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Review Article

Intestinal barrier function and absorption in pigs after weaning: a review

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Abstract

Under commercial conditions, weaning of piglets is associated with social, environmental and dietary stress. Consequently, small-intestinal barrier and absorptive functions deteriorate within a short time after weaning. Most studies that have assessed small-intestinal permeability in pigs after weaning used either Ussing chambers or orally administered marker probes. Paracellular barrier function and active absorption decrease when pigs are weaned at 3 weeks of age or earlier. However, when weaned at 4 weeks of age or later, the barrier function is less affected, and active absorption is not affected or is increased. Weaning stress is a critical factor in relation to the compromised paracellular barrier function after weaning. Adequate feed intake levels after weaning prevent the loss of the intestinal barrier function. Transcellular transport of macromolecules and passive transcellular absorption decrease after weaning. This may reflect a natural intestinal maturation process that is enhanced by the weaning process and prevents the pig from an antigen overload. It seems that passive and active absorption after weaning adapt accurately to the new environment when pigs are weaned after 3 weeks of age. However, when weaned at 3 weeks of age or earlier, the decrease in active absorption indicates that pigs are unable to sufficiently adapt to the new environment. To improve weaning strategies, future studies should distinguish whether the effect of feed intake on barrier function can be directed to a lack of a specific nutrient, i.e. energy or protein.

Key words: Pigs: Weaning: Intestinal barrier: Intestinal absorption

The small-intestinal epithelium has three major functions: (1) the digestion and absorption of nutrients; (2) the secretion and absorption of water and electrolytes to maintain a proper viscosity of the luminal content and to flush out noxious components; (3) serving as a barrier against noxious antigens and pathogens. Impaired intestinal barrier function or an increased intestinal permeability may promote the translocation of bacteria and the entering of allergenic compounds from the gut into the body. This results in immunological responses and an increased susceptibility to infections[1,2]. Weaning of pigs is associated with social, environmental and dietary stress[3], and in rats and humans, various stressors will deteriorate the small-intestinal barrier function[4,5]. Evidence for weaning stress in pigs is that cortisol and corticotropin-releasing factor concentrations in the blood plasma are increased after weaning[6,7]. Moreover, about 10 % of the pigs do not ingest any feed during the first 48 h after weaning[8], and most other pigs have a low feed intake. In addition, low feed intake after weaning is consistently associated with villous atrophy in the small intestine[3,9,10]. After weaning, pigs are especially susceptible to infections[3]. Oedema disease, caused by the Shiga-like toxin type II variant from some *Escherichia coli* strains, is associated with the process of weaning but requires a disturbed intestinal barrier function in order to enable large toxin molecules to pass through the intestinal epithelium[11]. Thus, the effect of weaning on intestinal morphology, susceptibility to infections and occurrence of oedema disease indicates that intestinal barrier function is disturbed after weaning. Moreover, a reduced villous surface after weaning implicates a reduction in intestinal absorptive capacity as well.

Abbreviations: GlySar, glycylsarcosine; HRP, horseradish peroxidase; Isc, short-circuit current; Na-Flu, sodium-fluorescein isothiocyanate; TEER, transepithelial electrical resistance.

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The present review discusses the effect of weaning and dietary treatments after weaning on the intestinal barrier function and absorption. The study opens with some background information regarding epithelial transport in the small intestine. Subsequently, the most commonly used techniques, Ussing chambers and orally administered marker probes, to assess intestinal barrier function and intestinal absorption are discussed. Eventually, the review concentrates on the effects of weaning and treatments after weaning on the intestinal barrier function and absorption in pigs.

Background of epithelial transport

Transport across the small-intestinal epithelium can be separated into paracellular and transcellular pathways. Paracellular transport represents diffusion between epithelial cells. Networks of proteins called tight junctions connect the epithelial cells and 'seal' the space between the cells. The tight junctions are selectively permeable for ions, small molecules and water[12]. Transcellular transport represents either the uptake of small molecules (e.g. nutrients) by carrier-mediated (active) or carrier-unmediated (passive) transport through absorptive small-intestinal cells (enterocytes) or the uptake of macromolecules by endocytosis. Endocytosis is specifically important with respect to maternal Ig uptake after birth and for antigen uptake. In healthy animals, antigen uptake is precisely regulated to train the immune system. Antigen uptake by endocytosis is more common in the follicle-associated epithelium, covering the Peyer's patches, than in enterocytes[13]. In the present review, we distinguish between small-intestinal barrier function and small-intestinal absorption. For this distinction, we associate a disturbed barrier function with increased paracellular transport and transepithelial transport (paracellular and transcellular) of macromolecules into the body.

Techniques to measure epithelial transport

Ussing chamber

Intestinal barrier function and absorption in pigs after weaning have mainly been assessed in *ex vivo* studies with Ussing chambers. In Ussing chambers, a section of intestinal mucosa is mounted between two chambers. Marker probes are added to the solution in the chamber at the mucosal site. The appearance of these marker probes in the chamber at the serosal site represents the permeability for these probes. Table 1 gives an overview of probes that have been used in studies with pigs after weaning. In addition, Table 1 gives three electrophysical parameters that can be determined in Ussing chambers. First, the transepithelial electrical resistance (TEER) of the mounted intestinal mucosa can be determined. This is considered to reflect the opening of the tight junctions between epithelial cells, i.e. the paracellular permeability of the intestinal mucosa[14]. An increased TEER reflects decreased paracellular permeability, and a decreased TEER reflects increased paracellular permeability. Second, the transepithelial electrical conductance can be determined. This is the inverse of the TEER[14]. Finally, the short-circuit current (Isc) over the mucosa can be measured. This is a measurement of active electrogenic ion transport across the epithelium. Increased Isc reflects either increased electrogenic anion secretion (e.g. Cl^- and HCO_3^-) or increased electrogenic cation absorption (Na^+). *In vivo*, the water flow over the intestinal epithelium follows the osmotic gradient induced

Table 1. Marker probe characteristics and electrophysical parameters

Probe	Molecule size (Da)	Transepithelial routes/description
Probes used in Ussing chambers		
Ovalbumin	45 000	Transcellular by endocytosis[40]
Horseradish peroxidase	40 000	Transcellular by endocytosis[32,63] as well as paracellular through tight junctions[31]
Na-Flu	376	Passive transcellularly[7,40,54]
Mannitol	182	Mainly via a paracellular route[16,29], passive transcellular routes cannot be excluded[30]
D-Glucose*	180	Carrier-mediated transcellular transport[34]
Glutamine*	146	Carrier-mediated transcellular transport[34]
Glycylsarcosine	146	Via a H^+ carrier-mediated transcellular route[43]
Electrophysical parameters used in Ussing chambers		
TEER	–	Reflects mainly paracellular transport[14]
Conductance	–	Inverse of TEER[14]
Short-circuit current	–	Measures active ion transport and gives indication of water movement and electrolyte-dependent glucose and amino acid absorption[14]
Probes used *in vivo*		
Bovine serum albumin	66 000	Transcellular by endocytosis[40]
Lactulose	342	Paracellular[64]
Mannitol	182	Mainly via a paracellular route[16,29], passive transcellular routes cannot be excluded[30]
L-Rhamnose	164	Transcellular (aqueous pores) or paracellular[16]
D-Xylose	150	Passive transcellular, paracellular[55] or carrier-mediated transcellular transport[16]

Na-Flu, sodium-fluorescein isothiocyanate; TEER, transepithelial electrical resistance.
* Determined by measuring the change in short-circuit current after the addition of glucose or glutamine to the mucosal site of the chamber.

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by actively transported electrolytes. Therefore, Isc can be used to indicate water movement over the epithelium[14]. Moreover, the change in Isc after the addition of specific nutrients to the mucosal solution (e.g. glucose and glutamine) is an indirect measure of Na-dependent nutrient absorption.

Orally administered marker probes

In vivo permeability tests have been used to assess intestinal permeability in human and medical research for many decades[15]. The principle of the test is that orally administered test substances (probes) that are not metabolised in the body pass through the intestinal epithelium and are excreted in the urine within a short time after administering. Most frequently used probes are those monosaccharides and disaccharides that are not degraded by digestive enzymes, that are hardly metabolised in the body but that are fermented by 'colonic' bacteria[16]. Because of these three characteristics, the sugars are almost exclusively absorbed by the small intestine, and the influences of digestion and metabolism are minimised. Therefore, depending on their permeation route, they can be used as specific markers for small-intestinal permeability function, absorption function or both[16].

In vivo permeability tests have been used in pigs around weaning. The most frequently used test in this respect is the D-xylose absorption test[9,17–21]. In studies with pigs, D-xylose is exclusively analysed in blood 1 h after an oral dose. In humans, both blood and urine are used for the D-xylose test. In animals, it is easier to sample blood than to perform a quantitative urine collection. However, a single blood measurement is affected by many factors, for instance, rate of gastric emptying, intestinal absorption rate and clearance rate from the blood[22]. Therefore, in general, a correct quantitative urine collection is better than a single blood sampling in order to get an accurate permeability or absorption estimate[22]. Even though, absorption and permeability tests based on quantitative urine collection can also be disturbed by several factors. Most important in this respect are a marked renal dysfunction, an incomplete urine collection and an increased luminal clearance of the marker probes by bacterial overgrowth[22,23].

It is evident that knowledge about the permeation routes of the used marker probes is required in order to draw conclusions with physiological relevance. Of the marker probes that are most commonly used with this technique, we have gathered the most relevant information in Table 1. The test results with this technique can, however, be influenced by many premucosal factors (gastric emptying, intestinal transit time and bacterial degradation) and postmucosal factors (metabolism, endogenous production, completeness of urinary collection and renal function)[16]. To reduce the effects of those premucosal and postmucosal factors, the theory of differential urinary excretion of marker probes has been introduced[16,24]. In this approach, it is common practice to use both a disaccharide and a monosaccharide, which are both transported over the epithelium by unmediated diffusion. The transport of monosaccharides used for this test occurs either through tight junctions between the epithelial cells or through aqueous pores in the cell. For disaccharides, the transport occurs through the tight junctions of the crypts[2,16]. The ratio of the urinary recovery of the two sugars provides information about the intestinal barrier function. The assumption is that both probes are affected by the premucosal and postmucosal factors to a similar extent, and their ratio is not disturbed by those factors[2]. In this so-called 'dual sugar test', most often, lactulose is used as a disaccharide to assess paracellular permeability and L-rhamnose or mannitol (a sugar alcohol) is used as a monosaccharide. As an example, an increase in the lactulose:L-rhamnose ratio indicates a decrease in the intestinal barrier function, whereas a decrease in the lactulose:L-rhamnose ratio indicates an improved intestinal barrier function. The use of the dual sugar test in pigs is to our knowledge limited to four studies. In two studies, the effect of parenteral *v.* enteral nutrition in neonatal piglets[25,26] has been addressed with lactulose and mannitol as marker probes. In another study, the effect of an lipopolysaccharide challenge in 20 kg gilts has been evaluated with lactulose and L-rhamnose as marker probes[27]. In the fourth study, enhanced dietary Zn concentrations in the weaner diet decreased the lactulose:mannitol ratio in pigs at 2 weeks after weaning[28].

Thus, studies in pigs shortly after weaning with orally dosed marker probes so far have been limited to a few that have assessed the absorptive small-intestinal function with D-xylose. Up to now, the use of the dual sugar tests to address small-intestinal barrier function in pigs shortly after weaning is limited to one study.

Comparison of techniques

With orally administered marker probes, intestinal barrier function and absorption itself can be measured without killing the pig. This enables repeated measurements on the same pig over time, which allows correlating permeability and absorption parameters with performance and health parameters over time. This is a clear advantage over the Ussing chamber technique for which the pigs need to be killed. However, Ussing chamber measurements enable the assessment of the permeability at a specific intestinal site. Thus, both techniques are complementary.

Intestinal barrier function after weaning

The effects of weaning and weaning conditions on the intestinal barrier function in pigs have been assessed in a small number of studies using Ussing chambers.

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Mannitol and TEER have been used to assess the barrier function related to paracellular transport, and horseradish peroxidase (HRP) has been used to assess the barrier function related to endocytosis. Mannitol is a sugar alcohol with a molecular mass of 182 Da. It is thought to cross the mucosa mainly via a paracellular route[16,29], but also transcellular routes cannot be excluded[30]. HRP is a 40 kDa protein with enzymatic activity. The enzymatic activity makes it possible to detect low concentrations of HRP. Therefore, HRP is a very sensitive marker to measure the transport of low amounts of macromolecules over the epithelium. The basal flux of intact HRP across the intestinal epithelium occurs mainly through transcellular transport via endocytosis[31,32]. A compromised barrier function may increase transcellular endocytosis of HRP[32] as well as paracellular transport of HRP through large pores in the tight junctions[31]. Therefore, HRP is primarily used as a marker of antigen uptake through endocytosis[33].

Transcellular transport (endocytosis)

In two studies with pigs, it has been revealed that in the proximal jejunum, the HRP flux was decreased at 2, 5 and 15 d after weaning[34] and at 4 and 7 d after weaning[35] compared with pre-weaning levels (Table 2). We suggest that after weaning, the natural maturation process, enhanced by weaning may reduce the permeability for macromolecules by a reduction in endocytosis rate. This is further supported by a 90 % lower HRP flux at 35 d after weaning compared with 15 d after weaning, as has been established by Boudry *et al.*[34]. This suggested that the maturation process may be a beneficial mechanism that prevents the animal suffering from an antigen overload. Such an antigen overload may result in an excessive activation of the immune system when the pigs are subjected to their new environment after weaning. In a study of van der Meulen *et al.*[7], in the mid-jejunum, the HRP flux increased at 4 and 7 d after weaning compared with 1 d after weaning. In that study, a pre-weaning measurement was not done (first measurement 1 d after weaning), and the pigs were transported and separated from the other pigs and fasted overnight before Ussing chamber measurements were performed. The handling stress in this experiment the day before the measurements can explain the contradiction with the results of the two studies that measured HRP flux in the proximal jejunum[34,35]. Stress has been shown to increase small-intestinal HRP flux in rats[33]. The handling stress the day before the Ussing chamber measurements on top of the effect of weaning may have generated the increased HRP flux over time in the experiment of van der Meulen *et al.*[7]. Unlike in the jejunum, the HRP flux in the ileum was not affected after weaning in the study of Boudry *et al.*[34]. These differences of the ileum compared with the jejunum may relate to the fact that the basal HRP flux at weaning in the ileum was only 35 % of that in the jejunum. This indicates that at weaning, antigen sampling is already at a lower level in the ileum, and a further reduction (maturation) in this respect may not be beneficial.

The HRP flux in the proximal jejunum was not affected by feed intake level after weaning[35], and in the mid-jejunum, it was not affected by feed intake level before weaning and by weaning age (4 *v.* 7 weeks)[7,35]. Boudry *et al.*[36] changed the diet of pigs of 25 kg (4–6 weeks after weaning) from a milk replacer to a barley- or wheat-based diet. At 4 d after this dietary change, the HRP flux in the proximal jejunum of the barley- and wheat-fed pigs was not different from that of control pigs fed the milk replacer. Also, extra tryptophan in the diet after weaning (5 g/kg diet) did not affect the HRP flux in the mid-small intestine at 4, 5 or 6 d after weaning of pigs at 25 d of age[37]. Egberts *et al.*[38] found no effect of enterotoxigenic *E. coli* infection on proximal jejunal permeability for HRP (measured *in vivo*) 48 h after the infection in 3-week-old pigs. In suckling piglets, Boudry *et al.*[39] have reported that the effect of mast cell degranulation (this is a stressor that increases permeability) on ileal permeability to HRP decreased with age. This effect of age occurred earlier in piglets of sows fed *n*-3 fatty acids (minimum at 7 d of age) than in piglets of sows fed the control diet (minimum at 28 d of age). In line with this, Rådberg *et al.*[40] have shown that oral administration of red kidney bean lectins in 2-week-old suckling piglets reduced the small-intestinal permeability for large molecules (bovine serum albumin, 67 kDa; ovalbumin, 45 kDa). Thus, dietary treatments were not able to affect HRP fluxes after weaning, but specific dietary treatments before weaning could reduce small-intestinal permeability for macromolecules. This indicates that the level of antigen uptake is higher before weaning than after weaning, creating a window that enables dietary treatments to have an effect. This further supports the hypothesis that antigen uptake decreases over time after weaning as a result of a maturation process.

Paracellular transport

Moeser *et al.*[41] have shown that mannitol flux and TEER over the mid-jejunum were not different at 1 d after weaning compared with unweaned controls for pigs weaned at 28 d of age. However, for pigs weaned at 3 weeks of age, Moeser *et al.*[6,41] and Boudry *et al.*[34] have reported that the mannitol flux over the proximal or mid-jejunum was significantly increased, and TEER was significantly decreased at 1 and 2 d after weaning compared with weaning or compared with unweaned controls (Table 2). This shows that weaning pigs at a higher age can prevent the loss of the paracellular barrier function after weaning. In addition, Moeser *et al.*[6,41] have shown that in 3-week-old pigs, TEER and mannitol flux were not affected by the weaning process when stress pathways were blocked with a corticotropin-releasing factor receptor

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Table 2. Small-intestinal barrier function in pigs after weaning as measured by horseradish peroxidase flux, mannitol flux and transepithelial electrical resistance in Ussing chambers

References	Intestinal segment	Weaning age (d)	Feed intake level at different days after weaning (fraction of energy requirement for maintenance)	Change after weaning (%)	Treatment description (first mentioned = control if unclear)	Change compared with control (%)
Horseradish peroxidase						
Boudry *et al.*[34]	Proximal jejunum	21	d2: 0, d5: 1, d8: 2, d15: 4	d2: −80*, d5: −64*, d8: −27, d15: −63*, d35: −96*		
	Ileum			d2: 25, d5: −53, d8: −39, d15: 29		
Verdonk *et al.*[35]	Proximal jejunum	26	Low: d1: 0, d2: 0, d3: 1, d4: 1·5, d5: 1·5, d6: 1·5, d7: 1·5	d4: −77*, d7: −51*	Low *v.* high feed intake	d4: 130, d7: 10
			High: d1: 0·5, d2: 1, d3: 1·5, d4: 2, d5: 2·5, d6: 2, d7: 2	d4: −47, d7: −47		
Van der Meulen *et al.*[7]	Mid-jejunum	28	Control: d1: 1, d2: 2, d3: 2, d4: 2, d5: 2, d6: 2, d7: 2 Creep feed: d1: 1, d2: 2, d3: 2, d4: 2, d5: 2, d6: 2, d7: 2	–	No creep feed or creep feed	Average d1, d4 and d7: 17
		49	Control: d1: 2, d2: 2, d3: 2, d4: 3, d5: 3, d6: 3, d7: 4 Creep feed: d1: 3, d2: 3, d3: 3, d4: 4, d5: 4, d6: 4, d7: 4	–	Weaning at 28 or 49 d of age	Average d1, d4 and d7: 9
Boudry *et al.*[36]	Proximal jejunum	28	d28–25 kg: 4·5 (milk diet). Diet switch at 25 kg, thereafter: d1: 0, d2: 1·5, d3: 3, d4: 4·5	–	Switch to the barley diet at 25 kg	d4: 20
					Switch to the wheat diet at 25 kg	d4: 33
Koopmans *et al.*[37]	Mid-jejunum	25	Average of d0 to d10: 1·5 (first days low intake)	–	5 g/kg Trp in the weaner diet	d4: −53, d5: −42, d6: 31
Egberts *et al.*[38]	Proximal jejunum	21†	–	–	Enterotoxigenic *Escherichia coli* infection	d2: not affected
Mannitol						
Verdonk *et al.*[35]	Proximal jejunum	26	Low: d1: 0, d2: 0, d3: 1, d4: 1·5, d5: 1·5, d6: 1·5, d7: 1·5	d4: 12, d7: −22	Low *v.* high feed intake	d4: 16 and d7: −37
			High: d1: 0·5, d2: 1·0, d3: 1·5, d4: 2, d5: 2·5, d6: 2, d7: 2	d4: 29, d7: −51		
Verdonk[44]	Proximal jejunum	26	Dry: d1: 0, d2: 0, d3: 0·5, d4: 1·5, d5: 1·5, d6: 1·5	d2: 216*, d6: 51	Dry *v.* wet feed	Average d2 and d6: 11
			Wet: d1: 0·5, d2: 0·5, d3: 1, d4: 1, d5: 1·5, d6: 1·5	d2: 180*, d6: 159*		
	Mid-jejunum	26	Low: d1: 0·5, d2: 1, d3: 1, d4: 1	d1: 53, d2: 82*, d4: 111*	Low *v.* high feed intake	Average d1, d2 and d4: −32*
			High: d1: 2·5, d2: 3·5, d3: 3·5, d4: 3·5	d1: 4, d2: 35, d4: 31		
Spreeuwenberg *et al.*[43]	Mid-jejunum	26	d1: 0·5, d2: 1, d3: 1, d4: 1	d1: 23, d2: 85*, d4: 80*	24 % lactose and 30 % dietary protein *v.* 41 % lactose and 15 % dietary protein	Average d1, d2 and d4: −25
					24 % lactose and 30 % dietary protein *v.* 8 % lactose and 45 % dietary protein	Average d1, d2 and d4: −10
Moeser *et al.*[41]	Mid-jejunum	19	*Ad libitum* available, intake not given	d1: 151* (compared with unweaned)	Injection with mast cell-stabilising drug cromolyn before and after weaning	d1: −38*

Table 2. *Continued*

References	Intestinal segment	Weaning age (d)	Feed intake level at different days after weaning (fraction of energy requirement for maintenance)	Change after weaning (%)	Treatment description (first mentioned = control if unclear)	Change compared with control (%)
		28	*Ad libitum* available, intake not given	d1: −4 (compared with unweaned)		
Moeser *et al.*[6]	Mid-jejunum	19	*Ad libitum* available, intake not given	d1: 148* (compared with unweaned)	Injection with corticotropin-releasing factor antagonist α-helical before and after weaning	d1: −29*
Lodemann *et al.*[47]	Mid-jejunum	28	Not given	d7: −44*, d28: 27*	Probiotic (*Enterococcus faecium* SF68) added to the sow and piglet diets	d0: −31*, d7: −7 and d28: −2
Lodemann *et al.*[48]	Mid-jejunum	28	Not given	Control: d7: −31, d28: −7	Probiotic (*Bacillus cereus* var. *toyoi*) added to the sow and piglet diets	Average d0, d7 and 28: −3
				Probiotic: d7: −25, d28: −5		
Transepithelial electrical resistance						
Boudry *et al.*[34]	Proximal jejunum	21	d2: 0, d5: 1, d8: 2, d15: 4	d2: −67*, d5: 21, d8: −6, d15: 3		
	Ileum			d2: −18, d5: 116*, d8: 64, d15: 91*		
Boudry *et al.*[36]	Proximal jejunum	28	d28–25 kg: 4·5 (milk diet). Diet switch at 25 kg, thereafter: d1: 0, d2: 1·5, d3: 3, d4: 4·5	–	Switch to the barley diet at 25 kg	d4: −14
					Switch to the wheat diet at 25 kg	d4: −15
Moeser *et al.*[41]	Mid-jejunum	19	*Ad libitum* available, intake not given	d1: −40* (compared with unweaned)		
		28	*Ad libitum* available, intake not given	d1: 9 (compared with unweaned)		
Moeser *et al.*[6]	Mid-jejunum	19	*Ad libitum* available, intake not given	d1: −74*, d2: −48*, d7: −18* (compared with unweaned)	Injection with corticotropin-releasing factor antagonist α-helical before and after weaning	d1: 52*
Lodemann *et al.*[48]	Mid-jejunum	28	Not given	Control: d7: −11, d28: 8	Probiotic (*Bacillus cereus* var. *toyoi*) added to the sow and piglet diets	Average d0, d7 and d28: 2
				Probiotic: d7: −2, d28: 11		
Carlson *et al.*[49]	Ileum	28	Not given but probably already nearly at the maintenance level the second day after weaning	d1–2: 12, d5–6: 32*, d14–15: 27*	(100 mg Zn, 20 mg Cu/kg) *v.* (100 mg Zn, 175 mg Cu/kg)	d5–7: −10
					(100 mg Zn, 20 mg Cu/kg) *v.* (2500 mg Zn, 20 mg Cu/kg)	d5–7: −10
					(100 mg Zn, 20 mg Cu/kg) *v.* (2500 mg Zn, 175 mg Cu/kg)	d5–7: 0
Carlson *et al.*[50]	Mid-jejunum	28	d0–d6: 1·5 (100 mg Zn) and 1 (2500 mg Zn)	–	100 mg Zn/kg *v.* 2500 mg Zn/kg	d5–6: 0
Hamard *et al.*[51]	Ileum	7	d1: 1, d2: 1, d3–14: 2	–	Adequate (9·3 g/kg) *v.* deficient (6·5 g/kg) dietary threonine	d14: −29

d, Day.
Values were significantly different from those at weaning or from the control treatment: *$P<0·05$.
† Piglets were delivered by caesarian section and subsequently housed in isolators and fed *ad libitum* condensed cows' milk until the treatment started at 3 weeks of age.

antagonists or with a mast cell-stabilising drug. This shows that stress is a major factor with respect to the disturbed intestinal barrier function after weaning. Moreover, it shows that the immune system through mast cell activation has a critical role in the loss of the intestinal barrier function after weaning. Several other studies have shown that intestinal barrier function was compromised in pigs weaned at an age of 26 d[35,42–44]. Verdonk[44] measured the mannitol flux in the mid-jejunum of pigs with a low *v.* a high intake level of milk replacer after weaning (Table 2). The mannitol flux was not affected in pigs with a high feed intake level at 1, 2 and 4 d after weaning. However, mannitol flux was increased for pigs with low feed intake levels at 2 and 4 d after weaning compared with pre-weaning. As an average over days 1, 2 and 4, this resulted in a higher mannitol flux for pigs with low feed intake compared with pigs with high feed intake (at least 2·5 times energy maintenance). In line with this, a 2 d fast of 23-d-old pigs increased transepithelial electrical conductance (the opposite of TEER)[45]. Thus, a sufficient feed intake after weaning prevents the loss of the barrier function of the tight junctions after weaning. This indicates the importance of a sufficient luminal nutrient supply to maintain the barrier function. Sufficient feed intake may be especially important for the proximal small intestine because this part depends more on luminal nutrient supply than the distal small intestine[46]. In the study of Verdonk[44], villus height in the proximal small intestine decreased after weaning for pigs at the low intake level and was hardly affected after weaning in pigs at the high intake level. This illustrates that luminal nutrient supply was adequate in the high-intake group and inadequate in the low-intake group. In line with the study of Verdonk[44] described earlier, Spreeuwenberg *et al.*[43] have shown that the mannitol flux in the mid-jejunum increased at 2 and 4 d after weaning compared with pre-weaning. Moreover, Verdonk *et al.*[35,44] published two other experiments in which the mannitol flux was measured over the epithelium of the proximal instead of the mid-jejunum in pigs after weaning (Table 2). In the first experiment, the mannitol flux was increased at 2 and 6 d after weaning compared with the pre-weaning flux. However, in the second experiment, the mannitol flux at 4 and 7 d after weaning was not different from pre-weaning fluxes and was not affected by feed intake level after weaning. Thus, the last study, in contrast to the first two studies of Verdonk[35,44] and the study of Spreeuwenberg *et al.*[43], has shown no increase in mannitol flux after weaning, although feed intake levels were reasonably low. This discrepancy is probably related to handling of the pigs before the start of the experiment. In the first two studies of Verdonk and in the study of Spreeuwenberg *et al.*[43], pigs were transported from another location before the start of the experiment. However, in the last experiment of Verdonk[44], piglets were weaned and kept at the same location at which the experiment was conducted. Thus, in this last experiment, the weaning process was probably less stressful because pigs were not transported in a trailer. Because stress is an important factor with respect to the intestinal barrier function (see above), this may explain the differences between the studies. Lodemann *et al.*[47,48] have found that at 7 d after weaning, the mannitol flux in the mid-jejunum was either significantly or numerically lower from before weaning, and TEER was not different than before weaning. In these experiments, the timing of permeability measurements may have been too late to detect a disturbed barrier function or, again, the stress level around weaning may have been lower than in the other experiments. In the ileum, TEER was increased at 5 and 15 d after weaning compared with pre-weaning levels in a study of Boudry *et al.*[34]. Carlson *et al.*[49] have shown similar results in the ileum with pigs weaned at 4 weeks of age. In their study, the transepithelial electrical conductance (the opposite of TEER) was not different from weaning at 1–2 d after weaning and was lower at 5–6 and 14–15 d after weaning compared with weaning. Thus, in the ileum, in contrast to the proximal and mid-jejunum, intestinal barrier function is not compromised due to the weaning process. Finally, Berkeveld *et al.*[21] administered pigs an oral mannitol dose before and at 0·5, 2, 4 and 7 d after weaning and measured plasma mannitol concentrations 1 h after each dose. They observed that the plasma mannitol concentrations decreased gradually after weaning, being significantly different from pre-weaning concentration at 4 d after weaning. This contradiction between the results of Berkeveld *et al.*[21] (orally administered mannitol) with the other studies (Ussing chambers) may relate to the different techniques used to determine intestinal mannitol transport. The study of Berkeveld *et al.*[21] may reflect mannitol permeability in the whole small intestine, and in this study, mannitol was measured in the blood instead of in the urine, which is a complicating factor in evaluating the results (see above).

In general, dietary treatments after weaning showed only minor effects on paracellular intestinal permeability (Table 2). Supplementing different probiotics to the diets of the sow and piglets had no effect on mid-jejunal mannitol flux and TEER of piglets after weaning[47,48]. In two studies of Carlson *et al.*[49,50], it was revealed that dietary Cu and Zn levels had no effect on the transepithelial electrical conductance of the jejunum and ileum at 5–7 d after weaning. However, in a study of Zhang & Guo[28], urinary latulose:mannitol ratios were decreased after feeding enhanced dietary Zn concentrations for a period of 2 weeks after weaning. The discrepancy between the studies of Carlson *et al.* and the study of Zhang & Guo may again relate to the stress level during the study. In the study of Zhang & Guo, pigs were separated from the other pigs, individually housed and fasted overnight before the permeability test. Boudry *et al.*[36] changed 25 kg pigs from a milk replacer diet to a barley- or

wheat-based diet on an equal intake level. The jejunal TEER was not different from the milk-fed pigs 4 d after this dietary change. Finally, the combination of an increased dietary lactose concentration (41 *v.* 24 %) and a decreased dietary protein concentration (15 *v.* 30 %) tended to decrease mannitol flux in the mid-small intestine[43]. Thus, no distinction could be made between a possible (positive) effect of lactose and a possible (negative) effect of protein on mannitol flux. In the same study, the combination of a decreased dietary lactose concentration (8 *v.* 24 %) and an increased dietary protein concentration (45 *v.* 30 %) had no effect on mannitol flux. In contrast to the studies described earlier, Hamard *et al.*[51] weaned pigs at 7 d of age and showed that dietary threonine deficiency, for a period of 2 weeks after weaning, increased the ileal permeability for fluorescein isothiocyanate dextran (4 kDa) and also tended to decrease TEER. The aforementioned studies indicate that diet composition in general has not a major effect on paracellular permeability. However, when diets deficient in nutrients (e.g. threonine) are fed, paracellular permeability deteriorates. In line with this, intestinal barrier function is compromised at low feed intake levels, and, in this respect, a low feed intake level is similar to a diet being deficient in all nutrients. In addition, diet composition may affect paracellular permeability when permeability measurements are accompanied with additional stressors (i.e. fasting and individual housing).

Conclusions regarding barrier function

The present review clarifies that small-intestinal barrier function in pigs is affected by the process of weaning. In the literature, four factors (i.e. weaning age, weaning stress, feed intake and diet composition) have been identified that can have a major effect on the barrier function after weaning. In addition, barrier function is differently affected after weaning in the proximal and mid-jejunum than in the ileum. The relationships between these different aspects of the intestinal barrier function after weaning are illustrated in Fig. 1. The stress that is associated with weaning manipulates the immune system, resulting in mast cell activation that has a critical role in the loss of the barrier function of the tight junctions in the small intestine. In addition, the loss of the paracellular barrier function is prevented when feed intake after weaning is at an adequate level such that the loss of the villous height is prevented. This shows that low feed intake is another factor that seems to have a critical role in the compromised barrier function after weaning. In contrast to the proximal and distal jejunum, paracellular barrier function after weaning is not compromised in the ileum. Because luminal nutrient supply is most critical in the proximal small intestine, this indicates that the loss of the barrier function due to low feed intake is due to a shortage of luminal nutrient supply. In line with this, it was shown that barrier function was compromised in pigs after feeding a threonine-deficient diet for a period of 2 weeks. This effect of nutrient supply on the barrier function may be a secondary effect as a result of a compromised intestinal architecture. The loss of the paracellular barrier function is almost exclusively found in the first week after weaning and returns to the pre-weaning level at 2 weeks after weaning. Pigs are less susceptible to a compromised barrier function when weaned at an older age and also in the long term have a better barrier function[41,52]. This is probably related to the earlier stage of maturation of the small intestine when pigs are weaned at a young age. Although paracellular barrier function is consistently compromised after weaning, one can argue whether this is a direct risk for the health of the pig. Moreover, transcellular barrier function for macromolecules through endocytosis improves after weaning. We suggest that this maturation process enhanced by weaning may prevent the animal

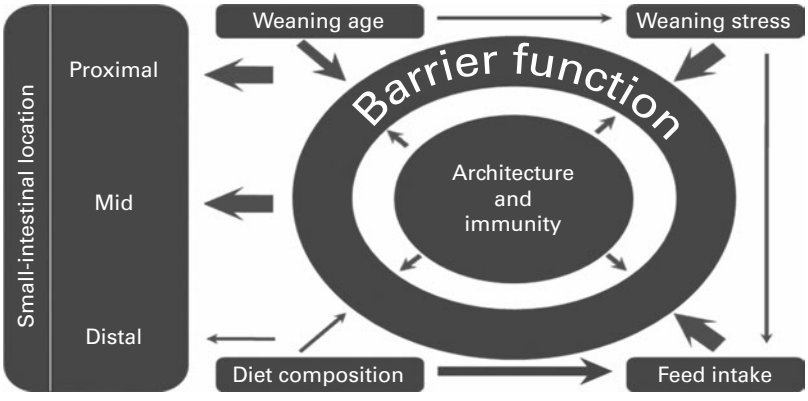

Fig. 1. Scheme representing the relationship between small-intestinal barrier function, small-intestinal location and factors (age, stress, feed intake or diet composition) that affect the barrier function. The thickness of the arrows indicates the significance of the relationship. Barrier function is less affected at high than at low weaning age, which relates probably to the intestinal maturation rate. Weaning stress compromises the paracellular barrier function indirectly through mast cell activation (immunity). Adequate feed intake levels after weaning prevent the loss of the barrier function probably indirectly through the preservation of intestinal architecture. The direct effect of diet composition on the intestinal barrier function seems to be limited unless diets are deficient in specific nutrients. Barrier function is most affected in the proximal and mid-small intestine and hardly in the distal small intestine.

suffering from an antigen overload. Such an antigen overload may result in an excessive activation of the immune system when pigs are subjected to their new environment after weaning. We hypothesise that the increased paracellular permeability also indicates that the intestine is extra susceptible for the disturbance of the transcellular barrier function when several stressors occur simultaneously.

Based on the present review, three different approaches can be followed to improve the intestinal barrier function after weaning by ways of dietary composition: first, the old-fashioned approach to improve the palatability of the diet to increase feed intake after weaning. This has only been partially successful up to now; second, to identify crucial nutrients (e.g. protein or specific amino acids) that may be supplied to pigs with low feed intake in a concentrated form or through the drinking-water in order to prevent the loss of the intestinal barrier function; third, to add specific biologically active components to the diet to modulate the stress response or the subsequent immune response, to prevent the loss of the barrier function. With this last approach, it is essential that the diet should be eaten, otherwise the active component needs to be supplied through the drinking-water.

Intestinal absorption after weaning

Small-intestinal transcellular absorption can be divided into active and passive absorption. Active absorption takes place in specific transporters that transport nutrients such as glucose or amino acids over the intestinal epithelium, which coincides with Na^+ transport. This Na^+ transport enables us to estimate the active transport over the intestinal epithelium in Ussing chambers. In pigs after weaning with this technique, the Na^+-dependent glucose and glutamine absorption have been investigated (Table 1). The change in Isc after the addition of glucose or glutamine to the mucosal site estimates the transport of glucose or glutamine to the serosal site of the chamber[14]. In addition, after weaning, glycylsarcosine (GlySar) has been used to assess active transport in Ussing chambers. GlySar is a dipeptide (146 Da, Table 1), which is believed to pass through the intestinal epithelium via a H^+ carrier-mediated transcellular route[43]. As addressed earlier, active absorption coincides with electrolyte transport (i.e. Na^+ and H^+) over the epithelium. The transport of electrolytes, but also the transport of nutrients, coincides with water movement through the tight junctions because water follows the osmotic gradient. Thus, active transport of electrolytes is also important with respect to fluid absorption. It should be noted that electrolytes are not only absorbed by the small intestine but also secreted into the lumen. Carey *et al.*[45] have shown that the net Na^+ and Cl^- movement in fasted pigs was only 25 % of the total mucosal to serosal movement. In addition, Miller & Skadhauge[53] have shown that weaning reduced Na^+ absorption but had hardly any effect on Na^+ secretion. These studies have shown that in addition to a net movement of charge, as measured by Isc, unidirectional electrolyte movements occur in the small intestine.

Sodium-fluorescein isothiocyanate (Na-Flu) and D-xylose have been used to study passive absorption in pigs after weaning. Na-Flu is a small (376 Da) fluorescent-labelled molecule with a 50:50 lipid–water solubility (Table 1)[7,54]. It has been used to study intestinal absorption with Ussing chambers. D-Xylose has been used to test the absorptive intestinal function before and after weaning *in vivo* in several studies. In these studies, D-xylose was orally administered, and plasma D-xylose concentrations were determined after 1 h. D-Xylose is a monosaccharide (150 Da, Table 1), which besides a passive transcellular route is also thought to pass through the intestinal mucosa by a carrier-mediated route or by paracellular diffusion[2,16,55].

Active absorption

In a study of Boudry *et al.*[34], pigs were weaned at 21 d of age. In that study, the Na^+-dependent glucose absorption increased in the proximal jejunum at 2 d after weaning and decreased at 15 d after weaning compared with pre-weaning absorption (Table 3). Moreover, in the same study, ileal glucose absorption decreased at 2, 5 and 15 d after weaning. In line with this, Smith[56] has found that the Na^+-dependent alanine uptake by enterocytes of the mid-small intestine (measured with a rapid uptake apparatus, using radiolabelled tracer amino acids) decreased considerably at 5 d after weaning at 2 or 3 weeks of age. Furthermore, in 4-week-old pigs, Na^+-dependent alanine uptake by enterocytes of the mid-small intestine was lower for weaned pigs (5 d after weaning, thus weaning at 23 d) than for unweaned pigs[57]. However, the alanine uptake of 6-week-old pigs (both weaned and unweaned) was similar to the alanine uptake of 4-week-old weaned pigs[57]. This study suggests that weaning (before 4 weeks of age) and ageing appear to decrease the number of enterocytes that are involved in active alanine uptake[57]. The aforementioned studies have shown that active small-intestinal absorption decreases after weaning when pigs are weaned between 14 and 23 d of age. However, when weaned after 4 weeks of age, active absorption is not affected by the weaning process. Miller *et al.*[57] have observed that the decreased absorption rates after weaning also occur in unweaned pigs but over a much longer time course. This suggests that this decrease in active absorption is part of a maturation process that is enhanced by the process of weaning. This may relate to a decrease in the relative demand for nutrients when pigs get older because weight gain expressed relative to body weight decreases when pigs get older. Several studies have investigated the active absorption of glucose, glutamine or GlySar in the proximal, mid and distal small intestine for pigs that were weaned at 26 or 28 d of age[35,43,44,47–49]. In all these studies, active absorption between 1 and 15 d after

Table 3. Small-intestinal molecular absorption in pigs after weaning as measured for Na^+-dependent glucose, Na^+-dependent glutamine, glycylsarcosine and sodium-fluorescein isothiocyanate absorption

References	Intestinal segment	Weaning age (d)	Feed intake level at different days after weaning (fraction of energy requirement for maintenance)	Change after weaning (%)	Treatment description (first mentioned = control if unclear)	Change compared with control (%)
Na^+-dependent glucose						
Boudry *et al.*[34]	Proximal jejunum	21	d2: 0, d5: 1, d8: 2, d15: 4	d2: 78*, d5: −32 d8: −43, d15: −83*		
	Ileum			d2: −62*, d5: −74*, d8: −13, d15: −54*		
Boudry *et al.*[36]	Proximal jejunum	28	d28–25 kg: 4·5 (milk diet). Diet switch at 25 kg, thereafter: d1: 0, d2: 1·5, d3: 3, d4: 4·5	–	Switch to the barley diet at 25 kg	d4: 76*
					Switch to the wheat diet at 25 kg	d4: 74*
Lodemann *et al.*[47]	Mid-jejunum	28	Not given	d7: 6, d28: −12	Probiotic (*Enterococcus faecium* SF68) added to the sow and piglet diets	d0: 27, d7: 24, d28: −16
Lodemann *et al.*[48]	Mid-jejunum	28	Not given	Control: d7: −14, d28: −28	Probiotic (*Bacillus cereus* var. *toyoi*) added to the sow and piglet diet	Average d0, d7 and d28: −7
				Probiotic: d7: −2, d28: −9		
Carlson *et al.*[49]	Ileum	28	Not given but probably already nearly at the maintenance level the second day after weaning	d1–d2: 111*, d5–d6: 112*, d14–d15: 130*	(100 mg Zn, 20 mg Cu/kg) *v.* (100 mg Zn, 175 mg Cu/kg)	d5–d7: 12
					(100 mg Zn, 20 mg Cu/kg) *v.* (2500 mg Zn, 20 mg Cu/kg)	d5–d7: 3
					(100 mg Zn, 20 mg Cu/kg) *v.* (2500 mg Zn, 175 mg Cu/kg)	d5–7: −d14
Gabler *et al.*[61]	Proximal jejunum	14–17	d1: 0	–	Sow gestation and lactation diets supplemented with either fish oil, DHA or coconut fat	Measurement at d1: fish oil: 355*, DHA: 510*, coconut fat: 190
Gabler *et al.*[60]	Proximal jejunum	15–19	d1: 0	–	Sow gestation/lactation diets supplemented with or without fish oil, resulting in four treatments: control/control, control/fish, fish/control, fish/fish	Measurement at d1: control/fish: 189, fish/control: 316*, fish/fish: 389*
Hamard *et al.*[51]	Ileum	7	d1: 1, d2: 1, d3–d14: 2	–	Adequate (9·3 g/kg) *v.* deficient (6·5 g/kg) dietary Thr	d14: 81
Glycylsarcosine						
Verdonk *et al.*[35]	Proximal jejunum	26	Low: d1: 0, d2: 0, d3: 1, d4: 1·5, d5: 1·5, d6: 1·5, d7: 1·5	d4: 29, d7: 33	Low *v.* high feed intake	d4: 18, d7: −15
			High: d1: 0·5, d2: 1, d3: 1·5, d4: 2, d5: 2·5, d6: 2, d7: 2	d4: 53, d7: 14		
Verdonk[44]	Proximal jejunum	26	Dry: d1: 0, d2: 0, d3: 0·5, d4: 1·5, d5: 1·5, d6: 1·5	d2: 169*, d6: 209*	Dry *v.* wet feed	Average d2 and d6: −28*
			Wet: d1: 0·5, d2: 0·5, d3: 1, d4: 1, d5: 1·5, d6: 1·5	d2: 109*, d6: 118*		
	Mid-jejunum	26	Low: d1: 0·5, d2: 1, d3: 1, d4: 1	d1: −13, d2: −2, d4: 9	Low *v.* high feed intake	Average d1, d2 and d4: −20
			High: d1: 2·5, d2: 3·5, d3: 3·5, d4: 3·5	d1: −35*, d2: −1, d4: −29		
Spreeuwenberg *et al.*[43]	Mid-jejunum	26	d1: 0·5, d2: 1, d3: 1, d4: 1	d1: −6, d2: 1, d4: 19	24 % Lactose and 30 % dietary protein *v.* 41 % lactose and 15 % dietary protein	Average d1, d2 and d4: 10

Table 3. *Continued*

References	Intestinal segment	Weaning age (d)	Feed intake level at different days after weaning (fraction of energy requirement for maintenance)	Change after weaning (%)	Treatment description (first mentioned = control if unclear)	Change compared with control (%)
					24 % Lactose and 30 % dietary protein *v.* 8 % lactose and 45 % dietary protein	Average d1, d2 and d4: 8
Na^+-dependent glutamine						
Lodemann *et al.*[47]	Mid-jejunum	28	Not given	d7: 17, d28: −44	Probiotic (*Enterococcus faecium* SF68) added to the sow and piglet diets	d0: 40, d7: 19, d28: 11
Lodemann *et al.*[48]	Mid-jejunum	28	Not given	Control: d7: −17, d28: 12 Probiotic: d7: 1, d28: −12	Probiotic (*Bacillus cereus* var. *toyoi*) added to the sow and piglet diets	Average d0, d7 and d28: 19
Gabler *et al.*[61]	Proximal jejunum	14–17	d1: 0	–	Gestation and lactation diets supplemented with either fish oil, DHA or coconut fat	Measurement at d1: fish oil: 400, DHA: 2425*, coconut fat: 1625
Sodium-fluorescein isothiocyanate						
Verdonk *et al.*[35]	Proximal jejunum	26	Low: d1: 0, d2: 0, d3: 1, d4: 1·5, d5: 1·5, d6: 1·5, d7: 1·5	d4: −48*, d7: −42*	Low *v.* high feed intake	d4: 6, d7: 21
			High: d1: 0·5, d2: 1, d3: 1·5, d4: 2, d5: 2·5, d6: 2, d7: 2	d4: −45*, d7: −30		
Van der Meulen *et al.*[7]	Mid-jejunum	28	Control: d1: 1, d2: 2, d3: 2, d4: 2, d5: 2, d6: 2, d7: 2 Creep feed: d1: 1, d2: 2, d3: 2, d4: 2, d5: 2, d6: 2, d7: 2	–	No creep feed or creep feed	Average d1, d4 and d7: −10
		49	Control: d1: 2, d2: 2, d3: 2, d4: 3, d5: 3, d6: 3, d7: 4 Creep feed: d1: 3, d2: 3, d3: 3, d4: 4, d5: 4, d6: 4, d7: 4	–	Weaning at 28 or 49 d of age	Average d1, d4 and d7: −18
Koopmans *et al.*[37]	Mid-jejunum	25	Average of d0–d10: 1·5 (first days low intake)	–	5 g/kg Trp in the weaner diet	d4: 11, d5: 38, d6: −1

d, Day.
Values were significantly different from those at weaning or from the control treatment: *$P<0·05$.

weaning was either similar to or higher than absorption before weaning. This confirms that active small-intestinal absorption after weaning is only suppressed when pigs are weaned before 4 weeks of age. The only contradiction to this is the study of Buddington *et al.*[58]. They measured carrier-mediated asparagine, leucine, lysine, methionine and proline absorption (per unit of wet mass) in the mid-small intestine for pigs weaned between 32 and 35 d of age. They showed that for all amino acids, active absorption was lower after weaning (42 d of age) than before weaning (at 28 d of age). The discrepancies of the study of Buddington *et al.*[58] with the other studies may relate to the fact that they did not use Ussing chambers or a rapid uptake apparatus.

In a study of Verdonk[44], energy intake during the first 2 d after weaning was either low (close to 0 for pigs on a dry diet) or moderate (0·5 times maintenance for pigs fed a wet diet). In this study, the average GlySar absorption at days 2 and 6 was higher for pigs with low feed intake than for pigs with moderate feed intake. In rats, starvation increased the expression of mRNA of peptide transporter 1, and along with the up-regulation of this transporter protein, the activity of GlySar uptake was enhanced[59]. This may explain why GlySar absorption increased in pigs with low feed intake levels after weaning. In another study of Verdonk[44], pigs were fed milk replacers, and the energy intake during the first 2 d after weaning was either moderate (0·5 times maintenance) or high (3·0 times maintenance). GlySar absorption in the mid-jejunum at 1, 2 and 4 d after weaning was not different from pre-weaning for treatments with moderate energy intake. However, GlySar absorption at day 1 was lower compared with pre-weaning in pigs with high energy intake. Thus, low feed intake stimulates active absorption. In agreement with this, active glucose absorption in the proximal jejunum was stimulated after pigs were fasted for a period of 2 d[45]. Moreover, Boudry *et al.*[34] have shown that active glucose absorption in the proximal jejunum increased when pigs were fasted for a period of 2 d after weaning. In contrast, in the same study, ileal glucose absorption decreased after weaning. This difference in glucose absorption between the proximal and distal small intestine is probably because the proximal region depends more on luminal nutrient supply than the distal region[46]. Boudry *et al.*[36] changed pigs of 25 kg (4–6 weeks after weaning) from a milk replacer to a barley- or wheat-based diet. It was observed that 4 d after this dietary change, the Na^+-dependent glucose absorption in the proximal jejunum of the barley- or wheat-fed pigs was higher than that of the control pigs fed the milk replacer. This indicates that after weaning, the shift from a milk- to a cereal-based diet increases active small-intestinal absorption. In conclusion, the short-term increase in active absorption in the proximal small intestine after weaning is due to the low feed intake after weaning. The long-term increase is related to the shift from a milk- to a cereal-based diet. The addition of fish oil or DHA to gestation diets of the sow increased Na^+-dependent glucose and glutamine absorption in the proximal jejunum at 24 h after weaning in 15- to 20-d-old pigs[60,61]. Dietary threonine deficiency tended to increase ileal Na^+-dependent glucose absorption at 14 d after weaning[51]. Studies with differences in dietary minerals (Cu and Zn), lactose and protein concentration and dietary probiotic addition have found no effect on active small-intestinal glucose, glutamine or GlySar absorption after weaning[43,47–50]. Thus, the shift from a milk- to a cereal-based diet and dietary fatty acid composition have a significant effect on active small-intestinal absorption, whereas some other dietary changes have no effect on absorption.

Passive absorption

Results of three studies[17,19,21] revealed that D-xylose absorption in piglets before weaning is hardly affected over time after 3 weeks of age. Results of five studies with pigs after weaning showed that absorption of D-xylose[9,17–20] decreased gradually to about 50 % of the pre-weaning level at 7 d after weaning. In line with this, the absorption of Na-Flu in the proximal jejunum decreased at 4 and 7 d after weaning compared with pre-weaning levels in pigs weaned at 26 d of age[44]. Berkeveld *et al.*[21], however, have observed that plasma D-xylose concentrations were significantly higher at 2 and 7 d after weaning than pre-weaning and were not different between pre-weaning and 0·5 and 4 d after weaning. Apart from the results of the study of Berkeveld *et al.*[21], passive absorption seems to decrease consistently after weaning. This seems to be a permanent effect because even at 14 d after weaning D-xylose absorption was only at 65 % of the absorption level measured before weaning[17]. We hypothesise that the reduced passive transcellular absorption after weaning is a defence mechanism that prevents uncontrolled transport of potential harmful agents to enter the body. This is more or less in line with what was described before with respect to the transcellular transport of macromolecules after weaning. In all these studies, weaning age, varying from 14 to 29 d of age, does not seem to have a major effect on the response after weaning. Creep feed intake of piglets before weaning had no effect on D-xylose absorption from 1 to 14 d after weaning[17,19,20]. Moreover, Na-Flu absorption was not affected by creep feed intake before weaning or by weaning age (4 or 7 weeks of age) at 1, 4 and 7 d after weaning[7]. Kelly *et al.*[62] have found no difference in D-xylose absorption at 5 d after weaning for tube-fed pigs fed at a low (0, 0·25, 0·5, 0·75, 1·0 times energy maintenance on days 1–5, respectively) or at a high (1·5, 1·75, 2·0, 2·25, 2·5 times energy maintenance on days 1–5, respectively) intake level of the diet. In addition, Pluske *et al.*[9] have shown that D-xylose absorption after weaning was not affected by the intake level of cows' milk after weaning (1·0, 2·5 or 4·0 times energy maintenance) and

was not different for pigs fed cows' milk *v.* pigs fed a dry weaner diet. Thus, passive absorption is not affected by feed intake level before or after weaning. Finally, the addition of extra tryptophan (5 g/kg diet) after weaning had no effect on Na-Flu flux in the mid-small intestines at 4, 5 or 6 d after weaning of pigs weaned at 25 d of age[37].

Conclusions regarding absorption

The present review shows that active and passive absorption are differently affected after weaning. Active absorption after weaning is influenced by three important factors (weaning age, feed intake level and feed composition). However, passive absorption decreases after weaning irrespective of these three factors. This reduced passive transcellular absorption after weaning may be a defence mechanism that prevents uncontrolled transport of potential harmful agents to enter the body. This is more or less in line with what was previously described with respect to the transcellular transport of macromolecules after weaning. The decreased passive absorption may reflect a natural maturation process of the intestine that occurs rapidly after weaning, as hypothesised earlier by other authors[56,57]. In general, active small-intestinal absorption decreases after weaning when pigs are weaned at 3 weeks of age or at a lower age. In line with the decrease in passive absorption, this decrease in active absorption may be part of a maturation process that is enhanced by the process of weaning. This may relate to a decrease in the relative demand for nutrients when pigs get older because of a decrease in weight gain relative to body weight. However, when weaned at an age of 4 weeks or later in life, active absorption is not affected by weaning or stimulated by the weaning process. This may indicate that with respect to active absorption, the small intestine is mature at 4 weeks of age. The shift from a milk- to a cereal-based diet and addition of fish oil or DHA to the diet increase active absorption. Thus, diet composition after weaning can have a significant effect on active small-intestinal absorption. Moreover, this indicates that the shift from a milk- to a cereal-based diet may be responsible for the long-term increase in active absorption after weaning. It seems that passive and active absorption after weaning adapt accurately to the changed environment after weaning with respect to feeding status when weaned after 3 weeks of age. Only when weaned at 3 weeks of age or earlier, the decrease in active absorption indicates an insufficient adaptation to the new environment that may result in an insufficient absorptive capacity. A diet that stimulates active absorption, for instance DHA, may help to overcome or prevent this sudden decrease in absorption.

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