

SUPPLEMENTARY INFORMATION

Functional properties of flagellin as a stimulator of innate immunity

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Supplementary Tables

Supplementary Table S1. The sequences of different flagellin variants.

| Variants | Protein sequence | DNA encoding sequence |
|-----------|---|---|
| Wild-type | MAQVINTNSLSLLTQNNLNKSQSAL GTAIERLSSGLRINSKDDAAGQAI ANRFTANIKGLTQASRNANDGISIA QTTEGALNEINNNLQRVRELAVQSA NSTNSQSDLLDSIQAEITQRLNEIDR VSGQTQFNGVKVLAQDNTLTIQVGA NDGETIDIDLKQINSQTLGLDNLNV QKRYKVSdTAAATVTGYADTTIALDN STFKASATGLGGTDQKIDGDLKFD TTGKYAKVTVTGGTGKDGYYEVS DKTNGEVTLAGGATSPLTGGLPATA TEVKNVQVANADLTEAKAALTAAG VTGTASVVKMSYTDNNGKTIIDGGLA VKVGDYYSATQNKDGSISINTTKY TADDGTSKLTALNKGADGKTEVVS IGGKTYAASKAEGHNFKAQPDLEA AATTTENPLQKIDAALAQVDTLRSD LGAQVNRFNSAITNLGNTVNNLTS RSRIEDSDYATEVSNMSRAQILQQA GTSVLAQANQVPPQNVLSLLRW | ATGGCACAAGTGATTAATACAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAACAAATCCCAGTCCGCTC TGGGCACCCTATCGAGCGTCTGTCTCCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGTACAGGC GATTGCTAACCGTTTTACCGCCAACAATAAAGGTTGACTACGGCTTCCCCTAACGCTAACGACGGTATCTCC ATTGGCAGACCCACTGAAGGCGCGTGAACGAAATCAACAACAACCTGCAGCGTGTGCGTGAACCTGGCGGTT CAGTCTGCTAACAGCACCCTCCAGTCTGACTCGACTCCATCCAGGCTGAAATCACCCAGCGCTGAACG AAATCGACCGTGTATCCGGCCAGACTCAGTCAACGGCGTGAAGTCTGGCGCAGGACAACCCCTGACCA TCCAGGTTGGTCCACAGCGGTTGAAACTATCGATATCGATCTGAAGCAGATCAACTCTCAGACCCCTGGGTCT GGATACCGCTGAATGTGCAACAAAAATAAAGGTCAGCGATACGGCTGCAACTGTTACAGGATATGCCGATAC TACGATTGCTTTAGACAATAGTACTTTAAAGCCTCGGCTACTGGTCTTGGTGGTACTACAGAAAATTGATG GCGATTTAAAATTTGATGATACGACTGGAAAAATTACGCCAAAGTTACCGTTACGGGGGGAACCTGGTAAAG ATGGCTATTGAAAGTTCCGTTGATAAGACGAAACGGTGAAGTGAAGTACTTGTCTGGCGGTGCGACTTCCCCTG TACAGGTGGACTACTCGCAGCAGCAACTGAGGATGTGAAAAATGTACAAGTTGCAAAATGCTGATTTGACAGA GGCTAAAGCCGATTGACAGCAGCAGGTGTTACCAGCAGCAGCATCTGTTGTTAAGATGCTTATACAGTAAAT AACGGTAAAACTATTGATGGTGGTTAGCAGTTAAGGTAGGCGATGATTACTATTCTGCAACTCAAAAATAAAG ATGGTCCATAAGTATTAATACTACGAAATACACTGCAGATGACGGTACACTTAAACCTGCACTAAACAACT GGGTGGCGCAGACGGCAAAACCGAAGTTGTTCTATTGGTGGTAAAACITACCGTGAAGTAAAGCCGAAGG TCACAACCTTAAAGCACAGCTGATCTGGCGGAAGCGGCTGTACAACCACCGAAAACCCGCTGCAGAAAAT TGATGCTGCTTGGCACAGGTTGACAGCTTACGTTCTGACCTGGGTGGGTACAGAACTGGTTCACAACTGACTA TTACCAACTGGGCAACACCGTAAACAACTGACTTCTGCCGTAGCCGTATCGAAGATTCCGACTACCGGAC CGAAGTTTCCAACTGCTCGCGCGAGATTCTGACGACGGCCGTTACCTCGTCTGGCGCAGGCGAACCCAG GTTCCGCAAAACGCTCTCTTTACTGCGT |
| | FEK | IGGAGCCATCCGCAGTTGAAAAATAA |
| | (HPG) | ATGGCACAAGTGATTAATACAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAACAAATCCCAGTCCGCTC TGGGCACCCTATCGAGCGTCTGTCTCCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGTACAGGC GATTGCTAACCGTTTTACCGCCAACAATAAAGGTTGACTACGGCTTCCCCTAACGCTAACGACGGTATCTCC ATTGGCAGACCCACTGAAGGCGCGTGAACGAAATCAACAACAACCTGCAGCGTGTGCGTGAACCTGGCGGTT CAGTCTGCTAACAGCACCCTCCAGTCTGACTCGACTCCATCCAGGCTGAAATCACCCAGCGCTGAACG AAATCGACCGTGTATCCGGCCAGACTCAGTCAACGGCGTGAAGTCTGGCGCAGGACAACCCCTGACCA TCCAGGTTGGTCCACAGCGGTTGAAACTATCGATATCGATCTGAAGCAGATCAACTCTCAGACCCCTGGGTCT GGATACCGCTGAATGTGCAACAAAAATAAAGGTCAGCGATACGGCTGCAACTGTTACAGGATATGCCGATAC TACGATTGCTTTAGACAATAGTACTTTAAAGCCTCGGCTACTGGTCTTGGTGGTACTGACCAAAAATTGATG GCGATTTAAAATTTGATGATACGACTGGAAAAATTACGCCAAAGTTACCGTTACGGGGGGAACCTGGTAAAG ATGGCTATTGAAAGTTCCGTTGATAAGACGAAACGGTGAAGTGAAGTACTTGTCTGGCGGTGCGACTTCCCCTG TACAGGTGGACTACTCGCAGCAGCAACTGAGGATGTGAAAAATGTACAAGTTGCAAAATGCTGATTTGACAGA GGCTAAAGCCGATTGACAGCAGCAGGTGTTACCAGCAGCAGCATCTGTTGTTAAGATCTTATACTGATAAT AACGGTAAAACTATTGATGGTGGTTAGCAGTTAAGGTAGGCGATGATTACTATTCTGCAACTCAAAAATAAAG ATGGTCCATAAGTATTAATACTACGAAATACACTGCAGATGACGGTACACTCAAAAATGCTACTAAACAACT GGGTGGCGCAGACGGCAAAACCGAAGTTGTTCTATTGGTGGTAAAACITACCGTGAAGTAAAGCCGAAGG TCACAACCTTAAAGCACAGCTGATCTGGCGGAAGCGGCTGTACAACCACCGAAAACCCGCTGCAGAAAAT TGATGCTGCTTGGCACAGGTTGACAGCTTACGTTCTGACCTGGGTGGGTACAGAACTGGTTCACAACTGACTA TTACCAACTGGGCAACACCGTAAACAACTGACTTCTGCCGTAGCCGTATCGAAGATTCCGACTACCGGAC CGAAGTTTCCAACTGCTCGCGCGAGATTCTGACGACGGCCGTTACCTCGTCTGGCGCAGGCGAACCCAG GTTCCGCAAAACGCTCTCTTTACTGCGT |
| III | KFDDTTGKYAKVTVTGGTGKDGYY EVSVDKTNGEVTLAGGATSPLTGGL PATATEDVKNVQVANADLTEAKAAL TAAGVTGTASVVKISYTDNNGKTI DGLAVKVGDDYYSATQNKDGSISIN TKYTADDGTSKLTALNKGADGKT EVVYIGGKTYAASKAEGHNFKAQPD LAEAAAATTTENPLQKIDAALAQVDT LRSDLGAQVNRFNSAITNLGNTVNN LTSARSIEDSDYATEVSNISRAQI LQOAGTSVLAQANQVPPQNVLSLLRW | ATGGCACAAGTGATTAATACAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAACAAATCCCAGTCCGCTC TGGGCACCCTATCGAGCGTCTGTCTCCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGTACAGGC GATTGCTAACCGTTTTACCGCCAACAATAAAGGTTGACTACGGCTTCCCCTAACGCTAACGACGGTATCTCC ATTGGCAGACCCACTGAAGGCGCGTGAACGAAATCAACAACAACCTGCAGCGTGTGCGTGAACCTGGCGGTT CAGTCTGCTAACAGCACCCTCCAGTCTGACTCGACTCCATCCAGGCTGAAATCACCCAGCGCTGAACG AAATCGACCGTGTATCCGGCCAGACTCAGTCAACGGCGTGAAGTCTGGCGCAGGACAACCCCTGACCA TCCAGGTTGGTCCACAGCGGTTGAAACTATCGATATCGATCTGAAGCAGATCAACTCTCAGACCCCTGGGTCT GGATACCGCTGAATGTGCAACAAAAATAAAGGTCAGCGATACGGCTGCAACTGTTACAGGATATGCCGATAC TACGATTGCTTTAGACAATAGTACTTTAAAGCCTCGGCTACTGGTCTTGGTGGTACTGACCAAAAATTGATG GCGATTTAAAATTTGATGATACGACTGGAAAAATTACGCCAAAGTTACCGTTACGGGGGGAACCTGGTAAAG ATGGCTATTGAAAGTTCCGTTGATAAGACGAAACGGTGAAGTGAAGTACTTGTCTGGCGGTGCGACTTCCCCTG TACAGGTGGACTACTCGCAGCAGCAACTGAGGATGTGAAAAATGTACAAGTTGCAAAATGCTGATTTGACAGA GGCTAAAGCCGATTGACAGCAGCAGGTGTTACCAGCAGCAGCATCTGTTGTTAAGATCTTATACTGATAAT AACGGTAAAACTATTGATGGTGGTTAGCAGTTAAGGTAGGCGATGATTACTATTCTGCAACTCAAAAATAAAG ATGGTCCATAAGTATTAATACTACGAAATACACTGCAGATGACGGTACACTCAAAAATGCTACTAAACAACT GGGTGGCGCAGACGGCAAAACCGAAGTTGTTCTATTGGTGGTAAAACITACCGTGAAGTAAAGCCGAAGG TCACAACCTTAAAGCACAGCTGATCTGGCGGAAGCGGCTGTACAACCACCGAAAACCCGCTGCAGAAAAT TGATGCTGCTTGGCACAGGTTGACAGCTTACGTTCTGACCTGGGTGGGTACAGAACTGGTTCACAACTGACTA TTACCAACTGGGCAACACCGTAAACAACTGACTTCTGCCGTAGCCGTATCGAAGATTCCGACTACCGGAC CGAAGTTTCCAACTGCTCGCGCGAGATTCTGACGACGGCCGTTACCTCGTCTGGCGCAGGCGAACCCAG GTTCCGCAAAACGCTCTCTTTACTGCGT |
| | SHPOFEK | IGGAGCCATCCGCAGTTGAAAAATAA |
| | (HPG) | ATGATACAAGTGATTAATACAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAACAAATCCCAGTCCGCTC TGGGCACCCTATCGAGCGTCTGTCTCCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGTACAGGC GATTGCTAACCGTTTTACCGCCAACAATAAAGGTTGACTACGGCTTCCCCTAACGCTAACGACGGTATCTCC ATTGGCAGACCCACTGAAGGCGCGTGAACGAAATCAACAACAACCTGCAGCGTGTGCGTGAACCTGGCGGTT CAGTCTGCTAACAGCACCCTCCAGTCTGACTCGACTCCATCCAGGCTGAAATCACCCAGCGCTGAACG AAATCGACCGTGTATCCGGCCAGACTCAGTCAACGGCGTGAAGTCTGGCGCAGGACAACCCCTGACCA TCCAGGTTGGTCCACAGCGGTTGAAACTATCGATATCGATCTGAAGCAGATCAACTCTCAGACCCCTGGGTCT GGATACCGCTGAATGTGCAACAAAAATAAAGGTCAGCGATACGGCTGCAACTGTTACAGGATATGCCGATAC TACGATTGCTTTAGACAATAGTACTTTAAAGCCTCGGCTACTGGTCTTGGTGGTACTGACCAAAAATTGATG GCGATTTAAAATTTGATGATACGACTGGAAAAATTACGCCAAAGTTACCGTTACGGGGGGAACCTGGTAAAG ATGGCTATTGAAAGTTCCGTTGATAAGACGAAACGGTGAAGTGAAGTACTTGTCTGGCGGTGCGACTTCCCCTG TACAGGTGGACTACTCGCAGCAGCAACTGAGGATGTGAAAAATGTACAAGTTGCAAAATGCTGATTTGACAGA GGCTAAAGCCGATTGACAGCAGCAGGTGTTACCAGCAGCAGCATCTGTTGTTAAGATCTTATACTGATAAT AACGGTAAAACTATTGATGGTGGTTAGCAGTTAAGGTAGGCGATGATTACTATTCTGCAACTCAAAAATAAAG ATGGTCCATAAGTATTAATACTACGAAATACACTGCAGATGACGGTACACTCAAAAATGCTACTAAACAACT GGGTGGCGCAGACGGCAAAACCGAAGTTGTTCTATTGGTGGTAAAACITACCGTGAAGTAAAGCCGAAGG TCACAACCTTAAAGCACAGCTGATCTGGCGGAAGCGGCTGTACAACCACCGAAAACCCGCTGCAGAAAAT TGATGCTGCTTGGCACAGGTTGACAGCTTACGTTCTGACCTGGGTGGGTACAGAACTGGTTCACAACTGACTA TTACCAACTGGGCAACACCGTAAACAACTGACTTCTGCCGTAGCCGTATCGAAGATTCCGACTACCGGAC CGAAGTTTCCAACTGCTCGCGCGAGATTCTGACGACGGCCGTTACCTCGTCTGGCGCAGGCGAACCCAG GTTCCGCAAAACGCTCTCTTTACTGCGT |
| IIII | KFDDTTGKYAKVTVTGGTGKDGYY EVSVDKTNGEVTLAGGATSPLTGGL PATATEDVKNVQVANADLTEAKAAL TAAGVTGTASVVKISYTDNNGKTI DGLAVKVGDDYYSATQNKDGSISIN TKYTADDGTSKLTALNKGADGKT EVVYIGGKTYAASKAEGHNFKAQPD LAEAAAATTTENPLQKIDAALAQVDT LRSDLGAQVNRFNSAITNLGNTVNN LTSARSIEDSDYATEVSNISRAQI LQOAGTSVLAQANQVPPQNVLSLLRW | ATGATACAAGTGATTAATACAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAACAAATCCCAGTCCGCTC TGGGCACCCTATCGAGCGTCTGTCTCCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGTACAGGC GATTGCTAACCGTTTTACCGCCAACAATAAAGGTTGACTACGGCTTCCCCTAACGCTAACGACGGTATCTCC ATTGGCAGACCCACTGAAGGCGCGTGAACGAAATCAACAACAACCTGCAGCGTGTGCGTGAACCTGGCGGTT CAGTCTGCTAACAGCACCCTCCAGTCTGACTCGACTCCATCCAGGCTGAAATCACCCAGCGCTGAACG AAATCGACCGTGTATCCGGCCAGACTCAGTCAACGGCGTGAAGTCTGGCGCAGGACAACCCCTGACCA TCCAGGTTGGTCCACAGCGGTTGAAACTATCGATATCGATCTGAAGCAGATCAACTCTCAGACCCCTGGGTCT GGATACCGCTGAATGTGCAACAAAAATAAAGGTCAGCGATACGGCTGCAACTGTTACAGGATATGCCGATAC TACGATTGCTTTAGACAATAGTACTTTAAAGCCTCGGCTACTGGTCTTGGTGGTACTGACCAAAAATTGATG GCGATTTAAAATTTGATGATACGACTGGAAAAATTACGCCAAAGTTACCGTTACGGGGGGAACCTGGTAAAG ATGGCTATTGAAAGTTCCGTTGATAAGACGAAACGGTGAAGTGAAGTACTTGTCTGGCGGTGCGACTTCCCCTG TACAGGTGGACTACTCGCAGCAGCAACTGAGGATGTGAAAAATGTACAAGTTGCAAAATGCTGATTTGACAGA GGCTAAAGCCGATTGACAGCAGCAGGTGTTACCAGCAGCAGCATCTGTTGTTAAGATCTTATACTGATAAT AACGGTAAAACTATTGATGGTGGTTAGCAGTTAAGGTAGGCGATGATTACTATTCTGCAACTCAAAAATAAAG ATGGTCCATAAGTATTAATACTACGAAATACACTGCAGATGACGGTACACTCAAAAATGCTACTAAACAACT GGGTGGCGCAGACGGCAAAACCGAAGTTGTTCTATTGGTGGTAAAACITACCGTGAAGTAAAGCCGAAGG TCACAACCTTAAAGCACAGCTGATCTGGCGGAAGCGGCTGTACAACCACCGAAAACCCGCTGCAGAAAAT TGATGCTGCTTGGCACAGGTTGACAGCTTACGTTCTGACCTGGGTGGGTACAGAACTGGTTCACAACTGACTA TTACCAACTGGGCAACACCGTAAACAACTGACTTCTGCCGTAGCCGTATCGAAGATTCCGACTACCGGAC CGAAGTTTCCAACTGCTCGCGCGAGATTCTGACGACGGCCGTTACCTCGTCTGGCGCAGGCGAACCCAG GTTCCGCAAAACGCTCTCTTTACTGCGT |
| | SHPOFEK | IGGAGCCATCCGCAGTTGAAAAATAA |
| | (HPG) | ATGATACAAGTGATTAATACAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAACAAATCCCAGTCCGCTC TGGGCACCCTATCGAGCGTCTGTCTCCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGTACAGGC GATTGCTAACCGTTTTACCGCCAACAATAAAGGTTGACTACGGCTTCCCCTAACGCTAACGACGGTATCTCC ATTGGCAGACCCACTGAAGGCGCGTGAACGAAATCAACAACAACCTGCAGCGTGTGCGTGAACCTGGCGGTT CAGTCTGCTAACAGCACCCTCCAGTCTGACTCGACTCCATCCAGGCTGAAATCACCCAGCGCTGAACG AAATCGACCGTGTATCCGGCCAGACTCAGTCAACGGCGTGAAGTCTGGCGCAGGACAACCCCTGACCA TCCAGGTTGGTCCACAGCGGTTGAAACTATCGATATCGATCTGAAGCAGATCAACTCTCAGACCCCTGGGTCT GGATACCGCTGAATGTGCAACAAAAATAAAGGTCAGCGATACGGCTGCAACTGTTACAGGATATGCCGATAC TACGATTGCTTTAGACAATAGTACTTTAAAGCCTCGGCTACTGGTCTTGGTGGTACTGACCAAAAATTGATG GCGATTTAAAATTTGATGATACGACTGGAAAAATTACGCCAAAGTTACCGTTACGGGGGGAACCTGGTAAAG ATGGCTATTGAAAGTTCCGTTGATAAGACGAAACGGTGAAGTGAAGTACTTGTCTGGCGGTGCGACTTCCCCTG TACAGGTGGACTACTCGCAGCAGCAACTGAGGATGTGAAAAATGTACAAGTTGCAAAATGCTGATTTGACAGA GGCTAAAGCCGATTGACAGCAGCAGGTGTTACCAGCAGCAGCATCTGTTGTTAAGATCTTATACTGATAAT AACGGTAAAACTATTGATGGTGGTTAGCAGTTAAGGTAGGCGATGATTACTATTCTGCAACTCAAAAATAAAG ATGGTCCATAAGTATTAATACTACGAAATACACTGCAGATGACGGTACACTCAAAAATGCTACTAAACAACT GGGTGGCGCAGACGGCAAAACCGAAGTTGTTCTATTGGTGGTAAAACITACCGTGAAGTAAAGCCGAAGG TCACAACCTTAAAGCACAGCTGATCTGGCGGAAGCGGCTGTACAACCACCGAAAACCCGCTGCAGAAAAT TGATGCTGCTTGGCACAGGTTGACAGCTTACGTTCTGACCTGGGTGGGTACAGAACTGGTTCACAACTGACTA TTACCAACTGGGCAACACCGTAAACAACTGACTTCTGCCGTAGCCGTATCGAAGATTCCGACTACCGGAC CGAAGTTTCCAACTGCTCGCGCGAGATTCTGACGACGGCCGTTACCTCGTCTGGCGCAGGCGAACCCAG GTTCCGCAAAACGCTCTCTTTACTGCGT |

(HPG) A Q V I N T N S L S L L T Q N N L N K S
 Q S A L G T A I E R L S S G L R I N S A K D D A A
 G Q A I A N R F T A N I K G L T Q A S R N A N D G
 I S I A Q T T E G A L N E I N N N L Q R V R E L A
 V Q S A N S T N S Q S D L D S I Q A E I T Q R L N
 E I D R V S G Q T Q F N G V K V L A Q D N T L T I
 Q V G A N D G E T I D I D L K I N S Q T L G L D
 T L N V Q Q K Y K V S D T A A T V T G Y A D T T I
 A L D N S T F K A S A T G L G G T D Q K I D G D L
 K F D D T T G K Y Y A K V T V T G **(HPG)** T G K
 D G Y Y E V S V D K T N G E V T L A G G A T S P L
HHII T G G L P A T A T E D V K N V Q V A N A D L T E A
 K A A L T A A G V T G T A S V V K I S Y T D N N G
 K T I D G G L A V K V G D D Y S A T Q N K D G S
 I S I N T T K Y T A D D G T S K T A L N K L G G A
 D G K T E V V S I G G K T Y A A S K A E G H N F K
 A Q P D L A E A A A T T E N P L Q K I D A A L A
 Q V D T L R S D L G A V Q N R F N S A I T N L G N
 T V N N L T S A R S I E D S D Y A T E V S N I S
 R A Q I L Q Q A G T S V L A Q A N Q V P Q N V L S
 L L R **W S H P Q F E K**
ATG G C A C A A G T G A T T A A T A C A A A C A G C T G T C G T G T T G A C C C A G A A T A A C C T G A A C A A A T C C C A G T C C G C T C
 T G G G C A C C G C T A T C G A G C G T C T G T C T T C C G G T C T G C G T A T C A A C A G C G C G A A A G A C G A T G C G G C A G G T C A G G C
 G A T T G C T A A C C G T T T A C C G C G A A C A T C A A A G G T C T G A C T C A G G C T C C C G T A A C C G T A A C G A C G G T A T C T C C
 A T T G C G C A G A C C A C T G A A G G C G C G T G A A C G A A A T C A A C A C A A C C T G C A G C G T G T G C G T G A A C T G G C G G T T
 C A G T C G C T A A C A G C A C C A C C C A G T C G A C C T C G A C T C C A T C C A G G C T G A A A T C A C C C A G C G C T G A A C G
 A A A T C G A C C G T G T A T C C G G C C A G A C T C A G T T C A A C G G C G T G A A A G T C C T G G C G C A G G A C A A C C C T G A C C A
 T C C A G G T T G G T G C C A A C G A C G G T G A A A C T A T C G A T A T C G A T C T G A A G C A G A T C A A C T C T C A G A C C C T G G G T C T
 G G A T A C G C T G A A T G T G C A A C A A A A A T A T A A G G T C A G C G A T A C G G C T G C A A C T G T T A C A G G A T A T G C C G A T A C
 T A C G A T T G C T T T A G A C A A T A G T A C T T T T A A A G C C T C G G C T A C T G G T C T T G G T G G T A C T G A C C A G A A A A T T G A T G
 G C G A T T T A A A A T T T G A T G A T A C G A C T G G A A A A T A T T A C C C A A A A G T T A C C G T T A C G G G G **ATG** A C T G G T A A A G
 A T G G C T A T T A T G A A G T T T C C G T T G A T A A G A C G A A C G G T G A G G T G A C T C T T G C T G C G G T G C G A C T T C C C G C T
 T A C A G G T G G A C T A C C T G C G A C A G C A A C T G A G G A T G T G A A A A T G T A C A A G T T G C A A G T T G C A A T G C T G A T T G A C A G A
 G G C T A A A G C C G A T T G A C A G C A G C A G G T G T T A C C G G C A C A G C A T C T G T T G T T A A G **ATC** T C T T A T A C T G A T A A T
 A A C C G T A A A A C T A T T A T G T G G T G T T A G C A G T T A A G G T A G G C G A T G A T T A C T A T T C T G C A A C T C A A A A T A A A G
 A T G G T T C C A T A A G T A T T A A T A C T A C G A A A T A C A C T G C A G A T G A C G G T A C A T C C A A A A C T G C A T A A A C A A A A C T
 G G G T G G C G C A G A C G G C A A A A C C G A A G T T G T T C T A T T G G T G G T A A A A C T T A C G C T G C A A G T A A A G C C G A A G G
 T C A C A A C T T T A A A G C A C A G C C T G A T C T G C G G A A G C G G C T G C T A C A A C C A C C G A A A C C C G C T G C A G A A A A T
 T G A T G C T G C T T T G G C A C A G G T T G A C A C G T T A C G T T C T G A C C T G G G T G C G G T A C A G A A C C G T T C A A C T C C G C T A
 T T A C C A C C T G G G C A A C A C C G T A A A C A A C C T G A C T T C T G C C C G T A G C G G A T C G A A G A T T C C G A C T A C C G G A C
 C G A A G T T T C C A A C **ATC** T C C G C G C A G A T T C T G A C A G C A G G C G G T A C C T C C G T T C T G G C C A G G C G A A C C A G
 G T T C C G C A A A A C G T C C T C T C T T A C T G C G T **TGGAGCCATCCGCAGTTTGAAAA** T A A

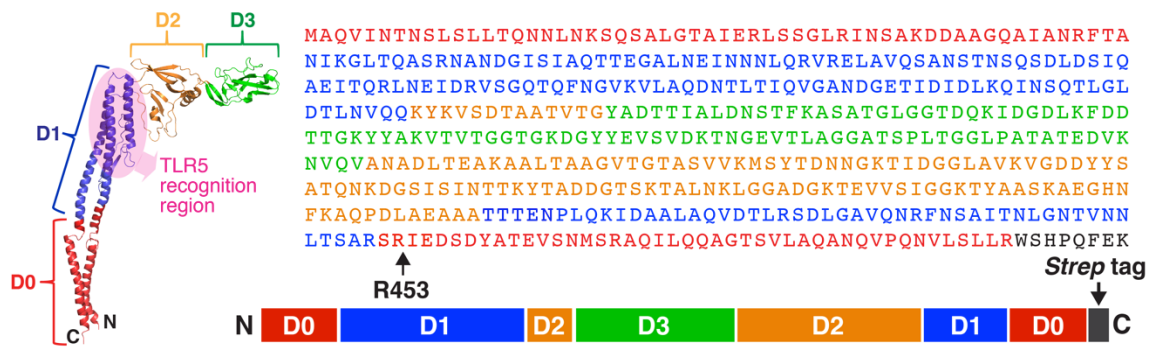
CAPS represent the flagellin. **Underlined BOLD CAPS** represent the *Strep*-tag II polypeptide. The mutations were marked in red color.

Supplementary Table S2. The sequence of Hbc protein.

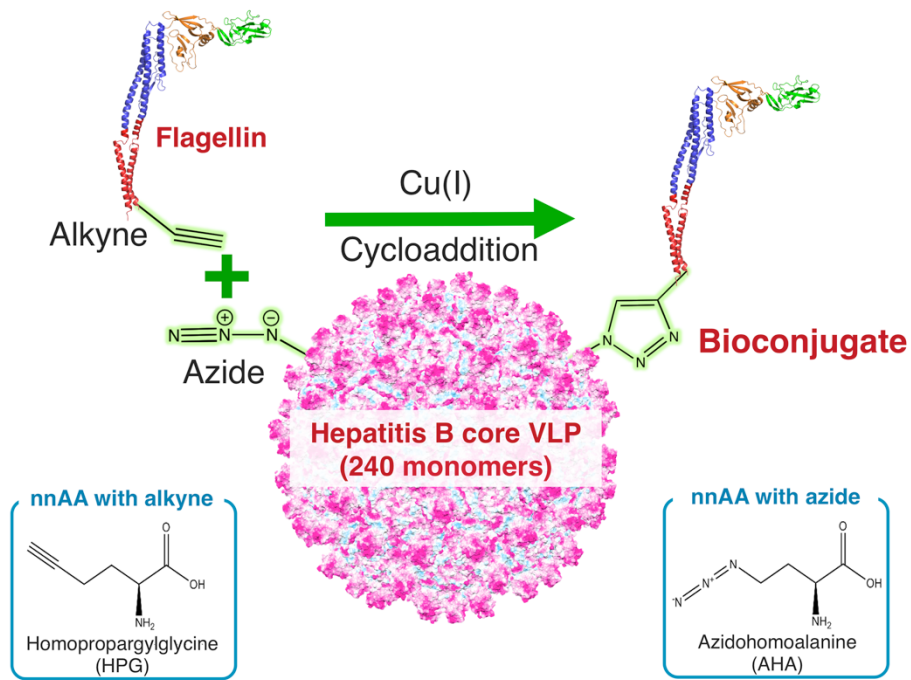
| Variant | Protein sequence | DNA encoding sequence |
|---------|--|--|
| Hbc | M D I D P Y K E F G A T V E L L S F L P S D F F P S V R CLLD T A A A L Y R D A L E S P E H C S P H H T A L R Q A I L C W G D L S T L A T W V G T N (AHA) E D P A S R D L V V S Y V N T N V G L K F R O L L W F H I S C L T F G R E T V L E Y L V S F G V W I CT P P A Y R P P N A P I L S T L P E T T V V | A T G G A T A T C G A C C C G T A C A A A G A A T T C G G C G C A C C G T T G A A C T G C T G T C T T T C C T G C C G T C T G A T T T C T C C C G T C T G T T C G T TGC C T G T G G A C A C C G C G C A G C A C T G T A C C G T G A C G C G C T G G A A T C T C C G G A A C A T T G T T C C C G C A T C A C A C T G C G T G C G T C A G G C G A T T C T G T G C T G G G G C G A C C T G agc A C C T G G C G A C T T G G G T T G G C A C C A A C atg G A A G A T C C G G C G C T C G T G A T C T G G T T G T T C T T A C G T T A A C A C T A A C G T T G G T C T G A A A T T C C G T C A G C T G C T G T G G T T C C A C A T C T C T T G C C T G A C T T C G G T C G T G A A A C C G T T C T G G A A T A C C T G G T T C T T T T G G T G T T G G A T T TGT A C T C C G C C G G C T T A C C G T C C G C C G A A C G C A C C G A T C C T G A G C A C C C T G C C G G A A A C C A C T G T T G I G T A A T A A |

CAPS represent the Hbc protein. **Underlined** represent the mutations. The mutations for introducing a disulfide bond network were marked in red color.

Supplementary Figures



Supplementary Figure S1. Positions of flagellin domains (D0, D1, D2 and D3) in amino acid sequence and possible truncation position (R453).



Supplementary Figure S2. Diagram of Cu(I)-catalyzed [3 + 2] cycloaddition click chemistry reaction for the direct coupling of flagellin proteins to HBc VLPs. The HBc VLP is stabilized by introducing a SS1(D29C-R127C) disulfide bond network¹.

Reference

1. Lu, Y., Chan, W., Ko, B. Y., VanLang, C. C. & Swartz, J. R. Assessing sequence plasticity of a virus-like nanoparticle by evolution toward a versatile scaffold for vaccines and drug delivery. *Proc. Natl. Acad. Sci. USA* **112**, 12360-12365 (2015).