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دراسة خلايا الليف من الأبقار الصحيحـــة اكلينيكيا بواسطة الميكروسكوب الالكتروني

محمود عبد الظاهر

عند دراسة الخليـا المكونة لليف من الابقار الصحيحة اكلينيكيا وجد أنهـا تتكون في الغالب من الخـلايا الليمفاوية . وهـذه الخلايا الليمفاوية يمكـن تقسيمها الى أربعة أنواع :

- خلايا الليمفاوية الصـــــغيرة .
- الخلايا الليمفاوية ذات الحجم المتوسط .
- الخلايا الليمفاوية الكبـــــــيرة .
- الخلايا الغـــير ممـــــــيزة .

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ULTRASTRUCTURAL STUDY OF LYMPHOCYTE FROM LYMPH OF THE THORACIC DUCT OF CLINICALLY NORMAL CATTLE

(With 5 Figs.)

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SUMMARY

Ultrastructural study of lymph trom the thoracic duct of clinically healthy cattle showed that the cellular Population of lymph consists mainly of Four types of cells of lymphiod series. Small medium sized and large lymphocytes as well as undifferentited cells. This classifection was based upon. The diameter of the cells, the nuclear cytoplosmic ratio and the ultrastructural morphology.

INTRODUCTION

There are few reports on ustrstructural study of lymphocytes of lymph nodes and spleen from cattle affected with leukosis (SORANSON and THELLEN 1963). UEBORSCHAR (1963-1966) describe certian ultrastructural feature of lymphocytes from haemopatic organs. Also there were few works on the ultrastructural morphology of blood leucocytes of clinically healthy cattle (D.E. BLECHOR MAN; A.N. SMURNOV; B.D. EGROUA 1968).

However analysis of literature revealed that there was no reports concerning the ultrasturctural morphology of lymphocyte of lymph of clinically healthy cattle.

This work was desighned to study the ultrastructural morphology of lymphocytes from clinically normal cattle.

MATERIAL and METHODS

This work was done at the department of pathology Moscow Veterinary akademy. Sex clinically healthy cows were used for this study. All cows were exposed for haemtological examination. The amount of leucocytes in the blood ranges from 5,000 to 12,000/mm^3 of blood. Directly after surgical apening of the tharocic duct and establishment of artificial fistula, 200 ml of lymph was obtained. Heparin was added as an anticoagulant at a rate of 1 mg/1 ml of lymph. As the lymph contian a certian amount of red blood cells. Distelled water was added to lymph in ratio of 1:2 per volume, then mixing and Nacl 4% solution was added in ratio 1:1 per volume to prevent swelling and rapture of lymphocytes. This lead to haemolysis of erythrocytes. The lymph was centrifugated at 1000 R/m for 5 m. The supernatant fluid was removed and the precipetate was then fixed in 2.5% solution of glutaraldhyde for 25-30 m. The disck of cell was then divided into small pellets, washed in phosphate buffer with PH. 7.4 several times. The pellets were post fixed in osmic acid, then washed with phosphate buffer 3-5 times. Dehydration was done in serial dilution of ethyl alcohol and acetone. The pelets were embrignated in mixture of epion and araldhyde. The pellets were then embeded in gelatine capsule contian the mixture of epion and araldhyde. Ultrathin

sections were obtianed on ultratome L K B. 8802 A using glass knife. The ultrathin section was stianed by urinal acetate and potassium citrate (method of REYNOLDS 1963).

RESULTS and DISCUSSION

Electron microscopic examination of lymph from thoracic duct of clinically healthy cattle showed that the cellular population of lymph consisted mainly of cells of lymhiod series with few amount of granulocytes, leucocytes and red blood corpuscles. The per centage was 99.4% lymphoid cells 0.2% neutrophil, 0.3% eosinophiles and 0.1% Monocytes. The majority of cell were those of lymphoid series. According to the size of the cells, ratio between the size of the nucleus and cytoplasm and the ultrastructural morphology, four types of cells could be distingushed.

Small lymphocyte:

This was the predominent type of lymphocyte observed. The diameter was 5-6 U km. The ratio between the nucleus and cytoplasm was high ranging from 5:1 or more. The nucleus was rouned or oval or may be indentended, usually centrally situated, coarse chromatine located at the periphary in either isolated masses or in continous band. This masses or bands somtimes extend towards the midle part of the nucleus. The nucleolus was absent. The fine chromatin was usually situated in the center and somtime extend to the periphary. Somtimes the coarse chrometine was diffusely distributed all over the nucleus with small amount of fine chromatine. The perinuclear area was distinct. Nuclear pores were not promenant. The cytoplasm was represented by narrow band, surrounding the nucleus. The organalles in the cytoplasm were very scanty (Fig. 1). Only free ribsomes could be demonstrated in the cytoplasm of this cells. Mitochonderia, smooth, rough endoplasmic reticulum and Golgi apparatus were not observed. Vacuoles of pinocytosis were observed.

Medium sized lymphocyte:

The diameter of this cell ranging from 6-8 U. This cell was observed in fewer amount than the small lymphocyte. The ratio between the size of the nucleus and cytoplasm was lesser than those of small lymphocyte (average 3.5:1). The nucleus was not centrally situated. The coarse chromatine was arranged in the form of clamps which usually observed at the periphary of the nucleus and somtimes extend towards the middle portion. The coarse chromatine occupied a small part of the nucleus. The nucleus was well developed. The perinuclear space was clear, while the nuclear pores were not observed. The cytoplasm appeared as a relatively broad band surrounding the nucleus. being broadest at the end where the cytoplasmic organelles were condensed (Fig. 2). The cytoplasm of this cell was relatively rich in organelles. Free ribsomes were relatively abundant. Mitochonderia with condensed crusts were also found. Its number was raning from 3-5 and were rounded in shape. Golgi apparatus were seen they were represented by electron clear small rounded or oval vacuoles. smooth and rough endoplasmic reticulum also not observed.

Large lymphocyte:

The amount of these cells was fewer than the preceeding two types. The diameter ranges from 8-10 U with nucleo-cytoplasmic ratio of 1.5:1. The nucleus in this cell was eccentrically situated being rounded or oval in shape. The coarse chromatine was either diffusly distributed in the nucleus or arranged in clumps at the periphary of the nucleus. Fine chromatine was abundant in amount and found usually in the middle part of the nucleus. The perinuclear area was

not distinct, while the nuclear pores were clear. The cytoplasm was more or less abundant and was very rich in organelles (Fig. 3). The number of mitochonderia ranges from 8-10. They were oval rounded, elongated or polyhydral. The mitochonderial crest were usually of electron clear type. Golgi apparatus was well developed. It was represented by electron clear vacoules or tubules arranged side by side. Few ill developed granular endoplasmic reticulum were also seen. The amount of free ribsome was larger if compared with the preceeding types. Sometimes ribsomes unite to form rossete shape polysomes. Vacoules of pinocytosis were few.

Undifferentated or blast cells:

The diameter of this cell was about 10 U or larger. The nucleocytoplasmic ratio was 1.9:1. The nucleous was oval or rounded occupying a large part of the cell. Coarse chromatine was arranged on the periphary near the nuclear membrane in form of small or large masses. The amount of fine chromatine was larger than that of coarse one and occupying the largest area of the nucleous being concentrated in the middle part. (Fig. 4). The nucleolus were constantly seen. The perinuclear space was clear and the nuclear pores were distinct. The cytoplasm was of mild electron density. It contians scanty amount of free ribsomes which may coalase to form rossete shape polysomes. The amount of mitochonderia (3-5) with electron dense crests were present. Golgi apparatus was not well developed. It was represented by few numbers of small electron clear vacoules. multivascular bodies were also observed. Single ill developed endoplasmic reticulum were detected while granular endoplasmic reticulium were absent.

Our classification of lymphocyte according to diameter was in agrement with the calssifilation given by Cimonin and AGACIN (1968). These authers classify lymphocytes from the blood of healthy and leucotic cattle into small, medium sized and large lymphocytes using light microscope. FEDROV (1967) stated that blood lymphocytes from healthy person according to the diameter and ultrstructural morphology could be divided into 4 types. Large lymphocytes, small light & dark lymphocytes and lymphoplasmocytes. He added that small light lymphocytes constitute 75% of all types. According to our result small lymphocytes could not be distinguished into dark and light type, lympho-plasmocytes were not observed in our material. However small lymphocytes constitute the main type of lymphoid cells of the lymph from thoracic duct of clinically healthy cattle as in healthy human blood. The ultrastructural morophology of large & small lymphocytes described by this auther are inagreement with the ultrastructural morphology encountered in our material with exception that the stated that lymphocyte exhibit 1-2 mitochonderia in its cytoplasm. This statment was given by BLEKRMAN B.E. et al. 1968. A general description of fine structure of lymphocyte from spleen and lymph node of clinically healthy cattle was given by FEDROV, N.A. et al. 1973, but they did not classify lymphocytes into different types.

From our result we can concluded that the cellular pouulation of lymph obtianed from the thoracic duct of clinically healthy cattle consists mainly of four types of cells of lymphoid series. small lymphocyte, medium sized lymphocyte, large lymphocyte and undifferentiated cells. The predominant type was the small lymphocyte. The ultrastructural morphology was described in details. This result may be of great help in comparative study of lymphocyte of lymph of the thoracic duct of cattle with lympholeukosis and lymphosarcoma in the preceeding work.

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DESCRIPTION OF FIGURES

Fig. (1 a,b): Ultrathin section of small lymphocyte with a cytoplasm cantion only few ribsomes.

Fig. (2): Ultrathin section of medium sized lymphocyte with cytoplasmic organelles condensed at the broadest end.

Fig. (3): Ultrathin section of large lymphocyte with cytoplasm rich in organelles.

Fig. (4): Ultrathin section of undifferintiated cell with large amount of fine chromatine occupying the middle part of the nucleus.

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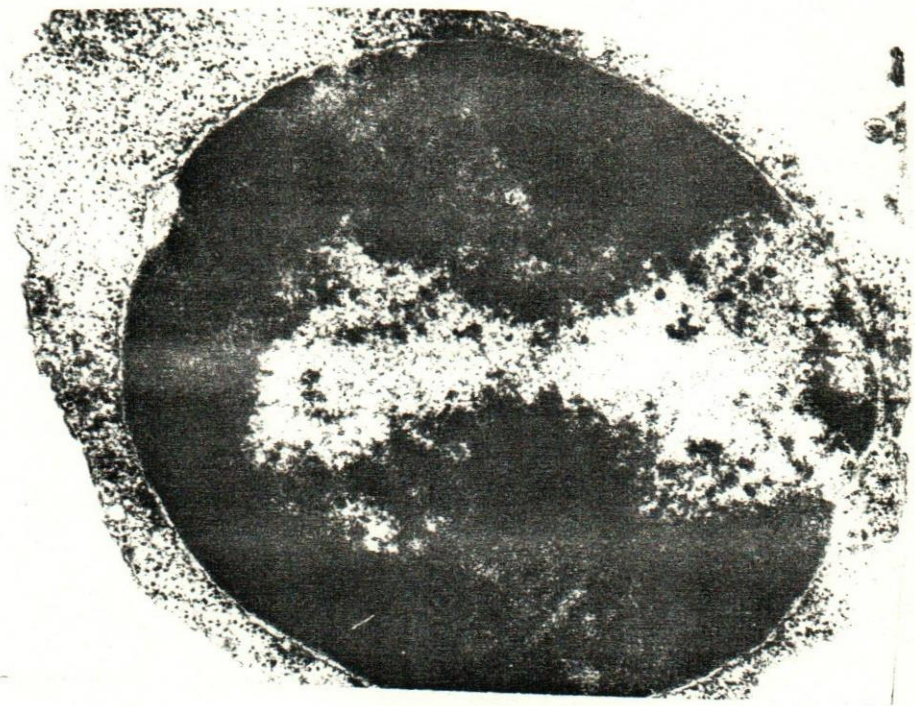

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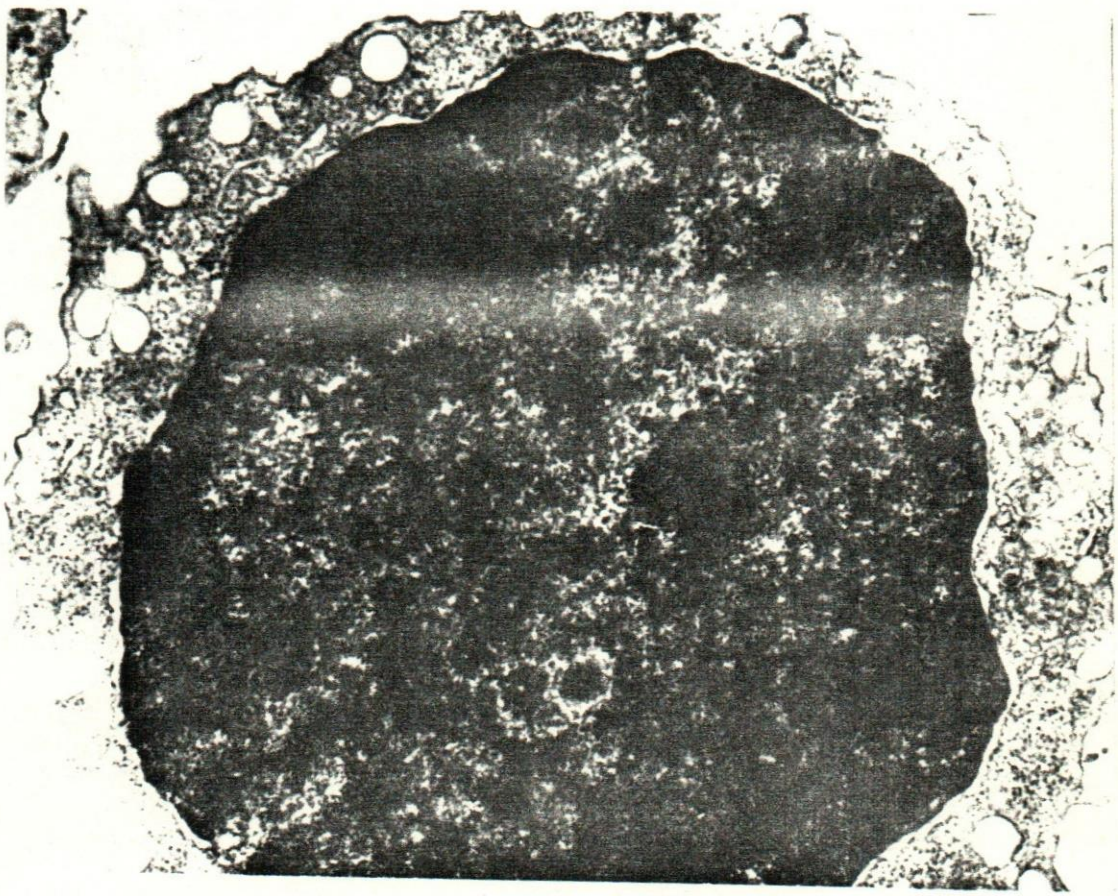

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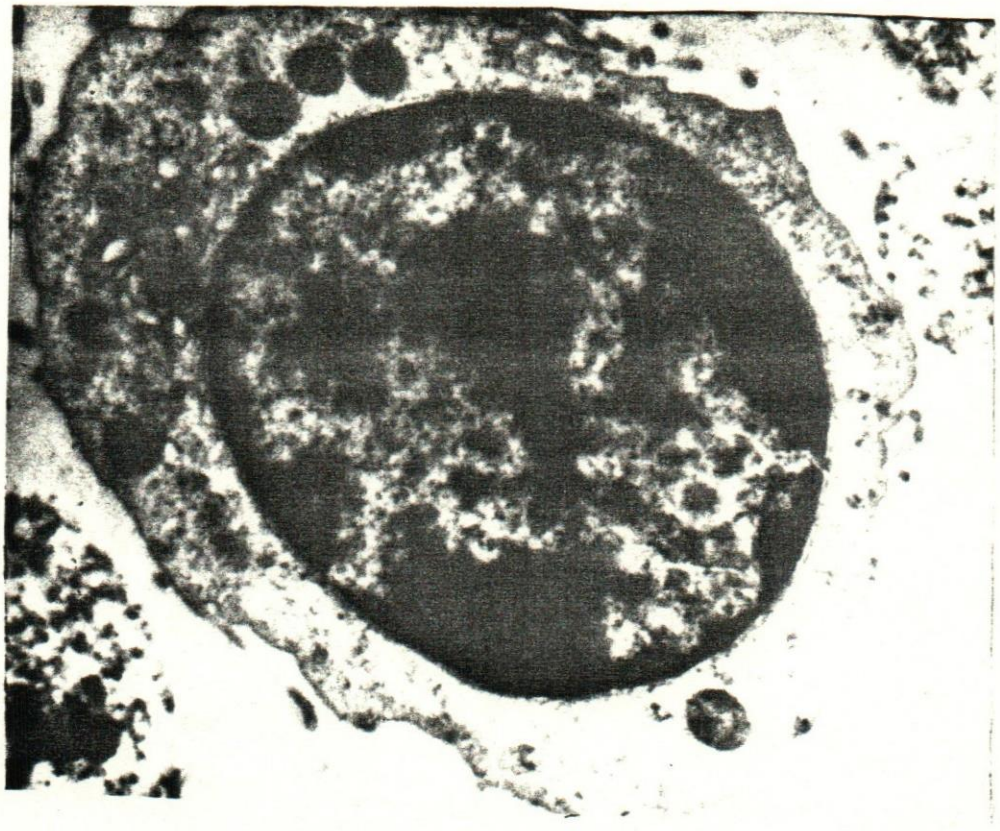

Fig. (2)

Fig. (3)

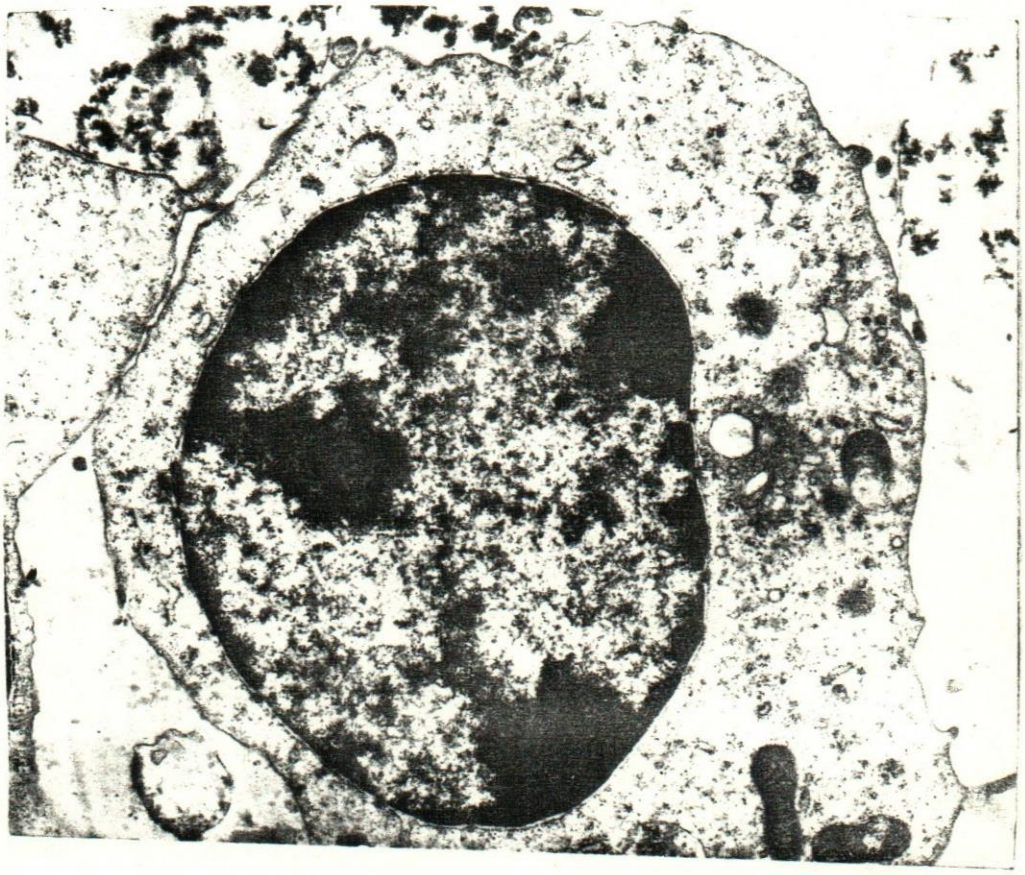

Fig. (4)