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Annotation of the dissertation
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«Relationship with vitamin D level and VDR gene polymorphism in adolescent girls with primary dysmenorrhea»

Educational program 8D10102 - "Medicine"

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ANNOTATION of the dissertation work of DONAYEVA AINUR YERGALIKYZY, Doctor of Philosophy (PhD) under the Educational program 8D10102 - "Medicine" entitled "Relationship with vitamin D level and VDR gene polymorphism in adolescent girls with primary dysmenorrhea"
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RELEVANCE

The future of the nation is determined, first of all, by the health of adolescents and their fertility. The state of reproductive health of adolescents is currently a matter of discussion not only among specialists, but also in society. The period of puberty, as the period of formation of sexual maturity of the body, takes an important place in the formation of the reproductive system of every woman and in many cases is directly related to the harmony and development of girls during the period of puberty. The review data of the last ten years shows that in addition to clearly unfavorable trends in health indicators, significant changes have been observed among adolescent girls, which are often associated with various factors such as unbalanced physical activity, chronic stress, and illiteracy of the population regarding their own health.

An important marker of the reproductive activity of adolescent girls is the characteristics of the menstrual cycle. It is an integral indicator of reproductive system, somatic health, endocrine system, mechanisms of regulation of menstrual cycle activity, mental health and social well-being.

According to WHO data, about 94% of adolescent girls aged 10 to 20 years experience pain during menstruation, and in 15% of cases it leads to reduced social activity and loss of work ability. According to foreign studies, the prevalence of primary dysmenorrhea among adolescents is from 8 to 90%, and in Europe and the USA it is 65-85%, similar studies in Asian countries showed a high frequency of this disease up to 90% [3-5]. Variations in the frequency of occurrence in different regions are due to the lack of standardized approaches and ethnic characteristics.

The diagnosis of primary dysmenorrhea N94.4 (BD) in teenage girls during puberty in the Republic of Kazakhstan as a functional disorder without organic changes is carried out according to the diagnostic and treatment criteria of pediatric and adolescent gynecologists according to the national clinical protocol. According to the data of domestic researchers, 12% of teenagers were treated by a pediatric gynecologist with BD.

Violation of uterine contractile activity as a result of excessive release of prostaglandins is a proven finding of primary dysmenorrhea. Regarding the role of vitamin D, it has been found that there is a strong correlation between menstrual irregularities in girls and vitamin D deficiency at the age of puberty and various other factors that determine a wide range of clinical manifestations. Vitamin D may reduce the production of proglastaglandins, derivatives of arachidonic acid, during uterine contractions in several ways. The level of 25(OH)D decreases in the luteal phase, as estradiol is degraded in the liver by 1-α-hydroxylase and 24-

hydroxylase, reducing the level of 25(OH)D3. Ovarian hormones peak during the luteal phase and cause vitamin D levels to drop. As a result, the cyclic disturbance of vitamin D metabolism with ovarian hormones leads to the appearance of symptoms of primary dysmenorrhea in adolescent girls. In the literature available to us, we found only a few studies on the genetic determination of BD. Genes responsible for the production of interleukins were found to be associated with BD. Of interest is the association between different genotypes/allele frequencies, VDR rs7975232, rs1544410, rs2228570, rs731236 and haplotypes with inflammatory diseases.

The high frequency of primary dysmenorrhea in adolescent girls, multifactorial nature, variety of mechanisms of pathogenesis and clinical manifestations indicate the relevance of this problem and the need for further research.

Research purpose:

Evaluation of the association between vitamin D, estradiol and progesterone hormone levels and the VDR (rs731236) gene polymorphism in primary dysmenorrhea in adolescent girls.

Research objectives:

1. Assessment of the level of vitamin D, estradiol and progesterone hormones of the studied adolescent girls.
2. Analysis of the frequency of VDR gene rs731236 genotypes according to the level of vitamin D in adolescent girls diagnosed with primary dysmenorrhea.
3. Determination of correlation between vitamin D level and VDR gene polymorphism during primary dysmenorrhea.

Scientific news:

- As part of the research, for the first time in Kazakhstan, the level of vitamin D and the frequency of VDR gene polymorphism were determined among adolescent girls diagnosed with primary dysmenorrhea;
- The association between vitamin D level and VDR gene polymorphism was determined among adolescent girls diagnosed with primary dysmenorrhea.

Theoretical and practical importance:

To evaluate the predictors of primary dysmenorrhea among adolescent girls, a probabilistic statistical modeling model was developed with the blood vitamin 25(OH)D receptor VDR gene.

During the protocol No. 109 of July 30, 2020, approved by the Ministry of Health of the Republic of Kazakhstan, the Joint Commission on Healthcare Quality, "For the diagnosis of primary dysmenorrhea", it is possible to consider the determination of the level of vitamin D as an additional criterion.

Many of the problems associated with menstrual disorders are related to the sequential physiological events of puberty and can be effectively addressed through practical health care management.

To protect the reproductive health of adolescents - early diagnosis allows to maintain and control the level of vitamin D during the sexual development of the growing adolescent body.

Rules issued for protection:

1. Identification of genetic polymorphisms affecting vitamin D levels allows early diagnosis of primary dysmenorrhea in adolescent girls prone to vitamin D deficiency.

2. The results of the VDR gene polymorphism in the development of primary dysmenorrhea in teenage girls with high probability.

3. Early diagnosis of the course of primary dysmenorrhea based on vitamin D deficiency prevents the development of secondary dysmenorrhea (endometriosis) among adolescent girls and allows prevention.

Approbation of work.

The main principles of the dissertation work were presented at extended meetings of the Scientific Council and the Scientific Organization of West Kazakhstan Marat Ospanov Medical University.

Research results were presented at scientific and practical conferences:

- At the international scientific conference of students and young scientists "Science: yesterday, today, tomorrow" "Clinical case describing mineral density and bone metabolism in an adolescent girl diagnosed with primary dysmenorrhea" (April 29, 2021 WKMU named after M.Ospanov);

- At the international scientific-practical conference "Modern medicine: new approach and current research" between near and far foreign medical educational institutions of the Republic of Kazakhstan, organized on the occasion of the World Day against Osteoporosis, "Polymorphism of the vitamin D receptor gene VDR and mineral density of bone tissue in Asian girls" adolescents with primary dysmenorrhea" and "Peculiarities of sexual development in adolescents with primary dysmenorrhea" Lithuania, Kaunas Medicine volume 57, Supplement 2, 2021 (February 20, 2021, Aktobe);

- At the VIII international scientific conference of young scientists and students entitled "Future of development of biology, medicine and pharmacy" in online format, the Science Council under the Nursultan Nazarbayev Foundation JSC "South Kazakhstan Academy of Medicine" "Prevalence of genes VDR, COL1A1, CALCR among adolescent girls with primary dysmenorrhea" (December 9-10, 2021);

- "Vitamin D deficiency during primary dysmenorrhea" at the II International Conference organized by the "Kazakhstan Association for Sexual and Reproductive Health (KMPA)" (April 7-8, 2023, Almaty).

Publications on the topic of the dissertation.

11 scientific publications were published on the topic of the dissertation. Among them - 3 works in international scientific and practical data, 3 articles in international journals indexed in the Web of Science and Scopus information

bases, 2 article in scientific publications recommended by the Committee for Quality Assurance in the Field of Education and Science of the Republic of Kazakhstan, data in the state register of rights to objects protected by copyright. implementation - 2, acts for the introduction of scientific research results into clinical practice - 1, a patent of the Republic of Kazakhstan for a useful model - 1 was received.

Personal contribution of the author.

Development of the idea, purpose and objectives of the scientific research, collection of data and consent to conduct clinical, laboratory and genetic research, statistical processing of research results, formulation of conclusions and recommendations, articles and theses were written.

RESEARCH OBJECT AND METHODS

Scientific and technical project funded by a grant of scientific and (or) scientific and technical projects under the main research work 2021-2023 program (12 months): AP09563004 "Features of bone mineral density and metabolism in teenage girls with primary dysmenorrhea", registration number 0121KP00549 and Within the framework of the 2021-2023 grant program established at the level of the West Kazakhstan Marat Ospanov Medical University, it was carried out in the Department of Normal Physiology as a fragment of the scientific and technical project "The state of mineral density and metabolism of adolescent girls with primary dysmenorrhea in the Kazakh population".

The research work was approved by protocol No. 10 of October 4, 2022 of the local ethical committee of West Kazakhstan Marat Ospanov Medical University. The clinical part of this scientific study was carried out at the Regional Perinatal Center of JSC "Consulting-Diagnostic Center", and the laboratory part was carried out on the basis of the "Invitro" laboratory located at the address of Aktobe city, Abilkayr Khan Ave. 89 and 12 microdistrict.

Research design: Case-control design.

Object of the study: According to the case-control design of the study, the study sample was adolescent girls of Aktobe between the ages of 12 and 17 years 5 months and 29 days, divided into comparison groups: in the case group (n=206) - adolescent girls diagnosed with primary dysmenorrhea, and in the control group (n=204) consisted of girls without pain during the menstrual cycle. The selection of subjects was carried out by random sampling method. The total number of teenage girls was 27,972. The sample size calculation for the case-control study design was calculated using the online calculator in the Epi info TM program, the range was expressed as proportions, and the estimated odds ratio was 1.0; confidence level was 95%. Research Power: For biomedical research, this figure is typically 80%. The ratio of case group to control group was taken as 1:1. A case and a control group were grouped using a matched case-control study. Further, according to the following main factors: age, gender, body mass index, residential address, city of Aktobe and Kazakh nationality, adolescent girls were paired.

Standard anthropometric instruments were used: a stadiometer, medical scales and a plastic measuring tape. The study design used pseudorandomization (propensity score matching - PSM) to divide adolescent girls into two groups, an effective method of eliminating the influence of various factors that could bias the results when comparing groups in a study. In this study, STATA 13 statistical software used the results of confounders determined by factors such as (age, body mass index, height, weight), direction of effect of the potential confounder, perceived and actual size. 410 girls took part in the study, with no differences between the values.

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Study criteria for the control group

Criteria for inclusion in the study: adolescent girls between the ages of 12 and 17 years 5 months and 29 days, girls of Kazakh nationality, regular menstrual cycle (within 21-35 days), menarche within 1 year, according to ultrasound (abdominal) examination, genitalia of the small pelvis in the case of undiagnosed pathology, girls without signs of pain on the visual analogue scale.

Exclusion criteria: female children aged 11 years 5 months to 29 days and 17 years of age who received exogenous hormones and drugs affecting the central nervous system for 1 year when early and late menarche, anomalies and diseases of the small pelvis, neurological and psychiatric abnormalities were detected. Adolescent girls older than 5 months and 29 days, in case of detection of genital pathology of the small pelvic cavity by ultrasound (abdominal) examination, girls with a pain mark higher than 1 point on the visual analogue scale.

I clinical stage of research:

According to the main criterion of the research, an (abdominal) ultrasound examination was conducted to rule out organic changes in the pelvic cavity of adolescent girls in the clinical stage.

As a result of this study, girls voluntarily informed their consent to participate in the study, and all examined adolescent girls were referred to the adolescent gynecologist. In addition, the researchers' parents also filled out a

written consent to conduct the research, and only the respondents who received consent were involved in the research.

Complaints and complete medical history were collected during the doctor's appointment, pain intensity was described on the Visual Analogue Scale (VAS), anthropometric parameters were measured (weight, height, DSI) and a gynecological examination was performed.

Primary dysmenorrhea was determined according to the 10-point scale of VASH. This adolescent girl presented the child's degree of pain, rated from painful to completely unbearable pain (on a scale of 0-10, respectively). The rating on the scale is as follows: 1-3 points - mild pain, 4-7 points - moderate pain and 8-10 - severe pain.

II laboratory stage of research:

The level of vitamin 25(OH)D, estradiol and progesterone hormones and genetic research were carried out to determine the genotypes of the VDR gene.

An indicator of vitamin D status in the body is 25(OH)D, the main metabolite of vitamin D in the blood. In the blood test, the level of 25(OH)D was determined from the blood serum. Collected blood was transferred to a tube with a red cap, mixed vigorously 4-6 times and kept in a vertical position for 30 minutes at room temperature, then centrifuged for 10 minutes and stored at +2...+8oC. The detection method was investigated by chemiluminescent immunological analysis of microparticles.

Blood serum is the material used to study the level of progesterone hormones. The detection method is a solid-phase chemiluminescent immunoassay. The analysis is carried out on the 22nd - 23rd day of the menstrual cycle.

Estradiol hormone level was determined from blood serum. The detection method is electrochemiluminescent immunoassay (ECLIA). Biomaterial for research was submitted on an empty stomach.

Genetic research: conducted by determining the genotypes of the VDR gene.
The vitamin D receptor is located in a single nucleotide polymorphism (SNP) in the VDR gene (the VDR gene is located at the 12q12q13 locus in the human body, on chromosome 12). At the time of genetic analysis: a genetic research questionnaire, an instruction form and an informed consent were completed by each respondent. Genotyping was determined using polymerase chain reaction (PCR) and restriction analysis. Blood sampling for genetic analysis was taken only when the subject gave his consent and joined the study. Blood was drawn into a 2.0 ml vacutainer (Becton Dickinson, BD Vacutainer) of pale purple, potassium ethylenediaminetetraacetic acid (EDTA)-treated whole blood. After blood collection, the tube was carefully moved 4-6 times, stored at +2...+8°C and transported. In our study, the genotypes of the vitamin D receptor VDR gene were classified into three types: T/T, T/C, and C/C. T/T is a normal version of the polymorphism, that is, a genotype without a risk of development; T/C - finding a mutation in one of two pairs of genes, the risk of development is a heterozygous genotype; C/C- the genotype located in the mutated paired alleles and associated with the risk of polymorphism homozygosity.

Methods of statistical analysis

Collection, compilation and systematization of primary information was done in MS Excel 2016 database.
Statistical processing of the research results was carried out using the SPSS 25 program.
Normality of distribution was checked using Kolmogorov-Smirnov and Shapiro-Wilk's W-test criteria. Parameters such as arithmetic mean (M), standard error (m) and standard deviation (SD) in case of normal distribution, and median (CI), interquartile range (25th to 75th quartiles, IQR) in case the data deviate from normal distribution was calculated.
Non-parametric methods were used due to the absence of normal distribution between indicators: Mann-Whitney U-test was used to compare two groups, Kruskal-Wallis h test was used for three and more groups. Nominal data are expressed in absolute values and percentage (N (%)).
Pearson's Chi-square test was used to compare nominal data, however, Fisher's exact test was calculated when the expected phenomenon had a value of at least 10 in a cell. Generalized linear regression models for analyzing the relationship of the studied results with possible predictors: for binary results - binary logistic regression (confidence interval (CI) as an estimate of the effect size, 95% probability of the corresponding ratio (OR) (OR), for ordinal results - proportional odds model (effect (proportional likelihood ratio with 95% CI) was used as an estimate of the size.
Mixed linear regression models were used to assess the influence of predictors on the dynamics of numerical indicators, including the interaction term, the confidence level for the interaction coefficient p was estimated by calculating the Spearman (r) correlation coefficient r to determine correlation relationships between variables.

Cox and Snella coefficients were also tested using Nigelkerk's significance criterion for further practical use of the model.

The GEN-expert online calculator file:///C:/Users/Acer/Downloads/GEN-эксперт%20(1).html was used to calculate genetic statistics in case-control studies.

Decision Tree/Classification Tree is a method for studying the relationship of one dependent variable with several independent variables. In this case, the connection is carried out not in the form of a predictive equation (as in the regression method), but in the form of a tree structure. The result of using the method is to divide the sample into several subgroups, each of which demonstrates a certain relationship with the dependent variable. A decision tree is a way of representing data in a hierarchical sequential structure, where each object corresponds to one decision node.

RESEARCH RESULTS

In the course of general work, according to the "Case-control" study, first of all, a descriptive and comparative analysis of the indicators of the general physical development of adolescent girls was conducted.

The number of teenage girls who took part in the entire study was 410. Study groups were divided into case and control groups.

Case group - the number of girls diagnosed with primary dysmenorrhea is n=206, i.e. girls who have pain during menstruation on the visual analogue scale, and the control group - the number of girls who have a normal menstrual cycle is n=204.

According to the descriptive analysis of subjects in the case and control groups: the mean age was 15.2 (95% CI: 15-15.5) in the case group and 14.9 (95% CI: 14.7-15.1) in the control group , (p= 0.19); height was 159.5 (95% CI: 158.5 - 160.5) cm in the case group and 160.9 (95% CI: 160.0 - 161.9) cm in the control group, (p= 0.06) ; body weight was 53.7 (95% CI: 52.5-55) kg in the case group and 54.3 (95% CI: 53.9 - 55.7) kg in the control group (p=0.1), body weight According to the index, the average value of teenage girls in the case group is 21.1 (95% CI: 20.7-21.5), and the body weight index of girls in the control group is 21.2 (95% CI: 20.8-21.5) was (p=0.17). There was no significant difference between the two groups in terms of age, height, body weight and body mass index. The median age of menarche was found to be 11 years in both groups.

Carrying out the research task, a descriptive and comparative analysis of the status of estradiol hormone in the follicular phase of the menstrual cycle and progesterone hormones in the lutein phase was carried out between the two groups.

The median estradiol hormone level in the blood test of adolescent girls was 188.0 [109.0; 335.0] pmol/l is the average amount, and in the control group the median is 262.0 [120.0; 398.0] pmol/l, that is, the indicated medians of estradiol hormone levels were within normal limits in both groups. During the comparative analysis, it was observed that this value decreased in the condition group, and it showed that there is a relatively significant difference between the two groups (p=0.03).

The median serum progesterone hormone of the adolescent girls participating in the study had a reference value between the two groups. The median progesterone hormone in blood serum of adolescent girls in the case group was 0.7 [0.4; 2.3] nmol/l, and in the control group 0.8 [0.4; 11.4] nmol/l, the result of the comparison showed that there was no difference between the study groups in terms of the median progesterone hormone (p=0.2).

The level of vitamin 25(OH)D determined in the blood serum of all adolescent girls included in the study was 15.3 [11.6; 21.1] ng/ml. Adolescent girls in the two groups, which were further divided, also had lower levels of vitamin 25(OH)D, that is, the level of vitamin 25(OH)D of adolescent girls in the condition group was 14.6 [10.6; 19.3] ng/ml, and in the control group 15.6 [13.0; 23.2] ng/ml. These values confirm vitamin 25(OH)D deficiency <20 ng/ml. The level of determined serum vitamin 25(OH)D was 0.8ng/ml lower in the case group than in the control group according to comparison analysis, respectively, a significant difference was found between the two groups (p= 0.0004).

Vitamin 25(OH)D levels were the focus of attention among adolescent girls diagnosed with primary dysmenorrhea. Because this vitamin acts as a metabolite on the one hand, and as a regulatory hormone on the other. Therefore, the range of its values is considered important.

According to the basic analysis of vitamin 25(OH)D, depending on the level of the body, it shows sufficient, insufficient and deficit conditions. Accordingly, the descriptive analysis of the primary indicator 25(OH)D vitamin level showed the following results: the median of sufficient level (>30 ng/ml) among adolescent girls in the case group was 34 [32.0; 41.0] ng/mL; the median of insufficient level (between 20 and 30 ng/ml) was 23 [21.0; 25.0] ng/ml; deficiency level (<20 ng/ml) median 13 [10.0; 16.0] ng/ml. A further descriptive analysis showed that among teenage girls in the control group, the median level of sufficient vitamin 25(OH)D (>30 ng/ml) was 42 [31.0; 45.0] ng/mL; the median of insufficient level (between 20 and 30 ng/ml) was 23 [21.0; 27.0] ng/mL; deficiency level (<20 ng/ml) median 14 [12.0; 17.0] ng/ml. The results of the comparison of vitamin D levels between these two groups: the level of vitamin D in the control group was 1.2 times lower than that of the control group by 1.2 ng/ml and a significant difference (p<0.001) was found. Next, vitamin D deficiency level was similar between the case-control groups, so no relative difference was found (p=0.2). The level of vitamin 25(OH)D deficiency in the control group was 1 ng/ml higher than that of the adolescent girls in the case group, and a relatively significant difference (p<0.01) was found.

The next analysis was a correction analysis with primary dysmenorrhea.

Correlation analysis showed that there is a strong negative correlation between the level of vitamin 25(OH)D and the visual analog scale, which is an indicator of the pain symptom of primary dysmenorrhea in adolescent girls, r=-0.9, p<0.0001, that is, as the level of vitamin D decreases It is characterized by an increase in the visual analog scale, an increase in the menstrual pain syndrome during the menstrual cycle.

A positive weak correlation was found between serum 25(OH)D vitamin and progesterone hormone r=0.15, p<0.001. This, in turn, leads to an increased production of the serum progesterone hormone, corresponding to the phase of the menstrual cycle, as the level of vitamin 25(OH)D rises above the level of deficiency or insufficiency.

The following negative weak relationship was found between the visual analog scale and estradiol hormone, that is, the lower the level of estradiol hormone in blood serum, the more the concussion leads to an increase in the visual analog scale r=-0.1, p=0.04.

The following relationship was conducted using logistic regression analysis.

In the logistic regression analysis model, the independent indicator dependent on the occurrence of primary dysmenorrhea, the odds ratio (OR) of insufficient level of vitamin 25(OH)D was 10.1 [95% CI: 2.869 - 35.665], and the odds ratio of insufficient level of vitamin 25(OH)D (OR) 11.6 [95% CI: 3.447-38.839] sensitivity of the logistic regression analysis model showed a statistically significant direct association among adolescent girls diagnosed with primary dysmenorrhea with a 12-fold increased risk. The coefficient in the logistic regression model of this finding: false R2 Nigelkirk value was equal to 0.84, the logistic regression model for predicting primary dysmenorrhea is based on vitamin D insufficiency and deficiency.

Analysis of frequencies of alleles and genotypes of the VDR rs731236 gene.

As a result of genotyping, it was found that the T allele was found at a frequency of 35.9% (n=295) in teenage girls diagnosed with primary dysmenorrhea, while the T allele was found 1.1 times less than the frequency of 41.3% (n=338) of girls in the control group. Next, primary dysmenorrhea on the C allele was found in adolescent girls with a frequency of 14.3% (n=117), compared to the control group it showed a frequency of 8.5% (n=70) and was found to be 1.6 times less common. However, the risk allele C was more frequent in the case group.

Further analyzing the frequency of VDR rs731236 gene genotypes, the T/T genotype occurred in 43% frequency (n=107) in the case group and 57% frequency (n=141) in the control group; T/C genotype was found in 59% frequency (n=81) in the case group, and 41% frequency (n=56) in the control group; C/C homozygous genotype was found in 72% frequency in the case group (n=18), and in 28% frequency (n=7) in the control group.

Comparative analysis of vitamin D levels in VDR gene genotypes between two groups.

Regarding the level of vitamin D in the control group, the T/T genotype was 43.5±3.6 ng/ml of vitamin D, the normal genotype was 43.5±3.6 ng/ml. genotype

showed a statistically significant difference between the two study groups of adolescent girls (p=0.003).

If the level of vitamin D is 24.2±3.1 ng/ml in the control group of the dangerous allele of T/C heterozygous genotype polymorphism; the level of vitamin D in adolescent girls of the condition group was 23.6±2.7 ng/ml and the relative difference was significant (p=0.01). The C/C homozygous polymorphism in the risk allele case group had a vitamin D level of 12.7±3.7 ng/ml and a control group had a vitamin D level of 13.4±3.2 ng/ml, with no significant difference between the two groups (p= 0.9).

A significant statistical difference was found between the case and control groups on the level of vitamin D in T/T and T/C genotypes: T/T (38.4±6.5/43.5±3.6ng/ml) p=0.003 and T/ C (23.6±2.7/24.2±3.1ng/ml) p=0.01, C/C (12.7±3.7 -13.4±3.2 ng/ml) p =0.9.

GEN-expert, an online calculator used for SNPs, was used to compare the results of the VDR gene study in a case-control design.

A multiplicative model of heritability was used between allele frequencies in the case-control groups while fulfilling Hardy-Weinberg conditions. The T allele occurred at a frequency of 0.716% in the case group and 0.828% in the control group, with an odds ratio (OR) of 0.52 [95% CI: 0.37–0.73]. The C allele was detected at a frequency of 0.284% in the case group and at a frequency of 0.172% in the control group, with an odds ratio (OR) of 1.92 [95% CI; 1.37-2.68], x2=14.71. During the Hardy-Weinberg equilibrium analysis, a statistically significant difference (p= 0.0001) in frequencies between the two groups, that is, in the case group compared to the control group, was revealed.

According to the general heritability model, the frequency of genotypes was used for case-control groups. The T/T genotype occurred at a frequency of 0.519% in the case group and 0.691% in the control group, with an odds ratio (OR) of 0.48 [95% CI: 0.32–0.72]. The frequency of the T/C genotype was 0.393% in the case group and 0.275% in the control group, with an odds ratio (OR) of 1.71 [95% CI; 1.13-2.60]. The C/C genotype occurred with a frequency of 0.087% in the case group and 0.034% in the control group, with an odds ratio (OR) of 2.69 [95% CI; 1.10-6.60] x2=14.05. During the Hardy-Weinberg equilibrium analysis, a statistically significant difference (p= 0.0009) was found between the two groups, i.e. the frequency of genotypes in the case group compared to the control group.

Decision tree prediction model for diagnosing primary dysmenorrhea.

The sensitivity of the resulting model decision tree was 78.2%, specificity - 64.7%. The overall percentage of correctly predicted values of the dependent variable is 71.5%. In general, the maximum number of all levels in this study was 5, the minimum number of observations per parent node was 2, and the minimum number of observations per child node was 1. The resulting number of nodes was 23, the final number of nodes was 13, and the depth was 5.

Of the 13 nodules, 3 significant nodules were associated with primary dysmenorrhea, i.e. 15th, 21st and 17th knots. The result obtained at the 15th node shows that the level of vitamin D deficiency is 100% predisposed to the BD group

due to the frequency of the VDR gene polymorphism of the T/C genotype (p = 0.018).

Estradiol levels ≤169.0 in 21 nodes increased the likelihood of having BD due to vitamin D deficiency by 100% (p=0.012). In the 17th node, the level of vitamin D deficiency increases the likelihood of PD in girls by 88.9% (p = 0.001) depending on the frequency of the T/C and C/C genotypes affected by the VDR gene polymorphism.

Based on the obtained results, the following conclusions were made.

CONCLUSION

1. The 25(OH)D level of teenage girls was significantly lower in the case group with primary dysmenorrhea compared to the control group p= 0.0004. The level of estradiol was significantly higher in the control group than in the case group with primary dysmenorrhea p=0.03.

2. T/T (p=0.003) and T/C (0.01) in the condition group the level of vitamin D in the genotypes was significantly lower compared to the control group, and the difference was not detected in the C/C genotype (p=0.9).

3. A strong negative correlation between 25(OH)D vitamin and VAS was determined r=-0.9 (p<0.0001), that is, the probability of developing primary dysmenorrhea is 10.1 times increased by vitamin D insufficiency [95% CI: 2.869 - 35.665], and the deficiency rate is 11.6 times [95% CI: 3.447; 38.839] significantly increases (Nigelkerka R2 = 0.84). 0.519% in T/T case group, 0.691% in control group, (OR) 0.48 [95% CI: 0.32-0.72]; T/C case group 0.393%, control group 0.275% (OR) 1.71 [95% CI; 1.13-2.60]; C/C case group 0.087%, control group 0.034% (OR) 2.69 [95% CI; 1.10-6.60]. The relationship between the development of primary dysmenorrhea in adolescent girls and VDR gene polymorphism was determined x^2=14.05. (p=0.0009). Vitamin D deficiency increases the likelihood of primary dysmenorrhea in girls by 88.9% (p = 0.001) depending on the frequency of the T/C and C/C genotypes affected by the VDR gene polymorphism.