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Clinical Signs of Seasonal Disease Dynamics in Calves Caused by Rotavirus and Coronavirus Infections

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Authors' contributions

This work was carried out in collaboration between both authors. All authors listed, have made substantial, direct and intellectual contribution to the work and approved it for publication. Both authors read and approved the final manuscript.

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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 Azerbaijan. An important roleour country. Their research is critical in the implementation of measures to combat rotavirus and coronavirus infections in infant calves. The primary goal of this study was to investigate the seasonal dynamics of accompanying diarrhea in calves caused by rotavirus and coronavirus infection. The research was carried out on farms located in the northwestern region of the Azerbaijan republic.

Fecal samples were used as material and tested by chromatographic immunoassay for the qualitative detection of rotavirus and coronavirus antigen.

The study revealed that the clinical signs of diarrhea in calves caused by rotavirus and coronavirus infections in farms located in the northwest region of the country are relatively similar. However, there was a relatively higher incidence of rotovirus infection, and mortality relative to coronavirus

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was at a lower level. The occurrence of secondary pneumonia in patients with coronavirus was noted as one of the main clinical signs. The study showed that the trend of infection and death from both diseases changes in different seasons of the year, which from season to season and, as a result, becoming massive causes serious damage to farms, becoming widespread in spring, autumn, and winter.

Keywords: Coronavirus; rotavirus; indigestion, seasonal; diarrhea; serotype.

1. 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 are more widespread during the winter months [1,2]. Seasonal changes repeated every year can cause changes in the biology of the susceptible organism and infections, causing epidemics [3]. However, other leading factors that can also lead to epidemics should be taken into account [4,5]. The coronavirus pandemic, which is a disease of viral origin that 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 reducing biological threats [6-8]. Rotavirus is a viral disease of calves characterized by dyspepsia, diarrhea, and dehydration [9,10]. At least two different 27 G and 37 P rotavirus genotypes have been identified that can lead to rotavirus infections [11]. Rotaviruses of the Reoviridae family are spherical viruses with a double-stranded, 1-segment RNA genome [12]. 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) [13-15]. The disease is common in developed countries with 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 (*Escherichia coli*), mortality among sick animals reaches 100% [16-19].

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 [20,21]. Betacoronaviruses are coronoviruses that cause SARS-CoV-2 in humans [22, 23]. 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 [24]. Calves become ill from day 10 to 8 weeks of age if they do not have inherited maternal antibodies [25, 26]. Pathogenic viruses that have spread in the environment in different serotypes, mutating and changing their serotypes, cause epizootics, epidemics, and even pandemics. Along with humans, coronaviruses also cause 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.

Rotavirus is one of the most widespread infectious agents worldwide, causing diseases, and it has been extensively studied in depth [27,28]. 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, the estimated prevalence was 24–47% in Sweden, 42% in outbreaks of diarrhea in the UK, and 37–47.4% in France [29-32].

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 goal of this study was to identify seasonal dynamics and clinical signs of rotavirus and coronavirus diseases in calves in the republic's northwestern region.

2. MATERIALS AND METHODS

2.1 Study Area and Climatic Characteristics

The study used a statistical database for 2017-2020 on diarrhea that spread among calves (ages 0-90) 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 Bazardyuzyu (4466 m). The area's snow cover lasts 45 days. The first autumnal frosts are observed in the 2nd decade of December and 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, and the number of days with temperatures above 10 degrees reaches 198 days. The 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.

2.2 Collected Samples

During this process, fecal samples taken from calves at different times of the year were checked by rapid testing (TMR nutrition calf test-5), 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; and in 2017: 357 samples.

2.3 Test Procedure

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

For testing according to the instructions (TMR Nutrition, calf test-5), 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 were dissolved in the solvent, and a pause of 30 seconds was made.
2. Dissolved samples are taken from the solution with a pipette.
3. Four drops of solution were added to each cell.
4. Results were read in 5-10 minutes.
5. Strips showing control and test lines were considered positive for rotavirus infection.
6. In clinical trials, other research methods were used using an electronic thermometer and a phonendoscope.

3. RESULTS

The 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 ^{0}C, and in the summer months it reaches a maximum of 29 ^{0}C. Such climatic conditions were regarded as more favorable for enteroviruses.

Tables 2 and 3 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.

As can be seen from Table 2, in the winter period of 2020, 99 heads of cattle were infected with coronovirus and rotavirus infections. Of these, 39 heads of cattle were infected with coronavirus and 60 heads of cattle with rotavirus. The deaths of 5 heads of cattle (with a percentage mortality of 13%) from coronavirus infection and 2 heads of cattle (with a percentage mortality of 3.3%) from rotavirus infection were recorded.

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

Climate of Gabala region												
Months	**January**	**February**	**March**	**April**	**May**	**June**	**July**	**August**	**September**	**October**	**November**	**December**
Maximum temperature ^{0}C (^{0}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 ^{0}C (^{0}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 2. The dynamics of rotavirus and coronavirus disease spread in Gilan Dairy Farms LLC by month in 2020

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 3. The month-by-month dynamics of the spread of rotavirus and coronavirus diseases in Gilan Dairy Farms LLC in 2019

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 9 (49%)
Rotavirus death	2 (6%)	2 (5%)	1 (4%)	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%)

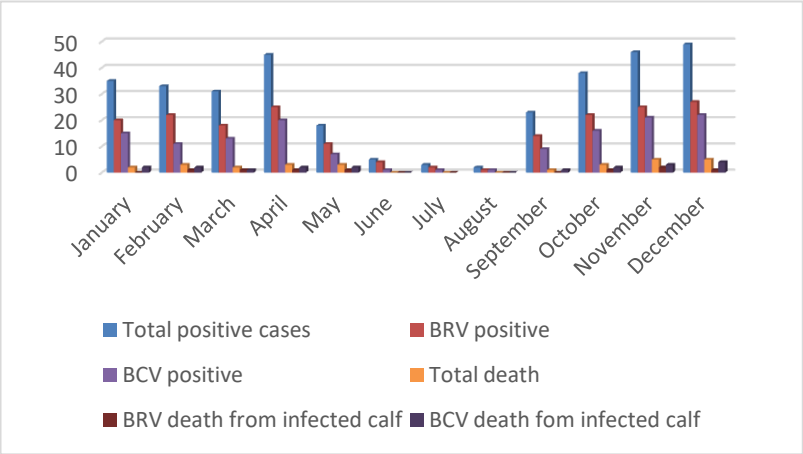

Fig. 1. Correlation of positive results in 2020 by months

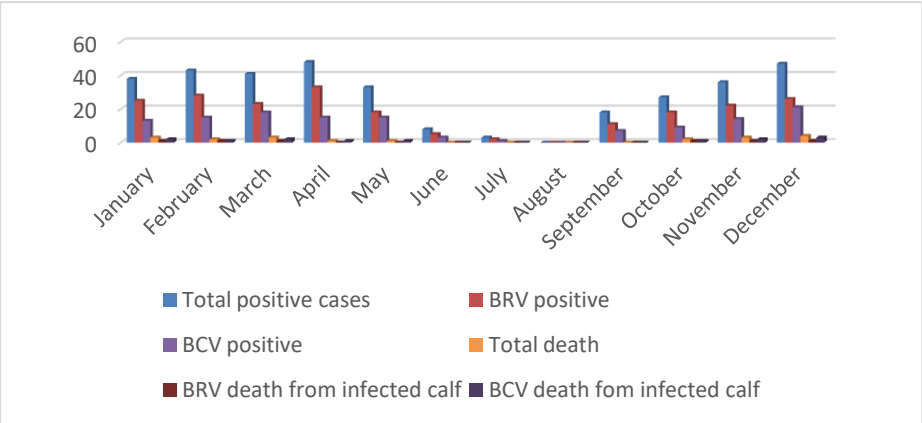

Fig. 2. The month-by-month correlation of positive results in 2019

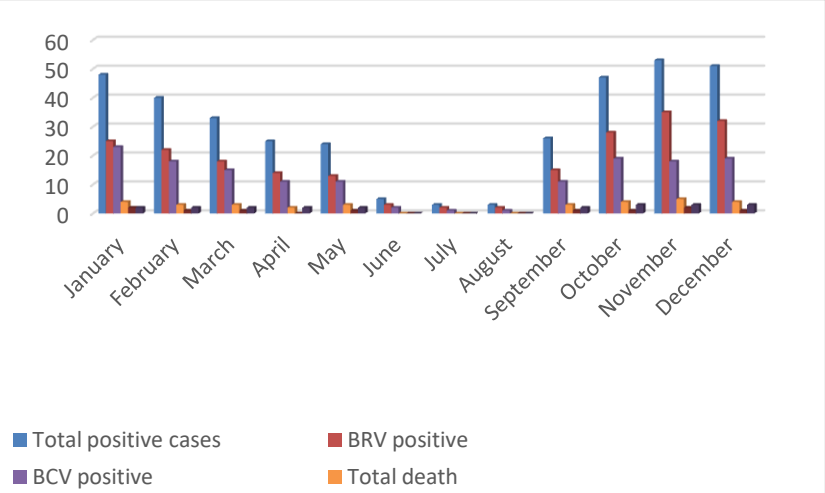

Fig. 3. Correlation of positive results in 2018 by months

In the spring of 2020, 68 cattle were infected with coronavirus and rotavirus infections, including 28 coronavirus-infected cattle and 40 rotavirus-infected cattle. The cattle losses from coronavirus infection were 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 coronovirus and rotavirus 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 affected 59 heads of cattle (with a mortality rate of 15%) for coronavirus infection and 74 heads of cattle (percentage with a mortality rate of 5.4%) for rotavirus infection.

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 month, including from rotavirus and coronavirus.

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 five cattle loss cases (10%) from infection with coronavirus and three cattle loss cases (3.9%). During the summer season, 89 heads of cattle were infected with coronovirus and rotavirus infections, of which 33 heads of cattle were infected with coronavirus and 56 heads of cattle with rotavirus. There were 2 cattle losses due to coronavirus infection (6% mortality) and 0 cattle losses due to rotavirus infection (0%). During the summer season, 8 heads of cattle were infected with the coronavirus (percentage mortality 0%), and 13 heads of cattle were infected with the rotavirus (percentage mortality 0%). In the autumn of 2019, 44 heads of cattle were infected with the coronavirus (percentage mortality: 13.6%), and 66 heads of cattle were infected with the rotavirus (percentage mortality: 4.5%).

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

4. DISCUSSION

To study the clinical symptoms and seasonality of rotavirus and coronavirus diseases in 2018-2021, 1385 samples were studied. Samples were taken from calves aged 0-90 days. According to Qinghe Zhu and others. studies, calf diarrhea is a multifactorial disease caused by a combination of infectious and non-infectious complex risk factors, which include enteropathogenic bacteria, viruses, environmental factors, immune status of the animal, genetic factors, nutrition, of course of labor, , childbirth, the structure of the nest of the calf, vaccination of the mother, and the state of health status calf [17]. In addition to these indicators, our studies have shown that the most common diarrhea of rotavirus and coronavirus origin is calf diarrhea, while the frequency of cases has different characteristics depending on the season and year. According to our observations, diarrhea caused by rotaviruses was registered most often at the age ofin children

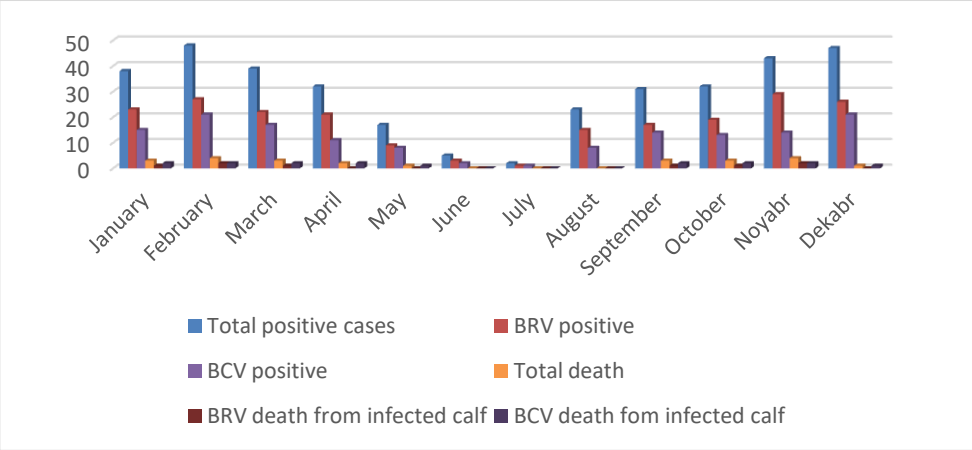

Fig. 4. Correlation of positive results in 2017 by months

Table 4. The month-by-month dynamics of the spread of rotavirus and coronavirus diseases in Gilan Dairy Farms, LLC in 2018

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 5. The month-by-month dynamics of the spread of rotavirus and coronavirus diseases in Gilan Dairy Farms, LLC in 2017

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%)	3 (12%)	3 (11%)	4 (10%)	1 (13%)

aged 5-–15 days. According to the literature, watery yellow diarrhea in calves is a clinical sign of rotaviruses [8,9,10,11, 33]. Depression, shock and dehydration occur more often in calves aged up to 5 days [6] of age [34]. The results presented by us showed that depression, decreased milking reflex, diarrhea and dehydration in the sick calf were among the main clinical signs. Calf diarrhea caused by coronaviruses has been reported in calves aged 2–21 days. Usually, the authors point out that the epithelial cells of the small and large intestine are sensitive to these viruses [12]. In addition, the virus [35]. Usually, a coronavirus infection causes watery diarrhea, and blood clots may appear in the stool. In our studies, decreased appetite and loss of fluid and electrolytes, dehydration, metabolic acidosis, and hypoglycemia were the most frequently registered clinical symptoms in calves. Calves sickinfected with the coronavirus developed complications in the form of pneumonia. Thus, rotaviruses were the most common viral cause of diarrhea in newborn calves, especially in the spring and winter. Clinical symptoms of rotavirus and coronavirus have been registered since the first 10 days of calf life, so it is important to keep both viruses under control.

5. CONCLUSION

During the study, samples were taken in different seasons and analyzed by express testing. As a result of the study of clinical signs of seasonal diseases dynamics in calves caused by rotavirus and coronavirus infection, both rotaviruses and coronaviruses cause disease in calves belonging to Gilan Dairy Farms LLC, located in the north-westnorthwest of Azerbaijan. According to the tables, rotaviruses are accompanied by a greater number of infections than caronaviruses, but the mortality rate from them is relatively lower. The spread of rotoviruses 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.

ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the author(s).

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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