pH/T duality – equation of state for plants

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Abstract We examined the pH/T (or μ/T) duality of acidic pH (or auxin-induced acidification) and temperature (T) for the growth of grass shoots in order to determine the equation of state (EoS) for living plants. By considering non-meristematic growth as a dynamic series of phase transitions in the extending primary wall, we identified the critical exponents for this phenomenon, which exhibit a singular behaviour at a critical temperature, critical pH and critical chemical potential (μ) in the form of four power laws: $F(\tau) \propto |\tau|^{\beta-1}$, $F(\pi) \propto |\pi|^{1-\alpha}$, $G(\tau) \propto |\tau|^{-2-\alpha+2\beta}$ and $G(\mu) \propto |\mu|^{2-\alpha}$. The power-law exponents $\alpha$ and $\beta$ are numbers, which are independent of pH (or μ) and T that are known as critical exponents, while $\pi$ and $\tau$ represent a reduced pH and reduced temperature, respectively. Various ‘scaling’ predictions were obtained – a convexity relation $\alpha + \beta \geq 2$ for practical pH-based analysis and a $\beta \equiv 2$ identity in microscopic representation. In the presented scenario, the magnitude that is decisive for growth is the chemical potential of protons. The EoS span areas of the biological, physical, chemical and Earth sciences cross the borders with the language of phase transitions.

Key words: cell wall, chemical potential, critical exponents, grass, maize, plant sciences, power laws, scaling relations, systems biology
Introduction

General outline This work deals with the derivation and determination of an equation of state (EoS) for plants that integrates the relationship between temperature and pH (or chemical potential, $\mu$) with growth. Based on reliable (published) data, we sought to identify the state variables that are necessary for optimal growth (at a constant turgor pressure). The limited number of parameters can be misleading; this approach does not oversimplify the complexity of the biological systems, but conversely, takes all of them into account by building an outer scaffold that cannot be surpassed. We cannot dismiss the possibility that this is the role of phenomenology in making universal statements, in this case imposing a physical constraint (i.e. EoS) on the extending primary wall.

A commonly held view is that biological systems are complex, and that therefore modelling has to cope with the fact that the degrees of freedom are numerous, and correspondingly, the number of parameters should be high. Though physical systems on a microscopic scale are also complex and the number of degrees of freedom in real systems is abundant (Avogadro number, as in the solid state physics), their behaviour (and time evolution) can usually be described by a few parameters and thermodynamic potentials (e.g., the Ginzburg-Landau theory of superconductivity (Ginzburg, 2004)) within the framework of phase transition theory. A similar argument also concerns microscopic (quantum) theories such as the Hubbard (1963) model, which has only two parameters: $t$ – for the “hopping” integral and $U$ – for the Coulomb interaction of electrons within narrow energy bands. In what follows, I argue that, similar to physical systems, a low number of relevant parameters may also be sufficient in biological systems.

The growth of plant cells and organs (such as extending cylindrical organs such as grass shoots) are affected by light and humidity, which along with pH and temperature are the most important factors that influence the growth of plants. It can be a rewarding enterprise to understand growth in terms of the time evolution of phase transitions that take place in the peripheral cell wall via the exchange of particles and energy-consuming metabolic processes (such as energy-conserving ATP) with the inside of cell compartment – the vacuole and cytoplasm, which are treated as molecules and the heat reservoir (thermostat). Hence, in this article we consider living organisms as open systems that dissipate energy and exchange entropy and matter with the environment in the sense that was summarised in Barbacci et al. (2015). We introduce a continuous phase transition approach as the realisation of the same
four properties that were established by Per Bak et al. (1998) that are linked to the dissipative structures, which we can use to understand the biology of living organisms. Even though we considered only one phase transition, a cascade (ratchet) of subsequent phase transitions, which constitute the extension and growth of the cell wall, can be imagined.

**Preliminaries** Plant developmental systems have evolved within the universal limitations that are imposed by the plant cell wall (Lintilhac, 2014). Plant cells encase themselves in a complex polysaccharide wall (Cosgrove, 2005), and characteristically obtain most of their energy from sunlight via the photosynthesis of the primary chloroplasts. The expansive growth of turgid cells, which is defined as an irreversible increase in cell volume, can be regarded as a physical process that is governed by the mechanical properties of the cell wall and the osmotic properties of the protoplast (Schopfer, 2006). The precise biochemical mechanism that regulates the ability of the growth-limiting walls to extend irreversibly under the force of turgor pressure has not yet been identified (Kutschera, 2000).

Growing plant cells characteristically exhibit acid growth (Rayle and Cleland, 1970; Hager et al., 1971; Cosgrove, 1989), which has been formulated in the form of the “acid growth hypothesis” (Hager, 2003; Taiz and Zeiger, 2006). The acid growth hypothesis postulates that both phytotoxin fusicoccin (FC) and the growth-promoting factor auxin (indole-3-acetic acid, IAA) cause wall-loosening and produce the concomitant induction of growth (growth enhancement) through the rapid acidification of the extension-limiting cell wall (Kutschera, 1994). These enigmatic “wall-loosening processes” are in fact minor changes within the polymer network of the extension limiting walls (i.e. the incorporation of proteins, the enzymatic splitting of polymer backbones or covalent cross-links or the disruption of non-covalent interactions between wall polymers).

Temperature is an underappreciated tool for research; in many cases the process of growth can be differentiated by the response of plants to temperature (Went, 1953). Actually, only a few papers in which temperature response is treated as a major problem can be mentioned (Yan and Hunt, 1999). However, cell growth, especially cell elongation, has a high $Q_{10}$ factor, which indicates that this is a chemically rather than physically controlled phenomenon (the $Q_{10}$ temperature coefficient is a measure of the rate of change in a biological or chemical system as a consequence of increasing the temperature by 10°C). Growth is accomplished through the enlargement of the cell volume owing to water uptake, which maintains the appropriate inside pressure in the vacuole as well as the irreversible extension
of the pre-existing (primary) cell wall. Hereafter, we will assume an almost constant or slowly varying turgor pressure, which by definition is a force that is generated by water pushing outward on the plasma membrane and plant cell wall that results in rigidity in the plant cell and the plant as a whole.

Expansive growth is the result of the coupling effects (Barbacci et al., 2013) between mechanical (pressure), thermal (temperature) and chemical energy (pH). The thermal sensitivity of biochemical processes refers to the coupling effect between the thermal and chemical energies. In the search for a plant-wall specific ‘equation of state’ in growing biological cells or non-meristematic tissues, we considered two ‘state variables’, namely temperature and pH at a constant turgor. We note that pH is not sensu stricto a fundamental physical quantity, and cannot be treated as a usual intense variable. Nonetheless, relying on the definition of pH and then considering the chemical potential (Baierlein, 2001), which is a proper state variable, we may propose the following.

Evidence has accumulated that the final goal of auxin action is to activate the plasma membrane (PM) H+-ATPase, a process which excretes H+ ions into the cell wall. The auxin-enhanced H+-pumping lowers the cell wall pH, activates pH-sensitive enzymes and proteins (such as expansins (Cosgrove, 1993), xyloglucans (Fry et al., 1992) or yieldins (Okamoto-Nakazato et al., 2001)) within the wall and initiates cell-wall loosening and extension growth (Hager, 2003). It has also been observed that when auxin-depleted segments were submerged in an acid buffer solution, the segments started their elongation growth immediately (“acid growth”), whereas the auxin-induced growth (“auxin growth”) began after a delay (lag phase), as is shown in Fig. 1 (curve 1) in Hager (2003); see also the remaining plots, which are of quite a different character, (curves 2 – 6) that correspond to “acid growth”. From the many investigations that have been done for more than four decades, it was deduced that protons, which are exerted into the wall compartment, are directly responsible for wall-loosening processes through the hydrolysis of covalent bonds, transglycosylation or the disruption of non-covalent bonds. The growth effect is illustrated in Fig. 2 in Hager (2003), where the “Zuwachs” (increment in %) is plotted against pH (SI Fig. 1a), which is defined as the decimal logarithm of the reciprocal of the hydrogen ion activity aH+ in a solution (pH = −log10 aH+ = log10 1/aH+). The pH value is a logarithmic measure of H+-activity (the tendency of a solution to take H+) in aqueous solutions and defines their acidity or alkalinity; aH+ denotes the activity of hydronium ions in units of mol/l. The logarithmic pH scale ranges from 0 to 14 (neutral water has a pH equal 7). However, pH can be expressed in terms of the more
elementary chemical potential $\mu_{\text{H}^+}$ of the proton (H$^+$) through the relation

$$\text{pH} = (\mu_{\text{H}^+}^0 - \mu_{\text{H}^+}) / RT \log 10,$$

where $R$ denotes the gas constant and $\mu_{\text{H}^+}^0$ is the reference potential.

Indirect measurements of the chemical potential ($\mu$) by means of pH can be compared to our previous measurements of the electromotive force (EMF) in order to detect phase transitions, which are localised by the “kinks” in the chemical potential in condensed matter physics (Matlak and Pietruszka, 2000; Matlak et al. 2001). pH is not a typical generalised coordinate, although the microscopic state of the system can be expressed through it in a collective way.

Temperature is among the most important environmental factors that determine plant growth and cell wall yielding (Pietruszka et al., 2007). Temperatures at which most physiological processes occur normally in plants range from approximately 0°C to 40°C, which determines the typical temperature scale (in Kelvin scale: [K] = [°C] + 273.15). The temperature responses of plants include all of the biological processes throughout the biochemical reactions. This is clearly visible in the relative rates of all of the development or growth processes of maize as a function of temperature, which is illustrated in Fig. 3 in Yan and Hunt (1999) and is reconstructed as SI Fig. 1b, to become self-contained. For future applications, we propose that the “absolute” temperature scale for plants [0 – 40°C] or [0 – 45°C], which corresponds to a [0, 1] interval after rescaling, be setup.

Astonishingly enough, the pH plots that are presented in Fig. 2 in Hager (2003) as well as the temperature plots in Fig. 3 in Yan and Hunt (1999) look similar (when mirror symmetry is applied). Providing that we perform a substitution $x \to (1 - x)$ and consider a function $f(x)$ of a variable $x$ and of its reflection $(1 - x)$, one might expect that they will look approximately the same after proper scaling. Not surprisingly, such a function exists in mathematics, and can be conveniently described by the Euler beta density distribution (or simply the Euler function). In this approach, we assume that $x$ equals either pH or temperature (both rescaled to a [0, 1] interval), an assignment which makes sense, since at relatively low pH, which corresponds to a relatively high T in this [0, 1] scaling, plant cells and organs grow (elongate) the fastest, which is clearly reproduced in SI Fig. 1.

Now, inspired by the Ansatz “educated guess” using Euler beta function $f(x, \alpha, \beta)$ for growth empirical data, we will show that the final action of temperature and pH on plant growth is effectively the same, though both triggers of these responses are apparently of a different nature.
Materials and methods

Based on the “acid growth hypothesis” and relevant experimental data (Yan and Hunt, 1999; Hager, 2003), we examined the pH/T (or μ/T) duality of acidic pH (or auxin-induced acidification) and temperature (T) for the growth of grass shoots, in order to determine the EoS for extending the primary wall of (living) plants.

When comparing data from different experiments (Yan and Hunt, 1999; Hager, 2003), we noticed that the x variable may, after rescaling, bear the following meaning – either acidity (basicity) $x = \text{pH}$ or temperature $x = T$ (see SI Fig. 2) with a characteristic beta function cut-off at $x = 0$ (SI Fig. 2a and the inset) and at $x = 1$ (SI Fig. 2b); the normalised scale we used is presented in SI Fig. 2b (inset).

Results

Derivation of the equation of state for plants

The probability density function of the beta distribution (also called the Euler integral of the first kind (Polyanin and Chernoutsan, 2011) for $0 \leq x \leq 1$ and shape parameters $\alpha, \beta > 0$ is a power function of the variable $x$ and of its reflection $(1-x)$

$$f(x; \alpha, \beta) = \text{const} \cdot x^{\alpha-1} (1-x)^{\beta-1} = \int_{0}^{1} \frac{x^{\alpha-1} (1-x)^{\beta-1}}{B(\alpha, \beta)} \, du$$

where $B = B(\alpha, \beta)$ is the normalisation constant. Based on the experimental observations as described above, equation (1) can be merged into a single expression

$$[c_{\text{pH}} \cdot \text{pH}^{\alpha-1} (1 - \text{pH})^{\beta-1}]_{T} = [c_{T} \cdot T^{\beta-1} (1 - T)^{\alpha-1}]_{\text{pH}}$$

where $c_{\text{pH}}$ and $c_{T}$ are constants and “pH” is treated here as a non-separable variable name. The lower indices “T” and “pH” in Eq. (2) should be read “at constant temperature”, and “at constant pH”, respectively, which is valid for preparation of strict experimental conditions.

By defining the state function $F$ at constant turgor pressure $P$ (isobaric: $\delta P \approx 0$)

$$F(T, pH; \alpha, \beta)_{P} \equiv \left( \frac{T}{1 - pH} \right)^{\beta-1} \left( \frac{1 - T}{pH} \right)^{\alpha-1}$$
and assuming adequate water uptake to fulfil this requirement, we determine from equation (2)

\[
\frac{T^{\beta-1}(1-T)^{\alpha-1}}{pH^{\alpha-1}(1-pH)^{\beta-1}} = \left(\frac{T}{1-pH}\right)^{\beta-1} \left(\frac{1-T}{pH}\right)^{\alpha-1} = \frac{c_{pH}}{c_T} = \Omega_S
\]

where \(\Omega_S\) is a dimensionless constant. We notice that Eq. (4) takes the usual form of an equation of state. Hence, the constitutive EoS for living plants explicitly reads

\[
\left(\frac{T}{1-pH}\right)^{\beta-1} \left(\frac{1-T}{pH}\right)^{\alpha-1} = \frac{c_{pH}}{c_T} = \Omega_S
\]

which has the elegant form of a double power law. Note that \(T\) is not the absolute temperature in the Kelvin scale, but the temperature rescaled to a \([0, 1]\) interval, as \(pH\) also is. Here \(\alpha\) and \(\beta\) are the shape exponents (SI Table 1 and SI Figure 3). The saddle-point type of solution that is presented in Fig. 1a represents a concave surface (Fig. 1b, for a contour plot), which is roughly accurate at low \(pH\) and moderate temperatures. Equation (5) becomes increasingly inaccurate (divergent) at very low or very high \(pH\) values (close to zero or one in the normalised scale, which we use throughout the article), though such excessive conditions, which constitute unacceptable extremes for life to come to existence, are excluded by Nature.

The so-called “threshold pH”, which corresponds to the minimum \(pH\) in Fig. 1a (or the horizontal line in the derivative, Fig. 1c), was already observed by Lüthen et al. (1990). The isotherms of Eq. (3) are presented in Fig. 2a, while the lines of constant \(pH\) are presented in Fig. 2b.

Even though we (apparently) launched our derivation from the “acid growth theory”, a closer look at the ‘initial conditions’ revealed that, in fact, it was established on the raw experimental data of \(pH\) and \(T\) dependent growth (SI Figure 1). For that reason, Eq. (5) is independent of whether the acid growth theory applies or not, and therefore seems universal, at least for tip-growing grass shoots. Note, Eq. (5) is not an evolution (growth) equation, but a system (cell wall) property equation. The solutions of Eq. (5) for \(\alpha\) and \(\beta\) in the form of contour plots are presented in SI Fig. 4 for comparison.

Essential thermodynamic relations are property relations and for that reason they are independent of the type of process. In other words, they are valid for any substance (here: the primary wall of a given plant species) that goes through any process (mode of extension). Yet, in our case, we should be aware of the empirical origin of Eq. (5) and memorise the fact that a
The fundamental state variable of the system is the Gibbs energy – the chemical potential $\mu$ at a constant pressure and temperature (here: $\mu = \mu_{H^+}$ for H$^+$ ions). Then

$$F(T, pH; \alpha, \beta) \approx G(T, pH(T); \alpha, \beta) = G(T, \mu_{H^+}; \alpha, \beta)$$

(6a)

where

$$G(T, \mu_{H^+}; \alpha, \beta)|_p = -\frac{(\log 10)^\alpha}{T-1} (\mu_0 - \mu_{H^+})^2 \frac{T-1}{(\mu_0 - \mu_{H^+})RT} \frac{RT^2}{RT + (\mu_0 - \mu_{H^+}) \log 10} \beta^{-1}$$

(6b)

$G_p(T, \mu)$ is another state function though expressed properly by intense, microscopic state variables; temperature $T$ here belongs to the [0, 1] interval. In approximation, since pH is measured in isothermal conditions (SI Fig. 1a), we may temporarily neglect the pH(T) dependence in equation (6a) and consider pH and T as intense (non-additive) variables.

The equation of state is the relation between state variables that describes the state of matter under a given set of physical conditions. It is a constitutive equation that provides a relationship between two or more state functions that are associated with the matter, such as its temperature, pressure, volume or internal energy. They are useful in describing the properties of gases (ideal gas law: $PV = nRT$), fluids (van der Waals equation of state, which is an improvement of the ideal gas law), condensed matter and even the interior of stars or (at present) the accelerated expansion of the Universe: $P = \omega \rho$ (Weinberg, 1972). In this context, equation (5) is the EoS that describes plant cell growth at constant turgor pressure $P$, where the ‘state functions’ are the temperature and pH of the extending cell volume. Here pH, which is intimately connected with the acid growth hypothesis, even though it is not an intense variable in the usual sense, introduces a direct link between the physical variables ($P, V$) that represent the thermodynamic state of the system (growing cell or tissue) and biological response (via pH altering – H$^+$-ATPase), which results in growth. Clearly, equation (5) can be further verified by the diverse experiments that have been conducted for many species in order to determine characteristic triads ($\alpha, \beta, \Omega_S$) that belong to different species or families (classes) taxonomically, or that are simply controlled by growth factors (growth stimulators such as auxin, fusicoccin or inhibitors such as CdCl$_2$). However, the real test of the utility of Eq. (5) would include predictive outputs for any perturbations that could then be subject to experimental validation. From these triads, Eq. (5) should also be possible to connect with either the underlying physiological processes or their molecular drivers. In particular, whether the underlying molecular mechanism is identical or different should be reflected in the critical
exponents (at phase transition, see below). Despite the fact that $F$ is not a generalised homogeneous function, it can also be used to check for a kind of “universality hypothesis” (Stanley, 1971) and to validate the evolutionary paradigm with genuine numbers.

**Calculation of critical exponents**

Phase transitions occur in the thermodynamic limit at a certain temperature, which is called the critical temperature $T_c$, where the whole system is correlated (radius of coherence $\xi$ becomes infinite at $T = T_c$). We want to describe the behaviour of the function $F(pH, T)$, which is expressed in terms of a double power law by equation (3) that is close to the critical temperature and, specifically in our case for practical reasons, also about the critical $pH = pH_c$. In physics, critical exponents describe the behaviour of physical quantities near continuous phase transitions. It is believed, though not proven, that they are universal, i.e. they do not depend on the details of the system. Since growth (such as cell wall extension or the elongation growth of the shoots of grasses, coleoptiles or, hypocotyls) may be imagined as advancing over the course of time, a series of a quasi-continuous phase transitions, which are encouraged by the form of equation (3), the critical exponents may be calculated.

Let us introduce the dimensionless control parameter ($\tau$) for the reduced temperature

$$\tau = 1 - \frac{T}{T_c}$$

(7)

and similarly ($\pi$), for the reduced pH

$$\pi = 1 - \frac{pH}{pH_c}$$

(8)

which are both zero at the phase transition, and calculate the critical exponents. It is important to keep in mind that critical exponents represent the asymptotic behaviour at phase transition, thus offering unique information about how the system (here: the cell wall) approaches a critical point. A critical point is defined as a point at which $\xi = \infty$, so in this sense $T = T_c$ and $pH = pH_c$ are bi-critical points of Eq. (5). We believe that this significant property can help to determine the peculiar microscopic mechanism(s) that allows for wall extension (mode of extension) and growth in the future research. The information that is gained from this asymptotic behaviour may advance our present knowledge of these processes and their mechanisms and help to determine the prevailing one.
By substituting equations (7) and (8) into equation (3), we can calculate the critical exponents (when $T \rightarrow T_c$ and $pH \rightarrow pH_c$) from definition (Stanley, 1971), which will give us

$$\lambda_1 = \lim_{\tau \to 0} \frac{\log F(\tau, \pi)}{\log(\tau)} = \beta - 1$$

(9)

$$\lambda_2 = \lim_{\pi \to 0} \frac{\log F(\tau, \pi)}{\log(\pi)} = 1 - \alpha$$

(10)

and

$$\lambda_3 = \lim_{\tau \to 0} \lim_{\pi \to 0} \frac{\log F(\tau, \pi)}{\log(\tau) \log(\pi)} = 0$$

(11)

Here $\lambda_1 = \beta - 1$ and $\lambda_2 = 1 - \alpha$ are the critical exponents ($\lambda_3 = 0$). The above equations result in two power relations that are valid in the immediate vicinity of the critical points

$$F(\tau_\pi) \propto |\tau|^{\beta - 1}$$

(12)

$$F(\pi_\tau) \propto |\pi|^{1 - \alpha}$$

(13)

where lower indices denote constant magnitudes. Equations (12) and (13) represent the asymptotic behaviour of the function $F(\tau)$ as $\tau \to 0$ or $F(\pi)$ as $\pi \to 0$. In fact, we can observe singular behaviour at $T = T_c$ (Fig. 3a) and $pH = pH_c$ (Fig. 3b). At this bi-critical point, the following convexity relation (Stanley, 1971) holds $- \alpha + \beta \geq 2$.

Retaining $\tau$ for the reduced temperature, let us introduce another microscopic control parameter, $\mu$, for the chemical potential

$$\mu = 1 - \frac{\mu_{\mu^*}}{\mu_c}$$

(14)

Substituting $\tau$ and equation (14) into equation (6b) for $G(T, \mu)$ and assuming the reference potential $\mu_0 = 0$, we can calculate the critical exponents (when $T \rightarrow T_c$ and $\mu_{\mu^*} \rightarrow \mu_c$) to get

$$\lambda_1 = \lim_{\tau \to 0} \frac{\log G(\tau, \mu)}{\log(\tau)} = -2 - \alpha + 2\beta$$

(15)

$$\lambda_2 = \lim_{\mu \to 0} \frac{\log G(\tau, \mu)}{\log(\mu)} = 2 - \alpha$$

(16)

$$\lambda_3 = \lim_{\tau \to 0} \lim_{\pi \to 0} \frac{\log G(\tau, \mu)}{\log(\tau) \log(\pi)} = 0$$

(17)
The above limits result in another two power relations, which are valid in the immediate vicinity of the critical points

\[ G(\tau, \mu) \propto |\tau|^{2-\alpha+2\beta} \]  
\[ G(\mu, \tau) \propto |\mu|^{2-\alpha} \]  

Equations (18) – (19) represent the asymptotic behaviour of the function \( G(\tau, \mu) \) as \( \tau \to 0 \) or \( G(\tau, \mu) \) as \( \mu \to 0 \). At the microscopic level, we observe a singular behaviour at \( T = T_c \) (Fig. 4a) and \( \mu = \mu_c \) (Fig. 4b – c).

For the bi-critical point, the exact relation \( \beta = 2 \) holds (compare with SI Table 1 and SI Fig. 3), thus leaving us with the only free parameter (\( \alpha \)) of the theory. Note a broad peak at the critical temperature (optimal growth occurs within a certain temperature range) and a sharp, more pointed peak for the critical chemical potential (a single value that corresponds to the optimum growth). In this representation, growth can be treated as a series of subsequent phase transitions that take place in the cell wall. It looks as though a cardinal magnitude that is decisive for growth – in this particular phase transition approach – is the chemical potential. During the time of evolution of the extending cell wall, the chemical potential may form asymmetrical ratchet mechanisms, oscillating biochemical reactions, which are common in cell dynamics, that may be closely related to the emergence of the phenomenon of life itself (Martin et al., 2009). In this context, the relations for the chemical potential may also belong to the most fundamental dynamic constraints for the origin of life.

Complex systems such as the cell wall of a growing plant are solid and liquid-like at the same time. At the molecular level, they are both ordered and disordered. The equation of state explains the properties of the wall that can be described in general way. Whether the structure is ordered or disordered, universal features can be identified using simple scaling laws.

Discussion

Erwin Schrödinger (1944) may have been the first to consider the thermodynamic constraints within which life evolves, thereby raising fundamental questions about organismal evolution and development. Here, we attempted to present a more formal description of life couched in terms of the equation of state (EoS). More precisely, this work seeks to model the relationship between plant growth and temperature (or pH) in terms of the EoS, thereby describing growth.
optima as functions of state variables and critical exponents. Apparently, there is already a simple and compelling explanation for growth optima of plant growth from the biological point of view. Growth is mediated both directly and indirectly by enzymes and increases broadly in line with the frequently observed effect of temperature on enzyme activities (Berg et al., 2002). However, once temperatures are sufficient to denature proteins, enzyme activities rapidly decrease and plasma membranes and some other biological structures also experience damage (compare SI Fig. 1b at a high temperature). In this context, the kink that is observed at $T = T_c$ in Fig. 3a may coincide with a temperature-driven phase transition, thus pointing to the critical temperature $T_c$, which is in agreement with the interpretation of the enhancement in the effective diffusion rates (Pietruszka, 2012; Pietruszka and Haduch-Sendecka, 2015). On the other hand, the plot in Fig. 3b resembles a “lambda” kind of phase transition in which the state function $F$ tends towards infinity as pH approaches the lambda point, which is similar to the heat capacity diffusive transition (Münster, 1969; Matlak and Pietruszka, 2001). Note, at the low-pH end, the plot does not approach zero, but tends to the finite value of one, thus naturally preferring a lower pH-value regime (below pH$_c$) for growth, as is predicted by acid growth hypothesis, while nonetheless allowing for limited (though diminished) growth at pH > pH$_c$. A continuous phase transition can be observed at the critical points.

A magnitude that infinitesimal changes effect in symmetry change at phase transition always exist in continuous phase transitions (Landau and Lifshitz, 1980). Seemingly, such (order/disorder) symmetry changes (De Gennes, 1991) in biological systems may be connected with a mechanism (Rojas et al., 2011) by which chemically mediated deposition causes the turnover of cell wall cross-links, thereby facilitating mechanical deformation. It may also reflect the pectate structure and distortion that was suggested by Boyer (2009) (Fig. 5c, d) in the Chara cell walls by placing wall polymers in tension and making the load-bearing bonds susceptible to calcium loss and allowing polymer slippage that irreversibly deforms the wall. Proseus and Boyer (2007) suggested that the ladder-like structure would be susceptible to distortion and that the distortion would increase the distance between adjacent galacturonic residues (Fig. 5D in Boyer (2009)) and therefore the bonds may lengthen and thus weaken and decrease their affinity for Ca$^{2+}$. Dissociation may then occur, thus allowing an irreversible turgor-dependent expansion (see also Fig. 4 in Pietruszka (2013); this is also applicable for non-isochronous growth in pollen tubes). The direction of the maximal expansion rate is usually regulated by the direction of the net alignment among cellulose
microfibrils, which overcomes the prevailing stress anisotropy (Baskin, 2005). The transient changes in the wall composition and the deformation of bonds may be a hallmark of symmetry change and a kind of phase transition (orchestrated instability) that is taking place in the cell wall. A situation, which was already pointed out in the Introduction, in which the protons that are excreted into the wall compartment are directly responsible for wall-loosening processes through the hydrolysis of covalent bonds or the disruption of non-covalent bonds may be also a signature for the symmetry change and phase transitions that occur in the wall compartment of the growing cell.

Though the underlying microscopic mechanisms are not well recognised as yet (and as a result the order parameters are difficult to identify), a pH-driven “lambda” phase transition may be attributed to the maximum activity of PM H⁺-ATPase, while temperature-driven phase transitions in the cell wall can be directly related to the maximum elevation of the effective diffusion rate ($k_2$ coefficient in Pietruszka (2012)) – we have considered primary (diffusive) growth throughout this article. As an aside, from the calculation of cross-correlations, we may draw further conclusions that are connected with the biochemical picture of the acid growth theory. The result presented in SI Fig. 5 is a clear characteristic that temperature-induced growth and auxin-induced acidification (PM H⁺-ATPase) growth correlate strikingly well (there is good experimental evidence that higher temperature induces auxin biosynthesis in some plants). However, the convolution of acidic pH growth and temperature growth is less shifted away from zero (lag) and is even more pronounced (see also SI Fig. 6 for cell wall pH (proton efflux rate) and growth rate, which are directly co-regulated in growing shoot tissue, thus strongly supporting the foundations of EoS). This strict quantitative result may contribute to the acid growth hypothesis that was developed by plant physiologists and contribute to resolving problems that arise in the long-lasting discussion. However, a problem still arises. It is unclear to what degree Eq. (5) applies universally as the published raw data that constitute the experimental basis are only for grass shoot elongation growth.

We now show that our findings may be embedded in the evolutionary context that is connected with the migration of plants away from the Equator (changes in latitudes) as the climate changed and their adaptation to the spatial distribution of pH in the soil as a substitute for high temperature. As an example that exhibits the potential strength of equation (5), let us consider the following. The dominant factors that control pH on the European scale are geology (crystalline bedrock) in combination with climate (temperature and precipitation) as
was summarised in the GEMAS project account (Fabian et al., 2014). The GEMAS pH maps mainly reflect the natural site conditions on the European scale, whilst any anthropogenic impact is hardly detectable. The authors state that the results provide a unique set of homogenous and spatially representative soil pH data for the European continent (note Fig. 5, ibid.). In this context, the EoS that is expressed by equation (5) may also gain an evolutionary dimension when considering the spatial distribution of pH data as affected by latitude (angular distance from the Equator) elevation on the globe, which is associated with climate reflection (ibid.). pH is strongly influenced by climate and substrate – the pH of the agricultural soils in southern Europe is one pH unit higher than that in northern Europe. See also Fig. 4 in Fabian et al. (2013) or equivalently SI Fig. 7, in which the pH (CaCl$_2$) of soil samples that are grouped by European climate zones are measured (ranging (descending) from pH ≈ 7 (Mediterranean), pH ≈ 6 (temperate) through pH ≈ 5.25 (boreal) to pH ≈ 4.80 (sub-polar)). This fact may be reinterpreted in terms of pH – temperature duality that is considered in this work. When the migration of plants away from the Equator took place, lower pH values further to the North might have acted in a similar way as high temperatures at the tropic zones on the growth processes. This situation could have led to energetically favourable mechanisms (adaptation) such as the amplified ATP-powered H$^+$ extrusion into the expanding wall of plant cells and the secondary acidification of the environment (released protons decrease pH in the incubation medium), irrespective of the initial pH level, which is the main foundation for the acid growth hypothesis. Apparently, the evolutionary aspect of equation (5), which suggests a possible link with self-consistent adaptation processes ("The individual organism is not computed, or decoded; it is negotiated" (Walsh, 2010), as one of its first applications, seems difficult to underestimate, and the potentially predicative power of EoS looks impressive considering the modest input we made by deriving equation (5) and consequently the true thermodynamic relation that is given by equation (6b). In this perspective, the pH-response could potentially provide a plant with an adaptive advantage under unfavourable climate conditions.

**Perspectives** The EoS for a biological system has not been reported until this study. To further verify the EoS and its implications, we note that based on the results that were obtained (calculated critical exponents), a kind of “universality hypothesis”, which is known from the physics of phase transitions (Stanley, 1971), can be tested experimentally in order to extract “classes of universality” for plant species ($\Omega_S$), thereby substantiating the taxonomic divisions
in the plant kingdom via quantitative measures (SI Table 2). This issue, however, demands further experimentation and is beyond the scope of this work.

It seems the EoS may explain a number of phenomena and is powerful in its simplicity. It positions itself at the brink of biology and nano-materials. We believe it may be helpful in the description of the processes of the self-assembly of polymers. In agricultural implementations, any departures from the optimum growth may simply be corrected by appropriately adjusting pH or T.

The discovery of the EoS for plants adds an important dimension to the biophysical search for a better explanation of growth-related phenomena in a coherent way through its applicability to the visco-elastic (or plastic) monotonically ascending and asymptotically saturated (Pietruszka, 2012) cell wall extension that is usually observed, as well as to pollen tube oscillatory growth. The underlying biochemical foundation, which is expressed in the form of acid growth hypothesis (SI Fig. 6), could also help to understand the growth conditions that are ubiquitous in biological systems through coupling to EoS. In this respect, our method is accompanied by its partial validation – its application to the important biological question concerning the acid growth hypothesis. In the above context, it is not surprising that the growth of the cell wall in plants can be thought of as a time-driven series of phase transitions (via the breaking of polymer bonds or other mechanisms that were mentioned earlier), although they are ultimately connected with physics and chemistry by the equation of state (equations (5) or (6b)), which are solved at subsequent time instants by Nature. Since non-equilibrium stationary states are achieved after discrete intervals of time for single cells like pollen tubes, such a mechanism can lead to a kind of ‘leaps’, macroscopically (of μm scale) observed growth rate oscillations (Geitmann and Cresti, 1998; Hepler et al., 2001; Zonia and Munnik, 2007). This phenomenon may be treated as a result of the subsequent phase transitions that take place in the cell wall. (Spontaneous bond polarisation/breaking may be synchronised through Pascal’s principle by pressure P or pressure fluctuations δP (Pietruszka and Haduch-Sendecka, 2015), thereby acting as a long-range (of the order of ξ) messenger at the phase change (the radius of coherence $\xi \to \infty$ at a $\Gamma$-interface (Pietruszka, 2013)). When the entire system is correlated and the stress-strain relations are fulfilled (ibid.), the simultaneous extension of the cell wall at the sub-apical region may correspond to pollen tube oscillation(s). Hence, the uncorrelated or weakly correlated extension would result in the bendings that are observed in the extension of the growing tube. In multi-cell systems (tissues) that have a higher organisation (coleoptiles or
hypocotyls in non-meristematic zones), the equation of state for plants will manifest itself by the emergent action of an acidic incubation medium, or will be induced by endogenous auxin acidification, as is presumed in the acid growth theory. In this aspect, the EoS may serve as a new tool for the further investigation and verification of claims about the acid growth hypothesis. This should greatly facilitate the analysis of auxin-mediated cell elongation as well as provide insight into the environmental regulation of auxin metabolism. It also seemingly delivers a narrative that provides a biophysical context for understanding the evolution of the apoplast, thereby uncovering hidden treasures in the as yet unscripted biophysical control systems in plants.

Conclusions

Cross-disciplinary research at the interface between the physical and life sciences was accomplished in this work. We first considered the Euler beta function enigma, which is a duality of acidic pH (or auxin-induced acidification) and temperature, in order to obtain the equation of state (EoS) for living plants. We started from the striking similarity (mirror symmetry) between the elongation growth of grass shoots that were incubated in different pH environments or auxin-induced elongation in water (or a neutral buffer) with the respective growth at different temperatures. We based these on the hypothesis that the action of temperature on the elongation growth is effectively equivalent for the relative growth increments such as those that are caused by the change of pH or endogenous auxin-induced acidification in the incubation medium. In order to resolve this ambiguity, we first used the beta function for both dependences, which were normalised prior to the comparison, in order to obtain the beta function shape parameters $\alpha$ and $\beta$. It turned out that even without referring to biochemical underpinning, we might have concluded that the acidic conditions of the incubation medium or auxin-induced acidification and environmental temperature can act interchangeably, at least when they are considered at phenomenological level, i.e. from the effectiveness of the plant growth point of view. The numerically verified high accuracy of these complementary representations allowed us, among others, to treat this dual approach as a new apparatus for predicting the outcomes in the swapping growth conditions, which is especially useful in changing climate surroundings. We presumed that by applying a beta distribution and continuously changing its character with the values of the shape parameters, our findings might be related to the evolutionary context that is connected with the migration of plants away from the Equator as the climate changed and their adaptation to spatial distribution of pH in the soil as a substitute for high temperature. The equation of state may be
also helpful in delivering solutions (the respective (α, β, Ω) - triads) for the assisted migration of plant species (Vitt et al., 2010) that are at risk of extinction in the face of rapid climate change. In this application, the EoS tool can constitute a complementary method for genetic modifications in assisted migration processes.

In physics, it is believed (Fisher, 1966; Griffiths, 1970) that the critical exponents are ‘universal’, i.e. independent of the details of the Hamiltonian (energy function) that describes a system. The implications of this, however, are far reaching. One could take a realistic and complicated Hamiltonian, ‘strip’ it to a highly idealised Hamiltonian, and still obtain precisely the same critical exponents. For instance, on these grounds, it is believed that carbon dioxide, xenon and the three-dimensional Ising model should all have the same critical exponents (Baxter, 1989). This appears to be the case to within experimental error (Hocken and Moldover, 1976). In this context, since growth phenomenology and the phase transition approach have converged to a form for a universal critical equation of state for plants, the potential role of critical exponents to discriminate the different modes of wall extension (Breidwood et al., 2014 for review) of growing cells or tissues seems promising in opening new avenues of research.

By resolving the duality of low pH or auxin action (producing acidic pH) against temperature, not only have we introduced equations of state for the realm of plants, but also, by considering wall extension growth (life?) as a dynamic cascade of phase transitions that are identified critical exponents for this phenomenon, which exhibit a singular behaviour at critical temperature and critical pH (or a critical value of the chemical potential) in the form of power laws. Furthermore, universal (and exact) “scaling relations” (such as β = 2) were introduced that hold at the bi-critical point, which is in agreement with the experiment. The EoS, which is strongly predicative, can either be helpful for resolving food resource problems on Earth, or could be used as an analytical tool (calculator) for optimising fresh food production in the manned exploration of space.

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Figures captions

**Figure 1** Plot of the state function $F$ given by equation (3) showing the optimum conditions for plant cell/organ growth. Both coordinates temperature ($T$) and acidity or basicity (alkaline medium) $pH$ are rescaled to unity. The best conditions for maximum growth are found at (a) the saddle point (a stationary point) that corresponds to (b) the lines that cross in the contour plot or in the plot (c) of $F$ – derivative. The simulation parameters used like those for auxin-induced growth – see SI Table 1.

**Figure 2** The (a) isotherms (scaled values indicated) as a function of $pH$ (scaled) of the state function $F$, Eq. (3); (b) constant $pH$ curves as a function of temperature (scaled). Simulation parameters: $\alpha = 1.7$ and $\beta = 3.52$ (SI Table 1).

**Figure 3** Critical behaviour at phase transitions that take place in the cell wall ($pH$, $T$ – variables). (a) A “kink” (discontinuity in the first derivative) at the critical temperature $T = T_c$. (b) a “log-divergent” (“$\lambda$” – type) solution at the critical $pH = pH_c$. Control parameters: reduced temperature ($\tau$) and reduced $pH$ ($\pi$). Simulation parameters (a) $\beta = 1.93$ and (b) $\alpha = 1.7$ (SI Table 1).

**Figure 4** Critical behaviour at phase transitions that take place in the cell wall (for $\mu$, $T$ – intense state variables). (a) A “log-divergent” (“$\lambda$” – type) solution at $T = T_c$. (b–c) Solution at the critical value of the chemical potential $\mu = \mu_c$; (b) “acid growth”, (c) “auxin growth” (SI Table 1). Control parameters: reduced temperature ($\tau$) and chemical potential ($\mu$). Simulation parameters (a) $\alpha = 3.52$, $\beta = 1.93$; (b) $\alpha = 1.87$, $\beta = 3.17$; (c) $\alpha = 1.7$ and $\beta = 3.52$ (SI Table 1).
Figure 1
Figure 2
Figure 3
Figure 4