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Studying transmission activation of aphidvectored *Cauliflower mosaic virus*

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It has been assumed for a long time that virus acquisition was simple contamination of the stylets occurring when aphids feed on infected plants. However, non-circulative Cauliflower mosaic virus (CaMV) forms during aphid punctures specific transmission morphs in infected cells. More precisely, the CaMV transmission helper protein P2 is dispatched from cytoplasmic transmission body inclusions and virus particles from virus factory inclusions to form together and transiently transmission morphs on microtubules that are acquired and transmitted by the vector. This 'Transmission Activation' (TA), discovered first for CaMV and recently described for the Potyvirus Turnip mosaic virus, requires that the virus recognizes the presence of aphids via the plant perception systems, and then induces TA. We want 1) to characterize TA reaction in living tissue after biotic (aphid punctures) and abiotic stresses, and 2) to capture and identify plant partners involved in TA. For this, we chose GFP, which serves as a fluorescent in vivo reporter, and can be used to immunocapture interaction factors by the GFP trap technique. Since the CaMV genome cannot accommodate the entire GFP sequence, we implemented the split GFP system, based on spontaneous auto-assembling of the non-fluorescent small GFP11 (16 amino acids) and big GFP1-10 (rest of the molecule) fragments to fluorescent GFP. A recombinant CaMV with GFP11 fused to P2 was constructed and used to infect transgenic Arabidopsis expressing GFP1-10. The virus was infectious and infected cells displayed P2-GFP fluorescence in transmission body-like structures. When validated, the system might help to identify TA elicitors by tracking GFP11-P2 behavior after application of candidate molecules. The same approach will be used for viral protein P6, with the goal to potentially detect early infection events at the site of aphid-inoculated cells. P6 is particularly suited for this because it is the first viral protein to appear during infection.