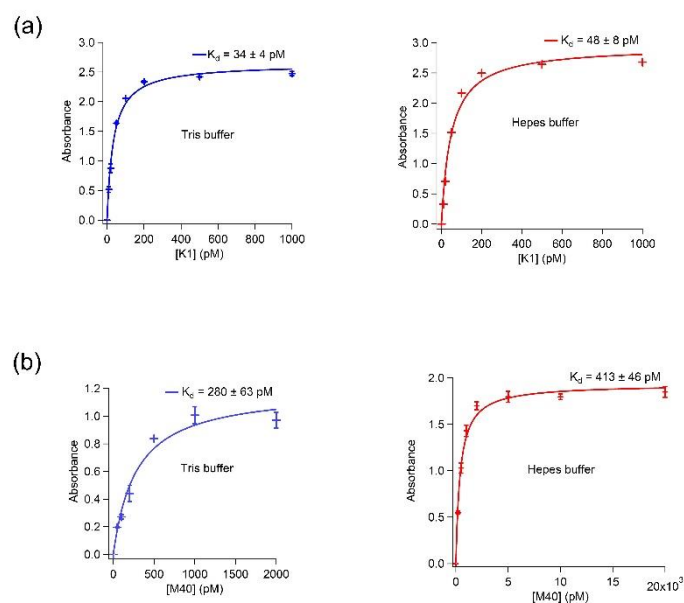


## Supplementary Information

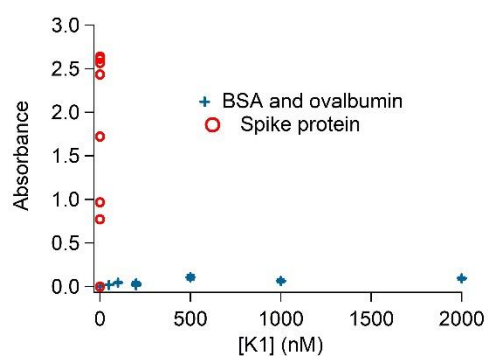
# Development of High Affinity Broadly Reactive Aptamers for Spike Protein of Multiple SARS-CoV-2 Variants

Thao T. Le, Donald J. Benton, Antoni G. Wrobel and Steven J. Gamblin

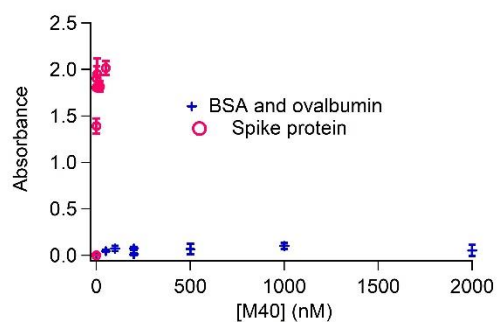


**Figure S11:** (a) Binding of aptamer K1 in Tris buffer and Hepes buffer. (b) Binding of aptamer M40 in Tris buffer and Hepes buffer.

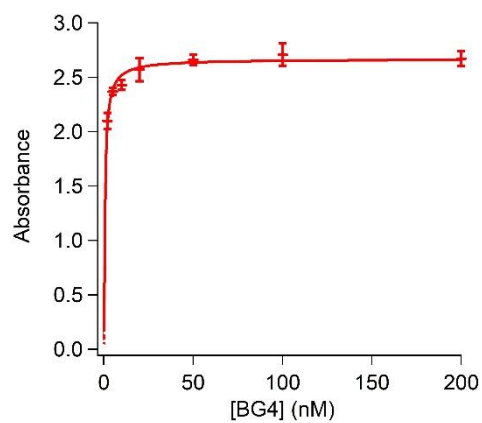
(a)



(b)

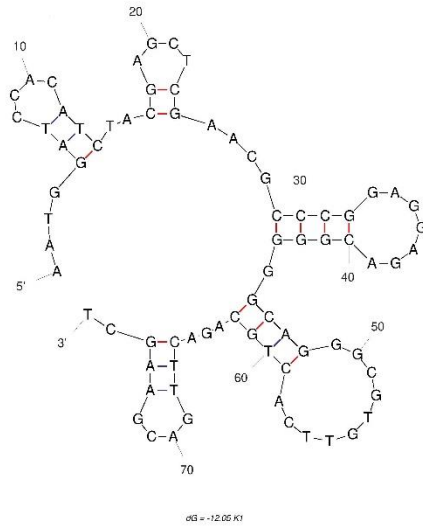


**Figure S12:** Control experiments. (a) Binding of aptamer K1 to BSA and ovalbumin. (b) Binding of aptamer M40 to BSA and ovalbumin.

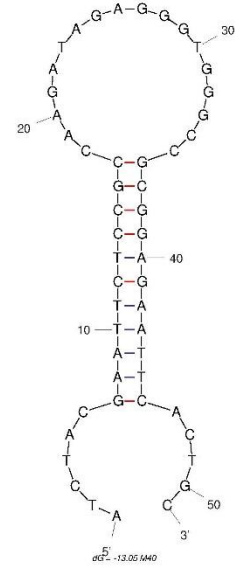


**Figure S13:** Binding of BG4 to biotinylated aptamer K1 (immobilised on plate) by ELISA. Aptamer K1 had a biotin tag at its 3'-end to facilitate its immobilisation on the streptavidin-coated microtiter plate.

(a)



(b)



**Figure S14:** Predicted secondary structures of the aptamers using MFold<sup>1</sup> at in solution having 150 mM Na<sup>+</sup> and 5 mM Mg<sup>2+</sup> and 25°C. (a) Predicted structure of aptamer K1. (b) Predicted structure of aptamer M40.

**Table S11:** A predicted G-quadruplex of aptamer K1 with its overlaps by QSGR-Mapper<sup>2</sup>

Predicted structure Sample	Position	Length	QGRS	G-Score
P1	32	13	<a href="#">GGAGGAGACGGGG</a>	17
P2	32	14	<a href="#">GGAGGAGACGGGGG</a>	18
P3	32	14	<a href="#">GGAGGAGACGGGGG</a>	16
P4	32	18	<a href="#">GGAGGAGACGGGGGCAGG</a>	17
P5	32	18	<a href="#">GGAGGAGACGGGGGCAGG</a>	17
P6	32	18	<a href="#">GGAGGAGACGGGGGCAGG</a>	16
P7	32	18	<a href="#">GGAGGAGACGGGGGCAGG</a>	15
P8	32	18	<a href="#">GGAGGAGACGGGGGCAGG</a>	14
P9	32	18	<a href="#">GAGGAGACGGGGGCAGG</a>	15
P10	32	18	<a href="#">GGAGGAGACGGGGGCAGG</a>	13
P11	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	16
P12	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	17
P13	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	16
P14	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	15
P15	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	14
P16	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	15
P17	32	19	<a href="#">GGAGGAGACGGGGGCAGGG</a>	13
P18	35	15	<a href="#">GGAGACGGGGGCAGG</a>	17
P19	35	15	<a href="#">GGAGACGGGGGCAGG</a>	18
P20	35	15	<a href="#">GGAGACGGGGGCAGG</a>	16
P21	35	16	<a href="#">GGAGACGGGGGCAGGG</a>	17
P22	35	16	<a href="#">GGAGACGGGGGCAGGG</a>	18
P23	35	16	<a href="#">GAGACGGGGGCAGGG</a>	16

**Table S12:** Sequences of the truncated oligos

<b>Oligo14</b>	CCACATCTACGA GCTCGAACGCCCGGAGGAGACGGGGGCAGGGCGTG TTCACTGCAGACTTGA
<b>Oligo30</b>	ACGA GCTCGAACGCCCGGAGGAGACGGGGGCAGGGCGTG TTCACTGC

## References

- 1 M. Zuker, Nucleic Acids Research, 2003. **31**(13): p. 3406-3415.
- 2 O. Kikin, L. D'Antonio, and P. S. Bagga, Nucleic Acids Research, 2006. **34**(suppl\_2): p. W676-W682.