Battling Canine Parvovirus with Antiviral Peptide Therapy

Nisha Naveen

Abstract

Canine Parvovirus (CPV), a gastrointestinal virus transmitted through feces that causes severe illness in infected individuals, remains incurable. Unlike its original counterpart, CPV-1, which only affected puppies, CPV-2 has crossed the species barrier numerous times and affects wild animals of all ages. CPV-2 has developed significantly, transforming from CPV-2A to CPV-2B and CPV-2C. Finding an effective treatment method is critical to limit the evolution of the virus and conserve endangered species. While there are preventative vaccines for domestic animals, efforts to address outbreaks in wild animals remain inadequate. This includes current therapies being tested, like monoclonal antibodies, which cannot serve as a long-term solution to combat the adaptive nature of the pathogen. Antiviral Peptide Therapy (APT) uses peptides, or short amino acid chains, designed to target specific viruses and prevent their replication. Here, we propose the novel application of APT for the global treatment of CPV-2 in endangered species and compare it with other therapies developed today. While APT has never been used on animals or to address CPV-2, this method may be more effective than current therapies being explored, and its potential to revive endangered species is very promising.

Keywords

Translational Medical Sciences; Disease Treatment and Therapies; Antiviral Peptide Therapy; Canine Parvovirus; Endangered Species

Introduction

The first CPV strain known as CPV-1, only affected young, domesticated dogs around three weeks old. In 1978, the first case of the second, more prominent strain (CPV-2) was discovered in Germany, causing a widespread epidemic among numerous species. It is still a concern worldwide and is considered endemic in many places, especially among susceptible populations like immuno-compromised species, or in areas where wildlife vaccination rates are low.[1] CPV-2 outbreaks among such populations can impact reproductive success, population growth, and genetic variability.[1] Efforts have been made to protect these endangered animals, including disease surveillance, reduction in contact with other species, vaccinations, and developments of antibody therapies. However, a more effective measure is necessary to fully combat the destructive effects of CPV-2 fully. In younger animals, CPV-2 has exhibited a mortality rate of up to 80%, resulting in substantial declines in endangered species populations.[2] The highly contagious nature of CPV-2 and its ability to spread rapidly among vulnerable animal populations makes it a pressing concern for conservation efforts.

This is why antiviral peptide therapy (APT) is a novel, yet promising proposal for the pressing concern of CPV-2. In recent years, APT has emerged as an effective approach in the battle against other viral infections such as influenza, herpes simplex virus, SARS-CoV-2, and human immunodeficiency virus. APT involves the use of short chains of amino acids, known as peptides, which are specifically designed to target and disrupt viral structures or processes. These peptides can interfere with viral entry into host cells, inhibit viral replication, or modulate the host's immune response to combat the infection.[8] Their ability to target specific viral components makes them particularly effective against a wide range of viruses, opening up the possibility that they can be effective against CPV. This modern therapy offers a promising avenue for developing antiviral treatments with potentially fewer side effects and lower resistance risk than traditional drugs.

Here, the focal point of our research centers on the development and application of APT as a potential treatment for CPV-2 in endangered species. With a specific emphasis on its comparative effectiveness against existing treatments, our research seeks to unravel the intricacies of APT application in conservation. We discuss how the unique challenges of treating endangered species can be addressed through the innovative application of APT, and how its efficacy measures up against conventional treatments currently available on the market. This investigation aims to shed light on the benefits of APT for endangered species and contribute insights into the broader landscape of CPV-2 as a whole.

Discussion

Genetic Diversity and Evolution of CPV

While CPV-2 is the most significant of the canine parvoviruses, a large variety of strains and modifications of this particular virus make its effects much more widespread. The virus is believed to have developed and evolved from the feline panleukopenia virus (FPV) in the late 1970s, with the first CPV-2 appearing in the early 1980s.[3] CPV-2 has also begun to affect wild animals and deteriorate their already critically endangered statuses due to contact between domestic dogs and wild animals, whether physically or by their feces. Detection of CPV-2 in wild animals has been reported in scientific literature since around the late 1900s. Alongside the well-known CPV-2 strain, researchers at Cornell University's Wildlife Health Lab reported related strains including RPV (Raccoon Parvovirus) and BFPV (Blue Fox Parvovirus), originating from the same Feline Panleukopenia lineage. These strains have specific genetic distinctions enabling their infectivity in an even greater diversity of wildlife populations, including raccoons, foxes, big cats, and other felids such as lions, tigers, leopards, cougars, and civets.[3] CPV-2 has spread among various families and species, including several endangered species as seen in Figure 1.

Figure 1. Phylogenetic Tree of CPV-2's Spread Among Species. A phylogenetic tree, not to scale, meant to showcase the spread of CPV-2 among species, with endangered species in blue.

Aside from having other branches from the common ancestor, CPV-2 has diversified into smaller variants like 2a, 2b, and 2c.[1] Each variant possesses distinct genetic traits influencing its impact and adaptation to different hosts. CPV-2a, the original variant, initiated the early outbreaks. Subsequently, CPV-2b emerged, exhibiting improved resilience against antibodies, eroding the effectiveness of existing vaccines. The most recent variant, CPV-2c, has unique genetic features and widespread prevalence in various regions and is still being studied today.[1] The development of different strains over time is described in Figure 2.

Figure 2. Canine Parvovirus Strain Evolution Timeline. This timeline from the 1960s to present is a visual representation of the information presented in research done by Gael Darren Maganga et al., 2023 regarding the different adaptations the virus has gone through before becoming CPV-2.

Specifically, the recent variant displays changes in certain amino acids and proteins.[4] The diverse genetic makeup of these variants dictates many characteristics such as their host cell recognition, environmental stability, and evasion of the host's immune response. This genetic diversity emphasizes the need for a comprehensive, adaptive vaccine and treatment development strategy. A treatment method targeted at one variant can open the door to addressing the entire spectrum of CPV variants and related viruses, a vital step in safeguarding many species.

CPV Pathology

The most common symptoms of a wild animal infected by CPV-2 include lethargy, lack of appetite, and a fever, along with results of the virus's effect on the gastrointestinal system such as soft, foul smelling, mucous, or bloody feces, vomiting, and diarrhea. These symptoms and the context of the virus's path within the host are much better understood. Once inside, CPV-2 targets the body's fastest-multiplying cells and uses the cell's machinery to replicate its own genetic material in a process called viral entry. The virus begins by destroying the bone marrow where these lymphocytes are found. Lymphocytes are a type of white blood cell found within the lymph nodes that help the body's immune system fight off foreign bacteria. By destroying these cells, the virus slows the production of lymphocytes and weakens the overall immune system. After targeting the immune system, the CPV-2 virus rides on the lymphocytes through the bloodstream. These lymphocytes are generally killed in the process, causing lymphopenia, an inadequate amount of lymphocytes. The resulting weakened immune system makes the host body vulnerable to the worst, most harmful attack, the gastrointestinal area. The virus enters the small intestine's epithelium lining and invades the reproduction site of these epithelial cells. Epithelial cells are crucial to replenishing the small intestinal lining to prevent fluid loss and bacterial infection from the gut. Severe diarrhea and nausea are initial symptoms until the gut bacteria invade the broken barrier of the small intestine. As a result, the gut bacteria later enter the bloodstream causing a potentially fatal widespread infection.[3] The virus then controls these rapidly dividing white blood cells and small intestinal cells to accomplish the most crucial step: viral entry. Once the virus enters these cells, it utilizes the cell's machinery to replicate its genetic material and produce more viral particles, an important step in its overall impact on the host animal.

Global Impact of CPV-2 on Wildlife

Global case studies conducted on a diverse range of animal species further demonstrate the profound impact of CPV-2. In Taiwan, from October 2017 to June 2019, researchers examined four Taiwanese pangolins that showed signs of gastrointestinal illness. They found that the sequences of nucleic acids from these four pangolins were identical to the CPV-2c strain found in China among domestic dogs. The team later used this information to identify the prevalence of CPV-2 virus across all Taiwanese pangolins; a species already deeply affected by habitat destruction and animal trafficking.[5] The impact of CPV-2 on endangered species doesn't stop there; it was discovered to have affected other species, including the Asian small-clawed otter. In Japan, researchers Kenichi Temukai and Shohei Minami, among others, conducted a post-mortem examination on one of two otters that had been imported from Indonesia. The two displayed distinctive gastrointestinal issues, and the medics found a necrotic small intestine along with swollen lymph nodes. An oral swab of the animal confirmed it was the CPV-2a strain, the first of its kind to affect small-clawed otters.[6] In South Africa, effects of CPV-2 were observed during a case study on an unvaccinated serval from South Africa. The serval presented with vomiting, anorexia, and diarrhea before dying around two weeks later. While this was the first known case of CPV-2 among the serval species, extensive research has been done to explore further the virus's impact on non-domestic species on the African continent.[7] Fundamentally, studies done on many species across different continents have furthered what is known about the effects of the virus which is visually represented in Figure 3.

Figure 3. Global Map of Canine Parvovirus Cases. This map of the entire planet pinpoints different locations of species that have been presented with CPV-2, to show the global impact of the virus.

The presence of the virus across several species globally further enforces the increased urgency of the matter and the need for a solution.

Applying Antiviral Peptide Therapy

Using antiviral peptides to target viruses is an important research development and opens the door to new possibilities. These peptide chains can be designed specific to a single virus and target the virus's specific proteins, a quality known as being virucidal. When developing antiviral peptides, scientists target specific stages in the viral process such as viral entry, viral synthesis, or assembly. Scientists view viral entry as the most crucial step in the CPV-2 process, making it the most optimal stage to target with APT. Specifically, as CPV-2 enters, it recognizes and binds to the receptors on the host cell using specific proteins, also known as a lock and key mechanism.[8] Within this lock and key mechanism, the virus's capsid protein binds specifically to sialic acid residues present on the host cell membrane.[1] This interaction triggers endocytosis, allowing the virus to enter the cell. With the current research and technology, the antiviral peptide can be essentially coded to interfere with this lock and key mechanism. Specifically, the antiviral can mimic the host cell's cellular receptor and compete with the binding site. If the CPV-2 virus binds to the antiviral rather than the cell, it has effectively prevented it from entering, replicating, or causing damage. For CPV-2, the primary cellular receptor is the canine transferrin receptor type 1 (TfR1), also known as the transferrin receptor protein 1 (TfR1).[8] The antiviral peptide would mimic the TfR1 to successfully guide the virus away from the cell, preventing it from replication and infecting the body. A promising and unique aspect of antiviral peptides is their interaction with bacterial membranes via electrostatic interactions, making it difficult for viruses to develop resistance to the peptide.[8] Especially considering the potential and history of CPV's various adaptive strains, this aspect proves to be a promising feature. Additionally, because CPV-2 directly attacks rapidly dividing areas such as the bone marrow and epithelial cells, cell-penetrating peptides (CPPs) can be added to the antiviral chain to facilitate the entry of the antiviral into these areas during administration.[8]

Advantages of APT Over Current Therapies

APT presents a targeted and potentially effective means to combat CPV-2 infections in these vulnerable populations. Today, the only significant therapy for CPV-2 being tested is an antibody-based solution known as Monoclonal Antibody Therapy. While antibodies are more efficient to develop, antivirals are much more specific and accurate in the long run. Unlike antibodies, which depend very heavily on cellular geometry and no longer work if the virus undergoes even the slightest of changes, antivirals target viral replication and reproduction. The reliability of antivirals is especially important considering the evolvability (or adaptability) of CPV-2 and its several strains. However, antivirals are much more complex to develop, hence why they can target complicated, distinct viruses. Peptides are biologically active molecules composed of a chain of amino acids, and they are also easily broken down by the enzyme, peptidase, making them less likely to accumulate in areas and cause toxic side effects like pituitary damage. These peptides are small, easy to synthesize, and can be made to be very specific, especially with the help of advanced research. The antiviral peptide can be designed to attack a specific stage, typically the most crucial in the viral replication process, consequently making it impossible for the virus to harm the animal further.[8] Overall, in light of the profound threat CPV-2 poses to endangered species and the pressing need for innovative solutions, the exploration of an APT targeting the CPV-2 cell entry stands as an incredible solution, offering hope in improving conservation efforts and mitigating the devastating impact of this pathogen and on the most vulnerable populations, with minimal negative side effects.

Potential Side Effects

Because peptides are biological molecules, they are known to work cohesively with other bodily functions. Therefore, antiviral peptides often have fewer side effects than traditional antiviral medications, with minimal impact on healthy cells, thereby reducing toxicity.[8] While research on the side effects of APT specifically for CPV does not exist, peptide-based therapeutics about other viruses such as HIV have caused mild symptoms, like headaches, rash, and mild nausea, as well as allergic reactions or irritation at the administration site.[9] Researchers attributed

the potential for side effects to variable factors such as gender, genetics, and age. When considering endangered species suffering from CPV, the type of species could also be a potential factor. Despite these, ongoing research on the topic aims to optimize these therapies, balancing their effectiveness against potential side effects for safe and widespread use against various viruses.

Open Questions

As APT progresses human medicine, it is important to consider the open questions it may raise. Wildlife conservation faces unique difficulties such as global biodiversity loss, the looming threat of a sixth mass extinction, and the accelerated spread of infectious diseases due to human encroachment into natural habitats. The relationship between the anthropogenic and wildlife world is ever-changing and requires advanced therapy to prevent the catastrophes that the pathogen promises. Unfortunately, the lack of resources, research, demand, and attention to the problems facing the animal world hinder treatments' crucial and necessary progression towards the deadly pathogen. It is for this reason that APT is a significant and groundbreaking proposal to a pressing, frequently overlooked issue. It is also important to acknowledge the potential errors and limitations in translating APT from lab to wild settings. A complex, intricate therapy such as APT may take years to come to promising stages in the lab. However, the proposal may raise questions about dosage optimization, delivery methods, and the broader ecological conflicts. Administering such a complex therapy to wild animals may be complicated and tedious. Though the likelihood of developing resistance to APT is very low, especially compared to other current treatment options, some risk still remains. Especially in such a fragile world, and as with all well-meaning interventions, the unknown outcomes of using APT in wildlife heed a cautious and calculated approach. Regardless of the possible complications, this novel proposal is just as promising as it is crucial to solving a pressing and urgent issue in the animal world. These open questions press for future exploration and may guide the promising potential of such a therapy in the right direction.

Conclusion

Overall, APT poses a promising avenue in combating the harmful pathogen, CPV. Currently, the most prevalent strain of the virus is CPV-2c, which has become increasingly common among domesticated animals as well as endangered species such as wolves, foxes, and more. Looking into the future, there are a variety of efforts being directed at conserving the species affected by CPV-2, including developing drugs, isolating infected species, improving supportive care, and enhancing vaccination strategies. The drastic impacts of CPV-2 make finding a long-lasting solution and combating its effects on endangered species before they reach extinction increasingly important. It is for this reason that antiviral peptide therapeutics appear to be an ideal solution. Currently, APT is being developed to target other impactful viruses, such as SARS-CoV-2, herpes simplex virus, and influenza virus, and the results are quite optimistic. The ability to adapt peptides specifically for the complexity and ever-changing nature of CPV-2 further makes it a reliable and stable possibility. By mimicking the lock and key mechanism of CPV, antiviral peptides pose a dependable method of protecting species this still-incurable pathogen's often fatal and long-lasting effects. As we navigate the potential difficulties of APT, the well-being of the planet's wildlife is illuminated by the quest for innovative treatments and preventative strategies, striving for a brighter and healthier future for struggling species.

Acknowledgements

I would like to thank Ambika Verma and Kate Marie Lagerstrom for providing mentorship and feedback on the manuscript.

References

(1) Nandi, S.; Kumar, M. Canine Parvovirus: Current Perspective. Indian J. Virol. 2010, 21 (1), 31–44. https://doi.org/10.1007/s13337-010-0007-y

(2) Horecka, K.; Porter, S.; Amirian, E. S.; Jefferson, E. A Decade of Treatment of Canine Parvovirus in an Animal Shelter: A Retrospective Study. Animals 2020, 10 (6), 939. https://www.mdpi.com/2076-2615/10/6/939

(3) Canine Parvovirus. Cornell University College of Veterinary Medicine. https://www.vet.cornell.edu/departments-centers-and-institutes/baker-institute/our-research/canine-parvovirus (accessed 2024-01-30).

(4) Moon, B.; Jang, J.; Kim, S.; Kim, Y.; Lee, H.; So, B.; Park, C.; Lee, K. Genetic Characterization of Canine Parvovirus Type 2c from Domestic Dogs in Korea. Transbound Emerg Dis 2020, 67 (4), 1645–1653. . https://onlinelibrary.wiley.com/doi/10.1111/tbed.13501.

(5) Chang, Y. C.; Lin, Z. Y.; Lin, Y. X.; Lin, K. H.; Chan, F. T.; Hsiao, S. T.; Liao, J. W.; Chiou, H. Y. Canine Parvovirus Infections in Taiwanese Pangolins (Manis Pentadactyla Pentadactyla). Vet Pathol 2021, 58 (4), 743–750. https://pubmed.ncbi.nlm.nih.gov/33866880/

(6) Tamukai, K.; Minami, S.; Kadekaru, S.; Mitsui, I.; Maeda, K.; Une, Y. New Canine Parvovirus 2a Infection in an Imported Asian Small-Clawed Otter (Aonyx Cinereus) in Japan. J Vet Med Sci 2021, 83 (3), 507–511. https://pubmed.ncbi.nlm.nih.gov/33473050/

(7) Oosthuizen, A.; Brettschneider, H.; Dalton, D. L.; Du Plessis, E. C.; Jansen, R.; Kotze, A.; Mitchell, E. P. Canine Parvovirus Detected from a Serval (Leptailurus Serval) in South Africa. J S Afr Vet Assoc 2019, 90 (0), e1–e6. https://pubmed.ncbi.nlm.nih.gov/31038325/

(8) Agarwal, G.; Gabrani, R. Antiviral Peptides: Identification and Validation. Int J Pept Res Ther 2021, 27, 149–168. https://doi.org/10.1007/s10989-020-10072-0

(9) HIV Antiviral Drugs - What are the common side effects?. Drugs.com. https://www.drugs.com/medical-answers/common-side-effects-hiv-antiviral-drugs-3565615/ (accessed 2024-01-30).

(10) Maganga, G. D.; Labouba, I.; Milendz Ikapi, S. Z.; Nkili-Meyong, A. A.; Ngonga Dikongo, A. M.; Boundenga, L.; Ngoubangoye, B.; Memvie, C.; Kumulungui, B. S. Molecular Characterization of Canine Parvovirus Variants CPV-2a and CPV-2c, Associated with Vaccinated Dogs at Libreville, Gabon. Viruses 2023, 15 (5), 1169. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10221576/

Author

Nisha Naveen is currently a sophomore at Wilcox High School in Santa Clara, California. She has an ever-growing passion for conservation medicine and aspires to pursue a career as a veterinarian in the future. Nisha also enjoys competitive gymnastics, Indian classical dance, traveling, and spending time with friends and family.