Impact and control of Infectious Bronchitis in layers and breeders

Eduardo Bernardi
Technical Services Manager - Pacificvet

In New Zealand and the South Pacific countries, we are more fortunate than many other poultry producing nations due to the lack of serious disease issues and our isolation from other continents. Such freedom from the ravages of many virulent diseases is an enormous economic advantage in terms of performance. It allows us a degree of ‘natural biosecurity’ of distance.

There are however some poultry diseases that do occur as either a sporadic or a constant problem within the region. One of the most common threats, and usually included in vaccination programmes for commercial layers or breeders, is Infectious Bronchitis (IB).

Unlike most world areas of massive poultry exploitation, Infectious Bronchitis in New Zealand has not been a threat for broilers or younger birds in rearing. The nature of the local strain of Infectious Bronchitis virus (IBV) has not shown to be as virulent as the classic or variant strains that constantly ravage the poultry industry of North and Latin America, Europe and Asia. No mortality has been observed on experimental infection of chickens with recently-isolated IBV from New Zealand, and only mild changes on the respiratory histology were observed. In addition, the high standards locally achieved in the sanitation of facilities, biosecurity and animal welfare have maintained the level of infection and injury to a minimum. Therefore, in New Zealand vaccines against Infectious Bronchitis are not used on broilers, for even when the IB virus may be infecting a broiler shed, any respiratory symptoms may not have been relevant enough to alter the overall economic performance of the flock.

Although the New Zealand strains of IBV virus have been thoroughly studied in terms of gene sequencing, immunological protection and pathogenicity on the respiratory system and kidneys, little is known about its effect on the reproductive system. A full study of the effects on IB infection on SPF adult layers in laboratory conditions is a very expensive trial, demanding time and subject to several variables. In the last 50 years, we have seen few complete studies on the effects of IB on adult birds, notably the works of Sevoyan (1957) and Bisgaard (1976).

Worldwide, IBV can cause damage in the reproductive system with minimum respiratory involvement (sub-clinical infection) or reach it through an infection in the digestive system. In laying birds, the virus will affect the reproductive organs, more specifically the uro-genital tract, with the possibility of causing significant economic losses. Hence its importance within a layer and breeder vaccination programmes.

The occurrence of Infectious Bronchitis in adult birds has often been called “silent” due to the elusiveness of the clinical symptoms, which may go unnoticed, unless there is a closer examination. Unprotected laying flocks on table egg operations or breeder farms will suffer from subtle peaks of seconds/undergrades, if not dips or oscillations in the rate of production. Consequences may not show themselves as obvious signs of IB, but an overall reduction of income/profits from the flock. These effects, although often cyclic (occurring after a whole cascade of cellular immunity from field infection has been triggered and receded), may persist for the life of the flock.
Effects of IB virus infections in commercial layers are basically reflected in egg numbers and egg quality.

Egg losses in terms of daily production or cumulative eggs per hen house may result from delayed sexual maturity, a decline in lay during or after an infection and the sub-optimal production during the recovery period post-infection. Dips of 3% or more may be observed in the rate of production of unprotected flocks. In some cases, recovery of production may never occur, due to the severity of the injuries in the reproductive system.

Egg size will be affected, with infected birds tending not to increase egg weight with age as expected, or even reducing egg weight.

Alternatively, or following these reductions in the rate of lay, spikes of downgrading in egg quality will reduce the amount of sellable eggs and may trigger negative reactions from the final customer.

The cascade of consequences from infected layers is shown on figure 1.

Undergrade eggs usually cost the same to be produced as good eggs, but they are below the agreed quality standards of the codes of practice by the Egg Producers Federation of New Zealand. Dirty, cracked or broken eggs must be removed from the collection system prior to the grading. Infectious Bronchitis virus will affect epithelial cells and cilia of the oviduct in different degrees, to the point of influencing:

- Shell deposition and egg motility
  - Shell-less (or soft shelled) eggs.
  - Thin shelled.
  - Cracks / leakers
  - Deformed eggs
  - Yolk in peritoneal cavity

- Synthesis of albumen proteins / albumen deposition
  - Lower Haugh Unit

Shell quality: Shell-less eggs are usually destroyed in the cage or equipment, making them not so easy to be accounted for, but they are certainly good contributors to the total amount of dirty eggs (see page 3). More commonly, shell abnormalities such as deformation, loss of colour, thinner shells and additional cracks will contribute to increases in undergrades.

Naturally, thinner shells are more likely to crack and leak in face of the normal mechanical insults that an egg suffers from the moment it comes out of the chicken to the moment the consumer uses it. Under acceptable conditions, cage floor slope, cage wires, conveyor belts, egg stoppers, manual handling, grading machines, packers and transportation will all contribute to an expected percentage of cracks and leakers, which will however increase when eggs have thinner shells.

Moreover, thinner shells have a high conductance, which is the ability to allow the flow of gases through the shell pores. If not detected and culled at egg grading, thin-shelled eggs will lose moisture and CO2 on a faster rate, contributing to a fast deterioration of internal quality while in storage (see Haugh Unit insert page 4).

Mottled eggshells have an irregular thickness, with translucent spots or areas, giving the surface a marbled appearance. They are more commonly found in eggs from older hens. However, excessive complaints from shell motting, even with younger flock, are associated with abnormal function of the shell gland. The causes include disease.

Deformed eggs can be observed, such as wrinkled and corrugated shells. These have likely resulted from inappropriate shell deposition on an unstable watery albumen base. Slab-sided eggs are also seen in cases of IB. They result from poor rotation at shell formation due to retention of another egg in the oviduct. This previously retained egg receives an extra coating of calcium and, by the time it is laid, this layer will be observed in the form of a white ring or just a generally rough surface.

For markets which aim at a uniform dark brown colour for table eggs, occurring infections of IBV will have detrimental effects due to an increase in loss of pigment, increasing the number of bleached appearance on shells.

### Possible effects of IB infections on table egg production

<table>
<thead>
<tr>
<th>Decrease in production</th>
<th>Decrease in egg quality</th>
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<tbody>
<tr>
<td>Delayed sexual maturity</td>
<td>Drop in shell quality</td>
</tr>
<tr>
<td>Drop in production</td>
<td>Shell-less eggs</td>
</tr>
<tr>
<td>Smaller eggs</td>
<td>More cracks</td>
</tr>
<tr>
<td>Egg yolk peritonitis</td>
<td>Deformed shells</td>
</tr>
<tr>
<td>Slow recovery</td>
<td>Thinner shells</td>
</tr>
<tr>
<td>Increased mortality</td>
<td>Loss of colour</td>
</tr>
<tr>
<td>Lower sellable egg output</td>
<td>Higher seconds/undergrades</td>
</tr>
<tr>
<td></td>
<td>Drop in internal quality</td>
</tr>
<tr>
<td></td>
<td>Slower market recovery regardless of brand</td>
</tr>
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<td></td>
<td>Consumer reaction</td>
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*Figure 1.*
Some indicators of IBV infections during egg production. Eggs such as shown below are bound to be encountered under variable percentages on cases of flocks being affected by IBV during the laying cycle, usually at higher rates than the expected for “normal” flocks. However, none of these alterations are pathognomonic signs of Infectious Bronchitis, for they may also appear under other infectious, nutritional or physiological conditions that could be affecting the birds’ egg formation in the oviduct.

The occurrence of an excessive amount of shell-less eggs (or soft-shelled eggs - pictures 1 & 2) may be difficult to detect. In floor/nest operations, usually they will be quickly eaten by the birds. In cage operations, they are easily pushed through the cage floor wiring, disappearing in manure pits or belts, or broken during transport on egg belts or elevators (pic.3). The residues of these broken eggs add to overall soiling of good eggs (pic.4). In addition, shells soiled with egg contents will stick to pulp from trays or cartons. Glued shells (pic.5) will generate new broken eggs, from the egg processing room, to the consumer’s kitchen.

Slab-sided eggs (or flat-sided eggs - pictures 6, 7, 8) result from improper rotation during shell formation. They occur when another egg is retained in the oviduct. While the retained egg gets an extra layer of calcium on its shell (pic.9), the second egg cannot rotate completely, becoming flattened at the point where the eggs touch. This phenomenon can also be, as opposed to an IB infection, a result from stress and sudden changes in the lighting pattern.

Wrinkled and corrugated eggs (pictures 10,11,12,13,14) are usually related to exceptionally thin albumen. Diseases such as Infectious Bronchitis, which may seriously affect albumen deposition, lead to a very fragile matrix of albumen for the shell to be deposited on. The wrinkles and corrugation are caused by the unstable basis and lack of firmness of albumen to support the shell.

Eggs with thin shells are more likely to crack and leak in face of the normal mechanical insults that an egg suffers from the moment it comes out of the chicken to the moment the consumer uses it. The three eggs on pictures 15, 16, 17 were only lightly poked on the shell by the author’s finger. Thinner shells facilitate the gaseous exchange between the egg’s interior and exterior. If not detected and culled at egg grading, thin-shelled eggs will lose moisture and CO2 on a faster rate, contributing to a fast deterioration of internal quality while in storage. Mottled eggs (pic. 18) have irregular - thinner - patches of shell deposition, making the shell structure unstable and more likely to crack. These mottled eggs also lose moisture at a faster rate.
Internal quality:
Although IBV has been reported to increase the occurrence of meat and blood spots, most relevance and studies have been put onto albumen deposition.

Fresh eggs sit up tall and firm when broken out onto a flat surface, while stale or older eggs spread out runny on a wide area, with a thinner albumen and a flattened yolk. As the egg ages, the albumen thins due to a change of protein character in the egg over time. Ocurring infections of IBV can deteriorate the internal quality of eggs, basically by affecting the deposition of albumen proteins during egg formation, resulting in a low rating of Haugh Units.

The low viscosity of the albumen negatively affects its foaming properties, which are essential for the processing industry. Moreover, low ratings of Haugh Units, regardless of the actual “freshness” of the egg, are clearly perceived by the consumer of table eggs as an inferior characteristic, unfit for the edible part of the egg.

When faced with very thin/liquid albumen, consumers will normally reject eggs. At values below 50 HU, they will often be unwilling to purchase eggs again, regardless of brand. This reaction causes lasting effects on the whole egg sector in general. Additionally, IBV is listed as one of the many possible causes of egg yolk peritonitis, or EODES (erratic oviposition and defective egg syndrome). Although this may be an often seen problem in mortality of laying flocks, we will not discuss further points in this article, but just point the matter as being possibly IBV-related.

The universally accepted methodology to measure and define true egg quality and freshness is the Haugh Unit (HU). This formula takes into account egg weight and albumen height and provides a range of HU values from <60 in extremely poor quality eggs to over 100 plus in very good quality fresh eggs. Acceptable standards may vary according to the demands of the market and consumers’ habits.

Commercial layers’ breed standards predict HU values of 103 (first eggs) to 75 (at the end of lay).

General guidelines are given on the table below:

<table>
<thead>
<tr>
<th>Haugh Unit (HU)</th>
<th>Quality of eggs</th>
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<tbody>
<tr>
<td>90+</td>
<td>Excellent</td>
</tr>
<tr>
<td>80</td>
<td>Very good</td>
</tr>
<tr>
<td>70</td>
<td>Acceptable</td>
</tr>
<tr>
<td>65</td>
<td>Fair</td>
</tr>
<tr>
<td>60</td>
<td>Consumer resistance point</td>
</tr>
<tr>
<td>55</td>
<td>Poor</td>
</tr>
<tr>
<td>50</td>
<td>Unacceptable</td>
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</table>

In a cross-Australia surveillance, Roberts observed that average HU for all layer breeds started at 92 at 20 weeks of age, decreasing to 80, at the age of 80 weeks. American researchers observed a natural decline from 89 to 68 HU over a period of 12 months on commercial white layers.

In another study, eggs from Korean commercial poultry farms decreased their HU rating below 70 as the birds got older than 50 weeks of age. Long storage time and high temperatures at storage also contribute to a faster degradation of albumen quality.

Pictured here are eggs with different albumen heights, therefore, different Haugh Units. On picture 1, an excellent egg with 92 HU. On picture 2, an acceptable egg with 76 HU. Pictures 3 and 4 show eggs with very poor HU of 40 and 0 respectively (Not from IB infection, these eggs have deteriorated with age, 2 days past date of expiry. The egg from picture 2 was kept in the fridge. The eggs from pictures 3 and 4 were not!)
Consequences on breeding stock go beyond the same drops in production that commercial egg layers would suffer or the suitability of misshapen eggs for incubation or not. In this case, thinner shells and watery albumen may have profound effects on the embryo development, hatchability, and overall chick quality.

The complex cascade of events triggered from infected breeders on hatchability and chick quality is illustrated on Figure 2. Bronchitis infections may impair male fertility leading to constant lower hatches. In addition, females may have slight drops in production and reduction of egg size—both aspects decrease the cumulative amount of hatchable eggs.

The same eggshell defects from infections of IBV that degrade eggs destined for human consumption make hatching eggs totally inadequate for setting. Cracked eggs should not, under any circumstances, be set into an incubator. They will dry up quickly and, if contaminated, they may pose a serious hazard of unnecessary contamination towards the good eggs and the healthy chicks which should be hatching. Good records of hatching egg quality at egg selection will determine the expected levels of cracks to be taken out before setting, and sudden variation in levels may indicate a deterioration of overall shell quality. Acceptable levels of cracks for hatching eggs vary from one operation to another. North American averages go from 0.4% in the USA to 0.1% in Canada. Deformed or misshapen eggs should always be considered non-hatchable. Although they may be fertile sometimes, these deformed eggs have very low hatchability and will certainly occupy useful incubating space and affect the overall circulation and distribution of heat within the machines, interfering with the incubation of good hatchable eggs. Failure in selecting the appropriate eggs for incubation will lead to an uneven hatch and a progeny with total lack of uniformity.

**Possible effects of IB infections on chick production**

- **Decrease in production**
  - Delayed sexual maturity
  - Drop in production
  - Smaller eggs
  - Egg yolk peritonitis
  - Increased mortality

- **Decrease in egg quality**
  - Drop in shell quality
  - Shell-less eggs
  - More cracks
  - Deformed shells
  - Thinner shells
  - Slow recovery
  - Higher non-hatchable

- **Reduced male fertility**
  - Drop in internal quality Reduced ovolabumin
  - Faster degradation on storage/bacterial penetration
  - Excessive loss of moisture and CO2
  - Higher embryo mortality
  - Higher % of “pips”
  - Stunting, small heart, retarded lung development, enlarged spleen, small chicks

**Less chicks hatched**

**Inferior quality chicks**

Figure 2.

Thinner shells and watery albumen resulting from infections of IB during production in broiler breeders may invariably lead to deficiencies in embryo development. Results will be summarised into less chicks hatched per hen housed and an overall drop on chick quality. Insufficient nutrients or moisture available to the developing embryos can lead to stunting in the broiler shed (above right).
The next and more likely threat on the list is related to thinner shells. Although not always detected by manual handling of hatching eggs, these thinner shelled eggs, when arriving still whole at the selection and pre-incubation stage, will affect incubation negatively in several manners.

Normal or thick-shelled eggs, with a low eggshell conductance (exchange of gases through the shell pores), always achieve the highest hatchability, irrespective of the age of the parent stock. Thus, the thin-shelled eggs have a higher eggshell conductance and lose moisture and CO2 on a higher intensity. The percentage of embryo mortality will be higher, with particular increase on late embryonic mortality especially at the time of pipping. The increased evaporation from thin shells will also cause the hatched chicks to be of smaller size than normal, leading to the presence of runts on the growing period.

Eggs with thinner shells are also more likely to carry bacterial contamination due to the easier penetration of organisms through the pores. Hatchery professionals are well aware of the disastrous effects of contaminated eggs in incubators and hatchers.

On unprotected flocks challenged by a field strain, each of the effects described above could be discreetly occurring under a subtle degree. It is the sum of all these factors indicating sub-optimal production, lower hatchability percentages and suffering chick quality, associated with the evidence of field exposure, which will point for the need of more efficacious flock protection and a monitoring procedure.

**Control and Monitoring**

IBV infections can be effectively prevented by a good immunisation of the layer or breeder flock. The lack of variant strains in New Zealand makes immunisation a fairly straightforward question to be solved and a simple choice of vaccination programmes.

<table>
<thead>
<tr>
<th>Programme 1: live + inactivated</th>
<th>Programme 2: live only</th>
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<tbody>
<tr>
<td><strong>Rearing (age)</strong></td>
<td><strong>Production</strong></td>
</tr>
<tr>
<td>4 weeks: live</td>
<td>(none)</td>
</tr>
<tr>
<td>10 weeks: live</td>
<td></td>
</tr>
<tr>
<td>16-20 weeks: inactivated</td>
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The simplest option is the programme with two live vaccines in the rearing phase and one inactivated just before the beginning of production. The second alternative is the replacement of the inactivated vaccines by repeated live vaccinations, every two months, during the whole period of lay.

Live vaccines are easy to administer on large flocks, for they can be given in the drinking water. They stimulate a good local protection in the target tissues of the bird where the field virus attaches to, inactivated vaccines by repeated live vaccinations, every two months, during the whole period of lay.

The vaccine available is Inacti/Vac IB, an oily emulsion containing inactivated IB antigen of the Massachusetts type, conferring broad protection, which includes the New Zealand IB strains. Inactivated vaccines against Infectious Bronchitis are individually injected on each bird. They are usually given four weeks prior to the onset of egg production to allow time for the immune response to fully develop. Inactivated vaccines establish a solid platform of immunity against the disease, providing the long-lasting protection desired throughout lay. Nevertheless, for a steady uniform and robust protection, the flock must be “primed” by a live vaccine.

**Practical solution:** Programmes that replace the inactivated vaccine are designed to avoid individual handling of the birds; therefore they require the constant exposure of the flock to a live attenuated virus and its shorter length of protection. The repetition of vaccination at every 7-8 weeks mimics the natural build-up and decrease of immunity in unprotected birds challenged by a field strain [such field challenges are often observed as drops in production and peaks of undergrades at every 7-8 weeks]. By offering controlled exposure to a mild vaccine strain, egg producers can prevent these cyclic production losses and deterioration of egg quality.
Monitoring of IB relies not only on the observation of symptoms and a good record keeping of production levels, but also on the follow up of antibody levels on the birds’ circulatory system. Indeed the local immunity has an important role on protection against infection on the mucosa, but also humoral antibodies have their essential role in immunity and their levels can be easily detected by ELISA immunoassays testing.

Birds vaccinated with the IB NZA live vaccine should indicate a titre of between 1000 and 3000 ELISA units before reducing it to lower levels at around 8 weeks post vaccination. Titres above 4000 should indicate a field challenge, especially if uneven individual results, at very high coefficients of variation (CVs’), with some levels going above 9000 ELISA units. On flocks vaccinated with the inactivated Mass type, ELISA titres should go up to 4000-6000, with a good uniformity throughout individuals.

Live vaccines are easy to administer on large flocks, for they can be given in the drinking water (above, left). They stimulate a good local protection in the target tissues of the bird where the field virus attaches to. However, the duration of this immunity is rather limited, when compared to inactivated vaccines. The latter are individually injected on each bird (above, right), establishing a solid platform of immunity against the disease, providing the long-lasting protection desired throughout lay.

Conclusions

Although the classic clinical signs of Infectious Bronchitis may not be clearly seen in susceptible adult birds, infections pre or during the laying cycle may lead to oscillations in egg production and egg quality. These will affect overall productivity, either by reduced revenues from eggs or hatched chicks, or by a negative perception from the clients when purchasing sub-optimum quality products. Due to the absence of the so-called IB variants in New Zealand and the South Pacific, IB control can and should be easily achieved by a comprehensive vaccination programme, covering both rearing and laying periods. Monitoring for IB is an important tool on the control of this disease, involving no more than good record keeping and serological profile.

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