

DOI: 10.12731/2658-6649-2025-17-6-2-1533

EDN: VGYUTD

UDC 636.08



Original article

EFFECT OF TGF AND VEGF PROTEINS ON PRENATAL DISEASES OF CALVES: CORRELATION ANALYSIS UNDER COMPLICATED PREGNANCY CONDITIONS IN COWS

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Abstract

Background. The study of the role of transforming growth factor (TGF) and vascular endothelial growth factor (VEGF) in the development of prenatal pathologies in calves, especially in the context of complicated pregnancy in cows, is relevant due to the high incidence of morbidity and mortality in newborn young animals, which significantly reduces the economic efficiency of the livestock industry. The study is based on analyzing placental tissue collected immediately after calving to identify relationships between the levels of these proteins and the incidence of pathologies in newborn calves. Changes in TGF and VEGF concentrations can serve as predictors of feto- and uteroplacental insufficiency, which in turn negatively affects the health of calves. Correlation analysis revealed statistically significant relationships between TGF and VEGF levels and clinical disease manifestations, highlighting their role in prenatal disorder pathogenesis. These studies underscore the importance of monitoring TGF and VEGF levels in veterinary practice, potentially enabling early risk identification and the development of effective disease prevention strategies in young animals.

Purpose. The objective of the present study is to develop new methods for predicting prenatal diseases in calves based on the correlation analysis of TGF and VEGF protein levels.

Materials and methods. At the SPC “Plemzavod Vtoraya Pyatiletka”, in the period from 2023 to 2024, a correlation analysis of the proteins TGF and VEGF in the context of complicated pregnancy was carried out.

For the experiment, 200 dry first-calf heifers aged 24 to 48 months were randomly selected. The animals were divided into two groups: Group A included 100

cows with a physiological pregnancy and uncomplicated births, and Group B consisted of 100 animals with complicated pregnancy, accompanied by feto- and uteroplacental insufficiency.

The distribution of livestock into groups was carried out on the basis of anamnesis data and clinical examination results.

Placental tissue collected immediately after calving in compliance with the temperature (cold) regime was used as research material.

The study analyzed the protein level of transforming growth factor (TGF), which regulates various cellular functions such as growth, development, immune responses, and tissue remodeling. The protein level of vascular endothelial growth factor (VEGF), which is involved in angiogenesis, was also assessed using enzyme immunoassay kits from the biotechnological company Cutimmune systems, located in the United States.

The licensed program “Primer of Biostatistics 4.03. For Windows” was used to process the data in the study.

The homogeneity of variances was assessed using the Fisher criterion, which allows determining the uniformity of variances in a group.

To test the statistical significance of differences between the compared groups, two criteria were used: the Student criterion and the nonparametric Mann-Whitney criterion. The Student criterion is a parametric method and is designed to compare the average values of two groups. The nonparametric Mann-Whitney criterion, in turn, is designed to compare two independent and unrelated small samples by a quantitative feature of two groups. Differences were considered reliable at $p < 0.05$.

To conduct a correlation analysis, the Spearman method was used, which allows determining the strength and direction of the relationship between two features.

Results. During the study, it was found that prenatal disorders in calves born from cows with feto- and uteroplacental insufficiency have a direct correlation with the level of protein transforming growth factor (TGF) and protein vascular endothelial growth factor (VEGF).

Conclusion. The results of the studies revealed significant differences in the levels of VEGF and TGF proteins in cows with complicated pregnancy. Data analysis showed that the level of VEGF in animals from group B was 24% higher compared to the indicators of group A ($p < 0.05$). At the same time, a significant decrease in the concentration of TGF in the placentas of cows with complicated pregnancy was observed - 2.4 times lower than in animals with a physiological course of pregnancy ($p < 0.01$). These changes may indicate a compensatory mechanism in which an increase in VEGF production is aimed at leveling out the consequences of a sharp decrease in the level of TGF.

Also, special attention was paid to the ratio of VEGF and TGF in the placenta. In group A, this coefficient was 1.1%, which indicates a balance of the processes regulated by these growth factors. At the same time, a significant increase in the VEGF/TFR ratio was observed in Group B – approximately 3.2 times compared to Group A. This indicates a pronounced imbalance in the regulation of angiogenesis and metabolic processes in cows with complicated pregnancy.

The obtained VEGF/TFR ratio can become a valuable tool for predicting physiological disorders in calves during postembryonic ontogenesis. Its use will optimize veterinary strategies and, possibly, prevent the development of diseases in the long term. This is important not only for ensuring animal welfare, but also for increasing the economic efficiency of animal husbandry by reducing losses associated with morbidity and mortality of young animals.

Keywords: transforming growth factor; vascular endothelial growth factor; prenatal diseases; pregnancy; cows

For citation. Samoylenko, V. S., Lapina, A. A., Zhivoderova, A. I., Pushkin, S. V., & Svetlakova, E. V. (2025). Effect of TGF and VEGF proteins on prenatal diseases of calves: Correlation analysis under complicated pregnancy conditions in cows. *Siberian Journal of Life Sciences and Agriculture*, 17(6-2), 81-94. <https://doi.org/10.12731/2658-6649-2025-17-6-2-1533>

Научная статья

ВЛИЯНИЕ БЕЛКОВ TGF И VEGF НА ПРЕНАТАЛЬНЫЕ ЗАБОЛЕВАНИЯ ТЕЛЯТ: КОРРЕЛЯЦИОННЫЙ АНАЛИЗ В УСЛОВИЯХ ОСЛОЖНЕННОЙ БЕРЕМЕННОСТИ У КОРОВ

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Аннотация

Обоснование. Изучение роли трансформирующего фактора роста (TGF) и фактора роста эндотелия сосудов (VEGF) в развитии пренатальных патологий у телят, особенно в условиях осложненной беременности у коров, актуально в связи с высокой заболеваемостью и смертностью новорожденного молодняка, что значительно снижает экономическую эффективность животноводческой отрасли. Исследование основано на анализе плацентарной ткани, взятой сра-

зу после отела, с целью выявления взаимосвязи между уровнем этих белков и частотой патологий у новорожденных телят. Изменения концентрации TGF и VEGF могут служить предикторами фето- и маточно-плацентарной недостаточности, что, в свою очередь, негативно сказывается на здоровье телят. Корреляционный анализ выявил статистически значимые связи между уровнями TGF и VEGF и клиническими проявлениями заболеваний, что подчеркивает их роль в патогенезе пренатальных нарушений. Эти исследования подчеркивают важность мониторинга уровней TGF и VEGF в ветеринарной практике, что может способствовать раннему выявлению рисков и разработке эффективных стратегий профилактики заболеваний у молодых животных.

Цель. Целью исследования является разработка новых методов прогнозирования пренатальных заболеваний у телят на основе корреляционного анализа уровней белков TGF и VEGF.

Материалы и методы. В НПП «Племзавод “Вторая Пятилетка”» в период с 2023 по 2024 год был проведен корреляционный анализ белков TGF и VEGF в условиях осложненной беременности.

Для эксперимента были случайным образом отобраны 200 сухостойных первотелок в возрасте от 24 до 48 месяцев. Животные были разделены на две группы: В группу А вошли 100 коров с физиологической беременностью и неосложненными родами, а в группу Б – 100 животных с осложненной беременностью, сопровождающейся фето- и маточно-плацентарной недостаточностью.

Распределение поголовья по группам проводилось на основании данных анамнеза и результатов клинического обследования.

В качестве материала исследования использовали плацентарную ткань, собранную сразу после отела с соблюдением температурного (холодового) режима.

В ходе исследования был проанализирован уровень белка трансформирующего фактора роста (TGF), который регулирует различные клеточные функции, такие как рост, развитие, иммунные реакции и ремоделирование тканей. Уровень белка фактора роста эндотелия сосудов (VEGF), участвующего в ангиогенезе, также оценивали с помощью наборов для иммуноферментного анализа биотехнологической компании Cutimmune systems, расположенной в США.

Для обработки данных в исследовании использовалась лицензионная программа «Primer of Biostatistics 4.03. For Windows» использовалась для обработки данных в исследовании.

Однородность вариаций оценивали с помощью критерия Фишера, который позволяет определить однородность вариаций в группе.

Для проверки статистической значимости различий между сравниваемыми группами использовали два критерия: критерий Стьюдента и непараметриче-

ский критерий Манна-Уитни. Критерий Стьюдента является параметрическим методом и предназначен для сравнения средних значений двух групп. Непараметрический критерий Манна-Уитни, в свою очередь, предназначен для сравнения двух независимых и несвязанных малых выборок по количественному признаку двух групп. Различия считались достоверными при $p < 0,05$.

Для проведения корреляционного анализа использовали метод Спирмена, который позволяет определить силу и направление связи между двумя признаками.

Результаты. В ходе исследования было установлено, что пренатальные нарушения у телят, рожденных от коров с фето- и маточно-плацентарной недостаточностью, имеют прямую корреляцию с уровнем белка трансформирующего фактора роста (TGF) и белка сосудисто-эндотелиального фактора роста (VEGF).

Заключение. Результаты исследований выявили значительные различия в уровнях белков VEGF и TGF у коров с осложненной беременностью. Анализ данных показал, что уровень VEGF у животных из группы В был на 24 % выше по сравнению с показателями группы А ($p < 0,05$). В то же время наблюдалось значительное снижение концентрации TGF в плацентах коров с осложненной беременностью - в 2,4 раза ниже, чем у животных с физиологическим течением беременности ($p < 0,01$). Данные изменения могут свидетельствовать о компенсаторном механизме, при котором увеличение продукции VEGF направлено на нивелирование последствий резкого снижения уровня TGF.

Также особое внимание было уделено соотношению VEGF и TGF в плаценте. В группе А этот коэффициент составил 1,1%, что свидетельствует о сбалансированности процессов, регулируемых этими факторами роста. В то же время в группе В наблюдалось значительное увеличение соотношения VEGF/TFR - примерно в 3,2 раза по сравнению с группой А. Это свидетельствует о выраженном дисбалансе в регуляции ангиогенеза и метаболических процессов у коров с осложненной беременностью.

Полученное соотношение VEGF/TFR может стать ценным инструментом для прогнозирования физиологических нарушений у телят в постэмбриональном онтогенезе. Его использование позволит оптимизировать ветеринарные стратегии и, возможно, предотвратить развитие заболеваний в долгосрочной перспективе. Это важно не только для обеспечения благополучия животных, но и для повышения экономической эффективности животноводства за счет снижения потерь, связанных с заболеваемостью и смертностью молодняка.

Ключевые слова: трансформирующий фактор роста; фактор роста эндотелия сосудов; пренатальные заболевания; беременность; коровы

Для цитирования. Самойленко, В. С., Лапина, А. А., Живодерова, А. И., Пушкин, С. В., & Светлакова, Е. В. (2025). Влияние белков TGF и VEGF на пренатальные заболевания телят: Корреляционный анализ в условиях осложненной беременности у коров. *Siberian Journal of Life Sciences and Agriculture*, 17(6-2), 81-94. <https://doi.org/10.12731/2658-6649-2025-17-6-2-1533>

Introduction

The birth of calves with intrauterine pathologies is a major issue in veterinary medicine and animal husbandry, leading to significant economic losses and health deterioration in young animals [1; 2]. Prenatal disorders that occur in the context of complicated pregnancy require immediate intervention and correction to minimize their negative consequences.

One of the key elements that determine the health of the fetus is uteroplacental insufficiency, which is often accompanied by hemodynamic disorders and oxygen starvation of the fetus. Such conditions can cause severe damage to the tissues and organs of newborns, including disorders in the functioning of the nervous system [3; 5]. The main clinical manifestations of uteroplacental insufficiency are hypoxia and intrauterine growth retardation, which is confirmed by numerous studies [1; 2].

An important aspect in the development of these pathologies is the disruption of angiogenesis processes in the early stages of embryo formation. These processes directly depend on the production of growth factors – regulatory polypeptides that affect the development of the placenta, its growth and the formation of the vascular network. Among the many known growth factors, special attention is paid to transforming growth factor (TGF) and specific extrafollicular growth factor (VEGF). These proteins, synthesized by the placenta, play a key role in the regulation of metabolic processes and angiogenesis, which is critical for the normal course of pregnancy [8; 9].

An imbalance of these endogenous regulators can cause various pregnancy complications, including uteroplacental insufficiency. Normal fetal development and the course of gestation directly depend on the state of angiogenesis and the production of angiogenic factors that control the formation and regression of blood vessels.

Thus, there is a need to develop new methods for predicting prenatal diseases in calves based on a correlation analysis of TGF and VEGF protein levels. This will allow timely implementation of adequate pathogenetic therapy, which will help reduce the frequency and severity of manifestations of prenatal disorders, which will ultimately improve the health of newborn young animals and increase the efficiency of animal husbandry.

Purpose. The objective of the present study is to develop new methods for predicting prenatal diseases in calves based on the correlation analysis of TGF and VEGF protein levels.

Materials and methods

At the SPC “Plemzavod Vtoraya Pyatiletka”, in the period from 2023 to 2024, a correlation analysis of the proteins TGF and VEGF in the context of complicated pregnancy was carried out.

For the experiment, 200 dry first-calf heifers aged 24 to 48 months were randomly selected. The animals were divided into two groups: Group A included 100 cows with a physiological pregnancy and uncomplicated births, and Group B consisted of 100 animals with complicated pregnancy, accompanied by fetoplacental insufficiency.

The distribution of livestock into groups was carried out on the basis of anamnesis data and clinical examination results.

Placental tissue collected immediately after calving in compliance with the temperature (cold) regime was used as research material.

The study analyzed the protein level of transforming growth factor (TGF), which regulates various cellular functions such as growth, development, immune responses, and tissue remodeling.

The protein level of vascular endothelial growth factor (VEGF), which is involved in angiogenesis, was also assessed using enzyme immunoassay kits from the biotechnological company Cutimmune systems, located in the United States.

The licensed program “Primer of Biostatistics 4.03. For Windows” was used to process the data in the study.

The homogeneity of variances was assessed using the Fisher criterion, which allows determining the uniformity of variances in a group.

To test the statistical significance of differences between the compared groups, two criteria were used: the Student criterion and the nonparametric Mann-Whitney criterion. The Student criterion is a parametric method and is designed to compare the average values of two groups. The nonparametric Mann-Whitney criterion, in turn, is designed to compare two independent and unrelated small samples by a quantitative feature of two groups. Differences were considered reliable at $p < 0.05$.

To conduct a correlation analysis, the Spearman method was used, which allows determining the strength and direction of the relationship between two features.

Results of the research

The level of protein TGF and growth factor VEGF in complicated pregnancy.

During the study, it was found that prenatal disorders in calves born from cows with feto- and uteroplacental insufficiency have a direct correlation with the level of protein transforming growth factor (TGF) and protein vascular endothelial growth factor (VEGF) (Table 1).

Table 1.

Level of growth factors in the placenta of cows from groups A and B (M±m)

Study groups	Contents of the SEFR	Contents of TFR
A group – pregnancy without complications	0.34+0.1* (ng/mg tissue)	0.31+0.95 (ng/mg tissue)
Group B – complicated (pathological) pregnancy	0.41+0.2* (ng/mg tissue)	0.13+0.4* (ng/mg tissue)

* – deviations from the norm in pathological pregnancy are statistically confirmed.

The obtained data presented in Table 1 and their analysis clearly demonstrate that the level of vascular endothelial growth factor protein in cows from group B significantly exceeded the indicators of cows from group A by 24% ($p < 0.05$). At the same time, the content of TGF significantly decreased in the placentas of the studied cows with complicated pregnancy from group B compared to group A by 2.4 times ($p < 0.01$). This may indicate that an increase in the production of VEGF compensates for a sharp drop in the TGF level. The identified changes in the production of growth factors during complicated pregnancy in the studied cows are associated with the suppression of key functions of the placenta, which in turn leads to the progression of irreversible processes in this organ, thereby worsening the physiological development of the fetus.

The established disorganization in the production of growth factors such as VEGF and TGF can create favorable conditions for the development of feto- and uteroplacental insufficiency, and fetal growth retardation. It is important to note that the imbalance in the level of growth factors, accompanied by an increase in the production of VEGF and TGF, can be a consequence of hypoxic conditions inherent in complicated pregnancy. These changes can affect the regulation of blood flow, metabolism and immune responses in the body of the cow and the fetus, leading to intrauterine growth retardation.

Taking into account the important complex effect of growth factors on the processes they control and the relationship between their effects, it is proposed to calculate the ratio of the concentration of vascular endothelial growth factor

(VEGF) to transforming growth factor (TGF) and study changes in this coefficient at different stages of calf ontogenesis. This analysis involves monitoring calves for 60 days to identify the dynamics of changes.

Changes in the VEGF/TGF ratio may reflect the interaction between two key growth factors and their role in regulating physiological adaptation processes in calves during early postnatal ontogenesis. A high or low level of this ratio may indicate disturbances in the balance between the two factors, which may affect animal productivity.

Studying the dynamics of this ratio during the 60-day period after birth will help identify possible patterns between its change and physiological processes occurring during postembryonic development of calves. This approach helps identify factors influencing the productivity of young animals, as well as in developing strategies for maintaining an optimal balance of growth factors in animals during early postnatal ontogenesis.

The period of early neonatal ontogenesis in young cattle of all groups was recorded without complications. Despite the absence of physiological disorders in newborn calves of all groups, special attention was paid to the ratio of certain growth factors (VEGF/TGF) in the placenta. In group A, this coefficient was 1.1%, indicating a certain balance between vascular endothelial and transforming growth factor. However, in group B, a significant increase in this coefficient was observed, approximately 3.2 times that of group A (Table 2).

Table 2.

Growth factor coefficient of cows from groups A and B

Study groups	TFR/SEFR ratio
A group – pregnancy without complications	1.1
Group B – complicated (pathological) pregnancy	3.54

One of the characteristic features of physiological adaptation disorders in calves in postembryonic ontogenesis is often the absence of noticeable clinical signs during the initial examination, which appear only a few hours after birth. Clinical manifestations of these disorders in newborn calves differ from those in older calves. In this regard, newborn calves from Group B were under the supervision of a veterinarian during the first two months of life.

Upon reaching the age of one and a half months, clinical signs of cerebral disorders (n=32) were detected in calves of the studied Group B, namely: an uncertain standing posture, decreased appetite and changes in eating behavior, increased excitability or, on the contrary, passivity, deviations in physical development. Thus, monitoring the coefficient of growth factors and regulating

their production can play a key role in improving pregnancy outcomes in cows, health and productivity of newborn offspring.

Discussion and conclusion

The results of the studies revealed significant differences in the levels of VEGF and TGF proteins in cows with complicated pregnancy. Data analysis showed that the level of VEGF in animals from group B was 24% higher compared to the indicators of group A ($p < 0.05$). At the same time, a significant decrease in the concentration of TGF in the placentas of cows with complicated pregnancy was observed - 2.4 times lower than in animals with a physiological course of pregnancy ($p < 0.01$). These changes may indicate a compensatory mechanism in which an increase in VEGF production is aimed at leveling out the consequences of a sharp decrease in the level of TGF.

Also, special attention was paid to the ratio of VEGF and TGF in the placenta. In group A, this coefficient was 1.1%, which indicates a balance of the processes regulated by these growth factors. At the same time, a significant increase in the VEGF/TFR ratio was observed in Group B — approximately 3.2 times compared to Group A. This indicates a pronounced imbalance in the regulation of angiogenesis and metabolic processes in cows with complicated pregnancy.

The obtained VEGF/TFR ratio can become a valuable tool for predicting physiological disorders in calves during postembryonic ontogenesis. Its use will optimize veterinary strategies and, possibly, prevent the development of diseases in the long term. This is important not only for ensuring animal welfare, but also for increasing the economic efficiency of animal husbandry by reducing losses associated with morbidity and mortality of young animals.

Conflict of interest information. The authors declare that they have no conflict of interest.

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AUTHOR CONTRIBUTIONS

The authors contributed equally to this article.

ВКЛАД АВТОРОВ

Все авторы сделали эквивалентный вклад в подготовку статьи для публикации.

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Поступила 07.07.2025

После рецензирования 26.08.2025

Принята 01.09.2025

Received 07.07.2025

Revised 26.08.2025

Accepted 01.09.2025