



AUSTRALIAN POULTRY CRC

FINAL REPORT

Program 3A: Welfare

Project No: 05-13

Queensland Department of Primary Industries and Fisheries

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Pilot trial – Mortality in free range flocks

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ISBN 1 921010 26 6

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Published in September 2008

Executive Summary

Free-range is the poultry housing system most actively supported by welfare groups. Currently there are 1.69 million free range hens in Australia (11% of total laying hens in Australia) with an estimated grocery market share of 14.5%. Free range eggs are worth 23% of the value of the Australian egg industry, which is more than \$71 million a year. Mortality in free range flocks can be caused by numerous factors including feather pecking and/or cannibalism, disease, predators and management (diet, housing, strain, rearing, light levels, vaccinations, flock size and density). There is limited scientifically sound information on causes of mortality in free range flocks in Australia and we will start to address this. This pilot study provides survey data on what producers believed were the causes of mortality and preliminary epidemiological data on causes of mortality in an intensive, albeit of limited geographical range, survey.

A survey of all free-range producers in Australia was undertaken to indicate causes of mortality in the industry and to tailor the epidemiological survey to ensure temporal/locality issues were covered. Fourteen free range producers replied to the nationwide survey, five from Queensland, one from New South Wales, and four each from South Australia and Victoria. Fowl cholera was named as the most important cause of mortality in Queensland. New South Wales and Victoria recorded spotty liver as one of their most important causes of mortality with South Australia giving it a low rank and Queensland producers not considering it a problem at all. It is to be thought that, similar symptoms shown by both fowl cholera and spotty liver might have confused producers in their presumption. Queensland and Victoria had problems with predators (foxes/crows and dogs/hawks, respectively) with Victoria also recording heat stress as an important cause of mortality. Egg peritonitis, prolapse/protusion, cannibalism and vent pecking were all seen as important causes of mortality in all states.

In 2006, five flocks of birds from free range farms in Southeast Queensland undertook an intensive epidemiological survey which included a detailed survey, serology and faecal samples of the flock, ongoing mortality records and gross pathology of all mortalities. Flock sizes of the five farms ranged from 1500 – 3500 with all hens (Bond Brown, Bond Black and Hyline Brown) allowed to range in daylight hours regardless of weather conditions. All farms had conventional sheds fixed in one location and fitted with individual nest boxes and perches. All farmers reported that they had vaccinated their flocks for infectious bronchitis (IB), Newcastle disease virus (NDV) and egg drop syndrome (EDS). Overall, all flocks had a positive average NDV, IB and EDS titre throughout the survey; however farms 4 and 5 did not have a uniform distribution of the titres and therefore not a good protection for their flocks. Reproduction tract lesions such as oophoritis, salpingitis, egg peritonitis, and salpingoperitonitis were the most frequently encountered necropsy findings, presumably causing death of laying hens. Cannibalism was the second most common cause of mortality. A follow-up investigation was carried out in 2008 to collect sterile samples from fresh sacrificed birds showing similar symptoms to previous trial for further microbiological tests. It was shown that 85% of birds sacrificed had similar reproductive tract problems. From samples collected (61 in total), 20 did not show presence of any bacteria; gram positive cocci were found in 30 (or 49%) of all samples, while 11 (or 18%) of isolates were gram negative cocci or rods. It is common in commercial poultry that egg laying can be interrupted by stress or an infectious disease. The frequency with which Staphylococcus spp. was isolated suggests an aetiological relationship with encountered lesions. However, only S. aureus is considered to be pathogenic in poultry. To date isolates from live birds have not been associated with human infections or food intoxications. Economic losses occur to producers because egg production drops and mortality increases. Bird welfare could also be compromised.

The nationwide survey questionnaire return rate was disappointing with only fourteen producers returning completed surveys. The survey was developed in consultation with free range producers and an opportunity for the industry itself to have a major input into the direction of free-range research in Australia. Apparently, the survey caused concern that we were unfairly targeting the free range industry and therefore in turn helping the cage layer industry. Recommendations include improvement

of hygienic conditions and biosecurity, approval of more antibiotics and anthelmintics for use in laying hens, and a detailed investigation into the microbial population of the reproduction tract in free range, barn and cage laying hens with the possibility of developing probiotics.

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Introduction

Industry Profile

Free-range is the poultry housing system most actively supported by welfare groups. Birds in free-range systems are housed in sheds and have access to an outdoor range (Model Code of Practice for the Welfare of Animals: Domestic Poultry 4th Ed). This is a traditional system for egg production which still today provides a vision of 'farm fresh eggs' associated with it, where hens can wander at will over green pastures with no environmental restrictions (Kilgour and Dalton, 1984).

Irrespective of a likely gradual increase in demand for free range eggs to eventually become a significant part of the Australian egg market, free range is nevertheless an important sector as it allows consumers the opportunity to exercise their buying preferences; this provision of adequate choice is likely to be an important factor in managing the industry welfare debate. Currently there are 1.69 million free range hens in Australia (11% of total laying hens in Australia) with an estimated grocery market share of 14.5%. Free range eggs are worth 23% of the value of the Australian egg industry, which is more than \$71 million a year.

Margins are slim due to higher production costs, and mortality in free-range systems can be high (21% Kjaer and Sorensen, 2002, 32% Sommer and Vasicek, 2000, and 15-20% Kristensen, 1998) which is unacceptable to the producer, industry and public where negative impacts can be monetary, ethical and/or environmental. Egg production is lower and feed intake is higher under current free range conditions than in cage systems for a number of reasons, including different environmental challenges, energy requirements, strains and husbandry. Large-scale free range production may also be environmentally unfriendly.

There is limited scientifically sound information on causes of mortality in free-range flocks in Australia.

Causes of mortality

Feather pecking and/or cannibalism

Feather pecking is one of the most serious behavioural problems in commercial laying hens, particularly in loose-housing systems where many hens can be affected by only a few hens that engage in feather pecking (McAdie and Keeling, 2000, and Pötzsch et al., 2001). There are four types of birdto-bird pecking. 1. Aggressive pecking is directed by dominant birds at subordinates, and its objective is to establish and maintain dominance and can lead to severe injury or death if the recipient cannot escape. 2. Feather pecking without removal of feathers causes little or no damage and is sometimes directed at particles of litter or food lying on the plumage or around the beak. 3. Feather pulling leading to feather loss is much more forceful pecking. The feather is grasped and firmly pulled, and this may cause the recipient to squawk and withdraw. Hens can become almost completely denuded as a result of this pecking (McAdie and Keeling, 2000). 4. Forceful pecking is often persistently directed at exposed skin and this can lead to haemorrhage. The removal of feathers by feather pecking can result in bleeding from the skin and follicles (especially with immature feathers), which may predispose hens to cannibalism (McAdie and Keeling, 2000). In the flock, feather pecking may result in increased mortality, decreased egg production and increased food consumption (Appleby and Hughes, 1991, Bilčík and Keeling, 1999 and Pötzsch et al., 2001). Huber-Eicher and Audige (1999) investigated the potential risk factors for the occurrence of feather pecking in laying hens raised under commercial conditions on a Swiss farm with more than 500 rearing places. The factors examined considered the most important as being likely to influence feather pecking included stocking density, light intensity, intensity of care, access to elevated perches, access to a roofed and littered outdoor area, time of access to feeding facilities of the housing system, stocking density in the restricted area at the beginning of the rearing period and air quality. The study identified two risk factors and concluded that chicks should be reared at low density and with access to elevated perches.

Feather pecking is considered a welfare problem because it potentially leads to cannibalism especially when exposed skin is injured (Allen and Perry, 1975 and Appleby and Hughes, 1991). Cannibalism has been defined in many ways, the most objective describing it as the "pecking and tearing of the skin and underlying tissues of another bird" (Keeling, 1994). Cannibalism is more common in non-cage systems than cage systems. The most common form is vent pecking. Vent pecking is characterised by damage to the cloaca, the surrounding skin and underlying tissue by a conspecific and can progress to evisceration and death (Pötzsch *et al.*, 2001). It generally occurs soon after the birds have come into lay and may be linked to hormonal changes at the time. Cannibalism causes severe welfare and economic problems in modern egg production.

Disease

The risk of disease spread by contact between birds, or by contact between birds and faeces, is generally regarded as more severe in non-cage systems (Appleby and Hughes, 1991). Contact between birds can include flock mates and wild birds. Free-range poultry and their eggs are more likely to be infected by virus and parasites than caged birds and their eggs (Glatz and Ru, 2004). These poultry are susceptible to the same metabolic diseases affecting intensively housed birds, but the environment can influence their severity and make the birds susceptible to syndromes rarely found in caged layers (Mostert *et al.*, 1995). A comparison of European cage and free-range systems found that free-range hens had more endoparasites and zoonoses (salmonella) (Bestman, 2005). It was found that if mortality exceeded 20% this was often caused by cannibalism however some other causes of mortality included coccidiosis, fatty liver, Infectious Bronchitis, Coli, Amyloidosis and Brachyspira (Bestman, 2005). Following are some brief notes of common diseases and parasites found in poultry.

Infectious coryza

Infectious coryza is an acute respiratory disease of chickens (Blackall, 1999). The main signs of the disease are inflammation of eyes and nose with foul-smelling discharges, conjunctivitis, sneezing and facial swelling. Bacterium causing this disease is *Haemophilus paragallinarum*. Feed and water intake is reduced, leading to loss of weight and egg production in laying birds will drop. Mortality will vary with the virulence of the infection but is generally low. Chickens of all ages are susceptible, with an incubation period of 1-3 days and disease duration of 2-3 weeks (The Merck Veterinary Manual, 2005).

Newcastle disease

Newcastle disease is a highly contagious disease that affects the digestive, respiratory and/or nervous systems of poultry and is caused by a virus of the family Paramyxoviridae (Kahn, 2005 and Saif, 2003). Newcastle disease causes high mortality with depression and death in 3 to 5 days as major signs. Labored breathing with wheezing and gurgling, accompanied by nervous signs, such as paralysis or twisted necks are the main signs. Egg production will drop 30 to 50% before returning to normal levels in about 2 weeks. The disease easily spreads by contact with infected or diseased birds, it is excreted in manure, is expired in the air and can be spread through contaminated equipment, carcasses, water, food and clothing. The virus survives for long periods at ambient temperature, and can persist for 12 months in sheds (faeces and dust) (McMullin, 2004).

Infectious bronchitis

Infectious bronchitis is an acute, rapidly spreading, viral disease of chickens spread by airborne droplets, ingestion of contaminated feed and water and contact with contaminated clothing and equipment (Kahn, 2005). In young chicks IB virus infection causes a cheesy exudate in the bifurcation of the bronchi, thereby causing asphyxia, preceded by severe respiratory distress. In chicks under 3 weeks of age mortality may be as high as 30 or 40 percent. In older birds IB does not cause mortality however egg production will decrease dramatically and deformed eggs with wrinkled shells will often be laid.

Avian influenza

In domestic poultry, avian influenza viruses are typically of low pathogenicity causing subclinical infections, respiratory disease or a drop in egg production (Kahn, 2005). However highly pathogenic avian influenza is a highly contagious viral infection with high mortality. Avian influenza is caused by a virus belonging to the family Orthomyxoviridae. Clinical signs will vary from a drop in egg production to swelling of the head and neck, swollen sinuses with nasal discharge with respiratory involvement. In very severe forms the disease appears suddenly and birds die quickly (McMullin, 2004). The virus is highly concentrated in manure, and in nasal and eye discharges of infected birds and can spread from one farm to the next through contaminated equipment or via faeces on shoes or clothes.

Marek's disease

Marek's disease is caused by a virus belonging to the Herpes virus group. Infected birds show weight loss, or may exhibit some form of paralysis with mortality usually occurring between the ages of 10-20 weeks. The classical form of paralysis with sciatic nerve involvement causes a bird to lie on its side with one leg stretched forward and the other backward. The usual mode of transmission is by aerosols containing infected dander and dust and once the virus is introduced into a chicken flock infection spreads quickly from bird to bird (Kahn, 2005).

Egg drop syndrome

Egg drop syndrome is caused by an adenovirus with the clinical disease occurring at sexual maturity (McMillan, 2005 and Kahn, 2005). Affected flocks show a failure to reach peak egg production or a drop in egg production accompanied by an inferior eggshell quality and in the case of brown eggs, a loss of shell colour. Birds may also appear anaemic, show transient diarrhoea and sometime have reduced feed intake.

Fowl cholera

Fowl cholera is a contagious bacterial disease (*Pasteurella multocida*) of world wide distribution. Affected birds are depressed, have decreased appetite with egg production dropping 5 – 15% and mortality high in acute cases. Birds that die from acute fowl cholera frequently have bluish combs and wattles. Chronically infected birds are considered to a major source of infection with rodents quite often carriers of *P. multocida* (Kahn, 2005). The route of infection is oral or nasal, primarily by excretions from the nose and mouth, along with faeces, contaminated soil, equipment and people (McMillan, 2004).

Salmonellosis

Avian salmonellosis is caused by a group of bacteria of the genus salmonella. Salmonella infections can be transmitted in many ways (contaminated eggs, bird-to-bird contact, contaminated environment and feed). Young birds are more likely to be susceptible to infection than older birds, with a high dose challenge causing intestinal colonization and spread to internal organs, and may be accompanied by diarrhoea. Chicks may show omphalitis.

Coccidiosis

Coccidiosis is one of the more common and costly diseases in poultry and is caused by protozoa of the family Eimeriidae, with most species in poultry being from the genus *Eimeria* (Kahn, 2005). It is characterized by droopiness, paleness of the comb, diarrhoea and occasionally blood in the droppings. The death rate may be quite high, both in chicks and adults. Coccidiosis is spread when one bird eats faecal matter from an infected bird, which contains the infective stage of the coccidia (oocysts). Both

clinically infected and recovered birds shed oocysts in their droppings, which contaminate feed, dust, water, litter and soil, with fresh oocysts not infective until they sporulate (Kahn, 2005).

Worms

Worms of poultry live in the digestive tract and spread from bird to bird via eggs passed out in the droppings. A worm infestation may be indicated by poor growth or decreased egg production, loss of appetite, emaciation, weakness, ruffled appearance, drooping wings, diarrhoea, anaemia and in extreme cases, death. Common types of worms found in poultry are:

- Roundworms (*Ascaridia galli*) Roundworms are white worms approximately 5 12 cm long that occur in the small intestine (McMullin, 2004). Eggs are passed out in the droppings where they are picked up by other birds and hatch in the intestine. In suitable conditions, the eggs remain infective in the soil for up to four months.
- Caecal worms (*Heterakis gallinarum*) These small (07 1.5cm long) worms are found in the lumen of the caeca of poultry. They have a direct life cycle with earthworms and houseflies acting as mechanical transport hosts (McMullin, 2004). These worms must be present in large numbers before a detrimental effect on the bird is noticed however they can harbour the organism which causes the protozoan disease "blackhead".
- Hair worms (*Capillaria* spp.) Hair worms are long thread-like worms found in the crop, oesophagus, small intestine and caecum. Eggs are passed out in droppings and must be ingested by an intermediate host (i.e. earthworm), with the life cycle completed when the bird eats the earthworm and the parasite are released into the gut (Permin and Hansen, 1998). Infections with hair worms can be highly pathogenic for birds kept in deep-litter or free range systems where big numbers of infective eggs may build up in the litter or soil (Permin and Hansen, 1998).
- Tapeworms Poultry reared under free range conditions can become infected with tapeworms. All tapeworms of poultry have indirect life cycles with intermediate hosts (i.e. earthworms, beetles and flies) essential to perpetuate the life cycle (Permin and Hansen, 1998). They range in size from 5 − 15 cm in length and are segmented and ribbon-like. Eggs form in the segments which break off when they are ripe and are passed out in droppings.

Endoparasites - mites

Three types of mites are of economic importance to the poultry industry, the tropical fowl mite, red mite and scaly leg mite. The tropical fowl mite (*Ornithonyssus bursa*) is found in tropical and subtropical areas where wild birds carry the parasite from farm to farm. These mites can be found on the skin of the birds throughout the day whereas the red mite (*Dermanyssus gallinae*) is only found on the host during the night. During the day it retreats into cracks and crevices in the poultry shed and equipment. Infected birds may have a change in behaviour due to the itching effect of the mites. Weight loss, decreased egg production, anaemia and death are clinical signs with mites also being able to transmit a number of diseases (i.e. fowl pox and Newcastle disease) (Permin and Hansen, 1998). The scaly leg mite (*Cnemidocoptes mutans*) is found under the scales of the legs of birds with birds being infected from the ground. These mites tunnel into the skin and cause the scales to lump up and form crusts (McMullin, 2004). This causes keratinisation of the legs and in chronic cases lameness and malformation of the feet are seen.

Predators

Birds kept in free-range systems are at risk of predation from foxes, wild cats, dogs, snakes, eagles and hawks. The establishment of a proper fence and closing pop-holes at night may prevent attacks from foxes, wild cats, and dogs; however, predation from eagles and hawks is harder to control.

Management

This includes factors such as diet, strain, rearing, light levels and social pressure arising from type of feeders, drinkers and nest boxes.

Diet

Feed intake is influenced by housing system, with birds on the floor, particularly free-range, consuming more feed than birds in cages. This increase is presumably in response to a greater requirement for energy resulting from increased activity and lower temperatures generally prevailing in non-cage environments. If birds do not achieve sufficient feed intake their bodyweight will be low. In commercial barn systems, mortality was markedly increased in flocks that did not reach the appropriate body weights at 26-29 weeks of age (Parkinson and Cransberg, 2002). The authors found that as the body weight of the pullets increased, egg production and mortality decreased. Diagnostic studies of the underweight flocks indicated problems with egg peritonitis and salpingitis (Parkinson and Cransberg, 2002). A change in diet can also cause problems in free-range systems. A new diet can cause reduced or increased palatability of the food and therefore result in decreased intake or increased competition for food, leading to stress and frustration, which are reported risk factors in vent and feather pecking (Pötzsch *et al.*, 2001). A new diet may also cause diarrhoea, leading to an inflamed and reddened vent and soiled areas around the vent; these may attract pecking from flock mates.

Strain

Within a given management system, strain of bird is an important factor affecting husbandry requirements and economic performance. This is especially true in alternative systems with their more diversified environments and a correspondingly wider range of opportunities to exhibit behaviours not shown in cages. A wide selection of strains is now available and many producers believe that some of these are more suitable than others for free-range egg production under Australian conditions. Strains may differ in behavioural traits such as feather pecking, foraging activity, flightiness, broodiness and inclination to lay in nest boxes. These differences have repercussions both for the welfare of the birds and the manageability of the system. Propensity for cannibalism is especially important as beak trimming is prohibited in some accreditation schemes for free-range production.

Drinker type

Flocks with hanging bell drinkers are more at risk of vent pecking and feather pecking than flocks provided with water via other systems (Pötzsch *et al.*, 2001). Bell drinkers provide a focus for crowding and competition and consequently increase the level of stress. They are usually located near nest boxes where activity, 'tension' and severe feather pecking are more frequent that in other parts of the hen house (Nicol *et al.*, 1999).

Rearing

It has been suggested that pullets should be reared in a system identical to that they are to be housed in during the production period. Access to perches from no later than 4 weeks of age decreases both the prevalence of floor eggs during the early production period and the prevalence of cloacal cannibalism during the whole production period (Gunnarsson *et al.*, 1999).

Light Levels

Low light intensity in the hen house is frequently used to control vent and feather pecking. Illuminated nest boxes are generally screened with curtains to provide a sharp light-gradient from the outside to the inside of the nest boxes (Pötzsch *et al.*, 2001). The change in light intensity experienced on entering an illuminated communal nest box may increase the visual attractiveness of the cloacal mucosa of a hen that has just laid an egg. Pötzsch *et al.* (2001) found that the use of light to encourage the use of nest boxes in comparison with no nest box light showed the strongest (positive) association with vent

pecking. Dimmed light also affects eyesight development which reduces the welfare of the bird (Prescott *et al.*, 2003).

Flock size and Density

While cage housing imposes a particular spatial organisation, alternative systems in most cases provide hens with the opportunity to space themselves in relatively unconstrained ways. In larger groups of birds vent pecking is more likely to become a problem since more birds can detect and attack minor wounds and prolapses (Allen and Perry, 1975). Nicol *et al.* (1999) studied laying hens in a perchery at four stocking densities (6, 14, 22, or 30 birds/m²). They found that as flock sizes and stocking density increased the number of aggressive pecks increased. It is possible in larger flocks some birds rarely or never gained access to the floor areas, and that the increased pecking on the perches represents a redirection of frustrated ground pecking in these birds (Nicol, *et al.*, 1999). Bilčík and Keeling (1999) also found a strong influence of group size, with poorer plumage condition in larger group sizes.

Vaccinations

Health management involves an integrated program of precautions, procedures and treatments. The aim is to prevent disease occurring in the flock and if it does occur, to reduce the physical and financial loss associated with it. Vaccination increases the bird's natural immunity against specific infectious micro-organisms, such as viruses, bacteria and protozoa. It protects bird health and enhances bird welfare. Some vaccinations are compulsory, for example, Newcastle Disease.

Beak trimming

Beak trimming is not wholly effective at preventing feather pecking and cannibalism, and there is continuing public concern that it may cause chronic pain. Beak trimming (debeaking, or partial beak amputation) is a procedure widely used by the poultry industry for reducing the incidence and harmful effects of feather pecking, aggressive peaking and cannibalism.

Beak trimmed fowl have a lower rate of mortality, eat slightly less food and have a slightly improved food conversion ratio. The improved feed conversion is because the birds waste less food, or because they have better plumage so there is less food required for maintenance of normal body temperature, or because they are less active.

Adverse effects for the producer are generally slight. There is a small cost in labour and materials for the actual trimming and there may be a small increase in chick mortality in the first few days of life if the trimming has been too radical. There are also widespread negative effects to the image of the poultry industry.

Aim

This pilot study is the first scientific evaluation of the major causes of mortality in commercial freerange flocks in Australia and provides extensive survey data on what producers believe are the causes of mortality and preliminary epidemiological data on causes of mortality in an intensive, albeit of limited geographical range, survey. These outcomes will be utilised together to develop a framework for a nationwide study into causes of free-range mortality; this proposed nationwide study is not part of this pilot project. Anticipated benefits include both industry education and targeted research ideas to improve hen welfare. This will address community concerns as well as increasing productivity and profitability of the free-range system. This framework document will be an outcome of this pilot project.

Objectives

The objective of this project was to undertake a pilot study to develop a framework for a national survey on causes of mortality in commercial free-range flocks through:

- A survey of all free-range producers in Australia to indicate causes of mortality in the industry
 to tailor the epidemiological survey to ensure temporal/locality issues are covered. This aspect
 of the project will also involve networking with industry veterinarians and state industry
 departments.
- A small focused epidemiological study on the causes of mortality in commercial free-range flocks in Southeast Queensland. This aspect of the project will indicate the level of sampling and testing that is required to provide rigorous data (for a more comprehensive epidemiological survey in the future) and will include a detailed survey of participating farms, ongoing mortality records, serology, faecal samples and gross pathology.
- A follow-up investigation into the causes of reproductive tract lesions found in hens in pilot
 epidemiological study (September to December 2006), and determine the type/strain of
 bacteria that causes this infection. Determination of the typing of bacterial infection in free
 range flocks (particularly of the flocks that were most suspected for infection in the initial
 survey) would allow for more specific information regarding prevention and control measures
 for producers.

Methodology

Nationwide survey

The first part of the study involved a nationwide survey of all (~250) free-range producers. A draft survey was developed and included questions such as size of farm/flock, type of shed, source of birds (reared on farm or bought at point of lay), age of farm, beak trimming (yes or no), vaccination protocol (list of common vaccinations – indicate which ones they do), wild birds (list of types of birds, seasonal effects), causes of mortality (list of causes – indicate in order what are the most common on their farm and age relatedness. As well as questions on who was determining cause of mortality – veterinarian, pathology laboratory or farmer and the use of professional services.

The draft survey was tested and revised using a focus group comprised of people who were likely to be interested in the survey outcomes and also people who represent the survey targets (Salant and Dillman, 1994) and included a free range producer, poultry epidemiologist, senior poultry extension officer and poultry scientist. After revision by the focus group the survey was accepted as completed (appendix A).

Prior to distribution of the survey, state free range associations and egg producer groups were notified of the survey (appendix B) and asked for number of surveys required for members and/or member addresses (appendix C). Flyers were also distributed at P.I.X. in April 2006 (appendix D). A total of 207 surveys were distributed along with a covering letter (appendix E) and reply paid envelope. A reminder was sent to producers and due to the low response rate, a letter was sent extending the time available to complete the survey (appendix F). Distribution of the surveys is shown in table 1.

Table 1: Distribution of nationwide free range mortality survey.

State	Contact	Number of surveys
South Australia	Farmers Federation	50
	Individual Producers	2
Victoria	Free range Association	55
	Individual Producers	10
New South Wales	Farmers Association	2
	Free range Association	12
	Individual Producers	25
Queensland	Free range Association	50
Western Australia	Individual Producer	1
TOTAL		207

Epidemiological survey

Five flocks of birds from free range farms in Southeast Queensland undertook an intensive epidemiological survey (five flocks were chosen as the appropriate number for a pilot trial based on the indicative overall budget, costs of post-mortems and obtaining and processing serology samples). Each flock completed a detail questionnaire relating to the flock being studied and included questions on breed, rearing, shed type, vaccinations, nutrition, egg production, age of flock, management practices (appendix G).

Serology

Blood samples were taken from flocks at the start and end of the experimental period to test for IB, NDV and EDS. ELISA titres of these diseases are very useful to monitor success of vaccination and can also be used for diagnosis of field infection. Rising EDS titres could indicate reinfection from wild birds which is important in relation to possible avian influenza infection from wild birds.

Newcastle disease is a highly contagious disease that affects the digestive, respiratory and/or nervous systems of domestic poultry, cage and aviary birds and wild birds. The National Newcastle Disease Management Plan requires that all states and territories of Australia make Newcastle disease vaccination compulsory. Blood samples were tested to ensure adequate serological titres in flocks.

Infectious bronchitis has been connected to declines in egg production and quality, including thinning of the albumen component, lightening of the shell colour, reduced shell thickness, decrease in egg weight and increased incidence of abnormal shell formation. Currently all pullets destined for commercial egg production in Australia are vaccinated against IB.

Birds were sampled at random based on any divisions (physical or theoretical) established within the shed. For example, feeders in rows may reduce movement of birds from one row to another. A proportion of all birds caught were sampled and birds caught by different catchers were adequately represented. The number of birds sampled was as recommended by Birling Avian Laboratory and Melbourne University International Avian Health Laboratory (ratio of 30 samples per flock of 4000 birds – 0.75%). Thus for the 5 flocks a total of 82 (0.84%) samples were taken. Samples were taken using individual syringes and individual blood tubes from the wing vein using a 27 gauge x 1 cm needle attached to a syringe. Each bird was appropriately restrained to ensure as little stress as possible on the bird and for ease of blood collection (Figure 1).

Figure 1: Appropriate restraint of free range layer hen for wing vein blood sampling.



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Feathers along the ventral part of the wing overlying the vein were plucked to accentuate the vein. The needle was then inserted toward the body along the length of the vein and blood withdrawn (Figure 2). The vein was held after removing the needle to prevent haematoma. The blood was then transferred from the syringe to a 10 ml heparinised vacutainer tube after removing the needle.

A flock profile was obtained by testing with commercially available ELISA (Enzyme-Linked Immunosorbent Assay) test kits (Guildhay Ltd, UK) that measured the amount of antibody titre to NDV, IB and EDS vaccines from a single plasma sample. Plasma was separated by centrifugation and stored in -20°C until the test was carried out. Antibody titre against IBV, NDV and EDS were measured in duplicates and results given as a Log10 of the titre in the plasma following the producer's instructions. Results were interpreted as negative, suspect and positive. However in interpreting results, factors like age and vaccination/disease history of the flock had to be taken into consideration.

Figure 2: The needle is inserted into the wing vein towards the body of the hen and blood withdrawn.



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Faecal samples

Faecal samples were collected to determine parasite burden including worm burden and coccidia oocycst counts per gram using methods described by Sloss *et al.* (1994) pp 9 and 79-87 (Figure 3).

Figure 3: Collection of faecal samples for determination of parasite burden.



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Necropsy

All dead birds from the survey shed were collected, recorded, labelled and sent for gross pathology for three months. Farms were provided with freezers to store dead birds and were required to record date of mortalities and label carcasses for identification (appendix H). All birds collected were delivered to the pathology lab where the cause of death was identified (appendix I). The necropsy included the body weight of the bird and an examination of the overall condition, as well as external and internal observations. The tentative diagnosis was based on crucial clinical macroscopic lesions on organs. Microbiological samples are not recommended for birds that are found dead for more than 3-6 h. Given that, the majority of dead birds submitted for post mortem examination showed lesions of reproductive tract such as oophoritis, salpingitis, egg peritonitis, salpingoperitonitis, and impacted oviduct, a follow-up investigation was undertaken to find a relationship between pathology findings and possible bacterial implications.

For the follow-up investigation, live birds showing similar symptoms to pilot trial or morbid-looking birds were sacrificed and examined. Microbiological samples were taken from all birds showing reproductive tract problems. The bacteriological examination followed the methodology adopted from "A laboratory manual for the isolation and identification of avian pathogens" 4th Edition published by The American Association of Avian Pathologists (Swanye *et al.*, 1998). Samples from liver, spleen, ovary, oviduct, intestinal content, and cloacae were collected aseptically and plated on nonselective (Columbia blood agar) and selective plating media (MacConkey agar, chromogenic Salmonella, chromogenic E. coli etc.). The plates were incubated at $37\pm0.5^{\circ}$ C overnight. At least three well separated colonies per plate were selected for transfer to more selective plating media.

Bacterial isolates were identified by growth requirements, colony morphology, and cell morphology and staining characteristics (see Appendix J). As Gram positive cocci were the most abundant isolate further tests were conducted for this isolate. *S. aureus* is fermentative for manitol, therefore, Gram positive cocci were further transferred onto a selective medium inhibitory for Gram negative bacteria such as Manitol Salt Agar (MSA) plates. This was also needed for differential recovery of isolates. To confirm the presence of coagulase positive or coagulase negative staphylococci and further differentiate *S. aureus* from other *Staphylococcus sp*, isolates were tested for coagulase reaction (Staphytect Plus; Oxoid). No further identification/typing was made for *S. aureus* and/or other isolates.

Results

Nationwide Survey

General

Fourteen free range producers replied to the nationwide survey, five from Queensland, one from New South Wales, and four each from South Australia and Victoria. Six of the poultry farms had been operational for less than 10 years, three for 10 - 20 years, 4 for 21 - 50 years and one for more than 50 years. However when asked how long they were operating as a free range farm, one farm in Queensland that had been a poultry farm for more than 50 years had only been a free range farm for less than 10 years. The producer from New South Wales had also been in the poultry industry for 21 - 50 years however had only had free range poultry for 10 - 20 years. The average number of free range layer hens per producer per year ranged from less than 1000 (6 producers) to 50 000 - 100 000 (1 producer), with the number of sheds per farm ranging from less than five (10 producers) to between 21 and 35 (2 producers). Forty-three percent of farms had a maximum of less than 500 layers per shed, with 7% having between 501 – 1000 layers per shed, 29% having 1001 – 300l layers per shed and 21% having more than 3001 layers per shed. The size of range area available ranged from 100 m² to 2500 acres.

All farms had wild birds regularly visiting (table 2) with 9 farms having water courses/dams on the property.

T 11 0	TT7.1 1	7 . 7	1 1		C		C	
Table 2.	Wild	hirds	regularly	visitino	tree	range	tarms	in Australia.
I cioic 2.	111111	outus	regularly	Visiting.	,,,,,	Tunge.	juinis	in Thisircitic.

	Number of farms					
Type	Summer	Autumn	Winter	Spring		
Ducks	3	4	4	3		
Magpies	9	9	9	10		
Crows	11	9	9	11		
Mynas	4	4	3	3		
Hawks	9	9	9	10		
Eagles	8	6	5	8		
Ibis	4	4	3	4		
Plovers	5	4	4	4		
Kookaburras	7	7	7	7		
Swallows	6	3	3	8		
Pigeons	7	7	7	7		
Swamp wading birds	1	1	1	1		
Ravens	2	2	2	2		
Sparrows	1	1	1	1		

Housing

Thirteen of the 14 farms had conventional sheds made of metal and timber with two farms having igloo type sheds, one made of canvas and one of polypropylene. Of the 10 farms that had permanently fixed sheds, only three used pasture rotation. Victoria and South Australia had some farms with mobile sheds and movement of these sheds ranged from every couple of days to 5 weeks. Two producers used automatic nest boxes with the remaining farms using individual or colony nest boxes that required manual pick up. Nest boxes were made of wood (4 producers), metal (9 producers) and plastic (4 producers). One producer in South Australia used slats, with all other producers using either A-frame or single level perches.

Hens

Table 3 shows the predominant breed/strain of birds that are used in the farms surveyed with all farms except one having hens of a single age in each shed.

Table 3: Predominant breed/strain of bird used in the free range farms surveyed.

	Breed/strain							
Location	Hyline	Bond HiSex Log Durant Commercial F						
	Brown	Brown	Brown	Isa Brown	X B's	Black		
Queensland	2	3	1			1		
New South Wales				1				
Victoria	2		2	2				
South Australia	1		1	1	1			
TOTAL	5	3	4	4	1	1		

Fifty percent of farms reared their own birds on farm. Regardless of whether birds were reared on or off farm only one farm had birds reared on wire, with all other birds reared on the floor. Of the farms that reared off farm, 72% bought in replacement pullets at 13 - 16 weeks of age with the remaining two farms buying their pullets in at 5 - 8 weeks old and greater than 17 weeks, respectively. Table 4 shows at what age pullets started ranging.

Table 4: Age pullets start ranging.

	Age pullets start ranging							
Location	1-3 weeks	4 – 6 weeks	7 – 12 weeks	13 – 18 weeks	>18 weeks			
Queensland	1	3	1					
New South Wales					1			
Victoria	1	1		1	1			
South Australia			1	1	2			
TOTAL	2	4	2	2	4			

Hens were disposed of at ages ranging from 55 weeks to greater than 95 weeks, with 64% of producers disposing of hens when they were 76-85 weeks of age. Eight out of the fourteen producers replaced hens on an all in all out basis. Eight producers also used a 16:8 hour lighting regime. The average rate of production ranged from 60-65% to greater than 85%, 72% of producers averaging between 66 and 85%. Floor eggs were not considered a problem with all producers reporting less than 2%.

Management

All producers reported that their hens were allowed to range in all daylight hours with all but one allowing hens to range regardless of weather conditions. Three producers beak trimmed their birds as a matter of routine. All producers undertook a regular rodent baiting programme with 50% baiting only as required. One producer did not undertake regular worming of their birds, with 54% of producers who undertook worming did so tri-monthly and the remaining producers worming as required. Eight of the 14 producers regularly treated their birds for external parasites, with 75% on an as required basis. Table 5 shows what measures are taken in between flocks of hens in a particular shed.

Table 5: Measures taken in between flocks of hens in a particular shed.

	Between flocks of hens							
Location	Remove litter	Disinfect shed	High pressure clean	Leave shed empty	Nothing			
Queensland	5	4	4	1	1			
New South Wales	1	1	1	1				
Victoria	2	4	4	4				
South Australia	2	2	3	3				
TOTAL	10	11	12	9	1			

Table 6 shows what diseases producers vaccinate their flocks against.

Table 6: Vaccinations received by free range flocks.

	Location (number of producers)							
Vaccination	QLD	NSW	VIC	SA	TOTAL			
	(5)	(1)	(4)	(3)	(13)			
IB (infectious bronchitis) virus	5	1	4	2	12			
ILT (infectious laryngotracheitis)	1	1	4	2	8			
Marek's disease	5	1	4	3	13			
NDV (Newcastle disease virus)	4	1	4	3	12			
Fowl pox	4	1	3	2	10			
Coccidiosis	3		2	1	6			
Infectious coryza	1		1		2			
MG (Mycoplasma gallisepticum)	3		1		4			
AE (Avian encephalomyelitis)	2	1	2	1	6			
EDS (Egg drop syndrome) inactivated	3	1	3	1	8			
Fowl cholera	3	1	1	1	6			
MS (Mycoplasma synoviae)	2				2			

Producers were asked, if known, what were the main causes of mortality in their birds (including deaths and culls) and at what age did they occur. They were asked to rank in order of importance from 1 (most often the cause of mortality) to 10 (least often the cause of mortality) (Table 7).

Table 7: Causes of mortality and rank of importance with 1, most often the cause of mortality, to 10 least often the cause of mortality.

	Location							
	(QLD NSW		7	VIC	SA		
Cause	Rank	Age(wks)	Rank	Age(wks)	Rank	Age(wks)	Rank	Age(wks)
Fowl cholera	1,1	>18						
Marek's disease			8	15	9		10	
Salmonella sp.					10			
Coccidiosis			7	Summer	8			
Chronic respiratory					7			
disease					/			
Spotty liver			2	Summer	1	All	8	30-40
Worms			6	15-20	6			
Egg peritonitis	2,2				3,5	>38		
Prolapse/protrusion			3	30-40	4,6,8		1,9	>24
Cannibalism	2,2,8	>18	4	30-40	1,4,7			
Vent pecking	3,9		1	30-40	2,3,5		2	>24
Physical injury	8		5	15	5,7			
Predators	1,2,8		9		1,1,3,6	All		
Heat Stress			10	Summer	2,2,2,4	Old		

The owner/manager of the free range farm most often determined the cause of mortality (80 - 100% of the time) with veterinarians and pathology labs determining cause of death 1 - 20% of the time.

Discussion

The objective of the nationwide survey of free range layer producers in Australia was to indicate causes of mortality in the industry to tailor the epidemiological survey to ensure temporal/locality issues were covered. Of the fourteen free range producers who replied to the survey the results show that housing conditions, hens and management did not show any particular trend when looking at individual states.

Fowl cholera was named as the most important cause of mortality in Queensland however was not considered a problem at all in the other states. Fowl cholera is mainly transmitted from bird to bird by water and feed contamination with rodents also playing a role in contamination. All farms recorded that they undertook regular rodent baiting. It is also found in puddles of water and dams in the range area and four of the five farms in Queensland reported that there are water courses/dams on their However all other states also recorded farms with water courses/dams and did not report cholera problems. Stress conditions (overcrowding, cold weather, unhygienic sheds and poor ventilation) can also trigger infection outbreaks. Cold weather is unlikely to be a problem in Queensland and all but one farm reported undertaking rigorous cleaning of sheds between flocks. Three of the five Queensland respondents vaccinated their flocks against fowl cholera however as this vaccine contains the three most common serovars in Australian poultry cross protection to any of the other 13 serovars is likely to be limited.

New South Wales and Victoria recorded spotty liver as one of their most important causes of mortality with South Australia giving it a low rank and Queensland producers not considering it a problem at all. Spotty liver generally occurs in flocks kept on the ground either in a shed (barn) or where they birds are free to range. Liver lesions look a lot like that produced by *Pasteurella multocida* (fowl cholera) and flocks respond well to antibiotic treatment however they often relapse (Critchley, 2002).

The similar symptoms shown by both fowl cholera and spotty liver suggest that it is possible some flocks reported as dying of fowl cholera are actually suffering from spotty liver (although the cause of the condition is not clear, but Campylobacter jejuni has been implicated as the aetiological agent) and vice versa. This would mean Queensland, New South Wales and Victoria could all have spotty liver/fowl cholera as one of their major causes of mortality. It was also found that 80 - 100% of the time that producers determine the cause of mortality on their farm and with the symptoms of the two conditions being similar they could report the one they are more familiar with.

Queensland and Victoria had problems with predators (foxes/crows and dogs/hawks, respectively) with Victoria also recording heat stress as an important cause of mortality. The heat stress result was interesting with temperatures in Queensland, New South Wales and South Australia generally considered higher than Victoria. It is possible in these states where high temperatures are more the norm; producers could employ measures in their sheds to help combat this problem (fans, foggers). Another possibility is that it is the extremes of temperature that are the problem with birds acclimatised to a milder temperature suddenly experience a very hot day/week causing heat stress mortality (Daniel and Balnave, 1981).

Egg peritonitis, prolapse/protusion, cannibalism and vent pecking were all seen as important causes of mortality in all states. Therefore the main differences between the states in causes of death were fowl cholera and spotty liver and heat stress in Victoria.

Epidemiological Survey

Flock sizes of the five farms ranged from 1500 – 3500 with all hens allowed to range in daylight hours regardless of weather conditions. All farms had conventional sheds made of steel/colourbond or corrugated iron, with sheds fixed in one location. Individual nest boxes were used on all farms with two farms having wooden nest boxes, two with plastic and one metal nest boxes (Figure 4). All farms provided perches for their hens (Figure 5).

Figure 4: Example of individual plastic nest boxes in a free range shed.



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Hens in the survey sheds were Bond Brown, Hyline Brown and Bond Black with all rearing on-farm on the floor (Figure 5 and 6). All hens were placed in laying sheds between 12 - 16 weeks with pullets allowed to start ranging between 3 - 6 weeks of age. Three of the five flocks were under a 16:8 hr lighting regime, with one flock beak trimmed at 14 weeks.

All farms undertook regular rodent baiting and only one farm did not undertake regular worming of the flock. All farms had dedicated footwear and signage as their biosecurity measure with two farms also having visitor sign-in books and one farm a stand-down period. All farms vaccinated their flocks for IB, Marek's, NDV, fowl pox, MG, EDS, fowl cholera and MS. Two farms also vaccinated against

ILT, with four farms vaccinating against AE and providing in-feed coccidiostat. Nutritionists developed diets for all flocks with all using starter, grower and layer diets.

Figure 5: Hyline layers, perches, feed and watering systems in a free range shed.



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Figure 6: Bond black layers on a free range farm.



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Serology

Eighty-two birds over the five farms had blood samples taken and analysis completed at the start of the reporting period. At the end of the survey 65 samples were taken from four farms (1 farm had depopulated before samples could be taken). This represented 0.84% and 0.86% of total birds surveyed at the start and end of the trial, respectively, compared to 0.75% as minimum recommended sampling. Results of the serological analysis are shown in Tables 8 and 9.

When evaluating ELISA titres, one always has to look at mean titre response of the tested birds, and the coefficient of variation (% CV). The mean titre of the tested birds within a flock tells you how strong the antibody response is of a flock after vaccination. It basically provides you with a measure of the immune response of your flock. The second parameter, CV%, provides you with an indication on how variable a mean titre response of a flock is (the lower the % CV, the more uniform distribution of the titres and the better the vaccination). For most diseases, the % CV after a correctly applied

inactivated vaccination should be less than 40%. For live vaccine applications, the % CV should be less then 60%. With live priming, complete sero-conversion is more important than % CV and one should check if all birds test positive.

All farms had positive initial EDS titres (average 3397) with farms 1 and 3 also having 100% of samples with a positive initial IBV titres (4721 and 3734, respectively). Farms 2 and 5 had an overall positive initial IBV titre however farm 2 had 20% of samples with suspect results, and farm 5 had 30% of samples with suspect results and 15% of samples with negative titres. Farm 4 had an overall negative IBV titre (985) with only four samples out of 15 showing a positive result. Farm 1 was the only farm with all samples showing positive initial NDV titres (average 4407). All other farms had an overall positive initial NDV titre however each farm had a percentage of samples that were suspect or negative (Farm 2-13% suspect, 7% negative; Farm 3-18% suspect, 18% negative; Farm 4-13% suspect, 33% negative and Farm 5-10% suspect).

All farms except farm 2 showed a decrease in EDS titre from the start to the end of the survey however the titres of the four farms sampled remained positive (average 3141). Farm 1 had all positive samples, farm 2 had two negative results, farm 4, two negative and one suspect result and farm 5 one suspect result. IBV titres were positive for all farms at the end of the survey (average 3053) with farms 1, 2, and 5 having all positive samples. Farm 4's titre had increased over the period of the survey from negative (985) to positive (1737), with the number of positive samples increasing from 4 to 7 out of 15. As with the initial NDV titre, all farms had an overall positive titre (average 3318) with farms 1 and 2 having all positive samples. Farm 4 had three suspect and one negative sample and farm 5 had two suspect and one negative sample.

One should take in consideration that, factors like age, vaccination method and disease history of the flock could have influenced the vaccination titre. However, except for Farm 4, all farms have had a good protection against IB, NDV, and EDS.

Table 8: Percentage of positive (+), suspect (s) and negative (-) EDS, IBV and NDV titres from blood samples taken at the start and end of the survey period $(ns - no \ sample)$.

	EDS (%)					IBV (%)				NDV (%)								
]	Initial	-		Final]	Initial			Final]	Initial	l		Final	
Farm	+	S	-	+	s	-	+	s	-	+	s	-	+	s	-	+	S	1
1	100			100			100			100			100			100		
2	100			87	13		80	20		100			80	13	7	100		
3	100			ns			100			ns			64	18	18	ns		
4	100			80	13	7	27		73	46	27	27	54	13	33	73	20	7
5	100			95		5	50	30	20	100			90	10		85	10	5

Table 9: Individual EDS, IBV and NDV titres from blood samples taken at the start and end of the survey period (ns – no sample).

		iou (ns	– no sampi	-)•	T:4:-1		Einal		T:4:-1		Einal	
Farm	Initial EDS (titre)	Status	Final EDS (titre)	Status	Initial IBV (titre)	Status	Final IBV (titre)	Status	Initial NDV (titre)	Status	Final NDV (titre)	Status
1	2062	+	1733		5756		3392		4087		3459	+
1	2209	+	2673	+	4515	+ +	4433	+	4297	+ +	1321	+
1	1993	+	2456	+	4213	+	3989	+	2211	+	3437	+
1	2768	+	1670	+	2144	+	3250	+	2287	+	6281	+
1	2792	+	3671		4176	+	3016	+	8444	+	3055	
1				+								+
1	1880 2161	+	2414 1679	+	5202 5371	+	3334 4856	+	4389 3060	+	3816 3367	+
1	3048		3139		5162		4799		2616		5160	+
1	2789	+	1478	+	3944	+	3036	+	4643	+		+
1	2475	+	2566	+	5531	+	4157	+	4043	+	4631 5597	+
1	3252	+	2197	+	5130	+	4742	+	2030	+	3901	+
1	1409	+	2601	+	3348	+	4511	+	7191	+	3764	+
1	3659		2139		4117		4256		4597		4199	
1		+		+	5990	+		+		+		+
1	3161 3643	+	1876 3548	+	6224	+	3414 3110	+	6867 5324	+	6110 3244	+
Mean	2620	+	2389	+	4721	+	3886	+	4407	+	4090	+
Std Dev	666	+	674	+	1092	+	691	+	1913	+	1299	+
CV %	25		28		23		18		43		32	
2	3493		4576		3593	1	2482	,	3351	+	3616	
	3107	+	3581	+	2165	+		+	5076		3223	+
2	3604	+	5482	+	2499	+	2183 4453	+	2338	+	3262	+
2	3441	+	5632	+	3649	+	4341	+	4505	+	4901	+
2	1929	+		+	3928	+	4441	+	3448	+	4901	+
		+	5667 5987	+	3561	+	4443	+	3876	+		+
2 2	2004 3499	+		+		+		+		+	4000	+
		+	6205	+	4109 4072	+	4784	+	4204	+	4899	+
2 2	3722 3607	+	4025 6730	+	1126	+	4219 3589	+	1139 1362	- suspect	2781 2764	+
2	3438	+	6368	+	1787	suspect	4657	+		suspect		+
2		+	4206	+		+		+	2262	+	2430	+
2	3666 3571	+	835	+	1185 1298	suspect	3621 4571	+	1768	suspect	2745	+
2	3275	+	984		2350	suspect	2263	+	2313 2690	+	2543 2401	+
2	2597	+	3757	-		+		+	4894	+	2346	+
2	2942	+	6600	+	3265 1961	+	2945 2742	+	3519	+	2110	+
Mean	3193	+	4709	+	2703	+	3716	+	3116	+	3207	+
Std Dev		+		+		+		+		+		+
CV%	582 18		1860 28		1092 23		691 18		1913 43		1299 32	
3					4094				2499	guamaat		
3	4015	+	ns			+	ns			suspect	ns	
3	3765 3705	+	ns		3585 2557	+	ns		2362 2362	suspect	ns	
3	3512	+	ns		3382	+	ns		4064	suspect +	ns	
3	2124	+	ns		2818	+	ns		3899	+	ns	
3	3838	+	ns		3205	+	ns		6650	+	ns	
3	3771	+	ns		5287	+	ns		3662	+	ns	
3	4171	+	ns		3672	+	ns		3231	+	ns	
3	3854		ns		1611		ns		2167	-	ns	
3	3195	+	ns		4543	+	ns		2938	-	ns	
3	3659	+	ns		4408	+	ns		2364	+	ns	
3	3659	+	ns ns		4408	+	ns ns		2591	+	ns	
3	3854	+			4648	+			4273	+	ns	
3	3795	+	ns ns		3447	+	ns ns		2211	+	ns ns	
3	3445				4536				7576			
3	4133	+	ns		3464	+	ns		2210	+	ns	
3	3578	+	ns		3805	+	ns				ns	
Mean	35/8 3651	+	ns ns		3734	+	ns		3757 3597	+	ns	
Std Dev	462	+	ns		898	+	ns		559	+	ns	
CV%	13				24	l		<u> </u>	43	l		<u> </u>

	Initial		Final		Initial		Final		Initial		Final	
Farm	EDS (titre)	Status	EDS (titre)	Status	IBV (titre)	Status	IBV (titre)	Status	NDV (titre)	Status	NDV (titre)	Status
4	3240	+	1640	+	900	-	1278	suspect	967	-	2449	+
4	3666	+	612	-	900	-	1139	suspect	790	-	2956	+
4	3868	+	3110	+	288	-	1453	suspect	3375	+	2631	+
4	3765	+	2304	+	283	-	1026	-	3133	+	1898	+
4	4127	+	3859	+	241	-	1069	-	5749	+	1430	suspect
4	3861	+	1045	suspect	669	-	3070	+	4872	+	4974	+
4	4453	+	1871	+	374	-	1282	suspect	281	ı	4630	+
4	3291	+	1019	suspect	221	-	2515	+	2913	+	3940	+
4	3778	+	2964	+	669	-	3037	+	2082	+	2223	+
4	2829	+	1788	+	955	-	1913	+	1634	suspect	1570	suspect
4	2774	+	3661	+	708	-	1833	+	1768	suspect	3316	+
4	3904	+	4483	+	2595	+	1010	-	1978	+	2010	+
4	4304	+	2739	+	2661	+	1082	-	4689	+	1430	suspect
4	4002	+	1498	+	2642	+	2246	+	1167	-	2780	+
4	4435	+	2401	+	2489	+	2105	+	315	-	1003	-
Mean	3753	+	2333	+	985	-	1737	+	2381	+	2616	+
Std Dev	521		1131		962		721		1703		1182	
CV%	14		48		87		41		72		45	
5	3477	+	2672	+	2989	+	2666	+	6059	+	2616	+
5	3738	+	2353	+	1298	suspect	2614	+	2515	+	2415	+
5	4401	+	2553	+	1003	-	2638	+	7255	+	3940	+
5	4277	+	2694	+	1228	suspect	2309	+	2768	suspect	3011	+
5	3607	+	2573	+	1065	-	2784	+	3734	+	1599	suspect
5	4174	+	2045	suspect	2873	+	3402	+	3182	+	4859	+
5	4191	+	2850	+	3068	+	2562	+	3158	+	3128	+
5	3542	+	5987	+	2736	+	3275	+	4227	+	6449	+
5	3828	+	2547	+	3553	+	2851	+	5971	+	2072	+
5	3761	+	3957	+	3214	+	2037	+	2715	+	2982	+
5	3355	+	2653	+	1126	suspect	3129	+	7298	+	2552	+
5	3814	+	2653	+	1019	-	3080	+	3757	+	3477	+
5	3399	+	2792	+	1141	suspect	3708	+	2987	+	1599	suspect
5	3938	+	2999	+	1019	-	2397	+	3255	+	2003	-
5	3941	+	2612	+	3137	+	2784	+	6147	+	3402	+
5	4157	+	2602	+	3033	+	2931	+	2335	suspect	3402	+
5	3432	+	6391	+	1214	suspect	3006	+	3495	+	3824	+
5	3451	+	6730	+	1185	suspect	3263	+	7405	+	7570	+
5	2992	+	6690	+	2330	+	2934	+	7789	+	3673	+
5	3848	+	6292	+	3248	+	3056	+	2313	+	3562	+
Mean	3766	+	3632	+	2074	+	2871	+	4418	+	3407	+
Std Dev	364		1693		997		396		1934		1492	
CV%	10		44		48		14		44		44	

Faecal Samples

Faecal samples were collected from 80 birds at the start of the survey period to determine worm burden and oocysts count, with 65 samples being collected at the end of the survey (1 farm had depopulated before samples could be taken). Again this represented a higher percentage of samples taken than the minimum recommended of 0.75% (0.82% - start and 0.86% - end) with results shown in table 10. All farms had nil or low oocyst counts at the start and end of the survey. Ascarids (roundworms) were present on all farms at the beginning of the survey and on the four farms sampled at the end of the survey. Of the four farms were sampling was undertake at the end of the trial, three showed an increase in roundworm burden. Capillaria (hairworms) were present in initial samples from farms 3 and 5 however they were present in very low numbers in a very small number of samples from each of the four farms sampled at the end of the survey.

Table 10: Worm (egg counts/g) and coccidia burden (oocysts/g) of faecal samples taken at the start and end of the survey period (ns - no sample).

-	Start	Finish	Start	Finish	Start	Finish
Farm	Coccidia (counts/g)	Coccidia (counts/g)	Ascarid (counts per g)	Ascarid (counts per g)	Capillaria (counts per g)	Capillaria (counts per g)
1	<25	0	450	<50	0	0
1	50	0	875	100	0	0
1	450	0	5725	2350	0	0
1	<25	0	975	1750	0	0
1	25	<25	750	7850	0	0
1	<25	0	800	3750	0	50
1	<25	0	550	250	0	0
1	<25	0	<25	800	0	0
1	<25	0	<25	4850	0	0
1	<25	<25	4975	550	0	0
1	<25	0	150	2450	0	0
1	<25	0	475	450	0	0
1	<25	0	1825	150	0	0
1	<25	0	1950	3450	0	0
1	125	0	2100	300	0	0
Mean			1443	1940	<u> </u>	
2	25	0	300	700	0	50
2	<25	0	675	350	0	0
2	<25	0	500	300	0	0
2	<25	0	2125	650	0	0
2	50	0	700	250	0	0
2	<25	0	75 75	1000	0	0
2	50	0	400	1100	0	0
2	<25	0	2025	950	0	0
2	775	0	450	700	0	0
2	<25	0	225	300	0	50
2	<25	0	175	1100	0	50
2	<25	0	200	600	0	0
2	<25	0	125	<50	0	0
2	<25	0	75	150	0	150
2	<25	0	<25	200	0	0
Mean	<u> </u>	0	538	560	0	<u> </u>
	-25				150	
3	<25	ns	100	ns	150	ns
3	<25	ns	75 125	ns	50 25	ns
3	0 -25	ns	125 575	ns	25 275	ns
3	<25	ns	575 -25	ns	275	ns
3	0	ns	<25	ns	25 -25	ns
3	0	ns	<25 850	ns	<25	ns
3	<25	ns	850 350	ns	75 125	ns
3	0	ns	350 75	ns	125	ns
3	0	ns	75 400	ns	75 225	ns
3	0	ns	400	ns	225	ns
3	0	ns	450	ns	125	ns
3	0	ns	1150	ns	100	ns
3	<25	ns	600	ns	375	ns
3	0	ns	50	ns	125	ns
3	0	ns	50	ns	100	ns

. <u>-</u>	Start	Finish	Start	Finish	Start	Finish
Farm	Coccidia (counts/g)	Coccidia (counts/g)	Ascarid (counts per g)	Ascarid (counts per g)	Capillaria (counts per g)	Capillaria (counts per g)
4	<25	0	3700	3150	0	0
4	<25	0	175	2400	100	50
4	0	0	9300	800	0	0
4	0	0	1025	2900	0	0
4	0	0	400	350	0	0
4	0	0	2725	400	0	0
4	0	0	1950	2550	0	50
4	0	0	<25	2200	0	0
4	0	0	425	600	0	0
4	0	0	200	900	0	50
4	0	0	725	400	0	50
4	0	0	275	2600	0	0
4	0	0	625	500	25	0
4	0	0	275	350	0	0
4	0	0	850	150	0	0
Mean			1512	1350		
5	<25	<25	225	< 50	125	0
5	0	0	200	250	125	0
5	<25	0	425	1400	150	0
5	0	<25	25	50	50	0
5	0	0	125	100	75	0
5	0	0	1550	< 50	50	0
5	0	0	<25	< 50	<25	0
5	0	0	25	1100	<25	0
5	0	0	225	500	<25	0
5	<25	0	225	< 50	25	0
5	<25	0	25	< 50	25	0
5	0	0	500	50	25	0
5	0	0	500	550	<25	0
5	<25	0	50	950	25	50
5	0	0	50	150	50	100
5	<25	0	75	2750	<25	50
5	<25	0	<25	750	<25	0
5	<25	0	<25	< 50	100	0
5	<25	0	175	< 50	150	0
5	<25	0	<25	200	<25	50
Mean			225	458		

Necropsy results

In the 2006 pilot trial, a total of 185 birds (out of approximately 9796 birds – 1.8%) were sent to the pathology lab for necropsy to determine cause of death (Tables 11 and 12). Reproduction tract lesions were the most common pathological finding (45.4% of birds necropsied, Figures 7 to 12) possibly causing death of birds due to other complications such as acute and chronic peritonitis, salpingoperitonitis and impacted oviduct. This was followed by cannibalism (38.9%, Figures 13 to 15) with these two causes responsible for 84.3% of all mortalities. Other pathology findings were heavy infestation with parasites (3.2%, Figures 16 to 18), injuries by predators (2.7%, figure 19), cachexia caused by stress and malnutrition (2.7%, Figure 20) which was also associated with atrophy of ovary and oviduct, and rupture of blood vessels associated with mechanical trauma (2.2%, Figure 21).

To better understand the cause of the ovary and oviduct pathologies a follow-up investigation in 2008 was carried out to take sterile samples from fresh sacrificed birds or morbid-looking birds (in total 61 birds were sacrificed from 3 farms during 3 months, see Appendix J) and conduct microbiological

tests. It was shown that 85% of birds did have similar reproduction tract problems. From samples collected 29% did not show growth of bacteria; gram positive cocci (*Staphylococcus spp.*) were found present in 53% of all isolates; 18% of samples were gram negative cocci or rods.

Table 11: Pathology findings for all farms as determined by necropsy

Most common causes of death	Number of birds
1. Cannibalism	72
2. Reproduction tract lesions	
• Clinical lesions on reproductive tract (oophoritis, yolk peritonitis, salpingitis, salpingoperitonitis)	84
3. Heavy infestation with parasites (Ascaridia galli) macroscopic evaluation	6
4. Bite by predators	5
5. Rupture of blood vessels (heart and liver)	4
6. Cachexia caused by stress and malnutrition	5
7. Unclear cases	9
TOTAL	185

Table 12: Pathology findings as a percentage per individual farm as determined by necropsy

Most common causes of death	Farm 1 (%)	Farm 2 (%)	Farm 3 (%)	Farm 4 (%)	Farm 5 (%)
1. Cannibalism	55.8	40	36	0	22.6
2. Reproduction tract lesions	34.9	35	44	73.9	61.3
 Clinical lesions on reproductive tract (oophoritis, yolk peritonitis, salpingitis, salpingoperitonitis) 	34.7	33	44	13.9	01.3
3. Heavy infestation with parasites (Ascaridia galli) macroscopic evaluation	2.3	0	12	4.3	0
4. Bite by predators	2.3	15	0	0	0
5. Rupture of blood vessels (heart and liver)	1.2	5	0	4.3	3.2
6. Cachexia caused by stress and malnutrition	1.2	5	4	4.3	3.2
7. Unclear cases	2.3	0	4	13.2	9.7

Figure 7: Chronic peritonitis: peritoneum covered with considerable fibrinous material and yolk



Figure 8: Follicles are degenerative and discoloured (oophoritis) caused by opportunistic infection.



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Figure 9: Follicles are misshapen, discoloured or haemorrhagic (oophoritis)



Figure 10: Case of salpingoperitonitis: oviduct blocked with caseous material.



Figure 11: Impacted oviduct associated with hypoplasia of ovary



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Figure 12: Enclosed yolks containing caseous material in a thickened capsule



Figure 13: Pecking around the vent associated with cannibalism.



Figure 14: Vent completely pecked out and oviduct and intestines eaten leading to death.



Figure 15: This bird has been vent pecked. After the oviduct and large intestine were eaten the rest of the small intestine filled with blood.



Figure 16: Intestine filled with parasite.



Figure 17: Intestine filled with Ascaridia galli (roundworms).



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Figure 18: Ascaridia galli from intestine.



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Figure 19: Head injuries associated with an attack by predators.



Figure 20: Ovary and oviduct hypoplasia associated with stress and cachexia.



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Figure 21: Haemorrhage in the abdomen caused by mechanical trauma.



Discussion

Rising EDS titres in a flock will indicate reinfection from wild birds which is important in relation to possible infection of flocks with avian influenza. Only one farm studied had a small increase in the average EDS titre with this farm and two others showing a decrease in the number of positive samples over the period of the survey. Currently all pullets destined for commercial egg production in Australia are vaccinated against IB with blood samples in this survey tested for circulating IB antibody titres as there has been some suggestion that vaccines are not as effective as previously thought, and the potential for a decline in immunity with time. Some farms did not show 100% positive samples for IBV titres at the start of the survey. The National Newcastle Disease Management Plan requires that all states and territories of Australia make Newcastle disease vaccination compulsory. In Queensland, at least 66% of the birds sampled in a group must reach the target titre of 2⁵ for birds over 18 weeks of age. Overall, all flocks had a positive average NDV, IB and EDS vaccine titre throughout the survey; however farms 4 and 5 did not have a uniform distribution of the titres and therefore inadequate protection for their flocks.

Vaccination failures through incorrect vaccination methods and/or degradation of vaccine efficacy are possible reasons for low titre results. Vaccines are dependent on conditions that will sustain their life or viability. Disinfectants, temperature and ultraviolet light can all alter or destroy the active component of vaccines. It is critical that vaccines are transported and stored under correct temperature conditions. If held in recommended conditions vaccines remain potent and effective until the expiry date. Stress may reduce a chicken's ability to mount an immune response and could include environmental extremes, inadequate nutrition, parasitism and other diseases. Therefore vaccinations should be delayed during periods of stress (Butcher and Miles, 2003). Poor distribution of live vaccine administered by water may result in chickens being 'missed' in parts of the shed. This means reliance on bird to bird transmission of vaccine however if using a killed vaccine no transmission will occur and birds will have no protection. In a poultry operation the objective should be disease prevention through effective biosecurity however if a breakdown occurs, the vaccination program needs to be adequate and effective to limit resulting losses (Butcher and Miles, 2003).

Reproduction tract lesions such as oophoritis, salpingitis, egg peritonitis, salpingoperitonitis, and impacted oviduct were the most frequently encountered necropsy finding diagnosed in 185 dead birds over a period of 3 months, presumably causing death of laying hens. Misshapen, discoloured cystic ova, frequently containing caseous material in a thickened capsule were found in the abdominal cavity showing extensive peritonitis with considerable fibrinous material or yolks. Enclosed yolks or caseous material were also found in the oviduct. No changes in egg quality were reported. There are innumerable factors that can initiate such pathologies however in most of cases they are associated with only a small number of mortalities caused by other complications such as acute and chronic peritonitis. Bacterial infection is a major contributory factor that should be taken into consideration if the frequency of these lesions increases in a flock. Various bacteria have been frequently reported to cause primary or secondary reproductive tract infections. It is also recognised that bacterial infections effect birds on the ground more than birds in cages. Although the route of infection is not clearly known, contamination of vent and cloaca/oviduct with faecal material has been seen as a source of such lesions.

To better understand the cause of the ovary and oviduct pathologies a follow-up investigation was carried out to take sterile samples from fresh sacrificed morbid-looking birds (in total 61 birds were sacrificed from 3 farms during 3 months, see Appendix J) and conduct microbiological tests. It was shown that 85% of birds did have similar reproductive tract problems as encountered in the pilot trial. From samples collected 33% did not show growth of bacteria; gram positive cocci (*Staphylococcus spp.*) were found present in 49% of all isolates; 18% of samples were gram negative cocci or rods. The frequency with which probable *S. aureus/S. hyicus* was isolated (15 from 30 isolates) suggests an aetiological relationship with encountered lesions. Additional factors such as immunosuppresion caused by stress and the implications from other bacterial and parasitic infections in free range flocks may have predisposed birds to opportunistic infections. Spotty liver and Coryza outbreaks were reported by producers in 3 free range flocks. In some cases parasite eggs, such as *A. galli* eggs, may

act as mechanical vectors of other bacterial infections. All flocks in the current survey were found to be infected with roundworms.

It has been known that a variety of bacteria such as coliforms, staphylococci, streptococci, pasteurella, and salmonella may infect birds of any age and cause local infections of the ovary in adult chickens. Oophoritis, salpingitis, and salpingoperitonitis are prominent feature of several viral and bacterial diseases and are associated with reduced egg production and/or poor eggshell quality (Riddell, 1996).

Mycoplasma gallisepticum, E. coli, Salmonella spp., P. multocida, Staphylococcus aureus, have frequently been isolated from lesions in the peritoneum and reproduction tract of laying chickens (Gross and Siegel, 1959; Sudhir *et al.*, 1968; Grimes, 1975; Jones and Owen, 1981; Riddell, 1996; Trampel *et al.*, 2007).

Apparently, systemic and local immunity (of the oviduct and ovary) play significant roles in the defence against primary infections and in the control of opportunistic bacteria normally living in the poultry environments. Locally secreted cytokines and chemokines might also affect the function of reproductive organs leading to ovary and oviduct dysfunctions.

It was concluded that, reproductive lesions often may cause drops in egg production and sudden and numerous death in laying hens. This has causes significant economic losses to producers and might affect the welfare of birds in free range.

Cannibalism was the second most common cause of morality. The reproduction tract lesions have been in most of cases associated with a dirty cloacae and vent, therefore leading to pecking and cannibalism. Pecking at the vent would also lead to the spread of the bacterial infection. Diseased hens attract cannibalistic pecks which could explain a reported correlation between infections of the reproductive tract and pecks to the cloacae (Randell *et al.*, 1977). Care must be taken to separate cause from effect, since cannibalistic attacks may also predispose surviving victims to subsequent infection.

General Discussion

The nationwide survey questionnaire return rate was disappointing with only fourteen producers returning completed surveys. The survey was developed in consultation with free range producers and an opportunity for the industry itself to have a major input into the direction of free-range research in Australia. Publicity included letters to industry associations and handouts at PIX and the survey was distributed through either state free range associations, producer groups or to individual producers. With surveys distributed through associations or producer groups we were reliant on them to inform their members of the survey and to ensure each member received a copy of the survey. Problems with this included the sending of surveys to all egg producers in a state regardless of production system. This makes it extremely hard to get an accurate measure of the number of free range producers in that state and to be able to follow up with these producers and remind them to complete the survey and that there is an extension. Some surveys were returned from producers who had not had free range layers for years however they were still on an association or producer group list. The other problem encountered was the topic that was being surveyed. Comments returned included 'difficult type of survey – like have you got aids', 'suspect you will only get the good ones as the other might not want to know/admit they have high chook mortalities' and more than once 'don't want the cage big boys to use this information against us'. The fact that not every egg production system was being surveyed caused concern that we were unfairly targeting the free range industry and therefore in turn helping the cage layer industry. Every assurance was given that the survey and project as a whole was to help the free range industry (and in fact, the industry asked for some research to be done) and confidentiality would be paramount however there seems to be some level of distrust.

Reasons for a low response to a survey can include confusing questions; a format that is not consistent and is unprofessional; telephone surveys (especially with the growth of telemarketing); no follow up and not letting people know the survey is to be conducted. The nationwide survey was widely

advertised prior to release, follow up letters and an extension were also provided. The survey itself was short with easy to understand questions in a consistent and professional format. As stated before every assurance was given that the results would be confidential and that we valued producer input and opinion. Apart from some telephone follow up, which is difficult especially when contact is made through producer groups and associations and not with individual producers; it is hard to say how we could have increased the return rate of the survey.

Implications

Bacterial infections effect birds on the ground more than birds in cages which impacts on their welfare and adds to the production costs in alternative systems. Significant economic losses to producers occur because egg production drops and mortality increases. Food safety could also be compromised.

Recommendations

- Provide advice to producers on how to prevent and combat bacterial infections. Bacterial infections are spread through live birds (droppings, feathers and other discharges), people (hands, clothing and footwear), contaminated equipment, eggs, air, feed and water, animals (dogs, cats, rats and mice) and insects (mosquitoes, flies and beetles). Ways to combat bacterial infections include biosecurity, medication, separation of birds that are infected, and birds of different ages, alleviation of stress (malnutrition, overcrowding and dirty conditions), vermin control and clean waterers and feeders.
- One problem is that there are virtually no medications for use against opportunistic infections causing reproduction tract problems. The only option is to improve hygienic conditions and increase the level of biosecurity in the sheds and on the farm in general. Following a vaccination program and preventing outbreaks of other bacterial diseases (such as fowl cholera and coryza) will help to decrease reproductive tract complications and improve egg production. Round worms are a potential vector for bacterial disease with piperazine the only anthelmintic available for hens in lay, and this should be addressed. As well there are limited options for external parasite control all of which increase disease potential.
- An investigation into microbial population of the oviduct in free range, barn and cage system
 could uncover potential for the identification of microflora that could be used as a future
 probiotic.
- With producers diagnosing 80 100% of 'cause of mortality' on-farm, the possibility of misdiagnosis is increased especially if symptoms are similar (e.g. fowl cholera and spotty liver). Increased producer education and improved use and availability of professionals may result in an early and correct diagnosis of 'cause of mortality' therefore decreasing the impact on the birds and production.

Acknowledgments

The authors would like to thank Karen Norris, Dr John Barnett, Mrs Ivy Inwood, Dr Pat Blackall, Dr Nigel Perkins and Avril Finn for their advice and help with the survey; the QDPI&F Biosecurity Sciences Laboratory; the participates who responded to the nationwide survey and particularly the five free range producers in South East Queensland who let us undertake the epidemiological survey on their properties.

Appendix A

Free-range layer mortality survey

This survey is being conducted on behalf of the Australian Poultry Co-operative Research Centre and has been designed to evaluate causes of mortality in the Australian free-range layer flock. It forms part of a larger project aimed at developing a national epidemiological survey with anticipated benefits of industry education and targeted research ideas to improve hen welfare. Your participation in this survey will be much appreciated and ensure results which will benefit the free-range sector and the industry as a whole. Please be assured that you will not be identified individually in the survey outcomes. If you would like to discuss this research survey further, please contact me: Tanya Nagle 07 3824 3081 or email Tanya.Nagle@dpi.qld.gov.au

Please return the completed survey in the reply paid envelope by

1.	GENERAL						
a)	Length of time poultry farm has been operational: years months						
b)	Length of time operation	ng as a free-ran	ge farm: as abo	ove 🗆 or			
c)	Average number of fre	e-range layers	each year:				
d)	Housed in:	num	ber of sheds with	n an outdoor area			
e)	Do wild birds regularly	visit your farm:	yes □ no □				
f)	If yes, what type and v	vhen:					
	Туре	Summer	Autumn	Winter	Spring		
D	ucks						
G	eese						
S	Swans						
Μ	Magpies						
С	Crows						
Μ	lynas						
Н	awks						
Е	agles						
Ib	lbis						
Ρ	Plovers						
K	Kookaburras						
S	Swallows						
D	Doves						
	Pigeons						
	Coastal wading birds						
S	Swamp wading birds						
0	ther						
a)	Are there any water courses/dams on the property: ves □ no □						

2.	HOUSING					
a)	Shed type:					
b)	Construction material:					
c)	Are sheds fixed in one location: yes \square no					
d)	If mobile, how often are the sheds moved: _					
e)	If permanent, do you use pasture rotation for	r each batch yes □ no □				
nat litte	If permanent, what type of floor does the she ural (dirt)	ed have:				
ind	Type of nest boxes: ividual □ ony □					
	gle deck □ Itiple deck □					
wo me pla						
i)	Type of perches:					
sin	rame (leaning)					
3.	HENS					
a)	What is the predominant breed/strain of bird	that you use:				
b)	Are hens in each shed a single age:	yes □ no □				
c)	Are hens reared on farm: yes □ no □					
d)	At what age are pullets placed in laying sheds:					
e)	At what age do you dispose of hens:					
f)	Are hens replaced on an all in all out basis:	yes □ no □				
g)	Do you use a lighting regime: yes □ no	☐ if yes, what is it				
h)	What is your average rate of production:					

4.	MANAGEMENT					
a)	Are birds beak trimmed: yes □ no □					
b)	If yes, at what age:					
c)	Do you undertake a regular rodent baiting programme: yes \square no \square					
d)	If yes, how often: weekly□ monthly□ tri-monthly□ yearly□ as required□					
e)	Do you undertake a regular worming programme: yes \square no \square					
f)	If yes, how often: weekly□ monthly□ tri-monthly□ yearly□ as required□					
g)	Do you undertake a regular external parasite eradication programme: yes \Box no \Box					
h)	If yes, how often:					
	weekly□ monthly□ tri-monthly□ yearly□ as required□					
ren dis hig lea	i) In between batches of hens do you: remove litter disinfect shed high pressure clean leave shed empty for period of time nothing other					
j)	What diseases are your flocks vaccinated against: (please tick)					
	IB (infectious bronchitis) ILT (infectious laryngotracheitis) Marek's disease Newcastle disease Fowl pox Coccidiosis Infectious coryza MG (Mycoplasma gallisepticum) AE (avian encephalomyelitis) EDS (inactivated egg drop syndrome) Fowl cholera MS (Mycoplasma synoviae)					

j) If known, what were the main causes of mortality (birds found dead and culls) on your farm, and at what age do they occur. Please rank in order of importance from 1 (most often the cause of mortality) to 10 (least often the cause of mortality):

Disease	Rank	Age (weeks)
Fowl cholera		
Colibacillosis		
Marek's disease		
Salmonella sp.		
Tracheitis (Mycoplasma sp)		
Coccidiosis		
Chronic respiratory disease		
Spotty liver		
Worms		
External parasites		
Egg peritonitis		
Ingluvitis (inflammation of the crop)		
Salpingitis (inflammation of the oviduct)		
Prolapse/protrusion		
Cannibalism		
Vent pecking		
Physical injury (ie. Broken leg)		
Predators		
Heat Stress		

k)	Other causes:		
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I) Who determines the cause of mortality:

	Percentage of time
Owner/Manager	
Veterinarian	
Pathology Lab	

Appendix B

Publicity Letter

The Australian Poultry Co-operative Research Centre (CRC) Free-range Layer Mortality Survey

The Australian free-range industry has an estimated grocery market share of 14.5%, with an estimated grocery value of \$22 million per annum. Profit margins are lower due to higher production costs and the potential for higher mortalities in this system are unacceptable to producers, industry and the general public. There is limited scientifically sound information on causes of mortality in free-range flocks in Australia and this survey will start to address this issue.

The Queensland Department of Primary Industries and Fisheries, has been contracted by the Australian Poultry Co-operative Research Centre to conduct a survey of all Australian commercial free-range producers during March /April 2006. The anticipated benefits include industry education and targeted research ideas to improve hen welfare.

This survey forms part of a larger project to evaluate causes of mortality in Australian commercial free-range flocks and will ensure, when developing a national epidemiological survey, temporal/locality issues are covered.

In order to achieve the best results, it is important that each farm manager/owner takes this opportunity to reply honestly to the survey questions so that the Free- range industry sector obtains maximum benefit from the results. Confidentiality is assured, and only collective data will be used in any reporting.

Should you wish to discuss any issues regarding the survey, please contact me on telephone 07 3824 3081 or email Tanya.Nagle@dpi.qld.gov.au

Yours sincerely

Tanya Nagle

Project Co-ordinator

J. Nagle

Scientist, Delivery - Animal Science

Appendix C

Letter requesting surveys required

Dear

As notified in previous correspondence the Queensland Department of Primary Industries and Fisheries, has been contracted by the Australian Poultry Co-operative Research Centre to conduct a survey of all Australian commercial free-range producers during March /April 2006.

This survey forms part of a larger project that will evaluate causes of mortality in Australian commercial free-range flocks and ensure, when developing a national epidemiological survey, temporal/locality issues are covered.

In order to achieve the best results, it is important that each farm manager/owner takes this opportunity to reply honestly to the survey questions so that the free-range industry sector obtains maximum benefit from the results. Confidentiality is assured, and only collective data will be used in any reporting.

Your assistance is sought in two ways. Firstly to advertise within your associations of our intention to conduct the survey and, secondly, to either:

- a) provide me with the addresses of farmers/farm managers within your association to forward surveys too, or
- b) provide me with the number of surveys you require to forward to members yourself and your address.

Should you wish to discuss any issues regarding the survey, please contact me on telephone 07 3824 3081 or email Tanya.Nagle@dpi.qld.gov.au

Yours sincerely Tanya

Tanya Nagle **Project Co-ordinator**Scientist, Delivery – Animal Science

Appendix D

Publicity flyer (P.I.X.)

Mortality in Commercial Free-range Layer Flocks Development of a framework for national survey

The Queensland Department of Primary Industries and Fisheries, has been contracted by the Australian Poultry Co-operative Research Centre (CRC) to conduct a survey investigating the major causes of mortality in Australian commercial free-range flocks. The survey will be conducted in two parts. The first is a nationwide survey of commercial free-range flocks and the second is a focused epidemiological study in Southeast Queensland. The anticipated benefits include industry education and targeted research ideas to improve hen welfare.

1. National Survey

The nationwide survey will be conducted in April/May 2006 and will ensure, when developing a national epidemiological survey, temporal/locality issues are covered. In order to achieve the best results, it is important that each farm manager/owner takes this opportunity to reply honestly to the survey questions so that the free-range industry sector obtains maximum benefit from the results. **Confidentiality is assured, and only collective data will be used in any reporting.**

2. Epidemiological Survey in Southeast Queensland

A precise epidemiological study on the causes of mortality in a selected number of Southeast Queensland commercial free-range flocks will be conducted.

Approximately five free-range flocks will be selected for this focused epidemiological study for approximately three to six months. Farms that agree to be involved in this part of the investigation will be required to complete a detailed questionnaire, allow collection of blood and faecal samples at the start and end of trial, keep accurate records and store all mortalities from the flock being studied. Farms will be provided with all equipment/supplies needed to undertake this work.

All mortalities will need to be stored on-farm in a freezer for regular collection by project team members for pathological assessment. The project can assist with freezer units for participating farms if required. **Again, confidentiality is assured, and only collective data will be used in any reporting.**

Should you wish to discuss any issues regarding the national survey or participate in the onfarm epidemiological survey in Southeast Queensland, please contact me on telephone 07 3824 3081 or email Tanya.Nagle@dpi.qld.gov.au

Tanya Nagle

Project Co-ordinator

Scientist, Delivery – Animal Science Poultry Research and Development Centre Queensland Department of Primary Industries and Fisheries





Appendix E

Survey covering letter

Dear Australian Free-range Industry Member,

Free-range Layer Mortality Survey

I am conducting this national industry survey on behalf of the Poultry Co-operative Research Centre to gather information on the causes of mortality in free-range flocks. The anticipated benefits include industry education and targeted research ideas to improve hen welfare.

This survey forms part of a larger project to evaluate causes of mortality in Australian commercial free-range flocks and will ensure, when developing a national epidemiological survey, temporal/locality issues are covered.

The information that you provide will be kept strictly confidential and only collated data will be used in the report.

Each survey has been numbered for the purpose of follow-up and to distinguish any differences between States when developing the national epidemiological survey.

In order to achieve the best results, it is important that each farm manager/owner takes this opportunity to reply honestly to the survey questions so that the free-range industry sector obtains maximum benefit from the results.

If you require any further information regarding this matter, please do not hesitate to contact me on telephone 07 3820 0504 or email Tanya.Nagle@dpi.qld.gov.au

Yours sincerely

Tanya Nagle

Project Co-ordinator

Scientist, Delivery - Animal Science

J. Nagle

Appendix F

Survey reminder letter

Dear Australian Free-range Industry Member,

Free-range Layer Mortality Survey

Industry response to the above survey to date has been relatively poor with an Australia wide overall return of 12 surveys (4 from each of Queensland, Victoria and South Australia).

In order to achieve the best result possible, I'm extending the deadline for returns until the 31st July 2006. If it is at all possible could you please complete and return your survey. The survey outcomes are important to the continued growth of the free-range industry.

If you require any further information regarding this matter, please do not hesitate to contact me on telephone 07 3820 0504 or email Tanya.Nagle@dpi.qld.gov.au

Yours sincerely

Tanya Nagle **Project Co-ordinator**Scientist, Delivery – Animal Science

Appendix G

Free-range Layer Mortality Survey

Your participation in this survey will be much appreciated and will ensure results which will benefit the free-range sector and the industry as a whole. Please be assured that you will not be identified individually in the survey outcomes. If you would like to discuss this research survey further, please contact me: Tanya Nagle 07 3820 0504 or email Tanya.Nagle@dpi.qld.gov.au.

1.	. GENERAL				
a)	Length of time poultry farm has been operational:years months				
b)) Length of time operating as a fre	e-range	e farm: as abo	ove 🗆 or	
c)) Average number of free-range la	yers ea	ich year:		
d)) Housed in:nu	mber of	sheds with an	outdoor area	
e)) Maximum number of hens per s	ned			
f)	Size of range area per shed				
g)) Do wild birds regularly visit your	farm?	yes □ no □]	
h)) If yes, what type and when (Plea	se tick)			
	Туре	mmer	Autumn	Winter	Spring
	Ducks				
	Geese				
	Swans				
	Magpies				
	Crows				
	Mynas				
	Hawks				
	Eagles				
	Ibis				
	Plovers				
	Kookaburras				
	Swallows				
	Doves				
	Pigeons				
	Coastal wading birds				
	Swamp wading birds				
	Other				
g) h)	Are there any water courses/darLayout of farm (rough sketch)	ns on th	e property:	yes □ no □	

2.	HOUSING OF SURVEY FLOCK			
a)	Shed type: conventional □ sawtooth □ igloo □ other			
b)	Construction material:			
c)	Is shed fixed in one location: yes \square no \square			
d)	If mobile, how often is the shed moved:			
e)	If permanent, do you use pasture rotation for each batch yes \square no \square			
f)	If permanent, what type of floor does the shed have: natural (dirt) □ litter □ other (please describe):			
g)	Type of nest boxes: individual □ colony □ single deck □ multiple deck □ automatic/belt □			
h)	Nest box construction material: wood □ metal □ plastic □ other (please describe):			
i)	Type of perches: A-frame (leaning) □ single-level □ other (please describe):			
3.	. HENS IN SURVEY FLOCK			
a)) What is the breed/strain and age of bird in the survey shed:			
b)	Were hens reared on farm: How are hens reared? Barn□ wire □ Floor □ If no, at what age do you buy in replacement pullets?			
c)	At what age were pullets placed in this shed:			
d)	At what age did pullets start ranging?			
e)	At what age do you dispose of hens?			
f)	Do you use a lighting regime? yes \square no \square if yes, what is it			
g)	What is your average rate of production in this shed?			
h)	What is your % of floor eggs?%			

4.	MANAGEMENT OF SURVEY FLOCK				
a)	What is the length of time hens are allowed to range per day?				
b)	Are hens allowed to range regardless of weather conditions? yes \square no \square				
c) Are birds beaks trimmed: yes □ no □					
	If yes, at what age:				
d)	Do you undertake a regular rodent baiting programme? yes □ no □ If yes, how often: weekly □ monthly □ tri-monthly □ yearly □ as required □				
e)	Do you undertake a regular worming programme? yes \square no \square If yes, how often:				
	weekly □ monthly □ tri-monthly □ yearly □ as required □				
f)	Do you undertake a regular external parasite eradication programme: yes \Box $$ no \Box If yes, how often:				
	weekly \square monthly \square tri-monthly \square yearly \square as required \square				
g)	What biosecurity measures are in place on your farm? footbaths□ showers□ stand down time□ dedicated footwear□				
	other				
h)	What diseases is this flock vaccinated against: (please tick)				
	IB (infectious bronchitis) ILT (infectious laryngotracheitis)				
	Marek's disease				
	Newcastle disease				
	Fowl pox				
	Coccidiosis				
	Infectious coryza				
	MG (Mycoplasma gallisepticum) AE (avian encephalomyelitis)				
	EDS (inactivated egg drop syndrome)				
Fowl cholera					
	MS (Mycoplasma synoviae)				
O11					
Utl	ner:				
i)	Do you use a nutritionist to develop diets for your flock? yes \square no \square				
j)	Do you use different diets as the hens age? yes \square no \square				
	If yes, what are they? starter □ grower □ developer □ prelayer □ layer (20-45wks) □ layer (46-65wks) □ layer (>65 wks) □				

Appendix H

Example of mortality record sheet

Farm: 5

Unknown cause of mortality			Known cause of mortality			
Label (farm no./mortality no.)	Date	No. of birds	Dead or cull (D or C)	Date	No. of birds	Cause
5/1						
5/2						
5/3						
5/4						
5/5						
5/6						
5/7						
5/8						
5/9						
5/10						
5/11						
5/12						
5/13						
5/14						
5/15						
5/16						
5/17						
5/18						
5/19						
5/20						

Appendix I

Necropsy record sheet

Owner's identification:	
Bird identification:	
Breed/Age:	
Weight:	
Date/Time of possible death:	
Cause of death: found dead/killed/injured etc	C
Relevant clinical signs prior to death:	
3 4 1 4 4 4 4	
Date of examination:	Preservation of body: fresh/frozen

1. External examination	Normal (Yes/No)	Describe abnormalities
Skin		
Feather/Comb condition		
External parasites		
Eyes/Ears/Nose		
Beak/Oral cavity		
Foot condition		
General carcass condition		

2. Examination of the organs/tissues/contents	Normal (Yes/No)	Describe abnormalities
Subcutaneous		
Musculoskeletal		
Peritoneal / Pleural cavities		
Oesophagus		
Crop		
Proventriculus		
Gizzard		
Intestine		
Cloaca		
Liver / Pancreas		
Spleen		
Thymus (if present)		
Bursa (if present)		
Air sacs/Lungs/Trachea		
Heart/circulatory		
Nervous system		
Genital/Ovary/Oviduct		
Kidney/ureter/adrenal gland		
Other		

3. Further records

	a.	Sampl	e collection:	Yes / No
		i.	Organ	
		ii.	Tissue	
		iii.	Content (fluid)
		iv.	Culture (swab	s)
	b.	Smea	r preparation:	
	C.	Picture	es:	
	d.	Other:		
4.	Ancill	ary dia	gnostics:	
	a.	Histop	athology	()
	b.	Toxico	logy	()
	C.	Cytolo	gy	()
	d.	Parasi	tology	()
	e.	Microb	oiology	()
	f.	Virolo	ЭУ	()
	g.	Other		()
5.	Dispo	sal arra	angements:	
6.	Tenta	tive dia	gnosis/comm	ents:
7.	Exami	iner:		
Sh	aniko S	Shini D	VSc, PhD	

Appendix J

Follow-up examination and post mortem results

Sample ID	Clinical examination & post- mortem results*	Data on flock (farm number same as in initial trial)	Sample origin	Growth and colony morphology
1	Wattle pale, liver normal, no egg in oviduct; ovary atrophied.	Farm 4 Strain: Hyline brown Age: 33 weeks	Oviduct	Columbia G+ cocci
2	Liver pale, ovary OK, egg in oviduct.		Spleen Oviduct	No growth
3	Fat in abdomen, liver pale, small haemorrhagic dots; intestine filled with worms (A. galli)		Liver	Columbia G- rods
4	Liver with numerous white foci, worms in intestine (A. galli)		Liver Oviduct	Columbia G+ cocci (MSA* & CPR**)
5	Eye swollen, liver with small haemorrhagic dots; no egg in oviduct; ovary inflamed.		Liver Ovary	Columbia, McConnkey G+ cocci (MSA & CPR)
6	Atrophy of ovary		Liver & Ovary	No growth
7	Liver pale; intestine filled with worms (A. galli and tape worms);		Liver & Ovary	No growth
8	Hen found dead. Ovary and oviduct hyperaemic; pale yellow fluid present in the abdominal cavity (peritonitis).	Farm 3 Shed 1 (not vaccinated for cholera) Strain: Hyline brown Age: 26 weeks Mortality: April 1.2% Production: 69%	Liver Ovary	Columbia*** McConnkey G+ cocci (MSA) G- short rods
9	Hen found dead. Liver enlarged, dark, spotted; Ovary and oviduct hyperaemic.		Liver & Ovary	No growth
10	Hen found dead. Big liver and spleen; ovary atrophy (inactive);		Liver	Columbia G- rods
11	Intestine, ovary, oviduct hyperaemic; liver normal		Ovary Intestine Peritoneum	No growth
12	Pecked cloacae; liver haemorrhagic dots, heart enlarged, walls/thin; ovary atrophied; yolk in peritoneum;	Farm 3 Shed 4 Strain: Hyline brown Age: 44 weeks Mortality: April 0.8% Production: 63%	Liver Heart Ovary	Columbia G+ cocci (MSA & CPR)
13	Pecked cloacae; no egg in oviduct; yolks solid in peritoneum; ovary inactive.		Ovary	Columbia McConnkey G- cocci (tiny)
14	Pecked cloacae; liver with haemorrhagic dots; ovary atrophied.		Ovary	Columbia G+ cocci

4.5	Farmal dead 1 has 120 forth 1	Farm 0	15	Onlymakis
15	Found dead. Liver white foci and big; ovary inflamed; pale yellow fluid present in abdomen	Farm 3 Shed 1 (not vaccinated for cholera) Strain: Hyline brown Age: 26 weeks Mortality: April 1.2% Production: 69%	Liver Spleen	Columbia G+ cocci (MSA & CPR)
16	Found dead. Liver normal; ovary and peritoneum haemorrhagic;		Ovary Peritoneum	No growth
17	Vent messy: liver Ok, no egg in oviduct; yolks deformed.		Ovary Oviduct	Columbia McConnkey G+ cocci (MSA & CPR)
18	Very cachectic, comb small; no egg in oviduct; atrophy of ovary;		Ovary Oviduct	No growth
19	Prolapse and dirty bum; normal liver reproduction tract		Ovary	No growth
20	Vent messy: diarrhoea; liver adhered to ribs; ovary/ oviduct atrophied; small intestine watery content, mucosa thickened.	Farm3 Shed 2 Strain: Hyline brown Age: 67 weeks Mortality: April 0.8% Production: 40%	Liver Ovary	Columbia G+ cocci (MSA & CPR)
21	Swollen eye (coryza); liver Ok; ovary atrophied; oviduct small		Liver Ovary	No growth
22	Yellowish and watery diarrhoea; vent pecked; liver Ok; ovary/ oviduct hyperaemic; intestine with worms (A. galli and capillaria in caecum);		Liver Peritoneum Ovary	No growth
23	Swollen eye (coryza); cachectic; liver Ok; atrophy of ovary	Farm 1 Shed 5 Strain: Hyline brown Age: 51 weeks	Liver Ovary Oviduct	Columbia McConnkey G+ cocci (MSA & CPR)
24	Comb very pale; liver Ok; oviduct hyperaemic; ovary deformed. A. galli in intestine;		Liver Ovary Oviduct	Columbia*** G+ rods G- tiny rods
25	Prolapse; cloacae messy; liver Ok; ovary and oviduct Ok; A. galli found in intestine		Ovary Oviduct	Columbia G+ cocci (MSA)
26	Swollen eye (coryza); Liver dark- brown; ovary hyperaemic, yolks deformed; mesentery thickened		Ovary Liver Spleen	Columbia McConnkey G+ cocci (MSA & CPR)
27	Liver dark; no egg in oviduct; ovary/yolks deformed;	Farm1 Shed 4 Strain: Bond brown Age: 48 weeks	Ovary Oviduct	Columbia McConnkey G+ cocci (MSA)
28	Prolapse; egg in oviduct; big/fat hen; liver pale; pancreas very large; yolks hyperaemic; Intestine filled with A. galli		Liver Ovary Oviduct	Columbia G+ rods
29	Old hen, over 70 wks; Ovary and oviduct hyperaemic; liver and spleen OK		Ovary	Columbia McConnkey G+ cocci (MSA)

30	Old hen/70 wks; peritoneum with inclusions/thickened; oviduct and ovary atrophied; A. galli in intestine.		Peritoneum Ovary	No growth
31	Wattle small and pale, liver normal, no egg in oviduct; ovary and oviduct atrophied	Farm 4 Shed 1 Strain: Hyline brown Age: 36 wks (treated with levamisol & nematape)	Ovary	Columbia G+ cocci (MSA)
32	Wattle small/pale; liver normal; ovary and oviduct atrophied		Ovary	Columbia G+ cocci (MSA)
33	Wattle small and pale; liver normal; ovary and oviduct atrophied		Ovary	No growth
34	Wattle small & pale; liver normal; ovary and oviduct atrophied		Ovary	No growth
35	Wattle small & pale; liver normal; Ovary and oviduct atrophied		Ovary	No growth
36	Big and healthy hen; Fatty liver; Ovary hyperemeic		Ovary	Columbia G+ cocci (MSA)
37	Prolapse and yellowish diarrhoea; liver normal, ovary & oviduct OK;	Farm 4 Shed 4 Strain: Hyline brown Age: 20 weeks (treated with piperazine)	Ovary; Liver	Columbia G+ cocci (MSA & CPR)
38	Prolapse and yellowish diarrhoea; Liver normal, ovary & oviduct OK;		Liver, Intestine Ovary	No growth
39	Prolapse and yellowish diarrhoea; deformed eggs		Intestine Ovary	No growth
40	Healthy hen Ovary OK		Ovary	Columbia G+ cocci (MSA)
41	Swollen eye (coryza); Yolks deformed, no egg in oviduct; heart slightly enlarged	Farm 3 Shed 1 (not vaccinated for cholera) Strain: Hyline brown Age: 30 weeks	Liver Ovary	McConnkey G- rods
42	External parasite (lice and mites); Liver pale; Ovary deformed.		Ovary	Columbia G+ cocci
43	Liver Ok; Ovary and oviduct redness; deformed yolks;		Ovary	No growth
44	Atrophied ovary; small oviduct; Organs OK		Liver Ovary	Columbia McConnkey G+ cocci (MSA)
45	Egg bound; all organs (intestine, liver, spleen small and pressed by eggs, content of abdominal cavity smelly	Farm 3 Shed 2 Strain: Hyline brown Age: 71 weeks	Ovary Oviduct Liver	Columbia McConnkey Cromogenic Salmonella G- tiny rods
46	Swollen eye and sinuses (coryza);	_	Liver	Columbia

	Repro organs atrophied; intestine small-empty		Ovary	McConnkey G+ cocci G- rods
47	Liver dark brown; ovary atrophied		Liver Ovary	Columbia G+ cocci (MSA & CPR)
48	Dirty vent; white spots on liver; ovary-yolks deformed; peritoneum thick-cloudy;	Farm 3 Shed 4 Strain: Hyline brown Age: 48 weeks	Liver Ovary	No growth
49	External parasite: lice; Liver dark- brown; Ovary/oviduct atrophied; intestine wall thin & hyperaemic		Ovary	Columbia Skirrow McConnkey G- rods (short)
50	Prolapse, dirty vent; old yolks in the abdominal cavity;		Ovary	Columbia G+ cocci
51	Liver normal; Ovary-oviduct atrophied;		Ovary	No growth
52	Wattle and comb pale; liver small; ovary & oviduct atrophied	Farm 1 Shed 5 Strain: Hyline brown Age: 55 weeks (treated with piperazine)	Liver Ovary	Columbia McConnkey G+ cocci (MSA & CPR)
53	Wattle and comb pale; liver Ok; ovary & oviduct atrophied		Ovary	Columbia G+ cocci
54	Swollen eye (coryza); Liver OK; ovary & oviduct atrophied		Liver Ovary	Columbia McConnkey G+ cocci (MSA & CPR)
55	Swollen ears and sinuses (coryza); liver Ok; ovary & oviduct atrophied		Liver Ovary	Columbia McConnkey G+ cocci (MSA & CPR)
56	Fat in abdominal cavity; liver with haemorrhagic dots, ovaries deformed; tapeworms in intestine		Ovary	No growth
57	Eye swollen (coryza); Big urine bladder; ovary atrophied	Farm 1 Shed 4 Strain: Bond brown Age: 52 wks	Ovary	Columbia G+ cocci (MSA & CPR)
58	Pale comb; big belly; in-capsulated eggs in oviduct; ovary still in function		Liver Ovary	McConnkey Cromogenic- Salmonella G- rods (short)
59	Eye and sinuses swollen; soft shell egg in oviduct (close to cloacae)		Ovary	Columbia McConnkey G+ cocci
60	Big hen-rooster-like; Ovary and oviduct present/atrophied		Ovary	Columbia McConnkey G+ cocci
61	Big-hen rooster-like; Ovary like a ball with haemorrhagic yolks		Ovary	Columbia McConnkey G+ cocci (MSA & CPR)

^{*}MSA: Manito Salt Agar, these isolates were grown in MSA plates;

From 61 (or 100%) of samples:

20 (33%) did not show any growth of bacteria; 11(18%) showed growth of other than Gram + cocci; 30 (49%) showed growth of Gram + cocci; from those 24 were grown on MSA plates and 15 were positive to Coagulase test.

Note that 2 of samples had mixed colonies.

^{**}CPR: Coagulase positive reaction; these isolates were positive to coagulase test;

^{***}In this case mixed colonies were grown

References

- Allen, J. and Perry, G.C. 1975. Feather pecking and cannibalism in a caged layer flock. *British Poultry Science* **16**: 441-451.
- Appleby, M.C. and Hughes, B.O. 1991. Welfare of laying hens in cages and alternative systems: environmental, physical and behavioural aspects. *World's Poultry Science Journal* **47**: 109-128.
- Bestman, M. 2005. Welfare in outdoor poultry and strategies to keep them healthy. In: *Proceedings of European Workshop for Scientists, Farmers, Environmental Specialists, Policymakers and Consumer Organisations Should Hens Be Kept Outside*. Nijmegen, The Netherlands.
- Bilčík, B. and Keeling, L.J. 1999. Changes in feather condition in relation to feather pecking and aggressive behaviour in laying hens. *British Poultry Science* **40**: 444-451.
- Blackall, P.J. 1999. Infectious coryza: overview of the disease and new diagnostic options. *Clinical Microbiology Reviews* **12(4)**: 627-632.
- Butcher, G.D. and Miles, R.D. 2003. Vaccine failure in poultry: factors to consider. *IFAS Extension* **VM82**: 1-4.
- Chadfield, M., Permin, A., Nansen, P. and Bisgaard, M. 2001. Investigation of the parasitic nematode *Ascaridia galli* (Shrank 1788) as a potential vector for *Salmonella enterica* dissemination in poultry. *Parasitological Research* **87**: 317-325.
- Critchley, K. 2002. *Spotty liver in barn and free range hens*. South Australian Animal Health Quarterly **5**: 6.
- Daniel, M. and Balnave, D. 1981. Responses of laying hens to gradual and abrupt increases in ambient temperature and humidity. *Australian Journal of Experimental Agriculture and Animal Husbandry* 21: 189-195.
- Glatz, P. and Ru, Y. 2004. Developing free-range animal production systems. *RIRDC Publication: Project Number* **SAR-30A**.
- Grimes, T.M. 1975. Causes of disease in two commercial flocks of laying hens. *Australian Veterinary Journal* **51(7)**:337-43.
- Gross, W.B. and Siegel, P.B. 1959. Coliform peritonitis of chickens. Avian Diseases 3(4): 370-373
- Gunnarsson, S., Keeling, L.J. and Svedberg, J. 1999. Effect of rearing factors on the prevalence of floor eggs, cloacal cannibalism and feather pecking in commercial flocks of loose housed laying hens. *British Poultry Science* **40**: 12-18.
- Huber-Eicher, B. and Audige, L. 1999. Analysis of risk factors for the occurrence of feather pecking in laying hen growers. *British Poultry Science* **40**: 599-604.
- Jones, H.G. and Owen, D.M.. 1981. Reproductive tract lesions of the laying fowl with particular reference to bacterial infection. *Veterinary Record* **108**:36-37.
- Kahn, C.M. (Ed). 2005. *The Merck Veterinary Manual*, 9th Edition. Merck&Co; Inc. United States of America.

- Keeling, L.J. 1994. Feather pecking who in the group does it, how often and under what circumstances? In: *Proceedings of the 9th European Poultry Conference*, 7-12th August, Glasgow, WSPA UK Branch, pp. 288-289.
- Kilgour, R. and Dalton, C. 1984. Livestock Behaviour: A Practical Guide. Methuen, New Zealand.
- Kjaer, J.B. and Sørensen, P. 2002. Feather pecking and cannibalism in free-range laying hens as affected by genotype, dietary level of methionine + cystine, light intensity during rearing and age at first access to the range area. *Applied Animal Behaviour Science* **76(1)**: 21-39.
- Kristensen, I. 1998. Organic egg, meat, and plant production bio-technical results from farms. In: Kristensen, T. (Ed). *Report of the Danish Institute of Agriculture Science* 1: 95-169.
- McAdie, T.M. and Keeling, L.J. 2000. Effect of manipulating feathers of laying hens on the incidence of feather pecking and cannibalism. *Applied Animal Behaviour Science* **68(3)**: 215-229.
- McMullin, P. 2004. *A Pocket guide to Poultry Health and Disease*. 5M Enterprises Ltd, United Kingdom.
- Mostert, B.E., Bowes, E.H. and van der Walt, J.C. 1995. Influence of different housing systems on the performance of hens of four laying strains. *South African Journal of Animal Science* **25**: 80-86.
- Nicol, C.J., Gregory, N.G., Knowles, T.G., Parkman, I.D. and Wilkins, L.J. 1999. Differential effects of increased stocking density, mediated by increased flock size, on feather pecking and aggression in laying hens. *Applied Animal Behaviour Science* **65(2)**: 137-152.
- Parkinson, G. and Cransberg, P. 2002. Cloacal haemorrhage, vent trauma and beak trimming in laying hens. *RIRDC Publication* **02/012**.
- Permin, A. and Hansen, J.W. 1998. *The Epidemiology, Diagnosis and Control of Poultry Parasites An FAO Handbook.* Food and Agriculture Organisation of the United Nations, Rome.
- Pötzsch, C.J., Lewis, K., Nicol, C.J. and Green, L.E. 2001. A cross-sectional study of the prevalence of vent pecking in laying hens in alternative systems and its associations with feather pecking, management and disease. *Applied Animal Behaviour Science* **74(4)**: 259-272.
- Prescott, N.B., Wathes, C.M., Jarvis, J.R. 2003. Light, vision and the welfare of poultry. *Animal Welfare* **12**: 269-288.
- Randall, C.J., Blandford, T.B., Borland, E.D., Brooksbank, N.H., Hall, S.A., Hebert, C.N. and Richards, S.R. 1977. A survey of mortality in 51 caged laying flocks. *Avian Pathology* **6**: 149-170.
- Riddell, C. 1996. In: *Avian Histopathology*, 2nd edition, The American Association of Avian Pathologists, p. 214-215.
- Sahu, S.P. and Munro, D.A. 1969. Observations of systemic and localized infection associated with the isolation of staphylococcus in chickens in North Carolina. *Avian Disease* **13(3)**:684-9.
- Salant, P. and Dillman, D.A. 1994. *How to Conduct Your Own Survey*. John Wiley and Sons Inc., Canada.
- Saif, M. (Ed.). 2003. Diseases of Poultry. M. Iowa State University Press, United States of America.
- Sloss, M.W., Kemp, R.L. and Zajac, A.M. 1994. *Veterinary Clinical Parasitology*, 6th Edition. Blackwell Publishing Professional, USA.

- Sommer, F. and Vasicek, L. 2000. Management and state of health in free-range poultry flocks. *Wiener Tierärztliche Monasschrift* **87(7)**: 202-212.
- Swayne, D.E., Glisson, J.R., Jackwood. M.W., Pearson, J.E. and Reed, W.M. (Ed). 1998. *Isolation and Identification of Avian Pathogens*, 4th Edition. American Association of Avian Pathologists, United States of America.
- Trampel, D.W., Wannemuehler, Y. and Nolan, L.K. 2007. Characterization of Escherichia coli isolates from peritonitis lesions in commercial laying hens. *Avian Disease* **51(4)**:840-4.

Plain English Compendium Summary Pilot trial - Mortality in free range flocks **Project Title:** Poultry CRC Project No.: 05.13 Researcher: Tanya Nagle Organisation: Department of Primary Industries & Fisheries Poultry Research & Development Centre ALEXANDRA HILLS QLD 4161 Phone: 07 3820 0504 07 3824 4316 Fax: Email: Tanya.Nagle@dpi.qld.gov.au **Objectives** Undertake a pilot study to develop a framework for a national survey on causes of mortality in commercial free-range flocks. **Background** Free range is the poultry housing system most actively supported by welfare groups. Currently there are 1.69 million free range hens in Australia (11% of total laying hens in Australia) with an estimated grocery market share of 14.5%. Free range eggs are worth 23% of the value of the Australian egg industry, which is more than \$71 million a year. Mortality in free range flocks can be caused by numerous factors including feather pecking and/or cannibalism, disease, predators and management (diet, housing, strain, rearing, light levels, vaccinations, flock size and density). There is limited scientifically sound information on causes of mortality in free range flocks in Australia and we will start to address this. Research A survey of all free-range producers in Australia was undertaken to indicate causes of mortality in the industry to tailor future epidemiological surveys to ensure temporal/locality issues were covered. A small focused epidemiological study on the causes of mortality in five commercial free range flocks in Southeast Queensland including a detailed survey of participating farms, ongoing mortality records, serology, faecal samples and gross pathology was completed. **Outcomes** Fourteen free range producers replied to the nationwide survey (OLD - 5, NSW - 1, VIC - 4, SA - 4) with fowl cholera named as the most important cause of mortality in Queensland and spotty liver the most important cause of mortality in New South Wales and Victoria. Queensland and Victoria had problems with predators with Victoria also recording heat stress as an important cause of mortality. Egg peritonitis, prolapse/protusion, cannibalism and vent pecking were all seen as important causes of mortality in all states. Of the five flocks that were studied in the intensive epidemiological survey in Southeast Queensland one farm had a small increase in the average egg drop syndrome vaccine titre with this farm and two others showing a decrease in the number of positive samples over the period of the survey. One farm initially showed a negative average infectious bronchitis vaccine titre however this had increased to become positive in the final sample. Overall, all flocks had a positive average Newcastle vaccine titre throughout the survey; however two farms had only 54% and 64% of samples returning positive titres. Fowl typhoid was the most common cause of mortality with cannibalism the second most common cause. **Implications** Bacterial infections effect birds on the ground more than birds in cages which impacts on their welfare and adds to the production costs in alternative systems. Significant economic losses to producers occur because egg production drops and mortality increases. Food safety could also be compromised. Recommendations include typing of salmonella present in current trial allowing specific prevention and control information to be provided to producers, approval of more antibiotics and anthelmintics for use

in laying hens, an investigation into the microbial population of the oviduct in free range, barn and cage laying hens with the possibility of developing probiotics, and increased producer education and improved use and

availability of professionals to ensure early and correct diagnosis of causes of

mortality.