Infection with a pathogenic turkey coronavirus isolate negatively affects growth performance and intestinal morphology of young turkey poults in Canada

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Turkey coronavirus (TCoV) is an important viral pathogen causing diarrhoea of young turkey poults that is associated with sizeable economic losses for the turkey industry. Using a field isolate that was found to be free from turkey astrovirus and avian reovirus we were able to reproduce the clinical disease associated with TCoV. Clinical signs and weight gain of poults during experimental infections were compared with age-matched, uninfected controls. Poults infected at 2 days of age had 100% morbidity and 10% mortality, and birds infected at 28 days of age showed 75% morbidity and no mortality. Diarrhoea was consistently seen in infected poults at 2 to 3 days post infection (d.p.i.) with a duration of about 3 to 5 days. Mean body weights of birds infected at 2 or 28 days of age were significantly reduced compared with uninfected birds by 7 d.p.i. and remained significantly lower for the duration of the study. At 44 days of age, poults infected at 2 or 28 days of age weighed only 68.1% or 77.7%, respectively, compared with uninfected turkeys of the same age on the same diet, a mean difference in body weights of 683 or 477g, respectively. Infected birds had profound villus atrophy with some compensatory crypt hyperplasia at 5 to 7 d.p.i. Villus heights in the duodenum were significantly reduced at 7 d.p.i. We were able to reproduce enteric disease using only a pathogenic field isolate (MG10) of TCoV that negatively affected growth performance and intestinal morphology of young turkey poults.

Introduction

Viruses in the family Coronaviridae (order Nidovirales) belong to one of three widely recognized groups that infect mammalian and avian hosts. This classification was developed to reflect the antigenic relationships among various coronaviruses as well as their genomic sequences and organizations (Lai & Cavanagh, 1997; Sawicki et al., 2007). With the exception of coronaviruses isolated from a dead Beluga whale (Mihindukulasuriya et al., 2008) and Asian leopard cats (Dong et al., 2007), all known Group 3 coronaviruses infect birds. Most Group 3 coronaviruses share a high degree of antigenic as well as genomic similarities based on sequencing data from full-length or partial genome sequences. Recently, the Group 3 coronaviruses were separated into three subgroups based on genome sequencing and organization (Woo et al., 2009). Group 3a coronaviruses include infectious bronchitis virus and turkey coronavirus (TCoV), as well as some more recently recognized viruses such as quail coronavirus, pigeon coronavirus, pheasant coronavirus, duck coronavirus and goose coronavirus. Group 3a includes viruses characterized by either induction of respiratory and urinary tract infections such as infectious bronchitis virus and pheasant coronavirus or by induction of enteric diseases such as TCoV and quail coronavirus (Pennycott, 2000; Cavanagh, 2001; Jonassen et al., 2005; Circella et al., 2007; Gomaa et al., 2008a; Woo et al., 2009). A unique coronavirus isolated from a whale (Mihindukulasuriya et al., 2008) is the only recognized member of Group 3b, while several recently described coronaviruses isolated from wild birds belong to Group 3c along with the coronavirus from Asian leopard cats (Dong et al., 2007; Woo et al., 2009). TCoV was first reported in several turkey farms in USA suffering from enteritis, and was referred to as 'mud fever' (Peterson & Hymas, 1951). Later, it was renamed “bluecomb disease” based on a similar clinical syndrome affecting chickens. Several outbreaks were reported throughout the USA and Canada, and were a cause of severe economic losses for the turkey industry during the 1950s and 1960s (Nagaraja & Pomeroy, 1997). TCoV can spread rapidly via the faecal-oral route (Naqi et al., 1972) and can infect turkey at different ages, resulting in high morbidity and varying mortality. Affected birds usually have diarrhoea, ruffled feathers, decreased feed and water consumption in addition to poor growth rate (Barnes et al., 1997; Yu et al., 2000; Ismail et al., 2003). Even after the disappearance of the
clinical signs, infected birds can still shed TCoV particles in their droppings up to 7 weeks post infection (Larsen, 1979; Breslin et al., 2000, Gomaa et al., 2009). Although TCoV is not frequently isolated from the faeces of healthy turkeys (Pantin-Jackwood et al., 2008), seroprevalence of the virus in commercial breeder and meat turkey flocks remains quite high (Gomaa et al., 2008).

The goals of the current study were to assess the impact of TCoV infection in young turkey poults using an isolate obtained from an outbreak on a commercial turkey farm (Gomaa et al., 2008a). Clinical signs and weight gain of poults were followed through the course of an infection and compared with age-matched uninfected controls. Histological observations and morphometric analysis of the duodenal mucosa was correlated with these clinical and growth observations.

Material and Methods

Virus. Turkey coronavirus (TCoV-MG10) was obtained from an Ontario turkey farm suffering from cases of acute enteritis and diarrhoea (Gomaa et al., 2008a). This TCoV could not be propagated successfully in cell culture or in embryonated turkey eggs. Therefore, all infections and challenges were initiated using a single filtrate obtained from the intestinal tissue of one turkey culled from this flock that was suffering from clinical disease consistent with TCoV. Briefly, intestinal tissue from this single infected bird was homogenized in 0.9% phosphate-buffered saline and then clarified by centrifugation at 4000 × g for 15 min. The supernatant was filtered sequentially through 0.45 µm and 0.22 µm membrane filters (Millipore, Bedford, Massachusetts, USA) and stored at −80 °C until use. To ensure uniformity, the same virus-containing filtrate was used throughout this study.

Extensive testing of the resulting TCoV-MG10 filtrate was carried out to exclude the possibility of the presence of any pathogens other than TCoV. Negative staining of the filtrate and subsequent transmission electron microscopic examination demonstrated only virus particles with morphological features and virion dimensions that were consistent with a coronavirus such as TCoV; no other viral particles of any other size or appearance were detected using transmission electron microscopic. The TCoV-MG10 filtrate was also submitted to the Animal Health Laboratory (University of Guelph, Canada) for detection of the presence of other potential pathogen(s). Bacterial culturing techniques indicated that the filtrate was free from bacterial contamination, excluding pathogens such as Salmonella, Escherichia coli, Campylobacter or Clostridium in the filtrate. No growth was reported on a wide variety of typical broth and plate cultures. The filtrate also tested negative for the presence of mycoplasmas (specifically Mycoplasma gallisepticum, Mycoplasma meleagridis and Mycoplasma synoviae). The filtrate was also tested for the presence of culturable viruses by egg inoculation and inoculation of several avian and mammalian cell cultures. No viral growth was reported in either eggs or cultures, indicating the absence of enteric viral pathogens that affect turkeys such as reoviruses, adenoviruses or enteroviruses that can be cultured. Undiluted viral inoculum also tested negative by reverse transcriptase-polymerase chain reaction (RT-PCR) for the presence of turkey astrovirus (see Sellers et al., 2004) as well as avian reovirus (see Bruhn et al., 2005). Furthermore, poults infected with this same inoculum never demonstrated any evidence of infection with turkey astrovirus or avian reoviruses when tested repeatedly using the same RT-PCR methods (see Gomaa et al., 2009).

The complete genomic sequence for the TCoV-MG10 isolate is available in GenBank (NC_010800; see Gomaa et al., 2008a). To provide an estimate of viral concentration, the TCoV-MG10 suspension was titrated by RT-PCR using primers specific for the nucleocapsid gene (see Gomaa et al., 2009). Virus-containing suspension diluted 10⁻⁴ was able to produce a weak PCR product of the expected 330 base pairs in size.

Turkeys and experimental design. Fifty 1-day-old commercial turkey poults were obtained from a high-biosecurity primary breeder facility (Hybrid Turkey, Kitchener, Ontario, Canada). Although the poults themselves were not tested specifically for the presence of Salmonella, Mycoplasma and other pathogens, this breeder routinely tests for Salmonella and Mycoplasma; thus, the poults are considered free of Salmonella and Mycoplasma. The poults were transported on their day of hatch directly to steam-cleaned and paraformaldehyde-sterilized isolation rooms within Biohazard Level 2 containment housing (Isolation Unit, Central Animal Facility, University of Guelph). All feed and litter that were used in the experimental infections were kept within each isolation room and only used for that room. Poults were provided feed and water ad libitum. Birds were handled and housed in accordance with the guidelines established by the Canadian Council on Animal Care.

Birds were wing-tagged, weighed and divided into control (30 birds) and infected (20 birds) groups that were housed separately in two identical isolation rooms maintained under identical conditions of lighting and heating. At 2 days of age, poults in the control group were each inoculated orally with 100 µl phosphate-buffered saline, and poults in the infected group were each inoculated orally with 100 µl viral suspension containing TCoV-MG10. At 21 days of age, birds in the infected group received a second dose of 100 µl filtrate containing TCoV-MG10 orally. At 28 days of age, eight birds from the uninfected control group were moved to the room housing the infected poults and there infected orally with TCoV-MG10 in the same manner as described above for 2-day-old poults.

Clinical observations and body weights. The body weight of each bird was recorded at 1 day of age and once weekly thereafter until 6 weeks of age. Throughout the infection trials, poults were monitored daily for the appearance of clinical signs. For poults inoculated orally with TCoV-MG10 at 2 days of age, birds were scored individually as positive/ negative for signs of diarrhoea (based on appearance of cloacal contents, faecal staining surrounding the vent and/or faecal consistency) at 1, 2, 3, 4, 6, 9, 11, 13, 16, 18 and 20 days post inoculation (d.p.i.). The same observations were made at 1 to 5 d.p.i. for birds inoculated for a second time at 21 days of age (20 d.p.i.). Poults inoculated for the first time at 28 days of age were assessed for the presence of diarrhoea at 1 to 5, 7, 9, 11, 13, 15 and 16 d.p.i., when the experiment was terminated.

Histopathological and morphometric examination. Two birds from the control and infected groups were selected at random at 3, 5, 7, 14 and 20 d.p.i. for the collection of tissues. In total, 20 birds (10 infected birds and 10 age-matched control birds) were subjected to post-mortem and histopathological examination. Birds were examined for the presence of macroscopic lesions in the small and large intestines. Pieces of the duodenum, jejunum, ileum and caeca were collected in 10% buffered formalin and then processed for paraffin embedment and sectioning by standard methods. Sections (approximately 5 µm thick) were stained with haematoxylin and eosin. Morphometric analysis of the intestinal villar architecture was conducted on tissues from all 20 birds. For each bird, measurements were taken of 10 crypt-villus units (longitudinally sectioned villus and associated crypt) from the approximate midpoint of the duodenal loop. For morphometric analysis, measurements were made with the aid of ImageJ Lite Version 7 software (IMT i-Solution Inc., Vancouver, British Columbia, Canada) using images captured on an Infinity3-1C camera (Lumenera Corp., Ottawa, Ontario, Canada) fitted to an Olympus AX70 Provis microscope (Olympus Optical Co. Ltd, Tokyo, Japan). The following measurements were obtained from 10 apparently complete crypt-villus units for each bird: villus length (VL); villus width (VW); crypt depth (CD); and crypt width (CW).

Statistical analysis. All measurements are expressed as mean ± standard deviation. Mean measurements (VL, VW, CD, CW) and mean body weights were compared between age-matched infected and uninfected turkey poults. Means were considered significantly different at P ≤0.05 using a one-tailed Student’s t test for two samples assuming unequal variances.

Results

Clinical observations. Birds inoculated at 2 days of age. Poults started developing clinical signs at 2 d.p.i. in the control group and at 5 d.p.i. in the infected group. Poults in the infected group received a second dose of 100 µl filtrate containing TCoV-MG10 orally. At 28 days of age, eight birds from the uninfected control group were moved to the room housing the infected poults and there infected orally with TCoV-MG10 in the same manner as described above for 2-day-old poults.
form of diarrhoea, frothy droppings, ruffled feathers and decreased food and water consumption. The control poult were alert and active, whereas infected birds were depressed, huddled and many had heavily soiled vents; the morbidity rate reached 100% at 3 d.p.i. when these clinical signs were most apparent. Two of 20 birds died at 3 d.p.i., giving a mortality rate of 10% with the MG10 strain of TCoV. The duration of diarrhoea was 3 to 5 days in primary infections of birds inoculated at 2 days of age (Figure 1).

Birds inoculated at 2 and at 21 days of age. Birds previously infected with TCoV-MG10 and then challenged with a second dose of virus at 21 days of age (20 d.p.i.) showed neither diarrhoea nor any other clinical sign throughout the experiment (Figure 1).

Birds inoculated at 28 days of age. Eight previously uninfected birds inoculated orally with TCoV-MG10 at 28 days of age were susceptible to infection, and six out of eight developed clinically apparent diarrhoea for a duration similar to poult first inoculated at 2 days of age (Figure 2).

Weight gain. Weekly mean body weights from 1 day of age until the end of this study at 44 days of age were compared between poult challenged at 2 or 28 days of age with uninfected control poult housed under identical conditions (Figure 3). Poult infected at 2 days of age had significantly reduced mean body weight (P < 0.05) in comparison with age-matched, uninfected control birds, starting as early as 1 week of age (6 d.p.i.). This significant reduction in mean body weight persisted for the entire 6 weeks of the experiment. At 44 days of age, turkeys that were infected with TCoV at 2 days of age weighed only 68.1% of the uninfected control birds on the same diet; a mean body weight difference of 683 g. Poult that were inoculated at 28 days of age showed a similar pattern of reduced weight gain after infection with TCoV-MG10. These TCoV-infected poult had significantly lower mean body weight (P < 0.05) by 7 d.p.i. (35 days of age), and by 44 days of age poult from this second infected group had mean body weights that were only 77.7% of uninfected birds of the same age, a mean difference of 477 g (Figures 3 and 4).

Post-mortem observations and histopathology. Poult that were inoculated at 2 days of age were sampled at random at 3, 5, 7, 14 and 20 d.p.i. (two poult from each of the infected and uninfected groups). During the examination of infected and control poult selected 3, 5 or 7 d.p.i., the intestines were found to be swollen and filled with yellow frothy digesta. The walls of the intestines were thin and pale compared with uninfected control poult of the same age. At 14 and 20 d.p.i., there were no obvious macroscopic lesions visible in the infected poult and their intestinal contents appeared comparable with the uninfected control poult sampled the same day.

Microscopically, the most obvious difference between the infected (Figure 5b to d and 6c to f) and control poult (Figure 5a and 6a,b) was the reduced duodenal villus heights (see Morphometric analysis, below) in the infected birds. At 3 d.p.i., some evidence of villus atrophy with reduced villus height and limited infiltration into the lamina propria was seen in infected poult (Figure 5b). By 5 d.p.i., infiltration of the lamina propria, particularly surrounding the crypts, was pronounced and there was evidence of sloughing of epithelial cells and some fusion of adjacent villi (Figures 5c and 6c,d). In limited areas of the duodenum there was evidence of complete denuding of the villar lamina propria with complete sloughing of the overlying epithe-
lium. In one of the two infected poults examined at 5 d.p.i. there was dramatic infiltration of lymphocytes and heterophils into the lamina propria of the villi and crypts. Within the crypt epithelium of infected poults at both 5 and 7 d.p.i., there was evidence of crypt hyperplasia with numerous crypt enterocytes undergoing division (Figure 6e). By 7 d.p.i. the inflammatory infiltration of the lamina propria continued, and there was widespread evidence of sloughing of large portions of the villi and surrounding lamina propria of the duodenal epithelium in the infected poults (Figure 5d and 6e). At 7 d.p.i. the cellular infiltrate also contained eosinophils. Eosinophils were also seen within the lamina propria and between villar enterocytes at 14 and 20 d.p.i. in the infected poults. By 14 d.p.i., the villus height appeared reduced when compared with the villi of uninfected control birds and there appeared to be fewer, but broader, villi remaining. Some of the remaining villi retained denuded tips that possessed only lamina propria devoid of overlying enterocytes (Figure 6f). At 14 d.p.i., there appeared to be many more crypts than villi. By 20 d.p.i., the villar architecture had largely returned to normal; however, the villi appeared shorter than in age-matched uninfected birds and villi continued to appear somewhat wider and less numerous. A few scattered villar tips remained denuded of enterocytes but no evidence of active epithelial sloughing was seen. Cellular infiltration of the lamina propria was reduced but not eliminated by 20 d.p.i.

Morphometric analysis. Upon histological examination, the most obvious changes were the apparent decrease in villus height in the duodenum of poults infected at 2 days of age compared with the heights of duodenal villi of uninfected, age-matched control birds. Morphometric analysis of at least 10 crypt-villus units from each poult that was sampled permitted comparisons between VL, VW, CD and CW within the duodenum of infected and uninfected poults (Table 1). A significant reduction in mean VL of the infected birds was found at 7 d.p.i. compared with age-matched uninfected poults (Figure 7). Although not supported statistically, the mean VL of infected birds remained numerically smaller at 14 and 20 d.p.i. when compared with age-matched uninfected poults. Likewise, at 5 d.p.i. CD was increased significantly in infected birds compared with the controls. The mean CD remained numerically greater at 7 d.p.i.

Figure 4. Gross appearance of turkey poults at 28 days of age that were infected with TCoV-MG10 (left) or uninfected (right) at 2 days of age. On average, turkeys infected with TCoV-MG10 weighed 228 g less than control birds at this age.

Figure 5. Histological appearance of the duodenal mucosa of young turkey poults. 5a: Uninfected turkey poult at 6 days of age. 5b: Infected turkey poult at 4 days of age (3 d.p.i.) had nearly normal appearance. 5c: Infected turkey poult at 6 days of age (5 d.p.i.) demonstrating cellular infiltration of the lamina propria of the villi and surrounding the crypt. There was some shortening of the villi with modest increase in crypt depth. 5d: Infected turkey poult at 8 days of age (7 d.p.i.) showing significant villus shortening with sloughing of denuded villar lamina propria (arrows) and some increase in crypt depth. Infiltration of the lamina propria, especially surrounding the crypts, with lymphocytes and monocytes was evident. Bars = 500 μm.
Discussion

Using pathogenic TCoV (TCoV-MG10) obtained from an Ontario turkey farm suffering from severe cases of acute enteritis and diarrhoea (Gomaa et al., 2008a), we followed the course of the virus infection with respect to clinical signs (diarrhoea) and body weights in addition to histopathological and morphometric changes to the intestinal mucosa over the course of a primary infection.

The majority of turkey poults inoculated with TCoV-MG10 at 2 days of age demonstrated clinically apparent disease in the form of diarrhoea with 100% morbidity and 10% mortality, whereas in birds infected for the first time at 28 days of age there was 75% morbidity and no mortality. Infection of young poults with TCoV-MG10 caused severe clinical signs with relatively high mortality rates in contrast to infection with egg-adapted TCoV isolates (Guy et al., 2000). Concurrent infection of even egg-adapted TCoV with enteropathogenic E. coli had resulted in high mortality as well as poor weight gain in doubly infected turkeys (Guy et al., 2000). In the latter study, neither enteropathogenic E. coli nor egg-adapted TCoV alone produced mortality that differed from uninfected control poults. Based on our testing for extraneous pathogens, conducted both during the experimentation as well as on the inoculum itself, we are reasonably confident that the only pathogenic agent present was TCoV and that this virus was solely

Figure 6. Histological appearance of the duodenal mucosa of young turkey poults. 6a: Uninfected turkey poult at 6 days of age. 6b: Uninfected turkey poult at 8 days of age. 6c: Infected turkey poult at 6 days of age (5 d.p.i.) showing typical infiltration of lymphocytes and heterophils in the lamina propria surrounding the crypts and extending into the villi (arrows). 6d: Higher magnification of villi in 6c showing locally affected villar enterocytes (arrows). These regions were relatively common (several regions per villi) at 5 d.p.i. within the proximal half of the villi. 6e: Crypts of an infected poult at 8 days of age (7 d.p.i.) showing large numbers of dividing cells within the crypt epithelium (white arrows) and modest cellular infiltration of the lamina propria (black arrow). 6f: Mucosa of a poult at 15 days of age (14 d.p.i.) with localized sloughing of entire, denuded villi. The underlying crypts remained largely intact. Bars =100 μm.
Table 1. Morphometric analysis of the duodenal mucosa of turkey poults infected at 2 days of age with TCoV-MG10 compared with age-matched control poults

<table>
<thead>
<tr>
<th></th>
<th>3 d.p.i. (mm)</th>
<th>5 d.p.i. (mm)</th>
<th>7 d.p.i. (mm)</th>
<th>14 d.p.i. (mm)</th>
<th>20 d.p.i. (mm)</th>
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<tr>
<td><strong>Villus length</strong></td>
<td></td>
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<tr>
<td>Control</td>
<td>2.026 ± 0.257</td>
<td>1.844 ± 0.455</td>
<td>2.167 ± 0.058</td>
<td>1.518 ± 0.035</td>
<td>1.835 ± 0.529</td>
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<tr>
<td>Infected</td>
<td>1.443 ± 0.576</td>
<td>1.176 ± 0.131</td>
<td>0.874 ± 0.148*</td>
<td>1.261 ± 0.388</td>
<td>1.127 ± 0.099</td>
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<td><strong>Villus width</strong></td>
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<td>Control</td>
<td>0.094 ± 0.025</td>
<td>0.120 ± 0.017</td>
<td>0.139 ± 0.038</td>
<td>0.132 ± 0.007</td>
<td>0.105 ± 0.010</td>
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<td>Infected</td>
<td>0.127 ± 0.019</td>
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<td>0.124 ± 0.017</td>
<td>0.186 ± 0.035</td>
<td>0.164 ± 0.050</td>
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<td><strong>Crypt depth</strong></td>
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<tr>
<td>Control</td>
<td>0.103 ± 0.025</td>
<td>0.093 ± 0.012</td>
<td>0.097 ± 0.033</td>
<td>0.128 ± 0.013</td>
<td>0.116 ± 0.006</td>
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<tr>
<td>Infected</td>
<td>0.103 ± 0.009</td>
<td>0.133 ± 0.012*</td>
<td>0.108 ± 0.017</td>
<td>0.078 ± 0.028</td>
<td>0.115 ± 0.042</td>
</tr>
<tr>
<td><strong>Crypt width</strong></td>
<td></td>
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<tr>
<td>Control</td>
<td>0.067 ± 0.024</td>
<td>0.092 ± 0.022</td>
<td>0.078 ± 0.008</td>
<td>0.096 ± 0.029</td>
<td>0.077 ± 0.010</td>
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<tr>
<td>Infected</td>
<td>0.071 ± 0.001</td>
<td>0.070 ± 0.001</td>
<td>0.070 ± 0.011</td>
<td>0.173 ± 0.049</td>
<td>0.196 ± 0.161</td>
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Measurements are expressed as mean ± standard deviation in millimetres. *Means in the infected group that differ significantly from age-matched control poults (Student’s t test assuming unequal variance, P < 0.05).

Attempts to re-infect poults previously exposed to TCoV resulted in no resumption of clinical signs (current study) or viral shedding (Gomaa et al., 2009), suggesting that protective immunity is acquired after a single infection with TCoV. This is consistent with earlier studies showing that turkeys recovered from infection with TCoV showed resistance to any further infection with the same virus (Pomeroy et al., 1975; Nagaraja et al., 1997).

In a previous study (Gomaa et al., 2009), the proportion of the infected turkey poults shedding TCoV particles was determined using RT-PCR methods. Interestingly, in poults infected at either 2 or 28 days of age, the appearance of clinical signs (diarrhoea) was concurrent with or slightly preceded viral shedding. Regardless of the age of turkey that was infected with TCoV-MG10, shedding of virus continued for some days (frequently more than 1 week) after the cessation of obvious clinical signs. This finding was consistent with a study by Breslin et al. (2000) who were able to detect virus shedding in the faeces of turkeys infected experimentally with TCoV for up to 7 weeks post infection, even after cessation of clinical signs.

To further investigate the effects of TCoV on the intestinal epithelium, morphometric analysis of the duodenal mucosa was performed (Figure 7). The mean villus height for each group was calculated and expressed as mean ± standard deviation. The results showed that infected poults had significantly reduced villus height compared with age-matched controls at 3, 5, 7, and 14 d.p.i., but not at 21 d.p.i. (Student’s t test assuming unequal variance, P < 0.05).

Figure 7. Mean duodenal villus height of turkey poults infected at 2 days of age with TCoV MG10 (white bars) at various days post infection compared with age-matched uninfected turkey poults (black bars) housed under identical conditions. Error bars represent the standard deviation of the mean villus height for each day; bar marked with an asterisk is significantly different (Student’s t test assuming unequal variance, P < 0.05) from the villus height of control poults on the same day.

Changes to the microscopic appearance of the duodenal mucosa seen in poults infected with TCoV-MG10 were consistent with previous observations for this virus (Brown et al., 1997; Guy, 1998; Breslin et al., 2000; Ismail et al., 2003; Teixeira et al., 2007). Villus atrophy and marked infiltration of the lamina propria of the villi and crypts with lymphocytes and heterophils was noted starting as early as 3 d.p.i., and was most notable at 7 d.p.i. when villus heights of infected birds were significantly reduced compared with age-matched controls.
Widespread sloughing of intestinal villi was noted in the infected poults at 7 and 14 d.p.i.; the intestinal mucosa was recovering normal architecture by 20 d.p.i., but some cellular infiltration of the lamina propria remained at this time.

In summary, experimental infections in young turkey poults suggest that at least some strains of TCoV are capable of producing significant depression in weight gain with severe clinical disease in the apparent absence of other infections. Infections initiated with TCoV alone in 2-day-old or in 28-day-old poults caused reductions in weight gain following infection. Significantly lower weights persisted for the duration of the trial (44 days), suggesting that turkey poults may be permanently affected with respect to weight gain even after recovering from TCoV infections (Dea & Tijssen, 1988; Brown et al., 1997; Berslin et al., 2000; Guy et al., 2000; Lin et al., 2002; Ismail et al., 2003; Cavanagh, 2005; Culver et al., 2006). Although feed conversion was not examined directly in the present study, the gross lesions and histological appearance of the intestinal tract of the infected turkey poults would suggest that feed conversion would be adversely affected by infection with TCoV-MG10 and would contribute to the poor weight gains demonstrated by infected birds. This would be consistent with the observed impact of TCoV infections on commercial meat turkey operations (Guy et al., 2000; Ismail et al., 2003).

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