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Review

Vaccination against enteric rota and coronaviruses in cattle and pigs: enhancement of lactogenic immunity

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Passive immunity against enteric viral infections is dependent upon the continual presence in the gut lumen of a protective level of specific antibodies. This article examines methods currently used to enhance the titre and duration of specific antibody in the mammary secretions of cows and pigs, with particular reference to rotavirus and coronavirus infections. In addition, some of the potential problems to be found in attempting to produce vaccines against these viral infections are outlined.

Keywords: Viruses; rotavirus; coronavirus; cattle; pigs lactogenic immunity

Introduction

Neonatal diarrhoea is a complex disease associated with a number of infectious agents occurring either singly or in combination[1-3]. In domestic animals economic losses are suffered, as a result of mortality (ranging between 0 and 80%), and also veterinary costs and decreased productivity of the survivors. The viral agents most commonly associated with this syndrome are rotavirus and coronavirus, both of which have been found to be primary pathogens in calves[4,5] and piglets[6-8]. These viruses are most frequently isolated during the period from birth to weaning, and animals of this age have been the most intensively studied because of the frequency and severity of these infections. Animals of all ages are, however, susceptible, with subclinical infections apparently common in both adult cows and pigs[9,10]. In neonatal calves the incidence of rotavirus and coronavirus associated diarrhoea is similar varying between 15 and 76%[3,11-13]. The situation in neonatal piglets is less clear, rotavirus infections are apparently common[6,14-16], whilst transmissible gastroenteritis virus (TGEV), the prototype enteric coronavirus in swine, is an example of a seasonal cold-weather disease, probably related to both the thermal sensitivity of the virus[17] and the effect of cold-stress on converting subclinical to clinical infections[18].

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Pathogenesis of infection

The pathogenesis of enteric rotavirus and coronavirus diseases of swine and cattle are similar. In contrast to TGEV, however, rotaviruses appear to be confined to the alimentary tract, predominantly the small intestine, although there is some evidence in both lambs and piglets for infection of the large intestine[19,20]. The infections are characterized by diarrhoea and dehydration caused by the functional and anatomical loss of the absorbtive cells of the intestine. The principal site of virus replication has been shown to be the intestinal villus epithelium. The infected cells are lost from the tips of the villi and are replaced with immature crypt cells. Generally, there is a dimunition in the number and size of the villi and a progressive replacement of the epithelium with squamous and cuboidal cells which lack a brush border[3-6,17,20-26]. Such immature cells have been shown to possess reduced levels of disaccharidases[27,28]. The loss of the absorptive cells of the intestine is assumed to result in the observed malabsorption syndrome. This is further exacerbated by the decreased ability to utilize dietary lactose, resulting in its accumulation in the large intestine, thereby preventing further absorption of water by exerting an osmotic effect.

Passive immunity in pigs and cows

As a result of the severity of these enteric viral infections during the first few weeks of life, passively acquired antibody is the major source of protection. In calves and pigs there is no selective transfer of immunoglobulins from the maternal to the foetal circulation during the last third of the pregnancy. Instead, during the period immediately following birth, maternal immunoglobulin is acquired from the colostrum of the dam[29,30]. Absorption of colostral immunoglobulins by the intestinal epithelial cells is a non-selective process[31-34] lasting 24-48 h[33,35]. Factors present in colostrum may influence the absorption of immunoglobulins[36,37] or help prevent their proteolytic degredation[38]. In addition to immunoglobulins, colostrum and milk have recently been shown to contain functional, immunocompetent cells including macrophages and T and B lymphocytes[39,40]. In contrast to colostral absorption, highly specific mechanisms operate in the colostrum-forming mammary glands of cattle and pigs causing large amounts of IgG (relative to IgA and

IgM) to concentrate in the colostrum[41–44]. IgG passively acquired by the neonate from colostrum persists in the serum for several weeks protecting against systemic infection. In TGEV infection of pigs[45] and rotavirus infection of calves[46] circulating antibody has been found to be of little value. Resistance to these infections appears to be mediated instead by local immunity at the epithelial surface of the intestine.

In cattle the selective transfer of IgG_1 from serum to milk continues throughout lactation, although at a reduced level when compared with colostrum formation. The concentration of all three classes of immunoglobulin is significantly reduced (30 to 60-fold) in milk and in consequence IgG_1 remains the primary immunoglobulin in bovine milk[47]. In contrast, in pigs the concentration of IgG_1 decreases about 30-fold during the first week of lactation, whilst that of secretory IgA declines only about three-fold, leaving it to become the predominant class of immunoglobulin in swine milk[41,48]. Most adult cattle are seropositive for both rotavirus[49,50] and coronavirus[51] antibodies. There is a dramatic decline in these colostral antibody titres during the transition to milk[46,49,51–55], reflecting this reduction in concentration of immunoglobulins. This partially explains the high incidence of rotavirus and coronavirus infections in calves older than five days, as the titres of passively derived protective antibody decline.

Antigenic variation and cross-protection

Despite the presence of one or more common antigens it has been demonstrated that rotaviruses isolated from different species can differ antigenically from each other[56–59]. More recently it has been shown that different serotypes exist within isolates obtained from single species. The existence of at least two different serotypes of porcine rotavirus[60] and at least three distinct bovine serotypes[61] have been described. Bridger *et al.* have suggested the occurrence of intermediate bovine rotavirus types[62], although more work is essential to clarify this situation.

Some recent isolates possessing the distinctive morphology of rotaviruses have been found to lack the common group antigen. To date, these atypical rotaviruses have been isolated from humans, birds, calves, lambs and pigs[63–68]. In pigs, preliminary results using two previously characterized atypical isolates[69,70] have indicated that these are distinct and do not share a common group antigen[62,71]. These observations have been extended by Snodgrass *et al.*[68] who suggest the occurrence of at least four distinct groups of rotaviruses based on their group antigen.

The significance of the serotypic differences observed between rotaviruses *in vitro* still needs to be fully assessed *in vivo*. Orbiviruses (also members of the Reoviridae) possess many serotypes and require the use of multivalent vaccines[72].

Many of the cross-protection studies carried out using different rotavirus serotypes are contradictory and the data inconclusive. For example, *in utero* vaccination of calves with a bovine rotavirus was found to protect against diarrhoea caused by challenge with human rotavirus serotype 2, although challenge virus was still shed[73]. In contrast, one out of three calves was protected against a bovine rotavirus challenge after vaccination with a human serotype 2 or an equine rotavirus[74] and this animal shed no detectable virus. Furthermore, piglets vaccinated with human rotavirus and challenged with porcine rotavirus were protected against the clinical disease but shed virus[75]. Using a more defined challenge system, evidence has been obtained indicating that rotavirus isolates from different animal species and of different serotypes show poor cross-protective properties *in vivo*[76]. This observation has been confirmed and extended by studies in gnotobiotic calves and piglets showing that cross-protection only occurred between rotaviruses of the same serotype, and that even a minor serotype difference could be sufficient to affect cross-protection[60,61]. Further evidence for a lack of cross-protection between rotavirus serotypes can be obtained from studies of sequential infections, where subsequent rotavirus infections were found to be associated with different serotypes[77].

The situation with coronaviruses is simpler. To date, the coronaviruses isolated from mammals and birds have been grouped into four antigenic classes, where little or no cross-reactivity can be demonstrated between classes[78]. TGEV is antigenically distinct from bovine enteric coronavirus[79] as well as from another as yet unclassified coronavirus causing diarrhoea in pigs (CV777)[80].

Vaccination against rotavirus and coronavirus infection in cattle

Two approaches have been used in an attempt to provide calves with protection against rotavirus and coronavirus infections. The first approach involves oral vaccination with live attenuated virus in order to stimulate active immunity in the calf (Scourvax II, Norden Laboratories). The incidence of diarrhoea in neonatal calves orally vaccinated with attenuated rotavirus was found to be reduced[81–83], but the vaccine was not proven to be effective in blind field trials[84–86]. There are a number of limitations associated with this approach. These include the potential of the vaccine to regain virulence; a high incidence of seropositive adult animals, leading to the possibility of interference with vaccine virus replication by maternally derived (milk) antibodies; and the relative immaturity of the neonate's immune system. The second approach utilizes passive protection produced through lactogenic immunity, stimulated by maternal vaccination. Attempts to vaccinate dams using an attenuated live vaccine (Calf Guard, Norden Laboratories) have failed to significantly enhance milk antibody titres[53,87] (*Table 1*), whilst in-

Table 1 Enhancement of rotavirus neutralizing antibody titres in whey, following intramuscular vaccination of heifers and cows with rotavirus preparations, 56 and 28 days before calving. (Crouch, C.F. and Acres, S.D. unpublished data obtained at the Veterinary Infectious Disease Organization)

Vaccine			Whey antibody titre (VN) (days post partum)		
Immunogen	Titre (ELISA units)	Adjuvant[a]	0	10	20
Formaldehyde inactivated rotavirus	800	FICA	13004	203	51
Formaldehyde inactivated rotavirus	200	FICA	32768	102	40
Formaldehyde inactivated rotavirus	800	$AlPO_4$	13004	32	21
Formaldehyde inactivated rotavirus	200	$AlPO_4$	4598	28	7
None	—	—	813	9	5
Calf Guard	—	—	4096	13	10

[a]FICA, Freund's incomplete adjuvant

Table 2 Enhancement of rotavirus neutralizing antibody in cows following different vaccination protocols

Dose per cow	No. of doses	Route[a]	Adjuvant[b]	Immunogen[c]	Colostral antibody titre	Ref.
$10^{8.1}$	2	i.m.	FICA	Formaldehyde inactivated rotavirus	20452[d]	89
—	2	i.m.	FICA	Tissue culture fluid	100[d]	89
$10^{8.8}$	2	nr	$Al(OH)_2$ + oil	Formaldehyde inactivated rotavirus	1580[d]	90
—	2	nr	$Al(OH)_2$ + oil	None	320[d]	90
nr	2	s.c.	nr	Inactivated rotavirus	30[e]	92
—	—	—	—	None	3[e]	92
$10^{6.0}$	2	i.m.	$Al(OH)_2$	Formaldehyde inactivated rotavirus	3236[d]	88
$10^{5.5}$	1	i.m.	Oil	Formaldehyde inactivated rotavirus	11481[d]	88
$10^{4.9}$	1	i.m.	Oil	Formaldehyde inactivated rotavirus	9120[d]	88
—	—	—	—	None	645[d]	88
nr	1	s.c.	Oil	BPL inactivated rotavirus	1995[f]	91
nr	2	s.c.	Oil	BPL inactivated rotavirus	6300[f]	91
—	—	—	—	None	795[f]	91
$10^{8.4}$	2	i.m.	FICA	BEI inactivated rotavirus	24401[d]	53
$10^{8.4}$	2	i.m.	FICA	BPL inactivated rotavirus	2374[d]	53
—	—	—	—	None	2865[d]	53

nr, not reported: [a]i.m., intramuscular; s.c., subcutaneous. [b]FICA, Freund's incomplete adjuvant. [c]BPL, β-propriolactone; BEI, binary ethylenimine. [d]Titres determined by virus neutralization. [e]Titres determined by complement fixation test. [f]Titres determined by haemagglutination inhibition test

activated, adjuvanted rotavirus preparations have been found to enhance levels of specific antibody in colostrum and milk (*Tables 1* and *2*).

A number of parameters need to be considered in attempting to optimize the enhancement of antibody production in mammary secretions.

Dose and form of vaccine. In considering inactivated vaccines, it is to be expected that relatively large amounts are necessary to achieve a satisfactory response. Further, the process of inactivation may decrease the immunogenicity of some viral polypeptides. *Table 2* shows that no significant differences in milk antibody titres were obtained following vaccination of cows with rotavirus preparations containing either 200 or 800 ELISA units (after inactivation) emulsified in Freund's incomplete adjuvant. In contrast, if the same preparations were used, but adjuvanted with aluminium phosphate, the higher dose resulted in a greater antibody response. A similar result using an oil adjuvanted rotavirus vaccine has been previously reported[88].

Formaldehyde inactivated rotavirus vaccines have been used to successfully enhance milk antibody titres as compared with controls[88-90]. Other workers have reported increased antibody responses using β-propriolactone as the inactivating agent[91], although Saif *et al.* found that antibody titres in mammary secretions were at least tenfold greater from cows vaccinated with binary ethylenimine inactivated rotavirus compared with those vaccinated with β-propriolactone inactivated rotavirus[53].

Adjuvant. Snodgrass *et al.*[88] found that oil-based adjuvants were more effective than alhydrogel for the enhancement of rotavirus antibody titres in mammary secretions. This concurs with the data presented in *Table 1*. Most workers have demonstrated a satisfactory immune response following vaccination using oil-based adjuvants, generally Freunds incomplete adjuvant (*Table 2*).

Route and timing of vaccination. To some extent the route and timing of vaccination are dependent upon the type of cattle being farmed. Thus the intramammary route used successfully by Saif *et al.*[53], whilst applicable to dairy cattle, may not be practical in beef cows. Similarly, from an administrative viewpoint a single vaccination would be preferable to a regime utilizing several doses. The majority of studies have reported a significant increase in rotavirus antibody titres in mammary secretions using either subcutaneous or intramuscular injection of oil-adjuvanted vaccines. All such vaccines have also proved to be effective when administered as either single or double doses injected prior to or at parturition[53,88-92] (*Table 1*).

The efficacy of immune milk as a mechanism for providing passive immunity against rotavirus challenge has been examined by a number of workers (*Table 3*). The

Table 3 Passive protection against rotavirus challenge of calves fed immune milk

Lactogenic Antibody					Protection				
					Virus shedding		Diarrhoea		
Origin[a]	Amount	Method of feeding[b]	Challenge[c]	No. of calves	%	onset[d]	%	onset[d]	Ref.
Vacc	—	Suckling	Exp	7	100	5.2	71	4.8	89
Cont	—	Suckling	Exp	9	100	2.0	100	3.0	89
Vacc	—	Suckling	Field	402	—	nr	93	5.2[e]	92
Cont	—	Suckling	Field	174	—	nr	90	4.9[e]	92
Vacc	2l 2×	Supp	Exp	7	—	nr	88	5.2	92
Cont	2l 2×	Supp	Exp	5	—	nr	100	6.0	92
Vacc	10%	Supp	Field	10	40	nr	40	6.0[f]	94
Normal	10%	Supp	Field	11	36	nr	73	4.1[f]	94
Cont	—	—	Field	21	76	nr	76	3.9[f]	94
Vacc	—	Suckling	Field	77	10	nr	8	nr	91
Cont	—	Suckling	Field	64	29	nr	27	nr	91
Vacc	nr	Supp	Exp	3	nr	6.0	0	—	91
Cont	nr	Supp	Exp	2	100	1.0	0	—	91
Vacc	1%	Supp	Exp	8	0	—	25	3.0	93
Vacc	0.1%	Supp	Exp	6	66	3.8	83	3.7	93
Vacc[g]	1%	Supp	Exp	6	100	1.8	100	1.6	93
Normal	1%	Supp	Exp	6	100	1.6	100	1.7	93
Cont	—	—	Exp	8	100	1.1	100	1.1	93

[a]Lactogenic antibody originated from either vaccinated (vacc), control (cont) or normal cows (normal). [b]Calves were either suckled naturally (suckled) or fed a supplemented diet containing antibody (supp). [c]Calves were either challenged experimentally (exp) or naturally exposed under field conditions (field). [d]Days after challenge. [e]Days after birth. [f]Days after start of experiment. [g]Cows vaccinated with commercial vaccine. nr, not reported

Table 4 Efficacy of different vaccination protocols for the stimulation of lactogenic immunity for the passive protection of newborn pigs against challenge by TGEV

Immunogen	Route[a]	Challenge	Protection	Class of major immunoglobulin in milk	Ref.
Live-attenuated TGEV	i.m.	Experimental	Moderate	IgG	97
Live-attenuated TGEV	i.mm.	Experimental	Good	IgG	97
Live-attenuated TGEV	i.m.	Experimental	Poor	IgG	111
Live-attenuated TGEV	Oral	Experimental	Poor	IgG	110
Live-attenuated TGEV	Oral	Experimental	Poor	IgG	100
Live FIPV	Oral	Experimental	Poor	IgG	109
Live-attenuated TGEV	Oral	Experimental	Poor	IgA	109

[a]i.m., intramuscular; i.mm., intramammary

results, however, are difficult to compare, due to variations in the feeding regime used for the immune milk and also the challenge systems used. The amount and the timing of the feeding of lactogenic antibody and the dose, virulence and serotype of the virus challenge strain used will all affect the apparent susceptibility of the calf to infection. Further, in situations where a field challenge has been used, failure of protection may be due to infection by rotavirus serotypes other than those used in the vaccine, or possibly by other agents capable of causing diarrhoea. Generally, these investigators reported either a reduced incidence of rotavirus shedding or diarrhoea or both. In only one study[92] did the feeding of lactogenic antibody fail to significantly affect the incidence or onset of diarrhoea. The majority of animals receiving passive immunity appear to be capable of developing active immunity during this period[93–95], consequently vaccination should lead to elimination of clinical disease rather than a delay in its onset. Investigation of the immunoglobulin isotypes associated with this protective antibody induced by vaccination in bovine milk and colostrum suggests that IgG_1 plays the major role[95,96]. These observations are in agreement with those discussed earlier concerning passive immunity in the bovine.

Vaccination against rotavirus and coronavirus infection in pigs

In contrast to the bovine system, evidence suggests that milk or colostral immunoglobulin of the IgA isotype is more effective than those of the IgG isotypes at protecting piglets against infection by TGEV[97–100]. High persisting levels of IgG may, however, provide some degree of

protection against virus challenge[97]. As a result of these observations, most studies have examined methods for optimising the stimulation of secretory IgA antibodies in milk. The origin of TGEV-specific IgA found in mammary secretions remains somewhat obscure, although there is a good correlation with the presence of an infection in the intestinal tract[97,99,100,101]. Secretory IgA in porcine milk is almost certainly locally produced in the mammary gland[102-104]. In order to explain this phenomenon, it has been suggested that specifically-sensitized IgA-secreting lymphocytes may migrate to the mammary gland following initial sensitization in the intestine[97-100]. Such an inter-relationship between the intestinal and the mammary immune systems has also been proposed in rabbits[105] and humans[106]. Direct evidence for such migration, under the influence of pregnancy-associated hormones, has been obtained in mice[107].

A summary of various investigations into the antibody response and efficacy of lactogenic immunity following different vaccination protocols is given in *Table 4*. Reduced immunogenicity in pigs of cell culture attenuated TGEV has been described[108]. Oral vaccination with a live, attenuated TGE vaccine, whilst producing neutralizing antibody, did not stimulate good lactogenic immunity in suckling pigs[100,109,110]. Intramuscular vaccination of sows with live, attenuated TGEV leads to the enhancement of specific IgG levels in colostrum and milk[97,111]. Higher titres of TGEV-specific IgG have been achieved using intramammary injection, with an associated increase in the protection provided to suckling pigs[97]. These results are supported by the observations of other workers[112-115]. Feline infectious peritonitis virus (FIPV) is a member of the same antigenic class as TGEV and consequently the two viruses are serologically related. Good levels of cross-protection, associated with high titres of TGEV-specific neutralizing antibody have been reported in sows vaccinated orally with FIPV[116]. In contrast, the results of a more recent study have shown that whilst TGEV neutralizing antibodies of the IgG subclass are stimulated in milk and colostrum, the survival rate for suckling pigs was low[117].

It may be possible to boost the level of IgA in mammary secretions. Preliminary investigations have revealed that specific secretory IgA levels in milk can be enhanced by the parenteral injection, at parturition, of TGEV or rotavirus into naturally infected (orally primed) animals[118,119]. A similar approach also combining oral with parenteral antigen administration has been proposed as a means of providing lactogenic immunity against colibacillosis in pigs[120].

Future considerations

IgG can be induced readily in the mammary secretions of cattle, by intramuscular or subcutaneous injection of adjuvanted immunogen. In pigs however, whilst live, virulent virus is capable of inducing high levels in IgA in milk, it is apparent that the ideal candidate vaccine virus must be sufficiently attenuated to produce only mild or no disease in neonatal pigs, whilst retaining sufficient virulence to infect the intestinal tract of adult swine. More work is essential in the possible use of inactivated vaccines for the boosting of existing IgA levels in mammary secretions. These may require prior natural infection of the sow, the incidence of which will vary between herds, with an associated affect upon the efficacy of such a vaccine.

Further investigation into the variety of strains and serotypes of rotaviruses is of obvious importance, as is the response to vaccination of cattle and swine by rotaviruses or coronaviruses. Current data suggests that cross-protection between rotavirus serotypes is limited, although there is little information concerning the specificities of the antibodies induced by vaccination of previously infected animals. Such animals naturally exposed to a variety of serotypes may produce a heterogeneous antibody response, capable of reacting with a broad spectrum of rotavirus serotypes.

It is apparent that the enhancement of lactogenic immunity through the vaccination of the dam provides a suitable mechanism by which neonatal pigs and calves can be protected against rotavirus and coronavirus infections. The production of truly effective vaccines, however, awaits further work in some of the areas outlined above.

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References

1 Acres, S.D., Laing, C.J., Saunders, J.R. and Radostits, O.M. Acute undifferentiated neonatal diarrhoea in beef calves 1. Occurrence and distribution of infectious agents. *Can. J. Comp. Med.* 1975, **39**, 116
2 Moon, H.W., McClurkin, A.W., Isaacson, R.E., Pohlenz, J., Skartedf, S.M., Gillette, K.G. and Baetz, A.L. Pathogenic relationships of rotavirus, *Escherichia coli*, and other agents in mixed infections in calves. *J. Am. Vet. Med. Assoc.* 1978, **173**, 577
3 Morin, M., Lariviere, S. and Lallier, R. Pathological and microbiological observations made on spontaneous cases of acute neonatal calf diarrhoea. *Can. J. Comp. Med.* 1976, **40**, 228
4 Mebus, C.A., Stair, E.L., Underdahl, N.R. and Twiehaus M.J. Pathology of neonatal calf diarrhoea induced by a reo-like virus. *Vet. Path.* 1971, **8**, 490
5 Mebus, C.A., Stair, E.L., Rhodes, M.B. and Twiehaus, M.J. Pathology of neonatal calf diarrhoea induced by a corona-like agent. *Vet. Path.* 1973, **10**, 45
6 Woode, G.N., Bridger, J.C., Hall, G.A., Jones, J.M. and Jackson, G. The isolation of reovirus-like agents (rotaviruses) from acute gastoenteritis of piglets. *J. Med. Microbiol.* 1976, **9**, 203
7 Doyle, L.P. and Hutchings, L.M. A transmissible gastro-enteritis in pigs. *J. Am. Vet. Med. Assoc.* 1946, **108**, 257
8 Lecce, J.C., King, M.W. and Mock, R. Reovirus-like agent associated with fatal diarrhoea in pigs. *Infect. Immun.* 1976, **14**, 816
9 Benfield, D.A., Stortz, I., Moore, R. and McAdaragh, J.P. Shedding of rotavirus in feces of sows before and after farrowing. *J. Clin. Microbiol.* 1982, **16**, 186
10 Crouch, C.F. and Acres, S.D. Prevalence of rotavirus and coronavirus antigens in the faeces of normal cows. *Can. J. Comp. Med.* 1984, **48**, 340
11 Crouch, C.F., Raybould, T.J.G. and Acres, S.D. Monoclonal antibody capture-enzyme-linked immunosorbent assay for the detection of bovine enteric coronavirus. *J. Clin. Microbiol.* 1984, **19**, 388
12 Langpap, T.J., Bergeland, M.E. and Reed, D.E. Coronaviral enteritis of young calves: Virologic and pathologic findings in naturally occurring infections. *Am. J. Vet. Res.* 1979, **40**, 1476
13 Marsolais, G., Assaf, R., Montpetit, C. and Marois, P. Diagnosis of viral agents associated with neonatal calf diarrhoea. *Can. J. Comp. Med.* 1978, **42**, 168
14 Bohl, E.H. Rotaviral diarrhoea in pigs: Brief review *J. Am. Vet. Med. Assoc.* 1979, **174**, 613
15 Corthier, G., Vautherot, J.F. and Vannier, P. Serologic detection of rotavirus in swine from the region of Brittany. *Ann. Rech. Vet.* 1979, **10**, 65

16 Lecce, J.G., King, M.W. and Dorsey, W.E. Rearing regimen producing piglet diarrhoea (rotavirus) and its relevance to acute infantile diarrhoea. *Science* 1978, **199**, 776

17 Woode, G.N. Transmissible gastroenteritis of swine. *Vet. Bull.* 1969, **39**, 239

18 Shimizu, M. and Shimizu, Y. Effects of ambient temperatures on clinical and immune responses of pigs infected with transmissible gastroenteritis virus. *Vet. Microbiol.* 1979, **4**, 109

19 Snodgrass, D.R., Angus, K.W. and Craig, E.W. Rotavirus infection in lambs: Pathogenesis and pathology. *Arch. Virol.* 1977, **55**, 263

20 Theil, K.W., Bohl, E.H., Cross, R.F., Kohler, E.M. and Agnes, A.G. Pathogenesis of porcine rotaviral infection in experimentally inoculated gnotobiotic piglets. *Am. J. Vet. Res.* 1978, **39**, 213

21 Pensaert, M., Haelterman, E.O. and Burnstein, T. Virus-intestinal cell interactions. 1) Immunofluorescence, histopathology and virus production in the small intestine through the course of infection. *Arch. Ges. Virusforsch.* 1970, **31**, 321

22 Mebus, C.A., Newman, L.E. and Stair, E.L. Scanning electron, light and immunofluorescent microscopy of intestine of gnotobiotic calf infected with calf diarrhoeal coronavirus. *Am. J. Vet. Res.* 1975, **36**, 1719

23 Mebus, C.A., Underdahl, N.R., Rhodes, M.B. and Twiehaus, M.J. Calf diarrhoea (scours) reproduced with a virus from a field outbreak. *Univ. Nebraska Res. Bull.* 1969, **233**, 1

24 Pearson, G.R., McNulty, M.S. and Logan, E.E. Pathological changes in the small intestine of neonatal calves naturally infected with reo-like virus (rotavirus). *Vet. Rec.* 1978, **102**, 454

25 Pearson, G.R. and McNulty, M.S. Pathological changes in the small intestine of neonatal pigs infected with a pig reovirus-like agent (rotavirus). *J. Comp. Pathol.* 1977, **87**, 363

26 Crouch, C.F. and Woode, G.N. Serial studies of virus multiplication and intestinal damage in gnotobiotic piglets infected with rotavirus. *J. Med. Microbiol.* 1978, **11**, 325

27 Bishop, R.F., Davidson, G.P., Holmes, I.H. and Ruck, B.J. Virus particles in epithelial cells of duodenal mucosa from children with acute nonbacterial gastroenteritis. *Lancet* 1973, 1281

28 Halpin, C.G. and Caple, I.W. Changes in intestinal structure and function of neonatal calves infected with reovirus-like agent and *Escherichia coli*. *Austr. Vet. J.* 1976, **52**, 438

29 Brambell, F.W.R. The transmission of passive immunity from mother to young. In: *Frontiers of biology* (Eds Neuberger, A. and Tatum, E.L.) North Holland Publishing Co. Amsterdam, 1970, pp. 301

30 Simpson-Morgan, M.W. and Smeaton, T.C. The transfer of antibodies by neonates and adults. *Adv. Vet. Sci. Comp. Med.* 1972, **16**, 355

31 Brandon, M.R. and Lascelles, A.K. Relative efficiency of absorption of IgG_1, IgG_2, IgA and IgM in the newborn calf. *Austr. J. Exp. Biol. Med. Sci.* 1971, **49**, 629

32 Balfour, W.E. and Comline, R.S. The specificity of the intestinal absorption of large molecules by the newborn calf. *J. Physiol.* 1959, **148**, 77

33 Lecce, J.G. and Morgan, D.O. Effect of dietary regimen on cessation of intestinal absorption of large molecules (closure) in the neonatal pig and lamb. *J. Nutrition* 1962, **78**, 263

34 Porter, P. Porcine colostral IgA and IgM antibodies to *Escherichia coli* and their intestinal absorption by the neonatal piglet. *Immunology* 1969, **17**, 617

35 Pierce, A.E. Electrophoretic and immunological studies on sera from calves from birth to weaning. *J. Hygiene* 1955, **53**, 247

36 Balfour, W.E. and Comline, R.S. Acceleration of the absorption of unchanged globulin in the newborn calf by factors in colostrum. *J. Physiol.* 1962, **160**, 234

37 Johnson, N.E. and Oxender, W.D. Effect of altered serum glucocorticoid concentrations on the ability of the newborn calf to absorb colostral immunoglobulin. *Am. J. Vet. Res.* 1979, **40**, 32

38 Pineiro, A., Ortega, F. and Uriel, J. Trypsin inhibitor from bovine colostrum. *Biochim. Biophys. Acta* 1975, **379**, 201

39 Parmerly, M.J. and Beer, A.E. Colostral cell-mediated immunity and the concept of a common secretory immune system. *J. Dairy Sci.* 1977, **60**, 655

40 Smith, J.W. and Schultz, R.D. Mitogen and antigen response in milk lymphocytes. *Cell. Immunol.* 1977, **29**, 165

41 Bourne, F.J. Symposium on the young farm animal: The immunoglobulin system of the suckling pig. *Proc. Nutr. Soc.* 1973, **32**, 205

42 Lascelles, A.K. and McDowell, G.H. Localised humoral immunity with particular reference to ruminants. *Transplant. Rev.* 1974, **19**, 170

43 Pierce, A.E. and Feinstein, A. Biophysical and immunological studies on bovine immunoglobulins with evidence for selective transport within the mammary gland from maternal plasma to colostrum. *Immunology* 1965, **8**, 106

44 Porter, P., Noakes, D.E. and Allen, W.D. Secretory IgA and antibodies to *Escherichia coli* in porcine milk and their significance in the alimentary tract of the young pig. *Immunology 1970*, **18**, 245

45 Haelterman, E.O. and Hooper, B.E. Transmissible gastroenteritis of swine as a model for the study of enteric disease. *Gastroenterology* 1967, **153**, 109

46 Woode, G.N., Jones, J. and Bridger, J.C. Levels of colostral antibodies against neonatal calf diarrhoea virus. *Vet. Rec.* 1975, **97**, 148

47 Butler, J.E. Synthesis and distribution of immunoglobulins. *J. Am. Vet. Med. Assoc.* 1973, **163**, 795

48 Porter, P. and Allen, W.D. Classes of immunoglobulins related to immunity in the pig: A review. *J. Am. Vet. Med. Assoc.* 1972, **160**, 511

49 Acres, S.D. and Babiuk, L.A. Studies on rotaviral antibody in bovine serum and lacteal secretions using radioimmunoassay. *J. Am. Vet. Med. Assoc.* 1978, **173**, 555

50 Flewett, T.H. and Woode, G.N. The rotaviruses. *Arch. Virol.* 1978, **57**, 1

51 Rodak, L., Babiuk, L.A. and Acres, S.D. Radioimmunological (RIA) and enzymimmunological (ELISA) detection of coronavirus antibodies in bovine serum and lacteal secretions. *J. Clin. Microbiol.* 1982, **16**, 34

52 Eichorn, W., Bachmann, P.A., Baljer, G., Plank, P. and Schneider, P. Vaccination of cows with a combined rotavirus/enterotoxigenic *Escherichia coli* K99 vaccine to protect newborn calves against diarrhoea. *Dev. Biol. Stand.* 1983, **53**, 237

53 Saif, L.J., Smith, K.L., Landmeier, B.J., Bohl, E.H. and Theil, K.W. Immune responses of pregnant cows to bovine rotavirus immunization. *Am. J. Vet. Res.* 1984, **45**, 49

54 Snodgrass, D.R., Fahey, K.J., Wells, P.W., Cambell, I. and Whitehall, A. Passive immunity in calf rotavirus infections. Maternal vaccination increases and prolongs immunoglobulin G_1 antibody secretion in milk. *Infect. Immun.* 1980, **28**, 344

55 Crouch, C.F. and Raybould, T.J.G. Comparison of different antigen preparations as substrates for use in passive haemagglutination and enzyme-linked immunosorbent assays for the detection of antibody against bovine enteric coronavirus. *J. Clin. Microbiol.* 1983, **18**, 146

56 Flewett, T.H., Bryden, A.S., Davies, H., Woode, G.N., Bridger, J.C. and Derrick, J.M. Relation between viruses from acute gastroenteritis of children and newborn calves. *Lancet* 1974, 61

57 Schoub, B.D., Lectsas, G. and Prozesky, O.W. Antigenic relationship between human and simian rotaviruses. *J. Med. Microbiol.* 1976, **10**, 1

58 Thouless, M.E., Bryden, A.S., Flewett, T.H., Woode, G.N., Bridger, J.C. Snodgrass, D.R. and Herring, J.A. Serological relationships between rotaviruses from different species as studied by complement fixation and neutralization. *Arch. Virol.* 1977, **53**, 287

59 Woode, G.N., Bridger, J.C., Jones, J.M., Flewett, T.H., Bryden, A.S., Davies, H.A. and White, G.B.B. Morphological and antigenic relationships between viruses (rotaviruses) from acute gastroenteritis of children, calves, piglets, mice, and foals. *Infect. Immun.* 1976, **14**, 804

60 Bohl, E.H., Theil, K.W. and Saif, L.J. Isolation and serotyping of porcine rotaviruses and antigenic comparison with other rotaviruses. *J. Clin. Microbiol.* 1984, **19**, 105

61 Woode, G.N., Kelso, N.E., Simpson, T.F., Gaul, S.K., Evans, L.E. and Babiuk, L. Antigenic relationships among some bovine rotaviruses: Serum neutralization and cross-protection in gnotobiotic calves. *J. Clin. Microbiol.* 1983, **18**, 358

62 Bridger, J.C., Brown, J.F., Arnold, P.S., Pocock, D.H. and

McCrae, M.A. Antigenic variation in porcine and bovine rotaviruses. In: *Proceedings Fourth International Symposium on Neonatal Diarrhoea* VIDO, 124, Veterinary Road, Saskatoon, Saskatchewan, Canada. 1983, pp. 48–55

63 Rodger, S.M., Bishop, R.F. and Holmes, I.H. Detection of a rotavirus-like agent associated with diarrhoea in an infant. *J. Clin. Microbiol.* 1982, **16**, 724

64 Nicholas, J.C., Cohen, J., Fortier, B., Lourento, M.H. and Bricout, F. Isolation of human pararotavirus. *Virology* 1984, **124**, 181

65 McNulty, M.S., Allan, G.M., Todd, D., McFerran, J.B., McKillop, E.R., Collins, D.S. and McCracken, R.M. Isolation of rotaviruses from turkeys and chickens: Demonstration of distinct serotypes and RNA electrophoresis. *Avian Pathol.* 1980, **9**, 363

66 Bridger, J.C. Detection by electron microscopy of calici-viruses, astroviruses and rotavirus-like particles in the faeces of piglets with diarrhoea. *Vet. Rec.* 1980, **107**, 532

67 Saif, L.J., Bohl, E.H., Theil, K.W., Cross, R.F. and House, J.A. Rotavirus-like, calicivirus-like and 23 nm virus-like particles associated with diarrhoea in young pigs. *J. Clin. Microbiol.* 1980, **12**, 105

68 Snodgrass, D.R., Herring, J.A., Campbell, I., Inglis, J.M. and Hargreaves, F.D. Comparison of atypical rotaviruses from calves, piglets lambs and man. *J. Gen. Virol.* 1984, **65**, 909

69 Bridger, J.C., Clarke, I.N. and McCrae, M.A. Characterisation of an antigenically distinct porcine rotavirus. *Infect. Immun.* 1982, **35**, 1058

70 Bohl, E.H., Saif, L.J., Theil, K.W., Agnes, A.G. and Cross, R.F. Porcine pararotavirus: detection, differentiation from rota-virus, and pathogenesis in gnotobiotic pigs. *J. Clin. Microbiol.* 1982, **15**, 312

71 Pedley, S., Bridger, J.C., Brown J.F. and McCrae, M.A. Molecular characterisation of rotaviruses with distinct group antigens. *J. Gen. Virol.* 1983, **64**, 2093

72 Andrews, C., Pereira, H.G. and Wildy, P. In: *Viruses of vertebrates*, fourth edition, 1986, Balliere, Tindall, London p 42

73 Wyatt, R.G., Mebus, C.A., Yolken, R.H., Kalica, A.R., James, Jr., H.D., Kapikian, A.Z. and Chanock, R.M. Rotaviral immunity in gnotobiotic calves: Heterologous resistance to human virus induced by bovine virus. *Science* 1979, **203**, 548

74 Woode, G.N., Bew, M.E. and Dennis, M.J. Studies on cross-protection induced in calves by rotaviruses of calves, children and foals. *Vet. Rec.* 1978, **103**, 32

75 Tzipori, S.R., Makin, T.J. and Smith, M.L. The clinical response of gnotobiotic calves, pigs and lambs to inoculation with human, calf, pig and foal rotavirus isolates. *Austr. J. Exp. Biol. Med. Sci.* 1980, **58**, 309

76 Gaul, S.K., Simpson, T.F., Woode, G.N. and Fulton, R.W. Antigenic relationships among some animal rotaviruses: Virus neutralization *in vitro* and cross-protection in piglets. *J. Clin. Microbiol.* 1982, **16**, 495

77 Yolken, R.H., Wyatt, R.G., Zissis, G., Brandt, C.D., Rodriguez, W.J., Kim, H.W. *et al.* Epidemiology of human rotavirus type 1 and type 2 as studied by enzyme-linked immunosorbent assay. *N. Engl. J. Med.* 1978, **299**, 1156

78 Wege, H., Siddell, S. and ter Meulen, V. The biology and pathogenesis of coronaviruses. *Curr. Top. Microbiol. Immunol.* 1982, **99**, 165

79 Brian, D.A., Hogue, B., Lapps, W., Potts, B. and Kapke, P. Comparative structure of coronaviruses. In: *Proceedings Fourth International Symposium on neonatal diarrhoea* VIDO, 124, Veterinary Rd, Saskatoon, Saskatchewan, Canada, 1983, pp. 100–115

80 Pensaert, M.B. and Debouk, P. A new coronavirus-like particle associated with diarrhoea in swine. *Arch. Virol.* 1978, **58**, 243

81 Mebus, C.A., White, R.G., Stair, E.L., Rhodes, M.B. and Twiehaus, M.J. Neonatal calf diarrhoea: Results of a field trial using a reo-like virus vaccine. *Vet. Med. Small Anim. Clin.* 1972, **67**, 173

82 Mebus, C.A., White, R.G., Bass, E.P. and Twiehaus, M.J. Immunity to neonatal calf diarrhoea virus. *J. Am. Vet. Med. Assoc.* 1973, **163**, 880

83 Twiehaus, M.J., Mebus, C.A. and Bass, E.P. Survey of the field efficacy of reoviral calf diarrhoea vaccine. *Vet. Med. Small Anim. Clin.* 1975, **24**, 23

84 Newman, F.S., Myers, L.L., Firchammer, B.D. and Catlin, J.E. Licensing and use of the calf scours vaccine. Part II An analysis of Scourvax-Reo. In: *Proceedings 77th Annual Meeting U.S. Animal Health Association* 1973, pp. 59–64

85 Acres, S.D. and Radostits, O.M. The efficacy of a modified live reo-like virus vaccine and an *Escherichia coli* bacterin for prevention of acute undifferentiated neonatal diarrhoea of beef calves. *Can. Vet. J.* 1976, **17**, 197

86 deLeeuw, P.W., Ellens, D.J., Talman, F.P. and Zimmer, G.N. Rotavirus infections in calves: Efficacy of oral vaccination in endemically infected herds. *Res. Vet. Sci.* 1980, **29**, 142

87 Myers, L.L. and Snodgrass, D.R. Colostral and milk antibody titres in cows vaccinated with a modified live rotavirus-coronavirus vaccine. *J. Am. Vet. Med. Assoc.* 1982, **181**, 486

88 Snodgrass, D.R., Nagy, L.K., Sherwood, D. and Campbell, I. Passive immunity in calf diarrhoea: Vaccination with K99 antigen of enterotoxigenic *Escherichia coli* and rotavirus. *Infect. Immun.* 1982, **37**, 586

89 Snodgrass, D.R., Fahey, K.J., Wells, P.W., Campbell, I. and Whitelaw, A. Passive immunity in calf rotavirus infections: Maternal vaccination increases and prolongs immunoglobulin G_1 antibody secretion in milk. *Infect. Immun.* 1980, **28**, 344

90 Van Opdenbosch, E., Wellemans, G., Strobbe, R., De Brabander, D.L. and Boucque, Ch.V. Evolution des anticorps anti rota dans le lait de vaches traitees en fin de Gestation soit par le vaccine anti-rota complet, soit par l'adjuvant seul. *Comp. Immun. Microbiol. Infect. Dis.* 1981, **4**, 293

91 Dauvergne, M., Brun, A. and Soulebot, J.P. Passive protection of newborn calves against rotavirus by vaccination of their dams. *Dev. Biol. Stand.* 1983, **53**, 245

92 Hartmann, H., Bechtel, D., Otto, P. and Schonhev, W. Die Wirksamkeitsprufung eimer inaktivierten rotavirus-vakzine in einem Milchviehbestand. *Arch. Exper. Veterinaermed.* 1982, **36**, 611

93 Saif, L.J., Redman, D.R., Smith, K.L. and Theil, K.W. Passive immunity to bovine rotavirus in newborn calves fed colostrum supplements from immunized or non-immunized cows. *Infect. Immun.* 1983, **41**, 1118

94 Snodgrass, D.A. and Wells, P.W. Rotavirus infection in lambs: Studies on passive protection. *Arch. Virol.* 1976, **52**, 201

95 Bridger, J.C. and Brown, J.F. Development of passive immunity to porcine rotavirus in pigs protected from disease by bovine colostrum. *Infect. Immun.* 1981, **31**, 906

96 Snodgrass, D.R., Stewart, J., Taylor, J., Krautil, F.L. and Smith, M.L. Diarrhoea in dairy calves reduced by feeding colostrum from cows vaccinated with rotavirus. *Res. Vet. Sci.* 1982, **32**, 70

97 Bohl, E.H., Gupta, P.K., Olquin, F.M.W. and Saif, L.J. Antibody responses in serum, colostrum and milk of swine after infection or vaccination with transmissible gastroenteritis virus. *Infect. Immun.* 1972, **6**, 289

98 Saif, L.J., Bohl, E.H. and Gupta, P.K. Isolation of porcine immunoglobulins and determination of the immunoglobulin classes of transmissible gastroenteritis viral antibodies. *Infect. Immun.* 1972, **6**, 600

99 Bohl, E.H. and Saif, L.J. Passive immunity in transmissible gastroenteritis of swine: Immunoglobulin characterisation of antibodies in milk after inoculating virus by different routes. *Infect. Immun.* 1975, **11**, 23

100 Saif, L.J. and Bohl, E.H. Passive immunity in transmissible gastroenteritis of swine: Immunoglobulin classes of milk after oral/intranasal inoculation of sows with a live low cell culture-passaged virus. *Am. J. Vet. Res.* 1979, **40**, 115

101 Bohl, E.H., Gupta, R.K.P., McCloskey, L.W. and Saif, L.J. Immunology of transmissible gastroenteritis. *J. Am. Vet. Med. Assoc.* 1972, **160**, 543

102 Porter, P. Transfer of immunoglobulins IgG, IgA and IgM to lacteal secretions in the parturient sow and their absorption by the neonatal piglet. *Biochim. Biophys. Acta* 1969, **181**, 381

103 Porter, P., Noakes, D.E. and Allen, W.D. Secretory IgA and antibodies to *Escherichia coli* in porcine colostrum and milk and their significance in the alimentary tract of the young pig. *Immunology* 1970, **18**, 245

104 Bourne, F.J. and Curtis, J. The transfer of immunoglobulins IgG, IgA and IgM from serum to colostrum and milk in the sow. *Immunology* 1973, **24**, 157

105 Montgomery, P.C., Cohn, J. and Lally, E.T. The induction and characteristics of secretory IgA antibodies. In: *The immunoglobulin A system,* (Eds Mestecky, J. and Lawton, A.R.) Plenum Press, 1974, pp. 453–462

106 Goldblum, R.M., Ahlstedt, S., Carlsson, B., Hanson, L.A., Jodal, U., Lidinjanson, G. and Sohl-Akerlund, A. Antibody-forming cells in human colostrum after oral immunization. *Nature* 1975, **257**, 797

107 Weisz-Carrington, P., Roux, M.E., McWilliams, M., Phillips-Quagliata, J.M. and Lamm, M.E. Hormonal induction of the secretory immune system in the mammary gland. *Proc. Natl Acad. Sci. USA* 1978, **75**, 2928

108 Harada, K., Furuuchi, S., Kumagai, T. and Sasahara, J. Pathogenicity, immunogenicity and distribution of transmissible gastroenteritis in pigs. *Nat. Inst. Anim. Hlth Quart.* 1969, **9**, 185

109 Bohl, E.H. Transmissible gastroenteritis In: *Diseases of swine* (Ed. Dunne, H.W.) Iowa State University Press, Ames, Iowa, 1970, pp. 158–176

110 Pensaert, M.B. Immunity in TGE of swine after infection and vaccination. In: *Viral enteritis in humans and animals.* (Eds Bricout, F. and Scherrer, R.) INSERM 1979, Vol. 90, pp. 281–293

111 Bohl, E.H., Frederick, G.T. and Saif, L.J. Passive immunity in transmissible gastroenteritis of swine: Intramuscular injection of pregnant swine with a modified live-virus vaccine. *Am. J. Vet. Res.* 1975, **36**, 267

112 Djurickovic, S. and Thorsen, J. Experimental immunization of sows against transmissible gastroenteritis. *Vet. Rec.* 1970, **87**, 62

113 Thorsen, J. and Djurickovic, S. Experimental immunization of sows with cell-cultured TGE virus. *Can. J. Comp. Med. Vet. Sci.* 1970, **34**, 177

114 Thorsen, J. and Djurickovic, S. Experimental immunization of sows with inactivated transmissible gastroenteritis (TGE) virus. *Can. J. Comp. Med. Vet. Sci.* 1971, **35**, 99

115 Tamoglia, T.W. Present status of products for use against transmissible gastroenteritis. *J. Am. Vet. Med. Assoc.* 1972, **160**, 554

116 Woods, R.D. and Pedersen, N.C. Cross-protection studies between feline infectious peritonitis and porcine transmissible gastroenteritis viruses. *Vet. Microbiol.* 1979, **4**, 11

117 Woods, R.D. Efficacy of vaccination of sows with serologically related coronaviruses for control of transmissible gastroenteritis in nursing pigs. *Am. J. Vet. Sci.* 1984, **45**, 1726

118 Saif, L.J. and Bohl, E.H. Passive immunity against enteric viral infections. In: *Proceedings Third International Symposium on Neonatal Diarrhoea* (Eds Acres, S.D., Forman, A.J. and Fast, H.) VIDO, 124, Veterinary Rd., Saskatoon, Saskatchewan, Canada, 1980, pp. 83–98

119 Bohl, E.H. and Saif, L.J. Passive immunity against enteric viral infections of piglets. In: *The mucosal immune system.* (Ed. Bourne, F.J.) Martinus Nijhoff, 1981, pp. 259–278

120 Chidlow, J.W. and Porter, P. Intestinal defence of the neonatal pig: Inter-relationships of gut and mammary function providing surface immunity against colibacillosis. *Vet. Rec.* 1979, **104**, 496