**Supplementary Information**

# Temporal integration of systemic signals governs biological size and phase transitions

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## Supplementary Note 1: Mathematical Foundations of the Epigenetic Integration Clock (EIC)

### (1) Spatial growth dynamics (the chalone model)

Traditional models, including the chalone hypothesis, assume that growth arrests when a systemic inhibitor reaches a spatial threshold. We first define the basic growth dynamics of muscle mass $M(t)$ under the inhibitory feedback of a Myostatin-like spatial limiter:

$$dM/dt = \alpha - \lambda M \quad (1)$$

where $\alpha$ is the intrinsic basal growth rate and $\lambda$ is the feedback coefficient proportional to the current muscle mass. Setting $dM/dt = 0$, growth intrinsically arrests at a steady-state mass $M_{SS} = \alpha/\lambda$. The analytical solution over time is:

$$M(t) = M_{SS} + (M_0 - M_{SS}) \exp(-\lambda t) \quad (2)$$

where $M_0$ is the initial mass. In this framework, the final tissue size is a strictly defined physical limit determined by the balance between $\alpha$ and $\lambda$. Because overall body size $S(t)$ scales with total muscle mass, final body size is approximated as proportional to $M_{SS}$. For the time dependence of $M(t)$, please refer to Extended data Fig. 1.

### (2) Mathematical decoupling of space and time via GDF8/GDF11 subfunctionalization

In vertebrates, two primary orthologs of insect Myo exists within the TGF-β family: Myostatin (GDF8) and GDF11 (Extended data Fig. 4). Strikingly, during vertebrate evolution, this integrated dual role of insect Myo underwent a complete subfunctionalization into two distinct paralogs[1]: Myostatin (GDF8) and GDF11. The subfunctionalization of Myo into Myostatin (GDF8) and GDF11 yields two coupled but functionally distinct mathematical modules:

**The Spatial Module (GDF8):** Myostatin (GDF8) functions primarily as a quantity limiter (spatial regulator). First, the spatial module mediated by Myostatin (GDF8) governs feedback

inhibition of muscle mass $M_i$ in tissue i:

$$S_{8,i}(t) \propto M_i(t) \quad (3)$$

where $S_{8,i}(t)$ represents the local size of tissue i, which is proportional to Myostatin-regulated muscle mass ($M_i$) in tissue *i*.

**The Temporal Module (GDF11):** GDF11 operates exclusively as the systemic regulator of developmental progression (spatiotemporal integrator). The systemic Epigenetic Integration Clock is driven exclusively by GDF11:

$$dA/dt = \gamma G_{11}(t) - \beta A \quad (4)$$

where

$$G_{11}(t) \propto \sum_i \omega_i M_i(t) \quad (5)$$

and

$$\sum_i \omega_i = 1 \quad (6)$$

In this formulation, exponential memory arises from chromatin turnover dynamics (governed by $\beta$), while the input signal $G_{11}(t)$ dynamically reflects the weighted systemic growth state of the organism.

This mathematical decoupling—removing GDF8 from the temporal integration equation—elegantly explains the phenotype of Myostatin (GDF8) null mutants[2]. In contrast to insects, in vertebrates, the loss of Myostatin (GDF8) abolishes the local size inhibition, leading to massive muscular hypertrophy. However, because the temporal integrator is driven solely by GDF11, which continues to be abundantly secreted from the hypertrophied muscles, the accumulation of epigenetic readiness ($A$) progresses normally. By delegating the temporal clock to GDF11, vertebrates were able to evolve diverse and extreme muscle allometries—such as the massive hypertrophy seen in certain dinosaurs or whales—without disrupting developmental schedules. In this view, GDF11 defines a systemic developmental competence window, while Myostatin (GDF8) independently shapes organ-specific spatial trajectories within that window. GDF11 plays essential roles in developmental progression, including axial patterning and organogenesis, and null mutations result in abnormal skeletal patterning and organ development[1,3].

The subfunctionalization of Myo into Myostatin (GDF8) and GDF11 yields two coupled but functionally distinct mathematical modules.

$$A(t) = \int_0^t e^{-\beta_i(t-\tau)} \gamma\, G_{11}(\tau) d\tau \quad (7)$$

In this formulation, exponential memory arises from chromatin turnover dynamics (governed by $\beta$), while the input signal $G_{11}(\tau)$ dynamically reflects the weighted systemic growth state of

the organism.

In this view, GDF11 defines a developmental competence window along the body axis, while Myostatin (GDF8) shapes organ-specific growth trajectories within that window. This mathematical decoupling—removing GDF8 from the temporal integration equation (Eq. 11)—elegantly explains the phenotype of *Myostatin (GDF8)* null mutants. In insects, null mutation of the single *myo* gene abolishes temporal integration, causing severe developmental arrest.

**Supplementary Note 2: Determination of Insect Body Size by Myoglianin (Myo)**

**(1) Experimental data on roles of Myo in hemimetabolous insects**

We have investigated insect development and evolution using the field cricket *Gryllus bimaculatus* (*G. bimaculatus*)[4]. Crickets are hemimetabolous insects, meaning they develop into adults without a pupal stage. Unlike holometabolous insects such as *Drosophila* or butterflies, their relatively gradual morphological changes allow body size to be readily observed throughout postembryonic development.

We demonstrated that Myoglianin (Myo) plays a central role in determining both the timing of metamorphosis (final molt) and final body size in crickets[5]. Myo was originally identified in *Drosophila* based on its expression in embryonic muscles and glial cells[6]. In *G. bimaculatus*, Myo acts on the corpora allata (CA) to suppress expression of *juvenile hormone acid methyltransferase* (*jhamt*), thereby reducing juvenile hormone (JH) biosynthesis and triggering the transition to adulthood (Extended data Fig. 2)[5]. This endocrine pathway provided the first evidence that Myo signaling can regulate organismal developmental progression via systemic hormonal control[7]. RNA interference (RNAi) mediated reduction of *myo* expression in crickets induces additional molts and produces markedly enlarged adults (Fig. 1)[5]. These phenotypes indicate that Myo is a key determinant of final body size during postembryonic development. Supporting this conclusion, studies in another hemimetabolous insect, the cockroach *Blattella germanica*, have also linked Myo signaling to developmental timing through the JH system[8].

**(2) Experimental data on roles of Myo in holometabolous insects**

In holometabolous insects, Myo likewise contributes to the definition of the critical size checkpoint that permits the initiation of the metamorphosis program, as shown in the Tobacco hornworm Manduca sexta[8] and the beetle *Henosepilachna vigintioctopunctata*[9]. Recent comprehensive reviews highlight that Myo is not merely a spatial growth brake, but a critical licensing signal that links muscle mass accumulation to the endocrine and epigenetic cascades driving metamorphosis.

As the larva grows and reaches a threshold weight, the elevated Myo secreted from skeletal muscles acts on the CA to suppress JH biosynthesis, while also potentiating prothoracic gland activity to release ecdysone. This systemic decline in JH is essential to remove the Kr-h1-mediated maintenance of the larval specifier *chinmo*. Concurrently, Myo and ecdysteroids are implicated in inducing the expression of the pupal specifier broad.

Without this Myo-driven endocrine shift, larvae fail to undergo metamorphosis. For instance, in the beetle *Tribolium castaneum*, RNAi-mediated knockdown of *myo* in penultimate-stage larvae results in permanent larvae; they grow well beyond the normal threshold weight and continue to molt into giant larvae, effectively uncoupling physical growth from developmental timing. Thus, although the developmental stage at which size is assessed differs between hemimetabolous and holometabolous insects, Myo signaling as a temporal integrator of developmental competence represents a strictly evolutionarily conserved framework.

**(3) Reconsidering the fundamental principles of insect size determination**

Building upon the conceptual framework proposed by Texada et al[10]. we adopted the formulation that the final size of an insect is determined by the product of its growth rate and growth duration. The final size of an insect is generally determined by the product of its growth rate and growth duration.

$$S \text{ (size)} = R \text{ (growth rate)} \times D \text{ (duration time)} \qquad (12)$$

Their review[10] provides comprehensive insights into insect size determination and growth regulation. However, while their reviews have emphasized the modulation of growth rate (*R*), they have not fully resolved the paradox between partial and complete pathway perturbations observed in signaling systems. Growth rate (*R*) is mainly governed by the insulin/TOR signaling pathway (IIS/TOR), where the fat body senses nutritional status and regulates the secretion of insulin-like peptides (ILPs) from the brain via various adipokine signals[11].

In crickets, we reported that the adult body size can be altered by systemic RNAi knockdown of *Gryllus insulin receptor* (*InR*), *insulin receptor substrate* (*chico*), *target of rapamycin* (*Tor*), *ribosomal proteinp70-S6 kinase* (*S6k*), *forkhead box O* (*FoxO*) and *epidermal growth factor receptor* (*Egfr*)[12], indicating that the IIS/TOR and EGF pathways contribute to cricket body size determination.

Conversely, the regulatory basis of growth duration (*D*) that determines when to terminate growth remains poorly understood. Our studies in crickets suggest that muscle-derived Myo is not just a size-dependent inhibitor but a factor that connects growth-generated signals to the endocrine system to define the conditions for developmental phase transition[13]. The

divergence between RNAi and KO phenotypes therefore suggests moving beyond simple "quantity thresholds" models toward a new framework based on temporal integration or history-dependent control.

The EIC decodes classical endocrine puzzles for classical surgical manipulations of *G. bimaculatus* perfectly mirror our model's logic. Allatectomy (CA removal) in early instars causes precocious metamorphosis19; in our framework, this equates to the physical destruction of the JH brake, bypassing the integration clock entirely and forcing an immediate, premature phase transition. Conversely, CA implantation into late-instar nymphs—which artificially sustains high JH levels—results in supernumerary molting and gigantism[19]. This occurs because the downstream brake is exogenously maintained, effectively ignoring the fact that the internal clock $A(t)$ has already surpassed Θ.

**(4) Integration with Contemporary Dynamical Models**

Recently, Tyson et al.[14] proposed a sophisticated dynamical model of *Drosophila* growth and maturation using coupled differential equations. Their work represents a significant advance in viewing metamorphosis as arising from continuous physiological states rather than a static size-threshold event. However, such models often rely on complex, species-specific variables.

In *Drosophila*, Myo has been proposed to function as a Myokine that mediates allometric relationship between muscles and associated appendages[15], although the relationship between Myo and JH may have diversified across insects. Thus, the role of Myo in insects can be coherently interpreted within our "Time-Integration Model", offering a parsimonious and evolutionarily grounded principle. By focusing on the accumulation of Myo signaling as a single integration variable $A(t)$, this framework can account for developmental dynamics in insects and suggests a potentially general mechanism of size determination and phase transitions across distant lineages.

**Supplementary Note 3: Epigenetic integration mechanisms in plants**

**(1) Vernalization**

The concept of the EIC provides a universal framework for understanding how organisms measure extended physiological time. A mathematically isomorphic system is well-documented in plants during vernalization: the prolonged exposure to cold temperatures is continuously integrated at the *FLC* locus via the gradual accumulation of Polycomb-mediated H3K27me3 repressive marks. Once this epigenetic silencing reaches a critical threshold, the plant is licensed to flower[16,17].

In this plant system, the systemic input variable shifts from internal body size to external environmental intensity (e.g., the magnitude of cold exposure, $E$, during winter). Consequently, the temporal integration yields a parallel rule: $E \cdot D \approx$ constant. This mathematical relationship perfectly explains the widely observed ecological phenomenon—seen in both Arabidopsis and peach blossoms[18]—where a more severe winter (higher $E$) accelerates the epigenetic silencing of floral repressors, requiring a shorter duration ($D$) to reach the critical threshold[19] and resulting in earlier spring blooming. Thus, macroscopic agricultural rules, such as "accumulated cold/temperature" (degree-days), and the animal epigenetic Area-Constancy Rule are fundamentally isomorphic manifestations of the same epigenetic integration mechanism.

**(2) NTL8 and Growth-Dependent Dilution**

Recent studies have revealed that the long-term cold integration in plants is initiated by the transcription factor *NTL8*. Strikingly, NTL8 acts as a temperature integrator not through direct thermo-sensing, but through "growth-dependent dilution"[18]. NTL8 is constitutively synthesized at a constant rate. During warm periods, rapid cell division and volumetric expansion dilute the NTL8 protein, keeping its concentration low. However, during winter, low temperatures halt cellular growth. Without this volumetric dilution, NTL8 gradually accumulates over time, effectively translating the duration of growth arrest into a high-concentration biochemical signal. This accumulated NTL8 then activates VIN3, triggering the PRC2-mediated epigenetic silencing of *FLC*[20,21]. In the context of the EIC framework, this elegant mechanism demonstrates that even environmental clocks are physically anchored to spatial size dynamics; the system integrates the absence of spatial expansion to measure biological time.

**(3) Mechanistic Explanation of the Source-Sink Ratio Rule in Fruit Growth**

The Epigenetic Area-Constancy Rule (EACR) relationship, expressed as $S = 2\Theta R/k$, provides a flawless mechanistic explanation for the "source-sink ratio" rule widely recognized in agronomy[22,23]. In a favorable, nutrient-rich environment (characterized by a high growth rate $R$), the fruit rapidly accumulates metabolic signals and reaches the epigenetic threshold ($\Theta$) for ripening early, which shortens the maturation duration ($D$). However, because the physical growth rate ($R$) outpaces this temporal shortening, the final fruit size ($S$) inherently becomes larger. This apparent paradox—a trade-off between accelerated maturation and increased final size—is a mathematical inevitability of our EIC/EACR model, wherein the metabolic signal itself functions as the primary integrator of the developmental clock[24,25].

**(4) The Thermal/Hydrotime models for seed germination**

Similar to animal development and plant vernalization, seed germination is governed not by an instantaneous trigger, but by the continuous integration of environmental history.

The classical agronomic Thermal-time model for seed germination, mathematically expressed as $(T - T_\beta) \times t_g = \Theta_T$. Similar to animal development and plant vernalization, seed germination is governed not by an instantaneous trigger, but by the continuous integration of environmental history. This empirical formula perfectly aligns with the fundamental EACR equation through the following parameter mapping:

- Stimulus Intensity ($S$) corresponds to the effective temperature $(T - T_\beta)$ where $T$ is the current environmental temperature and $T_\beta$ is the base temperature. This represents the continuous intensity of the active environmental input or acquired energy capacity (e.g., sugars).
- Duration ($D$) is equivalent to the time required for germination ($t_g$).
- Epigenetic Threshold ($\Theta$) corresponds to the thermal time constant $\Theta_T$. This constant mathematically represents the critical accumulation of epigenetic remodeling (such as progressive H3K27me3 accumulation) required to irreversibly release dormancy and license the developmental phase transition.

Seeds continuously integrate these modular environmental inputs over time. Under highly favorable conditions (a large $(T - T_\beta)$), the epigenetic integration clock ticks faster, requiring a shorter duration ($t_g$) to reach the necessary threshold ($\Theta$). Conversely, under marginal conditions, the clock slows down, necessitating a proportionally longer duration. This intrinsic trade-off perfectly maintains the constant "area" (Intensity × Time), demonstrating that the classical models of seed germination naturally emerge from the exact same mathematical logic of epigenetic temporal integration shared across distantly related taxa.

**Supplementary Note 4: Biophysical Basis of the Epigenetic Threshold (Θ): Liquid-Liquid Phase Separation (LLPS)**

In the EIC model, the epigenetic threshold ($\Theta$) is not merely an abstract mathematical constant, but a critical physical transition point governed by the biophysics of liquid-liquid phase separation (LLPS). As the epigenetic integration proceeds—driven by continuous systemic signals—the local density of repressive chromatin marks (such as H3K27me3) steadily increases at the target gene locus.

Once this density reaches a critical point ($\Theta$), multivalent chromatin-binding proteins recognize these marks and undergo spontaneous LLPS, driving the target locus into a highly

compacted, transcriptionally inaccessible condensate. This mechanism is highly conserved across kingdoms. In plants, during vernalization, the continuous accumulation of H3K27me3 at the *FLC* locus recruits LHP1 (Like Heterochromatin Protein 1), which undergoes LLPS to form a stable silencing condensate, thereby irreversibly locking the "memory of winter" (Borg et al., 2021). Similarly, in animals, the accumulation of H3K27me3 recruits PRC1 complex components (such as CBX2), which undergo LLPS to physically compact developmental genes[26]

Thus, the EIC threshold (Θ) mathematically represents the exact thermodynamic tipping point where the integrated epigenetic driving force overcomes local chromatin resistance, triggering an irreversible macroscopic phase transition (LLPS) that physically actualizes the biological phase change.

**Supplementary Note 5: Evolutionary Conservation of the TGF-β–Terpenoid Regulatory Axis**

**(1) The TGF-β–terpenoid regulatory axis (Extended data Table 1)**

The dual role of TGF-β signaling in governing both physical size and developmental transitions is deeply rooted in bilaterian evolution. In the nematode *C. elegans*, these roles are performed by two distinct TGF-β pathways[27,28]: the DBL-1 pathway acts as a spatial size regulator[29], while the DAF-7 pathway functions as a temporal phase switch, determining reproductive development versus dauer arrest by regulating the synthesis of the steroid hormone dafachronic acid (DA)[30].

This functional bisection in nematodes appears perfectly integrated in the insect Myo system. In *G. bimaculatus*, a single ligand, Myo, links spatial size sensing to phase transition control into a unified molecular axis. Myo secreted from muscles acts on the CA to suppress JH biosynthesis, thereby acting as a systemic gating mechanism that synchronizes developmental phase transitions across the entire organism.

The downstream endocrine effectors executing these phase transitions are remarkably conserved. While vertebrates lack a direct equivalent to insect JH, retinoic acid (RA) signaling plays a functionally comparable systemic role in developmental timing and axial patterning[28]. Crucially, DA, JH, and RA are all derived from the isoprenoid (terpenoid) biosynthetic pathway and function as lipophilic ligands for nuclear receptors. Just as insect Myo suppresses JH synthesis, vertebrate GDF11 modulates RA availability (e.g., via the RA-degrading enzyme CYP26B1, which controls growth plate chondrocyte maturation)[31,32]. Thus, from invertebrates to vertebrates, the underlying architectural principle—linking TGF-β signaling to terpenoid-

based endocrine control of developmental phase transitions—remains strictly conserved

**(2) Epigenetic Integration in Vertebrates**

In vertebrates, we review evidence suggesting an epigenetic cascade analogous to that described in plant vernalization[33]. Most studies of Myostatin (GDF8) focus on muscle phenotypes: *Myostatin* (*GDF8*) knockout mice[2] and mutant mice[34] exhibit pronounced muscular hypertrophy.

Integrated GDF11 signaling regulates RA activity in the posterior embryonic region in part by maintaining expression of the RA-degrading enzyme *Cyp26a1*. Disruption of this regulatory axis perturbs epigenetic control of *Hox* gene expression (the *Hox* code), producing anterior transformations of vertebral identity[31]. This provides a clear example of how temporally integrated signaling can rewrite developmental epigenetic programs through modulation of RA levels.

In the vertebrate growth plate, RA acts as a powerful accelerator of chondrocyte maturation. A local increase in RA signaling strongly upregulates the expression of *Runx2*, the transcription factor for chondrocyte hypertrophy[32]. This Runx2 activation drives terminal chondrocyte maturation and epiphyseal closure, thereby finalizing skeletal size. Thus, the temporal integration of GDF11, which modulates RA availability via CYP26B1, acts as the systemic trigger for this Runx2-mediated growth cessation (Fig. 3).

Interestingly, RA can induce digit formation in chicken limb buds[35]; local RA application mimics the polarizing region. Although RA was initially proposed to act as a direct morphogen for digit patterning, subsequent work demonstrated that RA does not function as a classical morphogen[36] but instead activates a digit-forming program in *Hoxd11-13* that is normally epigenetically repressed[37].

During vertebrate development, when cumulative GDF11 signaling reaches a threshold, chromatin states of posterior *Hox* genes shift from closed to open configurations, driving transitions from thoracic to lumbar/sacral identities. Gaunt et al.[38] proposed that GDF11 controls the timing of posterior *Hox* (*Hox10-13*) activation, while Matsubara et al.[39] and Aires et al.[40] showed that GDF11 alters chromatin accessibility at *Hox* loci, promoting trunk-to-tail program transitions. These processes are conceptually analogous and functionally comparable to epigenetic *FLC* silencing during plant vernalization. Thus, sustained signal input reflecting growth history may be recorded as cumulative epigenetic memory.

We therefore propose that the animal Myo/GDF11 system operates through an isomorphic epigenetic principle: GDF11 converts development time into spatial pattern (vertebrae number and identity) by altering *Hox* chromatin states. Although many aspects of GDF11 biology remain unresolved[3], this framework offers testable predictions.

In insects, strong evidence indicates analogous epigenetic transitions downstream of Myo signaling. The metamorphic master regulator E93 functions as a pioneer factor capable of large-scale chromatin remodeling[41,42], and E93-dependent metamorphic competence can occur independently of body size per se[43,44]. Ureña et al.[45] proposed that E93 rewrites the epigenetic state of metamorphic target genes during the larval-to-adult transition. Within our model, attainment to the Myo integral threshold triggers E93 activation and coordinated opening of developmental chromatin programs across tissues.

In this view, Myo/GDF11 signaling does not merely report instantaneous size; it drives an epigenetic integrator that converts growth history into a stable endocrine command for maturation. This perspective resonates with the epigenetic clock framework[46], in which DNA and histone methylation patterns encode cumulative biological time.

**Supplementary Note 6: Biophysical Rationale for H3K27me3 as the Physical Integrator of the EIC**

The requirement of the EIC for a stable temporal integrator naturally points to H3K27me3 over other epigenetic marks. The biological phase transitions governed by the EIC, such as metamorphosis or flowering, are typically triggered by the ultimate de-repression of developmental programs—specifically, by the permanent silencing of juvenile-maintaining genes (e.g., *jhamt* in insects, *FLC* in plants). This necessitates an inhibitory epigenetic mark[47-49].

Furthermore, the physical clock must filter out short-term environmental noise while retaining long-term signal memory[48,50]. Active marks like histone acetylation or H3K4me3 turn over too rapidly (acting as transient RAM) to integrate developmental time over weeks. Conversely, DNA methylation or H3K9me3 are often too rigidly irreversible (acting as read-only memory) to function as a responsive, signal-dependent continuous gear[47].

H3K27me3, deposited by the PRC2 complex (specifically the E(z)/EZH2 catalytic subunit), uniquely satisfies the mathematical requirements of the EIC ($S \cdot D \approx$ constant). PRC2 exhibits

a self-amplifying "read-and-write" mechanism, allowing the repressive domain to slowly spread across the target locus in a signal-dependent manner. This spatial spreading of chromatin modification across the 1D genomic sequence is the exact physical manifestation of the mathematical integration ($\int S \cdot D$). Once the accumulated density of H3K27me3 reaches a critical conformational threshold ($\Theta$), it induces a bistable, irreversible silencing of the locus, flawlessly executing the digital phase transition predicted by our model.

Crucially, this PRC2-mediated spreading does not occur in a vacuum; it is actively antagonized by active chromatin marks, particularly H3K36me2 deposited by enzymes such as NSD2[51,52]. H3K36me2 acts as a powerful molecular friction that allosterically inhibits PRC2 activity, preventing premature silencing. Thus, the EIC timer progresses only as integrating signals (like Myo) progressively downregulate this NSD2-mediated barrier, ultimately permitting PRC2 to overcome the friction and reach the phase-transition threshold.

During *Gryllus* embryogenesis the Polycomb group gene *E(z)*—the epigenetic writer of H3K27me3—is strictly required to establish the spatial domains of Hox genes in a time-dependent manner during posterior elongation, mirroring the vertebrate mechanism38. Crucially, this temporal integration dictates local spatial patterning as well.

While diverse stage-specific signals—such as Dpp/Gbb during early instars—serve as the initial inputs to drive early developmental progression, muscle-derived Myo ultimately takes over as the definitive systemic signal in the third instar, driving the final epigenetic integration required for metamorphosis. This take-over clearly interprets the phenotypes of the myo KO crickets which grow up normally to the third instar.

In the regenerating cricket leg, knockdown of *E(z)* abolishes the forward integration rate, stalling the epigenetic clock and causing the continuous expansion of the proximal developmental phase (extra-tibia formation)[39]. Conversely, loss of the H3K27 demethylase *Utx* eliminates the epigenetic turnover[39], artificially hastening the clock and resulting in truncated distal structures. Thus, the continuous leaky integration of epigenetic time dictates both microscopic tissue patterning and macroscopic organismal maturation.

**Supplementary Note 7: Implications for Allometry, Lifespan, and Rejuvenation**

**(1) Allometry, Lifespan and Rejuvenation**

The EIC framework not only explains developmental timing but also provides a precise mechanistic foundation for universal allometric scaling laws, such as the quarter-power

scaling rules linking body mass ($M$) and lifespan $LS$ ($LS \propto M^{1/4}$) across taxa.

We propose that the EIC does not stop ticking after metamorphosis or sexual maturation. Instead, the continuous integration of systemic physiological signals—reflected as the progressive accumulation of epigenetic marks (e.g., DNA methylation drift)—persists throughout adulthood. In this context, aging can be defined as the continuous temporal integration toward a final, terminal threshold ($\Theta_{terminal}$), at which point systemic senescence is triggered.

Our area-constancy relationship provides a novel mechanistic explanation for this universal allometry. Biologically, it is reasonable to postulate that the absolute epigenetic integration threshold scales linearly with final body size or mass ($\Theta \propto M$), as a larger organism requires a proportionally larger total systemic signal (due to isometrically scaling blood and extracellular fluid volumes) to coordinate phase transitions[53]. Furthermore, the rate of epigenetic integration (the ticking speed of the clock, or the sensitivity parameter $k$) must be intimately linked to the mass-specific metabolic rate, which scales as $M^{-1/4}$, [54,55].

Substituting these scaling constraints into our temporal integration equation ($k \times M \times D \propto \Theta$) yields.

$$M^{-1/4} \times M \times D \propto M$$

Solving for developmental duration yields.

$$D \propto M^{1/4}$$

Similarly, integrating this mass-dependent clock rate over the lifespan to reach the terminal threshold ($\Theta_{terminal}$) naturally derives the universal allometric lifespan law: $LS \propto M^{1/4}$. This suggests that species-specific developmental times and lifespans are mathematically dictated by the metabolic efficiency of recording temporal information.

By defining aging as the continuous temporal integration toward a terminal threshold, the EIC model clarifies that extending lifespan and achieving systemic rejuvenation are merely two distinct directional interventions—decelerating versus reversing—on the exact same epigenetic timer. Ultimately, shifting the paradigm from static size-sensing to continuous epigenetic time-integration elegantly unifies the microscopic molecular drivers of development with the macroscopic evolutionary laws of longevity.

**(2) Derivation of the Constant Lifetime Heartbeat Rule and Relativistic Biological Time**

The EIC framework further provides a mechanistic and mathematical basis for one of the

most intriguing allometric invariants in mammalian biology: the empirical observation that the total number of heartbeats per lifetime is remarkably constant across species (approximately $1.5 \times 10^9$ beats)[56], irrespective of body size. If we define aging and natural death as the ultimate developmental phase transition, it is triggered when the lifetime accumulation of metabolic signals reaches a terminal epigenetic threshold ($\Theta_{aging}$). The ticking rate of this clock—the systemic mass-specific metabolic rate—is directly proportional to the heart rate ($HR$). Thus, the EIC integral for lifespan ($LS$) becomes: $\int_0^{LS} HR(t)dt = \Theta_{aging}$. Assuming a relatively constant adult heart rate, this approximates to $HR \times LS \approx \Theta_{aging}$. According to quarter-power allometric scaling laws, heart rate scales as $M^{-1/4}$, [57] while lifespan scales as $M^{1/4}$. Taking their product cancels the mass dependency completely: $HR \times LS \propto M^{-1/4} \times M^{1/4} = M^0 = 1$. Consequently, the total lifetime heartbeat converges to a mass-independent constant, mathematically defined by the absolute epigenetic threshold ($\Theta_{aging}$). This derivation highlights a profound philosophical and physical implication of the EIC model: time, for biological organisms, is not an absolute Newtonian calendar. Rather, it is a relativistic dimension intrinsically scaled by the pace of metabolic and epigenetic integration.

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