UDC 616.398:616-083.98
DOI http://doi.org/10.30978/GS-2025-3-29

ISSN 2786-5584 PRINT
ISSN 2786-5592 ONLINE

Association between rs571312 MC4R and rs3810291 TMEM160 single-nucleotide polymorphisms and metabolic surgery outcomes

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OBJECTIVE – to determine the association between the *SNP rs571312 MC4R* and *SNP rs3810291 TMEM160* genes and the results of metabolic surgery.

MATERIALS AND METHODS. A prospective, observational, monocentric cohort study was conducted at the Department of General Surgery No2 of Bogomolets National Medical University. A total of 112 patients with surgical indications were offered bariatric surgery. Patients who agreed to undergo bariatric surgery (n = 53) comprised the main group. Those who refused surgical treatment (n = 59) formed the control group. The main group underwent bariatric interventions, including laparoscopic gastric bypass and laparoscopic sleeve gastrectomy, whereas patients with BMI ≥ 50 kg/m^2 had a two-stage surgical treatment involving intragastric balloon placement for a period of 6 months, followed by gastric bypass within 14 days after balloon removal. The results for both groups were evaluated one year after the start of treatment. In the control group, 59 patients suffering from obesity received conservative treatment. Before the start of treatment, all patients underwent anthropometric examinations and analysis of buccal epithelial scrapings. Genetic studies included DNA collection and isolation, as well as genotyping to determine polymorphisms of the *rs571312 MC4R* and *rs3810291 TMEM160* genes.

RESULTS. A strong correlation was found between bariatric surgery outcomes and the presence of *rs571312 MC4R* and *rs3810291 TMEM160* gene polymorphisms in the study group ($r = 0.622$; $p \geq 0.001$). There was no significant association between the degree of gene dominance and bariatric surgery outcomes ($r = 0.112$, $p \geq 0.5$).

CONCLUSIONS. In the studied cohort, a relationship was found between the effectiveness of bariatric procedures and the presence of the *rs571312 MC4R* ($r = 0.465$) and *rs3810291 TMEM160* ($r = 0.55$) polymorphisms, as well as the concurrent presence of both polymorphisms ($r = 0.622$). The detection of these polymorphisms is associated with bariatric surgery outcomes, regardless of the degree of their genetic dominance ($r = 0.467$).

KEYWORDS
treatment, excess weight loss, gene, polymorphisms.

ARTICLE • Received 2025-09-25 • Received in revised form 2025-10-13 • Published 2025-10-30

Nowadays, metabolic surgery is the safest and most effective method of obesity treatment in most countries throughout the world. According to the IFSO 8th Global Registry Report, 480,970 bariatric surgeries were performed worldwide in 2023.

The study of polymorphisms of certain genes has considerably improved disease prediction in many cases. To date, a significant number of studies have proven that genetic factors play a key role in the development of obesity. However, it remains insufficiently studied whether certain genetic factors affect the process of obesity treatment and its results.

According to the literature data, there is a relationship between obesity and the detection of polymorphisms of certain genes.

Lu Liu et al. carried out a meta-analysis involving 11,569 patients with stage III obesity and identified polymorphisms of the *FTO, TMEM18, NRXN3, MC4R, SEC16B, GNPDA2, TNNI3K, QPCTL,* and *BDNF* genes in 7,334 (63.4 %) patients, indicating an association between polymorphisms of these genes and obesity [9].

M. C. Y. Ng et al. studied the association between single-nucleotide polymorphisms of the *MC4R* gene and obesity in a multicentric study involving 8,860 patients. It was determined that the *MC4R* gene is a codon for melanocortin receptor proteins, which are agonists of leptin, a hormone that acts as a regulator in the circuit of humoral regulation of hunger and satiety. As a result, 59.1 % (5,236) of obese patients demonstrated an association between the detection of *MC4R* gene polymorphisms and obesity [12].

M. Bandstein et al. identified 7 single-nucleotide polymorphisms: *PTBP2, NUDT3, TFAP2B, ZNF608, MAP2K5, GNPDA2*, and *MTCH2* associated with obesity in 238 obese patients who underwent laparoscopic gastric bypass and found an 11 % higher percentage of excess body weight loss (%EWL) in patients who had at least one of the seven single-nucleotide polymorphisms compared to patients who did not have *SNP*s in the indicated genes in their genome [3].

S. Z. Lutz et al. identified a correlation between the presence in the genome of obese patients of single nucleotide polymorphisms (*SNP*s) *rs2235543, rs12565406*, and *rs4844880* of the HSD11B1 gene gene, which is a regulator of fat metabolism in liver cells) and non-alcoholic fatty liver disease in obese patients (r = 0.64) [11].

Thus, there is a certain relationship between the detection of gene polymorphisms, namely *SNP rs571312* of the *MC4R* gene and *SNP rs3810291* of the *TMEM160* gene, and the occurrence of obesity, regardless of gender and race. However, the relationship between these polymorphisms and obesity treatment outcomes has not yet been investigated and is extremely relevant. Therefore, this study aimed to determine the association between the *rs571312 MC4R* and *SNP rs3810291 TMEM160* gene polymorphisms and obesity treatment outcomes.

Objective – to determine the association between the *MC4R SNP rs571312* and *SNP rs3810291 TMEM160* genes and the results of metabolic surgery.

Materials and methods

The study method is a prospective, observational, monocentric cohort study conducted at the Department of General Surgery No2 of Bogomolets National Medical University.

A total of 112 patients were included in the study (clinical characteristics of the patients are presented in Table 1). All patients had indications for surgical treatment determined by International Federation for the Surgery and Other Therapies for Obesity (IFSO), namely body mass index (BMI) ≥ 35 kg/m^2 regardless of the presence and severity of comorbidities or BMI 30.0–34.99 kg/m^2 and the presence of concomitant comorbidities. Patients included in the study were offered metabolic surgery. 53 patients who agreed to undergo the bariatric procedure were included in the main group. 59 patients who refused surgical treatment and underwent conservative treatment formed the control group. The study result was evaluated one year after the start of treatment. The main group underwent bariatric interventions, including laparoscopic gastric bypass (n = 20; 37.7 %) and laparoscopic sleeve gastrectomy (n = 17; 32.2 %). Patients with BMI ≥ 50 kg/m^2 (n = 16; 30.1 %) had a two-stage surgical treatment. The first stage involved placing an intragastric balloon for 6 months, followed by gastric bypass within 14 days after balloon removal. The final result was assessed 12 months later. The control group consisted of 59 obese patients treated conservatively, namely with diet therapy, psychological support sessions, lifestyle correction, and dosed physical activity.

Before treatment, both groups underwent anthropometric measurements, routine general clinical observations, and analysis of buccal epithelium scrapings. Genetic research included DNA collection, isolation, and genotyping. DNA isolation was performed using reagents from the DNA kit (NeoPrep DNA, Ukraine). The manipulations were

Table 1. **Clinical charasteristics of the patients**

Parameter	All the patients (n = 112)	Main group (n = 53)	Control group (n = 59)
Age, years	48 ± 9 (27–68)	48 ± 9 (23–68)	49 ± 10 (29–67)*
Body mass, kg	144.8 ± 15.7 (110.1–210.2)	148.0 ± 18.4 (110.1–210.2)	141.8 ± 12.2 (117.2–172.2)*
Height, cm	166 ± 13 (150–188)	165 ± 12 (152–188)	166 ± 13 (150–185)
Initial BMI, kg/м^2	45.1 ± 6.1 (35.2–75.3)	48.6 ± 7.4 (35.2–75.3)	41.6 ± 3.1 (35.5–50.5)*
Ideal body mass, kg	62.2 ± 7.2 (51.5–74.5)	62.6 ± 7.1 (52.8–73.3)	61.2 ± 6.8 (51.1–74.5)*
Excess weight, kg	100.2 ± 19.0 (65.9–144.6)	101.5 ± 22.3 (70.5–144.6)	99.8 ± 18.5 (62.3–132.7)*

Note. The difference between the main and the control groups for all parameters is statistically insignificant (p > 0.05).
* In one or both groups, the data distribution differed from normal; the analysis was performed using the Wilcoxon T-test.

carried out in accordance with the protocol and step-by-step instructions provided with the kit.

All DNA samples were genotyped using allelic discrimination analysis on a Fast Real-Time PCR device (System 7500, USA) using TaqMan probes. For the isolation of *SNP rs571312 MC4R* and *SNP rs3810291 TMEM160*, pre-assembled TaqMan® genotype assays (Applied Biosystems, USA) were used.

To assess treatment outcomes, BMI parameters and the percentage of excess weight loss (%EWL) were used.

Results

Before treatment, the main group had an average BMI of 48.6 ± 20.1 kg/m^2, while the control group had an average BMI of 41.6 ± 16.0 kg/m^2. After treatment, the average BMI in the main group reduced to 29.5 ± 5.4 kg/m^2, but in the control group it decreased insignificantly to 40.2 ± 13.8 kg/m^2 (Table 2).

After 12 months of observation, the main group's average %EWL was 59.21 % ± 23 %. The highest %EWL was 77.2 %, while the lowest %EWL was 49.9 %. The control group had a significantly lower average %EWL – 9.92 ± 7.25 %. The interval between the highest and lowest %EWL was 19.45 % and 2.2 %, respectively. The average %EWL in obese patients varied depending on the surgical treatment method used. Patients who underwent Roux-en-Y gastric bypass had the highest average %EWL – 69.71 ± 20.0 %, while those who underwent sleeve gastrectomy had an average %EWL of 51.57 ± 5.9 %. After two-stage surgical treatment, patients had an average %EWL of 51.7 ± 7.92 %.

During genotyping analysis, we revealed the distribution of single-nucleotide polymorphisms *rs571312* of the *MC4R* gene and *rs3810291* of the *TMEM160* gene in the main and control groups.

The distribution of results by the presence of polymorphisms of the genes studied had the following characteristics. Among all patients included in the study (n = 112), the studied single-nucleotide polymorphisms were detected in 23.2 % (n = 26), and the remaining 76.8 % (n = 86) of patients did not have *SNP rs571312 MC4R* and *SNP rs3810291 TMEM160* in the studied samples. The distribution of single-nucleotide polymorphism detection by patient groups is presented as follows. The *rs571312 MC4R* polymorphism was detected more often in the main group – 19.5 % (n = 10, where n is the number of patients) than in the control group – 14.6 % (n = 8) (p = 0.007). The *rs3810291 TMEM160* polymorphism was detected more often in the main group – 9.75 % (n = 5) than in the control group – 5 % (n = 3), (p < 0.01). The presence of both polymorphisms was found only in the main group – 7.3 % (n = 3) (Table 3).

The distribution of genotype dominance was significant (Fig. 1). The G/G genotype of the *SNP rs571312* polymorphism of the *MC4R* gene was detected in 15 (83.4 %) patients with detected polymorphisms (p ≤ 0.01). The T/G genotype of the *SNP rs571312* polymorphism of the *MC4R* gene was detected in 3 (16.6 %) individuals out of all patients in whom this polymorphism was observed

Table 2. **BMI dynamics in both groups depending on the treatment method, kg/m^2**

Treatment method	Before treatment	After treatment	p
Roux-en-Y Gastric bypass	47.8 ± 10.1 (35.2–75.3)	28.5 ± 6.2 (26.2–30.1)	< 0.001
Sleeve gastrectomy	42.2 ± 7.4 (35.4–48.6)	30.1 ± 5.5 (29.2–31.0)	< 0.001
Intragastric balloon + RYGB	52.2 ± 6.4 (50.1–54.4)	28.3 ± 6.0 (26.1–30.5)	< 0.001
BMI in the main grop	48.6 ± 20.1 (35.2–75.3)	29.5 ± 5.4 (26.1–31.0)	< 0.001
Conservative treatment	43.6 ± 16.2 (35.5–50.5)	40.2 ± 13.8 (33.4–47.0)	0.06

Table 3. **Distribution of single nucleotide polymorphism detection by patient groups**

Group	SNP rs571312 MC4R	SNP rs3810291 TMEM 160	Dual polymorphism presence	Polymorphism absence	p
Main (n = 53)	10 (19.5 %)	5 (9.75 %)	3 (7.3 %)*	38 (70.75 %)	0.007
Control (n = 59)	8 (14.6 %)	3 (5.0 %)	0	48 (82.97 %)	< 0.01

Note. * n = 3 among patients in whom both polymorphisms were detected (n = 26).

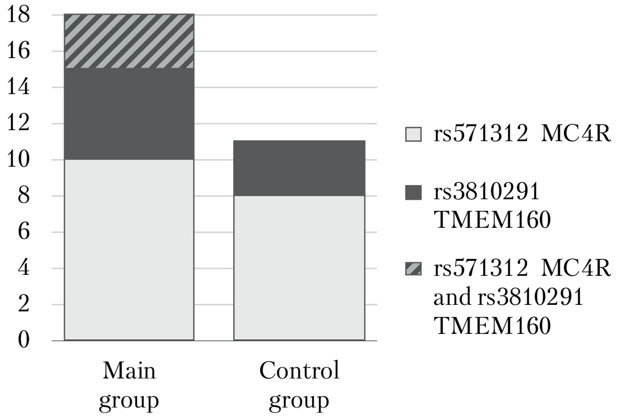

Figure 1. **Distribution into study groups depending on the available gene polymorphism**

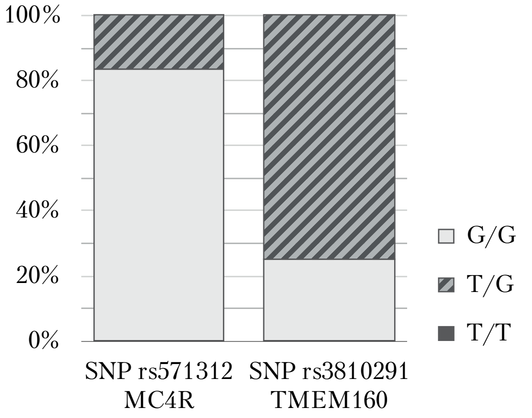

Figure 2. **Distribution of the genetic dominance**

(p = 0.0087), and has no statistical significance. The T/T genotype was not detected in any of the patients. The G/G genotype of the *SNP rs3810291* polymorphism of the *TMEM160* gene was detected in 2 patients (25 %) out of all identified carriers of this polymorphism, and has no statistical significance (p = 0.0077). The T/G genotype of the *SNP* polymorphism *SNP rs3810291* of the *TMEM160* gene was detected in 6 carriers of this polymorphism (75 %) (p ≤ 0.01). The T/T genotype was not detected in any of the patients (Fig. 2).

During the statistical processing of the obtained data by the rank correlation method, no relationship was found between the degree of gene dominance and the effectiveness of bariatric procedures (r = 0.112). Detection of the polymorphism *rs571312* of the *MC4R* gene and *rs3810291* of the *TMEM160* gene is strongly correlated (r = 0.467; p ≤ 0.05) with the effectiveness of surgical treatment of obesity, regardless of the degree of their genetic dominance.

In the main group, patients with single-nucleotide polymorphisms in their genome that were the subject of this research had an average %EWL of 60.47 ± 3.4 % and a decrease in BMI by 40.4 ± 15.4 % compared to the initial one 12 months after surgery. In the control group, such patients demonstrated a significantly lower average %EWL – 11.5 ± 1.5 % – and a decrease in BMI by 7.8 ± 2.1 % after 12 months of conservative therapy, which was not statistically significant.

Treatment outcomes were distributed by patient group as follows: average %EWL, BMI decrease, and the presence of single- nucleotide polymorphisms of the genes *rs571312 MC4R* and *rs3810291 TMEM160*. In the main group, patients with none of the studied *SNP*s (n = 38) had an average treatment efficacy index for %EWL of 52.9 ± 2.7 % and a BMI decrease of 33.3 ± 9.8 % 12 months after treatment. However, patients in the control group (n = 48) achieved a %EWL of 10.2 ± 1.1 % and a BMI decrease of 7.5 ± 1.1 % after conservative therapy. In the main group, patients with the *SNP rs571312*

Table 4. **Treatment outcomes by average percentage of excess weight loss, BMI reduction, depending on gene polymorphisms**

Gene polymorphism	**Main group**		**Control group**	
	%EWL, %	**BMI decrease, %**	**%EWL, %**	**BMI decrease, %**
No SNP	52.9 ± 2.7	33.3 ± 9.8	10.2 ± 1.1	7.5 ± 1.1
SNP rs571312 MC4R	55.4 ± 0.8	47.5 ± 12.2	9.96 ± 1.25	8.1 ± 1.4
SNP rs3810291 TMEM160	55.7 ± 1.4	45.5 ± 8.5	10.5 ± 1.3	9.1 ± 1.5
Dual SNP presence	68.4 ± 4.2	50.1 ± 10.2	0	0
p	≤ 0.05*		< 0.01*	

Note. * In one or both groups, the data distribution differs from normal; a comparison was performed using the Wilcoxon T-test. Comparison was performed between patients of the main and control groups.

MC4R ($n = 10$) had an average treatment efficacy index for %EWL of 55.4 ± 0.8 % and a BMI decrease of 47.5 ± 12.2 % after treatment. In the main group, patients with *SNP rs3810291 TMEM160* ($n = 5$) demonstrated an average %EWL of 55.74 ± 1.4 and a BMI decrease of 45.5 ± 8.5 %. Patients with both studied gene polymorphisms had the highest average treatment efficacy index for %EWL of 68.4 ± 4.2 % and a BMI decrease of 50.1 ± 10.2 %. In the control group, all obese patients with the studied gene polymorphisms had a low average treatment efficacy for %EWL of 11.5 ± 1.5 % and an insignificant BMI decrease of 7.5 ± 1.05 %. Patients with *SNP rs571312 MC4R* ($n = 8$) had %EWL indicators of 9.96 ± 1.25 % and a BMI decrease of 8.1 ± 1.4 %. Patients with *SNP rs3810291 TMEM160* ($n = 3$) demonstrated an average %EWL of 10.5 ± 1.3 % and a BMI decrease of 9.1 ± 1.5 % after treatment. In the control group, no patients had both studied gene polymorphisms (Table 4).

Statistical analysis using the method of linear correlation revealed a positive correlation ($r = 0.467$; $p \leq 0.05$) between the detection of polymorphisms *rs571312* of the *MC4R* gene and *rs3810291* of the *TMEM160* gene and treatment efficacy for %EWL, and a weak correlation ($r = 0.191$) between the absence of the studied polymorphisms in the sample and treatment efficacy for %EWL. A correlation was also found between the detection of *SNP rs571312* of the *MC4R* gene in the study group and the effectiveness of the treatment by %EWL ($r = 0.443$). A correlation was found between the presence of *SNP rs3810291* of the *TMEM160* gene in the main group and treatment efficacy for %EWL ($r = 0.5$). A strong correlation was noted between the presence of both studied polymorphisms in the main group and treatment efficacy for %EWL ($r = 0.608$). The presence of the studied polymorphisms in the control group does not affect treatment efficacy for %EWL ($r = 0.097$).

During the statistical processing of the obtained data using the rank correlation method, no relationship was found between the degree of gene dominance and the effectiveness of bariatric procedures ($r = 0.112$; $p \leq 00.5$), which indicates that the detection of polymorphisms *rs571312* of the *MC4R* gene and *rs3810291* of the *TMEM160* gene is strongly correlated ($r = 0.467$; $p \leq 0.05$) with the effectiveness of surgical treatment of obesity, regardless of the degree of their genetic dominance.

Discussion

During the course of the study, some patterns were identified. The polymorphisms that were the subject of this research, including *SNP rs571312* of the *MC4R* gene and *SNP rs3810291* of the *TMEM160* gene, were investigated to assess their association with metabolic surgery outcomes.

P. K. Thanos et al. in their study analyzed the genetic risk score in patients who underwent bariatric surgery, with the aim of using it as a predictive factor for weight loss parameters one year after surgery. Thirty patients were assessed using the Genetic Obesity Severity Scale (GARS), which analyzes neurogenetic polymorphisms associated with food addiction. Genetic and psychosocial data collected before surgery were compared with weight loss data, including weight change, BMI, and %EWL [16].

I. Ntalla et al. found that several genetic loci were associated with BMI and obesity. The aim of the study was to examine the impact of known loci associated with BMI in adults. For this purpose, 34 variants were selected for study among 7,787 adolescents of Greek origin. The cumulative impact of the variants was assessed by calculating a genetic risk score (GRS-34) for each participant. Variants at the FTO, *TMEM160*, FAIM2, RBJ, ZNF608, and QPCTL loci demonstrated nominal evidence of association with BMI and/or risk of overweight ($p < 0.05$) [15].

The results showed correlations between the «risk» alleles of individual genes, weight data after 1 year, and psychosocial indicators. Spearman correlation showed that the OPRM1 gene polymorphism (rs1799971) had a significant negative correlation with weight at 1 year ($r_s = -0.4477$, $p < 0.01$) and BMI ($r_s = -0.4477$, $p < 0.05$). In addition, the risk allele of the DRD2 gene (rs1800497) had a negative correlation with BMI at 1 year ($r_s = -0.4927$, $p < 0.05$), indicating that the presence of one copy of the «risk» allele was associated with lower BMI. However, this allele had a positive correlation with weight change ($r_s = 0.4077$, $p < 0.05$) and %EWL ($r_s = 0.5521$, $p < 0.05$) at 1 year after surgery. This suggests that people with a higher genetic risk of obesity are more likely to have better outcomes in obesity treatment, especially in the case of the DRD2 polymorphism.

Therefore, research on the association between genetic factors and surgical treatment efficacy for obesity is extremely relevant.

Conclusions

An association was found in the studied group of patients between the metabolic surgery outcomes and the presence of polymorphism of the genes *rs571312 MC4R* ($r = 0.465$); *rs3810291 TMEM160* ($r = 0.55$;); and the presence of both polymorphisms ($r = 0.622$; $p \leq 0.05$).

The degree of gene dominance has no relationship with bariatric surgery outcomes ($r = 0.112$).

The detection of polymorphisms *rs571312* of the *MC4R* gene and *rs3810291* of the *TMEM160* gene is associated ($r = 0.467$; $p \leq 0.05$) with the metabolic surgery outcomes, regardless of the degree of their genetic dominance.

Declaration of interests

The author declares no competing financial interests.

This study was made as a part of dissertational work for achievement of PhD grade.

Funding. This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

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Асоціація однонуклеотидних поліморфізмів генів rs571312 MC4R та rs3810291 TMEM160 з результатами метаболічної хірургії

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Мета — визначити зв'язок генів *SNP rs571312 MC4R* та SNP *rs3810291 TMEM160* з результатами метаболічної хірургії.

Матеріали та методи. Проспективне обсерваційне когортне моноцентричне дослідження проведене на кафедрі загальної хірургії № 2 Національного медичного університету імені О. О. Богомольця за участю 112 пацієнтів із показаннями до хірургічного лікування, яких розділено на дві групи. Основну групу становили 53 пацієнти, які погодилися на баріатричну процедуру, контрольну — 59 пацієнтів, які відмовилися від хірургічного лікування та отримали консервативне лікування. Результати оцінювали через рік після початку лікування. В основній групі було проведено такі баріатричні втручання: лапароскопічне шунтування шлунка, лапароскопічну рукавну резекцію шлунка, пацієнтам з індексом маси тіла ≥ 50 кг/м^2 — двохетапне хірургічне лікування (установлення внутрішньошлункового балона на 6 міс і шунтування шлунка протягом 14 днів після видалення балона). До початку лікування проводили антропометричні обстеження та дослідження зішкрібків букального епітелію. Генетичні дослідження передбачали виділення ДНК, а також генотипування для визначення поліморфізмів генів *rs571312 MC4R* і *rs3810291 TMEM160*.

Результати. Виявлено кореляцію між позитивними результатами баріатричних процедур і наявністю поліморфізмів генів *rs571312 MC4R* та *rs3810291 TMEM160* ($r = 0{,}622$, $p \geq 0{,}001$). Ступінь домінування генів не мав зв'язку з результатами баріатричної хірургії.

Висновки. У досліджуваній групі пацієнтів виявлено зв'язок між ефективністю проведених баріатричних процедур та наявністю поліморфізмів генів *rs571312 MC4R* ($r = 0{,}465$); *rs3810291 TMEM160* ($r = 0{,}55$); наявністю обох поліморфізмів ($r = 0{,}622$). Виявлення досліджуваних поліморфізмів пов'язане з результатами баріатричної хірургії, незалежно від ступеня їх генетичної домінантності ($r = 0{,}467$).

Ключові слова: лікування, втрачена надлишкова маса тіла, ген, поліморфізми.

FOR CITATION

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