COVID-19 is a systemic multiorgan disorder with major involvement of the lungs and heart leading to Interstitial Pneumonia, Diffuse Alveolar Damage (DAD) and Acute Respiratory Distress Syndrome (ARDS). An important mechanism responsible for the widespread COVID-associated mortality is presumed to be the ineffective immune responses to the SARS-CoV-2 virus along with an associated thrombotic microangiopathy that ultimately leads to multiorgan failure and death. Even COVID-19 survivors with preexisting comorbidities; especially the elderly, run a risk of secondary neurologic and cardiopulmonary complications and might sometimes succumb to sudden death. Autopsy findings are crucial to gaining a better understanding of the pathobiology of this “novel” disease as well as analyzing its long-term effects on target organs. In India, due to the prohibitive regulations regarding COVID autopsies; very little data is available on autopsy histopathology of patients dying of COVID-19; as well as those recovering from the disease, only to pass away during the recovery period. The present study aims to document the cardiopulmonary abnormalities found in autopsies of COVID-positive patients conducted at our institution while simultaneously conducting a review of the available international literature on the related topic. This will be particularly of interest for clinicians treating COVID-19 in Central India, as; of now, no similar studies have been reported from this region.

As rightly emphasized by Balachanda et al., the need of the hour is to conduct more longitudinal studies to assess the health status of the COVID-19 recovered patients. Follow-up survey of COVID-19 recovered patients will be helpful to evaluate any long-term changes in the other organs inhuman systems.

As more data emerges from the COVID-19 pandemic, it is gradually becoming clear that there is an important cardiovascular component to this disease as well. The heart frequently shows acute cardiomyocyte injury and, in some cases, pericarditis and/or myocarditis. Patients with fatal COVID-19 frequently are obese and have pre-existing cardiac disease, hypertension and/or diabetes mellitus. However, not much data is available on autopsies conducted on those patients who seem to have apparently recovered from COVID-19, yet inexplicably pass away during the post recovery period. In our country, this comundrum is further aggravated by an administrative ruling that prohibits autopsies in COVID positive cases barring a few special circumstances.

SARS-CoV-2 is a respiratory virus. Autopsies from patients with COVID-19 confirm that majority of severely affected patients have significant pulmonary pathology. COVID-19 produces an acute interstitial pneumonia, usually with a prominent diffuse alveolar damage (DAD) component. However, an important additional mechanism that contributes to death is thrombotic microangiopathy. Mediators released during cytokine storm overlap with those of thrombotic microangiopathy, suggesting that ineffective immune responses to SARS-CoV-2, severe interstitial pneumonia, ARDS, multiorgan failure and life-threatening microangiopathy are closely related to each other.

Numerous studies have shown that COVID-19 survivors with preexisting comorbidities; especially those who are of older age, have substantial physical as well as psychological disabilities and also run a risk of secondary neurologic and cardiopulmonary complications. Sometimes, sudden death may be encountered in such patients outside the hospital setup. Death may also occur unexpectedly in apparently healthy individuals after a brief period of trivial flu-like symptoms. In such doubtful cases, the forensic pathologist can help in defining the cause of death.

As rightly emphasized by Balachanda et al., the need of the hour is to conduct more longitudinal studies to assess the health status of the COVID-19 recovered patients. Follow-up survey of COVID-19 recovered patients will be helpful to evaluate any long-term changes in the other organs inhuman systems.

MATERIALS AND METHODS:
STUDY DESIGN: Bodies of three COVID-19 positive patients was received at the Forensic Medicine and Toxicology Department, CIMS, Bilaspur. Due to the medicolegal implications involved, autopsies were conducted on these patients in accordance with MOH&FW regulations and with the consent of the decedent’s kin. As our institution is a tertiary care non-COVID hospital, the only patients referred for treatment to our institute are those who had previously contracted and subsequently recovered from COVID-19. Thus, the autopsies were conducted to rule out post-COVID complications as the possible cause of death. Subsequently the heart and lungs of the autopsied patients were submitted to the Forensic Histopathology Section, Department of Pathology, CIMS, Bilaspur for histological examination.

INCLUSION AND EXCLUSION CRITERIA:
Due to the paucity of the number of cases no inclusion and exclusion criteria were set.

DATA COLLECTION:
Relevant information was collected regarding the age, sex, nature and duration of presenting complaints for which the patients sought medical intervention. Detailed medical history including history of comorbidities known to predispose to COVID-19 and treatment received prior to death was obtained.

GROSS EXAMINATION:
The received viscera were weighed, measured and detailed descriptions of grossly appreciable pathological findings were noted.

MICROSCOPIC EXAMINATION:
Tissue sections at 6μ were cut and slides prepared, which were analyzed. Descriptions of grossly appreciable pathological findings were noted.

KEYWORDS
COVID-19, Pneumonia, DAD, ARDS, Myocarditis, Microangiopathy, Autopsy, Histopathology.
subsequently stained with H&E stain and examined under the microscope.

**OBSERVATION:**

**Table 1: Clinical Presentation**

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Age</th>
<th>Sex</th>
<th>Known Comorbidities</th>
<th>COVID Disease Category</th>
<th>Symptom-free interval after Covid recovery</th>
<th>Post-COVID Symptoms</th>
<th>Duration from Hospitalization until Death (in days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>70</td>
<td>M</td>
<td>HT, DMII</td>
<td>ILI</td>
<td>1 month</td>
<td>Severe Chest Pain, Collapse</td>
<td>0 (brought dead)</td>
</tr>
<tr>
<td>2</td>
<td>90</td>
<td>F</td>
<td>HT</td>
<td>Mild ILI</td>
<td>14 days</td>
<td>Loss of Consciousness</td>
<td>1</td>
</tr>
<tr>
<td>3</td>
<td>75</td>
<td>M</td>
<td>COPD with PAH</td>
<td>SARI</td>
<td>-</td>
<td>Severe Breathlessness, Constriction feeling in chest</td>
<td>5</td>
</tr>
</tbody>
</table>

**Table 2: Autopsy Findings in Heart- Gross**

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Weight in gms</th>
<th>Dimensions in cms</th>
<th>External Surface</th>
<th>Cut Surface</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>492</td>
<td>11x11x6</td>
<td>Heart enlarged in size, bulky Vertical rupture in Anterolateral LV wall (8x2 cm) Hemopericardium with approx. 100ml blood in pericardial cavity</td>
<td>RV-0.8CM LV-1.4CM IVS-1.6CM RCA-thickened LCA-thickened, PM clot +</td>
</tr>
<tr>
<td>2</td>
<td>688</td>
<td>14.5x10.5x5</td>
<td>Heart enlarged in size, flabby in consistency</td>
<td>All chambers dilated RV-0.5CM LV-1CM IVS-1.3CM RCA &amp; LCA-grossly unremarkable</td>
</tr>
<tr>
<td>3</td>
<td>469</td>
<td>10x9.6x5.5</td>
<td>Heart slightly enlarged. Thick epicardial fat pad. Anterior descending artery appears thickened and prominent</td>
<td>RV-0.8 L.V-1.3 IVS-1.5 RCA-thickened, PM clot + LCA-thickened, cord-like, clot +</td>
</tr>
</tbody>
</table>

**Table 3: Autopsy Findings in Heart- Histopathology**

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Microscopic Findings</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Extensive areas of ischemic cardiomyocyte necrosis. Marked myxoid degeneration in necrotic areas in IVS and LV wall, especially on both sides of the ventricular lense. Diffuse infiltration of lymphocytes in the interstitium. Lipofuscin pigments and macrophages seen at places. Epicarditis noted in sections from RV. Microthrombi present in arterioles near infarcted areas. Other small caliber blood vessels show congestion. Both RCA and LCA show fibroatheroma with narrowing of lumen.</td>
<td>Cardiac Tamponade following rupture of Ventricular Aneurysm (complications of CAD &amp; MI)</td>
</tr>
</tbody>
</table>

**Table 4: Autopsy Findings in Lung- Gross**

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Weight in gms</th>
<th>Dimensions in cms</th>
<th>External Surface</th>
<th>Cut Surface</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>820 gm only rt. Lung received</td>
<td>13x12x5</td>
<td>Part of lower lobe not included (submitted for other forensic inv.) Lung appears heavy, external surface mottled with patchy, dark, brownish areas.</td>
<td>Loss of sponginess, decreased air entry, mottled blackish appearance</td>
</tr>
<tr>
<td>2</td>
<td>350g approx.</td>
<td>Bits of lung tissue together measuring 4x3x3cm.</td>
<td>Both lungs fibrotic and adherent to the chest wall; could only be removed in bits and pieces</td>
<td>Loss of sponginess, greyish white areas visible. Pleura thickened</td>
</tr>
<tr>
<td>3</td>
<td>Rt. Lung-980 gm Lt. Lung-820 gm</td>
<td>Rt. Lung-12x7x6 Lt. Lung-9x6x5</td>
<td>Part of lower lobe not included (submitted for other forensic inv.) Rt. Lung: appears heavy, external surface greyish, hemorrhagic Lt. Lung: external surface mottled with multiple blackish spots</td>
<td>Rt. Lung: solid, liver-like consistency in focal areas; loss of sponginess, decreased air entry. C/S appears greyish brown to brownish-black Lt. Lung: Loss of sponginess, decreased air entry, mottled blackish appearance</td>
</tr>
</tbody>
</table>

**Table 5: Autopsy Findings in Lung- Histopathology**

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Microscopic Findings</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Disruption of alveolar lining. Compression of alveolar spaces. Emphysematous dilatation of unaffected alveoli. Widening of interalveolar septae. Increased no. of Type II pneumocytes. Atypical changes of bronchiols</td>
<td>DAD, Organizing Proliferating Phase</td>
</tr>
</tbody>
</table>
Disease. Valvular heart disease has the greatest impact on heart cardiomegaly and an increase in heart weight, ranging from 340 g to 1010 g, especially in patients with history of cardiovascular disease. Valvular heart disease has the greatest impact on heart weight, followed by old myocardial infarction, coronary atherosclerosis, and hypertension.\\n
In our study however, the heaviest weighing heart was found in Dilated Cardiomyopathy as seen in Case No. 2 (688 g); followed by Coronary Atherosclerosis and Myocardial Infarction as seen in Case No.1 (492 g). The patient was a known diabetic and hypertensive. Case No. 3, having Coronary Atherosclerosis with Chronic Ischemic Heart Disease recorded a heart weight of 469 g. In all the three cases studied by us, cardiomegaly and increase in heart weight was found in accordance to these findings.

Older COVID patients with preexisting comorbidities like hypertension, coronary arteri disease, heart failure, and diabetes are prone to develop a significantly higher risk of sustaining myocardial injury and higher short-term mortality rate. This could be corroborated in both Case No. 1 & 3.

Case No. 1 was a known diabetic and hypertensive, who, one month after recovering from COVID, succumbed to a massive myocardial infarct. A rupture in the anterior wall LV wall was revealed upon autopsy, along with well as presence of calcified fibroatheromas in both coronary arteries. In Case No. 3, autopsy examination of the heart revealed a thick epicardial fat pad as well as a developing atheroma in the LCA.

Coronavirus may be associated with infectious dilated cardiomyopathy. As observed in Case No. 2 of our study, such cases are grossly observable as cardiomegaly with right and left ventricular dilatation with thinning of ventricular walls.

Lungs: (Table 3). Normal postmortem weights of right & left lungs are 608.32 g & 505.86 g respectively for men and 481.60 g & 410.90 g respectively for women, reaching peak weights of 60.79g for left and 573.11g for left lung in the 61-70yr age group. However, in most autopsy lung studies of COVID 19 decedents, the combined weight of was found to be >1300 g (average upper limit of normal). In our study, the individual and combined lung weights in Case No. 1 & 3 were found to be well above the upper limits of normal levels adjusted for age and sex and were comparable to the findings recorded by other authors. No conclusive opinion could be derived in Case No. 2 due to the paucity of the sample received.

Clinical examination of specimen in Case No. 1 showed heavy, congested and edematous lung with patchy, dark, brownish mottling of external surface with cut surface showing loss of sponginess, decreased air entry and mottled brownish to blackish hemorrhagic appearance. Similar findings were also seen in the lung specimens in Case No. 3 with the right lung additionally showing areas of patchy consolidation, while the left lung showed frank areas of parenchymal hemorrhage. Even though pleuritis and pleurisy have been reported in the setting of COVID 19, we were unable to conclusively attribute similar findings in Case No. 2 to COVID as other preexisting pathologies could not be ruled out due to lack of sufficient clinical information and tissue specimen.

Microscopic Examination: Heart: (Table 4). Patients with cardiovascular disease have the highest Case Fatality Rate (10.5%) among those COVID patients with medical comorbidities. However autopsy histopathology studies show a wide variety of abnormalities ranging from myocyte hypertrophy to isolated/focal cardiomyocyte necrosis to full blown myocardial infarction.

In accordance with the former, we found evidence of individual and small groups of necrotic cardiomyocytes surrounded by interstitial fibrosis in Case No. 2 & 3. With regard to Case No. 1, our findings correlated most closely with the findings of Elsoukkary S.S. et al where we found extensive areas of infarction with myxoid degeneration and rupture of the ventricular wall. (Figure 1). Fibroatheromas causing narrowing of lumen were found in RCA in Case No. 3 and both coronary arteries in Case No. 1, which was again in keeping with the findings of Elsoukkary S.S. et al. The elongated cardiomyocytes seen in Case No. 2 could be due to the shear stress on the dilated myocardium. There is wide variance in the references regarding the degree of and nature of inflammatory infiltrates; with Fox et al, Bryce et al, and Tian et al reporting insignificant or mild inflammatory infiltration, while others have found evidence of myocardiitis and pericarditis in their studies. All the three cases in
The hallmark pulmonary pathology in COVID-19 is an Acute Respiratory Distress Syndrome (ARDS) manifested histologically as Diffuse Alveolar Damage (DAD). ARDS/DAD is histologically characterized by 3 phases: Exudative, Proliferative/Organizing, And Fibrotic.

The Exudative Phase of ARDS is characterized by the destruction of Type 1 alveolar cells and the capillary endothelial cells with accumulation of protein-rich edema fluid and cellular debris in the alveolar spaces and collapse of alveolar sacs (Figure 2a, 2b). The most characteristic feature is the presence of hyaline membranes lining the alveolar ducts (Figure 2c). Additional features are viral cytopathic effects including viral inclusion bodies; septal widening due to congestion and edema; interstitial and intra-alveolar hemorrhage along with the presence of microthrombi (Figure 3) and megakaryocytes in pulmonary capillaries. All the aforementioned findings were seen in Case No. 3.

The Fibrotic Phase of ARDS was reported only in a few cases of COVID-related lung disease. In this phase, the inflammatory exudates are replaced by extensive alveolar and interstitial fibrosis. The remaining alveolar spaces are disorganized and surrounded by thick fibro-collagenous bands giving rise to a microscopic honeycomb-like change. Reactive squamous and even osseous metaplasia may be seen in the residual tissue. Intimal fibrosis of pulmonary vessels may lead to progressive vascular occlusion and pulmonary hypertension. In our study, even though the features in Case No. 2 closely resembled a majority of these findings, we could not conclusively attribute the findings to COVID due to our inability to rule out preexisting pathologies in the absence of sufficient clinical information and sample tissue.

Some cases of DAD in the autopsied specimens also showed evidence of superimposed pneumonia, a finding which was also reflected in Case No. 3 of our study.

**REVIEW OF LITERATURE:**

When a new or re-emergent pathogen, such as SARS-CoV-2, causes a major outbreak, rapid access to pertinent research findings is crucial for planning strategies and decision making. As the COVID-19 pandemic spread rapidly, causing widespread major morbidity and mortality, it became crystal clear to forensic pathologists and allied physicians that autopsy of deceased victims of the disease was of paramount importance for gaining knowledge of its pathogenesis and pathophysiology. Therefore, a detailed review of the postmortem gross and histopathological findings in lungs and heart, the organs most conspicuously affected by the SARS-CoV-2 virus, will help highlight the pathognomonic signs of COVID-19 disease.

Autopsies performed on COVID-19 patients may be broadly divided into two groups: minimally invasive autopsies and complete autopsies.

The earliest postmortem studies were minimally invasive autopsies conducted in the form of ultrasound-based minimally invasive autopsies (Dollinikoff et al.), postmortem transthoracic needle biopsies of the lungs (Zhang et al.), core biopsy samplings of lung, heart, and liver (Xu et al.) etc. Li et al. analyzed the three-dimensional histology reconstruction obtained from lung tissue samples of patients.
who died because of COVID-19 and were able to document viral cytopathic changes in pneumocytes as well as emphasize the presence of megakaryocytes & fibrin aggregates in pulmonary capillaries.  

Subsequent reports of complete autopsy findings by Elsoukkary et al., Buja et al., Tombolini et al., Fox et al. etc. helped to grasp a better understanding of the pathobiology of this “novel” disorder. 

The hallmark pulmonary pathology in COVID 19 is an Acute Respiratory Distress Syndrome (ARDS) / Diffuse Alveolar Damage (DAD). The features are similar to the pulmonary damage observed in the previous Coronavirus mediated epidemics SARS (Severe Acute Respiratory Syndrome) and MERS (Middle Eastern Respiratory Syndrome). However, unlike the SARS and MERS viruses, the SARS-CoV-2 virus disproportionately affects older patients with comorbidities such as hypertension, diabetes, obesity, and cardiovascular disease. 

Grossly; even though the most conspicuous morphological abnormalities were reported in the lungs, Elsoukkary et al., Buja et al., Fox et al., Fitzek et al. documented evidence of cardiomegaly and increase of heart weights above the normal range for age and sex in COVID 19 decedents. As expected, most autopsy lung studies found the total weight of >1300 g. with Boczarz et al. recording individual lung weights as heavy as 1100g.1,2,13

Histologically, the most frequent pathological finding is both exudative and proliferative DAD as documented by Fox et al. and others. 1,9,13,17,23,26 Though infrequently, late-stage DAD has been reported in the studies of Fox et al.,1 Remmelink et al.,2 Schaller et al. and Zhang et al. 1,9 Of particular interest is the report of reactive squamous and osseous metaplasia in a case of Fibrotic DAD studied by Schaller et al. 23

Konopka et al. 39 found heavy lung with mucous within the airways in an asthmatic patient who died of COVID 19 infection and documented chronic asthmatic alterations of the airways in addition to features of acute exudative DAD. Organizing pneumonia was found by Elsoukkary et al. and Buja et al.; while Menter et al.,29 Bradley et al.,30 Edler et al. 31 and others32,30,32 were able to recognize superimposed bacterial pneumonia upon histological examination. Another significant finding reported by most authors was viral cytopathic effect on the pneumocytes; with Tombolini et al. documenting marked cytological atypia, syncytiat formation and Cowdry Type A viral inclusion bodies in the pneumocytes.

Conversely, however, Lacy et al. and Aguilar et al. 30 were unable to find any viral inclusions or cytopathic changes even in the presence of exudative DAD with hyaline membranes. In such cases, conclusive evidence may be obtained by SARS-CoV-2-specific immunohistochemistry as done by Zhang et al., who were able to demonstrate viral particles in the alveolar epithelium that were almost undetectable on the interstitium and vessel walls.

Almost all studies reported the presence of microthrombi in the small and medium caliber pulmonary vessels, with Grimes et al.,13,27 Barton et al.26, Bryce et al.13 and Remmelink et al. 20 being able to find thrombi even in larger pulmonary vessels. Additionally, Tombolini et al., Aguilar et al., Duarte Neto et al. and Dolnikoff et al.29,32 were able to demonstrate naked megakaryocytes in pulmonary and other systemic capillaries, confirming an important pathogenic role of thrombotic microangiopathy in the pathophysiology of COVID 19.

Mago et al. 27 found relevant signs of systemic activation of the complement cascade and detected both SARS-CoV-2 spike glycoproteins and C4d and C5b-9 in the alveolar septa, indicating that activation of the complement cascade might also contribute to the pathogenesis. Varga et al. studied the endothelial damage to various organs and found evidence of lymphocytic endothelitis along with viral inclusions in the endothelial cells of various organs.

Significant cardiac histopathological abnormalities were reported by Elsoukkary S.S. et al., Buja et al., & Bryce et al. and Basso et al. 40 The most consistent finding in most autopsy studies of COVID hearts was a mild lymphocytic myocarditis12,25,33,37,38; though Ektundene et al. 22 and Bryce et al. also found macrophages in the myocardial interstitium. Bryce et al. also observed evidence of epicarditis along with Schaller et al.; while Buja et al. and Basso et al. documented evidence of lymphocytic pericarditis in their studies. However, contrary to the aforementioned findings, Fox et al. and Tian et al. 3 found little or no evidence of myocarditis in their studies.

While some authors found evidence of individual 12 or multifocal acute myocyte injury 12,34,35 Elsoukkary et al. 12 found evidence of remote myocardial injury in the form of patchy interstitial fibrosis as well as full blown myocardial infarction. Acute MI in a COVID patient was also reported by Remmelink et al. 12 Bryce et al. & Duarte-Neto et al. reported finding microthrombi in small vessels of the myocardium. Elsoukkary S.S. et al. along with Yan et al. 12 also reported cardiomyocyte hypertrophy. A peculiar finding reported by Menter et al. 26 was the increase in the incidence of senile cardiac amyloidosis in COVID decedents.

Review of available literature on the effect of SARS-COV-2 on other organs revealed discovery of viral particles in various organs including lungs, kidneys, liver, brain, intestines and brain. Similar to heart and lung; microthrombi were found in numerous vessels including brain, liver (portal venules), kidney (glomeruli) and skin. Some authors also found evidence of deep vein thrombosis 41 with varying grades of pulmonary embolism 12

CONCLUSION:
The study of autopsy histopathology of cardiopulmonary abnormalities in COVID 19 decedents has been a valuable source of information in understanding the pathophysiology as well as modelling the most appropriate therapeutic interventions to combat this new and “novel” disease. The available data point towards ARDS/DAD and thrombotic microangiopathy as the main pulmonary features of COVID 19, with microthrombi also being found in other vital organs like heart, brain, liver and kidney. Unfortunately, in India, not much data is forthcoming on COVID autopsy cases due to MOHFW guidelines that prohibit autopsies in COVID positive cases barring a few special circumstances. The need of the hour, therefore, is “defending science in a time of fear and uncertainty” 42 and allowing more autopsy studies to be conducted on COVID 19 patients in the larger interests of the lay public in general and the medical fraternity in particular.

REFERENCES:


