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Original Article

Gross and microscopic study of insertion of levator palpebrae superioris and its anatomical correlation in superior palpebral crease formation and its clinical relevance

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ABSTRACT

Background: To study insertion of LPS and correlate anatomically the formation of superior palpebral crease and its clinical relevance in section of Indian population.

Methods: Twenty-five human eyelids from cadavers ranging from 15–80 years were studied by dissection and histology.

Results: The levator aponeurosis traverses through interfascicular space of orbicularis oculi with twenty four inserting into subcutaneous tissue. Low septo aponeurotic sling or pre-aponeurotic fat was not observed. Lower one third tarsal insertion seen in twenty four specimens. Fibres become denser as it reaches the connective tissue anterior to tarsal plate. The crease was at different levels in relation to the tarsal insertion.

Conclusion: The aponeurosis insertion is either to the skin, the subcutaneous tissue or into the interfascicular space of the orbicularis. The present study reaffirms the insertion of LPS to the subcutaneous tissue of the eyelid with an extension reaching lower one third of the tarsal plate. The tissues in the anterior tarsal plate are closely packed but to act as a single complex to form a dynamic crease has not been confirmed in the study. No gross or histological difference was observed in the skin of the eyelid in the vicinity of the crease. The pattern of insertion of LPS aponeurosis plays a direct role in the formation of superior palpebral crease an important role in surgeries for Ptosis. The eyelids studied were of Indian origin with crease, and further comparative studies needs to be done for the precise anatomical explanation of single fold eyelids.

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Introduction

The extra ocular or extrinsic muscles of eyelid includes an elevator of the upper eyelid, the levator palpebrae superioris (LPS) which is a delaminated part of superior rectus (SR).¹ LPS is a thin triangular muscle which arises from inferior surface of lesser wing of sphenoid, above and anterior to optic canal and separated from it by the attachment of superior rectus. It has a short narrow tendon at its posterior attachment, broadens gradually as it passes anteriorly above the eyeball and ends in a wide aponeurosis. Some of its tendinous fibres pass straight into the upper eyelid to insert into the anterior surface of tarsus while rest of the fibres radiate, piercing the orbicularis oculi to reach the skin of upper eyelid.^{2,3} Close to the origin, the connective tissue coat of the adjoining surfaces of LPS and SR are fused and the fascia between them is attached to the superior conjunctival fornix, which is described as an additional attachment of LPS. The Superior Tarsal muscle (Muller's muscle), which arises as a smooth muscle ends in a fibrous extension to the upper margin of superior tarsus. This muscle, receives a sympathetic innervation from the superior cervical ganglion. The role of Superior Tarsal muscle is less clear, however in Horner's syndrome, it causes Ptosis of upper eyelid. The Muller's muscle has also been described as one of the attachments of LPS originating from the inferior surface of LPS,^{4,5} however, some studies do not mention of Superior Tarsal muscle as part of LPS,^{6,7} while some mention its insertion into the superior edge of tarsus.⁸

LPS has a levator action on the upper eyelid, its antagonist being palpebral part of orbicularis oculi.¹ LPS aponeurosis arising from the point of fusion of LPS and lower part of orbital septum (forming the septo aponeurotic sling), extends downwards into the lid and attaches with the tarsus and the connective tissue of the orbicularis in front of it.⁹ The aponeurosis has also been described to have three lamellae – superficial, middle and deep, getting inserted to front of superior tarsus, on the upper border of tarsus and superior fornix respectively.¹⁰ In addition, collagen fibres from the aponeurosis of LPS pass between bundles of orbicularis oculi, referred to as interfascicular space, and gets inserted into the skin of the upper eyelid.¹¹ Some other studies with similar interpretation justify their pretarsal part to attach into the skin of upper eyelid crease and sending septa in a diffuse pattern to the entire pretarsal portion of orbicularis¹² referred to as “superficial lamina”¹³ with a deeper lamina inserting on lower anterior surface of tarsal plate, while the deepest fibres reaching the superior conjunctival fornix as described previously.^{5,14} The LPS aponeurosis may also be absent in the pretarsal part by terminating in the superior tarsal border with some role in the elevation of the eyelid.^{15,16} Cheng et al¹⁷ studied the presence of the aponeurosis in the subcutaneous tissue of the upper eyelid leading to the formation of superior palpebral crease. Interestingly, eyelids without a crease have been postulated due to the inability of the aponeurosis to reach the skin due to excess preaponeurotic fat and lower fusion of orbital septum and levator aponeurosis^{16,18} while eyelids with crease may be due to strong adherence of pretarsal tissues.¹⁹ Recent studies on eyelid demonstrate the orbital septum fusion with the LPS fibres i.e. the septo aponeurotic sling above

the tarsal plate thus enabling the fibres to reach the pretarsal region of the eyelid²⁰ and the presence of crease attributed to thinner skin and orbicularis.²¹ The hypothesis of a strongly adherent pretarsal tissue has been refined to include a tight skin-orbicularis-tarsus complex which is lifted up as a single entity during opening of lid eye leading to formation of dynamic crease.²² The studies done till date on LPS aponeurosis and their varied patterns of insertions with reference to orbital septum, preaponeurotic fat, tight pretarsal tissue complexes, thinner skin etc exhibits multiplicity of hypotheses and findings. The studies have been done on double fold (with superior palpebral crease) eyelids of the west and single fold (without crease) eyelids of East Asians with peculiarities of each contributing to formation of crease with their associated clinical implications. However, there are no studies conducted amongst Indian population (which differs from the typical Western and East Asian race) to analyze and correlate the findings of the previous studies. The present aim of the study attempts to analyze the qualitative data obtained from cadaver eyelid by dissection and microscopic study of the levator insertion in a section of Indian population and to find the anatomical basis for the formation of superior palpebral crease. The study endeavours to prove that the superior palpebral crease is associated with the peculiar attachment of the LPS fibres into the upper eyelid and analyze the findings of different authors in an Indian context.

Material and methods

Twenty five human eyelids ranging from 15 years to 80 years were studied. The specimens were collected from cadavers obtained from a local hospital. Eyelid specimens were immediately kept in 10% formalin and dissection was carried out. All specimens were processed for paraffin section histological analysis. Orbits were cut in the midline parallel to the medial wall of the orbit by an electric saw (Fig. 1) and a macroscopic study of gross anatomy of LPS was done. The eyelids were extracted from the orbits and a strip was cut in the middle of the upper eyelid for histological study.

The specimens were then kept individually in vials containing 10% buffered formalin, decalcified (with 10% formic acid), and then prepared for sectioning through an Alcohol-Xylol-Paraffin wax sequence. Sections of 4–5 microns were examined with Haematoxylin and Eosin and Masson's trichrome stain (for collagen fibres).

Results

The observations are grouped under Cutaneous insertion, Tarsal insertion, Superior fornix and Muller's muscle. The data on insertion of the LPS has been recorded in a tabular form (Table 1).

The bony origin of LPS is not disputed. LPS continues as levator aponeurosis while the posterior lamella continues as the smooth muscle of Muller. The aponeurosis arises from the striated muscle of LPS. The preaponeurotic fat separates the levator aponeurosis from the connective tissue fibres of the orbital septum at upper levels (Fig. 2) and subsequently fuses

(Fig. 3). Fat has not been distinctly observed immediately anterior to lower part of tarsus (Fig. 4) and ends approximately at the level of eyelid crease. The aponeurosis of levator shows regular and linear arrangement unlike the fibres in orbital septum which presents no definite pattern and are comparatively irregular (Fig. 4). In twenty four specimens the fibres of levator penetrate the fibres of orbital septum at the point of fusion and enter the interfascicular space of orbicularis oculi (Fig. 5). In one specimen, the fibres of orbital septum were not visualized. The continuity of levator aponeurosis in interfascicular space is observed in all twenty five specimens (Fig. 5). The levator aponeurotic fibres become continuous with the subcutaneous tissue of the skin in twenty four eyelids (Figs. 6 and 7) while one showed the fibres in proximity to the dermis, suggesting a dermal insertion. Crease was below the level of superior tarsal border in sixteen specimens while it was at the level in nine specimens (Figs. 6 and 8). No change in the morphology of the skin was observed at the region of crease.

Twenty four specimens showed insertion at the lower one third of the tarsal plate (Fig. 4). The aponeurosis is loosely arranged in the upper tarsal plate and gradually becomes denser in the lower part. No direct relation was observed between the site of crease and aponeurotic fibres anterior to tarsal plate.

Twenty four specimens showed insertion to superior fornix from conjoined fascial sheath and Muller's muscle. The smooth muscle of Muller passes anterior to superior fornix, loses its muscular nature and finally inserted into the upper border of tarsal plate in twenty three specimens.

Discussion

The aponeurosis of LPS is important for the formation of eyelid crease and the anatomical basis for surgeries on eyelid crease and Ptosis. The LPS muscle continues as the levator aponeurosis below orbital septum¹⁹ to reach the eyelid skin,^{1,6,10,12,23} may insert into the interfascicular tissue of orbicularis oculi,^{5,24} may terminate in the subcutaneous tissue itself to form the crease,^{9,12,17} may insert into lower tarsus or superior border of tarsal plate at the level of crease^{2,14,25} or may not terminate at all at the tarsus. The single fold eyelid (without the crease) may be due to the inability of the levator fibres

reaching the sub dermal skin (subcutaneous tissue) due to the intervening pretarsal aponeurotic fat,¹⁹ thick subcutaneous fatty layer or a low septo aponeurotic sling.^{16,18} The crease may be formed by the strong adherence of the pretarsal tissue²⁰ while creaseless single fold eyelids may have no levator aponeurosis¹⁷ at all. Some recent studies have concluded the presence of levator aponeurosis in both kinds of eyelids, and the possibility of skin and orbicularis oculi in the single fold type to be much thinner, with an aponeurotic pull giving rise to a crease during lifting of the eyelid. The tight skin-orbicularis-tarsus complex has been hypothesized to form a dynamic crease while the complex being weak in single fold eyelids, thus unable to form a crease.²²

The classical surgeries for formation of crease in single fold eyelids and Ptosis has been performed based on "Supratarsal fixation" where the levator aponeurosis is anchored to the skin,²⁶ however in another procedure, portion of orbicularis oculi, connective tissue and pretarsal fat were removed and sutures put to eliminate the dead space, as the author postulated absence of aponeurosis insertion into tarsal plate.²⁷ In the modified approach the levator aponeurosis is sutured to the pretarsal skin eliminating the obstructive orbital septum and the preaponeurotic fat in between.¹⁸ Recently, fusion of orbicularis with levator aponeurosis with long lasting sutures for a dynamic crease has been performed.²² In Ptosis, the amount of levator aponeurosis to be resected is determined and after the required amount of levator is resected, the non-resected stump is advanced and sutured to the tarsus 5 mm or more below the upper tarsal border. This raises the upper eyelid and improves the ptotic eyelid¹².

In this study, the aponeurosis passed through orbital septum in twenty four eyelids and then traversed the space between the fascicles of orbicularis in twenty five specimens and subsequently blended with the connective tissue of the subcutaneous layer of the upper eyelid skin in twenty four eyelids. The study agrees with the insertion of the aponeurosis into the subcutaneous tissue with one specimen showing a possible insertion to the dermal layer of the pretarsal skin. The crease was observed to be below the superior border of tarsal plate in sixteen specimens and at the level of superior border of tarsal plate in nine specimens. The crease more or less coincided with the level at which the aponeurosis of levator blended with the orbital septum and its final termination in the subcutaneous layer of the eyelid in sixteen specimens. The significance of these findings suggests that the aponeurosis travels below the septo aponeurotic sling at the level of upper part of tarsal plate to contribute to formation of crease which anatomically has always been below these levels. In the present study, no single fold (Oriental) eyelid could be studied and none of the eyelids showed a low septo aponeurotic sling or an excess of fat in the preaponeurotic or the subcutaneous layer. As all the specimens studied had a crease and levator aponeurosis, it suggests strong possibility of formation of crease due to levator aponeurosis and is in agreement with similar studies. There was no gross morphological differences observed in the thickness of skin in the region of the crease or of the orbicularis oculi thus the observations are contrary to those of Kakizaki²¹. This study observed a prominent lower one third tarsal insertion in twenty four specimens. This finding is important for corrective surgeries of Ptosis. As the

Fig. 1 – Sagittal section of orbit cut in midline. LPS (levator palpebrae superioris) and SR (superior rectus) can be seen.

Table 1 – Table showing the pattern of LPS insertion. OO – Orbicularis oculi, OS – Orbital septum.

Specimen no.	Crease: below/above/at level with supra tarsal border of tarsal plate	Insertion of LPS					
		Aponeurotic part				Smooth part (Muller's muscle)	
		Insertion via OO interfascicular space reaching subcutaneous tissue, relation of LPS & OS	Insertion in skin (epidermis/dermis)	Tarsal plate (lower 1/3rd)	Superior fornix via collagenous fibres	Tarsal plate	
						Superior border	Anterior surface
1	Below	Yes. OS penetrated	Yes	No	Yes	Yes	Yes
2	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
3	Below	Yes. OS penetrated	No	Yes	Yes	Yes	yes
4	At level	Yes. OS penetrated	No	Yes	Yes	Not seen	Not seen
5	Below	Yes. OS penetrated	No	Yes	Not seen	Yes	Not seen
6	Below	Yes. OS penetrated by LPS at superior border of TP at level of crease	No	Yes	Yes	Yes	Not seen
7	At level	Yes. OS penetrated by LPS fibres	No	Yes.	Yes	Yes	Yes, few fibres
8	At level	Fibres seen through OO Interfascicular space only	No	Yes	Yes	No seen	Not seen
9	Below	Yes, OS penetrated	No	Yes	Yes	Yes	Not seen
10	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
11	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Yes
12	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
13	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
14	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
15	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
16	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
17	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
18	Below	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
19	At level	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
20	At level	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
21	Below	Yes. OS penetrated at level of crease.	No	Yes	Yes	Yes	Not seen
22	At level	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
23	At level.	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
24	At level	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen
25	At level	Yes. OS penetrated	No	Yes	Yes	Yes	Not seen

Fig. 2 – (×40). Showing Orbicularis Oculi (oo), Orbital Septum (os), Levator Aponeurosis (apo) and preaponeurotic fat (fat). Horizontal view of eyelid. Masson Trichrome stain. Aponeurosis piercing orbital septum in the upper right corner of the field. The aponeurosis and the orbital septum are separated by fat. The fibres seen in the inter fascicular space (ifs) of orbicularis oculi.

Fig. 4 – (×100). Showing aponeurosis (apo) and tarsal plate (tp). Horizontal view of the eyelids, right corner of the field being the superior part. Aponeurotic fibres are loosely arranged in the upper part of tarsus and gets inserted in the lower part of tarsus. Connective tissue is tightly arranged in the lower tarsus. Fat has not been distinctly observed immediately anterior to lower part of tarsus.

Fig. 3 – (×40). Showing orbicularis oculi (oo), orbital septum (os), levator aponeurosis (apo), smooth muscle of Muller (m) and conjunctiva (c). Horizontal view of the eyelid. Orbital septum blends with levator aponeurosis at the lower left corner of the field.

Summary and conclusions

There is a wide variation in the cutaneous insertion of the LPS aponeurosis. The insertion of the aponeurosis may be either to the skin, the subcutaneous tissue, into the inter fascicular connective tissue of the orbicularis oculi or the tarsal plate. Recent studies however show a subcutaneous insertion for the formation of upper eyelid crease. This study is in agreement with the recent studies and all the specimens exhibited crease with LPS aponeurosis, however as it was unable to confirm the presence of preaponeurotic fat or a low lying septo aponeurotic sling in any of the specimens, as it may have a role in single fold eyelids. No morphological change in the skin in the vicinity of the crease was observed in any of the specimens.

However, as the eyelids studied were of double fold eyelids of Indian origin, further comparative studies need to be done

aponeurotic fibres coursed anterior to the tarsal plate it became denser in the restricted space of the anterior part of the eyelid, thus confirming the observations of Saonanon.²² However, the dense skin-orbicularis-tarsus as the anatomical basis for crease formation could not be ascertained, but could have some role. No direct relation was observed between the site of crease and the type of arrangement of the connective tissue and aponeurotic fibres anterior to tarsal plate. The study could not ascertain, whether insertion to superior fornix had any clinical importance. The Muller muscle inserts to superior border of tarsal plate in twenty three eyelids and no extension to anterior surface of tarsal plate, as observed in twenty one eyelids. The insertion is not too extensive to elevate the upper eyelid in isolation or formation of crease.

Fig. 5 – (×100). Showing Orbicularis oculi (oo), Levator aponeurosis (apo) and inter fascicular space (ifs). Aponeurosis in inter fascicular space subsequently reaches the subcutaneous space.

Fig. 6 – (×40). Showing tarsal plate (tp), levator aponeurosis (apo) and crease (cr). Aponeurosis (apo) reaches subcutaneous tissue (sc) near the crease after traversing through interfascicular space (ifs).

for the precise anatomical explanation of single fold eyelids in similar settings. Limitation of procuring a creaseless eye has not gone unnoticed which otherwise would have made the observations more substantial.

Fig. 7 – (×200). Showing skin (sk) and subcutaneous tissue (sc). Levator aponeurosis (apo) lies in subcutaneous tissue but does not insert into skin. Levator aponeurosis fibres are more regularly arranged than the connective tissue of subcutaneous layer.

Fig. 8 – (×40). Showing subcutaneous tissue (sc), orbicularis oculi (oo), levator aponeurosis (apo) and upper eyelid crease (cr). Levator aponeurosis blending with the subcutaneous tissue in the area of upper eyelid crease.

Conflicts of interest

The authors have none to declare.

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