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Natural multiplicity of infection of plant cells by genomes of *Cauliflower mosaic virus* populations

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The number of viral genomes co-infecting host cells during the course of a systemic infection (the multiplicity of cellular infection, MOI) is a parameter of foremost importance for several aspects of the biology of viruses. While a low MOI may reduce the intra-cellular competition between genetic variants, it also greatly limits the extent of possible complementation, and *de facto* reduces the recombination frequency. In contrast, a high MOI would augment the possibility of trans-complementation between variants, and the recombination frequency, but potentially also enhance accumulation of defective genomes and increase competition for replication. These simple examples illustrate the predicted contrasted effects of MOI on viral genomes and populations, which open the possibility for important trade-offs on this particular trait.

Despite its importance, very little information is available on the MOI of viruses infecting eukaryotic hosts. A mean MOI value of 4 has been previously determined only for a baculovirus infecting insects. Also, an average value of 3 proviruses has been estimated in the HIV-infected spleen cells of two patients, suggesting an MOI above this value. In plants, the MOI has not been directly investigated at the cellular level for any virus species.

Using *Cauliflower mosaic virus* infecting turnip host plants, we measured both the ratio of two co-infecting genetic variants, and the rate of cellular co-infection within single leaves. A statistical model, based on maximum likelihood, allowed the calculation of mean MOI values from these data at different stages of the systemic infection. Indeed, under the hypothesis that the probability of cellular infection by a given variant depends on its relative frequency within the leaf, and that infection by one and/or the other variant are independent, the cellular co-infection rate for a given ratio of the two variants directly depends on the number of viral genomes infecting each cell.

This experiment yielded several remarkable results. The CaMV infects host cells at a very elevated MOI reaching values around 10, largely exceeding the rare estimates cited above. The MOI is not a constant; it greatly increases from leaf to leaf during the course of host invasion to drop late in infection close to initial values. This drop seems induced by flowering. Finally, at a given time point, different MOI values are measurable in different leaves.