



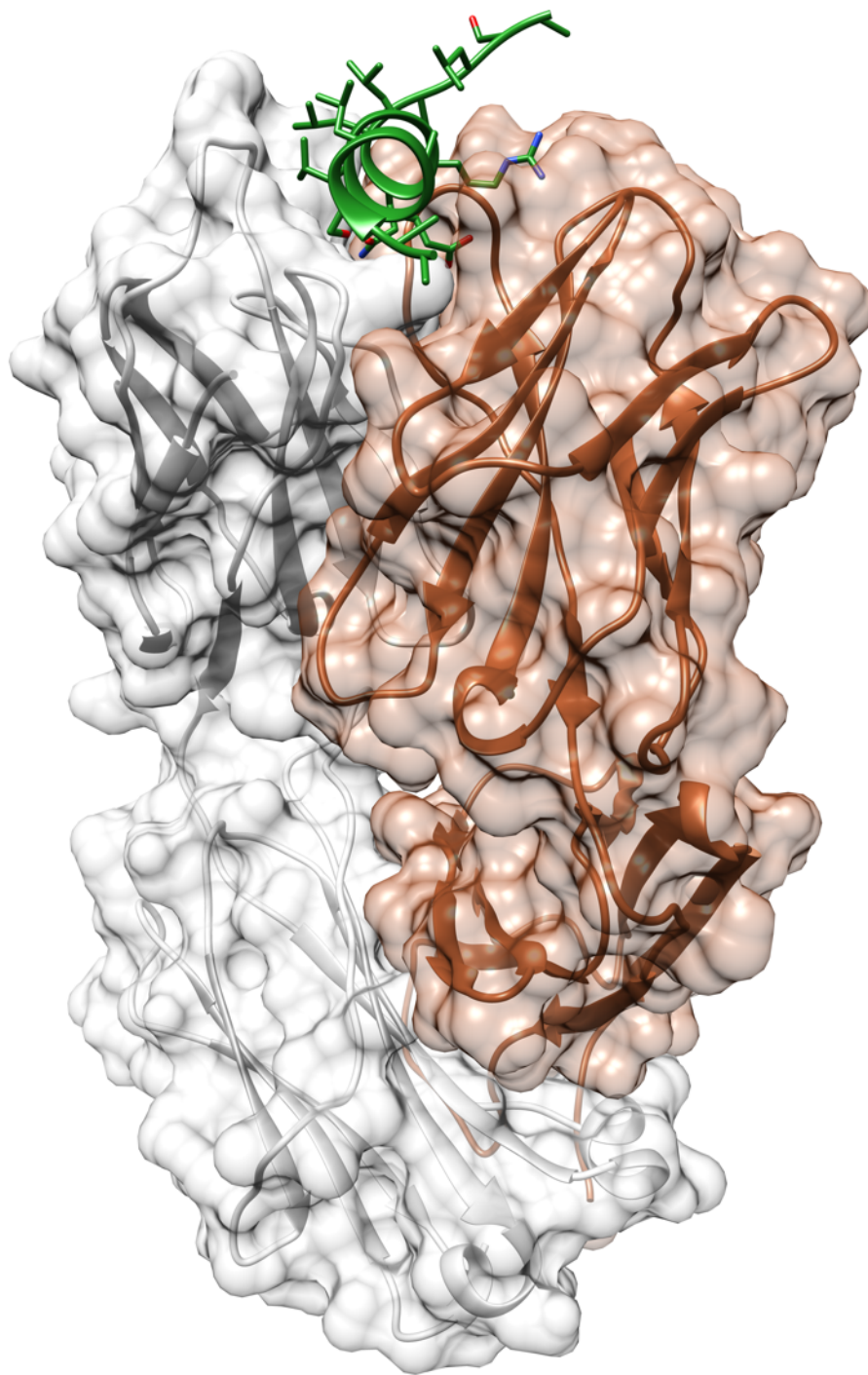
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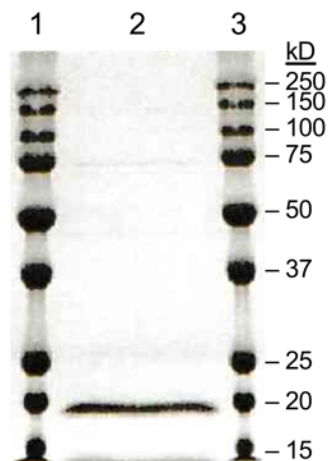
Supporting information for article:

Monoclonal antibody 7H2.2 binds the C-terminus of the cancer oocyte antigen SAS1B through the hydrophilic face of a conserved amphipathic helix corresponding to one of only two regions predicted to be ordered

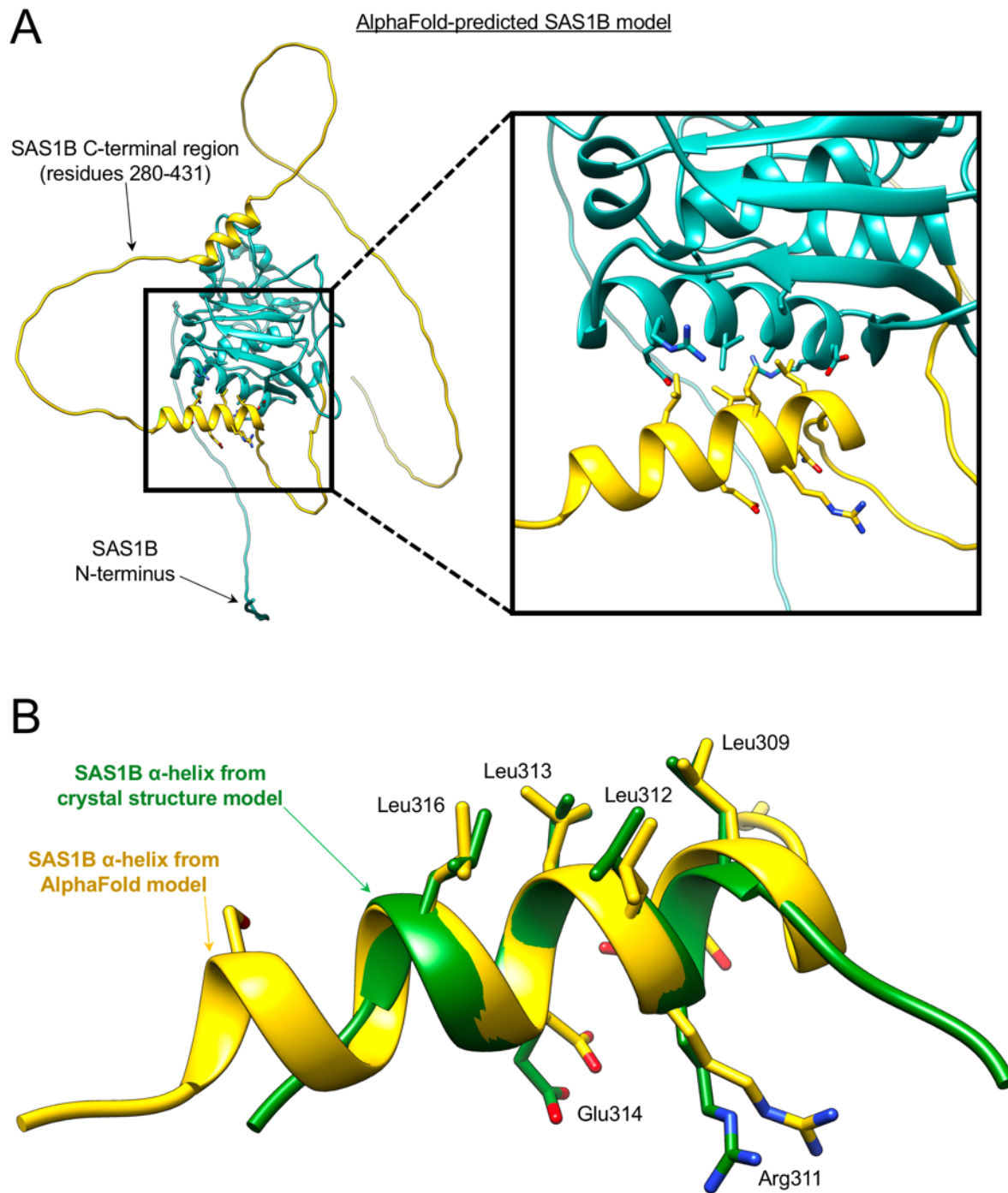
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Supplementary Figure S1. Crystal structure of mAb 7H2.2 Fab in complex with the SAS1B C-terminal antigen. One copy of the Fab molecule and one copy of the antigen molecule are present in the asymmetric unit. The Fab heavy and light chains are colored *brown* and *grey*, respectively, while the SAS1B antigen is colored *green*, with SAS1B residue sidechain atoms depicted in *green* for carbon, *red* for oxygen, and *blue* for nitrogen.



Supplementary Figure S2. SDS-PAGE of purified SAS1B C-terminal expression construct (*lane 2*) obtained after a two-step purification *via* immobilized metal affinity chromatography (HisPur™ Ni-NTA resin; Thermo Fisher Scientific) and cation-exchange chromatography (Shodex CM-825 column; Phenomenex Inc.). The Bio-Rad Precision Plus™ Dual Color Standard (shown in *lanes 1* and *3*) was used for molecular weight comparison with purified SAS1B sample.



Supplementary Figure S3. The AlphaFold-predicted model of human SAS1B contains a C-terminal α -helix corresponding to the antigenic structure observed in the SAS1B-7H2.2 Fab complex. **(A)** Ribbon representation of the AlphaFold-predicted structure of the SAS1B protein (Uniprot: Q6HA08) with the C-terminal region colored in *yellow*. **(B)** Overlap of the AlphaFold model α -helix in **(A)** with the α -helix structure of the SAS1B C-terminal region modeled in the SAS1B/7H2.2 Fab crystal structure.

Supplementary Table S1. Nucleotide and amino acid sequence comparison of the variable regions for antibody 7H2.2 and putative germline gene sequences IGHV8-8*01 F (heavy chain V-gene), IGHJ4*01 F (heavy chain J-gene), IGKV3-12*01 F (light chain V-gene) and IGKJ2*01 F (light chain J-gene) for which 7H2.2 exhibits the highest degree of homology.

Heavy chain variable region	
7H2.2 amino acid sequence	Q V T L K E S G P G I L Q P S Q T L S L T C S F S G F S L S T S G M
7H2.2 nucleotide sequence	caggttactctgaaagagtctggccctgggatattgcagccctcccagaccctcagctctgacttggttcttctctgggttttcaactgagcactctcggtagt <-----FR1-----><-----CDR1----->
<u>IGHV8-8*01</u> nucleotide sequence (94.0%)t.....
<u>IGHV8-8*01</u> amino acid sequence	Q V T L K E S G P G I L Q P S Q T L S L T C S F S G F S L S T F G M
7H2.2 amino acid sequence	G V G W I R Q P S G K G L E W L A H I W W D D V K R Y N P A L K S R
7H2.2 nucleotide sequence	ggtgtaggctggattcgtcagccatcaggaagggctggagtgctggcacacatttgggggatgatgtcaagcgtataatccagccctgaagagccga <-----FR2-----><-----CDR2----->
<u>IGHV8-8*01</u> nucleotide sequence (94.0%)T.....at...c.....t.g
<u>IGHV8-8*01</u> amino acid sequence	G V G W I R Q P S G K G L E W L A H I W W D D D K Y Y N P A L K S R
7H2.2 amino acid sequence	L T I S K D T S S S Q V F L K I A S G D T I D T A T Y Y C A R I P T
7H2.2 nucleotide sequence	ctgactatctccaaggatcctccagcagccaggtattcctcaagatcggccagctgggacactatagatactgcccacatactctgtgctogaatacctact <-----FR3-----><-----CDR3----->
<u>IGHV8-8*01</u> nucleotide sequence (94.0%)	..c..a.....aa.a.....a..t.....gc.....*****
<u>IGHV8-8*01</u> amino acid sequence	L T I S K D T S K N Q V F L K I A N V D T A D T A T Y Y C A R I
7H2.2 amino acid sequence	D D Y Y A L D H W G Q G A S V T V S S
7H2.2 nucleotide sequence	gatgattactatgctttggaccactggggtcaaggagcctcagtcaccgtctcctcag <-----CDR3-----><-----FR4----->
<u>IGHJ4*01</u> nucleotide sequence (94.4%)	****.....a.....t.....a.....
<u>IGHJ4*01</u> amino acid sequence	Y Y A L D H W G Q G A S V T V S S
Light chain variable region	
7H2.2 amino acid sequence	D I V L T Q S P A S L A V S L G Q R A T I S C R A S K S V S T S G Y
7H2.2 nucleotide sequence	gacatagtctgacacagctctcctgcttctcttagctgtatctctggggcagaggccaccatctcatgcaaggccagaaaagtgtcagtagacatctcggtagt <-----FR1-----><-----CDR1----->
<u>IGKV3-12*01</u> nucleotide sequence (98.6%)t.....C..
<u>IGKV3-12*01</u> amino acid sequence	D I V L T Q S P A S L A V S L G Q R A T I S C R A S K S V S T S G Y
7H2.2 amino acid sequence	S F M H W Y Q Q K P G Q P P K L L I Y L A S N L E S G V P A R F S G
7H2.2 nucleotide sequence	agttttatgcactggtaccacagaaccagcagaccacccaaactcctcatctcttgcacacatagaatctggggtccctgacaggttcagtgcc <-----FR2-----><-----CDR2----->
<u>IGKV3-12*01</u> nucleotide sequence (98.6%)	...a.....
<u>IGKV3-12*01</u> amino acid sequence	S Y M H W Y Q Q K P G Q P P K L L I Y L A S N L E S G V P A R F S G
7H2.2 amino acid sequence	S G S G T D F T L N I H P V E E E D A A T F Y C Q H S R E L P Y T F
7H2.2 nucleotide sequence	agtgggtctgggacagactcaccctcaacatccatcctgtggaggaggatgctgcaacctttactgtcagcagtagggagcttccgtacacgttc <-----FR3-----><-----CDR3----->
<u>IGKV3-12*01</u> nucleotide sequence (98.6%)a.....*****
<u>IGKV3-12*01</u> amino acid sequence	S G S G T D F T L N I H P V E E E D A A T Y Y C Q H S R E L P
<u>IGKJ2*01</u> nucleotide sequence (100.0%)	*****
<u>IGKJ2*01</u> amino acid sequence.	Y T F
7H2.2 amino acid sequence	G G G T K L E I K
7H2.2 nucleotide sequence	ggaggggggaccaagctggaataaaac <-----FR4----->
<u>IGKJ2*01</u> nucleotide sequence (100.0%)
<u>IGKJ2*01</u> amino acid sequence	G G G T K L E I K

In the germline nucleotide sequence, homology is indicated with a period (.) while a lack of sequence overlap is indicated with an asterisk (*). In the 7H2.2 amino acid sequence, residues that differ from the germline amino acid sequence are indicated in red-bold font.