




Changes in cardiovascular health and physical functioning in non-hospitalized, adult COVID-19 patients after 3 years of follow-up

Janneke I. A. Vloet¹  | Esmée A. Bakker² | Koen M. van der Sluijs¹ |
 Guilherme F. Speretta³  | Armando van der Horst⁴ | Thijs M. H. Eijsvogels¹ |
 Dick H. J. Thijssen^{1,5} 

¹Department of Medical BioSciences, Radboud University Medical Center, Nijmegen, The Netherlands

²Department of Primary and Community Care, Radboud University Medical Center, Nijmegen, The Netherlands

³Department of Physiological Sciences, Biological Sciences Center, Federal University of Santa Catarina, Florianópolis, Brazil

⁴Laboratory of Clinical Chemistry and Haematology, Jeroen Bosch Hospital, 's-Hertogenbosch, The Netherlands

⁵Research Institute for Sports and Exercise Sciences, Liverpool John Moores University, Liverpool, UK

Correspondence

Dick H. J. Thijssen, Department of Medical BioSciences, Radboud University Medical Center, Nijmegen, The Netherlands.

Email: dick.thijssen@radboudumc.nl

Funding information

ZonMW Lifestyle medicine and COVID -19 SA 2022, Grant/Award Number: 05550322210002; Dutch Heart Foundation Grant, Grant/Award Number: 2020T063; Established Investigator E -Dekker Grant - Dutch Heart Foundation, Grant/Award Number: 03 -002 -2023 -0036

Abstract

Long-term cardiovascular consequences of COVID-19 remain unclear, particularly in non-hospitalized individuals. We investigated longitudinal changes (~3 years) in cardiovascular risk factors, cardiac biomarkers, physical functioning, and activity in non-hospitalized COVID-19 individuals versus age- and sex-matched controls. Assessments were performed in 2021 (~6 months post-infection) and repeated 3 years after initial SARS-CoV-2 infection among 101 non-hospitalized COVID-19 participants and 101 healthy age- and sex-matched controls. Multiple imputation was adopted to adjust for missing data due to dropout. Based on the absence (–) or presence (+) of infections during follow-up, four groups were created: COVID-19/– ($n = 45$), COVID-19/+ ($n = 56$), Control/– ($n = 29$), and Control/+ ($n = 72$). Additionally, sex-stratified analyses were performed. No changes were found in parameters related to cardiovascular risk factors, cardiac biomarkers, physical functioning and physical activity across the 3-year follow-up, showing no time \times group effects for any of the outcomes (all p -values > 0.05). Sex-stratified analyses and complete-case sensitivity analysis reinforced our results. Overall, we found no changes in cardiovascular risk factors, cardiac biomarkers, physical functioning and activity across a 3-year follow-up in non-hospitalized COVID-19 individuals compared to healthy controls, independent of infections during follow-up. This suggests that non-hospitalized COVID-19 did not induce long-term deleterious changes in cardiovascular health or physical characteristics.

KEYWORDS

cardiovascular health, incident infection, long-term effects, non-severe COVID-19, physical functioning

This is an open access article under the terms of the [Creative Commons Attribution](https://creativecommons.org/licenses/by/4.0/) License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

© 2026 The Author(s). *Physiological Reports* published by Wiley Periodicals LLC on behalf of The Physiological Society and the American Physiological Society.

1 | INTRODUCTION

Coronavirus Disease 2019 (COVID-19) has been associated with a range of cardiovascular complications during and immediately following infection, particularly among patients with severe acute infections who were hospitalized (Clerkin et al., 2020; Long et al., 2020). As the COVID-19 pandemic transitioned into an endemic phase, attention has shifted towards the potential longer-term consequences of a SARS-CoV-2 infection. Even mild infections, that is, representing the majority of infections globally, trigger acute subclinical inflammation and/or subtle alterations in cardiovascular function (Puntmann et al., 2022; Todorovic, 2022). Therefore, it is important to understand the possible persistent effects of COVID-19.

Epidemiological data from the early pandemic suggest that hospitalized and non-hospitalized individuals with COVID-19 had an elevated 1-year risk for incident cardiovascular disease (e.g., cerebrovascular disease, dysrhythmias, inflammatory heart disease, and thrombotic disorders) and mortality compared to individuals free of a COVID-19 infection (Wan et al., 2023; Xie et al., 2022). Studies also examined the impact of COVID-19 on cardiovascular biomarkers and risk factors but reported conflicting results. For example, we (van der Sluijs et al., 2023) and others (Kilinc et al., 2025) found no differences in cardiac biomarker concentrations and/or physical functioning between non-hospitalized COVID-19 patients and non-COVID-19 controls 6 months post-infection. In contrast, a recent study found mild-to-severe COVID-19 to be associated with increased arterial stiffness 1-year following infection, a finding primarily observed in women (Bruno et al., 2025), suggesting potential sex-related differences in longitudinal changes. These discrepant findings may be related to the strong variation in follow-up (3–12 months), disease severity, and population health status, which complicates comparison across studies. Importantly, the impact of COVID-19 is scarcely studied beyond 12 months following infection. This limits our current understanding of the potential impact of COVID-19 on cardiovascular health.

Therefore, we examined longitudinal changes in cardiovascular risk factors, cardiac biomarkers, physical functioning, and physical activity in non-hospitalized individuals with COVID-19 across a 3-year follow-up period. For this purpose, we re-invited 101 individuals with a COVID-19 infection (in 2020/21) and 101 age- and sex-matched controls at 3 years following their initial SARS-CoV-2 infection to repeat assessments of their cardiovascular health and functional status (van der Sluijs et al., 2023). Additionally, we examined whether longitudinal changes were sex specific. We hypothesized that,

in line with our observations at 6 months post-infection, no longitudinal changes or differences between non-hospitalized individuals with COVID-19 and their controls in cardiovascular risk factors, cardiac biomarkers, physical functioning, and physical activity were present in this relatively healthy, non-hospitalized cohort.

2 | MATERIALS AND METHODS

2.1 | Study design and population

This is a longitudinal follow-up study of a sub-cohort of the Nijmegen Exercise Study (van der Sluijs, Bakker, et al., 2025), a large cohort study including relatively healthy and physically active participants, which originally tested 101 adults ~6 months after their initial SARS-CoV-2 infection in 2021 who were matched for age and sex with non-infected controls (van der Sluijs et al., 2023). An inclusion criterion for the COVID-19 group was evidence of a positive polymerase chain reaction (PCR) test for SARS-CoV-2, whereas an exclusion criterion was hospitalization due to the SARS-CoV-2 infection. Age and sex-matched controls were only included if they never had signs, symptoms, or suspicions of a SARS-CoV-2 infection nor a lifetime positive test of any sort for SARS-CoV-2 before study participation. Dutch language proficiency and residency were inclusion criteria for both groups. The follow-up assessments were performed in 2024, ~36 months following the initial COVID-19 infection and ~30 months following the original evaluation in 2021. Based on the initial presence of COVID-19 (COVID-19 versus Control), but also the absence (–) or presence (+) of incident infection during follow-up, subjects were allocated to 4 groups: COVID-19/–, COVID-19/+, Control/–, and Control/+. Participants were invited by e-mail for a single visit to our research center (Radboud university medical center, Nijmegen, The Netherlands). All study procedures were kept similar between the two assessments. Randomization was not applicable to the current and previous measurements of this observational cohort study. The study procedures were approved by the local ethics committee Oost-Nederland (2023-16721) and the study was conducted in accordance with the Declaration of Helsinki. All participants provided written informed consent prior to participation in the study.

2.2 | Sample size

As our study has an explorative character, no formal sample size calculation was performed a priori. In the primary study, the Cohen's D effect size for the outcome measure

mean arterial pressure was 5, with an alpha of 0.05 and beta of 0.2 (power of 0.8). According to G*power (Version 3.1), the (a posteriori) estimated sample size needed was 79 participants in each study arm, so 158 in total. As groups were further subdivided based on COVID-19 status in the present study, these analyses should be interpreted as exploratory.

2.3 | Measurements

2.3.1 | COVID-19 characteristics

Upon follow-up, participants completed a questionnaire to assess demographics, general and cardiovascular health status, as well as characteristics related to COVID-19, including information on the number of incident infections, vaccination status, and hospitalization due to infection.

2.3.2 | Cardiovascular risk factors

Height (m) and weight (kg; measured with Seca, Hamburg, Germany) were assessed, and body mass index (BMI, kg/m^2) was calculated. Noninvasive left brachial blood pressure (mmHg) and heart rate (beats/min) were measured twice after 5 min of rest in a supine position using a semi-automated blood pressure device (M3, OMRON, Healthcare Europe, Hoofddorp, The Netherlands). The average values of the two measurements were used for analysis. Venous blood was drawn from the antecubital vein (8.5 mL, BD Vacutainer SST II Advance) and coagulated for 45–60 min before being centrifuged at 3000 revolutions/min for 10 min at 4°C. Serum was then transferred to 2-mL microtubes and stored at -80°C until analysis. The following biomarkers were analyzed: total cholesterol, high-density lipoprotein (HDL), low-density lipoprotein (LDL), triglycerides, glucose hexokinase, insulin, creatinine, and C-reactive protein (CRP). Biochemical analyses were performed batchwise on Atellica and IMMULITE 2000 analyzers (Siemens Healthcare Diagnostics, Tarrytown, NY) at the Jeroen Bosch Hospital ('s-Hertogenbosch, The Netherlands). Smoking behavior was inquired via questionnaires.

2.3.3 | Cardiac blood biomarkers

From the venous blood samples, high-sensitive cardiac troponin I (hs-cTnI) and amino-terminal pro-B-type natriuretic peptide (NT-proBNP) were analyzed. Biochemical

analyses were performed batchwise on Atellica and IMMULITE 2000 analyzers (Siemens Healthcare Diagnostics, Tarrytown, NY) at the Jeroen Bosch Hospital ('s-Hertogenbosch, The Netherlands).

2.3.4 | Physical functioning and physical activity

Peak handgrip strength (kg) of the nondominant hand was measured three times, separated by 1-min intervals using the Jamar hydraulic, according to Webb et al. (1989). The single highest value was used in the analysis. Gait speed (km/h) was assessed twice over a 4-m stretch with 2-m acceleration and deceleration zones on either side to ensure a stable pace. The fastest try was used in the analysis. This method follows the 4-m gait speed protocol, as described in the Short Physical Performance Battery (Webb et al., 1989). Ambulant physical activity monitoring (8 days, 24 h) was performed with an ActivPAL3 micro-accelerometer (PAL Technologies, Glasgow, UK). During this period, participants were requested to keep a sleep/wake diary to enable automated analysis of 24-h activities. A minimum of 4 valid days was used as an inclusion criterion for analysis. Data were extracted via PALbatch (PAL software Suite, version 8, PAL Technologies) and analyzed using a modified version of the script by Winkler et al. (2016), Bakker et al. (2021) in SAS (Statistical Analysis System, RRID: SCR_008567, version 9.4; SAS Inst., Cary, NC) to compute daily light intensity (LIPA) and moderate-to-vigorous physical activity (MVPA) duration (min/day), sleeping and sitting time (h/day), and step count (steps/day).

Assessors were not blinded to group status. All measurements were conducted by trained staff using standardized procedures identical to the 2021 assessment to ensure consistency and minimize interrater variability.

2.4 | Statistical analyses

All parameters were tested for normality by inspecting Q-Q plots. Normally distributed data were reported as mean (SD), while non-normally distributed data were reported as median [Q1–Q3] for continuous data. Categorical variables were reported as numbers (%). Missing data were imputed with multiple imputations by chained equations with predictive mean matching, since $n=75$ participants had missing data of at least one of the follow-up measurements (~37% missing at follow-up). After checking for patterns of missing data, the assumption of “missing at random” was followed. All mentioned variables present from the assessment in 2021 and 2024 were used to predict

missing values in 5 imputed datasets with 20 iterations. Healthy convergence, imputed distribution, and plausibility were verified.

To compare differences between groups over time, two-way repeated measures analysis of Variance (ANOVA) was performed, where Control/– served as a reference group. Given two repeated measurements, no additional covariance structure was required. Level of education, age, and sex were considered as confounders in the analyses. Based on findings of recent research, stratified analyses were performed for males and females to explore a potential sex difference (Bruno et al., 2025). Sensitivity analyses were performed using only the complete cases available within the dataset, as well as sensitivity analyses to assess potential differences between participants who participated in the follow-up measurements and those who did not respond to the invitation. p -values <0.05 were considered significant. p -values of time, group, and interaction between time and group were presented. Statistical analyses were performed in RStudio (2023.12.1 + 402) using packages mice (Van Buuren & Groothuis-Oudshoorn, 2011) and ggplot2 (Wickham, 2011).

3 | RESULTS

Of the 202 participants originally enrolled in the study (van der Sluijs et al., 2023), 128 positively responded to the invitation for a follow-up assessment. Participants had a median age of 58 years [54, 65] at baseline, with 118 (58%) male and 84 (42%) female participants. No outliers were excluded. After multiple imputation by chained equations with predictive mean matching was performed, pooled estimates indicated 45 participants were in the COVID-19/– group, 56 in the COVID-19/+ group, 29 in the Control/– group, and 72 in the Control/+ group. All groups showed vaccination rates of 85% or higher at 3-year follow-up (Table 1). No significant differences were observed in participant characteristics, cardiovascular risk factors, cardiac biomarkers, physical functioning, and physical activity between groups, except for a higher handgrip strength in the COVID-19/+ group (47 [34, 53] kg) when compared to the Control/+ group (Table 1).

3.1 | Cardiovascular risk factors, cardiac biomarkers and physical functioning and activity

We found no changes (all p -values >0.05) in cardiovascular risk factors across follow-up in the 4 distinct groups (Figure 1). For all cardiovascular risk factors, analyses revealed no significant effect across time, group, or

time \times group-interaction. Similarly, no significant changes were found for cardiac biomarkers (all p -values >0.05). No time, group, or time \times group-interaction effects were found in hs-cTnI and NT-proBNP across (Figure 2). Lastly, no changes were found across time, group, or time \times group-interaction for any of the markers of physical functioning (Figure 3) or physical activity behaviors (Figure 4) (all p -values >0.05). Additional stratified analyses exploring potential sex-related differences revealed no significant effects of time, group, and time \times group-interaction for males or females in cardiovascular risk factors, cardiac biomarkers, and physical functioning (all p -values >0.05 , Figures S1–S8).

3.2 | Sensitivity analyses

Baseline characteristics were similar between participants included in the analytical cohort ($n=127$) versus participants lost to follow-up ($n=75$) for cardiovascular risk, cardiac biomarkers, and most, but not all, markers of physical functioning. For example, we found participants from the analytical cohort to demonstrate higher MVPA (109 [89, 137] vs. 95 [80, 120], $p=0.009$) and step count (7007 [5782, 8871] vs. 6264 [5227, 7687], $p=0.018$) compared to those lost to follow-up (Table S2). In a subsequent sensitivity analysis, we repeated our primary analysis using a complete case approach using data from participants who visited for follow-up measurements, leading to 29 participants in the COVID-19/– group, 40 in the COVID-19/+ group, 14 in the Control/– group, and 43 in the Control/+ group. These analyses reinforced the main analyses, showing no time, group, and/or time \times group-interaction effects in any of the parameters (all p -values >0.05 , data not shown).

4 | DISCUSSION

In this study we aimed to explore the long-term (i.e., 3-year) changes in cardiovascular risk factors, cardiac biomarkers, physical functioning, and physical activity following non-hospitalized COVID-19. First, we found no changes across a 3-year follow-up in cardiovascular risk factors, cardiac biomarkers, physical functioning, and physical activity in those with a non-hospitalized COVID-19 infection and their age- and sex matched controls. Furthermore, these observations were independent of a re-infection with SARS-CoV-2 during follow-up and similarly present between men and women. Taken together, these observations suggest that an initial SARS-CoV-2 infection not requiring hospitalization is not associated with deleterious changes in cardiovascular risk,

TABLE 1 Population characteristics of the study population across the different groups according to the pooled estimates. For exploring differences between groups for numeric variables (Median [IQR]), a Kruskal–Wallis test is performed. The chi-squared test or Fisher's exact test is used for categorical variables (n (%)).

	COVID-19/– ($n = 45$)	COVID-19/+ ($n = 56$)	Control/– ($n = 29$)	Control/+ ($n = 72$)	<i>p</i> -value
Age (years)	60 [54, 67]	59 [55, 65]	60 [56, 64]	57 [53, 64]	0.38
Sex (male, %)	25 (57%)	34 (61%)	18 (70%)	40 (56%)	0.57
Alcohol status (current, %)	38 (86%)	50 (90%)	24 (85%)	65 (91%)	0.28
Smoking status (never, %)	23 (48%)	38 (68%)	16 (55%)	43 (60%)	0.42
Vaccination status (vaccinated in 2024, %)	39 (85.2%)	52 (92.6%)	28 (95.2%)	62 (88.4%)	0.29
Cardiovascular risk factors					
BMI (kg/m^2)	23.8 [22.2, 25.8]	24.4 [22.3, 25.9]	25.1 [23.3, 26.3]	24.6 [22.8, 26.9]	0.41
MAP (mmHg)	97 [93, 109]	97 [92, 105]	101 [95, 108]	99 [92, 108]	0.78
HR (beats/min)	60 [53, 65]	56 [51, 61]	57 [51, 64]	57 [53, 64]	0.35
Total cholesterol (mmol/L)	5.3 [4.6, 6.2]	5.2 [4.9, 5.8]	5.2 [4.8, 5.7]	5.0 [4.6, 5.8]	0.54
LDL (mmol/L)	3.1 [2.4, 4.0]	3.2 [2.6, 3.5]	3.1 [2.7, 3.7]	3.0 [2.5, 3.6]	0.87
HDL (mmol/L)	1.6 [1.3, 1.8]	1.5 [1.2, 1.8]	1.5 [1.2, 1.7]	1.5 [1.3, 1.8]	0.46
Triglycerides (mmol/L)	1.0 [0.8, 1.4]	1.1 [0.9, 1.4]	1.2 [0.9, 1.6]	1.0 [0.8, 1.3]	0.38
Insulin (mIU/mL)	4.2 [2.2, 7.7]	4.2 [2.1, 8.1]	3.9 [2.1, 5.5]	3.5 [2.0, 5.8]	0.42
Glucose (mmol/L)	5.1 [4.8, 5.4]	5.0 [4.8, 5.3]	5.0 [4.8, 5.2]	4.9 [4.7, 5.2]	0.34
Creatinine ($\mu\text{mol}/\text{L}$)	79.5 [71.0, 89.0]	83.8 [71.2, 90.0]	81.2 [75.1, 92.1]	79.7 [69.7, 87.2]	0.45
CRP (mg/L)	4.0 [4.0, 4.0]	4.0 [4.0, 4.0]	4.0 [4.0, 4.0]	4.0 [4.0, 4.0]	0.48
Cardiac biomarkers					
NT-proBNP (pg/mL)	8.6 [5.4, 12.8]	8.5 [5.3, 14.3]	7.9 [5.0, 12.4]	6.6 [4.0, 10.7]	0.42
Hs-cTnI (ng/L)	4.2 [2.5, 7.1]	4.4 [2.7, 10.3]	4.0 [2.6, 5.8]	3.3 [2.5, 7.5]	0.19
Physical functioning					
Handgrip strength (kg)	41 [32, 52]	47 [34, 53]	42 [35, 49]	37 [30, 48]	0.018
4-m walking speed (km/h)	5.5 [5.1, 6.0]	5.5 [5.1, 6.3]	5.8 [5.1, 6.3]	5.8 [5.3, 6.3]	0.44
24-h Physical activity behavior					
Sitting time (h/day)	9.0 [8.1, 10.2]	9.3 [8.2, 10.2]	9.4 [8.8, 10.3]	9.2 [8.3, 10.4]	0.75
MVPA (min/day)	102 [87, 121]	105 [79, 129]	116 [92, 138]	108 [86, 132]	0.44
LIPA (min/day)	284 [222, 324]	273 [224, 317]	253 [196, 327]	243 [206, 308]	0.30
Step count (steps/day)	6577 [5612, 8221]	6732 [5101, 8249]	7164 [5765, 8597]	7018 [5707, 8525]	0.61
Sleeping time (h/day)	8.6 [7.6, 9.7]	8.8 [7.8, 9.2]	7.8 [7.1, 8.8]	8.3 [7.3, 9.6]	0.15

Abbreviations: BMI, Body Mass Index; CRP, C-Reactive Protein; HDL, high-density lipoprotein; HR, heart rate; Hs-cTnI, high-sensitivity cardiac Troponin I; LDL, low-density lipoprotein; LIPA, light intensity physical activity; MAP, mean arterial pressure; MVPA, moderate-to-vigorous physical activity; NT-proBNP, N-terminal pro Brain Natriuretic Peptide; SB, sedentary behavior.

cardiac biomarkers and physical functioning across 3 years of follow-up.

In line with our hypothesis and initial observations at 6 months post-infection (van der Sluijs et al., 2023), we found no change in cardiovascular risk factors, cardiac blood biomarkers, physical functioning, and physical activity across 3 years of follow-up after the initial COVID-19 infection. To date, only a few studies have explored the effects of COVID-19 beyond 1 year. Some studies report altered cardiovascular health in the first weeks or months after COVID-19 not requiring hospitalization,

including mild elevations in cardiac biomarkers, reduced exercise tolerance, or subtle changes in vascular function (Brown et al., 2022; Keefe et al., 2022). Longer follow-up, including 6- or 12-months, shows a return to normal values, suggesting at least a partial recovery of cardiovascular health over time (Russell et al., 2025; Willems et al., 2022, 2023; Yang et al., 2024). Also, studies with repeated measures up to 18 months post-COVID-19 support this notion when examining non-hospitalized patients for vascular function, inflammation and cardiac biomarkers, and CVD outcomes (Russell et al., 2025;

Cardiovascular Risk Factors – Total

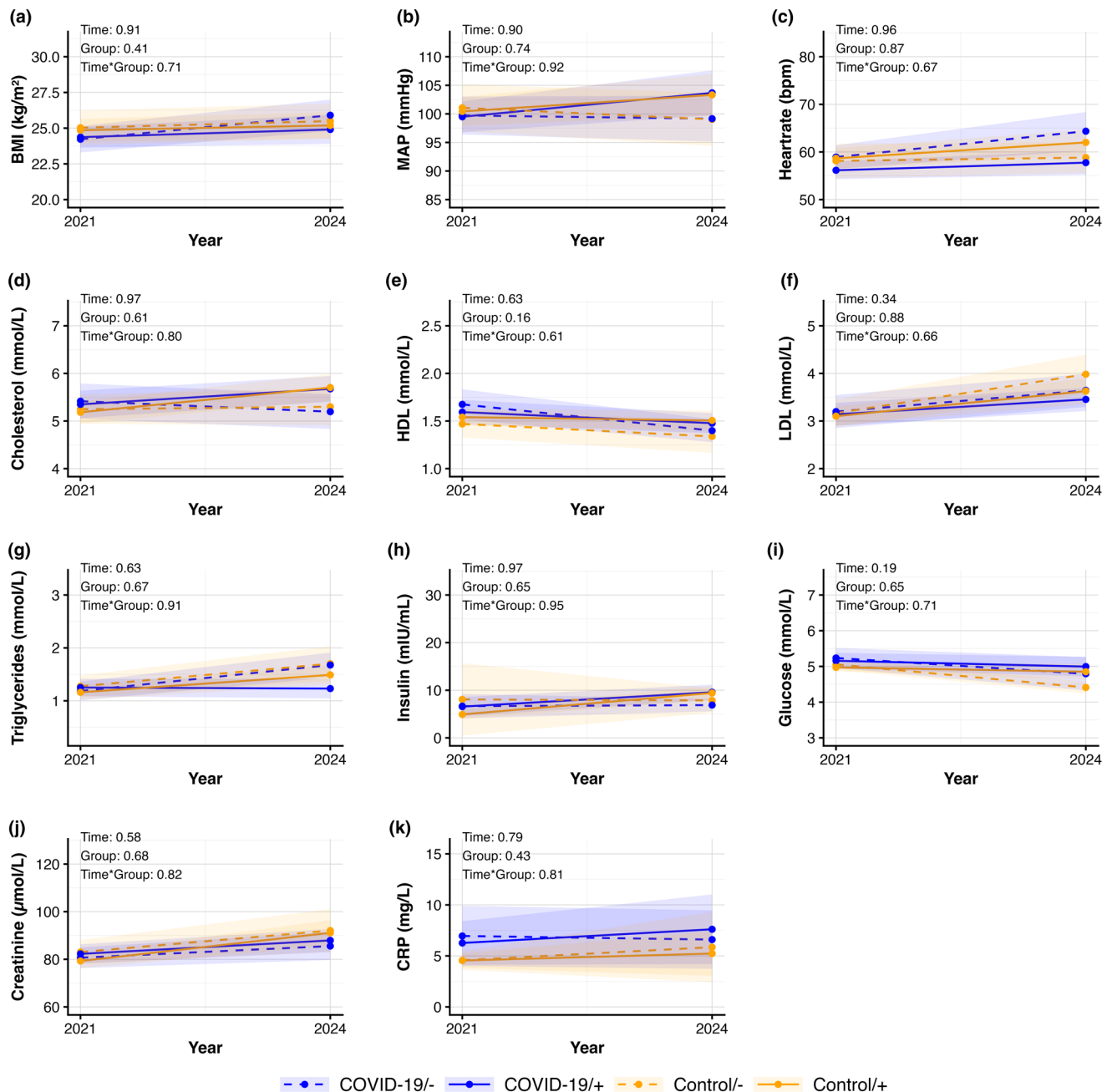


FIGURE 1 Lines represent data after multiple imputation for cardiovascular risk factors Body Mass Index (BMI, a), mean arterial pressure (MAP, b), Heartrate (HR, c), total cholesterol (d), HDL cholesterol (e), LDL cholesterol (f), triglycerides (g), Insulin (h), glucose Hexikinase (i), creatinine (j), and CRP (k) which are presented at 6-months and 3-years follow-up following initial COVID-19 infection (blue) and their age- and sex-matched controls (orange), with both groups divided into those with re-infection (+, solid lines) or free of re-infection during follow-up (–, dashed lines). Lines representing mean and 95% CI for the initial (2021) and follow-up assessment (2024) per group. *p*-values represent the outcomes of a two-way ANOVA reflecting the effects of time, group and time × group-interaction of the outcome.

Willems et al., 2022; Willems et al., 2023; Yang et al., 2024). We extend these observations by revealing no change in cardiovascular risk factors and cardiac biomarkers between 6 months and 3 years of follow-up in non-hospitalized COVID-19 patients. This suggests that

an initial SARS-CoV-2 infection without hospitalization is not associated with long-term (3-year) detrimental effects on cardiovascular health.

In understanding our results, it is important to consider that we have included cases with mild COVID-19

Cardiac Biomarkers – Total

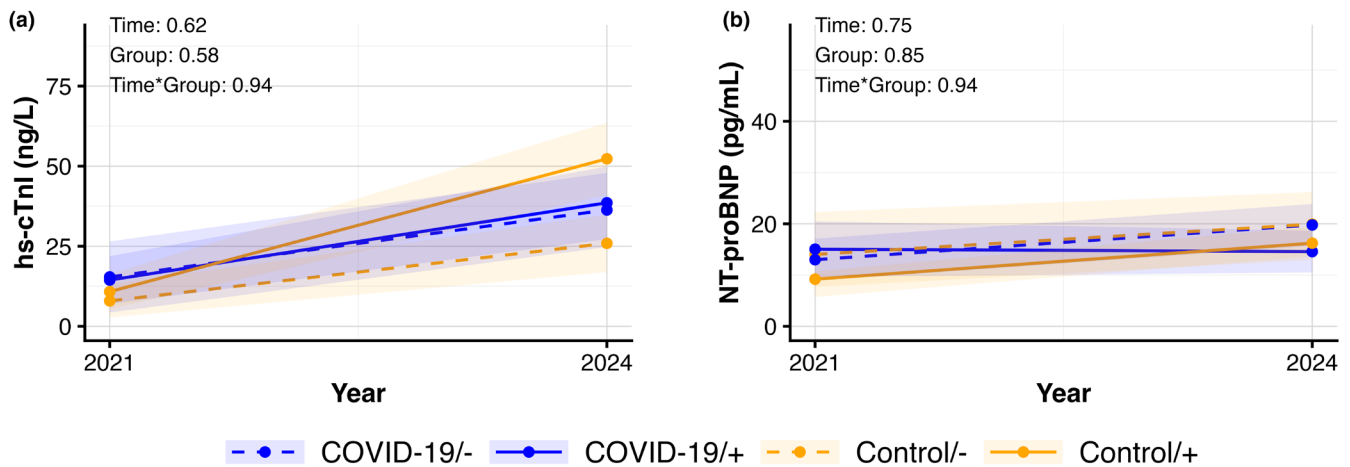


FIGURE 2 Lines represent data after multiple imputation for cardiac biomarkers hs-cTnI (a) and NT-proBNP (b) which are presented at 6-months and 3-years follow-up following initial COVID-19 infection (blue) and their age- and sex-matched controls (orange), with both groups divided into those with re-infection (+, solid lines) or free of re-infection during follow-up (–, dashed lines). Lines representing mean and 95% CI for the initial (2021) and follow-up assessment (2024) per group. *p*-values represent the outcomes of a two-way ANOVA reflecting the effects of time, group and time \times group-interaction of the outcome.

Physical Functioning – Total

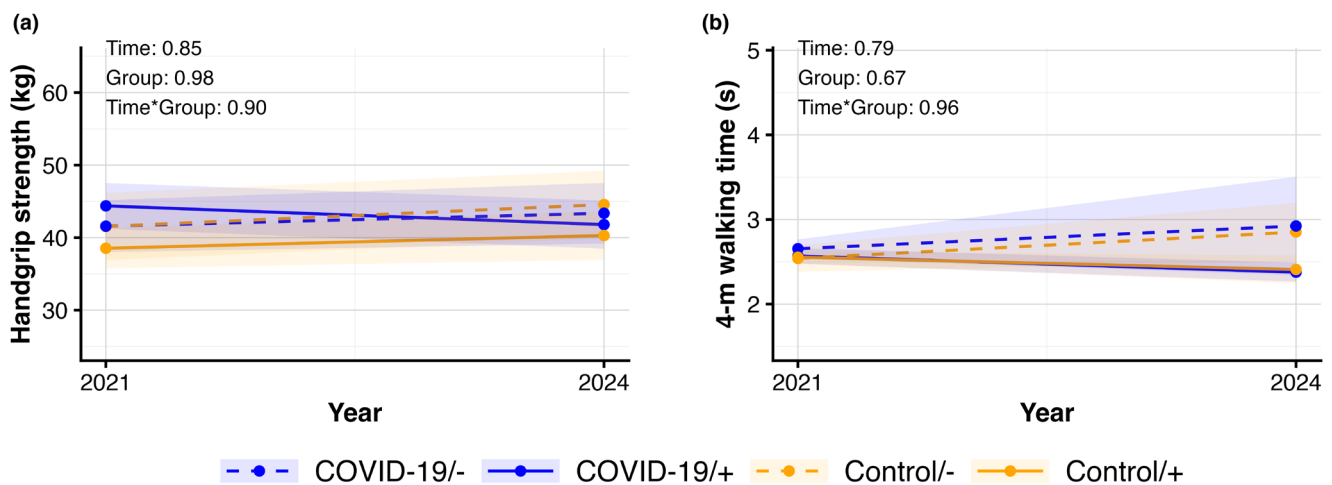


FIGURE 3 Lines represent data after multiple imputation for handgrip strength (a), and 4-m walking time (b) which are presented at 6-months and 3-years follow-up following initial COVID-19 infection (blue) and their age- and sex-matched controls (orange), with both groups divided into those with re-infection (+, solid lines) or free of re-infection during follow-up (–, dashed lines). Lines representing mean and 95% CI for the initial (2021) and follow-up assessment (2024) per group. *p*-values represent the outcomes of a two-way ANOVA reflecting the effects of time, group and time \times group-interaction of the outcome.

infections only. Requiring hospitalization and/or ICU admission may induce more severe and prolonged cardiovascular effects through impairment of microcirculatory and endothelial function (Poyatos et al., 2024; Sabioni et al., 2023). In support of this, Bruno et al. recently examined arterial stiffness in 2390 individuals ~6 months following COVID-19, and confirmed a more severe impact of COVID-19 on arterial stiffness in those requiring ICU admission (Bruno et al., 2025). Moreover, Bruno

et al. reported that their observations seem driven by sex differences, as the increased stiffness is primarily driven by observations in women. For this reason, we performed stratified analyses on our data. We could not identify a potential sex difference regarding the long-term effects of COVID-19 in non-hospitalized individuals on our measures of cardiovascular risk and cardiac biomarkers. Severity of disease, but also differences in outcome measures and duration of follow-up, may explain conflicting

Physical Activity Behaviour – Total

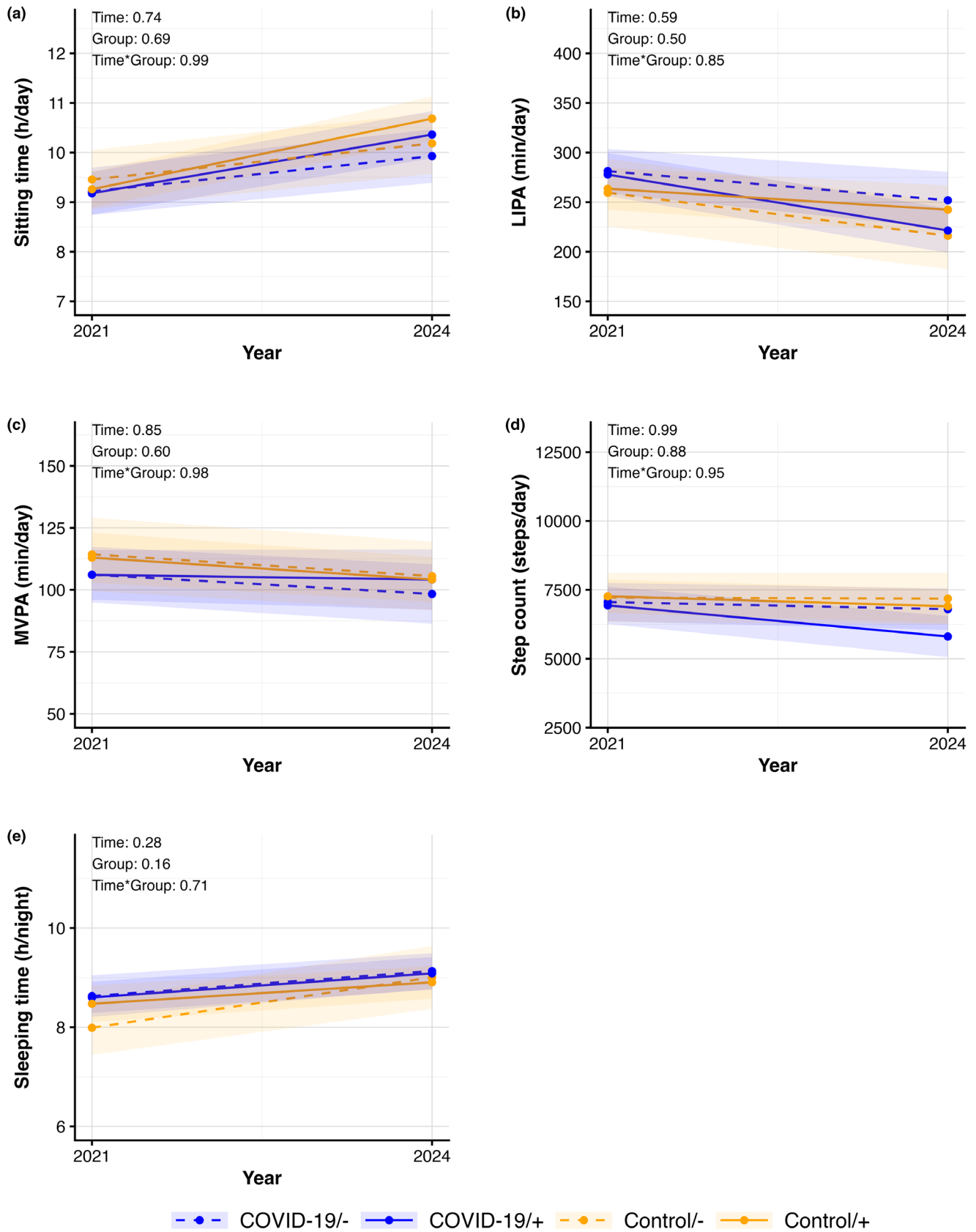


FIGURE 4 Lines represent data after multiple imputation for physical functioning for sitting time (a), Light intensity physical activity (LIPA, b), Moderate-to-vigorous physical activity (MVPA, c), step count (d), and sleeping time (e) which are presented at 6-months and 3-years follow-up following initial COVID-19 infection (blue) and their age- and sex-matched controls (orange), with both groups divided into those with re-infection (+, solid lines) or free of re-infection during follow-up (–, dashed lines). Lines representing mean and 95% CI for the initial (2021) and follow-up assessment (2024) per group. *p*-values represent the outcomes of a two-way ANOVA reflecting the effects of time, group and time × group-interaction of the outcome.

results between studies. To better understand the long-term effects of COVID-19, we recommend future studies to extend the follow-up beyond 12 months, but also to assess the role of disease severity, sex and other factors, such as microvascular function, thrombosis, and endothelial function.

A strong point of our study was the evaluation of habitual physical activity and physical functioning, especially given its strong relation with cardiovascular health (Lin et al., 2020). Previously, we observed that the lockdown substantially increased time spent in sedentary behavior (van Bakel et al., 2021), which did not return to pre-lockdown levels after 3 years of follow-up (Vloet et al., 2025). COVID-19 also substantially affects physical activity characteristics in the days and weeks following infection (Gupta et al., 2023; van Bakel et al., 2022). Nonetheless, and in line with our 6-month observations and our hypothesis, we found no difference in physical functioning and physical activity between non-hospitalized individuals with COVID-19 and their age- and sex-matched controls across 3 years of follow-up. An important consideration is that our participants are recruited from a relatively healthy cohort with participants who nearly all meet the international guidelines of physical activity (van der Sluijs, Bakker, et al., 2025). Given that higher levels of habitual physical activity may protect against long-term consequences of COVID-19 (Gomide et al., 2022; Halabchi et al., 2023), the observed long-term effects of COVID-19 on our outcomes may have been relatively small. Accordingly, our findings primarily apply to relatively healthy individuals with a low prevalence of pre-existing comorbidities and cardiovascular risk factors, limiting generalizability to less healthy or less physically active populations.

Another observation from our study is that incident infection(s) did not impact the change in cardiovascular risk factors, cardiac biomarkers, physical functioning, or physical activity between 6 months and 3 years of follow-up in the non-hospitalized COVID-19 participants and those without infection at initial inclusion. To our knowledge, no previous study addressed this topic, while this seems relevant given the relatively high prevalence of COVID-19 (re-)infections. An important factor to consider is that the severity of symptoms following a COVID-19 infection has

substantially dropped, including the risk for post-COVID complaints (Hu et al., 2023). Due to more mild complaints, we may have underestimated the true SARS-CoV-2 (re-) infection rates as we depend on self-reported re-infection rather than a PCR-based confirmation of re-infection. At the very least, our data suggest that SARS-CoV-2 re-infection, either in those with or without SARS-CoV-2 infection in 2020 or 2021, does not alter our outcome parameters across a 3-year follow-up.

Our study has several strengths, including the long follow-up, the within-subject design, and the comprehensive assessment of cardiovascular health, physical functioning and activity. However, some limitations should be discussed. First, not all participants returned to our laboratory for the 3-year follow-up testing. However, the return rate was high (63%), despite such a long follow-up, and we used multiple imputation techniques to include all participants in our analyses. As multiple imputation under a Missing-At-Random assumption may shrink between-group variability, this approach could bias findings towards the null. However, complete-case analyses showed similar results, indicating that any imputation-related bias is likely limited. Second, due to the observational nature of this study, causal relationships could not be established. Although we adjusted for a range of potential confounders, residual and unmeasured confounding cannot be ruled out. Third, our cohort consisted of relatively healthy and physically active adults. Previous work highlighted the benefits of regular physical activity on, for example, body composition and glucose metabolism (van der Sluijs et al., 2025). Therefore, including relatively active participants may attenuate the magnitude of potential long-term effects and limits generalizability to older or clinically more vulnerable populations. Additionally, detailed information on vaccination timing, vaccine type, and variant exposure was incomplete and partly self-reported; therefore, these variables were not included in analytical models.

In conclusion, we did not detect evidence that COVID-19 in non-hospitalized individuals, who were infected during the first waves of the pandemic in 2020/21, was associated with deleterious changes in cardiovascular risk factors, cardiac biomarkers, physical functioning, or physical activity behavior across 3 years of follow-up.

AUTHOR CONTRIBUTIONS

Janneke I. A. Vloet: Conceptualization; data curation; formal analysis; investigation; methodology; project administration; resources; software; visualization. **Esmée A. Bakker:** Conceptualization; methodology; supervision. **Koen M. van der Sluijs:** Conceptualization; investigation; methodology; project administration. **Guilherme F. Speretta:** Investigation. **Armando van der Horst:** Resources. **Thijs M. H. Eijsvogels:** Conceptualization; funding acquisition; supervision. **Dick H. J. Thijssen:** Conceptualization; funding acquisition; supervision.

ACKNOWLEDGMENTS

We sincerely thank all personnel involved in the data collection and Siemens Healthcare Diagnostics for providing the assay kits for the biomarker analyses. Additionally, we want to thank Prof. Dr. Maryam Kavousi, Prof. Dr. Geert-Jan Geersing, and Prof. Dr. Frans H. Rutten for their contributions to the design of the initial study.

FUNDING INFORMATION

This project was funded by ZonMW Lifestyle medicine and COVID-19 SA 2022 (#05550322210002) and by the Dutch Heart Foundation Grant 2020T063. TE was supported by an Established Investigator E-Dekker grant (#03-002-2023-0036) of the Dutch Heart Foundation.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT


Data will be made available upon reasonable request.

ETHICS STATEMENT

This study was conducted in accordance with the Declaration of Helsinki and was approved by the local Ethics committee (2023-16721). Written informed consent was obtained from all participants prior to their participation in the study.

ORCID

Janneke I. A. Vloet  <https://orcid.org/0009-0000-4001-5117>

Guilherme F. Speretta  <https://orcid.org/0000-0003-3474-1264>

Dick H. J. Thijssen  <https://orcid.org/0000-0002-7707-5567>

REFERENCES

- Bakker, E. A., van Bakel, B. M., Aengevaeren, W. R., Meindersma, E. P., Snoek, J. A., Waskowsky, W. M., & van Kuijk, A. A. (2021). Sedentary behaviour in cardiovascular disease patients: Risk group identification and the impact of cardiac rehabilitation. *International Journal of Cardiology*, 326, 194–201.
- Brown, J. T., Saigal, A., Karia, N., Patel, R. K., Razvi, Y., Constantinou, N., Steeden, J. A., Mandal, S., Kotecha, T., Fontana, M., Goldring, J., Muthurangu, V., & Knight, D. S. (2022). Ongoing exercise intolerance following COVID-19: A magnetic resonance-augmented cardiopulmonary exercise test study. *Journal of the American Heart Association*, 11(9), e024207.
- Bruno, R. M., Badhwar, S., Abid, L., Agharazii, M., Anastasio, F., Bellien, J., Burghuber, O., Faconti, L., Filipovsky, J., Ghiadoni, L., Giannattasio, C., Hametner, B., Hughes, A. D., Jeroncic, A., Ikonomidis, I., Lonnebakk, M. T., Maloberti, A., Mayer, C. C., Muiesan, M. L., ... Bates, L. (2025). Accelerated vascular ageing after COVID-19 infection: The CARTESIAN study. *European Heart Journal*, 46, 3905–3918.
- Clerkin, K. J., Fried, J. A., Raikhelkar, J., Sayer, G., Griffin, J. M., Masoumi, A., Jain, S. S., Burkhoff, D., Kumaraiah, D., Rabbani, L. R., Schwartz, A., & Uriel, N. (2020). COVID-19 and cardiovascular disease. *Circulation*, 141(20), 1648–1655.
- Gomide, E. B. G., Abdalla, P. P., Pisa, M. F., Schneider, G., Vieira, L. G., Mazzonetto, L. F., & de Sousa Oliveira, A. (2022). The role of physical activity in the clinical outcomes of people diagnosed with Covid-19: A systematic review. *JSAMS Plus*, 1, 100007.
- Gupta, V., Kariotis, S., Rajab, M. D., Errington, N., Alhathli, E., Jammeh, E., & Brook, M. (2023). Unsupervised machine learning to investigate trajectory patterns of COVID-19 symptoms and physical activity measured via the MyHeart counts app and smart devices. *npj Digital Medicine*, 6(1), 239.
- Halabchi, F., Mahdavian, B., Tazesh, B., Shab-Bidar, S., & Selk-Ghaffari, M. (2023). Association between physical activity and risk of COVID-19 infection or clinical outcomes of the patients with COVID-19: A systematic review and meta-analysis. *Journal of Preventive Medicine and Hygiene*, 64(2), E123.
- Hu, F. H., Jia, Y. J., Zhao, D. Y., Fu, X. L., Zhang, W. Q., Tang, W., Hu, S. Q., Wu, H., Ge, M. W., du, W., Shen, W. Q., Zhu, B., & Chen, H. L. (2023). Clinical outcomes of the severe acute respiratory syndrome coronavirus 2 omicron and Delta variant: Systematic review and meta-analysis of 33 studies covering 6 037 144 coronavirus disease 2019-positive patients. *Clinical Microbiology and Infection*, 29(7), 835–844.
- Keefe, J. A., Avadhanula, V., Nicholson, E. G., Devaraj, S., Piedra, P. A., Bozkurt, B., & Wehrens, X. H. T. (2022). Abnormalities in cardiac and inflammatory biomarkers in ambulatory subjects after COVID-19 infection. *International Journal of Cardiology Heart & Vasculature*, 43, 101144.
- Kilinc, O., Bisen, J. B., Maroun, A., Johnson, E., Quinn, S., Kim, D., Lee, D. C., Markl, M., & Allen, B. D. (2025). 4D flow MRI derived aortic pulse wave velocity and wall shear stress in Covid-19. *Clinical Imaging*, 124, 110524.
- Lin, H., Sardana, M., Zhang, Y., Liu, C., Trinquart, L., Benjamin, E. J., Manders, E. S., Fusco, K., Kornej, J., Hammond, M. M., Spartano, N. L., Pathiravasan, C. H., Khetarpal, V., Nowak, C., Borrelli, B., Murabito, J. M., & McManus, D. D. (2020). Association of Habitual Physical Activity with Cardiovascular Disease Risk. *Circulation Research*, 127(10), 1253–1260.
- Long, B., Brady, W. J., Koyfman, A., & Gottlieb, M. (2020). Cardiovascular complications in COVID-19. *The American Journal of Emergency Medicine*, 38(7), 1504–1507.
- Poyatos, P., Luque, N., Sabater, G., Eizaguirre, S., Bonnin, M., Orriols, R., & Tura-Ceide, O. (2024). Endothelial dysfunction

- and cardiovascular risk in post-COVID-19 patients after 6- and 12-months SARS-CoV-2 infection. *Infection*, 52(4), 1269–1285.
- Puntmann, V. O., Martin, S., Shchendrygina, A., Hoffmann, J., Ka, M. M., Giokoglu, E., Vanchin, B., Karyou, A., Laux, G. S., Arendt, C., de Leuw, P., Zacharowski, K., Khodamoradi, Y., Vehreschild, M. J. G. T., Rohde, G., Zeiher, A. M., Vogl, T. J., Schwenke, C., & Nagel, E. (2022). Long-term cardiac pathology in individuals with mild initial COVID-19 illness. *Nature Medicine*, 28(10), 2117–2123.
- Russell, S. L., Okwose, N. C., Rahman, M., Lee, B. J., McGregor, G., Raleigh, S. M., & Sandhu, H. (2025). The effect of COVID-19 on cardiovascular function and exercise tolerance in healthy middle-age and older individuals. *Scandinavian Cardiovascular Journal*, 59(1), 2468339.
- Sabioni, L., De Lorenzo, A., Castro-Faria-Neto, H. C., Estado, V., & Tibirica, E. (2023). Long-term assessment of systemic micro-circulatory function and plasma cytokines after coronavirus disease 2019 (COVID-19). *The Brazilian Journal of Infectious Diseases*, 27(1), 102719.
- Todorovic, V. (2022). Mild SARS-CoV-2 infection leaves long-lasting effects on cardiovascular health. *Nature Cardiovascular Research*, 1(10), 870.
- van Bakel, B. M. A., Bakker, E. A., de Vries, F., Thijssen, D. H. J., & Eijsvogels, T. M. H. (2021). Impact of COVID-19 lockdown on physical activity and sedentary behaviour in Dutch cardiovascular disease patients. *Netherlands Heart Journal*, 29(5), 273–279.
- van Bakel, B. M. A., van den Heuvel, F. M. A., Vos, J. L., Rotbi, H., Bakker, E. A., Nijveldt, R., Thijssen, D. H. J., & Eijsvogels, T. M. H. (2022). High levels of sedentary time in patients with COVID-19 after hospitalisation. *Journal of Clinical Medicine*, 11(4), 1110.
- Van Buuren, S., & Groothuis-Oudshoorn, K. (2011). Mice: Multivariate imputation by chained equations in R. *Journal of Statistical Software*, 45, 1–67.
- van der Sluijs, K. M., Bakker, E. A., Schoofs, M. C. A., Vloet, J. I. A., Verbeek, A. L. M., Hopman, M. T. E., Thijssen, D. H. J., & Eijsvogels, T. M. H. (2025). Cohort profile: The Nijmegen exercise study (NES). *International Journal of Epidemiology*, 54(4), dyaf092.
- van der Sluijs, K. M., Bakker, E. A., Schuijt, T. J., Joseph, J., Kavousi, M., Geersing, G. J., Rutten, F. H., Hartman, Y. A. W., Thijssen, D. H. J., & Eijsvogels, T. M. H. (2023). Long-term cardiovascular health status and physical functioning of nonhospitalized patients with COVID-19 compared with non-COVID-19 controls. *American Journal of Physiology. Heart and Circulatory Physiology*, 324(1), H47–h56.
- van der Sluijs, K. M., Vloet, J. I. A., Thijssen, D. H. J., Eijsvogels, T. M. H., Collaboration NES, & Bakker, E. A. (2025). Cardiometabolic benefit of replacing sedentary time with light-intensity physical activity: Compositional data analysis of the Nijmegen exercise study. *Medicine and Science in Sports and Exercise*, 57(8), 1711–1720.
- Vloet, J. I. A., Bakker, E. A., van Bakel, B. M. A., Kroesen, S. H., Thijssen, D. H. J., & Eijsvogels, T. M. H. (2025). Do cardiovascular disease patients return to pre-lockdown sedentary levels? A prospective cohort study. *Netherlands Heart Journal*, 33(7-8), 232–238.
- Wan, E. Y. F., Mathur, S., Zhang, R., Yan, V. K. C., Lai, F. T. T., Chui, C. S. L., & Li, X. (2023). Association of COVID-19 with short- and long-term risk of cardiovascular disease and mortality: A prospective cohort in UK biobank. *Cardiovascular Research*, 119(8), 1718–1727.
- Webb, A. R., Newman, L. A., Taylor, M., & Keogh, J. B. (1989). Hand grip dynamometry as a predictor of postoperative complications reappraisal using age standardized grip strengths. *Journal of Parenteral and Enteral Nutrition*, 13(1), 30–33.
- Wickham, H. (2011). ggplot2. *WIREs Computational Statistics*, 3(2), 180–185.
- Willems, L. H., Jacobs, L. M. C., Groh, L. A., Ten Cate, H., Spronk, H. M. H., Wilson-Storey, B., & Hannink, G. (2023). Vascular function, systemic inflammation, and coagulation activation 18 months after COVID-19 infection: An observational cohort study. *Journal of Clinical Medicine*, 12(4), 1413.
- Willems, L. H., Nagy, M., Ten Cate, H., Spronk, H. M. H., Groh, L. A., Leentjens, J., & Janssen, N. A. F. (2022). Sustained inflammation, coagulation activation and elevated endothelin-1 levels without macrovascular dysfunction at 3 months after COVID-19. *Thrombosis Research*, 209, 106–114.
- Winkler, E. A., Bodicoat, D. H., Healy, G. N., Bakrania, K., Yates, T., Owen, N., Dunstan, D. W., & Edwardson, C. L. (2016). Identifying adults' valid waking wear time by automated estimation in activPAL data collected with a 24 h wear protocol. *Physiological Measurement*, 37(10), 1653–1668.
- Xie, Y., Xu, E., Bowe, B., & Al-Aly, Z. (2022). Long-term cardiovascular outcomes of COVID-19. *Nature Medicine*, 28(3), 583–590.
- Yang, Q., Chang, A., Tong, X., Jackson, S. L., & Merritt, R. K. (2024). Long-term cardiovascular disease outcomes in non-hospitalized medicare beneficiaries diagnosed with COVID-19: Population-based matched cohort study. *PLoS One*, 19(5), e0302593.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Vloet, J. I. A., Bakker, E. A., van der Sluijs, K. M., Speretta, G. F., van der Horst, A., Eijsvogels, T. M. H., & Thijssen, D. H. J. (2026). Changes in cardiovascular health and physical functioning in non-hospitalized, adult COVID-19 patients after 3 years of follow-up. *Physiological Reports*, 14, e70868. <https://doi.org/10.14814/phy.2.70868>