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Experimental infection of broiler breeder hens with the intestinal spirochaete *Brachyspira* (*Serpulina*) *pilosicoli* causes reduced egg production

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The pathogenic potential of the anaerobic intestinal spirochaetes *Brachyspira* (*Serpulina*) *pilosicoli* and *Brachyspira innocens* was evaluated in adult chickens. Thirty 17-week-old Cobb broiler breeder hens were individually caged in three groups of 10 birds. Control birds (group A) were sham inoculated with sterile broth medium. Birds in the other two groups (groups B and C) were inoculated, respectively, with an isolate of *B. innocens* or of *B. pilosicoli*. Birds were monitored daily, and killed at 41 weeks of age. Infection had no consistent effect on body weight gain, but inoculation with *B. pilosicoli* resulted in a transient increase in faecal water content. *B. innocens* infection had no effect on egg production, but *B. pilosicoli* infection caused a delayed onset of laying, and a highly significant reduction in egg production over the first 11 weeks of lay. This study confirms that *B. pilosicoli* can cause serious egg production losses in adult chickens, while *B. innocens* is not obviously pathogenic.

Introduction

Spirochaetes belonging to the genus *Brachyspira* (formerly *Serpulina*) are anaerobic, spiral-shaped bacteria that colonize the large intestine and can cause enteric disease in a number of animal species (Hampson & Stanton, 1997). These intestinal spirochaetes can only be isolated after a minimum of 3 to 5 days incubation using specialized selective media and anaerobic growth conditions.

To date, colonization of layer and broiler breeder birds with intestinal spirochaetes has been recorded in continental Europe, the UK, the US and Australia (Davelaar *et al.*, 1986; Griffiths *et al.*, 1987; Dwars *et al.*, 1989; Swayne *et al.*, 1992, 1995; Trampel *et al.*, 1994; McLaren *et al.*, 1996; Stephens & Hampson, 1999). Colonization tends to be chronic in adult birds in infected flocks (Dwars *et al.*, 1990), and has been associated with a variety of symptoms including diarrhoea, increased faecal fat content, faecal staining of eggshells, delayed onset of egg laying, reduced egg weights, reduced growth

rates, increased feed consumption and poor digestion of feed (Davelaar *et al.*, 1986; Griffiths *et al.*, 1987; Dwars *et al.*, 1990; Swayne *et al.*, 1992; Dwars *et al.*, 1992a, 1993; Trampel *et al.*, 1994). Broiler flocks derived from the offspring of breeder flocks infected with spirochaetes have been shown to have poorer feed conversion, an increased number of weak chicks, slower growth and a poorer feed digestion than the offspring of flocks where spirochaetes are not present (Smit *et al.*, 1998). Experimentally infected broiler chicks show retarded growth (Dwars *et al.*, 1992b) but, despite several investigations, natural colonization of broilers with spirochaetes has not been detected in the field (Stephens & Hampson, 2001).

Several different species of intestinal spirochaetes naturally colonize chickens, and not all are necessarily capable of causing disease (McLaren *et al.*, 1997). The pathogenic species that workers in the UK and Europe appear to have been most concerned with is *Brachyspira intermedia* (Griffiths *et al.*, 1987; Dwars *et al.*, 1992a,b, 1993), although

strains of *Brachyspira pilosicoli* have also been identified (McLaren *et al.*, 1997). Reports from the US have involved either *B. pilosicoli* (Trampel *et al.*, 1994) or *Brachyspira alvinipulli* (Swayne *et al.*, 1992, 1995; Stanton *et al.*, 1998). In Australia, both *B. intermedia* and *B. pilosicoli* have been isolated from layer and broiler breeder flocks with production problems (Stephens & Hampson, 1999). Other intestinal spirochaete species, including *Brachyspira innocens*, have also been isolated from Australian chickens (McLaren *et al.*, 1997). *B. innocens* is generally considered to be a non-pathogenic spirochaete in conventional pigs (Kinyon & Harris, 1979), although porcine strains of this species have caused mucoid faeces and typhlocolitis in gnotobiotic pigs (Neef *et al.*, 1994).

In the Netherlands, experimental infection of adult birds with spirochaete strain 1380, later identified as *B. intermedia* (McLaren *et al.*, 1997), resulted in increased faecal water content, reduced egg production and poor performance in broilers hatched from the infected birds (Dwars *et al.*, 1989, 1992a, 1993). In Australia, experimental infection of layer hens with an Australian chicken isolate of *B. intermedia* resulted in prolonged caecal colonization, increased faecal moisture content and reduced egg production (Hampson & McLaren, 1999). In contrast to the situation with *B. intermedia*, there have been no reports of studies using either *B. pilosicoli* or *B. innocens* to experimentally infect adult birds. *B. pilosicoli* is of particular comparative interest because it colonizes many bird and animal species, including humans (Trott *et al.*, 1997a,b; Oxberry *et al.*, 1998; Trivett-Moore *et al.*, 1998; Brooke *et al.*, 2001). The existence of cross-species transmission of *B. pilosicoli* has important implications for control of the infection in chicken flocks, while the possibility of zoonotic transfer increases the need for further study of this spirochaete.

The overall aim of the current study was to investigate the pathogenic potential of Australian chicken strains of *B. pilosicoli* and *B. innocens* in broiler breeder hens. Strains of both species were obtained from a farm where ongoing egg production problems and wet litter had been observed in association with spirochaete colonization. These strains were then tested under experimental conditions on birds from the same farm, receiving the same diets, to determine whether the bacteria were capable of causing problems similar to those observed on the farm of origin.

Materials and Methods

Experimental birds

Thirty Cobb 500 broiler breeder females were obtained from a commercial producer at 13 weeks of age. The birds were placed in individual cages with mesh floors and egg roll-out trays. The cages were specially constructed to be large enough to accommodate breeder females. Each cage was provided with a waste tray for the collection of faeces. Clear plastic sheets were hung between cages to minimize the risk of transmission of infection between cages. The birds were kept in a controlled environment room with temperatures varying between 17 and 23°C. The day-length was set at 8 h until 19 weeks of age, then gradually increased to 15 h until 23 weeks of age and, thereafter, maintained at 16 h. The birds were fed commercial diets supplied by the farm of origin, and these contained 50 parts/10^6 zinc bacitracin. They received a pullet developer diet until 19 weeks of age, then a pre-breeder ration. When egg production in the control group reached approximately 15%, all the birds were given a breeder production mix. Feed intake was restricted, with the birds being given 62 g daily at 13 weeks of age, and this being gradually increased to a maximum of 165 g per day by 27 weeks of age. Water was provided *ad libitum* by means of individual water bottles with nipple drinkers.

Spirochaete strains used for experimental infection

B. pilosicoli strain CPSp1 and *B. innocens* strain CPSi1, which were used to infect the birds, were isolated from breeders on the same farm from which the test birds originated. The strains had been isolated 2 years before the current study, during the course of a disease investigation, and their species identity had been confirmed through biochemical testing and polymerase chain reaction (PCR) amplification of portions of their 16S rRNA and NADH oxidase genes (Stephens & Hampson, 1999). For experimental infection, the spirochaetes were grown to mid-log phase in Kunkle's anaerobic broth (Kunkle *et al.*, 1986). The inocula used to infect the birds contained approximately 10^8 bacterial cells/ml.

Experimental infection and monitoring of birds

The birds were acclimatized for 4 weeks. Over this period, individual faecal samples from each bird were taken weekly. These were cultured for spirochaetes on selective Trypticase Soy Agar (Micro Diagnostics, Australia), supplemented with 5% defibrinated bovine blood, 0.1% porcine mucin (Sigma, USA), 200 μg/ml spectinomycin and 6.25% μg/ml each of colistin and vancomycin (Sigma). Plates were incubated in an anaerobic environment generated by Anaerogen sachets (Oxoid, UK), and growth examined by dark-field microscopy after 5 and 10 days. The species identity of isolates obtained during the course of the study was determined using a species-specific PCR protocol, as previously described (Stephens & Hampson, 1999).

After 4 weeks, the birds were weighed and randomly assigned on the basis of body weight to one of three groups, each of 10 birds. Birds in group A (control group) were inoculated orally with 1 ml sterile broth. Birds in groups B and C were inoculated with 1 ml broth culture of either *B. innocens* or *B. pilosicoli*, respectively.

Eggs from each bird were collected, counted and weighed every day. Once a week, the birds were weighed, and individual faecal samples collected and cultured for intestinal spirochaetes. The faecal samples were weighed, dried to constant weight in a hot-air oven, and the percentage faecal moisture calculated.

Postmortem examination

At 41 weeks of age, the birds were killed by cervical dislocation, and subjected to routine postmortem examination. The caeca, ovaries and oviducts were examined for gross pathological lesions, and sections were fixed in 10% buffered formalin for subsequent histological examination. These were dehydrated through alcohol, embedded in paraffin and cut at 4 μm. Sections were stained with haematoxylin and eosin. Caecal contents were cultured for intestinal spirochaetes in the same way as for faecal samples.

In vitro susceptibility to zinc bacitracin

To determine the *in vitro* susceptibility of the two spirochaete strains to zinc bacitracin, each strain was tested in a broth dilution assay. For each strain, 20 test tubes each containing 10 ml volumes of Kunkle's anaerobic broth were prepared. For each series of 20 tubes, two contained no additives while the other pairs of tubes contained doubling concentrations of zinc bacitracin (Jurox Pty Ltd, Rutherford, Australia) from 1 to 256 μg/ml. For each series, each tube was

Table 1. *Group mean (+ standard error) body weight of chickens (g) in the three experimental groups at the start of the experiment, and on weeks where subsequent significant group effects were found*

Week	Group A (control)	Group B (*B. innocens*)	Group C (*B. pilosicoli*)	*P* value
18	1645 (48)	1695 (44)	1635 (61)	0.7627
26	3035 (76)^{A}	3030 (77)^{A}	3255 (46)^{B}	0.0518
28	3390 (91)^{AB}	3330 (81)^{A}	3611 (57)^{B}	0.0499
29	3330 (71)^{AB}	3170 (93)^{A}	3422 (54)^{B}	0.0771
32	3655 (81)^{AB}	3570 (77)^{A}	3822 (60)^{B}	0.0733

Means within a row having different superscripts differ at the 5% level of significance.

inoculated with 0.5 ml active culture of the appropriate spirochaete at 10^{7} cells/ml. The tubes were incubated at 37°C on a reciprocal shaker for 5 days. Turbidity was examined visually each day and, at the end of the incubation period, aliquots were removed from the tubes and examined under a phase contrast microscope for the presence of active viable spirochaetes.

Analysis

Weekly group bird weights and faecal moisture content were compared using one-way analysis of variance. Means were compared using Fisher's protected least-significant difference method, and significance was accepted at the 5% level. Group egg production per week was compared using Chi square tests, except where values per cell were less than five, when Fisher's exact test was used in 2×2 contingency tables. Full egg production was assumed to be one egg per bird per day. The weights of the eggs produced were analysed by calculating a mean weight of eggs produced from each bird per week (i.e. total weight/ number of eggs), then calculating group means of these bird means each week. These group means were compared by one-way analysis of variance.

Results

Body weights

There were few significant group differences in the body weights of the birds, and those that were present are shown in Table 1. On week 26, the birds in group C were significantly heavier than those in the other two groups. Group C birds were also significantly heavier than those in group B, but not in group A, on weeks 28, 29 and 32. Generally, the birds gained weight as expected for the type and age of bird, and remained comparable with birds of the same batch in commercial production.

Colonization

None of the birds were colonized with intestinal spirochaetes prior to the start of the experiment, and no control birds were colonized at any time. At week 18, 1 week following inoculation, *B. innocens* was isolated from one bird in group B. At week 19, an additional two birds in this group were positive for *B. innocens*, and all three remained positive for a further week. At week 21, 4 weeks after inoculation, only one of these three birds was positive. Thereafter, and for the duration of the trial, none of the birds in group B were culture positive.

At week 18, 1 week following inoculation, *B. pilosicoli* was isolated from faecal samples from three birds in group C. These birds were positive the following week, but only one was positive the next week. All birds were culture negative by week 21, and remained so throughout the rest of the experiment.

Faecal water content

In weeks 19, 20 and 32, the faecal moisture content of birds in group C was significantly higher than that of birds in group B, but not that of birds in group A (Table 2). At these times, the faeces of

Table 2. *Group mean (+ standard error) percent faecal water content of chickens in the three experimental groups at the start of the experiment, and on weeks where subsequent significant group effects were found*

Week	Group A (control)	Group B (*B. innocens*)	Group C (*B. pilosicoli*)	*P* value
18	55.33 (1.78)	53.29 (2.33)	58.52 (1.15)	0.1421
19	55.94 (1.11)^{A}	50.41 (2.36)^{B}	57.56 (1.28)^{A}	0.0143
20	54.0 (1.55)^{A}	48.7 (1.29)^{B}	55.4 (0.96)^{A}	0.0024
32	58.1 (0.64)^{AB}	57.4 (1.14)^{A}	60.5 (0.73)^{B}	0.0564

Means within a row having different superscripts differ at the 5% level of significance.

Table 3. *Total number of eggs produced per group of 10 chickens per week (maximum possible, 70/week)*

Age (weeks)	Group A (control)	Group B (*B. innocens*)	Group C (*B. pilosicoli*)	*P* value
23	1[A]	5[A]	0[B]	0.0272
24	4[A]	14[B]	0[C]	0.0001
25	15[A]	22[A]	5[B]	0.0015
26	22[A]	40[B]	5[C]	0.0000
27	31[A]	52[B]	17[C]	0.0000
28	44[A]	50[A]	26[B]	0.0014
29	49[A]	49[A]	33[B]	0.0525
30	64[A]	57[A]	37[B]	0.0000
31	55	46	44	0.2290
32	56[A]	57[A]	33[B]	0.0002
33	60[A]	58[A]	41[B]	0.0082
34	54	50	41	0.3066
35	51	48	40	0.5098
36	48	49	35	0.6473
37	52	45	34	0.1948
38	43	50	32	0.2222
39	48	48	41	0.8147
40	46	48	34	0.6519
41	44	35	34	0.2631
Total	787[A]	823[A]	532[B]	0.0000

Means within rows having different superscripts differ at the 5% level of significance

birds in group C were 2 to 7% wetter than those in the other two groups. There were no other significant differences in the faecal moisture content between the groups, and there was no overall group difference in faecal moisture content.

Egg production

Birds in groups A and B both commenced laying at 23 weeks of age; however, the onset of egg production in birds in group C was delayed for 2 weeks (Table 3). Ninety percent of birds in groups A and B had commenced laying by 27 and 26 weeks of age, respectively, but group C did not reach this figure until the birds were 30 weeks of age.

Egg production by birds in group C was significantly less than birds in the other two groups from weeks 23 to 33, except in week 31. Thereafter, no significant differences occurred. Overall, birds in group C produced highly significantly ($P < 0.0001$) fewer eggs than birds in the other two groups.

Egg weights

Comparisons of the mean weights of eggs layed revealed no significant group effects, either on a weekly basis or overall. Mean egg weights increased gradually from around 45 g at start of lay to approximately 67 g at week 41.

Bird health

Birds in group C had frothy brown faeces when sampled in the first 2 weeks following experimental inoculation, but not thereafter. The faeces of the birds in the other groups remained normal throughout.

Two birds in group C were euthanazed 1 week after they were noticed to be depressed and off their feed. The first, killed in week 24, was diagnosed with tibial dyschondroplasia, and the second, killed in week 34, with hepatoma. No other abnormalities were found in these birds, and no spirochaetes were isolated from their caecae.

Postmortem findings

At necropsy, the caeca of the birds from group C were observed to contain more gas, and their contents were more frothy, fluid and considerably paler than those of the birds in the other two groups. No gross or histological lesions were found in the caecae, ovaries or oviducts of any of the birds. There was no evidence of end-on attachment of spirochaetes to the caecal epithelium, and no spirochaetes were isolated from any of the birds.

Susceptibility to zinc bacitracin

No inhibition of growth was observed with either spirochaete strain at any of the dilutions of zinc bacitracin tested.

Discussion

In Australia, natural infection with both *B. pilosicoli* and *B. innocens* has been reported in layer and broiler breeder flocks (Stephens & Hampson, 1999). The current study is the first report of experimental infection of commercial adult birds with these organisms.

Overall, the study suggested that *B. innocens* is unlikely to have much pathogenic significance in adult birds. Experimental challenge had no influence on any of the production or health parameters measured. The presence of zinc bacitracin in the diet is unlikely to have influenced these results, as it was present in relatively low concentrations in the diet, acts only on the cell wall of Gram-positive organisms, and had no effect on growth of the spirochaetes *in vitro*. Furthermore, the zinc bacitracin was included in the diet to mimic the situation in the commercial breeder farm from which the spirochaetes originated, where ongoing production problems had been identified. Similarly, to mimic the situation in the field, the birds and diets were obtained from the same farm. This selection was made specifically to help determine the potential pathogenic significance of the spirochaete species in birds of the same genotype and immunological status as present on the farm, after they were experimentally infected with these bacteria under controlled conditions.

The presence of spirochaetes such as *B. innocens*, which appear to have little pathogenic significance, complicates the diagnosis of avian intestinal spirochaetosis. This emphasizes the need for reliable species-specific techniques, such as PCR, to differentiate pathogenic and non-pathogenic spirochaete isolates from chickens.

Following experimental inoculation of the birds with spirochaetes, only a small number became culture positive, and this colonization persisted for a maximum of 4 weeks. The birds were given only a single challenge with a relatively small number of spirochaetes, and this may help account for the low proportion of birds that became culture positive. Interestingly, there was no evidence of cross-transmission of spirochaetes between or within groups of birds, but again this may have failed to arise due to the careful hygiene that was practised and the low level of colonization overall. Similarly, the good husbandry conditions practiced in the experiment may have minimized potential production problems that would occur in a more stressful commercial situation.

Faeces were only cultured weekly, and it is possible that transient colonization of individual birds was missed. The culture detection method used may also not have been sufficiently sensitive to detect a low level of colonization in some birds. Atyeo *et al.* (1998) have shown that the use of PCR on growth from primary plates can substantially increase detection rates for *B. pilosicoli* in pig faeces, achieving detection limits of around 10^4 cells/g faeces. More recently, *B. pilosicoli* has been detected in human faeces using PCR on DNA extracted directly from the faeces (Mikosza *et al.*, 2001). Future studies of *B. pilosicoli* in chickens should include use of such PCR protocols to improve the sensitivity of detection.

The experimental challenge with *B. pilosicoli* did not cause a reduction in body weight and, on several occasions in the earlier part of the experiment, birds in group C were actually heavier than the other birds. This probably reflects the fact that the birds in group C were producing fewer eggs, and putting more energy into growth at this time.

The birds infected with *B. pilosicoli* showed a transient increase in faecal moisture in the first few weeks after infection, as well as having brown frothy faeces, but this effect did not persist. Had colonization with the spirochaetes continued, faecal moisture content might have remained elevated. Although the increase in moisture content was relatively small, commercially this could be sufficient to cause problems with mechanical cleaning of manure, faecal staining of eggs, increased odour and attraction of flies.

The most striking and significant finding in the study was the delay in both onset of egg production and in reduced total egg production in the birds inoculated with *B. pilosicoli*. When these birds did produce eggs, however, these were not significantly lighter than those produced by birds in the other two groups. The major losses in production occurred in the first 11 weeks of lay, where in 10 of these weeks total egg production was significantly reduced in birds of group C. Over this 11-week period, average egg productions per bird in groups A, B and C were 40.7, 45.0 and 26.8, respectively. Hence, over this initial period, birds in group C produced, on average, 14 less eggs than the birds in the control group, or had only two-thirds of their level of production. In a commercial situation, this loss would have an extremely serious economic impact. That such losses can occur in practice was seen in Iowa, where a layer flock (100 000 birds) infected with a spirochaete, which was later identified as *B. pilosicoli* (McLaren *et al.*, 1997), was shown to have an overall 5% reduction in egg production (Trampel *et al.*, 1994).

No gross or histological abnormalities were found in the caecae of any of the birds at postmortem examination. The lack of pathological changes at necropsy is not altogether surprising because the birds were not colonized at this time and they had had many weeks for any lesions to resolve. In future studies, it would be useful to kill birds at the time they were culture positive. A practical outcome of the lack of gross and histological changes in the caecae is that it may not necessarily always be possible to diagnose infection with intestinal spirochaetes purely on pathological grounds. Diagnosis may have to be

based on microbiological culture of appropriate samples from birds showing clinical signs. Previous studies have demonstrated that, in flocks infected with spirochaetes and experiencing production problems, not all birds are necessarily positive on culture (Stephens & Hampson, 1999). Thus, samples from a number of birds might have to have to be examined to enable a diagnosis to be made. Moreover, the anaerobic nature and fastidious growth requirements of intestinal spirochaetes make microbiological culture relatively difficult.

There was no obvious explanation for the delay and subsequent persistent reduction in egg production in the group of birds inoculated with *B. pilosicoli*, particularly as only three birds were confirmed to be colonized. It is possible that the other birds were colonized, but were not detected because samples were only tested once a week. Colonization had ceased by the time egg production had started, and it could be speculated that greater losses may have occurred had colonization persisted. At postmortem, no evidence was found for abnormalities in the ovaries or oviducts, although again by the time the birds were killed they were laying normally. It is possible that their intestinal function was temporarily impaired, resulting in reduced nutrient uptake, although this was not reflected in reduced body weight gain. The birds did develop transient frothy brown faeces with a slightly increased faecal water content, but did not exhibit obvious signs of diarrhoea. It was of interest that the birds in this group had altered caecal contents at postmortem, and this may reflect persistent subtle changes in the caecal microflora or in caecal function.

This investigation has confirmed that infection of commercial meat breeders with *B. pilosicoli* can significantly reduce egg production. Previously, we have shown that infection with this organism is widespread in commercial layer and meat breeder flocks in Australia (Stephens & Hampson, 1999). Thus, it would appear almost certain that the infection is causing important economic losses. Further experimental studies are needed to find reliable means to establish experimental colonization, to examine the pathological basis of production losses, and to improve diagnosis and control of *B. pilosicoli* infections.

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RÉSUMÉ

Infection expérimentale de reproductrices de type chair par des spirochètes intestinaux Brachyspira (Serpulina) pilosicoli entraînant une diminution de la production d'œufs

Le pouvoir pathogène des spirochètes intestinaux *Brachyspira (Serpulina) pilosicoli* et *Brachyspira innocens* a été évalué chez des animaux adultes. Trente reproductrices de type chair et de variété commerciale Cobb, âgées de 17 semaines, ont été hébergés en cage individuelle, et réparties en trois groupes de 10 sujets. Les oiseaux du groupe A témoin ont été inoculés avec du bouillon stérile. Les oiseaux des deux autres groupes B et C ont été inoculés, respectivement, avec un isolat de *B.innocens* ou de *B. pilosicoli*. Les oiseaux ont été observés tous les jours et ont été sacrifiés à l'âge de 41 semaines. L'infection n'a pas eu d'effet sur le gain de poids, mais l'inoculation de *B. pilosicoli* a entraîné une augmentation transitoire du contenu fécal en eau. L'infection par *Brachyspira innocens* n'a pas eu d'effet sur la production d'œufs, mais l'infection par *B. pilosicoli* a entraîné un retard du début de la ponte et une réduction très significative de la production d'œufs durant les 11 premières semaines de ponte. Cette étude confirme que *B. pilosicoli* peut entraîner de sérieuses pertes de production d'œufs chez les adultes, alors que *B. innocens* n'est manifestement pas pathogène.

ZUSAMMENFASSUNG

Die experimentelle Infektion von Mastelterntieren mit der Darmspirochäte Brachyspira (Serpulina) pilosicoli verursacht eine Reduzierung der Eierproduktion

Das pathogene Potential der anaeroben Darmspirochäten *Brachyspira (Serpulina) pilosicoli* und *Brachyspira innocens* wurde an ausgewachsenen Hühnern untersucht. Dreißig 17 Wochen alte Cobb-Mastelterntiere wurden in drei Gruppen von 10 Hennen einzeln in Käfigen gehalten. Die Kontrolltiere (Gruppe A) wurden mit steriler Nährbouillon scheininfiziert. Die Hennen in den anderen zwei Gruppen (B und C) wurden mit einem Isolat von *B. innocens* bzw. *B. pilosicoli* inokuliert. Die Tiere wurden täglich kontrolliert und im Alter von 41 Wochen getötet. Die Infektion hatte keinen konstanten Effekt auf die Körpergewichtszunahme, aber die Inokulation mit *B. pilosicoli* führte zu einer vorübergehenden Erhöhung des Wassergehalts im Kot. Die Infektion mit *Brachyspira innocens* hatte keinen Effekt auf die Eierproduktion, aber die Infektion mit *B. pilosicoli* bewirkte einen verzögerten Beginn der Legetätigkeit und eine hoch signifikante Verminderung der Eierproduktion in den ersten 11 Legewochen. Diese Studie bestätigt, dass *B. pilosicoli* bei ausgewachsenen Hühnern gravierende Eierproduktionsausfälle verursachen kann, während *B. innocens* nicht sichtlich pathogen ist.

RESUMEN

La infección experimental de reproductoras con la espiroqueta intestinal Brachyspira (Serpulina) pilosicoli causa reducción en la producción de huevos

Se evaluó el potencial patogénico de las espiroquetas anaeróbicas intestinales *Brachyspira (Serpulina) pilosicoli* y *Brachyspira innocens* en aves adultas. Treinta reproductoras Cobb de 17 semanas de edad se alojaron individualmente en tres grupos de 10 aves. Las aves control, grupo A, se inocularon con medio de caldo estéril. Las aves de los otros dos grupos, B y C, se inocularon respectivamente con una cepa de *B.innocens* o de *B.pilosicoli*. Las aves se monitorizaron diariamente y se sacrificaron a las 41 semanas de edad. La infección no tuvo un efecto manifiesto en la ganancia de peso diaria, pero la inoculación con *B. pilosicoli* resultó en un incremento transitorio del contenido hídrico fecal. La infección con *Brachyspira innocens* no tuvo efecto sobre la producción de huevos, pero la infección con *B. pilosicoli* causó un retraso en el comienzo de la puesta y una reducción significativa de la producción de huevos en las primeras 11 semanas de puesta. Este estudio confirma que *B.pilosicoli* puede causar pérdidas importantes en la producción de huevos en aves adultas, mientras que *B.innocens* no es claramente patógena.