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HOX genes: molecular genetics and effect of mutation

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In vertebrates, the axial skeleton, limbs, stomach, urogenital tract, and external genitalia all develop partly controlled by the homeobox-containing HOX gene families, which are deeply conserved throughout animal evolution. Much knowledge has been learned about the functions of HOX genes in many physiological and pathologic processes since their original discovery. There are 39 HOX genes in mammals, separated into the HOX A, B, C, and D clusters. Thirty-nine homeobox genes in mammals are clustered into four clusters: HOX A–D, with a correlation between mutations of HOXB13, most often G84E, with breast, colorectal, and early-onset prostate cancer in humans. Here, we discuss the current knowledge of homeobox signaling in genitourinary development and cancer and the role of homeobox protein cofactors in both development and cancer.

Keywords:
Bosley–Salih–Alorainy syndrome, human HOX disorders, prostate cancer, synpolydactyly type II

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Introduction

HOX genes, crucial for embryonic development, form a conserved family of transcription factors. Initially identified in the Drosophila fish cluster in 1984, they are structurally and functionally conserved across the animal kingdom [1]. Emerging from a single prototype HOX gene through tandem duplication and divergence, they gave rise to multiple clusters. Each fish cluster has 9–11 genes, with over a hundred homeobox-containing genes in humans, while humans and most vertebrates have 39 HOX genes divided into four groups (HOX A–D). Each fish cluster is â 120 kb long, with 9–11 genes arranged in the same 5′ to 3′ direction. The genes within each cluster are numbered based on sequence identity and relative locations. Despite losing distinct genes from each cluster, the same subgroups are maintained in all vertebrate clusters [2].

HOXA1

Molecular genetics

HOXA1, like other HOX genes, comprises two coding exons, with a missense mutation identified in exon one. Bosley–Salih–Alorainy syndrome is associated with three distinct homozygous mutations: c.185delG, c.175-176insG, and c.84CNG. While patients of Turkish ancestry exhibit the c.84CNG mutation, those of Saudi Arabian ancestry carry the c.185delG and c.175-176insG mutations. The Athabascan brainstem dysgenesis syndrome (ABDS) in individuals of Athabascan heritage is linked to the homozygous c.76CNT mutation. All four mutations are predicted to result in a translationally shortened protein lacking the necessary homeodomain for DNA binding [3].

Clinical features

Bosley–Salih–Alorainy syndrome

Initially diagnosed in nine Saudi Arabian and Turkish individuals, this condition is characterized by congenital horizontal gaze palsy, sensorineural hearing loss, inner-ear abnormalities, delayed motor milestones, and internal carotid artery malformations. Neuroimaging reveals common internal carotid artery malformations, ranging from unilateral hypoplasia to bilateral agenesis. Normal brain function relies on a healthy brainstem, cerebellum, and cerebrum. Additional, less frequent manifestations include facial paresis, limb anomalies, seizures, autism spectrum disorder, facial twitching, tetralogy of Fallot, total anomalous pulmonary venous return, interrupted aortic arch, and various cardiovascular

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malformations such as ventricular septal defect and tetralogy of Fallot. Dysmorphisms may involve bony facial asymmetry, low-set ears, flattened ear helices, and limb malformations like polydactyly, brachydactyly, and/or clubfoot [4,5].

Athabascan brainstem dysgenesis syndrome

ABDS, the more severe of the two HOXA1-related illnesses, has been observed exclusively in individuals with Athabascan heritage. Patients typically present with significant intellectual disability, horizontal gaze palsy, facial and bulbar weakness, central hypoventilation, and conotruncal cardiac abnormalities. Additional less frequent abnormalities may include internal carotid artery anomalies, facial twitching, face and bulbar paresis, and seizure disorders. The severe intellectual incapacity observed in ABDS could be attributed to global cerebral hypoxia resulting from hypoventilation, cerebrovascular abnormalities, and the high altitude of residence among the Athabascan tribe. Distinctive features of ABDS, as opposed to the related Brown–Sequard syndrome, include central hypoventilation, bulbar paresis, and the absence of limb abnormalities or facial dysmorphisms. Molecularly confirmed cases include 16 patients with Brown–Sequard syndrome and 13 with ABDS reported to date [5,6].

HOXD13

Molecular genetic

The etiology of HOXD13 limb morphopathies involves various mutation classes. Polyalanine expansion, similar to SPD type II, was the first mutation discovered in HOXD13. Transcriptional factors often contain conserved polyalanine tracts, typically with 20 or fewer alanine residues. The size of HOXD13 expansions correlates with the severity and penetrance of the phenotype, with seven alanine expansions required for synpolydactyly. These expansions likely induce disease through a dominant-negative mechanism, causing the mutant protein to aggregate within the cell. Mutations leading to atypical SPD with foot anomalies include truncating deletions and missense mutations, thought to induce disease via a dominant loss-of-function/haploinsufficiency pathway. The missense mutation c.964ANC, I322L, associated with brachydactyly classes D and E, has a complex etiology with selective loss of function depending on the target under investigation. Another mutation, c.947CNG; S316C, causes brachydactyly similar to the wild type and may also induce the disease through a dominant-negative process. Seven alanine contractions result in a brachydactyly–syndactyly phenotype, potentially through a dominant-negative mechanism or a loss-of-function process due to variations in the protein's secondary structure. This contraction was identified in a female patient with VACTERL association, including vertebral deformities, anal atresia, heart defects, tracheo-esophageal fistula, renal anomalies, and limb abnormalities. The significance of the HOXD13 mutation in the involvement of other organ systems is unknown, and more instances of the same contractions are needed to validate the proposed pathophysiology [7,8].

Clinical features

Following the discovery of HOXD13 polyalanine expansions in synpolydactyly type II (OMIM #186000), HOXD13 mutations were also identified to cause brachydactyly type D (OMIM #113200), brachydactyly type E (OMIM #113300), brachydactyly–syndactyly syndrome (OMIM #610713), and syndactyly type V (OMIM #186300). A HOXD13 intragenic deletion has also been found in a female patient with the VACTERL association (vertebral deformities, anal atresia, cardiac issues, tracheo-esophageal fistula, renal anomalies, and limb abnormalities), which is significant given the clinical overlap across the many illnesses. The range of limb abnormalities brought on by HOXD13 mutations has been dubbed "HOXD13 limb morphopathies" [7,9,10].

Synpolydactyly type II

Synpolydactyly, associated with HOXD13 gene alterations, represents the first limb abnormality linked to HOX gene mutations. Synpolydactyly type II (SPD), an autosomal dominant condition with incomplete penetrance (97%) and variable expressivity, manifests as soft-tissue syndactyly between the third and fourth fingers and the fifth and fourth toes, along with postaxial polydactyly affecting these digits. A sizeable Turkish pedigree with 182 affected individuals revealed four significant categories of malformations: postaxial polydactyly type A, preaxial and postaxial synpolydactyly, typical synpolydactyly (84.9% of the kindred), and bilateral, fully synpolydactyly due to HOXD13 gene homozygosity. Variable traits include the absence of middle phalanges, clinodactyly, camptodactyly, brachydactyly of the fifth digits, syndactyly, and brachydactyly of the second to fifth toes. Men with a significant polyalanine expansion also exhibit hypospadias symptoms. Patients with typical SPD often have structural or duplicational metacarpal abnormalities

(80.7%) and duplicational metatarsal abnormalities (83.9%). Homozygosity for the causative polyalanine expansion results in a more severe phenotype, including complete soft-tissue syndactyly of all four limbs, polydactyly of various digits, and severe bone abnormalities. Intragenic deletions and missense mutations (c.916CNT, R306W; c.683GNT, G228V) inside HOXD13 lead to atypical SPD cases. Frameshifting HOXD13 deletions exhibit a dominant hereditary pattern with unique foot deformities, while the missense mutant R306W results in a partially penetrant SPD phenotype. The G220V HOXD13 mutation causes a mild SPD phenotype similar to mild polyalanine tract expansions (Ala residues, 7–9 more), with outcomes ranging from synpolydactylous third and fourth digits to clinodactylous fifth fingers and camptodactylous fifth toes [11].

Brachydactyly types D and E

Missense mutations in the homeodomain of HOXD13 (c.947CNG, S316C; c.964ANC, I322L) overlap with brachydactyly types D and E (OMIM #13200 and 13300, respectively). Shorter metacarpals and metatarsals characterize brachydactyly type E, while brachydactyly type D features shorter and wider terminal phalanges on fingers and halluces. Individuals with the HOXD13 I322L mutation exhibit widespread brachydactyly and additional abnormalities, including phalangeal duplication in the fourth digit and syndactyly in the third and fourth fingers. Other observed features include clinodactyly in the fourth and fifth fingers and missing or immature nails on the fifth toe. The varied phenotypes induced by the HOXD13 missense gene S316C include distal phalangeal (typically the first and fifth) shortening or extension, metacarpal, and/or metatarsal shortening, and wrist bone fusion [9,12].

Syndactyly type V

A significant HOXD13 missense mutation (c.974ANG; Q325R) was identified in a large Han Chinese family with syndactyly type V, characterized by metatarsal and metacarpal union. Syndactyly type V was investigated in 23 individuals, revealing varying degrees of impairment with the extent of fusing differing among affected individuals. Expanding polyalanine in HOXD13 leads to synpolydactyly, which includes features such as syndactyly between the third and fourth digits, syndactyly with an extra digit between digits 3 and 4, and hypoplastic fifth middle phalanx with duplicated third, middle, and distal phalanges. Axially deformed thumbs exhibit short first metacarpals, duplicated third distal, middle, and proximal phalanges, bifid third metacarpals, and hypoplastic fifth middle phalanges. Syndactyly between the fourth and fifth digits, polysyndactyly with an extra toe, and synostosis of the phalanges of the fourth and fifth fingers may occur. Other hand deformities include ulnar deviation in fingers 2 through 5, cutaneous syndactyly of fingers 3 and 4, finger camptodactyly, and small distal phalanges affecting multiple digits. Foot findings involve varus deviation of the first metatarsals, hypoplasia and shortening of metatarsals 2 through 5, and significant cutaneous syndactyly of toes 2 and 3 and/or digits 3 and 4. Additionally, hypospadias and postaxial polydactyly were discovered in one male [13].

Brachydactyly–syndactyly A

A 21-base pair deletion (c.157_177del) in HOXD13, causing a contraction of seven alanine residues from the extensive 15-residue polyalanine region in exon 1, was identified as the cause of a new brachydactyly–syndactyly syndrome in a Han Chinese family. Individuals with this condition exhibit generalized brachydactyly of the hands and feet, wide and short thumb distal phalanges, cutaneous syndactyly of toes 2 and 3, absence of the middle phalanges of toes 2 through 5, and small middle phalanges of the fifth digit. While most limb anomalies are typically bilateral, other radiological anomalies may also be present. Overall, the appearance resembles syndactyly type I and brachydactyly types A4, D, and E [13].

VACTERL association

A female patient with VACTERL association underwent a de novo seven alanine contraction in the big polyalanine portion of HOXD13. Her characteristics included bilateral vesicoureteral reflux, anal atresia, tetralogy of Fallot, and fusion of the fourth and fifth finger's distal interphalangeal joints [13].

HOXB13

Molecular genetics

The predominant missense variants identified include the HOXB13 G84E mutation, which notably impacts individuals of European ancestry, particularly in Nordic nations with the highest carrier frequencies [14]. Various missense variants associated with an increased cancer risk have been identified in diverse ethnic groups. Exceptionally, one patient bears the c.853delT mutation, causing the wild-type protein to elongate by 96 amino acids and eliminate a stop codon,

with all other mutations being missense [15]. The G84E variant in exon 1 adds glycine at position 29 in the MEIS interaction domain, susceptible to missense mutations, as is the homeodomain [16]. The population-based prostate cancer risk of G84E was calculated risk of G84E based on carriers' age and year of birth. Despite the frequent occurrence of the G84E mutation, unknown pathogenic mechanisms persist. A gain-of-function mechanism was proposed due to the absence of truncating mutations and the prevalence of the G84E mutation [16,17]. Notably, HOXb13 loss-of-function mutant mice exhibit larger tail bud structures due to increased cell proliferation and reduced mortality [17,18].

Clinical features

The G84E HOXB13 missense mutation, first identified by researchers, is more prevalent in families with early-onset prostatic cancer [16]. Subsequent research confirmed these early findings, indicating that men with the G84E mutation face a 5–10 times increased risk of developing prostate cancer. This mutation is present in 1–5% of European families with prostate cancer, with a significantly higher prevalence in Nordic nations [19–21]. The association is more potent in individuals with notable family histories and a younger age at cancer diagnosis (55 years old). Notably, the G84E missense mutation has not been detected in many other ethnic groups, including Ashkenazi Jewish, Chinese, or African populations [20,21]. Most affected individuals are of European heritage. Chinese men, on the other hand, are more likely to be affected by prostate cancer due to a different, less common mutation (p.G135E) and 404GNA [22].

Breast cancer

Studies on the association between HOXB13 mutations and breast cancer are inconclusive. The bigger of the two studies failed to discover a link [22–24].

HOXD10

Molecular genetics

Genome-wide linkage and candidate gene studies were employed to identify a heterozygous missense mutation (c.956TNA; M319K) in the HOXD10 gene associated with the condition [25]. This HOXD10 homeodomain mutation has the potential to trigger the disease either through haploinsufficiency or a gain of new function. Mice lacking the HOXD10 gene displayed fewer nerves innervating the hindlimb muscles and alterations in the bones and spinal column of the hindlimb [26]. Mutant mice exhibited distinctive features, including aberrant patellar position, outward rotation of the lower leg, sacral vertebrae homeotic transition to the next-most anterior vertebrae, and sporadic formation of an anterior sesamoid bone [25,27].

Clinical features

A missense mutation in the HOXD10 gene was identified in 17 relatives with isolated CVT, CMT, or a combination of both conditions. Among these individuals, 71% had bilateral isolated CVT, 12% had bilateral CMT, 12% had both CVT and CMT, and 6% had bilateral CVT in one foot and bilateral CMT in the other. Importantly, each person exhibited a normal IQ and had no hand or spinal abnormalities [27].

HOXA2

Clinical description

Only four individuals have been reported with HOXA2-associated autosomal recessive microtia, all belonging to a single consanguineous Iranian family, with only three undergoing detailed clinical evaluation. Common facial features in these patients comprise grade II microtia, a short and narrow auditory canal, cleft palate, and occasional unilateral facial paresis. Radiographic findings may include unilateral or bilateral hypoplastic tympanic membranes, poorly developed mastoid air cells, a small middle ear cavity, and absent inner-ear structures. All affected individuals exhibit bilateral severe to profound mixed hearing impairment across all tested frequencies. Heterozygous carriers appear unaffected, displaying normal audiometric testing and external ear anatomy [28,29].

Molecular genetics

Through genome-wide linkage analysis, a homozygous mutation (c.556CNA; p.Q186K) was detected in affected individuals. The glutamine at position 186, crucially conserved in evolution, impacts position 44 of the homeodomain, particularly the recognition helix. Similar findings, including microtia and cleft palate, are observed in the HOXA2-/- mouse. Notably, sequencing of HOXA2 and SIX2, a downstream target of HOXA2 in mice, in eight Hispanic and African-American nonsyndromic microtia patients revealed no pathogenic mutations, indicating that HOXA2 is likely not a frequent cause of nonsyndromic microtia [30].

HOXA11

Clinical description

Afflicted patients primarily exhibit radioulnar synostosis and thrombocytopenia as the main symptoms. Two unrelated families identified by

Thompson and Nguyen each had men and their children affected by radioulnar synostosis, with three out of four children also experiencing symptomatic thrombocytopenia. The thrombocytopenia induced congenital bruising and bleeding, requiring treatment through bone marrow or umbilical cord stem cell transplantation [31].

Molecular genetics

All affected individuals experienced a one base-pair HOXA11 deletion (c.872delA), serving as the cause of RUSAT. This deletion results in a protein truncation by 22 amino acids, leading to a frameshift and a premature translational stop codon in the exon 2 homeodomain. HOXA11 mutant mice, whether homozygous or heterozygous, display forelimb and hindlimb deformities, but thrombocytopenia is not observed [31].

It was demonstrated that uterosacral ligaments were absent in HOXA11/ mice, suggesting that HOXA11 is essential for their formation. In contrast to a group of 10 women with normal pelvic support, a cohort of 18 women requiring surgery for symptomatic pelvic organ prolapse exhibited decreased uterosacral ligament HOXA11 expression [31].

HOXA13

Clinical description

Hand–foot–genital syndrome (HFGS, OMIM #140000) and Guttmacher syndrome (OMIM #176305) both result from heterozygous mutations in HOXA13 [32,33]. HFGS, an autosomal dominant syndrome, is characterized by limb deformities and urogenital abnormalities [33,34]. In addition to urogenital and limb deformities like those in HFGS, postaxial polydactyly is an extra hand finding in Guttmacher syndrome. Linked explicitly to Guttmacher syndrome are individuals with a particular missense mutation within the homeodomain. In contrast, HFGS is caused by numerous factors, including HOXA13 polyalanine expansions, point mutations, and uncommon heterozygous locus chromosomal deletions [35].

Hand–foot–genital syndrome

HFGS, an autosomal dominant disease, manifests with limb and urogenital abnormalities. Urogenital anomalies exhibit around 50% penetrance, while skeletal deformities are entirely penetrant. The characteristic feature of HFGS involves bilateral thumb and hallux hypoplasia, resulting from the shortening of the distal phalanx and/or the first metacarpal or metatarsal. Other limb abnormalities include hypoplasia of the thenar eminences, hallux varus, tiny hallux toenail, fifth finger clinodactyly, or short feet. Radiographically, HFGS is characterized by the thumb's pointed distal phalanges and first metacarpal, small calcaneus, and occasional bone fusions of the middle and distal phalanges of the second through fifth toes. Thumb and hallucis shortening is generally moderate but may be more severe in individuals with missense mutations. In afflicted females, incomplete Müllerian fusion, vesicoureteral reflux, urethral hypospadias, urinary incontinence, and trigonal hypoplasia can be present to varying degrees. Urogenital defects in boys may include hypospadias with or without chordee, ranging from moderate to severe. Notably, fertility remains normal in those affected by HFGS [35].

Guttmacher syndrome

Similar to HFGS but with extra limb abnormalities, Guttmacher syndrome is characterized by limb and urogenital anomalies. Postaxial polydactyly of the hands and small or uniphalangeal second toes with missing nails are particular skeletal anomalies [35].

Molecular genetics

In the first described HFGS family, Mortlock and Innis discovered a HOXA13 nonsense mutation (c.1107GNA, W369X). Since then, 16 distinct mutations have been identified in HOXA13, comprising 40% point mutations and 60% polyalanine expansions. Pathogenic expansions have been observed in three substantial polyalanine tracts within HOXA13, with reported extra Ala residues ranging from six in Tract II to 14 in Tract III. In contrast to HOXD13, HOXA13 polyalanine tract III contractions have been noted in healthy individuals, suggesting benign variations. Both missense and nonsense mutations are considered pathogenic point mutations [36].

The missense mutation c.1114ANC, altering the 51st homeodomain residue from asparagine to histidine, induces a more severe phenotype than nonsense mutations or polyalanine expansions. While the reported missense mutation likely acts through a mixed loss-of-function/gain-of-function mechanism, polyalanine expansions, and nonsense mutations predominantly lead to illness through a loss-of-function mechanism. The initial family associated with Guttmacher syndrome presented a specific missense mutation, c.1112ANT; Q371L was later discovered. On the same allele, a 2-base pair deletion in the promoter region (79-79delGC) was

found, though it has no phenotypic effects on its own. The substituted glutamine is a crucial residue in the homeodomain, expected to contribute to both functional gain and loss [36].

HOXB1

Clinical features

Four patients, organized into two sibling pairs, have been confirmed to possess homozygous HOXB1 mutations. Noteworthy characteristics associated with these mutations include congenital facial palsy, hearing loss, strabismus, midface retrusion, and an upturned nose. Additional observed features encompass a smooth philtrum, posteriorly oriented ears, eating difficulties, and speech delay. In one of the reported cases among the four patients, brain imaging revealed aberrant tapering of the cochlea's basal turn and facial nerve agenesis [37]. While one carrier father from a sibling pair had a history of bilateral sensorineural hearing loss and speech delay, with no brain imaging conducted, carriers generally appear unaffected [38].

Molecular genetics

Due to consanguinity in the parents of one pair of affected siblings, autosomal recessive inheritance was suggested in that particular case. Through homozygosity mapping and exome sequencing, a homozygous R207C mutation in HOXB1, specifically c.619CNT, was identified [38]. Another pair of affected siblings also exhibited homozygosity for the same mutation [39]. The mutation affects a highly conserved arginine in the protein's homeodomain, impacting DNA binding. The observed phenotype in the four reported individuals closely resembles that of HOXB1−/− mice, displaying abnormal contralateral vestibuloacoustic neuron migration and the absence or reduction of the facial motor nerve, potentially explaining the hearing loss [40]. Based on this information, the human mutation is considered an allele causing a loss of function [41].

HOXC13

Clinical features

HOXC13 mutations cause homozygous dysfunction resulting in ectodermal dysplasia 9, also known as pure hair and nail ectodermal dysplasia (OMIM #602032) [42]. This rare autosomal recessive disorder has been identified in consanguineous Chinese and Pakistani families [43]. In contrast to other HOX genes, HOXC13 exhibits a departure from the spatial collinearity principle, expressing itself at later developmental stages in the vibrissae, filiform papillae of the tongue, and all body hair follicles [44]. Ectodermal dysplasia 9, a hair/nail type, manifests primarily as nail dystrophy and hypotrichosis [45]. Hypotrichosis varies from mild hair loss to congenital atrichia, with noticeable nail degeneration from birth. Importantly, individuals with this condition exhibit normal neurological function, skeleton, sweat glands, eyes, teeth, and overall development. Interestingly, heterozygous carriers of loss-of-function mutations experience normal growth of hair and nails without any adverse effects [46].

Molecular genetics

All individuals affected by ectodermal dysplasia 9 exhibit homozygous, loss-of-function mutations in HOXC13. Notably, six families, including cases with mutations such as c.390CNA, p.Y130X; 27.6 kb deletion affecting exon 1 and part of the intron; c.355delC, p.L119Wfs*20; c.200_203 dupGCCA, p.H68Qfs*84; and c.404CNA, p. S135X, have been identified, all involving unique truncating mutations in exon 1 of HOXC13. An afflicted patient showed poor protein staining in hair follicles and reduced HOXC13 mRNA levels in the skin tissue, suggesting either nonsense-mediated mRNA decay or protein instability. In experiments with mice lacking the HOXC13 protein, baldness and nail abnormalities were observed, closely mirroring the human phenotype associated with HOXC13 mutations [47].

HOXD4

Clinical description

In a group of 86 children diagnosed with acute lymphoid malignancy, either with or without skeletal abnormalities, two individuals were identified with a heterozygous missense substitution in the HOXD4 gene, as reported in a study [48]. Among these two patients, only one exhibited a skeletal anomaly characterized by bilateral cervical ribs and right sacralization of L5. Notably, both patients had hematologic malignancies [49]. The identified mutation was inherited by both individuals from parents who did not show any apparent impact from the mutation [50].

Molecular genetics

Both affected patients inherited a missense mutation in HOXD4, specifically c.242ANT; p.E81V, from an unaffected parent [51,52]. The mutant, E81V, demonstrated â 40% lower transcriptional activity compared to another allele, as determined through a luciferase gene and HOXD4 autoregulatory enhancer reporter construct, indicating a partial loss-of-function

Table 1 Human HOX gene disorders

Conditions	Gene	Chr	OMIM #	Inheritance	Phenotype	Mechanism
Bosley–Salih–Alorainy syndrome	HOXA1	7	601536	AR	Horizontal gaze palsy, SNHL, ID, cardiac defects, facial dysmorphisms and limb anomalies	LOF
Athabascan brainstem dysgenesis syndrome	HOXA1	7	601536	AR	Horizontal gaze palsy, SNHL, ID, cardiac defects, central hypoventilation	LOF
Breast and prostate cancer susceptibility	HOXB13	17	-	AD	Increased incidence of prostate and breast cancer	?
Synpolydactyly type II	HOXD13	2	186000	AD	Hand and feet SPD, rarely hypospadias	Dominant negative (PA expansion) and LOF (deletions and missense mutations)
Brachydactyly types D and E	HOXD13	2	113200/ 113300	AD	Generalized brachydactyly, fifth finger distal hypoplasia/aplasia, phalangeal duplication, fingers 3–4 syndactyly, metacarpal/metatarsal shortening	LOF
Syndactyly type V	HOXD13	2	186300	AD	Metacarpal synostosis, fifth finger	Mixed LOF and GOF
Brachydactyly–syndactyly	HOXD13	2	610713	AD	clinodactyly, cutaneous syndactyly of fingers 3 and 4, mild cutaneous toe syndactyly, generalized brachydactyly of hands, broad and short distal thumb phalanges, cutaneous toe syndactyly, absence of middle phalanges of toes 2–5, 5th finger clinodactyly	Dominant negative and LOF

[52]. The identified HOXD4 mutation in both cases was found to be linked with three unknown HOXD cluster variants, suggesting a potential combinatorial effect [12,52,53]. Notably, no additional cases involving HOXD4 mutations have been documented to date. In experiments with mice, both heterozygous and homozygous HOXD4 mutants, while remaining alive and productive, exhibited cervical spine abnormalities akin to those observed in the single heterozygote person. Interestingly, HOXD4 -/- mice did not display hematological abnormalities, in contrast to the diminished bone marrow and liver hematopoietic stem cell proliferative ability seen in paralogous HOXB4-/- animals [54,55] (Tables 1 and 2).

Conclusion

In summary, this research delves into HOX genes and their impact on our physical development. It uncovers how mutations in these genes can result in various health issues, like limb abnormalities or hearing problems. The study offers valuable insights for further research, paving the way for a better understanding of genetic conditions.

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Table 2 Summary of HOX genes with corresponding syndrome

Conditions	Gene	Chromosome	Phenotype
Bosley–Salih–Alorainy syndrome	HOXA1	7	SNHL, cardiac defects, and limb anomalies
Synpolydactyly type II	HOXD13	2	Hand and feet SPD and rarely hypospadias
Breast and prostate cancer susceptibility	HOXB13	17	Increased incidence of prostate and breast cancer
Charcot–Marie–Tooth disease and hereditary vertical talu	HOXD1	2	Foot deformities (pes cavus and hammer toes)
Autosomal recessive microti	HOXA2	7	Narrowed auditory canal and cleft palate
Radioulnar synostosis and thrombocytopenia	HOXA11	7	Fusion or improper development of radius or ulna bone in forearm and thrombocytopenia
Hand–foot–genital syndrome	HOXA1	7	Small thumb and big toes, inward deviation of big toe, and hypospadias
Congenital facial palsy and hearing loss	HOXB1	17	Impaired facial movement and hearing loss

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Conflicts of interest
There are no conflicts of interest.

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