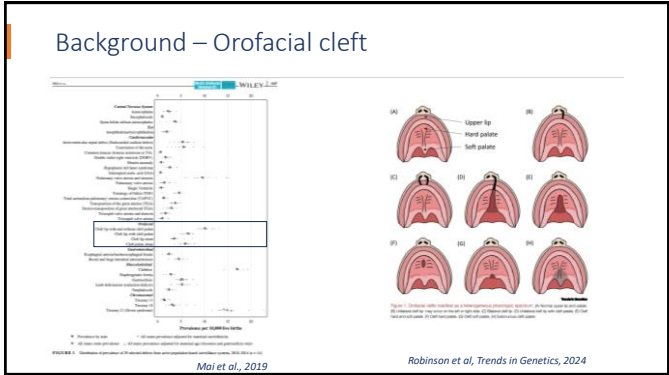




4



5

Mosaic Cornelia de Lange syndrome: genes, tissues and clinical severity

1 GENES IMPLICATED

- NIPBL**
+10-15% of affected individuals; most reported patient mosaicism is NIPBL.
- SMC1A**
Three affected FDM cases by 2025, including one female with classic CdLS.
- SMC3 / RAD21**
Patient mosaic variants are cited in consensus; evidence is case-level.

2 WHERE MOSAICISM IS FOUND

- Uncultured fibroblasts**
COMMONLY USED CHALLENGE
- Buccal epithelial DNA**
PRACTICAL HIGH-YIELD
- Peripheral blood**
CALCULATED CASE
- Other informative tissues**
WHEN AVAILABLE

3 DOES VAF TRACK SEVERITY?

NO

Mosaic ≠ mild

No dependable VAF-severity correlation has been demonstrated.

- VAF in an accessible tissue is a sampling proxy—not a whole-body measure.
- Variant effect, embryonic timing, tissue distribution and X-inactivation also matter.

Deardorff et al., GeneReviews 2026 • Kline et al., Nat Rev Genet 2018

6

ORIGINAL ARTICLE

Genetic heterogeneity in Cornelia de Lange syndrome (CdLS) and CdLS-like phenotypes with observed and predicted levels of mosaicism

Morad Ansari,¹ Gemma Poole,¹ Quentin Ferry,^{2,3} Kathleen Williamson,¹ Roland Adridge,¹ Alison M Maynard,¹ Henant Bengani,¹ Cheng Yee Chan,¹ Hülya Kayrakli,⁴ Sahin Avcı,⁴ Rosal C M Hennekam,⁵ Anne K Lampert,⁶ Egbert Redeker,⁷ Tessa Homfroy,⁸ Alison Ross,⁸ Marie Falkenberg Smørdal,⁹ Sahar Mansouri,¹⁰ Michael J Parker,¹⁰ Jacqueline A Cook,¹⁰ Miranda Spitt,¹¹ Richard B Fisher,¹¹ Alan Fryer,¹² Alex C Magee,¹³ Andrew Wilkie,¹⁴ Angela Barrios,¹⁵ Angela F Brady,¹⁶ Nicola S Cooper,¹⁷ Catherine Mearse,¹⁸ Chary Deshpande,¹⁹ Christopher P Bennett,²⁰ Daniela T Pilz,²¹ Deborah Rudzki,¹⁹ Deirdre Cilliers,²² Diana S Johnson,²³ Dragana Jaskova,²⁴ Elisabeth Rosser,²⁵ Elizabeth M Thompson,^{21,26} Emma Malsbenden,²⁶ Eghen Kinning,²⁷ Fana Stewart,²⁸ Frances Flinter,¹⁹ Karla M Gersh,²⁹ Helen Cox,¹⁹ Helen V Firth,²⁷ Helen Kingston,²⁸ Jamie S Wee,²⁹ Jane A Hurst,³⁰ Jill Clayton-Smith,³¹ John Tolmie,³² Julie Vogt,³³ Katrina Tamon-Brown,³⁴ Kate Chundrow,³⁵ Katrina Prescott,³⁶ Louise Wilson,³⁷ Mubiyeh Ibrahim,³⁸ Mireia McElduff,³⁹ Rosemarie Davidson,⁴⁰ Sally-Ann Lynch,⁴¹ Sanjay Soodhy,⁴² Sanju G Mehta,⁴³ Shane A McKee,⁴⁴ Shehla Mohammed,⁴⁵ Simon Holden,⁴⁶ Soo-Mi Park,⁴⁷ Susan E Holder,⁴⁸ Victoria Harrison,⁴⁹ Ververse McConnell,⁵⁰ Wayne K Lam,⁵¹ Andrew J Green,^{51,52} Dian Dorra,⁵³ Maria Birne-Glindicz,^{53,54} Deirdre E Donnelly,⁵⁵ Christopher Nelliker,⁵⁶ Martin S Taylor,¹ David R FitzPatrick¹

Is the mutation post-zygotic or has there been a revertant event?

3/46 NIPBL, 1/5 SMC1A and 1/5 SMC3 mutations in our cohort were mosaic. All three mosaic NIPBL mutations were in individuals with severe and typical CdLS

J Med Genet 2014



Yuan B, Neira J, Pehlivan D, et al. Genetics in Medicine. 2019;21(3):663–675.

Why heterozygous females develop STAG2-related disease

Mechanism	Molecular consequence	Clinical relevance
Random X-inactivation	Females become mosaics of cells expressing either the normal or variant STAG2 allele	A proportion of cells are STAG2-deficient
STAG2 dosage sensitivity	Reduced STAG2 disrupts cohesin-mediated chromatin organisation and transcription	One normal allele cannot fully rescue development
Tissue-specific X-inactivation	The proportion of STAG2-deficient cells differs between brain, craniofacial tissues, skin and blood	Variable organ involvement and severity
Skewed X-inactivation	Some tissues may preferentially retain the mutant X as the active chromosome	Markedly reduced or absent STAG2 expression
Variable XCI escape	STAG2 may partially escape X-inactivation in some tissues	Further modifies effective STAG2 dosage
Cellular selection	STAG2-negative fibroblasts may acquire a proliferative advantage	X-inactivation patterns can change over time
Partial compensation	Increased STAG1 and ectopic STAG3 can partially replace STAG2 within cohesin	Permits cellular survival but does not restore normal development

Moronta Gines et al., iScience, 2025

CHARGE syndrome

- Bilateral choanal atresia
 - Left facial palsy, resolving
 - Vestibular symptoms
 - Chorioretinal coloboma
 - no effect on vision
 - Minor ASD and VSD
- Bilateral choanal atresia
 - Major feeding difficulties, PEG
 - Pontocerebellar hypoplasia with poorly developed gyri/sulci
 - PDA and ASD
 - Hearing loss

missense variant CHD7 protein truncating

Verheij's diagnostic criteria (2005)
(Verheij A. 2005; Am J Med Genet A. 133:33A(3):306-8)

Major criteria (3 C's)
Coloboma (iris or choroid, with or without microphthalmia)
Choanal atresia
Hypoplastic semi-circular Canals

Minor criteria
Bilaterocephalic dysfunction (auditory dysfunction, cranial nerve VII to XII palsy and neurosensory deafness)
Regenerative hypophyseal dysfunction (including GH and gonadotrophin deficiencies)
Abnormal middle or external ear
Malformations of mediastinal organs (heart, esophagus)
Mental retardation

Diagnostic criteria interpretation
Typical CHARGE: 3 major or 2 major and 2 minor criteria
Partial/atypical CHARGE: 2 major and 3 minor criteria
Atypical CHARGE: 1 major or 1 major and 3 minor criteria



RTS1; OMIM #180849, CREBBP-RTS

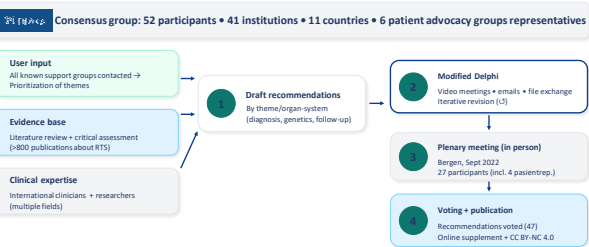
Distinctive Gestalt	Preeclampsia
Radial deviation of the thumbs	IUGR
Keloids	Microcephaly
Lower average cognitive skills	

A

RTS2; OMIM #613684, EP300-RTS

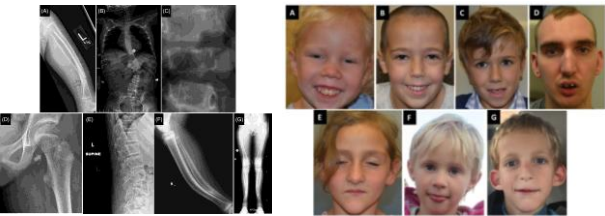
Distinctive Gestalt	Preeclampsia
Radial deviation of the thumbs	IUGR
Keloids	Microcephaly
Lower average cognitive skills	

Turning genomic answers into precision care pathways



Lacombe et al. J Med Genet 2024
Diagnosis and management in Rubinstein-Taybi syndrome: first international consensus statement.

Glass syndrome – SATB2 - AD



Clinical Genetics, 2017, DOI: (10.1111/cge.13121)

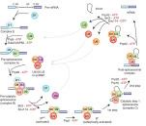
Gene Reviews

Developmental gene regulation and chromatin architecture

Syndrome	Gene(s)	Principal mechanism	Craniofacial and palatal phenotype
Cornelia de Lange	<i>NIPBL, SMC1A, SMC3, RAD21, HDAC8</i>	Altered cohesin loading, function or recycling disrupts chromatin looping and developmental transcription	Characteristic facial gestalt, micrognathia and high-arched palate; cleft palate occurs in a minority
STAG2-associated Mullegama-Klein-Martinez	<i>STAG2</i>	Cohesin subunit dysfunction alters chromatin topology and developmental gene expression	Microcephaly, prominent forehead, deep-set eyes, broad nasal bridge and micrognathia; palatal abnormalities vary
CHARGE	<i>CHD7</i>	ATP-dependent chromatin-remodelling dysfunction disrupts neural crest and placodal developmental programmes	Choanal atresia, characteristic ears, facial asymmetry and cranial-nerve abnormalities; cleft lip or palate in a subset
Rubinstein-Taybi	<i>CREBBP, EP300</i>	Reduced histone acetylation and transcriptional co-activator function alter enhancer activity	Characteristic facial and dental phenotype, high or narrow palate; cleft palate is uncommon
SATB2-associated	<i>SATB2</i>	Loss of chromatin organisation and transcriptional regulation of jaw, palate and osteoblast programmes	Cleft or high-arched palate, dental abnormalities, mandibular abnormalities and severe speech disorder

Craniofacial spliceosomopathies

- A ribonucleoprotein complex.
- The site in the cell nucleus where introns are excised from precursor messenger RNA and exons are joined together to form functional messenger RNA.
- A dynamic ribonucleoprotein (RNP) machine that performs the function of splicing.
- Distinguished in 2 different types in mammals (major and minor) that splice out mutually exclusive types of introns.
- Assembled in distinct subcellular compartments as part of a cellular quality control mechanism.



Guion-Almeida, AD, *EFTUD2*

Nager, AD, *SF3B4*

Cerebro-Costo-Mandibular, AD, *SNRPB*

Richieri-Costa-Pereira, AR, *EIF4A3*

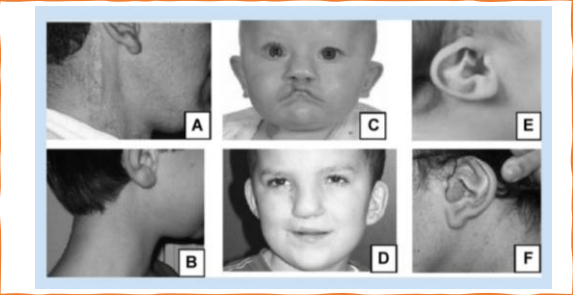
Burn-McKeown, AR, *TXNL4*

Podgett, 2012
Matera and Wang, 2014

Lehalle, 2015

Ribosome biogenesis and spliceosome dysfunction

Syndrome	Gene(s)	Principal mechanism	Craniofacial and palatal phenotype
Treacher Collins	<i>TCOF1</i> , <i>POLR1B</i> , <i>POLR1C</i> , <i>POLR1D</i>	Impaired rRNA transcription or processing causes nucleolar stress, p53 activation and loss of neuroepithelial and cranial neural crest progenitors	Bilateral malar and mandibular hypoplasia, microtia, external auditory-canal abnormalities and cleft palate
Mandibulofacial dysostosis with microcephaly	<i>EFTUD2</i>	US spliceosome dysfunction causes widespread mis-splicing, p53 dysregulation and apoptosis of cranial neural crest progenitors	Malar and mandibular hypoplasia, microtia, hearing loss, choanal abnormalities and variable cleft or high-arched palate
Burn-McKeown	<i>TXNL4A</i>	Reduced US snRNP assembly and spliceosome activity impair cranial neural crest formation and survival	Choanal atresia, lower-eyelid abnormalities, prominent nose, short philtrum and variable cleft lip or palate



Milunsky JM, Maher TM, Zhao G, et al.
American Journal of Medical Genetics Part A. 2011;155A:22–32.

Branchio-oculo-facial syndrome

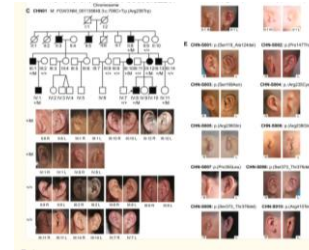
- AD **TFAP2A** disorder, chromosome 6p24.3
- Approximately **50–60% *de novo***
- Marked inter- and intrafamilial variability, without a reliable genotype–phenotype correlation
- Most reported missense variants affect the AP-2 α DNA-binding domain and can produce hypomorphic, null or dominant-negative effects
- Clinical diagnostic criteria
 - Do not cauterise the characteristic branchial skin defects.
 - Assess for an associated sinus tract and ectopic thymic tissue before excision.
 - Baseline assessment should include ophthalmology, hearing/ENT, cleft and dental evaluation, and renal ultrasonography.

Gene Reviews

22

FOXI3-related craniofacial microsomia

- AD with reduced penetrance
- Often bilateral and asymmetric, external ear abnormalities
- Rarer, CFM/Goldenhar



Mao, Borel et al, Nature Communications, 2022

23

Pharyngeal-arch, neural-crest and placodal patterning

Syndrome	Gene(s)	Principal mechanism	Craniofacial and palatal phenotype
Branchio-oculo-facial	TFAP2A	Disrupted AP-2 α regulation in surface ectoderm and cranial neural crest alters facial-prominence growth and fusion	Cervical branchial skin defects, ocular anomalies and characteristic cleft or pseudocleft of the upper lip; cleft palate can occur
FOXI3-related craniofacial microsomia	FOXI3	Abnormal pharyngeal ectoderm and endoderm signalling impairs the developmental niche supporting first- and second-arch neural crest	Microtia, mandibular hypoplasia and facial asymmetry; clefting is not a defining feature
Branchio-oto-renal spectrum	EYA1, SIX1	Disruption of the EYA1–SIX1 regulatory network affects pharyngeal pouches and otic placodes	Branchial cysts or fistulae, preauricular pits, ear abnormalities and hearing loss; cleft palate is uncommon

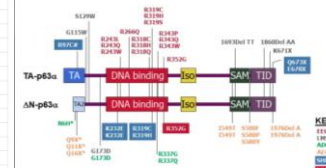
24

Van der Woude syndrome
Mimics non-syndromic clefting
AD with variable penetrance
Lip pits
Hypodontia
Ankyloblepharon
Syndactyly

Table 2.
TP63-Related Disorders: Comparison of Phenotypes by Select Features

Feature	TP63-Related Disorder					Onset/ cleft #
	AEC	ADULT	EECS	Limb- mammary	SHFM4	
Askylophorane filiform aduans	X					
Ectodermal dysplasia	X	X	X			Rare
Hypohidrosis ¹	X	X		X		
Nail dysplasia			X	Mild X		
Sparse hair		X	X	X		
Tooth abnormalities	X	X	X	X		
Cleft lip/palate	X					X
Split hand/foot malformation / syndactyly	X	X	X	X	X	
Lacrimal duct obstruction	X	X	X	X		
Dermal sinuses	X					
Hypopigmentation		X	X	X		
Hypogonads		X	X			
Triuma						
Excessive freckling	X					
Hypoplastic breasts		X	X			
Hypoplastic nipples	X	X				

ADULT = acro-dermato-ungual-lacral-tooth; AEC = ankylobiphano-ectodermal defects-cleft lip/palate; EEC3 = ectrodactyly (split hand/foot malformation), ectodermal dysplasia, cleft lip/palate syndrome 3; SHFM4 = split-hand/foot malformation type 4



Periderm, epithelial adhesion and facial fusion

Syndrome	Gene(s)	Principal mechanism	Craniofacial and palatal phenotype
Van der Woude and popliteal pterygium	<i>IRF6</i>	Impaired periderm differentiation and abnormal oral epithelial adhesion interfere with lip and palatal fusion	Cleft lip with or without cleft palate, isolated cleft palate and lower-lip pits
TP63-related ectodermal dysplasia and clefting spectrum	<i>TP63</i>	Disruption of the TP63-IRF6 regulatory feedback loop alters epithelial proliferation and differentiation	Cleft lip or palate accompanied by variable ectodermal and limb abnormalities
Blepharocheilodontic 1 and CDH1-related clefting	<i>CDH1</i>	Reduced E-cadherin-mediated adhesion affects epithelial integrity and cranial neural crest migration	Cleft lip or palate, dental abnormalities and lower-eyelid anomalies; penetrance is incomplete

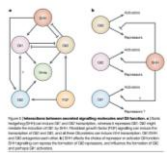
Culler Jones syndrome



Truncating and zinc-finger variants in *GLI3* are associated with hypopituitarism

Corder et al, 2022

- SHH protein acts as a crucial factor that patterns the ventral forebrain and is required for the division of the primordial eye field and brain into 2 distinct halves.
- Occasional pituitary hypoplasia and/or holoprosencephaly also seen in patients with Pallister-Hall syndrome caused by *GLI3* mutations



Variant Type	Percentage (%)
Truncating variants	~80
Zinc-finger variants	~20

Peters-Plus syndrome – *B3GLCT* - AR

FDNA

Peters-Plus Syndrome OMIM# 261540

Facial Gestalt



Associated Gene

B3GLCT



Most affected individuals tested to date are homozygous for a splice site pathogenic variant in intron 8 (c.660+1G>A) (Lesnik Oberstein et al 2006). However, several other loss-of-function variants have been identified, including pathogenic missense variants located in the putative catalytic domain of the enzyme.

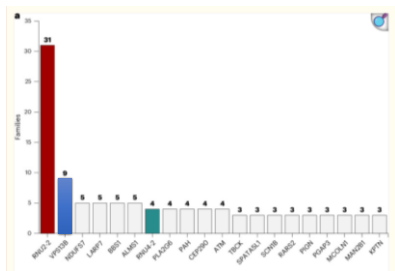
Gene Reviews

- non-progressive, mild-moderate developmental delay
- a variety of ophthalmic abnormalities; early onset myopia and a progressive retinopathy
- characteristic craniofacial features comprising microcephaly, thick hair, eyebrows and eyelashes, wave-shaped and downslanting eyelids, a short philtrum that does not cover the front teeth giving an open-mouth expression with prominent incisors, and micrognathia
- childhood hypotonia, joint hyperextensibility, neutropenia



Douzgou S et al, Am J Med Genet, 2011
Douzgou S and Petersen MB, Clin Genet, 2011

Cohen syndrome



Bar plot showing the 20 most frequent recessive diagnoses in the rare disease arm of 100KGP
Jackson A, Blakes AJM, Alhaddad B, et al. Nat Genet. 2026;58(4):798-809

Miscellaneous

Syndrome	Gene(s)	Principal mechanism	Craniofacial and palatal phenotype
Culler-Jones spectrum	GLI2	Reduced GLI activator function impairs Sonic Hedgehog transcriptional output and midline craniofacial patterning	Midface hypoplasia, solitary maxillary central incisor and occasional cleft lip or palate; pituitary abnormalities and polydactyly are important diagnostic clues
Peters-plus	B3GLCT	Loss of B1,3-glucosylation of O-fucosylated thrombospondin type-1 repeats disrupts ER quality control, protein secretion and extracellular-matrix function	Peters anomaly, cleft lip with or without cleft palate, characteristic face, short limbs and growth restriction
Cohen	VPS13B	Abnormal Golgi organisation, glycan maturation, lipid handling and endosomal-lysosomal trafficking	Short philtrum, thick hair and eyebrows, short upslanting palpebral fissures and high or narrow palate; clefting is not characteristic

Chromosomal Causes Of Craniofacial syndromes with clefting

- Prenatal CLP with multiple anomalies: trisomy 13, trisomy 18 and 4p deletion
- CP, submucous CP or VPI: 22q11.2 deletion
- CP plus severe speech and dental abnormalities: 2q33.1 deletion involving SATB2
- Cleft plus branchial and ocular anomalies: 6p24.3 deletion involving TFAP2A
- HPE plus midline cleft: 7q36, 13q32 or 18p deletion
- Pierre Robin sequence: 17q24.3 regulatory rearrangement affecting SOX9
- Craniosynostosis: 7p21, 9p deletion or 9q22.3 deletion



Take home messages

- Craniofacial syndromes converge anatomically on neural crest, pharyngeal arches, facial prominences and palatal fusion—but they originate at very different molecular levels: chromatin, ribosome, spliceosome, developmental patterning, epithelial fusion, signalling or secretory-system homeostasis
- Chromatin-organisation genes enriched in cleft palate rather than CL/P
- Significantly greater de novo protein-altering variant burden in syndromic than nonsyndromic cleft palate
- IRF6 enrichment predominantly in apparently nonsyndromic cases
- SATB2 shared across syndromic and nonsyndromic groups
- Supports a biological continuum rather than a rigid syndromic/nonsyndromic dichotomy

34



Manchester Rare
Conditions Centre

MANCHESTER EMBG

SYNDROMOLOGY AND
DYSMORPHOLOGY COURSE 2026

18-19 October 2026 • Manchester Academic Health Science Centre, Manchester, M13 9PL



ESHG
The European Society
of Human Genetics

Thank you !



NORWEGIAN CENTRE FOR
RARE DISEASES



HELSE BERGEN

35

References

1. Naqvi S, Hoskins H, Wilke F, et al. Decoding the human face: progress and challenges in understanding the genetics of craniofacial morphology. *Annu Rev Genomics Hum Genet.* 2022;23:383–412.
2. Selleri L, Rijli FM. Shaping faces: genetic and epigenetic control of craniofacial morphogenesis. *Nat Rev Genet.* 2023;24:610–626.
3. Robinson K, Curtis SW, Leslie EJ. The heterogeneous genetic architectures of orofacial clefts. *Trends Genet.* 2024;40(5):410–421.
4. Robinson KR, Curtis SW, Paschall JE, et al. Distinguishing syndromic and nonsyndromic cleft palate through analysis of protein-altering de novo variants in 818 trios. *Am J Hum Genet.* 2025;112(11):2679–2692.
5. Shull LC, Artinger KB. Epigenetic regulation of craniofacial development and disease. *Birth Defects Res.* 2024;116(1):e2271.
6. Jones NC, Lynn ML, Gaudenz K, et al. Prevention of the neurocristopathy Treacher Collins syndrome through inhibition of p53 function. *Nat Med.* 2008;14(2):125–133.
7. Calo E, Gu B, Bowen ME, et al. Tissue-selective effects of nucleolar stress and rDNA damage in developmental disorders. *Nature.* 2018;554(7690):112–117.
8. Griffin C. Modeling craniofacial spliceosomopathies: a pathway toward deciphering disease mechanisms. *Front Cell Dev Biol.* 2025;13:1624043.
9. Park BY, Tachi-Duprat M, Iheuevel C, Devotta A, Saint-Jeanet JP. The core splicing factors EFTUD2, SNRPB and TXNL4A are essential for neural crest and craniofacial development. *J Dev Biol.* 2022;10(3):29.
10. Mao K, Borel C, Ansar M, et al. FOXI3 pathogenic variants cause one form of craniofacial microsomia. *Nat Commun.* 2023;14:2026.
11. Milunsky JM, Maher TM, Zhao G, et al. Genotype–phenotype analysis of the branchio-oculo-facial syndrome. *Am J Med Genet A.* 2011;155A(1):22–32.
12. Smith RH, Azaleu H. Branchiootorenal spectrum disorder. updated 2025 Jun 26. In: Adam MP, Bick S, Mirzaa GM, et al., editors. *GeneReviews*®. Seattle: University of Washington
13. Thomason HA, Zhou H, Kouwenhoven EN, et al. Cooperation between the transcription factors p63 and IRF6 is essential to prevent cleft palate in mice. *J Clin Invest.* 2010;120(5):1561–1569.
14. Cox LL, Cox TC, Moreno Uribe LM, et al. Mutations in the epithelial cadherin–p120-catenin complex cause Mendelian non-syndromic cleft lip with or without cleft palate. *Am J Hum Genet.* 2018;102(6):1143–1157.
15. Xu J, Iyyanar PPR, Lan Y, Jiang R. Sonic hedgehog signaling in craniofacial development. *Differentiation.* 2023;133:60–76. doi:10.1016/j.diff.2023.07.002.
16. Vasudevan D, Takeuchi H, Johar SS, Majerus E, Haltiwanger RS. Peters plus syndrome mutations disrupt a noncanonical ER quality-control mechanism. *Curr Biol.* 2015;25(3):286–295. Vacca F, Valcin B, Ansar M. Exploring the pathological mechanisms underlying Cohen syndrome. *Front Neurosci.* 2024;18:1431400.

36
