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## MUSCLE ACTIVITY AND EMOTIONAL BEHAVIOR CHANGES IN RATS WITH CHRONIC BRAIN ISCHEMIA VIA COMPREHENSIVE PATHOGENETIC CORRECTION

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### Abstract

Cerebrovascular pathology has evolved from a medical concern to a social issue, with the efficacy of secondary neuroprotection remaining a pertinent question. While some investigations have been conducted to elucidate the effectiveness of a comprehensively grounded pharmacological correction scheme under conditions of experimentally induced chronic brain ischemia, further refinement is necessary to understand the behavioral effects of animals, dosages, routes of administration of pharmacological agents, and analysis of obtained data. The aim of this study was to assess the effectiveness of applying Semax and hopantenic acid in the comprehensive treatment of motor disorders and established neurological impairments in rats with experimentally induced chronic brain ischemia. We determined that rats with chronic brain ischemia exhibit muscular dysfunctions and experience pronounced disturbances in emotional behavior from the first day. It has been demonstrated that the separate and combined application of Semax and hopantenic acid contributes to the restoration of muscle activity, normalization of coordination, and emotional behavior in rats with chronic

brain ischemia. The most significant neuroprotective effect in rats with chronic brain ischemia was observed with the combined administration of Semax and hopantenic acid, starting from the 3<sup>rd</sup> day of the trial. The subsequent in the series of anti-ischemic effectiveness is the impact of Semax from the 3<sup>rd</sup> day of the experiment. The least pronounced neuroprotective effect was exhibited by hopantenic acid, starting from the 5<sup>th</sup> day of the experiment. We consider these findings to be an experimental foundation supporting the feasibility of clinically testing the effects of combined Semax and hopantenic acid administration, capable of restoring functional disorders caused by chronic brain ischemia. In conclusion, the observed effectiveness of the developed pathogenetically justified complex for the correction of post-ischemic muscle dysfunction and disorders indicates the development of an anti-ischemic effect, as well as the fundamental possibility of improving the treatment for patients with chronic brain ischemia through clinical intranasal administration of Semax, either alone or in combination with hopantenic acid. However, for a more comprehensive understanding of the temporal and dose-dependent effects of this complex pathogenetic correction scheme on other disorders and dysfunctions related to chronic brain ischemia and to formulate final conclusions, it is essential to conduct separate series of experimental studies.

**Key words: cerebrovascular pathology; chronic brain ischemia; muscle activity; emotional behaviour; Semax; hopantenic acid; pharmacological correction**

The prevalence and incidence of cerebrovascular pathology, particularly its chronic manifestations, continue to rise under current conditions. Medical statistics indicate that cerebrovascular pathology has transcended its status as a purely medical concern, becoming a significant social issue across most European countries and worldwide. It stands as a primary contributor to mortality, physical and occupational disabilities, and a decline in patients' quality of life [7, 9, 20]. Presently, the majority of individuals affected by cerebrovascular pathologies are represented by patients with chronic brain ischemia (CBI) [2, 20, 24].

Chronic brain ischemia is characterized by a disruption in regulatory activity within the brain and the body as a whole, resulting in impairment of numerous systems and organs. This includes sensory cortical projections such as vestibular, visual, cutaneous, and proprioceptive pathways, in addition to central control [2, 9, 21, 24, 27].

Efficient treatment for chronic cerebral bloodflow insufficiency is of utmost importance. While medical professionals' skills and the availability of diagnostic and therapeutic tools generally ensure the chance to save patients' lives in the majority of CBI cases [2, 20], the efficacy of secondary neuroprotection remains a pertinent concern. Although

rescuing the lives of CBI patients addresses the restoration of full motor function, emotional preservation, and modulation of various forms of behavior [2, 17, 20], inadequate understanding of the pathogenetic mechanisms underlying this intricate cerebrovascular collapse leads to suboptimal efforts in promoting recovery.

While we conducted certain studies on a pathogenetically justified pharmacotherapy scheme during experimental CBI reconstruction [6], refinement was required concerning the effects of pharmacological agents on animal behavior, dosing, administration routes, and data analysis. Thus, a series of meticulous trials were conducted to assess the efficacy of pharmacological correction for the muscular dysfunction inherent in CBI, coupled with associated emotional imbalances. These investigations involved determining temporal intervals and potential synergies among specific pharmacological agents. Pharmacological compounds were chosen as agents for secondary neuroprotection, targeting pathogenetic pathways initiated by ischemic neuronal damage [2, 4, 5, 15, 21].

**The aim of the work** is to evaluate the effectiveness of administering Semax and hopantenic acid in a comprehensive treatment approach for motor disorders and resultant neurological impairments in rats during experimental chronic brain ischemia.

### **Materials and Methods**

Experiments were conducted under controlled conditions on 55 male Wistar rats weighing 180-250 g and housed in a vivarium. Ethical considerations, as outlined by the 'General Ethical Principles in Animal Experiments' from the Fifth National Bioethics Congress (Kyiv, 2013), European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (Strasbourg, 1985), and State Expert Centre guidelines of the Ministry of Health of Ukraine on 'Preclinical Drug Development' (2001), were strictly adhered to throughout animal care, management, and manipulations.

The CBI model was induced by dissecting the skin, isolating, and bilaterally ligating the carotid arteries [13]. Animals were divided into distinct groups: Group 1 - Control (intact rats, only skin incision without carotid artery ligation, n=7); Group 2 - Experimental (rats with carotid artery ligation and CBI reconstruction, n=12); Group 3 - CBI rats treated with Semax (SEM; 0.1%, intranasally, 10 µl, n=12); Group 4 - CBI rats treated with hopantenic acid (HA; RIK-pharm Ltd, 100 mg/kg, intraperitoneally, n=12); Group 5 - CBI rats treated with combined SEM and HA (n=12).

Following carotid artery ligation, rats were observed for 7 days. Muscle activity was evaluated based on their ability to balance on two horizontally positioned rods using their front and hind limbs [22]. A motor performance test involved assessing their capacity to

remain on a horizontally rotating rotarod (25 mm diameter, 60 cm length, 6 sections with 5 discs) at a fixed rotation speed of 15 rpm for 120 seconds [17]. Coordination skills were evaluated using the 'grid test'.

Aggressive-defensive behavior intensity was gauged by analyzing animals' behavioral responses during routine handling, scored on a 6-point scale [16].

Emotional behavior was investigated using the radial arm maze test, positioned 1 meter above the floor [1]. Duration spent by rats in one maze arm (as a percentage of total time in all arms) and the number of entries into a specific arm (as a percentage of total entries) were determined. Entries into enclosed arm sections were also recorded as an indicator of nonspecific motor activity [18].

Statistical analysis employed parametric ANOVA and Newman-Keuls post-hoc test for pairwise comparisons, along with the non-parametric Kruskal-Wallis test. Statistical significance was set at  $p < 0.05$ .

## **Results**

### *1. Pharmacological Correction of Muscle Activity in Rats with CBI*

Twenty-four hours after inducing CBI, only 3 out of 12 rats were able to maintain their balance on two vertical rods, which was significantly lower compared to the control group ( $p < 0.01$ , Table 1). Similar trends were observed in the data obtained from rats in other experimental groups.

By the 3<sup>rd</sup> day of the pathological ischemic process, only 2 out of 9 rats were able to remain on two vertical rods without pharmacological correction, which was also significantly lower compared to the corresponding control observations ( $p < 0.01$ ). During this stage of the study, only CBI rats that received intranasal administration of SEM and combined SEM with HA (4 and 5 out of 10, respectively) demonstrated improved ability to maintain balance on two vertical rods, surpassing the corresponding indicator in those without pharmacological correction ( $p < 0.05$ ).

On the 5<sup>th</sup> day of the study, 7 out of 10 CBI rats that received combined SEM and HA were able to maintain balance on two vertical rods. This indicator significantly exceeded that of the group receiving HA alone ( $p < 0.05$ ). At this point in the investigation, CBI rats that received intranasal SEM (4 out of 9) demonstrated the highest test performance ( $p < 0.05$ ).

By the 7<sup>th</sup> day, rats' muscle activity, following separate and combined administration of the investigated compounds, had approached normal levels in the test of maintaining balance on two vertical rods ( $p < 0.05$ ).

Table 1

Impact of Separate and Combined Administration of Semax and Hopantenic Acid on  
Intensity of Muscle and Coordination Activity in Rats with CBI

Animal Groups	Absolute Values		
	Number of Rats Maintained Balance on Two Vertical Rods	Number of Rats Maintained Balance on a Rotating Rod	Number of Rats Maintained on an Elevated Grid
<i>1<sup>st</sup> day</i>			
1 Group – Control, n=7	7	7	7
2 Group – CBI, n=12	3**	1**	2**
3 Group – CBI + SEM, n=12	3**	2**	3**
4 Group - CBI + HA, n=12	2**	2**	2**
5 Group – CBI + SEM + HA, n=12	4**	3**	4**
<i>3<sup>rd</sup> day</i>			
1 Group – Control, n=7	7	7	7
2 Group – CBI, n=9	2**	1**	2**
3 Group – CBI + SEM, n=10	4*#	3***#	5*#
4 Group - CBI + HA, n=9	3**	2**	3**
5 Group – CBI + SEM + HA, n=10	5*#	5*#@	6#@
<i>5<sup>th</sup> day</i>			
1 Group – Control, n=7	7	7	7
2 Group – CBI, n=8	1**	1**	2**
3 Group – CBI + SEM, n=9	4#	3*#	5#
4 Group - CBI + HA, n=9	3***#	2**	4*
5 Group – CBI + SEM + HA, n=10	7###@	6###@	8#@
<i>7<sup>th</sup> day</i>			
1 Group – Control, n=7	7	7	7
2 Group – CBI, n=8	1**	2**	3**
3 Group – CBI + SEM, n=9	6###	5#	7#
4 Group - CBI + HA, n=9	5#	4*	5#
5 Group – CBI + SEM + HA, n=10	8###	7#	8#

*Designations.* Reduction in the number of rats in groups occurred due to their demise.

*Notes.* \* -  $p < 0.05$  and \*\* -  $p < 0.01$  – probable discrepancies in the investigated indicators compared to the data in the control group;

# -  $p < 0.05$  and ## -  $p < 0.01$  – probable discrepancies in the investigated indicators compared to the data in CBI rats without pharmacological correction;

@ -  $p < 0.05$  – probable discrepancies in the investigated indicators compared to the data in CBI rats treated with Semax and/or HA (Kruskal-Wallis test applied for all calculations)

On the 1st day after CBI induction, only 1-3 rats in all experimental groups were able to maintain their balance on a rotating rod, significantly worse compared to the corresponding indicator in the control observations ( $p < 0.05$  in all cases).

By the 3<sup>rd</sup> day of the study on the rotarod, 5 out of 10 CBI rats that received combined SEM and HA were able to maintain their balance, surpassing the corresponding data in those without pharmacological correction and those with separate HA administration ( $p < 0.05$  in all cases). Notably, on this day, 3 out of 10 CBI rats that received intranasal SEM were also able to maintain balance on the rotating rod – this indicator, although lower than the control, exceeded the corresponding indicator in the group without pharmacological correction ( $p < 0.05$ ).

On the 5th day of the study, a greater number of CBI rats were able to maintain balance due to SEM administration ( $p < 0.05$ ) and combined administration of SEM with HA ( $p < 0.01$ ), compared to those without pharmacological correction in the rotating rod test. The effectiveness of combined SEM and HA administration exceeded that of separate SEM and HA administration ( $p < 0.05$ ).

The rotating rod test on the 7th day revealed that CBI rats receiving injections of SEM, HA, or combined therapy were able to maintain balance ( $p < 0.05$ ).

The elevated grid test showed that only 2-4 rats in all experimental groups were capable of maintaining their balance on the 1<sup>st</sup> day, significantly lower compared to the corresponding control indicator ( $p < 0.01$  in all cases).

From the 3<sup>rd</sup> day onward, CBI rats with SEM and combined SEM and HA administration (3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> days of the study;  $p < 0.05$ ) and those receiving only HA (7<sup>th</sup> day;  $p < 0.05$ ) were able to maintain their balance. On the 7<sup>th</sup> day, the investigated indicator's value in all experimental groups of CBI rats was comparable to that in the control group ( $p < 0.05$ ).

## *2. Pharmacological Correction of Emotional Behaviour in Aggressive-Defensive Behavior Test in Rats with CBI*

Throughout the entire study, intact rats avoided researchers' attempts to handle them, displayed intense vocalization, and attempted to bite the approaching researcher's hand (Fig. 1). CBI rats with or without pharmacological treatment became immobilized on the 1<sup>st</sup> with significantly lower average indicators of their emotional behavior than the corresponding control ones ( $p < 0.001$  in all cases).

The investigated indicator in CBI rats that received intranasal SEM on the 3<sup>rd</sup> day of the study was  $1.1 \pm 0.2$  scores, which was 4.4 times lower than in control observations ( $p < 0.001$ ), but 2.2 times higher than the corresponding indicator in those without therapy ( $p < 0.05$ ). On the 3<sup>rd</sup> day, CBI rats receiving combined therapy attempted to escape when the researcher's hand approached. The average indicator of emotional behavior was less than the control ( $p < 0.001$ ), but it was 3.6 times higher compared to the group with no correction ( $p < 0.05$ ). Furthermore, this investigated indicator significantly exceeded such data in groups of CBI rats that received separate SEM and HA administrations ( $p < 0.05$  in all cases). On the 5<sup>th</sup> day of the study, the obtained factual data were comparable to those recorded on the 3<sup>rd</sup> one.

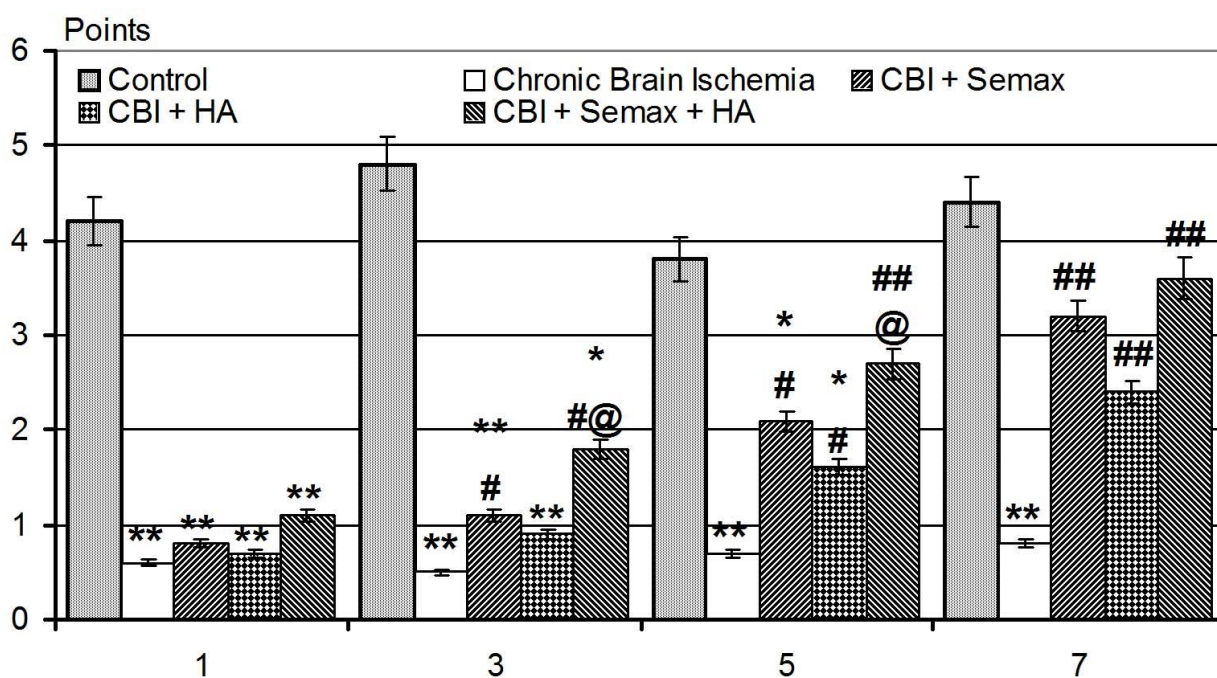


Fig. 1. Impact of Semax and Hopantenic Acid Separate and Combined Administration on the Emotional Behavior Expression in Rats with CBI

*Notes.* \*\* –  $p < 0,01$  – probable discrepancies of the investigated indicators compared to those in intact rats;

# –  $p < 0,05$  i ## -  $p < 0,01$  – probable discrepancies of the investigated indicators compared to those in rats with chronic brain ischemia without pharmacological correction (ANOVA + Newman-Keuls criterion applied in all calculations).

### *3. Pharmacological Correction of Emotional Behaviour in the Radial Arm Maze Test in Rats with CBI*

One day after CBI induction, significant differences were observed in all investigated indicators of rats' performance in radial arms – the number of entries into them and the duration of stay were 12 and 23 times lower, respectively ( $p < 0.001$ ; Table 2), compared to corresponding indicators in the control group. Rats did not move and enter enclosed sections of arms, but stayed in the center of the maze. The data obtained were similar to both the groups receiving CBI therapy with separate and combined SEM and HA administrations, as well as those without any pharmacological correction.

On the 3<sup>rd</sup> day, CBI rats treated with combined SEM and HA exhibited a 2.75-fold increase in the number of entries into the arms and a 2.8-fold increase in the duration of stay, in comparison to those without pharmacological correction ( $P < 0.05$ ). The observed indicators remained significantly lower than the corresponding control values ( $p < 0.001$ ) and surpassed the average values seen in groups of CBI rats treated with separate SEM and HA administrations ( $P < 0.05$ ). The number of entries into enclosed arm sections was uniformly lower across all experimental groups compared to the control group ( $p < 0.001$ ).

By the 5<sup>th</sup> day of the experiment, approximately 3-4 rats from the SEM and HA administration groups began to enter arms and remain there for distinct periods, resulting in a significant surge in the average entry indicators (91.7% and 75.0% respectively) and duration of stay (2.4 times and 2 times, respectively), as compared to the non-corrected group ( $p < 0.05$ ). The average number of arm entries for rats subjected to combined drug administration was  $5.6 \pm 0.5\%$ , with a corresponding stay duration of  $3.4 \pm 0.3\%$ , significantly exceeding the same parameters in groups receiving separate SEM and HA administrations ( $p < 0.05$ ). Similar trends were evident in the number of entries into enclosed arm sections.

Data presented in Table 2 highlighted parallel shifts in investigated emotional behavior indicators in the radial arm maze test on the 7<sup>th</sup> day of the experiment, signifying enhanced effectiveness of emotional behavior restoration during the post-ischemic period due to the combined SEM and HA administration.



Table 2

Impact of Semax and Hopantenic Acid Separate and Combined Administration on Emotional Behavior Intensity of CBI Rats in the 8-Radial Arm Maze Test

Animal Groups	Studied Indicators		
	Arms Entries, %	Time of Stay, %	Number of Entries into Enclosed Sections
<i>1<sup>st</sup> day</i>			
1 Group – Control, n=7	9.7±0.8	11.4±1.2	9.5±0.8
2 Group – CBI, n=12	0.8±0.1***	0.5±0.1***	0***
3 Group – CBI + SEM, n=12	1.1±0.1***	0.7±0.1***	0***
4 Group - CBI + HA, n=12	0.9±0.1***	0.5±0.1***	0***
5 Group – CBI + SEM + HA, n=12	1.3±0.1***	0.7±0.1***	0.4±0.1***
<i>3<sup>rd</sup> day</i>			
1 Group – Control, n=7	10.1±1.2	12.6±1.2	10.0±0.9
2 Group – CBI, n=9	0.8±0.1***	0.5±0.1***	0***
3 Group – CBI + SEM, n=10	1.2±0.1***	0.6±0.1***	0.4±0.1***
4 Group - CBI + HA, n=9	0.9±0.1***	0.7±0.1***	0***
5 Group – CBI + SEM + HA, n=10	2.2±0.3*** # @	1.4±0.1*** # @	0.7±0.1***
<i>5<sup>th</sup> day</i>			
1 Group – Control, n=7	9.6±0.8	13.1±1.3	10.0±0.9
2 Group – CBI, n=8	1.2±0.1***	0.8±0.1***	0.4±0.1***
3 Group – CBI + SEM, n=9	2.3±0.3***#	1.9±0.2***#	0.8±0.1***#
4 Group - CBI + HA, n=9	2.1±0.2***#	1.6±0.1***#	0.7±0.1***#
5 Group – CBI + SEM + HA, n=10	5.6±0.5** ## @	3.4±0.3*** ## @	1.5±0.1*** # @
<i>7<sup>th</sup> day</i>			
1 Group – Control, n=7	8.9±0.9	11.8±1.2	9.5±0.8
2 Group – CBI, n=8	1.9±0.2***	0.7±0.1***	0.4±0.1***
3 Group – CBI + SEM, n=9	4.7±0.4*##	2.4±0.3***##	1.7±0.2***##
4 Group - CBI + HA, n=9	3.6±0.4**#	1.9±0.2***##	1.6±0.2***##
5 Group – CBI + SEM + HA, n=10	8.1±0.6## @	5.3±0.4**## @	3.7±0.3***## @

*Designations.* Reduction in the number of rats in groups occurred due to their demise.

*Notes.* \* - p<0.05 and \*\* - p<0.01 – probable discrepancies in the investigated indicators compared to the data in the control group;

# - p<0.05 and ## - p<0.01 – probable discrepancies in the investigated indicators compared to the data in CBI rats without pharmacological correction;

@ - p<0.05 – probable discrepancies in the investigated indicators compared to the data in CBI rats treated with Semax and/or HA (Kruskal-Wallis test applied for all calculations)

## **Discussion**

In summary, the acquired data indicate that rats endure substantial muscular dysfunctions and emotional behavior disorders during the post-ischemic period, contingent upon its duration. The compromised functions were reinstated over the 7-day observation period, a process contingent upon the pharmacological agent type, route, and administration variant.

The data obtained in this study align, to some extent, with our prior findings on adynamia and hypodynamia in rats following bilateral ligation of the carotid arteries [6]. Consequently, during the course of cerebral ischemia, and its dynamics in both experimental and clinical settings, a marked reduction and/or derangement of motor and sensory functions [2, 13] transpire, in addition to the demise of certain biological entities, culminating in pronounced and clinically evident neurological deficits [19]. We posit that the analysis of the current data should also encompass the dynamics of horizontal and vertical motor activity in rats subjected to experimental ischemia.

We regard the reinstatement of studied muscle activity and emotional behavior, achieved via the combined administration of Semax and Hopantenic Acid, as a significant and pivotal outcome. The achieved restorative results became discernible from the 3rd day of the experiment and persisted through the observation period's culmination, corroborating data from prior studies [28].

Thus, we consider the combined administration of Semax and Hopantenic Acid the most efficacious anti-ischemic pharmacological intervention for muscle dysfunction and emotional disorders arising from experimental cerebral ischemia. Subsequently, intranasal Semax administration ranks next, with efficacy evident as early as the 3rd day in the majority of cases. HA administration ranks third in terms of successful anti-ischemic therapy, with observable impact commencing from the 5th day. Notably, a more pronounced efficacy of the SEM and HA combination was documented compared to their separate administrations. In this context, we provide empirical validation for the feasibility and clinical efficacy of intranasal drug delivery, a concept championed and substantiated for therapeutic purposes by Odessa neurologists [11]. In summary, restorative effects stemming from combined SEM and HA administration manifested earlier and significantly exceeded those resulting from separate administration of these two compounds.

We regard intranasal administration of therapeutic substances as a promising avenue for normalizing functions and ameliorating specific disorders induced by experimental cerebral ischemia. This route facilitates swift substance penetration into the brain, providing

elevated effective therapeutic concentrations and neuron contact in ischemic conditions, while circumventing the blood-brain barrier, whose traversal could otherwise compromise drug action speed and therapeutic efficacy [10, 12]. Within the framework of secondary neuroprotection, the swiftness of implementation and efficacy stand as paramount prerequisites for attaining a favorable therapeutic outcome. An analysis of the acquired data implies a correlation, in the majority of instances, between the anti-ischemic effects of intranasally administered Semax and combined Semax and HA therapy.

Comparable outcomes have previously been achieved under experimental conditions of ischemic and traumatic neuron injury [2, 14]. Intranasal administration of antiepileptic drugs, including those of peptide origin, suppressed both acute and chronic seizure activity [25], subsequently yielding an anti-ischemic effect [26]. With an emphasis on this, we posit that the acquired data offer supplementary experimental justification for the plausibility of clinically administering pharmacological agents with neuroprotective mechanisms of action via the intranasal route for cerebrovascular disorders.

The devised and validated pharmacological correction scheme for muscle dysfunction and emotional disorders is, in our estimation, pathogenetically sound, substantiated by the pharmacological profiles of the respective agents. Semax administration has demonstrated success in neuroprotective cerebrovascular disease therapy, complication prevention, and cognitive disorder management [4]. Owing to the peptide nature of Semax, it exhibits neuroprotective, neurometabolic, nootropic, and anti-asthenic effects [15], which impeccably align with the pathogenetic mechanisms governing neuronal injury and death in ischemic conditions [2, 21].

Hopantenic acid's efficacy has been corroborated in the treatment of combat veterans afflicted with chronic pain syndrome and post-traumatic stress disorder, contributing to memory and emotional state restoration, as well as anxiety reduction [8]. GABA's presence within the HA structure empowers it to engender anti-asthenic, stimulating, nootropic, and vegetotropic effects [3].

Consequently, our hypotheses have been substantiated, offering the prospect, as an integral facet of comprehensive pharmacotherapy for ischemic muscle dysfunction and emotional disorders, to apply a pathogenetic framework and achieve the reinstatement of motor and muscle activity (inclusive of coordination), along with emotional behavior, in rats with experimentally induced CBI. This, collectively, culminated in the realization of a restorative effect, achieved by stifling the ischemic pathological system's activity and activating regulatory antisystemic mechanisms that epitomize sanogenetic activity.

Though akin treatment modalities have been previously employed for cerebrovascular pathology [2, 5, 6, 14], our aim was to substantially augment the effectiveness of experimental correction for motor disturbances ensuing from CBI via intranasal drug administration.

Hence, the observed effectiveness of the developed pathogenetically validated complex for post-ischemic muscle dysfunction and disorder correction underscores the emergence of an anti-ischemic effect. Moreover, it underscores the fundamental potential to enhance the treatment of chronic brain ischemia in patients through clinical intranasal administration of Semax, either in isolation or in conjunction with hopantenic acid. However, in order to explore the temporal and dose-dependent ramifications of this multifaceted pathogenetic correction scheme for other dysfunctions and disorders associated with chronic brain ischemia and to establish definitive conclusions, we deem it imperative to conduct discrete series of experimental studies.

### **Conclusions**

1. Rats with a modeled chronic brain ischemia exhibited muscle dysfunction and pronounced disturbances in emotional behavior from the 1st day onward.
2. Separate and combined application of Semax and hopantenic acid contributed to the restoration of muscle activity, normalization of coordination, and emotional behavior in rats with chronic brain ischemia.
3. The most pronounced neuroprotective effect in rats with chronic brain ischemia was observed under conditions of combined administration of Semax and hopantenic acid, starting from the 3rd day of the experiment. Subsequently, the anti-ischemic effectiveness was evident with Semax administration from the 3rd day of the experiment. The least pronounced neuroprotective effect was exhibited by hopantenic acid, starting from the 5th day of the experiment.
4. The anti-ischemic efficacy of Semax was achieved through its intranasal administration, significantly enhancing its speed of action on neurons affected by ischemia.
5. The obtained data serves as an experimental basis for the feasibility of clinically testing the effects of combined administration of Semax and hopantenic acid, capable of restoring functional disorders caused by chronic brain ischemia. In this context, comprehensive pharmacological correction of behavioral manifestations in chronic brain ischemia is pathogenetically justified and aimed at activating sanogenetic mechanisms.

## References

1. Vastyanov R.S., Kopyova N.V. Peculiarities of the emotional behavior of rats in the dynamics of the development of pilocarpine-induced spontaneous seizures. Ukrainian medical almanac. 2009; 12(1): 28–33 [In Ukrainian].
2. Vastyanov R.S., Stoyanov A.N., Bakumenko I.K. Systemic pathological disintegration in chronic brain ischemia . Experimental-clinical aspects. - Saarbrucken : LAP Lambert Academic Publishing. 2015: 169 [In Russian].
3. Kanaeva L.S., Vazagaeva T.I., Yastrebova V.V., Shagiakhmetov F.Sh. Efficacy and tolerability of pantogam and pantogam active in patients with asthenic disorders. Clinical application and basic research. – M.: Triada-pharm. 2009: 104-120 [In Russian].
4. Kanunnikova N.P. Neuroprotective properties of neuropeptides. Journal of Grodno State Medical University. 2017; 5: 492-498 [In Russian].
5. Kaptalan A.O., Stoyanov O.M., Ostapenko I.O., Kirchev V.V. The effect of the combined use of Mexiprim and Semax on the behavioral correlates of post-traumatic epilepsy in rats. Actual problems of transport medicine. 2021; 1(63): 76-84 [In Ukrainian].
6. Kirchev V.V., Vastyanov R.S. Effect of semax and hopanthenic acid on locomotor activity and neurological deficit in rats under conditions of chronic brain ischemia. Bulletin of Marine Medicine. 2022; 2(95): 109-118 [In Ukrainian].
7. Muratova T.M., Khramtsov D.M., Stoyanov O.M., Babienko V.V., Vorokhta Yu.M. The influence of environmental factors on the risk of developing cerebrovascular pathology. Advances in biology and medicine. 2019; 2: 36-38 [In Ukrainian].
8. Paltsev A.I., Torgashov M.N., Voronova Yu.S., Bayandina E.V., Lunyaki on S.B. The role of combat stress in the formation of chronic pain syndrome in combatants and its treatment with pantogam active. Journal of Neurology and Psychiatry. 2010; 9: 43-46 [In Russian].
9. Sviridova N.K., Cherednychenko T.V. Diagnosis and treatment of cognitive disorders in comorbid patients with chronic brain ischemia . Medicines of Ukraine. 2020; 8(244): 50–53 [In Ukrainian].
10. Stoyanov A.N., Volokhova G.A., Vastyanov R.S., Prokopenko E.B. Possibilities of intranasal drug administration in CNS pathology. Bulletin of Physiotherapy and Balneology. 2009; 15(2): 37-39 [In Russian].
11. Stoyanov A.N., Vastyanov R.S., Burlya O.K., Bakumenko I.K., Drybina S.I. Intranasal peptidergic compounds administration in clinical neurovegetology. Journal of Health Sciences. 2014; 4(6): 157-170 [In Russian].

12. Stoyanov A.N., Vastyanov R.S., Volokhova G.A., Antonenko S.A., Burlya O.K., Bakumenko I.K., Oleinik S.M., Stoyanov A.A. Clinical and experimental possibilities of using peptidergic agents in neurovegetology. *Integrative Anthropology*. 2016; 1(27): 55-62 [In Russian].
13. Stoyanov A.N., Vastyanov R.S., Mashchenko S.S., Antonenko S.A., Skorobrekha V.Z. Possibilities of modeling cerebral ischemia in small animals. *International Journal of Neurology*. 2019; 6(108): 30-36 [In Russian].
14. Stoyanov O.M., Vastyanov R.S., Kolesnik O.O., Mashchenko S.S., Antonenko S.O. Experimental substantiation and dose-dependent effects of mexicor therapy for ischemic brain damage in rats. *Ukrainian Herald of Psychoneurology*. 2019; 27; 4(101): 25-31 [In Russian].
15. Sukhanov A.V. Experience in the use of the drug "Semax" in the treatment of cognitive impairment in young people who have undergone repeated closed craniocerebral trauma. *Astrakhan Medical Journal*. 2019; 14(1): 70-78 [In Russian].
16. Albert D.J., Richmond S.E. Septal hyperreactivity: a comparison of lesions within and adjacent to the septum. *Physiol. Behav.* 1975; 15: 339-347.
17. Christensen D., Gautron M., Guilbaud G., Kayser G. Combined systemic administration of the glycine/NMDA receptor antagonist, (+)-HA966 and morphine attenuates pain-related behaviour in a rat model of trigeminal neuropathic pain. *Pain*. 1999; 83: 433-440.
18. File S. The biological basis of anxiety. *Current Practices and Future Developments in the Pharmacology of Mental Disorders*. Y. Meltzer, D. Nerozzi (Eds.). Amsterdam : Excerpta Medica. 1991: 159-166.
19. Kalashnikov V.I., Stoyanov A.N., Pulyk O.R., Bakumenko I.K., Skorobrekha V.Z. Features of cerebrovascular reactivity in patients of young age with migraine. *Wiadomości Lekarskie*. 2020; 73(11): 2443-2446
20. Karger G. 30 years of Cerebrovascular Diseases. *Cerebrovasc Dis*. 2021; 50(1): 1. doi: 10.1159/000514372.
21. Moroz V.M. *Physiology* / V.M. Moroz, O.A. Shandra, R.S. Vastyanov, M.V. Yoltukhivsky, O.D. Omelchenko. Vinnytsia : Nova Knyha. 2016: 722.
22. Myslobodsky M., Kofman O., Mintz M. Convulsant specific architecture of postictal behavioral syndrome in the rat. *Epilepsia*. 1981; 27(4): 559–568.
23. Pellow S., Chopin P., File S. E., Briley M. Validation of open:closed arm entries in an elevated plus-maze as a measure of anxiety in the rat. *J. Neurosci. Meth.* 1985; 14: 149-167.

24. Sanchez-Bezanilla S., Hood R.J., Collins-Praino L.E., Turner R.J., Walker F.R., Nilsson M., Ong L.K. More than motor impairment: A spatiotemporal analysis of cognitive impairment and associated neuropathological changes following cortical photothrombotic stroke. *J Cereb Blood Flow Metab.* 2021; 41(9): 2439-2455.
25. Shandra A.A., Godlevskii L.S., Vastyanov R.S., Brusentsov A.I., Mikhaleva I.I., Prudchenko I.A., Zaporozhan V.N. Effect of intranigral dosage with delta sleep-inducing peptide and its analogs on movement and convulsive activity in rats. *Neurosci. Behav. Physiol.* 1996; 26(6): 567-571.
26. Shandra A.A., Godlevskii L.S., Brusentsov A.I., Vastyanov R.S., Karlyuga V.A., Dzygal A.F., Nikel B., Vastyanov R.S. Effects of delta-sleep-inducing peptide in cerebral ischemia in rats. *Neurosci. Behav. Physiol.* 1998; 28(4): 443-446.
27. Stoyanov O.M., Vastyanov R.S., Myronov O.O., Kalashnikov V.I., Babienco V.V., Hruzevskiy O.A., Turchin M.I. Vegetative system pathogenetic role in chronic brain ischemia, cerebral hemodynamics disorders and autonomous dysregulation. *World of medicine and biology.* 2022; 2(80): 162-168.
28. Vastyanov R.S., Kirchev V.V., Muratova T.M., Kashchenko O.A., Vastyanova O.V., Tatarko S.V., Zayats L.M. Comparative analysis of motor and emotional behavioral disorders in conditions of experimental chronic ischemic and chronic convulsive syndromes. *World of medicine and biology.* 2021; 2(76): 183-188.

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- conceptualization, methodology, formal analysis, data curation, writing—original draft preparation, writing—review and editing & supervision.

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**Informed Consent Statement**

The data of experimental studies are given. Written informed consent from the patients was not necessary to publish this paper.

**Data Availability Statement**

The data presented in this study are available on request from the author.

**Conflicts of Interest**

There is no conflict of interest.