Supplementary information

Rapid and Convenient Single-Chain Variable Fragment-Employed Electrochemical C-Reactive Protein Detection System

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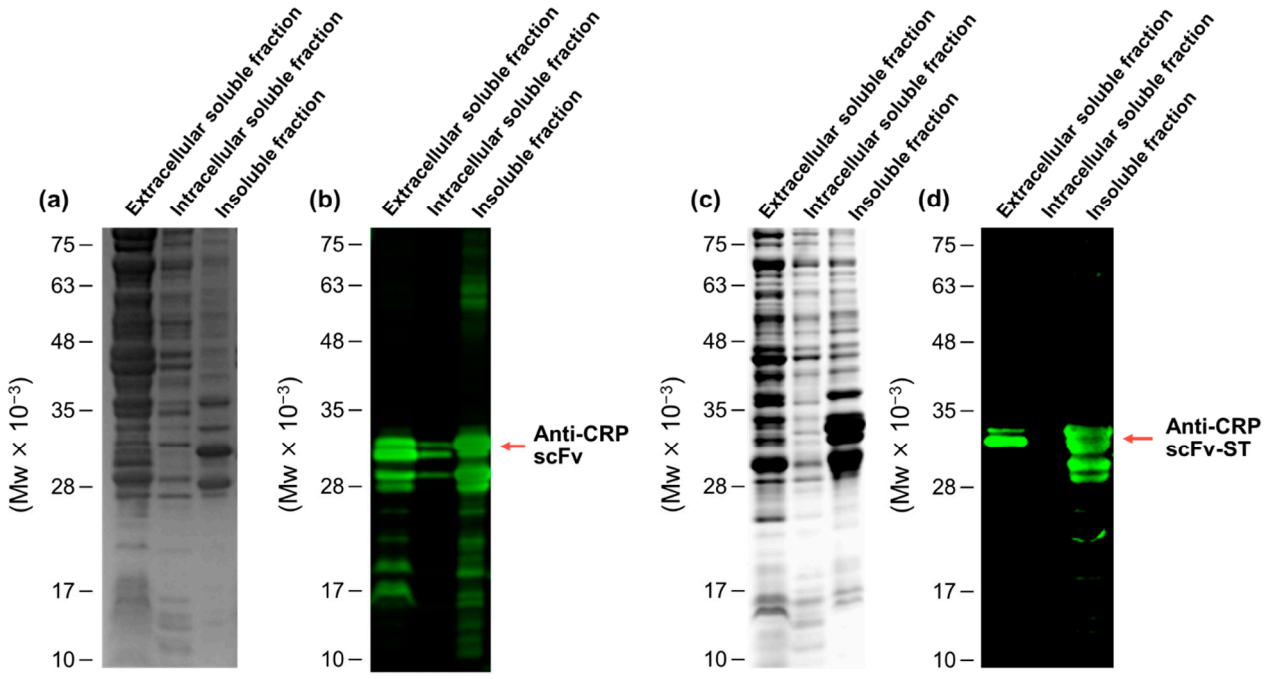

Figure S1 SDS-PAGE and Western blotting analysis for confirmation of expression of (a, b) anti-CRP scFv and (c, d) anti-CRP scFv-ST).

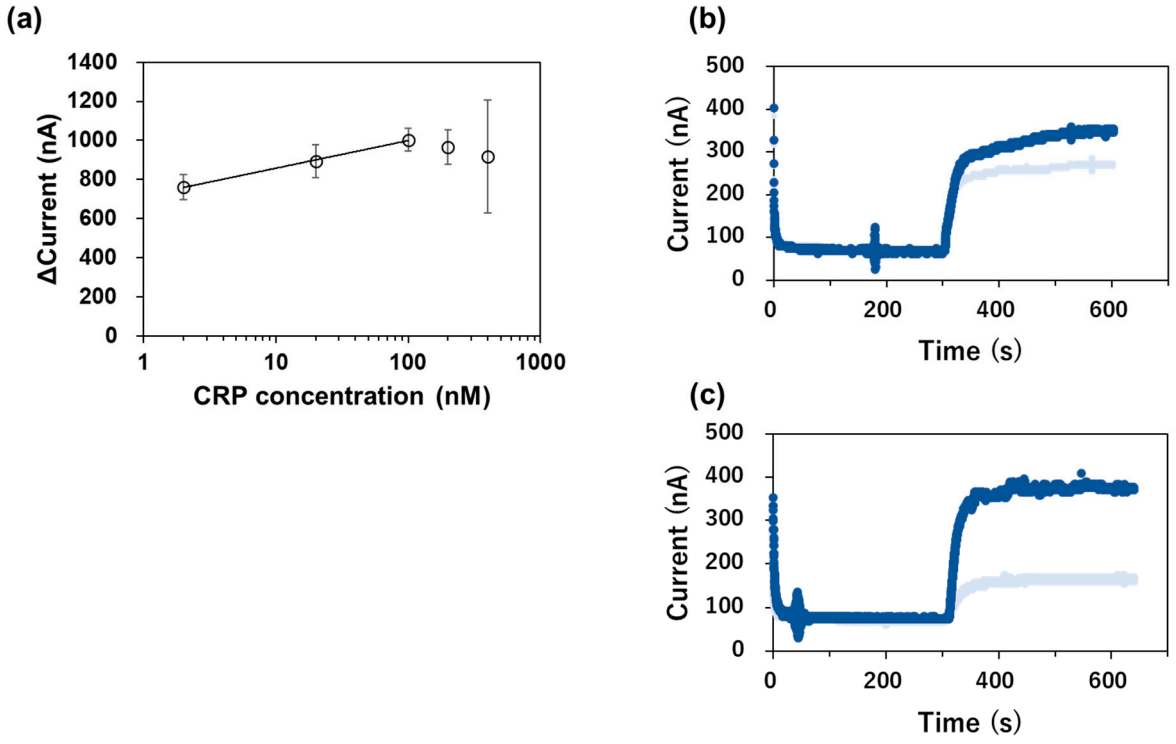

Figure S2 (a) A calibration curve of the electrochemical detection of CRP detection using our previous protocol (n = 3). Electrochemical CRP detection (b) without and (c) with washing after incubation with bivalent AEC, CRP, or scFv-immobilized magnetic beads (n = 1).

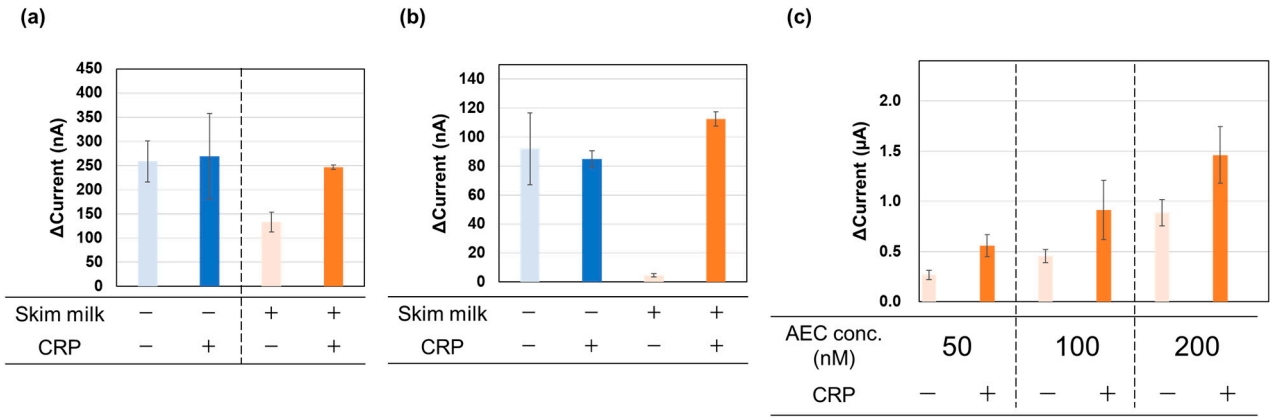

Figure S3 Investigation of the conditions for electrochemical detection of CRP. (a) The effect of skim milk during the incubation of AEC and CRP with scFv-captured magnetic beads on the prevention of non-specific binding of the bivalent AEC. (b) The effect of a washing procedure and addition of skim milk during the incubation of AEC and CRP with scFv-captured magnetic beads. (c) The effect of the concentration of the bivalent AEC. All data are presented as mean ± S.D. (n = 3).

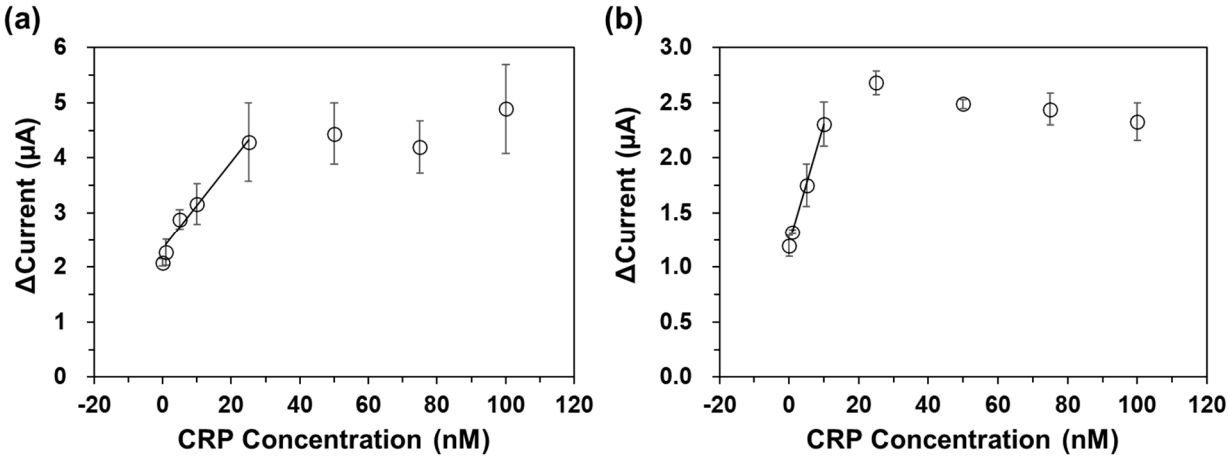

Figure S4 Calibration curves of detection (a) in buffer and (b) in serum. All data are presented as mean ± S.D. (*n* = 3).

Table S1 Amino acid sequences of the proteins used in this study.

Protein	Sequence
Anti-CRP scFv	MKYLLPTAAAGLLLLAAQPAMAHMASQVQLQQSGAELVKPGASVKLSCTASGFNIKDYYMHWVKQRTEQGLEWIGRIDPEDGETKYAPKFQGKATITADTSS NTAYLQLSSLTSEDTAVYYCARGYYGSEAMDYWGQGTSLTVSSGGGGSGGGGSGGGGSTGSIVMTQSHKFMSTSVGDRVSITCKASQDVNTAVAWYQQK PGQSPKLLIYWASTRHTGVPDRFTGSGFGTDYTLTISSVQAEDLALYYCQQHYSTPWTFGGGTKLEIKRADAAPKLAAAAEQKLISEEDLNLGGGMRGSHHHHH HH
Anti-CRP scFv-ST	MKYLLPTAAAGLLLLAAQPAMAHMASQVQLQQSGAELVKPGASVKLSCTASGFNIKDYYMHWVKQRTEQGLEWIGRIDPEDGETKYAPKFQGKATITADTSS NTAYLQLSSLTSEDTAVYYCARGYYGSEAMDYWGQGTSLTVSSGGGGSGGGGSGGGGSTGSIVMTQSHKFMSTSVGDRVSITCKASQDVNTAVAWYQQK PGQSPKLLIYWASTRHTGVPDRFTGSGFGTDYTLTISSVQAEDLALYYCQQHYSTPWTFGGGTKLEIKRADAAPKLAAAGSGAHIVMVDAYKPTKAEQKLISEE DLNLGGGMRGSHHHHHH
SC	MGAMVDTLSGLSSEQGQSGDMTIEEDSATHIKFSKRDEDGKELAGATMELRDSSGKTISTWISDGQVKDFYLYPGKYTFVETAAPDGYEVATAITFTVNEQGQ VTVNGKATKGDAHIHHHHHH
GDH-SC	MNTTTYDYIVVGGGTSGLVVANRLSENPDVSVLLLEAGASVFNNPDVTNANGYGLAFGSAIDWQYQSINQSYAGGKQQVLRAGKALGGTSTINGMAYTRAED VQIDVWQKLGNEGWTWKDLLPYYLKSENLTAPTSSQVAAGAAYNPACNGKEGPLKVGWSGSLASGNLSVALNRTFQAAGVPWVEDVNCGKMRGFNIYPST LDVDLNVREDAARAYYFPYDDRKNLHLLENTTANRLFWKNGSAEEAIADGVEITSADGKVTRVHAKKEVIISAGALRSPLILELSGVGNPTILKKNNITPRVDLPT VGENLQDQFNNGMAGEGYGVLAGASTVTYPSISDVFGNETDSIVASLRSQLSDYAAATVKVSNGHMKQEDLERLYQLQFDLIVKDKVPIAEILFHPGGGNAVS SEFWGLLPFARGNIHISSNDPTAPAAINPNYFMFEWDGKSQAGIAKYIRKILRSAPLNKLIAKETKPGLSEIPATAADEKWVEWLKANYRSNFHPVGTAAMMPR SIGGVVDNRLRVYGTSNVRVVDASVLPFQVCGHLVSTLYAVAERASDLIKEDAKSAGSGGAMVDTLSGLSSEQGQSGDMTIEEDSATHIKFSKRDEDGKELAG ATMELRDSSGKTISTWISDGQVKDFYLYPGKYTFVETAAPDGYEVATAITFTVNEQGQVTVNGKATKGDAHILEHHHHHH
SC-GDH-SC	MGSSHHHHHHSSGLVPRGSGAMVDTLSGLSSEQGQSGDMTIEEDSATHIKFSKRDEDGKELAGATMELRDSSGKTISTWISDGQVKDFYLYPGKYTFVETAA PDGYEVATAITFTVNEQGQVTVNGKATKGDAHIGSGHMNTTTYDYIVVGGGTSGLVVANRLSENPDVSVLLLEAGASVFNNPDVTNANGYGLAFGSAIDWQY QSINQSYAGGKQQVLRAGKALGGTSTINGMAYTRAEDVQIDVWQKLGNEGWTWKDLLPYYLKSENLTAPTSSQVAAGAAYNPACNGKEGPLKVGWSGSLA SGNLSVALNRTFQAAGVPWVEDVNCGKMRGFNIYPSTLDVDLNVREDAARAYYFPYDDRKNLHLLENTTANRLFWKNGSAEEAIADGVEITSADGKVTRVHA KKEVIISAGALRSPLILELSGVGNPTILKKNNITPRVDLPTVGENLQDQFNNGMAGEGYGVLAGASTVTYPSISDVFGNETDSIVASLRSQLSDYAAATVKVSNG HMKQEDLERLYQLQFDLIVKDKVPIAEILFHPGGGNAVSSEFWGLLPFARGNIHISSNDPTAPAAINPNYFMFEWDGKSQAGIAKYIRKILRSAPLNKLIAKETKP GLSEIPATAADEKWVEWLKANYRSNFHPVGTAAMMPRSIGGVVDNRLRVYGTSNVRVVDASVLPFQVCGHLVSTLYAVAERASDLIKEDAKSAGSGGAMVD TLSGLSSEQGQSGDMTIEEDSATHIKFSKRDEDGKELAGATMELRDSSGKTISTWISDGQVKDFYLYPGKYTFVETAAPDGYEVATAITFTVNEQGQVTVNGK ATKGDAHILEHHHHHH