Editorial

Advanced Expression Vector Systems: New Weapons for Plant Research and Biotechnology

Tzvi Tzfira, Stanislav V. Kozlovsky, and Vitaly Citovsky[1]*

Department of Molecular, Cellular, and Developmental Biology, University of Michigan, Ann Arbor, Michigan 48109 (T.T.); Department of Virology, Moscow State University, Moscow 119992, Russia (S.V.K.); and Department of Biochemistry and Cell Biology, State University of New York, Stony Brook, New York 11794–5215 (V.C.)

Scientific discoveries often coincide with the development of new and robust methodologies. Modern plant biology and biotechnology are of no exception to this rule, especially when it comes to production of new vector systems for gene expression. Thus, for example, the progress in plant genetic engineering could not have been as productive as it is today without the development of small, easy-to-manipulate, and simple-to-use *Agrobacterium* binary vectors (e.g. Komari et al., 2006; Komori et al., 2007), and studies of protein subcellular localization in plant cells have been greatly simplified and advanced with the introduction of vectors that express GFP fusions (Goodin et al., 2007). Indeed, the ability to transiently and stably express foreign genes, to genetically interfere with the expression of native genes, and to functionally study the expression, interaction, localization, and modification of proteins in cells, tissues, and whole plants are fundamental to modern plant basic research and biotechnology.

More than two decades had passed since the introduction of the first generation of plant transformation binary vectors (e.g. Bevan, 1984). Although revolutionary at their time, these vectors were rather simply designed, lacking cloning and expression versatility, and offered very little flexibility for their manipulation for specific research or application purposes. Vector technology has evolved throughout the years, and during this time plant transformation vectors have been a subject of constant improvements (e.g. Becker et al., 1992; Datla et al., 1992; Hajdukiewicz et al., 1994). Newer generations of plant transformation vectors provided plant biologists and biotechnologists with improved strategies for cloning and delivering their genes of interest into plant cells, typically using *Agrobacterium* as vehicle for the transformation process. Some of these vectors were developed as families of plasmids, and others represented single constructs designed for specific purposes. For example, the pCB mini-binary vector series of plasmids (Xiang et al., 1999) provided an excellent alternative for the relatively large first-generation binary plasmids, whereas the pBI121 plasmid—only recently sequenced (Chen et al., 2003) but no longer available commercially—was specifically designed to foster the use of the *GUS* gene as a reporter in genetic transformation experiments.

More recently, we have witnessed an impressive increase in the ''introduction'' of new and novel vectors suitable for performing various tasks for plant research and biotechnology (Fig. 1). These days, it seems that one can find a plasmid for every task, including such relatively unique applications as activation tagging (e.g. the pSKI015 and pSKI074 binary vectors; Weigel et al., 2000) or dexamethasone-inducible expression (e.g. the pOp/LhGR transcription activation system; Samalova et al., 2005). Also, vectors have been constructed to allow plant biologists to take advantage of radically new cloning methodologies, such as recombinase-mediated gene cloning (Gateway; e.g. the pMDC plasmid collection; Curtis and Grossniklaus, 2003), or of new approaches to modulate gene expression, such as RNA interference-mediated gene silencing (Meyer et al., 2004) and virus-induced gene silencing (Burch-Smith et al., 2006) for knocking out/down gene expression, and the use of viral RNA silencing suppressors to enhance expression of genes of interest (Voinnet et al., 2003). Overall, it appears that virtually every new gene expression technology developed for non-plant systems very quickly finds its application in plant biology via new vector systems. Some recent examples of such vector systems are those that introduce into plant biology the use of the bimolecular florescent complementation assay for protein-protein interaction (Bracha-Drori et al., 2004; Walter et al., 2004; Citovsky et al., 2006), C- and N-terminal protein tagging with various autofluorescent markers (Tzfira et al., 2005; Chakrabarty et al., 2007), CRE/loxP recombination to produce single-copy T-DNA inserts (De Buck et al., 2007), and many others. Besides adapting novel technologies for studies of gene expression and protein interactions, new vector systems are being produced to utilize transgenic technologies in an ever-expanding range of plant species, such as forest trees and transformation-recalcitrant crops (e.g. Meyer et al., 2004; Coutu et al., 2007). Furthermore, vectors for systemic gene expression without permanent genetic modification of the plant are

[1] Guest Editor and Monitoring Editor, *Plant Physiology.*

* Corresponding author; e-mail vitaly.citovsky@stonybrook.edu.

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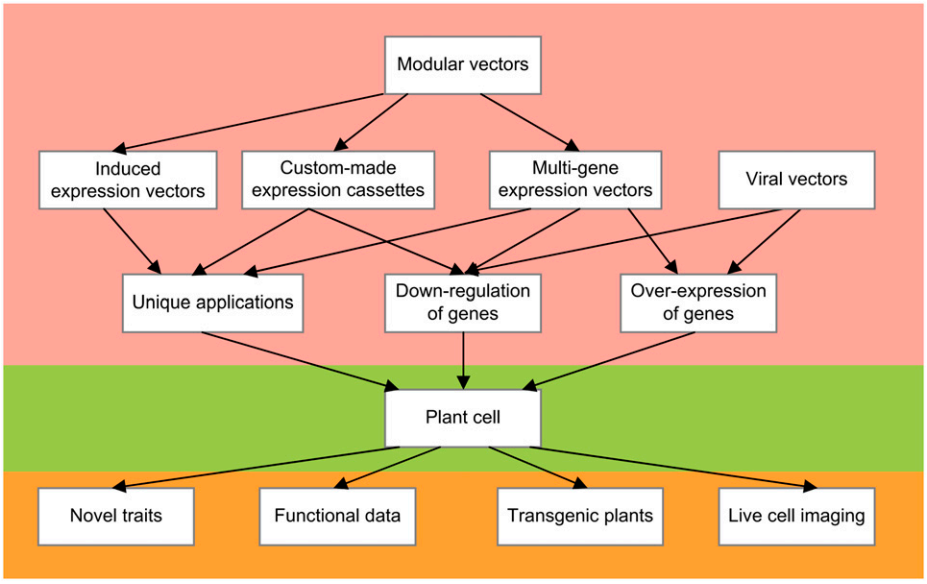

Figure 1. From vectors to applications to cellular functions. Introduction of genetic information into target plant cells and acquisition of new data as a result of transgene expression may require a network of modular vectors, flexible gene cloning and expression systems, and specialized plasmids that result in different modes of transgene expression. Modular vectors may represent a starting point for assembly of custom-made expression vectors, multigene expression vectors, and other types of plant transformation vectors. These vectors in turn provide the users with the abilities to overexpress and down-regulate genes, as well as with the capacity for specific, and often unique, applications, useful for obtaining novel traits and functional data, protein imaging in living plant cells, and generating transgenic plants for plant research and biotechnology.

being developed based on different plant viruses (e.g. Gleba et al., 2005; Marillonnet et al., 2005).

It is difficult to overestimate the effect a truly versatile yet simple-to-use expression vector system can have on many fields of plant research and biotechnology. As more and more vectors and vector systems for gene expression in plants become available, it also becomes essential to make their existence and the scope of their use known to the diverse community of plant researchers, basic and applied. This *Focus Issue* is a small step in this direction. It presents a collection of original articles describing the development of new vector systems useful for plant research and biotechnology, as well as a compilation of short review articles that highlight some of the major developments in vector-assisted plant research technologies. To name a few, the reader will find papers describing an extensive collection of MultiSite Gateway-based plant expression vectors (Karimi et al., 2007), updates on the use of bimolecular florescent complementation for analyses of protein-protein interactions in living plant cells (Ohad et al., 2007) and on the introduction of multiple genes into plant cells (Dafny-Yelin and Tzfira, 2007), a guide to vectors for chloroplast transformation (Lutz et al., 2007), and descriptions of a yeast-plant coupled system for detection of functional nuclear localization signals (Zaltsman et al., 2007), a virus-induced gene silencing system for reverse genetics of floral scent (Spitzer et al., 2007), a system of transformation vectors with the superpromoter (Lee et al., 2007), and a new cloning strategy for recombinase-mediated cassette exchange (Louwerse et al., 2007).

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