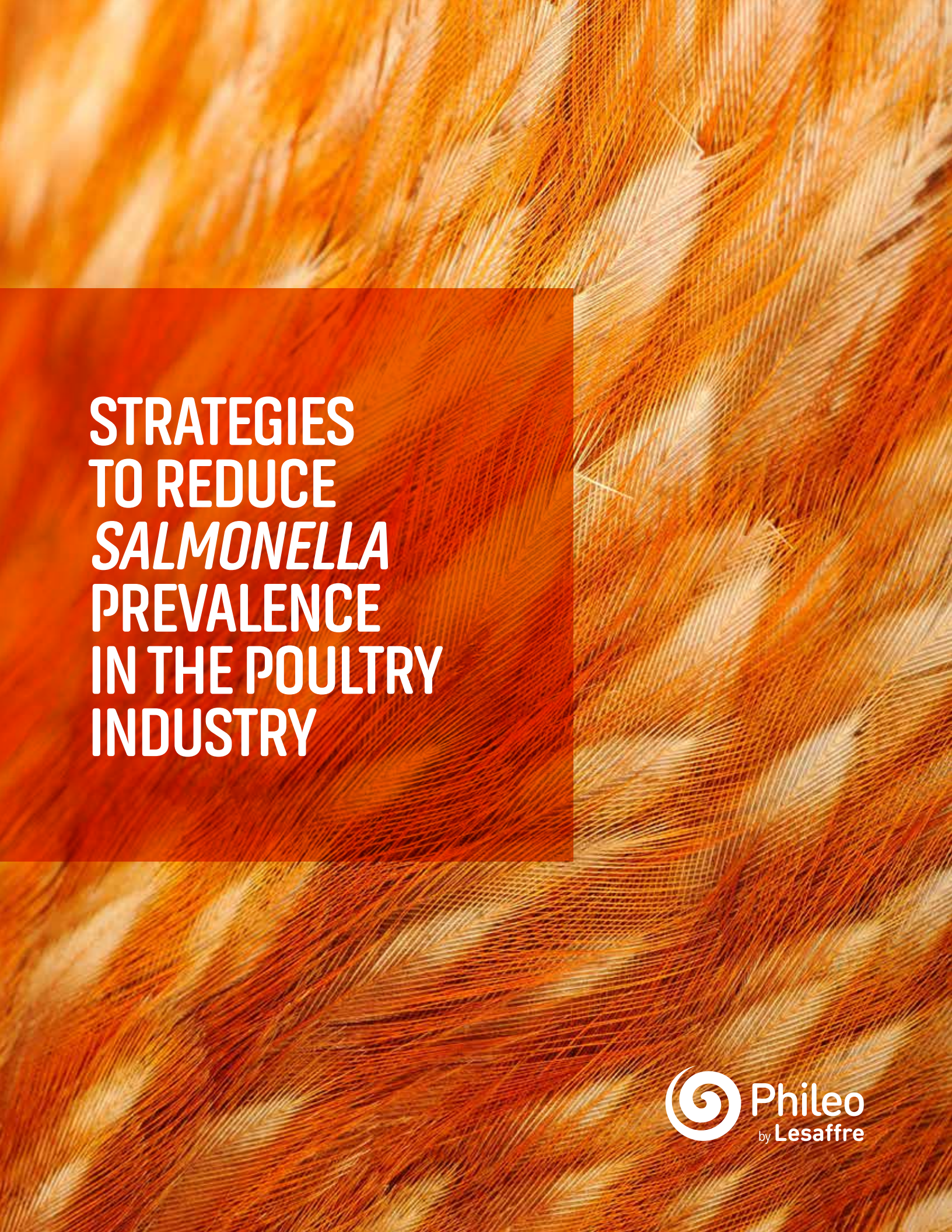


STRATEGIES TO REDUCE *SALMONELLA* PREVALENCE IN THE POULTRY INDUSTRY

Phileo by Lesaffre

SUMMARY

EDITORIAL

"Nothing is more precious than life"

Frederique Clusel
PHILEO GENERAL MANAGER

With the global population growth, **by the year 2050, our planet will be home to more than 9 billion people.**

How, with such numbers, will we succeed to nourish humanity and protect our planet?

Livestock farmers have to meet the growing demand for high-quality protein food products (meat, egg and milk), in large volumes, while supplying them at a reasonable price to feed the planet. Meanwhile, they also face challenges relating to **guaranteeing the food safety** of an increasingly demanding consumer base.

Salmonella contamination is **one of the most important food safety issues** when it comes to poultry production. Conventional chemical solutions have been shown to be effective; however, these solutions fail to meet the requirements of sustainable farming. This is due to their negative impacts on the environment during their manufacture, as well as across the different stages of the animal production chain (labor, animal) and, just as importantly, on the consumer.

Finding new solutions to support sustainable animal production is a challenge that Phileo by Lesaffre embraces – we strive to enhance the lives of animals in order to improve the lives of people.

Food safety is one of the major pillars in guaranteeing animal and consumer health, and accordingly, we have developed sustainable solutions for decreasing *Salmonella* prevalence in the poultry industry. **As such, we are able to enhance both animal welfare, and the quality of life of our consumers.**

Phileo is committed to delivering animal health and performance solutions based on live yeast, bacteria probiotics and purified yeast fractions.. By applying state-of-the-art fermentation technologies, our solutions are bringing global benefits to the environment, to animals, and to consumers, from their own manufacturing process to their application in animal production.

To decrease foodborne illness caused by poultry meat, or eggs, Phileo provides effective solutions to prevent *Salmonella* contamination risks. As we proclaim: **"prevention is better than cure".**

1. FOOD SAFETY CHALLENGES FACING THE POULTRY INDUSTRY TODAY

Studies indicate that poultry meat will take **the lead in feeding the growing global human population in upcoming years.** The poultry sector is already portraying a steep increase in growth, however, if it were to remain satisfactory against increasing consumer demand, **certain major challenges must be surmounted.** Increasing consumption triggers international trade. However, varying food safety standards in different countries complicate the trade process and fail to provide a solution around the globe, especially for developing countries with vulnerable economies[1].

1. Mottet, A., & Tempio, G. (2017). Global poultry production: Current state and future outlook and challenges. *World's Poultry Science Journal*, 73(2), 245-256

WORLD POPULATION BOOM: INCREASES IN MEAT CONSUMPTION ON THE HORIZON

The increasing human population, which is estimated to reach more than 9 billion in 2050, alongside urbanization, rising incomes, and changes in daily diets regarding the increased consumption of livestock products, are bringing about a rising demand for animal protein.

Alexandratos and Bruinsma[2] estimates that **the global demand until 2050 will increase 70% for animal source in general; 66% for beef, 43% for pork, 121% for poultry and 65% for eggs.**

Developing countries are expected to move towards the livestock-based diets that western cultures follow, although the transition time may well differ from one country to another. Some major countries such as Brazil and China[3] have transitioned rapidly, whereas certain others are adapting more slowly to the change due to strong adherences to food habits, or for religious reasons. Furthermore, the religious restrictions on beef meat in India and pork in Islamic countries favor the rapid growth of poultry. As chicken meat and eggs constitute the most common animal products consumed globally as a source of high-quality protein, vitamins and minerals, they are key to food security and nutrition.

2. Alexandratos, N., & Bruinsma, J. (2012). World agriculture towards 2030/2050: the 2020 revision (No. 12-03, p. 4). Rome, FAO : ESA Working paper
3. Refers to China Mainland and Taiwan Province of China

POULTRY: THE FASTEST GROWING LIVESTOCK

Consumer demands in terms of taste, true and meaning food reveal the power that consumers have taken over their food. True food refers to a more authentic, natural, healthier, but also safer food. Consumers are increasingly aware of the impact of food on their health. They are thus becoming more and more careful about what they eat[4].

The poultry industry matches consumers' increasing demands; poultry produce has exhibited a steep growth trend, owing to the short production cycles, low-cost -obtaining 1 kg of meat from a commercial broiler chicken in exchange of 1.7 kg of feed used- and accessibility. Furthermore, successful reinforcement of genetic poultry strains, better understanding of nutrition fundamentals, and **improved food safety measures** have also contributed to the consistent growth of worldwide poultry production.

In addition to **its contribution to food security and nutrition globally**, in developing countries, large-scale poultry production (broilers, egg-laying hens) and family poultry farms provide employment opportunities and income for families of poor backgrounds.

Whilst benefiting from a significant expansion, **the poultry sector is also facing challenges** in the form of newly arising risks. These include antimicrobial resistance reduction, evolving sustainability goals, and the ever-increasing expectations of contemporary consumers. Cost-effectiveness vs. performance outcomes and **feed safety and food safety are the major challenges that the poultry industry faces**.

4. Food360™ (2018). Getting the best of food. Available at https://www.tns-sofres.com/sites/default/files/brochure-food-360-2018-en.pdf

FOOD & FEED SAFETY MEASURES IN THE POULTRY INDUSTRY

The poultry industry has exploited the immediate effect of feed on poultry performance to promote growth, as well as to improve carcass yield and egg production. Moreover, genetic progress to obtain optimal bird growth has brought along changes in birds' nutrition requirements, something which has necessitated the widespread use of protein-rich and energy-dense feed[5].

New strategies have been developed to respond to increasing consumer demands for safety and sustainability, since feed safety is linked to animals' health, and as such to food safety.

In large-scale commercial systems in the poultry sector, 60-70% of the cost of production is feed. As a result, the quality and cost of feed impacts a company's profitability by no small measure. Essential criteria that determine whether a feed additive gets integrated into livestock industry or not include the return on investment, contribution to food safety, consistent outcomes and user-friendliness.

In modern farming, where the breeding density is high, one of the most important challenges is biosecurity control. Thus, poultry meat and eggs might pose a risk to human health **if precautions are not taken along the food production chain**.

5. Ravindran V. Poultry feed availability and nutrition in developing countries. Available at http://www.fao.org/3/a-al703e.pdf

2. *SALMONELLA* WATCH: CONTAMINATION RISKS REDUCTION ACTION PLANS

SALMONELLA CONTAMINATION IN KEY FIGURES

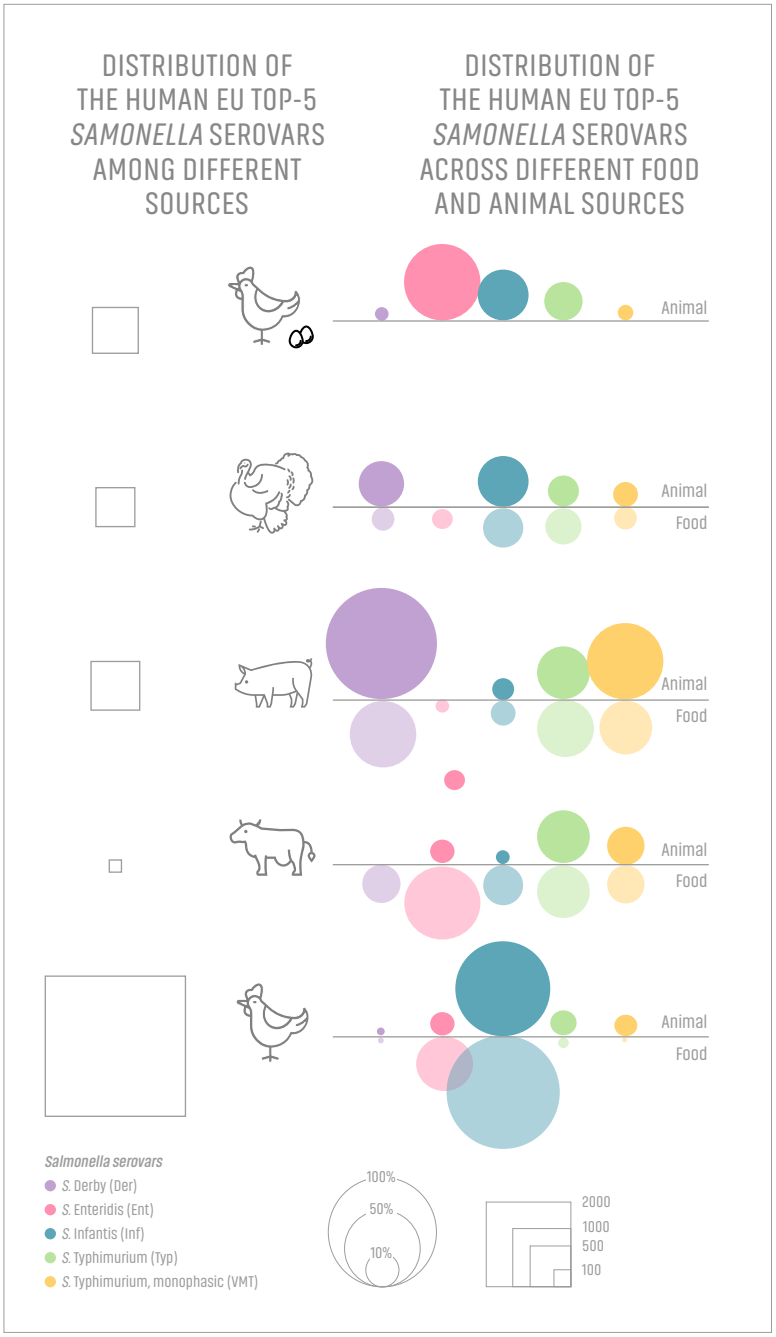
Salmonella is responsible for over 91,000 foodborne illnesses in Europe[6] and approximately 1 million in the U.S.[7] every year. *Salmonellosis* is the second most reported gastrointestinal infection caused by a foodborne pathogen after campylobacteriosis, and the highest levels of *Salmonella*-positive samples come from poultry.

A recent EFSA report on zoonoses reveals that *S.* Enteritidis, *S.* Infantis and *S.* Typhimurium are the 3 most common human-disease causing serovars in poultry products in Europe, whereas different serovars such as *S.* Heidelberg, *S.* Kentucky, *S.* Minnesota might also cause sickness of people elsewhere around the world.

Salmonella infections continue to persist because:

- *Salmonella* is a part of the normal intestinal flora of many animals and **it can survive for more than 9 days in faeces**
- It may contaminate a large variety of food products, including meat, eggs, vegetables, fruits, and more.
- Contamination can occur at any stage of the livestock food production chain, from the hen coops at the farm to the chopping board of the consumer
- Food safety policies focused on **control programs for the reduction of contamination** might take years to be put in practice[8]

To reduce the incidence of contamination, regulations have been introduced (such as Regulation (EC) No. 2160/2003[9]). For instance, as a result of the EU having introduced *Salmonella* control programs for poultry flocks, the number of cases of humans infected with *S.* Enteritidis **dropped by 60%** between 2007 and 2011.



6. The European Union One Health 2018 Zoonoses Report (2019). *EFSA Journal*, 17(12):5926
Available at: https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2019.5926
7. Centers for Disease Control and Prevention (2018). National enteric disease surveillance: *Salmonella* annual report 2016.
Available at: https://www.cdc.gov/nationalsurveillance/pdfs/2016-*Salmonella*-report-508.pdf
8. Centers for Disease Control and Prevention (2011). Vital Signs: Making Food Safer to Eat.
Available at https://www.cdc.gov/vitalsigns/pdf/2011-06-vitalsigns.pdf
9. Regulation (EC) No. 2160/2003 of the European Parliament and of the Council of 17 November 2003 on the control of *Salmonella* and other specified food-borne zoonotic agents

3 QUESTIONS TO

CHARLES HOFACRE

PRESIDENT OF THE SOUTHERN POULTRY RESEARCH GROUP & EMERITUS PROFESSOR AT THE COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF GEORGIA

What are the major food safety challenges faced in the poultry industry?

The poultry producers are facing 2 major foodborne illness issues worldwide: *Salmonella* and *Campylobacter*. In the U.S., the USDA-FSIS has increased the *Salmonella* regularity pressure and is writing new baseline standards for *Campylobacter* (due out summer of 2020) in poultry products.

The current *Salmonella* standards are testing whole carcasses, parts and ground chicken with the number of positive samples accumulative over a 52-week period. The results of this year-long testing is then summarized and the results determine if a company/processing plant is categorized as 1, 2 or 3. A 3 is not meeting USDA performance standards. All of these results are published online and available to the public to see how each processing plant ranks in their *Salmonella* results.

What preventive strategies would you recommend against *Salmonella*?

If I were to help a poultry company that has *Salmonella* issues, I would take a two-step approach to lower contamination:

- **Immediate:** the goal here is to **lower the *Salmonella* on the broiler farm** so that there is less *Salmonella* coming to the processing plant and hence we don't have the risk of foodborne illness.
- **Long term:** trying to eliminate the serovars that are most associated with human foodborne illnesses -S. Enteritidis, Typhimurium, Infantis, Heidelberg, Newport- **out of breeders**. We look at breeders because *Salmonella* can be transmitted from the mother hens to their offspring broilers. That's like a leaking faucet in your kitchen sink, **it drips *Salmonella* continuously from the breeders into the broiler environment.** If we don't stop that dripping, we may lower *Salmonella* coming into the processing plant, but we are not going to be as successful in the long term.

How do you see the future of *Salmonella* prevention in poultry?

We can reduce levels of the higher risk serovars through the use of inactive and live vaccines, probiotics, direct-fed microbiome bacteria, yeast fractions such as SafMannan® and organic acids; however, *Salmonella* is not going to be completely eliminated in poultry production because it is a part of the normal bacterial flora in the intestinal tracts of many animals. The paratyphoid *Salmonella* don't cause any disease to the poultry; therefore, their bodies don't try to eliminate the bacteria. What we are trying to do is to eliminate a normal flora bacterium.

PRACTICES AND STRATEGIES TO REDUCE THE RISK OF CONTAMINATION

The health of the birds in the flock, the type of the food chain (fresh or chilled), the efficiency of the management practices, quality of the hygiene measures (of water, feed, the house), presence of trained staff: all these factors contribute to the delivery of safe poultry, meat, and eggs to the market. Therefore, **risk control measures exist for all stages of the food chain**: from preharvest (on farm animals and their feed), to processing (cutting plants and slaughterhouses) and postharvest (storage, preparation, retail and catering).

PREHARVEST MONITORING AND INTERVENTION

In general, there are **three sources of *Salmonella* contamination on the farms:**

- **Feed:** interest of organic acids and thermisation of feed,
- **House:** which remains contaminated from a previous batch of birds and is poorly disinfected or contaminated because of rodents, insects etc.
- **Chicks:** contaminated by real vertical transmission -*Salmonella* in the egg- or by pseudo-vertical transmission -contamination of the surface of the egg shell when passing through the cloaca.

Therefore, appropriate measures are necessary to minimize *Salmonella* contamination. These include vaccination -mainly in primary breeders- to prevent vertical transmission, products such as probiotics to inhibit *Salmonella* growth via competitive exclusion with non-pathogenic bacteria, postbiotics (inactivated microorganism or fraction with proven benefits on health) to remove bacteria through binding while enhancing the immune system at the same time, and organic acids to lyse bacteria by altering the pH. Furthermore, management practices such as adequate cleaning and disinfection programs, feed and water hygiene, rodent/insect/vermin control all help reduce the risk of horizontal transmission between flocks and houses.

Successful reduction programs are based on a combination of these interventions, and when combining them, it is important to ensure that they have synergism and do not negatively impact the birds[10].

The EU adopts a **farm-to-fork food safety approach**, and a framework of legislation implemented across the Member States targets reduction of *S.* Enteritidis and *S.* Typhimurium contamination in broilers, breeders and turkeys. On the other hand, in the U.S., **long-established voluntary programs that are a part of the National Poultry Improvement Plan (NPIP)** and are supervised by the U.S. Department of Agriculture are in place and they include testing for *Salmonella* in the breeding stock[11]. There is no industry-wide legislation on *Salmonella* and the NPIP program does extend to other parts of the poultry sector in the U.S. In addition, in the EU, national *Salmonella* control plans also extend to feed production[12] whereas in the U.S., *Salmonella* monitoring at the feed mills is not required by the regulatory bodies but is carried out primarily by voluntary programs.

REGULATIONS AND STANDARDS DURING PROCESSING

As part of intestinal microflora, *Salmonella* is carried over to the processing plant on the feathers and in the intestines of birds, making cross-contamination a big problem. Biosecurity controls and hygiene are of the upmost importance in reducing the risk of carcass contamination during processing. As such, large-scale slaughter facilities are portioned into separate sections (at least three; a live birds' area, a slaughtering area, and a processing area including evisceration) and nearly **all procedures are automated to keep birds' contact with surfaces and staff members to an absolute minimum**[13].

Risk identification, knowing how, where and when contamination with microorganisms most commonly occurs **remains an essential first step in health risk control**. As a result, a food safety approach rooted on **Hazard Analysis Critical Control Points (HACCP)** procedures, and which is complemented by advanced regulations can be adopted. Poultry producers in low and middle-income countries follow HACCP procedures for export, whereas additional regulations and legislations exist for the poultry production units in high-income countries. The **Codex Alimentarius**[14] provides a collection of the international food safety standards for guidance.

The biggest poultry producers worldwide are the China, US, EU and Brazil. Harmonizing the CODEX standards between countries remains a challenge, and **food safety legislations also differ in these countries**. In the EU, the microbiological criteria set in legislation[15]

2 QUESTIONS TO

PAUL PRICE

NORTH AMERICA POULTRY MANAGER AT PHILEO BY LESAFFRE

Could you name the top 2 or 3 *Salmonella* serovars considered as foodborne pathogen per poultry species in the U.S.?

Most serovars recovered from broiler carcasses at the last FSIS survey in 2014: **S. Kentucky** 60%, **S. Enteritidis** 13.6%. Commonly identified human illness causing serovar is S. Enteritidis. In the egg industry, **S. Enteritidis** is the only *Salmonella* serovar monitored as eggs are the most common food commodity associated with S. Enteritidis.

What are the existing solutions for farms to reduce *Salmonella*, are they the same for breeder, layer and broiler farms?

The majority of all broiler, breeder hens & egg-laying hens in the U.S. are vaccinated against S. Enteritidis, usually with two live vaccines and 1 killed bacterium.

Most food safety control is directed at the **parent stock**. In broiler operations, this means that all incoming pullets are sourced from genetics companies who practice extremely high-level control for biosecurity, sanitation of feed and house, great grandparent vaccination etc. Broiler breeder operations also practice tight control for visitors, rodents/flies, egg cleanliness, hatchery cleanliness etc. When chicks are placed on the broiler farm, biosecurity is the top form of control and prevention. Litter management strategies to keep houses dry are employed, as well as acidification after flocks to reduce foodborne pathogens by altering pH.

10. The Pew Charitable Trusts Issue Brief (2019). Opportunities to Improve Food Safety From Farm to Fork. Available at https://www.pewtrusts.org/en/research-and-analysis/issue-briefs/2019/05/opportunities-to-improve-food-safety-from-farm-to-fork
11. Comparison of the Regulatory Framework and Key Practices in the Poultry Meat Supply Chain in the EU and USA (2016). Available at https://www.britishpoultry.org.uk/identity-cms/wp-content/uploads/2018/05/2016-ADAS-EU-US-comparison.pdf
12. Regulation (EC) No. 183/2005 of the European Parliament and of the Council laying down requirements for feed hygiene
13. Da Silva, M. V. (2013). Poultry and poultryproducts-risksforhumanhealth: Slaughtering and processing | Poultry DevelopmentReview.Availableathttp://www.fao.org/3/i3531e/i3531e.pdf
14. http://www.fao.org/fao-who-codexalimentarius/codex-texts/all-standards/en/
15. Commission Regulation (EC) No. 2073/2005 of 15 November 2005 on microbiological criteria for foodstuffs

for food products including poultry meat necessitate that *Salmonella* is absent in neck samples from chickens and turkeys after chilling. In the U.S., requirements are set out by the provisions and specific testing procedures established by the federal government for using a range of marketing terms for poultry such as "pullorum clean", "sanitation monitored", and '*salmonella* enteritidis monitored". Sampling requirements vary according to the status (category) of the facilities.

Nevertheless, the safety standards in these countries are all primarily based upon the analysis of *Salmonella* prevalence but with variations in sample sizes, sample timing and maximum percentage of *Salmonella* positive samples allowable. For example, a whole bird carcass rinse in buffered peptone water for 1 min. is performed in the U.S.[16], which can be compared to the pre-enrichment incubation of 1 g of meat in buffered peptone water at 37 °C for 24 h - followed by two further incubations - in China[17]. In the U.S., **9.8% is the maximum allowable for positive samples in whole birds,** whereas in Brazil the threshold **is 20%.**

POSTHARVEST CONTROL MEASURES

Refrigeration is a key factor in food safety to prevent bacterial multiplication on poultry parts and whole carcasses, as well as on comminuted meat. For this reason, meat should be refrigerated immediately after slaughter and refrigeration further down the production chain must be guaranteed for products to be put on the market by certification[18].

At the postharvest stage, national enforcement authorities perform control checks for proper refrigeration, labelling, overall hygiene and fraud. In the EU, temperatures for freezing and chilling poultry meat are specified and a use-by-date is stated on packaged meat as per marketing standards. Additionally, there are specifications for water-to-protein ratios in poultry cuts. Moreover, EU food safety regulations cover the import and trade of meat-based preparations and products. In comparison, in the U.S., when preparing for sale, the Poultry Products Inspection Act requirements that aim to prevent adulterated or mislabeled products from being sold and to ensure that poultry and poultry products are slaughtered and processed under sanitary conditions must be met.

16. FSIS Directive 10,250.1. *Salmonella* and Campylobacter Verification Program for Raw Meat and Poultry Products

17. China National Food Safety Standard GB4789.4-2016: food microbiological examination - *Salmonella*

18. Da Silva, M. V. (2013). Poultry and poultry products - risks for human health: Marketing | Poultry Development Review. Available at http://www.fao.org/3/i3531e/i3531e.pdf

3. SAFMANNAN®: AN EFFECTIVE, EFFICIENT, AND STRATEGIC FOOD SAFETY INTERVENTION

Minimising the prevalence of *Salmonella* coming into the slaughterhouse is the common objective of poultry producers and the regulatory authorities to prevent harmful products from reaching the market, as well as to avoid possible food recalls.

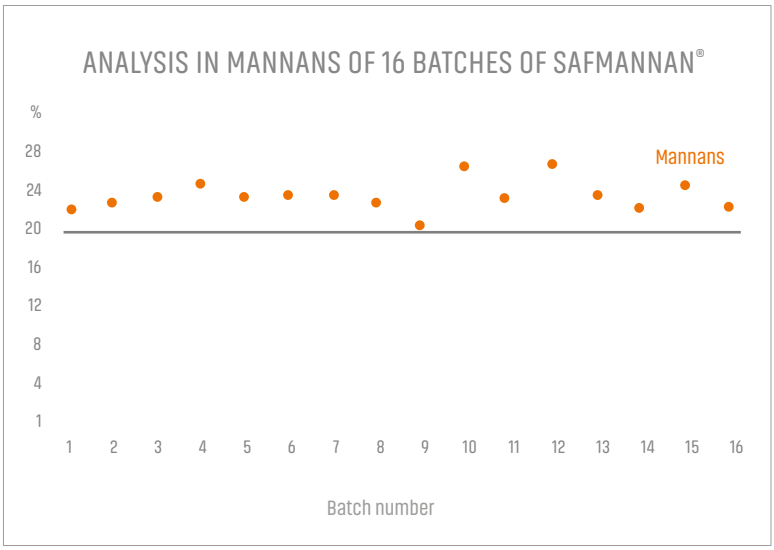
A series of research trials conducted on breeder, egg-laying hens and broilers demonstrate that Safmannan®, a selected yeast fraction, is an effective nutritional intervention that in addition to its main benefits on the health and performance of birds, also **reduces vertical and horizontal contamination on the farm as well as *Salmonella* prevalence or enumeration in carcasses**. In doing so, Safmannan® yields consistent results since the strain, manufacturing process and composition are well controlled.

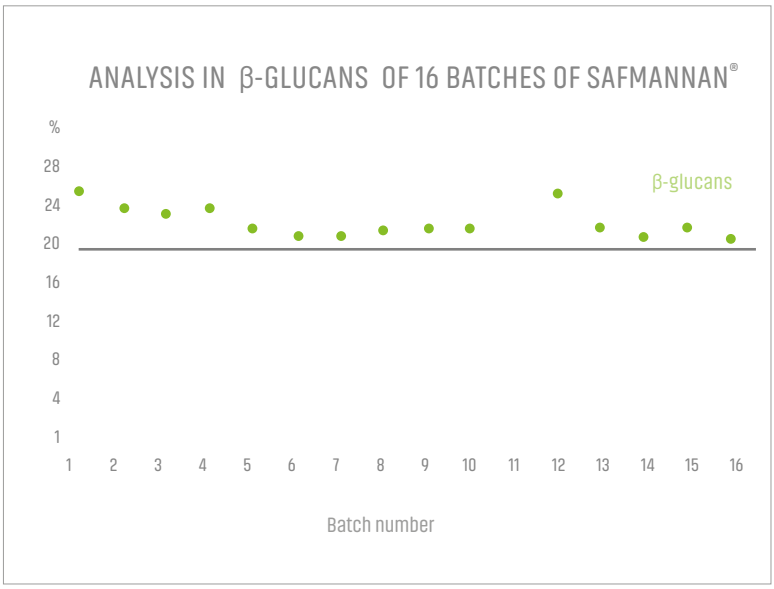
SCIENCE BEHIND SAFMANNAN®

Safmannan® is a yeast postbiotic rich in polysaccharides that are components of the cell wall, mannans and β-1,3/1,6-glucans, obtained by autolysis of *Saccharomyces cerevisiae* proprietary bakery strains. β-glucans stimulate the immune system by getting into contact with the immune cell receptors and make them activated to fight off pathogens[19], while mannans help diminish pathogen colonization by binding to a) the mannose receptors on the intestinal surface and blocking bacterial attachment b) the mannose binding fimbriae[20] on certain bacteria such as *Salmonella*.

The consistent concentration of mannans (≥ 20%) and β-glucans (≥ 20%) in the composition of Safmannan® yields predictable high-performance outcomes.

SAFMANNAN® POLYSACCHARIDE COMPOSITION AND MODE OF ACTION





19. Broadway, P.R., Carroll, J.A., Burdick Sanchez, N.C. (2015). Live yeast and yeast wall supplements enhance immune function and performance in food-producing livestock: a review. Microorganisms, 3:417-427

20. Spring, P., Wenk, C., Dawson, A.K., Newman, E.K. (2000). The effects of dietary mannanoligosaccharides on cecal parameters and the concentrations of enteric bacteria in the ceca of *Salmonella*-challenged broilers chicks. Poultry Science,79: 205-211

SAFMANNAN®, AN INNOVATION TO REDUCE THE CONTAMINATION RISK

SAFMANNAN® STRENGTHENS THE GUT BARRIER FUNCTION

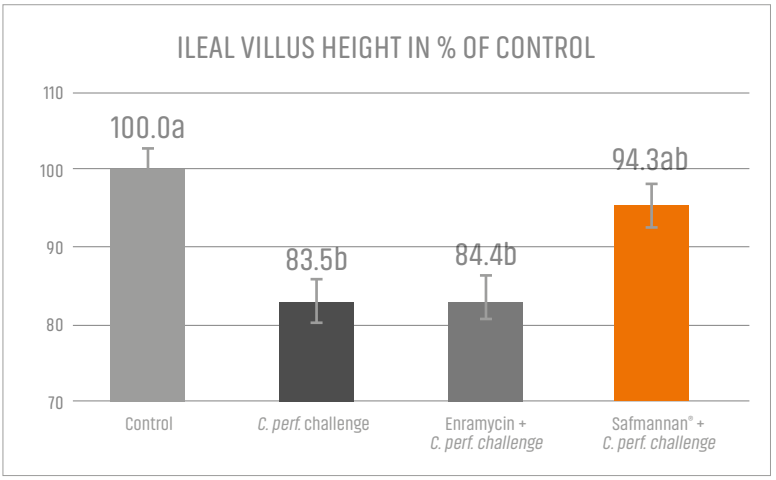
In an experiment conducted on broilers in the grower phase (at day 23), Safmannan® supplementation at 500 ppm was shown to significantly increase goblet cell production ($P<0.0001$) and villus length ($P<0.0001$), thereby promoting mucus secretion ($P<0.0001$) and strengthening the intestinal mucosal barrier against pathogen translocation in comparison to the control diet[21].

Harmful bacteria such as *Clostridium perfringens* may inhibit nutrient absorption in birds' intestines as a result of producing toxic metabolites that irritate the gut mucosa. Another study with *C. perfringens* challenged broilers demonstrated that Safmannan® at 500 ppm maintains the gut integrity more compared to a growth-promoting antibiotic (enramycin) treatment[22]. When morphometric measurements were done on intestinal epithelium samples from birds in the growing phase (day 16), it was found that birds which had received Safmannan® improved villus height to a level which was similar to the unchallenged birds (control). Increases in the absorptive surface of the intestines lead to superior gut health. This explains the mechanism behind the reduction in the ileal *C. perfringens* population of birds at the end of the experiment (day 30) supplemented with Safmannan® (1 Log^{10} CFU/g) as opposed to the control group (6 Log^{10} CFU/g).

EFFECT OF DIETARY YEAST CELL WALLS (SAFMANNAN®) SUPPLEMENTED TO THE DIET ON JEJUNAL MUCOSA MORPHOLOGY[1] AND ILEAL CONTENT VISCOSITY[2] OF 23 D AGE BROILER CHICKENS

		Jejunal mucosa		
	Ileal content viscosity (cps)	Villi heigh (µm)	Mucus thickness (µm)	Goblet cells (number)
YCW avg.				
0 mg/kg	5-01	1024.9[b]	39.1[b]	406.9[b]
500 mg/kg	4-12	1296.7[a]	70.7[a]	1213.9[a]

[1]n= 30 observations; [2]n=5 replicates; [a-b] values within a column not sharing a common superscript



21. Morales-López, R., Auclair, E., Van Immerseel, F., Ducatelle, R., Garcia, F., Brufau, J. (2010). Effects of different yeast cell wall supplements added to maize- or wheat-based diets for broiler chickens. Br Poult Sci, 51(3): 399-408

22. Abudabos, A.M., Yehia, H.M. (2013). Effect of dietary mannan oligosaccharide from Saccharomyces cerevisiae on live performance of broilers under Clostridium perfringens challenge. Italian Journal of Animal Science, 12(38): 231-235

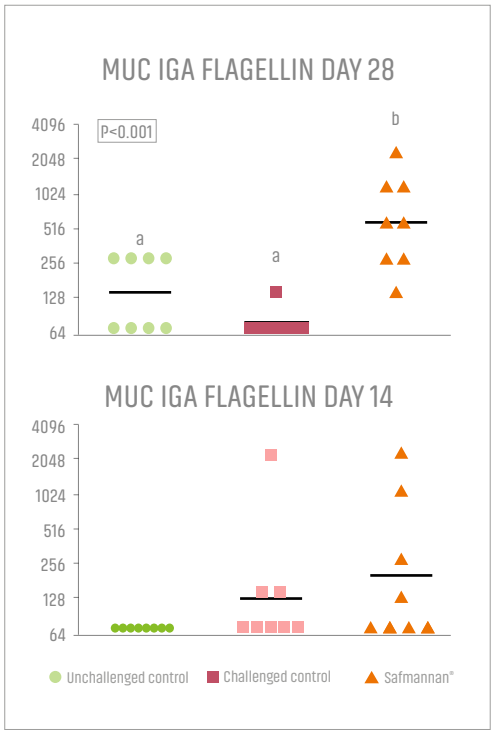
SAFMANNAN® ENHANCES IMMUNE RESPONSES

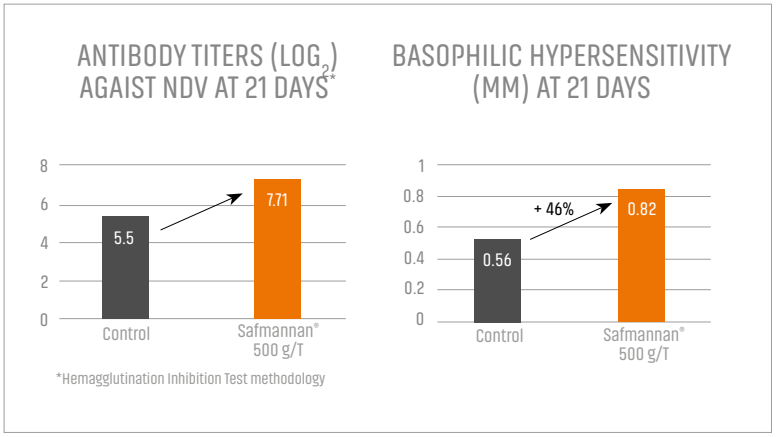
Immune stimulant effects of the selected yeast fraction Safmannan® on broilers were also identified in several experiments. One study conducted on 420 broilers per treatment group revealed that Safmannan® increases blood NDV antibody titers in chicks that have undergone simultaneous vaccination with attenuated live and inactive Newcastle disease virus (NDV) compared to the control group[23].

SafMannan® was also shown to enhance the production of intestinal IgA antibodies against *Salmonella* flagellin in growing broilers (28 d) significantly compared to the challenged and unchallenged controls.

Moreover, the delayed basophilic hypersensitivity test performed in parallel showed increased (46%, P<0.05) interdigital thickness in animals fed with Safmannan® as opposed to the no supplement control.

Safmannan® was also shown to enhance the production of intestinal IgA antibodies against *Salmonella* flagellin in growing broilers (28 d) significantly compared to the challenged and unchallenged controls[24].





2 QUESTIONS TO

PING WEI

MANAGING DIRECTOR AT THE COLLEGE OF VETERINARY MEDICINE, GUANGXI UNIVERSITY

Which 3 strategies would you suggest for *Salmonella* eradication?

In my experience, the three key solutions for *Salmonella* eradication are:

- The elimination of *Salmonella*-carrier birds from the breeder flock
- The elimination of *Salmonella* from the environment in which the birds are raised including any vectors (both biological and material) that can carry *Salmonella* and contaminate birds upon contact
- The use of products that can effectively eliminate or reduce *Salmonella* that bypass preventive barriers and infect the birds

Based on these criteria; I think pre-, pro-, postbiotics, organic acids, phytoceuticals, zinc and copper, fiber, non-starch polysaccharides, and phage therapy targeting the pathogenic bacteria could be used in the poultry industry now and in the future.

What are the proven advantages of nutritional solutions and do these products help reduce foodborne illnesses globally?

The probiotics, postbiotics and other products that supplement the drinking water or feed can reduce the colonization of foodborne pathogens such as *Salmonella* in the birds.

According to our investigations, preventing re-contamination of birds or meat products with foodborne pathogens like *Salmonella* post-raise, during slaughtering and other steps of the whole food chain, is also critical for food safety.

We compared bacterial isolates from birds and meat products and found out that they were not the same species and the genetic analysis further showed that they were not the same genotypes. **Any products or methods that can effectively reduce also the risk of re-contamination could contribute to food safety.**

23. Gómez-Verduzco, G., Cortés-Cuevas, A., & Coello, C.L., Menocal, J.A., Peláez, C.V., González, E.A. (2009). Productive performance and immune response in broilers fed a sorghum+soy diet supplemented with yeast (Saccharomyces cerevisiae) cell walls, in the presence or absence of aflotoxin B1. Tecnica Pecuaria en Mexico, 47(3):285-297.

24. Kiros et al. (2020). Administration of the yeast fraction SafMannan® reduces outcome of *Salmonella* Typhimurium challenge on broilers. Oral presentation - International Poultry Science Forum. Georgia World Congress Center, Atlanta, Georgia, U.S., January 27 - 28

REDUCING *SALMONELLA* WITH SAFMANNAN®: CONCRETE DATA

SAFMANNAN® BINDS BROAD SPECTRUM OF MAJOR PATHOGENS INCLUDING PARATYPHOID AND TYPHOID TYPES OF *SALMONELLA*

Posadas *et al.* (2017), an *in vitro* study, showed that **Safmannan® binds multiple pathogens** including several *S. enterica* serovars that cause human foodborne illnesses such as ***S.* Typhi and *S.* Typhimurium**[25].

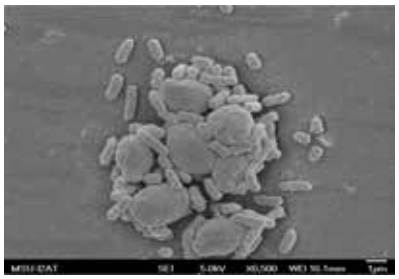
Furthermore, a study published on Poultry Science by Zhou *et al.* 2019 demonstrated that Safmannan® also reduces the population of ***Salmonella pullorum* (SP) and *Salmonella gallinarum* (SG)** that cause pullorosis and fowl typhoid diseases in poultry respectively, improves gut health and thus reduces the probability of *Salmonella* colonising the intestine[26].

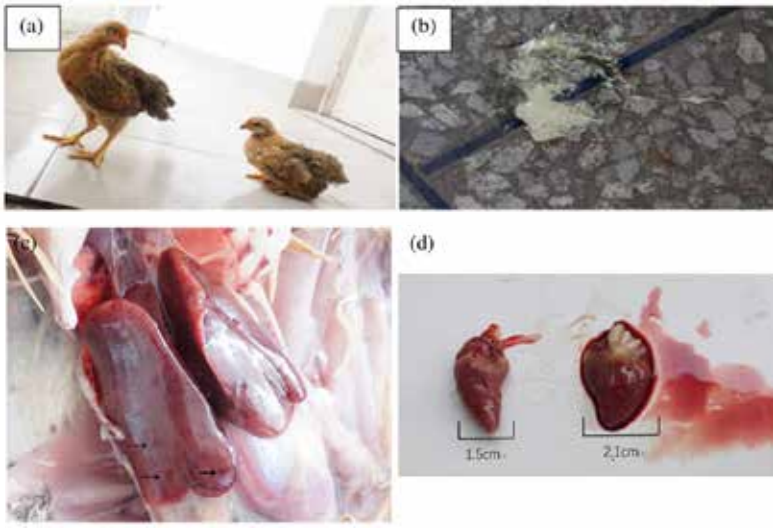
In this study, one hundred and sixty 1-day-old commercial chicks were divided into groups A and B, along with groups C and D, which served as the challenged but non-treated and non-challenged and non-treated control groups respectively. During this 42 d experiment, group A was fed a commercial diet (without any antibiotics) supplemented with Safmannan® (250 ppm), group B was fed the same commercial diet as group A but an organic acid (*Acidipure*, 1.5 mL/L) was added to the drinking water, and groups C and D were fed the same commercial diet without any supplements.

Whereas no birds died during the experiment, birds in groups C1 and C2 had white diarrhea, exhibited poor growth and weakness, and upon necropsy, livers with necrotic white foci as well as a softened heart with pericardial effusion were obser-

SEM AVERAGES FOR ADHERENCE OF SAFMANNAN® TO VARIOUS PATHOGENS (LEFT), TO *S.* TYPHIMURIUM (RIGHT)

Bacteria	% Adhere
S. Typhi	50.00
S. Typhimurium	98.11





(e)

Challenged strains	Groups	Clinical signs white diarrhea	Lesions: Necrotic white foci on the liver surface	Lesions: Distorted shape of the heart
Salmonella pullorum	A1	26.09%	8.70%	21.74%
	B1	30.43%	26.09%	26.09%
	C1	36.00%	44.00%	36.00%
Salmonella gallinarum	A2	21.74%	26.09%	8.70%
	B2	28.00%	36.00%	16.00%
	C2	43.48%	43.48%	17.39%

Illustrating the clinical signs (a) and hearts (d) of two birds from treatment groups A1 (left) vs. C1 (right), white diarrhea (b) and hemorrhage of liver (c) from group C1 and percentages from all treatment groups (e)
A1 - A2: Safmannan® supplemented, B1 - B2: Acidipure supplemented, C1 - C2: no supplement

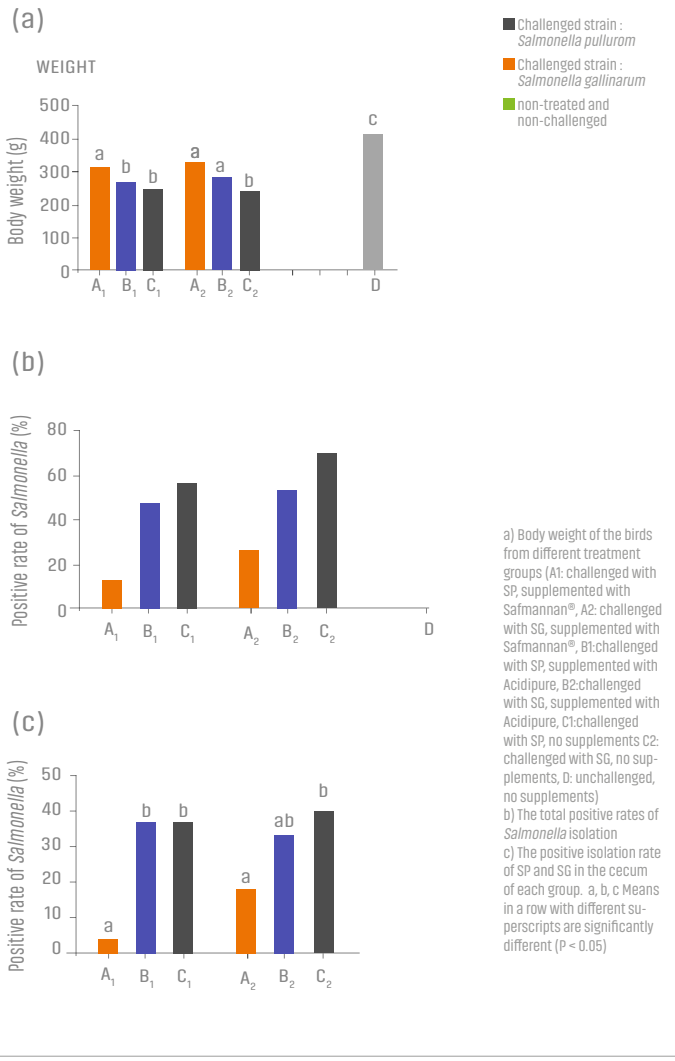
ved in these birds. Birds from group A1 were clinically normal and no lesions in the liver were observed.

Safmannan® was also able to decrease the cecal histology lesion score and *Salmonella* colonization in the liver and ceca of broilers challenged with *S.* Typhimurium[27].

Moreover, at the end of the study conducted by Zhou *et al.* (2019)[29], **bacterial isolation rates from different organs were significantly lower (P < 0.05) and body weights of birds** challenged with SP and SG **remained the highest when birds were fed Safmannan®** - supplemented diets (250 ppm) in comparison to organic *Acidipure* (1.5 mL/L) supplemented drinking water.

SIGNIFICANT DECREASE IN *SALMONELLA* PREVALENCE AND ENUMERATION

In Price *et al.*, 2019, published on the International Journal of Poultry Science, experiments were carried out in the U.S. with 24 **egg-laying hens** where half of the birds were fed a basal diet (control) and the other half were fed the same diet supplemented with Safmannan® at 500 ppm, *S.* Typhimurium in one-week post challenge ceca and ovary samples were enumerated by the MPN method. Cecal counts of *S.* Typhimurium were 4.71 Log^{10} CFU/mL vs. 3.41 Log^{10} CFU/mL in control (P=0.015) vs. Safmannan® - supplemented birds respectively, presenting evidence that Safmannan® is an effective intervention (reduction by > 1 Log^{10}) resulting in less *Salmonella* being shed into the environment[27].



a) Body weight of the birds from different treatment groups (A1: challenged with SP, supplemented with Safmannan®, A2: challenged with SG, supplemented with Safmannan®, B1:challenged with SP, supplemented with Acidipure, B2:challenged with SG, supplemented with Acidipure, C1:challenged with SP, no supplements C2: challenged with SG, no supplements, D: unchallenged, no supplements)
b) The total positive rates of *Salmonella* isolation
c) The positive isolation rate of SP and SG in the cecum of each group. a, b, c Means in a row with different superscripts are significantly different (P < 0.05)

25. Posadas, G.A., Broadway, P.R., Thornton, J.A., Carroll, J.A., Lawrence, A., Corley, J.R., Thompson, A., Donaldson, J.R. (2017). Yeast pro- and paraprobiotics have the capability to bind pathogenic bacteria associated with animal disease. Transl Anim Sci, 1:60-68

26. Zhou, C., Liang, J., Jiang, W., He, X., Liu, S., Wei, P. (2019). The effect of a selected yeast fraction on the prevention of pullorum disease and fowl typhoid in commercial breeder chickens. Poult Sci, 0:1-10

27. Price, P.T., Gaydos, T., Padgett, J.C., Gardner, K., Bailey, C. (2019). *Salmonella* Colonization of Production Hens Fed a Parietal Yeast Fraction with High Levels of Mannan and Beta-Glucan. Int. J. Poult. Sci., 18(9): 410-415

A recent study conducted in the U.S. by Price *et al.* 2020 investigating the effect of Safmannan® on *S.* Enteritidis colonization in **egg-laying hens** presents further evidence that Safmannan® offers a promising solution to *Salmonella* contamination of the environment and eggshells: a reduction of 41.6% in the cecal *S.* Enteritidis prevalence in the Safmannan® treated group vs. the untreated control group[28].

Furthermore, in a similar study where 30 **egg-laying hens** were fed with a control vs. Safmannan® - supplemented (500 ppm) diet, enumeration of *S.* Enteritidis in the ceca by the MPN method revealed an even greater reduction (1.44 Log^{10} MPN/g) in bacterial load as a result of Safmannan® addition: the control group had an enumeration value of 3.35 Log^{10} MPN/g whereas this was 1.91 Log^{10} MPN/g for the treated group ($P<0.05$)[29].

DECLINE IN HORIZONTAL CONTAMINATION

Salmonella can easily be transmitted across a flock with contaminated birds shedding the bacteria into the environment via their faeces. Results illustrate a significant decline in *S.* Heidelberg prevalence in the contact birds that didn't have bacterial inoculation but which acquired the bacteria through contact with their challenged penmates and supplemented with 500 ppm Safmannan® in their diet.

Treatment	Challenge status Indirect* (Contact birds)
Untreated	46/80 (57/5)
Safmannan® (125 ppm)	51/80 (63.8)
Safmannan® (250 ppm)	49/80 (61.3)
Safmannan® (500 ppm)	26/80 (32.5)
Yeast culture (1250 ppm)	42/80 (52.5)

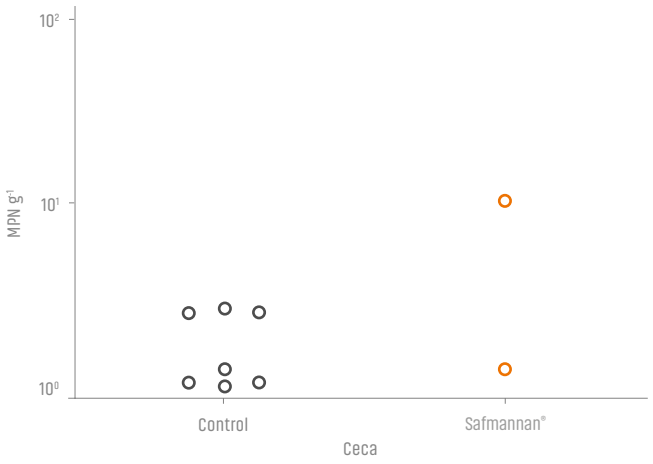
Salmonella prevalence in ceca samples collected on day 42

(a)

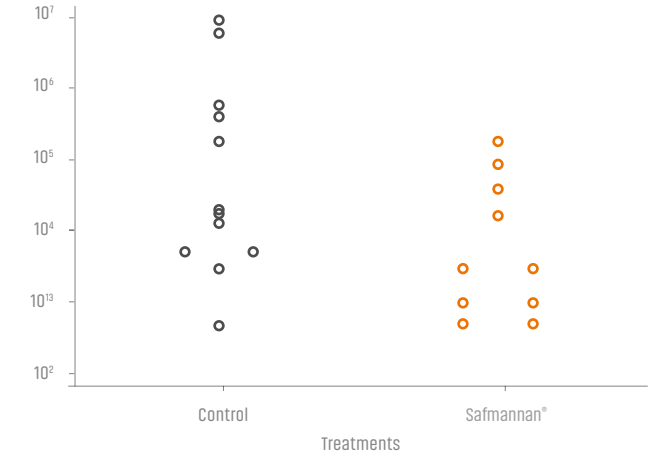
Treatments	No. samples	No. positive (%)	p-value
Control	12	12 (100.0)[a]	0.478
Safmannan®	12	10 (83.3)[a]	

Treatments	No. samples	No. positive (%)
Control	12	4 (33.3)
Safmannan®	12	3(25.0)

(b)



(c)



S. Typhimurium prevalence in ceca (a) and ovary (b) samples by percentage and dot plot (c) of bacterial counts in ceca samples per treatment group

28. Price, P.T., Gaydos, T., Berghaus, R., Hofacre, C. (2020). Reduction of *Salmonella* Enteritidis Colonization in Production Layers Fed High Levels of Mannan and Beta-glucan. Asian J. Poult. Sci., 14:1-5

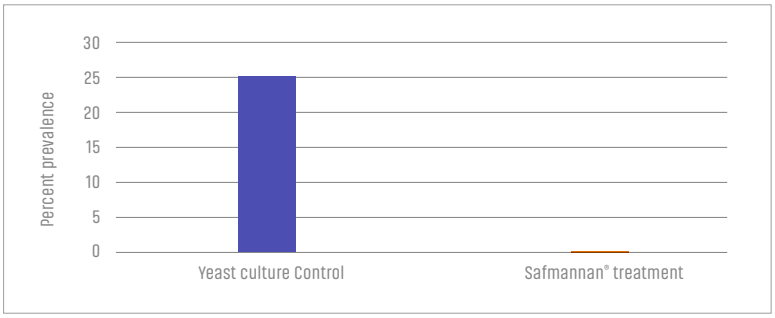
29. Price et al. (2020). Strategies to reduce colonization of *Salmonella* Enteritidis in layers. Poster presentation - International Poultry Science Forum. Georgia World Congress Center, Atlanta, Georgia, U.S., January 27 - 28

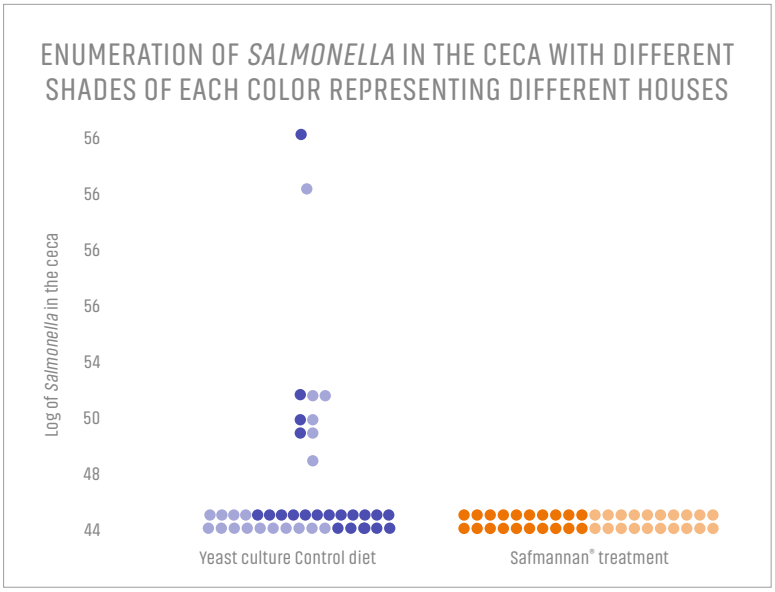
Moreover, enumeration of the bacterial colonization level in the ceca using the MPN method showed that Safmannan® at 500 ppm reduces the bacterial load in the ceca of positive-birds by ~1 Log^{10} compared to the control group (P=0.04).

In an experiment[30] that was carried out in two commercial **broiler** farms in the U.S. each containing four houses, birds in two houses, the control group, was fed a diet that consisted of commercial yeast culture, a proprietary additive blend, a *Bacillus* probiotic and butyric acid as opposed to the remaining birds in the treated group whose diet consisted of the same components, except the yeast culture and additive blend were replaced by Safmannan®. Prevalence and enumeration of *Salmonella* using the most probable number (MPN) method were determined from twenty cecal samples per house taken randomly at the processing plant, post evisceration.

25% of the birds fed with the control diet had *S.* Enteritidis in their ceca, whereas those fed with the Safmannan® -supplemented diet had no detectable levels.

Moreover, enumeration data from the birds in four houses of one of the farms (Farm 2 had no *Salmonella* detected in the ceca of the treatment and the control houses) were compared. Average load of *Salmonella* in the ceca samples from the control houses was 0.72 Log^{10} cfu/g (P<0.05) with two birds having more than 6 Log^{10} cfu/g whereas, it was minimal in samples from Safmannan® - supplemented birds.





30. Price et al. (2019). Yeast cell wall compared to yeast culture on performance and *Salmonella* prevalence in broiler ceca in a commercial setting. XXIst World Veterinary Poultry Association Conference. BITEC, Bangkok, Thailand, September 16 - 20

CONCLUSION

The poultry industry is pressured to meet the increasing and evolving demands of consumers while addressing matters such as food security, feed and food safety but also sustainability. Poultry company's profitability relies on high performance outcomes and health-risk free product obtained in a cost-effective manner.

In poultry production, it is essential to reduce the level of foodborne pathogens that enter the processing plant from the farm to save further costs and loss. To achieve long-term success in reducing *Salmonella* contamination in poultry meat and products**, the risk of both vertical and horizontal transmission needs to be lowered.** It is also essential to prevent the re-contamination of birds by different *Salmonella* serovars. Therefore, myriad of interventions is in use, mostly in combinations, to reduce contamination at preharvest stage. Furthermore, control programs such as for *Salmonella* are put in force by some of the largest poultry producers such as in the U.S. and the EU.

Large number of studies demonstrating their immunomodulatory effects, pathogen binding capacity and gut health improvement have led the poultry industry to integrate selected high-quality yeast fractions into birds' diets as an intervention strategy to contribute to the food safety early in the food production chain.

In addition to previously published benefits of yeast components, selected yeast fraction **Safmannan® provides promising solutions to vertical and horizontal transmission** by binding broad spectrum of pathogens while delivering consistent and high-performance outcomes in **broilers, egg-laying hens and breeders.**

Dietary supplementation of birds with Safmannan® helps consistently lower the prevalence and enumeration of both typhoid and paratyphoid *Salmonella*, and the serovars that are most commonly associated with human foodborne illnesses, like *S.* Enteritidis and *S.* Typhimurium .

The multifactorial benefits of Safmannan® on ensuring animal welfare and the safety of food on your plate contributes to the global sustainability, that is a 'must-pay-attention' factor for all food producers around the world right now.

Phileo by Lesaffre
137, rue Gabriel Péri - BP 3029
59703 Marcq-en-Baroeul, France

info@phileo.lesaffre.com

phileo-lesaffre.com