**Supplementary Figures**

Supplementary Figure S1: LY6G6D level in Human Immune Cell Subsets as Measured by Flow Cytometry. Human PBMCs from 12 healthy donors were stained for CD3, CD8, CD4, CD14, CD56, and Dylight 650-labeled rabbit anti-LY6G6D antibody or rabbit IgG isotype, followed by Flow. **A**, LY6G6D level in human CD4+ T Cells. **B**, LY6G6D level in human CD8+ T Cells. **C**, LY6G6D level in human Natural Killer Cells. **D**, LY6G6D expression in human Monocytes.

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Supplementary Figure S2: Distance mapping of LY6G6D peptide bound to 1G4 Fab complex with symmetry related complex. View and coloring are similar to Figure 3C. The symmetry related complex is represented as surface (with backbone in ribbon); for clarity, only the CDR L1 of the symmetry Fab is shown. Color gradient represents distance of both symmetry objects to LY6G6D peptide (red = 4 Å and yellow 8 Å). No clear interactions are observed between the two peptides although they share an estimated 164 A2 of buried surface area.

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Supplementary Figure S3: SEC-MALS analysis of recombinant LY6G6D alone or in complex with Fab 1G4. **A**, Untagged LY6G6D behaves as a 9.6 kDa (+/- 0.7 kDa) protein in solution (estimated molecular weight for monomeric protein with disulfides: 9952 Da). UV absorbance measured at 280nm (left axis) is in blue and calculated molar mass (right axis) in black. **B**, Molecular weight of the complex of untagged Ly6G6D bound to 1G4 Fab is estimated at 56.2 kDa (+/- 2.7 kDa); this data suggests that one Fab (47.9 kDa) binds one LY6G6D molecule.

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Supplementary Figure S4: Binding of LY6G6D-TDB to the engineered LY6G6D-expressing 293 cell line. **A**, LY6G6D-TDB binds to the engineered 293-LY6G6D cells, but not to LY6G6D-negative parental 293 cells by flow cytometry. **B**, LY6G6D level in the engineered 293-LY6G6D cell line by IHC and flow cytometry.

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Supplementary Figure S5: LY6G6D expression in the xenograft tumors by IHC. **A**, LS1034 xenograft tumor, IHC score of 3+. **B**, HT55 xenograft tumor, IHC score of 2+.

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Supplementary Figure S6:Pharmacokinetics (PK) of 1G4-CD3H TDB in mice.1). In a single dose PK study, 5 mg/kg of the antibody was administered as a single intravenous (IV) bolus dose in female C.B-17 SCID.bg mice (n=12) and PK were determined over a period of 28 days using sparse PK method. Sparse PK samples (n=3 animals per time point) were collected and the sampling time points included were 15 minutes, 2 and 6 hours, 1, 3, 7, 10, 14, 21, and 28 days (black solid line). The observed maximum serum concentration (Cmax) was 88.4 ± 2.15 µg/mL, area under concentration-time curve from time zero to 28 days (AUC0-28) was 408 ± 18.5 day.µg/mL, estimated clearance (CL) was 9.85 mL/day/kg and terminal elimination half-life (t1/2) was 13.1 days. This estimated CL of 1G4-CD3H TDB in SCID mice was approximately 2 times higher than the positive control antibody (non-binding humanized anti-gD 5B6) whose CL estimate was 4.53 mL/day/kg (black dashed line). 2). PK in HT55 tumor xenograft model in NSG mice. Animals were divided into two groups and were received a single IV dose of 1G4-CD3H TDB at 1 mg/kg, plus 10x106 human PBMCs (blue dashed line), or no PBMCs (red dashed line). While PBMC treated group has Day1 serum exposure of 6.83 ± 0.31 µg/mL and AUC0-14 of 52.6 ± 1.94 day.µg/mL, the group without PBMCs treatment had Day1 serum exposure of 6.13 ± 0.25 µg/mL and AUC0-14 of 52.0 ± 3.18 day.µg/mL.

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