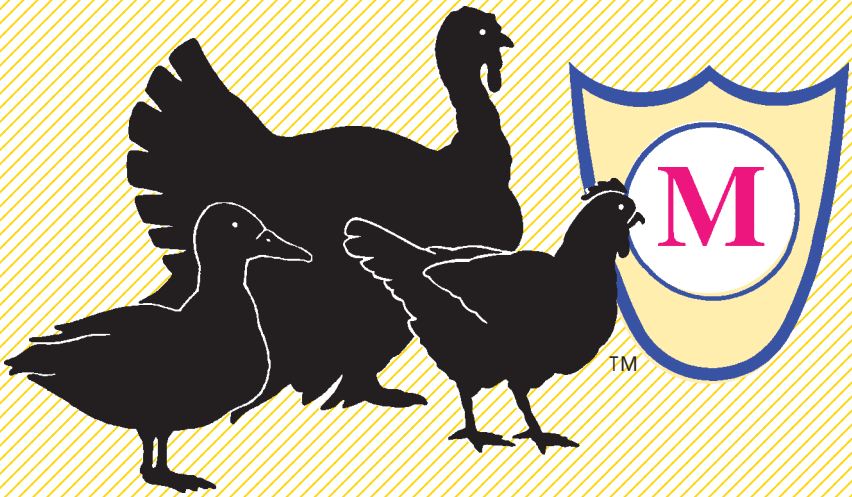
– USE MANUAL –

Merrick's

VACCI-GUARD®



Protects and Stabilizes In-Line Poultry Vaccines

– USE MANUAL –

WHY VACCI-GUARD?

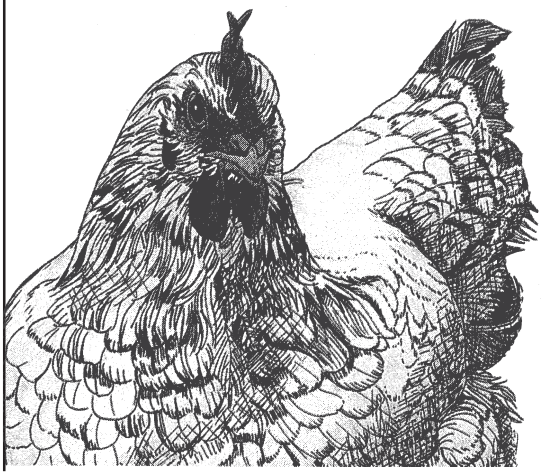
VACCI-GUARD is used to stabilize or neutralize the chlorine in water during vaccination of poultry. Water chlorination is done on many farms to kill bacteria in the water. Chlorine is well known as a bacteriocidal agent and is the active ingredient in many cleaning agents and sanitizers. Chlorine will also have a bacteriocidal effect on live, modified live bacterial and viral vaccines. When birds are going to be vaccinated, chlorination is normally stopped for several hours prior to vaccination. **VACCI-GUARD** is put through the water lines to help neutralize the negative effect of chlorination on bacterial and viral vaccines before, during and after the vaccination process. This neutralization of chlorine is independent of vaccine type.

VACCI-GUARD was developed after extensive testing of many different ingredients used at different concentrations to determine their ability to neutralize chlorine in water. Water chlorination concentrations were varied for testing. If the chlorine compounds were successfully neutralized, the vaccine would have stronger survivability as demonstrated in the testing of our **VACCI-GUARD** product. Pasteurella multocida vaccine was selected as a representative vaccine for initial testing.

VACCI-GUARD contains dye (coloring agent) so it can be seen when it goes through the water lines. The dye can also be observed on the beaks, tongues and feathers of birds that have consumed it. When mixed with vaccine solutions, the dye in **VACCI-GUARD** provides a excellent method of evaluating the number of birds that have consumed the vaccine solution.

VACCI-GUARD is better than other vaccine stabilizers on the market because it is the only one with research data to substantiate that it does reduce the negative effect of chlorine and does stabilize the vaccine in water. **VACCI-GUARD** is agglomerated (special processing) to enhance mixing into water and contains a dye to stain the beaks and tongues of the birds.

This guide is intended to highlight and discuss many of the variables affecting the success or failure of water vaccination programs.



ADMINISTRATING VACCINES

Administrating vaccines through the drinking water is a commonly used method of vaccinating poultry flocks around the world. When properly performed, this can be an effective and economical method of protecting poultry flocks. For diseases such as Avian Encephalomyelitis , Infectious Bursal Disease and Reovirus, oral entry is the natural route into the body, so oral vaccination may be the preferred route of vaccination. These disease organisms also grow and reproduce in the intestinal tract. Vaccines against Newcastle, Infectious Bronchitis and Infectious Laryngotracheitis are also commonly administered in water vaccination programs.

Many variables can affect the success and effectiveness of water vaccination programs. Some factors are directly influenced by the birds themselves, some are influenced by the vaccine and some are influenced by equipment and techniques used to supply the vaccine to the birds.

Bird factors include immune status, water consumption, antibody levels in the egg, social "pecking" order, and watering space. Vaccine factors include bacterial/viral concentration and stability in water and vaccination timing. Technique factors include water quality, water withdrawal time, type of waterers, method of delivery and time from vaccine mixing to administration and consumption, and ambient temperature.

In some areas of the world, certain vaccines are applied by aerosol spray. Newcastle and Infectious Bronchitis vaccines are now commonly applied by this method. Summary of a field trial using **VACCI-GUARD** during aerosol application of these two vaccines is presented in Appendix A.

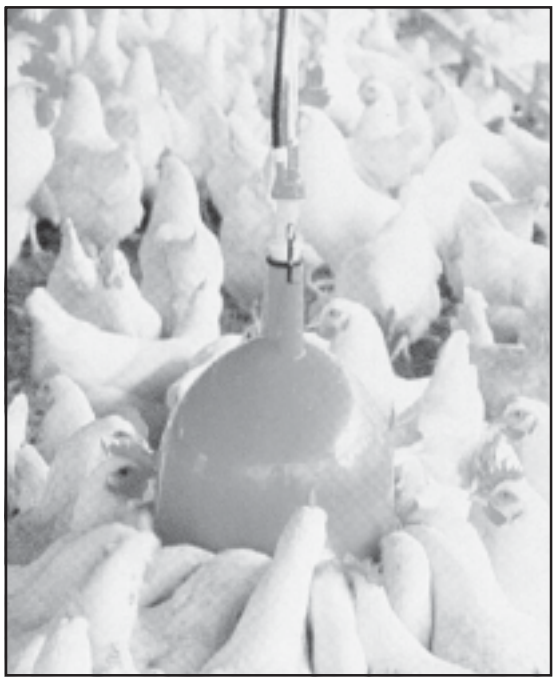
To be effective, all vaccination programs must be conducted in conjunction with a high-standard sanitation program. This is true whether we are talking about eggs, chicks, poults, pullets, full-grown chickens or turkeys. There is a very simple answer to what needs to be cleaned and disinfected – *everything*! Everything that may come into contact with the egg or bird, including personnel should be part of a thorough, consistent sanitation program.

Water Vaccination Methods

Preparing the Watering System for the Vaccine

Several methods have been adapted to administer vaccines in water. The two most common methods use a proportioner pump or water medicator system. These systems are used in closed watering systems with watering nipples. In areas with poor water quality and/or high dissolved solids, an in-line filter may be needed prior to the pump or medicator. Filtration may not be adequate or effective in some areas. For this reason, these two methods may require excessive maintenance to the point that they are impractical under these conditions. In these areas, mixing tanks with metered gravity flow or a straight gravity flow system into the water lines may be workable alternatives.

If bell-type waterers or trough drinkers are used in an open watering system, the waterers should be thoroughly scrubbed with sanitizers and rinsed with plain water a few days prior to vaccine administration. Vaccine stabilizers help neutralize chlorine and many other chemical agents but cannot neutralize large amounts of organic matter in dirty waterers or water lines. Water chlorination and the use of water-applied medications should be discontinued 3 days prior to vaccination to minimize the negative effect of these compounds on the vaccines.



Concentrated citric acid (2 pounds per gallon of water or 1 kg per 4 liters of water to make a stock solution for water medicator use) can be used 1 to 2 days before vaccination to clean the water lines. This treatment will help break up mineral deposits, rust and algae in the water lines. Acetic acid (vinegar) and household ammonia cleaners have also been used. The water lines should be thoroughly flushed with plain water following the citric acid (or other compounds) treatment to remove any loosened material and to reduce the negative effect of the cleanser residues on vaccine stability**.**

VACCI-GUARD should be run through the water lines for the 24 hours just prior to vaccination to neutralize any residual sanitizer or chlorination residues.

Critical Factors for Successful Flock Immunization

Thirsty Birds

The birds must be thirsty and ready to consume water when the vaccine is administered in the water to increase probability of uniform, successful immunization.

Withholding water for two hours prior to vaccine administration is a common practice. This water deprivation time must be adjusted to environmental conditions, of which air temperature at the bird level is the most important factor. Higher environmental temperatures (over 85 degrees F or 30 degrees C) may only require one hour of water deprivation, while cooler temperatures (less than 70 degrees F or 21 degrees C) may require four or more hours of water withdrawal to ensure adequate thirst conditions in the birds. If more than one barn is to be vaccinated, the withdrawal of water should be sequenced to ensure that the duration is similar among all barns. This duration should take into account the amount of time between introducing the vaccine in each barn. A standard method of withholding water is to turn off the water, raise the water lines above the reach of the birds as soon as the first few waterers are empty or drain the water lines. The water withdrawal time starts when the waterers are empty and the water lines are raised.

Care must be taken not to create excessive thirst in the birds, which may result in dominant birds consuming more water (and vaccine) while other birds (lower in the social pecking order) receive little or no vaccine. This uneven vaccination results in inadequate protection and varying degrees of vaccination reactions in the flock. For this reason, competition for drinking space must not be a limitation during vaccination.

Not inducing adequate thirst may result in lowered vaccine intake in the birds which results in less than satisfactory vaccination. This also means that the vaccine remains in the watering system for a longer time period. Once the vaccine is reconstituted, the stability of the diluted vaccine solution will vary considerably. When mixing vaccines, the water temperature should be less than 60 degrees F (15.5 degrees C). Some strains of Bronchitis viral vaccines can lose 50% viability within one hour as the water temperature in the water lines begins to rise above 60 degrees F. It is extremely important to read the information bulletin provided with each vaccine.

Water Intake

Typically, birds will consume about 1/4 to 1/3 of their daily water intake in the first 2-3 hours of morning activity. Daily water intake will determine the dilution rates for vaccination of birds at various ages and body weights. Water intake should be determined for each flock in each barn by using in-line water meters. Write down total daily water intake for the entire flock (written down every 24 hours at the same time of day) for 5 to 7 days prior to vaccination. Multiply the daily water intake by 30% (0.30) to determine the amount of water to be offered as a vaccine solution. By recording actual water consumption, the vaccine volume needed can be increased or decreased to ensure recommended dosages are administered over the desired time period. This also increases the chance of successful and cost-effective vaccination of each flock. There may be considerable differences in water consumption by flocks (of the same accomplished, water intake can be estimated from the **VACCI-GUARD** Use Rate Tables found in Appendix B. Typical water intake tables for various age birds and various temperatures are also presented in the flock management manuals provided by many poultry genetic companies.

When calculating the amount of water to be used during vaccination, keep in mind the amount of water needed to completely refill the water lines to and in the barns. If any distances are involved, this amount of water can be significant. Keep in mind that 500 feet (150 meters) of water line may contain 3 to 10 gallons (11 to 38 liters) of vaccine solution, depending on water line size.

If the water supply was turned off during vaccination (when pumps or gravity-feed are used to supply the vaccine solution to the water lines), the regular water supply should not be turned back on until all of the vaccine solution has been consumed.

Water Quality

There are several water quality factors that affect the stability and viability of vaccines, which, in turn, affect the degree of immunization of the flock. The factor with the greatest impact is the amount of chlorine in the water. Chlorine is a basic ingredient in most cleaners and disinfectants used in the poultry industry. It may also be found as a naturally occurring component in some water sources. Water supplies are frequently chlorinated to control water-borne bacteria. The intent of chlorine use in cleaners, disinfectants and water supplies is to kill bacteria and viruses in the environment. Chlorine is also effective in killing bacteria and viruses in vaccines. Water chlorinators as well as other water sanitizers should be turned off for 24 hours before, during and 24 hours after water vaccination.

There are workable solutions to reducing the amount of chlorine in the water and its negative effect on vaccines. The lowest cost method is to expose a large surface area of water to air at elevated temperatures (over 85 degrees F or 30 degrees C) and allow chlorine evaporation over a 48-72 hour period. This method is not very practical for commercial poultry operations due to the need for holding ponds or large open-top storage tanks and allows possible pathogen contamination from droppings of wild birds.

Distilled water can be used to make up the water-vaccine solution for the birds. Again, this is not very practical in commercial operations and has not been proven to increase vaccine viability and effectiveness.

Time of Day

Birds should be vaccinated soon after sunrise because early sunlight stimulates bird activity. On cloudy, overcast days, it may be necessary to use additional artificial light to stimulate flock activity. Under ideal conditions, the vaccine should be completely consumed within 2 hours. With less vaccination time, birds lower on the social order may not have an opportunity to drink adequate water to receive a full dose of vaccine. With greater vaccination time, the viability of the bacteria or virus in the vaccine may be depressed resulting in uneven vaccination of the flock.

Vaccine Stabilizers

Dried skim milk may help neutralize chlorine concentrations of up to 1 ppm when used at the rate of 3.2 ounces per 10 gallons (240 grams per 100 liters of water). In many areas, chlorine levels are well in excess of 1 ppm chlorine. Dried skim milk has been shown to be less effective in maintaining vaccine stability as compared to **VACCI-GUARD** (see research in Appendix C).

VACCI-GUARD is a unique blend of highly soluble ingredients formulated to neutralize chlorine and other water quality factors and increase vaccine viability. **VACCI-GUARD** should be run through the water lines along with the vaccine solution to minimize free chlorine during the 2 hour vaccination period. **VACCI-GUARD** contains a blue water-soluble food-grade dye to temporarily stain the beaks, tongues and feathers of the birds. This dye allows care-takers to monitor birds as they drink water and to monitor the vaccine solution in the waterers during the vaccination process.

VACCI-GUARD use rates are presented in Appendix B. Mix the required amount of **VACCI-GUARD** with water, add the required amount of vaccine to the **VACCI-GUARD** solution, mix well and administer the vaccine solution immediately. All water lines should be flushed immediately until the vaccine solution starts to flow out of the end of the line. Water filters, regulators and air-locks may need to be bypassed during the vaccination process. Water lines should be lowered to allow birds to start drinking the vaccine solution if the water lines were raised during the water withdrawal period.

Iodine is also frequently used as a component in sanitizing agents. Although **VACCI-GUARD** has not been specifically tested with iodine, it would be reasonable to expect similar responses due to the very similar chemical reactivity of chlorine and iodine.

Vaccine Dosage and Handling

It is essential for each bird to consume a full dose of vaccine to provide optimal immune response. This is especially important when the farm has a recent history of disease outbreaks and the birds are challenged with specific diseases. If the farm has not had recent history of a specific disease and the birds are not challenged with a specific disease, then the producer should consult with his veterinarian concerning specific vaccines and flock health programs.

Vaccines must be administered in accordance with manufacturer's directions for maximum effectiveness. Some vaccines will interfere with the effectiveness of other vaccines. In these cases, it would be much more cost effective to vaccinate twice rather than combine vaccines. Consult with the vaccine manufacturer's veterinarians before combining vaccines.

All vaccines must be shipped and stored according to manufacture's directions. This includes refrigerated shipment and storage to prevent degradation of the active vaccine. Vaccines should not be removed from refrigeration or cold storage until time of use on the farm. Do not allow exposure to direct sunlight before use.

Keep accurate records of all vaccines. These records should include vaccine name, manufacturer, supplier, expiration date, serial number, lot number, date and time of vaccination and identification of the vaccination crew. This provides a means of tracing the vaccine and personnel if problems should arise.

When the vaccine is reconstituted, it should be administered immediately in the drinking water. All vaccination programs should include **VACCI-GUARD** to minimize residual chlorine and other compounds that may reduce effectiveness of the vaccine. Make sure that the water is cool or cold when the vaccine is added. Use plastic buckets and tanks rather than metal tanks and pipes when practical. Mixing equipment should be cleaned, thoroughly rinsed with plain non-chlorinated water and allowed to completely dry before the next use.

Disposal of empty vaccine vials and partial bottles of reconstituted vaccine must be in accordance with all local, state and federal regulations.

Vaccinate only healthy birds unless advised differently by your flock veterinarian. There may be times when vaccination is needed during a disease outbreak. The added stress of vaccinations in sick birds may actually worsen the problem, reduce the effectiveness of the vaccine and lengthen the time needed for the birds to develop immunity. All animals, including birds, respond to stress conditions by increased immune system activity. This, in turn, reduces the bird's ability to respond to the additional stress of vaccination. Again, work closely with your flock veterinarian.

Keep in mind that many vaccinations actually induce a mild case of that disease in order to induce active immunity. In general, barn temperature should be increased by 5 degrees F (2 degrees C) over normal recommended temperatures during the first 24-48 hours after vaccination. As with humans, the initial response is usually fever and the birds may actually be chilled during the immune system response to the vaccine.

Other Factors to Consider

Care-takers should walk along the walls of the barns at least two times during the two-hour vaccination process to ensure that all birds move to the waterers and consume an adequate amount of vaccine solution. Allow the birds to consume as much water as possible during the vaccination process before turning the water back on. In nipple waterer systems, the water may need to be turned on to provide adequate pressure for the nipples to operate. **VACCI-GUARD** should be included with the water for 6 to 12 hours after the vaccination is complete to ensure that any residual live vaccine in the water lines is protected.

Poultry production facilities may also use municipal water supplies that are chlorinated. While municipal water is normally better quality, the same procedures and vaccine problems also apply to this water source.

Vaccinations of birds through drinking water can be a low-cost, effective means of disease protection provided that the method is monitored to ensure that vaccines are properly administered. **VACCI-GUARD** provides the ingredients to help protect vaccines in water and monitor the effectiveness of water intake in the birds.

Records should be kept of several management factors for future reference. These include barn temperatures, water withdrawal times, volumes of final vaccine solution, starting and ending time of vaccine administration through the proportioner pump, time (and amount of water) needed to drain and refill all water lines, when did the care-takers walk through the birds and the estimated percentage of birds with dye on beaks and tongues at each walk-through. Walk-throughs of the barns should be completed at 1 hour and 2 hours after the start of the vaccine. After 2 hours, a realistic target is over 80% of the birds being well-stained on the beak and tongue, and less than 10% of the birds having no stains from the dye. It will be rare to find 100% of the birds with dye stains on the beak and tongue.

With some flocks, it may be necessary to restrict feed intake by raising feeders to reduce the time needed for adequate water restriction prior to vaccine administration. Feeders should be lowered and feed consumption allowed when the waterers are lowered to allow water consumption.

Depending on production and environmental conditions, many poultry producers use water electrolytes such as Merrick's Blue Ribbon Poultry Electrolytes or Merrick's Blue Ribbon Poultry Stress Pack for 3 days prior to and 3 days after water vaccination. These electrolytes are also frequently used during starting and moving stress periods.

Typical Problems seen in Water Vaccination Programs

Too much time for vaccine solution to reach all birds: this results in potential reduction in vaccine stability, delayed delivery of vaccine to birds and reduction in effectiveness of water withdrawal – the solution is todrain water lines and refill the watering system under pressure using **VACCI-GUARD** (with blue dye) to reduce residual chlorine and to be able to determine when vaccine is reaching each waterer.

Vaccine solution is consumed in less than one hour: this results in uneven application of the vaccine with dominant birds getting an excess dose and less dominant birds receiving little or no vaccine. In this case, there would be a significant number of birds with no blue dye stains from the **VACCI-GUARD**. The solution is to increase the vaccine solution volume and decrease the water withdrawal time.

Vaccine solution is not consumed in 2 hours: this results in the potential for reduction in vaccine stability after 2 hours, may be a result of over-dilution of vaccine, not enough water withdrawal time and may also be a result of not draining the water lines prior to starting the vaccine and **VACCI-GUARD** solution. The solutions to this problem are to decrease the volume of vaccine solution, increase the water withdrawal period and to drain the water lines prior to starting the vaccine solution.

Airlocks: this results in uneven vaccination and may restrict vaccine delivery to some waterers in the barns – the solution to this problem is to raise the water lines during water withdrawal and introduce the vaccine solution into the water using pressure flow before lowering the water lines.

Birds in corners and along walls (or back areas of batteries) not getting vaccinated: this is a result of water lines that were not drained and recharged with vaccine solution, dominant birds consuming most of the vaccine solution, or excessive water withdrawal – the solution for care-takers to walk through the birds and along the walls during the vaccination period to mix the birds; or for caged birds – turn off the lights while filling the water lines and increase the volume of vaccine solution.

Inadequate watering space: this problem results in dominant birds consuming most of the vaccine solution and a large percentage of unvaccinated birds. The solution is to add additional waterers during vaccination.

VACCI-GUARD university trials are presented in Appendix C.

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APPENDIX A

Summary of field trial results in Central America using **VACCI-GUARD** with aerosol vaccination of birds against Newcastle Disease and Infectious Bronchitis.

Previous vaccination programs in a large poultry company had resulted in highly variable immunity and a large coefficient of variation in serology indicators of immunity. The vaccination program consisted of aerosol vaccination at day 1 and day 18 against Newcastle and Infectious Bronchitis. Using standard management practices of chlorinating the water supply, the water was maintained at 3 ppm chlorine at all times. Chlorine was thought to be interfering with the effectiveness of the vaccines.

VACCI-GUARD was added to the water used in the aerosol spray for 6-12 hours before and 6-12 hours after the aerosol vaccine application. Three to five (3-5) grams of **VACCI-GUARD** per liter of water were used to block the chlorine (3 ppm). Birds were again tested for the same serology indicators. When using **VACCI-GUARD** at the 5 gram per liter rate, the greatest immune response, highest titer levels and most uniform distribution of the vaccine were observed.

This large poultry operation now uses **VACCI-GUARD** in their aerosol vaccination program for Gumboro, Bronchitis and Newcastle Disease.

APPENDIX B

VACCI-GUARD USE RATES

Grams per 1000 birds

	Chickens Broilers/day		Leghorn Pullets/day		Turkeys/day	
Age (weeks)	Water (gallons)	**Vacci-Guard** (grams)	Water (gallons)	**Vacci-Guard** (grams)	Water (gallons)	**Vacci-Guard** (grams)
1	5	91	5	91	10	182
2	13	236	10	182	20	363
3	24	436	12	218	30	545
4	37	672	17	309	40	726
5	53	963	22	400	50	908
6	69	1253	25	454	60	1090
7	85	1544	28	509	75	1362
8	100	1816	30	545	95	1725
9			35	636	115	2088
10			38	690	125	2270
12			40	726	150	2724
15			42	763	160	2906
20			45	817	200	3632

Water intake may vary with temperature and diet composition.

VACCI-GUARD USE RATES

Kilograms per 1000 birds

Age (weeks)	Chickens Broilers/day Water (liters)	**Vacci-Guard** (kilograms)	Egg Strain Pullets/day Water (liters)	**Vacci-Guard** (kilograms)	Turkeys/day Water (liters)	**Vacci-Guard** (kilograms)
1	23	0.10	19	0.09	37	0.17
2	42	0.19	38	0.17	75	0.34
3	67	0.30	45	0.20	113	0.51
4	126	0.57	64	0.29	151	0.69
5	140	0.64	83	0.38	189	0.86
6	170	0.77	94	0.43	227	1.03
7	207	0.94	105	0.48	283	1.29
8	235	1.07	113	0.51	359	1.63
9			132	0.60	434	1.97
10			143	0.65	473	2.15
12			151	0.69	567	2.57
15			158	0.72	605	2.75
20			170	0.77		

Water intake may vary with temperature and diet composition.

VACCI-GUARD USE RATES

Pounds per 1000 birds

	Chickens Broilers/day		Egg Strain Pullets/day		Turkeys/day	
Age (weeks)	Water (gallons)	**Vacci-Guard** (pounds)	Water (gallons)	**Vacci-Guard** (pounds)	Water (gallons)	**Vacci-Guard** (pounds)
1	6	0.24	5	0.20	10	0.40
2	12	0.48	10	0.40	20	0.80
3	17	0.68	12	0.48	30	1.20
4	34	1.36	17	0.68	40	1.60
5	38	1.52	22	0.88	50	2.00
6	47	1.88	25	1.00	60	2.40
7	56	2.24	28	1.12	75	3.00
8	64	2.56	30	1.20	95	3.80
9			35	1.40	115	4.60
10			38	1.52	125	5.00
12			40	1.60	150	6.00
15			42	1.68	160	6.40
20			45	1.80		

Water intake may vary with temperature and diet composition.

Performance of **VACCI-GUARD** compared to dried skim milk as a poultry vaccine stabilizer.

Time (hours)	deionized water	dried skim milk	**VACCI-GUARD**
	(colony forming units per dose X 1000)		
0.0	6.5	17833.5	23700.0
0.5	0.0	19807.5	22250.0
1.0	0.0	16333.0	21000.0
1.5	0.0	14500.0	16150.0
2.0	0.0	14417.0	7835.0*****
2.5	0.0	11250.0	14350.0
3.0	0.0	12350.0	14500.0

Live Pasteurella multocida vaccine was used in all of the research for the development of **VACCI-GUARD**. The vaccine was inoculated at the rate of $1X10^6$ organisms per liter of chlorinated, deionized water (control) plus dried skim milk or **VACCI-GUARD**. Immediately after inoculation, samples were plated in duplicate on trypticase soy agar every 30 minutes for 3 hours. Plates were incubated at 37 degrees C for 24 hours, then colony forming units were counted to determine the survivability of Pasteurella multocida.

*****The low colony count observed with the **VACCI-GUARD** at the 2.0 hour sampling was most likely an anomaly due to lab or sampling error rather than being a true value. This logical conclusion is based on the higher levels seen in samples taken at the 2.5 and 3.0 hour sampling times. If **VACCI-GUARD** did indeed lose effectiveness as a vaccine stabilizer and lose effectiveness in chlorine neutralization, there would not have been higher colony forming unit counts at the 2.5 and 3.0 hour collections.

Evaluation of **VACCI-GUARD** and Dried Skim Milk in Enhancing the Effectiveness of Vaccines Administered by Water Systems to Broiler Chicks. University of Arkansas.

Experimental Protocol:

Part I: Two commercial broiler barns served as the test site. The three treatments were untreated water (control), control plus dried skim milk, and control plus **VACCI-GUARD**. Water chlorine level was 4 ppm and water temperature was 62 degrees F (16.5 degrees C). The vaccine used was Infectious Bronchitis. **VACCI-GUARD** and dried skim milk were used at the rate of 0.5 pounds (227 grams) per 12.5 gallons (47.5 liters) for test vaccine batches for 5000 birds. The water lines were charged with the **VACCI-GUARD** or dried skim milk for 24 hours prior to introduction of the vaccine solution. Approximately one gallon of water was drained from the end of the water line after the color of **VACCI-GUARD** or dried skim milk appeared. Duplicate samples of the vaccine solution were taken from the nipple waterers at the midpoint and end of the water lines using aseptic techniques at 0, 30, 60, 90 and 120 minutes after introduction of the vaccine solution into the watering system. Water flow through the system simulated the water consumption of 5000 two-week old broilers.

Observations and Results

The **VACCI-GUARD** readily dissolved into the water. The dried skim milk was not completely dissolved after 10 minutes and small clumps remained.

Water-vaccine samples (0.2 ml) were injected into embryonated eggs (10 days) via the chorio-allantoic route. Egg shells were sealed with wax, then the eggs were incubated at 37 degrees C (98.6 degrees F) at high humidity. Each water sample was injected into 5 eggs. Eggs were candled daily for 7 consecutive days. All embryos that died in the first 24 hours were discarded. Embryo deaths after 24 hours were considered in the final analysis for the presence of virus. Embryos were considered positive for the presence of the virus if they demonstrated at least 3 of the following: stunting, club feathering, urates, hemorrhage or mortality. Samples were considered positive if 3 of 5 embryos demonstrated lesions.

Table 1. Infectious Bronchitis Vaccine Recovery Following Water Administration.

Time	0		30		60		90		120	
Replicate	1	2	1	2	1	2	1	2	1	2
Control A	5/5 +	3/5 +	5/5 +	5/5 +	5/5 +	5/5 +	5/5 +	5/5 +	5/5 +	5/5 +
Control B	0/5 -	2/5 -	3/5 +	0/5 -	0/5 -	3/5 +	5/5 +	5/5 +	5/5 +	5/5 +
Vacci-Guard A	4/5 +	4/5 +	5/5 +	5/5 +	4/5 +	4/5 +	5/5 +	5/5 +	4/5 +	5/5 +
Vacci-Guard B	0/5 -	0/5 -	1/5 -	2/5 -	3/5 +	5/5 +	5/5 +	5/5 +	5/5 +	5/5 +
Skim Milk A	4/5 +	2/5 -	1/5 -	2/5 -	1/5 -	1/5 -	2/5 -	2/5 -	4/5 +	5/5 +
Skim Milk B	0/5 -	2/5 -	1/5 -	4/5 +	3/5 +	2/5 -	5/5 +	1/5 -	2/5 -	1/5 -

Raw data is expressed as the number of embryos demonstrating lesions of IBV infection over the total number of eggs inoculated for each repetition.

Positive lesions indicate active vaccine. It is of particular interest in this trial that the results for the control and **VACCI-GUARD** seem to be parallel (except for 60 minutes where **VACCI-GUARD** resulted in much higher active vaccine response), while the results using dried skim milk are consistently lower than either the control or **VACCI-GUARD.**

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