International Journal Dental and Medical Sciences Research
Volume 3, Issue 5, Sep-Oct 2021 pp 659-663 www.ijdmsrjournal.com ISSN: 2582-6018

Endothelial changes observed on Fine needle aspiration Cytology of cervical lymph node in a case of COVID-19: looking through the window

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Submitted: 15-09-2021 Revised: 25-09-2021 Accepted: 28-09-2021

ABSTRACT-
The role of endothelial cells in the pathogenesis of COVID-19 has been well established. The changes of endothelial cells in COVID-19 have been described in different histopathological and electron microscopic studies. We describe the endothelial changes observed on fine needle aspiration cytology of cervical lymph node of a 25 years COVID-19 positive young girl. The smears revealed cells of lymphoid series and histiocytes along with presence of capillary fragments and scattered as well as clusters of plump endothelial cells. The endothelial cells appeared hyperplastic and swollen with abundant pale eosinophilic or vacuolated cytoplasm. Some of the endothelial cells showed enlarged bizarre nuclei with coarsely granular chromatin. Few of these appeared protruding from their attachment, giving hobnail appearance and few showed intranuclear eosinophilic inclusions (suspected viral inclusions). These changes in endothelial cells observed on cytology are suggestive of viral infection of the endothelial cells in COVID-19 with viral cytopathic effect.

Key words- endothelial cells, COVID-19, viral cytopathic effect, fine needle aspiration cytology, viral inclusions

I. INTRODUCTION

The enigma of pathology and pathogenesis of Coronavirus disease (COVID- 19) is increasingly being deciphered by the various elaborate reports on autopsy and biopsy studies. Till now there have been many publications describing the histopathology of different organs in COVID-19 along with reports on special studies like immunohistochemistry and electron microscopy. [1] We present the cytological observations of cervical lymph node in a case of COVID-19 with emphasis on the unusual changes observed in endothelial cells, suggestive of viral cytopathic effect.

II. CASE REPORT

A 25 years girl visited Medicine OPD with complaints of fever, weakness and cough since three to four days. On examination she was febrile. Pulse rate was 80/minute. Respiratory rate was 22/minute. Blood pressure was 110/78mmHg. Systemic examination did not reveal any abnormality. Local examination of right cervical region revealed 3 small firm, mobile, non- tender lymph nodes largest measuring 1.5 x 1 cm.

A throat swab was collected for RT-PCR, which tested positive for COVID-19. CT chest was normal. Hematological parameters revealed Hemoglobin- 12gm%, total leucocyte count- 3760/cmm, with a differential count of Neutrophils-70%, Lymphocytes-26%, Monocytes-04%, Eosinophils-0%, Basophils-0%. Platelet count was 220,000/cmm. ESR was 12mm at 1 hour. Her inflammatory markers such as serum ferritin, interleukin-6, C reactive protein, procalcitonin and LDH were mildly raised. Other biochemical markers were within normal limits. FNAC of cervical lymph node was advised to rule out any other specific pathology.

USG guided FNAC of the largest cervical lymph node was performed taking all standard precautions and following the standard guidelines.[2] Four smears were prepared, fixed in ethyl alcohol and stained with hematoxylin and eosin. Microscopic examination revealed moderately cellular smears showing cells of lymphoid series admixed with histiocytes (Figure 1a), at places showing a background of necrotic debris and apoptotic bodies (Figure 1b, c). However the most prominent feature was the presence of capillary fragments (Figure 1d) along with scattered and clusters of endothelial cells. The endothelial cells lining the capillaries appeared plump and hyperplastic (Figure 1e) with focal clusters of endothelial cells attached to capillaries (Figure 1f). Some of the capillaries showed swollen endothelial cells with abundant pale eosinophilic or vacuolated cytoplasm (Figure 2a). Some of the endothelial cells showed enlarged, bizarre nuclei with coarsely granular chromatin (Figure 2b, c, d).

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Few of these appeared protruding from their attachment (Figure 2e), giving hobnail appearance. Few cells showed tiny intranuclear eosinophilic inclusions (suspected viral inclusions) (Figure 2f). A diagnosis of reactive lymphadenitis with changes in endothelial cells, suggestive of viral cytopathic effect was rendered.

The patient was administered oral medications including Favipiravir and Pantop for 6 days along with multivitamins, calcium and zinc preparations for 10 days. She completely recovered from the illness. The cervical lymph nodes also regressed in size.

III. DISCUSSION :

Various autopsy studies in COVID-19 have focused mainly on the changes seen in lungs. There are few studies which describe the findings of lymph nodes in COVID- 19. Xiang et al noted depletion of lymphocytes, with necrotic and apoptotic lymphocytes in hilar lymph nodes.[3] Elsoukkary et al observed relatively preserved architecture of lymph nodes with prominent vascular congestion, expansion of sinuses containing transformed immunoblast-like cells associated with apoptotic debris (in 2 cases).[4] Wang X et al noted primary lymphoid follicles with widened interfollicular areas, lymphatic sinus and vascular endothelial hyperplasia, focal necrosis, cellular degeneration and nuclear fragmentation.[5] Sinus histiocytosis and focal hemophagocytosis in thoracic lymph nodes has been reported by Bryce et al and Martines et al. [6,7] In the present case, as we could examine only the cytological smears, the architectural details of lymphoid tissue could not be appreciated. However we observed apoptotic bodies and necrosis. But the most prominent feature was the abundance of capillary fragments and endothelial cells with focal clusters of endothelial cells attached to the capillaries. These findings point towards vascular proliferation with endothelial hyperplasia.

In Severe acute respiratory syndrome coronavirus -2 (SARS - CoV-2) infections, the emerging research data has emphasized the role of endothelial cells in the pathophysiology of COVID-19. SARS- CoV-2 infects the host using Angiotensin converting enzyme 2(ACE 2) receptor which is widely expressed on endothelial cells. [8] Electron microscopic and immunohistochemical studies have evidenced direct viral infection of endothelial cells. Varga et al performed post mortem analysis of transplanted kidney of a COVID-19 patient by electron microscopy which revealed viral inclusion structures in endothelial cells.[8] Colmenero et al demonstrated the presence of viral particles in endothelial cells in lesional skin biopsies from seven pediatric patients with COVID-19 chilblains by immunohistochemistry, one of which was also confirmed by transmission electron microscopy.[9] Paniz-Mondolfi et al found viral particles in brain capillary endothelial cells by transmission electron microscopy.[10]

There are few reports describing the changes of endothelial cells on routine light microscopy. Carnevale et al described the histopathological findings in endoscopic biopsy of large bowel in a 40 years old SARS-CoV-2 positive woman, presenting with diarrhea and abdominal pain. They observed prominent multifocal vasculitis and bizarre modifications of endothelium of small and middle sized vessels along with hobnail modifications of endothelial cells. Immunohistochemistry confirmed the presence of virus particles in the cytoplasm of the endothelial cells with hobnail and bizarre features. They concluded that these modifications may represent a peculiar cytopathic effect of the virus.[11] In our case, the cytology smears showed endothelial cells with enlarged bizarre nuclei with coarsely granular chromatin and focal hobnail appearance. These findings are similar to those described by Carnevale et al, suggestive of viral cytopathic effect. [11]

In our case the endothelial cells also showed swelling and vacuolization. Copin et al observed endothelial vacuolization and detachment in small and medium sized pulmonary arteries as a prominent feature indicative of endothelial injury, in postmortem lung biopsies of COVID-19 patients.[12] Varga et al found evidence of endothelitis in kidney, lung, heart, liver or small intestine in three different cases.[8] Colmenero et al observed endothelitis with swelling of endothelial cells, that were confirmed for the presence of viral particles with immunohistochemistry in skin biopsies of patients with COVID-19 chilblains.[9] Thus the swelling of endothelial cells noted on cytology in our case could be indicative of endothelitis with endothelial injury.

Some of the cell nuclei in our smears showed presence of tiny intranuclear eosinophilic inclusions which may represent viral inclusions. Suspected intranuclear viral inclusions have been reported in pneumocytes by Tian et al.[13] Suspicious viral inclusions in cytoplasm of pneumocytes have also been reported by Wang X et al.[5]

In summary, the cytology findings of our case are consistent with similar findings on histopathology, noted and confirmed by other authors. The limitations of this case report are that

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the cytological observations could not be detailed by histopathological examination and the presence of virus in the cells could not be demonstrated.

To conclude, the present case report emphasizes the changes in endothelial cells observed on cytology of cervical lymph node in a COVID -19 patient, which are suggestive of viral infection of the endothelial cells with viral cytopathic effect. It also indicates that though SARS- CoV-2 enters the host through lungs, the effects on endothelial cells can be observed at remote sites.

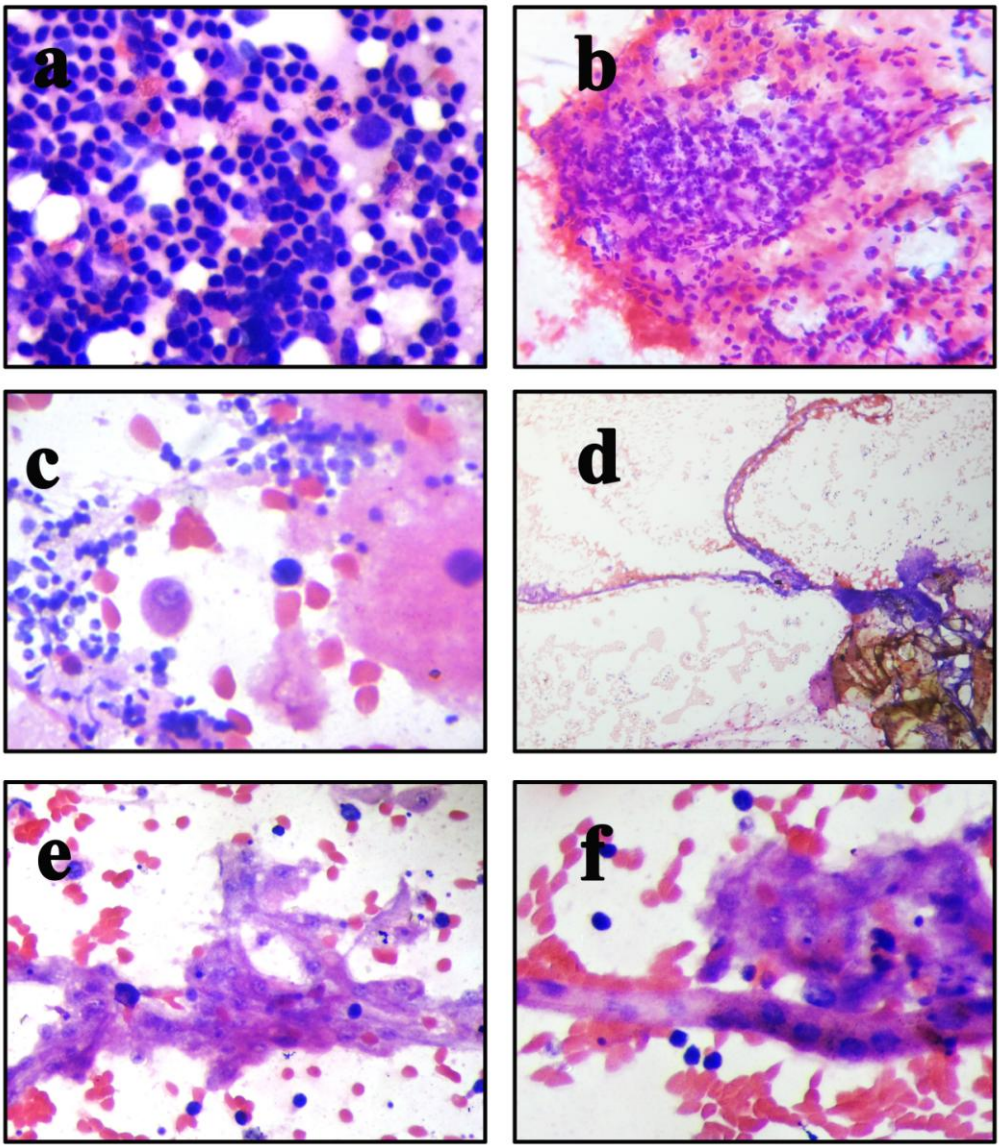

Figure 1- Photomicrograph of cytology smear showing
a- cells of lymphoid series (H&E 400X) **b-** necrotic debris (H&E 400X)
c- apoptotic bodies (H&E 400X) **d-** capillary fragments (H&E 100X)
e - plump and hyperplastic endothelial cells (H&E 400X)
f- cluster of endothelial cells attached to capillary (H&E 400X)

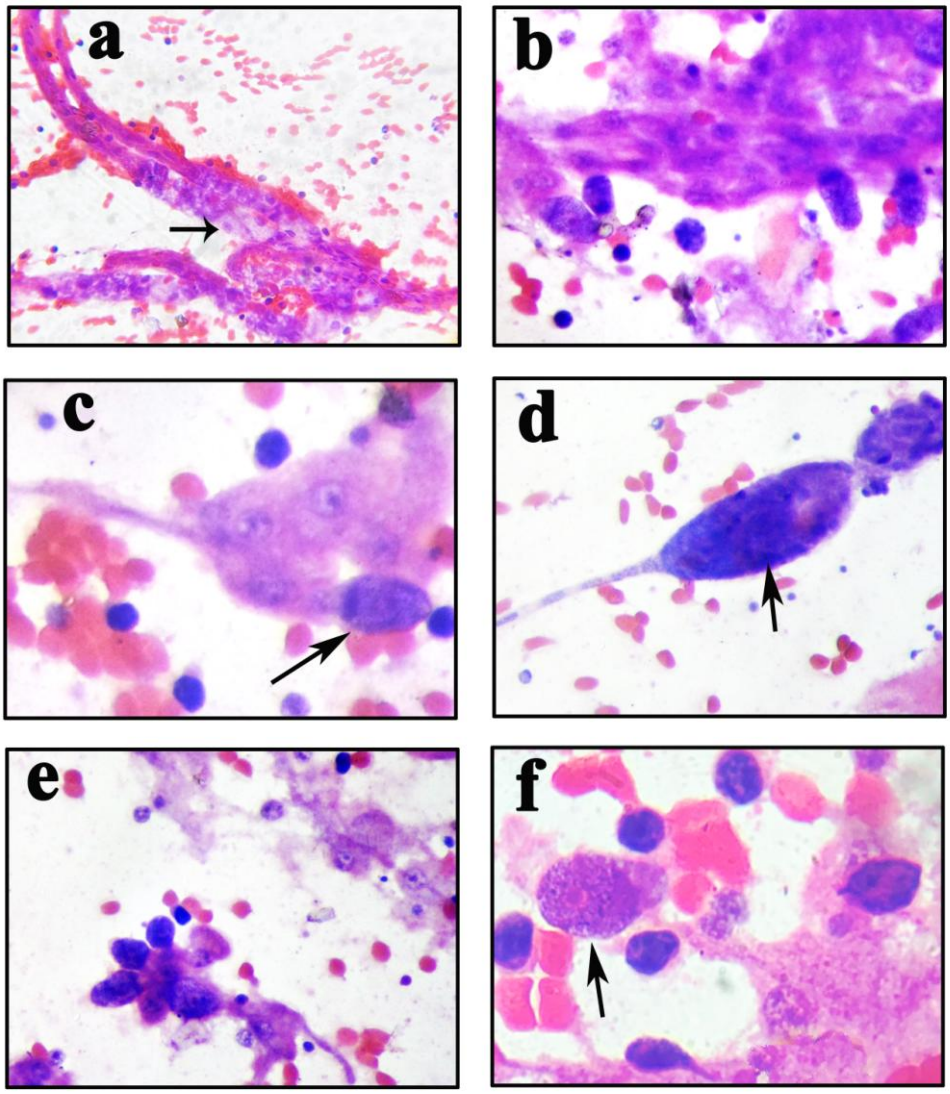

Figure -2 Photomicrograph showing
a- capillary fragment with swollen endothelial cells (H&E 400X)
b, c, d- endothelial cells with enlarged, bizarre nuclei with coarsely granular chromatin (H&E 400X)
e- endothelial cells with hobnail appearance (H&E 400X)
f- endothelial cell with intranuclear eosinophilic (suspected viral) inclusion (H&E 400X)

REFERENCES:

[1]. Vasquez-Bonilla WO, Orozco R, Argueta V, Sierra M, Zambrano LI, Muñoz-Lara F, et al. A review of the main histopathological findings in coronavirus disease 2019. Hum Pathol. 2020 Nov;105:74-83. doi: 10.1016/j.humpath.2020.07.023. Epub 2020 Aug 2. PMID: 32750378; PMCID: PMC7395947.

[2]. Srinivasan R, Gupta P, Rekhi B, Deb P, Nijhawan VS, Prasoon D, et al. Indian academy of cytologists national guidelines for cytopathology laboratories for handling suspected and positive COVID-19 (SARS-COV-2) patient samples. J Cytol 2020; 37:67-71
: https://www.jcytol.org/text.asp?2020/37/2/67/283147

[3]. Xiang Q, Feng Z, Diao B, Tu C, Qiao Q, Yang H, et al. SARS-CoV-2 Induces Lymphocytopenia by Promoting Inflammation and Decimates Secondary Lymphoid Organs. Front. Immunol. 12:661052. doi: 10.3389/fimmu.2021.661052

[4]. Elsoukkary S S, Mostyka M, Dillard A, Berman D R, Ma L X, Chadburn A, et al:

Autopsy Findings in 32 Patients with COVID-19: A Single-Institution Experience. Pathobiology 2021;88:56–68. DOI: 10.1159/000511325

[5]. Wang X, Shao C, Huang X, Sun L, Meng L, Liu H, et al. Histopathological features of multiorgan percutaneous tissue core biopsy in patients with COVID-19 J Clin Pathol 2021;**74**:522–527 doi:10.1136/jclinpath-2020-206623

[6]. Bryce, C., Grimes, Z., Pujadas, E. *et al.* Pathophysiology of SARS-CoV-2: the Mount Sinai COVID-19 autopsy experience. Mod Pathol **34,** 1456–1467 (2021). https://doi.org/10.1038/s41379-021-00793-y

[7]. Martines RB, Ritter JM, Matkovic E, Gary J, Bollweg BC, Bullock H, et al. Pathology and Pathogenesis of SARS-CoV-2 Associated with Fatal Coronavirus Disease, United States. Emerg Infect Dis. 2020;26(9):2005-2015. https://dx.doi.org/10.3201/eid2609.202095

[8]. Varga Z, Flammer AJ, Steiger P, Haberecker M, Andermatt R, Zinkernagelet AS, et al. Endothelial cell infection and endotheliitis in COVID-19. Lancet. 2020;395(10234):1417-1418. doi:10.1016/S0140-6736(20)30937-5

[9]. Colmenero I, Santonja C, Alonso-Riaño M, Noguera-Morel L , Hernandez-Martın A , Andina D,et al. SARS-CoV-2 endothelial infection causes COVID-19 chilblains: histopathological, immunohistochemical and ultrastructural study of seven paediatric cases. Br J Dermatol. 2020; 183(4):729-737. doi:10.1111/bjd.19327

[10]. Paniz-Mondolfi A, Bryce C, Grimes Z, Gordon RE, Reidy J, Lednicky J, et al. Central nervous system involvement by severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2). J Med Virol. 2020 Jul; 92(7):699-702. doi: 10.1002/jmv.25915. PMID: 32314810; PMCID: PMC7264598.

[11]. Carnevale S, Beretta P, Morbini P. Direct endothelial damage and vasculitis due to SARS-CoV-2 in small bowel submucosa of COVID-19 patient with diarrhea. J Med Virol. 2021 Jan;93(1):61-63. doi: 10.1002/jmv.26119. Epub 2020 Jun 24. PMID: 32492199; PMCID: PMC7300801.

[12]. Copin, M., Parmentier, E., Duburcq T, Poissy J, Mathieu D. Time to consider histologic pattern of lung injury to treat critically ill patients with COVID-19 infection. Intensive Care Med 46**,** 1124–1126 (2020). https://doi.org/10.1007/s00134-020-06057-8

[13]. Tian S, Hu W, Niu L, Liu H, Xu H, Xiao SY. Pulmonary Pathology of Early-Phase 2019 Novel Coronavirus (COVID-19) Pneumonia in Two Patients with Lung Cancer. J Thorac Oncol. 2020; 15(5):700-704. doi:10.1016/j.jtho.2020.02.010