Supporting Information

PNAS PNAS

Desmyter et al. 10.1073/pnas.1301336110

	10!20!30!40!50!abcdef60!
cAb-BP02	QVQLVESGGGLVQAGGSLRLS <mark>C</mark> AASE <mark>STFSNYAMGW</mark> FRQAPGPEREFVATISQTGSHTYYRNS
cAb-BP03	DVQLVESGGGLVQAGGSLRLS <mark>C</mark> VASG <mark>RTFSINTLSW</mark> FRQAPGKVREFVSGIEWSSGWPTYADS
cAb-BP07	DVQLVESGGGLVQAGTSRRLS <mark>C</mark> VASG <mark>TSFSRRQMGW</mark> FRQAPGKERTFVAAISGSGNWLDVTDS
cAb-BP09	QVQLVESGGGLVQPGGSLRLSCVHSGSIFSDNAMGWYRQAPGNQRERVALITSGGNTNYADS
cAb-BP11	QVQLVESGGGLVQAGDSLRLS <mark>C</mark> AVSG <mark>RTFSSNVIGW</mark> FRQAPGKEREFVAAISWSTGSTYYGRS
cAb-BP13	QVQLVESGGGLVQAGDSLRLS <mark>C</mark> AASG <mark>RTFSGYAVAWFR</mark> QAPGKEREFVAVIWRSGTGAYYVHADYAES
cAb-BP17	DVQLVESGGGLVQPGGSLRLS <mark>C</mark> EASG <mark>FSFDDYAIGW</mark> FRQAPGKEREGVSYISMSDGRTYVADS
cAb-BP18	QVQLVESGGGLVQPGGSLRLS <mark>C</mark> VASG <mark>SIFSRAVMG</mark> WYRQAPGKQRELVATITIHGYTTYADS
cAb-BP19	QVQLVESGGGLVQAGGSLRLS <mark>C</mark> AASG <mark>RTFSSYAMG</mark> WFRQAPGKEREFVAVISWSTGSTYYAGS
cAb-Tal11	QVQLVESGGGLVQPGGSLRLSCVASGRTFNTPDMGWFRQAPGNQREFVGRIRTSDGRTYYADS
cAb-Tal18	QVQLVESGGGLVHPGGSLRLS <mark>C</mark> AASG <mark>RTFSDYALGWFRQ</mark> APGKDREFVAAISWSGGSTYYADS
cAb-Tal41	QVQLVESGGGLVQAGDSLRLS <mark>C</mark> AYSG <mark>RTFSGYRMG</mark> WFRQAPGKEREYVGGPTSSSGKIYYADS
	70!80!abc90!100!abcdefghijklmnop110!
cAb-BP02	VKGRFTISRDNAKNTVYLQMNNMKPEDTAVYYCAAGDNYYYTRTYEYDYWGQGTQVTVSS
cAb-BP03	VKGRFTISREPAKNTVYLQMTNLTPEDTAVYYCAARRSAVGTYEYWGQGTQVTVSS
cAb-BP07	LQGRFTISRDNAKNTVYLQMNDLKPEDTAVYYCVSKEPTRWDSTLSGDVRGYSYWGPGTQVTVSS
cAb-BP09	VKGRFTISRDNSKSTVYLQMDSLMPEDTAVYYCNTPYMRKEDEYWGPGTQVTVSS
cAb-BP11	MKGRCAASRDNAKNTVALQLNSLKPEDTAVYYCAATLDWGKTLSDEYDYWGQGTQVTVSS
cAb-BP13	VRGRFTISIDNAKNTVYLQMNSLKLEDTAVYYCAATTRGVALIRPAGYDYWGQGTQVTVSS
cAb-BP17	VTGRFTISSDNAKNTVYLQMNSLKLEDTAVYYCAAGRFVTFGSAWSFVGGGPYGIDYWGKGTLVTVSS
cAb-BP18	VKGRFTISRDSAKNTVYLQMNSLKPEDTAVYYCNALRGIPNDYWGPGTQVTVSS
cAb-BP19	VKGRFTISRDNAKNTVYLQMNSLKPEDTAVYYCAATLDWGKTLSYEYDYWGQGTQVTVSS
cAb-Tal11	VKGRFTISRDAAKKTMYLQMNSLKPEDTAVYYCASRERIGQDYDYWGQGTQVTVSS
cAb-Tal18	VQGRFTISRDNDKNSVYLQMNSLKPEDTAVYYCAADKYYTGPGGESVYDYWGRGTQVTVSS
cAb-Tal41	VNGRFTISRDYAKNTVYLOMNSLKPEDTAVYYCAAARTTTALYEYWGOGTOVTVSS

Fig. S1. Sequences of the nanobodies raised against TP901-1 baseplate or tail-associated lysozyme (Tal) protein (numbering according to the work by Kabat et al.) (1). CDR1, -2, and -3 are red, green, and blue, respectively.

1. Kabat EA, Wu TT, Perry HM, Gottesman KS, Foeller C (1991) Sequences of Proteins of Immunological Interest, ed Services USDoHaH (Public Health Service, National Institutes of Health, Bethesda, MD), 5th Ed.



Nanobodies 2 & 17





Fig. S2. EM of the TP901-1 baseplate in complex with nanobodies. Views of the raw images and the EM maps of the baseplate (grid surface) with the ribbon representation of the X-ray structure inside. The position of the nanobodies is provided by the difference map between the complex and the native baseplate (violet). The identification of the molecular species is provided.



Fig. S3. Ribbon views of complexes of receptor binding proteins (RBPs) with various binders. (A) TP901-1 RBP with nanobody 2. (B) TP901-1 RBP with nanobody 11. (A and B) The structures in gray are not seen in the electron density maps. (C) TP901-1 RBP with nanobody 17. (D) TP901-1 RBP with nanobody 2 and the complex between p2 RBP and vhh5 (1) superimposed (beige ribbon). (E) View at 90° from D. (F) TP901-1 RBP in complex with DARPIN 20 (2).

1. Spinelli S, et al. (2006) Lactococcal bacteriophage p2 receptor-binding protein structure suggests a common ancestor gene with bacterial and mammalian viruses. Nat Struct Mol Biol 13:1):85–89.

 Veesler D, et al. (2009) Crystal structure and function of a DARPin neutralizing inhibitor of lactococcal phage TP901-1: Comparison of DARPin and camelid VHH binding mode. J Biol Chem 284(44):30718–30726.

Table 31. Tryulogen bonus established between hanoboules 2, 11, and 17 and the RDF in the comple	Table S1.	Hydrogen bonds established betwee	en nanobodies 2, 11, and	17 and the RBP in the comp	lexes
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Nanobody 2	Distance (Å)	RBPmon1	Nanobody 11	Distance (Å)	RBPmon1	Nanobody 17	Distance (Å)	RBPmon1
TYR58 [OH]	2.6	GLY74 [O]	TYR58 [OH]	2.50	GLY74 [O]	TYR50 [OH]	2.61	SER3 [OG]
TYR58 [OH]	3.13	GLU93 [O]	TYR58 [OH]	3.18	GLU93 [O]	TYR58 [OH]	2.72	ILE4 [O]
ASN61 [ND2]	3.27	SER95 [O]	ARG61 [N]	3.20	GLU93 [OE2]	SER100b [N]	3.21	ASP20 [OD2]
TYR98 [OH]	2.7	SER68 [OG]	ARG61 [NE]	2.50	ILE94 [O]	SER100b [OG]	3.43	ASP20 [OD2]
THR100a [OG1]	3.73	GLY92 [O]	ARG61 [NH2]	3.89	GLY 152 [O]	TRP100d [N]	2.86	SER3 [O]
ARG100b [NH1]	3.17	THR153 [OG1]	ARG61 [NH2]	3.66	GLU93 [OE1]	SER100e [OG]	2.64	GLU28 [OE2]
TYR58 [OH]	3.04	GLU93 [N]	THR 100a [OG1]	3.35	GLY92 [O]	TYR100I [N]	2.84	ASN34 [OD1]
TYR99 [O]	2.83	GLY92 [N]	TYR58 [OH]	3.11	GLU93 [N]	GLY100m [N]	3.1	GLY33 [O]
THR100a [OG1]	2.99	ARG155 [NH2]	TRP 98 [O]	3.82	SER78 [OG]	TYR58 [OH]	3.4	ILE4 [N]
GLU100e[OE1]	2.94	ARG155 [NH2]	GLY 99 [O]	3.00	GLY92 [N]	VAL59 [O]	3.27	LYS5 [NZ]
GLU100e [OE2]	2.89	ARG155 [NE]	GLU 100e [OE1]	2.85	ARG 155 [NH2]	GLY100i [O]	2.93	THR43 [N]
GLU100e [OE2]	2.89	RBPmon2	GLU 100e [OE2]	2.80	ARG 155 [NE]			RBPmon2
TYR100 [OH]	3.33	ARG118 [O]	ARG61 [NH2]	3.66	GLU93 [OE1]	GLY 100j [O]	3.09	LYS 38 [NZ]
TYR100 [OH]	2.78	SER120 [OG]			RBPmon2	GLY 100h [O]	2.90	LYS 38 [NZ]
ASP96[OD1]	2.73	SER138 [OG]	LYS 100 [NZ]	3.48	ARG 118 [O]			
ASP96 [OD2]	2.83	SER138 [N]	ASP 97 [OD2]	2.92	ARG 118 [N]			
ASN97 [OD1]	2.87	ARG118 [N]	ASP 100d [O]	3.43	ARG 118 [NH2]			
TYR100 [OH]	3.69	SER120 [N]	ASP 100d [OD2]	3.35	GLN 143 [NE2]			
TYR100d [O]	3.81	ARG118 [NH2]						
TYR100d [O]	2.97	GLN143 [NE2]						
GLU100e [O]	3.47	ARG118 [NE]						
ASP100g [OD1]	3.23	ARG118 [NH2]						