

Research Article

Influence of Enhanced Amino Acid Compositions on Human Cognitive Functions after COVID-19

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Abstract

This study aimed to demonstrate the effect of enhanced Amino Acid (AA) compositions on human cognitive functioning after Covid-19 damaging effect. The research study design included an open, randomized, placebo-controlled trial for 60 days. Participants in the study were measured initially, randomly divided into three groups: people who had Covid in the previous year, people who never had Covid, and placebo. Experimental and non-Covid groups used amino-acid compositions, while placebo group used sugar pills. All people were measured after 30 and 60 days. 70 healthy people aged 35-65, men and women, divided into three groups: 35 people in experimental Covid group, 15 people in non-covid group, and 20 people in placebo group. People in placebo group all had Covid. People in experimental covid and non-covid groups consumed daily specially prepared aminoacidic composition, charged with the radiation of a cold plasma. In the first test Covid groups demonstrated a statistically significant difference with a non-Covid group. After 30- and 60-days results of the experimental Covid group had no statistical difference with the results of non-Covid group, while results of both these groups demonstrated a statistically significant difference with the results of placebo group. The results showed that using AA compositions during the longitude period significantly affected cognitive functions in particular memory, speed of reactions and attention and allow to overcome negative effects of the Covid inflammation.

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Keywords: Cognitive functions; Cold plasma; Covid-19; Non-doping methods; Open; Placebo-controlled; Randomized

Introduction

On December 12, 2019, the first cases of the SARS-CoV-2 virus appeared in Wuhan, China. This virus is responsible for the development of Severe Respiratory Distress Syndrome (SARS) [1-3]. Human-to-human transmission of the virus occurs through direct or indirect routes. The direct route is associated with droplet transmission, while the indirect route is associated with contact with contaminated surfaces. The entry sites are the nose, mouth, and eyes [4-7].

Millions of people are still suffering the implications of the covid infection. The syndrome is known as post-acute sequelae of COVID-19 or PASC. In everyday practice it is referred to as long COVID. People with long COVID have symptoms as pain in different parts of the body. Difficulty with memory and decrease of cognitive functions, as well as extreme fatigue. The syndrome was estimated to affect about 16 million adults in the USA. Women and those at socio-economic disadvantage face higher risk. As do people who smoke and obese or have any of an array of health conditions. Particularly autoimmune disease [8,9]. The most common persistent and disabling effects of long COVID are neurological.

It is well known that amino acids (AAs) are cell signaling molecules and regulators of gene expression and the protein phosphorylation cascade. Additionally, AA is an essential precursor for synthesizing hormones and low-molecular-weight nitrogenous substances having enormous biological importance [10,11]. However, elevated levels of AA and its products are pathogenic factors for neurological disorders, oxidative stress, and cardiovascular disease. Thus, an optimal balance between AA in the diet and circulation is crucial for whole-body homeostasis.

A growing body of literature leads to a new concept of functional AA, defined as those AA regulating vital metabolic pathways to improve the health, survival, growth, development, lactation, and reproduction of organisms [12]. AA are essential for cognitive functions and may be used in clinical applications [13]. Dietary supplementation with one or a mixture of AA may benefit optimal health and well-being. Serotonin and dopamine amino acid precursors in the form of 5-HTP and l-tyrosine are highly effective treatments for depressed mood [14-16]. A study in [17] supported the use of oral supplementation of AAs for the cognitive function. Tryptophan is an AA that represents a key element for brain functioning, because of its role as a precursor for production of neurotransmitter serotonin (5-hydroxytryptamine), it has known cognition positive effects according to many studies [18-21]. The non-proteinic AA L-theanine in combination with caffeine showed to improve focus attention during a demanding cognitive task according to study [22]. Current findings in [23] suggested that ingestion of the seven essential AAs (granular powder, namely, leucine, phenylalanine, and lysine) led to improved attention and cognitive flexibility and psychosocial functioning, which is expected to prevent cognitive decline (Figure 1).

Plasma is a matter that exists in the form of ions and electrons. It is a gas that's been partially ionized. It is a mixture of neutral atoms, atomic ions, electrons, molecular ions, and molecules in excited and ground states. The number of particles in plasma depends on the gas pressure at which it was formed. The charges (positive and negative) balance each other; therefore, a good number of these charges are electrically neutral.

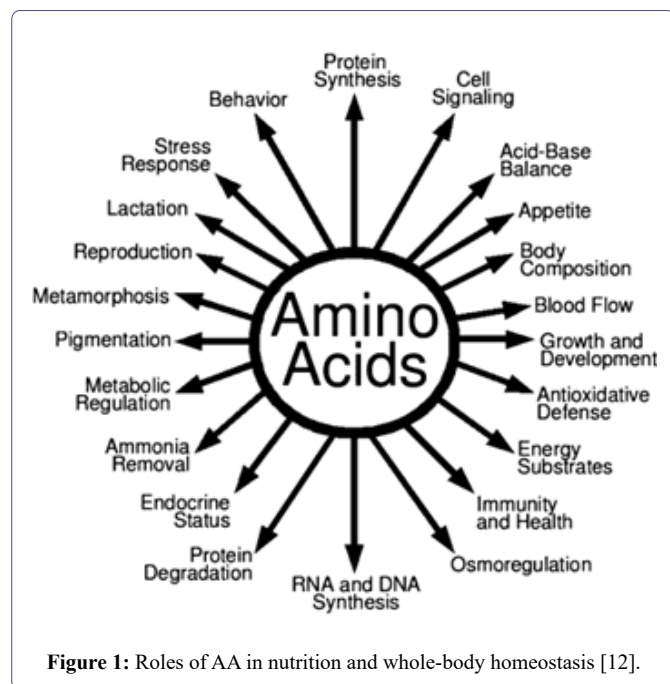


Figure 1: Roles of AA in nutrition and whole-body homeostasis [12].

Cold plasma has recently been widely used for food processing [24]. It is used to reduce the presence of microbiological contaminations [25]. Significantly extend the shelf-life of products [26], food drying performance [27], and even retain or potentially improve the organoleptic and biomolecular properties of food after processing [28].

In this paper, we present results of testing the influence of AA compositions on human cognitive functions and the improved effects of AAs processed by the radiation of a cold plasma over non-processed AAs.

Materials and Methods

An open, randomized, placebo-controlled trial for 60 days was conducted. Participants were measured initially, randomly divided into three groups (people who had Covid in the previous year, people who never had Covid and placebo). Experimental and non-Covid groups used AA compositions, while placebo group used sugar pills. All people were measured after 30 and 60 days later the start of the study.

Seventy adults of different sexes, ages, and professions, aged 50 +/- 14 years, participated in the study. Each subject received information about the study before the study and signed the Informed Consent Form. Information obtained during the investigation that identifies the identity of the subjects was kept secret and may be disclosed only within limits established by the law. Protocol was approved by the North-Western Medical University, St. Petersburg, Russia ethics committee. The study included adults meeting the following criteria:

- an absence of severe chronic medical conditions
- good mental health
- the ability to comply with the procedures set out in the inquiry protocol

There was no early abandonment of the subjects from the experiment.

Participants were randomly selected into three groups:

- Group 1: 35 people in experimental Covid group
- Group 2: 15 people in non-covid group
- Group 3: 20 people for 60 days consumed daily placebo

People in experimental covid and non-covid groups consumed daily specially prepared AA compositions, charged with the radiation of a cold plasma. Initially and every month, participants had access to online computer tests that were designed to test cognitive functions. The results of the tests were evaluated and statistically processed in the SPSS pro-gram by the research team.

The amino acid composition had the following content:

- Citicoline 500 mg
- Alfa-GPC 20 mg
- N-Acetyl L-Tyrosine
- NADH 20 mg,
- Phosphatidylserine 100 mg
- CoQ10 100 mg

Capsule made of fermented tapioca starch and distilled water.

The ethics committee of the Federal State Budget Institution "Saint-Petersburg Scientific-Research Institute for Physical Culture," St. Petersburg, Russia, approved the study protocol. All participants signed an informed consent form, where a written and oral explanation of the research protocol was provided.

Statistical Analysis

Several cognitive computer tests were designed with the objective of measuring the cognitive functions of the participants. The main cognitive functions measured were: – memory, speed of reactions, and attention. The following online computer tests were designed for this study:

- **Schulte Tables test:** These were designed based on German psychiatrist and psychotherapist Walter Schulte that created the Schulte Table as a psycho-diagnostic exam to evaluate attention and brain reaction. Generally, it is a grid with randomly distributed numbers or letters [29]. A Schulte table with the numbers 1 to 25 not in order was used. Participants were required to perform a task as quickly as possible and without mistakes to find and mark with the mouse all the numbers in order from 1 to 25. The number of errors (attention) and time-on task (speed of reactions) were collected as the main metrics for this test
- **Short-Term Memory test:** multi-digit numbers were presented on the screen. The task was to memorize this number and enter it into the input field. After entering, the following number was

introduced, one digit longer than the previous one, which should also be re-membered and entered, and so on. The number of right answers was the main metrics collected for this test after ten repetitions. Short-term memory could be defined as the memory mechanism that allows us to retain a certain amount of information over a short period of time. Short-term memory temporarily retains processed information that either fades quickly or turns into long-term memory [30]

- **Thorndike’s test:** The Thorndike test, used by psychologists to measure attention selectivity or focus on one thing at a time, is a table with 100 three-digit numbers in random order in 10 by 10 cells and ten random numbers that the subject must find and highlight as quickly as possible. The subject should focus as much as possible on the Thorndike exam to get an objective assessment of his attention [31]. A table of 100 three-digit numbers was presented. At the top, in front of the table, there were ten three-digit numbers, which one must find and mark on the table with the mouse as quickly as possible. The total amount of missing numbers in the correction (attention) and time-on task test (speed of reactions) were the main metrics collected
- **Correction test with numbers:** A table of a random set of 1080 numbers contained 36 lines of 30 characters each and a digit at the top. The task was to look through the table as quickly as possible and find and mark with the mouse the number indicated at the top. The total amount of missing numbers in the correction (attention) and time-on task test (speed of reactions) were the main metrics collected

Two set of hypotheses are considered for this study. The first set of hypotheses (H1 to H9) consist of probing that AAs have a positive effect in cognitive functions: memory, speed of reactions, and attention. The second set hypotheses (H10-H12) establish that difference on AAs effect on cognitive functions for Covid and non-Covid infected people. The complete set of hypotheses below:

- H1: There is no difference between 0- and 30-days AAs intake effect for the memory function H2: There is no difference between 0 and 60-days AAs intake effect for the memory function H3: There is no difference between 30- and 60-days AAs intake effect for the memory function H4: There is no difference between 0- and 30-days AAs intake effect for the speed of reactions function H5: There is no difference between 0 and 60-days AAs intake effect for the speed of reactions function
- H6: There is no difference between 30- and 60-days AAs intake effect for the speed of reactions function
- H7: There is no difference between 0- and 30-days AAs intake effect for the attention for COVID infected people
- H8: There is no difference between 0 and 60-days AAs intake effect for the attention function
- H9: There is no difference between 30- and 60-days AAs intake effect for the attention function
- H10: There is no difference between Covid and non-Covid infected people for AAs intake effect for the memory function
- H11: There is no difference between Covid and non-Covid infected people for AAs intake effect for the speed of reactions function

- H12: There is no difference between Covid and non-Covid infected people for AAs intake effect for the attention function

The first set of hypotheses (H1 to H9) are tested by using a t-student test for a difference of the mean values over the 4 periods of time (0, 30 and 60 days) in the study (T test 1-2, (0-30) T test 1-3 (0-60), T test 2-3 (30-60). For the second set of hypotheses (H10 to H12), the mean values of the AAs results are compared to check if there is a difference of effect between Covid and non-Covid infected people in the cognition functions with the use of AAs and tested by using the t-Student test (T-test Covid-non Covid). All the tests were performed with an alpha of 5%. If a p-value reported from a t test is less than 0.05, then that result is said to be statistically significant. If a p-value is greater than 0.05, then the result is insignificant.

Results

The initial test results and after every month are presented in tables. Table 1 shows the mean total time (minutes) on task-time for the Schulte Tables test, Short-Term Memory test and Correction test with numbers that measure the speed of reactions of the brain for the 3 groups in the study (Covid, non-Covid and placebo). The table also contains the p values for the t-student test for a difference of the mean values over the 3 periods of time (0, 30 and 90 days) in the study (T test 1-2 p value, T test 1-3 p value, T test 2-3 p value).

	Initial Test (1)			After 1 month (2)			After 2 month (3)		
	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo
Average	39,54 ± 9,8	32,8 ± 7	39 ± 8,3	37 ± 9,1	32 ± 8,2	38 ± 8,4	34 ± 8,4	32 ± 9	38 ± 8,1
T-test 1-2	< 0.05	0.95	0.86						
T-test 1-3	< 0.05	0.87	0.87						
T-test 2-3	< 0.001	0.98	0.96						
T-test Covid-Non Covid	< 0.05			< 0.05			0.87		
%% difference 1-3	-14.0	-2.4	-2.6						

Table 1: Mean Total on task-time the Schulte Tables test, Short-Term Memory test and Correction test, minutes (Speed of reactions). Total Time. min

Table 2 shows the number of right answers for the short-term memory test for the 3 groups in the study. This metric measures the memory cognitive function.

	Initial Test (1)			After 1 month (2)			After 2 month (3)		
	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo
Average	24.8 ± 7.2	33.5 ± 6.4	31.1 ± 7.3	36.3 ± 6.8	37 ± 6.6	33.6 ± 7.4	38 ± 6.4	37.5 ± 7.1	34.0 ± 7.1

T-test 1-2	< 0.001	< 0.05	0.78						
T-test 1-3	< 0.001	< 0.05	0.67						
T-test 2-3	0.87	0.78	0.89						
T-test Covid-Non Covid	< 0.05			0.76			0.82		
%% difference 1-3	53.2	11.9	9.3						

Table 2: Right Answers in memory test (Short term memory).

Table 3 shows the number of errors in Shuttle tables tests for the 3 groups. The number of errors measure the attention cognitive function.

	Initial Test (1)			After 1 month (2)			After 2 month (3)		
	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo
Average	6.97 ± 0.2	2.38 ± 0.1	6.6 ± 0.3	2.2 ± 0.2	2.1 ± 0.1	6.4 ± 0.4	2.2 ± 0.2	1.9 ± 0.2	6.0 ± 0.4
T-test 1-2	< 0.001	0.78	0.87						
T-test 1-3	< 0.001	0.09	0.89						
T-test 2-3	0.87	0.87	0.89						
T-test Covid-Non Covid	< 0.001			0.76			0.82		
%% difference 1-3	-68.4	-20.0	-9.1						

Table 3: Number of errors in Schulte Tables (Attention).

Table 4 shows the response time of Thorndike's test (seconds) for the 3 groups. This metric measures the attention cognitive function.

	Initial Test (1)			After 1 month (2)			After 2 month (3)		
	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo
Average	30.90 ± 4.2	14.56 ± 3.4	16.0 ± 4.3	9.3 ± 3.8	6.60 ± 1.6	14 ± 2.4	2.25 ± 0.4	2.34 ± 1.1	12 ± 2.1
T-test 1-2	< 0.001	< 0.001	0.87						
T-test 1-3	< 0.001	< 0.001	0.07						
T-test 2-3	< 0.001	< 0.05	0.09						
T-test Covid-Non Covid	< 0.001			< 0.05			0.88		

%% difference 1-3	-92.7	-83.9	-25.0						
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Table 4: Number of missing numbers in Thorndike's test (Attention).

Table 5 shows the total amount of missing numbers in the correction test for the 3 groups. This metric measures the attention cognitive function.

	Initial Test (1)			After 1 month (2)			After 2 month (3)		
	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo	Covid	Non Covid	Placebo
Average	0.94 ± 0.2	1.00 ± 0.04	0.9 ± 0.3	1.5 ± 0.08	1.6 ± 0.06	1.0 ± 0.4	2.3 ± 0.04	2.4 ± 0.01	0.9 ± 0.1
T-test 1-2	< 0.05	0.99	0.87						
T-test 1-3	< 0.05	< 0.05	0.98						
T-test 2-3	< 0.05	< 0.05	0.88						
T-test Covid-Non Covid	0.87			0.88			0.86		
%% difference 1-3	144.7	140.0	-3.3						

Table 5: Attention (Correction test).

Discussion

According to table 6, the presented results show that an intake of AAs has a positive effect in the memory function for Covid and non-Covid participants, however, this positive effect reaches its threshold at 30 days. The same table presents that an intake of charged AAs has a positive effect in the speed of reactions function for Covid participants. This last suggests that AAs have a more potential positive effect for Covid infected users.

The Schulte tables show that that an intake of AAs has a positive effect in the attention function for Covid participants, but this positive effect reaches its threshold at 30 days of the first intake for non-Covid patients. However, Thorndike's test presents that an intake of charged AAs has a positive effect for Covid and non-Covid participations in the attention function and this effect is present during the 3 periods tested and similar results are presented for the correction test with exception that for non-Covid participants this positive effect is only presented after 30 days of use. The difference of the results for both tests might be attributed to the better performance of Thorndike's test to test attention [32].

While presented results demonstrate positive effect in cognitive functions for the intake of AAs for Covid and non-Covid participants, the Covid participants benefit the most from the intake of AAs. The placebo group results practically did not change. This testifies that regular intake of the amino acid combinations positively affected cognitive function – memory, speed of reactions, and attention.

Hypothesis	COVID	Non COVID	Placebo	Type of tests
H1	Reject	Reject	Accept	Short-Term Memory
H2	Reject	Reject	Accept	Short-Term Memory
H3	Accept	Accept	Accept	Short-Term Memory
H4	Reject	Accept	Accept	Schulte Tables, Short-Term Memory and Correction
H5	Reject	Accept	Accept	Schulte Tables, Short-Term Memory and Correction
H6	Reject	Accept	Accept	Schulte Tables, Short-Term Memory and Correction
H7	Reject	Accept	Accept	Schulte Tables
H8	Reject	Accept	Accept	Schulte Tables
H9	Accept	Accept	Accept	Schulte Tables
H7	Reject	Reject	Accept	Thorndike's
H8	Reject	Reject	Accept	Thorndike's
H9	Reject	Reject	Accept	Thorndike's
H7	Reject	Accept	Accept	Correction test
H8	Reject	Reject	Accept	Correction test
H9	Reject	Reject	Accept	Correction test

Table 6: H1-H9 testing for COVID, Non COVID and placebo AAs.

Table 7 shows a significant difference in positive benefits for Covid participants against non-Covid participants, but this difference is only visible for the first period (0-30 days). After this period, there is almost no difference for Covid and non-Covid participants although both groups still benefit from the intake after 30 days. This last can be attributed to the fact that most participants improve their Covid condition after 30 days.

Hypothesis	Results from test 1	Results from test 2	Results from test 3	Type of tests
H10	Reject	Accept	Accept	Short-Term Memory
H11	Reject	Accept	Accept	Schulte Tables, Short-Term Memory and Correction
H12	Reject	Accept	Accept	Schulte Tables
H12	Reject	Reject	Accept	Thorndike's
H12	Accept	Accept	Accept	Correction

Table 7: H10-H12 Testing for results from test 1, 2 and 3.

It was shown that Covid virus could linger in the brain for months [5]. The autopsy study of people who died from Covid found viral DNA in the brain, significantly influencing cognitive functions. Presented results confirm this conclusion: in the initial test results for both Covid experimental and placebo groups were typically worth than the results of non-Covid group. After one and two months of the aminoacidic composition results of the experimental group became like the results of the non-Covid group, while results of the placebo group remain on the same level.

At the same time, one wants to understand the reason for such an additional significant effect (see the Table 8, will be discussed in the next paper) of enhanced supplements on cognitive functions compared to intact supplements. There are hypotheses that plasma radiation can generate a particular type of non-electromagnetic field (or quasi- particles) - known as ignitons, related to tachyon or scalar field [34,35]. This field is associated with hypothetical quasi-particles - tachyons or ignitons, which have a size and speed comparable to neutrinos. Gerald Feinberg coined the term tachyon in 1967. He studied the kinematics of such particles according to special relativity. In his paper, he also introduced fields with imaginary mass (now referred to as tachyons) to understand the microphysical origin of such particles. Although in some theories, the mass of tachyons is regarded as imaginary, in some modern formulations, the mass is considered real, the formulas for the momentum and energy being redefined to this end. Moreover, since tachyons are constrained to the space-like portion of the energy-momentum graph, they could not slow down to subluminal (meaning slower-than-light) speeds. In 1985, Chodos proposed that neutrinos can have a tachyonic nature [36].

Cognitive test	Amino Acids enhanced in plasma (Igmi Cognition)	Amino Acids non-irradiated in plasma	Placebo
Overall memory	100% improvement	20% improvement	1% improvement
Short Term memory	28% improvement	10% improvement	1% improvement
Operational memory	25% improvement	14% improvement	8% improvement
Attention	51% improvement	0% improvement	9% improvement
Quality of performance	83% improvement	9% improvement	39% improvement

Table 8: Clinical tests results for 80 volunteers results for 30 days. Group taken enhanced in plasma Amino Acids (Igmi Cognition) significantly outperformed both placebo and unenhanced supplements.

Obviously, applying these ideas to the experiments considered in this paper is a purely hypothetical concept.

Limitation of the study

One limitation of this study is its lack of generalizability since it involved only one set of data. Many critics question the academic value of the case study method as they argue that the finding or results cannot be generalized [33,37]. The limitations of this study include the sample size since we are using convenience sampling, and this can compromise the accuracy of the results. Longer periods of time for the AAs intake would also be beneficial to find the thresholds for the positive effect for usage recommendation purposes.

Conclusion

The study suggests that regular intake of enhanced AA compositions during the longitude period significantly affected cognitive functions: memory, speed of reactions, and attention for both Covid and non-Covid infected people. However, it was found that *regular intake of AA* compositions during the longitude allow to overcome negative effects of the Covid inflammation.

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