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Review

The effect of environmental diesel exhaust pollution on SARS-CoV-2 infection: The mechanism of pulmonary ground glass opacity

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ABSTRACT

Diesel exhaust particles (DEP) are the major components of atmospheric particulate matter (PM) and chronic exposure is recognized to enhance respiratory system complications. Although the spread of SARS-CoV-2 was found to be associated with the PMs, the mechanism by which exposure to DEP increases the risk of SARS-CoV-2 infection is still under discussion. However, diesel fine PM (dPM) elevate the probability of SARS-CoV-2 infection, as it coincides with the increase in the number of ACE2 receptors. Expression of ACE2 and its colocalized activator, transmembrane protease serine 2 (TMPRSS2) facilitate the entry of SARS-CoV-2 into the alveolar epithelial cells exposed to dPM. Thus, the coexistence of PM and SARS-CoV-2 in the environment augments inflammation and exacerbates lung damage. Increased TGF-β1 expression due to DEP accompanies the proliferation of the extracellular matrix. In this case, “multifocal ground-glass opacity” (GGO) in a CT scan is an indication of a cytokine storm and severe pneumonia in COVID-19.

1. Introduction

Coronavirus disease 2019 (COVID-19) is an acute pulmonary illness derived from the severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) infection, which has rapidly attained pandemic status worldwide. This pulmonary disease was firstly announced at the end of December 2019 in Wuhan, China (Zhu et al., 2020a, 2020b). It has been determined that the combined effect of meteorological conditions and rising air pollution and particulate matter (PM) is important in the spread of virus infection. Recently, different reports from Europe, China, and the USA underlined the contribution of PMs to the course of COVID-19 (Frontera et al., 2020; Sciomer et al., 2020). In a study conducted by Zhu et al. (2020a) in China covering 120 cities, an increase was recorded in more than 10 μg/m³ of aerodynamic particles, PM2.5, resulting in more than 2% increase in new cases. Additionally, increases in atmospheric pollution with PM2.5, PM10, nitrogen dioxide (NO2), and O3 cause between 1.76–6.94 % rising in the daily amounts of cases (Zhu et al., 2020a, 2020b). Indeed, 78 % of COVID-19 deaths observed in five districts of northern Italy and central Spain, occurred where NO2 was recorded as being at a higher intensity compared with other regions. Short or long-term exposure to PM2.5 and NO2 is equally efficient in increasing SARS-CoV-2 infection and associated mortality (Ali and Islam, 2020). Vehicle exhaust is the major source of NO2 and this gas gives rise to adverse outcomes on the respiratory system by causing cytokine-mediated pulmonary injury (Grange et al., 2019; Petit et al., 2017). The information from US Environmental Protection Agency Environmental Justice Screen (EPAEJS) indicated that COVID-19 prevalence and mortality frequencies are markedly connected with diesel PM (Hendryx and Luo, 2020). Moreover, the results of environmental analyses from 25 cities in India demonstrated a positive correlation between the concentrations of PM2.5 and COVID-19 mortality (Mele and Magazzino, 2021). The most frequent serious clinical presentation of this infection is pneumonia and 67 % of these patients demonstrate serious manifestations of acute respiratory distress syndrome (ARDS) (Yang et al., 2020) (Fig. 1A).

Indeed, ARDS is a high-mortality clinical manifestation which is emerged most probably via the excessive activation of the renin-angiotensin system (RAS) as a result of the SARS-CoV-2 infection. It has been suggested that the angiotensin-converting enzyme 2 (ACE2) receptor normally protects against lung injury through antagonizing the activation of the classical RAS ACE-Angiotensin II (Ang II)-Angiotensin II receptor type 1 (AT1R) axis. However, the protective function of ACE2

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2. Diesel exhaust particles and SARS-CoV-2 infection

Diesel fine PM (dPM) 2.5 exposure has been well documented to disturb alveolar epithelial cell differentiation, in parallel with the increased nicotinamide adenine dinucleotide phosphate oxidases expression, as well as inflammation. In response to the dPM2.5 exposure, ACE2 receptor via which SARS-CoV-2 enters the target cells, and

Fig. 1. A. Typical chest computed tomography (CT) findings in SARS-CoV-2 infection are multifocal, bilateral, ground-glass opacities (GGO) (shown with green circles) with thickened inter- and intralobular septa. (A 42-year-old male patient admitted to the hospital during the COVID-19 outbreak on April 24th, 2020 with history of fever, cough, diffuse myalgia and fatigue. After fourth day of PCR test positivity, CRP: 65 mg/Lt (Normal value:0-5 mg/Lt), IL-6: 27 pg/mL (Normal value:0-7 pg/mL)).

B. Molecular mechanism of typical pulmonary lesions development due to environmental DEP and SARS-CoV-2 exposure.

Abbreviations. ACE2: Angiotensin-converting enzyme 2; Aer: Aerosol; AngI: Angiotensin I; AngII: Angiotensin II; AT1R: Angiotensin II receptor type 1; CT: Computed tomography; DEP: Diesel exhaust particle; dPM2.5: Diesel fine particulate matter 2.5; ECMGS: Extracellular matrix ground substance; EGF: Epidermal growth factor; FGF: Fibroblast growth factor; IgG: Immunoglobulin G; IL-6R: Interleukin-6 receptor; NADPH: Nicotinamide adenine dinucleotide phosphate; NF-κB: Nuclear factor kappa-light-chain-enhancer of activated B cells; NO2: Nitrogen dioxide; NPs: Nanoparticles; PDGF: Platelet-derived growth factor; RNS: Reactive nitrogen species; ROS: Reactive oxygen species; SARS-CoV-2: Severe acute respiratory syndrome corona virus-2; TGF-β1: Transforming growth factor beta 1; TMPRSS2: Transmembrane protease serine 2; TNF-α: Tumor necrosis factor-alpha; TNFαR: Tumor necrosis factor alpha receptor.
the cofactor transmembrane protease serine 2 (TMPRSS2) are highly upregulated in the alveolar epithelial cells (Kim et al., 2020a, 2020b). Importantly, generation and emission of DEPs from diesel engines are one of the main components of aerial PM (Ohtoshi et al., 1998). DEP upregulates adhesion molecules and induces endothelial cell activation. Thus, inflammatory mediators are released from endothelial cells and alveolar macrophages. In daily life, co-exposure of people to different PMs, including variety of metal oxide nanoparticles (NPs) within DEP potentially eventuates in a diverse toxicity pattern compared to the responses induced by the sole metal oxide NP (Engin, 2021). Furthermore, the respiratory epithelium and alveolar macrophages are main destinations of these DEP-derived PMs. It has been observed that elevated morbidity and mortality from exacerbation of chronic obstructive pulmonary diseases are closely dependent on the exposure to PM2.5–10 (Ohtoshi et al., 1998). However, it has been claimed that this increase in COVID-19 infection is associated with the high population density in contrast to the short-term exposure to DEP contamination (Copiello and Grillenzoni, 2020). Nevertheless, epidemiological studies revealed that the amount of atmospheric PM10 and PM2.5 declines in correlation with the number of COVID-19 cases in Xiaoshan, China (Li et al., 2020a). Recent reports of the statistical data on pandemic displayed that Lombardi and Emilia Romagna in northern Italy had higher incidence of COVID-19 mortalities compared to other zones of Italy (Conticini et al., 2020). Consequently, the viruses are adsorbed onto dPMs and remain in the air for a long time (Martelletti and Martelletti, 2020). Thereby the accumulation of virus concentration increases via inhaled PM in the respiratory tract. Atmospheric intensity of PMs and NO2 has a significant relationship with pulmonary diseases in humans. In brief, contamination of dPM2.5 provides a suitable medium to “keep” and “carry” the SARS-CoV-2 during the transportation via respiratory air (Fig. 1B). Since the dimensions of SARS-CoV-2 varies between approximately 70–90 nm, it can be carried to many intracellular organelles after entrance to the human’s cells (Kim et al., 2020a, 2020b). Following the post-“Stay Home Order” period Northwestern US city, the median traffic intensity and road occupancy declined approximately by one-third to half. Considering the atmospheric contaminants, the median black carbon and PM2.5 concentrations have markedly diminished by one-fourth and one-third. Simultaneous fall in nitric oxide (NO), NO2, nitrogen oxides (NOx), and carbon monoxide (CO) contents are 33 %, 29 %, 30 %, and 17 %, respectively (Xiang et al., 2020). Paital et al. demonstrated that there is association among NO2 emissions, PM2.5 concentrations, ACE-2 upregulation and COVID-19 infection severity (Paital and Aigraval, 2020). Thus, type II pneumocytes express ACE2 receptor, which enables viral entry (Harmer et al., 2002). The spike glycoprotein of SARS-CoV-2 has a higher binding affinity for ACE2 in human cells in comparison to alternative viral entry sites (Neuropilin 1 (NRP1) and BASIGIN (BSG)) (O’Donovan et al., 2021; Vankadari and Wilce, 2020; Wrapp et al., 2020), thereby, ACE2 receptors provide an entry zone for SARS-CoV-2 to spread over the host cells (Brake et al., 2020). Another route is an alternative or additional molecule (CD147/BSG), which is a new receptor glycoprotein of the immunoglobulin superfamily. This molecule binds to the spike (S) protein and serves as a mediator of viral invasion (Ulrich and Pillat, 2020). On the other hand, SARS-CoV-2 also enters the host cells by binding with its S protein to the NRP1 receptor, and the olfactory epithelial cells show high expression of NRP1 in COVID-19 patients. In these patients, NRP1, FURIN and TMPRSS11A levels are elevated in SARS-CoV-2 infected cells (Daly et al., 2020).

ACE2 receptors are principally upregulated in transient secretory cells of the pulmonary subsegmental bronchial branches (Lukassen et al., 2020). In addition, contrary to the S protein, which is synthesized inside the cell before its transport to the cell surface, TMPRSS2 is indispensable for the activation of viral fusion proteins (Engin et al., 2020a, 2020b; Matsuyma et al., 2010). Indeed, TMPRSS2 proteolytically splits and activates the S protein in subunit S1, which enables viral binding to the receptors of host cells (Glowacka et al., 2011; Hoffmann et al., 2020). Although ACE2 is upregulated in both transient secretory cells and pneumocytes, TMPRSS2 is expressed in pneumocytes originated from subsegmental bronchial branches (Lukassen et al., 2020). Therefore, coexistence of TMPRSS2 with ACE2 facilitates SARS-CoV-2 entrance to host cells (Shulla et al., 2011). Thus, dPMs increase the probability of SARS-CoV-2 infection via ACE2 over-expression. The integration of SARS-CoV-2 to PMs, exacerbates pulmonary damage by enhancing inflammatory process (Tung et al., 2021). RAS blockers like ACE inhibitors and Ang II receptor antagonists hinder the destructive (ACE-Ang II) action of the RAS cascade in the respiratory system (Zhang et al., 2020).

3. Immunological aspects of dPM and SARS-CoV-2 crostalk

In COVID-19 cases, SARS-CoV-2 principally infects the type II pneumocytes. The innate immune response to viral infection of type II pneumocytes results in their death by apoptosis, despite alveolar macrophage activation. The inflammatory M1 phenotype polarized macrophages activate vascular endothelial cells which take part in the development of coagulopathy, systemic sepsis, cytokine storm and ARDS (Engin et al., 2021; Morris et al., 2020). Injury of the alveolar pneumocytes is the primary ground of COVID-19-related ARDS, while endothelial cells are damaged to a smaller extent (Li and Ma, 2020). Extreme proinflammatory cytokines synthesis and release into the circulation results in subsequent ARDS exacerbation and extensive tissue injury leading to multi-organ dysfunction and death (Ragab et al., 2020). In initial phase of ARDS excessive synthesis and release of the pro-inflammatory cytokines are dependent on the activation of alveolar macrophages (Borish et al., 1992; Gosset et al., 2003). Of these cytokines, increased production of tumour necrosis factor-alpha (TNF-α) induces pulmonary inflammation (Dricoll, 2000; Driscoll et al., 1995). Over-expression of TNF-α induces nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) activation in cells expressing TNF-α receptors, tumor necrosis factor receptor 1 (TNFR-1) and TNFR-2 (Fiers, 1991).

Since, urban air particulates, including dPM provoke apoptosis of alveolar macrophages, DEP exposure results in depression of alveolar macrophage phagocytosis (Hiura et al., 1999; Jakab et al., 1990). Exposure to dPM2.5–10 induces excessive synthesis and secretion of the inflammatory cytokine interleukin-8 (IL-8), an efficient neutrophil chemoattractant in human monocytes (Monn et al., 2002). Therefore, acute exposure to DEP increases IL-8 formation in human respiratory system (Salvi et al., 2000). Increased serum concentrations of CRP, classical acute-phase protein, is a response to exposure of excessively accumulated DEP pollution in humans (Kim et al., 2005; Peters et al., 2001). Macrophages synthesize high levels of IL-6 suggesting they take part in the excessive inflammation in COVID-19 disease. “Macrophage Activation Syndrome” can shed a light on and clarify the reason of elevated serum concentrations of CRP, that are normally absent in viral infections (Paces et al., 2020) (Fig. 1B). The maximum concentration of IL-6, in addition to the elevated CRP concentrations, is an indicator of the requirement for mechanical ventilation. Thus, serum IL-6 or CRP concentrations are significant markers used in the follow-up of treatment in patients with COVID-19-related cytokine storm (Herold et al., 2020). Consequently, the concentrations of these markers demonstrate the disease seriousness and prognosis in patients with COVID-19 (Liu et al., 2020). In this condition, COVID-19 pneumonia with ARDS is distinctively described by hyperinflammatory syndrome. It has been suggested that tocilizumab, a monoclonal antibody that blocks the IL-6 receptor, provides a rapid and significant clinical improvement in respiratory failure associated with hyperinflammation (Toniatì et al., 2020). Due to the lack of standard treatment guidelines for COVID-19 patients, the treatment strategy should be designed by a multidisciplinary in-hospital group. As shown in many clinical trials, in addition to antiviral, antibacterial and glucocorticoid therapies, the combination with tocilizumab has favorable outcomes in seriously ill patients with respiratory failure (Chen et al., 2020).
2020a, 2020b; Luis et al., 2021; Salama et al., 2021). In this respect, tocilizumab, prevents the cytokine storm, increases oxygenation and also improves lung lesion opacity in COVID-19 patients (Sheppard et al., 2017; Xu et al., 2020).

However, Stone et al. (2020) have claimed that Tocilizumab is not an efficient treatment option in COVID-19. In this clinical trial, since the impact of risk factors are predominant in the tocilizumab group, it is difficult to decide whether early IL-6 receptor blockade is an ineffective treatment strategy in COVID-19 cases. Furthermore, because of the wide range of the confidence intervals in statistical comparisons, the benefit or harm of tocilizumab treatment in some patients is not clear (Stone et al., 2020).

On the one hand DEP exposure activates alveolar macrophages by inducing the release of the inflammatory cytokine TNF-α, on the other hand, TNF-α release is controlled by the NF-κB expression. Excess TNF-α release leads to macrophage apoptosis through the upregulation of NF-κB. In this vicious cycle, while DEP stimulates TNF-α gene expression, it provokes an apoptotic response in alveolar macrophages. Consequently, PM-induced pulmonary inflammation and injury proceeds (Kafoury and Madden, 2005). In this process, uptake of DEP by alveolar macrophages culminates in respiratory disturbances which lead to superoxide and H₂O₂ release. Released H₂O₂ and superoxide activates the mitogen-activated protein kinases/extracellular-regulated kinase kinase kinase-1 (MEKK-1) downstream from Ras. MEKK-1 has been demonstrated to activate inhibitor of nuclear factor kappa B (IkB) kinase (IKK) and the c-Jun N terminal kinases (JNK), that both induce NF-κB downstream. The activation of NF-κB and the transcription and release of TNF-α creates an autocrine loop (Kafoury and Madden, 2005; Malinin et al., 1997). Changes in the activities of NF-κB have a direct effect on the ability of TNF-α to provoke cell survival or death. On the other hand, death receptor stimulation of caspase-8 triggers apoptotic cell death (Varfolomeev et al., 2005). TNF stimulation also causes the necrosum formation. Later, the necrosum stimulates the mitochondrial complex I-mediated oxidative stress (reactive oxygen species (ROS) generation) and cytotoxicity. Increase in the NADPH (the reduced form of nicotinamide adenine dinucleotide phosphate) oxidases mediated ROS synthesis and its effect on the membrane-associated TNF receptor complex I results in calcium release, activation of phospholipase A2, lipid oxygenases, and acid sphingomyelinas. All these lead to lysosomal destabilization (Vandenabeele et al., 2010). Alveolar macrophage death by the mechanisms mentioned above is a principal determinant in the progression of pulmonary inflammation through its effect on the remaining immune cell populations in the respiratory system (Fan and Fan, 2018). Abnormal cellular immunity and humoral immunity; including neutrophilia, lymphocytopenia, low CD4 + T cells, and diminished C3 are prognostic factors predicting mortality of patients suffering from COVID-19 (Zhao et al., 2020). Exposure to DEP firstly activates the uptake of neutrophils and alveolar macrophages into the respiratory tract, then suppresses their function. Furthermore, DEP-induced migration of alveolar macrophages into the respiratory tract is together with reduction in CD3+CD25+ cells (Rudell et al., 1999). The reactivity, toxicity, and fibrogenic potential of particles in the lung depends on a variety of particle-related factors. Fibrotic reactions due to particles lead to the progressive deposition of collagen by pulmonary fibroblasts (Bonner, 2007). Thus, long-term DEP exposure increases the level of transforming growth factor-β (TGF-β), collagen content and lung fibrosis, and goblet cell hyperplasia (Kim et al., 2016). Consequently, DEP exposure contributes to the induction of the pulmonary innate immune system. Thus, exposing to dPM10 results in inflammation and fibrosis of the airway epithelium (Maglione et al., 2019). Indeed, TGF-β1 is predominantly expressed in the pathogenesis of pulmonary fibrosis. This cytokine regulates the formation and accumulation of all extracellular matrix (ECM) molecules such as collagen, fibronectin, elastic fibers, and ground substances (Coker et al., 1997; Roberts et al., 1999).

4. Clinical outcomes of dPM impact on SARS-CoV-2 infection

Considering the molecular mechanisms described above, SARS-CoV-2 infection can progress with both pulmonary and systemic inflammation. In the course of the COVID-19, cardio-pulmonary insufficiency, sepsis, ARDS and multi-organ dysfunction are the most prevalent serious complications in high-risk patients (Chen et al., 2020a, 2020b).

With the more severe cases requiring hospitalization, increased respiratory rate and decreased oxygen transport may advance to ARDS, frequently. Since, the ACE2 is intensely expressed in the human brainstem which consists of the medullary respiratory centers of the brain, it may cause additional respiratory distress in many COVID-19 patients (Lukiv et al., 2020). Thereby, mechanical ventilation is indispensable in these patients (Grasselli et al., 2020; Huang et al., 2020; Zhou et al., 2020). Bilateral pulmonary ground glass opacity (GGO) is a sign of lung involvement on computed tomography (CT) imaging in most COVID-19 patients. Indeed, these radiological findings are consistent with pulmonary inflammatory infiltration, and fibrosis (Fig. 1A). In a group of 99 cases with SARS-CoV-2 pneumonia, 74 patients presented bilateral pulmonary infiltration, 14 patients had numerous patchy hyperinflation zones and GGO, and one patient admitted with a diagnosis of pneumothorax, ARDS developed in 17 % of these patients. When the serial thorax CT images taken at least 6 days after the positive polymerase chain reaction (PCR) test are scored semi-quantitatively according to the grade of pulmonary pathologies, it has been demonstrated that higher CT scores indicate a higher mortality incidence (Chen et al., 2020a, 2020b; Huang et al., 2020; Li et al., 2020a, 2020b). Emerging multifocal GGO in CT scans are detected simultaneously with lymphopenia, high levels of alanine aminotransferase and aspartate aminotransferase, increased high-sensitivity CRP, procalcitonin, fibrinogen and Dimmers concentrations. Consistent with the above mentioned data, the histopathological examination of GGO revealed that the infiltrating cells comprise mainly of plasma cells and macrophages, lymphocytes are rare in addition to thickened alveolar septum and fibrous connective tissue proliferation (Cai et al., 2020). As there is currently no proven effective treatment option against pulmonary fibrosis in COVID-19, measures to reduce and limit the severity of the disease and preserve the lungs from other risk factors should be considered (Ojo et al., 2020).

5. Conclusion

Although SARS-CoV-2 infection shares some similarities with that of other viral diseases of lung, its pulmonary CT appearance and serum CRP levels are quite specific based on consecutive immune response of host to both DEP and causal virus exposure. Inter- and intralobular septal thickening with increased fibroplasia and connective tissue eventuate with the bilateral pulmonary parenchymal ground-glass appearance. DEP increases COVID-19 mortality by aggravating the pulmonary inflammation caused by SARS-CoV-2 with similar mechanisms.

CRediT authorship contribution statement

Baris Mustafa Poyraz: Writing - CT interpretation. Evren Doruk Engin: Writing - original draft. Ayse Basak Engin: Writing - original draft. Atila Engin: Review & editing & figure design-preparation.

Declaration of Competing Interest

The authors report no declarations of interest.

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