

# zootechnica

INTERNATIONAL

**Poultry house environmental control during cold weather**

**Role of diet on odour emissions from meat chickens**

**The challenges and future of diagnostics in poultry medicine**



"Il Fiore delle Dolomiti" Organic Layer Farms (Italy) built and equipped by SKA

**12**  
**2017**



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IMPIANTI AVICOLI  
POULTRY EQUIPMENT



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# EDITORIAL



The world's poultry industry, similar to other sectors, continues to face tough market situations.

During more than half a century of history, our industry, for those who have had the personal privilege of being able to converse about it, there has always been talk of crisis.

Yet in those same fifty years what profound transformations have occurred!

Thanks to genetics, and to the evolution of methodologies and techniques in breeding and processing, growth in the industry has reached a high level of specialization.

The quality of the products has improved enormously and has succeeded in meeting the growing demands of the consumer. Thanks to this evolution the poultry industry of today can offer the consumer a wide range of products at affordable prices.

There are however some matters for consideration by those who will oversee the future of the entire poultry chain.

For those involved in the poultry business their reflections are quite different because for a long time, we hear of less profitability and greater competitiveness.

What are the possible deductions? The rapid evolution of the market has led to new rules, which many operators have not yet been able to adapt to. Question remains, why is it that so many companies are in critical condition while it is equally true that there are others that are growing.

Interpreting events is not always easy to understand.

The answer probably lies not only in embracing the great synergies occurring in the industry but equally in knowing how to keep up with the pace of technological evolution. Maybe there is something obvious that escapes us.

Perhaps we are dealing with a new approach to reality that goes beyond the rules of profit only.

A handwritten signature in black ink, appearing to read "Peter Hauke". The signature is fluid and cursive, with a large, stylized initial 'P' on the left.



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## THE MOST INNOVATIVE RANGE FOR POULTRY FEEDING :

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- Breeders      • Layers      • Ducks
- Cocks      • Turkeys



# MODULA

## The turkey feed pan



Model for adult birds without cover



Model with cover



First days chicks

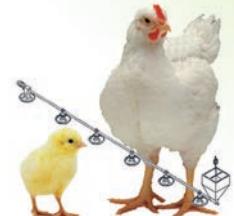


Young birds

- Sturdy and easy to manage.
- Designed for both one day old chicks and heavy male turkeys.
- No chick inside the pan.
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- High quality product at competitive prices.

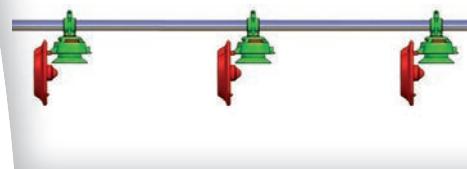
# PRATIKA

## The broiler feed pan



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## Poultry health course 9 to 20 April, 2018

*Poultry rearing for meat and eggs is now a global activity, which continues to adapt to the changing national and international economic and political climate. Infectious agents and other causes of disease also continue to change and adapt and it is important for those involved in the industry to keep abreast of the latest applied and pure scientific developments, which can affect the industry.*

The Poultry Health Course was established at the Houghton Poultry Research Station in the 1970s and has been run every year since then. In 2015 the course of 55 lectures was delivered by 30 international experts in their fields of pathology, poultry rearing, nutrition, pathology, parasitology, virology and bacteriology coupled with applied subjects such as vaccination, post-mortem examination and laboratory diagnosis.

In 2015, The University of Nottingham partnered with the Pirbright Institute to also develop the Poultry Health Course as a completely online course to be run in parallel with the residential course involving the same set of lectures and lecturers. The online course is available from October to June.

This one from 9 to 20 of April 2018 is a residential course for veterinary surgeons, technical staff and managers from the poultry industry who wish to learn more about the nature, diagnosis and control of infectious diseases of poultry. The course is also suitable for non-specialist graduates who wish to gain a thorough knowledge of the poultry sector while working towards the postgraduate awards in Agrifood.



**Week 1 (9 – 13 April 2018), based near the Pirbright Institute,** contains lectures based around cutting edge scientific aspects of poultry health including anatomy, immunity, genetics and infectious disease, delivered by international experts from the UK, Europe and the US.

**Week 2 (16 – 20 April 2018)** is based at the University of Nottingham's School of Veterinary Medicine and Science (SVMS), Sutton Bonington Campus and comprises a series of lectures and practical activity on the more applied aspects of poultry health including the turkey, duck and game bird industries, vaccination and use of antibiotics, problems of the broiler and layer industries, lameness, problems in the field and the art of field investigation.

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For course registration, enquiries, and further information on course organization and content  
[www.poultryhealthcourse.com](http://www.poultryhealthcourse.com)

# The 2018 International Production & Processing Expo is coming!



This year the IPPE show will bring together more than 1,200 exhibitors and 30,000 visitors in Atlanta, Ga. USA from Jan. 30 to Feb. 1, 2018. The trade show focuses on **Innovation** - bringing together buyers and sellers of the latest technology of products and services to make companies' business successful, **Education** - learning from the experts in free - and fee-based world-class programs beginning Mon., Jan. 29, on topics that cross industry interests, **Global Reach** - attracting more than 8,000 International visitors from 129 countries, and **Network-**

ing - meeting new and rekindling old relationships with leaders across the industries.

Made up of the three integrated tradeshows - International Poultry Expo, International Feed Expo and International Meat Expo - the IPPE is the world's largest annual feed, meat and poultry trade show.

The event is sponsored by the U.S. Poultry & Egg Association (USPOULTRY), the American Feed Industry Association (AFIA) and the North American Meat Institute (NAMI).

#### 2018 Dates:

Jan. 30 - Feb. 1, 2018

#### Show Times:

Tues., 10 A.M. - 5 P.M.

Wed., 9 A.M. - 5 P.M.

Thur., 9 A.M. - 3 P.M.

**Venue:** Georgia World Congress Center  
285 Andrew Young International Blvd NW  
Atlanta, Georgia USA

#### U.S. Poultry & Egg Association

1530 Cooledge Road Tucker, GA USA

**Phone:** 770.493.9401 - **Fax:** 770.493.9257

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# IFIF and the FAO continue to strengthen collaboration on critical feed issues

The International Feed Industry Federation (IFIF) and the Food and Agriculture Organization of the United Nations (FAO) held their 16<sup>th</sup> annual meeting at FAO Headquarters on October 2017 to further strengthen their collaboration on critical issues to ensure safe, nutritious and sustainable feed and food.



The meeting was officially opened by **Dr. Berhe Tekola**, Director of the FAO Animal Production & Health Division and Joel G. Newman, IFIF Chairman, who welcomed delegates and reiterated their commitment to this longstanding partnership and agreed to continue to strengthen their work together to tackle the challenges facing the feed and food chain.

**Mr. Newman, IFIF Chairman**, highlighted that “the FAO focus on five strategic objectives, emphasis on working in a

goal-oriented manner, and the FAO strong efforts reaching out to the private sector have made a tangible and positive difference in our already longstanding collaboration.”

Mr. Newman added that “together with the dedicated colleagues at the FAO we have achieved very important milestones, including the International Feed Regulators Meetings (IFRM), the Livestock Environmental Assessment and Performance (LEAP) partnership, as well as the Global Feed & Food Congress (GFFC) series and our relationship continues to strengthen year to year.”

**Dr. Daniel Bercovici, IFIF Chairman**, elect for 2018/19, said “our joint meeting with the FAO once again underlined our strong partnership and IFIF is committed to continue to support the FAO initiatives on capacity development for feed safety, the Global Agenda for Sustainable Livestock and LEAP, as well joint efforts on feed and food safety at the Codex Alimentarius. IFIF looks forward to our upcoming IFIF FAO 11<sup>th</sup> International Feed Regulators Meeting (IFRM) in Atlanta, USA, in January 2018, which is another great example of IFIF FAO collaboration positively impacting the feed and food chain.”

**Daniela Battaglia, Animal Development Officer at the Animal Production and Health Division of the FAO**, said “FAO and IFIF have a long standing partnership and this meeting addressed a number of critical issues of common interest, such as the need for capacity development to ensure feed safety and the importance of collaborating to tackle the containment of antimicrobial resistance (AMR). FAO is committed to work with the private sector and feed operators and believes that they can valuably contribute to make the livestock and food sectors more responsible and sustainable to achieve important goals such as public health, and animal health and welfare.”

## About IFIF

The International Feed Industry Federation (IFIF) is made up of national and regional feed associations, feed related organizations, and corporate members from around the globe, representing over 80% of the feed production worldwide. IFIF provides a unified voice and leadership to represent and promote the global feed industry as an essential participant in the food chain offering sustainable, safe, nutritious and affordable food for a growing world population.

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For more information on IFIF please visit [www.ifif.org](http://www.ifif.org) or contact Alexandra de Athayde, Executive Director, at [info@ifif.org](mailto:info@ifif.org).



# #towardsFieragricola - Verona, Italy

## 31 Jan. to 3 Feb. 2018

*Fieragricola has been a landmark in the international agricultural panorama for over a century and is the only event in Italy ensuring a complete coverage of agricultural topics.*



The wide-ranging exhibits, debates, technical and scientific meetings, performances and dynamic tests ensure that Fieragricola achieves impressive media impact and seeks to anticipate the needs of the market by creating relationships between exhibitors, visitors and sector associations and involving operators thanks to its dynamism and interactivity.

Always paying close attention to policies for shared growth and sustainability, Fieragricola has also accompanied the evolution of the Common Agricultural Policy since 1962, helping to stimulate debate between agricultural systems and the world of national and European institutions through dialogue focusing on planning the main directives to be adopted for sustainable growth, while also keeping abreast of the needs of producers.

### A crossway event focusing on agriculture through a vertical and complete trade offering:

- technologies and products for animal farming, livestock and genetics
- animal shows and auctions
- technologies and products for the renewable energy sector in agriculture
- agricultural machinery, equipment and technologies

- specialised machinery and equipment for vineyards and orchards
- demo areas (open field and vineyards)
- seeds and agricultural chemicals, plants and equipment for protected crops
- products and equipment for green management and forestry activity
- services for agriculture and livestock farming

### Exhibition trends - 2016 edition

The 2016 edition was the best Fieragricola for the last 10 years (5 editions). These results confirm, as was evident during the four days of the show, that agriculture has the potential to revive the economy. Fieragricola provided the answers that exhibitors were looking for in terms of specialist visitors from all Italian regions and impressive incoming attendance from abroad, particularly Austria, Switzerland, Germany, Romania, Russia, Finland, France and Japan, but also the Balkans and North Africa.

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## Cooperation Vencomatic Group BV and COBOT Automation

Vencomatic Group BV and COBOT Automation have decided to work together for the sale of her **M'eggbot**. This COBOT automatic palletizer complements Vencomatic Group sales program. With the extensive dealer network of Vencomatic Group, global coverage is achieved. The sale of the M'eggbot in the Netherlands will continue to be provided by COBOT Automation.

The addition to the sales program is aimed at further automation of egg collection and packing in poultry farms with a processing capacity up to 30,000 eggs per hour. In combination with the Prinzen packers type PSPC30 and Smartpack, the M'eggbot offers an automatically loading of pallets. The system uses the standard plastic tray, divider and pallet. For poultry farmers, the system not only provides labor savings but also pleasant working in the daily task of egg collection. And this, in return, contributes to safe transportation of the egg to the consumer.

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For more details:

**E-mail:** marketing@vencomaticgroup.com

**Website:** [www.vencomaticgroup.com](http://www.vencomaticgroup.com) / [www.cobot.eu/meggbott](http://www.cobot.eu/meggbott)

## Frédéric Fagnoul appointed as Director Hubbard R&D

*Global - Hubbard is pleased to announce the appointment of Frédéric Fagnoul as Director of R&D. Frédéric will report directly to Olivier Rochard, General Manager of Hubbard.*



Frédéric Fagnoul now heads up the Hubbard R&D team and succeeds Thomas de Bretagne, who decided to go back to swine breeding.

Frédéric graduated in 2000 as Agricultural Engineer in Liege, Belgium, with a specialisation in animal genetics. He first spent one year in Belgian Blue beef cattle breeding, before he joined Hubbard in 2001 as poultry geneticist.

During his 16 years with Hubbard, Frédéric first has been focusing on further developing genetic models for Hubbard's breeding program. Since 2008, he has been in charge of the Hubbard Premium products breeding program including a close practical follow up on pure line hatches and brooding, product testing and redesigning the selection process. More recently, the Hubbard R&D department has been implementing new technologies, such as genomics, RFID for a more accurate measurement of FCR, CT scanner, etc.

Frédéric's in-depth knowledge of the Hubbard broiler lines, selection process and (new) genetic tools, together with his proven management skills, assure the continuation and further development of all Hubbard R&D activities now and in the future.

The whole Hubbard team wishes Frédéric and his team a lot of success and a great future.




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For further information, please contact:  
[contact.emea@hubbardbreeders.com](mailto:contact.emea@hubbardbreeders.com)

# Koch Foods chooses Pas Reform and NatureForm for single-source hatchery expansion in the USA

*Chicago-based Koch foods, the USA's fifth largest poultry integrator has chosen to work with Pas Reform and NatureForm, the world's only single-source supplier of integrated hatchery solutions, to expand their hatchery at Crossville, Alabama. The news comes following an eighteen month trial at the company's Henagar operation.*



Pas Reform

Hatchery Technologies



KOCH FOODS

With a current weekly processing capacity of 14 million broilers, Koch Foods is starting its extensive renovation project at its Crossville hatchery in Alabama, where adding 48 SmartSet-Pro™ setters and 48 SmartHatchPro™ hatcher will increase capacity by 2.1 million eggs set per week, to bring total weekly capacity at this hatchery alone up 3.8 million eggs.

"Koch Foods is", says NatureForm's president Steve Warren, "fully accessing his company's single-source capabilities. This includes a complete HVAC system for both the new expansion and the egg and chick handling areas as well as chillers, pressure controls and humidification system, for a state-of-the-art climate control system throughout the hatchery".

The installation will also incorporate an entire range of hatchery automation systems, with farm to setter rack transfer systems,

auto-candling and transfer, chick separator, animal-friendly chick counters, washers, stackers and de-stackers.

Advanced, web-based SmartCenterPro™ hatchery information software will enable Koch foods to control setter, hatcher, climate control and hatchery automation (transfer, candling and chick counting) operations from a single computer, as well as providing remote access capabilities to manage and respond to system alerts or provide technical and troubleshooting support.

Don Davis, Complex Manager at Koch Foods, comments: "We have worked with NatureForm for many years now and were always of a mind to change over to single-stage incubation. Our goal is to put the absolute best chick possible in the field. So, we started trials with Pas Reform single-stage equipment in our Henagar operation and the results have clearly shown the advantages of the SmartPro™ system. This is the main reason we decided to proceed with Pas Reform and NatureForm."

Gary Davis, Vice President Eastern Division at Koch Foods, adds: "We were also very impressed by the dedication and expertise of their team during both the trial period and the discussions of the new project. It has become our firm belief that NatureForm and Pas Reform have the best team on the ground in the USA."

Steve Warren remarks: "We have worked with Gary Davis and his team for many years now and we are extremely proud of earning the confidence and trust of Koch Foods."

Bouke Hamminga, Director international sales and business development at Pas Reform, concludes: "This is a major milestone in Pas Reform's quest to truly become a world player and the world's first and only single-source supplier of fully integrated hatchery solutions. We are achieving this with robust, high quality technical and equipment solutions, but also with the introduction of a brand new, 360-degree approach to service and support - SmartCare™ - that is leading the way in our industry for delivering exactly the levels of support our customers need. We have seen how this matters to our customers – and as hatcheries increase in size and complexity, it will become pivotal to the future of our business. We look forward to building a long-lasting partnership with Koch Foods over the coming years."

Construction is now underway at Crossville, where the first chicks from the expanded hatchery are expected in June 2018.



## Danish Royal's visit to Japan with a special stop at the SANOVO TECHNOLOGY GROUP's stand

SANOVO TECHNOLOGY GROUP welcomed the Danish Royal couple's visit to their stand in Japan, where they could see the state-of-the-art company's technology for egg processing and talk with the company's CEO Michael Midskov.

From 10 to 13 October 2017 the Danish Royal Crown Prince Couple visited Japan to celebrate 150 years of diplomatic ties between Denmark and Japan.

On October 11, it was a great honor for the SANOVO TECHNOLOGY GROUP to welcome HRH the Crown Princess of Denmark to their stand. She had a talk with the company's CEO - Michael Midskov - about SANOVO technology and connection to the Japanese egg industry. In the afternoon, a signing luncheon with Sansyu Syokuhin Co. Ltd. and Chairman of the Board Thor Stadil took place. HRH the Crown Princess of Denmark also visited the Hummel stand where she had a talk with Christian Stadil, Owner of THORNICO.

On October 12, HRH the Crown Prince of Denmark visited a SANOVO's long-term valued customer, who is the largest mayonnaise producer in Japan. During the visit, he just started up the production of a new egg-breaking machine.

---

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Website: [www.sanovogroup.com](http://www.sanovogroup.com)

## Clark named Supply Chain Manager for Chore-Time



**Judi Clark has been named Supply Chain Manager for Chore-Time, according to Jeff Miller, Vice President and General Manager for the CTB, Inc. business unit.**

In her new position, Clark will be responsible for leading all aspects of purchasing, planning and scheduling as well as inventory control and forecasting.

Clark joined the Chore-Time team in 2016 as Planning and Scheduling Manager, and she quickly became an integral part of the continuous improvement efforts underway within the operations team. She has also led initiatives to increase efficiency in materials handling and inventory control.

Prior to her employment with Chore-Time, Clark gained more than 25 years of related work experience, including 17 years of experience in industrial materials and supply chain management. She holds a degree in organizational leadership from Purdue University in West Lafayette, Indiana, and, during the course of her career, she has achieved certification through the American Production and Inventory Control Society (APICS), Six Sigma Green Belt Certification and has become a Certified Purchasing Manager (CPM).

Clark is a native of Brownstown, Indiana, and currently resides in Leesburg, Indiana.

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## **“Il Fiore delle Dolomiti”: organic eggs produced using SKA equipment**

*Mountain organic eggs from hens grown according to the principles of the 5 H Club. Respecting the animal, the environment and the health of the consumer.*

“Il Fiore delle Dolomiti” is a fully evolving reality. This family business was built from scratch in 2015 in Limana (Belluno). The farm layout consists of 5 sheds (which look like the petals of a flower from above), each of which accommodates 3000 laying hens, with the offices and packaging plant located in the centre.

The SKA company, which specializes in the production of equipment for layers housed on the floor and in alternative systems, have been closely involved in the project through the design and building phases as well as supplying all the interior equipment.

The owners explained that they chose the SKA company for their decades of experience in the development of alternative systems; their expertise in the organic field and their vast product range





designed to provide a natural and comfortable environment for livestock. In addition, the proximity of the SKA headquarters to "Il Fiore delle Dolomiti" reduced the incidence of transport and, consequently, the potential pollution from CO<sub>2</sub> emissions. Respect for the environment is, in fact, one of the cornerstones of the company's business philosophy.

Production began in January 2016 and every day around 14,000 eggs are packaged and then distributed directly by the company to specialized stores, delicatessens and some supermarket chains under the brand name LUOVO, which summarizes their business philosophy of managing their flocks of laying hens with respect for the animal, the environment and the health of the consumer.

## The 5 H Club

Hens are reared according to the principles of **the 5 H Club** which is an exclusive **organic farming method**. The 5 H Club sums up a series of values five in total, which represent and translate into good everyday practices guaranteeing the laying hens a state of complete health, in which the bird is in harmony with its environment.

*"The 5 H Club is a trademark, a private warranty certificate, which we have created and registered at European level, with reference back to the USA youth organization 4 H (Head, Heart, Hands and Health)," explained the owners. "This youth movement founded over 100 years ago, exists in 80 countries and has as its foundation the harmonious development of the individual which can be achieved by the continuous improvement of the intellect (Head) while at the same time, the strengthening of one's manual craft (Hand). In addition, you must always preserve your physical well-being (Health) combined with great care and respect for your neighbour, our planet, and the animals that give us food (Heart). Using this concept as a base we have entered a fifth H (Hen) to give birth to a healthy food, born out of respect for the livestock, the land and the consumer."*

## In the name of Eco-sustainability

Hens reared on the Limana farm are fed GM-free rations. In addition to feeding outdoors in the open air birds are fed a mix



of grit, essential to the functioning of the digestive system, combined with organic maize and soya.

*"In addition, we enrich the feed with a special variety of dark mountain corn, known as 'Bellunese Tambar Gold', which is nourishing and rich in fibre and which is cultivated biologically in the Belluno area. Although its yield per hectare is very low, we choose this because the corn cobs are small and tasty and add an impressive colour to the yolk of our eggs", say the owners.*

Extreme attention is also paid to the drinking system with the water being dynamized thanks to the innovative AquaPhi™ natural system where the latest technology manages to retain all the original properties of the water.

Eggs are selected and packed by hand in wooden pulp boxes on the farm thereby eliminating the need to transport them to external packing centres. This guarantees the freshness of the product, greater protection of the environment and the total absence of the risk that organic eggs will come into contact with eggs of other types.

The whole company operates in an environmentally friendly manner with the installation of photo-voltaic systems installed on the roof for self-production of energy which is supplied through NWG Energia, the only company in Italy that, by statute, provides energy exclusively from renewables. In addition, only LED lights are used in all the farm buildings.

## SKA, a reliable and competent partner

With its traditional vocation to customize plants, SKA has successfully managed at "Il Fiore delle Dolomiti" to build an innovative and highly technological set up.

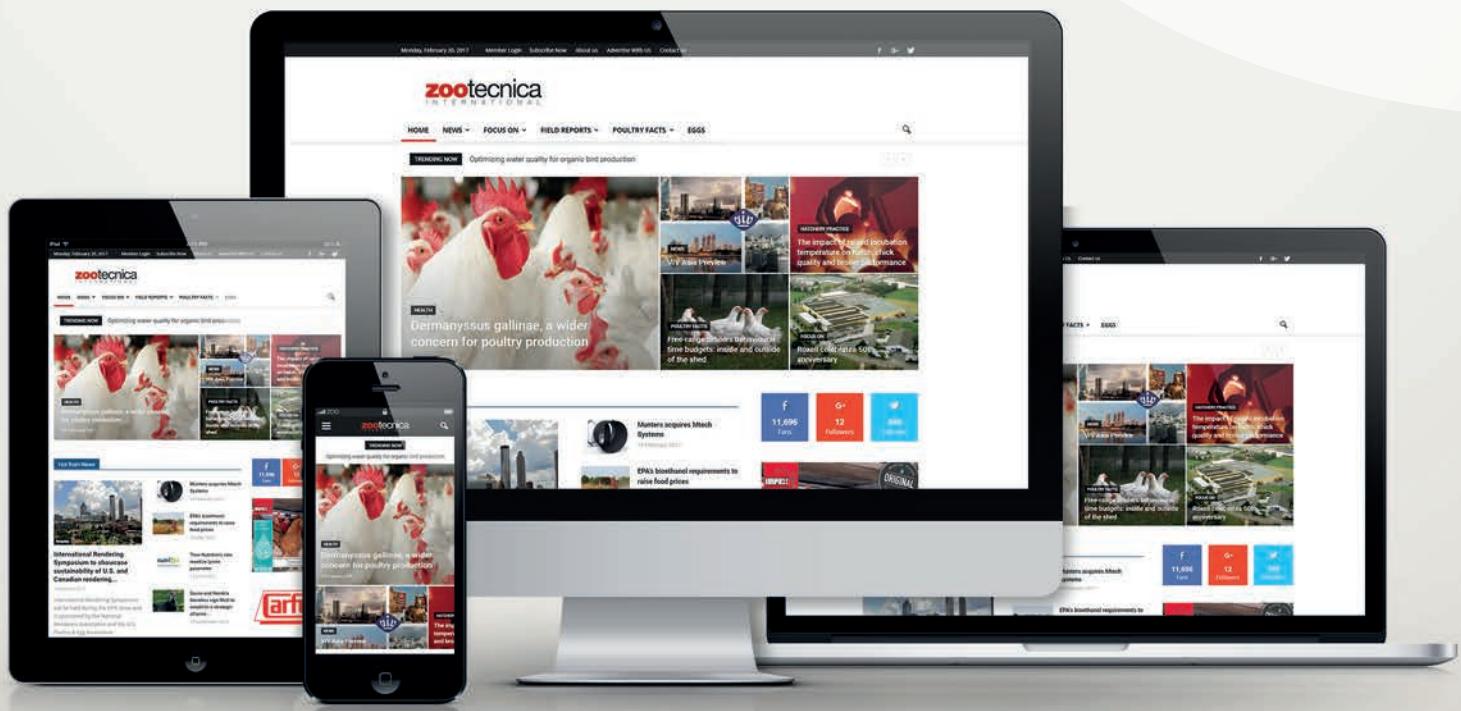
All the structures are galvanized steel, and are equipped with poly-carbonate windows. The "AVIO 500" communal automatic nests, arranged in a single longitudinal central row in the shed, providing the hens with a very comfortable environment for laying eggs. Particular attention has been paid in designing the nests to ensure eggs are not damaged and are kept clean. Hens have the choice of two entry points to the nest set-up and lay their eggs on a special mat made of synthetic material "AstroTurf®" and the eggs roll gently into a central collecting channel, set between the two rows of nest boxes. Eggs are conveyed on a wide rigid polypropylene belt with a variable speed traction unit to the designated egg collection table.

The SKA cross conveyor "SPIRALINE 90®" system has been installed as the automatic system for transporting feed from the silo to the in-house feeding layout. For the "Il Fiore delle Dolomiti" company SKA decided to install the "FLATLINE" chain feeder system renowned for its proven reliability for fast, uniform transportation of feed. Thanks to this system, all hens are able to feed evenly and at the same time.

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## The USA on their way to cage-free egg production

### ***The Red River Valley Egg Farm in Bogata, Texas, USA***

---

Hans-Wilhelm Windhorst  
Prof. Emeritus and  
Scientific Director of the  
Science and Information  
Centre Sustainable  
Poultry Production  
(WING),  
University of Vechta,  
Germany

*In June and July 2017, the Author had the chance to visit new large cage-free egg farms in Texas and Arizona. The main results of this research project are presented in two different papers. This article will focus on the Red River Valley Egg Farm in Bogata, Texas.*

# FIELD CASES

## The cage-free discussion in retrospect

The discussion about abandoning conventional cages in laying hen husbandry gained in momentum in 2008 when 63% of the voters in California approved the so-called Proposition 2 which would prohibit conventional cages from 2015 on. In addition, a law was passed which prohibited the import and sale of egg which had been produced in this housing system also from 2015 on.

Because of the possibility that similar ballots in other states might have the same result, United Egg Producers, in co-operation with the Humane Society of the United States, the leading animal welfare organisation in the USA, in 2011 tried to pass a law in the US Congress, the Egg Bill, which would have regulated the transformation to alternative housing systems. The

*“Leading egg producing companies began to build large cage-free farms or to restructure older farms, even though this was less important. One of these new farms is located in Bogata, Texas”*

bill did not pass Congress in 2013. The discussion began to gain in importance in April 2016 when several food retailers and restaurant chains published statements in which they declared that they would not sell or use eggs produced in conventional cages from 2022 respectively 2015 on. Until April 2017, 219 companies published similar declarations. If their plans would be realised, over 71 % of the U.S. layer flocks would then be

needed to produce the annual demand of almost 72 billion cage-free eggs.

Leading egg producing companies began to build large cage-free farms or to restructure older farms, even though this was less important. One of these new farms is located in Bogata (Texas).

## The Red River Valley Egg Farm in Bogata (Texas)

Bogata, a small community with less than 1,100 inhabitants, is located in the fertile Red River valley in northern Texas. Farmers grow corn, soybeans and cotton. Until 10 to 15 years ago, Bogata was a central city of some importance. But when a new highway was built and with the increasing mobility of the inhabitants, it very fast lost this function and the Main Street degraded. The decision of Cal-Maine and Rose Acre, the two leading

egg producing companies in the USA, to build a layer farm with 1.7 mill. layer places, was like a struck of luck. About 100 new jobs were created and the community began to recover.

Plans to build the farm started in 2014 and towards the end of the year, construction began. The farm is a joint venture of the two leading egg producing companies in the USA: Cal-Maine and Rose Acre. They decided to build a cage-free farm together



**Photo 1** - A total view of the Red River Valley Egg Farm in Bogata (Texas)



**Photo 2** - Once the feedmill will be completed, feed will be transported to the barns via conveyor belts

## FIELD CASES



**Photo 3** - The high outside temperatures make large evaporation cooling systems necessary



**Photo 4** - Large vents are installed to generate the necessary air flow through the long barns

***"Plans to build the farm started in 2014 and towards the end of the year, construction began. The farm is a joint venture of the two leading egg producing companies in the USA: Cal-Maine and Rose Acre. They decided to build a cage-free farm together as both companies had no experience with cage-free laying hen husbandry. The location was selected because of a rather central position in the USA and of the excellent traffic infrastructure."***

as both companies had no experience with cage-free laying hen husbandry. The location was selected because of a rather central position in the USA and of the excellent traffic infrastructure. Other aspects were the fact that no poultry farms existed nearby in the valley and that feed could be bought from adjacent farms and the dry manure be delivered to these farms.

In total, the farm consists of seven houses with 240,000 places each (Photo 1). In addition, two pullet farms were built in a distance of about one mile. The farms will be able to supply the layer farms with the needed pullets. At an age of 16 weeks they are transferred to the layer farms. The available space for white hens in the layer farm is 930 cm<sup>2</sup> and 1.116 cm<sup>2</sup> for brown hens.

The feedmill will be completed by the end of this year and have a weekly capacity of 2,700 t. So it was still necessary to haul the feed by truck from Sulphur Springs to the farm over a distance of 65 km. Once the mill will be in operation, the feed will be transported to the bins in front of the single houses by conveyor belts (Photo 2).

In the barns, two housing systems are used; one from the Italian company FACCO and one from the German company BIG DUTCHMAN. The barns have three stories. In each sto-

ry aviaries with three layers are installed. The barns are about 40 m wide, 16 m high and 160 m long. Horizontally they are divided in ten units, each 16 m long. This is necessary to keep the hens from piling up at the cooler end of the barn.

The temperature at the end with the evaporation coolers (Photo 3) was between 22 and 24 °C, at the other end where the huge vents are located (Photo 4) it was about 3 to 5 degrees higher. A very comfortable climate inside compared to the almost 40 °C outside. In winter, when the outside temperature decreases considerably, the ventilation system can be shut down during the night hours.

On the premise, the processing facility is connected online with the layer houses. Here the eggs are washed, graded and packed. A packing in units of 6, 12 and 18 eggs is possible. White eggs can be separated from brown eggs and the eggs can also be processed separately for the two owners. The majority of the employees are working here.

### First experiences with the cage-free systems

The farm manager and the production manager confessed that the handling of the cage-free systems had been a challenge in

the beginning but did no longer cause problems. The customers demand that twice a day the whole farm has to be inspected story by story and aviary by aviary. This is time consuming but guarantees an excellent overview on the situation of the flocks.

Feed conversion and mortality were higher than in farms with conventional cages. When visiting the farm, 3.5 pounds (1,6 kg) of feed were needed to produce a dozen of eggs which equals a conversion rate of 1.9 to 2.0. Because of the lasting low egg prices in the USA, the production costs could not be covered by the market prices. This is the reason for not building a similar farm in a distance of 25 km despite the building permission. The project will only be started when profits can be expected.

The eggs are marketed through the headquarters of Cal-Maine (Jackson, Mississippi) and Rose Acre (Seymour, Indiana). When visiting the farm, it was not possible to market all eggs as "cage-free", as the demand for such eggs was lower than production. Obviously, consumers were not willing to pay the considerably higher price for these eggs compared to the price for eggs produced in conventional cages.

### Perspectives

After visiting the farm, the Author could discuss recent problems of the U. S. egg industry with representatives of Cal-Maine and the vice president of the Texas Poultry Federation, James Grimm. One major problem in their opinion is the un-coordinated transformation to alternative housing systems and the lack of a nationwide valid definition of "cage-free". Presently, the installed systems are only certified by the customers who purchase the eggs. This is a high risk, they declared, because other customers may not accept the system as cage-free.

A second major problem is the high oversupply of eggs resulting from the building of large new non-cage farms without closing down older farms with conventional cages. Regarding the extremely low prices at farm gate, it would not make sense, however, to close the old farms because production costs were considerably lower with the old system. Nevertheless, such a step would be necessary to bring production and demand into balance again, was their unanimous statement.

The observed reduction of placed hatching eggs was a first positive indicator. A stabilisation of the egg prices was not expected by them before the second quarter of 2018.

### Additional references

Windhorst, H.-W.: Cage-free heißt das Zauberwort. Ausstieg aus der Käfighaltung in den USA steht bevor. In: Deutsche Geflügelwirtschaft und Schweineproduktion 68 (2016), Nr. 27, S. 3-5.

Windhorst, H.-W.: Ist das Chaos vorprogrammiert? USA: Umstellung der Legehennenhaltung hin zu käfiglosen Alternativen. In: Deutsche Geflügelwirtschaft und Schweineproduktion 69 (2017), Nr. 19, S. 3-5.



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# A model for promoting poultry industry development in Sub-Saharan Africa

*In commercial poultry husbandry practice, the hatchery takes over the incubation of bird eggs in order to provide as many day-old chicks as needed at any time to farmers. The main bottleneck for poultry industry development in Togo, and its neighbouring countries, is the lack of day-old chick supply.*

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K. Tona, M. Gbeassor,  
E. Decuyper,  
A. Agbonon, P. Simons,  
N. Everaert

## FIELD CASES

Indeed, there is no proficient hatchery, which can cover the needs of the farmers because of lack of information about hatchery management or people trained as hatchery managers. There is also lack of information on management practice aspects and feed rations formulation and other. With the aim to promote and develop poultry industry in these countries, an interuniversity project [Catholic University of Leuven (KUL) and University of Lome (UL)] as a model of poultry industry development is running.

### Specific objectives of the current project are:

1. to provide insights and disseminate guidelines and information on adapted methods to improve poultry production and,
2. to focus on development of new technologies in poultry production and implementation of research on poultry management practices.

Livestock, especially poultry, make a substantial contribution to household food security by providing income, quality food, fertilizer and assets in over 80 % of rural households in de-

could substantially improve productivity and income generation.

In developing countries, two main systems of poultry production can be distinguished. These include rural and commercial farming systems. The rural system concerns mainly poultry-yard, called village chickens, and is widely practiced. Although the production of village chickens is still an important activity and helps preservation of within species biodiversity, its commercial impact is decreasing every year in favour of increasing commercial poultry production system.

The objective of the current project is to investigate different aspects of poultry production, especially adapted incubation conditions, improved management practices and the use of feed components available in Togo in formulating adequate chicken feed rations. On the other hand, this project will provide information for poultry production improvement in Togo and its neighbouring countries. Also, it will focus on the training of local staff in order to build up the research facilities at UL. This will help the sustainability of the project as well as the extension of the results at the farm level and/or new small-scale industry as spin-off of the project.

*“Although the production of village chickens is still an important activity and helps preservation of species biodiversity, its commercial impact is decreasing every year in favour of increasing commercial poultry production systems”*

veloping countries. Constraints faced by the rural producer in resource-poor areas include: goods and services, weak institutions, lack of access to markets, lack of skills, knowledge and appropriate technologies. As a result, both production and productivity remain below potential and losses and wastage can be high.

However, imported breeds can be adapted and local feed resources are available, along with proven technologies that include preservation and value-added product processing which

Day-old chicks' importation from developed countries leads to long delay in feed and water access. However, a long delay in feed access and/or inappropriate climatic conditions during the first days of life for day-old chicks contribute to lower post-hatch performance. Therefore, the establishment of a local hatchery industry may reduce day-old chick delay and provide enough day-old chicks to the farmers and decrease significantly the time delay for access to feed and water. In addition, feeding chickens with feed rations incorporating a large amount of



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# FIELD CASES



poultry houses and office for the project for its feasibility were provided by UL. Several studies, capacity building and extension activities were run in the framework of the project.

## Research activities

They included a relationship between management practices and production performance of chicken lines or commercial crosses in Togo, incubation conditions of hatching eggs and rearing managements of hatchlings, evaluation and description of the effects of delayed feed and water access on post-hatch performance, and determination of nutritive values of local feed components, formulation of suitable rations with incorporation of plant products with prebiotic activity.

## Capacity building

Training of scientists as well as technicians with regard to poultry science and poultry production management is part of the project activities. These activities were possible by setting up a Laboratory of Poultry Science, which was the first spin off of the project.

In total, 19 scientists were trained in

1. analyzing and interpreting poultry production parameters and relate these to management practices,
2. formulation of feed rations according to the need of chickens and in analyzing and interpreting relationship between rations and poultry production parameters,

*"The objective of the current project is to investigate different aspects of poultry production, especially adapted incubation conditions, improved management practices and the use of feed components available in Togo in formulating adequate chicken feed rations"*

local feed components as well as plant products with possible antimicrobial or prebiotic activity may improve production performance. But, no study on adapted incubation conditions and hatchery management has been done in Togo and its neighbouring countries. Furthermore, there is no scientific information about the effects of local environmental conditions on poultry production. Such information is also scarce in the neighbouring countries (Ghana or Benin), and there is few information on eggs and poultry meat production management practices.

## Project facilities

To address the objectives of the project, incubators and its accessories were acquired as well as some laboratory equipments provided by Flemish Interuniversity Council (VLIR) through KU Leuven. Infrastructures such as incubation room,

3. interpretation of the relationship between incubation conditions and embryonic or post-hatch performance parameters and,
4. analysis and interpretation of the poultry products' quality (egg, meat). Different levels of training and education including Bachelor or Animal husbandry technician with Poultry technician (6 students), Bio-Engineer (6 students), Master of Science (4 students) and 1 PhD and 5 doctoral students were involved in the project activities.

## Extension activities

These activities are mainly focused on development of strategies for increased chicken population and productivity resulting in a sustainable improvement of poultry production in Togo. These strategies are disseminated through seminars and

## FIELD CASES



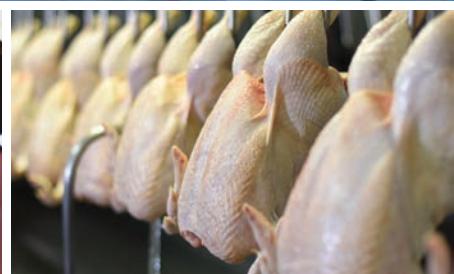
training courses at different levels to the attention of stakeholders. Previously, a branch of World Poultry Science Association (WPSA) was established in order to create an appropriated technical and scientific environment for improvement of poultry production. Through this channel and an existing organization of those involved in poultry industry, a programme of short term training courses was developed - and it is still running - with existing facilities and several thematic seminars were organized.

### Conclusion

The outcome of the project provided basic information for poultry industry. Also a Laboratory of Poultry Science was set up in UL. This laboratory together with WPSA-Togo takes over the research and development activities in poultry science. With regard to technology and knowledge transfer between developed and developing countries the project can be considered as a model. Recently, the laboratory has been selected to become a Centre of Excellence for Poultry Science in West and Central Africa.

From the Proceedings of the Potential for Poultry Production in Developing Countries, Antalya

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# Isolation, bioactivity and applications of sulfated polysaccharides from poultry by-products

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Mirko Betti, PhD

Department of  
Agricultural Food and  
Nutritional Science  
University of Alberta,  
USA

*The broiler chicken meat processing industry produces a large amount of by-products rich in connective tissue (i.e. keel cartilage, skin, and bone residues) which can be exploited for isolating important natural health ingredients. Connective tissue contains the extracellular matrix that includes collagen, glycoproteins, proteoglycans and glycosaminoglycans (GAGs).*

Proteoglycans are the major components of this extracellular matrix and are comprised of two types of molecules: the glycosylated protein core and covalently attached sulfated GAGs. These acid polysaccharides from various animal sources have been extensively studied. They are described based on their disaccharide composition and degree of sulfation; on one hand, the sulfated GAGs, such as chondroitin sulfate (CS), dermatan sulfate (DS), keratin sulfate (KS) and heparan sulfate (HS) are all bound glycans to the protein core; and on the other hand, hyaluronic acid (HA), the non-sulfated GAG polysaccharides, all exist as a free polymer (Nakano *et al.*, 2010).

Sulfated GAG polysaccharides can be liberated from the extracellular matrix by enzymatic or chemical hydrolysis and are considered biologically active compounds. They have a wide range of applications in pharmaceutical and food industries. For instance, CS is used as a supplement for treating osteoarthritis due to its anti-inflammatory and chondro-protective effects.

HS and DS also have anticoagulant activities. Besides these known properties, recent *in vitro* studies suggest that GAG polysaccharides can enhance non-heme iron absorption, thereby improving one's nutritional iron status. For instance, the GAG-containing fraction of cooked haddock has increased the iron uptake of epithelial Caco-2 cells in a simulated gastrointestinal model. Hence, this opens the possibility of extracting GAGs from poultry processing by-products with the primary aim to produce bioactive compounds that can be used in product formulation to produce functional foods.

## Extraction and separation of GAGs

Different selective extraction procedures and precipitation techniques have been used to isolate and obtain different types of sulphated GAG polysaccharides. Various attempts have been made to minimize the use of organic toxic solvents and chemicals during the extraction and separation processes. A food grade extraction according to the methodology of Nakano *et al.* (2012) was conducted on pre-cleaned chicken skin and cartilage tissues. This extraction involves the use of an economical proteinase for liberating the sulphated GAG polysaccharides from the extracellular matrix and a subsequent membrane ultrafiltration at 10 kDa molecular weight cut off (MWCO) to replace the common hazardous chemical deproteinization step involving trichloroacetic acid. The extraction of food grade GAGs from poultry by-products is reported in the *Figure 1*.

### Tissues

Cartilaginous tissues are the major source of CS, while most galactosaminoglycans in non-cartilaginous connective tissues (e.g. skin, tendon and skeletal muscle epimysium) are CS/DS. Tissue samples are cut into small pieces. Cartilage tissues with relatively low fat content may be used for GAG extraction without defatting.



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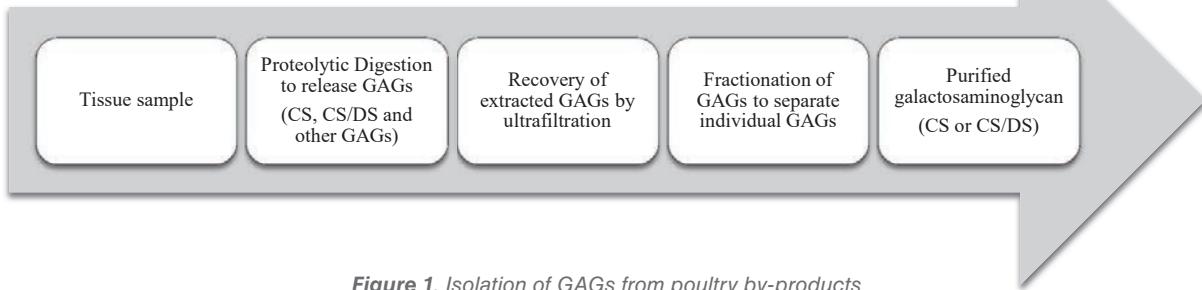




BABY AREA  
AVIARY SYSTEM FOR PULLET

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YOUTUBE

**Figure 1.** Isolation of GAGs from poultry by-products

Bones are crushed with a hammer, frozen in liquid nitrogen and ground while still frozen in a Wiley mill to obtain a small tissue size. Bone tissues need to be decalcified using EDTA or acid (e.g. HCl). Without decalcification, efficient proteolysis of bone tissues is difficult to achieve.

### Proteolysis

Whole GAGs can directly be liberated from tissues by hydrolysis with exogenous enzymes (i.e. pancreatin). GAGs can also be liberated from cartilage by activation of endogenous enzymes (autolysis) without using exogenous proteinase.

### Recovery and fractionation

The GAGs liberated by proteolysis are subjected to an ultrafiltration step with 10 kDa MWCO. GAGs have a MW of around

20 kDa, therefore they can be easily separated from the peptides (mostly collagen peptides).

After ultrafiltration, ion exchange chromatography using a gradient of NaCl is used to fractionate the GAGs. Two main fractions are obtained: the 0.4 M, containing mostly unsulfated GAGs and the 2.0 M NaCl which contains the sulphated GAGs (i.e. CS and DS). The 2.0 M fraction is then subjected for selective precipitation by using different concentration of food grade ethanol in order to recover different types of sulphated GAGs.

### Bioactivity of GAGs

GAGs have been used clinically for the treatment of chronic diseases such as degenerative arthritis, cirrhosis and chronic photo damage due to the emerging bioactivities such as antiox-

***“Sulfated GAG polysaccharides can be liberated from the extracellular matrix by enzymatic or chemical hydrolysis and are considered biologically active compounds. They have a wide range of applications in pharmaceutical and food industries”***



dant, anti-atherogenesis, anticoagulation, prevention and cure of arthritis. Their bioactivities depend on their specific chemical structure, including the patterns and degree of sulfation, molecular mass, relative amounts of iduronic (IdoA), glucuronic acids (GlcA) and hexosamine. For instance, HS is used as an anti-coagulant, while HA as a component in the synovial fluid lubricant in body joints. CS is used in the treatment of cartilage and tendons. Despite these known applications of GAGs, novel function and uses are emerging.

Of interest is the possibility of using low molecular weight GAGs (i.e. oligosaccharides) as a prebiotic or for their ability to increase micronutrient bioavailability.

### GAGs and iron bioavailability

Fe deficiency is a common and debilitating primary nutritional disorder, and can result in anaemia and brain development abnormalities. Fe deficiency is still common in the developed world and keeps many people from thriving. The promotion of



Fe absorption is a practical way of dealing with modern micro-nutrient deficiencies. Certain components in muscle tissue can enhance Fe absorption, a phenomenon called the “meat factor”.

Active molecules that enhance Fe bioavailability have been isolated from fish and meat processing by-products, like fish skin collagen. Recently, Wang and Betti (2017) demonstrated that GAGs extracted from poultry skin and cartilage possesses a certain antioxidant activity and ability to increase iron bioavailability in intestinal cell culture model.

Ferritin formation in intestinal culture model was also enhanced by the hydrolysis treatment, indicating that the GAGs-derived oligosaccharides have ability to increase the iron bioavailability. These results are of practical importance and may represent a very significant way to add value and increase sustainability of the meat processing industry. For instance, current studies in my research group are focusing on the supplementation of GAGs in several food matrix including milk products. The aim is to create food product that have ability to increase the bioavailability of key-micronutrient like iron.

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#### References are available on request

From Proceedings of Midwest Poultry Federation Convention,  
St. Paul, Minnesota, USA

An advertisement for Victoria Incubators. It features a large, stainless steel industrial incubator with multiple shelves and a control panel. The Victoria logo, which consists of a circle with red horizontal stripes and a black bird silhouette, is prominently displayed. Below the logo, the text "VICTORIA" is written in large, bold, black letters, followed by "INCUBATORS SPECIALIST SINCE 1924" in smaller letters. At the bottom left, the text "New Line Setter:  
EXTRA LARGE SERIES" is written. At the bottom right, the address "22070 Guanzate - COMO - ITALY" is listed, along with the company's email "victoria@victoria-srl.com" and website "www.incubaticivictoria.com".

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## Jamesway's last webinar talking about the Hatcher

*The hatcher is often the forgotten sister of the incubator. Most presentations focus on getting everything right in the incubator, assuming that if the incubation portion is right, the hatcher will just finish off all that good work.*

The reality though, is that the hatcher needs to be used to continue the progress in incubation and even compensate for some potential shortcomings, such as temperature gradients, experienced in the incubator.

### **There are 3 major phases involved in the incubator to hatcher process:**

- Transfer
- Maintenance
- Hatching

As well there is the option to further customize what is happening through the use of profiling on your Platinum hatchers, or multi-stage hatchers through Hatchcom III.

Transfer is defined as the process which moves the eggs from the incubator to the hatcher; including the movement between egg flat and hatcher basket. This process encompasses more and

specific transfer patterns and placement into the hatcher from the incubator can be used to improve the overall hatch. An example for our Single-Stage Platinum would take those eggs that were in a slightly cooler portion of the incubator, often the outside walls, and move them towards the center and the ECU in the hatcher, thereby moving them to the warmest area. Jamesway has specific transfer pattern guidance available for all combinations of equipment, which can be received by contacting the nearest Jamesway consultant or [webinars@jamesway.com](mailto:webinars@jamesway.com).

A key point brought up by Jamesway Hatchery Consultant, Phillip Perry, was that with the push towards automated transfer hatchery managers need to pay close attention to the transfer pattern their equipment provides, and ensure that it meets Jamesway's recommended transfer guidelines for whatever equipment you have installed.

### Other key tips for transfer:

- Handle eggs with care
- Do not remove all incubator racks from one side of a Platinum incubator – this disrupts airflow
- Minimize the time the eggs are out of the incubator; do not allow them to cool excessively
- Do not transfer into wet baskets
- Complete transfer for all the racks in one cabinet without stopping
- Ensure dollies are loaded and positioned properly according to the manufacturer's recommendations
- Ensure alarms are active once the hatcher has reached operational temperature and humidity set points

Maintenance is simply the process where we keep the eggs in a stable environment until the hatch begins. Generally this involves stepping down the temperature set point in the hatcher, such as from 97.6°F to 97.3°F. This process is important to ensure eggs are kept warm. If there is a need, the temperature can be kept higher to speed up the hatch or the humidity may be dropped to increase the weight loss of the egg.

The hatching process then begins. Jamesway expects a hatch window of 24 hours from first chicks to last finish for Platinum equipment; 30 hours for multi-stage.



In single-stage equipment, we can expect to see a humidity bell curve which tracks the hatch window exactly. In order to view this, Jamesway recommends against the use of humidification in the hatchers (providing a room humidification level is maintained at 45%RH), as it will hide the actual humidity bell curve. As seen in *Picture 1*, there is a clear increase in humidity that occurs as the chicks hatch out, due to their natural processes. As the chicks dry out and fewer are hatching, the humidity drops. This is visible both on the Platinum interface graphs as well as in Hatchcom.



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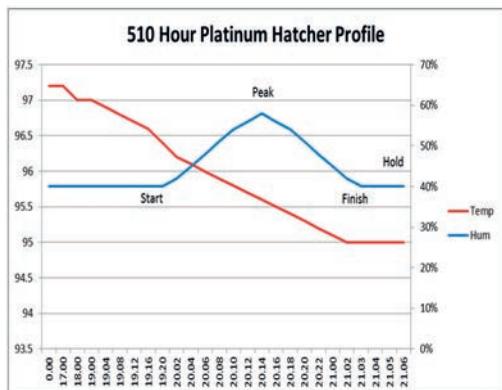
**Picture 1 - Platinum display hatch graph**

In multi-stage, the hatch is generally monitored by visual inspection. 12 hours prior to pull, 50-60% of chicks should be hatched, with 10% of those chicks wet or just hatched. However, Jamesway has discovered - you can also see in Hatchcom - the effects of the humidity bell curve created by the chicks by monitoring the spray usage. As the hatch progresses to its peak, the sprays will drop to 0 usage, and then as we head to the finish of the hatch, the sprays will begin to come back on. This allows the hatchery manager to check progress without opening the cabinet and disrupting the internal environment.

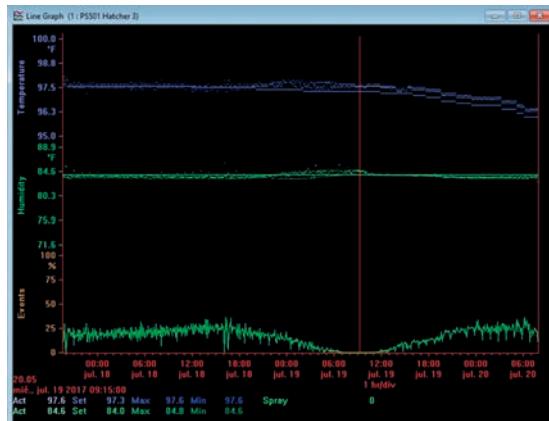
Now that we can identify when the hatch is happening, we can profile our machines to best suit those chicks as they hatch out. Profiles for single-stage are standard and included by default in your hatcher to suit the needs of the chick. Remember that humidity sprays should be disabled in the single-stage hatcher to allow the clear view of the bell curve. The profile includes a humidity rise and peak as shown below to approximate where the bell curve should be occurring; this also reduces the incidence of high humidity alarms.

## It is also possible to profile in the multistage hatcher provided the following equipment is in place:

- PS501 controller or existing PT100 controller,
- Hatchcom computer with a stable communication link to the equipment.



**Picture 3 - Typical Platinum Hatcher Profile**



**Picture 2 - Hatchcom Multi-Stage Hatcher Graph with Spray (Humidity) Duration**

Hatchcom allows for temperature and humidity profiles in the multistage hatchers. Jamesway recommends that the temperature decrease as the hatch progresses, rather than using a single set point or manually changing the set point throughout the duration of the time in the hatcher. Before embarking on profiling your multistage hatchers, please contact Jamesway's experts for advice and guidance. It is essential there is a stable connection as the profile is not stored in the multistage controller but instead only in Hatchcom.

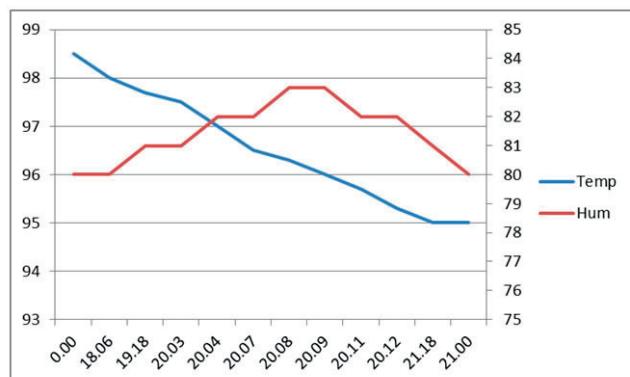
You can watch a live replay of the webinar at <https://youtu.be/VEbIZ4tGGFo> and also view the next two webinars: 5 Things You May Not Know about Hatchcom (at <https://youtu.be/I2-gVLQNhCY>) and a special edition "Ask Us Anything" open question and answer forum with a panel of Jamesway Hatchery Experts at <https://youtu.be/c11INIFpkI>.

## Nov 15: Poultry Industry Trends: Metabolic Heat

Metabolic heat is a hot topic in hatchery science today, becoming the focus of much research as we improve our genetics, creating the new "high heat" birds. Join Jerry Garrison to learn about Metabolic Heat and what it means for your hatchery.

## Dec 13: Biosecurity in your Hatchery

On the heels of recent avian flu, salmonella and other infections, biosecurity must always remain at the forefront of everyone's minds. Join Jamesway as we discuss hatchery biosecurity and what it means for us, as the manufacturer, as well as what it means for you, the hatchery manager.

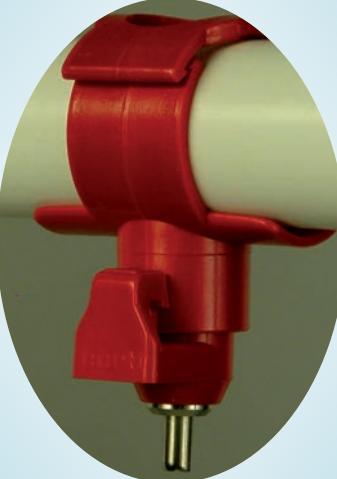


**Picture 4 - 2-Door Hatcher Hatchcom Profile**

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## Identifying feather pecking and feather eating using artificial feather presentation

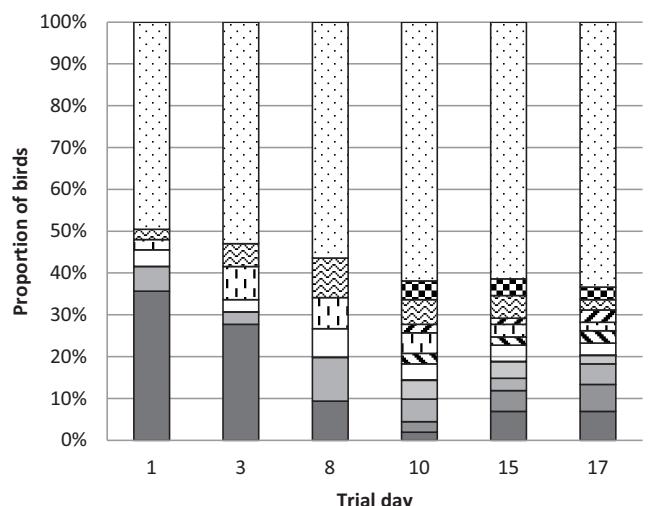
*Feather pecking and subsequent cannibalism outbreaks are the two of the main welfare concerns associated with free range egg production. Both have been linked to dietary nutrient deficiencies with Wylie et al. (2003) suggesting that feather eating behaviour indicates an attempt by the bird to obtain something that the feather can provide.*

---

K.M. Prescilla<sup>1</sup>,  
G.M. Cronin, S. Liu  
and M. Singh

<sup>1</sup>Poultry Research Foundation, The University of Sydney, Camden, NSW, Australia

*Although feather keratin typically represents 85% of feather protein, digestibility of keratin by the bird is very poor and it is unclear whether birds are able to digest feathers to any degree. However, consumption of feathers does suggest potential nutritional motivations behind the behaviour, either for a specific nutrient, or for insoluble structural fibre.*



**Figure 1** - Mosaic plot of the proportion of total birds ( $n=202$ ) removing feathers at each proportion level across each trial day. Each fill pattern represents the proportion of feathers removed from presented substrates after 2 hours of presentation.

## Method

A total of 202 individually housed Isa Brown hens were obtained from a commercial supplier at 16 weeks of age and given four weeks to habituate. Hens were fed standard commercial mash diet provided *ad libitum* and water was provided from nipple drinkers located at the back of the cages. Clear plastic lids measuring 17 x 12 cm were drilled with a total of ten 1.8 mm-diameter holes and suspended in front of cages for 14 days to familiarise the hens to the device.

Latency to peck was limited to 30 s, with observations stopping if birds did not peck within 30 s. All other measurements were taken within a 30-s period after the first pecking bout was observed. Statistical analyses were computed using GenStat 16<sup>th</sup> Edition (VSN International Ltd, Hemel Hempstead, UK). Means were compared using ANTOVA, and were considered significantly different at  $P < 0.05$ . Differences between means were compared using Fishers Protected Least Significant Difference Test.

## Results

Significant differences were found between birds ( $P < 0.001$ ) in relation to the proportion of feathers removed from substrate within 2 h of presentation on all days. Approximately 49.5% of the birds showed a strong interest in presented feathers, based on all feathers being removed from the substrate within 2 h on day 1. This proportion increased to 63.4% on day 17. Significant differences in the proportion of feathers removed were also observed between trial days with 55.7% feathers removed on day 1 of the trial, increasing to 76.2% by day 17 (Table 1).

Figure 1 shows the segregation of the population of birds based on the varying levels of feather removal as the trial progressed. The proportion of birds that did not remove feathers was initially high, at 35.6% on day 1, but decreased to 9.4% on day 8, where levels remained low for the remainder of the trial. In contrast, the proportion of birds removing all presented feathers

## *"This study identified feather peckers and feather eaters in the flock based on behavioural differences of individual birds"*

Feathers were presented in the form of 4-6 cm long semi-plumes and downy feathers placed in the holes in the plastic lids and suspended in front of the cages on days 1, 3, 5, 8, 10, 15, and 17 of the trial. Hens were initially presented with 5 feathers, but this was later increased to 10 feathers for the final 3 days when feathers were presented in the trial. The number of feathers remaining on the substrate was counted after 30 minutes, 1 hour, and 2 hours. A total of 59 birds were randomly selected and individually observed upon presenting the feathers. Latency to peck, and number of pecking bouts, feathers pecked, feathers pulled and feathers eaten were recorded.

increased gradually from 49.5% on day 1 to 63.4% on day 17 of the trial.

The average feather removal was significantly different ( $P < 0.001$ ) with 7.2 feathers removed within the first 30 min of presentation of feathers, compared to only 0.330 feathers between 30 min and 1 h, and 0.287 feathers between 1 and 2 h. Sixty-six percent of birds ate at least one feather during the period of observation and these birds were classified as feather eaters (FE). Both the number of FE birds and the average number of feathers eaten on any given day increased linearly as the trial progressed (Figure 2).

**Table 1** - Mean proportion of feather removal 2 hours after feather presentation and the proportion of birds removing none or all of the feathers after 2 hours.

| Trial day                 | 1                  | 3                   | 8                   | 10                 | 15                 | 17                 | SEM   | P value |
|---------------------------|--------------------|---------------------|---------------------|--------------------|--------------------|--------------------|-------|---------|
| Mean feathers removed (%) | 55.74 <sup>c</sup> | 63.86 <sup>Bc</sup> | 73.27 <sup>ab</sup> | 80.00 <sup>A</sup> | 76.98 <sup>A</sup> | 76.19 <sup>A</sup> | 0.026 | < 0.001 |

<sup>a,b,c</sup> Means in a row not sharing a common superscript are significantly different. Means compared using Fishers Protected Least Significant Difference test

# MANAGEMENT



## Discussion

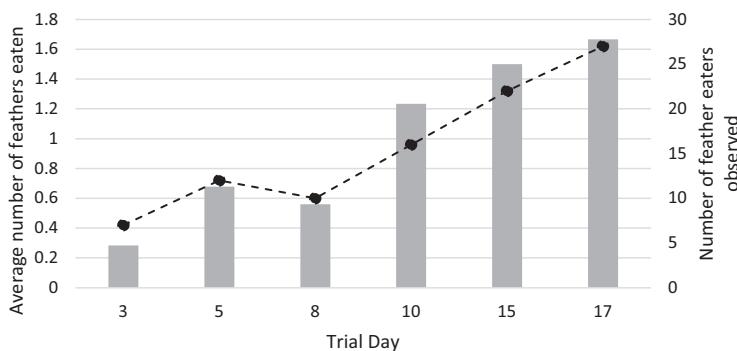
This study identified feather peckers and feather eaters in the flock based on behavioural differences of individual birds. Significant differences between birds are expected due to the variability of hen behaviour. The higher proportion of birds removing no feathers on days 1 and 3 compared to subsequent days indicates neophobia to the presented feathers. However, as the number of birds removing no feathers decreased substantially on day 17, and remained low for the remainder of the trial, it is likely that neophobia was overcome. In contrast, the proportion of birds removing all feathers from substrates after 2 h increased as the trial progressed.

The proportion of feathers removed in 30 min, 1 h and 2 h was significantly different. Based on the average number of feathers removed within each time period, it is clear that the majority of feathers were removed within the first 30 min, with an average difference of <1 feather between 30 min and 1 h, as well as 2 h. This is likely due to the majority of birds removing all feathers

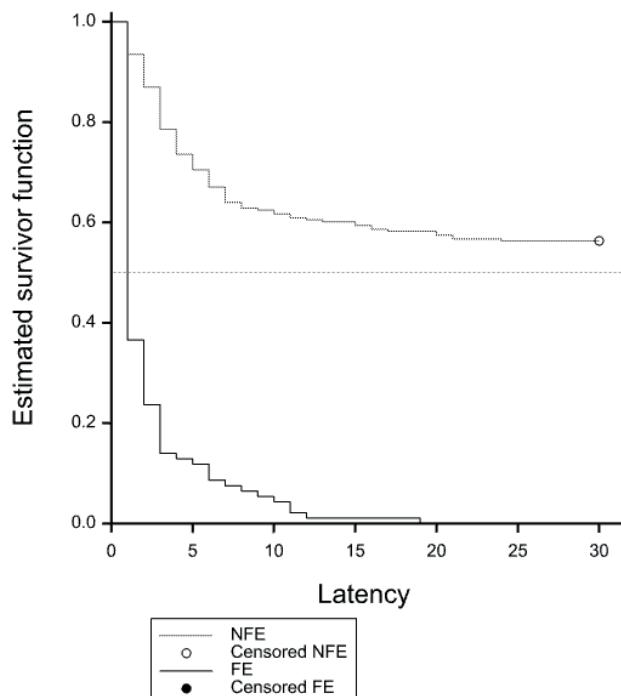
***“Differences in feather interest and feather appetite between individual birds, as well as the latency to peck for known feather eaters (FE) and non-feather eaters (NFE) birds, provides a behaviour-dependant basis for selection of FE and NFE birds in future research”***

Comparison of survival curves using non-parametric tests indicated that latency to peck at feathers was significantly lower ( $P < 0.001$ ) for FE birds than NFE birds (Figure 3).

The probability of FE birds to peck at the feathers was 0.5 after 1 s of presentation, and 0.75 after 2 s. In contrast, the probability of NFE birds to peck at feathers after 4 s of presentation was 0.25. Similarly, when considering latency to peck across all birds, significant differences were also found between days with latency to peck decreasing as the trial progressed ( $P < 0.05$ ).



**Figure 2** - The average number of feathers eaten per hen per day within a 30 s period for individually observed birds ( $n = 59$ ) is plotted as columns (primary y-axis), and the total number of birds observed to eat at least one feather during observation periods is shown as a dashed line (secondary y-axis).



**Figure 3** - Plot of Kaplan-Meier curve of survivor function estimate of latency to peck (s) of non-feather eating (NFE) and feather eating (FE) birds.



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on the substrate, leaving no feathers for subsequent time periods, as well as a general disinterest in the feathers after 30 min.

Depending on the size and orientation of the feather, McKeegan and Savory (2001) observed that feathers typically required manipulation before being swallowed by the bird. In this trial, feather manipulation sometimes resulted in birds dropping feathers. In these cases, feathers were either picked up by the same bird, picked up by a bird in an adjacent cage, or feathers fell through the cage floor which probably affected the ability of the bird to eat the feathers.

Behaviour observations indicate that feather eating is not the final outcome for all pecked feathers. Although strong interest in feathers was observed, it is unlikely that all birds tested and selected for future trials will be feather eaters. However, due to the limited number of birds observed, the possibility that some unobserved birds are feather eaters cannot be ruled out.

Differences in feather interest and feather appetite between individual birds, as well as the latency to peck for known FE and NFE birds, provides a behaviour-dependant basis for selection of FE and NFE birds in future research. Feather pecking and feather eating birds will be used in subsequent trials to determine potential nutritional motivations behind feather eating behaviour.

**Acknowledgments:** The Authors would like to thank the Poultry CRC for funding the project and the Poultry Research Foundation for technical support.

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## Poultry house environmental control during cold weather

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Extension Poultry Scientist  
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USA

*Most people think minimum ventilation is all about air quality but that is only partially correct. First one should define good air quality. Many people define good air quality as ammonia ( $NH_3$ ) concentrations of 25 ppm or less, carbon dioxide ( $CO_2$ ) concentrations of 5,000 ppm or less and carbon monoxide ( $CO$ ) concentrations of 50 ppm or less.*

Minimum ventilation rates are not based on oxygen concentration. However, the main purpose of minimum ventilation is moisture control, which can be achieved by maintaining relative humidity (Rh) levels between 40 and 60%. NH<sub>3</sub>, CO<sub>2</sub> and Rh concentrations are positively correlated, meaning that if Rh is high then NH<sub>3</sub>, and CO<sub>2</sub> will be elevated as well (*Figure 1*).

Maintaining Rh between 40-60% increases the probability that the NH<sub>3</sub> and CO<sub>2</sub> gases will be less. So maintaining Rh in this range is a good form of prevention. In many cases prevention is cheaper than having to correct a problem. For example, it is usually more expensive to ventilate a house enough to reduce high ammonia than it would have been to just prevent it with optimum minimum ventilation rates and litter treatments. Litter moisture will increase as the relative humidity exceeds 60%.



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*"It is in the best interest of bird health, performance and welfare to strive for optimal litter and air quality during the flock grow-out period. There are some key options that can be utilized to limit litter moisture accumulation and contribute to an environment that will allow the broiler to reach its full genetic potential"*

Consequently litter quality deteriorates as litter moisture increases. Litter moisture can increase for a number of reasons which include but are not limited to increased bird stocking density, coccidiosis, necrotic enteritis, diet, inadequate ventilation, insufficient litter volume, and improper drinker management. Lousy litter quality leads to increased ammonia and increased numbers of bacteria including pathogens. Increased litter moisture also leads to increased incidence and severity of footpad dermatitis, which not only can be an economic loss but is also a factor considered in animal welfare audits.

It is in the best interest of bird health, performance and welfare to strive for optimal litter and air quality during the flock grow-out period. Below are options that can be utilized to limit litter

moisture accumulation and contribute to an environment that will allow the broiler to reach its full genetic potential.

### Bedding depth

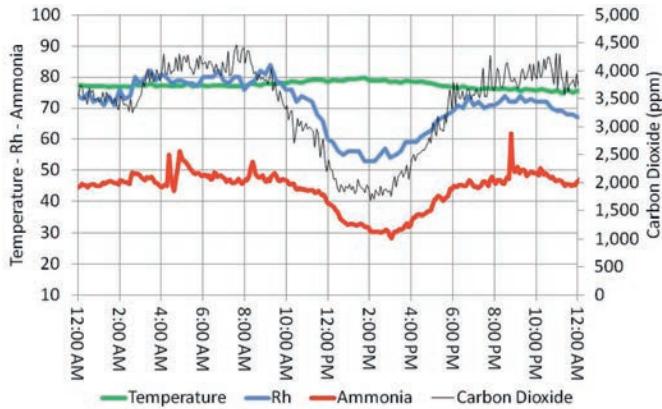
Whether it is fresh bedding or used litter a minimum of three inches is required. As bird density increases or if the farm has a history of litter quality issues, a litter depth greater than three inches should be utilized. The litter depth/volume will need to increase with bedding materials that do not absorb moisture as well such as peanut hulls and rice hulls. Typically, the bedding in houses that use these materials is deeper than those that utilize pine shaving or sawdust. Used litter, when properly managed and worked between flocks serves as good litter base but

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**Figure 1** - House temperature, relative humidity (Rh), carbon dioxide and ammonia levels in a house with 21 day old birds (December).

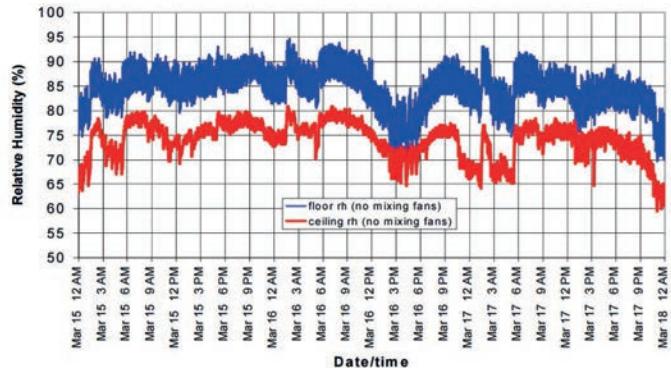
also has a smaller margin of error when it comes to managing for ammonia generation and Rh.

## Circulation fans

A circulation fan system is used not only to break up temperature and Rh stratification (*Figures 2 and 3*) and create more uniform conditions from one end of a house to the other but just as importantly is used to increase the level of air movement over the litter to aid in moisture removal. A proper circulation fan system moves air gently across the litter, producing air speeds generally in the neighborhood of 50 ft/min so as not to cause detrimental drafts on the birds. Circulation fans should operate continually to maximize moisture removal but may need to be temporarily turned off when the timer fans operate if they interfere with the air circulation pattern created by the air inlets.

## Attic inlets

Though attic inlets can lead to slightly lower heating costs, the primary objective of an attic system is to maximize fan runtime,

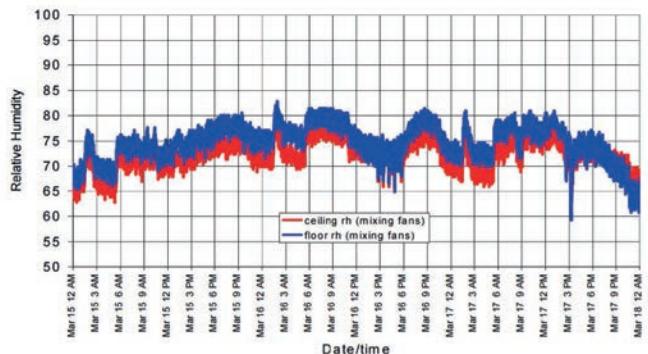


**Figure 2** - Relative humidity of the air at floor and ceiling level in house without circulation fans.

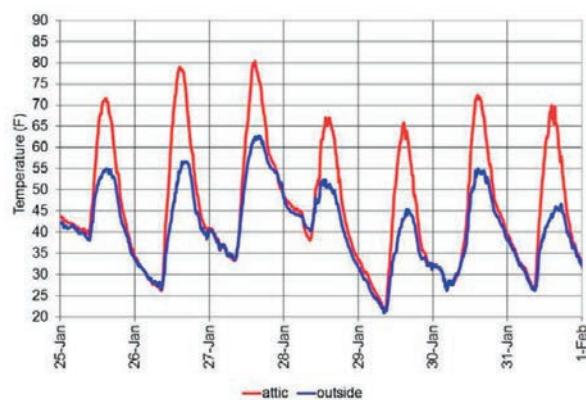
which will lead to improved air quality and litter conditions. Attic inlets tend to do a better job of conserving heat in a poultry house than conventional side wall inlets. First, fresh air is introduced at the peak of the ceiling where the warm air produced by the heating system and birds tends to collect. Second, air entering through an attic inlet moves parallel to the ceiling and not parallel to the side wall which tends to maximize the distance the air travels along the ceiling (*Figures 2 and 3*). The longer the air travels along the ceiling the more it will heat up and dry out. Last but not least, during the daylight hours, drawing heated air out of the attic at a minimum reduces house cooling and sometimes leads to increased house temperatures causing more fans to operate. The combination of all these factors has been shown to increase the amount of air brought into a house by 20% or more compared to a house using sidewall inlets (*Figure 4*).

## Ventilation rates

Keeping litter dry is a significant challenge in broiler houses. For every pound of feed a broiler eats it will drink roughly one quart of water, 80% of which will end up in the litter and the



**Figure 3** - Relative humidity of the air at floor and ceiling level in house with circulation fans.



**Figure 4** - Chart illustrates the increase ventilation (20%) that can be achieved with attic inlets without increased fuel costs

# MANAGEMENT

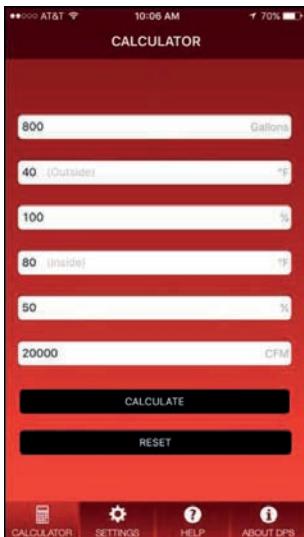


Figure 5 - Example of input values.

air in the house. To prevent the buildup of moisture in the litter, producers must constantly exchange the moist inside air in a poultry house with the relatively dry outside air. If a producer can remove the same amount of moisture from their houses each day that the birds are adding, they can maintain a constant moisture level in their houses and problems associated with excessive moisture will be minimized.

Recently an app titled "CHKMINVENT" was developed for the iPhone to enable producers to determine how much they need to ventilate their houses during cold weather to remove the moisture their birds have added on a daily basis. The app utilizes psychometric equations to determine the air exchange rate required to remove a user-specified amount of water based on inside/outside conditions (Figure 5).

## The information required by the CHKMINVENT app includes:

- Inside temperature (F/C)
- Inside relative humidity (%)
- Outside temperature (F/C)
- Outside relative humidity (%)
- Amount of water needed to be removed from the house (Gallons/Liters)

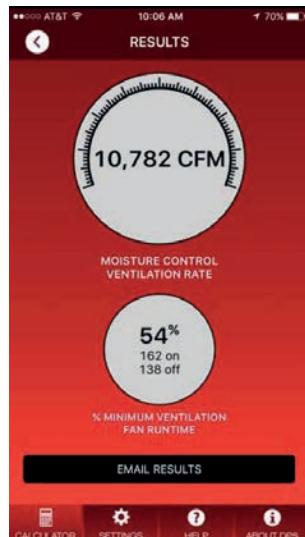


Figure 6 - Calculated moisture balance moisture balance ventilation rate.

- Total air-moving capacity of minimum ventilation fans to be used (cfm/cmh)
- From this the App will calculate a moisture balance ventilation rate as well as a minimum ventilation fans runtime (% runtime as well as seconds on/off out of 300) (Figure 6).

This app can be used to confirm whether ventilation rates currently being used are adequate to remove the moisture being added according to water usage by the birds. However, nothing is full proof so house conditions such as Rh and litter condition should continue to be monitored. A combination of the app, Rh and litter conditions can be used to fine-tune the minimum ventilation rates as needed. More information on the app and its use can be found by at: [www.poultryventilation.com](http://www.poultryventilation.com).

It is important to monitor relative humidity and bird water usage each day and adjust the ventilation rate as needed. The combination of the options discussed in this paper can lead to an optimum environment in modern poultry houses so that broilers can achieve their full potential.

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## Effect of production system and flock age on egg quality

*Egg quality parameters were measured in eggs from flocks reared together and then allocated to different production systems. Eggs were processed for measurement of egg quality variables, scoring of ultrastructural mammillary layer features, completeness of cuticle cover and protoporphyrin IX (PPIX) quantification.*

In Australia, the cage production system is the most efficient and cost effective and accounts for approximately 53% of the total table egg production (AECL, 2014). Modern cage production systems typically consist of multiple tiers of cages installed in environmentally controlled poultry houses. According to the current Australian Model Code of Practice (Primary Industries Standing Committee, 2002), a minimum floor space allowance of 550 cm<sup>2</sup> per hen must be provided for 3 or more birds per cage where the birds weigh less than 2.4 kg. The barn system offers access to a deep litter system, automated feeding and drinking systems, perches, and stepping rails to automated egg collection nest boxes.

The barn egg production system's average contribution to total eggs produced in Australia is about 8% (AECL, 2014). Free range production is increasing in Australia, reported as being 38% of total egg production. In a typical free range production system, nest boxes, perches, feed and water are available in the house which is often very similar to the barn production system but birds also have access to a grassed outdoor free-range area.

The main objective of the present study was to compare the performance of pullets reared together and then placed into three commercial production systems at different flock ages for traditional egg quality measurements, scoring of

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mammillary layer ultrastructural features, cuticle estimation and protoporphyrin IX quantification from eggshell.

## Materials and methods

Eggs were collected from Hy-Line brown egg laying flocks housed in conventional cage, barn and free range commercial production systems. Each of the three flocks was sampled at 44, 64 and 73 weeks of age. Each flock had pullets which were reared together and then allocated to one of cage, barn or free range commercial production systems.

From each flock, at each age, 30 eggs were processed for the measurement of traditional eggshell and egg internal quality variables, 30 for estimation of the completeness of cuticle cover and 30 for the amount of PPIX present in the shell. From the eggshells used for traditional quality measurements, 10 eggshells from each age group were randomly selected and processed for the scoring of eggshell mammillary layer ultrastructural features.

**Table 1 - Effect of production system and flock age on eggshell and egg internal quality variables**

| Variable           | Production System      |                        |                        | Flock Age (week)       |                        |                        | P Value |         |
|--------------------|------------------------|------------------------|------------------------|------------------------|------------------------|------------------------|---------|---------|
|                    | Cage                   | Barn                   | Free range             | 44                     | 64                     | 73                     | P       | A       |
| Shell reflectivity | 25.2±0.46 <sup>c</sup> | 29.7±0.66 <sup>a</sup> | 27.3±0.62 <sup>b</sup> | 24.3±41 <sup>c</sup>   | 27.3±0.48 <sup>b</sup> | 30.7±0.72 <sup>a</sup> | <0.0001 | <0.0001 |
| Egg weight         | 60.6±048 <sup>a</sup>  | 58.6±0.41 <sup>b</sup> | 61.2±0.54 <sup>a</sup> | 59.1±0.39 <sup>a</sup> | 60.0±0.52 <sup>b</sup> | 61.4±0.53 <sup>a</sup> | 0.0003  | 0.0031  |
| Shell weight       | 5.5±0.05               | 5.5±0.07               | 5.7±0.0                | 5.6±0.04               | 5.6±0.07               | 5.5±0.06               | 0.0765  | 0.3585  |
| Shell Percentage   | 9.2±0.09               | 9.3±0.09               | 9.2±0.08               | 9.5±0.07 <sup>a</sup>  | 9.2±0.1 <sup>b</sup>   | 9.0±0.09 <sup>b</sup>  | 0.2652  | 0.0002  |
| Shell thickness    | 406.5±2.8              | 408.8±3.6              | 408.2±3.2              | 414.9±2.6 <sup>a</sup> | 405.5±3.5 <sup>b</sup> | 403.0±3.4 <sup>b</sup> | 0.8780  | 0.0221  |
| Breaking strength  | 40.5±0.88              | 41.7±1.02              | 40.0±0.89              | 44.1±0.72 <sup>a</sup> | 38.4±0.99 <sup>b</sup> | 38.8±0.91 <sup>b</sup> | 0.3705  | <0.0001 |
| Deformation unit   | 262.8±4.07             | 279.4±10.3             | 274.9±8.69             | 284.1±3.71             | 265.0±8.79             | 267.9±10.3             | 0.3293  | 0.2030  |
| Albumen height     | 6.1±0.14 <sup>b</sup>  | 6.1±0.15 <sup>b</sup>  | 7.4±0.15 <sup>a</sup>  | 7.1±0.15 <sup>a</sup>  | 6.4±0.15 <sup>b</sup>  | 6.1±0.17 <sup>b</sup>  | <0.0001 | <0.0001 |
| Haugh unit         | 76.9±1.04 <sup>b</sup> | 77.4±1.2 <sup>b</sup>  | 85.5±1.43 <sup>a</sup> | 83.9±0.98 <sup>a</sup> | 79.4±1.03 <sup>b</sup> | 76.4±1.75 <sup>c</sup> | <0.0001 | <0.0001 |
| Yolk color         | 10.8±0.11 <sup>a</sup> | 10.6±0.09 <sup>a</sup> | 9.9±0.19 <sup>b</sup>  | 10.7±0.11 <sup>a</sup> | 10.8±0.08 <sup>a</sup> | 9.8±0.18 <sup>b</sup>  | <0.0001 | <0.0001 |

P = Production system; A = Flock age; Values are Means ±SE

a,b,c Across a row, values with different superscripts among different production systems and age groups are significantly different ( $P < 0.05$ )

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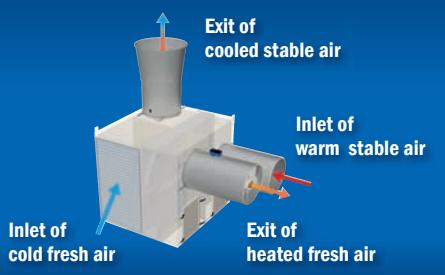


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were sputter coated in a Neocoater for 8 minutes, and viewed under the SEM at magnifications of 22~500. Mammillary ultrastructural variables were scored following the method of Solomon (1991).

## Cuticle cover estimation

For the cuticle cover estimation, shell color ( $L^*a^*b^*$ ) was measured on eggs before staining, using a Konica Minolta spectrophotometer. Eggs were soaked in MST cuticle blue stain for 1 minute, and rinsed in tap water to remove excess stain. Shell color was measured again on stained eggs. The amount of cuticle was estimated by converting the  $L^*a^*b^*$  values into a single score. The single score, measures the  $L^*$ ,  $a^*$ , and  $b^*$  values, before and after staining, and calculates a single value as  $\Delta E^*_{ab}$ :

$$\Delta E^*_{ab} = J[(\Delta L^*) + (\Delta a^*y + (\Delta b^*)^2)]$$

A higher  $\Delta E^*_{ab}$  denotes a higher staining affinity and hence more cuticle coverage (Roberts et al., 2013).

## Protoporphyrin IX quantification from eggshell with and without cuticle

Shell reflectivity and shell color were measured on eggs before and after removal of the cuticle layer. From the  $L^*a^*b^*$  values, only  $L^*$  values were used for analysis, as this reflectance component indicates the difference between light and dark brown shell color. Each egg, individually, was soaked in an EDTA solution (0.34 M, pH 7.5) for 5 minutes, and the cuticle of the soaked longitudinal half side of the shell was carefully scrubbed off using a soft brush in running tap water. Shells were cut longitudinally into two halves (with and without cuticle). Shell membrane was removed manually and shells were allowed to dry thoroughly. Shell reflectivity and shell color ( $L^*$ ) were measured again on the shells with cuticle removed.

A 0.25 g sample was weighed into a 10 mL centrifuge tube into which 4 mL of methanol - HCl (2:1) solvent was added. The tubes were stored for 3 hours to allow the shell pieces to digest completely. The tubes were centrifuged at 800xg at 4°C for 0.5 hour, the supernatant solution was decanted into 4 mL cuvettes, and the absorbance of the supernatant read at 412 nm in a spectrophotometer.

**Table 2** - Effect of production system and flock age on protoporphyrin IX ( $mM \times 10^{-8}$ )

| Variable                      | Production System |                   |                   | Flock age (week)  |                   |                   | P value |         |
|-------------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|---------|---------|
|                               | Cage              | Barn              | Free range        | 44                | 64                | 73                | P       | A       |
| PPIX in whole shell           | 9.49 <sup>a</sup> | 8.24 <sup>b</sup> | 8.64 <sup>b</sup> | 9.35 <sup>a</sup> | 8.74 <sup>b</sup> | 8.26 <sup>b</sup> | <0.0001 | 0.0001  |
| PPIX in shell without cuticle | 7.90 <sup>a</sup> | 6.90 <sup>b</sup> | 7.28 <sup>b</sup> | 7.84 <sup>a</sup> | 7.36 <sup>b</sup> | 6.88 <sup>c</sup> | <0.0001 | <0.0001 |
| PPIX in cuticle               | 1.60              | 1.35              | 1.36              | 1.51              | 1.38              | 1.42              | 0.1603  | 0.6284  |

<sup>a,b,c</sup> Across a row, indicates significant difference among different production systems and age groups;

P = Production system; A = Age; PPIX is protoporphyrin IX

Data were subjected to one-way and two-way analysis of variance (ANOVA) and General Linear Model (GLM) Procedures (SAS 2004) to test the probability of significant differences between the means values. The Level of Significance was assumed at 95% confidence ( $P < 0.05$ ).

## Results

### Eggshell and egg internal quality measurements

For eggshell quality measurements, there was a significant difference among production systems for shell reflectivity and egg weight. However, there was no statistically significant difference in the other parameters of shell weight, shell percentage, shell thickness, shell breaking strength and deformation unit (Table 1). The egg internal quality was significantly affected ( $P < 0.0001$ ) by production system. The hens kept in cage and barn production systems laid eggs of darker yolk color as compared with the free range system.

### Microscopic observations of the shell mammillary layer ultrastructure

For all the sixteen ultrastructural variables of mammillary layer observed, there was a significant main effect ( $P > 0.05$ ) of production system on the variability of mammillary cap size, amount of confluence, incidence of early fusion, changed membrane and erosion.

The mammillary caps arise from the deposition of calcium carbonate into the membrane fibres such that the shell membranes are attached to the mammillary caps. Confluence results from the attachment of the mammillary caps to each other, thus forming a smooth blanket on the surface of the mammillary caps. A high incidence of early fusion increases the bonding strength between mammillary cones and has a positive effect on mammillary layer strength.

Flock age significantly affected the incidence of early fusion and changed membrane. The interaction between production system and flock age was only significant for mammillary cap size, early and late fusion. The variability of the mammillary cap size increased with increased flock age and was significantly higher in cage eggs. The amount of confluence was significantly higher in cage eggs compared with barn and free range eggs. The incidence of alignment was not significantly different

among production systems and its values were closer to "isolated" on the measuring scale. Other variables like type A bodies, type B bodies, aragonite, cubic, depression and erosion varied with production system and flock age.

### Cuticle cover estimation

When the completeness of cuticle cover was estimated by the single score value ( $\Delta E^*_{ab}$ ), the barn production system showed a significantly higher amount of cuticle cover compared with cage and free range eggs. There was no significant interaction between production system and flock age for the amount of cuticle present on the eggshell surface. Comparing the three flock ages, the 44-week flock eggs had a significantly higher amount of cuticle followed by 73, whereas 64- and 73- week flocks were not significantly different.

### Shell Reflectivity and L\* measurements

There was a significant effect of production system and flock age on shell reflectivity and L\* values measured on shells with and without cuticle. When the values of shell reflectivity and L\* were compared among different age groups, the 44-week flock eggs had significantly lower values and the values increased linearly with increasing flock age. A similar pattern was observed for shells from which cuticle layer had been removed.

### Protoporphyrin IX quantification from shell with and without cuticle

There was a significant main effect ( $P > 0.05$ ) of production system and flock age, but no significant interaction between the two, on the amount of PPIX in 1 g of whole eggshell and eggshell from which the cuticle layer had been removed (Table 2). Eggs from the cage production system showed significantly higher amounts of PPIX in 1 g of whole eggshell and shell without cuticle. The amount of PPIX in 1 g of whole eggshell was significantly higher in eggs from the 44-week flock, followed by the 64 and 73-week old flocks.

### Discussion

There was a significant main effect ( $P < 0.05$ ) of production system on shell reflectivity, egg weight and egg internal quality and significant effects of flock age on most measurements. The mammillary layer ultrastructural variables showed no clear relationship with production system and flock age. Cuticle cover was significantly higher in barn eggs, followed by free range and cage eggs. Completeness of cuticle cover was significantly higher in eggs from the 44 week old flock than for 64 week and 73 week old flocks. There was a significant main effect of both production system and flock age, but no significant interaction for shell reflectivity, L\* and amount of PPIX. There was no statistically significant difference for cuticle cover.

In one gram of shell with and without cuticle, there was more protoporphyrin IX in cage eggs followed by free range and barn eggs. Similar trends were recorded for the amount of PPIX in 1 g of cuticle, but the difference was not statistically significant. The amount of PPIX decreased significantly with increasing flock age.

When birds reared together were allocated to different production systems prior to the onset of lay, there were relatively few differences among production systems for egg quality measurements. Eggs from the cage production system had darker shell colour and contained more protoporphyrin mainly within the calcareous part of the shell.

For the barn production system, the completeness of cuticle cover was higher and egg weight generally lower. The differences in egg quality in relation to flock age are similar to those reported previously.

**Acknowledgments:** This study was supported by funding from Australian Egg Corporation Limited.

### References are available on request

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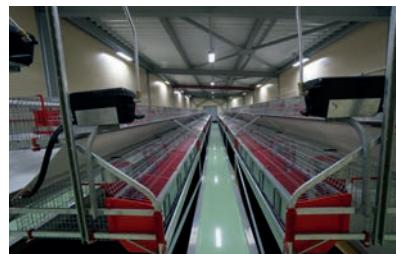
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## Role of diet on odour emissions from meat chickens

*Abatement of odour emissions has become an important consideration to agricultural industries, including poultry production. Odours are generated in meat chicken houses primarily from the microbial decomposition of faecal matter in chicken litter as well as directly from the birds. In recent years, odour emissions have become a growing concern to poultry producers.*

*In order to study the link between diet and odour emissions, an experiment was conducted using twelve Ross 308 broiler chickens.*

*At the age of 22 days, birds of uniform body weight were selected from a total of 288 male birds, adapted to metabolic chambers for six days and fed their respective diets for 15 days. Two treatments were compared using three replicates of two birds per chamber. The two wheat-soy diets were formulated according to the 2007 Ross 308 nutrient specifications for digestible amino acids but they differed in ingredient composition and metabolisable energy content.*

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Investigations by Murphy *et al.* (2014) and Dunlop *et al.* (2011) resulted in a comprehensive list of odorous chemical compounds that are of interest to the poultry industry. Murphy *et al.* (2014) identified eight major volatile organic compounds from tunnel ventilated meat chicken sheds that were considered as important predictors of odour. These were dimethyl sulphide (DMS), dimethyl trisulphide (DMTS), 2-3 butanedione, 3-methyl-butanal, 1-butanol, 3-methyl-1-butanol, 3-hydroxy-2-butanone (acetoin), and 2-butanone.

In an effort to address odour issues from poultry farms, there have been attempts to develop mitigation strategies including litter treatments, biofilters, neutralising agents, air scrubbers, ozone treatment, windbreak walls and short stacks but these techniques are generally costly or impractical due to the required high ventilation rates in poultry farms. There is little information available linking diet composition to odour emissions.

Diets can be formulated to more closely meet the bird's nutritional requirements to avoid overfeeding and to reduce excretion of undigested components. This will decrease the available substrates that the microbes metabolise to odour compounds. A real time odour measuring device, such as the Fourier transform infrared (FTIR) spectrometer, is feasible to measure odorous gases and quantify their individual constituents. Van Kempen *et al.* (2002) and Witkowska (2013) successfully used FTIR to detect and quantify odours from swine and turkey houses respectively. The objective of this study was to use FTIR to compare odour emissions from broilers fed two diets differing in ingredient and nutrient composition.

## Materials and methods

At the age of 22 days, 12 birds of uniform body weight were selected from a pool of 288

Ross 308 male broilers. The birds were adapted to the metabolic chambers for six days in a climate controlled room and

fed their respective test diets. The experiment started when the birds were 28 days of age and finished on day 42. Feed and water were provided *ad libitum*. Each diet was replicated three times with two birds per chamber.

Two wheat-soy diets were formulated according to the Ross 308 nutrient specifications for digestible amino acids (Aviagen, 2007). Diets were isonitrogenous but differed in ingredient composition and ME (Diet A with 60 g / kg canola and no corn, 13.39 MJ / kg ME; Diet B with 150 g / kg corn and no canola, 12.90 MJ / kg ME). Diets were analysed for nutrients including sulphur content.

A Fourier transform infrared (FTIR) spectrometer (Gasmet™ Model DX-4015, Gasmet Technologies, Finland) was calibrated for 30 compounds previously reported as odorants or volatile compounds from poultry production facilities. Gaseous samples were collected at day 42 from the excreta in the presence of birds. Chamber lids were closed for approximately 20 min before sample collection. Water was used to seal the chambers. At that time there was zero air exchange and odorants were allowed to concentrate prior to sampling.

Carbon dioxide and oxygen levels inside the chambers were recorded during the period of closure.

The emissions from the chambers were analysed using FTIR with the following set- up: flushing time- 30s, pumping time-1 min, measuring time-3 min. The gas samples were drawn at a flow rate of 2 L/min with the in-built FTIR instrument pump (i.e. 2 litres of gases were measured from each chamber). After sampling, the FTIR was flushed with pure nitrogen for 15 min before taking measurements for the next group. After the measurements, quantitative analysis was conducted in a laboratory with the use of Calcmet Professional software with a library of reference spectra for 30 gases.

The data were analysed by one-way ANOVA using the GLM procedure (SAS Institute Inc., Cary, NC). Differences were determined using the t-test. Variability in the data is expressed as

**Table 1 - Odorous compounds emitted from meat chickens fed two commercial diets**

| Compounds                    | Diet A, ppm (+canola seed) | Diet B, ppm (+corn) | SEM   | P-value |
|------------------------------|----------------------------|---------------------|-------|---------|
| 2, 3-butanedione/diacetyl    | 1.099 <sup>b</sup>         | 2.307 <sup>a</sup>  | 0.286 | 0.005   |
| 2-butanone                   | 0.923                      | 0.704               | 0.157 | 0.548   |
| Dimethyldisulfide            | 3.242                      | 3.079               | 0.154 | 0.651   |
| Methanethiol/methylmercaptan | 19.393 <sup>a</sup>        | 15.607 <sup>b</sup> | 0.940 | 0.014   |
| 2-butanol                    | 0.000                      | 0.344               | 0.109 | 0.116   |
| 3-methyl-butanal             | 0.317                      | 0.496               | 0.166 | 0.645   |
| Phenol                       | 0.880 <sup>b</sup>         | 0.981 <sup>a</sup>  | 0.026 | 0.027   |
| m-cresol                     | 0.582 <sup>b</sup>         | 1.051 <sup>a</sup>  | 0.112 | 0.006   |
| Excreta moisture, %          | 76.20 <sup>a</sup>         | 68.25 <sup>b</sup>  | 1.530 | 0.035   |

<sup>a,b</sup> Means in the same row with different superscripts differ ( $P < 0.05$ ) or ( $P < 0.01$ )



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the standard error means (SEM) and a probability level of  $P < 0.05$  was considered to be statistically significant.

## Results and discussion

The total duration allocated for concentration and measurement of odour (i.e. 25 min) was sufficient to capture the volatiles with acceptable levels of  $\text{CO}_2$  and  $\text{O}_2$  (< 2% and > 18%, respectively) inside the chambers. The temperature and humidity at the time of sampling were recorded as 22–23°C and 75%, respectively. Since FTIR provides results in parts per million (ppm), slight changes in temperature or humidity at sampling would not affect results. Altogether 24 volatile organic compounds were detected and quantified. Out of these, the eight considered as key odorants are listed in Table 1. The first six compounds are considered to be important poultry odorants. Two important odour predictors not detected by FTIR were dimethylsulfide and dimethyltrisulfide but methylmercaptan was detected. The mercaptans are unstable and can be oxidized easily to other sulphur compounds upon storage or during carbon capture sampling techniques using a thermal desorption analysis process. Murphy *et al.* (2014) used a thermal desorption process to quantify the volatiles and it is possible that the mercaptans were oxidized to dimethylsulfide and dimethyltrisulfide in those studies.

In the current study, very short sampling intervals were used, thus it could be stated with reasonable confidence that the mercaptans did not have time to oxidise into other sulphur

compounds. The concentration of methylmercaptan therefore might have been higher and oxidised products lower in this study than previously reported. Mercaptans and thiols are considered to be important odorants in animal production facilities.

Methylmercaptan, dimethyldisulfide, 2,3-butanedione, phenol and m-cresol were measured at higher concentrations than the odour detection threshold. Methylmercaptan has a rotten cabbage smell (Merck Index, 1968) and was produced at higher levels ( $P < 0.05$ ) from broilers fed Diet A. Dimethyldisulfide was also detected in chamber air from both diets but the concentration of the total sulphur compounds was higher with Diet A. These results suggest that the use of 60g/kg canola seed led to a higher level of sulphur containing odorants compared to Diet B that did not have canola seed. The calculated digestible methionine plus cysteine were similar in both diets (7.3 g / kg vs. 7.0 g / kg). This small difference in dietary sulfur amino acid levels is unlikely to produce differences in odour among the treatments. However, a higher excreta moisture content was observed in chambers from birds fed Diet A ( $P < 0.05$ ). The litter moisture content does not correlate with odour emissions but correlates with odorant characteristics. Increased litter moisture is associated with higher concentrations of organosulfides, aldehydes, and alcohols due to increased anaerobic degradation. Therefore, the higher organosulfide emission from Diet A in this study may be related to a higher excreta moisture content.

Diacetyl (2, 3-butanedione) has a rancid butter smell. This compound was produced at higher levels ( $P < 0.01$ ) in chambers



from broiler fed Diet B than those fed Diet A. Diacetyl is considered an important chicken odorant due to its relatively low human detection threshold. Diacetyl is a product of fermentation. Future investigations of intestinal metabolites and microbiota may help elucidate the mechanism by which diet composition influences diacetyl production. Diet B produced higher levels of phenol ( $P < 0.05$ ) and m-cresol ( $P < 0.01$ ) compared to Diet A. These compounds are considered to be strong odorants by some researchers and weak odorants by others. Phenol originates from the microbial degradation of tyrosine in the intestinal tract of animals and from phenolics contained in litter.

## Discussion

This study clearly showed that diet has an impact on odours produced from broiler chickens. Using closed circuit metabolic chambers coupled with FTIR allowed for accurate detection and quantification of the odorous compounds that are of interest to the poultry industry. Minor changes in diet composition were found to change the relative abundance of gases associated with odours. Further investigation is warranted to more fully understand the effect of microbial metabolism of nutrients and metabolites in the gut and litter on odour formation.

**Acknowledgements:** The authors are grateful to the financial support of the Poultry CRC and to Dr. Gavin Parcsi (UNSW) and Mark Dunlop (QDAFF) for their valuable suggestions.

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# The challenges and future of diagnostics in poultry medicine

*A review of the past helps to frame the future of poultry diagnostics. In the United States, the Land-Grant universities provided a fertile environment for faculty members in the early 20<sup>th</sup> century to apply the scientific method to poultry disease diagnosis. A natural affiliation and partnership of the USDA Bureau of Animal Industry and state departments of agriculture developed with these fledgling diagnostic programs. A mutual need existed for reagents and testing to support programs for tuberculosis, hog cholera, and brucellosis, among others. For poultry, this era marked the launch of the National Poultry Improvement Plan (NPIP) in the 1930's to control Pullorum disease.*



an Pathologists (AAAP) that same year. Through the 1960s and 70s, state and federal diagnostics were substantially moved from research laboratories into dedicated facilities. Their core mission was disease regulation, but general diagnostic services were provided for animal industries, including poultry. Private sector laboratories also were in operation during this time, some as technical service extensions of pharmaceutical and biological companies serving the poultry sector.

The state and university diagnostic laboratories were also the first line of support for the emerging field of companion animal medicine. A natural tension often developed within this setting because of finite diagnostic resources need to cover regulatory programs, animal industries, and companion animals as well as public health, wildlife and various other added services and programs. This was accentuated by the common practice of offering free or reduced-cost diagnostic services for agriculture to increase passive surveillance for regulated diseases. In this competitive environment, the amount of resources dedicated to poultry, state-by-state, was often contingent upon poultry producers who could leverage the largesse of university administrators and state agricultural authorities. Animal disease emergencies (Pullorum disease, chronic respiratory disease, tuberculosis, Newcastle disease, influenza, chronic wasting disease, concerns of agroterrorism) often provided the stimuli for renewed investment in diagnostic services for the animal industries.

The poultry diagnostic expertise in the Land-Grant universities led to the characterization of important poultry diseases in the years leading up to World War II: Newcastle disease, infectious bronchitis, mycoplasmosis, coccidiosis and others. The first printing of Diseases of Poultry appeared in 1943. Parallel to these activities was the formation of the United States Animal Health Association in 1897, from which was formed the American Association of Veterinary Laboratory Diagnosticicians (AAVLD) in 1957, coinciding with the founding of the American Association of Avi-

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Veterinary Diagnostic  
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Poultry diagnostics today is spread across multiple organizational structures. Necropsy examination and much specimen collection occurs on the farm. The most comprehensive diagnostic capabilities reside in state and university-affiliated laboratories in various administrative configurations. These labs refer cases to National Veterinary Services Laboratory (NVSL) as needed, and serve as nodes on the National Animal Health Laboratory Network (NAHLN). Government and university research laboratories provide diagnostics that are more narrowly defined and reflect the lab mission or the interests of the investigator. Private sector poultry diagnostics variously provide microbiology, serology, pathology and toxicology services. Poultry production company laboratories provide quality assurance testing of incoming feed ingredients, NPIP testing, and support of the processing plant, among other services.

Advances in poultry diagnostics stem chiefly from academic and corporate research laboratories, and from diagnostic laboratories. Quality system implementation has permeated poultry diagnostics in recent decades through AAVLD laboratory accreditation, membership in the NAHLN, and corporate quality programs, all based on standards defined by the International Organization for Standardization (ISO). This trend is sure to continue because it promotes trust among diagnosticians, producers, customers, and regulators worldwide.

## Traditional poultry diagnostics

A traditional diagnostic investigation is a response to problem, be it mortality, impaired production parameters, or a spike in losses at processing. The occurrence of a single disease such as influenza or a novel viral neoplasia can be highly significant. More often, the problem is multifactorial. A diagnosis of coccidiosis is useful, but for commercial poultry, there may be contributing diseases (enteritis, immunosuppression), or factors other than disease (litter mismanagement, drug resistance and coccidia vaccination errors), that serve to promote the coccidiosis problem. Thus, poultry diagnostics requires not just a knowledge of a disease process but an understanding of the poultry production system. Excluding regulatory tests that require a positive or negative result, poultry diagnostics requires identification of the presenting disease and the contributing diseases, and formulating a collective pathogenesis as the basis of control and prevention. It depends on the experience of the diagnostician to rank the duration and severity of each component.

A challenge in poultry diagnostics is first, for the right information to be generated and second, delivered to the right person. The weakness becomes evident when the diagnostician is unable to investigate deeply enough because of technical, economic, or intellectual restraints. Conversely, the recipient of diagnostic information may be unable to understand the interrelationships or to take corrective action. Traditional diagnostics can be repetitive, with many similar cases submitted and investigated, and resources utilized to arrive at the same endpoint.

The lower the fee for service, the more likely this is to occur. The captured data however remains valuable if uniformly recorded and analyzed for epidemiological trends.

## Production-integrated diagnostics

Another way to investigate disease interactions is to integrate diagnostics with production. This model is based on methods used in toxicologic pathology, which uses laboratory animals of identical genetics raised with uniform housing and nutrition, thus comprising a homogenous population that is sampled at defined ages to assess treatment effects. This model closely resembles integrated commercial poultry production. The poultry population (complex, farm, or house) can be sequentially sampled over time for lesion identification and quantitative (measurements, counts) and semiquantitative (lesion scores) characterization (Figure 1). It identifies the age of onset for collection of samples for etiologic agent detection, and thereby guides mitigation. It opens the process to better understanding of subclinical factors contributing to poor uniformity, at-risk subpopulations, and erosion of genetic potential and welfare. The numerical data, properly analyzed, provides visual representation that invites input from the perspective of health, nutrition, and management. It lends to comparative analysis over time because it is subject to quality management procedures for assessment.

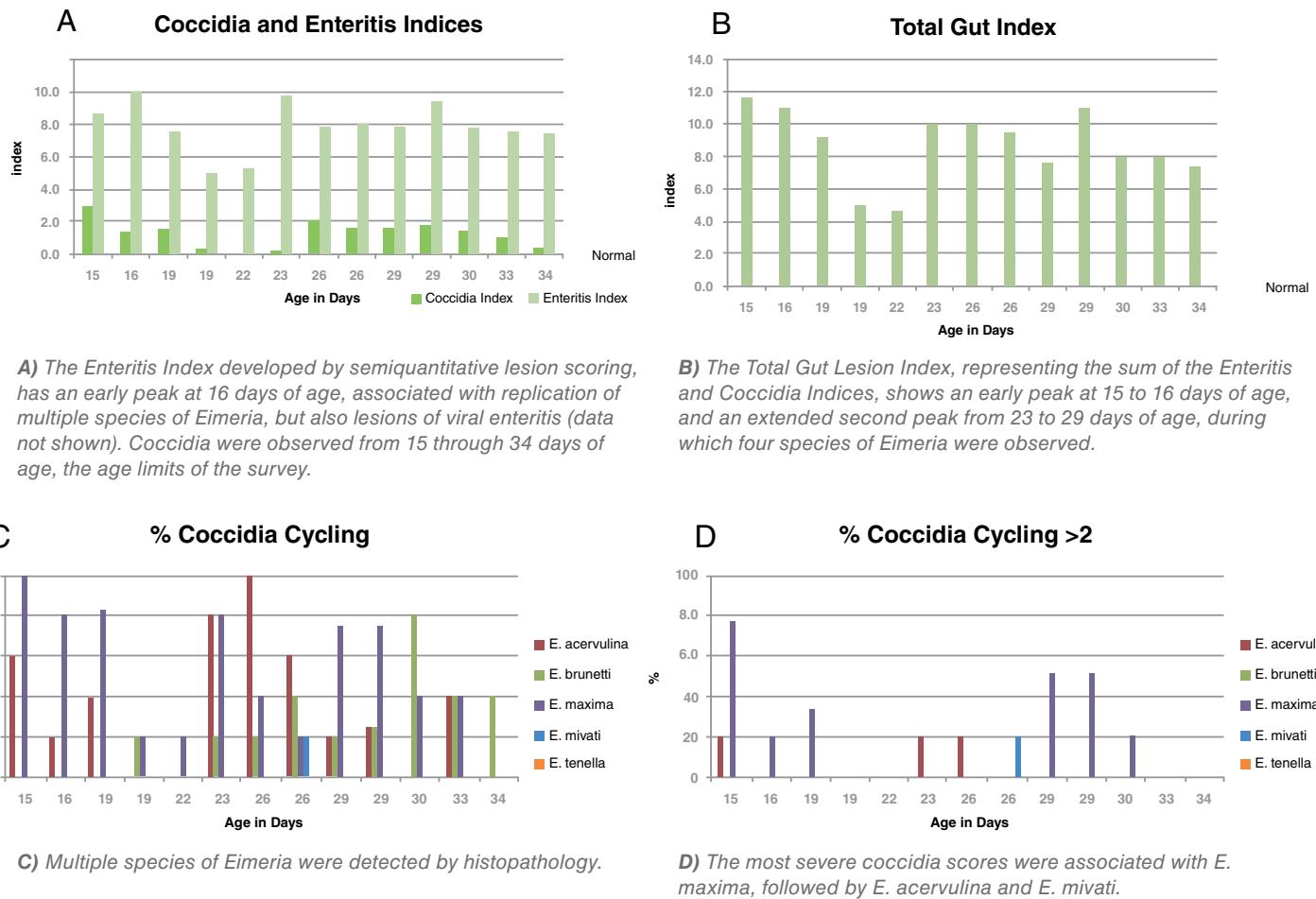
Integrated poultry diagnostics can begin with on-site data collection of necropsy findings, supported by analytical software (Elanco Health Tracking System™, Indianapolis, IN; VDP Path-Pro®, Fort Valley, VA). This can be supplemented with a wide range of analyses. Quantitative and semiquantitative histopathology provides understanding of lesion definition, development, and impact. Sample collection for serology and infectious or toxicological agent characterization yields data to correlate with lesion development, with defined age of onset and severity. Production-integrated diagnostics adds value to the decision-making process.

Production-integrated pathology can help define the relationship of the gut biome to anatomical development and pathological degradation of the gut mucosa. It can define the sequence of immunosuppressive diseases and their interactions through analysis of multiple systems at different ages. Integrated pathology can be applied to critical events in skeletal development, as well as provide correlates for wellness examinations involving footpad quality and gait assessment. In mature poultry, it has found application in mortality surveys and reproductive assessment at various ages.

## Diagnostics in the post-antibiotic era

The ongoing reduction in antibiotic usage and the emergence of the nutraceuticals (direct fed microbials, pre- and postbiotics, fermentation products, botanical extracts) create a challenge for bacterial diagnostics. To a considerable extent, nutraceuti-

**Figure 1.** Production-integrated pathology for a broiler program, with duodenum, jejunum, and cecum from clinically normal broilers examined histologically at 15 through 34 days of age.



cals are in development as substitutes to achieve the health benefits derived by antibiotics. Antibiotics have defined absorption, distribution, and excretion patterns and specific modes of action. Bacterial isolates are routinely tested for antibiotic sensitivity to guide judicious usage and successful therapy. From a regulatory perspective, nutraceuticals in general are on the FDA Generally Recognized as Safe (GRAS) list. Although some have data for *in vitro* activity against poultry pathogens, claims of treatment efficacy are purposely absent, as opposed to regulated antibiotics. Trial data replaces registered claims in demonstrating comparable effects to antibiotics or antibiotic-drug combinations. Compared to antibiotics, the mechanisms of action and interactions of the bioactive components, their distribution and spectrum of activity may have minimal to extensive characterization.

The nutraceuticals are coming onto the poultry market at the same time as high-throughput sequencing and metagenomic analysis of the gut microbiome are becoming mainstream in poultry research laboratories. An inherent issue with nutraceuticals is the consistency of beneficial results in the production

environment. Diagnostic laboratories may have a role in refining the process. Suppression of *Clostridium perfringens*, *Salmonella*, and *Campylobacter* is a desired outcome, as well as counteracting dysbacteriosis through modulating and maintaining a healthy balance of gut bacterial for optimum production efficiency. The bacterial populations are assessed by metagenomic analysis rather than traditional culture methods. This will require an expansion of capability for traditional bacteriology laboratories. Many of the botanical extracts show inhibitory effects *in vitro*, but standardized testing as applied to antibiotic sensitivity is less understood. The digestive tract is the logical first application; however, substantial new information would be needed to assess therapeutic possibilities for septicemia and deep tissue infections.

## Metagenomic analysis and organ-specific microbiota

The gut microbiome is now generally recognized to include viruses, and high-throughput sequencing has shown that more are present than previously recognized. This confirms diag-



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nostic detection of multiple viruses in cases of viral enteritis, including astrovirus, rotavirus, reovirus, parvovirus. Other viruses become apparent with metagenomic analysis, such as picornaviruses and bacteriophages. While the concept of one virus acting as a primary pathogen remains valid, the respective identities, tissue loads, and interactive virulence of these viruses as enteric pathogens will continue as an emerging story. Certain bacterial and viral taxa show the ability to shift together in dominance within the microbiota. Effacement of the gut mucosa caused by parasites and toxins further affect the balance of the microbiota, the manner of which is just now being revealed. The microbiomes of poultry skin (carcass rinse), and respiratory, urogenital, and skeletal systems invite research investigation and diagnostic applications, and can be anticipated to find a place in the poultry diagnostic lexicon.

## Discussion

In addition to traditional state and university-based diagnostic laboratories, there are opportunities to provide diagnostic services through an emerging private sector. While poultry consultants have long played a role in poultry health, privately-owned, independent diagnostic service providers create additional options for those needing services, as well as those seeking careers in diagnostics. The advantages of the private sector diagnostics are rapid, flexible approaches to project beyond that possible in a traditional diagnostic setting. Some disadvantages are a work environment more distant from academic resources and collegial interaction, the need to outsource services, and working without the operational infrastructure typically found in state and university laboratories.

Two keys to a successful career in poultry diagnostics are sound knowledge and skills in a diagnostic discipline, and the ability to merge this with an understanding of integrated poultry production. The core development of these future diagnosticians resides in student programs supported by the AAAP Foundation, support for graduate student research training, and postgraduate training programs and residencies, such as those recognized by the American College of Poultry Veterinarians.

The critical test for the future of poultry diagnostics is the addition of value to production efficiency, poultry welfare, and food safety. Within this framework, technology will continue to advance but the value will depend on successful application to mechanisms of disease and to efficient and humane poultry production. Quality system principles and practices will be integral to trustworthy communication and collaboration among diagnosticians and animal health regulators, and facilitation of domestic and global movement of healthy poultry and wholesome poultry products.

**Acknowledgement:** the author thanks John McCarty, Luis Gomez, Joan Schrader, Floyd Wilson, Joel Cline and Marty Hoerr for contributions to, and ongoing support of, production-integrated pathology.

## References are available on request

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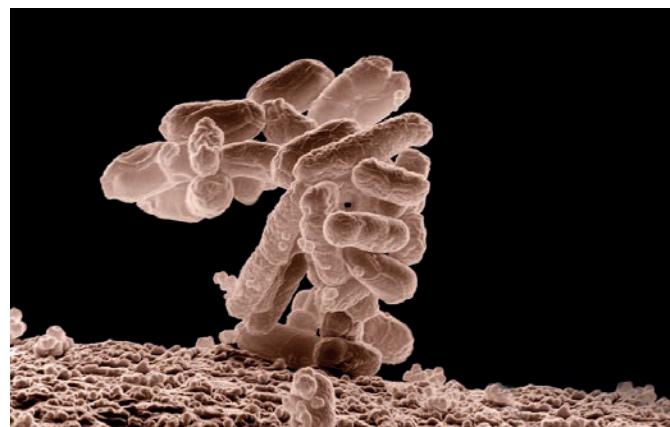
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## How to counteract *Escherichia coli*

*Reducing the challenge in turkeys through  
an antibiotic alternative strategy*

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*Colibacillosis is a systemic multifactorial disease caused by *E. coli* that still represents a major problem in the turkey industry worldwide because of its severe impact on mortality, average daily gain and feed conversion of animals.*

The disease can occur in different clinical forms, including colisepticemia, airsacculitis, peri-hepatitis and peri-carditis. The frequency and the severity of the disease depend on three main factors: the health status of the flock, the presence of predisposing environmental factors and the virulence of the strain involved. Antimicrobials have been and are used to counteract *E. coli* replication; however, in relation to the increasing percentages of antibiotic resistant *E. coli* strain isolated, poultry producers have been encouraged to reduce the use of antibiotic and to identify antibiotic alternative strategy (1). Therefore, the aim of the present study was to test the effects of an enhanced organic acid based product on the replication of an antibiotic resistant *E. coli*.

## Materials and methods

### Trial design

Ninety-six turkey poult of one day of age (females, Big 6 Aviagen®) were randomly assigned to 4 poultry isolators (24 animals/isolator), corresponding to 4 different dietary treatments. Four animals in each isolator were left untagged. Feed and water administered to birds were previously tested for *E. coli*, Enterobacteriaceae, *Clostridium spp.* and *Salmonella spp.* absence. From day 1 to day 3, all the birds were treated with colistin via drinking water (colistin 12%; 2 ml / l as recommended by the manufacturer).

From day 4 until the end of the trial (30 days), the 4 groups received the following dietary treatments:

- **negative control (NC):** standard diet, no additional feed or water supplements provided;
- **positive control (PC):** standard diet organic acid based additive at 0,5 ml / l of water;
- **trial group I (TGI):** standard diet organic acid based additive at 1 kg / ton of feed;
- **trial group II (TGII):** standard diet organic acid based additive at 2 kg / ton of feed.

The acid based feed additive tested is a commercial product (Biotronic® Top 3) composed of a Permeabilizing Complex™ a combination of organic acids (blend of formic, propionic and acetic acid) and a phytochemical (cinnamaldehyde). On day 11, all the birds in each group were orally challenged with  $1,38 \times 10^8$  cfu/mL of *E. coli*. The strain

used was a field one, O78 serotype, isolated during an outbreak of colisepticemia occurred in 2014 in a turkey flock. The strain was resistant to enrofloxacin. Before the *E. coli* challenge and following the treatment with colistin, the 4 untagged animals/isolator were sacrificed by cervical dislocation and examined by bacteriological analysis in order to

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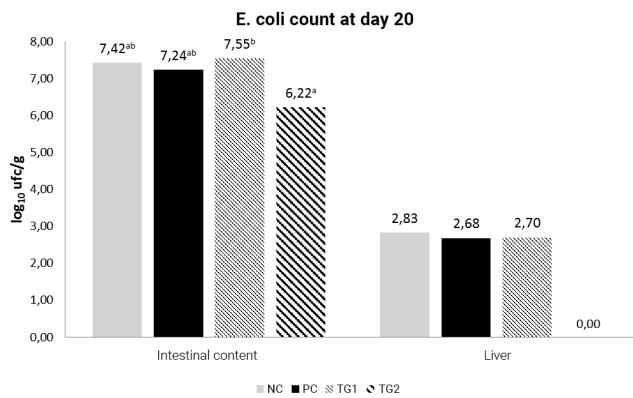
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**Fig. 1** - *E. coli* average count of the intestinal and liver content (g.) at day 20 of the experiment

<sup>a,b</sup> = Means with different superscripts differ significantly;  $P < 0.05$

confirm that they were free either from the challenged O78 serotype of *E. coli* as well as from any other *E. coli* strains. Therefore, since day 4, 20 animals were left in each isolator and grown for the rest of the trial. At day 20 and 30 of the trial, 10 birds from each isolator were sacrificed by cervical dislocation and examined by bacteriological analysis.

#### Lesion score

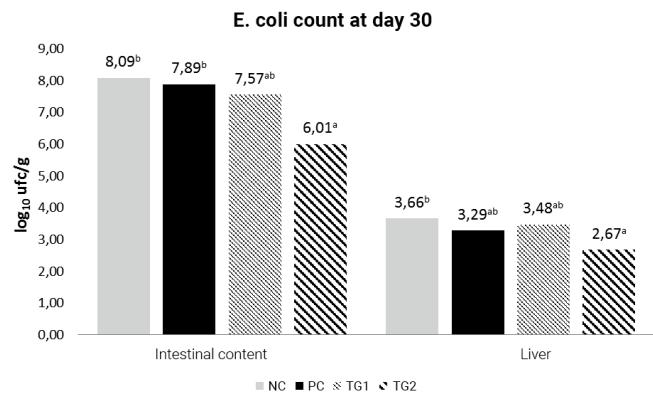
Sacrificed birds were submitted to necropsy examination. The gross lesions of the liver were scored using the following keys, modified from that described by Van Eck and Goren (1991) (2): 0, no lesions; 0.5, one yellow or brown pin-head sized inflammatory spot; 1, two or more pin-head sized inflammatory spots; 2, thin layer of fibrinous exudate on various locations; 3, thick and extensive fibrinous exudation. Mean lesion score per group were calculated.

#### *E. coli* and Enterobacteriaceae count

One gram of intestinal content and 1 g of homogenized liver were ten-fold diluted in Buffered Peptone Water (BPW) and serially diluted to appropriate levels for plating. From each sample, 100 µL were spiral plated on Tryptone Bile Agar with glucuronide (TBX-agar) for the isolation and enumeration of *E. coli* and on Violet Red Bile Glucose Agar (VRBGA) for the isolation and enumeration of Enterobacteriaceae. The results were expressed as cfu/g of tissue or of intestinal content. At day 20 and 30, one TBX plate/bird yielding isolated *E. coli* colonies was subcultured and serologically typed (by slide-agglutination test using specific antisera) for "O" antigen in order to verify if the isolated serotype matched the *E. coli* serotype challenged (O78).

#### Statistical analysis

Bacterial counts were log<sub>10</sub> transformed and analyzed by the non-parametric Kruskal-Wallis test using SPSS 20.0 software (SPSS Inc., Chicago, IL). Number of birds with colibacillosis



**Fig. 2** - *E. Coli* average count of the intestinal and liver content (g.) at day 30 of the experiment

<sup>a,b</sup> = Means with different superscripts differ significantly;  $P < 0.05$

lesions was compared with the Chi-Square test, adjusted by the Yates correction, while mean lesion scores were compared using Mann-Whitney U test. Statistical significance was considered at a probability of  $P < 0.05$ .

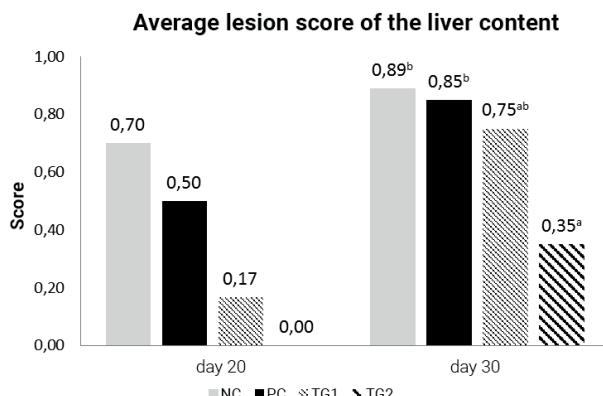
## Results

#### Bacterial count

At any interval of the experiment, the *Enterobacteriaceae* count in the intestinal tract of animals of TGII was lower ( $P < 0.05$ ) compared to animals of TGI and PC. At day 30 of the experiment, the cfu/g of intestine isolated from TGII animals was significantly lower than those isolated from the NC and PC groups ( $P < 0.05$ ). Also *E. coli* colonies were found in the gut of all challenged animals, despite the dietary treatment (Fig. 1-2). At any interval after the *E. coli* challenge, there were no differences in the *E. coli* population between the NC, PC and TGI groups. At day 20, TGII birds presented a lower ( $P < 0.05$ ) intestinal *E. coli* population than TGI animals. At day 30, the number of *E. coli* colonies isolated from TGII animals was significantly lower ( $P < 0.05$ ) than that isolated from both control groups. With regard to the detection of *E. coli* in the liver (Fig. 1-2), at day 20, no *E. coli* colonies were isolated from challenged animals supplemented with 2 kg / ton of acidifier. At day 30, *E. coli* colonies were isolated from livers of TGII birds but they were numerically lower compared with the PC group and TGII and significantly lower than the NC group ( $P < 0.05$ ). Results from the serotyping indicated that the O78 serotype of *E. coli* challenged was recovered in the gut of the birds at all intervals after the challenge.

#### Lesion score

At day 20, 60% of animals from NC and PC group and 20% of animals from TGI presented lesions of colibacillosis, with a mean lesion score of 0.75, 0.50 and 0.17 respectively (Fig. 3). At day 30, also animals of TGII presented lesions (60% of birds) with a mean lesion score of 0.30, whereas the 100%



**Fig. 3 - Average lesion score of the liver content**  
a,b = Means with different superscripts differ significantly; P < 0.05

of animals of the other groups presented lesions with a mean lesion score of 0.89 for NC, 0.85 for PC groups and 0.75 for TGII (Fig. 3). Between control groups and TGII, mean lesions score and percentage of affected turkeys differed significantly ( $P < 0.05$ ).

## Discussion

The effects of the active ingredients of the organic acids based product (Biotronic® Top 3, Biomin) on Gram-negative bacterial growth have been previously tested *in vitro* via a microplate assay (3). It resulted that the antimicrobial mixture inhibited the bacterial growth more effectively than the components alone, demonstrating a synergism of the effects. The Permeabilizing Complex™, in fact, destabilizing Gram-negative outer membrane, facilitates the penetration of cinnamaldehyde and organic acids into bacterial cells. The cinnamaldehyde is reported to have antimicrobial effects, as it targets the FtsZ protein of pathogens, which plays an important role in the cell division of bacteria. Thus, this substance, binding to the FtsZ, perturbs the formation of the Z-ring and inhibits the process of cell division (4). The antimicrobial mode of action of organic acids is two-fold: first, organic acids lead to a reduction in pH inhibiting the growth of microorganisms; second, in their non-dissociated form, they penetrate through the bacterial cell wall and destroy some bacterial vital cell functions (5). In this *in vivo* study, a 26 days continuous diet's supplementation with 2 kg / ton of the product reduced the number of *Enterobacteriaceae* and *E. coli* cfu isolated from the intestinal tract of the treated birds compared with NC and PC groups. Thus, 2 kg / ton of product resulted effective on controlling the *E. coli* replication, furthermore on controlling an antibiotic resistant one.

On the contrary, supplementation with 1 kg / ton did not have any effect. In a previous study, we tested the efficacy of the product on controlling *Salmonella Enteritidis* colonization after eye-drop inoculation of SPF chickens and the inclusion of the acidifier at both the dosages (1 kg / ton and 2 kg / ton of

feed) significantly decreased the *S. Enteritidis* cecal concentration (unpublished work). Considering that both *E. coli* and *S. Enteritidis* are Gram-negative bacteria, we can suppose that, in this study, the less efficacy of the product at a low dosage could be due to the high resistance of the *E. coli* used for the challenge, i.e an antibiotic resistant strain naturally selected. Although we obtained a significant reduction of *E. coli* cfu in the intestinal tract of TGII animals, this did not completely prevent the bacterial translocation in the liver, in particular at day 30 of the trial. However, it is more important, in our opinion, that the inclusion of the product has kept low the replication of the pathogen in TGII turkeys, as demonstrated by the low lesion score attributed during the necropsy (Fig. 3). Finally, although the supplementation of the product did not allow a complete intestinal clearance of *E. coli*, we can suppose that a 2 log<sub>10</sub> cfu reduction could allow less chance of environmental contamination (6) and, consequently, limit the horizontally diffusion of the pathogen. Thus, since feed acidifiers in general are included in poultry diets as alternative to antibiotic growth promoters, for preventing and controlling the gastrointestinal colonization of pathogens, the results obtained in this study with the tested enhanced organic acid based product, under experimental conditions and with a high challenge dose, are promising.

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## **Salmonella, creating the most undesirable environment**

*The ability for Salmonella to survive and grow within farm and processing environments are directly related to their characteristics. Salmonella is a gram negative bacteria, which has over 2,500 different serovars falling into three different categories based on the host.*

---

### **Background**

---

Angela Shaw - PhD  
Assistant Professor  
Extension and Outreach  
Food Safety Specialist

Department of Food  
Science and Human  
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USA

Salmonella non-typhoidal is among the top foodborne bacteria that cause illness within the United States annually. With 1.4 million illness (11% of foodborne bacteria), 19,336 hospitalizations (35% of foodborne bacteria), and 378 deaths (28% of foodborne bacteria), *Salmonella* is estimated at costing the U.S. \$2.65 billion in economic loss. Within the poultry industry it is estimated that 47 billion shell eggs are produced annually with close to 2.3 million of these eggs *Salmonella enteritidis* positive. U.S. Department of Agriculture's Animal and Plant Health Inspection Services, National Poultry Improvement Plan and Veterinary Services are the organizations that coordinate during outbreak scenarios with poultry and poultry products to ensure the safety of products produced within the U.S.

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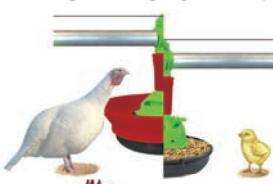


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## Characteristics of *Salmonella*

Some serovars of *Salmonella* only cause infection in humans, others are host-adapted (meaning they may be only found in poultry or cattle) and while the three categories represents the largest group of *Salmonella* that are Unadapted and can survive within multiple host, the Unadapted category of *Salmonella* represents the majority of foodborne serovars, which includes *enteritidis*. Another key characteristic to *Salmonella* is the presence of over 40 different secreted virulence factors. Virulence factors are molecules that are excreted by bacteria so they can colonize and attach to the host, invade the host immune system, inhibit the host immune system response, allows for entry and exiting the internal cells and aids with obtaining nutrients from the host. With such a high number of these factors, *Salmonella* has a wide range of attack and survival mechanisms to affect poultry (broilers and layers). It is well researched that *Salmonella*'s primary method for transmission is through contaminated feed, water, the environment (air), and/or the mother to the offspring. Once *Salmonella* has entered the host, it passes through the stomach and colonizes the gut and adheres to the

Moisture levels, temperature control, and acidification of the environment are keys to this undesirable environment. Humidity and temperature control during the hotter and colder months can be controlled through ventilation system and location of facility to optimize the air-flow through the unit. Reduction of dust within the air is optimum for controlling *Salmonella* but significant increases in added moisture to manure can cause problematic for environmental *Salmonella* survivability. Acidification of water sources along with use of acidified sanitizers, have been proven to be effective at creating an undesirable condition for *Salmonella*. It must be noted that *Salmonella* can become acid adaptive so rotation of sanitizers is critical to reduce selection of specific strains of *Salmonella*.

Flock health, sanitation, pest management, personnel, and incoming sources must be modified to prevent and control the presence of *Salmonella*. Flock health and selection of flock's reliable sources for all phases of production is the first step to pathogen free environment. Reduction of stress levels is another method for creating an undesirable environment for *Salmonella*.

---

*"It is well researched that *Salmonella*'s primary method for transmission is through contaminated feed, water, the environment (air), and/or the mother to the offspring. Once *Salmonella* has entered the host, it passes through the stomach and colonizes the gut and adheres to the mucosa layer"*

---

mucosa layer. Once colonization occurs, *Salmonella* has the ability to invade and infect other organs. Nesting material, dust, shipping containers, and rodents have also been linked to post laying contamination to the environment. *Salmonella* optimally grows between 37°C (5 and 45°C), at pH between 6.5-7.5, in moist conditions (Aw 0.93 or higher), with or without oxygen (facultative anaerobe). *Salmonella* has been shown to be more heat and acid resistant when the water activity and temperature are increased. With the growing conditions of layers and broilers being high humidity, temperatures, ample oxygen, and neutral pHs in cage conditions, *Salmonella* is within their optimal growth conditions.

## Intervention strategies

With optimal *Salmonella* growing/survival conditions within the layer and boiler environment, creation of undesirable environmental conditions must be strategically planned. Reduction of critical elements within production along with practical measures to ensure that *Salmonella* is not brought into the facility from outside sources combined provides this strategic plan.

*Salmonella* is an opportunistic bacteria. It only causes problems in humans and animals having their immune system compromised and showing the opportunity to take over the host. When animals are stressed during production due to management practices (i.e. change in staff, change in environment, temperature or moisture changes, feed withdrawal) their immune systems are weakened and the ability of *Salmonella* to infect the animal is significantly increased (this has been shown in many different studies). Ensuring the chicken has a healthy immune system along with minimal stress, is ideal scenario even if *Salmonella* is present. Antibiotic treatments along with probiotics have been proven in several animal based studies to reduce the prevalence of infections with *Salmonella* when *Salmonella* is present.

Nesting materials, dust, air sources, and feedstuffs are all controllable vectors to minimize the presence of *Salmonella* as they all are testable environmental samples to determine risk of contamination. Sanitization practices should include a cleaning and sanitizer step to reduce *Salmonella* within the environment. Cleaning is the removal of visual materials, which must be done prior to sanitizing or the killing of the bacteria.



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If the visual materials are not removed first then the sanitizing step is **not** effective. Rotation of sanitizers is critical to ensure that *Salmonella* strains are not selected for due to use of one category (not brand) of sanitizer. Another element that is critical to create an undesirable environment is to control the personnel flow and hygiene. Policies that limit the personnel's ability to work on other farms, raise their own poultry, and take their vehicle to other farms should be written and enforced by management. Additionally, clothing and boot use and sanitation should be provided and understood policies on health conditions that would exclude them from work.

## Microbial testing strategies

If a facility has a positive sample for *Salmonella*, it is critical to perform microbial swabbing in the areas, which should possess the highest populations if present. High-risk areas include air exchange fans, pathways from the exterior, personnel flow paths and rodent and birdpathways.

Transmission of *Salmonella* through the air is seen by many experts as the primary method of exchange between the environment to the birds and from bird to bird. Capturing samples from the air inlets (inside and outside) will reveal if it is an internal problem or external contamination. Pathways from the exterior (outside the building) along with personnel flow paths will provide information about cross contamination, effectiveness of boot cleaners and policies modification that may need

to occur with personnel flow. Rodents and birds are additional critical vector to transfer many different foodborne pathogens throughout a farm. Presence of rodents and birds increase the risk of *Salmonella* within the facilities and capturing of these critters may reveal *Salmonella* presence and the need for better pest management systems.

Feed stuff and the flock are other key samples that can provide evidence of initial contaminations. Feed stuff and flock testing for *Salmonella* should be part of company policies for acceptable shipments; therefore these two elements during an outbreak or positive *Salmonella* sample scenario would not relieve as much information about the environment.

## Conclusion

*Salmonella* characteristics assess that moisture levels, temperature control and acidification are key elements causing undesirable conditions. Teamed with healthy flocks, sanitation program, pest management, supply verification program and personnel policies will significantly reduce the risk of *Salmonella*. If *Salmonella* is detected, then focusing sampling on air exchange fans, pathways from the exterior, personnel flow paths and rodent and bird pathways will give the most information about the source of contamination.

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# UPCOMING EVENTS

2018

## February, 4 to 7

### 29<sup>th</sup> Annual Australian Poultry Science Symposium

Big Picture, Big Data, Big Future  
Sheraton on the Park, Sydney  
*The University of Sydney*

**Contact:**

Tel.: +61 2 9351 1656

Email: jo-ann.geist@sydney.edu.au

## February, 5 to 7

### VIV-MEA 2018

Abu Dhabi National Exhibition Company (ADNEC)  
*Khaleej Al Arabi Street*  
P.O. Box 5546  
Abu Dhabi, United Arab Emirates

**Contact:**

VIV Worldwide

VNU Exhibitions Europe

Ms. Renate Wiendels

P.O. Box 8800

3503 RV Utrecht, The Netherlands

Email: viv.me@vnuexhibitions.com

## February, 6 to 8

### AgroFarm

All-Russian Exhibition Centre (VVC Grounds) in Hall 75  
Moscow, Russia

**Contact:**

International exhibitor service and stand rental:

#### Gennady Mindru

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Fax: +49 (0) 69 - 24788-138

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Frankfurt, Germany

Tel.: +49(0)69/24 788-257

Fax: +49(0)69/24 788-138

Email: info@dlg-international.com

Website: www.dlg-international.com

## March, 13 to 15

### MPF Convention

New location for 2018:  
Minneapolis Convention Center  
*Minneapolis, Minnesota*

**Contact:**

Midwest Poultry Federation

108 Marty Drive

Buffalo, MN 55313-9338, USA

Website: www.midwestpoultry.com

Stoneleigh Park Warwickshire  
CV8 2LG, United Kingdom

**Contact:**

**Switchboard:** +44 (0) 24 7669 6969

**Fax:** +44 (0) 24 7685 8393

**Email:** teresag@stoneleighevents.com

## May, 15 to 17

### World and Russian poultry breeding development trends, the realities and future challenges

Federal Scientific Centre  
«All-Russian Research and Technological Institute of Poultry»  
Russian Science Academy  
(FSC “VNITIP” RSA),  
*Serguiyev Posad, Moscow Region*

**Contact:**

Russian branch of the WPSA:

Mrs. Tatiana Vasilieva

Email: vasilievatv@gmail.com

## June, 20 to 22

### VIV-Europe 2018

*Jaarbeurs, Utrecht, The Netherlands*

**Contact:**

Ruhan Berculo

Project Manager

Tel.: +31 30 295 2879

Email: ruwan.berculo@vnuexhibitions.com

Website: www.viveurope.nl

## September, 17 to 19

### VIV China 2018

VIV China 2018 will move to Nanjing

The VIV China Organizers

VIV Worldwide

### VNU Exhibitions Europe

Ms. Anneke van Rooijen

P.O. Box 8800

3503 RV Utrecht

The Netherlands

Tel.: +31 30 295 2772

### China

Ms. SHAO Jennifer

Rm 2013, Kelaowo Building

No. 23, Huixindongjie

Chaoyang District, Beijing 100029

P.R. of China

Tel.: +86 10 6498 8358

Fax: +86 10 6497 2776

## May, 9 to 12

### International Poultry Congress

Cultural and Convention Center  
*Omer Halisdemir University*  
Nidge, Turkey

**Contact:**

Prof Dr Ahmet Sekeroglu

Email: ahmet.sekeroglu@ohu.edu.tr

Website: www.ipc2018.org

## May, 15 to 16

### British Pig & Poultry Fair

NAEC Stoneleigh,  
Stoneleigh Park, Warks  
CV8 2LG Grandstand  
Stoneleigh Events Ltd

# Internet Guide

|                                |                                  |                                  |
|--------------------------------|----------------------------------|----------------------------------|
| ABVista                        | emea@abvista.com                 | www.abvista.com                  |
| Agritech                       | agritech@agritech.it             | www.agritech.it                  |
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| Avimpanti                      | info@avimpia.it                  | www.avimpia.it                   |
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| Carfed Italian Branch          | info@carfed.it                   | www.carfed.it                    |
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| Codaf                          | info@codaf.net                   | www.codaf.net                    |
| Corti Zootecnici s.r.l.        | info@cortizootecnici.com         | www.cortizootecnici.com          |
| DSM Nutritional Products       |                                  | www.dsm.com                      |
| Eurosilos SIRP                 | contatti@eurosilos.it            | www.eurosilos.it                 |
| EuroTier                       | eurotier@dlg.org                 | www.eurotier.com                 |
| Facco Poultry Equipment        | facco@facco.net                  | www.facco.net                    |
| Farmer Automatic               | info@farmerautomatic.de          | www.farmerautomatic.de           |
| FIEM                           | fiem@fiem.it                     | www.fiem.it                      |
| Fiera di Forlì                 | info@fieravicola.com             | www.fieravicola.com              |
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| Impex Barneveld BV             | info@impex.nl                    | www.impex.nl                     |
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| Jansen Poultry Equipment       | info@jpe.org                     | www.jpe.org                      |
| LAE-Anlagenbau GmbH            | info@lae-cuxhaven.de             | www.lae-cuxhaven.de              |
| Linco Food Systems A/S         | linco@baader.com                 | www.baader.com                   |
| Lohmann Animal Health          |                                  | www.lohmann.de                   |
| Lohmann Animal Nutrition       |                                  | www.lohmann-an.de                |
| Lohmann Tierzucht              | info@ltz.de                      | www.ltz.de                       |
| Lubing MaschinenFabrik         | info@lubing.de                   | www.lubing.com                   |
| Marel Poultry                  | info.poultry@marel.com           | www.marel.com/poultry-processing |
| Maxitech                       | info@maxitech.it                 | www.maxitech.it                  |
| Mbe Breeding Equipment         | info@mbefabriano.it              | www.mbefabriano.it               |
| Menci                          | commerciale@menci.it             | www.menci.it                     |
| Meyn                           | sales@meyn.com                   | www.meyn.com                     |
| MS Technologies                | sales@MSTegg.com                 | www.MSTegg.com                   |
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| Officine Meccaniche Vettorello | luciano@officinevettorello.it    | www.officinevettorello.it        |
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| Ska                            | ska@ska.it                       | www.ska.it                       |
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| Specht Ten Elsen GmbH & Co. KG | info@specht-tenelsen.de          | www.specht-tenelsen.de           |
| Tecno Poultry Equipment        | info@poultryequipment.com        | www.poultryequipment.com         |
| TPI                            | info@tpi-polytechniek.com        | www.tpi-polytechniek.com         |
| U.S. Poultry & Egg Association | info@uspoultry.org               | www.uspoultry.org                |
| Val-co                         | intl.sales@val-co.com            | www.val-co.com                   |
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| Victoria                       | victoria@victoria-srl.com        | www.incubatricivictoria.com      |
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| Vostermans                     | ventilation@vostermans.com       | www.vostermans.com               |

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## Licence

Registrazione Tribunale di Firenze  
n.3162 Spedizione in A.P. Art.2 comma  
20/B legge 662/96 - Filiale di Firenze

ISSN 0392-0593

## Subscription Rates (1 year / 11 issues):

|                   |         |
|-------------------|---------|
| Europe            | Euro 44 |
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## Art Direction & Layout

Laura Cardilichia - ellegrafica.com

## Cover Image:

"Il Fiore delle Dolomiti" Organic Layer  
Farms (Italy) built and equipped by SKA

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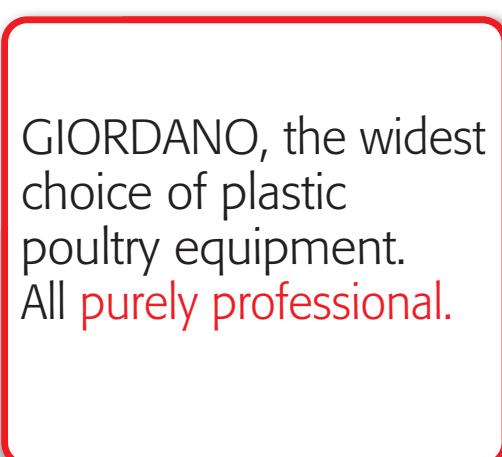
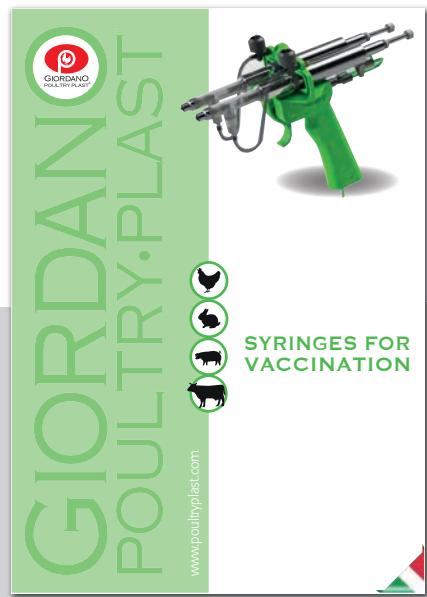
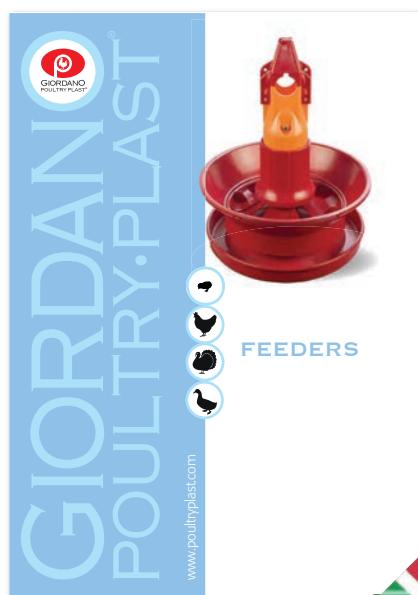
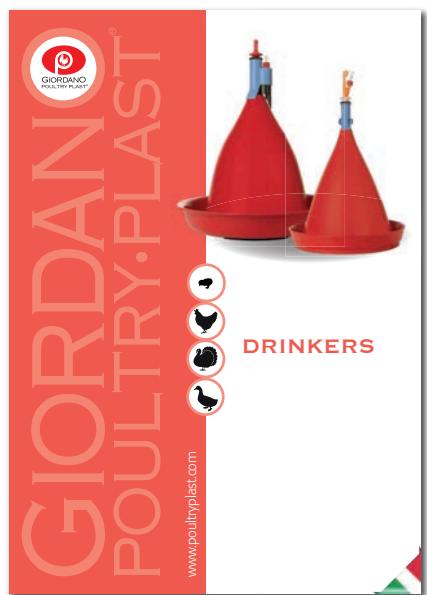
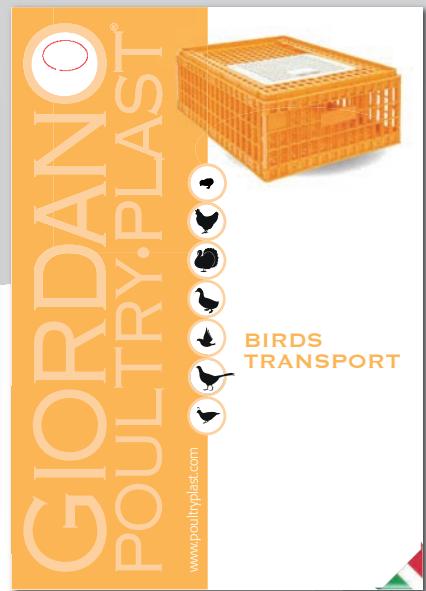
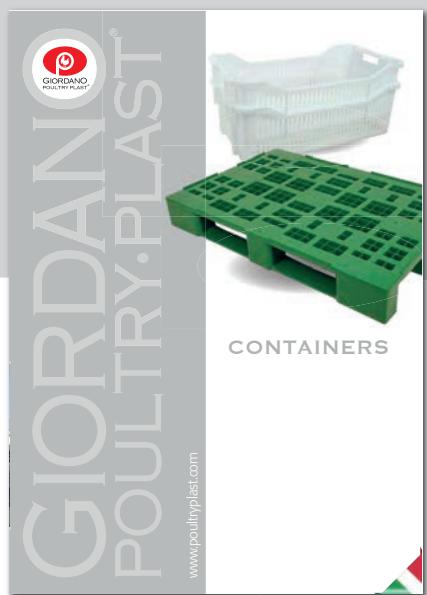
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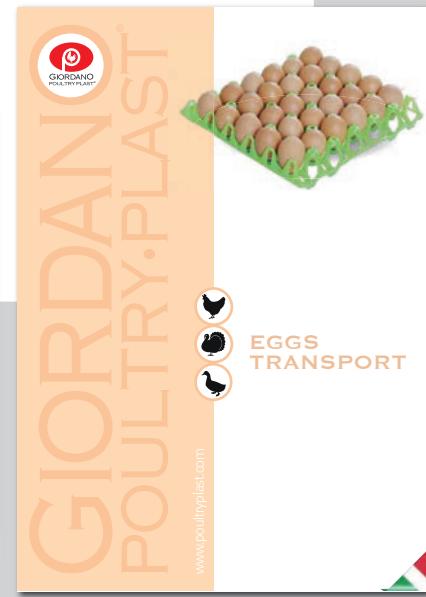


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