Effect of 4-Week Heartfulness Meditation on Stress Scores, Sleep Quality, and Oxidative and Inflammatory Biochemical Parameters in COVID-19 Patients after Completion of Standard Treatment – A Randomized Controlled Trial

Abstract

Context: COVID-19-affected patients showed increased stress, impaired sleep quality, altered complete blood count, and increased inflammatory and oxidative parameters. Yoga is an add-on nonpharmacological treatment that is established to normalize the abovementioned parameters. Heartfulness meditation is a form of Raja yoga. Aims: The present study aimed to study the effects of 4 weeks of heartfulness meditation on the abovementioned parameters in COVID-19 patients following treatment completion. Settings and Design: The present study was a randomized controlled trial carried out in the Department of Physiology, AIIMS, Mangalagiri, Andhra Pradesh. Subjects and Methods: Out of 50 COVID-19-treatment-completed patients recruited for the study, 25 were randomly assigned to the study group who received 4-week app-based heartfulness meditation. Other 25 patients were assigned to the control group who received app-based relaxation for 4 weeks. Perceived stress score, Pittsburgh Sleep Quality Index questionnaire, baseline cardiovascular parameters, complete blood count, serum cortisol, inflammatory parameters, oxidative stress parameters, and antioxidant parameters were assessed before and after 4 weeks of intervention in both the groups. The outcome assessor was blinded in the present study. Statistical Analysis Used: The mean difference between the two groups was tested using the Student’s t-test or Mann–Whitney U-test based on data distribution. Effect of intervention was analyzed using paired Student’s t-test for dependent samples test or Wilcoxon signed-rank test based on data distribution. Results: The groups were comparable before intervention for all the variables. After 4 weeks of intervention, we observed a significant decrease in stress, circulating cortisol, inflammatory markers, and oxidative stress biomarker in both the groups. Further, we observed improved sleep quality and antioxidant biomarkers in both the groups. These beneficial alterations following intervention were high in the study group compared to the control group. Conclusions: Our results suggest that app-based heartfulness meditation/relaxation can be used as a nonpharmacological adjuvant to hasten the recovery process in patients who have completed the COVID-19 treatment protocol. Beneficial effects in subjects practicing heartfulness meditation were more than that observed in subjects practicing relaxation.

Keywords: COVID-19, heartfulness meditation, relaxation therapy

Introduction

The recent COVID-19 pandemic has affected all walks of life globally, including health and psychological well-being, especially in COVID-positive subjects.[1,2] Common psychological symptoms are anxiety, depression, self-reported stress, and disturbed sleep.[3] Emerging evidence implicates inflammatory and oxidative stress in depression pathophysiology.[4,5] COVID-19 subjects show dysregulated cytokine responses with inflammation.[6,7] The severity of depressive symptoms in COVID-19 subjects correlated well with inflammatory indicator C-reactive protein (CRP).[8] On the other hand, sleep disturbance and perceived stress observed during this pandemic can lead to increased inflammation and oxidative stress.[9,10] Hence, this pandemic creates a clinical scenario that perpetuates itself and its effects prolong even after following the treatment guidelines. It has been noted that around 96% of subjects recovering from
COVID-19 suffer from posttraumatic stress symptoms.\(^2\) Hence, it becomes imperative to address the psychological aspects of COVID-19 in overcoming this global pandemic.\(^{11}\)

In the present scenario, where COVID-positive subjects will be quarantined or when the travel options are minimal, a free online option to manage their stress would empower them to overcome this situation. Many scientific works of literature have shown that meditation helps in reducing negative dimensions of psychological stress,\(^{12}\) especially in posttraumatic stress.\(^{13}\) Heartfulness meditation is a contemplative Raja yoga-based practice that is available on the android platform free of cost.\(^{14}\) This technique is shown to reduce perceived stress,\(^{15}\) decrease burnout, improve emotional wellness,\(^{16}\) and improve sleep quality\(^{17}\) previously.

We have planned this study to assess the effect of 4-week app-based heartfulness meditation on perceived stress, sleep quality, complete blood count, serum cortisol, inflammatory parameters, oxidative stress parameters, and antioxidant parameters in COVID-19 patients following treatment completion.

Subjects and Methods

Study design and setting

It was a randomized controlled trial conducted in the Department of Physiology, in collaboration with the Department of Medicine, Biochemistry and Community Medicine, All India Institute of Medical Sciences, Mangalagiri, Andhra Pradesh, India. The study was commenced after obtaining approval from the Institutional Ethical Committee for human studies (AIIMS/MG/IEC/2021-22/100). The trial was registered in the Clinical Trials Registry-India (CTRI registration number – CTRI/2021/06/034304).

Recruitment of the participants

Study population

COVID-19 patients in the moderate category in the age group of 18–50 years of either gender who were diagnosed and treated as per the guidelines of the Ministry of Health and Family Welfare, Government of India, in XXXX, XXXXXXX COVID-19 care center, during the period of June 2021 to November 2021 were considered for the study. We explained the study details to the patients who were about to get discharged. A consecutive sampling method was used. A total of 50 subjects without any comorbid condition such as diabetes/hypertension, not on any other treatments, and psychiatric illness were enrolled in the study after obtaining their written informed consent. We considered only subjects with moderate category severity who had steroid therapy and have not taken remdesivir or any other antiviral treatments.\(^{18}\) The participants were requested to visit the Department of Physiology, XXXXXXX, XX, between 8.00 AM to 9.00 AM on the day of their discharge.

Randomization and allocation

We have planned two groups; the study group would follow heartfulness meditation and the control group would follow relaxation. The participants were allotted to groups based on a computer-generated randomization method using the sequentially numbered sealed opaque envelopes. Twenty-five were assigned to the study group, and 25 were assigned to the control group. Figure 1 shows the study flow.

Parameters measured

We collected demographic and personal details such as age, gender, and contact number. Following this, the Perceived Stress Questionnaire\(^{19}\) and Pittsburgh Sleep Quality Index questionnaire\(^{20}\) were to assess their stress level and sleep quality, respectively. Anthropometric measurement – weight was measured using a digital weighing scale (MS 4900, Charder Electronics Co. Ltd, Taichung, Taiwan) to the nearest half a kilogram, and height was measured using a wall-mounted stadiometer (BHH6, Easy Care, Mumbai, India). Body mass index (BMI) was obtained using Quetelet index (BMI = weight [kg]/height [m] 2). Heart rate (HR) and blood pressure were recorded following 10 min of rest in the sitting posture with a backrest using an automated blood pressure monitor (OMRON HEM-8712, Omron Healthcare Co. Ltd, Kyoto, Japan). Oxygen saturation (SpO2) levels were measured using a digital pulse oximeter (ZEB-FPO600, Zebronics India Pvt. Ltd, India).

We collected 10 mL of venous blood in the aseptic method from the brachial vein for biochemical analysis after overnight fasting between 8 am and 9 am. We used 3 mL vacuum tubes with anticoagulant (ethylenediaminetetraacetic acid) for complete blood cell count analysis. Complete blood cell count parameters (hemoglobin, red blood cell [RBC] count, white blood cell [WBC] count, and differential WBC count) were carried out using a hematologic analyzer based on volume conductivity light scatter and flow cytometry method on the same day. We collected the remaining blood in blood collection tubes without anticoagulants to separate the serum. The separated serum was allocated in different vials and stored at −80°C. The following biochemical tests were done on the stored serum. Determination of malondialdehyde (MDA) was carried out using the thiobarbituric acid (TBA) reaction method.\(^{21}\) Superoxide dismutase (SOD) was measured using Beauchamp and Fridovich method.\(^{22}\) Vitamin C was measured using the oxidation method, and Vitamin E was measured using Emmerie-Engel method.\(^{23}\) Circulating concentrations of ferritin and cortisol were measured using Electrochemiluminescence immunoassay (Elecsys® Ferritin, and Elecsys® Cortisol, Roche Diagnostics International Ltd, Switzerland).
was measured using the immunoturbidimetric method (CRP Latex kit, Beckman Coulter, Inc., USA).

The same was repeated after 4 weeks of the intervention period in both the groups.

**Intervention and follow-up**

Participants were introduced to their respective interventions by the Program Increment (PI) during their meeting in the Department of Physiology. The heartfulness-based android app (HeartsApp) which is freely available on Google Play was installed on the subject’s phone. The app had instructions for both meditation and relaxation. The details of the practice are given in Supplementary File. After the introduction, participants were expected to follow their respective practice for 30 min daily preferably in the morning for 30 days at home with at least twice per week in person/live session (video call)/over the phone. Participants were expected to maintain a diary of their practice; only those who had practiced the meditation/relaxation for at least 25 days were considered for analysis. Participants who did attend in-person/live sessions were immediately contacted by PI to ensure the practice. The practice dairy was reviewed weekly and queries regarding practice were cleared by the PI. Participants were instructed not to practice any other form of adjuvant therapies without informing the PI. Of the 50 participants, 6 participants were not included in the final analysis (3 from the control group and 3 from the study group). Two participants from the control group adapted other adjuvant therapies. Three participants from the study group and one from the control group were not able to do the practices for a minimum of 25 days. There was no study-related injury or adverse events.

**Statistical analysis**

The normality of the data was assessed using Kolmogorov–Smirnov tests. We expressed data in mean ± standard deviation for normally distributed data and median (interquartile range) for nonnormally distributed data. The mean difference between the two groups was tested using the Student’s $t$-test or Mann–Whitney $U$-test based on data distribution both before and after the intervention period. Effect of intervention was analyzed using paired Student’s $t$-test for dependent samples test or Wilcoxon signed-rank test based on data distribution.
We used the Statistical Package for the Social Sciences version 19.0 for Windows (SPSS, Chicago, IL, USA). \( P < .050 \) was set as statistically significant.

**Results**

Age was comparable between the groups (control \(- 38.14 \pm 6.92; \) study group \(- 39.59 \pm 8.22; P = 0.509\)). Gender distribution was comparable between the groups (control group: M \(- 19, F = 3; \) study group: M \(- 20, F = 2\)). The duration of stay was comparable between the groups with a median of 15 days in both the groups.

Table 1 shows the comparison of SpO2, BMI, and basal cardiovascular parameters between the groups and the effect of the intervention on these parameters. Groups were comparable based on SpO2, BMI, HR, systolic blood pressure (SBP), and diastolic blood pressure (DBP) before intervention. SpO2 increased significantly in both the groups after the intervention period, and there was no difference between the groups after the intervention period. There was no significant change in BMI after the intervention period in both the groups. HR, SBP, and DBP decreased significantly after intervention in both the groups. The decrease in HR was more in the study group as compared to the control group.

Table 2 shows the comparison of complete blood count parameters between the groups and the effect of the intervention on these parameters. Before the intervention, groups were comparable based on hemoglobin levels, RBC count, WBC count, and differential leukocyte count. We observed a nonsignificant increase in hemoglobin levels in the study group after intervention. RBC count increased significantly in the study group after intervention. WBC count decreased significantly in both the groups after the intervention period, and the decrease was slightly more in the study group based on effect size (Cohen’s d control group \(- 1.22, \) study group \(- 1.42\)).

Table 3 shows the comparison of perceived stress and sleep quality parameters and the effect of the intervention on these parameters. Before the intervention, groups were comparable based on perceived stress and sleep quality. Perceived stress decreased significantly in both the groups after the intervention period; however, the decrease was significantly more in the study group. Sleep quality improved significantly in both the groups after the intervention period, and the improvement was comparable between the groups.

Table 4 shows the comparison of inflammatory markers, serum cortisol, oxidative stress parameters, and antioxidants. Before the intervention, groups were comparable based on ferritin, CRP, cortisol, MDA, SOD, Vitamin E, and Vitamin C. Ferritin, CRP, cortisol, and MDA decreased significantly in both the groups after the intervention period. SOD, Vitamin E, and Vitamin C increased significantly after the intervention period in both the groups. However, the decrease in CRP and MDA was significantly more in the study group. The decrease in ferritin and cortisol was more in the study group based on effect size estimation. The increase in SOD and Vitamin C was more in the study group based on effect size estimation.

| Table 1: Demographic profile and basic cardiovascular parameters |
|---------------------------------|---------------|---------------|---------------|
| Parameters                     | Group         | Mean±SD       | P (pre vs. post) |
| SpO2                           | Control       | 97.31±0.79    | <0.001         |
|                                | Study         | 97.79±1.2     | <0.001         |
|                                | Control versus study P | 0.124 | 0.522 |
|                                | Study         | 24.25±1.22    | 0.962          |
|                                | Control versus study P | 24.24±1.17 | 24.33±1.27 | 0.853 |
|                                | Study         | 0.890         | 0.238          |
| BMI                            | Control       | 24.25±1.22    | 0.962          |
|                                | Study         | 24.24±1.17    | 0.853          |
|                                | Control versus study P | 0.890 | 0.238 |
|                                | Study         | 82.18±7.96    | <0.001         |
|                                | Control versus study P | 82.49±7.58 | 82.70±7.62 | 0.002 |
| HR (beats per minute)          | Control       | 122.77±12.86  | 0.005          |
|                                | Study         | 119.32±11.87  | 0.005          |
|                                | Control versus study P | 0.432 | 0.229 |
|                                | Study         | 77.18±7.16    | 0.023          |
| SDBP (mmHg)                    | Control       | 83.68±8.82    | 0.001          |
|                                | Study         | 85.91±10.17   | 0.001          |
|                                | Control versus study P | 0.377 | 0.501 |
| DBP (mmHg)                     | Control       | 77.18±7.16    | 0.023          |
|                                | Study         | 75.77±6.58    | 0.001          |
|                                | Control versus study P | 0.377 | 0.501 |

Data are expressed in mean±SD. Statistical tests done: Comparison between control and study groups was done using unpaired Student’s \( t \)-test. Comparison of pre- and postintervention data was done using paired Student’s \( t \)-test. \( P<0.05 \) was considered statistically significant. BMI=Body mass index, HR=Heart rate, SBP=Systolic blood pressure, DBP=Diastolic blood pressure, SpO2=Oxygen saturation, SD=Standard deviation.
Table 2: Comparison of control and study groups – complete blood count

| Parameters                        | Group      | Pre       | Post      | P (pre vs. post) |
|----------------------------------|------------|-----------|-----------|-----------------|
| Hemoglobin (g/dL), mean±SD       | Control    | 13.93±2.11| 14.00±1.61| 0.831           |
|                                  | Study      | 13.57±1.96| 14.04±1.50| 0.083           |
|                                  | Control versus study | 0.562 | 0.923 |
| RBC (×10^6/µL), mean±SD          | Control    | 4.83±0.53 | 4.93±0.46 | 0.419           |
|                                  | Study      | 4.78±0.53 | 4.94±0.42 | 0.041           |
|                                  | Control versus study | 0.764 | 0.981 |
| WBC (×10^3/µL), mean±SD          | Control    | 9.52±3.95 | 6.02±0.98 | 0.001           |
|                                  | Study      | 9.89±3.79 | 5.96±0.99 | <0.001          |
|                                  | Control versus study | 0.754 | 0.849 |
| Neutrophils (%), median (IQR)    | Control    | 71.25 (12.5) | 63 (6.5) | <0.001          |
|                                  | Study      | 70.5 (10.25) | 59.5 (6) | <0.001          |
|                                  | Control versus study | 0.991 | 0.029 |
| Lymphocyte (%), median (IQR)     | Control    | 20.5 (13) | 28.5 (8) | <0.001          |
|                                  | Study      | 19 (10.75) | 30.5 (4) | <0.001          |
|                                  | Control versus study | 0.851 | 0.025 |
| Monocytes (%), median (IQR)      | Control    | 6.5 (4) | 6.5 (3) | 0.895           |
|                                  | Study      | 6 (2) | 6 (3.75) | 0.634           |
|                                  | Control versus study | 0.739 | 0.577 |
| Eosinophils (%), median (IQR)    | Control    | 2 (2) | 3 (1.75) | 0.008           |
|                                  | Study      | 2 (1.75) | 3 (1) | 0.023           |
|                                  | Control versus study | 0.659 | 0.788 |
| Basophils (%), median (IQR)      | Control    | 0 | 0 | 1 |
|                                  | Study      | 0 | 0 | 1 |
|                                  | Control versus study | 1 | 1 |

Data are expressed in mean±SD for normally distributed data and in median (IQR) for nonnormally distributed data. Statistical tests done: Comparison between control and study groups was done using unpaired Student’s t-test for normally distributed data and using Mann–Whitney U-test for nonnormally distributed data. Comparison of date pre- and postintervention was done using paired Student’s t-test for normally distributed data and using Wilcoxon signed-rank test for nonnormally distributed data. P<0.05 was considered statistically significant. RBC=Red blood cell, WBC=White blood count, IQR=Interquartile range, SD=Standard deviation

Discussion

The major finding in our study is that after 4 weeks of intervention, we observed a significant decrease in stress, circulating cortisol, inflammatory markers, and oxidative stress biomarker in both the groups. Further, we observed improved sleep quality and antioxidant biomarkers in both the groups. These beneficial alterations following intervention were high in the heartfulness meditation group as compared to the relaxation group.

Based on the India situation report 2022 by the WHO, there were 43,045,527 confirmed cases of COVID-19, and the psychiatric epidemic would concur with this pandemic. Stress and sleep disturbances seen in this pandemic increase oxidative stress and inflammation which perpetuates the morbidity of the disease even after the acute phase is overcome with standard treatments. Interactions between psychological stress and peripheral inflammation are important contributors to heart diseases. Various studies have documented the beneficial effects of meditation which include alleviating stress and improving sleep quality. Hence, the present study was conceived to study the effect of heartfulness meditation on the perceived stress, sleep quality, and inflammatory and oxidative parameters in patients who have been treated for COVID-19.

After 4 weeks of heartfulness meditation or relaxation programs, we did not observe any changes in BMI in either of the groups which could be due to short-term intervention.

The inability of a person to cope with emotional or physical challenges is called psychological stress (perceived stress) and systemic stress (cortisol), respectively. We observed that the study participants were in moderate perceived stress levels (as per PSS score). Psychological stress leads to systemic stress by activating the hypothalamic-pituitary-adrenal axis and releasing stress hormones such as cortisol; higher cortisol levels observed in stress are anti-inflammatory. Benson et al.’s “relaxation response” model is widely accepted to explain the anti-stress effect of mind–body medicine therapies. Heartfulness meditation and relaxation intervention were able to reduce the perceived stress levels from moderate to low. Cortisol levels also decreased significantly following both interventions. Quantitatively (effect size), the decrease was more in the heartfulness meditation group. Thus, meditation plays a role other than a relaxation response to bring down both psychological and systemic stress. Taylor et al. have proposed that increased bidirectional communication...
between the cerebral cortex, limbic system, and brain stem structures could regulate emotional and behavioral responses and optimize neuroendocrine regulations. Our study finding is in line with Goyal et al. who also reported reduced stress following meditation.

Kim et al. reviewed that psychological stress reduces slow-wave sleep, rapid eye movement sleep, and sleep efficiency. Increased cortisol levels are known to decrease slow-wave sleep. In our study, perceived stress and cortisol levels were high which could have reduced the sleep quality. Our participants experienced decreased sleep quality in terms of decreased sleep latency, decreased sleep duration, decreased sleep efficiency, use of sleep medication, and increased daytime dysfunction. We observed that sleep quality improved in both the groups following the intervention. The decrease in stress (psychological and systemic) may be one of the reasons for improved sleep quality observed in both the groups. Moreover, stress is a well-known factor that has been documented to inhibit melatonin levels. Since mediation is known to balance melatonin levels, we expected heartfulness meditation can increase sleep quality better than relaxation. However, the effect of both interventions on sleep quality was comparable.

Both stress and sleep disturbances are shown to affect the autonomic nervous system in terms of increased sympathetic activity and decreased parasympathetic activity. A decrease in cardiac vagal tone is considered a physiological index of stress, and based on Thayer and Lane’s “neurovisceral integration model,” the central autonomic network regulates stress physiology. We observed a significant decrease in HR, SBP, and DBP after both interventions. This shows that both relaxation technique and heartfulness meditation decrease sympathetic tone and increase parasympathetic activity which could be due to decreased stress and improved sleep quality observed. However, the decrease in HR is more in the heartfulness meditation group indicating a higher parasympathetic tone at rest in this group.

Inflammation in COVID-19 plays a critical role in pathogenesis and morbidity. Inflammatory parameters (ferritin and CRP) were high in our participants even though anti-inflammatory cortisol levels were high.

| Parameters          | Group       | Median (IQR)        | P (pre vs. post) |
|---------------------|-------------|---------------------|------------------|
|                     | Pre         | Post                |                  |
| PSS                 | Control     | 20 (3.75)           | 11.5 (4.5)       | <0.001 |
|                     | Study       | 20 (5.5)            | 9.5 (3)          | <0.001 |
| Control versus study | P          | 0.768               | 0.021            |        |
| Subjective sleep quality | Control     | 2 (2)               | 0 (1)            | 0.001  |
|                     | Study       | 2 (1)               | 1 (2)            | 0.047  |
| Control versus study | P          | 0.262               | 0.110            |        |
| Sleep latency       | Control     | 1 (1)               | 1 (1)            | 0.946  |
|                     | Study       | 1 (2)               | 1 (0.75)         | 0.044  |
| Control versus study | P          | 0.442               | 0.289            |        |
| Sleep duration      | Control     | 1 (2)               | 1 (1.75)         | 0.787  |
|                     | Study       | 1 (1.75)            | 1 (1)            | 0.186  |
| Control versus study | P          | 0.575               | 0.705            |        |
| Habitual sleep efficiency | Control     | 1 (2)               | 1 (1.75)         | 0.601  |
|                     | Study       | 1 (2)               | 1 (1)            | 0.791  |
| Control versus study | P          | 0.871               | 0.421            |        |
| Sleep disturbances  | Control     | 1 (1.75)            | 1 (2)            | 0.444  |
|                     | Study       | 2 (2)               | 0 (1.75)         | 0.013  |
| Control versus study | P          | 0.302               | 0.184            |        |
| Use of sleep medication | Control     | 0.5 (1)             | 0                | 0.002  |
|                     | Study       | 1 (1.75)            | 0                | <0.001 |
| Control versus study | P          | 0.081               | 1                |        |
| Day time dysfunction | Control     | 2 (1)               | 1 (1.75)         | 0.009  |
|                     | Study       | 1 (2)               | 1 (2)            | 0.470  |
| Control versus study | P          | 0.231               | 0.564            |        |
| Global score        | Control     | 8.5 (3.75)          | 6 (2.75)         | <0.001 |
|                     | Study       | 9 (3)               | 6 (2.75)         | <0.001 |
| Control versus study | P          | 0.568               | 0.567            |        |

Data are expressed in median (IQR). Statistical tests done: Comparison between control and study groups was done using Mann–Whitney U-test. Comparison of date pre- and postintervention was done using Wilcoxon signed-rank test. P<0.05 was considered statistically significant. PSS=Perceived stress score, IQR=Interquartile range.
Table 4: Comparison between control and study groups – biochemical parameters

| Parameters                  | Group                  | Mean±SD             | P (pre vs. post) | Effect size (Cohen’s d) |
|-----------------------------|------------------------|---------------------|-----------------|------------------------|
|                             | Preintervention        | Postintervention    |                  |                        |
| Ferritin (ng/mL)            | Control                | 263.77±128.31       | 186.48±149.21   | 0.014                  | 0.56                  |
|                             | Study                  | 304.74±85.96        | 141.87±116.35   | <0.001                 | 1.59                  |
| Control versus study P      |                        | 0.220               | 0.275           |                        |                       |
| CRP (mg/L)                  | Control                | 25.00±9.56          | 11.36±3.79      | <0.001                 | 1.88                  |
|                             | Study                  | 29.82±11.51         | 8.45±2.91       | <0.001                 | 2.55                  |
| Control versus study P      |                        | 0.138               | 0.007           |                        |                       |
| Cortisol (nmol/L)           | Control                | 444.75±175.52       | 310.11±277.07   | 0.004                  | 0.58                  |
|                             | Study                  | 505.63±120.04       | 295.11±241.12   | <0.001                 | 1.11                  |
| Control versus study P      |                        | 0.187               | 0.849           |                        |                       |
| Malondialdehyde (nmol/mL)   | Control                | 8.47±2.09           | 5.75±0.99       | <0.001                 | 1.66                  |
|                             | Study                  | 8.67±1.60           | 3.98±2.22       | <0.001                 | 2.43                  |
| Control versus study P      |                        | 0.719               | 0.001           |                        |                       |
| Superoxide dismutase (U/mL) | Control                | 148.23±32.07        | 203.09±19.92    | <0.001                 | 2.06                  |
|                             | Study                  | 141.18±22.57        | 211.09±14.70    | <0.001                 | 3.67                  |
| Control versus study P      |                        | 0.404               | 0.137           |                        |                       |
| Vitamin E (µg/mL)           | Control                | 6.16±2.04           | 11.10±2.93      | <0.001                 | 1.96                  |
|                             | Study                  | 6.32±1.67           | 12.48±4.20      | <0.001                 | 1.92                  |
| Control versus study P      |                        | 0.769               | 0.216           |                        |                       |
| Vitamin C (mg/dL)           | Control                | 0.03±0.02           | 0.15±0.05       | <0.001                 | 2.94                  |
|                             | Study                  | 0.04±0.03           | 0.17±0.05       | <0.001                 | 3.22                  |
| Control versus study P      |                        | 0.118               | 0.319           |                        |                       |

Data are expressed in mean±SD. Statistical tests done: Comparison between control and study groups was done using unpaired Student’s t-test. Comparison of date pre- and postintervention was done using paired Student’s t-test. P<0.05 was considered statistically significant. CRP=C-reactive protein, SD=Standard deviation

This might be due to decreased sensitivity of cortisol receptors in response to continuously higher levels of cortisol observed in COVID-19. In the present study, inflammatory markers such as CRP and ferritin decreased significantly in both the heartfulness meditation and relaxation groups. Our findings corroborate with previous studies which has reported decreased level of IL-6 and CRP following meditation. Further our study findings are in line with Chang et al. and Bhasin et al. who reported inhibition of inflammation following relaxation practice. Stress, sleep disturbances, and autonomic dysfunction are known to increase inflammation. A decrease in inflammatory markers after the intervention period in both the groups might be due to decreased perceived stress and increased sleep quality observed in our study. However, the reduction of inflammatory markers was significantly more in the heartfulness meditation group compared to the relaxation group.

SARS-CoV-2 induces oxidative stress in endothelial cells by increasing circulating levels of angiotensin II by depleting angiotensin-converting enzyme 2. Before the intervention, we observed increased oxidative stress (MDA) and reduced antioxidant (SOD and Vitamin C) in both the groups. The decrease in antioxidants could be due to direct inhibition of antioxidant pathways by the SARS-CoV-2 or indirectly due to increased utilization to counterbalance increased oxidative stress. Following the intervention, oxidative stress parameters (MDA) were decreased, and antioxidant parameters (SOD, Vitamin E, and Vitamin C) were increased in both the groups. However, the effect was significantly more in the heartfulness meditation group. This increase in antioxidant parameters could be due to a decrease in viral load (SOD) and a decrease in utilization (Vitamin E and C) as there were no supplements taken during this period by the participants. Vitamin C plays a myriad of functions which helps the COVID-19-positive subjects such as scavenging of oxidative species, regeneration of Vitamin E, regulation of inflammatory mediators, T-cell gene regulation, phagocytosis, and signaling cascade modification; it also increases neutrophil motility to the site of infection. Hence, Vitamin C not only plays a role in wound healing, but also it enhances the antiviral properties of lung epithelial cells. During the recovery period, since the viral load is reducing, and the oxidative stress is coming down, the utilization of the Vitamin C is reduced, and this could be the reason for the increased levels of Vitamin C observed in our study after the intervention. The clinical correlation of a decrease in oxidative stress-induced tissue damage (lung) is the improvement in SpO2 observed in both the groups.

Infection with SARS-CoV-2 can trigger inflammation by hyperactivating immune cells, especially the monocyte/macrophage system which, in turn, results in neutrophil infiltration of the tissue (inflammation) leading to oxidative stress at tissue levels. We observed neutrophilia in our subjects, and the percentage of neutrophils decreased significantly in both the groups after
the intervention period. The primary reason is the reduction in the viral load and thereby decreased inflammation as expected in our subjects.

In addition to the increase in neutrophil count, Palladino noted an increase in total leucocyte count, decrease in lymphocyte, eosinophils, RBC count, and hemoglobin in COVID-19 participants in his narrative review. In our study, the total leucocyte count was higher in both the groups, while hemoglobin and RBC count were within normal limits. We observed a significant reduction in total leucocyte count in both the groups as expected as the viral load will be decreasing during this period. Recent evidence shows that SARS-CoV-2 can directly attack the RBC membrane and indirectly damage it by oxidative stress, thereby reducing the RBC count. Even though RBC count was normal, we observed a significant increase in RBC count and a nonsignificant increase in hemoglobin in the heartfulness meditation group indicating the possibility of RBC destruction in our study subjects (moderate COVID-19 subjects). This improvement might be due to a decrease in oxidative stress and an increase in antioxidants in the heartfulness meditation group which was higher than the relaxation group. Lymphopenia induced by viral attachment or by the immune-mediated inflammatory mediator in COVID-19 is associated with delayed recovery and the need for an intensive care unit. There was lymphopenia in both the groups. Following the intervention, we observed that lymphocyte percentage increased significantly in both the groups which could be due to the recovery process or intervention or a combination of both. This hypothesis of our study findings needs further exploration.

To summarize, mental health problems during this pandemic are mainly due to the perceived psychological stress and increased systemic stress as observed by higher cortisol, inflammatory, and oxidative stress parameters. Practicing heartfulness meditation or relaxation could decrease the perceived stress and systemic stress, thereby reducing the psychological burden of the disease. Further, practicing heartfulness meditation resulted in a higher decrease in inflammatory and oxidative stress markers and an increase in antioxidant status, thereby speeding up the recovery process in COVID-19 and thereby reducing the morbidity associated with the disease. Hence, we suggest that heartfulness meditation may be advised to subjects who are suffering from COVID-19 to improve the recovery process.

Limitations

First, we were able to collect data from a modest sample size of participants which may limit the generalizability of the study. Second, we did not do a subgroup analysis in each group based on gender. Our study throws light on beneficial aspects of heartfulness meditation as an adjuvant therapy, but further studies can be carried out replicating our protocol using long-term intervention in a large-scale population. The intervention was given through an app considering the COVID scenario using the same app for both meditation and relaxation and hence strict adherence to one form of intervention could not be ensured.

Conclusions

Our results suggest that app-based heartfulness meditation/relaxation can be used as a nonpharmacological adjuvant to hasten the recovery process in patients who have completed the COVID-19 treatment protocol. Beneficial effects in subjects practicing heartfulness meditation were more than that observed in subjects practicing relaxation.

Ethical clearance

The study was approved by the institutional Ethics Committee of… (Name of institute) (Approval No – AIIMS/MG//IEC2021-22/100) All India Institute of Medical Sciences, Mangalagiri, Andhra Pradesh, India.

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Conflicts of interest

There are no conflicts of interest.

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Supplementary File – Details of Intervention

Intervention details

Duration: Both the interventions were given for 4 weeks.

Link for the techniques: https://heartfulness.org/en/meditation-practice

HeartsApp: When the participants visited the Department of Physiology, heartfulness practice coordinator (PI) would explain the techniques of meditation and relaxation to the respective participants. A standardized heartfulness-based android app (HeartsApp) which is freely available on Google Play was then installed on the subject’s phone, and instructions to use the same were given. The app had instructions for both meditation and relaxation. Three orientation sessions would be given within the 1st 3 days which can be in-person/live session (video call).

Heartfulness meditation: In home, participants should practice the technique daily following the instructions from the app, which are as follows
- “Gently close your eyes and relax.
- Turn your attention inwards and take a moment to observe yourself.
- Make a gentle suggestion that the source of light already present in your heart is illuminating it from within and drawing you in.
- Do this in a gentle and natural way.
- Rather than trying to visualize the light, simply tune in to your hearts, and open to any experience that you may have.
- There is no need to concentrate.
- If you find your awareness drifting to other thoughts, gently come back to the idea of the light in your heart.
- Feel immersed in the light in your heart, and let yourself become absorbed.
- Remaining still and quiet, rest there for as long as you want, until you feel ready to come out.”

The participants were requested to follow the techniques (relaxation or meditation) for 30 minutes in the early morning hours. Participants should follow the same morning meditation technique with a heartfulness trainer for 30 min at least twice per week in person/live session (video call)/over the phone. If the participants did not turn up in the scheduled weekly sessions, a reminder mail/call would be done by the heartfulness coordinator

Relaxation technique: In this technique, subjects are asked to sit comfortably and gently focus their attention on each part of the body and relax it consciously starting from the foot to the head guided with the audio from the android app which is as follows:
- “Sit comfortably. Gently close your eyes.
- Wiggle your toes.
- Feel the energy entering into your toes from Mother Earth. See its relaxing effect.
- Let this energy move upward to your feet and ankles. Feel how it rejuvenates and relaxes this part of the body.
- Let this energy move upward to your lower legs. Feel the relaxing effect of this energy moving upward to your calf muscles … your knees … your upper legs … and the entire area touching the chair, including the back. Feel this energy relaxing the entire back.
- Let this energy move forward relaxing the abdominal muscles … the chest area … your shoulders. Here especially feel the melting effect, the de-tensioning effect, in your shoulders. Feel that they are melting away.
- Let this energy move to your arms, feeling its effect though your biceps … and elbows … your wrist area, your palms, fingers and fingertips. Let this energy move and rejuvenate the entire length of your arms. Let this energy ooze out through your fingertips.
- Pay attention to the neck muscles. Feel the energy relaxing all the neck muscles.
- Let this energy move upward, relaxing all the facial muscles … forehead … your eyes … lips … earlobes … the top of your head.
- Feel this energy now flowing in a very gentle way from the feet, rising slowly upward through the steps we just followed, to the crown of your head.
- If you feel like revisiting a stressed area of your body, you can pay extra attention there now until that part of the body is also fully relaxed.
- Then scan the whole system now from toe to top.
- Remain still and quiet, and slowly become absorbed in yourself.
- Remain absorbed for as long as you want, until you feel ready to come out.”

The participants were requested to follow the techniques (relaxation or meditation) for 30 minutes in the early morning hours. Participants should follow the same morning relaxation technique with a heartfulness trainer for 30 min at least
twice per week in person/live session (video call)/over the phone. If the participants did not turn up in the scheduled weekly sessions, a reminder mail/call would be done by the heartfulness coordinator.

**Follow-up:** Participants were given with practice follow-up sheets in which they will mention their daily practice and guided sessions for attendance. This would be reviewed weekly by the PI and any clarifications on the practice if required would be addressed. Only participants who have 80% attendance, i.e., 25 days out of 30 days, would be considered for data analysis. Participants were instructed not to practice any other form of adjuvant therapies without informing the PI.