

## **Supplementary material**

### **Integrating Single Domain Antibodies into Field-Deployable Rapid Assays**

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Figure S1. Evaluation of different ACVE capture formats

Figure S2. Evaluation of two ACVE-based constructs on AuNPs

**Table S1.** Tail names and amino acid sequences

Tail name	Tail sequence
Hop tail	GAGGSGGAPASNRCSQGSCWN
Spy tag (ST)	AHIVMVDAYKPTK
Gold binding peptide (gbp) 1	MHGKTQATSGTIQS
Gbp2	LKAHLPPSRLPS
Synthetic zipper (synzip) 5	NTVKELKNYIQELEERNAELKNLKEHLKFAKAELEFELAAHKFE
synzip 6	QKVAQLKNRVAYKLKENAKLENIVARLENDNANLEKDIANLEKDIANLERDVAR
synzip 17	NEKEELKSKKAELRNRIEQLKQKREQLKQKIANLRKEIEAYK
synzip 18	SIAATLENDLARLENENARLEKDIANLERDLAKLEREEAYF
E34	ITIRAAFLEKENTALRTEIAELEKEVGRSENIIVSKYETRYGPL
Rhizavidin (RZ)	FDASNFKDFSSIASASSWQNSQSGSTMIIQVDSFGNVSGQYVNRAQGTGCQN SPYPLTGRVNGTFIAFVGWNNSTENCNSATGWTGYAQVNGNNTIEVTSWNL AYEGGSGPAIEQGQDTFQYVPTTENKSLLKD

**Table S2.** Sequences of sdAb used in the VFA

SdAb name	SdAb sequence
ACVE	DVQLVESGGGLVQPGGSLRLTCAASGLIFGSYAMGWFRQAPGKAREFVAAISWSGGDTYADSVK GRFTISRDNANTVYLQMNLSLEPEDTAVYSAAVGSKYISKDAKDYGWGGGTQVTVSS
A3H2	EVQLVESGGGLVQAGDSLRLSCTASGRIFSRVAMGWFRQAPGKAREFVAAISAAPGTAYYAFYADS VRGRFIISADSAKNTVYLQMNLSLPEDTAVYYCAADLKMQVAAYMNQRSVDYWGGGTQVTVSS
D12f	EVQLQASGGGLVQDGGSLRLSCAVAGRPLSDYGVGWFRQASGKAREFVAVISGSGIVTDYANSVK GRFTISRDVAKNVVHLQMNLSLPEDTAVYYCAALTNPVYAASRNSNDYGWGGGTQVTVSS
F6H2Y	QVQLVESGGGLVQAGGSLRLSCATSGGTFSDYGMGWFRQAPGKAREFVAAIRRNGNGNGIEYA DSVKGRFTISRDNANTVYLQMNLSLPEDTAVYYCAASISGYAYNTIERYNYWGGGTQVTVSS
E2	EVQLQASGGGLVQAGGSLRLSCAASGRTDSTQHMAWFRQAPGKAREFVTAIQWRGGGTSYTDS VKGRFTISRDNANTVYLEMNSLPEDTAVYYCATNTRWTYFSPTVPDRYDYWGGGTQVTVSS
C2	EVQLQASGGGLVQAGGSLRLSCAASGFTSSYAMMWVRQAPGKLEWVSAINGGGGTSYADS VKGRFTISRDNANTLYLQMNLSLPEDTAVYYCAKYQAAVHQEKEDYWGGGTQVTVSS
B6	EVQLQASGGGLVQAGDSLRLSCVAVSGRTISTFAMGWFRQAPGKAREFVATINWSGSSARYADPV EGRFTISRDDAKNTVYLEMSSLKPGDSAVYYCASGRYLGGITSYSQGFAPWGGGTQVTVSS

**Figure S1.**



Evaluation of different ACVE capture formats. Upper left spot = ACVE; upper right spot = ACVE-ACVE; lower left spot = SCVE-ST; lower right spot = ACVE-synthetic zipper 17

**Figure S2.**



Evaluation of two ACVE-based constructs on AuNPs. Left is ACVE-hop and right is the trivalent construct ACVE-ACVE-ACVE. Miriad test cartridges were spotted with a monoclonal anti-SEB (mAb 2F2) on the top, and SEBv on the bottom. Next 100  $\mu$ L of SEBv (10  $\mu$ g/mL) was applied to the cartridge, followed by a wash and the AuNPs. Cartridges were washed again and photographed.