*Hypothesis I Results: Growth in carbon-stressed environments*

| Bacteria strain | | Replicates (n) | Molecules of cobalamin per cell | | | |
|---|---|---|---|---|---|---|
| | | | Methylcobalamin | Adenosylcobalamin | Hydroxocobalamin | Combined |
| *R. pomeroyi DSS-3* | High carbon | 3 | Not detected | 444 ± 33 | 52 ± 27 | 248 ± 216 |
| | Low carbon | 3 | Not detected | 366 ± 68 | 227 ± 142 | 297 ± 126 |
| *Sulfitobacter sp. SA11* | High carbon | 3 | 2.25 ± 0.64 | 10 ± 2 | **5 ± 0.6** | 6 ± 4 |
| | Low carbon | 3 | Not detected | 8 ± 0.2 | **3 ± 0.2** | 6 ± 3 |

Table 1. Observed molecules per cell of three major types of cobalamin with standard deviations, as tested by the first hypothesis. Bolded/underlined figures refer to those observations that were statistically significant.

*Hypothesis II Results: Enriched medias*

| Bacteria strain | | Replicates (n) | Molecules of cobalamin per cell | | | |
|---|---|---|---|---|---|---|
| | | | Methylcobalamin | Adenosylcobalamin | Hydroxocobalamin | Combined |
| *R. pomeryoi DSS-3* | S-adenosylmethionine enriched media | 3 | Not detected | **310 ± 27** | 485 ± 394* | 398 ± 255* |
| | S-adenosylhomocysteine enriched media | 3 | Not detected | **302 ± 43** | 666 ± 400* | 484 ± 323* |
| | Unenriched seawater tryptone | 3 | Not detected | **235 ± 17** | 212 ± 72 | 224 ± 48 |
| *Sulfitobacter sp. SA11* | S-adenosylmethionine enriched media | 3 | Not detected | 5 ± 0.1 | 5 ± 2.1 | 5 ± 1 |
| | S-adenosylhomocysteine enriched media | 3 | Not detected | 6 ± 1 | Not detected | 6 ± 1 |
| | Unenriched seawater tryptone | 3 | Not detected | 20 ± 24 | Not detected | 20 ± 24 |

Table 2. Observed molecules per cell of three major types of cobalamin with standard deviation, as tested by the second hypothesis. Bolded/underlined figures refer to those observations that were statistically significant. *These estimates of molecules per cell reflects an average of three replicates, two of which fell below the detection limit on the LCMS. These figures may actually be much higher, and will require a repeated experiment to resolve.

*Hypothesis I Statistics*

| Bacteria strain | | Cobalamin type | | | |
|---|---|---|---|---|---|
| | | Methylcobalamin | Adenosylcobalamin | Hydroxocobalamin | Combined |
| *R. pomeryoi DSS-3* | High/low carbon | N/A | Insignificant difference | Insignificant difference | Insignificant difference |
| *Sulfitobacter sp. SA11* | High/low carbon | N/A | Insignificant difference | Significant difference | Insignificant difference |

Table 3. Statistical analysis of results for Hypothesis I. Average cobalamin molecules/cell are compared using t-tests between high and low carbon conditions for each species and cobalamin type, as well as pooled cobalamin comparisons.

*Hypothesis II Statistics*

| | | Cobalamin Type | | | |
|---|---|---|---|---|---|
| Bacteria strain | | Methylcobalamin | Adenosylcobalamin | Hydroxocobalamin | Combined |
| *R. pomeryoi DSS-3* | SAM enriched media, SAH enriched media, Seawater Tryptone | N/A | Significant differences between overall results.<br><br>SAM = SAH<br>SAH ≠ SWT<br>SAM ≠ SWT | Insignificant differences between overall results.<br><br>SAM = SAH<br>SAH = SWT<br>SAM = SWT | Insignificant differences between overall results.<br><br>SAM = SAH<br>SAH = SWT<br>SAM = SWT |
| *Sulfitobacter sp. SA11* | SAM enriched media, SAH enriched media, Seawater Tryptone | N/A | Insignificant differences between overall results.<br><br>SAM = SAH<br>SAH = SWT<br>SAM = SWT | N/A | Insignificant differences between overall results.<br><br>SAM = SAH<br>SAH = SWT<br>SAM = SWT |

Table 4. Statistical analysis of results for Hypothesis II. Average cobalamin molecules/cell are compared using ANOVA-F tests between SAM, SAH and SWT conditions for each species and cobalamin type, as well as pooled cobalamin comparisons.

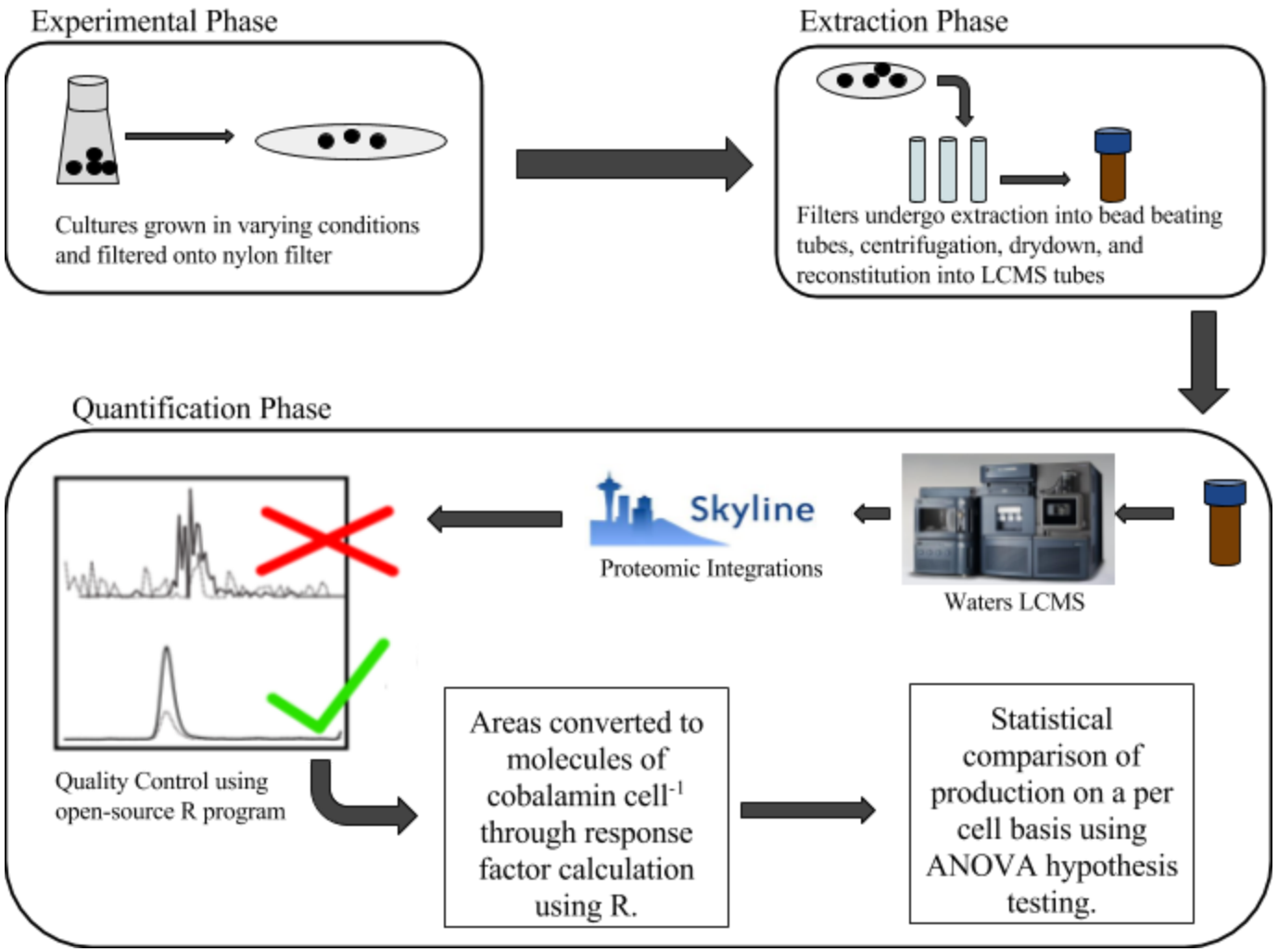


Fig 1. Methods flowchart.

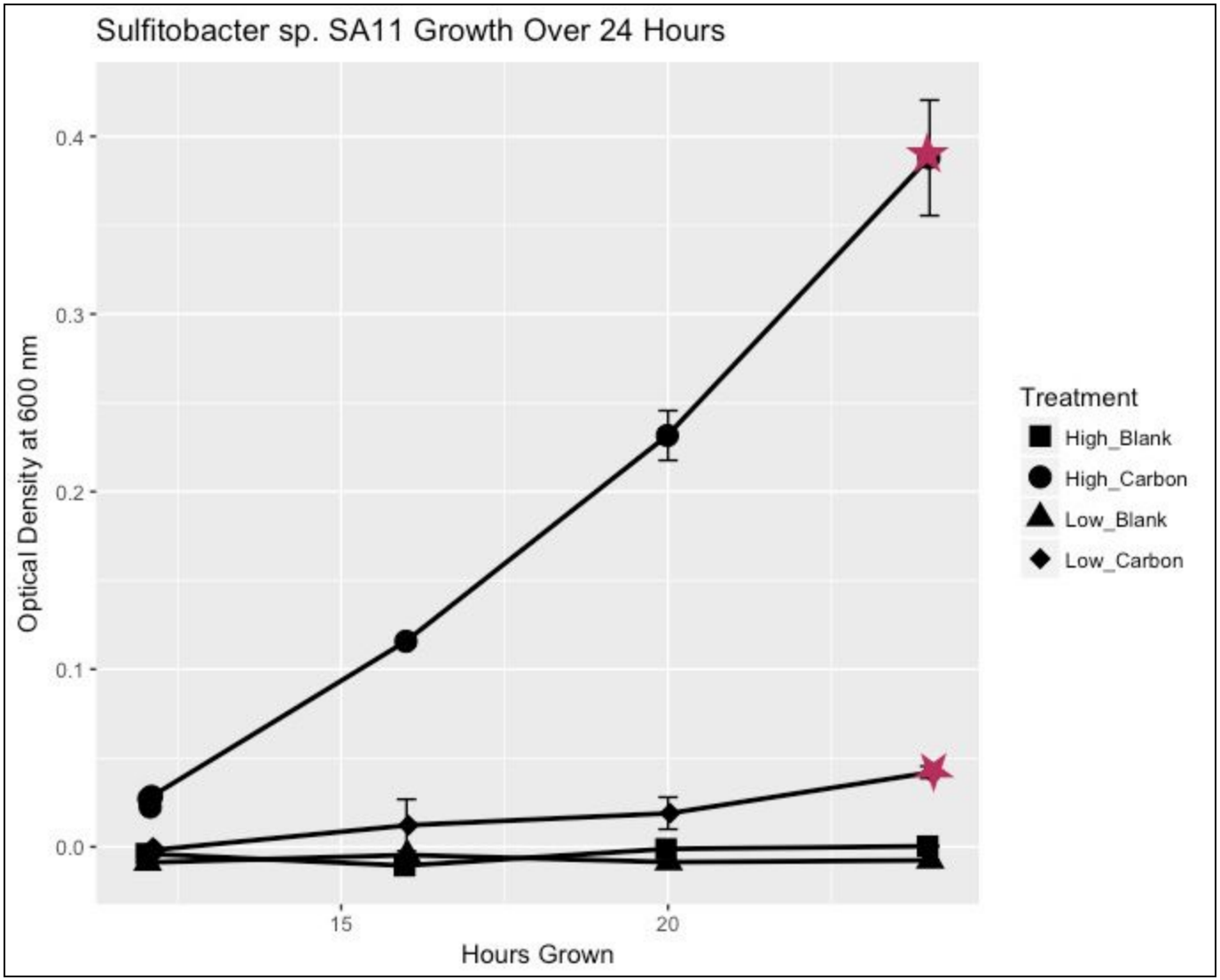


Fig 2. Optical density of *Sulfitobacter sp. SA11* at 600 nm, grown in high and low carbon conditions over a period of 24 hours. Blanks are included for comparison. Pink stars mark point of harvesting.

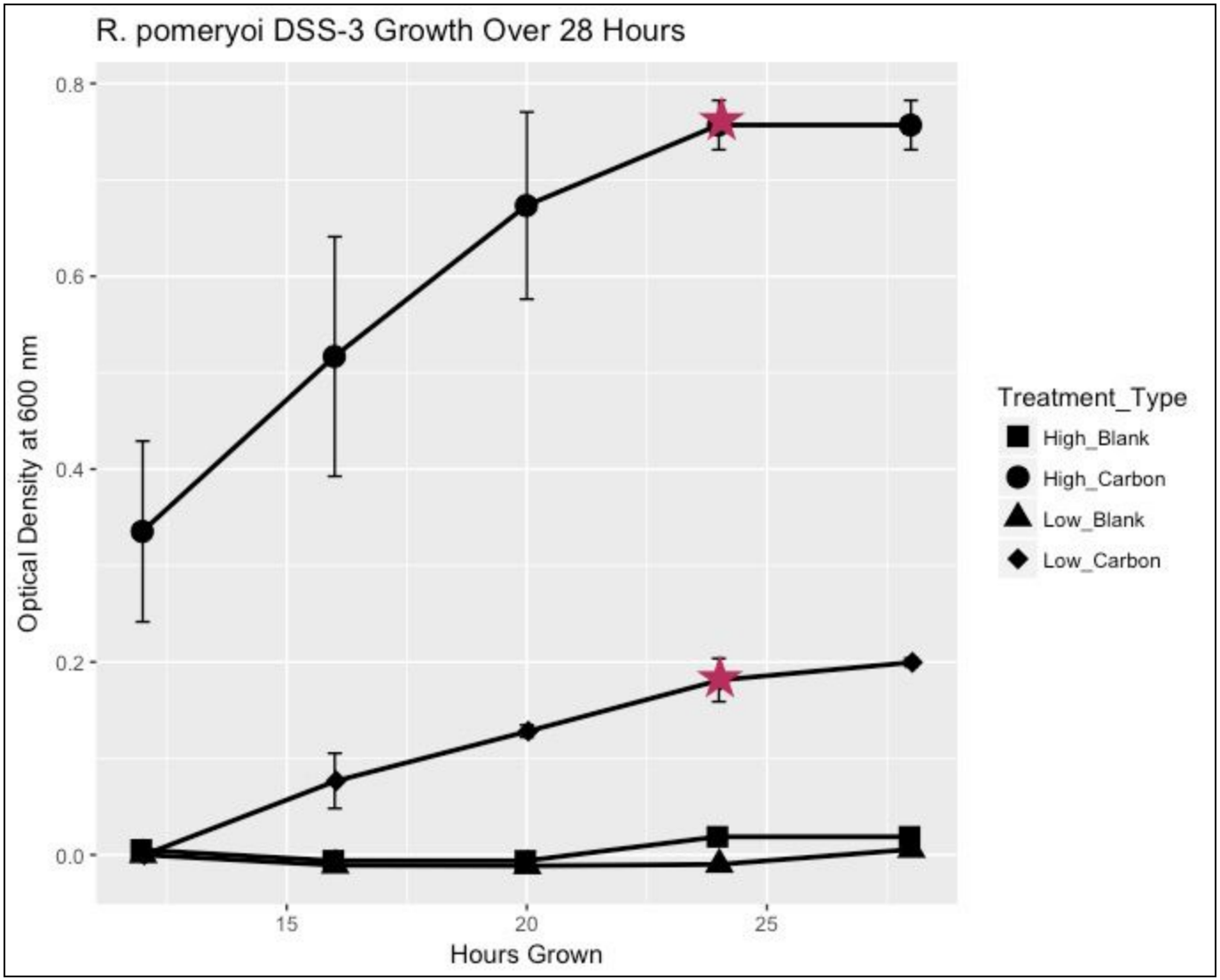


Fig 3. Optical density of *R. pomeryoi DSS-3* at 600 nm, grown in high and low carbon conditions over a period of 28 hours. Blanks are included for comparison. Pink stars mark harvesting points.

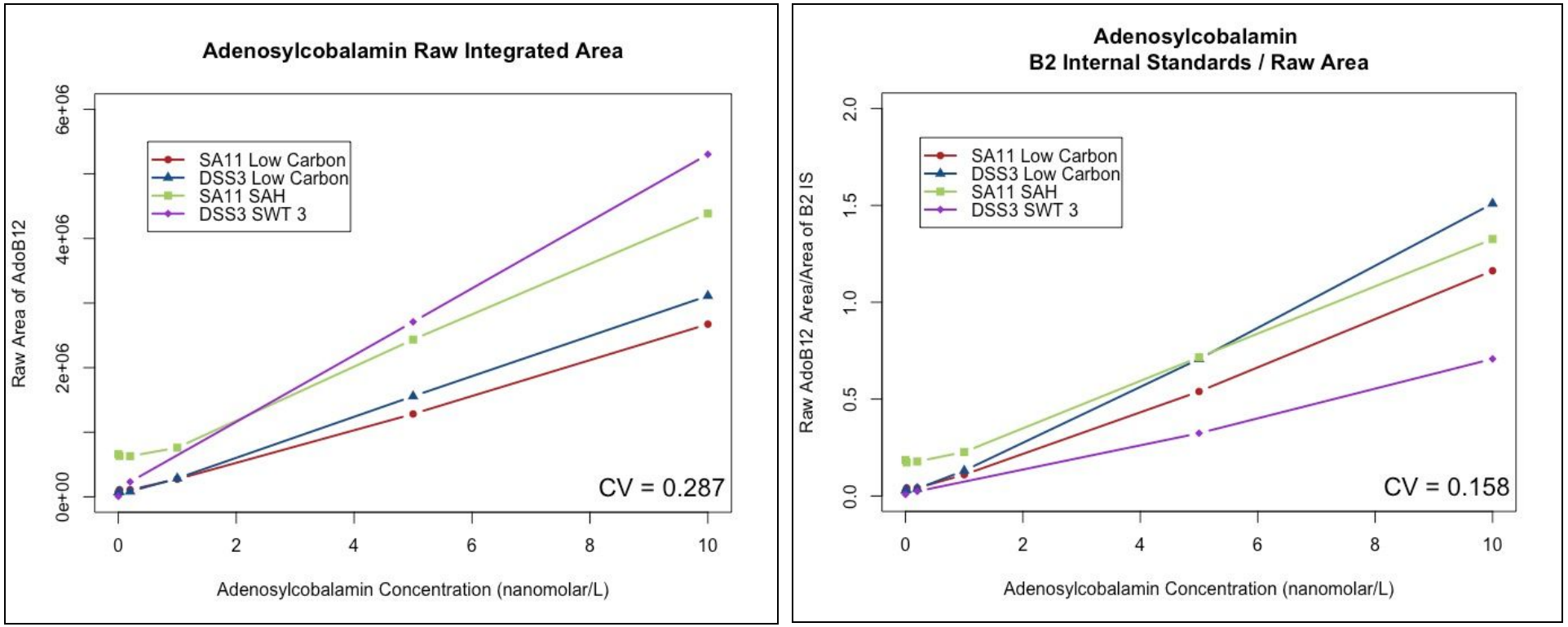


Fig 4. Comparison of the coefficient of variation (CV) between adenosylcobalamin raw area response factor and response factors modified by internal standard areas.

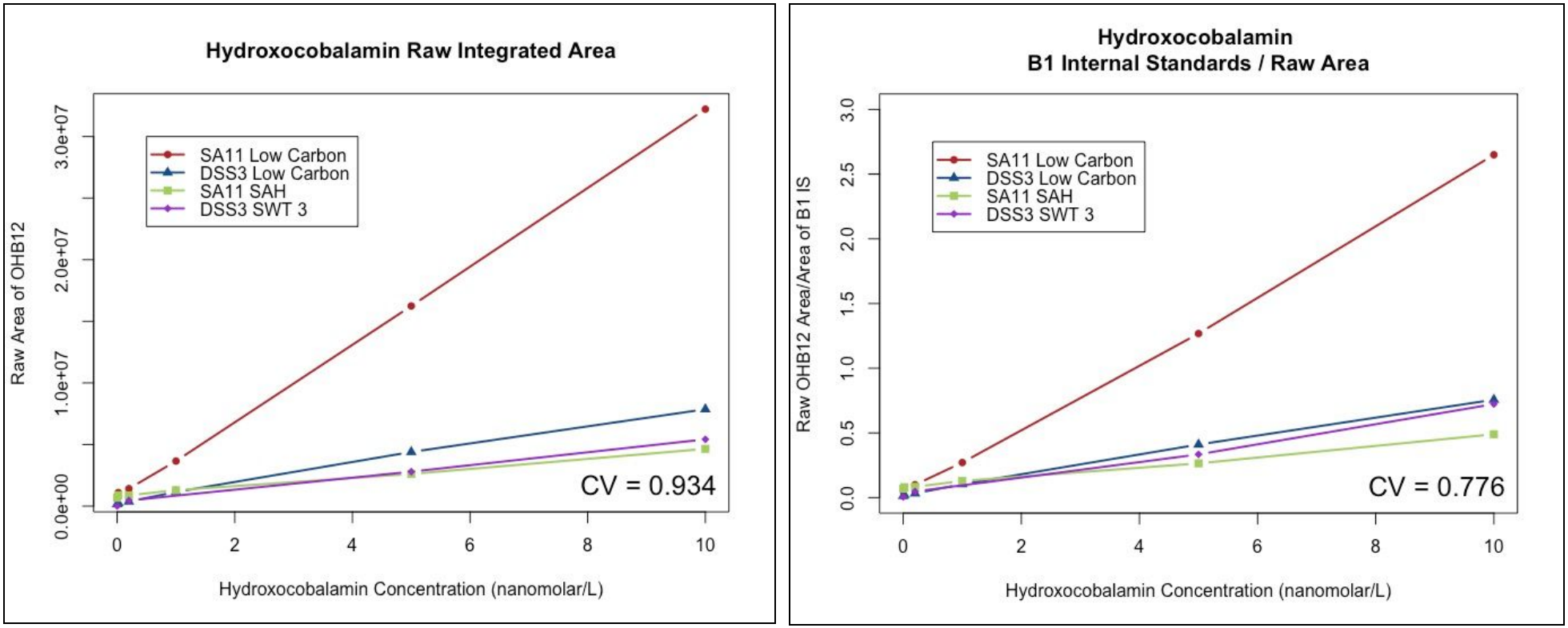


Fig 5. Comparison of the coefficient of variation (CV) between hydroxocobalamin raw area response factor and response factors modified by internal standard areas.