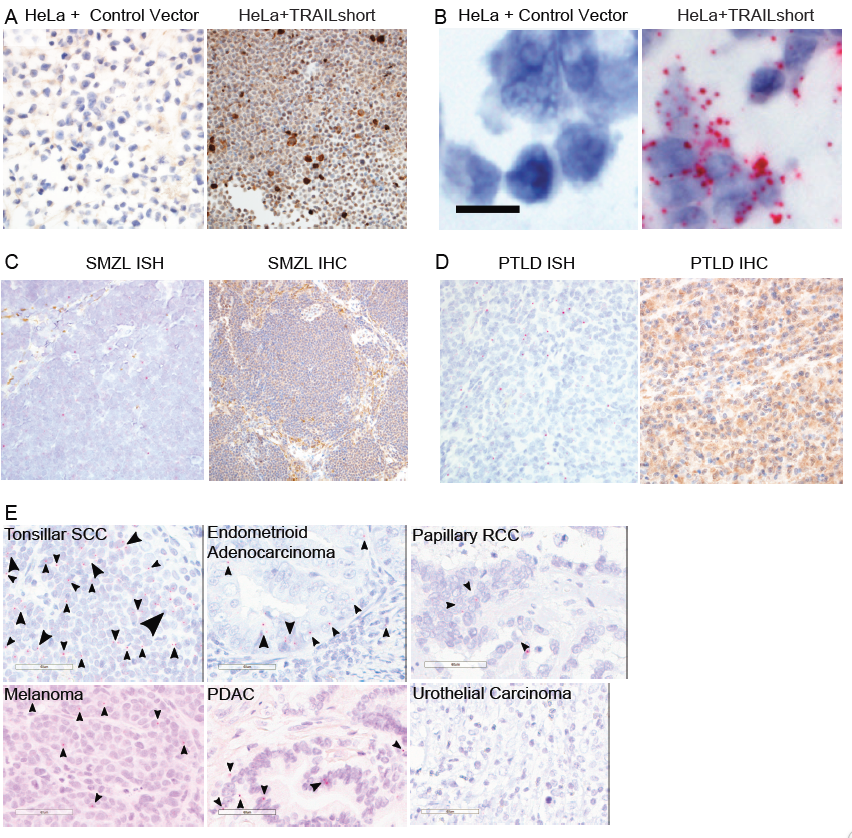
|  |  |
| --- | --- |
| *token* | *Enrichment Score TRAILshort* |
| *severe\_dengue* | 323 |
| *dengue\_fever* | 274.1124 |
| *clear\_cell\_renal\_carcinoma* | 109.0092 |
| *papillary\_renal\_cell\_carcinoma* | 88.88715 |
| *prostate\_adenocarcinoma* | 75.94247 |
| *ductal\_carcinoma* | 75.83678 |
| *cervical\_squamous\_cell\_carcinoma* | 72.38082 |
| *squamous\_cell\_carcinoma* | 68.17859 |
| *diarrhea* | 64.0066 |
| *lung\_squamous\_cell\_carcinoma* | 62.43683 |
| *lobular\_carcinoma* | 36.71969 |
| *Ltbi (latent tuberculosis infection)* | 36.7162 |
| *renal\_cell\_carcinoma* | 23.36674 |
| *asthmatic* | 21.58051 |
| *influenza* | 19.54017 |
| *breast\_cancer* | 17.8911 |
| *ulcerative\_colitis* | 15.7166 |
| *glioblastoma* | 15.7166 |
| *hepatocellular\_carcinoma* | 15.7166 |
| *endometrial\_adenocarcinoma* | 14.59189 |
| *oral\_squamous\_cell\_carcinoma* | 14.28489 |
| *prostate\_cancer* | 13.5306 |
| *brucellosis* | 12.04984 |
| *systemic\_lupus\_erythematosus\_sle* | 10.69982 |
| *lupus* | 10.185 |

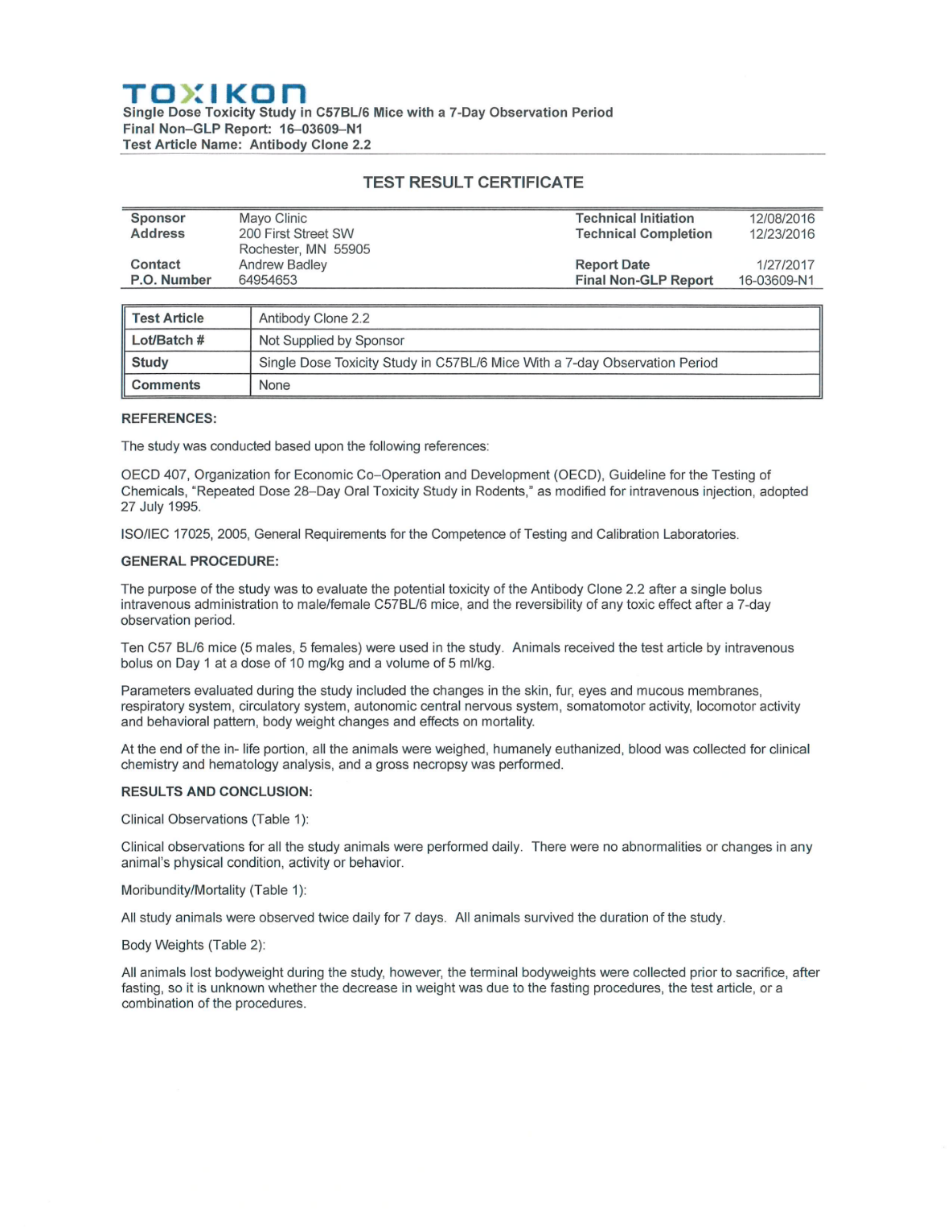
**Supplemental Table S1. Results of TRAILshort enrichment scores in Gene Expression Omnibus datasets by natural language processing derived disease term.** To determine disease enrichments for each transcript, the top expressing samples were compared to the lowest expressers for disproportionate representation of disease phenotypes. The final output was an enrichment score (reported as -log(p-val)) for each disease phenotype. Represented are the disease phenotypes with a TRAILshort enrichment score >10.

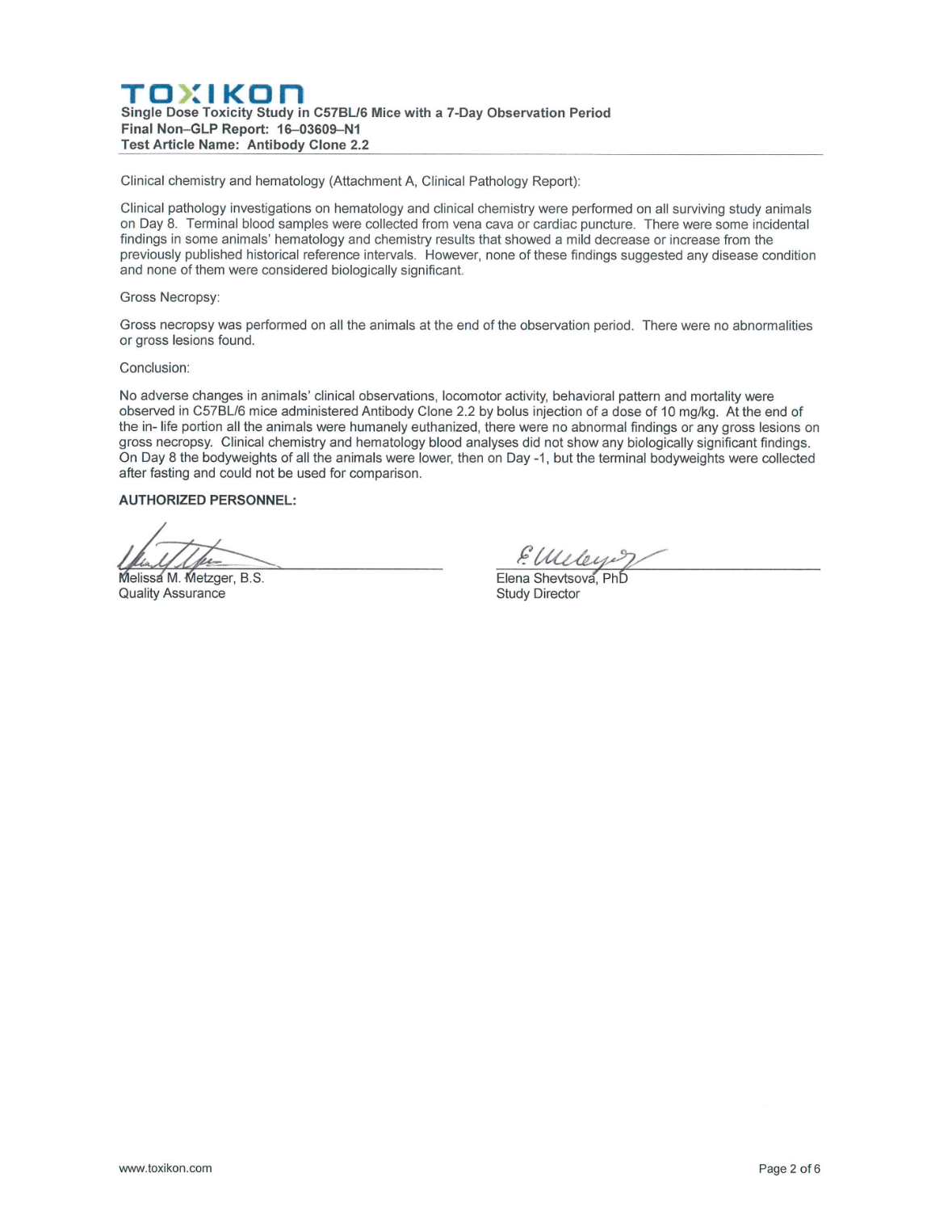


**Supplemental Figure S1. Primary human malignancies express TRAILshort protein and contain TRAILshort mRNA.** The indicated tissues were stained by immunohistochemistry (IHC) for TRAILshort protein expression (brown staining), or by *in situ* hybridization (ISH) for TRAILshort mRNA expression (red dot-like staining with black arrowheads). (A) Parental HeLa cells were negative for TRAILshort protein by IHC (left, 400X original magnification), whereas transfected HeLa cells show strong expression of TRAILshort protein (right, 200X original magnification). (B) RNA ISH within the TRAILshort transfected HeLa cells (right, 400X original magnification). Cells transfected with the empty EGFPC1 vector (left, 200X original magnification) are negative. (C-D) Primary samples of splenic marginal zone lymphoma (SMZL) and post-transplant lymphoproliferative disorder (PTLD) tissue reveal TRAILshort message by ISH and TRAILshort protein by IHC. (E) Tonsillar squamous cell carcinoma, melanoma, papillary renal cell carcinoma, endometrioid endometrial carcinoma and pancreatic ductal adenocarcinoma (PDAC) all were positive for TRAILshort mRNA. Red dots indicate TRAILshort mRNA within the various neoplastic cells. In contrast, the representative urothelial carcinoma was negative for TRAILshort mRNA. Scale bar = 60 µm for all panels. (F) TRAILshort mRNA expression was positive by ISH in two representative hematologic malignancies.

|  |  |  |
| --- | --- | --- |
| **Tumor Type** | **Number of Cases Tested by ISH** | **Positive Cases for TRAILshort RNA ISH** |
| Pancreatic Ductal Adenocarcinoma | 9 | 5 |
| Hepatocellular Carcinoma | 15 | 1 |
| Oropharyngeal Squamous Cell Carcinoma | 7 | 5 |
| Tonsil Squamous Cell Carcinoma | 3 | 3 |
| Melanoma | 6 | 2 |
| Peripheral T Cell Lymphoma, NOS | 4 | 3 |
| Angioimmunoblastic T Cell Lymphoma | 3 | 1 |
| Follicular Lymphoma | 4 | 2 |
| Mantle Cell Lymphoma | 3 | 1 |
| Chronic Lymphocytic Leukemia | 1 | 1 |
| Marginal Zone Lymphoma | 4 | 1 |
| Burkitt Lymphoma | 2 | 1 |
| Classic Hodgkin Lymphoma | 5 | 0 |
| Cervical Carcinoma, Invasive SCC | 4 | 2 |
| Endometrioid Adenocarcinoma | 4 | 2 |
| Papillary Renal Cell Carcinoma | 3 | 1 |
| Urothelial Carcinoma | 3 | 0 |
| Gastric Carcinoma | 2 | 0 |
| Invasive Intestinal Adenocarcinoma | 1 | 0 |
| Malignant Mullerian Mixed tumor(carcinsarcoma) | 1 | 0 |
| Ovarian, serous carcinoma | 1 | 0 |

**Supplemental Table S2. Results of TRAILshort ISH in human tissue sections.**

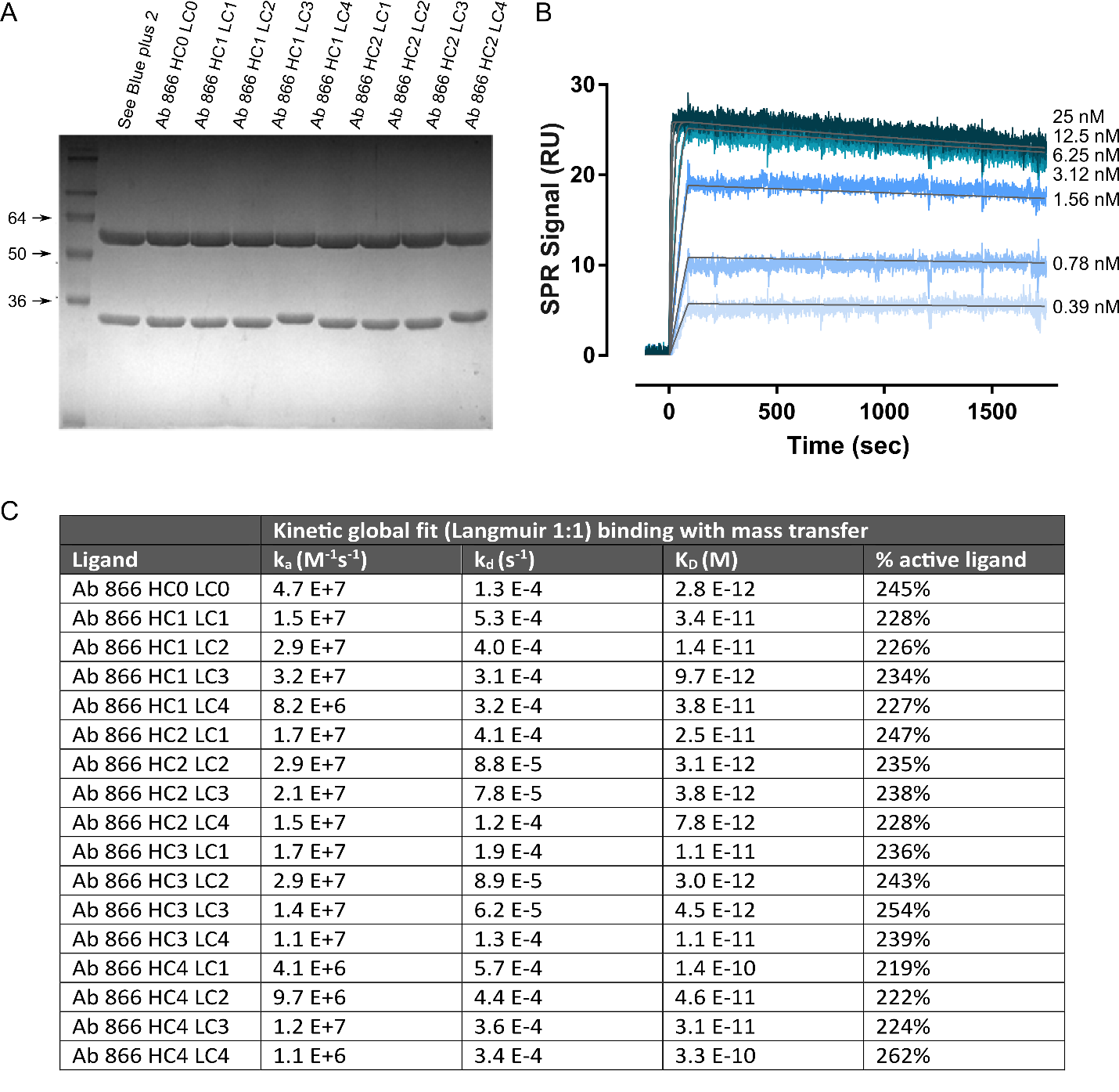




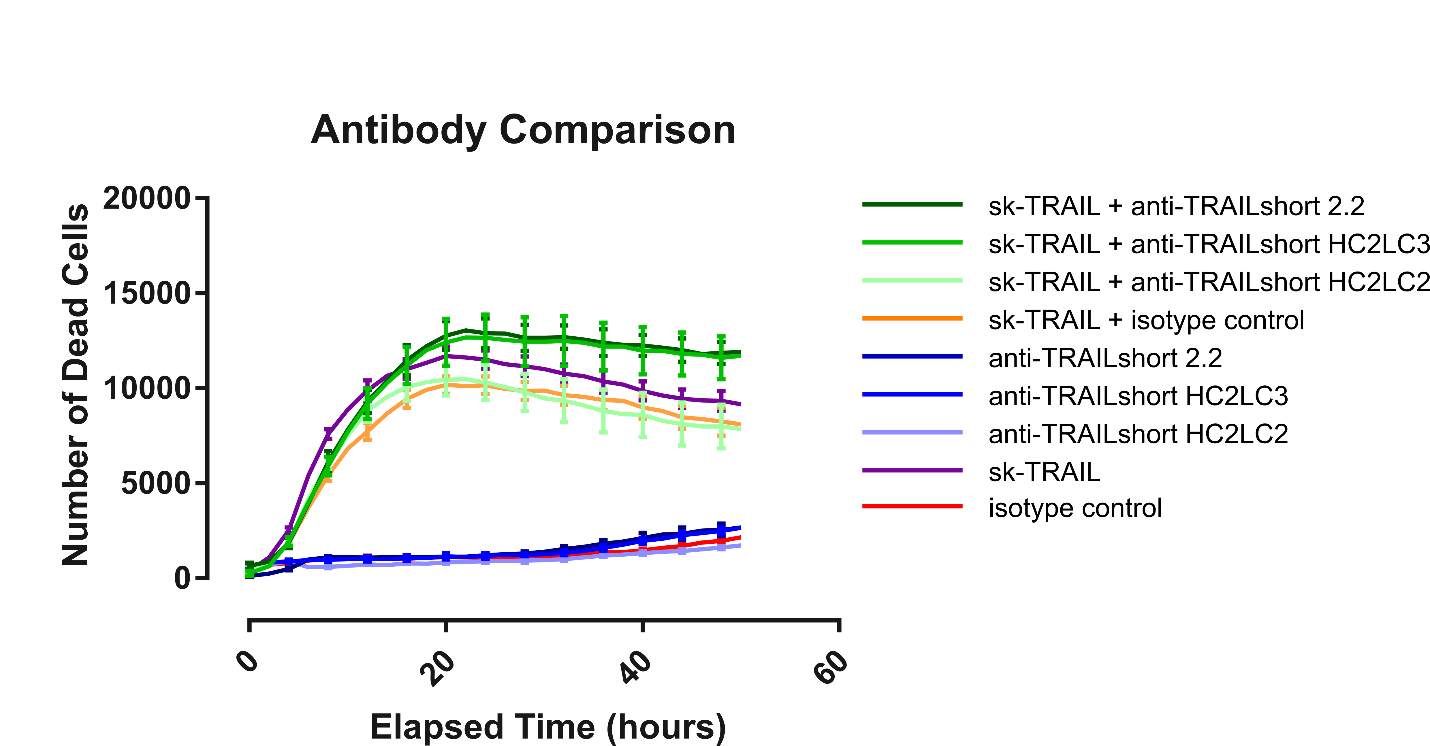
**Supplemental Document S1.** **Report of safety evaluation of TRAILshort antibody administered to mice and followed for 7 days.**

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**Supplemental Figure S2. Single dose anti-TRAILshort antibody in combination with a TRAIL agonist has antitumor effects in a Jurkat T cell xenograft model (supplemental).** Non-obese diabetic, severe combined-immunodeficient, common γ-chain deficient (NSG) mice were implanted with Jurkat human T leukemia cells expressing luciferase by IV injection, and tumors were allowed to become established. Mice were then administered a single injection of mouse anti-TRAILshort antibody (clone 2.2) followed 24 hours later by a single injection of anti-TRAIL-R2 antibody, and imaged twice weekly. Whole mouse imaging on day 45 shown.

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**Supplemental Figure S3. Characterization of humanized anti-TRAILshort antibodies.** (A) Comassie stain of different humanized anti-TRAILshort variants, (B) surface plasmon resonance (SPR) profile of HC2LC3 antibody binding to TRAILshort, and (C) affinity data for different humanized anti-TRAILshort variants.



**Supplemental Figure S4.** **Comparison of anti-tumor effect of mouse anti-TRAILshort 2.2 versus humanized anti-TRAILshort HC2LC2.** Functional effect of mouse anti-TRAILshort clone 2.2, humanized anti-TRAILshort clone HC2LC2, and humanized anti-TRAILshort clone HC2LC3 on Jurkat cell survival with and without sk-TRAIL. Data are represented as mean ± SE.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Diagnosis** | **Responsive to sk‑TRAIL + anti‑TRAILshort** | **Responsive to Anti-TRAILshort** | **Fold Increase in Number of Dead cells** | **P-value** | **TRAILshort RNA-ISH** |
| Non Hodgkin Lymphoma B cell lineage Cyclin D1 Positive | Yes | Yes | 1.19 | 0.019 | Positive |
| Splenic diffuse small B cell lymphoma | Yes | Yes | 1.98 | 0.014 | Positive |
| Mantle Cell lymphoma | Yes | Yes | 1.21 | 0.029 | Negative |
| Mantle cell Lymphoma | No | No | 1.28 | 0.182 | Negative |
| EBV associated lymphoproliferative disorder | Yes  (dose dependent) | No | 1.68 | 0.006 | Positive |
| Burkitt Lymphoma | Yes | No | 2.46 | 0.002 | Positive |
| Angioimmunoblastic T-cell Lymphoma | Yes | No | 1.83 | 0.009 | Negative |
| Follicular Lymphoma | Yes | No | 2.68 | 0.002 | Negative |
| Follicular Lymphoma | Yes | Yes | 1.38 | 0.05 | Positive |
| Follicular Lymphoma | Yes | No | 1.37 | 0.045 | N/A |
| Splenic marginal zone lymphoma | No | No | 1.28 | 0.217 | Negative |
| Splenic marginal zone lymphoma | No | No | 0.95 | 0.727 | Negative |
| Splenic marginal zone lymphoma | No | No | 0.96 | 0.822 | N/A |
| Splenic marginal zone lymphoma | No | No | 0.98 | 0.875 | N/A |
| Splenic marginal zone lymphoma | No | No | 0.81 | 0.316 | N/A |
| Classic Hodgkin Lymphoma, nodular sclerosis | No | No | 0.95 | 0.794 | Negative |
| Classic Hodgkin Lymphoma, nodular sclerosis | No | No | 0.85 | 0.193 | N/A |
| Hodgkin's Lymphoma no | No | No | 1.24 | 0.159 | N/A |
| CLL/SLL | No | No | 1.05 | 0.558 | Positive |
| CLL/SLL | No | No | 0.96 | 0.771 | N/A |
| Recurrent large B-cell lymphoma with extensive T-cell infiltrate | No | No | 1.16 | 0.437 | N/A |
| Reactive Follicular Hyperplasia | No | No | 1.33 | 0.092 | N/A |
| Reactive Follicular Hyperplasia | No | No | 0.92 | 0.560 | N/A |
| Non-lymphoma ITP | No | No | 0.93 | 0.673 | N/A |
| Non-lymphoma ITP | No | No | 1.25 | 0.348 | N/A |
| Non-lymphoma | No | No | 1.04 | 0.823 | N/A |

**Supplemental Table S3. Summary of effects of anti-TRAILshort antibody HC2LC3 on primary human tumors.** Response of primary splenocytes from human subjects with and without malignant diagnoses to cell killing induced by humanized anti-TRAILshort plus sk-TRAIL, or humanized anti-TRAILshort antibody alone, TRAILshort as measured by *in situ* hybridization.