RESEARCH PAPER

Gene Regulatory Networks and Molecular Mechanisms of Embryogenesis in Model Organisms: Pharmaceutical Perspectives and Therapeutic Implications

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ABSTRACT

Embryonic development is a highly coordinated and tightly regulated process governed by precise spatial and temporal control of gene expression. Model organisms, including Drosophila melanogaster, Caenorhabditis elegans, Danio rerio, and Mus musculus, have significantly advanced our understanding of the molecular mechanisms underlying developmental gene regulation. These systems have revealed conserved genetic pathways that regulate cell fate determination, tissue patterning, and organogenesis. This review comprehensively discusses the key regulatory mechanisms controlling gene expression during embryogenesis, including transcriptional regulation, epigenetic modifications, chromatin remodeling, and post-transcriptional processes. Special emphasis is placed on gene regulatory networks (GRNs), morphogen gradients, and major developmental signaling pathways such as Wnt, Notch, Hedgehog, and BMP/TGF-β, which collectively orchestrate embryonic pattern formation and cellular differentiation. Recent advancements in high-throughput and integrative technologies, including single-cell transcriptomics, CRISPR–Cas genome editing, and computational modeling, have enabled deeper insights into the dynamic regulatory landscapes of early development. Comparative analyses across model organisms highlight the evolutionary conservation of these regulatory mechanisms and their relevance to human developmental biology. Understanding these complex regulatory systems provides critical insights into the molecular basis of congenital disorders and supports the development of novel strategies in regenerative medicine and developmental therapeutics.

Keywords: Embryonic development; Gene regulation; Model organisms; Gene regulatory networks; Developmental signaling pathways; Epigenetic regulation

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INTRODUCTION

Embryonic development is a highly intricate and precisely coordinated biological process through which a single fertilized cell gives rise to a complex multicellular organism composed of specialized tissues and organs [1]. This transformation is directed by tightly controlled spatial and temporal patterns of gene expression that regulate essential cellular events such as proliferation, differentiation, migration, and tissue organization [2]. The accurate modulation of gene activity is crucial for ensuring that developmental processes occur in a sequential and orderly fashion, allowing cells to attain specific identities and functional roles during embryogenesis [3]. Any disturbance in these regulatory systems may result in developmental defects and congenital abnormalities, emphasizing the critical need to understand the mechanisms governing gene expression during early development [4].

Over the years, extensive research using model organisms has significantly advanced our understanding of the molecular basis of embryonic gene regulation [5], [6], [7]. Experimental systems such as *Drosophila melanogaster*, *Caenorhabditis elegans*, *Danio rerio* (zebrafish), and *Mus musculus* (mouse) are widely used due to their genetic manipulability, well-defined developmental stages, and conservation of key biological pathways. Investigations in these models have led to the identification of crucial developmental regulators, including transcription factors and signaling cascades involved in embryonic pattern formation. Notably, pathways such as Wnt, Notch, Hedgehog, and BMP/TGF-β play pivotal roles in orchestrating developmental processes and are highly conserved across species, contributing significantly to both normal human development and disease pathogenesis [8], [9], [10].

Gene regulation during embryogenesis operates through multiple interconnected levels of control. These include transcriptional regulation driven by transcription factors and cis-regulatory DNA elements, epigenetic mechanisms such as DNA methylation and histone modifications, chromatin remodeling processes, and post-transcriptional regulation affecting RNA maturation and stability [11], [12], [13]. Collectively, these mechanisms form complex gene regulatory networks that integrate intracellular signals and positional cues to direct developmental outcomes. Morphogen gradients and signaling pathways further contribute to spatial organization by enabling cells to interpret positional information and activate specific gene

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expression programs. Recent advancements in technology have greatly enhanced our capacity to investigate gene regulation during development [1], [14], [15]. Techniques such as high-throughput sequencing, including single-cell transcriptomics and epigenomics, allow detailed analysis of gene expression patterns at high resolution. Additionally, genome editing technologies like CRISPR-Cas systems provide powerful tools for targeted manipulation of genes, facilitating in vivo functional studies. Computational and systems biology approaches are also being increasingly applied to reconstruct regulatory networks and predict developmental behaviors [16].

Despite these advancements, several aspects of embryonic gene regulation remain poorly understood, particularly the integration of diverse regulatory mechanisms that coordinate developmental events across both spatial and temporal dimensions [17]. Gaining a deeper understanding of these processes is essential not only for uncovering fundamental principles of developmental biology but also for enabling advances in biomedical fields such as regenerative medicine and the management of congenital disorders [18]. This review aims to present a comprehensive overview of the molecular mechanisms underlying gene regulation during embryonic development, with a particular focus on findings derived from model organisms. It highlights key areas including transcriptional and epigenetic regulation, gene regulatory networks, developmental signaling pathways, and emerging technologies that continue to transform our understanding of embryogenesis.

2. ORGANISMS AS PLATFORMS FOR STUDYING DEVELOPMENTAL GENE REGULATION

Model organisms have been instrumental in deepening our understanding of gene regulation during embryonic development. These systems offer experimentally manageable platforms that enable researchers to explore fundamental biological processes under well-controlled conditions [1]. Their application in developmental biology has facilitated the discovery of conserved genetic pathways, regulatory circuits, and signaling mechanisms that govern embryogenesis. Notably, many of these regulatory frameworks are evolutionarily conserved, underscoring the relevance of model organisms in studying human development and associated diseases [19]. The emergence of advanced technologies, including high-throughput sequencing and single-cell analytical approaches, has further refined our ability to examine these regulatory processes at an unprecedented level of detail [20]. Nevertheless, the inherent complexity of developmental systems, especially those characterized by extended lifespans and large genomes, poses significant challenges in fully perturbing gene networks and extrapolating findings from traditional model systems [21]. Despite these limitations, comprehensive transcriptional profiling across a range of embryonic models—from invertebrates such as *Drosophila* and nematodes to vertebrates including zebrafish, mice, and humans—has established a strong framework for comparative developmental analyses.

A key advantage of model organisms is their high degree of genetic tractability. Many widely used models possess well-characterized genomes, rapid life cycles, and established methodologies for genetic manipulation [22]. These attributes enable the application of techniques such as gene knockouts, transgenic modifications, and targeted mutagenesis to investigate gene function during development. Furthermore, recent advancements in genome editing technologies, particularly CRISPR–Cas systems, have significantly enhanced the precision with which developmental genes can be manipulated, thereby advancing functional genomics research in embryogenesis [23].

Another significant strength of model organisms lies in their relatively simplified and thoroughly characterized developmental processes [24]. In several models, embryonic development has been mapped with remarkable precision, allowing detailed tracking of cell lineage specification and differentiation. Additionally, the presence of transparent embryos in certain species facilitates real-time imaging of developmental events, enabling direct observation of gene expression dynamics, cellular movements, and tissue morphogenesis during early stages [25]. When combined with single-cell molecular profiling techniques, these approaches provide in-depth mechanistic insights that can be translated to human developmental biology through comparative genomic analyses [26].

Research utilizing model organisms has been pivotal in uncovering fundamental principles that regulate embryonic development. Classical genetic screening approaches in organisms such as fruit flies and nematodes have led to the identification of numerous genes involved in body patterning, segmentation, and organogenesis [27]. Likewise, vertebrate models including zebrafish and mice have provided valuable insights into complex signaling pathways and gene regulatory networks that are directly relevant to mammalian and human systems [28].

The evolutionary conservation of developmental genes further emphasizes the importance of model organisms in biological research. A wide range of transcription factors, signaling pathways, and regulatory mechanisms initially identified in model systems have subsequently been validated in human embryonic development [29]. As a result, model organisms serve not only as essential tools for elucidating basic developmental mechanisms but also as critical platforms for investigating the molecular basis of congenital abnormalities and developmental disorders.

3. MAJOR MODEL ORGANISMS IN EMBRYONIC DEVELOPMENT

Model organisms have significantly advanced our understanding of the genetic and molecular frameworks that control embryonic development. These systems provide distinct experimental benefits, such as well-defined developmental stages, ease of genetic manipulation, and conservation of key regulatory pathways across species [24]. Investigations in various model organisms have led to the elucidation of core principles underlying gene regulation, cellular differentiation, and tissue organization. Among the most extensively utilized models in developmental biology is *Drosophila melanogaster*, *Caenorhabditis elegans*, *Danio rerio*, and *Mus musculus*, each contributing uniquely to the study of embryogenesis and gene regulatory networks [30]. A comparative overview of these commonly used model organisms and their experimental advantages is presented in Table 1.

3.1 *Drosophila melanogaster*

The fruit fly *Drosophila melanogaster* represents one of the most extensively investigated model organisms in the field of developmental genetics [31]. Its compact genome, rapid life cycle, and availability of sophisticated genetic tools make it an ideal system for studying gene regulation during early embryonic stages [31]. Pioneering genetic studies in *Drosophila* have led to the identification of crucial developmental regulators involved in body axis formation and segmentation, including maternal effect genes, gap genes, pair-rule genes, and segment polarity genes [32].

A landmark contribution of *Drosophila* research is the identification of morphogen gradients, particularly the Bicoid gradient, which establishes positional information along the anterior–posterior axis of the embryo [33]. Additionally, studies in this organism have been fundamental in uncovering several highly conserved developmental signaling pathways, such as Hedgehog, Notch, and Wnt signaling pathways [32]. These pathways are not only critical for invertebrate development but also play analogous roles in vertebrate systems, reflecting the evolutionary conservation of gene regulatory mechanisms. The semi-transparent nature of *Drosophila* embryos, combined with advanced imaging technologies, enables real-time observation of gene expression patterns and cellular dynamics during embryogenesis. This provides valuable insights into developmental processes at both molecular and cellular levels [34], [35]. Moreover, the feasibility of performing large-scale genetic screens in *Drosophila* has enabled the discovery of numerous novel genes and regulatory networks involved in diverse developmental processes, including organ formation and neural patterning [27].

3.2 *Caenorhabditis elegans*

The nematode *Caenorhabditis elegans* is a well-established model organism extensively employed in the study of developmental gene regulation. Its transparent body, fixed cell lineage, and relatively simple structural organization make it an ideal system for investigating cell fate determination and developmental events at single-cell resolution [7], [24], [36]. The entire cell lineage of *C. elegans* has been comprehensively mapped, enabling researchers to follow each cell division from the fertilized egg to the fully developed organism.

Research in *C. elegans* has uncovered key regulatory mechanisms governing processes such as cell differentiation, programmed cell death (apoptosis), and developmental timing. Notably, this organism played a central role in the discovery of RNA interference (RNAi), a gene regulatory mechanism mediated by small RNA molecules that suppress gene expression. RNAi has since emerged as a fundamental tool for gene silencing and functional genomics across a wide range of biological systems [27], [37].

3.3 *Danio rerio* (Zebrafish)

The zebrafish *Danio rerio* is a prominent vertebrate model organism extensively utilized for investigating embryonic development and gene regulation. Its embryos are transparent and undergo external development, enabling direct visualization of key developmental events such as gastrulation, organ formation, and tissue patterning [38], [39]. These characteristics make zebrafish particularly advantageous for examining gene expression dynamics and cellular migration during embryogenesis.

Molecular and genetic studies in zebrafish have substantially contributed to elucidating vertebrate developmental mechanisms, including those governing neural development, cardiovascular system formation, and skeletal patterning. This model organism is also widely employed to study major developmental signaling pathways such as Wnt, FGF, and BMP, all of which play critical roles during embryogenesis. Furthermore, the advent of advanced genome editing technologies has enabled precise functional analysis of gene regulatory networks in zebrafish embryos [40], [41].

In addition, the availability of extensive genomic databases and functional annotation resources has further strengthened the position of zebrafish as a powerful system for developmental biology research, supporting both fundamental studies and translational applications [42].

3.4 *Mus musculus*

The laboratory mouse *Mus musculus* is the most extensively utilized mammalian model organism for studying embryonic development and gene regulation. Due to its high genetic homology with humans, it serves as a crucial system for understanding mammalian developmental biology as well as the molecular basis of human diseases. Mouse models have played a pivotal role in elucidating the functions of transcription factors, signaling pathways, and epigenetic mechanisms that govern embryogenesis [43], [44].

The establishment of transgenic and gene knockout technologies in mice has enabled detailed investigation of the functional roles of individual genes during developmental processes. More recently, the application of CRISPR–Cas genome editing has significantly enhanced the precision and efficiency of genetic modifications in mouse systems. These advanced methodologies have facilitated comprehensive studies of gene regulatory networks involved in cellular differentiation, organogenesis, and stem cell maintenance [23], [45].

Table 1. Major model organisms used in developmental gene regulation studies [46]

Model Organism	Characteristics	Developmental Contributions	Relevance to Human Biology
Drosophila melanogaster	Short life cycle, powerful genetics	Segmentation genes, morphogen gradients	Discovery of conserved signaling pathways
Caenorhabditis elegans	Transparent body, defined cell lineage	Cell fate determination, RNA interference	Understanding apoptosis and gene silencing
Danio rerio	Transparent embryos, vertebrate system	Organogenesis, neural development	Vertebrate developmental studies
Mus musculus	Mammalian model, genetic similarity to humans	Gene knockout studies, disease modeling	Translational research in human diseases

4. Molecular Mechanisms of Gene Regulation in Embryogenesis

Embryonic development is governed by highly coordinated regulatory mechanisms that control gene expression in a precise spatial and temporal manner [1]. These mechanisms ensure that specific genes are selectively activated or suppressed at distinct stages of development, allowing cells to differentiate into specialized types with unique functions. Gene regulation during embryogenesis occurs through multiple interconnected layers, including transcriptional control, epigenetic modifications, chromatin remodeling, and post-transcriptional regulation [47]. Collectively, these regulatory processes form complex networks that integrate environmental factors, signaling pathways, and developmental cues to direct embryonic pattern formation and cellular differentiation.

4.1 Transcriptional Regulation

Transcriptional regulation is a fundamental mechanism governing gene expression during embryonic development. This process is primarily mediated by transcription factors that recognize and bind to specific DNA sequences within promoters, enhancers, or silencers associated with developmental genes [1]. These regulatory elements interact with the transcriptional machinery to either stimulate or inhibit gene expression [48].

During embryogenesis, transcription factors play a pivotal role in initiating developmental programs and guiding cell fate determination [49]. Many of these factors belong to evolutionarily conserved protein families, including homeobox (HOX), basic helix–loop–helix (bHLH), and zinc-finger proteins. They regulate genes responsible for critical processes such as body axis formation, tissue specification, and organogenesis [1], [50]. The combined activity of multiple transcription factors often forms regulatory networks or modules that ensure precise control of gene expression during development [1].

Enhancers are also key components of transcriptional regulation in embryogenesis [51]. These DNA regulatory elements can exert their effects over considerable genomic distances, enabling tissue-specific and stage-specific gene activation [51], [52]. The dynamic interaction between enhancers and promoters allows cells to interpret positional cues and activate appropriate genetic programs, thereby contributing to accurate embryonic patterning and development.

4.2 Epigenetic Regulation

Epigenetic mechanisms constitute an additional layer of gene regulation that modulates chromatin organization and gene accessibility without altering the primary DNA sequence [53]. These modifications are essential for regulating developmental gene expression and preserving cellular identity during embryogenesis [54].

Among the key epigenetic processes, DNA methylation plays a central role and is commonly associated with transcriptional repression by restricting the binding of transcription factors to DNA [55]. During early embryonic development, DNA methylation patterns undergo dynamic reprogramming, which is crucial for controlling gene expression during cell differentiation [56], [57]. In addition to DNA methylation, histone modifications are major contributors to epigenetic regulation [58]. Various chemical modifications of histone proteins, including acetylation, methylation, phosphorylation, and ubiquitination, influence chromatin structure and accessibility [59].

For instance, histone acetylation is generally linked with transcriptional activation, whereas histone methylation can either promote or inhibit gene expression depending on the specific residues modified and their context within the genome [60]. Collectively, these epigenetic mechanisms enable embryonic cells to maintain stable gene expression profiles while retaining the flexibility to respond to developmental cues.

4.3 Chromatin Remodelling and Genome Organization

Chromatin organization plays a critical role in the regulation of gene expression during embryonic development [61]. In eukaryotic cells, DNA is packaged into chromatin, which can adopt either a tightly condensed or a more relaxed configuration, thereby influencing gene accessibility [58]. Chromatin remodeling complexes regulate nucleosome positioning and alter chromatin structure, ultimately controlling the accessibility of transcription factors to their target genes.

Beyond localized chromatin modifications, higher-order genome organization also plays a significant role in developmental gene regulation. The three-dimensional arrangement of the genome within the nucleus enables distal regulatory elements, such as enhancers, to physically interact with their corresponding promoters through chromatin looping mechanisms. These interactions are often structured within defined regions known as topologically associating domains (TADs), which promote coordinated regulation of gene expression.

Dynamic alterations in chromatin architecture during embryogenesis allow cells to selectively activate or repress specific developmental genes in response to various signaling inputs, thereby ensuring precise control of developmental processes [62], [63].

4.4 Post-Transcriptional Regulation

Post-transcriptional mechanisms provide an additional level of control over gene expression by acting on RNA transcripts after they have been synthesized. These processes regulate various aspects of messenger RNA (mRNA) fate, including its maturation, stability, intracellular transport, and efficiency of translation into proteins. One of the key mechanisms involved is alternative splicing, through which a single gene can produce multiple protein variants with different functional properties. This significantly enhances protein diversity and plays a vital role in regulating developmental processes [64], [65].

In addition, small non-coding RNAs, particularly microRNAs (miRNAs), are crucial regulators of gene expression at the post-transcriptional level. MicroRNAs exert their effects by binding to complementary sequences on target mRNAs, leading to their degradation or suppression of translation. Numerous miRNAs have been identified that control essential developmental genes and signaling pathways during embryogenesis. Through these regulatory actions, post-transcriptional mechanisms ensure precise modulation of gene expression, maintaining appropriate timing and levels of gene activity required for normal development [66], [67].

4.5 Integration of Regulatory Mechanisms

Although these regulatory mechanisms are often discussed independently, they function in a highly coordinated and interconnected manner during embryonic development [54], [61]. Transcriptional control, epigenetic modifications, chromatin organization, and post-transcriptional regulation collectively contribute to the formation of complex gene regulatory networks that govern developmental gene expression [54], [61]. These networks integrate inputs from signaling pathways and morphogen gradients, allowing embryonic cells to interpret positional cues and commit to specific developmental lineages [54]. A comprehensive understanding of the interactions among these regulatory layers is crucial for elucidating the molecular basis of embryogenesis and overall organismal development [61]. The key molecular mechanisms involved in regulating gene expression during embryonic development namely transcriptional regulation, epigenetic modifications, chromatin architecture, and post-transcriptional control are summarized in Figure 1.

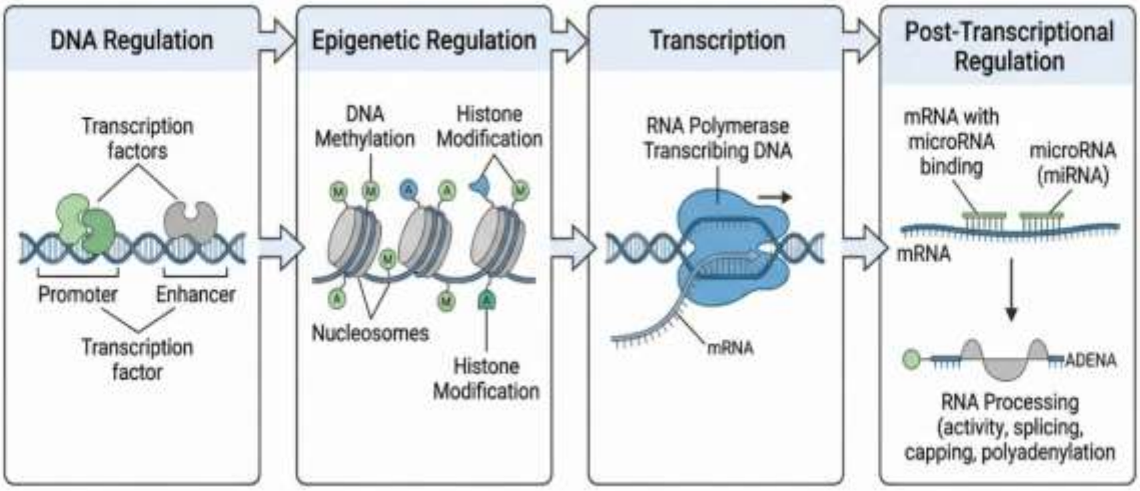

Figure 1. Molecular Mechanisms Regulating Gene Expression During Embryonic Development

5. Gene Regulatory Networks and Pattern Formation

5.1 Gene Regulatory in Development

Gene regulatory networks (GRNs) are complex systems composed of interacting genes, transcription factors, regulatory DNA sequences, and signaling pathways that collectively coordinate gene expression [68]. Within these networks, transcription factors function as central regulatory elements that either activate or suppress the expression of specific target genes, thereby directing developmental processes [17]. These interactions are often organized in hierarchical frameworks, where upstream regulatory genes govern the activity of downstream genes involved in specialized developmental roles [69].

During embryogenesis, GRNs play a vital role in controlling essential processes such as cell fate determination, body axis establishment, and tissue differentiation [17]. These networks are characterized by feedback and feedforward loops, which contribute to the stabilization of gene expression patterns while still permitting adaptive responses to developmental cues [70]. The dynamic nature of GRNs allows embryonic cells to integrate environmental and positional information, ultimately ensuring accurate tissue organization and organ development.

5.2 Maternal-to-Zygotic Transition

One of the earliest and most crucial regulatory events in embryonic development is the maternal-to-zygotic transition (MZT). During this phase, developmental control shifts from maternally inherited RNA and proteins to gene expression governed by the zygotic genome [71]. In the initial stages, maternal gene products deposited within the oocyte direct early developmental processes, including rapid cell divisions and initial pattern formation [72].

As embryogenesis advances, the zygotic genome becomes transcriptionally activated, resulting in the progressive degradation of maternal transcripts and the initiation of zygotic gene expression [73]. This transition is tightly controlled by specific transcription factors and chromatin remodelling processes that trigger widespread transcriptional activation required for further development [74]. The maternal-to-zygotic transition represents a fundamental step in establishing functional gene regulatory networks that drive subsequent stages of embryogenesis [24].

5.3 Morphogen Gradients and Positional Information

Morphogen gradients are fundamental in establishing spatial organization of gene expression during embryonic development [75]. Morphogens are diffusible signaling molecules that spread across embryonic tissues, forming concentration gradients that provide positional information to cells [76]. Cells within the embryo interpret these gradients to determine their location and accordingly activate specific developmental gene expression programs [14].

Well-known examples of morphogen gradients include the Bicoid gradient in *Drosophila* embryos and signaling molecules such as Sonic Hedgehog and BMP in vertebrate systems [77]. These gradients influence gene expression by triggering distinct sets of target genes at different concentration thresholds, leading to the formation of defined developmental regions within the embryo [77]. Through this mechanism, morphogen gradients enable coordinated pattern formation across extensive developmental fields, ensuring proper spatial arrangement of tissues and organs [14], [25].

5.4 Feedback and Regulatory Loops in Development

Gene regulatory networks often incorporate feedback and feedforward loops that play a crucial role in stabilizing gene expression patterns during development [70]. Positive feedback mechanisms enhance and sustain gene activation, ensuring that once a particular cell fate is initiated, the associated developmental regulators remain consistently expressed [78]. In contrast, negative feedback loops act to limit gene activity, preventing overexpression and maintaining appropriate developmental equilibrium [79]. These regulatory circuits contribute significantly to the robustness and reliability of developmental processes, enabling embryos to preserve stable gene expression patterns despite internal variability or external environmental fluctuations [80]. Through these integrated mechanisms, gene regulatory networks establish a coordinated regulatory framework that ensures precise and consistent embryonic development [26].

6. Signaling Pathways in Embryonic Gene Regulation

Embryonic development is regulated not only by intrinsic genetic mechanisms but also by extracellular signaling pathways that mediate intercellular communication and coordinate developmental patterning. These pathways influence gene expression by transmitting external molecular signals to the nucleus, thereby modulating transcriptional activity and guiding developmental outcomes. Several evolutionarily conserved signaling pathways are critically involved in embryogenesis, including Wnt, Notch, Hedgehog, BMP/TGF-β, and fibroblast growth factor (FGF) pathways. These signaling cascades interact closely with gene regulatory networks to regulate key processes such as cell proliferation, differentiation, tissue organization, and organ formation [27]. A summary of the major signaling pathways involved in embryonic gene regulation, along with their developmental roles, is provided in Table 2. Additionally, the principal signaling pathways that coordinate embryonic gene regulation and pattern formation namely Wnt, Notch, Hedgehog, BMP/TGF-β, and FGF signaling are illustrated in Figure 2.

6.1 Wnt Signaling Pathway

The Wnt signaling pathway is one of the most evolutionarily conserved regulatory systems involved in embryonic development [27]. It plays a crucial role in controlling key developmental processes such as axis formation, cell fate specification, tissue patterning, and stem cell maintenance across model organisms including *Drosophila*, zebrafish, and mice [81], [82]. The pathway primarily functions through the canonical mechanism, in which binding of Wnt ligands to Frizzled receptors leads to stabilization and nuclear accumulation of β-catenin. This β-catenin subsequently interacts with TCF/LEF transcription factors to regulate the expression of target genes [27].

During early embryogenesis, Wnt signaling is essential for establishing the anterior–posterior axis—for example, via Wingless signaling in *Drosophila* and also contributes to the regulation of stem cell populations and tissue differentiation [27]. In addition to the canonical pathway, non-canonical Wnt signaling pathways, such as the planar cell polarity pathway, are involved in regulating cell movement and tissue organization during development [28]. Dysregulation of Wnt signaling is strongly associated with developmental defects and various human diseases, highlighting its critical biological significance [8].

6.2 Notch Signaling Pathway

The Notch signaling pathway is a highly conserved mechanism of direct cell-to-cell communication that plays a central role in regulating cell fate decisions during development [83]. Unlike many signaling pathways, Notch signaling requires physical interaction between adjacent cells, allowing precise spatial control of developmental processes within tissues [84].

Activation of the Notch receptor results in the release of the Notch intracellular domain (NICD), which translocates to the nucleus and modulates the transcription of target genes involved in differentiation and tissue organization [85]. Notch signaling is essential for processes such as neural development, segmentation, and maintenance of stem cell populations [29]. It is particularly important for binary cell fate decisions and proper organ formation, with tightly regulated activity being crucial for both embryonic development and tissue homeostasis [30], [31].

6.3 Hedgehog Signaling Pathway

The Hedgehog signaling pathway is another highly conserved system that regulates embryonic patterning, morphogenesis, and organ development across a wide range of organisms [27], [31]. This pathway is activated when Hedgehog ligands (Hh in *Drosophila* or Sonic Hedgehog (Shh) in vertebrates) bind to the Patched receptor, relieving its inhibitory effect on Smoothened. This activation leads to the nuclear localization of Gli transcription factors, which regulate target gene expression [86].

Hedgehog signaling is essential for numerous developmental processes, including neural tube patterning, limb formation, body axis development, and organogenesis [28], [31]. For instance, Sonic Hedgehog plays a critical role in establishing ventral–dorsal patterning in vertebrate embryos, and its disruption can lead to severe defects such as cyclopia [28]. Proper spatial and temporal regulation of Hedgehog signaling is crucial for normal development, while its dysregulation is associated with developmental abnormalities and diseases, including cancer [27], [33]. Additionally, this pathway can be reactivated in adults during tissue repair and stem cell maintenance [32], [34].

6.4 BMP and TGF-β Signaling Pathways

Bone morphogenetic protein (BMP) and transforming growth factor-β (TGF-β) signaling pathways, which belong to the TGF-β superfamily, are key regulators of cell proliferation, differentiation, apoptosis, and tissue patterning during embryonic development [27], [31]. These pathways are initiated when ligands bind to type I and type II serine/threonine kinase receptors, leading to phosphorylation of receptor-regulated SMAD proteins (SMAD1/5/8 for BMP and SMAD2/3 for TGF-β). These SMAD complexes associate with SMAD4 and translocate to the nucleus to regulate gene expression [31].

During embryogenesis, BMP signaling is critical for dorsal–ventral axis formation and germ layer patterning, while TGF-β signaling is involved in mesoderm formation, left-right asymmetry, epithelial–mesenchymal transition, and organ development [28], [30], [31]. These pathways often interact with other signaling systems such as Wnt and Hedgehog to ensure coordinated developmental outcomes. Disruption of BMP/TGF-β signaling can result in significant developmental abnormalities, emphasizing their essential roles in embryogenesis [28].

6.5 Fibroblast Growth Factor (FGF) Signaling Pathway

The fibroblast growth factor (FGF) signaling pathway is a highly conserved regulatory system involved in embryonic patterning, cell proliferation, differentiation, survival, and morphogenesis across various organisms [28], [31]. FGF ligands bind to receptor tyrosine kinases (FGFRs), triggering receptor dimerization and autophosphorylation, which in turn activates downstream signaling cascades such as the MAPK/ERK pathway. These signaling events regulate the expression of genes essential for developmental processes.

Table 2. Signaling pathways involved in embryonic gene regulation

Signaling Pathway	Components	Developmental Functions	Organisms Studied
Wnt	β-catenin, Frizzled receptors	Axis formation, stem cell regulatio	Drosophila, Zebrafish, Mouse[87], [88], [89]
Notch	Notch receptor, Delta ligand	Cell fate determination, tissue patterning	Drosophila, Zebrafish, Mouse
Hedgehog	Hedgehog ligand, Smoothened, Gli	Neural patterning, limb development	Drosophila, Mouse[90], [91]
BMP/TGF-β	SMAD proteins, BMP ligands	Dorsal–ventral patterning, tissue differentiation	Zebrafish, Mouse
FGF	FGF ligands, receptor tyrosine kinases	Cell proliferation, organogenesis	Zebrafish, Mouse

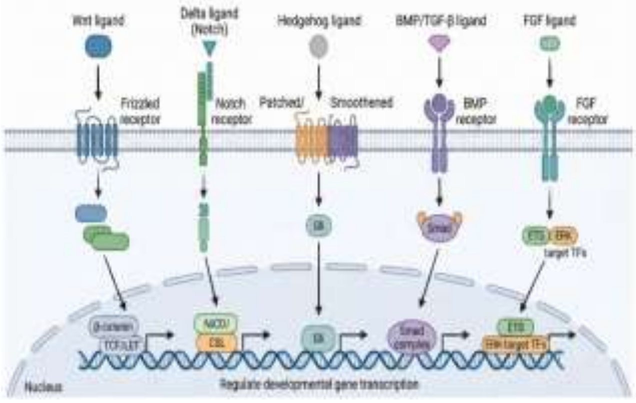

Figure 2. Signaling Pathways Regulating Gene Expression

7. Gene Regulation in Specific Developmental Processes

Gene regulation is fundamental to controlling essential developmental events during embryogenesis, including cell fate determination, tissue patterning, organ formation, and stem cell differentiation. In the early stages of development, embryonic cells undergo tightly regulated genetic programming that directs them toward specific cellular identities. This process is governed by coordinated interactions among transcription factors, signaling pathways, and epigenetic regulators, which activate lineage-specific genes while suppressing alternative developmental programs a phenomenon referred to as cell fate determination [92], [93].

As development progresses, gene regulatory networks in conjunction with morphogen gradients establish distinct spatial domains of gene expression. These domains guide tissue patterning and structural organization, ensuring that cells interpret positional cues accurately and differentiate according to their anatomical context. Gene regulation also plays a critical role in organogenesis, where precise temporal and spatial gene expression patterns drive organ formation through regulated processes such as cell proliferation, migration, and differentiation.

Similarly, embryonic stem cell maintenance and differentiation are controlled by intricate transcriptional and epigenetic networks that balance self-renewal with lineage commitment. Throughout these developmental stages, key signaling pathways including Wnt, Notch, BMP, and FGF interact with transcriptional regulators and chromatin-modifying factors to fine-tune gene expression and ensure proper developmental progression. Collectively, these integrated regulatory mechanisms enable the transformation of a single fertilized cell into a complex organism composed of diverse cell types and organized tissues [94], [95], [96].

8. Emerging Technologies for Gene Regulation in Embryonic Development

Recent advancements in molecular biology and genomics have significantly enhanced our ability to study gene regulation during embryonic development. Conventional genetic and biochemical methods are now complemented by advanced high-throughput and precision technologies that provide detailed insights into gene expression and regulatory mechanisms. Among the most impactful innovations are single-cell omics approaches, which allow gene expression analysis at the resolution of individual cells. Techniques such as single-cell RNA sequencing (scRNA-seq) and single-cell assay for transposase-accessible chromatin sequencing (scATAC-seq) have enabled detailed characterization of cellular heterogeneity and developmental trajectories during embryogenesis [35]. These methodologies facilitate the identification of lineage-specific transcriptional programs and enable reconstruction of dynamic gene regulatory networks that drive cell differentiation and lineage progression. In parallel, genome editing technologies particularly CRISPR–Cas systems have transformed functional genomics by allowing precise and targeted manipulation of DNA sequences [23], [97]. These tools are widely used to investigate the roles of genes, regulatory elements, and signaling pathways including Wnt, BMP/TGF-β, FGF, Notch, and Hedgehog in developmental processes such as patterning, differentiation, and morphogenesis.

Spatial transcriptomics has further expanded the analytical scope by enabling the study of gene expression within intact tissue architectures while preserving spatial context. This approach allows mapping of signaling pathway activity across entire embryos, revealing tissue-specific gene expression patterns and developmental organization.

In addition to experimental innovations, computational modeling and systems biology approaches are increasingly employed to integrate large-scale genomic datasets and reconstruct gene regulatory networks governing development [98]. Together, these technologies have significantly advanced our understanding of the regulatory complexity underlying embryogenesis and continue to open new avenues for investigating developmental biology at unprecedented resolution.

The integration of single-cell and spatial omics technologies, combined with advanced computational frameworks, is now enabling detailed analysis of spatiotemporal gene regulation during cell lineage specification and tissue morphogenesis. Furthermore, combining lineage tracing techniques with single-cell sequencing approaches provides deeper insights into developmental relationships among different cell types. For example, single-cell CUT&TAG has been utilized to profile histone modifications in post-implantation embryos, linking enhancer activity with gene expression patterns within spatial contexts. Beyond spatial and cellular resolution, emerging technologies also facilitate the investigation of temporal dynamics in gene regulation, offering a more comprehensive understanding of developmental processes [99].

9. Evolutionary Conservation of Developmental Gene Regulation

9.1 Developmental Genes and Regulatory Pathways

A large number of genes involved in embryonic development belong to evolutionarily conserved gene families that regulate body patterning and tissue specification across a wide range of species. One of the most notable examples is the homeobox gene family, which plays a central role in establishing the anterior–posterior axis in organisms ranging from *Drosophila* to vertebrates [100]. The structural organization and functional conservation of HOX genes highlight the preservation of essential genetic mechanisms that govern body plan formation through evolutionary processes.

In a similar manner, major developmental signaling pathways including Wnt/Wingless, BMP/TGF-β, FGF, Notch, and Hedgehog exhibit strong conservation in terms of their molecular components, receptors, downstream effectors, and biological functions across metazoans. These pathways regulate key processes such as cell differentiation, tissue patterning, proliferation, survival, and morphogenesis. For example, they are involved in the formation of germ layers, body axes, and organ systems in both invertebrate and vertebrate embryos, with ligands expressed in specific spatial and temporal patterns during development [32].

The high degree of conservation observed in gene regulatory networks and developmental programs enables researchers to apply findings from model organisms such as *Drosophila*, zebrafish, and mouse to more complex systems, including humans. Approaches such as comparative genomics and evolutionary developmental biology (Evo–Devo) have been instrumental in identifying these conserved developmental toolkits, thereby enhancing our understanding of the fundamental principles underlying embryogenesis across different species [101].

9.2 Evolutionary Developmental Biology

Evolutionary developmental biology, commonly referred to as Evo–Devo, focuses on understanding how changes in gene regulation drive the evolution of morphological diversity. Rather than arising from extensive alterations in gene sequences, many evolutionary variations are attributed to modifications in regulatory elements such as enhancers and promoters, which determine the spatial and temporal patterns of gene expression.

Such regulatory changes can alter developmental processes, leading to variations in body structure, organ formation, and overall morphology among species. By analyzing and comparing gene regulatory networks across different organisms, Evo–Devo provides valuable insights into how conserved developmental programs have been modified over evolutionary time to generate biological diversity [102].

9.3 Relevance to Human Development

The conservation of developmental gene regulation has significant implications for understanding human biology and disease. Numerous genes and signaling pathways initially identified in model organisms have been shown to perform similar functions during human embryonic development. As a result, model systems serve as powerful platforms for investigating the molecular basis of congenital disorders and developmental abnormalities.

Furthermore, insights derived from comparative developmental studies have contributed to advancements in regenerative medicine, stem cell research, and therapeutic strategies. By examining conserved regulatory mechanisms across species, researchers can better understand the genetic basis of human development and identify potential targets for the treatment of developmental disorders [35], [103].

10. Implications for Human Development and Disease

Research on gene regulation during embryonic development has profound implications for understanding human health and disease. Many of the molecular pathways and regulatory mechanisms that govern embryogenesis in model organisms are highly conserved in humans. Consequently, studies conducted in systems such as *Drosophila melanogaster*, *Caenorhabditis elegans*, zebrafish (*Danio rerio*), and mouse (*Mus musculus*) have been instrumental in identifying genes and signaling pathways that regulate human developmental processes.

These discoveries have provided critical insights into how disruptions in gene regulatory networks can result in developmental abnormalities and congenital disorders, thereby advancing our understanding of disease mechanisms and potential therapeutic interventions [104], [105].

10.1 Developmental Disorders and Congenital Abnormalities

Disruptions in gene regulation during embryonic development can lead to a broad spectrum of congenital abnormalities. Mutations affecting transcription factors, signaling molecules, or regulatory DNA elements may interfere with normal developmental programs and impair proper tissue formation. For instance, altered expression of HOX genes can disrupt body patterning, while defects in signaling pathways such as Wnt, Hedgehog, and BMP are associated with skeletal deformities, neural defects, and organ malformations [100], [106].

Insights into the molecular basis of these conditions have been greatly enhanced through studies in model organisms, where developmental genes can be experimentally manipulated. Such comparative approaches allow identification of conserved gene regulatory network components as well as lineage-specific modifications that have evolved over time [107].

10.2 Gene Regulatory Networks in Human Disease

Gene regulatory networks that orchestrate embryonic development also play essential roles in maintaining cellular function throughout life. Dysregulation of these networks can contribute not only to developmental abnormalities but also to complex diseases such as cancer. Many cancers are associated with aberrant activation of developmental signaling pathways, including Wnt, Notch, and Hedgehog pathways [8], [108].

These pathways, which normally regulate controlled cell proliferation and differentiation during development, may lead to uncontrolled cell growth and tumor formation when misregulated. Therefore, studying gene regulation during embryogenesis provides critical insights into disease mechanisms and offers potential targets for therapeutic intervention [1], [95].

10.3 Applications in Regenerative Medicine and Stem Cell Research

Understanding gene regulation in embryonic development has significantly advanced the fields of regenerative medicine and stem cell biology. Knowledge of transcriptional and epigenetic mechanisms that govern cell fate decisions allows researchers to manipulate stem cells to produce specific cell types for therapeutic applications. Technologies such as induced pluripotent stem cells (iPSCs) rely on the controlled expression of key developmental transcription factors to reprogram differentiated cells into a pluripotent state. These innovations have created new opportunities for developing cell-based therapies aimed at tissue repair and the treatment of degenerative diseases [109], [110].

11. Future Perspectives and Emerging Directions

Rapid progress in molecular biology, genomics, and computational sciences is reshaping our understanding of gene regulation during embryonic development. Although considerable advancements have been made in identifying major regulatory genes and signaling pathways, many aspects of developmental gene regulation remain to be fully elucidated. Future research is expected to focus on integrating multiple layers of regulation—including transcriptional, epigenetic, and chromatin-based mechanisms—to better understand how complex gene regulatory networks coordinate developmental processes.

One of the most promising directions involves the application of single-cell and spatial omics technologies. These approaches enable detailed analysis of gene expression, chromatin accessibility, and epigenetic modifications at single-cell resolution while preserving spatial context within tissues. Such technologies provide powerful tools for studying cell lineage specification, developmental trajectories, and dynamic regulatory changes during embryogenesis.

Another emerging area is the use of advanced genome editing technologies, particularly CRISPR-based systems, to investigate gene function and regulatory elements. These tools allow precise modification of genes, enhancers, and other regulatory sequences, enabling detailed dissection of gene regulatory networks. Additionally, CRISPR-based screening approaches are increasingly being used to identify novel regulators involved in developmental processes [23], [111].

Computational modeling and systems biology approaches are also expected to play a significant role in future research. By integrating large-scale genomic and transcriptomic data, these methods can reconstruct gene regulatory networks and predict how interactions among genes and signaling pathways influence developmental outcomes [112].

Furthermore, emerging experimental systems such as organoids and synthetic developmental models provide innovative platforms for studying human development in vitro. These systems replicate key aspects of embryogenesis, enabling controlled investigation of gene regulation in human tissues and supporting advances in regenerative medicine and stem cell research. Collectively, these developments are paving the way toward a more comprehensive understanding of gene regulatory networks, including their spatial and temporal organization within a three-dimensional framework of embryonic development [113], [114].

12. Conclusion

Embryonic development is governed by highly coordinated gene regulatory mechanisms that control spatial and temporal patterns of gene expression. These mechanisms involve complex interactions among transcription factors, regulatory DNA elements, epigenetic modifications, and signaling pathways that collectively guide cell fate determination, tissue patterning, and organ formation. Studies in model organisms such as Drosophila melanogaster, Caenorhabditis elegans, zebrafish (Danio rerio), and mouse (Mus musculus) have been instrumental in identifying conserved developmental genes and signaling pathways, including Wnt, Notch, Hedgehog, and BMP/TGF-β, which regulate key embryonic processes. Recent advances in technologies such as single-cell genomics, CRISPR-based genome editing, and computational modeling have significantly enhanced our ability to study dynamic gene regulatory networks during development. These approaches are providing deeper insights into the molecular mechanisms underlying embryogenesis and their relevance to human developmental biology. Continued research integrating experimental and computational methods will further improve our understanding of developmental gene regulation and may contribute to new therapeutic strategies for congenital disorders and regenerative medicine.

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Conflict of Interest

The authors declare that there is no conflict of interest

REFERENCE

[1] A. Lin, "Mechanisms of Gene Expression Regulation in Embryonic Development," *BIO Web of Conferences*, vol. 174, p. 1009, Jan. 2025, doi: 10.1051/bioconf/202517401009.

[2] U. Kadiyala, "Spatiotemporal Coordination of Proliferation and Patterning in Early Embryonic Development," *Deep Blue (University of Michigan)*, Jan. 2025, doi: 10.7302/27155.

[3] M. Lagha, J. P. Bothma, and M. Levine, "Mechanisms of transcriptional precision in animal development," *Trends in Genetics*, vol. 28, no. 8. Elsevier BV, p. 409, Apr. 16, 2012. doi: 10.1016/j.tig.2012.03.006.

[4] G. Peng *et al.* , "Molecular architecture of lineage allocation and tissue organization in early mouse embryo," *Nature* , vol. 572, no. 7770, p. 528, Aug. 2019, doi: 10.1038/s41586-019-1469-8.

[5] J. Briggs *et al.* , "The dynamics of gene expression in vertebrate embryogenesis at single-cell resolution," *Science* , vol. 360, no. 6392, Apr. 2018, doi: 10.1126/science.aar5780.

[6] B. Prud'homme, N. Gompel, and S. B. Carroll, "Emerging principles of regulatory evolution," *Proceedings of the National Academy of Sciences* , vol. 104, p. 8605, May 2007, doi: 10.1073/pnas.0700488104.

[7] A. G. Cole, T. Hashimshony, Z. Du, and I. Yanai, "Gene regulatory patterning codes in early cell fate specification of the C. elegans embryo," *eLife* , vol. 12, May 2023, doi: 10.7554/elife.87099.

[8] X. Chen, Q. Chu, Q. Shi, Y. Zeng, J. Lu, and -Jun Li-, "Wnt signaling pathways in biology and disease: mechanisms and therapeutic advances," *Signal Transduction and Targeted Therapy*, vol. 10, no. 1, p. 106, Apr. 2025, doi: 10.1038/s41392-025-02142-w.

[9] M. C. K. Leung *et al.* , "Applying evolutionary genetics to developmental toxicology and risk assessment," *Reproductive Toxicology* , vol. 69. Elsevier BV, p. 174, Mar. 05, 2017. doi: 10.1016/j.reprotox.2017.03.003.

[10] N. Perrimon, C. Pitsouli, and B. Shilo, "Signaling Mechanisms Controlling Cell Fate and Embryonic Patterning," *Cold Spring Harbor Perspectives in Biology*, vol. 4, no. 8. Cold Spring Harbor Laboratory Press, Aug. 01, 2012. doi: 10.1101/cshperspect.a005975.

[11] A. S. Doane and O. Elemento, "Regulatory elements in molecular networks," *WIREs Systems Biology and Medicine*, vol. 9, no. 3, Jan. 2017, doi: 10.1002/wsbm.1374.

[12] M. Englund, "Identification and Characterization of Cis-Regulatory Elements in the Human Genome," *Deep Blue (University of Michigan)* , Jan. 2023, doi: 10.7302/7349.

[13] A. A. Pai, J. K. Pritchard, and Y. Gilad, "The Genetic and Mechanistic Basis for Variation in Gene Regulation,"*PLoS Genetics* , vol. 11, no. 1, Jan. 2015, doi: 10.1371/journal.pgen.1004857.

[14] M. F. Simsek and E. M. Özbudak, "Patterning principles of morphogen gradients," *Open Biology* , vol. 12, no. 10, p. 220224, Oct. 2022, doi: 10.1098/rsob.220224.

[15] K. S. Stapornwongkul and J. Vincent, "Generation of extracellular morphogen gradients: the case for diffusion," *Nature Reviews Genetics* , vol. 22, no. 6, p. 393, Mar. 2021,

doi: 10.1038/s41576-021-00342-y.

[16] J. Nam, P. Dong, R. Tarpine, S. Istrail, and E. H. Davidson, “Functional cis -regulatory genomics for systems biology,” *Proceedings of the National Academy of Sciences* , vol. 107, no. 8, p. 3930, Feb. 2010, doi: 10.1073/pnas.1000147107.

[17] J. E. Valencia and I. S. Peter, “Combinatorial regulatory states define cell fate diversity during embryogenesis,” *Nature Communications* , vol. 15, no. 1, p. 6841, Aug. 2024, doi: 10.1038/s41467-024-50822-y.

[18] J. Soczyńska *et al.* , “Medical embryology and regenerative medicine: research and applications in clinical practice,” *Frontiers in Cell and Developmental Biology* , vol. 13. Frontiers Media, Aug. 26, 2025. doi: 10.3389/fcell.2025.1619036.

[19] P. T. Sharpe and I. Mason, *Molecular Embryology* . Humana Press, 1999. doi: 10.1385/1592592708.

[20] J. W. Ding *et al.* , “Dissecting gene regulatory networks governing human cortical cell fate,” *Nature* , Jan. 2026, doi: 10.1038/s41586-025-09997-7.

[21] J. Zeiltinger *et al.* , “Perspective on recent developments and challenges in regulatory and systems genomics,”*arXiv (Cornell University)* , Nov. 2024, doi: 10.48550/arxiv.2411.04363.

[22] D. Prasad, “Hormonal Regulation of Drosophila Accessory Gland Growth,” *Deep Blue (University of Michigan)*, Apr. 2022, doi: 10.7302/6448.

[23] G. K. Varshney and S. M. Burgess, “CRISPR-based functional genomics tools in vertebrate models,” *Experimental & Molecular Medicine* , vol. 57, no. 7, p. 1355, Jul. 2025, doi: 10.1038/s12276-025-01514-0.

[24] U. Irion and C. Nüsslein-Volhard, “Developmental genetics with model organisms,” *Proceedings of the National Academy of Sciences* , vol. 119, no. 30, Jul. 2022, doi: 10.1073/pnas.2122148119.

[25] J. D. Amack, “Cellular dynamics of EMT: lessons from live in vivo imaging of embryonic development,” *Cell Communication and Signaling* , vol. 19, no. 1. BioMed Central, Jul. 22, 2021. doi: 10.1186/s12964-021-00761-8.

[26] M. Haniffa *et al.* , “A roadmap for the Human Developmental Cell Atlas,” *Nature* , vol. 597, no. 7875, p. 196, Sep. 2021, doi: 10.1038/s41586-021-03620-1.

[27] J. Kimble and C. Nüsslein-Volhard, “The great small organisms of developmental genetics: Caenorhabditis elegans and Drosophila melanogaster,” *Developmental Biology* , vol. 485, p. 93, Mar. 2022, doi: 10.1016/j.ydbio.2022.02.013.

[28] A. Gil-Gálvez *et al.* , “Gain of gene regulatory network interconnectivity at the origin of vertebrates,” *Proceedings of the National Academy of Sciences* , vol. 119, no. 11, Mar. 2022, doi: 10.1073/pnas.2114802119.

[29] A. Pérez-Posada *et al.* , “Hemichordate cis-regulatory genomics and the gene expression dynamics of deuterostomes,” *Nature Ecology & Evolution* , Oct. 2024, doi: 10.1038/s41559-024-02562-x.

[30] F. Bertile, S. Matallana-Surget, A. Tholey, S. Cristóbal, and J. Armengaud, “Diversifying the concept of model organisms in the age of -omics,” *Communications Biology* , vol. 6, no. 1. Nature Portfolio, Oct. 19, 2023. doi: 10.1038/s42003-023-05458-x.

[31] N. S. Tolwinski, “Drosophila—A Model System for Developmental Biology,” *Journal of Developmental Biology* , vol. 12, no. 2, p. 15, May 2024, doi: 10.3390/jdb12020015.

[32] M. Peifer, “40 years since the Heidelberg genetic screen that revolutionized developmental and cell biology.,” *Carolina Digital Repository (University of North Carolina at Chapel Hill)* , Oct. 2024, doi: 10.17615/wzs2-ay72.

[33] M. Weber, *Philosophy of Developmental Biology* . 2022. doi: 10.1017/9781108954181.

[34] E. Clark, M. Battistara, and M. Benton, “A timer gene network is spatially regulated by the terminal system in the Drosophila embryo,” *eLife* , vol. 11, Dec. 2022, doi: 10.7554/elife.78902.

[35] R. M. Williams, “Leveraging chicken embryos for studying human enhancers,” *Developmental Biology* , vol. 524, p. 123, May 2025, doi: 10.1016/j.ydbio.2025.05.009.

[36] P. Pan *et al.* , “A spiral microfluidic device for rapid sorting, trapping, and long-term live imaging of Caenorhabditis elegans embryos,” *Microsystems & Nanoengineering* , vol. 9, no. 1, Feb. 2023, doi: 10.1038/s41378-023-00485-4.

[37] A. Hernández-Soto, F. Echeverría-Beirute, and T. Hernández, “The RNAi as a tool to control tropical pathogens,” *Agronomía Mesoamericana* , p. 326, Jan. 2021, doi: 10.15517/am.v32i1.40896.

[38] R. S. Angom and N. M. R. Nakka, “Zebrafish as a Model for Cardiovascular and Metabolic Disease: The Future of Precision Medicine,” *Biomedicines* , vol. 12, no. 3, p. 693, Mar. 2024, doi: 10.3390/biomedicines12030693.

[39] D. Lanka and J. Nellore, “A Comprehensive Review on Zebrafish and Zebrafish Embryonic Stem Cells (Zescs) as the Versatile Biotechnological Green Tool for Detecting Environmental Pollutants,” *Biosciences Biotechnology Research Asia* , vol. 20, no. 3. Oriental Scientific Publishing Company, p. 773, Oct. 05, 2023. doi: 10.13005/bbra/3131.

[40] S. C. Fox and A. J. Waskiewicz, “Transforming growth factor beta signaling and craniofacial development: modeling human diseases in zebrafish,” *Frontiers in Cell and Developmental Biolog* , vol. 12, Feb. 2024, doi: 10.3389/fcell.2024.1338070.

[41] C. P. Choe *et al.*, “Transgenic fluorescent zebrafish lines that have revolutionized biomedical research,” *Laboratory Animal Research* , vol. 37, no. 1. BioMed Central, Sep. 08, 2021. doi: 10.1186/s42826-021-00103-2.

[42] D. Baranašić *et al.* , “Multiomic atlas with functional stratification and developmental dynamics of zebrafish cis-regulatory elements,” *Nature Genetics* , vol. 54, no. 7, p. 1037, Jul. 2022, doi: 10.1038/s41588-022-01089-w.

[43] S. D. M. Brown, “Advances in mouse genetics for the study of human disease,” *Human Molecular Genetics* , vol. 30, Jun. 2021, doi: 10.1093/hmg/ddab153.

[44] X. Huang *et al.* , “Single-cell, whole-embryo phenotyping

of mammalian developmental disorders," *Nature* , vol. 623, no. 7988, p. 772, Nov. 2023, doi: 10.1038/s41586-023-06548-w.

[45] M. Osterwalder *et al.* , "Characterization of Mammalian In Vivo Enhancers Using Mouse Transgenesis and CRISPR Genome Editing," *Methods in molecular biology* , vol. 2403, p. 147, Dec. 2021, doi: 10.1007/978-1-0716-1847-9_11.

[46] J. Lalanne *et al.* , "Multiplex profiling of developmental cis-regulatory elements with quantitative single-cell expression reporters," *Nature Methods* , vol. 21, no. 6, p. 983, May 2024, doi: 10.1038/s41592-024-02260-3.

[47] C. D. Todd *et al.* , "Epigenetic priming of mammalian embryonic enhancer elements coordinates developmental gene networks," *Genome biology* , vol. 26, no. 1, p. 214, Jul. 2025, doi: 10.1186/s13059-025-03658-8.

[48] M. Bauer, "How does an organism extract relevant information from transcription factor concentrations?," *Biochemical Society Transactions* , vol. 50, no. 5. Portland Press, p. 1365, Sep. 16, 2022. doi: 10.1042/bst20220333.

[49] T. Fan and Y. Huang, "Accessible chromatin reveals regulatory mechanisms underlying cell fate decisions during early embryogenesis," *Scientific Reports* , vol. 11, no. 1, Apr. 2021, doi: 10.1038/s41598-021-86919-3.

[50] C. Mau *et al.* , "How enhancers regulate wavelike gene expression patterns," *eLife* , vol. 12, Jul. 2023, doi: 10.7554/elife.84969.

[51] J. Yang and A. S. Hansen, "Enhancer selectivity in space and time: from enhancer–promoter interactions to promoter activation," *Nature Reviews Molecular Cell Biology* , vol. 25, no. 7, p. 574, Feb. 2024, doi: 10.1038/s41580-024-00710-6.

[52] B. P. de Almeida, C. Schaub, M. Pagani, S. Secchia, E. E. M. Furlong, and A. Stark, "Targeted design of synthetic enhancers for selected tissues in the Drosophila embryo," *Nature* , vol. 626, no. 7997, p. 207, Dec. 2023, doi: 10.1038/s41586-023-06905-9.

[53] N. M. A. Aboud, C. Tupper, and I. Jialal, "Genetics, Epigenetic Mechanism," Aug. 2021.

[54] A. L. Wilkinson, I. Zorzan, and P. J. Rugg-Gunn, "Epigenetic regulation of early human embryo development," *Cell stem cell* , vol. 30, no. 12, p. 1569, Oct. 2023, doi: 10.1016/j.stem.2023.09.010.

[55] S. Kaluscha *et al.* , "Evidence that direct inhibition of transcription factor binding is the prevailing mode of gene and repeat repression by DNA methylation," *Nature Genetics* , vol. 54, no. 12, p. 1895, Dec. 2022, doi: 10.1038/s41588-022-01241-6.

[56] Z. Zhang *et al.* , "Whole-Genome DNA Methylation Dynamics of Sheep Preimplantation Embryo Investigated by Single-Cell DNA Methylome Sequencing," *Frontiers in Genetics* , vol. 12, Dec. 2021, doi: 10.3389/fgene.2021.753144.

[57] Q. Zhou, Y. Xiong, B. Qu, A. Bao, and Y. Zhang, "DNA Methylation and Recurrent Pregnancy Loss: A Mysterious Compass?," *Frontiers in Immunology* , vol. 12. Frontiers Media, Oct. 21, 2021. doi: 10.3389/fimmu.2021.738962.

[58] R. Halabian, V. Arshad, A. Ahmadi, P. Saeedi, S. A. Jamalkandi, and M. R. Alivand, "Laboratory methods to decipher epigenetic signatures: a comparative review," *Cellular & Molecular Biology Letters* , vol. 26, no. 1. BioMed Central, Nov. 11, 2021. doi: 10.1186/s11658-021-00290-9.

[59] D. Jiang, T. Li, C. Guo, T.-S. Tang, and H. Liu, "Small molecule modulators of chromatin remodeling: from neurodevelopment to neurodegeneration," *Cell & Bioscience* , vol. 13, no. 1. BioMed Central, Jan. 16, 2023. doi: 10.1186/s13578-023-00953-4.

[60] M. Krasteva, "Environmental Stress, Epigenetic Modifications, Adaptation, and Disease: A Fine Interplay," in *IntechOpen eBooks*, IntechOpen, 2024. doi: 10.5772/intechopen.1004285.

[61] Y. Liu and R. Schneider, "Histone modifications in development," *Development* , vol. 152, no. 12, Jun. 2025, doi: 10.1242/dev.204384.

[62] G. Yu *et al.* , "Establishment of chromatin architecture interplays with embryo hypertranscription," *Nature* , vol. 646, no. 8083, p. 208, Aug. 2025, doi: 10.1038/s41586-025-09400-5.

[63] F. Cardamone *et al.* , "Chromatin landscape at cis-regulatory elements orchestrates cell fate decisions in early embryogenesis," *Nature Communications* , vol. 16, no. 1, p. 3007, Mar. 2025, doi: 10.1038/s41467-025-57719-4.

[64] M. Nazim, "Post-transcriptional regulation of the transcriptional apparatus in neuronal development," *Frontiers in Molecular Neuroscience* , vol. 17, p. 1483901, Dec. 2024, doi: 10.3389/fnmol.2024.1483901.

[65] Y. Tao, Q. Zhang, H. Wang, X. Yang, and H. Mu, "Alternative splicing and related RNA binding proteins in human health and disease," *Signal Transduction and Targeted Therapy* , vol. 9, no. 1. Springer Nature, Feb. 02, 2024. doi: 10.1038/s41392-024-01734-2.

[66] L. Lorenzo-Orts and A. Pauli, "The molecular mechanisms underpinning maternal mRNA dormancy," *Biochemical Society Transactions* , vol. 52, no. 2. Portland Press, p. 861, Mar. 13, 2024. doi: 10.1042/bst20231122.

[67] M. Arnott, N. F. Sampilo, and J. L. Song, "Transcription of microRNAs is regulated by developmental signaling pathways and transcription factors," *Frontiers in Cell and Developmental Biology* , vol. 12, p. 1356589, Apr. 2024, doi: 10.3389/fcell.2024.1356589.

[68] H. T. Otal, A. Subaşı, F. Kurt, M. A. Canbaz, and Y. Uzun, "Analysis of Gene Regulatory Networks from Gene Expression Using Graph Neural Networks," *arXiv (Cornell University)* , Sep. 2024, doi: 10.48550/arxiv.2409.13664.

[69] A. Grishechkin, A. Mukherjee, and O. Karin, "Hierarchical Control of State Transitions in Dense Associative Memories," *arXiv (Cornell University)* , Dec. 2024, doi: 10.48550/arxiv.2412.11336.

[70] A. J. Massri, B. D. McDonald, G. A. Wray, and D. R. McClay, "Feedback circuits are numerous in embryonic gene regulatory networks and offer a stabilizing influence on evolution of those networks," *EvoDevo* , vol. 14, no. 1, Jun.

2023, doi: 10.1186/s13227-023-00214-y.

[71] M. Kojima, C. Hoppe, and A. J. Giráldez, "The maternal-to-zygotic transition: reprogramming of the cytoplasm and nucleus," *Nature Reviews Genetics* , vol. 26, no. 4, p. 245, Nov. 2024, doi: 10.1038/s41576-024-00792-0.

[72] M. A. Hagadorn, F. K. Hunter, T. DeLory, M. M. Johnson, T. L. Pitts-Singer, and K. M. Kapheim, "Maternal body condition and season influence RNA deposition in the oocytes of alfalfa leafcutting bees (Megachile rotundata)," *Frontiers in Genetics* , vol. 13, Jan. 2023, doi: 10.3389/fgene.2022.1064332.

[73] T. Guan *et al.* , "Single-cell analysis of preimplantation embryonic development in guinea pigs," *Research Square (Research Square)* , Aug. 2024, doi: 10.21203/rs.3.rs-4820011/v1.

[74] M. Gao *et al.* , "Pluripotency factors determine gene expression repertoire at zygotic genome activation," *Nature Communications* , vol. 13, no. 1, p. 788, Feb. 2022, doi: 10.1038/s41467-022-28434-1.

[75] A. Kicheva and J. Briscoe, "Control of Tissue Development by Morphogens," *Annual Review of Cell and Developmental Biology* , vol. 39, no. 1, p. 91, Jul. 2023, doi: 10.1146/annurev-cellbio-020823-011522.

[76] T. Kühn *et al.* , "Single-molecule tracking of Nodal and Lefty in live zebrafish embryos supports hindered diffusion model," *Nature Communications* , vol. 13, no. 1, Oct. 2022, doi: 10.1038/s41467-022-33704-z.

[77] H. Greenfeld, J. Lin, and M. C. Mullins, "The BMP signaling gradient is interpreted through concentration thresholds in dorsal–ventral axial patterning," *PLoS Biology* , vol. 19, no. 1, Jan. 2021, doi: 10.1371/journal.pbio.3001059.

[78] A. Srivastava and M. Rashid, "Logic-dependent emergence of multistability, hysteresis, and biphasic dynamics in a minimal positive feedback network with an autoloop," *arXiv (Cornell University)* , Apr. 2024, doi: 10.48550/arxiv.2404.05379.

[79] M. J. K *et al.* , "A negative feedback loop of TOR signaling balances growth and stress-response trade-offs in plants," *Cell Reports* , vol. 39, no. 1, p. 110631, Apr. 2022, doi: 10.1016/j.celrep.2022.110631.

[80] M. Parihar *et al.* , "Retinoic Acid Fluctuation Activates an Uneven, Direction-Dependent Network-Wide Robustness Response in Early Embryogenesis," *Frontiers in Cell and Developmental Biology* , vol. 9, Oct. 2021, doi: 10.3389/fcell.2021.747969.

[81] E. Rismani, N. Haghighi-Najafabadi, B. E. Far, B. Shahbazi, and L. Mafakher, "Insights into the Wnt Signaling Pathway Evolution," in *Physiology* , IntechOpen, 2022. doi: 10.5772/intechopen.108012.

[82] L. Hu, W. Chen, A. Qian, and Y. Li, "Wnt/β-catenin signaling components and mechanisms in bone formation, homeostasis, and disease," *Bone Research* , vol. 12, no. 1. Springer Nature, Jul. 10, 2024. doi: 10.1038/s41413-024-00342-8.

[83] S. J. Bray and A. Bigas, "Modes of Notch signalling in development and disease," *Nature Reviews Molecular Cell Biology* , vol. 26, no. 7. Nature Portfolio, p. 522, Mar. 10, 2025. doi: 10.1038/s41580-025-00835-2.

[84] A. S. Yatsenko and H. R. Shcherbata, "Distant activation of Notch signaling induces stem cell niche assembly,"*PLoS Genetics* , vol. 17, no. 3, Mar. 2021, doi: 10.1371/journal.pgen.1009489.

[85] D. Verma, A. Singh, J. Singh, M. Mutsuddi, and A. Mukherjee, "Regulation of Notch signaling by non-muscle myosin II Zipper in Drosophila," *Cellular and Molecular Life Science*, vol. 81, no. 1, Apr. 2024, doi: 10.1007/s00018-024-05142-1.

[86] J. Jing *et al.*, "Hedgehog signaling in tissue homeostasis, cancers, and targeted therapies," *Signal Transduction and Targeted Therapy* , vol. 8, no. 1. Springer Nature, Aug. 18, 2023. doi: 10.1038/s41392-023-01559-5.

[87] G. A. Albano and A. S. Hackam, "Repurposing development genes for axonal regeneration following injury: Examining the roles of Wnt signaling," *Frontiers in Cell and Developmental Biology*, vol. 12, May 2024, doi: 10.3389/fcell.2024.1417928.

[88] C. Colin-Pierre *et al.*, "Regulation of stem cell fate by HSPGs: implication in hair follicle cycling," *npj Regenerative Medicine* , vol. 7, no. 1. Nature Portfolio, Dec. 28, 2022. doi: 10.1038/s41536-022-00267-y.

[89] C. Engel-Pizcueta and C. Pujades, "Interplay Between Notch and YAP/TAZ Pathways in the Regulation of Cell Fate During Embryo Development," *Frontiers in Cell and Developmental Biology*, vol. 9. Frontiers Media, Aug. 19, 2021. doi: 10.3389/fcell.2021.711531.

[90] H. Dear, "Investigating the Role of the GLI Transcription Factors in Limb Development," *Deep Blue (University of Michigan)* , Jan. 2024, doi: 10.7302/25045.

[91] J. Marcetteau, "The role of the small GTP-binding protein Arf6 during Drosophila development," *HAL (Le Centre pour la Communication Scientifique Directe)* , Jul. 2021, Accessed: Oct. 2025. [Online]. Available: https://theses.hal.science/tel-04136997

[92] P. Madrigal *et al.* , "Epigenetic and transcriptional regulations prime cell fate before division during human pluripotent stem cell differentiation," *Nature Communications* , vol. 14, no. 1, p. 405, Jan. 2023, doi: 10.1038/s41467-023-36116-9.

[93] J. Lee, N. Kim, and K. Cho, "Decoding the principle of cell-fate determination for its reverse control," *npj Systems Biology and Applications*, vol. 10, no. 1, p. 47, May 2024, doi: 10.1038/s41540-024-00372-2.

[94] H. Lv, F. Sun, and Y. Chen, "Decoding congenital heart disease: a multi-omic framework for cardiac lineage and regulatory dysfunction," *Frontiers in Cell and Developmental Biology*, vol. 13. Frontiers Media, Sep. 18, 2025. doi: 10.3389/fcell.2025.1659884.

[95] A. Maytum, N. Obier, P. Cauchy, and C. Bonifer, "Regulation of developmentally controlled enhancer activity by extrinsic signals in normal and malignant cells: AP-1 at the

centre," *Frontiers in Epigenetics and Epigenomics* , vol. 2, Sep. 2024, doi: 10.3389/freae.2024.1465958.

[96] G. Furlan, A. Huyghe, N. Combémorel, and F. Lavial, "Molecular versatility during pluripotency progression," *Nature Communications*, vol. 14, no. 1, p. 68, Jan. 2023, doi: 10.1038/s41467-022-35775-4.

[97] E. Akıncı, M. C. Hamilton, B. Khowpinitchai, and R. I. Sherwood, "Using CRISPR to understand and manipulate gene regulation," *Development*, vol. 148, no. 9, Apr. 2021, doi: 10.1242/dev.182667.

[98] B. Noordijk, M. L. G. Gomez, K. ten Tusscher, D. de Ridder, A. D. J. van Dijk, and R. W. Smith, "The rise of scientific machine learning: a perspective on combining mechanistic modelling with machine learning for systems biology," *Frontiers in Systems Biology*, vol. 4. Frontiers Media, Aug. 02, 2024. doi: 10.3389/fsysb.2024.1407994.

[100] P. C. K. Chow and P. J. Bentley, "Development necessitates homeodomain factors," *Research Square (Research Square)* , Oct. 2024, doi: 10.21203/rs.3.rs-5010355/v1.

[101] Y. Uchida and N. Irie, "Toward mechanisms underlying the developmental hourglass: evolvability and phylotypic conservation," *EvoDevo* , vol. 17, no. 1, p. 2, Dec. 2025, doi: 10.1186/s13227-025-00255-5.

[102] T. Kohsokabe, S. Kuratanai, and K. Kaneko, "Developmental hourglass: Verification by numerical evolution and elucidation by dynamical-systems theory,"*PLoS Computational Biology* , vol. 20, no. 2, Feb. 2024, doi: 10.1371/journal.pcbi.1011867.

[103] B. I. Li, M. J. Alvarez, H. Zhao, N. Chirathivat, A. Califano, and M. M. Shen, "The regulatory architecture of the primed pluripotent cell state," *Nature Communications*, vol. 16, no. 1, p. 3351, Apr. 2025, doi: 10.1038/s41467-025-57894-4.

[104] Q. Kong, H. Peng, Q. Zhao, H. Jiang, and X. Zhu, "The Xenopus model as a tool for investigating craniofacial developmental disorders," *Frontiers in Medicine*, vol. 12. Frontiers Media, Sep. 08, 2025. doi: 10.3389/fmed.2025.1671687.

[105] S. R. F. Twigg, N. D. E. Greene, D. J. Henderson, P. Mill, and K. Liu, "The power of mouse models in the diagnostic odyssey of patients with rare congenital anomalies," *Mammalian Genome* . Springer Science+Business Media, Mar. 18, 2025. doi: 10.1007/s00335-025-10114-2.

[106] T. M. Hoard, "Identification of Semaphorin Receptors as Novel Regulators of Hedgehog and Wnt Signaling," *Deep Blue (University of Michigan)* , Jan. 2024, doi: 10.7302/24087.

[107] J. R. York *et al.* , "Shared features of blastula and neural crest stem cells evolved at the base of vertebrates," *Nature Ecology & Evolution* , vol. 8, no. 9, p. 1680, Jul. 2024, doi: 10.1038/s41559-024-02476-8.

[108] M. Kumar, "The Precision Oncology Approach to Molecular Cancer Therapeutics Targeting Oncogenic Signaling Pathways is a Means to an End," *SSRN Electronic Journal* , Jan. 2024, doi: 10.2139/ssrn.4693638.

[109] M. Yagi, J. E. Horng, and K. Hochedlinger, "Manipulating cell fate through reprogramming: approaches and applications," *Development*, vol. 151, no. 19, Sep. 2024, doi: 10.1242/dev.203090.

[110] M. Matiukhova *et al.* , "A comprehensive analysis of induced pluripotent stem cell (iPSC) production and applications," *Frontiers in Cell and Developmental Biology* , vol. 13, p. 1593207, May 2025, doi: 10.3389/fcell.2025.1593207.

[111] R. Rouco, A. Rauseo, G. Sapin, O. Bompadre, F. Darbellay, and G. Andrey, "Temporal constraints on enhancer usage shape the regulation of limb gene transcription," Mar. 2024, doi: 10.1101/2024.03.22.585864.

[112] M. van der Sande, S. Frölich, and S. J. van Heeringen, "Computational approaches to understand transcription regulation in development," *Biochemical Society Transactions*, vol. 51, no. 1. Portland Press, p. 1, Jan. 25, 2023. doi: 10.1042/bst20210145.

[113] M. Yu *et al.*, "Integrative analysis of the 3D genome and epigenome in mouse embryonic tissues," *Nature Structural & Molecular Biology*, vol. 32, no. 3, p. 479, Dec. 2024, doi: 10.1038/s41594-024-01431-2.

[114] P. Xie *et al.*, "Digital reconstruction of full embryos during early mouse organogenesis," *Cell*, vol. 188, no. 17, p. 4754, Jun. 2025, doi: 10.1016/j.cell.2025.05.035.