Original Article

Medial and lateral canthal ligaments shown in P45 sheet plastination and dissection

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Purpose: Ocular suspensory ligament is an important part of the lower eyelid retractors. However, there is a scarcity of studies examining detailed en-block histologies of ocular suspensory ligaments. **Methods:** In this study, we included the cadavers of Chinese adults as subjects. These cadavers of Chinese adults were processed using P45 plastination techniques. The polymer resulted in transparent plastination, and the P45 sheet-plastinated sections of the lower eyelid were observed. The gross anatomy results of three Chinese adult heads (six hemifaces) were included as gross dissection data. All photographic documentation was performed via a Canon EOS 7D Mark camera. **Results:** The results showed that the inferior rectus muscle, inferior oblique muscle, ocular suspensory ligament, and its arcuate expansion are under the eyeball. The medial and lateral parts of the ocular suspensory ligament end at the medial and lateral canthal ligament. The middle part, a hammock-like shape, is slightly lower. The ocular suspensory ligament holds up the inferior oblique muscle, inferior rectus muscle, and the eyeball. As the inferior oblique muscle passes through the sheath of the inferior rectus, the fascia is thickened, forming the ocular suspensory ligament. The ocular suspensory ligament connects to the intermuscular septum, the inferior tarsal muscle, and the medial and lateral check ligaments. **Conclusion:** This study observed the ocular suspensory ligament and arcuate expansion through P45 sheet plastination for the first time and identified the distribution of the lower eyelid ligaments, thus laying the foundation for further research.

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The ocular suspensory ligament is an important part of the lower eyelid retractors.[1] Gray's Anatomy describes the sheath of the inferior rectus as thickened on its underside and blending with the sheath of the inferior oblique. These two, in turn, are continuous with the sheaths of the medial and lateral recti. Since the latter is attached to the orbital walls by check ligaments, a continuous fascial band, the suspensory ligament of the eye, is slung like a hammock below the eye, providing sufficient support.[2] It is connected from the medial canthal ligament to the lateral canthal ligament on the horizontal plane and has the function of keeping eyeballs in a normal position.[3,4]

A study reported that the ocular suspensory ligament can cause fat relaxation in the lower eyelid orbital septum due to the aging process.[5] Another study introduced suspending the orbicularis oculi on the ocular suspensory ligament during lower blepharoplasty without removing the fat.[6] Despite the importance of the ocular suspensory ligament in lower eyelid surgeries, few studies have shown detailed en-block histology of ocular suspensory ligaments.

Therefore, we conducted this study to elucidate a precise systematic and comprehensive anatomical study of the ocular suspensory ligament of Chinese people using P45 sheet plastination and gross dissection.

Methods

Subjects

In this study, we included cadavers of Chinese adults as the subjects, processed using the P45 plastination techniques. The polymer resulted in transparent plastination, enabling the P45 sheet-plastinated sections of the low eyelid to be observed. This study was conducted in accordance with the Declaration of Helsinki and approved by the institutional ethics committee of our university.

Gross dissection

After removal of the skin from the heads, the submuscular fibroadipose layer of the orbicularis oculi muscle in the lower eyelid region was exposed. The preseptal fat was removed from its region, exposing the orbital structures. The types of relationship between the ocular suspensory ligament and the capsulopalpebral fascia were observed and marked. Photographic documentation was carried out using a Canon EOS 7D Mark camera.

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P45 sheet plastination

Slicing

The embalmed specimens of the head and neck were frozen at -70°C for 2 weeks, and then embedded in polyurethane foam and frozen at -70°C, again for 2 days. After freezing, sagittal slices were made at 3 mm from side to side using a high-speed band saw. The volume of residual dust was 1 mm. The slices were placed in an orderly fashion on polyethylene grids with a piece of fly screen. A light stream of running water was used to remove any dust. The grids were then stacked on top of each other and tied using twine to hold the grids together as one unit. All the units were then labeled and put into square polyethylene pails.

Bleaching

All the slices were rinsed overnight in cold running water, after which the slices were immersed in 5% hydrogen peroxide overnight.

Dehydration

After bleaching, the slices were dehydrated using a freeze-substitution method. First, the slices were precooled at 5°C to avoid the formation of ice crystals and shrinkage before being placed into cold acetone. The slices were then placed in the first bath of 100% acetone at -25°C for 1 week and then transferred into a second bath of 100% acetone at -15°C for 10 days. Following these 10 days, they were then put into 100% acetone at room temperature for 1 week. The slices were finally submerged in fresh 100% acetone at room temperature. After 1 week, the slices were taken out for impregnation. The purity of the acetone was monitored with an acetonometer daily. Provided the purity remained similar on three observations, the slices were moved to a fresh dehydrating solution.

Casting and forced impregnation

After dehydration, the casting mold was prepared. This was a vertical chamber consisting of two plates of 5-mm tempered glass, flexible 4-mm latex tubing, and several large fold-back clamps. The slices were lifted out of the acetone bath and placed between the two glass plates. The molds were then filled with polyester (Hoffen polyester P45, China) via a funnel. The components of Hoffen polyester P45 were mixed at a ratio of 1000 mL of polyester P45 monomer to 10 g of P45a to 30 mL of P45b to 5 g of P45c. P45a and P45c were used as plasticizers, and P45b was used as a hardener for sheet plastination. After casting was completed, the filled mold was placed upright into a vacuum chamber at room temperature for impregnation. Larger bubbles were removed manually via a piece of 1-mm stainless steel wire. Absolute pressure was slowly decreased to 20 mmHg, 10 mmHg, 5 mmHg, and 0 mmHg according to the release of bubbles. A pressure of 0 mmHg was maintained until bubbling ceased. The duration of impregnation was more than 8 h.

Curing

After the vacuum was released, the air bubbles within the sheets were checked and removed. The alignment of the slices was checked and corrected using the steel wire. The top of the mold was clamped with large fold-back clamps, and the sheet was then ready for curing. The sheets were cured using heated bathwater. The sheets were placed upright in the bathwater at 40°C for 3 days. To equilibrate the temperature of the water, a small circulatory pump was used to circulate the water around the bath.

Cutting and sanding the molds

After curing, the sheets were removed from the bath and cooled to room temperature on a rack. The slices were then taken from the vertical chamber and were covered appropriately with adhesive plastic wrap for protection. A bend saw was used to cut and trim the plastic along the edges approximately 1 mm outside of the slices. Following this, a wool sander was used to remove the sharp edges of the slices, and after sanding, the adhesive plastic wraps were removed and the slices were put into plastic wraps to avoid scratches.

The head and neck sheets were then observed and photographed using LED reading lights.

Results

The general characteristics

In this study, a total of 14 heads (28 hemifaces) cadavers of Chinese adults were used (22 hemifaces for P45 sheet plastination,[6] hemifaces for gross dissection).

Gross dissection

Origin and insertion

The medial and lateral parts of the ocular suspensory ligament end at the medial and lateral canthal ligaments. The middle part is slightly lower, appearing as a hammock-like shape, holding up the inferior oblique muscle, inferior rectus muscle, and the eyeball [Fig. 1].

Adjacent structures

Muscle

The structures below the eyeball are the inferior rectus muscle, the inferior oblique muscle, the ocular suspensory ligament, and its arcuate expansion. Below the inferior oblique muscle, there is a fascia structure that issues forward, forming the ocular suspensory ligament.

Fat

The inferior oblique muscle and arcuate expansion, respectively travel inward and downward. They divide the postseptal fat of the lower eyelid into three independent parts: lateral, middle, and medial.

P45 Sheet-plastinated sections

The head of the capsulopalpebral fascia (CPF) splits open superiorly and inferiorly, wrapping around the inferior oblique muscle and meeting anteriorly. As the inferior oblique muscle passes through the sheath of the inferior rectus, the fascia is thickened, forming the ocular suspensory ligament. The CPF enters the inferior border of the tarsus, merging with the anterior border of the inferior tarsal muscles. The intermuscular septum, the inferior tarsal muscle, and the medial and lateral check ligaments are connected to the ocular suspensory ligament.

Sagittal plane: (10 hemifaces observed)

Lateral part [Fig. 2]

The CPF is a layer of dense fiber that enters the inferior border of the tarsus, merging with the anterior border of the inferior tarsal muscles. The head of the CPF splits open superiorly and inferiorly, wrapping around the inferior oblique muscle and meets anteriorly. As the inferior oblique muscle passes through the sheath of the inferior rectus, the fascia is thickened and forms the ocular suspensory ligament. A fibrous structure can be observed in the lower border of the ocular suspensory ligament, which is postseptal fat.

Midpupillary part [Fig. 3]

The inferior oblique muscle becomes smaller than the lateral part. All cases indicate that the fascia between the inferior oblique muscle and the inferior rectus muscle is thickened and forms the ocular suspensory ligament.

Medial part [Fig. 4]

The inferior oblique muscle becomes smaller than the midpupillary part. The fascia between the inferior oblique muscle and the inferior rectus muscle forms the ocular suspensory ligament, and the ocular suspensory ligament is thinner than the midpupillary part, connecting the inferior oblique muscle and the inferior rectus muscle. The upper border of the ocular suspensory ligament is loose tissue, and the lower border is postseptal fat.

Coronal plane: Six hemifaces observed [Fig. 5]

Lateral part

A single layer of dense fiber can be observed around the outer border of the inferior part of the eyeball, which is the ocular suspensory ligament in the coronal plane. The inferior side of the end of the inferior oblique muscle can be observed in the postseptal fat, issuing dense fibers and traveling laterally and inferiorly. The lateral check ligaments are connected to the ocular suspensory ligament.

Midpupillary part

A single layer of dense fiber can be observed around the outer border of the inferior part of the eyeball, which is the ocular suspensory ligament in the coronal plane. It can be observed in the postseptal fat. The inferior oblique muscle tissues from the lower part of the eyeball and travels medially and inferiorly.

Medial part

A single layer of dense fiber can be observed around the outer border of the inferior part of the eyeball. It can be observed in the postseptal fat, and the inferior oblique muscle continues to travel medially and inferiorly, stopping at the periorbiatal in a 7 o'clock direction. The medial check ligaments were connected to the ocular suspensory ligament.

Horizontal plane: Six hemifaces observed [Fig. 6]

Lateral part

A layer of dense fiber, which runs along the outer edge of the anterior side of the eyeball from the lateral canthal ligament, is the ocular suspensory ligament. There is also a layer of dense fiber running on the outer side of the ocular suspensory ligament, which merges with the ocular suspensory ligament. It is an arcuate expansion of the ocular suspensory ligament.

Midpupillary part

There is a layer of dense fiber on the outer edge of the anterior side of the eyeball, which is the ocular suspensory ligament. No branch was observed on the middle side of the ocular suspensory ligament.

Medial part

A layer of dense fiber, which runs along the outer edge of the anterior side of the eyeball and ends at the medial canthal ligament, is the ocular suspensory ligament. No branch was observed on the medial side of the ocular suspensory ligament.

Discussion

In this study, we first observed the ocular suspensory ligament using P45 plastination technology, which is suitable for the preservation and transparency of large and thin fault specimens. In the lower eyelid, the only ligament currently recognized is the ocular suspensory ligament.[5-8]

According to Whitnall, distal to the point where the inferior oblique crosses the inferior rectus, their blended sheaths send forwards thin lamellae, one of which follows the tendon of the latter muscle to help form the anterior part of the capsule of Tenon, while the other can be traced with difficulty into the lower eyelid. The sheaths of the muscles become confluent with each other by their adjacent margins immediately before the fusion with the capsule; the sheath of the inferior rectus is no exception to the rule, and the thickening of its sheath can be traced laterally and medially up to the sheaths of the lateral and medial recti, and since these are fixed to the orbital walls by expansions there is the formation of a continuous band about one-tenth of an inch thick beneath the globe, supporting it as in a hammock (ocular suspensory ligament of Lockwood) [Table 1].[9]

Fink and Duke-Elder thought that the ocular suspensory ligament is a blending of the sheaths of the inferior oblique and inferior rectus muscles extending upward and laterally to join the sheath of the lateral rectus and upwards and medially to join that of the medial rectus. There are also connections between this ligament and the intracapsular ligaments of the capsule of Tenon at the sites where the lateral and medial rectus muscles pass through the capsule.[10-13] The fibers at the lateral and medial ends of the ligament converge to their insertion, but, centrally, the fibers diverge to form a suspensory hammock which supports the eyeball. Fink believed that the check ligament of the inferior oblique may be regarded as part of the ocular suspensory ligament, and it throws out extensions into the lower lid where it is attached anteriorly to the orbital septum, the tarsal plate, and the cul-de-sac formed by the inferior fornix. Meanwhile, the fascicular bands pass from it to the skin, the intermuscular tissue of the lower lid, and the periorbital of the orbital margin and floor [Table 1].[14,15]

Beard and Hornblass described the ocular suspensory ligament as having few attachments to the medial and lateral retinacula, with major support coming from these retinacula. The fascial confluences that occur as the inferior oblique muscle passes through the capsulopalpebral head contribute significantly to the ocular suspensory ligament. Besides, the ocular suspensory ligament has contributions from the intermuscular septum, the inferior tarsal muscle, and the medial and lateral check ligaments [Table 1].[16]

Our observations concord well with studies by Whitnall, Fink, Duke-Elder, Beard, and Hornblass. The previous studies were mainly gross dissections and histologies of a part of an orbit. In our study, our serial cross-sections included whole hemifaces; consequently, we could observe the orbital bone as well as the soft tissue structure adjacent to the ocular suspensory ligament.

Hawes and Dorzbach *et al*. termed the fascial tissue anterior to the ocular suspensory ligament of Lockwood as "capsulopalpebral fascia."[17] In our observation, the head of the CPF split open superiorly and inferiorly, wrapping around the inferior oblique muscle and meeting anteriorly. As the inferior oblique muscle passes through the sheath of the inferior rectus, the fascia is thickened and forms the ocular suspensory ligament.

Kakizaki *et al*.[5] insisted that they observed the ocular suspensory ligament as having two layers; the other layer is not a muscle but does include some smooth muscle cells. However, other studies do not agree that these are unique structures in the lower eyelids.[18] Our research, both via Hornblass's book and our previous and present studies, have not revealed two layers of the ocular suspensory ligament. Kakizaki *et al*. also observed two branches of ocular suspensory ligaments (superior ligament and inferior ligament). However, we could not find these branches in any serial cross-sections [Table 2].

In this study, we identified the distribution of the lower eyelid ligaments. We observed the ocular suspensory

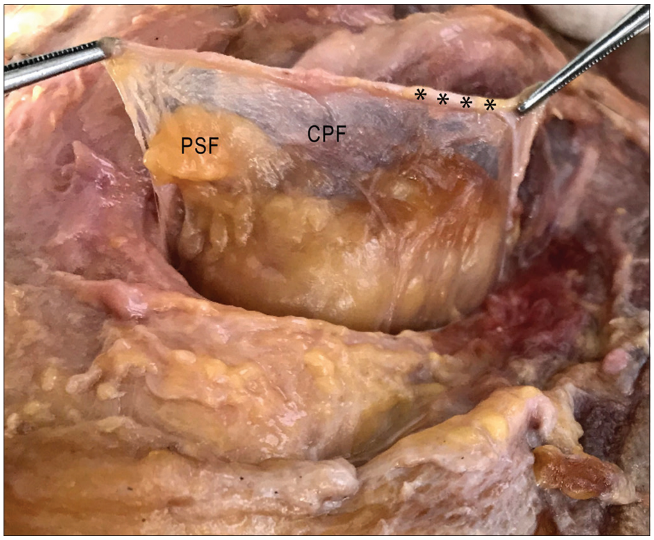

Figure 1: Gross anatomy of the lower eyelid. CPF: capsulopalpebral fascia, PSF: postseptal fat, asterisk: ocular suspensory ligament

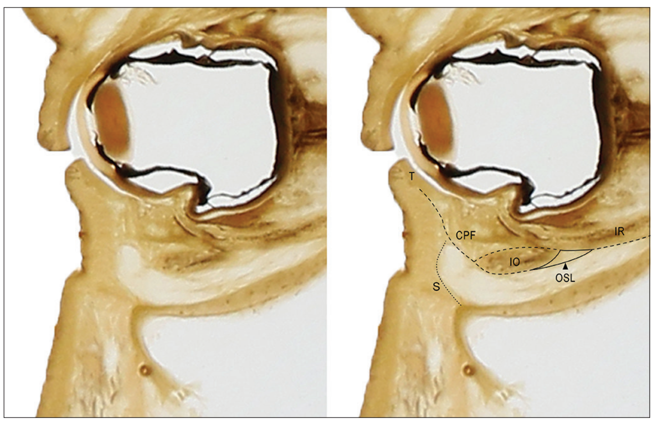

Figure 3: Sagittal section of the midpupillary part of the lower eyelid. CPF: capsulopalpebral fascia, IO: inferior oblique muscle, IR: inferior rectus muscle, OSL: ocular suspensory ligament, S: orbital septum, T: tarsal plate

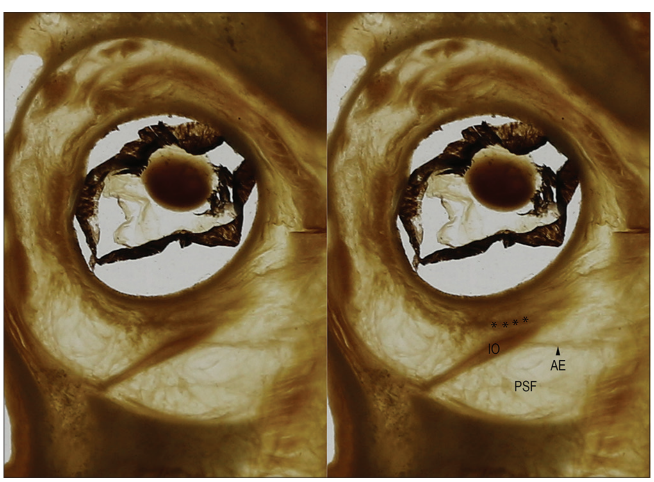

Figure 5: Coronal section of the lower eyelid. AE: arcuate expansion, IO: inferior oblique muscle, IR: inferior rectus muscle, PSF: postseptal fat, asterisk: ocular suspensory ligament

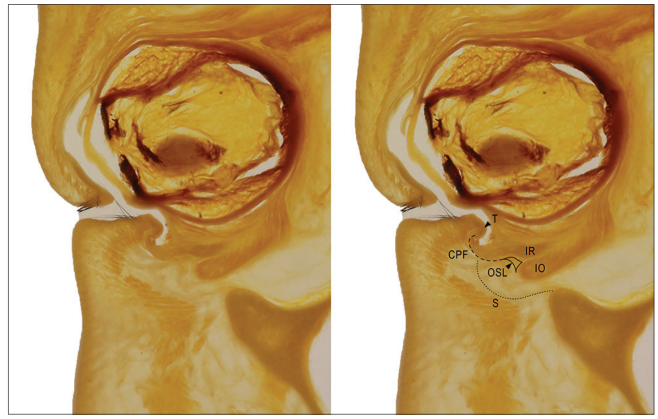

Figure 2: Sagittal section of the lateral part of the lower eyelid. CPF: capsulopalpebral fascia, IO: inferior oblique muscle, IR: inferior rectus muscle, OSL: ocular suspensory ligament, S: orbital septum, T: tarsal plate

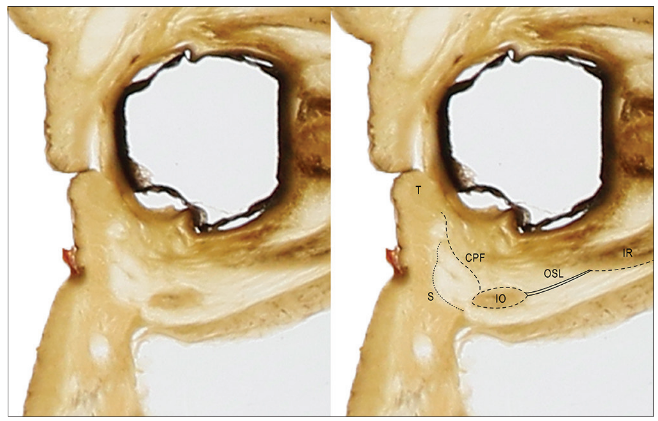

Figure 4: Sagittal section of the medial part of the lower eyelid. CPF: capsulopalpebral fascia, IO: inferior oblique muscle, IR: inferior rectus muscle, OSL: ocular suspensory ligament, S: orbital septum, T: tarsal plate

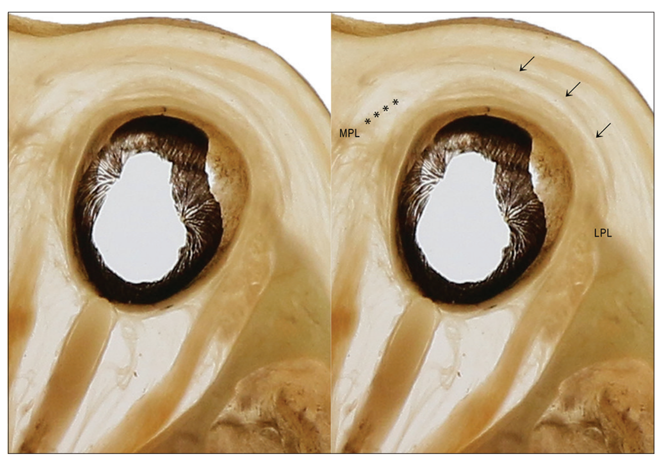

Figure 6: Horizontal section of the lower eyelid. LPL: lateral canthal ligaments, MPL: medial canthal ligament, asterisk: ocular suspensory ligament, arrow: arcuate expansion

ligament and arcuate expansion through P45 sheet plastination for the first time, laying the foundation for

Table 1: Structure and function of the ocular suspensory ligament

Author	Year	Tissue	Lateral	Medial	Extension	Function
Lockwood	1886	Band of fibrous tissue	Zygoma on the lateral side of the orbit	Lacrimal bone		
Whitnall	1932	Sheath of IR 1/10 inch thick	Sheath of LR to retinaculum oculi laterale	Sheath of MR to retinaculum oculi mediale	Fornix	Anchor the globe in position draw the fornix backward
Fink Duke-Elder	1948 1955	Blending of sheath of IO and IR	Upward and laterally to sheath of LR	Upward and medially to sheath of MR	Check lig., orbital septum, tarsal plate, cul-de-sac formix	Suspensory hammock
Beard Hornblass	1977 1988	Fascia of IO pass through CPH			Intermuscular septum, inferior tarsal muscle, medial and lateral check ligs.	
Present	2020	Multiple layers of dense fibers in front of IO	Lateral palpebral lig.	Medial palpebral lig.	CPF, Arcuate expansion	Hammock holding IO, IR, and eyeball

IO: inferior oblique muscle, IR: inferior rectus muscle, CPH: capsulopalpebral head, LR: lateral rectus, MR: medial rectus

Table 2: Ocular suspensory ligament (OSL)

	Kakizaki (2005)		Present (2020)
Branches	Superior ligament	Inferior ligament	No other branches found
Origin	From the posterior lacrimal crest at almost the same site as the OSL	From the posterior lacrimal crest	Medial palpebral lig.
Insertion	Periosteum of the lateral orbital rim	Posterior aspect of the IO, Not clear in the lateral margin of the CPF	Lateral palpebral lig.
Location	Superior to the OSL	Inferior to the OSL along the medial margin of the CPF	In front of IO

IO: inferior oblique muscle, CPF: capsulopalpebral fascia

further research. The ocular suspensory ligament should be approximated in place even after excision or fracture of the upper part of the jaw.

Conclusion

This study observed the ocular suspensory ligament and arcuate expansion through P45 sheet plastination for the first time and identified the distribution of the lower eyelid ligaments, laying the foundation for further research.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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