Supplemental Methods

Physicochemical rules for identifying monoclonal antibodies with drug-like specificity

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Individual chemical rules for identifying antibodies with low specificity. The chemical rules were generated using threefold cross validation methods and were required to meet a number of constraints. First, the clinical-stage mAbs (137) were split into training (80%) and test (20%) sets in ten different ways using stratified sampling (Table S6). The training sets were further divided into three partitions (folds), and two partitions were used for training and one for validation. Individual rules were required to satisfy the following constraints: i) adjusted accuracy (herein simply referred to as accuracy) of preclinical antibodies (training set) >55%; ii) % mAbs flagged with high specificity (preclinical, training set) < % mAbs flagged with low specificity (preclinical, training set); iii) accuracy (clinical mAbs, training set) >55% in each fold (three constraints for each of the ten 80/20 splits); iv) difference between the accuracy of training (two folds) and validation (one fold) < 5% (three constraints for each of the ten 80/20 splits, clinical mAbs); v) % mAbs flagged with high specificity (defined by each individual assay) < % mAbs flagged with low specificity (defined by each individual assay; five constraints that were evaluated using the entire 80% of the clinical antibody training data for each of the ten 80/20 splits); vi) average validation accuracy for each of the ten 80/20 splits > 60% where the average validation accuracy is the average of the validation accuracy for all observed flag values within each 80/20 split; vii) average test accuracy for each of the ten 80/20 splits > 50% where the average test accuracy is the average of the test accuracies for all observed flag values within each 80/20 split. These constraints are summarized in Table S7. Finally, the rules were required to be observed in each of the ten 80/20 splits, although different values for the rules were allowed.

Combined rules for enhancing the identification of mAbs with low specificity. Sets of rules were generated by combining single rules together (up to six single rules per combined set were evaluated), as explained in the Results section. Each mAb was considered to have low specificity if flagged by four or five rules (as specified). Sets of rules in the first round of analysis were only accepted if they met several requirements: i) accuracy >60% (preclinical mAbs, training set); ii) % mAbs flagged with high specificity <10% (preclinical mAbs, training set); iii) accuracy (clinical mAbs, training set) >60% in each fold (three constraints for each of the ten 80/20 splits); iv)

difference between the accuracy of training (two folds) and validation (one fold) <10% (three constraints for each of the ten 80/20 splits, clinical mAbs); v) flag <5% clinical-stage mAbs with high specificity in training sets (as defined combination of five assays); vi) % accuracy (clinical mAbs, training set) >60% (defined by each individual assay; five constraints that were evaluated using the entire 80% of the clinical antibody training data for each of the ten 80/20 splits); and vii) % accuracy (clinical mAbs, training set) >60% (defined by combination of five assays; one constraint was evaluated using the entire 80% of the clinical antibody training data for each of the ten 80/20 splits). These constraints are summarized in Table S7. Finally, the combined rules (with the same values for each rule) were required to be observed in each of the ten 80/20 splits. The best sets of combined rules (Table S9) in the first round of analysis were identified as those with the lowest coefficients of variation for the average validation accuracy (ten 80/20 splits).

Next, mAbs that were not flagged as polyspecific in the first round of analysis (Set A in Table S9, <4 flags, 121 of 137 clinical mAbs and 375 of 424 preclinical mAbs) were evaluated in round 2 of analysis. First, single rules were generated using the same constraints as used in the first round of analysis. Next, combined sets of rules using the six rules in Set A (Table S9) and up to six additional rules were required to meet a number of constraints: i) accuracy >70% (preclinical mAbs, training set); ii) accuracy (clinical mAbs, training set) >75% in each fold (three constraints for each of the ten 80/20 splits); iii) difference between the accuracy of training (two folds) and validation (one fold) <10% (three constraints for each of the ten 80/20 splits, clinical mAbs); iv) flag <10% clinical-stage mAbs with high specificity in training sets (as defined by the combination of five assays); v) % accuracy (clinical mAbs, training set) >70% (defined by each individual assay; five constraints that were evaluated using the entire 80% of the clinical antibody training data for each of the ten 80/20 splits); and vi) % accuracy (clinical mAbs, training set) >75% (defined by combination of five assays; one constraint that was evaluated using the entire 80% of the clinical antibody training data for each of the ten 80/20 splits). These constraints are summarized in Table S7. Finally, the combined rules (with the same values for each rule) were required to be observed in each of the ten 80/20 splits. The best five sets of combined rules in concert with Set A (Table S9) were identified as those with the lowest coefficients of variation for the average validation accuracy (ten 80/20 splits; Table S12).

Deep sequencing and data analysis. The antibody libraries were evaluated using deep sequencing, as described in the Methods section. The V_H region of the scFab gene was amplified via two-step PCR. The first step was performed using primers that were complementary to V_H and added the Illumina adapter sequences and barcodes. The second reaction used the purified PCR

product from the first reaction and primers identical to the Illumina adapter sequences. The primers are summarized below:

Primer Name	Sequence	Function
Key	p5-i5-filler-plasmid_complement	Forward Primer for Sample Preparation
CSpr90	AATGATACGGCGACCACCGAGATCTACACATCGTACGTCACTCGCTACCGCTGGAGCTTCTTCTGGC	
CSpr91	AATGATACGGCGACCACCGAGATCTACACACTATCTGTCACTCGCTACCGCTGGAGCTTCTTCTGGC	
CSpr92	*AATGATACGGCGACCACCGAGATCTACACTAGCGAGTTCACTC*GCTACCGCTGGAGCTTCTTCTGGC	
CSpr93	AATGATACGGCGACCACCGAGATCTACACCTGCGTGTTCACTCGCTACCGCTGGAGCTTCTTCTGGC	
CSpr94	AATGATACGGCGACCACCGAGATCTACACTCATCGAGTCACTCGCTACCGCTGGAGCTTCTTCTGGC	
CSpr95	AATGATACGGCGACCACCGAGATCTACACCGTGAGTGTCACTCGCTACCGCTGGAGCTTCTTCTGGC	
Key	p7-i7-filler-plasmid_complmement	
MSpr14	CAAGCAGAAGACGGCATACGAGATAACTCTCGGTGACTGCCAATGGAAAAACAGAGGGCCC	Reverse Primer for Sample Preparation
MSpr15	CAAGCAGAAGACGGCATACGAGATACTATGTCGTGACTGCCAATGGAAAAACAGAGGGCCC	
MSpr16	CAAGCAGAAGACGGCATACGAGATAGTAGCGTGTGACTGCCAATGGAAAAACAGAGGGCCC	
MSpr17	CAAGCAGAAGACGGCATACGAGATCAGTGAGTGTGACTGCCAATGGAAAAACAGAGGGCCC	
MSpr18	CAAGCAGAAGACGGCATACGAGATCGTACTCAGTGACTGCCAATGGAAAAACAGAGGGCCC	
MSpr19	CAAGCAGAAGACGGCATACGAGATCTACGCAGGTGACTGCCAATGGAAAAACAGAGGGCCC	
CSpr96	TCACTCGCTACCGCTGGAGCTTCTTCTGGC	Read1 Primer
MSpr12	GGTGACTGCCAATGGAAAAACAGAGGGCCC	Read2 Primer
MSpr13	GGGCCCTCTGTTTTTCCATTGGCAGTCACC	Index1 Primer

The raw sequencing files from Illumina MiSeq (300 bp paired-end sequencing reaction) were merged together using BBMerge[1] with the qtrim parameter set to 15 and all other parameters set to default values. The resulting merged ".fastq" file was converted to a ".fasta" file and analyzed line by line. Sequences were translated with BioPython[2] if they were the correct size (378 bp) without any 'N' base calls. The frequency of each set of mutations (herein referred to as a mutational string) was counted if the first residue of the translation was the correct amino acid ('A') and there were no stop codons. If the first residue was incorrect or there was a stop codon, the translation of the reverse complement was checked. The frequency of each mutational string was determined and exported into a ".csv" file for calculation of the enrichment ratios.

References

1. Bushnell, B.; Rood, J.; Singer, E. BBMerge - Accurate paired shotgun read merging via overlap. *Plos One* **2017,** *12*, (10).
2. Cock, P. J. A.; Antao, T.; Chang, J. T.; Chapman, B. A.; Cox, C. J.; Dalke, A.; Friedberg, I.; Hamelryck, T.; Kauff, F.; Wilczynski, B.; de Hoon, M. J. L. Biopython: freely available

Python tools for computational molecular biology and bioinformatics. *Bioinformatics* **2009,** *25*, (11), 1422-1423.

Table S5. Maximum and minimum values for the observed counts of amino acids (weighted by their solvent accessibilities) and net charges (pH 7.4) of different regions within the variable domains of clinical-stage and preclinical mAbs in the training sets. Glycine is assumed to be fully exposed (SASA value of one). The net charges were calculated by assigning values of +1 for R and K, +0.1 for H, and -1 for D and E. The CDRs were defined using a combination of Chothia and Kabat numbering, and heavy chain CDR3 was defined to also include two additional N-terminal residues. It is expected that the performance of the rules generated in this study will be highest for mAbs that do not violate any of these limits.

Max limits	A	C	D	E	F	G	H	I	K	L	M
H1	0.81	0	1.12	0.64	0.78	6	0.62	1.11	0.73	0.28	0.10
H2	0.98	0	2.01	1.79	0.81	5	0.80	1.48	1.79	0.32	0.64
H3	1.15	0.05	1.88	0.70	0.93	4	0.76	1.07	0.92	1.67	0.90
H123	1.37	0.05	2.61	2.41	1.47	9	1.49	1.69	2.16	1.67	0.91
L1	0.82	0.02	2.43	0.80	0.68	4	0.92	1.30	1.74	0.61	0.01
L2	0.45	0	1.95	0.94	0.60	2	0.36	0.75	0.62	0.54	0.24
L3	0.68	0.05	0.78	0.85	0.49	4	0.67	0.69	0.53	0.92	0.50
L123	1.02	0.05	2.71	1.56	0.73	7	0.99	1.30	1.74	1.25	0.50
CDR	1.95	0.05	4.22	3.04	1.47	12	1.80	1.79	2.89	1.78	0.91
VH	3.39	0.05	3.63	4.34	1.47	18	1.49	2.18	4.73	2.65	1.02
VL	2.98	0.06	4.54	4.31	1.22	17	1.26	1.59	4.05	2.67	0.50
Fv	5.94	0.06	6.70	6.99	1.67	30	1.80	2.80	7.41	4.29	1.02
framework	4.66	0.03	4.43	5.51	0.99	19	0.88	1.59	6.37	3.37	0.64

Max limits	N	P	Q	R	S	T	V	W	Y	# of residues	charge
H1	1.28	0.61	0.02	0.68	2.01	1.54	0.63	0.41	2.02	12	1.1
H2	2.33	0.81	2.10	1.47	2.82	1.50	0.67	0.63	1.59	19	3
H3	0.81	0.79	0.60	1.13	1.67	1.20	1.83	1.06	2.07	23	3.1
H123	2.88	1.09	2.10	1.87	4.60	2.54	1.83	1.18	3.59	50	4.1
L1	1.51	0.61	1.31	1.73	3.28	1.37	0.84	0.47	1.07	17	3.2
L2	1.41	0.77	0.59	1.40	2.39	1.44	0.33	0.64	0.86	7	2.1
L3	0.97	0.72	0.57	0.69	1.30	1.03	0.59	0.47	0.95	12	2.1
L123	2.94	0.94	1.31	2.70	5.36	2.06	1.16	0.71	1.74	33	5.3
CDR	3.84	1.68	3.12	2.74	8.22	3.58	1.96	1.34	4.05	79	7.1
VH	4.11	3.47	4.68	3.84	11.50	7.26	2.82	1.47	3.80	130	7.1
VL	3.28	3.67	3.32	4.16	12.61	5.22	2.56	0.71	2.09	113	5.2
Fv	6.86	5.36	7.26	5.57	20.64	10.93	4.29	1.69	4.70	238	9.1
framework	4.02	4.75	6.70	4.52	15.17	9.43	3.39	0.54	1.52	161	5.1

Min limits	A	C	D	E	F	G	H	I	K	L	M
H1	0	0	0	0	0	0	0	0	0	0	0
H2	0	0	0	0	0	0	0	0	0	0	0
H3	0	0	0	0	0	0	0	0	0	0	0
H123	0	0	0	0	0	1	0	0	0	0	0
L1	0	0	0	0	0	0	0	0	0	0	0
L2	0	0	0	0	0	0	0	0	0	0	0
L3	0	0	0	0	0	0	0	0	0	0	0
L123	0	0	0	0	0	0	0	0	0	0	0
CDR	0	0	0	0	0	1	0	0	0	0	0
VH	0.35	6E-05	0.29	0.30	0	7	0	0	1.17	0.02	0
VL	0.13	3E-05	0.03	0.60	0.01	7	0	0.01	0.76	0.22	0
Fv	0.75	3E-04	1.15	1.54	0.04	17	0	0.08	2.79	0.31	0
framework	0.63	3E-04	0.10	1.54	0.01	13	0	0.01	1.85	0.31	0

Min limits	N	P	Q	R	S	T	V	W	Y	# of residues	charge
H1	0	0	0	0	0	0	0	0	0	10	-2
H2	0	0	0	0	0	0	0	0	0	16	-3
H3	0	0	0	0	0	0	0	0	0	5	-4
H123	0	0	0	0	0	0	0	0	0.19	32	-4.9
L1	0	0	0	0	0.25	0	0	0	0	10	-4
L2	0	0	0	0	0	0	0	0	0	7	-2
L3	0	0	0	0	0	0	0	0	0	7	-2
L123	0	0	0	0	0.32	0	0	0	0	24	-5
CDR	0	0	9E-04	0	0.82	0.21	0	0	0.31	60	-7
VH	0	0.53	1.17	0	5.26	1.25	0.27	0.01	0.29	112	-4.9
VL	0	0.88	0.74	0.52	6.19	1.99	0.10	0.00	0.22	104	-5.9
Fv	0	1.74	2.26	0.70	12.88	4.36	0.95	0.02	1.02	220	-5
framework	0	1.55	1.90	0.70	8.78	2.92	0.74	0.01	0.13	158	-4

Table S6. Summary of segregation of clinical-stage mAbs into training and test sets. Clinical-stage mAbs (as numbered in Table S1) were divided into ten sets of 80% training mAbs and 20% test mAbs. For each 80/20 split, the training sets of mAbs (80%) was further divided into three groups for threefold cross validation.

80/20 Split	1				2				3				4			
	Test	Training set			Test	Training set			Test	Training set			Test	Training set		
	set	A	B	C	set	A	B	C	set	A	B	C	set	A	B	C
Clinical-stage mAbs with high specificity	16	1	2	4	7	1	9	3	1	5	6	2	4	3	5	1
	20	3	8	5	11	2	12	6	4	7	11	3	10	6	7	2
	34	10	11	6	20	4	24	8	9	8	12	15	13	15	8	9
	35	14	12	7	23	5	26	10	10	13	16	25	14	17	11	21
	38	18	17	9	25	14	28	13	14	17	19	26	16	18	12	23
	42	31	19	13	27	15	29	16	24	18	38	28	28	20	19	24
	45	37	25	15	30	18	32	17	35	20	39	29	41	22	27	29
	51	48	27	21	49	31	34	19	43	21	40	30	48	25	30	31
	52	49	29	22	50	33	35	21	57	22	44	33	54	26	32	33
	54	50	33	23	51	37	41	22	58	23	47	36	62	40	34	36
	55	56	36	24	58	38	42	36	60	27	49	37	64	42	35	37
	61	59	39	26	59	40	44	39	63	31	50	41	77	46	38	39
	68	63	40	28	62	48	45	43	64	32	51	42	78	50	44	43
	70	65	41	30	64	52	54	46	65	34	54	48	80	52	45	51
	71	67	44	32	78	53	63	47	73	45	56	66	88	57	47	53
	76	69	47	43	82	55	69	57	80	46	61	69	89	59	49	55
	80	72	53	46	89	56	71	60	84	52	62	70	93	66	60	56
	82	73	57	60	90	61	72	66	86	53	67	76	94	67	65	58
	94	74	58	62	95	65	75	68	93	55	68	78	95	69	76	61
		77	75	64		67	76	73		59	71	79		73	81	63
		78	79	66		70	79	74		81	72	82		75	82	68
		84	81	86		80	83	77		85	74	83		79	83	70
		88	83	87		84	87	81		87	75	88		84	87	71
		89	85	90		88	91	85		90	77	89		86	90	72
		91	93	96		94	92	86		94	95	91		92	91	74
		92	95	97		96	97	93		96	97	92		97	96	85
Clinical-stage mAbs with low specificity	Test	Training set			Test	Training set			Test	Training set			Test	Training set		
	set	A	B	C	set	A	B	C	set	A	B	C	set	A	B	C
	98	106	101	99	99	101	107	98	99	100	103	98	105	98	100	99
	100	108	103	105	103	106	109	100	101	102	106	104	113	102	104	101
	102	109	107	112	104	108	119	102	108	107	111	105	115	109	116	103
	104	110	114	113	114	111	121	105	109	112	113	110	117	110	119	106
	115	111	118	123	118	112	124	110	115	116	114	118	123	111	120	107
	120	116	122	125	130	116	125	113	130	117	119	122	126	112	122	108
	121	117	127	129	131	117	132	115	132	123	120	125	128	114	127	121
	124	119	128	131	134	120	133	122	133	124	121	126	129	118	131	124
		126	132	133		123	135	126		127	128	131		130	132	125
		130	134	135		127	136	129		129	136	134		135	133	137
		137	136			128	137			135	137			136	134	

80/20 Split	5				6				7				8			
	Test set	Training set			Test set	Training set			Test set	Training set			Test set	Training set		
		A	B	C		A	B	C		A	B	C		A	B	C
Clinical-stage mAbs with high specificity	3	6	2	1	12	1	3	2	3	6	2	1	3	5	1	2
	9	8	4	7	21	5	4	16	9	8	4	7	4	8	23	6
	23	10	5	11	23	6	7	18	23	10	5	11	7	12	24	9
	24	12	13	14	25	8	10	19	24	12	13	14	10	13	28	11
	26	16	19	15	34	9	13	20	26	16	19	15	16	14	29	15
	36	28	20	17	41	11	14	26	36	28	20	17	18	19	30	17
	39	29	25	18	44	15	17	31	39	29	25	18	26	22	36	20
	45	30	27	21	45	22	29	33	45	30	27	21	31	27	41	21
	48	34	31	22	59	24	30	35	48	34	31	22	32	34	45	25
	51	42	32	37	60	27	36	40	51	42	32	37	40	37	57	33
	54	44	33	40	62	28	37	42	54	44	33	40	42	38	61	35
	57	47	35	41	63	32	46	48	57	47	35	41	46	43	63	39
	61	49	38	43	65	38	47	54	49	38	43	61	52	44	65	48
	66	52	53	46	66	39	49	55	52	53	46	66	58	47	66	50
	71	55	58	50	69	43	51	61	55	58	50	71	59	49	69	53
	78	59	64	56	75	50	52	64	59	64	56	78	62	51	75	60
	91	62	65	60	81	56	53	67	62	65	60	91	64	54	76	67
	94	69	67	63	82	57	58	74	69	67	63	94	68	55	77	70
	96	76	72	68	92	68	70	76	76	72	68	96	80	56	81	72
		77	75	70		71	86	78		77	75	70		71	83	78
		80	82	73		72	87	83		80	82	73		73	87	79
		81	84	74		73	88	84		81	84	74		74	88	82
		85	87	79		77	89	85		85	87	79		86	89	84
		86	88	83		79	93	90		86	88	83		91	95	85
		89	90	92		80	94	96		89	90	92		92	96	90
		95	93	97		91	95	97		95	93	97		93	97	94
Clinical-stage mAbs with low specificity	Test set	Training set			Test set	Training set			Test set	Training set			Test set	Training set		
		A	B	C		A	B	C		A	B	C		A	B	C
	99	100	101	98	100	99	98	102	99	100	101	98	100	98	104	101
	105	102	103	104	112	101	105	103	105	102	103	104	102	99	105	107
	117	106	110	108	114	107	106	104	117	106	110	108	106	103	108	110
	118	107	111	112	115	109	108	111	118	107	111	112	118	114	109	111
	122	109	113	114	123	122	110	116	122	109	113	114	119	116	112	115
	127	115	116	119	128	124	113	118	127	115	116	119	123	117	113	120
	132	121	120	123	135	126	117	120	132	121	120	123	126	121	122	124
	134	124	125	133	136	129	119	121	134	124	125	133	137	132	125	127
		128	126	135		132	125	131		128	126	135		133	128	129
		129	130	136		134	127	133		129	130	136		135	131	130
		131	137			137	130			131	137			136	134	

80/20 Split	9				10			
	Test set	Training set			Test set	Training set		
		A	B	C		A	B	C
Clinical-stage mAbs with high specificity	17	1	3	2	2	4	3	1
	21	10	6	4	6	8	10	5
	32	18	12	5	7	9	12	15
	37	26	14	7	11	19	13	16
	38	28	15	8	14	26	23	17
	39	44	19	9	18	32	25	22
	42	45	22	11	20	33	28	27
	43	49	25	13	21	39	29	31
	66	52	29	16	24	40	34	36
	74	54	36	20	30	41	35	37
	75	55	41	23	43	47	51	38
	77	57	47	24	46	49	52	42
	79	58	50	27	55	50	53	44
	80	59	51	30	58	59	56	45
	81	60	53	31	72	63	64	48
	84	61	56	33	73	70	65	54
	87	62	63	34	87	71	67	57
	93	64	65	35	88	75	69	60
	97	71	68	40	89	78	76	61
		78	69	46		81	77	62
		82	70	48		83	79	66
		88	72	67		85	80	68
		90	83	73		90	84	74
		91	86	76		92	86	82
		92	95	85		93	91	94
		94	96	89		95	96	97
Clinical-stage mAbs with low specificity	Test set	Training set			Test set	Training set		
		A	B	C		A	B	C
	100	99	98	101	102	100	98	99
	105	107	102	103	105	106	101	103
	118	108	104	111	108	107	111	104
	127	110	106	113	115	109	122	112
	129	112	109	115	124	110	123	113
	132	117	114	116	125	116	126	114
	135	123	119	121	128	118	127	117
	137	124	120	122	135	119	129	131
		126	125	134		120	130	132
		131	128	136		121	134	133
		133	130			137	136	

Table S7. Summary of the constraints used to generate the single and combined sets of rules for flagging mAbs with low specificity.

	Constraints for single rules	**1st round**	**2nd round**
1	Accuracy for preclinical mAbs (PSR assay)	>55%	>55%
2	% mAbs (preclinical, training set) flagged with low specificity - high specificity (PSR assay)	>0	>0
3	Accuracy of training set (clinical mAbs) in each fold	>55%	>55%
	* Three constraints for each of the ten 80/20 splits		
4	Accuracy of training set (clinical mAbs, two folds) - accuracy of validation set (one fold)	<5%	<5%
	* Three constraints for each of the ten 80/20 splits		
5	% mAbs (clinical, training set) flagged with low specificity - high specificity for each assay	>0	>0
	* Five constraints for each individual assay (PSR, ELISA, BVP, AC-SINS, CSI)		
6	Average validation accuracy for clinical mAbs	>60%	>60%
	* Average validation accuracy is evaluated based on the results for each flag value observed in each of the ten 80/20 splits		
7	Average test accuracy for clinical mAbs	>50%	>50%
	* Average test accuracy is evaluated based on the results for each flag value observed in each of the ten 80/20 splits		

	Constraints for combined rules	**1st round**	**2nd round**
1	Accuracy for preclinical mAbs (PSR assay)	>60%	>70%
2	% mAbs (preclinical, training set) flagged with high specificity (PSR assay)	<10%	
3	Accuracy of training set (clinical mAbs) in each fold	>60%	>75%
	* Three constraints for each of the ten 80/20 splits		
4	Accuracy of training set (clinical mAbs, two folds) - accuracy of validation set (one fold)	<10%	<10%
	* Three constraints for each of the ten 80/20 splits		
5	% mAbs (clinical, training set) flagged with high specificity	<5%	<10%
6	Accuracy of training set (clinical mAbs) for each assay (PSR, ELISA, BVP, AC-SINS, CSI)	>60%	>70%
	* Five constraints for each individual assay (PSR, ELISA, BVP, AC-SINS, CSI)		
7	Accuracy for training set (clinical mAbs)	>60%	>75%

Table S9. Summary of the best combined sets of rules generated in the first round of analysis that flag mAbs with low specificity. Five sets (A, B, C, D and E) of rules are reported. The rules are numbered as defined in Table S8. mAbs with low and high specificity are defined as in Tables S1 and S2.

Set of flags	# of flags for mAs with low specificity	Individual rule numbers for each set of flags						Flag values for corresponding individual rule of each set of flags					
A	≥4	1	11	14	21	24	37	5.0	4.0	4.6	4.7	12.2	4.9
B	≥4	7	11	15	19	21	33	3.6	4.0	4.2	3.0	4.8	17.8
C	≥5	9	13	19	21	23	33	3.6	7.5	3.0	4.8	2.1	17.8
D	≥4	7	11	15	19	21	33	3.6	4.0	4.2	3.0	4.8	16.7
E	≥5	3	7	11	21	23	33	4.7	3.6	4.0	4.8	2.1	17.8

	Clinical mAbs (non-specific and self-interactions, five assays)								
Set of flags	**% mAbs flagged (high specificity group)**	**% mAbs flagged (low specificity group)**	**Average validation accuracy (%)**	**Std. dev. of validation accuracy (%)**	**COV for validation accuracy (%)**	**Average test accuracy (%)**	**Test accuracy Stdev (%)**	**COV for test accuracy (%)**	***p*-value for entire panel of clinical mAbs**
A	2.06	35.00	66.17	0.87	1.32	67.43	3.68	5.45	3.78E-07
B	2.06	40.00	69.07	0.93	1.35	68.42	3.74	5.46	2.15E-08
C	2.06	40.00	68.48	0.96	1.41	70.92	3.91	5.51	2.15E-08
D	0.00	37.50	68.71	1.06	1.55	68.75	4.17	6.06	1.04E-09
E	3.09	47.50	72.18	1.22	1.69	72.01	4.98	6.92	1.31E-09

	Preclinical mAbs (non-specific interactions, PSR assay)			
Set of flags	**% mAbs flagged (high specificity group)**	**% mAbs flagged (low specificity group)**	**Training accuracy (%)**	***p*-value for preclinical mAbs**
A	9.32	44.44	67.56	7.16E-06
B	9.07	44.44	67.69	5.61E-06
C	5.04	33.33	64.15	1.86E-05
D	6.55	29.63	61.54	5.54E-04
E	8.56	40.74	66.09	2.12E-05

Table S10. Segregation of mAbs that pass the combined specificity rules in the first round of analysis into training and test sets. Clinical-stage mAbs (as numbered in Table S1) that pass the combined rules in Set A (<4 flags, 121 out of 137 mAbs; Table S8) were divided into ten sets of 80% training mAbs and 20% test mAbs. For each 80/20 split, the training sets of mAbs (80%) was further divided into three groups for threefold cross validation.

80/20 Split	1				2				3				4			
	Test set	Training set			Test set	Training set			Test set	Training set			Test set	Training set		
		A	B	C		A	B	C		A	B	C		A	B	C
Clinical-stage mAbs with high specificity	9	4	1	2	2	6	3	1	3	1	6	2	9	1	2	4
	10	5	3	16	4	12	5	8	13	5	7	4	11	6	3	5
	11	6	7	20	7	13	16	9	28	9	11	8	14	13	8	7
	18	13	8	22	11	19	17	10	29	12	18	10	23	16	20	10
	23	14	12	25	14	20	23	15	35	15	19	14	25	21	24	12
	26	17	15	27	18	25	30	22	43	17	20	16	29	33	26	15
	28	19	24	29	21	26	35	24	53	26	21	22	35	37	28	17
	39	21	31	34	34	27	42	28	54	33	27	23	38	42	32	18
	42	30	33	38	40	29	45	33	55	38	30	24	39	47	34	19
	47	32	35	40	51	31	52	41	57	40	32	25	41	49	36	22
	51	37	36	41	59	32	56	43	63	41	37	31	43	50	44	27
	58	44	45	43	61	36	58	47	65	42	46	34	51	52	45	30
	64	54	46	48	66	37	68	48	71	44	47	36	53	54	46	31
	68	55	49	50	74	38	71	49	76	59	48	39	55	61	48	40
	74	56	59	52	76	39	72	50	78	61	51	45	64	63	56	62
	76	57	63	53	86	44	78	53	85	62	73	49	66	65	57	69
	77	65	66	61	87	46	80	55	87	64	74	50	81	67	58	70
	90	71	70	62	91	54	81	57	91	66	77	52	84	72	59	74
	94	75	73	67	96	63	82	62	96	67	79	56	91	79	68	76
		78	83	69		65	83	64		68	80	58		83	71	77
		80	84	72		73	84	67		70	83	69		85	73	80
		81	85	79		79	85	69		72	88	75		86	75	82
		86	87	82		89	88	70		82	92	81		90	78	87
		91	88	89		95	90	75		86	94	84		92	89	88
		96	92	95		97	94	77		89	97	90		95	96	94
				97				92				95				97
	Test set	Training set			Test set	Training set			Test set	Training set			Test set	Training set		
		A	B	C		A	B	C		A	B	C		A	B	C
Clinical-stage mAbs with low specificity	102	99	98	107	103	102	110	98	108	102	99	98	107	102	110	98
	103	108	115	110	107	118	111	99	113	103	106	112	112	103	111	99
	106	111	121	112	113	119	117	106	115	107	110	117	117	106	118	108
	118	113	127	129	115	121	128	108	134	119	111	121	135	115	119	113
	122	117	128	131	135	122	131	112	137	122	118	127	136	127	121	122
		119	134	135		133	134	127		128	129	133		128	134	129
		133	136	137		137	136	129		131	136	135		131	137	133

80/20 Split	5				6				7				8			
	Test set	Training set			Test set	Training set			Test set	Training set			Test set	Training set		
		A	B	C		A	B	C		A	B	C		A	B	C
Clinical-stage mAbs with high specificity	6	8	2	1	4	5	1	2	3	2	1	5	11	2	1	9
	9	10	11	3	7	8	6	3	6	8	4	12	19	3	4	12
	13	14	12	4	14	17	9	10	7	10	9	13	20	5	7	13
	19	17	15	5	15	19	13	11	11	15	20	14	23	6	8	17
	20	23	16	7	39	20	23	12	22	21	29	16	27	14	10	22
	25	33	21	18	44	21	24	16	25	24	33	17	29	15	16	24
	28	36	26	22	48	22	25	18	27	26	35	18	30	18	21	28
	31	37	27	24	56	26	29	27	30	28	36	19	38	32	25	31
	38	47	29	30	59	37	32	28	32	31	38	23	40	34	26	36
	43	48	32	35	72	40	33	30	43	37	40	34	48	39	33	42
	56	49	34	40	73	49	41	31	50	41	42	39	49	41	35	46
	62	58	39	50	78	50	42	34	52	47	53	44	58	43	37	47
	65	61	41	52	80	51	47	35	58	48	55	45	62	53	44	50
	66	64	42	53	81	53	61	36	63	59	57	46	63	54	45	56
	80	67	44	54	83	55	63	38	68	61	64	49	67	55	51	61
	81	69	45	55	86	58	64	43	73	66	70	51	77	57	52	64
	82	70	46	63	87	62	65	45	85	67	75	54	81	59	66	71
	88	71	51	68	90	66	74	46	86	71	76	56	89	65	69	72
	92	72	57	75	92	67	76	52	94	72	80	62	95	68	75	74
		73	59	78		69	82	54		74	83	65		70	76	78
		76	74	83		70	84	57		79	84	69		73	79	80
		77	84	86		79	89	68		87	88	77		82	91	83
		79	89	87		85	91	71		92	90	78		84	94	85
		85	91	90		96	94	75		96	91	81		87	96	86
		95	94	96		97	95	77		97	95	82		92	97	88
				97				88				89				90
	Test set	Training set			Test set	Training set			Test set	Training set			Test set	Training set		
		A	B	C		A	B	C		A	B	C		A	B	C
Clinical-stage mAbs with low specificity	99	107	102	98	108	102	99	98	99	98	107	102	106	98	103	99
	108	110	103	106	111	103	113	106	119	112	110	103	113	108	107	102
	111	112	118	115	118	107	115	110	128	113	111	106	118	110	117	111
	113	117	122	119	119	117	121	112	131	115	118	108	119	112	127	122
	121	128	129	127	133	131	122	128	134	121	122	117	121	115	131	128
		133	131	135		136	127	129		129	133	127		135	133	129
		136	134	137		137	135	134		137	135	136		136	134	137

80/20 Split	9				10			
	Test set	Training set			Test set	Training set		
		A	B	C		A	B	C
Clinical-stage mAbs with high specificity	2	3	1	9	4	10	2	1
	14	6	4	10	7	11	3	5
	22	11	5	13	8	15	13	6
	23	12	7	17	9	17	19	16
	38	19	8	18	12	18	25	20
	42	20	15	24	14	24	27	21
	46	26	16	25	22	30	31	23
	51	28	21	27	28	37	32	26
	56	32	35	29	34	40	36	29
	58	33	37	30	35	41	38	33
	61	36	40	31	39	48	46	42
	63	44	41	34	43	58	47	45
	69	48	43	39	44	59	50	53
	70	52	45	49	49	67	52	56
	83	53	47	50	51	69	54	57
	84	55	54	67	55	70	61	63
	87	57	64	68	72	74	62	64
	91	59	65	76	85	78	66	65
	96	62	71	77	87	79	68	76
		66	74	78		80	71	77
		72	80	79		83	73	81
		73	85	81		86	75	82
		75	92	82		88	84	90
		86	94	89		91	89	92
		88	95	90		95	96	94
				97				97
	Test set	Training set			Test set	Training set		
		A	B	C		A	B	C
Clinical-stage mAbs with low specificity	103	106	98	111	103	102	98	106
	108	107	99	113	108	107	99	110
	115	112	102	118	113	111	112	118
	122	117	110	119	127	117	115	121
	129	121	131	127	136	122	119	134
		134	133	128		128	129	135
		135	136	137		131	133	137

Table S12. Summary of the best combined sets of rules generated after the second round of analysis that identify mAbs with low specificity. Each of the reported sets of rules generated in the second round of analysis (F, G, H, I and J) were derived in combination with Set A from the first round of analysis (as defined in Table S8). The rules are numbered as defined in Table S11. mAbs with low and high specificity are defined as in Tables S1 and S2.

Set of flags	# of flags for mAbs with low specificity	Individual rule numbers for each set of flags						Flag values for corresponding individual rule of each set of flags					
F	≥8	11	18	31	32	36	39	23.2	2.8	2.2	3.9	19.2	19.8
G	≥8	11	18	31	32	36	42	23.2	2.8	2.2	3.9	19.2	19.8
H	≥8	10	21	30	31	36	41	25.5	35.7	2.9	2.2	19.9	19.0
I	≥8	10	18	21	31	36	41	25.5	2.8	35.7	2.2	19.9	19.0
J	≥8	6	21	31	32	36	37	24.3	35.7	2.2	3.9	19.3	3.6

	Clinical mAbs (non-specific and self-interactions, five assays)								
Set of flags	**% mAbs flagged (high specificity group)**	**% mAbs flagged (low specificity group)**	**Average validation accuracy (%)**	**Std. dev. of validation accuracy (%)**	**COV for validation accuracy (%)**	**Average test accuracy (%)**	**Test accuracy Stdev (%)**	**COV for test accuracy (%)**	***p*-value for entire panel of clinical mAbs**
F	8.25	77.50	83.20	1.44	1.73	90.16	5.84	6.48	1.55E-15
G	8.25	77.50	83.20	1.44	1.73	90.16	5.84	6.48	1.55E-15
H	7.22	77.50	83.59	1.72	2.06	91.22	6.97	7.64	3.50E-16
I	7.22	77.50	83.52	1.73	2.08	91.48	7.06	7.72	3.50E-16
J	7.22	77.50	83.71	1.76	2.10	90.69	7.18	7.91	3.50E-16

	Preclinical mAbs (non-specific interactions, PSR assay)			
Set of flags	**% mAbs flagged (high specificity group)**	**% mAbs flagged (low specificity group)**	**Training accuracy (%)**	***p*-value for preclinical mAbs**
F	16.22	55.00	70.27	1.71E-05
G	16.52	55.00	70.02	2.27E-05
H	19.47	60.00	70.11	3.04E-05
I	18.58	60.00	70.74	1.56E-05
J	17.70	55.00	70.99	1.18E-05

Table S15. Sets of four mutations in Fab sub-libraries of emibetuzumab that are most strongly correlated with reduced binding to polyspecificity reagents [PSR and ovalbumin (OVA)]. The libraries were designed and evaluated as described in Fig. 8. The *p* -values are for the Spearman's correlation coefficients (ρ).

CDR	H1	H2					H3		# of clones		Spearman's correlation coefficient and *p*-value								Avg ρ
											Repeat 1				Repeat 2				
	33	50	54	55	56	95	97	102				*p*-value		*p*-value					
WT	Y	R	R	R	G	A	W	Y	with 4WT	with 4MT	ρ (PSR)	(PSR)	ρ (OVA)	(OVA)	ρ (PSR)	*p*-value (OVA)	ρ (OVA)	*p*-value (OVA)	
1	F		T		**D**			A	10	12	0.81	1E-05	0.83	6E-05	0.71	2E-03	0.83	1E-06	0.80
2	F		T		**D**			**D**	10	14	0.73	3E-04	0.87	4E-05	0.76	7E-05	0.78	7E-06	0.79
3	V		T	G	**D**				26	24	0.69	5E-05	0.85	9E-10	0.61	4E-05	0.76	1E-08	0.73
4					**D**	S	L	V	10	10	0.75	2E-04	0.69	2E-03	0.75	7E-04	0.69	7E-04	0.72
5	V				**D**		L	**D**	13	16	0.68	2E-04	0.70	4E-04	0.85	3E-08	0.65	2E-04	0.72
6		**K**			**D**		L	**D**	14	17	0.67	2E-04	0.73	7E-05	0.83	1E-08	0.63	3E-04	0.71
7	V		T		**D**			**D**	10	14	0.64	4E-03	0.75	3E-04	0.73	6E-04	0.72	2E-04	0.71
8	F			G	**D**		G		42	21	0.63	2E-06	0.81	3E-11	0.68	7E-08	0.64	3E-08	0.69
9				G	**D**		L	V	10	16	0.66	5E-04	0.72	4E-04	0.70	3E-04	0.68	2E-04	0.69
10	F			G	**D**		L		42	16	0.66	2E-06	0.75	1E-07	0.63	6E-07	0.69	1E-08	0.68
11				G	**D**	S	L		41	39	0.64	1E-07	0.69	1E-07	0.63	2E-08	0.73	3E-13	0.67
12				G	**D**		S	S	10	18	0.66	2E-04	0.62	1E-03	0.67	1E-04	0.67	9E-05	0.65
13					**D**	S	S	S	10	23	0.64	3E-04	0.66	3E-04	0.62	2E-04	0.63	8E-05	0.64

Table S16. Theoretical net charges and isoelectric points for the high specific clinical-stage antibodies in this study. The net charges (pH 7.4) were calculated by assigning values of +1 for R and K, +0.1 for H, and -1 for D and E. The CDRs were defined using a combination of Chothia and Kabat numbering, and heavy chain CDR3 was defined to also include two additional N-terminal residues. The values are averages with their corresponding standard deviations. These ranges (average ± standard deviation) are defined only for the high specific mAbs (97 of 137 mAbs), while the ranges in Table S5 (minimum and maximum) are defined for both high and low specific mAbs (137 mAbs). Antibodies with charge and isoelectric properties outside of these ranges are not expected to be well described by the chemical rules. In particular, antibodies with more negatively charged CDRs or variable regions as well as lower isoelectric points than those for the high specific antibodies in this study are at risk for abnormal behavior, such as viscous behavior at high antibody concentrations.

Antibody region	**pI** avg ± stdev	**net charge (pH 7.4)** avg ± stdev
H1		-0.17±0.54
H2		0.01±1.32
H3		-0.45±1.07
H123		-0.61±1.71
L1		0.43±1.16
L2		0.12±0.97
L3		0.01±0.79
L123		0.56±1.90
CDR		-0.05±2.44
VH	7.4±1.4	0.75±1.89
VL	7.5±1.4	0.79±1.97
Fv	7.7±1.3	1.54±2.48
framework	7.9±1.0	1.59±1.52

Table S17. Number of chemical flags for the clinical-stage antibodies calculated using two different SASA machine learning models. The two models for calculating solvent-accessible surface areas were generated using a published method [Jain et al., *Bioinformatics*, 33, 3758-3766 (2017)]. Model A is used for calculating the chemical rules for the clinical-stage antibodies in Table S1, and Model B is used for calculating the chemical rules for preclinical antibodies in Table S2 and S13.

Antibody	Specificity	# of chemical flags from model A	# of chemical flags from model B	Antibody	Specificity	# of chemical flags from model A	# of chemical flags from model B
abituzumab	high	7	7	mepolizumab	high	4	5
abrilumab	high	6	6	mogamulizumab	high	2	2
adalimumab	high	2	2	motavizumab	high	3	3
alemtuzumab	high	3	3	muromonab	high	8	8
alirocumab	high	1	1	natalizumab	high	7	7
anifrolumab	high	6	6	necitumumab	high	6	7
bapineuzumab	high	5	5	nimotuzumab	high	4	4
benralizumab	high	7	7	nivolumab	high	3	3
bevacizumab	high	5	5	obinutuzumab	high	5	5
brentuximab	high	3	3	ocrelizumab	high	7	7
brodalumab	high	9	9	ofatumumab	high	6	6
canakinumab	high	7	7	olaratumab	high	9	9
certolizumab	high	6	6	olokizumab	high	3	3
cetuximab	high	3	3	omalizumab	high	6	6
clazakizumab	high	2	1	onartuzumab	high	3	3
crenezumab	high	4	3	otelixizumab	high	3	3
dacetuzumab	high	6	6	otlertuzumab	high	5	5
daclizumab	high	6	6	palivizumab	high	3	3
daratumumab	high	6	6	panitumumab	high	3	4
dinutuximab	high	7	8	panobacumab	high	5	5
eculizumab	high	2	2	pembrolizumab	high	5	5
efalizumab	high	5	6	pertuzumab	high	7	7
eldelumab	high	5	5	pinatuzumab	high	6	6
elotuzumab	high	1	1	polatuzumab	high	5	5
enokizumab	high	8	9	radretumab	high	4	4
epratuzumab	high	4	4	ramucirumab	high	4	4
evolocumab	high	5	6	ranibizumab	high	6	6
farletuzumab	high	6	6	reslizumab	high	4	4
fasinumab	high	5	6	rilotumumab	high	7	8
fezakinumab	high	4	4	rituximab	high	6	6
ficlatuzumab	high	6	6	romosozumab	high	5	5
fletikumab	high	5	5	sarilumab	high	4	4
fresolimumab	high	5	5	secukinumab	high	6	7
fulranumab	high	5	5	seribantumab	high	4	5
galiximab	high	3	2	sifalimumab	high	7	7
gemtuzumab	high	2	2	siltuximab	high	6	6
gevokizumab	high	6	6	tabalumab	high	8	8
girentuximab	high	4	4	tanezumab	high	6	6
glembatumumab	high	8	8	tigatuzumab	high	5	4
golimumab	high	7	7	tildrakizumab	high	6	6
ibalizumab	high	4	4	tocilizumab	high	5	5
ipilimumab	high	5	4	tovetumab	high	9	9
lampalizumab	high	2	2	tralokinumab	high	2	2
lebrikizumab	high	3	3	trastuzumab	high	3	3
lintuzumab	high	6	6	tremelimumab	high	11	11
lumiliximab	high	3	3	vedolizumab	high	7	7
matuzumab	high	5	5	veltuzumab	high	7	7
mavrilimumab	high	2	2	zalutumumab	high	6	6

Antibody	Specificity	# of chemical flags from model A	# of chemical flags from model B
zanolimumab	high	5	5
atezolizumab	low	9	9
basiliximab	low	9	9
bavituximab	low	10	10
belimumab	low	8	9
bimagrumab	low	9	9
blosozumab	low	5	5
bococizumab	low	11	11
briakinumab	low	9	8
carlumab	low	6	6
cixutumumab	low	8	8
codrituzumab	low	7	7
dalotuzumab	low	8	8
denosumab	low	6	7
drozitumab	low	4	4
duligotuzumab	low	6	6
dupilumab	low	8	7
emibetuzumab	low	8	8
etrolizumab	low	8	8
figitumumab	low	8	8
foralumab	low	9	9
ganitumab	low	8	8
gantenerumab	low	8	8
guselkumab	low	8	8
imgatuzumab	low	8	8
infliximab	low	7	7
inotuzumab	low	10	10
ixekizumab	low	9	9
lenzilumab	low	9	9
lirilumab	low	10	10
ozanezumab	low	8	8
parsatuzumab	low	8	8
patritumab	low	9	9
ponezumab	low	8	8
robatumumab	low	7	7
simtuzumab	low	10	10
sirukumab	low	8	8
teplizumab	low	8	8
urelumab	low	9	9
ustekinumab	low	6	6
visilizumab	low	9	9

	H1	H2				H3		
	33	50	54	55	56	95	97	102
Degenerate codon	KHY	RVR	RVR	RVR	RVY	KHY	KBG	KHY
WT	Y	R	R	R	G	A	W	Y
1	F	K	K	K	A	F	V	F
2	V	A	A	A	D	Y	L	V
3	A	G	G	G	N	V	A	A
4	D	E	E	E	S	D	G	D
5	S	T	T	T	T	S	S	S

Figure S1. Design of a mutant V_H library for single-chain Fab fragments of emibetuzumab aimed at reducing the number of chemical flags. Eight sites in the CDRs flagged by the maximum chemical rules in Fig. 4 were mutated using degenerate codons that are designed to sample the wild-type residue and five mutations that reduce the number of chemical flags.

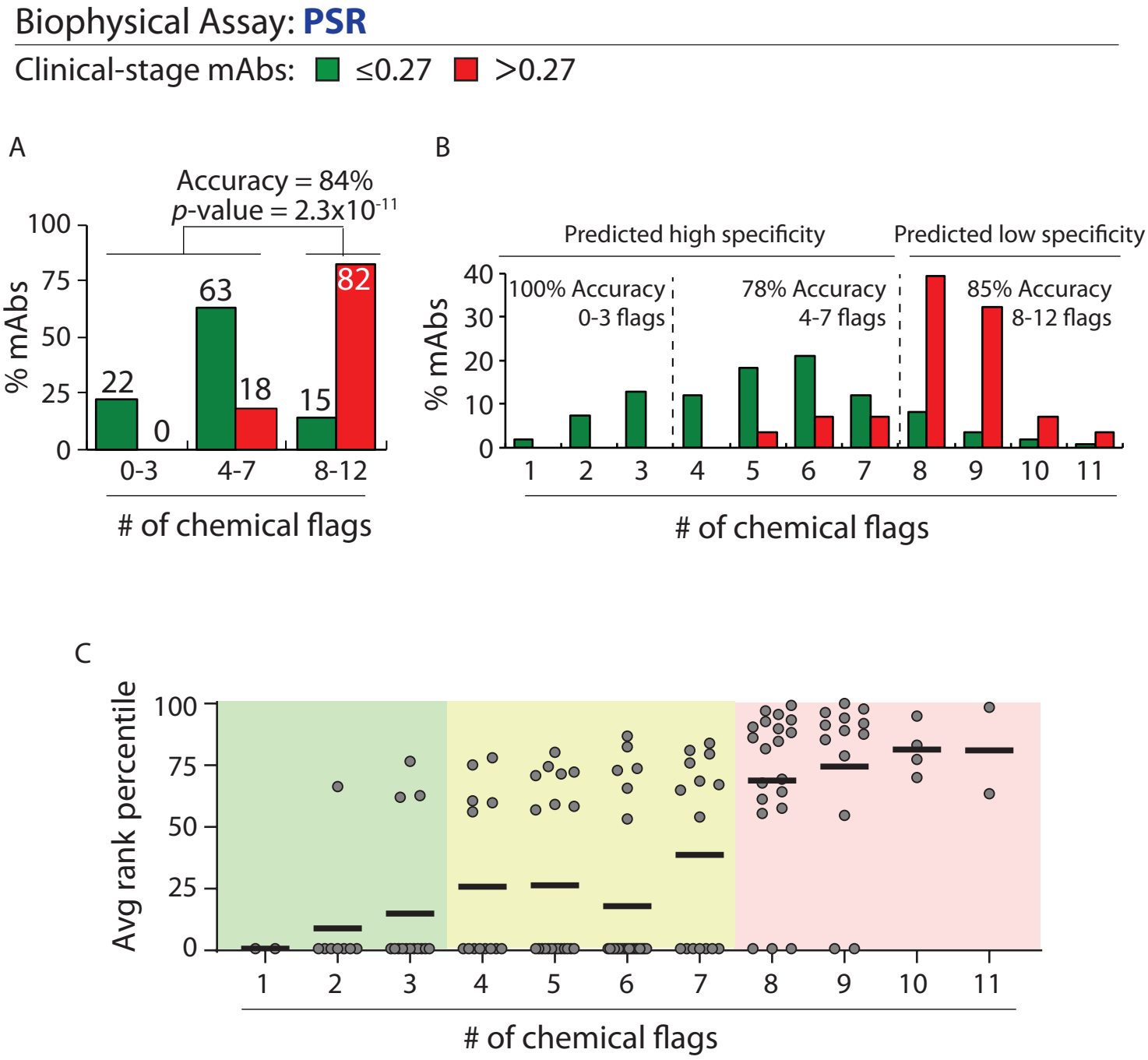

Figure S2. Performance of combined chemical rules for identifying clinical-stage mAbs with high levels of non-specific interactions detected using the PSR assay. (A, B) Evaluation of the percentage of mAbs with high and low specificity flagged by the combined set of chemical rules for (A) grouped numbers of chemical flags and (B) individual numbers of chemical flags. (C) Comparison of the rank for clinical-stage mAbs based on PSR measurements and the corresponding number of chemical flags. In (A) and (B), the *p*-values were calculated using a 2x2 contingency table (Fisher's exact test). In (C), three regions are shown, one with predicted high specificity (0-3 chemical flags), a second one with intermediate specificity (4-7 chemical flags), and a third one with low specificity (8-12 chemical flags).

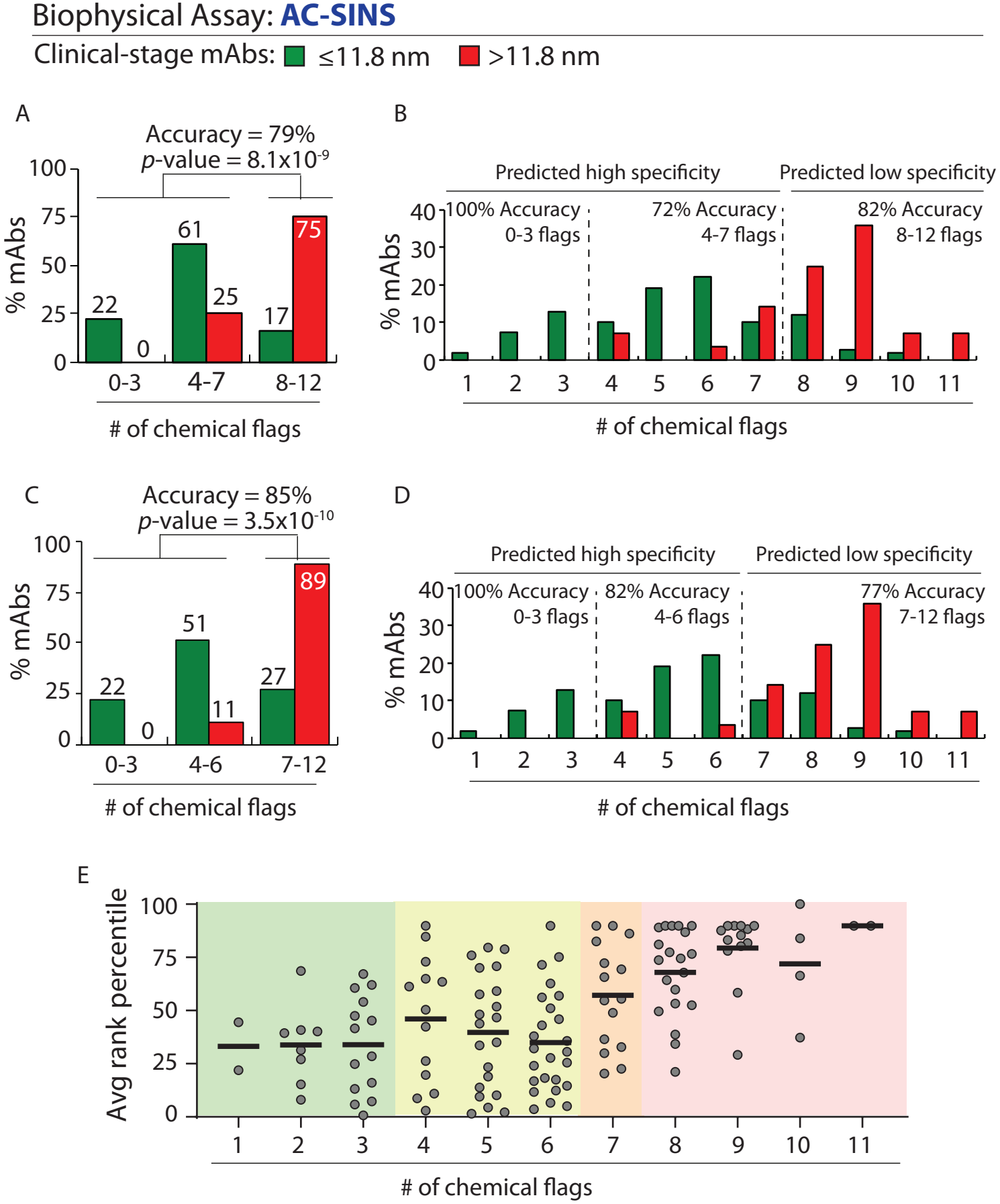

Figure S3. Performance of combined chemical rules for identifying clinical-stage mAbs with high levels of self-interactions detected using the AC-SINS assay. (A-D) Evaluation of the percentage of mAbs with high and low specificity flagged by the combined set of chemical rules for (A, C) grouped numbers of chemical flags and (B, D) individual numbers of chemical flags. (E) Comparison of the rank for clinical-stage mAbs based on AC-SINS measurements and the corresponding number of chemical flags. In (A-D), the *p*-values were calculated using a 2x2 contingency table (Fisher's exact test). In (E), three regions are shown, one with predicted high specificity (0-3 chemical flags), a second one with intermediate specificity (4-6 or 4-7 flags chemical flags), and a third one with low specificity (7-12 or 8-12 chemical flags).

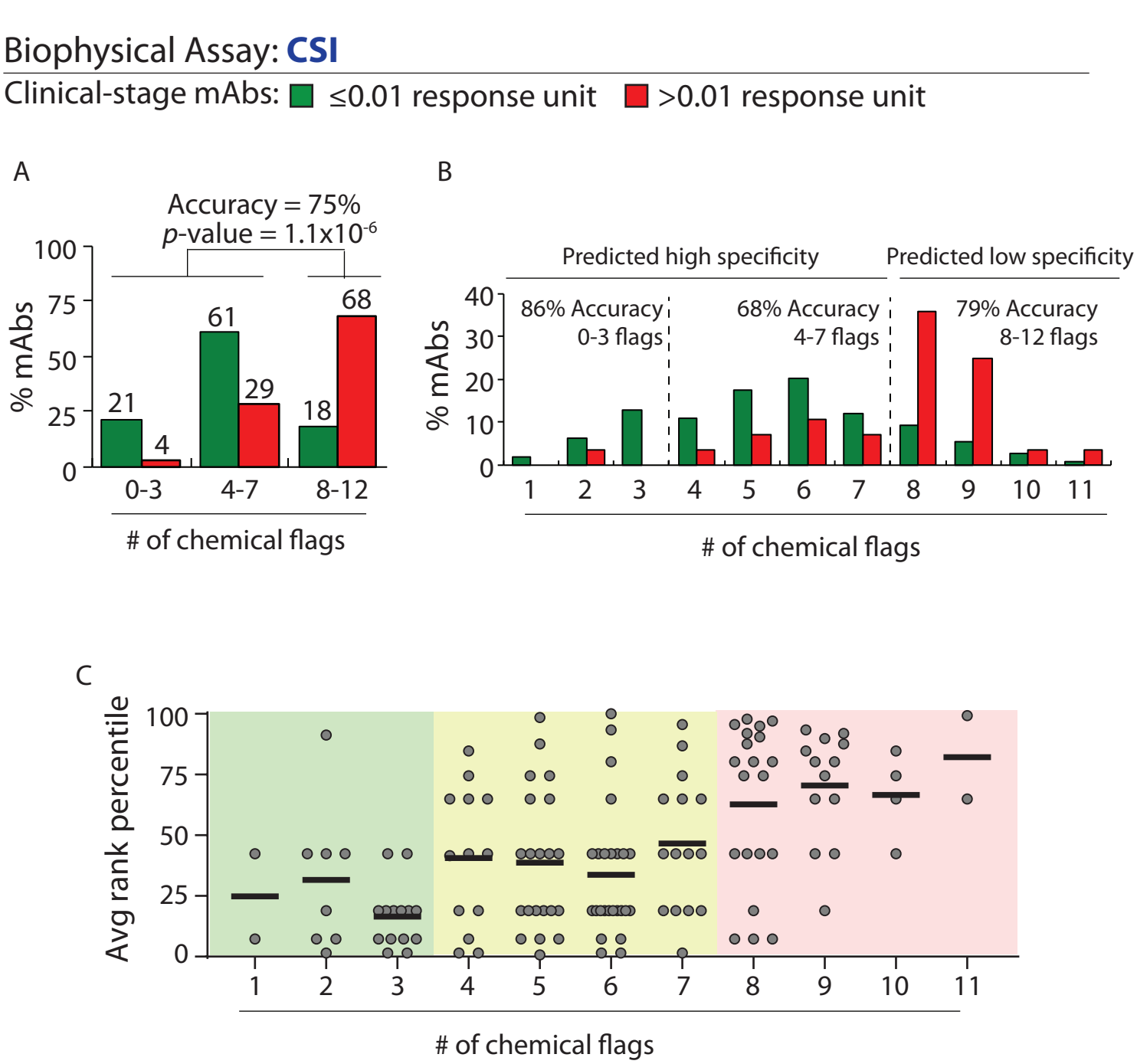

Figure S4. Performance of combined chemical rules for identifying clinical-stage mAbs with high levels of self-interactions detected using the CSI assay. (A, B) Evaluation of the percentage of mAbs with high and low specificity flagged by the combined set of chemical rules for (A) grouped numbers of chemical flags and (B) individual numbers of chemical flags. (C) Comparison of the rank for clinical-stage mAbs based on CSI measurements and the corresponding number of chemical flags. In (A) and (B), the *p*-values were calculated using a 2x2 contingency table (Fisher's exact test). In (C), three regions are shown, one with predicted high specificity (0-3 chemical flags), a second one with intermediate specificity (4-7 chemical flags), and a third one with low specificity (8-12 chemical flags).

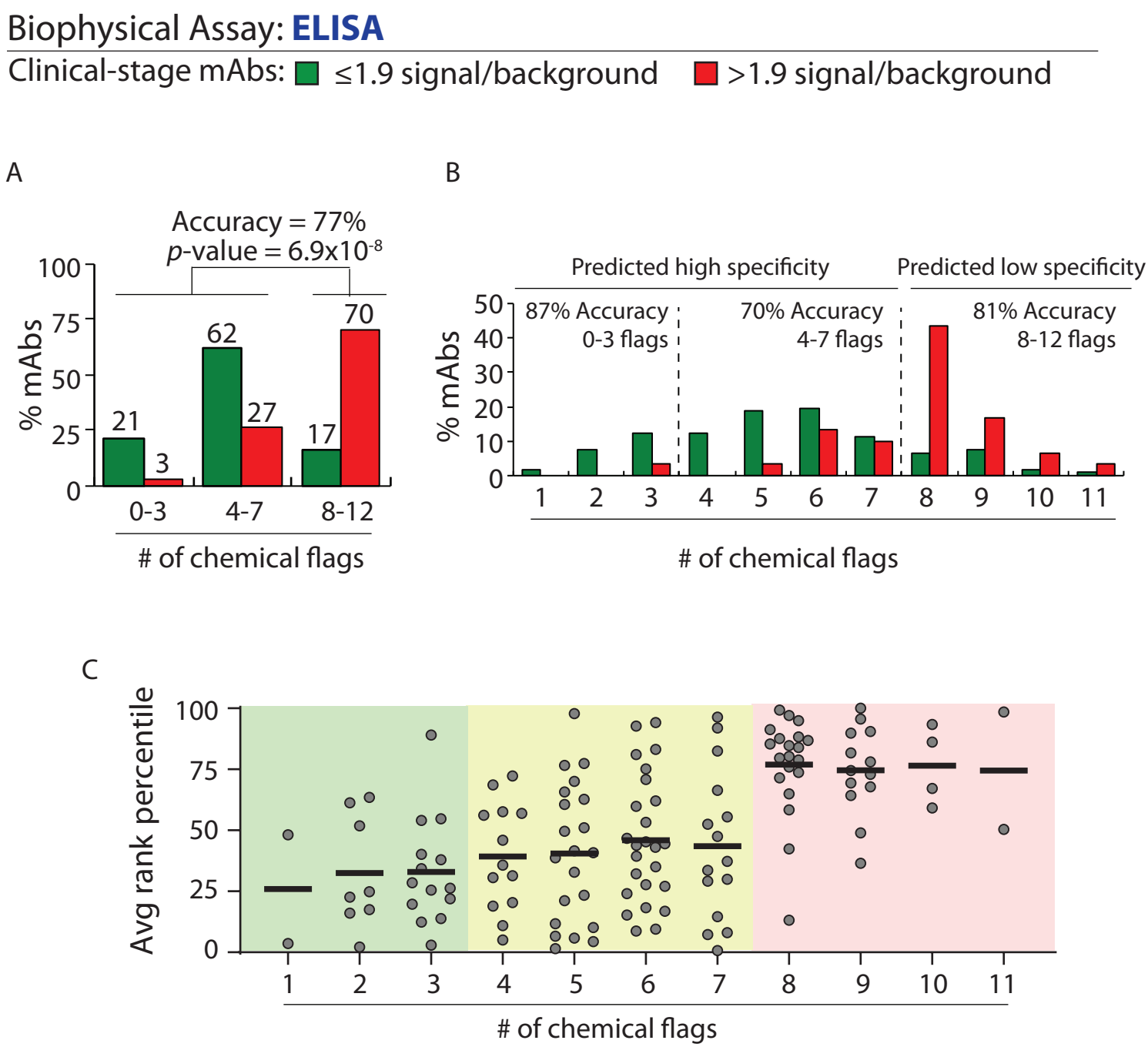

Figure S5. Performance of combined chemical rules for identifying clinical-stage mAbs with high levels of non-specific interactions detected using the ELISA non-specific binding assay. (A, B) Evaluation of the percentage of mAbs with high and low specificity flagged by the combined set of chemical rules for (A) grouped numbers of chemical flags and (B) individual numbers of chemical flags. (C) Comparison of the rank for clinical-stage mAbs based on ELISA measurements and the corresponding number of chemical flags. In (A) and (B), the *p*-values were calculated using a 2x2 contingency table (Fisher's exact test). In (C), three regions are shown, one with predicted high specificity (0-3 chemical flags), a second one with intermediate specificity (4-7 chemical flags), and a third one with low specificity (8-12 chemical flags).

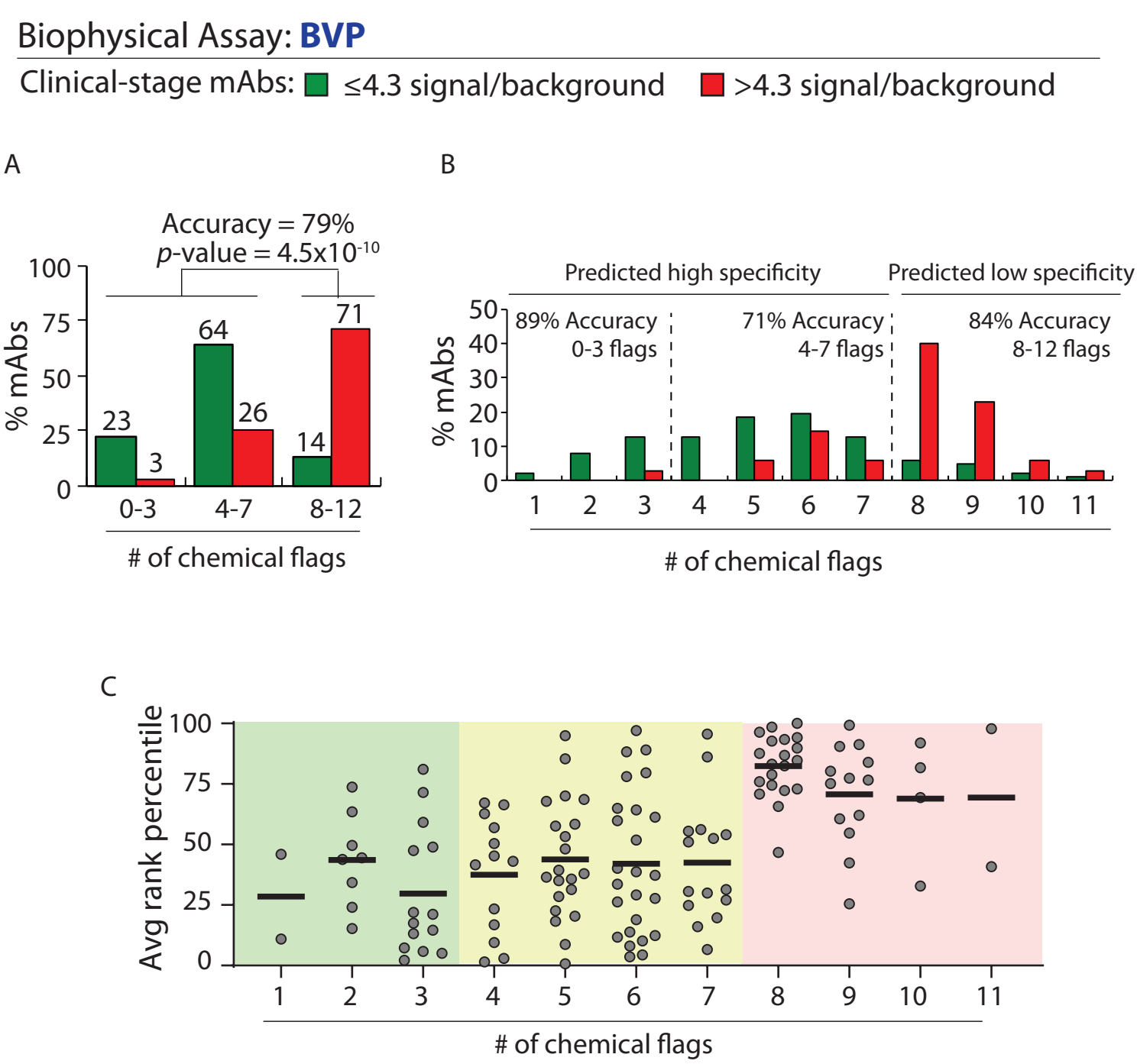

Figure S6. Performance of combined chemical rules for identifying clinical-stage mAbs with high levels of non-specific interactions detected using the BVP assay. (A, B) Evaluation of the percentage of mAbs with high and low specificity flagged by the combined set of chemical rules for (A) grouped numbers of chemical flags and (B) individual numbers of chemical flags. (C) Comparison of the rank for clinical-stage mAbs based on BVP measurements and the corresponding number of chemical flags. In (A) and (B), the *p*-values were calculated using a 2x2 contingency table (Fisher's exact test). In (C), three regions are shown, one with predicted high specificity (0-3 chemical flags), a second one with intermediate specificity (4-7 chemical flags), and a third one with low specificity (8-12 chemical flags).

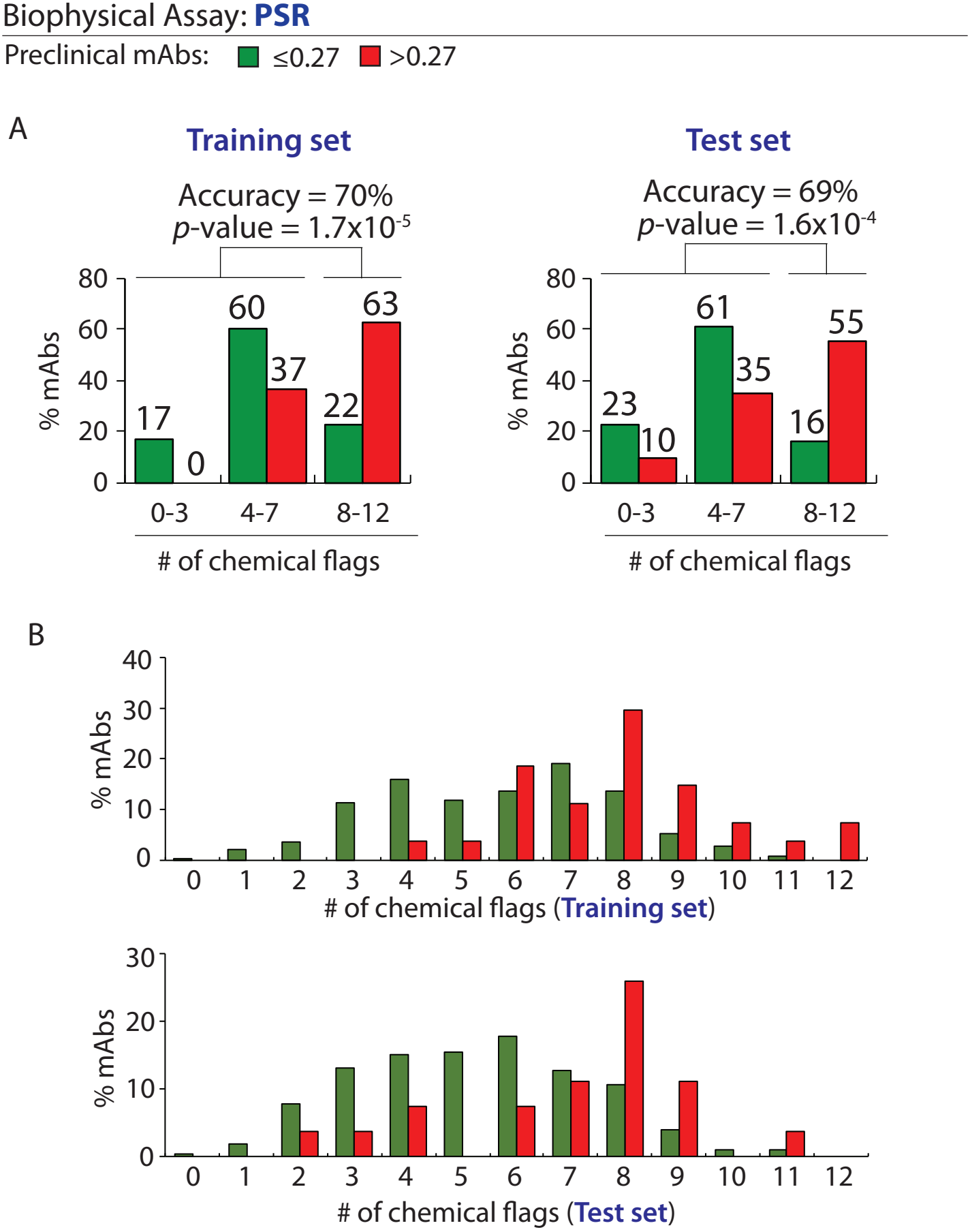

Figure S7. Combined chemical rules selectively flag preclinical mAbs with high levels of non-specific interactions. Antibodies with predicted high specificity (as described in Fig. 4) display reduced levels of non-specific binding to a poly-specificity reagent (PSR). (A) The selectivity of the combined chemical rules is similar for the training and test sets of preclinical antibodies. (B) Distributions of the number of flags for the training and test sets of antibodies with low and high specificity. The *p*-value was calculated using a 2x2 contingency table (Fisher's exact test).