Committed TO INNOVATION
PROVEN BY Science

SPRAY-VAC® VAC-PAC® VAC-PAC PLUS® OPTI-VAC®

INSTANTLY GUARDS AGAINST: OXIDIZERS • pH IMBALANCE • LOW TONICITY

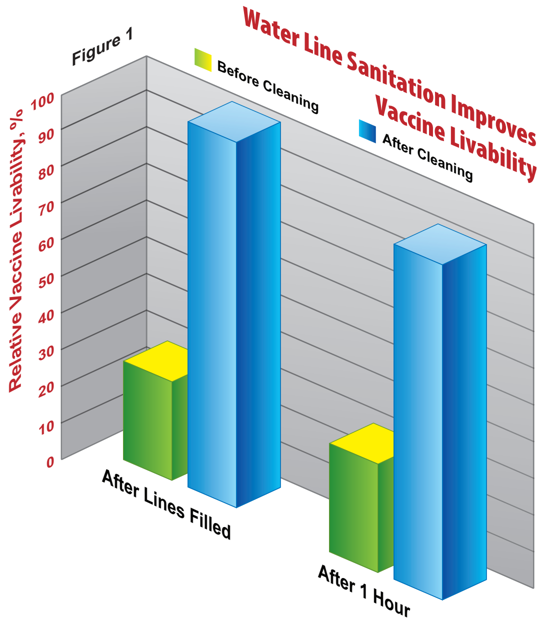
Providing Ideal Solutions... Everyday!

IDEAL MANAGEMENT PRACTICES TO OPTIMIZE IMMUNIZATION

Don't Just Vaccinate, Immunize

Oral mass vaccination of poultry has become the most widespread method of delivering vaccine to birds, mainly because of its low labor requirement. Unfortunately, various factors interact to influence the success or failure of the vaccination program. It has been correctly stated that vaccination is not immunization. The two have a cause and effect relationship, and the simple act of vaccination does not guarantee that good immunity will result. Doing a poor job of vaccination will assure poor protection from disease, while those who take pains to do vaccination well can benefit from the highest level of protection.

Some of the factors that impact vaccination success are not under the control of the vaccinator. Examples are the "bird factors" such as genetic hardiness, maternal antibody interference, stress level, nutritional adequacy, strength of the disease challenge, and even social dominance. Other factors impacting success are related to the vaccine itself. These "vaccine factors" include vaccine hardiness, the effectiveness of the vaccine's stabilizing and preserving additives, the initial potency of the vaccine, and its handling and storage conditions.



Outside of these bird factors and vaccine factors, there are the "management factors" that are under direct control of the vaccination team. This is where vaccination will either succeed or fail. The "man" is the indispensable part of vaccination management, and without good control of the management factors, immunity will be eroded. Erosion is the best way to think of how vaccines fail. There are many cumulative layers of good management practices that all stack up to help vaccines build solid immunity. Neglecting any of these practices means a layer of disease protection is stripped away, leaving less than 100% of the desired immunity and resulting in less than 100% of the potential productivity. It is helpful to split the successful vaccination program into 3 areas: preparing the house, preparing the bird, and preparing/delivering the vaccine.

PREPARING THE HOUSE

The best way to start preparing the facility is not really a start at all, but rather a continuation of your routine water line sanitation practices. Because organic matter buildup in the water line can reduce the vaccine's effectiveness, routine line sanitation must be done to realize full protection from your vaccine. A constant flow of chlorinated drinking water and routine use of acids are helpful in reducing or eliminating harmful organic growth in the water line. If organic matter has accumulated in the lines, it is very important to remove it before vaccination.

To demonstrate the harmful effect that trapped organic matter has on a vaccine, researchers measured vaccine titers in the drinking water before and after cleaning and flushing the lines (Figure 1). The results suggested that the organic matter was inactivating over 75% of the bronchitis fraction of a Newcastle/bronchitis vaccine used on this farm (Heins-Miller, 1993).

The "man" is the indispensable part of vaccination management, and without good control of the management factors, immunity will be eroded.

THE CLEAR CHOICE FOR IMPROVING WATER FLOW RATES

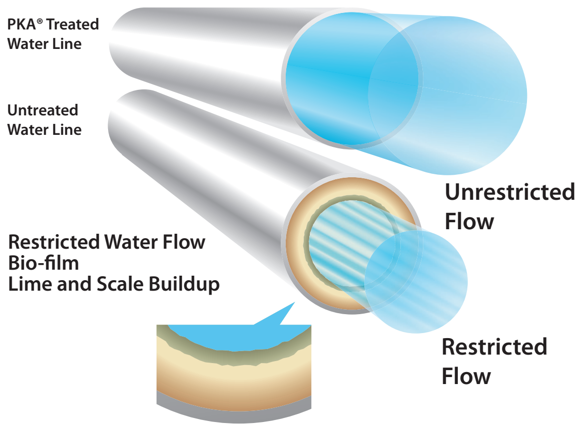
The build-up of scale, algae and bio-film in water supply lines negatively impacts water line function in poultry production facilities. Recent work by Dr. Berry Lott, et al., Mississippi State University, provided useful information on the management of water intake and the impact on broiler performance. In addition to posing a risk to vaccines, mineral deposits and bio-film can result in flow restrictions, especially in nipple drinkers.

REMOVING SCALE AND BIO-FILM BETWEEN FLOCKS USING PKA®

Lime and mineral scale, bio-film build-up and contamination of a watering system will occur with routine usage. Birds consuming this water will be exposed to an increased microbial load through the contaminated system.

Removal of accumulated bio-film layers and mineral deposits from water lines and nipple drinkers will improve water flow, decrease microbial challenges, and reduce the negative impact on vaccines. PKA® dissolves lime and scale, helping to restore and maintain full function to affected water systems. PKA is approved by the U.S. National Sanitation Foundation (NSF) for pH adjustment, corrosion and scale control.

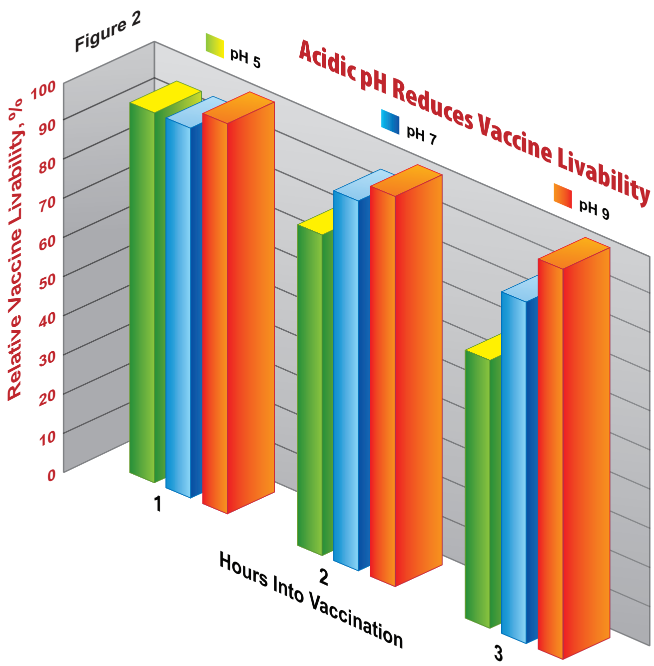
Effect of Scale and Bio-film on Water Flow



An untreated water line contains biofilm, causing potential health risks and water flow restriction

MAINTAINING CLEAN WATER LINES DURING GROWOUT

A PKA® water acidification program will prevent mineral scale build-up and maintain optimum water flow resulting in enhanced flock performance. Between flocks, add 1 pack of PKA to each 970 liters of water. Allow the solution to cleanse water lines for a minimum of 8 hours, up to a maximum of 24 hours. Flush thoroughly with fresh water after cleansing is complete.



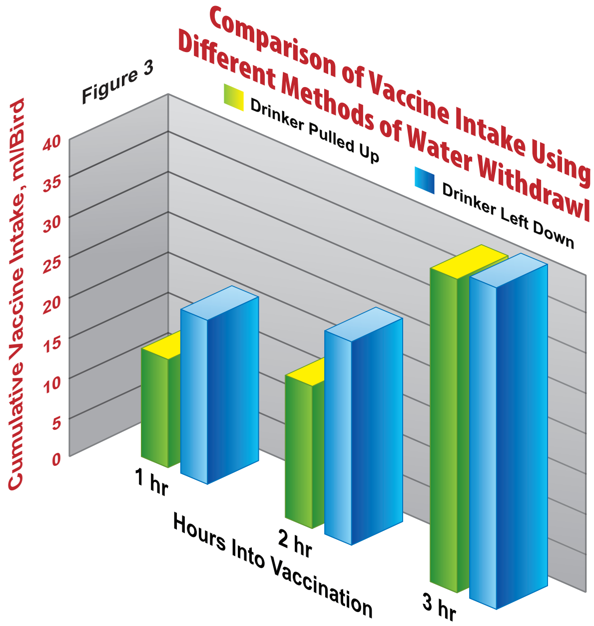
After treating the water lines with PKA® to remove organic matter, it is important to remove or bypass filters that might expose the vaccine to trapped organic matter before vaccinating. Rinsing as much acid as possible from the lines is also key, since residual acids are potentially harmful to the vaccine's potency. The test results in Figure 2 show the loss of potency of an infectious bronchitis vaccine in different pH conditions over a 1, 2, or 3 hour period. The moderately low (acid) pH of 5 greatly reduced the livability of the bronchitis vaccine compared to a more basic pH at 7 or higher (Jordan and Nassar, 1973).

Cleaning the vaccination equipment such as pails, tanks, stirrers and hoses also reduces the transmission of incidental pathogens that might be present. Certain vaccines are also inactivated by water or feed medications. Two days prior to vaccination, these medications should be withdrawn if recommended by the vaccine manufacturer. The final action in preparing the house is to purge the lines with stabilizer to neutralize residual negative water quality factors. Your aim is to build a "buffer zone" of stabilizer to precede the vaccine. Schedule the pre-vaccination stabilizer dose to make sure the water that occupies the lines immediately before the vaccination starts is protected with stabilizer.

PREPARING THE BIRDS

Accurately estimating water consumption in advance will help assure the proper vaccine dose is delivered. This can be done several ways, depending on whether the vaccine is to be delivered by header tank, proportioner, or pumped into the lines. The "dry run" can consist of observing how much blank solution disappears from the header tank or vaccination pail over a mock vaccination period.

Figure 3



When planning vaccinations, consider that sick or stressed birds can have a poorer response to the vaccine, so it is generally best to avoid vaccinating until they are healthy and unstressed. There are exceptions, occasionally your veterinarian may want to vaccinate against a disease in response to a severe outbreak. Typically, birds are orally vaccinated in the morning, when they drink more. Withdrawing water induces thirst in the birds and helps ensure all birds drink more aggressively and consume a higher dose of vaccine. A rule of thumb is to have the birds drink about a fourth of their normal daily water intake in a 2-4 hour period. Vaccinations lasting less than 2 hours may not be long enough to let all the birds consume a protective dose of vaccine, while vaccinating longer than 4 hours risks exceeding the lifespan of the vaccine. In either case, some of the birds may not get a fully protective dose of vaccine. Properly timing the water withdrawal requires some skill in planning. Thirsting the birds for as little as 1 hour in hot weather may be enough, while in cold weather, it may take up to 4 hours.

There are two thoughts on how to withhold water from the birds. The most popular way is to remove the water source by raising the lines, and the other is to turn off the water and let the birds drink the water lines dry. Both ways are effective, and each may have its place in a particular management system. Figure 3 shows the results of a test comparing the two methods. Birds that were thirsted by drinking the lines dry consumed vaccine more aggressively over the first two hours than those that had the water removed, but by the third hour of vaccinating, both groups had consumed the same overall amount of vaccine (Merial, 2002).

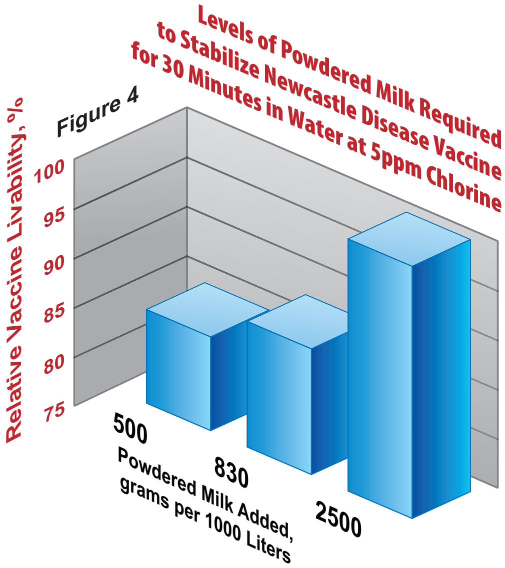
PREPARING THE VACCINE

Live vaccines must remain viable to keep their infectivity and protect your flock. They are vulnerable to several risks, including the triple threat of oxidizers in the water, pH imbalance and low tonicity.

The list of vaccine hazards is a long one. Fortunately, ideal management strategies are available to address each risk, and understanding the obstacles is the first step in conquering them.

STABILITY AGAINST OXIDIZERS

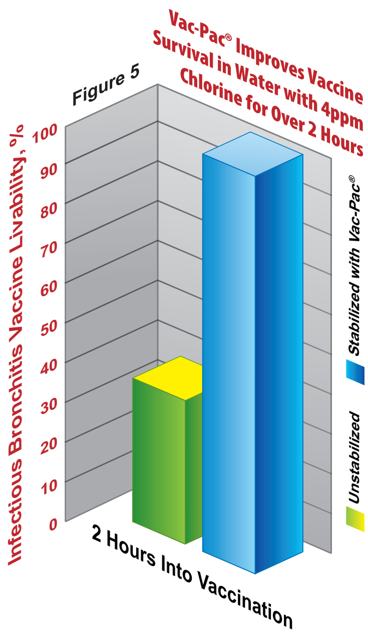
Effective drinking water sanitation is a vital good manufacturing practice in poultry production. Constant chlorination is the most common means of sanitizing drinking water, as it reduces the microbiological load on the birds. However, chlorine and other oxidizers also harm live vaccines and rob flocks of valuable immunity. An advantage of Vac-Pac Plus® is heightened vaccine stability over the entire course of the vaccination. Vac-Pac Plus is manufactured using patent-pending technologies designed to stabilize vaccines against this threat. Vac-Pac Plus rescues vaccines from chlorination spikes that other less powerful products cannot absorb.



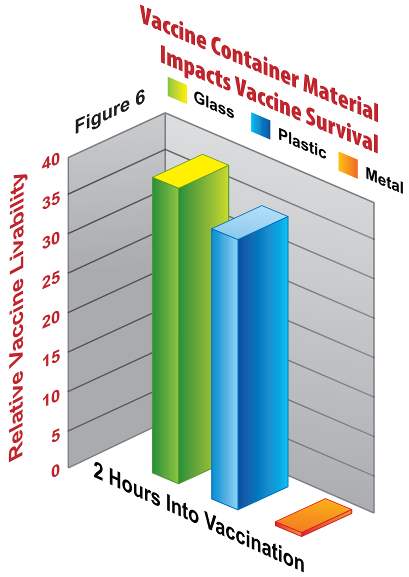
Enough stabilized vaccine should be prepared to last throughout the predetermined vaccination period. Early researchers recognized that chlorine, the chief negative water quality factor that inactivates vaccine, could be neutralized with unsophisticated milk-based stabilizers. Unfortunately, the fact that it takes a lot of milk powder to fully neutralize chlorine was often overlooked. Tests measuring the power of milk to stabilize Newcastle vaccine were performed by Gentry and Braune as early as 1971. Calculations from their data demonstrate that it would take over 2500 grams of powdered skim milk per 1000 liters of drinking water to fully protect vaccine with chlorine at 5 ppm. Figure 4 shows that when using less than 2500 grams per 1000 liters, the vaccine cannot be expected to fully survive in 5 ppm chlorine for even 30 minutes.

SOLUBILITY AND CONCENTRATION

Another limitation of milk-based stabilizers is the poor solubility in water, which slows the vaccination process. Few vaccinators are willing to add the full 2500 grams of powdered milk per 1000 liters of drinking water (or 2 gallons of stock solution), and then wait the recommended 10-15 minutes before adding vaccine. This results in less stabilizer being used, less time being allowed for stabilizer activation, and even warm water being used to dissolve the milk. All these shortcomings contribute to decay in vaccine effectiveness and potential vaccination failures.



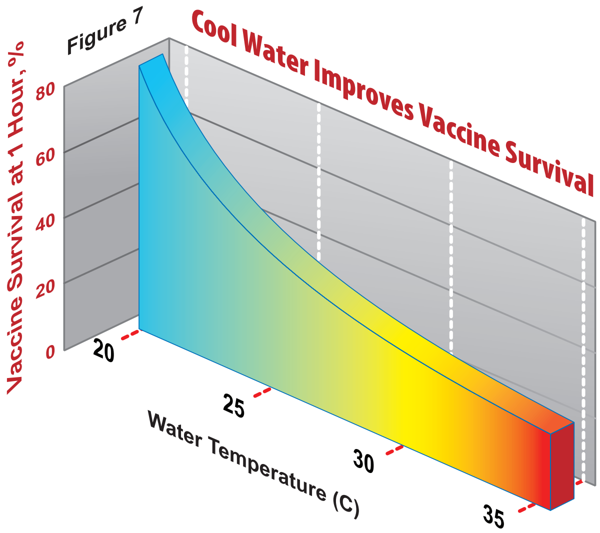
The recent advent of "new generation" Vac-Pac® stabilizers has provided significant improvements over earlier, less sophisticated milk-based stabilizers. The research investment that has gone into new generation products has focused on improving the concentration and effectiveness of the stabilizer. It is now possible to achieve 100% vaccine stability in 1000 liters of water with as little as 100 grams of Vac-Pac (Figure 5). It was previously shown in Figure 4 how milk-based stabilizers could require over 2500 grams per 1000 liters of water to reach a similar level of effectiveness. Further improvements include increasing the "wetting", or how quickly water takes to the stabilizer. Milk powders have a tendency to repel water initially, causing the stabilizer to float in lumps. Once these lumps are wet on the outside, their centers can remain very dry, even if they fall to the bottom of the container. The Vac-Pac® family of stabilizers is "instantized", meaning they get wet and begin to dissolve immediately on contact with water. They not only wet faster, but their ultimate solubility is 100%, even without the use of hot water that would be harmful to the vaccine. The perfect solubility means there is no stabilizer to settle out of the vaccination tank or stock solution. Finally, because the new generation stabilizers are more concentrated, they can be formulated with additional ingredients to address a wider range of negative water quality factors without harming the vaccine.



Vaccines are most often mixed and dispensed from plastic containers today, but occasionally one sees metal tanks or drums in use. It is important to note that some metal containers are not suitable for holding vaccine and should not be used. Research into different containers found that glass and plastic are equally suited, while some metal is poorly suited to keeping vaccines effective. The results of this research are presented in Figure 6, showing the infectious bronchitis vaccine could not survive in a galvanized container for 2 hours (Jordan and Nassar, 1973).

Water temperature is another critical factor that must be managed well for optimum vaccine potency, and it is usually under our control. Always use cool water to make the stock solution instead of warmer water from storage tanks. The harmful impact of increased temperature is shown in Figure 7, which demonstrates that every increase in temperature reduces the amount and duration of bronchitis vaccine livability (Jordan and Nassar, 1973).

When it is time to begin vaccinating, remove the vaccine from cold storage. If it must be reconstituted, protect its potency by using the cool stabilized stock solution as the diluent instead of unstabilized water. Always use the full dose recommended by the vaccine manufacturer. There are already more than enough obstacles to overcome in getting the best vaccine performance, cutting the dose risks further diluting the birds' protection. Another great way to get more value out of your vaccine is to fully rinse the vial. It has been stated that rinsing the vaccine vial can recover up to 14 % more doses compared to an unrinsed vial (Halvorson, 1984). Again, only rinse with stabilized water.



When you are ready to begin vaccinating, charge the lines with the stabilized vaccine laden drinking water, taking care to bleed airlocks and check drinker operation. Lower the lines to start vaccinating, and walk the birds to encourage them all to drink. Occasionally stir the vaccine solution, because some vaccine may migrate to the top or bottom of the container and the doses may be unevenly distributed throughout the vaccination period and the flock. Achieving full and uniform dosage is key to getting 100% of the immunity you desire.

POST-VACCINATION STABILIZER FLUSH

When the vaccine is all delivered, do not refill the lines with unstabilized water. The amount of unused vaccine that remains in the lines can be significant, and unstabilized water following the vaccine may inactivate these valuable doses. The only way to take full advantage of these remaining doses is to make sure the fresh water coming behind the vaccine is also stabilized. Stabilizing the incoming water with a proportioner full of stabilizer will provide a protective buffer zone between your vaccine and the unprotected fresh water supply. Since the lifespan of some vaccines can exceed 6 hours, the incoming fresh water should be stabilized for at least that long.

The list of vaccine hazards is a long one. Fortunately, ideal management strategies are available to address each risk, and understanding the obstacles is the first step in conquering them. In our cause and effect relationship between vaccination and immunization, a healthy and productive flock is your reward for carefully managing every aspect of the vaccination process.

VAC-PAC PLUS®

WATER VACCINATION

Profit From Protection

ADVANTAGES OVER MILK POWDER AND EFFERVESCENT PRODUCTS

The Vac-Pac family of vaccine stabilizers represents the newest generation in vaccine protection technologies. Advances in stabilizer technology continue to push these top-performers farther in front of the field. In producing Vac-Pac Plus®, Animal Science Products dominates the stabilizer market with stabilizers that are safer for vaccines and more convenient and profitable for users.

EASIER TO USE

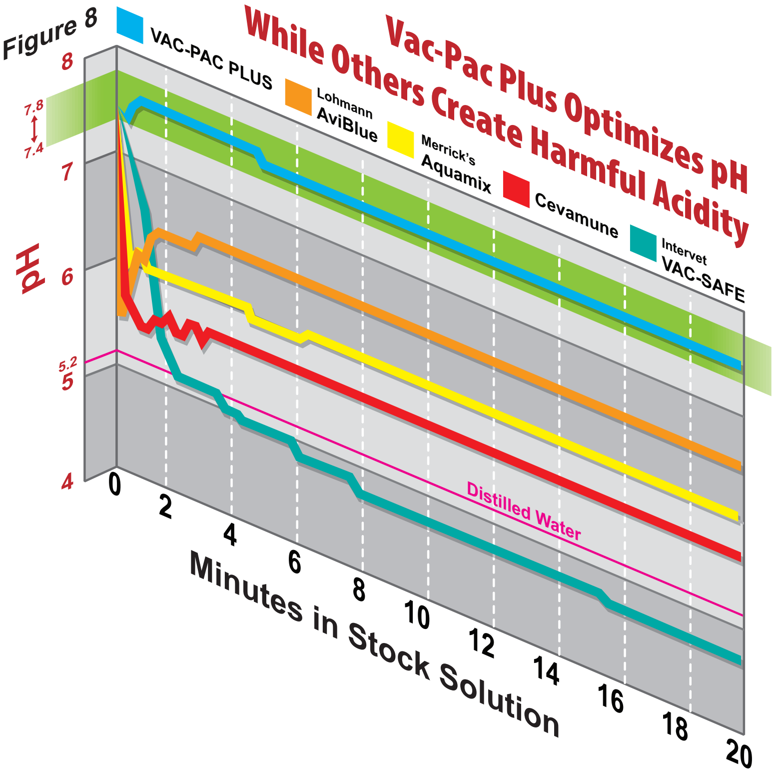
Vac-Pac Plus® is more concentrated and more soluble than obsolete technologies such as slow-dissolving effervescent tablets or animal-derived milk proteins. Vac-Pac Plus' higher stabilizing power and solubility offers faster vaccine preparation time. Slow-acting competitors carry label warnings to wait 15 minutes after mixing the stabilizer before adding the vaccine. Vac-Pac Plus needs no such warning because its stabilizing action is immediate. There is no waiting required. The stabilized vaccine diluent is immediately safe and ready to receive the vaccine as soon as Vac-Pac Plus has been stirred into the solution. Another benefit that makes Vac-Pac Plus more convenient is the new, dust-free formulation. Thanks to advanced granulation technologies, customers now enjoy the same intense blue color in the vaccine solution in a form that is even easier to use.

It also offers the strongest protection from a wider array of risk factors. These benefits all combine to build the greatest protection for vaccines, poultry and profit.

VAC-PAC PLUS®

SAFER FOR VACCINES

Figure 8



The triple threat of oxidizers in water, pH imbalance and low tonicity are key risks that can inactivate live vaccines, making them worthless. Vac-Pac Plus protects vaccines from this triple threat, while other products cannot stabilize against all these risks.

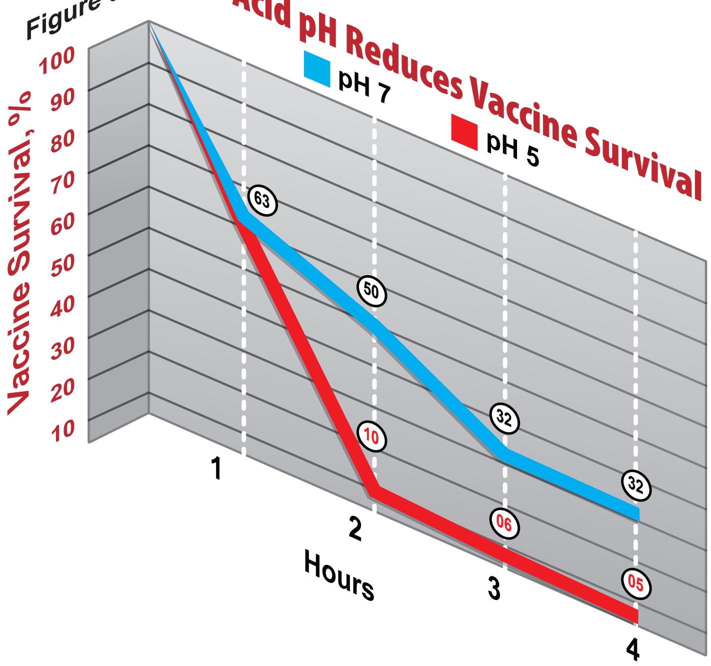
Vac-Pac Plus protects vaccines from oxidizers at much higher concentrations than you will typically find in water supplies (refer to Ideal Management Practices section, Figure 5). Vac-Pac Plus also absorbs shocks from occasional chlorine spikes, giving your vaccines an added measure of safety. Researchers have proven Vac-Pac Plus stabilizes sensitive vaccines in chlorinated water, retaining all of the original doses for over two hours.

Vac-Pac Plus also contains a sophisticated buffering system that sustains an ideal pH for vaccines at about 7.8. Vac-Pac Plus maintains this ideal pH regardless of whether the water used in the stock solution is acidic or alkaline. Several competitors ignore the vaccine's need for buffering and actually take the opposite approach. They attempt to overcome poor solubility in their powders and tablets by making them effervescent. Strong acids in these effervescent stabilizers generate violent bubbling. Excess acid immediately builds, driving the pH to harmful levels for vaccines. The graph (Figure 8) compares the optimized pH of a stock solution stabilized with Vac-Pac Plus to the acidic pH generated by several competitive effervescent tablets or powders.

The effervescent products all caused the stock solution's pH to drop within 15 seconds. They all required a wait of 10-15 minutes before dissolving and the acid condition persisted after the stabilizer was dissolved. Any vaccines added to these effervescent solutions would be exposed to excess acid which puts vaccine titers at risk.

How much titer decay might you expect from acidic effervescent products? Research shown in Figure 9 illustrates that only 10% of a vaccines' original titer remained after 2 hours in a pH 5 solution, and the titer fell to 6% by 3 hours. It highlights the enormous risk effervescent products pose to vaccines, even during the course of a normal vaccination period.

Figure 9



VAC-PAC PLUS®

WATER VACCINATION

BROADER SPECTRUM

For optimal stability, bacterial vaccines such as Mycoplasma and Salmonella have different biological requirements than viral vaccines such as Newcastle, Bronchitis and Influenza. These bacterial vaccines require a more tonically balanced environment to maintain optimal potency. Vac-Pac Plus helps satisfy this biological need by providing more ideal tonicity or osmolarity. Vac-Pac Plus' improved tonic balance broadens the spectrum of vaccine protection by stabilizing both viral and bacterial vaccines. No effervescent tablet, powder or milk protein offers the same osmotic balance as that provided by Vac-Pac Plus.

Vac-Pac Plus represents the newest generation in vaccine stabilizing technology. It provides significant advances over lesser alternatives such as effervescent powders and tablets or animal-derived milk proteins. Vac-Pac Plus is more convenient for the user because it is highly soluble, provides instant protection, does not contain strong effervescing acids and is now cleaner and more dust-free. It also offers the strongest protection from a wider array of risk factors. Vac-Pac Plus absorbs greater shocks from possible chlorine spikes, it is acid-free and buffered, and it is more tonically balanced to protect a broader range of both viral and bacterial vaccines. These benefits all combine to build the greatest protection for vaccines, poultry and profit.

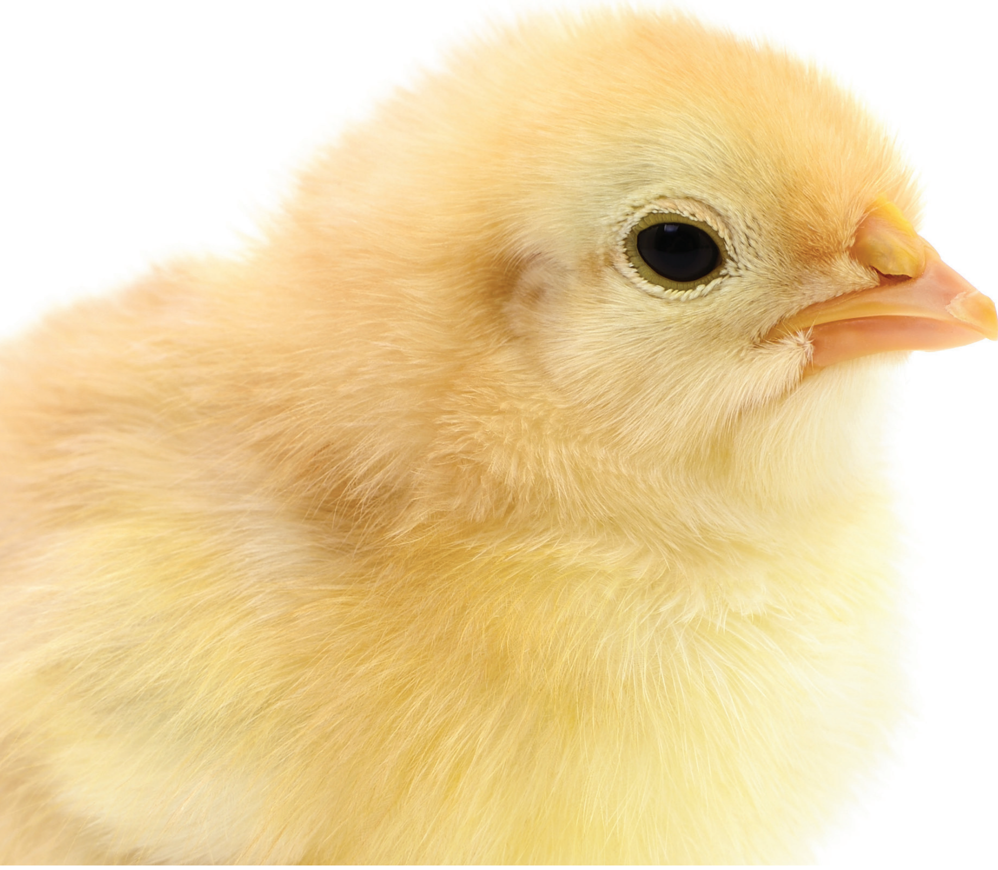
Vac-Pac Plus maintains this ideal pH regardless of whether the water used in the stock solution is acidic or alkaline. Several competitors ignore the vaccine's need for buffering and actually take the opposite approach.

Figure 10 Vac-Pac Plus Benefits Over Competitive Stabilizers

Stabilizing power	Vac-Pac Plus	Effervescent tablet	Effervescent powder	Milk powder
Instantly soluble	+ + +	- - -	- -	- - -
No wait before adding vaccine	+ + +	- - -	- - -	- - -
Surge protection	+ + +	-	-	- - -
Acid-free and buffered	+ + +	- - -	- - -	-
Broadest spectrum	+ + +	- - -	- -	- -
No animal protein	+ + +	+ + +	+ + +	- - -
Dust-free granulation	+ + +	+ + +	-	- - -
Sugar	+ + +	+ + +	- - -	- - -

+ + +	Very Postitive
- -	Very Negative
-	Negative
- - -	Extremely Negative

SPRAY-VAC®



Defend Your Vaccine Investment

Spraying vaccines has become a popular, labor-saving means of administration, and Spray-Vac stabilizer improves the immunity that spray vaccines deliver. Before the new generation of vaccine stabilizing technologies became available, vaccine manufacturers recommended that distilled water be used to spray the vaccine antigen on the birds. They recommended distilled water because it was less toxic to vaccines than chlorinated tap water. One trouble with using distilled water as a diluent is the large volumes of costly spray water required. Among other advantages, new generation Spray-Vac rescues vaccines from oxidizers like chlorine in water, it allows vaccinators to use local tap water to spray vaccines instead of hauling large volumes of costly distilled water. Recently, researchers have discovered that distilled water itself is more harmful to vaccines than many people realized. Distilled water threatens vaccine survival because of inappropriate pH, low tonicity, and temperatures that are often too warm to support optimum immunity. Adding only 32 ml of Spray-Vac to each liter of cool tap water overcomes all these threats and makes an ideal solution for spray vaccines.

RESCUING SPRAY VACCINES FROM OXIDIZERS

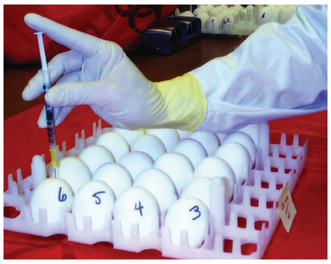
The following research conducted by Lasher Associates proved that Spray-Vac rescues vaccines from the threat of oxidizers in the water, making chlorinated tap water safe and effective for spray vaccines. The purpose of the experiment was to determine (1) if Spray-Vac® Stabilizer affects the viability of a live infectious bronchitis vaccine and (2) if Spray-Vac® Stabilizer exerts a stabilizing influence on the bronchitis vaccine rehydrated in chlorinated water.

In their experiment, a live freeze-dried infectious bronchitis vaccine was rehydrated in water alone, water containing Spray-Vac® Stabilizer, or chlorinated (4 and 8 ppm) water containing Spray-Vac® Stabilizer. Titrations of the virus were conducted in specific-pathogen-free (SPF) embryos at 0, 0.5, and 2.0 hours after rehydration. Spray-Vac® Stabilizer had no deleterious effect on the bronchitis vaccine while chlorine at both concentrations inactivated as much as 80% of the virus. Further, it was shown Spray-Vac® Stabilizer completely rescued the vaccine from degradation by chlorine at both levels tested for as long as 2.0 hours.

SPRAY-VAC®

SPRAY VACCINE PREPARATION

A live commercially available infectious bronchitis vaccine was used. It was reconstituted in various diluents to one label dose per 0.1 ml.



Diluents. Four (4) diluents were employed: (a) water alone, (b) water containing Spray-Vac® Stabilizer, (c) water containing sodium hypochlorite, (d) water containing Spray-Vac® Stabilizer and sodium hypochlorite. The water used was commercially available distilled water. The sodium hypochlorite was added to the diluent water to attain either 4 or 8 ppm available chlorine.

Titrations. Titrations (determinations of live virus concentration) were conducted using specific-pathogen-free (SPF) embryonated eggs. The method was the one commonly used by vaccine manufacturers described in Title 9, Code of Federal Regulations, section 113.327. Briefly, 0.1 ml of 10-fold serial dilutions of vaccine virus were inoculated in the allantoic cavity of groups of six 9- to 11-day-old embryos. Embryo deaths occurring during the first 24 hours after inoculation were disregarded. After 6 or 7 days incubation, surviving embryos were examined for signs of infection, to include, stunting, curling, and clubbing. A satisfactory titer was obtained when at least 4 embryos survived in each dilution, one dilution produced 50 to 100 percent positives, and one dilution, 0 to 50 percent positives. The method of Reed and Muench was used to calculate the EID_{50}* per dose. All titrations were replicated.

> *"One of the most critical aspects of performing spray vaccination is to maintain the viability of the vaccine virus throughout the process."*
> *- Vergil S. Davis, D.V.M., Ph. D.*

EXPERIMENTAL DESIGN

A total of three experiments was conducted.

In the first experiment, in order to ensure that Spray-Vac® Stabilizer did not pose a hazard to the relatively fragile vaccine virus, the effect of Spray-Vac® Stabilizer on the vaccine was compared to the effect of distilled water alone. The lyophilized vaccine was reconstituted in distilled water at the rate of 1000 doses per 100 ml (1 dose/0.1 ml) and then divided equally into two vials. To one of the vials, Spray-Vac® Stabilizer was added at the same dilution recommended by the manufacturer for field use. After 30 and 120 minutes, titrations of vaccine in each of the two vessels, respectively, were conducted.

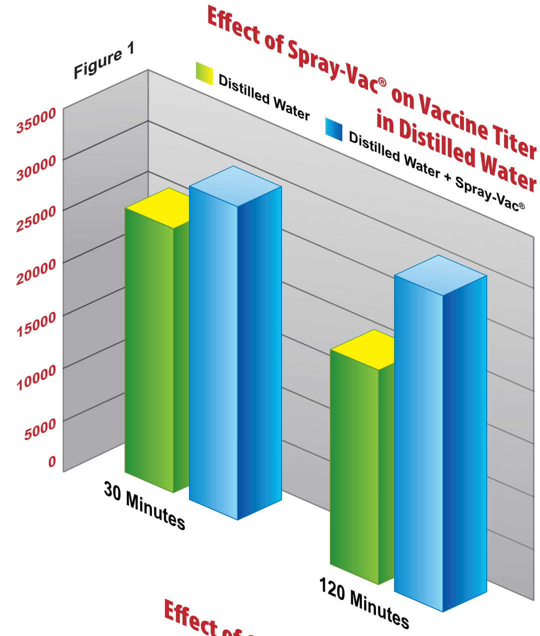
The second experiment evaluated the capacity of Spray-Vac® Stabilizer to protect the vaccine against the detrimental effect of chlorinated water. The experiment compared the viability of vaccine virus rehydrated in chlorinated distilled water containing Spray-Vac® Stabilizer to that of vaccine virus rehydrated in chlorinated distilled water alone. The available free chlorine was adjusted to 4 ppm. The methodology of the second experiment was the same as that of the first except for the addition of sodium hypochlorite to the water used for rehydration.

The purpose and methodology of the third experiment were identical to the second, except that the level of available chlorine was adjusted to 8 ppm.

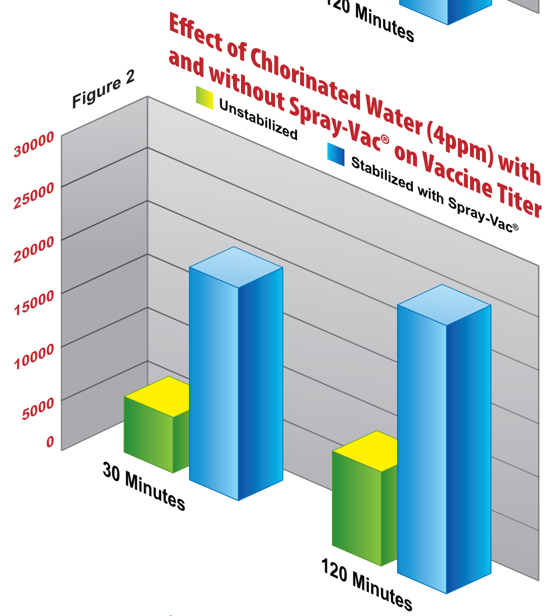
SPRAY-VAC®

PROVEN SAFE AND EFFECTIVE

Effect of Spray-Vac® Stabilizer on the Vaccine. The titers, or concentrations, of the virus rehydrated in water were determined to be $10^{4.4}$ EID_{50} /dose and $10^{4.3}$ EID_{50}/dose at 30 and 120 minutes, respectively. It is interesting to note that the virus titer remained stable in sterile distilled water near neutral pH at room temperature for as long as 2 hours. One reason for this finding might be that the freeze-dried vaccine presented by the manufacturer is mixed with preserving sugars and proteins and these are rehydrated along with the virus. If this preserving quality of the vaccine preparation were actually at work in this instance, it would likely be less of a factor in field applications where vaccine is mixed in much greater quantities of water. For instance, instead of a concentration of 1000 doses per 100 ml as in this experiment, the virus might be used in a back pack sprayer at up to 1000 doses per 1000 ml.

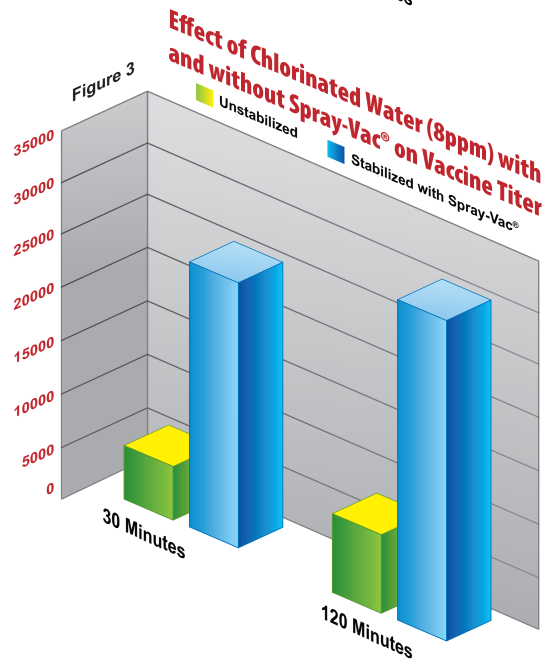


The corresponding titers in water plus Spray-Vac® Stabilizer were $10^{4.5}$ EID_{50} at both time intervals. Note that this titer is essentially equivalent to that of the vaccine in water only, indicating that Spray-Vac® Stabilizer itself posed no detriment to vaccine viablilty (Figure 1).



Effect of Spray-Vac® Stabilizer on the Vaccine in Chlorinated Water (4 ppm). As expected, chlorine at 4 ppm significantly degraded virus titer to $10^{3.7}$ EID_{50} /dose and $10^{3.9}$ EID_{50} /dose at 30 and 120 minutes, respectively. In contrast, addition of Spray-Vac® Stabilizer to the rehydrated virus prior to the introduction of chlorine prevented the virus degradation at both the 30- and 120-minute intervals (Figure 2).

Effect of Spray-Vac® Stabilizer on the Vaccine in Chlorinated Water (8 ppm). As was observed for the addition of chlorine at 4 ppm, the effect of the 8 ppm level was to lower the expected virus titer by 0.7 - 0.8 log10 to $10^{3.7}$ EID_{50} /dose and $10^{3.8}$ EID_{50} /dose at 30 and 120 minutes, respectively. Also, as observed in the second experiment, the addition of Spray-Vac® Stabilizer resulted in complete protection of the virus at the 30-minute, and the 120-minute interval, as well (Figure 3).



Spray-Vac® Stabilizer was developed to allow the poultryman to utilize regular chlorinated tap water as vaccine diluent rather than special deionized or distilled water.

*Titers are commonly expressed as EID_{50}, meaning "(embryo infectious dose)50", which is the dose expected to result in infection of 50% of the embryos. Because the actual numbers can be quite large, often reaching the 6- to10- figure range, they are commonly written in logarithm base 10. Thus, a titer of 10,000 EID_{50} is more commonly seen as $10^{4.0}$ EID_{50}.

SPRAY-VAC®

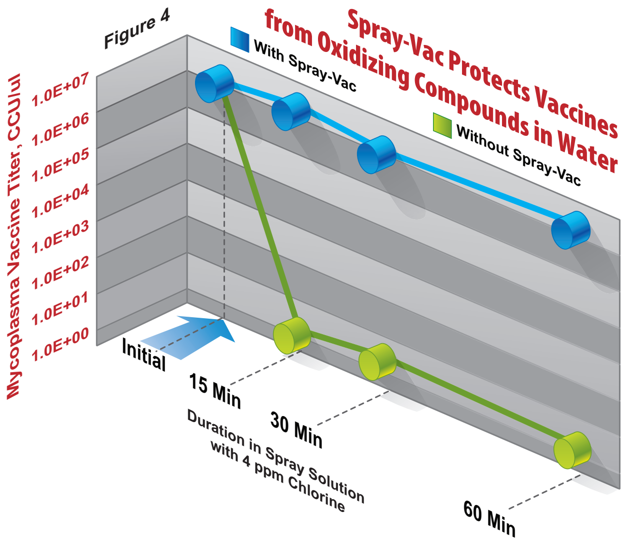
DISCUSSION

Administration of live virus vaccines to poultry by coarse spray is an established practice of modern husbandry. One of the most critical aspects of performing spray vaccination is to maintain the viability of the vaccine virus throughout the process. Among other things, in order to assure viability, the water used for dilution must not be toxic to the vaccine virus. Thus, the recommendation is often made to use deionized or distilled water in the sprayer, especially if the house is on a chlorinated water supply system. In large operations, the task of providing suitable water in adequate quantities to complete a vaccination run can be quite cumbersome and expensive.

Spray-Vac® Stabilizer was developed to allow the poultryman to utilize regular chlorinated tap water as vaccine diluent rather than special deionized or distilled water. This experiment evaluated the stabilizer's effect on one typical live virus vaccine administered by spray and the stabilizer's ability to neutralize chlorine in water so that the vaccine virus remained viable, and thus, fully infective.

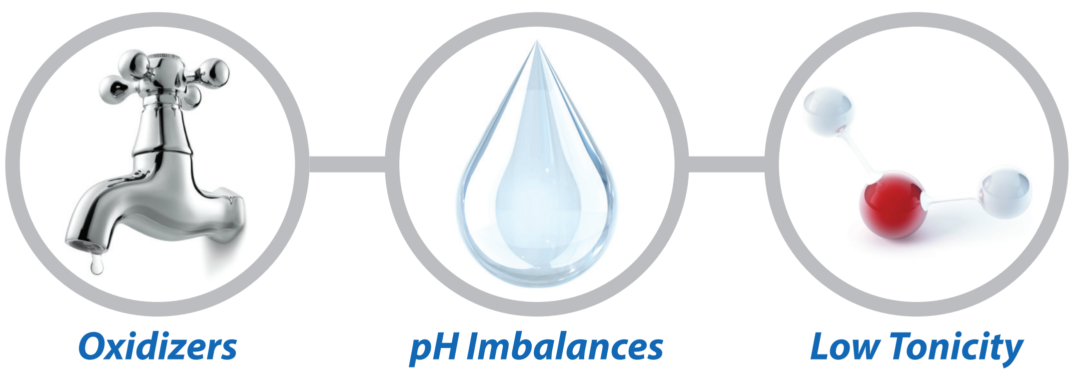
As can be noted from the results, Spray-Vac® Stabilizer is completely safe to the vaccine itself, compared to water alone. As can also be seen from the results, Spray-Vac® Stabilizer completely protected the live bronchitis virus vaccine from degradation by chlorinated water. The vaccine virus in water chlorinated at either 4 (a level typical of public water supplies) or 8 ppm lost as much as 0.7 log10, or 80%, of its original titer. This loss falls within the range of biological significance, especially in situations where the titer of the vaccine at the point of use is close to the minimum protective dose. In contrast, the vaccine's "true" virus titer of $10^{4.4}$ - $10^{4.5}$ EID_{50} /dose was maintained by Spray-Vac® Stabilizer in the presence of the same levels of chlorine.

SPRAY-VAC® ALSO PROTECTS BACTERIAL VACCINES FROM TRIPLE THREATS POSED BY DISTILLED WATER AND TAP WATER



Spray-Vac vaccine stabilizer reduces threats to your vaccines, whether you are using modified live products or bacterins. Risks to fragile vaccines come from all types of water, even distilled. Spray-Vac incorporates the next generation of stabilizing technology to protect vaccines from negative water quality factors, such as, oxidizers, low tonicity, and pH swings. Distilled water, frequently recommended as a spray diluent, is far from ideal for some vaccines. Spray-Vac stabilizer, added to your tap water, allows you to replace distilled water with a more ideal solution.

Protect your vaccine investment from the Triple Threat



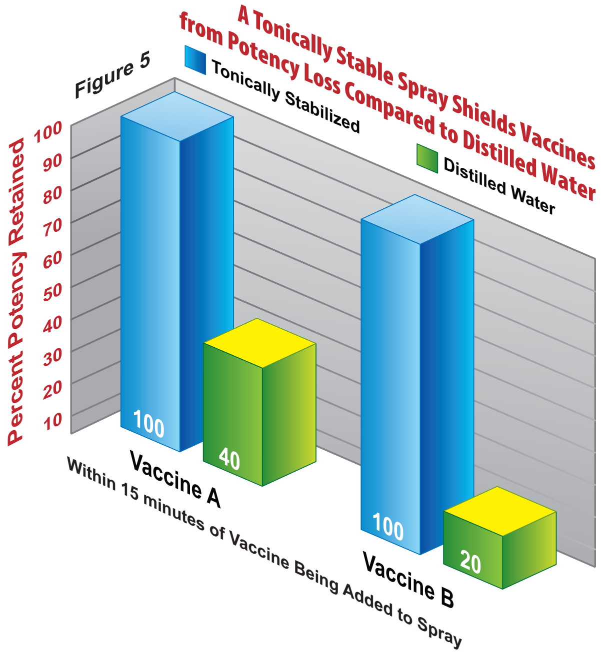
SPRAY-VAC®

DECAY FROM OXIDIZING COMPOUNDS

Just as Davis and Lasher (2000) found that Spray-Vac protects live virus vaccine from chlorine in water, other researchers have discovered a broader range of benefits. Recently, as part of an on-going vaccine stability project, USDA researchers determined that Spray-Vac prevented inactivation of a fragile, live mycoplasma vaccine diluted in chlorinated water (Leigh and Branton, unpublished). Their data, depicted in Figure 4, shows the vaccine's dramatic stability improvement in tap water solutions containing Spray-Vac. Chlorinated water completely inactivated the vaccine in less than 15 minutes, while Spray-Vac® protected it for at least an hour.

THREATENED BY LOW TONICITY, OR OSMOLARITY

Whole-cell bacterial vaccines survive best in an environment where tonicity, or osmolarity, is properly balanced. Vaccine spray solutions with low tonicity are also called "hypotonic". This can be an especially harmful condition occurring when electrolytes are in short supply. Distilled water has low tonicity because the distilling process removes electrolytes. When placed in distilled water, the cells of whole-cell vaccines adapt by pulling water in through their outer membranes. Water uptake continues as the cell's electrolytes are diluted to the same tonicity as the spray solution. Within minutes, some vaccine cells take in enough water to swell and, ultimately, burst. What remains is ruptured cell debris instead of effective vaccine. In addition to distilled water, tap water has low tonicity. Adjusting the tonicity of the spray is especially critical for live whole-cell vaccines that lose their protective power almost instantly in distilled water.



USDA researchers Drs. Scott Branton and Spencer Leigh demonstrated this effect by diluting two popular mycoplasma vaccines in distilled water. The vaccines lost 60-80% of their immunizing power in as quickly as 15 minutes, compared to the same vaccines in tonicity-adjusted water (Figure 5). Adding 32 ml of Spray-Vac per liter of tap water corrects the tonicity and shields vaccines from excessive potency losses.

You've invested good money in valuable vaccines to protect your birds from costly diseases. Don't let one of the triple threats from distilled or tap water rob your vaccine of its value.

VULNERABLE TO pH SWINGS

A third risk factor is the pH of the spray water. Vaccines are sensitive to pH swings that stray from the optimal pH, or "sweet spot", with rapid cell death being reported below 6.5 (Rodwell and Mitchell, 1979). The pH of distilled water is well below ideal, and is much more acidic than many realize. Distillation demineralizes water, but it does not bring about a neutral pH. The harmful acidity comes from a natural and unavoidable reaction between carbon dioxide and distilled water. Carbon dioxide from the air contacts distilled water in manufacturing and storage. It then reacts with the water to create carbonic acid, bringing the pH well below the "sweet spot." Recent surveys of distilled water pH confirm the unavoidable presence of acid. The acid pH distribution shown in Figure 6 is from tests performed on distilled water samples from different sources across the U.S. The pH of the distilled water averaged 5.2, well below the optimal pH of 7.8 as specified in B. W. Calnek's Diseases of Poultry.

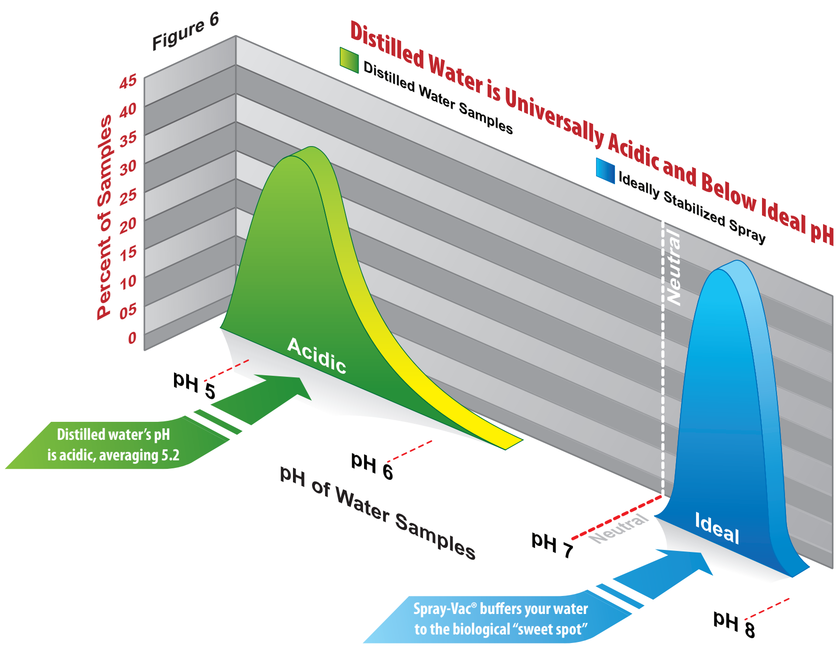
SPRAY-VAC®

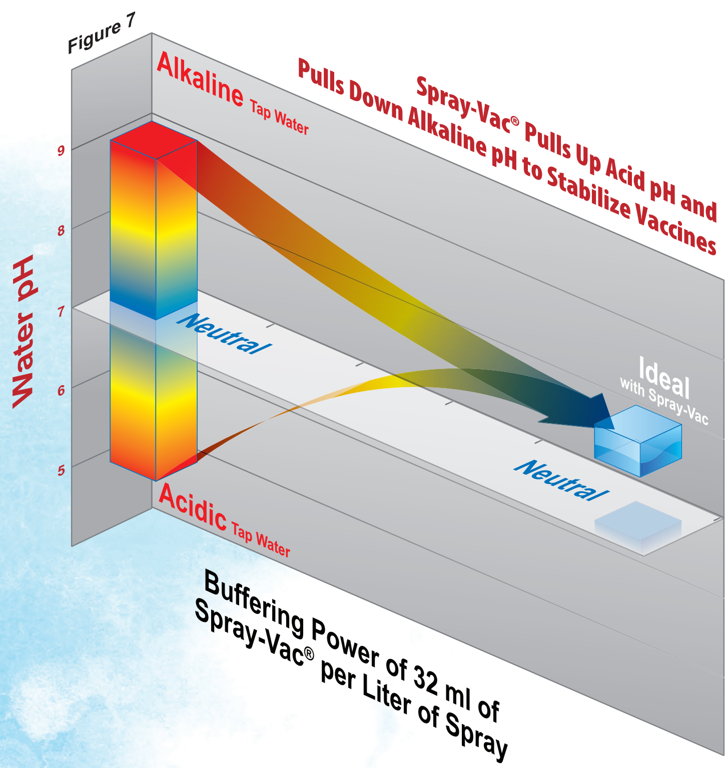
SPRAY-VAC® RESCUES VACCINES FROM STRAY pH

Spray-Vac's buffer system rescues vaccines when pH strays from the ideal value. Regardless of whether your water's pH is acidic or alkaline, Spray-Vac can make it more biologically ideal. Figure 7 shows how effectively 32 ml of Spray-Vac in each liter of tap water can correct pH levels that are as high as 9 or as low as 5. It is unlikely that you will encounter water outside these extremes, but rest assured that if you do, increasing the Spray-Vac concentration will correct it too.

- *Protects Vaccine Investment*
- *Maximizes Immune Response*
- *Economic & Effective*

As more vaccines are applied by spray, unrecognized risks threaten to derail vaccination success. These even include risks from distilled water often used as spray diluent. Researchers are responding to the demands of a poultry industry that won't settle for the tired, old performance of yesterday's technology. You've invested good money in valuable vaccines to protect your birds from costly diseases. Don't let one of the triple threats from distilled or tap water rob your vaccine of its value. Protect your efforts and your vaccine investment with the newest generation: Spray-Vac stabilizer.





AEROSOL VACCINATION

OPTI-VAC®

Because Every Drop Counts

Poultry eye-drop vaccines are designed to deliver maximum immunization, and Opti-Vac® eye-drop vaccine stabilizer works to protect every dose. Even though eye-drop application assures each bird gets vaccinated, it does not always guarantee optimal vaccine stability or resulting immunity. This is because inappropriate diluents are frequently used to deliver eye-drop vaccine. When eye-drop vaccines are rehydrated with distilled water or ordinary sterile diluent, they immediately begin to lose large portions of their activity. The vaccines die quickly because the water used to dilute them is not biologically ideal. Ocular vaccines and the immunity they provide are too valuable to sacrifice performance by administering them with an unsuitable diluent. Opti-Vac provides an environment that is designed to meet the vaccine's needs, preserving valuable titers drop after drop.

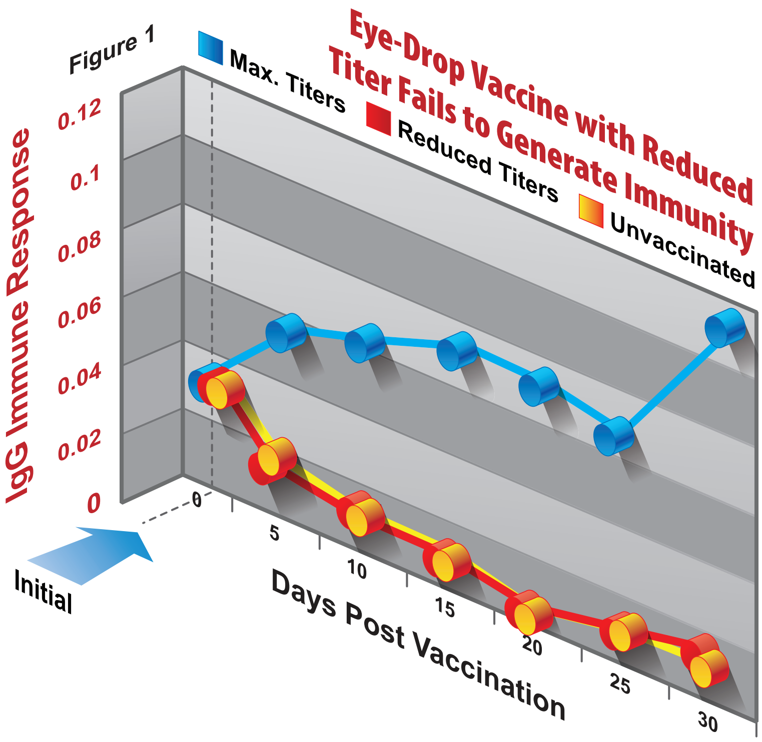
Rehydrated eye-drop vaccines must be stable if they are to retain optimum potency and generate full immunity. Toro et al. (1997) demonstrated how a decline in eye-drop vaccine titers can produce an inadequate immune response. Hens that were vaccinated with an optimal dose of infectious bronchitis virus (IBV) vaccine (10^6 EID_{50}/ml) generated significantly higher immune responses than hens receiving a vaccine in which titers had dropped to a lower concentration (10^4 EID_{50}/ml). The hens receiving the low-titer vaccine produced an immune response that was just as poor as the birds that had received no vaccine at all (Figure 1). An erosion in vaccine titer rendered the vaccine completely useless. Stabilizing eye-drop vaccines with Opti-Vac is an important step in preventing titer loss and ensuring that the first dose delivered is as potent as the last.

Modern vaccines and the performance they offer are too valuable to waste. Opti-Vac eye drop stabilizer works with vaccines, stabilizing and protecting them to optimize the efficacy of each dose delivered.

OPTI-VAC®

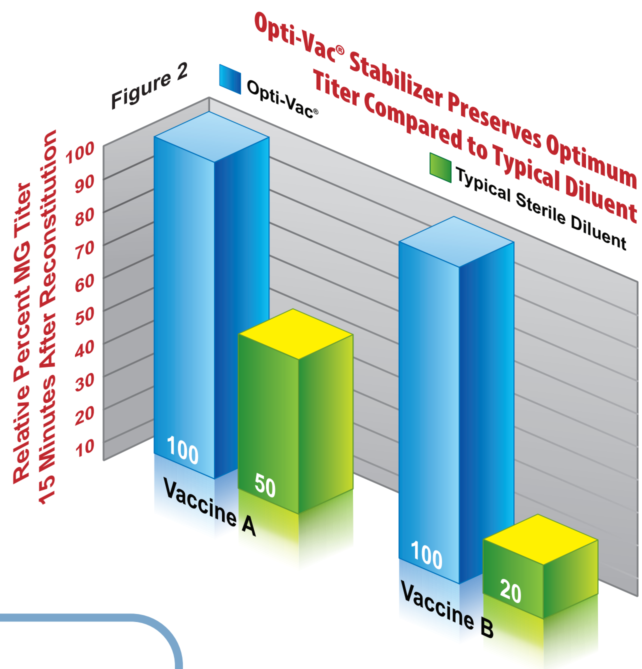
OPTIMUM PH FOR ANTIGEN SURVIVAL

Unstable sterile diluents pose several hazards to live vaccines. Ordinary distilled water diluents are acidic by nature. This causes a rapid loss of vaccine titer after reconstitution. A survey completed by the United States Department of Agriculture (USDA) showed that distilled water diluents have an average pH of 5.2 with a range of 4.9 to 6.2. Most live vaccines require a more neutral pH (7.4 - 7.8) to survive. Vaccines that are diluted with an acidic diluent die very rapidly, meaning most of the flock will not receive the proper dose to develop good immunity. Because Opti-Vac eye-drop stabilizer keeps vaccines at the ideal pH, titers are maintained until the very last drop.



ISOTONIC ENVIRONMENT TO PROTECT VACCINE TITER

In addition to requiring a proper pH, many live vaccine antigens also require an isotonic environment. Low tonicity is particularly detrimental to fragile bacterial vaccines, such as Mycoplasmas. USDA researchers compared the survival of two MG vaccines diluted with isotonic Opti-Vac stabilizer or a typical hypotonic diluent to demonstrate this effect (Leigh et al., 2008). Using Opti-Vac to rehydrate the vaccine prevented a loss of titer, preserving the full strength of each dose (Figure 2). The ordinary sterile diluent caused a 50-80% loss of the vaccines' titers when compared to vaccine rehydrated with Opti-Vac. Birds receiving the vaccine mixed with ordinary diluent would receive only 20-50% of the proper dose needed to develop a solid immune response.



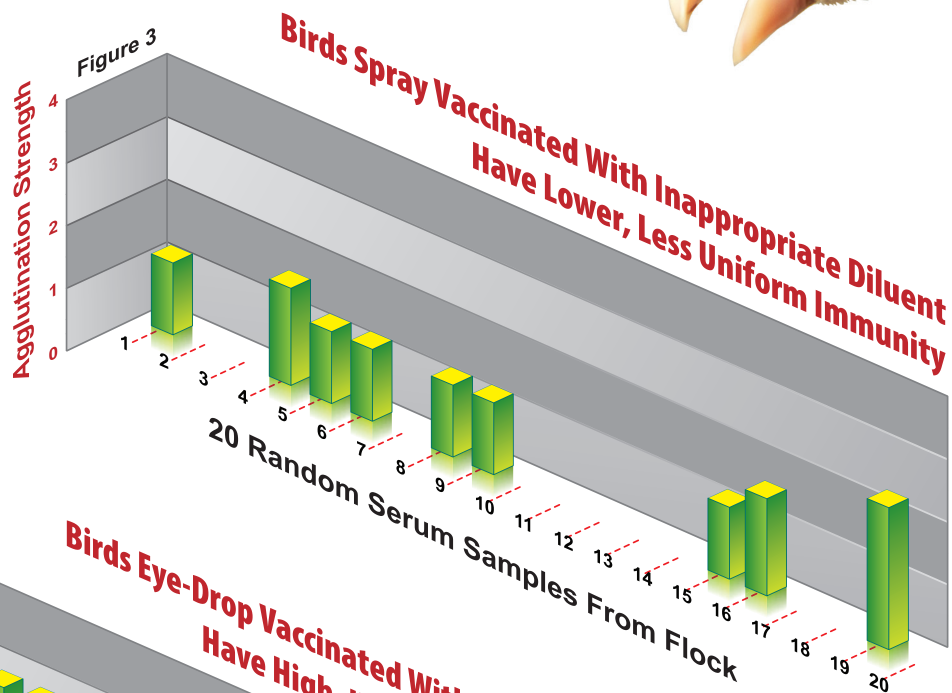
- *Optimum pH for Antigen Survival*
- *Isotonic Environment to Protect Vaccine Titer*
- *Protects Antigen Conformation*
- *Prevents Vaccine Clumping for Even Dosing*
- *Provides Unsurpassed Uniformity*

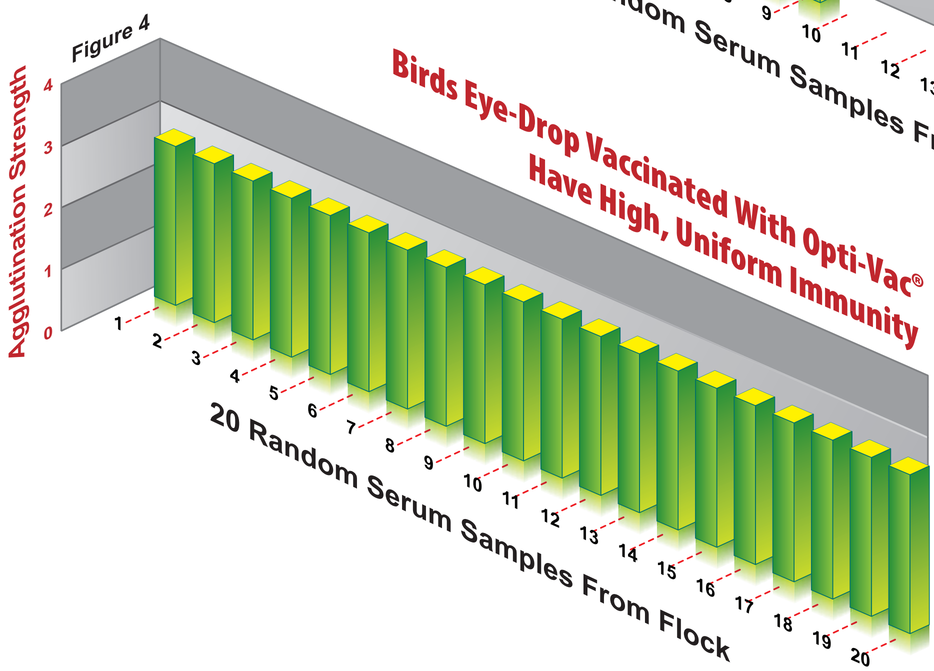
OCULAR VACCINATION

OPTI-VAC®

Opti-Vac's ability to produce strong, uniform immunity has also been demonstrated in the field. Serum plate agglutination (SPA) results using data from commercial laying hens illustrate Opti-Vac's positive effect on vaccine titer and uniformity. Figure 3 represents a flock vaccinated via spray, with a typically non-uniform immune response and weak agglutination scores. Results like these indicate that many birds in the flock did not receive a sufficiently high titer of vaccine. These birds are then susceptible to MG infection. The birds in the flock shown in Figure 4 were vaccinated via eye-drop using a vaccine mixed with Opti-Vac. This Opti-Vac combination provided 100% of the birds with the proper dose of the vaccine resulting in stronger, more uniform titers and more protection from any exposure to MG.

- *Protects Vaccine Investment*
- *Maximizes Immune Response*
- *Economic & Effective*





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Vaccine STABILIZERS

ANIMAL Science PRODUCTS

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VAC-PAC PLUS®
OPTI-VAC®

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