## **Supplementary Materials**

Table S1. The sequences of PCR primers used in this study				
Name	Sequences (5' to 3')			
MinD-For	ATCCCTTTTTAACAAGGAATTTTTATGGCACGCATTATT- GTGTAGGCTGGAGCTGCTTC			
MinD-Rev	AAAATCCAGTAATGCCATAATTTATCCTCCGAACAGGCG- ATGGGAATTAGCCATGGTCC			
araD-C	CCAGATTCATCAACGCGCCCCCATGGGACGCGTTTTTAGAGGCA- TTAGTGGTGGTGGTGGTGG			
araB-C	TCTCTACTGTTTCTCCATACCTGTTTTTCTGGATGGAGTAAGACG- ATGACTATGACAAGACTGAAGA			

Table S1. The sequences of PCR primers used in this study
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Table S2. The details of aptamer sequences and modification sites

Name	5'- modify	3'- modify	Sequences (5' to 3')
Cy3-DC-Apt	Cy3	cholesteryl	Cy3-GGGAGGUGUGUUAGCACACGAU UCAUAAUCAGCUACCCUCCC-choleste ryl
DC-Apt	-	cholesteryl	GGGAGGUGUGUUAGCACACGAUUC AUAAUCAGCUACCCUCCC-cholesteryl

Table S3. The amino acid sequences of OsmY signal peptide and C1

Name	Amino acid sequence
OsmY signal peptide	MTMTRLKISKTLLAVMLTSAVATGSAFA
	SIKEDVQFGGGSTLHDMGIFSITSSDSGGGSDPKRTIQKKSGGGSGLS
	IITPPEGGYESKTKDTPSQNNPKNDAQKTEIQPTQVIDGPFAGGKDT
	VVNIFRLNTNADGTIRVGGFKASLTTNAAHLHIGEGGVNLSNQASG
C1	RTLLVENLTGNITVEGALRVNNQVGGAAVAGSSANFEFKAGTDTNN
	GTATFNNDIHLGKAVNLRVDAGGGSVADKYDVQVAIHTDTKKMEG
	VLIPAGFIKVTILEPGGGSMEIQQTHRKINRPIISLALVGVLMGTELGA
	NTPNDPIHSESRAFFTGGGSEQILQNQGYKVIS

Figure S1. The structure diagram of OsmY-C1. The red arrow indicates the OsmY signal peptide; the green arrow represents the peptide combination of C1; the purple arrow shows the location of His-tag; and the gray arrow indicates the termination codon.



Figure S2. Proliferation and cytotoxicity effects of wild-type (WT) minicells, TA-2m, and Apt-TA-2m on DC2.4 cells. DC2.4 cells were incubated with varying concentrations of WT minicells, TA-2m, or Apt-TA-2m at 37 °C for 24 hours with 5% of CO<sub>2</sub>. Cell viability was assessed using the CCK-8 assay. Significance was determined using One-Way ANOVA and Tukey's multiple comparison with an adjusted *p*-value < 0.05. Groups labeled with the same letter (e.g., 'a' vs. 'a', or 'a' vs. 'ab') are not significantly different ( $p \ge 0.05$ ), as they share at least one common letter. Conversely, groups labeled with different letters (e.g., 'a' vs. 'b') are considered significantly different (p < 0.05).



Figure S3. Pathological changes in the spleens of mice immunized with the highest dose  $(2 \times 10^8)$  of wild-type minicells (WT), TA-2m, or Apt-TA-2m. A total of 48 female BALB/c mice (6–8 weeks old) were randomly divided into four groups (n = 12 per group). On Day 0, mice in Groups 1, 2, and 3 received an oral administration of 100 µL PBS containing  $2 \times 10^8$  WT minicells, TA-2m, or Apt-TA-2m, respectively. Group 4 served as the control and received 100 µL of PBS via the same route. At 1, 3, 5, and 7 days post-immunization (dpi), three mice from each group (n = 3) were euthanized for histopathological examination. No significant pathological changes were observed in the spleens of any treatment group at the indicated time points.



Figure S4. The FAC scatter plots. Thirty-two mice were randomly divided into four groups (n=8 per group). On Day 0, mice in each group were orally administered 100 μL of PBS or PBS containing 2×10<sup>8</sup> wild-type minicells, TA-2m, or Apt-TA-2m, followed by booster immunizations on Days 14 and 28. One week after the final immunization, all mice were anesthetized and sacrificed for FCA. (a) Percentages specific CD4+ T cells or CD8+ T cells producing IFN-γ. (b) Percentages specific CD4+ T cells or CD8+ T cells producing IL-4. (c) Percentages specific CD4+ T cells or CD8+ T cells producing IL-6. (d) Percentages specific CD4+ T cells or CD8+ T cells producing IL-17a.

