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Genetics Corner: A Familial Variant in *CDH1* Connects the Dots between Oral Clefts in Two Sisters and Gastric Cancer in their Mother

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Clinical Summary:

The Genetics team evaluated an 18-year-old female with repaired bilateral cleft lip and palate in the Craniofacial clinic. She was in good health with normal growth and development. She had no other dysmorphic features. Her family history was significant for unilateral cleft lip and palate in her elder sister, age 25. Their mother died at age 36 when the patient was about ten years old. She had gastric cancer, diagnosed at age 31. No one else in the family had gastric cancer, other cancers, or oral clefts.

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Hereditary cancer predisposition gene testing in the patient's mother detected a likely pathogenic splice site variant in *CDH1*: c.1565+dupT. This established the diagnosis of autosomal dominant Hereditary Diffuse Gastric Cancer (HDGC) syndrome in her mother. There is an increased risk for oral clefts in HDGC families. The patient and her sister were tested and are heterozygous for the familial variant in *CDH1.*

Discussion:

Cleft lip with or without cleft palate (CL/P) is the most common craniofacial anomaly in humans. The incidence, 1 in 700–1000 live births, varies with sex, race and ethnicity. There is a higher incidence in people of Asian and Native American ancestry and a lower incidence in Black/African Americans. More males are affected than females in a 2:1 ratio. Unilateral cleft lip occurs more commonly, typically on the left side, while bilateral cleft lip occurs less often. Midline and lateral cleft lips are the rarest forms of oral clefts.

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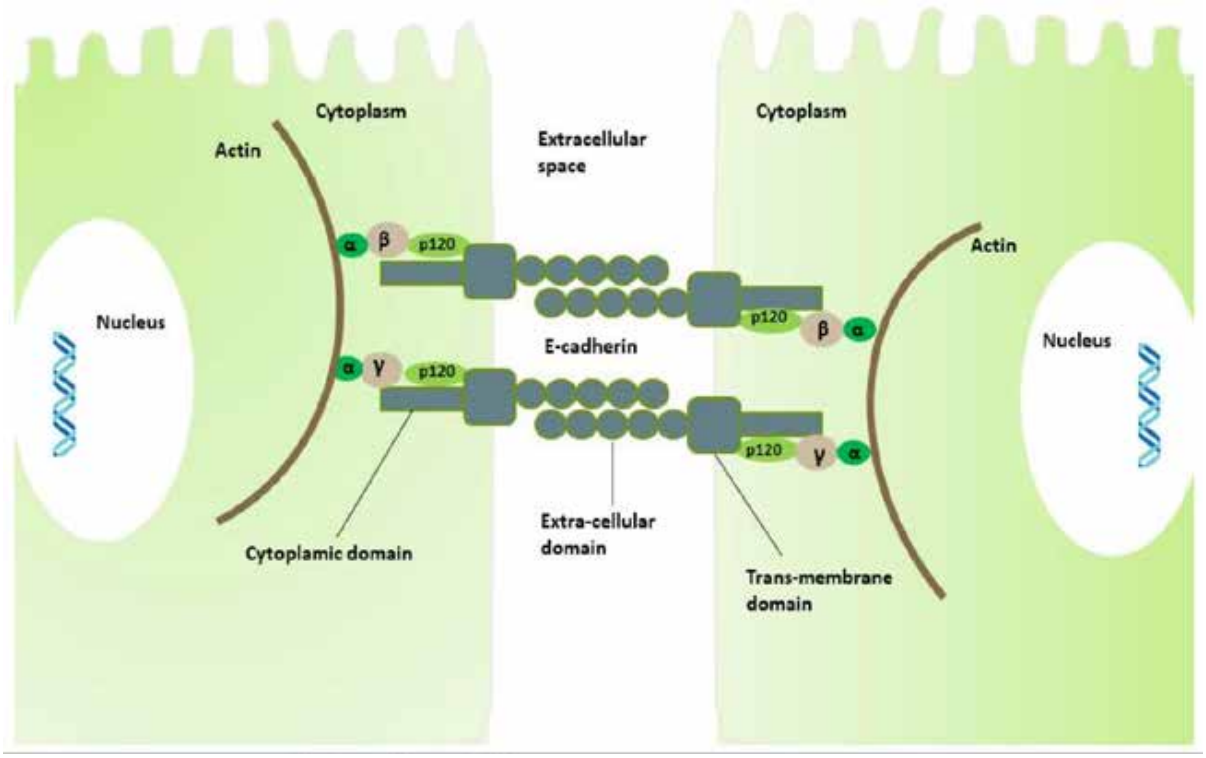
In about 70% of patients, CL/P is an isolated, sporadic disorder transmitted as a multifactorial trait, in which genetic variants and environmental and socioeconomic factors contribute incrementally to the overall risk. Genome-wide association studies have implicated 40 genomic loci in the etiology of CL/P, each with a relatively small effect size. Recurrence risks for future affected offspring of individuals with multifactorial CL/P are 2–5% based on empiric data, with the lowest risks for males with sporadic unilateral CL/P and the highest for females with sporadic bilateral CL/P.

As in our patient and her family, CL/P may also occur as part of a monogenic syndrome. In these cases, the recurrence risk can be much higher, up to 50%. Over 400 Mendelian traits are known to include oral clefts. In order to detect syndromic clefting disorders, a thorough assessment is warranted, especially when the family history is positive for CL/P or when the proband is a girl with bilateral CL/P, the rarest combination in the multifactorial group.

Figure 1. (available for public use, taken from Ansari et al. (2) Cell-cell adhesion occurs when extracellular domains of e-cadherin molecules from neighboring cells bind to each other in a trans manner.



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Heterozygous pathogenic variants in *CDH1* have been implicated in both nonsyndromic cleft lip with or without palate (NS-CL/P) and in the cancer predisposition syndrome, Hereditary Diffuse Gastric Cancer syndrome (HDGC, OMIM 137215)(1). *CDH1* encodes E-cadherin (E for Epithelial), a transmembrane calcium ion-dependent cell adhesion molecule that is the principal adhesion protein in epithelial adherens junctions. Its dimers promote cell-cell adhesion by homophilic binding between the extracellular domains protruding from neighboring cells. Five extracellular domains (EC1-5) are separated by calcium-binding hinge regions. Adjacent E-cadherin molecules from the same cell can bind in a cis manner, and E-cadherin molecules from neighboring cells can bind in a trans manner (See Figure 1).

Dysregulation of E-cadherin affects cell adhesion in the adherens junction and plays a role in cleft lip and palate etiology. E-cadherin is expressed early in embryogenesis during the critical lip and palate development stages in susceptible tissues (3). It is present in the frontonasal prominences at 4 and 5 weeks of gestation and lateral and medial nasal prominences at six weeks. Alvizi and colleagues (4) showed that loss-of-function variants in E-cadherin impair cell migration in human and *Xenopus* neural crest cells. In whole exome sequencing studies of multigeneration NS-CL/P families, Cox et al. (5) identified pathogenic or likely pathogenic variants in 5 functionally linked genes that encode proteins that regulate the epithelial adhesion complex, including *CDH1*. They found missense variants in 5 out of 6 *CDH1*-positive NS-CL/P families.

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Germline variants in *CDH1* were first reported as the cause of Hereditary Diffuse Gastric Cancer (HDGC) in 1998 (6). In 2006, Frebourg et al. published the first report on CL/P in two families with HDGC syndrome (3). The first family (family A) included four individuals in two generations with both CL/P and gastric cancer or peritoneal carcinosis and a familial *CDH1* variant at the intron four splicing donor site: c.531+2T>A. The second family (family B) had a different *CDH1* splicing site variant: c.1137G>A. In family B, gastric cancer segregated separately from CL/P and other birth defects: the father and two of his two daughters had gastric cancer, another carrier daughter had a cleft lip, and a carrier son had aplasia cutis and partial acrania. Both variants generated several aberrantly spliced transcripts in affected individuals.

The penetrance for oral clefts is reduced in families with HDGC syndrome. In a series of 299 individuals from 153 families with confirmed pathogenic or likely pathogenic *CDH1* variants who were enrolled in a prospective trial at the National Cancer Institute, Green et al. (7) report that the rate of CL/P was 19% (29/153) among families in which at least one family member had CL/P and 2.7% of the entire cohort (8/299). They did not detect a genotype-phenotype pattern correlated with CL/P in these HDGC families. Results of a methylation-wide association study from Brazil suggest the mechanism for the decreased penetrance of CL/P. Alvizi et al. (8) compared an age-matched Brazilian cohort of 67 NS-CL/P patients and 59 controls. They found significantly more evidence of hypermethylation of the *CDH1* promoter in penetrant (n=8) vs. nonpenetrant carriers (n=7) and noncarrier (n=3) family members in *CDH1*+ families with NS-CL/P (p=0.0112). This suggests that hypermethylation of the *CDH1* promoter acts as a second hit that increases the expression of CL/P in *CDH1*+ carriers.

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Germline variants in *CDH1* cause pleiotropic effects ranging from gastric cancer, hereditary lobular breast cancer without gastric cancer, colorectal cancer, and nonsyndromic CL/P (9). *CDH1* is also associated with a rare syndromic type of oral clefting with dysmorphic features but no gastric cancer. In 2017, Ghoumid et al. (10) reported that variants in *CDH1* and a related gene, *CTNND1*, cause blepharocheilodontic syndrome (BCD, OMIM 119580). BCD is a rare disorder consisting of bilateral cleft lip/palate, eyelid abnormalities, distichiasis (double row of eyelashes) of the upper eyelids, ectropion of the lower eyelids, and ectodermal defects of hair and teeth, such as hypodontia and conical teeth. Other anomalies include syndactyly, imperforate anus, hypothyroidism, dermoid cysts, and neural tube defects. None of the BCD families express gastric or other cancers. These distinct phenotypes suggest a genotype-phenotype correlation, though, at this time, the *CDH1* phenotypes of gastric cancer or CL/P cannot be predicted reliably based on the genotype alone (11).

The *CDH1* variant in our patient, which is a splice site (truncating) variant, is responsible for autosomal dominant HDGC syndrome in her family, causing gastric cancer in her mother and CL/P in herself and her sister. Truncating mutations in *CDH1* are found in 30% to 50% of HDGC cases. Gastric cancer in HDGC syndrome usually occurs at a young age and has a histologically characteristic signet ring cell. *CDH1* heterozygotes have a lifetime risk for gastric cancer (by age 80) estimated to be 70% for men and 56% for women (12). The average age at diagnosis of gastric cancer is 37 years. Females with *CDH1* mutations are at increased risk of about 42% of developing lobular carcinoma of the breast. Other cancers (esp. colon) have been reported.

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Current cancer surveillance recommendations for *CDH1* heterozygotes published by the National Comprehensive Cancer Network (NCCN Guidelines Version 3.2024. Breast, Ovarian, and Pancreatic; Version 1.2024 Gastric Cancer) include:

- Prophylactic total gastrectomy between ages 18 and 40 when there is a positive family history of gastric cancer. A baseline endoscopy is indicated prior to prophylactic total gastrectomy.
- *CDH1* mutation carriers who do not undergo prophylactic gastrectomy should be offered screening every 6–12 months by upper endoscopy with multiple random biopsies.
- For women with *CDH1* mutations, annual mammograms with consideration of tomosynthesis and breast MRI starting at age 30 (or 5–10 years earlier than the youngest breast cancer diagnosis in the family, but no later than age 30).
    - There is insufficient evidence to support risk-reducing mastectomy based on *CDH1* mutation status alone; management should be based on personal risk factors and family history.

Preventative total gastrectomy is recommended, but some patients opt for cancer surveillance. We discussed both options with our patient. Her sister had chosen surveillance with regular endoscopies and gastric biopsies rather than prophylactic

gastrectomy. In either case, these high-risk patients will benefit from the coordinated long-term care and team-based expertise available at comprehensive cancer centers.

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Oral clefts, especially in females, warrant a thorough inquiry of family history for oral clefts, other congenital anomalies, and young-onset cancers, especially gastric cancer. In this case, our patient's mother had not yet developed gastric cancer at the time of her birth, but there was a strikingly positive family history for CL/P in an older sister, offering evidence of a familial disorder. A formal genetic consultation is warranted when there is a positive family history of oral clefts, even when there are no associated anomalies, and the clinical picture appears nonsyndromic. This family offers the lesson that a family history of CL/P may be the first sign of HDGC syndrome, appearing even before the first case of gastric cancer.

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Practical Applications:

1. Understand that 70% of cases of cleft lip with or without palate (CL/P) are sporadic and multifactorial. Most of these affected patients are males with unilateral cleft lip.
2. Recall that females with CL/P, especially females with bilateral cleft lip, are the less commonly affected sex and are more likely to have a genetic etiology for their CL/P.
3. Suspect a genetic etiology when there is a positive family history of oral clefts, especially when the affected first-degree relative (parent, sibling) is female.
4. Recall that oral clefts occur in families with hereditary diffuse gastric cancer (HDGC) syndrome, a cancer predisposition syndrome caused by pathogenic variants in *CDH1*, which encodes E-cadherin, a component of the major cell-cell adhesion process.
    a. Understand that nonsyndromic CL/P and a syndromic form of CL/P, blepharocheilodontic syndrome, can also be caused by pathogenic variants in *CDH1*.
5. Take a detailed family history when an infant has an oral cleft; ask about other relatives with oral clefts, congenital anomalies, and cancer, especially early onset and gastric cancers.
    a. Consider genetic consultation whenever there is a positive family history of oral cleft or early onset gastric or other cancer.

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