

Trends of *Carnivore protoparvovirus 1* and *Canine morbillivirus* infection in domestic dogs in South Africa

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DECLARATION

I, Dr K Krishnalall, student number u15008526 hereby declare that this dissertation, “Trends of Carnivore protoparvovirus 1 and Canine morbillivirus infection in domestic dogs in South Africa,” is submitted in accordance with the requirements for the Master of Science degree at the University of Pretoria, is my original work and has not previously been submitted to any other institution of higher learning. All sources cited or quoted in this research paper are indicated and acknowledged with a comprehensive list of references.



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ABSTRACT

The study aimed to try and identify trends in both *Carnivore protoparvovirus 1* and *Canine morbillivirus* in South Africa to better equip veterinarians to respond to the overwhelming caseload we experience in this country.

Infection with *Carnivore protoparvovirus 1* often causes fatal haemorrhagic gastroenteritis in young puppies. The condition is exacerbated in dogs with poor immune status and insufficient supportive therapy. With supportive care, of which the most important intervention is fluid therapy, puppies can make a full recovery with no long-term side effects. Canine distemper, caused by *Canine morbillivirus* is a disease that affects multiple organ systems of puppies. This disease can also affect adults who do not have pre-existing immunity. It may cause characteristic neurological signs and may have a high mortality rate. Even with supportive therapy, the disease is often fatal. If the puppies are fortunate enough to survive, they are often left with long-term neurological deficits. The two diseases are common problems encountered in a rural setting.

Five years' worth of data collected from compulsory community service veterinarians working throughout South Africa were used to identify spatial and temporal patterns of these two diseases on a national scale. On a local scale, dogs that presented at Mdzananda Clinic based in Khayelitsha, a township in the Western Cape were used to determine the proportion of canine distemper and parvovirus cases encountered from July to December 2021 of the total animals seen.

The data showed a seasonal trend in *Carnivore protoparvovirus 1* infection with higher caseloads experienced in the summer months whereas *Canine morbillivirus* infections did not show any significant trend, but some provinces experienced higher caseloads than others. The data from the Mdzananda clinic followed the same trend in *Carnivore protoparvovirus 1* cases seen at a national and provincial level.

A trend can be seen with *Carnivore protoparvovirus 1* that may be the result of environmental factors such as precipitation and temperature and animal factors such as seasonal births, however, no such trends were seen with *Canine morbillivirus*

Keywords: parvovirus, distemper, canine, South Africa, epidemiology, distribution, rapid antigen test

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LIST OF ABBREVIATIONS

CCS	Compulsory community service
CPV	Carnivore protoparvovirus 1
CMV	Canine morbillivirus

1. Literature review

Carnivore protoparvovirus 1 (CPV), commonly known as parvovirus, is closely related to feline panleukopenia virus but is adapted to the canine host. Parvovirus is a widespread disease that affects mainly young puppies and causes acute and often fatal gastroenteritis (Ogbu et al., 2021) and sometimes myocarditis (Ezeibe et al., 2010). While adults may show clinical signs of enteritis, the disease is often not as severe and these animals often recover fully (Ezeibe et al., 2010). The virus affects both domestic and wild canids of both genders and all breeds.

Parvovirus is a small, non-enveloped, single-stranded, DNA virus that emerged in the late 1970s in companion animals but has since undergone mutations and now circulates globally (Rika-Heke et al., 2015). Non-enveloped viruses lack a lipid membrane (Dimitrov, 2004) and are more stable and resistant in the environment than enveloped viruses (Howie et al., 2008). Non-enveloped viruses survive well in the environment especially if organic matter such as faeces is present (Howie et al., 2008). Carnivore protoparvovirus 1 can survive for up to one year in the environment and is highly resistant to disinfectants. Special care must be taken to use the correct disinfectant such as glutaraldehyde and ensure a sufficient contact time with surfaces of at least ten minutes to inactivate the virus (Sykes, 2014).

There are three antigenic types of CPV, namely CPV-2a, CPV-2b and CPV-2c, which differ only slightly in a few of the viral capsid's amino acid structure known as the VP2 protein (Steinel et al., 1998). The VP2 is made up of 584 amino acids, changes in the sequence of these amino acids give rise to new variants. The CPV-2c type was first detected in Italy in 2000 and in Asia in 2012 (Temuujin et al., 2019). In recent years this strain has been detected more frequently in China and in 2019 the complete genome was sequenced (Li et al., 2019). A study was conducted from 1995 to 1998 to determine which strains were dominant in South Africa. All the samples were found to be either CPV-2a or CPV-2b and none were the original CPV-2 (Steinel et al., 1998). A study conducted in 2020 looked at CPV in north-central Nigeria and it was found that the prevalence was higher than what other studies reported, with the highest prevalence in Plateau State which is the hub of dog breeding in Nigeria (Ogbu et al., 2021).

The virus is transmitted via the faecal-oral route either by direct contact or indirectly via fomites with mechanical vectors such as rodents aiding in its spread. The virus can persist in the environment for long periods (Sykes, 2014).

There are many risk factors associated with CPV infection, including stress, immune status, and the presence of other infectious organisms (Ogbu et al., 2021). Season, rainfall, and temperature have been speculated as risk factors, but no definitive relationship has been established. In 2022 a study was conducted in India that looked at the oestrus patterns in street dogs, the humane society collected data over four years and found most pregnancies occurred in June to September (which is a post-monsoon season) and a peak in litters born followed soon after (Brill et al., 2022). The data in the study supports the existence of seasonal

oestrus in these dogs, and therefore a seasonal rise in the puppy population in winter through to early summer which in this case is February to March. The canine breeding cycle can explain the seasonal veracity as most litters of puppies are born between the spring and summer months (Rika-Heke et al., 2015) which in South Africa would be September to February. Not all the triggers for reproductive dynamics are known but factors such as photoperiod, rainfall and temperature as well as species-specific triggers may be involved (Brill et al., 2022).

A study by Shima et al (2015) demonstrated that there could be climatic factors associated with seasonality that could impact the local spread of the virus, with most cases occurring during the early dry season. This could be due to the negative effects the environmental stress of the dry season has on the immune system. If left untreated survival rates from CPV infection can be as low as 9.1% (Ogbu et al., 2021).

A drop in rainfall can have many negative effects on a population including water restrictions and a decrease in livestock production, however, an increase in rainfall can also have negative impacts. A study conducted in Australia between January 2010 and December 2013 investigated the relationship between rainfall and CPV in the country. A positive cross-correlation was found between cases of CPV and rainfall in the same month. Viral particles can remain in the environment for up to seven months which means the viral load can build up in the environment in the drier months and after rainfall, the water can spread the virus, leading to an increase in the number of cases seen (Rika-Heke et al., 2015).

The most used test for CPV in private practice is the in-house faecal antigen ELISA, a rapid test using a rectal swab as the sample. The test has a high specificity of nearly 100% and a sensitivity of 86% with false negative results being fairly common (Sykes, 2014). Unfortunately in the CCS context, there is not enough funding available to purchase these frequently and diagnosis is reliant upon clinical signs.

Canine morbillivirus (CMV) is a highly contagious virus that belongs to the genus Morbillivirus and the family Paramyxoviridae. It is a single-stranded, enveloped RNA virus that causes high mortality in dogs worldwide (Dorji et al., 2020). The survival of the virus in the environment depends on a multitude of factors one of them being temperature, with colder temperatures prolonging viability. Spread will also depend on the dispersion of virus particles either by rain or mechanical vectors such as people moving through the area. Contaminated environments and fomites serve as a source of distemper virus (Bucko and Gieger, 2019).

Canine distemper can affect a large range of carnivores and wildlife can serve as a reservoir if domestic dogs encounter wild species such as ferrets (which are becoming common household pets) (Zhang et al., 2020). The disease occurs commonly in younger dogs and can affect multiple organ systems. The highest fatality rates occur in puppies who are highly susceptible to the disease (Cunha et al., 2020) but CMV can also be contracted by vaccinated animals. The disease is readily spread within stray dog communities in South Africa (Dorji et

al., 2020). Townships house many free-roaming and often unvaccinated canines who serve as a maintenance population for both CPV and CMV (McRee et al., 2014).

Canine distemper is characterised by biphasic fever, gastrointestinal, respiratory, and nervous signs. While distemper causes a serous nasal discharge and mucopurulent ocular discharge, these are not necessarily seen in all cases which may lead to some cases not being diagnosed correctly. A leukopaenia, specifically lymphopaenia, may be seen and this can predispose affected animals to secondary bacterial infections (Degirmencay et al., 2021).

There is a rapid in-house ELISA test for CMV commonly used in private veterinary practice. This CMV rapid antigen test has a sensitivity of 92% and specificity of 95% but false positives because of vaccination are possible (Bucko and Gieger, 2019).

Both CPV infection and distemper are major causes of morbidity and mortality in our canine companions, but vaccines are available for these diseases. There are many different vaccines and vaccine protocols available but adherence to the recommended schedule can have a profound effect on the success rate of vaccination (Cunha et al., 2020).

South Africa experiences differences in rainfall due to the El Nino–Southern Oscillation which means that rainfall patterns can be difficult to predict. Summer and winter rainfall periods can be defined, with summer rainfall occurring from October to April, which accounts for the rainy season in most of the country. The winter rainfall period is from May to September and occurs in the Western Cape (Marumbwa et al., 2019)

2. Problem statement

Carnivore protoparvovirus 1 and canine morbillivirus are diseases that affect many domestic dogs in South Africa daily. Knowledge about these diseases and in particular if they are more prevalent at certain times of the year will allow veterinarians to be better equipped to handle the caseload.

2.1. Research question

Are there any trends of CPV and CMV infection in domestic dogs in South Africa?

2.2. Aims

- Determine if there are trends of CPV and CMV infection in domestic dogs in South Africa using the data collected from compulsory community service veterinarians from 2016 to 2020.
- Determine if there is a trend in the data from our local study and compare it to the national study data.
- Determine if there are differences in the trends seen between provinces.

2.3. Objectives

- Complete the local study between June and December 2020 and analyse it for trends. in Excel to create graphs for better visual representation.
- Analyse the data obtained on a national level in excel to create graphs and create maps in ArcGIS to better visualise the data.
- Compare the graphs created for each province to ascertain if there are differences or similarities.
- If any trends are visible write a short communication to share with the veterinary community

3. METHODOLOGY

3.1. Literature review

A literature review was performed by identifying key terms and synonyms. The key terms were “epidemiology”, “Carnivore protoparvovirus 1” and “Canine morbillivirus”. For epidemiology, alternate words such as “prevalence”, “incidence”, “occurrence” and “distribution” were considered, for Carnivore protoparvovirus 1 the term “parvo” was used as well and for Canine morbillivirus, the other search term was “distemper”. When the search term “parvo” was used it brought up many articles about parvo of different species and in these cases, the search was filtered by adding the term “canine” or “dog”.

Articles were found using CABI and Web of Science, available via the University of Pretoria library website. Broad search terms such as “parvo or parvovirus” or “distemper” and “prevalence or incidence or occurrence” and the Boolean operators and/or were used to combine terms or widen the search parameters. All the articles which had titles or abstracts that contained both key search terms were downloaded and reviewed.

Web of Science is a very user-friendly search engine but unfortunately, many of the article links did not work or the article did not have an online version available, so the online search platform ScienceDirect was also employed. ScienceDirect has the added benefit that all the articles found in your search have links to direct downloadable PDF copies.

To gather the required data on South Africa the terms “South Africa geography”, “South Africa and climate” and then finally “South Africa and rainfall”. To determine if there were articles that already demonstrated a correlation between parvo or distemper and weather or climate the search terms “parvo and climate” and “parvo and rainfall” were used on the ScienceDirect database. The same strategy was used with the terms “distemper and climate” and “distemper and rainfall”. The average temperatures for each month in South Africa were obtained from the website ‘climateknowledgeportal.worldbank.org’ was used and this gave the monthly average temperatures in South Africa using data from 1991 to 2020.

3.2. National study

Compulsory community service (CCS) is a governmental programme where newly graduated veterinarians are placed in a variety of regions and perform the function of state veterinarians for a year. Each province submits a list of locations to the government that needs a veterinarian and then the government will allocate each province a number of students. The places vary from year to year based on the number of students available and which locations take priority. The number of medical and surgical cases seen at each (CCS) clinic and the GPS

coordinates of each clinic were available electronically from the online CCS reporting system LimeSurvey (SourceForge, USA).

The GPS coordinates given did not relate to a specific clinic but only to the province in which that clinic was located. The data were collected using forms sent to all the CCS veterinarians in the country every month to complete as an online survey. No steps were taken to locate possible duplicates and remove them, each monthly survey can only be completed once by each veterinarian so duplicates were unlikely. The data were exported to a Microsoft Excel spreadsheet (Microsoft, USA) to make use of the pivot function. The number of CPV and CMV cases seen was based on clinical findings, there were no inclusion or exclusion criteria. These were all the suspected cases that the veterinarian saw, and dependent on their judgement, there was no definition of 'case'. When finding the temperature and rainfall data to compile Table 7.1 and Table 7.2, monthly aggregates for rainfall were able to be found but unfortunately temperature aggregates could only be found for yearly intervals. This does affect the precision of the results obtained.

3.3. Local study

3.3.1. Sample size

A sample size to estimate the prevalence of these diseases in the Khayelitsha dog population with a 99% level of confidence and a precision of 0.04 was calculated: $n = Z_{\alpha}^2 * P * (1-P) / d^2$ (Equation 1), where n = sample size, $Z = (1 - \alpha/2)$ percentile of a normal distribution = 2.576, P = expected prevalence and d = precision of the estimate and equal to half the desired length of a confidence interval = 0.04 (1/5th of the estimated prevalence)). The expected prevalence of these diseases can vary between 0.2 and 0.8 (Böhm et al., 2004) depending on the country and period during which the study is conducted. We used an estimated prevalence of 0.2 which resulted in a larger sample size but was also a more accurate representation of true prevalence. Using the above formula and values we calculated a sample size of 664.

It was expected that the clinic would use between 8 - 15 snap tests per day and estimated that there would be approximately 100 CPV and CMV cases a month as in previous years. It was expected that within six months it was possible to see 600 CPV and CMV cases which did not include all the routine cases seen.

Our sample size was constrained by the budget available for this project and the amount of Bionote (Bionote, South Korea) rapid antigen tests that could be purchased and supplied to the clinic. We were able to purchase 430 parvo test kits and 320 distemper test kits. Only suspected cases were tested, animals who came in for other ailments were not tested using the rapid tests. There were some cases where the animals seen by the clinic were suspected of having both CPV infection and distemper and in those cases both tests were performed.

3.3.2. Inclusion criteria

The inclusion criteria were all animals that visited the Mdzananda Clinic based in Khayelitsha, a township in the Western Cape, from July to December 2021 and showed clinical signs of disease. For CPV this included all animals, regardless of their age, who presented with vomiting, diarrhoea, lethargy, or anorexia. Canine distemper is often more difficult to diagnose, so the inclusion criteria included animals with nasal or ocular discharge, nervous symptoms such as circling, head tilts or nystagmus, or animals that presented with coughing or hyperkeratosis of any age group.

3.3.3. Exclusion criteria

Clinically healthy animals or animals that presented to the clinic that showed clinical signs other than those commonly seen in CPV and CMV.

3.3.4. Diagnosis

If the animal showed clinical signs of CPV infection, the Rapid CPV antigen test (Bionote, Korea) was performed which entailed using a rectal swab to collect the sample. This sample was then added to the specimen tube that contained 1 ml of the assay diluent. After the sample was mixed with the diluent, the disposable dropper was used to add four drops of the diluent into the sample hole in the reading device. The reading device was placed on a flat surface to ensure accurate results and read after five to ten minutes of incubation. If the test was not read after 20 minutes, it was discarded, and a new test was performed as the result of the former test was no longer valid. Figure 3.1 demonstrates how the test is to be read. A line at the 'test' section and well as the 'control' section indicates a positive result. if there is only a line at 'control' the result is negative and if no line appears at 'control' the test is invalid.

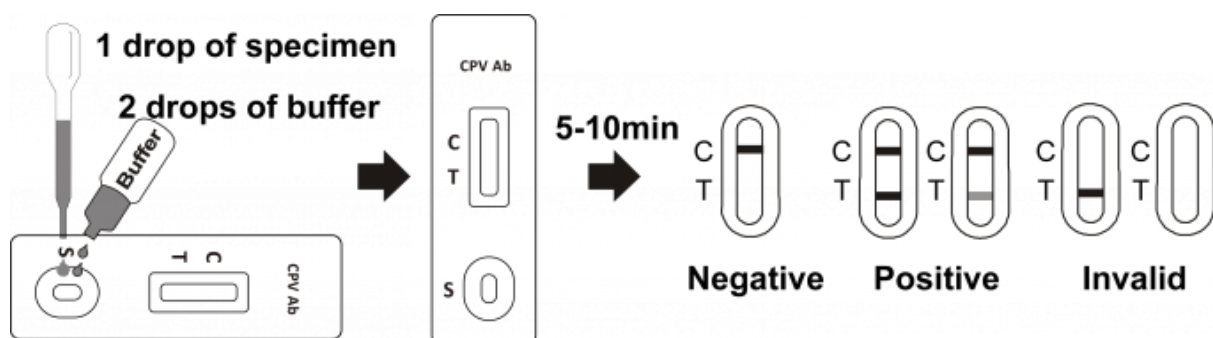


Figure 3.1. A schematic representation of how to use the *Carnivore protoparvovirus 1* rapid test and interpret the result. Image source: Hangzhou AllTest Biotech CO., LTD. Available at: <https://www.custom-monoclonalantibody.com/sale-10980996-cpv-rapid-antibody-test-cassette-serum-plasma-with-24-months-self-time.html> (Accessed: January 23, 2023).

If an animal showed clinical signs of distemper, a conjunctival swab was collected. This sample was mixed with the diluent provided in the Rapid CMV antigen test kit (Bionote, Korea) and added to the test device. The procedure is the same as that of the CPV test except that the specimen tube only contains 0.3 ml of diluent. The test device was read after five to ten minutes of incubation. The CMV rapid test is read the same way the CPV test is read. The test results were recorded on the spreadsheets the clinic uses to log their cases, it is highly unlikely that duplicates were created as only the person who examined logged the case.

3.4. Analysis

The number of CPV and distemper cases was defined as the cases the CCS thought were positive based on clinical signs of the respective diseases. These were then compared to the number of canine medical cases seen. The total medical cases seen did not include vaccinations, surgeries, or feline patients.

The spatial analysis was performed using ArcGIS 10.6 (ESRI, USA). In Excel, all the values in the array (province | monthly CPV/CDV averages) were taken, the maximum and minimum values determined and then the percentiles in 10% intervals were calculated to determine the break values of the classes.

Correlation coefficients for CPV and CMV and temperature and rainfall were done in Microsoft Excel using the function CORREL (array1, array2), this function was performed on the datasets contained in Table 7.1 and Table 7.2 in the Appendix.

The Mdzananda Clinic did not have any data capture software in place and all the data of animals who presented at the clinic were recorded in hospitalization charts and logged manually in books before the data were transferred to an Excel spreadsheet (Microsoft, USA).

4. RESULTS

4.1. National level

The CCS data from 2016 to 2020 showed that there was an increase in the number of CPV cases seen as well as a slight decline in CMV cases (Figure 4.1). Table 7.1 and Table 7.2 in the Appendix show the total number of medical cases as well as CPV and CMV cases seen that were used to calculate the proportions seen in Figure 4.1 and Figure 4.2 below.

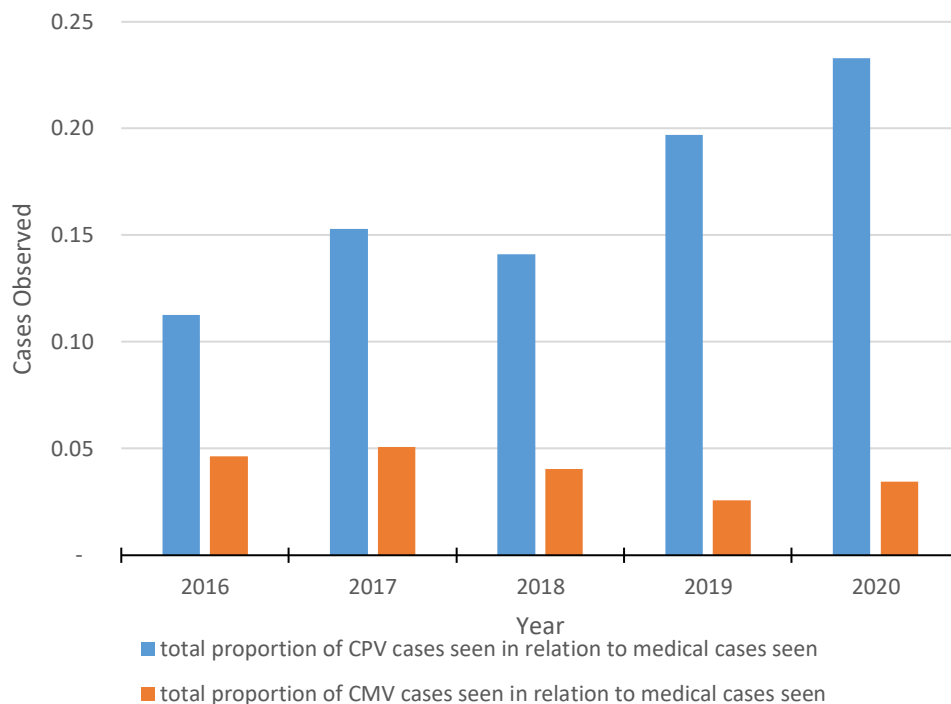


Figure 4.1. The average proportion of *Carnivore protoparvovirus 1* and *Canine morbillivirus* cases seen in relation to the overall proportion of medical cases seen in South Africa between 2016 and 2020. The number of total medical cases seen in 2016 = 26,920, 2017 = 37,305, 2018 = 45,791, 2019 = 59,647 and 2020 = 60,708.

The mean CPV and CMV cases were categorised by month of the year (Figure 4.2). The CPV cases decreased between April and August with the lowest recording in May to July, i.e., during winter. Most of the country experiences its rainy season in the summer months which is from November through to March. The CMV cases did not fluctuate throughout the year and did not show any visible trend. The same trend occurred every year during the study period, but some months stand out. In January 2019 the first big spike in the number of CPV cases occurred, followed by peaks in January 2020 and December 2020. The data used to calculate the different correlation values can be found in Figures 6.1 and 6.2 in the Appendix.

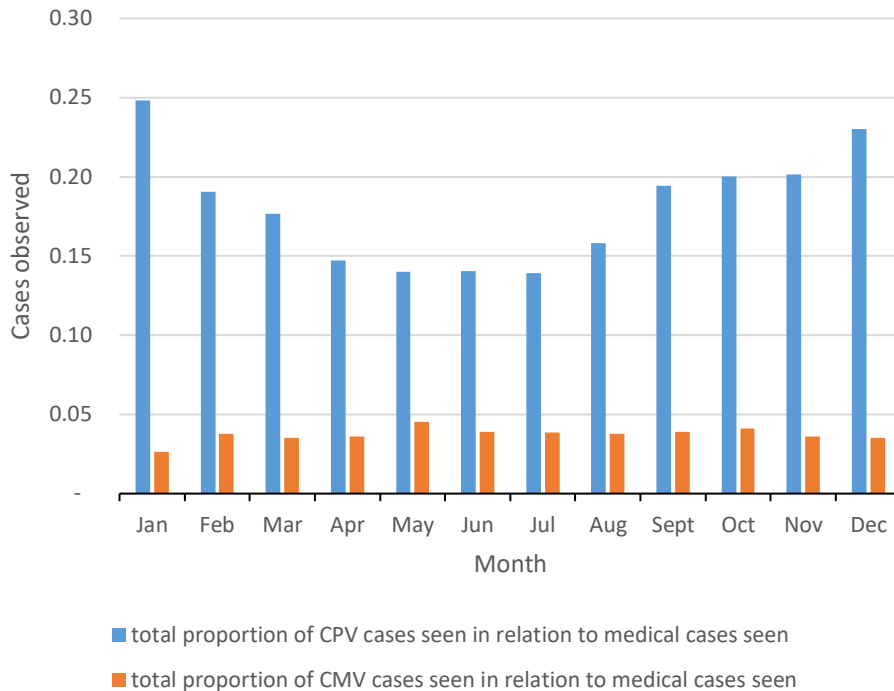


Figure 4.2. The average proportion of *Carnivore protoparvovirus 1* and *Canine morbillivirus* cases seen in relation to the overall proportion of medical cases seen in South Africa monthly between the period of 2016 and 2020.

The correlation coefficient between monthly rainfall and CPV cases was 0.35 (weak positive correlation) (Figure 4.3).

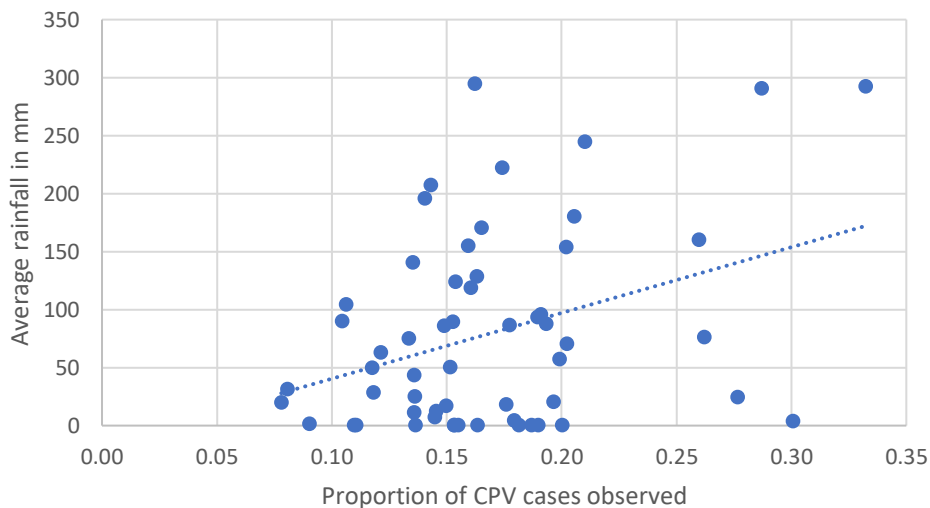


Figure 4.3. A scatter plot of the average rainfall the country received in mm in relation to the proportion of *Carnivore protoparvovirus 1* cases to total medical cases seen in the same period. The dotted line represents the trend line demonstrated in this dataset.

The correlation coefficient between rainfall and CMV caseload was -0.21 (weak negative correlation) (Figure 4.4).

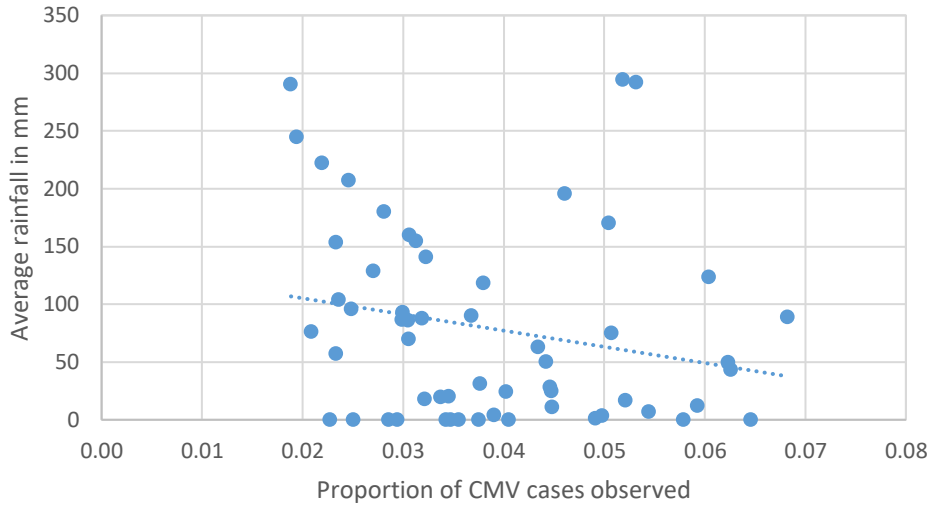


Figure 4.4. A scatter plot of average rainfall the country received in mm in relation to the proportion of *Canine morbillivirus* cases to total medical cases seen in the same period. The dotted line represents the trend line demonstrated in this dataset.

The correlation coefficient between caseload and temperature was determined. The correlation coefficient between CPV cases and temperature was 0.804, a strong positive correlation (Figure 4.5).

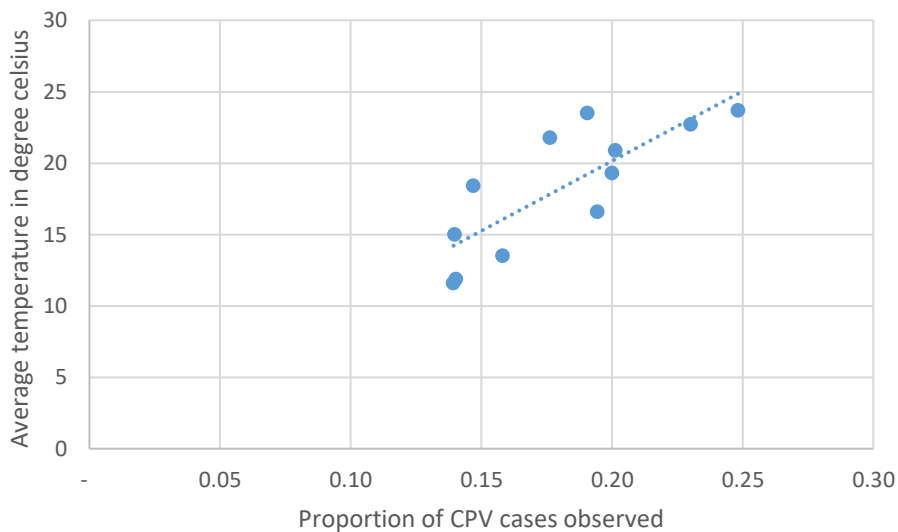


Figure 4.5. A scatter plot showing the average temperature in the country in degrees Celsius in relation to the proportion of *Carnivore protoparvovirus 1* cases to total medical cases seen in the same period. The dotted line represents the trend line demonstrated in this dataset.

The correlation coefficient between CMV and temperature was -0.573, a moderate negative correlation (Figure 4.6).

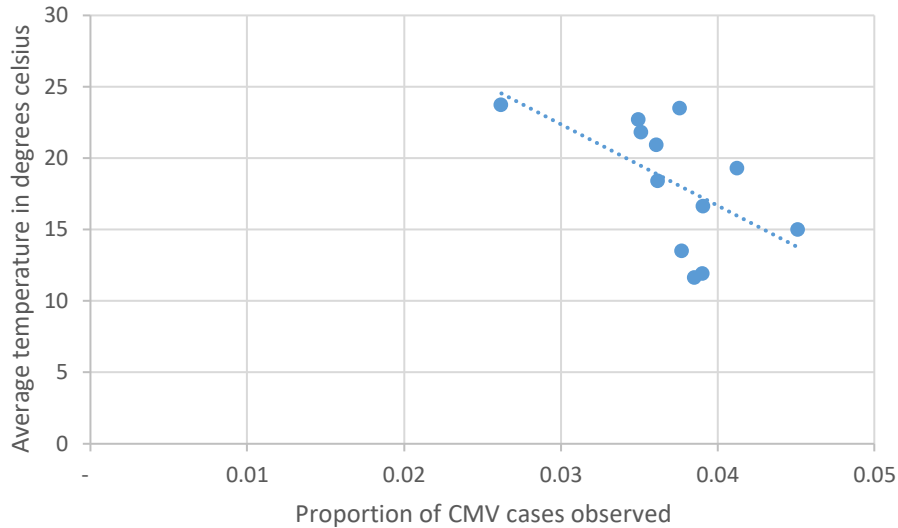


Figure 4.6. A scatter plot of the average temperature in the country in degrees Celsius in relation to the proportion of *Canine morbillivirus* cases to total medical cases seen in the same period. The dotted line represents the trend line demonstrated in this dataset.

4.2. Provincial level

The data analysed at a provincial level showed a similar trend seen on a national level, with only small differences in the months when the number of cases increased or decreased. In most of the country, the highest CPV caseload occurred in December and January with the lowest caseload between April and June (Figure 4.7). For CMV there was no obvious trend, however, some provinces such as Limpopo and the Northern Cape had the greatest caseload between May and September (Figure 4.8). Individual provincial data for monthly and yearly CPV and CMV cases are shown in the Appendix in Figure 7.1 to Figure 7.14. The only outlier in this data set is the Western Cape province (Figure 7.15 and Figure 7.16) which shows a CPV peak from November to February which did not coincide with the rainfall pattern in that province.

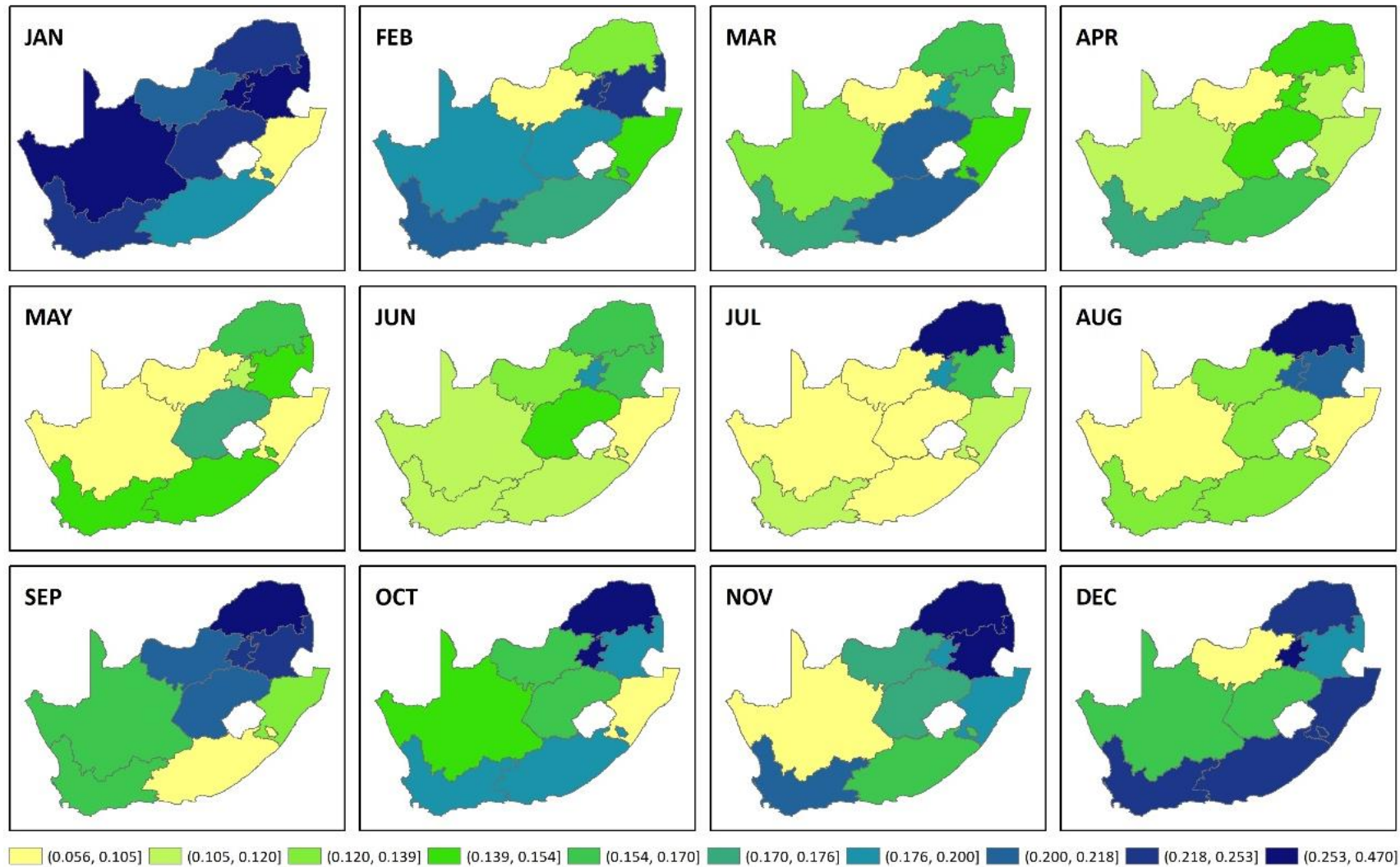


Figure 4.7. *Carnivore protoparvovirus 1* monthly proportion of CPV cases in relation to medical cases seen using yearly averages from 2016 - 2022. The minimum value was 0.056 and the maximum 0.470, the intervals were calculated as 10% percentiles and colours were assigned to each interval.

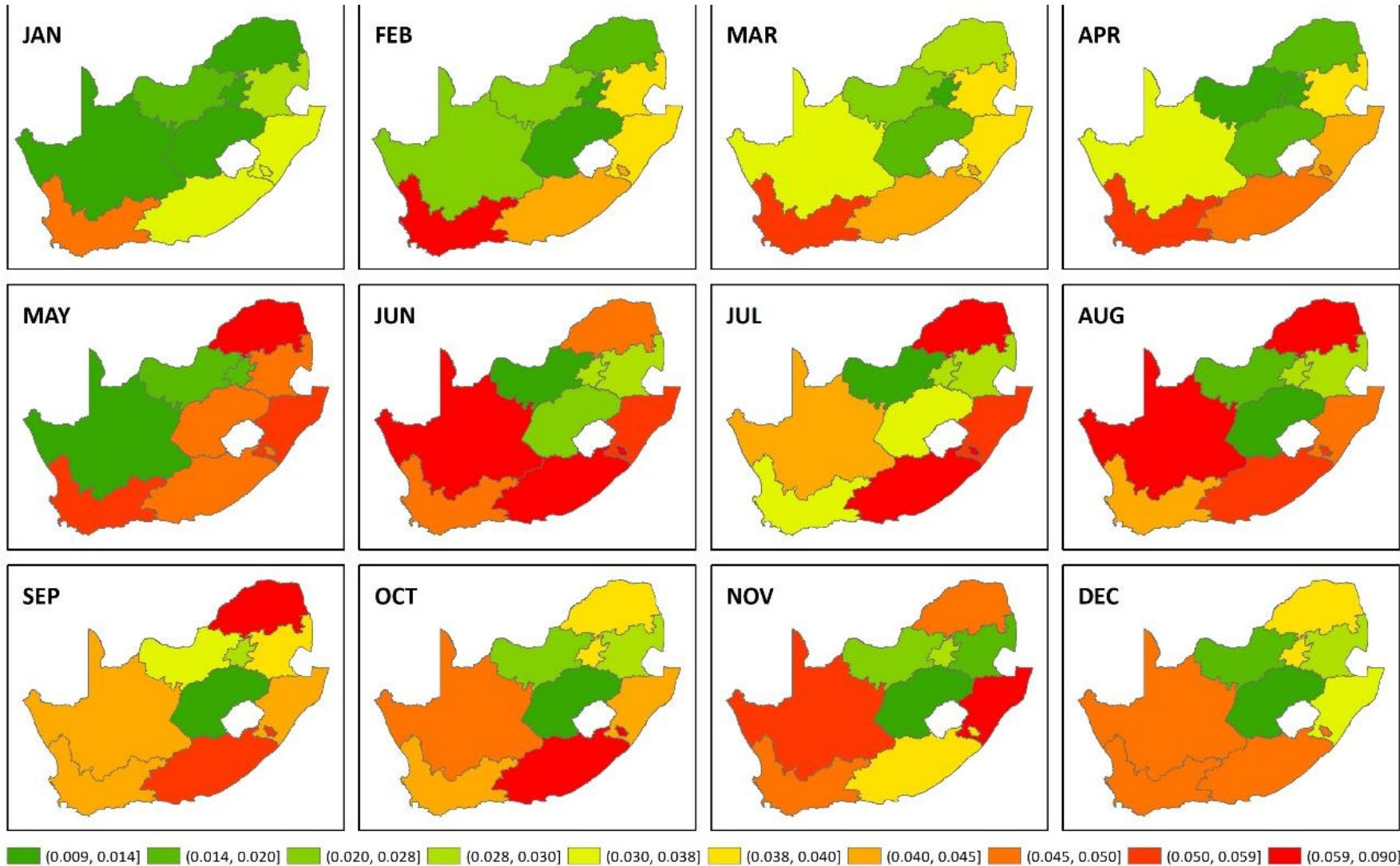


Figure 4.8. *Canine morbillivirus* monthly proportion of CMV cases in relation to medical cases seen using yearly averages from 2016 - 2022. The minimum value was 0.009 and the maximum 0.090, the intervals were calculated as 10% percentiles and colours were assigned to each interval.

4.3. Local scale

Khayelitsha is a township in the Western Cape province located on the Cape Flats this area is known to have warm dry summers and cool damp winters. The Mdzananda clinic which is located here was used to conduct the study that ran from July through to December.

The numbers of CPV and CMV cases seen monthly were analysed in relation to the number of animals seen that month. Of the total 360 CPV suspected cases seen at Mdzananda Clinic, 249 were positively diagnosed using the snap test, therefore the test positivity for CPV was 69%. Of the 155 CMV-suspected cases seen at Mdzananda clinic, 64 were positively diagnosed using the snap test, therefore the test positivity for CMV was 41% (Table 6.3). Figure 4.9 shows an increase in cases in October and November at the Mdzananda clinic which coincides with Spring in South Africa.

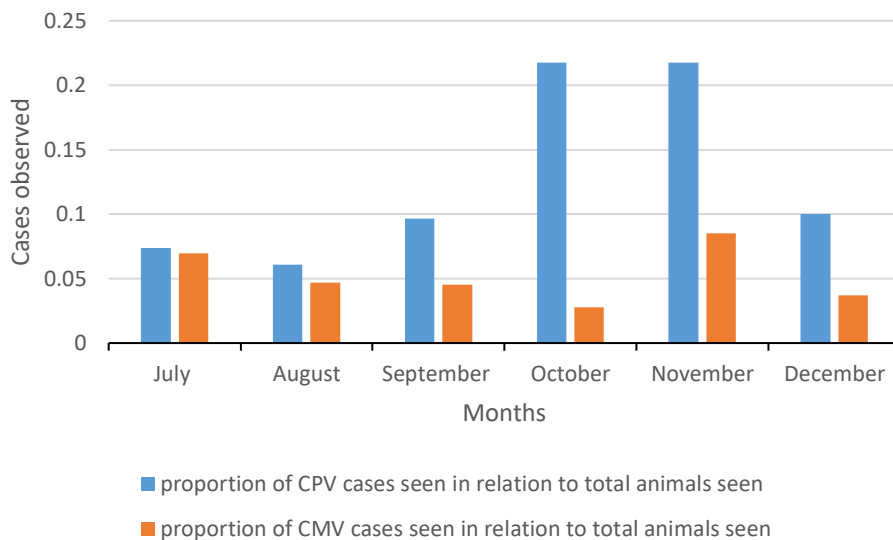


Figure 4.9. The proportion of *Carnivore protoparvovirus 1* and *Canine morbillivirus* cases seen in relation to the total proportion of animals seen at Mdzananda clinic from July 2021 to December 2021.

This matches what is on a provincial level as well as on a national level. There is more variation seen in the CMV cases throughout the study period.

5. DISCUSSION

Carnivore protoparvovirus 1 is a small non-enveloped virus that results in haemorrhagic gastroenteritis in young dogs (Ogbu et al., 2021). A non-enveloped virus is not covered by a lipid layer (Dimitrov, 2004) which means the virus is more resistant to stressors such as heat stress and disinfectants. This enables CPV to survive longer in its environment without a host (Howie et al., 2008). Even small amounts of infected faeces can harbour the virus and as such contaminate the environment. Environmental factors such as rainfall can then aid in the spread of the disease but help to disseminate viral particles and increase the contaminated surface area. It is also possible that dogs to some degree have a seasonal reproductive season with the main contributing factors being photoperiod, temperature, and rain (Brill et al., 2022). A peak in puppy births in the early summer months would lead to a larger pool of animals who could potentially become infected with diseases such as CPV.

For most (77%) of the provinces in the country, we saw an increase in cases in the summer months, which are the high rainfall periods except for the Western Cape. The peaks in CPV in January 2019, January 2020 and December 2020 coincided with high precipitation of 100 – 200 mm in a large section of the country, whereas in May 2020, the lowest CPV caseload was recorded, and most of the country received 0 – 10 mm of rainfall that month with a few parts of the Western Cape receiving 25 – 50 mm of rain. The correlation coefficient between monthly rainfall in South Africa and monthly CPV cases was 0.35 (Table A1), while this is a positive correlation it is not a strong correlation. A study conducted in Australia between 2010 and 2013 did find positive cross-correlation between CPV and rainfall in the same month (Rika-Heke et al., 2015).

The correlation between CPV cases and the average monthly temperature in the country was also calculated (Table A2) and resulted in a correlation coefficient of 0.804 which is a strong positive correlation. Higher proportions of suspected CPV cases were observed during the warmer months in this dataset. The correlation coefficient between temperature and CPV is much stronger than that of rainfall and CPV so temperature could be the bigger influence on caseload. This allows veterinarians to know when to have more test kits and consumables available to manage a rise in cases.

Limpopo Province and the Western Cape Province are the only two provinces where the highest number of cases did not coincide with the months with the highest rainfall. However, these provinces still have their temperature peaks at the same time as the rest of the country. This variability could be due to the different biomes that make up the provinces and the wide variety of different climatic conditions within the province. Unfortunately, due to the inaccuracy of the coordinates in the CCS data, we could not plot the cases to look at the distribution within the different biomes. The exact locations of the CCS posts are often difficult to locate on software such as Google maps because they are mostly located in rural

areas that have no access to other veterinary care. The CCS posts often vary from year to year depending on the number of students available and where provincial directors think the students are needed the most.

Limpopo had the highest number of CPV cases in relation to the number of canine medical cases seen in the whole country with up to 47% of cases on average in September, on average no other province has higher than 35% of their canine medical cases diagnosed as CPV. More studies should be done to try and determine why Limpopo is so greatly affected by CPV.

In the Mdzananda study, we noted that the CPV peak in October and November were similar at provincial and national levels. The Western Cape has very erratic weather it is often very windy and has rainfall throughout the year, but most of it is in winter. At the clinic, 69% of animals who had clinical signs of CPV tested positive with the rapid antigen test. The test is highly specific but not very sensitive which means a negative test result does not rule out the disease and that the disease was most likely under-diagnosed. This means that on a national level clinics that use these rapid tests may be underdiagnosing the disease, and in CCS conditions where the diagnosis is based on clinical signs we could be under- or over-diagnosing.

Canine morbillivirus is an enveloped virus and as such does not survive long in the environment (Dorji et al., 2020). The virus thrives better in cold temperatures (Bucko and Gieger, 2019) and would not last long in South Africa's summer months. Looking at the monthly data for South Africa there does seem to be a trend of lower CMV caseloads seen between the periods of December to January, this is the height of summer and temperatures can be anywhere up to 32 – 38 degrees Celsius. This virus which thrives better in cooler conditions would then survive longer in our environment in the winter months and hence infect more animals resulting in a higher caseload. The correlation coefficient between monthly rainfall in South Africa and monthly CMV cases was -0.21 (Table A1). This is a weak negative correlation which would mean that as rainfall increased, we would see a decrease in CMV cases. The correlation coefficient between CMV and the average monthly temperature in South Africa was -0.573 (Table A2) which is only a moderate negative correlation. In some cases, a temperature rise coincided with a decrease in the number of CMV cases seen but we cannot say if these two events are related. The trend we see in more cases being present in the winter months tells us we should try to increase our vaccination drives in the months leading up to that to try and improve the immunity of these at-risk animals. There is a lack of data to see if the trends in this virus can be seen in other African countries.

The Mdzananda data did not exhibit any trends in the number of CMV cases throughout the six-month trial period and only 41% of animals suspected of having CMV had a positive rapid test result (Table 7.3). The CMV rapid antigen test is both highly sensitive and highly specific meaning that a positive result means that the animal is infected, and a negative result can rule out disease. This result suggests that if the diagnosis is based solely on clinical signs,

distemper can be overdiagnosed. In CCS conditions where there are not always tests available clinical signs are relied upon to make a diagnosis. Clinics using these rapid tests should be able to make an accurate diagnosis of CMV.

During this project, we encountered some challenges, one being that some of the test kits we supplied to the Mdzananda clinic were stolen halfway through the study. It was estimated that 170 parvo snap tests (17 kit boxes) and 110 distemper snap tests (11 kit boxes) were stolen from the clinic premises. Fortunately, more funding was secured and a further 15 boxes of parvo snap tests and 12 boxes of distemper snap tests (150 and 120 tests respectively) could be provided to the clinic for the duration of the study. The number of test kits we were able to buy was ultimately limited by the funding available.

Mzananda is only one small clinic in the Western Cape province and as such, it is difficult to make generalized statements about the province based solely on this. If we had more resources more clinics could be used in the study making the results more accurate.

Another problem encountered was the inaccuracy of the GPS locations of the different community service posts around the country. As a result, the locations could not be mapped as accurately as we would have liked to see hotspot areas within each province as we originally intended, however, the season variation within each province and the country can still be demonstrated.

The data from the CCS veterinarians provided good insight into the current situation in South Africa but it was not the whole picture. It does not consider the animals seen at private practices in the area or other animal welfare organizations that treat animals but do not have a CCS veterinarian assigned to them. There are also rural communities in South Africa that unfortunately don't have any CCS vet facility nearby and do not receive any veterinary support. With the limited number of veterinarians in the country and constrained resources, it is not feasible currently to have services available that everyone in the country has access to.

Another problem with the CCS data is that there was no definition or criteria for what a case was, it was based entirely on the opinion of the veterinarian at the specific post. If a new study were to be done in the future precise inclusion and exclusion criteria should be given to all participating members.

In conclusion, we can see that CMV and CPV are multifaceted diseases with many factors influencing the spread of disease. We can see that CPV peaks in the summer months and should start vaccination campaigns in late winter to give puppies the best chance of having an adequate immune response and not succumbing to the virus. Veterinarians should be on high alert in the summer months and expect an influx in CPV cases. More research should be conducted to find the cause of the CMV caseload in Limpopo province. Government should

invest more of its resources in vaccination campaigns and educating owners on the importance of vaccination.

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7. APPENDIX

Table 7.1. The proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) infections in South Africa in relation to total medical cases and the average monthly rainfall (mm) in the country from 2016 to 2020.

Month Year	Medical cases seen	CPV cases seen	CDV cases seen	CPV cases seen/ Medical cases seen	Average rainfall	CDV cases seen/ Medical cases seen
Apr-16	1381	207	72	0.15	17	0.05
May-16	1336	162	58	0.12	63	0.04
Jun-16	4589	358	155	0.08	20	0.03
Jul-16	3926	317	148	0.08	31	0.04
Aug-16	3805	344	187	0.09	1	0.05
Sep-16	4533	606	230	0.13	75	0.05
Oct-16	4284	623	254	0.15	12	0.06
Nov-16	1832	280	125	0.15	89	0.07
Dec-16	762	81	18	0.11	104	0.02
Jan-17	2577	426	130	0.17	170	0.05
Feb-17	2873	466	149	0.16	295	0.05
Mar-17	2824	428	125	0.15	50	0.04
Apr-17	2588	352	162	0.14	43	0.06
May-17	3385	398	211	0.12	50	0.06
Jun-17	3173	348	205	0.11	0	0.06
Jul-17	3224	381	144	0.12	28	0.04
Aug-17	2693	413	156	0.15	0	0.06
Sep-17	3098	556	121	0.18	4	0.04
Oct-17	3900	581	119	0.15	86	0.03
Nov-17	2763	444	105	0.16	119	0.04
Dec-17	1370	277	32	0.20	154	0.02
Jan-18	3836	727	115	0.19	93	0.03
Feb-18	4202	647	254	0.15	124	0.06
Mar-18	4208	592	194	0.14	196	0.05
Apr-18	3751	392	138	0.10	90	0.04
May-18	4113	560	184	0.14	25	0.04
Jun-18	3599	522	196	0.15	7	0.05
Jul-18	3477	473	156	0.14	11	0.04
Aug-18	4207	465	158	0.11	0	0.04
Sep-18	3759	513	129	0.14	0	0.03
Oct-18	4583	620	148	0.14	141	0.03
Nov-18	3739	596	117	0.16	155	0.03
Dec-18	2034	291	50	0.14	207	0.02
Jan-19	6106	1753	115	0.29	291	0.02
Feb-19	5407	1137	105	0.21	245	0.02
Mar-19	5954	1186	139	0.20	57	0.02
Apr-19	4928	858	108	0.17	222	0.02

May-19	5149	790	183	0.15	0	0.04
Jun-19	5303	962	133	0.18	0	0.03
Jul-19	5099	790	150	0.15	0	0.03
Aug-19	4787	895	109	0.19	0	0.02
Sep-19	5328	1048	184	0.20	20	0.03
Oct-19	4154	841	127	0.20	70	0.03
Nov-19	3106	601	99	0.19	88	0.03
Dec-19	1367	261	34	0.19	96	0.02
Jan-20	6793	1780	142	0.26	76	0.02
Feb-20	6192	1272	174	0.21	180	0.03
Mar-20	4618	819	138	0.18	87	0.03
Apr-20	2291	374	62	0.16	129	0.03
May-20	3224	527	112	0.16	0	0.03
Jun-20	4442	782	143	0.18	18	0.03
Jul-20	4839	919	196	0.19	0	0.04
Aug-20	5597	1121	160	0.20	0	0.03
Sep-20	5366	1484	216	0.28	24	0.04
Oct-20	7106	2137	354	0.30	4	0.05
Nov-20	6658	1729	204	0.26	160	0.03
Dec-20	3476	1155	185	0.33	292	0.05

Table 7.2. The proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) infections in South Africa in relation to total medical cases and the average monthly temperature (°C) in the country from 2016 to 2020.

Month	CPV cases seen	CMV cases seen	Medical cases seen	Total proportion of CPV cases seen in relation to medical cases seen	Total proportion of CMV cases seen in relation to medical cases seen	Average temperature (°C)
Jan	4907	518	19760	0.25	0.03	23.7
Feb	3640	718	19089	0.19	0.04	23.5
Mar	3189	635	18072	0.18	0.04	21.8
Apr	2255	555	15324	0.15	0.04	18.4
May	2529	816	18076	0.14	0.05	15
Jun	3045	847	21685	0.14	0.04	11.9
July	2982	825	21404	0.14	0.04	11.6
Aug	3336	796	21087	0.16	0.04	13.5
Sep	4496	904	23122	0.19	0.04	16.6
Oct	4890	1008	24422	0.20	0.04	19.3
Nov	3713	666	18437	0.20	0.04	20.9
Dec	2112	321	9173	0.23	0.03	22.7

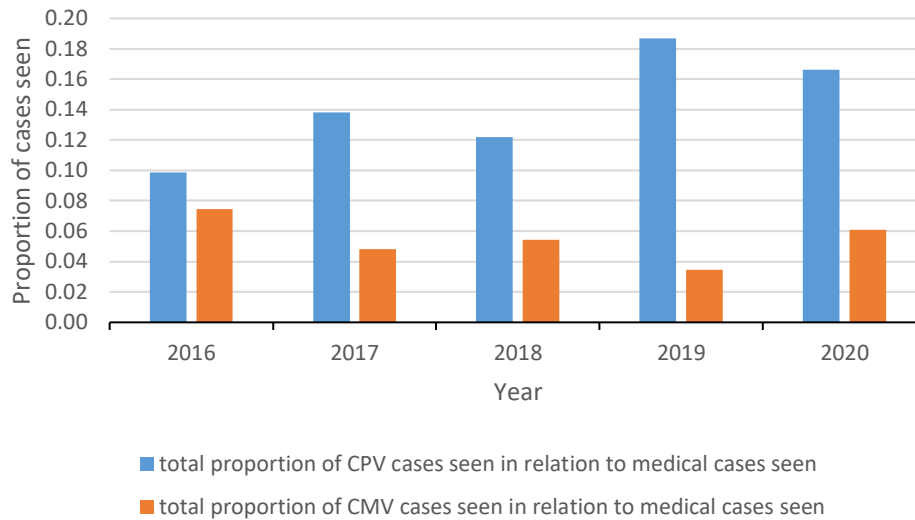


Figure 7.1. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Eastern Cape province every year between 2016 and 2020.

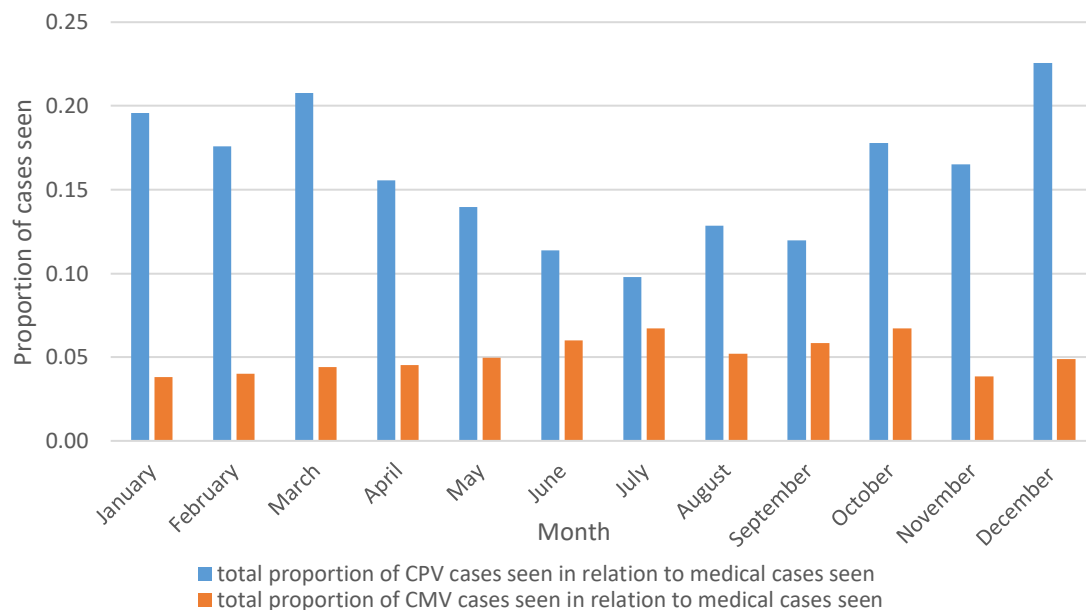


Figure 7.2. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Eastern Cape province monthly averages were collected using the data from 2016 to 2020.

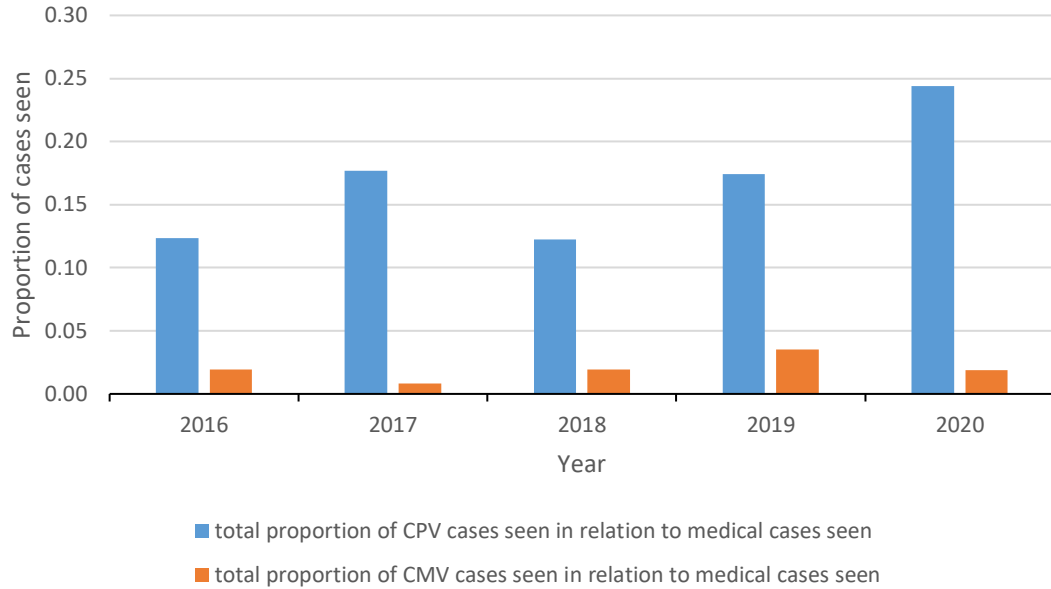


Figure 7.3. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Free State province every year between 2016 and 2020.

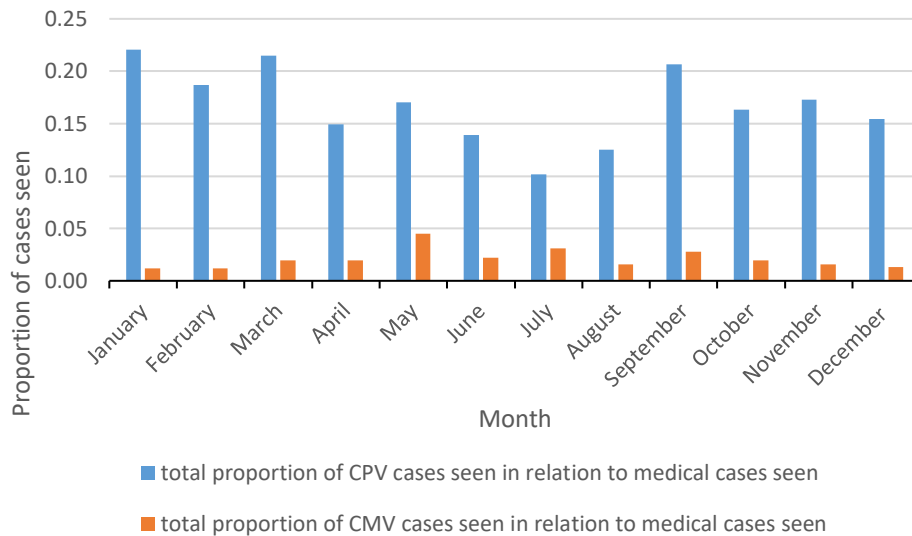


Figure 7.4. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Free State province monthly averages were collected using the data from 2016 to 2020.

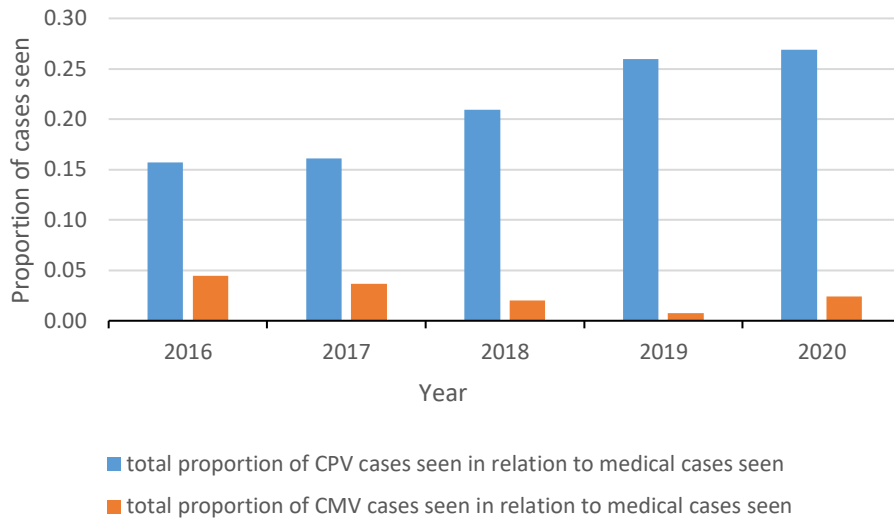


Figure 7.5. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in Gauteng province on a yearly between 2016 and 2020.

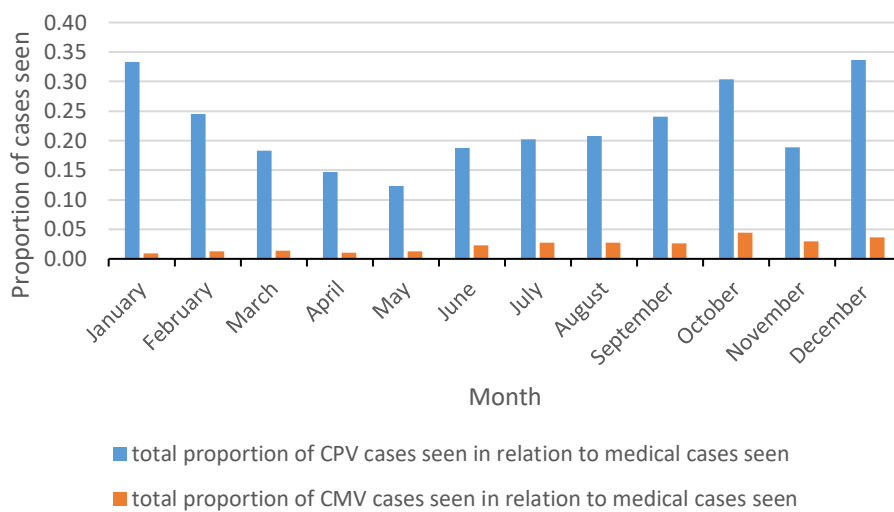


Figure 7.6. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Gauteng province monthly averages were collected using the data from 2016 to 2020.

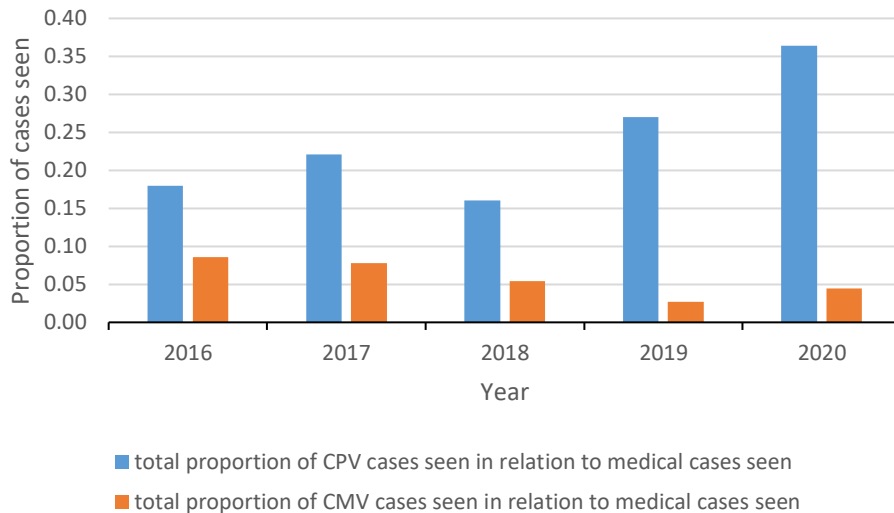


Figure 7.7. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in Limpopo province every year between 2016 and 2020.

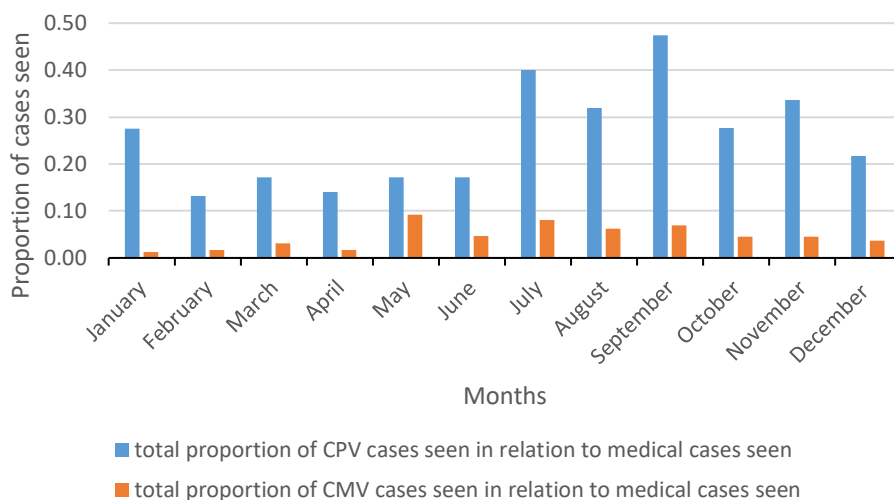


Figure 7.8. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Limpopo province monthly averages were collected using the data from 2016 to 2020.

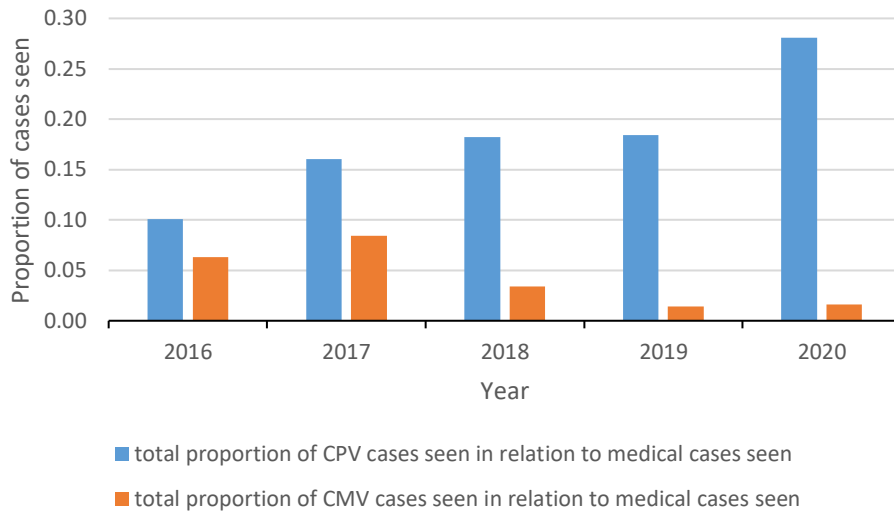


Figure 7.9. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in Mpumalanga province every year between 2016 and 2020.

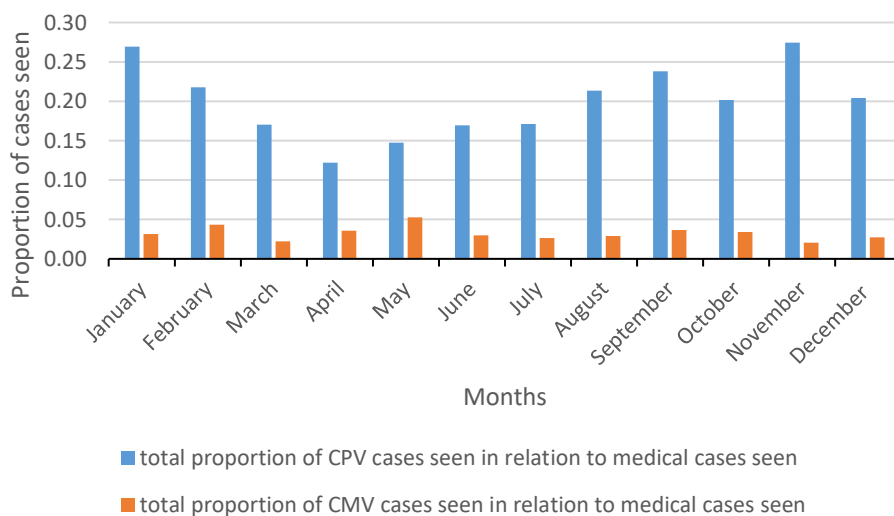


Figure 7.10. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Mpumalanga province monthly averages were collected using the data from 2016 to 2020.

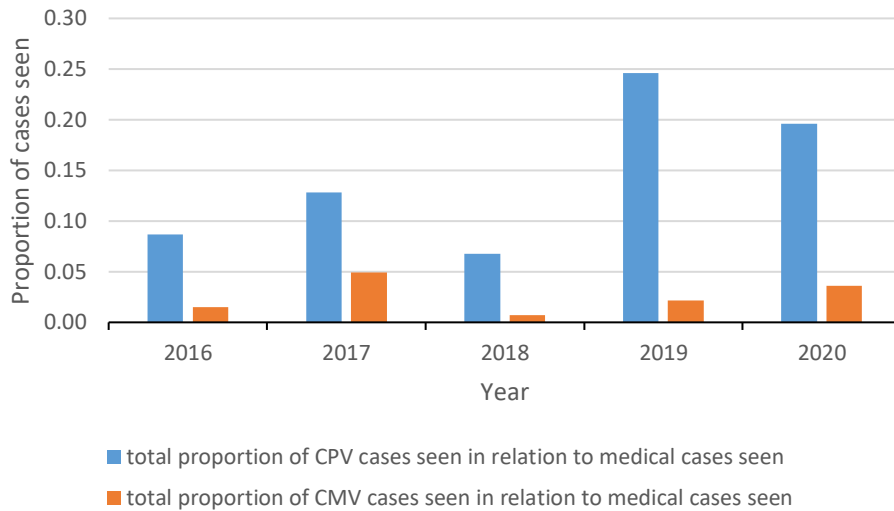


Figure 7.11. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Northwest province every year between 2016 and 2020.

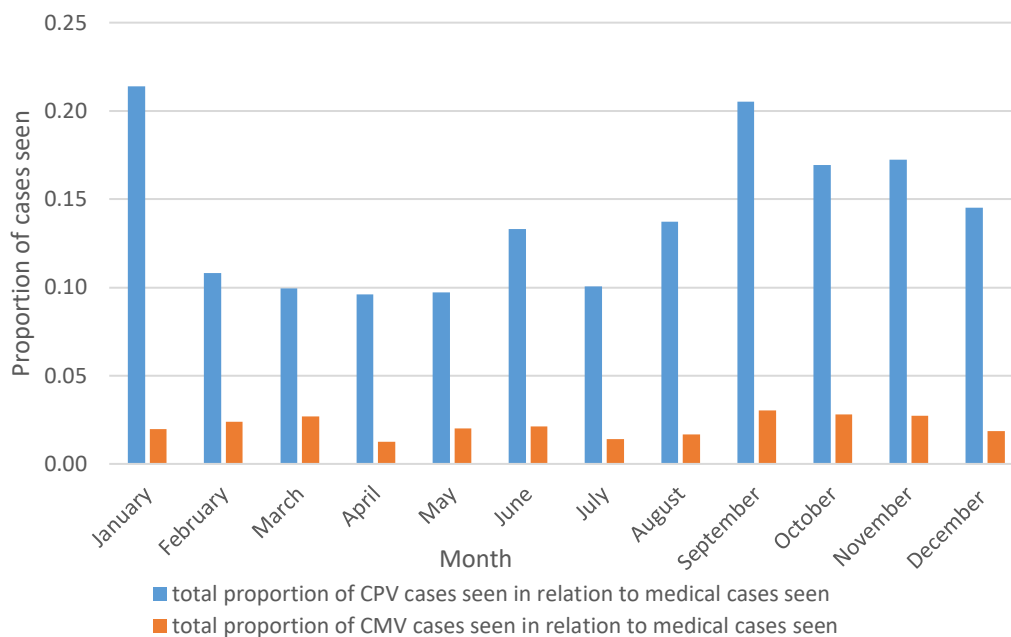


Figure 7.12. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Northwest province monthly averages were collected using the data from 2016 to 2020.

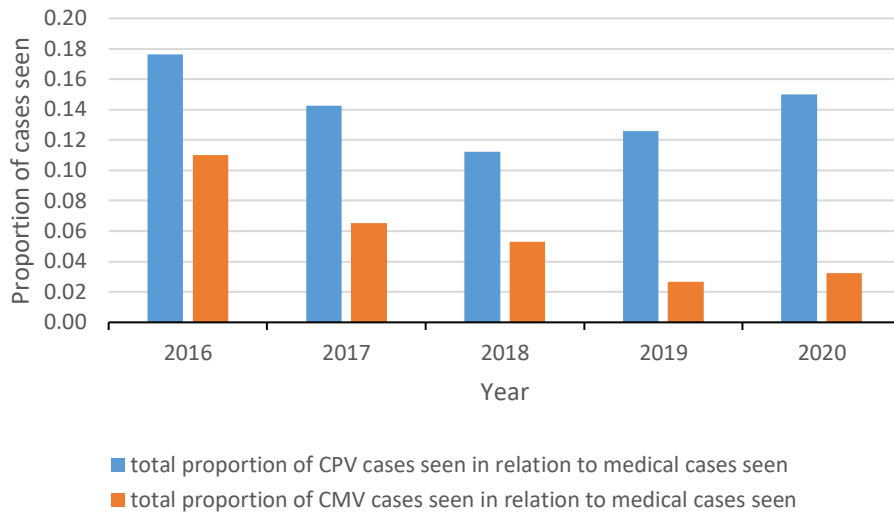


Figure 7.13. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Northern Cape province every year between 2016 and 2020.

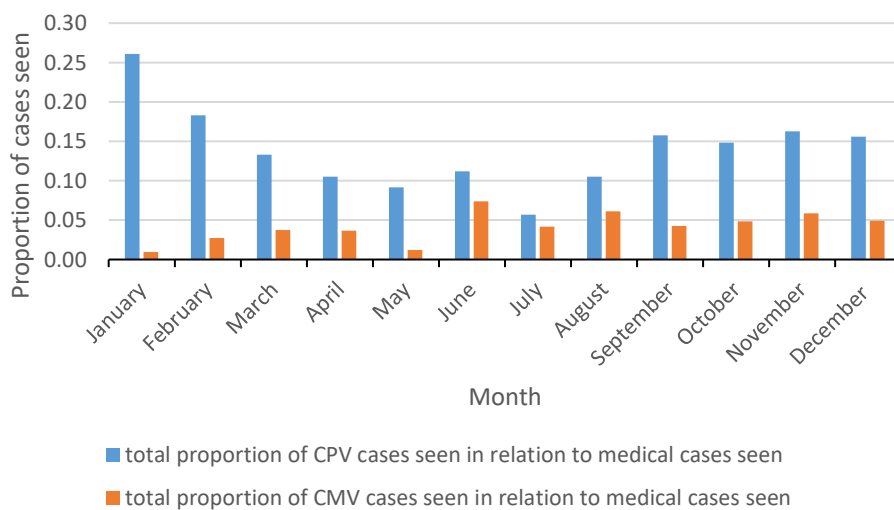


Figure 7.14. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Northern Cape province monthly averages were collected using the data from 2016 to 2020.

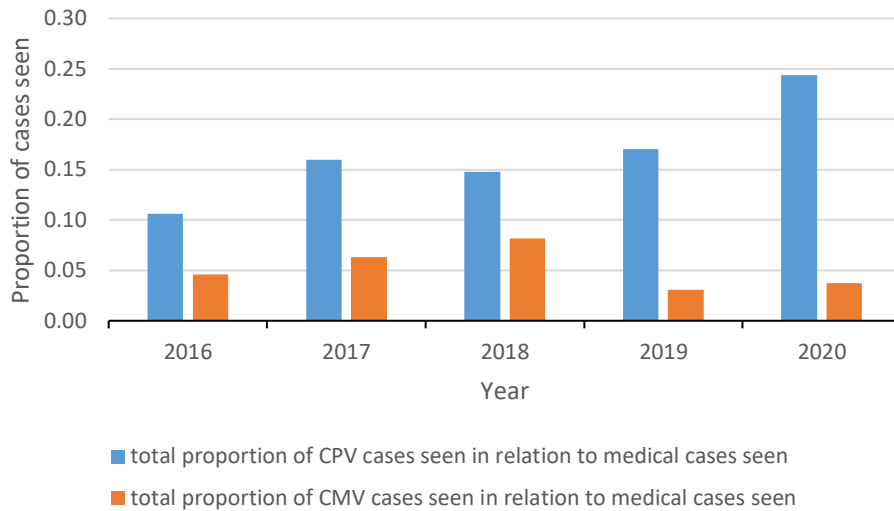


Figure 7.15. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Western Cape province every year between 2016 and 2020.

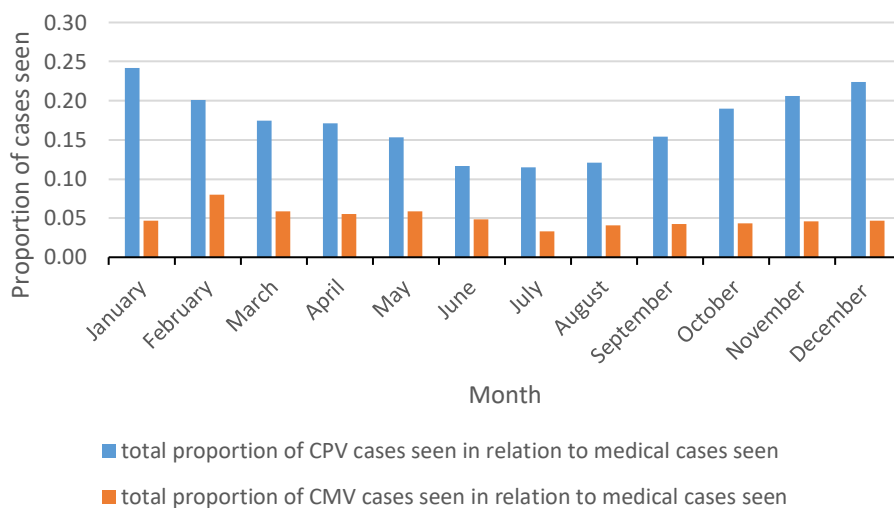


Figure 7.16. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Western Cape province monthly averages were collected using the data from 2016 to 2020.

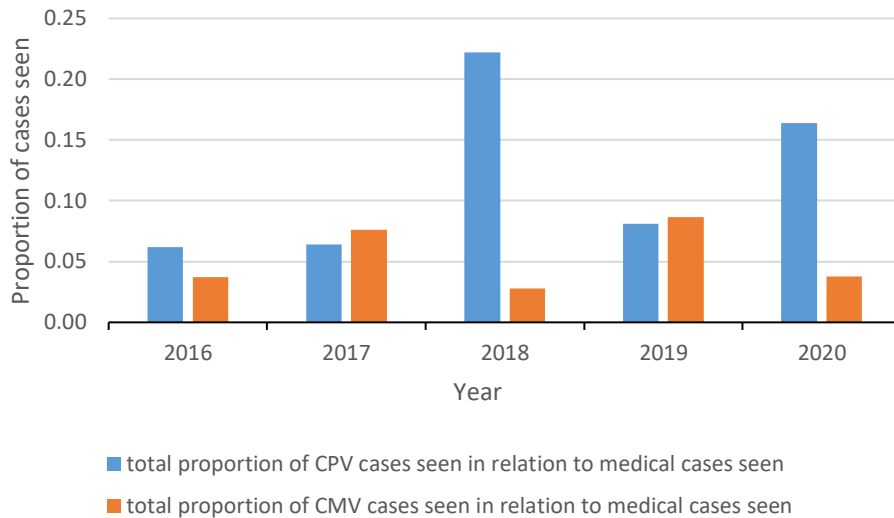


Figure 7.17. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Kwa-Zulu Natal province every year between 2016 and 2020.

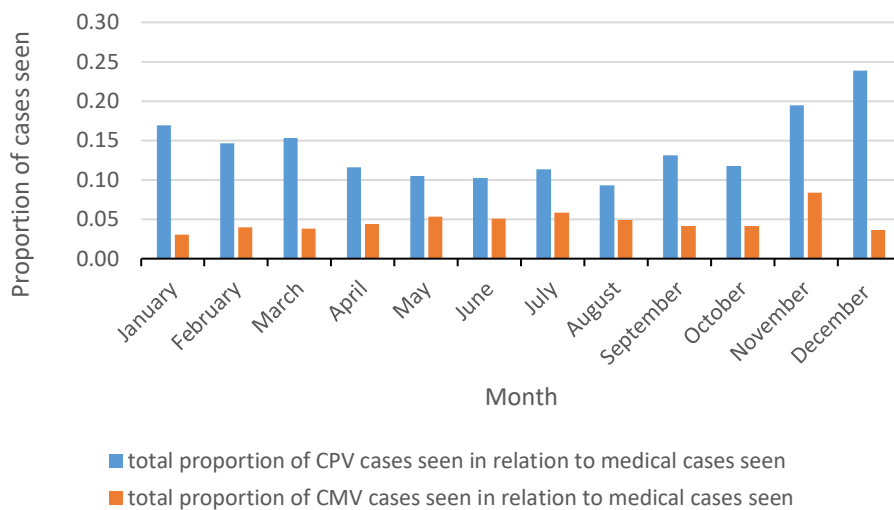


Figure 7.18. The average proportion of *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases seen in relation to the overall proportion of medical cases seen in the Kwa-Zulu Natal province monthly averages were collected using the data from 2016 to 2020.

Table 7.3. The total number of animals seen at Mdzananda clinic between July 2021 and December 2021, showing the number of animals seen at the clinic and the number of suspected *Carnivore protoparvovirus 1* (CPV) and *Canine morbillivirus* (CMV) cases. Records of the number of rapid tests used to test for *Carnivore protoparvovirus 1* and *Canine morbillivirus* respectively and how many of those gave positive results. There is also the number of *Carnivore protoparvovirus 1* and *Canine morbillivirus* cases seen in relation to the total animals seen at the clinic for each month during the 6-month study period.

Month	Total number of animals seen	Suspect CPV cases seen	Suspect CMV cases seen	Suspect CPV and CMV cases seen	Tests used for CPV	Tests used for CMV	CPV snap positive	CMV snap positive	Number of CPV cases seen in relation to total animals seen	Number of CMV cases seen in relation to total animals seen	
July	502	33	31	4	68	37	35	18	13	0.0737052	0.06972112
August	643	36	27	3	66	39	30	33	13	0.0606532	0.0466563
September	664	55	21	9	85	64	30	40	12	0.0963855	0.04518072
October	432	91	9	3	103	94	12	75	4	0.2175926	0.02777778
November	294	58	19	6	83	64	25	38	12	0.2176871	0.08503401
December	619	58	19	4	81	62	23	45	10	0.1001616	0.0371567
TOTAL	3154	331	126	29	486	360	155	249	64	0.1141	0.0491