The true story of the HD-Zip family

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The HD-Zip family of transcription factors is unique to the plant kingdom. These proteins exhibit the singular combination of a homeodomain with a leucine zipper acting as a dimerization motif. They can be classified into four subfamilies, according to a set of distinctive features that include DNA-binding specificities, gene structures, additional common motifs and physiological functions. Some HD-Zip proteins participate in organ and vascular development or meristem maintenance. Others mediate the action of hormones or are involved in responses to environmental conditions. Here, we review recent data for this family of transcription factors from a wide variety of plant species to unravel their crucial role in plant development.

Homeodomains and the homeotic phenomena

A homeobox (HB) encodes a protein domain, the homeodomain (HD), which is a conserved 60-amino acid motif present in transcription factors found in all the eukaryotic organisms. This 60-amino acid sequence folds into a characteristic three-helix structure that is able to interact specifically with DNA. Most HDs are able to bind DNA as monomers with high affinity, through interactions made by helix III (the so-called recognition helix) and a disordered N-terminal arm located beyond helix I. The high degree of conservation of this type of domain among diverse proteins from different kingdoms indicates that this structure is crucial to maintain the HD functionality and that the role played by this domain is vital [1].

The importance of being homeobox but not homeotic

Homeobox genes, first identified in *Drosophila* [2], were named after the homeotic effect, a fundamental developmental reorganization, which is caused by the mutation or ectopic expression of these genes. In animals, overexpression or mutation of homeotic genes generally results in the transformation of one complete body part into another. The HB-containing genes were discovered in plants by Erik Volbrecht and coworkers in 1991 [3]. The maize (*Zea mays*) *KNOTTED1* was identified by transposon tagging and resulted in the isolation of the first plant HB-containing gene. This gene earned its name from its constitutive expression in transgenic plants, which resulted in knotted leaves. Since then, many HB-containing genes have been isolated from a wide variety of plants, including mono- and dicots. They were classified into several families according to specific distinguishing features. Nevertheless, the legacy of the family name (homeobox genes) from the animal kingdom seems rather unsuitable, because none of the known HB-containing genes in plants exert any homeotic effect.

The plant homeodomain superfamily

Since the discovery of *KNOTTED1*, a high number of plant genes encoding HDs have been isolated, but, of those, only a few have been characterized. Members of this superfamily differ not only in the sequence encoding the HD, but also in their size, HD location, association with other domains and in their genes structures. Moreover, HD-containing transcription factors participate in a wide variety of developmental processes. The full description of model plant genomes, such as those of rice (*Oryza sativa*) and *Arabidopsis thaliana*, enabled us to classify the plant HD-containing proteins in six families, according to the distinguishing features mentioned above. These families are homeodomain associated to a leucine zipper (HD-Zip), plant homeodomain associated to a finger domain (PHD finger), Bell (named after the distinctive Bell domain), zinc finger associated to a homeodomain (ZF-HD), Wuschel related homeobox (WOX) and Knotted related homeobox (KNOX). The analysis of the HD sequences from unrelated organisms suggested that some plant HDs are more related to defined HDs from animals and fungi than to HDs from different plant families. Therefore, HDs seem to have diverged before the separation of the branches leading to plants, animals and fungi [4,5].

Figure 1a and Table 1 show a schematic representation of the main structural features and the functionality of the six families constituting the superfamily of plant HDs.

During the past years, our understanding of HD-Zip-encoding genes has dramatically increased. Hence, we devote this article to this specific gene family, which is unique to plants.

The HD-Zip family

Members of the HD-Zip family have a leucine zipper motif (LZ) immediately downstream of the HD. The two motifs are present in transcription factors found in species belonging to other eukaryotic kingdoms, but their association in a single protein is unique to plants [6]. The HD is responsible for the specific binding to DNA, whereas LZ acts as a dimerization motif. HD-Zip proteins bind to DNA as

Glossary

Orthologous genes: genes from different organisms that diverged after a speciation event and exhibit a high homology.
Paralogous genes: genes from the same organism that have arisen by a recent duplication event of a common ancestral gene.

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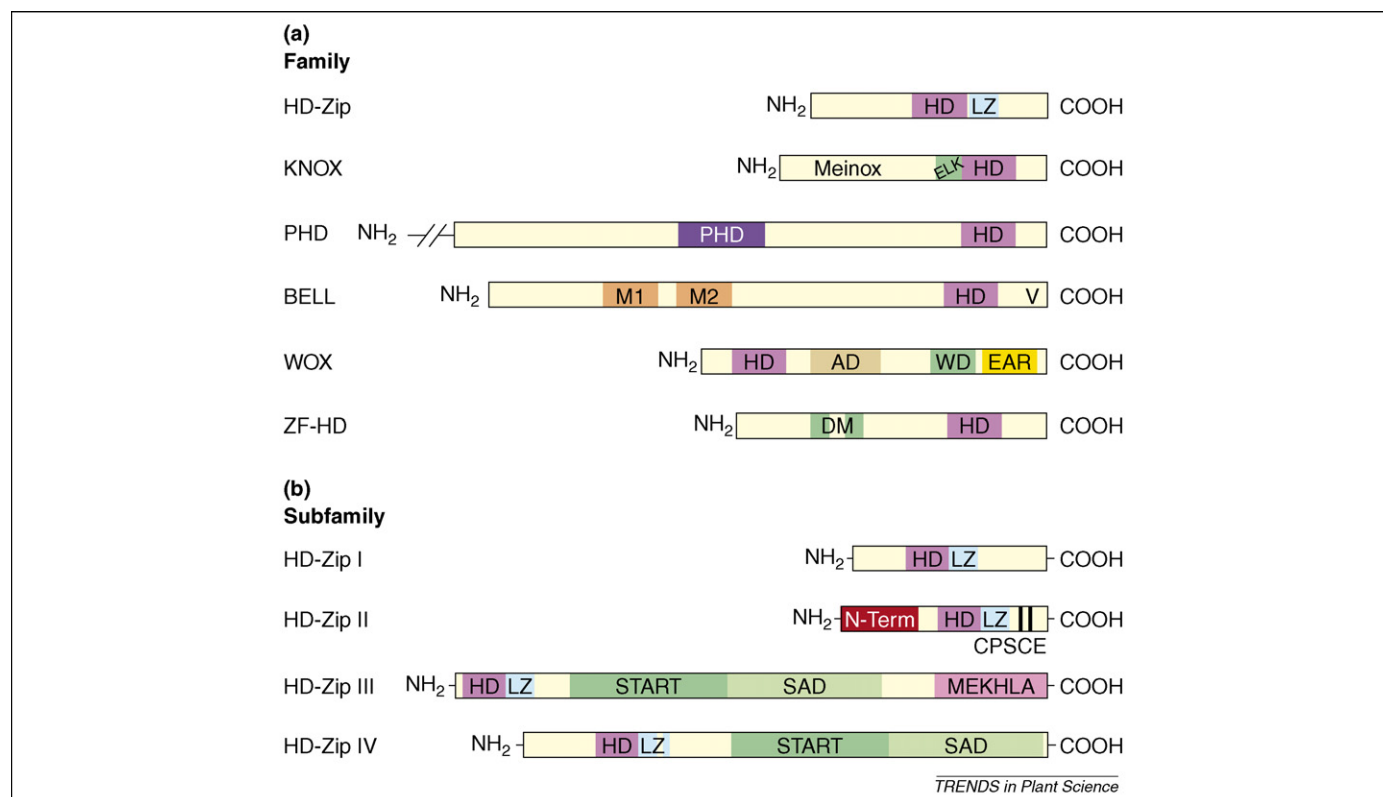Available online 16 August 2007.

Figure 1. **(a)** Schematic representation of the distinctive domains exhibited by each family of HD-containing proteins. Abbreviations: AD, acidic domain; DM, dimerization motif; EAR, amphiphilic repression motif; ELK motif, named after the three conserved amino acids Glu, Leu and Lys in the one letter code; HD, homeodomain; LZ, leucine zipper; M1 and M2 conform the Meinox (association between MEIS and KNOX domains) interaction domain (MID); PHD, plant homeodomain; V, 'VSLTLGL' box; WD, WUS domain. **(b)** Schematic representation of the distinctive features exhibited by each HD-Zip subfamily. Abbreviations: MEKHLA domain, named after the highly conserved amino acids Met, Glu, Lys, His, Leu, Ala; N-term, N-terminus consensus; SAD, START adjacent domain; START, steroidogenic acute regulatory protein-related lipid transfer domain.

dimers, and the absence of LZ absolutely abolishes their binding ability, which indicates that the relative orientation of the monomers, driven by this motif, is crucial for an efficient recognition of DNA.

It is important to note that the existence of an atypical HD-Zip transcription factor [homeodomain leucine zipper encoding gene (HOMEZ)] in vertebrates has been reported recently [7]. However, a sequence comparison between this protein and plant HD-Zip proteins as well as with other LZ-containing proteins indicates that the structure of the so-called LZ in HOMEZ greatly differs from the definition of a typical leucine zipper.

The classification of HD-Zip proteins in four subfamilies is supported by the following four distinguishing characteristics: (i) conservation of the HD-Zip domain that determine DNA-binding specificities, (ii) genes structures, (iii) additional conserved motifs and (iv) functions. Figure 1b shows a schematic representation of the main structural features that characterize each subfamily; in addition, a phylogenetic tree of *Arabidopsis* HD-Zip proteins is shown in Figure 2.

Structural characteristics and target sequences of members of each HD-Zip subfamily

Subfamily I

In Arabidopsis, subfamily I (HD-Zip I) is composed of seventeen members (*ATHB1/HAT5, ATHB3/HAT7, ATHB5– ATHB7, ATHB12, ATHB13, ATHB16, ATHB20–*

Table 1. The six families of plant HD-encoding genes

Family	Functions[a]	Model member gene[b]	Refs
HD-Zip	Response to environmental conditions	ATHB1 (At1g20280)	[9]
	Meristem regulation	ATHB2 (At4g16780)	[39]
	Organ and vascular development	ATHB8 (At1g52150)	[48]
	Hormones action mediation	ATHB10 (At1g79840)	[19]
PHD-finger	Pollen maturation	MMD1 (At1g66170)	[74]
BELL	Maintenance of the shoot apical meristem	BEL1 (At5g41410)	[75]
			[76]
ZF-HD	Regulation of floral development	ATHB25 (At5g65410)	[8]
WOX	Embryogenesis	WUSCHEL (At2g17950)	[77]
	Maintenance of the inner core of the shoot apical meristem		[78]
	Activation of organ identity genes in floral development		
KNOX	Initiation and maintenance of the shoot apical meristem	KNAT1 (At4g08150)	[79]
	Determination of the inflorescence architecture	KNAT6 (At1g23380)	[80]

[a]Physiological events in which HD-encoding genes are involved.
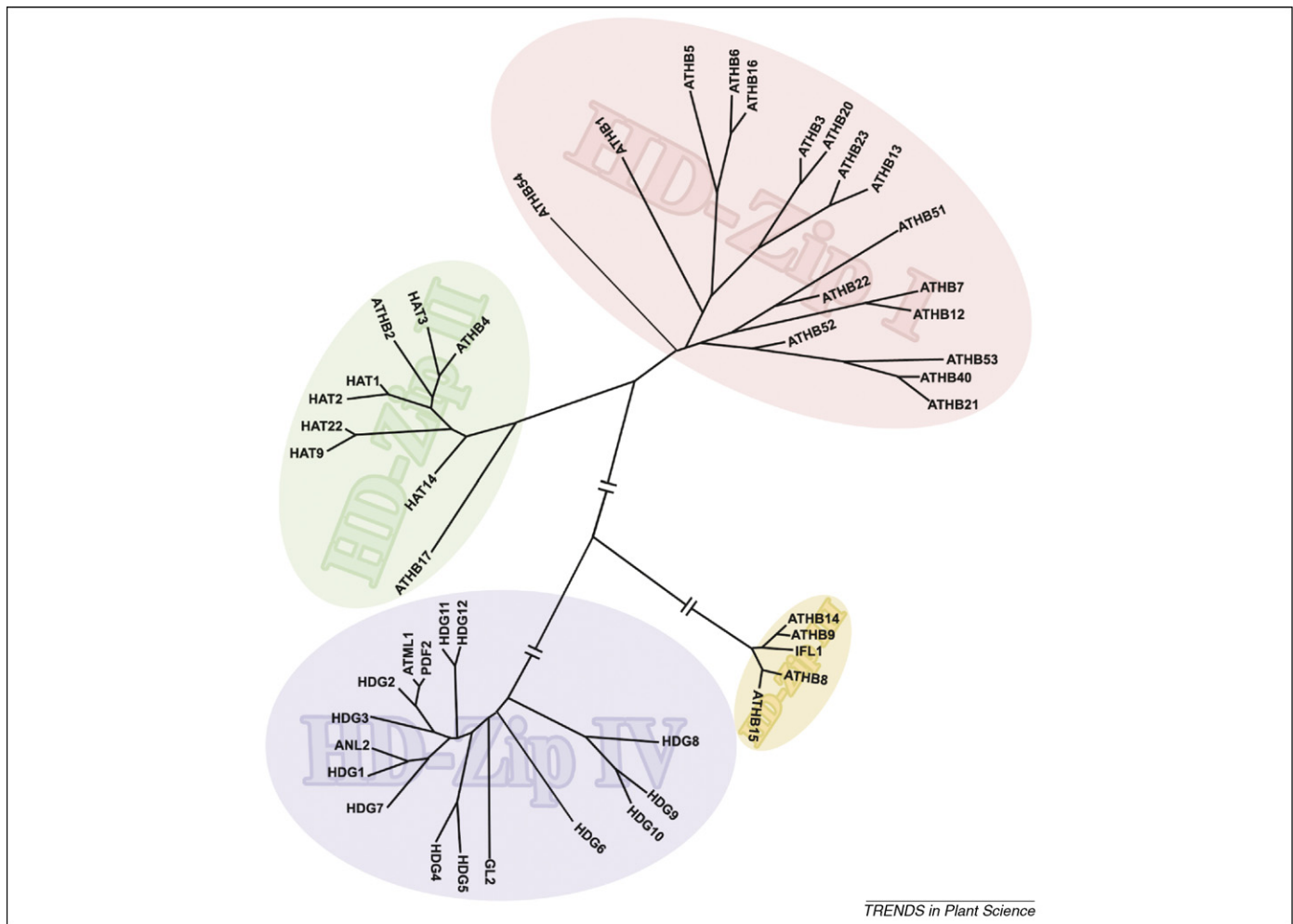[b]Examples of model genes for each function and gene family.

Figure 2. Phylogenetic tree of *Arabidopsis* HD-Zip proteins based on the alignment of sequences by using the program Tree-Puzzle 5.2. The comparison among HD-Zip proteins to construct the phylogenetic tree was carried out with Tree-Puzzle 5.2 [73] loading the available full length protein sequences.

ATHB23, ATHB40, ATHB51– ATHB54). It is worth noting that the names A*THB21–ATHB23* (*At2g18550*, *At2g36610* and *At1g26960*) were also given to three genes belonging to the ZF-HD family (*At2g02540*, *At4g24660* and *At5g39760*) [8], which can lead to confusion. The genes of HD-Zip I have a common intron/exon distribution in agreement with their phylogenetic relationships [9]. The encoded proteins are of ~35 kDa, and exhibit a highly conserved HD, a less conserved LZ and no other similarity [4].

A full *in vitro* description, consisting of PCR-assisted binding site selection and footprinting assays, determined that proteins encoded by HD-Zip I genes form dimers that recognize the pseudopalindromic sequence CAAT(A/T)ATTG [4,10,11]. The only two members that are apparently unable to bind to DNA *in vitro* are *ATHB7* and *ATHB12* [12]. Biochemical *in vitro* analysis performed with a sunflower member of this subfamily indicated that the N-terminal flexible arm plays an important role in the DNA–protein interaction, increasing its affinity without changing the specificity [11]. On the other hand, the inability of HD-Zip proteins to bind DNA in their monomeric form as the majority of HD proteins is determined by the composition of the loop located between helixes I and II [13].

Subfamily II
Subfamily II (HD-Zip II) consists of nine members (*ATHB2 / HAT4*, *ATHB4*, *HAT1–HAT3*, *HAT9*, *HAT14, HAT17,* and *HAT22*) that are of similar size compared to those of subfamily I and exhibit two introns within the HB sequence. They show high conservation in the HD and LZ domains, as well as in two additional motifs: the CPSCE (named after the five conserved amino acids Cys, Pro, Ser, Cys, Glu in the one letter code) adjacent to and downstream of LZ, and an N-terminal consensus sequence. The CPSCE domain is responsible for redox cell state sensing. *In vitro* assays demonstrated that, in an oxidant environment, these transcription factors form high molecular weight multimers through intermolecular Cys–Cys bridges. This kind of macromolecule is probably not transportable to the nucleus to exert its role. This suggests that the redox state of the cell might regulate the activity of HD-Zip II proteins [14].

The proteins encoded by this gene subfamily form dimers that bind to the pseudopalindromic sequence CAAT (C/G)ATTG differing from members of subfamily I only in the recognized nucleotide located in the center of the target site. The binding specificity to the central nucleotide of the pseudopalindromic sequence seems to be conferred in part by amino acids 46 and 56 of helix III (Ala and Trp in HD-Zip

I; Glu and Thr in HD-Zip II), together with a different spatial orientation of the conserved Arg55 in both proteins. Arg55 would be directly responsible for the interaction [15].

Subfamily III

Subfamily III (HD-Zip III) is composed of five members (*ATHB8*, *PHAVOLUTA/ATHB9*, *PHABULOSA/ATHB14*, *CORONA/ATHB15* and *REVOLUTA/IFL1*). The binding domain of this subfamily has four additional amino acids between the HD and LZ domains. Among them, more than a half of the amino acids are conserved and exhibit a common START (steroidogenic acute regulatory protein-related lipid transfer) domain followed by an adjacent conserved region called SAD (START-adjacent domain). Although many START-containing proteins found in the animal kingdom have been well characterized, up to now no lipid ligands have been identified in plants [16]. However, the high conservation of this motif achieved throughout evolution indicates that it is likely to play a significant role in the activity regulation. Additionally, all members have a conserved domain in the C-terminus, called MEKHLA after the goddess of lightning, water and rain. This domain shares significant similarity with the PAS domain, found in many proteins throughout all kingdoms of life and involved in light, oxygen and redox potential sensing [17].

The interaction between these proteins and DNA is less studied than in the case of the other subfamilies. Nonetheless, GTAAT(G/C)ATTAC was determined to be the extended sequence for which *ATHB9* has the highest affinity *in vitro* [18].

Subfamily IV

GL2/subfamily IV (HD-Zip IV) constitutes a large subfamily of genes, composed of 16 members (*GLA-BRA2/ATHB10*, *ATML1*, *ANL2*, *PDF2, HDG1–HDG5, HDG6/FWA* and *HDG7–HDG12*). The proteins encoded by these genes are also of the type HD-Zip–START–SAD, like those of HD-Zip III, although their binding and dimerization structures bear a closer resemblance to the subfamilies I and II. Their most distinguishable features are the presence of a loop in the middle of the LZ domain and the lack of the MEKHLA motif. This group of proteins has also been named HD-Zip GL2 or simply GL2 family after its founding member, the *Arabidopsis* GLABRA2 protein [19].

Proteins belonging to this subfamily show a binding preference for alternative sequences. CATT(A/T)AATG was identified as a preferential target of the sunflower (*Helianthus annuus*) member HAHR1 [20]. On the other hand, the results of binding site selection experiments using HDG7, HDG9, ATML1 and PDF2 recombinant proteins revealed a GCATT(A/T)AATGC consensus sequence. This sequence is overlapping with the L1 box sequence TAAATG(C/T)A recognized *in vitro* by ATML1 [21]. Together, the obtained data reveals that the sequences targeted by HD-Zip IV proteins are all characterized by a TAAA core. This motif is in fact present in a target site of GL2 within the phospholipase Dξ1 gene promoter [22].

Expression and functional studies of the members of each HD-Zip subfamily

Subfamily I

The expression of these genes is regulated by external factors such as drought, extreme temperatures, osmotic stresses and illumination conditions, and is specific to different tissues and organs of the plant. Their role as transcription factors is related to developmental events in response to such environmental conditions, particularly those in which abiotic factors generate stress [23–28].

Different subsets of *Arabidopsis* HD-Zip I genes that bear a close phylogenetic resemblance exhibit a common organ expression pattern and are responsive to the same environmental factors [9].

ATHB1, the first isolated member, acts as a mediator in the leaf cell fate determination [29], whereas *ATHB16* is involved in blue-light perception signaling [30]. Another group of genes was proposed to be involved in ABA-related and abiotic stress responses. Evidence was obtained from expression studies and transgenic plants, indicating that *ATHB7* and *ATHB12* are upregulated and *ATHB5* and *ATHB6* are downregulated by water-deficit conditions and/or externally applied abscisic acid (ABA) [31–36]. Under the effect of these stimuli, HD-Zip I genes behave as developmental and growth regulators. The sunflower *HAHB4* gene, on the other hand, confers drought tolerance to transgenic *Arabidopsis* plants when it is expressed under the control of constitutive or drought-inducible promoters [35,36]. This is the result of its action as an ethylene-mediated senescence repressor and its ability to protect plants from photooxidative stress by downregulating the biogenesis of the photosynthetic machinery [37]. Because the sunflower genome sequence is not available, and taking into consideration that *HAHB4* does not behave like *ATHB7* or *ATHB12*, it is probable that they are not orthologous genes (see Glossary), even if they exhibit high sequence homology and are regulated by the same external factors [23,27,35].

Overexpression of *ATHB3*, *ATHB13*, *ATHB20* or *ATHB23* genes suggests that they are involved in the regulation of cotyledon and leaf development [9,38].

Most of the studies reporting either the overexpression or knocking out of these genes support their postulated role as negative or positive developmental regulators.

Subfamily II

The expression of the genes of this subfamily is generally regulated by illumination conditions in photosynthetic tissues, and their function in plant development is associated with this factor, particularly in the case of the shade avoidance response [39–41].

ATHB2/HAT4 has been described as a negative regulator of paralogous genes (see Glossary). It recognizes its own promoter region and its endogenous expression is repressed in overexpressing transgenic plants [42,43]. Sunflower *HAHB10* bears a close resemblance to *Arabidopsis ATHB2* with regards to structural features; moreover, these two genes show an analogous expression pattern in their natural genomic neighboring. The effect of their overexpression in *Arabidopsis* indicates that they

both act as developmental regulators in response to illumination conditions. [28].

On the other hand, *HAT2,* another member of this subfamily, has been characterized as an auxin-inducible gene in seedlings by DNA microarray screening [44]. Its overexpression produces long hypocotyls, epinastic cotyledons, long petioles and small leaves, which are typical characteristics of auxin-overproducing mutants [45]. The authors concluded that this gene plays opposite roles in the shoot and root tissues in the regulation of auxin-mediated morphogenesis.

Subfamily III

It has been determined that different subsets of genes belonging to the HD-Zip III subfamily are involved in different developmental events, playing overlapping, antagonistic or distinct roles. In general, subfamily III members are well characterized functionally as developmental regulators of the apical meristem, the vascular bundles, auxin transport and the adaxial domains of lateral organs (*REV/FL1, PHB/ATHB9, PHV/ATHB14*), or alternatively as regulators of vascular development (*ATHB8* and *ATHB15*). Their expression patterns have been fully described, and are in complete accordance with the roles they play [46].

Arabidopsis revoluta (rev) plants are the only single mutants of this subfamily that exhibit a phenotype. This is a result of the functional overlap of the five members of this subfamily. The *rev* phenotype shows defects in shoot and leaves development, stem cell specification, vascular development and auxin transport [46–48]. Even though *athb8* mutants do not display any detectable phenotype, the constitutive expression of the *ATHB8* gene results in the overproduction of xylem [49]. Moreover, it has been shown that *ATHB8* regulates the early events of procambial development [50].

PHB, *PHV* and *REV* exhibit overlapping functions during embryogenesis and in the determination of leaf polarity. These three genes are involved in the establishment of apical bilateral symmetry and of the shoot apical meristem. A triple mutant of these genes exhibits seedling lethality. *PHB*, *PHV* and *CNA/ATHB15* play a critic role in meristem regulation, and their action in this pathway turned out to be independent from *REV* and *ATHB8*. On the other hand, *ATHB8* and *CNA* downregulate the action of *REV* in lateral shoot meristems as well as during floral meristem formation. The plant stature is redundantly regulated by each of the five HD-Zip III proteins, although it remains unclear how this is achieved [48].

The ectopic expression of each of these genes could not rescue the phenotype of the singles mutants, suggesting that the HD-Zip III members do not play equivalent roles.

All HD-Zip III genes are both transcriptionally and post-transcriptionally regulated. Their mRNAs are targets of the miRNAs *miR165* and *miR166* [51,52]. In plant lines with high levels of these miRNAs, *PHB*, *PHV* and *CNA* have been shown to be downregulated [53,54]. Accordingly, when their target sequences are mutated in the *PHB* and *PHV* mRNAs, a gain-of-function of both genes is observed, revealing their participation in the leaf polarity determination and in meristem maintenance [55,56]. The same effect is observed when genes involved in miRNA-mediated regulation, such as *ARGONAUTE1* or *SERRATE,* are mutated. Besides, miRNA targeting to the *PHB* transcript also influences the methylation status of the *PHB* locus [57]. Considering that the *PHV* gene is also methylated in the 3′ region, one might expect that all HD-Zip III genes are regulated at multiple levels [58]. Further analyses performed in *Nicotiana sylvestris* support these observations [59].

Subfamily IV

A detailed description of the expression patterns of these genes was achieved by analyzing their transcript levels in each organ and by studying the activity of their promoters fused to the *GUS* reporter gene. All HD-Zip IV proteins are expressed specifically in the outer cell layer of the plant organs in which they respectively play a role [19].

GLABRA2/ATHB10, *ATML1* and *PDF2* were the first characterized. They are apparently involved in establishing cell fates in the epidermal layer through the regulation of cell layer-specific gene expression. *atml1 pdf2* double mutants fail to survive after germination and their leaves seem to lack epidermis [60]. *ANL2* affects anthocyanin accumulation in the subepidermal layer of the leaf, suppressing it when mutated. It is also involved in the determination of cell identity in the roots. *HDG11* is another HD-Zip IV family member whose single mutants show an abnormal phenotype, that is, an excess of branching of the trichome. Such effect is enhanced in double mutants of *HDG11* and *HDG12*, suggesting that both genes act by repressing the outgrowth of trichomes. *hdg12* single mutants, however, show no trichome phenotype, indicating that this gene plays a subsidiary role in this process. Alternatively, an epistatic relationship between *HDG11* and *HDG12* might be established during branch repression. For their part, *gl2* mutants exhibit trichome abortion and an increase in the number of root hair. As a result of the phenotype observed in *gl2 hdg11* and *gl2 hdg12* double mutants, *GLABRA2* seems to be epistatic to *HDG11* and *HDG12*. The remaining single mutants of HD-Zip IV genes exhibit a phenotype indistinguishable from that of the wild type. Loss-of-function analysis suggest that paralogous members no longer have overlapping functions, because double mutants do not show any phenotypical alterations. Instead, expression patterns indicate that this might actually occur between less related members. This is the case of *HDG2* and *HDG5* or *FWA* and *HDG8*, which are not paralogous but whose expression patterns bear close resemblance [19].

A summary of functional roles played by the members of each HD-Zip subfamily is given in Table 2.

Evolutionary history of the HD-Zip family

HD-Zip-encoding genes have been isolated from a wide variety of plants, such as *Solanum lycopersicum, Craterostigma plantagineum, Zea mays, Pisum sativum, Glycine max, Daucus carota, Heliantus annuus, Nicotiana sylvestris, Silene latifolia, Picea excelsa, Zinnia elegans, Lotus japonicus, Medicago truncatula, Brassica napus* and *Physcomitrella patens* (a moss), which includes monocots and dicots, C3 and C4 plants. Many of the identified proteins

Table 2. The four HD-Zip subfamilies

Subfamily	Functions[a]	Refs
HD-Zip I	Response to abiotic stress	[27]
	Response to ABA	[9,23,24]
	De-etiolation	[9]
	Blue-light signaling	[30]
HD-Zip II	Response to illumination conditions	[28]
	Shade avoidance	[39–41]
	Response to auxins	[44,45]
HD-Zip III	Embryogenesis	[46]
	Meristem regulation	[47,49,54,55]
	Lateral organs initiation	[47]
	Leaf polarity	[46,51,55]
	Vascular system development	[49,51,53]
	Auxin transport	[38,41]
HD-Zip IV	Epidermal cells differentiation	[19,60]
	Anthocyanin accumulation	[19,81]
	Root development	[19,22,81]
	Trichomes formation	[19,82,83]

[a]Physiological events in which HD-Zip-encoding genes are involved.

have been well studied and seem to bear not only structural but also functional resemblance to those of the model plants [14,28,37,61–68].

The numbers of the known members constituting the four HD-Zip subfamilies are variable and dependent on each plant species. However, these numbers should not significantly differ from those of the plants whose genomes have been completely sequenced. Naturally, structural and functional characterization of HD-Zip proteins from species other than *Arabidopsis* and rice is taking longer [9,26].

HD-Zip I genes have evolved through a series of gene duplications to a level of complexity that has resulted in subsets of paralogous genes. These genes share intron/exon distribution, amino-acidic sequences and expression patterns. Ectopic expression of each HD-Zip I subset of genes leads to different phenotypic effects, however, in some cases, this also happens among members from the same subset [9]. Phylogenetic analysis showed that, in both monocots and dicots, the evolutionary rate of the HD-Zip I genes was considerably faster than that of the HD-Zip II genes [66].

START domain-containing proteins were first classified in subfamilies III and IV, based on the presence of the loop in the middle of the LZ motif [16]. Phylogenetic analysis confirmed this separation, by revealing an important divergence at the N-terminal part of the START domain [69].

Regarding the HD-Zip IV genes in particular, homologs have been identified in representatives of many land plants. Most of *Arabidopsis* HD-Zip IV members form paralogous gene pairs. A comparison with other plant species suggests that some of the gene duplications took place before the divergence of gymnosperms and angiosperms [19].

HD-Zip III genes show a remarkable conservation. Indeed, homologs have been identified in representatives of all plant lineages. Phylogenetic analysis has strongly suggested that HD-Zip III genes were first associated with basic growth and patterning in ancient land plants. Throughout evolution, they diversified and acquired new functions that contributed to the modification of land plant development and to the origin of new tissues and organs,

such as the vascular system and leaves. Nonetheless, whether HD-Zip III genes evolved before the origin of land plants or uniquely within the land plant lineage remains an unresolved question [48,70,71].

Not all functions are conserved between angiosperms and gymnosperms. A thorough phylogenetic analysis of HD-Zip proteins in a wide group of land plants indicates that a gene duplication event took place before the division of angiosperm–gymnosperm, leading to two gene lineages. Such lineages diversified during angiosperm plant radiation [46,48]. The heterologous expression of a member of subfamily III from moss in a *rev Arabidopsis* mutant did not complement the phenotype, indicating the existence of a functional difference between flowering and non-flowering plant homologs. Although the moss HD-Zip III gene and *REV* do not perform the same function in specifying organ polarity, they still play a similar role in vascular development and organ initiation [48]. Finally, the alignment of mRNA sequences of HD-Zip III genes from different plant lineages showed that the target sequences of two miRNAs, miRNA 165 and miRNA 166, (and, therefore, the miRNA-mediated regulation of gene expression) are well conserved [70,72]. The authors of these studies concluded that this gene regulation mechanism seems to predate the emergence of flowering plants and might have played an important role in the evolution of land plants.

Concluding remarks and future perspectives

HD-Zip proteins have been well studied in the past few years. Functional and DNA-binding studies have demonstrated that each of the HD-Zip proteins studied participates in alternative signal transduction pathways and mediates a cross-talk between some of the pathways. Nonetheless, many questions are still unanswered. How can these proteins that have similar binding specificity and that show overlapping expression patterns, function in different pathways? What are the functions of these transcription factors in plant development? Do they interact with any partners for their function?

Although HD-Zip proteins have crucial functions, analogous to the rest of plant HD-containing proteins, they do not play homeotic roles. The existence of such a large multigenic family involved in a fine regulation of the developmental program might be the result of the evolutionary pressure.

Hopefully, some of these questions will be answered in the near future, and it is possible that the non-conserved regions might hide additional information to the already identified common motifs. Comparison of ortholog members to be isolated from a broader range of species should shed light on this subject.

Throughout history, all famous families have kept their own secrets. Time (with hard work and good luck) will tell if we are able to reveal those of the HD-Zip family.

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