RESEARCH ARTICLE
Open Access

Journal of Veterinary and Animal Sciences
ISSN (Print): 0971-0701, (Online): 2582-0605
https://doi.org/10.51966/jvas.2022.53.4.

Occurrence and distribution of gross and microscopic gastrointestinal lesions in pigs in Northern Kerala#

V. Elizabeth[1], R. Anoopraj[2*], J.G. Ajith[3], M. Pradeep[2], R. Chintu[4], P. Hamza[2], T. Dhanya[1] and M.T.K. Alin[1]
Department of Veterinary Pathology,
College of Veterinary and Animal Sciences, Pookode, Wayanad-673576,
Kerala Veterinary and Animal Sciences University, Kerala, India.

Citation: Elizabeth, V., Anoopraj, R., Ajith, J.G., Pradeep, M., Chintu, R., Hamza, P., Dhanya, T. and Alin, M.T.K. 2022. Occurrence and distribution of gross and microscopic gastrointestinal lesions in pigs in Northern Kerala. *J. Vet. Anim. Sci.* **53**(4):
DOI: https://doi.org/10.51966/jvas.2022.53.4.

Received: 04.04.2022 *Accepted: 09.06.2022* *Published: 31.12.2022*

Abstract

Gastroenteritis in swine herds pose a big constraint in pig farming due to the economic losses accompanying it. The present study utilised 57 pig carcasses to assess the occurrence and distribution of various gastrointestinal lesions in pigs in Northern Kerala. Gross changes in stomach were observed in 84.21 per cent of carcasses in which mucosal congestion (61.40 per cent) was the most frequent lesion. Principal microscopic gastric lesions observed were mucosal necrosis, cellular infiltration in lamina propria, hyperplastic epithelium, hyperkeratinisation and fibrino-necrotic membrane. Gross changes in at least one region of intestine were evident in all the samples. The most common lesions in intestines were engorgement of mesenteric blood vessels (80.70 per cent) followed by catarrhal enteritis (73.68 per cent). Salient gross lesions such as mucosal ulcers were observed only in caecum and colon, diphtheritic membrane formation was observed in ileum, caecum and colon. Major histological lesions observed in intestines were mucosal necrosis (89.47 per cent) followed by cellular infiltration in lamina propria (68.42 per cent). Histologically, the major site-specific lesions observed were Brunner gland hyperplasia in duodenum, necrosis, hyperplasia, and lymphocytic depletion of Peyer's patches in ileum, granulomatous infiltration in ileum and colon, neutrophilic colitis and goblet cell depletion in colon. The present study observed a higher incidence of gastrointestinal variations in pigs irrespective of age and further research is needed to understand the etiological factors contributing to such heavy burden of gastrointestinal lesions.

Keywords: *Stomach, intestine, pigs, lesions*

 # Part of MVSc thesis submitted to Kerala Veterinary and Animal Sciences University, Pookode, Wayanad, Kerala

1. *MVSc Scholar*
2. *Assistant Professor*
3. *Professor and Head*
4. *Associate Professor, Department of Veterinary Microbiology*

** Corresponding author: anoopraj@kvasu.ac.in, Ph: 9495361872*

Pig farming once considered least acceptable among Indian livestock farmers has gained momentum over the past few years. Maximum production with less input has turned the tide in favour of pig farming in India. However, gastroenteritis continues to threaten the emerging piggery sector, resulting in substantial financial losses to marginal farmers (Hartadi *et al*., 2020). Gastroenteritis drastically affects productivity in pigs and if left untreated, results in increased morbidity and mortality rates in pigs (Si *et al*., 2021). It can affect pigs of all ages and is a serious cause of concern particularly in neonates and weaned piglets. According to Sinha *et al*. (2019), gastrointestinal disorders are the leading cause of morbidity and mortality in piglets in India. Among the different diseases impairing swine production in India, gastrointestinal disorder such as diarrhoea results in decreased weight gain in piglets especially in first week of birth (Kylla *et al*., 2019). A thorough and updated knowledge on the incidence and distribution of the different gastrointestinal lesions in pigs will aid in effective management strategies. Although several pieces of literature explain the pathology associated with gastroenteritis (Sithara *et al*., 2016; Prasad *et al.,* 2018), only a few describe the incidence and distribution of different gastrointestinal lesions in pigs in Kerala. Thus, the present study was aimed at a systematic recording and characterisation of gross and histomorphological changes observed in the stomach and intestines in pigs.

Materials and methods

A total of 57 pig carcasses brought for post mortem to the Department of Veterinary Pathology, College of Veterinary and Animal Sciences, Pookode, Wayanad, from organised and unorganised farms in and around Wayanad, Calicut, and Thrissur, from January 2021 to November 2021, formed the material of the study. The piglets were divided based on the age into different groups such as neonates (0-<3weeks), pre-weaners (≥3weeks-<2months), weaners (≥2months-≤5months), growers (>5months-<8months) and finishers (≥8 months). Gross lesions were recorded and intestinal tissue samples were collected in 10 per cent neutral buffered formalin for histopathological examination and processed following standard histopathological procedures (Suvarna *et al.*, 2013). The stained sections were examined under a light microscope (Zeiss Axio Scope A1 microscope) and microscopic lesions were recorded.

Results and discussion

Gross changes in stomach were observed in 84.21 per cent cases. Among the different gross lesions observed in stomach, vascular change such as gastric mucosal congestion was the predominant lesion observed in 61.40 per cent cases. The next major gross lesion was mucosal erosions (28.07 per cent) seen in at least one out of the two growers and finishers. Hyperkeratinisation in *pars oesophagea* as well as gastric mucosal ulceration were also mostly observed in the growers. Managemental practices, nutrition and infectious diseases predispose to the formation of gastric ulcers in swine (Cybulski *et al*., 2021). Chronic exposure to any of these risk factors that increased the fluid nature of the stomach, results in increased influx of the acidic contents into the non-glandular portion (De Witte *et al*., 2018). Studies conducted by Majekodunmi *et al*. (2013) reported hyperkeratinisation and gastric ulcers in younger piglets of three- six months of age with the common site of lesion being the *pars oesophagea* followed by cardia and fundus. In the present study, hyperkeratinisation which is prequel to ulcer formation was observed in the *pars oesophagea*. However, the ulcerations and erosions were more pronounced in the glandular mucosa than in *pars oesophagea*. The development of these lesions in glandular mucosa might be attributed to infectious diseases such as hog cholera, salmonellosis and PCV2 infections as well as dietary hygienic practices that disrupted the normal glandular mucosal epithelium (Correa *et al*., 2008; Omotosho *et al*., 2016). Another notable lesion in stomach was diphtheritic membrane formation in the glandular mucosa which was higher among growers. The different gross changes observed in stomach and their occurrence in different age groups is summarised in Table 1.

Histologically, gastric changes were evident in 96.49 per cent cases. The major

Table 1. Age-wise occurrence of gross changes in stomach

Lesions (No. of animals with lesions)	Age groups				
	Neonates (n=5)	Pre-weaners (n=5)	Weaners (n=41)	Growers (n=4)	Finishers (n=2)
Erosions (16)	2 (40%)	0	11 (26.83%)	2 (50%)	1 (50%)
Ulcers (9)	0	1 (20%)	7 (17.07%)	1 (25%)	0
Hyperkeratinisation (10)	0	0	9 (21.95%)	1 (25%)	0
Diphtheritic membrane (5)	1 (20%)	0	3 (7.32%)	1 (25%)	0
Mucosal haemorrhage (2)	0	0	2 (4.88%)	0	0
Serosal congestion (11)	1 (20%)	2 (40%)	8 (19.51%)	0	0
Mucosal congestion (35)	4 (80%)	2 (40%)	24 (58.54%)	3 (75%)	2 (100%)

histopathological lesions in stomach comprised of cellular infiltration in lamina propria followed by mucosal necrosis. Gastric mucosa could be considered normal if few inflammatory cells are found in lamina propria with no apparent changes in the glandular and mucosal epithelium (Mendes *et al*., 1991). In the present study, the diffused distribution of inflammatory cells varying from mild to intense infiltration with glandular degeneration indicated a significant inflammatory response. Histologically, hyperkeratinised epithelium, mucosal erosions and ulcers of varying degrees of severity were observed and these could be associated with multifactorial etiologies (Cybulski *et al.*, 2021). Cytoplasmic vacuolisation in gastric epithelial cells as observed in the present study might be attributed to an adaptive response at cellular level (Miller and Zachary, 2017). Diphtheritic membranes observed in gastric mucosa were histologically presented as pseudo membranes with fibrino-necrotic debris and degenerated inflammatory cells replacing the mucosa in 8.77 per cent cases. The different vascular changes observed were mucosal and submucosal congestion and haemorrhage, mucosal and submucosal edema in 61.40 per cent, 19.30 per cent and 28.07 per cent cases respectively. These vascular changes might be associated with venous infarction or endotoxaemia (Robbins *et al*., 2014). The different histopathological changes in stomach and their occurrence in different age groups is summarised in Table 2.

Table 2. Age-wise occurrence of histopathological changes in stomach

Lesions (No. of animals with lesions)	Age groups				
	Neonates (n=5)	Pre-weaners (n=5)	Weaners (n=41)	Growers (n=4)	Finishers (n=2)
Mucosal necrosis (32)	3 (60%)	4 (80%)	22 (53.66%)	3 (75%)	0
Cellular infiltration in lamina propria (38)	2 (40%)	3 (60%)	29 (70.73%)	4 (100%)	0
Mucosal and submucosal edema (16)	2 (40%)	1 (20%)	13 (31.71%)	0	0
Mucosal and submucosal congestion (35)	4 (80%)	2 (40%)	26 (63.42%)	2 (50%)	1 (50%)
Mucosal and submucosal haemorrhage (11)	1 (20%)	1 (20%)	9 (21.95%)	0	0
Epithelial vacuolization (2)	0	0	2 (4.88%)	0	0
Fibrino-necrotic membrane (5)	1 (20%)	0	3 (7.32%)	1 (25%)	0
Hyperkeratinisation (10)	0	5 (100%)	5 (12.20%)	0	0
Hyperplastic epithelium (4)	0	0	4 (9.76%)	0	0
Mucosal erosions (16)	2 (40%)	0	11 (26.83%)	2 (50%)	1 (50%)
Glandular degeneration (21)	2 (40%)	3 (60%)	15 (36.59%)	1 (25%)	0
Mucosal ulcers (9)	0	1 (20%)	7 (17.07%)	1 (25%)	0

Table 3. Age-wise occurrence of gross changes in intestines

Lesions (No. of animals with lesions)	Age groups				
	Neonates (n=5)	Pre-weaners (n=5)	Weaners (n=41)	Growers (n=4)	Finishers (n=2)
Mucosal ulcerations (15)	0	1 (20%)	11 (26.83%)	3 (75%)	0
Catarrhal enteritis (42)	3 (60%)	5 (100%)	29 (70.73%)	3 (75%)	2 (100%)
Haemorrhagic enteritis (17)	2 (40%)	2 (40%)	13 (31.71%)	0	0
Diphtheritic membrane (5)	0	0	5 (12.20%)	0	0
Flaccid intestines (35)	5 (100%)	5 (100%)	21 (51.21%)	2 (50%)	2 (100%)
Thin- walled intestines (26)	4 (80%)	2 (40%)	20 (48.78%)	0	0
Engorged mesenteric blood vessels (46)	5 (100%)	4 (80%)	32 (78.05%)	3 (75%)	2 (100%)
Mesocolonic edema (15)	0	4 (80%)	11 (26.83%)	0	0
Serosal congestion (39)	3 (60%)	5 (100%)	28 (68.29%)	1 (25%)	2 (100%)
Mucosal congestion (41)	5 (100%)	3 (60%)	29 (70.73%)	2 (50%)	2 (100%)

In the present investigation, diarrhoea was reported in 49.12 per cent of cases. However, gross and histopathological changes in intestines were observed in all the samples that were presented with and without a history of diarrhoea. In the present study, the consistent macroscopic changes observed in intestines in almost all age groups were diffuse catarrhal enteritis characterised by mucoid contents in lumen and engorgement of mesenteric blood vessels. Haemorrhagic enteritis with blood mixed contents in the intestinal lumen was observed in 29.82 per cent cases and were common in neonates and pre-weaners. Flaccid and thinning of intestines, characteristic of viral infections, were mostly evident in the proximal intestines (Kongsted *et al*., 2013; Chen *et al*., 2020). Mesocolonic edema was observed in 26.32 per cent cases belonging to pre-weaners and weaners. The occurrence of observed gross lesions in intestines in different age groups is represented in Table 3.

In the present study, mucosal ulcers with circular to irregular borders and depressed centre were predominant in the caecum and colon. Diphtheritic membrane formation characterised by the presence of diffuse yellowish plaques of fibrinous necrotic deposits was observed in the ileum, caecum, and colon of 8.77 per cent cases, all belonging to the weaner age group. Similar lesions in weaners have been reported in porcine salmonellosis (Sithara *et al.,* 2016; Prasad *et al*., 2018) and classical swine fever (Sudo *et al*., 2021).

Histologically, the lesions in intestine can be characterised into lesions involving the epithelium, mucosal and submucosal architecture and the type of inflammatory cell infiltrate (Erben *et al*., 2014). Lesions involving the epithelium observed in the present study included hyperplastic crypts, goblet cell hyperplasia, epithelial vacoulisation, crypt and glandular degeneration, crypt abscess and goblet cell depletion. Goblet cell hyperplasia was a consistent histopathological finding in cases presented with catarrhal enteritis. These epithelial changes indicated an attempt at repair and replacement of enterocytes (Gelberg, 2017). In the current study, certain lesions such as epithelial vacuolisation was observed only in duodenum and jejunum and goblet cell depletion was restricted to colon.

Lesions involving the mucosal and submucosal architecture included necrosis, hyperplastic submucosal glands, villous atrophy and fusion, mucosal ulcers, fibrino-necrotic membrane, depleted/hyperplastic or necrotic Peyer's patches and vascular changes such as edema, lymphangiectasis, congestion and haemorrhage. The predominant histopathological change observed in intestine was mucosal necrosis in 89.47 per cent cases. Villous atrophy in small intestine was a common finding in cases presented with thin-walled intestines. Another significant finding in duodenum was submucosal gland hyperplasia which could be a duodenal reaction to hypersecretion of acid into intestinal

lumen (Franzin *et al*., 1985). Hyperplasia or coagulative necrosis of Peyer's patches in ileum and lymphocytic depletion as observed in the present study has also been reported in porcine circovirus 2 infections by Rovira *et al.* (2002). Villous fusion and atrophy, submucosal lymphangiectasis, hyperplastic Peyer's patches, mucosal haemorrhages, submucosal edema were also described as the histopathological changes associated with transmissible gastroenteritis virus and porcine epidemic diarrhoea virus (Jung *et al*., 2014; Robbins *et al*., 2014). Mucosal ulcers presented in caecum and colon was observed histologically as multiple necrotic foci with accumulation of degenerated inflammatory cells. Diphtheritic membrane in ileum, caecum and colon was histologically presented as fibrino necrotic membrane replacing the mucosa in the 8.77 per cent cases.

The types of inflammatory cell infiltrates observed were mononuclear, polymorphonuclear or granulomatous. Mononuclear and polymorphonuclear cell infiltration of varying degrees of severity was observed in 68.42 per cent cases. Granulomatous infiltration characterised by the infiltration of multinucleated giant cells, epitheliod macrophages and histiocytes was observed only in the ileum and colon in 5.26 per cent cases. Neutrophilic colitis characterised by the necrotic mucosa along with degenerated neutrophils and severe goblet cell depletion in colon was observed in 26.32 per cent cases. Similar histopathological changes were reported in piglets infected with *Clostridium difficile* without any clinical history of diarrhoea (Yaeger *et al*., 2007). The occurrence of different histopathological changes observed

Table 4. Age-wise occurrence of histopathological changes in intestines

Lesions (No. of animals with lesions)	Age groups				
	Neonates (n=5)	Pre-weaners (n=5)	Weaners (n=41)	Growers (n=4)	Finishers (n=2)
Mucosal necrosis (51)	5 (100%)	3 (60%)	38 (92.68%)	4 (100%)	1 (50%)
Cellular infiltration in lamina propria (39)	3 (60%)	4 (80%)	28 (68.29%)	3 (75%)	1 (50%)
Mucosal and submucosal edema (19)	1 (20%)	4 (80%)	13 (31.71%)	1(25%)	0
Mucosal and submucosal congestion (15)	1 (20%)	1 (20%)	13 (31.71%)	0	0
Mucosal and submucosal haemorrhage (19)	2 (40%)	2 (40%)	14 (34.15%)	1(25%)	0
Epithelial vacuolization (3)	0	1 (20%)	2 (4.88%)	0	0
Fibrino-necrotic membrane (5)	0	0	5 (12.20%)	0	0
Brunner gland hyperplasia (4)	0	0	3 (7.32%)	0	1 (50%)
Goblet cell hyperplasia (33)	2 (40%)	5 (100%)	23 (56.09%)	3 (75%)	0
Hyperplastic crypts (11)	1 (20%)	1 (20%)	9 (21.95%)	0	0
Crypt and glandular degeneration (23)	4 (80%)	3 (60%)	15 (36.59%)	1 (25%)	0
Lymphangiectasis (11)	1 (20%)	0	9 (21.95%)	0	1 (50%)
Hyperplastic Peyer's patches (2)	0	0	2 (4.88%)	0	0
Necrotic Peyer's patches (2)	0	0	2 (4.88%)	0	0
Lymphocytic depletion in Peyer's patches (17)	0	1 (20%)	15 (36.59%)	1 (25%)	0
Granulomatous enteritis (3)	2 (40%)	0	1 (2.44%)	0	0
Villous atrophy (26)	4 (80%)	2 (40%)	20 (48.78%)	0	0
Villous fusion (9)	0	0	9 (21.95%)	0	0
Crypt abscess (4)	0	0	4 (9.76%)	0	0
Neutrophilic colitis (15)	0	1 (20%)	12 (29.27%)	2 (50%)	0
Goblet cell depletion (15)	0	2 (40%)	13 (31.71%)	0	0
Mucosal ulcer (15)	0	1 (20%)	11 (26.83%)	3 (75%)	0

in intestines in different age groups is given in Table 4.

In the present study, the weaned piglets comprised the major age group brought in for necropsy followed by neonates and pre-weaners. However, gastric lesions were observed mostly in neonates, growers and finishers. Since the enterocyte turnover rate is the highest compared to other organ systems, compensatory lesions in intestines were observed in all the age groups presented for post mortem (Gelberg, 2017). The present investigation observed a wide array of gross and histopathological changes in stomach and intestines that could be attributed to different viral and bacterial etiological agents, that might have occurred alone or in combination.

Conclusion

Gastroenteritis in pigs presented a concoction of pathological changes. The present study pointed out the severe incidences of gastric and intestinal lesions in pigs irrespective of age, and concludes that gastroenteritis which culminates in severe production losses, poses a serious risk to the swine industry. Further research should look into the early diagnosis of the different etiological factors for proper interventions and to cut short the production losses.

Acknowledgement

The authors thank the Kerala Veterinary and Animal Sciences University for providing the research grant and other facilities for the conduct of this study.

Conflict of interest

The authors declare no conflict of interest.

References

Chen, Y.M., Helm, E.T., Gabler, N., Hostetter, J.M. and Burrough, E.R. 2020. Alterations in intestinal innate mucosal immunity of weaned pigs during porcine epidemic diarrhoea virus infection. *Vet. Pathol.* **57**: 642–652.

Correa, A.M.R., Zlotowsk, P., de Barcellos, D.E.S.N., da Cruz C.E.F. and Driemeier, D. 2008. Gastric ulcers in pigs affected with postweaning multisystemic wasting syndrome. *Pesq. Vet. Bras*. **28**: 601-605.

Cybulski, P., Larska, M., Wozniak, A., Jabłonski, A. and Stadejek, T. 2021. The dietary risk factors of gastric ulcers in finishing pigs from 16 polish farms. *Agriculture* [online]**. 11:** 719. Available: https://doi.org/10.3390/agriculture11080719.

De Witte, C., Ducatelle, R. and Haesebrouck, F. 2018. The role of infectious agents in the development of porcine gastric ulceration. *Vet. J.* **236**: 56-61.

Erben, U., Loddenkemper, C., Doerfel, K., Spieckermann, S., Haller, D., Heimesaat, M.M., Zeitz, M., Siegmund, B. and Kuhl, A.A. 2014. A guide to histomorphological evaluation of intestinal inflammation in mouse models. *Int. J. Clin. Exp. Pathol.* **7**: 4557-4576.

Franzin, G., Musola, R., Ghidini, O., Manfrini, C. and Fratton, A. 1985. Nodular hyperplasia of Brunner's glands. *Gastrointest. Endosc.* **31**: 374-378.

Gelberg, H.B. 2017. Alimentary system and the peritoneum, omentum, mesentery and peritoneal cavity. In: Zachary, J.F. (ed.), P*athologic Basis of Veterinary Disease.* (7th Ed.). Elsevier, Missouri, pp. 324-405.

Hartadi, E.B., Effendi, M.H., Plumeriastuti, H., Sofiana, E.D., Wibisono, F.M. and Hidayatullah, A.R. 2020. A review of enterotoxigenic *Escherichia coli* infection in piglets: public health importance. *Sys. Rev. Pharm.* **11**: 687-698.

Jung, K., Wang, Q., Scheuer, K.A., Lu, Z., Zhang, Y. and Saif, L.J. 2014. Pathology of US porcine epidemic diarrhoea virus strain PC21A in gnotobiotic pigs. *Emerg. Infect. Dis.* **20**: 662-665.

Kongsted, H., Jonach, B., Haugegaard, S.,

Angen, O., Jorsal, S.E., Kokotovic, B., Larsen, L.E., Jensen, T.K. and Nielsen, J.P. 2013. Microbiological, pathological and histological findings in four Danish pig herds affected by a new neonatal diarrhoea syndrome. *BMC Vet. Res.* **9**: 206p.

Kylla, H., Dutta, T.K., Roychoudhury, P. and Subudhi, P. K. 2019. Coinfection of diarrheagenic bacterial and viral pathogens in piglets of Northeast region of India. *Vet. World*. **12**: 224-230.

Majekodunmi, M.M., Omotosho, O.O., Emikpe, B.O. and Olufemi, B.E. 2013. The incidence of gastric lesions in slaughtered pigs in Ibadan, Nigeria. *Sokoto. J. Vet. Sci*. **11**: 22-27.

Mendes, E.N., Queiroz, D.M., Rocha, G.A., Nogueira, A.M., Carvalho, A.C., Lage, A.P. and Barbosa, A.J. 1991. Histopathological study of porcine gastric mucosa with and without a spiral bacterium ("*Gastrospirillum suis*"). *J. Med. Microbiol*. **35**: 345–348.

Miller, M.A. and Zachary, J.F. 2017. Mechanisms and morphology of cellular injury, adaptation and death. In: Zachary, J.F. (ed.), *Pathologic Basis of Veterinary Disease*. (7th Ed.). Elsevier, Missouri, pp. 2-43.

Prasad, C., George A.J., Nair, N.D., Krithiga, K. and Mani, B.K. 2018. Pathogenesis study of porcine salmonellosis in mouse model. *J. Vet. Anim. Sci.* **49**: 44 - 49.

Omotosho, O.O., Emikpe, B.O., Lasisi, O.T. and Jarikre, T.A. 2016. Prevalence, distribution and pattern of gastric lesions in slaughtered pigs in south-western Nigeria. *Onderstepoort. J. Vet. Res.* **83**: 1063-1067.

Robbins, R.C., Almond, G. and Byers, E. 2014. Swine diseases and disorders. In: Alfen, N.K.V. (ed.). *Encyclopaedia of Agriculture and Food Systems.* (2nd Ed.). Academic Press, Elsevier. pp. 261-276.

Rovira, A., Balasch, M., Segales, J., Garcia, L., Plana-Duran, J., Rosell, C., Ellerbrok, H., Mankertz, A. and Domingo, M. 2002. Experimental inoculation of conventional pigs with porcine reproductive and respiratory syndrome virus and porcine circovirus 2. *J. Virol.* **76**: 3232-3239.

Si, G., Niu, J., Zhou, X., Xie, Y., Chen, Z., Li, G., Chen, R. and He, D. 2021. Use of dual priming oligonucleotide system-based multiplex RT-PCR assay to detect five diarrhoea viruses in pig herds in South China. *AMB Express*. **11**: 1-10.

Sinha, R., Sahoo, N.R., Shrivastava, K., Kumar, P., Qureshi, S., Kumar, U., Kumar, A., Kumar, G.V.P.P.S.R. and Bhushan, B. 2019. Resistance to ETEC F4/F18–mediated piglet diarrhoea: opening the gene black box. *Trop. Anim. Health Prod.* **51**: 1307-1320.

Sithara, M.S., George, A.J., Vijayan, N., Manomohan, C.B., Abraham, M.J. and Mani, B.K. 2016. Pathology of bacterial gastro-enteritis in piglets. *J. Vet. Anim. Sci*. **47**: 66-69.

Sudo, K., Yamada, M., Ochiai, M., Iwamoto, S., Sakaki, H., Kushida, C., Yamashita, M., Yamamoto, A., Nagasaka, T., Oidemizu, M., Saito, A. and Yamamoto, K. 2021. The formation process of button ulcers in pigs experimentally infected with a subgenotype 2.1 isolate of classical swine fever virus. *J. Vet. Med. Sci.* **83**: 990-993.

Suvarna, S.K., Layton, C. and Bancroft, J.D. 2013. *Bancroft's theory and practice of histological techniques* (7th Ed.). Churchill Livingstone Elsevier, Philadelphia. 654p.

Yaeger, M.J., Kinyon, J.M. and Songer, J.G. 2007. A prospective, case control study evaluating the association between Clostridium difficile toxins in the colon of neonatal swine and gross and microscopic lesions. *J. Vet. Diagn. Invest*. **19**: 2-59. ■