

Stem cells in plants are differentiated too

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ABSTRACT

Stem cells in plants, better known as meristems, can determine the number and fate of cells, structure and fate of tissues, shape and type of organs, phases of plant vegetative and sexual reproduction, and general plant architecture. In this review I propose that, contrary to the common view, stem cells in plants in both their broad definition (all meristems) or even according to the narrower one (central part of the shoot and root apical initial-cell zone) are differentiated. I list the various plant meristems and discuss why they should be considered differentiated. I also discuss their alleged totipotency *in vivo*, the question of whether all plant cells are differentiated, the types of cell differentiation and the issue of re-differentiation by plant cells. The question of whether there are several types of stem cells in plants that differ in their level of "stemness" is also discussed. Plant meristems (including apical ones) are differentiated from structural (they include several cell types) and functional points of view. Their various types of origin, many times following re-differentiation of other cell types also indicate that meristems are differentiated. Finally, now it is well established that their development and physiology depend on the up- or down-regulation of specific regulatory genes that induce stem cell production and regulate their amount. All these facts clearly indicate that they are differentiated and that they specialize in the production of new cells, tissues and organs.

INTRODUCTION

Stem cells in animals became a hot scientific, medical, economic and political issue because of the moral implications of certain scientific and technological

innovations. Stem cells offer a unique opportunity to advance our understanding of cell biology, especially answering many complicated questions related to the nature of cell and tissue differentiation. Of course, there are many practical opportunities that arise from a better understanding of stem cell biology.

There is a general misconception that plant stem cells are undifferentiated. To correct this misconception, there are several major issues that warrant discussion: Are there non-differentiated cells in plants at all? What is a differentiated cell? What is the distinction between terminal and non-terminal differentiation of cells? Are dividing cells either differentiated or not, or can their status vary? Does differentiation mean that the cell was dividing at the beginning of the process? Does plant cell differentiation imply that the differentiating cells were previously undifferentiated? Are there pre-patterns in the meristems that determine pattern formation in their derivatives, or do the patterns develop only later and independently? Are stem cells totipotent?

DEFINITION OF STEM CELLS IN ANIMALS AND PLANTS

A major characteristic of stem cells in animals is their potential to give rise to other cell types depending upon their developmental history. Embryonic animal stem cells are pluripotent, whereas in the adult their potential for differentiation is usually restricted to specific cell types [1,2]. Potten and Loeffler [1] defined the characters of animal stem cells: Undifferentiated cells capable of proliferation, self-maintenance, production of large numbers of differentiated functional progeny, regeneration of the tissue after injury, and flexibility in the use of these options. They explained that, in practice, this undifferentiated state means that the cells

can be identified only by the absence of a differentiation marker in comparison with some other cells. They used the term undifferentiated in its morphological sense, that the cell has no physical features commonly attributed to specialized chemical or physical functions. More specifically, Potten and Loeffler [1] defined differentiation as a qualitative change in the cellular phenotype that is the consequence of the onset of synthesis of new gene products, i.e., the new changes in gene expression that led ultimately to functional competence. Following the accumulation of a wealth of new molecular data, Blau et al. [2] proposed that stem cells fulfill a function that may be performed by an array of various cell types, but are not an entity. As will be discussed later, plant meristems clearly fit the definition of differentiated cells because they are structurally and functionally differentiated. For instance, the largest plant meristem, the vascular cambium, is differentiated into two specific cell types (fusiform and ray initials) [3, 4]. Steeves and Sussex [5] defined differentiation of plant cells as a process in which cells become structurally distinguishable and functionally distinctive, a definition that describes stem cells even according to their narrower definition. Clark's [6] study, unlike most others mentioning plant stem cells, specified the criteria for undifferentiated cells: Cells that are morphologically distinct from differentiated ones in that they are small (~5 µm across), densely cytoplasmic, lack large vacuoles, and have a great developmental potential. Francis [7] used the definitions given in Potten and Loeffler [1]. However, most recent authors who have mentioned plant stem cells in their papers referred only to the central part of the shoot or root apex (the promeristem) as stem cells. Barlow [8, 9] (who introduced the stem cell concept to plant biology) referred to plant meristems in general as being made of stem cells. Because of the different attitudes toward the identity of plant stem cells, their current definition is not clear and this issue needs clarification. Here, I refer to all meristematic cells in plants as stem cells, as did Barlow [8, 9].

THE NARROW DEFINITION OF STEM CELLS IN PLANTS

In the vast majority of papers mentioning plant stem cells, they are mentioned briefly without discussing the issue of what stem cells are. These papers are usually the outcome of the studies of scientists who are concerned more with the genetic and molecular aspects

than with the classic developmental ones. Moreover, it is obvious that many of them are less trained in the broad and complicated issues of classic plant development. Most of the 78 papers mentioning plant stem cells refer only to the apical initials, a small region within the apex, as stem cells. This approach seems to have originated in the influential book by Steeves and Sussex [5]. However, Steeves and Sussex [5], who referred only to the central parts of apices as stem cells, never discussed or explained their approach in depth and why they have decided to change from Barlow's original definition. This issue has been never given enough attention, and the "paper evolution" of the narrower definition for stem cells in plants seems to have gained momentum because of the "founder effect" of the use of the narrower definition by the small group of people who did the first successful and important modern studies on this topic.

PLANT CELL DIFFERENTIATION AND RE-DIFFERENTIATION

The question of whether there are undifferentiated plant cells has been addressed by Sachs [10], who clearly demonstrated that even plant tumors are composed of differentiated cells, although in a tissue structure that differs from a normal one. Sachs also stated that all known plant cells are differentiated - a position with which I fully agree. This view implies that there is no such thing as de-differentiation. When differentiated cells change their identity and become a different cell type, the process should be referred to as re-differentiation. Sometimes, the term trans-differentiation is used, but this definition is redundant. Although some authors also use the term "de-differentiation", I consider it erroneous because it suggests that cells become undifferentiated. One of these new fates is to re-differentiate into a stem cell.

There are, however, two major types of cell differentiation in plants: terminal and non-terminal. Terminal differentiation means that the cell can no longer change its fate, because it died [vessel member, phellem (cork) cell], had a thick secondary wall that does not allow for further changes (sclereid, fiber), or lost its nucleus (sieve element). Non-terminal differentiation occurs when a cell can later change its fate if it is exposed to the right signals.

A GENERAL OVERVIEW OF STEM CELLS (MERISTEMS) IN PLANTS

Plants have a set of several groups of cells, known as meristems, that can divide and give rise to new tissues

(including other meristems in certain cases) and organs. Meristems are classified according to their origin, the type of tissues they give rise to, and their position in the plant [11, 12]. At the root and shoot apex, and in similar structures (branching at leaf axils and emerging side roots, but also in barks of many trees and shrubs, calli, in association with wounds, or leaf margins in certain taxa), there are apical initials (promeristems) that give rise to five primary meristems (protoderm, ground meristem and procambium in the shoot; protoderm, ground meristem, procambium, pericycle and calyptrogen in the root) that form the primary body. In the base of internodes and leaves, especially in many monocotyledons, but also in some dicotyledons, there are regions in which islands of the apical primary meristems remain meristematic and later induce intercalary growth. These five primary meristems (all histologically and functionally distinct from one another, and in some, even the genetic and molecular basis for such distinction is partly known) produce primary tissues and gives rise to certain parts of the secondary meristem, the vascular cambium. The development of the secondary body is the outcome of the activity of several secondary meristems (the vascular cambium, phellogen - cork cambium, and dilatation meristem). The vascular cambium in the root originates in part by re-differentiation from primary meristems (the procambium and pericycle). In the shoot, the vascular cambium originates from the procambium in fascicular regions and following the re-differentiation of non-meristematic parenchyma cells in the interfascicular region. In addition, the cambium may originate from callus in both root and shoot.

In addition to these meristems, leaves and fruits have their special primary meristems, involved in their expansion, and some woody monocotyledons have their variation of the cambium [3, 11, 12]. Adventitious apical meristems of both shoots and roots also re-differentiate after wounding from cell groups that had other fates, i.e. callus or parenchyma [see 13, 14].

THE SECONDARY MERISTEMS: THE VASCULAR CAMBIUM, PHELLOGEN (CORK CAMBIUM) AND DILATATION

The most common and best-known secondary meristem is the vascular cambium that gives rise to the secondary xylem and phloem. In the stem, the origin of several strands of the vascular cambium is from the procambial strands, but the vascular cambium that develops in the gaps between these strands originates by re-

differentiation of the inner cortical parenchyma [12]. Similarly, in the roots, the procambial strands re-differentiate and form sectors of the vascular cambium, and the remaining parts of the vascular cambium that develop in the gaps between these strands originate by re-differentiation of the pericycle [12]. In addition, in both shoots and roots, a wound-related vascular cambium can re-differentiate from callus parenchyma. In most vascular seed plants, the vascular cambium has two types of initials (fusiform that form the axial system that always develops, and ray initials, which may be lacking in the juvenile stage or during the full length of life in certain plant species, that form the radial system - the rays) [3, 4, 11, 12, 15]. Moreover, there are cases in which there are higher levels of order in the vascular cambium, such as domains (regions that divide to the left or to the right) [16] and storied cambia in which large regions and sometimes all the vascular cambium are arranged in horizontally aligned, well-defined stratified cell files [4, 17]. Thus, it is obvious that the vascular cambium is differentiated both structurally and functionally.

A second important secondary meristem is the phellogen (cork cambium), the meristem that forms cork. Phellogen always re-differentiates from non-meristematic cells that had already differentiated and functioned for a short or long time as one of several cell types: epidermis, hypodermis, cortical collenchyma, photosynthetic and storage parenchymas, phloem axial parenchyma, xylem and phloem ray (radial) parenchyma, pith parenchyma or callus. Structurally and functionally there are two types of phellogen cells: the common type that gives rise to regular cork cells (phellem cells, phelloids or sclereids), and regions that form lenticels (parenchymatous ventilation shafts in the bark).

In the bark of many plants, there is an additional special secondary meristem - the dilatation meristem that allows the already differentiated cortex and/or phloem to expand and grow in girth in accordance with the increase in the girth of the xylem, to avoid cracks in the bark [18]. Dilatation meristem is also always formed by re-differentiation of cells: cortical collenchyma, photosynthetic and storage parenchymas; and from axial and ray (radial) secondary phloem parenchyma [18, 19].

THE HISTORY OF STUDYING STEM CELLS IN PLANTS

The early attempt to introduce the concept of stem cells

to plant biology [8] was, unfortunately, generally overlooked by the main stream of developmental botany until recently. Barlow [8] was the pioneer that recognized the fundamental functional similarity, in nature and role, of plant initial cells in meristems to animal stem cells. Barlow [8] focused on several primary and secondary meristems: shoot and root apical meristems and two lateral meristems, the cambium and phellogen (cork cambium) and considered all these stem cells to be undifferentiated. The characteristics of stem cells according to Barlow [8] are: a slower rate of division and residence at G₀ phase, the capability to repopulate the meristem when damaged, and pluripotency (but sometimes in the text proposed totipotency). Later, in their short discussion of plant stem cells, Steeves and Sussex [5] pointed to the basic difference between plants and animals, i.e., that in animals, post embryonic stem cells are associated with the replacement of tissues within the existing body and the additional new cells are balanced by cell loss, whereas in plants there is a continued addition not only of replacement cells, but also of new cells and organs throughout the life of the plant. In the last 25 years, the concept of stem cells was gradually introduced into developmental botany [5-9, 13, 20-92]. A basic developmental view expressed in most of these studies and essays, usually as a short phrase without discussing it, is that plant stem cells are not differentiated. I propose that this view should be revised based on the evidence that plant meristems in general and even the apical stem cells in particular are, in fact, differentiated.

GENES INVOLVED IN PLANT STEM CELL DIFFERENTIATION

The action of several genes (and probably many others not mentioned here and ones that await discovery) has already been shown to be involved in the regulation of differentiation of shoot apices [46, 58]. I will briefly give only a few cases, just to document that plant stem cells (specifically in shoot apices that have been studied more than other meristems using modern approaches) are differentiated. Although pleiotropic effects may be involved in the developmental processes of mutants, I still refer to them. As early as in the globular to torpedo stages of the *Arabidopsis* embryo, the differentiation of the apical part depends on the activity of specific genes. Moreover, it has been found that even at the globular embryo stage, the apical portion is composed of domains demarked by expression of different genes [93, 94]. The gene products of both

SHOOTMERISTEMLESS (STM) and *PINHEAD* [23, 63, 95], the activities of *TUMOROUS SHOOT DEVELOPMENT (TSD)* genes [72] and *KANADII* or *KANADI2* and *YABBY* genes [96], *CLAVATA (CLV)* [58, 83] or *REVOLUTA* [66, 97] are required for the development of normal shoot apices in *Arabidopsis thaliana*. Similarly, the genes *shoot meristemless*, *wuschel* and *zwille/pinhead* are involved in the development of shoot apex of somatic embryos of *Arabidopsis thaliana* [98]. The putative receptor kinase *ACR4* gene of *Arabidopsis thaliana* expressed in the outer cell layers of embryos and plants [99] indicate that the L1 layer differs from the L2 and L3. These findings clearly indicate that shoot development, including the stem cells it contains, is a regulated process. Cells in the position of the shoot apical meristem, in spite of their position, will not function as a shoot apical meristem if the genes involved in shoot apical meristem identity do not function. In the apical meristem itself, the stem cells have been proposed to provide signals that determine adaxial (upper side of leaf) cell fate [96, 100], a specific regulatory function that, if it exists, is sufficient to mark these cells as differentiated. Further indications for differentiation in the shoot apical meristem are evident in the spatial regulation of several other genes, including an asymmetric expression of genes related to brassinolide biosynthesis within it [46, 101], or cytokinins [102]. A similar indication for differentiation in the shoot apex of *Arabidopsis* was the analysis of fluorescent tracer movement via plasmodesmata that revealed that there was spatial and temporal regulation of symplastic domains within the apex [103], i.e., the shoot apex is not a physiologically uniform tissue. *Brostm*, a *Brassica* homologue of the *Arabidopsis SHOOTMERISTEMLESS (STM)* gene, and a cyclin box-derived gene *Broscyc* marked the *de novo* formation of adventitious shoot meristems in stem segments in *Brassica oleracea*. The developmental switching from phloem parenchyma into a meristem occurred before cell divisions began [104], thus the stem cells of the adventitious shoot meristems were the outcome of re-differentiation of parenchyma cells that changed their fate. In maize, it has been found that in seedlings homozygous for both *sml* (shootmeristemless) and *dgr* (distorted growth) loci a shoot-apical-meristem-less phenotype occurs [81]. Again, the mutual action of these two genes is required to regulate apical meristem development. Similarly, the maize homeobox gene *knotted1* is required for the normal development and

maintenance of its shoot apical meristem [105, 106]. Overexpression of the maize homeo box gene *KNOTTED-1* in tobacco caused, among other phenotypic alterations, the differentiation of ectopic growth apices (including stem cells) on leaves [107, 108]. Looking at the figures in Sinha et al. [107], it seems that the ectopic growth apices re-differentiate from L1 and L2 layers of the leaves, i.e. from differentiated cells. Moreover, the expression, or lack of expression, of growth apices in certain positions in the plant, as revealed by the experiments with *KNOTTED-1* in tobacco [107], is another strong indication that growth apices that include stem cells are differentiated. Growth apex development is thus up or down regulated by the action of specific genes, which in turn may be up or down-regulated by various hormonal or other developmental signals.

RE-DIFFERENTIATION BY PLANT CELLS

Plant cells can re-differentiate (sometimes redundantly described as trans-differentiation) even if they were previously differentiated. Re-differentiation may or may not involve cell division. Many differentiated but non-meristematic plant cells can switch between different cell fate alternatives better than animal cells can. This does not mean that they are undifferentiated. They simply keep cell fate machinery and other aspects of their biology, such as cell wall structure, in a state that enables them to re-differentiate when they are exposed to the right developmental signals. In addition, it is also note worthy that many plant cells can directly re-differentiate to form vascular elements, fibers and sclereids without re-entering the cell cycle. The common situation, in which the same cell type, at the same position and age, may have several alternative fates according to the signals (not all known yet) it is exposed to, is demonstrated here by a parenchyma cell in the cortex of a dicotyledonous plant. Such a cell may directly re-differentiate into one of the following cell types: (1) phellogen (meristematic), (2) callus (meristematic), (3) dilatation meristem (meristematic), (4) dilatation cell via expansion without becoming meristematic, (5) a vessel member (without cell division), (6) a tracheid (without cell division), (7) a sieve element (without cell division), (8) a fiber, (9) a sclereid, (10) an initial gum duct [12]. Some of these fates require re-entrance into the cell cycle, some only a change in cell shape and physiology, some change their secondary cell-wall structure (with or without a change in cell shape), some require programmed cell death

(always in vessel members and tracheids) or a not yet defined type of cell death (in certain fibers and sclereids), or a loss of the nucleus (in sieve elements). Thus, here again, becoming meristematic, or in other words, becoming a stem cell, is a specific case of re-differentiation, sometimes shortly and sometimes even many years after the original parenchymatic cell differentiated. All these examples clearly indicate that plant meristems (stem cells) are commonly formed by re-differentiation.

ARE THERE SEVERAL TYPES OF STEM CELLS IN PLANTS THAT DIFFER IN "STEMNESS" LEVEL?

Certain plant cells, such as apical initials, are considered by all to be stem cells. Such cells are fully occupied by their stem cell identity. Gradually, they re-differentiate to become other cell types and lose their stem cell identity. During the process of re-differentiation they may still resume their full stem cell function as the result of wounding or following the exposure to various developmental signals. As mentioned before, several cell types, especially non-lignified ones, including the epidermis, hypodermis, cortical collenchyma, photosynthetic and storage parenchymas, xylem and phloem axial parenchyma, xylem and phloem ray (radial) parenchyma and pith parenchyma, may become meristematic following normal ontogeny, wounding or other changes in physiology. It seems that these cells (unlike certain other cell types in plants) retain a stem cell potential along with their other functions. In animals there is evidence that stem cells might show specific expression of a large group of genes (most of which still have an unknown function) [109, 110]. In plants this possibility has not yet been examined. If plants share this phenomenon, it is tempting to suggest that cell types that have the potential to re-differentiate express at least a subset of such genes.

DETERMINATION OF PRE-PATTERNING BY PLANT STEM CELLS

Another aspect that may indicate differentiation is the determination of patterns in tissues by pre-patterns within the meristem that give rise to patterns in the daughter tissue. Unlike animals, in plants there are many cases when lineages do not determine the exact fates of cells but positional cues determine it. However, statements that lineages do not determine cell fates in meristem derivatives [i.e., 55] do not describe

the full, actual, complicated situation of plant development. A clear case of pre-patterns in a meristem that determine pattern formation in its derivatives and cell lineages is the vascular cambium and its derivatives, the secondary xylem and phloem. In the vascular cambium there are two types of initials: The fusiform initials (usually the majority) that divide and later differentiate into the axial system of the secondary xylem and phloem, and the ray initials that divide and later differentiate into the radial system of the secondary xylem and phloem [4, 11, 12, 15]. Cell size and pattern in the secondary xylem, for instance, is a good copy of the patterns of initials in the vascular cambium including their production and elimination. Thus, serial tangential sections in the layers of the secondary xylem enable reconstruction of the ontogeny of the vascular cambium over time [4], even over millennia [111]. The pre-patterning that is evident to a certain extent (it is not clear whether all the cellular patterns within the axial components of the secondary xylem and phloem are determined only in the vascular cambium or whether some are determined only in the zone of differentiating cambial derivatives) in the vascular cambium does not necessarily indicate similar pre-patterning for all or some of the other meristems. In the root apex, for instance, laser ablation of cells enabled a good demonstration that positional signaling determines cell fate [112]. However, in the *Extra cell layers*1 mutation of maize the timing of cell divisions in a primary meristem can determine whether the cells will acquire a fate according to their position or lineage [113]. In any case, the cell layer within the shoot apical meristem (L1, L2, L3) greatly influences cell-type fate [114]. The certain flexibility in switching from L1 to L2 and from L2 to L3 by periclinal divisions and the ability to regenerate L1 from L2 and L3 after wounding [see 115] and the random directions and numbers of cell divisions [116] do not allow for a fixed determination of fates. The fates are, however, determined by a partly flexible system that has greater tendencies for certain patterns than for others in the derivatives of the stem cells [i.e., 117].

ARE STEM CELLS ACTUALLY TOTIPOTENT *IN VIVO*?

The issue of the so-called totipotency of stem cells also needs clarification. Stem cells in their *in vivo* condition (in the shoot apex for instance) and possibly also in *in vitro* culture do not express totipotency. They only produce a limited number of cell types, which in turn,

with the right developmental signals may re-differentiate to whatever cell type they still have the potential to become. So in actual tissues, stem cells differentiate into a limited number of cell types. In this respect stem cells are not different from many parenchyma, collenchyma or epidermal cells. Bowman and Eshed [46], for instance, were more careful than many others who discussed plant stem cells and considered them a population of pluripotent cells, a much more accurate description than the common view as undifferentiated cells.

PERSPECTIVES TO CHANGE THE CORRENT VIEW

Currently, the list of papers in which plant stem cells are considered undifferentiated because of this misconception includes several dozen. It is still possible to correct the errant view and consider it tittering pains in a new, exciting and very important field. Over time, however, "paper evolution" will make it hard to correct the misconception. Because meristems are differentiated, the critical mechanisms that determine cell fate to become a meristem should be studied. Under the current dogma, there is a risk that the nature of the differentiation necessary to become a plant stem cell will be overlooked. More than a decade ago, Potten and Loeffler [1] wrote in their review of stem cells in animals "The present concepts involve a number of pitfalls, weaknesses and logical, semantic and classification problems. This indicates the necessity for new and well-defined concepts that are amenable to experimental analysis..... The terms that describe stem cell functions are often not well defined and are used loosely, which can lead to confusion." This criticism of animal stem cell research is equally valid for the current dogma on the nature of plant stem cells.

Most plant biologists today are excellent molecular biologists. The great achievements of the modern approaches in advancing our understanding of plant biology in the last 15 years clearly indicate the fruitfulness of these approaches. However, there is a risk that research groups will be unbalanced by the lack of the needed review and advise of organismic plant biologists. The number of brilliant scientists who know organismic plant biology is decreasing steadily. Many of the good tenured botanists are not updated with the new approaches (both technically and as a way of thinking and analysis) or are already busy with academic administration. I see this dangerous trend

expanding over the majority of the plant biology community. Under such conditions, botanical misconceptions may spread with no one to weed them out. Considering meristems or stem cells as undifferentiated is a specific case of this problem. Moreover, the shift from considering all meristems as stem cells (as defined by Barlow, [8]) to only a small group of apical cells has been introduced into plant science in what seems to be a casual manner with no deep and broad discussion that this issue deserves.

In the previous decade I argued for the use of *Arabidopsis thaliana* as a model for wood and fiber production in forest trees. For five years (1994-1999) I met a loud, common and strong opposition that among other views declared that *Arabidopsis* has no secondary growth. Now, when *Arabidopsis thaliana* is used as a model for wood and fiber formation in many laboratories, it seems obvious. I expect to see the same process of change of perspective about both issues: (1) stem cells in plants are not limited to a small subpopulation of the meristematic cells, and (2) stem cells in plants are differentiated.

CONCLUSIONS

I conclude that in plants, meristems are differentiated tissues and stem cells are differentiated cells. This is true from the structural point of view (being organized in distinct layers or having several cell types and expressing other higher levels of organization [i.e., 117]. This is also true from a functional point of view (there are various types of meristems). They are differentiated from a physiological point of view (active, dormant, determinate, indeterminate). They are differentiated from the point of view of origin during development (primary, secondary, traumatic, originating from other meristems or from non-meristematic cells). The considerable variability in meristem differentiation is expressed, for instance, in the many types of apical meristems: apical vegetative shoot meristems, apical reproductive shoot meristems, lateral vegetative and reproductive shoot meristems, adventitious shoot meristems, apical root meristems, side root meristems and adventitious root meristems [see 12, 118-122]. As discussed above, there are several known genes that regulate apical meristem differentiation and certain genes that are differentially expressed in certain meristems or domains within meristems [i.e., 123]; or specifically the *Athb-8* homeobox gene in the procambium [124]. So the new genetic data of the last decade largely supports the view

that meristems are differentiated. Meristems "assess" their position in the plant and function accordingly. Meristems have evolved and currently differentiate to function as a source of new cells and organs. Doing it in an organized, efficient and reliable way is a complicated function that is crucial to the fitness of a plant. Such critical functions could not have been left to non-specific, "undifferentiated" cells. Barlow's [8] pioneer recognition that plants have stem cells as do animals occurred some two decades before the similarities in certain gene activity made it clearer [41]. It is a pity that his important contribution had not stimulated research in plant meristems for over a decade of critical years when great developments occurred in the understanding of differentiation of animal cells. It will be no less sad if the misconception about meristem cells being undifferentiated, that was probably hard to notice with the meager knowledge about plant cell biology and gene action about a quarter of a century ago, will be the most cited and remembered phrase from such an important contribution. Differentiation to become a stem cell is a complicated and largely unknown process that deserves focused and realistic scientific attention. The misconception that plant stem cells are undifferentiated may result in wrong attitudes and a considerable delay in the expected understanding of their biology.

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REFERENCES

- [1] Potten, C. S., and Loeffler, M. 1990, Development, 110, 1001.
- [2] Blau, H. M., Brazelton, T.R., and Weimann, J. M. 2001, Cell, 105, 829.
- [3] Philipson, W. R., Ward, J. M., and Butterfield, B. G. 1971, The vascular cambium, Chapman & Hall, London.
- [4] Larson, P. R. 1994, The vascular cambium. Development and structure, Springer, Berlin.
- [5] Steeves, T. A., and Sussex, I. M. 1989, Patterns in plant development, 2nd ed., Cambridge University, Cambridge.
- [6] Clark, S. E. 1997, Plant Cell, 9, 1067.
- [7] Francis, D. 1997, Stem cells, C. S. Potten (Ed.), Academic, London, 59.
- [8] Barlow, P. W. 1978, Stem cells and tissue homeostasis, B. I. Lord, C. S. Potten, and R. J. Cole