

The Development of the Curve of Spee in Australian Twins

Submitted in partial fulfilment for the degree of Doctor of Clinical
Dentistry (Orthodontics)

by

Antonio Gagliardi

B.H.Sc, B.D.S, B.Sc. Dent (Hons).



THE UNIVERSITY
of ADELAIDE

School of Dentistry
Faculty of Health Sciences
The University of Adelaide
South Australia
5005

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Summary

The objectives of this study were to investigate the development of the curve of Spee in Australian twins as well as to quantify the genetic contribution to the shape of the curve of Spee. The material used in the following study is part of an ongoing project at the University of Adelaide, investigating teeth and faces of twins. The sample investigated comprised pairs of Australian twins from the primary dentition stage through to the permanent dentition stage.

Dental study models of the primary (T1), mixed (T2) and permanent (T3) dentitions for each twin pair were mounted and photographed. Landmarks were then digitized and a 2-dimensional interpretation of the curve was analysed. Linear distances were then taken as a representation of the depth of the curvature. By digitizing each landmark, orthogonal polynomials were then fitted to the curve to allow a description of the shape of the curvature. To further investigate the genetic contribution on the development of the curve of Spee a classical twin model was used, broad sense heritability estimates were derived to quantify the extent of genetic contribution to the observed phenotypic variation.

The result indicated that the greatest change in the depth of the curve of Spee occurred between mixed and permanent dentitions while the primary to mixed dentitions showed a relatively flat curve. Depth changes were found to be larger in males during the transition to a permanent dentition. Heritability estimates indicated that there is a moderate to high genetic influence on the phenotypic variation of the curve of Spee.

Declaration

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Dr Antonio Gagliardi

Dated

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Section 1

Literature Review

4.1 Historical Background

The curve of Spee was first described by the German embryologist, Ferdinand Graf Von Spee. His original 1890 publication entitled, “The Gliding path of the mandible along the skull,” has since been translated (Biedenbach *et al.* 1980) and has helped dental professionals in understanding Von Spee’s original description, as the curve has been described and interpreted differently by many.

Von Spee defined the curve in humans by constructing an arc which runs from the anterior surfaces of the mandibular condyles to the occlusal surfaces of the second molars and the incisal edges of the mandibular teeth. This arc lies at a tangent in the sagittal plane. The centre of this cylinder in the sagittal plane has a radius of 6.5 to 7cm. Von Spee described the forward and backward gliding movement of the mandible as taking place in a circular motion along an arc (Biedenbach *et al.* 1980) (Figure 4.1). This motion of gliding performed by the mandible can also occur with a slight deviation to either the left or right during function.

The skull profile in Figure 4.1 shows the occlusal surfaces of the molars which are aligned in a downward convex curve in the maxilla and an upper concave curve in the mandible (Biedenbach *et al.* 1980).

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Figure 4.1. The curve of Spee (Biedenbach et al. 1980)

Figure 4.1 also displays the effect of attrition, whereby the molars have lost their cusps and allowed the upper and lower arches to fit together in a smooth curved line (Biedenbach *et al.* 1980). Looking closely at the material used by Von Spee, it can be seen that the skulls he assessed in his paper displayed extensive dental wear.

Hitchcock (1983) clinically described the curve of Spee as the result of the distal marginal ridges of the posterior teeth and the incisal edges of the central incisors. Wheeler (1940) believed the curve exists in occlusal balance and is dependent on the individual curvature of the teeth, taking into account crown shape. Upper and lower teeth with completely intact crown structure also appear to have the same curve-like relationship of their occlusal surfaces, although the curvature can be visualised with greater ease in a worn dentition (Biedenbach *et al.* 1980).

Von Spee described the following findings from his examination of the occlusal curvature (Biedenbach *et al.* 1980).

1. *The total visible contact line of the molar occlusal surfaces was on the same arc of a circle.*
2. *Within the sagittal plane, the posterior continuation of the arc will touch the most anterior point of the condyle of the mandible.*

Monson (1932) developed a spherical theory, based on the description of the curve of Spee. He considered the ideal arrangement of the teeth within the dental arches, describing the occlusal curvatures not only in the sagittal but also the frontal planes, therefore giving rise to Monson's spherical geometry. He described two schools of thought in which one considered that shape and movement of the condyles dictated the occlusion of the teeth. The second concept described the occlusion of the teeth as the dominant guiding factor which also determined the shape and the movements of the condyles in the glenoid fossa. Monson's curve is defined as a series of planes as can be seen in Figure 4.2. In which the upper molar buccal cusps lie slightly higher to the lingual cusps and the cusps of the upper second premolars are flat and on the same plane with the buccal cusps of the upper first premolars lying lower to the lingual cusps (Freer, 1999).

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Figure 4.2. The Curve of Monson (Freer, 1999)

4.2 Significance of the curve of Spee

The curve of Spee is retained as a relatively constant structure through life. Monson's curve, however, tends to reverse with age and increased occlusal wear. Mathematical models have been used to describe the effects of changing the direction of the bite force and the consequences on the mandibular joints. Baragar and Osborn (1987) used mathematical models to investigate the sagittal plane. They found that the molar long axis was orientated in a way that would result in the most efficient use of muscle force. The orientation of the masseter muscle appears to be suited to the direction of crushing forces between molars in the posterior region which tend to be mesially inclined with forward facing occlusal surfaces. They described these as 'work efficient' angles, which alludes to the most efficient part of the curve of Spee being its posterior portion and that raising the occlusal plane increased the efficiency of the muscles. The authors noted that this increase in the molar region was 7% with a 20% increase in the incisor region, which helped quantify possible reasons for the upward

tilt of the dentition anteriorly. It was also believed that the orientation of the dentition developed during the tooth eruption (Figure 4.3).

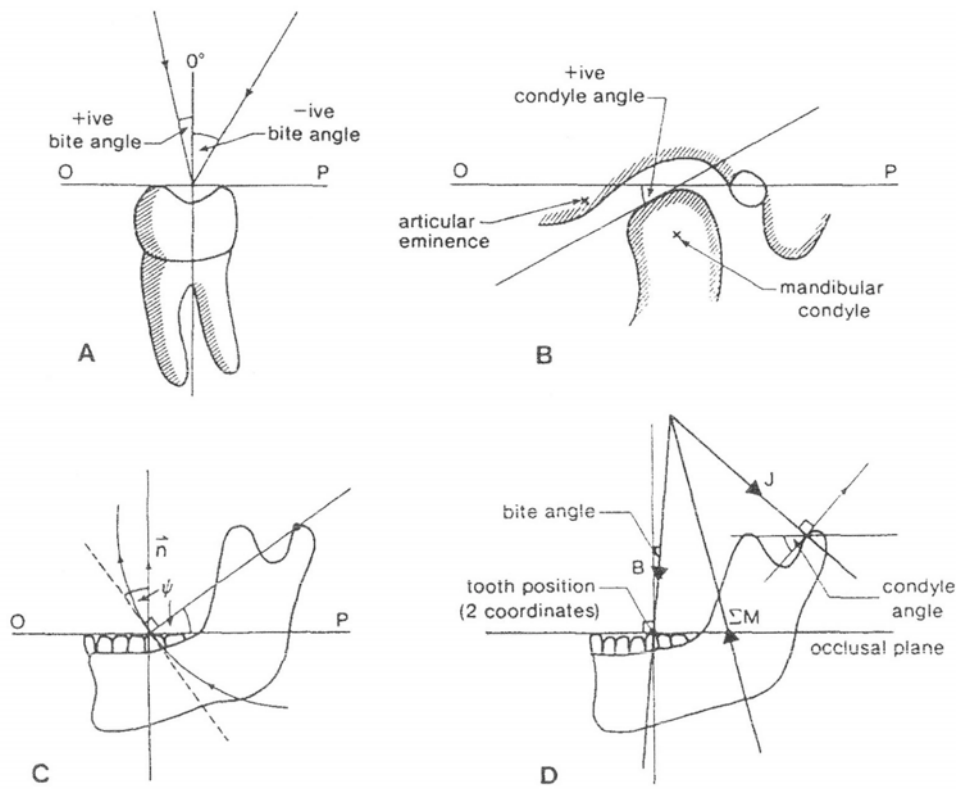


Figure 4.3. Positive and negative angulations of the dentition (Baragar and Osborn, 1989)

Baragar and Osborn (1987) suggested that an increase in the crush-shear ratio of the posterior dentition would, therefore, maximise the efficiency of the occlusal forces during mastication. Therefore, the growth and development of the curve of Spee may be related to a combination of factors which includes the growth of the orofacial structures, eruption of the teeth and the development of the neuromuscular system (Marshall *et al.* 2008).

It has also been proposed that the mandibular sagittal and vertical positions relative to the cranium have an influence on the curve of Spee. In humans this can be reflected in facial types; for example, brachyfacial patterns tend to display an exaggerated or

increased curvature (Marshall *et al.* 2008). This again supports the idea of the muscular effects of on the development of the curve of Spee, whereby brachyfacial types tend to display higher muscular tonicity when compared to dolicofacial types with a more flaccid musculature. Wylie (1944) also supported this observation, describing changes in the curve of Spee in humans in relation to depth of curvature being commonly found in brachyfacial patterns.

Osborn (1993) also alluded to this idea in his primate studies suggesting that the curvature is under the influence of the masseter muscle and its delivery of crushing forces to the molar teeth. Similarly, Salem *et al.* (2003) looked at mandibular morphology and its relationship to the antegonial notch and the curve of Spee and found that an increased curve is associated with a shortened mandibular body, a feature of brachyfacial types.

4.3 Modern interpretations and clinical significance of the curve of Spee

In modern orthodontics, the curve of Spee refers to the natural progression upwards of the curvature of the teeth from the incisors through the premolars and molars. Freer (1999) describes the average curve as flat and not exceeding a depth of 1.5 mm. It is recommended that a relatively flat curve is best suited to normal occlusion as described by the six keys of a normal occlusion. Leveling of the curve of Spee has become a universally adopted treatment goal by clinicians as a flat occlusal plane allows Angle class I canine relationship and interdigitation to be attained with relative ease by reducing occlusal interference.

These modern ideas for a stable occlusion may be attributed to Andrews (1972) in his description of the 6 ideal characteristics of normal occlusion. The curve of Spee was found to range from flat to mild, as it gave the best interdigitation. Following orthodontic treatment the curvature tends to deepen; therefore, by treating to a flat or reverse curvature allowances can be made for changes following treatment.

Andrews (1972) six keys include the following:

1. *Molar Relationship*

- *The mesiobuccal cusp of the maxillary permanent first molar contacts and occludes with the buccal groove of the lower first permanent molar.*

2. *Crown Angulation*

- *Refers the mesiodistal tip and long axis of the crown.*

3. *Crown Inclination*

- *Labio/bucco-lingual inclination and long axis of the crown. Represented by + and – ve degrees of torque to a line perpendicular to the occlusal plane.*

4. *Rotations*

- *Teeth should be free of undesirable rotations.*

5. *Tight Contacts*

- *No space should be present at the contact points.*

6. *Occlusal plane*

- *Flat to mild curve of Spee.*

In modern clinical orthodontics the leveling of the curve of Spee is part of all treatment regimes. Leveling, as described by Carcara *et al.* (2001), involves bringing the incisal edges of the anterior teeth and the buccal cusps of the posterior teeth into a horizontal plane alignment. By leveling the curve of Spee, orthodontic treatment objectives and occlusal ideals may be achieved. This treatment goal of a flat occlusal

plane was outlined initially by Andrews (1972) in his observations. Andrews (1972) described a deep curve as one which did not facilitate a normal occlusion, as the upper teeth were contained in a smaller area.

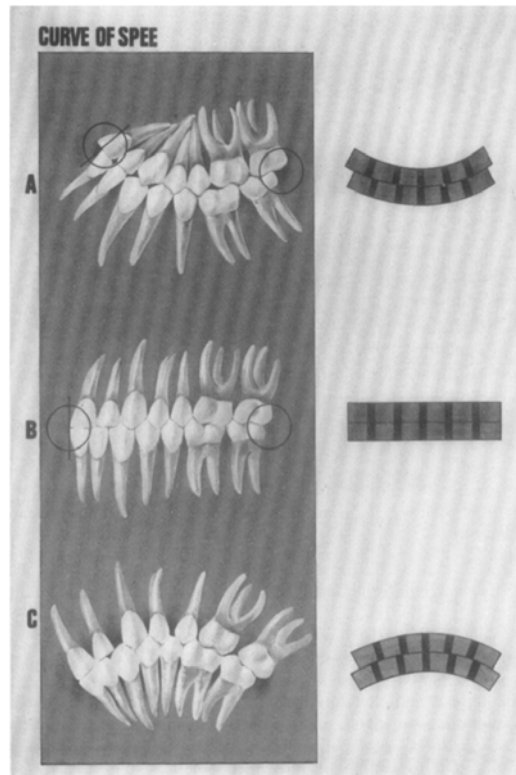


Figure 4.4. Andrews description of the curve of Spee

The above diagram (Figure 4.4) taken from Andrews (1972), indicates the orthodontic relevance of the curve of Spee to the clinician. Andrews' descriptions give the extreme ends of the spectrum.

A: A deep curve of Spee resulting in a more confined area for the upper teeth, therefore creating a spillage in both the mesial and distal directions, with a proclination of the anteriors and distal tipping of the posterior teeth.

B: A flat plane of occlusion is conducive to achieve orthodontic treatment goals, such as the correct tooth angulations and interdigitations.

C: A reverse curve of Spee results in excessive room for the maxillary teeth. This can be seen diagrammatically as a spacing of the upper teeth and crowding in the lower arch.

In a clinical environment this has been observed in different occlusal types. For example, AlQabandi *et al.* (1999) attributed a deep bite to an exaggerated curve of Spee. In order for a deep bite to be successfully treated, the exaggerated curve must be flattened by the intrusion of the anterior segment and extrusion of the posterior segment to varying degrees. As a result, arch length dimensional changes will lead to an increase in arch length and proclination of the lower labial segment or crowding within the dental arch. Alexander (2001) similarly described the importance of leveling in orthodontic treatment and outlines the usefulness and success of leveling in opening bites while also noting its relative stability.

Bishara *et al.* (1989) reviewed changes in mandibular and maxillary arch length over time and found that the curve of Spee remains relatively stable post-adolescence in their non-orthodontic sample. This has not been the case with treatment of deep and open bites where some degree of relapse and changes to the curve of Spee post treatment have been described. Sengupta *et al.* (1999) considered the effects of dental wear on the eruption of third molars and its influence on the curve of Spee and noted that the curve of Spee was not maintained with ageing and dental wear and that this

would have clinical implications, particularly in the area of prosthodontics. Orthodontists manage the curve of Spee on a daily basis and aspects of long-term stability must be considered. De Praeter *et al.* (2002) investigated the long-term stability of orthodontic leveling of the curve of Spee and reported that a stable curve is shown to be observed during orthodontic treatment and remains relatively stable post-treatment. Carcara *et al.* (2001) was also unable to predict relapse but found that a completely level post-treatment curve of Spee had a significantly reduced chance of relapse. When considering relapse, occlusion must also be considered, as the degree of relapse will vary depending on the original occlusion as well as face and muscular types.

In contrast to Andrews, Lie *et al.* (2006) examined the curve of Spee in 135 post-treatment subjects. Utilising cephalometric measures, they were able to describe a 40-48% stability post-treatment. They found that the curve of Spee was most unstable at its deepest point following treatment and the smaller changes displayed less stability than a curve that has undergone greater change. They concluded that a final curve (post-treatment) of 1.9 mm depth provided the highest incidence of long-term stability, therefore alluding to the existence of the optimal curve rather than a flat curve.

Preston *et al.* (2008) investigated the treatment of Angle Class II Division I subjects by two different approaches. The long-term effectiveness of leveling was examined and similar results were found in both groups, leading to greater relapse potential if the curve of Spee was not completely level post-treatment. The above studies indicate

an underlying biological or functional need of a curvature as there is a tendency for the curve to re-establish itself through biological influences.

The function of the curve of Spee has been considered previously. Baragar and Osborn (1989) considered the efficiency of mastication and described the curve as contributing to the efficiency of occlusal forces during mastication. This efficiency is increased by the greater crush-shear ratio of the posterior teeth.

Furthermore, Carcara *et al.* (2001) described an imbalance of the posterior and anterior teeth leading to the curvature. This imbalance would result in lower incisor overeruption, premolar infra-eruption and increased mesial angulation of the molars. The curve of Spee, therefore, may be considered as having an evolutionary biological role and an alteration of the curvature in a biologically dynamic environment would result in a re-establishment of the curve to its functional role.

4.4 Measuring the curve of Spee

Wheeler (1940) considered the curve from the perspective of occlusal balance, describing the curve as establishing this balance. Observations were based on examination of the crown of dissected specimens with the curvature of each individual tooth taken into account. Contrary to this, Von Spee's original work described the curve as being present in the worn dentition.

Braun *et al.* (1996) employed a measuring device and computer to accurately determine the arch circumference of 27 dental casts with a moderate to severe curve

of Spee, concluding that arch circumference reduction is low, therefore implying that the lower incisor protrusion is not primarily due to arch length changes but rather the treatment mechanics. Leveling of the curve of Spee has been criticised for its unwanted effects on lower incisor proclination; however, it has been found that orthodontic treatment has no significant effect on the flaring of lower incisors (Woods, 1986).

Previous authors have attempted to measure and describe changes that occur in the curve of Spee during treatment and normal development. Ferrario *et al.* (1999) investigated the dental casts of 50 adults aged between 19-22 years, and the dental casts of 20 adolescents aged between 12 to 14 years. Using three co-ordinates of cusp tips and a 3D digitizer, a spherical model was derived to describe the occlusal curvature. With these measurements, it was possible to assess associated dental changes from adolescence to adulthood. It was found that the radius of the curves of Spee, Monson and Wilson changed approximately 20 mm between the two age groups from 80 mm in the adolescent to 101 mm in the young adult group. This process was influenced by a progressive rotation of the major axis of teeth which moved the occlusal plane to a more buccal position. These findings may provide an explanation for the relapse of the curve of Spee observed post-orthodontic treatment.

Similarly, Cheon *et al.* (2008) used a 3D method to describe changes in the curve of Spee and dentofacial morphology. Dental models and lateral cephalograms were used to determine the depth of the curve using virtual models and computer software. They observed that the depth of the curve was significantly related to overbite, overjet and

the sagittal position of the mandible with respect to the anterior cranial base. No differences were observed between the sexes.

Marshall *et al.* (2008) recently described the development of the curve of Spee using 16 male and 17 female subjects from the Iowa Facial Growth Study. The depth of the curve of Spee was measured at seven time points for each subject with the relative eruption of the mandibular teeth and contribution to the curve being examined with lateral cephalograms. It was concluded that the curve of Spee initially developed as a result of the mandibular permanent first molar and incisor eruption. It maintained its depth until the mandibular permanent second molars erupted, after which the curvature deepened. Once the adolescent dentition was established, the curve deepened only slightly and remained relatively stable into early adulthood. This observation was also described by Ferrario *et al.* (1999).

Some authors have determined the depth of the curve of Spee as a measure of perpendicular distance (Figures 4.5 & 4.6). Marshall *et al.* (2008) recently described a method of evaluation which is based on a 2D analysis model.

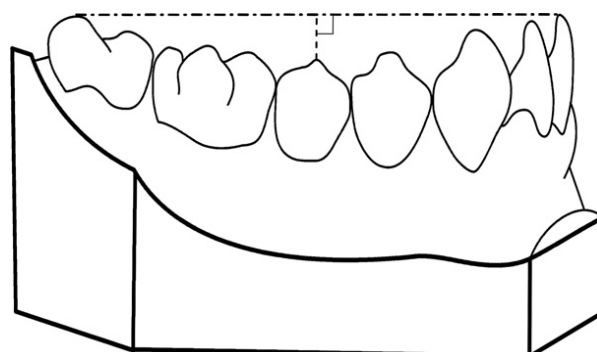


Figure 4.5. Measurement of the maximum depth of the curve of Spee (Marshall *et al.* 2008).



Figure 4.6. Digital calipers vertically mounted on a dental surveyor used to measure the maximum depth on leveled study models as described in Figure 4.5. (Marshall *et al.* 2008).

Similarly, other authors have approached the description of the curve of Spee from a 2D perspective. AlQabandi *et al.* (1999) and Baydas *et al.* (2004) used a perpendicular measure similar to that of Marshall *et al.* (2008) in which a perpendicular line was measured to the deepest cusp tip from the occlusal plane, constructed from the incisal edges to the most distal cusp tips (Figure 4.5).

Bishara *et al.* (1989) similarly analysed the curve of Spee from study models. The method utilised differed from that of other authors as more than one reference line was used and an average was taken of the perpendicular lines. Tooth and arch dimensions were also investigated.

Similarly, Dager *et al.* (2008) also considered the long-term changes to the dental arches and the curve of Spee and reported on a dynamic system which is constantly changing and adapting well into the sixth decade of life, although the degree to which these changes occur will decrease with time. Dager *et al.* (2008) measured the curve of Spee from dental casts using digital calipers. A flat plane was constructed over the incisal edges of the mandibular incisors to the mandibular first molar cusps and perpendicular distances were then measured from this plane to the most inferior aspect. This was repeated on both sides and an average taken. Various measuring devices have been developed and adapted for research purposes, such as the precision co-ordinate measuring machines shown in Figure 4.7.

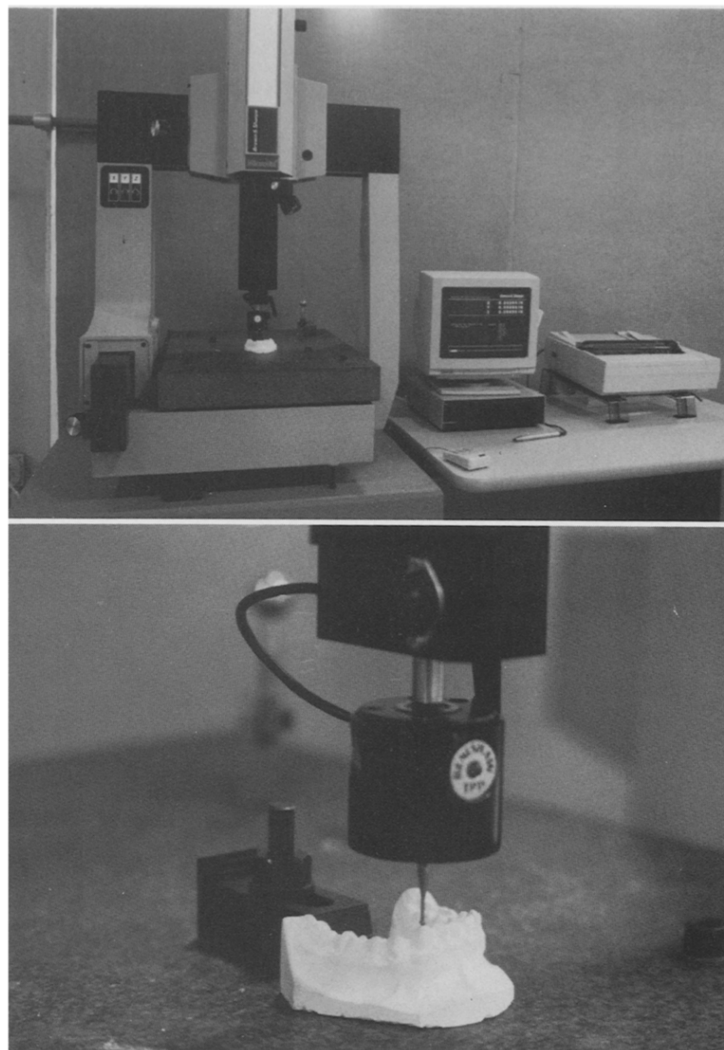


Figure 4.7. Precision co-ordinate measuring machine (Braun *et al.* 1996).

Braun *et al.* (1996) measured the depth of the curve of Spee from a flat plane which was formed by the tips of the mandibular incisors anteriorly and the distal cusp tips of the second molars. The maximum depths of both the right and left sides were taken using a Brown and Sharp Precision Co-ordinate measuring machine (Figure 4.8) where the touch trigger probe has an accuracy of 0.006mm.

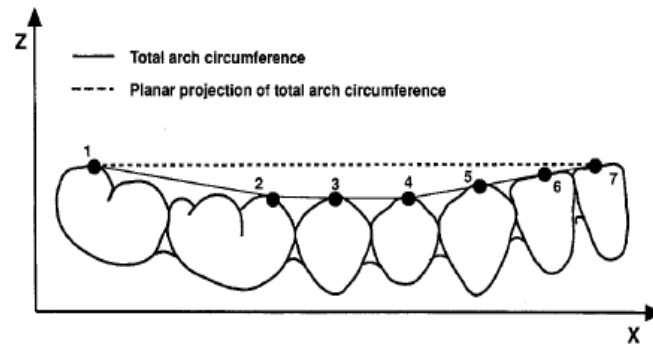


Figure 4.8. 2D view of total arch circumference in relation to planar projection (Braun *et al.* 1996).

Similarly, Shannon and Nanda (2004) utilised similar reference points and methodology and compared the changes in the curve of Spee during treatment and 2 years post-treatment. Their machine displayed an accuracy of up to 0.0025 mm, but they did not find any significant differences between males and females and, overall reported a relatively stable curve after treatment with only about 16% relapse of the levelled curve.

Photographic methods of assessment have also been proposed. De Praeter *et al.* (2002) utilised photographs of study models and the creation of reference lines from the incisal edge of the central to the distal cusp tip of the first molar. This reference line served as a reference point from which perpendicular reference planes could be constructed to the lateral incisor, canine, first and second premolar and the mesio-buccal cusp of the first molar. These lines served as a means of measuring the depth of the curve.

This photographic approach was also used by Farella *et al.* (2002). In their study, the distal cusp of the second molar to the cusp of the canine was used to create a reference plane forming the x axis. The y axis was constructed at the midpoint of the x axis as a perpendicular (Figure 4.9). These co-ordinates were then mathematically analysed and were correlated to other facial features. Weak correlations were reported between posterior face height, anterior facial height ratio and the curve of Spee. They found a deeper curve to be evident in short-faced individuals compared to shallow curves in long-faced individuals, indicating a possible difference between dolicofacial and brachyfacial types. Strong correlations were found to exist between the position of the condyle and the occlusal plane. A reduced curve was evident in subjects whose dentition was more anteriorly placed in relation to the condyle.

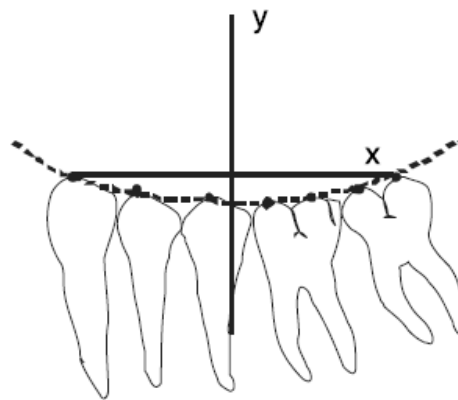


Figure 4.9. X-Y axis reference lines used for measurement of the curve of Spee (Farella *et al.* 2002).

Cephalometric measures have also been used by many authors to investigate the curve of Spee. Bernstein *et al.* (2007) employed the use of lateral cephalometric radiographs to create a perpendicular for curve depth analysis, similar to methods utilising study models.

Lie *et al.* (2006) used standardised photographs of mandibular study models out of occlusion, from the left and right sides, with the buccal tooth surfaces aligned in the same plane. These photographs were taken in a plane perpendicular to the occlusal plane tangent to the buccal surface of the first molar and canine and centered on the first premolar. A ruler was included in all photographs to determine magnification. Fixed landmarks on the photographs were then digitised and measurements were carried out for each model of the left and right curve.

In a study which took into account both arches, Xu *et al.* (2004) reported the maxillary arch possessed a flatter curve compared to the mandibular arch. Comparisons were made by using digital images of dental casts which were then measured and analysed. Similarly, Liu *et al.* (2006) measured perpendicular distances from an occlusal reference plane from the distal cusp tip of the last molar to the mid-incisal edge of the lower incisor. A ruler was used in each photograph to determine magnification. The deepest point marked by the lowest cusp tip was taken from the reference plane and measured. The following criteria were used to define the curve as either flat (<1mm), normal (1-2 mm) or deep (>2mm).

The curve of Spee has been interpreted differently by many authors and this is reflected in their approach to quantification. Opinions and descriptions range from that of the anatomist to the clinician. This broad approach may be related to the goals that each author seeks in quantifying and altering the curvature. One deficiency is the physiological reason of why the curve of Spee exists and of what purpose it serves.

Authors have been exploring the possibility of 3D methods of investigation. Ferrario *et al.* (1999) analysed digitised dental casts, excluding the third molars with three coordinates of cusp tips, recorded with a three-dimensional digitizer and used to derive a spherical model of the curvature of the occlusal surfaces. From the best interpolating sphere, the radii of the left and right curves of Spee (quasi-sagittal plane) and of the molar curve of Wilson (frontal plane) were computed.

In a more recent study, Cheon *et al.* (2008) described a method of evaluating the curve of Spee and dentofacial morphology using computer software, 3D models and lateral cephalograms. The sample consisted of 18 Korean men and 31 Korean women with no history of orthodontic treatment, TMD and crossbites with minimal crowding and spacing. The measurements of the curve of Spee were taken from the occlusal plane and to the deepest perpendicular point to the buccal cusp tip and these distances were averaged between the right and left sides. Cheon *et al.* (2008) defined the occlusal plane as the midpoint of the centres of the right and left incisor edges as well as the distobuccal cusp tips of the right and left molars, these landmarks have served as the basis of most of the analyses of the curve of Spee. Figure 4.10 outlines a suggested approach for 3D analysis by Cheon *et al.* (2008),

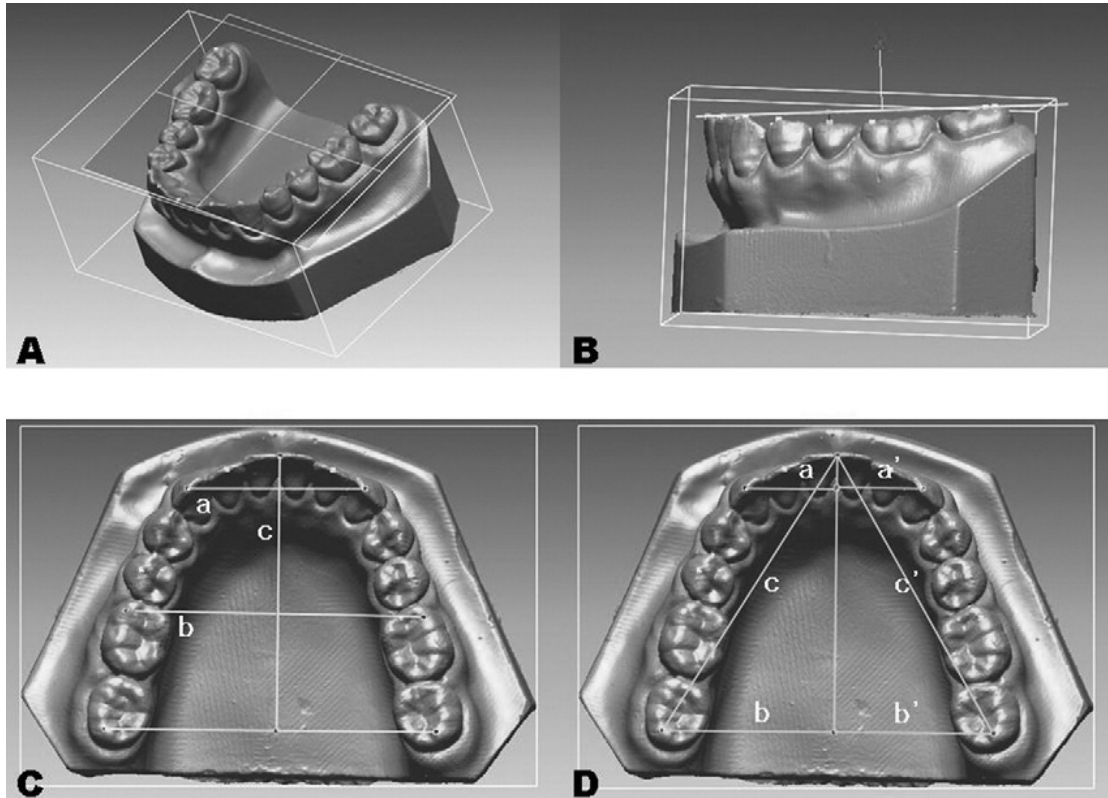


Figure 4.10. Reference plane descriptions. (Cheon *et al.* 2008).

Reference plane descriptions

- A. Occlusal plane
- B. Curve of Spee (deepest point from the occlusal plane to buccal cusp tip of each lateral tooth)
- C. Intercanine width (a), Intermolar width (b), arch length (c)
- D. Dental arch form (symmetry of the dental arch evaluated by using the perpendicular distance at the canine (a) and the second molar (b) and the distances from the midpoint of the right and left incisor edge to the tips of the right and left second molar distobuccal cusps (c))

Cheon *et al.* (2008) reported that a correlation existed between the depth of the curve of Spee with SNB and ANB angles, suggesting that the radius of the curve of Spee was shorter in class III malocclusions. Overbite, overjet and the depth of the curve of Spee were also shown to be correlated by which an increase in the overjet and overbite showed an increase in the depth of the curve of Spee. The position of the mandible was also found to have a positive correlation, in that the further posteriorly the mandible was positioned the greater the depth of the curve of Spee. Imtiaz *et al.*

(2011) reported on the influence of malocclusion and found that the curve of Spee was deepest in class II division I malocclusion and a flat curve of Spee was observed in class III malocclusions.

Dental arch changes have been considered to cause changes in the curve of Spee depth, reports have suggested that 1 mm of arch circumference is necessary for every millimetre of curve depth (Graber, 1994; Bishara, 2001). Eskine (1992) employed a geometric approach to investigate the arch requirements of leveling and concluded that a shorter arch length will require greater space and that less than 1 mm of arch length is required for 1 mm arch levelling; therefore, a non-linear relationship existed.

Germane *et al.* (1992) employed the use of a mathematical model to help determine the arch requirements that will be necessary to flatten the curve of Spee. From their work it was suggested that 1 mm in the reduction of the curve's depth would increase the circumference of the arch by 0.06mm and a depth of 9 mm would be equivalent to an increase of 4.55 mm of arch circumference.

The literature describes diverse approaches to interpreting and measuring the curve of Spee. Some of this may be attributed to the differing interpretations of Spee's original publication. The methods used by authors range from photographic, study models, radiographic to 3D dimensional scans. The ability to provide a universal standardisation approach varies due to the diverse definitions and approaches. While most literature tends to focus on the mandibular arch it would be beneficial to investigate the relationship between both arches and difference, which may exist between the two curvatures.

Clinicians and anatomists differ in their interpretation, as most clinicians would regard the curve of Spee as being isolated to the dentition only whereas an anatomist may take into account the condyles and their function. A deficiency in clear universal definitions and methodological variation indicates a deficiency in understanding the development of the curve of Spee and the possible biological effects and implications of levelling during routine treatment.

4.5 Twin Models

Currently, there appears to be no literature which considers underlying genetic or epigenetic influences on the curve of Spee. Various models have been used in the literature to assess twin data for particular biological features and to gain an insight into genetic, epigenetic and environmental contributions to phenotypic variation. The following are the most common models used (Townsend *et al.* 2009);

1. ***The traditional or classical twin model-*** *this model can be used to assess the phenotypic variation between monozygotic (MZ) and dizygotic (DZ) twins and the contributions of genetic and environmental factors.*
2. ***The MZ co-twin model-*** *this model enables the comparison of MZ twins who are discordant for a selected feature; for example, a missing tooth. It allows insights into both environmental and epigenetic contributions.*
3. ***Twins reared apart model-*** *this model aims to minimise common environmental effects. Assuming twins are separated shortly after birth and reared in separate homes, they are not influenced by a common family environment. Therefore, any similarities in MZ co-twins can be attributed to genetic influences.*

4. ***The analysis of twins and family members (parents and siblings)***- this approach aims to further establish genetic and environmental contributions to phenotypic variation.
5. ***The MZ half-sibling model***- this model considers the genetic contributions by looking at half-siblings. This approach involves studying genetic and environmental contributions to variation of MZ twins, their spouses and offspring. The children of MZ co-twins are genetically half-siblings. This therefore offers the opportunity to detect maternal effects.
6. ***The DZ opposite sex model***- this model considers the hormonal effects in-utero. This approach focuses on male-female twin pairs and tests whether there are differences in mean values and variances for selected features between these twins and other twin types and singletons. The different levels of hormones that co-twins are exposed to in utero may have observable effects post-natally.

The above models have been used to describe various dento-facial features in twins. These include the relationship of the maxilla and the mandible, tooth size and position as well as arch size and shape (Townsend *et al.* 2009). The effects of environmental influences on occlusal variation have been reported in the literature. For example, Harris and Johnson (1990) reported that much of the variation in the permanent dentition is acquired rather than inherited. Hughes *et al.* (2001) reported on the occlusal variation of the primary dentition of Australian twins and singletons, suggesting that the observed variation of the primary dentition can be attributed to a moderate to high genetic contribution. In the classical twin design, the extent to which

phenotypic variation in a trait is due to genetic and environmental influences can be estimated.

4.6 Describing dental arch shape – use of polynomials

A polynomial can be described as an expression of a finite length which may be constructed from a number of variables and constants. Richards *et al.* (1990) reported the use of polynomials as accurately representing the dental arch providing no irregularities or displacements were present, reporting low correlations within twin pairs and attributing a greater influence of environmental factors in arch asymmetry.

Ferrario *et al.* (1994) has used mathematical definitions to describe the shape of human dental arches. The maxillary and mandibular arches were both constructed using a fourth order polynomial to geometrically describe arch shape.

A fourth-order polynomial is represented by the following equation:

$$Y = a + bx + cx^2 + dx^3 + ex^4$$

Further to the use of simple fourth order polynomials, Hughes *et al.* (2002) have described the use of orthogonal polynomials as a robust method of quantifying dental arch shape mathematically with good results.

An equation in the form $Y = a - b_1\phi_1 + b_2\phi_2 + b_3\phi_3 + b_4\phi_4$ can be used to describe arch shape where ϕ_1 , ϕ_2 , ϕ_3 and ϕ_4 are orthogonal polynomials and b_1 , b_2 , b_3 and b_4 are coefficients. The different coefficients can be interpreted, with some describing arch shape (second and fourth coefficients) and others asymmetry (first and third coefficients). Orthogonal polynomials allow values to be assigned to the relative contributions to symmetry and asymmetry to overall arch shape. This was first

presented by Lu (1966) and has since been mathematically refined as presented by Hughes *et al.* (2002).

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Figure 4.11. Arch shape described by an orthogonal fourth order polynomial (Hughes et al. 2002)

Figure 4.11 derived from Hughes *et al.* (2002), illustrates how arch shape can be described by an orthogonal fourth order polynomial, with antimeric points joined to illustrate the degree of asymmetry.

Section 2

5. Statement of Purpose

It is generally understood that the development of the curve of Spee is relatively flat in the primary dentition but develops more curvature during the transition from the primary to the permanent dentition.

From the literature it can be seen that various methods have been used to quantify the curve of Spee. These methods range from common 2D approaches measuring either directly from casts or digitized images to the beginnings of 3D analysis. There is also a deficiency in our understanding of the role of genetics, as most authors are attempting to describe the curve of Spee from a single linear arbitrary measurement. The purpose of this study is to further our knowledge of the development of the curve of Spee by adapting previous methods of quantification while introducing some new approaches from a genetic perspective. The current study will be one of the first to investigate genetic influence using a twin model. The curve of Spee will be first quantified by the common 2D approach of a linear measure for depth. This will be taken as an average of the depth at various points along the curve rather than a single maximum depth. As well, shape coefficients will be used to describe the curvature by using digitized cusp tip landmarks. In paper 2 the underlying genetic influence will be considered by using a monozygotic (MZ) and dizygotic (DZ) classical twin model approach.

This study will allow confirmation of interpretations proposed within the earlier literature but also provide genetic perspective.

6. Aims of proposed study

6.1 Paper 1

- To investigate the development of the curve of Spee during the primary dentition, mixed dentition and permanent dentition
- To describe sex differences
- To fit orthogonal polynomials to curves and interpret coefficients

6.2 Paper 2

- To use a classical twin model to describe the differences between monozygotic (MZ) and dizygotic (DZ) twin pairs, to clarify the role of genetic and environmental contributions to variation in the development of the curve of Spee.
- To use an MZ co-twin model to illustrate differences in the curve within twin pairs.

Whereas previous studies have utilised singleton samples, the following papers assess samples comprising both monozygotic (MZ) and dizygotic (DZ) twins at three time points including the primary dentition, early mixed dentition and permanent dentition. By using a twin model we have a unique opportunity to observe genetic influences on the curve of Spee.

The present studies will be amongst the first to describe and quantify the relative contributions of genetic influences to observed variation in the development of the

curve of Spee. The findings may have implications from a clinical perspective, particularly with regard to orthodontic treatment philosophy.

The following papers are set out in the form for publication; the papers will be submitted to HOMO Journal of Comparative Human Biology.

6.3 Hypotheses

Paper 1

- It is hypothesised that the development of the curve of Spee remains relatively flat in the primary dentition but develops more curvature during the transition from the primary to the permanent dentition.

Paper 2

- It is hypothesised that the curve of Spee will display a genetic contribution to variation in its final depth and shape.

7. Materials and Methods

The following methodology section will apply to both papers 1 and 2.

7.1 Selection of Subjects

The material used in the following study is part of an ongoing project at the University of Adelaide, investigating teeth and faces of twins. The sample investigated comprised pairs of Australian twins, including children and young adults from the primary dentition stage through to the permanent dentition stage.

T1= Primary (Deciduous) Dentition

T2= Mixed Dentition

T3= Secondary Dentition

Initially, models for 100 pairs of twins were selected (50 MZ and 50 DZ) based on availability of models at the three different developmental stages. Inclusion criteria for the final study sample included the presence of the primary dentition, mixed dentition and early permanent dentition, as well as complete tooth eruption with no restorations, missing teeth or dental anomalies at the particular time point. Subjects who had undergone orthodontic treatment were excluded. The selection was also based on the availability of complete records. Individuals with missing time points were excluded from the sample. The breakdown of the study sample for each time point and sex is as follows; T1 N= 128 (M= 53, F= 75, MZ= 84, DZ= 44), T2 N= 99 (M= 45, F= 54, MZ= 58, DZ= 41), T3 N= 98 (M= 44, F= 54, MZ= 58, DZ= 40). The decreased number of MZ twins at T2 compared with T1 was due to unerupted permanent teeth at this stage in one or both of the twins.

7.2 Evaluation of Digital photographs

Digital photographic images (2D) were obtained utilising the following method adapted from Brook *et al.* (1999).

This system consisted of the following:

- 1) Canon EOS D50 digital camera with a 55 mm lens (*Canon Inc, Tokyo, Japan*).
- 2) Desktop Windows Based PC directly connected to the Canon EOS D50 using a Canon EOS digital utility software interface allowing remote control. These images were stored in RAW and JPEG formats.
- 3) Copy stand attached with adjustable lighting (*manufactured by Kaiser, Odenworld*).
- 4) Model clamp with a universal joint system.
- 5) Table allowing vertical adjustments to be made.
- 6) Occlusal Plane leveling jig (*Constructed by the University of Adelaide Engineering Faculty*).
- 7) Adjustable 30 mm scale ruler.

- 8) ImageJ software for image analysis (*Public domain, Java based image processing program developed by Wayne Rasband, National Institutes of Health*).

7.3 Standardisation of dental casts

A consistent and reproducible alignment of each dental cast was achieved by the use of a levelling jig which ensured consistently accurate and reproducible cast placement prior to photography.

Reference points were chosen to enable a consistent positioning of the cast via a fixed reference plane.

This dental cast positioning apparatus is shown in the following Figures 7.1-7.3.

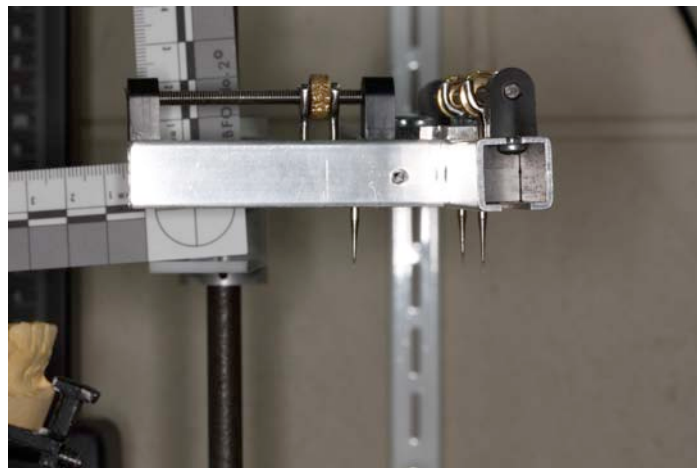


Figure 7.1. Occlusal plane leveling jig in position.

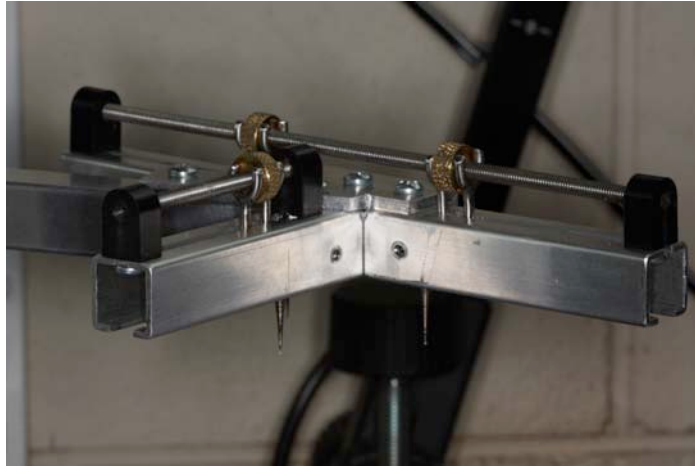


Figure 7.2. Adjustable thumb wheels to allow registration of the precision points.

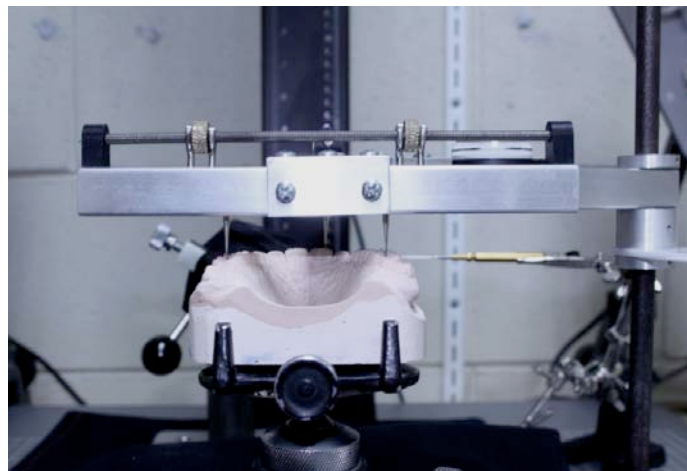


Figure 7.3. Positioning of the occlusal plane levelling jig precision points to standardidise the occlsal plane.

The device was composed of three aluminium arms positioned in a ‘T’ configuration. Each arm consisted of a thread with adjustable thumb wheel allowing the free movement of the three precision points to locate the relevant landmarks. These could be oriented to correspond to the central fossae of the lower left and lower right first molars (or deciduous second molars) and midpoint of the lower central incisor edge. The device was checked frequently between uses to ensure that it remained parallel to the camera lens. This was achieved via the use of a spirit level placed on the device.

In order to facilitate the location of the landmarks and establish a standardised occlusal plane, casts were mounted on a dental surveyor with an adjustable universal

joint. Fine adjustments were made in both horizontal and vertical planes with the aid of a spirit level and then locked in so as to ensure that all three precision points were in contact with the relevant landmarks.

Once the occlusal plane was established the device was removed from the cast which was then placed below the reference scale, and the following landmarks were lined up with the scale and camera perpendicular to the buccal line of occlusion.

- 1) **Primary (Deciduous) Dentition-** Disto-buccal cusp of lower primary second molar and the incisal tip of the primary canine were first used as the reference points. This was then repeated with the canine landmark being replaced with the central incisor. Both right and left sides were photographed.
- 2) **Mixed Dentition-** Disto-buccal cusp of the lower permanent 1st molar and the incisal tip of the permanent canine were first used as the reference points. This was then repeated with the canine landmark being replaced with the central incisor. Both right and left sides were photographed.
- 3) **Permanent Dentition-** Disto-buccal cusp of the lower 2nd molar and the incisal tip of the permanent canine were first used as the reference points. This was then repeated with the canine landmark being replaced with the central incisor. Both right and left sides were photographed.

To ensure consistency, the scale was checked between each photograph with use of a spirit level and set-square to ensure it remained perpendicular to the camera lens.

As can be seen in the following Figures 7.4 and 7.5 a study model is mounted and standardised ready for photographing, a further mounted model illustrates the positioning of the scale.



Figure 7.4. Mounted model with standardised occlusal plane, the figure illustrates the ability of the study model occlusal plane to be adjusted 360°.



Figure 7.5. Illustrates the scale used once a study model has been mounted.

The above method was applied to all dental casts to ensure lateral views and reference point location was standardised in each photograph. Camera settings and lighting conditions were also maintained to ensure all digital images were standardised.

7.4 Evaluation of Photographs

The evaluation of digital images was carried out with Image J and Microsoft Excel software programs. Image J is a public domain image processing program capable of calculating user defined areas, distances and angles. Once the scale points and landmarks including each cusp tip between landmarks was digitised in image J, they were exported into Microsoft Excel for distance calculation (mm) from the measurement plane.

7.5 Variables Studied

Rather than using a single linear distance at the maximum curvature concavity, the maximum depth of the curve of Spee was measured as the average of the sum of the perpendicular distances. These distances fell between the most posterior disto-buccal cusp tip of the mandibular molar teeth and a measurement plane described by the central incisors/canine and distal cusp tip of the most posterior tooth in the mandibular arch. This method was modified from that reported by Marshall *et al.* (2008).

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Figure 7.6. Measurement plane with cusp tip landmarks (Modified from Marshall et al. 2008).

Each cusp tip between landmarks was also digitized to allow descriptions of the curvature as viewed from a 2D buccal perspective using orthogonal polynomials.

8. Statistical Methods

Statistical parameters including the sample size (N), mean (average of a series of values), standard deviation (SD the deviation of the data values from the mean) and the range (minimum and maximum values) were calculated. One randomly selected twin from each pair was excluded in order to obtain unbiased estimates. This was done because, if there was a genetic influence on the study variables, including both members of a twin pair would mean a ‘doubling up’ of the data. A mixed linear effects model was then fitted to the data, taking account of any missing values. Two modeling approaches were then applied, the first approach modeled sex and zygosity together and the second approach modeled sex and zygosity separately. This allowed the average linear distances over time to be compared according to the side of the mouth, zygosity and sex group.

Equations based on two orthogonal polynomials were also used to quantify the curve shape mathematically, with the b1 and b2 coefficients allowing a description of curve shape and also asymmetry within the curve. The b1 coefficient allowed for the presence of any asymmetry within the curvature to be quantified, i.e. whether there was a dip more to the anterior part of the arch or more posteriorly while the b2 coefficient allowed the shape of the curve to be determined, i.e. whether the curve was deeper or shallow.

In order to investigate the presence of an underlying genetic basis to variation of the curve of Spee, heritability estimates were calculated based on a derivative of Falconer's formula (Falconer and Mackay 1996). The genetic heritability of a particular trait can be determined based on the differences between twin correlations.

$$h^2 = 2(r_{MZ} - r_{DZ})$$

The above formula describes the heritability estimate, h^2 . In addition, r_{MZ} and r_{DZ} represent the monozygotic (identical) twin and dizygotic (fraternal) twin correlations between MZ pairs and DZ pairs. Therefore by comparing MZ and DZ twins, an estimate of the strength of the genetic contribution to observed variation can be made.

9. Error of the Methods

In order to determine the extent of error in the analysis of the digital images, re-assessments of photographic images were performed. Fifteen dental casts were randomly selected at each time point for re-assessment. All casts were re-mounted and standardised as described in the above method to quantify the degree of method error. Both systematic and random errors were able to be quantified.

- *Dahlberg Statistic- to determine random error (Dahlberg 1940)*

$$Se = \sqrt{\sum d^2 / 2n}$$

Percentage of error due to variation $Se^2/So^2 \times 100\%$

Where:

$Se^2 =$ Error variance

$So^2 =$ Observed variance

- *Paired t-test- To determine systematic error*

$$t = \frac{\bar{d}}{sd/\sqrt{n}}$$

No significant differences were found, using paired t-tests, between values for first and second measurements for systematic errors. The Dahlberg statistic for the measurements of the distances between the cusp tips and the measurement plane (that quantified random error) varied from 0.1 mm to 0.6 mm, with the error of the average distance being 6% of the total observed variation. These findings indicate that errors were small and unlikely to bias the results.

The Development of the Curve of Spee

Paper 1

by

Antonio Gagliardi

B.H.Sc, B.D.S, B.Sc. Dent (Hons).



THE UNIVERSITY
of ADELAIDE

School of Dentistry
Faculty of Health Sciences
The University of Adelaide
South Australia
5005

2013

10. Paper 1: The Development of the curve of Spee

Gagliardi, A.

10.1 Aims

- To investigate the development of the curve of Spee during the primary dentition, mixed dentition and permanent dentition.
- To describe sex differences.
- To fit orthogonal polynomials to curves and interpret coefficients.

10.2 Abstract

The curve of Spee was first described by German embryologist, Ferdinand Graf Von Spee. Von Spee defined the curve in humans by constructing an arc which runs from the anterior surfaces of the mandibular condyles to the occlusal surfaces of the mandibular teeth. The forward and backward gliding movements of the mandible were believed to take place in a circular motion along this arc.

The aim of this study was to investigate the development of the curve of Spee during the primary, mixed and permanent dentition and describe any sex differences. Orthogonal polynomials were also fitted to curves to describe curve shape.

The sample comprised of pairs of Australian twins. The breakdown of the study sample for each time point and sex is as follows; T1 N= 128 (M= 53, F= 75), T2 N= 99 (M= 45, F= 54), T3 N= 98 (M= 44, F= 54). Subjects had dental models at 3 time points; the primary dentition, mixed dentition and early permanent dentition. The maximum depth of the curve of Spee was measured as the average of the perpendicular distances of depth from a measurement plane between the disto-buccal

cuspid tip of the most posterior mandibular tooth and the central incisors. This was then repeated to the canine cuspid tip. All cuspid tips between these reference cusps were digitized to allow a curve to be fitted. Descriptive statistics were generated and comparisons made. Time point 1 and 2 showed a relatively flat curve, while Time point 3 showed the greatest change in the depth of the curve. The results indicated that the greatest changes occurred between time point 2 and 3 with males recording a larger depth during this transition to the permanent dentition than females. The curve of Spee was also found to be greater in depth on the left side across all time points. The curve of Spee has clinical importance in dentistry, but there exists little conclusive information as to exactly define when, how and why it develops. The descriptions presented in this paper offer the opportunity to gain a better insight and understanding into the development of the curve of Spee by taking advantage of three time points demonstrating the transition of the dentition.

10.3 Introduction

The curve of Spee was first described by German embryologist, Ferdinand Graf Von Spee. His original 1890 publication entitled 'The gliding path of the mandible along the skull' has since been translated (Biedenbach *et al.* 1980) which assisted in understanding Von Spee's original description, as the curve has been described and interpreted differently.

Von Spee defined the curve in humans by constructing an arc which ran from the anterior surfaces of the mandibular condyles to the occlusal surfaces of the mandibular teeth, such that the arc lay at a tangent to the surface of a cylinder as viewed in the sagittal plane. The forward and backward gliding movements of the

mandible were believed to take place in a circular motion along this arc (Biedenbach *et al.* 1980)

This motion of gliding performed by the mandible can also occur with a slight deviation to either the left or right during function. The skull profile in Figure 10.1, shows the occlusal surfaces of the molars which are aligned in a downward convex curve in the maxilla and an upper concave curve in the mandible (Biedenbach *et al.* 1980).

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Fig 10.1. The Curve of Spee (Biedenbach et al. 1980).

Figure 10.1 also displays the effect of attrition, whereby the molars have lost their cusps thus allowing the upper and lower arches to fit together in a smooth curved line (Biedenbach *et al.* 1980). Upper and lower teeth with completely intact crown structure also appear to have the same curve-like relationship of their occlusal

surfaces, although the curvature can be visualised with greater ease in a worn dentition (Biedenbach *et al.* 1980). Von Spee described the following findings from his examination of the occlusal curvature (Biedenbach *et al.* 1980):

- The total visible contact line of the molar occlusal surfaces was on the same arc of a circle.
- Within the sagittal plane, the posterior continuation of the arc will touch the most anterior point of the condyle of the mandible.

Mathematical models have been used to describe changing direction of the bite force and its consequences on the mandibular joints. Baragar and Osborn (1987) used such a model to investigate the sagittal plane. They found that the molar long axis was orientated in a way that it would result in the most efficient use of muscle force. They described these as ‘work efficient’ angles which alluded to the most efficient part of the curve of Spee being its posterior portion and that raising the occlusal plane increased the efficiency of the muscles. They also believed that this orientation of the dentition was developed during the eruption of the dentition.

The growth and development of the curve of Spee has been related to a combination of factors which include the growth of the orofacial structures, eruption of the teeth and the development of the neuromuscular system (Marshall *et al.* 2008). It has also been proposed that the mandibular sagittal and vertical position relative to the cranium have an influence on the curve of Spee. In humans, brachyfacial patterns tend to display an exaggerated or increased curvature of Spee (Marshall *et al.* 2008).

It is hypothesised that the development of the curve of Spee remains relatively flat in the primary dentition but develops more curvature during the transition from the primary to the permanent dentition.

Previous work considering the development of the curve of Spee has been carried out by Marshall *et al* (2008). From their work it was concluded that the curve of Spee is relatively flat in the primary dentition and that a distinctive curve is observed with the eruption of the mandibular molars, they found no statistically significant differences between the right and left side and the sexes. The purpose of this first part of the study is to adapt some of the methodology presented by Marshall *et al.* (2008) by taking an average of all the perpendicular distances from the reference plane to each cusp tip rather than a single depth measure, this method is believed to provide a broader description of the depth as it provides an average depth of multiple points. As well as an average measure of depth, each cusp tip landmark was digitized and orthogonal polynomials were fitted to further investigate the development of the curve of Spee longitudinally across three time points.

10.4 Materials and Methods

Please refer to previous section for detailed description.

As discussed previously, linear measurements of the curve of Spee were determined. The methodology was adapted to that presented by Marshall *et al* (2008) where a single linear measurement was taken at the maximum depth directly on mounted study models with digital calipers. The methodology was adapted to allow study models to be mounted and the occlusal plane standardized. These were photographed

and landmarks digitized with Image J software to allow a 2-dimensional interpretation of the curve to be analysed. Rather than a single linear measurement, all cusp tips were digitized between the reference landmarks and the average of the perpendicular distances from the measurement plane was taken as the representation of the depth of the curvature. This was repeated for both the right and left sides and using the most posterior tooth distal cusp tip and incisal edge as landmark. This was then repeated with the canine cusp tip replacing the incisal edge. By digitizing each landmark, orthogonal polynomials could also be fitted to the curve to allow a description of the shape of the curvature (Figure 10.2). The equation that provided the best description of the curve of Spee included two orthogonal polynomials and their associated coefficients, b_1 and b_2 .

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Figure 10.2 Curve fitting (adapted from Marshall et al. 2008).

To compare the change in the depth of the curve of Spee over time, according to side and zygosity/sex (male MZ, female MZ, male DZ, female DZ, male DZOS (opposite sex) and female DZOS), a linear mixed effects model was fitted to the data. Two approaches were used, approach 1 modeled zygosity and sex together while approach 2 modeled zygosity and sex separately. In the model, side (right and left), time point

(T1, T2 and T3) and sex group (males and females) were investigated. Two-way interactions involving time were fitted as fixed effects (predictor variables). Random effects for twin pair, individual within twin pair, time point within individual and side within individual were fitted to the model. This was done in order to account for the multiple levels of dependency within the data. Where appropriate, non-significant interaction terms were removed from the models to allow for interpretation of main effects.

10.5 Results

Tables 10.1-10.4 show a Type 3 test of fixed effects. These fit all the variables first, and then consider individual variance for the variables of interest, therefore showing the significance of fixed effects in the linear mixed effects model. Looking at the interaction terms, it can be seen that the interaction between side and time was not statistically significant ($F(2, 194) = 0.87, p = 0.42$). Thus, there was no evidence that the difference in average non-zero distance between left and right sides varied over time. In order to interpret the main effect of side, a second linear mixed effects model excluding the non-significant interaction between side and time was fitted to the data. The non-significant interaction between zygosity/sex type and time ($p = 0.26$) was also removed from this second model so that the main effect of zygosity/sex type could be interpreted. Statistical significance was set at $P < 0.05$.

Table 10.1. Test of fixed effects to the canine landmark using Approach 1.

Type 3 Tests of Fixed Effects (Approach 1)				
Effect	Num DF	Den DF	F Value	P Value
Time	2	195	161.66	<.0001
Side	1	127	6.26	0.0137
Zyg/sex type	5	196	1.36	0.2425
Side*Time	2	194	0.87	0.4210
Zyg/sex type*Time	10	194	1.25	0.2643

Table 10.1 shows that independent of zygoty/sex type and time, there was a statistically significant difference in average depth of the curve of Spee between left and right sides ($p= 0.014$). Independently of zygoty/sex and side there was a statistically significant change in average depth of the curve of Spee over time ($p< 0.0001$). Independently of side and time, there was no evidence for a difference in average depth of the curve of Spee between zygoty/sex ($p= 0.24$).

Table 10.2. Test of fixed effects to the canine landmark using Approach 2.

Type 3 Tests of Fixed Effects (Approach 2)				
Effect	Num DF	Den DF	F Value	P Value
Time	2	193	168.37	<.0001
Side	1	127	6.24	0.0138
Zyg type	2	196	0.06	0.9451
Sex	1	196	8.14	0.0048
Sex*Time	2	196	3.46	0.0335

Table 10.2 shows the final model. From this independently of sex, side and time there were no differences in average depth of the curve of Spee between the MZ, DZ and DZ (opposite sex) zygoty types ($p = 0.95$). Independently of sex, zygoty and time there was a statistically significant difference in average depth of the curve of Spee

between the right and left sides ($p= 0.014$). There was also a significant interaction between sex and time ($p= 0.034$). This suggested that the difference in average depth of the curve of Spee males and females also differed over time.

The same model was then repeated using the incisor as the landmark.

Table 10.3. Test of fixed effects to the incisor landmark using Approach 1.

Type 3 Tests of Fixed Effects (Approach 1)				
Effect	Num DF	Den DF	F Value	P Value
Time	2	195	320.09	<.0001
Side	1	125	5.07	0.0261
Zyg/sex type	5	175	0.42	0.8325
Side*Time	2	175	3.72	0.0263

Table 10.3 shows that independently of side and time, there was no evidence for a difference in average depth of the curve of Spee between zygoty/sex ($p= 0.83$). Independently of zygoty/sex, there was a statistically significant interaction between side and time ($p= 0.026$), suggesting that average depth of the curve of Spee between left and right sides depended on time.

Table 10.4. Test of fixed effects to the incisor landmark using Approach 2.

Type 3 Tests of Fixed Effects (Approach 2)				
Effect	Num DF	Den DF	F Value	P Value
Time	2	195	320.44	<.0001
Side	1	125	5.15	0.0250
Zyg type	2	177	0.00	0.9959
Sex	1	177	1.57	0.2117
Side*Time	2	177	3.63	0.0285

Table 10.4 shows the final model. This shows that the average depth of the curve of Spee was found to depend on the combination of side and time (interaction $p= 0.03$), but was not related to sex ($p= 0.21$) or zygoty ($p> 0.99$).

Post hoc tests

To further explore the changes over the three time points, additional post-hoc tests were carried out and these can be seen in Table 10.5. The post-hoc comparisons show that the difference in average depth of the curve of Spee was statistically significant across all three time points (increasing from 0.41mm at T1 to 1.25mm at T3).

Table 10.5. Post-hoc test across T1, T2 and T3.

Differences of Adjusted Means (mm)							
Effect	Time	Time	Estimate	Standard Error	DF	t Value	P Value
Time	1	2	-0.4346	0.04627	195	-9.39	<.0001
Time	1	3	-0.8334	0.04646	195	-17.94	<.0001
Time	2	3	-0.3988	0.04811	195	-8.29	<.0001

The following Tables 10.6-10.9 show the descriptive statistics for the average curve of Spee depth (mm) across all three time points. Tables 10.6 and 10.7 give the descriptive statistics for the average curve of Spee depth (mm) for T1 through to T3 using the canine cusp tip landmark. T1, representing the primary dentition, shows a minimal curve of Spee with a statistically significant ($p<0.0001$) change through to T3. This change through the transitional dentition to the permanent, corresponds to the eruption of the permanent mandibular molars and canines. The curve of Spee increased from a right and left side average at T1 in males of 0.425 mm to 1.222 mm

at T3 and, for T1 females, from 0.392 mm to 1.129 mm. This represented a statistically significant change in the curve of Spee depth ($p < 0.0001$). There was also a statistically significant difference between the right and left sides ($p = 0.014$). It can be seen that the curve of Spee was deeper on the left hand side at all time points for both males and females. Plots of the data distribution can be seen in Figure 10.3.

Table 10.6. Average curve of Spee depth for right side canine landmark.

	Average depth (mm)		
	T1	T2	T3
All Subjects			
N	128	99	98
Mean	0.374	0.846	1.207
SD	0.267	0.567	0.422
Minimum	-0.27	-1.24	0.15
Maximum	1.27	1.92	2.83
Male Subjects			
N	53	45	44
Mean	0.385	0.936	1.349
SD	0.244	0.476	0.418
Minimum	-0.11	-1.24	0.58
Maximum	1.01	1.83	2.83
Female Subjects			
N	75	54	54
Mean	0.366	0.771	1.094
SD	0.283	0.628	0.394
Minimum	-0.27	-1.19	0.15
Maximum	1.27	1.92	1.85

Table 10.7. Average curve of Spee depth for left side canine landmark.

	Average depth (mm)		
	T1	T2	T3
All Subjects			
N	128	99	98
Mean	0.437	0.860	1.289
SD	0.243	0.611	0.415
Minimum	-0.19	-1.47	0.01
Maximum	1.12	2.39	2.15
Male Subjects			
N	53	45	44
Mean	0.464	0.928	1.442
SD	0.242	0.602	0.376
Minimum	0.02	-1.47	0.72
Maximum	1.12	2.39	2.15
Female Subjects			
N	75	54	54
Mean	0.418	0.804	1.164
SD	0.244	0.619	0.407
Minimum	-0.19	-1.38	0.01
Maximum	0.96	1.86	1.88

Tables 10.8 and 10.9 give the descriptive statistics for the curve of Spee depth for T1 through to T3 using the incisor cusp tip landmark. T1, representing the primary dentition, showed a minimal curve of Spee with a statistically significant ($p < 0.0001$) change through to T3. This change through the transitional dentition to the permanent, corresponds to the eruption of the permanent mandibular molars and central incisors. The curve of Spee increased from a right and left side average at T1 in males of 0.677 mm to 1.465 mm at T3. In females, the change ranged from -0.205 mm at T1 to 1.216 mm at T3. This change was statistically significant ($p < 0.0001$). There was also a statistically significant difference between the right and left sides.

($p= 0.014$). It can be seen that the curve of Spee was deeper on the left hand side at all time points for both males and females. These results reflect the same findings as the canine landmark. Plots of the data distribution can be seen in Figure 10.4.

Table 10.8. Average curve of Spee depth for right side incisor landmark.

	Average depth (mm)		
	T1	T2	T3
All Subjects			
N	128	99	98
Mean	-0.085	0.898	1.254
SD	0.552	0.454	0.596
Minimum	-1.98	-0.39	-0.53
Maximum	1.60	1.18	2.91
Male Subjects			
N	53	45	44
Mean	0.051	0.969	1.402
SD	0.474	0.418	0.606
Minimum	-1.35	0.24	-0.05
Maximum	1.60	1.81	2.91
Female Subjects			
N	75	54	54
Mean	-0.180	0.838	1.133
SD	0.585	0.478	0.566
Minimum	-1.98	-0.39	-0.53
Maximum	1.38	1.76	2.33

Table 10.9. Average curve of Spee depth for left side incisor landmark.

	Average depth (mm)		
	T1	T2	T3
All Subjects			
N	128	99	98
Mean	-0.070	0.950	1.401
SD	0.773	0.450	0.570
Minimum	-2.99	-0.28	-0.51
Maximum	5.23	2.14	2.71
Male Subjects			
N	51	45	44
Mean	0.173	1.034	1.527
SD	0.858	0.337	0.490
Minimum	-1.51	0.34	0.36
Maximum	5.23	1.64	2.71
Female Subjects			
N	77	54	54
Mean	-0.231	0.881	1.299
SD	0.669	0.519	0.613
Minimum	-2.99	-0.28	-0.51
Maximum	1.01	2.14	2.56

Figure 10.3. Maximum mean curve depth (Canine Landmark).

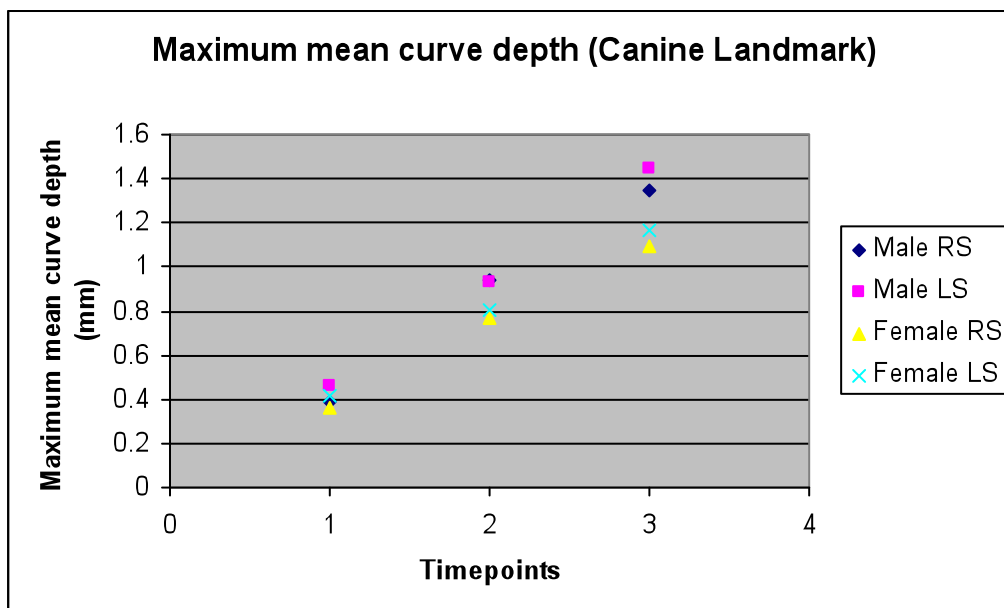
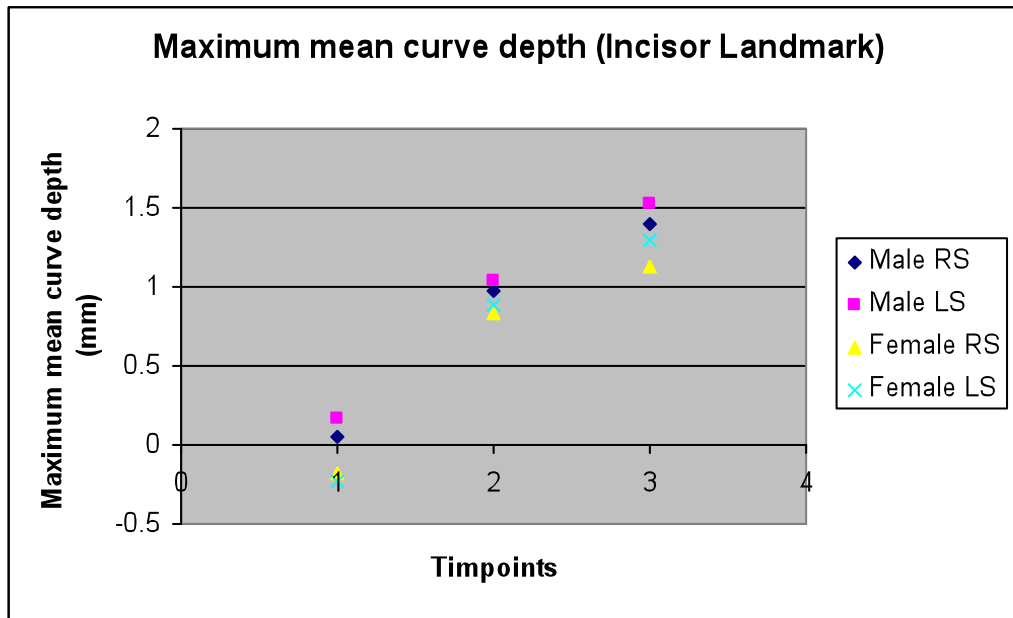


Figure 10.4. Maximum mean curve depth (Incisor Landmark).



Curve fitting b1 and b2 coefficients

As was previously carried out for the linear measurement of curve of Spee depth, the following description presents Type 3 tests of fixed effects. These fitted all the variables first, and then considered individual variance for the variables of interest, therefore showing the significance of fixed effects in the linear mixed effects model. Two approaches were used; Approach 1 modeled zygoty and sex together while Approach 2 modeled zygoty and sex separately.

The b1 coefficient enabled the presence of any asymmetry within the curvature to be quantified i.e. whether there was a greater dip in the anterior region of the arch

compared with the posterior region. The b2 coefficient represented the shape of the curve, i.e. whether the curvature was deep or shallow.

b 1 Coefficient

The b1 coefficient depended on both side and time. While no differences were found between right and left sides at T1 ($p= 0.29$). The differences were statistically significant at T2 and T3. The b1 value increased with time on the right side and decreased on the left. The final model showed that the differences between right and left sides were statistically significant and depended on time ($p< 0.0001$). Post hoc tests confirmed that b1 values increased on the right side over time and decreased on left side.

b 2 Coefficient

Independently of side and zygoty/sex and, similarly, time and zygoty/sex, the b2 coefficient was found to change over time. There was a significant difference in b2 values between the left and right sides. The b2 values were less negative on the right side. It was observed that b2 values decreased over time indicating a greater depth of curvature with time. The final model showed that b2 values were associated independently with time ($p< 0.0001$), side ($p= 0.04$) and sex ($p= 0.04$). Values were found to be more negative in males than females (-0.050 vs. -0.045 respectively, $p= 0.04$) as were on the left side compared to the right side (-0.050 vs. -0.047 respectively, $p= 0.04$). This would indicate that males have a more pronounced curve of Spee than females with greater depth being observed on the left hand side. Post hoc tests confirmed that b2 values decreased over time in both males and females.

Tables 10.10-10.13 give the descriptive statistics for b1 and b2 coefficients, describing changes in any asymmetry within the curve as well as the depth of the curvature. The depth of the curvature can be seen to increase across all three time points as the b2 value increases. As this value becomes more negative the curvature is further accentuated. This accentuation of curve shape can be seen across all time points in both males and females. This change appears to be more pronounced on the left hand side, indicating a greater curvature on the left. This observation appears in both males and females, with males recording larger changes. The b1 coefficient, which is describing any asymmetry within this curvature, also appears to follow a similar pattern. A value of zero would indicate a symmetrical curvature, and as the value increases over time the curvature shows an increasing degree of asymmetry in the posterior molar region of the curve. This was observed to be greater on the left side and greater in males. The description of asymmetry refers to any observed change in the curvature when viewed from the lateral aspect taking into account the left and right sides individually, i.e. the extent to which the curvature dips in the anterior versus the posterior region. The change refers to the differences between the anterior (incisor and canines) and posterior region (first and second molars) of the left and right sides separately.

Table 10.10. Curve fitting coefficients for right side canine landmark.

Time point	T1		T2		T3	
Coefficients	b1	b2	b1	b2	b1	b2
All Subjects						
N	128	128	99	99	98	98
Mean	0.002	-0.029	0.085	-0.048	0.098	-0.064
SD	0.006	0.021	0.042	0.029	0.045	0.025
Minimum	-0.01	-0.11	-0.00	-0.12	-0.04	-0.16
Maximum	0.02	0.03	0.23	0.05	0.21	0.01
Male Subjects						
N	53	53	45	45	44	44
Mean	0.001	-0.029	0.089	-0.053	0.099	-0.071
SD	0.006	0.019	0.032	0.025	0.045	0.026
Minimum	-0.01	-0.08	0.04	-0.11	-0.04	-0.16
Maximum	0.02	0.01	0.17	0.05	0.17	-0.02
Female Subjects						
N	75	75	54	54	54	54
Mean	0.003	-0.028	0.082	-0.045	0.097	-0.059
SD	0.007	0.022	0.048	0.032	0.046	0.023
Minimum	-0.01	-0.11	-0.00	-0.12	-0.02	-0.10
Maximum	0.02	0.03	0.23	0.05	0.21	0.01

Table 10.11. Curve fitting coefficients for left side canine landmark.

Time point	T1		T2		T3	
Coefficients	b1	b2	b1	b2	b1	b2
All Subjects						
N	128	128	99	99	98	98
Mean	-0.002	-0.033	-0.076	-0.048	-0.090	-0.067
SD	0.006	0.019	0.046	0.031	0.042	0.024
Minimum	-0.03	-0.09	-0.26	-0.15	-0.19	-0.11
Maximum	0.01	0.03	0.11	0.06	0.01	0.01
Male Subjects						
N	53	53	45	45	44	44
Mean	-0.001	-0.036	-0.083	-0.052	-0.091	-0.075
SD	0.004	0.019	0.028	0.031	0.038	0.023
Minimum	-0.01	-0.09	-0.19	-0.15	-0.17	-0.11
Maximum	0.01	-0.00	-0.04	0.06	-0.00	-0.02
Female Subjects						
N	75	75	54	54	54	54
Mean	-0.003	-0.031	-0.070	-0.045	-0.090	-0.061
SD	0.007	0.019	0.057	0.030	0.046	0.024
Minimum	-0.03	-0.07	-0.26	-0.11	-0.19	-0.10
Maximum	0.01	0.03	0.11	0.05	0.01	0.01

Table 10.12. Curve fitting coefficients for right side incisor landmark.

Time point	T1		T2		T3	
Coefficients	b1	b2	b1	b2	b1	b2
All Subjects						
N	128	128	99	99	98	98
Mean	-0.007	0.005	0.045	-0.046	0.073	-0.045
SD	0.038	0.026	0.022	0.023	0.031	0.022
Minimum	-0.08	-0.06	-0.01	-0.10	-0.02	-0.12
Maximum	0.11	0.09	0.10	0.02	0.16	0.02
Male Subjects						
N	53	53	45	45	44	44
Mean	-0.014	-0.004	0.045	-0.050	0.071	-0.051
SD	0.037	0.023	0.017	0.020	0.029	0.023
Minimum	-0.08	-0.06	0.00	-0.10	-0.02	-0.12
Maximum	0.11	0.05	0.08	-0.02	0.12	0.01
Female Subjects						
N	75	75	54	54	54	54
Mean	-0.003	0.012	0.046	-0.042	0.075	-0.040
SD	0.039	0.027	0.025	0.025	0.033	0.021
Minimum	-0.08	-0.06	-0.01	-0.09	0.01	-0.09
Maximum	0.11	0.09	0.10	0.02	0.16	0.02

Table 10.13. Curve fitting coefficients left side incisor landmark.

Time point	T1		T2		T3	
Coefficients	b1	b2	b1	b2	b1	b2
All Subjects						
N	128	128	99	99	98	98
Mean	0.021	0.004	-0.034	-0.047	-0.061	-0.050
SD	0.043	0.029	0.024	0.023	0.029	0.022
Minimum	-0.13	-0.06	-0.09	-0.11	-0.12	-0.10
Maximum	0.12	0.12	0.05	0.01	0.01	0.03
Male Subjects						
N	53	53	45	45	44	44
Mean	0.019	-0.007	-0.035	-0.052	-0.062	-0.054
SD	0.041	0.023	0.023	0.017	0.027	0.019
Minimum	-0.08	-0.05	-0.09	-0.09	-0.11	-0.09
Maximum	0.10	0.05	0.01	-0.01	0.00	-0.01
Female Subjects						
N	75	75	54	54	54	54
Mean	0.022	0.011	-0.034	-0.043	-0.061	-0.047
SD	0.044	0.030	0.025	0.027	0.031	0.023
Minimum	-0.13	-0.06	-0.09	-0.14	-0.12	-0.10
Maximum	0.12	0.12	0.05	0.01	0.01	0.03

10.6 Discussion

The principal Paper 1 findings of the study indicate that the depth of the curve of Spee on the left side appeared to be greater in both males and females. The curvature also increased across all the three time points, indicating a greater accentuation to the curvature with time, as well as an increase in asymmetry. The depth of the curve of Spee appeared to be minimal at the first time point, representing the primary dentition, and supports descriptions of the primary dentition curve of Spee by Ash (1993). Time point 3, represented by a permanent dentition, showed the greatest increase in the depth of the curve of Spee. The average change from T1 to T3 was 0.8

mm in males and 0.7 mm in females and this may be explained by the eruption of the second permanent molars and canines. Their possible contribution to the development of the curve of Spee supports the findings presented by Marshall *et al.* (2008). This may also explain the observed changes in the degree of curvature and increase in asymmetry and the contribution of molar eruption to the shape of the curve. The primary dentition, therefore, may be described as having a flat or mild curve of Spee which progressively develops with the eruption of the permanent dentition. This has been previously described and reported by others (Bishara *et al.* (1989); Carter and McNamara (1998)).

The depth of the curve of Spee showed no statistically significant differences between males and females at both time points 1 and 2, although time point 3 showed a significantly greater average value in males. This is in contrast to other studies (Shannon and Nanda 2004, Marshall *et al.* 2008) which have found no significant sex differences between males and females. The curve of Spee has been described by authors as being relatively stable following the increase in T3 which coincides with the eruption of the second molars. This stability into early adulthood has been reported by Marshall *et al.* (2008).

Difficulties and limitations in modelling the curve of Spee can be attributed to its form. As it is a 3D dynamic structure, there are difficulties in developing an accurate definition of the curve of Spee. Most authors have attempted to model the curve as a simple 2D curvature defined by the length of an arc of a circle. By using landmarks on this arc (cusp tips) it is possible to establish reference planes and the maximum depth of the arc can be determined. It may be better to consider the curve of Spee as a

3D ribbon encompassing the entire occlusal surfaces, with multiple planes of different surface area existing within its entire structure.

In this study the curve of Spee was measured from a 2D perspective. Thus, the changes in curve depth were measured during a change in arc length over 3 time points where the greatest changes may be attributed to the eruption of the first and second molars.

Molar eruption may result in an increase in the length of the curvature as the landmark will be moved further posteriorly, resulting in a movement of the reference plane, for example, from first molar to second molar, effectively altering the height of the reference plane and increasing the maximum depth of the curvature from the plane. The changes observed across T1, T2 and T3, therefore, may be attributed to both an increase in the length of the curve and a change in shape of the curve. The curve of Spee, therefore, may be considered largely the result of dental development rather than being of skeletal origin as the eruption of the molars and central incisors appears to have an important influence over the development of the curvature. Mandibular dental development also precedes the development of the maxillary dentition, thus a difference in these dental events may allow the unopposed over-eruption of the central incisors and mandibular molars past the mandibular occlusal plane to create a curvature. This event, occurring at both T2 and T3 with the eruption of the first and second molars respectively, allows up to 6 month for supra-eruption to occur past the occlusal plane.

Osborn (1987, 1993) has described the development of the curve of Spee as a result of masseter muscle orientation and forward tilting of the posterior mandibular teeth. Thus there may also be a combination of contributions from dental eruption, craniofacial development and biomechanics (Baragar and Osborn 1987). Osborn (1987, 1993) and Baragar and Osborn (1987) also reported on the presence of the curve of Spee in primates. Regardless of the amount of tooth wear, the curve of Spee tends to be maintained. Farella *et al.* (2002) have considered the curve of Spee and skeletal facial morphology and found the curve to be influenced by the horizontal position of the condyle with respect to the dentition, sagittal position of the mandible with respect to the anterior cranial base and the posterior and anterior facial height ratios.

As a possible explanation of the observed accentuated curvature on the left side, preferential mastication side may be considered, and possible influences of dental eruption, craniofacial development and muscle biomechanics. Martinez-Gomis *et al.* (2009) reported on chewing side preference, suggesting that lateral asymmetry of bite force and asymmetry of occlusal contacts at the intercuspital position may contribute to preferential chewing side. Baragar and Osborn (1987) used mathematical models to investigate the sagittal plane. They found that the molar long axis was orientated in a way that would result in the most efficient use of muscle force. The orientation of the masseter muscle appears to be suited to the direction of crushing forces between molars in the posterior region which tend to be mesially inclined with forward facing occlusal surfaces. They described these as 'work efficient' angles, which alludes to the most efficient part of the curve of Spee as its posterior portion. They proposed that raising the occlusal plane would increase the efficiency of the muscles. Baragar and

Osborn (1989) suggested that an increase in the crush-shear ratio of the posterior dentition would, therefore, maximise the efficiency of the occlusal forces during mastication.

This, in turn, may result in greater masseter muscle activity on the preferred chewing side to increase the work efficient angles and mesially inclined molars to increase the crush-shear ratio which would, therefore, reflect a deeper curvature on the left side.

These findings support previous authors who related development of the curve of Spee, as assessed from a 2D perspective, to the eruption of the mandibular molars; particularly, the mandibular second molar. These results give the clinician a greater understanding of the natural development of the curvature and the significant influence that the inclusion of lower second molars in orthodontic treatment has on leveling and eliminating the occlusal curvature to achieve bite opening and orthodontic treatment alignment goals. The stability of leveling has been investigated by various authors. De Praeter *et al.* (2002) investigated the stability of leveling the curve of Spee, before, during and post-treatment and found that in some cases the curve became even deeper after treatment. This relapse may be due to effects of the biological system and re-establishing of the natural curve.

Quantifying a 3D structure via 2D measures poses limitations, particularly due to the dynamic nature of the curve and the ability to locate stable reference points. A 3D computer scanning method of analysis may prove to be a more accurate descriptor of the changes affecting the curve.

The findings provide an insight into the development of the curve and possible influences. Orthodontists treat the curve of Spee on a daily basis and an understanding of the curve of Spee changes across the transitional phases of the dentition has showed some guidance in leveling mechanics, particularly in cases of exaggerated curvatures. Bonding of the second molars would aid in leveling the curvature and bite opening mechanics during orthodontic treatment, conversely some degree of curvature retained at the end of treatment may also be associated with a more biologically stable occlusion.

10.7 Conclusion

The curve of Spee was relatively flat in the primary T1 stage and showed an increase in curvature through all three time points T1-T3. The greatest increase was observed at the T3 stage and this increase may be attributed to the eruption of the second molars and central incisors beyond the occlusal plane resulting in further lengthening and deepening of the curve of Spee. This observation was supported as the curvature was accentuated with time and also showed evidence of further asymmetry contributed to by second molar eruption.

Significant differences were observed between right and left sides, with the curve being deeper on the left side than the right. This deepening on the left side may be accounted for by bite forces due to preferential chewing and it may be considered as developing due to an interaction of dental eruption, craniofacial development and muscle biomechanics contributing to the crush-shear ratio for maximum chewing efficiency.

The aims of the present study were achieved and the development of the curve of Spee was observed over three time points with side and sex differences being reported. Orthogonal polynomials were used to describe changes in curvature as well as linear measurements, with the changes in curvature and asymmetry within the curve being explained by possible changes in dental development. T2 to T3 was associated with the greatest changes in the curve of Spee, which coincided with the eruption of the second molars.

The genetic basis of the curve of Spee in Australian twins.

Paper 2

by

Antonio Gagliardi

B.H.Sc, B.D.S, B.Sc. Dent (Hons).



THE UNIVERSITY
of **ADELAIDE**

School of Dentistry
Faculty of Health Sciences
The University of Adelaide
South Australia
5005

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11. Paper 2: The genetic basis of the curve of Spee in Australian twins.

Gagliardi, A.

11.1 Aims

- To use a classical twin model to describe the differences between monozygotic (MZ) and dizygotic (DZ) twin pairs, to clarify the role of genetic and environmental contributions to the development of the curve of Spee.
- To use an MZ co-twin model to illustrate differences in the curve of Spee within twin pairs.

11.2 Abstract

The curve of Spee was first defined in humans by Ferdinand Graf Von Spee, who constructed an arc which ran from the anterior surfaces of the mandibular condyles to the occlusal surfaces of the mandibular teeth. Other authors have attempted to better understand the development of the curve of Spee over time as well as to identify factors that contribute to its shape. In this follow up to Paper 1, the underlying genetic contribution will be quantified. Changes in the curve of Spee over time, both as a linear measure of depth and also its shape were reported in Paper 1. From this, it was concluded that the curvature was more accentuated in shape in the permanent dentition and that differences in curvature were indicated between right and left sides. This may be related to dental development; in particular, the eruption of the mandibular second molars and differential eruption of the incisors as well muscular, biomechanical and craniofacial development parameters. To further investigate

genetic influences on the development of the curve of Spee, the same sample of untreated subjects from an ongoing twin study at the University of Adelaide was assessed. The study sample consisted of study casts monozygotic (MZ) and dizygotic (DZ) twin pairs across three time points ranging from the primary dentition through to the permanent dentition. The breakdown of the study sample for each time point and sex is as follows; T1 N= 128 (M= 53, F= 75), T2 N= 99 (M= 45, F= 54), T3 N= 98 (M= 44, F= 54). In order to clarify the genetic influences between MZ and DZ twins, a classical twin model was used as well as an MZ co-twin model. Broad sense heritability estimates were derived to identify the total contribution of genetic factors (additive and non-additive) to the observed variation. The results indicated that a moderate to high heritability is likely and that there are environmental factors acting on both the average depth of the curve and the $b2$ coefficient describing its extent of curvature. There appeared to be evidence of genetic dominance of the $b1$ coefficient describing the extent of asymmetry within the curve.

11.3 Introduction

Features of human dental variation can be further investigated from a genetic basis by utilizing data derived from twins and their families. Various models have been proposed with their own advantages and limitations. The most common approach is to use a classical twin model. This model allows for the use of both monozygotic (MZ) as well as dizygotic (DZ) twin samples. From a genetic perspective, MZ co-twins share the same genes while the DZ co-twins only share 50% of their genes on average. If both the MZ and DZ pairs have been sampled from the same gene pool and have been under the influence of the same environmental factors, the extent that

the observed variation is influenced by genetic and environmental factors can be ascertained.

Heritability (h^2) indicates the relative importance of nature (genetics) compared with nurture (environment) on a given trait's variability (Harris, 2008). Estimating h^2 depends on some form of analysis of variance (ANOVA) or correlation analysis that partitions the observed variability (total phenotypic variance) into one part due to genetic similarities and the other part where the remaining variation is collectively labeled as environmental.

The MZ co-twin model allows the comparison of MZ twins where one member of the pair has been exposed to different environmental effects than the other. A particular feature, such as tooth size, can be considered and inferences can be made about the relative contributions of genetic and environmental factors to the phenotypes. If MZ twins exhibit differences for a particular feature, it is likely that environmental and/or epigenetic factors are exerting an influence leading to the differences.

The genetic variance can then be broken down to additive and non-additive genetic effects. Most of the twin studies consider narrow sense heritability, this being the proportion of variation due to additive genetic variance (Van Dongen *et al.* 2012). Additive genetic variation causes DZ correlations to be about half the MZ correlations. Dominance would tend to decrease the DZ correlation to less than half the MZ value, and common environment increases it over half the MZ value (Dempsey *et al.* 1995).

In this second paper, the focus is on identifying the presence of any underlying genetic influence on the curve of Spee. The purpose of the heritability estimates is not to establish an exact percentage of genetic contribution but rather to ascertain whether there is some evidence of an underlying genetic contribution due the depth and shape of the curve of Spee. The descriptive statistical analyses for these parameters have been previously reported in Paper 1. It is hypothesised that the curve of Spee shape has a genetic contribution with some environmental influence on its final depth and shape.

11.4 Material and Methods

Please refer to previous section 6 for detailed description.

Broad sense heritability estimates were derived to identify the total contribution of genetic factors (additive and non-additive) to the observed variation.

h^2 was calculated according to the following formula (Falconer and MacKay 1996):

$$h^2 = 2(rMZ - rDZ)$$

In order to quantify the extent of the genetic contribution to the observed phenotypic variation, heritability estimates were calculated. These values can range from 0 to 1 (0-100%). A heritability estimate of 1 would indicate that all the phenotypic variation can be explained by genetic factors while an estimate of zero would indicate that there are no genetic factors acting on the phenotypic variation. It must be emphasized that the h^2 values in this study are provided to indicate a trend and clarify whether there is

a genetic contribution to the curve of Spee. The values presented are only estimates of the true genetic contribution.

Further to the heritability estimates, a selection of MZ pairs is presented to further illustrate similarities in curve of Spee form.

11.5 Results

Table 11.1. Heritability estimates (0-1 = 0-100%) for variables describing the Curve of Spee at three different ages.

Time points	Landmark	Side	Heritability Depth	Heritability (b1)	Heritability (b2)
T1	Canine	Right	0.44	1	0.22
		Left	0.22	0.92	0.2
	Incisor	Right	0.68	0.54	0.82
		Left	0.42	0	0.54
T2	Canine	Right	1	0.76	1
		Left	0.24	0.68	0.6
	Incisor	Right	0.86	0.74	1
		Left	0.28	0.32	0.6
T3	Canine	Right	0.36	0.98	0.7
		Left	1	1	1
	Incisor	Right	0.34	1	0.58
		Left	0.74	1	1

b1 is the first shape coefficient (describes the degree of asymmetry of the curve).

b2 is the second shape coefficient (describes how rounded the curve is).

The above table reports the broad sense (additive + dominance) heritability estimates for each of the parameters; average depth, b1 and b2 on a per age/variable/side basis.

The heritability estimates have no estimate of confidence (e.g. standard error). The

limitations presented by the broad sense estimates will be addressed in future with

more sophisticated analysis. The purpose of these estimates is to establish whether there are any consistent trends.

It can be seen from the data that the heritability estimates for depth at T1 were centered about 0.5 (50% of phenotypic variation explainable by genetic influence), slightly increasing across T2 and T3, therefore indicating a moderate to high influence of genetic factors on the depth of the curve of Spee. Heritability estimates for the first shape coefficient b_1 can be seen to follow a similar trend in T1 and show a moderate to high influence of genetic factors in T2 with further increase in T3. T2 to T3 displays the greatest change, which corresponds to development of the permanent dentition. It is at this stage that the second mandibular molars are erupted and greater asymmetry is present between the anterior and posterior portions of the curve of Spee from a 2D perspective.

Heritability estimates for the second shape coefficient, b_2 , can be seen to reflect a similar trend to b_1 . Heritability estimates across T2 and T3 tend to be centered around 0.8 (80% of phenotypic variation explained by genetic influence). This would indicate that there is a relatively high genetic influence on the extent of curvature in the curve of Spee.

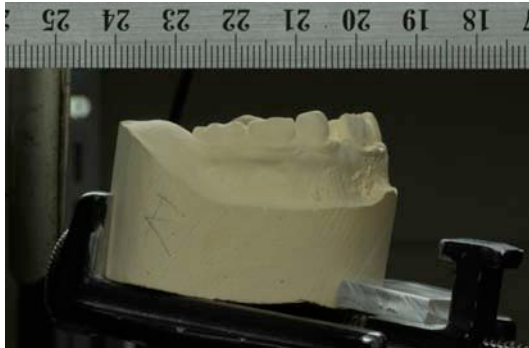
The results indicate that a moderate to high heritability is likely and that there may be common environmental factors acting on both the average depth of the curve and the b_1 and b_2 coefficient describing the extent of asymmetry and curvature within the curve.

11.6 MZ twin case examples

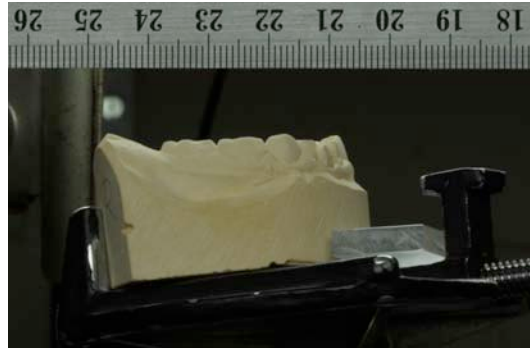
The following case examples are included to illustrate dissimilarities between twin MZ pairs. Only one side is shown to illustrate the changes in the curve of Spee. Photographs represent the right side.

Case 1

Twin A
T1



Twin B



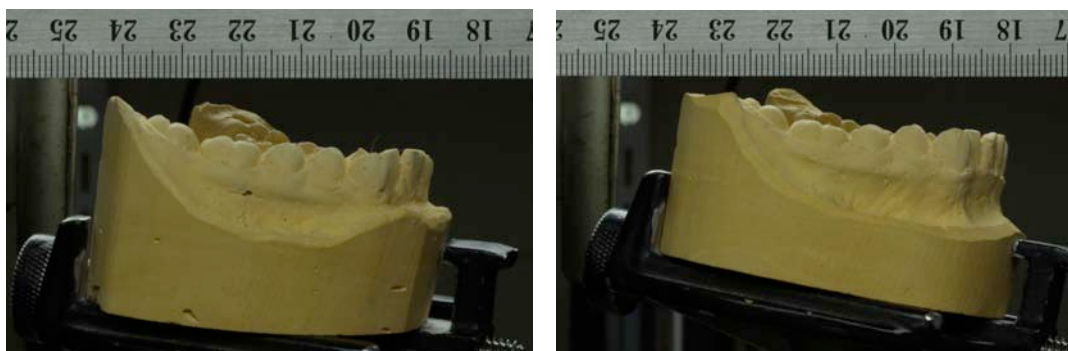
Figures 11.1 and 11.2. It can be seen that at T1, the curve of Spee is relatively flat, both twin A and B recorded the same depth of 0.2mm.

T2



Figures 11.3 and 11.4. It can be seen that at T2, the curve of Spee remains relatively flat, both twin A and B recorded the similar depths with a difference of 0.1mm. Twin B also appears to show differences in eruption timing and exfoliation.

T3



Figures 11.5 and 11.6. It can be seen that at T3, the curve of Spee curvature is becoming more distinct. Both twin A and B recorded the greatest increase in depth. Of the pair, twin B displayed the greatest increase in depth, compared to its co twin, with twin B appearing to have a more distinct curvature.

Case 1 illustrates the change across T1 to T3. The values for depth, b1 and b2 are presented in the following Table 11.2.

Table 11.2. Twin A and B values for depth, b1 and b2.

Time point	Depth (mm)	b 1	b 2
T1 Twin A	0.20	-0.00018	-0.01296
T1 Twin B	0.20	-0.00099	-0.00653
T2 Twin A	0.60	0.083391	-0.00278
T2 Twin B	0.70	0.106397	0.003554
T3 Twin A	0.85	0.14922	-0.04544
T3 Twin B	1.27	0.128577	-0.07536

Table 11.2 illustrates dissimilarities between twin pairs for all values; changes in curve of Spee depth and shape follow a similar trend in both twin A and B. It can be seen that the depth of the curve of Spee is slightly increased in twin B. On closer

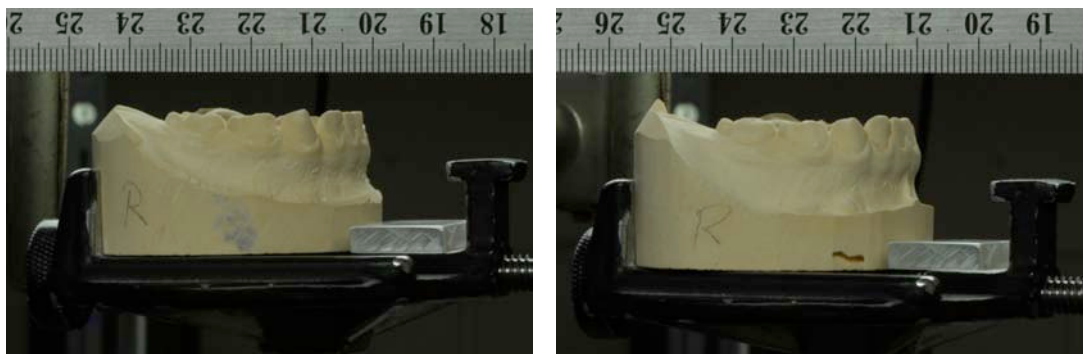
inspection during T2 there is some evidence of eruption timing differences within the pair and by T3 a more distinct curvature is discernable in twin B. The case highlights the subtle differences which can exist in a MZ twin pair.

Case 2

Twin A

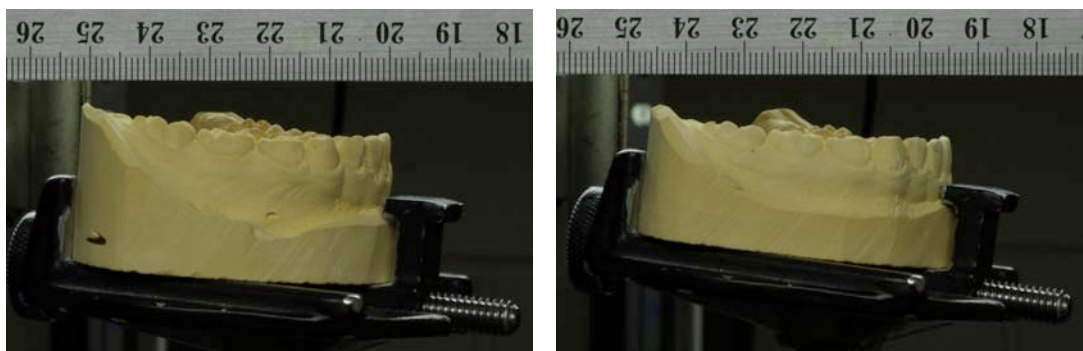
Twin B

T1



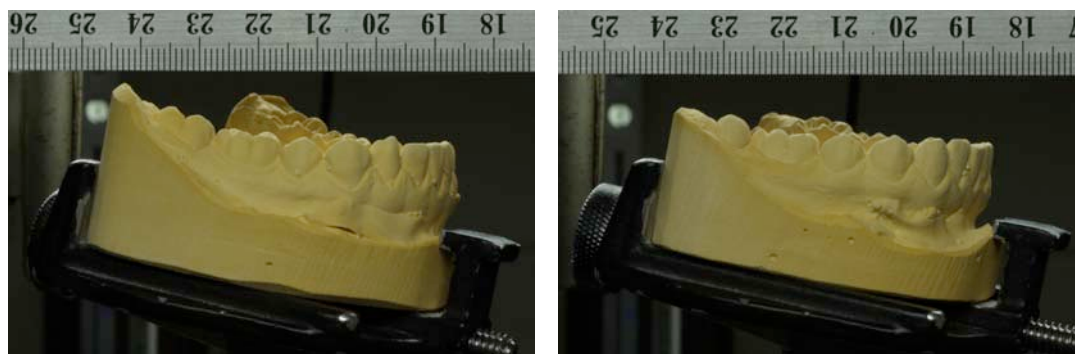
Figures 11.7 and 11.8. It can be seen that at T1, the curve of Spee is relatively flat with both twin A and B recording a similar depth.

T2



Figures 11.9 and 11.10. It can be seen that at T2, the curve of Spee remains relatively flat, with both twin A and B recording a slight increase in depth.

T3



Figures 11.11 and 11.12. It can be seen that by T3, the curve of Spee curvature is becoming more distinct. Both twin A and B recorded the greatest increase in depth at this time. Of the pair, twin A displayed the greatest increase in depth, compared to his co-twin, with twin A appearing to have a more distinct curvature. The eruption and position of the second molars as well as the crown morphology of the canine differ, these differences being reflected in the shape of the curve of Spee.

Case 2 illustrates the change across T1 to T3. The value for depth, b1 and b2 are presented in the following Table 11.3.

Table 11.3. Twin A and B values for depth, b1 and b2.

Time point	Depth (mm)	b 1	b 2
T1 Twin A	0.57	-0.00532	-0.04007
T1 Twin B	0.56	-0.00715	-0.03922
T2 Twin A	0.99	0.141207	-0.05192
T2 Twin B	0.67	0.105363	-0.03579
T3 Twin A	1.53	0.129538	-0.08462
T3 Twin B	0.67	0.112167	-0.04154

Table 11.3 illustrates the dissimilarities between twin pairs for all values. Changes in curve of Spee depth and shape follow a similar trend in both twin A and B. It can be seen that the depth of the curve of Spee differs in T3 representing minimal change in twin B. On closer examination it can be seen that the lower right second molar position differs between twin A and B in T3 as well as canine cusp tip wear pattern. These factors contribute to the observed differences in the curve of Spee. The case further highlights the subtle differences which can exist in a MZ twin pair, highlighting the contribution of genetics and environment.

While these pairs are regarded as sharing 100% of their genes, subtle phenotypic variation can still be observed by comparing the study models. Variations in dental wear patterns, tooth angulation and eruption rates can all be regarded as external environmental influences and these may influence the curve of Spee, but there still seems to be a relatively strong genetic influence still present contributing to variation.

11.7 Discussion

The application of twin studies to improve understanding of dental development, tooth size, tooth morphology, arch shape and dimensional changes has clarified the roles of genetic, epigenetic and environmental influences (Townsend *et al.* 2012). The curve of Spee is another such biological parameter that can be investigated. Many of the studies that have been carried out to date have not investigated the curve of Spee from a genetic perspective, rather they have used arbitrary measures of length to describe the depth of the curve and its variation. The advantage of using the twin

model is that it has allowed the initial steps to be taken towards explaining the curve of Spee from a genetic perspective.

The aims of the study were achieved to the extent that a relationship was found to exist between genetic factors and the final phenotypic variation of the curve of Spee. This relationship was found to be positive with evidence of moderate to high genetic influence. It must be emphasized that the identification of a trend is of importance as further studies using more sophisticated statistical tests and larger sample sizes will be necessary.

For the purpose of this study, heritability (h^2) estimates were generated. Harris (2008) has highlighted the use of such estimates and emphasized that the estimate provides a method of assessing the extent to which genetics affects a trait's variation in a specific population at a given time. As such it is a collective parameter not applicable to the individual. While it is possible to assess to a certain degree the extent of genetic influence on phenotypic variation, environmental factors also exert an influence. These factors consist of all the non-genetic influences and may include early tooth loss, extensive wear or even intervention such as restorative dentistry and orthodontic treatment.

The findings from the present study indicate that there is a genetic influence on phenotypic variation of the curve of Spee, although the estimates of heritability varied across the time points due to the relatively small sample sizes and limitations of the methods for calculating heritability values. The depth of the curve of Spee was found to increase across all three time points. It was found that about 50% of the phenotypic

variation could be explained by genetic factors at T1 and this was found to increase during T2 and T3. These results indicate a moderate to high genetic influence. Curve of Spee depth to date has been assessed by linear measurements to determine the maximum depth and to quantify the time point of greatest change. Studies by Marshall *et al.* (2008), Qabandi *et al.* (1999), Bishara *et al.* (1989) and Baydas *et al.* (2004) have considered environmental factors in the development of the curve of Spee while assessing the development of the curve with descriptive measurements.

Similarly to depth, the b1 and b2 asymmetry and shape coefficients have shown there to be a moderate to high genetic influence on the shape of the curvature. Particularly the extent of the curvature and asymmetry within the curve (differences between the anterior and posterior region of the curve of Spee). Approximately up to 80% of the variation in the values of the b1 and b2 coefficients may be explained by genetic influences.

The sample used in this study and the previous study are from a larger population group enrolled in an on going study of twins from the School of Dentistry, University of Adelaide. Previous studies using the same sample group have considered traits such as Carabelli cusp, tooth size, tooth morphology and arch shape, successfully identifying an underlying genetic influence. Higgins *et al.* (2009) have reported on hypocone expression within this sample and have found strong genetic influences. Narrow sense heritability estimates were used to determine a strong genetic influence on phenotypic variation. Dempsey *et al.* (2001) have also shown there to be strong genetic contributions to variation in human dental morphology, which is supported by other studies indicating an underlying genetic influence (Dempsey *et al.* 1995,

Hughes *et al.* 2000, Dempsey *et al.* 2001 and Higgins *et al.* 2009). Richards *et al.* (1990) described dental arch morphology in South Australian twins and suggested that genetic factors contributed to variation in both maxillary and mandibular arch shape. From these studies a common finding is the relationship between the various biological parameters and genetic influence. While no studies have been identified as assessing the curve of Spee's genetic influence, the findings in this paper provide a positive step forward in identifying a genetic influence on the development of the curve of Spee.

Research has tended to focus on dental similarities between MZ pairs rather than differences but, despite having the same genetic make up, studies have shown that phenotypic differences can occur with similar genotypes, whereby the phenotypic differences may be explained by environmental influences on the genetic expression. Townsend *et al.* (1995) reported findings of MZ pairs where one exhibited small, peg-shaped incisors and even agenesis where as the other exhibited a normal expression despite both having the same genetic make-up. This has further been investigated by Townsend *et al.* (2005) who postulated that minor variations in local epigenetic events during tooth formation could account for the variation observed in MZ twin pairs. Such factors may also be applied to the curve of Spee whereby some of the observed variation between twin pairs may be due to environmental factors such as early loss of a tooth, restorative intervention or tooth wear rate, as well as possible epigenetic influences. Corruccini and Potter (1980) have also stressed the need to consider environmental determinants. By investigating a series of occlusal traits in MZ and DZ pairs they found that significant heritability could not be demonstrated for overbite, overjet, buccal segment relation, tooth displacement and occlusal

discrepancy in arch shape, suggesting environmental determinants. The curve of Spee can also be considered in relation to these findings as it is associated with the above occlusal traits, thus suggesting environmental determinants on its phenotype.

11.8 Conclusion

From the results it can be seen that there is evidence of a genetic influence to phenotypic variation of the curve of Spee. This preliminary work, identifying a positive relationship, is statistically limited but is supported by findings from other genetic studies sharing this same sample group. Other occlusal features have shown that a genetic influence is present in final phenotypic variation. This paper has identified trends as the heritability values were only estimates and these values do have errors associated with them. Its intended to carry out more detail genetic modeling in the future.

Future directions for research could include increasing the sample size and performing further statistical tests of greater sophistication. Further clarification of genetic, environmental and epigenetic factors will need to be considered as well as the use of 3D scanning systems to provide more detailed descriptions of phenotypic variation. Further exploration of the MZ co-twin model should enable differences in expression of dental traits such as the curve of Spee to be identified. This could be further explored by assessing the epigenetic profiles of their DNA.

12. Concluding Remarks

Papers 1 and 2

The aims of the study were achieved whereby the curve of Spee development was described over three time points. The result from paper 1 showed that the curve of Spee development changes over time where the curve initially starts as a relatively flat structure in T1, deepening further at T2 and T3. Not only were changes in depth found to occur but also the extent of the curvature was found to increase and the degree of asymmetry within the curvature with respect to the anterior and posterior portion of the curve. This deepening of the curvature in T3 may be explained by differential tooth eruption, particularly the eruption of the lower mandibular second molar. This asymmetry also appears to be more marked with the eruption of the lower second molar with a further deepening of the curve.

Paper 2 further investigated the development of the curve of Spee by attempting to determine to some extent the genetic influence in the observed variation. From this preliminary investigation of the curve of Spee, evidence confirming a positive relationship was established. A moderate to high genetic influence can be seen from heritability estimates. Although the calculations are limited in that only broad sense heritability estimates were generated, other published evidence derived from this sample has indicated a positive relationship between genetic influence and biological parameters. Further research would increase the sample size and use more powerful statistical methods of analysis to enable epigenetic and environmental influences to be further quantified. Detailed dental histories, radiographic and photographic records

combined with 3D scanning techniques and DNA analysis would establish possible links and further expand our knowledge of dental development.

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