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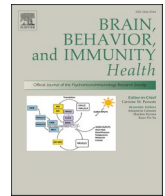
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## Blood parameters differentiate post COVID-19 condition from Myalgic Encephalomyelitis/Chronic Fatigue Syndrome and Fibromyalgia

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### ABSTRACT

Post-COVID-19 condition, such as Myalgic Encephalomyelitis/Chronic Fatigue Syndrome (ME/CFS) and Fibromyalgia (FM), are characterized by fatigue, pain, shortness of breath, sleep disturbances, cognitive dysfunction and other symptoms, heavily impacting on patients daily functioning. Moreover, over half of patients end up fulfilling ME/CFS and/or FM clinical criteria after a few months of SARS-CoV-2 infection. Expression of the toxic human endogenous retrovirus (HERV)-W ENV protein can be induced by viral infection and HERV-W detection was correlated with acute COVID-19 severity and found significantly expressed in post-COVID-19 condition. This study shows that HERV-W ENV may also be present in prepandemic cases of ME/CFS, FM or co-diagnosed with both clinical criteria, suggesting viral participation in these chronic diseases. To learn whether associated antiviral mechanisms may also show differing patterns of immunological responses, we measured IgM, IgG, IgA and IgE antibody isotypes against SARS-CoV-2 spike and nucleocapsid antigens, the levels of IL-6, IL-8, IL-10, IFN $\gamma$  and TNF $\alpha$  cytokines, the level of NfL, a neural damage biomarker, as well as some blood cell markers potentially related with fatigue. Importantly, some of the measured variables showed a capacity to discriminate post-COVID-19 condition cases from all other participants, with 100 % sensitivity and up to 71.9 % specificity providing a new tool for a differential diagnosis between diseases or syndromes with so many overlapping clinical symptoms. Interestingly, the detected markers showed moderate-to-strong correlations with patient symptoms pointing at novel therapeutic opportunities.

### 1. Introduction

Post-COVID-19 condition (ICD-11 code RA02), also known as Long COVID, is a post-viral syndrome first described by Soriano et al., in September 2020 (Soriano et al., 2022). It is characterized by a wide range of debilitating symptoms, including fatigue, pain, shortness of breath, sleep disturbances, and cognitive dysfunction, significantly impairing patients' daily functioning (Venkatesan, 2021). This chronic idiopathic condition triggered by SARS-CoV-2 infection lacks diagnostic biomarkers and current treatments are palliative and focused on alleviating symptoms, resembling the scenario of Myalgic

Encephalomyelitis/Chronic Fatigue Syndrome (ME/CFS) (ICD-11, code 8E49) (Sukocheva et al., 2022; Unger et al., 2024) and Fibromyalgia (FM) (ICD-11, code MG30.01) (Khoja et al., 2024). In fact, over 50 % of patients diagnosed with post-COVID-19 condition eventually meet the diagnostic criteria for ME/CFS (Dehlija and Guthridge, 2024) and/or FM (Khoja et al., 2024) after several months of illness progression. An additional aspect that further complicates the diagnosis of these patients is the frequent co-appearance of ME/CFS and FM, with co-diagnosis rates reaching at or over 50 % of the cases (Giménez-Orenga et al., 2022; Natelson, 2019) posing significant challenges for healthcare systems.

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Prior to the COVID-19 pandemic, the estimated prevalence of ME/CFS ranged from 0.17 to 0.89 % of the general population (Lim et al., 2020), and it was over 2 % for FM (Clauw, 2014; Lim et al., 2020), affecting preferentially women by at least 1.5 (female/male ratio) (Chen et al., 2022; Clauw, 2014; Fors et al., 2024; Lim et al., 2020). In the aftermath of the widespread global infection by SARS-CoV-2 and with estimates of 10 % or more of infected individuals that will develop chronic symptoms (Chen et al., 2022), the future epidemiological landscape of these chronic conditions remains uncertain. Furthermore, the continuous appearance of novel SARS-CoV-2 variants, the established presence of flu-seasonal viruses, and the increased exposure of the population to other infectious agents due to globalization, highlights the urgency to understand why and how some individuals fail to fully recover following infection.

Similar to other viral-triggered diseases such as Multiple Sclerosis (MS), patients with post-COVID-19 express the toxic envelope protein of the human endogenous retrovirus (HERV-W ENV) (Bjornevik et al., 2022; Charvet et al., 2023; Giménez-Orenga et al., 2022). Moreover, SARS-CoV-2 has been shown to activate HERV expression (Marston et al., 2021) and HERV-W ENV levels correlated with patient severity in the acute phase (Balestrieri et al., 2021; Petrone et al., 2023), suggesting a detrimental contribution to the response in certain individuals. Beyond SARS-CoV-2 and the MS-associated Epstein-Barr virus, other viral infections have also been linked to aberrant HERV expression (Macchietto et al., 2020) by mechanisms affecting, at least partly, epigenetic homeostasis (Chen et al., 2023; Giménez-Orenga and Oltra, 2021), which, if unreversed, may lead to a chronic state of the host (Giménez-Orenga and Oltra, 2021; Ovejero et al., 2020).

Upregulation of HERV expression for the Long COVID-related ME/CFS and FM diseases had also been reported at the transcriptional level (Ovejero et al., 2020; Rodrigues et al., 2019; Giménez-Orenga et al., 2025). Furthermore, preliminary findings by our group showed that HERV-W ENV is also overexpressed in some pre-pandemic cases of ME/CFS at the protein level (Giménez-Orenga et al., 2022) raising the possibility of a common mechanism triggered by viral infections in susceptible individuals, leaving the host in a prolonged state of immune activation while driving multi-symptom illness, name it post-COVID-19 condition, ME/CFS or FM.

To further investigate molecular similarities and/or differences that that could aid in the subtyping or stratification of patients affected by one or more of these three diseases, we extended our studies to the analysis of HERV-W ENV soluble oligomeric protein circulating in the serum of 193 post-pandemic females. This included 147 patients either diagnosed with post-COVID-19 condition ( $n = 33$ ), ME/CFS ( $n = 12$ ), FM ( $n = 42$ ), or co-diagnosed with MECFS and FM ( $n = 60$ ); and 46 matched healthy female controls. In this same cohort we measured SARS-CoV-2 antigenemia (IgM, IgG, IgA and IgE), circulating levels of selected cytokines (IL-6, IL-8, IL-10, IFN $\gamma$  and TNF $\alpha$ ) and concentration of neurofilament light chain (NFL), a marker of neural damage. All potentially relevant factors to characterize a chronic post-viral status. In addition, we measured red blood cell markers, previously reported as altered in some COVID-19 cases (Lechuga et al., 2023) to explore possible correlations with infection-associated molecular markers.

The results confirmed detection of HERV-W ENV antigenemia in subgroups of patients across all four patient study groups (post-COVID-19 condition, ME/CFS, FM and co-diagnosis) with the highest expression observed in post-COVID-19 condition. Importantly some of the measured variables showed ability to discriminate post-COVID-19 condition cases from all other participants, achieving 100 % sensitivity and up to 71.9 % specificity thereby providing utility for its differential diagnosis. Interestingly, the detected markers showed moderate-to-strong correlations with patient symptoms supporting their relevance to disease mechanisms.

## 2. Materials and Methods

### 2.1. Study design

This cross-sectional observational study involves the analysis of platelet-poor plasma from 193 post-pandemic adult female donors to measure HERV-W ENV protein levels and assess their association with SARS-CoV-2 serology, cytokine levels, and neurofilament light chain (NFL), as well as their correlation with associated clinical data. Participant study groups consisted of post-COVID-19 condition cases ( $n = 33$ ), patients having received diagnosis of ME/CFS ( $n = 12$ ), FM ( $n = 42$ ), or both ( $n = 60$ ), in addition to matched healthy controls ( $n = 46$ ). Clinical diagnosis for ME/CFS and FM preceded COVID-19 pandemic and complied Canadian (Carruthers et al., 2003) and/or International criteria (Carruthers et al., 2011), and/or American College of Rheumatology (ACR) 2011 criteria (Wolfe et al., 2010), respectively. It must be highlighted that all ME/CFS, FM and co-diagnosed cases had received clinical diagnosis before pandemic even though all samples were collected post-pandemic. Each participant donated about 10 ml of blood under fasting conditions, having dropped all medication or supplements at least 12 h before venipuncture. Reasons for exclusion included pregnancy, diagnosis of autoimmune disease, or receiving chronic treatment with monoclonal or immunomodulatory or immunosuppressive drugs, including systemic corticosteroids. Patients filled standard validated questionnaires including FIQ (Fibromyalgia Impact Questionnaire) (Burckhardt et al., 1991; Rivera and González, 2004), MFI (Multi Fatigue Inventory) (Smets et al., 1995), for assessment of pain and fatigue related domains, SF-36 (McHorney, Ware and Raczek, 1993) for quality of life, and COMPASS-31 (Sletten et al., 2012) for evaluation of dysautonomia. In addition, a small aliquot of patient donations was subjected to routine clinical blood analytics for a fine phenotyping of participants (Supplementary Table S1).

The study was approved by the Public Health Research Ethics Committee DGSP-CSISP of Valencia, Valencia, Spain, ref. 20220128/05/02. All samples and data from patients included in this study were provided by the Biobank IBSP-CV, integrated in the ISCIII Biobanks and Biomodels Platform and in the Valencian Biobanking Network and they were processed following standard operating procedures with the appropriate approval of the Ethics and Scientific Committees. All participants signed the corresponding informed consent.

### 2.2. Blood collection and poor-platelet plasma isolation

Blood, obtained by standard phlebotomy in K2EDTA tubes (BD cat. 366643 or Vacuette cat. 455045), was kept at RT and processed within 4 h. First it was centrifuged at  $500\times g$  for 15 min to separate plasma from the remaining components. Then, the upper phase was transferred to another tube and re-centrifuged at  $8000\times g$  for 10 min to remove contaminating platelets and other debris, and aliquoted in cryovials (0.5–1 ml) and kept frozen at  $-80\text{ }^{\circ}\text{C}$  until use, as previously described (Ovejero et al., 2020).

### 2.3. Detection of HERV-W envelope protein and SARS-CoV-2 antigen serology

For the detection of HERV-W ENV antigen in plasma and the quantification of its circulating soluble form, all analyses were performed according to the conditions provided in the patent published under ref. WO2019201908 (A1) and entitled "Method for the detection of the soluble hydrophilic oligomeric form of HERV-W envelope protein", as previously described (Tamouza et al., 2021). The specific signal was expressed as the signal to noise (S/N) ratio, where the noise represents the mean+2SD of the background signals yielded by a panel of healthy control samples. Both HERV-W ENV antigenemia and SARS-CoV-2 serology were performed by Simple Western technology according to immunoglobulin class. Specific antibodies against SARS-CoV-2 in

plasma were detected using recombinant nucleocapsid and spike proteins as target antigens. All samples were stored in freezers at  $-80^{\circ}\text{C}$  for less than a year. All samples were kept frozen at all times after their initial freezing until use (single freeze/thaw cycle before the immunoassay).

#### 2.4. Cytokine and NFL levels

Levels of cytokines and NFL were quantified using the fully automated immunoassay platform, Ella (ProteinSimple/Bio-technie, San Jose, CA, USA) (Rodrigues et al., 2019; Lechuga et al., 2023). All proteins were quantified using a single disposable microfluidic SimplePlex™ cartridge, which holds up to 32 serum samples per assay. The serum samples were thawed on ice and diluted in sample according to manufacturer's instructions. Within the cartridge, each serum sample gets further divided into four unique microfluidic parallel channels that are specific for each of the four proteins being analyzed. Each protein channel contains three analyte-specific glass nanoreactors (GNRs), which allows for each serum sample to be run in triplicate for each of the four protein samples. The obtained data were displayed as pg/mL and automatically calculated by Explorer Ella software.

#### 2.5. Statistical analysis and plotting

All statistical analysis and plots were performed with RStudio software version 4.2.1. Continuous data are expressed as means  $\pm$  SD, as indicated. Normal data distribution was assessed using the Shapiro-Wilk test. Statistical differences were determined using two-tailed unpaired Student's t-tests or Mann-Whitney-Wilcoxon tests, depending on whether the data followed or not normal distribution, respectively. Variable plots were drawn with ggplot2 v 3.5.1. Correlation analysis was performed with the package Hmisc v5.1. and plotted with ggpubr v0.6.0.999. Response operating characteristic (ROC) analysis with area under curve (AUC) were used to assess the discriminating capacity of the variable or set of variables and were performed with the R package PROC version 1.18.5.

### 3. Results

#### 3.1. Demographics and clinical characteristics of patient cohorts

A total of 193 female subjects were evaluated in this study corresponding to 33 post COVID-19 condition cases; 12 ME/CFS cases, not fulfilling FM ACR criteria (Wolfe et al., 2010); 42 FM cases, not fulfilling ME/CFS criteria (Carruthers et al., 2003; Carruthers et al., 2010); 60 cases fulfilling both, ME/CFS and FM criteria (Carruthers et al., 2003; Wolfe et al., 2010), which from now on will be called "co-diagnosis" study group; and 46 healthy matched subjects. Mean age by group in years  $\pm$  SD [range], were  $49.3 \pm 6.2$  [40–63],  $47.8 \pm 13.7$  [20–64],  $55.1 \pm 9.0$  [36–72], and  $53.1 \pm 9.7$  [26–71] respectively, for the four patient groups, and  $54 \pm 9.5$  [39–69] for the healthy control group (Supplementary Table S1).

Patient phenotyping using validated standard questionnaires to assess pain impact, fatigue, quality of life and dysautonomia is illustrated on Table 1. The highest total FIQ scores were observed in the co-diagnosed group, followed by those with FM, ME/CFS and Post-COVID-19. Based on established thresholds (FIQ scores  $<39$  indicating mild impact, 39–59 moderate, and  $>59$  severe (Rivera and Gonzalez, 2004) approximately 82 % of cases in the cohort fell into the severe category, with 12 % classified as moderate (Supplementary Table S1). According to MFI and SF-36 scores, the ME/CFS and co-diagnosed study groups seem the most affected. In contrast, the highest COMPASS31 scores were associated with FM and co-diagnosed study groups, suggesting a higher degree of dysautonomia in these populations. Statistical analysis revealed significant differences across several subdomains of these instruments, particularly between the post-COVID-19 condition and the

co-diagnosed groups. Notably, differences were found in 6 out of 8 SF-36 subdomains and 6 out of 7 COMPASS-31 subdomains (Supplementary Fig. S1). In contrast, fewer differences were observed in pain using the FIQ (3 out of 4 subdomains) and in fatigue assessment via the MFI, where only the 'General Fatigue' subdomain (1 out of 5) showed significant variation.

#### 3.2. Increased HERV-W ENV expression in all patient groups

Building on previous findings that demonstrated expression of the HERV-W ENV protein in acute COVID-19 patients with severe disease (Balestrieri et al., 2021; Petrone et al., 2023) and in post-COVID-19 individuals (Charvet et al., 2023; Giménez-Orenga et al., 2022), along with literature suggesting a potential progression from post-COVID-19 conditions to ME/CFS and/or FM (Khoja et al., 2024; Sukocheva et al., 2022; Unger et al., 2024), this study aimed to compare HERV-W ENV levels and their associations with additional variables—including SARS-CoV-2 serology, circulating cytokines, clinical red blood cell parameters, and patient-reported symptoms—between post-COVID-19 cases and patients diagnosed with ME/CFS, FM, or both. The goal was to explore potential molecular relationships that could inform diagnostic differentiation.

For this, circulating levels of HERV-W ENV protein were measured in our four study groups described above by Simple Western technology (see Methods section, and (Giménez-Orenga et al., 2022) for details). As shown in Fig. 1, almost half of post COVID-19 cases presented HERV-W ENV antigenemia (48.8 %) (16/33), compared to only 8.9 % (4/45) in the healthy control group. However, HERV-W ENV detection in plasma did not seem to be an exclusive feature of the post-COVID-19 group, as 30.7 % of the compared cases (35/114) also showed detection of this toxic protein. In particular, the prevalence of HERV-W ENV protein for each of the compared subgroups was 25 % (3/12) in ME/CFS cases, 33.3 % (14/42) in the FM group, and 30 % (18/60) in the co-diagnosed group (Fig. 1), clearly showing detectable antigenemia in all patient groups, with higher prevalence in the post-COVID-19 condition. Overall, this indicates that the expression of HERV-W ENV is not specific to one of these conditions only but may affect a sub-group in each one. This likely reflects HERV-W activation triggered by infectious agents, identified or not, in susceptible individuals.

#### 3.3. SARS-CoV-2 serology according to immunoglobulin class

To explore the potential relationship between HERV-W ENV protein expression and the immune response to SARS-CoV-2, we measured circulating levels of IgG, IgM, IgA and IgE anti-SARS-CoV-2 immunoglobulins in post-COVID-19 condition cases and compared them to those of patients with ME/CFS, FM, co-diagnosed, and healthy controls (Fig. 2).

Patient study groups showed alterations across all immunoglobulin types. The differences observed were modest for IgG and IgA types (Fig. 2A and C) as compared to IgM and IgE differences (Fig. 2B and D), with significance for increased anti-spike IgG in the post-COVID-19 condition and IgA in the co-diagnosed group, suggesting a more sustained class-specific response in these patients.

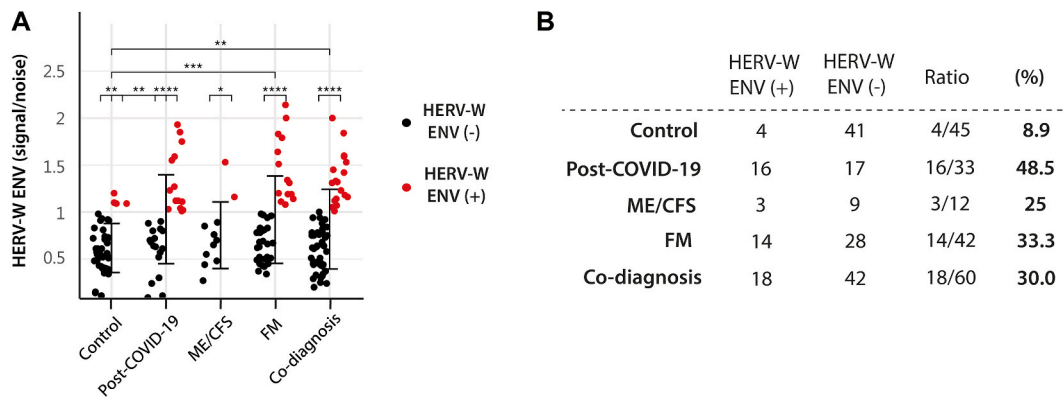
Interestingly, total IgM antigenemia was detected in all disease groups, with prominent values for anti-spike IgM levels in post-COVID-19 condition (Fig. 2B), which could relate to the potential SARS-CoV-2 reservoirs pointed by some authors (Proal et al., 2025).

With respect to total and anti-spike IgE antigenemia, both appeared significantly increased in the post-COVID-19 condition compared to healthy subjects, with significant differences across all other study groups for total IgE antigenemia. However, the small ME/CFS study group showed only a tendency ( $p \leq 0.1$ ) (Fig. 2D). This suggests a potential specific participation of IgE antigenemia in the post-COVID-19 condition, further supporting our previous findings (Giménez-Orenga et al., 2022).

**Table 1**  
Patient health status assessment with FIQ, MFI, SF-6 and COMPASS31 questionnaires (Burckhardt, Clark, and Bennett, 1991; McHorney, Ware and Raczek, 1993; Rivera and González, 2004; Sletten et al., 2012; Smets et al., 1995), by study group.

Questionnaire	Post-COVID-19 (n = 33)			ME/CFS (n = 12)			FM (n = 42)			Co-diagnosis (n = 60)		
	Mean	SD ± SE	Range	Mean	SD ± SE	Range	Mean	SD ± SE	Range	Mean	SD ± SE	Range
<b>FIQ</b>												
Total FIQ*	60.55	21.15 ± 3.8	(7.09 - 88.54)	67.24	12.31 ± 3.55	(48.85 - 87.51)	71.90	18.64 ± 2.88	(9.86 - 97.5)	76.41	16.05 ± 2.11	(14.61 - 97.5)
Function*	4.61	2.33 ± 0.42	(0 - 8.25)	6.71	2.07 ± 0.60	(1.65 - 8.58)	5.52	2.74 ± 0.42	(0 - 9.9)	6.79	1.98 ± 0.26	(1.32 - 10.56)
Overall*	6.14	2.51 ± 0.45	(0 - 8.58)	7.51	1.94 ± 0.56	(2.86 - 8.58)	6.98	2.26 ± 0.35	(0 - 8.58)	7.79	1.4 ± 0.18	(1.43 - 8.58)
Symptoms	5.67	3.17 ± 0.57	(1.43 - 10.01)	8.10	2.68 ± 0.77	(1.43 - 10.01)	5.99	3.02 ± 0.47	(1.43 - 10.01)	7.17	2.51 ± 0.33	(1.43 - 10.01)
<b>MFI</b>												
General Fatigue*	17.29	2.77 ± 0.5	(11 - 20)	18.17	2.62 ± 0.76	(12 - 20)	16.69	3.77 ± 0.58	(8 - 20)	18.52	2.67 ± 0.35	(8 - 20)
Physical Fatigue	17.48	2.9 ± 0.52	(8 - 20)	18.33	2.39 ± 0.69	(13 - 20)	16.26	4.42 ± 0.68	(5 - 20)	18.28	2.8 ± 0.37	(4 - 20)
Reduced Activity	15.39	3.67 ± 0.66	(8 - 20)	17.83	2.95 ± 0.85	(12 - 20)	15.79	4.56 ± 0.7	(4 - 20)	16.72	3.78 ± 0.5	(4 - 20)
Reduced Motivation	12.94	4.11 ± 0.74	(4 - 20)	13.17	3.07 ± 0.89	(8 - 18)	13.05	4.4 ± 0.68	(4 - 20)	14.67	4.22 ± 0.55	(5 - 20)
Mental Fatigue	14.77	4.94 ± 0.89	(4 - 20)	16.50	3.63 ± 1.05	(10 - 20)	15.38	4.71 ± 0.73	(4 - 20)	16.24	4.17 ± 0.55	(4 - 20)
<b>SF-36</b>												
Physical Functioning*	48.55	23.1 ± 4.15	(5 - 90)	31.25	16.94 ± 4.89	(0 - 50)	33.1	22.82 ± 3.52	(0 - 85)	24.48	11.95 ± 1.57	(0 - 50)
Role Physical	2.14	1.09 ± 0.2	(1 - 4.5)	1.41	0.63 ± 0.18	(1 - 3)	1.96	1.01 ± 0.16	(1 - 4.75)	1.47	0.6 ± 0.08	(1 - 3.25)
Bodily Pain*	44.03	27.79 ± 4.99	(10 - 100)	63.12	27.57 ± 7.96	(10 - 100)	30.6	18.84 ± 2.91	(10 - 80)	29.78	19.5 ± 2.56	(10 - 100)
General Health*	29.84	12.81 ± 2.3	(10 - 70)	25.83	13.29 ± 3.84	(0 - 50)	26.31	15.1 ± 2.33	(0 - 75)	18.88	14.11 ± 1.85	(0 - 55)
Vitality*	30.48	15.94 ± 2.86	(10 - 65)	25.42	13.73 ± 3.96	(10 - 50)	23.93	16.17 ± 2.5	(10 - 70)	18.28	11.64 ± 1.53	(10 - 50)
Social Functioning	36.37	23.21 ± 4.17	(0 - 90)	22.92	18.85 ± 5.44	(0 - 57.5)	30.83	24.97 ± 3.85	(0 - 90)	24.01	21.14 ± 2.78	(0 - 90)
Role Emotional*	3.59	1.11 ± 0.2	(1 - 5)	3.75	1.16 ± 0.33	(2 - 5)	2.87	1.28 ± 0.2	(1 - 5)	2.91	1.28 ± 0.17	(1 - 5)
Mental Health	51.61	16.28 ± 2.92	(8 - 84)	54.67	19.69 ± 5.69	(28 - 84)	42.29	16.44 ± 2.54	(8 - 76)	43.86	20.1 ± 2.64	(8 - 84)
<b>COMPASS-31</b>												
OI score*	17.81	8.25 ± 1.48	(0 - 32)	22.00	10.58 ± 3.06	(0 - 36)	24.86	9.25 ± 1.43	(4 - 40)	29.79	7.76 ± 1.02	(0 - 40)
Vasomotor	0.67	1.32 ± 0.24	(0 - 4.17)	0.56	1.03 ± 0.30	(0 - 2.5)	0.83	1.34 ± 0.21	(0 - 4.17)	0.98	1.44 ± 0.19	(0 - 4.17)
Secretomotor*	5.6	3.57 ± 0.64	(0 - 10.71)	5.90	4.68 ± 1.35	(0 - 12.9)	9.4	4.09 ± 0.63	(0 - 15)	9.95	3.14 ± 0.41	(0 - 12.9)
GI*	8.09	3.31 ± 0.59	(1.79 - 14.29)	9.38	3.91 ± 1.13	(0 - 13.39)	10.8	4.38 ± 0.68	(1.79 - 21.43)	12.04	3.57 ± 0.47	(1.79 - 17.86)
Bladder*	1.15	1.33 ± 0.24	(0 - 5.56)	2.58	2.85 ± 0.82	(0 - 10)	3.35	2.95 ± 0.45	(0 - 10)	4.17	2.71 ± 0.36	(0 - 10)
Pupillomotor*	2.14	1.05 ± 0.19	(0 - 3.67)	2.11	1.43 ± 0.41	(0 - 4)	3.02	0.84 ± 0.13	(0 - 4)	3.07	0.83 ± 0.11	(0.3 - 4)
Total COMPASS-31*	35.46	12.56 ± 2.26	(7.12 - 56.86)	42.33	17.87 ± 5.16	(0 - 69)	52.31	16.07 ± 2.48	(12 - 86)	59.98	12.68 ± 1.67	(18 - 88)

(\*) indicate statistical differences ( $p \leq 0.05$ ) between groups, as detailed on [Supplementary Table S1](#).



**Fig. 1.** HERV-W envelope (ENV) expression in post-pandemic subjects ( $n = 193$ ), healthy controls (HC) ( $n = 46$ ); post-COVID-19 ( $n = 33$ ); ME/CFS ( $n = 12$ ); FM ( $n = 42$ ); and co-diagnosed ( $n = 60$ ). **(A)** for HERV-W ENV protein antigenemia are labeled in red, defined as those values above the mean + 2SD of the background signals yielded by a panel of healthy control samples. \* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ , \*\*\*\* $p < 0.0001$  according to T-Test. **(B)** Summary table displaying number of cases expressing HERV-W-ENV (+) or with normal levels (-), ratios and percentages of HERV-W-ENV detection by study group.

As an overall, these results highlight that a subgroup of post-COVID-19 condition patients present a sustained immune response against SARS-CoV-2 antigens, with increased IgM and IgE levels.

To determine whether HERV-W ENV presence was related to anti-SARS-CoV-2 antigenemia, we stratified participants according to HERV-W ENV levels and then evaluated possible differences between anti-SARS-CoV-2 immunoglobulin levels. No such relationship could be identified for any of the immunoglobulin isotypes (Supplementary Fig. S2), concluding that SARS-CoV-2 humoral response is unrelated to HERV-W ENV levels.

### 3.4. Cytokine and NFL differences across patient groups

Given that cytokine levels can reflect immune disturbances potentially linked to the observed skewed anti-SARS-CoV-2 humoral response in some patients, we next assessed circulating levels of IL-6, IL-8, IL-10, IFN $\gamma$ , and TNF $\alpha$  in these same cohort. In addition, NFL plasma levels were measured to evaluate potential neuronal damage and peripheral neuropathy, which are frequently reported in these patient populations (Maalmi et al., 2023). The results revealed elevated IL-6 levels in all patient groups except those with post-COVID-19 condition as compared to healthy controls. IL-8 levels were significantly increased in FM and co-diagnosis groups relative to ME/CFS; while IL-10 levels were elevated in all groups except ME/CFS. Notably IFN $\gamma$  levels were reduced in the post-COVID-19 condition group. No significant differences were detected in circulating levels of NFL and TNF $\alpha$ .

Overall, these results indicate altered immune function, with a distinct reduction of IFN $\gamma$  levels in the post-COVID condition group. Cytokine profiles also revealed differences between diagnostic groups, although FM and co-diagnosed patients could not be distinguished based on these markers.

To determine whether cytokine or NFL concentration are influenced by HERV-W ENV expression levels, samples were first stratified by HERV-W ENV expression levels and then reanalyzed. However, no significant differences with HERV-W ENV expression could be detected for any of the measured variables (Supplementary Fig. S3).

### 3.5. Blood analytics across patient groups

To further search for potential disease-associated factors, we studied patient hemograms by study group in reference to normal population levels (Fig. 4). Normal range values are indicated by horizontal dotted lines. A limitation encountered was that among the biobanked post-COVID-19 condition samples, this information was available from only

14 out of the 33 participants (Supplementary Table S1). As shown on Fig. 4, patients in the post-COVID-19 condition group had significantly lower ferritin levels and significantly higher mean corpuscular hemoglobin concentration (MCHC) compared to all other patient groups (Supplementary Table S1).

Once again, stratification of samples by HERV-W ENV levels did not reveal any significant differences in hemogram parameters or platelet counts (Supplementary Fig. S4).

### 3.6. Potential diagnostic value of findings for the post-COVID-19 condition group

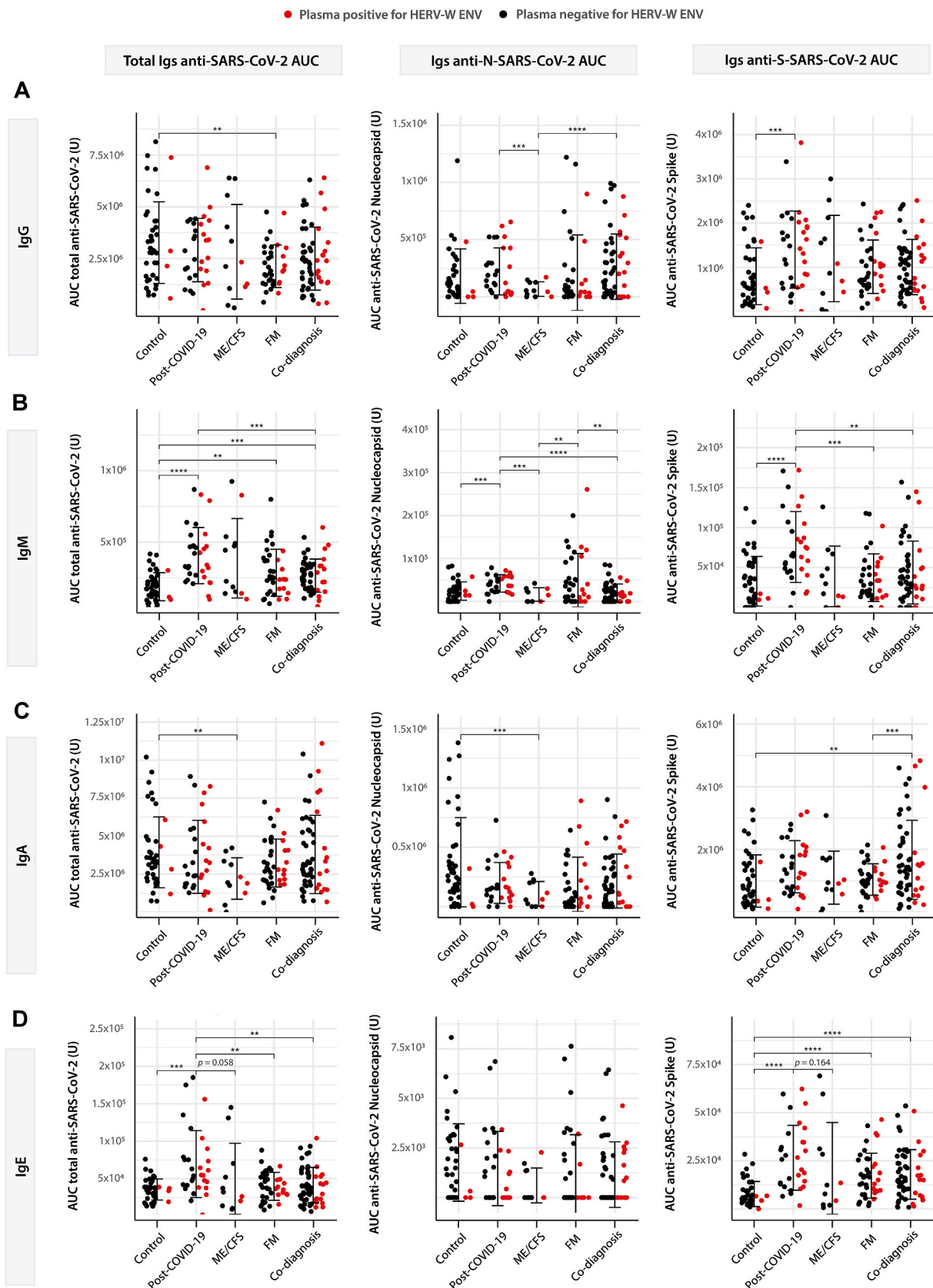
Since the post-COVID-19 condition group exhibited distinct differences, not only compared to healthy subjects, but also with respect to the other patient groups, we sought to evaluate the potential diagnostic value of these differences using Receiver Operating Characteristic (ROC) curve analysis.

To this end, we selected combinations of two, three or four variables most specifically associated with the post-COVID-19 condition, namely SARS-CoV-2 IgE antigenemia, IFN- $\gamma$ , Ferritin and MCHC, hereafter referred to as post-COVID-19 condition diagnostic variables. Individually, Ferritin, MCHC (Fig. 4), IFN $\gamma$  (Fig. 3) and anti-SARS-CoV-2 IgE antigenemia (Fig. 2) effectively distinguished post-COVID-19 condition cases from both, healthy individuals and patients with ME/CFS, FM and co-diagnoses.

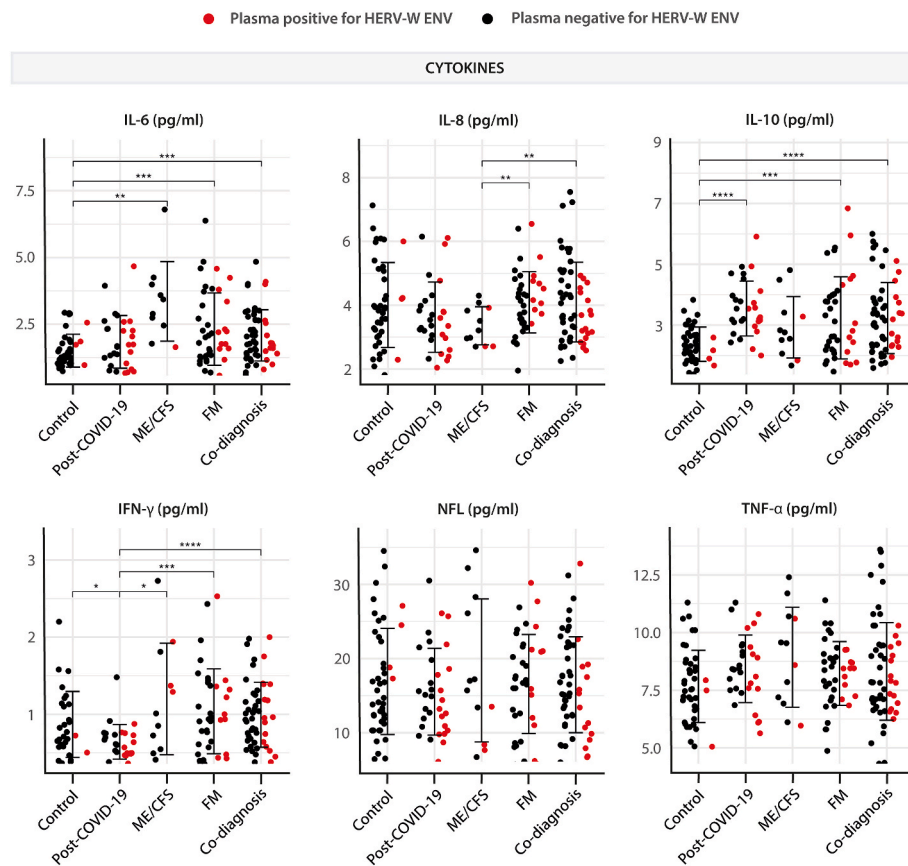
Fig. 5 presents the ROC curve built with just 2 variables yielding the highest AUC value. Notably Ferritin and MCHC alone already provided a robust diagnostic tool (Fig. 5A), achieving high sensitivity (92.3 %) and acceptable specificity (74.8 %). Surprisingly, these two red blood cells-related parameters outperformed IFN- $\gamma$  and IgE antigenemia in differentiating post-COVID-19 condition cases from other patient groups. Furthermore, when IgE and IFN $\gamma$  values were added to the model, sensitivity increased to 100 %, ensuring no true positive cases were missed. However, this came at the expense of a reduced specificity of 70.8 % (Fig. 5B) indicating a higher risk of false positives. Additional ROC curves exploring various combinations of these biomarker candidates are shown on Supplementary Fig. S5.

### 3.7. Correlation of HERV-W ENV expression with post-COVID-19 diagnostic variables (SARS-CoV-2 IgE antigenemia, IFN- $\gamma$ , Ferritin and MCHC)

Since the highest proportion of HERV-W ENV-positive individuals was observed in the post-COVID-19 condition group (48 %, Fig. 1), we



**Fig. 2.** Anti-SARS-CoV-2 multi-isotype serology of healthy controls (n = 46), in comparison to post-COVID-19 condition (n = 33), ME/CFS (n = 12), FM (n = 42), and co-diagnosed (n = 60) cases. Levels of IgG (A), IgM (B), IgA (C) and IgE (D) against the SARS-CoV-2 nucleocapsid, spike or the combination of both (total) are shown as the mean of the area under the curve (AUC) according to electropherograms. Cut-off for each isotype was calculated as the mean of pre-pandemic controls +3 standard deviations (SD). Samples positive for HERV-W ENV protein antigenemia are labeled in red. \*\*p < 0.01, \*\*\*p < 0.001, \*\*\*\*p < 0.0001 according to Mann-Whitney Test.



**Fig. 3.** Circulating IL-6, IL-8, IL-10, IFN $\gamma$ , NFL and TNF $\alpha$  levels (pg/ml) in healthy controls (n = 46), in comparison to post-COVID-19 condition (n = 33), ME/CFS (n = 12), FM (n = 42), and co-diagnosed (n = 60) cases. Samples positive for HERV-W ENV protein antigenemia are labeled in red. Group differences statistical significance is indicated with \*p < 0.05, \*\*p < 0.01, \*\*\*p < 0.001, \*\*\*\*p < 0.0001 according to Mann-Whitney Test.

next investigated potential correlations of HERV-W ENV expression levels and the four post-COVID-19 diagnostic variables. As shown in Fig. 6A, moderate to strong negative correlations ( $R > 0.5$ ,  $p \leq 0.05$ ) were identified between HERV-W ENV levels and both MHCH and IFN $\gamma$  in the post-COVID-19 condition group. No such correlations were identified in any of the other patient groups (Supplementary Table S2). Moreover, the diagnostic performance of the ROC curve based on the four post-COVID-19 diagnostic variables (SARS-CoV-2 IgE antigenemia, IFN- $\gamma$ , Ferritin and MCHC) showed a modest improvement when HERV-W ENV was included as an additional variable. The AUC increased from 88.4 to 89.3 %, accompanied by a slight reduction in the false positive rate from 29.2 % to 28.1 % (Fig. 6B).

### 3.8. Correlations of post-COVID-19 diagnostic variables and HERV-W ENV levels with patient symptoms

Finally, to identify potential connections between post COVID-19 condition diagnostic variables and HERV-W ENV values with patient symptoms, we assessed their statistical associations with subdomains from the four-health assessment and phenotyping instruments (FIQ, MFI, SF-36, and COMPASS-31). Fig. 7 displays a heatmap summarizing moderate to strong correlations (Spearman  $R > 0.3$ ,  $p \leq 0.05$ ,  $n = 14$ ) between the five diagnostic variables (SARS-CoV-2 IgE antigenemia, IFN- $\gamma$ , Ferritin, MCHC and HERV-W ENV) and each of the symptom subdomains. Strikingly, MHCH showed the strongest associations with symptom severity. Lower hemoglobin concentration values correlated with higher “Total FIQ” ( $R = -0.51$ ) and MFI “Reduced Activity” ( $R = -0.77$ ) scores, indicating poorer health status. Additionally, it also associated with lower SF-36, “Role Physical” domain ( $R = 0.63$ ) score, where lower scores represent poorer quality of life. IFN $\gamma$  also showed a

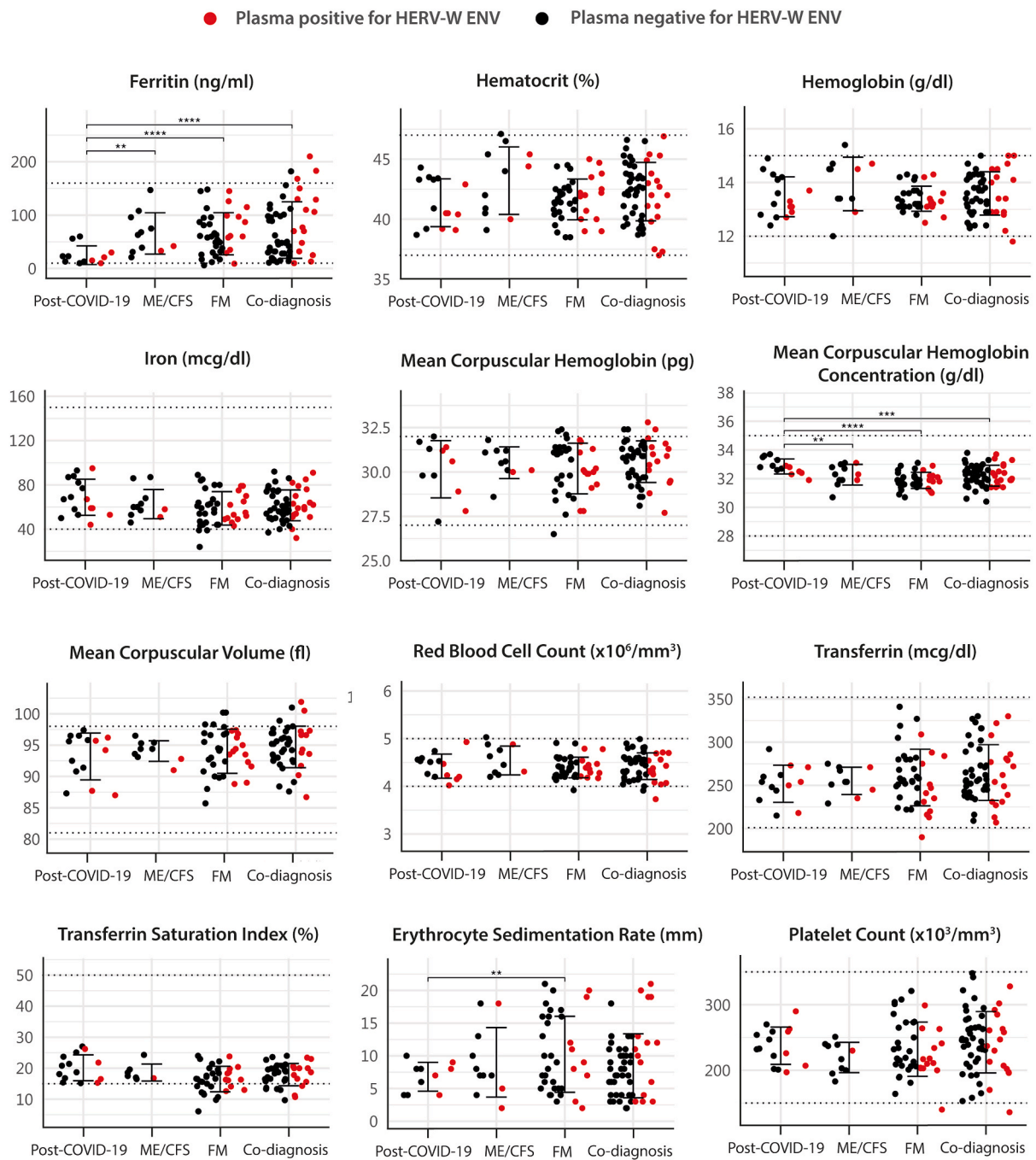
consistent, though moderate trend ( $R < 0.4$ ,  $p \leq 0.1$ ). Lower IFN $\gamma$  levels were associated with worse scores in FIQ subdomains (“Overall” and “Symptoms”), and MFI (“Physical Fatigue” and “Reduced Activity”) subdomains, and positively correlated with the SF-36 ‘Vitality’ domain, suggesting a link between reduced IFN- $\gamma$  and poorer health status.

In contrast, HERV-W ENV levels, SARS-CoV-2 IgE antigenemia and Ferritin showed fewer significant associations. Notably, ferritin levels strongly correlated with the COMPASS-31 ‘Secretomotor’ subdomain. ( $R = 0.62$ ), suggesting increased dysautonomia symptoms on the secretomotor system (sweat and salivary glands leading to hyperhidrosis or anhidrosis) in patients with elevated ferritin levels.

Interestingly, high HERV-W ENV levels negatively correlated with the “Pupillomotor” domain of the COMPASS-31 questionnaire suggesting vision problems and/or signs of neurodegeneration), although to a moderate extent. Also moderate were the correlations between SARS-CoV-2 IgE antigenemia and SF-36 “Bodily Pain” and “Mental Health” subdomains, suggesting worse health status with higher levels of this antibody. No significant correlations were observed between these five diagnostic variables and symptom subdomains in the ME/CFS, FM, or co-diagnosed groups (Supplementary Table S3).

## 4. Discussion

Motivated by the need to better understand the potential relationships between post-COVID-19 condition, ME/CFS and FM, and supported by preliminary evidence of increased HERV-W ENV expression in these conditions (Charvet et al., 2023; Giménez-Orenga et al., 2022; Ovejero et al., 2020; Rodrigues et al., 2019), this study aimed to measure and compare circulating levels of HERV-W ENV among female patients from a single geographic region (n = 147), alongside matched healthy



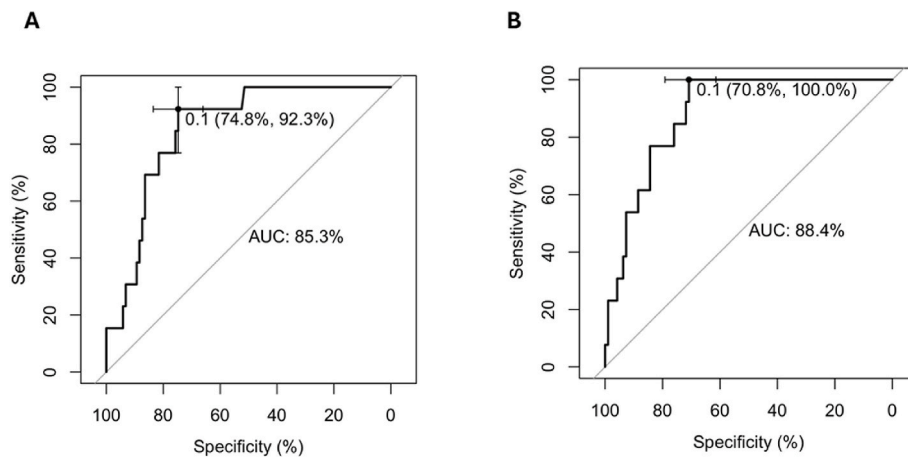
**Fig. 4.** Red blood cell analytics and platelet counts by patient study group. Post-COVID-19 condition (n = 14), ME/CFS (n = 12), FM (n = 42), and co-diagnosed (n = 60) cases. Variables are grouped in relation to red blood cells and platelets (upper) or belonging to white blood cells (lower). Units for each variable are indicated. Samples positive for HERV-W ENV protein antigenemia are labeled in red. Group differences statistical significance is indicated with \*\*p < 0.01, \*\*\*p < 0.001, \*\*\*\*p < 0.0001 according to Mann-Whitney Test.

female controls (n = 46). The objective was to identify potential differences in the expression of this toxic protein in relation to SARS-CoV-2 antigenemia, blood parameters, and patient symptoms. Male exclusion seemed justified in favor of minimizing sex-associated confounding factors (Athnaiel et al., 2023; Gamer et al., 2023; Kitselman et al., 2024).

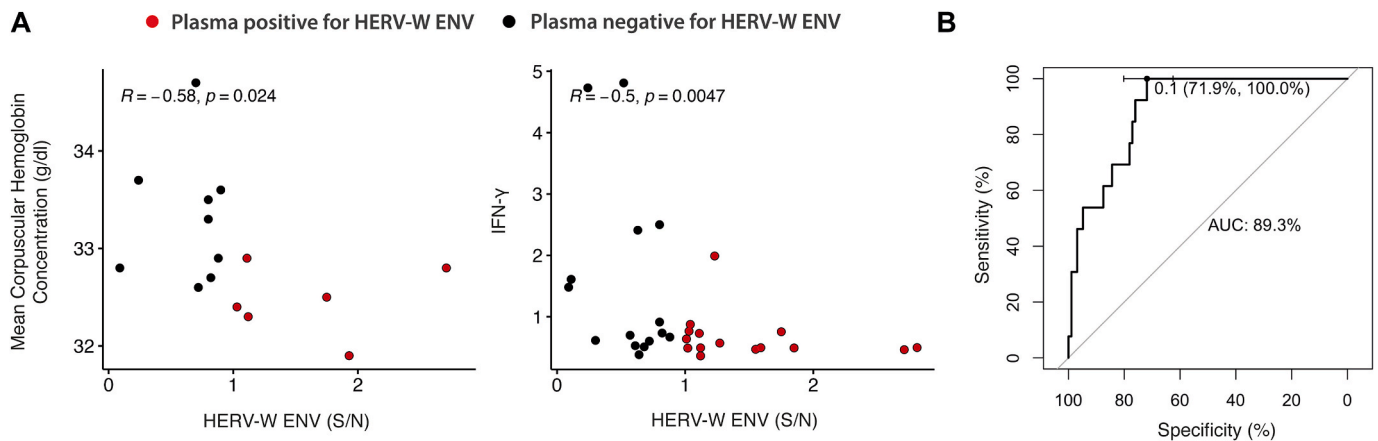
Our initial findings showed HERV-W activation in subsets of individuals for each compared group, with highest prevalence in the post-COVID-19 condition group (48.5 %). This was notably higher than in the ME/CFS (25 %), FM (33.3 %), and the co-diagnosis group (30 %), all of which exceeded the prevalence observed in healthy controls (8.9 %). These results suggest a widespread biological process possibly driven by

post-infectious or inflammatory cues, as proposed in the context of other neurological, psychiatric and autoimmune diseases (Tamouza et al., 2021; Rangel et al., 2024), and/or other yet unknown mechanisms perpetuating or fueling chronicity in a sub-group of patients.

Whether inflammation leads to HERV-W ENV expression, or vice versa, may be context dependent. *In vitro* controlled experiments replicating acute SARS-CoV-2, or the related Middle East respiratory syndrome coronavirus (MERS), or influenza A virus (IAV) infections have shown viral-specific induction of HERV expression profiles. These patterns were partially overlapping and largely independent of the host cell line (Marston et al., 2021), supporting the idea of a general viral-driven



**Fig. 5.** Receiving Operating Characteristic (ROC) curves for post-COVID-19 condition, using Ferritin levels and Mean Corpuscular Hemoglobin Concentration (MCHC) values (A) or adding anti-SARS-CoV-2 IgE antigenemia and circulating interferon gamma (INF $\gamma$ ) levels (B). Area Under the Curve (AUC) values, together with Specificity and Sensitivity readings of the model are displayed.



**Fig. 6.** Correlations of HERV-W ENV with post-COVID-19 MCHC and INF- $\gamma$  values ( $R > |0.5|$ ,  $p \leq 0.05$ , Spearman) (A), and ROC curve for post-COVID-19 condition with five variables (SARS-CoV-2 IgE antigenemia, INF- $\gamma$ , Ferritin, MCHC, and HERV-W ENV levels). Area Under the Curve (AUC) values, together with Specificity and Sensitivity readings of the model are displayed (B).

mechanism. They also suggest that blood-based detection of HERVs may serve as a viable “liquid biopsy” approach. The study also showed that the elements becoming activated were enriched in binding sites for transcription factors involved in immune responses. How exactly is the HERV epigenetic silencing machinery controlled by SARS-CoV-2, or other viral agents, in acute vs. chronic stages, remains to be defined. A comprehensive analysis of SARS-CoV-2 antigenemia, by immunoglobulin type (IgG, IgM, IgA and IgE) and antigen (nucleocapsid and spike) after stratification by HERV-W ENV levels, was conducted in search of potential links between SARS-CoV-2 antigenemia and HERV-W ENV activation. However, no significant intragroup differences were found for any immunoglobulin or the rest of variables measured with HERV-W ENV levels. Samples with elevated HERV-W ENV expression were marked with red dots across figures to facilitate visual tracking of these analysis.

Interestingly our findings reaffirmed previous observations of elevated IgE responses against SARS-CoV-2 (Giménez-Orenga et al., 2022) predominantly in the long COVID-19 group. IgEs are capable of activating mast cells and basophils, degranulating, and thus signaling inflammation (Collins and Jackson, 2013). They are strong effectors of humoral responses against parasitic infections (Hadadianpour et al., 2022), associated with allergies (Alexiou et al., 2022). Prolonged exposure to antigens, typical of parasite infection or sustained presence of allergens, can induce IgE class switching.

Although inclusion/exclusion criteria ruled out individuals presenting symptoms for ongoing acute infections, or those who had received a vaccine within the previous month, an interval chosen due to concerns about potential anaphylactic reactions to PEG or other (Khalid and Frischmeyer-Guerrero, 2023), the possibility of an unnoticed ongoing infection cannot be entirely discarded.

Despite uncommon, certain viral protein antigens, such as those from the RSV (respiratory syncytial virus) (Tesari Crnković et al., 2023), or varicella zoster (Smith-Norowitz et al., 2009) have been shown to elicit IgE responses. In the context of SARS-CoV-2, our previous findings (Giménez-Orenga et al., 2022), along with two additional studies support the occurrence of increased IgE antigenemia following SARS-CoV-2 infection (Plüme et al., 2022; Portilho et al., 2024). However, to the best of our knowledge, our studies are the only demonstrating perpetuation of these antibodies with long COVID-19. SARS-CoV-2 serology for other Ig types appeared less relevant in distinguishing patient groups, with the exception of total IgM, which appears to discern all patient groups from controls. As mentioned, vaccination cannot explain this difference, and the likelihood of ongoing recent SARS-CoV-2 infections in all patient groups, but not in the healthy control group, seems less plausible than the alternative explanation of viral reactivation or persistent viral protein presence in our chronic patient groups.

Transient hematological alterations with SARS-CoV-2 infection as with any other virus were expected, however, numerous studies have

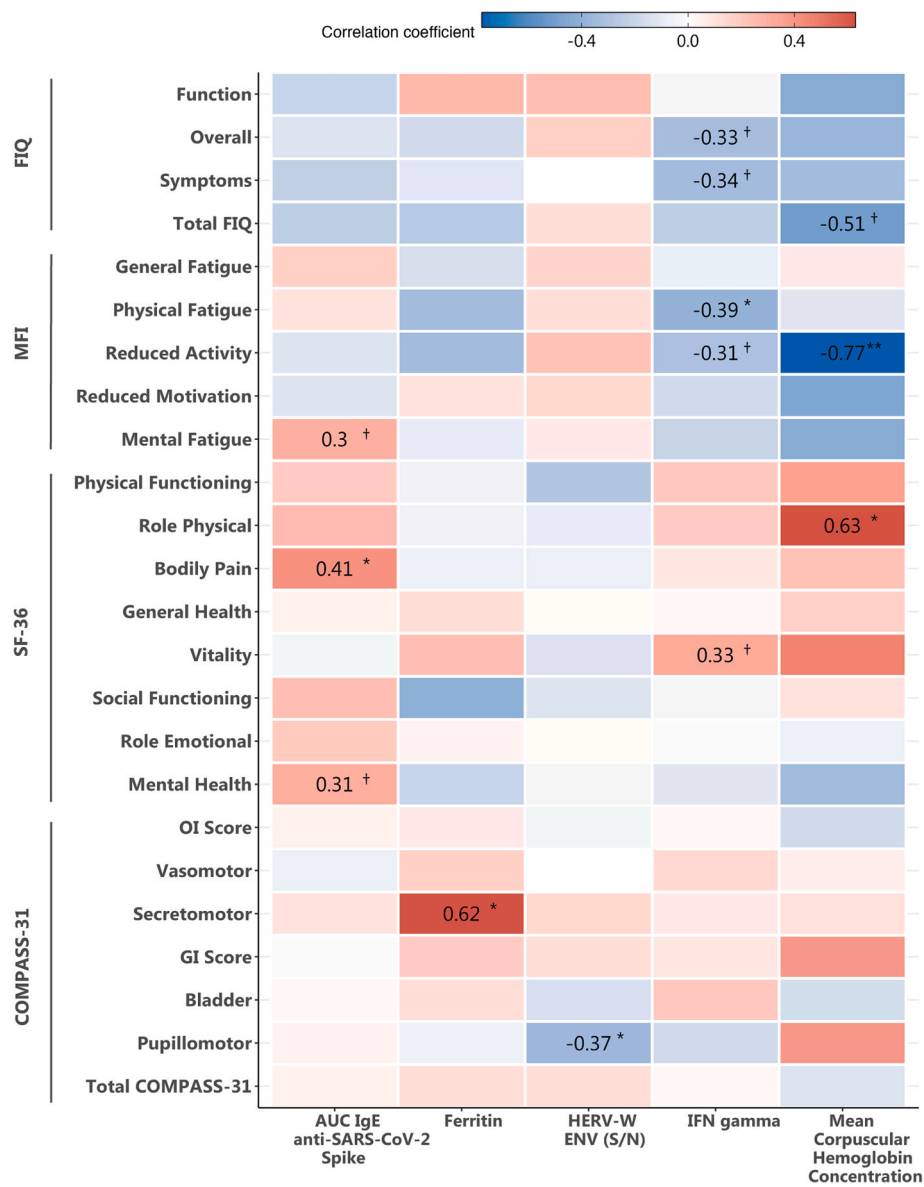


Fig. 7. Summary heatmap to represent the significance and intensity of correlations between SARS-CoV-2 IgE antigenemia, IFN- $\gamma$ , Ferritin, MCHC and HERV-W ENV levels and FIQ, MFI, SF-36 and COMPASS-31 questionnaire scores. Instrument subdomains are indicated. The color palette shows positive correlations in pink/red while blue displays negative correlations. R values of significant correlations (\*\*p  $\leq$  0.001; \*p  $\leq$  0.05; and †p  $\leq$  0.1) are displayed.

reported that such changes may persist in individuals with post-COVID-19 condition, sometimes with contradictory findings (Lechuga et al., 2023). In this study, the most notable result from cytokine profiling was the low levels of IFN $\gamma$  in the post-COVID-19 condition group with respect to all other groups (the three patient groups as well as the healthy control). This reduction, alongside elevated IgE levels, suggests potential differential diagnostic value across patient groups, an aspect pursued by this study. Our IFN $\gamma$  findings are in good agreement with those by (Williams et al., 2022), but not with those by (Queiroz et al., 2022), who did not detect changes in the levels of this cytokine.

Additional cytokine differences were observed in IL-10 and IL-6 levels. IL-10, an anti-inflammatory cytokine, was elevated in all patient groups, except ME/CFS. In contrast, ME/CFS patients exhibited increased levels of the proinflammatory IL-6, a pattern also seen in the FM and co-diagnosed group but not in post-COVID-19 condition. Our IL-10 findings in post-COVID-19 condition contradict the two previously cited studies (Queiroz et al., 2022; Williams et al., 2022) who report

decreased and unchanged levels, respectively. This disagreement also includes IL-6 and IL-8 levels, found significantly decreased by Williams et al. in long COVID-19 patients, which did not show significant differences in our group. The significance of these disagreements is unknown at present. However, both studies coincide with our results in reporting unchanged TNF $\alpha$  levels in the post-COVID condition group. According to unchanged NFL levels, none of the patient groups seem to hold major neurologic damage (Maalmi et al., 2023).

Given previous reports of altered red blood cell (RBC) characteristics, including smaller red blood cell membrane rigidity, smaller size and increased MCHC, as well as hyperferritinemia in long COVID (Lechuga et al., 2023), we included selected RBC parameters in our analysis. Our findings revealed significantly lower ferritin levels and increased MCHC values in the post-COVID-19 condition group, distinguishing it from the ME/CFS, FM, and co-diagnosed groups. The results contrast with those of (Alfadda et al., 2022) regarding MCHC, and with six other studies on ferritin levels, which themselves reported inconsistent findings (Lechuga

et al., 2023). Despite these discrepancies, our data strongly support the potential of ferritin and MCHC as distinguishing biomarkers. When combined with IgE anti-spike antigenemia and IFN- $\gamma$ , these variables demonstrated high diagnostic potential for post-COVID-19 condition. ROC curve analysis confirmed this, with certain combinations achieving 100 % sensitivity and up to 74.8 % specificity, suggesting a promising tool for stratifying post-COVID-19 patients in individualized medicine programs.

In a final effort to understand potential connections between the high prevalence of HERV-W ENV levels in the post-COVID-19 condition group and these four markers found to hold differential diagnostic value (IgE anti-spike antigenemia, IFN- $\gamma$ , ferritin, and MCHC), with the highly related ME/CFS and FM syndromes, we analyzed their correlations with HERV-W ENV expression. Interestingly, moderate-to-strong negative correlations were observed for IFN $\gamma$  ( $R = -0.5$ ,  $p = 0.0047$ ) and MCHC ( $R = -0.58$ ,  $p = 0.024$ ). Furthermore, incorporating HERV-W ENV into the diagnostic model modestly improved the AUC and increased specificity by approximately 1 %, reinforcing its potential as a complementary biomarker.

A limitation of this study was the restricted availability of complete questionnaire data and red blood cell analytics for the post-COVID-19 condition group ( $n = 14$  out of 33 cases). Despite this reduced statistical power for two of the five diagnostic variables, the analysis still identified a robust combination of markers capable of distinguishing post-COVID-19 condition from ME/CFS and FM, with an AUC of 89.3 %. This finding is highly relevant for improving diagnostic accuracy, reducing misdiagnosis, and advancing the development of individualized treatment strategies for post-COVID-19 patients.

Our detailed patient phenotyping, using four validated questionnaires to assess different aspects of health status, revealed a range of positive and negative correlations in different subdomains. We focused our analysis on moderate-to-strong correlations to identify the most meaningful associations. Among all correlations stand out those of ferritin levels with secretomotor dysfunction (e.g. sweat and salivary glands leading to hyperhidrosis or anhidrosis), a hallmark of dysautonomia (Larsen et al., 2022). MCHC levels also stood out with lower hemoglobin concentration values associating with poorer health status, particularly increased pain and reduced physical activity. While hemoglobin itself is not directly linked to pain, its reduction can lead to hypoxia, fatigue, and inactivity, which in turn may contribute to chronic pain (Rodríguez-Sánchez-Laulhé et al., 2024). Similarly, elevated IgE anti-spike antigenemia was associated with increased bodily pain, further supporting its relevance as a biomarker. The key remaining question for all this work is how the identified biomarkers reflect patient status as the ultimate goal is to uncover therapeutic targets that correspond to an upstream driver of the disease. This may then allow healing toward the pre-infectious health status of the patient.

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## CRedit authorship contribution statement

**Karen Giménez-Orenga:** Writing – review & editing, Writing – original draft, Investigation, Funding acquisition, Formal analysis, Data curation. **Justine Pierquin:** Writing – review & editing, Investigation. **Joanna Brunel:** Writing – review & editing, Investigation. **Benjamin Charvet:** Writing – review & editing, Investigation. **Eva Martín-Martínez:** Writing – review & editing, Resources, Investigation. **Margot Lemaire:** Writing – review & editing, Investigation. **Steven Fried:** Writing – review & editing, Investigation. **Alexandre Lucas:** Writing – review & editing, Investigation. **Hervé Perron:** Writing – review & editing, Supervision, Resources, Methodology, Conceptualization. **Elisa**

**Oltra:** Writing – review & editing, Writing – original draft, Supervision, Resources, Methodology, Funding acquisition, Formal analysis, Conceptualization.

## Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Herve Perron reports financial support was provided by GeNeuro Innovation. Justine Pierquin reports financial support was provided by GeNeuro Innovation. Joanna Brunel reports financial support was provided by GeNeuro Innovation. Benjamin Charvet reports financial support was provided by GeNeuro Innovation. Elisa Oltra reports financial support was provided by Generalitat of Valencia Ministry of Education Culture Universities and Employment to cover reagents and external services only. Karen Gimenez-Orenga reports financial support was provided by Generalitat of Valencia Ministry of Education Culture Universities and Employment in the form of a PhD fellowship. All other authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper. Neither Generalitat of Valencia Ministry of Education Culture Universities and Employment, nor the Spanish public National Health Service or GeNeuro Innovation, participated in the design, investigation, data analysis and interpretation, or writing of this article.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.bbih.2025.101058>.

## Data availability

Data is provided as Supplementary Files as supplementary information information

## References

- Alexiou, A., Höfer, V., Döller-Bierke, S., Grünhagen, J., Zuberbier, T., Worm, M., 2022. Elicitors and phenotypes of adult patients with proven IgE-mediated food allergy and non-immune-mediated food hypersensitivity to food additives. *Clin. Exp. Allergy* 52 (11), 1302–1310. <https://doi.org/10.1111/cea.14203>.
- Alfadda, Assim A., Rafiullah, Mohamed, Alkhowaiter, Mohammad, Alotaibi, Naif, Alzahrani, Musa, Binkhamis, Khalifa, Siddiqui, Khalid, Youssef, Amira, Altalhi, Haifa, Almaghlouth, Ibrahim, Alarifi, Mohammed, Albanyan, Saleh, Alosaimi, Mohammed F., Isnani, Arthur, Nawaz, Shaik Sarfaraz, Alayed, Khalid, 2022. Clinical and biochemical characteristics of people experiencing post-coronavirus disease 2019-Related symptoms: a prospective Follow-up investigation. *Front. Med.* 9. <https://doi.org/10.3389/fmed.2022.1067082>.
- Athnaiel, Onella, Cantillo, Santiago, Paredes, Stephania, Knezevic, Nebojsa Nick, 2023. The role of sex hormones in pain-related conditions. *Int. J. Mol. Sci.* 24 (3).
- Balestrieri, Emanuela, Minutolo, Antonella, Petrone, Vita, Fanelli, Marialaura, Iannetta, Marco, Malagnino, Vincenzo, Zordan, Marta, Vitale, Pietro, Charvet, Benjamin, Horvat, Branka, Bernardini, Sergio, Garaci, Enrico, di Francesco, Paolo, Vallebona, Paola Sinibaldi, Sarmati, Loredana, Grelli, Sandro, Andreoni, Massimo, Perron, Hervé, Matteucci, Claudia, 2021. Evidence of the pathogenic HERV-W envelope expression in T lymphocytes in association with the respiratory outcome of COVID-19 patients. *EBioMedicine* 66. <https://doi.org/10.1016/j.ebiom.2021.103341>.
- Bjornevik, Kjetil, Cortese, Marianna, Healy, Brian C., Kuhle, Jens, Mina, Michael J., Leng, Yumei, Elledge, Stephen J., Niebuhr, David W., Scher, Ann L., Munger, Kassandra L., Ascherio, Alberto, 2022. MULTIPLE SCLEROSIS Longitudinal Analysis Reveals High Prevalence of Epstein-Barr Virus Associated with Multiple Sclerosis, 375.

- Burckhardt, C.S., Clark, S.R., Bennett, R.M., 1991. The Fibromyalgia Impact Questionnaire: development and validation. *J. Rheumatol.* 18 (5), 728–733. PMID: 1865419.
- Carruthers, B.M., Jain, A.K., De Meirleir, K.L., Peterson, D.L., Klimas, N.G., Lerner, A.M., Bested, A.C., Flor-Henry, P., Joshi, P., Powles, A.C.P., Sherkey, J.A., van de Sande, M.L., 2003. Myalgic encephalomyelitis/chronic fatigue syndrome: clinical working case definition, diagnostic and treatment protocols. *J. Chronic Fatigue Syndrome* 11 (1), 7–115. [https://doi.org/10.1300/J092v11n01\\_02](https://doi.org/10.1300/J092v11n01_02).
- Carruthers, B.M., Van de Sande, M.L., De Meirleir, K.L., Klimas, N.G., Broderick, G., Mitchell, T., Staines, D., Powles, A.C.P., Speight, N., Vallings, R., Bateman, L., Baumgarten-Austrheim, B., Bell, D.S., Carlo-Stella, N., Chia, J., Darragh, A., Jo, D., Lewis, D., Light, A.R., Marshall-Gradisbik, S., Mena, I., Mikovits, J.A., Miwa, K., Murovska, M., Pall, M.L., Stevens, S., 2011. Myalgic encephalomyelitis: international consensus criteria. *J. Intern. Med.* 270 (4), 327–338.
- Charvet, Benjamin, Brunel, Joanna, Pierquin, Justine, Iampietro, Mathieu, Decimo, Didier, Queruel, Nelly, Lucas, Alexandre, Mar Encabo-Berzosa, María del, Arenaz, Izaskun, Perez Marmolejo, Tania, Gonzalez, Arturo Ivan, Maldonado, Armando Castorena, Mathieu, Cyrille, Küry, Patrick, Flores-Rivera, Jose, Torres-Ruiz, Fernanda, Avila-Rios, Santiago, Montes de Oca, Gonzalo Salgado, Schoorlemmer, Jon, Perron, Hervé, Horvat, Branka, 2023. SARS-CoV-2 awakens Ancient retroviral genes and the expression of proinflammatory HERV-W envelope protein in COVID-19 patients. *iScience* 26 (5). <https://doi.org/10.1016/j.isci.2023.106604>.
- Chen, Chen, Haupt, Spencer R., Zimmermann, Lauren, Shi, Xu, Fritsche, Lars G., Mukherjee, Bhramar, 2022. Global prevalence of post-coronavirus disease 2019 (COVID-19) condition or long COVID: a meta-analysis and systematic review. *JID (J. Infect. Dis.)* 226 (9), 1593–1607. <https://doi.org/10.1093/infdis/jiac136>.
- Chen, Xun, Pacis, Alain, Aracena, Katherine A., Gona, Saideep, Kwan, Tony, Groza, Cristian, Lin, Yen Lung, Sindeaux, Renata, Yotova, Vania, Pramatarova, Albena, Simon, Marie Michelle, Pastinen, Tomi, Barreiro, Luis B., Guillaume, Bourque, 2023. Transposable elements are associated with the variable response to influenza infection. *Cell Genomics* 3 (5). <https://doi.org/10.1016/j.xgen.2023.100292>.
- Clauw, D.J., 2014. Fibromyalgia: a clinical review. *JAMA* 311 (15), 1547–1555. <https://doi.org/10.1001/jama.2014.3266>.
- Collins, Andrew M., Jackson, Katherine J.L., 2013. A temporal model of human IgE and IgG antibody function. *Front. Immunol.* 4 (AUG). <https://doi.org/10.3389/fimmu.2013.00235>.
- Dehlia, Ankush, Guthridge, Mark A., 2024. The persistence of Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS) after SARS-CoV-2 infection: a systematic review and meta-analysis. *J. Infect.* 89 (6).
- Fors, Egil A., Wensaas, Knut Arne, Helvik, Anne Sofie, 2024. Prevalence and characteristics of fibromyalgia according to three fibromyalgia diagnostic criteria: a secondary analysis study. *Scandinavian J. Pain* 24 (1). <https://doi.org/10.1515/sjpain-2023-0143>.
- Gamer, Jackson, Van Booven, Derek J., Zarnowski, Oskar, Arango, Sebastian, Elias, Mark, Kurian, Asha, Joseph, Andrew, Perez, Melanie, Collado, Fanny, Klimas, Nancy, Oltra, Elisa, Nathanson, Lubov, 2023. Sex-dependent transcriptional changes in response to stress in patients with myalgic encephalomyelitis/chronic fatigue syndrome: a pilot project. *Int. J. Mol. Sci.* 24 (12). <https://doi.org/10.3390/ijms241210255>.
- Giménez-Orenga, K., Oltra, E., 2021. Human endogenous retrovirus as therapeutic targets in neurologic disease. *Pharmaceuticals* 14 (6), 495. <https://doi.org/10.3390/ph14060495>.
- Giménez-Orenga, Karen, Justine, Pierquin, Joanna, Brunel, Benjamin, Charvet, Eva, Martín-Martínez, Hervé, Perron, Elisa, Oltra, 2022. HERV-W ENV antigenemia and correlation of increased Anti-SARS-CoV-2 immunoglobulin levels with Post-COVID-19 symptoms. *Front. Immunol.* 13. <https://doi.org/10.3389/fimmu.2022.1020064>.
- Giménez-Orenga, K., Martín-Martínez, E., Nathanson, L., Oltra, E., 2025. HERV activation segregates ME/CFS from fibromyalgia while defining a novel nosologic entity. *eLife* 14, RP104441. <https://doi.org/10.7554/eLife.104441>.
- Hadadianpour, Azadeh, Jacob, Daniel, Zhang, Jian, Spiller, Benjamin W., Makaraviciute, Asta, DeWitt, Åsa M., Walden, Heather S., Hamilton, Robert G., Stokes Peebles, R., Nutman, Thomas B., Smith, Scott A., 2022. Human IgE MABs identify major antigens of parasitic worm infection. *J. Allergy Clin. Immunol.* 150 (6), 1525–1533. <https://doi.org/10.1016/j.jaci.2022.05.022>.
- Khalid, Muhammad Bilal, Frischmeyer-Guerrero, Pamela A., 2023. The conundrum of COVID-19 MRNA vaccine-induced anaphylaxis. *J. Allergy Clin. Immunol.: Globalizations* 2 (1), 1–13.
- Khoja, Omar, Mulvey, Matthew, Astill, Sarah, Tan, Ai Lyn, Sivan, Manoj, 2024. New-onset chronic musculoskeletal pain following COVID-19 infection fulfils the fibromyalgia clinical syndrome criteria: a preliminary study. *Biomedicine* 12 (9), 1940. <https://doi.org/10.3390/biomedicine12091940>.
- Kitselman, A.K., Bédard-Matteau, J., Rousseau, S., Tabrizchi, R., Daneshmand, N., 2024. Sex differences in vascular endothelial function related to acute and long COVID-19. *Vasc. Pharmacol.* 154, 107250. <https://doi.org/10.1016/j.vph.2023.107250>.
- Larsen, N.W., Stiles, L.E., Shaik, R., Schneider, L., Muppidi, S., Tsui, C.T., Geng, L.N., Bonilla, H., Miglis, M.G., 2022. Characterization of autonomic symptom burden in long COVID: a global survey of 2,314 adults. *Front. Neurol.* 13, 1012668. <https://doi.org/10.3389/fneur.2022.1012668>.
- Lechuga, Guilherme C., Morel, Carlos M., De-Simone, Salvatore Giovanni, 2023. Hematological alterations associated with long COVID-19. *Front. Physiol.* 14.
- Lim, Eun Jin, Ahn, Yu Chan, Jang, Eun Su, Lee, Si Woo, Lee, Su Hwa, Son, Chang Gue, 2020. Systematic review and meta-analysis of the prevalence of chronic fatigue syndrome/myalgic encephalomyelitis (CFS/ME). *J. Transl. Med.* 18 (1).
- Maalmi, Haifa, Strom, Alexander, Petrer, Agnese, Hauck, Stefanie M., Strassburger, Klaus, Kuss, Oliver, Zaharia, Oana-Patricia, Bönhof, Gidon J., Rathmann, Wolfgang, Trenkamp, Sandra, Burkart, Volker, Szendroedi, Julia, Ziegler, Dan, Roden, Michael, Herder, Christian, 2023. Serum neurofilament light chain: a novel biomarker for early diabetic sensorimotor polyneuropathy. *Diabetologia* 66, 579–589. <https://doi.org/10.1007/s00125-022-05846-8/Published>.
- Macchietto, Marissa G., Langlois, Ryan A., Shen, Steven S., 2020. Virus-induced transposable element expression up-Regulation in human and mouse host cells. *Life Sci. Alliance* 3 (2). <https://doi.org/10.26508/lsa.201900536>.
- Marston, Jez L., Greenig, Matthew, Singh, Manvendra, Bendall, Matthew L., Duarte, Rodrigo R.R., Feschotte, Cédric, Iniguez, Luis P., Nixon, Douglas F., 2021. SARS-CoV-2 infection mediates differential expression of human endogenous retroviruses and long interspersed nuclear elements. *JCI Insight* 6 (24). <https://doi.org/10.1172/jci.insight.147170>.
- McHorney, C.A., Ware Jr, J.E., Raczek, A.E., 1993. The MOS 36-Item Short-Form Health Survey (SF-36): II. Psychometric and clinical tests of validity in measuring physical and mental health constructs. *Med. care* 31 (3), 247–263. <https://doi.org/10.1097/00005650-199303000-00006>.
- Natelson, Benjamin H., 2019. Myalgic encephalomyelitis/chronic fatigue syndrome and fibromyalgia: definitions, similarities, and differences. *Clin. Ther.* 41 (4), 612–618. <https://doi.org/10.1016/j.clinthera.2018.12.016>.
- Ovejero, Tamara, Sadones, Océane, Sánchez-Fito, Teresa, Almenar-Pérez, Eloy, Espejo, José Andrés, Martín-Martínez, Eva, Nathanson, Lubov, Oltra, Elisa, 2020. Activation of transposable elements in immune cells of fibromyalgia patients. *Int. J. Mol. Sci.* 21 (4). <https://doi.org/10.3390/ijms21041366>.
- Petrone, Vita, Fanelli, Marialaura, Giudice, Martina, Toschi, Nicola, Conti, Allegra, Maracchioni, Christian, Iannetta, Marco, Resta, Claudia, Cipriani, Chiara, Miele, Martino Tony, Amati, Francesca, Andreoni, Massimo, Sarmati, Loredana, Rogliani, Paola, Novelli, Giuseppe, Garaci, Enrico, Rasi, Guido, Sinibaldi-Vallebona, Paola, Minutolo, Antonella, Matteucci, Claudia, Balestrieri, Emanuela, Grelli, Sandro, 2023. Expression profile of HERVs and inflammatory mediators detected in nasal mucosa as a predictive biomarker of COVID-19 severity. *Front. Microbiol.* 14. <https://doi.org/10.3389/fmicb.2023.1155624>.
- Plüme, Janis, Galvanovskis, Artis, Smite, Sindija, Romanchikova, Nadezhda, Zaykin, Pawel, Liné, Aija, 2022. Early and strong antibody responses to SARS-CoV-2 predict disease severity in COVID-19 patients. *J. Transl. Med.* 20 (1). <https://doi.org/10.1186/s12967-022-03382-y>.
- Portillo, Amanda Izeli, Silva, Valéria Oliveira, Costa, Herman Hermes Monteiro Da, Yamashiro, Rosemeire, Oliveira, Isabela Penteriche de, Campos, Ivana Barros de, Prudencio, Carlos Roberto, Matsuda, Elaine Monteiro, Brígido, Luís Fernando de Macedo, Gaspari, Elizabeth De, 2024. An unexpected IgE anti-receptor binding domain response following natural infection and different types of SARS-CoV-2 vaccines. *Sci. Rep.* 14 (1). <https://doi.org/10.1038/s41598-024-71047-5>.
- Proal, A.D., Aleman, S., Bomsel, M., Brodin, P., Buggert, M., Cherry, S., Chertov, D.S., Davies, H.E., Dupont, C.L., Deeks, S.G., Ely, E.W., Fasano, A., Freire, M., Geng, L.N., Griffin, D.E., Henrich, T.J., Hewitt, S.M., Iwasaki, A., Krumholz, H.M., Locci, M., et al., 2025. Targeting the SARS-CoV-2 reservoir in long COVID. *Lancet Infect. Dis.* 25 (5), e294–e306. [https://doi.org/10.1016/S1473-3099\(24\)00769-2](https://doi.org/10.1016/S1473-3099(24)00769-2).
- Queiroz, Maria Alice Freitas, Neves, Pablo Fabiano Moura das, Lima, Sandra Souza, Lopes, Jefferson da Costa, Torres, Maria Karoliny da Silva, Izaura Maria Vieira Cayres Vallinoto, Bichara, Carlos David Araújo, Ferreira dos Santos, Erika, Mioni Thieli Figueiredo Magalhães de Brito, Andréa Luciana Soares da Silva, Mauro de Meira Leite, Flávia Póvoa da Costa, Maria de Nazaré do Socorro de Almeida Viana, Rodrigues, Fabíola Brasil Barbosa, Sarges, Kevin Matheus Lima de, Cantanhede, Marcos Henrique Damasceno, Silva, Rosilene da, Bichara, Clea Nazare Carneiro, Berg, Ana Virgínia Soares van den, Verissimo, Adriana de Oliveira Lameira, Carvalho, Mayara da Silva, Henriques, Daniele Freitas, Pinheiro dos Santos, Carla, Nunes, Juliana Abreu Lima, Costa, Iran Barros, Viana, Giselle Maria Rachid, Carneiro, Francisca Regina Oliveira, Palacios, Vera Regina da Cunha Menezes, Quaresma, Juarez Antonio Simões, Brasil-Costa, Igor, Melo dos Santos, Eduardo José, Falcão, Luiz Fábio Magno, Vallinoto, Antonio Carlos Rosário, 2022. Cytokine profiles associated with acute COVID-19 and long COVID-19 syndrome. *Front. Cell. Infect. Microbiol.* 12. <https://doi.org/10.3389/fcimb.2022.922422>.
- Rangel, Sara Coelho, Silva, Michelly Damasceno da, Filho, Décio Gilberto Natrielli, Nascimento Santos, Samuel, Bussador do Amaral, Jonatas, Russo Victor, Jefferson, Silva, Kevin Cezar Nascimento, Izabela, Dorota Tuleta, França, Carolina Nunes, Shio, Marina Tiemi, Neves, Lucas Melo, Bachi, André Luis Lacerda, Nali, Luiz Henrique da Silva, 2024. HERV-W upregulation expression in bipolar disorder and schizophrenia: unraveling potential links to systemic immune/inflammation status. *Retrovirology* 21 (1). <https://doi.org/10.1186/s12977-024-00640-3>.
- Rivera, J., González, T., 2004. The fibromyalgia impact questionnaire: a validated Spanish version to assess the health status in women with fibromyalgia. *Clin. Exp. Rheumatol.* 22 (5), 554–560.
- Rodrigues, Lucas S., Da Silva Nali, Luiz H., Leal, Cibele O.D., Sabino, Ester C., Lacerda, Eliana M., Kingdon, Caroline C., Nacul, Luis, Romano, Camila M., 2019. HERV-K and HERV-W transcriptional activity in myalgic encephalomyelitis/chronic fatigue syndrome. *Autoimmunity Highlights* 10 (1). <https://doi.org/10.1186/s13317-019-0122-8>.
- Rodríguez-Sánchez-Laulhé, Pablo, Heredia-Rizo, Alberto Marcos, Salas-González, Jesús, Piña-Pozo, Fernando, Fernández-Seguín, Lourdes María, García-Muñoz, Cristina, 2024. Exploring factors associated with changes in pain and function following MHealth-Based exercise therapy for chronic musculoskeletal pain: a systematic review with meta-analysis and meta-regression. *Appl. Sci.* 14 (15).

- Sletten, D.M., Suarez, G.A., Low, P.A., Mandrekar, J., Singer, W., 2012. COMPASS 31: a refined and abbreviated Composite Autonomic Symptom Score. *Mayo Clin. Proc.* 87 (12), 1196–1201. <https://doi.org/10.1016/j.mayocp.2012.10.013>.
- Smets, E.M., Garssen, B., Bonke, B., De Haes, J.C., 1995. The Multidimensional Fatigue Inventory (MFI): psychometric qualities of an instrument to assess fatigue. *J. Psychosom. Res.* 39 (3), 315–325. [https://doi.org/10.1016/0022-3999\(94\)00125-O](https://doi.org/10.1016/0022-3999(94)00125-O).
- Smith-Norowitz, T.A., Josekutty, J., Silverberg, J.L., Lev-Tov, H., Norowitz, Y.M., Kohlhoff, S., Nowakowski, M., Durkin, H.G., Bluth, M.H., 2009. Long term persistence of IgE anti-varicella zoster virus in pediatric and adult serum post chicken pox infection and after vaccination with varicella virus vaccine. *Int. J. Biomed. Sci.* 5 (4), 353–358. PMID: PMC3614806.
- Soriano, Joan B., Murthy, Srinivas, Marshall, John C., Relan, Pryanka, Diaz, Janet V., 2022. A clinical case definition of Post-COVID-19 condition by a Delphi consensus. *Lancet Infect. Dis.* 22 (4), e102–e107.
- Sukocheva, Olga A., Maksoud, Rebekah, Beeraka, Narasimha M., Madhunapantula, Sabba Rao V., Sinelnikov, Mikhail, Nikolenko, Vladimir N., Neganova, Margarita E., Klochkov, Sergey G., Kamal, Mohammad Amjad, Staines, Donald R., Marshall-Gradsnik, Sonya, 2022. Analysis of post COVID-19 condition and its overlap with myalgic encephalomyelitis/chronic fatigue syndrome. *J. Adv. Res.* 40, 179–196.
- Tamouza, Ryad, Meyer, Urs, Foiselle, Marianne, Richard, Jean Romain, Lu, Ching lieng, Boukouaci, Wahid, Le Corvoisier, Philippe, Barrau, Caroline, Lucas, Alexandre, Perron, Hervé, Leboyer, Marion, 2021. Identification of inflammatory subgroups of schizophrenia and bipolar disorder patients with HERV-W ENV antigenemia by unsupervised cluster analysis. *Transl. Psychiatry* 11 (1). <https://doi.org/10.1038/s41398-021-01499-0>.
- Tesari Crnković, H., Bendelja, K., Drkulec, V., Gjergja Juraški, R., Turkalj, M., 2023. Respiratory syncytial virus-specific antibodies and atopic diseases in children: a 10-year follow-up. *Pathogens (Basel, Switzerland)* 12 (4), 546. <https://doi.org/10.3390/pathogens12040546>.
- Unger, Elizabeth R., Lin, Jin Mann S., Wisk, Lauren E., Yu, Huihui, L'Hommedieu, Michelle, Lavretsky, Helen, Montoy, Juan Carlos C., Gottlieb, Michael A., Rising, Kristin L., Gentile, Nicole L., Santangelo, Michelle, Venkatesh, Arjun K., Rodriguez, Robert M., Hill, Mandy J., Geyer, Rachel E., Kean, Efrat R., Saydah, Sharon, McDonald, Samuel A., Huebinger, Ryan, Idris, Ahamed H., Dorney, Jocelyn, Hota, Bala, Spatz, Erica S., Stephens, Kari A., Weinstein, Robert A., Elmore, Joann G., 2024. Myalgic encephalomyelitis/chronic fatigue syndrome after SARS-CoV-2 infection. *JAMA Netw. Open* 7 (7), e2423555. <https://doi.org/10.1001/jamanetworkopen.2024.23555>.
- Venkatesan, Priya, 2021. NICE guideline on long COVID. *Lancet Respir. Med.* 9 (2), 129. [https://doi.org/10.1016/S2213-2600\(21\)00031-X](https://doi.org/10.1016/S2213-2600(21)00031-X).
- Williams, E.S., Martins, T.B., Shah, K.S., Hill, H.R., Coiras, M., Spivak, A.M., Planelles, V., 2022. Cytokine deficiencies in patients with Long-COVID. *J. Clin. Immunol.* 13 (6), 672.
- Wolfe, F., Clauw, D.J., Fitzcharles, M.A., Goldenberg, D.L., Katz, R.S., Mease, P., Russell, A.S., Russell, I.J., Winfield, J.B., 2010. The American College of Rheumatology preliminary diagnostic criteria for fibromyalgia and measurement of symptom severity. *Arthritis Care Res.* 62 (5), 600–610. <https://doi.org/10.1002/acr.20140>.