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دراسة هستولوجية وهستوكيمائية للجفن في الجاموس المصرى

عبد الله حفني ، أحمد حسن ، عزيزة سليم ، محمد نبيل

أجرى البحث على ٢٠ جفنا جمعت من الجاموس المصرى :

- يمتاز جلد الجفن العلوى والسفلي بأنه رقيق ويصبح سميكا وداكنا عند حافة كل جفن .
- يغطى السطح الخلفي للجفن بنسيج طلائي انتقالي به خلايا كأسية .
- تم تمييز أربعة أنواع من الخلايا الدهنية ، وأنواع عده من نهايات الأعصاب .
- درست نشاطات انزيم الفوسفاتيز القاعدى والحامضي والسكسنيك دى هيدروجيناز .

Dept. of Anatomy and Histology,
Faculty of Veterinary Medicine, Assiut University,
Head of Dept. Prof. Dr. M.H. Badawi.

HISTOLOGICAL AND HISTOCHEMICAL STUDIES ON THE EYELIDS OF THE BUFFALOES IN EGYPT

(Wit One Table and 17 Figures)

By
A. HIFNY, A.H.S. HASSAN, AZIZA, A.A. SELIM and M.N. MOUSTAFA
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SUMMARY

Histological details of the eyelids from 20 adult Egyptian buffaloes were observed. The epidermis is relatively thin in both the superior and inferior eyelids, but becomes thicker and heavily pigmented on the free margin between the cilia and the tarsal gland openings. The palpebral surface is lined with transitional epithelium containing goblet cells. Morphologically, four types of sebaceous glands could be recognized. Dermoepidermal free nerve endings, free nerve endings surround the hair follicles and krause's end bulbs were demonstrated. The distribution and activities of alkaline phosphatase, acid phosphatase and succinic dehydrogenase were investigated.

INTRODUCTION

The available data on the histological structure of the eyelids of buffaloes are scanty and incomplete. PRASAD and SINHA (1979) studied the sebaceous glands and their modification in the eyelids of the Indian buffaloes. LAST (1961) recognized three types of sebaceous glands in the palpebral skin of mammals. Little information on the eyelids of cats was revealed by STRICKLAND and CALHOUN (1963) and American goats by SAR and CALHOUN (1966). So, the present investigation was performed to characterize the histological and histochemical features of the eyelids of buffaloes in Egypt.

MATERIAL and METHODS

The specimens used in the present study, were obtained from 20 adult Egyptian buffaloes of both sexes. Specimens were fixed in Helly's fluid, dehydrated, cleared, embedded in paraffin and sectioned at 8-10 Um thick. Representative specimens were fixed in 10% formol saline and cryostat sections of about 15-20 Um thickness were prepared for some histochemical studies. Fresh frozen cryostat sections (10-15 Um) from representative samples of the examined specimens were prepared for some enzymatic histochemical examinations.

The prepared sections were stained with: Harris's hematoxylin and eosin, Weighert's elastic tissue and Van Gieson's connective tissue stains, Bielschowsky's silver stain, Periodic acid Schiff (PAS) technique, Sudan black- B stain, Gomoril's method for alkaline phosphatase, Gomori's method for acid phosphatase, Nitro-blue tetrazolium method for succinic dehydrogenase.

RESULTS

The skin covering the eyelids is thin, corrugated, and contains fine hairs (Fig. 1). The epidermis measures about 63 Um in thickness (Fig. 8). The stratum granulosum shows dense PAS reaction while the upper cell layers of stratum spinosum demonstrates a moderate reaction.The stratum corneum shows a sudanophilic reaction. The dermis of the skin of the eyelid measures about 2055 Um in thickness. The dermal papillae are small, short and measure about 27 Um in depth. Dermoepidermal free nerve endings appear bulbous in shape and run tortuouslly parallel to the dermoepidermal junction (Fig. 6 A). The reticular layer of the dermis consists of large coarse interwoven bundles of collagenous and elastic fibers which spray out towards the epidermis (Fig. 3). The dermis contains hair follicles, sweat and sebaceous glands. Tactile hairs and cilia are numerous in the superior than in the inferior eyelid.

The hair follicles are less dense, short and accompanied with Arrector pili muscles, sweat and sebaceous glands. The sebaceous glands are multilobulated and nearly encircling the hair follice (Fig. 2). The tactile hairs are mostly observed near the cilia (Fig. 4) associated with poorly developed sebaceous glands, and Arrector pili muscle but lack sweat glands. Myelinated nerve fibers pierce the connective tissue sheath of the tactile hair follicle and run along the connective tissue trabeculae, crossing the blood sinus and give off free nerve endings surround the hair follicle at a level below the sebaceous glands (Fig. 6 D). The cilia are arranged in 2 or 3 rows and are more numerous in the superior than in the inferior eyelid, and disappear at the medial and lateral commissures. The follicle of each cilium is associated with an apocrine tubular gland and two sebaceous glands (Fig. 5 A). The apocrine tubular glands (glands of Moll) are less coiled spiral tubular glands about 155 Um in diameter (Fig. 5 C). The secretory epithelium is simple columnar and surrounded by large myoephithelial cells and rests upon a thick basement membrane. Clear cells with large rounded nuclei are observed inbetween the secretory cells. The secretory portion becomes narrow toward the surface and emerges directly into the duct. There is no transition between the secretory epithelium and that lining the duct. The duct runs parallel to the hair follicle and opens into it above the level of the sebaceous glands. The initial portion of the duct is wide where it joins the secretory portion, then narrows abruptly where it is lined with cuboidal and myoepithelial cells. Each cilium is associated with two large, lobulated sebaceous glands (glands of Zeis). Each gland opens by a separate duct on each side of the follicle. Mitotic figures are usually observed within the peripheral cells of these glands (Fig. 5 B). Free nerve endings are also recognized around the follicles of the cilia (Fig. 6 E). Circular striated and smooth muscle fibers are situated deeply in skin of the eyelids. The tarsal glands are arranged in a linear series and are embedded in the tarsal plate. These glands are of the multilobular sebaceous type (Fig. 7). Each lobule opens by a short duct into a large central duct. Both ducts are lined with stratified squamous epithelium, which aquires a heavy stratum corneum at the opening onto the posterior margin of the eyelid. The tarsal glands are larger and more numerous in the superior than in the inferior eyelid. However, near the palpebral commissure they are few and smaller in size. Large nerve fibers give off numerous small retiform branches around the tarsal glands (Fig. 6 C). These glands react positivey to PAS technique. Coarse purple granules are demonstrated within the basal cell layer, however fine ones are scattered within the other cells of these glands. Also the ducts show an intensive reaction towards PAS technique.

At the free portion of the eyelid (between the cilia and the openings of the tarsal glands), the epidermis increases in thickness (about 99 Um) and demonstrates numerous melanin pigments (Fig. 9). The dermis is devoid of both hair follicles and their associated glands.

It is composed mainly of circularly disposed collagenous bundles and contains circularly arranged skeletal muscle bundles within its deeper portion. Krause's end bulbs are also observed (Fig. 6 B). Posterior to the openings of the tarsal glands, the covering epithelium (about 98 Um) is of the stratified squamous noncornified type (Fig. 10) with numerous pigment cells at its basal layer (Fig. 11). This stratified squamous non-cornified epithelium is reflected posteriorly to cover the posterior margin of the free portion of the eyelid, as well as a short distance of the palpebral surface (Fig. 12). Then it changes into transitional epithelium with goblet cells (about 80 Um) which covers the rest of the posterior surface. Goblet cells are gradually increased in number towards the conjunctival fornix forming intraepithelial glands (Fig. 13). The activities of the alkaline phosphatase, acid phosphatase and succinic dehydrogenase enzymes in the eyelids are given in table (1) and Figs. (14, 15, 16 & 17).

DISCUSSION

The epidermis of the eyelids is relatively thin in both the superior and inferior eyelids of buffaloes. However, in the eyelids of horses, the epidermis contains long branched, heavily pigmented epidermal pegs (TALUKDAR et al. 1972). The present investigation revealed that the epidermis of the eyelids of buffaloes contains a relatively small amount of non-glycogen. No alkaline phosphatase reaction could be demonstrated. Moderate acid phosphatase reaction in the stratum granulosum and even in the stratum corneum were observed. The reaction of succinic dehydrogenase was moderate within the stratum basale and slight within the lower layers of stratum spinosum. Similar results were demonstrated within the normal epidermis whether in man (MONTAGNA and PARAKKAI, 1974), beagle dogs (CONROY and GREEN, 1975) and pigs (MEYER and NEURAND, 1976).

Posterior to the openings of the tarsal glands, the epidermis changes into non-cornified stratified squamous epithelium, then reflected on the posterior margin of the eyelids for further support and protection of the free margin. This epithelium continues with the transitional epithelium of the palpebral conjunctiva. The latter epithelium adaptes the palpebral conjunctiva which is subjected to continuous stretching and relaxation. The presence of the goblet cells within the transitional epithelium provide a smooth gliding movement between the palpebral and bulbar conjuctiva. In domestic animals the palpebral conjunctiva is lined with either stratified squamous, stratified cuboidal, stratified columnar, transitional or pseudostratified epithelium with goblet cells (CALHOUN and STINSON, 1976). However, in man it is lined with stratified columnar epithelium among which goblet cells are situated (HAM and CORMACK, 1979). The dermis contains abundant elastic fibers with a fine architectural distribution particularly in the papillary layer. The sebaceous glands were observed in the eyelids of buffaloes either associating the hair follicles, the cilia, and the tactile hairs or as tarsal glands. These findings are similar to those described by PRASAD and SINHA (1979). However, LAST (1961) did not mention any sebaceous glands associating the tactile hair and suggested that only 3 locations of sebaceous glands are recognized in the palpebral skin of mammals. The follicles of the tactile hairs observed near the cilia marked by a blood filled sinus, traversed by trabeculae, were similar to those of ungulates and coinside to those mentioned by TALUKDAR et al. (1972) and CALHOUN and STINSON (1976) in horses, ruminants and pigs. These tactile follicles lacked the sinus pad described in carnivores by STRICKLAND and CALHOUN (1963) and MULLER and KIRK (1969). The cilia were found to be numerous

in the superior than in the inferior eyelids of buffaloes. Similar results were obtained by CALHOUN and STINSON (1976) in ruminants and horses.

The secretory portions of the tarsal glands are small, few, embedded in the tarsal plate and open onto the posterior portion of the free margin of the eyelids. Similar observations were recorded in the Indian buffaloes (PRASAD and SINHA, 1979). MONTAGNA and ELLIS (1959 b) as well as MONTAGNA and FORD (1969).

The present investigation revealed a network of nerve fibers and abundant smooth muscle fibers around the tarsal glands. So, it can be suggested that these muscle bundles may play a role in discharging the secretion of these glands. However, MONTAGNA and ELLIS (1959 b) as well as MONTAGNA and FORD (1969) demonstrated a complex network of nerves encircling the Meibomian glands in man and in some animals, but did not explain probable function of these nerves. They also stated that, no muscles could be observed around the Meibomian glands other than those of the palpebrae. Similar to what was observed in man, intraepidermal nerve fibers were observed in the epidermis of the eyelids of buffaloes (MONTAGNA and FORD, 1969). At the inner portion of the papillary layer of the dermis of the buffalo, numerous nerve fibers are oriented horizontally to form a network of different densities depending on its location simulating what was mentioned by TAMPONI (1940), MILLERet al. (1960) and WINKELMANN (1960 a).

In the present investigation, the dermoepidermal nerve fibers, the free nerve endings which surround the hair folicle and the krause's end bulbs represent the cutaneous sensory receptors in the eyelids and are greatly similar to that mentioned by WINKELMANN (1959 b , 1960 a,b), MONTAGNA and PARRKKAL (1974) HAM and CORMACK (1979).

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Table (1): Distribution of alkaline phosphatase (AKP), acid phosphatase (AP) and sucinic dehydrogenase (SDH) enzymes in the eyelids of buffalo. (Figs. 14, 15, 16, 17).

Outer skin	AKP	AP	SDH	Free part	AKP	AP	SDH	Lower part of posterior surface	AKP	AP	SDH	Palpebral Conjunctiva	AKP	AP	SDH
Epidermis															
Stratum basale	+	-	+++	Stratum basale	-	-	++	Basal cell layer	-	-	++	Transitional epithelium	+++	+	+++
Stratum spinosum	-	-	++	Stratum spinosum	-	-	+	Polyhedral cell-layer	-	-	+				
Stratum granulosum	-	+	+	Stratum granulosum	+	++	+								
Stratum corneum	-	++	-	Stratum corneum	+	++	-	Squamous cell layer	+	++	+				
Apocrine glands:				Tarsal gland:											
Secretory cell	+	++	+++	Peripheral cells	++	++	++								
Myoepithelium	+++	-	++	Central cells	+	++	+								
Sebaceous glands:				Duct epith.	++	++	+								
Peripheral cells	++	++	++												
Central cells	+	++	+												

(+++). Strong reaction. (++) Moderate reaction (+) Weak reaction (-) No visible reaction.

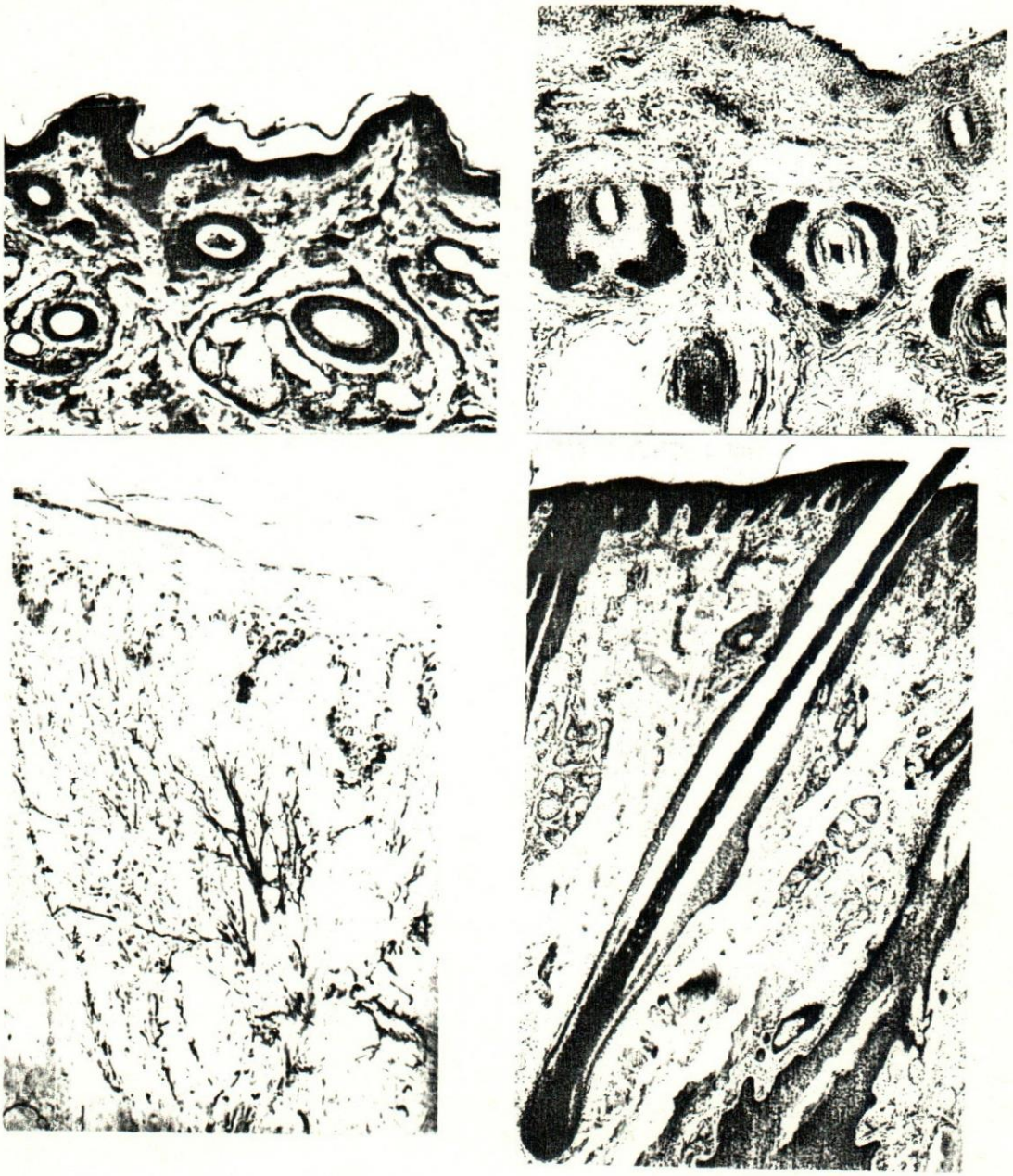

Fig. (1): Vertical section in the skin covering the eyelids. (H&E. X43).

Fig. (2): Vertical section in the skin. (Sudan black. X 43).

Fig. (3): The elastic fibers in the skin of eyelids. (Weigert's elastic tissue stain X 100).

Fig. (4): Vertical section in the skin at the rostral margin of the free part of the eyelid.

Fig. (5): The cilia of the eyelid. A, the follicle of the cilia (H&E. X 100). B, glands of Zeis with clear mitotic figure (arrow) (H&E. X 160). C, glands of Moll; secretory cells (S), myoepithelium (m), clear cells (C) and duct cells (d). (H&E. X 240).

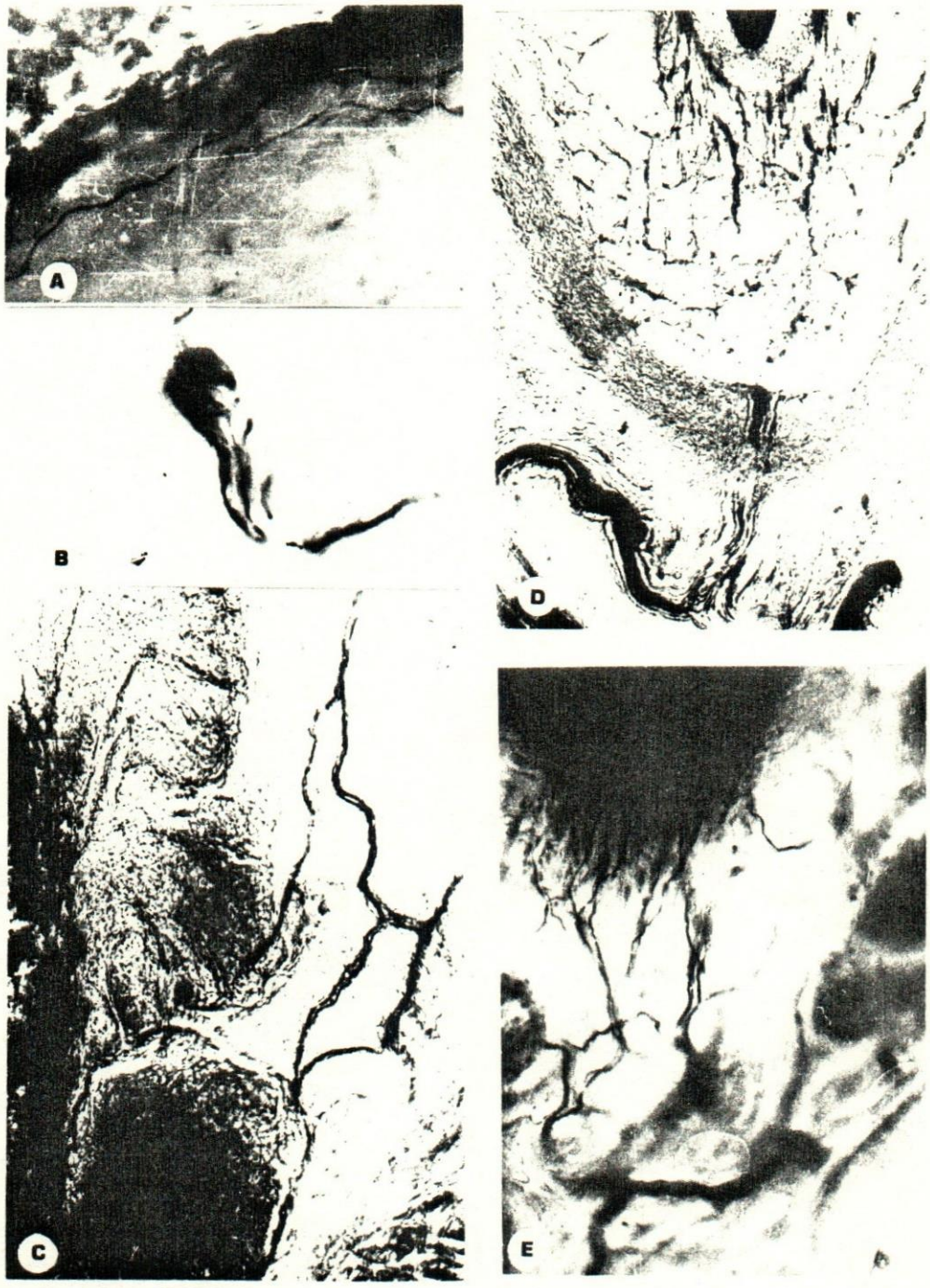

Fig. (6): Nerves and nerve endings in the eyelids. A., dermoepidermal free nerve fiber X 250. B, Krause's end bulb X 160. C, myelinated nerve fibers around the tarsal gland, X160. D, innervation of the tactile hair X 100. E, innervation of the folliele of the cilia, X 160. (Bielschowsky's stain).

Fig. (7): Longitudinal section in the tarsal gland. (H&E. X 43).

Fig. (8): The epidermis of the skin covering the eyelid. (H & E. X 160).

Fig. (9): The epidermis covering the free portion of the eyelid. (H&E. X 160).

Fig. (10): The epithelium covering the posterior margin of the eyelid. (H&E. X 160).

Fig. (11): Pigment cells in the basal cell layer. (H&E. X 240).

Fig. (12): The area of junction between the epithelium lining the posterior margin of the eyelid (P) and palpebral conjunctiva (C). (H&E. X 43).

Fig. (13): The epithelium of the palpebral conjunctiva, containing goblet cells (g). (PAS. X 100).

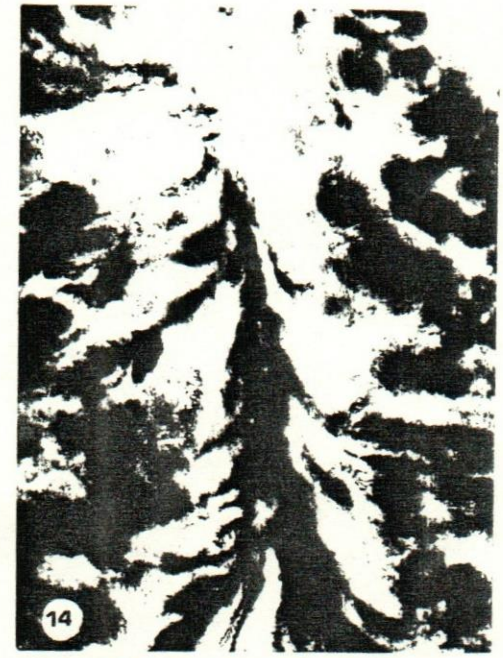

Fig. (14): Alkaline phosphatase activity in the tarsal gland. (Gomori's method. X 43).

Fig. (15): Acid phosphatase activity in the tarsal gland. (Gomori's stain. X 43).

Fig. (16): Succinic dehydrogenase activity in the tarsal gland. (N-BT method X 160).

Fig. (17): Acid phosphatase activity in the skin covering the eyelid. (Gomori's method. X 43).