

Aftereffects in Epigenetic Age Related to Cognitive Decline and Inflammatory Markers in Healthcare Personnel with Post-COVID-19: A Cross-Sectional Study

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Purpose: Epigenetic age and inflammatory markers have been proposed as indicators of severity and mortality in patients with COVID-19. Furthermore, they have been associated with the occurrence of neurological symptoms, psychiatric manifestations, and cognitive impairment. Therefore, we aimed to explore the possible associations between epigenetic age, neuropsychiatric manifestations and inflammatory markers (neutrophil-lymphocyte ratio [NLR], platelet-lymphocyte ratio [PLR], monocyte-lymphocyte ratio [MLR], and systemic immune-inflammation index [SII]) in healthcare personnel with post-COVID condition.

Patients and Methods: We applied the Montreal Cognitive Assessment (MoCA) and Mini-Mental State Examination (MMSE) tests to 51 Mexican healthcare workers with post-COVID-19 condition; we also estimated their epigenetic age using the PhenoAge calculator.

Results: The participants had a post-COVID condition that lasted a median of 14 months (range: 1–20). High NLR (>1.73) had association with mild cognitive impairment by MMSE ($p=0.013$). Likewise, high MLR (>0.24) were associated with language domain in MOCA ($p=0.046$). Low PLR (<103.9) was also related to delayed recall in MOCA ($p=0.040$). Regarding comorbidities, hypertension was associated with SII ($p=0.007$), overweight with PLR ($p=0.047$) and alcoholism was associated with MLR ($p=0.043$). Interestingly, we observed associations of low PLR (<103.9) and low SII (<1.35) levels with increased duration of post-COVID condition ($p=0.027$, $p=0.031$). Likewise, increases in PhenoAge were associated with high levels of SII ($OR=1.11$, $p=0.049$), PLR ($OR=1.12$, $p=0.035$) and MLR ($OR=1.12$, $p=0.030$).

Conclusion: We observed neurocognitive changes related to inflammatory markers and increases in epigenetic age in healthcare personnel with post-COVID-19 condition. Future research is required to assess mental and physical health in individuals with post-COVID-19 symptoms.

Keywords: post-COVID-19, cognitive manifestation, inflammatory markers, epigenetic age

Introduction

Aging is a biological process that has been related to disease and mortality, which is reflected in molecular characteristics, epigenetic modifications, and telomere wear and tear.¹ However, individuals with the same chronological age exhibit differences

in their underlying biological aging process that could reflect different susceptibilities to age-related diseases.² The biological differences between individuals can be expressed via epigenetic age, an estimation of age in years resulting from a mathematical algorithm based on the DNA methylation.³ Although DNA methylation is the primary method for estimating epigenetic age, clinical biomarkers (biochemical parameters and blood cell count) can be used as a surrogate measure,² by using a calculator for epigenetic age. The Horvath-Levine PhenoAge Calculation combines the results of 9 blood parameters and uses the Horvath-Levine Blood Component PhenoAge Equation; its results correlate strongly with age in every tissue and cell tested.^{2,4} The epigenetic age could be altered due to viral infections and DNA methylation, which has been shown to correlate with the aging process and can be used to estimate epigenetic aging in tissues.¹ Similarly, several studies have shown increased epigenetic ages in COVID-19 survivors;^{5,6} these changes due to SARS-CoV2 could be short-lasting or permanent. Therefore, there should be a post-COVID-19 monitoring of survivors.⁶

Evidence indicates that COVID-19 is associated with neurologic symptoms, acute and chronic psychiatric manifestations, as well as cognitive impairment.^{7–9} COVID-19 induces persistent symptoms after the acute phase such as muscle pain and weakness, headache, sleep disturbances, palpitations, sleep problems, anxiety, low-grade fever, and physical and mental fatigue.^{7–9} Importantly, cognitive impairment is observed as one of the consequences in patients with COVID who developed mild symptoms.^{10–13} It has been reported that 48 to 56% of individuals who presented COVID-19, have a cognitive decline afterwards, particularly after a severe infection by SARS-CoV-2.^{14,15}

On the other hand, several studies have shown that a disruption of immune and inflammatory responses caused by SARS-CoV-2 infection can lead to long-term disorders.¹⁶ In this sense, inflammatory markers such as the systemic immune-inflammation index (SII), neutrophil-lymphocyte ratio (NLR), platelet-lymphocyte ratio (PLR) and monocyte-lymphocyte ratio (MLR) have been prognostic biomarkers in tumors, cardiovascular diseases, and COVID-19.^{17,18} Recently, the “post-COVID-19 condition” is considered when after three months of having the SARS-CoV2 infection, the individual has persistent symptoms for at least two months, and no alternative diagnosis has been made.^{8,19} Additionally, high levels of inflammatory markers have been observed in individuals with post-COVID-19 symptoms, indicating that there is a relationship between immunity and post-COVID condition.¹⁶

Evidence that individuals have post-COVID condition is increasing every day. However, to the best of our knowledge, there is a paucity of studies relating phenotypic age, inflammatory markers and cognitive characteristics in healthcare workers with post-COVID-19 condition. In this study, our aim was to explore the possible associations between neurocognitive manifestations, inflammatory markers and epigenetic age, in Mexican healthcare workers who present post-COVID-19 condition.

Materials and Methods

Participants

This is a descriptive and cross-sectional study. We recruited 51 healthcare workers with a previous diagnosis of COVID-19 who work at the Mental Health Regional High Specialty Hospital in Tabasco, Mexico; the data collection was from August to December 2021. We included healthcare personnel (physicians, nurses, chemists, and pharmacists) of 18–60 years of age, who had been diagnosed with a positive PCR test for SARS-CoV-2 and recovered from COVID-19 at least four weeks before the evaluation. The participants were diagnosed between March 2020 and October 2021 (range: 1–20 months post-COVID-19). Exclusion criteria were recent infection with SARS-CoV-2 confirmed by PCR test, diagnosis of chronic or recent acute infectious diseases, clinical history of autoimmune diseases or cancer, history of a mental health disorder, and previous diagnosis of neurocognitive impairment. A psychiatrist evaluated the mental health and neurocognitive impairment history of the participants.

Study Design

We interviewed the participants and gathered information about their medical history, sociodemographic data, symptoms of acute COVID-19 disease, and anthropometric measurements. We conducted face-to-face surveys to 51 healthcare workers: the Mini-Mental State Examination (MMSE) and Montreal Cognitive Assessment (MoCA) neurocognitive scales.^{20,21} We employed the Spanish version of the MoCA test, a validated scale for the Mexican population.²² MoCA

scores ≥ 26 indicate normal cognitive functioning, while lower scores indicate a mild neurocognitive impairment; 1 point is added to patients with ≤ 12 years of schooling. The MMSE scale used was a validated version for the Mexican population.²³ MMSE is associated with the identification of moderate to severe cognitive impairment; it has a maximum score of 30, scores > 24 points are normal, and ≤ 24 is neurocognitive impairment moderate to severe. Both the MoCA and the MMSE were obtained from the guidelines of the National Institute of Geriatrics in Mexico (INGER).²⁴

Epigenetic Age and Inflammatory Markers

From each participant, venous blood samples were taken in tubes with and without EDTA to determinate the inflammatory markers and epigenetic age. The hemogram test was determined with CELL-DYN Ruby (Abbott). Data from the hemogram was used to calculate SII, NLR, PLR, and MLR using the following formulas: NLR= neutrophils count/lymphocytes count, PLR= platelets count/lymphocytes count, MLR= monocytes count/lymphocytes count, and SII index= (platelets count \times neutrophils count)/lymphocytes count. We divided all inflammatory markers into low, medium, and high categories using tertiles: $< 3.16 \times 10^3$, $3.16 - 4.74 \times 10^3$ and $> 4.743 \times 10^3$ for SII, < 1.35 , $1.35 - 1.73$ and > 1.73 for NLR, < 103.9 , $103.9 - 134.8$ and > 134.8 for PLR, and < 0.1860 , $0.1860 - 0.2487$ and > 0.2487 for MLR, respectively. Stratifying the inflammatory ratios in tertiles is commonly used in literature.^{25,26}

We determined albumin (g/L), alkaline phosphatase (U/L), creatinine ($\mu\text{mol/L}$), fasting glucose (mmol/L), C-Reactive Protein (mg/L), using the Cm250 (Wiener Lab[®]). These biochemical parameters were required to calculate the phenotypic age. We utilized the Horvath-Levine PhenoAge Calculator to estimate phenotypic age, DNA methylation, Δ age (Phenotypic age - Chronological age), and 10-year mortality.⁴

Statistical Analysis

We expressed numerical variables as mean \pm standard deviation for variables with normal distribution, and median (interquartile range [IQR]) for variables with non-normal distribution. Categorical data was expressed as frequencies and percentages (%). We used the Shapiro–Wilk test to assess normality in our variables. To compare inflammatory markers tertiles, we used the Chi-squared test for categorical variables and ANOVA test for numerical data. Later, we performed ordinal logistic regression analysis for SII, NLR, PLR, and MLR; all models included the duration (in months) that the post-COVID-19 condition lasted, MMSE total score, PhenoAge, and associated features as confounding variables. Ordinal logistic regression results were expressed as odds ratio and 95% CI for the highest tertile of inflammatory markers. Comparisons of the PhenoAge estimations and the neurocognitive scores were made using ANOVA test and Kruskal–Wallis test. The effect size was estimated using Cramér’s V (V), Cohen’s D (d), Eta-squared (η^2), Cliff’s delta (δ) and ranked Eta-squared (η^2_H) (for Chi-squared, t-Student, ANOVA, Mann–Whitney U and Kruskal–Wallis tests, respectively). A p-value < 0.050 was considered significant for all the analyses. Shapiro–Wilk, Chi-squared, t-Student, ANOVA, Mann–Whitney U, and Kruskal–Wallis tests were made with SPSS v. 26. The effect size and logistic regression analyses were performed with R v.4.2. Word cloud of comorbidities was performed with the wordcloud package for R. Density plots and bump charts were performed with ggplot2, ggbum, and patchwork packages for R.

Ethical Considerations

We followed the Official Mexican Standard NOM-012-SSA3-2012 guidelines and the ethical principles of the Declaration of Helsinki to perform this research. Additionally, this study was approved by the ethics committee of the Mental Health Hospital “Villahermosa” in Mexico (HRAESM/DG/UEI/467/2021). The participants received verbal and written information about the objectives of this study. Every person participated voluntarily without receiving any remuneration and signed an informed consent.

Results

Sociodemographic and Clinical Characteristics of Healthcare Personnel with Post-COVID-19 Condition

This study included 51 healthcare workers (29 females and 22 males); their mean age was 40.57 (10.96) years. We also evaluated variables such as schooling (16.32 [4.08] years) and Body Mass Index (29.72 [6.52]). We observed that 92.15% (n=47) of

participants had mild symptoms of COVID-19 and required home treatment in social isolation, and only 1.96% (n=1) were treated in hospital. The duration of the post-COVID condition showed a median of 14 (4.5–16.5) months. According to the MoCA test, we identified 76.47% (n=47) healthcare personnel with mild cognitive impairment. Nonetheless, the MMSE showed that 7.84% (n=4) of healthcare workers had scores indicating mild neurocognitive impairment. (Table 1)

Inflammatory Ratios in Post-COVID-19 Healthcare Personnel

A high NLR was associated with mild cognitive impairment measured by MMSE ($p=0.013$, $V=0.36$); nonetheless, we did not observe an association with MoCA. Figure 1A shows the proportion of patients with and without cognitive impairment among inflammatory marker tertiles.

Table 1 Sociodemographic, Clinical Characteristics and Cognitive Assessment in Post-COVID-19 Mexican Healthcare Personnel

Variables	Post-COVID-19 (n=51)
Age (years)	40.57 (10.96)
Schooling (years)	16.32 (4.08)
Body Mass Index	29.72 (6.52)
Pharmacotherapy, n (%)	47 (92.15%)
Post-COVID months	14 (4.5–16.5)
Cognitive assessment	
Cognitive impairment MoCA, n (%)	39 (76.5%)
Cognitive impairment MMSE, n (%)	4 (23.52%)
Neurocognitive domains by MoCA	
Visuospatial	3 (3–3)
Attention	5 (3–6)
Language	2 (1–2)
Abstraction	2 (1–2)
Delayed recall	2 (1–3)
Orientation	6 (6–6)
MoCA score	22.86 (3.34)
Neurocognitive domains by MMSE	
Orientation	10 (10–10)
Registration	3 (3–3)
Attention	5 (4–5)
Language	5 (5–5)
Recall	5 (4–6)
MMSE score	28 (27–29)
PhenoAge Calculator	
Phenotypic Age	39.47 (13.07)
DNA _m Phenoage	38.86 (13.23)
Δ age	–1.10 (5.48)
10-year mortality	2.20 (0.94–4.61)
Inflammatory indexes	
SII	430294 (288,358–514,052)
NLR	1.51 (1.22–2.02)
PLR	122.05 (42.64)
MLR	0.22 (0.17–0.26)

Notes: Numerical variables are expressed as mean (standard deviation) for variables with normal distribution, and median (interquartile range) for variables with non-normal distribution.

Abbreviations: n, frequency; MoCA, Montreal Cognitive Assessment; MMSE, Mini Mental State Examination; SII, systemic immune-inflammation index; NLR, neutrophil-lymphocyte ratio; PLR, platelet-lymphocyte ratio; MLR, monocyte-lymphocyte ratio.

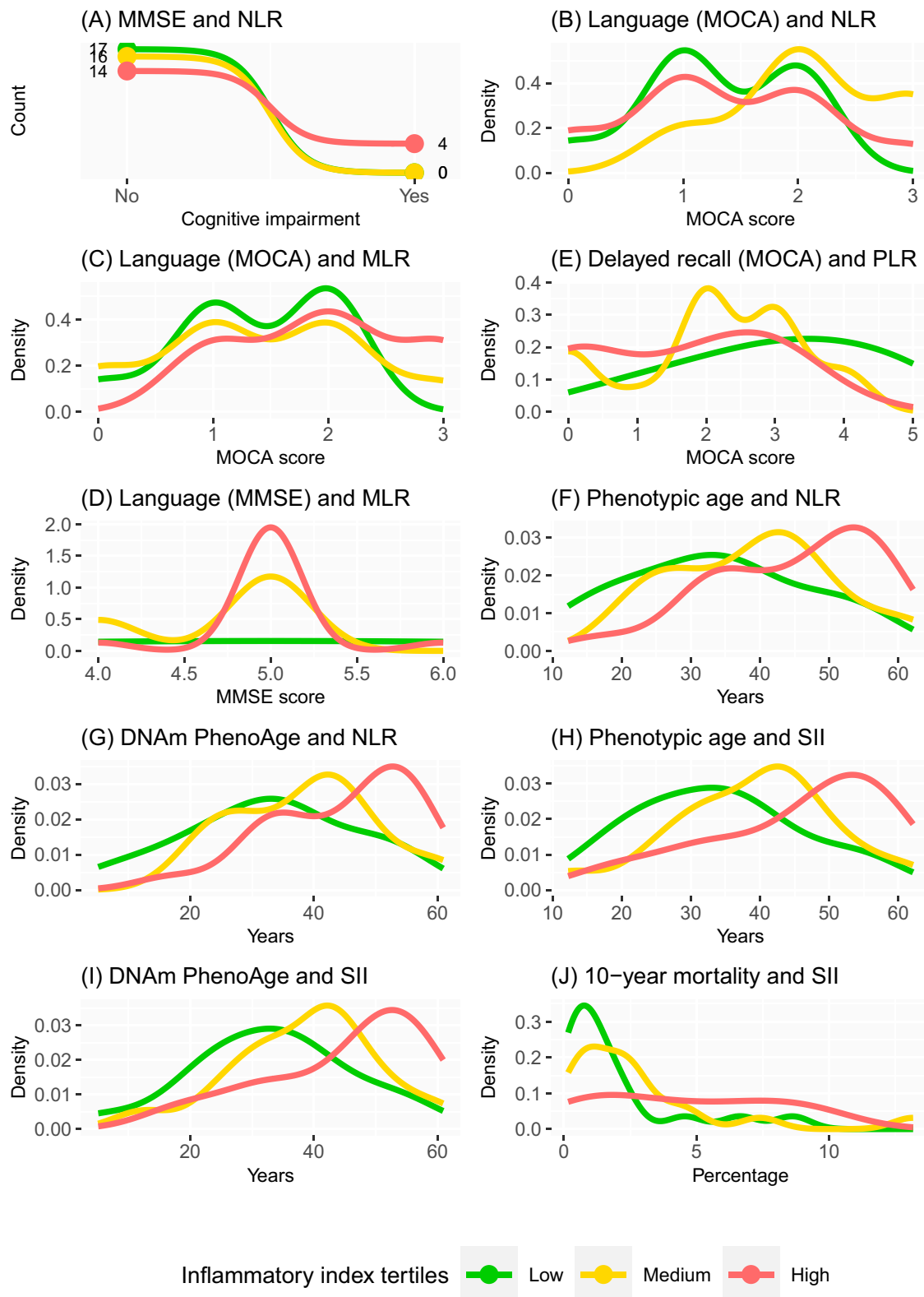


Figure 1 Associations between inflammatory markers' tertiles, cognitive and epigenetic age features.

Notes: (A–E) Cognitive domains and inflammatory markers' distributions. (F–J) PhenoAge features and inflammatory markers' distributions. All features were significant in Chi-square and ANOVA test ($p < 0.05$).

Cognitive domains showed different associations. Figure 1B–E shows the association between the inflammatory markers tertiles and the cognitive domains scores. The language domain in MoCA was associated with medium NLR and high MLR (respectively: $p=0.022$, $\eta^2_H=0.12$; $p=0.047$, $\eta^2_H=0.12$); likewise, low PLR levels were associated with a delayed recall in MoCA ($p=0.040$, $\eta^2_H=0.09$). In addition, high MLR levels were significant in language score measured by MMSE ($p=0.046$, $\eta^2_H=0.02$).

The most common comorbidities presented in this population were overweight and obesity ($n=22$, 43.14%), hypertension ($n=11$, 21.57%), alcoholism ($n=7$, 13.73%), and diabetes ($n=4$, 7.84%). The analysis of inflammatory markers indicated that high levels of SII were associated with hypertension ($p=0.007$, $V=0.40$) (Supplementary Figure S1A). Overweight (BMI 25–29.9) had significant association with high PLR ($p=0.047$, $V=0.29$). Notably, individuals with overweight in our sample had higher mean NLR, MLR and SII compared with obesity and normal weight; nonetheless, we found not significant differences. Also, alcoholism was associated with medium MLR levels ($p=0.043$, $V=0.29$). Regarding pharmacotherapy, we found that medium MLR values were associated with the use of hydroxychloroquine for COVID-19 acute treatment ($p=0.041$, $V=0.29$). Overall, the most frequent post COVID-19 symptoms were dysgeusia ($n=41$, 80.39%), anosmia ($n=39$, 76.47%), and headaches ($n=36$, 70.59%). Some symptoms were associated with different levels of NLR: high NLR levels were associated with sudden onset ($p=0.005$, $V=0.40$), rhinorrhea ($p=0.005$, $V=0.41$) and abdominal pain ($p=0.025$, $V=0.33$), while low levels of NLR were associated with arthralgia ($p=0.048$, $V=0.29$). (Supplementary Figure S1B–S1H). Interestingly, we observed associations of low PLR and SII levels with longer duration (in months) of the post-COVID-19 condition (respectively: $p=0.027$, $\eta^2=0.14$; $p=0.031$, $\eta^2=0.13$). (Supplementary Figure S1I–S1J)

Epigenetic Age in Healthcare Personnel with Post-COVID-19 Condition

We observed a slightly higher chronologic age (40.55 [11.0]) than the phenotypic age (39.38 [13.12]) and DNAm PhenoAge (38.78 [13.28]) in healthcare workers with post-COVID-19 condition. The mean Δ age was -0.62 (5.55), and the mortality for 10 years was 3.16 (3.17). Notably, we found associations between high SII levels and higher phenotypic age, as well as DNAm PhenoAge (respectively: $p=0.048$, $\eta^2=0.12$; $p=0.038$, $\eta^2=0.13$); while high NLR was associated with DNAm PhenoAge ($p=0.049$, $\eta^2=0.12$). Likewise, high SII levels increased the 10-year mortality ($p=0.044$, $\eta^2_H=0.12$) (Figure 1F–J).

Logistic Regression: Epigenetic Age and Inflammatory Markers

The duration (in months) of post-COVID-19 condition showed strong associations with decreased levels of SII (OR=0.87, 95% CI: 0.78–0.96, $p=0.008$), NLR (OR=0.85, 95% CI: 0.75–0.96, $p=0.008$) and PLR (OR=0.86, 95% CI: 0.77–0.96, $p=0.005$). Regarding cognitive impairment, increases in MMSE total score resulted in higher PLR (OR=1.19, 95% CI: 1.00–1.42, $p=0.046$). Remarkably, arthralgia reduced NLR (OR=0.19, 95% CI: 0.05–0.83, $p=0.028$). On the other hand, rhinorrhea (OR=5.46, 95% CI: 1.11–26.82, $p=0.037$) and abdominal pain (OR=29.76, 95% CI: 2.23–396.23, $p=0.010$) increased risk for high NLR. In comorbidities, hypertension was associated with high SII (OR=10.34, 95% CI: 1.49–72.02, $p=0.018$). Interestingly, one-year increases in PhenoAge were associated with high levels of SII (OR=1.11, 95% CI: 1.00–1.23, $p=0.049$), PLR (OR=1.12, 95% CI: 1.01–1.25, $p=0.035$) and MLR (OR=1.12, 95% CI: 1.01–1.24, $p=0.030$).

Epigenetic Age and Neurocognitive Domains

We associated the neurocognitive scores with the PhenoAge estimations. Interestingly, we found that lower delayed recall scores (MoCA) were related to older phenotypic age, DNAm Phenoage, and chronological age ($p<0.005$, $\eta^2=0.33$ in all tests); also, higher 10-year mortality showed lesser scores in this domain ($p=0.004$, $\eta^2_H=0.27$) (Table 2). In addition, healthcare personnel with mild cognitive impairment by MoCA scores had higher phenotypic age (41.91 [12.91] vs 31.54 [10.51], $p=0.014$, $d=0.84$), DNAm PhenoAge (41.45 [12.63] vs 30.45 [11.99], $p=0.010$, $d=0.88$), chronological age (42.56 [10.60] vs 34.08 [9.89], $p=0.017$, $d=0.81$) and 10-year mortality (2.41 [1.0–5.8] vs 0.95 [0.43–1.38], $p=0.011$, $\delta=0.49$). We did not find associations between MMSE domains and the PhenoAge estimations.

Table 2 Associations Between PhenoAge Estimations, Chronological Age, and Delayed Recall Cognitive Domain

Delayed Recall (MoCA)							F / H	p	η^2
Score	0	1	2	3	4	5			
Phenotypic age	47.78(8.40)	42.21(17.06)	46.81(11.31)	31.91(12.29)	29.72(5.83)	36.08(10.93)	4.53	0.002	0.33
DNAm PhenoAge	47.47(2.71)	41.66(7.46)	46.09(3.04)	30.92(3.57)	29.53(2.16)	35.75(6.21)	4.48	0.002	0.33
Chronological age	46.22(3.70)	44.20(13.97)	47.15(9.75)	35.14(10.23)	32.00(5.77)	34.33(15.04)	4.36	0.003	0.33
10-year mortality	35.61	29.2	34.35	17.89	13.17	22.17	17.24	0.004	0.27

Notes: Results for phenotypic age, DNAm PhenoAge and chronological age are expressed as mean (standard deviation), ANOVA test was performed. Results for 10-year mortality are expressed as mean rank, Kruskal–Wallis test was done.

Abbreviation: MoCA, Montreal Cognitive Assessment.

Discussion

Epigenetic Age and Inflammatory Markers in Healthcare Personnel with Post-COVID-19

Data from this research may contribute to understand the post-COVID condition shown by health care workers in relation to epigenetic age, inflammatory markers and cognitive characteristics. For instance, we obtained an association between increased epigenetic age and high SII, NLR, MLR in healthcare workers with post-COVID-19 condition. Literature indicates that methylation derived NLR is a marker of high cardiovascular risk; while high SII and NLR values were found in non-surviving COVID-19 patients.²⁷ Koestler et al have suggested that DNA methylation could be used to estimate NLR and MLR in cancer.²⁸ In our study, the 10-year mortality risk (calculated with PhenoAge) was associated with SII in individuals with post-COVID-19 condition; in this sense, it has been suggested that epigenetic age acceleration and inflammatory markers are associated with the severity of COVID-19.^{29,30} It is known that epigenetic age in blood reflects both, the chronological age and aspects of the biological age; a higher epigenetic age is associated with a higher mortality and the risk of age-related comorbidities. The epigenetic changes due to COVID-19 include increases in epigenetic age, methylation regulation of Angiotensin Converting Enzyme 2, and immune response genes.³¹ In this regard, it has been observed that SARS-CoV-2 induces changes in DNA methylation, generating the expression of immune response inhibitor genes; also, individuals with COVID-19 present a downregulation of ACE2, while in individuals with post-COVID-19 condition the expression of ACE2 mRNA is reduced. However, in young individuals with post-COVID-19 condition, an accelerated DeltaAge correlated with lower ACE2 mRNA levels, indicating an adverse effect of DNAmAge on peripheral blood ACE2 density.³²

Cao et al suggested that the DNA methylation changes due to SARS-CoV-2 infection may affect the expression of the metabolic process and epigenetic aging due to COVID-19;⁵ these authors found a reversible influence of epigenetic aging and COVID-19 in some patients with post-COVID-19 condition. Similarly, previous studies have shown that HIV and SARS-CoV-2 infections alter the host cell epigenetic landscape. All these findings suggest that COVID-19 may be involved in the epigenetic clock.¹ Nonetheless, some studies have been unsuccessful in associating increased epigenetic age with a higher risk for severe COVID-19.^{32,33}

Neurocognitive Symptoms and Inflammatory Ratios in Post-COVID-19 Healthcare Personnel

We found a neurocognitive decline in healthcare workers with post-COVID-19 condition when assessed with MoCA. Various studies suggest that COVID-19 is associated with neurological, psychiatric manifestations and cognitive impairment.^{34,35} Bolattürk & Soyulu (2022) showed that an early-stage cognitive impairment was detected in 15% of patients with COVID-19 by MMSE, and neurocognitive impairment was detected in 55% of patients by MOCA.³⁶ Other findings have shown some dysfunction of episodic memory, visuospatial domain, slowness for ideation, and alteration of executive functions in patients previously hospitalized for COVID-19.³⁷ Crunfli et al, have reported that 34 to 56% of individuals who recovered from COVID-19 (not hospitalized) presented impaired cognitive function after the diagnosis of COVID-19 (59 days later).³⁸ Interestingly, a study in northern Israel found a characteristic cognitive profile of post-COVID condition related to executive function, language, and attention.¹³ Cecchetti et al suggested that COVID-19

increases the fragility of brain structures involved in memory processes.³⁹ Other research on post-COVID-19 individuals showed that executive function, language and attention scores were significantly worse than in individuals without post-COVID-19 symptoms.⁴⁰ In this sense, other findings have shown the dysfunction of episodic memory, visuospatial domain, slowness for ideation and alteration of executive functions in patients previously hospitalized for COVID-19.³⁷

In our study, higher NLR, PLR and MLR were associated with neurocognitive decline (language and delayed recall. Many authors have suggested that inflammation is associated with cognitive dysfunction; in fact, a high NLR has recently been described in mild cognitive impairment. Some studies have shown that NLR is significantly higher in patients with dementia than in controls. Another study reported a correlation between NLR and cognitive decline in individuals with small cerebral blood vessel disease.^{41–43} High levels of neutrophils characterize chronic inflammation, reflecting the intensity of stress and systemic inflammation (chronic activation of the innate immune system); while a low level of lymphocytes may be indicative of a reduced response to pathogens.⁴⁴ Maamar et al mentioned that post-COVID-19 condition involve inflammatory mechanisms, immune responses and psychological factors.¹⁶ Consistent with our findings, a study showed that high SII was a predictor of developing depression and anxiety in patients with TB.⁴⁵ In contrast, a reduction in SII observed between admission and discharge may predict the alleviation of depression among survivors of coronavirus disease.⁵ These findings suggest a consistent association between inflammation and mental disorders.

We found that a longer duration of post-COVID condition (measured in months) was associated with lower SII, NLR, and PLR indices. Previous studies indicate that PLR, NLR and platelets could be efficacious markers of the severity and prognosis of COVID-19.^{46,47} NLR, MLR, PLR and SII have also been proposed as biomarkers to assess the severity, morbidity, mortality and prognosis of psychiatric and non-psychiatric illnesses.⁴⁸ Likewise, it has been reported that NLR and other inflammatory markers decrease after a few weeks compared to acute COVID-19 levels.^{49,50} Although SII, NLR and PLR are suggested to predict the risk of severe COVID-19 and mortality, the inflammatory indices could decrease over time in COVID-19 survivors.^{30,49,51} According to this, individuals with post-COVID condition have shown a relationship between the severity of deterioration and age, where younger patients show impairment in some cognitive domain more frequently than older ones.^{15,52} Surprisingly though, young people were considered to have better expectations, because a younger age induces a better prognosis in dementia and acquired brain damage. Herrera et al mentions that cognitive difficulties after SARS-Cov2 infection continue for up to two years or more.¹⁵ In contrast, patients with a longer evolution time showed better results in verbal learning and visual memory recall.

At the same time, we identified that hypertension was associated with SII in healthcare personnel with post-COVID-19 condition. Other authors found that SII may be a better indicator than NLR and PLR in treatment-naïve hypertensive patients¹⁷ and patients with coronary heart disease.⁵³ We also found associations between overweight and PLR. Metabolic syndrome and other comorbidities combined with obesity (eg polycystic ovary syndrome, osteoporosis and cancer) are related with increased levels of inflammatory markers.^{54–57} Although some studies report that overweight and obesity alone increase the levels of NLR, PLR and SII, these associations are not consistent.^{58–60} Figure 2 shows the most frequent comorbidities with the largest font, while less frequent comorbidities appear with decreasing font size. Various studies have shown that individuals with comorbidities or chronic diseases are more likely to develop post-COVID-19 condition.^{19,61} Finally, NLR was associated with rhinorrhea, abdominal pain and arthralgia in our analysis. Maamar et al found differences in inflammatory markers and frequencies of symptoms in mild COVID-19; nonetheless, they reported that high NLR was related to anosmia, ageusia, and fatigue.¹⁶

Some studies indicate that acute viral infection with SARS-CoV-2 alters the inflammatory and immune response, and this could also cause post-COVID 19 condition.¹⁶ Proinflammatory factors released by peripheral cells (lymphocytes, monocytes, neutrophils) are known to participate in neuroinflammation by activating astrocytes and microglia and modulating brain areas involved in mood regulation. Proinflammatory factors also decrease the concentration of intracerebral monoamines, trigger neuroendocrine responses, promote excitotoxicity (increased glutamate) and impair brain plasticity.⁶² Evidence links cortical (frontal, temporal and parietal cortex) and blood brain barrier dysfunction in individuals with post-COVID-19 condition with cognitive impairment. This dysfunction has been suggested to be related to vasculitis, hyperinflammation (cytokines), coagulopathy, and cerebral complications (cerebral hypoperfusion,



Figure 2 Comorbidities in post-COVID-19 Mexican healthcare workers.

Notes: The font size differences indicate changes in frequencies of comorbidities. The most frequent comorbidity, overweight (n=24), is eight times bigger than the least frequent comorbidity, asthma (n=2).

hypometabolism, and hypoxia) induced by SARS-CoV-2 infection. Altogether, proinflammatory factors can be involved in cognitive impairment in individuals with post-COVID 19 condition.^{39,63,64}

This study has some limitations. We observed a limited participation of healthcare personnel with post-COVID-19 condition; additionally, healthcare personnel showed lack of free time to participate in the study, then, our sample size is small. Another limitation is that possible cofactors could not be considered in our multivariable analysis. We could not establish causal relationships because of the cross-sectional nature of this study. Finally, our results cannot be extrapolated to the Mexican population as they come from one specific sample.

Conclusion

We observed that epigenetic age and 10-year estimated mortality were associated with altered concentrations of inflammatory markers (NLR, PLR, MLR and SII). In addition, inflammatory markers were associated with the duration of post-COVID condition, neurocognitive decline (language and delayed recall) and comorbidities. With all these findings, we suggest that the assessment of epigenetic age, inflammatory markers and neurocognitive scales could help to elucidate the neuropsychiatric sequelae in healthcare personnel with post-COVID-19 condition. Future research is required to evaluate if these alterations are maintained, improved, or worsen over time in post-COVID-19 condition.

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Disclosure

The authors report no conflicts of interest in this work.

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