Aalen University

Clinical Effect of Tear Layer Thickness on Corneal Edema During Scleral Lens Wear

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Declaration

I, Christiane Arlt, assure that my final thesis with the topic "Clinical Effect of Tear Layer Thickness on Corneal Edema During Scleral Lens Wear" was issued by myself and that I did not use any other sources or help other than those indicated.

Sentences or parts of sentences quoted literally are marked as quotations; identification of other references with regard to the statement and scope of the work is quoted.

The thesis in this form or in any other form has not been submitted to an examination board and has not been published.

Buchbach, July 11, 2015

Christiane Arlt

Preface

The data included in the present study was collected and analyzed between September 2014 and July 2015, at the Augen-MVZ Landshut, Germany. I want to acknowledge several individuals who assisted me and acted as mentors during my thesis preparation:

Special thanks go to Ms. Prof. Dr. Nagl and Mr. Prof. Kümmel for the organization and mentoring of the course of studies.

Sincere thanks also go to my supervisor, Prof. Pat Caroline of Pacific University, and his colleagues, Prof. Dr. John Hayes (Pacific University) and Dr. Maria Walker of the University of Houston. I also thank my second supervisor, Bernd Brückner, and the company Appenzeller Kontaktlinsen, who supplied me with knowledge and donated the scleral lenses that were necessary for the measurements.

I would like to thank my bosses at the clinic of the Augen-MVZ, for allowing me to collect the data and use the technical equipment at the clinic. I would also like to express my appreciation for the ten volunteer subjects, without whom this study would not have been possible.

Last but not least, I cordially thank my family for their support and confidence. Most notably I thank my fiancé, motivating me again and again, my brother for providing me with his scientific experience, and my parents giving me a safe background but educating me to be independent.

Table of Contents

Abbreviations

ANOVA	Analysis of variance
BC	Base curve
CL	Contact lens
COP	Center of pupil
Dk	Oxygen permeability of a material
HVID	Horizontal visible iris diameter
n.p.p.	no publishing place
n.y.	no year
OCT	Optical coherence tomographer/tomography
OU	Oculus uterque – both eyes
OD	Oculus dexter – right eye
OS	Oculus sinister – left eye
PMD	Pellucidal marginal degeneration
SAG	Sagittal depth

Abstract

Purpose: Although the frequency in which practitioners are fitting scleral contact lenses is increasing, the recommendation for proper tear layer depth (thickness) varies amongst experts. The main goal of this paper is to clinically verify the effect of varying tear layer depths on induced corneal edema during lens wear.

Methods: Ten subjects with healthy eyes were fitted with scleral lenses on their right eye. Each of them was fit with two different lenses: one with an apical clearance of 200 µm and another with an apical clearance of 600 µm. They wore the lenses for 8 hours on two different days, with at least a one week wash-out period. Lenses were applied at 8 a.m. on each of the testing days. Pachymetry measurements were taken one day prior to lens wear at 4 p.m., on the day of wear prior to lens application, and after removal of the lenses at 4 p.m. Measurements were collected using both the Pentacam® HR Corneal Tomographer, as well as the Visante Anterior Segment Optical Coherence Tomographer (OCT). The apical clearance was measured using the Visante OCT at two intervals during the test day: immediately after application of the lens and immediately prior to the removal of the lens.

Results: In this study, there was found to be no significant difference in corneal edematous response during lens wear between the two test groups. The study shows that the eyes with the lenses have a statistically significantly thicker cornea compared to the non-lens-wearing eye after wearing either lens for 8 hours, lying within clinically and physiologically acceptable limits.

Conclusion: Our clinical results do not correlate with current theoretical calculations, which predict a greater amount of corneal swelling with increasing tear layer thickness. It has to be evaluated if the effect on corneal edema changes with longer wearing periods, larger samples or other influences.

Key words: scleral (contact) lens, corneal edema, pachymetry, tear layer thickness, vaulting, apical clearance

1 Introduction

1.1 Motivation and Objective

Scleral lenses are gaining more and more importance in places where contact lenses are fitted. Nevertheless, there are unknown components that are discussed in literature and need further research.

One large chapter is the influence of the tear lens beneath the scleral lenses. The uncertain exchange, debris within and the influence on the oxygen supply are only some factors that could be studied. Referring to the last mentioned element, there was a noticeable development concerning the contact lens material, which was a crucial factor for the revival of scleral lenses. When extending *"existing single lens models to circumstances where two lenses offer resistance to oxygen in series"* (Weissman, 2006), it shows up that the oxygen permeability of the tear lens stays, for physiological reasons, on the same level. The theoretical findings of Michaud (2012) are a milestone in the evaluation of oxygen availability to the cornea during scleral lens wear. Due to their findings, they recommend a combination of minimal vaulting, thin lenses and high Dk, which might limit the practitioner's scope and is difficult to implement in special corneal conditions.

This study aims to evaluate the clinical effect of the tear layer thickness on corneal edema during scleral lens wear. In particular, it investigates the amount of edema during lens wear, and whether or not there is a significant difference in edema with different values of corneal vaulting.

1.2 Structure

The introductory section shows the general initial situation, the motivation and the goal of the study. The second chapter contains fundamental anatomical knowledge, and discusses the current available knowledge in fitting scleral lenses. This serves as a basis for all further considerations and conclusions. Subsequently, the third chapter describes the methods, respectively the course of the study. The fourth chapter describes the results of the statistical analysis while the fifth chapter shows pachymetry changes in percent and cares about their clinically relevance. This prefaces the concluding chapter which summarizes the study, reviews the difficulties of the practical implementation of the study and compares itself to other studies. It also gives a prospective view to further possibilities for studies in this field.

2 Terms and State of the Art

2.1 Anatomy and Physiology of the Anterior Ocular Surface

Contact lenses in general are, as the name suggests, in “contact” with the anterior segment of the eye. That is why this chapter gives an understanding of these different structures.

The anterior segment of the eye refers to all ocular components extending from the front surface of the vitreous humor to the front surface of the cornea (Millodot, 2009). In the present study, the main focus is on the visible surface of the eye within the palpebral fissure. This contains the sclera, conjunctiva, and the cornea (including the limbus).

2.1.1 Sclera

The sclera, which is the rigid collagenous shell encasing the majority of the intraocular contents (Snell, 1998), is influenced by forces from inside and outside and is for that reason significantly responsible for the roughly spherical shape of the eye (Berke, 1999). Maidowsky (1980) describes that from a histological point of view, the scleral stroma consists of bundles of flat white collagen fibers, which are made out of many very fine fibrils. Additional elastic fibers allow the sclera to respond to eventually deforming forces. As a result of the irregular crisscross pattern of the fibers, the sclera gains its opacity (Snell, 1998) and mechanical stability (Berke, 1999). While the scleral stroma itself has very few blood vessels and nerves (Maidowsky, 1980), the overlying layer, the episclera, has a rich blood supply. These vessels are normally inconspicuous but become very red and congested in the presence of inflammation (Snell, 1998).

The low nerve density within the sclera is fortuitous for the scleral lens wearing individual, since the scleral lens rests on this relatively insensitive tissue. Due to this low sensitivity, these lenses do generally not cause significant foreign body symptoms.

The thickness of the sclera varies considerably in different areas of the eyeball: at the corneoscleral junction it is 0.8 mm thick, thinning to 0.3 mm immediately posterior to the tendinous insertions of the recti muscles. At the equator it is 0.6 mm thick and it is thickest (1 mm) posteriorly. In addition, the insertions of the muscles contribute to the non-rotational shape of the eyeball, in particular the recti muscles, which insert 5.5 mm to 7.7 mm posterior to the limbus (see figure 2-1).

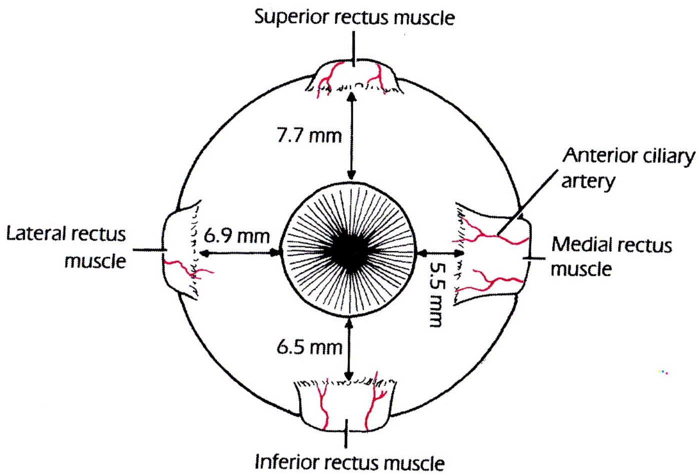

Figure 2-1: Insertion of the muscles (Snell, 1998)

The insertions of the superior oblique and inferior oblique muscles are posterior to the scleral equator (see figure 2-2). (Snell, 1998)

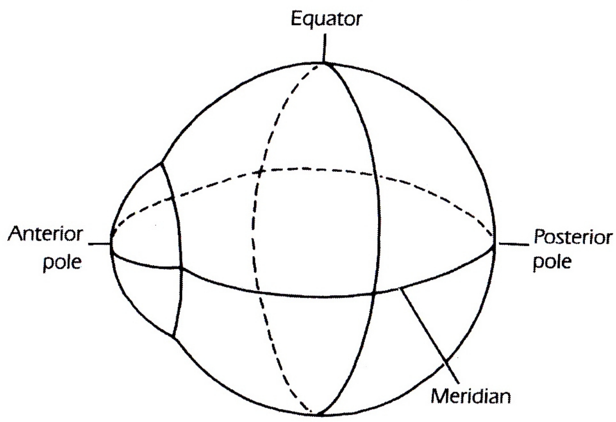

Figure 2-2: Eyeball, showing the poles, equatorial and meridional planes (Snell, 1998)

Tina Graf (2010) studied the limbal and anterior scleral shape and concluded that the anterior scleral surface is asymmetric:

> *“The limbal portion is flatter than the scleral one except in the nasal direction which also shows the flattest average angles and is more often concave than all other directions.*
>
> *Superior and temporal both the limbal and the scleral average angles are roughly in the same range. In the inferior direction where most of the surfaces are rated straight, the limbal and the scleral average angles do not differ.“*

This has to be considered when fitting scleral contact lenses, especially when fitting large diameter scleral lenses (greater than 15.0 mm) (van der Worp, 2010).

2.1.2 Conjunctiva and Limbal Area

The bulbar conjunctiva overlies the anterior globe of the eye, in which the sclera designates the form of the eye. It is loosely bound to the scleral tissue beneath it, and is malleable and susceptible to blood or other fluid accumulation (Kaufman, 1988). Together with the eyelids, the conjunctiva provides physical and metabolic protection for the cornea (Kaufman, 1988).

The limbal area is the transition area between conjunctiva and cornea and sclera and cornea. The conjunctiva thins out and becomes the corneal epithelium while the irregular collagen fibers of the sclera change to form more regular collagen patterns, guaranteeing the transparency of the corneal stroma. Blood and lymphatic vessels end here. (Maidowsky, 1980)

While the radius of curvature of the cornea is about 8 mm, the sclera with its 12 mm is much flatter, which results in a more or less distinct sulcus sclerae (Berke, 1999), also known as corneo-scleral profile (see figure 2-3).

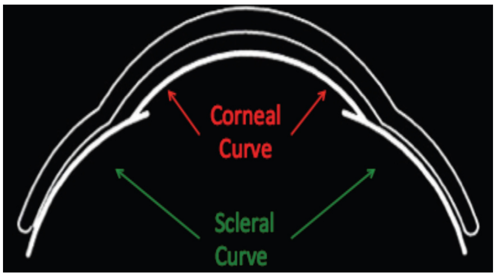

Figure 2-3: Corneo-scleral profile - Traditional (Caroline, 2013)

This is, however, a very traditional way of thinking. While the cornea is not spherical to its edges, the current knowledge (Pacific Scleral Lens Study) shows a tangential transition in the limbal area (see figure 2-4).

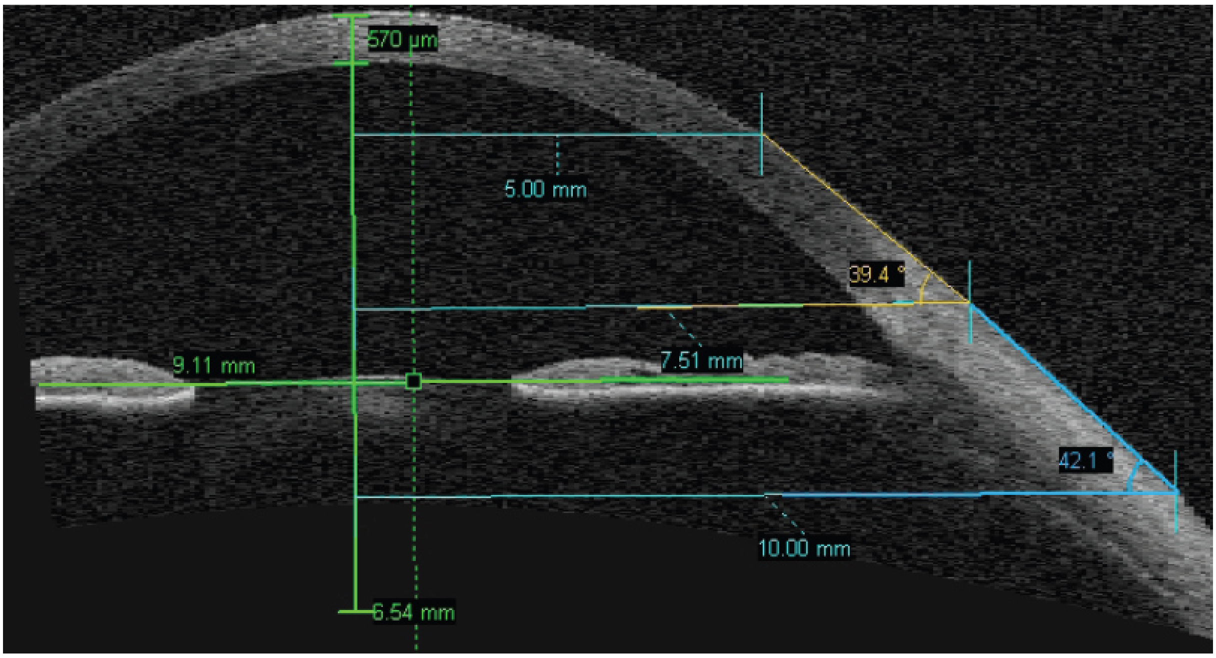

Figure 2-4: Corneo-scleral profile - Modern (Caroline, 2013)

Palisades of Vogt are located in the limbal area. This is the home of the epithelial stem cells, which guarantee epithelial regeneration and maintain the corneo-conjunctival border (Naumann, 2008). If limbal stem cells are partially or totally exhausted, this results in varying degrees of stem cell deficiency, ending in ocular discomfort and reduced vision (Dua, 2000). In opposition to the cornea, which does not contain blood or lymphatic vessels *"(so-called 'corneal antiangiogenic privilege')"* the limbal area is home for an intense network of them (Naumann, 2008). The occurrence of limbal stem cell deficiency is usually indicated by conjunctival epithelial ingrowth, vascularization and chronic inflammation, followed by recurrent erosions and persistent ulcer as well as destruction of the basement membrane which leads in its totality to pertinent functional disturbance (Puangsricharern, 1995 & Holland, 1996 as cited in Naumann, 2008). In order not to aggravate the situation of the eventually already compromised eye when fitting a scleral lens, the practitioner should strive to avoid the scleral lens resting on the limbus (Walker, 2014; van der Worp, 2010).

2.1.3 Cornea

Lying in the center of the palpebral fissure, the cornea is responsible for 2/3 of the refractive power in the eye. This importance *"to the ocular structure and visual system is often overlooked because of the cornea's unassuming transparent nature"*. (DelMonte, 2011)

The average horizontal corneal diameter, or also called horizontal visible iris diameter (HVID), is about 11.7 mm according to Rüfer (2005) and Naumann (2008). The vertical diameter is smaller. Over all, the cornea comprises 1/6 of the outermost ocular tunic (Kaufman, 1988). The anterior surface of the cornea is approximately spherical in the center and gets flatter in the periphery (see figure 2-5) while the posterior surface is more spherical which leads to a greater corneal thickness in the periphery than in the center of the cornea (Kaufman, 1988) (see figure 2-6).

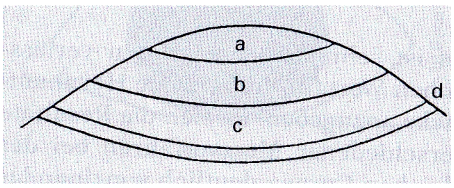

Figure 2-5: Corneal surface, showing the asphericity (Watkins-Profile) (Baron, 2008)

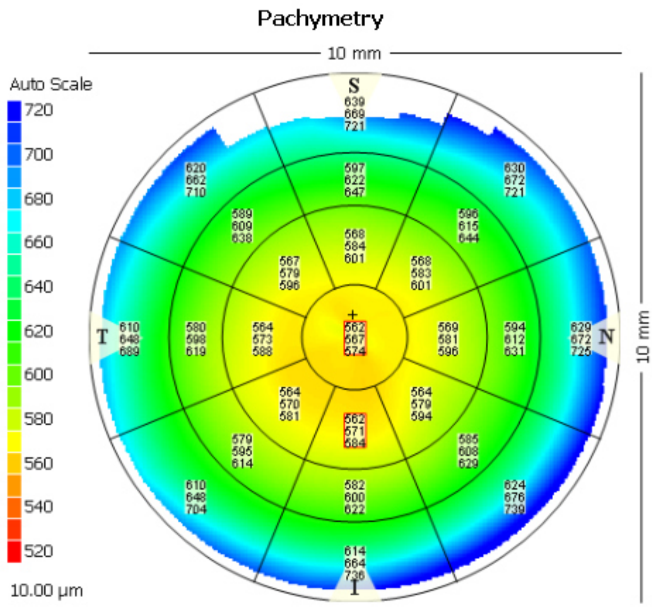

Figure 2-6: Increase of the pachymetry from the center to the periphery

The cornea of human beings consists of 5 recognized layers (DelMonte, 2011). These are all crucially involved in keeping the cornea avascular, transparent and in an anti-inflammatory state. Altogether, they play an important role in keeping good visual quality (Naumann, 2008). Each layer is explained separately hereinafter.

Epithelium

The body’s cornea is one of the most densely innervated and sensitive tissues. (DelMonte, 2011) and these numerous nerve endings are located among the epithelial cells (Naumann, 2008). The epithelium is the outer layer of the cornea and has a first barrier property (Kaufman, 1988). It prevents infections by its 50 - 100 µm[1] thick (Naumann, 2008), stratified, non-keratinizing squamous surface, characterized by extreme uniformity from limbus to limbus (DelMonte, 2011). The whole epithelium renews itself every 7 days (Dohlman, 1970 as cited in Kaufman, 1988) which guarantees a good wound healing without scarring. The primary function of the corneal epithelium, however, is to guarantee adherence of the mucin layer of the tear film in order to evenly spread moisture on the corneal surface to prevent evaporation and optical degradation (Kauf-

[1] µm: micron = 10^{-6} m

man, 1988). The innermost layer of epithelial cells is attached to a basement membrane which is adherent to the Bowman's layer (Snell, 1998).

Bowman's layer

Bowman's layer, which is historically referred to as a membrane, is a 8 - 12 µm (Kaufman, 1988) thin amorphous band. It is responsible for maintaining most of the tensile strength of the corneal tissue (Naumann, 2008) but following injury it scars and does not regenerate. Collagen fibrils of the Bowman's layer guarantee a tight attachment to the corneal stroma by merging into the anterior stroma (Kaufman, 1988).

Stroma

Ninety percent of the corneal thickness is comprised of the corneal stroma (Kaufman, 1988) which is made up of highly arranged collagen fibrils, allowing it to maintain transparency (Naumann, 2008). It has a normal water content of about 78% (Smolin, 1987; Kaufmann, 1988; Berke, 1999), which is higher than that found in most of the body's connective tissue, and is attributed to the high water-holding capacity of the proteoglycans within the stromal matrix (Smolin, 1987). The tendency to absorb water, called the stromal swelling pressure, has to be counteracted constantly to keep a deturgesced state in order to guarantee the transparency. There are several mechanisms for the regulation, including barrier function of epithelium and endothelium, water-pumping mechanism of the endothelium, evaporation from corneal surface and intraocular pressure. (Smolin, 1987) If disturbances occur to this regularly arranged structure, cloudiness within the cornea is caused (Naumann, 2008).

Corneal hypoxia can be one etiology for excess water within the stroma, which ultimately can result in increased corneal thickness and corneal opacification. Early objective findings, visible using a biomicroscope, are vertical striae which are not likely to affect visual acuity. At approximately 5 - 6% corneal swelling, single central striae may become visible. If the cornea reaches a level of 6 - 10% swelling, several striae, mainly in vertical orientation, are likely to be seen. If the level of edema is raised above 10%, dark folds can be seen in the area of the Descemet's Membrane and above 20%, evident loss of transparency (haze) will likely occur, which cannot be accepted during contact lens wear under any circumstances. (Müller-Treiber, 2009)

Descemet's Membrane

Located posterior to the stroma, Descemet's membrane is the basement membrane of the endothelium, and is about 10 µm in thickness (Snell, 1998). The membrane thickens with age (Smolin, 1987*).* Breaks in this tissue (i.e. corneal hydrops occurring in Keratoconus) can result in fluid migration from the aqueous humor into the corneal stroma. This can significantly reduce the tensile strength and the function of the cornea (Naumann, 2008).

Endothelium

The posterior surface of the human cornea consists of single layer pump cells that are essential for optimal corneal functioning (Kaufman, 1988) because they are responsible for maintaining proper hydration of the cornea (Naumann, 2008). The endothelium appears as a *"honeycomb-like mosaic"* (DelMonte, 2011), and its cells are post-mitotic and usually not able to regenerate (Naumann, 2008). Influenced by age, intraocular surgery or trauma, the cells change their shape (pleomorphism), size (polymegethism) and number (cell density) (Kaufman, 1988). While pleomorphism and polymegetism can also be observed due to contact lens wear, the cell density seems to remain stable (Orsborn, 1988). According to Müller-Treiber (2009), the pathogenesis of the changes is not completely clear, but is likely triggered by chronic hypoxia that leads to the accumulation of lactate and carbon dioxide in the cornea.

2.1.4 Physiology of the Cornea

Endothelial cells control the hydration of the corneal stroma by two mechanisms. First, they act as a physical barrier between the corneal stroma and the aqueous humor, disallowing fluid from the aqueous to inadequately flood into the corneal tissue. Second, they have active transport mechanisms that pump water in or out of the cornea to control hydration. (Snell, 1998)

While the metabolism of the cornea usually proceeds on the edge of the physiologically tolerable, small disturbances can seriously change the circumstances within the cornea. Alterations in the epithelium or formation of corneal edema can be the consequence. (Berke, 1999)

2.2 Scleral Lenses

2.2.1 History

The idea of scleral lenses dates back to the thoughts of Leonardo da Vinci in 1508, who described an optically neutralizing effect by a liquid reservoir directly in front of the corneal surface (da Vinci, circa 1508 as cited in Bückner, n.y.). The first scleral lenses at the end of the nineteenth century were glass blown shells to ensure the vaulting effect (Fick, 1888). Some important breakthroughs had to follow in order to develop this lens modality. Smaller, gas permeable lenses, only resting on the cornea were developed, followed by soft lenses. This temporarily interrupted further progression of scleral lens fitting, but the thus developed high gas permeability of materials and improved manufacturing technologies allowed a revival of the old lens modality (van der Worp, 2010; 2014). Several studies showed that the central corneal thickness increased less with gas permeable compared to polymethylmethacrylate (PMMA) lenses (Pullum, 1990).

2.2.2 Nomenclature

While scleral lenses have a long historical background, there are many different kinds of naming schemes and fitting recommendations. The Scleral Lens Education Society (2013) recommends internationally recognized nomenclature. The bearing of the lens is the defining feature in this terminology: resting completely on the cornea, a lens is called corneal lens. If a lens rests partly on the cornea and partly on the sclera, it is called a corneo-scleral lens and if a lens vaults the cornea and only rests on the sclera, professionals are talking of a scleral lens. The recommended nomenclature does not intend a differentiation due to diameter within the scleral lens group. When dividing the scleral lens group into mini-scleral and large-scleral lenses, the HVID should be used as a reference: lenses that are up to 6 mm larger than the HVID are classified as mini-scleral lenses. Larger than that, lenses are classified as large-scleral lenses (Scleral Lens Education Society, 2013), summarized in table 2-1.

Lens Type	Description	Definition of Bearing Area
Corneal		Lens rests entirely on the cornea
Corneo-scleral		Lens rests partly on the cornea, partly on the sclera
Scleral	Mini-Scleral *Lens is up to 6mm larger than HVID*	Lens rests entirely on the sclera
	Large Scleral *Lens is more than 6mm larger than HVID*	

Table 2-1: Nomenclature (Scleral Lens Education Society, 2013)

The lenses used in the present study are part of the mini-scleral category. Their diameter is chosen dependent on the horizontal visible iris diameter, according to the fitting guidelines of the manufacturer: Appenzeller Kontaktlinsen, Switzerland.

2.2.3 Applications of Scleral Lenses

The initial idea of optically neutralizing the corneal surface is an important property of scleral lenses. Though, the existing tear lens underneath the contact lens has other positive effects, too. It grants a stable wetting of the corneal surface, which is an important factor in severe dry eye conditions. The following section describes these mentioned and some more applications in detail, divided into categories according to the model of the “Guide to Scleral lens fitting” of van der Worp (2010).

Vision improvement

While a corneal rigid gas permeable lens also ensures the optical neutralization, there might be contraindications that make the fitting of those unfeasible. Foreign body sensation, drying of the peripheral cornea, inacceptable decentration of the lens or in very irregular or prominent cases even the fact that the lens does not stay on the cornea at all, are only some reasons. But especially because these corneas are highly irregular, they need a visual rehabilitation, which the scleral lens is able to guarantee. Among other studies, that’s confirmed by Visser (2007) who determined that keratoconus was the number one indication for scleral lens wear, followed by patients who had undergone a penetrating keratoplasty. Altogether, in the following cases, the visual rehabilitation plays an important role:

- Primary Corneal Ectasia:
  - Keratokonus
  - Keratoglobus
  - Pellucidal Marginal Degeneration (PMD)
- Secondary Corneal Ectasia:
  - Post-LASIK
  - Post-LASEK
  - Post-PRK
  - Post-RK
  - Post-Trauma
- Corneal Transplant
- Corneal Scars Post-infection
- Corneal Dystrophies or Degenerations

Corneal protection

Scleral lenses need to be filled up with preservative free saline, before being placed on the eye in order to keep the surface of the cornea moist to prevent damage to the epithelium (Kaufman, 1988). This tear lens appears to have minimal tear exchange over an eight-hour wear period (Morrison, 2014), but maintains the hydration of the affected cornea over the whole wearing time.

Patients who suffer from abnormalities linked with severe dryness of the ocular surface benefit of this wetting effect:

- Ocular Surface disease:
  - Sjörgen’s Syndrome
  - Steven’s Johnson Syndrome
  - Graft versus Host Disease
  - Neurotrophic Corneal Disease
  - Ocular Cicatricial Pemphigoid
  - Atopic Keratoconjunctivitis

- Incomplete Lid Closure
  - Eyelid Coloboma
  - Exophthalmus
  - Ectropion
  - Nerve Palsies
  - After Lid Retraction Surgery

Cases of entropion, trichiasis and distichiasis also benefit of the protective role of the scleral lens itself.

Cosmetics /Sports

Scleral lenses can also be helpful in disfiguring diseases, both in primary abnormalities like atrophia bulbi and secondary to trauma or equivalent.

For those who are participating in active water sports or are exposed to dusty environments, the scleral lenses are also useful.

In a study of Romero-Rangel (2000), 92% of patients who were fitted with scleral contact lenses showed visual improvement and an increased quality of life.

2.2.4 Fitting Scleral Lenses

Since scleral lenses completely vault the cornea, they are generally fit to accommodate the sagittal depth of the anterior segment (van der Worp, 2010). The shape or condition of the cornea underneath is thus less important. In classic measurement methods, there is no way to measure the sagittal depth of an eye based on a diameter larger than the cornea. Instrumentation such as the anterior segment Optical Coherence Tomographer, or very recently the Eye Surface Profiler (ESP) might one day change the methods of fitting scleral lenses.

Currently, it is clinical practice and recommended by manufacturers to use a fitting set of the particular lens type (van der Worp, 2014).

Van der Worp describes in his Guide to Scleral Lens fitting (2010) a Five Steps Fitting Approach, independent of manufacturer and design. As the lenses fitted in this present study are manufactured by Appenzeller Kontaktlinsen, Switzerland, called i-MATRIX, and their Fitting Guide correlates with the mentioned fitting approach, the lens design is introduced before their recommendations are elaborated in five steps (both according to Brückner, n.y.).

The back surface of the i-MATRIX consists of three zones as shown in figure 2-7: the central, spherical zone (labeled BOZ) with the sagittal depth x0 and according to the findings of the Pacific Scleral Lens Study two tangential zones in the periphery: the limbal zone (labeled BPZ 1) with sagittal depth x1 and the scleral zone (labeled BPZ 2) with the sagittal depth x2.

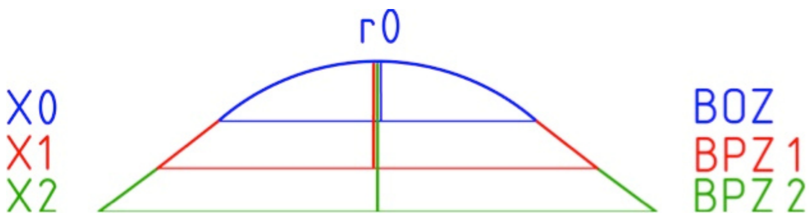

Figure 2-7: Zones of the i-MATRIX (Brückner, n.y.)

Subsequently, the five steps are delineated.

Choice of the diameter

The diameter of the final scleral lens is dependent on the individual corneal diameter as mentioned above. The Appenzeller fitting guide recommendations are shown in the following table 2-2. Accordingly, the diameter of the first lens should be chosen.

i-Matrix diameter choice in relation to HVID

HVID	i-Matrix diameter
Smalls (≤ 11.5 mm)	16.0 mm
Medium (11.6 mm–12.1 mm)	16.5 mm
Large (≥ 12.2 mm)	17.0 mm

Table 2-2: Table for i-Matrix diameter choice (Brückner, 2015)

Choice of the central base curve (BC)

Because the complete cornea is vaulted by this type of lens, opposed to a classical corneal lens, the radius of the cornea does not play a role in choosing the central base curve of the lens. Appenzeller Kontaktlinsen primarily recommends deciding for one out of three base curves of the fitting set due to the indication. Additionally, the practitioner might incorporate very extreme obvious sagittal depths.

They provide practitioners with the following table 2-3 including first choice and possible reaction counsel.

BC/SAG (10 mm chord) selection based on indications

Indication	Description	BC/SAG
Indications linked to irregular corneas	• Corneal ectasia (Keratoconus, PMD), • Intacs • post-surgical ectasia • Keratoplasic • trauma	Start with BC 7.2 (medium SAG 2.02) A) If the CL rests on a widespread area, change to BC 6.6 (steep SAG 2.29) B) If the CL bridges excessively, change to BC 7.8 (flat SAG 1.81)
Indications linked to dry eye	• Pronounced Sicca-Syndrome • Steven-Johnson Syndrome • Sjörgen Syndrome • chemical burn • Neurotrophic-Keratitis • Keratopathy Filamentosa	Start with BC 7.8 (flat SAG). If the CL rests on a widespread area, change to BC 7.2 (medium SAG 2.02)

Table 2-3: Table for i-MATRIX BC choice (Brückner, 2015)

As soon as these two main parameters are determined, the lens of the fitting set has to be assessed on the patient's eye. For this purpose, the lens has to be applied filled up with sterile saline and fluorescein because both of them cannot be brought underneath the scleral lens when the lens is once in place. While assessing the lens, the following aspects should be respected:

Vaulting of the cornea

The flourescein already applied together with the lens and the saline allows assessing the vaulting of the cornea the classical way with the slit lamp. At first, the vaulting should be assessed by looking at the overall fluorescein picture (see figure 2-8): direct focal illumination, slit with maximum size and low magnification should guarantee an overview of the whole lens if brightness is maximum and lids are not touching the lens as far as possible.

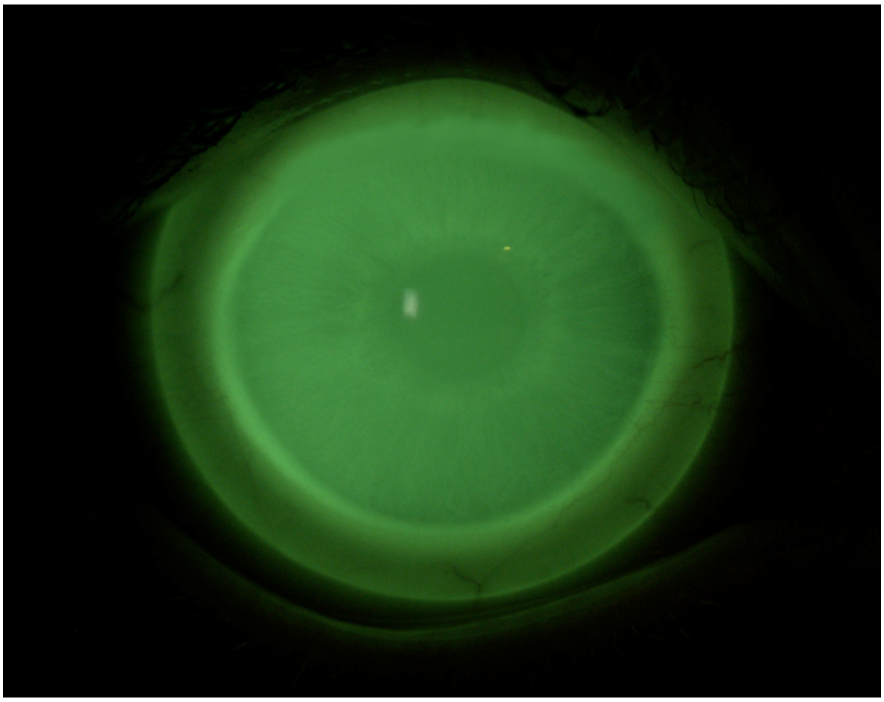

Figure 2-8: Fluorescein assessment of a scleral lens

This observation method allows a quick check for air bubbles, which would not disappear over time, and if the whole cornea is vaulted sufficiently. If there are dark areas, which indicates the lens is in contact with the cornea or the limbus, the sagittal depth of the lens is inadequate and has to be increased.

Knowing that as clinicians we are only able to observe 20 µm or more of fluorescein beneath a lens on the eye (van der Worp, 2014), this first method does not allow a statement concerning the depth of the vaulting. Although, the translucence of the pupil might be a small hint, when fitting scleral lenses, a more exact method is in demand.

Using the optical section of the slitlamp (see figure 2-9) and knowing the center thickness of the applied lens, the corneal clearance over the whole cornea can be estimated. For practical purposes, this is, in the author’s viewpoint, sufficient.

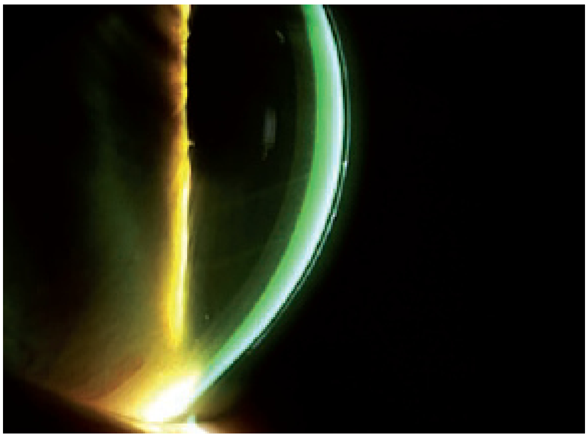

Figure 2-9: Optical section of a cornea, tear lens with fluorescein and scleral lens

Advanced visualization methods like OCT (Optical coherence tomography) or Scheimpflug allow direct measurements of the vaulting as shown in figure 2-10:

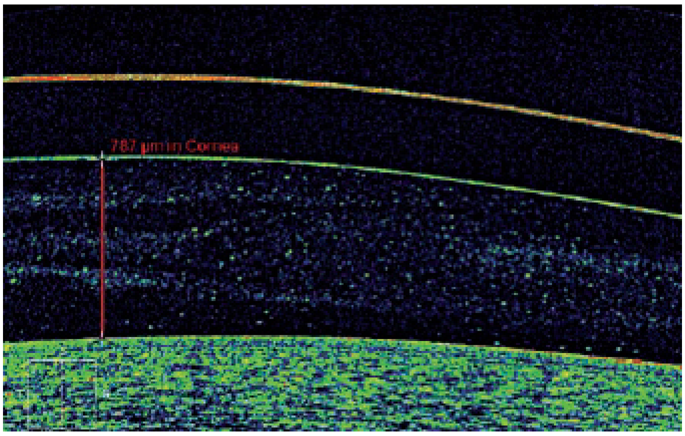

Figure 2-10: OCT visualization of cornea (bottom of picture), tear lens and scleral lens (Brückner, n.y.)

When measuring or estimating the vaulting, one should keep in mind the fact of artefacts produced by OCT and triggered by deviant refractive indices (Chhablani, 2014). Additionally, the sinking of the scleral lens into the conjunctiva has to be kept in mind, which is subject to a high individual variance (van der Worp, 2014). The amount of vaulting changes with measurements at different wearing times.

Appenzeller Kontaktlinsen recommends an assessment directly after appliance and a wearing time of 2 hours to the next measurement if initial clearance was acceptable.

Bauer (2015) from Ferris State University presented an equation how to calculate the resulting value after 5 days:

$$\textit{Final Settling} = 4\ (\textit{Initial vault} - \textit{Vault at 20 minutes})$$

There is no general recommendation for the ideal corneal clearance. In practice, usually the clearance of 200 µm to 600 µm is applied (van der Worp, 2014). Appenzeller Kontaktlinsen recommends an initial corneal clearance of around 350 µm.

This value is suspected to be critical concerning the oxygen supply as discussed in this study in detail in the following chapters.

Just of the same importance as the central corneal clearance is the peripheral corneal and limbal clearance. As described above, the important stem cells reside in this area. They should never be exposed to pressure from the scleral lens. Fitting the i-MATRIX and seeing peripheral corneal or limbal rest, the choice of the diameter should be checked. If correct, the sagittal depth has to be increased in the limbal zone x1 (see figure 2-7).

Fitting of Landing Zone

Vaulting the whole cornea and limbus, the total weight of the lens is on the sclera. The goal for the scleral zone is to land on the sclera without excessive edge lift and blood vessel barring (van der Worp, 2014). In the objective slit lamp evaluation in white light, the small vessels of the conjunctiva should be assessed. Whitened areas imply blood restriction and show due to their position, if the scleral zone is too steep or too flat.

Indications for alternate geometries

If the lens or the eye shows nonrotational signs like lift off or blanching in certain quadrants, lenses with a scleral-toric (i-MATRIX ST) or scleral-quadrant specific (i-MATRIX S-QSD) back surface become necessary.

2.2.5 Oxygen Supply with Scleral Lenses

Because scleral lenses have other characteristics of fit than corneal rigid lenses, the altered oxygen supply is in question. There is, if any, only minimal tear exchange (Morrison, 2014), typically no or very little movement (van der Worp, 2010) and a significantly thicker tear lens compared to corneal lenses. Similar to piggyback systems, the scleral lens system can be seen as two lenses, offering resistance to oxygen in series (Weissman, 2006).

The oxygen permeability of contact lenses is commonly described by Dk, while D displays a diffusion coefficient and k gives evidence of the oxygen solubility (Tranoudis, 1995 as cited in Michaud, 2012). Dk is an individual attribute of every material depending on the oxygen-permeable moieties, determined by *"polymer physic-chemical characteristics, water content and silicone composition"* (Michaud, 2012).

The resistance to oxygen in series has been previously studied, mainly by theoretical approaches: Weissmann (2006) extended single lens models to resistance in series by two lenses applied in piggyback fittings. They found acceptable values applying modern contact lens materials under open eye conditions.

Michaud (2012) put the focus on scleral lenses and calculated tables of combinations of scleral lens material and thickness of both scleral and tear lens, referring to the classic Holden-Mertz and Harvitt-Bonanno criterias, to avoid corneal swelling during lens wear. They assumed the Dk of tears being 80 Dk units (Benjamin, 1994 as cited in Michaud, 2012). Thus they found *"the ideal combination of scleral lens/tear clearance should be as follows: a lens made of the highest Dk available, designed with a maximal central thickness of 250 µm, and fitted in a manner to achieve a clearance that does not exceed 200 µm"* (Michaud, 2012).

In 2014, another research group studied the subject split in two methods. On the one hand also in a theoretical model, on the other hand in a clinical trial. The theory did not show any combination which avoids hypoxic effects, whereas the clinical trial showed edemas within at least physiological levels. In the practical trial, a group of eight subjects was fitted randomly with a post-lens tear film thickness of 150 µm and with a post-lens tear film thickness of 350 µm (Dk 100). It showed statistically significant corneal thickness changes (see figure 2-11), in which the deeper tear reservoir induced the highest value of swelling.

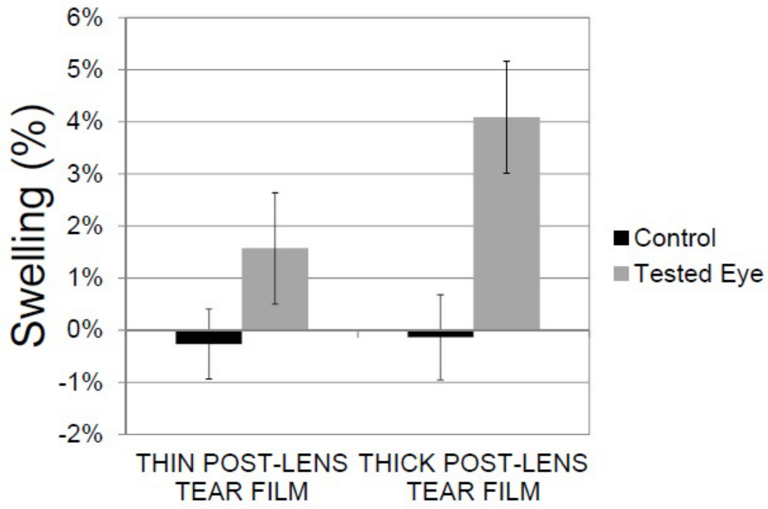

Figure 2-11: The mean and standard deviation values of corneal swelling (Compañ, 2014)

The figure shows a higher corneal swelling of the eye that wore the lens with the thick (350 µm) post-lens tear film (right column) compared to the eye that wore the lens with the thin (150 µm) post-lens tear film (left column).

They concluded, that scleral lenses must be comprised of a Dk of at least 125 and be no more than 200 µm thick. The post-lens tear film layer should be below 150 µm in order to avoid clinically significant edema. (Compañ, 2014)

Achong-Coan (2014) compared corneal thickness variation post scleral lens wear of different materials with different Dks without referring to tear lens thickness and found no striking differences in corneal swelling from Dk 65 to Dk 125. Compared to their overnight corneal swelling study, the swelling with all materials was within this limit.

Despite the results of these studies, few clinicians have observed clinically significant corneal edema in their scleral lens wearing patients (Achong-Coan, 2014). Therefore, the present study will clinically analyze the effect of tear layer thickness on corneal edema. The conditions and methods are exposed in the following chapter.

3 Methods

3.1 Design of the Study

This study was a prospective experimental study to prove what was hypothesized in advance. The data was collected by the author according to the hypothesis.

3.2 Subjects

The subjects are all employees at the eye clinic "Augenmedizinisches Versorgungszentrum" in Landshut, Bavaria, Germany. They all took part voluntarily and without financial refund.

Ten female subjects were included in the clinical trial. The average age within the sample was 34.5 years with a standard deviation of 11.06 years at the time of the measurements.

Selection criterion was, that both eyes of the subjects were healthy and without any corneal abnormalities. Post-surgery corneas were excluded, just as contact lens wearing subjects. Additionally, it was obligatory, that a rotationally symmetric geometry of scleral lens could be fitted adequately. If not possible, these subjects had to be excluded in order to create equal conditions.

3.3 Measuring Tools

Pachymetry

The thickness of the cornea (= pachymetry) was measured at all times with two different devices: Pentacam® HR (OCULUS, Germany) and Visante OCT Model 1000, Software-Version 3.0 (Carl Zeiss Meditech, Germany).

They apply completely different measuring principles: The Pentacam® HR is a combination of a slitlamp and Scheimpflug system rotating around the eye (see figure 3-1) that allows precise measurement of the corneal topography. The camera, pivoted out of the axis of the observation unit, takes pictures of the optical sections which are generated by the slitlamp. Twenty-five of these radially oriented images are combined. This enables high quality mappings, picturing the whole anterior segment of the eye including the posterior surface of the crystalline lens (if mydriatratic drops were instilled). Most importantly in this study, the pachymetry of the cornea can be displayed, which was used for the analysis of the collected data. (OCULUS Optikgeräte GmbH, n.y.)

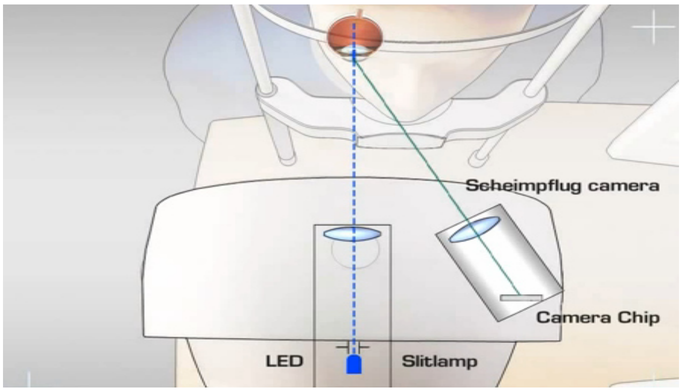

Figure 3-1: Setup of the measurement principle of the Pentacam® HR (Qvision, 2011)

The Zeiss Visante OCT however works on the principle of optical coherence tomography which is a noninvasive technique taking high precision recordings of the human tissue. With its echo technique, the OCT is similar to ultrasound imaging but it uses light instead of sound which allows high resolution. By detecting the intensity of the reflected light from the relevant layers via interferometry, OCT develops clear three-dimensional models (see figure 3-2). (Garg, 2014)

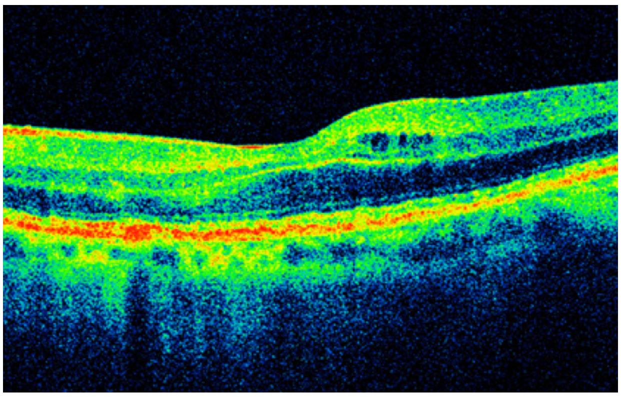

Figure 3-2: OCT image of the retina, showing the individual layers, detected by different reflection of light

OCT scanners have become commercially available for retinal imaging but a few modifications allowed the imaging of the anterior surface, for which the Visante OCT is designed. (Muscat, 2002)

Muscat (2002) found that OCT measurements of the human pachymetry correlate to those of recognized measuring methods and that those measurements are repeatable, without the need to take multiple readings for a certain value.

In the present study, the scan-type “Global Pachymetry Map” was used for the pachymetry measurement. All scans are line scans, called scan primitives. Contrary to the “Pachymetry Map”, the “Global Pachymetry Map” uses 16 instead of 8 scan primitives and the separation angle accordingly is halved (Carl Zeiss Meditec, 2012).

Tear Layer Thickness

The amount of vaulting, which was the crucial factor in the study, was measured with the Visante OCT. According to the author, it allows, contrary to other devices or methods, the most exact and reproducible values: The Zeiss Cirrus OCT (Carl Zeiss Meditech) allows very exact measurements but it is, in the author’s opinion and with the equipment at the Augen-MVZ, not possible to take re-measurements at the same spot. Examination at the slit-lamp as it was done in the study of Compañ (2014) by estimating the tear layer thickness appears having low reproducibility. Due to the fact that the OCT is initially designed for measuring the cornea, not being vaulted by a contact lens with a deviant refractive index, artefacts have to be expected (Chhablani, 2014). The consequence is, that measured distances do not originally reflect the truth but best possible with the current technical possibilities. The Visante OCT allows a measurement called ‘Anterior Segment Single’ where the anterior segment can be fully shown. The scan shows a width of 16 mm and a depth of 6 mm and measurement within the recorded picture is possible. In figure 3-3 you see the scan of an eye involved in the study in inverse colors wearing a scleral lens where the measurement tool is already placed.

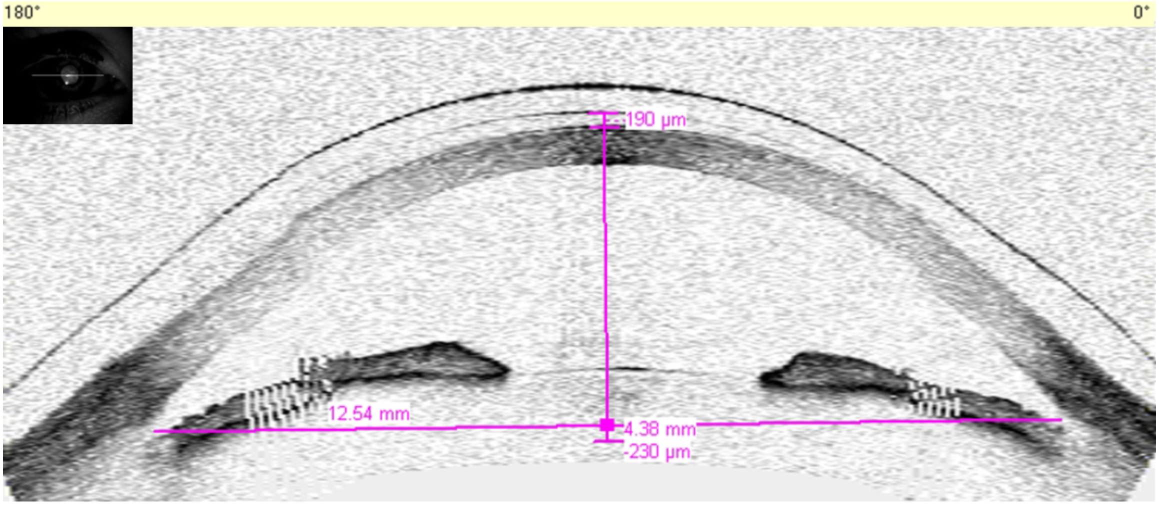

Figure 3-3: Measurement of the tear layer thickness

All measurements were taken in the vertical center of the pupil, adjusted with the help of the live video image, depicted in the upper left corner of figure 3-3. After the scan was taken, the ‘chamber tool’ was applied (in pink color in figure 3-3). The horizontal line of the chamber tool had to be placed in the chamber angles as if the width of the anterior chamber should be measured. The tool automatically halves this width and a perpendicular line allows the measurement of either the corneal thickness, what it was designed for, or, what was used in this study, the tear layer thickness. This had to be done manually because the software can not recognize the different layers as it was designed to measure the corneal thickness that is expected within normal limits.

3.4 Scleral Lens Design

As described previously, the study was executed with the lenses of Appenzeller Kontaktlinsen AG, Switzerland. Their scleral lens design and the fitting recommendations for their lens i-Matrix was illustrated in chapter 2.2.4. The fitting of the lenses within the study followed these instructions.

The lenses of Appenzeller Kontaktlinsen AG are recognized by the requirements of the Medicinal Devices Act whereby an approval of the ethics committee is not required.

All lenses were produced of the material Optimum Extreme (Dk 125) with the same central thickness of 0.35 mm (+/- 0.015 mm manufacturing tolerance) and in plano (+/- 0.0125 dpt manufacturing tolerance) in order to create equal conditions.

3.5 Procedure

While Fonn (1999; 2004) found an affection of the contralateral eye by a swelling response of the lens-wearing eye, the subjects of this study only wore one lens at once and all on the right eye. Before wearing the second lens with another tear layer thickness underneath, there was a required wash-out time of at least one week.

In order to evaluate the data regarding the findings of Fonn (1999; 2004) all measurements of the pachymetry were taken on both eyes (OU).

Pre-fitting

Subject to the condition that every patient had to wear two different lenses with determined tear layer thicknesses before the final relevant data could be evaluated, the subjects had to pass the pre-fitting. Using the provided fitting set, the ideal diameter and base curve were chosen. If the evaluation with fluoresce-in at the slit lamp did not look as expected (see conditions in chapter 2.2.4), modifications were made in cooperation with the manufacturer. After having measured the initial vaulting of the trial lens, Appenzeller Kontaktlinsen calculated the deviation of the base curve in order to achieve a vaulting of one lens of 200 µm and 600 µm of the other lens.

Having received the individual lenses, the subjects had to apply these before the actual wearing day, in order to control the initial vaulting and the limbal and peripheral fit. Only if all requirements were fulfilled, the lens could be used in the study.

Sequence of the Measurements

1) Baseline pachymetry “day before”

   On the day before the lens wearing day at 4 p.m., a measurement of the pachymetry was done OU using the Zeiss Visante OCT and the Oculus Pentacam.

2) Baseline pachymetry OU “morning”

   On the lens wearing day at 8 a.m., prior to any lens wear, a pachymetry was done OU using the Zeiss Visante OCT and the Oculus Pentacam.

3) Application of scleral lens

   One of the scleral lenses determined in the pre-fitting was applied at 8 a.m. in random order, filled with preservative-free saline (Oté Ampullen) and without fluorescein.

4) Measurement of the central clearance

   The initial vaulting was measured using the Visante OCT as described in chapter 3.3.

5) Scleral lens wear for 8 hours

   The lens was worn under open-eye conditions from 8 a.m. to 4 p.m.

6) Re-measurement of the central clearance

   After 8 hours of scleral lens wear, the central clearance of the lens was re-measured using the Visante OCT as before.

7) Removal of the scleral lens

   The scleral lens was removed.

8) Endpoint pachymetry OU “post 8 hours”

   After 8 hours of open eye scleral lens wear, at 4 p.m., a pachymetry was done OU using the Zeiss, Visante OCT and the Oculus Pentacam.

After at least one week of wash-out time, the sequence started again with the second lens.

All measurement results were documented in a spreadsheet (see appendix A 1).

3.6 Statistical Methods

The measured values were documented with the software “Microsoft Office Excel for Windows 2007 (12.0.6718.5000) SP3 MSO” and statistically evaluated with “SPSS Version 11, IBM” and “Minitab 16.2.0”

Testing for statistical significance, a decision concerning the research hypothesis could be brought about. For this reason, the author established the following null hypothesis which is in competition to the research hypothesis:

Null hypothesis H_0: There is no difference in corneal edema due to tear layer thickness during scleral lens wear.

Alternative hypothesis H_1: There is a difference in corneal edema due to tear layer thickness during scleral lens wear.

The acceptable level of statistical significance was defined on 5%. This level $\alpha = 0.05$ bounds the probability of a type I error, which is the rejection of a true H_0 in favor of H_1. A type II error is committed if a false null hypothesis is not rejected. (Bortz, 2008)

4 Statistics

All of the ten subjects succeeded to finish the two wearing sessions completely. In order to be allowed to apply tests for statistical significance like the applied t-test and ANOVA (analysis of variance) (Kuckartz, 2010), the normal distribution of the data collection was tested.

4.1 Normal Distribution Testing

Within every time category ("day before", "morning" and "post 8 hours") and apical clearance group (200 µm, 600 µm) the pachymetry values are not significantly different from normal on a 95% confidence level, because all p-values are larger than 0.05 (see appendix A 2).

4.2 Adequate Number of Measurements

Questioning how many measurements would be adequate in order to get reliable data in the study, the data of a independent measurement series was verified. For this purpose, one subject was measured without any influence of a lens twice (morning/evening) a Monday, twice (morning/evening) a Friday, each 12 times OD and OS.

In the Pentacam measurement, the mean pachymetry in the center of the pupil (COP) was 549.32 µm with a standard deviation of 5.13 µm, which does, with a confidence level of 95%, not go above 6.39 µm. Measured with the Visante OCT, the mean central pachymetry is 539.16 µm with a maximum standard deviation (on the 95% confidence level) of 5.26 µm.

This corresponds to deviations of 1.16% (Pentacam) and 0.98% (Visante) which is far below the physiological and thus the clinical relevant edema. Due to this observation, it is adequate to measure the pachymetry of each subject at each defined time only once.

The deviations, measuring the apical clearance with the Visante OCT are larger, one measurement to the next, because of the deviant initial application purpose of the device. For that reason, the measurements of the apical clearance are repeated three times.

4.3 Two Groups of Apical Clearance

Hereinafter, the amounts of apical clearance are divided into two groups as described in the methods. There is one group including all amounts of vaulting of around 200 µm and one bracketing those around 600 µm.

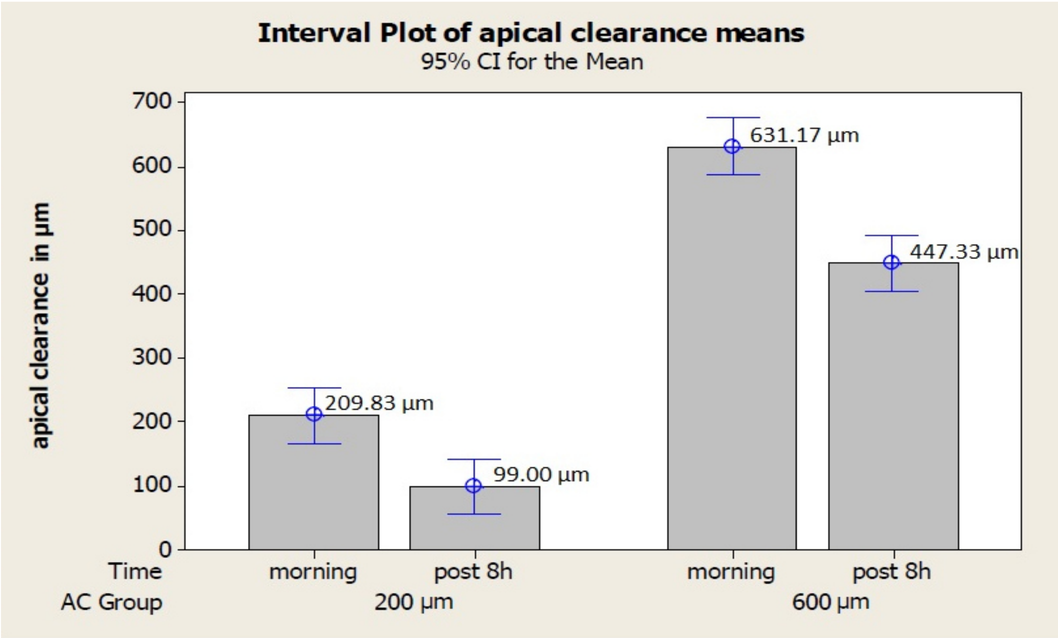

Figure 4-1: Apical clearance groups

The figure shows the two groups of apical clearance, initially after applying the lens ("morning") and after 8 hours ("post 8 h"). The numbers above the columns represent the mean values of lenses fitted in this category, the whiskers illustrate the 95% confidence interval.

Figure 4-1 shows all amounts of apical clearance dependent on the group, each in the morning and after 8 hours scleral lens wear. Due to the pre-described settling of the scleral lens, the second value is lower. Obviously, the fitting was successful in regard of keeping the groups strictly separated. This allows us speaking of two independent groups hereinafter.

4.4 Statistical Analysis

For the statistical approach of answering the research hypothesis, the change in corneal thickness, difference "post 8 hours" - "morning" and "post 8 hours" - "day before" dependent on the group of apical clearance (200 µm vs. 600 µm) has to be considered. By this means, the individual level of pachymetry is neglected which enables a more neutral approach.

This refers to chapters 4.4.1 to 4.4.3. Chapter 4.4.4 applies the raw, measured pachymetry values.

4.4.1 Normal Distribution Testing

The difference values underwent a normality test which did not show any difference from normal (see figure 4-2 showing values of Pentacam and Visante OCT for the difference "post 8 hours" - "morning" and figure 4-3 for the difference "post 8 hours" - "day before") and can thus be used for statistical analysis.

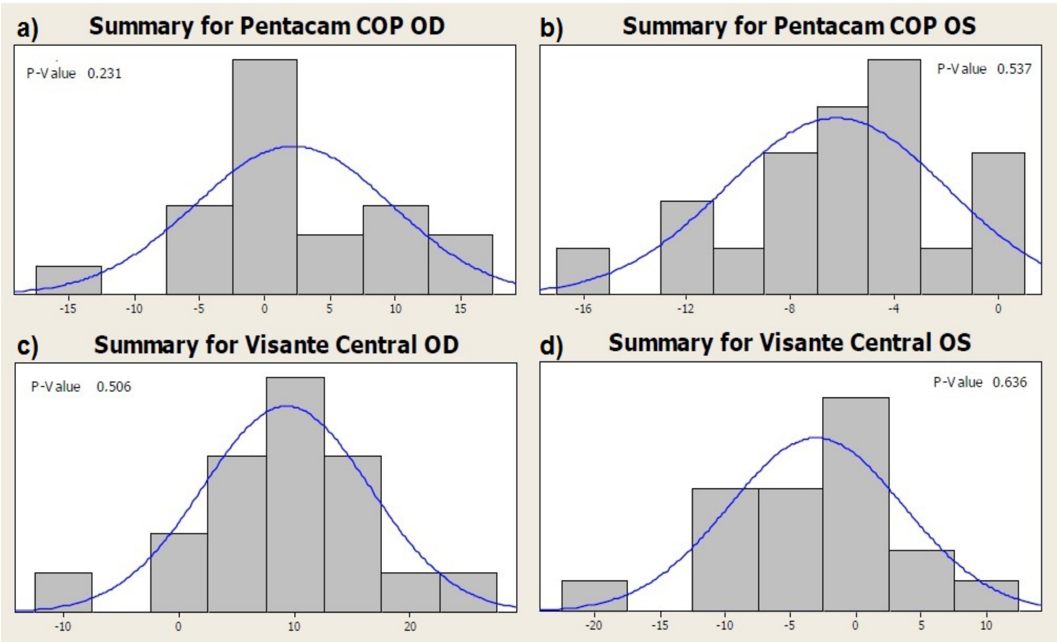

Figure 4-2: Test of normal distribution of pachymetry differences ("post 8 hours" - "morning")[2]

a) p-value Pentacam COP OD: 0.231; b) p-value Pentacam COP OS: 0.537; c) p-value Visante Central OD: 0.506; d) p-value Visante Central OS: 0.636. Each difference sequence itself is not significantly different from normal on a level of significance of 5%.

[2] Histograms with tests for normal distribution show the difference of pachymetry in µm on the x-axis and the number of subjects (n) on the y-axis. The statistical software does not allow printing this within the diagram. This applies for figures 4-2 and 4-3.

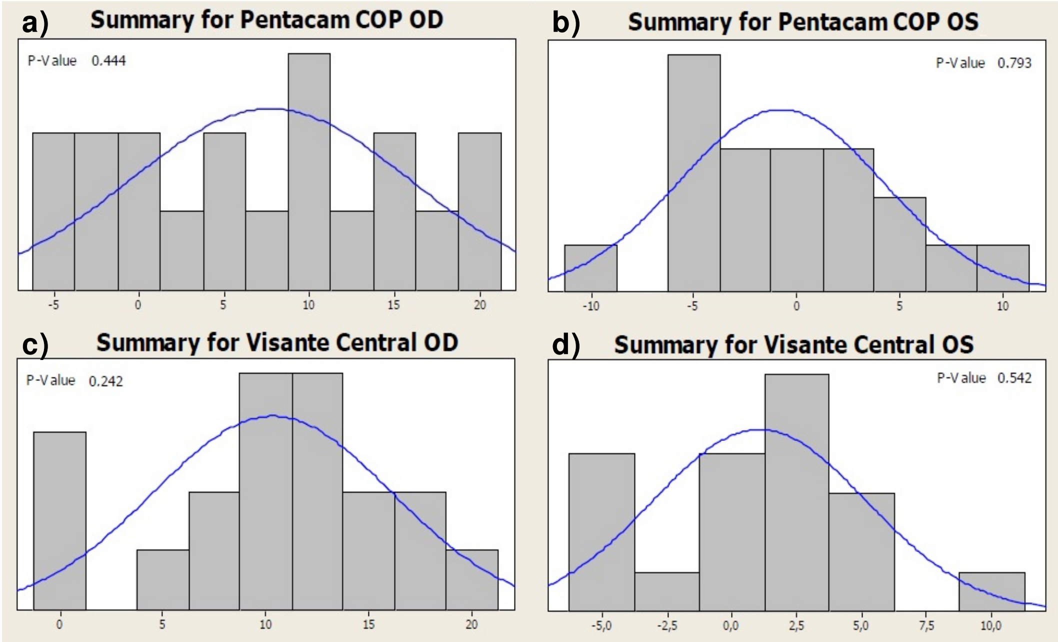

Figure 4-3: Test of normal distribution of pachymetry differences (“post 8 hours” - “day before”)

a) p-value Pentacam COP OD: 0.444; b) p-value Pentacam COP OS: 0.793; c) p-value Visante Central OD: 0.242; d) p-value Visante Central OS: 0.542. Each difference sequence itself is not significantly different from normal on a level of significance of 5%.

4.4.2 T-Test of Corneal Thickness Change - Pentacam

A two-sample t-test for the change in corneal thickness, difference “post 8 hours” - “morning” and “post 8 hours” - “day before”, was performed, dependent on the group of apical clearance (200 µm vs. 600 µm).

Testing the difference “post 8 hours” - “morning”:

Illustrated in figure 4-4, the group of 200 µm apical clearance has a mean change in pachymetry of the lens-wearing right eye (OD) of 2.22 µm with a standard deviation of 6.55 µm, the group of 600 µm has a mean of 2.10 µm with a standard deviation of 8.62 µm. The p-value of 0.973 shows that there is no statistically significant difference in pachymetry change on the lens-wearing eye whether a lens with an apical clearance of 200 µm or 600 µm is worn, ceteris paribus. Thus, the null hypothesis can be confirmed.

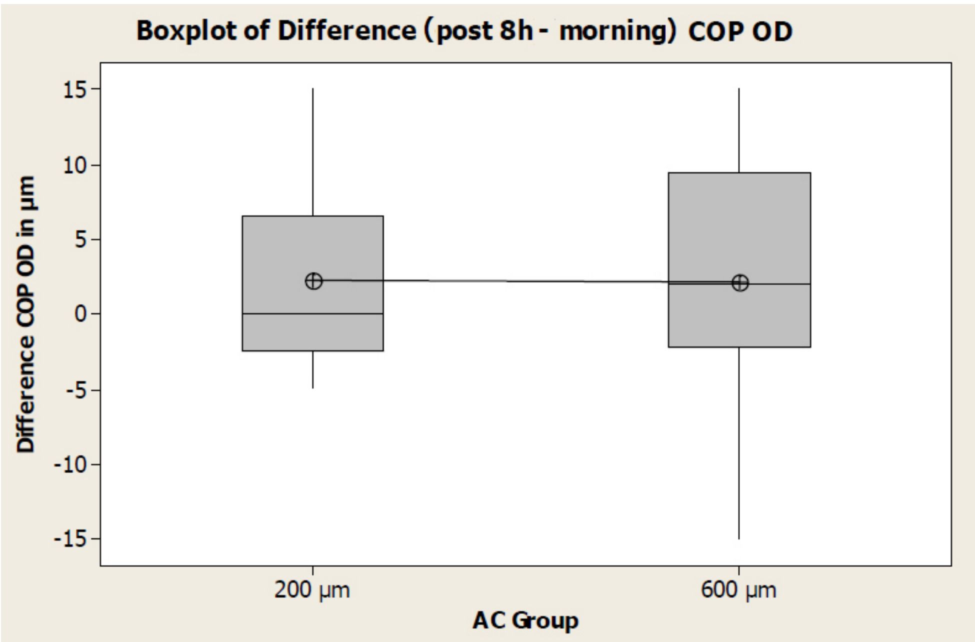

Figure 4-4: Corneal thickness change OD dependent on apical clearance; Pentacam (“post 8 hours” - “morning”)

The figure shows the corneal thickness change “post 8 hours” - “morning” of the right (lens-wearing) eye dependent on the apical clearance groups, measured with the Pentacam, COP values shown. The mean of the 200 µm group is 2.22 µm change, the mean of the 600 µm group is 2.10 µm.

Figure 4-5 illustrates the corneal thickness change of the non-lens-wearing left eye. The mean differences are negative. This can be justified in the overnight edema that is included in the measurement of the morning. The group of 200 µm apical clearance results in a mean change in pachymetry of the non-lens-wearing left eye (OS) of -5.89 µm, with a standard deviation of 5.11 µm. The group of 600 µm has a mean of -6.80 µm with a standard deviation of 3.74 µm.

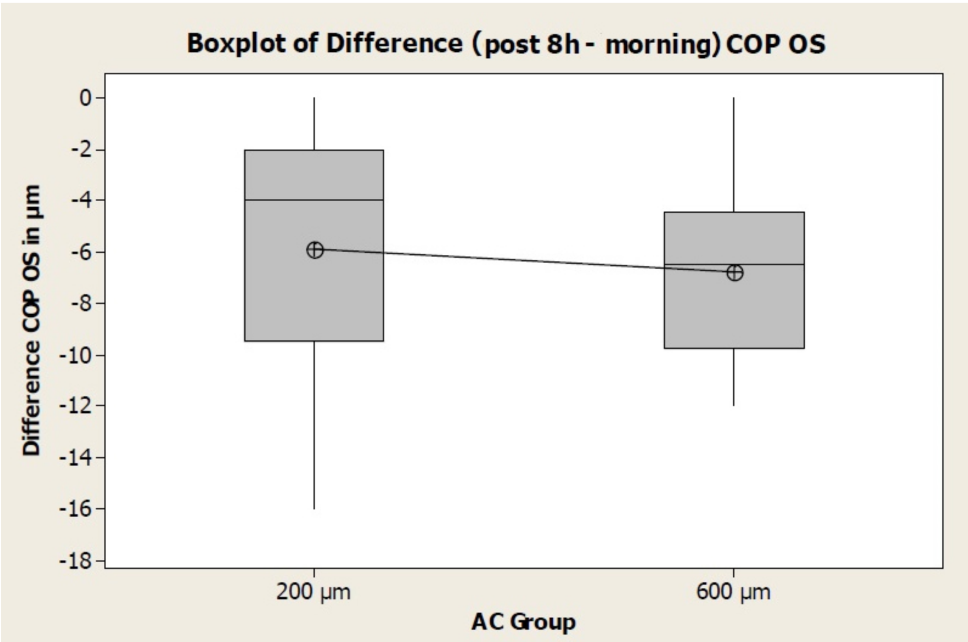

Figure 4-5: Corneal thickness change OS dependent on apical clearance; Pentacam (“post 8 hours” - “morning”)

The figure shows the corneal thickness change “post 8 hours” - “morning” of the left (non-lens-wearing) eye dependent on the apical clearance groups, measured with the Pentacam, COP values shown. The mean change of the 200 µm group is -5.89 µm, the mean of the 600 µm group is -6.80 µm.

The p-value of 0.667 shows that there is no statistically significant difference in pachymetry change on the non-lens-wearing eye whether a lens with an apical clearance of 200 µm or 600 µm is worn on the contralateral eye, ceteris paribus. Thus, the null hypothesis also can be confirmed for the left eye.

Testing the difference OD versus OS, a statistically significant difference (p = 0.000) shows up (see figure 4-6):

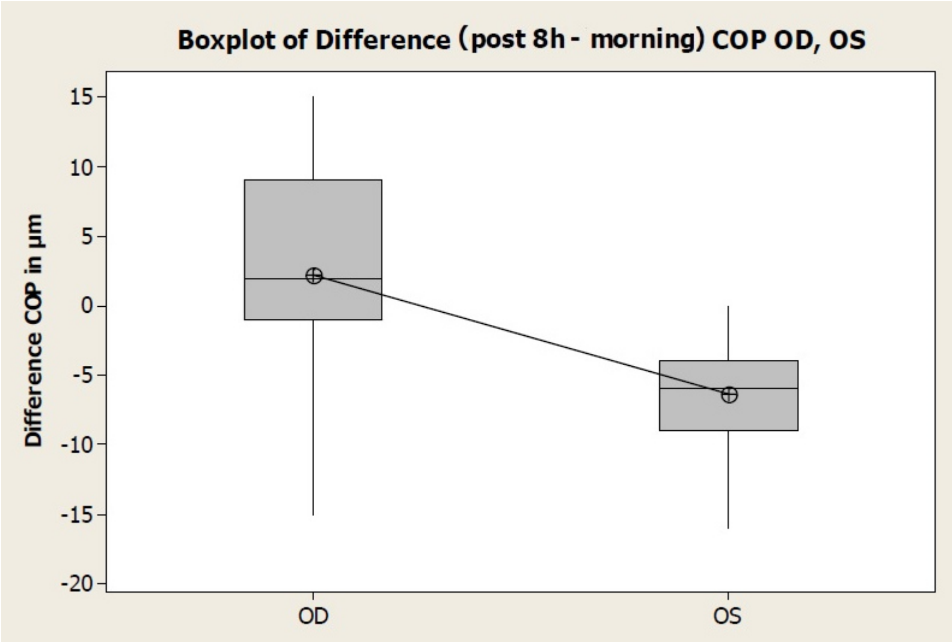

Figure 4-6: Corneal thickness change, all apical clearance groups, OD vs. OS; Pentacam (“post 8 hours” - “morning”)

The figure shows the corneal thickness change “post 8 hours” - “morning” dependent on the eye (OD vs. OS) including all apical clearance groups, measured with the Pentacam, COP values shown. The mean change OD (lens-wearing) is 2.16 µm, the mean change OS (non-lens-wearing) is -6.37 µm.

The mean difference between corneal thickness change OD and OS is 8.53 µm. This evaluation does not show if there is any or no influence of wearing a lens to the contralateral eye but at least, there is shown, that there is significantly less influence on the non-lens-wearing eye than on the lens-wearing eye.

Testing the difference "post 8 hours" - "day before":

In order to exclude the physiological overnight swelling, the change in pachymetry post 8 hours of scleral lens wear was additionally analyzed compared to the day before.

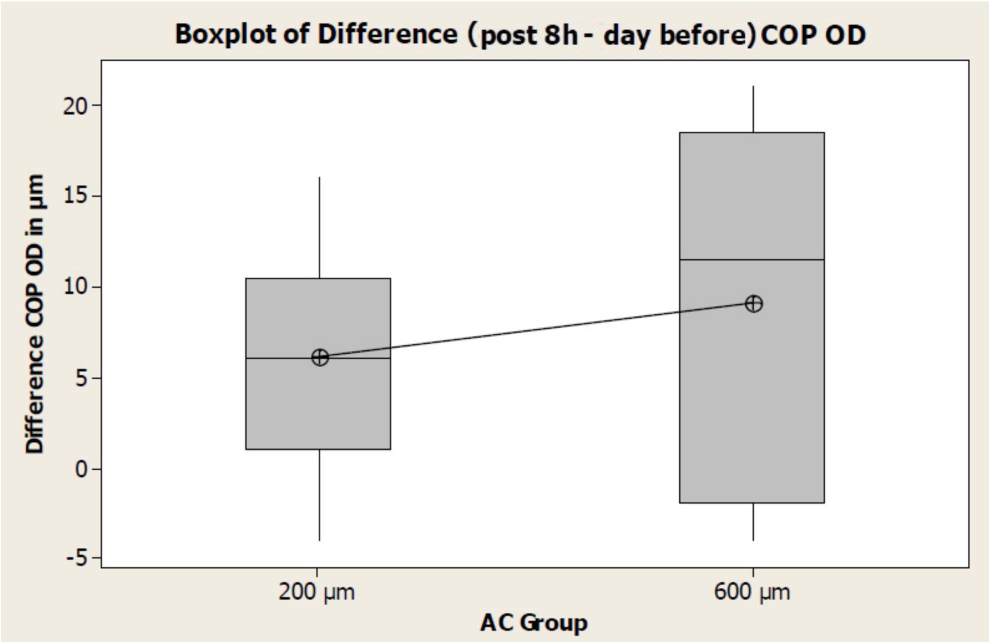

Figure 4-7: Corneal thickness change OD dependent on apical clearance; Pentacam ("post 8 hours" - "day before")

The figure shows the corneal thickness change "post 8 hours" - "day before" of the right (lens-wearing) eye dependent on the apical clearance groups, measured with the Pentacam, COP values shown. The mean of the 200 µm group is 6.11 µm change, the mean of the 600 µm group is 9.10 µm.

Figure 4-7 illustrates the group of 200 µm apical clearance compared to 600 µm. The mean change in pachymetry of the lens-wearing right eye is 6.11 µm with a standard deviation of 6.09 µm, the group of 600 µm has a mean of 9.10 µm with a standard deviation of 9.90 µm. The p-value of 0.436 shows that there is no statistically significant difference in pachymetry change on the lens-wearing eye whether a lens with an apical clearance of 200 µm or 600 µm is worn, ceteris paribus. Thus, the null hypothesis can be confirmed.

Figure 4-8 illustrates the corneal thickness change of the non-lens-wearing left eye. The group of 200 µm apical clearance results in a mean change in pachymetry of the non-lens-wearing left eye of 0.67 µm, with a standard deviation of 6.12 µm. The group of 600 µm has a mean of -2.10 µm with a standard deviation of 3.35 µm.

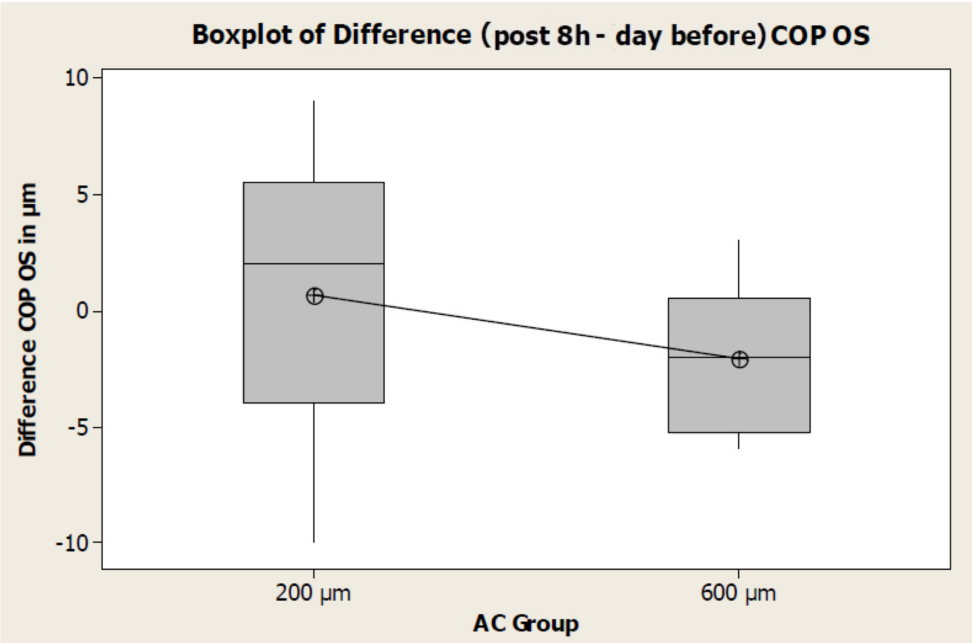

Figure 4-8: Corneal thickness change OS dependent on apical clearance; Pentacam (“post 8 hours” - “day before”)

The figure shows the corneal thickness change “post 8 hours” - “day before” of the left (non-lens-wearing) eye dependent on the apical clearance groups, measured with the Pentacam, COP values shown. The mean of the 200 µm group is 0.67 µm change, the mean of the 600 µm group is -2.10 µm.

The p-value of 0.252 shows that there is no statistically significant difference in pachymetry change on the non-lens-wearing eye whether a lens with an apical clearance of 200 µm or 600 µm is worn on the contralateral eye, ceteris paribus. Thus, the null hypothesis also can be confirmed for the left eye.

Testing the difference OD versus OS, again a statistically significant difference (p = 0.001) shows up (see figure 4-9):

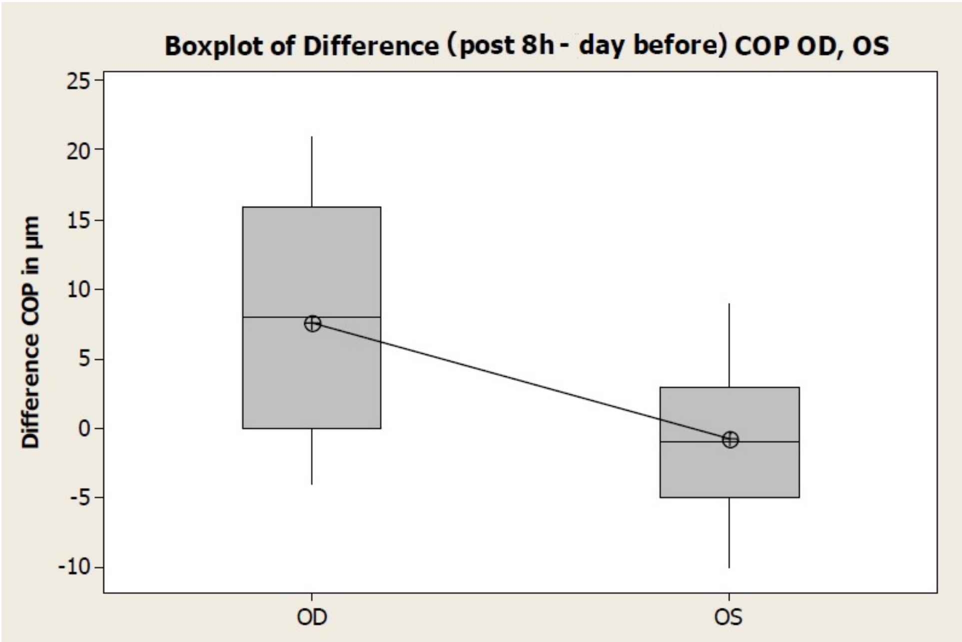

Figure 4-9: Corneal thickness change, all apical clearance groups, OD vs. OS; Pentacam ("post 8 hours" - "day before")

The figure shows the corneal thickness change "post 8 hours" - "day before" dependent on the eye (OD vs. OS) including all apical clearance groups, measured with the Pentacam, COP values shown. The mean change OD (lens-wearing) is 7.68 µm, the mean change OS (non-lens-wearing) is -0.79 µm.

The mean difference between corneal thickness change OD and OS is 8.47 µm. This evaluation does not show if there is any or no influence of wearing a lens to the contralateral eye but at least, there is shown, that there is significantly less influence on the non-lens-wearing eye than on the lens-wearing eye.

4.4.3 T-Test of Corneal Thickness Change – Visante OCT

The findings of the Pentacam above can be confirmed with the analysis of the data measured with the Visante OCT (see figures 4-10 to 4-15).

Testing the difference "post 8 hours" - "morning":

Figure 4-10 shows, that on a level of p = 0.933, the change in corneal thickness is statistically equal, no matter if a lens with an apical clearance of 200 µm or 600 µm is worn. Figure 4-11 shows the same on a level of p = 0.712 for the left eye, which did not wear a lens. Thus, the null hypothesis also can be confirmed for the measurements of the Visante OCT.

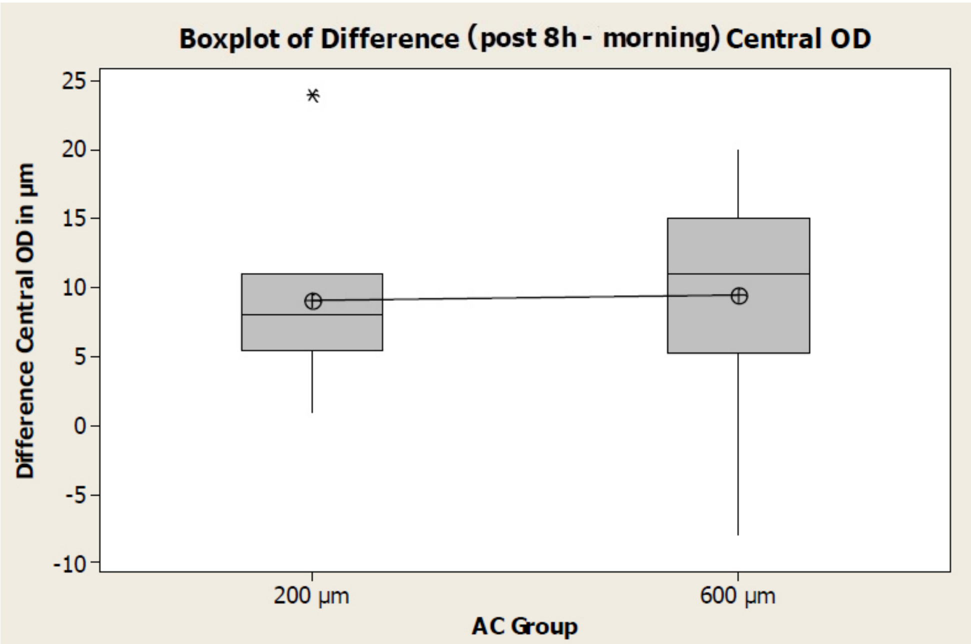

Figure 4-10: Corneal thickness change OD dependent on apical clearance; Visante OCT ("post 8 hours" - "morning")

The figure shows the corneal thickness change "post 8 hours" - "morning" of the right (lens-wearing) eye dependent on the apical clearance groups, measured with the Visante OCT, Central values shown. The mean of the 200 µm group is 9.11 µm change, the mean of the 600 µm group is 9.40 µm.

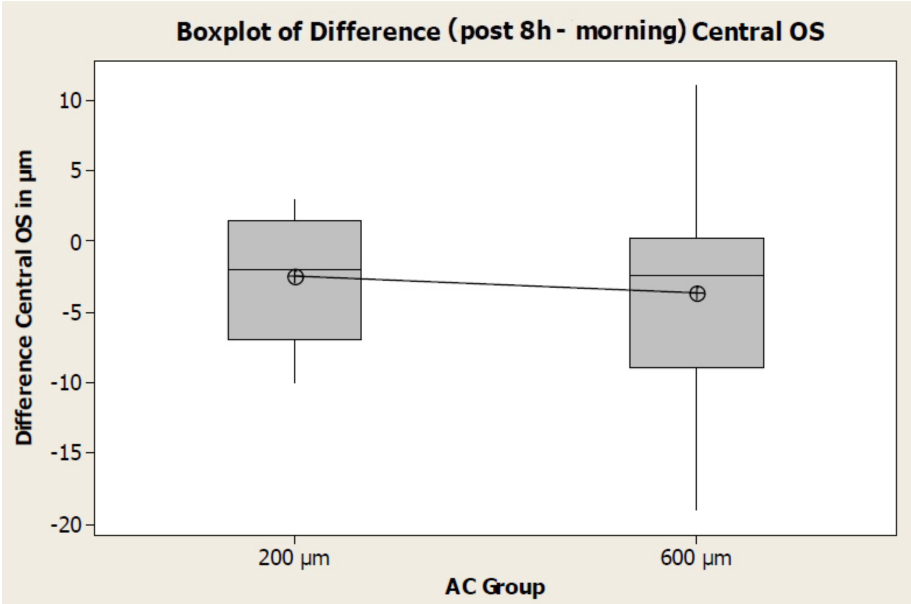

Figure 4-11: Corneal thickness change OS dependent on apical clearance; Visante OCT ("post 8 hours" - "morning")

The figure shows the corneal thickness change "post 8 hours" - "morning" of the left (non-lens-wearing) eye dependent on the apical clearance groups, measured with the Visante OCT, Central values shown. The mean of the 200 µm group is -2.44 µm change, the mean of the 600 µm group is -3.60 µm.

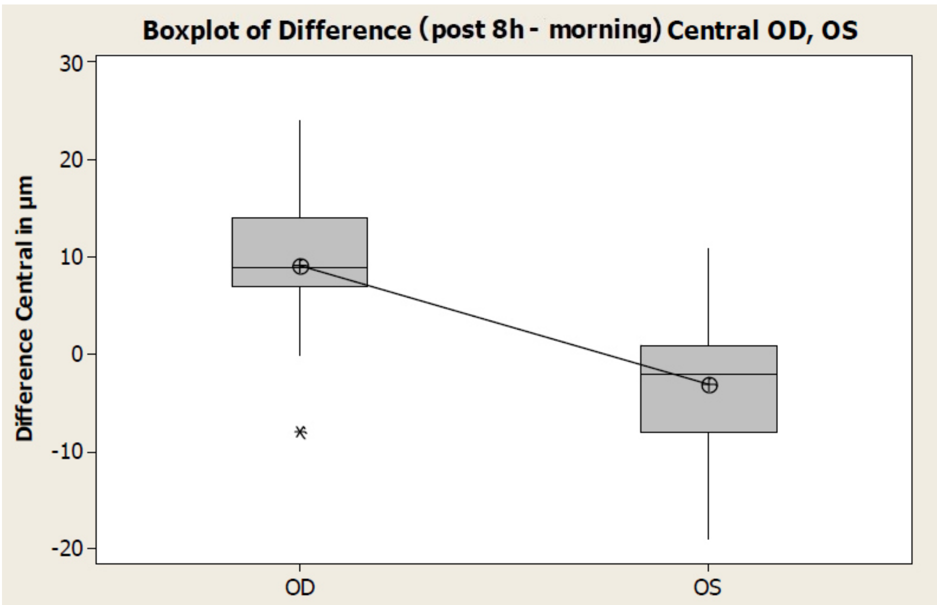

Figure 4-12: Corneal thickness change, all apical clearance groups, OD vs. OS; Visante OCT ("post 8 hours" - "morning")

The figure shows the corneal thickness change "post 8 hours" - "morning" dependent on the eye (OD vs. OS) including all apical clearance groups, measured with the Visante OCT, Central values shown. The mean change OD (lens-wearing) is 9.26 µm, the mean change OS (non-lens-wearing) is -3.05 µm.

Figure 4-12 shows, similar to the measurements of the Pentacam illustrated in figure 4-6, a statistically significant difference OD versus OS ($p = 0.000$). The mean difference between corneal thickness change OD and OS is 12.32 µm. Additionally, this kind of measurement shows that there is significantly less influence on the non-lens-wearing eye than on the lens-wearing eye.

Testing the difference “post 8 hours” - “day before”:

Figure 4-13 shows, that on a level of $p = 0.352$, the change in corneal thickness is statistically equal, no matter if a lens with an apical clearance of 200 µm or 600 µm is worn. Figure 4-14 shows the same on a level of $p = 0.873$ for the left eye, which did not wear a lens. Thus, the null hypothesis also can be confirmed for the measurements of the Visante OCT.

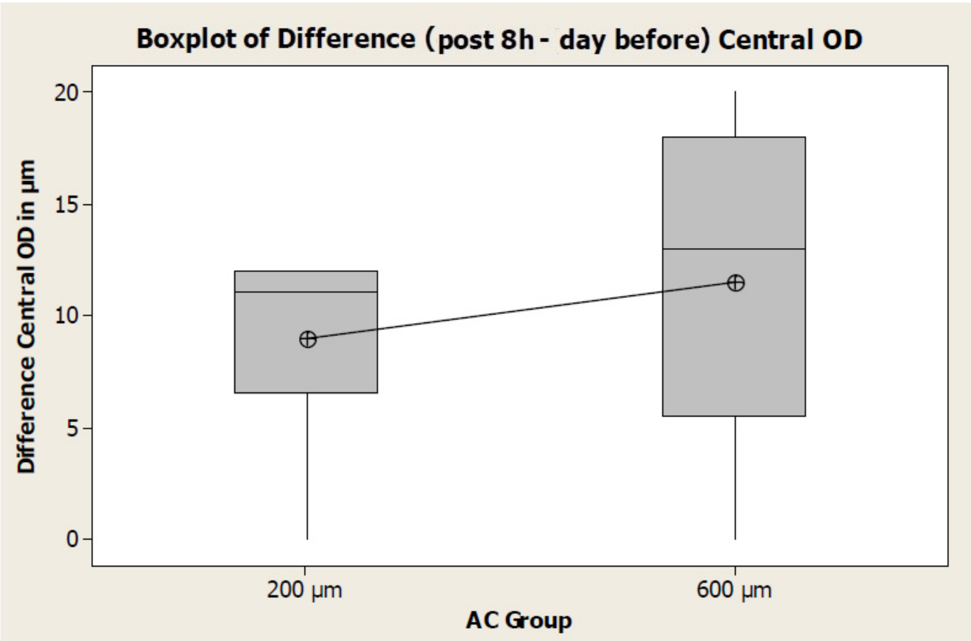

Figure 4-13: Corneal thickness change OD dependent on apical clearance; Visante OCT (“post 8 hours” - “day before”)

The figure shows the corneal thickness change “post 8 hours” - “day before” of the right (lens-wearing) eye dependent on the apical clearance groups, measured with the Visante OCT, Central values shown. The mean of the 200 µm group is 9.00 µm change, the mean of the 600 µm group is 11.50 µm.

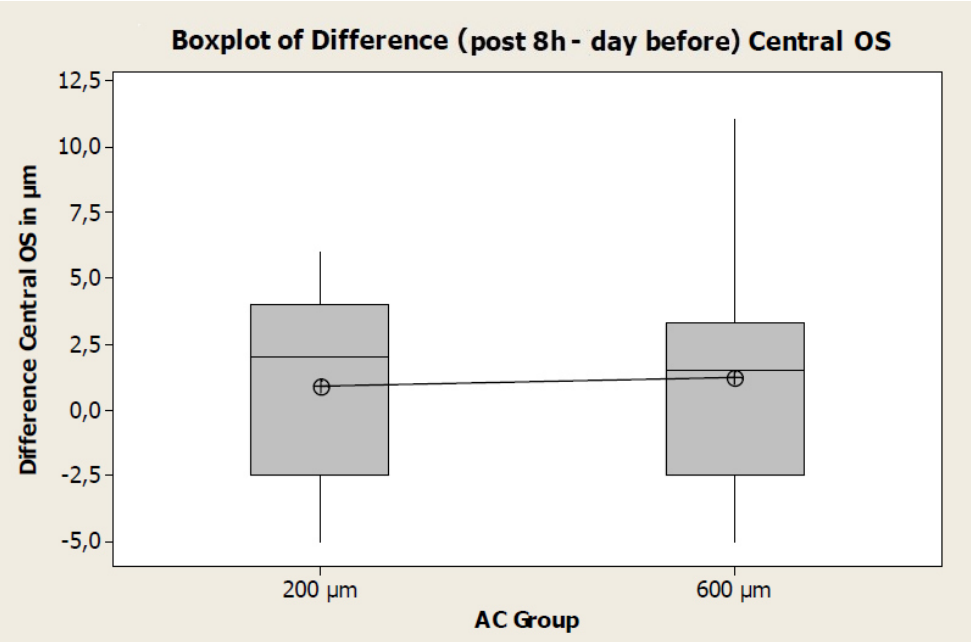

Figure 4-14: Corneal thickness change OS dependent on apical clearance; Visante OCT (“post 8 hours” - “day before”)

The figure shows the corneal thickness change “post 8 hours” - “day before” of the left (non-lens-wearing) eye dependent on the apical clearance groups, measured with the Visante OCT, Central values shown. The mean of the 200 µm group is 0.89 µm change, the mean of the 600 µm group is 1.20 µm.

In this study, all in all, the amount of apical clearance (200 µm compared to 600 µm) does not show a statistically significant influence on central corneal thickness change. The null hypothesis can be confirmed for all central measurements (Pentacam and Visante OCT).

Figure 4-15 shows, similar to the measurements of the Pentacam illustrated in Figure 4-9, a statistically significant difference OD versus OS ($p = 0.000$). The mean difference between corneal thickness change OD and OS is 9.26 µm. Additionally, this kind of measurement shows that there is significantly less influence on the non-lens-wearing eye than on the lens-wearing eye.

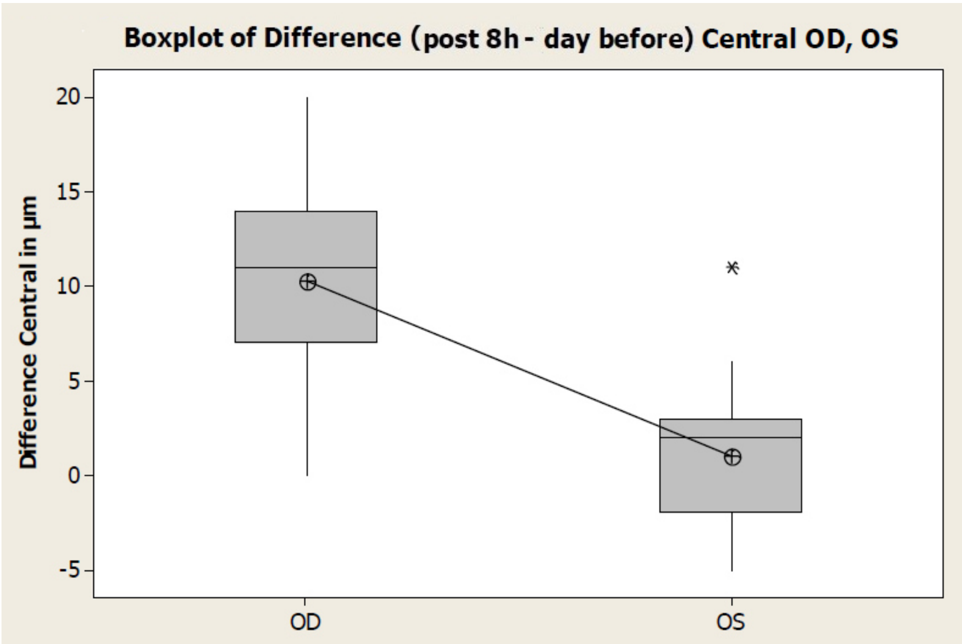

Figure 4-15: Corneal thickness change, all apical clearance groups, OD vs. OS; Visante OCT ("post 8 hours" – "day before")

The figure shows the corneal thickness change "post 8 hours" – "day before" dependent on the eye (OD vs. OS) including all apical clearance groups, measured with the Visante OCT, Central values shown. The mean change OD (lens-wearing) is 10.32 µm, the mean change OS (non-lens-wearing) is 1.05 µm.

In chapter 5, the clinical relevance of the differences between corneal thickness change OD and OS will be analyzed.

4.4.4 Three-way ANOVA of Corneal Thickness Data

The three-way ANOVA model evaluated the same data including all peripheral aspects. It evaluated corneal thickness based on two levels of apical clearance, 3 times of measurement, and the eye with or without the lens. “Group” describes the amount of apical clearance and it was found that there are no statistical differences in corneal thickness based on the type of lens that was used, visible in all p-values being above 0.05, see table 4-1. This was true for the main effect and any interaction involving group. The null-hypothesis can be confirmed.

	Pentacam COP	Pentacam TP	Visante Central	Visante Superior	Visante Inferior	Visante Temporal	Visante Nasal
Source	p	p	p	p	p	p	p
Group	0.050	0.044	0.244	0.439	0.606	0.354	0.783
Time	0.000	0.001	0.000	0.641	0.002	0.004	0.010
Eye	0.001	0.028	0.000	0.042	0.235	0.757	0.000
Group * Time	0.943	0.909	0.814	0.786	0.997	0.361	0.892
Group * Eye	0.190	0.193	0.529	0.397	0.116	0.855	0.780
Time * Eye	0.001	0.001	0.000	0.026	0.023	0.005	0.031
Group * Time * Eye	0.436	0.271	0.926	0.596	0.918	0.538	0.758
Effect Size OD/OS at Post	0.74	0.66	1.33	0.65	0.43	0.57	0.71

Table 4-1: Results of three-way ANOVA

It appeared, that time, eye, and time by eye interaction were frequently significant as p-values are below 0.05. This means, that there are statistically significant results concerning the lens-wearing and non-lens-wearing eye as well as for the aspect time, meaning that pachymetry after 8 hours of scleral lens wear was from a statistical viewpoint significantly higher than on the 2 other times measured (“day before” and “morning”). This is demonstrated in Figure 4-16 for Visante Central:

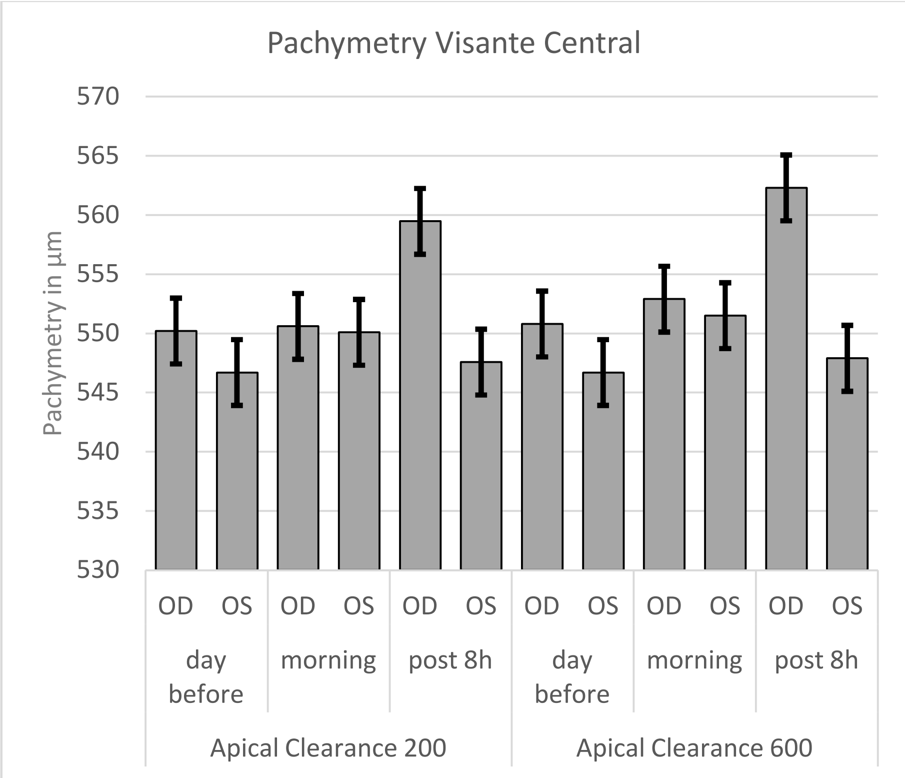

Figure 4-16: Overview Pachymetry Visante Central

This figure illustrates that the eye with the lens has a thicker cornea after wearing either lens for 8 hours.

In this graph it is shown that the eye with the lens has a significantly thicker cornea after wearing either lens for 8 hours.

One can assume due to the height of the columns that there is no difference between the lenses, which was proved statistically in chapter 4.4.2 and 4.4.3 and which is resulting of the ANOVA (see table 4-1).

The patterns are similar in the other measurements. This also results in a confirmation of the null hypothesis.

5 Clinical Relevance of Corneal Thickness Change

There was no statistical influence of the amount of apical clearance upon the corneal thickness found, but a general effect on the pachymetry due to the fact that a scleral lens is worn. For that reason it has to be considered if this resulting edema is clinically relevant, including exclusively the values which are not influenced by the overnight swelling. The corneal thickness change (“post 8 hours” - “day before”) is converted in percent, relative to the individual pachymetry of each subject.

In order to assess the clinical relevance of the corneal thickness change, the assumption is applied that the everyday occurring no-lens-wearing overnight edema of the widely accepted ~4% (Cox, 1990) does not harm the cornea.

5.1 Individual Overnight Swelling in this Study

Looking at the individual overnight swelling in this study, the mean value of the central measurements is 0.77% with a standard deviation of 1.69%. The maximum measured with the Pentacam is 3.19% and 3.73% measured with the Visante OCT. It should be added here, that the measurement in the morning was always taken at 8 o’clock, regardless of the time the individual subject awoke. Estimating a time of being awake of at least 1 hour in every subject, the induced overnight swelling could already have declined to a certain degree. This might be the reason why the measured value is below the widely accepted value of no-lens-wearing overnight corneal swelling of approximately 4% (Cox, 1990).

5.2 Corneal Thickness Change in Percent

The following figures 5-1 and 5-2 give a practical overview of the corneal thickness change by illustrating this in percent. They allow comparing the lens-wearing eye and the non-lens-wearing eye as well as the two groups of apical clearance.

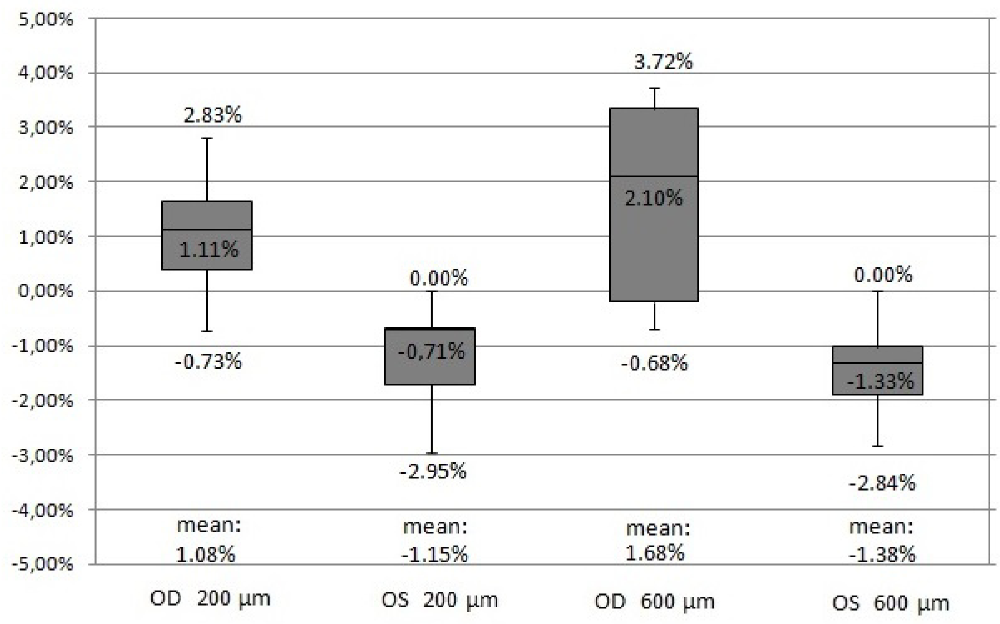

Figure 5-1: Corneal thickness change in percent; Pentacam COP

The figure shows the change in pachymetry "post 8 hours" - "day before" measured with the Pentacam, COP values shown. The whiskers go to the minimum and maximum which is never beyond the 4% mark.

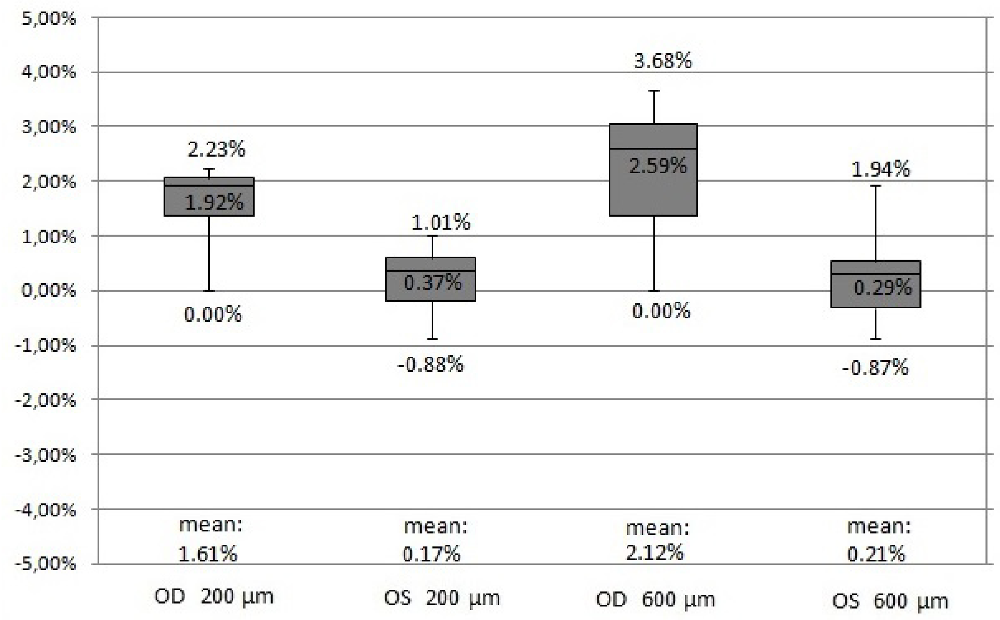

Figure 5-2: Corneal thickness change in percent; Visante Central

The figure shows the change in pachymetry "post 8 hours" - "day before" measured with the Visante OCT, Central values shown. The whiskers go to the minimum and maximum which is never beyond the 4% mark.

The columns show higher amounts of corneal swelling in the lens-wearing eye compared to the non-lens-wearing eye. This is proved by the statistical analysis in chapter 4, illustrated in figures 4-9 and 4-15. The found mean difference (Pentacam) between corneal thickness change OD and OS which is 8.47 µm in absolute numbers, corresponds to 1.52%, based on a mean corneal pachymetry of 558.8 µm (Pentacam) of the subjects in this study. The mean difference of the Visante OCT of 9.26 µm corresponds to 1.69%, based on the measured mean corneal pachymetry of 548.6 µm (Visante). Both of them are below the critical 4%.

Concerning the apical clearance groups, these figures (5-1, 5-2) are not enough to make a statement because the whole range of values of the 200 µm group is similar to the range of values of the 600 µm group while the means and moreover the medians, differ. The statistical analysis presented in chapter 4 shows that there is no statistical significant difference which can be transferred to no clinically relevant difference because of the negligible small differences which lie below the 4% mark. Thus, the null-hypothesis also can be confirmed from a clinical viewpoint.

All in all, none of the corneas swelled more than 4% (maximum 3.72% measured with the Pentacam, 3.68% measured with the Visante OCT). In the author’s opinion, the measured level of edema is clinically reasonable, provided that this level stays constant with several successive days of wearing, that the patient is compliant inserting days off the lens and that the consequences not-wearing a lens are not worse than accepting a certain level of edema.

6 Summary and Discussion

The ideal tear layer thickness underneath scleral lenses is an unknown parameter when fitting this type of lens. This present study practically contributes to gradually solve this issue in terms of corneal edema.

Fundamentals like the anatomy and physiology of the anterior ocular surface, especially of the cornea, and those concerning scleral lenses provide a basis to look into the oxygen supply with scleral lenses.

The null hypothesis that there is no difference in corneal edema due to tear layer thickness during scleral lens wear could not be rejected on the basis of the data collected. There was no statistically significant difference in corneal edema wearing either lens. The non-significant result however is not a final evidence that the null hypothesis is correct because the study is limited in its validity due to the sample size. Although all ten subjects succeeded to finish the two wearing sessions completely, one result of one subject (subject 6, visit #1) had to be excluded because a deviating contact lens material has been used in the study.

This result does not correlate with the theoretical estimates of Michaud (2012), referring to the classic Holden-Mertz and Harvitt-Bonanno criterias, to avoid corneal swelling during lens wear. Similarly there is no correlation with the findings of Compañ (2014) who approached theoretically and clinically to this issue. There were eight subjects included in his trial, wearing the lenses for 3 hours under open-eye conditions. In contrast, in the present study, the same conditions for all subjects were ensured in using lenses in plano, standardized thickness and equal Dk.

Compañ considered steady-state conditions and negated tear exchange beneath the scleral lens but this might be an explanation why few problems with corneal edemas during scleral lens wear occur in practice.

When designing this experiment, there was some difficulty choosing the right focus when measuring the vaulting of the scleral lens. Anterior OCTs are designed for measuring the anterior segment of the eye, not expecting a scleral lens resting on top of the eye. Due to this reason, the automatisms of the devices oftentimes report errors (e.g. unusual corneal thickness because the software recognizes the whole system cornea – tear lens – contact lens as one unit). Altering the focus point when measuring caused a reasonably different value. As it is not known which value is closer to the true vaulting, the focus point was always chosen the same way. Although, compared to the clinical trial of Compañ (2012), it is a non-invasive and objective way of measurement.

The study additionally assessed the resulting edema, irrespective of the apical clearance. The eyes with the lenses had statistically significantly higher edematous responses compared to the non-lens-wearing eyes after wearing the lenses for 8 hours. All induced edemas did not go beyond 4% which was assumed physiologically acceptable. The statement of Fonn (1999; 2004) that there is corneal swelling of the contralateral eye can neither be confirmed nor rejected because the changes in the non-lens-wearing eyes were too small.

Over and above that, these results have to be considered with caution in corneas or eyes varying from the norm. Lenses used in the trial were all manufactured in plano. Higher refractive errors cause thicker contact lenses, either in the center or the optical periphery which can have an influence on the oxygen supply of the cornea. Corneas with a highly irregular surface will always have spots with small and spots with high vaulting. Comprised corneal metabolism might cause higher demand for oxygen as the healthy corneas which were involved in the study. As these lenses are mainly applied in diseased ocular surface conditions, the corneal reaction might be diverging. Nevertheless, there might be conditions which are considered to be more destructive to the cornea than the corneal hypoxia caused by the scleral lens (Smith, 2004). The advantages and disadvantages have to be weighed up in order to find the best solution for the patient.

We recommend conducting further clinical trials with larger samples which maybe can disprove long-established criterias concerning the oxygen demand of the cornea. As this study only intended the subjects wearing the lens for 8 hours on one day, more clinical research is necessary to observe the long-term-consequences of scleral lenses on corneal physiology.

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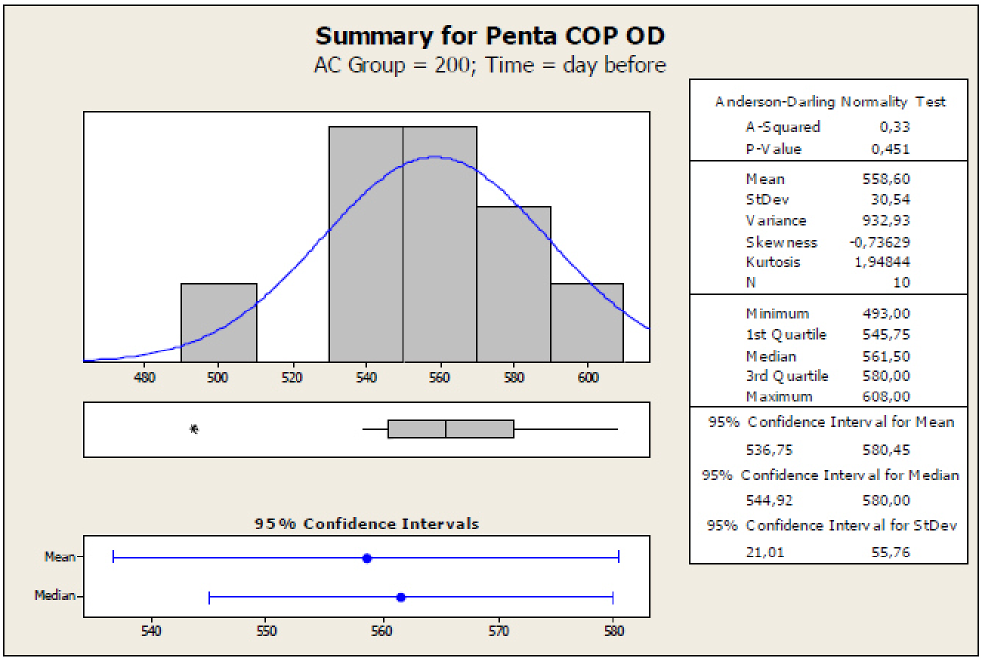
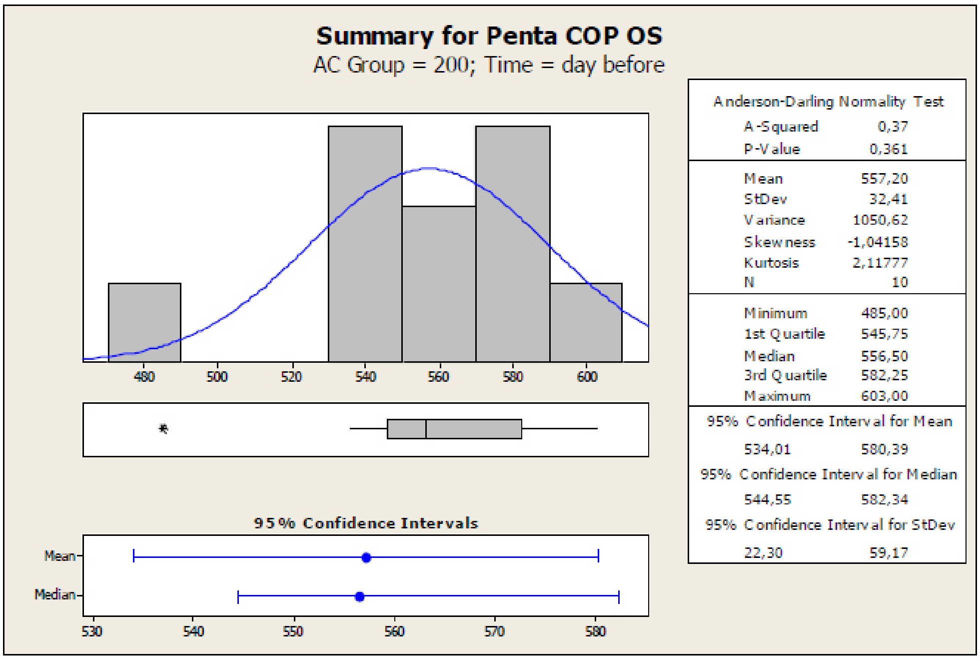
Appendix

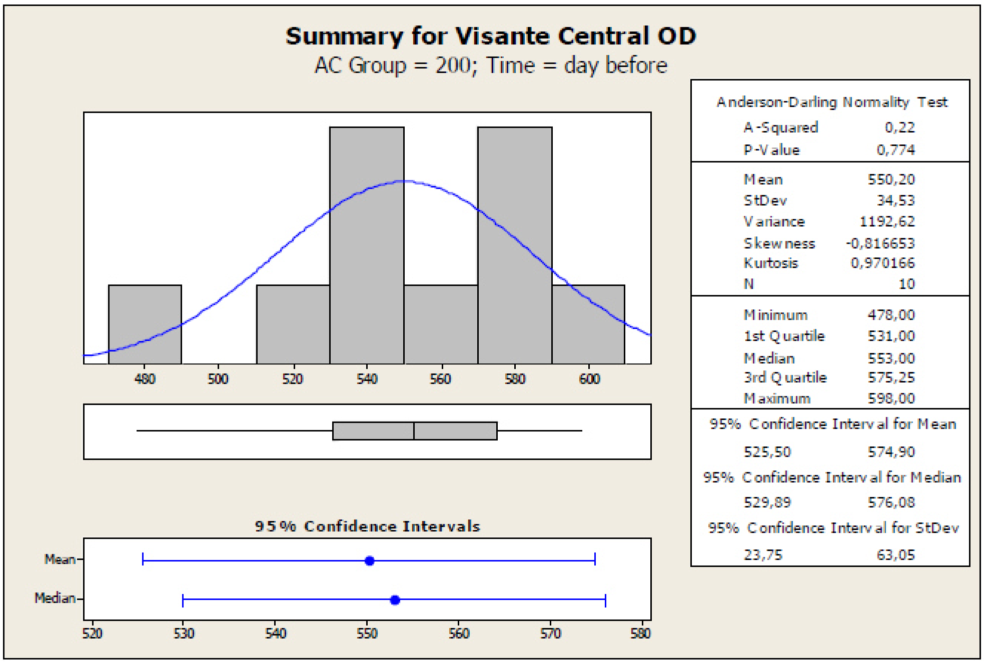
A 1: Spreadsheet

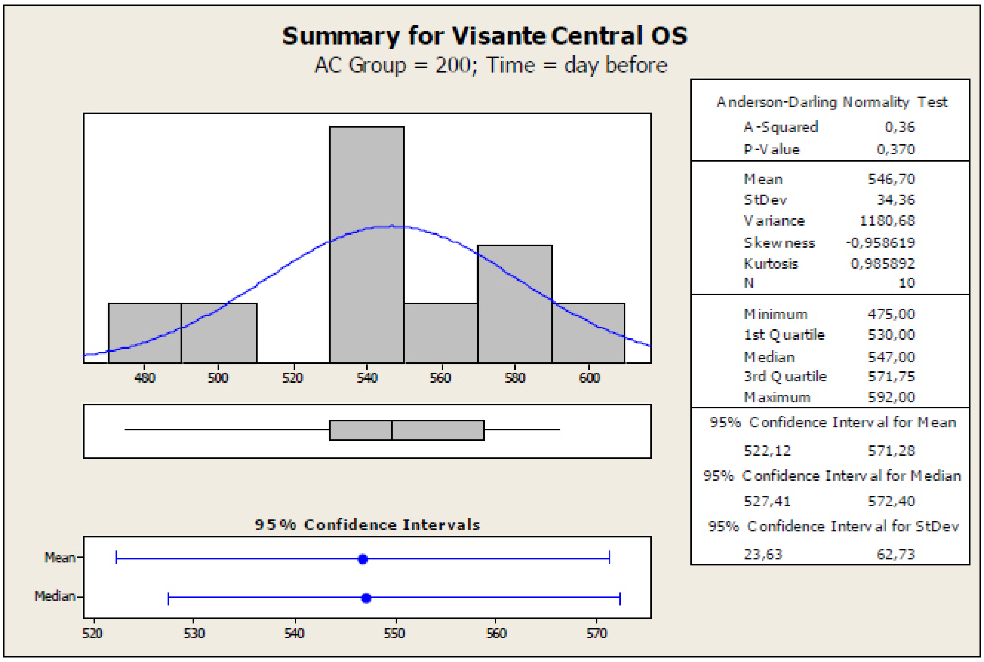
Subject #	Visit #	Date	Time	apical clearance (AC)			Diagnostic len	Base curve	Diameter	Pachymetry Pentacam COP OD	Pachymetry Pentacam COP OS	Pachymetry Pentacam TP OD	Pachymetry Pentacam TP OS	Pachymetry Visante Central OD	Pachymetry Visante Central OS	Pachymetry Visante Superior OD	Pachymetry Visante Superior OS	Pachymetry Visante inferior OD	Pachymetry Visante inferior OS	Pachymetry Visante temporal OD	Pachymetry Visante temporal OS	Pachymetry Visante nasal OD	Pachymetry Visante nasal OS
1	1	12.1.14	day before							587	583	584	580	568	562	715	696	662	648	645	657	680	656
1	1	12.2.14	morning	670	680	-	NAHF5	7,06	16,5	592	590	589	585	577	574	723	722	694	664	648	658	707	704
1	1	12.2.14	post 8h	600	-	-	NAHF5	7,06	16,5	607	578	605	574	586	566	740	690	679	673	680	646	687	671
1	2	12.8.14	day before							580	572	577	568	573	568	733	712	629	654	660	619	676	702
1	2	12.9.14	morning	270	260	-	NAHF6	7,35	16,5	588	585	585	583	573	572	737	721	674	662	663	652	699	696
1	2	12.9.14	post 8h	220	230	-	NAHF6	7,35	16,5	588	581	586	578	584	563	773	698	679	656	656	649	715	670
2	1	1.20.15	day before							603	603	602	602	594	590	683	718	643	650	648	661	685	679
2	1	1.21.15	morning	670	670	700	NA3LI	7,48	16,0	616	615	614	614	603	612	712	742	657	628	648	713	704	696
2	1	1.21.15	post 8h	520	540	530	NA3LI	7,48	16,0	601	606	600	604	595	593	694	701	634	652	669	656	681	689
2	2	2.3.15	day before							608	603	607	602	598	592	708	733	639	657	660	661	687	683
2	2	2.4.15	morning	170	210	200	NA3LG	8,05	16,0	603	611	603	608	606	600	730	725	651	661	660	659	699	703
2	2	2.4.15	post 8h	40	60	50	NA3LG	8,05	16,0	618	607	618	604	610	598	693	734	640	637	676	683	705	687
3	1	1.15.15	day before							564	563	562	560	553	546	732	683	605	625	647	627	644	643
3	1	1.16.15	morning	710	740	740	NAG4F	7,2	17,0	574	570	567	563	558	553	734	715	613	607	681	635	639	663
3	1	1.16.15	post 8h	620	580	-	NAG4F	7,2	17,0	585	563	582	558	571	549	715	649	660	674	640	658	693	617
3	2	1.27.15	day before							567	562	563	558	558	547	718	685	610	619	641	641	669	640
3	2	1.28.15	morning	220	240	280	NAG4E	7,8	17,0	565	568	559	565	552	551	696	706	629	615	639	637	671	646
3	2	1.28.15	post 8h	80	110	120	NAG4E	7,8	17,0	567	564	563	560	563	552	733	711	615	611	651	648	680	638
4	1	1.21.15	day before							548	549	544	545	538	537	695	684	613	613	619	623	670	660
4	1	1.22.15	morning	200	200	190	NA647	7,98	16,5	556	553	554	548	541	537	689	680	620	629	622	624	677	680
4	1	1.22.15	post 8h	80	110	90	NA647	7,98	16,5	559	547	554	543	550	539	715	710	647	590	611	635	701	651
4	2	2.5.15	day before							557	554	552	548	543	544	704	702	598	595	612	614	671	671
4	2	2.6.15	morning	480	450	450	NA648	7,6	16,5	564	564	560	560	543	541	691	690	617	621	613	610	686	680
4	2	2.6.15	post 8h	430	340	330	NA648	7,6	16,5	558	548	553	544	563	540	738	690	607	606	645	604	695	675
5	1	2.19.15	day before							548	551	543	548	534	545	666	713	594	584	592	621	642	649
5	1	2.20.15	morning	210	200	200	NA3LF	8,02	17,0	549	562	543	559	538	555	695	717	606	600	627	624	640	682
5	1	2.20.15	post 8h	100	100	100	NA3LF	8,02	17,0	544	558	539	554	545	545	722	694	588	595	646	611	629	659
5	2	3.3.15	day before							537	558	533	550	537	549	677	698	578	589	603	631	637	642
5	2	3.4.15	morning	590	580	580	NA3LD	7,63	17,0	553	565	550	562	547	563	690	717	606	603	587	617	682	703
5	2	3.4.15	post 8h	370	370	370	NA3LD	7,63	17,0	555	553	552	549	547	551	717	698	586	602	622	597	660	692
6	1	4.24.15	day before							557	549	553	546	548	547	666	701	629	616	618	639	666	634
6	1	4.25.15	morning	150	210	150	NA1NG	7,7	16,0	564	556	561	552	553	551	680	671	639	620	632	619	661	670
6	1	4.25.15	post 8h	60	30	60	NA1NG	7,7	16,0														
6	2	1.27.15	day before							558	551	556	548	552	545	644	614	635	647	611	625	675	644
6	2	1.28.15	morning	650	700	680	NA1NH	7,25	16,0	566	559	563	557	558	548	675	?	645	643	647	664	668	629
6	2	1.28.15	post 8h	310	450	380	NA1NH	7,25	16,0	570	551	568	549	567	543	669	648	664	632	648	621	672	640
7	1	2.5.15	day before							493	485	487	474	478	475	597	584	528	553	528	541	580	583
7	1	2.6.15	morning	200	280	230	NA1NI	7,95	17,0	495	491	487	485	471	476	591	615	536	533	536	561	571	571
7	1	2.6.15	post 8h	110	100	110	NA1NI	7,95	17,0	495	482	477	477	478	477	590	582	559	571	525	541	598	581
7	2	2.12.15	day before							482	486	474	481	478	480	603	625	533	528	520	562	595	571
7	2	2.13.15	morning	670	650	600	NA3LD	7,63	17,0	489	489	478	485	475	477	600	600	535	556	531	558	588	567
7	2	2.13.15	post 8h	340	370	380	NA3LD	7,63	17,0	498	483	486	478	490	481	609	629	544	525	548	546	585	585
8	1	1.15.15	day before							580	582	576	574	582	577	732	722	625	622	637	642	711	699
8	1	1.16.15	morning	180	190	190	NA3L0	8,32	17,0	591	586	585	579	566	570	706	726	631	634	641	632	697	716
8	1	1.16.15	post 8h	70	90	-	NA3L0	8,32	17,0	586	586	581	582	590	573	778	720	619	623	679	643	707	695
8	2	2.3.15	day before							590	588	584	584	583	567	738	710	619	639	627	631	716	693
8	2	2.4.15	morning	660	650	700	NAG4E	7,8	17,0	587	590	585	586	576	567	719	710	627	634	651	654	697	685
8	2	2.4.15	post 8h	410	420	410	NAG4E	7,8	17,0	586	587	582	580	583	578	744	733	631	620	656	662	707	679
9	1	1.22.15	day before							538	523	527	514	525	511	671	652	564	585	593	568	622	622
9	1	1.23.15	morning	580	540	520	NAG4E	7,8	17,0	549	530	520	523	524	511	650	651	577	591	593	588	618	609
9	1	1.23.15	post 8h	470	430	430	NAG4E	7,8	17,0	549	525	541	516	539	510	673	671	608	579	607	584	631	607
9	2	2.5.15	day before							539	536	530	529	522	509	651	652	581	584	580	569	617	611
9	2	2.6.15	morning	240	170	190	NA9AI	8,18	17,0	535	542	514	539	524	515	639	650	608	576	573	568	627	633
9	2	2.6.15	post 8h	70	100	100	NA9AI	8,18	17,0	545	526	529	511	532	512	633	632	642	588	595	567	640	623
10	1	2.24.15	day before							584	585	580	581	575	573	727	681	633	694	648	656	732	707
10	1	2.25.15	morning	680	620	650	NA3LI	7,48	16,0	580	579	574	575	568	569	698	681	675	676	657	666	731	709
10	1	2.25.15	post 8h	400	400	420	NA3LI	7,48	16,0	582	579	579	574	582	568	693	690	669	644	667	661	755	697
10	2	3.3.15	day before							566	583	561	579	571	570	712	728	667	625	640	680	724	664
10	2	3.4.15	morning	200	190	210	NARH0	7,93	16,0	582	588	578	584	582	574	704	709	668	643	627	675	767	709
10	2	3.4.15	post 8h	80	90	90	NARH0	7,93	16,0	582	578	577	574	583	569	710	671	671	653	659	647	746	693

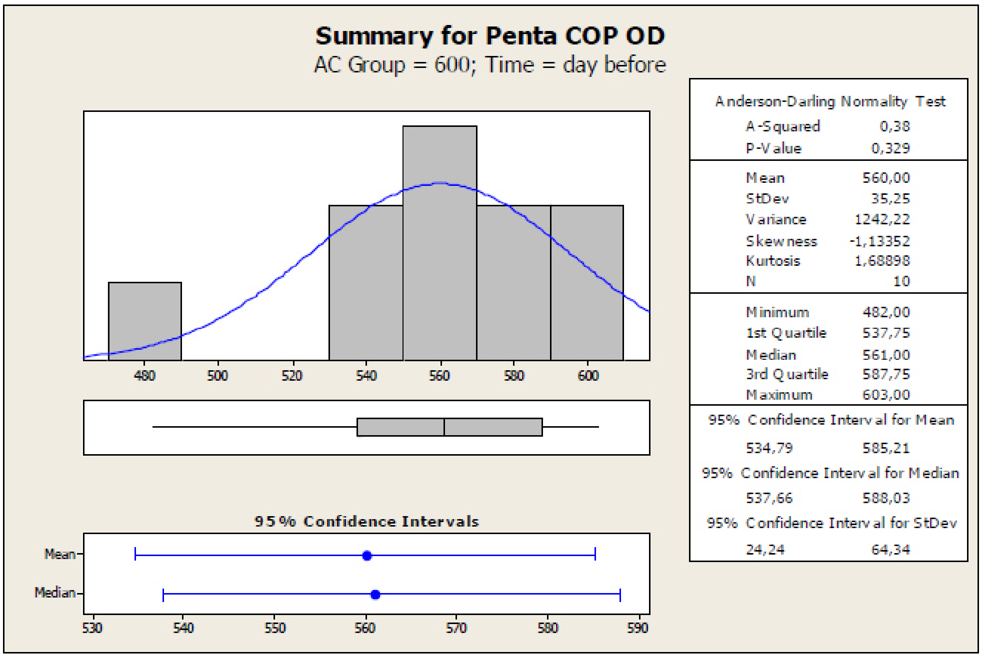
A 2: Normal Distribution Testing

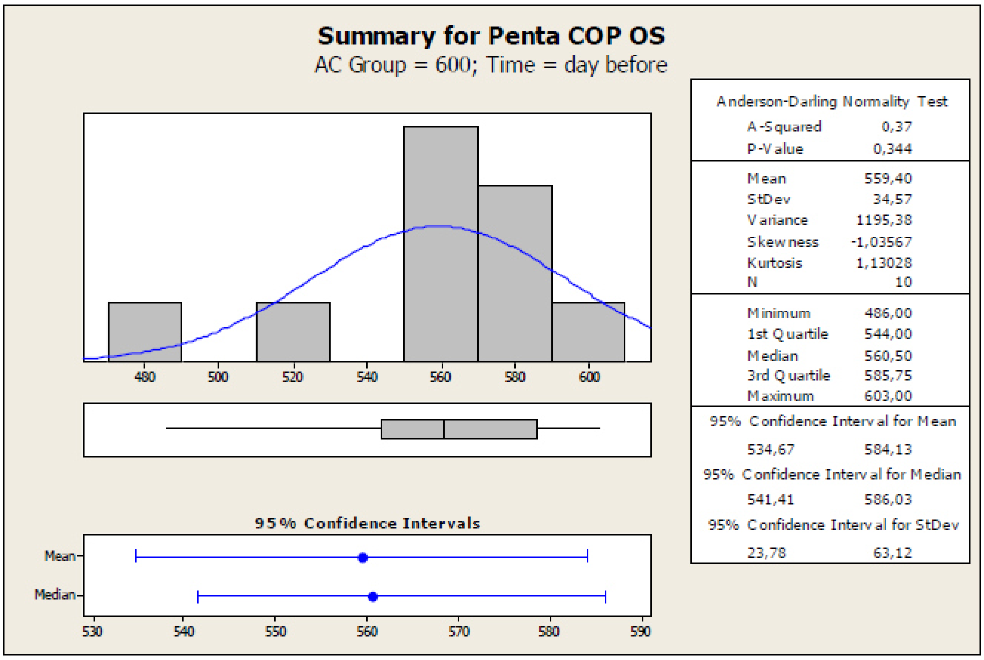


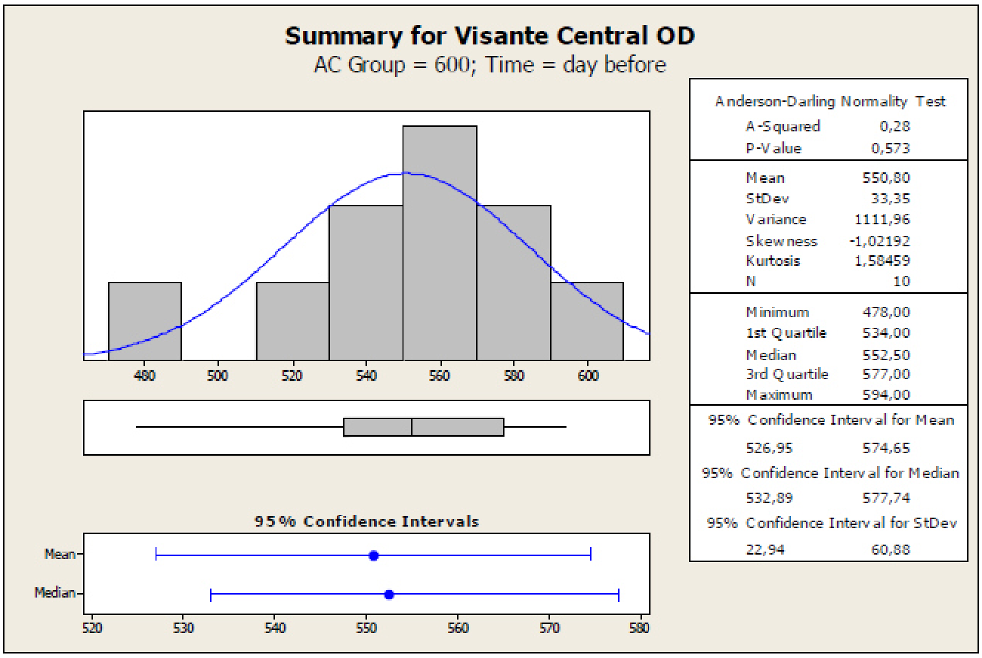


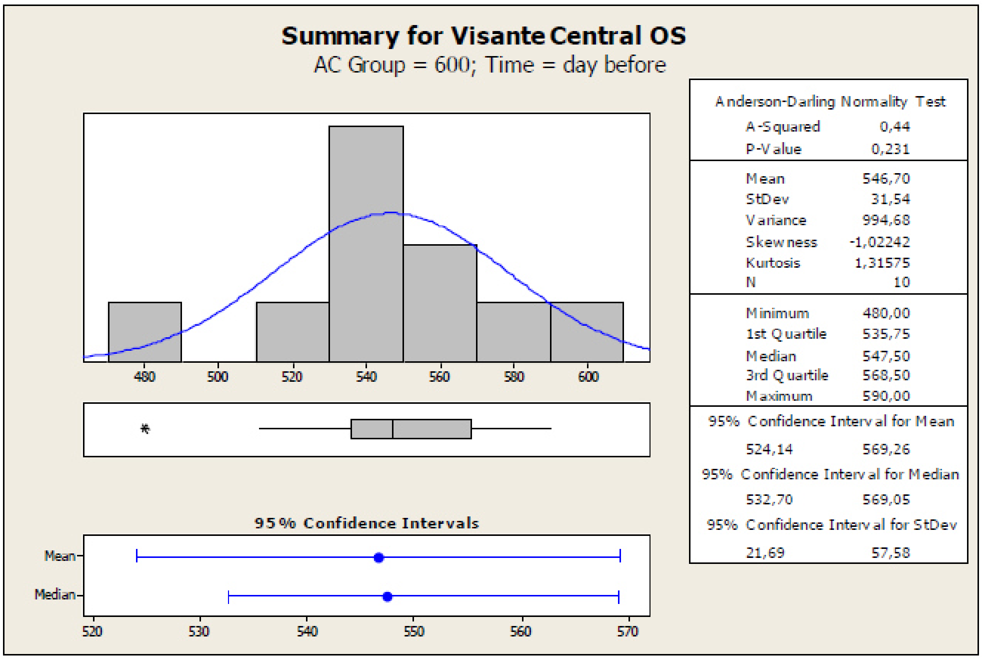

Summary for Visante Central OD
AC Group = 200; Time = day before
Anderson-Darling Normality Test
A-Squared 0,22
P-Value 0,774
Mean 550,20
StDev 34,53
Variance 1192,62
Skewness -0,816653
Kurtosis 0,970166
N 10
Minimum 478,00
1st Quartile 531,00
Median 553,00
3rd Quartile 575,25
Maximum 598,00
95% Confidence Interval for Mean
525,50 574,90
95% Confidence Interval for Median
529,89 576,08
95% Confidence Interval for StDev
23,75 63,05
95% Confidence Intervals
Mean
Median

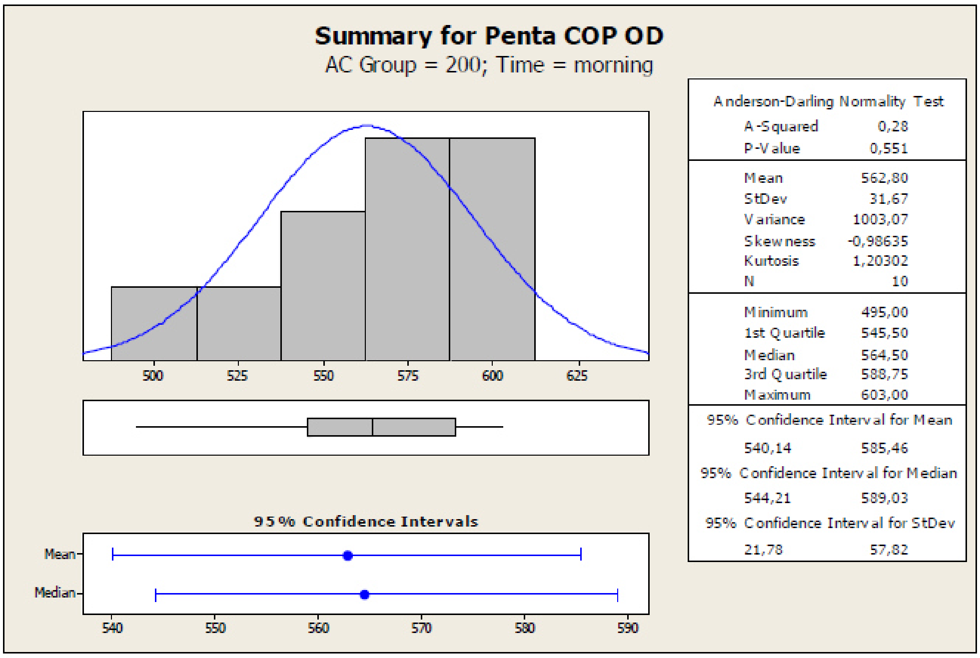

Summary for Visante Central OS
AC Group = 200; Time = day before
Anderson-Darling Normality Test
A-Squared 0,36
P-Value 0,370
Mean 546,70
StDev 34,36
Variance 1180,68
Skewness -0,958619
Kurtosis 0,985892
N 10
Minimum 475,00
1st Quartile 530,00
Median 547,00
3rd Quartile 571,75
Maximum 592,00
95% Confidence Interval for Mean
522,12 571,28
95% Confidence Interval for Median
527,41 572,40
95% Confidence Interval for StDev
23,63 62,73
95% Confidence Intervals
Mean
Median

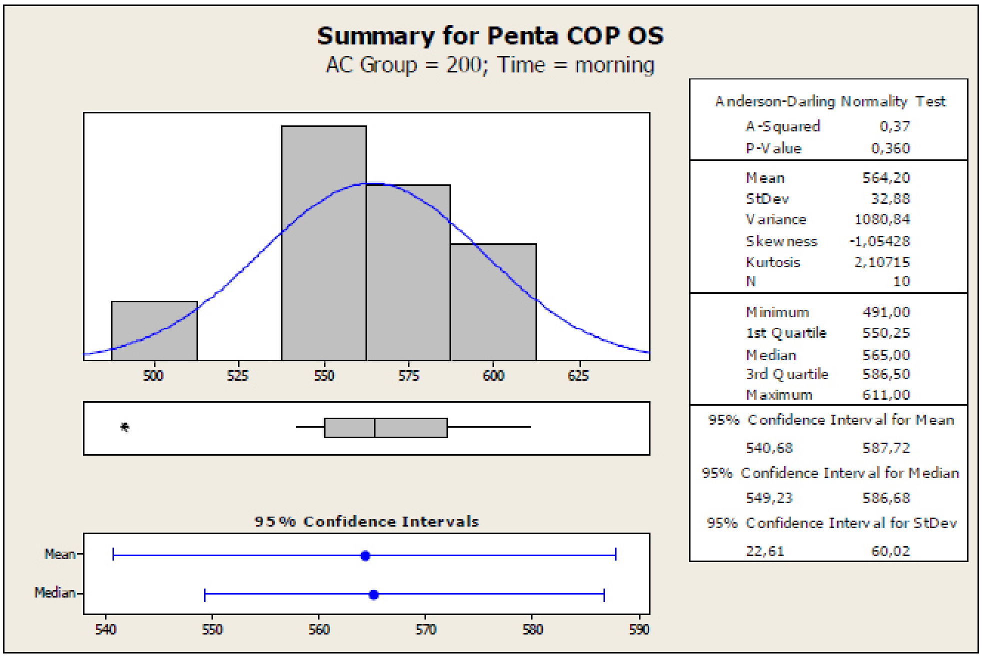

Summary for Penta COP OD
AC Group = 600; Time = day before
Anderson-Darling Normality Test
A-Squared 0,38
P-Value 0,329
Mean 560,00
StDev 35,25
Variance 1242,22
Skewness -1,13352
Kurtosis 1,68898
N 10
Minimum 482,00
1st Quartile 537,75
Median 561,00
3rd Quartile 587,75
Maximum 603,00
95% Confidence Interval for Mean
534,79 585,21
95% Confidence Interval for Median
537,66 588,03
95% Confidence Interval for StDev
24,24 64,34
95% Confidence Intervals
Mean
Median
480 500 520 540 560 580 600
530 540 550 560 570 580 590

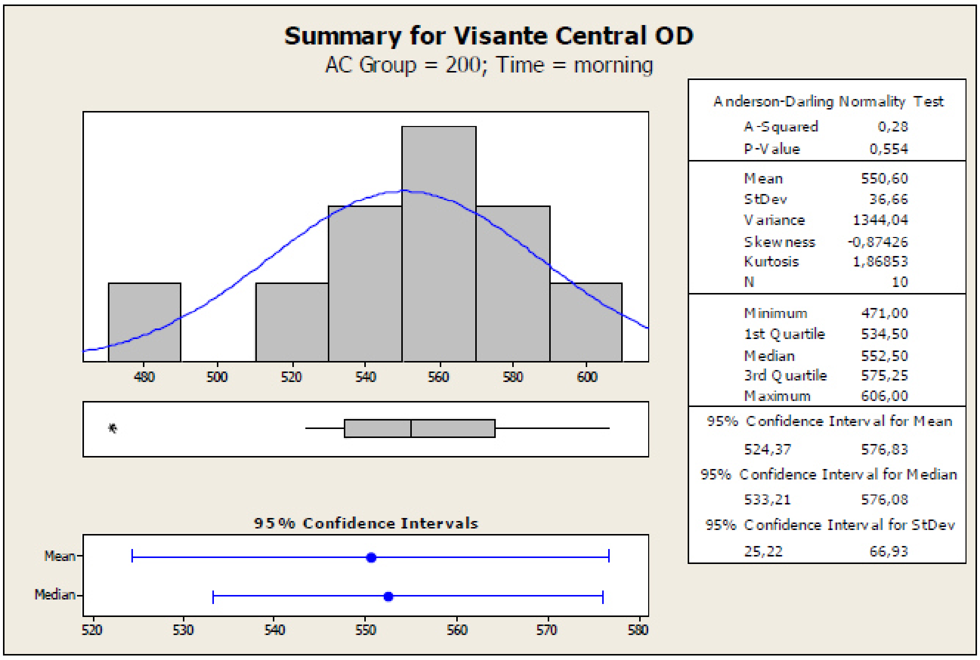

Summary for Penta COP OS
AC Group = 600; Time = day before
Anderson-Darling Normality Test
A-Squared 0,37
P-Value 0,344
Mean 559,40
StDev 34,57
Variance 1195,38
Skewness -1,03567
Kurtosis 1,13028
N 10
Minimum 486,00
1st Quartile 544,00
Median 560,50
3rd Quartile 585,75
Maximum 603,00
95% Confidence Interval for Mean
534,67 584,13
95% Confidence Interval for Median
541,41 586,03
95% Confidence Interval for StDev
23,78 63,12
95% Confidence Intervals
Mean
Median
480 500 520 540 560 580 600
530 540 550 560 570 580 590

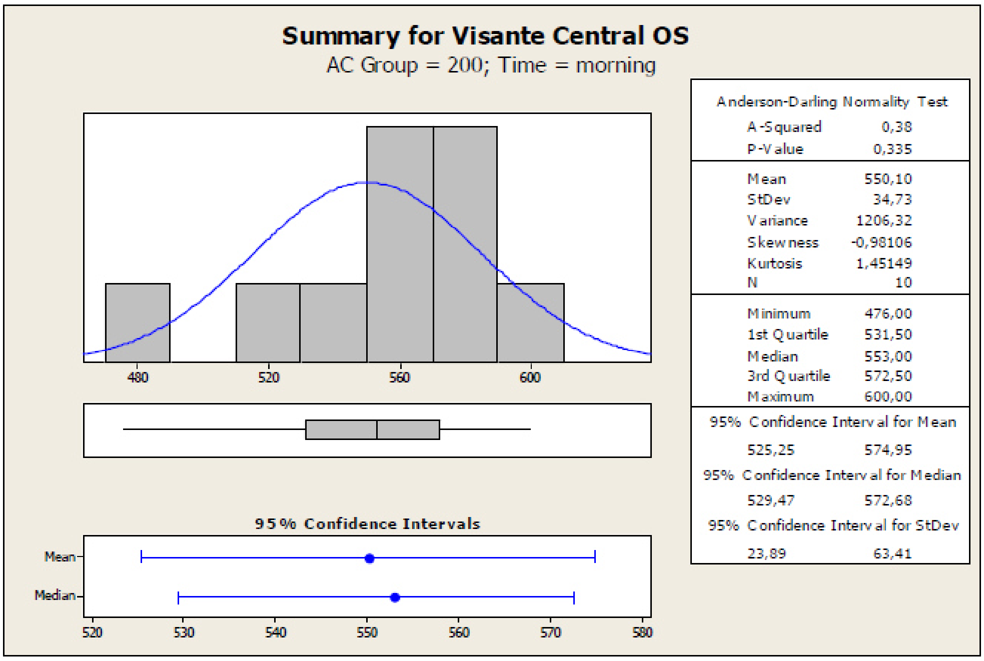

Summary for Visante Central OD
AC Group = 600; Time = day before
Anderson-Darling Normality Test
A-Squared 0,28
P-Value 0,573
Mean 550,80
StDev 33,35
Variance 1111,96
Skewness -1,02192
Kurtosis 1,58459
N 10
Minimum 478,00
1st Quartile 534,00
Median 552,50
3rd Quartile 577,00
Maximum 594,00
95% Confidence Interval for Mean
526,95 574,65
95% Confidence Interval for Median
532,89 577,74
95% Confidence Interval for StDev
22,94 60,88
480 500 520 540 560 580 600
95% Confidence Intervals
Mean
Median
520 530 540 550 560 570 580

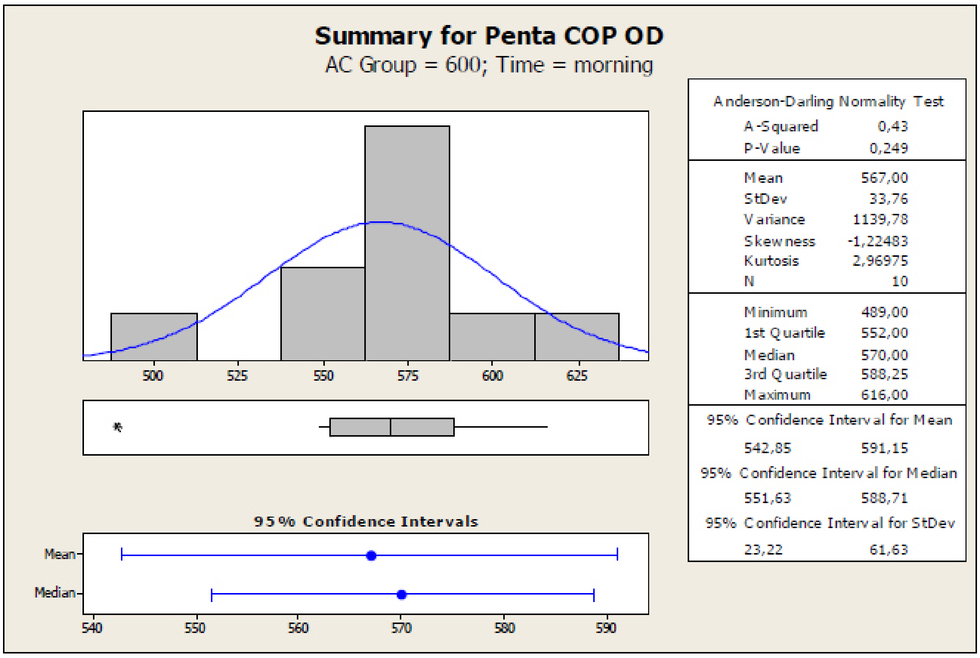

Summary for Visante Central OS
AC Group = 600; Time = day before
Anderson-Darling Normality Test
A-Squared 0,44
P-Value 0,231
Mean 546,70
StDev 31,54
Variance 994,68
Skewness -1,02242
Kurtosis 1,31575
N 10
Minimum 480,00
1st Quartile 535,75
Median 547,50
3rd Quartile 568,50
Maximum 590,00
95% Confidence Interval for Mean
524,14 569,26
95% Confidence Interval for Median
532,70 569,05
95% Confidence Interval for StDev
21,69 57,58
480 500 520 540 560 580 600
95% Confidence Intervals
Mean
Median
520 530 540 550 560 570

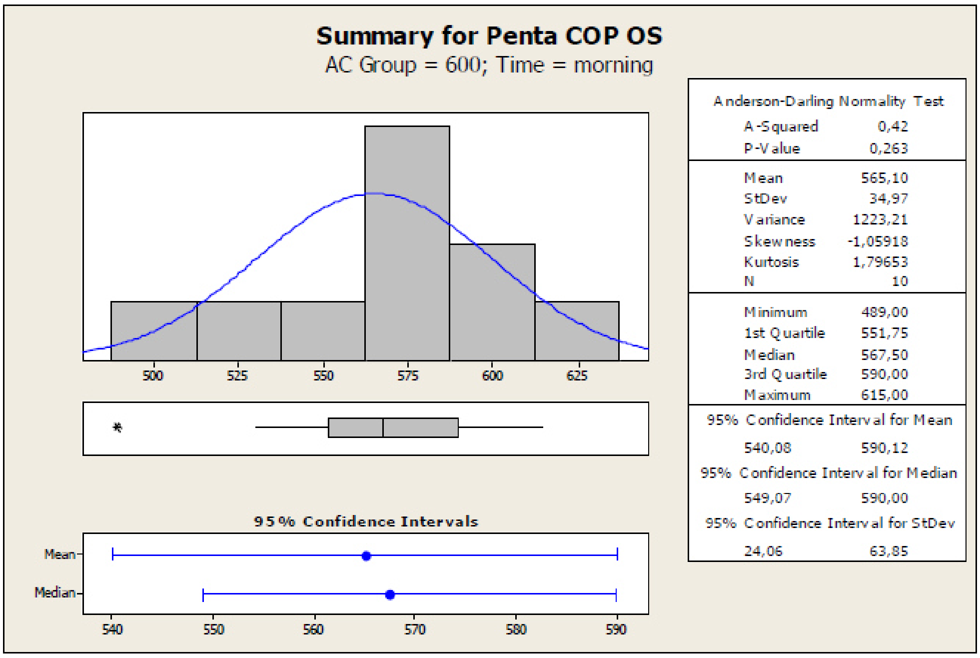

Summary for Penta COP OD
AC Group = 200; Time = morning
Anderson-Darling Normality Test
A-Squared 0,28
P-Value 0,551
Mean 562,80
StDev 31,67
Variance 1003,07
Skewness -0,98635
Kurtosis 1,20302
N 10
Minimum 495,00
1st Quartile 545,50
Median 564,50
3rd Quartile 588,75
Maximum 603,00
95% Confidence Interval for Mean
540,14 585,46
95% Confidence Interval for Median
544,21 589,03
95% Confidence Interval for StDev
21,78 57,82
500 525 550 575 600 625
95% Confidence Intervals
Mean
Median
540 550 560 570 580 590

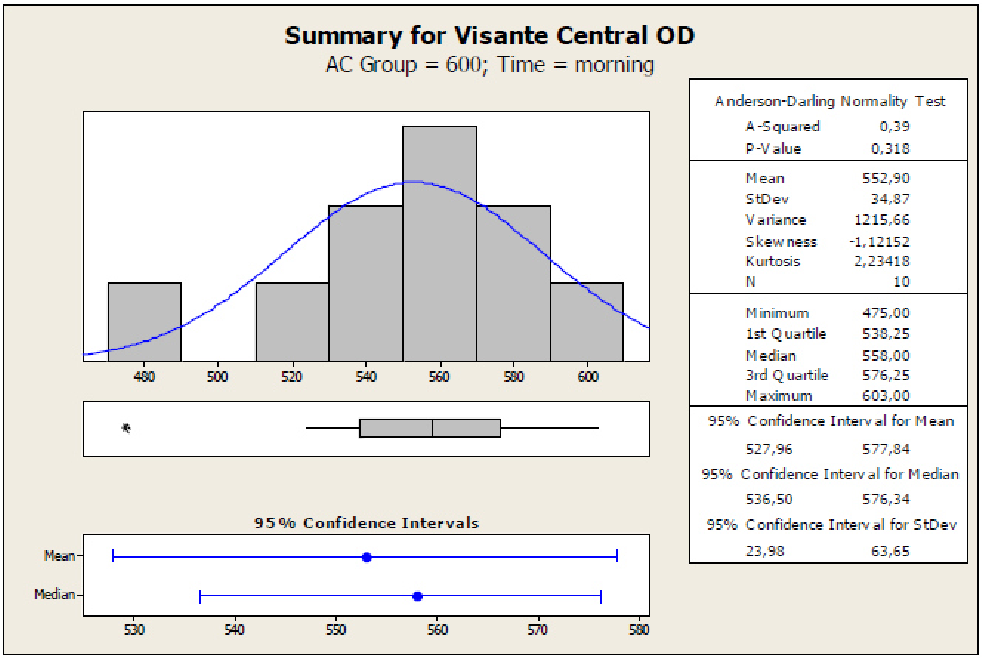

Summary for Penta COP OS
AC Group = 200; Time = morning
Anderson-Darling Normality Test
A-Squared 0,37
P-Value 0,360
Mean 564,20
StDev 32,88
Variance 1080,84
Skewness -1,05428
Kurtosis 2,10715
N 10
Minimum 491,00
1st Quartile 550,25
Median 565,00
3rd Quartile 586,50
Maximum 611,00
95% Confidence Interval for Mean
540,68 587,72
95% Confidence Interval for Median
549,23 586,68
95% Confidence Interval for StDev
22,61 60,02
500 525 550 575 600 625
95% Confidence Intervals
Mean
Median
540 550 560 570 580 590

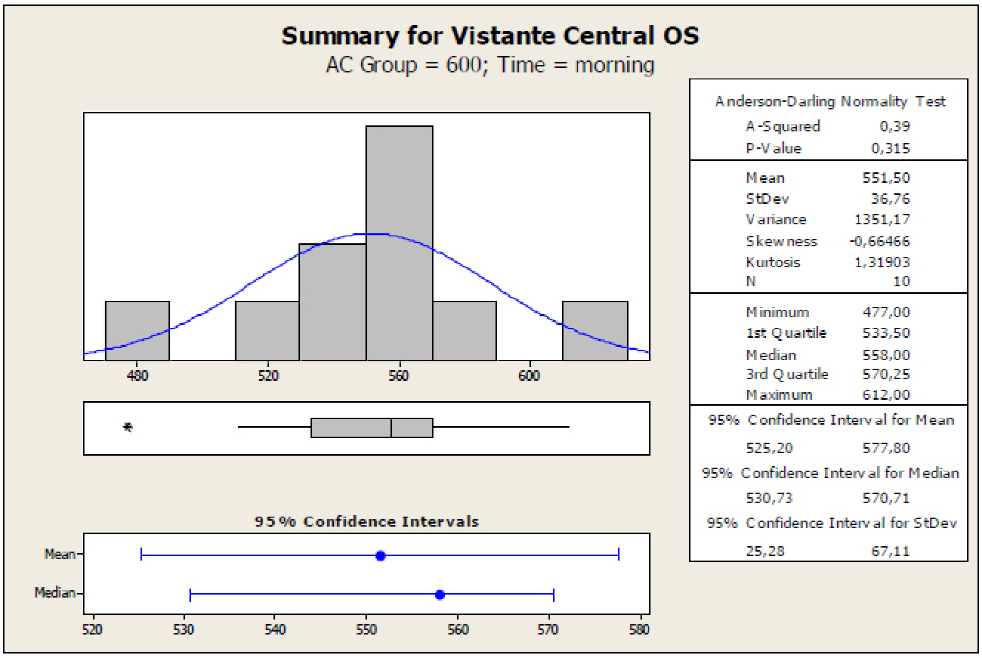

Summary for Visante Central OD
AC Group = 200; Time = morning
Anderson-Darling Normality Test
A-Squared 0,28
P-Value 0,554
Mean 550,60
StDev 36,66
Variance 1344,04
Skewness -0,87426
Kurtosis 1,86853
N 10
Minimum 471,00
1st Quartile 534,50
Median 552,50
3rd Quartile 575,25
Maximum 606,00
95% Confidence Interval for Mean
524,37 576,83
95% Confidence Interval for Median
533,21 576,08
95% Confidence Interval for StDev
25,22 66,93
480 500 520 540 560 580 600
95% Confidence Intervals
Mean
Median
520 530 540 550 560 570 580

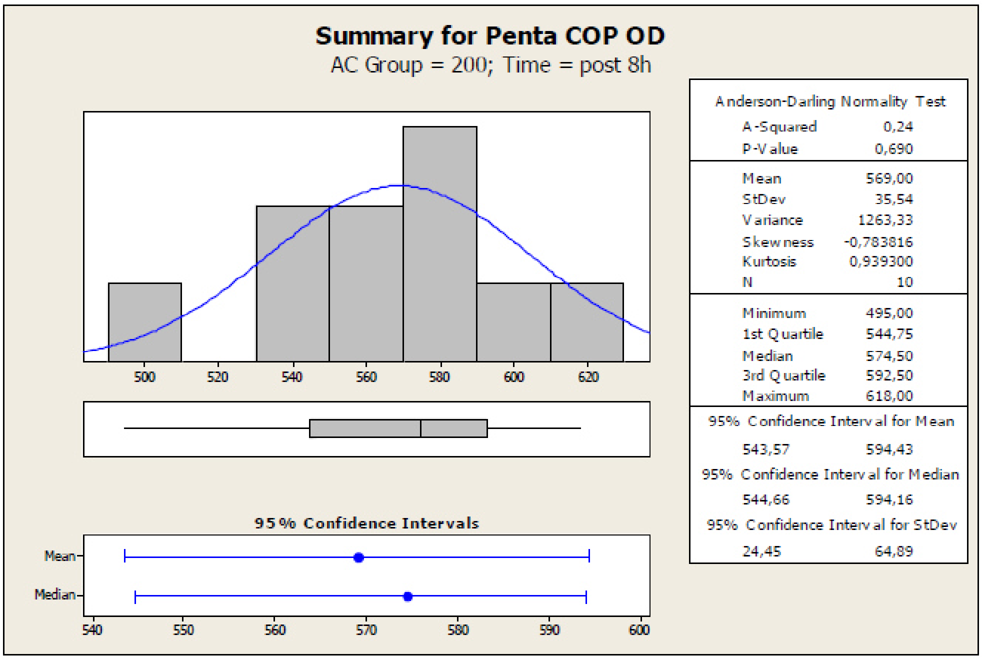

Summary for Visante Central OS
AC Group = 200; Time = morning
Anderson-Darling Normality Test
A-Squared 0,38
P-Value 0,335
Mean 550,10
StDev 34,73
Variance 1206,32
Skewness -0,98106
Kurtosis 1,45149
N 10
Minimum 476,00
1st Quartile 531,50
Median 553,00
3rd Quartile 572,50
Maximum 600,00
95% Confidence Interval for Mean
525,25 574,95
95% Confidence Interval for Median
529,47 572,68
95% Confidence Interval for StDev
23,89 63,41
480 520 560 600
95% Confidence Intervals
Mean
Median
520 530 540 550 560 570 580

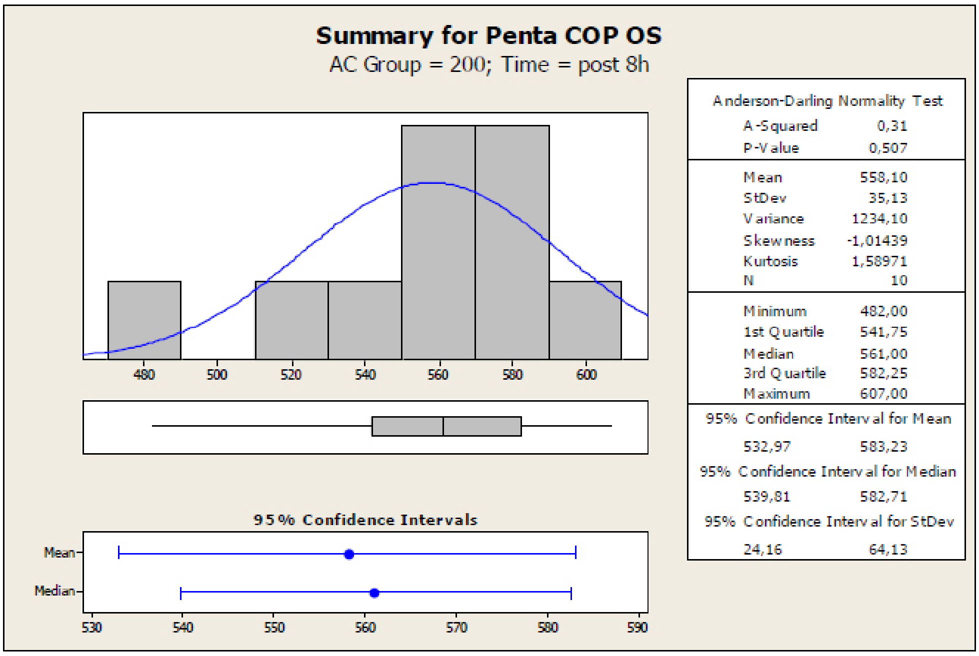

Summary for Penta COP OD
AC Group = 600; Time = morning
Anderson-Darling Normality Test
A-Squared 0,43
P-Value 0,249
Mean 567,00
StDev 33,76
Variance 1139,78
Skewness -1,22483
Kurtosis 2,96975
N 10
Minimum 489,00
1st Quartile 552,00
Median 570,00
3rd Quartile 588,25
Maximum 616,00
95% Confidence Interval for Mean
542,85 591,15
95% Confidence Interval for Median
551,63 588,71
95% Confidence Interval for StDev
23,22 61,63
95% Confidence Intervals
Mean
Median
500 525 550 575 600 625
540 550 560 570 580 590

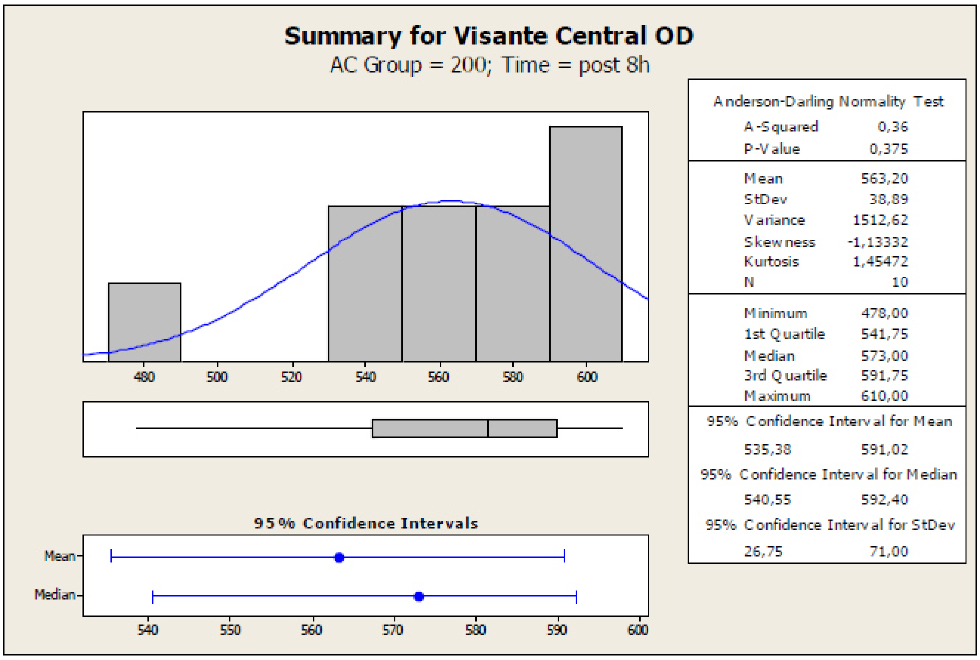

Summary for Penta COP OS
AC Group = 600; Time = morning
Anderson-Darling Normality Test
A-Squared 0,42
P-Value 0,263
Mean 565,10
StDev 34,97
Variance 1223,21
Skewness -1,05918
Kurtosis 1,79653
N 10
Minimum 489,00
1st Quartile 551,75
Median 567,50
3rd Quartile 590,00
Maximum 615,00
95% Confidence Interval for Mean
540,08 590,12
95% Confidence Interval for Median
549,07 590,00
95% Confidence Interval for StDev
24,06 63,85
95% Confidence Intervals
Mean
Median
500 525 550 575 600 625
540 550 560 570 580 590

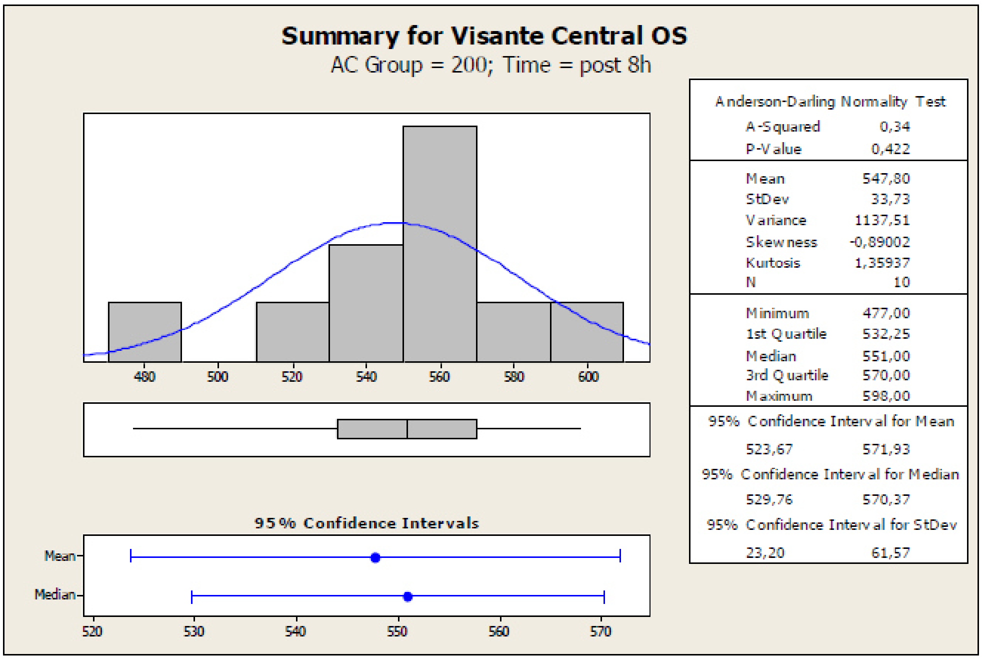

Summary for Visante Central OD
AC Group = 600; Time = morning
Anderson-Darling Normality Test
A-Squared 0,39
P-Value 0,318
Mean 552,90
StDev 34,87
Variance 1215,66
Skewness -1,12152
Kurtosis 2,23418
N 10
Minimum 475,00
1st Quartile 538,25
Median 558,00
3rd Quartile 576,25
Maximum 603,00
95% Confidence Interval for Mean
527,96 577,84
95% Confidence Interval for Median
536,50 576,34
95% Confidence Interval for StDev
23,98 63,65
95% Confidence Intervals
Mean
Median
480 500 520 540 560 580 600
530 540 550 560 570 580

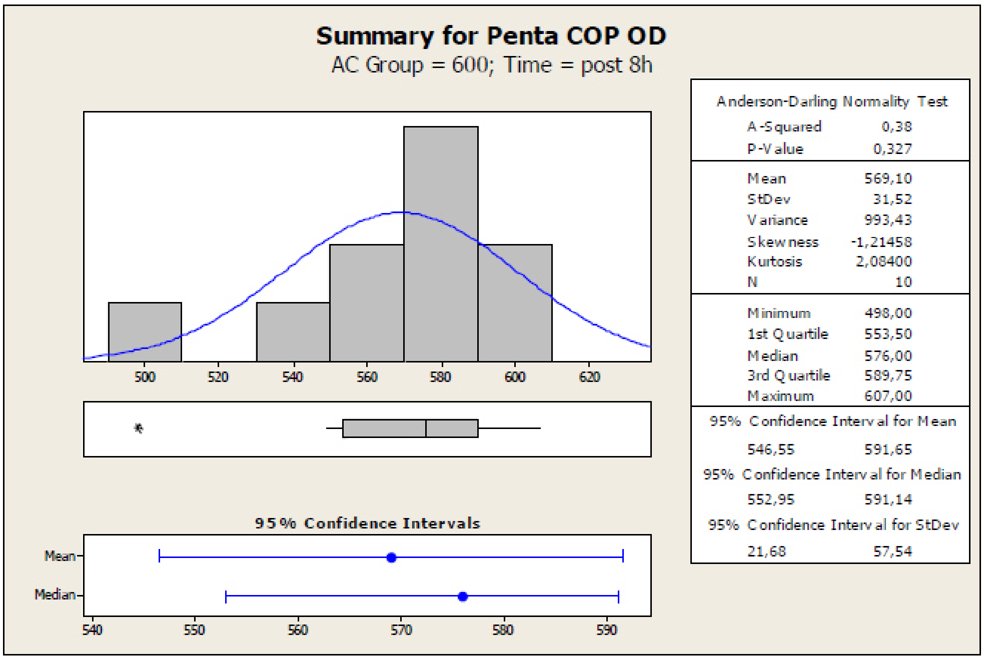

Summary for Vistante Central OS
AC Group = 600; Time = morning
Anderson-Darling Normality Test
A-Squared 0,39
P-Value 0,315
Mean 551,50
StDev 36,76
Variance 1351,17
Skewness -0,66466
Kurtosis 1,31903
N 10
Minimum 477,00
1st Quartile 533,50
Median 558,00
3rd Quartile 570,25
Maximum 612,00
95% Confidence Interval for Mean
525,20 577,80
95% Confidence Interval for Median
530,73 570,71
95% Confidence Interval for StDev
25,28 67,11
95% Confidence Intervals
Mean
Median
480 520 560 600
520 530 540 550 560 570 580

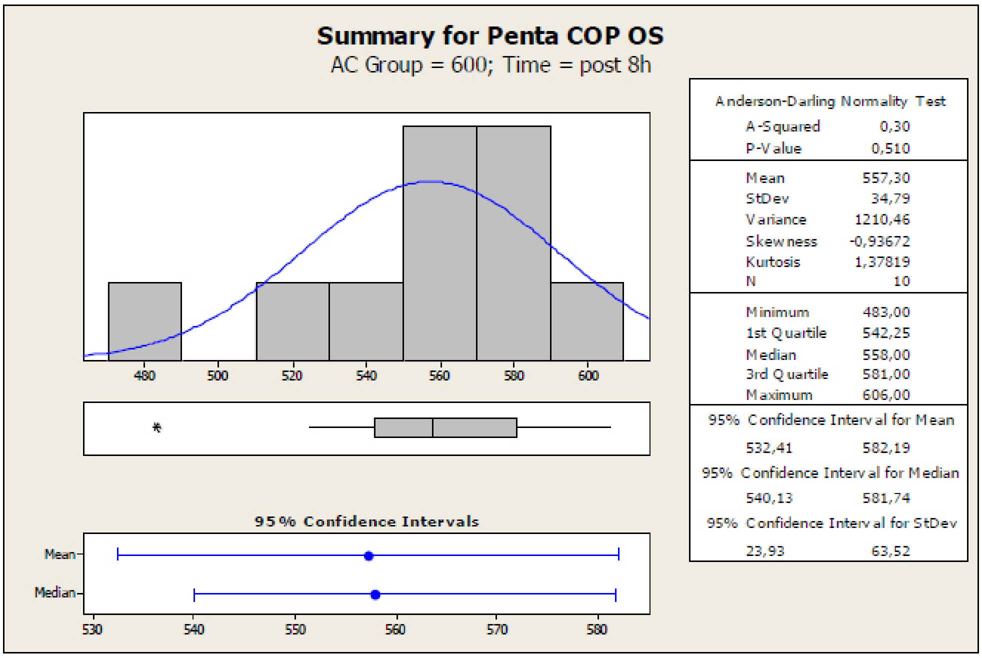

Summary for Penta COP OD
AC Group = 200; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,24
P-Value 0,690
Mean 569,00
StDev 35,54
Variance 1263,33
Skewness -0,783816
Kurtosis 0,939300
N 10
Minimum 495,00
1st Quartile 544,75
Median 574,50
3rd Quartile 592,50
Maximum 618,00
95% Confidence Interval for Mean
543,57 594,43
95% Confidence Interval for Median
544,66 594,16
95% Confidence Interval for StDev
24,45 64,89
95% Confidence Intervals
Mean
Median
500 520 540 560 580 600 620
540 550 560 570 580 590 600

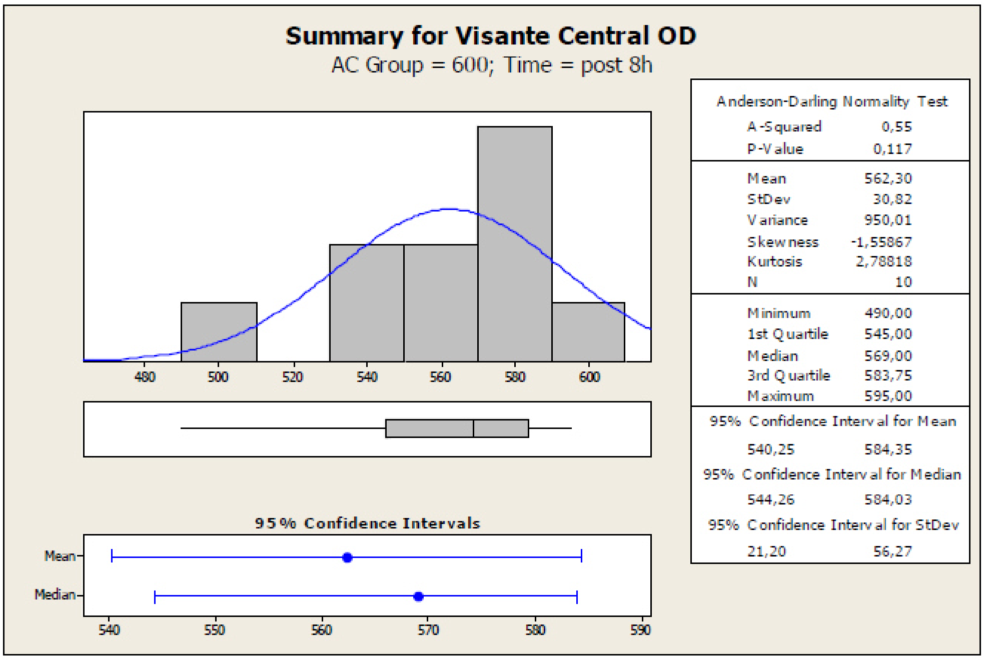

Summary for Penta COP OS
AC Group = 200; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,31
P-Value 0,507
Mean 558,10
StDev 35,13
Variance 1234,10
Skewness -1,01439
Kurtosis 1,58971
N 10
Minimum 482,00
1st Quartile 541,75
Median 561,00
3rd Quartile 582,25
Maximum 607,00
95% Confidence Interval for Mean
532,97 583,23
95% Confidence Interval for Median
539,81 582,71
95% Confidence Interval for StDev
24,16 64,13
95% Confidence Intervals
Mean
Median
480 500 520 540 560 580 600
530 540 550 560 570 580 590

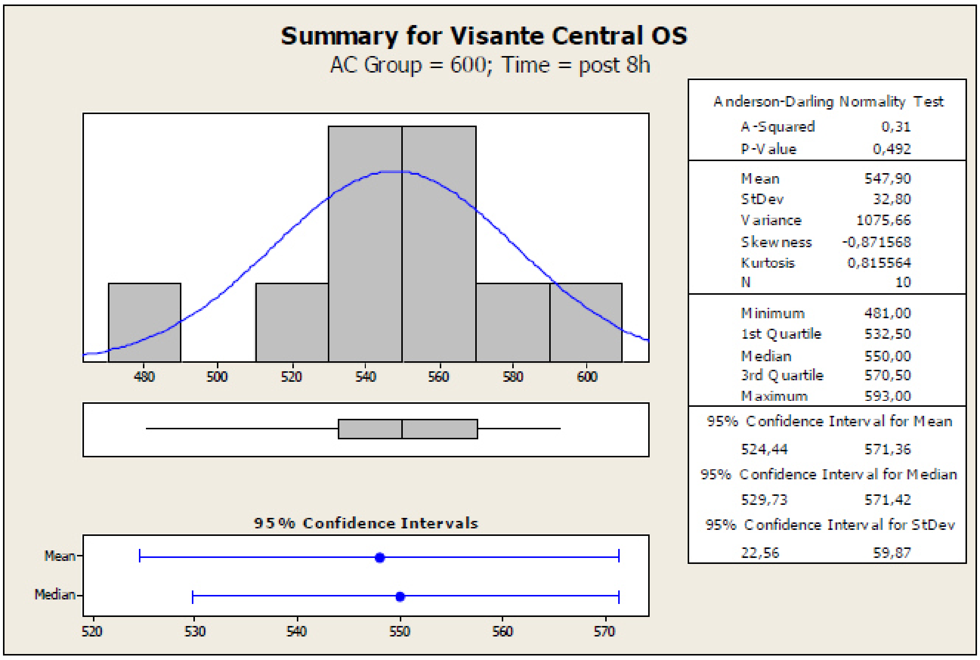

Summary for Visante Central OD
AC Group = 200; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,36
P-Value 0,375
Mean 563,20
StDev 38,89
Variance 1512,62
Skewness -1,13332
Kurtosis 1,45472
N 10
Minimum 478,00
1st Quartile 541,75
Median 573,00
3rd Quartile 591,75
Maximum 610,00
95% Confidence Interval for Mean
535,38 591,02
95% Confidence Interval for Median
540,55 592,40
95% Confidence Interval for StDev
26,75 71,00
480 500 520 540 560 580 600
95% Confidence Intervals
Mean
Median
540 550 560 570 580 590 600


Summary for Visante Central OS
AC Group = 200; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,34
P-Value 0,422
Mean 547,80
StDev 33,73
Variance 1137,51
Skewness -0,89002
Kurtosis 1,35937
N 10
Minimum 477,00
1st Quartile 532,25
Median 551,00
3rd Quartile 570,00
Maximum 598,00
95% Confidence Interval for Mean
523,67 571,93
95% Confidence Interval for Median
529,76 570,37
95% Confidence Interval for StDev
23,20 61,57
480 500 520 540 560 580 600
95% Confidence Intervals
Mean
Median
520 530 540 550 560 570


Summary for Penta COP OD
AC Group = 600; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,38
P-Value 0,327
Mean 569,10
StDev 31,52
Variance 993,43
Skewness -1,21458
Kurtosis 2,08400
N 10
Minimum 498,00
1st Quartile 553,50
Median 576,00
3rd Quartile 589,75
Maximum 607,00
95% Confidence Interval for Mean
546,55 591,65
95% Confidence Interval for Median
552,95 591,14
95% Confidence Interval for StDev
21,68 57,54
95% Confidence Intervals
Mean
Median
500 520 540 560 580 600 620
540 550 560 570 580 590


Summary for Penta COP OS
AC Group = 600; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,30
P-Value 0,510
Mean 557,30
StDev 34,79
Variance 1210,46
Skewness -0,93672
Kurtosis 1,37819
N 10
Minimum 483,00
1st Quartile 542,25
Median 558,00
3rd Quartile 581,00
Maximum 606,00
95% Confidence Interval for Mean
532,41 582,19
95% Confidence Interval for Median
540,13 581,74
95% Confidence Interval for StDev
23,93 63,52
95% Confidence Intervals
Mean
Median
480 500 520 540 560 580 600
530 540 550 560 570 580


Summary for Visante Central OD
AC Group = 600; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,55
P-Value 0,117
Mean 562,30
StDev 30,82
Variance 950,01
Skewness -1,55867
Kurtosis 2,78818
N 10
Minimum 490,00
1st Quartile 545,00
Median 569,00
3rd Quartile 583,75
Maximum 595,00
95% Confidence Interval for Mean
540,25 584,35
95% Confidence Interval for Median
544,26 584,03
95% Confidence Interval for StDev
21,20 56,27
95% Confidence Intervals
Mean
Median


Summary for Visante Central OS
AC Group = 600; Time = post 8h
Anderson-Darling Normality Test
A-Squared 0,31
P-Value 0,492
Mean 547,90
StDev 32,80
Variance 1075,66
Skewness -0,871568
Kurtosis 0,815564
N 10
Minimum 481,00
1st Quartile 532,50
Median 550,00
3rd Quartile 570,50
Maximum 593,00
95% Confidence Interval for Mean
524,44 571,36
95% Confidence Interval for Median
529,73 571,42
95% Confidence Interval for StDev
22,56 59,87
95% Confidence Intervals
Mean
Median