The role of FLI-1-EWS, a fusion gene reciprocal to EWS-FLI-1, in Ewing sarcoma

**Supplementary Materials**

**Table S1** Quantitative RT-PCR analysis of EWS-FLI-1, FLI-1-EWS, EWS, and FLI-1 expression in A673 cells.

|  |  |  |
| --- | --- | --- |
|  | average\* | STD (standard deviation) |
| EWS-FLI-1 | 0.416493 | 0.032531 |
| FLI-1-EWS | 0.002903 | 0.000302 |
| EWS | 1.237203 | 0.153295 |
| FLI-1 | 0.002466 | 0.000193 |

\*Average of triplicate measurements normalized against RNA polymerase II transcript levels.

**Table S2** Quantitative RT-PCR analysis of FLI-1 expression in human mesenchymal stem cells vs. A673 cells.

|  |  |  |
| --- | --- | --- |
|  | average\* | STD |
| MSC FLI-1 | 0.985257 | 0.086769 |
| A673 FLI-1 | 0.002119 | 0.001002 |

\*Average of triplicate measurements normalized against RNA polymerase II transcript levels

**Table S3** Quantitative RT-PCR analysis of EWS-FLI-1, FLI-1-EWS, EWS, and FLI-1 expression in Ewing sarcoma tumors.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **FNY** |  | **FXU** |  | **FNZ** |  | **FXZ** |  |
|  | average\* | STD | average\* | STD | average\* | STD | average\* | STD |
| EWS-FLI-1 | 1.146272 | 0.061594 | 1.205752 | 0.017396 | 1.200642 | 0.091347 | 0.759027 | 0.113066 |
| FLI-1-EWS | 0.030707 | 0.002039 | 0.004003 | 0.001541 | 0.007592 | 0.002824 | 0.025450 | 0.006256 |
| EWS | 1.163902 | 0.096999 | 1.115651 | 0.060653 | 1.750794 | 0.221265 | 2.176045 | 0.083319 |
| FLI-1 | 0.466359 | 0.039897 | 0.087819 | 0.014144 | 1.212170 | 0.104840 | 1.169748 | 0.298578 |

\*Average of triplicate measurements normalized against RNA polymerase II transcript levels.

**Table S4** List of genes displaying differential expression upon FLI-1-EWS silencing.

419 differentially expressed genes derived from DESeq algorithm, with RPKM of one condition (luciferase shRNA-expressing sample or FLI-1-EWS shRNA-expressing sample) > 1, absolute Log2-FoldChange > 1, and *p*-value < 0.05. Columns are 1) Gene Symbol, 2) baseMean (mean normalized counts of luciferase shRNA-expressing sample and FLI-1-EWS shRNA-expressing sample), 3) baseMean1 (normalized counts of luciferase shRNA-expressing sample), 4) baseMean2 (normalized counts of FLI-1-EWS shRNA-expressing sample), 5) fold-change (the ratio of baseMean2 to baseMean1), 6) Log2-FoldChange, 7) *p*-value (*p*-value for the statistical significance of the gene expression change), and 8) adj. *p*-value (*p*-value adjusted for multiple testing with the Benjamini-Hochberg procedure). The table is pre-sorted by *p*-values.

**Table S5** Quantitative RT-PCR analysis of EWS-FLI-1, FLI-1-EWS, EWS, and FLI-1 expression in human mesenchymal stem cells infected with lentiviruses expressing EWS-FLI-1 and/or FLI-1-EWS.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **vector** |  | **EWS-FLI-1** |  | **EWS-FLI-1****+****FLI-1-EWS** |  | **FLI-1-EWS** |  |
|  | average\* | STD | average\* | STD | average\* | STD | average\* | STD |
| EWS-FLI-1 | NA | NA | 0.740486 | 0.009417 | 0.834132 | 0.056353 | NA | NA |
| FLI-1-EWS | NA | NA | NA | NA | 0.009797 | 0.000799 | 0.013017 | 0.001496 |
| EWS | 1.417851 | 0.083748 | 1.461751 | 0.064151 | 1.311592 | 0.013690 | 1.505269 | 0.112527 |
| FLI-1 | 0.437177 | 0.011827 | 0.362344 | 0.022263 | 0.391498 | 0.018944 | 0.375621 | 0.025337 |

\*Average of triplicate measurements normalized against RNA polymerase II transcript levels.