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**Supplementary information**

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**The CRL5–SPSB3 ubiquitin ligase targets nuclear cGAS for degradation**

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In the format provided by the authors and unedited

## The CRL5<sup>SPSB3</sup> ubiquitin ligase targets nuclear cGAS for degradation

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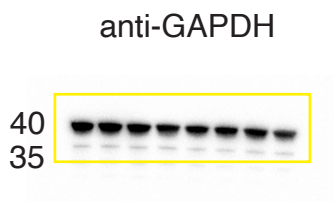
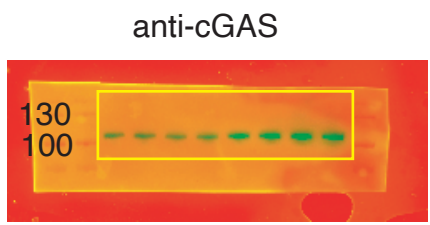
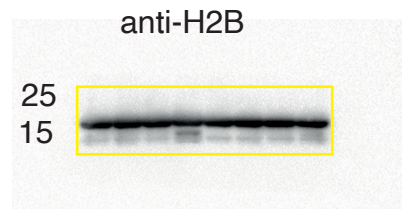
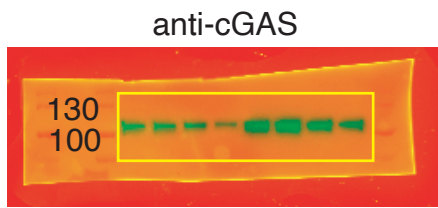
Supplementary Figure 1 | Full scans for all immunoblots and Coomassie Blue stainings.

Supplementary Figure 2 | Flow cytometric gating strategy

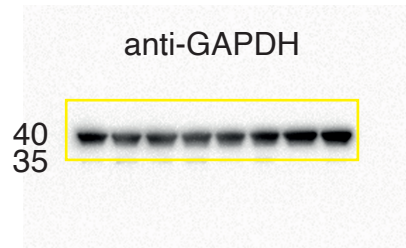
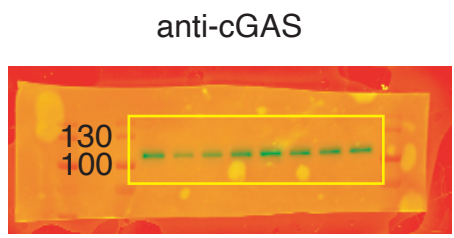
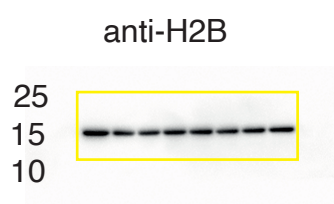
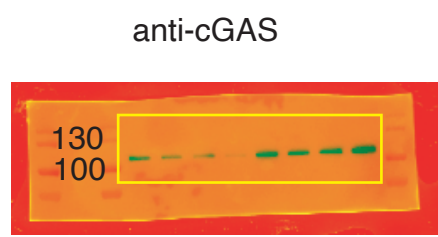


**Fig.1**

**i**

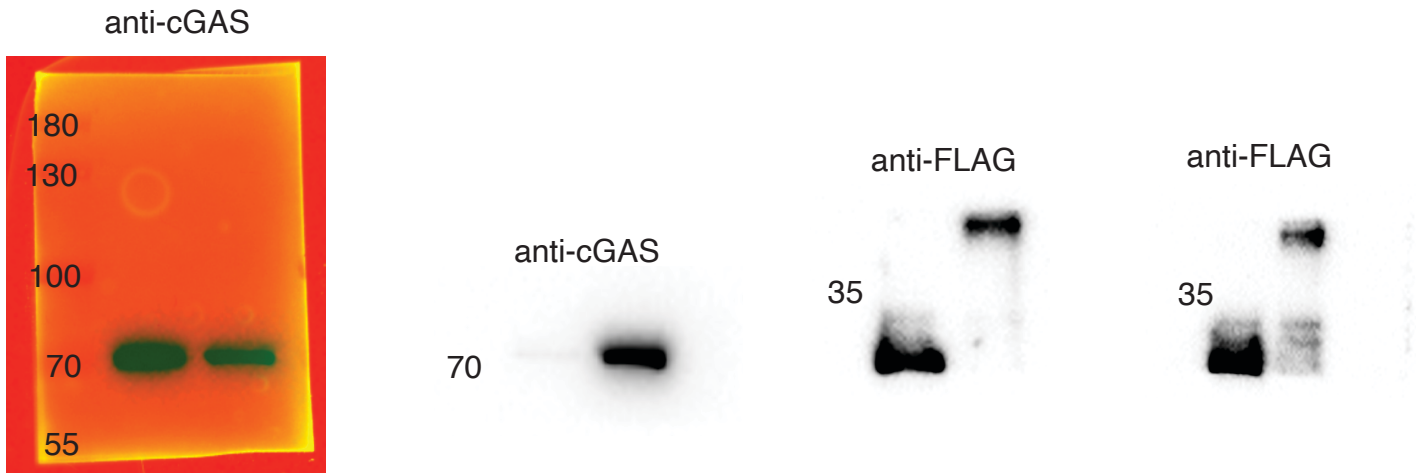


**j**

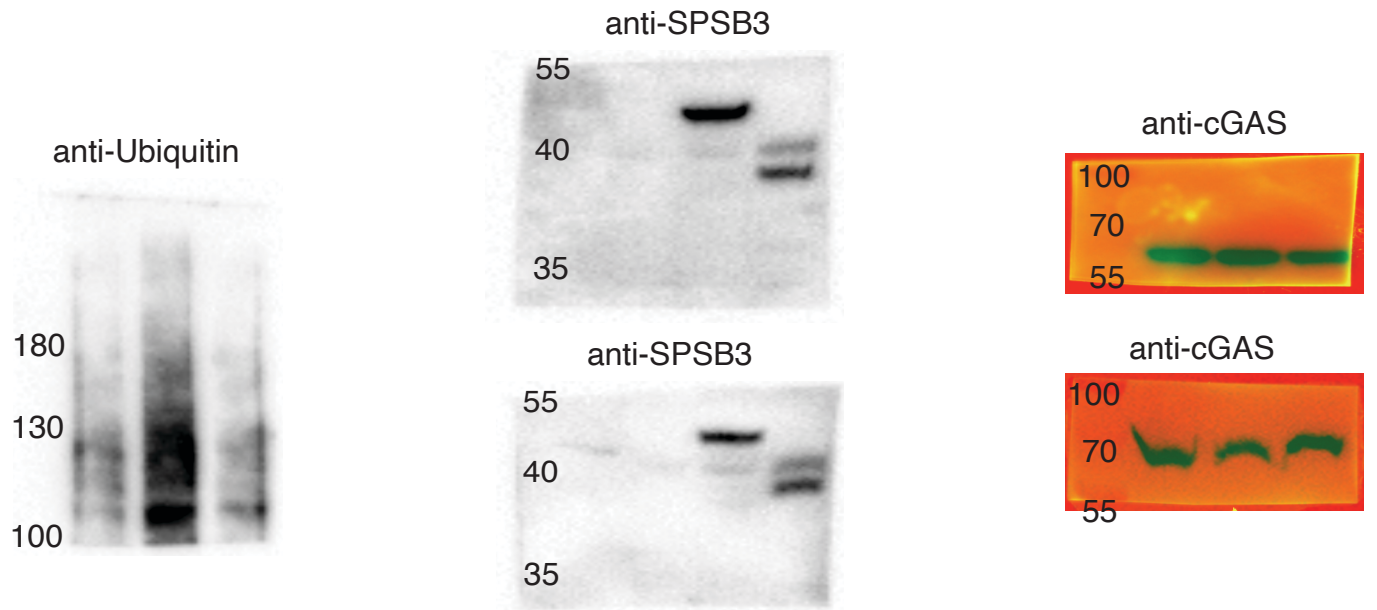


**Fig.2**

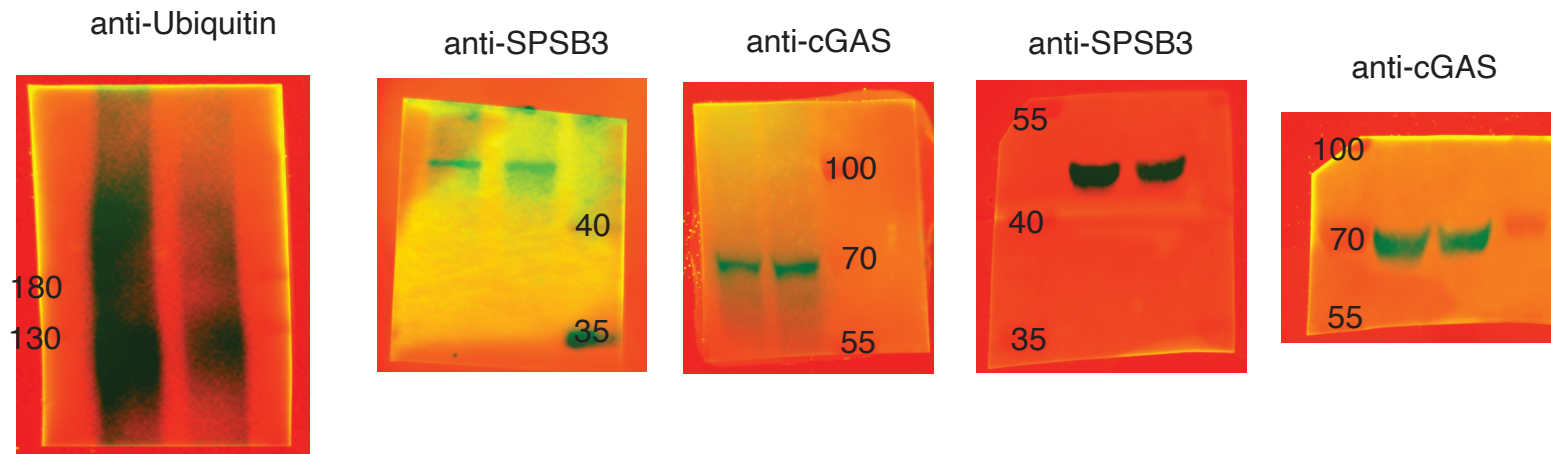
**a**



**b**



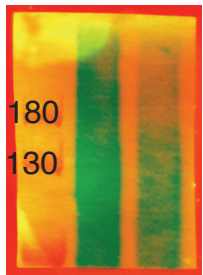
**c**



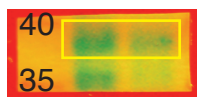
**Fig.2**

**d**

anti-k48Ubiquitin



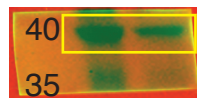
anti-SPSB3



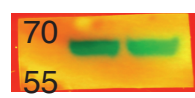
anti-cGAS



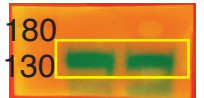
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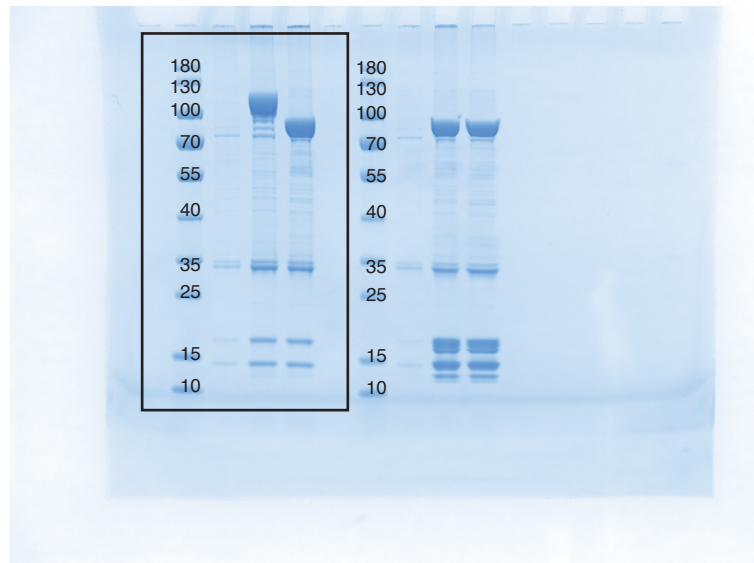
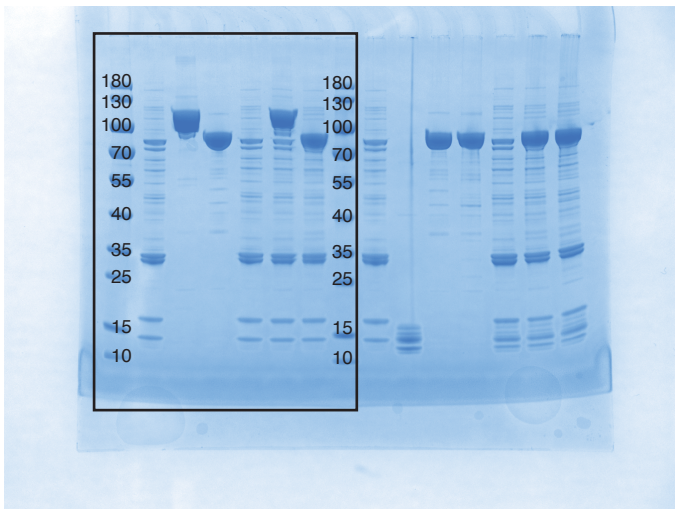
anti-cGAS



anti-Vinculin

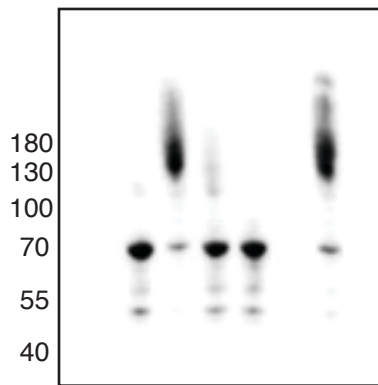


**e**



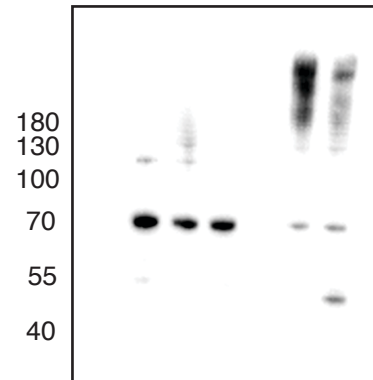
**f**

anti-cGAS



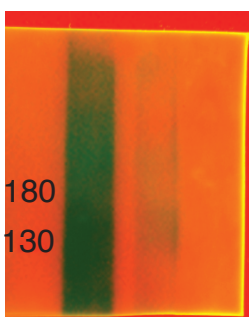
**h**

anti-cGAS

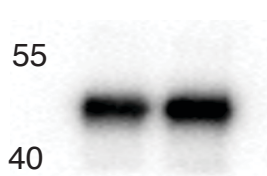


**i**

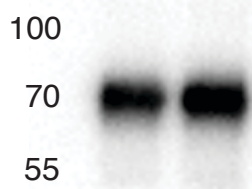
anti-Ubiquitin



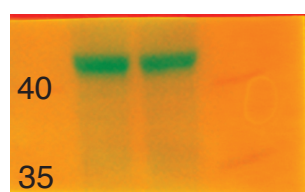
anti-SPSB3



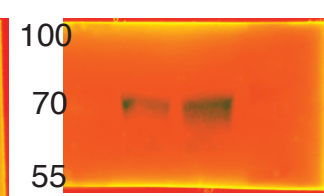
anti-cGAS



anti-SPSB3



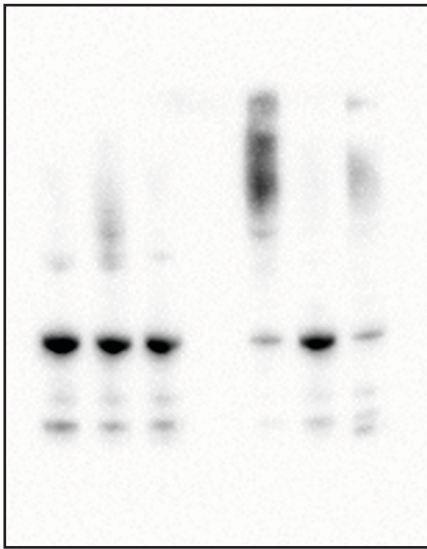
anti-cGAS



**Fig.4**

anti-cGAS

**b**



**c**

anti-Ubiquitin

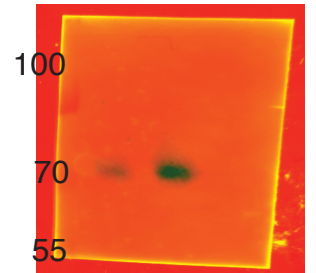
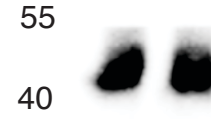
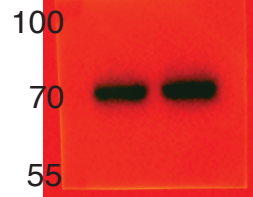
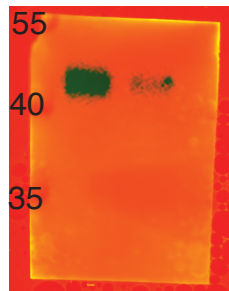
anti-SPSB3

anti-cGAS

anti-SPSB3

anti-cGAS

180  
130  
100

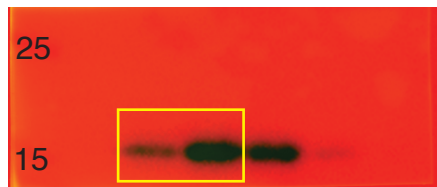
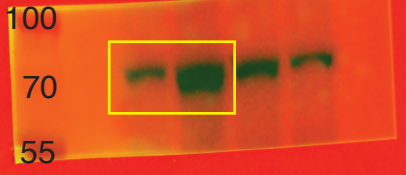


**d**

anti-cGAS

anti-ISG15

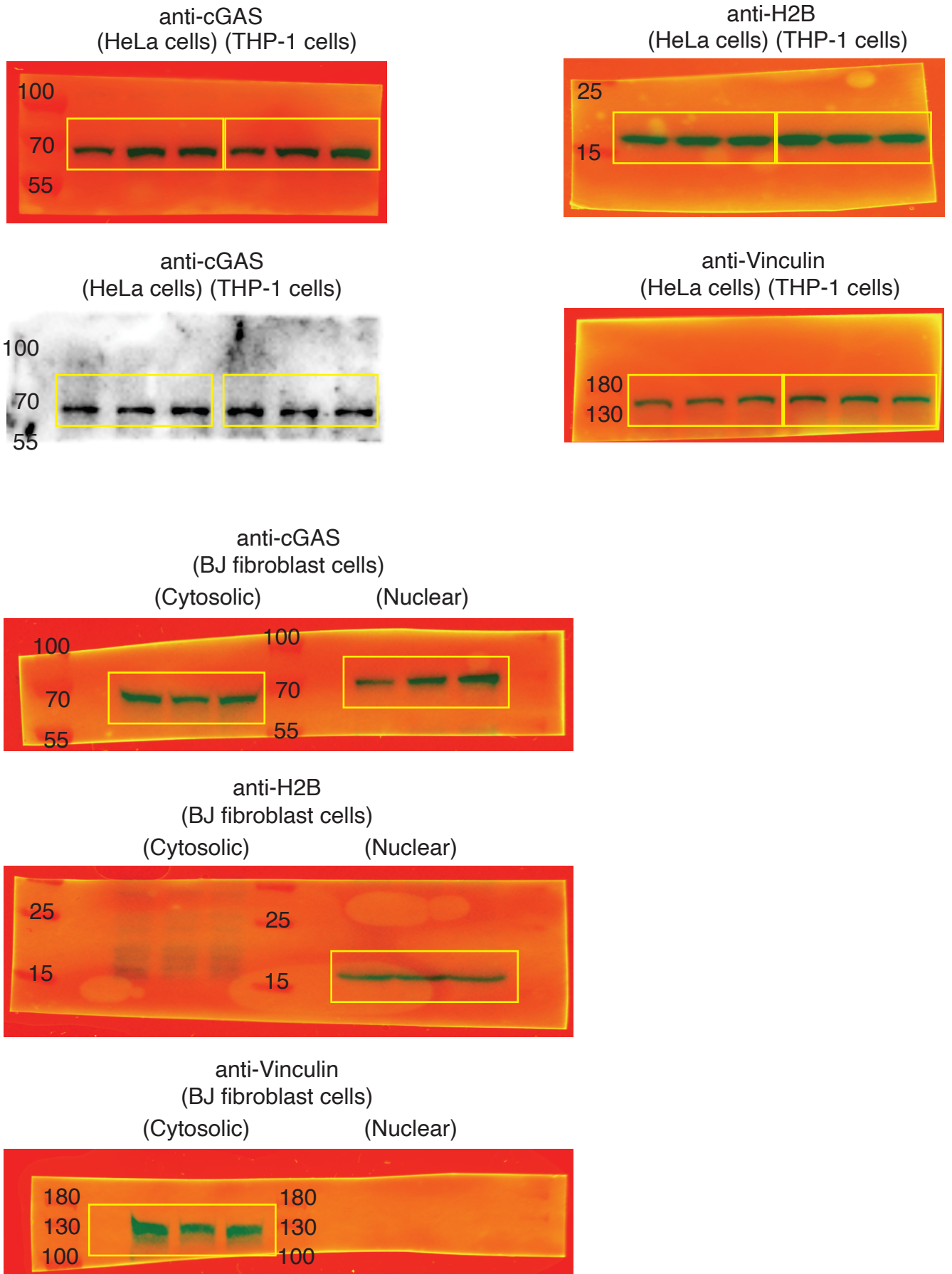
anti-GAPDH





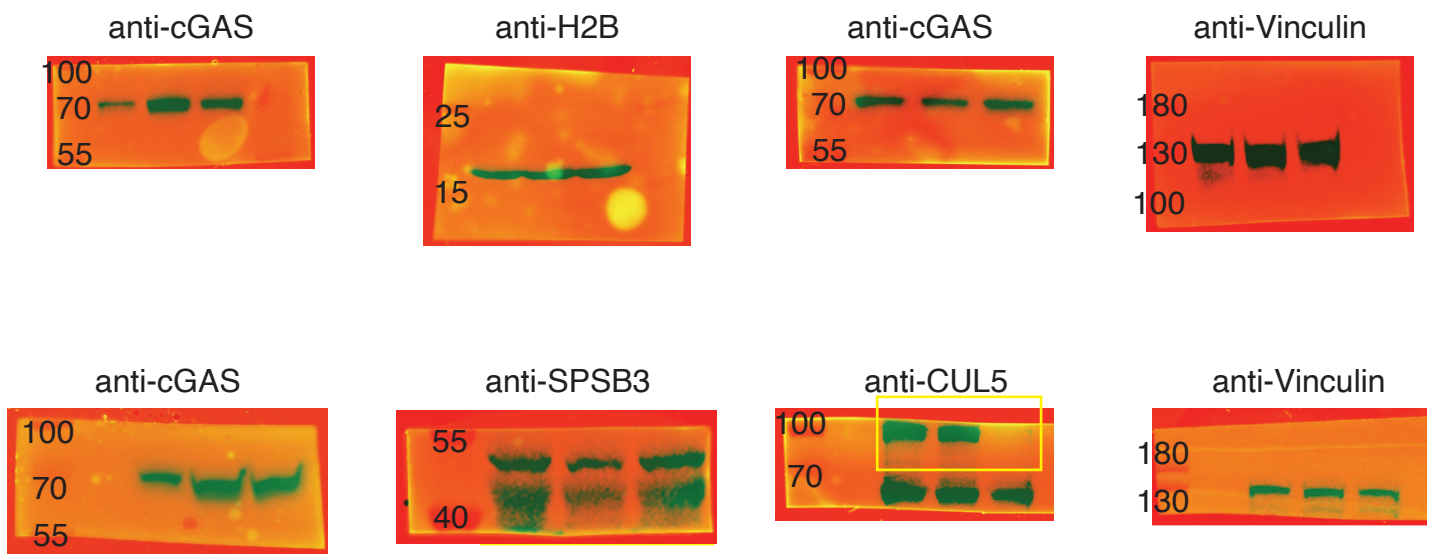
Extended Fig.2

f

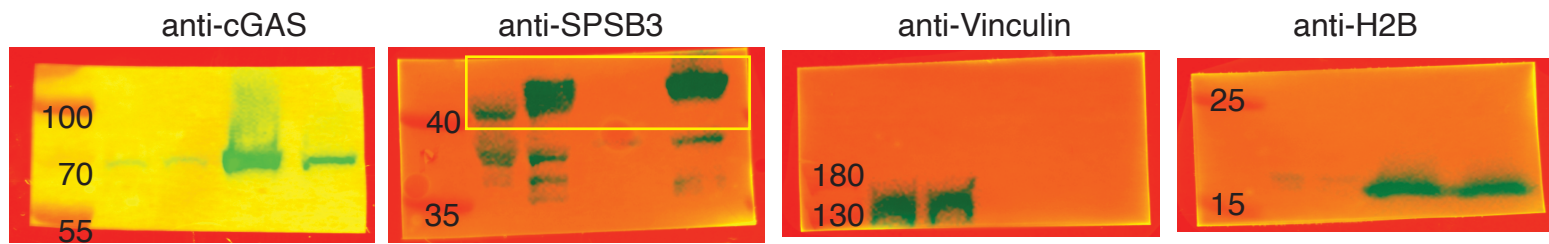


Extended Fig.3

e

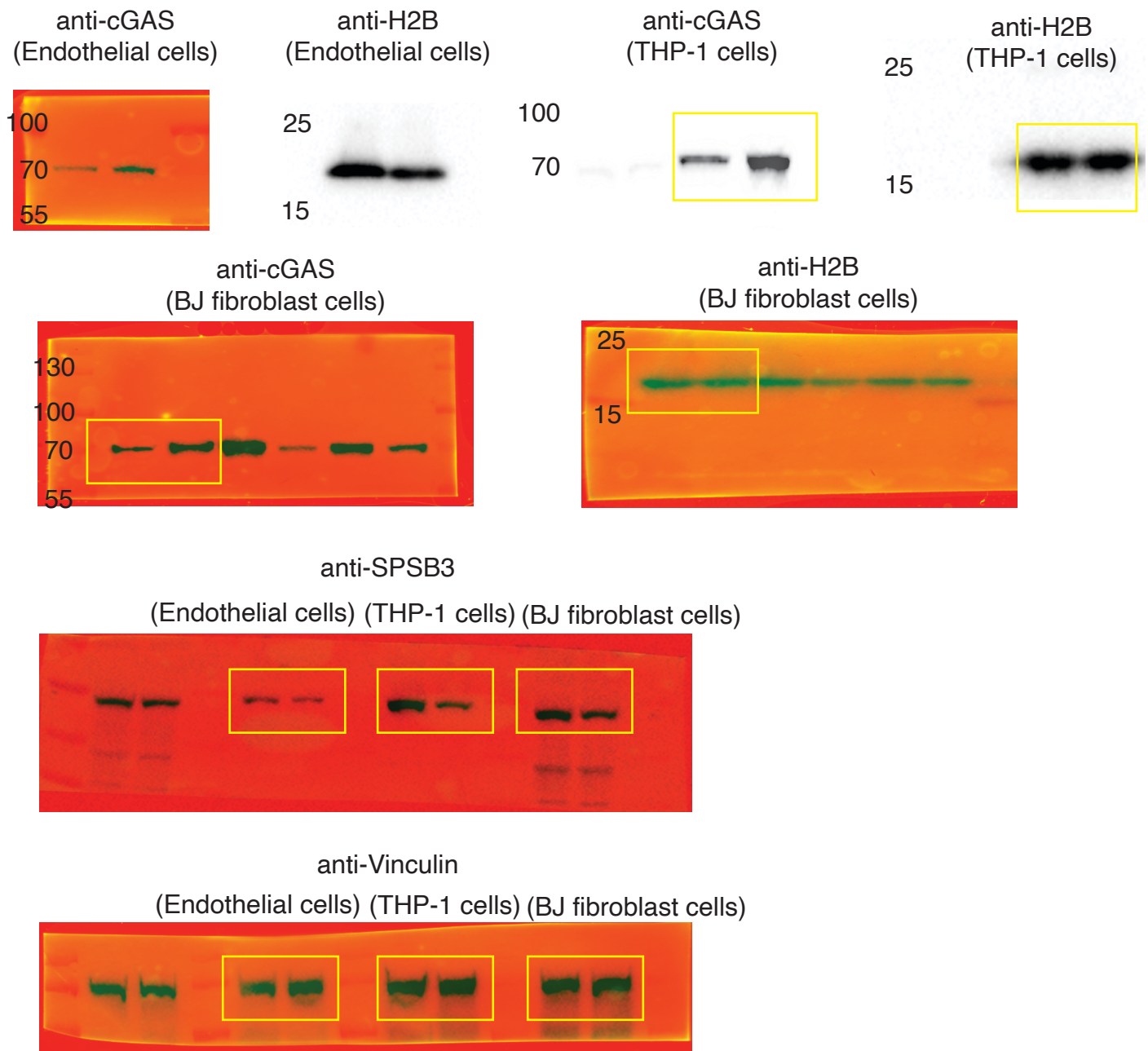


k

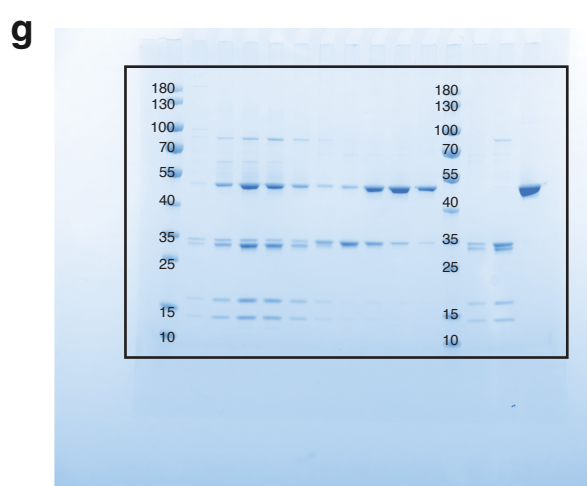
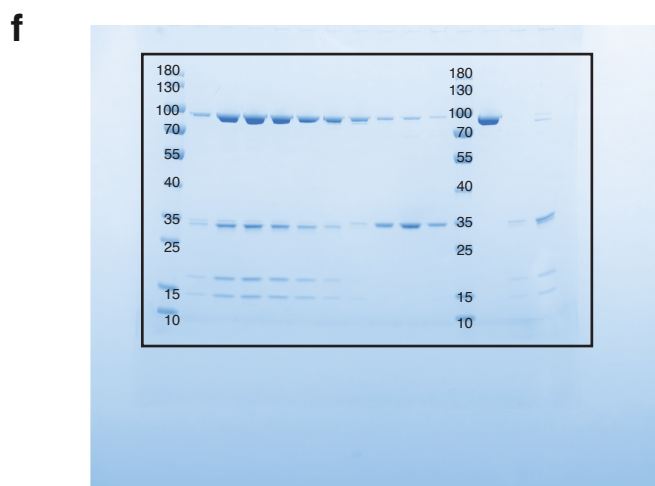
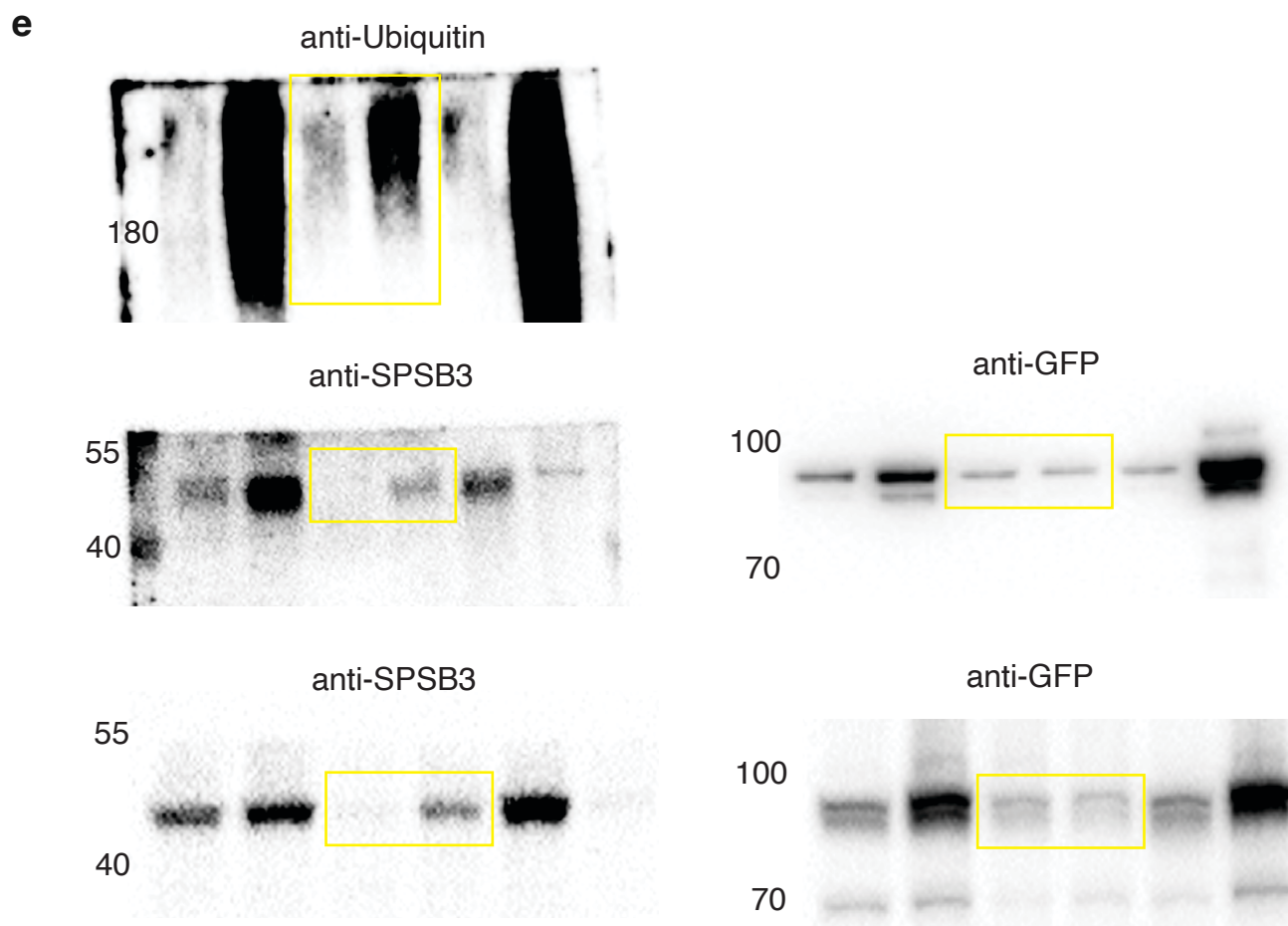
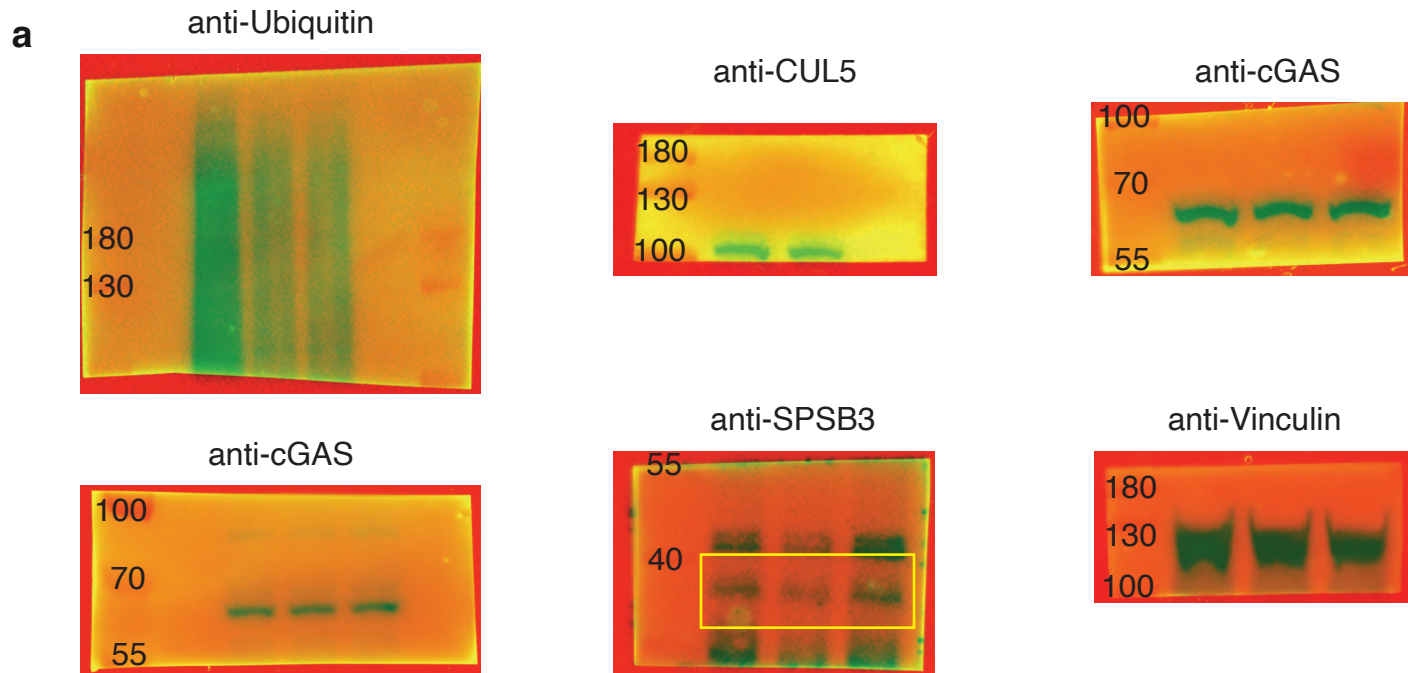


Extended Fig.3

i

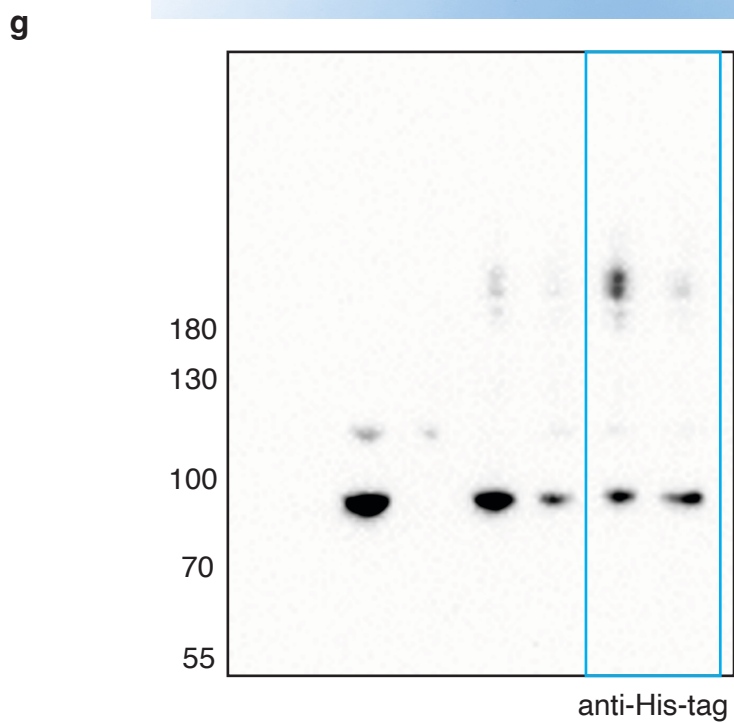
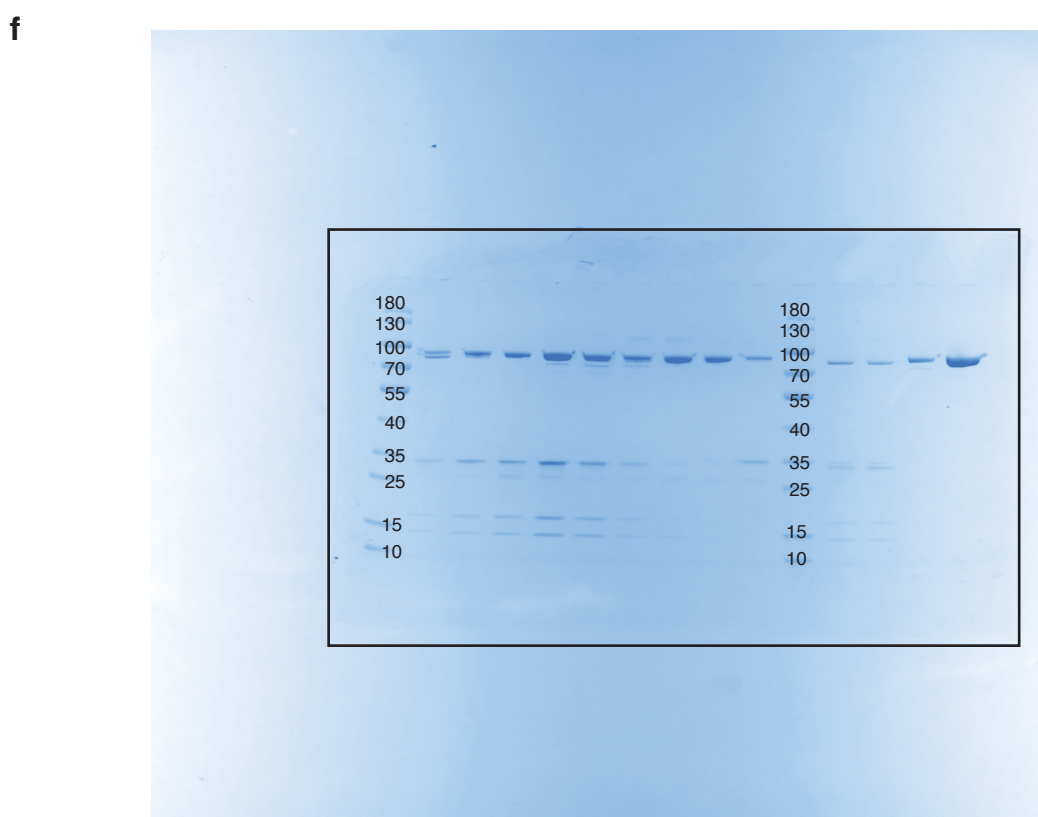
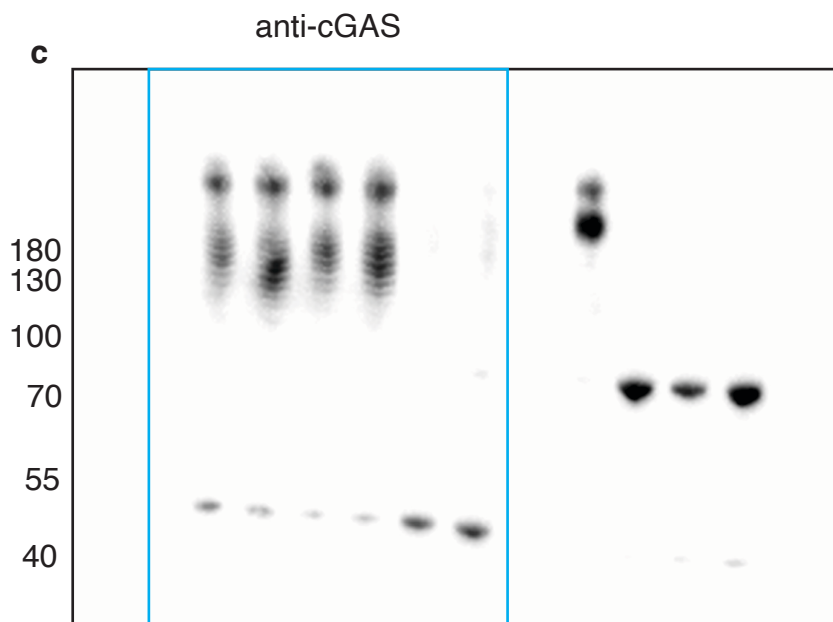
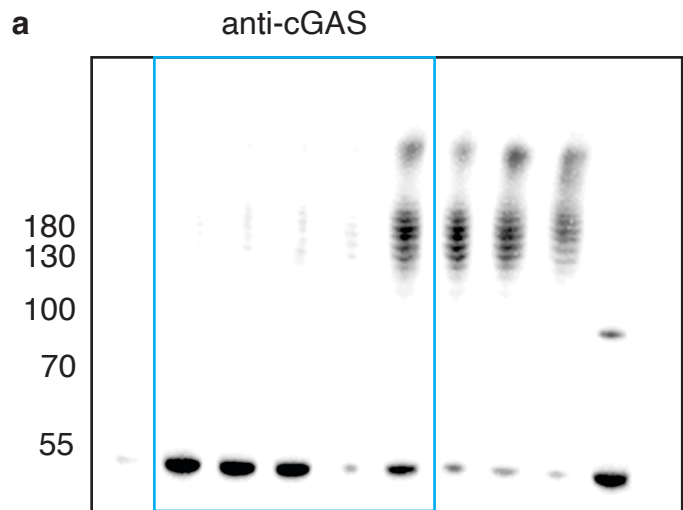


Extended Fig.4



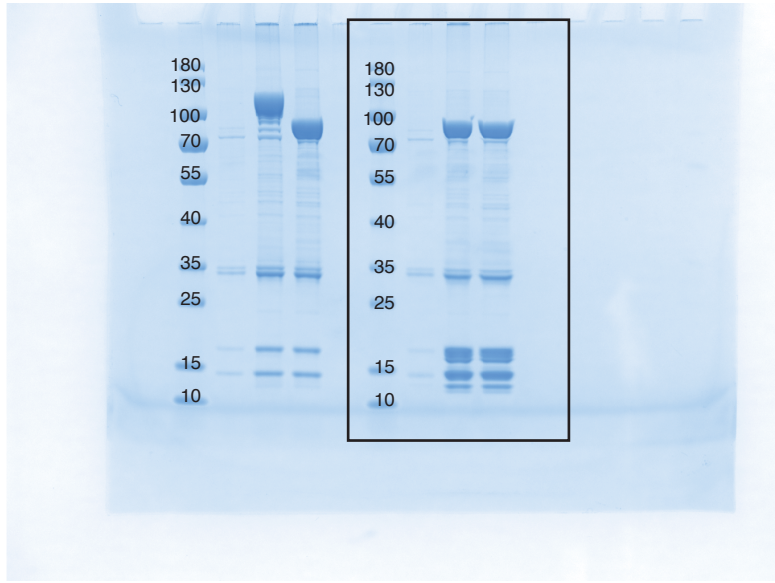
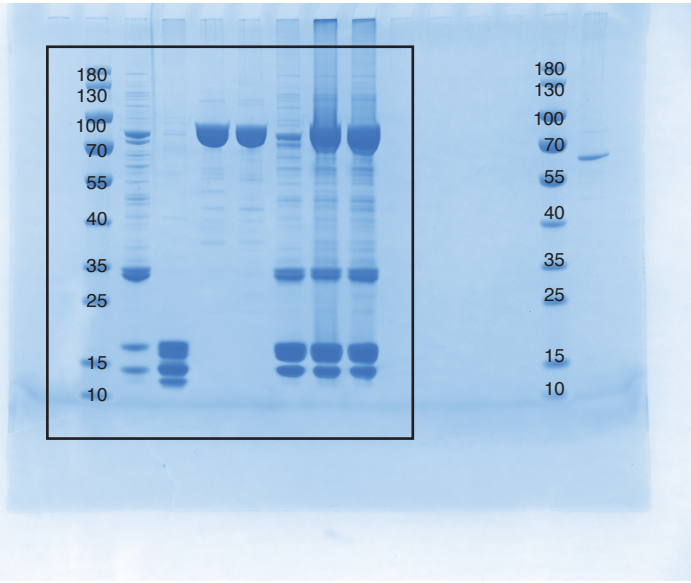


Extended Fig.5

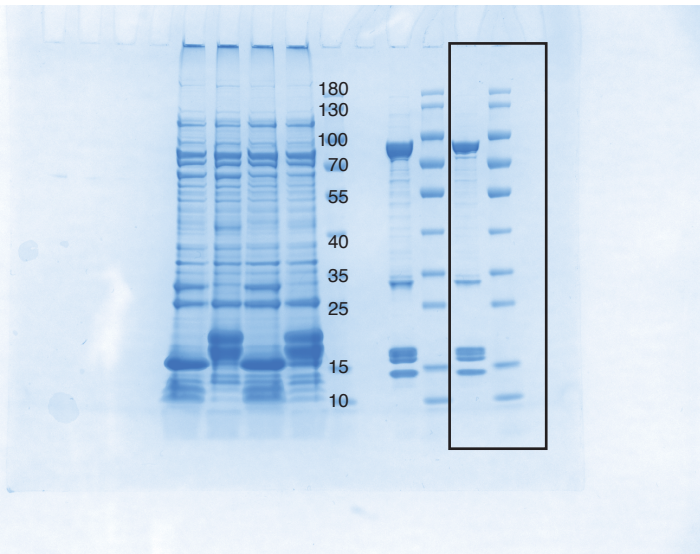


# Extended Fig.6

**a**

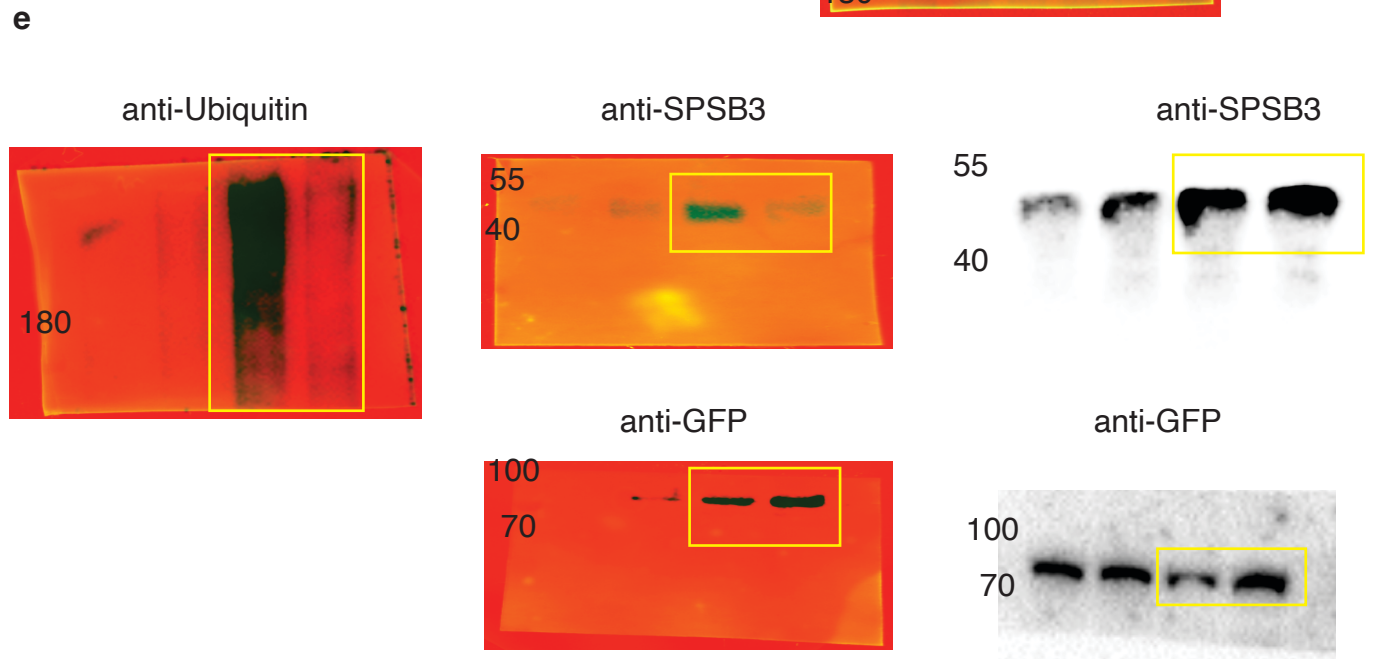
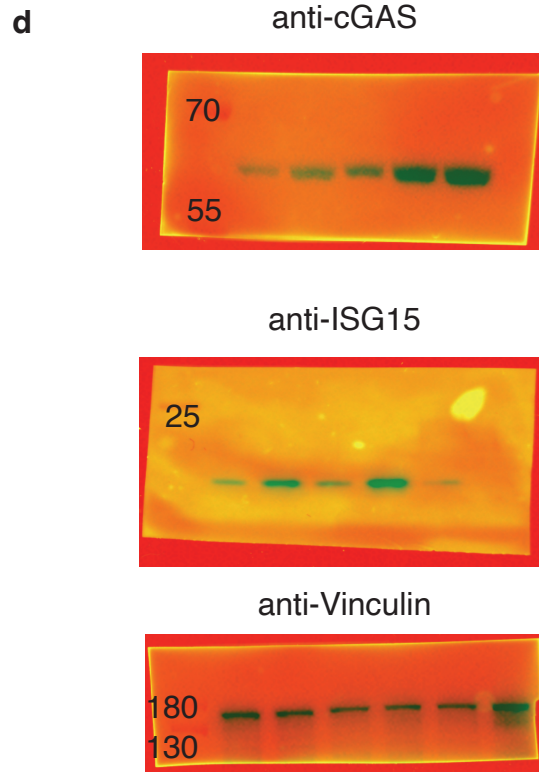
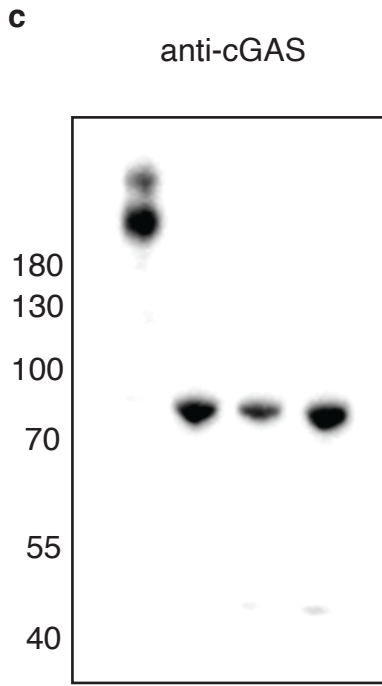


**c**



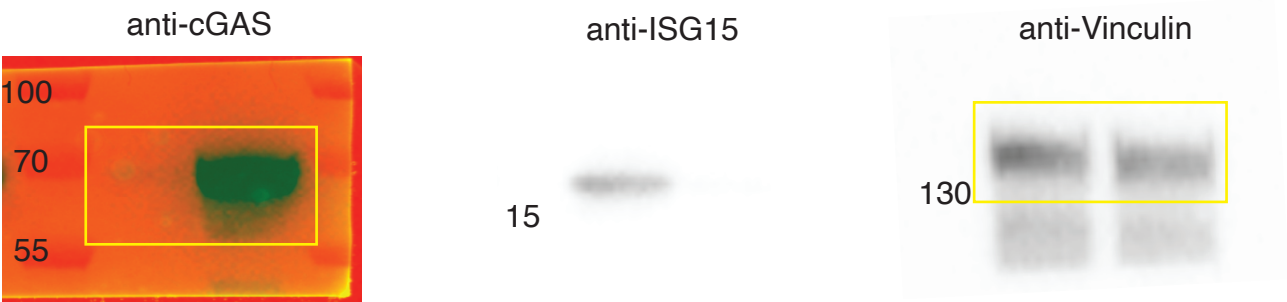


Extended Fig.9

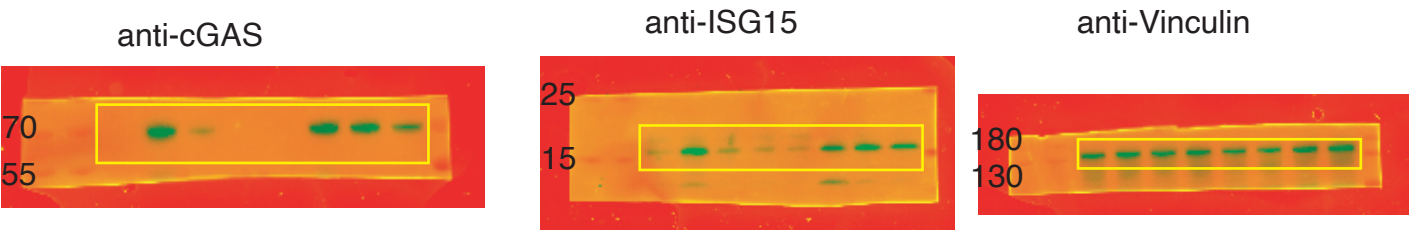


Extended Fig.10

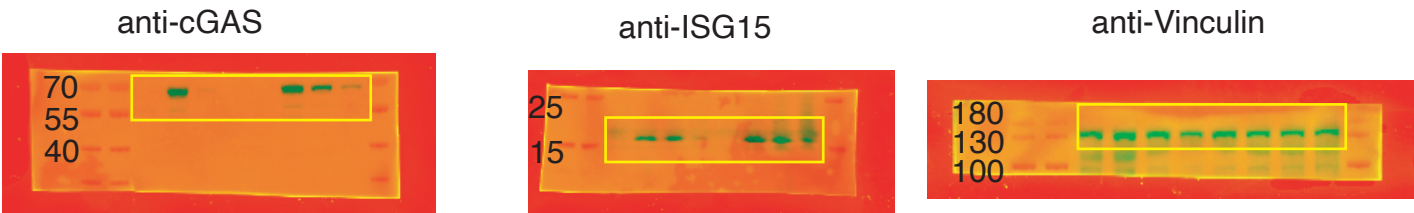
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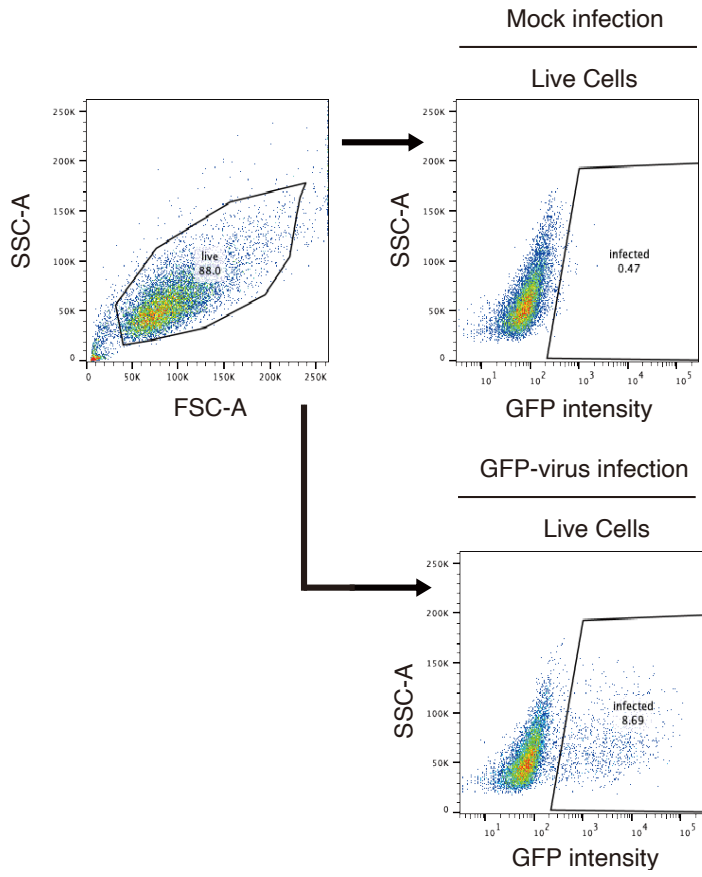
d



e



Infected HeLa cells gating strategy



Supplementary Fig.2 | The flow cytometry gating strategy for quantification of GFP<sup>+</sup> cells, related to Figure 4i, 4j and Extended Data Figure 11k, is shown.