European Journal of Orthodontics 26 (2004) 375–384

European Journal of Orthodontics vol. 26 no. 4

Parental craniofacial morphology in orofacial clefting

G. T. McIntyre and P. A. Mossey
Orthodontic Department, Dundee Dental Hospital and School, UK

SUMMARY The parental craniofacial morphology in orofacial clefting (OFC) has been shown to differ from that of the non-cleft population when evaluated using conventional cephalometric analyses comprising a variety of linear, angular, and area measurements. In spite of this, the shape of the parental craniofacial complex is of greater importance in the search for the morphogenes involved in OFC.

This retrospective case–control study employed three morphometric techniques [discriminant analysis of the principal components of shape (PCS), Euclidean distance matrix analysis (EDMA), and thin-plate spline analysis (TPS)] to localize the craniofacial skeletal shape differences between (a) the parents of children with OFC and a comparison group, (b) the parents of children with cleft lip and palate [CL(P)] and cleft palate (CP), and (c) the male and female parents of children with OFC. The postero-anterior (PA) cephalograms of 92 parents of children with non-syndromic OFC and 43 comparison group volunteers were scanned and digitized. The configurations of 24 reproducible landmarks were optimally superimposed using Procrustes algorithms to allow shape data to be derived using PCS, EDMA, and TPS.

The parental craniofacial shape statistically significantly differed from that of the comparison group using PCS ($P < 0.001$) and EDMA ($P = 0.001$). However PCS, EDMA, and TPS differed in their localization of the shape differences, explainable by the different mathematical methods used by the individual techniques. Interestingly, the parental craniofacial shapes in CL(P) and CP were morphologically similar when tested using PCS ($P = 0.03$) and EDMA ($P = 0.027$). However, there was no shape-related sexual dimorphism in parental craniofacial morphology in OFC when tested using PCS ($P = 0.35$) and EDMA ($P = 0.525$).

Thus, the parental craniofacial shape in OFC differs from the non-cleft population, the parental craniofacial shape does not differ between CL(P) and CP and there is no sexual dimorphism in the parental craniofacial morphology in OFC, as viewed on PA cephalograms.

Introduction

Non-syndromic orofacial clefting (OFC) comprises cleft lip with or without cleft palate [CL(P)] and isolated cleft palate (CP). The aetiology of OFC is considered to be polygenic and multifactorial, with the relative contributions from genetic and environmental sources varying between cases. The genetic contribution will be minimal in some cases, heavily weighted to one parent in other cases, and approximately equal where, by chance, each parent happens to possess the same degree of predisposing factors (Ward *et al.*, 1989).

Embryonic craniofacial morphology influences the development of the orofacial region and OFC. Specifically, a wider face and head may prevent palatal shelf contact (Fraser and Pashayan, 1970). There is a high correlation in parent–child craniofacial morphology (Saunders *et al.*, 1980), while parental craniofacial data can predict the craniofacial growth of their children (Suzuki and Takahama, 1991). Because the parental craniofacial morphology in OFC reflects the genetic influences on the development of OFC in their offspring, the parents of children with OFC offer an ideal opportunity to investigate and characterize the craniofacial morphology in OFC. This will assist in the search for OFC morphogenes and in the identification of parents 'at risk' of producing further children with OFC.

Fraser and Pashayan (1970) found that the parental facial shape in OFC differed from that of a control group. A systematic review of the subsequent cephalometric studies identified that the parental craniofacial morphology in OFC differed from the non-cleft population and the parental craniofacial morphology in CL(P) and CP are distinct (McIntyre and Mossey, 2002). Nevertheless, the parental craniofacial morphology in OFC was not consistent and several conflicting results were detected. One explanation was the reliance on conventional cephalometric analyses (CCA) to measure craniofacial morphology (McIntyre and Mossey, 2002). CCA measure an arbitrary group of linear distances, angles, and areas; no two studies having measured the same variables. Even multivariate statistical techniques amalgamating CCA variables cannot appropriately measure shape (McIntyre and Mossey, 2003), while the natural craniofacial variability resulting from sex and ethnicity can drastically reduce the statistical power of CCA studies with low subject numbers. Surprisingly, few studies have used the postero-anterior (PA) cephalogram, potentially of greater relevance in investigating

the aetiopathogenesis of a defect principally affecting the transverse oronasal morphology.

Morphometric techniques overcome the 'shape from size' problem associated with CCA. However, despite the availability of suitable software, no study has analysed the parental craniofacial shape in OFC. Procrustes superimposition standardizes location, size, and orientation, permitting unambiguous shape information to be derived (Kendall, 1989). Subsequently, multivariate statistical techniques such as discriminant analysis of the principal components of shape (PCS) can identify any shape difference between groups. Euclidean distance matrix analysis (EDMA; Lele and Richtsmeier, 1991) also compares biological shape using homologous landmark co-ordinates. This technique produces a matrix of the ratios of Euclidean distances. Thin-plate spline analysis (TPS) can display shape differences as the deformation of the surface of an infinitely thin metal plate, draped over the landmarks. The deformation of the surface at each landmark is related to the form difference, calculated using the mathematics of surface spline interpolations (Bookstein, 1991). The magnitude of TPS function is colloquially known as the 'bending energy', and the TPS transformation between the respective forms is visualized using transformation grids.

The objectives of this study were to (1) investigate the shape of the parental craniofacial morphology in OFC, identifying the regions of the craniofacial skeleton morphologically distinguishing the parents of children with OFC from the non-cleft population; (2) determine if the parental craniofacial shape in CL(P) and CP differ; and (3) investigate shape-related sexual dimorphism in parental craniofacial morphology in OFC.

Subjects and methods

The biological parents of a completely ascertained sample of all children with non-syndromic CL(P) and CP born in the west of Scotland between January 1980 and December 1984 were invited to participate in a study (approved by Glasgow Dental Hospital and School Ethics Committee) investigating the parental craniofacial morphology in OFC. Of 196 potential parental pairs, 136 parents replied. However, 32 subjects defaulted for record collection. Fourteen of the 106 parental volunteers were excluded (previous facial trauma, poor quality PA cephalogram) leaving 92 parental PA cephalograms available for this study. Fifty-two were parents of children with CL(P) and 40 of children with CP. The ratio of the cleft types is representative of the high CL(P) to CP ratio of 1:1 within the Scottish and Northern Irish populations (Fitzpatrick *et al.*, 1994; Gregg *et al.*, 1994) compared with a ratio of 2:1 in many other European centres (Jensen *et al.*, 1988). This parental sample was representative of the population when compared with census data (1981 census), no bias being detected with respect to age or social class.

No series of PA cephalograms are available in the UK that could be used as comparison group material in this investigation. Furthermore, in a study involving ionizing radiation, it is unethical to sample the population randomly. Therefore, following ethical and radiological approval from the Tayside Committee on Medical Research Ethics and the Area Radiation Safety Committee, the staff and dental students of the University of Dundee Dental School were invited to volunteer as comparison group subjects. Volunteers were excluded if (1) they were not Scottish Caucasians, (2) a positive personal or family history of OFC or other congenital abnormality was present, (3) previous cranio-maxillo-mandibular surgery had been undertaken, and (4) pregnancy was suspected. Of the total of 44 comparison group PA cephalograms thus obtained, one was discarded due to poor image quality.

The parental and comparison group PA cephalograms were captured using Orthoceph 10 and Sirona Orthophos Plus DS digital cephalometers, respectively (Siemens Dental Systems, Bensheim, Germany). The source–transporionic axis distance was 152 cm and the transporionic axis–film distance was 12 cm. Ear-rods were used and the transporionic axis and Frankfort plane were parallel with the floor (Grummons and Kappeyne, 1987). The nasal rest eliminated rotational errors. The PA cephalograms were scanned at 600 dpi to produce digital images. The co-ordinates of 29 skeletal landmarks (Table 1) were digitized using a 17 inch monitor under identical conditions by one investigator (GMcI). The pixel size was 0.051 mm, smaller than the recommended 0.1 mm maximum (Quintero *et al.*, 1999). Twenty-five per cent ($n = 34$) of the sample was redigitized 1 month later (using the protocol of Houston, 1983) to evaluate individual landmark intra-operator reproducibility by quantifying random and systematic errors using the co-efficient of reliability and a two-sample *t*-test, respectively. The level of significance was $P < 0.95$ for the random error values (Stirrups, 1993) and $P < 0.1$ for systematic errors (Houston, 1983). Twenty-four of the 29 landmarks were reproducible (Table 1).

Procrustes superimposition

Geometrically superimposed landmark configurations were produced using the tpsSmall program (version 1.15) (ftp://life.bio.sunysb.edu/morphmet/tpssmalw32.exe). This uses Procrustean algorithms to scale the configurations of the 24 landmarks to uniform size, translating them to superimpose the centroids, and iteratively rotating the configurations to minimize the squared differences between landmarks (Auffray *et al.*, 1999). This is the 'best-fit' position of the landmark

Table 1 Postero-anterior cephalogram landmarks.

Landmark		Definition
1	RSO	The most superior point on the inner cortical plate of the right orbital rim
2	RGWSO	The intersection of the right greater wing of the sphenoid and the inner cortex of the superolateral orbital rim
3	RMZF	The most medial point of the right zygomaticofrontal suture
4	LSO	The most superior point on the inner cortical plate of the left orbital rim
5	LGWSO	The intersection of the right greater wing of the sphenoid and the inner cortex of the superolateral orbital rim
6	LMZF	The most medial point of the left zygomaticofrontal suture
7	RMO	The most medial point on the inner cortical plate of the right orbital rim
*	CG	The most superior point on the crista galli
8	N	The intersection of the nasal septum and the anterior cranial base—nasion
9	LMO	The most medial point on the inner cortical plate of the left orbital rim
*	RIO	The most inferior point on the inner cortical plate of the right orbital rim
*	LIO	The most inferior point on the inner cortical plate of the left orbital rim
10	RZ	Zygion—the most lateral point on the right zygomatic arch
*	RCond	Condylar—the most superior point on the right mandibular condyle
11	RCor	The most superior point on the right mandibular coronoid process
12	RMast	The most inferior point on the right mastoid process (apex)
13	LZ	Zygion—the most lateral point on the left zygomatic arch
*	LCond	Condylar—the most superior point on the left mandibular condyle
14	LCor	The most superior point on the left mandibular coronoid process
15	LMast	The most inferior point on the left mastoid process (apex)
16	RMx	Maxillare—the most medial point on the right maxillary buttress
17	LMX	Maxillare—the most medial point on the left maxillary buttress
18	RC	The most lateral point on the inner cortex of the right anterior nasal aperture
19	RIN	The most inferior point on the inner cortex of the right anterior nasal aperture
20	ANS	Anterior nasal spine—the centre of the intersection of the nasal septum and the palate
21	LIN	The most inferior point on the inner cortex of the left anterior nasal aperture
22	LC	The most lateral point on the inner cortex of the left anterior nasal aperture
23	RGo	Right gonion—the most outward inferior point on the angle of the mandible
24	LGo	Left gonion—the most outward inferior point on the angle of the mandible

*Landmark not reproducible: deleted from further analyses.

configurations. These were then used to determine the shape differences between (1) the parents of OFC and the comparison group, (2) the parents of CL(P) and the parents of CP, and (3) the male and female parents, using PCS, EDMA, and TPS.

PCS were evaluated using APS (a Procrusté software, version 2.21, http://www.cpod.com/monoweb/aps). Like tpsSmall, APS uses Procrustes algorithms to superimpose specimens. In addition to scaling, translation, and rotation, reflection is also employed. Because differences due to reflection represent asymmetric shape differences (potentially of clinical importance in the investigation of an asymmetric defect such as OFC), tpsSmall was used in advance of APS. Following scaling and superimposition, the Procrustes mean or the consensus configuration (essentially the mean shape) was calculated. The displacement between each landmark and the Procrustes mean was also calculated—producing a matrix of Procrustes residuals for analysis. The shape variance around the landmarks for the combined parental and comparison group subjects is shown in Figure 1.

The shape components were computed by an analysis of the Procrustes residuals covariance matrix. This collates the structure of the data set as new variables, linear combinations of the original variables. Each new variable, a shape component, is a global movement of all the landmarks. The shape components were sorted by magnitude with the null space relegated to the trailing components. For each of the three tests, the first shape components accounted for most of the variance (Table 2). Although the *F* statistic is greater than with

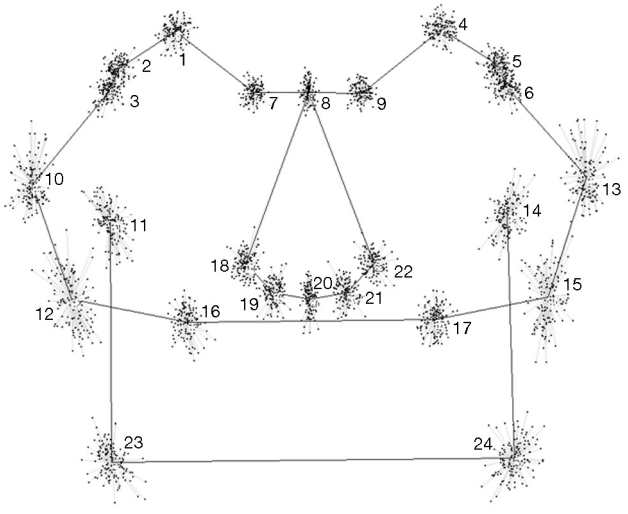

Figure 1 Shape variance for the parents of orofacial clefting and comparison group subjects (for landmark numbers see Table 1).

Table 2 Principal components of shape.

Test	Components	% variance	R^2 value	*F* value	*P* value
Parents of orofacial clefting/comparison group	2	46	0.12	9.0	0.0002
Parents of CL(P)/parents of CP	2	45	0.07	3.47	0.03
Male parents/female parents	2	43	0.02	1.04	0.35

CL(P), cleft lip with or without cleft palate; CP, isolated cleft palate.

the full model, the use of two components reduces the number of degrees of freedom, while still being representative of the shape. Multivariate regression and discriminant analysis then estimated the best linear combination of the principal components separating the test groups.

EDMA

EDMA software (Cole, 1999; http://faith.med.jhmi.edu/) performed a form difference analysis using the mean *x*, *y* co-ordinates of the landmark configurations following initial Procrustes superimposition. This program generates a form matrix for the numerator and denominator landmark configurations by calculating all the possible Euclidean distances between landmark pairs. Each pair of homologous Euclidean distances from the numerator and denominator form matrices are then systematically compared as a ratio, producing the form difference matrix (FDM). This is then sorted to compare the numerator and denominator morphologies by identifying the elements of the FDM that have the smallest and largest values, corresponding to the Euclidean distances that differ by the greatest amount at both extremes. The *T* statistic for form difference testing was calculated as the ratio of the largest to the smallest of the elements of the FDM and represents the total range of shape differences between the two forms. The statistical significance of *T* was assessed by comparing the observed value with the distribution of *T* values using a non-parametric bootstrap procedure (Richtsmeier and Lele, 1993), based on 1000 resamples (pseudosamples), with the denominator as the reference sample. The *T* statistic and the median ratio summarize the FDM, and are reported along with the 10 per cent extremities of each FDM, which represent clinically significant shape differences between the forms (McIntyre and Mossey, 2003).

TPS analysis

The mean parental group landmark configuration was deformed into the comparison group consensus configuration using TPS software (ftp://life.bio.sunysb.edu/morphmet/tpssplnw.exe). This produced the 'total spline', which was decomposed into affine and non-affine transformations. The affine transformation delineates the changes due to size, rotation, and uniform shape change. The non-affine transformations delineating non-uniform or local deformations were further decomposed into p-3 partial warps or localized components (where p is the number of landmarks). These 21 partial warps correspond to deformations at differing geometric scales. The systematic comparison of individual partial warps towards the total spline determines the contribution of each partial warp to the morphology under test.

Results

PCS analysis (Table 2)

The global shape of the parental OFC and comparison group landmark configurations differed statistically significantly ($P < 0.001$), while the parental craniofacial morphology in CL(P) and CP ($P = 0.03$), and the paternal and maternal craniofacial shape ($P = 0.35$) did not differ significantly.

Figure 2 demonstrates the features that discriminated the parents of children with OFC from the comparison

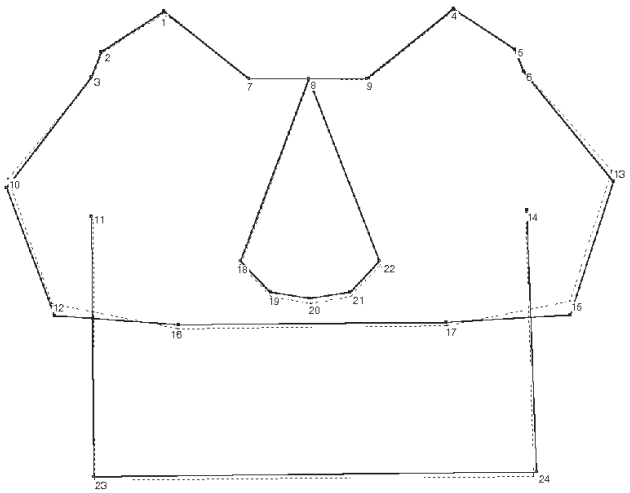

Figure 2 Principal components of shape discrimination between the parents of children with orofacial clefting and the comparison group (for landmark numbers see Table 1).

group. These were relative inferolateral positioning of the right and left zygion, inferior positioning of both mastoids, superior positioning of the nasal floor and both maxillare, lateral expansion of the right side of the nose, superior positioning of the gonia, with minimal changes at the superolateral orbit, nasal roof and medial orbit. Principal component 1 (33 per cent of the variance; Figure 3) was characterized by inferior positioning of the mastoids, superior positioning of the nasal floor and both maxillare; principal component 2 (13 per cent of the variance; Figure 4) by inferolateral positioning of the right and left zygion, superolateral positioning of both coronoidale, the right nasal cavity and both gonia.

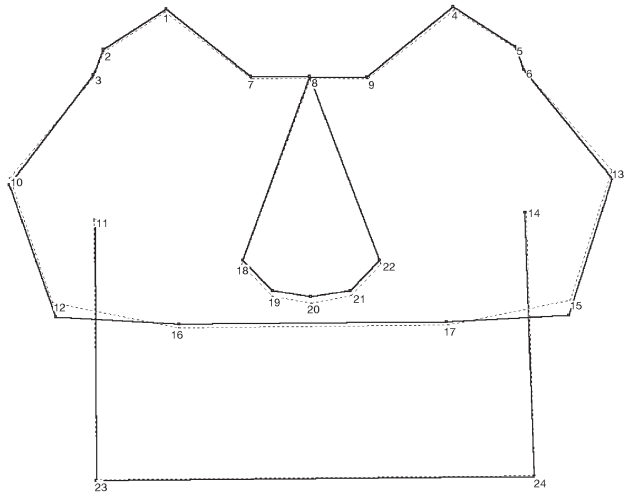

Figure 3 Principal components of shape analysis principal component 1 (for landmark numbers see Table 1).

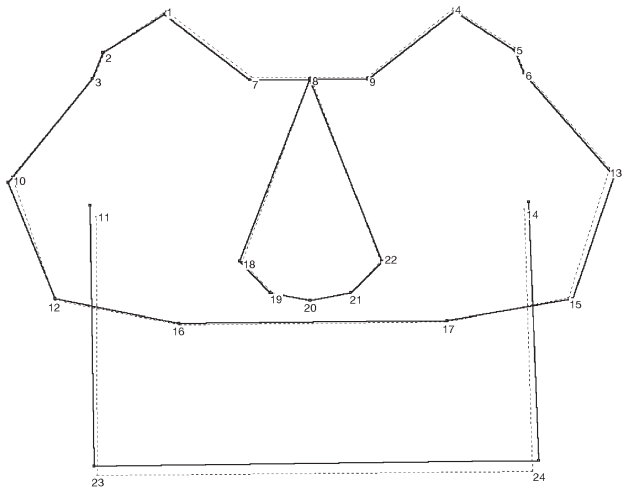

Figure 4 Principal components of shape analysis principal component 2 (for landmark numbers see Table 1).

EDMA

Although each FDM comprised 276 elements, the 10 per cent extremities for the parental OFC/comparison group, parental CL(P)/CP and male/female parents are contained in Tables 3–5. For the parents of OFC/comparison group, the *T* statistic was 1.763, demonstrating significant morphological variation. As only 0.1 per cent of the bootstrapped *T*s were greater than 1.763, this shape difference was statistically significant ($P = 0.001$). The median ratio was 0.997: between RMZF and LMZF, with eight ratios of 1.000. Seventeen ratios were clinically important, being either 10 per cent larger or smaller (Figure 5). The remaining 259 ratios

Table 3 Parental orofacial clefting/comparison group form difference matrix: 10 per cent extremities and median statistic. See Table 1 for landmark definitions.

Euclidean distance	Ratio
LMast–LGo	0.835
RMast–RGo	0.849
LGWSO–LMZF	0.864
LZ–LCor	0.882
RMZF–LMZF	**0.997**
RZ–RMast	1.100
RGWSO–RMast	1.113
LSO–LMast	1.117
RGWSO–RZ	1.118
LSO–LZ	1.120
RMZF–RMast	1.129
LGWSO–LMast	1.140
RMZF–RZ	1.143
LMZF–LMast	1.174
LGWSO–LZ	1.183
RCor–RMast	1.232
LMZF–LZ	1.255
LCor–LMast	1.472

T statistic (maximum/minimum): 1.763 ($P = 0.001$).
Median ratio in bold.

Table 4 Parental cleft lip and palate/cleft palate form difference matrix: 10 per cent extremities and median statistic. See Table 1 for landmark definitions.

Euclidean distance	Ratio
LMast–LGo	0.881
RIN–ANS	0.886
RMast–RGo	0.926
RCor–RMX	0.930
RIN–LIN	0.938
RZ–RC	**0.998**
LGWSO–LMast	1.074
LMZF–LMast	1.076
LZ–LMast	1.094
RCor–RMast	1.125
LCor–LMast	1.159

T statistic (maximum/minimum): 1.316 ($P = 0.027$).
Median ratio in bold.

Table 5 Maternal/paternal form difference matrix: 10 per cent extremities and median statistic. See Table 1 for landmark definitions.

Euclidean distance	Ratio
RCor–RMX	0.939
LCor–LMX	0.944
RCor–RMast	0.944
RZ–RMast	0.947
RIN–ANS	0.948
LGWSO–LC	**1.001**
RSO–RMZF	1.062
LSO–LMZF	1.067
LMX–LC	1.071
RSO–RGWSO	1.072
LSO–LGWSO	1.078

T statistic (maximum/minimum): 1.147 ($P = 0.525$).
Median ratio in bold.

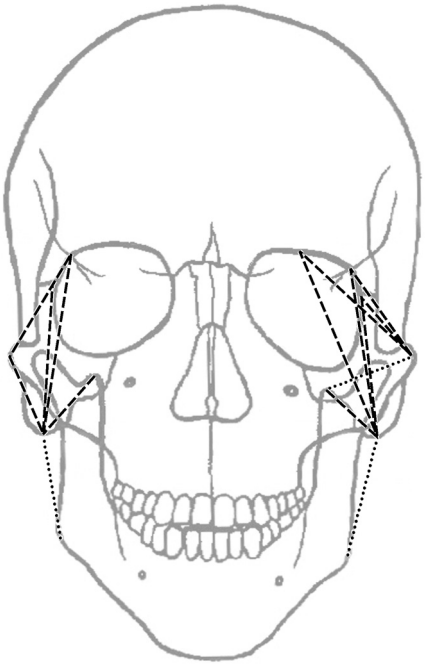

Figure 5 Clinically important ratios of Euclidean distances for parents of orofacial clefting/comparison group (dashed lines = larger ratios of Euclidean distances in the parents of children with orofacial clefting; dotted lines = smaller ratios of Euclidean distances in the parents of children with orofacial clefting).

(94 per cent of the total) involved less than a 10 per cent difference in morphology between the parental and comparison groups.

For the parental CL(P)/CP groups and male/female parents the median ratios were 0.998 and 1.001, respectively. The *T* values of 1.316 ($P = 0.027$) and 1.147 ($P = 0.525$), respectively, were not statistically significant.

TPS analysis

The total spline is depicted in Figure 6, demonstrating superolateral expansion around the orbits and zygoma, inferolateral expansion at both gonia, with relative horizontal constriction at the level of the maxilla and mastoids, and vertical constriction at the nasal floor and maxilla. The bending energy of the total spline was 0.088164. The bending energy of the affine change was zero with only minor tilting of the plate. The affine change has been described as 'the parallel lines remain parallel' (Slice *et al.*, 1998) (Figure 7). Because the affine change contributed no bending to the total spline, the non-affine change (Figure 8) accounted for all the bending. Non-affine transformations delineate non-uniform or local deformations. These were decomposed into localized components, represented by the 21 partial

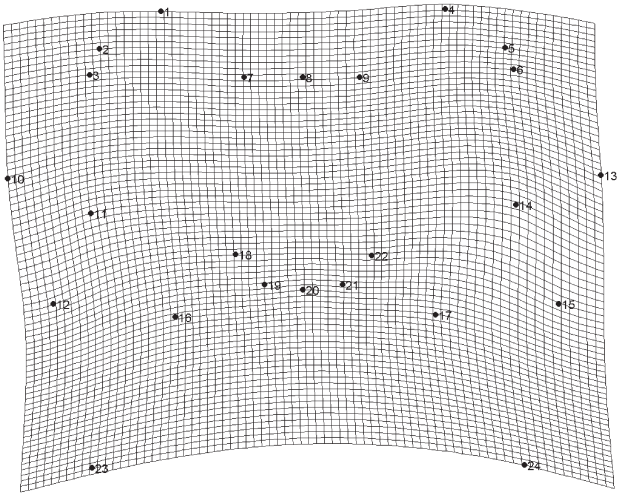

Figure 6 Total spline (for landmark numbers see Table 1).

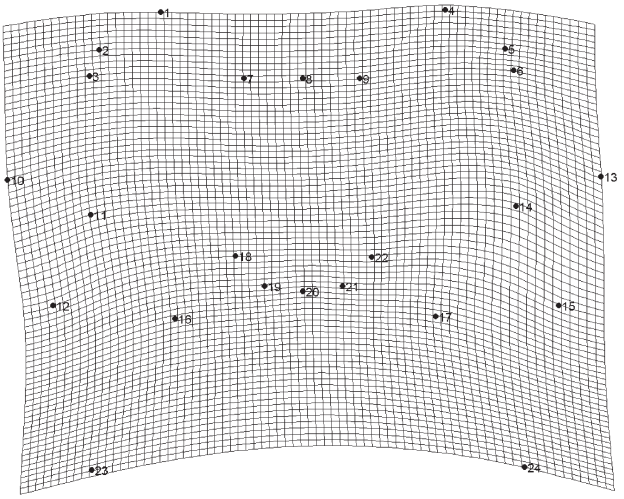

Figure 7 Affine transformation (for landmark numbers see Table 1).

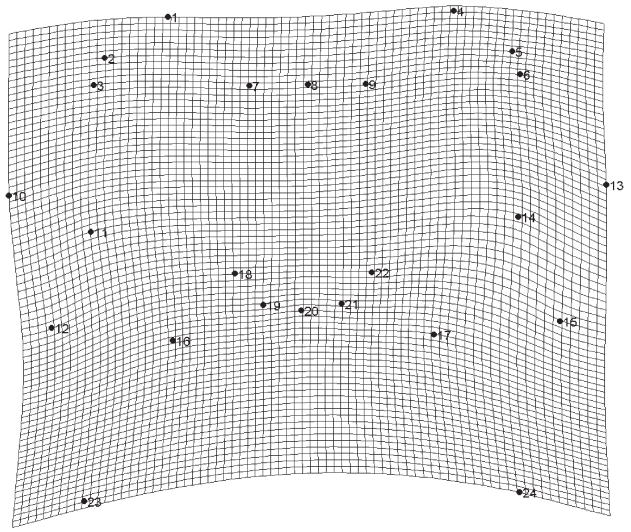

Figure 8 Non-affine transformation (for landmark numbers see Table 1).

warps (Table 6). The eigenvalue is an inverse measure of the spatial scale of the deformation for the partial warp. Large eigenvalues correspond to small-scale deformations and *vice versa*. The bending energy value and its relative contribution to the total spline are displayed, as well as a measure of the strength ('magnitude') of each partial warp, which is independent of spatial scale (Table 6). The partial warps contributing greater than 10 per cent to the non-affine change were determined to be of clinical importance. Partial warp 6 (Figure 9) was characterized by vertical compression of the nasal floor and maxilla, partial warp 9 (Figure 10) by supraorbital expansion, and partial warp 13 (Figure 11) by superolateral orbital expansion. These three partial warps contributed 62 per cent of the non-affine change.

Discussion

The statistically significant craniofacial shape differences found between the parents of children with OFC and the comparison group involve variations in the vertical and transverse location of the craniofacial anatomy. A disproportion between the relative size of the developing palatal processes to the face and head could mitigate against the potential for primary and secondary palatal fusion and structural continuity (Fraser and Pashayan, 1970). Although their relative size is of crucial importance in their ability to contact, the shape features identified in this study may be of greater significance in the aetiopathogenesis of OFC. Thus, the subtle shape difference in the vertical position of the nasal floor and maxillary base in relation to the larger dimensions of the superolateral aspect of the face could represent that the OFC morphogenes specify a wider face, adversely influencing the propensity of the palatal processes to contact. This morphological feature in the presence of, perhaps, an additional OFC environmental factor, could have resulted in the development of an overt cleft in the children of this parental group.

In a systematic review of the published cephalometric studies, McIntyre and Mossey (2002) determined the nasal width to be clinically significantly larger in the parents of children with OFC. Importantly, the present study identified a nasal shape difference: expansion

Table 6 Partial warps.

Partial warp	Eigenvalues	Energy	% contribution	Magnitude
1	606.14908	0.0033242	1.89	0.0000054841
2	491.61096	0.0045657	2.59	0.0000092871
3	483.87443	0.00096665	0.55	0.0000019977
4	281.27268	0.002329	1.32	0.0000082801
5	240.93655	0.0015	0.85	0.0000082801
6	**103.43682**	**0.039904**	**22.63**	**0.00038578**
7	95.86703	0.0047701	2.71	0.000049757
8	85.53706	0.005917	3.36	0.000069175
9	**77.71397**	**0.032195**	**18.26**	**0.00041428**
10	61.21303	0.0066563	3.77	0.00010874
11	58.87188	0.0010925	0.62	0.000018556
12	56.09876	0.005812	3.3	0.000018556
13	**45.54867**	**0.037526**	**21.28**	**0.00082387**
14	36.22681	0.0018811	1.07	0.000051926
15	29.16057	0.0012383	0.7	0.000042465
16	12.97262	0.0038735	2.2	0.00029859
17	11.43774	0.013866	7.86	0.0012123
18	10.1372	0.00031447	0.18	0.000031022
19	4.75979	0.0010023	0.57	0.00021058
20	2.64672	0.000093145	0.05	0.000035192
21	2.2406	0.0075	4.25	0.0033473

Bold denotes greater than 10 per cent contribution to the non-affine component.

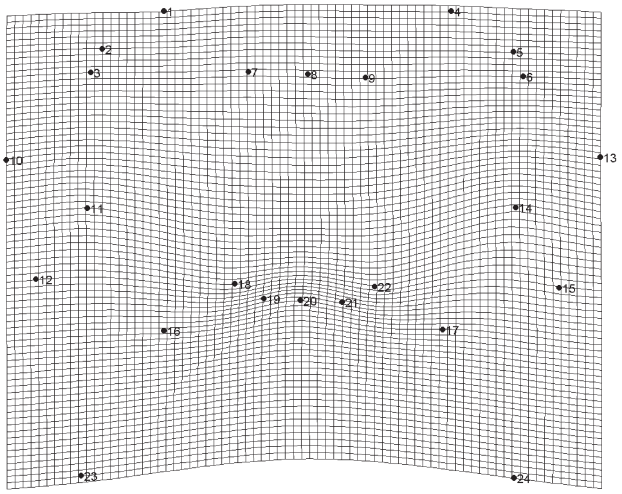

Figure 9 Partial warp 6 (for landmark numbers see Table 1).

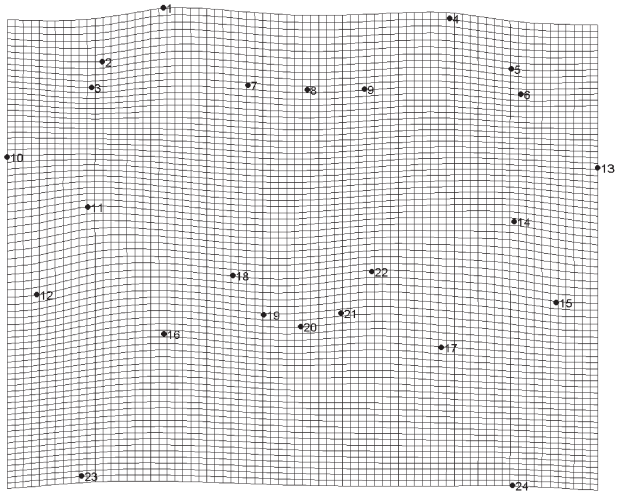

Figure 10 Partial warp 9 (for landmark numbers see Table 1).

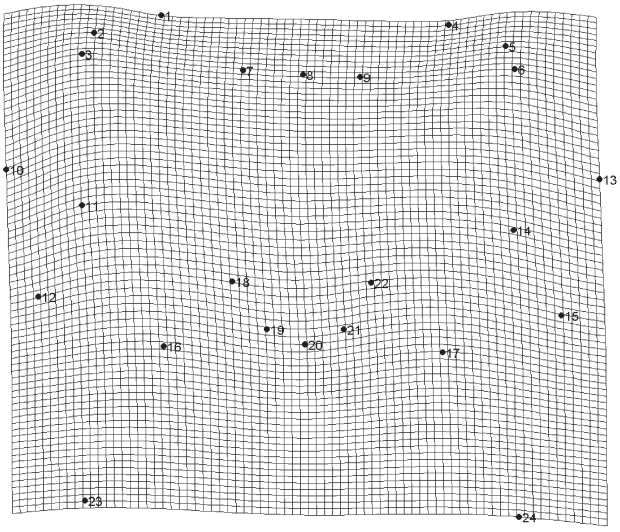

Figure 11 Partial warp 13 (for landmark numbers see Table 1).

located at the right side of the anterior nasal aperture. Although this feature appears at first to conflict with the predilection for left-sided clefting, it should be remembered that the experimental group was a mixture of the parents of children with right- and left-sided clefts.

Mandibular growth is a prime mover in facilitating secondary palatogenesis (Diewert, 1985) by assisting in the withdrawal of the tongue from the oronasal chamber, allowing the palatal shelves to overpower the resistance of the tongue and 'flip up'. Could it be, that the mandibular morphology possessed by this parental group, principally in the vertical plane, has the potential to hinder the physiological overpowering of the tongue by the secondary palatal shelves? On the other hand, the shape difference located at the mastoids is one of several general dysmorphic features that characterize the parental craniofacial morphology in OFC.

Homeotic genes, such as morphoregulatory genes controlling craniofacial morphogenesis (Slavkin, 2000), specify the 'geometry' of orofacial form. These could be crucial in the development of OFC, by patterning an aberration in the embryonic craniofacial morphology. Several OFC candidate genes are currently under investigation, including TGFα, TGFβ and MSX1. However, only one study has combined parental craniofacial morphology with genotypic data (Mossey *et al.*, 1998a). It remains that phenotypic data from studies of the parental craniofacial morphology could be used to investigate OFC morphogenes.

Using three different morphometric techniques, the parental craniofacial shapes in CL(P) and CP were not statistically significantly different. This was a surprising finding, considering that CL(P) and CP are aetiologically distinct, while the lateral cephalometric morphology of the parents of CL(P) and CP children differ (McIntyre and Mossey, 2002). However, no study has, to date, compared the shape of the parental craniofacial skeleton in CL(P) with CP using PA cephalograms. The absence of a statistically significant parental PA cephalometric shape difference between these two distinct conditions could mean that the OFC morphogenes merely specify an aberration in oronasal morphology, while other environmental and anatomical factors determine between the CL(P) and CP phenotypes. Alternatively, distinct morphogenes could be involved in the aetiopathogenesis of CL(P) and CP, yet produce similar craniofacial phenotypes as viewed using the PA cephalogram.

There was no statistically significant sexually dimorphic shape difference in this parental group when investigated using three morphometric techniques. This conflicts with cephalometric studies identifying size-related sexual dimorphism in the craniofacial complex (Riolo *et al.*, 1974; Bhatia and Leighton, 1993). Furthermore, Mossey *et al.* (1997) identified size-related sexual dimorphism using CCA of lateral cephalograms

obtained from the same parents in this study. However, the present study investigated shape from the frontal perspective and the absence of shape-related sexual dimorphism is in keeping with Ferrario *et al.* (1995) who reported size- but not shape-related sexual dimorphism in soft tissue morphology when assessed using the Fourier series. Nevertheless, the significance of size-related antero-posterior and vertical sexual dimorphism is of relevance in the female predilection to CP. This is because the relative time delay for palatal shelf elevation in female embryos when compared with male embryos (Burdi and Silvey, 1969) adversely influences palatal closure. Therefore, continued transverse head growth could increase the disproportion between the palatal shelves and overall head width. Interestingly, Blanco *et al.* (2001) found genetic variation at the MSX1 locus to be a predisposing gene involved in the sex-dependent susceptibility to OFC. As a consequence, it is postulated that there could be a relationship between MSX1 variants and paternal or maternal craniofacial sizes in OFC.

Despite the presence of a demographic imbalance between the parental and comparison groups, with 80 per cent of the comparison group aged under 30 years, this would exert little significance in a study investigating craniofacial shape. This is because the majority of transverse facial growth is complete before 20 years of age (Björk and Skieller, 1974; Işeri and Solow, 2000).

Three different co-ordinate based morphometric techniques were used in this study, because each on its own may not fully describe form, and as a result, the synthesis of the results from different techniques is appropriate (McIntyre and Mossey, 2003). Not surprisingly, although the morphometric techniques produced similar results, they were not identical. This is because each has its individual mathematical *modus operandi.* Only the first three principal components in the PCS were examined and a 10 per cent threshold was applied to the EDMA and TPS data to identify the clinically important shape differences. The rest of the data represented variation due to 'noise' and were discarded. Ten per cent was selected as the threshold, because area measurements have been accorded as a sensitive measure of shape (Mossey *et al.* 1998b), and a 10 per cent threshold for clinically significant area measurements was used in the systematic review by McIntyre and Mossey (2002).

No study of the parental craniofacial complex in OFC has, to date, employed geometric morphometrics. This could partially explain the absence of corroboration among previous studies (McIntyre and Mossey, 2002). Using PCS, EDMA, and TPS to evaluate shape and shape changes is of greater relevance in an investigation of the aetiopathogenesis of OFC than an analysis of the size difference of individual craniofacial components. Nevertheless, parental cephalometric information derived using both CCA and geometric morphometric techniques could be synthesized in investigations of OFC morphogenes and in the identification of parents at risk of producing further children with OFC. Because there are no clinical or laboratory tests to indicate if parents are predisposed to produce children with OFC, genetic counselling currently relies on an empirical recurrence risk. As this method is relatively crude, it is hoped that when sufficient parental craniofacial phenotypic data have been accrued, a model of the cleft-specific parental craniofacial morphological features could be applied to the cephalometric data of potential at risk parents who wish to determine if they possess the cleft-specific craniofacial phenotypic features.

It is surprising that as OFC principally affects the transverse oronasal dimensions, only eight of the previous parental cephalometric investigations have used PA cephalograms. PA cephalograms were used to image the craniofacial skeleton in this study because the anterior transverse and vertical craniofacial morphology are likely to have a greater relevance to the development of a midline defect, such as OFC. This is in contrast to the antero-posterior and posterior vertical craniofacial morphology assessed using lateral cephalograms.

Further studies investigating parental craniofacial morphology in different geographical regions and diverse ethnic populations are required.

Address for correspondence

Grant McIntyre
Orthodontic Department
Glasgow Dental Hospital and School
378 Sauchiehall Street
Glasgow G2 3JZ
UK

Acknowledgement

This study was supported by a European Orthodontic Society research grant.

References

Auffray J-C, Debat V, Alibert P 1999 Shape asymmetry and developmental stability. In: Chaplain M A, Singh G D, McLachlan J C (eds) On growth and form spatio-temporal pattern formation in biology. Wiley, Chichester, pp. 309–324

Bhatia S N, Leighton B C 1993 A manual of facial growth. A computer analysis of longitudinal cephalometric growth data. Oxford University Press, Oxford

Björk A, Skieller V 1974 Growth in width of the maxilla studied by the implant method. Scandinavian Journal of Plastic and Reconstructive Surgery 8: 26–33

Blanco R *et al.* 2001 Evidence of a sex-dependent association between the MSX1 locus and nonsyndromic cleft lip with or without cleft palate in the Chilean population. Human Biology 73: 81–89

Bookstein F L 1991 Morphometrics tools for landmark data. Cambridge University Press, Cambridge

Burdi A R, Silvey R G 1969 Sexual differences in closure of the human palatal shelves. Cleft Palate Journal 6: 1–7

Cole TM III 1999 *Win*EDMA: software for Euclidean distance matrix analysis. University of Missouri, Kansas City School of Medicine, Kansas City

Diewert V M 1985 Development of human craniofacial morphology during the late embryonic and early fetal periods. American Journal of Orthodontics 88: 64–76

Ferrario V F, Sforza C, Schmitz J H, Miani Jr A, Taroni G 1995 Fourier analysis of human soft tissue facial shape: sex differences in normal adults. Journal of Anatomy 187: 593–602

Fitzpatrick D R, Raine P A, Boorman J G 1994 Facial clefts in the west of Scotland 1980–84: epidemiology and genetic diagnoses. Journal of Medical Genetics 31: 126–129

Fraser F C, Pashayan H 1970 Relation of face shape to susceptibility to congenital cleft lip. Journal of Medical Genetics 7: 112–117

Gregg T, Boyd D, Richardson A 1994 The birth prevalence of cleft lip and palate in Northern Ireland from 1980 to 1990. British Journal of Orthodontics 21: 387–392

Grummons D C, Kappeyne M A 1987 A frontal asymmetry analysis. Journal of Clinical Orthodontics 21: 448–465

Houston W J B 1983 The analysis of errors in orthodontic measurements. American Journal of Orthodontics 83: 382–390

Işeri H, Solow B 2000 Changes in the width of the mandibular body from 6 to 23 years of age: an implant study. European Journal of Orthodontics 22: 229–238

Jensen B L, Kreiborg S, Dahl E, Fogh-Andersen P 1988 Cleft lip and palate in Denmark 1976–1981. Epidemiology, variability and early somatic development. Cleft Palate Journal 25: 258–269

Kendall D G 1989 A survey of the statistical theory of shape. Statistical Science 4: 87–120

Lele S, Richtsmeier J T 1991 Euclidean distance matrix analysis: a coordinate-free approach for comparing biological shapes using landmark data. American Journal of Physical Anthropology 86: 415–427

McIntyre G T, Mossey P A 2002 The craniofacial morphology of the parents of children with orofacial clefting: a systematic review of cephalometric studies. Journal of Orthodontics 29: 23–29

McIntyre G T, Mossey P A 2003 Size and shape measurement in contemporary cephalometrics. European Journal of Orthodontics 25: 231–242

Mossey P A, McColl J H, Stirrups D R 1997 Differentiation between cleft lip with or without cleft palate and isolated cleft palate using parental cephalometric parameters. Cleft Palate–Craniofacial Journal 4: 27–35

Mossey P A, Arngrimsson R, McColl J H, Vintiner G M, Connor J M 1998a Prediction of liability to orofacial clefting using genetic and craniofacial data from parents. Journal of Medical Genetics 35: 371–378

Mossey P A, McColl J, O'Hara M 1998b Cephalometric features in the parents of children with orofacial clefting. British Journal of Oral and Maxillofacial Surgery 36: 202–212

Quintero J C, Trosien A, Hatcher D, Kapila S 1999 Craniofacial imaging in orthodontics: historical perspective, current status, and future developments. Angle Orthodontist 69: 491–506

Richtsmeier J T, Lele S 1993 A coordinate-free approach to the analysis of growth patterns: models and theoretical considerations. Biological Reviews of the Cambridge Philosophical Society 68: 381–411

Riolo M L, Moyers R E, McNamara Jr J A, Hunter W S 1974 An atlas of craniofacial growth: cephalometric standards from the University of Michigan growth study. Monograph No. 2, Craniofacial Growth Series. Center for Human Growth and Development, University of Michigan, Ann Arbor

Saunders S R, Popovich F, Thompson G W 1980 Family study of craniofacial dimensions in the Burlington Growth Centre sample. American Journal of Orthodontics 78: 394–403

Slavkin H 2000 Toward understanding the molecular basis of craniofacial growth and development. American Journal of Orthodontics and Dentofacial Orthopedics 117: 538–539

Slice D E, Bookstein F L, Marcus L F, Rohlf F J 1998 A glossary for geometric morphometrics: part 1 (http://129.49.19.42/morph/glossary/gloss1html)

Stirrups D R 1993 Guidance on presentation of cephalometry-based research studies. A personal perspective. British Journal of Orthodontics 20: 359–365

Suzuki A, Takahama Y 1991 Parental data used to predict growth of craniofacial form. American Journal of Orthodontics and Dentofacial Orthopedics 99: 107–121

Ward R E, Bixler D, Raywood E R 1989 A study of cephalometric features in cleft lip–cleft palate families. I: Phenotypic heterogeneity and genetic predisposition in parents of sporadic cases. Cleft Palate Journal 26: 318–325