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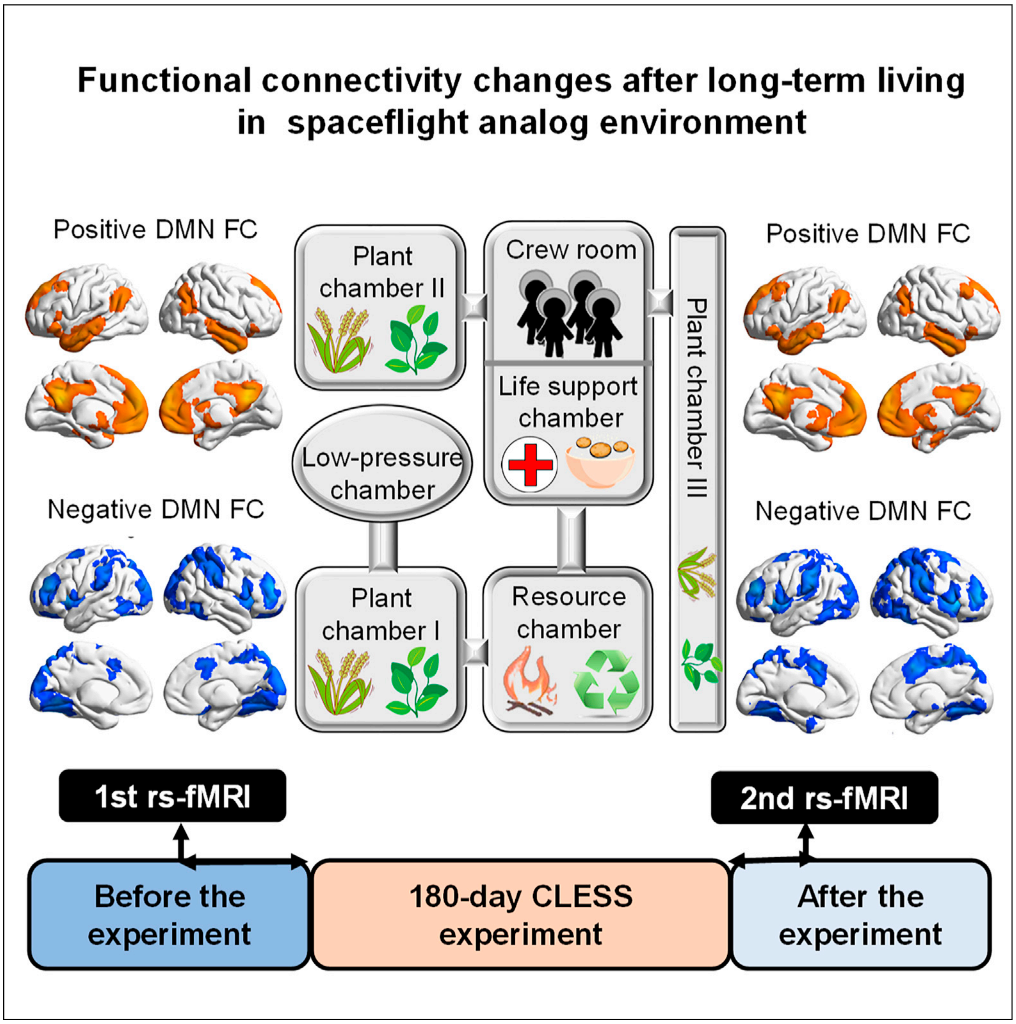


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## Highlights

Long-term living in closed spaceflight environment will lead to brain functional changes

FC of default mode network is significantly altered after CELSS experiment

Individual differences of FC changes are supported by evidence from behavioral analyses

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Article

# Effects of long-term closed and socially isolating spaceflight analog environment on default mode network connectivity as indicated by fMRI

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## SUMMARY

**Long-term manned spaceflight and extraterrestrial planet settlement become the focus of space powers. However, the potential influence of closed and socially isolating spaceflight on the brain function remains unclear. A 180-day controlled ecological life support system integrated experiment was conducted, establishing a spaceflight analog environment to explore the effect of long-term socially isolating living. Three crewmembers were enrolled and underwent resting-state fMRI scanning before and after the experiment. We performed both seed-based and network-based analyses to investigate the functional connectivity (FC) changes of the default mode network (DMN), considering its key role in multiple higher-order cognitive functions. Compared with normal controls, the leader of crewmembers exhibited significantly reduced within-DMN and between-DMN FC after the experiment, while two others exhibited opposite trends. Moreover, individual differences of FC changes were further supported by evidence from behavioral analyses. The findings may shed new light on the development of psychological protection for space exploration.**

## INTRODUCTION

Human beings have never stopped the pace of the exploration of space. During the progress, long-term manned spaceflight and the extraterrestrial planet settlement are no doubt the inevitable trends of space technologies and the focus of space powers.[1] In advancing the long-term manned spaceflight, establishing a controlled ecological life support system (CELSS) has been widely accepted to be the fundamental way to achieve the continuous supply of life support materials.[2] CELSS is an artificial closed ecosystem constructed for long-term space exploration. It has a significant material closure and circulation[3] and can provide enough breathable air, clean water, essential food, and safe working-living places for human beings (crewmembers). Various closed ecological system experiments have been conducted to date, including Biosphere 2 project,[4,5] the BIOS-3,[6] the Closed Ecological Experimental Facility,[7] and the 2-person-30-day CELSS in Beijing.[8] However, challenges still persist, one of which is the potential influence of closed spaceflight environment on the brain functioning of the crewmembers.

It has been observed that crewmembers tend to perform worse on measures of locomotor function and postural stability after a long-duration spaceflight mission compared to their pre-flight performance,[9–13] indicating that the long-term spaceflight can adversely affect sensorimotor performance. In addition, crewmembers have to spend all day working and living in a closed and socially isolating environment, which may affect mood and cognition. Previous studies have suggested that brain connectivity was reorganized within an environment of microgravity.[14–17] However, the potential influence of long-term closed and socially isolating environment on brain connectivity of the crewmembers remains unclear.

The default mode network (DMN) has been widely considered to directly support internal mentation that is largely detached from the external world.[18,19] Activation of the DMN has been consistently observed in the absence of tasks (i.e., during rest conditions), and the DMN is deactivated during the perception of the external world. More specifically, the DMN is active during passive rest and mind-wandering, which usually involves thinking about others or oneself, remembering the past, and envisioning the future.[20–22] In healthy individuals, positive functional connectivity (FC) was observed within the DMN, and negative FC was observed between the DMN and task-positive network, including the dorsal attention network.[23] The positive FC of the DMN was considered to reflect the functional homogeneity and facilitate

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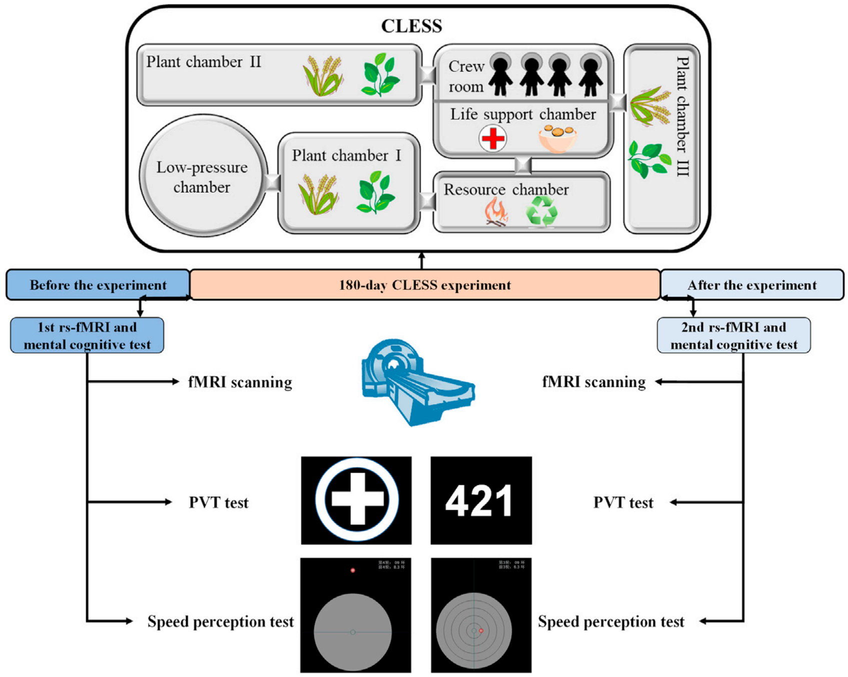


**Figure 1. Schematic of the 4-person-180-day CELSS integrated experiment**

The resting-state fMRI scans occurred at 25.1 ± 4.2 days before and 11.3 ± 7.5 days after the CELSS experiment, respectively.

the coordination and organization of information processing during rest,[24] while the negative FC of the DMN or the between-network anti-correlation was considered to reflect the effective capacity to switch between an extrospectively oriented and an introspectively oriented mode of function.[25,26] In our previous studies, a 7-day −6° head down tilt bed rest experiment was used to simulate microgravity, and enhanced FC within the DMN was observed, implying a self-adaption or compensatory enhancement to fulfill the complex demands of spatial navigation and motor control functions in a microgravity environment.[15] It is possible that long-duration living in a closed and socially isolating spaceflight environment may influence the functioning of the DMN.

In the current study, a 4-person-180-day CELSS integrated experiment was conducted,[27] and the crewmembers underwent resting-state functional magnetic resonance imaging (fMRI) scanning at two time points: the pre- and the post-experiment time points, respectively. We hypothesized that living in the closed and socially isolating environment for a long time would change the functioning of the human brain. Selecting the DMN as a network of interest, we examined within-network and between-network FC changes in the crewmembers using both seed-based and network-based analysis methods.

## RESULTS

The schematic of the 4-person-180-day CELSS integrated experiment and the detailed information of all participants are demonstrated in Figure 1 and Table 1, respectively. The positive and negative DMN masks in the standard Montreal Neurological Institute space are shown in Figure 2. Within these masks, the average positive DMN FC and negative DMN FC were calculated for each subject, respectively.

The individual DMN FC changes during the 180-day experiment are demonstrated in Figure 3A, and it can be clearly observed that the magnitude of the FC change in the three crewmembers is visually greater than those in the control group. Statistical analyses were performed with a modified t test procedure as Crawford and Howell recommended.[28] As shown in Figure 3B, the three crewmembers all exhibited significant changes in positive DMN FC (Crewmember#1: $t$-value = −4.49, $p < 0.001$; Crewmember#2: $t$-value = 1.88, $p = 0.04$; Crewmember#3: $t$-value = 2.34, $p = 0.02$), and two in negative DMN FC (Crewmember#1: $t$-value = 2.46, $p = 0.02$; Crewmember#2: $t$-value = −2.43, $p = 0.02$; Crewmember#3: $t$-value = −1.52, $p = 0.07$), compared with the mean level of normal controls. However, three crewmembers did not show the same trends in the change of average DMN FC. Crewmember#1, who was the leader of the crewmembers and worked at the China Astronaut Research and Training Center for many years, showed an opposite trend both in the positive and negative DMN FC compared with the two other crewmembers.

As illustrated in Figure 4, the three crewmembers showed the similar alterations of within-DMN and between-DMN connectivity strength as the seed-based analysis results (within-DMN connectivity strength change, Crewmember#1: $t$-value = −1.05, $p = 0.15$; Crewmember#2: $t$-value = 1.60, $p = 0.07$; Crewmember#3: $t$-value = 3.63, $p = 0.002$. Between-DMN connectivity strength change, Crewmember#1: $t$-value = 1.59, $p = 0.07$; Crewmember#2: $t$-value = −1.53, $p = 0.08$; Crewmember#3: $t$-value = −4.37, $p < 0.001$), and the Crewmember#1 also showed the opposite trend compared with the two others. These results were consistent with seed-based results.

Table 1. The characteristics of the participants in this study

| Subject | Sex (male/female) | Age (years) | Education (years) | The first fMRI scanning date (days before the CELSS experiment) | The second fMRI scanning date (days after the CELSS experiment) |
|---|---|---|---|---|---|
| Crewmember #01 | Male | 43 | 22 | 33 | 2 |
| Crewmember #02 | Male | 31 | 16 | 20 | 2 |
| Crewmember #03 | Male | 29 | 18 | 27 | 1 |
| Control #01 | Male | 34 | 22 | 32 | 10 |
| Control #02 | Male | 31 | 16 | 32 | 8 |
| Control #03 | Male | 31 | 18 | 28 | 8 |
| Control #04 | Male | 40 | 22 | 27 | 28 |
| Control #05 | Male | 42 | 22 | 26 | 10 |
| Control #06 | Male | 34 | 18 | 26 | 6 |
| Control #07 | Male | 46 | 22 | 23 | 27 |
| Control #08 | Male | 30 | 22 | 22 | 8 |
| Control #09 | Male | 31 | 22 | 22 | 6 |
| Control #10 | Male | 32 | 18 | 19 | 4 |
| Control #11 | Male | 28 | 18 | 19 | 12 |
| Control #12 | Male | 36 | 22 | 25 | 9 |
| Control_Group (mean ± SD) | Male | 34.6 ± 5.2 | 20.2 ± 2.2 | 25.1 ± 4.2 | 11.3 ± 7.5 |

In addition, evidence from the behavioral analysis further supported the results of resting-state fMRI. Specifically, in the psychomotor vigilance task (PVT) tests, the reaction times of Crewmember#2 and #3 were significantly higher after the closed experiment, while Crewmember#1 did not demonstrate the similar change (two-sample t test, Crewmember#1: $p = 0.66$, Crewmember#2: $p < 0.001$, Crewmember#3: $p = 0.003$) (Figure 5A). Moreover, in the speed perception tests, the Crewmember#1 was with a significant increase in error rate, while the other two crewmembers demonstrated the opposite alteration trend (two-sample t test, Crewmember#1: $p = 0.01$, Crewmember#2: $p = 0.12$, Crewmember#3: $p = 0.84$) (Figure 5B). In total, these results suggested that the CELSS experiment did not produce the same effect on all three crewmembers, where Crewmember#1 showed different behavioral changes compared with the other two.

## DISCUSSION

In this study, seed-based and network-based FC analyses were performed to examine the possible effect of closed and socially isolating spaceflight analog environment on the FC of the DMN, respectively. The results showed that at a voxel-wise level, the crewmembers exhibited significant changes both in the positive and negative DMN FC compared with the normal controls, which can be replicated in a network-based analysis manner. To the best of our knowledge, this is the first study to investigate the potential influence of long-duration closed and socially isolating living on spaceflight crewmembers' brain connectivity, providing new evidence for the development of psychological protection and personnel selection for space exploration.

It should be noted that the leader of the crewmembers, i.e., Crewmember#1, has suffered a significant reduction in both positive and negative DMN FC when completing the spaceflight analog experiment. In previous studies, the reduced DMN FC was observed in the subjects with cognitive impairments.[29–32] Accordingly, the current results indicated that Crewmember#1 might suffer from a mild cognitive decline after exposed in this socially isolating environment for 180 days. Actually, as the leader of the crewmembers and the eldest participant, Crewmember#1 has been trained for this experiment for a long time in advance, and the cognitive decline was more likely to occur due to the longest duration of closed training. The significant reduction of DMN FC was also observed in the real astronaut's brain after long-duration spaceflight,[33] which further demonstrated the reliability of our results.

Compared with Crewmember#1, the two other crewmembers did not show reduced FC of the DMN. Instead, an enhancement of positive and negative DMN FC was observed in these two crewmembers. This phenomenon might be a result of functional reorganization in response to early possible brain damage.[34] In specific, the reorganization of functional networks could be explained by the compensatory mechanisms. Before the onset of clinically apparent cognitive deterioration, the compensatory mechanisms play an important role in counteracting the slight decline in cognitive performance, in which the hyper-FC might be observed in the DMN.[35–37] Then, if the functional reorganization fails, functional networks will become disrupted, and the decompensation will occur with a significant FC reduction. In this study, therefore, the long-duration isolated living in the spaceflight analog might be the origin of the increase in both the DMN positive and negative FC, with the compensatory enhancement working of Crewmember#2 and Crewmember#3. Furthermore, the reduced DMN FC of Crewmember#1 was consistent with the manifestation of decompensation due to the excessive training received. It is possible that the period of

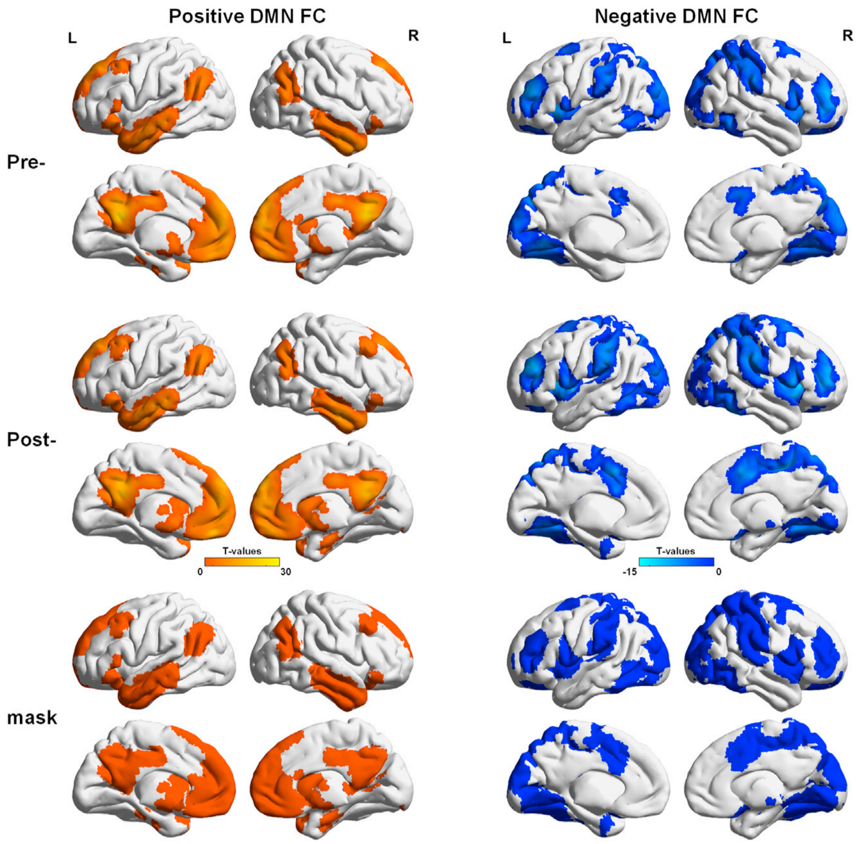


**Figure 2. Functional connectivity patterns of the DMN from all the subjects**

The regions with the orange color showed significant positive FC with the seeds of the DMN (left panel). The regions with the blue color showed significant negative FC with the seeds of the DMN (right panel). The conjunction mask included the voxels either in the maps of pre-experiment time point or in the maps of post-experiment time point. (One-sample t tests, $p < 0.05$, FDR corrected, cluster size $\geq 30$ voxels).

compensatory enhancement working of Crewmember#1 might occur before the experiment, which may be the reason that he exhibited the relatively high DMN FC strength compared with the rest of the subjects at baseline.

Because of the individual differences of FC changes, we hypothesized that the long-term closed and socially isolating living during the CELSS experiment might not produce an equal effect on all three crewmembers. Actually, this hypothesis could be partially supported by behavioral analysis results. In the PVT tests, both the Crewmember#2 and #3 had significantly increased reaction times after the experiment, indicating the reduction of personal alertness and watchfulness. However, similar change was not observed in Crewmember #1 who maintained the same reaction times as before the experiment, which could be explained by his extra vigilance-related training experience at the China Astronaut Research and Training Center. On the other hand, in the speed perception tests, only Crewmember#1 performed much worse after the experiment, which might be due to his functional decompensation and resulting early cognition impairment. Therefore, we speculate that long-term closed and socially isolating experiment affects three crewmembers differently, which may result from the differences of their work experience and task engagement.

## Limitations of the study

Some limitations in this study should be noted. Firstly, this experiment has a relatively small sample size, which makes it difficult to correlate the current findings with individual behavioral measurements. Therefore, future studies with large samples are needed. Secondly, the effects of other factors on FC changes were not taken into account, including circadian rhythm, noise, and indoor air quality. These factors could be investigated in future research. Finally, due to the lack of follow-up, it is unknown whether the crewmembers will recover after a certain period of adjustment.

## STAR★METHODS

Detailed methods are provided in the online version of this paper and include the following:

- KEY RESOURCES TABLE
- RESOURCE AVAILABILITY

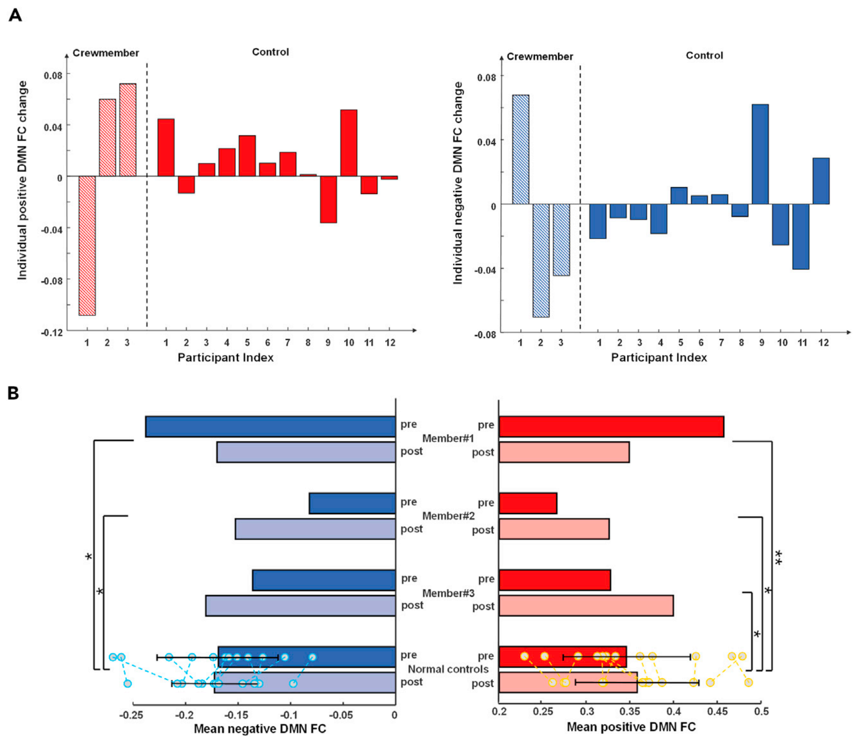


**Figure 3. The comparison results of seed-based functional connectivity analysis**

(A) The magnitude of the DMN FC change in three crewmembers was visually greater than in the control group.

(B) All of the three crewmembers exhibited significant changes both in positive DMN FC and in negative DMN FC, compared with the mean level of normal controls. The solid circles represented the FC values of normal controls. Positive FC change: Crewmember#1 (t(11) = −4.49, $p < 0.001$), Crewmember#2 (t(11) = 1.88, $p = 0.04$), and Crewmember#3 (t(11) = 2.34, $p = 0.02$); negative FC change: Crewmember#1 (t(11) = 2.46, $p = 0.02$), Crewmember#2 (t(11) = −2.43, $p = 0.02$), and Crewmember#3 (t(11) = −1.52, $p = 0.07$). (*$p < 0.05$; **$p < 0.005$). Error bars indicate variability (±1 standard deviation).



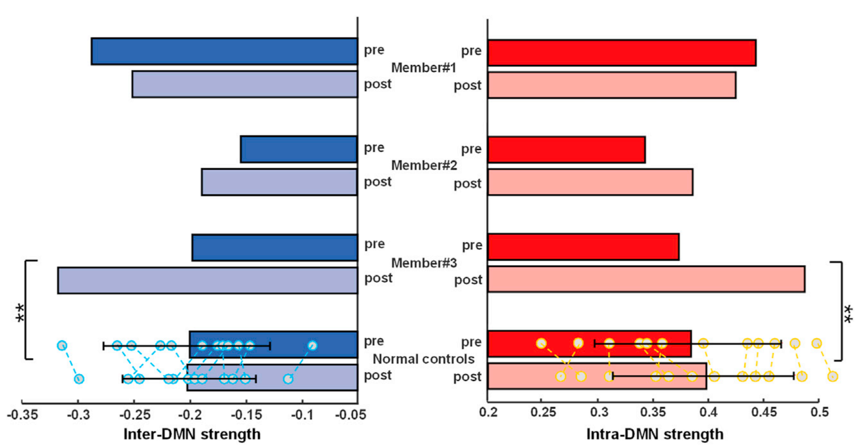


**Figure 4. The comparison results of network-based functional connectivity analysis**

The three crewmembers exhibited the similar changes with the seed-based analysis results both in the within- and between-DMN connectivity strength. The solid circles represented the connectivity strengths of normal controls. Within-DMN connectivity strength change: Crewmember#1 (t(11) = −1.05, $p = 0.15$), Crewmember#2 (t(11) = 1.60, $p = 0.07$), and Crewmember#3 (t(11) = 3.63, $p = 0.002$); between-DMN connectivity strength change: Crewmember#1 (t(11) = 1.59, $p = 0.07$), Crewmember#2 (t(11) = −1.53, $p = 0.08$), and Crewmember#3 ((11) = −4.59, $p < 0.0001$). (**$p < 0.005$). Error bars indicate variability (±1 standard deviation).

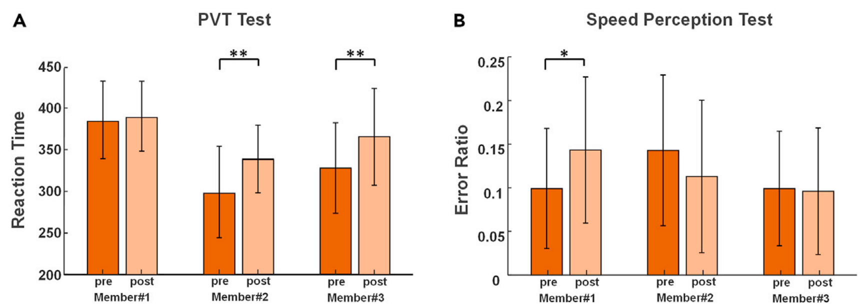


**Figure 5. The results of behavioral analyses**

(A) In the PVT test, the reaction times of three crewmembers in pre- and post-experiment time point, respectively.

(B) In speed perception test, the error ratios of three crewmembers in pre- and post-experiment time point, respectively. (Two-sample t test: *$p < 0.05$; **$p < 0.005$).

Error bars indicate variability ($\pm$1 standard deviation).

  - Subjects
  - Experiment design
- METHOD DETAILS
  - Mental cognitive tests
  - Resting-state fMRI acquisition and preprocessing
  - Seed-based functional connectivity analysis
  - Network-based functional connectivity analysis

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## AUTHOR CONTRIBUTIONS

Y.S., H.C., Z.X., and M.L. collected the data and wrote the manuscript. L.P. and L.-L.Z. performed the data analysis and wrote the manuscript. P.X., K.L., H.S., G.J., J.X., D.H., and Y.L. contributed to the discussion and manuscript revision. L.-L.Z., L.Q., and M.L. made contributions to the design of the experiment and revised the manuscript. L.-L.Z., L.Q., and M.L. are all the guarantors of this work and, as such, had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

## DECLARATION OF INTERESTS

The authors declare no competing interests.

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# STAR★METHODS

## KEY RESOURCES TABLE

| REAGENT or RESOURCE | SOURCE | IDENTIFIER |
|---|---|---|
| Software and algorithms | | |
| MATLAB | MathWorks | R2015-2022a |
| Statistical parametric mapping software package | http://www.fil.ion.ucl.ac.uk/spm | v8.0 |

## RESOURCE AVAILABILITY

### Lead contact

Further information and requests for resources should be directed to and will be fulfilled by the lead contact, Ling-Li Zeng (zengphd@nudt.edu.cn).

### Materials availability

This study did not generate new unique reagents.

### Data and code availability

- All data reported in this paper will be shared by the lead contact upon reasonable request.
- This paper does not report original code.
- Any additional information required to reanalyze the data reported in this paper is available from the lead contact upon reasonable request.

## EXPERIMENTAL MODEL AND STUDY PARTICIPANT DETAILS

### Subjects

Four crewmembers (three males and one female) participated in this study, with the ages from 29 to 43 years. The crewmembers were all selected through physiological and psychological evaluations, and the personality, cognitive ability and operational capacity were also tested and evaluated. All of the crewmembers had extensive educational experience and were trained for living in isolated and confined environments. The female crewmember was unable to undergo fMRI scanning due to the detection of a metal object in her body, so she was excluded from the study. Twelve normal controls matched for age and education were recruited for the fMRI comparison. The inclusion criteria for this study were as follows: (a) male, (b) age 28-45 years, (c) normal intelligence, (d) right-handed, (e) 20/20 vision, (f) ability to provide written informed consent and to comply with the study procedures, (g) no history of neuropsychiatric disorders, (h) abstaining from alcohol, coffee, and tobacco consumption for 24 hours before scanning, and (i) exclusion of subjects with magnetically active prosthetics, plates, pins, permanent retainers, or bullets for safety reasons. Detailed information of all the participants was provided in Table 1. This study was approved by the Institutional Review Board of Longgang Central Hospital of Shenzhen, China. All participants submitted written informed consent.

### Experiment design

The "4-person-180-day CELSS integrated experiment" took place in Shenzhen, China. The area of the CELSS integration chamber is 370 square meters, and the volume is 1340 cubic meters. As demonstrated in our previous study, it consists of 8 interconnected cabins:[38] three plant chambers, two crew rooms, one low-pressure plant chamber, one life support chamber, and one resource chamber. The plant chambers were mainly used for cultivating plants; the crew rooms were used for sleeping, medical examinations and daily work; the life support chamber provided food, oxygen and water for the entire crew; the resource chamber mainly recycled the waste goods and wastewater generated by the whole CELSS integration chamber, using plants and microorganisms as central recycling components. The crewmembers had their own professional duties. Crewmember#1 was the leader of the crew group, and was responsible for the overall daily arrangement, coordination, and task management. Crewmember#2 was in charge of cultivating and harvesting plants; Crewmember#3 was in charge of the equipment's operation and maintenance. Crewmember#4 (female) was responsible for the medical support and resource management. More details of the experiments can be seen in our previous studies.[27,38]

## METHOD DETAILS

### Mental cognitive tests

To investigate whether the change of brain functional organization would influence individual behavioral performance, we conducted the behavioral analysis by introducing two mental cognitive tests.

#### *Psychomotor vigilance task test*

The psychomotor vigilance task (PVT) test has been widely used to evaluate the ability of sustained and vigilant attention. In simulated and practical aerospace operations, PVT test is also a useful method for assessing the central fatigue of crewmembers and astronauts.[39–41] During the test, the participant was first instructed to focus on a crosshair displayed on a small screen. Then, a digital stimulus would appear on the screen after a random interstimulus interval ranging between 0.5 and 10 seconds. The participant was required to press a button as quickly as possible upon the appearance of the digital stimulus. The reaction time, measured from the button press, was displayed on the screen and automatically recorded. Each PVT test consisted of approximately 100 stimulus presentations and lasted for 10 minutes. In this study, a standard 10-minute PVT test was conducted individually for each crewmember before and after the CELSS experiment. A two-sample *t*-test was performed to assess the differences in the test results between these two time points.

#### *Speed perception test*

The speed perception test was conducted to evaluate an individual's ability for spatial-temporal integration. In practical aerospace operations, this capability is important for maintaining daily cabin life and achieving successful extra-vehicular activities. During the test, a dartboard with a radius of 200 mm was displayed at the center of the screen. Additionally, a red ball with a radius of 12 mm randomly appeared at the top, bottom, left, and right sides of the screen, with a distance of 330 mm from the screen center. The participant started the test by pressing a button, after which the ball would move towards the screen center with a specific velocity and continue moving if it passed through the center point. Once the ball moved into the dartboard area, it would become invisible, and the participant was instructed to judge whether the ball has reached the screen center and press the button accordingly. When the button was pressed, the ball would stop, and its position and the error rate would be recorded. After one second, a new trial would begin with the ball appearing randomly in another position. Each speed perception test consisted of 40 trials and lasted approximately three minutes. In this study, the speed perception test was conducted individually for each crewmember before and after the CLESS experiment, and a two-sample *t*-test was performed to evaluate the difference of test results between these two time points.

### Resting-state fMRI acquisition and preprocessing

The resting-state fMRI data were collected before and after the CELSS integrated experiment, respectively (see Figure 1). The three male crewmembers and 12 normal controls were instructed to keep eyes open, and keep conscious and still during the scanning, not thinking of anything in particular. The resting-state data were collected using a Siemens Prisma 3.0-T MRI scanner, and the T2*-weighted gradient-echo planar imaging parameters were as follows: repetition time/echo time = 2000/30 ms, slice thickness / gap = 4.0/0.4 mm, flip angle = 90°, field of view = 192×192 mm, matrix = 64×64, and number of slices = 37. For each subject, there were two resting-state runs at each time point with each lasting 6 min and 40 sec, and 200 volumes were obtained for each run.

The fMRI data were preprocessed using previously described procedures[42–44] with a statistical parametric mapping software package (SPM8, Welcome Department of Cognitive Neurology, Institute of Neurology, London, UK, http://www.fil.ion.ucl.ac.uk/spm). The first 10 volumes of each scan were discarded to avoid the magnetic saturation effect. Then, slice-timing and head motion correction were performed, in which the remaining images were realigned to the first volume within a run for the motion correction. It should be noted that all the subjects had less than 2 mm translation and 2° of rotation in any of the x-, y-, and z-axes. Next, the images were normalized to the standard EPI template in the Montreal Neurological Institute (MNI) space (3 mm isotropic voxels). The resulting images were spatially smoothed with a Gaussian filter kernel of 6 mm full-width half-maximum and temporally filtered with a Chebyshev bandpass filter (0.01-0.08 Hz). Finally, to reduce the spurious variance unlikely to reflect neuronal activity, the filtered data were regressed with the six head motion parameters and the mean signals from white matter (WM), the cerebrospinal fluid (CSF) and the whole brain, as well as their first-order derivative terms. The residuals of the regression were used for further analysis.

### Seed-based functional connectivity analysis

For each time point, the two runs of preprocessed images were timely-concatenated to obtain the whole fMRI data for each subject. To examine the effect of closed and socially isolating environment on the functional patterns of the DMN, a seed-based correlation analysis was performed. In this study, two seeds were defined as spheres with 5-mm radius around peak coordinates of the two main DMN nodes, i.e., medial prefrontal cortex (–1, 54, 27) and posterior cingulate cortex (0, –52, 27).[45] The time series of the voxels within each seed region were extracted and then averaged. For each individual, we obtained the functional correlation maps by calculating the Pearson's correlation coefficients between the averaged signal and the time series of each voxel in the entire brain. It should be noted that roughly averaging the time series from the voxels of spatially separated ROIs may cause possible problems in the FC calculation. However, we averaged the time series from the voxels of the posterior cingulate cortex and medial prefrontal cortex here because of the high temporal correlation between the two critical DMN nodes, as the previous studies.[25] Subsequently, Fisher's *r*-to-*z* transformation was applied to the resulting maps to improve the normality.

For each time point, one-sample *t*-tests ($P < 0.05$, FDR corrected, cluster size $\geq$ 30 voxels) were conducted first on the *z*-maps of all the subjects to identify the brain regions showing significant positive FC with the DMN and the regions showing significant negative FC with the DMN, respectively. By combining the binary spatial maps of the two time points together, a positive spatial mask and a negative spatial mask were then obtained, respectively. Within these two masks, averaged positive and negative DMN FC were calculated for each subject. Finally, separately for each crewmember, a modified *t*-test ($P < 0.05$) procedure[28,46,47] was conducted for comparing the FC change of a single case with the changes of a small control sample.

### Network-based functional connectivity analysis

To further understand the effect of spaceflight analog environment on the resting-state brain networks, a network-based analysis was supplemented. Twenty-four ROIs within the DMN and 14 ROIs within the dATN were involved in the analysis, with all of the ROIs defined by Yeo and his colleagues.[48] Similar case study was used to compare the alteration of within-DMN and between-DMN connectivity strengths of the three crewmembers with those of normal controls. In particular, the within-DMN strength was calculated by averaging the significantly positive FC between the ROIs within the DMN, and the between-DMN strength was calculated by averaging the significantly negative FC between the ROIs respectively in the DMN and dATN (one-sample t-test, $P < 0.05$, FDR corrected).