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Anthropometric and cephalometric measurements in X-linked hypohidrotic ectodermal dysplasia

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Structured Abstract

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Objective – To describe the somatic development and craniofacial morphology in males affected with hypohidrotic ectodermal dysplasia (HED) and female carriers and to find clinical markers for early clinical diagnosis of possible female carriers.

Design – A clinical and radiographic examination of the affected males and the female carriers.

Setting and sample population – Twenty-four affected males and 43 female carriers with a known mutation in the *ED1* gene were examined in a dental clinic in either Copenhagen or Aarhus, Denmark.

Experimental variables – Height, body mass index (BMI) and head circumference. Cephalometric analysis of the craniofacial morphology.

Outcome measure – Data on the somatic and craniofacial development in the affected males and female carriers.

Results – No difference was observed regarding body height in the affected males and female carriers, BMI values were lower than the mean in most affected boys and adolescence and head circumference was somewhat decreased in both groups compared to normative data. The cephalometric analysis showed a reduced maxilla length and prognathism, a normal size and shape of the mandible and a reduced sagittal jaw relationship in both HED groups. Furthermore, affected males had a retroclined nasal bone and a more anteriorly inclined maxilla. A short nose, protruding lips, reduced facial convexity and facial height, characterized the soft tissue profile of the affected males. In female carriers, the lips were significantly retruded when compared with controls.

Conclusion – No specific somatic or cephalometric markers could be observed, in the female carrier group.

Key words: anthropometry; cephalometry; clinical markers; ectodermal dysplasia; female carriers; hypohidrotic

Introduction

To date more than 150 distinct conditions in which ectodermal derivated tissue is affected have been identified (1). Most of these conditions are rare and manifest defects in the morphogenesis of ectodermal derivated structures; e.g. the hair, skin, nails, teeth and glands. The most common

type of ectodermal dysplasia (ED) is the X-linked hypohidrotic ectodermal dysplasia (HED) where males are usually more severely affected, and females show variable degrees of symptoms because of the X-chromosome inactivation (2, 3).

The X-linked form of HED is caused by mutations in the *ED1* gene, located at Xq12–q13.1. This gene is normally expressed in tissues derived from the ectoderm and eight different splice forms are known. The *ED1* gene encodes the transmembrane protein, ectodysplasin-A, which is a member of the tumour necrosis factor family (4–6). To date, more than 85 different *ED1* mutations have been identified (The Human Gene Mutation Database, Cardiff). The autosomal recessive and the autosomal dominant forms of HED, which are caused by mutations in the gene encoding the receptor for ectodysplasin-A (*EDAR*) or in the death domain adapter signalling molecule EDARADD, have an identical phenotype to the X-linked HED (7, 8).

Only a few studies have examined the somatic growth characteristics of children with ED, and found that body height in children with HED was not different from the normal population (9). Motil et al. (9) also reported that children with ED, in general, had lower body mass index (BMI) than normal children, but no specific data for HED were presented. Ward and Bixler (10) observed that the head circumference was reduced in HED.

Clinical studies have reported on facial dysmorphology in affected males, e.g. frontal bossing, saddle nose, small nose, maxillary hypoplasia and prominent lips (11, 12), and these traits have even been employed in prenatal diagnosis of an affected male from 3D ultrasonography at 30 weeks gestation (13).

A few studies have attempted to quantify the facial dysmorphology in both affected males and female carriers by anthropometry (10, 14–16) or roentgencephalometry (16–20). The roentgencephalometric studies showed that affected males have a reduced facial height, a retrusive short maxilla and a prognathic mandible. Johnson et al. (20) suggested that the number of missing maxillary permanent teeth was significantly related to craniofacial dysmorphology in the ED population. However, in none of the aforementioned studies was a DNA-analysis carried out and some studies pooled different types of ED and males and females. In general, the sample sizes were small and the age range wide.

The purpose of the present study, based on a nation-wide Danish survey, was to analyse the somatic development and the craniofacial morphology, including the soft tissue profile, in affected males and female carriers with a mutation in the *ED1* gene. The dental characteristics of these groups have recently been published (21).

We hypothesized that:

- Body height, BMI and head circumference in affected males and female carriers do not differ from those in the normal population.
- Craniofacial morphology in affected males and female carriers differs from that of the normal population, and the deviations are the most pronounced for the males.
- There is a positive correlation between the severity of the deviations in craniofacial morphology and the severity of hypodontia in female carriers.

In a further perspective, it was anticipated that the study could contribute to the identification of cephalometric biomarkers, which could aid the clinical identification of potential female carriers of HED.

Materials and methods

Study group

With the existence of a well-organized Danish community dentistry system covering all children in the country (since 1972), it has been possible to include a large group of affected males and female carriers of HED. Families with a proband who was clinically diagnosed by the dentist as having HED were contacted and invited to be included in the study. Informed consent was obtained in all instances according to Danish law. Probands and possible female carriers were examined clinically.

The examination included measurements of body height, weight and head circumference and radiographic examination (panoramic radiographs and cephalometric radiographs). Blood samples (9.5 ml) were drawn for mutation and chromosome analysis, which was made by PCR, SSCP and direct sequencing to detect mutations in the *ED1* gene. Sixteen different mutations were detected including: missense, and frame-shift mutations in frame deletions as well as deletions of one or more exons and splice site mutations. Only the affected males and the female carriers with known mutation in the *ED1* gene were included in the present study with 24 affected males and 43 female

carriers. The mean age and range of the affected males was 19 years (2–49 years) and for the female carriers 35 years (6–73 years).

The ethical committee of Copenhagen and Frederiksberg in Denmark approved the study [reference number (KF)03-005/03].

Measurements of height, weight and head circumference

Body height (in cm) and weight (in kg) were measured without footwear. The head circumference (in cm) was measured with a metal tape placed over the glabella and the opisthocranion. To define underweight, normal weight, overweight and obesity, the weight-for-height measurement BMI was used as it is associated with total body fat and can therefore, be used to calculate the morbidity risk because of, e.g. underweight or obesity. BMI is calculated as weight divided with height squared (kg/m^2) (22, 23). Data regarding body height were compared with normative data of Andersen et al. (24); head circumference data of children and adolescents up to the age of 16 years with those of Jansen (25), while data concerning adults over the age of 18 years were compared with those of Solow (26) and Ingerslev and Solow (27). BMI data for children and adolescents (up to the age of 18 years) were compared with those of He et al. (28), and for adults, with the WHO BMI classification (23).

Cephalometric analysis

In this part of the study, only affected males and female carriers above the age of 16 years were included. Table 1 indicates the gender and age distribution as well as information about the number of missing permanent teeth. Children and adolescents younger than 16 years of age were not included as the samples in the different age groups were small.

To reduce the travel distance for the participants, the examinations were conducted in dental clinics at two locations (Copenhagen and Aarhus). At both locations, the roentgencephalometric exposures were made at 70–72 kV and a grid, and an intensifying screen was employed to minimize the participants' exposure to X-rays as much as possible; furthermore, an aluminium filter was used to enhance the soft tissue profile. The distance from the mid-sagittal plane of the head to the film was 10 cm; however, the focus to film distance was 190 cm in the Copenhagen clinic and 160 cm in the Aarhus clinic. Thus, the enlargement of the mid-sagittal plane was 5.6% and 6.7%, respectively. During exposure, the participants were instructed to occlude; however, in two affected males and one female carrier this was not possible, because of extensive number of missing permanent teeth. Therefore, they were instructed to relax the jaw elevator muscles, so the cephalometric exposure was made with the mandible in the resting position and with closed lips. Twenty-seven anatomical reference points (Table 2) were marked on the lateral cephalometric radiographs by an experienced orthodontist (SK). The films were digitized and analysed using the software package TIOPS™ (Department of Orthodontics, Postgraduate studies, University of Copenhagen, Denmark). All films were digitized by the same investigator (MOL). Thirty-one variables were calculated to describe the craniofacial skeleton and the soft tissue profile of the affected males

Table 1. **Characteristics of the group included in the cephalometric analysis**

	Affected males	*p*-values	Female carriers	*p*-values
Number of subjects	10		33	
Age in years	34 (19–49)		42 (17–75)	
Hypodontia (Yes/no/NA)	10/0/0 (100%)		24 / 4 / 5 (86%)	
Mean number of missing teeth	22 (16–26)		4 (0–20)	
Mean body height in cm.	177.1 ± 3.4*	NS	166.5 ± 6.6‡	NS
Median BMI in kg/m^2 (range)	22.5 (20.7–37.7)*		24.9 (19.4–40.7)‡	
Mean head circumference in cm. (SD)	56.9 ± 1.5†	NS	54.7 ± 1.8§	<0.001

NA denotes that the cause for the missing tooth was unknown (agenesis, extraction). With respect to dental and other oral findings see Lexner et al. (21). For some HED males and female carriers not all the somatic measurements were obtained, therefore, *n = 9, †n = 7, ‡n = 31 and §n = 28.
p-Values for female carriers and affected males compared with normative data were obtained by *t*-tests.
NS, not significant.

Table 2. **Definition of the references points and lines used in this study**

References point/lines	Definition
Skeletal points	
ar – Articulare	The intersection between the external contour of the cranial base and the dorsal contour of the condylar head or neck
ba – Basion	The most posterior-inferior point on the anterior margin of foramen magnum
gn – Gnathion	The most inferior point on the mandibular symphysis
go – Gonion*	The midpoint of the angle of the mandible. It is founded by bisecting the angle between the mandibular line and the ramus line
n – Nasion	The most anterior point of the frontonasal suture
na – Nasale	The tip of the nasal bone
pg – Pogonion	The most anterior point on the mandibular symphysis
pgn – Prognathion	The point on the mandibular symphysis farthest from *cd*
pm – Pterygomaxillaris	The intersection between the nasal floor and the posterior contour of the maxilla.
s – Sella	The centre of sella turcica
sp – Spinal point	The apex of the anterior nasal spine
ss – Subspinale	The most posterior point on the anterior contour of the upper alveolar process
Reference lines	
ML – mandibular line	The tangent to the lower border of the mandible through gnathion
NL – nasal line	The line through spinal point and ptygomaxillara
NSL – nasion- sella line	The line through nasion and sella
RL – ramus line	The tangent to the posterior border of the mandible
Soft tissue points	
ct – Chin tangent point	The lower tangent point of the nose chin line (NCL)
gns – Soft tissue gnathion	The soft tissue point overlying *gn*
gs – Soft tissue glabella	The most anterior point on the soft tissue glabella
li – Labrale inferior	The most prominent point on the prolabium of the lower lip
llt – Lower lip tangent point	The upper tangent point of the tangent to the lower lip through *sms*
lnt – Lower nasal tangent point	The upper tangent point of the nose chin line (NCL)
ls – Labrale superior	The most prominent point on the prolabium of the upper lip
ns – Soft tissue nasion	The deepest point in the fronto-nasal curvature
nst – Nasal septum tangent point	The anterior tangent point of the tangent to the nsal septum through *sn*
pgs – Soft tissue pogonion	The soft tissue point overlying *pgn*
prn – pronasale	The most prominent point on the apex of the nose
sgs – Soft tissue supraglabella	The soft tissue point overlying sg
sms – Soft tissue supramentale	The deepest point in the mento-labial sulcus
sn – Subnasale	The deepest point in the naso-labial curvature
sto – Stomiom	The deepest point in the rima oris
unt – Upper nasal tangent point	The nasal tangent point of the nose-frontal line (NFL)
Reference lines	
NCL – nose chin line	The line through lower nasal tangent point and chin tangent point
NFL – nose frontal line	The line through frontal tangent point and Upper nasal tangent point

Definitions according to Kreiborg (29).
*Definition according to Riolo (30).

and female carriers and all linear measurements were corrected for radiographic enlargement depending on the clinic they were taken to. Mean facial diagrams were produced for each *gender* and superimposition of mean diagrams was in every case carried out on the nasion-sella line (NSL) and registered on the sella point according to the method of Kreiborg (29).

The cephalometric analyses were made in different stages. First, the affected males and female carriers, respectively, were compared with normative data from Dahl (31), Kreiborg (29) and Thilander et al. (32), all control data were corrected for the enlargement. Then, to examine the hypothesis regarding the correlation between the number of missing permanent teeth and the craniofacial dysmorphology, the female carrier group was subsequently dichotomized into two sub-groups according to the number of missing permanent teeth, with a cut off point, chosen by us, of four missing permanent teeth, which was the mean number of missing teeth in this group (21), and the cephalometric data in the two groups were subsequently compared. Lastly, the affected males were compared with the female carriers with regard to the angular measurements to examine the hypothesis that the deviations in the affected male group are more severe than in female carriers.

Precision

No calibration was performed regarding the placement of the reference points as the examiner placing these points had been calibrated on numerous occasions previously (29). The digitization was calibrated by choosing 10 randomly selected radiographs where seven selected reference points (X- and Y-values), given a total of 140 measurements, were digitized twice, 2 weeks apart. The results showed a nearly complete agreement within the same examiner ($r = 0.99$). The percentage difference within the same examiner in relation to the mean was 0.2 ± 0.5%.

Statistical analysis

The R 2.2.1. Statistical software (R development Core Team, Vienna, Austria 2006) and Excel (Microsoft Corporation, USA) were used to analyse the results. Differences between means were analysed by a two-sample t-test. For comparison with normative data from more than 100 subjects, a one-sample t-test was used with normative data as μ. To correct for multiple tests of statistical significance on the related data (cephalometric measurements and growth related variables), the level of significance was set at 1% throughout.

Results

Anamnestic and clinical data

Anamnestic and clinical data for the affected males and female carriers included in the cephalometric analysis are given in Table 1. This table includes information about the size of the study groups, and data regarding the average number of missing permanent teeth and the range. The clinical and radiographic examination showed that all affected males (100%) and 86% of the female carriers had hypodontia. In comparison, values for hypodontia in the normal population, are 7.5% for males and 9% for females (33), so the frequencies in this study population were highly increased. [For more information regarding dental findings, see Lexner et al. (21)].

Measurements of height, BMI and head circumference

The number of children and adolescents with HED was small for each age group and, therefore, their data were simply plotted in gender specific normative curves for body height, BMI and head circumference (Fig. 1), and no actual statistical testing was attempted. In the group of affected boys, it was observed that the values for body height and BMI were all within the 97% confidence interval, of the normative mean; however, while the values for body height were equally distributed around the mean, the majority of affected boys had BMI values below the normative mean. The values regarding the head circumference in the affected boys all fell within ±2 SD of the mean for the normal population, but they were all lower than the mean. In the carrier girl group, the measurements of body height and BMI were all within the 97% confidence interval, of the normative data and all equally distributed, around the mean. The values regarding the head circumference were all within ±2SD of the mean for the normal population, but all values were equal to or lower than the normative mean.

Table 1 shows the data regarding height, BMI and head circumference in the adult groups of affected males and female carrier. Two female carriers were not

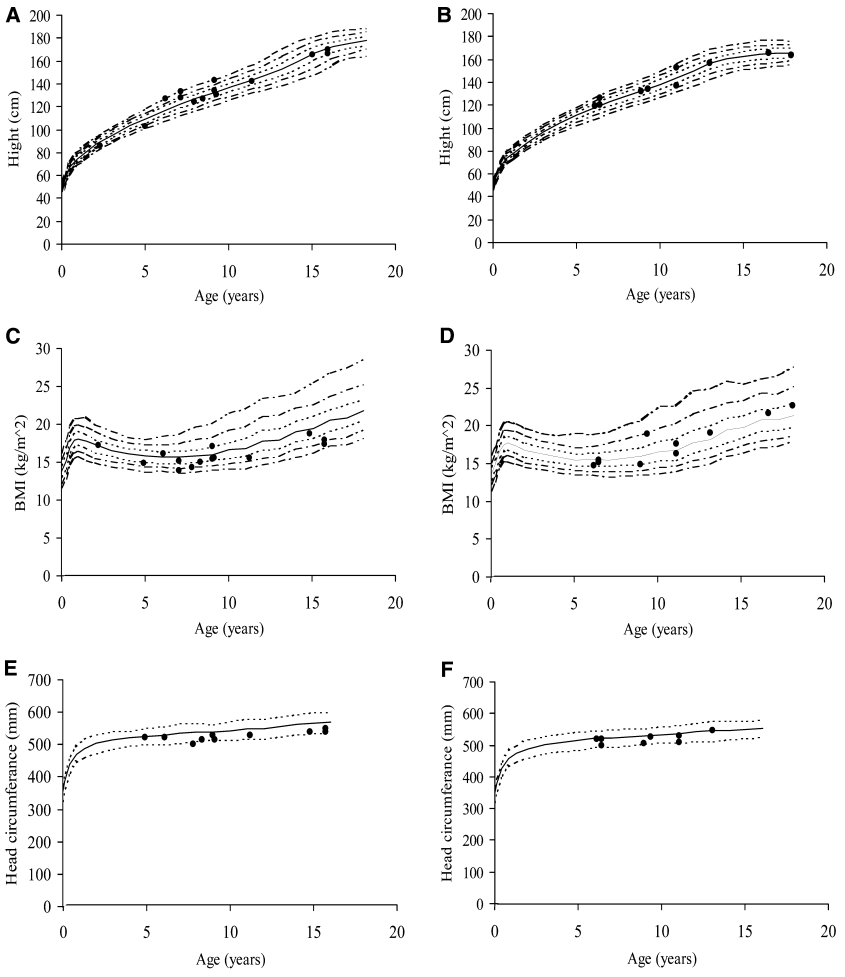

Fig. 1. Body height of affected boys plotted against normative curves for body height (A), body height of female carrier girls plotted against normative curves for body height (B), body mass index (BMI) of affected boys plotted against normative curves for BMI (C), BMI of female carrier girls plotted against normative curves for BMI (D), head circumference of affected boys plotted against normative curves for head circumference (E), and head circumference of female carrier girls plotted against normative curves for head circumference (F). The normative curves for body height and BMI are represented in 3%, 10%, 25%, 50%, 75%, 90%, 97% (A–D) and the normative curve for head circumference is represented by mean ± 2 SD (E and F).

included in this table as they were under the age of 18 years and one affected male and three female carriers did not have all the somatic measurements taken. It was observed that there was no difference regarding the average body height between the affected males and the female carriers, respectively, compared with normative data. The BMI measurements showed that both the affected males and female carriers had median BMI values of 22.5 and 24.9 kg/m^2, respectively. The head circumference of the female carriers was 1.5 cm smaller than that of the normal population and this was statistically significant ($p < 0.001$); the affected males also showed a smaller mean head circumference by 1.6 cm compared with the normative data, but the difference was not significant at the 1% level.

Cephalometric findings in affected males

Table 3 illustrates the cephalometric data for affected males and female carriers as well as the normative data for both gender and the *p*-values. A visual impression of the findings can be obtained from the superimposed mean diagrams shown in Fig. 2.

In the affected male group, the size and shape of the *cranial base* were within normal limits. The length of the *nasal bone* was normal, but the nasal bone was significantly retroclined in relation to the anterior cranial base (*s-n-na*).

The *maxilla* was significantly shorter (*sp-pm*), retrognatic (*s-n-ss*) and anteriorly inclined (*NSL/*NL) in relation to the anterior cranial base than in controls.

The length (*pgn-ar*), posterior height (*ar-go*) and gonial angle (*ML/*RL) of the *mandible* were within normal limits. The mandible was, however, significantly anteriorly inclined in relation to the anterior cranial base (*NSL/*ML) and it was significantly more prognathic (*s-n-pg*) than in the controls.

The mean *sagittal jaw relationship* (*ss-n-pg*) was negative (−9.0°) and significantly decreased in the affected male group. The mean *vertical jaw relationship (NL/*ML) was also decreased, but this difference was not significant.

Table 3. **Cephalometric measurements in affected males and female carriers compared to normative data**

	HED affected males			Male controls*			HED female carriers			Female controls*		
	Number of subjects	Mean	SD	Mean	SD	P-value	Number of subjects	Mean	SD	Mean	SD	P-value
Skeletal morphology												
Cranial base												
n-s	10	71.0	3.4	70.1	3.0	NS	33	68.0	3.5	67.1	2.3	NS
n-ba	10	105.5	3.5	106.2	4.1	NS	33	101.5	5.1	100.4	3.2	NS
n-s-ar	10	123.0	5.0	123.8	5.2	NS	33	125.1	3.9	123.3	5.1	NS
n-s-ba	10	128.8	5.2	130.2	5.6	NS	33	132.1	3.9	129.3	4.9	0.01
Nasal bone												
n-na	10	24.7	4.2	23.7	2.8	NS	33	21.8	2.5	23.1	2.3	NS
s-n-na	10	109.8	5.7	117.4	6.1	0.01	33	111.5	6.2	114.4	6.0	NS
Maxilla												
sp-pm	10	49.5	3.1	55.6	2.5	<0.001	33	50.3	3.0	52.6	2.6	<0.001
s-n-ss	10	78.9	2.7	82.0	3.2	0.01	33	78.8	4.1	81.5	3.5	0.01
NSL / NL	10	2.5	4.8	7.7	3.2	0.01	33	7.4	3.1	7.1	3.2	NS
Mandible												
pgn-ar	10	115.1	8.1	111.4	4.8	NS	33	103.9	4.8	104.6	4.7	NS
ar-go	10	51.5	5.9	50.7	4.6	NS	33	46.3	4.9	46.5	3.7	NS
s-n-pg	10	87.9	6.1	81.2	3.1	0.01	33	80.7	4.9	80.5	3.2	NS
ML / RL	10	120.3	9.9	120.4	6.3	NS	33	119.7	7.4	121.2	5.2	NS
NSL / ML	10	20.7	7.2	28.2	5.9	0.01	33	27.9	7.7	29.6	5.7	NS
Jaw relations												
ss-n-pg	10	−9.0	6.9	0.4	3.1	0.01	33	−2.0	4.4	1.0	1.9	<0.001
NL / ML	10	18.2	5.7	21.0	5.9	NS	33	20.5	6.5	22.0	5.1	NS
Facial height bone												
n-gn	10	110.0	6.5	119.9	6.1	<0.001	33	108.4	7.0	113.3	6.0	0.01
n-sp	10	49.8	4.3	53.1	2.9	NS	33	49.1	2.7	50.0	2.6	NS
sp-gn	10	60.7	5.3	68.4†	4.9†	0.01	33	60.3	5.8	59.9‡	4.4‡	NS
Soft tissue morphology												
Glabella												
sgs-gs-ns	10	149.6	4.6	154.7	6.5	NS	33	163.3	6.8	167.6	4.9	0.01
Facial convexity												
NFL / NCL	10	157.4	9.8	140.2	5.0	<0.001	33	144.6	7.0	143.6	3.7	NS
Nose												
ns-sn	10	48.0	4.4	54.0	3.7	<0.001	33	48.5	3.4	49.1	3.0	NS
ns-prn	10	43.1	4.3	48.7	4.0	0.01	33	42.7	3.8	43.5	3.0	NS
ns-unt / n-s	10	113.6	3.6	116.5	3.9	NS	33	114.2	4.6	116.8	3.7	NS
Lips												
ls-NCL	10	−6.8	3.0	−6.5	2.0	NS	33	−8.9	2.9	−5.4	1.8	<0.001
li-NCL	10	−6.9	3.7	−4.6	2.2	NS	33	−7.1	3.0	−3.1	2.0	<0.001

Table 3. **Continued.**

	HED affected males			Male controls*			HED female carriers			Female controls*		
	Number of subjects	Mean	SD	Mean	SD	P-value	Number of subjects	Mean	SD	Mean	SD	P-value
llt-sms-pgs	10	103.7	29.0	120.9	13.5	NS	33	131.0	16.7	128.7	13.6	NS
nst-sn-ls	10	79.3	16.1	109.1	8.0	<0.001	33	105.5	14.7	111.6	11.7	NS
Facial height soft												
ns-gns	10	113.9	9.3	124.9	5.8	0.01	33	112.4	8.0	114.7	4.1	NS
sto-gns	10	49.1	7.1	52.5	3.6	NS	32§	46.4	4.3	48.4	2.8	NS
ns-sto	10	66.7	5.9	74.7	3.7	<0.001	32§	67.3	5.2	68.9	2.4	NS

p-Values obtained by *t*-test.
NS, not significant.
*Normative data obtained from Kreiborg (29).
†Normative obtained from Dahl (31)
‡Normative data from Thilander et al. (32).
§One female carrier excluded because of open mouth at the X-ray session.

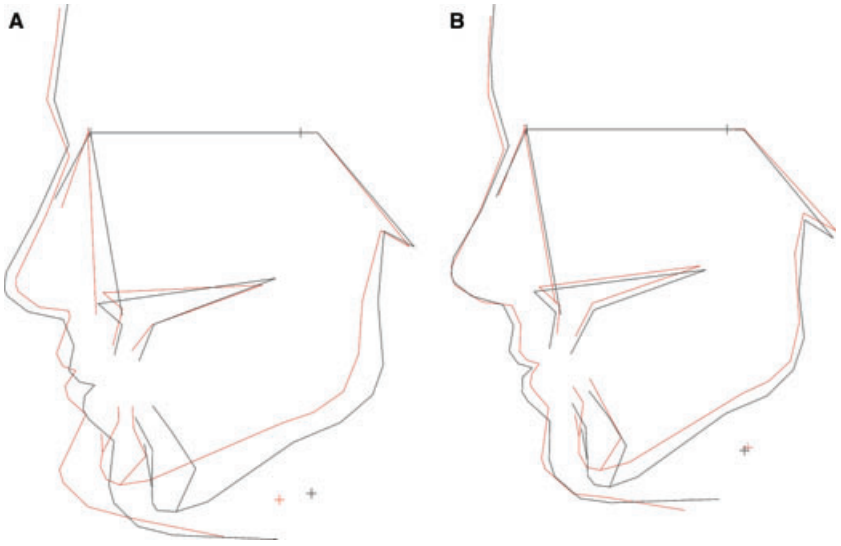

Fig. 2. (A) Mean tracing of the affected males (colour red) superimposed on the mean tracing of the male controls (colour black). (B) Mean tracing of the female carriers (colour red) superimposed on the mean tracing of the female controls (colour black).

The mean total *facial height* (*n-gn*) was about 1 cm shorter in the affected male group, and this difference was statistically significant. Both *upper (n-sp) and lower (sp-gn) facial height* were reduced; however, only the reduction in the lower facial height was statistically significant.

As for the *soft tissue profile*, the *glabella region* (*sgs-gs-ns*) was somewhat more protruding than in controls, but the difference was not statistically significant. The *facial convexity* was reduced in the affected males as the mean *NFL/*NCL angle was significantly increased.

The mean length of the *nose* was significantly shorter (*ns-prn*) in the affected males and the nose was flatter (*unt-ns/*NSL), but this was not statistically significant. The *naso-labial* angle (*nst-sn-ls*) was significantly reduced in the affected males; the *mento-labial* angle (*llt-sms-pgs*) was also markedly reduced (about 17°), but this difference was not statistically significant.

The mean *soft tissue total facial height* (*ns-gns*) was significantly reduced by about 1 cm in the affected male group. Both *upper* (*ns-sto*) and *lower* (*sto-gns*) *soft tissue facial height*s were smaller in the affected males,

but the difference was only significant for the upper facial height.

Cephalometric findings in female carriers

In the female carriers, the size of the *cranial base* (*n-s; n-ba*) was within normal limits. The cranial base angle measured to the reference point basion (*n-s-ba*) was slightly, but significantly increased.

The *maxilla* was significantly shorter and more retrognatic in relation to the anterior cranial base, while the inclination of the maxilla in relation to the anterior cranial base was normal.

The *mandible* was of normal size and prognathism in relation to the anterior cranial base. The gonial angle and the inclination of the mandible in relation to the anterior cranial base were somewhat decreased, but these differences were not statistically significant.

The mean *sagittal jaw relationship* was negative (−2.0) and significantly decreased, while the *vertical jaw relationship* was also somewhat decreased, but this difference was not statistically significant. The mean *total facial height* (n-gn) was significantly shorter (about ½ cm) in female carriers than in controls.

As for the soft *tissue profile*, it was observed that the *glabella region* was significantly more protruded in the female carriers; whereas the *facial convexity* and the shape and size of the nose were within normal limits. The *naso-labial* angle was decreased, but this difference was not significant. Both the *upper and lower lips* were significantly retruded in relation to the *nose-chin line* (*ls-NCL; li-NCL*). The mean soft tissue total facial height was somewhat shorter, but this difference was not significant.

As mentioned above, the female carrier group was sub-divided into two groups according to the number of missing permanent teeth. Five females were excluded from this part of the study, as the cause of the missing teeth was unknown (agenesis, extraction). Most females (n = 21) belonged to the group with less than or equal to four missing permanent teeth. The only significant difference in facial morphology between the two groups was that the length of the *nasal bone* was smaller in the group with agenesis of more than four permanent teeth ($p = 0.01$), but there was a tendency towards reduced anterior facial height in the group with agenesis of more than four permanent teeth, but the difference was not significant.

Gender difference in cephalometric findings

Figure 3 illustrates the superimposed mean diagram for the affected males and female carriers, and, for comparison, a similar superimposition of the mean diagrams for the male and female control groups. In addition, the angular variables for the affected males and female carriers are presented in Table 4. The major differences between the affected males and female carriers were that affected males had anterior

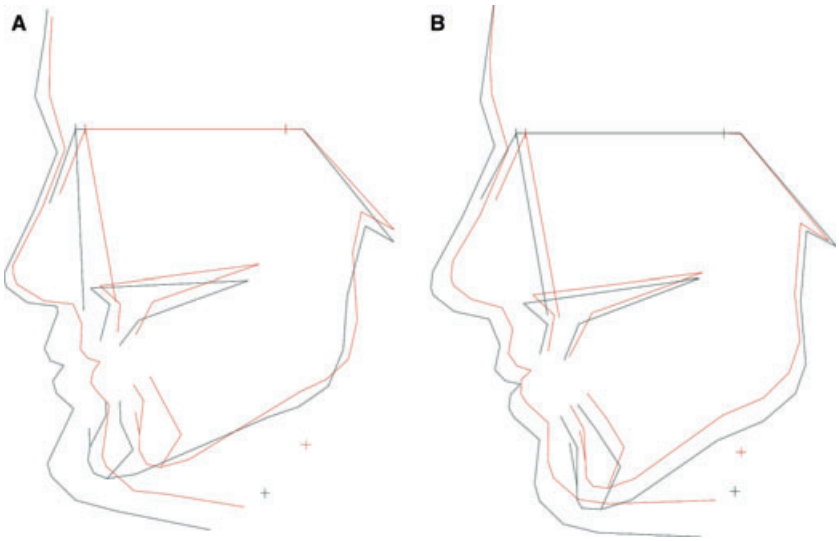

Fig. 3. (A) Mean tracing of the affected males (green) superimposed on the mean tracing of the female carriers (red). (B) Mean tracing of the male control group (green) superimposed on the mean tracing of the female control group (red).

Table 4. **Cephalometric measurements of affected males and female carriers**

	HED affected males			HED female carriers			
	Number of subjects	Mean	SD	Number of subjects	Mean	SD	P-value
Skeletal morphology							
Cranial base							
n-s-ar	10	123.0	5.0	33	125.1	3.9	NS
n-s-ba	10	128.8	5.2	33	132.1	3.9	NS
Nasal bone							
s-n-na	10	109.8	5.7	33	111.5	6.2	NS
Maxilla							
s-n-ss	10	78.9	2.7	33	78.8	4.1	NS
NSL / NL	10	2.5	4.8	33	7.4	3.1	0.01
Mandible							
s-n-pg	10	87.9	6.1	33	80.7	4.9	0.01
ML / RL	10	120.3	9.9	33	119.7	7.4	NS
NSL / ML	10	20.7	7.2	33	27.9	7.7	0.01
Jaw relations							
ss-n-pg	10	–9.0	6.9	33	–2.0	4.4	0.01
NL/ML	10	18.2	5.7	33	20.5	6.5	NS
Soft tissue morphology							
Glabella							
sgs-gs-ns	10	149.6	4.6	33	163.3	6.8	<0.001
Facial convexity							
NFL / NCL	10	157.4	9.8	33	144.6	7.0	<0.001
Nose							
ns-unt / n-s	10	113.6	3.6	33	114.2	4.6	NS
Lips							
llt-sms-pgs	10	103.7	29.0	33	131.0	16.7	0.01
nst-sn-ls	10	79.3	16.1	33	105.5	14.7	<0.001

p-Values obtained by *t*-test.
NS, not significant.

inclination of the maxilla as well as a significantly more pronounced anterior inclination and prognathism of the mandible. The affected males also had a significantly more pronounced negative sagittal jaw relationship; a more protruding soft tissue glabella region, a flatter soft tissue facial profile (reduced convexity) and both upper and lower lip were significantly more protruding than they were in the females.

Discussion

Because of a highly variable clinical picture in the female carriers of the X-linked HED, there is a need to utilize several available diagnostic tools in an attempt to try and identify potential female carriers of HED clinically. Therefore, this study investigated the somatic and cephalometric findings in affected males and female carriers, with a known mutation in the *ED1*

gene. To the best of our knowledge, this is, the first study to report, in detail, on these findings in a group with a known mutation in the *ED1* gene. Furthermore, the sample size of especially the female carriers was comparatively larger than in the previous studies. However, as different types of ED (including HED) as well as males and females were pooled in previous studies it was not possible to compare our findings directly with the findings of previous studies.

When examining the somatic data in the group of affected male children and adolescents, the majority had BMI values lower than the normative mean. These findings are in agreement with the previous study of Motil et al. (9). However, in both the male adult groups and the female group in this study, it was observed that the median BMI were within the normal range according to WHO BMI classification (23). This could mean that although the affected boys have a low BMI in childhood and adolescence they catch up later in life. The explanations for this observation could be the recurrent infections in early childhood (34) and also the severe hypodontia resulting in a reduced chewing ability. It was further observed that the head circumference in affected males and female carriers was somewhat decreased, but only statistically significant in the female carriers. This observation is in line with the anthropometric study of Ward and Bixler (10). The reason for the non-significance of the head circumference in the affected male group could be the small sample size.

In this study, we hypothesized that body height, BMI and head circumference of the affected males and female carriers did not differ from normative data. However, the hypothesis regarding head circumference could be partly rejected.

Our findings of cephalometric analysis in the affected males, such as a retroclined nasal bone, a short retrognathic and anteriorly inclined maxilla, and a anteriorly inclined and prognathic mandible as well as reduced sagittal and vertical jaw relationship, and facial height are in good agreement with findings of previous studies (16, 18, 18–20, 35). Most of the findings related to the *mandibular* position could be explained by the extensive number of missing permanent teeth as similar findings have been reported in patients with advanced hypodontia without an ED diagnosis (35–37). The short retrognatic and anteriorly inclined *maxilla* could probably, to a certain extent, also be explained by the missing teeth, as somewhat similar deviations were previously reported in persons with hypodontia (35, 36, 38), However, the maxillary deviations could also be part of a more extensive anomaly pattern of the midface in HED with the retroclined nasal bone and flattening of the nose. The observations regarding the retrusive midface, including the malar region and retroclined nasal bone in HED were previously reported by Bixler et al. (16) and Bondarets and McDonald (35). The reduced sagittal jaw relationship in the affected males could be explained by the combination of a retrognathic maxillary and prognathic mandible and similar observations were previously made in persons with advanced hypodontia (35–37). Our study showed that the overall size and shape of the mandible in the affected males and female carriers were normal. In contrast, Johnson et al. (20) suggested that the mandibular length was decreased in affected males. This difference could be because of sampling differences and differences in the variables measured. The observation that the mandible was of normal size and shape, while the maxilla was reduced in size could be explained by the different growth mechanisms in the two jaws. Mandibular growth occurs primarily as endochondral growth at the mandible condyles, while the maxillary growth is characterized by sutural growth (39).

In the female carrier group, it was observed that the maxilla was short and retrognathic, and the sagittal jaw relationship and total facial height were reduced. A retrusive maxillary region in female carriers was reported by Saksena and Bixler (17). The cranial base angle was significantly increased in the female carrier group in the present study. This finding has not been reported before in studies of affected males and female carriers; however, it has been reported in studies of individuals with extensive hypodontia (36). The flattened cranial base could probably explain part of the retrognatic maxilla in our female carrier group (39).

As mentioned before, several of the findings in males affected with the X-linked HED and female carriers, in this study, have also observed in individuals with extensive non-syndromic hypodontia. However, it would seem that, some of the deviations in the facial morphology, especially related to the midface, could be specific for HED. This hypothesis is supported by a study of Montonen et al. (40) who examined the

expression of *ED1* mRNA in different tissues in foetuses and adults; they suggested that the localization of mRNA in neuroectoderm and the expanding bone could account for some of the characteristic features of the skull and facial bones in the X-linked HED.

Most of the previous studies of the soft tissue profile of affected males and female carriers have been made with anthropometric analysis (10, 14, 15). In this study, the soft tissue profile was measured by cephalometric variables. The affected males showed a somewhat more protruding glabella region, a flattened nasal root, a reduced facial convexity, a short relatively flat nose and protruding lips. The protruding lips were also observed in the HED group in the study of Bondarets and McDonald (35). The total facial height, as well as the upper facial height, was significantly reduced. This is opposite to the finding in the skeletal measurements, where the lower facial height was more reduced than the upper facial height.

In the female carrier group, the soft tissue profile was not as distinct, but they had significantly retruded lips. Retruded lips have also been observed in patients with hypodontia (35, 41). It was further hypnotized that there is a positive correlation between the severity of the deviations in craniofacial morphology and the severity of hypodontia in female carriers. Based on the present study this hypothesis was rejected.

The difference in the angular measurements, between the affected males and female carriers showed that, both skeletal morphology variables and especially, the soft tissue morphology variables were more pronounced in the affected male group. Our observations regarding the craniofacial morphology agree with our hypothesis that the affected males and female carriers differ from the normal population in the craniofacial morphology, and the deviations are more pronounced for the males.

In conclusion, this is the first study of somatic development and craniofacial morphology in a genetically diagnosed group of affected males and female carriers. The somatic analysis showed that body height is within normal limits, while BMI would seem to be in the low normal range for boys in childhood and adolescence and within normal range for adults. Head circumference was significantly decreased in the female carriers. The observed deviations in craniofacial morphology for the affected males were in good agreement with the findings of previous studies where the genotype was not established. This situation probably reflects that the distinct clinical phenotype of this disorder makes the diagnosis of affected males reliable. Most of the findings in the affected male group could probably be explained by the extensive number of missing permanent teeth, but the flat midface with the retroclined nasal bone, retrognatic maxilla and the flattened short nose might be caused by the gene defect. The female carriers showed, as a group, rather subtle deviations in craniofacial morphology, but probably none of the deviations can be said to be syndrome specific. Therefore, we cannot at the present time suggest any specific cephalometric biomarkers, which could aid the clinical identification of potential female carriers of HED.

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