Doctoral School in Medical Sciences

Manuscript title
U.D.C: 616.831-005.4-074/-076(043.2)

COSTRU-TAȘNIC Elena

STUDY OF POTENTIAL BIOMARKERS FOR PREDICTION OF HAEMORRHAGIC TRANSFORMATION OF CEREBRAL ISCHEMIC STROKE

321.05 – CLINICAL NEUROLOGY

Summary of the doctoral thesis in medical sciences

Chisinau, 2024

The thesis was elaborated at the Department of Neurology no. 1 of the Public Institution „Nicolae Testemitanu" State University of Medicine and Pharmacy and the Public Institution „Diomid Gherman" Institute of Neurology and Neurosurgery Institute.

Scientific adviser:
Gavriliuc Mihail,
Habilitated Doctor in medical sciences, university professor _____ signature

Members of the guidance group:
Lisnic Vitalie,
Habilitated Doctor in medical sciences, university professor _____ signature

Sangheli Marina,
PhD, associate professor _____ signature

Manole Elena,
PhD, associate professor _____ signature

The thesis defense will take place on September 11th, 2024, at 14:00, in the State University of Medicine and Pharmacy „Nicolae Testemitanu", 165, Stefan cel Mare si Sfant bd., Chisinau, MD-2004, 204 office, in the meeting of the Commission for public defense of the doctoral thesis, approved by the decision of the Scientific Council of the Consortium from 19.03.2024 (protocol Nr. 36).

Nominal structure of the Commission for public defense of the doctoral thesis:
Chair:
Groppa Stanislav,
Habilitated Doctor in medical sciences, university professor,
Academician of the MSA _____ signature

Members:
Gavriliuc Mihail,
Habilitated Doctor in medical sciences, university professor _____ signature

Zota Eremei,
PhD, associate professor _____ signature

Official reviewers:
Lisnic Vitalie,
Habilitated Doctor in medical sciences, university professor _____ signature

Odobescu Stela,
Habilitated Doctor in medical sciences, associate researcher _____ signature

Glavan Iulian,
PhD, associate professor _____ signature

Tuță Sorin,
PhD, associate professor,
Neurology discipline, University of Medicine and
Pharmacy „Carol Davila", Bucharest, Romania _____ signature

Author: Costru-Taşnic Elena _____ signature

CONTENT

INTRODUCTION

The research actuality. Cerebral ischemic stroke represents the second mortality cause and the third reason of disability worldwide [1,2]. In 2018, the stroke incidence in Republic of Moldova was 21.8 cases by 10.000 population, being the leading cause of disability among adults in our country [1,3].

The brain tissue sensibility to ischemia imposes rapid diagnosis and treatment approach, in the limited timeframe of the stroke therapeutic window. Haemorrhages, both systemic, but particularly the cerebral ones by haemorrhagic transformation of ischemic stroke (IS), are among the most important adverse reactions of the thrombolytic treatment [4,5].

Haemorrhagic transformation (HT) of IS represents the bleeding within the primary cerebral ischemic infarction territory, being a frequent complication among patients with ischemic stroke, and an incidence varying from 13 to 70% annually (according to radiologic and morphologic studies) [6].

Actual HT prediction scores in acute IS patients include mostly composed clinical and/or imaging parameters. Particular attention, according to literature data, is given to the markers of blood-brain barrier (BBB) integrity and functionality, the functional insufficiency of which proved to be crucial in the evolution towards haemorrhagic transformation of cerebral infarcts and which could serve as predictive factors of this complication.

Matrix metalloproteinases – MMPs, can degrade the basal membrane of the BBB and numerous fundamental studies emphasize the link between the increased expression of MMP-2 and MMP-9 in case of HT, with or without thrombolytic treatment [7]. Clinical validation of these studies could optimize the management of IS patients in the acute stage of the disease.

The aim of the research is the study of risk factors, clinical-biochemical correlations, and relevance of some clinical and paraclinical biomarkers in the prognosis of haemorrhagic transformation of acute ischemic stroke.

Objectives of the research:

1. Evaluation of clinical risk factors for haemorrhagic transformation of cerebral infarcts and calculation of disease severity scores: *NIHSS* (National Institutes of Health Stroke Scale), *SPAN-100* (Stroke Prognostication Using Age and NIHSS), *THRIVE* (Totalled Health Risks in Vascular Events), to highlight the persons with negative prognosis and increased risk for haemorrhagic transformation of ischemic stroke.
2. Establishing the role of the *ASPECTS* score (Alberta Stroke Program Early CT Scale) in the prognosis of haemorrhagic transformation of cerebral ischemic stroke, which would allow the early identification of haemorrhagic transformation cases.
3. Determination of the correlations between the MMP-2 and MMP-9 plasma levels and the risk of haemorrhagic transformation in ischemic stroke patients.
4. Assessment of the correlation between standard laboratory parameters determined in ischemic stroke patients (plasma level of fibrinogen, INR, total cholesterol, LDL-cholesterol, HDL-cholesterol, triglycerides, glucose, leukocytes, neutrophils, lymphocytes, platelets) and the risk of developing haemorrhagic transformation of cerebral infarction.
5. Evaluation of the ischemic stroke patient's functional independence, according to the *mRS* scale (modified Rankin Scale), at discharge and 3 months after the disease onset.

Scientific novelty of the research. A prospective clinical study was conducted with a comprehensive correlational analysis of multiple clinical and paraclinical risk factors (including imaging and laboratory - specific/nonspecific), as well as clinical severity assessment scores (clinical and imaging), in patients with acute cerebral infarction with the aim of establishing the relationship with the risk for haemorrhagic transformation (HT), as well as with the evolution of neurological recovery at discharge and 3 months after the ischemic cerebrovascular event. Biomarkers of blood-brain barrier integrity impairment (MMP-2, MMP-9) and their correlation with the post-stroke patient evolution (HT, unfavourable evolution) were studied.

Thus, the scientific problem solved in the study consists of highlighting the risk factors for HT in patients with acute ischemic stroke, which will allow the stratification of the risk for this complication, otherwise determining the optimization of therapeutic measures and post-stroke secondary prophylaxis. Clinical, imaging, and laboratory prognostic factors of patients with acute cerebral infarction were identified, with recommendations for in-hospital management and prevention.

The theoretical importance of the work consists in the evaluation of clinical, imaging, and biochemical characteristics in patients with acute ischemic stroke as risk factors for haemorrhagic transformation and unfavourable clinical evolution. The study provided additional information regarding the application of clinical, imaging, and laboratory biomarker scores in the prognosis of complications through HT in acute cerebral infarctions.

Applicative value of the thesis. The results of the study encourage the systematic application of clinical scores (NIHSS, SPAN-100, THRIVE) and imaging scores (ASPECTS), along with laboratory biomarkers of blood-brain barrier integrity, for stratifying patients with acute cerebral infarction at increased risk for unfavourable evolution, including haemorrhagic transformation. This will lead to more active monitoring of these patients and limit post-stroke complications.

Keywords: acute stroke, haemorrhagic transformation, risk factors, biomarkers, predictive scores, blood-brain barrier integrity, matrix metalloproteinases, prognosis of neurological recovery.

RESEARCH METHODOLOGY

The actual research represents an observational-analytic, prospective cohort study, with the main purpose to analyse ischemic stroke patients for the registration of haemorrhagic transformation of cerebral infarction cases.

The volume of the research cohort was calculated based on the formula for estimating observational-analytical research samples, cohort study type:

$$n = \frac{1}{(1-f)} \times \frac{2(Z_\alpha + Z_\beta)^2 xP(1-P)}{(P_o - P_1)^2}$$

where:

P_o = Proportion of patients with ischemic strokes with haemorrhagic transformation. According to the bibliographic data, the mean percentage of patients in whom this phenomenon is determined is 45,0% (P_0=0,45).

P_1 = Proportion of ischemic stroke patients with haemorrhagic transformation in the study group. We assume that their ratio will constitute 80.0% (P_1 =0,80).

$P = (P_0 + P_1)/2$=0,625

Zα – tabular value. When „α" – the significance threshold is 5%, then the coefficient Zα =1.96

Z_β – tabular value. When „*β*" – the statistical power of the comparison is 5%, then the coefficient Z_β = 1.65

f = Proportion of subjects expected to drop out of the study for various reasons q = 1/(1-f), f=10,0% (0,1).

Entering the data into the formula we obtained:

$$n = \frac{1}{(1-0.1)} \times \frac{2(1.96+1.65)^2 x0.625x0.375}{(0.45-0.8)^2} = 55.39$$

Two groups were designed for the research: the research group - L_A which required the inclusion of no less than 55 patients with cerebral infarctions with haemorrhagic transformation; and the control group - L_B which had to include no less than 55 patients with cerebral infarctions without haemorrhagic transformation.

Thus, at least 110 adult patients with acute ischemic stroke, in the super-acute period of the disease according to the research design, were to be included in the study (figure 1).

The inclusion criteria for the research:

1. Signing of the informed consent for participation in the research.
2. Adult age (greater than or equal to 18 years).
3. The history of the disease and the clinical manifestations suggestive for stroke, in the super-acute period of the disease (the first 24h from the onset of the first clinical signs).
4. The presence of early manifestations of cerebral infarction on the primary native computed tomography and/or the absence of signs of intracerebral bleeding.

Exclusion criteria from the research:

1. Lack of the informed consent to participate in the research.
2. Minors under the age of 18.
3. The presence of signs of intracerebral bleeding on the primary native computed tomography.

RESEARCH STUDY DESIGN

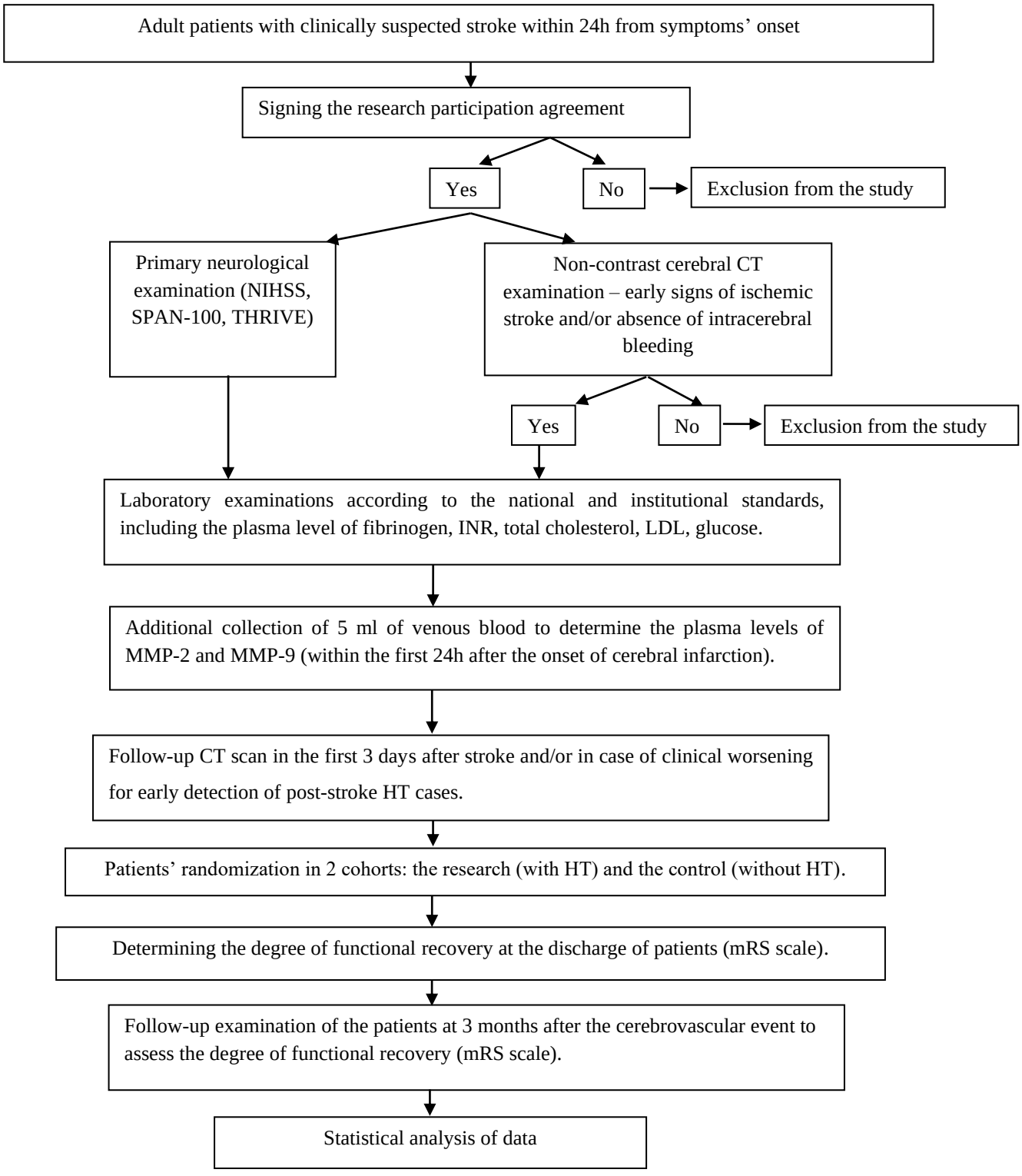

Figure 1. **The study design.**

Note: NIHSS – National Institute of Health Stroke Scale, SPAN-100 – Stroke Prognostication using Age and NIHSS, THRIVE – The Totaled Health Risks in Vascular Events, INR – International Normalised Ratio), LDL – Low Density Lipoproteins, MMP-2 – matrix metalloproteinase–2, MMP-9 – matrix metalloproteinase–2, CT – computed tomography, HT – haemorrhagic transformation, mRS scale – modified Rankin Scale.

SUMMARY OF THE CHAPTERS

CHAPTER 1 („HAEMORRHAGIC TRANSFORMATION OF CEREBRAL ISCHEMIC STROKE – UPDATES OF THE PATHOGENETIC MECHANISMS, CLINICAL, RADIOLOGICAL AND LABORATORY RISK FACTORS"), includes a detailed bibliographic analysis of the clinical-temporal characteristics of haemorrhagic transformation of cerebral vascular accidents, in the pathophysiological mechanisms of haemorrhagic transformation post-stroke, the risk and predictive factors for haemorrhagic transformation of cerebral infarction.

In the subsection, dedicated to the pathophysiological mechanisms of HT, are analysed the important structural elements of the blood-brain barrier (BBB), their lesion can induce the haemorrhagic complication; the neuroinflammatory mechanisms and the molecules associated with HT, but also the specific enzyme group – matrix metalloproteinases, in particular MMP-2 and MMP-9, which, based on the bibliographic sources, have an important role in the remodelling of the basement membrane of the blood-brain barrier, their increased proteolytic activity being associated with a higher rate of post-cerebral infarction haemorrhagic transformation [4].

The section dedicated to the bibliographic analysis of the risk and prediction factors for the haemorrhagic transformation of cerebral infarction includes the evaluation of data from the specialized literature regarding: clinical parameters associated with an increased risk of HT of acute ischemic stroke [8], including comorbidities and pre-stroke medication; brain imaging changes that increase the rate of HT complication of cerebral infarcts [6], as well as plasma biomarkers associated with an increased risk of HT after ischemic stroke, including non-specific and specific parameters (related to blood-brain barrier integrity impairment) [7].

CHAPTER 2 („STUDY MATERIAL AND METHODS") describes in detail the research methodology conducted for the thesis topic including a comprehensive description of the general characteristics of the study (the research sample, stages, and design of the study), as well as the applied investigation methods (clinical assessment, laboratory investigations, instrumental methods).

The data from the subjects included in the research was evaluated using computerized processing through descriptive, dispersion, and correlational analysis methods, applying the R program, version 4.1.1. To determine if the difference between samples was statistically significant, the independent samples t-test was applied for non-parametric data, and one-way ANOVA (analysis of variance) for parametric data. Values with $p<0.05$ were considered significant.

To assess the predictive value of the investigated parameters in predicting haemorrhagic transformation, as well as unfavourable clinical evolution during hospitalization and at 3 months following ischemic stroke (quantified by the mRS scale value), ROC analysis (Receiver Operating Characteristic analysis) was performed. This involved graphical representation in the form of ROC curves and calculation of the area under the ROC curve (AUC). Additionally, for each parameter included in the ROC analysis, a predictive threshold value was established.

CHAPTER 3 („CLINICAL, RADIOLOGICAL AND LABORATORY PARAMETERS OF CEREBRAL ISCHEMIC STROKE PATIENTS WITH AND WITHOUT HAEMORRHAGIC TRANSFORMATION") consists of 2 subchapters, with the first focusing

on the clinical and imaging characteristics of patients with ischemic stroke from the studied cohorts, and the second presenting a comparative assessment of laboratory parameters and biomarkers of blood-brain barrier integrity in the studied acute ischemic stroke patients.

3.1. Clinical and radiological particularities in the researched ischemic stroke patients

The conducted study included 150 persons, comprising 55 patients in the research cohort (Group A) - patients in whom haemorrhagic transformation of cerebral infarction was detected during hospitalization, and 95 patients in the control cohort (Group B) - patients with ischemic stroke that did not complicate with haemorrhagic transformation during their stay in the hospital.

The mean age of patients in the overall cohort is 71 years (minimum 41 years, maximum 96 years), with the mean age in Group A being 70 years (minimum 43 years, maximum 96 years), and in Group B - 71 years (minimum 41 years, maximum 94 years), without statistically significant differences, $p>0.05$.

Analysis *based on gender* indicates a roughly equal distribution in the overall cohort (54% female = 81 patients, 46% male = 69 patients), with similar results in the control group (49% female = 47 patients, 51% male = 48 patients) and a slight predominance of females in the research group (62% female = 34 patients, 38% male = 21 patients), which is statistically insignificant.

*The etiopathogenic classification TOAST (*Trial of ORG 10172 *in Acute Stroke Treatment, 1993*) for ischemic stroke has highlighted a predominance of type 2 cerebral infarction, determined by embolism of cerebral vessels of cardiac origin (figure 2).

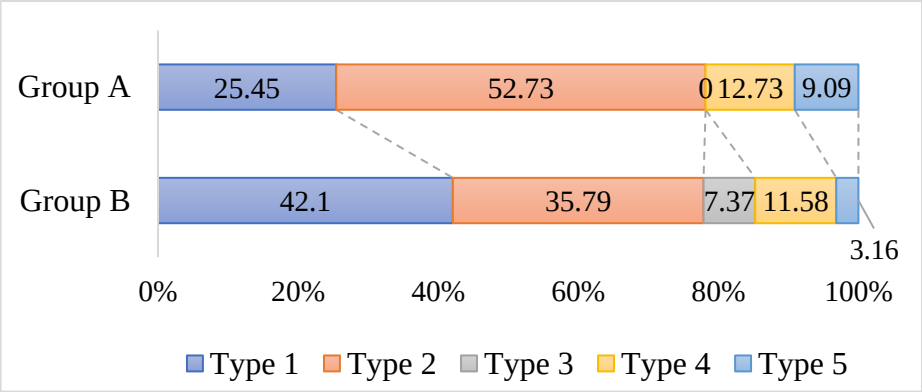

Figure 2. **Distribution of patients according to the etiopathogenic type of ischemic stroke based on TOAST classification (%). Group A = research cohort, Group B = control cohort.**

When we compared the research groups, we obtained the following results: in the control cohort, type 1 stroke - atherothrombotic (42.1%) predominates, closely followed by type 2 - cardioembolic stroke (35.79%). In the research group, patients with cardioembolic stroke clearly dominate - 52.73%, followed by patients with atherothrombotic stroke with a proportion twice as small - 25.45%.

After analysing the imaging parameters, within the conducted study, 55 patients (36.66%) presented haemorrhagic transformation of cerebral infarctions, the majority being of type I HT, namely haemorrhagic infarction (HI) type 1 - 23 patients, followed by a roughly equal number of cases for HT of HI type 2 (11 patients) and parenchymal hematoma (PH) type 1 (12 patients). The fewest cases were recorded for parenchymal hematomas of type 2 (9 patients).

Out of the 55 patients with HT in the current study, 17 (30.9% of HT cases and 11.33% of the total number of patients) presented worsening of the clinical condition requiring re-evaluation through cerebral imaging, where signs of intracerebral bleeding on a background of formed ischemic lesions were detected - symptomatic haemorrhagic transformation. On average, there was an increase of 5.12±0.98 points according to the NIHSS scale (minimum 1 point, maximum 14 points). Dichotomizing the results into 2 categories: an increase of 1-3 points, equivalent to minor symptomatic HT, and ≥4 points (major symptomatic HT), highlighted 8 patients with minor neurological deterioration (5.33% of the overall cohort), and 9 with deterioration (6% of the overall cohort).

We aimed to analyse if there is a *correlation between the etiopathogenetic type of cerebral infarction and the risk of haemorrhagic transformation* in the active working cohort. By logistic regression analysis, it was found that HT is significantly more frequent in patients with cardioembolic stroke compared to atherothrombotic stroke (p=0.026) and in patients with cryptogenic stroke compared to those with stroke determined by atherosclerosis of medium and large arteries (p=0.049).

Analysis of the spectrum and frequency of *risk factors for stroke* in the studied patients revealed that high blood pressure (HBP) was the most prevalent risk factor for developing a stroke, being present in 97% of the studied cases (146/150 patients). Comparative analysis of the frequency of risk factors in the studied groups (with and without subsequent HT of cerebral infarction) showed statistically significant differences only in the case of atrial fibrillation: 73% in the research group versus 47% in the control group, p = 0.003.

According to bibliographic data, there is a correlation between the treatments received prior to ischemic stroke and the risk of HT in the case of cerebral infarction. In this context, we analysed the medications received by patients prior to the development of ischemic events, overall and comparatively, in patients with (group A) and without subsequent HT (group B).

Antiplatelet and anticoagulant therapy are of particular interest in studies regarding the risk of haemorrhagic transformation, which is why we aimed to investigate this aspect in our research. The obtained results didn’t show statistically significant differences between the groups.

We have also recorded the patients who underwent cerebral *revascularization therapy by medical (thrombolysis) and/or surgical (thrombectomy)* upon hospital admission. Out of the total of 150 patients, 21 received such treatment (14%), of which 13 (24%) were in the research group and 8 (8.4%) were in the control group, with the difference being statistically significant (p = 0.01).

To highlight possible factors influencing the rate of haemorrhagic transformation of cerebral infarctions in the included patients, we compared the specific cerebral revascularization treatment methods applied in the two studied groups. Thus, we have established that all 8 patients in the control group received treatment with intravenous thrombolysis with rtPA, while in the research group, 5 patients received alteplase treatment, 5 underwent mechanical thrombectomy intervention, and 3 patients received initially rtPA (alteplase), followed by surgical thrombectomy due to lack of clinical improvement on medical therapy.

The severity of cerebral infarction, clinically, was quantified using several clinical scores, including the NIHSS scale (*National Institutes of Health Stroke Scale*), the SPAN-100 index (*Stroke Prognostication by Using Age and NIHSS score*), and the THRIVE score (*The Totaled Health Risks In Vascular Events*).

Upon admission, on average, in the general cohort, the NIHSS score was 13 points (minimum 2 points, maximum 30 points), while at discharge it was 9 points (minimum 0 points, maximum 24 points). When we comparatively analysed the evolution of the NIHSS scale across the study groups, we obtained statistically significant differences ($p<0.001$) both at admission and discharge. The results are depicted in Figures 3a and 3b.

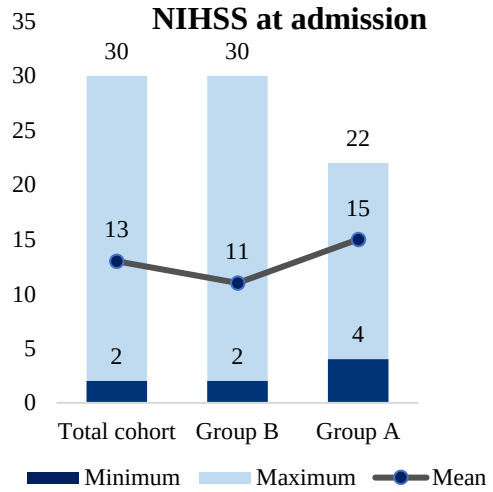

Figure 3a. **The comparative analysis of the study groups according to the accumulated NIHSS score at admission (mean values, minimum, maximum - points), with statistically significant difference between groups A and B (* p<0.001).**

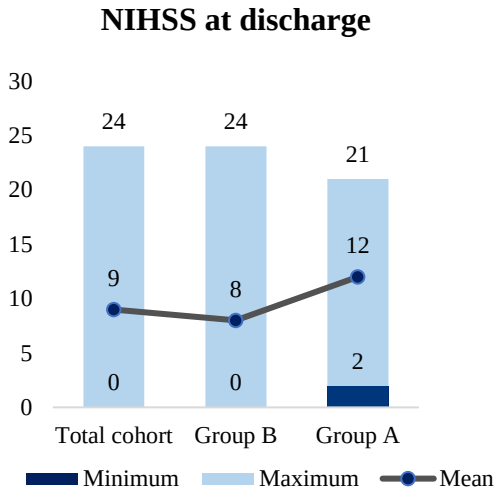

Figure 3b. **The comparative analysis of the study groups according to the accumulated NIHSS score at discharge (mean values, minimum, maximum - points), with statistically significant difference between groups A and B (* p<0.001).**

SPAN-100 index (Stroke Prognostication by Using Age and NIHSS score), which is obtained by summing the values of the NIHSS scale at admission (points) with the age in years of patients with cerebral infarction. When comparing patients in groups A and B, similar results were obtained, namely 84 points on average in patients with subsequent haemorrhagic transformation and 83 points in those without haemorrhagic transformation, with $p = 0.44$.

Analysis of the *THRIVE score* (The Totaled Health Risks In Vascular Events) values at admission, revealed statistically significant differences between the research groups. The mean score, according to this index, in the research group was 5 points (minimum 1 point, maximum 9 points), while in the control group it was 4 points (minimum 1 point, maximum 8 points), with $p = 0.019$. An increase in the score is associated with increased cardiovascular and cerebrovascular risks, as well as a decreased rate of favourable evolution in patients with stroke.

Assessment of the *post-stroke functional recovery degree*, according to the *mRS scale*, showed that at discharge only 3 out of 150 patients (2%) had excellent recovery (mRS 0-1 points). Favourable outcomes (mRS 0-2 points) were observed in 16 out of 150 patients (10.67%). Although at 3 months post-cerebral ischemic event, there is a shift to the right in the accumulated scores of patients, according to the mRS scale, the proportion of patients with excellent evolution (mRS 0-1 points) - 8.1% and favourable evolution (mRS 0-2 points) - 24.32% remains considerably lower compared to patients with unfavourable evolution (Figure 4).

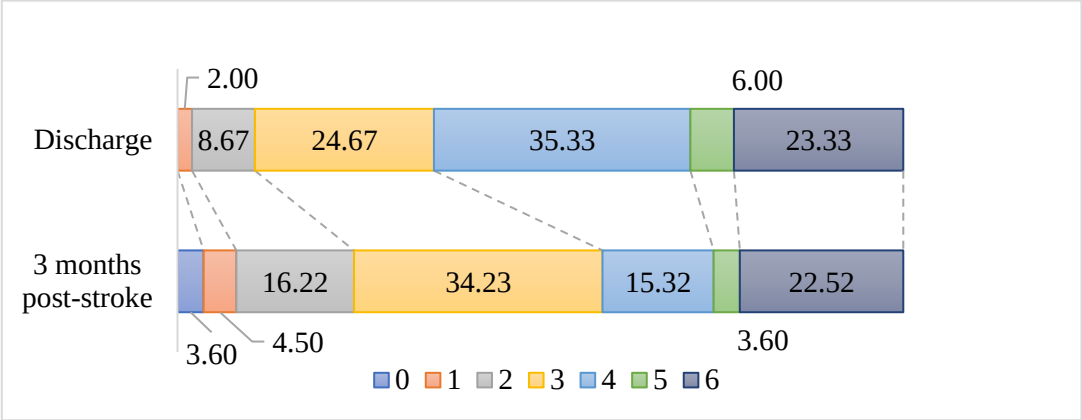

Figure 4**. The evolution of the score, according to the mRS scale (0-6 points), accumulated by the analysed patients (%), at discharge and 3 months follow-up.**

The separate analysis, by study groups, highlighted a less favourable evolution in the research cohort (with haemorrhagic transformation), where, both at discharge and at 3 months after the stroke, no cases of excellent neurological recovery (mRS 0-1 points) were recorded, and only one out of 55 patients (1.82%) had favourable recovery (mRS 0-2 points) at discharge. At the follow-up visit, the number of patients in the research group who showed favourable functional neurological recovery (mRS 0-2 points) reached a rate of 13.51% (5 out of 37 patients). Patients in the control group showed a different distribution of scores according to the mRS scale. The proportion of patients with excellent recovery (mRS 0-1 points) was 3.16% at discharge (3 out of 95 patients) and 12.16% (9 out of 74 patients) at 3 months. When analysing the rate of favourable evolution (mRS 0-2 points), we obtain 15.79% (15 out of 95 patients) at discharge and 29.73% (22 out of 74 patients) at 3 months.

The cerebral imaging study allowed the quantification of the extension of cerebral ischemic changes by calculating the *ASPECTS score*, both at the initial imaging examination and at the repeated one, by a neuroradiologist blinded to other investigated parameters in patients.

In our study, on average, at admission, patients presented an ASPECTS score of 8.2 ± 0.2 points (mean ± standard error), with significantly lower values in the research cohort where the accumulated score was 7.44 ± 0.34 points versus the control group with 8.65 ± 2.33 points, p = 0.004 (Figure 5).

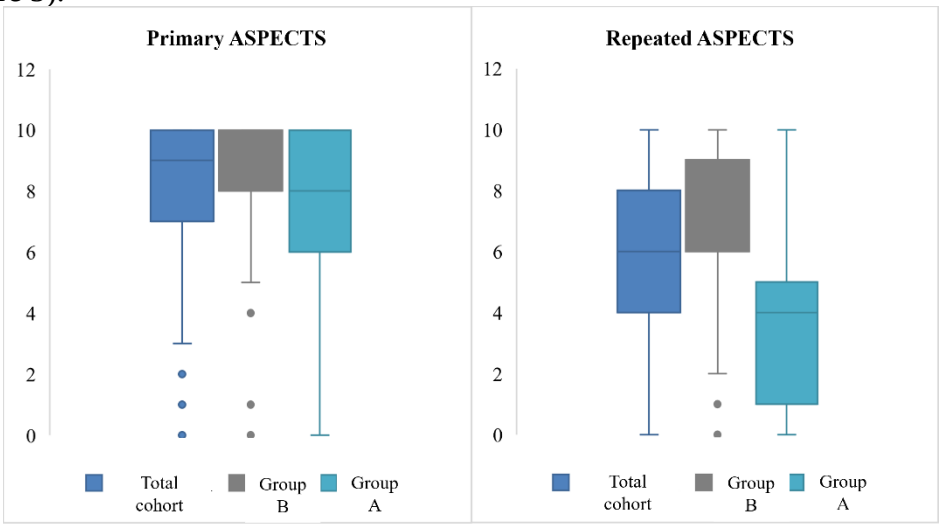

Figure 5. **Distribution by percentile in the boxplot diagram of the accumulated score by the studied patients, according to the ASPECTS score at admission and based on repeated cerebral imaging (points).**

The same trend is observed in the dynamic analysis of the ASPECTS score, calculated based on repeated cerebral imaging. Thus, overall, patients had 5.63 ± 0.26 points. Patients in the haemorrhagic transformation group had much lower results (3.68 ± 0.37 points) compared to those without haemorrhagic transformation (6.96 ± 0.27 points), $p < 0.001$ (Figure 5).

In Figure 5, we can observe a clearly negative inclination towards the lower percentiles of the values accumulated by patients in the research cohort, both at admission, but especially on repeated cerebral imaging.

3.2. Comparative analysis of the laboratory data and brain-blood barrier integrity biomarkers of the studied acute ischemic stroke patients

The average *blood glucose level at admission* in the studied patients was 7.6 ± 0.28 mmol/l (minimum 2.41 mmol/l, maximum 21.42 mmol/l), with similar values in the control group - 7.3 ± 0.37 mmol/l (minimum 2.41 mmol/l, maximum 21.42 mmol/l) and slightly higher but statistically insignificant figures ($p = 0.26$) in the research cohort - 8.0 ± 0.41 mmol/l (minimum 3.97 mmol/l, maximum 17.2 mmol/l).

The *analysis of coagulation tests* data performed on the studied patients allowed the inclusion of three laboratory parameters in the study, namely: INR value, plasma fibrinogen level, and platelet count. Overall, the mean INR value was 1.34 ± 0.02, fibrinogen level was 3.75 ± 0.03 g/l, and platelet count was 234 ± 6.4 10^9/l. Among the evaluated parameters, only for the INR value statistically significant differences were recorded ($p = 0.003$) between the compared groups, with mean values of 1.27 ± 0.03 in Group A and 1.38 ± 0.03 in Group B.

The study of lipidogram parameters in the patients included in the research revealed elevated values of total cholesterol, with an average of 5.42 ± 0.1 mmol/l, LDL cholesterol - 3.3 ± 0.07 mmol/l, serum triglycerides - 1.76 ± 0.06 mmol/l; but also slightly decreased values of HDL cholesterol - 1.33 ± 0.03 mmol/l, without significant differences between the analysed groups.

In the conducted study, we aimed to analyse the *parameters of the inflammatory reaction* from the complete blood count performed on patients upon admission to the hospital, namely the total number of plasma *leukocytes*, plasma *lymphocytes* level, *neutrophils*, and the *neutrophil/lymphocyte ratio* (NLR).

Table 1 shows the results of the total cohort of patients included in the study, indicating the mean values, medians, as well as the ranges within which these values fall (minimum-maximum values).

The comparative analysis of the hemoleucogram results, by study groups, reveals that patients in the research group had higher median values of both total leukocytes in the blood (9.3*109/l vs 8.5*109/l) and plasma neutrophils (6.7*109/l vs 5.7*109/l), without reaching the threshold required to become statistically significant, with p-values of 0.082 for leukocytes and 0.068 for neutrophils. The mean number of lymphocytes was also insignificantly higher in the research group (1.76±0.16*109/l) compared to patients in the control group (1.7±0.1*109/l), $p=0.75$. When calculating the neutrophil/lymphocyte ratio (NLR) in Group A patients (with HT), the mean value was 5.7±0.54, and in the control group - 5.2±0.47, with the difference between the compared groups being statistically insignificant ($p=0.47$). Additionally, Pearson correlation analysis didn't identify a significant negative correlation between this biomarker and the risk for haemorrhagic transformation ($r(148)=0.05$, $p=0.46$) or the patients' status at discharge ($r(148)=0.13$, $p=0.09$) and at 3 months post-stroke ($r(109)=0.09$, $p=0.34$).

Table 1. **The parameters of the inflammatory response in the patients included in the study**

Statistical indicators	**Analysed laboratory parameters**			
	Leukocytes (10^9/l)	*Neutrophils* (10^9/l)	*Lymphocytes* (10^9/l)	*NLR*
Mean (SD)	9.2 (3.8)	6.7 (3.4)	1.72 (1.08)	5.4 (4.4)
Median (IQR)	8.6 (4.3)	6.2 (4.2)	1.50 (0.90)	3.9 (4.3)
Range	3.5 - 27.2	1.8 - 21.1	0.40 - 6.90	0.3 - 21.7

[1]n (%); SD = standard deviation; IQR = interquartile range.
[2]Pearson's Chi-squared test; Fisher's exact test; Two Sample t-test.
NLR - neutrophil/lymphocyte ratio.

The following studied laboratory parameters were *matrix metalloproteinases 2 and 9 - MMP-2 and MMP-9*, these biomarkers being included in the research, given their influence on the proper functioning of the blood-brain barrier, the integrity of which is compromised in case of haemorrhagic transformation.

When applying the t-test for two independent samples, it was observed that there were no statistically significant differences in means between the studied cohorts, both for MMP-2 (p=0.72) and for MMP-9 (p=0.37). Thus, patients who developed haemorrhagic transformation during hospitalization did not have significantly higher levels of MMP-2 and MMP-9, assessed within the first 24 hours of admission, compared to individuals in the control group.

Additionally, to determine if there is any dependency between the variable plasma MMP-2 value and the variable haemorrhagic transformation, in the research and control groups, the one-way ANOVA test (one-way analysis of variance) was applied. The test results demonstrated that the relationship between the mentioned variables, even if positive, is not statistically significant, with the following values obtained: F (3, 51) = 2.19, with p=0.1. Similar results were obtained when applying the one-way ANOVA test for the analysis of the dependency between plasma MMP-9 value and the haemorrhagic transformation variable, where F (3, 51) = 0.36 and p=0.8, also being statistically insignificant.

Applying the t-test for independent samples, in the case of MMP-2, statistically significant differences were identified when comparing subgroups of patients with HT of haemorrhagic infarction types 1 and 2, with higher values observed in patients with type 2 haemorrhagic infarction, p=0.04. A similar analysis (on subtypes of HT) was performed for MMP-9, with no statistically significant differences recorded when comparing subgroups of patients with HT.

Given the multitude of factors that could influence/increase the risk of haemorrhagic transformation in patients with ischemic strokes, we applied the *regression analysis method*, which allows for establishing a possible relationship between multiple variables and the probability of a specific event, e.g., the risk of haemorrhagic transformation in our study.

Thus, we performed linear regression analysis to establish the relationship and possible correlations between the individual variables analysed and the risk of HT in the studied ischemic stroke patients, with the obtained data being represented graphically in Figure 6.

In the studied patients, the following parameters were associated with an increased risk of developing haemorrhagic transformation: NIHSS score at admission - OR = 1.11, 95% CI: 1.04-1.2, p = 0.003; THRIVE score at admission - OR = 1.25, 95% CI: 1.02-1.56, p = 0.04; history of atrial fibrillation - OR = 3.51, 95% CI: 1.63-7.94, p = 0.002; and the application of medicinal

and/or surgical cerebral revascularization treatment - OR = 2.76, 95% CI: 1.05-7.62, p = 0.04. Only 2 of the analysed parameters showed an inverse proportional relationship with the risk of developing post-cerebral infarction HT, higher values being associated with a decreased risk of haemorrhagic transformation, namely higher ASPECTS score at admission - OR = 0.77, 95% CI: 0.64-0.91, p = 0.003; and INR value in primary analyses - OR = 0.11, 95% CI: 0.02-0.54, p = 0.01.

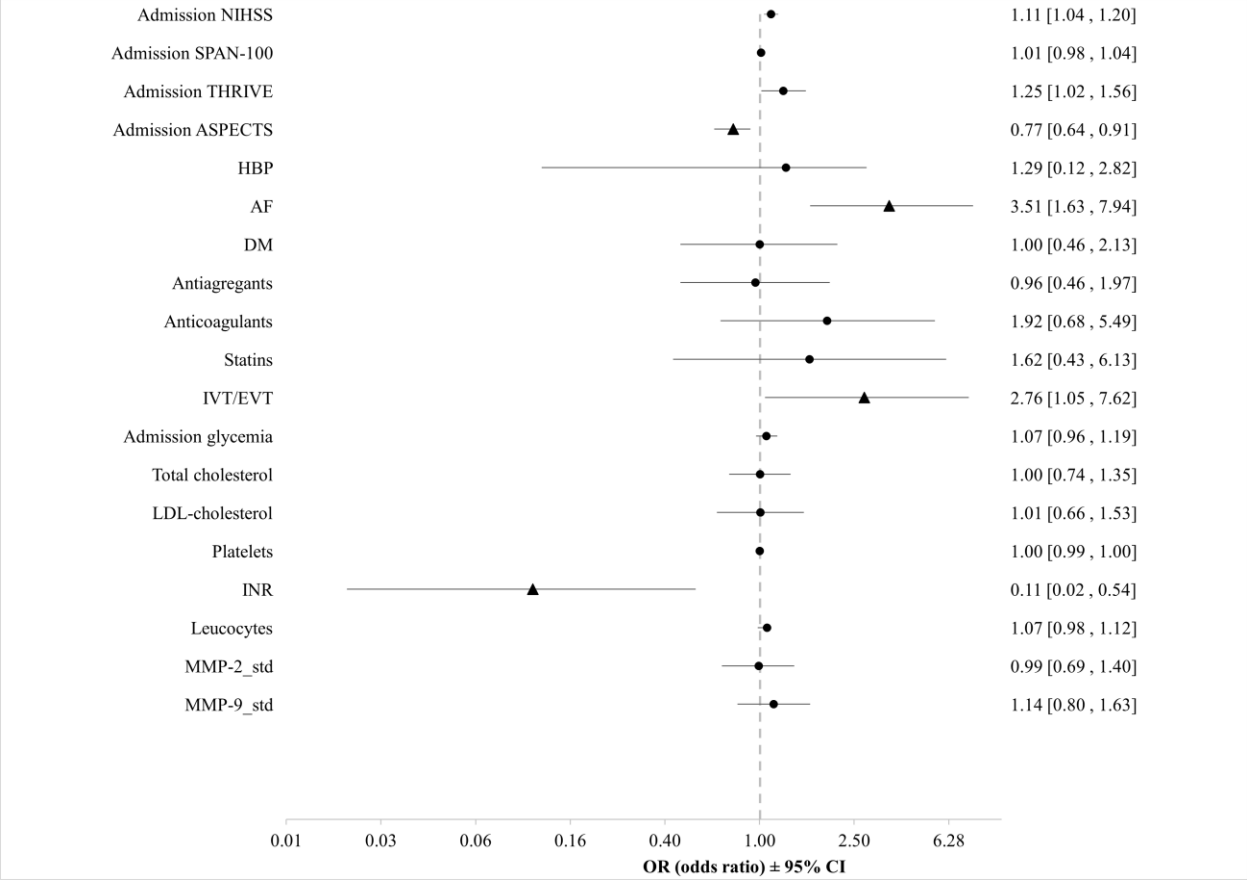

Figure 6. **Forest plot diagram of correlations between studied risk factors and the development of haemorrhagic transformation of cerebral infarctions in the studied patients, by the linear regression analysis. HBP – high blood pressure, AF – atrial fibrillation, DM – diabetes mellitus, IVT – intravenous thrombolysis, EVT – endovascular treatment, std – standardized, OR - odds ratio, CI - confidence interval.**

Following the same analysis algorithm, we investigated the relationship between the selected factors and the neurological status at discharge of the patients studied, by the mRS scale for assessing functionality and independence of post-cerebral infarction patients. As a result, according to the linear regression analysis, the parameters associated with unfavourable outcomes at discharge (higher mRS score) included: NIHSS score at admission - OR = 1.3, 95% CI: 1.13-1.56, p = 0.001; SPAN-100 score at admission - OR = 1.06, 95% CI: 1.01-1.12, p = 0.04; THRIVE score at admission - OR = 1.9, 95% CI: 1.26-3.13, p = 0.005; and history of atrial fibrillation - OR = 4.91, 95% CI: 1.38-23.02, p = 0.02. The ASPECTS score presented an inverse proportional relationship with patients' outcomes, with higher values associated with better functional status in the studied patients - OR = 0.52, 95% CI: 0.22-0.87, p = 0.05.

Adhering to the initially established research algorithm, the patients included in the study were to be evaluated at 3 months distance from the onset of stroke symptoms to determine the degree of recovery and functionality according to the mRS scale.

According to the data obtained from the linear regression analysis, the parameters significantly associated with unfavourable outcomes at 3 months post-stroke (higher mRS score) included: NIHSS score at admission - OR = 1.17, 95% CI: 1.06-1.3, p = 0.004; SPAN-100 score at admission - OR = 1.07, 95% CI: 1.02-1.12, p = 0.008; THRIVE score at admission - OR = 1.62, 95% CI: 1.19-2.3, p = 0.004; as well as the presence of atrial fibrillation as comorbidity - OR = 3.68, 95% CI: 1.41-10.21, p = 0.009. Similarly to the obtained results in the analysis of the risk for HT and the functional status at discharge of the studied patients, the ASPECTS score showed an inverse proportional relationship with the patients' outcomes at 3 months, with higher values being associated with better functional status - OR = 0.65, 95% CI: 0.42-0.91, p = 0.03.

In the final stage of the statistical analysis, our aim was to determine the predictive value of clinical, imaging, and laboratory parameters with significant results in previous stages of statistical analysis, in identifying patients at high risk of developing haemorrhagic transformation of cerebral infarction during hospitalization through the application of Receiver-Operating Characteristic (ROC) analysis and calculating the Area Under the Curve (AUC). The AUC values are represented as percentages for a more suggestive interpretation of the data.

According to the results from the multiple regression and logistic analysis, applied in the evaluated patients' data examination in the research, 3 parameters strongly correlated with the risk for HT: specifically, the admission ASPECTS score, the presence of atrial fibrillation in the concomitant diseases and the application of cerebral revascularization treatment (intravenous thrombolysis - IVT, endovascular treatment - EVT). To establish the predictive value of these parameters for haemorrhagic transformation, we performed ROC analysis and calculated the AUC, obtaining values of 76.51% (95% CI: 68.16%-84.86%) (figure 7), with a reasonable predictive power of HT.

The predictive capability of a diagnostic or prognostic test, as indicated by AUC values, is categorized as follows: 0.9-1.0 – excellent capability; 0.8-0.9 – good capability; 0.7-0.8 – fair capability; 0.6-0.7 – poor capability; 0.5-0.6 – no predictive/discriminatory capability (the result can be considered random) [9]. By multiplying the values by 100, they can be represented as percentages.

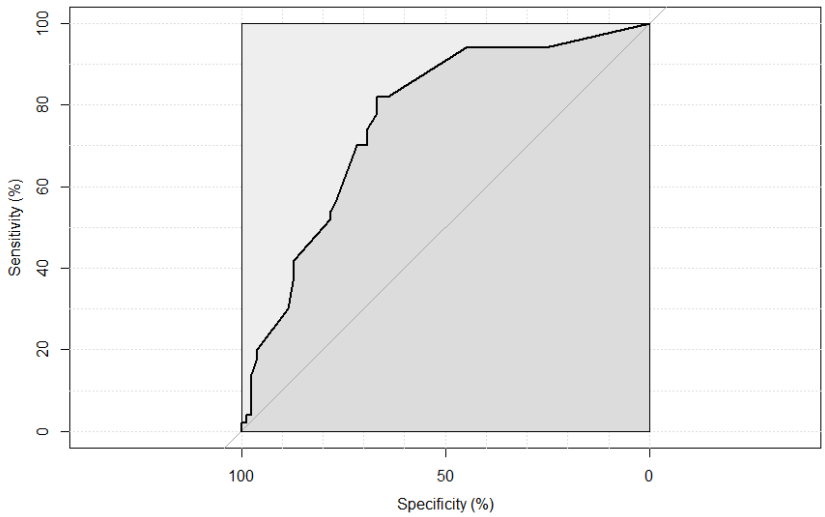

Figure 7**. ROC curve for assessing the predictive values in HT prognosis of ASPECTS score at admission, atrial fibrillation, and cerebral revascularization treatment**.

By multiple regression analysis, the strongest correlation with the risk of unfavourable outcomes (quantified by the mRS score) at discharge and 3 months post-stroke was found to be for admission NIHSS score. Therefore, we conducted ROC analysis and calculated AUC to determine the predictive power of the NIHSS score, obtaining, at discharge, values of 80.68% (95% CI: 67.29%-94.06%) - indicating a high prognostic capacity, and at 3 months post-stroke - 71.12% (68.16%-84.86%) – showing a reasonable prognostic capacity.

CHAPTER 4 („SYNTHESIS OF THE OBTAINED RESULTS") includes the comparative analysis of the results we obtained with those from the literature, highlighting the similarities and differences observed.

In the conducted research, the proportion of HT was 36.66% (55/150 patients), corresponding to current epidemiological data from the literature where the frequency of haemorrhagic transformation of ischemic stroke varies, in clinical studies, from 10 to 40% of the total number of patients, regardless of thrombolytic treatment [6,10], with slightly higher figures when studying revascularized patients (41.2% in patients treated by mechanical thrombectomy [11]; and 32% in patients undergoing reperfusion therapy with rtPA [12]).

The proportion of symptomatic haemorrhagic transformation cases (sHT) - defined as HT associated with worsening of neurological clinical status, quantified by an increase of at least 4 points according to the NIHSS scale [13], in the literature is reported to be 6% in patients treated with rtPA and <1% in the placebo groups of the respective studies. In the conducted study, we obtained a proportion of 6% of sHT cases (according to the ECASS II definition), data that are consistent with the analysed bibliographic resources, and the significant clinical worsening (≥4 NIHSS points) associated with HT was comparable to studies in reference publications.

The description of the patients included in the study was conducted comparatively between the study cohorts and began with the analysis of patients' ages, with the results indicating the following basic demographic traits: patients with HT had a mean age of 70 years (minimum 43 years, maximum 96 years), while those without HT had a mean age of 71 years (minimum 41 years, maximum 94 years), with no statistically significant differences between the groups. Literature data are controversial regarding the role of age as a predictor of HT, with studies where age is not a predictor factor, meaning there are no age differences between patients with and without HT, like our data [14], but also research where older age is an independent risk factor for haemorrhagic transformation [15].

Regarding gender distribution among patients with HT, in the current study, females predominated (62%), but the differences weren't statistically significant, while in the control group there was an approximately equal gender distribution (49% females, 51% males). The obtained results differ from those in the literature, where consistently, among patients with HT, males predominate, without gender being considered an independent risk/predictive factor for haemorrhagic transformation [16].

Given the correlation between the etiological subtype of cerebral infarction and the risk of HT [17], with ischemic strokes of cardioembolic type being associated with a higher rate of HT according to literature data [18], we compared the study cohorts based on this principle of analysis. The study results, similar to literature data, indicate a higher rate of cardioembolic type of ischemic stroke in patients with HT - 52.73%, compared to the control group - 35.79%.

In the context of achieving the research objective to highlight possible clinical risk and predictive factors for HT, we analysed the risk factor profile for stroke in the studied patients, noting that only atrial fibrillation (AF) showed statistically significant differences between

groups, being more common in patients with subsequent HT (73% versus 47%, p = 0.003). In the literature, AF is also reported among the most frequent risk factors associated with a major risk for TH [19]. It is worth mentioning that in the logistic regression analysis applied to the patients included in the study, there was an approximately 3.5-fold increase in the odds ratio for developing post-stroke HT in patients with AF (OR=3.5, p=0.004).

Furthermore, atrial fibrillation was associated with unfavourable outcomes during hospitalization (OR=4.9, 95% CI: 1.38-23.02, p=0.022), as well as at 3 months following the ischemic cerebral event (OR=3.68, 95% CI: 1.41-10.21, p=0.009), corresponding to other published studies, which indicate that AF not only increases the risk of stroke by about 5 times but is also associated with a doubling of mortality rate in these patients [20].

Of particular importance in assessing risk factors for HT is specific cerebral revascularization therapy, including IVT and EVT, which, based on data from the literature, is associated with an increase in the frequency of haemorrhagic transformation cases [21]. The data obtained in the present study indicate a significant difference between the compared groups in terms of the rate of application of cerebral revascularization therapy (IVT and EVT), with 24% in the research group and 8.4% in the control group, p=0.01.

To establish whether revascularization therapy increases the risk of HT, we conducted regression analysis, which established a certain correlation and an odds ratio of OR=4.31, p=0.07. At the same time, multiple regression analysis showed that cerebral revascularization therapy not only did not have a negative impact on the patients' condition at discharge but also had a favourable influence on it (lower mRS score at discharge), without reaching the threshold of statistical significance - OR=0.02, 95% CI: 0.00-0.75, p=0.072, a trend that became more evident and statistically significant at the 3-month post-stroke reassessment - OR=0.18, 95% CI: 0.03-0.91, p=0.05. The obtained data correspond to current literature data, especially when analysing studies with bridging therapy [22].

The comparative study of the NIHSS scale in the included cohorts revealed statistically significant differences (p<0.001), both at admission and discharge, similar to specialized bibliographic data, and the regression analysis identified a direct proportional relationship between the NIHSS score at admission and the risk of HT, with a statistically significant probability rate - OR=1.11, p=0.003 (univariate regression). This correlation persisted when other possible risk factors for HT were included in the multivariate regression analysis - OR=1.26, p=0.005. Additionally, the NIHSS score correlated with patients' condition at discharge and at 3 months post-stroke, with the obtained data being akin to those in the literature [23].

Literature analysis presents variable data regarding the value of the SPAN-100 score in predicting HT [21]. Patients in the compared groups in our study had similar inter-group results, with an average of 84 points in patients with subsequent HT and 83 in those without haemorrhagic transformation, p=0.44. Regression analysis did not establish a correlation between the SPAN-100 score calculated at admission and the risk of HT in the enrolled subjects, but a relationship was observed between this score and patients' condition at discharge, with a higher score being associated with unfavourable outcomes (OR=1.06, p=0.035), a relationship confirmed in the repeated assessment at 3 months following stroke (OR=1.06, p=0.008), resembling data from other studies that applied this score, which has a higher value in predicting the functional recovery of stroke patients than the risk of HT [24].

The third clinical score assessing stroke severity at admission, calculated for patients in the compared groups, was the THRIVE score, and its value calculated at admission revealed statistically significant differences between the study groups, p=0.019.

In line with findings in the specialized literature, the THRIVE score in the current study correlated with the risk of haemorrhagic transformation in the regression analysis (OR=1.25, p=0.038). The relationship between the THRIVE score and patients' condition at discharge and subsequently at 3 months post-stroke (quantified by the mRS score) also persisted in the regression analysis (at discharge - OR=1.9, p=0.005; at 3 months - OR=1.62, p=0.004) [25].

The quantification of the extent of cerebral ischemic changes in the patients in the current study was performed by calculating the ASPECTS score. According to literature data, the ASPECTS score correlates inversely with the risk of HT [26].

In the conducted study, patients had significantly lower ASPECTS scores at admission in the study cohort compared to the control group, p=0.004, consistent with findings in similar research [26]. Additionally, regression analysis highlighted the relationship between higher ASPECTS scores at admission and the reduction in HT risk (OR=0.7, p=0.001), as well as the likelihood of favourable outcomes at discharge (OR=0.52, p=0.05), and at 90 days post-stroke (OR=0.65, p=0.03).

Another research objective of the current study was the analysis of a series of standard laboratory biochemical parameters determined in patients with ischemic stroke, in the context of evaluating them as biomarkers of patients' outcomes, including their role in predicting haemorrhagic transformation complications.

Among the coagulation profile parameters, only the INR value showed statistically significant differences (p=0.003) between the compared groups, with lower mean values in the research group (with HT). Paradoxically and contrary to literature data [27], patients with haemorrhagic transformation in our study had lower INR values at admission, whereas higher values of this parameter are usually associated with haemorrhagic events. One possible explanation for this situation could be the prothrombotic effect of low INR values, leading to the formation of larger thrombi, which could result in severe cerebral infarction, independently posing a major risk for haemorrhagic transformation.

In the context of evaluating the role of blood-brain barrier integrity in the evolution of patients with acute ischemic stroke, as a research objective in the current study, plasma levels of MMP-2 and MMP-9 were determined in patients with ischemic stroke, with and without haemorrhagic transformation, to assess their correlation with the risk of HT.

Numerous studies in the literature have been published on the role of MMPs in predicting the outcome of patients with cerebral infarction, including a study conducted in the Republic of Moldova by Ciobanu N. (2020), which established a correlation between increased MMP levels and the evolution of patients with acute ischemic stroke, including the risk of haemorrhagic transformation [28].

However, in the patients from the research cohort in the current study, no statistically significant differences were found between MMP-2 (p=0.72) and MMP-9 (p=0.37) values in the group A (with HT) compared to the control group (group B). When we analysed the relationship between matrix metalloproteinases values and patients' outcomes at discharge, we observed an increased risk of unfavourable outcomes in the context of elevated MMP-2 and MMP-9 values at admission, with OR=1.45, 95% CI: 0.78-2.96, p=0.28 (MMP-2) and OR=1.13, 95% CI: 0.63-

2.13, p=0.67 (MMP-9), but statistically insignificant, results that can be explained by the relatively small number of analysed patients.

At 3 months after stroke, the correlation remained only for MMP-9 (OR=1.1, 95% CI: 0.69-1.8, p=0.65), but crossed the line of no effect, therefore being statistically insignificant.

Similar results were reported in a recent study (Cui Y. et al., 2022), where the authors recorded lower MMP-2 levels associated with an increased risk of haemorrhagic transformation, similar to the mean standardized data presented in the patients analysed by us, but also the lack of a correlation with the risk of haemorrhagic transformation for MMP-9 [29].

The predictive role of the studied parameters for the development of hemorrhagic transformation and unfavourable clinical evolution

The evolution of patients with ischemic stroke presents a high degree of uncertainty, hence the necessity of clear and feasible prognostic tools that could serve as support in managing these patients. According to literature data, the most promising results in predicting post-stroke evolution, including haemorrhagic transformation, are provided by the NIHSS score, in diverse variations (at admission, at 24 hours, the score variation from day to day, etc.) [30], similar data being obtained in the analysis conducted in the current research. Thus, the NIHSS score, separately, has demonstrated a weak predictive value, and when used in combination with clinical scores such as SPAN-100, THRIVE, and the imaging score ASPECTS, the predictive value increased to a reasonable one, which can be used as a prognostic tool for post-ischemic stroke haemorrhagic transformation. Reasonable prediction results of HT were also obtained by combining the ASPECTS score at admission, the presence of atrial fibrillation in comorbidities, and the application of cerebral revascularization therapy.

Regarding the prediction of unfavourable clinical evolution during hospitalization and at 3 months post-stroke (mRS score ≥2), the NIHSS score has shown high and reasonable prognostic values, akin to literature data [31], indicating that it can be used as a prognostic tool for the neurological status of patients at discharge and at 3 months after stroke.

The limitations of the research are as follows: patient recruitment was carried out at a single stroke center, and it would be necessary in the future to include patients from multiple stroke centers across the country, a situation that was not possible at the initiation stage of the study but could be implemented at present, when multiple acute stroke management centers have been organized in regions. Regarding laboratory biomarkers, an important limitation is the single measurement of MMP-2 and MMP-9 within the first 24 hours, pre-cerebral revascularization, with no possibility of sequential, repeated measurements during hospitalization, which would have increased their sensitivity/specificity in predicting the evolution of patients with acute ischemic stroke, including the risk of HT.

CONCLUSIONS

1. In patients with acute stroke, who were clinically, biochemically, and radiologically investigated, the following clinical risk factors and therapeutic management factors associated with an increased rate of hemorrhagic transformation were identified: insufficient primary antithrombotic prophylaxis, atrial fibrillation, elevated NIHSS and THRIVE clinical scores at admission (above the mean of 15 points and 5 points, respectively), cerebral revascularization treatment (pharmacological and/or surgical), administration of oral and parenteral anticoagulants within the first 72 hours of inpatient treatment.
2. The cerebral radiologic investigations in the analyzed patients revealed that the ASPECTS score, calculated at admission, exhibits an inversely proportional relationship with the risk of post-stroke haemorrhagic transformation (OR=0.77, p=0.003).
3. The biochemical analysis established that the plasma levels of MMP-9, but not of MMP-2, within the first 24 hours of stroke symptoms' onset, positively correlate with the risk of haemorrhagic transformation.
4. In the researched patients, mean INR levels below 1.27 at admission significantly inversely correlate with the risk of post-cerebral infarction HT (OR=0.11, p=0.01). Laboratory biochemical parameters: plasma fibrinogen level, INR, total cholesterol, LDL-cholesterol, HDL-cholesterol, triglycerides, glucose, leukocytes, neutrophils, lymphocytes, and platelets don't correlate with the risk of HT and unfavourable clinical evolution in patients with acute stroke.
5. According to our cohort, the mRS score and mortality rate are significantly higher in patients with HT, both at discharge (p=0.038) and at 3 months post-stroke (p<0.001). History of atrial fibrillation and elevated NIHSS, SPAN-100, and THRIVE scores at admission are associated with unfavourable evolution, while higher ASPECTS score at admission and cerebral revascularization therapy lead to lower disability and mortality rates at discharge and 3 months post-stroke.
6. The individual prediction formula for HT in patients with acute ischemic stroke, which includes variables such as ASPECTS score at admission, presence/absence of atrial fibrillation, and presence/absence of cerebral revascularization treatment, increases the predictive value to 76.51% compared to the combined formula of clinical scores (SPAN-100, THRIVE, NIHSS) and imaging score (ASPECTS) with a predictive value of 72.58%.

PRACTICAL RECOMMENDATIONS

1. To reduce the incidence of acute ischemic stroke and the rate of haemorrhagic transformation, efficient primary antiarrhythmic prophylaxis (maintaining sinus rhythm) and optimal antithrombotic treatment are recommended for patients with atrial fibrillation.
2. Systematic calculation of the ASPECTS imaging score is indicated for all patients with acute ischemic stroke to identify those at increased risk for unfavourable clinical evolution during hospitalization and at 3 months post-stroke, including the HT complication.
3. Parallel calculation of NIHSS, SPAN-100, and THRIVE scores is recommended for patients with acute ischemic stroke to increase the sensitivity of predicting HT and unfavourable evolution during hospitalization and at 3 months post-stroke.
4. Assessment of the blood-brain barrier integrity plasma biomarkers, specifically MMP-9, within the first 24 hours of stroke symptom onset, is recommended to predict the acute cerebral infarction patients' outcomes and assess the increased risk of haemorrhagic transformation.

5. Delayed initiation of oral anticoagulant therapy post-stroke is recommended for patients with moderate (from day 7), moderate-severe and severe cerebral infarction, with extensive lesions on primary brain imaging (from day 14), given the increased risk of haemorrhagic transformation in these patients.

6. The increased risk of HT shouldn't delay the implementation of specific cerebral revascularization treatment in patients within the therapeutic window, considering the positive correlation with functional recovery at discharge and 3 months post-stroke in the studied groups.

BIBLIOGRAPHY (selective)

1. Groppa S, Zota E, Crivorucica I, Gavriliuc M, Manole El. Accidentul vascular cerebral. Protocol clinic national. 2020;
2. Manole E, Lisnic V, Groppa S, Costru-Tasnic E, Filioglo A, Odainic O, et al. REGISTRUL RES-Q ÎN REPUBLICA MOLDOVA – PRIMELE REZULTATE NAȚIONALE ÎN CADRUL UNUI PROIECT INTERNAȚIONAL. *Bul Acad Științe, Științe Medicale*. 2017;
3. Groppa S, Zota E, Bodiu A, Gasnas A, Manole E, Ciobanu N. Diagnosis and management of ischemic stroke: time is critical. *Mold Med J*. 2020;63(4):65–74.
4. Wang W, Li M, Chen Q, Wang J. Hemorrhagic Transformation after Tissue Plasminogen Activator Reperfusion Therapy for Ischemic Stroke: Mechanisms, Models, and Biomarkers. *Mol Neurobiol [Internet]*. 2015;52(3):1572–9. Available from: http://link.springer.com/10.1007/s12035-014-8952-x
5. Drago D, Manea MM, Dobri AM, Stoican IC, Enache II, Ghenu MI, Tuta S. Risk factors for the outcome after thrombolysis in acute ischemic stroke - the prominent role of kidney dysfunction: A retrospective cohort observational study. *Med (United States)*. 2023;102(43):E35688.
6. Costru-Tasnic E, Plescan T, Gavriliuc M, Manole E, Odainic O. Complicația prin transformare hemoragică a accidentului vascular cerebral ischemic: factori predictivi clinici, imagistici și de laborator (Revista literaturii). *Bul Acad Științe, Științe Medicale [Internet]*. 2015;2(47):50–9. Available from: http://www.crri.acad.md/administrator/fisiere/editii/f35.pdf
7. Lakhan SE, Kirchgessner A, Tepper D, Leonard A. Matrix metalloproteinases and blood-brain barrier disruption in acute ischemic stroke. *Front Neurol*. 2013;4 APR(April):1–15.
8. Thomas SE, Plumber N, Venkatapathappa P, Gorantla V. A Review of Risk Factors and Predictors for Hemorrhagic Transformation in Patients with Acute Ischemic Stroke. *Int J Vasc Med*. 2021;2021.
9. Hosmer DWJ, Lemeshow S, Sturdivant RX. Assessing the Fit of the Model. In: *Applied Logistic Regression*. 2013. p. 153–225.
10. Meinel TR, Branca M, De Marchis GM, Nedeltchev K, Kahles T, Bonati L, et al. Prior Anticoagulation in Patients with Ischemic Stroke and Atrial Fibrillation. *Ann Neurol*. 2021;89(1):42–53.
11. Tian B, Tian X, Shi Z, Peng W, Zhang X, Yang P, et al. Clinical and Imaging Indicators of Hemorrhagic Transformation in Acute Ischemic Stroke after Endovascular Thrombectomy. *Stroke*. 2022;53(5):1674–81.
12. Honig A, Percy J, Sepehry AA, Gomez AG, Field TS, Benavente OR. Hemorrhagic Transformation in Acute Ischemic Stroke: A Quantitative Systematic Review. *J Clin Med*. 2022;11(5):1162.
13. Larrue V, Von Kummer R, Müller A, Bluhmki E. Risk factors for severe hemorrhagic transformation in ischemic stroke patients treated with recombinant tissue plasminogen activator: A secondary analysis of the European-Australasian Acute Stroke Study (ECASS II). *Stroke*. 2001;32(2):438–41.

14. Sun J, Lam C, Christie L, Blair C, Li X, Werdiger F, et al. Risk factors of hemorrhagic transformation in acute ischaemic stroke: A systematic review and meta-analysis. *Front Neurol*. 2023;14.
15. Guo Y, Yang Y, Zhou M, He L. Risk factors of haemorrhagic transformation for acute ischaemic stroke in Chinese patients receiving intravenous recombinant tissue plasminogen activator : a systematic review and meta-analysis. *Stroke Vasc Neurol*. 2018;May 26(3(4)):203–8.
16. Wen L, Zhang S, Wan K, Zhang H, Zhang X, Omboni S. Risk factors of haemorrhagic transformation for acute ischaemic stroke in Chinese patients receiving intravenous thrombolysis: A meta-analysis. *Med (United States)*. 2020;99(7).
17. Ge WQ, Chen J, Pan H, Chen F, Zhou CY. Analysis of Risk Factors Increased Hemorrhagic Transformation after Acute Ischemic Stroke. *J Stroke Cerebrovasc Dis [Internet]*. 2018;27(12):3587–90. Available from: https://doi.org/10.1016/j.jstrokecerebrovasdis.2018.08.028
18. Dang H, Ge WQ, Zhou CF, Zhou CY. The Correlation between Atrial Fibrillation and Prognosis and Hemorrhagic Transformation. *Eur Neurol*. 2020;82(1–3):9–14.
19. Meinel TR, Wilson D, Gensicke H, Scheitz JF, Ringleb P, Goganau I, et al. Intravenous Thrombolysis in Patients With Ischemic Stroke and Recent Ingestion of Direct Oral Anticoagulants. *JAMA Neurol*. 2023;80(3):233.
20. Jensen M, Schlemm E, Cheng B, Lettow I, Quandt F, Boutitie F, et al. Clinical Characteristics and Outcome of Patients With Hemorrhagic Transformation After Intravenous Thrombolysis in the WAKE-UP Trial. *Front Neurol*. 2020;11(August):1–8.
21. Teekaput C, Thiankhaw K, Tanprawate S, Teekaput K, Chai-Adisaksopha C. Outcomes of asymptomatic recombinant tissue plasminogen activator associated intracranial hemorrhage. *PLoS One [Internet]*. 2022;17(8 August):1–13. Available from: http://dx.doi.org/10.1371/journal.pone.0272257
22. Wang X, Ye Z, Busse JW, Hill MD, Smith EE, Guyatt GH, et al. Endovascular thrombectomy with or without intravenous alteplase for acute ischemic stroke due to large vessel occlusion : a systematic review and meta- - analysis of randomized trials. *Stroke Vasc Neurol*. 2022;Dec(7(6)):510–7.
23. Yang C, Zhang J, Liu C, Xing Y. Comparison of the risk factors of hemorrhagic transformation between large artery atherosclerosis stroke and cardioembolism after intravenous thrombolysis. *Clin Neurol Neurosurg [Internet]*. 2020;196(3):106032. Available from: https://doi.org/10.1016/j.clineuro.2020.106032
24. de Andrade JBC, Mohr JP, Ahmad M, Lima FO, Barros LCM, Silva GS. Accuracy of predictive scores of hemorrhagic transformation in patients with acute ischemic stroke. *Arq Neuropsiquiatr*. 2022;80(5):455–61.
25. Flint AC, Faigeles BS, Cullen SP, Kamel H, Rao VA, Gupta R, et al. Thrive score predicts ischemic stroke outcomes and thrombolytic hemorrhage risk in vista. *Stroke*. 2013;44(12):3365–9.
26. Raychev R, Saver JL, Jahan R, Nogueira RG, Goyal M, Pereira VM, et al. The impact of general anesthesia, baseline ASPECTS, time to treatment, and IV tPA on intracranial hemorrhage after neurothrombectomy: Pooled analysis of the SWIFT PRIME, SWIFT, and STAR trials. *J Neurointerv Surg*. 2020;12(1):2–6.
27. Jiao Y, Li G, Xing Y, Liu X. In fl uencing factors of hemorrhagic transformation in non-thrombolysis patients with cerebral infarction. *Clin Neurol Neurosurg [Internet]*. 2019;181(February):68–72. Available from: https://doi.org/10.1016/j.clineuro.2019.04.018
28. Ciobanu N. Particularitățile patogenice ale accidentului vascular cerebral ischemic la pacienții cu sindrom metabolic. Chisinau: C.Z.U.: [616.831-005/009.86]:616-008.9; 2020. 1–167 p.
29. Hong CT, Chiu WT, Chi NF, Lai LY, Hu CJ, Hu HH, et al. Low-density lipoprotein level

on admission is not associated with postintravenous thrombolysis intracranial hemorrhage in patients with acute ischemic stroke. *J Investig Med*. 2019;67(3):659–62.
30. Chen L, Chen N, Lin Y, Ren H, Huang Q, Jiang X, et al. Glucose to Platelet Ratio: A Potential Predictor of Hemorrhagic Transformation in Patients with Acute Ischemic Stroke. *Brain Sci*. 2022;12(9):1–11.
31. Matsumoto K, Nohara Y, Soejima H. Stroke Prognostic Scores and Data-Driven Prediction of Clinical Outcomes After Acute Ischemic Stroke. *Stroke*. 2020;51(5):1477–83.

LIST OF PUBLICATIONS AND SCIENTIFIC EVENTS

where were presented the research results of the PhD thesis in medical sciences on the topic „Study of potential biomarkers for prediction of haemorrhagic transformation of cerebral ischemic stroke", realized within the Department of neurology nr.1, *Nicolae Testemitanu* State University of Medicine and Pharmacy, by Mrs Costru-Tașnic Elena.

SCIENTIFIC PUBLICATIONS

- **Articles published in journals from abroad:**
  - ✓ **Articles in ISI, SCOPUS journals and other international databases***

    1. **Costru-Tașnic E.**, Gavriliuc M., Manole E. Serum biomarkers to predict haemorrhagic transformation and ischemic stroke outcome in a prospective cohort study. In: *Journal of Medicine and Life*. 2023 June; 16(6):908-914. ISSN: 1844-122X (Print), ISSN: 1844-3117 (Online), DOI: 10.25122/jml-2023-0148 (IF: 2,1).
- **Articles published in accredited national journals:**
  - ✓ **Articles published in B+ category journals:**

    2. **Costru-Tașnic E.**, Gavriliuc M., Manole E., The importance of matrix metalloproteinases in the prognosis of acute ischemic stroke patients. In: *Moldovan Medical Journal,* September 2021;64(3):2, ISSN 2537-6373 (Print), ISSN 2537-6381 (Online), https://doi.org/10.52418/moldovan-med-j.64-3.21.09.
  - ✓ **Articles published in B category journals**

    3. Pleșcan T., **Costru-Tasnic E.**, Gavriliuc M., Arion M., Dacin I. Hemorrhagic transformation of ischemic stroke – prediction and evaluation with different computed tomography modalities. În: *Curierul Medical*. 2015; (4): 63-73. ISSN 1857-0666.

    4. **Costru-Tașnic E.**, Pleșcan T., Gavriliuc M., Manole E., Odainic O. Complicația prin transformare hemoragică a accidentului vascular cerebral ischemic: factori predictivi clinici, imagistici și de laborator. În: *Buletinul Academiei de Științe a Moldovei. Științe Medicale*. 2015; (2): 50-59. ISSN 1857-0011.

    5. Pleșcan T., **Costru-Tașnic E.**, Gavriliuc M. Clinical application of perfusion computed tomography in the early diagnosis of acute ischemic stroke and hemorrhagic transformation prediction. În: *Curierul Medical*. 2015; (5): 24-32. ISSN 1857-0666.

    6. Pleșcan T., **Costru-Tașnic E.**, Gavriliuc M., Gavriliuc P., Odainic O. Reperfuzia sectorului masiv al nucleului infarctului cerebral ischemic acut emisferial cu utilizarea tratamentului trombolitic în fereastra terapeutică, confirmată prin perfuzie CT (tomografie computerizată). În: *Buletinul Academiei de Științe a Moldovei. Științe Medicale*. 2017; (2): 278-282. ISSN 1857-0011.

    7. **Costru-Taşnic E.**, Pleșcan T., Manole E., Gavriliuc M., Olesea O. Corelații clinico-imagistice la pacienții cu infarct cerebral în circulația posterioară cerebrală. În: *Buletinul Academiei de Științe a Moldovei. Științe Medicale.* 2017; (5): 63-68. ISSN 1857-0011.

    8. Manole E., Lisnic V., Groppa S., **Costru-Tasnic E.**, Filioglo A., Odainic O. et al. Registrul RES-Q în Republica Moldova – primele rezultate naţionale în cadrul unui proiect

internaţional. În: *Buletinul Academiei de Ştiinţe. Ştiinţe Medicale*, 5(57), 2017, p. 72-77. ISSN 1857-0011.

- **Abstracts published for national and international scientific conferences:**

9. **Costru-Tasnic E.**, Gavriliuc M., Lisnic V., Assessment of risk factors for haemorrhagic transformation in ischemic stroke patients. In: *Journal of the Neurological Sciences*, Vol. 357, Supplement e393. Published in issue: October 15, 2015. ISSN: 0022-510X (Print), ISSN 1878-5883 (Online), DOI: https://doi.org/10.1016/j.jns.2015.08.1394 (IP: 2.126).

10. Lisnic V., Groppa S., Efremova D., Manole E., **Costru-Tasnic E.** Stroke peculiarities in Moldova's population. Implementation of the ESO-EAST project. In: *Materials of the XIIth Congress of the Society for the Study of Neuroprotection and Neuroplasticity*. Tbilisi, Georgia, 6-9 October 2016, pp.18-19.

11. **Costru-Tasnic E.**, Pleșcan T., Manole E., Gavriliuc M., Odainic O. Computed tomography permeability to predict hemorrhagic transformation in ischemic stroke. In: *European Journal of Neurology*. 2017; 24(Suppl. 1): p706. Online ISSN: 1468-1331 (IF: 4.621).

12. **Costru-Tasnic E.**, Manole E., Gavriliuc M., Odainic O., Gavriliuc P., Filioglo A., Stroke registries to improve patient's outcome - first data of institutional stroke registry. In*: Journal of the Neurological Sciences*, Vol. 381, Supplement p406. Published in issue: October 15, 2017 (IP: 2.295).

13. **Costru-Tasnic E.**, Biomarkeri plasmatici ai accidentelor vasculare cerebrale ischemice. În: *Buletinul informativ al Conferinței Naționale cu participare internațională*. Iași, România, 2017, nr. 5, pp. 90-91.

14. **Costru-Tasnic E.**, Gavriliuc M. Serum biomarkers of blood brain barrier integrity to predict hemorrhagic transformation of ischemic stroke: a prospective study. In: *European Stroke Journal*, 2018, Vol. 3(1S), p.514. ISSN: 23969873 (Print), eISSN: 23969881 (Online) (IF: 1.095).

15. Manole E., Groppa S., **Costru-Tasnic E.** et al. In-hospital management of acute stroke in the Republic of Moldova – analysis of first data of the RES-Q as a part of ESO-East Project. In: *European Stroke Journal*, 2018, Vol. 3(1S), pp. 321-322 ISSN: 23969873 (Print), eISSN: 23969881 (Online) (IF: 1.095).

16. **Costru-Tasnic E.**, Gavriliuc P., Plescan T., Manole E., Odainic O. Posterior reversible encephalopathy syndrome mimicking stroke in a young woman. In: *Abstract book, 7th International Medical Congress for Students and Young Doctors*, 2018, p. 29.

17. Gavriliuc P., **Costru-Tasnic E.**, Plescan T., Dacin I., Intracerebral hemorrhage in a patient with moyamoya syndrome: case report. In: *Abstract book, 7th International Medical Congress for Students and Young Doctors*, 2018, pp. 29-30.

18. Plescan T., **Costru-Tasnic E.**, Gavriliuc P., Current achievements in reporting brain imaging in ischemic stroke: zones and score ASPECTS. In: *Abstract book, 7th International Medical Congress for Students and Young Doctors*, 2018, p.55-56.

19. Pleșcan T., **Costru-Tașnic E.**, Gavriliuc P., Manole E., Gavriliuc M., Arion M. Predicția transformării hemoragice a infarctului cerebral prin perfuzie CT. The IVth Congress of Radiology and Medical Imaging of the Republic of Moldova with international participation. In: *The Moldovan Medical Journal*. 2018; 61: 92. ISSN 2537-6373 (Print), ISSN 2537-6381 (Online).

20. **Costru-Tașnic E.**, Gavriliuc M. Clinical and laboratory biomarkers to predict haemorrhagic transformation of ischemic stroke: first data of a prospective study. In: *Moldovan Medical Journal.* September 2021;64(Neuro Congress Issue). ISSN 2537-6373 (Print), ISSN 2537-6381 (Online).

21. **Costru-Tașnic E.** Aplicarea scorurilor clinice în predicția transformării hemoragice a accidentelor vasculare cerebrale ischemice. În: *Culegere de rezumate, Conferința*

științifică anuală „Cercetarea în biomedicină și sănătate: calitate, excelență și performanță" Revista de Ştiinţe ale Sănătăţii din Moldova, 29(3)/2022, ANEXA 1, p. 260.

22. **Costru-Taşnic E.**, Gavriliuc M., Manole E. Infectious complications correlate with both discharge and 3 months follow-up functional status of ischemic stroke patients with haemorrhagic transformation. In: *European Stroke Journal*. 2023, Vol. 8(2S) 3–669, p. 566. ISSN: 23969873 (Print), eISSN: 23969881 (Online), DOI: https://doi.org/10.1177/23969873231169660 (IF: 5.894).

23. **Costru-Taşnic E.**, Gavriliuc M., Manole E. Previous statins use and the risk for haemorrhagic transformation in acute ischemic stroke patients. In: *European Journal of Neurology*, 2023, 30: 330-742, p. 349. ISSN:1468-1331 (Online). DOI: https://doi.org/10.1111/ene.15950 (IF: 6.228).

24. **Costru-Taşnic E.**, Tratamentul cu anticoagulante orale pre-stroke și evoluția post-infarct cerebral la pacienții cu fibrilație atrială. În: *Culegere de rezumate, Conferința științifică anuală „Cercetarea în biomedicină și sănătate: calitate, excelență și performanță" Revista de Ştiinţe ale Sănătăţii din Moldova*, 2023, 10(3) / ANEXA 1, p. 353.

- **Invention patents, patents, registration certificates, materials for invention salons:**

25. Costru-Taşnic E., Gavriliuc M., Manole E. Aplicarea scorurilor clinice NIHSS, SPAN-100 și THRIVE în evidențierea pacienților cu infarct cerebral cu risc majorat pentru transformare hemoragică. Certificat de inovator nr. 6101, 27.06.2023.

- **Active participation with oral presentations within scientific forums:**

✓ **international organised abroad**

26. Costru-Taşnic E. Biomarkerii plasmatici ai accidentelor vasculare cerebrale ischemice. *Conferința Națională de Neuroștiințe cu participare internațională, Congresul reunit de neurologie, Iași-Chișinău 2017, ediția a XV-a*. Iași, România, 19-22 octombrie 2017.

✓ **international organised in Republic of Moldova**

27. Costru-Taşnic E., Gavriliuc P., Pleșcan T., Manole E., Odainic O. Posterior reversible encephalopathy syndrome mimiking stroke in a young woman. *7th International medical congress for students and young doctors MedEspera*. Chișinău, 3-5 mai 2018.

28. Pleșcan T., Costru-Taşnic E., Gavriliuc P. Current achievements in reporting brain imaging in ischemic stroke: zones and score ASPECTS. *7th International medical congress for students and young doctors MedEspera*. Chișinău, 3-5 mai 2018.

✓ **national**

29. Costru-Taşnic E., Pleșcan T., Manole E., Gavriliuc M., Odainic O. Corelații clinic-imagistice la pacienții cu infarct cerebral în circulația posterioară cerebrală. *Congresul al VI-lea al Neurologilor și Neurochirurgilor din Republica Moldova*. Chișinău, 2-5 octombrie 2017.

30. Costru-Taşnic E. Rolul biomarkerilor în managementul accidentelor vasculare cerebrale ischemice. *Conferința Științifică: Actualități în tratamentul patologiilor sistemului nervos, sesiunea I – Actualități în patologia vasculară medulară și cerebrală. Management terapeutic și chirurgical*, Chișinău, 14 Septembrie 2018.

31. Costru-Taşnic E., Gavriliuc M. Biomarkeri clinici și de laborator de predicție a transformării hemoragice a infarctului cerebral: rezultate preliminare ale unui studiu prospectiv. *Congresul VII al Neurologilor din Republica Moldova*, Chișinău, 16-18 septembrie, 2021.

32. Costru-Taşnic E., Aplicarea scorurilor clinice în predicția transformării hemoragice a accidentelor vasculare cerebrale ischemice. *Conferința științifică anuală „Cercetarea în biomedicină și sănătate: calitate, excelență și performanță",* Chișinău, 19-21 octombrie 2022.

33. Costru-Taşnic E., Tratamentul cu anticoagulante orale pre-stroke și evoluția post-infarct cerebral la pacienții cu fibrilație atrială. *Conferința științifică anuală „Cercetarea în*

biomedicină și sănătate: calitate, excelență și performanță", Chișinău, 18-20 octombrie 2023.

- **Poster presentations within scientific forums:**
  - ✓ **international**

34. Costru-Tasnic E., Manole E., Gavriliuc M., Odainic O., Gavriliuc P., Filioglo A., Stroke registries to improve patient's outcome - first data of institutional stroke registry. *Congresul Mondial de Neurologie*, Kyoto, Japonia, 16-21 septembrie 2017.

35. Costru-Tasnic E., Gavriliuc M. Serum biomarkers of blood brain barrier integrity to predict hemorrhagic transformation of ischemic stroke: a prospective study. *4th European Stroke Organisation Conference*, Gothenburg, Suedia, 16-18 Mai 2018.

36. Manole E., Groppa S., Costru-Tasnic E. et al. In-hospital management of acute stroke in the Republic of Moldova – analysis of first data of the RES-Q as a part of ESO-East Project. *4th European Stroke Organisation Conference,* Gothenburg, Suedia, 16-18 Mai 2018.

37. Costru-Tasnic E., Gavriliuc M., Cojocaru L., Odainic O., Chetrari O., Plescan T., Manole E. Occult malignancy to cause embolic stroke in a young patient – How often can it be? *4th Congress of the European Academy of Neurology*, Lisabona, Portugalia, 16-19 Iunie 2018.

38. Costru-Tașnic E., Gavriliuc M., Manole E. Infectious complications correlate with both discharge and 3 months follow-up functional status of ischemic stroke patients with haemorrhagic transformation. *9th European Stroke Organisation Conference*, Munich, Germany, 24-26 Mai 2023.

39. Costru-Tașnic E., Gavriliuc M., Manole E. Previous statins use and the risk for haemorrhagic transformation in acute ischemic stroke patients. *9th Congress of the European Academy of Neurology*, Budapesta, Ungaria, 01-04 Iulie 2023.

ADNOTARE

Costru-Taşnic Elena, „Studiul potențialilor biomarkeri de predicție a transformării hemoragice a infarctului cerebral”, teză de doctor în științe medicale, Chișinău, 2024

Structura tezei: introducere, 3 capitole, sinteza rezultatelor obținute, concluzii generale, recomandări practice, 112 pagini text de bază, un indice bibliografic cu 224 de surse. Materialul ilustrativ include 42 de figuri, 10 tabele și 5 anexe. Rezultatele obținute sunt publicate în 24 de lucrări științifice și au fost prezentate în cadrul a 14 forumuri științifice naționale și internaționale

Cuvinte-cheie: accident vascular cerebral acut, transformare hemoragică, factori de risc, biomarkeri, scoruri de predicție, integritatea barierei hematoencefalice, metaloproteinaze matriceale, prognosticul recuperării neurologice.

Domeniul de studiu al tezei: neurologie.

Scopul lucrării: studiul factorilor de risc, al corelațiilor clinico-biochimice și al relevanței unor biomarkeri clinici și paraclinici în prognosticul transformării hemoragice a infarctului cerebral acut.

Obiectivele lucrării: 1. Evaluarea factorilor de risc clinici pentru transformarea hemoragică a infarctelor cerebrale și calcularea scorurilor de apreciere a severității bolii: NIHSS (*National Institutes of Health Stroke Scale),* SPAN-100 (*Stroke Prognostication Using Age and NIHSS),* THRIVE (*Totaled Health Risks in Vascular Events*), pentru evidențierea persoanelor cu prognostic negativ și risc sporit pentru transformare hemoragică a infarctelor cerebrale; 2. Stabilirea rolului scorului ASPECTS (*Alberta Stroke Programme Early CT Scale*) în prognosticul transformării hemoragice a accidentelor vasculare cerebrale ischemice, care ar permite evidențierea timpurie a cazurilor de transformare hemoragică; 3. Determinarea corelațiilor dintre nivelului plasmatic al MMP-2 și MMP-9 la pacienții cu accidente vasculare cerebrale ischemice și riscul de transformare hemoragică a infarctelor cerebrale; 4. Aprecierea corelației unor parametri de laborator determinați în mod standard la pacienții cu accident vascular cerebral ischemic (nivelul plasmatic de fibrinogen, INR, colesterol total, LDL-colesterol, HDL-colesterol, trigliceride, glucoză, leucocite, neutrofile, limfocite, trombocite) cu riscul dezvoltării transformării hemoragice a infarctului cerebral; 5. Aprecierea ratei de independență funcțională, conform scalei mRS (*modified Rankin Scale*), a pacienților cu evenimente vasculare cerebrale ischemice, cu şi fără transformare hemoragică, la externare și la 3 luni distanță de la debutul bolii.

Noutatea şi originalitatea ştiinţifică: a fost realizată o analiză corelațională amplă a multiplilor factori de risc clinici, paraclinici (imagistici și de laborator – specifici/nespecifici), cât și a scorurilor de apreciere a severității clinice (clinici și imagistici), la pacienți cu infarct cerebral acut cu scopul de a stabili relația cu riscul pentru TH, dar și cu evoluția recuperării neurologice la externare și 3 luni distanță de la evenimentul ischemic cerebral. Au fost studiați biomarkerii afectării integrității barierei hematoencefalice (MMP-2, MMP-9) și corelația acestora cu evoluția pacienților post-AVC (TH, evoluție nefavorabilă).

Problema științifică soluționată în studiu: evidențierea factorilor de risc pentru TH la pacienții cu AVC ischemic acut, ceea ce va permite stratificarea riscului pentru această complicație, determinând altfel optimizarea măsurilor terapeutice și de profilaxie secundară post-stroke. S-au identificat factori de prognostic clinici, imagistici și de laborator al pacienților cu infarct cerebral acut, cu recomandări de management intraspitalicesc și prevenție.

Semnificația teoretică: evaluarea particularităților clinice, imagistice și biochimice la pacienții cu AVC ischemic acut drept factori de risc pentru TH și evoluție clinică nefavorabilă. Studiul a adus informații suplimentare privind aplicarea scorurilor clinice, imagistice și biomarkerilor de laborator în prognosticul complicației prin TH a infarctelor cerebrale acute.

Valoarea aplicativă: rezultatele studiului încurajează aplicarea sistematică a scorurilor clinice (NIHSS, SPAN-100, THRIVE) și imagistice (ASPECTS) analizate pentru stratificarea pacienților cu infarct cerebral acut cu risc sporit pentru evoluție nefavorabilă, inclusiv pentru TH, ceea ce va determina monitorizarea mai activă a acestora și limita complicațiile post-stroke.

Implementarea rezultatelor științifice: Rezultatele studiului au fost implementate în activitatea clinică a Institutului de Neurologie și Neurochirurgie „Diomid Gherman”, secția Neurourgențe, precum și în activitatea didactică a Catedrei de Neurologie nr.1 a USMF „Nicolae Testemițanu”.

În baza rezultatelor tezei a fost obținut un Certificat de inovator, nr. 6101 din 27 iunie 2023, pentru inovația cu titlul „Aplicarea scorurilor clinice NIHSS, SPAN-100 și THRIVE în evidențierea pacienților cu infarct cerebral cu risc majorat pentru transformare hemoragică”.

ANNOTATION

Costru-Tasnic Elena, „Study of potential biomarkers for predicting haemorrhagic transformation of cerebral infarction", doctoral thesis in medical sciences, Chisinau, 2024

Thesis structure: introduction, 3 chapters, synthesis of the obtained results, general conclusions, practical recommendations, 112 pages of main text, a bibliography index with 224 sources. Illustrative material includes 42 figures, 10 tables, and 5 annexes. The obtained results were published in 24 scientific papers and have been presented at 14 national and international scientific forums.

Keywords: acute stroke, haemorrhagic transformation, risk factors, biomarkers, prediction scores, blood-brain barrier integrity, matrix metalloproteinases, prognosis of neurological recovery.

Thesis field of study: neurology.

The aim of the research: study of risk factors, clinical-biochemical correlations, and relevance of some clinical and paraclinical biomarkers in the prognosis of haemorrhagic transformation of acute ischemic stroke.

Objectives of the research: 1. Evaluation of clinical risk factors for haemorrhagic transformation of cerebral infarcts and calculation of disease severity scores: *NIHSS* (National Institutes of Health Stroke Scale), *SPAN-100* (Stroke Prognostication Using Age and NIHSS), *THRIVE* (Totalled Health Risks in Vascular Events), to highlight the persons with negative prognosis and increased risk for haemorrhagic transformation of ischemic stroke; 2. Establishing the role of the *ASPECTS* score (Alberta Stroke Program Early CT Scale) in the prognosis of haemorrhagic transformation of cerebral ischemic stroke, which would allow the early identification of haemorrhagic transformation cases; 3. Determination of the correlations between the MMP-2 and MMP-9 plasma levels and the risk of haemorrhagic transformation in ischemic stroke patients; 4. Assessment of the correlation between standard laboratory parameters determined in ischemic stroke patients (plasma level of fibrinogen, INR, total cholesterol, LDL-cholesterol, HDL-cholesterol, triglycerides, glucose, leukocytes, neutrophils, lymphocytes, platelets) and the risk of developing haemorrhagic transformation of cerebral infarction; 5. Evaluation of the ischemic stroke patient's functional independence, according to the *mRS* scale (modified Rankin Scale), at discharge and 3 months after the disease onset.

Scientific novelty and originality of the research: a prospective clinical study was conducted with a comprehensive correlational analysis of multiple clinical and paraclinical risk factors (including imaging and laboratory - specific/nonspecific), as well as clinical severity assessment scores (clinical and imaging), in patients with acute cerebral infarction with the aim of establishing the relationship with the risk for haemorrhagic transformation (HT), as well as with the evolution of neurological recovery at discharge and 3 months after the ischemic cerebrovascular event. Biomarkers of blood-brain barrier integrity impairment (MMP-2, MMP-9) and their correlation with the post-stroke patient evolution (HT, unfavourable evolution) were studied.

Scientific problem solved in the study: the research highlighted the risk factors for HT in patients with acute ischemic stroke, which will allow the stratification of the risk for this complication, otherwise determining the optimization of therapeutic measures and post-stroke secondary prophylaxis. Clinical, imaging, and laboratory prognostic factors of patients with acute cerebral infarction were identified, with recommendations for in-hospital management and prevention.

Theoretical importance: evaluation of clinical, imaging, and biochemical characteristics in patients with acute ischemic stroke as risk factors for haemorrhagic transformation and unfavourable clinical evolution. The study provided additional information regarding the application of clinical, imaging, and laboratory biomarker scores in the prognosis of complications through HT in acute cerebral infarctions.

Applicative value: the results of the study encourage the systematic application of clinical scores (NIHSS, SPAN-100, THRIVE) and imaging scores (ASPECTS), along with laboratory biomarkers of blood-brain barrier integrity, for stratifying patients with acute cerebral infarction at increased risk for unfavourable evolution, including HT. This will lead to more active monitoring of these patients and limit post-stroke complications.

Implementation of scientific results: the study results have been implemented in the clinical activity of the „Diomid Gherman" Institute of Neurology and Neurosurgery, Neuroemergency section, as well as in the teaching activity of the Neurology Department nr. 1 of the „Nicolae Testemitanu" SUMPh. Based on the thesis results, an Innovation Certificate, nr. 6101 of June 27, 2023, was obtained for the innovation entitled „Application of clinical scores NIHSS, SPAN-100, and THRIVE in identifying patients with cerebral infarction at increased risk for haemorrhagic transformation".

АННОТАЦИЯ

Костру-Ташник Елена, „Исследование потенциальных биомаркеров для прогнозирования геморрагической трансформации церебрального инфаркта”, диссертация доктора медицинских наук, Кишинев, 2024

Структура диссертации: введение, 3 главы, обработка полученных результатов, общие выводы, практические рекомендации, 112 страниц основного текста, библиографический указатель с 224 источниками. Иллюстративный материал включает 42 рисунка, 10 таблиц и 5 приложений. Полученные результаты опубликованы в 24 научных статьях и были представлены на 14 национальных и международных научных форумах.

Ключевые слова: острый инсульт, геморрагическая трансформация, факторы риска, биомаркеры, факторы прогнозирования, целостность гематоэнцефалического барьера, матричные металлопротеиназы, прогноз неврологического восстановления.

Область исследования: неврология.

Цель: изучение факторов риска, клинико-биохимических корреляций и актуальности клинических и параклинических биомаркеров в прогнозировании геморрагической трансформации острого ишемического инсульта.

Цели исследования: 1. Оценка клинических факторов риска для геморрагической трансформации церебральных инфарктов и расчет оценочных шкал тяжести заболевания: NIHSS, SPAN-100, THRIVE, для выявления лиц с отрицательным прогнозом и повышенным риском для геморрагической трансформации церебральных инфарктов; 2. Установление роли шкалы ASPECTS в прогнозировании геморрагической трансформации ишемических инсультов, что позволит ранее выявлять случаи геморрагической трансформации; 3. Определение корреляций между плазмотическим уровнем MMP-2 и MMP-9 у пациентов с ишемическими инсультами и риском геморрагической трансформации церебральных инфарктов; 4. Оценка корреляции стандартных лабораторных параметров у пациентов с ишемическим инсультом с риском геморрагической трансформации церебрального инфаркта; 5. Оценка уровня функциональной независимости, согласно шкале mRS (модифицированная шкала Рэнкина), у пациентов с ишемическими цереброваскулярными событиями, с и без геморрагической трансформации, при выписке и через 3 месяца заболевания.

Научная новизна и оригинальность: был проведен комплексный корреляционный анализ множественных клинических, параклинических (изображений и лабораторных – специфических / неспецифических) и клинических оценочных шкал тяжести у пациентов с острым церебральным инфарктом для установления связи с риском геморрагической трансформации, а также с неврологическим восстановлением при выписке и через 3 месяца после церебрального ишемического события. Были изучены биомаркеры нарушения целостности гематоэнцефалического барьера (MMP-2, MMP-9) и их корреляция с результатами пациентов после инсульта (геморрагическая трансформация, неблагоприятный исход).

Решенная научная проблема: выявление факторов риска геморрагической трансформации у пациентов с острым ишемическим инсультом, что позволит стратифицировать риск данного осложнения, оптимизируя тем самым терапевтические мероприятия и вторичную профилактику послеинсультного периода.

Теоретическая значимость: исследование предоставило дополнительную информацию о применении клинических, радиологических и лабораторных биомаркеров для прогнозирования осложнения геморрагической трансформации острого церебрального инфаркта.

Прикладная ценность: результаты исследования стимулируют систематическое применение клинических шкал (NIHSS, SPAN-100, THRIVE) и радиологической шкалы (ASPECTS), анализируемых для стратификации пациентов с острым церебральным инфарктом с повышенным риском неблагоприятного развития, включая геморрагическую трансформацию, что приведет к более активному мониторингу и ограничению осложнений после инсульта.

Реализация результатов: результаты исследования были внедрены в клиническую деятельность Института Неврологии и Нейрохирургии имени Диомида Германа, а также в учебную деятельность Кафедры неврологии № 1 Государственного Университета Медицины и Фармации имени Николая Тестемицану. На основе результатов диссертации было получено Свидетельство об инновации № 6101, 27 июня 2023 года, за инновацию под названием „Применение клинических шкал NIHSS, SPAN-100 и THRIVE для выявления пациентов с церебральным инфарктом с повышенным риском геморрагической трансформации”.

COSTRU-TAȘNIC Elena

STUDY OF POTENTIAL BIOMARKERS FOR PREDICTION OF HAEMORRHAGIC TRANSFORMATION OF CEREBRAL ISCHEMIC STROKE

321.05 – CLINICAL NEUROLOGY

Summary of the doctoral thesis in medical sciences

Aprobat spre tipar: 17.06.2024
Hârtie ofset. Tipar ofset.
Coli de tipar: 2,0

Formatul hârtiei: 60x84 1/16
Tiraj 20 ex.
Comanda nr. 22

SRL S.C. TIPOGRAFIA NR.1
MD-2001, mun. Chişinău, sect. Centru,
str. 31 August 1989, 46, ap.(of.) 9,
Tel. +373 69104435, +373 79471245