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Genetics of Congenital Hand Anomalies

Die Genetik angeborener Handfehlbildungen

Abstract

Congenital limb malformations exhibit a wide spectrum of phenotypic manifestations and may occur as an isolated malformation and as part of a syndrome. They are individually rare, but due to their overall frequency and severity they are of clinical relevance. In recent years, increasing knowledge of the molecular basis of embryonic development has significantly enhanced our understanding of congenital limb malformations. In addition, genetic studies have revealed the molecular basis of an increasing number of conditions with primary or secondary limb involvement. The molecular findings have led to a regrouping of malformations in genetic terms. However, the establishment of precise genotype-phenotype correlations for limb malformations is difficult due to the high degree of phenotypic variability. We present an overview of congenital limb malformations based on an anatomic and genetic concept reflecting recent molecular and developmental insights.

Key words
Limb development · malformation · syndactyly · polydactyly · brachydactyly · genetics

Zusammenfassung

Angeborene Handfehlbildungen sind durch ein breites Spektrum an phänotypischen Manifestationen gekennzeichnet. Sie treten als isolierte Malformation oder als Teil verschiedener Syndrome auf. Die einzelnen Formen kongenitaler Handfehlbildungen sind selten, besitzen aber aufgrund ihrer Häufigkeit insgesamt und der hohen Belastung für Betroffene erhebliche klinische Relevanz. Die fortschreitende Erkenntnis über die molekularen Mechanismen der Embryonalentwicklung haben in den letzten Jahren wesentlich dazu beigetragen, die genetischen Ursachen kongenitaler Malformationen besser zu verstehen. Der hohe Grad an phänotypischer Variabilität kongenitaler Handfehlbildungen erschwert jedoch eine Etablierung präziser Genotyp-Phänotyp-Korrelationen. In diesem Übersichtsartikel präsentieren wir das Spektrum kongenitaler Malformationen, basierend auf einer entwicklungsbiologischen, anatomischen und genetischen Klassifikation unter Berücksichtigung der Bedeutung neuerer molekularer und entwicklungsbiologischer Erkenntnisse.

Schlüsselwörter
Angeborene Handfehlbildungen · Genetik · Embryologie · Syndaktylie · Polydaktylie · Brachydaktylie

Introduction

Congenital hand malformations are a genetically and clinically heterogeneous group of disorders which present as an isolated trait or as part of a syndrome. They exhibit a wide spectrum of manifestations, phenotypic variability and genetic heterogeneity. For a review on skeletal malformations see Kornak and Mundlos [30]. They are individually rare, but due to their overall frequency and severity they are of clinical relevance. Congenital limb malformations may be caused by genetic or environmental

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Received: 11. 9. 2003 · **Accepted:** 2. 11. 2003

Bibliography
Handchir Mikrochir Plast Chir 2004; 36: 85 – 97 © Georg Thieme Verlag KG Stuttgart · New York · ISSN 0722-1819 · DOI 10.1055/s-2004-817884

factors disturbing the regular developmental program during embryogenesis. The study of the molecular mechanisms underlying human genetic diseases, the analysis of animal model organisms and the use of transgenic mouse technology have considerably helped elucidate the genetic basis of limb development and malformation in the last years. These findings have strongly influenced the genetic classification of congenital limb malformations (see Glossary). In addition, a number of non-genetic conditions exist that are mainly caused by external factors such as amniotic bands, vascular disruptions or teratogens. In this article we present an overview covering hand malformations in an isolated form or as part of a syndrome, based on anatomic and genetic observations and taking into account the significance of recent molecular and developmental data (see Fig. **1** and Table **1**). Accordingly, we will focus on conditions with known molecular and genetic causes, refraining from a surgical classification.

Embryonic Limb Development

During development, the limb forms as a result of a dual contribution from the lateral plate and the somitic mesoderm (Fig. **2**; [28, 35]). The limb buds of the embryo are formed at the lateral flank by proliferation of cells from the lateral plate. Shortly thereafter, cells from the lateral edges of adjacent somites invade the limb bud to form limb muscles, nerves and vessels. The limb bud increases in size and the individual skeletal elements are laid down in a proximodistal fashion. Different signaling centers control the outgrowth and patterning of the limbs in three dimensions (Fig. **2**). The outgrowth of the limb along the proximodistal axis is directed by signals from the overlying ectoderm which forms a specialized epithelial structure at its distal tip, the apical ectodermal ridge (AER). Experimental removal of the AER leads to limbs with different severities of truncations, depending on the time of removal. Subsequently, it has been shown that fibroblast growth factors *(Fgfs)* represent key factors that mediate the outgrowth of the limb.

The anteroposterior patterning of the limb is controlled by the zone of polarizing activity (ZPA), consisting of mesenchymal cells at the posterior margin of the limb bud. Experimental grafting of the ZPA to the anterior margin of the limb bud leads to mirror image duplications of skeletal elements. *Sonic hedgehog (Shh),* a secreted molecule related to Drosophila *Hedgehog (Hh),* has been identified as the molecule responsible for the morphogenetic properties of the ZPA. *Shh* regulates its target genes by controlling the balance of *Gli3* repressor and activator.

Dorsoventral patterning results in the formation of the dorsal versus the palmar side of the hand including the formation of an ordered structure of muscles, tendons and nails. Originally, surgical manipulations involving rotation of the chick limb bud ectoderm suggested that the non-AER ectoderm is responsible for specifying the fate of cells along the dorsoventral axis. Subsequent findings now indicate that the dorsal identity of the limb is controlled by the genes *Wnt7a* and *Lmx1,* whereas *En1* is responsible for the ventral identity [43].

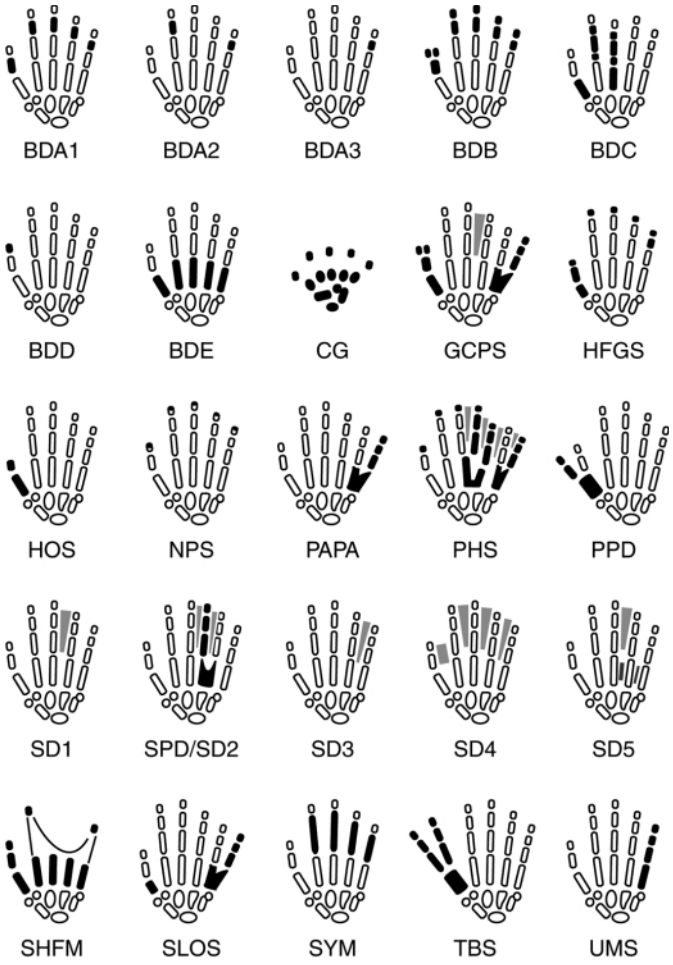

Fig. **1** Congenital limb anomalies and their anatomic basis. Schematic representation of congenital limb malformations appearing as an isolated trait or in association with a syndrome. Abbreviations: BDA – Brachydactyly Type A; BDB – Brachydactyly Type B; BDC – Brachydactyly Type C; BDD – Brachydactyly Type D; BDE – Brachydactyly Type E; CG – Chondrodysplasia Grebe; GCPS – Greig cephalosynpolydactyly; HFGS – Hand-foot-genital syndrome; HOS – Holt-Oram syndrome; NPS – Nail-patella syndrome; PAPA – Postaxial polydactyly; PHS – Pallister-Hall syndrome; PPD – Preaxial Polydactyly; SD – Syndactyly; SHFM – Split-hand/-foot malformation; SLOS – Smith-Lemli-Opitz syndrome; SYM – proximal symphalangism; TBS – Townes-Brocks syndrome, UMS – Ulna-mammary syndrome.

Despite our knowledge of outgrowth and patterning, the mechanisms underlying the specification of cells in the early limb bud remain unclear. In order to form distinct skeletal elements, the cells in the early limb bud are specified and receive positional information during development. Two models, the "progress zone model" and the "early specification model", have been proposed to explain the underlying developmental mechanisms (Fig. **2**). The progress zone model suggests that cells acquire their positional information progressively in a proximodistal manner. In this model, the AER facilitates specification by maintaining the cells in the underlying progress zone in an immature condition. The cells are specified as they leave the PZ, indicating that cells that leave the PZ late will acquire a more distal fate. According to the early specification model, a specified cell population expands as the limb grows and is determined in a proximodistal fashion in response to cell-cell interactions [15].

Table **1** Congenital limb anomalies and their molecular basis

Classification	*Disorder*	*Genomic locus*	*Gene*
Polydactylies			
Preaxial			
– *isolated*	PPD I		
	PPD II/TPT	7q36	C7orf2/ZRS
	PPD III		
	PPD IV	7p13	GLI3
– *associated*	Carpenter		
	OFD II		
	SRPS II		
	Townes-Brocks	16q12.1	SALL1
Postaxial			
– *isolated*	PAPA1	7p13	GLI3
	PAPA2	13q21-q32	
	PAPA3	19p13.2-p13.1	
	PAPB	7p13	GLI3
– *associated*	Ellis van Creveld	4p16	EVC
	Smith-Lemli-Opitz	11q12-q13	DHCR7
	McKusick-Kaufmann	20p12	BBS6
	SRPS I		
	OFD III		
	BBS1	11q13	BBS1
	BBS2	16q21	BBS2
	BBS3	3p13	
	BBS4	15q22	BBS4
	BBS5	2q31	
	BBS7	4q27	BBS7
	Meckel-Gruber	17q21q24	
	Jeune	15q13	
– *central*	Pallister-Hall	7p13	GLI3
– *pre- and postaxial*	Greig	7p13	GLI3
Reduction anomalies			
– *mesoaxial isolated*	SHFM1	7q21.3-q22.1	DLX5/6
	SHFM2	Xq26	
	SHFM3	10q24–35	
	SHFM4	3q27	P63
	SHFM5	2q31	
	Acheiropodia	7q36	C7orf2
– *associated*	EEC, AEC, LMS, ADULT	3q27	P63
– *preaxial*	Nager		
	LADD		
	Fanconi anemia		
	TAR		
	Baller-Gerold		
	Holt-Oram	12q24.1	TBX5
	HFGS	7p14.2-p15	HOXA13
– *postaxial*	Cornelia de Lange	3q26	
	FFU		
	UMS	12q24.1	TBX3
Brachydactylies			
– *isolated*	BDA1	2q35	IHH
	BDA2	4q23-q24	BMPR1B
	BDB	9q34	ROR2
	BDC	20q11.2	CDMP1
	BDD	2q31	HOXD13
	BDE	2q31	HOXD13
– *associated*	Robinow	9q34	ROR2
	Grebe	20q11.2	CDMP1
	Rubinstein-Taybi	16p13.3	CREBBP
– *associated BDE*	Turner	45X0	
	Albright HO	20q13.2	GNAS
	Albright-like HO	2q37	
	BCNS	9q22.3	PTC
	HTNB	12 p	
– *Cone shape epiphysis*	TRPS	8q24.12	TRPS
	Acrodysostosis		
Syndactylies			
– *isolated*	SD1	2q34-q36	
	SD2/SPD	2q31-q32	HOXD13
	SD3/ODDD	6q22-q24	CX43
– *associated*	Apert	10q26	FGFR2
	Saethre-Chotzen	7p21	TWIST
	Pfeiffer	8p11.2	FGFR1
		10q26	FGFR2
	F-syndrome	2q36	
	Fraser	4q21	FRAS1

Abbreviations: ADULT – Acro-dermato-ungual-lacrimal-tooth syndrome; AEC – Ankyloblepharon-ectodermal dysplasia defects-cleft lip/palate; BBS – Bardet-Biedl syndrome; BCNS – Basal cell nevus syndrome; BDA – Brachydactyly Type A; BDB – Brachydactyly Type B; BDC – Brachydactyly Type C; BDD – Brachydactyly Type D; BDE – Brachydactyly Type E; EEC – Ectrodactyly ectodermal dysplasia, and cleft lip/palate syndrome; FFU – Femur-Fibula-Ulna syndrome; HFGS – Hand-foot-genital syndrome; HTNB – Hypertension with brachydactyly; LADD – Lacrimoauriculodentodigital syndrome; LMS – Limb-mammary syndrome; OFD – Orofaciodigital syndrome; PAPA – Postaxial polydactyly; PPD – Preaxial Polydactyly; SD – Syndactyly; SHFM – Split-hand/-foot malformation; SRPS – Short rib-polydactyly syndrome; UMS – Ulna-mammary syndrome; TAR – Thrombocytopenia-absent radius syndrome; TPT – Triphalangeal thumb, TRPS – Trichrhinophalangeal syndrome

Multiple genes are involved in order to control growth and polarization of the limb and the subsequent formation of skeletal elements. *Homeobox (Hox-)* genes of the 5′ region of the A- and D-clusters on human chromosome 7 and chromosome 2, respectively, are expressed in the developing limb bud and are involved in the determination of shape and identity of skeletal elements. *Hox* genes influence limb patterning, mesenchymal condensation and the rate and timing of cartilage proliferation and differentiation. The specification of fore and hind limb identity is mediated by certain *T-box (Tbx)* genes [1]. *Tbx* genes are related to the murine transcription factor *T*. In addition, signals deriving from the interdigital mesenchyme between the digital rays including *Bone morphogenetic proteins (Bmp)* and their antagonist *Noggin (Nog)* have recently been shown to influence the number of phalanges. These findings underscore the impact of local signaling during development of cartilage templates.

Classification of Congenital Hand Anomalies

Congenital anomalies in the embryo occur as malformations, deformations, disruptions and dysplasias. Malformations are defined as an abnormal intrinsic embryonic development due to a genetic defect as a consequence of exposure to teratogens or ma-

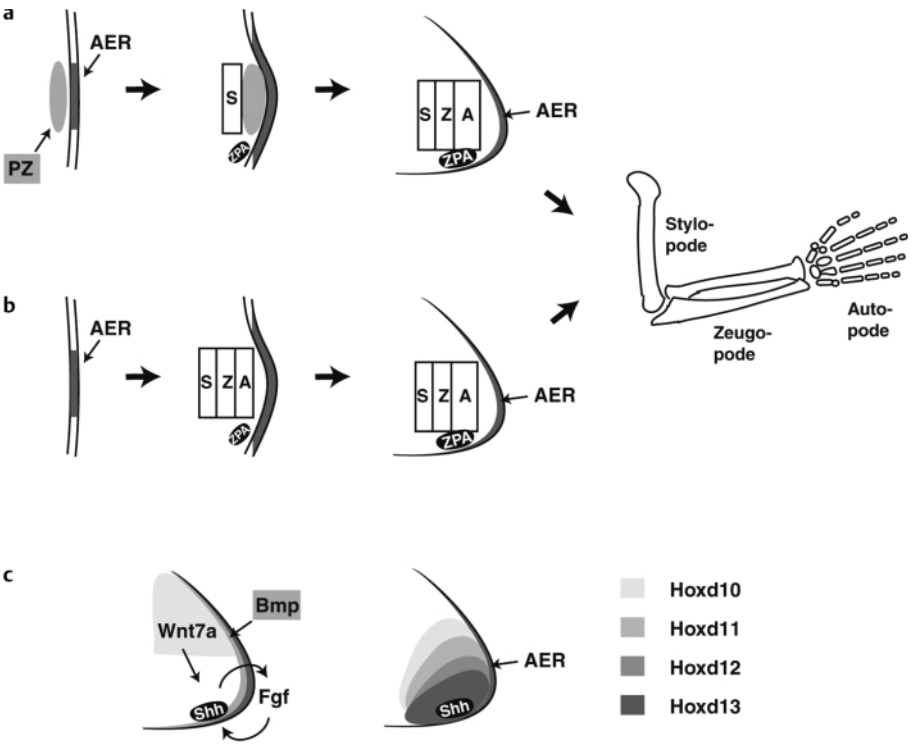

Fig. **2 a** to **c** Limb development and patterning. The limb buds form at the lateral flank of the embryo by proliferation of cells of the lateral plate. Signaling centers are important for outgrowth and patterning of the limb bud. Proximodistal outgrowth of the limb along the axis is controlled by the apical ectodermal ridge (AER). Anteroposterior patterning of the limb is established by the zone of polarizing activity (ZPA). **a** The progress zone model suggests that mesodermal cells are specified as they leave the so-called progress zone (PZ), indicating that the first cells leaving the PZ will form the stylopode (S), followed by the zeugopode (Z) and the autopode (A). **b** The early specification model proposes that in the early limb bud cells are specified to form all three compartments (S, Z, A), then expand and are finally determined to form the distinct skeletal elements. **c** Gene expression patterns corresponding to signaling centers direct the three-dimensional outgrowth and patterning of the limb bud. *Fgfs* mediate proximodistal outgrowth and mutually influence *Shh*, a key factor for establishment of anteroposterior polarity. *Wnt7a* regulates the dorsal identity of the limb. *HoxD10-13* genes are expressed in the developing limb bud and are involved in the determination of shape and identity of skeletal elements.

ternal metabolic disease. Deformations result from external mechanical influences, for example an oligohydramnion leading to joint contractures or facial compression of the fetus. Disruptions refer to the pathology of organs or tissues that have initially developed normally but have been disrupted in their development by external influences such as infections or ischemia. Disruptions normally affect different types of tissues not restricted to a developmental pattern. Dysplasias are characterized by tissue specific defects, indicating that the initial patterning process is normal, while organogenesis is disturbed, for example harmatomas.

In this review, we focus on limb malformations with known molecular and genetic causes. The large spectrum of remaining limb malformations are likely to have a wide range of different etiologies, ranging from external factors, polygenic mutations to single gene mutations with low penetrance. We present an overview of congenital limb malformations, classified on an anatomic and genetic basis, taking recent molecular and developmental insights into account (see Fig. **1** and Table **1**). For a human chromosome karyogram with loci of limb malformations see Fig. **4**.

I. Polydactyly

Polydactyly refers to the occurrence of supernumerary digits or parts of them, ranging from complete duplication of a limb or a part of a limb to duplications of single digits. Isolated polydactyly is commonly found as preaxial or postaxial polydactyly. Polydactyly of central rays is rare. Higher degrees of polydactylies include defects/agenesis of hands/feets or mirror image duplications.

In syndromic association, postaxial polydactylies are found in Ellis-van-Creveld syndrome (EVC; MIM 225500, for explanation of MIM numbers see Glossary), Smith-Lemli-Opitz syndrome (SLOS, MIM 270400), McKusick-Kaufmann syndrome (MKKS; MIM 236700), short rib-polydactyly syndrome I (SRPS I; MIM 263530), orofaciodigital syndrome III (OFD III, MIM 258850), Bardet-Biedl syndrome (BBS; MIM 209900), Meckel-Gruber syndrome (MKS1; MIM 249000) and many others. Preaxial polydactyly occurs in acrocephalopolysyndactyly syndrome (ACPS II; MIM 201000), orofaciodigital syndrome II (OFD II, MIM 252100), short rib-polydactyly syndrome II (SRPS II; MIM 263520) and Townes-Brocks syndrome (TBS; MIM 107480). Greig cephalosynpolydactyly (GCPS; MIM 175700) presents with both pre- and postaxial polydactyly, whereas Pallister-Hall syndrome (PHS; MIM 146510) is characterized by central or postaxial polydactyly. On the molecular level, many forms of polydactyly have been shown to be more or less directly linked to the *Shh* signal transduction pathway, which plays a major role in anteroposterior patterning of the limb.

Shh signal transduction pathway

Shh belongs to the Hedgehog family of intercellular signaling proteins and functions as a key morphogen in a dose dependent manner to induce cell fates and play a central role in growth, patterning and morphogenesis of a variety of tissues [24]. Many of the signaling mechanisms for Hh have been identified and extensively studied in Drosophila and are essentially conserved among flies, mice and other species (Fig. **3**). After synthesis, the N-terminal part of Hh is autocatalytically cleaved, secreted and processed in an active form by covalent coupling of cholesterol to the C terminus of Hh-N. Hh exerts its function by binding to its

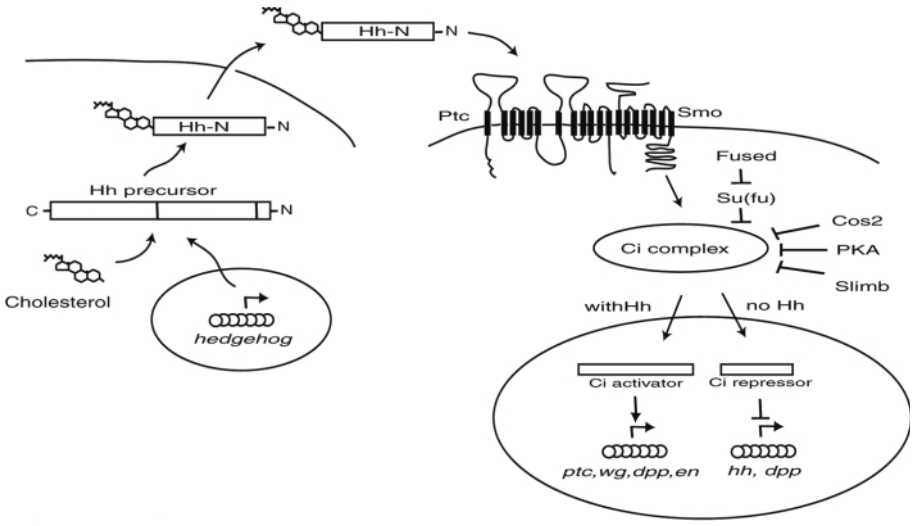

Fig. **3** Hedgehog pathway (adapted from McMahon [37]). Hh is autocatalytically cleaved, secreted and processed in an active form by cholesterol. Hh binds to the Ptc receptor which inhibits downstream signaling by forming a complex with Smo. When Shh binds to Ptc, Smo is released from Ptc inhibition and leads to stabilization of the transcription factor Ci, which activates downstream Hh targets. Ci acts as an activator and repressor. In the absence of Hh, Ci protein is cleaved, converting it to a repressor form. Hh activity leads to stabilization of full length Ci, resulting in transcription of Hh targets.

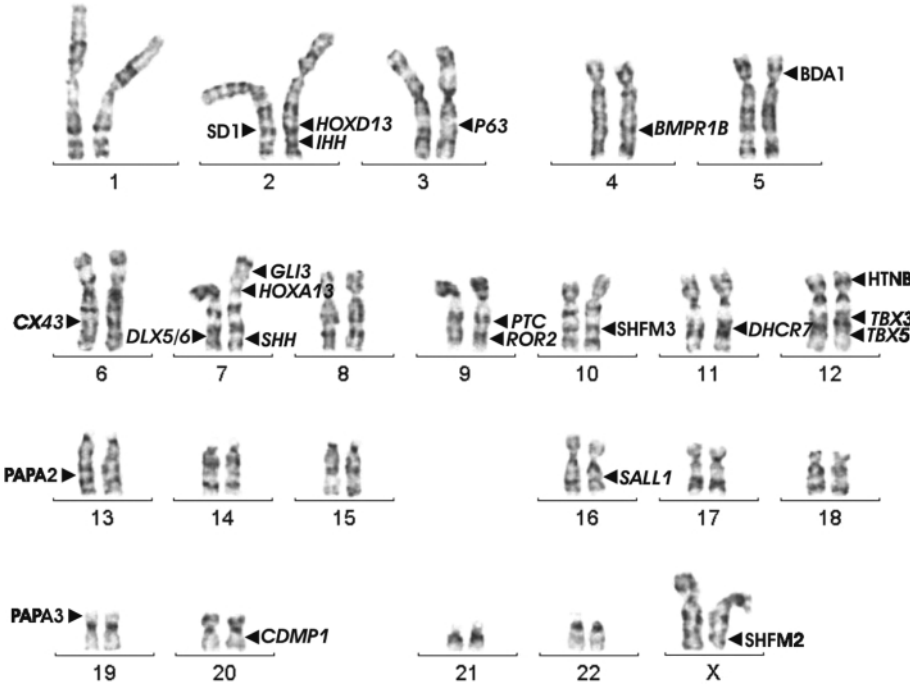

Fig. **4** Chromosomal karyogram of a normal female with loci of genetic limb malformations. Genetic loci and genes (in italics) of traits with limb malformations are indicated with an arrowhead. Due to space limitations, loci and genes are not indicated on the second parental chromosome.

receptor Patched (Ptc). Ptc normally forms a complex with the seven span transmembrane protein Smoothened (Smo), thereby inhibiting downstream signaling. As a result of Shh binding, Ptc releases Smo resulting in stabilization of the transcription factor cubitus interruptus (Ci). Ci represents a zinc finger transcription factor acting as an activator of Hh target genes in the presence of Hh or a repressor in the absence of Hh. During mouse embryonic development, *Shh* is expressed in the notochord, the floorplate of the neural tube, the gut and the zone of polarizing activity (ZPA), where it mediates anteroposterior limb patterning.

Preaxial polydactyly

Preaxial polydactyly is a genetically heterogeneous group. The following classification has been suggested: 1) Thumb polydactyly (PPD1; MIM 174400) is characterized by duplications of one or more skeletal elements of a biphalangeal thumb. 2) Polydactyly of a triphalangeal thumb (PPD2/TPT; MIM 174500; Fig. **5**) shows a triphalangeal opposable thumb with facultatively duplicated phalanges. 3) Polydactyly of the index finger is characterized by replacement of the thumb by one or two triphalangeal digits with an accessory complete or partial metacarpal (PPD3; MIM 174600). 4) In preaxial polydactyly type 4 (PPD4; MIM 174700), preaxial polydactyly and facultative syndactyly of various degrees are present involving the third and fourth finger in the hand and the second and third toe in the foot.

It is conceivable that a large proportion of these different forms of preaxial polydactyly represents a variable disease spectrum involving the *Shh-Ptc-Gli* pathway. TPT and PPD3 occur in the same families and have been mapped to the *Shh* locus on chromosome 7q36 ([57]. In addition, mutations in a *Shh* regulatory element have been described for PPD1 [33]. PPD4 has been

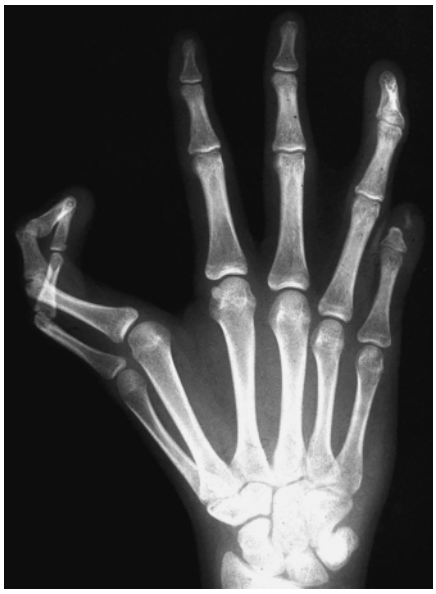

Fig. **5** Right hand of an individual with preaxial polydactyly of a triphalangeal thumb (PPD2/TPT). Note that the metacarpal and the phalanges of the thumb are duplicated; both thumbs have three phalanges. In addition, in the fifth digit the middle phalanx is absent and the distal phalanx is hypoplastic.

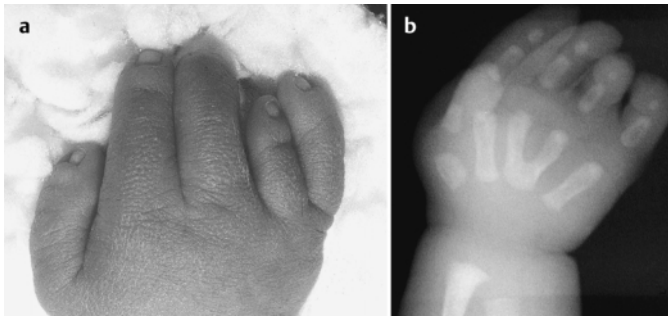

Fig. **6 a** and **b** Pallister-Hall syndrome. **a** Right hand of female newborn individual. The fingers are plump and shortened with emphasis on distal phalanges including nail dysplasia. The fourth finger is extremely shortened and hypoplastic. **b** The corresponding radiograph of the right hand shows metacarpal synostosis. The middle phalanges are absent in all digits and the distal phalanges are hypoplastic.

shown to be caused by heterozygous frameshift mutations in *Gli3* [46]. For an explanation of mutation terminology see Glossary. These findings underline the key function of the *Shh-Ptc-Gli* pathway in the development of various forms and the variability of preaxial polydactyly.

Postaxial polydactyly

Postaxial polydactyly is characterized by supernumerary digits on the ulnar or fibular side of the hands or feet. Two phenotypically and possibly genetically different varieties exist: Postaxial polydactyly type A is inherited as a dominant trait with marked penetrance and is characterized by a rather well formed digit articulating with the fifth or an extra metacarpal. Three different loci exist for postaxial polydactyly on chromosomes 7p13 (PAPA1; MIM 174200), 13q21-q32 (PAPA2; MIM 602085) and 19p13.2-p13.1 (PAPA3; MIM 607324). Heterozygous frameshift mutations in *Gli3* have been demonstrated for autosomal dominant PAPA1 [47]. In postaxial polydactyly type B (PAPB; MIM 174200) exhibiting a more complicated trait, the extra digit is not well formed and frequently appears in the form of a skin tag (pedunculated postminimus). In postaxial polydactyly A/B, some patients present with polydactyly type A and polydactyly type B, indicating phenotypic variability of the trait. Heterozygous *Gli3* frameshift mutations have been described for PAPA/B [46], indicating a more complex genotype-phenotype correlation than previously expected.

Greig cephalopolysyndactyly

Greig cephalopolysyndactyly (GCPS; MIM 175700) is an autosomal dominant disorder affecting limb and craniofacial development. GCPS is characterized by 1) malformations of the hands (broad and sometimes duplicated thumbs, postaxial polydactyly, pedunculated postminimus, syndactyly of the third and fourth finger) and feet (preaxial polydactyly, broad toes, occasionally postaxial polydactyly, syndactyly particularly of the first to third toes) and 2) craniofacial abnormalities including macrocephaly, frontal bossing, hypertelorism, a broad base of the nose, occasionally associated with late closure of cranial sutures, mild mental retardation and a mild degree hydrocephalus. GCPS is caused by deletions, truncations or point mutations before the zinc finger region of the *Gli3* gene, which is likely to lead to haploinsufficiency of *GLI3* [58].

Pallister-Hall syndrome

Pallister-Hall syndrome (PHS; MIM 146510) is characterized by 1) malformations of the central nervous system, including hypothalamic harmatomas, hypopituitarism and occasional underdevelopment of the thyroid, 2) craniofacial anomalies including a flat midface with midline capillary hemangioma, anteverted nares, external ear anomalies, micrognathia, multiple frenuli between the alveolar ridge and buccal mucosa, 3) limb malformations with nail dysplasia, variable degrees of syndactyly and polydactyly (central and postaxial) involving hands and feet, shortened and distally placed fourth metacarpal and distal shortening of limbs (Fig. **6**) and 4) anal defects including imperforate anus with variable degrees of rectal atresia. PHS has been shown to be caused by *Gli3* frameshift mutations located after the zinc finger domain. If stable, these proteins are able to bind DNA, leading to disturbances of the Hh pathway.

Townes-Brocks syndrome

Townes-Brocks syndrome (TBS; MIM 107480) is an autosomal dominant disorder with marked phenotypic variability. TBS includes 1) craniofacial anomalies consisting of auricular anomalies ranging from large ears to poorly formed ears with microtia and preauricular tags and variable degrees of facial hypoplasia resembling hemifacial microsomia (Goldenhar syndrome; MIM 141400), 2) limb anomalies including hypoplastic, broad, triphalangeal and supernumerary thumbs, fusion of carpal and metatarsal bones, 3) imperforate anus, anterior placement and stenosis of the anus, 4) renal anomalies including renal hypoplasia. TBS is caused by heterozygous nonsense mutations, insertions or deletions in *SALL1,* a homolog of the Drosophila homeotic gene *spalt (Sal)* that encodes a putative C2H2 zinc finger transcription factor [29]. The mutations are predicted to lead to haploinsufficiency as they result in a prematurely terminated SALL1 protein lacking all putative DNA binding domains. In the fish, *Spalt* has been demonstrated to be a *Hedgehog* target gene and is expressed in the *Hh* signaling centers of the embryo [31].

Smith-Lemli-Opitz syndrome

Smith-Lemli-Opitz syndrome (SLOS, MIM 270400) is a relatively frequent autosomal recessive malformation syndrome (incidence 1 : 20 000 live births) that is characterized by 1) craniofacial abnormalities including microcephaly, slanted or low set ears, ptosis, inner epicanthal folds and micrognathia and 2) limb malformations, such as simian crease, high frequency of digital whorl dermal ridge patterning, syndactyly of the second and third toe, asymmetrically short fingers or pre- or postaxial polydactyly. Other limb malformations include a short thumb with a short first metacarpal, ectrodactyly, monodactyly, oligodactyly, radial agenesis, various forms of syndactyly and rhizomelic or mesomelic limb shortening. In addition, patients with SLOS exhibit 3) genitourinary abnormalities and 4) severe mental retardation and different morphogenic defects of the central nervous system with seizures. Occasionally, cardiovascular defects, renal and gastrointestinal anomalies are present.

Smith-Lemli-Opitz syndrome (SLOS) is caused by mutations in the *DHCR7* gene, leading to deficient activity of 7-dehydrocholesterol reductase (DHCR7), the final enzyme of the cholesterol biosynthetic pathway, resulting in low cholesterol and high concentrations of its direct precursor 7-dehydrocholesterol in plasma [59]. The mutations in *DHCR7* lead to decreased protein stability with a genotype-phenotype correlation existing between clinical severity scores and mutation classes [60].

II. Absence Deformities

Terminal transverse defects

Terminal transverse defects refer to absence or hypoplasia of distal structures of the limb with more or less normal proximal structures. Ectrodactyly (Greek: ektroma-abortion, daktylos-finger) describes a number of genetic and non-genetic malformations with absence deformities of more distal segments such as certain phalanges (aphalangia), fingers (adactylia) or the entire hand (acheiria). More severe degrees of terminal transverse defects are hemimelia and amelia. Most of these cases occur sporadically with one hand involved and the feet not affected. The majority of these malformations are considered to be non-genetic, i.e. caused by external factors such as amniotic bands or vascular disruptions. However, inheritance of unilateral hypoplasia has been described. It is conceivable that many conditions currently considered to be non-genetic are in fact polygenic, i.e. they occur only if several (three or more) mutated genes are present or they are more likely to be caused by single gene mutations with low penetrance. If the threshold of a mutation to produce a certain phenotype is low, random factors may determine the outcome. This may then result in only one limb affected and no obvious pattern of inheritance.

Split-hand/-foot malformation

Split-hand/-foot malformation (SHFM) belongs to the group of ectrodactylies and may occur as an isolated trait or in combination with other defects. Phenotypically, two types of SHFM are recognized: 1) deficiency of the central ray with a cleft and 2) monodactyly with absence of radial rays and no cleft with only the fifth digit remaining (Fig. **7**). There is reduced penetrance and considerable phenotypic variability in SHFM.

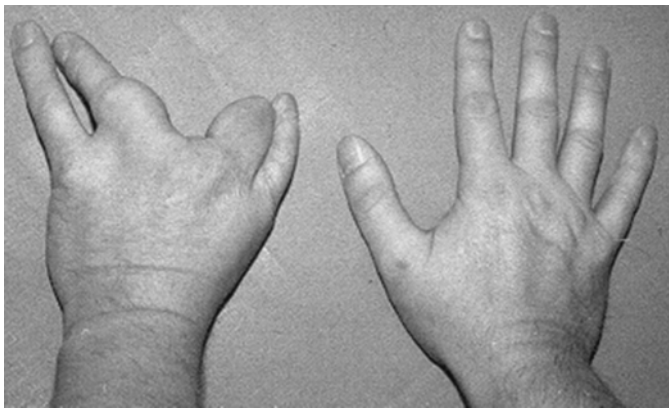

Fig. **7** Split-hand/-foot malformation. Patient with a unilateral split-hand malformation showing a severe central reduction defect of the left hand. There is aplasia of the third finger, hypoplasia of the first, second and fourth fingers and nail hypoplasia in all affected digits. The fingers of the right hand are normal.

Four genetic loci for SHFM have been described to date: SHFM1 (MIM 183600) maps to chromosome 7q21.3-q22.1 [48], harbouring the *Distalless Homeobox* genes *DLX5* and *DLX6* that are expressed in the limb and represent candidate genes for SHFM1. SHFM2 (MIM 313350) is located on chromosome Xq26 [17]. SHFM3 (MIM 600095) maps to chromosome 10q25, a region corresponding to mouse chromosome 19, where the mouse mutant *dactylaplasia (Dac)* is located [27]. *Dac* is a member of the *F-box/WD40* gene and the expression of *Dac* is regulated by a modifier locus *(mdac)* on mouse chromosome 13 [27]. SHFM4 (MIM 605289) is caused by mutations in *P63* [11]. Interestingly, *P63* mutations have been found in four other phenotypically related diseases including ectrodactyly ectodermal dysplasia and cleft lip/palate syndrome (EEC1; MIM 604292; [11]), ankyloblepharon ectodermal defects-cleft lip/palate (AEC; MIM 106260; [36]), limb-mammary syndrome (LMS; MIM 603543) and acro-dermato-ungual-lacrimal-tooth syndrome (ADULT; MIM 103285; [16]). The mutations are predicted to lead to dominant negative and gain of function mechanisms rather than loss of function.

Radial ray defects

Radial ray defects occur as an isolated malformation or in a number of syndromes. Syndromic radial defects comprise a large group of diseases and can be classified into radial defects with 1) orofacial malformations such as in Nager syndrome (AFD1; MIM 154400) or lacrimoauriculodentodigital syndrome (LADD; MIM 149730), 2) aplastic anemias such as Fanconi anemia (FA; MIM 227650), thrombocytopenia and absent radius (TAR; MIM 274000) syndrome, 3) craniosynostosis-radial dysplasia syndrome (BGS; MIM 218600), 4) Holt-Oram syndrome (HOS; MIM 142900), 5) hand-foot-genital syndrome (HFGS; MIM 140000) and others. HOS and HFGS and their molecular pathogenesis are discussed in more detail.

Holt-Oram syndrome

Holt-Oram syndrome (HOS; MIM 142900) is a dominantly inherited condition characterized by 1) skeletal and 2) cardiovascular abnormalities [7]. The limb phenotype is variable and involves a wide spectrum including defects in the upper limb and shoulder girdle ranging from thumb hypoplasia to phocomelia sometimes

with asymmetric presentation. The syndrome presents with finger-like hypoplastic to absent or triphalangeal thumbs with syndactyly, hypoplasia to absence of the first metacarpal and radius, defects of the ulna, humerus, clavicle, scapula and sternum. Cardiovascular anomalies occur in 95 percent of the patients and include auricular septal defects, sometimes with arrhythmia and ventricular septal defects.

HOS is caused by mutations in *TBX5,* a member of the *Tbx* gene family, predicted to lead to haploinsufficiency of the gene [34]. *Tbx* genes encode a group of transcription factors characterized by a highly conserved DNA-binding motif (T-box). Tbx genes play key roles during organogenesis and pattern formation in vertebrate and invertebrate embryos [42]. They are directly involved in both the processes of limb initiation at early stages and in the specification of limb identity at later stages [1].

Hand-foot-genital syndrome

Hand-foot-genital syndrome (HFGS; MIM 140000) is a dominantly inherited condition affecting the distal limbs and the lower genitourinary tract. Individuals with HFGS display short hands and feet with short proximally placed thumbs, hypoplastic thenar eminences and short tibially deviated greater toes with small pointed first distal phalanges and short first metacarpals/metatarsals. Moreover, delayed ossification, fusion and deformity of carpal and tarsal bones is present. The limb abnormalities are fully penetrant, bilateral and symmetrical with little variation in severity. Urogenital tract malformations are phenotypically variable with incomplete penetrance and include hypospadias, micropenis in males and Müllerian duct fusion defects, leading to a double uterus or double cervix in females. In some patients, ectopic ureteric orifices, vesico-ureteric reflux and pelvi-ureteric junction obstruction has been described. HFGS is caused by nonsense or frameshift mutations in *HOXA13* leading to a truncated protein or polyalanine tract expansions similar to those in *HOXD13* that cause synpolydactyly [38].

Ulnar defects

Ulnar defects occur as isolated disorders or as parts of syndromes. Syndrome-related ulnar defects include ulnar defects 1) with orofacial malformations, 2) Cornelia de Lange syndrome (CDL1; MIM 122470), 3) Femur-Fibula-Ulna syndrome (FFU; MIM 228200) and 4) Ulnar-mammary syndrome (UMS; MIM 181450) which is presented in more detail.

Ulnar-mammary syndrome

Ulnar-mammary syndrome (UMS; MIM 181450) is a rare autosomal dominant disorder associated with a wide range of phenotypes. UMS is characterized by 1) hypoplasia of the ulnar side of the upper extremity or ulnar longitudinal deficiency ranging from symphalangism, absence or duplication of the fifth digit to absence of the ulna and reduction of the humerus, 2) hypoplasia or aplasia of mammary glands and nipples and of apocrine glands in both sexes and 3) underdeveloped external genitalia and delayed puberty in males and uterine anomalies in females. UMS is caused by haploinsufficiency of the *TBX3* gene [6], coding for another T-box transcriptional factor. *Tbx3* is widely expressed in a variety of tissues including forelimbs and hindlimbs, epithelium of the mammary gland, the genital tubercle and the uterus. Mice with loss of *Tbx3* function show deficiency of mammary gland induction and forelimb and hindlimb abnormalities [13].

III. Defects in Dorso-Ventral Patterning

Nail-patella syndrome

Nail-patella syndrome (NPS; MIM 161200) is an autosomal dominant disorder characterized by developmental defects affecting the nails, skeleton, kidneys and eyes. Onychoskeletal features include hypoplastic or absent nails and patellae, patella dislocations, elbow abnormalities and iliac horns on radiographs. Renal manifestations include a defect in the glomerular basal membrane. In the eye, open-angle glaucoma may present as a feature. NPS has a high penetrance and phenotypic variability. Additional skeletal defects have been noted such as foot and ankle abnormalities including clubfoot, dislocation or subluxation of the hip or of the radial head.

Loss of function mutations in the LIM-homeodomain protein *LMX1B* lead to NPS [14]. *Lmx1b* is involved in determination of dorso-ventral patterning of the limb. *Lmx1b* is induced by dorsal ectoderm expression of *Wnt7a* and repressed by ventral ectoderm expression of *En1* [28]. *Lmx1b* knock-out mice show complete absence of the dorsal side of the autopode including nails and patellae indicating that *Lmx1b* is needed for the specification of dorsal limb fate [12]. Although the murine phenotype is more pronounced than in the NPS patient, the parallels are obvious.

IV. Brachydactyly

Brachydactyly may occur as an isolated trait or in association with other malformations. According to their patterns of skeletal involvement isolated brachydactylies have been classified on an anatomic and genetic basis into five groups, A to E, including three subgroups A1 to A3 [8]. Isolated brachydactylies usually occur as autosomal dominant traits and show a high degree of phenotypic variability.

Brachydactyly type A1, A2, A3

The type A brachydactylies according to the Bell classification are characterized by hypoplasia/aplasia of the middle phalanges. In brachydactyly type A1 (BDA1; MIM 112500) all middle phalanges are affected and shortened or fused with terminal phalanges. BDA1 was the first trait in man to be interpreted in Mendelian terms [19]. For BDA1 heterozygous missense mutations have been identified in the amino-terminal signaling domain of *Indian hedgehog (IHH)* [21]. A second locus for BDA1 has been mapped to chromosome 5p13.3-p13.2 [3].

BDA2 (BDA2; MIM 112600) is characterized by a triangular shaped middle phalanx with or without radial deviation and clinodactyly of the fifth finger and can be caused by missense mutations in *BMPR1 B* [32]. Brachydactyly type A3 (BDA3; MIM 112700) is characterized by shortening of the middle phalanx of the little finger but no genetic location has been found so far.

Brachydactyly type B

Brachydactyly type B (BDB; MIM 113000) is an autosomal dominant disorder characterized by shortening/hypoplasia of the distal phalanges, nail dysplasia, hypoplasia of middle phalanges and variable degrees of distal and proximal symphalangism (Fig. **8**). Additional findings are broad thumbs with or without distal duplication/clefts and syndactyly. Brachydactyly type B is caused by mutations within two distinct regions of the receptor tyrosine kinase *ROR2* [41,50] that are predicted to result in truncation of the receptor before or after the tyrosine kinase (Tk) domain. Mutations located after the Tk domain are associated with a more severe phenotype. *ROR2* is expressed in the limb bud, throughout the nervous, cardiovascular, respiratory, digestive, urogenital and skeletal systems [2,52].

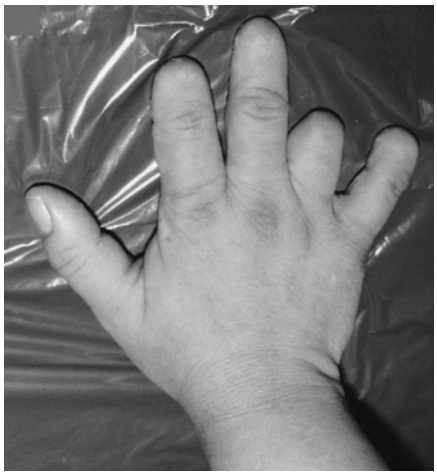

Fig. **8** Brachydactyly type B. Right hand of a patient exhibiting shortening/absence of distal and middle phalanges of the second to fifth fingers and aplasia of nails.

Brachydactyly type C

Brachydactyly type C (BDC; MIM 113100) is characterized by brachymesophalangy of the second, third and fifth fingers, hyperphalangy of the second and/or third finger and shortening of the first metacarpal. Generally, the fourth finger is not affected and it is therefore the longest digit in BDC. Accompanying skeletal or non-skeletal features such as short stature, talipes valgus, Perthes disease, hip dysplasia or spine deformities have also been described. Heterozygous frameshift or nonsense mutations affecting the *cartilage-derived morphogenetic protein-1 (CDMP1)* also called *GDF5* and leading to a loss of function are responsible for dominant BDC [45]. A recessive form of BDC has recently been described, caused by a homozygous missense mutation located within the *CDMP1* prodomain [51]. *Gdf5,* the murine homologue of *CDMP1,* is expressed along early cartilage condensations in the joint interzone and in the perichondrium during limb development. *Gdf5* has been shown to be a key regulator during skeletal and joint development. Treatment of embryonic mouse and chicken limbs with recombinant Gdf5 results in an altered pattern of bones and articulations in the digits [20].

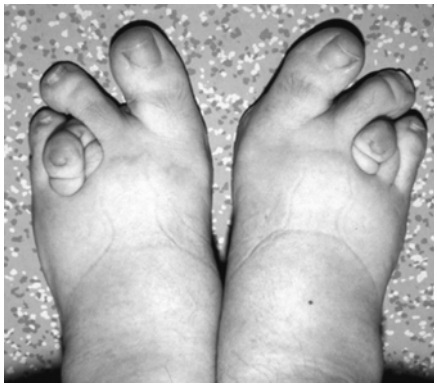

Fig. **9** Brachydactyly type E in a patient with Turner (45, X0) syndrome. Feet exhibiting bilateral shortening of the third and fourth toes due to shortening of the third and fourth metatarsals. The nails exhibit a convex shape.

Brachydactyly type D

In Brachydactyly type D (BDD; MIM 113200) the distal phalanx of the thumb is shortened to various degrees. Recently, a mutation in *HOXD13* has been described in an individual with BDD [26].

Brachydactyly type E

Isolated Brachydactyly type E (BDE; MIM 113300) is inherited as a dominant trait and is characterized by variable shortening of metacarpals. In one individual with BDE a mutation in *HOXD13* has been described [26]. The majority of cases with BDE occur in association with syndromes such as Turner syndrome (Fig. **9**), Albright hereditary osteodystrophy (AHO; MIM 103580), an AHO-like disorder on chromosome 2q37 (MIM 600430), basal cell nevus syndrome (BCNS; MIM 109400), hypertension-brachydactyly syndrome (HTNB; MIM 112410) and others.

Brachydactyly as part of syndromes

Different forms of brachydactyly, some of them resembling isolated brachydactyly traits, occur as part of a syndrome in a number of bone dysplasias such as achondroplasia (ACH; MIM 100800), Weill-Marchesani syndrome (MIM 277600), Robinow syndrome (RS; MIM 268310), Grebe syndrome (CG; MIM 200700) and others. Short thumbs are a characteristic feature of Robinow syndrome (RS; MIM 268310) and Rubinstein-Taybi syndrome (MIM 180849) and BDB is present in Sorsby syndrome (MIM 136900).

Other types of syndromal forms of brachydactyly include brachytelephalangy, characterized by shortening of distal phalanges and cone shape epiphyses. Cone shape epiphyses result from ossification anomalies due to invagination of the epiphysis and physis into the metaphysis leading to brachydactyly as found in trichorhinophalangeal syndrome (TRPS1; MIM 190350), Langer-Giedion syndrome (LGS; MIM 150230) and acrodysostosis (MIM 101800). Some of the mentioned syndromes with brachydactyly are discussed below in more detail.

Acromesomelic chondrodysplasia Grebe, Hunter Thompson, Du Pan

Chondrodysplasia Grebe (CG; MIM 200700), Hunter-Thompson (HT; MIM 201250) and Du Pan syndrome (DP; MIM 228900) are autosomal recessive inherited chondrodysplasias. Of the three, CG is the most severe chondrodysplasia and is characterized by shortening of upper and lower limb skeletal elements in a proximodistal fashion with absence of carpal bones and severe reduction of metacarpals and phalanges. Patients with HT are not as severely affected as in CG. DP is characterized by fibular hypoplasia with complex brachydactyly. All three forms of chondrodysplasia are caused by homozygous mutations of *CDMP1* in the active domain of *CDMP1* [18,55,56]. The mouse mutant *brachypodism (bp),* caused by a loss of function mutation of *Gdf5,* exhib-

its similar cartilage and joint formation and serves as a model for CG and HT [53].

Hypertension with brachydactyly

Hypertension with brachydactyly, also referred to as Bilginturan syndrome (HTNB; MIM 112410), is characterized by shortening of both phalanges and metacarpals associated with hypertension. Affected adults present with short height and blood pressure increasing with age. In a large Turkish kindred the disease locus was mapped to chromosome 12p12.2-p11.2. [49]. Interestingly, a deletion at 12p in a Japanese child with brachydactyly but without hypertension overlaps the assigned locus suggesting that possibly two different genes are involved for brachydactyly and hypertension [5].

V. Segmentation Disorders

Proximal symphalangism

Proximal symphalangism (SYM1; MIM 185800) is an autosomal dominant malformation characterized by absent proximal interphalangeal joints in some fingers and fusion of talus and navicular bones in the foot. In a subset of patients, fusion of ossicles of the middle ear occur, resulting in conductive hearing loss. Heterozygous missense mutations in the Bmp antagonist *Noggin (NOG)* lead to SYM1 and multiple synostoses syndrome (SYNS1; OMIM 186500) indicating that SYM1 and SYNS1 are allelic [22]. During development, *Nog* is expressed in multiple organs including cartilage; *Nog* modulates activity of bone morphogenetic proteins (Bmps) [10].

VI. Syndactyly

Syndactyly is the most common congenital malformation of the hand in Europe and North America. It is characterized by the apparent fusion of soft tissues of the fingers and toes with or without bony fusion and is caused by the absence of programmed cell death in the interdigital mesenchyme. Syndactyly is referred to as partial when it involves proximal segments of the digits and as complete when it extends to the finger tips. Syndactylies may also present in association with constriction rings at the finger tips as acrosyndactyly which involves soft tissue with occasional fenestration and is not likely to have a genetic basis.

Syndactyly may occur as an isolated malformation or as part of a syndrome. The syndromal forms comprise 1) Poland syndrome (MIM 173800) associated with symbrachydactyly, 2) several types of acrocephalosyndactyly (in Apert syndrome; MIM 101200), Saethre-Chotzen syndrome (MIM 101400), Waardenburg syndrome type III (MIM 148820), Pfeiffer syndrome (MIM 101600), Summitt syndrome (MIM 272350), 3) the F-syndrome (MIM 102510) and 4) Fraser syndrome (MIM 219900). Isolated syndactylies have been classified into five groups with each syndactyly type affecting different interdigital spaces as presented below [54].

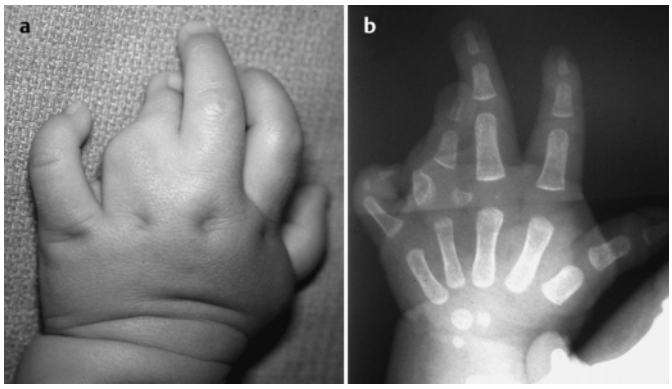

Fig. **10 a** and **b** Synpolydactyly. **a** Left hand of an individual with synpolydactyly showing soft tissue syndactyly of the third and fourth fingers and brachydactyly of all fingers. The fourth digit is severely shortened and laterally deviated. There is clinodactyly of the fifth finger. **b** The corresponding hand radiograph shows that the phalanges of all digits are shortened. The proximal phalanx of the fourth digit exhibits a delta shape and there is an additional rudimentary proximal phalanx of the fourth digit in the syndactylous web. Shortening of the middle phalanx of the fifth digit leads to clinodactyly.

Syndactyly type I (zygodactyly)

Syndactyly type I (SD1; MIM 185900) is an autosomal dominant limb malformation and the most common type of syndactyly. It is characterized by syndactyly of the third and fourth fingers and the second and third toes. Syndactyly may be complete or partial; occasionally other digits may be affected. The malformation is not always bilateral or symmetric and in some cases may affect either hands or feet. A special form of SD1 frequently involving the second to fourth fingers was mapped to human chromosome 2q34-q36 in a large German family [9].

Syndactyly type II (synpolydactyly)

Synpolydactyly (SPD; MIM 186000) is a dominantly inherited malformation of the distal limbs. It is characterized by soft tissue syndactyly between the third and fourth fingers and between the fourth and fifth toes with a supernumerary digit in the syndactylous web (Fig. **10**). Clinodactyly or camptodactyly of the fifth fingers, syndactyly and brachydactyly of the second to the fifth toes are optional findings. The penetrance of SPD may be incomplete with variable expressivity and asymmetrical involvement. Since in SPD syndactyly can occur without polydactyly but polydactyly does not occur without syndactyly, SPD has been classified as one of the syndactylies. SPD is caused by heterozygous mutations of *HOXD13* [39] resulting in expansions of the 15-residue polyalanine tract in the N-terminal region of the protein. The resulting protein can be expected to regularly bind DNA, but interactions through its N-terminal portion are likely to be disturbed, suggesting a dominant negative effect. Interestingly, both penetrance and severity of the SPD phenotype correlate with expansion size, with large expansion leading to a greater number of limbs involved [23]. The polyalanine tract mutations in SPD have been shown to be meiotically stable over at least seven generations [23]. These findings distinguish *HOXD13* mutations from dynamic repeat expansions found in Huntington disease, Fragile X syndrome and myotonic dystrophy which increase in size when transmitted and lead to a more severe course of the disease in successive generations [4].

Glossary

Anatomy and development	*Molecular genetics*	*Major limb malformations*
Apical Ectodermal Ridge (AER) – a specialized ectoderm covering the distal tip of the limb	Allele – one or alternative forms of a gene occurring at a chromosomal locus	Aphalangia – absence/deformities of certain phalanges
Acromelic – concerning the distal segment of the limb (autopode)	DNA binding domain – permits specific binding of a transcription factor to its target	Adactylia – absence/deformities of certain fingers
Autopode – distal segment of the limb, i.e. hand/foot	Dominant negative – mutational effect inhibiting the function of the wildtype gene product	Acheiria – absence/deformities of a complete hand
Early specification model – a specified cell population expands during limb outgrowth and is determined in a proximodistal sequence	Enhancer – positive regulatory element that stimulates level of transcription, possibly located at a far distance from the gene	Amelia – absent limb
Mesomelic – concerning the middle segment of the limb (zeugopode)	Haploinsufficiency – 50% reduction of gene function, i.e. loss of one allele	Brachydactyly – short digits
Preaxial – concerning the radial side of the limb	Homeobox – characteristic DNA binding motif	Camptodactyly – flexion contracture of finger at the proximal interphalangeal joint
Postaxial – concerning the ulnar side of the limb	Knock-out – targeted inactivation of a gene	Clinodactyly – lateral deviation of finger usually due to a short middle phalanx
Progress zone (PZ) – immature mesenchymal cells in the limb bud beneath the AER	Linkage – genes inherited together due to their locus proximity	Hemimelia – terminal transverse defect with absence of a large portion of the distal segment
PZ zone model – cells are specified as they leave the PZ; cells that leave the PZ late will acquire a more distal fate	MIM – Medelian Inheritance of Men Database http://www.ncbi.nlm.nih.gov/	Polydactyly – supernumerary digits
Rhizomelic – concerning the proximal segment of the limb (stylopode)	Mutation – a nucleotide exchange resulting in a phenotype: A frame-shift mutation alters the translational reading frame of the mRNA, a nonsense mutation results in a stop codon, a missense mutation results in an amino acid exchange	Split-Hand-Foot-Malformation (SHFM) – absence/hypoplasia of median autopode rays
Stylopode – proximal segment of the limb, i.e., humerus/femur	Locus heterogeneity – determination of the same phenotype at different genetic loci	Symphalangism – fused phalanges due to absent interphalangeal joints
Zeugopode – middle segment of the limb, i.e., radius/ulna, tibia/fibula	Promoter – short sequence elements located in the upstream region of a gene that serve to initiate transcription	Syndactyly – fusion of digits
ZPA (zone of polarising activity) – mesenchymal cells at the posterior margin of the limb expressing Shh, involved in anteroposterior patterning of the limb	Transcription factor – recognizes and binds short DNA sequences and regulates transcription	Synostosis – fusion of bones
	Zinc finger – DNA binding motif with a central zinc atom bound by two conserved cysteine/histidine residues	Triphalangeal thumb – thumb with three phalanges instead of two

Syndactyly type III (syndactyly of the fourth and fifth fingers)
Syndactyly type III (SD3) affects the fourth and fifth fingers with the middle phalanx of the fifth finger being rudimentary or absent. When expressed completely, the ring finger is always partially flexed. Isolated SD3 is part of a disease spectrum which leads to oculodentodigital dysplasia disease (ODDD; MIM 164200) if completely expressed, characterized by craniofacial and limb dysmorphisms, spastic paraplegia and neurodegeneration. Mutations in *Connexin 43 (Cx43)* also termed *GJA1* have been shown to be responsible for ODDD and SD3 [44]. During development *Cx43* is strongly expressed in the limb bud, developing digits and cartilage condensation. Six connexins form a connexon that surrounds a pore as a specialized structure and two connexons in adjacent cell membranes can form a gap junction which is important for the passage of ions and small molecules.

Syndactyly type IV (complete syndactyly of all fingers) and type V (associated with metacarpal and metatarsal synostosis)
Syndactyly type IV (SD4) and type V (SD5) are extremely rare disorders. SD4 is characterized by complete syndactyly of all fingers without bony fusion. SD5 is associated with metacarpal and metatarsal fusion, most commonly of the fourth and fifth or the third and fourth digits [54].

Summary and Conclusion

Congenital limb malformations may occur as an isolated trait or in association with a malformation syndrome. On an anatomic and genetic basis they can be divided into six groups consisting of polydactylies, absence deformities, defects in dorsoventral patterning, segmentation disorders, brachydactylies and syndactylies. The presented disorders are mainly caused by mutations in genes which are important during embryonic development of the limb and other organs. These findings have led to a regrouping of limb malformations in genetic terms. Due to the considerable phenotypic variability of congenital limb malformations, the establishment of exact genotype-phenotype correlations for limb malformations is difficult. On the other hand, non-genetic conditions such as amniotic bands or vascular disruptions are caused by external factors. Many conditions currently considered to be non-genetic are likely to be polygenic, meaning that they present only if several genes are mutated or that they may be caused by single gene mutations with low penetrance. Future research of the interaction of genes and their genetic and epigenetic control may help to better understand the pathogenesis and phenotypic variability of congenital limb malformations.

Acknowledgements

This work was supported by a grant from the Deutsche Forschungsgemeinschaft to S. M. We thank Britta Fiebig for support in preparing photographs and X-rays.

Announcement

The authors offer their help for clinical and molecular diagnostics to physicians seeing patients with hand anomalies. Please contact S. M. and provide a photograph, if possible a hand radiograph and a pedigree of the patient.

References

[1] Agarwal P, Wylie JN, Galceran J, Arkhitko O, Li C, Deng C, Grosschedl R, Bruneau BG: Tbx5 is essential for forelimb bud initiation following patterning of the limb field in the mouse embryo. Development 2003; 130: 623 – 633

[2] Al-Shawi R, Ashton SV, Underwood C, Simons JP: Expression of the Ror1 and Ror2 receptor tyrosine kinase genes during mouse development. Dev Genes Evol 2001; 211: 161 – 171

[3] Armour CM, McCready ME, Baig A, Hunter AG, Bulman DE: A novel locus for brachydactyly type A1 on chromosome 5p13.3-p13.2. J Med Genet 2002; 39: 186 – 188

[4] Ashley CT, Warren ST: Trinucleotide repeat expansion and human disease. Ann Rev Genet 1995; 29: 703 – 728

[5] Bähring S, Nagai T, Toka HR, Nitz I, Toka O, Aydin A, Muhl A, Wienker TF, Schuster H, Luft FC: Deletion at 12 p in a Japanese child with brachydactyly overlaps the assigned locus of brachydactyly with hypertension in a Turkish family. Am J Hum Genet 1997; 60: 732 – 735

[6] Bamshad M, Lin RC, Law DJ, Watkins WC, Krakowiak PA, Moore ME, Franceschini P, Lala R, Holmes LB, Gebuhr TC, Bruneau BG, Schinzel A, Seidman JG, Seidman CE, Jorde LB: Mutations in human TBX3 alter limb, apocrine and genital development in ulnar-mammary syndrome. Nat Genet 1997; 16: 311 – 315

[7] Basson CT, Cowley GS, Solomon SD, Weissman B, Poznanski AK, Traill TA, Seidman JG, Seidman CE: The clinical and genetic spectrum of the Holt-Oram syndrome (heart-hand syndrome). N Engl J Med 1994; 330: 885 – 891

[8] Bell J: On brachydactyly and symphalangism. In: Penrose LS (Ed): The Treasury of Human Inheritance. Vol. 5. Cambridge: Cambridge University Press, 1951: 1 – 31

[9] Bosse K, Betz RC, Lee YA, Wienker TF, Reis A, Kleen H, Propping P, Cichon S, Nothen MM: Localization of a gene for syndactyly type 1 to chromosome 2q34-q36. Am J Hum Genet 2000; 7: 492 – 497

[10] Capdevila J, Johnson RL: Endogenous and ectopic expression of noggin suggests a conserved mechanism for regulation of BMP function during limb and somite patterning. Dev Biol 1998; 197: 205 – 217

[11] Celli J, Duijf P, Hamel BC Bamshad M, Kramer B, Smits A, Newbury-Ecob R, Hennekam RC, Van Buggenhout G, van Haeringen A, Woods C, van Essen A, de Waal R, Vriend G, Haber DA, Yang A, McKeon F, Brunner H, van Bokhoven H: Heterozygous germline mutations in the p53 homolog p63 are the cause of EEC syndrome. Cell 1999; 99: 143 – 153

[12] Chen H, Lun Y, Ovchinnikov D, Kokubo H, Oberg KC, Pepicelli CV, Gan L, Lee B, Johnson RL: Limb and kidney defects in Lmx1b mutant mice suggest an involvement of LMX1 B in human nail patella syndrome. Nat Genet 1998; 19: 51 – 55

[13] Davenport TG, Jerome-Majewska LA, Papaioannou VE: Mammary gland, limb and yolk sac defects in mice lacking Tbx3, the gene mutated in human ulnar mammary syndrome. Development 2003; 130: 2263 – 2273

[14] Dreyer S, Zhou G, Baldini A, Winterpacht A, Zabel B, Cole W, Johnson R, Lee B: Mutations in LMX1 B cause abnormal skeletal patterning and renal dysplasia in nail patella syndrome. Nat Genet 1998; 19: 47 – 50

[15] Dudley AT, Ros MA, Tabin CJ: A re-examination of proximodistal patterning during vertebrate limb development. Nature 2002; 418: 539 – 544

[16] Duijf PH, Vanmolkot KR, Propping P, Friedl W, Krieger E, McKeon F, Doetsch V, Brunner HG, van Bokhoven H: Gain-of-function mutation in ADULT syndrome reveals the presence of a second transactivation domain in p63. Hum Mol Genet 2002; 11: 799 – 804

[17] Faiyaz ul Haque M, Uhlhaas S, Knapp M, Schuler H, Friedl W, Ahmad M, Propping P: Mapping of the gene for X-chromosomal split-hand/split-foot anomaly to Xq26-q26.1. Hum Genet 1993; 91: 17 – 19

[18] Faiyaz-Ul-Haque M, Ahmad W, Zaidi SH, Haque S, Teebi AS, Ahmad M, Cohn DH, Tsui LC: Mutation in the cartilage-derived morphogenetic protein-1 (CDMP1) gene in a kindred affected with fibular hypoplasia and complex brachydactyly (DuPan syndrome). Clin Genet 2002; 61: 454 – 458

[19] Farabee WC: Hereditary and Sexual Influence in Meristic Variation: A Study of Digital Malformation in Man. Harvard University: Ph. D. thesis, 1903

[20] Francis-West PH, Abdelfattah A, Chen P, Allen C, Parish J, Ladher R, Allen S, MacPherson S, Luyten FP, Archer CW: Mechanisms of GDF-5 action during skeletal development. Development 1999; 126: 1305 – 1315

[21] Gao B, Guo J, She C, Shu A, Yang M, Tan Z, Yang X, Guo S, Feng G, He L: Mutations in IHH, encoding Indian hedgehog, cause brachydactyly type A-1. Nat Genet 2001; 28: 386 – 388

[22] Gong Y, Krakow D, Marcelino J, Wilkin D, Chitayat D, Babul-Hirji R, Hudgins L, Cremers CW, Cremers FP, Brunner HG, Reinker K, Rimoin DL, Cohn DH, Goodman FR, Reardon W, Patton M, Francomano CA, Warman ML: Heterozygous mutations in the gene encoding noggin affect human joint morphogenesis. Nat Genet 1999; 21: 302 – 304

[23] Goodman FR, Mundlos S, Muragaki Y, Donnai D, Giovannucci-Uzielli ML, Lapi E, Majewski F, McGaughran J, McKeown C, Reardon W, Upton J, Winter RM, Olsen BR, Scambler PJ: Synpolydactyly phenotypes correlate with size of expansions in HOXD13 polyalanine tract. Proc Natl Acad Sci USA 1997; 94: 7458 – 7463

[24] Ingham PW, McMahon AP: Hedgehog signaling in animal development. Genes Dev 2001; 15: 3059 – 3087

[25] Innis JW, Goodman FR, Bacchelli C, Williams TM, Mortlock DP, Sateesh P, Scambler PJ, McKinnon W, Guttmacher AE: A HOXA13 allele with a missense mutation in the homeobox and a dinucleotide deletion in the promoter underlies Guttmacher syndrome. Hum Mutat 2002; 19: 573 – 574

[26] Johnson D, Kan S, Oldridge M, Trembath RC, Roche P, Esnouf RM, Giele H, Wilkie AOM: Missense mutations in the homeodomain of HOXD13 are associated with brachydactyly types D and E. Am J Hum Genet 2003; 72: 984 – 997

[27] Johnson KR, Lane PW, Ward-Bailey P, Davisson MT: Mapping the mouse dactylaplasia mutation, Dac, and a gene that controls its expression, mdac. Genomics 1995; 20: 457 – 464
[28] Johnson RL, Tabin CJ: Molecular models for vertebrate limb development. Cell 1997; 90: 979 – 990
[29] Kohlhase J, Wischermann A, Reichenbach H, Froster U, Engel W: Mutations in the SALL1 putative transcription factor gene cause Townes-Brocks syndrome. Nat Genet 1998; 18: 81 – 83
[30] Kornak U, Mundlos S: Genetic Disorders of the Skeleton: A Developmental Approach. Am J Hum Genet 2003; 73: 447 – 474
[31] Koster R, Stick R, Lossli F, Wittbrodt J: Medaka spalt acts as a target gene of hedgehog signaling. Development 1997; 124: 3147 – 3156
[32] Lehmann K, Seemann P, Stricker S, Sammar M, Meyer B, Süring K, Majewski F, Tinschert S, Muller D, Knaus P, Nürnberg P, Mundlos S: Mutations in bone morphogenetic protein receptor1 B cause brachydactyly type A2. Proc Natl Acad Sci USA 2003; 100: 12277 – 12282
[33] Lettice LA, Heaney SJ, Purdie LA, Li L, de Beer P, Oostra BA, Goode D, Elgar G, Hill R, de Graaff E: A long-range Shh enhancer regulates expression in the developing limb and fin and is associated with preaxial polydactyly. Hum Mol Genet 2003; 12: 1725 – 1735
[34] Li QY, Newbury-Ecob RA, Terrett JA, Wilson DI, Curtis AR, Yi CH, Gebuhr T, Bullen PJ, Robson SC, Strachan T, Bonnet D, Lyonnet S, Young ID, Raeburn JA, Buckler AJ, Law DJ, Brook JD: Holt-Oram syndrome is caused by mutations in TBX5, a member of the Brachyury (T) gene family. Nat Genet 1997; 15: 21 – 29
[35] Mariani FV, Martin GR: Deciphering skeletal patterning: clues from the limb. Nature 2003; 423: 319 – 325
[36] McGrath JA, Duijf PH, Doetsch V, Irvine AD, de Waal R, Vanmolkot KR, Wessagowit V, Kelly A, Atherton DJ, Griffiths WA, Orlow SJ, van Haeringen A, Ausems MG, Yang A, McKeon F, Bamshad MA, Brunner HG, Hamel BC, van Bokhoven H: Hay-Wells syndrome is caused by heterozygous missense mutations in the SAM domain of p63. Hum Mol Genet 2001; 10: 221 – 229
[37] McMahon AP: More surprises in the hedgehog signaling pathway. Cell 2000; 100: 185 – 188
[38] Mortlock DP, Innis JW: Mutation of HOXA13 in hand-foot-genital syndrome. Nat Genet 1997; 15: 179 – 180
[39] Muragaki Y, Mundlos S, Upton J, Olsen BR: Altered growth and branching patterns in synpolydactyly caused by mutations in HOXD13. Science 1996; 272: 548 – 551
[40] Nunes ME, Schutt G, Kapur RP, Luthardt F, Kukolich M, Byers P, Evans JP: A second autosomal split hand/split foot locus maps to chromosome 10q24-q25. Hum Mol Genet 1995; 4: 2165 – 2167
[41] Oldridge M, Fortuna AM, Maringa M, Propping P, Mansour S, Pollitt C, DeChiara TM, Kimble RB, Valenzuela DM, Yancopoulos GD, Wilkie AO: Dominant mutations in ROR2, encoding an orphan receptor tyrosine kinase, cause brachydactyly type B. Nat Genet 2000; 24: 275 – 278
[42] Papaioannou VE: T-box genes in development: from hydra to humans. Int Rev Cytol 2001; 207: 1 – 70
[43] Parr BA, McMahon AP: Dorsalizing signal Wnt-7 a required for normal polarity of D-V and A-P axes of mouse limb. Nature 1995; 374: 350 – 353
[44] Paznekas WA, Boyadjiev SA, Shapiro RE, Daniels O, Wollnik B, Keegan CE, Innis JW, Dinulos MB, Christian C, Hannibal MC, Jabs EW: Connexin 43 (GJA1) mutations cause the pleiotropic phenotype of oculodentodigital dysplasia. Am J Hum Genet 2003; 72: 408 – 418
[45] Polinkovsky A, Robin NH, Thomas JT, Irons M, Lynn A, Goodman FR, Reardon W, Kant SG, Brunner HG, van der Burgt I, Chitayat D, McGaughran J, Donnai D, Luyten FP, Warman ML: Mutations in CDMP1 cause autosomal dominant brachydactyly type C. Nat Genet 1997; 17: 18 – 19
[46] Radhakrishna U, Bornholdt D, Scott HS, Patel UC, Rossier C, Engel H, Bottani A, Chandal D, Blouin JL, Solanki JV, Grzeschik KH, Antonarakis SE: The phenotypic spectrum of GLI3 morphopathies includes autosomal dominant preaxial polydactyly type-IV and postaxial polydactyly type-A/B; No phenotype prediction from the position of GLI3 mutations. Am J Hum Genet 1999; 65: 645 – 655
[47] Radhakrishna U, Wild A, Grzeschik KH, Antonarakis SE: Mutation in GLI3 in postaxial polydactyly type A. Nat Genet 1997; 17: 269 – 271
[48] Scherer SW, Poorkaj P, Massa H, Soder S, Allen T, Nunes M, Geshuri D, Wong E, Belloni E, Little S, Zhou L, Becker D, Kere J, Ignatius J, Niikawa N, Fukushima Y, Hasegawa T, Weissenbach J, Boncinelli E, Trask B, Tsui LC, Evans JP: Physical mapping of the split hand/split foot locus on chromosome 7 and implication in syndromic ectrodactyly. Hum Mol Genet 1994; 3: 1345 – 1354
[49] Schuster H, Wienker TE, Bahring S, Bilginturan N, Toka HR, Neitzel H, Jeschke E, Toka O, Gilbert D, Lowe A, Ott J, Haller H, Luft FC: Severe autosomal dominant hypertension and brachydactyly in a unique Turkish kindred maps to human chromosome 12. Nat Genet 1996; 13: 98 – 100
[50] Schwabe GC, Tinschert S, Buschow C, Meinecke P, Wolff G, Gillessen-Kaesbach G, Oldridge M, Wilkie AO, Kömec R, Mundlos S: Distinct mutations in the receptor tyrosine kinase gene ROR2 cause brachydactyly type B. Am J Hum Genet 2000; 67: 822 – 831
[51] Schwabe GC, Türkmen S, Leschik G, Palanduz S, Meinecke P, Stöver B, Goecke TO, Mundlos S: A homozygous missense mutation in the prodomain of *CDMP1* causes a pronounced form of brachydactyly type C. Am J Med Genet 2004; 124 A: 356 – 363
[52] Schwabe GC, Trepczik B, Süring K, Brieske N, Tucker AS, Sharpe PT, Minami Y, Mundlos S: Ror2 knock out mouse as a model for the developmental pathology of autosomal recessive Robinow syndrome. Dev Dyn 2004; 229: 400 – 410
[53] Storm EE, Huynh TV, Copeland NG, Jenkins NA, Kingsley DM, Lee SJ: Limb alterations in brachypodism mice due to mutations in a new member of the TGF beta-superfamily. Nature 1994; 368: 639 – 643
[54] Temtamy SA, McKusick VA: The Genetics of Hand Malformations. New York: The National Foundation March of Dimes, 1978
[55] Thomas JT, Kilpatrick MW, Lin K, Erlacher L, Lembessis P, Costa T, Tsipouras P, Luyten FP: Disruption of human limb morphogenesis by a dominant negative mutation in CDMP1. Nat Genet 1997; 17: 58 – 64
[56] Thomas JT, Lin K, Nandedkar M, Camargo M, Cervenka J, Luyten FP: A human chondrodysplasia due to a mutation in a TGF-beta superfamily member. Nat Genet 1996; 12: 315 – 317
[57] Tsukurov O, Boehmer A, Flynn J, Nicolai JP, Hamel BC, Traill S, Zaleske D, Mankin HJ, Yeon H, Ho C: A complex bilateral polysyndactyly disease locus maps to chromosome 7q36. Nat Genet 1994; 6: 282 – 286
[58] Vortkamp A, Gessler M, Grzeschik K: GLI3 zinc-finger gene interrupted by translocations in Greig syndrome families. Nature 1991; 352: 539 – 540
[59] Wassif CA, Maslen C, Kachilele-Linjewile S, Lin D, Linck LM, Connor WE, Steiner RD, Porter FD: Mutations in the human sterol delta7-reductase gene at 11 q12 – 13 cause Smith-Lemli-Opitz syndrome. Am J Hum Genet 1998; 63: 55 – 62
[60] Witsch-Baumgartner M, Fitzky BU, Orogelkova M, Kraft HG, Moebius FF, Glossmann H, Seedorf U, Gillessen-Kaesbach G, Hoffmann GF, Clayton P, Kelley RI, Utermann G: Mutational spectrum in the Delta7-sterol reductase gene and genotype-phenotype correlation in 84 patients with Smith-Lemli-Opitz syndrome. Am J Hum Genet 2000; 66: 402 – 412