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ARABIDOPSIS: A RICH HARVEST 10 YEARS AFTER COMPLETION OF THE GENOME SEQUENCE

The development of Arabidopsis as a model plant

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SUMMARY

Twenty-five years ago, *Arabidopsis thaliana* emerged as the model organism of choice for research in plant biology. A consensus was reached about the need to focus on a single organism to integrate the classical disciplines of plant science with the expanding fields of genetics and molecular biology. Ten years after publication of its genome sequence, Arabidopsis remains the standard reference plant for all of biology. We reflect here on the major advances and shared resources that led to the extraordinary growth of the Arabidopsis research community. We also underscore the importance of continuing to expand and refine our detailed knowledge of Arabidopsis while seeking to appreciate the remarkable diversity that characterizes the plant kingdom.

Keywords: Arabidopsis, community resources, history, model organism, plant biology.

INTRODUCTION

Of all the known species of flowering plants, *Arabidopsis thaliana* stands alone as the most thoroughly studied. Measured by the total number of journal publications, other plants such as maize, soybean, petunia, tomato, pea, and snapdragon, once considered as promising candidates to guide plant research into the future, all lag far behind. Not even rice (*Oryza sativa*) has kept pace with Arabidopsis, using research publications as the benchmark. In 2008 alone, more than 3500 papers on Arabidopsis were added to the PubMed database (http://www.ncbi.nlm.nih.gov/pubmed). By contrast, just seven Arabidopsis publications were listed for 1979 and 65 for all preceding years combined. The growth of Arabidopsis research over the last 30 years has been remarkable, rewarding, and transformative.

Arabidopsis was originally adopted as a model organism because of its usefulness for genetic experiments. Important features included a short generation time, small size that limited the requirement for growth facilities, and prolific seed production through self-pollination. Although these features are still important, other attributes that could not be foreseen 40 years ago have allowed Arabidopsis to remain the premiere model for plant biology. A change from individual research efforts focused on specific disciplines to more interdisciplinary, multi-investigator studies, requiring extensive community resources, was an essential factor in the growth of Arabidopsis as a favoured organism. Traditional plant science was subdivided into discrete, classical disciplines, including anatomy, morphology, physiology, and biochemistry. When molecular biology emerged as a major force in the 1970s, plants were not the organism of choice for experimentation. One problem was the limited availability of shared resources needed to bring plants up to the same level of sophistication enjoyed by other model organisms. The agricultural foundation for plant research at the time also made it difficult to gain support for elevating an 'outsider' to special research status.

The increased role of genetics in discipline integration (Pruitt *et al.*, 2003) and the availability of powerful tools in molecular biology resulted in the gradual realization that plant biologists needed to focus attention on a single organism most amenable to detailed analysis. That this concept of discipline integration took time to be accepted by the classical disciplines can be illustrated by the rejection of a paper (Koornneef *et al.*, 1984) on abscisic acid (ABA)-insensitive mutants of Arabidopsis by one of the main journals in plant physiology at that time. The only argument

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for rejection was that a paper on mutants should be published in a genetics journal. This attitude was surprising in the light of past research on Drosophila and a wide range of microorganisms, where genetics had long been championed to address fundamental questions in biochemistry and development.

The emergence of *A. thaliana* as a model organism has been documented several times in the past, through historical retrospectives written either by those who participated in the research (Rédei, 1992; Fink, 1998; Meinke *et al.*, 1998; Dean, 2001; Meyerowitz, 2001; Somerville and Koornneef, 2002; Pruitt *et al.*, 2003; van Lijsebettens and van Montagu, 2005; Koncz, 2006) or by interested observers (North, 1985; Patrusky, 1991; Moffat, 1992; Pennisi, 2000; Endersby, 2007; Leonelli, 2007). Readers are encouraged to consult Meyerowitz (2001) and Somerville and Koornneef (2002) for additional historical details. Several major reviews detailing the breadth of Arabidopsis research in the pre-genomics age have also been published (Rédei, 1970, 1975; Meyerowitz, 1987, 1989). Our objective in the present review is to be more reflective than comprehensive in discussing factors that contributed to the establishment of the Arabidopsis research community. Because we could not include every major advance in such a brief review, we ask for understanding from those individuals whose important contributions are not mentioned. We also encourage those currently working on Arabidopsis to remember and continue to utilize when appropriate the rich historical foundation of Arabidopsis research. To underscore this point, we include updates on the classical genetic map and the analysis of natural variation in Arabidopsis, both of which trace their origins back to detailed phenotype information and seed stocks collected more than 40 years ago.

A BRIEF HISTORY OF ARABIDOPSIS RESEARCH

The early years (before 1975)

Without question, Friedrich Laibach (Figure 1) is the founder of experimental Arabidopsis research. He described the correct chromosome number during his PhD research (Laibach, 1907) and returned to this species in the 1930s when he was an established botanist. In a seminal paper, Laibach (1943) made a clear case for the suitability of Arabidopsis for genetic studies. Laibach and his students also emphasized the use of natural variation for the analysis of physiological traits such as flowering time (Laibach, 1951) and seed dormancy (Kugler, 1951). In addition, Laibach initiated experiments on the treatment of Arabidopsis with X-rays. This enabled his PhD student, Erna Reinholz, to isolate the first induced Arabidopsis mutants. Laibach, who had to leave his position at Frankfurt University in 1945, continued his research in a private laboratory in the nearby town of Limburg/Lahn called 'Biologisches Forschungsinstitut Limburg'. This facility consisted of several rooms in the pharmacy of

Figure 1. Central figures in the early years of Arabidopsis research. Left to right: Friedrich Laibach (photo from George Rédei, courtesy of Elliot Meyerowitz); Gerhard Röbbelen (photo courtesy of the European Association for Plant Breeding Research); and George Rédei (photo courtesy of Csaba Koncz).

his co-worker, Franz Josef Kribben, who was probably the first to call Arabidopsis the botanical Drosophila (Kribben, 1964). The study of mutants was already a focus of Arabidopsis research when John Langridge described the first auxotrophs in higher plants in an influential paper in *Nature* (Langridge, 1955).

Interest in Arabidopsis research increased throughout the 1950s. One landmark event was the emigration in 1956 of George Rédei from Hungary to the United States (Koncz, 2006). From his laboratory at the University of Missouri, Rédei became the leading proponent of Arabidopsis research in the United States. He joined researchers in Germany (Napp-Zinn, Röbbelen, Müller), the Czech Republic (Veleminsky, Gichner, Cetl), the Netherlands (Feenstra, van der Veen), and Belgium (Jacobs, Bouharmont), to form an active research community in the 1960s (Somerville and Koornneef, 2002). Gerhard Röbbelen led this initiative and organized the first international Arabidopsis conference in 1965 in Göttingen, Germany. Röbbelen also published the Arabidopsis Information Service (AIS) newsletter beginning in 1964 and maintained a seed stock centre that included Laibach's collection of ecotypes (called accessions later on) as well as a number of induced mutants.

Although it was expected that such a promising start would lead to further growth of Arabidopsis research, in practice the opposite occurred in the early 1970s, when many of the individuals mentioned above left the field. In retrospect, there were probably several reasons why the future of Arabidopsis research was viewed with scepticism in the mid 1970s. Principal among them was the impression that success in plant tissue culture was central to the future of plant biology. Petunia and tobacco, where the desired manipulation of cells in culture was somewhat routine, were therefore viewed in a more positive light than Arabidopsis, which seemed to resist most initial attempts to proliferate and regenerate in culture. The small size of Arabidopsis chromosomes was also viewed by some as a disadvantage, primarily for cytogenetic analyses, rather than an advantage, as later understood for molecular studies. Röbbelen became more involved in rapeseed research and plant breeding,

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ultimately becoming a leading figure in this field in Germany. Eventually, Röbbelen transferred his responsibility for the AIS to Albert Kranz at the University of Frankfurt, who served in this capacity from 1974 until his retirement in 1990. Arabidopsis also received negative publicity from a controversial set of experiments (Ledoux *et al.*, 1974) that claimed to demonstrate complementation of thiamine-deficient mutants following treatment of seeds with DNA from *Escherichia coli* (Koncz, 2006). Interest in Arabidopsis research declined throughout this period, as reflected in decreased publication rates and fewer participants at the second international Arabidopsis conference in 1976 (Meyerowitz, 2001).

The renaissance period (1976–89)

Renewed interest in Arabidopsis began in the late 1970s with the search for a suitable plant model for research in molecular genetics. It gained momentum when several groups (Figure 2) began to exploit the genetic potential of Arabidopsis to characterize plant-specific processes. The excellent review article by Rédei on Arabidopsis as a genetic organism, published in the well-known *Annual Review of Genetics*, played a pivotal role in the resurgence of interest in Arabidopsis and the recruitment of young investigators to the field (Rédei, 1975). In the area of plant development, David Meinke began to work on embryo-lethal mutants as a graduate student with Ian Sussex at Yale in 1976, expanding upon the work of Andreas Müller 15 years before in Gatersleben, Germany (Müller, 1963). This ultimately led to publications in *Developmental Biology* on the use of Arabidopsis as a model system for the study of plant embryo development (Meinke and Sussex, 1979a,b). Meinke was first introduced to the details of Müller's work when he convinced the landlady of his apartment in New Haven to translate the original paper from German in return for cleaning out her basement. This anecdote illustrates a barrier to advances in Arabidopsis research that is often overlooked: the language problem encountered by some when attempting to access the early literature. Another Yale graduate student, Elliot Meyerowitz, was first introduced to Arabidopsis during this time as well, although his interests were focused then on Drosophila.

Figure 2. Some early contributors to the renaissance period of Arabidopsis research. Left to right; top: Chris Somerville and Elliot Meyerowitz; bottom: Maarten Koornneef and David Meinke.

Chris Somerville and colleagues were instrumental in promoting Arabidopsis throughout the 1980s and beyond. Their early work illustrated the value of mutant analysis for plant physiology and biochemistry and included a series of elegant studies on photorespiration (Somerville and Ogren, 1980), starch and lipid biosynthesis, plant hormone responses, and cell wall architecture (Estelle and Somerville, 1986). Informative mutant screens were being performed at the same time in Wageningen by Maarten Koornneef and Jaap van der Veen. Their work on a number of plant-specific substances and processes, from phytohormones and photoreceptors to flowering time, resulted in several influential publications in the early 1980s, along with the first comprehensive genetic map (Koornneef *et al.*, 1983).

Several key events in the growth of the Arabidopsis research community took place in the mid 1980s. The most noteworthy was the realization that the small size of the Arabidopsis genome (Leutwiler *et al.*, 1984) was a distinct advantage in the age of molecular genetics (Meyerowitz and Pruitt, 1985). This observation was instrumental in attracting the attention of investigators working on other model organisms. Flowering plants, once considered marginally significant by many of those interested in advancing the fundamentals of cell and molecular biology, started to be viewed in a more positive light, especially when the small number of funded investigators in the field was seen as an opportunity and not a hindrance. Another important factor involved presentations and discussions about Arabidopsis at scientific conferences, including the Gordon Conference on Plant Cell and Tissue Culture in 1983 and the Keystone Symposium on Plant Genetics in 1985. A picture from the Keystone meeting has been published before (Meyerowitz, 2001), with Shauna and Chris Somerville, Elliot Meyerowitz, David Meinke, and Maarten Koornneef in attendance.

A third factor in the dramatic rise of Arabidopsis research in the mid 1980s was the vision and support provided by key administrators at US funding agencies, most notably DeLill Nasser, Machi Dilworth, and Mary Clutter at the National Science Foundation (NSF). Their combined efforts were instrumental in helping to encourage young investigators, establish shared resources, and guide the Arabidopsis community into the modern age of genomics. These factors and others, such as advances in plant transformation

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methods noted later, energized the Arabidopsis community and convinced a number of leading scientists from other disciplines, including Ron Davis, Howard Goodman, Gerald Fink, and Fred Ausubel, to pursue and promote research on Arabidopsis. This added level of distinction proved helpful in several ways, including the recruitment of talented students and post-docs who represented the next generation of Arabidopsis biologists. The fact that Elliot Meyerowitz, and later on Gerd Jürgens, leading proponents of using Arabidopsis for research in molecular and developmental genetics, were respected young investigators from the Drosophila world, further reinforced the message that plants were indeed amenable to molecular genetic analysis. Clearly, Arabidopsis research efforts around this time benefited from the demonstration with other model organisms that combining genetics and molecular biology represented a powerful approach to addressing biological questions (Meyerowitz, 1987). The cloning of the first Arabidopsis gene by Chang and Meyerowitz (1986) further confirmed that molecular genetic approaches to plant biology were starting to yield results.

The revival of Arabidopsis research was on full display at the Michigan State conference on Arabidopsis in 1987 organized by Chris Somerville and colleagues. Even so, the number of participants (about 200) and small booklet of abstracts (85) were unimpressive by modern standards. The most frequent entries in the abstract index reflected a different set of priorities at the time: *Agrobacterium*, amino acid analogues, embryo lethals, heterologous probes, lambda libraries, tissue culture, and transformation. But this meeting helped to define the modern age of Arabidopsis research and establish the atmosphere of collaboration that remains in effect today. An *ad hoc* meeting of a handful of principal investigators held during that conference also laid the foundation for the community infrastructure that would play an important role in future efforts to sequence the Arabidopsis genome. When Chris Somerville described the breadth of Arabidopsis research at a subsequent meeting in Bloomington, IN, two years later, it was clear that Arabidopsis was poised to enter the modern age and compete with other model organisms for recognition and respect (Somerville, 1989).

THE ARABIDOPSIS TRANSFORMATION STORY

An important breakthrough for Arabidopsis research involved the development of efficient transformation procedures. Arabidopsis transformation made possible the introduction back into plants of cloned genes of interest for subsequent analysis and the production of insertion mutants through random disruption of endogenous genes. At first, transformation methods based on *Agrobacterium tumefaciens* infection of leaf explants in culture seemed the way to go (Lloyd *et al.*, 1986). Soon this method was improved (Valvekens *et al.*, 1988) by using a two-step tissue culture procedure and root explants as the starting material. The success of these tissue culture methods depended much on the parental genotype, and unfortunately the most common laboratory accessions of Arabidopsis were not very responsive. For this reason, investigators turned to other accessions to generate transgenic plants. This caused problems with subsequent genetic studies, where phenotypes were often affected by genetic background.

Shortly thereafter, Feldmann and Marks (1987) described a method that did not require tissue culture and was based on incubating mature seeds with *Agrobacterium*, growing plants from these treated seeds, and then screening for antibiotic resistance among the progeny seedlings. This method relied on the continuous growth of *Agrobacterium* within the plant until it could infect the egg cell inside an ovule. Recognizing this underlying biology led to improved methods where *Agrobacterium* was introduced into the plant when flower buds had already formed (Bechtold *et al.*, 1993; Clough and Bent, 1998). The large populations of T-DNA insertion lines that Ken Feldmann generated using the original method, first at DuPont (Wilmington, DE) and later at the University of Arizona, were nevertheless a pivotal advance that allowed the identification of a wide range of insertion mutants amenable to gene isolation. One of the first T-DNA mutants characterized in detail appeared on the cover of *Science* (Feldmann *et al.*, 1989).

Transformation of Arabidopsis is now highly reproducible and not that genotype dependent, allowing for the generation of thousands of transformants when needed. Apart from the use of transformation to generate plants that express (or over-express) known genes of interest and to establish large collections of insertion lines for reverse genetics, the same technology has been used to introduce reporter gene constructs into plants, thereby allowing localization and quantification of expression patterns and development of lines with localized expression in specific tissues (Haseloff and Amos, 1995) and subcellular structures (Cutler *et al.*, 2000). The power of such an approach was later demonstrated again in combination with cell sorting (Birnbaum *et al.*, 2003). The high efficiency of plant transformation was not predictable when Arabidopsis was adopted as a model in the early 1980s. But dramatic improvements in this important genetic tool enhanced the stature and utility of Arabidopsis as an experimental organism, leading Somerville and Koornneef (2002) to conclude that adopting Arabidopsis as a model organism was indeed a fortunate choice.

THE IMPORTANCE OF MUTANT SCREENS

Mutant screens played an important role in the emergence of Arabidopsis as a model genetic organism. The short life cycle, small plant size, and efficient reproduction through self-pollination made Arabidopsis an early favourite for studying induced mutations in plants. In the late 1950s and

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1960s, the efficiency of mutagenic treatments was readily gauged using the Müller (1961) embryo test and the frequency of either chlorophyll-deficient seedlings or sterile plants. Ethylmethane sulphonate (EMS) was first introduced for mutation studies in those early days and remains an effective and popular mutagen today. That saturation mutagenesis was an option in Arabidopsis was shown by finding multiple mutant alleles of the same gene in a reasonably sized population of M_2 seeds (Koornneef *et al.*, 1982). A variety of phenotypic screens were developed, yielding large collections of mutants, with those deficient in chlorophyll among the most prevalent (Röbbelen, 1957). The suitability of Arabidopsis for biochemical genetics was confirmed through exhaustive studies of thiamine auxotrophs with a seedling lethal phenotype (Feenstra, 1964; Li and Rédei, 1969). An early example of a directed biochemical screen aimed at a specific metabolic defect was the use of chlorate resistance to obtain mutants affected in nitrate uptake or metabolism (Oostindier-Braaksma and Feenstra, 1973). Selection for resistance was thereafter also applied to plant hormones (Koornneef *et al.*, 1984; Bleecker *et al.*, 1988). Screens for a deviating phenotype under conditions where the process being studied was critical or limiting were also devised, as demonstrated with photorespiration mutants at high CO_2 levels (Somerville and Ogren, 1980) and photoreceptor mutants with altered hypocotyls under specific light conditions (Koornneef *et al.*, 1980).

Because mutation frequencies were high and scores of mutagenized individuals could be sampled with minimal effort, large-scale forward genetic screens for specific biochemical defects soon became routine, as demonstrated in a convincing manner by the Somerville group. This ultimately led to the statement that when no mutants are found in 2000 M_2 plants, there is something wrong with the design of the screen (Estelle and Somerville, 1986). Surprisingly, the issue of gene redundancy, which often prevents the appearance of a mutant phenotype when a related gene with a similar function remains unaltered, was not widely discussed, perhaps because it was assumed that gene duplications would be rare in a plant with a small genome. Additional variations in mutant screens were developed over the years, as described by Page and Grossniklaus (2002). These included screens for genetic enhancers and suppressors of a specific mutant phenotype and the use of reporter lines to screen for altered reporter expression, as demonstrated for abiotic stress with a luciferase construct (Ishitani *et al.*, 1997). The most significant advance in the design of mutant screens resulted from development of random T-DNA (and transposon) mutagenesis procedures, which followed the establishment of efficient transformation protocols. One of the most highly publicized examples of how mutants advanced our understanding of plant biology was the pioneering work of Elliot Meyerowitz and colleagues at the California Institute of Technology on a small collection of floral mutants that helped to identify global regulators of floral organ identity in Arabidopsis and beyond (Weigel and Meyerowitz, 1994).

A mutant screen is normally performed with an inbred line that represents the reference (wild-type) genotype. For Arabidopsis, the Columbia (Col) and Landsberg *erecta* (L*er*) accessions, most probably derived (Rédei, 1992) from the Landsberg accession collected in the Landsberg an der Warthe (Gorzów Wielkopolski) region of Poland, have long been used. The Wassilewskija (Ws-1) accession was added later for some experiments because it was believed to be more suitable for transformation. All of these genotypes are early flowering, which is convenient for mutagenesis experiments. Sequencing data (Clark *et al.*, 2007; Ossowski *et al.*, 2008) and the analysis of natural variation (Alonso-Blanco *et al.*, 2009) indicate that there is no single wild-type accession for Arabidopsis. However, based on extensive mutant collections, high-quality sequence (AGI, 2000) and microarray (Zimmermann *et al.*, 2004) data, and much physiological and biochemical knowledge, Columbia (Col) is generally viewed as the reference genotype.

THE GENETIC MAP OF ARABIDOPSIS

Genetic linkage maps reveal the estimated orders and positions of genes along the chromosome. Before physical maps based on contigs of cloned DNA segments could be constructed, linkage (recombination) analysis was the principal method used to obtain information on gene locations. Initially, single gene mutants acting as morphological markers were used to construct such genetic maps. The first linkage groups of Arabidopsis were presented at the Göttingen conference for part of chromosome 1 (McKelvie, 1965) and in a journal article for chromosome 2 (Rédei, 1965). At Göttingen, Rédei also described a number of markers that defined six distinct linkage groups. These linkage groups formed the basis for the chromosome nomenclature used nowadays for Arabidopsis. When it was later realized that linkage groups 1 and 4 were both on chromosome 1, linkage group 6 was renamed chromosome 4 (Koornneef and van der Veen, 1983). Surprisingly, hardly any progress was reported on linkage analysis between 1965 and 1983. A post-doctoral fellowship application submitted in 1978 by Meinke and Rédei to enhance the genetic map using embryo-lethal mutants as genetic markers was not funded. Eventually, a complete genetic map covering all five chromosomes with 76 markers was published (Koornneef *et al.*, 1983). Further evidence that not everyone believed a linkage map was relevant to the study of Arabidopsis can be shown by the difficulty encountered in publishing this map for what one reviewer commented was a questionable genetic model (Somerville and Koornneef, 2002).

Trisomic lines with a distinctive phenotype caused by an extra copy of a single chromosome were useful in the initial

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assignment of linkage groups to cytological chromosomes. The methods involved have been reviewed elsewhere (Koornneef *et al.*, 2003). Telotrisomic lines with just one chromosome arm duplicated allowed the assignment of morphological markers to chromosome arms and thereby aided in locating some of the centromeres (Koornneef, 1983). Later on, these positions were refined by tetrad analysis using the *quartet* mutant, which enables all four products of a single meiosis to be identified (Copenhaver *et al.*, 1998). Because of the small size of the Arabidopsis genome, individual chromosomes do not exhibit the cytological details that often proved useful in cytogenetic studies with crop plants (Koornneef *et al.*, 2003). However, by combining pachytene chromosomes, which appear longer than mitotic chromosomes and have distinct heterochromatic and euchromatic regions, with sensitive *in situ* hybridization methods, Arabidopsis chromosomes finally became amenable to cytogenetic analysis (Fransz *et al.*, 1998), leading to a number of advances such as the discovery of chromosome inversions in some accessions (Fransz *et al.*, 2000).

The introduction of molecular (restriction fragment length polymorphism, RFLP) markers to the field of genetics in the 1980s made it important to generate maps of such markers in Arabidopsis (Chang *et al.*, 1988) and to integrate them with the classical map (Hauge *et al.*, 1993). Subsequent advances in marker development included the analysis of microsatellite (Bell and Ecker, 1994) and cleaved amplified polymorphic sequence (CAPS; Konieczny and Ausubel, 1993) markers. Although these initial maps were all based on limited F_2 and F_3 populations, it soon became important to generate immortal mapping populations involving recombinant inbred lines (Rils). The first of these was published by Reiter *et al.* (1992). An especially important resource was the L*er* × Col Ril population (Lister and Dean, 1993), which was widely used to link markers from the physical map being constructed in the 1990s (Meinke *et al.*, 1998) to the genetic map. Additional Ril populations were later developed to investigate natural variation and to incorporate other marker types such as amplified fragment length polymorphisms (AFLPs; Alonso-Blanco *et al.*, 1998). Mapping is no longer used in Arabidopsis to establish the definitive orders of genes, which is based instead on the genome sequence. However, mapping is still needed to locate mutants on the genetic map as a prerequisite for map-based cloning (Lukowitz *et al.*, 2000). The latest developments in marker technology include single nucleotide polymorphism assays based on comparative sequencing of Arabidopsis accessions (Nordborg *et al.*, 2005; Clark *et al.*, 2007) and single feature polymorphisms based on microarrays (Borevitz *et al.*, 2007). Deep sequencing will bring a fresh perspective to mapping because sequence comparisons between mutant and wild-type plants should indicate the positions of mutations involved (Lister *et al.*, 2009; Schneeberger *et al.*, 2009).

Ultimately, the classical genetic map of Arabidopsis will be replaced by a sequence-based map of genes with mutant phenotypes. An initial effort along these lines was published several years ago (Meinke *et al.*, 2003). Efforts to update this map and establish a comprehensive dataset of all known Arabidopsis genes with a loss-of-function phenotype are ongoing. In the meantime, we should note that all but 12 of the 76 morphological markers included on the original genetic map of Koornneef *et al.* (1983) have been cloned over the past 25 years (Meinke *et al.*, 2009). The total number of morphological markers included on the updated classical genetic map, which is limited to mutants mapped in relation to each other, stands at 335 (Meinke *et al.*, 2009). The most common markers on this map are *EMB* genes required for normal embryo development (Franzmann *et al.*, 1995). Although many of these genes remain to be cloned, recent progress was made by aligning the genetic and physical maps and performing genetic complementation tests between mapped (but not cloned) and cloned (but not mapped) mutants with similar map locations and phenotypes (Meinke *et al.*, 2009).

NATURAL VARIATION

Variation in physiological traits among natural accessions was one reason why Laibach (1943) first promoted work on Arabidopsis. Sixty-five years later, this research potential is starting to be realized. At first, natural variation controlled by multiple genes and influenced by environmental factors was resistant to molecular dissection. The initial breakthroughs came in the 1990s with the cloning of monogenic disease resistance genes, which had a simple inheritance pattern (Slusarenko and Schlaich, 2003). Subsequent developments in quantitative genetics enabled the identification of genomic regions of interest for complex traits by association of specific trait values with segregating molecular markers known as quantitative trait loci (QTLs). This eventually led to cloning of the underlying genes (quantitative trait genes, QTGs). The steps involved (Alonso-Blanco and Koornneef, 2000) include confirmation and validation of QTLs in near-isogenic lines (Nils) followed by fine-mapping and complementation. In comparison with mutant approaches, where sequencing the region encompassing the mapped locus often identifies the target gene of interest, sequencing a QTL region does not immediately point to the causal locus because of the high degree of polymorphism involved (Clark *et al.*, 2007). Despite these complications, a large number of QTGs have been identified in Arabidopsis (Alonso-Blanco *et al.*, 2009). Research on natural variation can also lead to the identification of functional alleles of genes already mutated in laboratory accessions. One example is *FRI*, a major gene in the control of flowering time, which is mutated both in Col and L*er* but was identified in late-flowering natural accessions (Johanson *et al.*, 2000). Even when genes are not mutated in the lab accessions, strong alleles

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present in natural accessions may lead to their cloning on the basis of QTL analysis (Bentsink *et al.*, 2006). The high level of genetic variation between Arabidopsis accessions can be deduced from direct sequence comparisons (Nordborg *et al.*, 2005; Clark *et al.*, 2007). At a single developmental stage, genetic variation for expression of 20% of the genes can be detected (Keurentjes *et al.*, 2007; Kliebenstein, 2009). Some of this variation probably has little effect at the phenotypic level (Fu *et al.*, 2009), in part because plants have mechanisms involving molecular chaperones (Queitsch *et al.*, 2002) that buffer against visible expression of underlying variation.

One disadvantage of biparental mapping populations, which are commonly used to study natural variation, is that only genetic differences between the two parents can be analysed. Furthermore, the resulting map positions of target loci are rather inaccurate. Both problems can be solved by using genome-wide association (GWA) mapping, which exploits variation in a collection of genotypes and can improve the chances of identifying causal polymorphisms (Myles *et al.*, 2009). The high level of homozygosity found in Arabidopsis accessions, in combination with the high density of molecular markers, make it a suitable organism for this approach (Nordborg and Weigel, 2008). In the future, GWA mapping will benefit from improved sequence technologies and ongoing efforts to sequence large numbers of accessions (Weigel and Mott, 2009). One alternative to GWA and biparental populations is multiparent populations, in which linkage (disequilibrium) is higher but additional variation can be screened (Paulo *et al.*, 2008; Kover *et al.*, 2009).

Apart from being a valuable resource for analyzing gene function, natural variation provides an opportunity to study important features of evolutionary ecology at the molecular level. Arabidopsis has not historically been at the centre of this discipline, although the wide geographical distribution of accessions (Hoffmann, 2002), coupled with a full toolbox of molecular resources, make it a suitable model for such studies (Mitchell-Olds and Schmitt, 2006). One example of insights obtained by combining genetic tools, ecological experiments, and modelling studies based on known details of Arabidopsis flowering time control, was recently published (Wilczek *et al.*, 2009). Further advances in this area may require that more Arabidopsis biologists move out of their laboratories and into the field.

Another fundamental question in biology that relates to natural variation concerns the identities of genes and allelic variants that underlie differing features of related species. The considerable variation that exists among different members of the Brassicaceae is a valuable resource that remains to be exploited through comparative studies with Arabidopsis. Two recent examples of success include the analysis of heavy metal accumulation in *Arabidopsis hallerri* (Hanikenne *et al.*, 2008) and the control of flowering in the perennial species *Arabis alpina* (Wang *et al.*, 2009). Recently, experiments have been extended beyond the Brassicaceae, with comparisons of flowering time control between Arabidopsis and selected grasses (Greenup *et al.*, 2009). Genomic tools in Arabidopsis have also enabled the identification of variation that may underlie speciation events (Bikard *et al.*, 2009). The natural variation in Arabidopsis accessions first studied by Laibach has therefore resulted in major advances on multiple fronts.

COMMUNITY INFRASTRUCTURE AND SHARED RESOURCES

Throughout its brief history, the Arabidopsis community has exhibited an admirable level of collegiality and cooperation. There have been scattered disputes and disappointments, but overall these have not defined the field or impeded progress. Most plant biologists seemed to sense that collaboration was central to making Arabidopsis a viable model. And being a 'simple weed' helped at times to minimize conflicts of interest over the practical applications of shared resources. Even when unexpected results were published (Lolle *et al.*, 2005) and later questioned (Peng *et al.*, 2006), cool heads generally prevailed (Gallagher, 2008). At critical points, dedicated individuals (Figure 3) stepped forward to help advance not only their own research interests but the community as a whole. It began in Europe with the Laibach seed collection, later maintained by Röbbelen and Kranz, continued with the annual publication of the AIS, and was evident at the international conferences in Göttingen and Frankfurt. The focus later shifted to the United States, where Chris Somerville and colleagues organized the Michigan State conference and established an electronic newsgroup to help researchers keep in touch. Soon thereafter, Arabidopsis research became truly global, necessitating the formation of a broader organizational structure. This was realized with the establishment of the first Multinational Arabidopsis Steering Committee (MASC), which included members from several different countries and continents: Marc van Montagu (Belgium), Jim Peacock (Australia), Caroline Dean and Dick Flavell (United Kingdom), Howard Goodman, Elliot Meyerowitz, and Chris Somerville (United States), Maarten Koornneef (the Netherlands), and Yoshiro Shimura and Kiyotaka Okada (Japan). National steering committees were formed as well, most notably the North American Arabidopsis Steering Committee (NAASC), to facilitate interactions with national funding agencies. These oversight committees played an important role in organizing the community, establishing stock centres and databases, identifying shared resources that still needed to be developed, proclaiming the goal of sequencing the genome by the end of the millennium, and keeping those sequencing efforts on track (Somerville and Koornneef, 2002).

Stock centres for seeds and molecular biology materials were established on two continents to serve investigators

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Figure 3. Representative contributors to community infrastructure and shared resources. Left to right; top: Randy Scholl (ABRC) and Eva Huala (TAIR); middle: Machi Dilworth (NSF) and Sean May (NASC); bottom: Joe Ecker (Functional genomics tools) and Kazuo Shinozaki (Functional genomics tools).

worldwide. The European Arabidopsis Stock Centre (NASC) in Nottingham, founded in 1990 with support from the British government, was headed first by Mary Anderson and Bernard Mulligan and thereafter by Sean May, who continues to serve in this capacity. The US counterpart, the Arabidopsis Biological Resource Center (ABRC), at Ohio State University, which serves Asia and Australia in addition to the Americas, was founded in 1991 with support from the NSF. This resource centre was overseen for 19 years by Randy Scholl, whose contributions to the community were recognized at the Arabidopsis conference in Edinburgh, Scotland (July, 2009). Several individuals contributed to the establishment of DNA facilities, including Jeff Dangl, Keith Davis, and Doreen Ware. In Japan, the Riken BRC Experimental Plant Division set up an important resource centre in 2002 for full-length cDNA clones and specialized collections of plant materials. Factors that played a role in the development and success of these stock centres, and in the creation of common standards for Arabidopsis genetics, have been described elsewhere (Meinke and Koornneef, 1997; Meinke and Scholl, 2003).

Excitement over Arabidopsis research was soon coupled with the realization that someone needed to keep track of the information being generated. At first, it seemed possible to accomplish this through traditional methods. The result was 1300 pages of information assembled into a definitive book on Arabidopsis published by Cold Spring Harbor Laboratory Press (Meyerowitz and Somerville, 1994). That was impressive when compared to the 150 pages found in the monograph published 24 years earlier in *Bibliographica Genetica* (Rédei, 1970) but it soon became outdated. With support and guidance from the NSF, an informal committee representing database experts, funding agencies, and the Arabidopsis community, met at Dallas–Fort Worth in 1993 to discuss the future of Arabidopsis genome databases. This began a long series of meetings and discussions about database needs and design, and culminated in a call for proposals some years later to establish a central database suitable for the genomics age. Formal work in database design began with Michael Cherry, who created the first *Arabidopsis thaliana* database (AtDB) and was instrumental in database efforts prior to completion of the genome sequence (Flanders *et al.*, 1998). Later on, it was a group headed by Sue Rhee and Chris Somerville at the Carnegie Institute of Plant Biology in Stanford, CA, in collaboration with database experts at the National Center for Genomic Research (NCGR) in Santa Fe, NM, that was charged with compiling all of the known information on Arabidopsis into one central location, which eventually became known as The Arabidopsis Information Resource (TAIR) at http://www.arabidopsis.org/ (Rhee *et al.*, 2003). Through the continued efforts of Eva Huala and dedicated support personnel, this central database remains a focal point for Arabidopsis research.

The usefulness of knockout mutants generated by T-DNA insertion mutagenesis was demonstrated 20 years ago with the cloning of *AG* (Yanofsky *et al.*, 1990) and *GL1* (Herman and Marks, 1989). Additional insertion mutants, combined sometimes with transposable elements from maize (Aarts *et al.*, 1993), were generated by tissue culture methods (Koncz *et al.*, 1989) and seed transformation (Feldmann, 1991). Efficient procedures were also developed to recover genomic sequences flanking insertion sites (Liu *et al.*, 1995). This led to the realization that large collections of insertion mutants, when combined with public seed stocks and flanking sequence information, could be invaluable tools for reverse genetics. Several groups contributed over the next 15 years to make this dream a reality, including Joe Ecker at the Salk Institute (Alonso *et al.*, 2003), Michel Caboche at INRA in France (Samson *et al.*, 2002), Michael Sussman at the University of Wisconsin (Sussman *et al.*, 2000), Csaba Koncz (Szabados *et al.*, 2002) and Bernd Weisshaar (Rosso *et al.*, 2003) in Germany, Kazuo Shinozaki in Japan (Kuromori *et al.*, 2004), and Syngenta, a multinational company with research facilities in California and North Carolina, which eventually donated seeds and sequence information for two distinct populations (McElver *et al.*, 2001; Sessions *et al.*, 2002). Additional technologies for generating loss-of-function phenotypes such as RNAi and miRNA (Schwab *et al.*, 2006) have also

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become available. Tilling methods (Colbert *et al.*, 2001) were developed to combine EMS mutagenesis with sequence information to find mutants of specific target genes not represented in knockout collections. Random insertion libraries have also been generated using activation tagging (Weigel *et al.*, 2000; Marsch-Martinez *et al.*, 2002) for dominant mutants, and promoterless reporter constructs for selection of insertions at desired intragenic locations coupled with visualization of expression patterns (Sundaresan *et al.*, 1995). In keeping with long-standing policies, these materials have been made widely available to encourage future advances.

THE GENOME SEQUENCE AND BEYOND

Many individuals played a role in the planning, sequencing, and bioinformatics phases of the Arabidopsis genome project (Somerville and Koornneef, 2002). A meeting held at the NSF in 1989 led to a report calling for a wide range of research initiatives and a completed genome sequence by the year 2000. This NSF report and subsequent annual publications describing major goals and accomplishments of the Arabidopsis research community can be accessed through TAIR. What seemed like a risky proclamation at first, given the modest portfolio of sequencing accomplishments at the time, turned out in retrospect to be a milestone in plant biology. Dozens of individuals contributed over the ensuing decade to genome sequencing efforts on three continents. These efforts were spearheaded by several key participants: Michael Bevan, for a European consortium involving multiple countries, Francis Quetier at Genoscope in France, Satoshi Tabata at the Kazusa DNA Research Institute in Japan, and three groups in the United States: (i) Joe Ecker (Salk Institute), Ron Davis (Stanford), and Sakis Theologis (USDA Plant Gene Expression Center in California); (ii) Rob Martienssen and Dick McCombie (Cold Spring Harbor Laboratory) in collaboration with Richard Wilson (Washington University, St Louis); and (iii) Steve Rounsley and Craig Venter at The Institute for Genome Research (TIGR). Informatics teams at multiple locations were also heavily involved with genome annotation efforts, which were coordinated by Klaus Mayer at the Munich Information Centre for Protein Sequences (MIPS) in Germany. When the combined results were published (AGI, 2000) and released to the press in December, 2000, on schedule and within budget, the plant biology community experienced a rare moment of distinction. Public attention in the United States, however, was focused more on the Supreme Court decision about the contested presidential election, which was released the afternoon before the Arabidopsis news conference in Washington, DC.

Realizing that a sequenced genome was of limited use without additional functional details, workshops were scheduled to put forth a plan for the next phase of Arabidopsis research. In the United States, these workshops, along with critical support from the NSF, resulted in the Arabidopsis 2010 project, a vision to characterize the function of each gene by the year 2010 (Chory *et al.*, 2000). Related efforts were advanced in other countries, with progress once again noted in annual reports of the Multinational Arabidopsis Steering Committee. Some of the scientific achievements made possible by those combined efforts are celebrated in this special issue. Further community resources, including full-length cDNAs, knockout collections for reverse genetics, and microarray chips and datasets, to mention just a few, were also developed to support the pending 'omics' revolution that fundamentally changed the nature of plant research. With a centralized database, stock centres, and internet resources to disseminate information and materials worldwide, it was not just the large laboratories at leading research institutions that benefited. Everyone finally had access to the information and resources needed to advance diverse research interests. Of course, much still remains to be done, both in terms of resource development and hypothesis-driven research, but the first Arabidopsis seeds planted years ago have without question brought about a plentiful harvest.

CONCLUSIONS AND FUTURE PROSPECTS

Thirty years ago, when dramatic advances in molecular genetics fundamentally changed the landscape of biology, it was not obvious that plant science would play a central role in the approaching revolution. Plant genomes were large and complex, life cycles were long, and most of the favoured genetic models at the time were difficult to transform. Even the future of plant genetics as a discipline was uncertain, despite an illustrious history that included well-known figures such as Mendel and McClintock. Ultimately, it was a combination of factors, including the choice of Arabidopsis as a plant model, advances in *Agrobacterium*-mediated transformation, the influx of talented and collaborative individuals into plant biology, and increased funding to support experimental breakthroughs that enabled plant biologists to remain at the forefront of modern biology. Discipline integration in plant biology was finally realized, with significant accomplishments that extended far beyond a simple weed, including applications to human health (Jones *et al.*, 2008). Genetic variants have remained at the centre of Arabidopsis research throughout this time, along with improved methods in cell and molecular biology. Computational techniques, including modelling at many different levels, have also become important in recent years (Prusinkiewicz and Rolland-Lagan, 2006). Most private companies interested in the practical applications of plant science have grown to appreciate and utilize Arabidopsis as well, especially in the area of gene discovery (Gutterson and Zhang, 2004; Century *et al.*, 2008). The future of Arabidopsis research should indeed look bright, with a well-established model organism providing the foundation for continued

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breakthroughs in our understanding of how plants work and the possibility that further advances in regulating plant growth and development might soon enable plant breeding by design to become a reality.

Yet despite this impressive record of accomplishments and the remarkable path that Arabidopsis helped to pave, there is reason to be concerned about the future. The vast majority of people worldwide have never heard of Arabidopsis and have no idea what role it should continue to play in improving the lives of ordinary people. This is painfully obvious to anyone who ventures out in public wearing one of the Arabidopsis T-shirts distributed at past conferences. Education and outreach efforts notwithstanding, there is a considerable amount of work that remains to be done in educating those outside of the plant science community about what has been gained by focusing research efforts on a single model organism. The articles found elsewhere in this special issue of *The Plant Journal* should provide much of the detail required for such an education campaign. But new approaches may be needed to make connections between Arabidopsis research programmes and their practical benefits more relevant to the average person.

Even within the community of research biologists, there are troubling signs that not everyone agrees with the premise that Arabidopsis should continue to attract special attention and funding. Major grants used to support critical databases and other shared resources have in some cases been curtailed and funding programmes used to develop Arabidopsis as a model plant eliminated. With initial genome efforts completed, costs of sequencing other genomes reduced, and global problems competing for limited resources, questions are being raised again about the special role of Arabidopsis in plant research. Although the desire to analyse a wider spectrum of angiosperm species is clearly justified, the accumulated knowledge base and broad availability of vast community resources for Arabidopsis should continue to make this model organism the focal point of plant biology. Support for Arabidopsis must therefore not be abandoned. Future advances in our broad understanding of plant diversity are best enabled by continued advances in the detailed characterization of how a single plant works. That is what the past 25 years of Arabidopsis research have so elegantly demonstrated. Twelve years ago, Gerald Fink began his 'perspectives' contribution to a special issue of *Genetics* devoted to Arabidopsis research with the following statement: 'With this volume, *Genetics* announces that Arabidopsis has joined the Security Council of Model Genetic Organisms. These favoured few form the standard to which all other organisms are compared' (Fink, 1998). We hope that future generations of plant biologists will continue to have the vision and resources needed to keep Arabidopsis engaged as a respected member of this select group of organisms. Because only then will the true potential of Arabidopsis be realized, and its importance measured not just by the total number of research publications, but also by the transformative effects on modern biology that became possible when a single model plant was elevated to special research status.

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REFERENCES

Aarts, M.G.M., Dirkse, W.G., Stiekema, W.J. and Pereira, A. (1993) Transposon tagging of a male-sterility gene in Arabidopsis. *Nature*, **363**, 715–717.

AGI. (2000) Analysis of the genome sequence of the flowering plant *Arabidopsis thaliana*. *Nature*, **408**, 796–815.

Alonso, J.M., Stepanova, A.N., Leisse, T.J. *et al.* (2003) Genome-wide insertional mutagenesis of *Arabidopsis thaliana*. *Science*, **301**, 653–657.

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

Alonso-Blanco, C., Peeters, A.J.M., Koornneef, M., Lister, C., Dean, C., van den Bosch, N., Pot, J. and Kuiper, M.T.R. (1998) Development of an AFLP based linkage map of Ler, Col and Cvi *Arabidopsis thaliana* ecotypes and construction of a Ler/Cvi recombinant inbred line population. *Plant J.* **14**, 259–271.

Alonso-Blanco, C., Aarts, M.G.M., Bentsink, L., Keurentjes, J.J.B., Reymond, M., Vreugdenhil, D. and Koornneef, M. (2009) What has natural variation taught us about plant development, physiology, and adaptation? *Plant Cell*, **21**, 1877–1896.

Bechtold, N., Ellis, J. and Pelletier, G. (1993) In-planta Agrobacterium-mediated gene-transfer by infiltration of adult *Arabidopsis thaliana* plants. *C. R. Acad. Sci. III*, **316**, 1194–1199.

Bell, C.J. and Ecker, J.R. (1994) Assignment of 30 microsatellite loci to the linkage map of Arabidopsis. *Genomics*, **19**, 137–144.

Bentsink, L., Jowett, J., Hanhart, C.J. and Koornneef, M. (2006) Cloning of *DOG1*, a quantitative trait locus controlling seed dormancy in Arabidopsis. *Proc. Natl Acad. Sci. USA*, **103**, 17042–17047.

Bikard, D., Patel, D., Le Metté, C., Giorgi, V., Camilleri, C., Bennett, M.J. and Loudet, O. (2009) Divergent evolution of duplicate genes leads to genetic incompatibilities within *A. thaliana*. *Science*, **323**, 623–626.

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.

Bleecker, A.B., Estelle, M.A., Somerville, C. and Kende, H. (1988) Insensitivity to ethylene conferred by a dominant mutation in *Arabidopsis thaliana*. *Science*, **241**, 1086–1089.

Borevitz, J.O., Hazen, S.P., Michael, T.P. *et al.* (2007) Genome-wide patterns of single-feature polymorphism in *Arabidopsis thaliana*. *Proc. Natl Acad. Sci. USA*, **104**, 12057–12062.

Century, K., Reuber, T.L. and Ratcliffe, O.J. (2008) Regulating the regulators: the future prospects for transcription-factor-based agricultural biotechnology products. *Plant Physiol.* **147**, 20–29.

Chang, C. and Meyerowitz, E.M. (1986) Molecular cloning and DNA sequence of the *Arabidopsis thaliana* alcohol dehydrogenase gene. *Proc. Natl Acad. Sci. USA*, **83**, 1408–1412.

Chang, C., Bowman, J.L., Dejohn, A.W., Lander, E.S. and Meyerowitz, E.M. (1988) Restriction Fragment Length Polymorphism linkage map for *Arabidopsis thaliana*. *Proc. Natl Acad. Sci. USA*, **85**, 6856–6860.

Chory, J., Ecker, J.R., Briggs, S. *et al.* (2000) National Science Foundation-sponsored workshop report: ''The 2010 Project'' – Functional genomics and the virtual plant. A blueprint for understanding how plants are built and how to improve them. *Plant Physiol.* **123**, 423–425.

Clark, R.M., Schweikert, G., Toomajian, C. *et al.* (2007) Common sequence polymorphisms shaping genetic diversity in *Arabidopsis thaliana*. *Science*, **317**, 338–342.

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Clough, S.J. and Bent, A.F. (1998) Floral dip: a simplified method for Agrobacterium-mediated transformation of *Arabidopsis thaliana*. *Plant J.* **16**, 735–743.

Colbert, T., Till, B.J., Tompa, R., Reynolds, S., Steine, M.N., Yeung, A.T., McCallum, C.M., Comai, L. and Henikoff, S. (2001) High-throughput screening for induced point mutations. *Plant Physiol.* **126**, 480–484.

Copenhaver, G.P., Browne, W.E. and Preuss, D. (1998) Assaying genome-wide recombination and centromere functions with Arabidopsis tetrads. *Proc. Natl Acad. Sci. USA*, **95**, 247–252.

Cutler, S.R., Ehrhardt, D.W., Griffitts, J.S. and Somerville, C.R. (2000) Random GFP::cDNA fusions enable visualization of subcellular structures in cells of Arabidopsis at a high frequency. *Proc. Natl Acad. Sci. USA*, **97**, 3718–3723.

Dean, C. (2001) A science career on two continents. *Plant Physiol.* **127**, 4–5.

Endersby, J. (2007) *A Guinea Pig's History of Biology*. London: William Heinemann.

Estelle, M.A. and Somerville, C.R. (1986) The mutants of Arabidopsis. *Trends Genet.* **2**, 89–93.

Feenstra, W.J. (1964) Isolation of nutritional mutants in *Arabidopsis thaliana*. *Genetica*, **35**, 259–269.

Feldmann, K.A. (1991) T-DNA insertion mutagenesis in Arabidopsis: mutational spectrum. *Plant J.* **1**, 71–82.

Feldmann, K.A. and Marks, M.D. (1987) Agrobacterium-mediated transformation of germinating seeds of *Arabidopsis thaliana*: a non-tissue culture approach. *Mol. Gen. Genet.* **208**, 1–9.

Feldmann, K.A., Marks, M.D., Christianson, M.L. and Quatrano, R.S. (1989) A dwarf mutant of Arabidopsis generated by T-DNA insertion mutagenesis. *Science*, **243**, 1351–1354.

Fink, G.R. (1998) Anatomy of a revolution. *Genetics*, **149**, 473–477.

Flanders, D.J., Weng, S., Petel, F.X. and Cherry, J.M. (1998) AtDB, the *Arabidopsis thaliana* database, and graphical-web-display of progress by the Arabidopsis Genome Initiative. *Nucleic Acids Res.* **26**, 80–84.

Fransz, P., Armstrong, S., Alonso-Blanco, C., Fischer, T.C., Torres-Ruiz, R.A. and Jones, G. (1998) Cytogenetics for the model system *Arabidopsis thaliana*. *Plant J.* **13**, 867–876.

Fransz, P.F., Armstrong, S., de Jong, J.H., Parnell, L.D., van Drunen, C., Dean, C., Zabel, P., Bisseling, T. and Jones, G.H. (2000) Integrated cytogenetic map of chromosome arm 4S of *A. thaliana*: structural organization of heterochromatic knob and centromere region. *Cell*, **100**, 367–376.

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.

Fu, J., Keurentjes, J.J.B., Bouwmeester, H. *et al.* (2009) System-wide molecular evidence for phenotypic buffering in Arabidopsis. *Nat. Genet.* **41**, 166–167.

Gallagher, R. (2008) Cool heads and hothead. Model behavior on a model organism. *The Scientist*, **22**, 13.

Greenup, A., Peacock, W.J., Dennis, E.S. and Trevaskis, B. (2009) The molecular biology of seasonal flowering-responses in Arabidopsis and the cereals. *Ann. Bot.* **103**, 1165–1172.

Gutterson, N. and Zhang, J.Z. (2004) Genomics applications to biotech traits: a revolution in progress? *Curr. Opin. Plant Biol.* **7**, 226–230.

Hanikenne, M., Talke, I.N., Haydon, M.J., Lanz, C., Nolte, A., Motte, P., Kroymann, J., Weigel, D. and Kramer, U. (2008) Evolution of metal hyperaccumulation required cis-regulatory changes and triplication of HMA4. *Nature*, **453**, 391–395.

Haseloff, J. and Amos, B. (1995) GFP in plants. *Trends Genet.* **11**, 328–329.

Hauge, B.M., Hanley, S.M., Cartinhour, S. *et al.* (1993) An integrated genetic RFLP map of the *Arabidopsis thaliana* genome. *Plant J.* **3**, 745–754.

Herman, P.L. and Marks, M.D. (1989) Trichome development in *Arabidopsis thaliana*. II. Isolation and complementation of the GLABROUS1 gene. *Plant Cell*, **1**, 1051–1055.

Hoffmann, M.H. (2002) Biogeography of *Arabidopsis thaliana* (L.) Heynh. (Brassicaceae). *J. Biogeogr.* **29**, 125–134.

Ishitani, M., Xiong, L.M., Stevenson, B. and Zhu, J.K. (1997) Genetic analysis of osmotic and cold stress signal transduction in Arabidopsis: interactions and convergence of abscisic acid-dependent and abscisic acid-independent pathways. *Plant Cell*, **9**, 1935–1949.

Johanson, U., West, J., Lister, C., Michaels, S., Amasino, R. and Dean, C. (2000) Molecular analysis of *FRIGIDA*, a major determinant of natural variation in Arabidopsis flowering time. *Science*, **290**, 344–347.

Jones, A.M., Chory, J., Dangl, J.L., Estelle, M., Jacobsen, S.E., Meyerowitz, E.M., Nordborg, M. and Weigel, D. (2008) The impact of Arabidopsis on human health: diversifying our portfolio. *Cell*, **133**, 939–943.

Keurentjes, J.J.B., Fu, J.Y., Terpstra, I.R., Garcia, J.M., van den Ackerveken, G., Snoek, L.B., Peeters, A.J.M., Vreugdenhil, D., Koornneef, M. and Jansen, R.C. (2007) Regulatory network construction in Arabidopsis by using genome-wide gene expression quantitative trait loci. *Proc. Natl Acad. Sci. USA*, **104**, 1708–1713.

Kliebenstein, D. (2009) Quantitative genomics: analyzing intraspecific variation using global gene expression polymorphisms or eQTLs. *Annu. Rev. Plant Biol.* **60**, 93–114.

Koncz, C. (2006) Dedication: George P. Rédei. Arabidopsis geneticist and polymath. *Plant Breed. Rev.* **26**, 1–33.

Koncz, C., Martini, N., Mayerhofer, R., Koncz-Kalman, Z., Korber, H., Rédei, G.P. and Schell, J. (1989) High-frequency T-DNA-mediated gene tagging in plants. *Proc. Natl Acad. Sci. USA*, **86**, 8467–8471.

Konieczny, A. and Ausubel, F.M. (1993) A procedure for mapping Arabidopsis mutations using codominant ecotype-specific PCR-based markers. *Plant J.* **4**, 403–410.

Koornneef, M. (1983) The use of telotrisomics for centromere mapping in *Arabidopsis thaliana* (L) Heynh. *Genetica*, **62**, 33–40.

Koornneef, M. and van der Veen, J.H. (1983) Trisomics in *Arabidopsis thaliana* and the location of linkage groups. *Genetica*, **61**, 41–46.

Koornneef, M., Rolff, E. and Spruit, C.J.P. (1980) Genetic control of light-inhibited hypocotyl elongation in *Arabidopsis thaliana* (L) Heynh. *Z. Pflanzenphysiol.* **100**, 147–160.

Koornneef, M., Dellaert, L.W.M. and van der Veen, J.H. (1982) EMS-induced and radiation-induced mutation frequencies at individual loci in *Arabidopsis thaliana* (L) Heynh. *Mutat. Res.* **93**, 109–123.

Koornneef, M., van Eden, J., Hanhart, C.J., Stam, P., Braaksma, F.J. and Feenstra, W.J. (1983) Linkage map of *Arabidopsis thaliana*. *J. Hered.* **74**, 265–272.

Koornneef, M., Reuling, G. and Karssen, C.M. (1984) The isolation and characterization of abscisic-acid insensitive mutants of *Arabidopsis thaliana*. *Physiol. Plant.* **61**, 377–383.

Koornneef, M., Fransz, P. and de Jong, H. (2003) Cytogenetic tools for *Arabidopsis thaliana*. *Chrom. Res.* **11**, 183–194.

Kover, P.X., Valdar, W., Trakalo, J., Scarcelli, N., Ehrenreich, I.M., Purugganan, M.D., Durrant, C. and Mott, R. (2009) A multiparent advanced generation inter-cross to fine-map quantitative traits in *Arabidopsis thaliana*. *PLoS Genet.* **5**, e1000551.

Kribben, F.J. (1964) *Arabidopsis thaliana* (L.) Heynh. eine botanische Drosophila. *Naturwiss. Rundsch.* **17**, 139–145.

Kugler, I. (1951) Untersuchungen über das Keimverhalten einiger Rassen von *Arabidopsis thaliana* (L.) Heynh. Ein Beitrag zum Problem der Lichtkeimung. *Beitr. Biol. Pflanz.* **28**, 211–243.

Kuromori, T., Hirayama, T., Kiyosue, Y., Takabe, H., Mizukado, S., Sakurai, T., Akiyama, K., Kamiya, A., Ito, T. and Shinozaki, K. (2004) A collection of 11 800 single-copy Ds transposon insertion lines in Arabidopsis. *Plant J.* **37**, 897–905.

Laibach, F. (1907) Zur Frage nach der individualität der Chromosomen im Pflanzenreich. *Beit. Bot. Zentralbl.* **22**, 191–210.

Laibach, F. (1943) *Arabidopsis thaliana* (L.) Heynh. Als Objekt für genetische und entwicklungsphysiologische Untersuchungen. *Bot. Arch.* **44**, 439–455.

Laibach, F. (1951) Summer- and winter-annual races of *A. thaliana*. A contribution to the etiology of flower development. *Beitr. Biol. Pflanzen*, **28**, 173–210.

Langridge, J. (1955) Biochemical mutations in the crucifer *Arabidopsis thaliana* (L) Heynh. *Nature*, **176**, 260–261.

Ledoux, L., Huart, R. and Jacobs, M. (1974) DNA-mediated genetic correction of thiamineless *Arabidopsis thaliana*. *Nature*, **249**, 17–21.

Leonelli, S. (2007) Arabidopsis, the botanical Drosophila: from mouse cress to model organism. *Endeavour*, **31**, 34–38.

Leutwiler, L.S., Houghevans, B.R. and Meyerowitz, E.M. (1984) The DNA of *Arabidopsis thaliana*. *Mol. Gen. Genet.* **194**, 15–23.

Li, S.L. and Rédei, G.P. (1969) Thiamine mutants of the crucifer, Arabidopsis. *Biochem. Genet.* **3**, 163–170.

van Lijsebettens, M. and van Montagu, M. (2005) Historical perspectives on plant developmental biology. *Int. J. Dev. Biol.* **49**, 453–465.

Lister, C. and Dean, C. (1993) Recombinant inbred lines for mapping RFLP and phenotypic markers in *Arabidopsis thaliana*. *Plant J.* **4**, 745–750.

Lister, R., Gregory, B.D. and Ecker, J.R. (2009) Next is now: new technologies for sequencing of genomes, transcriptomes, and beyond. *Curr. Opin. Plant Biol.* **12**, 107–118.

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Liu, Y.G., Mitsukawa, N., Oosumi, T. and Whittier, R.F. (1995) Efficient isolation and mapping of *Arabidopsis thaliana* T-DNA insert junctions by thermal asymmetric interlaced PCR. *Plant J.* **8**, 457–463.

Lloyd, A.M., Barnason, A.R., Rogers, S.G., Byrne, M.C., Fraley, R.T. and Horsch, R.B. (1986) Transformation of *Arabidopsis thaliana* with *Agrobacterium tumefaciens*. *Science*, **234**, 464–466.

Lolle, S.J., Victor, J.L., Young, J.M. and Pruitt, R.E. (2005) Genome-wide non-Mendelian inheritance of extra-genomic information in Arabidopsis. *Nature*, **434**, 505–509.

Lukowitz, W., Gillmor, C.S. and Scheible, W.R. (2000) Positional cloning in Arabidopsis. Why it feels good to have a genome initiative working for you. *Plant Physiol.* **123**, 795–805.

Marsch-Martinez, N., Greco, R., Van Arkel, G., Herrera-Estrella, L. and Pereira, A. (2002) Activation tagging using the En-I maize transposon system in Arabidopsis. *Plant Physiol.* **129**, 1544–1556.

McElver, J., Tzafrir, I., Aux, G. *et al.* (2001) Insertional mutagenesis of genes required for seed development in *Arabidopsis thaliana*. *Genetics*, **159**, 1751–1763.

McKelvie, A.D. (1965) Preliminary data on linkage groups in Arabidopsis. In *Arabidopsis Research* (Röbbelen, G., ed). University of Göttingen, Germany: Gerd Wasmund Co, pp. 79–84.

Meinke, D. and Koornneef, M. (1997) Community standards for Arabidopsis genetics. *Plant J.* **12**, 247–253.

Meinke, D. and Scholl, R. (2003) The preservation of plant genetic resources. Experiences with Arabidopsis. *Plant Physiol.* **133**, 1046–1050.

Meinke, D.W. and Sussex, I.M. (1979a) Embryo-lethal mutants of *Arabidopsis thaliana*: a model system for genetic analysis of plant embryo development. *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.

Meinke, D.W., Cherry, J.M., Dean, C., Rounsley, S.D. and Koornneef, M. (1998) *Arabidopsis thaliana*: a model plant for genome analysis. *Science*, **282**, 662–682.

Meinke, D.W., Meinke, L.K., Showalter, T.C., Schissel, A.M., Mueller, L.A. and Tzafrir, I. (2003) A sequence-based map of Arabidopsis genes with mutant phenotypes. *Plant Physiol.* **131**, 409–418.

Meinke, D., Sweeney, C. and Muralla, R. (2009) Integrating the genetic and physical maps of *Arabidopsis thaliana:* identification of mapped alleles of cloned essential (*EMB*) genes. *PLoS ONE*, **4**, e7386.

Meyerowitz, E.M. (1987) *Arabidopsis thaliana*. *Annu. Rev. Genet.* **21**, 93–111.

Meyerowitz, E.M. (1989) Arabidopsis, a useful weed. *Cell*, **56**, 263–269.

Meyerowitz, E.M. (2001) Prehistory and history of Arabidopsis research. *Plant Physiol.* **125**, 15–19.

Meyerowitz, E.M. and Pruitt, R.E. (1985) *Arabidopsis thaliana* and plant molecular genetics. *Science*, **229**, 1214–1218.

Meyerowitz, E.M. and Somerville, C.R. (1994) *Arabidopsis*. New York: Cold Spring Harbor Laboratory Press.

Mitchell-Olds, T. and Schmitt, J. (2006) Genetic mechanisms and evolutionary significance of natural variation in Arabidopsis. *Nature*, **441**, 947–952.

Moffat, A.S. (1992) Gene research flowers in *Arabidopsis thaliana*. *Science*, **258**, 1580–1581.

Müller, A.J. (1961) Mutationen mit Embryonaler Manifestation bei *Arabidopsis thaliana*. *Naturwissenschaften*, **48**, 579.

Müller, A.J. (1963) Embryonentest zum Nachweis rezessiver Letalfaktoren bei *Arabidopsis thaliana*. *Biol. Zentralbl.* **82**, 133–163.

Myles, S., Peiffer, J., Brown, P.J., Ersoz, E.S., Zhang, Z., Costich, D.E. and Buckler, E.S. (2009) Association mapping: critical considerations shift from genotyping to experimental design. *Plant Cell*, **21**, 2194–2202.

Nordborg, M. and Weigel, D. (2008) Next-generation genetics in plants. *Nature*, **456**, 720–723.

Nordborg, M., Hu, T.T., Ishino, Y. *et al.* (2005) The pattern of polymorphism in *Arabidopsis thaliana*. *PLoS Biol.* **3**, 1289–1299.

North, G. (1985) A plant joins the pantheon at last? *Nature*, **315**, 366–367.

Oostindier-Braaksma, F.J. and Feenstra, W.J. (1973) Isolation and characterization of chlorate-resistant mutants of *Arabidopsis thaliana*. *Mutat. Res.* **19**, 175–185.

Ossowski, S., Schneeberger, K., Clark, R.M., Lanz, C., Warthmann, N. and Weigel, D. (2008) Sequencing of natural strains of *Arabidopsis thaliana* with short reads. *Genome Res.* **18**, 2024–2033.

Page, D.R. and Grossniklaus, U. (2002) The art and design of genetic screens: *Arabidopsis thaliana*. *Nat. Rev. Genet.* **3**, 124–136.

Patrusky, B. (1991) Drosophila botanica (the fruit fly of plant biology). *Mosaic*, **22**, 32–43.

Paulo, M.J., Boer, M., Huang, X.Q., Koornneef, M. and van Eeuwijk, F. (2008) A mixed model QTL analysis for a complex cross population consisting of a half diallel of two-way hybrids in *Arabidopsis thaliana*: analysis of simulated data. *Euphytica* **161**, 107–114.

Peng, P., Chan, S.W., Shah, G.A. and Jacobsen, S.E. (2006) Increased outcrossing in *hothead* mutants. *Nature*, **443**, E8.

Pennisi, E. (2000) Arabidopsis comes of age. *Science*, **290**, 32–35.

Pruitt, R.E., Bowman, J.L. and Grossniklaus, U. (2003) Plant genetics: a decade of integration. *Nat. Genet.* **33**, 294–304.

Prusinkiewicz, P. and Rolland-Lagan, A.G. (2006) Modeling plant morphogenesis. *Curr. Opin. Plant Biol.* **9**, 83–88.

Queitsch, C., Sangster, T.A. and Lindquist, S. (2002) Hsp90 as a capacitor of phenotypic variation. *Nature*, **417**, 618–624.

Rédei, G.P. (1965) Non-Mendelian megagametogenesis in Arabidopsis. *Genetics*, **51**, 857–872.

Rédei, G.P. (1970) *Arabidopsis thaliana* (L.) Heynh. A review of the genetics and biology. *Bibliogr. Genet.* **20**, 1–151.

Rédei, G.P. (1975) Arabidopsis as a genetic tool. *Annu. Rev. Genet.* **9**, 111–127.

Rédei, G.P. (1992) A heuristic glance at the past of Arabidopsis genetics. In *Methods in Arabidopsis Research* (Koncz, C., Chua, N.H. and Schell, J., eds). Singapore: World Scientific, pp. 1–15.

Reiter, R.S., Williams, J.G.K., Feldmann, K.A., Rafalski, J.A., Tingey, S.V. and Scolnik, P.A. (1992) Global and local genome mapping in *Arabidopsis thaliana* by using recombinant inbred lines and random amplified polymorphic DNAs. *Proc. Natl Acad. Sci. USA*, **89**, 1477–1481.

Rhee, S.Y., Beavis, W., Berardini, T.Z. *et al.* (2003) The Arabidopsis Information Resource (TAIR): a model organism database providing a centralized, curated gateway to Arabidopsis biology, research materials and community. *Nucleic Acids Res.* **31**, 224–228.

Röbbelen, G. (1957) Untersuchungen an Strahleninduzierten Blattfarbmutanten von *Arabidopsis thaliana* (L) Heynh. *Z. Abst. Vererbungslehre*, **88**, 189–252.

Rosso, M.G., Li, Y., Strizhov, N., Reiss, B., Dekker, K. and Weisshaar, B. (2003) An *Arabidopsis thaliana* T-DNA mutagenized population (GABI-Kat) for flanking sequence tag-based reverse genetics. *Plant Mol. Biol.* **53**, 247–259.

Samson, F., Brunaud, V., Balzergue, S., Dubreucq, B., Lepiniec, L., Pelletier, G., Caboche, M. and Lecharny, A. (2002) FLAGdb/FST: a database of mapped flanking insertion sites (FSTs) of *Arabidopsis thaliana* T-DNA transformants. *Nucleic Acids Res.* **30**, 94–97.

Schneeberger, K., Ossowski, S., Lanz, C., Juul, T., Petersen, A.H., Nielsen, K.L., Jorgensen, J.E., Weigel, D. and Andersen, S.U. (2009) SHOREmap: simultaneous mapping and mutation identification by deep sequencing. *Nat. Methods* **6**, 550–551.

Schwab, R., Ossowski, S., Riester, M., Warthmann, N. and Weigel, D. (2006) Highly specific gene silencing by artificial microRNAs in Arabidopsis. *Plant Cell*, **18**, 1121–1133.

Sessions, A., Burke, E., Presting, G. *et al.* (2002) A high-throughput Arabidopsis reverse genetics system. *Plant Cell*, **14**, 2985–2994.

Slusarenko, A.J. and Schlaich, N.L. (2003) Downy mildew of *Arabidopsis thaliana* caused by *Hyaloperonospora parasitica* (formerly *Peronospora parasitica*). *Mol. Plant Pathol.* **4**, 159–170.

Somerville, C. (1989) Arabidopsis blooms. *Plant Cell*, **1**, 1131–1135.

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

Somerville, C.R. and Ogren, W.L. (1980) Photorespiration mutants of *Arabidopsis thaliana* deficient in serine-glyoxylate aminotransferase activity. *Proc. Natl Acad. Sci. USA*, **77**, 2684–2687.

Sundaresan, V., Springer, P., Volpe, T., Haward, S., Jones, J.D.G., Dean, C., Ma, H. and Martienssen, R. (1995) Patterns of gene action in plant development revealed by enhancer trap and gene trap transposable elements. *Genes Dev.* **9**, 1797–1810.

Sussman, M.R., Amasino, R.M., Young, J.C., Krysan, P.J. and Austin-Phillips, S. (2000) The Arabidopsis knockout facility at the University of Wisconsin-Madison. *Plant Physiol.* **124**, 1465–1467.

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Szabados, L., Kovacs, I. and Oberschall, A. *et al.* (2002) Distribution of 1000 sequenced T-DNA tags in the Arabidopsis genome. *Plant J.* **32**, 233–242.

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.

Wang, R.H., Farrona, S., Vincent, C., Joecker, A., Schoof, H., Turck, F., Alonso-Blanco, C., Coupland, G. and Albani, M.C. (2009) PEP1 regulates perennial flowering in *Arabis alpina*. *Nature*, **459**, 423–427.

Weigel, D. and Meyerowitz, E.M. (1994) The ABCs of floral homeotic genes. *Cell*, **78**, 203–209.

Weigel, D. and Mott, R. (2009) The 1001 genomes project for *Arabidopsis thaliana*. *Genome Biol.* **10**, 107.

Weigel, D., Ahn, J.H. and Blazquez, M.A. *et al.* (2000) Activation tagging in Arabidopsis. *Plant Physiol.* **122**, 1003–1013.

Wilczek, A.M., Roe, J.L., Knapp, M.C. *et al.* (2009) Effects of genetic perturbation on seasonal life history plasticity. *Science*, **323**, 930–934.

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.

Zimmermann, P., Hirsch-Hoffmann, M., Hennig, L. and Gruissem, W. (2004) GENEVESTIGATOR. Arabidopsis microarray database and analysis toolbox. *Plant Physiol.* **136**, 2621–2632.

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