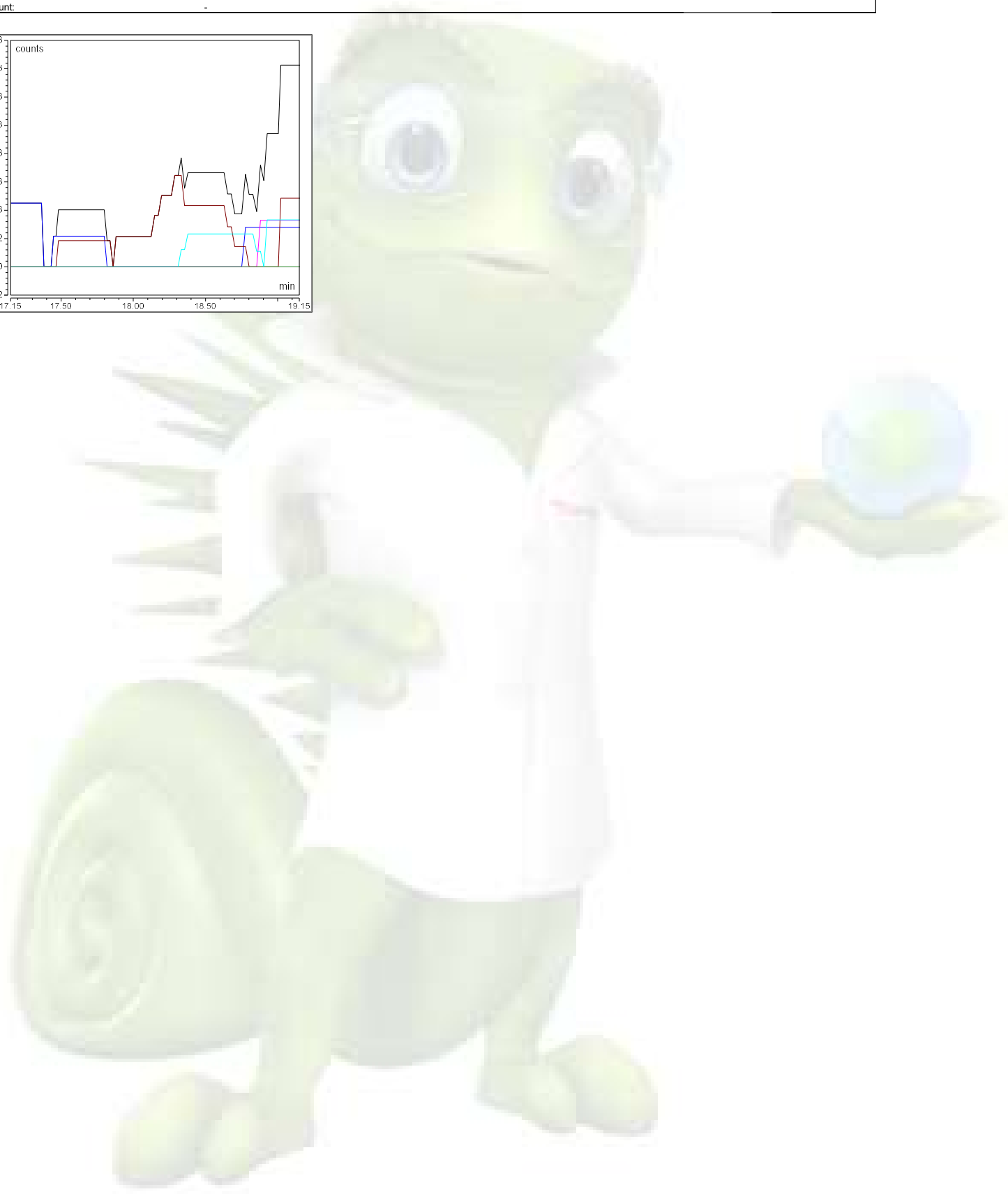
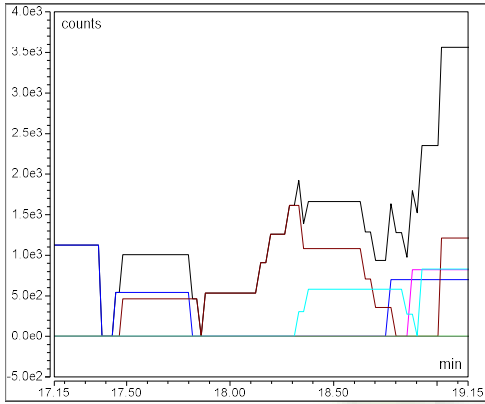
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

Quantitation ions: 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792

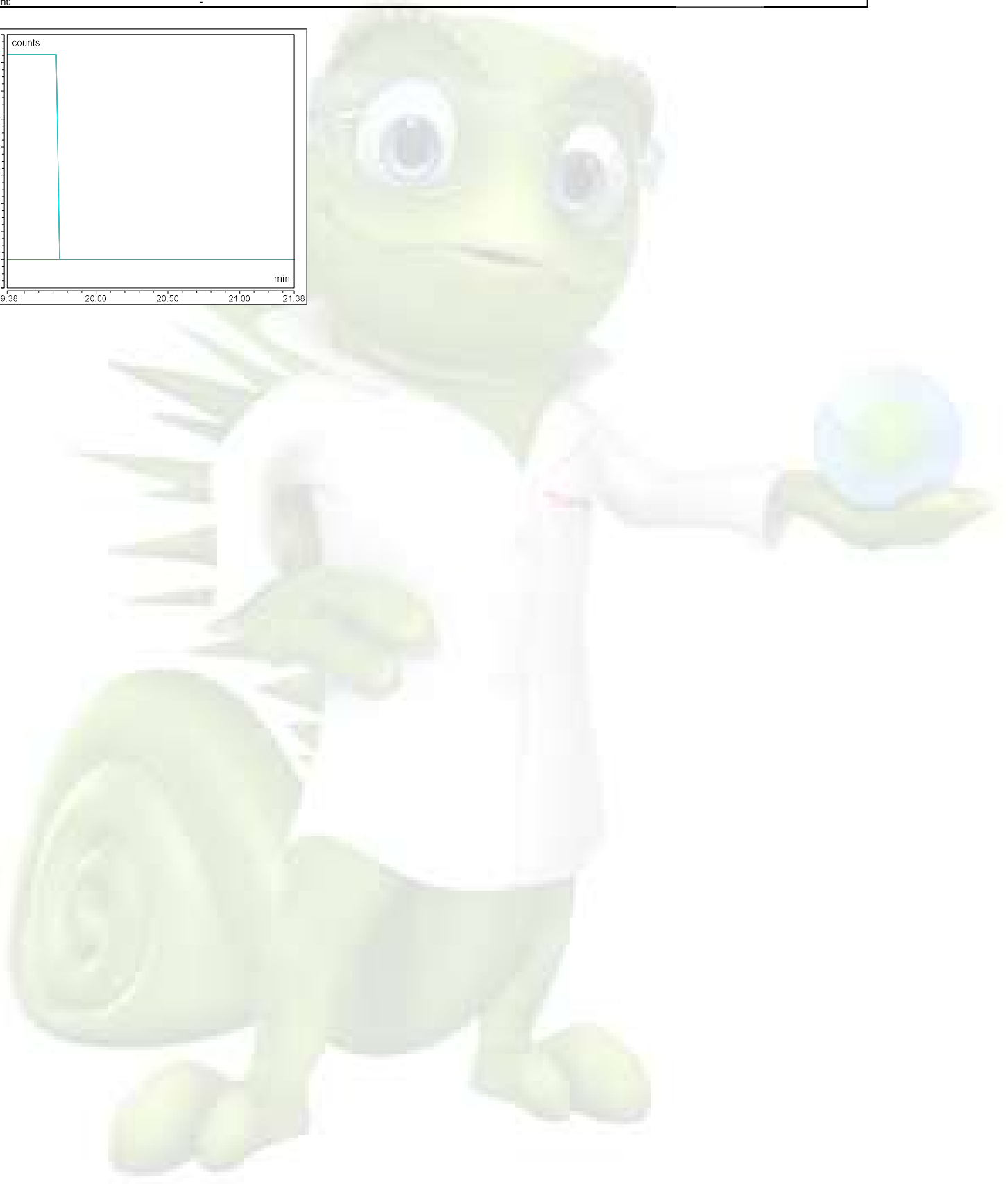
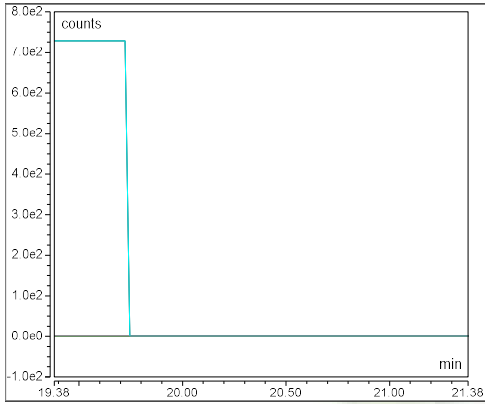
Retention time (min): 20.38

Expected Mass: 59973.780

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

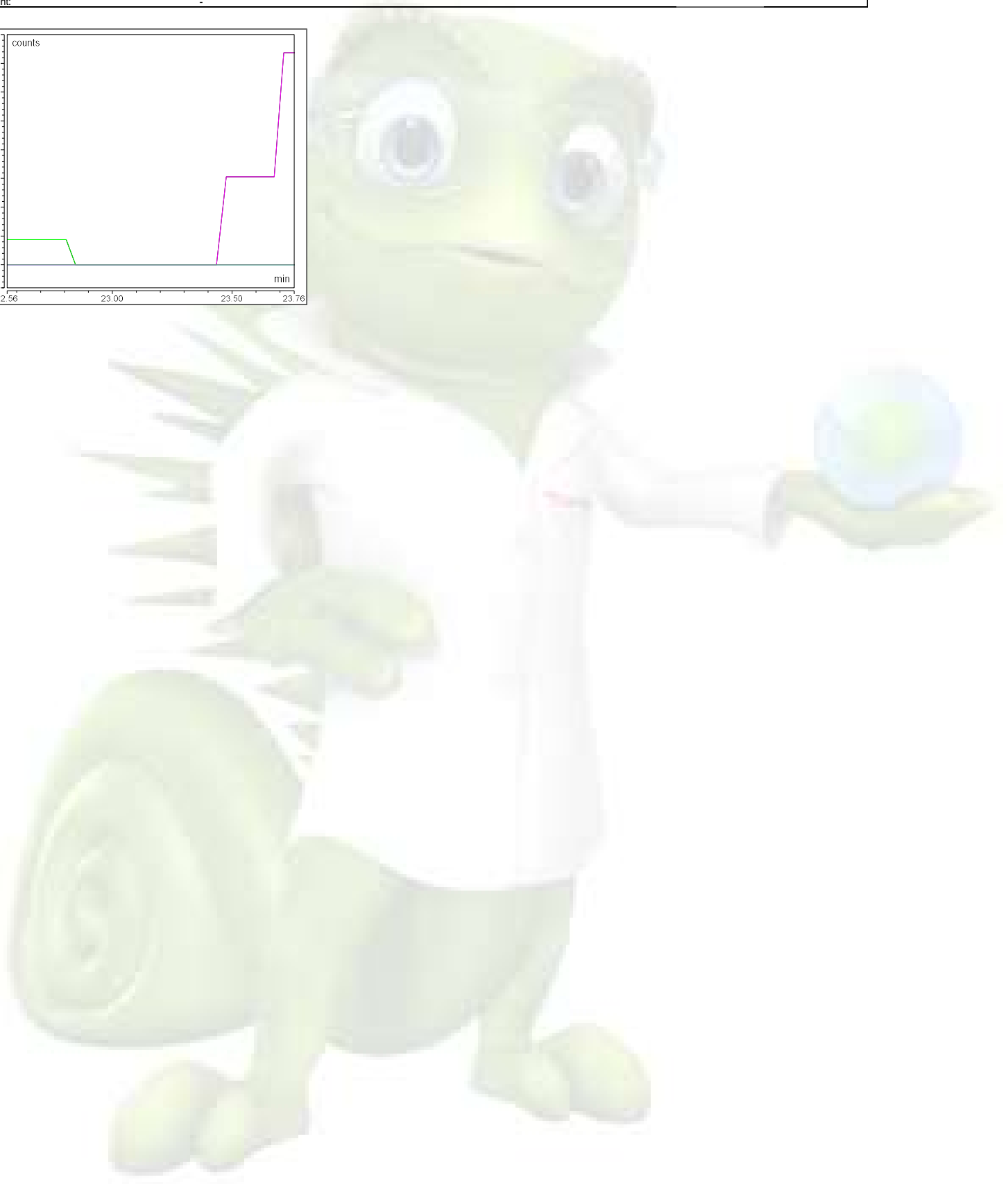
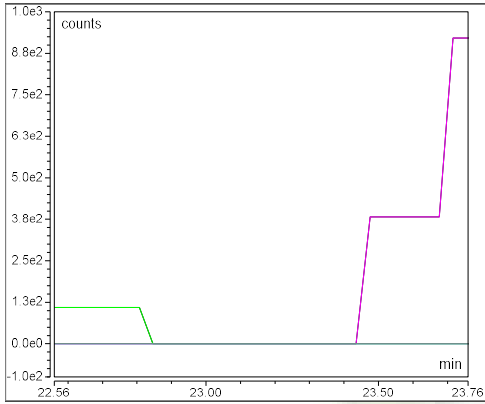
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

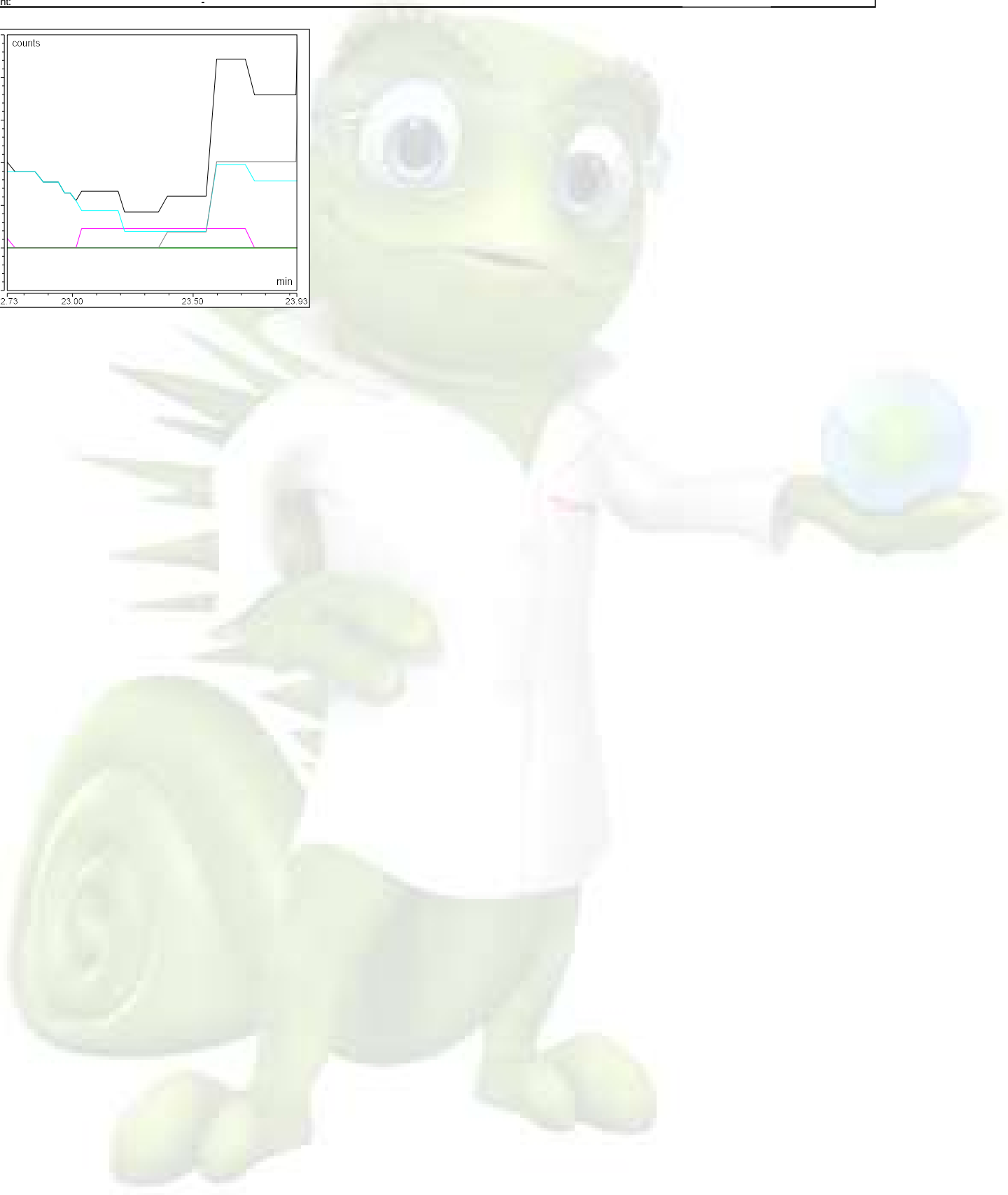
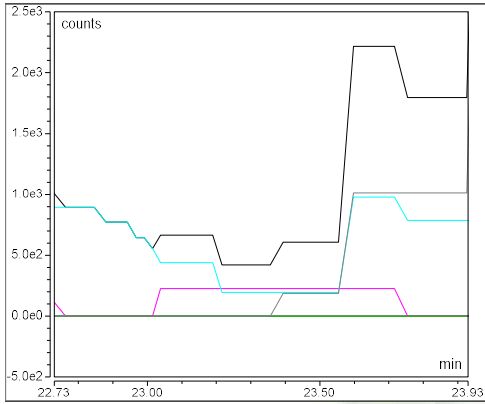
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

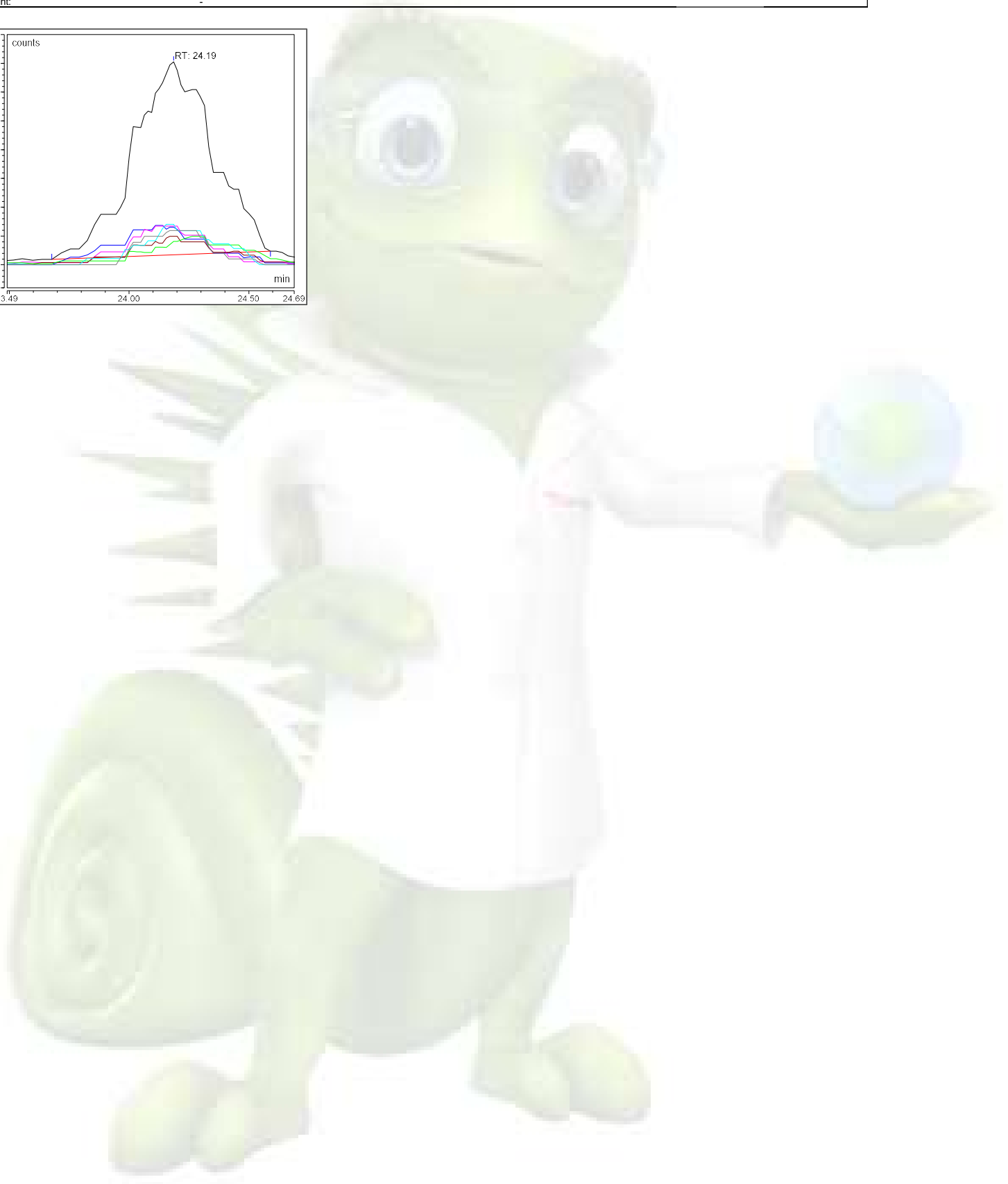
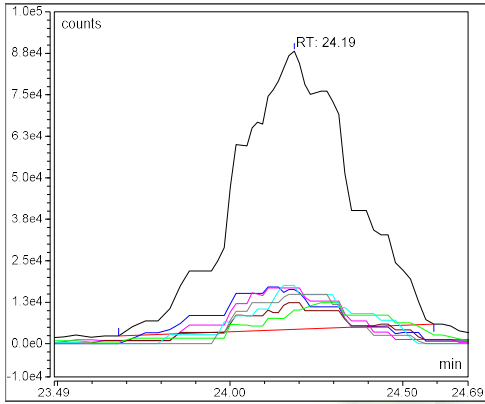
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): 32189

Intensity (counts): 83792

Amount: -



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

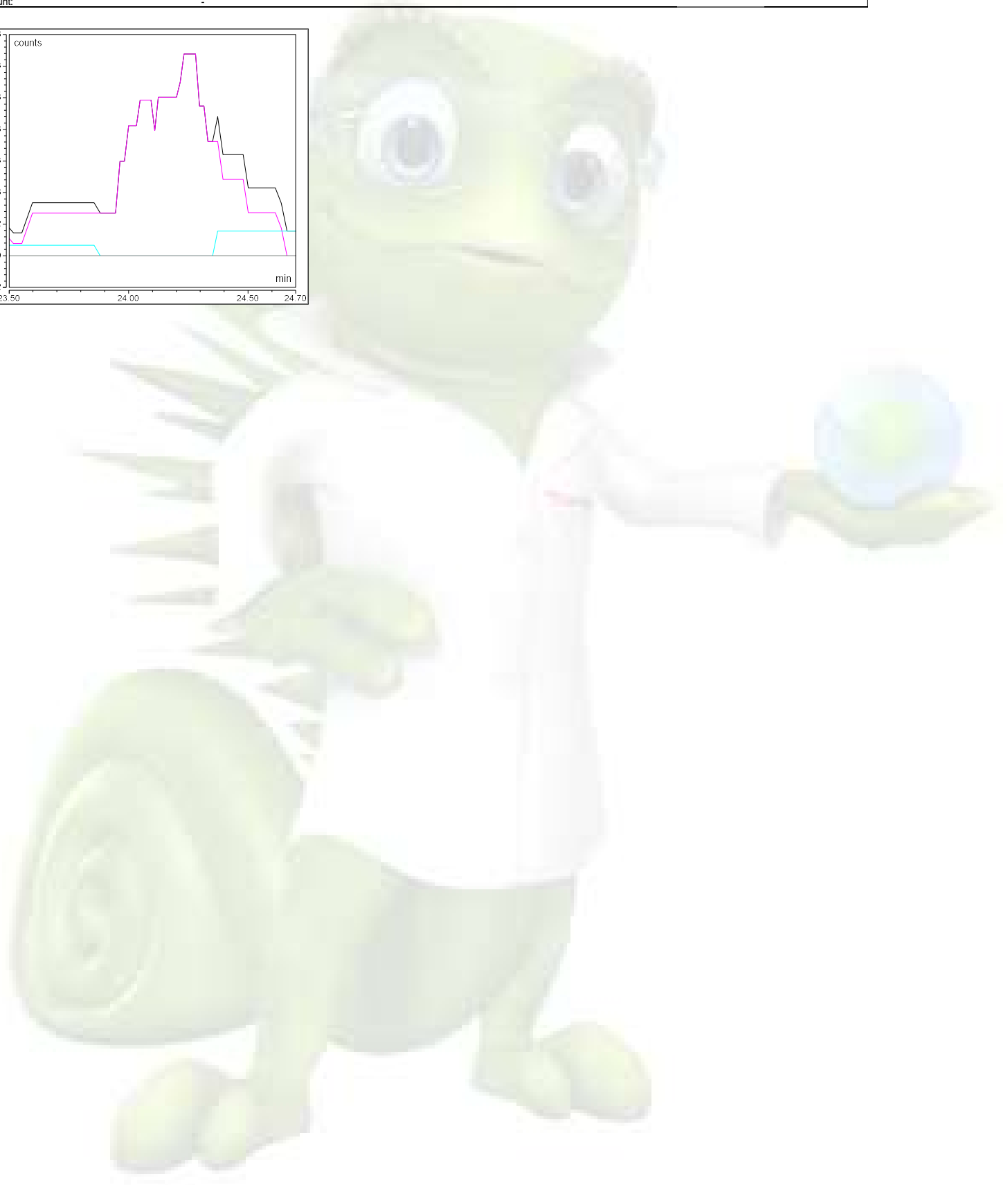
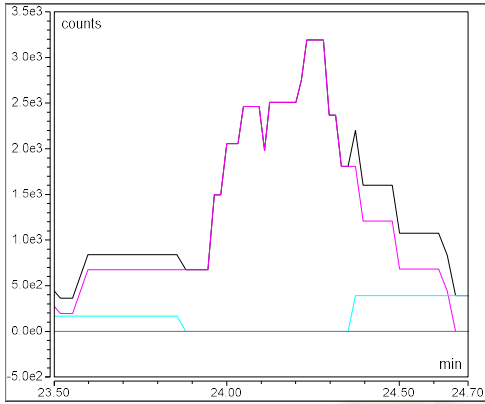
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

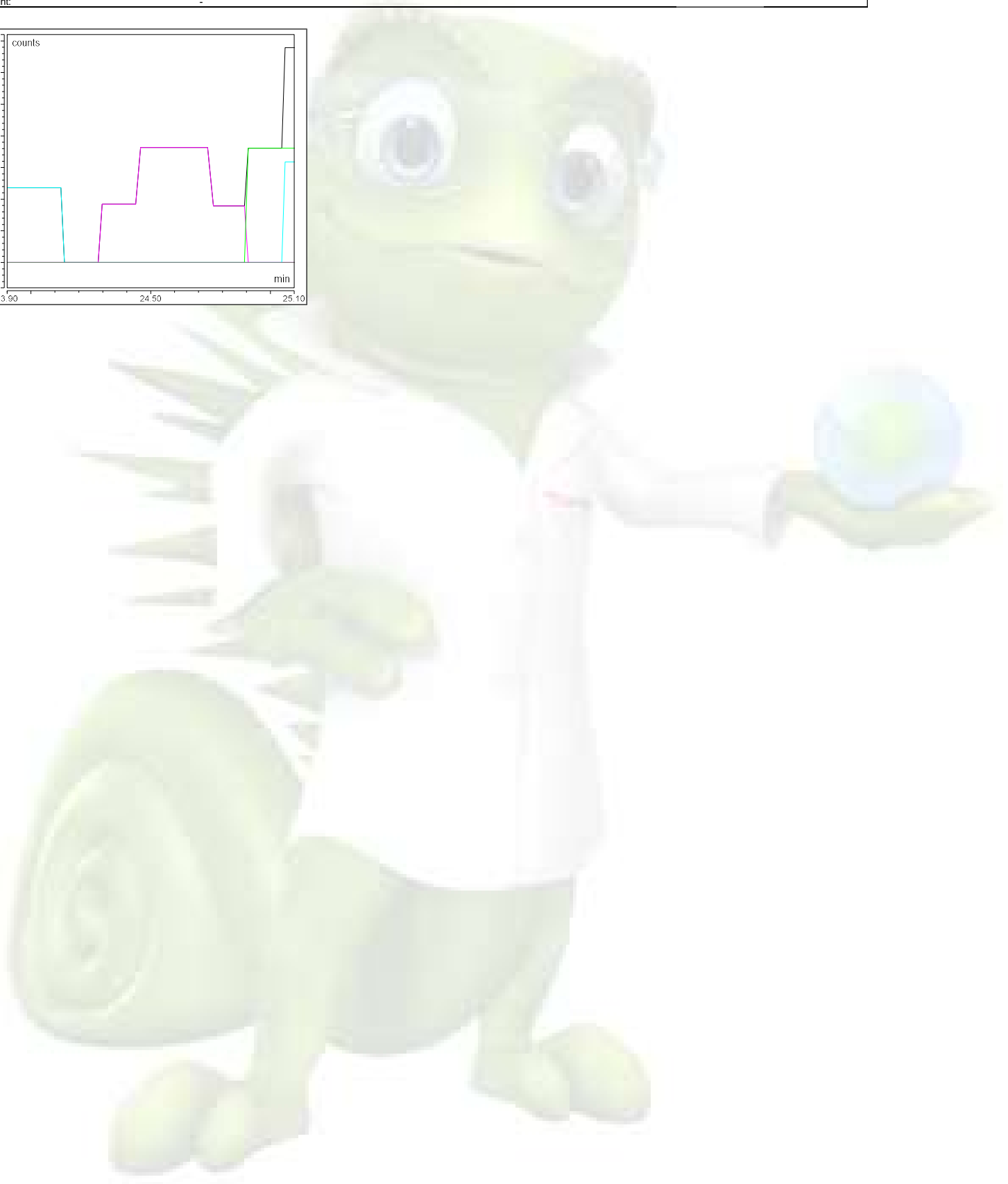
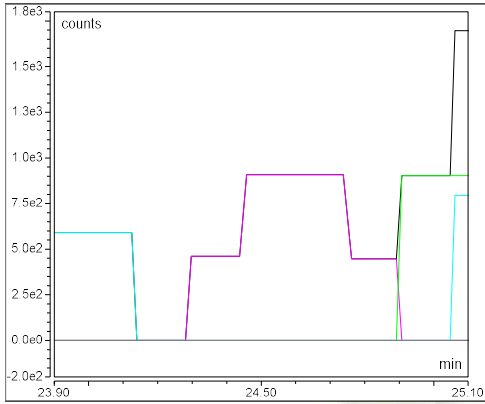
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

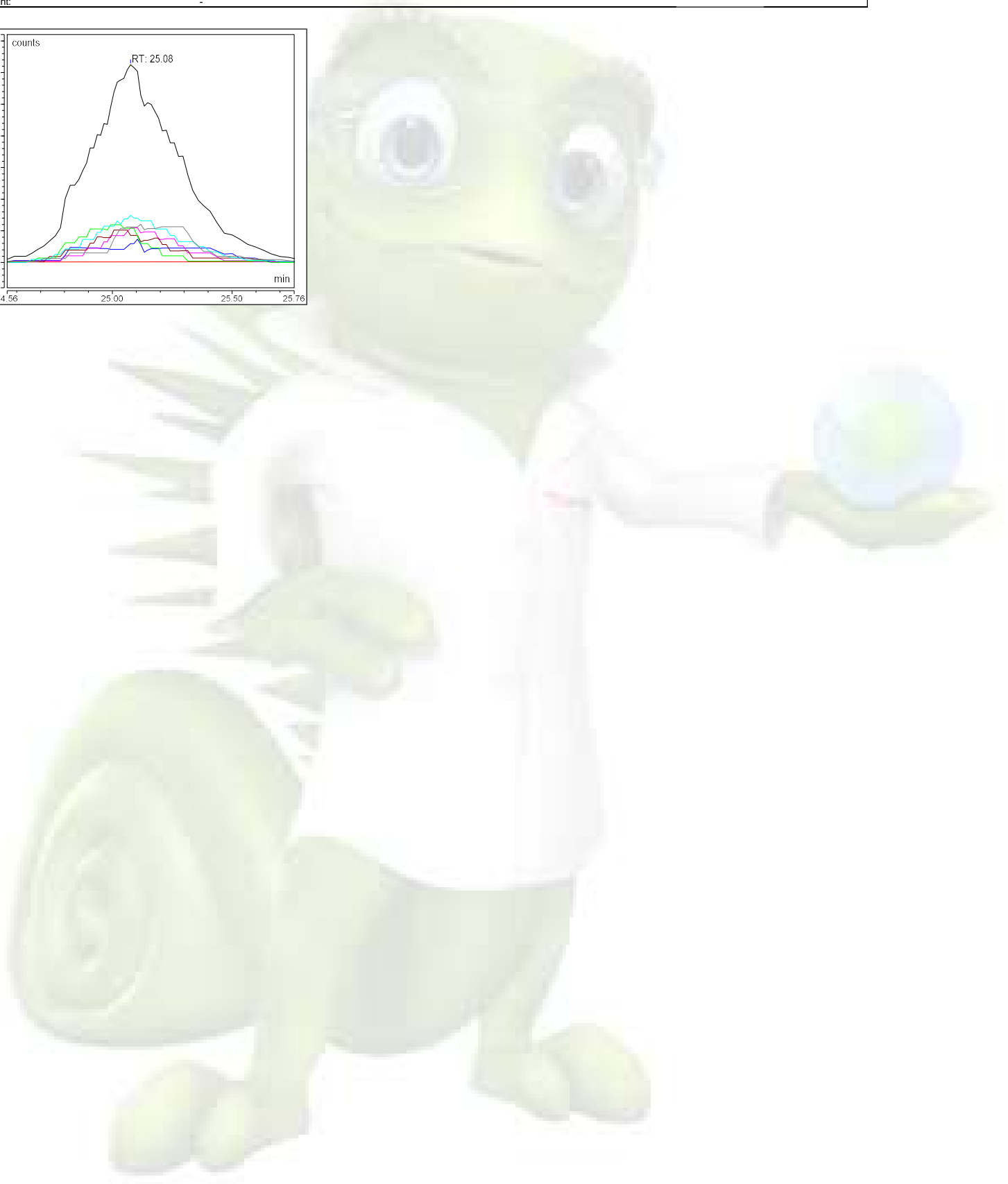
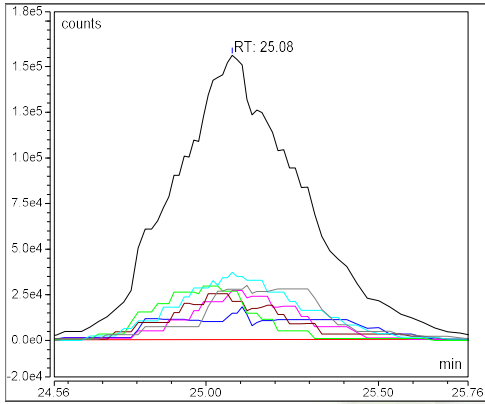
Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727 |
| Retention time (min): | 25.16   |
| Expected Mass:        | 65282.690   |
| Area (count*min):     | 67495   |
| Intensity (counts):   | 155649  |
| Amount:               | -   |



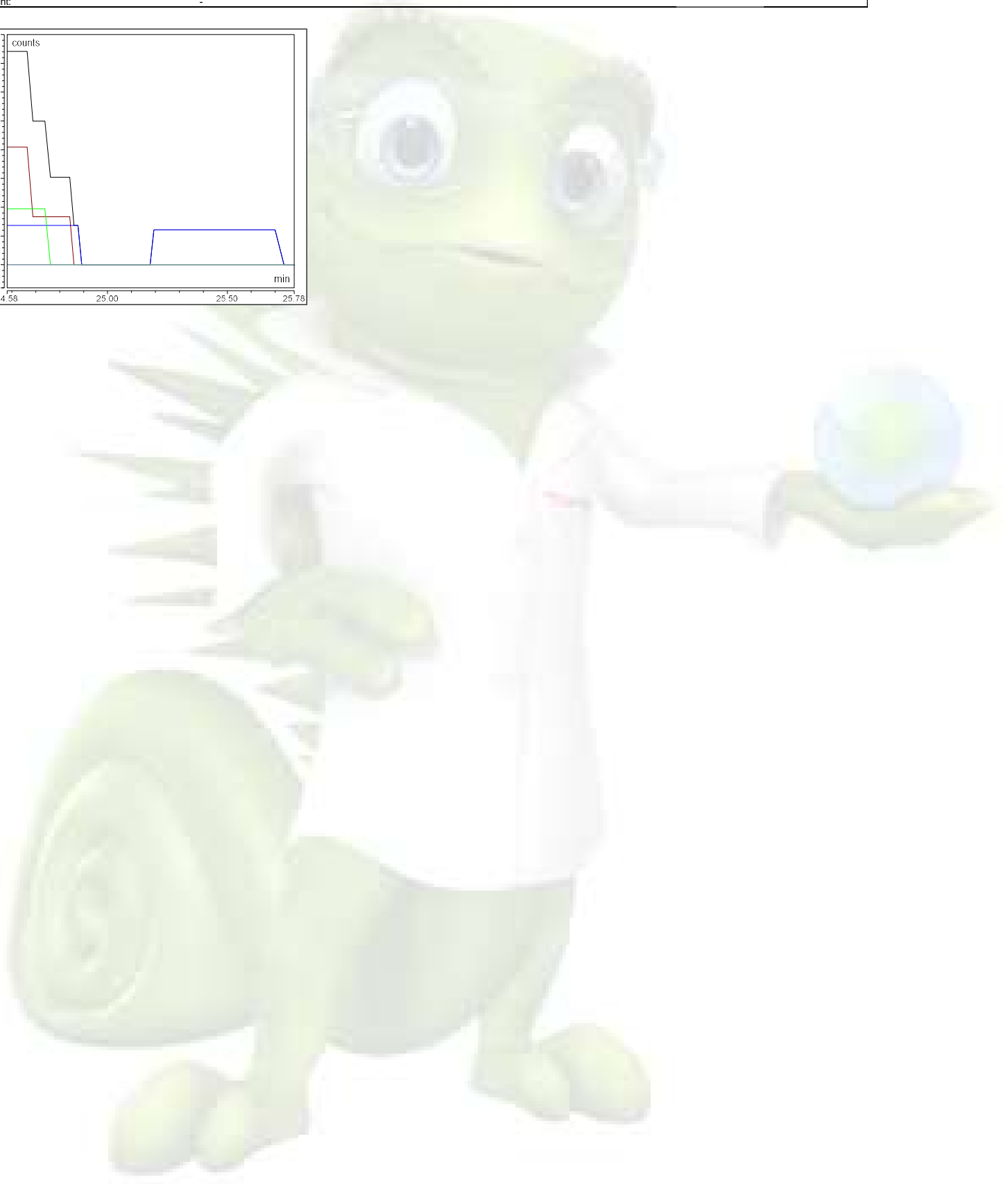
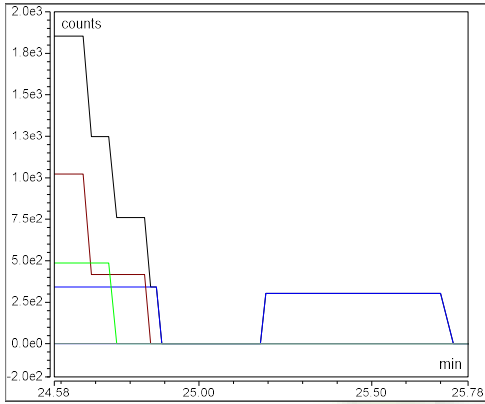
**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): -  
Intensity (counts): -  
Amount: -



**Component Name: AAV2 VP2**

Quantitation ions: 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686

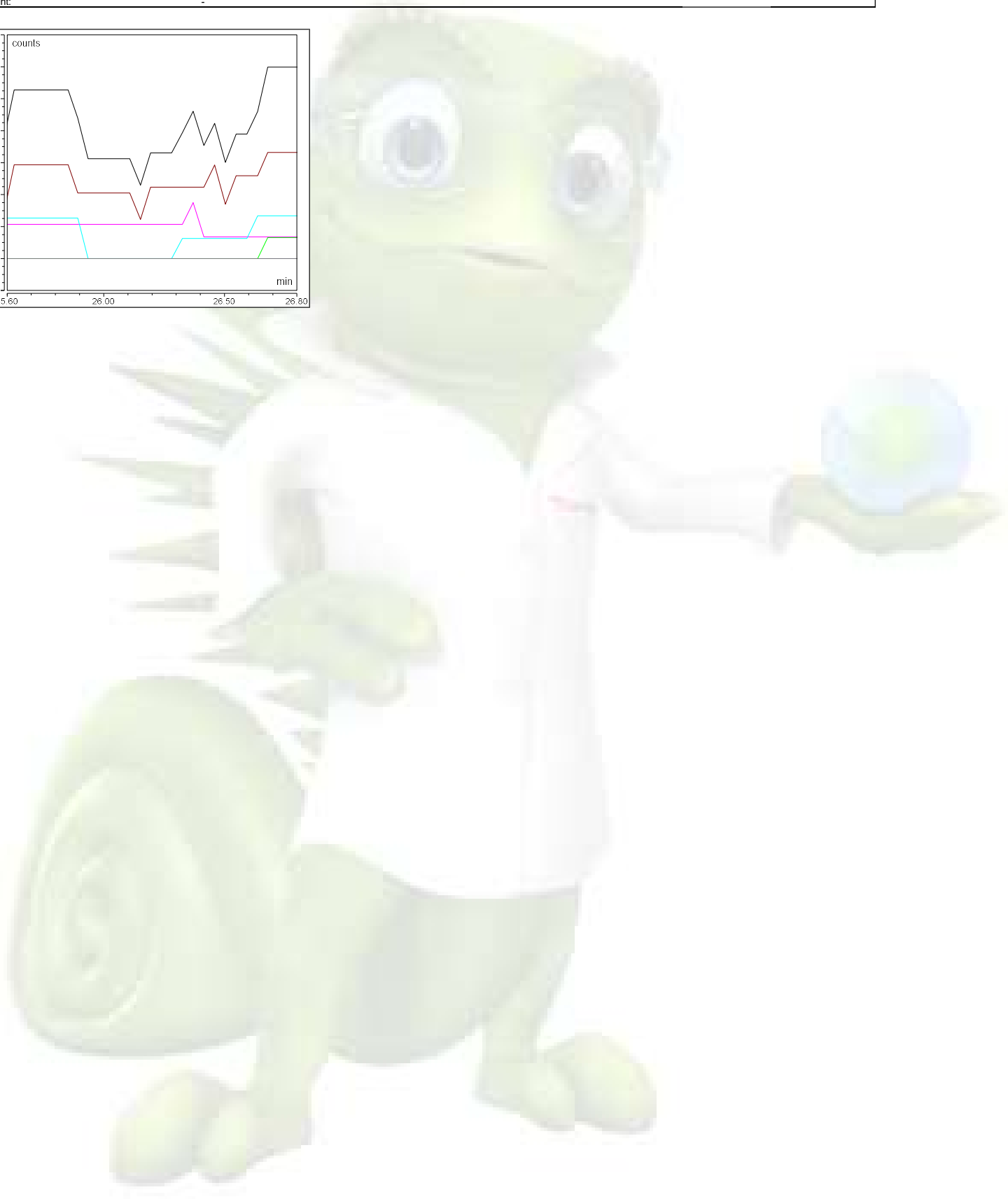
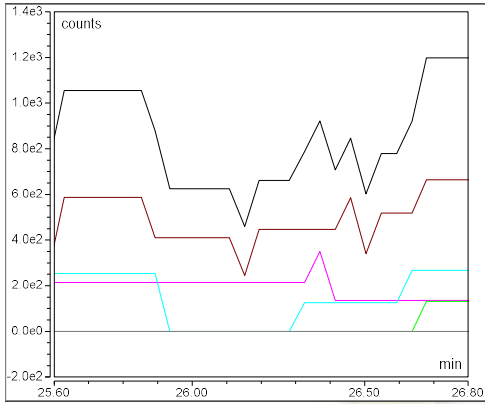
Retention time (min): 26.20

Expected Mass: 66487.900

Area (count\*min): -

Intensity (counts): -

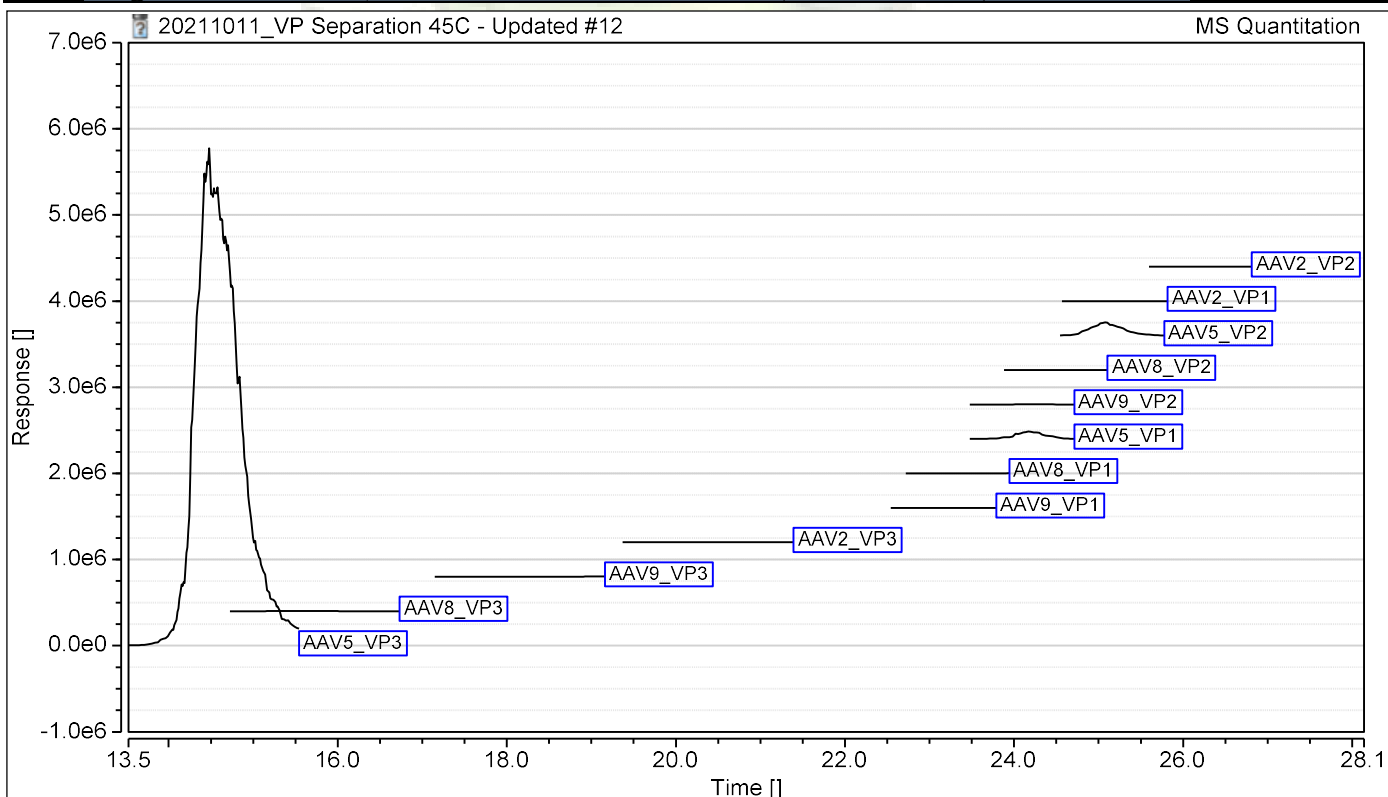
Amount: -



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV5 E_45C_65-59B_Mod1_1ug_3  | Run Time (min): 45.00   |
| Vial Number:         | R:A7   | Injection Volume: 3.17  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                         |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 13/Oct/21 06:58  | Sample Weight: 1.0000   |

### Chromatogram



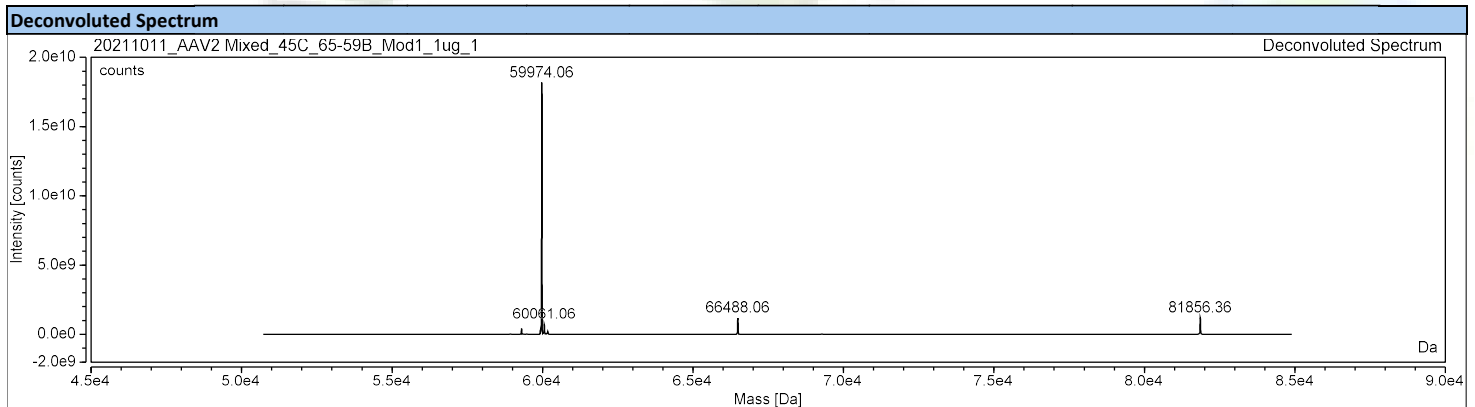
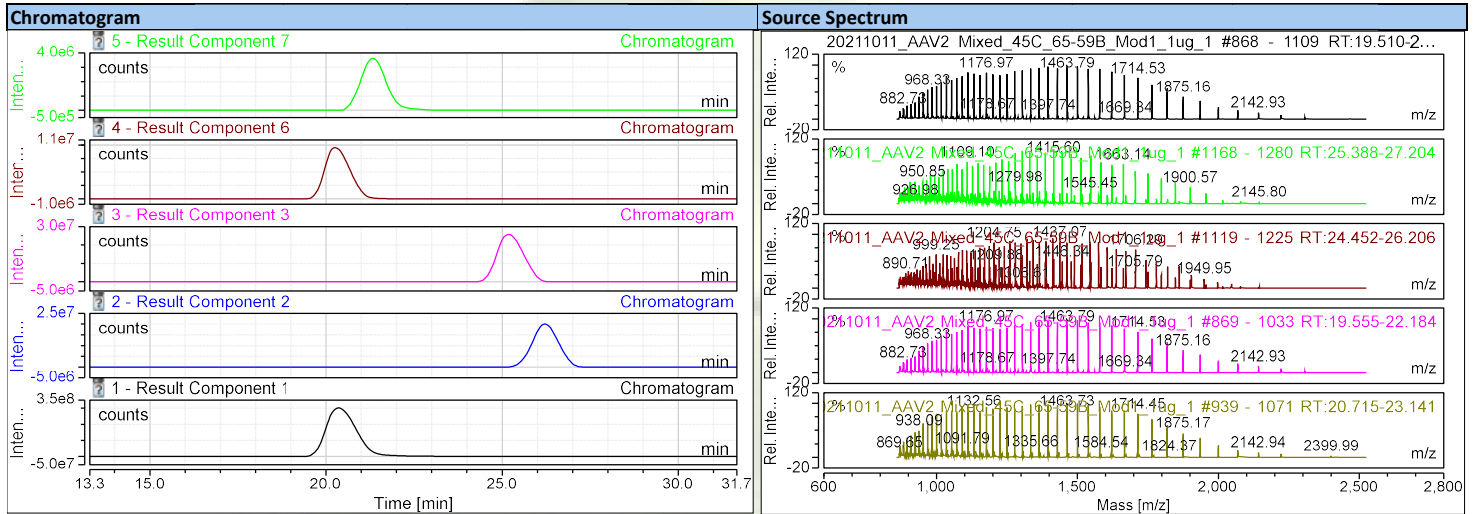
### SST Results

| No.  | Name | Inj. Condition | Peak | Test Result                 | Injection |
|--|------|----------------|------|-----------------------------|-----------|
| <b>Number of executed test cases: n.a.</b> |      |                |      | <b>Total Result: Passed</b> |           |



## Protein Deconvolution Summary

| Injection Details |   |   |
|-------------------|---|---|
| Injection Name:   | 20211011_AAV2 Mixed_45C_65-59B_Mod1_1ug_1 | Injection Volume:                         |
| Vial Number:      | R:A9                                      | Instrument Method:                        |
| Injection Type:   | Unknown                                   | Processing Method:                        |
| Run Time (min):   | 45.00                                     | Injection Date/Time:                      |
|                   |   | 8.45                                      |
|                   |   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
|                   |   | MS Quantitative_45C_65-59B                |
|                   |   | 12/Oct/21 18:13                           |



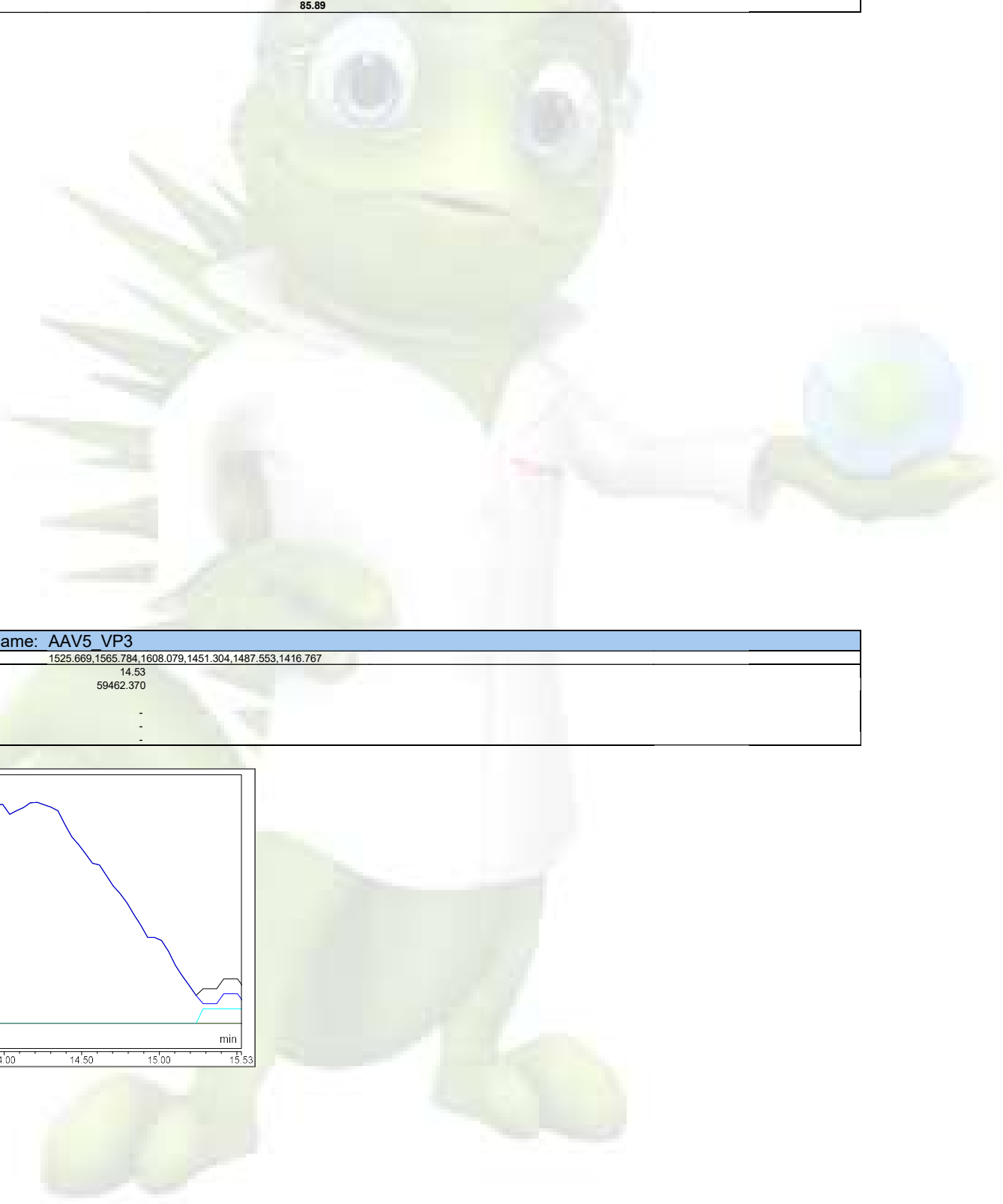
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

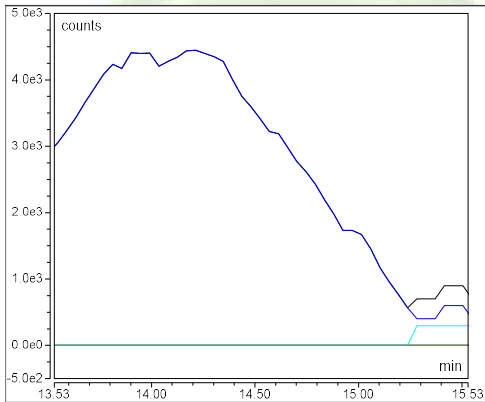
|  |   |  |
|--|---|--|
| <i>Injection Name:</i> 20211011_AAV2 Mixed_45C_65-59B_Mod1 | <i>Injection Volume:</i> 8.45   |  |
| <i>Vial Number:</i> R:A9                                   | <i>Instrument Method:</i> AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| <i>Injection Type:</i> Unknown                             | <i>Processing Method:</i> MS Quantitative_45C_65-59B                            |  |
| <i>Run Time (min):</i> 45.00                               | <i>Injection Date/Time:</i> 12/Oct/21 18:13                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3     | 20.399                | 3830196.738        | 79.17              | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 | 59973.780 | PASS                           |
| AAV2_VP1     | 25.230                | 145835.140         | 3.01               | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 | 81854.900 | PASS                           |
| AAV2_VP2     | 26.206                | 179370.301         | 3.71               | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 | 66487.900 | PASS                           |
| <b>85.89</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

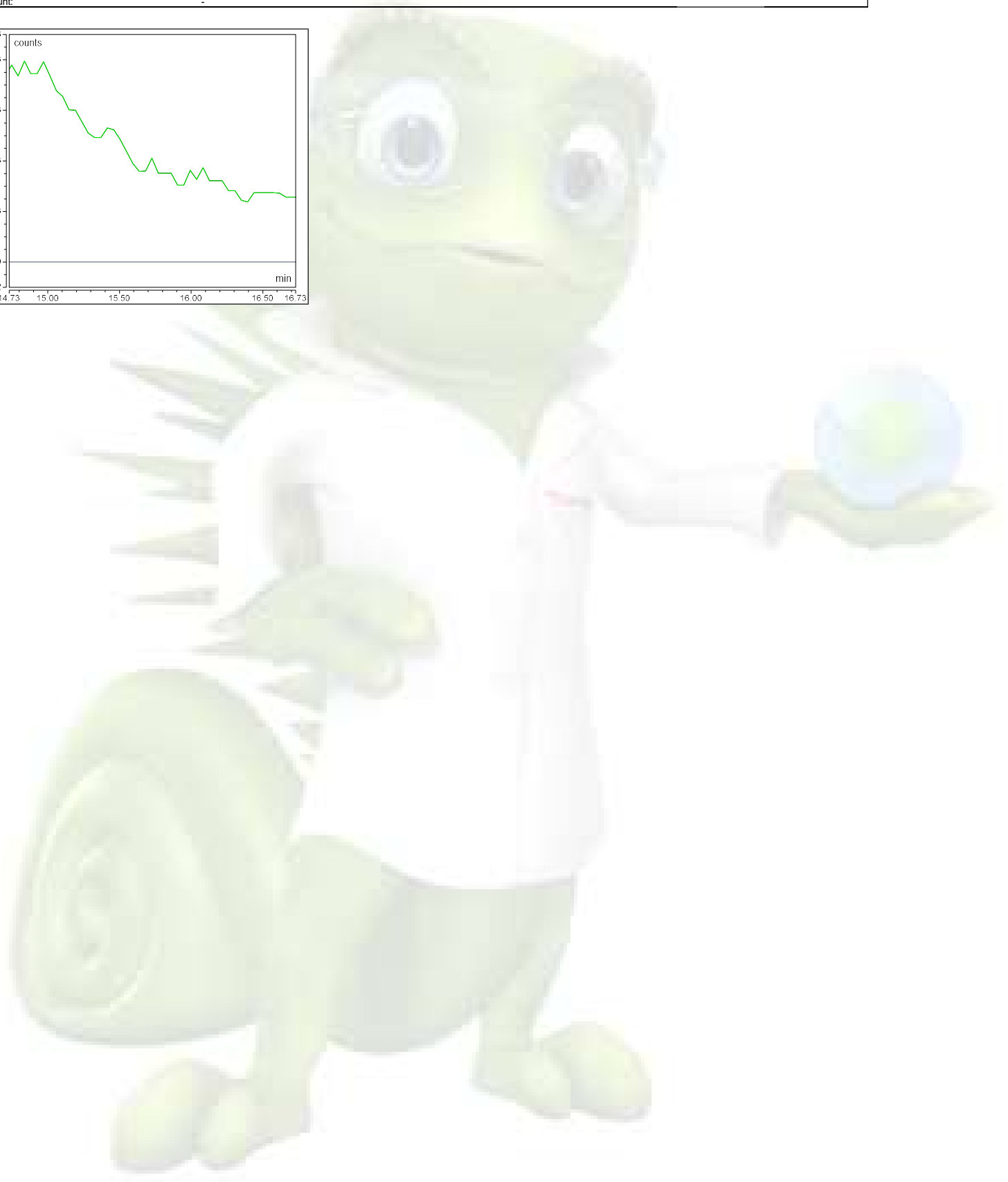
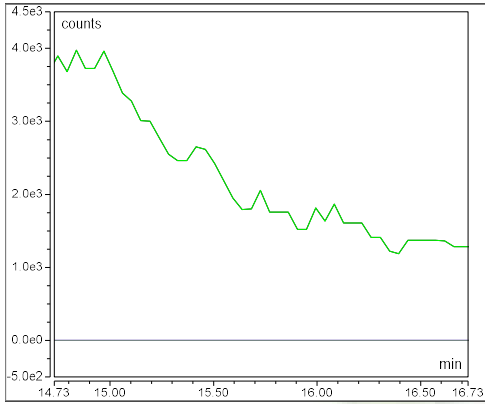
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

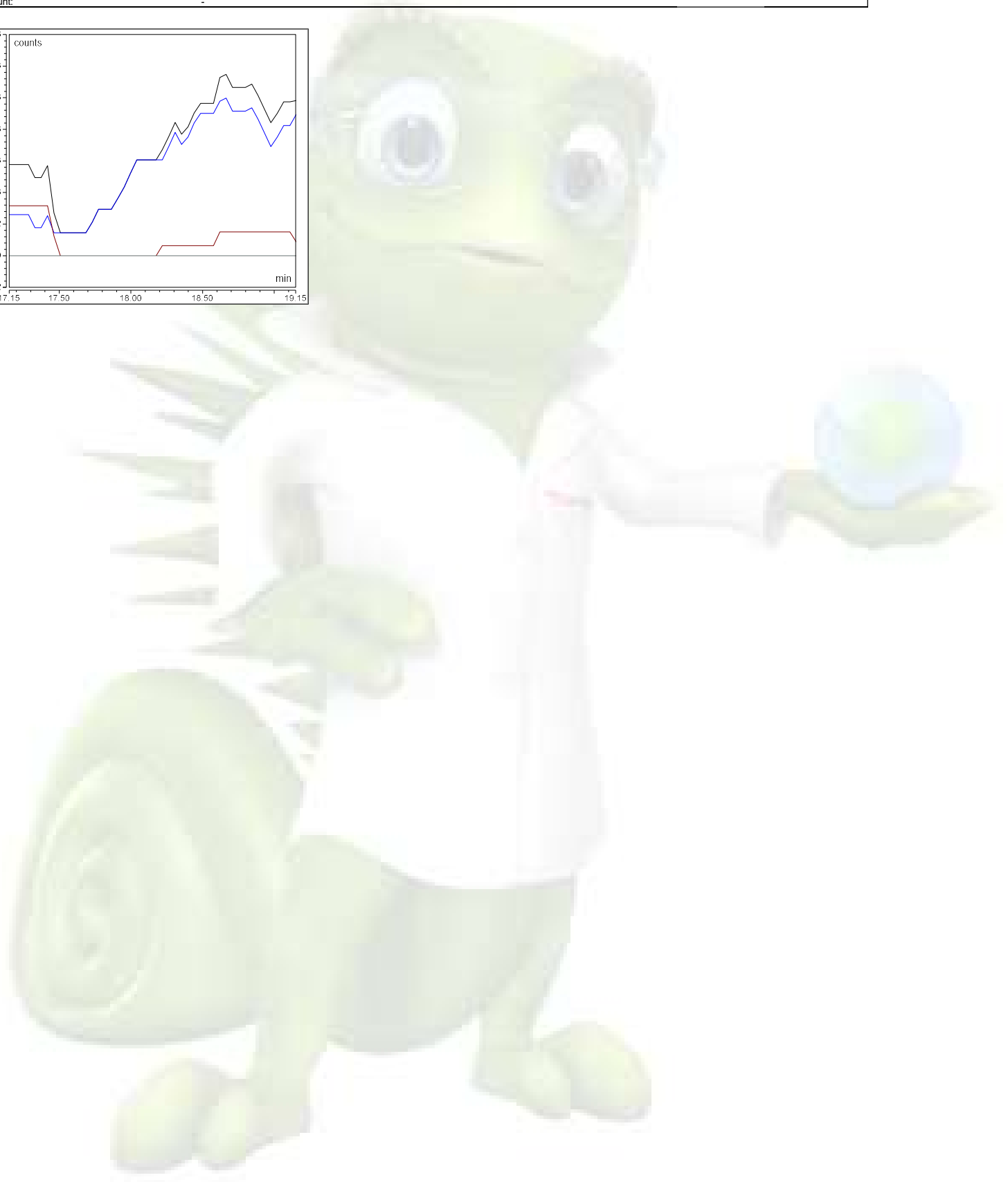
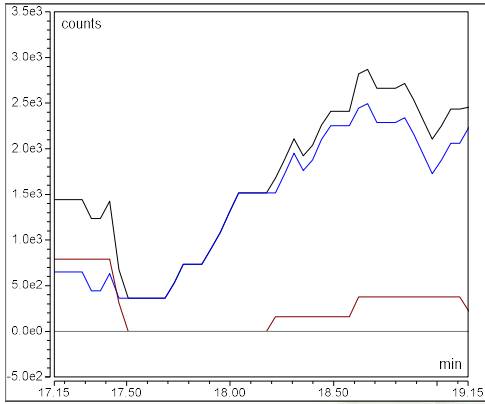
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

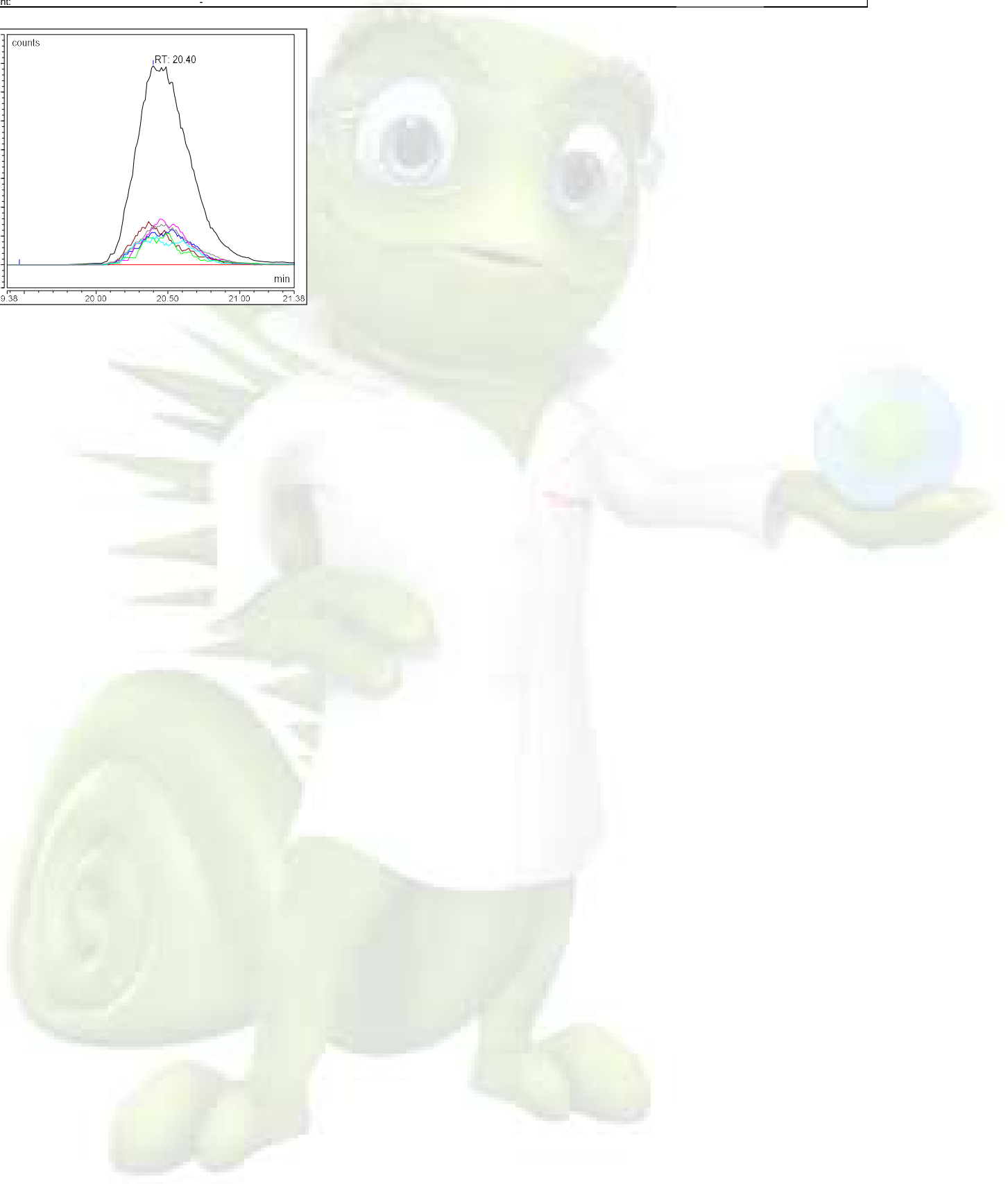
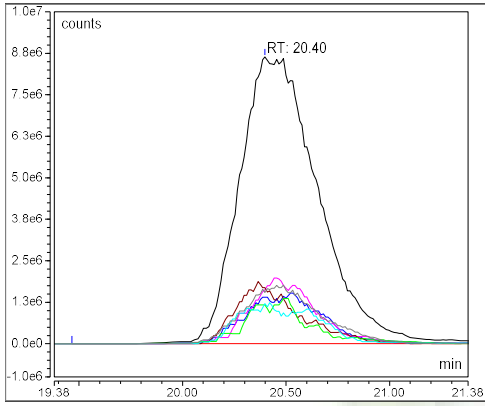
Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 |
| Retention time (min): | 20.38   |
| Expected Mass:        | 59973.780   |
| Area (count*min):     | 3830197   |
| Intensity (counts):   | 8641216   |
| Amount:               | -   |



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

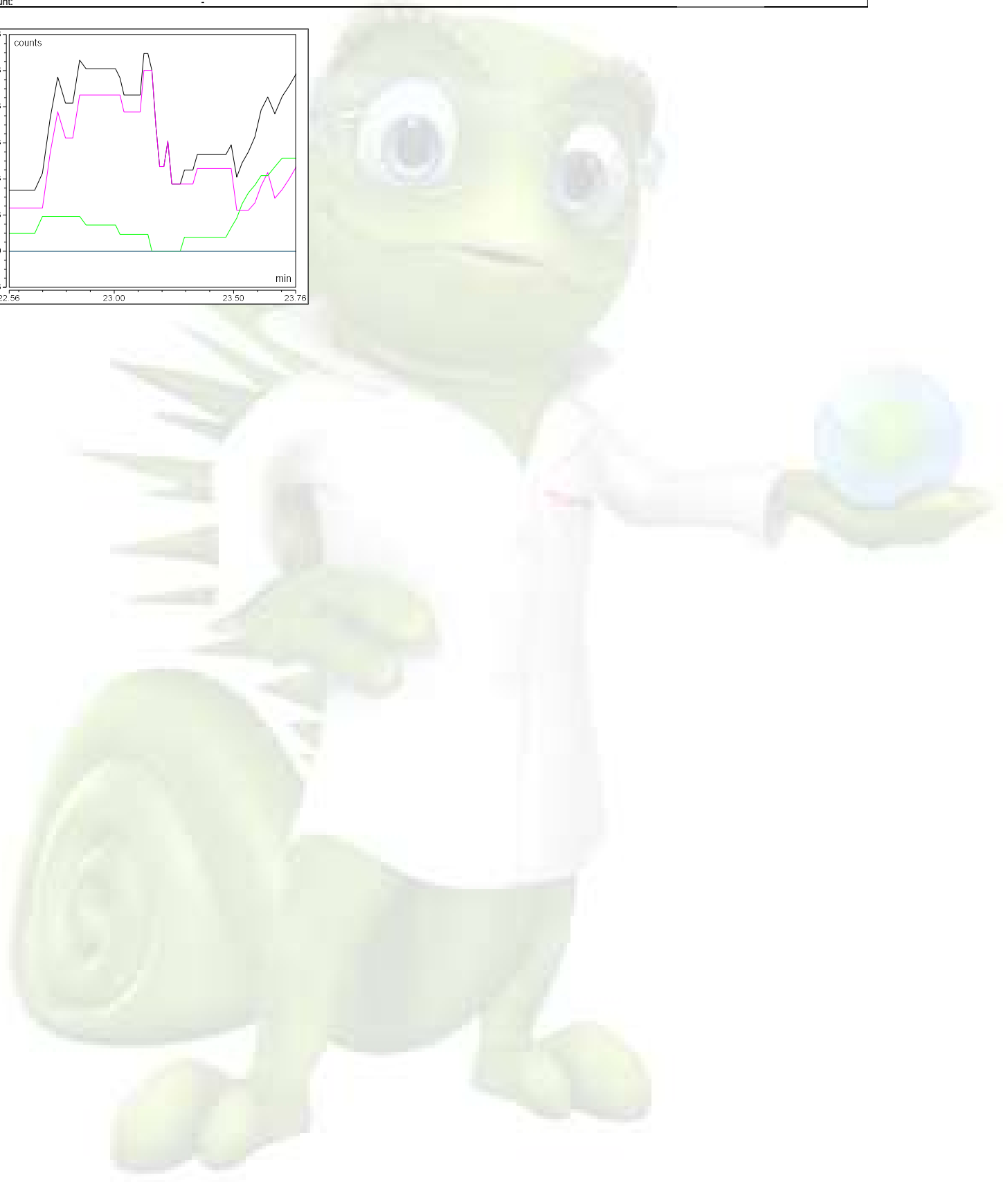
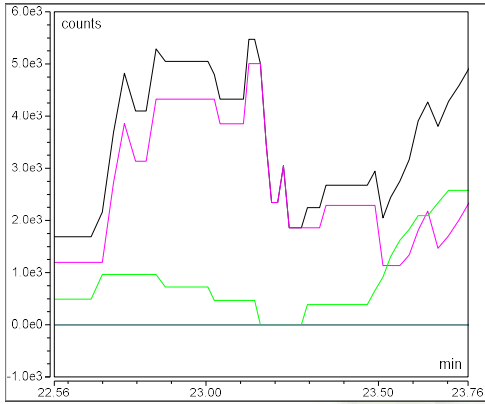
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

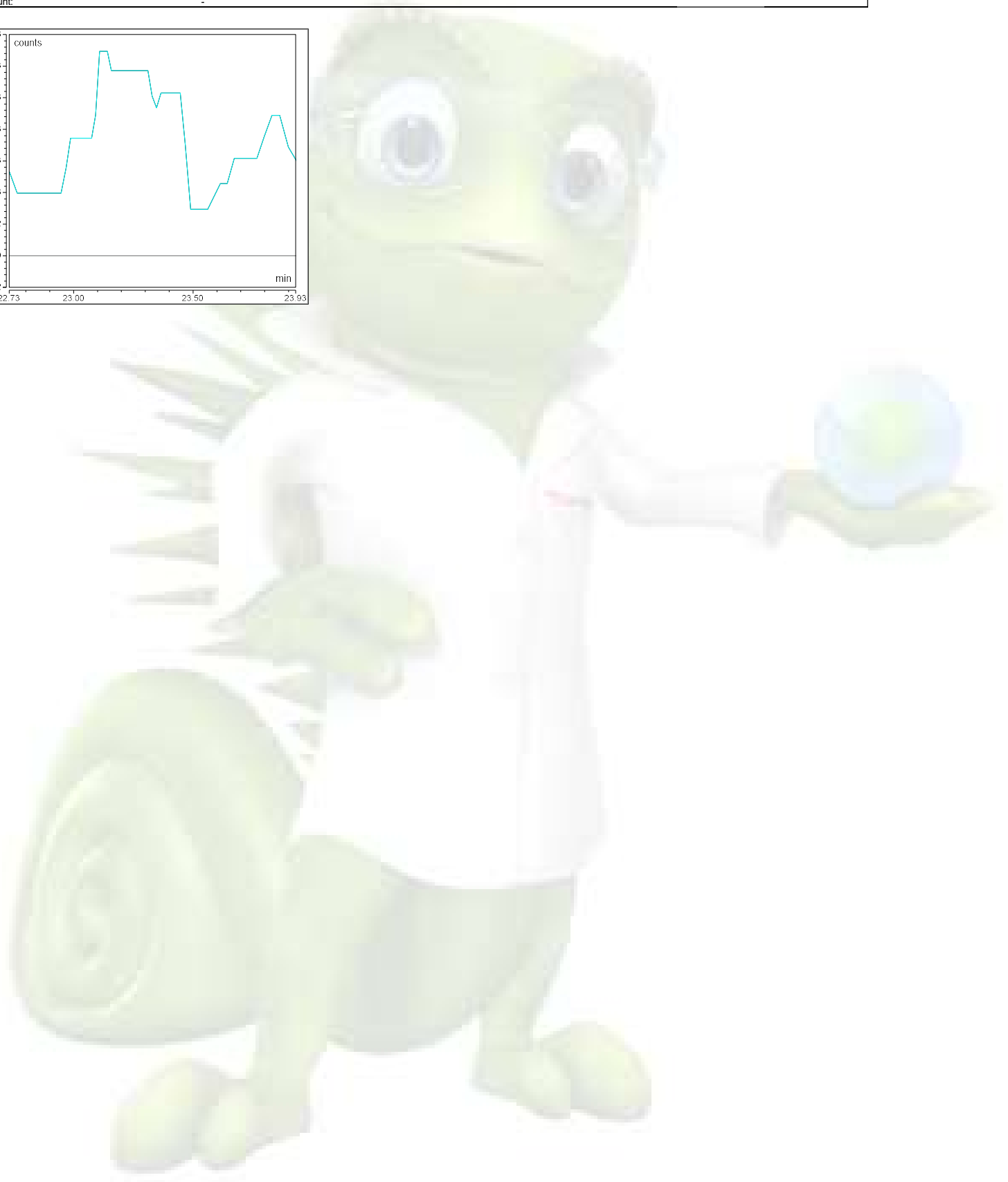
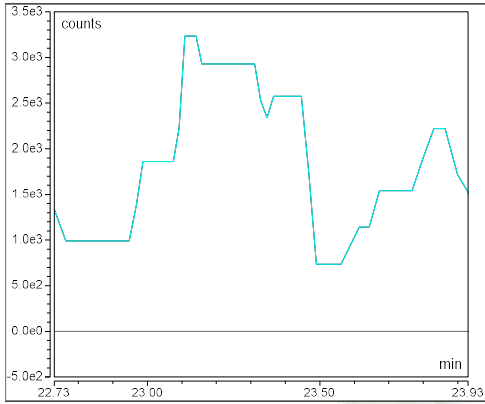
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

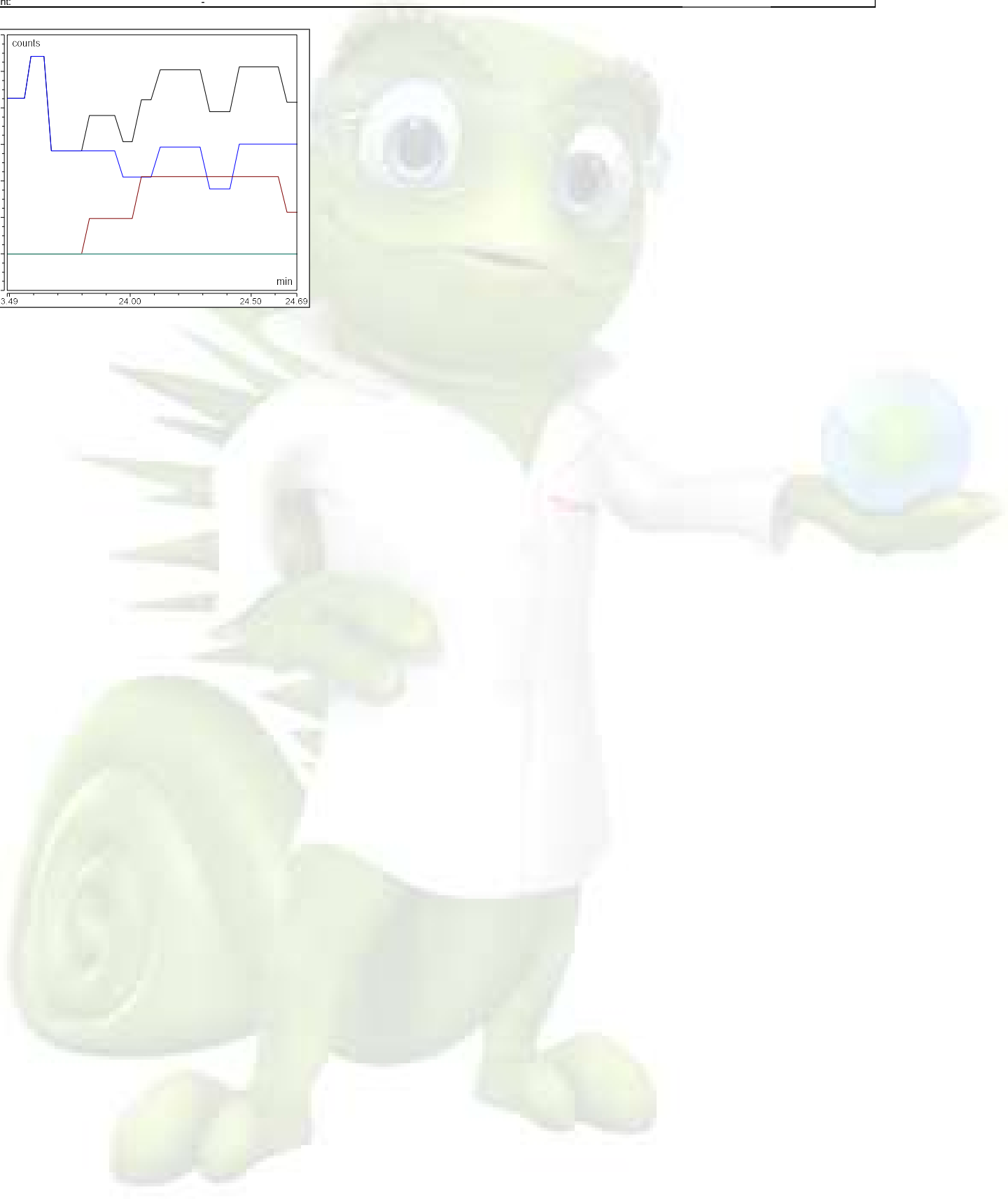
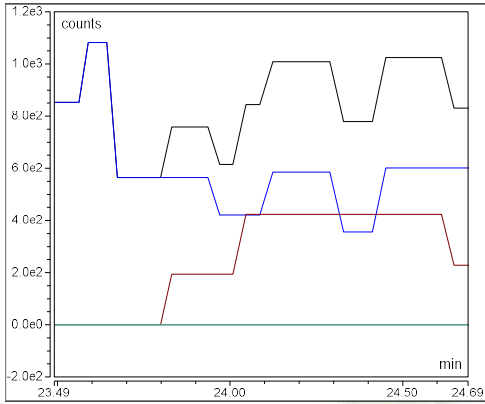
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -



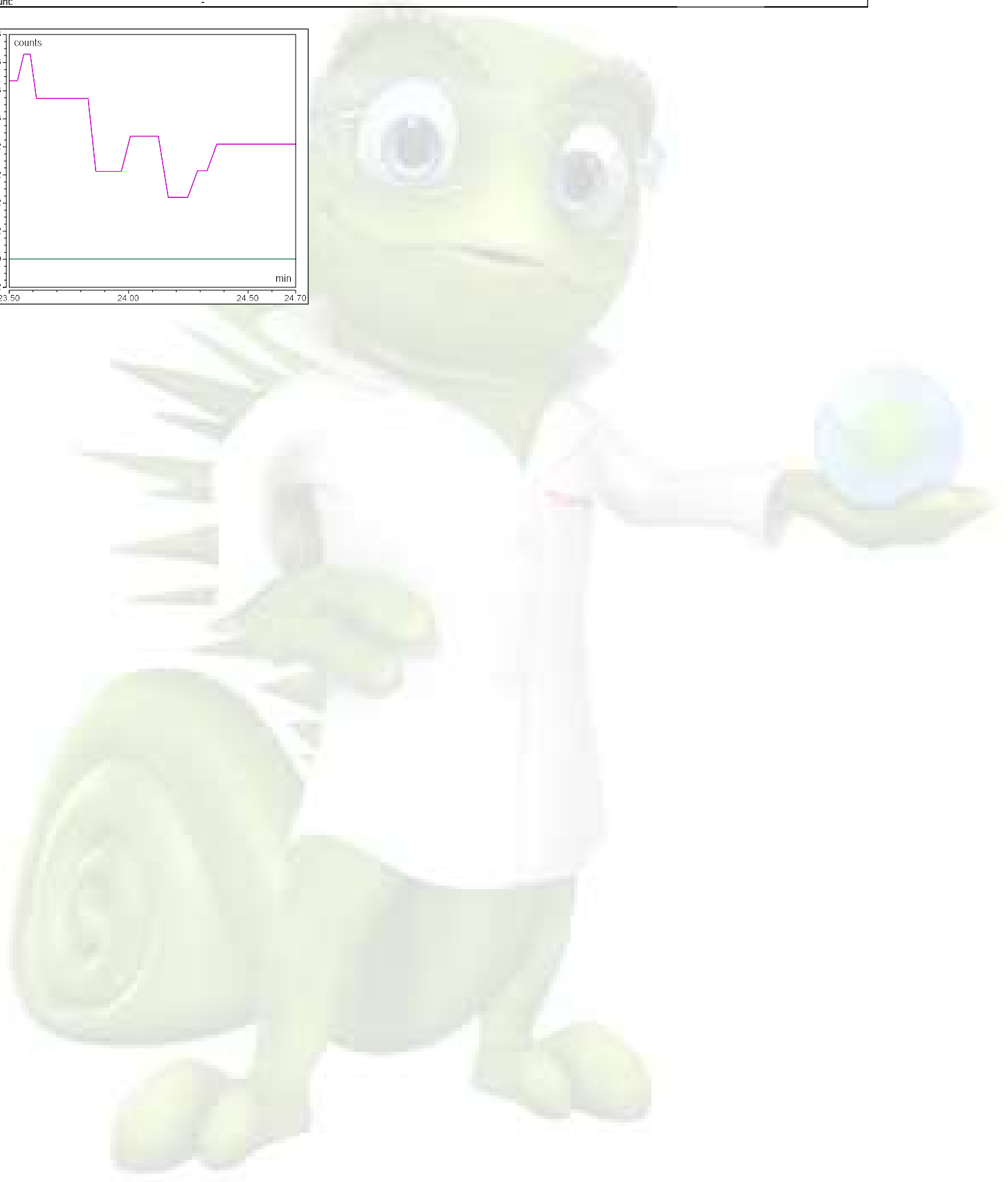
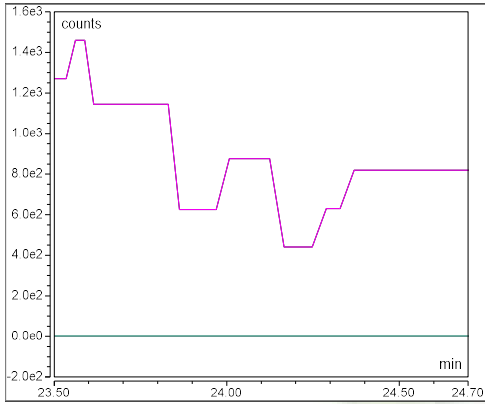


**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

Retention time (min): 24.10  
Expected Mass: 66209.760

Area (count\*min): -  
Intensity (counts): -  
Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

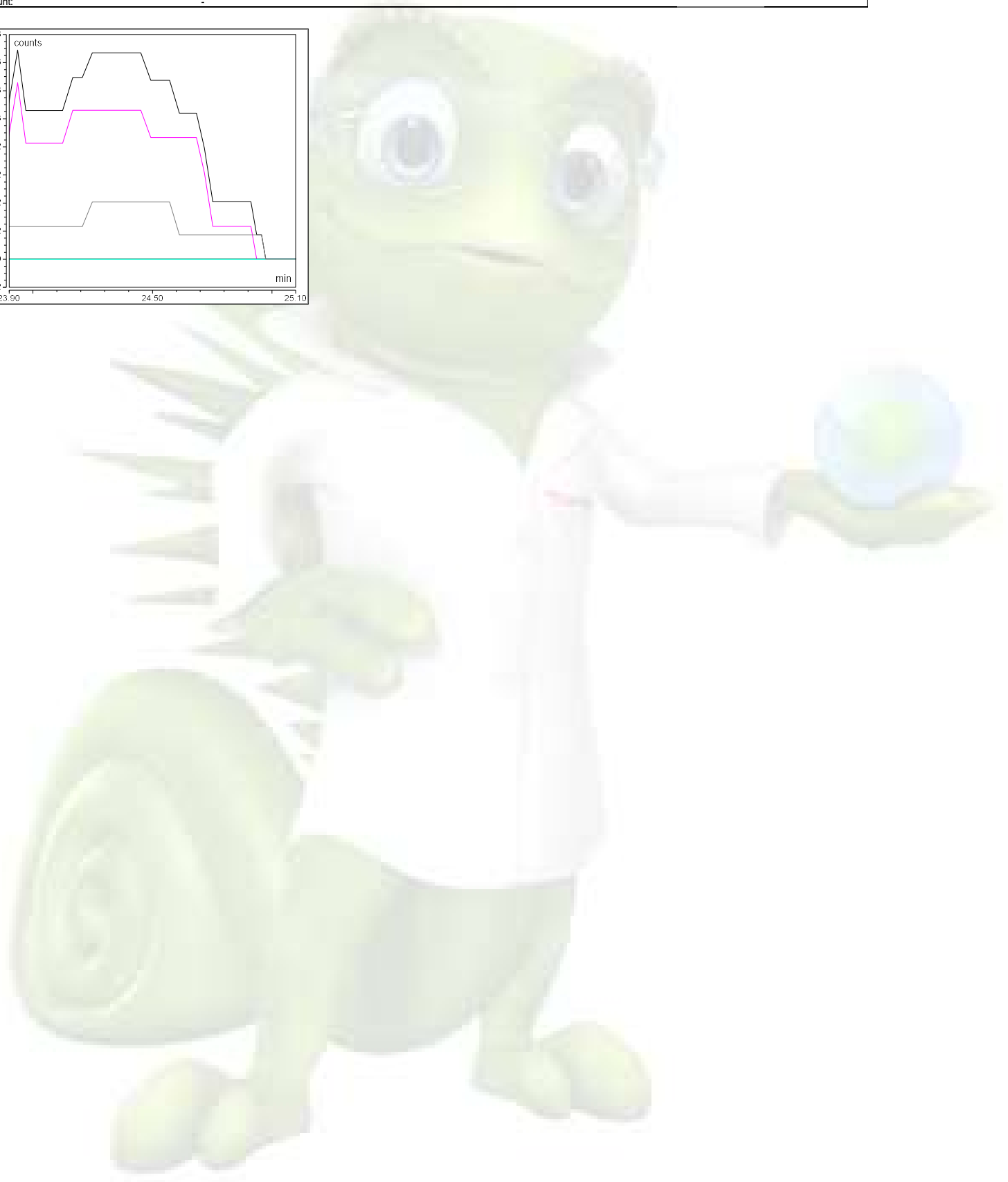
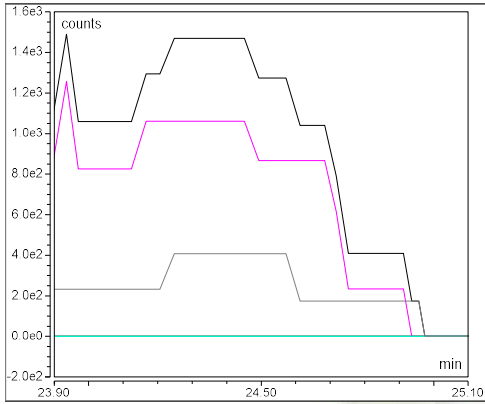
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

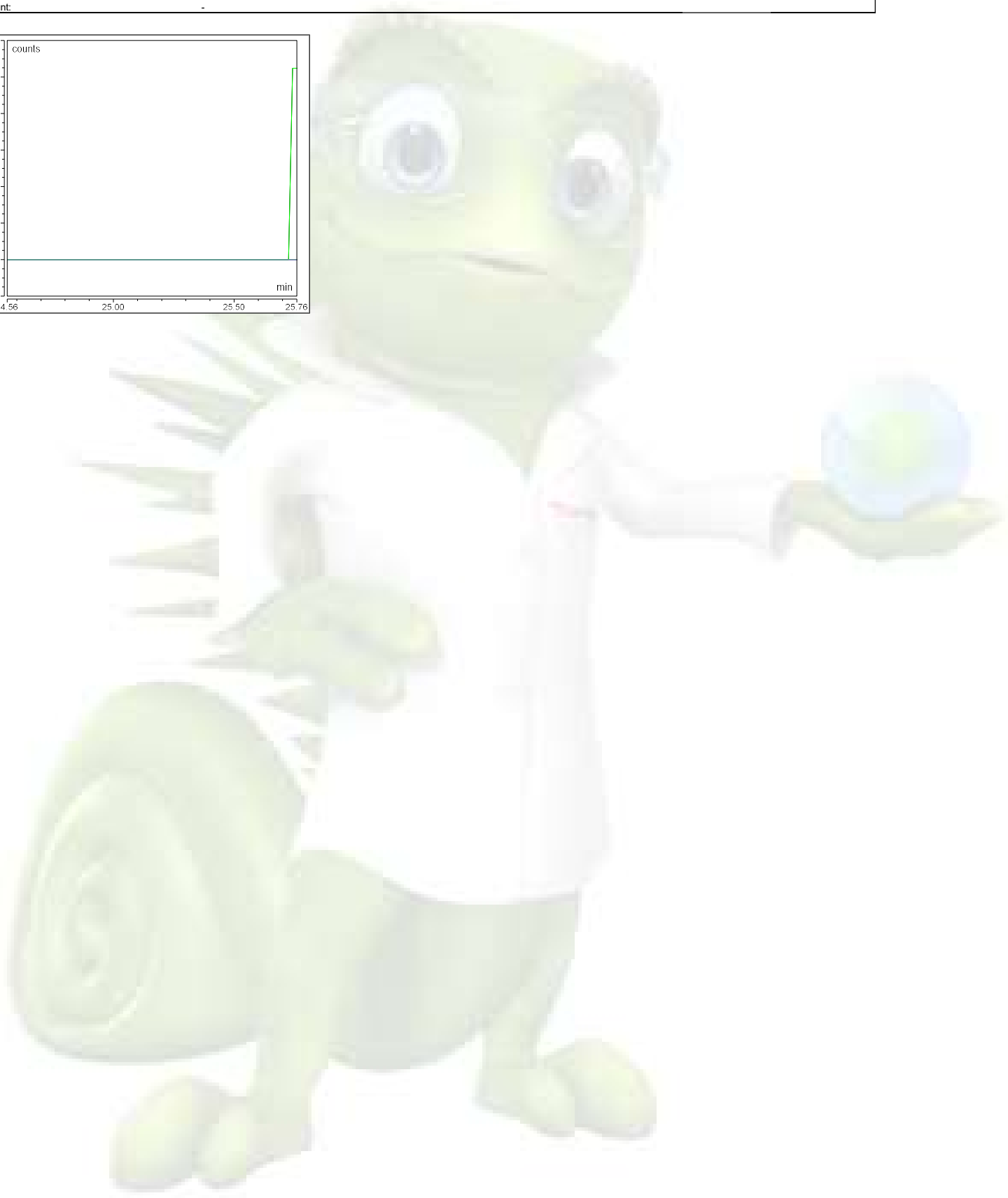
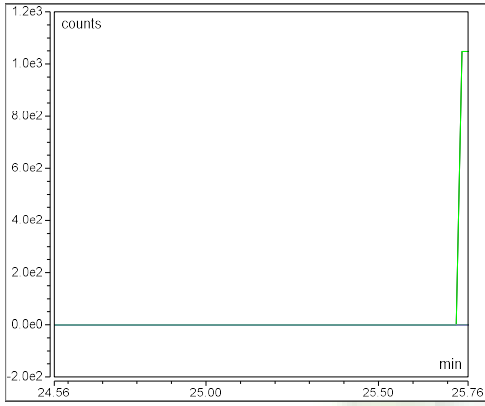
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2\_VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

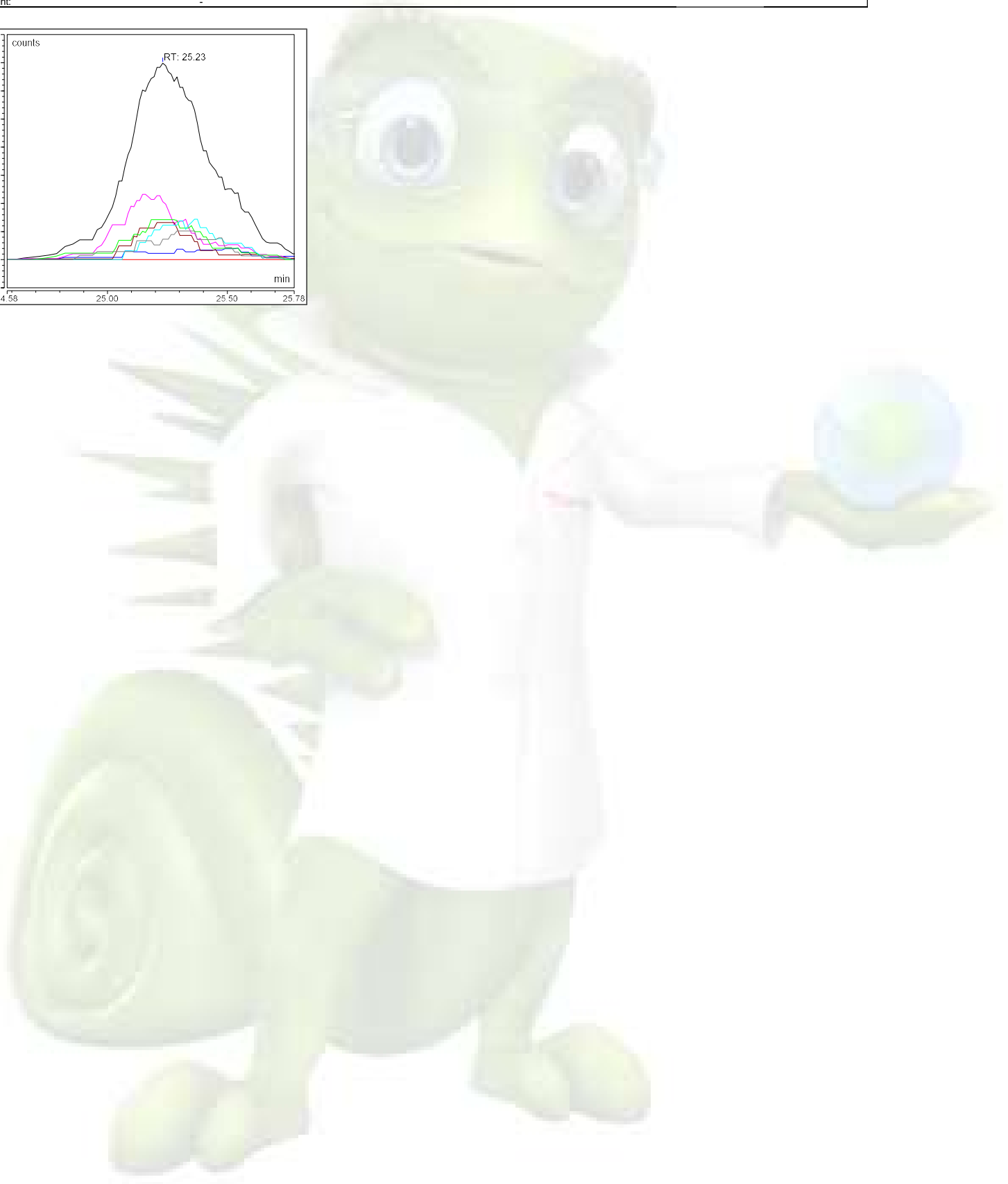
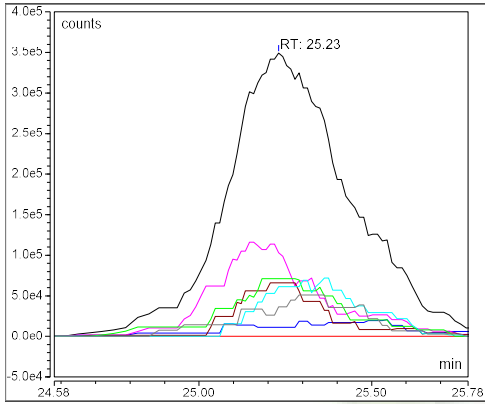
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): 145835

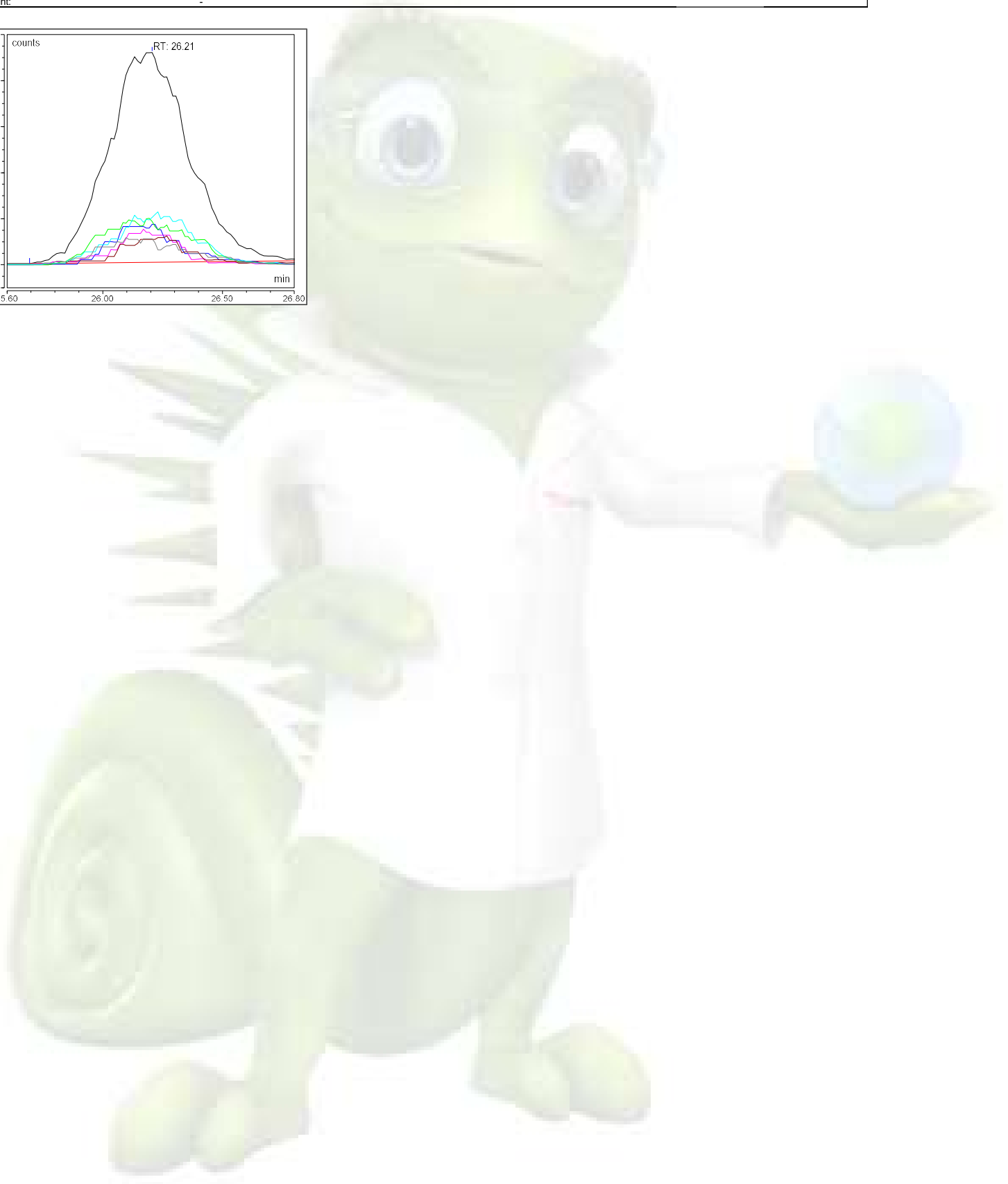
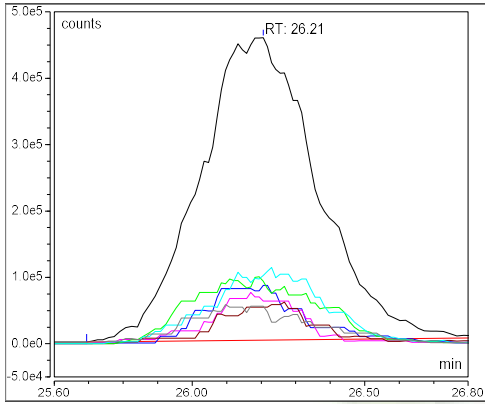
Intensity (counts): 349122

Amount: -



**Component Name: AAV2 VP2**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 |
| Retention time (min): | 26.20   |
| Expected Mass:        | 66487.900   |
| Area (count*min):     | 179370  |
| Intensity (counts):   | 455688  |
| Amount:               | -   |

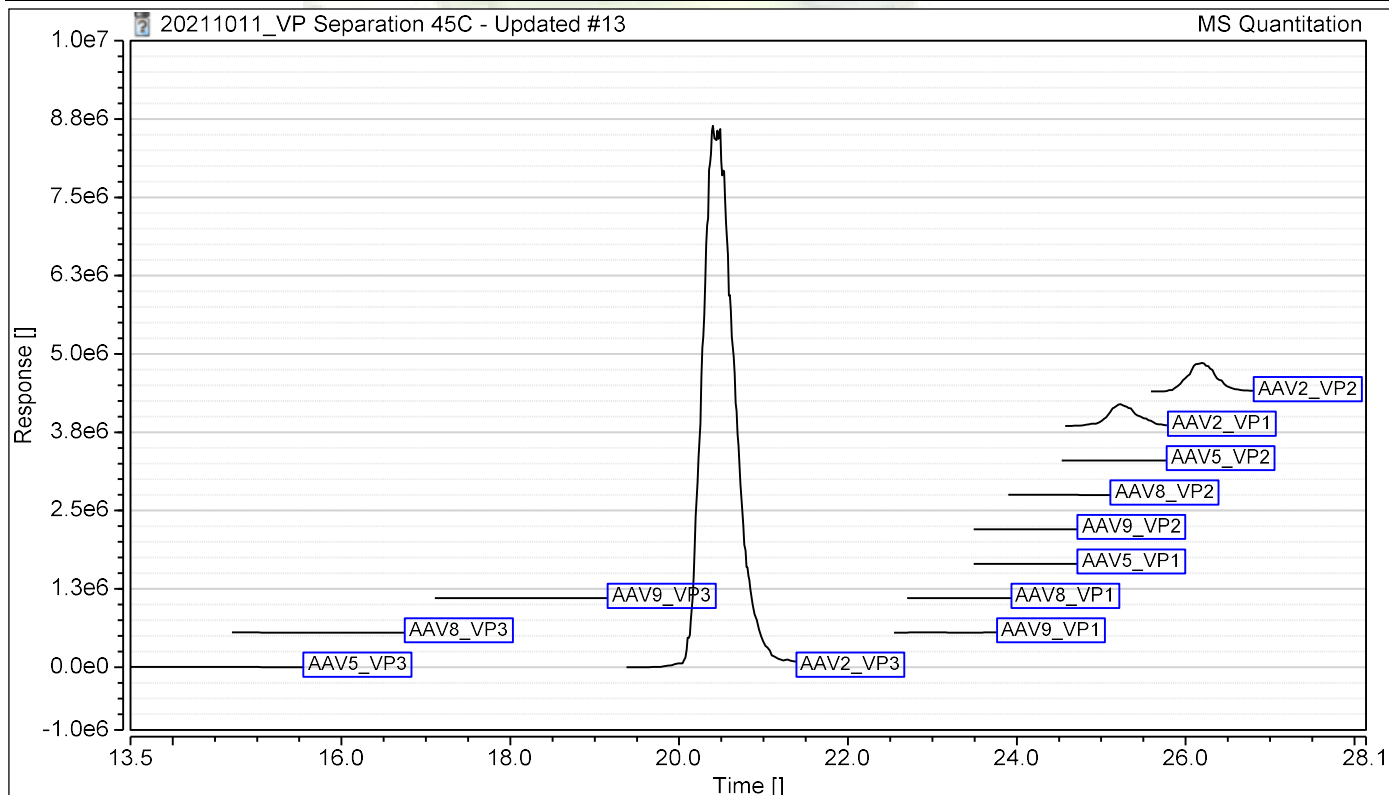


## Chromatogram and SST Results

### Injection Details

|                      |  |                   |        |
|----------------------|--|-------------------|--------|
| Injection Name:      | 20211011_AAV2 Mixed_45C_65-59B_Mod1_1ug_1                                    | Run Time (min):   | 45.00  |
| Vial Number:         | R:A9   | Injection Volume: | 8.45   |
| Injection Type:      | Unknown  |                   |        |
| Calibration Level:   |  |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 S |                   |        |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 12/Oct/21 18:13  | Sample Weight:    | 1.0000 |

### Chromatogram

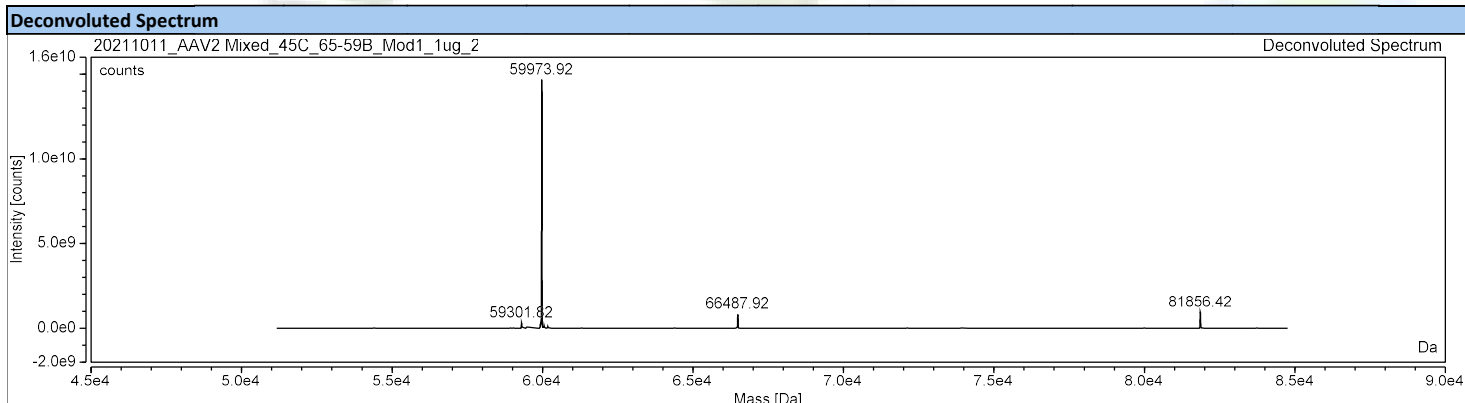
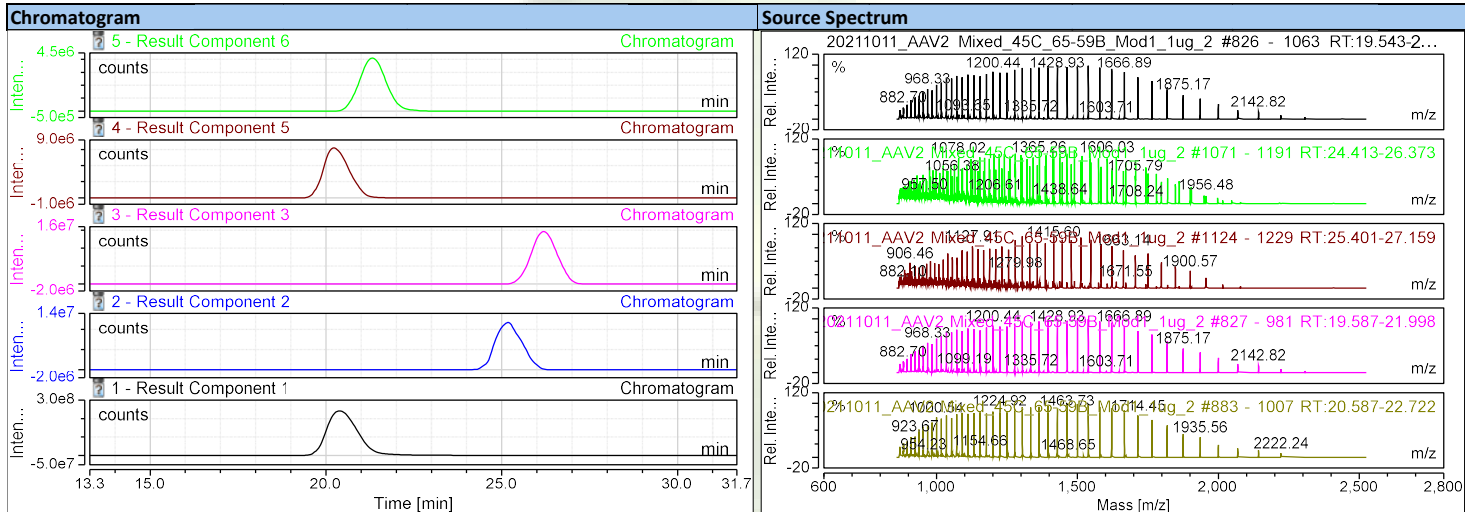


### SST Results

| No.                                 | Name | Inj.Condition | Peak | Test Result   | Injection |
|-------------------------------------|------|---------------|------|---------------|-----------|
| Number of executed test cases: n.a. |      |               |      | Total Result: | Passed    |

## Protein Deconvolution Summary

| Injection Details |   |  |
|-------------------|---|--|
| Injection Name:   | 20211011_AAV2 Mixed_45C_65-59B_Mod1_1ug_2 | Injection Volume: 8.45                                       |
| Vial Number:      | R:A9                                      | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                                   | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 44.98                                     | Injection Date/Time: 12/Oct/21 18:59                         |



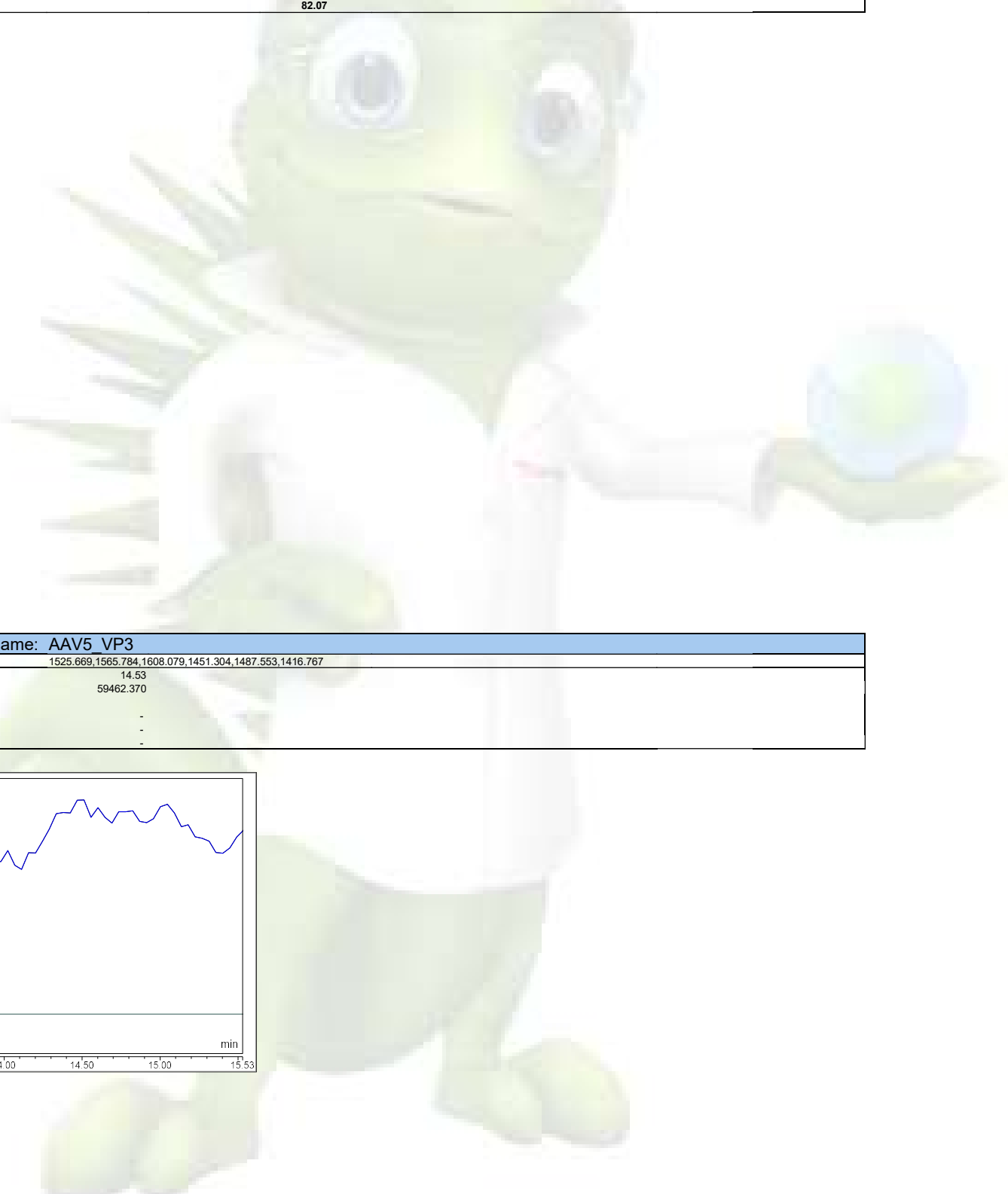
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

### Component Summary

#### Injection Details

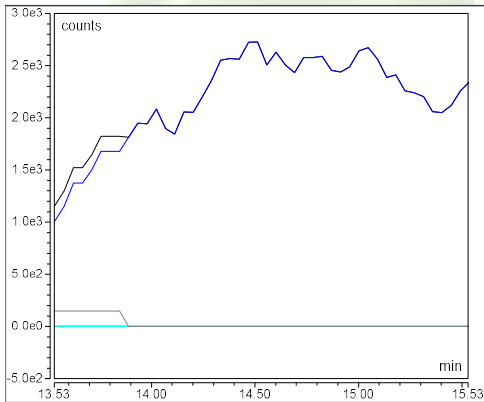
|   |  |  |
|---|--|--|
| Injection Name: 20211011_AAV2 Mixed_45C_65-59B_Mod1 | Injection Volume: 8.45   |  |
| Vial Number: R:A9                                   | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |  |
| Injection Type: Unknown                             | Processing Method: MS Quantitative_45C_65-59B                            |  |
| Run Time (min): 44.98                               | Injection Date/Time: 12/Oct/21 18:59                                     |  |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3     | 20.453                | 2637005.867        | 75.85              | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 | 59973.780 | PASS                           |
| AAV2_VP1     | 25.142                | 91155.345          | 2.62               | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 | 81854.900 | PASS                           |
| AAV2_VP2     | 26.152                | 124930.221         | 3.59               | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 | 66487.900 | PASS                           |
| <b>82.07</b> |                       |                    |                    |   |           |                                |



#### Component Name: AAV5\_VP3

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| Retention time (min): | 14.53   |
| Expected Mass:        | 59462.370   |
| Area (count*min):     | -   |
| Intensity (counts):   | -   |
| Amount:               | -   |





**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

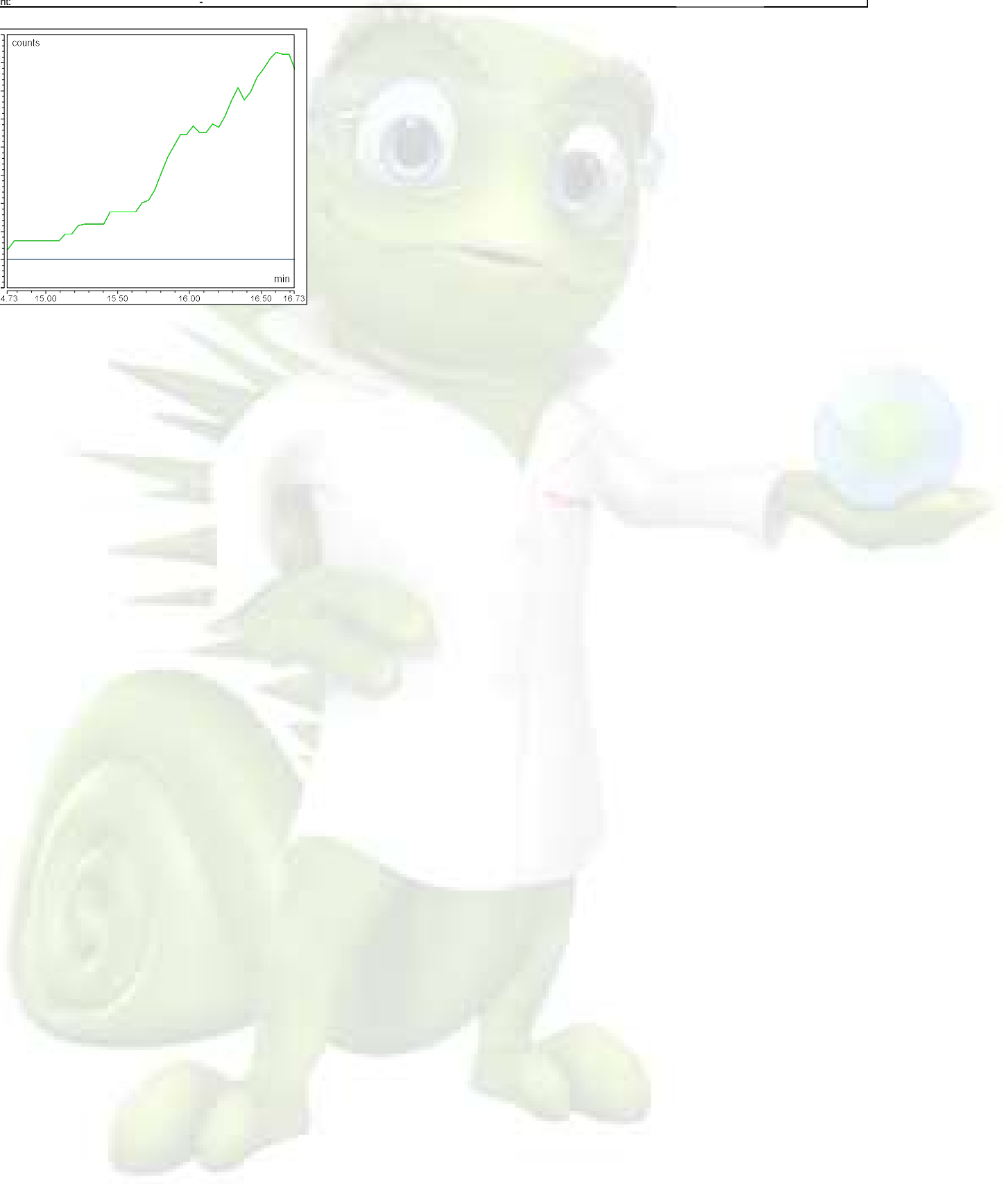
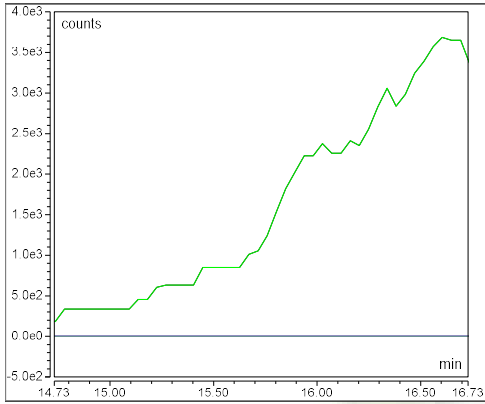
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

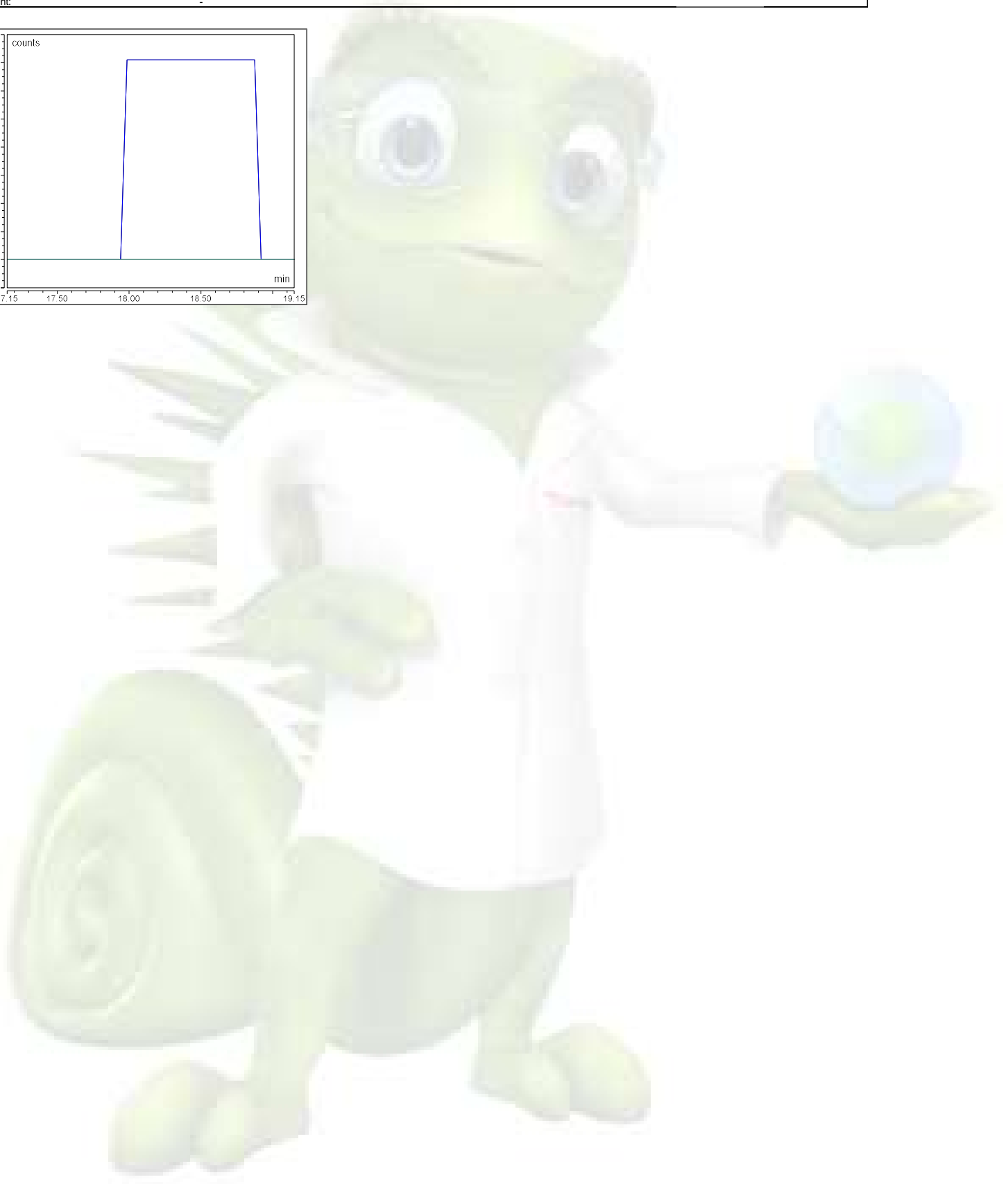
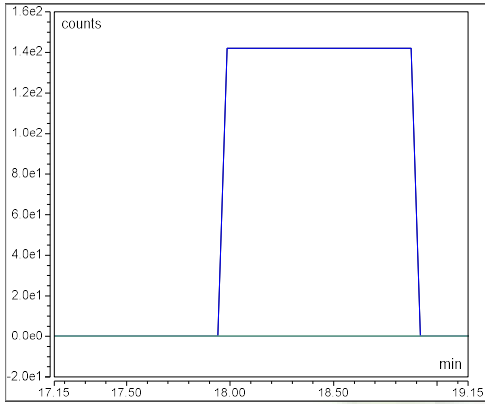
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

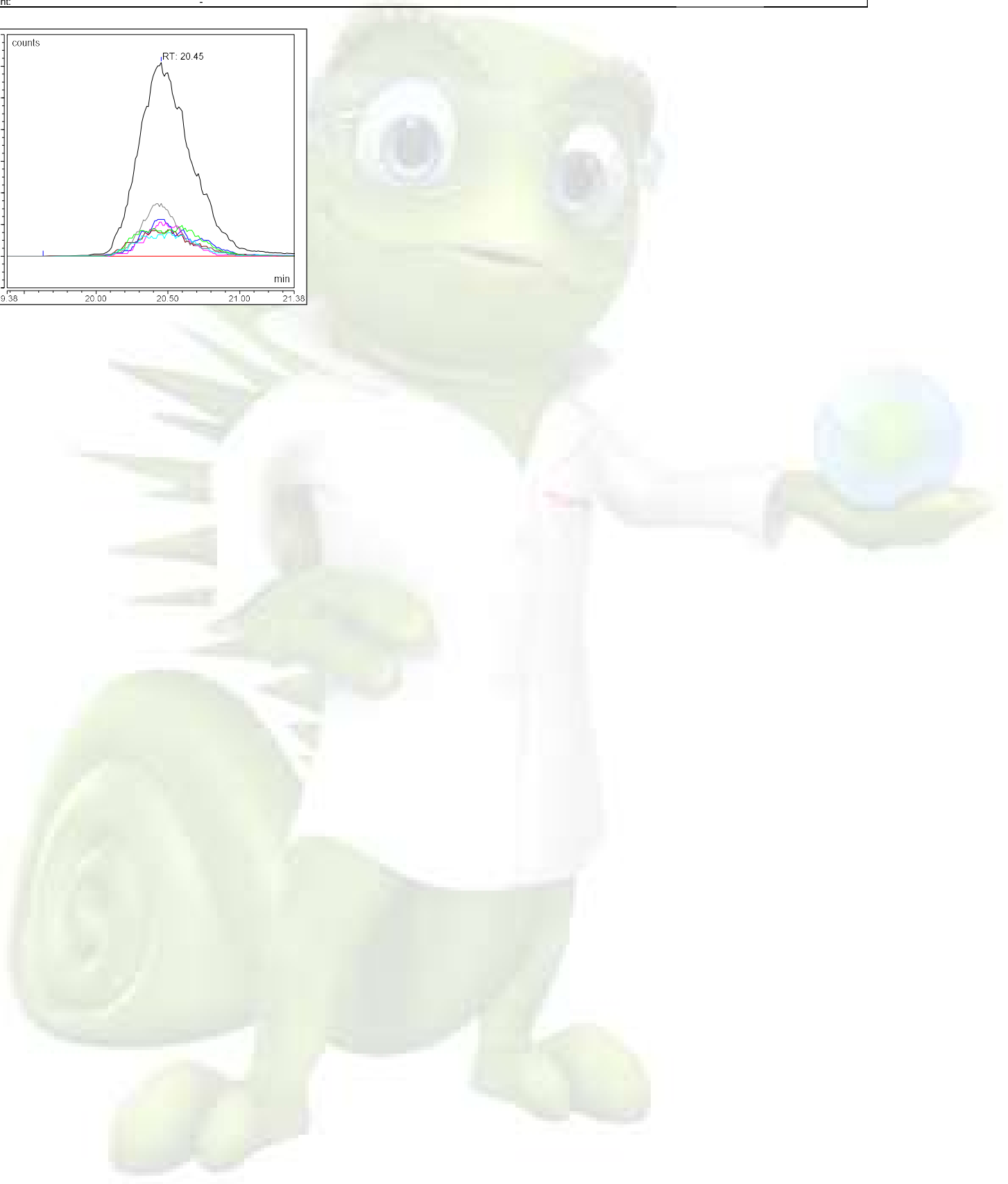
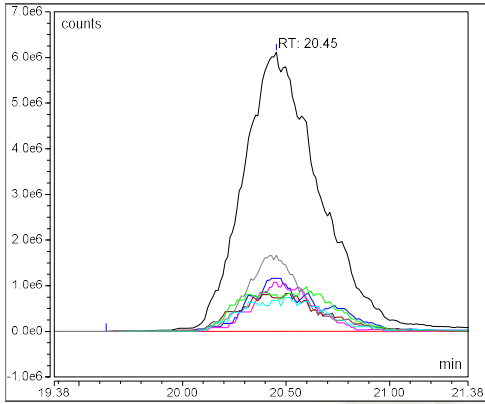
Intensity (counts): -

Amount: -



**Component Name: AAV2 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 |
| Retention time (min): | 20.38   |
| Expected Mass:        | 59973.780   |
| Area (count*min):     | 2637006   |
| Intensity (counts):   | 6113568   |
| Amount:               | -   |



**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

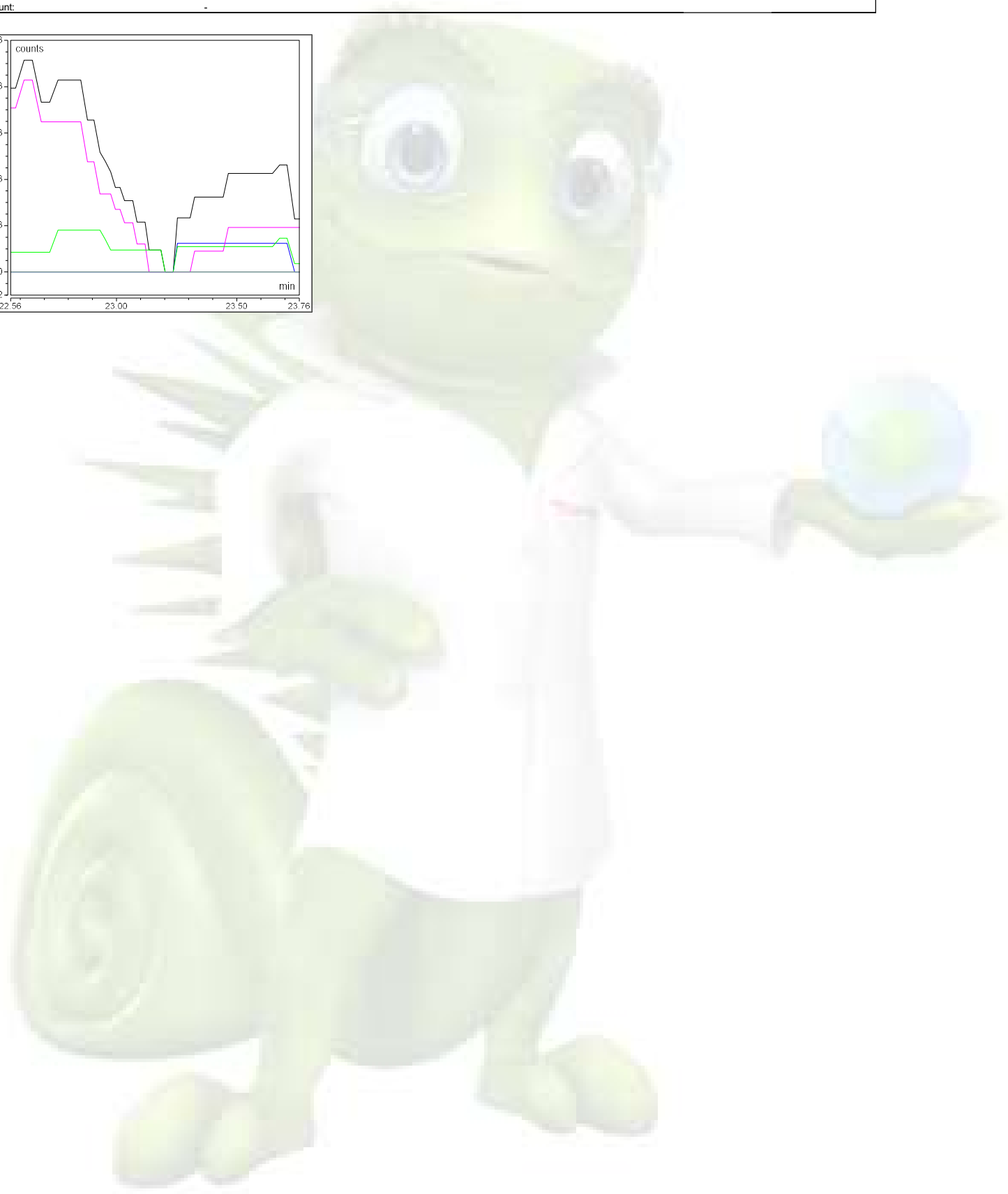
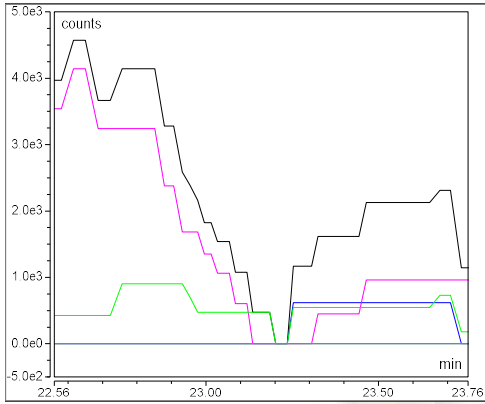
Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

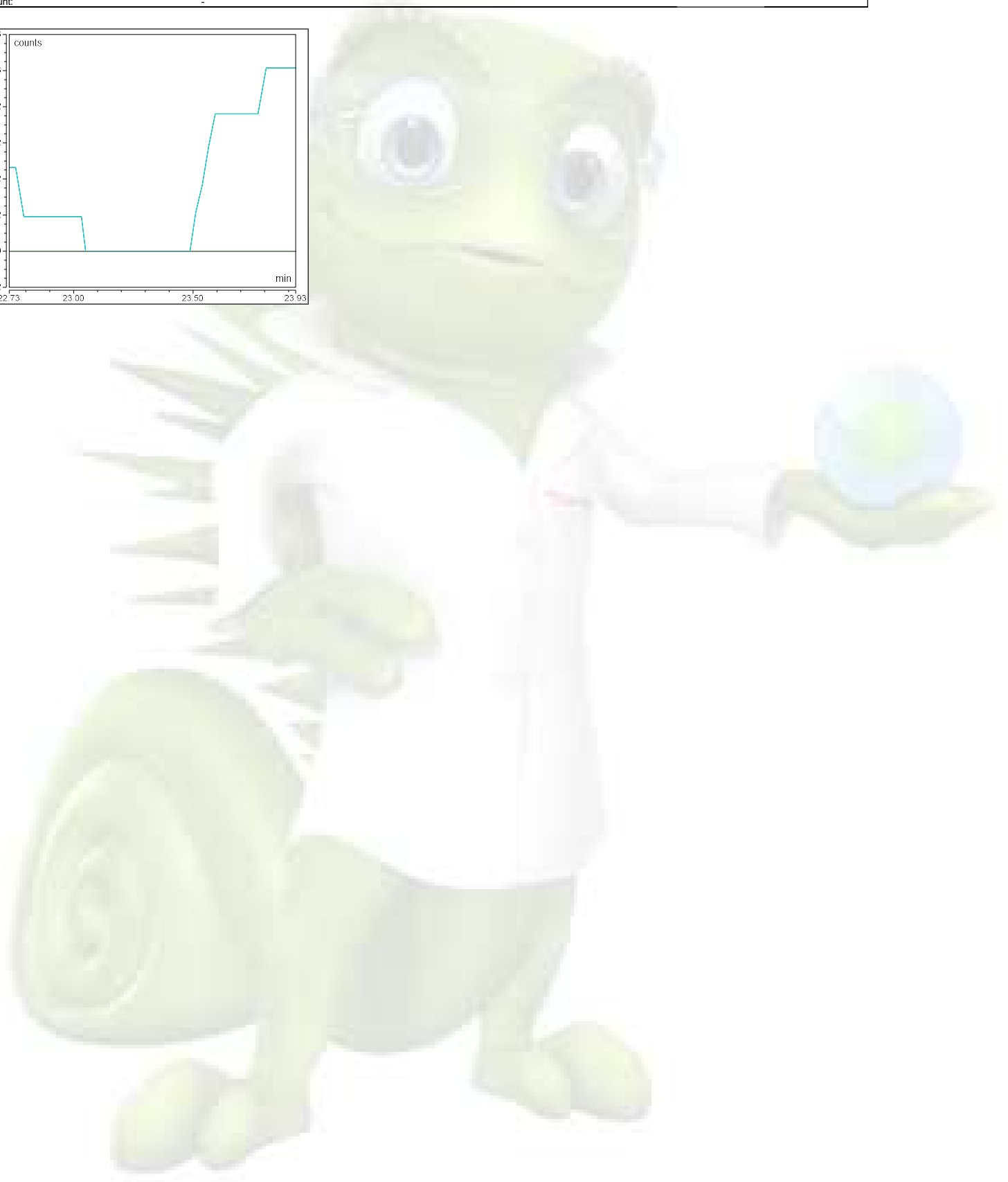
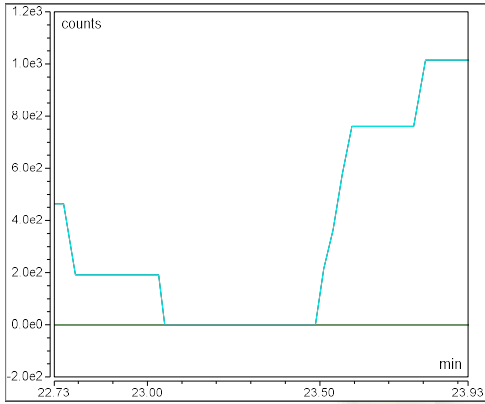
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

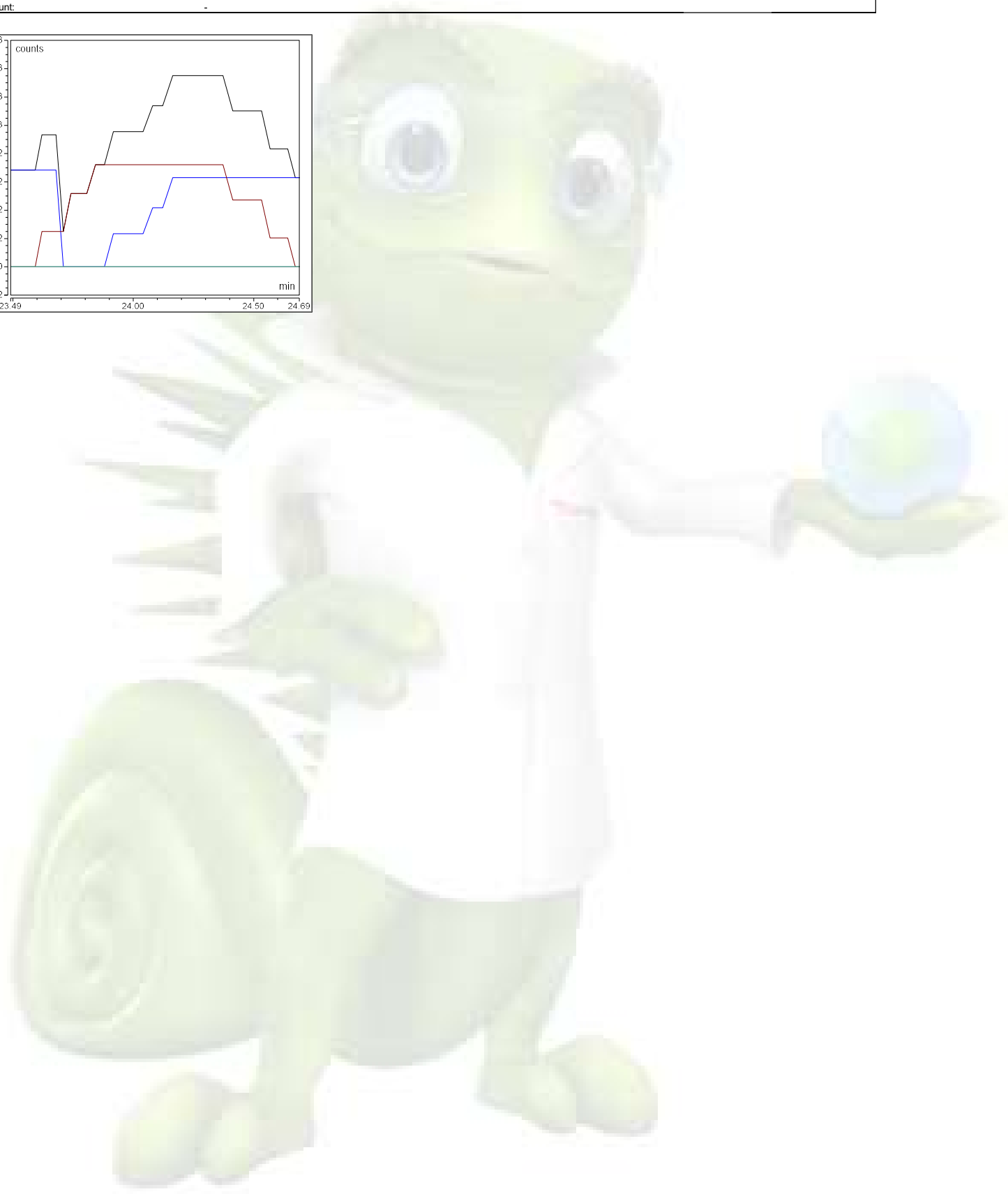
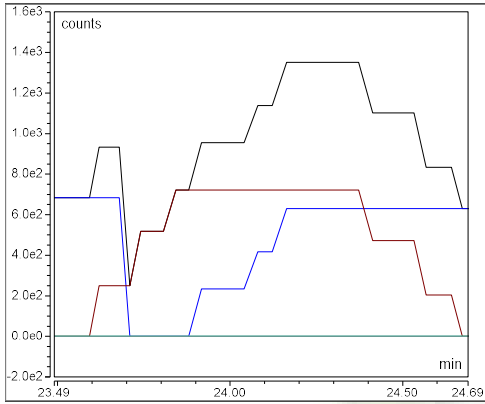
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

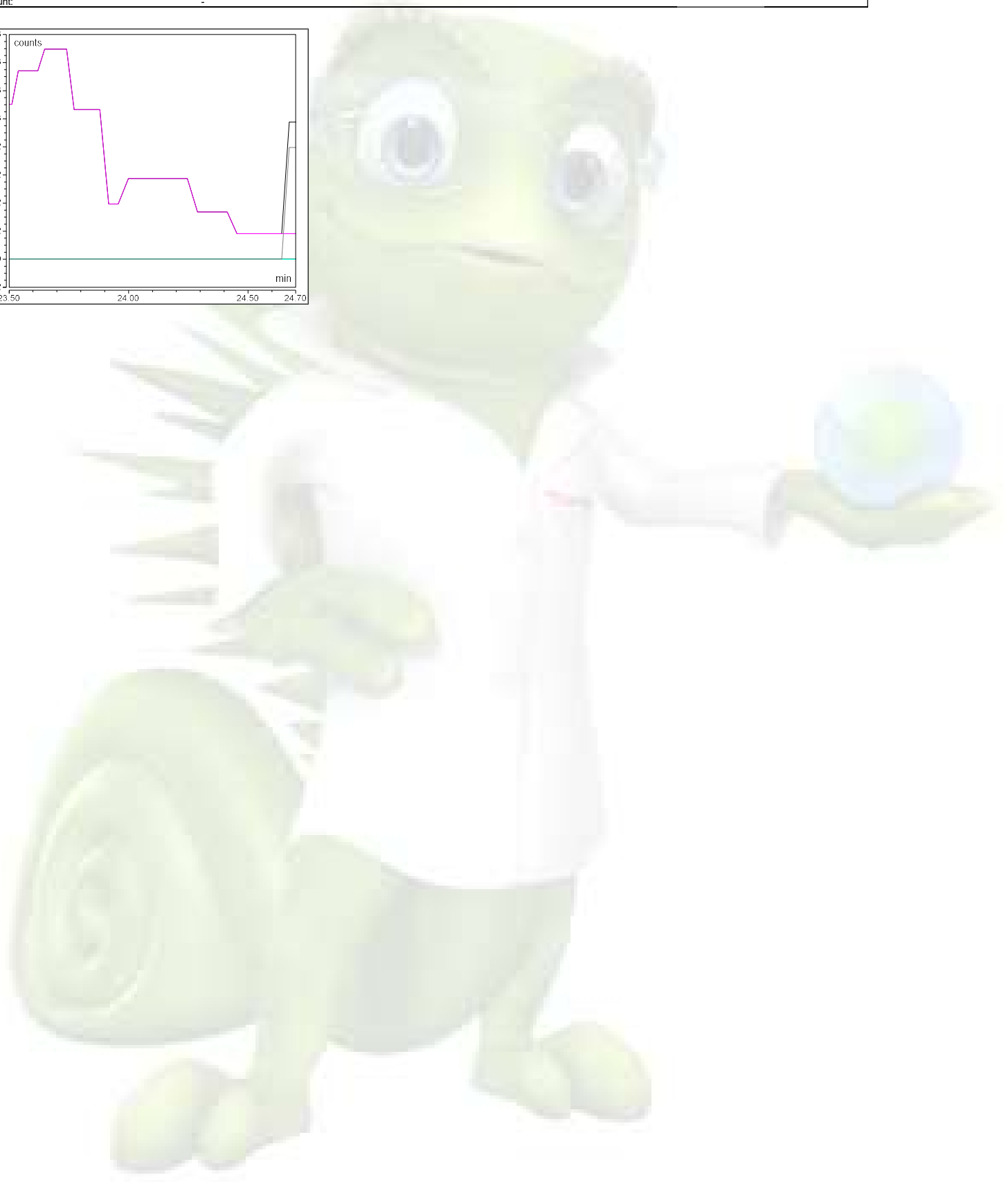
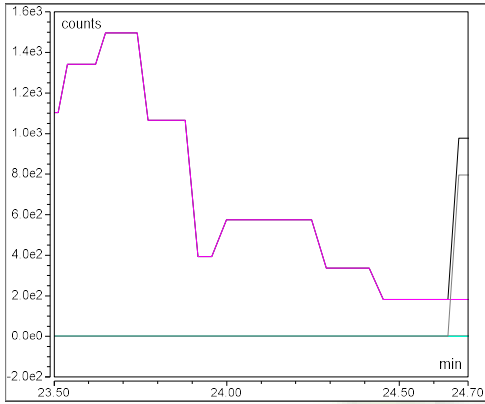
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

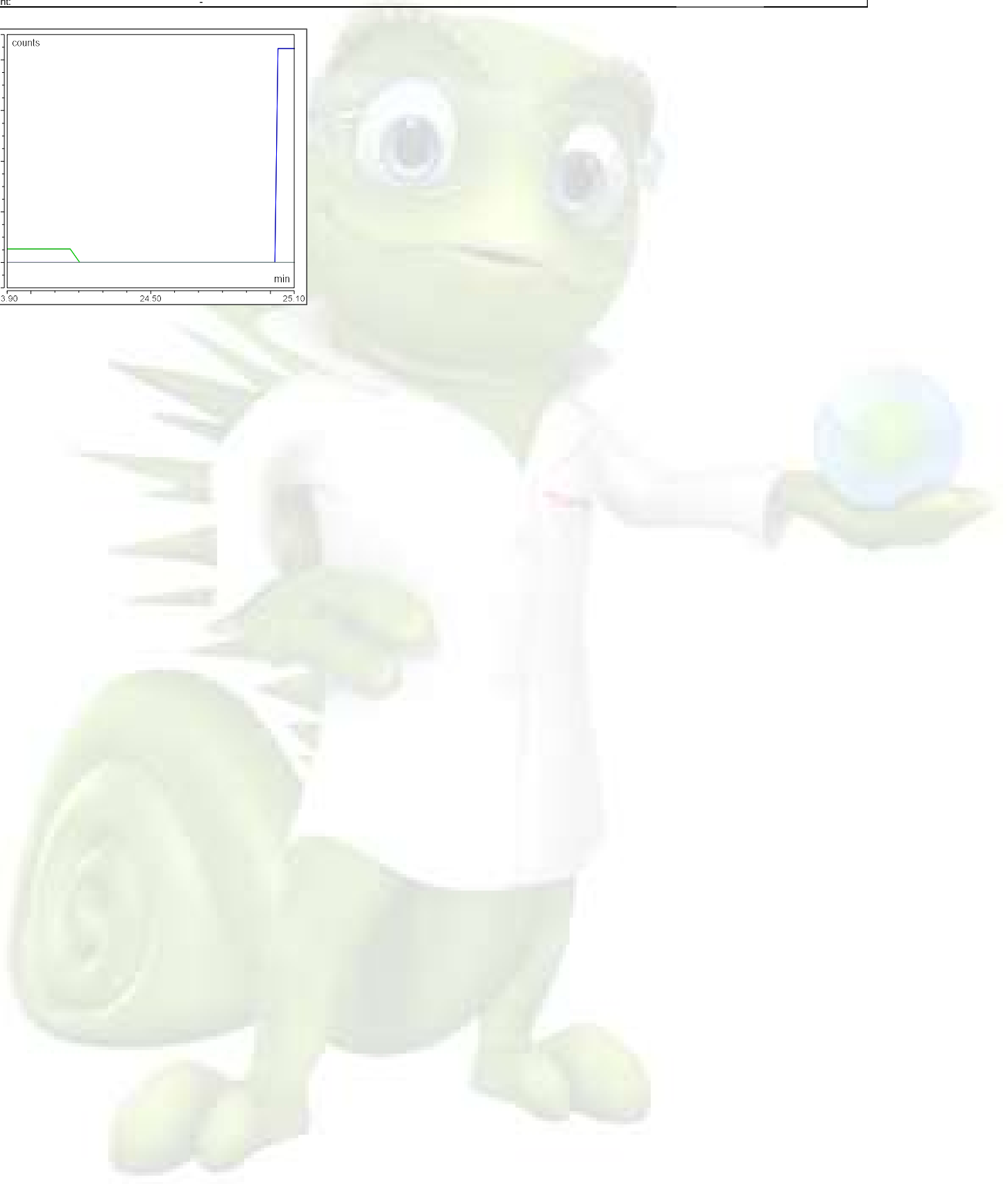
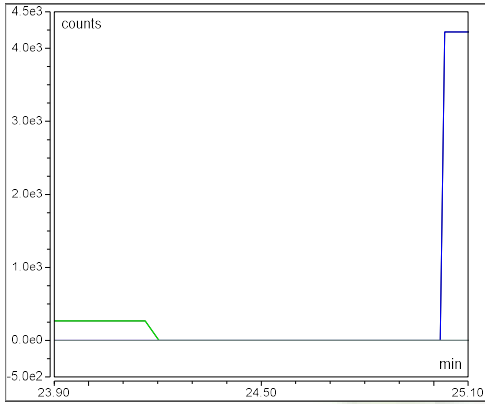
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -





**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

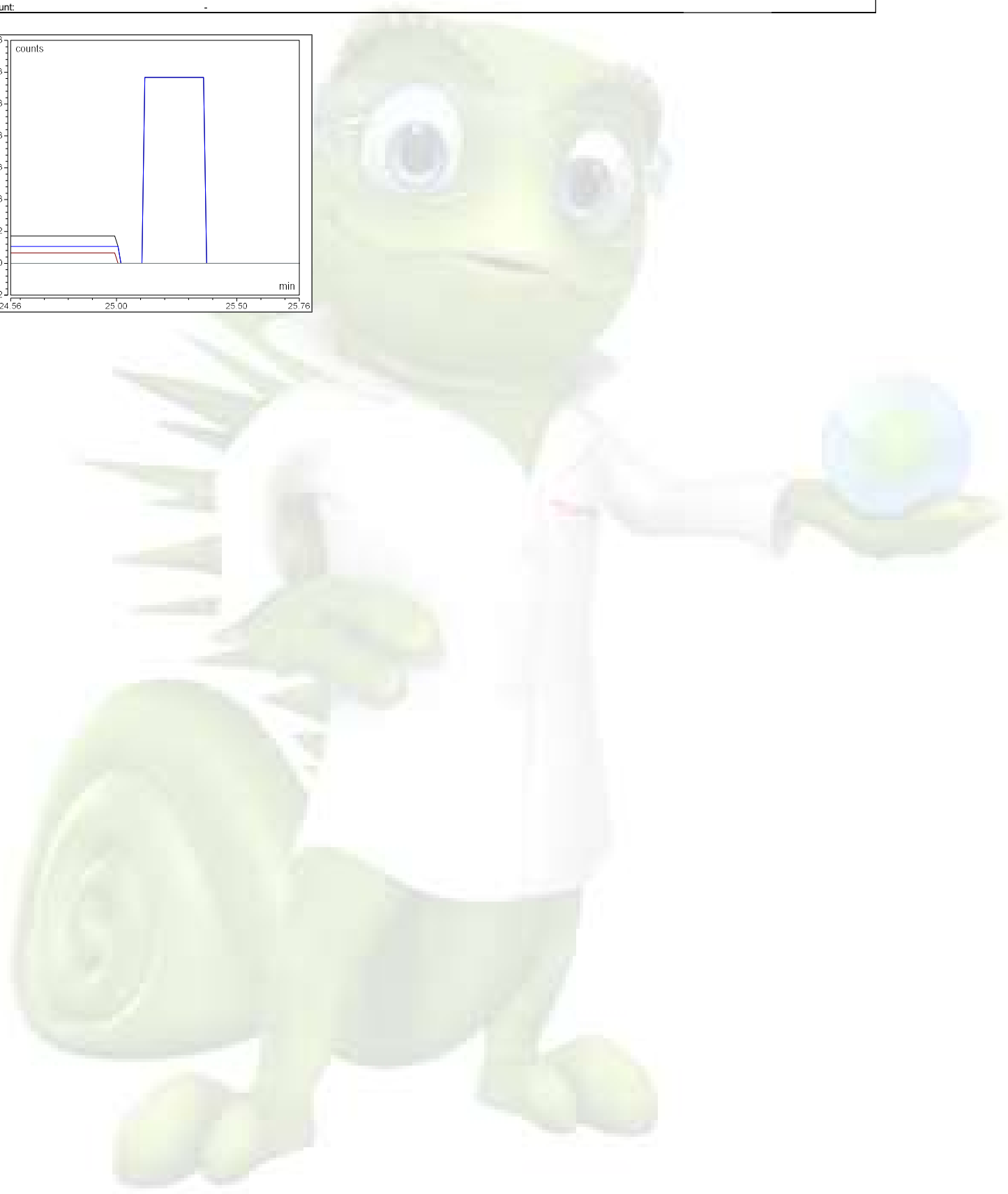
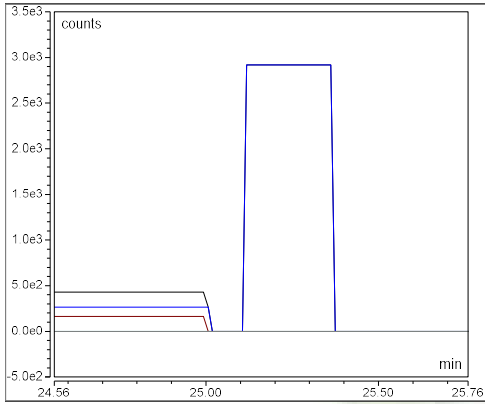
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

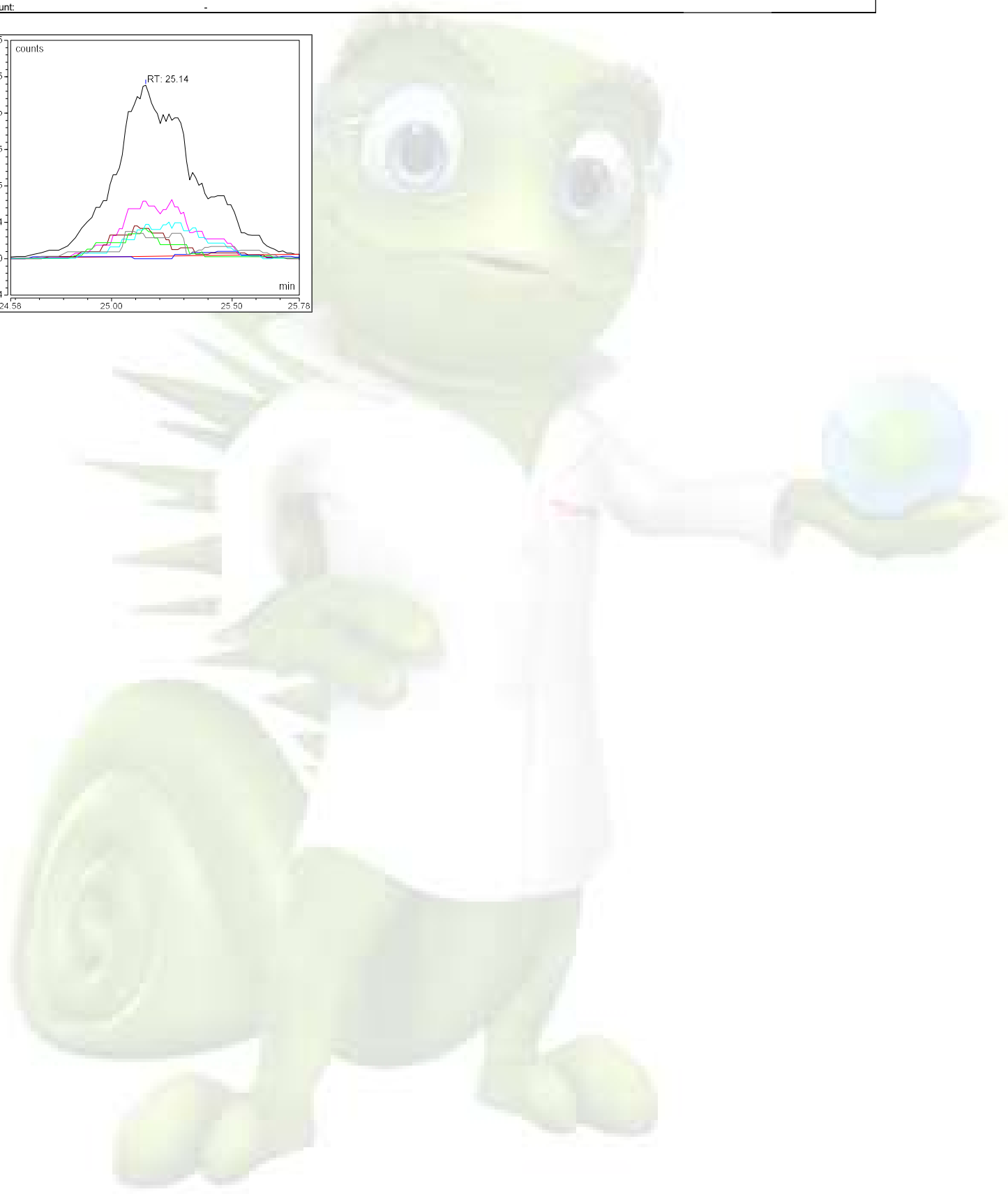
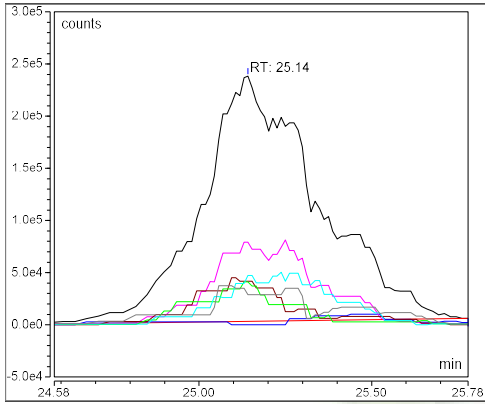
Intensity (counts): -

Amount: -



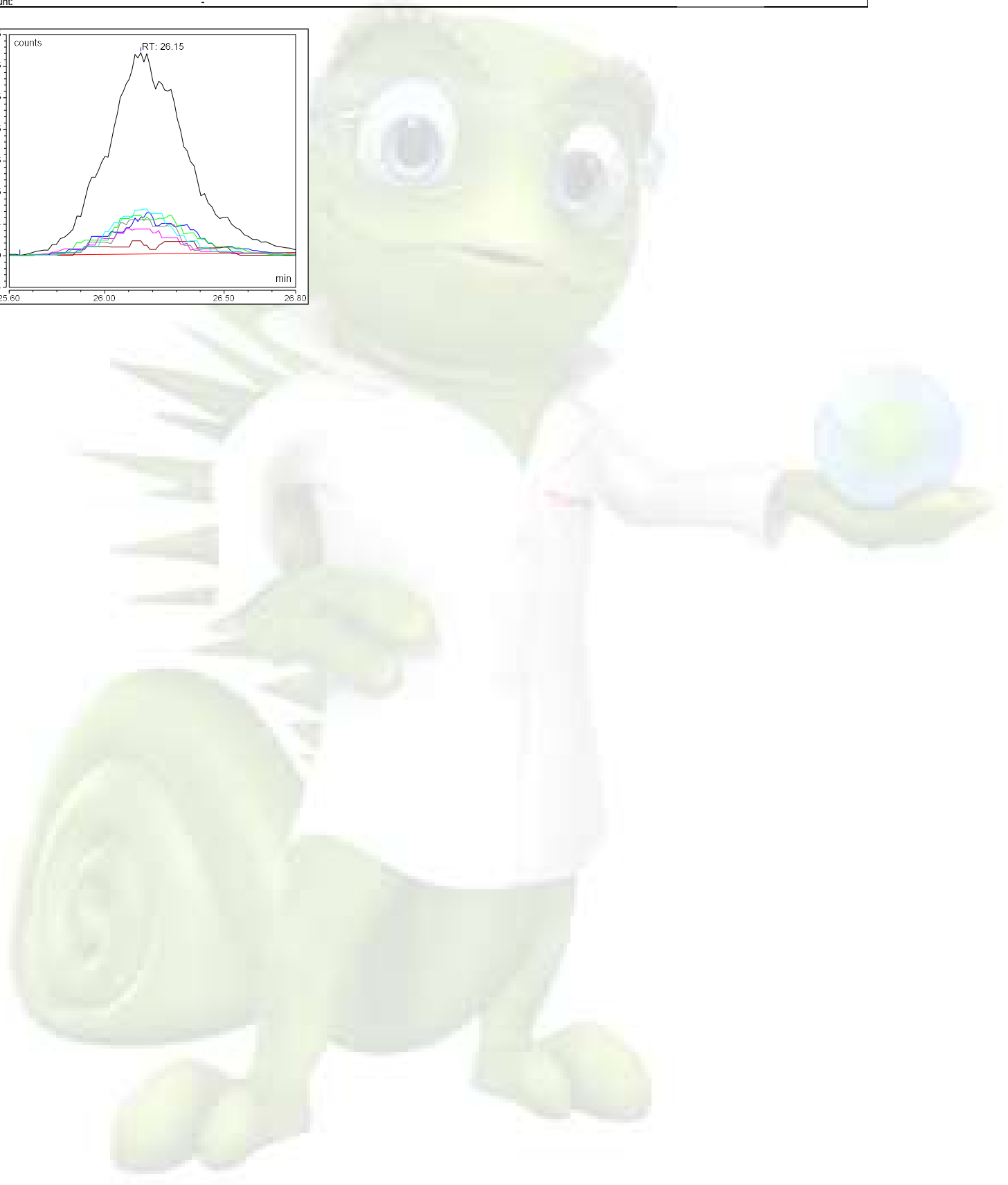
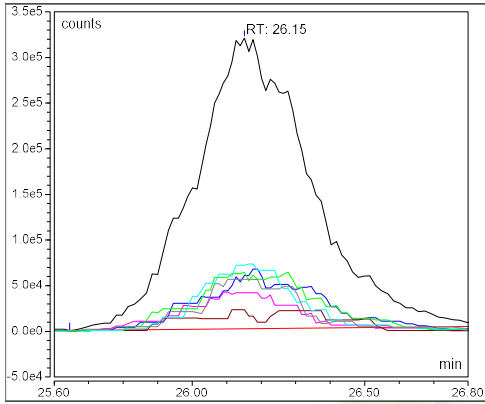
**Component Name: AAV2 VP1**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 |
| Retention time (min): | 25.18   |
| Expected Mass:        | 81854.900   |
| Area (count*min):     | 91155   |
| Intensity (counts):   | 235208  |
| Amount:               | -   |



**Component Name: AAV2 VP2**

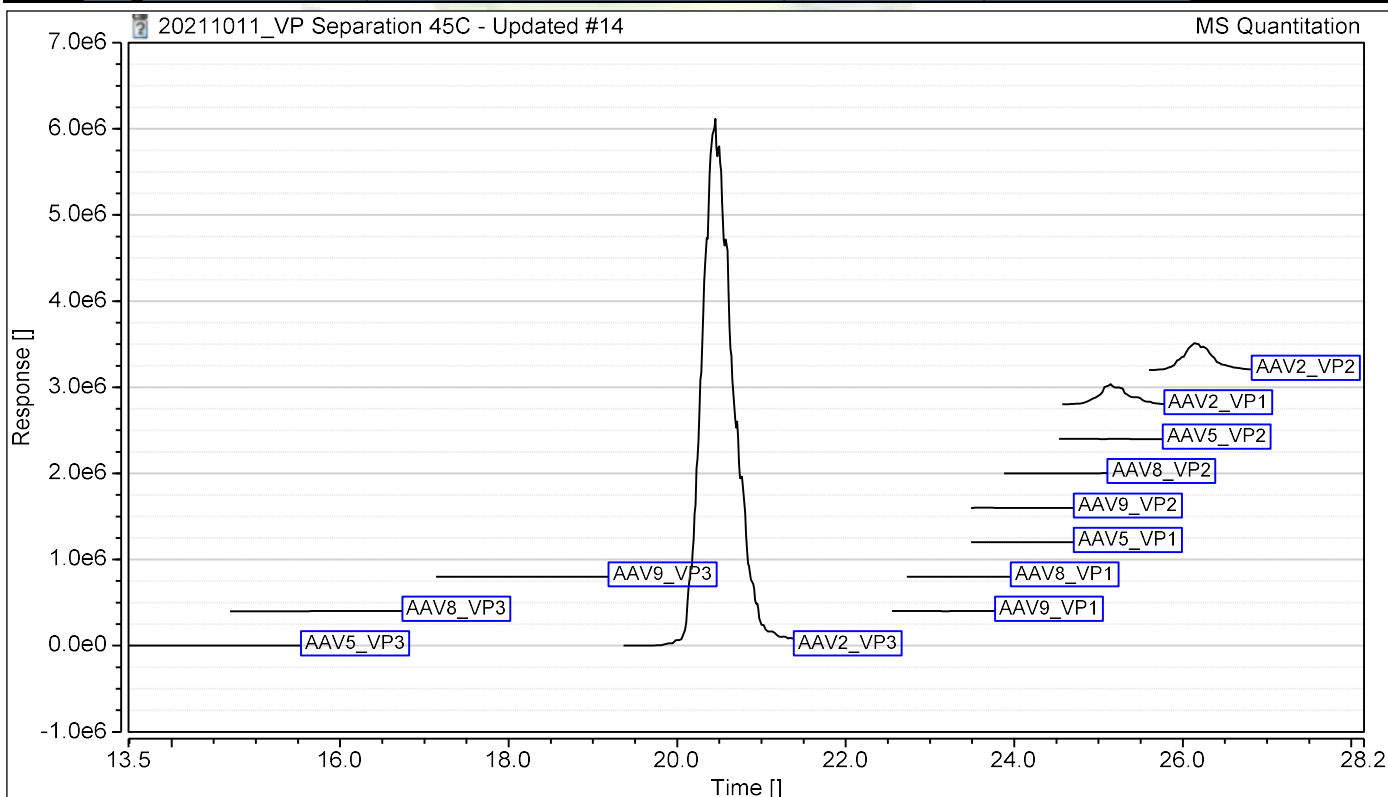
|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 |
| Retention time (min): | 26.20   |
| Expected Mass:        | 66487.900   |
| Area (count*min):     | 124930  |
| Intensity (counts):   | 318436  |
| Amount:               | -   |



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV2 Mixed_45C_65-59B_Mod1_1ug_2                                  | Run Time (min): 44.98   |
| Vial Number:         | R:A9   | Injection Volume: 8.45  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 | S                       |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 12/Oct/21 18:59  | Sample Weight: 1.0000   |

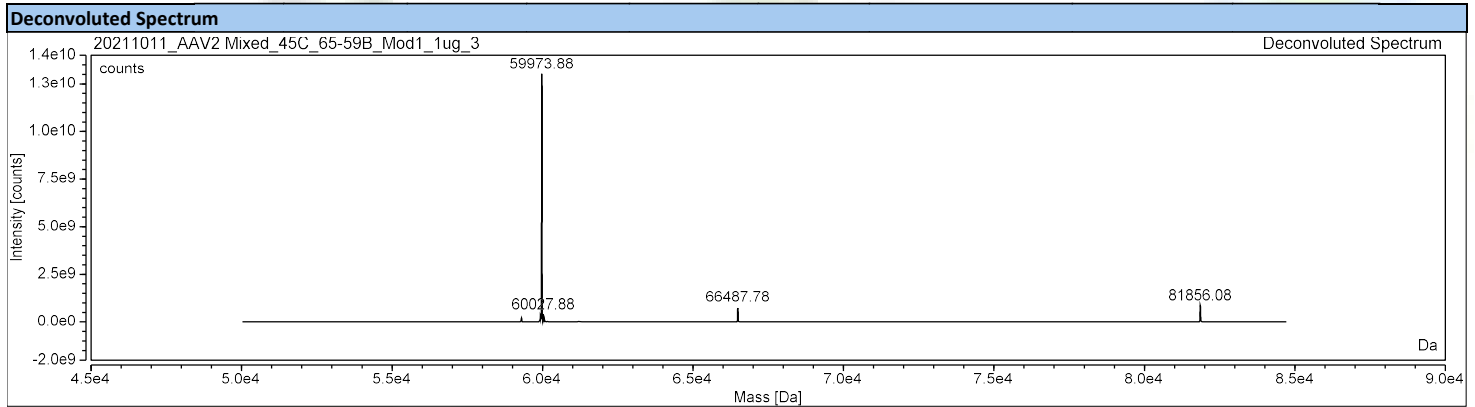
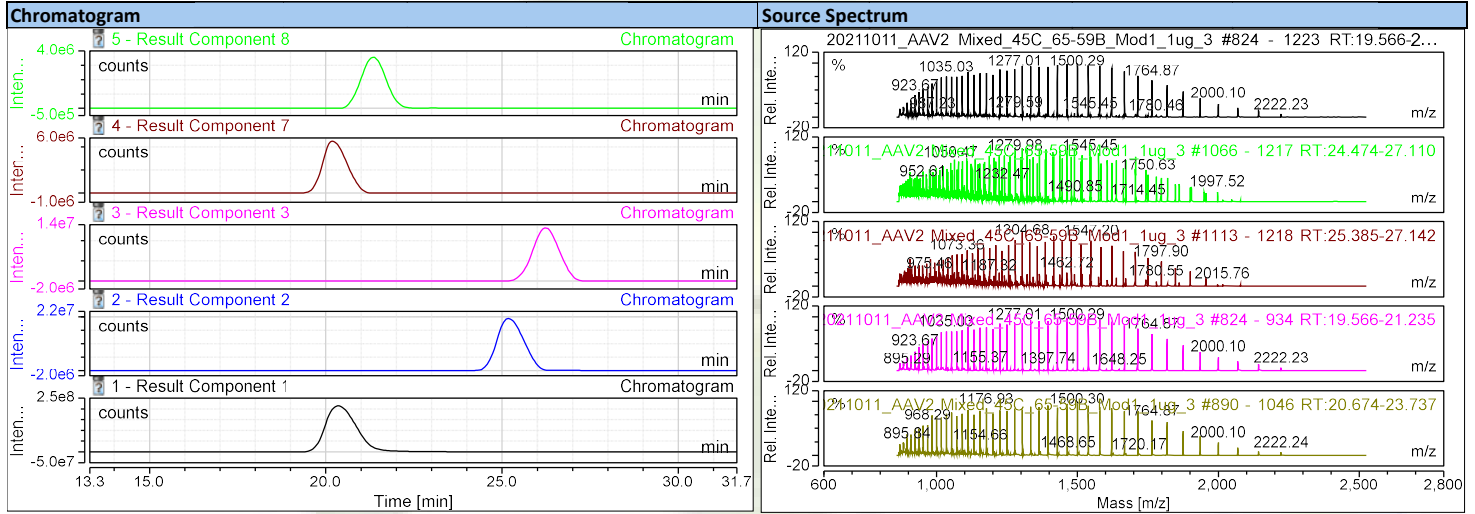
### Chromatogram



| SST Results                         |      |                |               |             |           |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

## Protein Deconvolution Summary

| Injection Details |   |  |
|-------------------|---|--|
| Injection Name:   | 20211011_AAV2 Mixed_45C_65-59B_Mod1_1ug_3 | Injection Volume: 8.45                                       |
| Vial Number:      | R:A9                                      | Instrument Method: AAV Cap_Prot_Sep_BEH Glycoprotein Amide H |
| Injection Type:   | Unknown                                   | Processing Method: MS Quantitative_45C_65-59B                |
| Run Time (min):   | 44.99                                     | Injection Date/Time: 12/Oct/21 19:45                         |



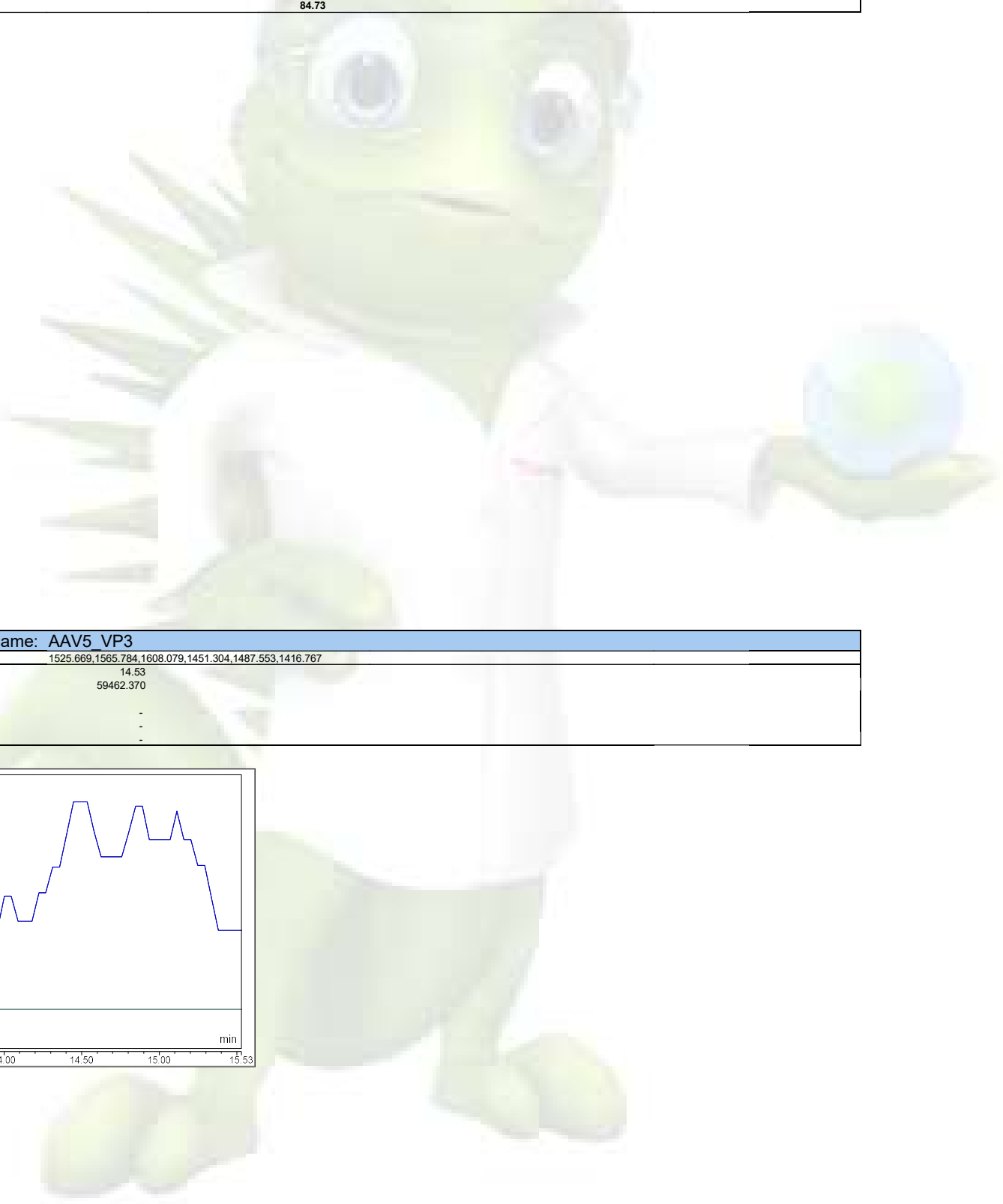
| Deconvolution Results |        |           |              |               |               |                  |                             |                  |                         |        |
|-----------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component      | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
| Result Component 1    | 18.11  | 59733.033 | 0.40         | 6.68          | 0.000         | 27034264760      | 80.1                        | 100.00           | 45                      | 238.14 |
| Result Component 2    | 18.22  | 59820.302 | 0.95         | 15.91         | 87.269        | 1527586438       | 4.5                         | 5.65             | 40                      | 170.79 |
| Result Component 3    | 23.18  | 81291.415 | 0.91         | 11.13         | 21558.382     | 643910272        | 1.9                         | 2.38             | 56                      | 291.15 |
| Result Component 4    | 19.20  | 59813.944 | 0.85         | 14.29         | 80.911        | 471832605        | 1.4                         | 1.75             | 43                      | 234.57 |
| Result Component 5    | 18.27  | 59686.960 | 1.22         | 20.37         | -46.073       | 390164875        | 1.2                         | 1.44             | 32                      | 176.14 |
| Result Component 7    | 24.10  | 66210.268 | 0.85         | 12.85         | 6477.235      | 340089971        | 1.0                         | 1.26             | 44                      | 239.41 |
| Result Component 8    | 21.47  | 59731.938 | 0.90         | 15.10         | -1.095        | 300593658        | 0.9                         | 1.11             | 41                      | 217.18 |
| Result Component 9    | 13.31  | 59733.070 | 0.64         | 10.75         | 0.037         | 298107673        | 0.9                         | 1.10             | 42                      | 257.50 |
| Result Component 11   | 19.18  | 59933.627 | 1.10         | 18.41         | 200.594       | 200302659        | 0.6                         | 0.74             | 40                      | 199.85 |
| Result Component 13   | 17.92  | 59687.388 | 1.55         | 25.99         | -45.645       | 162773683        | 0.5                         | 0.60             | 31                      | 170.85 |
| Result Component 15   | 21.21  | 59731.785 | 0.70         | 11.70         | -1.249        | 129722977        | 0.4                         | 0.48             | 41                      | 225.58 |
| Result Component 17   | 19.13  | 59766.513 | 1.08         | 18.11         | 33.479        | 91587642         | 0.3                         | 0.34             | 38                      | 197.64 |
| Result Component 18   | 23.54  | 81372.238 | 1.92         | 23.54         | 21639.205     | 84227912         | 0.2                         | 0.31             | 50                      | 158.32 |
| Result Component 19   | 18.85  | 59816.211 | 1.65         | 27.63         | 83.178        | 73795145         | 0.2                         | 0.27             | 39                      | 170.82 |
| Result Component 20   | 19.45  | 59690.811 | 1.05         | 17.65         | -42.222       | 72092544         | 0.2                         | 0.27             | 37                      | 195.30 |
| Result Component 21   | 24.66  | 66290.558 | 1.28         | 19.26         | 6557.525      | 43412692         | 0.1                         | 0.16             | 41                      | 172.17 |

**Component Summary**

**Injection Details**

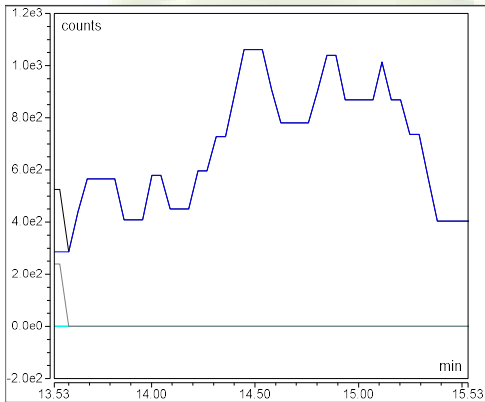
|                        |                                     |                             |   |
|------------------------|-------------------------------------|-----------------------------|---|
| <i>Injection Name:</i> | 20211011_AAV2 Mixed_45C_65-59B_Mod1 | <i>Injection Volume:</i>    | 8.45  |
| <i>Vial Number:</i>    | R:A9                                | <i>Instrument Method:</i>   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm |
| <i>Injection Type:</i> | Unknown                             | <i>Processing Method:</i>   | MS Quantitative_45C_65-59B                            |
| <i>Run Time (min):</i> | 44.99                               | <i>Injection Date/Time:</i> | 12/Oct/21 19:45                                       |

| Peak Name    | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|--------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3     | 20.403                | 2673211.186        | 78.52              | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 | 59973.780 | PASS                           |
| AAV2_VP1     | 25.250                | 107448.559         | 3.16               | 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362 | 81854.900 | PASS                           |
| AAV2_VP2     | 26.172                | 104060.303         | 3.06               | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 | 66487.900 | PASS                           |
| <b>84.73</b> |                       |                    |                    |   |           |                                |



**Component Name: AAV5\_VP3**

|                              |   |
|------------------------------|---|
| <i>Quantitation ions:</i>    | 1525.669,1565.784,1608.079,1451.304,1487.553,1416.767 |
| <i>Retention time (min):</i> | 14.53   |
| <i>Expected Mass:</i>        | 59462.370   |
| <i>Area (count*min):</i>     | -   |
| <i>Intensity (counts):</i>   | -   |
| <i>Amount:</i>               | -   |



**Component Name: AAV8 VP3**

Quantitation ions: 1574.800,1496.109,1534.442,1459.645,1273.445,1391.802

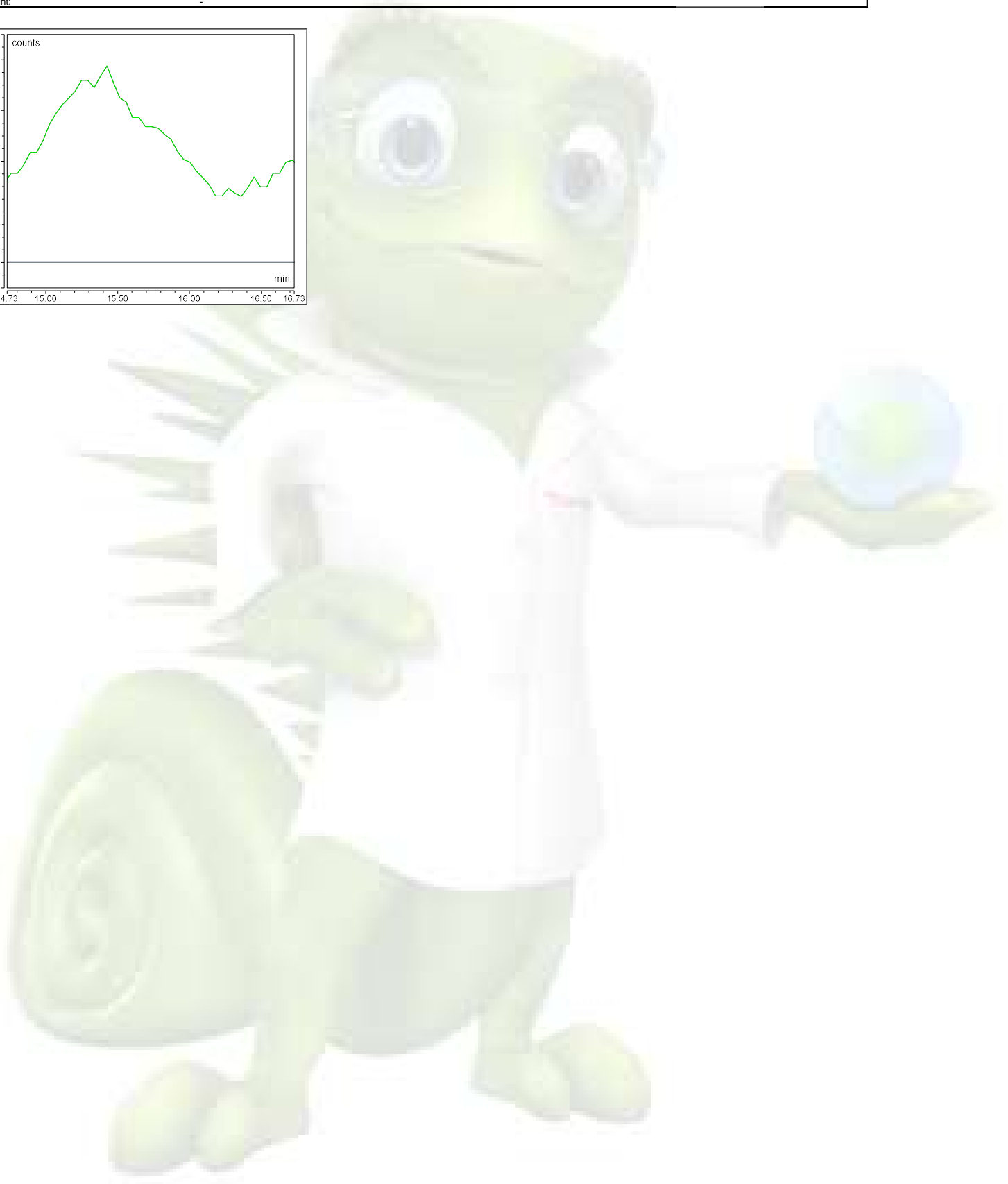
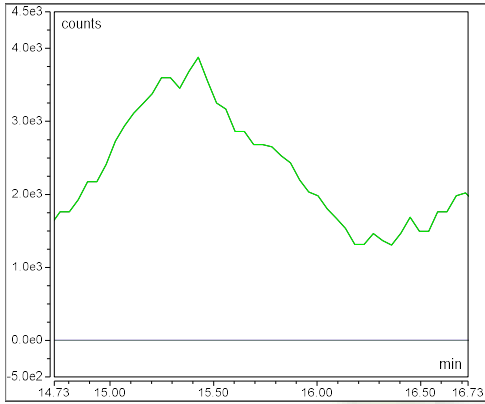
Retention time (min): 15.73

Expected Mass: 59804.680

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP3**

Quantitation ions: 1358.581,1390.150,1299.556,1328.415,1271.930,1457.907

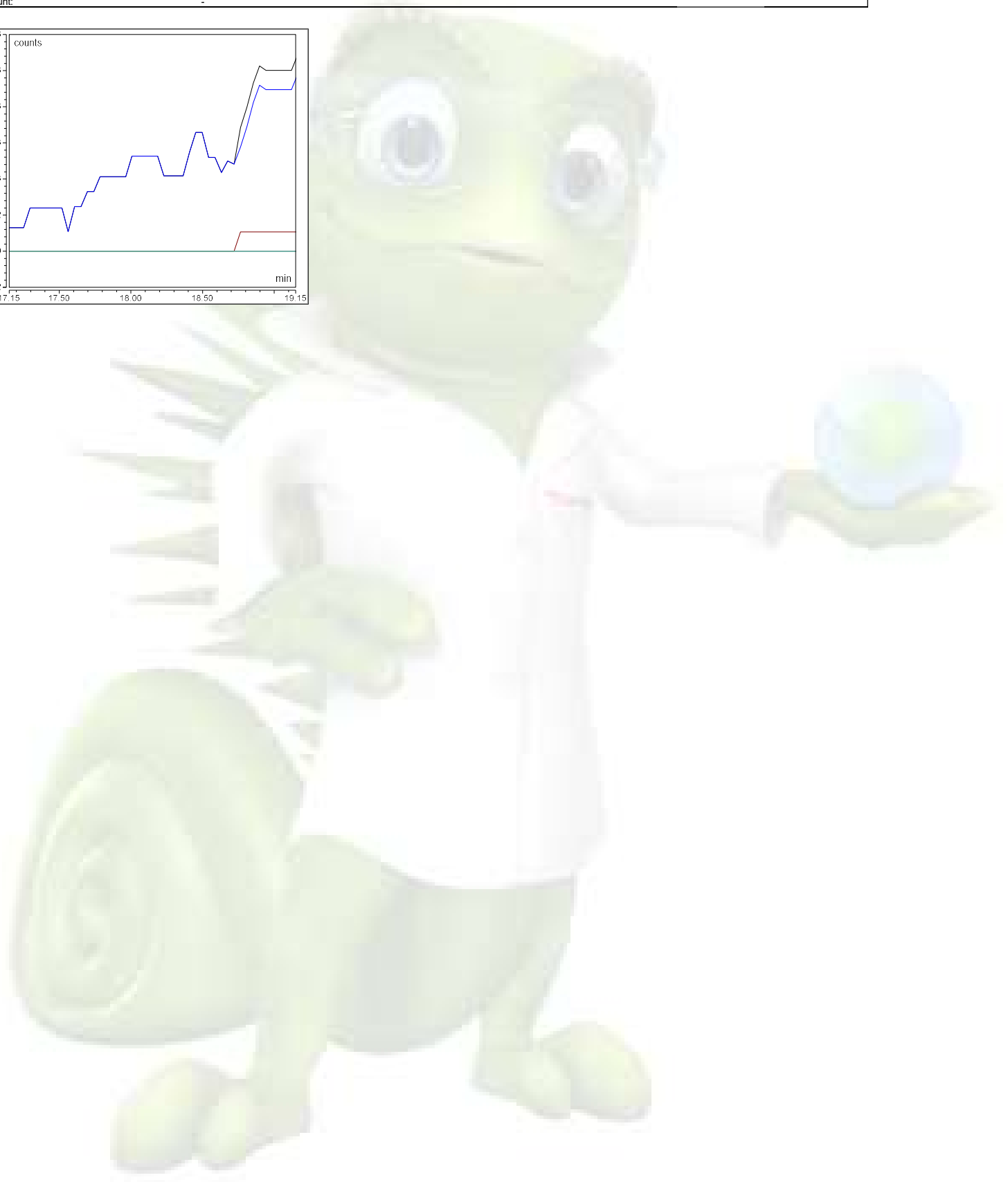
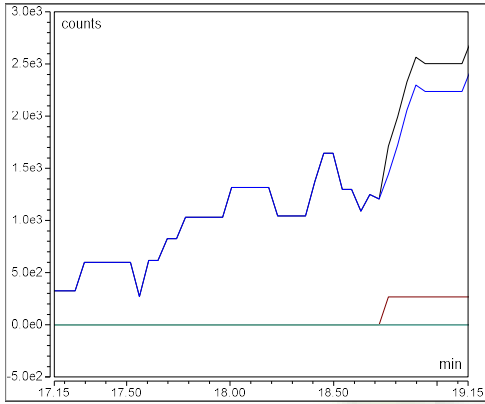
Retention time (min): 18.15

Expected Mass: 59732.620

Area (count\*min): -

Intensity (counts): -

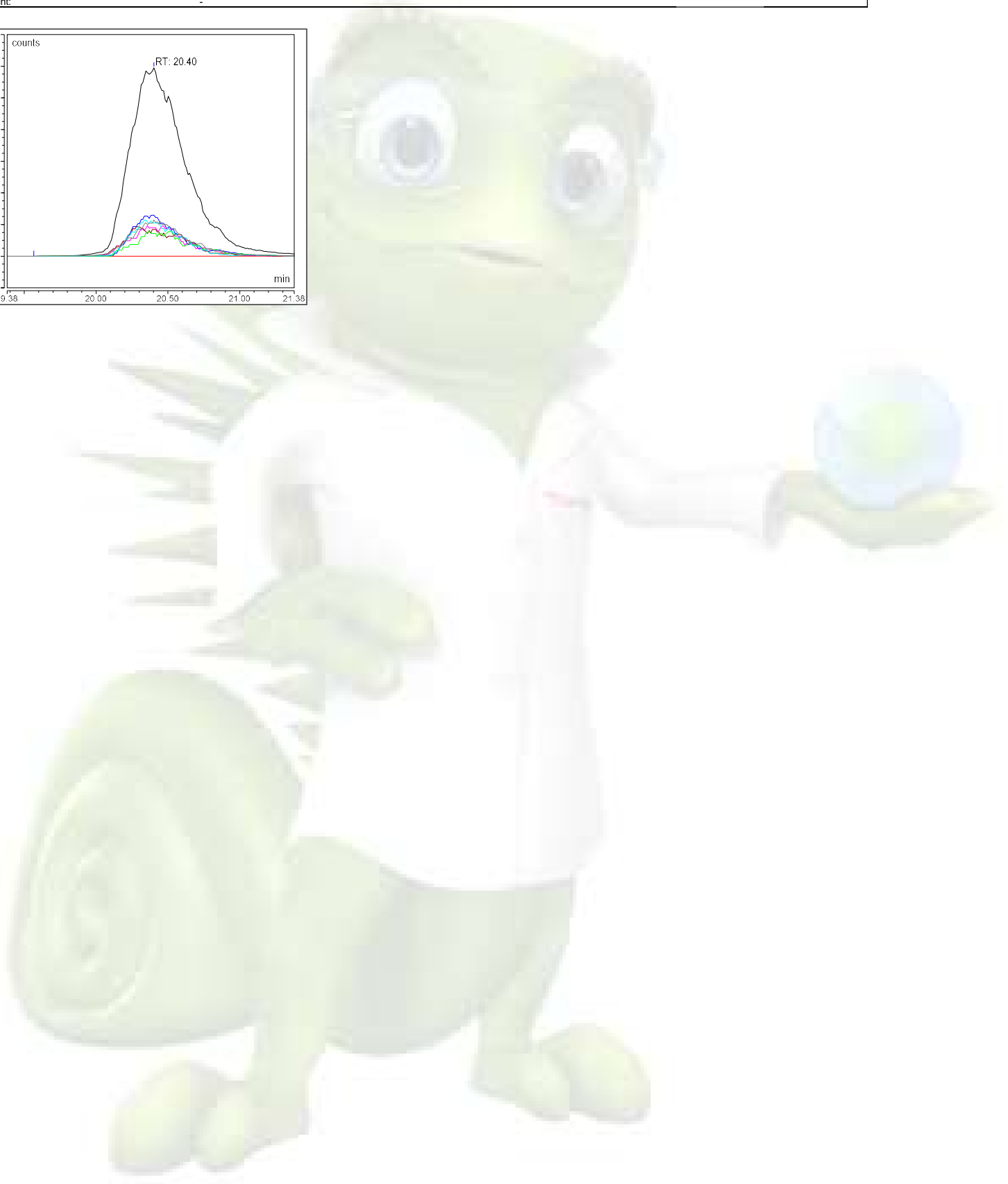
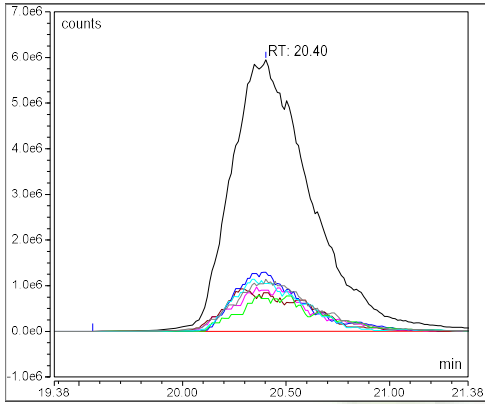
Amount: -





**Component Name: AAV2 VP3**

|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1428.955,1500.355,1463.789,1395.748,1364.055,1538.792 |
| Retention time (min): | 20.38   |
| Expected Mass:        | 59973.780   |
| Area (count*min):     | 2673211   |
| Intensity (counts):   | 5948923   |
| Amount:               | -   |



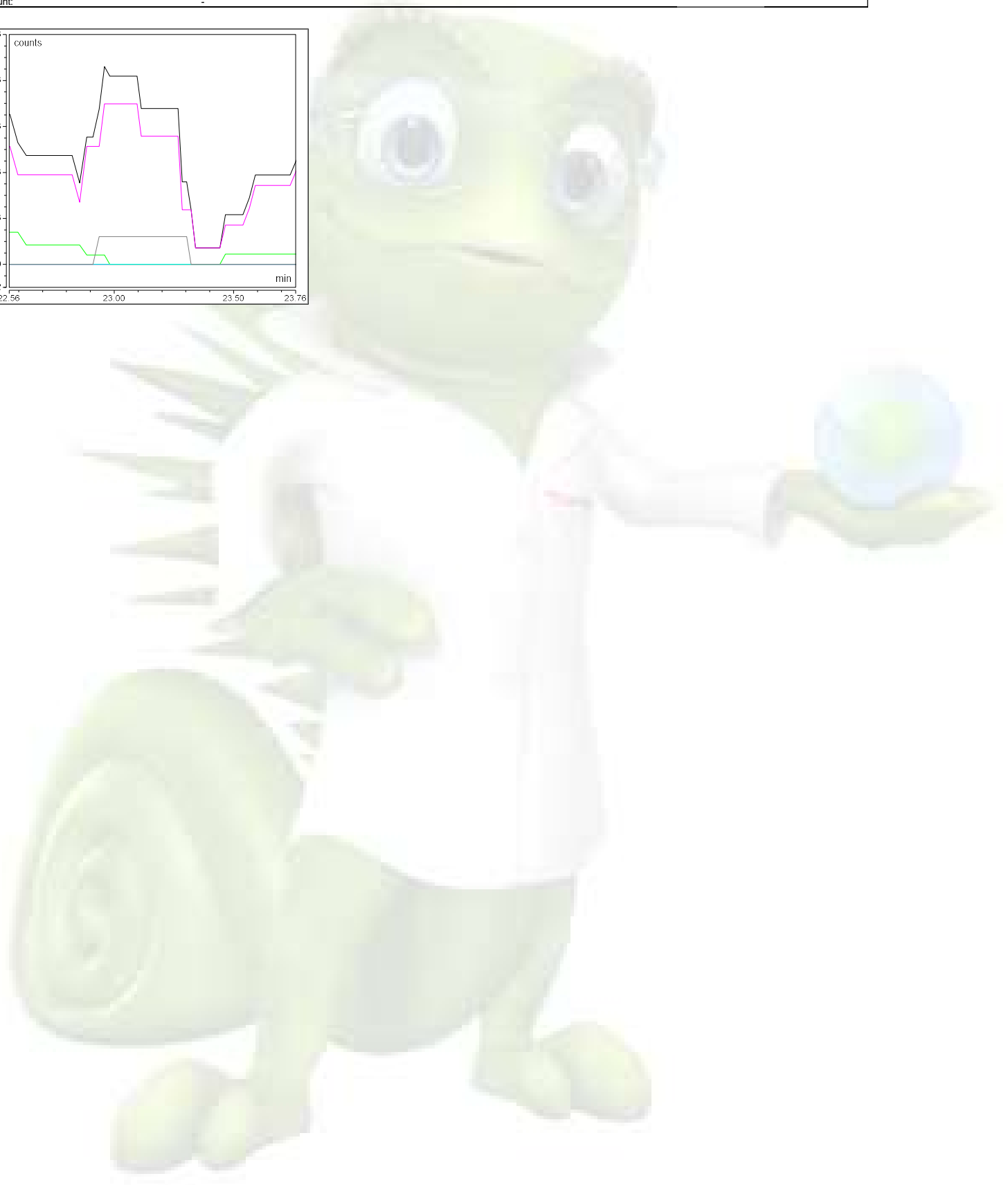
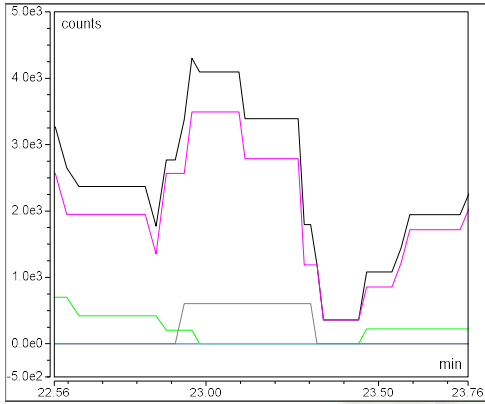
**Component Name: AAV9 VP1**

Quantitation ions: 1378.820,1333.651,1355.868,1402.584,1534.808,1452.636

Retention time (min): 23.16

Expected Mass: 81290.340

Area (count\*min): -  
Intensity (counts): -  
Amount: -



**Component Name: AAV8 VP1**

Quantitation ions: 1409.080,1459.382,1385.209,1513.388,1184.605,1339.828

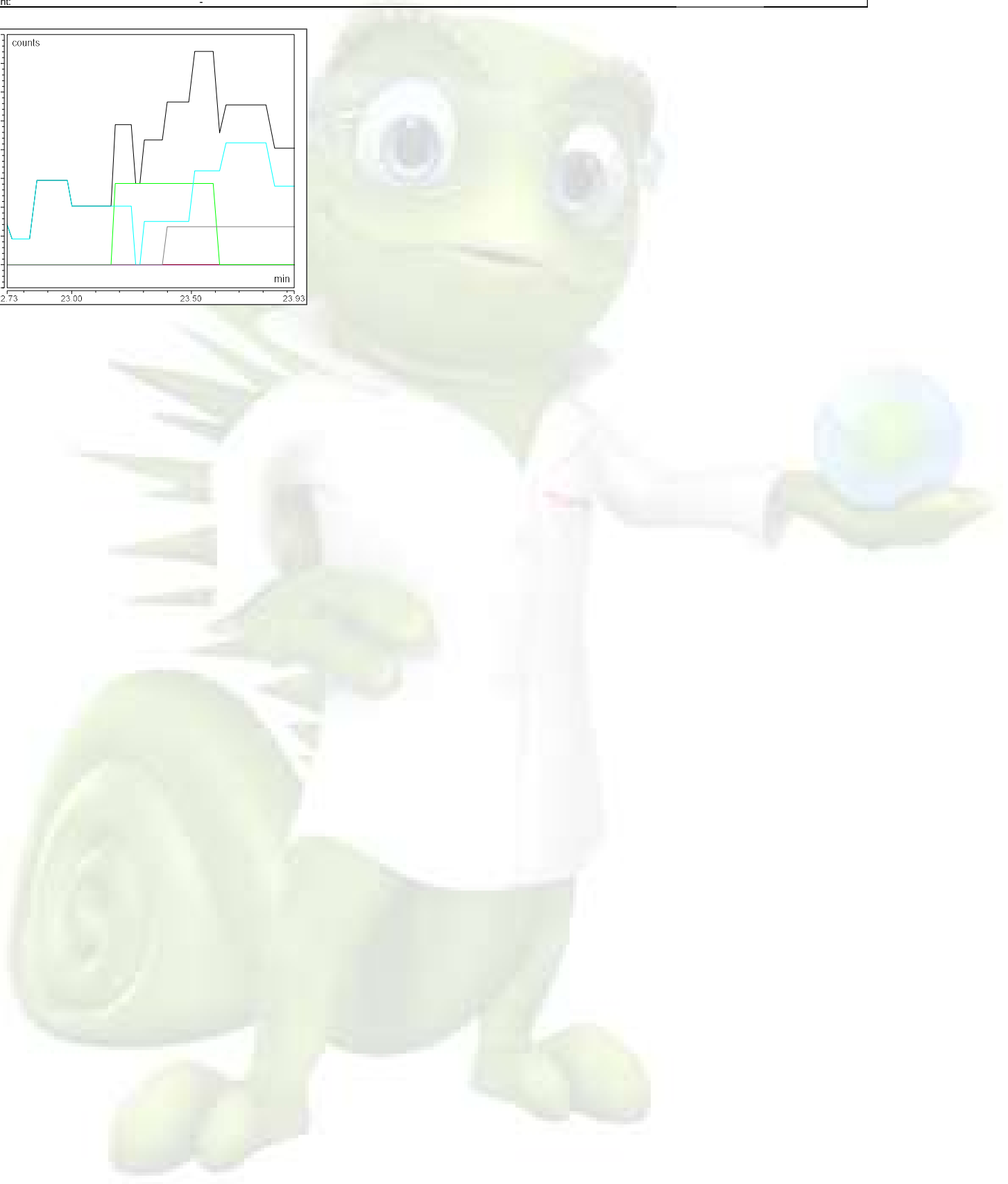
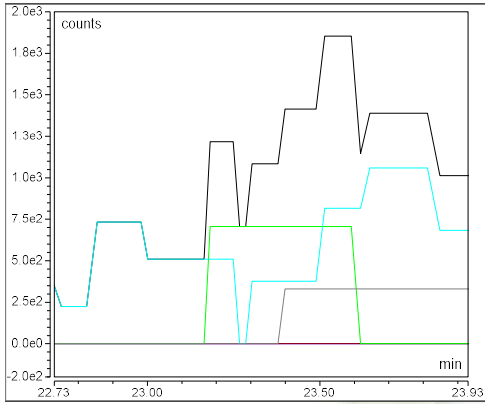
Retention time (min): 23.33

Expected Mass: 81666.710

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP1**

Quantitation ions: 1362.628,1410.415,1435.577,1317.996,1339.949,1488.697

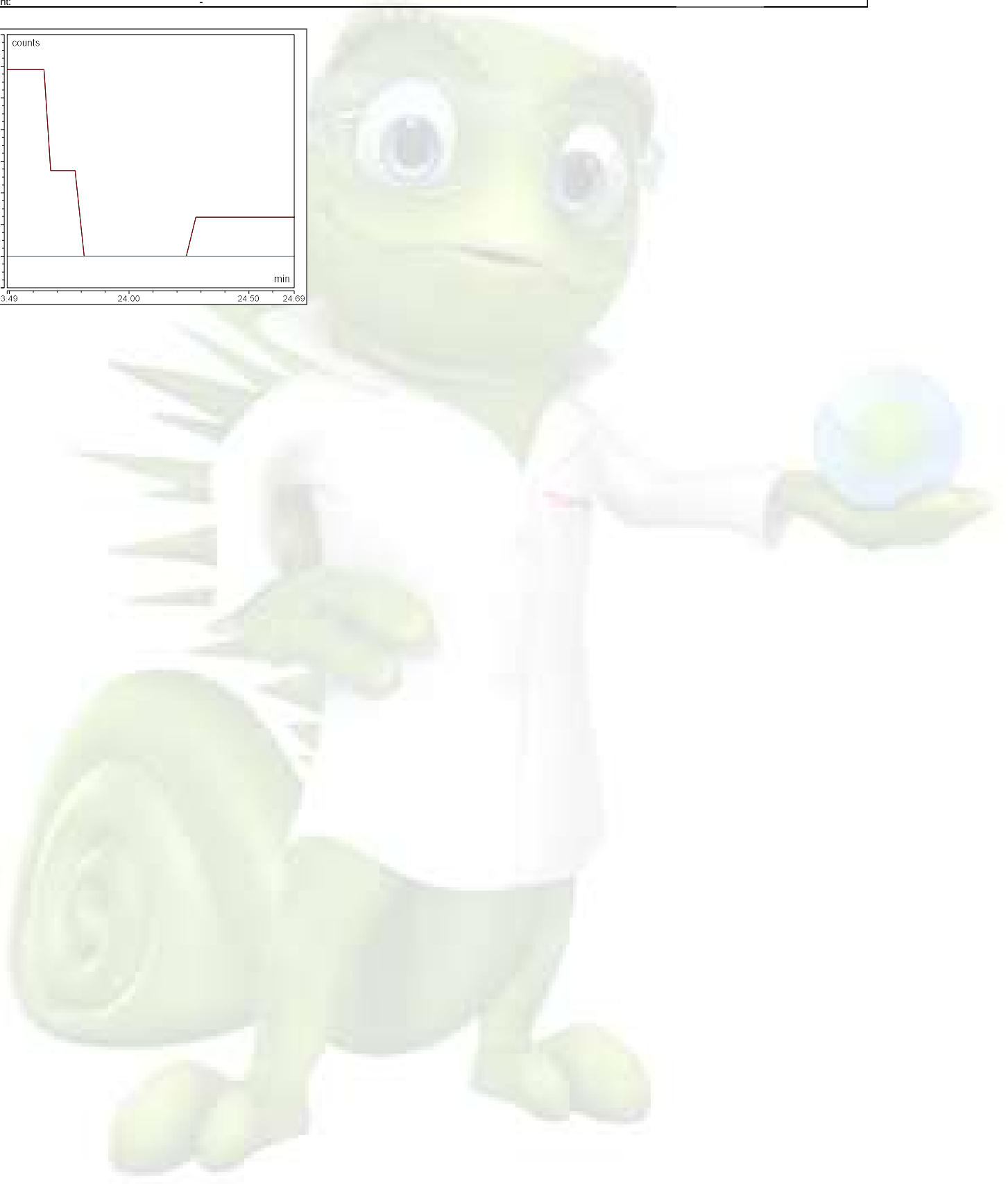
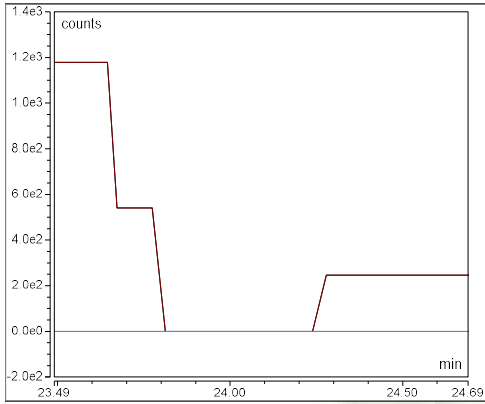
Retention time (min): 24.09

Expected Mass: 80335.080

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV9 VP2**

Quantitation ions: 1409.730,1162.589,1472.334,1274.287,1380.371,1352.231

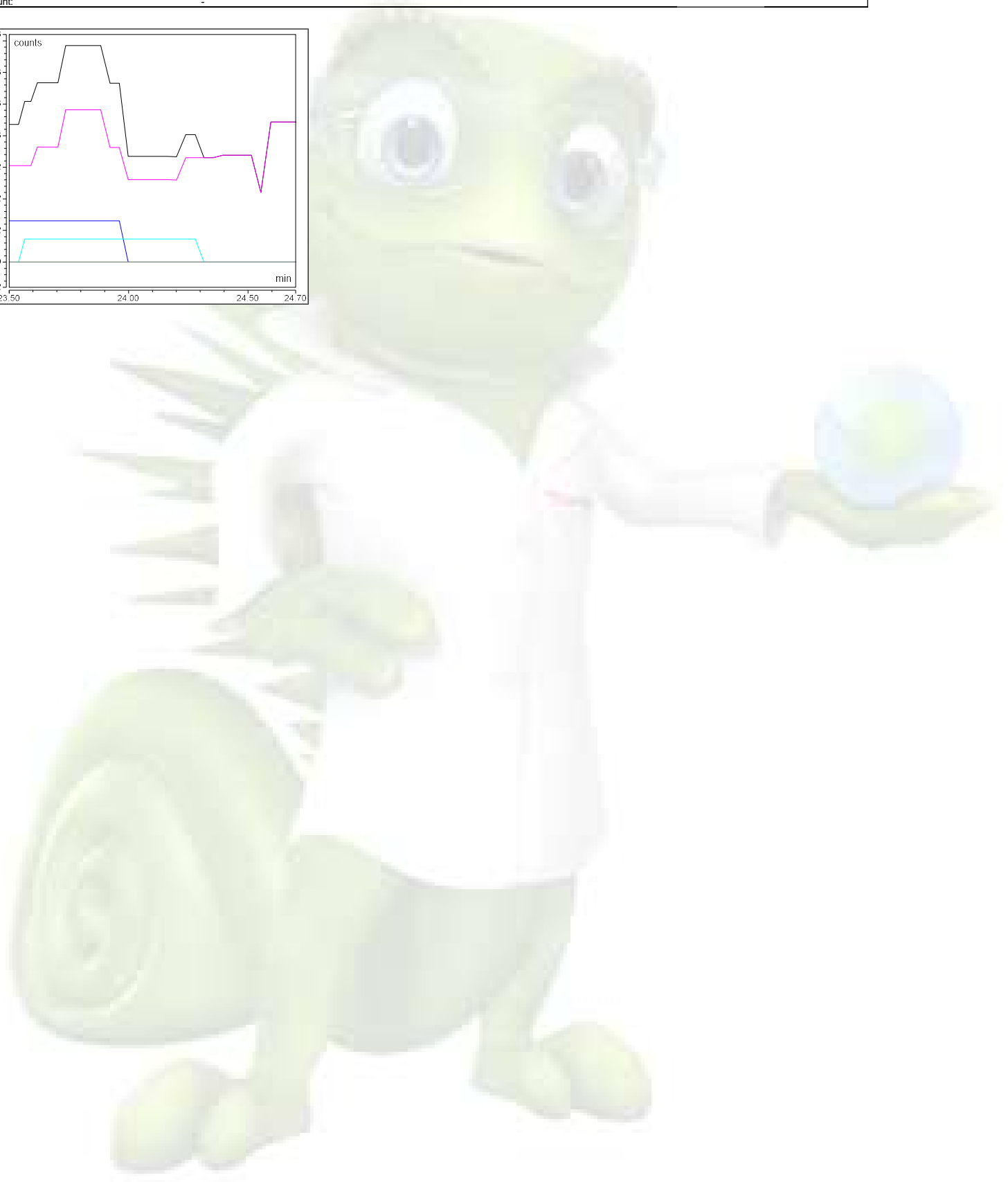
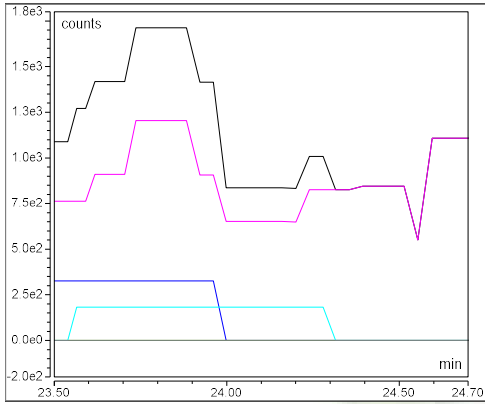
Retention time (min): 24.10

Expected Mass: 66209.760

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV8 VP2**

Quantitation ions: 1386.809,1358.524,1331.393,1305.308,1416.294,1623.397

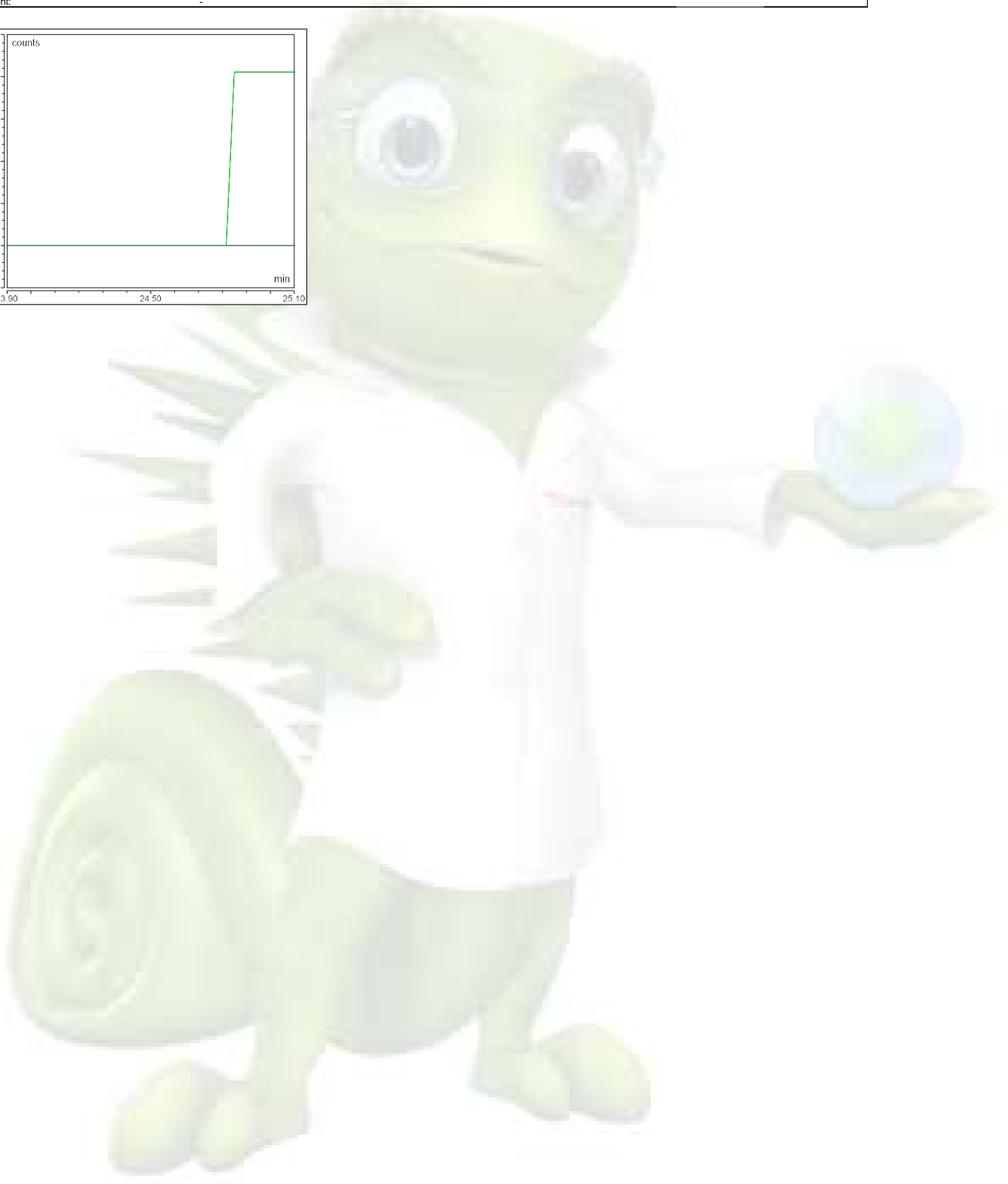
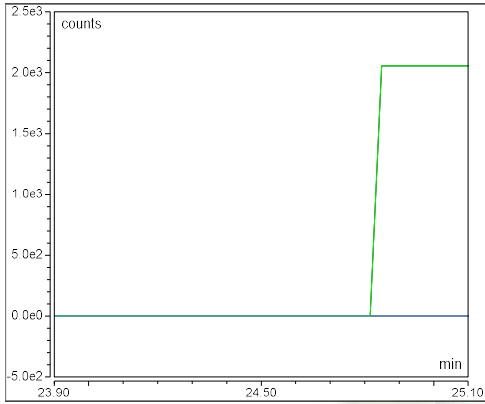
Retention time (min): 24.50

Expected Mass: 66518.100

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV5 VP2**

Quantitation ions: 1389.997,1519.197,1484.692,1420.196,1555.348,1451.727

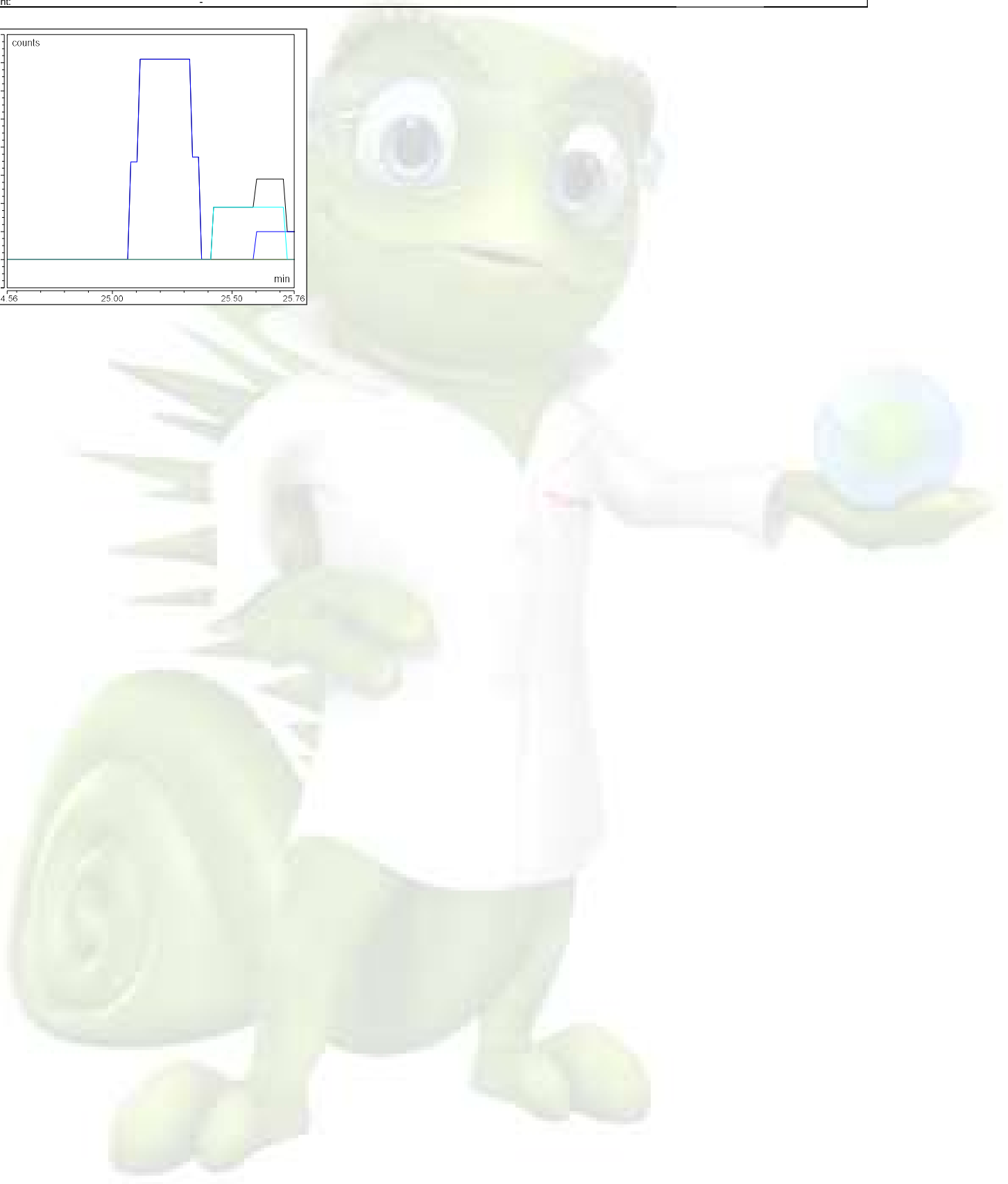
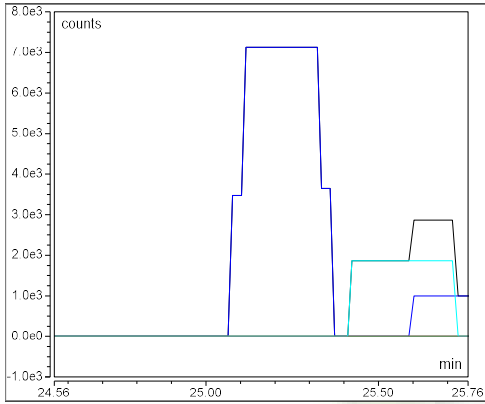
Retention time (min): 25.16

Expected Mass: 65282.690

Area (count\*min): -

Intensity (counts): -

Amount: -



**Component Name: AAV2 VP1**

Quantitation ions: 1388.434,1545.445,1489.336,1342.917,1365.283,1412.362

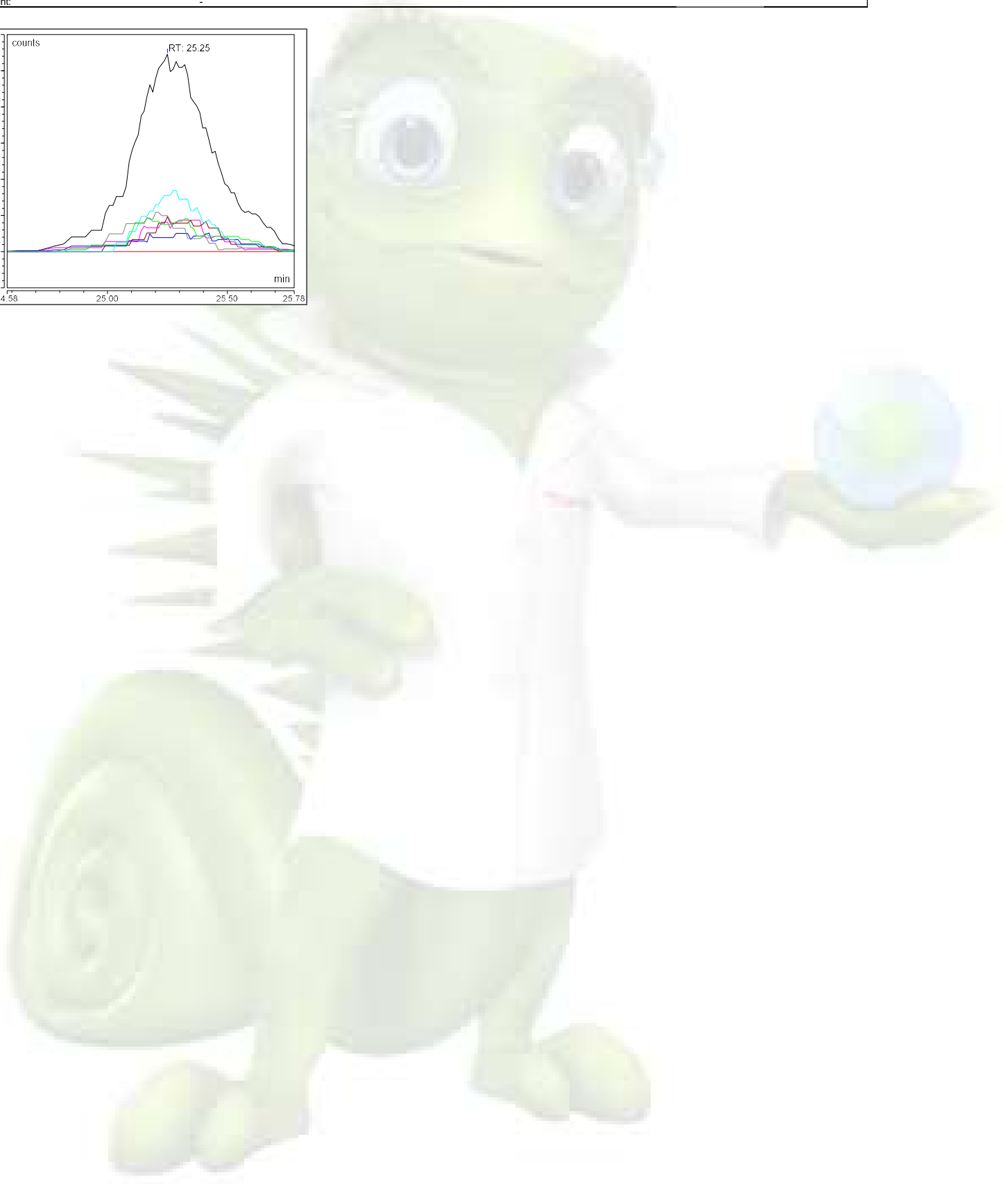
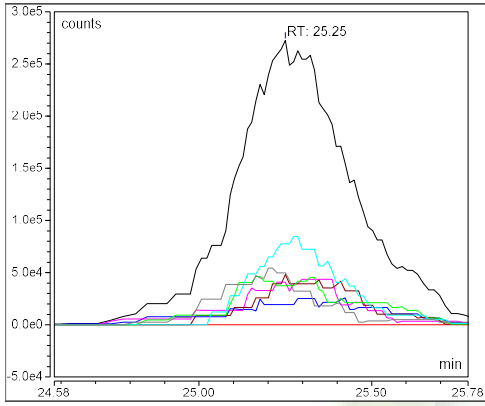
Retention time (min): 25.18

Expected Mass: 81854.900

Area (count\*min): 107449

Intensity (counts): 272716

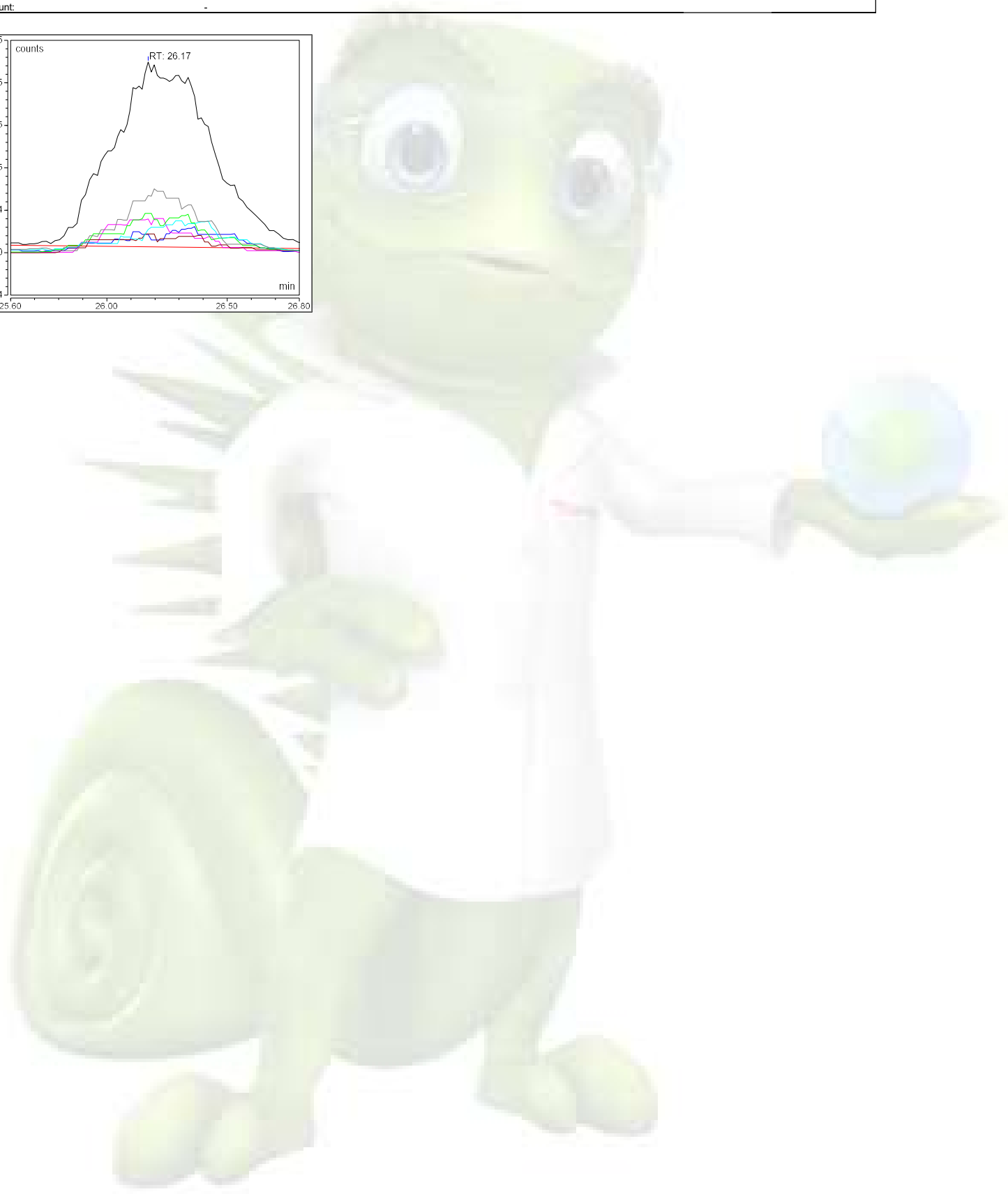
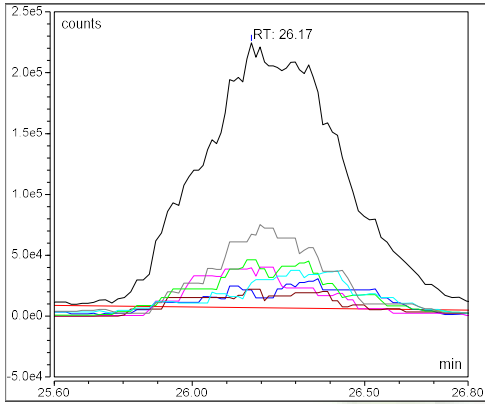
Amount: -





**Component Name: AAV2 VP2**

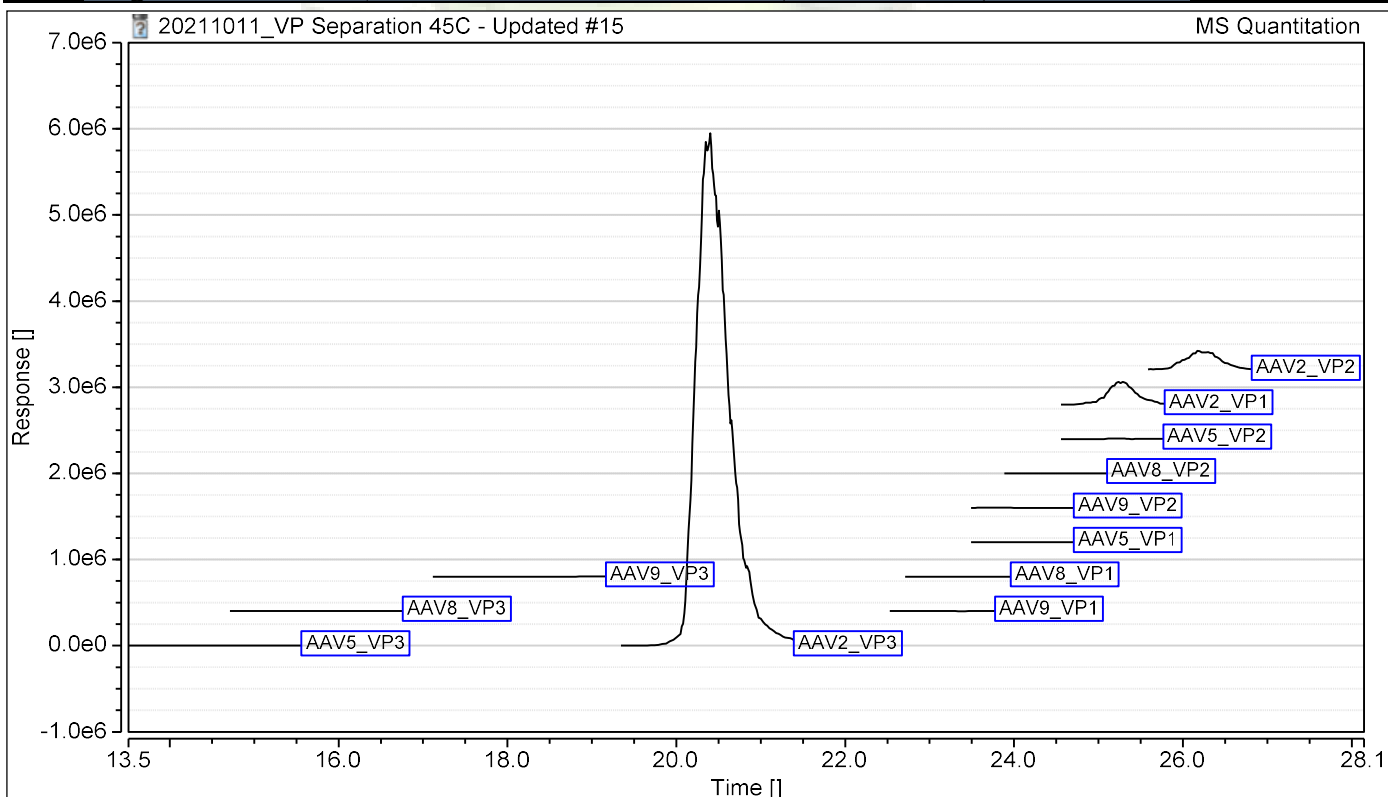
|                       |   |
|-----------------------|---|
| Quantitation ions:    | 1415.636,1446.415,1255.530,1478.506,1279.609,1304.686 |
| Retention time (min): | 26.20   |
| Expected Mass:        | 66487.900   |
| Area (count*min):     | 104060  |
| Intensity (counts):   | 217576  |
| Amount:               | -   |



## Chromatogram and SST Results

| Injection Details    |  |                         |
|----------------------|--|-------------------------|
| Injection Name:      | 20211011_AAV2 Mixed_45C_65-59B_Mod1_1ug_3                                  | Run Time (min): 44.99   |
| Vial Number:         | R:A9   | Injection Volume: 8.45  |
| Injection Type:      | Unknown  |                         |
| Calibration Level:   |  |                         |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_CT45C_65-59 | S                       |
| Processing Method:   | MS Quantitative_45C_65-59B   | Dilution Factor: 1.0000 |
| Injection Date/Time: | 12/Oct/21 19:45  | Sample Weight: 1.0000   |

### Chromatogram



### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

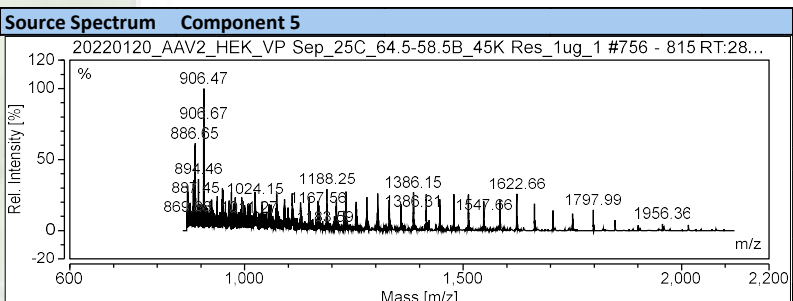
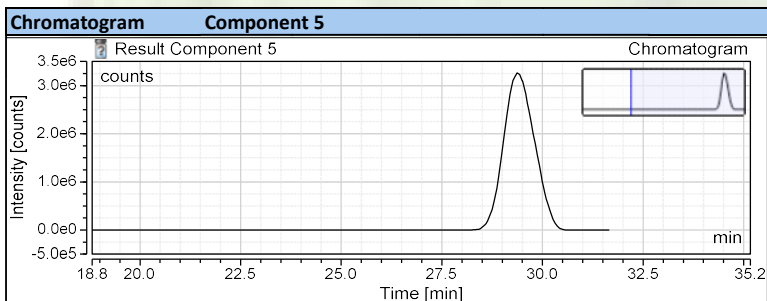
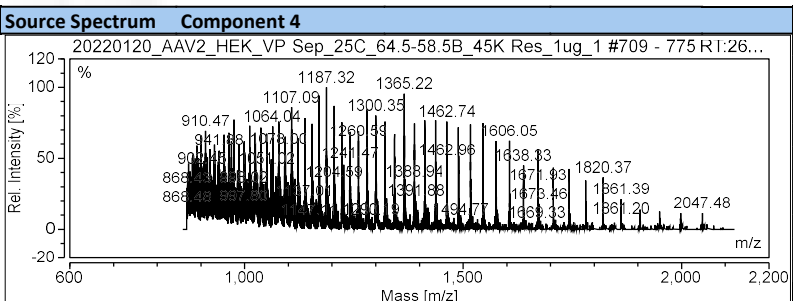
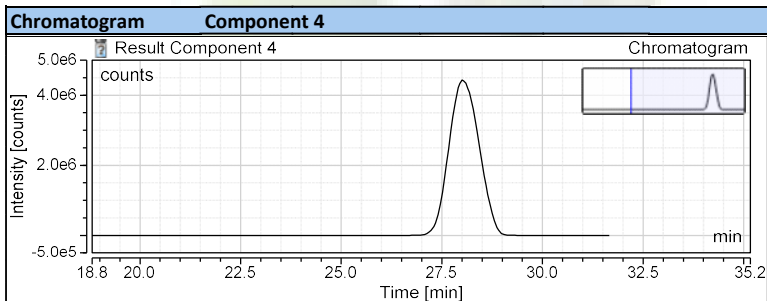
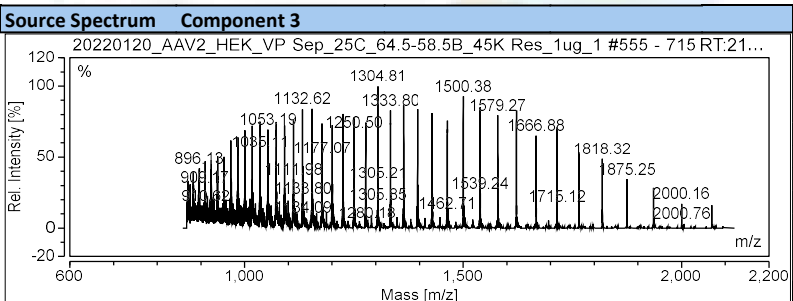
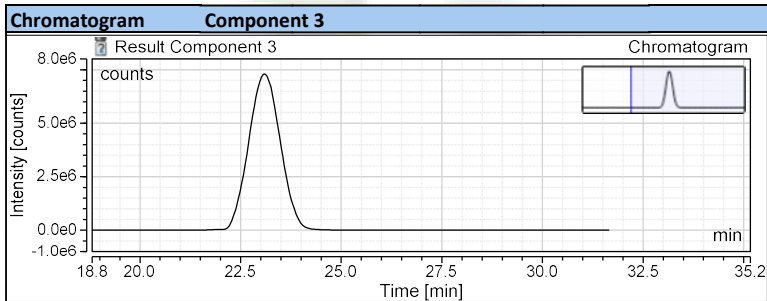
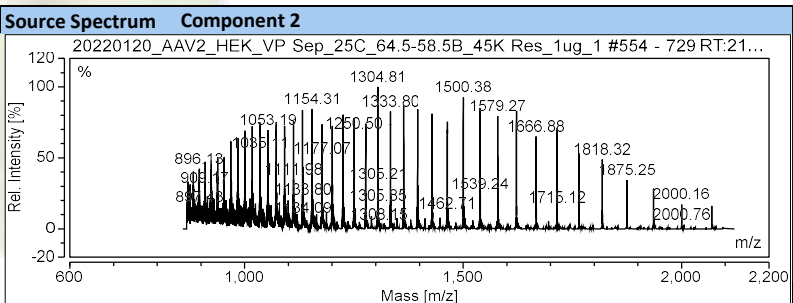
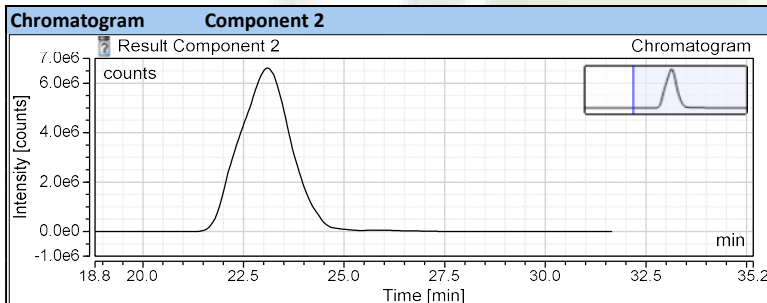
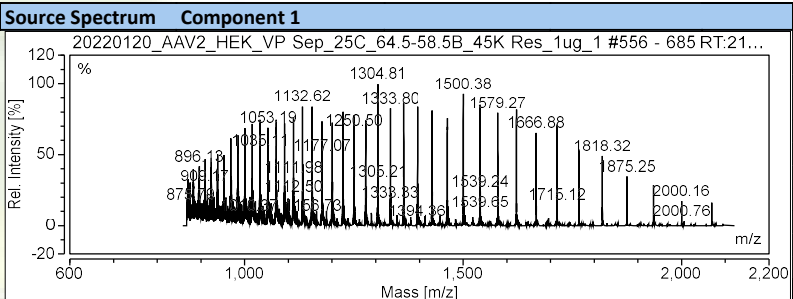
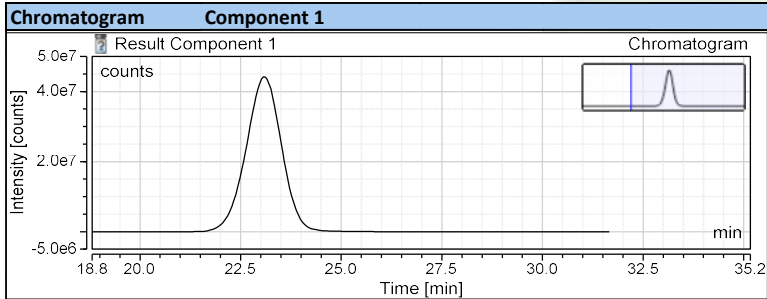
**Supplementary Report 2:**

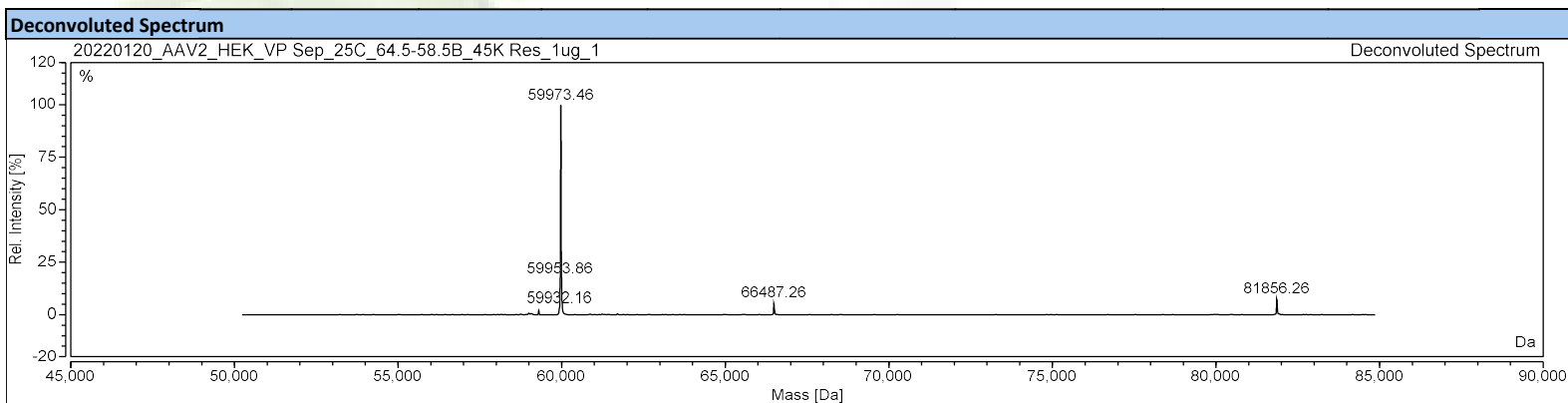
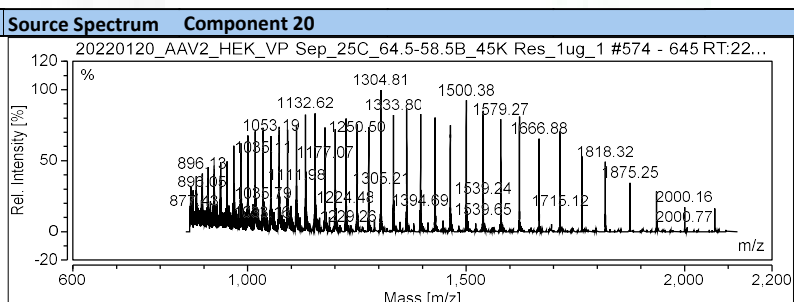
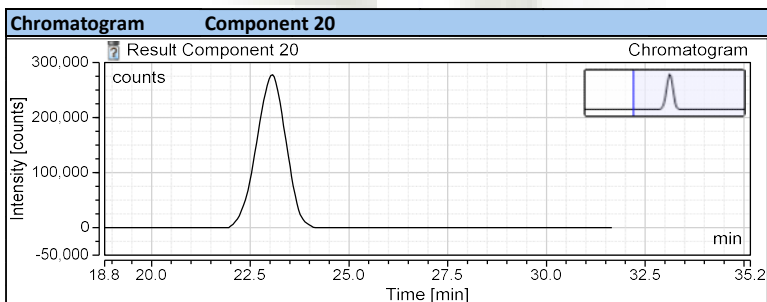
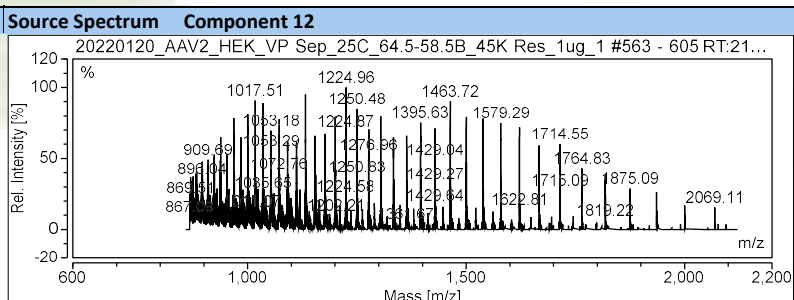
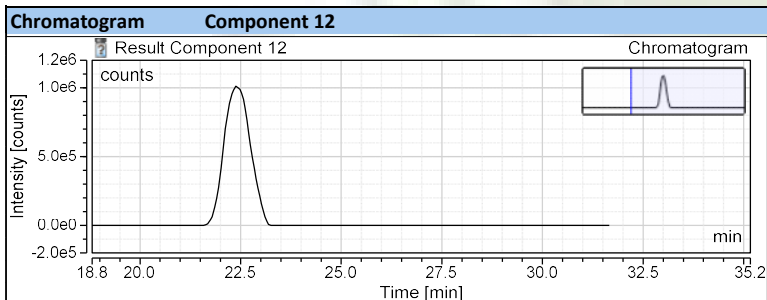
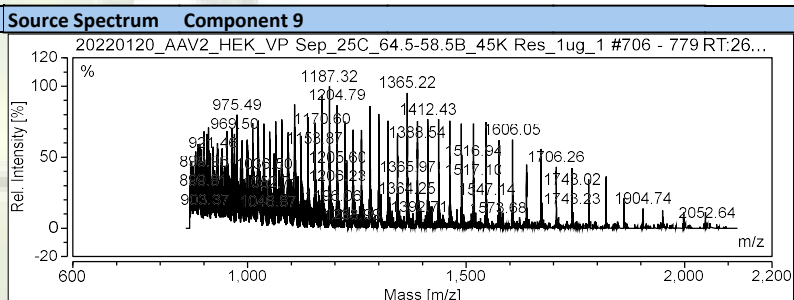
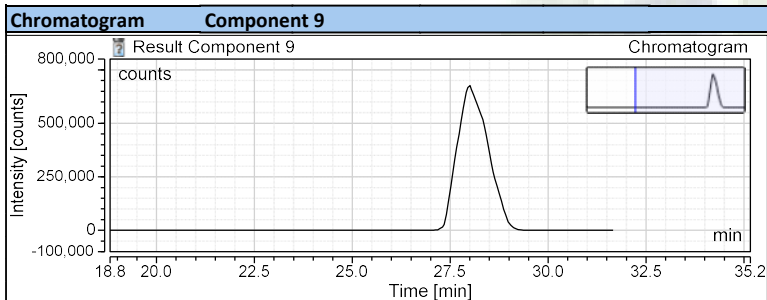
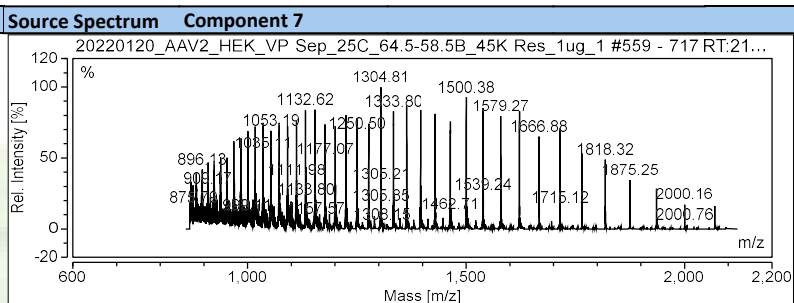
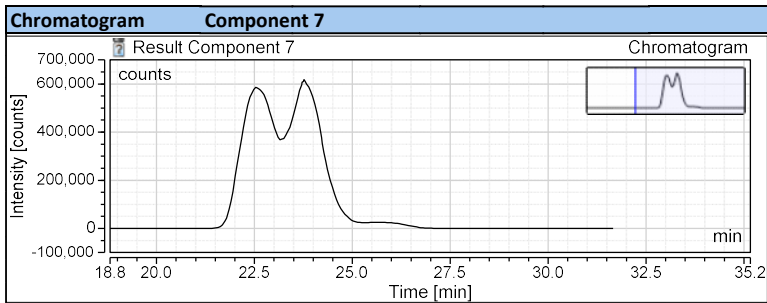
Report of PTM monitoring method applied to AAV2 generated in Chromeleon

### Protein Deconvolution Summary

#### Injection Details

|                 |   |                      |  |
|-----------------|---|----------------------|--|
| Injection Name: | 20220120_AAV2_HEK_VP Sep_25C_64.5-58.5B_45K Res_1ug_1 | Injection Volume:    | 8.56   |
| Vial Number:    | R:A1  | Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide H      |
| Injection Type: | Unknown   | Processing Method:   | AAV2 MS Quantitative In-depth Characterization |
| Run Time (min): | 45.00   | Injection Date/Time: | 21/Jan/22 07:32                                |





**Deconvolution Results**

| Result Component   | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
|--------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component 1 | 23.08  | 59973.436 | 0.84         | 13.99         | 0.000         | 1959279259       | 46.2                        | 100.00           | 41                      | 242.04 |
| Result Component 2 | 23.11  | 59991.819 | 1.81         | 30.24         | 18.383        | 430877006        | 10.2                        | 21.99            | 41                      | 161.22 |
| Result Component 3 | 23.08  | 59953.840 | 1.53         | 25.59         | -19.596       | 302960887        | 7.1                         | 15.46            | 34                      | 154.28 |
| Result Component 4 | 28.00  | 81855.271 | 1.70         | 20.79         | 21881.835     | 149335479        | 3.5                         | 7.62             | 55                      | 180.65 |
| Result Component 5 | 29.37  | 66487.295 | 1.24         | 18.66         | 6513.860      | 115071618        | 2.7                         | 5.87             | 42                      | 195.77 |
| Result Component 6 | 23.08  | 60018.234 | 1.74         | 28.98         | 44.799        | 61338748         | 1.4                         | 3.13             | 25                      | 68.82  |
| Result Component 7 | 23.76  | 60006.121 | 1.86         | 30.94         | 32.686        | 59932437         | 1.4                         | 3.06             | 39                      | 79.35  |
| Result Component 8 | 29.37  | 66504.638 | 1.95         | 29.34         | 6531.202      | 43476745         | 1.0                         | 2.22             | 41                      | 102.72 |
| Result Component 9 | 27.98  | 81872.829 | 2.11         | 25.73         | 21899.393     | 38271649         | 0.9                         | 1.95             | 44                      | 90.47  |

|                      |       |           |      |       |           |          |     |      |    |        |
|----------------------|-------|-----------|------|-------|-----------|----------|-----|------|----|--------|
| Result Component 11  | 22.91 | 59299.871 | 1.48 | 25.01 | -673.565  | 28850826 | 0.7 | 1.47 | 30 | 69.26  |
| Result Component 12  | 22.40 | 59954.253 | 1.72 | 28.73 | -19.183   | 27440115 | 0.6 | 1.40 | 33 | 139.46 |
| Result Component 19  | 27.67 | 81874.453 | 2.73 | 33.39 | 21901.017 | 10900834 | 0.3 | 0.56 | 50 | 109.08 |
| Result Component 77  | 24.00 | 60020.804 | 1.54 | 25.71 | 47.368    | 3356483  | 0.1 | 0.17 | 33 | 78.05  |
| Result Component 152 | 29.15 | 66467.903 | 1.85 | 27.80 | 6494.467  | 1942465  | 0.0 | 0.10 | 19 | 65.67  |



## Component Summary

### Injection Details

|                 |                                     |                      |                                       |
|-----------------|-------------------------------------|----------------------|---------------------------------------|
| Injection Name: | 20220120_AAV2_HEK_VP_Sep_25C_64.5-5 | Injection Volume:    | 8.56                                  |
| Vial Number:    | R:A1                                | Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein     |
| Injection Type: | Unknown                             | Processing Method:   | AAV2 MS Quantitative In-depth Charact |
| Run Time (min): | 45.00                               | Injection Date/Time: | 21/Jan/22 07:32                       |

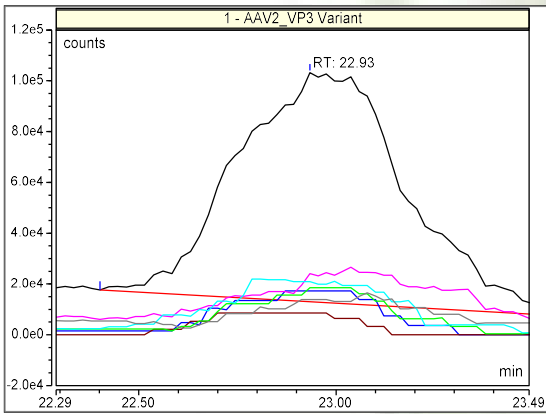
| Peak Name                  | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|----------------------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3 Variant           | 22.934                | 44616.104          | 6.75               | 1447.374,957.488,1380.090,1163.736,1099.120,973.145   | 59301.030 | PASS                           |
| AAV2_VP3[2xOxidation (MW)] | 22.934                | 89046.902          | 13.47              | 1133.193,1092.027,1112.217,1072.520,1539.665,1464.606 | 60005.780 | PASS                           |
| AAV2_VP3[1xSuccinimide D]  | 22.955                | 133805.141         | 20.24              | 952.674,923.374,968.020,937.789,1017.182,1000.250     | 59955.770 | PASS                           |
| AAV2_VP3[1xOxidation (MW)] | 23.079                | 125455.836         | 18.97              | 1277.428,1396.179,1464.229,1250.823,1305.183,1225.324 | 59989.780 | PASS                           |
| AAV2_VP3                   | 23.100                | 122079.437         | 18.46              | 1364.311,1500.570,1333.966,1395.925,1463.892,1225.374 | 59973.780 | PASS                           |
| AAV2_VP1[1xOxidation (MW)] | 28.073                | 34246.303          | 5.18               | 1343.191,1154.150,1388.700,1187.572,1412.610,1170.616 | 81870.890 | PASS                           |
| AAV2_VP1                   | 28.117                | 52640.275          | 7.96               | 1342.896,1388.388,1365.271,1300.301,1187.321,1279.998 | 81854.900 | PASS                           |
| AAV2_VP2[1xOxidation (MW)] | 29.331                | 24845.730          | 3.76               | 1446.760,1305.022,1167.738,1358.263,1147.628,1331.107 | 66503.900 | PASS                           |
| AAV2_VP2                   | 29.400                | 34429.457          | 5.21               | 1446.392,1415.628,1304.683,1478.503,1209.871,1386.167 | 66487.900 | PASS                           |
| <b>100.00</b>              |                       |                    |                    |   |           |                                |

### Component Name: AAV2\_VP3 Variant

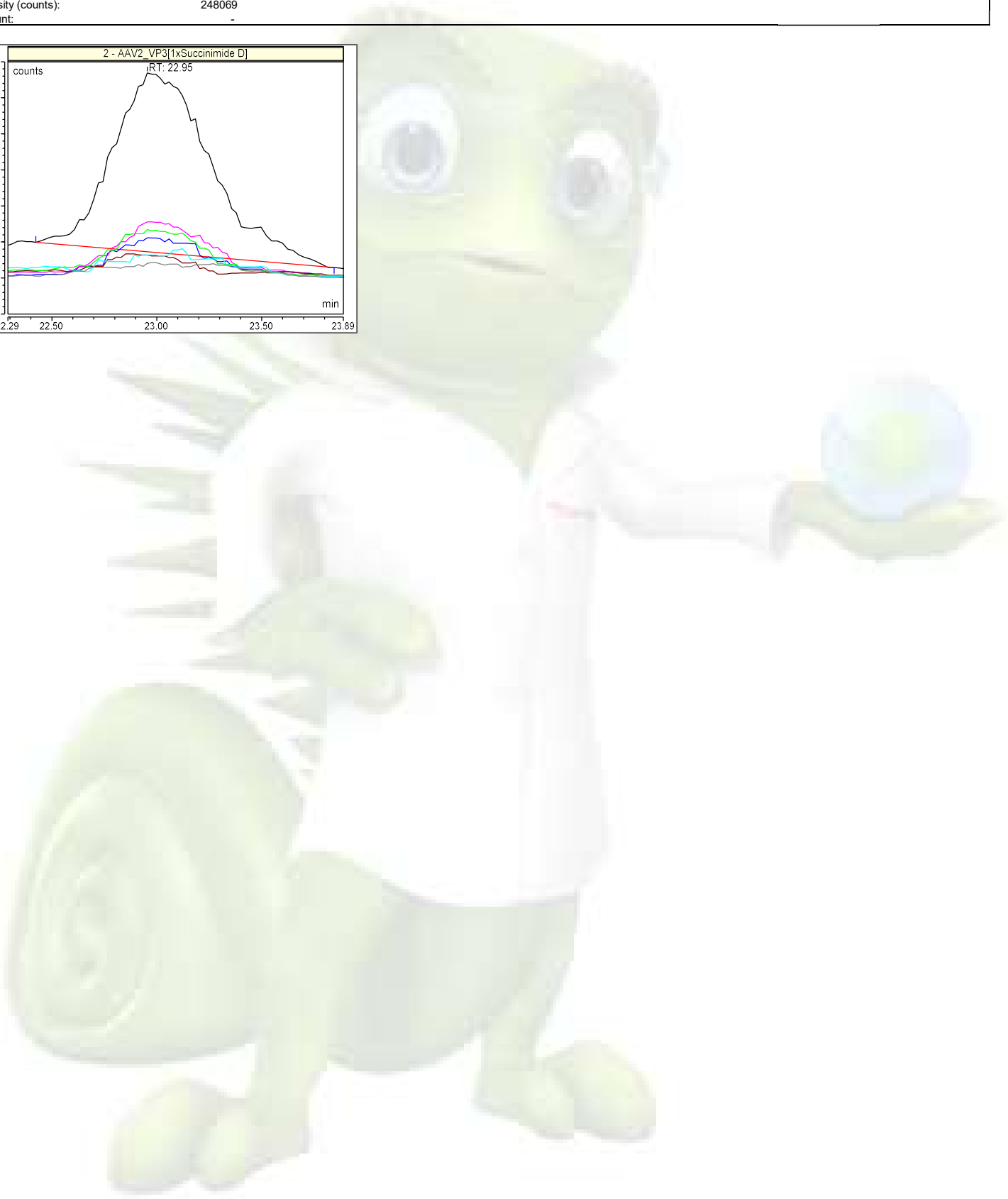
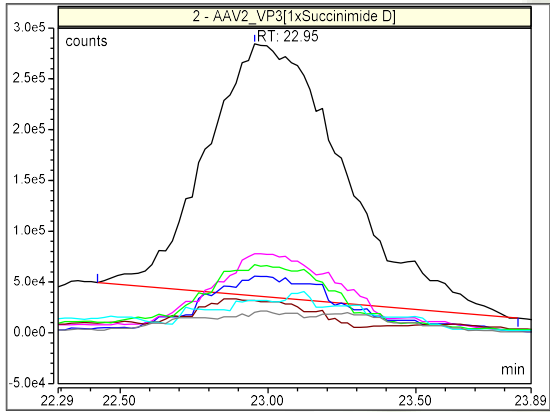
Quantitation ions: 1447.374,957.488,1380.090,1163.736,1099.120,973.145

Retention time (min): 22.89  
Expected Mass: 59301.030

Area (count\*min): 44616  
Intensity (counts): 90177  
Amount: -

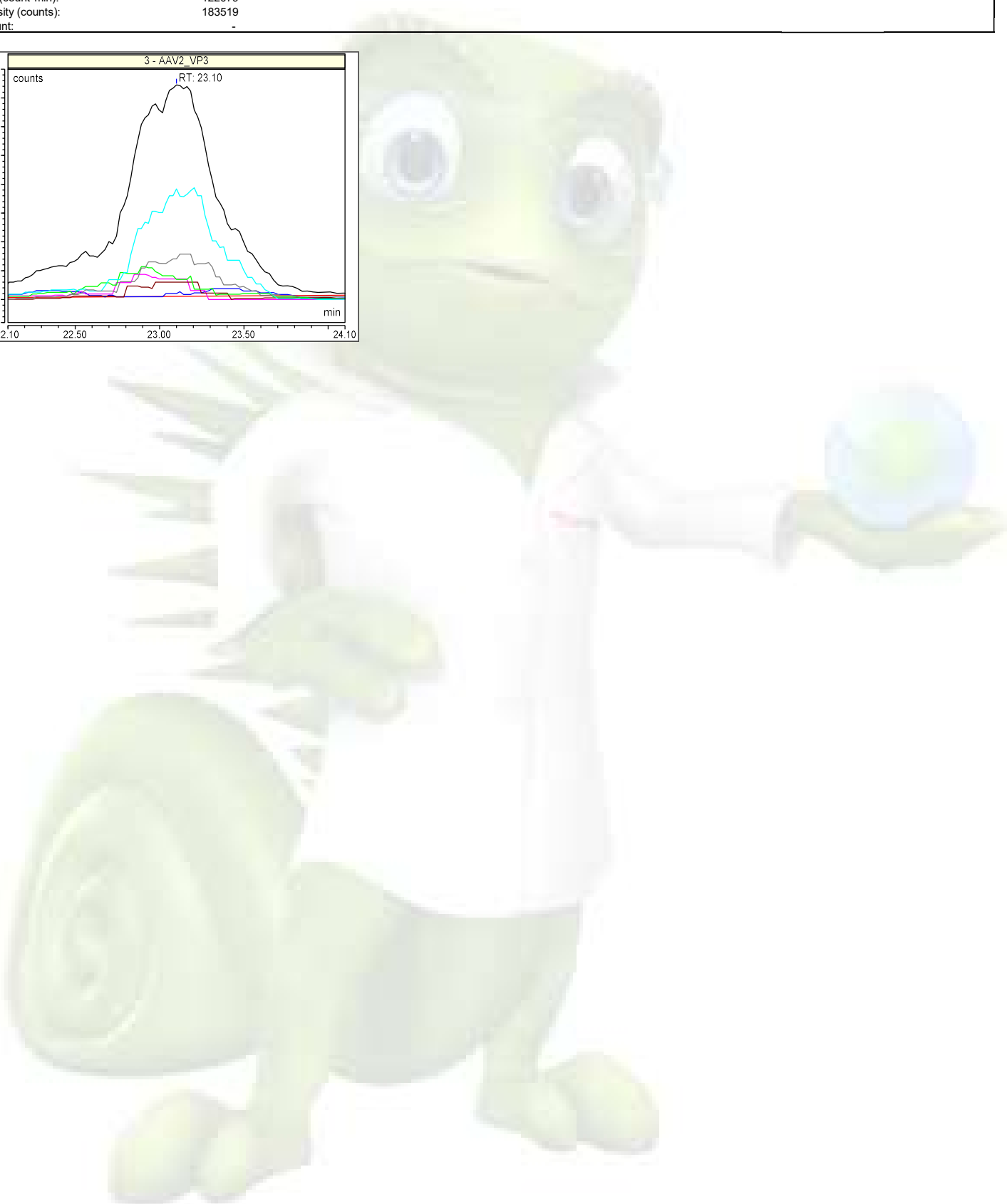
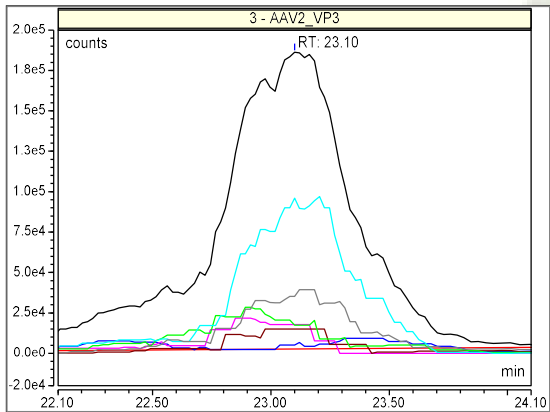


|  |   |
|--|---|
| <b>Component Name: AAV2 VP3[1xSuccinimide D]</b> |   |
| Quantitation ions:                               | 952.674,923.374,968.020,937.789,1017.182,1000.250 |
| Retention time (min):                            | 23.09   |
| Expected Mass:                                   | 59955.770   |
| Area (count*min):                                | 133805  |
| Intensity (counts):                              | 248069  |
| Amount:  | -   |





|                                 |   |
|---------------------------------|---|
| <b>Component Name: AAV2 VP3</b> |   |
| Quantitation ions:              | 1364.311,1500.570,1333.966,1395.925,1463.892,1225.374 |
| Retention time (min):           | 23.10   |
| Expected Mass:                  | 59973.780   |
| Area (count*min):               | 122079  |
| Intensity (counts):             | 183519  |
| Amount:                         | -   |



**Component Name: AAV2 VP3[2xOxidation (MW)]**

Quantitation ions: 1133.193,1092.027,1112.217,1072.520,1539.665,1464.606

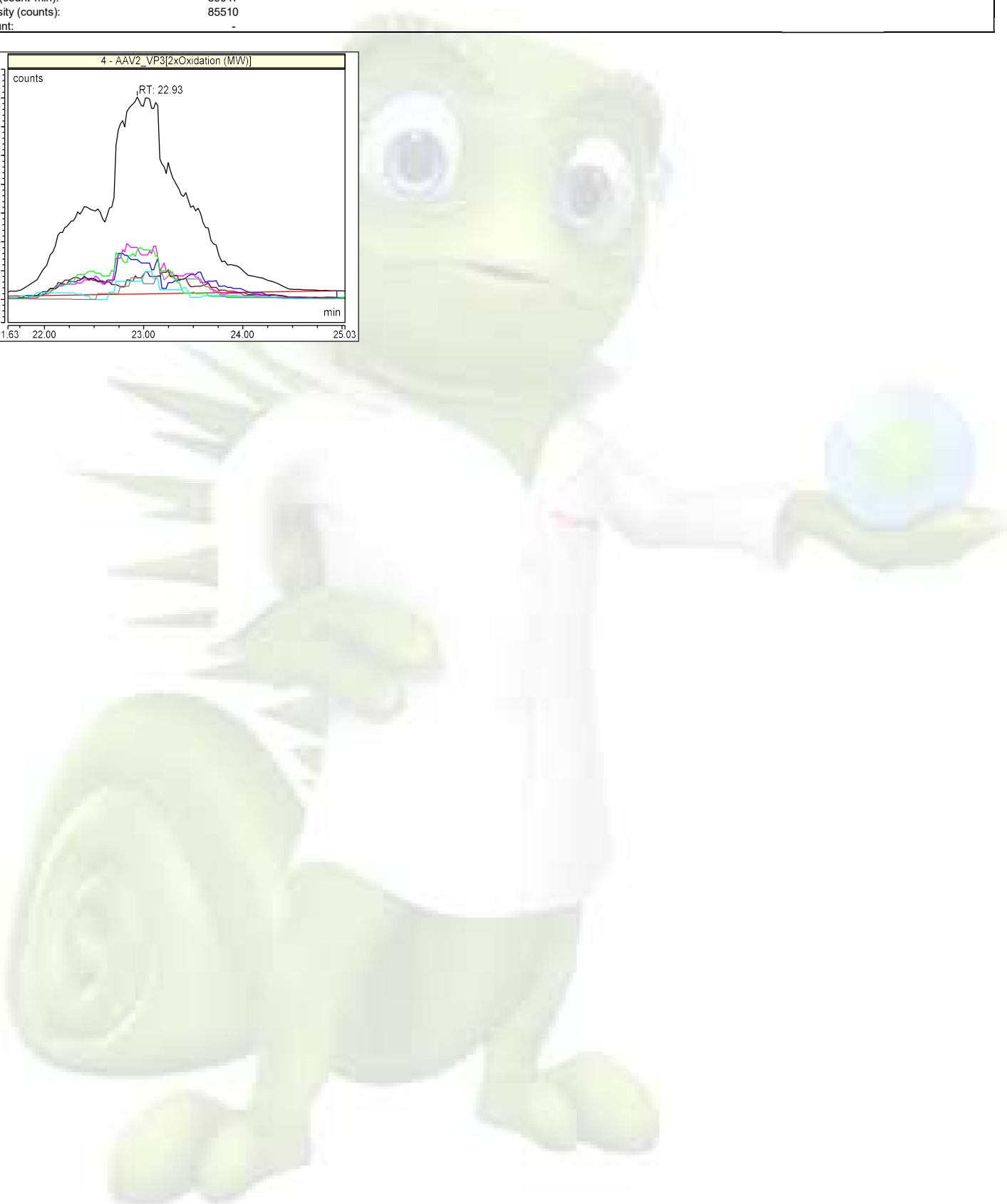
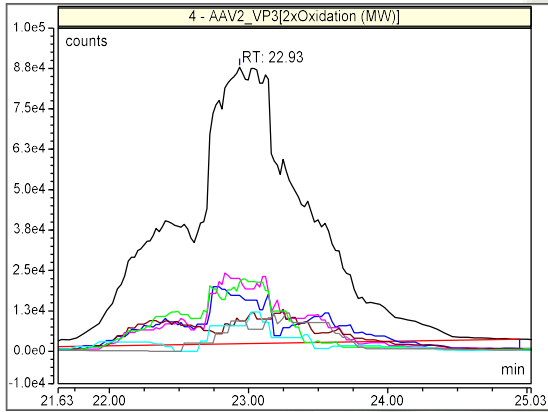
Retention time (min): 23.33

Expected Mass: 60005.780

Area (count\*min): 89047

Intensity (counts): 85510

Amount: -



**Component Name: AAV2 VP3[1xOxidation (MW)]**

Quantitation ions: 1277.428,1396.179,1464.229,1250.823,1305.183,1225.324

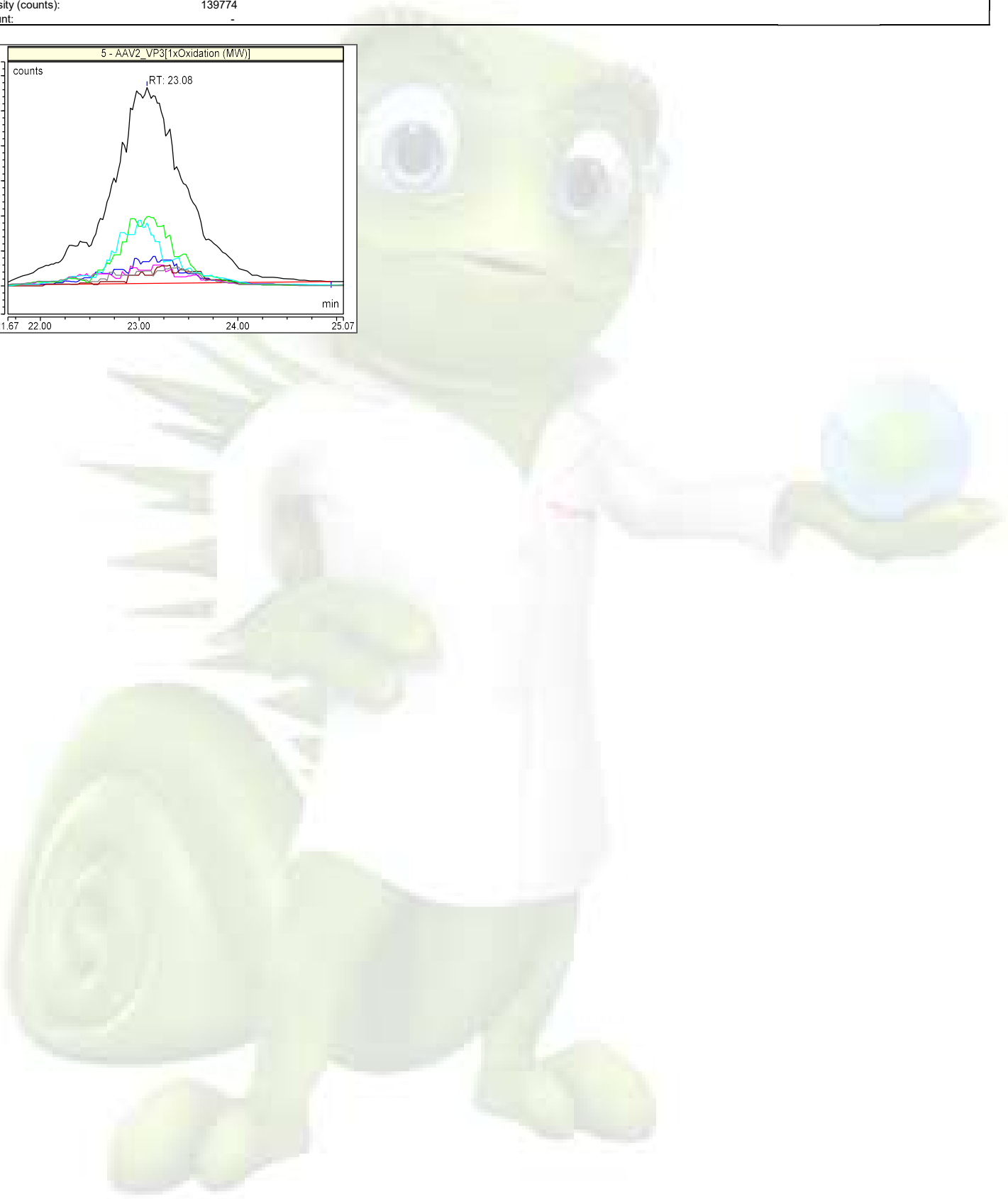
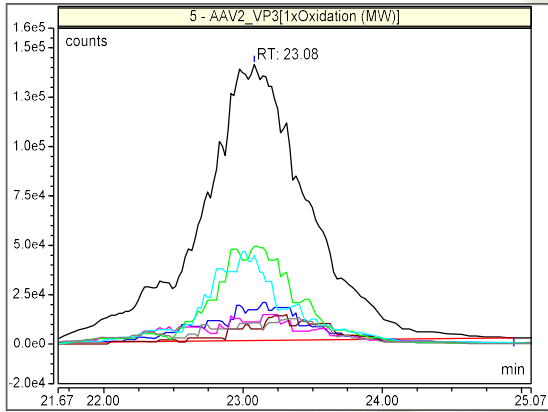
Retention time (min): 23.37

Expected Mass: 59989.780

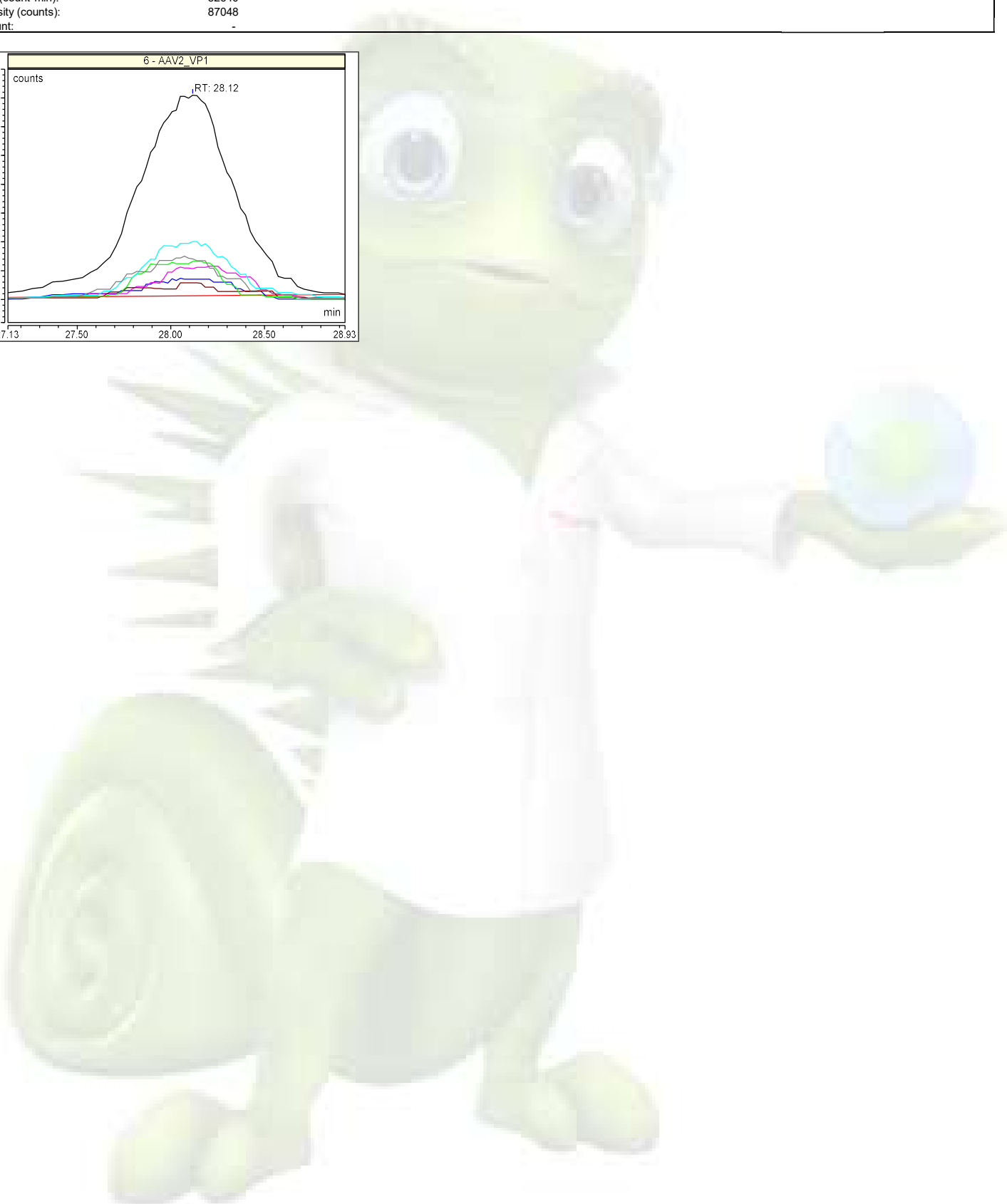
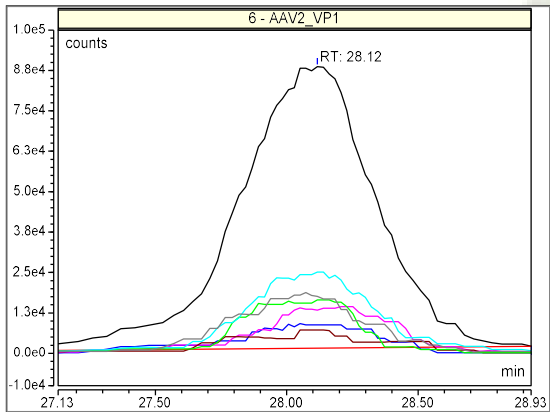
Area (count\*min): 125456

Intensity (counts): 139774

Amount: -



|                                 |   |
|---------------------------------|---|
| <b>Component Name: AAV2 VP1</b> |   |
| Quantitation ions:              | 1342.896,1388.388,1365.271,1300.301,1187.321,1279.998 |
| Retention time (min):           | 28.03   |
| Expected Mass:                  | 81854.900   |
| Area (count*min):               | 52640   |
| Intensity (counts):             | 87048   |
| Amount:                         | -   |



**Component Name: AAV2 VP1[1xOxidation (MW)]**

Quantitation ions: 1343.191,1154.150,1388.700,1187.572,1412.610,1170.616

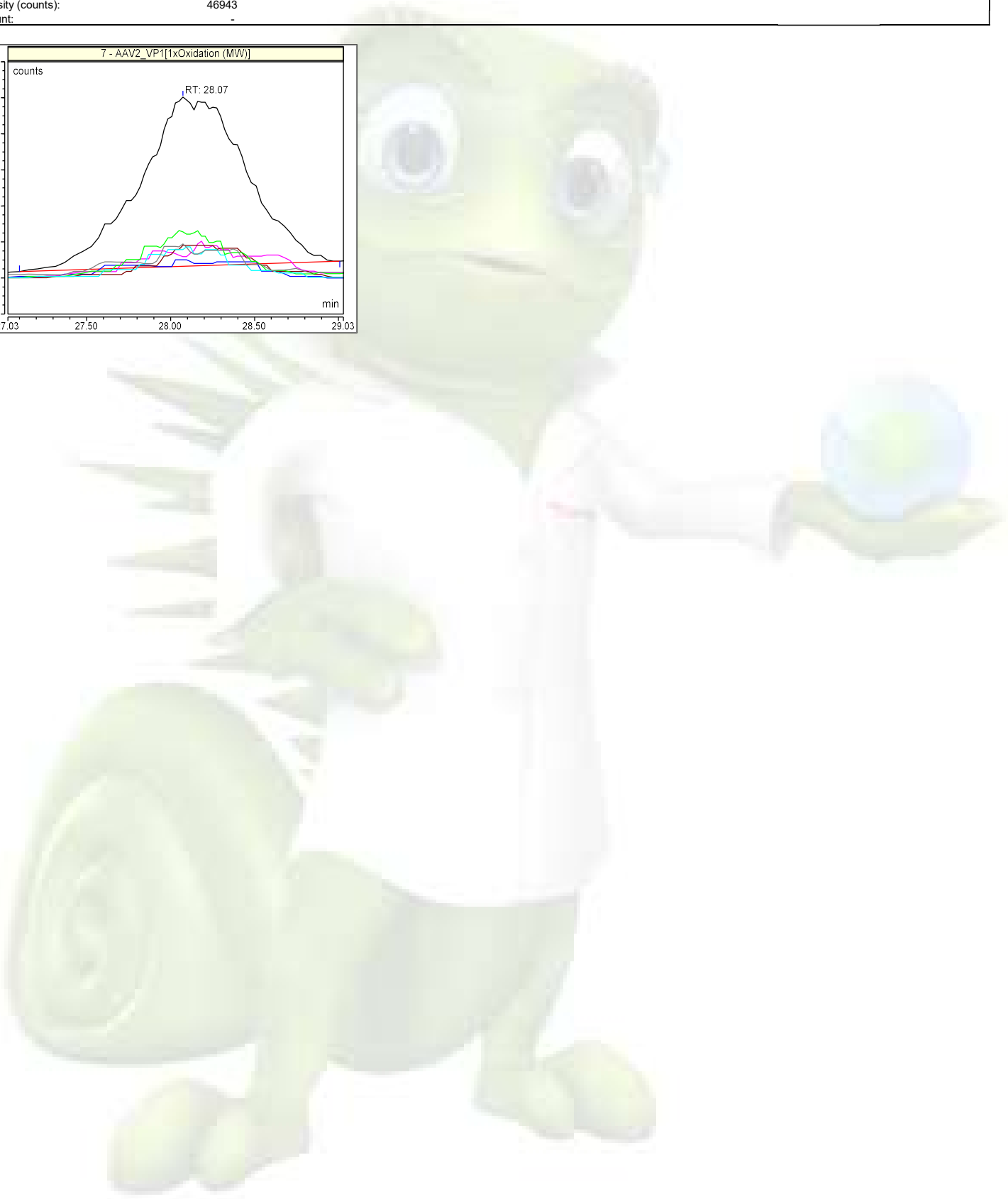
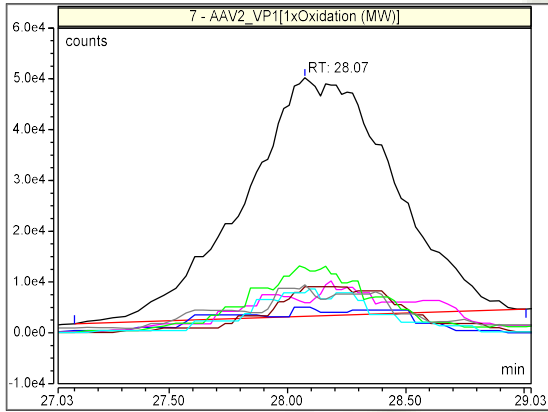
Retention time (min): 28.03

Expected Mass: 81870.890

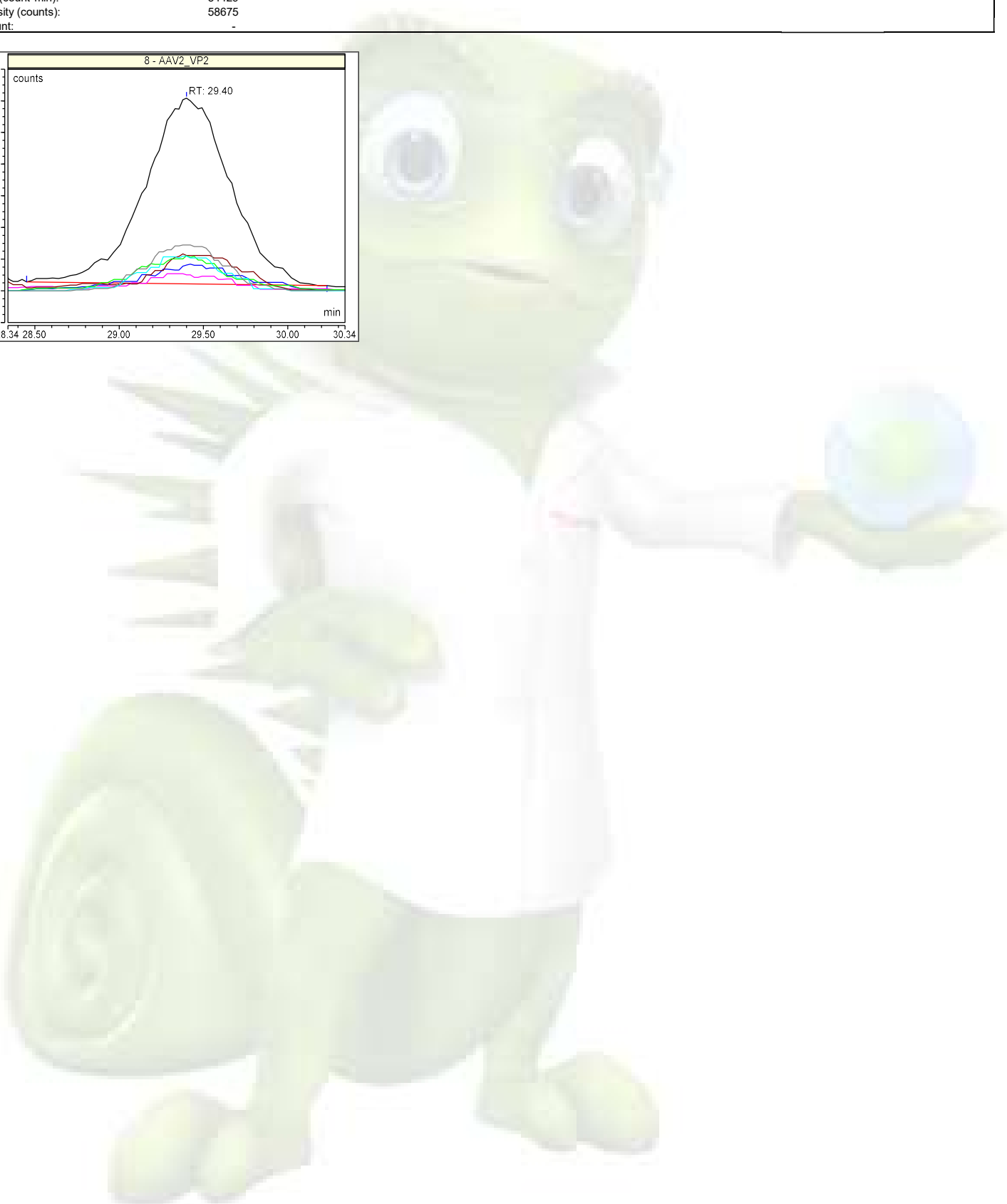
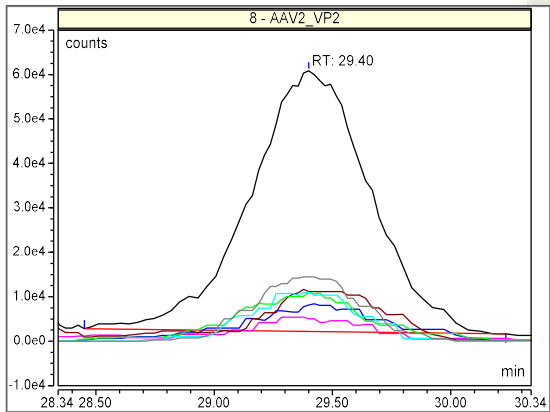
Area (count\*min): 34246

Intensity (counts): 46943

Amount: -



|                                 |  |
|---------------------------------|--|
| <b>Component Name: AAV2 VP2</b> |  |
| Quantitation ions:              | 1446.392, 1415.628, 1304.683, 1478.503, 1209.871, 1386.167 |
| Retention time (min):           | 29.34  |
| Expected Mass:                  | 66487.900  |
| Area (count*min):               | 34429  |
| Intensity (counts):             | 58675  |
| Amount:                         | -  |



**Component Name: AAV2 VP2[1xOxidation (MW)]**

Quantitation ions: 1446.760, 1305.022, 1167.738, 1358.263, 1147.628, 1331.107

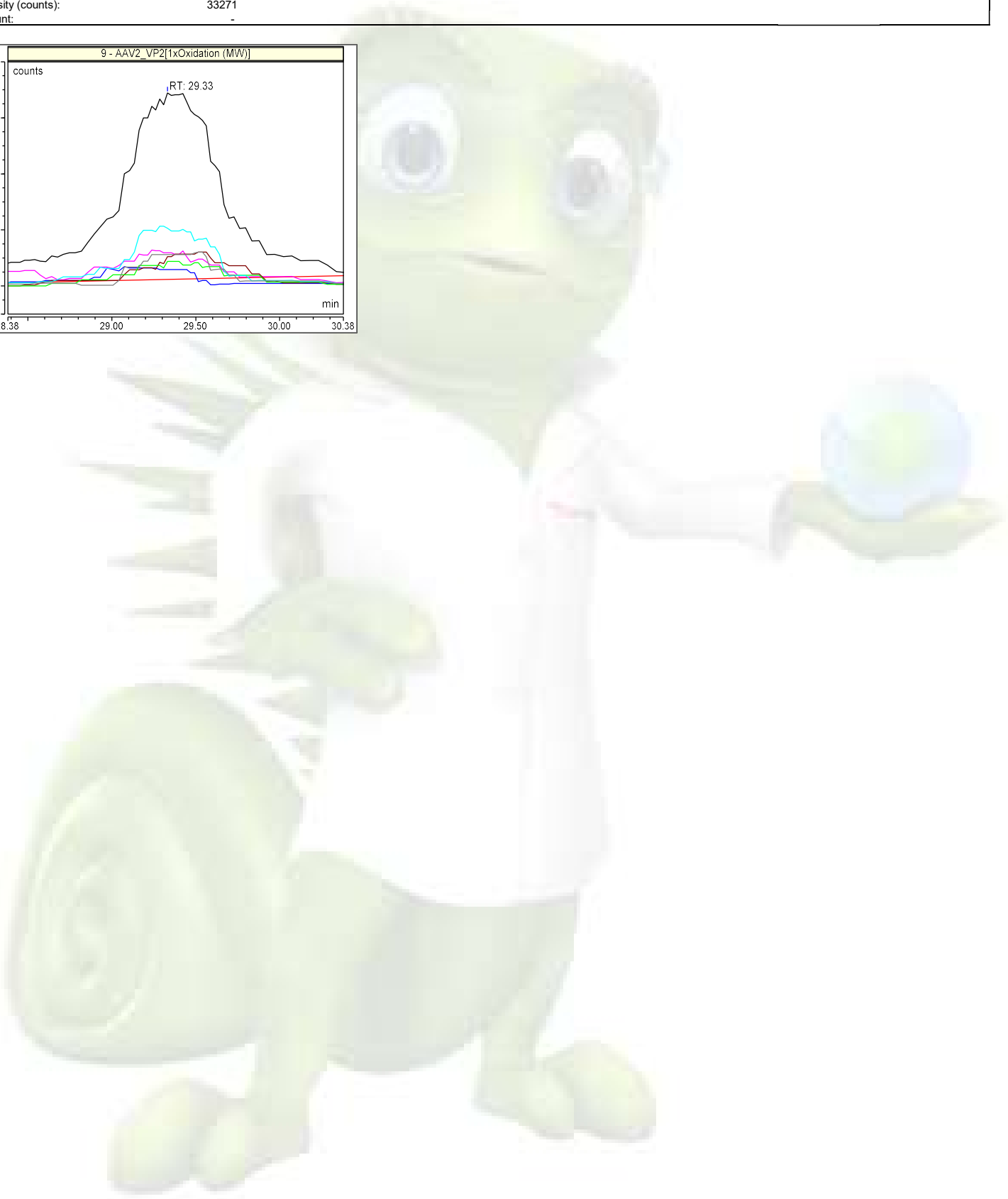
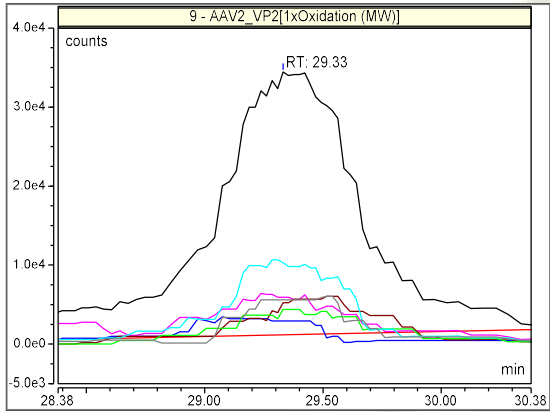
Retention time (min): 29.38

Expected Mass: 66503.900

Area (count\*min): 24846

Intensity (counts): 33271

Amount: -

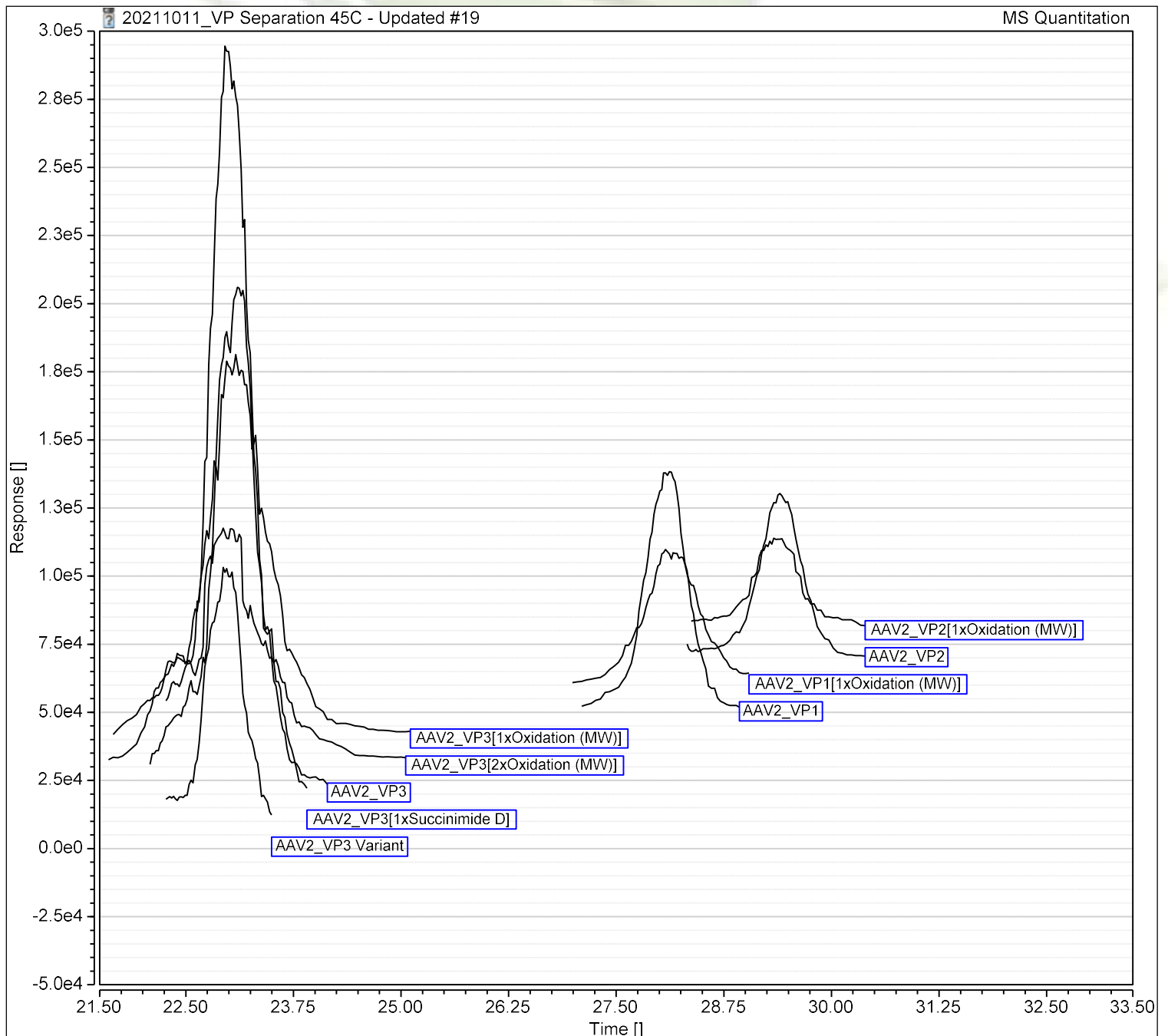


## Chromatogram and SST Results

### Injection Details

|                      |   |                   |        |
|----------------------|---|-------------------|--------|
| Injection Name:      | 20220120_AAV2_HEK_VP Sep_25C_64.5-58.5B_45K R                                 | Run Time (min):   | 45.00  |
| Vial Number:         | R:A1  | Injection Volume: | 8.56   |
| Injection Type:      | Unknown   |                   |        |
| Calibration Level:   |   |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_45K Res_CT25C_ |                   |        |
| Processing Method:   | AAV2 MS Quantitative In-depth Characterization                                | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 21/Jan/22 07:32   | Sample Weight:    | 1.0000 |

### Chromatogram





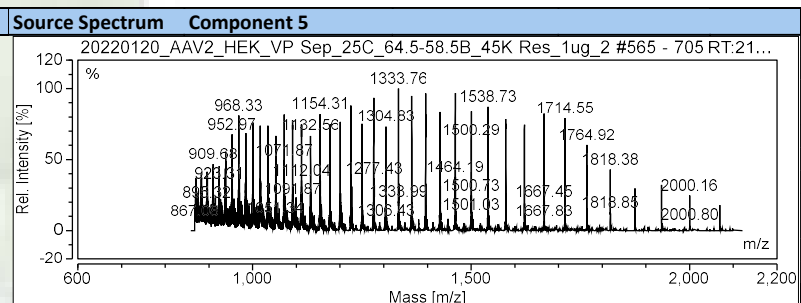
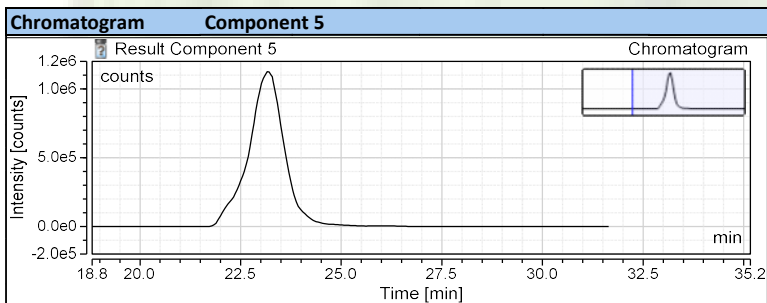
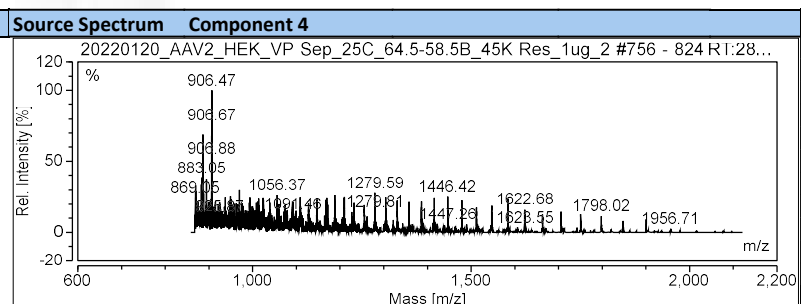
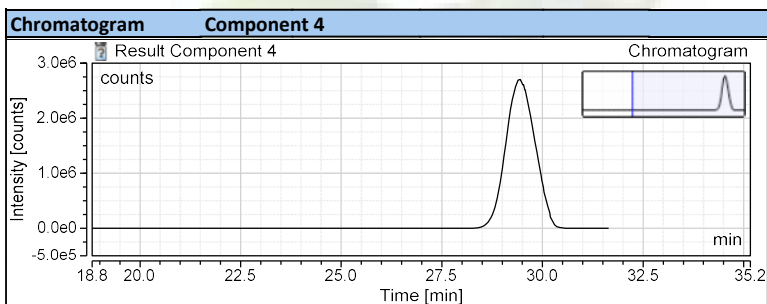
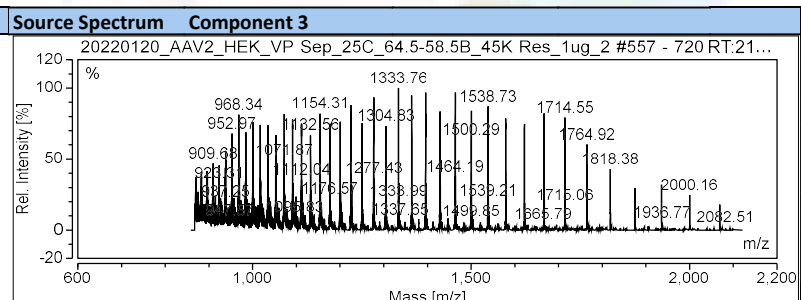
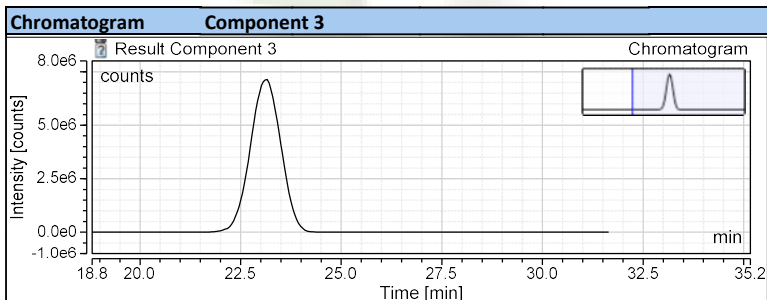
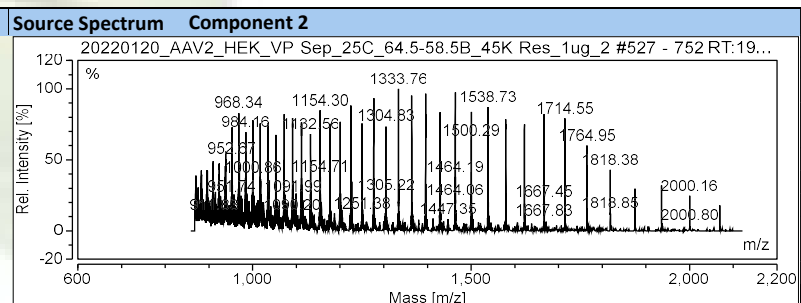
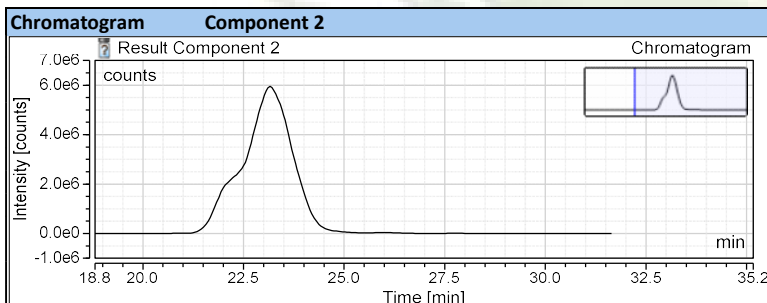
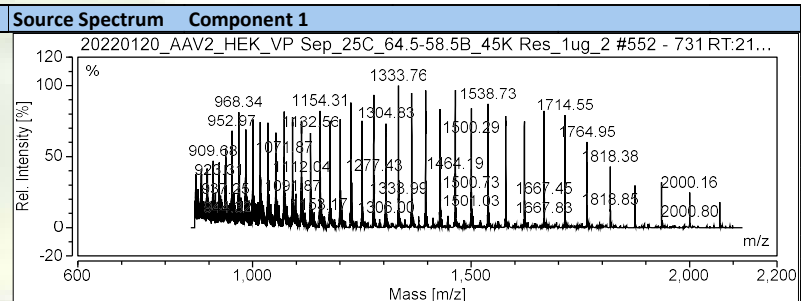
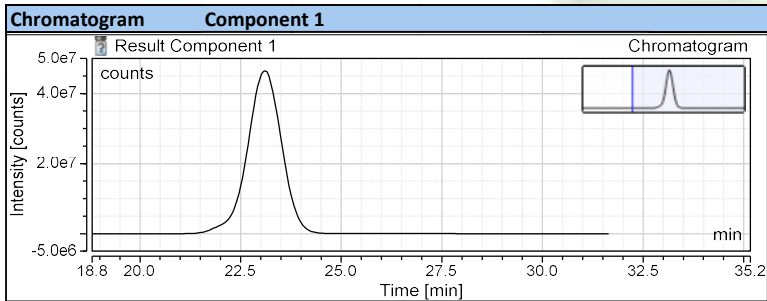
| SST Results                         |      |                |               |             |           |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

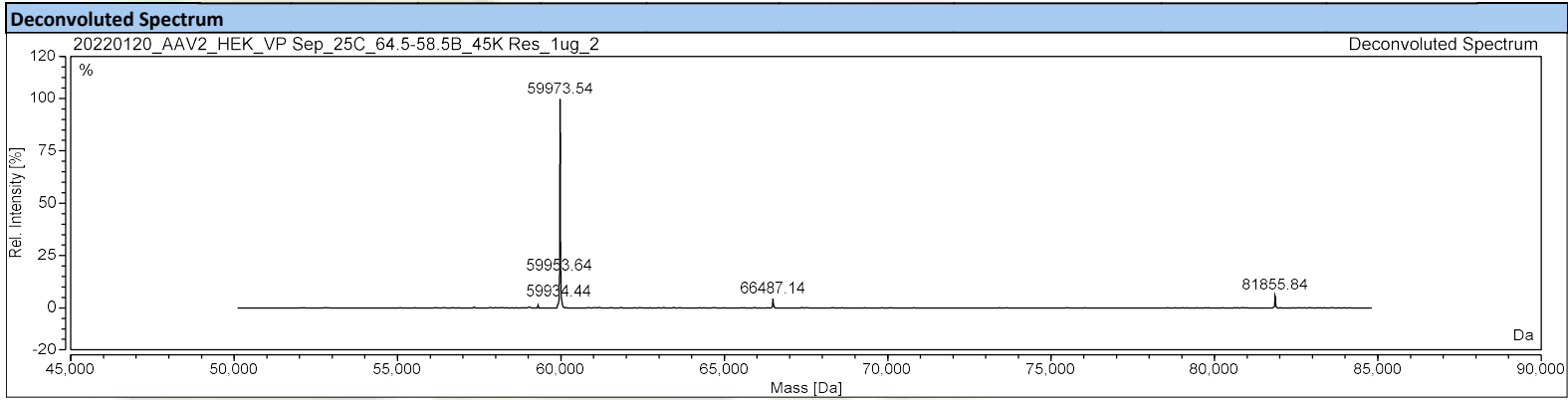
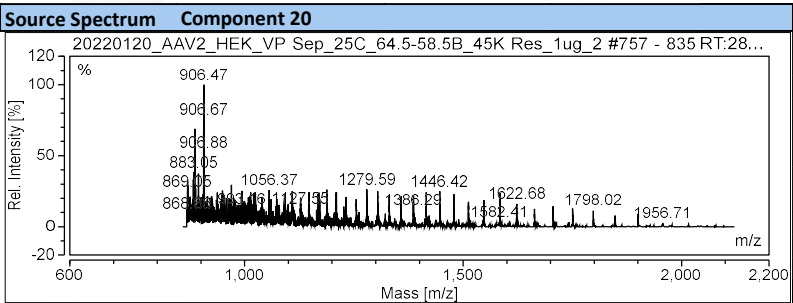
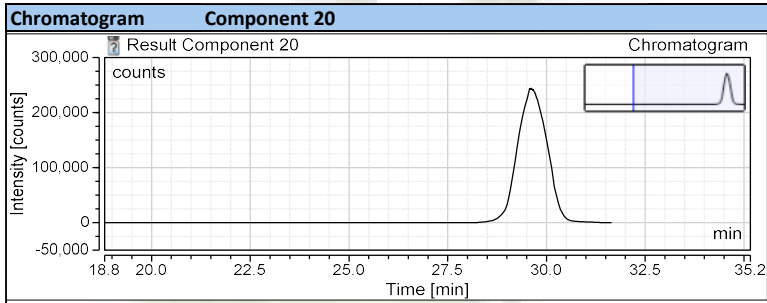
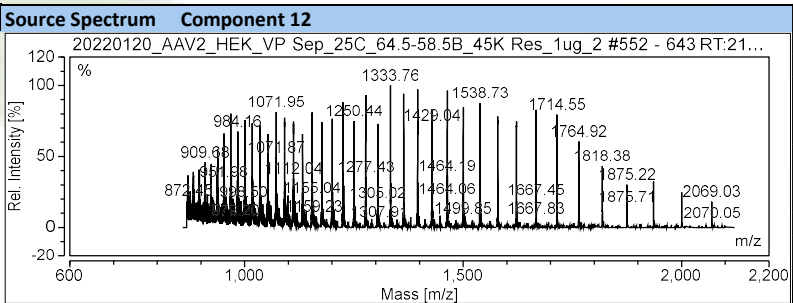
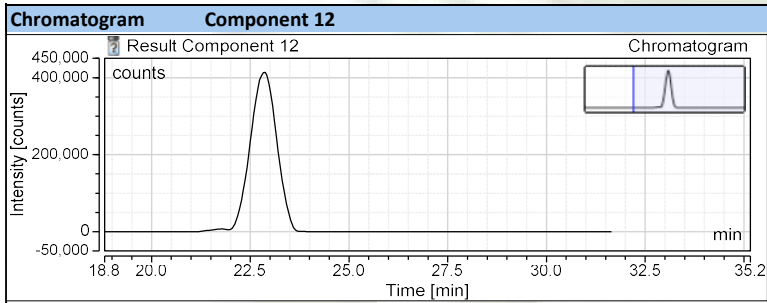
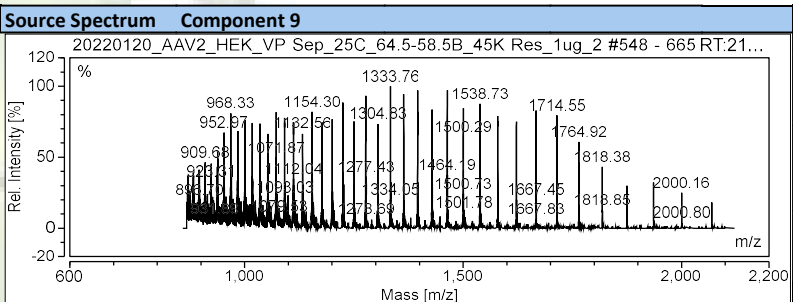
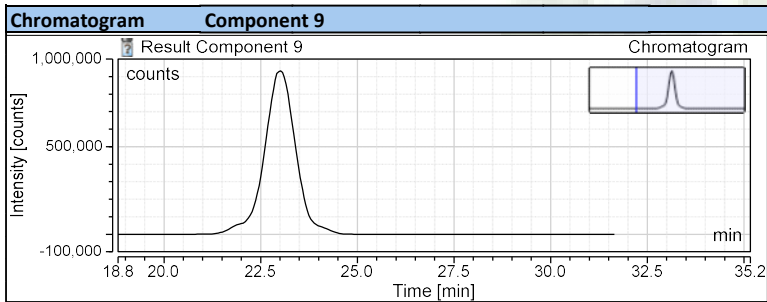
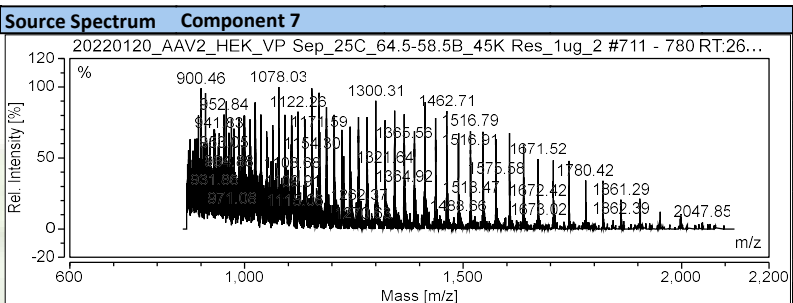
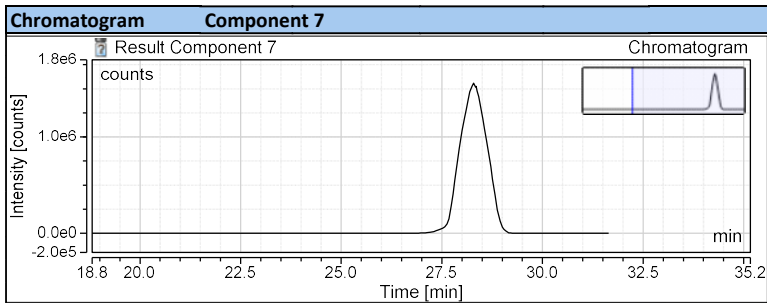


### Protein Deconvolution Summary

#### Injection Details

|                 |   |                      |  |
|-----------------|---|----------------------|--|
| Injection Name: | 20220120_AAV2_HEK_VP Sep_25C_64.5-58.5B_45K Res_1ug_2 | Injection Volume:    | 8.56   |
| Vial Number:    | R:A1  | Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide H      |
| Injection Type: | Unknown   | Processing Method:   | AAV2 MS Quantitative In-depth Characterization |
| Run Time (min): | 44.99   | Injection Date/Time: | 21/Jan/22 08:18                                |





| Result Component   | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
|--------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component 1 | 23.08  | 59973.436 | 0.84         | 13.99         | 0.000         | 1959279259       | 46.2                        | 100.00           | 41                      | 242.04 |
| Result Component 2 | 23.11  | 59991.819 | 1.81         | 30.24         | 18.383        | 430877006        | 10.2                        | 21.99            | 41                      | 161.22 |
| Result Component 3 | 23.08  | 59953.840 | 1.53         | 25.59         | -19.596       | 302960887        | 7.1                         | 15.46            | 34                      | 154.28 |
| Result Component 4 | 28.00  | 81855.271 | 1.70         | 20.79         | 21881.835     | 149335479        | 3.5                         | 7.62             | 55                      | 180.65 |
| Result Component 5 | 29.37  | 66487.295 | 1.24         | 18.66         | 6513.860      | 115071618        | 2.7                         | 5.87             | 42                      | 195.77 |
| Result Component 6 | 23.08  | 60018.234 | 1.74         | 28.98         | 44.799        | 61338748         | 1.4                         | 3.13             | 25                      | 68.82  |
| Result Component 7 | 23.76  | 60006.121 | 1.86         | 30.94         | 32.686        | 59932437         | 1.4                         | 3.06             | 39                      | 79.35  |
| Result Component 8 | 29.37  | 66504.638 | 1.95         | 29.34         | 6531.202      | 43476745         | 1.0                         | 2.22             | 41                      | 102.72 |
| Result Component 9 | 27.98  | 81872.829 | 2.11         | 25.73         | 21899.393     | 38271649         | 0.9                         | 1.95             | 44                      | 90.47  |

|                      |       |           |      |       |           |          |     |      |    |        |
|----------------------|-------|-----------|------|-------|-----------|----------|-----|------|----|--------|
| Result Component 11  | 22.91 | 59299.871 | 1.48 | 25.01 | -673.565  | 28850826 | 0.7 | 1.47 | 30 | 69.26  |
| Result Component 12  | 22.40 | 59954.253 | 1.72 | 28.73 | -19.183   | 27440115 | 0.6 | 1.40 | 33 | 139.46 |
| Result Component 19  | 27.67 | 81874.453 | 2.73 | 33.39 | 21901.017 | 10900834 | 0.3 | 0.56 | 50 | 109.08 |
| Result Component 77  | 24.00 | 60020.804 | 1.54 | 25.71 | 47.368    | 3356483  | 0.1 | 0.17 | 33 | 78.05  |
| Result Component 152 | 29.15 | 66467.903 | 1.85 | 27.80 | 6494.467  | 1942465  | 0.0 | 0.10 | 19 | 65.67  |



## Component Summary

### Injection Details

|                 |                                     |                      |                                       |
|-----------------|-------------------------------------|----------------------|---------------------------------------|
| Injection Name: | 20220120_AAV2_HEK_VP_Sep_25C_64.5-5 | Injection Volume:    | 8.56                                  |
| Vial Number:    | R:A1                                | Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein     |
| Injection Type: | Unknown                             | Processing Method:   | AAV2 MS Quantitative In-depth Charact |
| Run Time (min): | 44.99                               | Injection Date/Time: | 21/Jan/22 08:18                       |

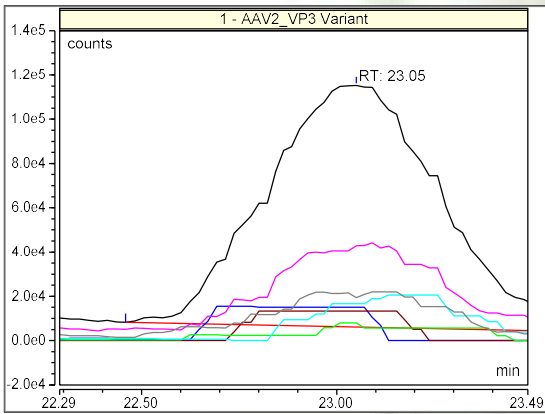
| Peak Name                  | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|----------------------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3[1xOxidation (MW)] | 22.988                | 172743.238         | 23.92              | 1277.428,1396.179,1464.229,1250.823,1305.183,1225.324 | 59989.780 | PASS                           |
| AAV2_VP3 Variant           | 23.050                | 55540.588          | 7.69               | 1447.374,957.488,1380.090,1163.736,1099.120,973.145   | 59301.030 | PASS                           |
| AAV2_VP3[1xSuccinimide D]  | 23.071                | 196617.847         | 27.23              | 952.674,923.374,968.020,937.789,1017.182,1000.250     | 59955.770 | PASS                           |
| AAV2_VP3                   | 23.071                | 92029.695          | 12.74              | 1364.311,1500.570,1333.966,1395.925,1463.892,1225.374 | 59973.780 | PASS                           |
| AAV2_VP3[2xOxidation (MW)] | 23.092                | 75537.982          | 10.46              | 1133.193,1092.027,1112.217,1072.520,1539.665,1464.606 | 60005.780 | PASS                           |
| AAV2_VP1[1xOxidation (MW)] | 28.146                | 38902.016          | 5.39               | 1343.191,1154.150,1388.700,1187.572,1412.610,1170.616 | 81870.890 | PASS                           |
| AAV2_VP1                   | 28.168                | 40665.176          | 5.63               | 1342.896,1388.388,1365.271,1300.301,1187.321,1279.998 | 81854.900 | PASS                           |
| AAV2_VP2                   | 29.391                | 30432.656          | 4.21               | 1446.392,1415.628,1304.683,1478.503,1209.871,1386.167 | 66487.900 | PASS                           |
| AAV2_VP2[1xOxidation (MW)] | 29.437                | 19629.625          | 2.72               | 1446.760,1305.022,1167.738,1358.263,1147.628,1331.107 | 66503.900 | PASS                           |
| <b>100.00</b>              |                       |                    |                    |   |           |                                |

### Component Name: AAV2\_VP3 Variant

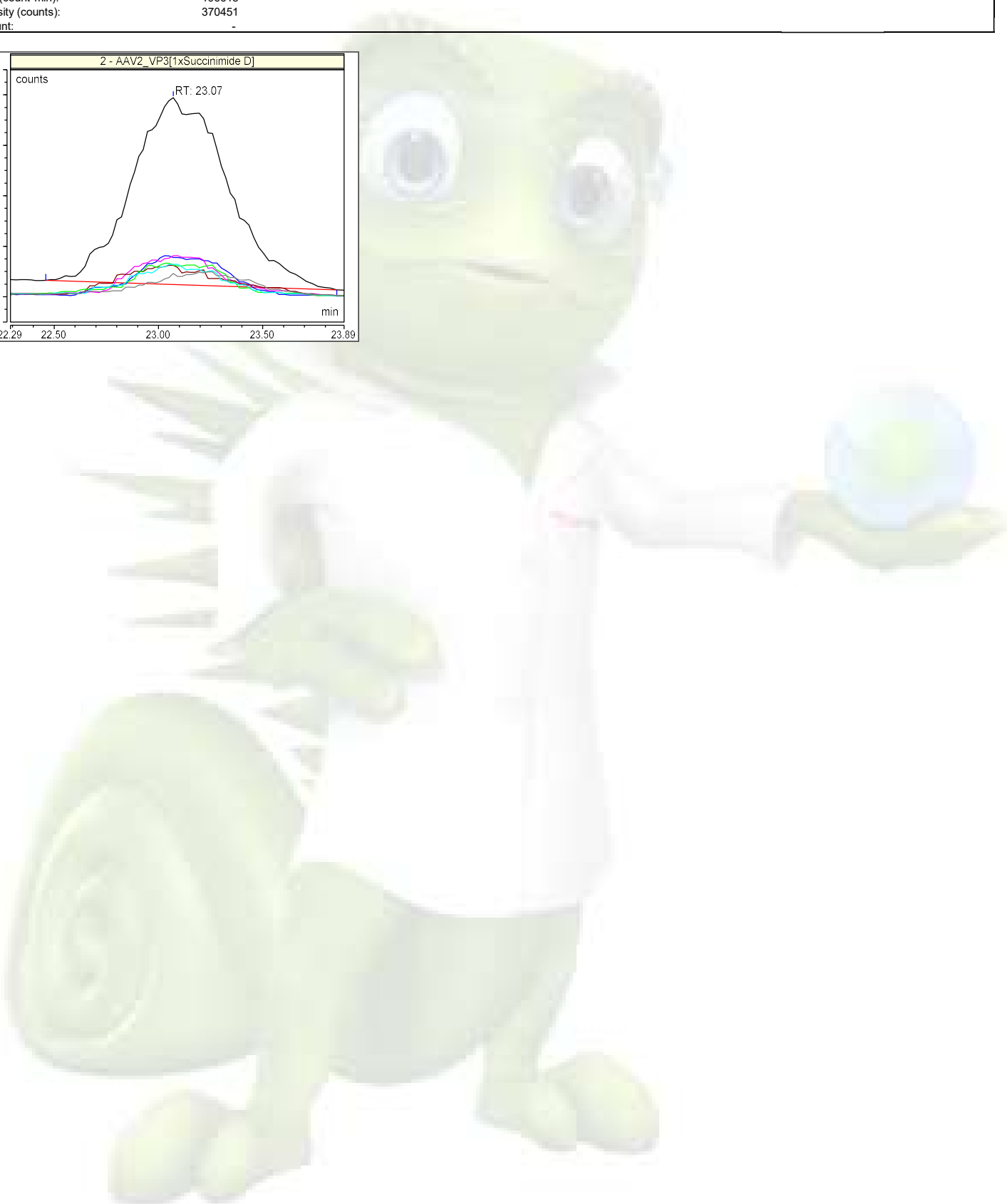
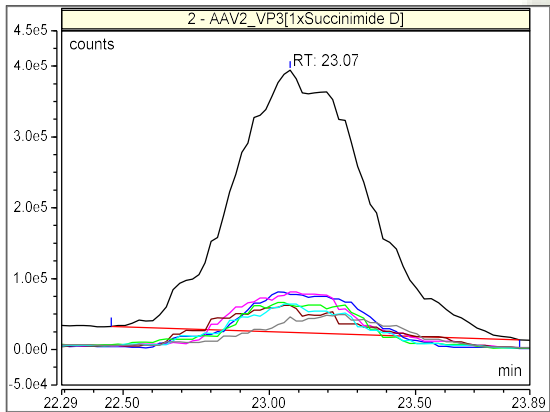
Quantitation ions: 1447.374,957.488,1380.090,1163.736,1099.120,973.145

Retention time (min): 22.89  
Expected Mass: 59301.030

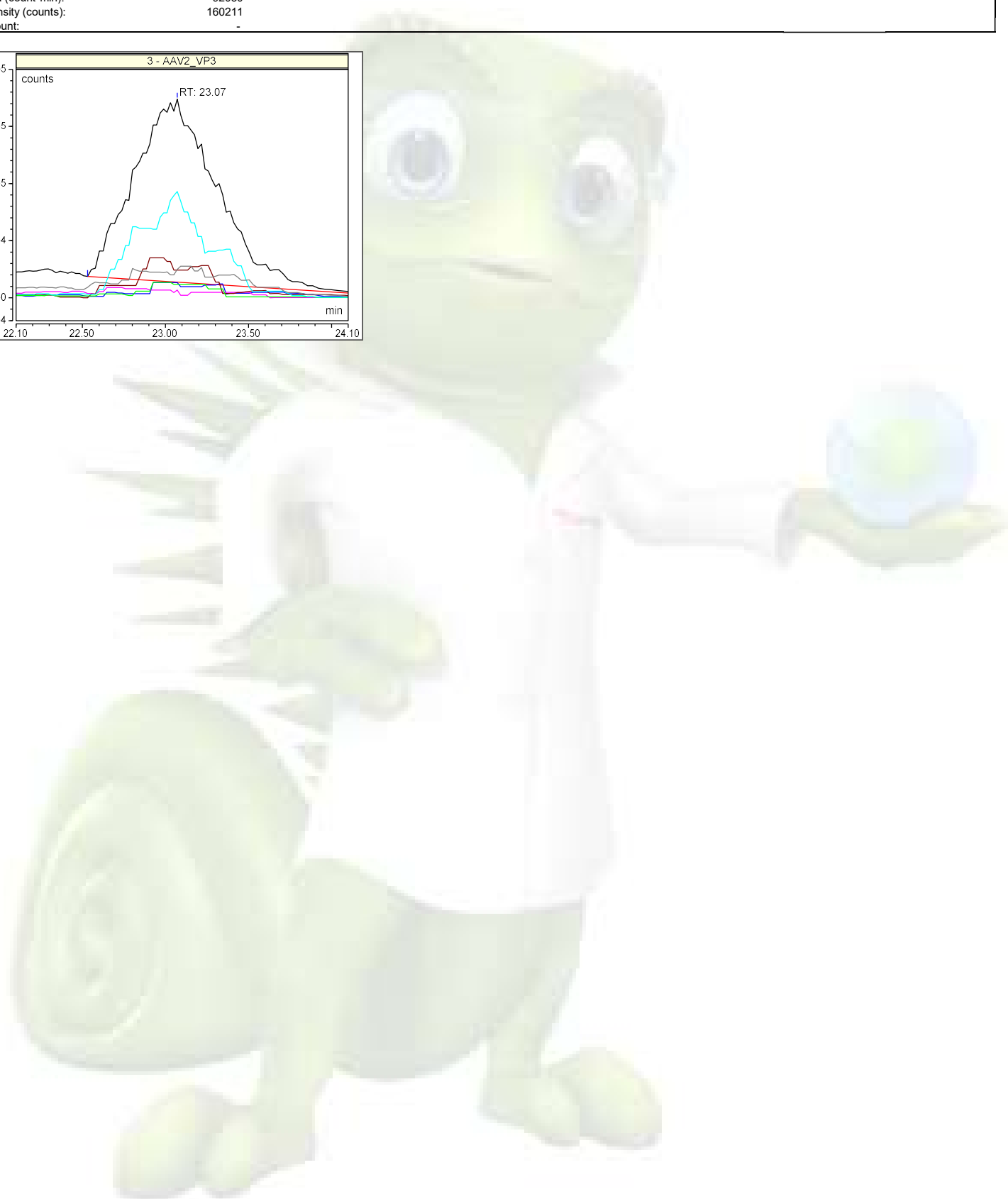
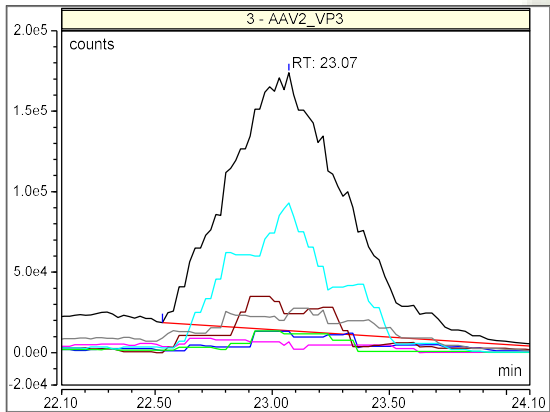
Area (count\*min): 55541  
Intensity (counts): 109089  
Amount: -



|  |   |
|--|---|
| <b>Component Name: AAV2 VP3[1xSuccinimide D]</b> |   |
| Quantitation ions:                               | 952.674,923.374,968.020,937.789,1017.182,1000.250 |
| Retention time (min):                            | 23.09   |
| Expected Mass:                                   | 59955.770   |
| Area (count*min):                                | 196618  |
| Intensity (counts):                              | 370451  |
| Amount:  | -   |



|                                 |   |
|---------------------------------|---|
| <b>Component Name: AAV2 VP3</b> |   |
| Quantitation ions:              | 1364.311,1500.570,1333.966,1395.925,1463.892,1225.374 |
| Retention time (min):           | 23.10   |
| Expected Mass:                  | 59973.780   |
| Area (count*min):               | 92030   |
| Intensity (counts):             | 160211  |
| Amount:                         | -   |



**Component Name: AAV2 VP3[2xOxidation (MW)]**

Quantitation ions: 1133.193,1092.027,1112.217,1072.520,1539.665,1464.606

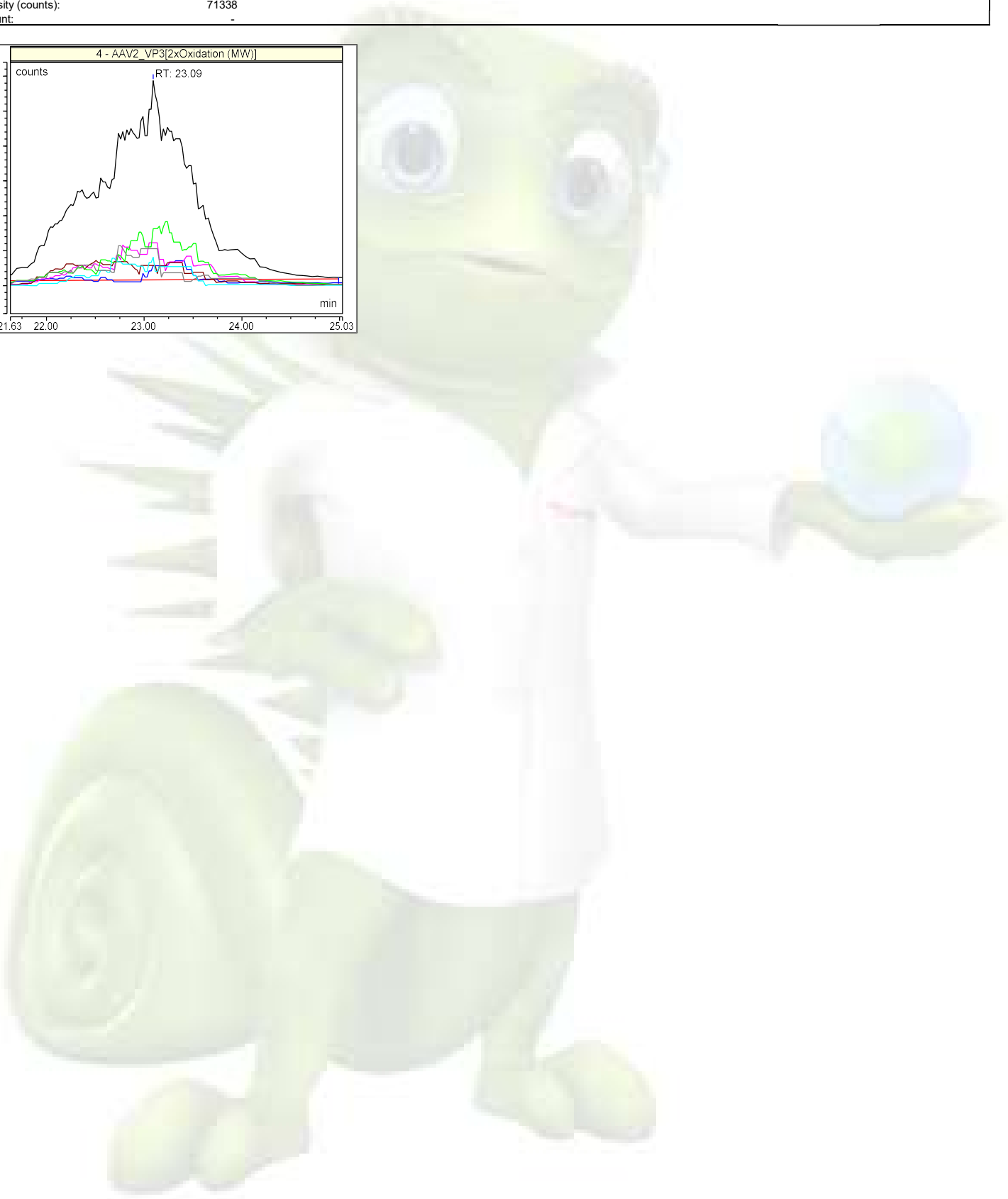
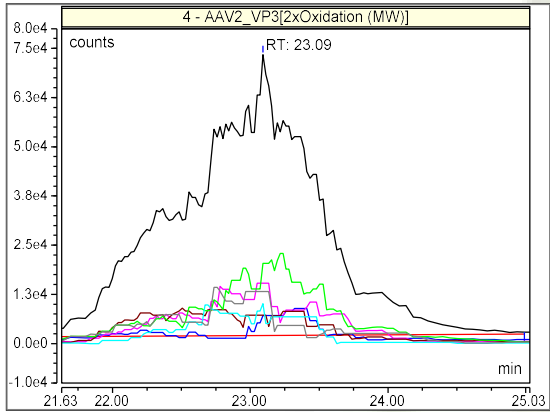
Retention time (min): 23.33

Expected Mass: 60005.780

Area (count\*min): 75538

Intensity (counts): 71338

Amount: -





**Component Name: AAV2 VP3[1xOxidation (MW)]**

Quantitation ions: 1277.428,1396.179,1464.229,1250.823,1305.183,1225.324

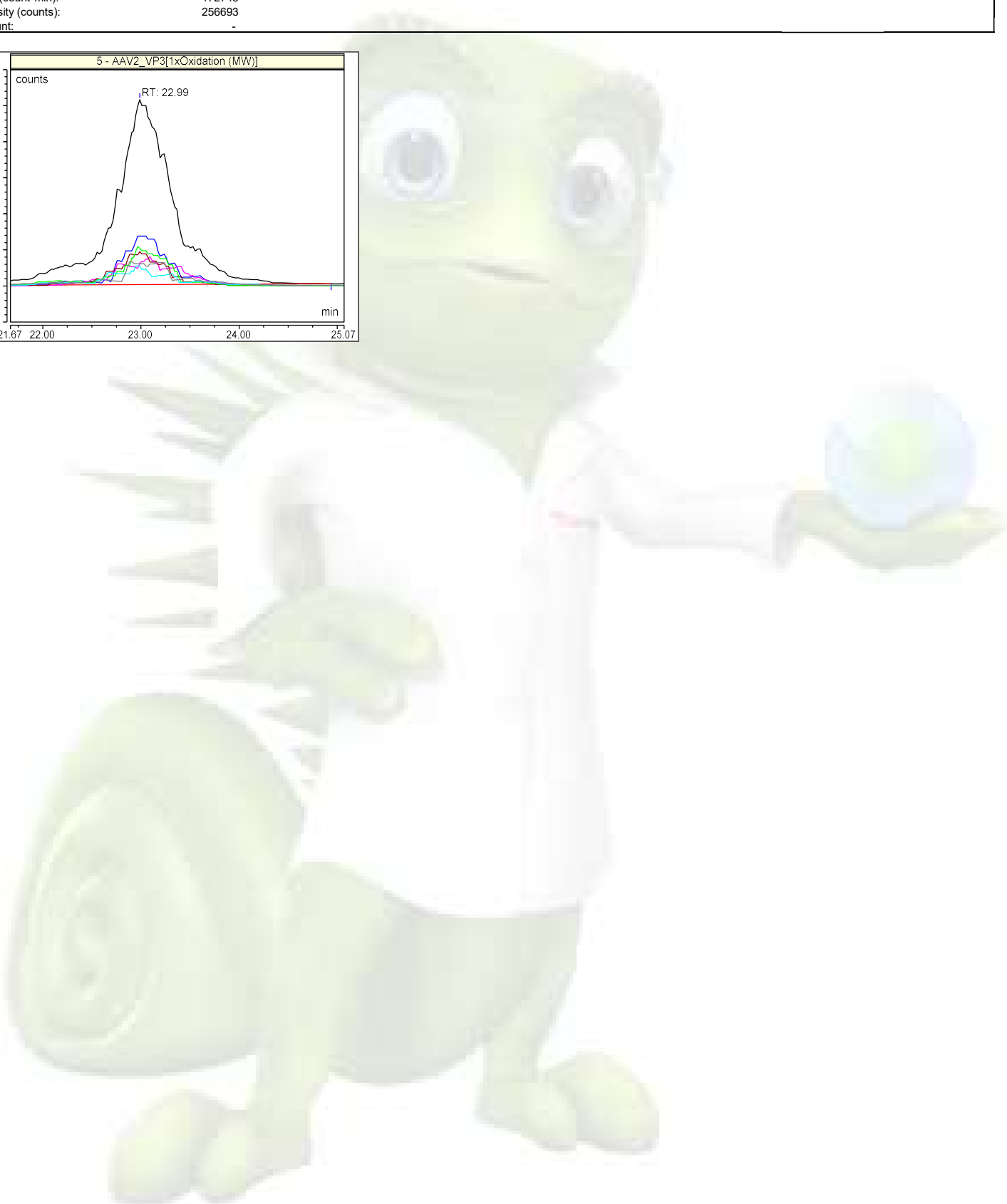
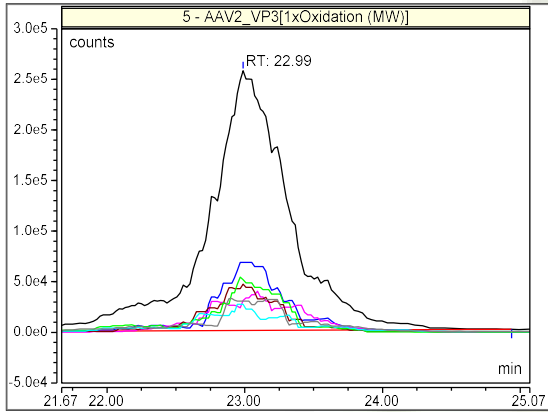
Retention time (min): 23.37

Expected Mass: 59989.780

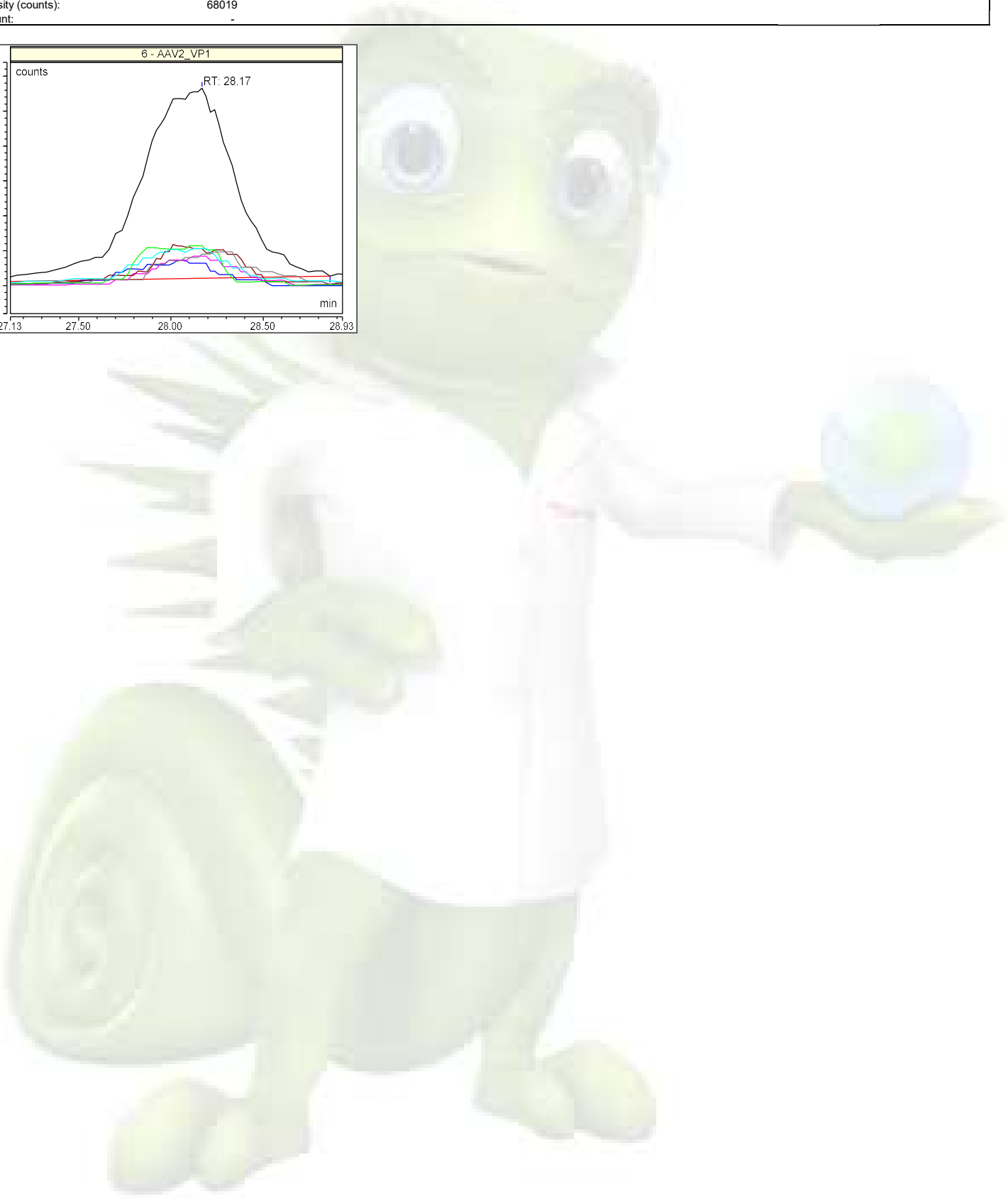
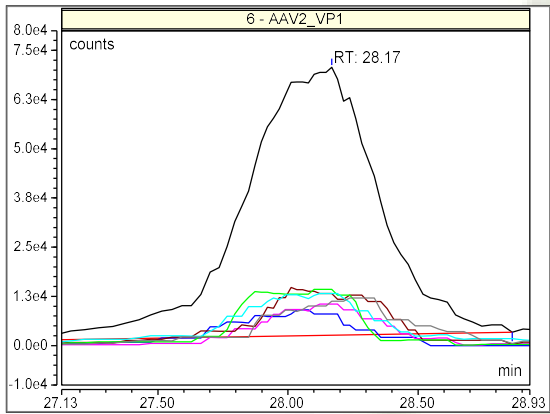
Area (count\*min): 172743

Intensity (counts): 256693

Amount: -



|                                 |   |
|---------------------------------|---|
| <b>Component Name: AAV2 VP1</b> |   |
| Quantitation ions:              | 1342.896,1388.388,1365.271,1300.301,1187.321,1279.998 |
| Retention time (min):           | 28.03   |
| Expected Mass:                  | 81854.900   |
| Area (count*min):               | 40665   |
| Intensity (counts):             | 68019   |
| Amount:                         | -   |



**Component Name: AAV2 VP1[1xOxidation (MW)]**

Quantitation ions: 1343.191,1154.150,1388.700,1187.572,1412.610,1170.616

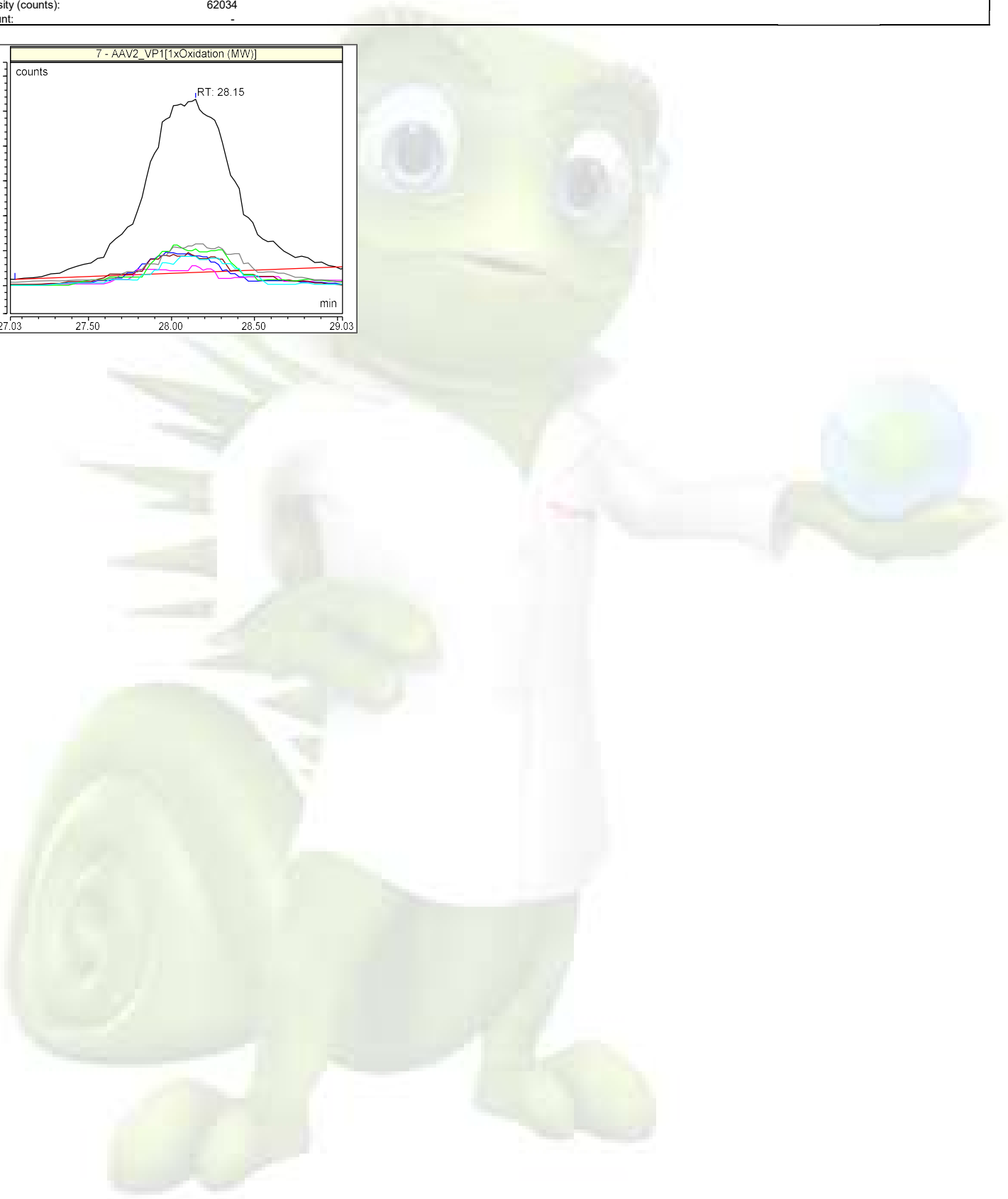
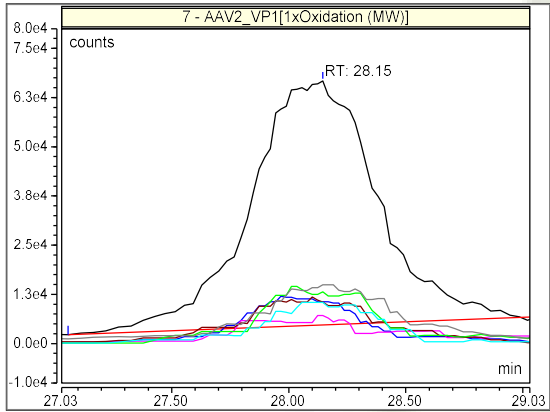
Retention time (min): 28.03

Expected Mass: 81870.890

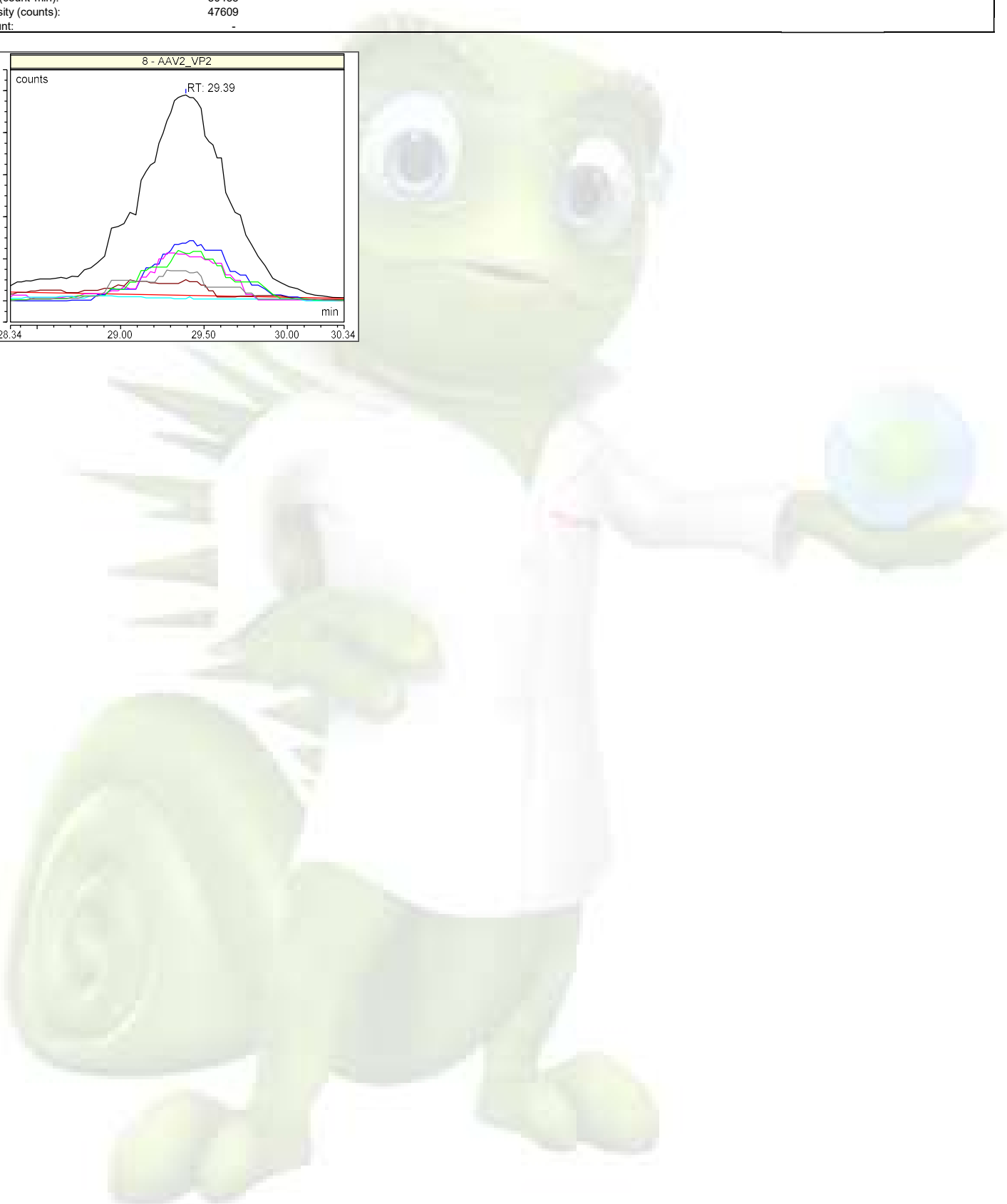
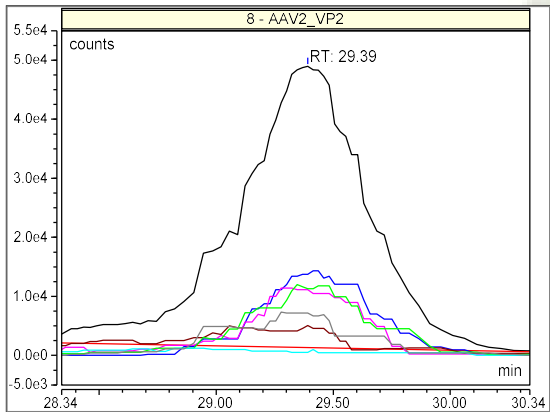
Area (count\*min): 38902

Intensity (counts): 62034

Amount: -



|                                 |  |
|---------------------------------|--|
| <b>Component Name: AAV2 VP2</b> |  |
| Quantitation ions:              | 1446.392, 1415.628, 1304.683, 1478.503, 1209.871, 1386.167 |
| Retention time (min):           | 29.34  |
| Expected Mass:                  | 66487.900  |
| Area (count*min):               | 30433  |
| Intensity (counts):             | 47609  |
| Amount:                         | -  |



**Component Name: AAV2 VP2[1xOxidation (MW)]**

Quantitation ions: 1446.760, 1305.022, 1167.738, 1358.263, 1147.628, 1331.107

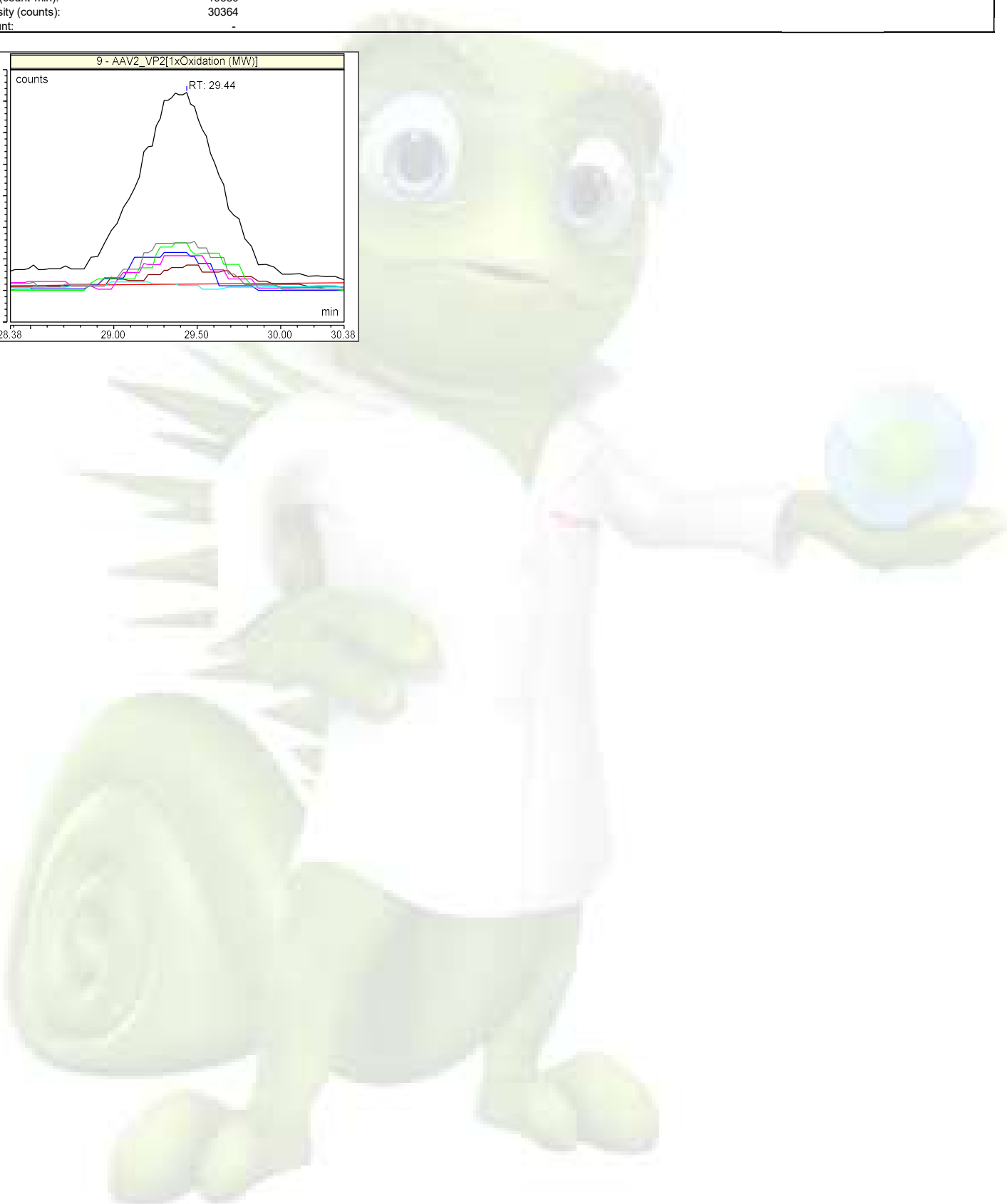
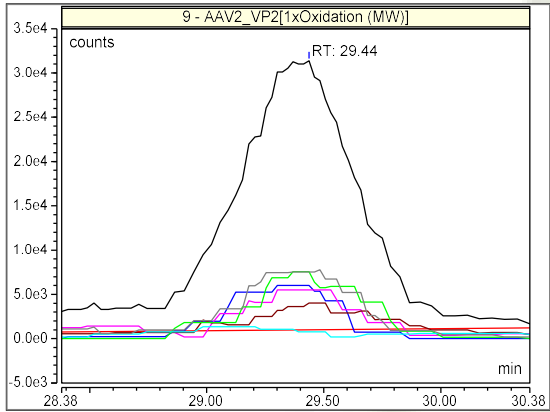
Retention time (min): 29.38

Expected Mass: 66503.900

Area (count\*min): 19630

Intensity (counts): 30364

Amount: -

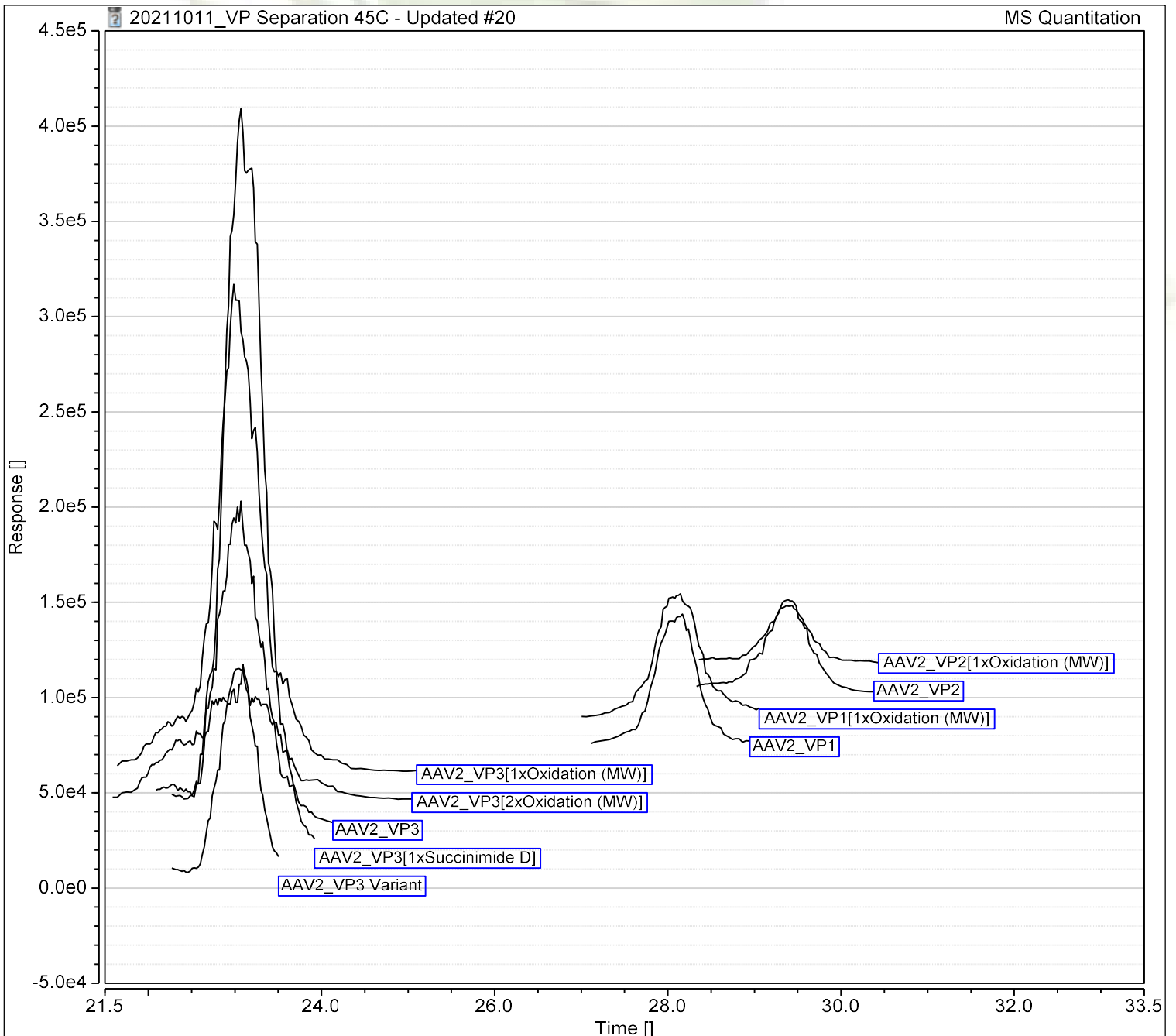


## Chromatogram and SST Results

### Injection Details

|                      |   |                   |        |
|----------------------|---|-------------------|--------|
| Injection Name:      | 20220120_AAV2_HEK_VP Sep_25C_64.5-58.5B_45K R                                 | Run Time (min):   | 44.99  |
| Vial Number:         | R:A1  | Injection Volume: | 8.56   |
| Injection Type:      | Unknown   |                   |        |
| Calibration Level:   |   |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_45K Res_CT25C_ |                   |        |
| Processing Method:   | AAV2 MS Quantitative In-depth Characterization                                | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 21/Jan/22 08:18   | Sample Weight:    | 1.0000 |

### Chromatogram



### SST Results

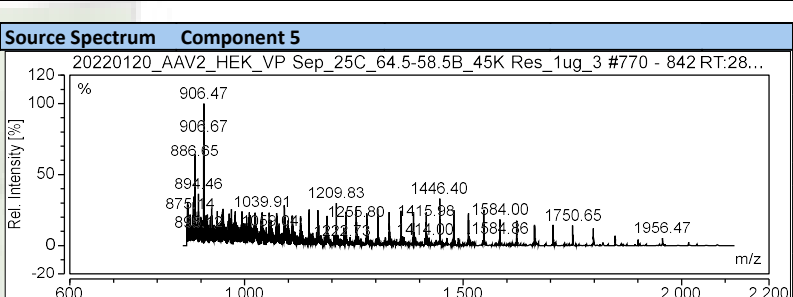
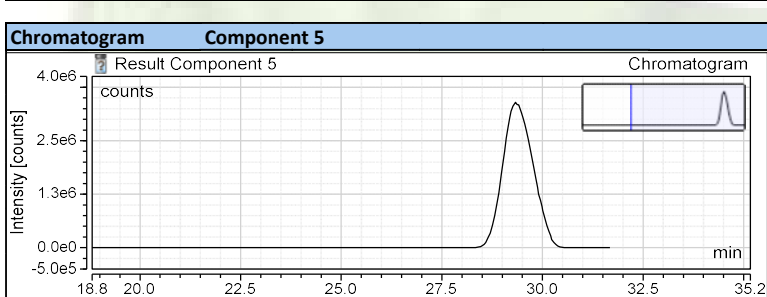
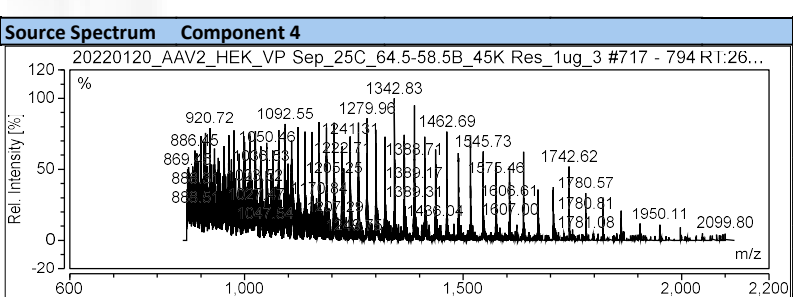
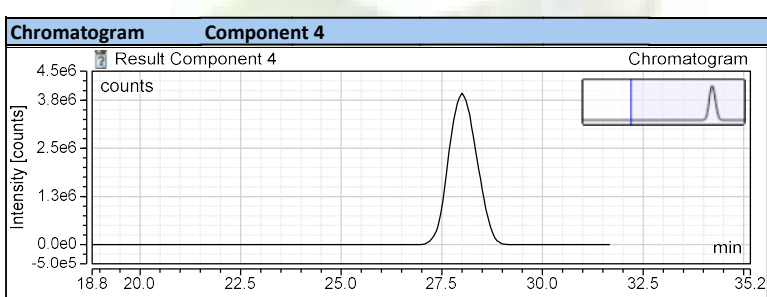
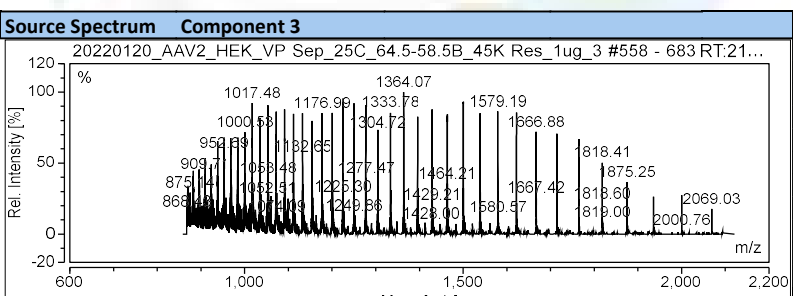
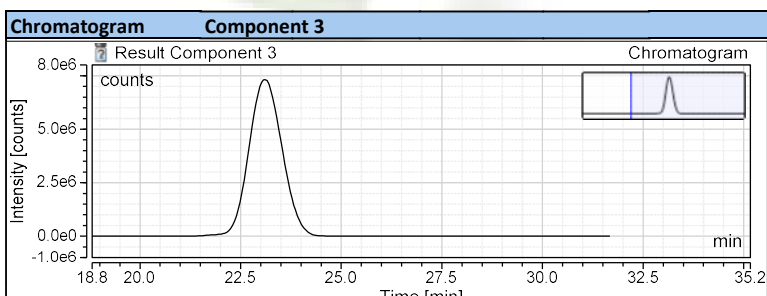
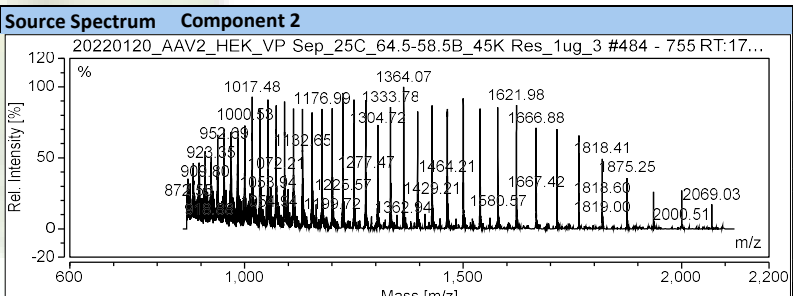
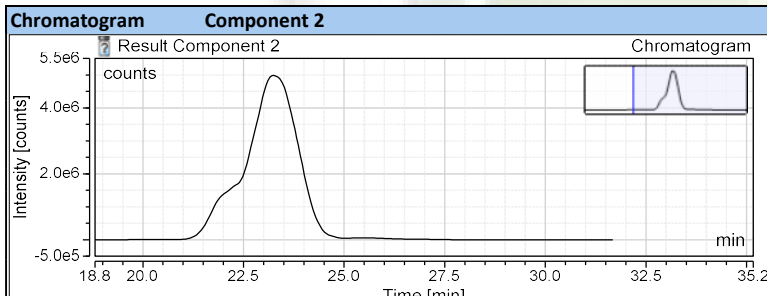
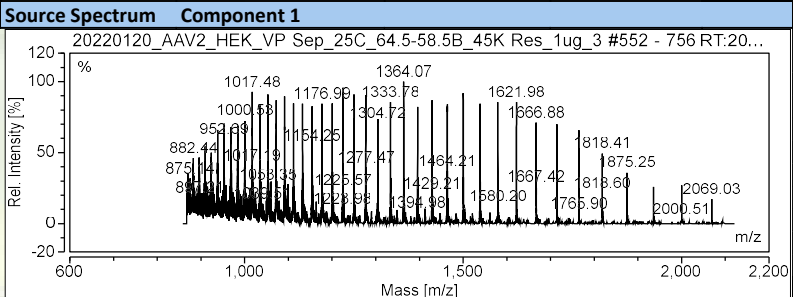
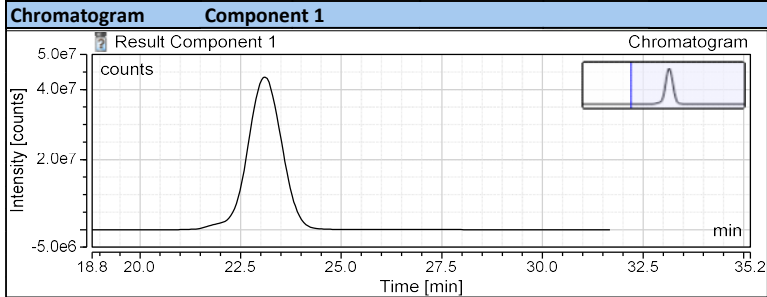
| No.                                 | Name | Inj.Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|---------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |               | Total Result: | Passed      |           |



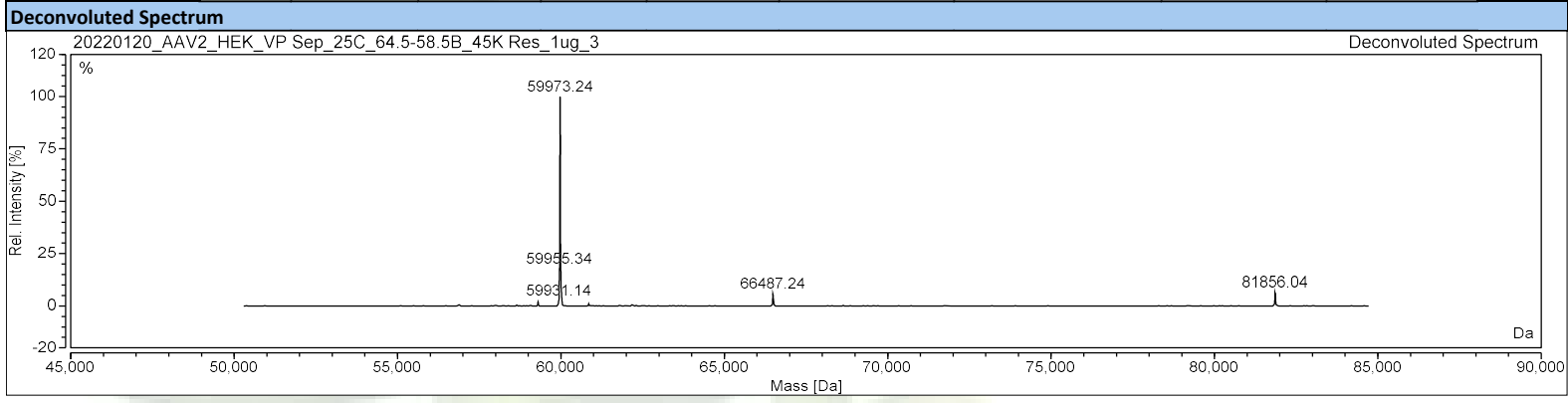
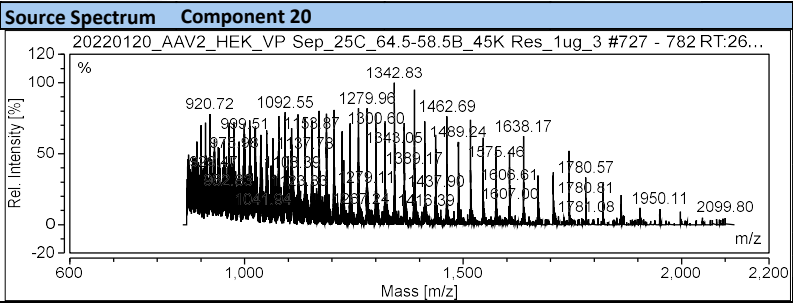
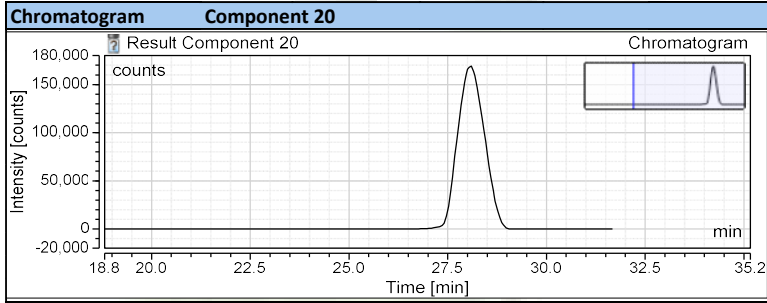
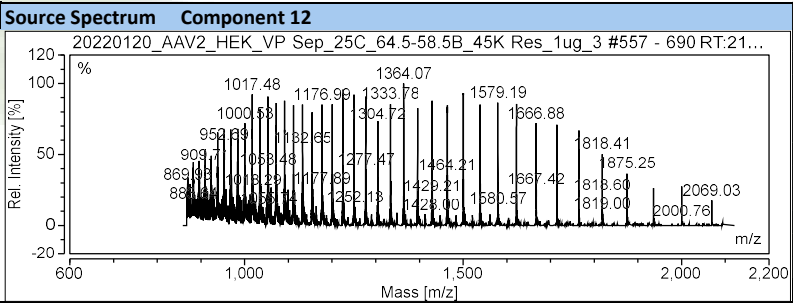
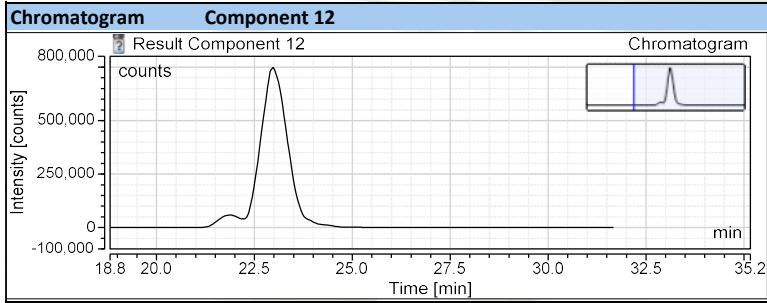
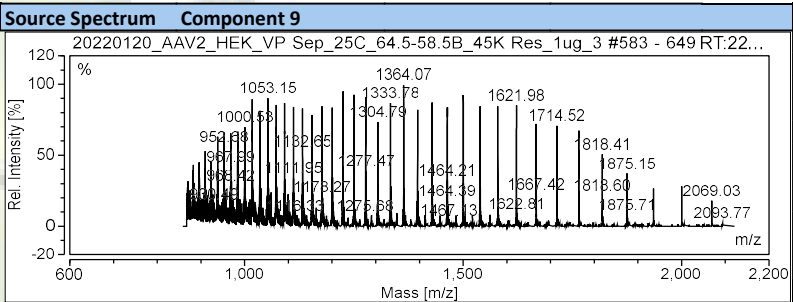
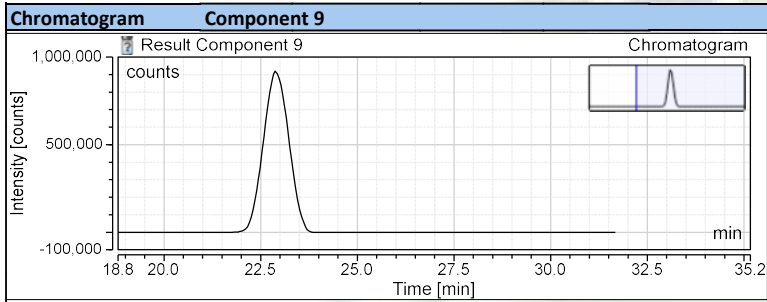
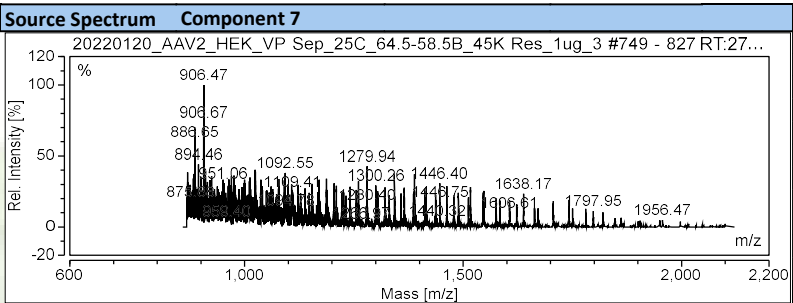
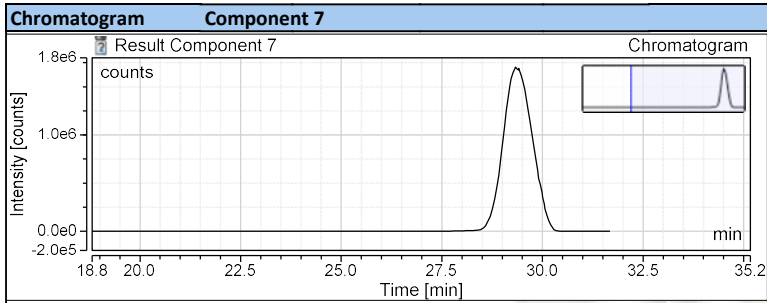
### Protein Deconvolution Summary

#### Injection Details

|                 |   |                      |  |
|-----------------|---|----------------------|--|
| Injection Name: | 20220120_AAV2_HEK_VP Sep_25C_64.5-58.5B_45K Res_1ug_3 | Injection Volume:    | 8.56   |
| Vial Number:    | R:A1  | Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide H      |
| Injection Type: | Unknown   | Processing Method:   | AAV2 MS Quantitative In-depth Characterization |
| Run Time (min): | 45.00   | Injection Date/Time: | 21/Jan/22 09:04                                |







| Result Component   | RT min | Mass Da   | Std. Dev. Da | Std. Dev. ppm | Delta Mass Da | Intensity counts | Fractional Abundance counts | Rel. Abundance % | Number of Charge States | Score  |
|--------------------|--------|-----------|--------------|---------------|---------------|------------------|-----------------------------|------------------|-------------------------|--------|
| Result Component 1 | 23.08  | 59973.436 | 0.84         | 13.99         | 0.000         | 1959279259       | 46.2                        | 100.00           | 41                      | 242.04 |
| Result Component 2 | 23.11  | 59991.819 | 1.81         | 30.24         | 18.383        | 430877006        | 10.2                        | 21.99            | 41                      | 161.22 |
| Result Component 3 | 23.08  | 59953.840 | 1.53         | 25.59         | -19.596       | 302960887        | 7.1                         | 15.46            | 34                      | 154.28 |
| Result Component 4 | 28.00  | 81855.271 | 1.70         | 20.79         | 21881.835     | 149335479        | 3.5                         | 7.62             | 55                      | 180.65 |
| Result Component 5 | 29.37  | 66487.295 | 1.24         | 18.66         | 6513.860      | 115071618        | 2.7                         | 5.87             | 42                      | 195.77 |
| Result Component 6 | 23.08  | 60018.234 | 1.74         | 28.98         | 44.799        | 61338748         | 1.4                         | 3.13             | 25                      | 68.82  |
| Result Component 7 | 23.76  | 60006.121 | 1.86         | 30.94         | 32.686        | 59932437         | 1.4                         | 3.06             | 39                      | 79.35  |
| Result Component 8 | 29.37  | 66504.638 | 1.95         | 29.34         | 6531.202      | 43476745         | 1.0                         | 2.22             | 41                      | 102.72 |
| Result Component 9 | 27.98  | 81872.829 | 2.11         | 25.73         | 21899.393     | 38271649         | 0.9                         | 1.95             | 44                      | 90.47  |

|                      |       |           |      |       |           |          |     |      |    |        |
|----------------------|-------|-----------|------|-------|-----------|----------|-----|------|----|--------|
| Result Component 11  | 22.91 | 59299.871 | 1.48 | 25.01 | -673.565  | 28850826 | 0.7 | 1.47 | 30 | 69.26  |
| Result Component 12  | 22.40 | 59954.253 | 1.72 | 28.73 | -19.183   | 27440115 | 0.6 | 1.40 | 33 | 139.46 |
| Result Component 19  | 27.67 | 81874.453 | 2.73 | 33.39 | 21901.017 | 10900834 | 0.3 | 0.56 | 50 | 109.08 |
| Result Component 77  | 24.00 | 60020.804 | 1.54 | 25.71 | 47.368    | 3356483  | 0.1 | 0.17 | 33 | 78.05  |
| Result Component 152 | 29.15 | 66467.903 | 1.85 | 27.80 | 6494.467  | 1942465  | 0.0 | 0.10 | 19 | 65.67  |



## Component Summary

### Injection Details

|                 |                                     |                      |                                       |
|-----------------|-------------------------------------|----------------------|---------------------------------------|
| Injection Name: | 20220120_AAV2_HEK_VP_Sep_25C_64.5-5 | Injection Volume:    | 8.56                                  |
| Vial Number:    | R:A1                                | Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein     |
| Injection Type: | Unknown                             | Processing Method:   | AAV2 MS Quantitative In-depth Charact |
| Run Time (min): | 45.00                               | Injection Date/Time: | 21/Jan/22 09:04                       |

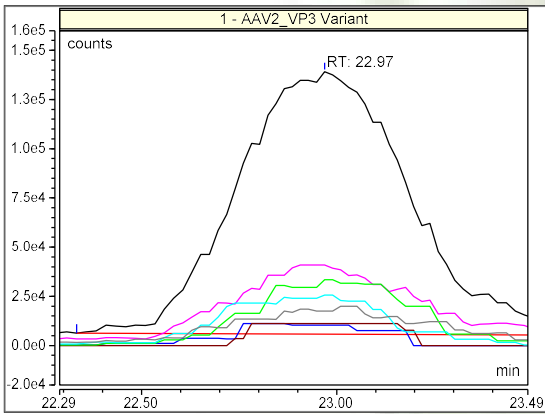
| Peak Name                  | Retention Time<br>min | Area<br>counts*min | Relative Area<br>% | Quantitation Ions<br>m/z                              | Mol.Mass  | Mass Confirmation<br>Pass/Fail |
|----------------------------|-----------------------|--------------------|--------------------|---|-----------|--------------------------------|
| AAV2_VP3 Variant           | 22.969                | 67967.176          | 8.71               | 1447.374,957.488,1380.090,1163.736,1099.120,973.145   | 59301.030 | PASS                           |
| AAV2_VP3[1xOxidation (MW)] | 23.010                | 139936.624         | 17.94              | 1277.428,1396.179,1464.229,1250.823,1305.183,1225.324 | 59989.780 | PASS                           |
| AAV2_VP3[2xOxidation (MW)] | 23.010                | 129323.456         | 16.58              | 1133.193,1092.027,1112.217,1072.520,1539.665,1464.606 | 60005.780 | PASS                           |
| AAV2_VP3                   | 23.052                | 130658.262         | 16.75              | 1364.311,1500.570,1333.966,1395.925,1463.892,1225.374 | 59973.780 | PASS                           |
| AAV2_VP3[1xSuccinimide D]  | 23.135                | 171338.149         | 21.97              | 952.674,923.374,968.020,937.789,1017.182,1000.250     | 59955.770 | PASS                           |
| AAV2_VP1[1xOxidation (MW)] | 28.028                | 38381.637          | 4.92               | 1343.191,1154.150,1388.700,1187.572,1412.610,1170.616 | 81870.890 | PASS                           |
| AAV2_VP1                   | 28.050                | 39880.541          | 5.11               | 1342.896,1388.388,1365.271,1300.301,1187.321,1279.998 | 81854.900 | PASS                           |
| AAV2_VP2[1xOxidation (MW)] | 29.333                | 30324.198          | 3.89               | 1446.760,1305.022,1167.738,1358.263,1147.628,1331.107 | 66503.900 | PASS                           |
| AAV2_VP2                   | 29.355                | 32159.860          | 4.12               | 1446.392,1415.628,1304.683,1478.503,1209.871,1386.167 | 66487.900 | PASS                           |
| <b>100.00</b>              |                       |                    |                    |   |           |                                |

### Component Name: AAV2\_VP3 Variant

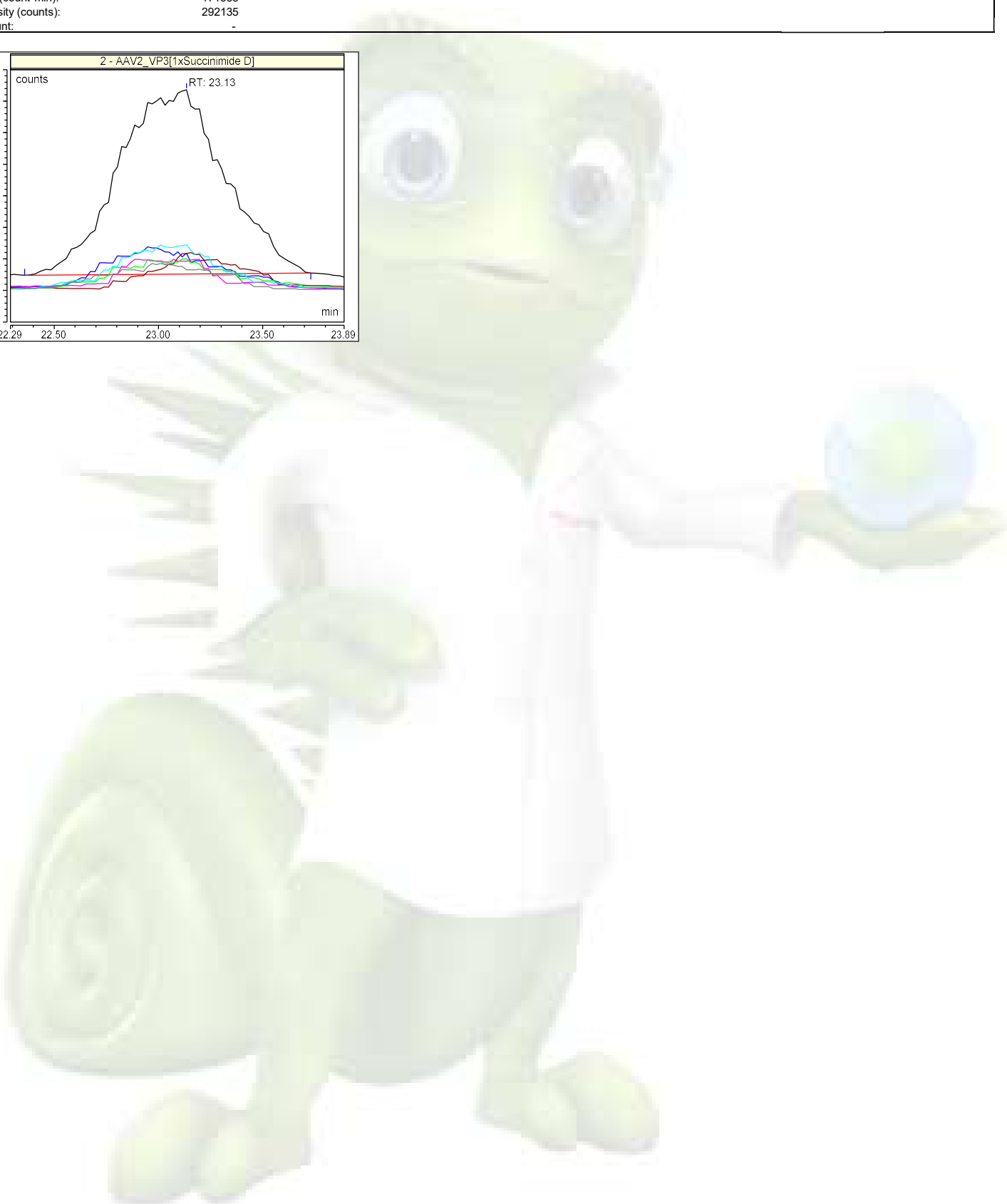
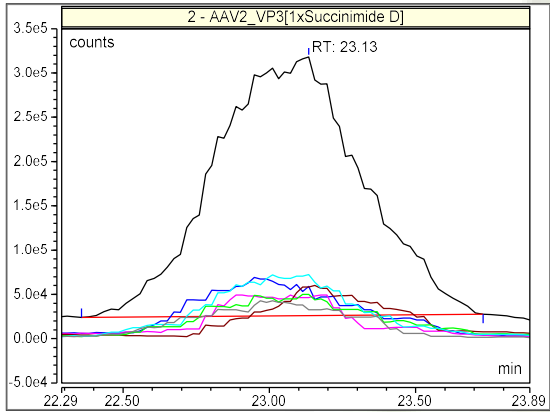
Quantitation ions: 1447.374,957.488,1380.090,1163.736,1099.120,973.145

Retention time (min): 22.89  
Expected Mass: 59301.030

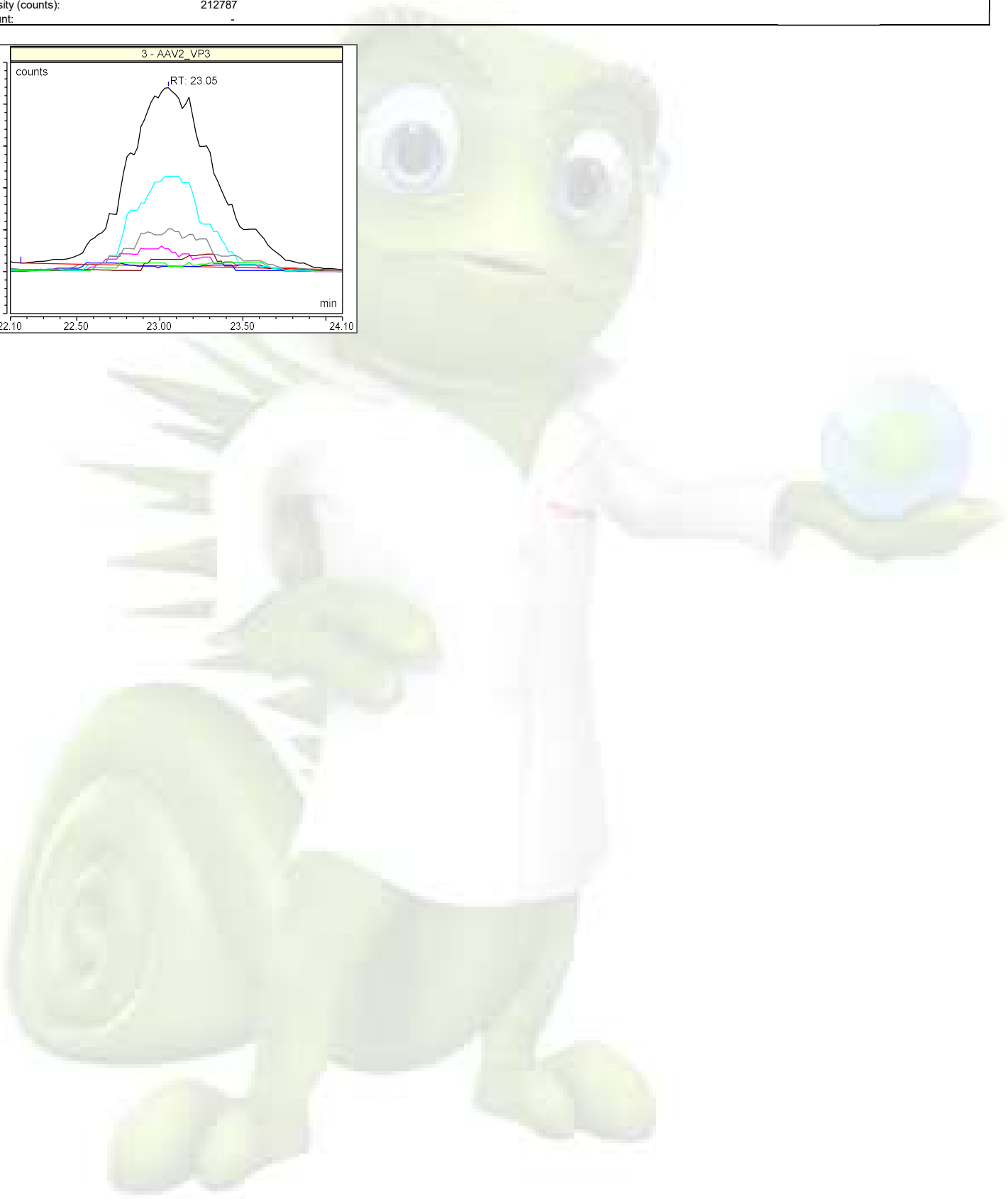
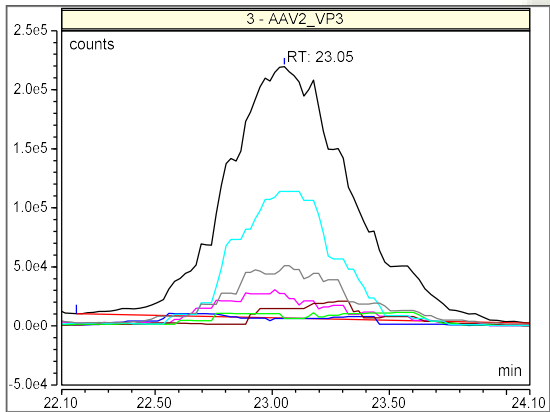
Area (count\*min): 67967  
Intensity (counts): 133312  
Amount: -



|  |   |
|--|---|
| <b>Component Name: AAV2 VP3[1xSuccinimide D]</b> |   |
| Quantitation ions:                               | 952.674,923.374,968.020,937.789,1017.182,1000.250 |
| Retention time (min):                            | 23.09   |
| Expected Mass:                                   | 59955.770   |
| Area (count*min):                                | 171338  |
| Intensity (counts):                              | 292135  |
| Amount:  | -   |



|                                 |   |
|---------------------------------|---|
| <b>Component Name: AAV2 VP3</b> |   |
| Quantitation ions:              | 1364.311,1500.570,1333.966,1395.925,1463.892,1225.374 |
| Retention time (min):           | 23.10   |
| Expected Mass:                  | 59973.780   |
| Area (count*min):               | 130658  |
| Intensity (counts):             | 212787  |
| Amount:                         | -   |



**Component Name: AAV2 VP3[2xOxidation (MW)]**

Quantitation ions: 1133.193,1092.027,1112.217,1072.520,1539.665,1464.606

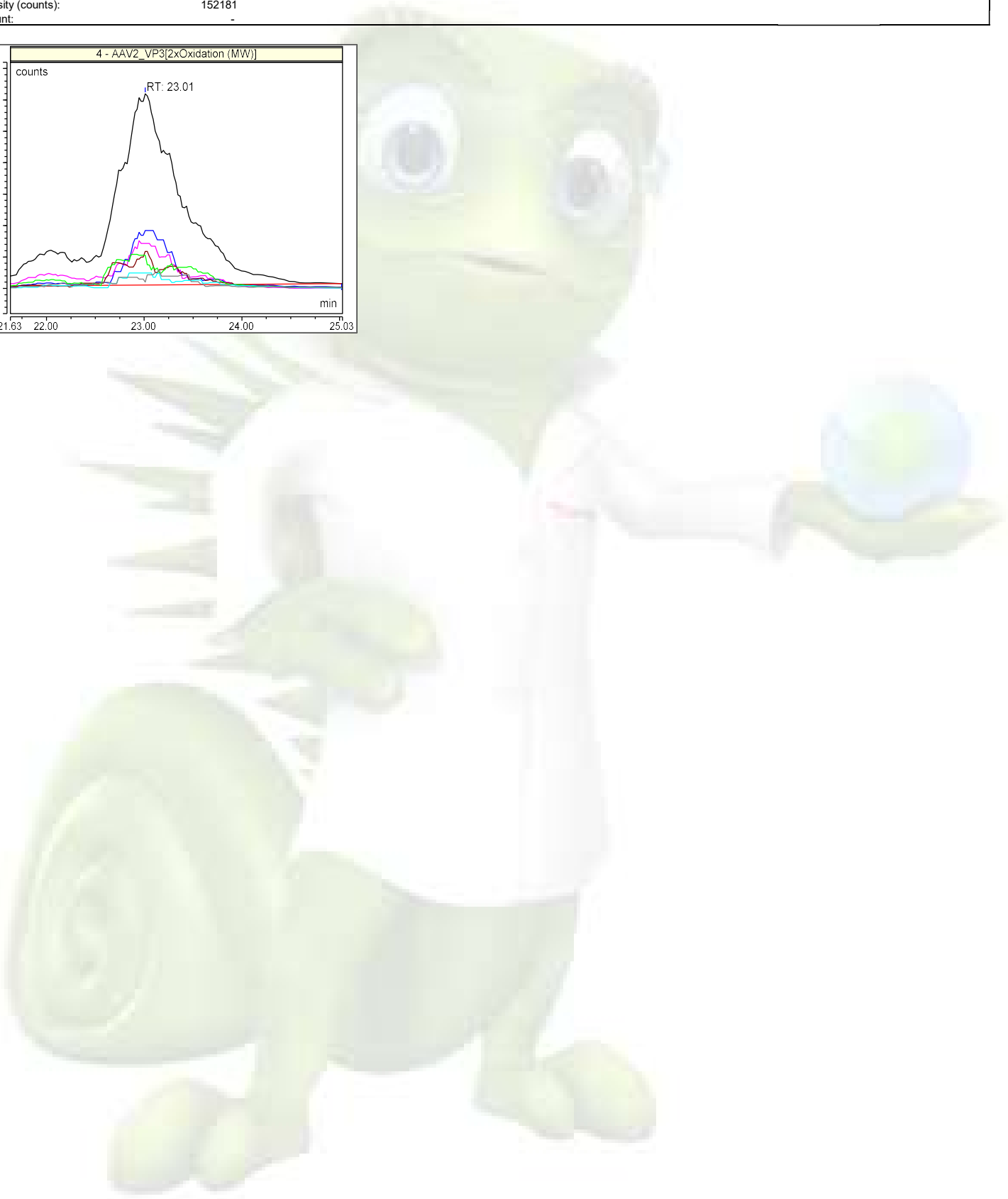
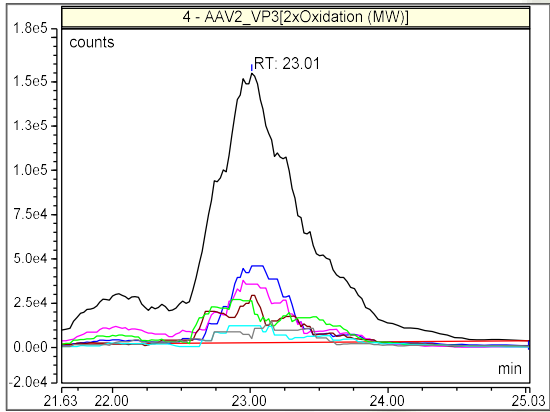
Retention time (min): 23.33

Expected Mass: 60005.780

Area (count\*min): 129323

Intensity (counts): 152181

Amount: -



**Component Name: AAV2 VP3[1xOxidation (MW)]**

Quantitation ions: 1277.428,1396.179,1464.229,1250.823,1305.183,1225.324

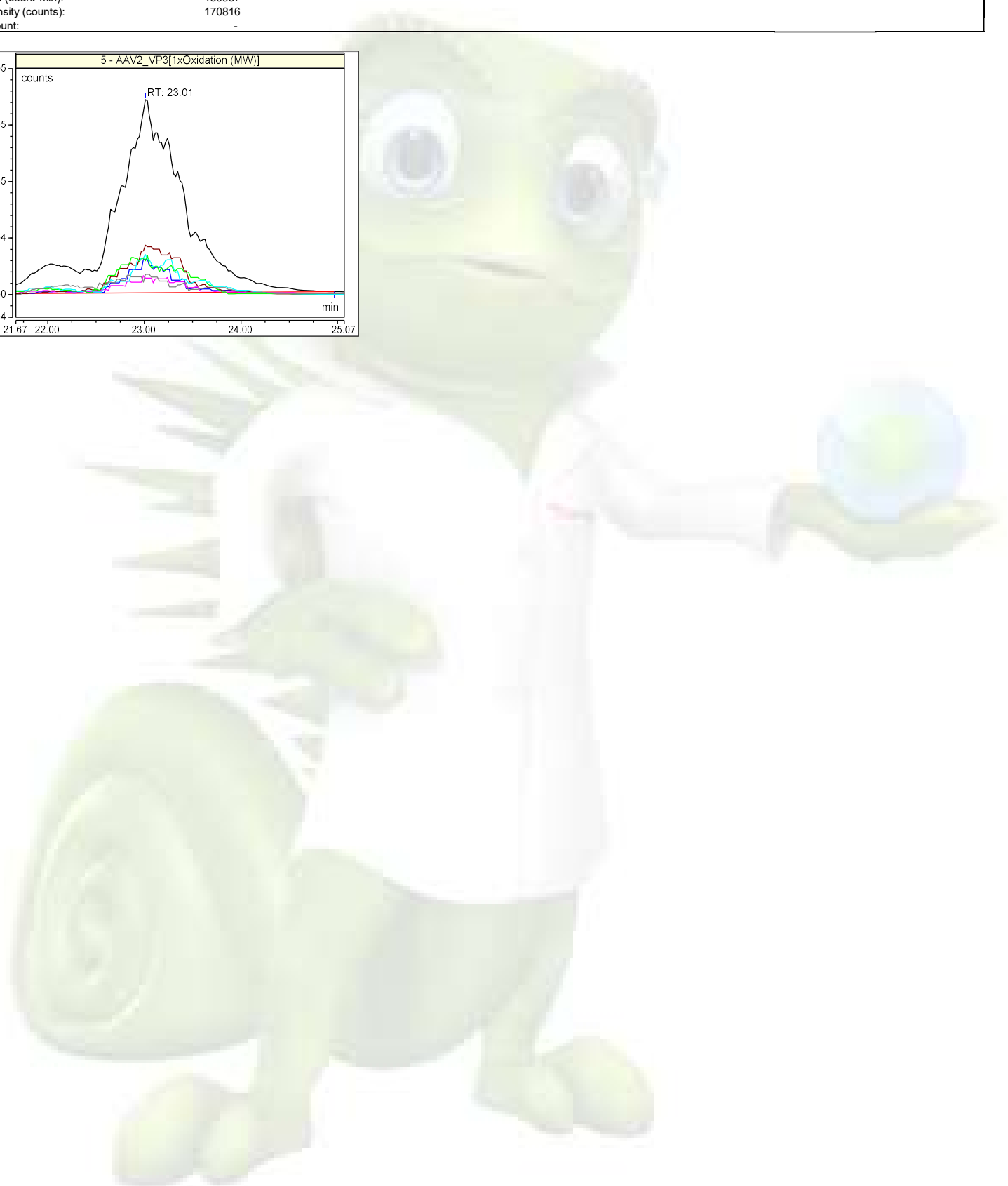
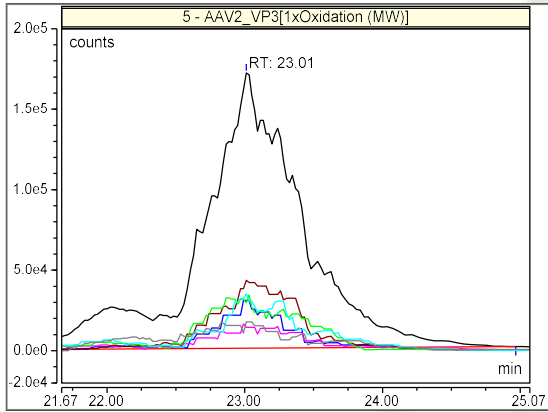
Retention time (min): 23.37

Expected Mass: 59989.780

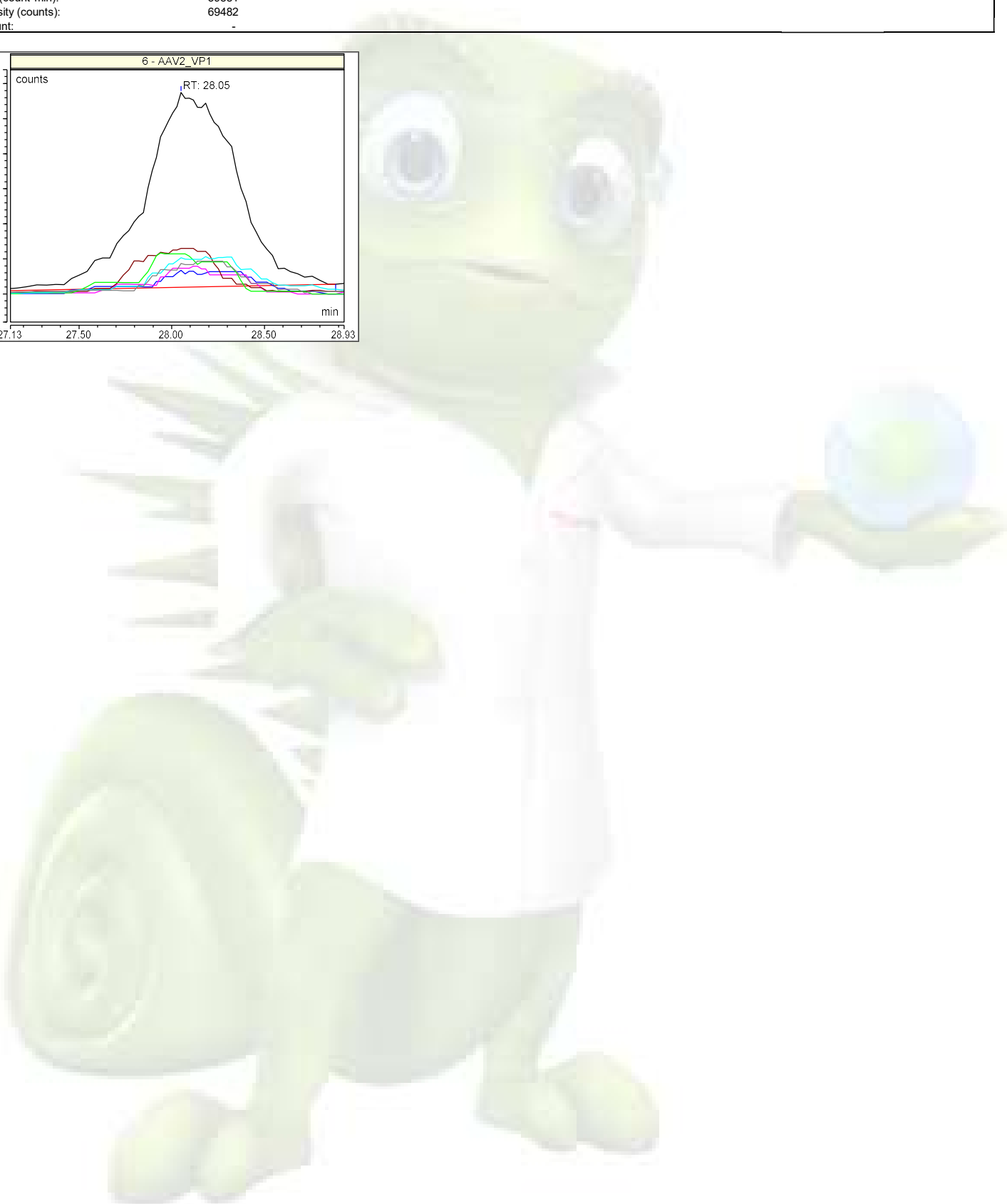
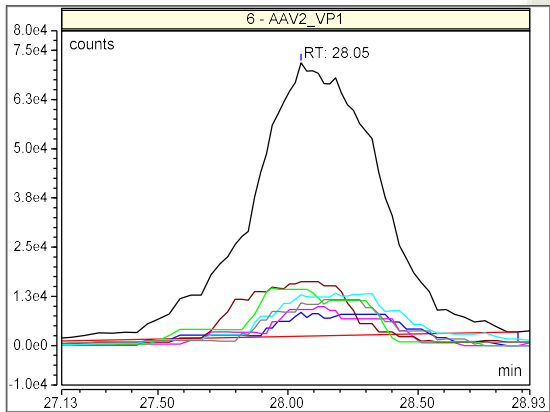
Area (count\*min): 139937

Intensity (counts): 170816

Amount: -



|                                 |   |
|---------------------------------|---|
| <b>Component Name: AAV2 VP1</b> |   |
| Quantitation ions:              | 1342.896,1388.388,1365.271,1300.301,1187.321,1279.998 |
| Retention time (min):           | 28.03   |
| Expected Mass:                  | 81854.900   |
| Area (count*min):               | 39881   |
| Intensity (counts):             | 69482   |
| Amount:                         | -   |





**Component Name: AAV2 VP1[1xOxidation (MW)]**

Quantitation ions: 1343.191,1154.150,1388.700,1187.572,1412.610,1170.616

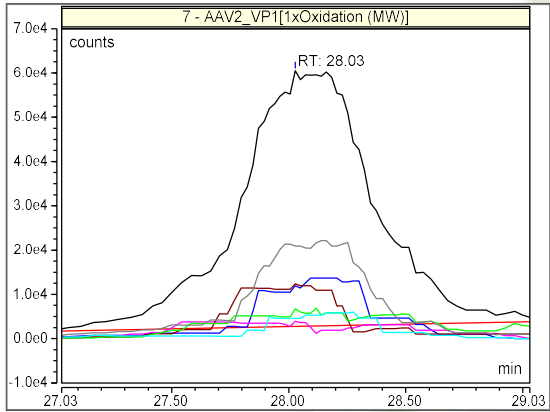
Retention time (min): 28.03

Expected Mass: 81870.890

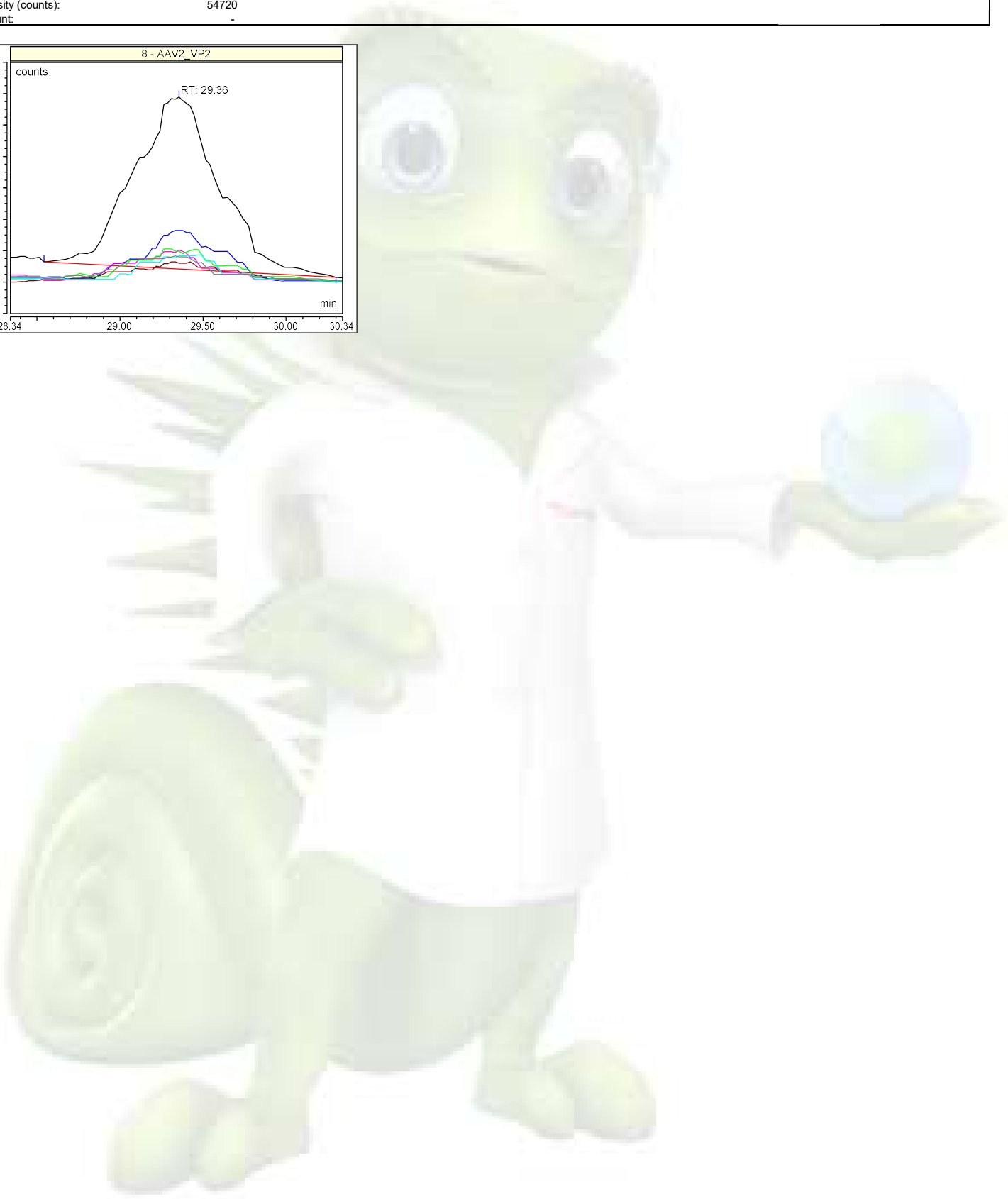
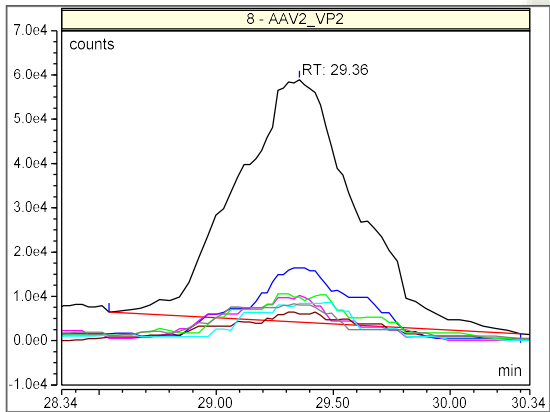
Area (count\*min): 38382

Intensity (counts): 57736

Amount: -



|                                 |  |
|---------------------------------|--|
| <b>Component Name: AAV2 VP2</b> |  |
| Quantitation ions:              | 1446.392, 1415.628, 1304.683, 1478.503, 1209.871, 1386.167 |
| Retention time (min):           | 29.34  |
| Expected Mass:                  | 66487.900  |
| Area (count*min):               | 32160  |
| Intensity (counts):             | 54720  |
| Amount:                         | -  |



**Component Name: AAV2 VP2[1xOxidation (MW)]**

Quantitation ions: 1446.760, 1305.022, 1167.738, 1358.263, 1147.628, 1331.107

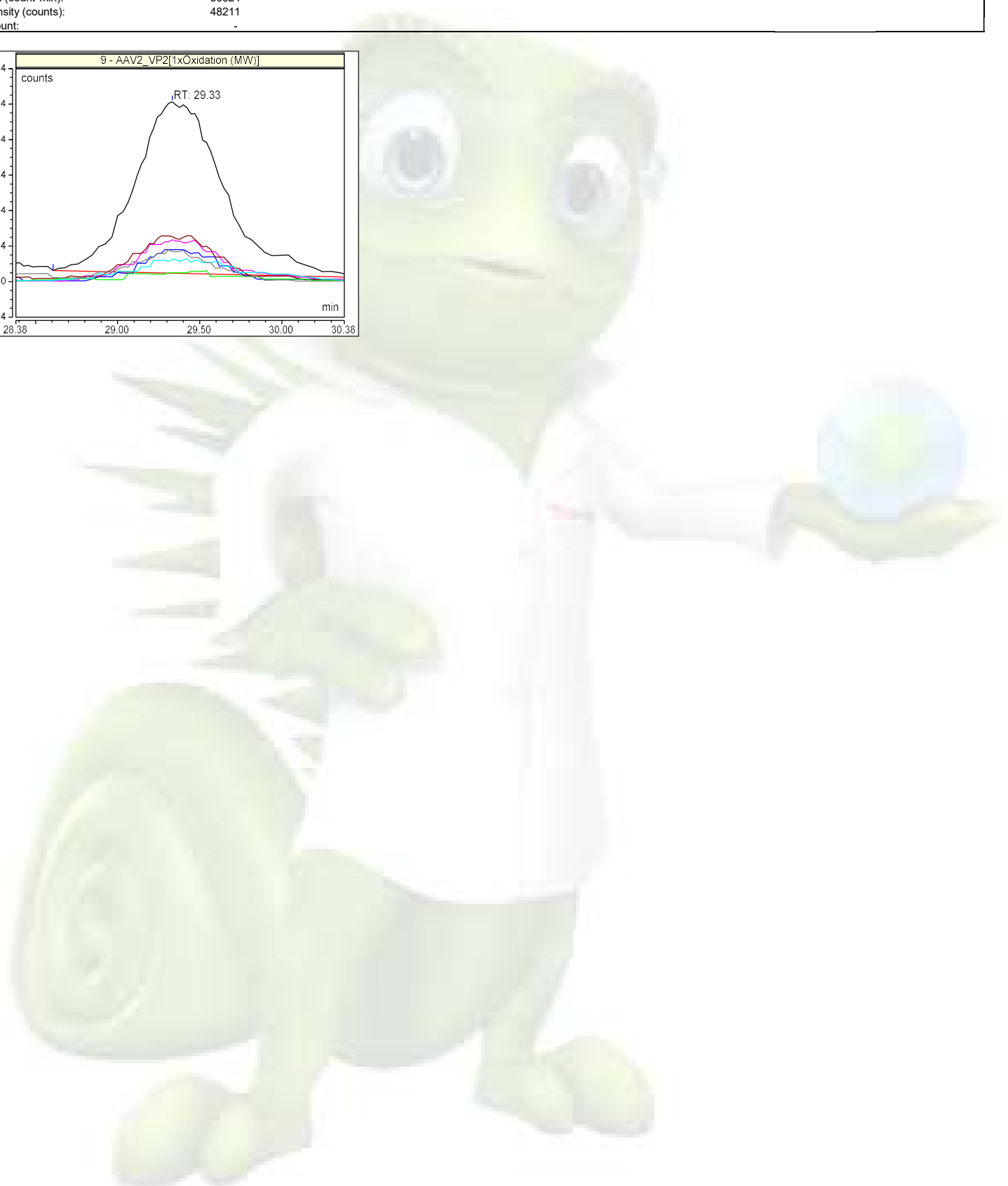
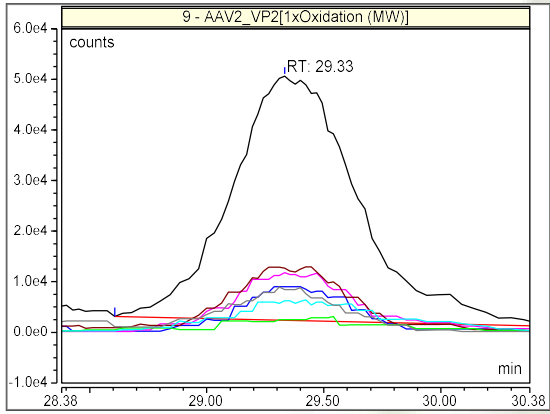
Retention time (min): 29.38

Expected Mass: 66503.900

Area (count\*min): 30324

Intensity (counts): 48211

Amount: -

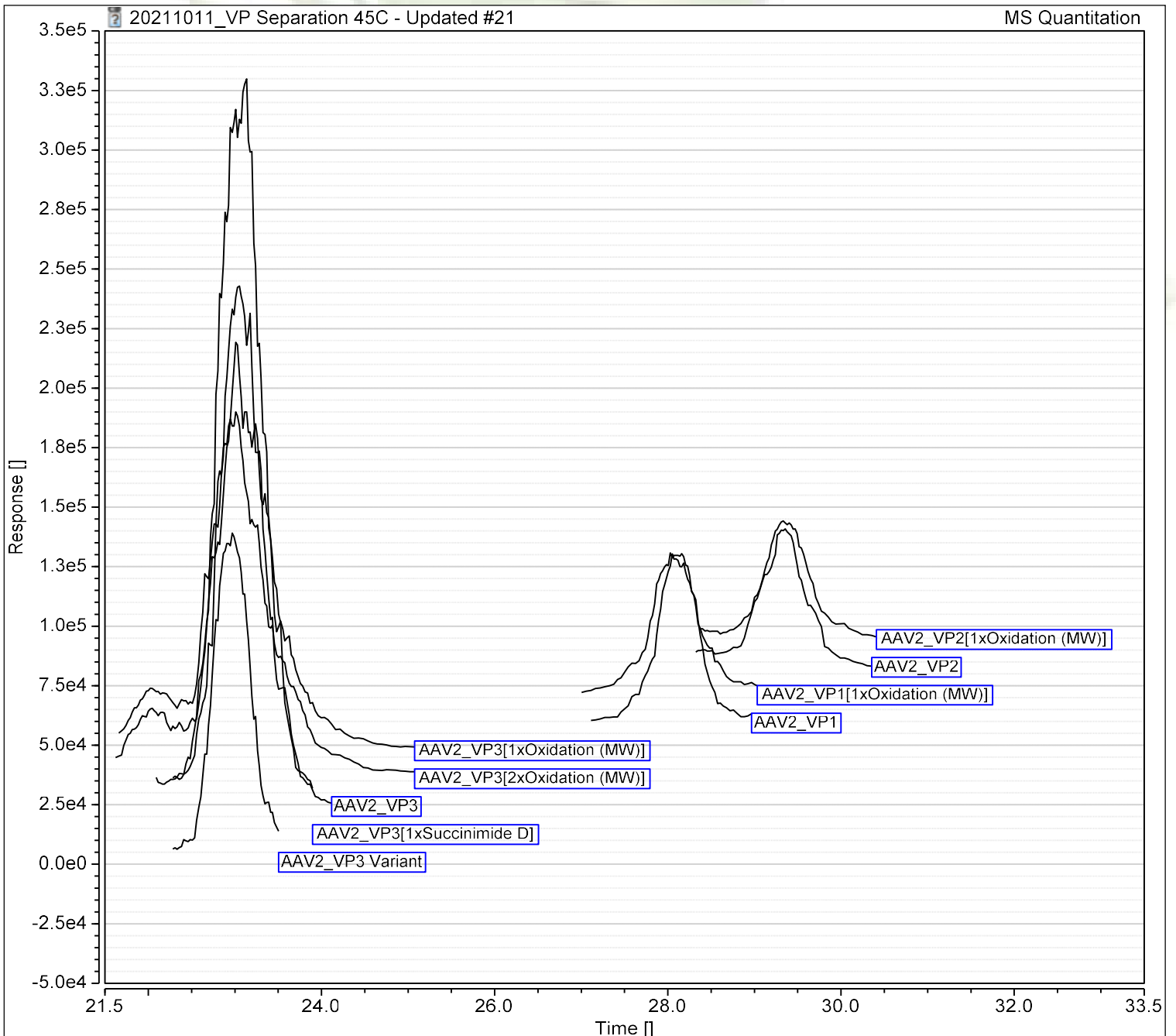


## Chromatogram and SST Results

### Injection Details

|                      |   |                   |        |
|----------------------|---|-------------------|--------|
| Injection Name:      | 20220120_AAV2_HEK_VP Sep_25C_64.5-58.5B_45K R                                 | Run Time (min):   | 45.00  |
| Vial Number:         | R:A1  | Injection Volume: | 8.56   |
| Injection Type:      | Unknown   |                   |        |
| Calibration Level:   |   |                   |        |
| Instrument Method:   | AAV Cap_Prot_Sep_BEH Glycoprotein Amide HILIC_1x150mm_FLR + MS_45K Res_CT25C_ |                   |        |
| Processing Method:   | AAV2 MS Quantitative In-depth Characterization                                | Dilution Factor:  | 1.0000 |
| Injection Date/Time: | 21/Jan/22 09:04   | Sample Weight:    | 1.0000 |

### Chromatogram



### SST Results

| No.                                 | Name | Inj. Condition | Peak          | Test Result | Injection |
|-------------------------------------|------|----------------|---------------|-------------|-----------|
| Number of executed test cases: n.a. |      |                | Total Result: | Passed      |           |

