Supplementary data

Studies on the Japanese soil-borne wheat mosaic virus movement protein highlight its ability to bind plant RNA

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Table S1: Primer sequences

| Primer name | Sequence 5'-3' | Determination |
|---------------------|--|---|
| MP_GW_For1 | GGG GAC AAG TTT GTA CAA AAA AGC AGG CTT AAA CAT GAC AAC TAA AGA TGT TTC | GATEWAY-cloning of MP ^{JSBWMV} , addition of |
| MP_GW_Rev1 | GGG GAC CAC TTT GTA CAA GAA AGC TGG GTC CAC TAT GTT TTC AGA GTC | attB-sites GATEWAY-cloning of MP ^{JSBWMV} , addition of attB-sites |
| MP_fw_GG | TTA AGG TCT CGT TGC ATG ACA ACT AAA GAT GTT TCA AGT GAT TCC | GoldenGate cloning of MPJSBWMV:GFP into |
| | | E.coli expression plasmid pET28a |
| eGFP_rev_GG | TTA AGG TCT CGA AGC TTA CTT GTA CAG CTC GTC CAT GC | GoldenGate cloning of MP ^{JSBWMV} :GFP into <i>E.coli</i> expression |
| | | plasmid pET28a |
| eGFP_For | GGGG ACA AGT TTG TAC AAA AAA GCA GGC TTC ATG GTG AGC AAG GGC GAG | GATEWAY-cloning of free GFP |
| eGFP_Rev | GGGG AC CAC TTT GTA CAA GAA AGC TGG GTC TTA CTT GTA CAG CTC GTC CAT | GATEWAY-cloning of free GFP |
| MP_toRFP_NSp_For1 | CCG AAG ATG CTC CCT GAA CA | Modification of pJS1 |
| Bgl2_inRFP_NSp_Rev1 | GAA GAT CTT TAG GCG CCG GTG G | Modification of pJS1 |
| RNA2_for | GAT CTA ATA CGA CTC ACT ATA GTT GAG TGT TAA CTC TTC TTG | Full length RNA2 synthesis |
| RNA2_rev | CTC CGG TTC GGG GGG | RNA2 full length and tRNA-like structure- RNA2 synthesis |
| tRNA2_for | GAT CTA ATA CGA CTC ACT ATA TGT ATG TTC TGT TGA ACT ACT GT ATG | tRNA-like structure- RNA2 synthesis |
| tRNA1_for | GAT CTA ATA CGA CTC ACT ATA CAT AGT GTG ATT ATT ACT ATT ATG | tRNA-like structure- RNA1 synthesis |
| tRNA1_rev | CTC CGG TTC AGG GGG | tRNA-like structure- RNA1 synthesis |
| Non_tRNA1_for | GAT CTA ATA CGA CTC ACT ATA AGT CGT TAG ACT GCT TAA AG | non tRNA1-like- structure synthesis |
| Non_tRNA1_rev | TCA CAC TAT GTT TTC AGA GTC CAA | non tRNA1-like- structure synthesis |
| Non_tRNA2_for | GAT CTA ATA CGA CTC ACT ATA GTT GAG TGT TAA CTC TTC TTG | non tRNA2-like- structure synthesis |
| Non_tRNA2_rev | TCA CAG AGG TTT AGA CTT CCT T | non tRNA2-like- structure synthesis |

24 °C 17 °C



Figure S1. JSBWMV forms visible infection sites on *Chenopodium quinoa* leaves at **17** °C but not at **24** °C. *Chenopodium quinoa l*eaves of five to six-week-old plants were infected with RNA in-vitro transcribed from pJS1 and pJS2 cDNA clones (Yamamiya and Shirako, 2000) and kept in the greenhouse with 16h light and 8h dark cycles and at least 200 W/m² light intensity either at 24°C or at 17°C. Multicellular infection sites visible as chlorotic spots were only obtained at 17°C. Pictures were taken 12 dpi.

Figure S2:

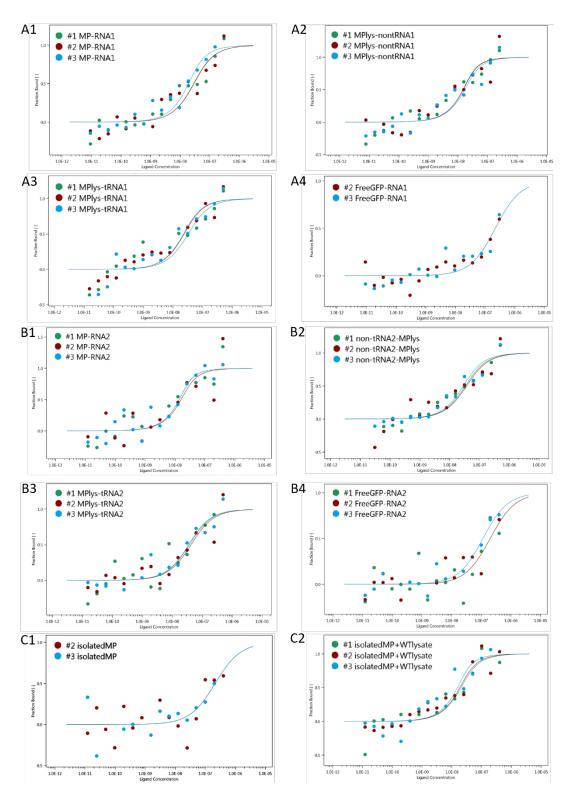


Figure S2: Microscale thermophoresis binding curves for MP^{JSBWMV}:GFP or GFP, respectively to viral RNAs

N. benthamiana leaf crude extracts expressing MP^{JSBWMV} and GFP after agro-inoculation were used to analyze the binding affinity of (**A1**) MP^{JSBWMV}:GFP to RNA1^{JSBWMV}, (**A2**) MP^{JSBWMV}:GFP to RNA1^{JSBWMV} non-tRNA, (**A3**) MP^{JSBWMV}:GFP to tRNA1^{JSBWMV}, (**A4**) GFP to RNA1^{JSBWMV}, (**B1**) MP^{JSBWMV}:GFP to RNA2^{JSBWMV}:GFP to tRNA2^{JSBWMV} of the RNA2^{JSBWMV} of the RNA2^{JSBWM}

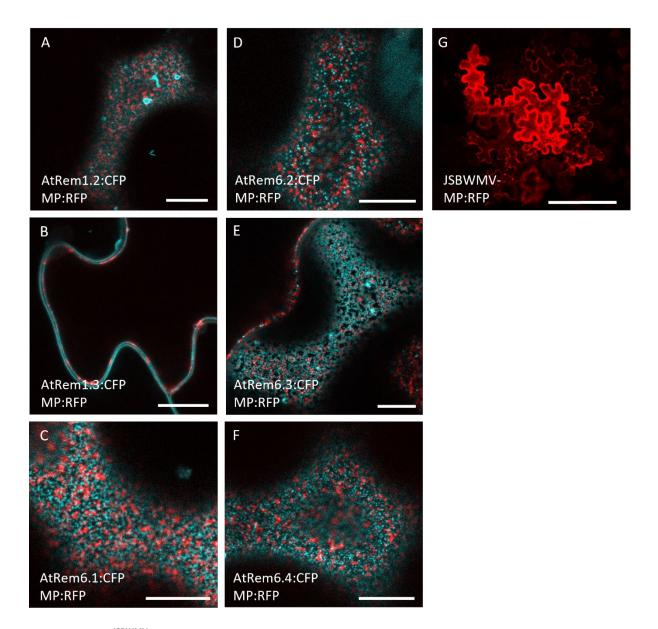


Figure S3: MPJSBWMV:RFP localizes to membrane rafts.

(A-F) MP^{JSBWMV}:RFP was ectopically expressed in *N. benthamiana* epidermal cells via agroinoculation and co-expressed with different markers. (A-F) MP^{JSBWMV}:RFP (red) co-expressed with (A) AtREM1.2:CFP (cyan); (B) AtREM1.3:CFP (cyan); (C) AtREM6.1:CFP (cyan); (D) AtREM6.2:CFP (cyan); (E) AtREM6.3:CFP (cyan); (F) AtREM6.4:CFP (cyan), remorins and MP^{JSBWMV}:RFP localize in a patchy pattern at the cell periphery. The localizations do not overlap. Images taken two to five days post infiltration. Scale bars are 10 μm. (G) JSBWMV-MP:RFP infection site seven days after rub inoculation of RNA^{JSBWMV-MP:RFP} into *N. benthamiana* epidermal cells. Scale bar is 200 μm.