

## Original Research Article

### CLINICAL SIGNS OF SEASONAL DISEASES DYNAMICS IN CALVES CAUSED BY ROTAVIRUS AND CORONAVIRUS INFECTION

#### Abstract

The study of animal diseases increases the relevance of the livestock industry, which is the main leading component of the strategy for the development of agriculture in our country. An important role in the implementation of measures to combat diseases of infant calves caused by rotavirus and coronavirus infection is played by their study. The main goal that we set ourselves in carrying out this study was to study the seasonal dynamics of accompanying diarrhea in calves caused by rotavirus and coronavirus infection. The research was carried out in farms located in the northwestern region of the republic.

The study revealed that the clinical signs of diarrhea in calves caused by rotavirus and coronavirus infection in farms located in the northwestern region of the country are relatively similar. However, there was a relatively higher incidence of rotavirus infection, and mortality relative to coronavirus was at a lower level. The occurrence of secondary pneumonia in patients with coronavirus was noted as one of the main clinical signs. The trend of infection and mortality from both diseases varies throughout the year from season to season and, as a result of this, becoming massive, causes serious damage to farms in spring, autumn and winter.

*Keywords: coronavirus, rotavirus, indigestion, seasonal, diarrhea, serotype.*

#### INTRODUCTION

Seasonal changes are one of the most important and predictable systems influencing humans and ecosystems. The spread of many pathogenic infections is seasonal, for example, respiratory diseases in humans and rotaviruses in children, which are more widespread during the winter months [4,31]. Seasonal changes repeated every year can cause changes in the biology of the susceptible organism and infections and cause epidemics [21]. However, other leading factors that can also lead to epidemics should be taken into account [5, 7]. The coronavirus pandemic, which is a disease of viral origin, which the planet is facing in modern times, has shown that in order to be prepared to prevent such diseases and avoid similar risks in the future, it is extremely important to comprehensively investigate and study diseases of viral origin from the point of view of reduce biological threats [19, 26]. Rotavirus is a viral disease of calves characterized by dyspepsia, diarrhea and dehydration [17, 22]. At least different 27 G and 37 P rotavirus genotypes have been identified that can lead to rotavirus infections [12]. Rotaviruses of the Reoviridae family are spherical viruses with a double-stranded and 11-segment RNA genome [27]. Genomic segments encode 12 proteins, six of which are structural proteins (VP1, VP2, VP3, VP4, VP6, and VP7) and six are nonstructural proteins (NSP1–NSP6) [2, 14,

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18]. The disease is widespread in countries developed in terms of industrially oriented animal husbandry. Usually, 2–3-day old calves are more susceptible to the disease.

When calves are infected with contagious secondary infections of the gastrointestinal tract (*E. coli*), mortality among sick animals reaches 100% [11,19].

Coronaviruses are divided into 4 genotypic groups according to their genetic proximity: Alphacoronaviruses, Betacoronaviruses, Gamacoronaviruses and Deltacoronaviruses. Coronaviruses in cows are membrane-enveloped RNA viruses with positive polarity belonging to the Coronaviridae family [21, 28]. Betacoronaviruses are coronaviruses that cause SARS-CoV-2 in humans [24,26]. The virus has a common antigen with strains of human coronavirus, hepatitis in mice and rats, and porcine encephalomyelitis. Transmission of the virus is widespread among animals (25). Calves become ill from day 10 to 8 weeks of age if they do not have inherited maternal antibodies (20, 33). Pathogenic viruses that have spread in the environment in different serotypes, mutating, change their serotypes and cause epizootics, epidemics, and even pandemics. Along with humans, coronaviruses are also accompanied by diarrhea in animals, including calves. In the United States, BCoV-WD is more prevalent in the northern states [16].

Rotavirus causes death of nursing calves and leads to serious economic losses for farmers around the world [33].

Rotavirus is one of the most widespread infectious agents worldwide, causing diseases, and it has been extensively studied in depth [34]. The prevalence of rotavirus among calves with diarrhea in India ranges from 11.8% to 26.8% [6]. Rotavirus infection has also been widely studied in European countries. Between 1993 and 2006, estimated prevalence was 24–47% in Sweden, 42% in outbreaks of diarrhea in the UK, and 37–47.4% in France [3, 8, 23].

According to available statistics in Azerbaijan, 75-95% of infectious diarrhea in calves is caused by *E. coli*, *Rotavirus*, *Coronavirus* and *Cryptosporidiosis*, with rotavirus accounting for 27-36% and coronavirus for 20-26%.

The study of both viruses can play an important role in reducing risks on farms. The purpose of this study was to identify seasonal dynamics and clinical signs of rotavirus and coronavirus diseases accompanied by diarrhea in calves in the northwestern region of the republic.

## **MATERIALS AND METHODS**

### **Study area and climatic characteristics**

The study used a statistical database for 2017-2020 on diarrhea that spread among calves from the "Holstein-Friesian" breed of cattle imported from abroad to "Gilan Dairy Farms" LLC in the Gabala region, which belongs to the northwestern region republics. The farm has 2,000 head of cattle, and the number of dairy cows ranges from 900. The complex has 7 stalls, a feed production workshop, feed dryers, ancillary buildings and 120 calf pens. The complex is located in the village of Boyuk-Emily, Gabala region, the height of the village is 389 meters above sea level, the average temperature in winter is 3.9-9.4 °C, and in summer it is in the range of 15.4-29.5 °C. According to academician B.A. Budagov, the hottest place in the region is marked in the direction of the villages of Kushlar-Kurd-Bayramkokhaly. And the minimum temperature reaching up to -50°C in the region is observed at the top of Bazardyuzu (4466 m). Snow cover in the area lasts 45 days. The first autumn frosts are

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observed in the 2nd decade of December and in January, and the last spring frosts, as a rule, usually occur on April 8th. The average annual number of frost-free days is 241 days, the number of days with temperatures above 5 degrees during the flowering period is 265 days, while the number of days with temperatures above 10 degrees reaches 198 days. Southern slope of the Greater Caucasus, which includes the Gabala region, was divided into six agro-climatic regions. Each of these areas occupies its own place and is important for the development of agricultural territories.

### Collected samples

During this process, fecal samples taken from calves at different times of the year were checked by rapid testing, their registration was carried out, and the seasonality of infection was determined.

Fecal samples were collected only within the first 24 hours of the first clinical signs in calves aged 4 to 30 days by digital fecal release after the formation of a rectal reflex.

In 2020, 328 samples were taken, in 2019 - 342 samples, in 2018 - 358 samples, in 2017 - 357 samples.

### Test procedure

In order to clarify the results of the studies, clinical and pathoanatomical methods of research were used, pathoanatomical autopsies of dead animals were performed. During the autopsy, scissors, a surgical knife, a mask, gloves, and sample containers were used.

For testing according to the instructions, kits for express testing were used. Thus, when diagnosing, the BoviD-4/5 rapid test sampling stick, D4 Diarrhea Ag, test strips, sterile gauze, test flask, ready-made solvent inside the flask, test pipette and sterile gloves were used in the following sequence:

1. The selected samples are dissolved in the solvent and a pause is made for 30 seconds.
2. Dissolved samples are taken from the solution with a pipette.
3. 4 drops of solution are added to each cell.
4. Results are read in 5-10 minutes
5. Strips showing control and test lines are considered positive for rotavirus infection.
6. In clinical trials, other research methods were used using an electronic thermometer and a phonendoscope.

### RESULTS

Gabala region is distinguished by a large number of cold days a year. As can be seen from Table 1, the highest temperature in the winter months ranges from 5-7 °C, and in the summer months it reaches a maximum of 29 °C. Such climatic conditions were regarded as more favorable for enteroviruses.

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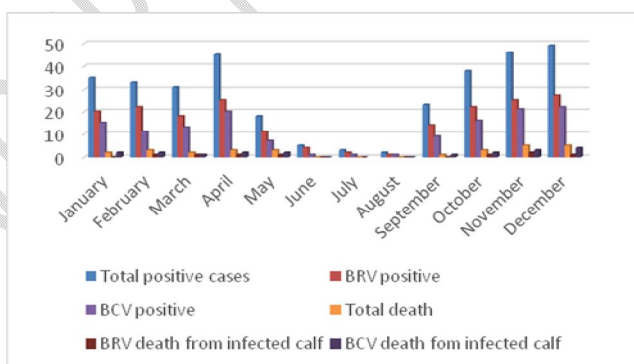
Climate of Gabala region												
Months	January	February	March	April	May	June	July	August	September	October	November	December
Maximum temperature °C (°F)	4.4 (39.9)	5.0 (41.0)	9.4 (48.9)	17.3 (63.1)	21.1 (69.9)	26.0 (78.8)	29.5 (85.1)	28.6 (83.3)	24.8 (76.6)	17.4 (63.3)	11.4 (52.5)	7.0 (44.6)
Minimum temperature °C (°F)	-3.9 (24.98)	-2.8 (41.0)	1.0 (48.92)	6.7 (50.4)	11.2 (59.9)	15.4 (70.3)	18.2 (76.3)	17.4 (76.8)	13.9 (73.0)	8.5 (65.1)	3.0 (56.8)	-1.4 (50.9)
Number of sunny days	10.0	11.0	12.0	13.0	14.0	15.0	15.0	14.0	12.0	11.0	10.0	9.0

**Table 1. Temperature indicators in the Gabala region throughout the year.**

Tables 2 and 3, Diagram 1 shows the indicators of calves that tested positive for rotavirus and coronavirus, as well as cases of their death due to the disease in 2020 by months and seasons of the year, as well as a comparative analysis of cattle loss cases from rotavirus and coronavirus.

Results of 2020	January	February	March	April	May	June	July	August	September	October	November	December
Total positive cases	35	33	31	45	18	5	3	2	23	38	46	49
BRV positive	20 (57.1%)	22 (66.7%)	18 (58.1%)	25 (55.6%)	11 (61.1%)	4 (80.0%)	2 (66.7%)	1 (50.0%)	14 (60.9%)	22 (57.9%)	25 (54.3%)	27 (55.1%)
BCV positive	15 (42.86%)	11 (33.33%)	13 (41.94%)	20 (44.44%)	7 (38.89%)	1 (20.00%)	1 (33.33%)	1 (50.00%)	9 (39.13%)	16 (42.11%)	21 (45.65%)	22 (44.90%)
Total death	2 (5.7%)	3 (9.1%)	2 (6.5%)	3 (6.7%)	3 (16.7%)	0	0	0	1 (4.3%)	3 (7.9%)	5 (%)	5 (10.2%)
BRV death from infected calf	0	1 (4.5%)	1 (5.6%)	1 (4.0%)	1 (9.1%)	0	0	0	0	1 (4.5%)	2 (8.0%)	1 (3.7%)
BCV death from infected calf	2 (13%)	2 (18%)	1 (8%)	2 (10%)	2 (29%)	0	0	0	1 (11%)	2 (13%)	3 (14%)	4 (18%)

**Table 2. Dynamics of the spread of rotavirus and coronavirus diseases in "Gilan Dairy Farms" LLC by months in 2020**



**Fig.1. Correlation of positive results in 2020 by months**

As can be seen from Table 2, in the winter period of 2020, 99 heads of cattle were infected with coronavirus and rotavirus infection. Of these, 39 heads of cattle were infected with coronavirus and 60 heads of cattle with rotavirus. The death of 5 heads of cattle (percentage mortality 13%) from

coronavirus infection and 2 heads of cattle (percentage mortality 3.3%) from rotavirus infection was recorded.

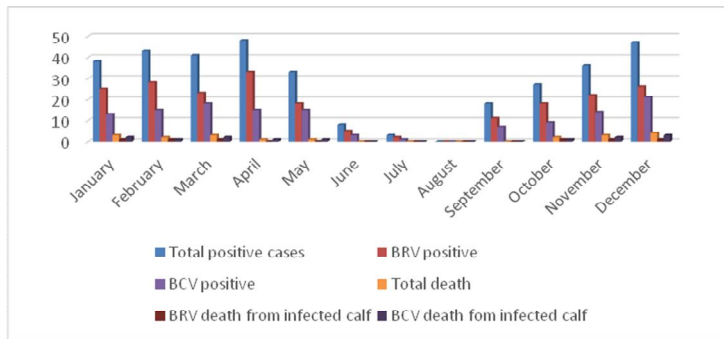
In the spring season of 2020, 68 heads of cattle were infected with coronavirus and rotavirus infection, including 28 heads of cattle were infected with coronavirus and 40 heads of cattle with rotavirus infection. The cattle loss from coronavirus infection was 4 heads of cattle (13%) and 2 heads of cattle from rotavirus infection (5%).

During the summer season of 2020, the death of 1 head of cattle from infection with coronavirus and rotavirus infection was registered (percentage mortality 4%) and not a single case of cattle loss from rotavirus infection. During the autumn season, cases of infection with coronavirus and rotavirus infection affected 59 heads of cattle (percentage mortality 15% ) for coronavirus infection and 74 heads of cattle (percentage mortality 5.4% ) for rotavirus infection.

Results of 2019	January	February	March	April	May	June	July	August	September	October	November	December
Total positive cases	65	70	48	36	33	8	3	2	32	80	88	82
Rotavirus positive	35 (54%)	40 (57%)	28 (58%)	20 (56%)	18 (55%)	5 (63%)	2 (67%)	1 (50%)	18 (56%)	43 (54%)	46 (52%)	42 (51%)
Coronavirus positive	30 (46%)	30 (43%)	20 (42%)	16 (44%)	15 (45%)	3 (38%)	1 (33%)	1 (50%)	14 (44%)	37 (46%)	42 (48%)	40 (49%)
Rotavirus death	2 (6%)	2 (5%)	1 (4%)	0	0	0	0	0	2 (11%)	3 (7%)	5 (11%)	3 (7%)
Coronavirus death	3 (10%)	3 (10%)	2 (10%)	1 (6%)	1 (7%)	0	0	0	4 (29%)	5 (14%)	6 (14%)	6 (15%)
Total death	5 (8%)	5 (7%)	3 (6%)	1 (3%)	1 (3%)	0	0	0	6 (19%)	8 (10%)	11 (13%)	9 (11%)

**Table 3. Dynamics of the spread of rotavirus and coronavirus diseases in "Gilan Dairy Farms" LLC by months in 2019**

Table 3 shows the indicators of calves that tested positive for rotavirus and coronavirus by months and seasons in 2019, as well as the mortality rates from diseases by months, including from rotavirus and coronavirus.

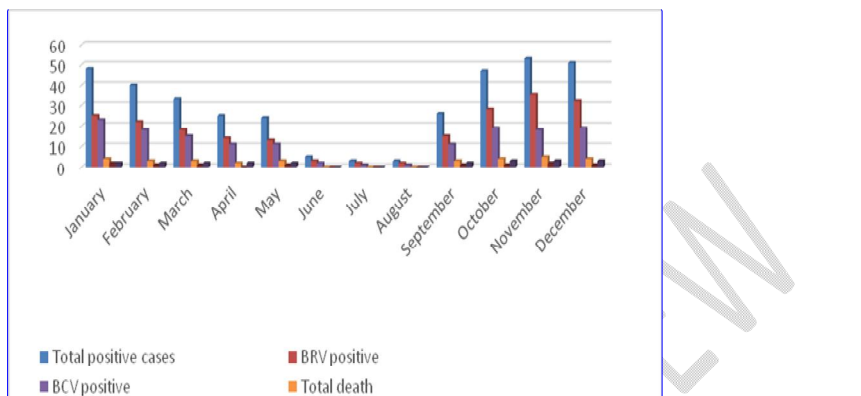


**Fig.2. Correlation of positive results in 2019 by months**

Table 4 shows that during the winter season of 2019, 122 heads of cattle were infected with coronavirus and rotavirus infection, including 46 heads of cattle infected with coronavirus and 76 heads of cattle with rotavirus infection. There were a cattle loss cases of 5 heads of cattle (10%) from infection with coronavirus infection and 3 heads of cattle from infection with rotavirus infection 3 (3.9%). During the summer season, 89 heads of cattle were infected with coronavirus and rotavirus infection, of which 33 heads of cattle were infected with coronavirus and 56 heads of cattle with rotavirus. There was cattle loss case of 2 heads of cattle from infection with coronavirus infection (percentage mortality 6%) and 0 cattle loss cases of infection with rotavirus infection (0%). During the summer season, 8 heads of cattle were infected with the coronavirus (percentage mortality 0%), 13 heads of cattle were infected with rotavirus (percentage mortality 0%). In the autumn season of 2019, 44 heads of cattle were infected with the coronavirus (percentage mortality 13.6%), 66 heads of cattle were infected with rotavirus (percentage mortality 4.5%).

Results of 2018	January	February	March	April	May	June	July	August	September	October	November	December
Total positive cases	48	40	33	25	24	5	3	3	26	47	53	51
BRV positive	25 (52%)	22 (55%)	18 (55%)	14 (56%)	13 (54%)	3 (60%)	2 (67%)	2 (67%)	15 (58%)	28 (54%)	35 (58%)	32 (55%)
BCV positive	23 (48%)	18 (45%)	15 (45%)	11 (44%)	11 (46%)	2 (40%)	1 (33%)	1 (33%)	11 (42%)	19 (46%)	18 (42%)	19 (45%)
BRV death from infected calf	2 (8%)	1 (8%)	1 (6%)	0 (0)	1 (8%)	0 (0%)	0 (0%)	0 (0%)	1 (7%)	1 (9%)	2 (6%)	1 (10%)
BCV death from infected calf	2 (9%)	2 (11%)	2 (13%)	2 (18%)	2 (18%)	0 (0%)	0 (0%)	0 (0%)	2 (18%)	3 (13%)	3 (16%)	3 (16%)
Total death	4 (8%)	3 (8%)	3 (9%)	2 (8%)	3 (13%)	0 (0%)	0 (0%)	0 (0%)	3 (12%)	4 (11%)	5 (10%)	4 (13%)

**Table 4. Dynamics of the spread of rotavirus and coronavirus diseases in "Gilan Dairy Farms" LLC by months in 2018.**



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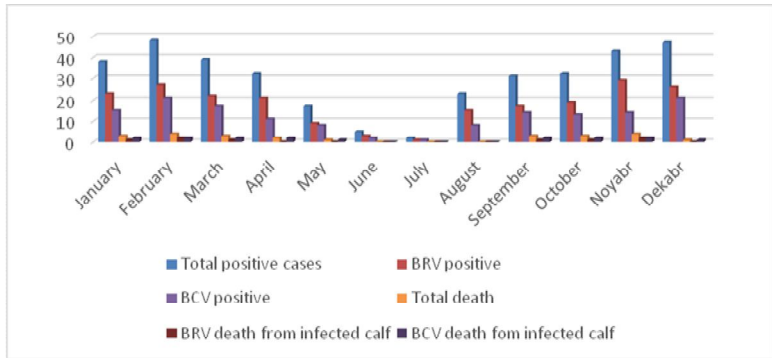
**Fig.3. Correlation of positive results in 2018 by months**

Table 4 shows that in the winter season of 2018, 56 heads of cattle were infected with coronavirus (percentage mortality 10.7%), 65 heads of cattle were infected with rotavirus (percentage mortality 6.1%). During the spring season of 2018, 24 heads of cattle (percentage mortality 16.6%) were infected with coronavirus and 30 heads of cattle (percentage mortality 3.3%) were infected with rotavirus. In the summer season, 13 heads of cattle were infected with coronavirus (percentage mortality 15.3%), 19 heads of cattle (percentage mortality 5.2%) - with rotavirus. In the autumn season, 56 heads of cattle were infected with coronavirus (percentage mortality 16%), 695 heads of cattle were infected with rotavirus (percentage mortality 4.2%).

Results of 2017	January	February	March	April	May	June	July	August	September	October	November	December
Total positive cases	38	48	39	32	17	5	2	23	31	32	43	47
BRV positive	23 (52%)	27 (55%)	22 (55%)	21 (56%)	9 (54%)	3 (60%)	1 (67%)	15 (67%)	17 (58%)	19 (54%)	29 (58%)	26 (55%)
BCV positive	15 (52%)	21 (55%)	17 (55%)	11 (56%)	8 (54%)	2 (60%)	1 (67%)	8 (67%)	14 (58%)	13 (54%)	14 (58%)	21 (55%)
BRV death from infected calf	1 (8%)	2 (5%)	1 (6%)	0 (0%)	0 (8%)	0 (0%)	0 (0%)	0 (0%)	1 (7%)	1 (9%)	2 (6%)	0 (10%)
BCV death from infected calf	2 (9%)	2 (11%)	2 (13%)	2 (18%)	1 (18%)	0 (0%)	0 (0%)	0 (0%)	2 (18%)	2 (13%)	2 (16%)	1 (16%)

Total death	3 (8%)	4 (8%)	3 (9%)	2 (8%)	1 (13%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	3 (12%)	3 (11%)	4 (10%)	1 (13%)
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**Table 5. Dynamics of the spread of rotavirus and coronavirus diseases in "Gilan Dairy Farms" LLC by months in 2017.**



**Fig.4. Correlation of positive results in 2017 by months**

## DISCUSSION

According to Qinghe Zhu et al., diarrhea in calves is a multifactorial disease caused by a combination of infectious and non-infectious complex risk factors, the manifestation of which is influenced by enteropathogenic bacteria, viruses, factors that cause peripheral conditions, the immune status of the animal, genetic factors, nutrition, course childbirth, the structure of the alveoli of the calf, vaccination of the mother, the state of health of the calf. Our field studies have shown that diarrhea in calves is the most common type of diarrhea caused by rotavirus and coronavirus, and the frequency of cases varied depending on the season and year. According to our observations, diarrhea caused by rotaviruses was recorded most often at the age of 5-15 days. According to the literature, watery yellow diarrhea in calves is a clinical sign of rotaviruses [13]. Depression, shock, and dehydration are more common in calves under 5 days of age [1]. The main clinical signs in the affected calf were depression, decreased sucking reflex, diarrhea, and dehydration. Diarrhea in calves caused by coronaviruses has been reported in calves aged 2-21 days. The epithelial cells of the small and large intestine are sensitive to the virus [14]. Usually, a coronavirus infection causes watery diarrhea, and blood clots can be seen in the feces. In our studies, the most commonly reported clinical symptoms in calves are decreased appetite and loss of fluid electrolytes, dehydration, metabolic acidosis, and hypoglycemia. Calves with coronavirus then developed complications caused by pneumonia. Thus, rotaviruses were the most dominant cause of viral diarrhea in newborn calves, especially in spring and winter.

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## CONCLUSION

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During the study, samples were taken in different seasons and analyzed by express testing. According to the results obtained, both rotaviruses and coronaviruses cause disease in calves belonging to Gilan Dairy Farms LLC, located in the north-west of our republic. According to the tables, rotaviruses are accompanied by a greater number of infections than coronaviruses, but the mortality rate from them is relatively lower. The spread of rotaviruses and coronaviruses among calves on farms becomes widespread in the spring, autumn and winter months of the year. A stressful change in environmental temperature can be a factor influencing the increase in the dynamics of the spread of diseases among animals.

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