The American College of Poultry Veterinarians Presents:

What's New & Coming in Poultry Vaccination? Production, Administration, & Monitoring of Tomorrow's Poultry Vaccines

April 2, 2019

Sheraton Buganvilias Resort & Convention Center

Puerto Vallarta, Jalisco, Mexico

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Dear Colleagues,

Welcome to Puerto Vallarta! On behalf of the American College of Poultry Veterinarians (ACPV), and the ACPV Continuing Education Committee, I welcome you to the workshop, "What's New & Coming in Poultry Vaccination?" In recent years, there have been many advances in poultry vaccination. The objective of this meeting is to highlight the future of poultry vaccines. The focus is on what new technology and R&D is progressing in the specific areas of production, administration, and monitoring of tomorrow's poultry vaccines.

This year we invited a select group of speakers with recognized expertise in the area of poultry vaccination previously mentioned. I would like to express my sincere thanks to these distinguished professionals for accepting our invitation and for sharing their knowledge, experiences and perspectives with our diplomates. I am sure that they will provide us with valuable and updated information that will be very useful to our attendees.

I also want to express my appreciation to the companies and individuals that generously contributed financially to make our workshop possible. With your collaboration, you are helping the ACPV to accomplish its mission. Thank you so much for supporting our college!

This year, I had the opportunity to collaborate with a wonderful group of diplomates as part of our Continuing Education Committee. All your ideas, suggestions and collaboration were so helpful, and at the end of this process, we put together a very diverse and interesting agenda.

I also want to express special recognition to Janece and Nathan Bevans-Kerr and Diana Kerr for all their logistic support.

Finally, we would deeply appreciate all our attendees providing us with their candid feedback to improve future workshops. We hope that you enjoy this workshop, and that the knowledge shared by our outstanding panel of speakers is useful and valuable to be applied in your professional activities.

Sincerely,
Kelli H. Jones, DVM, MAM, Dipl. ACPV, Dipl. ACVM Chair, Continuing Education Committee (2019) American College of Poultry Veterinarians

Agenda

Moderator: Kelli Jones

7:30 – 7:40 AM
Introduction
Dr. Kelli Jones

7:40 – 8:10 AM
New vaccine technologies coming for the future.
John El-Attrache, Director of Scientific Services, Ceva Animal Health, Lenexa, Kansas

8:10 – 8:40 AM
Protozoan vaccination strategies coming.
Robert Beckstead, Associate Professor, Prestage Department of Poultry Science, NC State University

8:40 – 9:10 AM
Vaccine techniques for improving application.
Chris Williams, Director Technical Service, Zoetis, Durham, North Carolina

9:10 – 9:40 AM
Questions and Answers

9:40 – 10:00 AM
Break

Moderator: Jenny Nicholds

10:00 – 10:30 AM
Future of bacterial vaccination.
Margie Lee, Professor and head of the Department of Biomedical Sciences and Pathobiology, Virginia-Maryland College of Veterinary Medicine at Virginia Tech, Blacksburg, Virginia

10:30 – 11:00 AM
Improving vaccine monitoring accuracy using diagnostic tools and techniques.
Kristen Roza Sutherland, Professional Services Veterinarian, IDEXX, Washington D.C.

11:00 – 11:30 AM
New technologies coming for vaccine application.
Jay Halliday, Product Owner of Sales and Marketing, Nova-Tech Engineering, Willmar, Minnesota

11:30 AM – 12:00 PM
Questions and Answers

12:00 – 1:30 PM
Lunch Break

Moderator: Mark Bland

1:30 – 2:00 PM
Monitoring for disease challenges & post-vaccination.
Milos Markis, Laboratory Manager, AviServe LLC, Newark, Delaware

2:00 – 2:30 PM
A field veterinarian perspective on vaccinating layer type chickens.
Ian Rubinoff, Director of Sales and Technical Service, Europe, Hyline International, West Des Moines, Iowa

2:30 – 2:50 PM
Questions and Answers

2:50 – 3:10 PM
Break

Moderator: Ivan Alvarado

3:10 – 3:40 PM
A field veterinarian perspective on vaccinating meat type chickens.
Travis Cigainero, Poultry Technical Service, Ceva Animal Health, Pittsburg, Texas

3:40 PM – 4:10 PM
Vaccination as part of the control against avian influenza in Mexico.
Susano Medina, Gerente de Operaciones, Buenaventura Grupo Pecuario, Puebla, México

4:10 – 4:30 PM
Questions and Answers

Dr. John El-Attrache

Global Director of Science and Innovation Direction, Ceva Animal Health

Technologies Advancing Future Vaccines

7:40 AM

Biography

John El-Attrache, Ph.D is a virologist with over 25 years experience working with animal viruses at a clinical, classical and molecular level. Dr. El-Attrache has garnered diverse experience working with academia, private sector groups and foreign governments. Experiences include all areas of biological product development, clinical assessment, high throughput molecular and virological diagnostic development, laboratory review and design, environmental and facility Bio-security review, personnel review and associated recommendations.

John El-Attrache is currently the Global Director of the Scientific Services Division of Ceva Animal Health in North America. Responsibilities include further investigation of licensed biological products through laboratory diagnostics, field trials and controlled studies, in addition to guidance of the Genomic Platform and Autogenous Vaccine Research and Development team for both the swine and poultry industries.

Abstract

Some of the first methodologies of human immunization were first recorded on or before the 10th century. Smallpox variolation was most notably performed in China, Africa, India and the Middle East at this time. Primary forms of variolation included either collecting pus from scabs of recently infected individuals and scratching the skin or the pulverizing of scabs into dust form and delivery through nasal insufflation. This practice was later introduced into Europe and it wasn't until the late 1700's when E. Jenner famously utilized a cowpox virus to vaccinate humans against the disease of smallpox. Advancements in vaccinology were then made by L. Pasteur and others in the understanding of virus attenuation and human immunization with live attenuated vaccines soon began.

Since the 10th century, advancements in vaccines and vaccine technologies have primarily been led by breakthroughs on the human side of medicine. However, over the past few decades any advancement discovered through human vaccine research has been applied more readily and quickly through veterinary medicine. One could easily make this argument by speaking to the number of vectored vaccines that exits in the veterinary medicine market.

The history of animal vaccines most notably began with E. Jenner's discovery. Several vaccination farms were developed for cowpox and the practice of inoculating lambs with sheep pox coincided with such events. Serendipitous events in L. Pasteur's lab led to the discovery of an attenuated strain of fowl cholera that preceded the multitude of attenuated vaccines also discovered by Pasteur and other connected researchers. The 1913 U.S. Virus Serum Toxin Act allowed for the procedures to license animal vaccines for commercial availability in the U.S. Soon thereafter, University of California Davis recorded through this act a license for fowlpox. In the 1930s, vaccines began to appear in the United States and in Europe for Newcastle Disease, Infectious Laryngotracheitis, and Infectious Bronchitis.

Since the introduction of the Virus Serum Toxin Act, many have identified the evolution of vaccines into three major generations. The first generation being the creation of whole microbial organism vaccines which

are either live-attenuated or killed (inactivated). The second generation of vaccines is comprised of vectors and/or subunits expressing proteins. The third generation of vaccines would include vaccination by the transfer of nucleic acids into the host that then express the appropriate antigen.

All three generations of vaccines will impact the future of veterinary medicine through the advancements of technology across adjuvant formulations, delivery mechanisms and recent molecular developments.

Adjuvant Formulations

Traditional adjuvant formulations for the poultry industry have not changed much over the past several decades. These formulations can be grouped in the following categories.

- Water in Oil (WO)
- Oil in Water (OW)
- Aluminum Hydroxide

These three adjuvants have been the backbone of most killed vaccines produced. A producer had one simple choice to make with these three types of adjuvants. This choice often had to be made between safety and efficacy. However, recent advancements and registration of new adjuvants have now enabled the production veterinarian to find a safe compromise between efficacy and safety. Formulations of recently registered adjuvants have replaced mineral oils and have inserted immunomodulators that not only enhance the humoral system response, but also help drive cellular immunity as well. Other ingredients such as triglycerides, acrylic polymers and cholesterol have been shown to be helpful in recent formulations for both adjuvant effect and delivery.

Delivery Formulations

Ingredients such as triglycerides, acrylic polymers and cholesterol that have been utilized in adjuvant formulations are often found to be a key component for optimal product delivery (often nucleic acids and proteins). Formulations of these key ingredients are known as Lipid Nanoparticle Platforms (LNPs). LNPs can have single to multiple ingredients and can be optimized to carry RNA, mRNA, DNA, plasmids, immunomodulators and select antigenic proteins. Recent advancements

of LNPs have allowed for much more efficient delivery (less product) and therefore, have reduced the cost of manufacturing an efficacious dose.

Molecular Advancements

Molecular advancements, within this past decade, will allow for the rapid change and improvements of vaccines within the animal health industry. Innovations in Next Generation Sequencing (NGS), DNA printing and assembly and CRISPR-cas9 could predictably provide to the industry several opportunities. They include the potential of providing a completely safe and efficacious live/killed program for long lived birds as well as quick development and regulatory approval for rapidly evolving microbial organisms.

Over the past two decades, NGS has evolved to become a more set and standard technique utilized in most research and diagnostic laboratories. This technology has allowed researchers to effectively sequence the complete genomes of bacteria, viruses and parasites and compare whole genomes rather than a specific genome segment. This has allowed for the elucidation of the multi-genomic factors associated with pathogenicity and/or virulence. Analysis of this data is still often dependent on manual curation, however, this is rapidly being overcome by intense bioinformatic analyses of recently available data and the creation of better algorithms to summarize this data. This in silico information can rapidly feed genes of interest (GOI) for insertion into established vector platforms.

DNA printing and rapid assembly developments have allowed for the creation of both DNA and RNA microorganisms in vitro without the use of any animal origin materials. Benefits from this technology enhance the ability to produce both small and large volumes of vaccine with less production issues and regulatory hurdles. In addition, rapid changes (deletions, insertions, mutations) can be accurately implemented into a microbial genome. Therefore, allowing for vector optimization, microbial genome deletion, genome optimization and genome compartmentalization.

The newest discovery of Clustered Regularly Interspaced Short Palindromic Repeats-associated protein 9 (CRISPR-cas9) technology has exploded onto the scene of vaccine development. Utilization of this

technology allows the microbial genome to be cut at a desired location. Existing microbial genes can then be edited, removed and/or new ones added. The use of CRISPR-cas9 technology is another method to allow for vector optimization and microbial genome deletion.

Future Vaccines

Undoubtedly, future vaccines within the poultry industry will be comprised of all three described generations. Recent technology improvements will enhance vaccines in each of these categories. Examples from each vaccine generation and how technology will improve these vaccines will be discussed. Furthermore, traditional first generation live attenuated vaccine development will probably decrease as new technologies enhance nucleic acid, subunit and vector vaccines.

Dr. Robert Beckstead

North Carolina State University

Histomoniasis (Blackhead Disease): Vaccination Potential and Hurtles

8:10 AM

Biography

Robert B. Beckstead received his bachelor of science from Brigham Young University, his Ph.D. from Baylor College of Medicine in Houston, Texas and performed postdoctoral work at the University of Utah School of Medicine. He was a faculty member in the Department of Poultry Science at The University of Georgia from 2007-2016 and is currently an associate professor in the Prestage Department of Poultry Science at North Carolina State University. Research in his lab focuses on molecular diagnostics and treatment strategies related to flagellated protozoal infections in poultry and the development of transgenic technology in chickens. Dr. Beckstead has mentored 60 undergraduate, 12 Masters, and 5 Ph.D. students. He currently is a member of the board of directors for the Poultry Science Association. Dr. Beckstead has been awarded the Poultry Science Association Early Achievement in Teaching Award and Novus International Inc. Teaching Award, the Richard B. Russell Excellence in Undergraduate Teaching, the First-Year Odyssey Seminar Teaching Award, and the UGA Outstanding Undergraduate Academic Advisor Award.

Abstract

Histomonas meleagridis, an anaerobic protozoan parasite of the order Trichomonadida, infects a wide range of gallinaceous birds causing histomoniasis, commonly referred to as blackhead disease (3). H. meleagridis is transmitted horizontally through the eggs of an infected cecal nematode, *Heterakis gallinarum*, or through direct contact (lateral transmission) between infected and uninfected birds. Blackhead disease is common in turkey production units, often resulting in high mortality, with clinical signs including: drooping head and wings, prolonged standing, closed eyes, ruffled feather and emaciation (5). Pathology post-mortem includes necrotic lesions in the liver and ulceration of the ceca. In chicken broiler breeder stocks and layers, blackhead disease has been suggested to cause financial loss through mortality, morbidity, increased culling, and loss of flock uniformity. Commercial layers infected with *H. meleagridis* during lay resulted in decreased egg production (6). Although the number of chickens that show signs of disease is low. Historically, blackhead disease could be treated by nitroimidazole drugs or prevented by inclusion of nitarsone (4-nitrophenylarsonic acid) in the feed. Currently there is no approved vaccine, preventative or treatment for blackhead disease.

A decrease in virulence with passage in culture has been represented in literature multiple times in an attempt to develop a vaccination for this parasite using attenuation (4, 7). Research in my laboratory has indicated that different field isolates of *H. meleagridis* are more prone to attenuation than other, with some strains attenuating after 30-50 passages and other remaining virulent after 100+ passages in culture. Attenuated strains of *H. meleagridis* do not appear to regain virulence upon passage through either the chicken or the turkey suggesting that attenuated strains used for vaccination should not cause future outbreaks of blackhead disease or disease in non-vaccinated birds in the flock (9).

Vaccination against *H. meleagridis* has shown variable results. Vaccines using a killed or inactive H. meleagridis does not provide immune protection to the turkey confirming that serum antibodies alone cannot protect birds from blackhead disease (1, 4). Early vaccination studies by Lund (7) using attenuated strains of H. meleagridis demonstrated that

high passed parasites which had lost their ability to cause disease also had limited immunizing capability when chickens or turkeys were reinfected with a pathogenic strain at 21 days post vaccination. Recent work by multiple groups has demonstrated that attenuated monocultures of *H. meleagridis* are capable of producing immunity in both chickens and turkeys (4, 6, 8). The underlying discrepancy between the recent work and Lund's studies may be due to the strain used or the methodology, with the later studies using of a booster vaccine at day 14 days and virulent challenge strain giving at 4 to 6 weeks post initial vaccination.

Several technical and financial challenges associated with the use of a live attenuated *H. meleagridis* vaccine. First, *H. meleagridis* in an anaerobic protozoan that is grown at 42ºC in a very specific media and is prone to desiccation (2). For the vaccine to work, the live parasite must be delivered to the ceca of the bird. Therefore, conditions that are inhospitable to the parasite will inactivate the vaccine. This precludes the inclusion of the vaccine/parasite in the water. Birds that are consuming feed cannot be orally infected, requiring cloacal inoculation. Thus, if a booster is required to develop full immunity, it will be impractical to cloacally inoculate turkeys and chickens in the field. Based on the literature, full immunity due to vaccination is not observed for 4 weeks post infection in the turkey leaving the young poults vulnerable during the brooding period. Research is needed to determine if an adjuvant can shorten this gap in immunity. Lastly, the nature of blackhead disease makes the financial return on the vaccine low. Blackhead disease sporadically affects the small turkey market and has limited impact on the broiler breeder and layer industry, thus to obtain a return on investment, the cost of the vaccine would need to be high.

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Dr. Christopher J Williams

Director of Technical Services, Zoetis

Vaccine Techniques for Improving Application

8:40 AM

Biography

Dr. Williams serves as Director of Technical Service for Zoetis. He was employed with Embrex, Inc. from 1988 to 2007 as Director of Global Technical Development and has continued with the company through its transformation from Pfizer to Zoetis.

In his current role, Chris supports the veterinary services team to optimize in ovo devices and vaccine applications in the United States. He has received ten patents for novel in ovo concepts and application ideas. In addition, he has donated his time as adjunct professor at NC State University and Mississippi State University. He has contributed to and authored more than 50 articles and studies in trade journals and publications on several topics, including in ovo technology, hatchery sanitation, Aspergillus, candling and clear egg removal.

Dr. Williams received Master of Science and Doctorate degrees in Avian Physiology from North Carolina State University, as well as a Bachelor of Science degree in Poultry Science. He currently resides with his wife in Apex, NC.

Abstract

There exists a wide range of techniques to apply poultry vaccines, but with all of them, achieving consistent success requires attention to details. Administration techniques are significantly driven by labor costs and time, and these factors must be considered heavily as they most negatively affect quality. Most birds raised for meat production are given vaccines through mass applications via egg injection, aerosol spray and dilution into drinking water with relatively low labor costs. Adult breeding stock are handled individually and present a relatively high cost in labor and time. Understanding environmental or equipment contaminants, embryonic/bird age, personnel and their ongoing training needs, and the principles underlying vaccination process all play significant roles in improving application irrespective of the method used. For the purposes of this review, hatchery applied vaccines will be the primary focus. Broiler vaccination trends in the US (US Broiler-Health Market Tracker, Rennier 2017) over the past 10 years have shown a marked increase in vaccine dosages and cost administered at the hatchery compared to field application. The value of hatchery applied vaccines has grown by approximately $80 M from 2007 to 2017. With these facts in mind, discussions will focus on what we can do to improve the safety and efficacy of vaccination to maximize both return on investment and bird performance.

Arguably, the most important key to improving vaccination techniques is managing the communication and culture of the application team and how team responsibilities regarding bird health are viewed. Successful vaccination in the hatchery is a coordinated live production effort involving incubation, sanitation, in ovo equipment operation and hatch pull/placement. These processes and the people who perform and manage them interact and affect bird immunocompetence. The most successful vaccination programs have key supervisory roles managing incubation, sanitation, in ovo vaccination/transfer, and hatch pull/placement. Routine veterinary and live production interaction with the hatchery personnel adds another layer of quality control. It is important to establish a working paradigm whereby communication, relationships and understanding details of production across the hatchery groups are considered routine components of quality assessment and not just during challenging or problem-solving periods. Best practices by habit

are formed by a culture of open communication, routine training and monitoring vaccine applications.

Fundamental to improving hatchery vaccination techniques is understanding and controlling microbial challenges. Excessive microbial challenges to the vaccine preparation area, the environment supporting the hatching chick, and the equipment used in applying vaccines can negatively affect vaccination. Discussions will include how routine quantitative bacterial and fungal assessments (daily, weekly, monthly) of critical control points and equipment should be conducted to provide comprehensive data to direct management decisions.

Environmental sampling from vaccine preparation areas (>150 US hatcheries) show significant challenges from air and water sources that negatively impact quality. We need to improve the construction and biosecurity of vaccine preparation areas and facilities. Sterile work surfaces areas can be created in our vaccine preparation areas for in ovo vaccines by utilizing laminar flow 'sterile' hoods. Point of use water treatment, improved room ventilation, restricted entry and inventory control can all significantly reduce microbial challenges in the vaccine preparation area.

Hatching environments and ventilation equipment that support late stage embryo development (after transfer/in ovo vaccination) play a role in controlling part of a 'background' of microflora. It is imperative that the cleaning and disinfection programs for hatchers, hatching baskets and hatcher hallway ventilation is robust on a day to day basis. Microbial monitoring should be routine and unannounced. Sanitation programs and chemicals should be tailored for specifics of the hatchery including incubation type, water quality, and seasonality.

Monitoring vaccine application equipment for sterility after usage and cleaning is also an important part of quality control. Microbial sampling of 120 separate vaccine spray cabinets (fluid pathway, 450 total samples) in 57 separate hatcheries was conducted to evaluate clean storage conditions. The fluid pathways of the vaccine spray cabinets were contaminated with bacteria more than 40% of the time during overnight storage after cleaning. Routine monitoring of vaccine pathways of in ovo vaccination equipment rarely show contamination; however,

fluid containers that support the clean in place systems are continually challenged with environmental contamination. We need to do a better job with daily, weekly, and monthly sanitation and maintenance routines of our vaccination equipment.

With the removal of antibiotics from hatchery applied vaccines there exists a greater risk to contamination during the vaccine mixing and application processes, specifically with respect to in ovo vaccination. The greatest risk is associated with two critical steps. First, the opening and removal of concentrated Marek's vaccine from the ampule after thawing, and second, during the 'hanging' process when the vaccine is connected to the injection device vaccine delivery system via 'spiking' the bag. Inadvertent contamination of the vaccine during reconstitution can happen due to transfer of contaminated thaw bath water to the vial or more commonly by over filling the vial during ampule 'rinsing'. Microbial sampling of Marek's vaccine thaw baths at 169 hatcheries during 2017/2018 showed >40% of them were contaminated with bacteria. It is recommended to add chlorine to the thaw bath (15.0 ml of 5-6% bleach per gallon distilled water resulting in ~200 ppm Cl) to prevent contamination of the thaw bath. Discussions and data to support this recommendation will be presented. It is recommended to thoroughly dry the ampules before opening. Similarly, when the bag of mixed vaccine is attached to the dispensing (pump) system on the machine, the bag and connection port should be dried thoroughly before IV spiking of the bag. A recent large-scale evaluation of MD vaccine vial rinsing efficacy shows that only a nominal increase in viable cells are salvaged during ampule and cap rinsing. Rinsing MD vaccine ampules with fresh diluent after withdrawing concentrated vaccine results in ~ 1.5 % 'salvage' of chick embryo fibroblast cells from the ampule and the caps. Perhaps we should consider the risks of bacterial contamination due to this procedure keeping in mind the hundreds of times this task needs to be performed without error and how negative the impact of worse-case scenario can be.

Finally, we must consider the developmental stage of the embryo and hatched chick at the time of vaccination to optimize and improve the vaccination response. Monitoring hatch progression via pre-hatch observation (12-15 hours prior to pull) establishes that incubation has produced a 'normal' population. Under-developed chicks ('dragging'

hatch) pose significant problems to subsequent spray vaccination as not only are the chicks in a weakened state, but their immune status is compromised. Similarly, embryonic staging prior to in ovo vaccination can reveal challenges to the developing embryo with respect to incubation quality. Discussions will be made as to optimizing the timing of in ovo application and quality of spray vaccination.

Dr. Margie D. Lee

Virginia-Maryland
College of
Veterinary Medicine

The Future of Bacterial Vaccines

10:00 AM

Emeritus Professor
Poultry Diagnostic and Research Center
College of Veterinary Medicine
The University of Georgia, USA

Professor and Head
Biomedical Sciences and Pathobiology
VA. MD. College of Veterinary Medicine
Virginia Tech, USA

Education:
Postdoctoral, mentored by Dr. Roy Curtiss, III, Biology Dept., Washington University, 1990-1992
Ph.D., Medical Microbiology, The University of Georgia, 1990.
M.S., Medical Microbiology, The University of Georgia, 1988.
D.V.M., Va. Md. Regional College of Veterinary Medicine, 1986.
B.S., Biology (major), Chemistry (minor), Virginia Tech, 1982.

Career Accomplishments:
Research focus on epidemiology of meat and poultry-borne food safety pathogens, microbial pathogenesis, and ecology of poultry intestinal microbiome resulting in 77 peer-reviewed publications as well as chapters in the Merck Veterinary Manual and the Laboratory Manual for Isolation, Identification and Characterization of Avian Pathogens. In addition to research, she teaches microbiology to veterinary students, graduate

and undergraduate students and veterinary residents. She is formerly the coordinator of Graduate Affairs for the UGA College of Veterinary Medicine and graduate coordinator for the Masters of Avian Medicine program.

Abstract

Despite the fact that they are among the most studied organisms known to man, bacterial pathogens have resisted many disease control measures. Antimicrobials have offered a short-term solution but from an evolutionary perspective, the pathogen tends to quickly develop resistance. Epidemiological studies have shown that we can reduce the incidence of many bacterial diseases by improving sanitation and reducing transmission. Vaccines have played a significant role in reducing transmission and infection rates thereby aiding in disease resistance. However, throughout the millennia, bacteria evolved to be able to counteract the immune system. Our uninformed, simplified attempts to design and apply vaccines have not significantly reduced the impact of some of the most relevant bacterial pathogens.

E. coli is one of the most costly bacterial pathogens in poultry and Salmonella is an economic challenge to control because of its food safety importance. These gram-negative bacteria have evolved a variety of surface molecules that exhibit a large antigenic diversity which enables them to persist as commensals in vertebrate hosts. The terminal O-antigen of the lipopolysaccharide is one of the most antigenic molecules possessed by the bacteria, but it is also one of the most variable. Similarly, flagella make up a large proportion of the surface molecules present on these bacteria, but it too is very variable. The variability of these molecules is used to as a serotyping system for isolate grouping and epidemiological studies which illustrates the vast array of antigenic types present among E. coli and Salmonella. However they do possess antigenically conserved molecules within the core LPS which can be exploited to design bacterins with cross-protection. Similarly, there is serological conservation among some flagellar antigen types within serotypes of Salmonella, which offers an opportunity to utilize bacterins to control dissimilar O-serogroups.

Current literature regarding cross-protection in Salmonella and E.

coli has shown that conserved surface molecules can serve as protective antigens. Methods to remove the serologically variable O-antigen has produced evidence of cross protection and this approach is likely to be effective for other bacterial pathogens as well. Old-school approaches to develop serological tools need to be resuscitated to help diagnostic labs characterize clinical isolates within a production system in order to describe the serological diversity of the pathogens. Strain typing will also help to define strain distribution to enable better decisions regarding vaccine development for outbreaks or opportunistic infections. Ultimately, diagnostic laboratories have to be more effective in informing veterinarians of whether disease isolates are members of an outbreak strain. In addition, the serological diversity of disease isolates needs revealing in order for effective vaccine formulations to be available. Better vaccines result from an informed view of the status of disease distribution, its main routes of transmission and the serological diversity of the pathogens causing the disease.

Dr. Kristen Roza-Sutherland

IDEXX Livestock

Improving Vaccine Monitoring Accuracy Using Diagnostic Tools & Techniques

10:30 AM

Biography

Dr. Kristen Roza- Sutherland attended Veterinary school at North Carolina State University, where she also completed her undergraduate degrees in Poultry Science (BS) and Animal Science (BS).

She began working for IDEXX in 2011, specializing in Poultry Veterinary Professional Services. During this time, she has travelled extensively to the labs around the US and Canada and has attended and presented at educational events across the country. She also works with Bovine, Swine and Equine diagnostic testing in her current position.

Her previous positions in the veterinary field include working with the Population Health and Pathobiology group in Poultry Medicine at NCSU CVM and as a field veterinarian at Rood and Riddle Equine Hospital in Lexington, KY. Her professional interests include diagnostics and disease epidemiology in commercial and backyard populations.

Abstract

As animal husbandry technology and practices improve, we are presented with many new options for monitoring poultry flocks for disease and vaccine take. It is important that the practitioner and animal managers are able to gain useful and actionable information from the testing that is done, while maintaining rearing costs and preventing illness. Health monitoring programs should include a combination of diagnostic testing results, flock performance metrics and the results of regular examination of the flock and any natural mortality. Rapid and accurate disease detection is imperative when a field infection is suspected.

Currently there are many options for diagnostic testing, which can be generally divided into Screening and Confirmatory testing methods. Screening methods, such as Ab ELISA, AGID, and HI are typically lower in cost, quick and robust tests. Confirmatory testing, such as PCR, Culture, Virus Neutralization and Histopathology may have higher specificity, but often have a longer time to results and have higher cost of testing per sample. When determining the best test to be used in the design of a testing program, many factors should be considered, including test cost, time to results, and accuracy of the test.

When a new testing modality is considered, it is important to consider the following questions to determine the suitability of the test to the flock health program;

- Information needed (vaccine take vs field infection presence)
- What is the value of the information being generated
- Does this enhance or replace current testing?
- Can I make better decisions with the information this test produces?

Often costs are a deciding factor in these decisions. When managing the costs of a diagnostic program it can be approached as a management strategy, in which the information being generated is used to direct health activities in future flocks, or as a defensive strategy, in which diseases with the highest rick of mortality or morbidity are prioritized for monitoring. Any regulatory or required testing must always be prioritized when making decisions on diagnostics spending.

One strategy for providing cost savings without potentially sacrificing animal health in diagnostics is to better utilize current or fundamental testing methods such as ELISA testing. This depends on appropriate number of samples being tested to assure result accuracy. Using the statistics generated from ELISA Testing software, such as the flock Mean, Standard deviation (SD), and Coefficient of Variation (CV) can provide additional information on flock status and provide earlier detection of changes in serologic profile that can be indicative of early changes in health. One example of this is shared in the slides presented that outlines the Standard Deviation (SD) of a flock raising prior to increase in titers or clinical disease signs. Tracking serologic results for like flocks over time is the most detailed way to utilize serology results and can provide additional value for testing already being done. Addition of newer technologies for confirmation of these early changes is an excellent strategy to maintain best possible animal health through diagnostic testing.

Mr. Jay Halliday

Nova-Tech
Engineering

Technological Advances in the Applications of Poultry Medicine

11:00 AM

Biography

Jay has an M.B.A. in international business and is fluent in Spanish and Portuguese. He has been working in the animal health industry for 12 years as the regional business manager for Latin America at Nova-Tech Engineering. His primary focus is on animal welfare, market development, and leadership development. He lives in Minnesota with his wife, son (3), daughter (1), and British red lab (8).

Abstract

Although numerous forms of vaccination applications currently exist, the industry has not seen much innovation in this sector for decades. Standard subcutaneous or intramuscular systems, respiratory sprays, and eye drops are used throughout the lifecycle of the bird, from the hatchery to the grow-out facility. Specifically, in the hatchery, we commonly find spray gels and in-ovo vaccinations. All of these systems have purpose, and many are very efficient means of applying vaccinations, yet not without downsides. For instance, spray gels are reliant on the birds' ability to peck and preen, thereby ingesting the vaccine, and hoping the bird consumed the adequate dose to be effective. The future of vaccine application will

no longer rely on hope as a strategy; rather we will use technology to apply a specific quantity of vaccine to a specific location on the bird. This prescriptive treatment will greatly increase the effectiveness of vaccine application and therefore maximize the potential of its efficacy within each individual bird, maximizing protection for the entire flock.

In this presentation, we will take a brief review of current poultry vaccination tools, highlighting the major pro's and con's. Then, for its first appearance on the public scene, you will be introduced to the new Nutrient Delivery System. This automated technology has the ability to apply vaccines, probiotics, prebiotics, etc. directly into the crop of poultry at day of age within the hatchery. It allows for full control of dosage, giving poultry veterinarians confidence in the ability to populate a targeted gut bacteria in earliest stages of a bird's lifecycle, and therefore establish the best means for maximizing the genetic potential of the bird.

Dr. Milos Markis

Vice President,
AviServe LLC

Monitoring of Disease Challenges Post-Vaccination

1:30 PM

Biography

Dr. Markis received his Bachelor's degree in Biological Sciences in 2008 and Master's degree in Animal Science in 2010 from the University of Delaware. His thesis research was on molecular biology of Marek's disease virus. Dr. Markis received his Ph.D. degree from the University of Delaware in 2014 while conducting his research at AviServe LLC investigating the roles of reoviruses and astroviruses in viral enteritis/enteropathy in chickens. Following graduation, Dr. Markis was employed by AviServe LLC, where he still conducts research. Majority of his research involves studying variant reoviruses and infectious bursal disease viruses of economic significance to the poultry industry.

Abstract

AviServe LLC is a privately owned, independent diagnostic and research company located in Delaware. AviServe was established to serve the needs of the poultry and allied industries for recognition and control of conventional and complex diseases. The vast experience, willingness, and freedom to work with all poultry and allied industries has given us a large historic and geographic perspective on avian diseases and associated

pathogens. The majority of contract research conducted at AviServe is on emerging and re-emerging diseases and disease complexes often associated with antigenic and pathotype variants. Antigenic variants of reovirus, infectious bursal disease virus, and infectious bronchitis virus have comprised much of our research efforts for the past several years. We have isolated and characterized multiple antigenic variants that have been used by multiple poultry companies to produce autogenous vaccines that supplement existing vaccination programs.

"Breaks" of diseases that are otherwise controllable through vaccination are often the first indications that a vaccination program is not working properly. These "breaks" may be results of poor vaccine administration, use of inadequate vaccines, or occasionally due to emergence of antigenic variants that are no longer covered by commercial vaccines. Vaccination programs can be evaluated, modified, and monitored utilizing several approaches that will be discussed.

The first step in evaluation of a disease "break" is to isolate and identify the pathogen evolved. Viral pathogens circulating in commercial poultry that result in clinical disease are isolated in our laboratory from general laboratory submission of field samples, placement of sentinel chickens on commercial farms, and controlled surveys. Virus isolations are most successful when samples are collected at the beginning or peak of infection, when virus is present at highest concentration, however, this can be difficult in cases where the infection precedes development of overt disease or clinical signs, such as reovirus infections.

Laboratory submissions of field samples are useful for isolation of pathogens involved in particular disease "break", but lab submissions are not ideal for every scenario. Reovirus isolations from synovial fluids and digital flexor tendons collected from affected chicken legs are routinely done in our laboratory. Infectious bronchitis virus (IBV) can be isolated from tracheal swabbings of afflicted commercial chickens, but sentinel chickens are often necessary to isolate emerging antigenic variants without the contamination with vaccine viruses that may persist in commercial flocks.

Surveys can be useful for virus isolations when a poultry company is interested in evaluating performance, disease prevalence, and virus

challenges across an entire complex or company. In case of infectious bursal disease (IBD), a bursal survey can be conducted to monitor the strength of vaccination program and identify potential antigenic variant viruses. Bursal surveys are completed by many poultry companies once a year on about thirty farms per complex.

Antigenic and pathogenic characterization of virus isolates is necessary for a reliable modification of any vaccination program. Infectious bursal disease virus (IBDV) and reovirus variants are often utilized by poultry and vaccine companies to produce autogenous vaccines, which supplement breeder vaccination programs by expanding antigenic diversity and providing progeny with appropriate maternal immunity. In our laboratory, isolated reoviruses are pathotyped through inoculation of day-old chicks via oral/intratracheal route and foot pads. Pathogenic reovirus isolates are serotyped using a microneutralization assay and a panel of 20-30 polyclonal antisera developed in our laboratory. IBDV isolates are antigenically characterized (serotyped) via virus neutralization assay with Delaware Variant E antiserum in embryonated SPF chicken eggs. Delaware Variant E antiserum is utilized for virus neuralization because this virus serotype is present in most commercial vaccines in the USA. Additionally, antigenic diversity of IBDV isolates will be evaluated via extended virus neutralization assays utilizing multiple polyclonal antisera, and correlations made to genomic sequence. IBV isolates can be easily genotyped and potential variants can be identified this way. IBV isolates can be used to challenge chickens immunized with different commercial IBV vaccines and vaccine combinations to identify the most suitable vaccination program. Often, a commercial vaccine against a particular IBV serotype is available, however, new IBV vaccines may need to be developed when a variant IBV emerges. Occasionally, viruses isolated from commercial birds are not antigenically different from already utilized vaccines, and in those cases evaluation of vaccine administration, vaccine dose, and age of vaccination is recommended.

When emerging antigenic variants of IBDV and reovirus are discovered, autogenous vaccines containing these variants can be produced to expand antigenic diversity and coverage of the vaccination program. Vaccination programs that were altered can be re-evaluated in several ways, but it is always important to allow several broiler flocks to cycle through production prior to sample collection. Bursal surveys and

leg surveys can be repeated to evaluate presence or absence of infectious bursal disease and viral arthritis/tenosynovitis, respectively. Any virus isolated from the survey samples should be serotyped to determine if additional variants are circulating in the field, which can be added to subsequent serials of autogenous vaccine. Also, progeny challenge can be done for IBDV evaluation utilizing chicks from several vaccinated breeder flocks and evaluate protection against several contemporary IBDV isolates.

Efficacious vaccination programs are economically important to poultry companies. Inadequate administration of vaccines or emergence of variant viruses not covered by vaccination programs can have a devastating impact on performance. At AviServe, much time is spent on isolation of emerging viruses and their further characterization. Often, new characterization assays and challenge models have to be developed to fully assess emerging viruses. The findings from these evaluations aid in selection of the appropriate commercial vaccines or selection of variant isolates to be used in autogenous vaccine production.

Dr. Ian Rubinoff

Director of Global Technical Services, Hy-Line International

A Field Veterinarian Perspective on Vaccinating Layer Type Chickens

2:00 PM

Biography

Dr. Ian Rubinoff is the Director of Global Technical Services with Hy-Line International. Since 2009 he has worked on disease prevention, lighting, management, nutrition, export, and biosecurity issues with global distributers, and speaks at seminars for a variety of organizations. Dr. Rubinoff works at all genetic levels from the research farms down to the commercial customers with Hy-Line to help internal flocks and customers achieve the best performance.

Dr. Rubinoff is originally from Rhode Island, and attended the University of Rhode Island for undergrad. He earned his D.V.M and M.P.H degrees from the University of Minnesota, while working with the layer industry and researching avian influenza. Dr. Rubinoff is a Diplomate of the American College of Poultry Veterinarians.

Abstract

Introduction

Vaccination of laying hens is critical to help protect flocks against disease challenges. While laying hens have always been longer lived

than broiler breeders or turkeys, recent changes in the egg industry have cast a new light on the need for vaccination protocols. Single cycle production, cage free systems, and antibiotic free marketing has all disrupted the egg industry and fundamentally changed the way we need to look at protecting hens. With the exception of vectored vaccines, most vaccination technology is decades old and potentially in need of an update.

Disease families

Viral vaccines have the widest effective range of products available, with vectored, live, and killed products all proving to be efficacious. While there are a variety of products available, several challenges still exist for viral vaccines.

•Infectious bronchitis still causes problematic production drops and egg quality issues if flocks are not properly vaccinated or not vaccinated with the correct strains. False layer syndrome has emerged in several regions where it had not appeared before.
•Virulent strains Newcastle disease are reemerging in many parts of the world. Current vaccination programs have helped to limit mortality in flocks, however the impact on production can be dramatic.
•Highly pathogenic avian influenza is very difficult to vaccinate against because of the unique nature of each strain and serotype if the country does not support eradication efforts.

Bacterial vaccines are critical to the future of egg production, especially in the absence of antimicrobial usage in many countries. The challenge to create effective bacterial vaccines without resorting to utilizing inactivated vaccine methods is also becoming more important. Bacterial infections in chickens represent both animal health and food safety issues.

•Salmonella is always an important pathogen for human and bird health
•E. coli has been causing greater levels of mortality and is becoming resistant to treatment and potentially vaccines as well
•Enterococcus/staphylococcus especially for chicks coming from antibiotic free hatcheries
•Campylobacter and food safety for egg layers has not been a strong current issue but may become more important in the future

•Coryza/cholera and the need for homologous strains makes it difficult to match commercial vaccines to on farm challenges
•Clostridium and the impact on flocks in the form of necrotic enteritis and focal duodenal necrosis make these bacteria a prime target for future vaccine innovation
•Mycoplasma for multi-age and any at risk flock ideally should be done on a bird by bird basis to ensure proper titers
Protozoal vaccines along with bacterial vaccines have become more important as the utilization of chemical and ionophore coccidiostats and medications are becoming harder to use in some countries.
•Coccidial vaccines can be efficacious but are expensive and not always effective due to management difficulties
•Histomonas has no current vaccine and also no legal treatments in some areas

Methods of application

The traditional methods of vaccine application are injection, eye drop, wing web, spray, and water. Spray and water vaccines are very useful because of the mass application opportunities with the detriment of the potential for uneven exposure to the vaccinated disease. Injection, wing web, and eye drop vaccines are very useful for delivering an exact titer to each bird with the detriment being the stress of having to handle each bird in the house. As labor becomes more difficult to find for agriculture, utilizing more mass application vaccines will allow for greater levels of protection.

Dr. Travis Cigainero

Ceva Animal Health

A Field Veterinarian's perspective on Vaccinating Meat Type Chickens

3:10 PM

Biography

Obtained veterinary degree from the LSU School of Veterinary Medicine. After practicing in a mixed animal practice for a year in Tennessee, was accepted to Auburn University College of Veterinary Medicine for a rotating internship position in small animal medicine and surgery. Upon completing responsibilities at Auburn, was employed as an associate in a mixed animal practice in Pittsburg, TX. After six years of practice, Travis accepted a position with Pilgrim's Pride Poultry with the intent of learning the integrated poultry business from the "inside out" combining hands on daily management of poultry farms with a diverse understanding of veterinary medicine. This ultimately lead to practicing as a production veterinarian for Pilgrim's Pride Poultry (meat type and leghorns) for 24 years with production and processing responsibilities in both the US and Mexico. In 2014, joined Ceva Animal Health as a technical service veterinarian.

Abstract

Many significant changes have occurred over the past ten to twenty years with regard to producing meat type chickens. Contributing factors include changes in husbandry, consolidation, nutrition, and vaccination strategies to name a few. The objective of this presentation will be to share some of the more relevant issues surrounding successful vaccination of meat type chickens. Vaccination is not a standalone process for efficient production as many factors feed into the final results. It is not uncommon for different companies or poultry veterinarians to have somewhat different opinions on how to control similar problems. A brief evolution of some disease challenges, vaccines, vaccine administration, and production practices will be addressed.

Dr. Susano Medina Jaramillo

National Autonomous University of Mexico

Review of the Aspects of Avian Ifluenza in Mexico and its Control Through Vaccination

3:40 PM

Biography

Avian pathologist with more than 40 years of professional experience in poultry. His experience includes commercial layers, grandparents, broiler breeders, incubation, broiler production, training projects, instrumentation of poultry health and biosecurity programs, business, marketing, and strategic planning.

Abstract

BRIEF DESCRIPTION OF TYPE A INFLUENZA VIRUSES:

Type A avian influenza viruses are members of the Ortomyxoviridae family, these viruses are enveloped and pleomorphic, with a tendency to be spherical. The virus contains in its interior 8 segments of ribonucleic acid (RNA) joined together by the nucleoprotein (NP); this condition of having a segmented genome is fundamental for reassortment, mutations and insertions, between viruses in the same host or with different avian hosts. They are negative-sense single-stranded RNA virus. On their surface are two glycoproteins: a hemagglutinin (HA or H) and a neuraminidase (NA or N). The protein HA has three special functions; it bears the ligand that will bind to the cell to start the infection process, binding the virus

with the sialic acid on the surface of cell to be infected, it also contains the fusion site, which favors the fusion of the viral envelope with the endosome membrane, that allows the delivery of the nucleic acids into the cytoplasm, which are directed to the nucleus to carry out the process of viral replication and translation. In addition, HA is the most important antigenic protein that elicits an important response in the host against the virus. The neuraminidase is a protein that has the function of breaking the sialic acid of the infected cell, allowing the release of viral progeny particles to infect other cells. The virus also has two membrane proteins (M2 and M1).

16 different subtypes of HA and 9 different NAs have been identified in birds.

BACKGROUND OF THE OUTBREAKS of AVIAN INFLUENZA IN MEXICO:

In 1994, the National Council on Animal Health (Consejo Nacional de Salud Animal, CONASA) created a committee to investigate the emergence of respiratory problems of undetermined etiology, and that were difficult to control in commercial flocks. Doctors, Juan Gay and Ricardo Cuetos, after a presumptive diagnosis of Avian Influenza, elaborated a type A antiserum and confirmed the infection. They also carried out pathogenicity tests. They notified their results to the Mexican animal health authorities and they also transferred their antisera and viral isolates for their characterization. Through sequence analysis it was indicated the presence of a low pathogenicity avian influenza virus subtype H5N2 (IABPH5N2). At that time, proposals to develop a vaccine and carry out a massive vaccination program were raised, but these proposals were not approved. In December of the same year, the virus mutated to highly pathogenic avian influenza virus (VIAAPH5N2) and it was officially reported to the authorities by the poultry producers of the states of Querétaro and Puebla. Rigorous measures were implemented for the management of infected flocks and for the adequate disposal of carcasses and slaughtered birds, for the management of poultry products and by-products, mobilization controls, establishment of quarantines, depopulation programs, disinfection of facilities and equipment. A vaccination program was focused on the birds at risk, in the affected areas, with a homologous inactivated vaccine, elaborated with the strain: A/ck/Puebla/2859-474/1995-H5N2. In June of 1995, the eradication of highly pathogenic influenza was accomplished. With this important

milestone, eradication of low pathogenic avian influenza would have been possible. However, this did not happen, because with the drafting of the Official Mexican Code (Norma Oficial Mexicana, NOM), for the prevention, control and eradication of Avian Influenza, H5N2 (NOM-044-ZOO-1965) only eradication was contemplated, and massive monitoring and vaccination programs were not implemented, that were especially needed in the areas of poultry population at high risk. Furthermore, an evaluation focused to determine the size of the affectation was not carried out. The outcome to date, more than 24 years after the emergence of low pathogenic avian influenza, is that the disease is still endemic in many parts of the country where vaccination continues in place.

In February of 2006, academic personnel from the National Institute of Forestry Research (Instituto Nacional de Investigaciones Forestales) and the College of Veterinary Medicine and Animal Sciences (Facultad de Medicina Veterinaria y Zootecnia) of the National Autonomous University of Mexico (UNAM) detected an low pathogenic avian influenza virus subtype H7N3 (VIABPH7N3) in wetlands of Valle de Bravo, in the State of Mexico. That was the first report in our country of an official isolation, by a natural route from an Anas cyanoptera duck (cinnamon tail). However, no monitoring or follow-up program was established after this notification (in 1980, Hinshaw et al., reported the perpetuation of avian influenza viruses in free-living aquatic birds).

On June 13th, of 2012, poultry producers from the State of Jalisco made the first official report of three outbreaks of highly pathogenic influenza subtype H7N3 (IAAPH7N3), in commercial layers (Tepatitlán and Acatic), the authorities confirmed the isolations, and a plan for eradication was activated. By July 24th, 358 farms had been monitored in the area, with an approximate population of 17 million birds (60% commercial layers, 24.6% broilers, 8.5% backyard birds and the rest broiler breeders). Due to the magnitude of the problem, and the level of damage of this important poultry producing area the animal health authorities decided to work on the preparation of an inactivated vaccine, prepared with the viral strain isolated in year 2006 (A/Duck/2817/2006H7N3), the vaccine was ready to initiate a vaccination program under official control on July 27th, only in the affected areas. However, from June to August of 2012, 44 infected farms were identified in 8 municipalities of Jalisco and in January of 2013, reports of outbreaks appeared in the State of Aguascalientes

and in more flocks of commercial layers resulted infected in Jalisco. In February, outbreaks are reported in the State of Guanajuato in broiler breeder hens, commercial layers, and with the first reports in broiler chickens. Depopulation efforts are carried out in the affected farms with proper disposition of carcasses, cleaning and disinfection of facilities and equipment, a vaccination program was implemented in the area, as well as control of the mobilizations, quarantines and depopulation of the affected farms.

In March, an outbreak is reported in the State of Puebla, in commercial layers, molted hens, and the corresponding depopulation program was established.

Based on official data, only in the year 2012, 24 million of commercial birds were lost or sacrificed (this figure corresponds to 8% of the Mexican national production).

Today we have the experience of 24 years in the use of vaccines against avian influenza subtype H5N2. Our country rapidly accomplished the control and eradication of the highly pathogenic virus, however, the infection with the low pathogenicity virus remains established. From 2012 to date, we are being challenged with the control of viruses of the H7N3 subtype of high pathogenicity.

WHAT IS HAPPENING, WITH THE SUBTYPE H5N2 OF LOW PATHOGENICITY?

The initial strategy to control the by low pathogenic avian influenza virus subtype H5N2 (VIABPH5N2) was based on the use of a killed virus emulsion vaccine, using the official seed (PRONABIVE 94), whose technical guidelines were specified in terms of its antigenic content. Few years later, a vector vaccine was introduced with a fowlpox vector expressing the hemagglutinin of a H5 virus. Years later, vector vaccines with Newcastle disease and Marek's disease viruses as vectors were available.

Great efforts have been made, unfortunately, these have been based on trial and error in the search of better forms of control. In 2000, Dr. David Suarez, documented that the antigenic drift suffered in the field viruses was significant, measured both by cross HI testing HI against the

official antigen, and also by providing evidence of a lack of protection based in the reduction of viral shedding. After this report, the Mexican government authorized two new official seeds, a virus isolated in year 2006, and a more recent virus from 2016, the attempts with both vaccine seeds have been insufficient to maintain the required antigenic similarity for optimal protection. We know that to elaborate a vaccine of good quality, it is extremely important the antigenic homology with the subtype of the challenge, as well as the antigen concentration, the quality of the emulsion and the type of adjuvant that is being used.

STATUS OF THE CONTROL OF HIGHLY PATHOGENIC AVIAN INFLUENZA SUBTYPE H7N3:

The efforts to control highly pathogenic avian influenza subtype H7N3 (VIAAPH7N3) (H7N3/chicken/Jalisco/CPA/12283-12/ 2012) started with the development of an inactivated vaccine containing a seed from the field isolate of year 2006 that was demonstrated to be of low pathogenicity, obtained from a cinnamon teal, which initially showed to induce protection, using vaccine doses of 102 and 256 hemagglutinating units per bird (HAU), but similary to what happened with low pathogenic avian influenza H5N2 (VIABPH5N2), antigenic drift was documented after one year of using the vaccine. According to this scenario, a working group was formed with the participation of animal health officials, producers and the pharmaceutical industry, to certify whether the authorized vaccine was protective or not, a fact that was completely demonstrated and led to the approval of new vaccines, and a new official seed, of which there are two forms; one of year 2015 and other of year 2016. These vaccines were produced with a dose per bird of 512 HAU. Despite the use of these vaccines and the new vaccination programs, clinical problems are still manifest in both broilers and commercial layers. However, it must be acknowledged that some places in the country have showed much better results. Recently, at the end of last year, a new vector vaccine with fowlpox was introduced, this vaccine expresses the hemagglutinin of the H7 virus, with the expectation that the program with a prime stimulation would achieve the benefits of protection and the reduction of the viral excretion, which are sought with vaccination.

The lessons acquired during these years of vaccine use seem to show that dual programs using both vaccines developed by reverse genetics or vector (recombinant) vaccines, and a program of emulsion

killed vaccines including a recent homologous seed, with high antigenic mass, perform much better in controlling problems in the field. The aspects to be considered are the homology (a minimum of 97%) and to guarantee the immunogenicity (that elicits high levels of antibodies) against the field strain. Inactivated vaccines basically provide humoral immunity. On the other hand, reverse genetics vaccines and vector vaccines are responsible for generating specific cellular immunity and nonspecific natural immunity, which contributes to the control and protection against mortality, morbidity, and also significantly reducing the replication and excretion of the virus.

Regarding the recombinant H7 vaccine with fowlpox as vector, it can be applied from day one of age with no neutralization by maternal antibodies and it is compatible to be applied, together with Marek's disease vaccine, in both broilers and commercial pullet. Its effectiveness in conjunction with the use of emulsion vaccines is currently being evaluated in the field. Currently, H7 vaccines with a Newcastle disease virus as vector and reverse genetics vaccines are the most widely used, accompanied by different schedules using inactivated vaccines with high homology.

All the work done through vaccination must be supported by a joint program between the poultry industry and the authorities, with clear goals, clear objectives and great sanitary discipline. This program must include an excellent control of the movement of live birds, products and poultry by-products, it should also consider internal and external mobilizations, with a precise situational analysis, timely outbreak reporting, programs of indemnity, compensation, and support to companies affected by outbreaks. Only with these conditions better results can be obtained. Currently, we are still far from reaching a definitive solution.

Vaccination is only a mean, by itself, it will not completely solve the problems that avian influenza represents to the Mexican poultry industry.

Translated from Spanish into English by Dr. Alejandro Banda

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