Illuminating SBWMV-host interaction – Subcellular localization and function of CP-RT during virus infection

Nico Sprotte, Claudia Janina Strauch and Annette Niehl

Julius Kühn Institute (JKI) – Federal Research Centre for Cultivated Plants, Institute for Epidemiology and Pathogen Diagnostics, Braunschweig

E-mail of corresponding author: nico.sprotte@julius-kuehn.de

Soil-borne cereal viruses cause substantial crop losses and therefore represent an extensive threat for agriculture in Europe, Asia and America. The Furovirus Soil-borne wheat mosaic virus (SBWMV) infects several crop species like wheat, rye or barley and is transmitted by a soil-borne plasmodiophorid, called Polymyx graminis. Resistances against Furovirus infections are barely described; in wheat, the Sbm1 and Sbm2 genes code for a translocation resistance, which restricts the infection to the plant roots. The viral movement protein (MP) and coat protein –readthrough (CP-RT) protein are believed to play a major role in infection of plant roots and the translocation of the virus into upper plant tissues.

We modified a SBWMV cDNA clone to express fluorescent CPRT fusion proteins (CP-RT:FP) to illuminate their subcellular localization and uncover the function of the CP-RT protein during SBWMV infection. The subcellular localization of the CP-RT:FP was compared with fluorescent markerproteins expressed in Nicotiana benthamiana mutants as well as wild-type plants, which were transiently transformed by Agrobacterium-infiltration prior to virus infection. Fluorescent infection sites were studied by confocal laser scanning microscopy.

Our results show that the CP-RT:FP co-localizes with the endoplasmic reticulum (ER) and forms dot-like structures along the ER, tightly associated with the ER membrane. Following this observation, we investigated the role of CP-RT for the plant secretory pathway, as this is a common route for virus spread in higher plants. This was done by leaf infiltration of Brefeldin A (BFA) in transiently transformed N. benthamiana leaves. BFA is a fungal toxin, which blocks the vesicle formation between ER and Golgi and thus leading to the emergence of distinctive hybrid compartments. Moreover, co-expression of CP-RT:FP with GTP-locked and GFP-tagged Sar1, a small GTPase which regulates COPII-Vesicle formation at the ER, revealed co-localization at dot-like structures, indicating that the ER-associated dot-like CP-RT:FP structures might represent sub-ER sites at which CP-RT accumulates during virus infection. We hope that this knowledge will provide new ideas and targets for the development of novel resistance strategies against soil-borne virus disease.