Int. J. Dev. Biol. 49: 453-465 (2005)
doi: 10.1387/ijdb.041927ml

Historical perspectives on plant developmental biology

MIEKE VAN LIJSEBETTENS* and MARC VAN MONTAGU

Department of Plant Systems Biology, Flanders Interuniversity Institute for Biotechnology (VIB), Ghent University, Gent, Belgium

The pre-plant developmental biology era

Around 1950, B. McClintock's and P. Peterson's analyses in maize led to the description of mobile DNA in the genome. McClintock correlated chromosome breakpoints at specific positions with mobile DNA elements, called *Dissociator (Ds)* and *Activator (Ac)*, that caused specific changes in phenotypes explained by the altered expression status of known gene loci such as the C locus (McClintock, 1950). This new vision of the genome being dynamic was confirmed in bacteria, animals and other plant species. The molecular basis of mobile DNA in several plant species was demonstrated later (Fedoroff *et al.,* 1983; Döring *et al.,* 1984; Pohlman *et al.,* 1984; Pereira *et al.,* 1985). The cloning of the bronze locus in maize with the Ac transposable element was one of the first examples of «gene tagging» in plants (Fedoroff *et al.,* 1984). Moreover, the *Ac/Ds* and *En/Spm* elements, endogenous to the monocotyledon maize, were shown to be active in the dicotyledonous tobacco (Baker *et al.,* 1986; Masson and Fedoroff, 1989; Pereira and Saedler, 1989). These studies pioneered the use of mobile DNA in large-scale mutagenization programs of the plant genome for gene discovery. Introduction of mobile DNA into heterologous plant genomes required a transformation step that was solved by the study of the tumor-inducing (Ti) principle of the plant-colonizing bacterium, *Agrobacterium tumefaciens*. In 1974, it was demonstrated that a plasmid was present in oncogenic *Agrobacterium* strains and absent from non-oncogenic strains (Zaenen *et al.,* 1974). It resulted in the hypothesis that this plasmid was the Ti principle and it was indeed shown that a fragment of the plasmid, the so-called T-DNA was transferred to the plant genome (Chilton *et al.,* 1977; De Beuckeleer *et al.,* 1978). The T-DNA contained a number of genes, the so-called oncogenes, encoding plant hormone-synthesizing enzymes that were driven by eukaryotic promoters that became active in the plant cell upon infection (Joos *et al.,* 1983; Zambryski *et al.,* 1989). Only the T-DNA borders and the virulence genes on the Ti plasmid were essential for T-DNA transfer and integration into the plant genome. All the T-DNA genes could be replaced by chimeric selectable marker genes or other genes and stably integrated and expressed into the plant genome (Bevan *et al.,* 1983; Fraley *et al.,* 1983; Zambryski *et al.,* 1983; De Block *et al.,* 1984; Horsch *et al.,* 1984). The T-DNA was further engineered as a versatile vector for plant transformation and such vector construction is still ongoing today (Karimi *et al.,* 2002). The plant transformation procedures benefited from earlier research on *in vitro* propagation and regeneration of explants on sterile mineral salt solutions (Murashige and Skoog, 1962) that contained different ratios of phytohormones to promote either callus, root, or shoot growth from explants (Linsmaier and Skoog, 1965). Digestion of explants to single protoplasts and subsequent regeneration into fertile plants was a great advancement because these regenerated plants were clonal (Nagata and Takebe, 1970; Nagy and Maliga, 1976; Lörz *et al.,* 1979). The integration of the

***Address correspondence to:** Dr. Mieke Van Lijsebettens. Department of Plant Systems Biology, VIB2-Universiteit Gent, Technologiepark 927, B-9052 Gent, Belgium. Fax 32-9-331-3809. e-mail: mieke.vanlijsebettens@psb.ugent.be

0214-6282/2005/$25.00

Printed in Spain
www.intjdevbiol.com

protoplast regeneration procedure with *Agrobacterium* infection opened the way to produce clonal transgenic cell lines in tobacco at first (Márton *et al.,* 1979). Some plants appeared to be recalcitrant to *in vitro* regeneration and *Agrobacterium* transformation. It took more than a decade to succeed in a wide variety of plant species. Efficient *Agrobacterium tumefaciens*-mediated transformation methods that were tissue culture-based and accessible to the entire academic community were established for the model plants *Arabidopsis thaliana, Oryza sativa* and *Zea mays* (Valvekens *et al.,* 1988; Hiei *et al.,* 1994; Frame *et al.,* 2002). In the meantime, a number of important technological breakthroughs were made in molecular biology, such as the cloning into plasmid vectors, the determination of the DNA sequence of the first viral organism (Fiers *et al.,* 1978) and gene expression analysis (Kamalay and Goldberg, 1984). The subsequent automatization of the sequencing technology resulted in the whole genome sequence of the first flowering plant, namely that of *Arabidopsis* (Arabidopsis Genome Initiative, 2000). High density micro-arrays allowed for quantitative genome-wide expression analyses and contributed to the systems biology approach of biological questions (Lipshutz *et al.,* 1999). The *in vitro* DNA amplification via the polymerase chain reaction (PCR) revolutionized plant biology because the large genomes were made accessible for experimentation (Mullis and Faloona, 1987). The β-glucuronidase gene of *Escherichia coli* was the first reporter gene adapted for use in plants (Jefferson *et al.,* 1987) and was replaced ten years later by the green fluorescent protein (GFP) from jelly fish because of its

TABLE 1

TIMELINE OF PLANT DEVELOPMENTAL BIOLOGY MILESTONES INTEGRATED WITH RELEVANT TECHNOLOGICAL BREAKTHROUGHS

Year	Discovery	Scientist
1859	Speciation through genetic alteration and natural selection	C. Darwin and A.R. Wallace
1950	Mobile DNA in the maize genome and its mutagenicity	B. McClintock
1952	Regenerative capacity of the potato shoot apex shown by microsurgery	I. Sussex
1955	Dependence of leaf dorsoventrality on communication with the shoot apex in potato	I. Sussex
1965	Organogenetic capacity of phytohormones in tobacco *in vitro* regeneration	E.M. Linsmaier and F. Skoog
1970	Evolution by gene duplication	S. Ohno
1974	Plasmid present in oncogenic agrobacteria	J. Schell and M. Van Montagu
1977	Tumor≠-inducing principle of *Agrobacterium tumefaciens* was a plasmid fragment that integrated into the plant genome	M.D. Chilton, M.P. Gordon and E.W. Nester
1978	Complete sequence of the viral SV40 genome	W. Fiers
1983-1984	Regeneration of the first transgenic plants in tobacco using antibiotic resistance markers and disarmed T-DNA vectors	M. Bevan, M. De Block, M. Van Montagu, J. Schell and R. Horsch
1983-1984	Isolation of *Ac/Ds* elements and sequence determination	P. Starlinger and N. Fedoroff
1986	Maize transposons function in tobacco	B. Baker and N. Fedoroff
1987	DNA synthesis *in vitro* via polymerase chain reaction	K. Mullis and F. Faloona
1987	*E. coli* β glucuronidase used as reporter gene in plants	R.A. Jefferson and M. Bevan
1989-1990	Floral homeotic mutants in *Arabidopsis* and *Antirrhinum* and the ABC model for flower formation	J. Bowman and E. Meyerowitz R. Carpenter and E. Coen
1990	Cloning of the first floral homeotic gene in *Antirrhinum* Cloning of the first floral homeotic gene in *Arabidopsis*	H. Sommer, H. Saedler and S. Scharz-Sommer M. Yanofsky and E. Meyerowitz
1991	Cloning of *KNOTTED* in maize important for indeterminacy in the SAM	E. Vollbrecht and S. Hake
1991	Genetic proof of embryonic axis formation	U. Mayer and G. Jürgens
1993	Genetic control of meristem size: the *CLAVATA* signaling pathway	E. Meyerowitz and S. Clark
1993-1994	Cell patterning in the root epidermis and meristem	L. Dolan and B. Scheres
1995	Trafficking of *KNOTTED1* transcription factor through plasmodesmata	W. Lucas and S. Hake
1995-1997	Cell identity in the root apical meristem dependent on short range signaling as shown by laser cell ablation	P. Weisbeek and B. Scheres
1997	Gene silencing in plants	D. Baulcombe
1997	Use of green fluorescent protein as reporter in plants	J. Haseloff
1999	High-density micro-array for monitoring genome-wide expression	R. Lipchutz and S. Fodor
1999-2000	Conservation of genetic control of leaf initiation	M. Tsiantis, J. Langdale, M. Timmermans, T. Nelson, A. Hudson and M. Byrne
2001	Leaf polarity genes in *Arabidopsis*,	J. Bowman, R. Kerstetter and S. Poethig
2000	Sequence of the *Arabidopsis thaliana* genome	Arabidopsis Genome Initiative
2002	microRNAs in plants	D.J. Bartel, B. Bartel and M.W. Rhoades
2003	Regulation of phyllotaxis by polar auxin transport	D. Reinhardt and C. Kuhlemeier
2003	Sorting of cells by laser capture or flow cytometry	T. Nelson and P. Benfey
2003-2004	Mathematical modeling and computer simulation of patterning	E.Coen and P. Prusinkiewicz

Technological breakthrough in red

application in living explants using confocal microscopy (Haseloff *et al.,* 1997). A timeline is presented in Table 1.

Milestones in plant developmental biology

Milestones during the last 25 years were the cloning of genes corresponding to mutations affecting key steps in developmental processes (forward genetics), their molecular analysis and the study of their genetic interactions in order to build genetic models for a given process (reviewed for *Arabidopsis* by Somerville and Koornneef, 2002; Van Lijsebettens *et al.,* 2002). This molecular-genetic approach is now replaced by a large-scale functional genomics approach in which the function of all members of a gene family is analyzed by reverse genetics. In the following sections, we will exemplify a number of specific cases, with milestones listed in Table 1.

Embryogenesis

Embryogenesis in plants starts with the asymmetric division of the zygote resulting in an upper cell that will develop into an embryo and in a lower cell that generates the suspensor, which is the connection to the maternal tissue. For ease of conceptualization approximately 20 stages have been distinguished in embryo formation of which the early ones are important for axis formation and patterning and the later ones for growth and maturation. Mature plant embryos have a very simple body plan, in which the apical-basal and radial axes are specified. At the end of the apical-basal axis the root and shoot apical meristems (SAM) are situated that become active upon germination and generate the primary root and shoot, respectively. Along the radial axis, patterning in progenitor tissue layers occurs during early embryogenesis. One of the key questions has been the identification of regulators that control the switches from globular to heart stage or from heart to torpedo stage. However, such master switches have not been detected despite extensive mutagenization programs for embryo-lethal *(emb)* mutants in *Arabidopsis* (Meinke and Sussex, 1979a, 1979b; Franzmann *et al.,* 1995; McElver *et al.,* 2001) and in maize and extensive studies of the *Daucus carota* embryogenic cell suspension (Giuliano *et al.,* 1984; De Jong *et al.,* 1992). Currently these structures are assumed to arise progressively. Approximately 750 *EMB* loci have been described to date that are essential for embryogenesis (Franzmann *et al.,* 1995; McElver *et al.,* 2001). Some of these genes are important in the communication between suspensor and embryo, in the control of cell number in the embryos and in the control of embryo maturation. A number of them will reveal essential enzymes for primary metabolism and numerous loci correspond to unknown proteins (Berg *et al.,* 2004). A comprehensive database has been developed containing information on genes that give a seed phenotype upon mutation in *Arabidopsis* (Tzafrir *et al.,* 2003). The conclusion is that the development of the zygote into the embryo is a progressive process in which the action of many genes together is required.

Key regulatory genes that control axis formation have been identified: phenotypes predicted from defects along the apical-basal or the radial axis were obtained upon mutagenization and screening for seedling lethals (Mayer *et al.,* 1991); their corresponding genes are involved in cytokinesis and auxin transport (Shevell *et al.,* 1994; Lukowitz *et al.,* 1996; Hardtke and Berleth, 1998; Assaad *et al.,* 2001). These studies confirmed the model for embryo formation obtained through cell biology and clonal analyses describing the different domains and boundaries within a developing embryo and the embryonal origin of the different parts of the germinating seedling (Dolan *et al.,* 1993; Scheres *et al.,* 1994).

The self-regulatory shoot apical meristem

The SAM has an embryonic origin based on the expression of the *SHOOTMERISTEMLESS (STM)* SAM marker gene and the stm knockout phenotype that produces seedlings without SAM

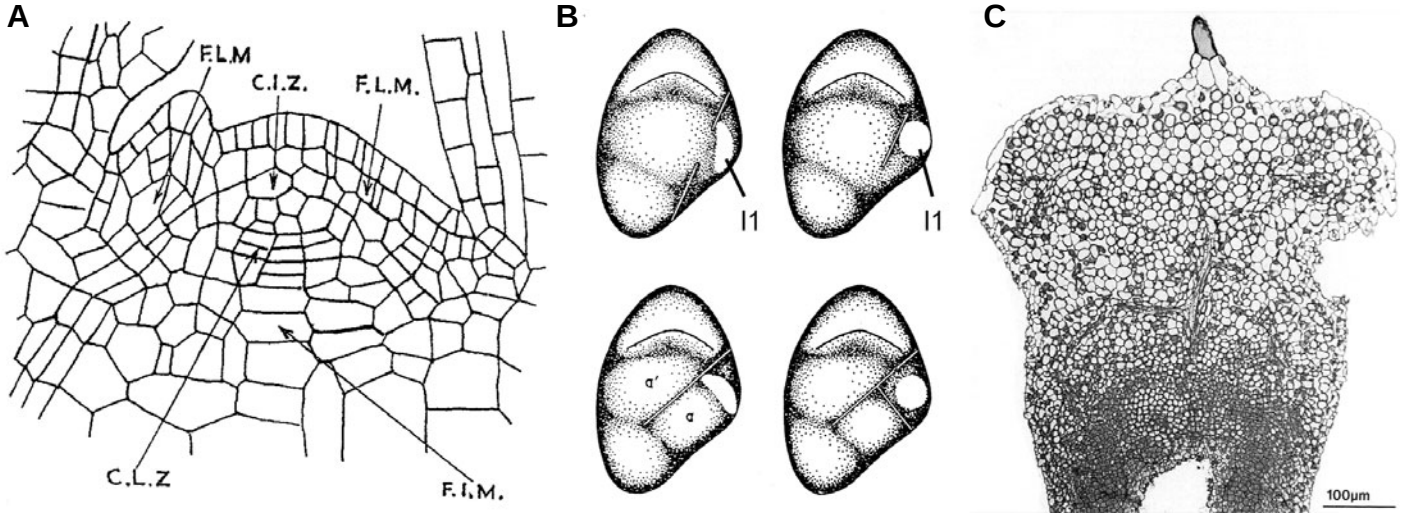


Fig. 1. Shoot apical meristem organization and leaf development. (A) *Drawing of an* Arabidopsis thaliana *median longitudinal histological section of the SAM with the different zones (FLM, flank meristem; CIZ, central initiation zone; CLZ, cambium-like zone; FIM, file meristem) (according to Vaughan, 1952).* **(B)** *Microsurgical incisions, represented by the white lines, in the potato SAM (top view) have an effect on the symmetry of the next leaf primordium (I1) to be formed (according to Sussex, 1955).* **(C)** *Paradermal section through an expanding* Arabidopsis thaliana *leaf lamina showing gradients of cell division at the basal zone and cell expansion at the tip (according to Pyke* et al., *1991).*

(Barton and Poethig, 1993; Long *et al.,* 1996; Long and Barton, 1998). The *STM* gene is the orthologue of the maize KNOTTED1 *(KN1)* homeobox gene that upon ectopic expression in maize leaves reverts the determinate to the indeterminate state, resulting in the production of knots (Vollbrecht *et al.,* 1991). KN1 was the first plant protein for which plasmodesmal trafficking has been shown to occur (Lucas *et al.,* 1995); this report was one of the first to emphasize the importance of plasmodesmal cell-to-cell communication in developmental processes. After germination, the SAM starts to produce the lateral organs and stem tissue that are organized in the so-called phytomers. The SAM consists of zones that are distinct with respect to their cell division activity and developmental destination and was subject of early developmental research in plants and is still today (Vaughan, 1952; Steeves and Sussex, 1989; Potten and Loeffler, 1990; Laufs *et al.,* 1998) (Figure 1A). In the central zone, stem cells stay in an indeterminate state. Upon division, stem cells replenish themselves but also produce daughter cells that are displaced into the peripheral zone where they are recruited to initiate leaf primordia or into the rib zone where they contribute to the formation of stem tissue. The SAM is also layered: L1, L2 and L3 layers that are the progenitor of epidermal tissue, of palisade and spongy parenchyma (and the sporogenic cells) and of vascular tissue, respectively. A genetic model has been proposed for the self-regulation of the SAM. The *CLAVATA (CLV)* genes are responsible for the repression of the growth in the central zone (Clark *et al.,* 1993, 1997; Fletcher *et al.,* 1999; Brand *et al.,* 2000) and encode components of a signaling cascade that regulates *WUSCHEL (WUS)* activity (Laux *et al.,* 1996; Trotochaud *et al.,* 1999; Schoof *et al.,* 2000). WUS is a homeodomain protein that keeps stem cells in their indeterminate state through a negative feedback loop with CLV3 (Mayer *et al.,* 1998). The site of expression of the *WUS* domain, just beneath the stem cell zone is called the «organizing center» and is comparable to the quiescent center in the root apical meristem (van den Berg *et al.,* 1997; for review, see Weigel and Jürgens, 2002).

Leaf phyllotaxis, initiation and polarity

Leaf primordia initiate at the SAM peripheral zone and have a multicellular origin because cells are recruited from the different SAM layers. The leaf initiation site or phyllotaxis is delineated by molecular markers, such as *ASYMMETRIC1*, whose position at the periphery of the SAM depends on the position of previously formed primordia. Phyllotaxis is species specific, can be opposite, decussate, or spiral according to the mathematical Fibonacci series (Mitchison, 1977). «Biophysical forces» regulating local epidermal cell wall extensibility was one of the mechanisms proposed to explain phyllotaxis (Green, 1996; Fleming *et al.,* 1997). Recently mutational analysis and pharmacological tests combined with micro-manipulation have demonstrated that the hormone auxin is crucial in determining the leaf initiation site (Reinhardt *et al.,* 2000, 2003).

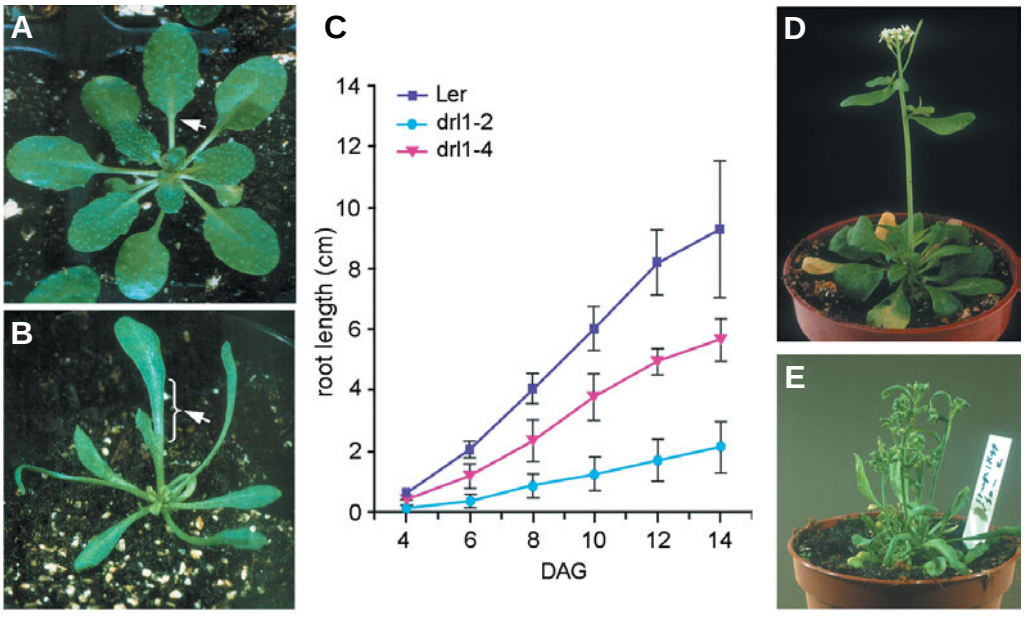
At the leaf inception site, no *STM* expression fits the exit from the proliferative state into a differentiation state of the primordium founder cells with a de-repressed *AS1* gene activity. AS1 in *Arabidopsis* (Byrne *et al.,* 2000), its orthologue *PHANTASTICA (PHAN)* in *Antirrhinum* (Waites and Hudson 1995; Waites *et al.,* 1998) and *ROUGH SHEATH2 (RS2)* in maize (Timmermans *et al.,* 1999; Tsiantis *et al.,* 1999) are important for promoting adaxial fate in leaf primordia, their function being conserved in monocots and dicots. Upon recessive mutation of the *AS1/PHAN/RS2* genes, some of the *KNOX* genes are ectopically expressed in the leaves where they are normally inactive (Schneeberger *et al.,* 1998; Byrne *et al.,* 2000; Ori *et al.,* 2000). Microsurgical experiments on the potato shoot apex have shown that the SAM communicates with leaf primordia and that a signal is required to induce polarity in the leaf primordium. Incisions between the SAM and the primordium resulted in radial symmetrical rather than dorsoventral asymmetrical leaves (Figure 1B) (Sussex, 1951, 1955). Although the signal is still not known today, the genetic factors for polarity have been identified: these are *AS1* and transcription factors of the HD-ZIPIII and GARP class (Sawa *et al.,* 1999; Siegfried *et al.,* 1999; Kerstetter *et al.,* 2001; McConnell *et al.,* 2001). Analysis of their genetic interactions resulted in a model for dorsoventrality in leaves (for review, see Bowman, 2004).

Organ size and shape

A very intriguing question in organ formation is how size and shape are determined. Cell expansion and its direction have been considered for a long time to be the major determinants. However, recent work has demonstrated that cell division activity, rate of cell division and termination of division activity are also important determinants for organ morphology as shown by mutational analysis and manipulation of the cell cycle. Two theories have been postulated: the Cell Theory which states organ size and shape are merely determined by their building blocks, the cells; in the Organismal Theory, cells just fill up the organ form that is determined by higher order control (for review, see Tsukaya, 2002; Beemster *et al.,* 2003).

The leaf has been exploited as a model to study the genetic and environmental factors that control size and shape. Early leaf growth is mainly due to cell division processes that cease gradually from the tip to the base of the organ, from its margin to the midvein and from the ventral to the dorsal side of the lamina (Figure 1C) (Pyke *et al.,* 1991; Van Lijsebettens and Clarke 1998; Donnelly *et al.,* 1999). Interference with early growth by modulation of cell cycle regulatory genes has resulted in changes in leaf size and shape (De Veylder *et al.,* 2001; Fleming, 2002; Wyrzykowska *et al.,* 2002; Dewitte *et al.,* 2003). The *AINTEGUMENTA* transcription factor controls organ size by regulating the number and the extent of cell divisions during organogenesis (Mizukami and Fischer, 2000). Later growth is assumed to be due to polar and non-polar cell expansion processes. Expansion growth is perturbed by modifying the expression of genes coding for enzymes involved in hormone biosynthesis or cell wall composition and results in altered leaf size and shape (Fleming *et al.,* 1997; Kim *et al.,* 1999; Cho and Cosgrove, 2000; Pien *et al.,* 2001; Fleming, 2002). *ANGUSTIFOLIA*, a transcriptional co-repressor, is required for polar cell expansion (Folkers *et al.,* 2002; Kim *et al.,* 2002). Over 100 gene loci have been identified to date with a function in the making of the leaf, of which 94 originate from an ethyl methane sulfonate mutagenization program (Berná *et al.,* 1999; Tsukaya, 2002). The systematic cloning and molecular-genetic analyses of these genes will further our knowledge on the molecular mechanisms directing organ formation.

In addition to the above-mentioned internal factors, leaf growth is also modulated by environmental factors, such as water, light and CO_2 availability, that affect leaf size and shape. These parameters influence the number of cell cycles during leaf formation



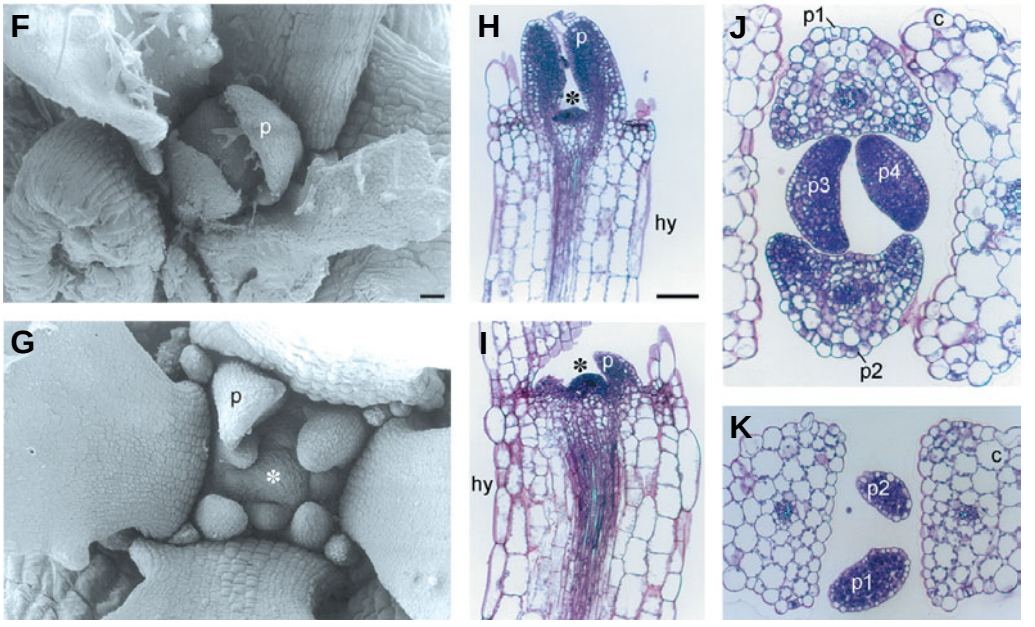


Fig. 2. Developmental phenotypes of the *drl1-2* mutant affected in the *DRL1* gene that is a putative regulator of the plant Elongator histone acetyltransferase complex. (A,B) *Full grown rosettes of wild type, resp.* drl1-2. **(C)** *Primary root growth kinetics.* **(D,E)** *Inflorescence architecture of wild type, resp.* drl1-2. **(F,G)** *Scanning electron micrograph of a wild type, resp.* drl1-2 *SAM.* **(H,I)** *Longitudinal section through a 6-day-old shoot apex of wild type, resp.* drl1-2. **(J,K)** *Transverse section through 12-day-old shoot apices of wild type, resp.* drl1-2 *(according to Nelissen* et al., *2003). Asterisks indicate the SAM. c, cotelydon; DAG, days after germination; hy, hypocotyls; p, leaf primordium; p1 to p4, first to fourth leaf primordium; Bar in F,G = 25 μm; in H to K, 50 μm.*

(Tardieu and Granier, 2000) and the polar and non-polar cell expansion processes that contribute to leaf organogenesis as well (Granier and Tardieu, 1998).

In *Arabidopsis*, mutational analysis showed that leaf growth is controlled at the transcriptional level not only by transcription factors but presumably also by chromatin modification. Functional analysis of structural components and a putative regulator of the Elongator histone acetyltransferase complex, associated with the RNA polymerase II transcription elongation complex resulted in plants with a leaf phenotype (Nelissen *et al.,* 2005), suggesting that the chromatin status is important during organogenesis (Figure 2).

Universal flower model

Flowers consist of four types of organs that are arranged in whorls. The genetic control of flower organ identity was a major discovery of plant developmental biology in the nineties and was based on the study of homeotic flower mutants with normal floral organs at ectopic positions, which replace the flower organs usually present. Such mutants were described in a number of species in ancient literature all over the world. Homeotic mutants and their genetic interactions have been studied extensively in snapdragon (Carpenter and Coen, 1990) and *Arabidopsis* (Komaki *et al.,* 1988; Meyerowitz *et al.,* 1989). This research resulted in the famous ABC flower model (Bowman *et al.,* 1989; Schwarz-Sommer *et al.,* 1990; Coen and Meyerowitz, 1991). In this model, floral organs are specified by the action of A, B and C genes, the so-called floral organ identity genes, that are active in two subsequent whorls. From 1990 on, the genes corresponding to the flower homeotic genes were cloned in *Antirrhinum*, *Arabidopsis* and *Petunia* by making use of the first mutant collections tagged either with endogenous transposable elements (Carpenter and Coen, 1990), with the *Agrobacterium* T-DNA (Azpiroz-Leehan and Feldmann, 1997), with the endogenous transposon *dTph1* in petunia; or using reverse genetics strategies (Vandenbussche *et al.,* 2003a, 2004). The first homeotic genes cloned were the C function gene *AGAMOUS* in *Arabidopsis* (Figure 3B) (Yanofsky *et al.,* 1990) and the B function gene *DEFICIENS* in snapdragon (Sommer *et al.,* 1990; Schwarz-Sommer *et al.,* 1990). In the following years all the floral homeotic genes were cloned and appeared to be MADS-box transcription factors, except for *APETALA2* that identified a plant-specific transcription factor class (Jofuku *et al.,* 1994). The

Fig. 3. Homeotic flower mutants. (A) *Wild-type* Arabidopsis thaliana *flower.* **(B)** Arabidopsis agamous c^- *mutant (according to Yanofsky* et al., *1991).* **(C)** Arabidopsis $a^-b^-c^-$ *triple mutant (according to Weigel and Meyerowitz, 1994).*

ABC model has been verified by double and triple mutants, overexpression constructs and gene expression analyses; it still stands today even though it has been extended with D and E function genes (Colombo *et al.,* 1995; Pelaz *et al.,* 2000; Honma and Goto, 2001). The identity of the floral organs has been postulated to be specified by tetrameric complexes of floral organ identity gene products that bind to promoters of downstream targets thereby activating or repressing their activity and resulting in specific floral organ identities (Honma and Goto, 2001; Theiβen and Saedler, 2001). ABCDE genes were identified in a lot of species of flowering plants, monocots and dicots, confirming the conservation of the overall molecular mechanism of flower organ formation in evolution (for recent review, see Ferrario *et al.,* 2004). All genes, except for A-type function, are present in gymnosperms indicating ancient mechanisms for reproductive organ formation (Tandre *et al.,* 1995; Theissen *et al.,* 2000).

In $a^-b^-c^-$ triple mutants, every flower organ is converted into a leaf and overexpression of B and D function genes is sufficient to transform rosette leaves into petals, which is genetic proof of leaves being the ground state (Figure 3C) (Weigel and Meyerowitz, 1994; Honma and Goto, 2001). von Goethe (1790) had already pointed out that different types of organs, such as leaves, petals and stamens, were variations on a common underlying theme. His theory was also based on the study of abnormal flower morphologies in which one organ type was replaced by another one.

Symmetry

The famous botanist C. Linnaeus (1749) noticed that occasionally individuals of a plant species had altered flower morphologies. He described a naturally occurring mutant of *Linaria vulgaris* (toadflax) with radial flower symmetry instead of dorsoventral asymmetry; the mutant flower was called peloric (Greek for monstruous). Similar mutations have been obtained in *Antirrhinum* from the large transposon-mutagenized population generated at Norwich (Carpenter and Coen, 1990; Coen, 1996). In the mutant flowers, the bilateral symmetry was converted into a radial symmetry with ventral-type of petals and stamens, indicating that a dorsalizing factor had been affected. The dorsalizing factor represented two closely related genes, *CYCLOIDEA (CYC)* and *DICHOTOMA (DICH)* (Luo *et al.,* 1996; Luo *et al.,* 1999), both members of the so-called TCP class of plant-specific DNA-binding proteins (Cubas *et al.,* 1999a). The peloric mutant described by Linnaeus had an epigenetic mutation in a *CYC* orthologue (Cubas *et al.,* 1999b). The *CYC* and *DICH* genes are expressed very early in flower meristems before flower organ initiation. Superimposition of a dorsal domain onto the radial symmetry of the flower meristem is necessary to create dorsoventral asymmetry. Differences in flower morphology between the closely related species *Antirrhinum* and *Mohavea* have been explained by ectopic expression patterns of the *CYC* and *DICH* genes (Hileman *et al.,* 2003), which is one of the first examples that explains evolutionary morphological divergence in terms of variations in gene expression.

The cellular organization in the primary root

The development of the primary root has been neglected for a long time and it was only until *Arabidopsis* started to be a model that people got interested in root biology. Until then, the root was mainly studied for its role in gravitropism. The root is a good model for cell biology because it is transparent (no chlorophyll) and, hence amenable to confocal microscopy on living explants by using fluorescent dyes or reporter genes. Tissue patterning and cellular communication have been studied extensively and with great success in the root. Clonal analyses showed the embryonic origin of the root meristem initials and its radial organization in a constant cell number with root cell initials giving rise to one or two cell layers (Dolan *et al.,* 1993; Scheres *et al.,* 1994). The root meristem consists of quiescent center cells that keep the surrounding initials in an indeterminate state (van den Berg *et al.,* 1997). The daughter cells of the initials differentiate into specific tissue, the identity of which is reinforced by signals from more mature cells (van den Berg *et al.,* 1995). Patterning in the root epidermis was subject to cell biology and genetic analyses (Dolan *et al.,* 1994) and a number of regulatory genes have been identified (Larkin *et al.,* 2003). Mutagenization programs were initiated to look for regulatory genes of root cell specification (Benfey *et al.,* 1993; Scheres *et al.,* 1995). *SCARECROW* and *SHORT ROOT* are essential for the asymmetric cell division in the generation of the cortex/endodermal tissue layers (Di Laurenzio *et al.,* 1996). A huge number of marker lines exist with cell type-specific expression in the root, which have been obtained by promoter trapping with a modified GFP reporter gene (Haseloff *et al.,* 1997). A major breakthrough technology was the use of these marker lines in cell sorting to purify specific cell types and study their transcript profiles (Birnbaum *et al.,* 2003). The technology will become applicable to a large number of cell types or cell domains *in planta* that can be distinguished by marker genes. Cell differentiation studies were restricted in plant research because *in vitro* culture of a specific cell type has not been achieved so far.

This restriction has been alleviated by the above-mentioned approach.

Perspectives in plant developmental biology

Although the survey of milestones is far from complete it is obvious that in the past 15 years a lot of progress has been made in the identification of the genetic control of pattern formation during embryogenesis, organ (leaf and flower) formation and in tissue differentiation. Many of the transcription factors involved have been cloned and studied, however much less effort was investigated so far in the study of the upstream signaling cascades and intrinsic and external stimuli that direct these patterning processes through transcription factor activation or repression (Hay *et al.,* 2004). It will be a future challenge to link the genetics to the physiology of the plant.

A lot of research needs to be done on the communication processes between the cells of the multicellular plant as well as between its different tissues and organs. Hormones have been shown to be important to direct developmental processes at the whole organism level, but the molecular mechanism of their circulation through the plant is still poorly understood. In addition other signaling molecules have been recognized as important communicators such as small peptides, oligosaccharides and metabolites such as salicylic acid. The peptides appear to have crucial functions in tissue domain interaction such as CLV3 in the regulatory loop for self-maintenance of the SAM and in cell-cell interaction such as SCR in the self-incompatibility response (Matsubayashi, 2003). A lot of small open reading frames are out there in the genome and their function remains to be solved. The role of volatiles such as jasmonic acid in plant development needs to be further explored. The regulation of plasmodesmata formation and closure between cells and tissue domains has been shown to be important in communication and needs further attention.

Another big question to be solved is how organ size and shape are determined. Over the last years it became clear that not only cell expansion but also cell division is important. At some point in development cells in meristems need to know when to leave the cell cycle and start the differentiation process. The signals and molecular mechanisms need to be determined that control the switch between cell cycle entry and exit during development and in response to environmental cues (Gutierrez, 2005).

It took a decade to functionally analyze 10% of the *Arabidopsis* genes using forward genetics. In the meantime large mutagenized seed collections have been generated that are exploited for reverse genetics of gene families. Within the next five years of *Arabidopsis* research the aim is to uncover the function of every gene; the National Science Foundation 2010 project is the leading initiative. This will be possible because there is a shift to large-scale experimentation in which not a single gene but rather its whole gene family is functionally analyzed. From the genome sequence, all the members of a given gene family can be retrieved; by reverse genetics, mutations can be looked for in the available collections and be analyzed for their phenotype. The function of large gene families such as the cellulose synthase-like genes (Bonetta *et al.,* 2002) or myb-type transcription factors (Meissner *et al.,* 1999) are analyzed by reverse genetics. In large gene families functions might be redundant because of recent gene duplication resulting in the lack of phenotypes by single gene knockout. In order to define functions, double or even triple mutant combinations of knockouts will have to be made in the respective paralogs. A few nice examples illustrate this approach, such as for the MADS-box *SEPALLATA* genes (Pelaz *et al.,* 2000) and the B-function genes in petunia (Vandenbussche *et al.,* 2004). Unknown proteins for which a mutant phenotype has been obtained are analyzed for their interactions with other proteins by means of yeast-two-hybrid analysis or TAP tagging to get a clue to their molecular function. A number of unknown proteins identified by embryo-lethals are studied in this way (Berg *et al.,* 2004). The wealth of information on gene function in model systems will serve to improve plant product quality and adaptation of plants to changing environments. Genes from model systems have been overexpressed in other species with success (Weigel and Nilsson, 1995); however, they mainly serve to isolate and study the orthologs in crops (Byzova *et al.,* 2004). The synteny of large chromosomal domains between related species has been exploited to use gene knowledge obtained in model systems such as *Arabidopsis* for molecular breeding in related crops such as Brassica species (Lagercrantz *et al.,* 1996). Synteny between cereal genomes is high and the rice genome sequence is used as reference to aid for instance in positional cloning of genes in maize (Devos, 2005). Quantitative trait loci analysis is an approach to identify and clone genes that contribute to complex phenotypes such as seed weight or leaf size and shape and has been successfully used in a number of plant species (Alonso-Blanco and Koornneef, 2000; Pérez-Pérez *et al.,* 2002; Morgante and Salamini, 2003; Tanksley, 2004).

More model species for developmental studies are emerging such as *Medicago truncatula* to study nodule formation upon symbiosis with Sinorhizobium (Cook, 1999; Young *et al.,* 2003) and the tree model *Populus* to study wood formation and, more recently, cambium activity (Bhalerao *et al.,* 2003; Brunner *et al.,* 2004). These model systems fulfill a number of criteria such as diploidy, easy transformation, small genome, ongoing genome sequencing, big consortia for coordinated international research and maintenance and availability of genetic resources. In this new research tendency the diversity in plant developmental processes is recognized to exceed the potential of just a few model systems.

Soon there will be a shift from the model species to a wide range of species to study species-specific development or morphologies as for instance the «cluster» root (Shane *et al.,* 2004) and to study processes for which *Arabidopsis* is not a good model such as for domestication, mycorrhizae interaction or nodule formation. With the increasing functional analyses of genes from model species, comparative analysis will become more important and powerful. DNA sequencing technology is automated and its efficiency has improved tremendously over the last five years, so that not the amount of work but rather the cost and bioinformatics tools will be the limiting factors for sequence analysis of a specific species in the near future (Venter, 2004). New areas of research, such as comparative genetics, will exploit this sequence information and couple it to questions related to gene function conservation or divergence. A well-studied case is the homeobox gene function divergence between plants and animals (Meyerowitz, 2002). The conclusion is that similar processes of pattern formation are used in plant and animal developmental programs; however different classes of regulatory genes have been re-

cruited for it during evolution. Comparative genetics relies on DNA sequence information and aims at studying a genetic trait within a plant family or even between incompatible species and overcomes the genetic barrier of crossing inhibition.

Another emerging field is evolutionary developmental biology the so-called "Evo-Devo" that also exploits DNA sequence information to explain morphological diversity. Function conservation of key regulators in development, such as the MADS-box transcription factors with a role in flower organ specification, begins to explain the main aspects of flower morphology in different species, such as the different floral organ types and the floral whorls. Gene duplication and function divergence by coding sequence changes in addition to ectopic expression patterns clarify the diversity in flower morphology in a number of cases (Kramer and Irish, 1999; Vandenbussche *et al.,* 2003b; Ferrario *et al.,* 2004). Evolutionary developmental biology studies have investigated some aspects of diversity in leaf morphology as well (Cubas *et al.,* 1999b; Bharathan *et al.,* 2002; Kanno *et al.,* 2003; Hileman *et al.,* 2003; Tsiantis and Hay, 2003). Bioinformatics research showed that diploid genomes, such as that of *Arabidopsis* and other model systems contain large genome duplications (Arabidopsis Genome Initiative, 2000; Simillon *et al.,* 2002; Blanc and Wolfe, 2004). Genome duplications have been postulated to allow for diversification in gene function and to be the major mechanism to achieve morphological diversity in the flowering plants and also in the animal kingdom in combination with natural selection as postulated by C. Darwin in the late 19th century (Darwin, 1859; Ohno, 1970). Computational approaches to unveil ancient genome duplications are under development and may contribute to new insights into evolutionary genetics (Van de Peer, 2004).

Significant progress in the unraveling of molecular networks is to be expected from the systems biology approach in which the entire transcriptome, proteome, or metabolome is analyzed upon perturbation rather than single genes. The aim is to identify the complex networks responsible for biological processes and their mutual interactions (Gutiérrez *et al.,* 2005). Integration with computational science and mathematics will be indispensable to interpret the large data sets, generate network visualization and build models. The number of computer programs for visualization and integration of different data sets, such as MAPMAN (Thimm *et al.,* 2004) is increasing and is a prerequisite to understand the biology. The integration of biological data into regulatory networks will allow further testing and predictions (Ideker *et al.,* 2001; Davidson *et al.,* 2002). Models on plant growth and development are being generated for plant architecture, organs and tissues and incorporating genetic regulatory networks. These models are an integration of mathematical modeling and computer simulations with biological components such as modules for architecture, growth parameters for organs and tissues, or genes and their domains of action and genetic interaction for regulatory networks (Rolland-Lagan *et al.,* 2003; Kwiatkowska and Dumais, 2003; Gielis, 2003; Prusinkiewicz, 2004). Future goals are the integration of models for architecture with those for organs and tissues and for genetic regulatory networks in order to obtain in-depth understanding of the mechanisms of plant development from genes to phenotypes (Prusinkiewicz, 2004).

Plant cell sorting has recently been achieved by several groups to purify living cells of the same type with cell-specific GFP markers, or alternatively small tissue domains with laser technology (Kerk *et al.,* 2003; Birnbaum *et al.,* 2003). Plant cell sorting is a breakthrough technology since research on cell differentiation in plants was limited to molecular-genetic analysis because of the inability to culture differentiated plant cells *in vitro* unlike in mammalian systems. The genome-wide profiling techniques are applied to this sorted plant material and the results will undoubtedly further our knowledge on the progressive process of cell specification to differentiation and cell function. Increasing the resolution of sorting and systems biology up to single cell level will open up new opportunities in the study of cell specification. Then, genetic programs would be analyzed that distinguish, for instance, between the different fate of the daughter cells after asymmetric cell division, such as in the case of the first zygotic division or upon lateral root induction or in stomatal development.

Another challenge for future research on plant development will be to understand other mechanisms besides the transcriptional control of genetic programs exerted by transcription factors. Recently, microRNAs have been discovered in plants and a number of them are complementary to transcription factors with a function in developmental processes (Reinhart *et al.,* 2002; Rhoades *et al.,* 2002; Bonnet *et al.,* 2004). For instance in leaf development several transcription factors, such as *PHABULOSA, PHAVOLUTA* and *CINCINNATA* -like genes are targeted by miRNAs (Nath *et al.,* 2003; Palatnik *et al.,* 2003; Juarez *et al.,* 2004; Kidner and Martienssen 2004). Temporal and spatial regulation of expression of miRNAs is of utmost importance for the proper destruction of transcription factor mRNAs during developmental processes and it is based on the silencing pathway (Baulcombe, 2004). However, the regulation of expression of the miRNAs is still unknown and needs to be explored because it adds another level to gene expression regulation and it may contribute to the delineation of boundaries and domains in developing organisms.

Protein degradation through the ubiquitination pathway is an important control mechanism for developmental pathways. E3 ubiquitin ligases target specific substrates for degradation at the proteasome and more than 460 are represented in the *Arabidopsis* genome (Stone *et al.,* 2005). A number of their targets will be important in developmental control and their nature will be revealed in the coming years by functional genomics.

Epigenetic control of developmental transitions and morphogenetic processes needs to be further explored (Reyes *et al.,* 2002). The naturally occurring peloric mutant of *Linaria vulgaris* described by Linnaeus more than 250 years ago has an epigenetic mutation in a CYC ortholog (Linneaus, 1749; Gustafsson, 1979; Cubas *et al.,* 1999b). Other well-studied epimutations are at the P locus in maize and at the *SUPERMAN* locus in *Arabidopsis* (Das and Messing, 1994; Jacobsen and Meyerowitz, 1997). From these studies it became clear that the DNA methylation status has a great impact on gene expression and can be transferred to subsequent generations in plants. As mentioned, the *DRL1* and *ELO* genes studied in our unit (Nelissen *et al.,* 2003, 2005) identified a histone acetyltransferase complex, named Elongator that associates with the RNA polymerase II transcription elongation complex. The *drl1-2* and *elo* mutants have a narrow leaf phenotype indicating that leaf form is also regulated by reversible chromatin modification. The *drl1-2* and *elo* mutants have in addition reduced root growth, a stunted inflorescence and an altered phyllotaxis (Figure 2). Reversible histone modifications,

such as acetylation/deacetylation, are of critical importance to make DNA available for transcription or to repress transcription. A well-studied case is the vernalization-dependent deacetylation and, hence, inactivation of the *FLOWERING LOCUS C* gene that codes for a repressor of flowering (Sung and Amasino, 2004). A number of histone acetylases and deacetylases (HDAC) are present in the plant genome, amongst them the plant-specific HD2 subfamily of HDACs (Lusser *et al.,* 1997; Pandey *et al.,* 2002). It will be a challenge to find out about their upstream signaling, downstream targets and function in plant development.

On the longer term, more than 10 years from now, it is difficult to predict the future because a major input of technology and expertise from other disciplines in biological research is to be expected. The biology-driven research relies to a great extent on breakthrough technologies to take the research to the next level. No doubt, technology will have a huge impact on experimentation and thinking in biological research in the next decade.

Summary

The early studies of plant growth and development focused on embryogenesis. In the past twenty five years, it became possible to successfully analyze many more developmental processes, hence plant developmental biology became the generally accepted terminology. It refers to a multidisciplinary approach using expertise and tools from genetics, molecular biology and cell biology to study processes in development also beyond the formation of the embryo. Around that time, initiatives were taken to address biological questions in just a few model systems, such as *Arabidopsis thaliana*, *Zea mays*, *Antirrhinum majus* and *Petunia hybrida*, while the «old» model systems, i.e. potato, tobacco, used in regeneration and grafting experiments, were increasingly abandoned. International research programs were initiated in *Arabidopsis* at first to create stock centers and databases to proceed faster with the scientific research and to get deeper insight into plant biology. During the last five years the maize community made tremendous progress in developing tools and resources for their system. Milestones in plant developmental biology discussed relate to the molecular-genetic approach to study embryogenesis, autoregulation of meristems, leaf and flower initiation, leaf and flower formation and cell specification in the root. Developmental biology changed the research from descriptive to causal resulting in a number of genetic models. Future developments in research will focus on the study of a specific gene activity in a genome-wide context. The building of molecular networks will allow computer modeling of biological processes and its use for predictions and further experimentation. Sequence information derived from the multiple genome projects will be exploited in comparative biology.

KEY WORDS: *model plant, regulatory network, forward and reverse genetics, evo-devo, systems biology and modeling*

Acknowledgements

We thank Marc De Block and Tom Gerats for critical reading of the manuscript and helpful suggestions and Martine De Cock and Karel Spruyt for help in preparing it. This work was supported in part by grants from the Interuniversity Poles of Attraction-Belgian Science Policy (P5/13) and the European Community's Human Potential Program under contract HPRN-CT-2002-00267 (DAGOLIGN).

References

ALONSO-BLANCO, C. and KOORNNEEF, M. (2000). Naturally occurring variation in Arabidopsis: an underexploited resource for plant genetics. *Trends Plant Sci.* 5, 22-29.

The ARABIDOPSIS GENOME INITIATIVE (2000). Analysis of the genome sequence of the flowering plant *Arabidopsis thaliana*. *Nature* 408: 796-815.

ASSAAD, D.D., HUET, Y., MAYER, U. and JÜRGENS, G. (2001). The cytokinesis gene *KEULE* encodes a Sec1 protein that binds the syntaxin KNOLLE. *J. Cell Biol.* 152: 531-543.

AZPIROZ-LEEHAN, R. and FELDMANN, K.A. (1997). T-DNA insertion mutagenesis in *Arabidopsis* : going back and forth. *Trends Genet.* 13: 152-156.

BAKER, B., SCHELL, J., LÖRZ, H. and FEDOROFF, N. (1986). Transposition of the maize controlling element «Activator» in tobacco. *Proc. Natl. Acad. Sci. USA* 83: 4844-4848.

BARTON, M.K. and POETHIG, R.S. (1993). Formation of the shoot apical meristem in *Arabidopsis thaliana:* an analysis of development in the wild type and in the *shoot meristemless* mutant. *Development* 119: 823-831.

BAULCOMBE, D. (2004). RNAi silencing in plants. *Nature* 431: 356-363.

BEEMSTER, G.T.S., FIORANI, F. and INZÉ, D. (2003). Cell cycle: the key to plant growth control? *Trends Plant Sci.* 8: 154-158.

BENFEY, P.N., LINSTEAD, P.J., ROBERTS, K., SCHIEFELBEIN, J.W., HAUSER, M.-T. and AESCHBACHER, R.A. (1993). Root development in *Arabidopsis:* four mutants with dramatically altered root morphogenesis. *Development* 119: 57-70.

BERG, M., ROGERS, R. and MEINKE, D. (2004). *Functional analysis of EMB genes using epitope-tagged proteins.* Abstract presented at the 15th International Conference on Arabidopsis Research, July 11-14, 2004, Berlin (Germany), T10-042.

BERNÁ, G., ROBLES, P. and MICOL, J.L. (1999). A mutational analysis of leaf morphogenesis in *Arabidopsis thaliana. Genetics* 152: 729-742.

BEVAN, M.W., FLAVELL, R.B. and CHILTON, M.-D. (1983). A chimaeric antibiotic resistance gene as a selectable marker for plant cell transformation. *Nature* 304: 184-187.

BHALERAO, R., NILSSON, O. and SANDBERG, G. (2003). Out of the woods: forest biotechnology enters the genomic era. *Curr. Opin. Biotechnol.* 14: 206-213.

BHARATHAN, G., GOLIBER, T.E., MOORE, C., KESSLER, S., PHAM, T. and SINHA, N.R. (2002). Homologies in leaf form inferred from *KNOXI* gene expression during development. *Science* 296: 1858-1860.

BIRNBAUM, K., SHASHA, D.E., WANG, J.Y., JUNG, J.W., LAMBERT, G.M., GALBRAITH, D.W. and BENFEY, P.N. (2003). A gene expression map of the *Arabidopsis* root. *Science* 302: 1956-1960.

BLANC, G. and WOLFE, K.H. (2004). Widespread paleopolyploidy in model plant species inferred from age distributions of duplicate genes. *Plant Cell* 16: 1667-1678.

BONETTA, D.T., FACETTE, M., RAAB, T.K. and SOMERVILLE, C.R. (2002). Genetic dissection of plant cell-wall biosynthesis. *Biochem. Soc. Trans.* 30: 298-301.

BONNET, E., WUYTS, J., ROUZÉ, P. and VAN DE PEER, Y. (2004). Detection of 91 conserved plant microRNAs in *Arabidopsis thaliana* and *Oryza sativa* identifies important new target genes. *Proc. Natl. Acad. Sci. USA* 101: 11511-11516.

BOWMAN, J.L. (2004). Establishment of polarity in lateral organs of seed plants. In *Polarity in Plants*, K. Lindsey (Ed.). Oxford, Blackwell Publishing, pp. 288-316.

BOWMAN, J.L., SMYTH, D.R. and MEYEROWITZ, E.M. (1989). Genes directing flower development in *Arabidopsis*. *Plant Cell* 1: 37-52.

BRAND, U., FLETCHER, J.C., HOBE, M., MEYEROWITZ, E.M. and SIMON, R. (2000). Dependence of stem cell fate in *Arabidopsis* on a feedback loop regulated by *CLV3* activity. *Science* 289: 617-619.

BRUNNER, A.M., BUSOV, V.B. and STRAUSS, S.H. (2004). Poplar genome sequence: functional genomics in an ecologically dominant plant species. *Trends Plant Sci.* 9: 49-56.

BYRNE, M.E., BARLEY, R., CURTIS, M., ARROYO, J.M., DUNHAM, M., HUDSON, A. and MARTIENSSEN, R.A. (2000). Asymmetric leaves1 mediates leaf

patterning and stem cell function in *Arabidopsis*. *Nature* 408: 967-971.

BYZOVA, M., VERDUYN, C., DE BROUWER, D. and DE BLOCK, M. (2004). Transforming petals into sepaloid organs in *Arabidopsis* and oilseed rape: implementation of the hairpin RNA-mediated gene silencing technology in an organ-specific manner. *Planta* 218: 379-387.

CARPENTER, R. and COEN, E.S. (1990). Floral homeotic mutations produced by transposon mutagenesis in *Antirrhinum majus*. *Genes Dev.* 4: 1483-1493.

CHILTON, M.-D., DRUMMOND, M.H., MERLO, D.J., SCIAKY, D., MONTOYA, A.L., GORDON, M.P. and NESTER, E.W. (1977). Stable incorporation of plasmid DNA into higher plant cells: the molecular basis of crown gall tumorigenesis. *Cell* 11: 263-271.

CHO, H.-T. and COSGROVE, D.J. (2000). Altered expression of expansin modulates leaf growth and pedicel abscission in *Arabidopsis thaliana*. *Proc. Natl. Acad. Sci. USA* 97: 9783-9788.

CLARK, S.E., RUNNING, M.P. and MEYEROWITZ, E.M. (1993). *CLAVATA1*, a regulator of meristem and flower development in *Arabidopsis*. *Development* 119: 397-418.

CLARK, S.E., WILLIAMS, R.W. and MEYEROWITZ, E.M. (1997). The *CLAVATA1* gene encodes a putative receptor kinase that controls shoot and floral meristem size in *Arabidopsis*. *Cell* 89: 575-585.

COEN, E. (1996). Floral symmetry. *EMBO J.* 15: 6777-6788.

COEN, E.S. and MEYEROWITZ, E.M. (1991). The war of the whorls: genetic interactions controlling flower development. *Nature* 353: 31-37.

COLOMBO, L., FRANKEN, J., KOETJE, E., VAN WENT, J., DONS, H.J.M., ANGENENT, G.C. and VAN TUNEN, A.J. (1995). The petunia MADS box gene *FBP11* determines ovule identity. *Plant Cell* 7: 1859-1868.

COOK, D.R. (1999). *Medicago truncatula* - a model in the making! *Curr. Opin. Plant Biol.* 2: 301-304.

CUBAS, P., LAUTER, N., DOEBLEY, J. and COEN, E. (1999a). The TCP domain: a motif found in proteins regulating plant growth and development. *Plant J.* 18: 215-222.

CUBAS, P., VINCENT, C. and COEN, E. (1999b). An epigenetic mutation responsible for natural variation in floral symmetry. *Nature* 401: 157-160.

DARWIN, C. (1859). *On the origin of species by means of natural selection, or the preservation of favoured races in the struggle for life.* John Murray, London.

DAS, O.P. and MESSING, J. (1994). Variegated phenotype and developmental methylation changes of a maize allele originating from epimutation. *Genetics* 136: 1121-1141.

DAVIDSON, E.H., RAST, J.R., OLIVERI, P., RANSICK, A., CALESTANI, C., YUH, C.-H., MINOKAWA, T., AMORE, G., HINMAN, V., ARENAS-MENA, C., OTIM, O., BROWN, C.T., LIVI, C.B., LEE, P.Y., REVILLA, R., RUST, A.G., PAN, Z.J., SCHILSTRA, M.J., CLARKE, P.J.C., ARNONE, M.I., ROWEN, L., CAMERON, R.A., McCLAY, D.R., HOOD, L. and BOLOURI, H. (2002). A genomic regulatory network for development. *Science* 295: 1669-1678.

DE BEUCKELEER, M., DE BLOCK, M., DE GREVE, H., DEPICKER, A., DE VOS, G., DE VOS, R., DE WILDE, M., DHAESE, P., DOBBELAERE, M., ENGLER, G., GENETELLO, C., HERNALSTEENS, J.P., HOLSTERS, M., JACOBS, A., MESSENS, E., SCHELL, J., SEURINCK, J., SILVA, A., VAN HAUTE, E., VAN MONTAGU, M., VAN VLIET, F., VILLAROEL, R. and ZAENEN, I. (1978). The use of the Ti plasmid as a vector for the introduction of foreign DNA into plants. In *Proceedings of the 4th International Conference on Plant Pathogenic Bacteria,* M. Ridé (Ed.). Angers, Institut National de la Recherche Agronomique, pp. 115-126.

DE BLOCK, M., HERRERA-ESTRELLA, L., VAN MONTAGU, M., SCHELL, J. and ZAMBRYSKI, P. (1984). Expression of foreign genes in regenerated plants and their progeny. *EMBO J.* 3: 1681-1689.

DE JONG, A.J., CORDEWENER, J., LO SCHIAVO, F., TERZI, M., VANDEKERCKHOVE, J., VAN KAMMEN, A. and DE VRIES, S.C. (1992). A carrot somatic embryo mutant is rescued by chitinase. *Plant Cell* 4: 425-433.

DE VEYLDER, L., BEECKMAN, T., BEEMSTER, G.T.S., KROLS, L., TERRAS, F., LANDRIEU, I., VAN DER SCHUEREN, E., MAES, S., NAUDTS, M. and INZÉ, D. (2001). Functional analysis of cyclin-dependent kinase inhibitors of Arabidopsis. *Plant Cell* 13, 1653-1667.

DEVOS, K.M. (2005). Updating the "crop circle". *Curr. Opin. Plant Biol.* 8, 155-162.

DEWITTE, W., RIOU-KHAMLICHI, C., SCOFIELD, S., HEALY, J.M.S., JACQMARD, A., KILBY, N.J. and MURRAY, J.A.H. (2003). Altered cell cycle distribution, hyperplasia and inhibited differentiation in Arabidopsis caused by the D-type cyclin CYCD3. *Plant Cell* 15: 79-92.

DI LAURENZIO, L., WYSOCKA-DILLER, J., MALAMY, J.E., PYSH, L., HELARIUTTA, Y., FRESHOUR, G., HAHN, M.G., FELDMANN, K.A. and BENFEY, P.N. (1996). The *SCARECROW* gene regulates an asymmetric cell division that is essential for generating the radial organization of the Arabidopsis root. *Cell* 86: 423-433.

DOLAN, L., JANMAAT, K., WILLEMSEN, V., LINSTEAD, P., POETHIG, S., ROBERTS, K. and SCHERES, B. (1993). Cellular organisation of the *Arabidopsis thaliana* root. *Development* 119: 71-84.

DOLAN, L., DUCKETT, C.M., GRIERSON, C., LINSTEAD, P., SCHNEIDER, K., LAWSON, E., DEAN, C., POETHIG, S. and ROBERTS, K. (1994). Clonal relationship and cell patterning in the root epidermis of *Arabidopsis*. *Development* 120: 2465-2474.

DONNELLY, P.M., BONETTA, D., TSUKAYA, H., DENGLER, R.E. and DENGLER, N.G. (1999). Cell cycling and cell enlargement in developing leaves of *Arabidopsis*. *Dev. Biol.* 215: 407-419.

DÖRING, H.-P., TILLMAN, E. and STARLINGER, P. (1984). DNA sequence of maize transposable element Dissociation. *Nature* 307: 127-130.

FEDOROFF, N., WESSLER, S. and SHURE, M. (1983). Isolation of the transposable maize controlling elements *Ac* and *Ds*. *Cell* 35: 235-242.

FEDOROFF, N.V., FURTEK, D.B. and NELSON, O.E. JR (1984). Cloning of the bronze locus in maize by a simple and generalizable procedure using the transposable controlling element Activator *(Ac). Proc. Natl. Acad. Sci. USA* 81: 3825-3829.

FERRARIO, S., IMMINK, R.G.H. and ANGENENT, G.C. (2004). Conservation and diversity in flower land. *Curr. Opin. Plant Biol.* 7: 84-91.

FIERS, W., CONTRERAS, R., HAEGEMAN, G., ROGIERS, R., VAN DE VOORDE, A., VAN HEUVERSWYN, H., VAN HERREWEGHE, J., VOLCKAERT, G. and YSEBAERT, M. (1978). Complete nucleotide sequence of SV40 DNA. *Nature* 273: 113-120.

FLEMING, A.J. (2002). The mechanism of leaf morphogenesis. *Planta* 216: 17-22.

FLEMING, A.J., McQUEEN-MASON, S., MANDEL, T. and KUHLEMEIER, C. (1997). Induction of leaf primordia by the cell wall protein expansin. *Science* 276: 1415-1418.

FLETCHER, J.C., BRAND, U., RUNNING, M.P., SIMON, R. and MEYEROWITZ, E.M. (1999). Signaling of cell fate decisions by *CLAVATA3* in *Arabidopsis* shoot systems. *Science* 283: 1911-1914.

FOLKERS, U., KIRIK, V., SCHÖBINGER, U., FALK, S., KRISHNAKUMAR, S., POLLOCK, M.A., OPPENHEIMER, D.G., DAY, I., REDDY, A.R., JÜRGENS, G. and HÜLSKAMP, M. (2002). The cell morphogenesis gene *ANGUSTIFOLIA* encodes a CtBP/BARS-like protein and is involved in the control of the microtubule cytoskeleton. *EMBO J.* 21: 1280-1288.

FRALEY, R.T., ROGERS, S.G., HORSCH, R.B., SANDERS, P.R., FLICK, J.S., ADAMS, S.P., BITTNER, M.L., BRAND, L.A., FINK, C.L., FRY, J.S., GALLUPPI, G.R., GOLDBERG, S.B., HOFFMANN, N.L. and WOO, S.C. (1983). Expression of bacterial genes in plant cells. *Proc. Natl. Acad. Sci. USA* 80: 4803-4807.

FRAME, B.R., SJOU, H., CHIKWAMBA, R.K., ZHANG, Z., XIANG, C., FONGER, T.M., PEGG, S.E.K., LI, B., NETTLETON, D.S., PEI, D. and WANG, K. (2002). *Agrobacterium tumefaciens* -mediated transformation of maize embryos using a standard binary vector system. *Plant Physiol.* 129: 13-22.

FRANZMANN, L.H., YOON, E.S. and MEINKE, D.W. (1995). Saturating the genetic map of *Arabidopsis thaliana* with embryonic mutations. *Plant J.* 7: 341-350.

GIELIS, J. (2003). A generic geometric transformation that unifies a wide range of natural and abstract shapes. *Am. J. Bot.* 90: 333-338.

GIULIANO, G., LO SCHIAVO, F. and TERZI, M. (1984). Isolation and developmental characterization of temperature-sensitive carrot cell variants. *Theor. Appl. Genet.* 67: 179-183.

GRANIER, C. and TARDIEU, F. (1998). Spatial and temporal analyses of expansion and cell cycle in sunflower leaves. A common pattern of development for all zones of a leaf and different leaves of a plant. *Plant Physiol.* 116: 991-1001.

GREEN, P.B. (1996). Expression of form and pattern in plants - a role for biophysical fields. *Semin. Cell Dev. Biol.* 7: 903-911.

GUSTAFSSON, A. (1979). Linnaeus' peloria: the history of a monster. *Theor. Appl.*

Genet. 54: 241-248.

GUTIERREZ, C. (2005). Coupling cell proliferation and development in plants. *Nat. Cell Biol.* 7: 535-541.

GUTIERREZ, R.A., SHASHA, D.E. and CORUZZI, C.M. (2005). Systems biology for the virtual plant. *Plant Physiol.* 138: 550-554.

HARDTKE, C.S. and BERLETH, T. (1998). The *Arabidopsis* gene *MONOPTEROS* encodes a transcription factor mediating embryo axis formation and vascular development. *EMBO J.* 17: 1405-1411.

HASELOFF, J., SIEMERING, K.R., PRASHER, D.C. and HODGE, S. (1997). Removal of a cryptic intron and subcellular localization of green fluorescent protein are required to mark transgenic *Arabidopsis* plants brightly. *Proc. Natl. Acad. Sci. USA* 94: 2122-2127.

HAY, A., CRAFT, J. and TSIANTIS, M. (2004). Plant hormones and homeoboxes: bridging the gap? *BioEssays* 26: 395-404.

HIEI, Y., KOMARI, O.S. and KUMASHIRO, T. (1994). Efficient transformation of rice (*Oryza sativa* L.) mediated by *Agrobacterium* and sequence analysis of the boundaries of the T-DNA. *Plant J.* 6: 271-282.

HILEMAN, L.C., KRAMER, E.M. and BAUM, D.A. (2003). Differential regulation of symmetry genes and the evolution of floral morphologies. *Proc. Natl. Acad. Sci. USA* 100: 12814-12819.

HONMA, T. and GOTO, K. (2001). Complexes of MADS-box proteins are sufficent to convert leaves into floral organs. *Nature* 409: 525-529.

HORSCH, R.B., FRALEY, R.T., ROGERS, S.G., SANDERS, P.R., LLOYD, A. and HOFFMANN, N. (1984). Inheritance of functional foreign genes in plants. *Science* 223: 496-498.

IDEKER, T., THORSSON, V., RANISH, J.A., CHRISTMAS, R., BUHLER, J., ENG, J.K., BUMGARNER, R., GOODLETT, D.R., AEBERSOLD, R. and HOOD, L. (2001). Integrated genomic and proteomic analyses of a systematically perturbed metabolic network. *Science* 292: 929-934.

JACOBSEN, S.E. and MEYEROWITZ, E.M. (1997). Hypermethylated *SUPERMAN* epigenetic alleles in *Arabidopsis*. *Science* 277: 1100-1103.

JEFFERSON, R.A., KAVANAGH, T.A. and BEVAN, M.W. (1987). GUS fusions: β-glucuronidase as a sensitive and versatile gene fusion marker in higher plants. *EMBO J.* 6: 3901-3907.

JOFUKU, K.D., DEN BOER, B.G.W., VAN MONTAGU, M. and OKAMURO, J.K. (1994). Control of Arabidopsis flower and seed development by the homeotic gene *APETALA2*. *Plant Cell* 6: 1211-1225.

JOOS, H., INZÉ, D., CAPLAN, A., SORMANN, M., VAN MONTAGU, M. and SCHELL, J. (1983). Genetic analysis of T-DNA transcripts in nopaline crown galls. *Cell* 32: 1057-1067.

JUAREZ, M.T., KUI, J.S., THOMAS, J., HELLER, B.A. and TIMMERMANS, M.C.P. (2004). microRNA-mediated repression of *rolled leaf1* specifies maize leaf polarity. *Nature* 428: 84-88.

KAMALAY, J.C. and GOLDBERG, R.B. (1984). Organ-specific nuclear RNAs in tobacco. *Proc. Natl. Acad. Sci. USA* 81: 2801-2805.

KANNO, A., SAEKI, H., KAMEYA, T., SAEDLER, H. and THEISSEN, G. (2003). Heterotopic expression of class B floral homeotic genes supports a modified ABC model for tulip *(Tulipa gesneriana). Plant Mol. Biol.* 52: 831-841.

KARIMI, M., INZÉ, D. and DEPICKER, A. (2002). GATEWAY™ vectors for *Agrobacterium*-mediated plant transformation. *Trends Plant Sci.* 7: 193-195.

KERK, N.M., CESERANI, T., TAUSTA, S.L., SUSSEX, I.M. and NELSON, T.M. (2003). Laser capture microdissection of cells from plant tissues. *Plant Physiol.* 132: 27-35.

KERSTETTER, R.A., BOLLMAN, K., TAYLOR, R.A., BOMBLIES, K. and POETHIG, R.S. (2001). *KANADI* regulates organ polarity in *Arabidopsis*. *Nature* 411: 706-709.

KIDNER, C.A. and MARTIENSSEN, R.A. (2004). Spatially restricted microRNA directs leaf polarity through ARGONAUTE1. *Nature* 428: 81-84.

KIM, G.-T., TSUKAYA, H., SAITO, Y. and UCHIMIYA, H. (1999). Changes in the shapes of leaves and flowers upon overexpression of cytochrome P450 in *Arabidopsis*. *Proc. Natl. Acad. Sci. USA* 96: 9433-9437.

KIM, G.-T., SHODA, K., TSUGE, T., CHO, K.-H., UCHIMIYA, H., YOKOYAMA, R., NISHITANI, K. and TSUKAYA, H. (2002). The *ANGUSTIFOLIA* gene of *Arabidopsis*, a plant *CtBP* gene, regulates leaf-cell expansion, the arrangement of cortical microtubules in leaf cells and expression of a gene involved in cell-wall formation. *EMBO J.* 21: 1267-1279.

KOMAKI, M.K., OKADA, K., NISHINO, E. and SHIMURA, Y. (1988). Isolation and characterization of novel mutants of *Arabidopsis thaliana* defective in flower development. *Development* 104: 195-203.

KRAMER, E.M. and IRISH, V.F. (1999). Evolution of genetic mechanisms controlling petal development. *Nature* 399: 144-148.

KWIATKOWSKA, D. and DUMAIS, J. (2003). Growth and morphogenesis at the vegetative shoot apex of *Anagallis arvensis* L. *J. Exp. Bot.* 54: 1585-1595.

LAGERCRANTZ, U., PUTTERILL, J., COUPLAND, G. and LYDIATE, D. (1996). Comparative mapping in *Arabidopsis* and *Brassica*, fine scale genome collinearity and congruence of genes controlling flowering time. *Plant J.* 9: 13-20.

LARKIN, J.C., BROWN, M.L. and SCHIEFELBEIN, J. (2003). How do cells know what they want to be when they grow up? Lessons from epidermal patterning in Arabidopsis. *Annu. Rev. Plant Biol.* 54: 403-430.

LAUFS, P., GRANDJEAN, O., JONAK, C., KIÊU, K. and TRAAS, J. (1998). Cellular parameters of the shoot apical meristem in Arabidopsis. *Plant Cell* 10: 1375-1389.

LAUX, T., MAYER, K.F.X., BERGER, J. and JÜRGENS, G. (1996). The *WUSCHEL* gene is required for shoot and floral meristem integrity in *Arabidopsis*. *Development* 122: 87-96.

LINNAEUS, C. (1749). De Peloria. Diss. Ac. Amoenitates Academicae III, Uppsala, Sweden.

LINSMAIER, E.M. and SKOOG, F. (1965). Organic growth factor requirements of tobacco tissue cultures. *Physiol. Plant.* 18: 100-127.

LIPSHUTZ, R.J., FODOR, S.P., GINGERAS, T.R. and LOCKHART, D.J. (1999). High density synthetic oligonucleotide arrays. *Nat. Genet.* 21: 20-24.

LONG, J.A. and BARTON, M.K. (1998). The development of apical embryonic pattern in *Arabidopsis*. *Development* 125: 3027-3035.

LONG, J.A., MOAN, E.I., MEDFORD, J.I. and BARTON, M.K. (1996). A member of the KNOTTED class of homeodomain proteins encoded by the *STM* gene of *Arabidopsis*. *Nature* 379: 66-69.

LÖRZ, H., WERNICKE, W. and POTRYKUS, I. (1979). Culture and plant regeneration of *Hyoscyamus* protoplasts. *Planta Med.* 36: 21-29.

LUCAS, W.J., BOUCHÉ-PILLON, S., JACKSON, D.P., NGUYEN, L., BAKER, L., DING, B. and HAKE, S. (1995). Selective trafficking of KNOTTED1 homeodomain protein and its mRNA through plasmodesmata. *Science* 270: 1980-1983.

LUKOWITZ, W., MAYER, U. and JÜRGENS, G. (1996). Cytokinesis in the Arabidopsis embryo involves the syntaxin-related KNOLLE gene product. *Cell* 84: 61-71.

LUO, D., CARPENTER, R., VINCENT, C., COPSEY, L. and COEN, E. (1996). Origin of floral asymmetry in *Antirrhinum*. *Nature* 383: 794-799.

LUO, D., CARPENTER, R., COPSEY, L., VINCENT, C., CLARK, J. and COEN, E. (1999). Control of organ asymmetry in flowers of *Antirrhinum*. *Cell* 99: 367-376.

LUSSER, A., BROSCH, G., LOIDL, A., HAAS, H. and LOIDL, P. (1997). Identification of maize histone deacetylase HD2 as an acidic nucleolar phosphoprotein. *Science* 277: 88-91.

MÁRTON, L., WULLEMS, G.J., MOLENDIJK, L. and SCHILPEROORT, R.A. (1979). In vitro transformation of cultured cells from *Nicotiana tabacum* by *Agrobacterium tumefaciens*. *Nature* 277: 129-130.

MASSON, P. and FEDOROFF, N.V. (1989). Mobility of the maize Suppressor-mutator element in transgenic tobacco cells. *Proc. Natl. Acad. Sci. USA* 86: 2219-2223.

MATSUBAYASHI, Y. (2003). Ligand-receptor pairs in plant peptide signaling. *J. Cell Sci.* 116, 3863-3870.

MAYER, K.F.X., SCHOOF, H., HAECKER, A., LENHARD, M., JÜRGENS, G. and LAUX, T. (1998). Role of WUSCHEL in regulating stem cell fate in the *Arabidopsis* shoot meristem. *Cell* 95: 805-815.

MAYER, U., TORRES RUIZ, R.A., BERLETH, T., MISÉRA, S. and JÜRGENS, G. (1991). Mutations affecting body organization in the *Arabidopsis* embryo. *Nature* 353: 402-407.

McCLINTOCK, B. (1950). The origin and behavior of mutable loci in maize. *Proc. Natl. Acad. Sci. USA* 36: 344-355.

McCONNELL, J.R., EMERY, J., ESHED, Y., BAO, N., BOWMAN, J. and BARTON, M.K. (2001). Role of *PHABULOSA* and *PHAVOLUTA* in determining radial

patterning in shoots. *Nature* 411: 709-713.

McELVER, J., TZAFRIR, I., AUX, G., ROGERS, R., ASHBY, C., SMITH, K., THOMAS, C., SCHETTER, A., ZHOU, Q., CUSHMAN, M.A., TOSSBERG, J., NICKLE, T., LEVIN, J.Z., LAW, M., MEINKE, D. and PATTON, D. (2001). Insertional mutagenesis of genes required for seed development in *Arabidopsis thaliana*. *Genetics* 159: 1751-1763.

MEINKE, D.W. and SUSSEX, I.M. (1979a). Embryo-lethal mutants of *Arabidopsis thaliana*. *Dev. Biol.* 72: 50-61.

MEINKE, D.W. and SUSSEX, I.M. (1979b). Isolation and characterization of six embryo-lethal mutants of *Arabidopsis thaliana*. *Dev. Biol.* 72: 62-72.

MEISSNER, R.C., JIN, H., COMINELLI, E., DENEKAMP, M., FUERTES, A., GRECO, R., KRANZ, H.D., PENFIELD, S., PETRONI, K., URZAINQUI, A., MARTIN, C., PAZ-ARES, J., SMEEKENS, S., TONELLI, C., WEISSHAAR, B., BAUMANN, E., KLIMYUK, V., JONES, J.J.D., PEREIRA, A., WISMAN, E. and BEVAN, M. (1999). Function search in a large transcription factor gene family in Arabidopsis: assessing the potential of reverse genetics to identify insertional mutations in R2R3 *MYB* genes. *Plant Cell* 11: 1827-1840.

MEYEROWITZ, E.M. (2002). Plants compared to animals: the broadest comparative study of development. *Science* 295: 1482-1485.

MEYEROWITZ, E.M., SMYTH, D.R. and BOWMAN, J.L. (1989). Abnormal flowers and pattern formation in floral development. *Development* 106: 209-217.

MITCHISON, G.J. (1977). Phyllotaxis and the Fibonacci series. *Science* 196: 270-275.

MIZUKAMI, Y. and FISCHER, R.L. (2000). Plant organ size control: *AINTEGUMENTA* regulates growth and cell numbers during organogenesis. *Proc. Natl. Acad. Sci. USA* 97: 942-947.

MORGANTE, M. and SALAMINI, F. (2003). From plant genomics to breeding practice. *Curr. Opin. Biotech.* 14, 214-219.

MULLIS, K.B. and FALOONA, F.A. (1987). Specific synthesis of DNA in vitro via a polymerase-catalyzed chain reaction. In Recombinant DNA, Part F, (*Methods in Enzymology,* Vol. 155), R. Wu (Ed.). San Diego, Academic Press, pp. 335-350.

MURASHIGE, T. and SKOOG, F. (1962). A revised medium for rapid growth and bio assays with tobacco tissue cultures. *Physiol. Plant.* 15: 473-497.

NAGATA, T. and TAKEBE, I. (1970). Cell wall regeneration and cell division in isolated tobacco mesophyll protoplasts. *Planta* 92: 301-308.

NAGY, J.I. and MALIGA, P. (1976). Callus induction and plant regeneration from mesophyll protoplasts of *Nicotiana sylvestris*. Z. *Pflanzenphysiol.* 78: 453-455.

NATH, U., CRAWFORD, B.C.W., CARPENTER, R. and COEN, E. (2003). Genetic control of surface curvature. *Science* 299: 1404-1407.

NELISSEN, H., CLARKE, J.H., DE BLOCK, M., DE BLOCK, S., VANDERHAEGHEN, R., ZIELINSKI, R.E., DYER, T., LUST, S., INZÉ, D. and VAN LIJSEBETTENS, M. (2003a). DRL1, a homolog of the yeast TOT4/KTI12 protein, has a function in meristem activity and organ growth in plants. *Plant Cell* 15: 639-654.

NELISSEN, H., FLEURY, D., BRUNO, L., ROBLES, P., DE VEYLDER, L., TRAAS, J. MICOL, J.L., VAN MONTAGU, M., INZÉ, D. and VAN LIJSEBETTENS, M. (2005). The *elongata* mutants identify a functional Elongator complex in plants with a role in cell proliferation during organ growth. *Proc. Natl. Acad. Sci. USA* 102: 7754-7759.

OHNO, S. (1970). *Evolution by gene duplication.* Springer, New York.

ORI, N., ESHED, Y., CHUCK, G., BOWMAN, J.L. and HAKE, S. (2000). Mechanisms that control *knox* gene expression in the *Arabidopsis* shoot. *Development* 127: 5523-5532.

PALATNIK, J.F., ALLEN, E., WU, X., SCHOMMER, C., SCHWAB, R., CARRINGTON, J.C. and WEIGEL, D. (2003). Control of leaf morphogenesis by microRNAs. *Nature* 425: 257-263.

PANDEY, R., MÜLLER, A., NAPOLI, C.A., SELINGER, D.A., PIKAARD, C.S., RICHARDS, E.J., BENDER, J., MOUNT, D.W. and JORGENSEN, R.A. (2002). Analysis of histone acetyltransferase and histone deacetylase families of *Arabidopsis thaliana* suggests functional diversification of chromatin modification among multicellular eukaryotes. *Nucleic Acids Res.* 30: 5036-5055.

PELAZ, S., DITTA, G.S., BAUMANN, E., WISMAN, E. and YANOFSKY, M.F. (2000). B and C floral organ identity functions require *SEPALLATA* MADS-box genes. *Nature* 405: 200-203.

PEREIRA, A. and SAEDLER, H. (1989). Transpositional behavior of the maize *En/Spm* element in transgenic tobacco. *EMBO J.* 8: 1315-1321.

PEREIRA, A., SCHWARZ-SOMMER, ZS., GIERL, A., BERTRAM, I., PETERSON, P.A. and SAEDLER, H. (1985). Genetic and molecular analysis of the Enhancer (En) transposable element system of *Zea mays*. *EMBO J.* 4: 17-23.

PÉREZ-PÉREZ, J.M., SERRANO-CARTAGENA, J. and MICOL, J.L. (2002). Genetic analysis of natural variations in the architecture of *Arabidopsis thaliana* vegetative leaves. *Genetics* 162, 893-915.

PIEN, S., WYRZYKOWSKA, J., McQUEEN-MASON, S., SMART, C. and FLEMING, A. (2001). Local expression of expansin induces the entire process of leaf development and modifies leaf shape. *Proc. Natl. Acad. Sci. USA* 98: 11812-11817.

POHLMAN, R.F., FEDOROFF, N.V. and MESSING, J. (1984). The nucleotide sequence of the maize controlling element *Activator*. *Cell* 37: 635-643.

POTTEN, C.S. and LOEFFLER, M. (1990). Stem cells: attributes, cycles, spirals, pitfalls and uncertainties: lessons for and from the crypt. *Development* 110: 1001-1020.

PRUSINKIEWICZ, P. (2004). Modeling plant growth and development. *Curr. Opin. Plant Biol.* 7: 79-83.

PYKE, K.A., MARRISON, J.L. and LEECH, R.M. (1991). Temporal and spatial development of the cells of the expanding first leaf of *Arabidopsis thaliana* (L.) Heynh. *J. Exp. Bot.* 42: 1407-1416.

REINHARDT, D., MANDEL, T. and KUHLEMEIER, C. (2000). Auxin regulates the initiation and radial position of plant lateral organs. *Plant Cell* 12: 507-518.

REINHARDT, D., PESCE, E.-R., STIEGER, P., MANDEL, T., BALTENSPERGER, K., BENNETT, M., TRAAS, J., FRIML, J. and KUHLEMEIER, C. (2003). Regulation of phyllotaxis by polar auxin transport. *Nature* 426: 255-260.

REINHART, B.J., WEINSTEIN, E.G., RHOADES, M.W., BARTEL, B. and BARTEL, D.P. (2002). MicroRNAs in plants. *Genes Dev.* 16: 1616-1626.

REYES, J.C., HENNIG, L. and GRUISSEM, W. (2002). Chromatin-remodeling and memory factors. New regulators of plant development. *Plant Physiol.* 130: 1090-1101.

RHOADES, M.W., REINHART, B.J., LIM, L.P., BURGE, C.B., BARTEL, B. and BARTEL, D.P. (2002). Prediction of plant microRNA targets. *Cell* 110: 513-520.

ROLLAND-LAGAN, A.-G., BANGHAM, J.A. and COEN, E. (2003). Growth dynamics underlying petal shape and asymmetry. *Nature* 422: 161-163.

SAWA, S., WATANABE, K., GOTO, K., KANAYA, E., MORITA, R.H. and OKADA, K. (1999). *FILAMENTOUS FLOWER,* a meristem and organ identity gene of *Arabidopsis*, encodes a protein with a zinc finger and HMG-related domains. *Genes Dev.* 13: 1079-1088.

SCHERES, B., WOLKENFELT, H., WILLEMSEN, V., TERLOUW, M., LAWSON, E., DEAN, C. and WEISBEEK, P. (1994). Embryonic origin of the *Arabidopsis* primary root and root meristem initials. *Development* 120: 2475-2487.

SCHERES, B., DI LAURENZIO, L., WILLEMSEN, V., HAUSER, M.-T., JANMAAT, K., WEISBEEK, P. and BENFEY, P.N. (1995). Mutations affecting the radial organisation of the *Arabidopsis* root display specific defects throughout the embryonic axis. *Development* 121: 53-62.

SCHNEEBERGER, R., TSIANTIS, M., FREELING, M. and LANGDALE, J.A. (1998). The *rough sheath2* gene negatively regulates homeobox gene expression during maize leaf development. *Development* 125: 2857-2865.

SCHOOF, H., LENHARD, M., HAECKER, A., MAYER, K.F.X., JÜRGENS, G. and LAUX, T. (2000). The stem cell population of *Arabidopsis* shoot meristem is maintained by a regulatory loop between the *CLAVATA* and *WUSCHEL* genes. *Cell* 100: 635-644.

SCHWARZ-SOMMER, Z., HUIJSER, P., NACKEN, W., SAEDLER, H. and SOMMER, H. (1990). Genetic control of flower development by homeotic genes in *Antirrhinum majus*. *Science* 250: 931-936.

SHANE, M.W., CRAMER, M.D., FUNAYAMA-NOGUCHI, S., CAWTHRAY, G.R., MILLAR, A.H., DAY, D.A. and LAMBERS, H. (2004). Developmental physiology of cluster-root carboxylate synthesis and exudation in harsh hakea. Expression of phosphoenol pyruvate carboxylase and the alternative oxidase. *Plant Physiol.* 135: 549-560.

SHEVELL, D.E., LEU, W.-M., GILLMOR, C.S., XIA, G., FELDMANN, K.A. and CHUA, N.-H. (1994). *EMB30* is essential for normal cell division, cell expansion and cell adhesion in Arabidopsis and encodes a protein that has similarity to Sec7. *Cell* 77: 1051-1062.

SIEGFRIED, K.R., ESHED, Y., BAUM, S.F., OTSUGA, D., DREWS, G.N. and BOWMAN, J.L. (1999). Members of the *YABBY* gene family specify abaxial cell fate in *Arabidopsis*. *Development* 126: 4117-4128.

SIMILLION, C., VANDEPOELE, K., VAN MONTAGU, M.C.E., ZABEAU, M. and VAN DE PEER, Y. (2002). The hidden duplication past of *Arabidopsis thaliana. Proc. Natl. Acad. Sci. USA* 99: 13627-13632.

SOMERVILLE, C. and KOORNNEEF, M. (2002). A fortunate choice: the history of *Arabidopsis* as a model plant. *Nat. Rev. Genet.* 3: 883-889.

SOMMER, H., BELTRÁN, J.-P., HUIJSER, P., PAPE, H., LÖNNIG, W.-E., SAEDLER, H. and SCHWARZ-SOMMER, Z. (1990). *Deficiens*, a homeotic gene involved in the control of flower morphogenesis in *Antirrhinum majus:* the protein shows homology to transcription factors. *EMBO J.* 9: 605-613.

STEEVES, T.A. and SUSSEX, I.M. (1989). *Patterns in plant development,* 2nd ed. Cambridge, Cambridge University Press.

STONE, S.L., HAUKSDOTTIR, H., TROY, A., HERSCHLEB, J., KRAFT, E. and CALLIS, J. (2005). Functional analysis of the RING-type ubiquitin ligase family of Arabidopsis. *Plant Physiol.* 137: 13-30.

SUNG, S. and AMASINO, R.M. (2004). Vernalization and epigenetics: how plants remember winter. *Curr. Opin. Plant Biol.* 7: 4-10.

SUSSEX, I.M. (1951). Experiments on the cause of dorsiventrality in leaves. *Nature* 167: 651-652.

SUSSEX, I.M. (1955). Morphogenesis in *Solanum tuberosum* L.: experimental investigation of leaf dorsoventrality and orientation in the juvenile shoot. *Phytomorphology* 5: 286-300.

TANDRE, K., ALBERT, V.A., SUNDÅS, A. and ENGSTRÖM, P. (1995). Conifer homologues to genes that control floral development in angiosperms. *Plant Mol. Biol.* 27: 69-78.

TANKSLEY, S.D. (2004). The genetic, developmental and molecular bases of fruit size and shape variation in tomato. *Plant Cell* 16: S181-S189.

TARDIEU, F. and GRANIER, C. (2000). Quantitative analysis of cell division in leaves: methods, developmental patterns and effects of environmental conditions. *Plant Mol. Biol.* 43: 555-567.

THEIβEN, G. and SAEDLER, H. (2001). Floral quartets. *Nature* 409: 469-471.

THEISSEN, G., BECKER, A., DI ROSA, A., KANNO, A., KIM, J.T., MÜNSTER, T., WINTER, K.-U. and SAEDLER, H. (2000). A short history of MADS-box genes in plants. *Plant Mol. Biol.* 42: 115-149.

THIMM, O., BLÄSING, O., GIBON, Y., NAGEL, A., MEYER, S., KRÜGER, P., SELBIG, J., MÜLLER, L.A., RHEE, S.Y. and STITT, M. (2004). MAPMAN: a user-driven tool to display genomics data sets onto diagrams of metabolic pathways and other biological processes. *Plant J.* 37: 914-939.

TIMMERMANS, M.C.P., HUDSON, A., BECRAFT, P.W. and NELSON, T. (1999). ROUGH SHEATH2: A Myb protein that represses *knox* homeobox genes in maize lateral organ primordia. *Science* 284: 151-153.

TROTOCHAUD, A.E., HAO, T., WU, G., YANG, Z. and CLARK, S.E. (1999). The CLAVATA1 receptor-like kinase requires CLAVATA3 for its assembly into a signaling complex that includes KAPP and a Rho-related protein. *Plant Cell* 11: 393-405.

TSIANTIS, M. and HAY, A. (2003). Comparative plant development: the time of the leaf? *Nat. Rev. Genet.* 4: 169-180.

TSIANTIS, M., SCHNEEBERGER, R., GOLZ, J.F., FREELING, M. and LANGDALE, J.A. (1999). The maize *rough sheath2* gene and leaf development programs in monocot and dicot plants. *Science* 284: 154-156.

TSUKAYA, H. (2002). Interpretation of mutants in leaf morphology: genetic evidence for a compensatory system in leaf morphogenesis that provides a new link between cell and organismal theories. *Int. Rev. Cytol.* 217: 1-39.

TZAFRIR, I., DICKERMAN, A., BRAZHNIK, O., NGUYEN, Q., McELVER, J., FRYE, C., PATTON, D. and MEINKE, D. (2003). The *Arabidopsis* SeedGenes project. *Nucleic Acids Res.* 31: 90-93.

VALVEKENS, D., VAN MONTAGU, M. and VAN LIJSEBETTENS, M. (1988). *Agrobacterium tumefaciens* -mediated transformation of *Arabidopsis thaliana* root explants by using kanamycin selection. *Proc. Natl. Acad. Sci. USA* 85: 5536-5540.

VAN DEN BERG, C., WILLEMSEN, V., HAGE, W., WEISBEEK, P. and SCHERES, B. (1995). Cell fate in the *Arabidopsis* root meristem determined by directional signalling. *Nature* 378: 62-65.

VAN DEN BERG, C., WILLEMSEN, V., HENDRIKS, G., WEISBEEK, P. and SCHERES, B. (1997). Short-range control of cell differentiation in the *Arabidopsis* root meristem. *Nature* 390: 287-289.

VAN DE PEER, Y. (2004). Computational approaches to unveiling ancient genome duplications. *Nat. Genet.* 5: 752-763.

VAN LIJSEBETTENS, M. and CLARKE, J. (1998). Leaf development in *Arabidopsis*. *Plant Physiol. Biochem.* 36: 47-60.

VAN LIJSEBETTENS, M., TERRYN, N. and VAN MONTAGU, M. (2002). *Arabidopsis*. In *Encyclopedia of Molecular Biology,* Vol. 1, T.E. Creighton (Ed.). New York, Wiley & Sons [http://www.mrw.interscience.wiley.com/embm].

VANDENBUSSCHE, M., ZETHOF, J., SOUER, E., KOES, R., TORNIELLI, G.B., PEZZOTTI, M., FERRARIO, S., ANGENENT, G.C. and GERATS, T. (2003a). Toward the analysis of the petunia MADS box gene family by reverse and forward transposon insertion mutagenesis approaches: B, C and D floral organ identity functions require *SEPALLATA* -like MADS box genes in petunia. *Plant Cell* 15: 2680-2693.

VANDENBUSSCHE, M., THEISSEN, G., VAN DE PEER, Y. and GERATS, T. (2003b). Structural diversification and neo-functionalization during floral MADS-box gene evolution by C-terminal frameshift mutations. *Nucleic Acids Res.* 31: 4401-4409.

VANDENBUSSCHE, M., ZETHOF, J., ROYAERT, S., WETERINGS, K. and GERATS, T. (2004). The duplicated B-class heterodimer model: whorl-specific effects and complex genetic interactions in *Petunia hybrida* flower development. *Plant Cell* 16: 741-754.

VAUGHAN, J.G. (1952). Structure of the angiosperm apex. *Nature* 169: 458-459.

VENTER, J.C. (2004). *Genomes and Life.* Talk presented at the Symposium «Life, a Nobel story», organized by the The Royal Flemish Chemical Society, Section Biotechnology, held in Brussels (Belgium), April, 28, 2004.

VOLLBRECHT, E., VEIT, B., SINHA, N. and HAKE, S. (1991). The developmental gene *Knotted-1* is a member of a maize homeobox gene family. *Nature* 350: 241-243.

VON GOETHE, J.W. (1790). Versuch die Metamorphose der Pflanzen zu erklären. Gotha, C.W. Ettinger.

WAITES, R. and HUDSON, A. (1995). *phantastica* : a gene required for dorsoventrality of leaves in *Antirrhinum majus. Development* 121: 2143-2154.

WAITES, R., SELVADURAI, H.R.N., OLIVER, I.R. and HUDSON, A. (1998). The *PHANTASTICA* gene encodes a MYB transcription factor involved in growth and dorsoventrality of lateral organs in *Antirrhinum*. *Cell* 93: 779-789.

WEIGEL, D. and JÜRGENS, G. (2002). Stem cells that make stems. *Nature* 415: 751-754.

WEIGEL, D. and MEYEROWITZ, E.M. (1994). The ABCs of floral homeotic genes. *Cell* 78: 103-209.

WEIGEL, D. and NILSSON, O. (1995). A developmental switch sufficient for flower initiation in diverse plants. *Nature* 377: 495-500.

WYRZYKOWSKA, J., PIEN, S., SHEN, W.H. and FLEMING, A.J. (2002). Manipulation of leaf shape by modulation of cell division. *Development* 129: 957-964.

YANOFSKY, M.F., MA, H., BOWMAN, J.L., DREWS, G.N., FELDMANN, K.A. and MEYEROWITZ, E.M. (1990). The protein encoded by the *Arabidopsis* homeotic gene *agamous* resembles transcription factors. *Nature* 346: 35-39.

YOUNG, N.D., MUDGE, J. and ELLIS, T.H.N. (2003). Legume genomes: more than peas in a pod. *Curr. Opin. Plant Biol.* 6: 199-204.

ZAENEN, I., VAN LAREBEKE, N., TEUCHY, H., VAN MONTAGU, M. and SCHELL, J. (1974). Supercoiled circular DNA in crown gall inducing *Agrobacterium* strains. *J. Mol. Biol.* 86: 109-127.

ZAMBRYSKI, P., JOOS, H., GENETELLO, C., LEEMANS, J., VAN MONTAGU, M. and SCHELL, J. (1983). Ti plasmid vector for the introduction of DNA into plant cells without alteration of their normal regeneration capacity. *EMBO J.* 2: 2143-2150.

ZAMBRYSKI, P., TEMPÉ, J. and SCHELL, J. (1989). Transfer and function of T-DNA genes from *Agrobacterium* Ti and Ri plasmids in plants. *Cell* 56: 193-201.