Supplementary Information (SI) for Journal of Materials Chemistry B.

Supporting raw data of Western Blot

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Enhanced gene transfection ability of sulfonylated low molecular weight PEI and its application in anti-tumor immunotherapy

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Comment of the Publishing Editor: The reported Western blot results in Figure 4 and figure S4 appear to lack supporting raw data, and are not reported in line with our experimental data guidelines. I am therefore writing to request all raw data files to support your manuscript. This should include full, uncropped data showing a representative portion of the gel, with control markers and full molecular weight scales included. Please send these files by return email (to materialsb@rsc.org) as soon as possible.

Response: Thanks for the reminding, here we provide the uncropped Western blot images for Figure 4C and Figure S4 (as Fig. R1 and R2 below). In this assay, the PVDF membrane was first cut into pieces according to the molecular weight of the target proteins after the membrane transfer procedure. Then these pieces of membrane were incubated with their respective primary and secondary antibodies, and finally stained by ECL substrate and recorded by a chemiluminescence imaging system.

The background of these images is not exactly the same, the reason is as follows: the intracellular levels of the internal control proteins (like GAPDH) are usually higher than other target proteins. When recorded by an imaging system, the corresponding Western Blot strips of the internal control proteins on the membrane are prone to overexposure. In other words, if the exposure times of all proteins were set to be the same, either the internal control protein strips would be overexposed, or the strips of other proteins would not obvious. Therefore, these membranes of different proteins were recorded by the imaging system individually, and the exposure time of GAPDH was set shorter than other proteins, making the background of GAPDH brighter than other proteins.

Based on this, in Fig. 4C and Fig. S4, it's suitable for comparison between different groups of the same protein, but not suitable for comparison between different proteins. If necessary, relevant descriptions may be added to the caption of the figure.

In the last well of Fig. R1, we did a parallel experiment of the Ns-P/pOVA/Man group, and the results were consistent, so we retained only one set of data. In Fig. R2, we also verified the activation of STING pathway by other materials synthesized in this study, such as Ts-P and Fs-P. Since these results were not relevant to the purpose of this study, only the results in the red dashed box are shown in Fig. S4.

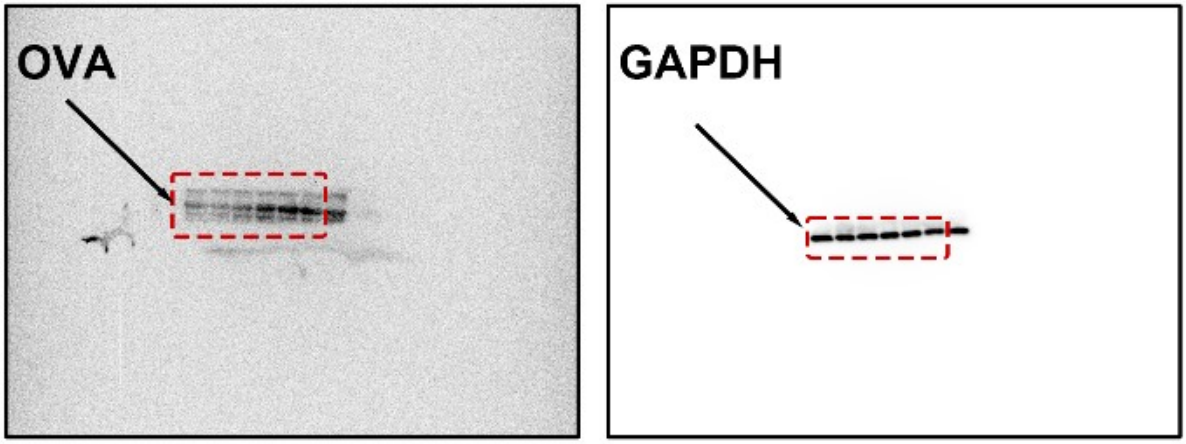

Fig. R1. Supporting uncropped western blot images for OVA and GAPDH (an internal control). The samples within the red dashed box from left to right are groups i – vi. (i) Control; (ii) pOVA; (iii) Ns-P/pOVA; (iv) Ns-P/pOVA/Man; (v) PEI 25k; (vi) Lipo 2000.

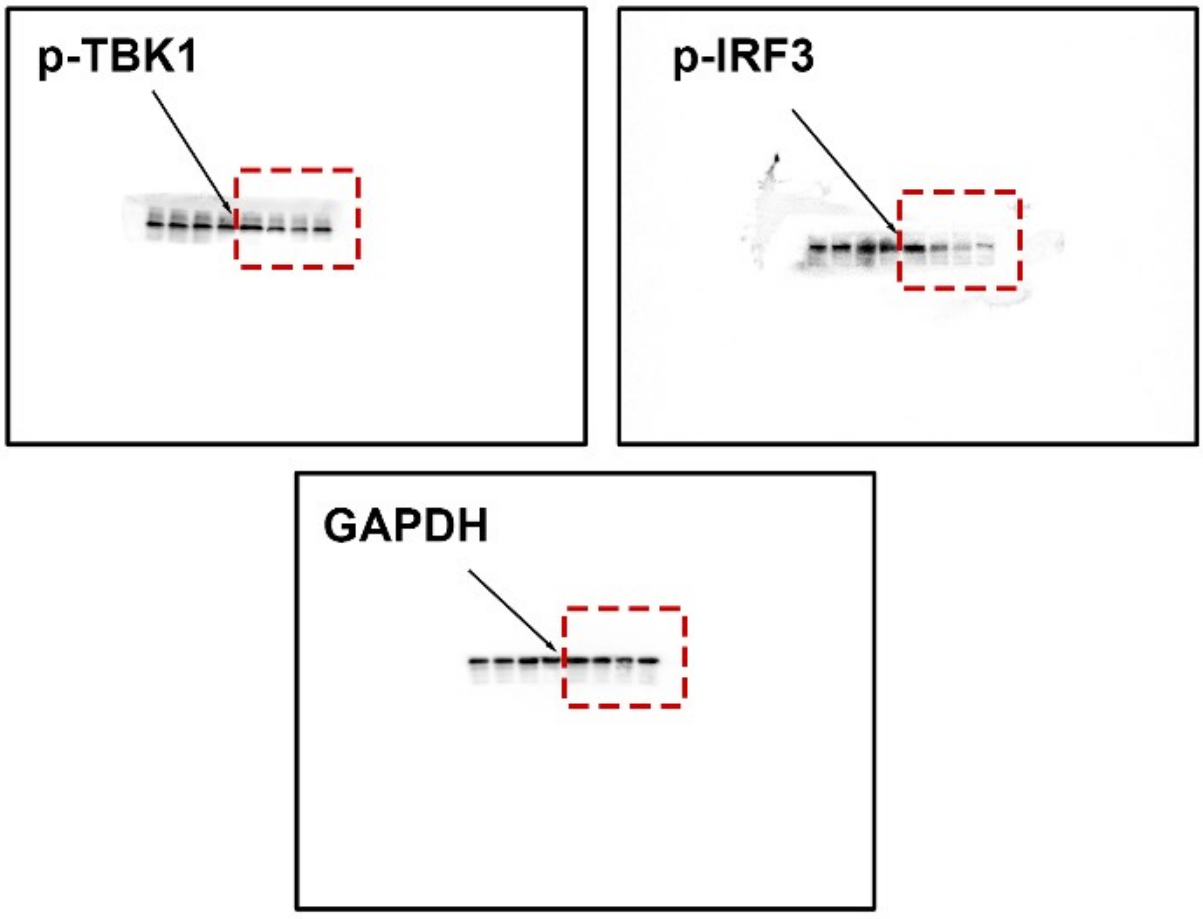

Fig. R2. Supporting uncropped western blot images for p-TBK1, p-IRF3 and GAPDH (an internal control). The samples within the red dashed box from left to right are groups i – iv. (i) Ns-P; (ii) Ns-P/DNA; (iii) DNA; (iv) Control.