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HAZARDOUS EFFECT OF SOME INDUSTRIAL POLLUTANT ON TILAPIA NILOTICA (OREOCHROMS NILOTICUS)

(With One Table and 17 Figures)

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التأثيرات الضارة لبعض المخلفات الصناعية على أسماك بلطي النيل

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ان مشكلة تلوث المياه في العلم تعد من أخطر مشاكل العصر لا سيما بالملوثات الكيميائية والناشئة من تواجد مصبات المصانع في الانهار الهامة · ويعتبر تلوث نهر النيل بمخلفات مصانع السوبر فوسفات بمنقباد بأسيوط واحدا من تلك المصانع والتي ثبتت كافة الدراسات السابقة مدى تلوث تلك المياه بالفلورين والكبريت ، وتعد الثروة السمكية واحدة من أهم مصادر البروتين الحيواني وسمك البلطي من أهمها ونظرا لقلة المراجع العلمية عن الآثار الباثولوجية المترتبة على معيشة تلك الاسماك في المياه الملوثه والشكوى المستمرة لصيادي هذه المنطقة من نفوق اعداد وفيرة من هذه الاسماك بصفة دائمة مما أوجب معه القيام بهذا البحث والذي استهدف دراسة الآثار الضارة للفلورين والكبريت على الاعضاء المختلفة لاسماك بلطي النيل · ومن ثم تم أخذ اسماك من هذه المنطقة (٣٠) كما تم اخذ اسماك في نفس الحجم والوزن من منطقة أسيوط والتي تبعد عشرة كيلو مترات في الاتجاه العكسي لتيار المياه كضابط للتجربة وعمل الصفة التشريحية واخذ عينات من القلب والكبد والكلى والعدة والامعاء والمخ والخياشيم وتثبيتها في محلول ١٠٪ فورمالين متعادل ثم صبغها بالهيماتوكسيلين وايوسين بعد تقطيعها · كما تم حساب النسب المئويه لكرات الدم البيضاء ·وقد أظهرت النتائج الميكروسكوبية مظاهر عديدة أهمها الخلل الواضح في معظم الأعضاء ويصل أحياناً إلى موت خلوي منتشر خاصة في الكبد والكلى والعدة مع إرتشاحات خلوية في الكلى بالنيوتروفيل ، كما لوحظ إحتقان شديد في الطحال مع قلة عدد الخلايا البيضاء به وتحطيم مراكز الميلانوماكروفاج كما لوحظ زيادة في إفرازات الميوسين بالخياشيم مع موت خلوي بها ووجود تفاعلات خلوية واضحة · وقد ترسبت أجسام في داخل الكلى كما لوحظ إلتهابات بعضلة القلب · أما كرات الدم البيضاء فقد إنخفضت معدلات الخلايا الليمفاوية وإرتفعت المتعادلة منها · مما سبق يتضح خطورة هذه الملوثات على سائر أعضاء أجسام الكائنات الحية المائية مع زيادة إحتمالات تناقص وإنخفاض الجهاز المناعي الخاص بها الأمر الذي يدعونا للتساؤل لأي مدى يتأثر الجهاز المناعي في الإنسان أو الحيوان في هذه المنطقة والذي يستحق مزيداً من البحث ·

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SUMMARY

Organs and blood from Tilabia nilotica fish suspected to be intoxicated with fluorine and sulphur were studied clinically and pathologically. Differential Leucocytic count revealed moderate degree of Lymphopenia and neutrophilia. Pathological changes were observed in some parenchymatous organs including toxic hepatitis, Ulcerative and necrotic gastritis, tubular nephrosis, degenerative myocarditis and bronchitis. Degenerative and destructive lesions were also observed in the spleen. In conclusion the study evaluated the hazardous effect of industrial intoxication on Tilabia nilotica by fluorine and sulphur emitting from factory of fertilizer. Moreover it has been established that fluorosis and sulphurosis constitutes a serious hazardous to the health of Tilabia nilotica fish as it causes destruction of immune system with a subsequent lowering of the resistance of the affected fish. Further more lesions observed in the parenchymatous organs of the intoxicated fish was too sever to kill it.

INTRODUCTION

The acidification of fresh waters by acidic pollutants is a primary environmental problem. Studies throughout the world have documented the loss of fish populations in lakes and rivers acidified by atmospheric fall out of pollutants (GLASS, et al. 1979 and SCHOFIELD, 1976) and acidic run off from emittion of H_2SO_4 from super- phosphate factory (IBRAHIM, 1992) at river Nile water.

In supsequent studies, IBRAHIM, et al. (1985) recorded the contamination of the environment at the area surrounding the supper-phosphate factory at Manquabad, Assiut, Egypt by both fluorine and sulphur. The previous studies indicated that the highest degree of water pollution of the river Nile up to 4 Kilometers to the north and 1.5 Kilometers to the south of the factory. This pollution occured as a result of the drainge of wast water of the factory (mainly diluted sulphuric acid and hydrogen fluoride) directly into the river Nile.

Although there have been many reports of pathological lesions caused by pollutants in fish, there have been few systemic descriptions of the toxic syndromes associated with exposure to pollutants. Such descriptions are clearly desirable as an aid to understanding the mechanism of action of pollutants, and could be diagnostic aids in the investigation of fish mortalities where the accidental or negligent dischange of pollutant is suspected (DREWETT and ABEL, 1983).

The scanty litrature on the pathological lesion on Nile fish by industrial polluntant (fluorine and inorganic sulphate) in addition to presistance complaint of fishermen, regarding natural fish moribund in river Nile at the area surrounding the superphosphate factory in Assiut. The present study was carried out to investigated the toxic effect of fluorine and sulphur in Tilapia nilotica which considered one of the important types of fish widely consumed by Egyptian people.

MATERIAL and METHODS

Thirty Tilapia nilotica exposed to fluorine and sulphur weighing from 200 to 250 gms collected from the River Nile in El-Tawabiya area, which is located 0.5 Km north the super-phosphate factory. Twenty Tilapia nilotica of the same weight were collected from the River Nile at Assiut City, located 10 Km south to the factory and used as control.

Blood samples from the investigated and control fish was made and stained with Giemsa for differential leucocytic count (COLES, 1980).

Tissue samples from different organs including heart, liver, kidneys, stomach, intestine, brain and gills were fixed in 10% neutral buffer formaline, washed, dehydrated and embeded in soft parafine. The tissues were blocked, sectioned at 5 um and then stained with hematoxyline and eosin sectioned with examined and phytographed. The obtained data were statistically analysed according to KALTON (1967).

RESULTS

The obvious clinical symptoms appeared on fish were recognised as dulness and restlessness, congestion of the gills, died within 2 minutes after leaving water.

Gross examination of fish revealed a certain degree of congestion of the internal organs including heart, liver, kidney, stomach and gills. Other organs were grossly unchanged. Examination of the bony tissue revealed no specefic changes.

Microscopic examination of the liver revealed hepatic cell degeneration, which may reach up to necrosis. Necrobiotic changes of hepatic cells are focal or sometimes diffuse (Fig. 1). Necrotic changes sometimes extend to involve the pancreatic island of the liver (Fig. 2). Necrobiotic changes in most cases were usually associated with mononuclear cells reactions, which was usually observed arround the central vein in the middle of hepatic lobules (Fig. 3). In some cases along with the hepatic cells degeneration which was manifested by cytoplasmic vaculation and nuclear pyknosis. A crystal like material consist of central nucleus from which radiate a short process which ended in a dense basophilic layer surrounding the nucleus with its radiating short process. These crystals were observed between the hepatic cells.

Microscopic change in the kidneys were observed in the tubular epithelium. The epithelium lining tubules were individulized, sometimes showed necrobiotic and or desqumative changes. Mild leucocytic infiltration of the interstitial tissue was constantly observed by neutrophil and mononuclear cells (Fig. 5). Depletion of haematobiotic center in the kidneys was observed in all cases examined (Fig. 6). Also, deposition of crystal like material was observed. Deposition of these material lead to degeneration and necrosis of the tubular epithelium, which sometimes followed by regeneration of this epithelium.

Microscopic examination of the stomach revealed extensive necrotic changes which commonly associated with mononuclear cellular reactions in the gastric mucosa

(Fig. 7). Errossions were continuosly seen in the mucosa of the stomach. The cellular reactions in stomach was consisted of mononuclear cells. In some cases this cellular reaction extend deep into the submucosa (Fig. 8).

The myocardium showed small focal areas of myocardial degeneration, which in most cases were accompined by mononuclear cell reaction (Fig. 9).

Microscopic examination of the brain revealed a mild degree of vaculation which was focal and seen mostly in the grey matter of the cerberum (Fig. 10).

The spleen in most cases showed severe congestion of the red pulb, however, the white pulb showed a decreased lymphocytic populations (Fig. 11). A severe exhaustions of melanomacrophage cell (in the melanomacrophages centers) were also constant findings (Fig. 12).

Microscopically the bronchial tissue showed extensive oedema which was associated with hemorrhages in some areas. The blood vessels were severely dilated and congested. Mononuclear cells infiltration varies among different cases from light to heavy (Fig. 13). The epithelium of the secondary gill lamaellae showed increased amount of mucin producing cells (Fig. 14). In some cases the epithelium of the secondary gill lamellae showed desqumative and necrotic changes. In such cases the blood vesseles of the primary gill lamallae were congested with very mild leucocytic reactions (Fig. 15). Howevere, we must point to the fact that, extensive inflammatory reaction manifested by hyperemia and mononuclear cells infiltration in most cases was allways observed in the bronchial tissue (Fig. 16).

Differential leucocytic count was done for all cases under investigation and the results were represented in table 1. Differential leucocytic count of all cases revealed a moderate degree of lymphopenia and neutrophilia (Fig. 17).

DISCUSSION and CONCLUSION

Gross and histopathological examination of Tilapia nilotica fish organs revealed pathognomic changes for intoxication.

Necrobiotic changes of the liver and kidney observed in examined cases were of chronic nature and constant in all cases under investigation. A similar lesions were observed in the liver and kidney of cattle and sheep affected with fluorosis and sulphurosis and reported by many authers (KESSABI, et al. 1986; OBEL and ERNE, 1971 and SHUPE, 1960).

Chronic toxic hepatitis and chronic tubular nephritis observed in the liver and kidneys of Tilapia Nilotica fish were apperently due to hypoxia. This openion may be supported by the hypothesis that fish *subjected to acid stress may experience a decrease in arterial* oxygen level and respond to this hypoxaemia by increasing the oxygen carring capacity of the *circulating blood (VAALA and* MITCHALL, 1970 and *VAALA, 1972). Also, the* recorded degenerative changes in kidney and liver, reported by BLOOD and HENDERSON (1983), in severe cases of fluorine toxicity support the indicated results.

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A crystal like material with a special morphological structure was observed in the liver and kidney. In our openion this material is suspected to be due to deposition of sulphur containing compounds. Moreover, it has been observed that this lead to degeneration and necrosis of the surrounding tissues.

Errossive and necrotic gastritis seen in most examined cases was apperently due to exposure of fish to polluted water. However, the cellular reaction was secondary response to the alternative changes. Also, focal areas of degenerative myocardities together with the mild degree of demylination of the brain were observed as a result of the toxic effect of these pollutants, which was previously detected in high levels in fish blood (IBRAHIM, 1992). Severe gastroentritis, degenerative changes in kidney, liver, heart muscle, adrenal glands and central nervous system have been reported in severe cases of fluoride toxicity by BLOOD and HENDERSON (1986).

Exhustion of the lymphocytic population as well as degeneration of the most cellular element of the melanomacrophages centers of the spleen may indicate immune depression. It also point to the chronic exhustive effect of the pollutant on the immune system. The latter concept was proved by the lymphocytopenia and neutrophilia observed in differential leucocytic count of the most cases examined.

Oedema, hemorrhage and degenerative changes observed in the gills and bronchial tissues were due to the effect of hypoxia. WESTFALL (1945) concluded that the lethal effect of acid water to goldfish was directly attributable to reduced oxygen permeability of the gills caused by excess formation of mucus. DAYE and GARSIDE (1976) showed that damage to the epithelium of the gill lamelae also occured at pH levels below 5.2 or above 9.0 in conjunction with hypertrophy of the mucous cell. Exposure to low pH can also decrease the blood pH (PACKER and DUNSON, 1970) which might in turn cause a decreased oxygen carrying capacity of the blood.

Depopulation of hemopiotic centers of the kidneys might be partial responsable for neutrophilia constantly observed in most cases under investigations.

In conclusion the study evaluated the effect of industrial intoxication on Tilapia Nilotica fish by fluorine and sulphur emitting into river Nile from fertelizer factory. Furthermore the severity of histopathological lesion (which is too severe to cause death of the most affected fish) together with immune suppresive effect of the pollutant confirm the pollution hazard on the health condition of aquatic life.

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Table 1: Differential leucocytic percentage in the blood of investigated fish

Samples	Number	Mean ± S.E				
		Lymphocyte	Neutrophil	Basophil	Eosinophil	Monocyte
Investigated fish	30	68.50±3.45**	28.50±3.20**	1.75±0.20	0.75±0.10	0.50±0.05
Control	20	88.25±5.60	9.25±1.20	1.50±0.30	0.50±0.10	0.50±0.15

** : Significant at P / 0.01

S.E. : Standard error.

LIST OF THE FIGURE

Fig. 1: Diffuse necrosis and necrobiosis of the hepatic cells (25X) H & E.

Fig. 2: Necrotic changes sometime extend to involve the pancreatic islands of the liver H & E (16X).

Fig. 3: Mononucleo cellular reaction arround the central zone surrounded with area of necrobiotic changes of the hepatic cells H & E (25X).

Fig. 4: Showed a crystaline material in the hepatic tissue H & E (25X).

Fig. 5: The epithelium of the tubules showed necrobiotic and desqumative changes H & E (16X).

Fig. 6: Depletion of hematopiotic center in the kidney H & E (10X).

Fig. 7: Necrotic changes of the gastric mucosa associated with cellular reactions H & E (10X).

Fig. 8: Showed mononuclear cells infiltration in the submucosa H & E (10X).

Fig. 9: Focal areas of myocardial degeneration H & E (25X).

Fig. 10: Brain showed mild degree of vaculation in the grey matter (10X).

Fig. 11: Shown congestion of the red pulb and decreased population of lymphycytosis in the white pulp H & E (10X).

Fig. 12: Spleen showed exhaustion of melanomacrophage cells in melano macrophage centers H & E (10X).

Fig. 13: Oedema, haemorrhage and leucocytic infiltration in H & E (25X).

Fig. 14: Increased amount of mucin producing cells in the secondary gill lamallae H & E (10X).

Fig. 15: Showed desqumative and necondary gill lamellae H & E (10X).

Fig. 16: Hyperemia and mononuclear cells reactions of the bronchial tissues H & E (16X).

Fig. 17: Blood smear showed one lymphoid and one neutrophil cells, Gimsa stain (100X).

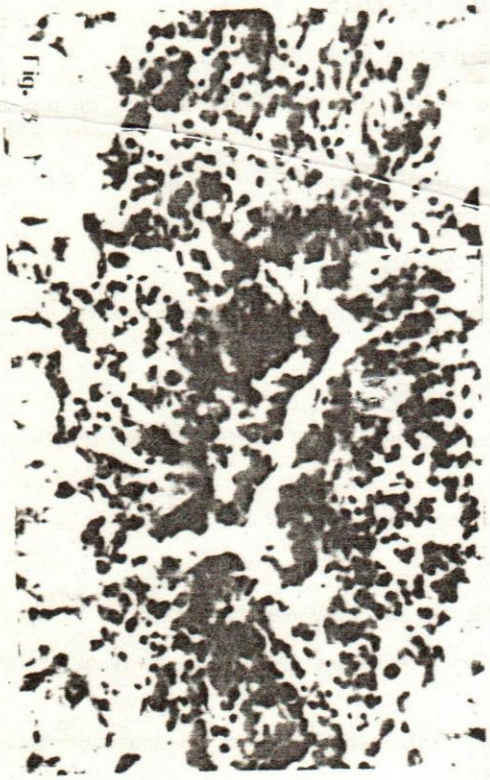

Fig. 3

Fig. 1

Fig. 2

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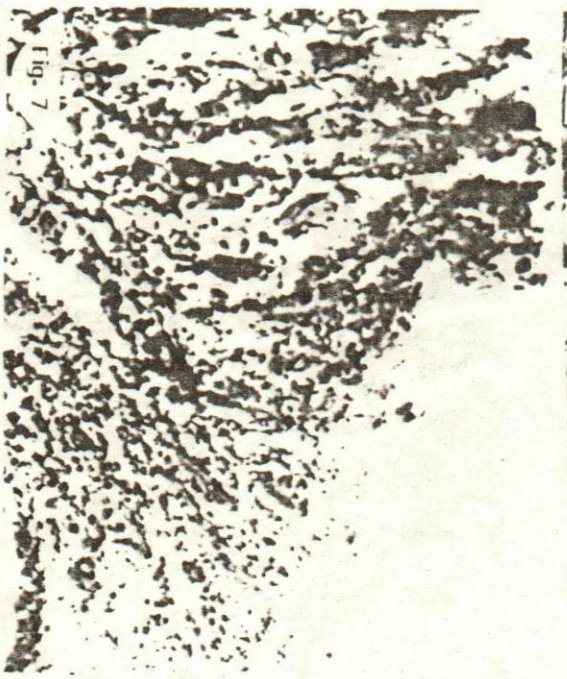

Fig. 7

Fig. 5

Fig. 8

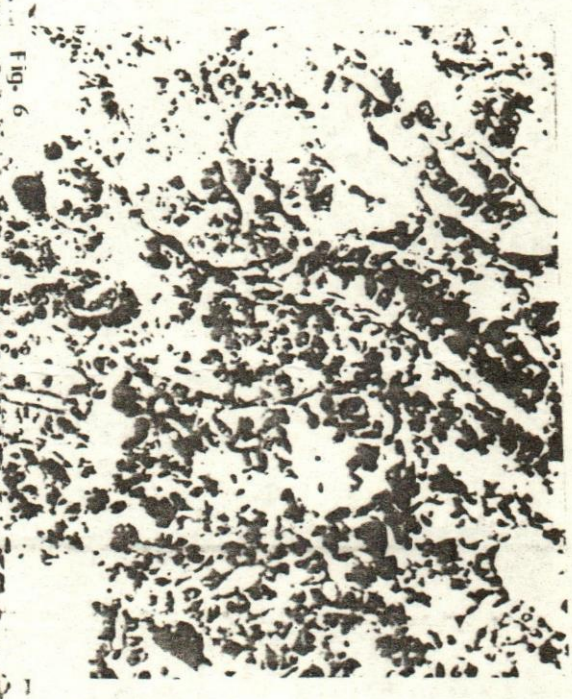

Fig. 6

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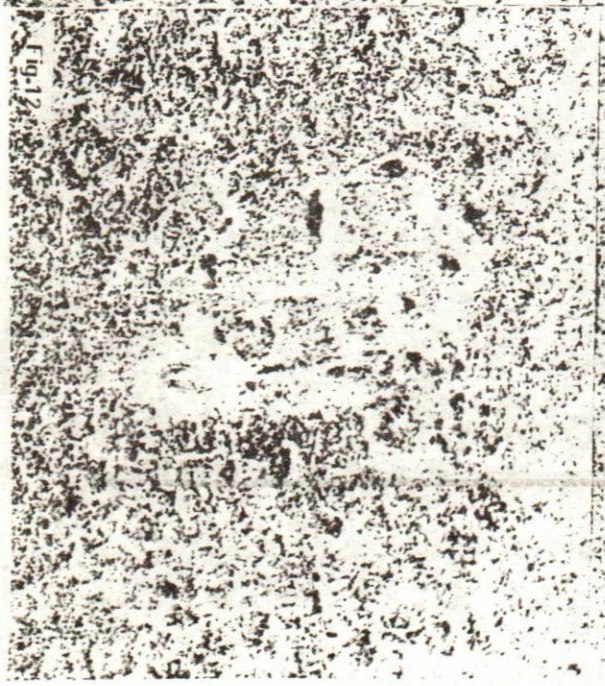
Fig. 9



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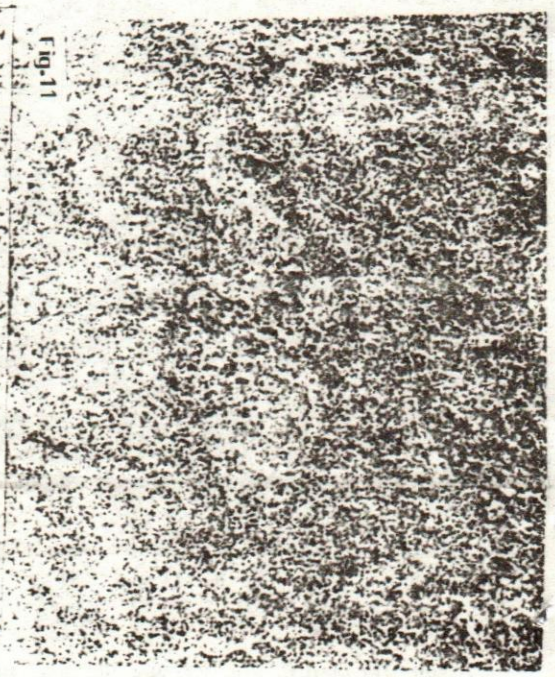
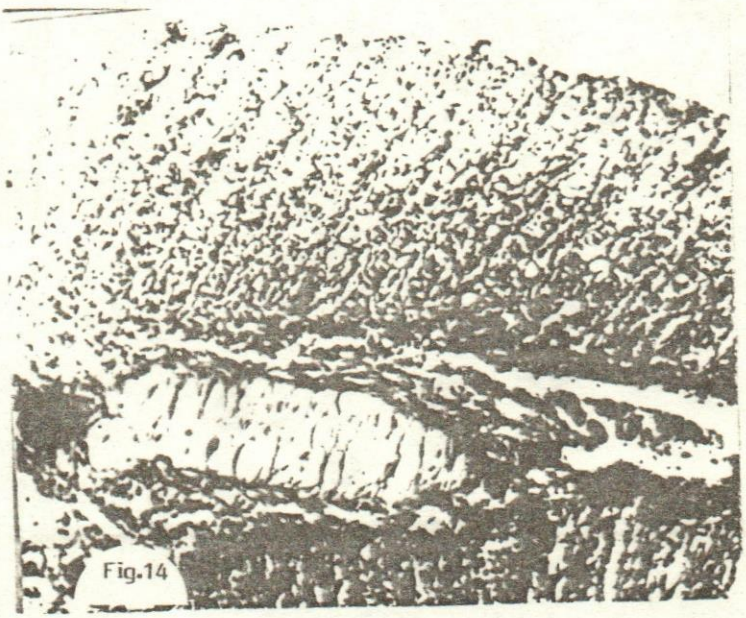


Fig. 11

Fig. 13



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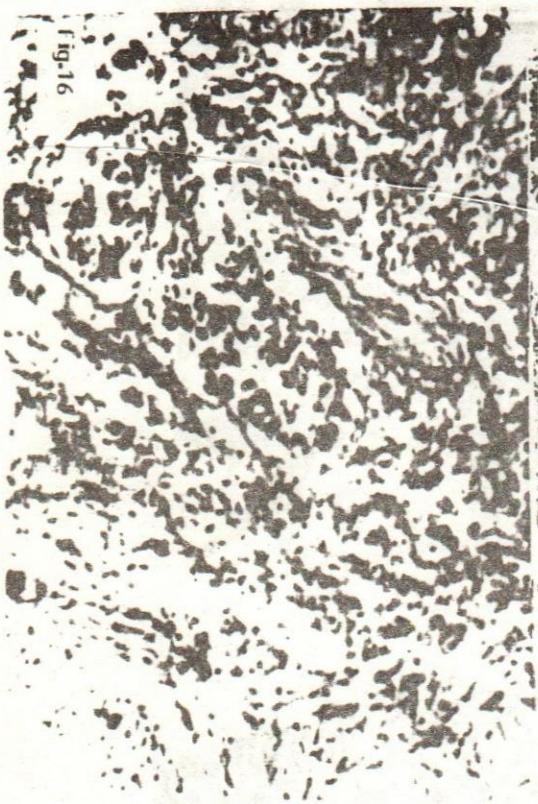

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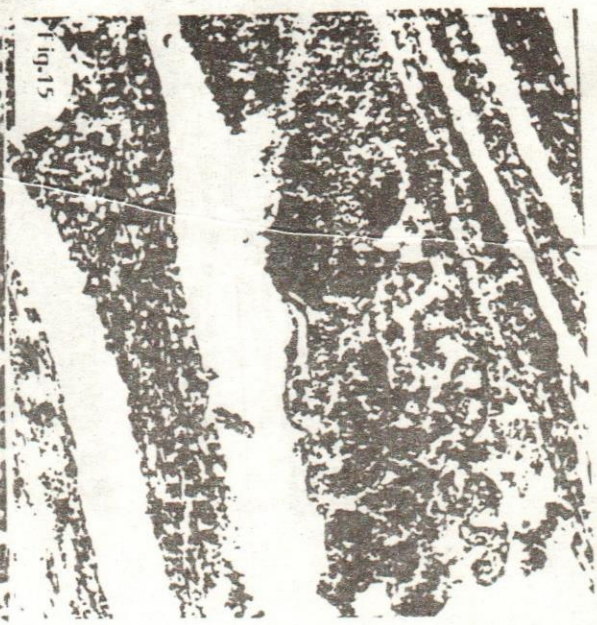

Fig.15

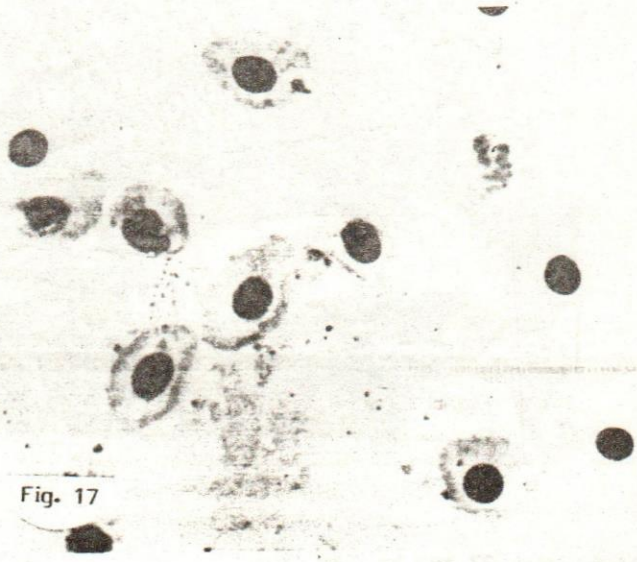

Fig. 17