Individuals with nonsyndromic orofacial clefts have increased asymmetry of fingerprint patterns

Katherine Neiswanger, Nandita Mukhopadhyay, Shwetha Rajagopalan, Elizabeth J. Leslie, Carla A. Sanchez, Jacqueline T. Hecht, Iêda M. Orioli, Fernando A. Poletta, Javier Enríquez de Salamanca, Seth M. Weinberg, Mary L. Marazita

1 Center for Craniofacial and Dental Genetics, Department of Oral Biology, School of Dental Medicine, University of Pittsburgh, Pittsburgh, Pennsylvania, United States of America, 2 Department of Pediatrics, University of Texas McGovern Medical Center, Houston, Texas, United States of America, 3 Laboratory of Congenital Malformation Epidemiology, Oswaldo Cruz Institute, Rio de Janeiro, Brazil, 4 Center for Medical Education and Clinical Research, Estudio Collaborativo Latino Americano de Malformaciones Congénitas, Buenos Aires, Argentina, 5 Sección de Cirugía Plástica, Hospital Infantil Universitario Niño Jesús, Madrid, Spain, 6 Department of Human Genetics, Graduate School of Public Health, University of Pittsburgh, Pittsburgh, Pennsylvania, United States of America, 7 Department of Anthropology, University of Pittsburgh, Pittsburgh, Pennsylvania, United States of America, 8 Clinical and Translational Science, School of Medicine, University of Pittsburgh, Pittsburgh, Pennsylvania, United States of America

☯ These authors contributed equally to this work.
* Current address: Department of Human Genetics, Emory University, Atlanta, Georgia, United States of America
* knacct@pitt.edu

Abstract

Dermatoglyphic patterns on the fingers often differ in syndromes and other conditions with a developmental component, compared to the general population. Previous literature on the relationship between orofacial clefts—the most common craniofacial birth defect in humans—and dermatoglyphics is inconsistent, with some studies reporting altered pattern frequencies and/or increased asymmetry and others failing to find differences. To investigate dermatoglyphics in orofacial clefting, we obtained dermatoglyphic patterns in a large multiethnic cohort of orofacial cleft cases (N = 367), their unaffected family members (N = 836), and controls (N = 299). We categorized fingerprint pattern types from males and females who participated at five sites of the Pittsburgh Orofacial Cleft study (Hungary, United States of America (Pennsylvania, Texas), Spain, and Argentina). We also calculated a pattern dissimilarity score for each individual as a measure of left-right asymmetry. We tested for group differences in the number of arches, ulnar and radial loops, and whorls on each individual’s hands, and in the pattern dissimilarity scores using ANOVA. After taking sex and site differences into account, we did not find any significant pattern count differences between cleft and non-cleft individuals. Notably, we did observe increased pattern dissimilarity in individuals with clefts, compared to both their unaffected relatives and controls. Increased dermatoglyphic pattern dissimilarity in individuals with nonsyndromic orofacial clefts may reflect a generalized developmental instability.
Introduction

Orofacial clefts are formed by improper or failed fusion of tissues during early development (Fig 1). They are among the most common birth defects worldwide and can present in

Fig 1. Types of nonsyndromic oral facial clefts. Nonsyndromic clefts can involve the lip only (A), both lip and palate (B), or the palate only (C). They range in severity from small lip notches and submucous cleft palates to the severe case shown in (B). CL = cleft lip; CP = cleft palate.
isolation or be syndromic [1]. Nonsyndromic cleft lip with or without cleft palate (CL/P) has birth prevalence rates ranging from a high of 1/500 in Asian and Native American populations to lower prevalences of 1/2500 in Africans, while the prevalence of nonsyndromic cleft palate only (CPO) is about half that of CL/P worldwide [1,2]. CL/P and CPO are both complex multifactorial traits, controlled by multiple genes and environmental factors [1–6].

The frequencies of dermatoglyphic patterns on the fingers and palms have been studied for many years as potentially sensitive indicators of generalized prenatal developmental delay or instability [7–10]. Because there exists an embryological overlap in the chronologies of the formation of the lip and palate in utero (4th–9th week) [11,12] and dermatoglyphics (6th–24th week) [13–16], numerous studies have examined dermatoglyphic traits in nonsyndromic clefting in multiple populations. Many have reported altered frequencies of dermatoglyphic patterns [16–30] or increased pattern asymmetry [22,24,27,28,31–36] between cleft cases, unaffected relatives, and/or controls. However, results have been inconsistent, with other studies reporting no dermatoglyphic differences in individuals with orofacial clefts [37–41].

The Pittsburgh Oral Facial Cleft study (POFC) began in 1993 and aims to identify genes for nonsyndromic orofacial clefting through a number of strategies, including deep phenotyping of affected cases, their unaffected relatives, and controls from around the world [42,43]. We hypothesize that, due to the shared embryological chronology between the formation of the lip/palate and fingerprints in the first trimester, individuals with nonsyndromic clefts may show altered dermatoglyphic pattern counts and/or increased pattern asymmetry, when compared to individuals without clefts. Our aim is to test these two hypotheses in the POFC sample by ANOVA analysis of dermatoglyphic pattern counts and asymmetry among individuals with clefts, their unaffected relatives, and controls.

Materials and methods

The University of Pittsburgh Institutional Review Board approved this study (IRB protocol numbers STUDY19030367 and STUDY19090156). Written consent was obtained from all participants.

Sample characteristics

The POFC study focuses on identifying genes for nonsyndromic clefting [5,43]. Collaborating clinicians at hospitals, cleft centers, and field sites worldwide refer their nonsyndromic cleft patients and families to the study, or screen potential participants at cleft surgery clinics, to exclude individuals with suspected syndromes, e.g., Van der Woude. Control families and individuals are recruited through advertisements and word of mouth. They are screened for the absence of any cleft, and have no known history of cleft in their first- or second-degree relatives. After obtaining written informed consent from the University of Pittsburgh IRB and other international IRBs or Institutional Ethics Committees, trained research staff collect several additional phenotypes. From 1999–2011, dermatoglyphic prints were collected at five sites internationally (Hungary, Pennsylvania in the U.S.A. (USA-PA), Texas in the U.S.A. (USA-TX), Spain, and Argentina). POFC participants from these five sites with 9 or 10 scoreable fingerprints were included in this study, for a total of 1502 individuals (801 female; 701 male).

All forms of nonsyndromic clefting—cleft lip only, cleft lip and palate, and cleft palate only—were included in this analysis. POFC initially concentrated on nonsyndromic cleft lip.

With or without cleft palate, so there are comparatively few people with cleft palate only. Since alterations in fingerprints are more likely to reflect generalized early developmental instability, as opposed to specific mechanisms of lip or palate formation, we combined individuals with all types of cleft in this analysis. The POFC study is enriched for multiplex cleft
families, i.e. those with more than one affected family member, and the sample includes all available family members, with or without a cleft, as well as multiple family members from the control families. [43]. We divided study participants into three groups: 1) Cases– 367 individuals with a nonsyndromic CL/P or CPO; 2) Unaffected Family Members (UFMs)– 836 individuals from case families who did not have a cleft; and 3) Controls– 299 individuals from control families who were recruited at the Hungary and USA-PA sites only.

**Fingerprint collection**

We took fingerprints using the standard ink method [44,45]. Three trained raters each scored all the patterns on every finger as arch (A), ulnar loop (UL), radial loop (RL), whorl (W), or accidental/other uncommon print. All prints that were not A, UL, RL, or W were grouped into a single “Other” (O) category. If the three initial scores did not agree, a fourth, experienced rater re-evaluated the print. Spot-checks were also performed on sub-sets of prints. Only individuals with 9–10 ratable prints were included in this analysis. See Fig 2 for examples of fingerprint patterns from our participants.

**Pattern count ANOVA**

We used analysis of variance (ANOVA) to model the joint effects of recruitment site, sex, cleft status, and their interactions on the dermatoglyphic pattern count, defined as the number of common patterns (i.e., A, UL, RL, W) present on the hands of the 1502 individuals in the sample. Each individual was assigned a four-part pattern count score. For example, a score of (5,3,1,1) shows that this person has 5 UL, 3 W, 1 A, and 1 RL on their hands. People can have extreme scores of (10,0,0,0), (0,10,0,0), (0,0,10,0), or different combinations of pattern types. This composite variable totaled 10 for most people, but could total 9 or fewer for those people who were missing a print and/or had rare prints.

ANOVA was run on the full model that included the effects of site, sex, cleft status + interactions on dermatoglyphic pattern count, and compared to all possible nested models, i.e., models that excluded one or more of the independent variables and their interactions. The most parsimonious model, determined by the Akaike Information Criterion (AIC), was selected as the best fitting model. For those variables with more than two categories (e.g., five sites and three cleft statuses), we also used the Tukey’s post hoc Honest Significant Differences (HSD) test to determine which sites or cleft statuses were driving the association. The significance threshold for p values was set to 0.05. All statistical analyses were conducted using the R statistical package [46].

Because the cleft status variable did not include controls at three of the sites, we performed two ANOVAs on slightly different samples. First, we used the “All Sites model,” which included all five sites, but only cases and UFMs for the cleft status variable. Second, we ran the “Hungary + USA-PA model,” which included only the Hungary and USA-PA sites, but included all three cleft statuses—cases, UFMs, and controls.

**Pattern dissimilarity ANOVA**

Next, we examined whether patterns differed between the left and right hands according to site, sex, and/or cleft status. For this analysis, patterns were assigned to one of four different types—arches, loops (ulnar and radial loops collapsed into one type), whorls, and other patterns (accidentals and other rare patterns collapsed into one type). Only study participants with known patterns on all ten digits were included (N = 1,476). Pattern asymmetry between right and left hands was determined by calculating a dissimilarity score for each individual, following [35]. For each pair of digits, e.g., right and left thumb, we assigned a score of ‘0’ if the pattern type was the same on both digits, and a score of ‘1’ if the pattern types did not match.
We then summed the digit scores over all five pairs of digits. An individual’s dissimilarity score could range from 0 (all 5 digit pairs with matching pattern types) to 5 (all 5 pairs of digits with dissimilar pattern types).

As done previously, we conducted ANOVA for the full and nested models including site, sex, cleft status, and their interactions, followed by Tukey’s post hoc HSD test for pairwise differences between sites and cleft statuses. The significance threshold for p values was set to 0.05. All statistical analyses were conducted using the R statistical package [46]. ANOVAs were run separately for the “All Sites Model” and the “Hungary + USA-PA Model.”

Results

Sample demographics and pattern frequencies

To describe the sample, Tables 1 and 2 provide the number of individuals and the number of common patterns, respectively, by site, sex, and cleft status. Table 1 shows that in the complete
sample of 1,502 individuals, the overall male-to-female ratio is close to equal (701 male vs. 801 female (47% vs. 53%)). However, the male-to-female ratios within each cleft status are more skewed. There is a preponderance of males among the cleft cases (210 male vs. 157 female (57% vs. 43%)), whereas there are more females than males among the UFMs (377 male vs. 459 female (45% vs. 55%)), and the controls (114 male vs. 185 female (38% vs. 62%)). We observed these sex differences in cleft status for the sites taken separately as well.

Table 2 provides the frequencies of the common dermatoglyphic patterns by site, sex, and cleft status. The 1,502 individuals contributed 14,893 common patterns to Table 2, with 101 rare patterns and 26 missing prints not included. In Table 2, UL is the most frequent pattern, followed by W, A, and RL. Visual inspection of Table 2 suggests that the frequencies of common dermatoglyphic patterns vary by site, sex, and/or cleft status.

### Pattern count ANOVA—Effects of site, sex, and cleft status

The dermatoglyphic pattern counts per individual showed the same general trends as the pattern frequencies in Table 2. UL are present on an average of at least six fingers, with W averaging 2–3.5 fingers, and A and RL averaging less than one finger per individual. Even so, there are a few individuals in the sample with ten arches on their fingers.

Table 3 provides the results of the ANOVA analyses for those nested models which had a p value ≤ 0.05. For each pattern type and model, Table 3 summarizes the most optimal models based on AIC, including overall and pairwise p values, observed group means, and estimated pairwise group differences plus 95% confidence intervals for the pairwise comparisons. In the All Sites Model, for example, the categorical site variable represents five sites; therefore, its overall p value, if significant, is included in addition to the significant pairwise site differences from the Tukey HSD test. Similarly, test outcomes for the cleft status variable, which has three categories in the Hungary + USA-PA Model, include both pairwise and overall test outcomes that are significant.

Arches did not show any significant pattern count differences by site, sex, or cleft status. For UL, RL and W, we observed significant count differences depending on both recruitment site and sex. Site differences were most significant for W (p < 0.001) using the All Sites Model, but were also significant for UL and RL. The Spanish sample had significantly more UL than...
Hungary (6.76 vs. 6.06; p = 0.03), and significantly fewer W (1.77 vs. 2.90; p < 0.001). Even though the average overall RL count is < 1 per individual, there were significantly more RL in the USA-PA sample than in Hungary (0.56 vs. 0.41) for both the All Sites Model (p = 0.003) and for the Hungary-USA-PA Model (p < 0.001).

The all-sites ANOVA also found significant sex differences for UL, RL, and W, although they were not as pronounced as the site differences. Females had significantly more UL than males (6.34 vs. 6.08; p = 0.02), while males had significantly more W (2.79 vs. 2.54; p = 0.03)

Table 2. Number of common patterns by site, sex, and cleft status.

| Site     | Cleft Status | N   | Number (%) of Pattern Types |
|----------|--------------|-----|-----------------------------|
|          |              |     | A   | UL | RL | W   |
| Hungary  | Cases        | 163 | 95 (6%) | 952 (59%) | 73 (5%) | 491 (30%) |
|          | Male         | 91  | 41 (5%) | 547 (61%) | 37 (4%) | 275 (31%) |
|          | Female       | 72  | 54 (8%) | 405 (57%) | 36 (5%) | 216 (30%) |
|          | UFM          | 392 | 204 (5%) | 2394 (62%) | 154 (4%) | 1130 (29%) |
|          | Male         | 174 | 74 (4%) | 1033 (60%) | 80 (5%) | 534 (32%) |
|          | Female       | 218 | 130 (6%) | 1361 (63%) | 74 (3%) | 596 (28%) |
|          | Controls     | 124 | 60 (5%) | 770 (63%) | 49 (4%) | 348 (28%) |
|          | Male         | 47  | 32 (7%) | 280 (60%) | 19 (4%) | 135 (29%) |
|          | Female       | 77  | 28 (4%) | 490 (64%) | 30 (4%) | 213 (28%) |
| USA-PA   | Cases        | 81  | 51 (6%) | 479 (59%) | 45 (6%) | 231 (29%) |
|          | Male         | 45  | 22 (5%) | 257 (57%) | 25 (6%) | 144 (32%) |
|          | Female       | 36  | 29 (8%) | 222 (62%) | 20 (6%) | 87 (24%) |
|          | UFM          | 204 | 123 (6%) | 1334 (66%) | 125 (6%) | 445 (22%) |
|          | Male         | 91  | 47 (5%) | 557 (62%) | 73 (8%) | 227 (25%) |
|          | Female       | 113 | 76 (7%) | 777 (69%) | 52 (5%) | 218 (19%) |
|          | Controls     | 175 | 101 (6%) | 1078 (62%) | 88 (5%) | 472 (27%) |
|          | Male         | 67  | 37 (6%) | 426 (64%) | 38 (6%) | 166 (25%) |
|          | Female       | 108 | 64 (6%) | 652 (61%) | 50 (5%) | 366 (29%) |
| USA-TX   | Cases        | 43  | 30 (7%) | 287 (67%) | 17 (4%) | 92 (22%) |
|          | Male         | 29  | 21 (7%) | 197 (69%) | 11 (4%) | 58 (20%) |
|          | Female       | 14  | 9 (6%) | 90 (65%) | 6 (4%) | 34 (24%) |
|          | UFM          | 152 | 81 (5%) | 957 (63%) | 63 (4%) | 408 (27%) |
|          | Male         | 70  | 29 (4%) | 435 (63%) | 34 (5%) | 198 (28%) |
|          | Female       | 82  | 52 (6%) | 522 (64%) | 29 (4%) | 210 (26%) |
| Spain    | Cases        | 35  | 31 (9%) | 246 (71%) | 19 (6%) | 49 (14%) |
|          | Male         | 18  | 16 (9%) | 128 (72%) | 11 (6%) | 23 (13%) |
|          | Female       | 17  | 15 (9%) | 118 (71%) | 8 (5%) | 26 (16%) |
|          | UFM          | 82  | 76 (9%) | 545 (67%) | 37 (5%) | 158 (19%) |
|          | Male         | 38  | 39 (10%) | 214 (57%) | 22 (6%) | 103 (27%) |
|          | Female       | 44  | 37 (6%) | 331 (76%) | 15 (3%) | 55 (13%) |
| Argentina| Cases        | 45  | 20 (5%) | 254 (57%) | 14 (3%) | 156 (35%) |
|          | Male         | 27  | 15 (6%) | 155 (58%) | 7 (3%) | 89 (33%) |
|          | Female       | 18  | 5 (3%) | 99 (56%) | 7 (4%) | 67 (38%) |
|          | UFM          | 6   | 0 (0%) | 38 (63%) | 0 (0%) | 22 (37%) |
|          | Male         | 4   | 0 (0%) | 29 (72%) | 0 (0%) | 11 (28%) |
|          | Female       | 2   | 0 (0%) | 9 (45%) | 0 (0%) | 11 (55%) |

N = Number of individuals from which pattern frequencies are derived; A = Arch; UL = Ulnar Loop; RL = Radial Loop; W = Whorl; UFM = Unaffected Family Member

https://doi.org/10.1371/journal.pone.0230534.t002
and RL (0.51 vs. 0.41; p = 0.006). Thus, individual pattern counts differ by both site and sex in this data set.

After taking site and sex differences into account, the mean number of UL, RL, and W per individual did not differ significantly by cleft status in either the All Sites sample or the Hungary-USA-PA sample. However, for both UL and RL, there appears to be a significant interaction between cleft status and sex in the UFM sample. Across all five sites, female UFM have a higher mean number of UL than male UFM (mean UL females vs. males: 6.54 vs. 6.03; p = 0.02), and a lower frequency of RL (mean RL females vs. males: 0.38 vs. 0.56; p = 0.001). We observed the RL association in the Hungary-USA-PA sample as well (mean RL female UFM vs. males: 0.38 vs. 0.58; p = 0.01). The addition of controls in the latter analysis did not change the results.

### Pattern dissimilarity ANOVA—Effects of site, sex, and cleft status

Table 4 provides the mean dissimilarity scores by site, sex, and cleft status for the 1476 individuals with complete sets of 10 readable prints, and Table 5 gives the results of the ANOVA. Under the All Sites Model, we observed a significant increase in the mean dissimilarity score...
of cleft cases compared to UFMs (1.26 vs 1.08; p = 0.01). This observation was consistent within each site and within males and females, although the individual comparisons did not reach significance. For the Hungary-USA-PA Model, individuals with clefts showed significantly more dissimilarity than either the UFMs (mean dissimilarity score cases vs. UFMs: 1.30 vs. 1.08; p = 0.04) or the controls (mean dissimilarity score cases vs. controls: 1.30 vs. 1.08; p = 0.03). For the All Sites Model, males also had significantly higher dissimilarity scores than females (1.20 vs. 1.06; p = 0.02). This difference was observed within all cleft statuses, although it was not significant. No significant sex effects were observed in the Hungary-USA-PA model.

In contrast to the pattern count analysis, cleft status was a significant predictor of pattern dissimilarity scores. This is evident in Table 4, which shows the mean pattern dissimilarity scores for different sites, sexes, and cleft statuses.

### Table 4. Mean pattern dissimilarity scores (N) by site, sex, and cleft status.

| Site          | Cleft Status | Cases     | UFMs       | Controls  | Total      |
|---------------|--------------|-----------|------------|-----------|------------|
| Hungary       |              | 1.30 (162)| 1.17 (382) | 1.04 (120)| 1.17 (664) |
|               | Male         | 1.23 (91) | 1.21 (169) | 1.00 (47) | 1.18 (307) |
|               | Female       | 1.39 (71) | 1.14 (213) | 1.07 (73) | 1.17 (357) |
| USA-PA        |              | 1.31 (81) | 1.02 (202) | 1.12 (173)| 1.11 (456) |
|               | Male         | 1.38 (45) | 1.07 (90)  | 1.27 (66) | 1.20 (201) |
|               | Female       | 1.22 (36) | 0.98 (112) | 1.02 (107)| 1.03 (255) |
| USA-TX        |              | 1.26 (43) | 1.03 (152) | -          | 1.08 (195) |
|               | Male         | 1.38 (29) | 1.10 (70)  | -          | 1.18 (99)  |
|               | Female       | 1.00 (14) | 0.96 (82)  | -          | 0.97 (96)  |
| Spain         |              | 1.00 (34) | 0.90 (80)  | -          | 0.93 (114) |
|               | Male         | 1.22 (18) | 1.18 (38)  | -          | 1.20 (56)  |
|               | Female       | 0.75 (16) | 0.64 (42)  | -          | 0.67 (58)  |
| Argentina     |              | 1.24 (41) | 1.00 (6)   | -          | 1.21 (47)  |
|               | Male         | 1.48 (25) | 0.75 (4)   | -          | 1.38 (29)  |
|               | Female       | 0.88 (16) | 1.50 (2)   | -          | 0.94 (18)  |
| All sites     |              | 1.26 (361)| 1.08 (822) | 1.09 (293)| 1.12 (1476)|
|               | Male         | 1.31 (208)| 1.15 (371) | 1.16 (113)| 1.20 (692) |
|               | Female       | 1.20 (153)| 1.02 (451) | 1.04 (180)| 1.06 (784) |

N = Number of individuals in each group that contribute to the mean dissimilarity score; UFMs = Unaffected Family Members

The pattern dissimilarity scores were also analyzed using ANOVA models. Table 5 summarizes the optimal models and their P values.

### Table 5. Pattern dissimilarity scores—ANOVA optimal models with P values ≤ 0.05.

| Optimal Model | P Value | Observed Group Means Gp1, Gp2 | Estimated Group Difference and [95% CI] |
|---------------|---------|-------------------------------|----------------------------------------|
| **All Sites Model:** Sex + Cleft Status |         |                               |                                        |
| Sex (Male–Female) | 0.02    | 1.20, 1.06                    | 0.14 [0.02–0.26]                        |
| Cleft Status     | 0.01    |                               |                                        |
| Case–UFM²        | 0.01    | 1.26, 1.08                    | 0.16 [0.03–0.28]                        |
| **Hungary + USA-PA Model:** Cleft Status |         |                               |                                        |
| Cleft Status     | 0.02    |                               |                                        |
| Case–Control²    | 0.03    | 1.30, 1.08                    | 0.22 [0.01–0.43]                        |
| Case–UFM²        | 0.04    | 1.30, 1.08                    | 0.19 [0.01–0.37]                        |

¹Based on AIC. For each pairwise test, group1 is on the left and group2 is on the right in columns 1 and 3
²Tukey HSD Pairwise Group
GP1 = Group 1; GP2 = Group 2; CI = Confidence Interval; UFMs = Unaffected Family Members

https://doi.org/10.1371/journal.pone.0230534.t005

Individuals with nonsyndromic orofacial clefts have increased asymmetry of fingerprint patterns...
dissimilarity between left and right hands, along with sex, while there were no observed differences due to recruitment site, nor any interaction effects.

**Discussion**

We analyzed fingerprint patterns from 1502 individuals from the POFC Study, to determine if individuals with clefts have altered pattern counts on their hands or increased pattern type asymmetry, compared to their relatives without clefts or to controls. In general, our results revealed little evidence that cleft status was associated with differences in the number of common patterns on an individual’s hands. However, we did find evidence of increased left-right pattern dissimilarity in affected cleft cases, particularly if they were male.

**Differences in pattern frequency by recruitment site and sex**

Dermatoglyphic pattern frequencies vary extensively by race and ethnicity [7, 47], and we were not surprised to observe site-specific differences in our sample. Most notably, the overall frequency of whorls in the sample from Spain (18%) was low while the frequencies of ulnar loops (68%) and arches (9%) were high, compared to the other sites, in particular Hungary and Argentina. As demonstrated in Table 6, our Spanish frequencies are relatively extreme, even when compared to other published samples taken from specific regions of Spain [48–50]. Many of the Spanish families in our sample traveled some distance to participate in POFC, and thus they probably do not reflect any single ethnic or regional population. Furthermore, the Spanish sample size of 117 is not exceptionally small. Although fingerprint patterns often vary significantly between relatively close geographical regions and similar ethnicities (see Table 6), the Spanish samples in this study remain unusual, compared to other sites in our study and to other published Spanish samples.

The sample had different proportions of males and females in the cleft cases, UFMs, and controls, with 57% males in the cleft cases, 45% males in the UFMs, and 38% males in the controls. This is not unexpected, since nonsyndromic cleft lip with or without cleft palate is more common in males [11,51], while controls, both family derived and unrelated, often include more females. Dermatoglyphic patterns vary between males and females in general [44,52]. We observed an increase in ulnar loops in females, and an increase in radial loops in males, a trend seen in other studies of Spain (Table 6 and [53]). Thus, we included both site and sex as covariates in the analysis.

**Little evidence of association between clefting and specific fingerprint patterns**

Fingerprint patterns are formed in early development, overlapping the period of lip and palate formation, and different pattern frequencies may reflect changes in developmental timing [13]. Arches, for example, are considered to be slower forming patterns, and may be seen more frequently in conditions involving developmental delay [54]. Frequencies of dermatoglyphic patterns have been studied in individuals with clefts and their family members or controls for decades. In the majority of reports, the frequency of whorls is decreased in individuals with nonsyndromic clefts and/or their relatives, compared to controls, while the corresponding frequencies of ulnar/radial loops and/or arches is increased, although not all differences are significant or occur in both sexes. This is to be expected, since by definition, a decreased count for one pattern will lead to increased counts in some of the other patterns. These trends have been observed in studies of Japanese [17,23,30], Austrians [29], Indians [16, 18–20,25,26], Israelis [24], Filipinos [28], and Iranians [21,22]. Other studies, however, have failed
to replicate these findings, including studies of U.S. Caucasians [41], Belgians [37], Chinese [39], and Indians [38,40].

This study, one of the largest to date, failed to confirm the observation seen in the majority of studies—that whorls are less frequent in individuals with clefts, while loops and/or arches are more frequent. Because the frequencies of both clefting and dermatoglyphic patterns are known to vary significantly in different ethnic groups and by sex, we performed an ANOVA that took all of these factors into account jointly. The analysis compared a full model (site, sex, and cleft status variables) to nested models, and determined that site and sex accounted for most of our observed differences in pattern counts, while cleft status did not. We did observe an interaction between cleft status and sex in the case of radial and ulnar loops. Unaffected male relatives have more radial loops, while unaffected female relatives have more ulnar loops than expected. The biological significance of this observation is not clear, but the implication is that in families with nonsyndromic clefting, the process by which a loop becomes lateralized differs between males and females, but only in individuals who do not have a cleft.

Earlier studies have not taken our analytical approach, so it is possible that not all studies have adequately controlled for the effects of ethnic variation and/or sex. The consistent nature

| Site                                          | Sex   | N  | Pattern Type Frequency (%) | A    | UL  | RL  | W    |
|-----------------------------------------------|-------|----|----------------------------|------|-----|-----|------|
| Spain (current study)                         |       |    |                            |      |     |     |      |
| Male                                          | 56    |    | 9.89                       | 61.51| 5.93| 22.66|      |
| Female                                        | 61    |    | 8.60                       | 74.21| 3.80| 13.39|      |
| Alberche/Tormes Valley [50]                   |       |    |                            |      |     |     |      |
| Male                                          | 187   |    | 3.32                       | 58.82| 4.12| 33.74|      |
| Female                                        | 219   |    | 4.29                       | 61.19| 3.06| 31.46|      |
| Basque Alava Region, Llanada population [48]  |       |    |                            |      |     |     |      |
| Male                                          | 99    |    | 2.13                       | 62.01| 4.04| 31.82|      |
| Female                                        | 82    |    | 4.87                       | 61.58| 2.68| 20.86|      |
| Basque Guipuzcoa Region, Goierri population   |       |    |                            |      |     |     |      |
| Male                                          | 100   |    | 5.40                       | 62.70| 4.80| 27.10|      |
| Female                                        | 101   |    | 3.37                       | 65.25| 4.15| 27.23|      |
| Basque Navarra Region, Baztan population      |       |    |                            |      |     |     |      |
| Male                                          | 92    |    | 6.52                       | 61.96| 5.87| 25.65|      |
| Female                                        | 66    |    | 5.15                       | 71.06| 4.09| 19.69|      |
| Basque Vizcaya Region, Markina population     |       |    |                            |      |     |     |      |
| Male                                          | 93    |    | 7.31                       | 59.78| 4.52| 28.39|      |
| Female                                        | 198   |    | 7.58                       | 61.41| 3.84| 27.17|      |
| Tierra de Campos [49]                         |       |    |                            |      |     |     |      |
| Male                                          | 417   |    | 3.95                       | 60.48| 4.17| 31.03|      |
| Female                                        | 416   |    | 6.06                       | 61.70| 3.73| 28.47|      |
| La Alcarria [49]                              |       |    |                            |      |     |     |      |
| Male                                          | 339   |    | 5.13                       | 60.56| 4.84| 29.47|      |
| Female                                        | 314   |    | 8.28                       | 64.08| 3.47| 24.17|      |
| Murcia [49]                                   |       |    |                            |      |     |     |      |
| Male                                          | 163   |    | 5.23                       | 60.47| 4.60| 28.11|      |
| Female                                        | 184   |    | 8.75                       | 65.90| 4.28| 21.06|      |

N = Number of males and females at each site; A = Arch; UL = Ulnar Loop; RL = Radial Loop; W = Whorl

https://doi.org/10.1371/journal.pone.0230534.t006
and direction of the majority of findings in the literature (increased loops/arches and decreased whorls in clefing) lends strength to the hypothesis that dermatoglyphic pattern frequencies reflect a developmental delay or perturbation during the formation of the lip and palate that increases the risk of a nonsyndromic cleft. Since our sample sizes are relatively large, this would imply either that these prior studies were flawed, perhaps by low power due to small sample sizes, or that our analysis was overly conservative.

**Fingerprint pattern asymmetry increased in cleft cases**

The embryologic development of many bilaterally symmetric anatomic features is presumed to be identical on the left and right sides, whether such features are influenced by genes, the prenatal environment, or some combination of both. This study observed a significant increase in dermatoglyphic asymmetry—measured by a pattern-type dissimilarity score—in individuals with clefts compared to UFMs and controls. We observed an independent increase in pattern dissimilarity in males compared to females. These results were consistent across most sites, indicating that both cleft status and sex are associated with greater dermatoglyphic asymmetry in our sample. However, in Hungary the female cases have more dissimilarity than the male cases, which may explain why the sex effect was not significant in the Hungary-USA-PA model.

Increased asymmetry in multiple dermatoglyphic traits taken from either fingers or palms has been widely reported in different groups of individuals, with or without clefts. Some studies find increased dermatoglyphic asymmetry in individuals with clefts, compared to UFMs and/or controls \[24,33\]. Others report increased dermatoglyphic asymmetry in cleft individuals and/or their UFMs from multiplex cleft families, compared to either simplex families or controls \[31,35,36,39\]. There are reports of increased dermatoglyphic asymmetry in parents of individuals with clefts, compared to controls \[22 (mothers only), 32 (mothers only), 33,34\]. In contrast, \[28\] reported increased dermatoglyphic asymmetry in female unaffected relatives, compared to cases. Taken together, these studies suggest that dermatoglyphic asymmetry reflects a generalized developmental disturbance that also impacts cleft formation. More specifically, there is evidence of shared genetic mechanisms between limb or appendage developmental processes and cleft formation. For example, a number of cleft-related syndromes include limb defects as well (OMIM search term = "cleft lip/palate and limb"), and a recent GWAS study of human facial shape reported that associated loci were enriched for genes involved in limb development \[55\]. Whether both phenotypes are the result of mutations in genes for the robustness of early embryonic development or reflections of prenatal environmental insults or an interaction between the two remains to be determined.

**Limitations**

Even though our sample sizes are large, recruiting from multiple sites with different racial backgrounds introduced ethnicity as a potential confounder, which could have reduced the power of the sample. Likewise, the decision to define cleft status broadly as CL and/or CP might have introduced additional heterogeneity into the sample, which could also reduce power. Another consideration is that our study sample contains related individuals. Cleft statuses of related individuals may be correlated due to shared genetic as well as environmental factors, the degree of correlation being highest for first-degree relatives (parents, siblings and children). We verified, prior to analysis, that the number of first-degree relatives included from each family is small (2 or 3 for nearly all pedigrees), and that there are no large clusters of correlated subjects in any sub-group. We therefore concluded that the likelihood of observing spurious significant associations due to this correlation is small. The analysis was designed to
take these factors into account, but it may have missed small difference in pattern counts, which have been reported in other studies of dermatoglyphics and nonsyndromic clefting. This is countered by the significant laterality differences, in which individuals with clefts showed increased pattern dissimilarity, regardless of recruitment site or sex, showing that there is ample power in this sample for that phenotype. In any event, additional studies in large, well-characterized samples are warranted.

**Conclusions**

In this study of nonsyndromic clefting and dermatoglyphic patterns, there were no significant differences in pattern counts for individuals with clefts, their unaffected relatives, or controls. However, individuals with clefts had significantly more dermatoglyphic asymmetry in their pattern types, as measured by a dissimilarity score, than unaffected relatives or controls. These results lend support to the idea that generalized developmental disturbances acting early in pregnancy may increase the risk of orofacial clefting.

**Supporting information**

S1 Data.
(XLSX)

**Acknowledgments**

We thank our colleagues both within and outside the U.S. who have dedicated themselves to collecting these data. In particular, Drs. Andrew E. Czeizel, (Foundation for the Community Control of Hereditary Diseases, Budapest, Hungary), and Eduardo E. Castilla (Center for Medical Education and Clinical Research, Buenos Aires, Argentina and Latin American Collaborative Study of Congenital Malformations, Rio de Janeiro, Brazil), now deceased, collected fingerprints from their cleft family samples and controls over many years. At the University of Pittsburgh Center for Craniofacial and Dental Genetics, Eleanor Feingold, Ph.D., John R. Shaffer, Ph.D., Jenna C. Carlson, Ph.D. (now in the Dept. of Biostatistics at the University of Pittsburgh), Myoung Keun Lee, M.S., Jonathan M. Chernus, M.S, Margaret E. Cooper, M.S, Toshiki Soejima, B.S., Kathy Bardi, M.S.W., Megan Branning, M. L.I.S., and Judith M. Resick, B.S. provided essential help with recruitment, data collection, and fingerprint analysis. We could not have completed study without the many participants throughout the world who have given their time and energies to the Pittsburgh Oral Facial Cleft Study.

**Author Contributions**

**Conceptualization:** Katherine Neiswanger, Shwetha Rajagopalan, Elizabeth J. Leslie, Carla A. Sanchez, Seth M. Weinberg, Mary L. Marazita.

**Data curation:** Nandita Mukhopadhyay, Carla A. Sanchez, Jacqueline T. Hecht, Iêda M. Orioli, Fernando A. Poletta, Mary L. Marazita.

**Formal analysis:** Katherine Neiswanger, Nandita Mukhopadhyay, Shwetha Rajagopalan.

**Funding acquisition:** Jacqueline T. Hecht, Seth M. Weinberg, Mary L. Marazita.

**Investigation:** Katherine Neiswanger, Carla A. Sanchez, Iêda M. Orioli, Fernando A. Poletta, Javier Enríque de Salamanca.

**Methodology:** Katherine Neiswanger, Nandita Mukhopadhyay, Shwetha Rajagopalan, Seth M. Weinberg.
Project administration: Seth M. Weinberg, Mary L. Marazita.

Resources: Jacqueline T. Hecht, Iêda M. Orioli, Fernando A. Poletta, Javier Enríquez de Salamanca, Mary L. Marazita.

Supervision: Katherine Neiswanger, Mary L. Marazita.

Writing – original draft: Katherine Neiswanger, Nandita Mukhopadhyay, Shwetha Rajagopalan.

Writing – review & editing: Katherine Neiswanger, Nandita Mukhopadhyay, Shwetha Rajagopalan, Elizabeth J. Leslie, Carla A. Sanchez, Jacqueline T. Hecht, Iêda M. Orioli, Fernando A. Poletta, Javier Enríquez de Salamanca, Seth M. Weinberg, Mary L. Marazita.

references
1. Leslie EJ, Marazita ML. Genetics of cleft lip and cleft palate. Am J Med Genet C Semin Med Genet. 2013; 163C: 246–258. https://doi.org/10.1002/ajmg.c.31381 PMID: 24124047
2. Dixon MJ, Marazita ML, Beatty TH, Murray JC. Cleft lip and palate: Understanding genetic and environmental influences. Nat Rev Genet. 2011; 12: 167–178. https://doi.org/10.1038/nrg2933 PMID: 21331089
3. Deroo LA, Wilcox AJ, Drevon CA, Lie RT. First-trimester maternal alcohol consumption and the risk of infant oral clefts in Norway: a population-based case-control study. Am J Epidemiol. 2008; 168: 639–646. https://doi.org/10.1093/aje/kwn186 PMID: 18667525
4. Honein MA, Rashid DT, Romiti RM, Lammer EJ, Sun L, et al. Maternal smoking and environmental tobacco smoke exposure and the risk of orofacial clefts. Epidemiol. 2007; 18: 226–233.
5. Leslie EJ, Carlson JC, Shaffer JS, Wehby GJ, Laurie CA, et al. A multi-ethnic genome-wide association study identifies novel loci for non-syndromic cleft lip with or without cleft palate on 2p24.2, 17q23 and 19q13. Hum Mol Genet. 2016; 25: 2862–2872. https://doi.org/10.1093/hmg/ddw104 PMID: 27033726
6. Munger RG, Tamura T, Johnston KE, Feldkamp ML, Phister R, Carey JC. Plasma zinc concentration of mothers and the risk of oral clefts in their children in Utah. Birth Defects Res A Clin Mol Teratol. 2009; 85: 151–155. https://doi.org/10.1002/bdrc.20516 PMID: 19067407
7. Jantz RL. Anthropological dermatoglyphic research. Ann Rev Anthropol. 1987; 16: 161–177.
8. Livshits G, Kobyliansky E. Fluctuating asymmetry as a possible measure of developmental homeostasis in humans: a review. Hum Biol. 1991; 63: 441–466. PMID: 1889795
9. Miller JR. Dermatoglyphics. J Invest Dermatol. 1973; 60: 435–442. https://doi.org/10.1111/1523-1747.ep12702906 PMID: 4351102
10. Naugler CT, Ludman MD. A case-control study of fluctuating dermatoglyphic asymmetry as a risk marker for developmental delay. Am J Med Genet. 1996; 66: 11–14. https://doi.org/10.1002/(SICI)1096-8628(19961202)66:1<11::AID-AJMG3>3.0.CO;2-Z PMID: 8957503
11. Mossey PA, Little J, Munger RG, Dixon MJ, Shaw WC. Cleft lip and palate. Lancet. 2009; 374: 1773–1785. https://doi.org/10.1016/S0140-6736(09)60695-4 PMID: 19747722
12. Smarius B, Loozen C, Manten W, Bekker M, Pistorius L, Breugem C. Accurate diagnosis of prenatal cleft lip/palate by understanding the embryology. World J Medical. 2017; 9: 93–100. https://doi.org/10.5662/wjm.v7.i3.93 PMID: 29026689
13. Babler WJ. Embryologic development of epidermal ridges and their configurations. Birth Defects Orig Artic Ser. 1991; 27: 95–112. PMID: 1786361
14. Holt S. The genetics of dermal ridges. Springfield, IL: Charles C. Thomas; 1968.
15. Seidenberg-Kajabova H, Pospisilova V, Vranakova V, Varga I. An original histological method for studying the volar skin of the fetal hands and feet. Biomed Pap Fac Univ Palacky Olomouc Czech Repub. 2010; 154: 211–218.
16. Mathew L, Hegde AM, Rai K. Dermatoglyphic peculiarities in children with oral clefts. J Ind Soc Pedod Prev Dent. 2005; 23: 179–182.
17. Azumi J, Shiono H. Dermatoglyphic diagnosis of patients with cleft lip, palate. Nihonjiishinpo. 1966; 2900: 43–44.
18. Baligir RS. Congenital oral clefts and dermatoglyphics. Isr J Med Sci. 1984; 20: 622–624. PMID: 6469586
19. Balgir RS. Dermatoglyphics in cleft lip and cleft palate anomalies. Indian Pediatr. 1993; 30: 341–346. PMID: 8365784

20. Deshmukh RN, Grewal MS, Sidhu SS. Cleft lip with or without left palate and isolated cleft palate, as two aetiological entities—a dermatoglyphic evidence. Indian J Med Res. 1981; 73: 584–589. PMID: 7262931

21. Eslami N, Jahanbin A, Ezzati A. Palm and finger print characteristics in nonfamilial cleft lip and palate patients and their parents. J Craniof Surg, 2013; 24: 769–772.

22. Jahanbin A, Mahdavishahri N, Naseri MM, Sardari Y, Rezaian S. Dermatoglyphic analysis in parents with nonfamilial bilateral cleft lip and palate children. Cleft Palate Craniof J. 2010; 47: 9–14.

23. Kanematsu N, Yoshida Y, Kishi N, Kawata K, Kaku M, Maeda K, et al. Studies on abnormalities in the appearance of finger and palm prints in children with cleft lip, alveolus, and palate. J Maxillofac Surg. 1986; 14: 74–82. https://doi.org/10.1016/s0301-0503(86)80265-x PMID: 3457887

24. Kobyliansky E, Bejerano M, Yakovenko K, Katznelson MBM. Relationship between genetic anomalies of different levels and deviations in dermatoglyphic traits. Part 6: dermatoglyphic peculiarities of males and females with cleft lip (with or without left palate) and cleft palate–family study. Coll Antropol. 1999; 23: 1–51. PMID: 10402704

25. Maheshwari N, Bansal K, Rao D, Chopra R. Comparison of dermatoglyphic traits and dental anomalies associated with cleft lip or cleft lip and palate patients with normal healthy children. J Ind Soc Pedod Prev Dent. 2013; 31: 260–264.

26. Saxena RS, David MP, Indira AP. Dermatoglyphic evaluation in subjects and parents of cleft lip with and without cleft palate. Cleft Palate Craniof J. 2013: 50: e105–e110.

27. Scott NM, Weinberg SM, Neiswanger K, Brandon CA, Daacke-Hirsch S, Murray JC, et al. Dermatoglyphic fingerprint heterogeneity among individuals with nonsyndromic cleft lip with or without cleft palate and their unaffected relatives in China and the Philippines. Hum Biol. 2005; 77: 257–266. https://doi.org/10.1353/hub.2005.0042 PMID: 16201141

28. Scott NM, Weinberg SM, Neiswanger K, Daacke-Hirsch S, O’Brien S, Murray JC, et al. Dermatoglyphic pattern types in subjects with nonsyndromic cleft lip with or without cleft palate (CL/P) and their unaffected relatives in the Philippines. Cleft Palate Craniof J. 2005; 42: 362–366.

29. Vormittag W, Weninger M, Hoffmann K, Hoffmann D. Heterogeneity of cleft lip and/or palate and dermatoglyphics. Birth Defects Orig Artic Ser. 1979; 15: 649–659. PMID: 552890

30. Yamagata Y. Dermatoglyphic study of hare lip and cleft palate. Shikoku Acta Medica. 1973; 29: 143–148. Esteban E, Moral P. Finger dermatoglyphics in a Mediterranean population (Murcia, Spain): Pattern types and pattern intensity index. Anthropol Anz. 1993;51: 159–167.

31. Crawford FC, Sosafer JA. Cleft lip with or without cleft palate: identification of sporadic cases with a high level of genetic predisposition. J Med Genet. 1987; 24: 163–169. https://doi.org/10.1136/jmg.24.3.163 PMID: 3572999

32. Leite BDGL Queiroz IN, Aquino SM Machado RA, Paranaiba LMR, Martelli DRB, et al. Evaluating fluctuating asymmetry in a Brazilian population with non-syndromic cleft lip and/or palate. J Plast Surg Hand Surg. 2015; 49: 289–294. https://doi.org/10.1016/j.bjps.2015.10.023 PMID: 25967927

33. Ma H, Qi Y, Zhu W, Chao H, Shi B. Dermatoglyphic features in nonsyndromic cleft lip and/or palate patients and their parents in China. 2014. Cleft Palate Craniof J. 2014; 51: 76–82.

34. Singh P, Nathani DB. Dermatoglyphics and cheiloscopy as key tools in resolving the genetic correlation of inheritance patterns in cleft lip and palate patients: an assessment of 160 patients. Cleft Palate Craniof J. 2017; 54: 588–594.

35. Woolf CM, Gianadas AD. Congenital lip and fluctuating asymmetry. Am J Hum Genet. 1976; 28: 400–403.

36. Woolf CM, Gianadas AD. A study of fluctuating dermatoglyphic asymmetry in the sibs of parents of cleft lip patients. Am J Hum Genet. 1977; 29: 503–507. PMID: 900124

37. DeBlie S, Hayashi M, Matton MT, Matton G, Vrijdagh S, Lejour M, et al. Dermatoglyphic analysis of primary and secondary cleft palate patients. Cleft Palate J. 1977; 14: 222–225. PMID: 267521

38. Mayall SS, Chaudhary S, Kaur H, Manuja N, Ravishankar T, Sinha AA. Comparison of dermatoglyphic pattern among cleft and noncleft children: a cross-sectional study. Int J Clin Pediatr Dent. 2017; 10: 245–249. https://doi.org/10.5005/jp-journals-10005-1444 PMID: 29104983

39. Neiswanger K, Cooper ME, Weinberg SM, Flodman P, Keglovits AB, Liu YE, et al. Cleft lip with or without cleft palate and dermatoglyphic asymmetry: Evaluation of a Chinese population. Orthodont Craniof Res. 2002; 5: 140–146.

40. Seujanya K, Prasad MG, Sushma B, Kumar JR, Reddy YSN, Niranjani K. Cheiloscopy and dermatoglyphic as genetic markers in the transmission of cleft lip and palate: a case-control study. J Ind Soc Pedodont Prevent Dent. 2016; 34: 48–54.
41. Silver WE. Dermatoglyphics and cleft lip and palate. Cleft Palate J. 1966; 3: 368–375. PMID: 5222744
42. Marazita M. Subclinical features in non-syndromic cleft lip with or without cleft palate (CL/P): review of the evidence that subepithelial orbicularis oris muscle defects are part of an expanded phenotype for CL/P. Orthod Craniof Res. 2007; 10: 82–87.
43. Weinberg SM, Neiswanger K, Martin RA, Mooney MP, Kane AA, Wenger SL, et al. The Pittsburgh Oral-Facial Cleft Study: Expanding the cleft phenotype. Background and justification. Cleft Palate Craniof J. 2006; 43: 7–20.
44. Cummins H, Midlo C. Finger prints, palms and soles. Philadelphia: Blakiston; 1943.
45. Schauermann B, Alter M. Dermatoglyphics in medical disorders. New York-Heidelberg-Berlin: Springer-Verlag; 1976.
46. R Core Team. R: A Language and Environment for Statistical Computing. Vienna, Austria: R Foundation for Statistical Computing. 2019. www.R-project.org
47. Meier RJ. Applications of dermatoglyphics to anthropologic populations. Birth Defects Orig Artic Ser. 1991; 27: 253–265. PMID: 1786354
48. Arrieta I, Martı́nez B, Criado B, Te´lez M, Ortega B, Peñagarikano O, et al. Dermatoglyphic variation in Spanish Basque populations. Hum Biol. 2003; 75: 265–291. https://doi.org/10.1353/hub.2003.0029 PMID: 12943163
49. Esteban E, Moral P. Finger dermatoglyphics in a Mediterranean population (Murcia, Spain): Pattern types and pattern intensity index. Anthropol Anz. 1993; 51: 159–167. PMID: 8333736
50. Martı́n J, Mesa MS, Fuster V, Moral P. Dermatoglyphics of inhabitants of Alberche/Tormes valley (Sierra de Gredos—Central Spain): 1. Finger pattern types and pattern intensity. Am J Hum Biol. 1996; 8: 305–316. https://doi.org/10.1002/(SICI)1520-6300(1996)8:3<305::AID-AJHB1>3.0.CO;2-Z PMID: 28557255
51. Martelli DR, Machado RA, Swerts MS, Rodrigues LA, Aquino SM, Martelli-Júnior H. Non syndromic cleft lip and palate: relationship between sex and clinical extension. Braz J Otorhinolaryngol. 2012; 78: 116–120.
52. Buchwald W. The morphological diversity of dermatoglyphic patterns on fingers—A simple and objective method for measurement. Homo. 2015; 66: 60–78. https://doi.org/10.1016/j.jchb.2014.07.001 PMID: 25541233
53. Arquimbau R, Esteban E, Fañabá L. Finger dermatoglyphics in Delta de l'Ebre: a Mediterranean Spanish population. Anthropol Anz. 1993; 51: 267–274. PMID: 8215262
54. Rosa A, Gutiérrez B, Guerra A, Aria B, Fañanas L. Dermatoglyphics and abnormal palmar flexion creases as markers of early prenatal stress in children with idiopathic intellectual disability. J Intellect Disabil Res. 2001; 45: 416–423. https://doi.org/10.1046/j.1365-2788.2001.00351.x PMID: 11679047
55. Claes P, Roosenboom J, White JD, Swigut T, Sero D, Li J, et al. Genome-wide mapping of global-to-local genetic effects on human facial shape. Nat Genet. 2018; 50: 414–423. https://doi.org/10.1038/s41588-018-0057-4 PMID: 29459680