

ACTIVATED IMMUNE MECHANISMS OF THE VETERINARY RATTLESNAKE VACCINE
AND ITS DETERMINATION OF ADVERSE REACTIONS IN CANINE PATIENTS

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ABSTRACT

This paper examines the information basis behind such wellness decisions regarding administration of the *crotalus atrox toxoid* in canines based on recurring instances of adverse reactions, such as anaphylaxis, highly attributed to this vaccine. Presentation of previous research conducted on the rattlesnake vaccine as well as case management studies are outlined in the form of a literature review. Additional surveying of veterinary professionals and owners with patients who have experienced hypersensitivity reactions as a result of the vaccine may provide new insights as to potential underlying commonalities. Analysis of the correlation between the rattlesnake vaccine to secondary adverse reactions will follow discussion from a toxicological and immunological perspective. Priorities for research in order to decrease the risk associated with administration of the *crotalus atrox toxoid* are presented. These findings will hopefully lend more insight into the potential risk factors surrounding the *crotalus atrox toxoid* as well as pioneer developments in animal health and solutions for addressing a larger spectrum of pre-drug efficacy screening.

INTRODUCTION

Routine health and wellness form the core of preventative medicine within the veterinary field, most notably through vaccines. Compromised preventative care has presented a major challenge to maintaining health and safety standards, as limited knowledge and controversy over the *crotalus atrox toxoid*, known as the rattlesnake vaccine, has created an unstable foundation in the management of its administration. Controversy over the protective qualities of the vaccine, and its suspected association to anaphylactic reactions with its administration compared to other routine vaccinations, has called into question its true benefits. As pet owners seek to find more established cautionary measures against the risk of rattlesnake toxin, it is imperative to have an understanding of why these reactions are seemingly more prone to occurring, especially for the pet population based in the arid climate of the southwest.

As this doubt persists, there are no known determinants for these triggers, meaning that there is no explicit information behind why these adverse effects happen, or how to tell if a patient is at risk for reacting negatively. There are two main portions comprising this project. The first involves consolidating what published research exists within the scientific and veterinary community regarding the vaccine or any relation to rattlesnake envenomation in canines. Incorporating primary accounts from veterinarians who have encountered cases of rattlesnake envenomation in relation to vaccination status with the *crotalus atrox toxoid* and rates of observed secondary anaphylaxis forms the proposed second basis of information, which would be achieved by surveying from the surrounding Pima and Yuma County areas. These findings may build into further research which investigates down to a genomic level. Any possible indicators gathered from surveying, such as common signalment of patients like breed, age, or weight, may provide a genetic basis for activation of the immune system to the rattlesnake vaccine. Being able to differentiate a particular gene trigger has the potential for devising a test which screens patients so as to definitively know whether administration of the rattlesnake vaccine would cause a reaction. Overall, these considerations create a better understanding of the epigenetic relationship between genetics, a patient's history, and vaccines, with the potential to develop a more effective rattlesnake vaccine product. Given the parameters of this study, investigating the concepts surrounding the current understanding of the *crotalus atrox toxoid* through a comprehensive literature review, as well as primary accounts from local veterinarians, may lend more insight into potential risk factors of the vaccine, which can pioneer developments in animal health and devise solutions for addressing a larger spectrum of routine preventative care.

IDEAS IN CONTEXT

In theory, the *crotalus atrox toxoid* functions to stimulate antibody production against the rattlesnake toxin of the Western Diamondback rattlesnake, *Crotalus atrox*. Three primary

ingredients compose the vaccine: inactivated *Crotalus atrox* venom, aluminum hydroxide which serves as the adjuvant, and thimerosal that acts as the preservative. In the context of vaccines, adjuvants are meant to improve the immune response of the vaccine while preservatives are added to increase its vitality under the refrigerated conditions it is kept in. No compounding is needed for administration of this vaccine which comes in a 1 cc dose given subcutaneously.¹

Pathophysiology of *Crotalid* venom centers on myotoxic enzymes: phospholipase A2 and metalloproteinases. Phospholipase A2 is commonly known in the context of snake venom, as it works to block calcium-channels at the neuromuscular junction, effectively acting as a paralytic. Thrombocytopenia is often seen as platelet membranes are destroyed due to the phospholipase. On another front, metalloproteinases compromise the integrity of endothelial cells by cleaving the peptide bonds of the basement membrane. As a result, endothelial cells are morphologically affected by creating gaps which allows for fluid accumulation in the surrounding tissue.

Metalloproteinases have also been found to increase the release of TNF- α within the host, causing significant localized inflammation. Necrosis progresses through the mechanisms of venom proteins that reduce perfusion and promote secondary ischemia due to bleeding.²

Cases of rattlesnake bites, regardless of vaccination status, are required to be medically treated as an emergency, often including hospitalization. Unlike most other routine wellness vaccines, which work to stimulate antibodies to prevent the contraction of disease, the rattlesnake vaccine does not prove a formal preventative, and instead aids the canine in not developing as severe of a reaction to the venom upon intoxication, affording more time to reaching a hospital and receiving treatment.



Figure 1. Map showing the California distribution of the Western Diamondback rattlesnake (*Crotalus atrox* - black), North Pacific rattlesnake (*Crotalus oreganus oreganus* - light gray), and South Pacific rattlesnake (*Crotalus oreganus helleri* - dark gray) adapted from Cates, Charles C et al.⁶.

Antivenin acts as the standard treatment for rattlesnake envenomation, which is derived from the serum of horses who have been vaccinated with *crotalus atrox toxoid*. Essentially, the secondary antibodies afforded from the horse's serum in antivenin aid the body in negating the effects of the toxin. Vaccines function on the same principle, as they are meant to stimulate the production of antibodies against the specific rattlesnake toxin. A central question is then posed, considering what may be the disconnecting factor between successful antivenin treatment and the *crotalus atrox toxoid* if the basis for both relies on antibodies. Examining the research behind both within the context of rattlesnake envenomation provides measures in which to compare and contrast the two processes.

Information regarding the active ingredients of the *crotalus atrox toxoid* presents an opportunity to review functionality and the immunological mechanisms that may be the cause of triggered secondary anaphylaxis in these patients. If these reactions should occur, appropriate protocol includes administration of intravenous fluids, epinephrine and possible additional anti-anaphylactic therapies such as corticosteroids or atropine.³ Adverse reactions in association to the veterinary rattlesnake vaccine should be questioned if they appear as a development of cross-reactivity, in which the antigen of the vaccine is similar to the structure of a protein the patient is actually allergic to, causing an immune-mediated response.³ Furthermore, what information is available to the general public regarding the rattlesnake vaccine since its release in 2004 is an important facet to consider.⁴ Owners often make decisions based on what recommendations are readily available, allowing popular and media sources to be an important tool to cross-examine within the context of the study. Understanding what information owners would most like to know when considering this vaccine allows for more educated discussion with their veterinarian.

LITERATURE REVIEW

Much of the validity concerns surrounding the *crotalus atrox toxoid* has to do with a lack of definitive data and research proving its safety as well as its beneficial effects. Red Rock Biologics, the manufacturer of the rattlesnake vaccine, outlines the purpose and theorized immune interactions that encourage a first-response effect to the rattlesnake toxin in order to improve treatment outcomes. Bites from pit vipers, including the rattlesnake, entail varying degrees of injury.⁴ Swelling and bruising may characterize local tissue injury at the site of injection, but the venom itself is known for its anti-coagulative properties and progressive systemic effects such as hemorrhage, hypotension, altered mental state, nerve damage as well as tissue necrosis.⁵

Red Rock Biologics devised the vaccine to supplement the host with antibodies that would bind to the proteins present within the toxin and ultimately neutralize the venom. The presentation of their research claims that vaccination with the *crotalus atrox toxoid* works to reverse the venom immediately from injection, where mild reactions like swelling are diminished upon presentation to a clinic compared to unvaccinated dogs who may continue to have progressive swelling 1-2 days after the event. While claims addressing that only 71 out of 24,975 cases of side effects were noted, and no anaphylactic reactions upon commercial usage in its first year were reported, none of the case studies were discussed in the context of the document. Vaccine efficacy was demonstrated purely through *in vitro* cell cultures which confirmed the ability of the post-vaccinal serum to neutralize the *Crotalus atrox* venom.⁴

In a later developed study analyzing higher critical cases of rattlesnake envenomation from 2006-2012, a total of 82 canine patients were examined regarding the protective effects of the *crotalus atrox toxoid*. Record of prior vaccination was only noted in 17% of subjects; however, no statistically significant difference was associated between vaccinated and unvaccinated patients upon clinical presentation, length of hospitalization, or amount of antivenin needed. Based on the parameters of the investigation, higher instances of morbidity correlated with lower body weight were the only signalment element found relevant. Though they were also determined to not be statistically significant, bite location in the cephalic region as well as unvaccinated dogs were 2.7 times more likely to be scored higher in morbidity.⁵

A study by the American Journal of Veterinary Research tested vaccination with the *crotalus atrox toxoid* (CAT) against venoms of *Crotalus atrox* and two other snakes (*Crotalus oreganus oreganus* and *Crotalus oreganus helleri*) using comparative mice models. Mice were inoculated with only adjuvant or CAT at 0 and 4 weeks of age, before being challenge-exposed to one of the three venoms four weeks later. Vaccination held up in protection against *Crotalus atrox* venom, resulting in the survival of 6/15 mice, but afforded no cross protection to other snake venoms.

Survival rates and times after exposure were improved with CAT vaccination, but ultimately no significant difference in the survival curves for those exposed specifically to *Crotalus atrox* venom. Vaccination with adjuvant-only also was not effective against envenomation in any of the three cohort groups involved within the experiment.⁶

Notably enough, a majority of the research that has been conducted regarding the rattlesnake vaccine has to do with what clinical protection is afforded by it. Clear distinctions between what patients are and are not vaccinated have yet to decipher claims that the *crotalus atrox toxoid* carries a higher risk of adverse reactions than other routine vaccinations. Additional speculation over causation of anaphylaxis given vaccination status in canine patients has led to further study into a simple phenomena: does administration of the vaccine itself cause a reaction in certain patients, or are adverse reactions triggered by the vaccine after natural envenomation?

Two cases of secondary anaphylaxis after a rattlesnake bite in CAT vaccinated dogs were discussed at length by one study with their symptoms and recovery processes. Due to atypical clinical presentations based on anecdotal experiences of rattlesnake envenomation in the Sonoran Desert, anaphylactic shock secondary to components of the venom was proposed as an alternative mechanism responsible for these symptoms. Researchers in that study proposed that vaccination enabled a developed hypersensitivity to envenomation required for the progression of anaphylaxis.⁷

Seeing as treatment for *Crotalus atrox* venom is derived from the secondary antibodies afforded from natural envenomation in horses, insight into clinical protective mechanisms in horses as patients provides an interesting comparison. Horses previously bitten had significantly higher antibody titers than vaccinated horses. Serum and plasma samples were collected at one week and one month before being assayed using ELISA (enzyme linked immunosorbent assay) in order to evaluate immunological responses. TNF-a concentrations were elevated, thus thought to be the cause of observed cardiac damage. Cardiac troponin I and electrocardiography were

used to evaluate sustained cardiac effects of venom.⁸ A comparative study analyzed antibody titers between natural envenomation horses (bitten) and those that were vaccinated. Antibody titers were evaluated using ELISA, and horses that were bitten had a higher antibody titer than those that were vaccinated. There was no demonstrated difference in effects between pregnant and nonpregnant horses, which may indicate that pregnancy does not act as a relevant signalment component when considering potential adverse effects of vaccination.⁹

ANTIVENOM (ANTIVENIN) TREATMENT

Investigating research surrounding the sole treatment for rattlesnake envenomation was deemed relevant given the similar properties of vaccination and antivenin. Both function on a principle of antibodies, where one is marketed as a preventative/suppressor and the other a neutralizing agent. However, while antivenin continues to be used in clinical context without fault, the rattlesnake vaccine remains dependent on region as well as preference.

Some adverse effects have been noted in previous cases with regard to the administration of antivenom. A comparison study between three commercially available antivenin treatment products was conducted and graded based on observed efficacy. No benefit was found of one particular antivenom, though treatment with the whole immunoglobulin product (IgG) only required one unit. Lower probability of survival was associated with older age, lower body weight, being bitten in the thoracic area, and possible antivenom infusion reaction.¹⁰ Other articles argue that further study of the pathophysiology associated with rattlesnake envenomation may provide better insight into developing appropriate treatment protocols. Instances of 34 canine and feline patients with evidence of neurotoxicity secondary to antivenin treatment was 5.4%. Evaluation of these cases revealed that there was no statistical significance in the type of antivenin or vials administered on neurologic status or survival rate.¹¹

GENERAL PUBLIC

What is important to consider is that evidence remains circumstantial in light of what the owner believes is best for their pet. Much of the efficacy research regarding the vaccine is still ongoing within the veterinary community, but what information is readily available to the public remains paramount to how they understand the rattlesnake vaccine. Most likely, what is guiding their decision-making in how to best protect their pet comes from the media or their veterinary care provider.

Several veterinary hospital websites have an information page dedicated to the *Crotalus atrox* toxoid, each varying in their span of information. Most outline the general purpose of the vaccine, the risk associated with rattlesnake bites, as well as the consideration that immunization is not a 100% preventative. Two distinct articles relay that adverse reactions occur in less than 1% of canines vaccinated.¹² Opinion between sites varies as many outline that there is a significant lack in published evidence and definitive support for the vaccine, while others communicate that the benefit outweighs the risk of an adverse reaction based on the pet's lifestyle and geographical risk.

POTENTIAL SURVEYING

What current research exists mostly involves examining envenomation responses and measured clinical protection afforded by the vaccine. While these findings seem to reach similar consensus, the information afforded to the public through veterinary information pages carry much more subjective and contradictory opinion. An aim of this research was to investigate the reasoning behind triggered anaphylaxis due to administration of the rattlesnake vaccine, and the immune-mediated mechanisms behind clinical adverse reactions. The premise of this analysis seeks to garner insight as to the risk factors behind vaccine-triggered anaphylaxis, while calling into question the current understanding of the rattlesnake vaccine that exists among veterinary professionals as well as owners. These surveys were conducted through voluntary participation.

VETERINARIAN

An important component of this research relies in understanding what current premise the veterinary community has regarding the *crotalus atrox toxoid*. These considerations relate to developing a veterinarian's perspective on vaccinations in general and its relationship to anaphylactic reactions. Each question is devised as a way to understand each veterinarian's experience with adverse reactions in relation to routine vaccinations, as well as specifically related to the rattlesnake vaccine. These responses give a general sense as to the role the rattlesnake vaccine is supposed to function in theory versus actuality.

Questioning how frequently they administered or if they've ever seen a negative reaction to the vaccine distinguishes the level of familiarity with the *crotalus atrox toxoid* at work. Any reported adverse reactions linked to other standard vaccinations allows for cross-analysis of the ingredients within that vaccine, as well as the rattlesnake one, to observe if there are commonalities that may be the issue behind these occurrences. If there proves to be a higher incidence rate with the rattlesnake vaccine, it demonstrates the point of compromised wellness and something particular about this vaccine is the issue. Patient history may indicate what signalment is important in cases of vaccination. Considerations into discussing with owners elucidates what information is available to the public.

All responses remain open-ended to allow for maximum input. The questions are as follows:

<i>VETERINARIAN INTERVIEW</i>
Question
What is the purpose of the rattlesnake vaccine?
In general, what is vaccine triggered anaphylaxis in canines?
Have you ever had a patient that had an adverse reaction to a vaccine? If so, what vaccine was it?
How frequently have you administered the rattlesnake vaccine per year?
Have you ever encountered a patient that developed an anaphylactic reaction to the rattlesnake vaccine? If so, what symptoms were prominent?
In your experience, have you seen that canine patients have a higher chance of reacting negatively to the rattlesnake vaccine than to another vaccine?
What aspects of patient history are important to note in order to avoid a potential adverse reaction to a vaccine?
Would you recommend canine patients to receive this vaccine? Why or why not?
What are your considerations when discussing the merits of this vaccine with clients?
Why do you believe there hasn't been more research into why particular patients develop adverse reactions to the rattlesnake vaccine?

Figure 2. Veterinarian Interview regarding the *crotalus atrox toxoid* vaccine.

Vaccine triggered anaphylaxis was reported as a type 1 hypersensitivity reaction. Defined, type 1 hypersensitivity is an exaggerated immunoglobulin E-mediated (IgE) response of the immune system. The high influx of inflammatory-mediators such as histamine that can be triggered from vaccines have the ability to cause “severe vasodilatory shock”, resulting in anaphylaxis.

One veterinarian's account reported having witnessed an adverse reaction to the rattlesnake vaccine, leptospirosis vaccine, and the rabies vaccine. However, the anaphylaxis was triggered only after being bitten by the rattlesnake, not upon injection of the vaccine itself. It is their opinion that patient's do appear to have a higher chance of reacting negatively to the *crotalus atrox toxoid* rather than any other vaccine:

“There are many cases of the vaccine acting as the initial sensitization which then causes anaphylaxis when the patient is bit [by the rattlesnake]. There is also no strong evidence that there are any long lasting antibodies in these patients with any benefit.”

- Dr. Kaelyn Petras

When regarding patient history, it is important to note any previous experience of an adverse reaction to any vaccine. Most veterinarian's in my personal history have expressed that they would not recommend patients receive this vaccine. Each testimony claimed that there was no proven effect in minimizing the severity of symptoms that accompany a rattlesnake bite, or higher incidence of survival for vaccinated patients. The anaphylactic risk of the *crotalus atrox toxoid* may actually pose a threat to the life of the animal, as well as accumulate cost for the owner in the long-run.

In asking about the proposed reasoning behind the lack of research into this phenomena, one account references incentive. Rattlesnake envenomation cases are treated the same regardless of the patient's vaccination status, undermining the relevancy of the vaccine. Dr. Petras proposes that geography may be an influential factor as well: risk of a rattlesnake bite only applies to particular regions, because highly venomous snakes only exist in a small proportion of the United States. Rather than an adverse reaction seen upon administration, Petras considers that secondary anaphylaxis is seemingly triggered post-bite in vaccinated patients. As such, doctors

may potentially mistake the symptoms as shock from natural envenomation rather than an anaphylactic reaction (Dr. Kaelyn Petras, personal communication).

A conclusionary finding of this process revealed unknown differences in offering the rattlesnake vaccine as a service. Through these investigations, I was able to discern that the rattlesnake vaccine is not a commonly offered service, even in regions at high risk for rattlesnake bites. Many of the hospitals contacted within the Tucson area did not carry the *crotalus atrox toxoid*, which differed from hospitals contacted in Yuma County.

RATTLESNAKE ENVENOMATION

Further reflection on the parameters of the original survey developed an idea for a more simplistic approach. Questions detailing an investigation on the correlation of the *crotalus atrox toxoid* in cases of rattlesnake envenomation and observed secondary anaphylaxis would provide a basis which revealed incidences of shock stimulated by a rattlesnake bite and any connection to vaccination status. The idea of focusing the response process opened up the survey for any veterinarian to answer- not limited to those who have and/or frequently administer the vaccine. These questions detail the basis of the relationship between rattlesnake bites, secondary anaphylaxis, and whether the patient was previously vaccinated with the *crotalus atrox toxoid*. Detailing the order of events is important to differentiate one of the ideas introduced in previous research: can secondary anaphylaxis be caused as a result of a vaccinated canine being bitten. In this scenario, cases can be evaluated in order to ascertain molecular interactions between rattlesnake venom as a negative catalyst for whatever protective mechanisms are afforded by the vaccine.

Rattlesnake Envenomation Investigation Survey

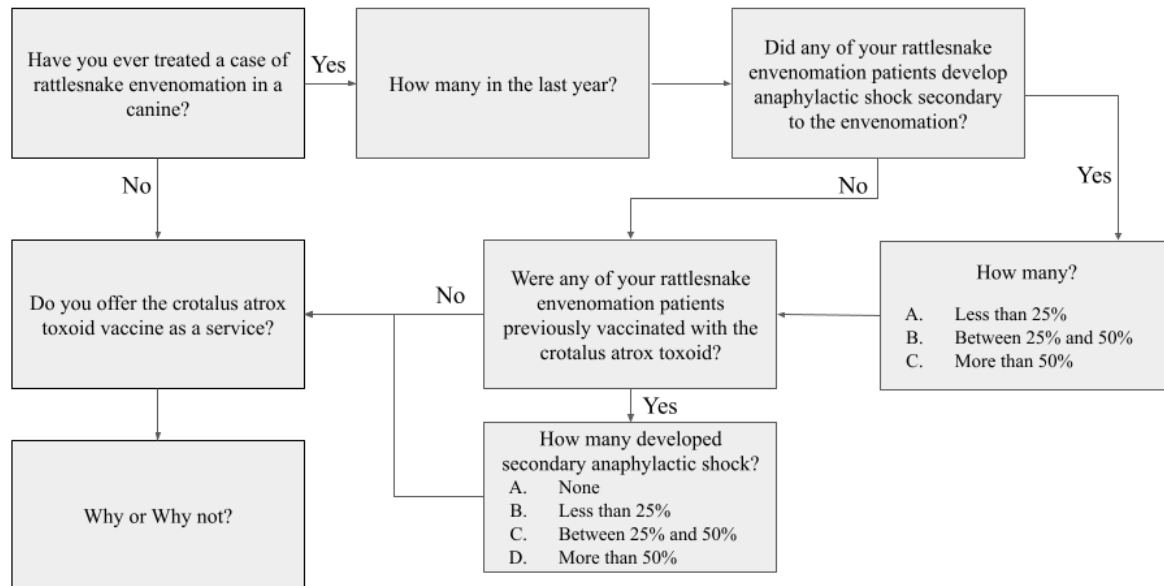


Figure 3. Response-based questionnaire relating to incidences of rattlesnake envenomation and secondary anaphylaxis.

This survey was not able to be conducted based on approval processes given the time constraints of the research.

OWNER

The purpose of this survey was to further study patients who had a confirmed negative reaction to the rattlesnake vaccine, *crotalus atrox toxoid*. Owner testimony would elucidate on signalment and history of their animals which allowed for cross-examination between patients who had an adverse reaction. Based on responses, these surveys could be analyzed for commonalities in terms of age, weight, or even pre-existing conditions which may be a determinant for an adverse reaction to the *crotalus atrox toxoid*. Furthermore, owners would be able to provide greater testimony about the specificities of these reactions, such as the onset, severity, and span of observable symptoms. The extensive list of symptoms to select from was

devised based on reported clinical signs of anaphylactic and vaccine-based reactions from various sources. Ultimately, any detectable trend would then be able to be examined for validity by devising an experiment which compared control subjects with similar signalment in order to compare adverse reactions to those that did not. These specific questions include:

OWNER INTERVIEW	
Question	Response
What breed is your pet?	
Is your pet male or female?	<ul style="list-style-type: none"> • Female • Male
How old is your pet?	
What is your pet's weight?	
Please select any and all symptoms your pet experienced to the rattlesnake vaccine:	<ul style="list-style-type: none"> • Fever • Weakness/Lethargy • Decrease appetite • Facial swelling • Swelling of the extremities • Swelling at vaccination site • Redness at vaccination site • Rash/Hives • Itching • Difficulty breathing • Coughing • Vomiting • Collapse • Pale gums • Blood from the anus • Diarrhea • Other (Fill-in Response)
Which symptom did you first notice?	
How long was it before you observed a reaction? (immediately, minutes, days, weeks, etc.)	
Does your pet have any underlying health conditions? If yes, please list all known pre-existing conditions:	
Does your pet have any known allergies? If yes, please list all known allergies:	
Has your pet ever had an allergic reaction to a vaccine in the past? If so, which one(s)?	

Figure 4. Owner Interview of Adverse Reaction Patients.

No owner testimonies were able to be conducted for this study.

DISCUSSION

The culmination of this data illustrates limited consensus involving the *crotalus atrox toxoid* in cases of rattlesnake envenomation, both in terms of clinical protection afforded as well as interactions with host immune mechanisms that may denote secondary adverse effects. As a result, little can be determined proving efficacy and thorough testing of the vaccine. A cascading effect is then experienced within the provision of health care. Veterinarians have not been met with definitive proof regarding the rattlesnake vaccine, nor are clear on the risk and rates of adverse reaction, and thus are unable to provide clear recommendations. Owners have to base decisions on what is best for their animal, mostly pertaining to avoiding a risk potentially associated with administration of the *crotalus atrox toxoid*, and must find other preventative measures for rattlesnake safety.

Most studies were able to differentiate some level of higher morbidity associated with unvaccinated canines, though no data in any study was concluded as statistically significant. Vaccination with the *crotalus atrox toxoid* affords the most clinical protection against *Crotalus atrox* venom rather than all pit vipers associated with the southwest region. No specific signalment was determined to have a connection to instances of adverse reactions such as anaphylaxis or higher morbidity in canine patients; however, lower weights were correlated with higher severity scores upon clinical presentation. Despite these considerations, there is a clear lack in data evaluating the particular immune mechanisms associated with vaccination status and hypersensitivity reactions. A question still remains of why the vaccine cannot generate enough antibodies to neutralize the venom on its own. Devising whether these adverse reactions were local, systemic, or allergic was unclear given the scope of data.

Many studies have been conducted in an attempt to characterize the properties of specific metalloproteinases as hemorrhagic toxins. Further analysis of the *Crotalus atrox* toxin proteins

is required to gain a more conclusive understanding of its interactions with the host as well as how that compares to vaccination with the *crotalus atrox toxoid*. In addition, analyzing the rattlesnake vaccine in terms of its specific components has the potential for identifying other therapeutic targets as well as ingredients that have a causative effect to the adverse reactions that have been demonstrated. Other vaccines which contain ingredients like the aluminum hydroxide adjuvant can be investigated for reports of induced anaphylaxis as a comparison. These insights have implications in creating a study to evaluate reactivity on a subcellular level such as IgE interactions, targeted antigen receptors, and TNF- α concentrations with envenomation versus the vaccine.

Due to the lack of survey data, no formulations were able to be made connecting anecdotal case reports to the information present in past research studies. An important distinction in discussing morbidity associated with a venom is that these cases are manifestations of an intoxication, not a pathogenic molecule. Toxicology outlines a principle known as dose-dependency: the route and amount of toxin delivered will affect the rate the toxin reaches systemic circulation and severity of clinical symptoms. For example, a bite on the extremities, such as the leg, will take longer to reach systemic circulation than a bite to the body cavity such as the thorax. This may account for the variations in case study reports as well as lack of reliability in efficacy demonstrations. Most of the studies involved in the literature review called for further surveying utilizing subject groups of a larger sample size (ex: n=400) in order to be statistically significant.

As there was no surveying, no commonalities were observed between vaccination with the *crotalus atrox toxoid* and specific signalment or patient history. Further study would involve conducting these surveys, with the addition of including more extensive interviews that incorporate veterinarian testimony and patient profiles beyond the state of Arizona as well as a larger sample size. These canines should be evaluated for numerous aspects that expound on

classic signalment to incorporate elements such as history of litters, blood type, etc. that may be in connection to observed hypersensitivity reactions. The results of the survey would indicate what patient group would need to be analyzed to evaluate for the potential of a specific gene trigger. Essentially, this would shape knowledge on any commonalities underlying activated immune-mediated mechanisms against the rattlesnake vaccine for future investigative research.

By revealing any specific genetic or molecular trigger which correlates with a predisposition for adverse reactions specific to the rattlesnake vaccine, therein lies the implication in developing pre-drug efficacy screening. Essentially, if negative immune responses can be tied to a particular factor, we can test for that factor in order to determine if that patient would have a negative reaction to the vaccine. It provides a definitive answer as to whether or not an individual patient will experience negative side effects as a consequence of vaccination with the *crotalus atrox toxoid*.

Technology predicting drug efficacy outcomes prior to administration can be applied in a broader sense to veterinary medicine as a whole. Theoretically, this would entail analyzing how well a patient would respond to a treatment plan before it is conducted. Many cases seen within a veterinary practice hinge on the successful interaction between a drug and the host's response, such as the prescription of steroids to control edema, autoimmune disease, and so on. If this interaction can be determined to be fruitless before any treatment is initiated through a pre-efficacy screening test, many resources are spared for both hospital and owner. Without the cycle of trying a new treatment when the previous proves unsuccessful, treatment time, such as length of hospital stay, can be shortened (which may be valuable for quality of life cases), finances can be spared for the owner by paying for only the treatment that is needed, and patients are less worn out, reducing stress, fear, and anxiety. On the other hand, hospitals are able to address patients quicker and move on to new cases, saving valuable time and/or space;

resources such as pharmaceuticals are allocated appropriately, saving money, and clients are more satisfied with the care being provided.

Evaluating the mechanisms of the current rattlesnake vaccine further, as well as identifying any deficiencies between venom neutralizing antibodies in antivenin as a treatment and the *crotalus atrox toxoid* as a preventative, may lead to the development of an improved vaccine affording more clinical protection.

CONCLUSION

There are many concepts to consider with such a decision as the priority level of routine vaccinations in regards to welfare, and as such, we must consider the alternative perspectives, concluding with a position that seems most justifiable under the circumstances of case experience. While no confirmation exists that observed adverse reactions are directly caused by the veterinary rattlesnake vaccine, finding any correlation to an inappropriate immune response remains paramount in evaluating risk. In recognition of this issue, several steps can be taken to mitigate the lack of consensus and data surrounding the *crotalus atrox toxoid*. Following up with the proposed survey accounts will provide needed testimony through case experience. Conducting these surveys in multiple counties of Arizona, as well as expanding to other states and regions where the rattlesnake vaccine is prevalent, may supply enough data for cross-examination of underlying commonalities as well as key differences in reports. Apart from further research, creating protocols for managing adverse reactions as well as more descriptive characterization of anaphylactic allergy presentation are encouraging prospects. Once veterinary professionals are familiar with the clinical signs of a hypersensitivity reaction, this will serve as a resource for differentiating these symptoms from that of the rattlesnake toxin. Not only do these considerations provide a better way for understanding the *crotalus atrox toxoid* interactions with the host on a molecular level, but owners and veterinarians who are unfamiliar with the consequences of immune-mediated reactions due to the rattlesnake vaccine are better equipped

to handle these cases upon presentation. With the comprehensive knowledge that may be generated from conducting this research, both the veterinary community and the general public can have more informed conversations regarding the benefits and detriments of the *crotalus atrox toxoid* as it applies to maintaining the health of family pets and mitigating the risk of rattlesnake envenomation.

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