INTRODUCTION

The occurrence of diarrhoea in piglets within three to 10 days of weaning (postweaning diarrhoea; PWD) has long been associated with the proliferation of certain serotypes of beta-haemolytic *Escherichia coli* in the proximal small intestine of affected animals (Richards and Fraser, 1961). Many of these *E. coli* isolates have been found to be enterotoxigenic [ETEC], producing either heat stable toxin 'a' [STa], heat stable toxin 'b' [STb], heat labile toxin [LT], or combinations of these; some strains also produce shiga-like vero cell cytotoxins [VT] (Smith et al., 1983; Morris and Sojka, 1985). Of these toxins, LT and STb cause fluid accumulation in intestinal loops of weaned pigs, but STa does not (Burgess et al., 1978). Forms of VT may be involved in the production of oedema disease in weaned pigs (Dobrescu, 1983; Smith et al., 1983), but their role in PWD is uncertain. Many ETEC from PWD lack the well-characterized adhesins found on *E. coli* from neonatal diarrhoea (i.e. K88, K99, 987P or F41), but may possess other uncharacterized adhesins (Nakazawa et al., 1987; Okerman, 1987). The haemolysin of *E. coli* isolates from PWD does not appear to play a part in the aetiology of the diarrhoea (Smith and Linggood, 1971), and non-haemolytic ETEC may on occasions also be recovered from natural cases of PWD (Hoblet et al., 1986). Initial infection with PWD-producing strains of ETEC can occur in the farrowing house, the organisms then being carried into the weaner house undetected in the intestinal tract (Miller et al., 1984a); alternatively the ETEC infecting weaned pigs can originate in the contaminated environment of the weaner house (Hampson et al., 1987).

Despite the strong association between infection with haemolytic ETEC and occurrence of PWD, oral dosing of weaned pigs with these bacteria does not invariably reproduce the condition (Smith and Jones, 1963; Kenworthy and Allen, 1966; Armstrong and Cline, 1977). The organisms can also be found in the intestinal tract of healthy litter mates of pigs with PWD, although usually in lower numbers (Kenworthy and Crabb, 1963; Svendsen et al., 1974, 1978). These observations have led to a search for other factors influencing the occurrence of PWD, generally with the presumption that "predisposing factors" allow ETEC to establish themselves in susceptible portions of the intestinal tract in sufficient numbers to initiate diarrhoea. Although many such predisposing factors have been suggested, for example fluctuating environmental temperatures, chilling, crowding etc., the purpose of this paper is only to outline reported influences of diet and closely related factors on susceptibility to PWD, especially in relation to pigs weaned at three to four weeks of age ("early weaning").

Loss of sow milk at weaning

By definition, PWD only occurs in newly-weaned pigs. Therefore a common factor in its occurrence is recent withdrawal of sow milk. Milk has a variety of potentially important specific and non-specific protective effects against pathogenic *E. coli*. For example, if present in sufficient concentration, milk antibody may inhibit growth of *E. coli* (Wilson and Svendsen, 1971), block adhesion to enterocytes (Nagy et al., 1979) or neutralise LT (Brandenburgh and Wilson, 1973). Iron-binding proteins such as transferrin and lactoferrin may inhibit bacterial growth (Bullen et al., 1972), whilst fat globules may compete as receptor sites for certain *E. coli* adhesive factors (Atroshi et al., 1983). Withdrawal of such potential protection allows enteropathogens to proliferate, provided that they possess some selective growth advantage over other members of the intestinal flora. This protective influence of milk in pigs of weaning age has been demonstrated by Deprez...
et al. (1986), who inhibited postweaning proliferation of haemolytic E. coli in weaners by supplementing their diet with sows' milk.

**Influence of Weaner Diet**

The weaner diet differs from the preweaning one in physical state, nutrient composition, availability, and the amount of it which is consumed. Since these differences have all been suggested as factors influencing susceptibility to PWD, and despite the fact that such aspects of the diet may be interrelated, an attempt will be made to address these facets separately.

**Physical state of the weaner diet**

Weaner diets are usually offered either as a dry meal or as dry pellets. A number of authors have found, however, that liquid weaner diets have benefits over dry ones; these include reducing the number of coliform organisms in the intestinal tract (Decuypere and Van de Hyde, 1972), improving food conversion ratio (Efird, 1982a), and preventing postweaning growth checks (Leece et al., 1979). Therefore, physical transition from a liquid to a dry diet may predispose weaners to PWD. For example, Lawrence (1983) noted that wet feeding reduced gastric pH, thereby maintaining a greater bacteriocidal barrier to ingested ETEC, and influenced gastric emptying and the characteristics of the digesta and its movements in other parts of the intestinal tract. It is possible that in turn this could influence the extent of proliferation of ETEC at these sites.

Experiments have been undertaken comparing the influence of solid and liquid diets on PWD. Miller et al. (1984a) challenged one-week-old piglets with marked naladixic-acid-resistant ETEC, and then weaned them at three weeks of age onto a liquid diet of cows' milk fed twice daily. The piglets remained healthy and did not excrete ETEC. However, when the pigs were transferred two weeks later to a dry commercial diet fed *ad libitum* they developed PWD and excreted the ETEC in their faeces. Interpretation of these results is confounded by the possibility that the cows’ milk may have had some non-specific antibacterial activity, and intake of the milk was restricted, whilst the dry feed was offered *ad libitum*.

Byrne and Halls (1984) noted that germ-free gnotobiotic piglets which were transferred from a liquid diet to a solid one developed a transient reduction in height of villi and a decrease in crypt cell production rate in the proximal small intestine. When Tzipori et al. (1980a) transferred gnotobiotic piglets from a liquid to a solid diet, they found that they became less susceptible to challenge with an ETEC, although more susceptible to challenge with rotavirus. Working with caesarian-derived SPF piglets and another strain of the same serotype of ETEC, Tzipori et al. (1984) found different effects. Firstly, piglets killed two or three days after being transferred from a liquid diet to a solid one showed few alterations in height of villi or depth of crypt in the small intestine. Secondly, when challenged with the ETEC, the piglets on the dry diet developed severe diarrhoea, showed heavy bacterial adherence to the intestinal mucosa, severe blunting and fusion of villi, and crypt hyperplasia; those on the liquid diet had only minor mucosal changes, little bacterial adherence and mild or no diarrhoea. Similar results were obtained by H.S. Chang and S. Tzipori (unpublished data, quoted by Tzipori, 1985) using four-week-old caesarian derived SPF piglets transferred from a diet of cows' milk to either a wet or a dry milk-based diet. The two diets were also offered to pigs on a commercial unit with a history of PWD, and again those on the liquid diet tended to do better, with less mucosal changes, lower bacterial counts in the lumen and less adhesion to the mucosa. The benefits of a liquid diet were not, however, observed when the experiment was undertaken using a soya-bean based diet. To summarize, the advantages of the liquid form of the diet appeared to depend on the type of diet used (soyabean based diets being detrimental whether fed wet or dry), were independent of gross effects on the mucosal structure, and apparently acted by reducing the usual postweaning proliferation and adhesion of ETEC. In piglets given dry weaner diet there was both greater proliferation and more extensive adhesion of ETEC, which in turn appeared to cause mucosal changes and initiate diarrhoea.

The relationship between the changes reported above in the intestinal mucosa of caesarian-
derived SPF milk-reared piglets suffering proliferation of ETEC, and those changes normally seen after weaning in conventionally-reared piglets, is not clear. In the latter animals, there are reductions in height of villi, increases in depth of crypts and decreases in certain enterocyte brush-border enzyme activities throughout the small intestine immediately after weaning (Gay et al., 1976; Kenworthy, 1976; Smith, 1984; Hampson and Kidder, 1986; Hampson, 1986a; Miller et al., 1986). Similar changes have been seen in conventional pigs on a number of diets, including a liquid one (Hampson, 1986b), with or without the occurrence of diarrhoea, and also with or without the presence of ETEC and/or rotavirus (Hampson et al., 1985; Hampson, 1986a). Changes in ultrastructure of the small intestinal epithelium of weaned pigs also occur, and these are greatest in pigs with severe PWD and a high proportion of ETEC in their intestinal tracts (Kenworthy et al., 1967). Although ETEC were originally thought not to alter small intestinal structure (Moon, 1974), STb has been shown to induce a degree of villus atrophy in piglet intestinal loops (Rose et al., 1986), and this activity might help to explain the mucosal changes seen in the SPF piglets. Further investigation is required into the relationship between liquid weaner diets based on milk and their potential inhibition of proliferation and adhesion of ETEC, and on the possible role of ETEC in causing mucosal changes.

Nutrient composition of the weaner diet, and ability to digest and absorb this material

Numerous studies have been directed at manipulating the nutrient composition of pig weaner diets, and most have been undertaken from the point of view of attempting to optimize growth rates in healthy pigs, rather than examining their influence on PWD. These trials have been conducted under different husbandry conditions, using animals of various breeds and ages, with a variety of different sources, combinations and levels of nutrients used. Not surprisingly, a variety of sometimes conflicting results have been produced. In general, the diet should be tailored to match the ability of the young pig to deal with it.

Digestive capacity of young pigs: A variety of mucosal carbohydrases (Kidder and Manners, 1980) and pancreatic enzyme activities (Corring et al., 1978) are incompletely developed in three-week-old pigs. Furthermore, certain enzyme activities may be reduced for a short period after weaning. For example, Hartmann et al. (1961) found depressed pancreatic lipase and protease activities after weaning at one week of age. In pigs weaned at 16 days of age Efird et al. (1982b) recorded reductions in pancreatic trypsin and chymotrypsin activities, but increases in intestinal chymotrypsin, and a greater proteolytic activity of the stomach compared to unweaned pigs. Lindemann et al. (1986) also found a general depression in pancreatic enzyme activities, but not in gastric proteolytic activity in pigs during the first week following weaning at four weeks of age. Pigs weaned at two, three or five weeks of age also show reductions in some, but not all enterocyte brush-border enzyme activities (Gay et al., 1976; McCracken, 1984; Kelly et al., 1984; Hampson and Kidder, 1986; Miller et al., 1986).

Absorptive capacity of young pigs: There is evidence suggesting that there may also be an overall loss of absorptive capacity in young pigs immediately after early weaning. As previously indicated, small intestinal villus height is usually reduced after weaning, there is a change in villus morphology (Plates 1 and 2), and an increase in crypt depth associated with a more rapid rate of enterocyte turnover (Gay et al., 1976; Kenworthy, 1976; Smith, 1984; Hampson, 1986a; Miller et al., 1986). The early-weaned pig shows a reduced capacity to absorb xylose (Miller et al., 1984a; Hampson and Kidder, 1986; Hampson and Smith, 1986), and villus enterocytes show reduced sodium-dependent alanine uptake (Smith, 1984; Miller et al., 1986). It is therefore not surprising that a “malabsorption syndrome” may be seen in newly-weaned pigs (Kenworthy and Allen, 1966a). The outcome of such changes in digestive and absorptive capacity depend on the total capacity of the intestines to absorb, and on the amount and digestibility of the diet consumed immediately after weaning.
Protein levels and sources in the weaner diet: Three-week-old piglets need a high protein diet if their potential for deposition of lean meat is to be exploited in full. Campbell (1977) found that with a diet containing 15.12 MJ DE/kg, 19% protein gave optimal growth whilst 21.5% gave the best conversion to lean carcass by eight weeks of age. Where PWD is a problem, however, it has been suggested that reducing protein levels in the diet may be beneficial (Bertschinger et al., 1979; Prohaszka and Baron, 1980). Kornegay et al. (1974) noted a non-significant "trend" for faeces to be firmer in weaned pigs fed a 14% protein diet compared to others given 18%, and Namioka and Murata (1965) found that the intestines of pigs fed very high protein levels (29.59%) contained greater numbers of coliform organisms than those fed a lower level (20.86%). Prohaszka and Baron (1980) ascribed the increased susceptibility of pigs weaned onto a "high" protein diet (21%) to an inability to produce sufficient gastric acid to digest the protein. This in turn led to an elevation in gastric pH to a level which permitted selective proliferation of ingested ETEC. Other workers have noted elevated gastric pH values in newly-weaned pigs, but not necessarily associated with consumption of high levels of dietary protein [Thomlinson (1969) in the protected centre of dry-food boluses, and Lawrence (1970) after frequent feeding, or feeding finely-ground barley].

Armstrong and Cline (1976) demonstrated that the amount of protein in the intestinal contents of weaned pigs is related to the level in their diet. It has been suggested that the antigenic nature of this protein may influence the effects of *E. coli* in the pig (Kenworthy, 1970), and be the cause of structural changes in the small intestine, which in turn predispose to PWD (Miller et al., 1984a). The latter possibility will be discussed in the section on "creep food, intestinal structure and hypersensitivity to diet". It should, however, also be pointed out that many workers have found that dietary protein levels have had no influence on either coliform numbers in the intestinal tract or on incidence of PWD (Smith and Jones, 1963; Palmer and Hulland, 1965; Smith and Halls, 1968; Armstrong and Cline, 1976; Armstrong and Clawson, 1980).

The source of dietary protein for weaner diets has also received considerable attention. Products with high digestibility such as dried skim milk or whey have been recommended (Armstrong and Clawson, 1980; Lecece et al, 1985), but these are expensive and have not always proved beneficial (Meade et al., 1965; Kornegay et al., 1974). Pouteaux et al. (1982) compared buttermilk powder, soyabean meal and pea protein concentrate as protein sources for weaner diets, and found that they caused no differences in either faecal water content or incidence of diarrhoea. However, pigs fed herring meal may be more liable to develop PWD (Smith and Halls, 1968).
Bertshinger et al., 1979), and high levels of soya protein may also cause problems (e.g. Anon., 1987). H.S. Chang and S. Tzipori (unpublished data quoted by Tzipori, 1985) noted that soya meal protein adversely affected small intestinal mucosal morphology in weaned pigs challenged with ETEC, and feeding soya protein has been shown to be associated with villus atrophy, crypt hyperplasia and an increased flow of digesta through the small intestine of the calf, possibly as a result of hypersensitivity to globulin components of the soya meal (Kilshaw and Slade, 1982). Pigs fed soya-containing weaner diets usually have poorer growth rates than do those fed milk based ones (Lecce et al., 1985), and this may reflect a poorer digestibility of the soya protein (Wilson and Leibholz, 1981), or possibly the development of intestinal hypersensitivity to this material (Newby et al., 1985). Soya extracts alone may also cause fluid accumulations in piglet ligated small intestinal loops (Nabuurs, 1986), so it is not surprising that high levels of soya protein are unsuitable for newly-weaned pigs.

The number of protein sources used in the diet may also influence PWD, with most authors finding that simple diets result in less diarrhoea than do complex diets which contain many protein sources (Okai et al., 1976; Ball and Aherne, 1982; Etheridge et al., 1984b). However, Meade et al. (1965) found that diet complexity may make little difference to postweaning performance and complex diets have resulted in faster growth rates, mainly because they tend to cause increased consumption of the weaner diet (Bayley and Carlson, 1970).

Energy levels and sources in the weaner diet: Suckling piglets receive approximately 60% of their dietary energy as fat, but they appear less able to utilize lipids after weaning, and relatively low levels are usually incorporated in commercial diets (around five to 10%). Optimal digestible energy levels for such diets are 15.12 MJ/kg where tallow is incorporated at 5.6% (Campbell et al., 1975). The energy source of weaner diets therefore consists mainly of a variety of carbohydrates, of varying complexity and digestibility. Although it has been said that diets that are high in energy may predispose to selective colonization of the small intestine with pathogenic E. coli (Moon, 1974), there unfortunately appears to be little direct evidence available on this topic. O’Grady and Bowland (1972) found that pigs weaned at two weeks of age had lower mortality on a high energy diet compared to a low energy one (16.0 and 11.8 MJ DE/kg, respectively), and inclusion of 5% molasses in the formulation reduced mortality. Tzipori et al. (1980b) found that a high-energy weaner diet contributed to earlier postweaning consumption of the diet, and a delay in onset of PWD compared to pigs fed a commercial diet. Once pigs have adjusted to the weaner diet, their voluntary intake (to 50 kg live weight) is mainly limited by their gut capacity, and not by the energy concentration of the diet (Campbell and Taverner, 1986).

To test the possibility that ETEC are selectively able to utilize excess energy sources in the intestine, Vasenius (1969) orally inoculated weaned pigs with sucrose-fermenting ETEC, with or without dietary sucrose supplements (10%). Pigs receiving the sucrose developed diarrhoea, whilst those not getting it had only transient or no diarrhoea, and had somewhat lower numbers of ETEC in their faeces. These results suggest that the bacteria used the substrate, proliferated and initiated diarrhoea, although another interpretation could be that the sucrose itself started an ‘osmotic’ diarrhoea which in turn facilitated recovery of the ETEC. Even though most E. coli strains from PWD are capable of fermenting sucrose (Larsen, 1976), Palmer and Hulland (1965) found that the presence of 15% sucrose in a weaner ration did not influence proliferation of haemolytic E. coli. Bayley and Carlson (1970) found that supplementing a weaner ration with 5% glucose increased “digestive disturbances”, whilst Armstrong and Cline (1976) noted no effects of 20% or 30% glucose in the diet. In contrast, Entringer et al. (1975) found that grower pigs offered glucose or lactose suffered more diarrhoea than those fed starch, and they ascribed this to the more rapid intestinal passage of the readily digestible disaccharides.

Dietary fibre in the weaner diet: There have been a number of reports indicating that both ETEC proliferation and susceptibility to diarrhoea after weaning may be reduced by adding crude fibre (generally derived from oats or barley) to the weaner diet (Richards and Fraser, 1961; Palmer and Hulland, 1965; Smith and Halls, 1968; Armstrong and Cline, 1976; Bertshinger et al., 1979).
However Rivera et al. (1978) found that adding oat fibre to the diet conferred no benefits either in terms of performance or diarrhoea, and English (1981) found less PWD in pigs fed a highly digestible diet with low fibre content (0.8%) than in those on commercial diets (3% crude fibre).

Assuming that ETEC do have the potential for selective utilization of unabsorbed nutrient in the intestinal lumen (as suggested by Kenworthy and Crabb, 1963, and Moon, 1974), the addition of fibre to the diet could act by both reducing the nutrient density of the diet and by increasing its rate of passage through the intestinal tract (Lawrence, 1983). Alternatively it is known that both the source and level of fibre in the diet may rapidly influence bacterial production of volatile fatty acids (VFAs) in the large intestines of growing pigs (Stanogias and Pearce, 1985). In the pig, VFAs are mainly produced by Bacteroides spp., and these colonize the gut within two days of birth (Smith and Jones, 1963). VFAs constitute an important energy source in the pig, and their absorption in the large intestine may also facilitate water and electrolyte absorption (Crump et al., 1980). Their function in this capacity in the large intestine may be of great importance since this appears to be the major site for water and electrolyte absorption in the weaned pig (Hamilton and Roe, 1977). VFAs may also be beneficial because, in concert with pH and oxidation potential, they may have a suppressive effect on Enterobacteriaceae in the porcine intestine (Prohaszka, 1986). Weaned pigs do appear to have a much more active intestinal bacterial fermentation than do pigs remaining with the sow (Etheridge et al., 1984b). The protective role of fibre could therefore be mediated by its influence on VFA production by the normal intestinal flora. Such a protective role for the normal flora has been speculated upon by Palmer and Hulland (1965), who found greater proliferation of ETEC and more PWD in weaned pigs fed bacitracin than in those not receiving the antimicrobial. Oral chloramphenicol also increases incidence of diarrhoea in weaned pigs orally inoculated with ETEC (Cox et al., 1986), again presumably by suppressing the normal flora.

It should be noted however that Etheridge et al. (1984b) found that weaned pigs fed a corn-soyabean meal diet had greater intestinal bacterial fermentation, more VFA production and more diarrhoea than did animals fed a more digestible diet based on oat-groats and casein. Further work is required to observe the relationship between bacterial fermentation, VFA production and proliferation of ETEC in pigs on various diets over the critical postweaning period.

**Availability and intake of weaner diets:** It has been argued that the often poor performance of piglets immediately after weaning is a reflection of inadequate intake, and that regular feeding of a liquid diet might improve performance without overloading a limited digestive capacity (Leece et al., 1979). This problem of limited digestive and absorptive capacity was demonstrated by Leece et al. (1983) when they infected newly-weaned pigs with rotavirus and ETEC, and fed them either a diet containing 30% protein, 40% lactose and 20% animal fat fed in three equal meals over the day, or the same fed in 24 equal increments. Two other groups received one third of this ration, diluted with water to give similar quantities. The high nutrient intake fed three times a day resulted in the most prolonged PWD, the most persistent shedding of rotavirus, and the greatest colonization with ETEC. A less severe response was found in the high intake group fed in 24 instalments, and the groups receiving the low nutrient intake were least affected. In this experiment rotavirus was presumably contributing to the postweaning malabsorption.

Although these results do not identify what components of the diet were involved, they do suggest that feeding too much highly digestible, good quality nutrient to newly-weaned pigs may predispose to PWD, possibly by encouraging proliferation of ETEC. Feeding small amounts of the diet at regular intervals improved performance, presumably because this feeding method does not overwhelm the limited and temporarily reduced absorptive and digestive capacities of young weaned pigs. Ball and Aherne (1982) also found that pigs fed once a day suffered more diarrhoea than those fed ad libitum, whilst animals given a limit on total intake performed best. The greater incidence of diarrhoea in the time-limited group was ascribed to their more erratic feed intake and a tendency for pigs to gorge themselves. Where pigs on ad libitum intake did develop diarrhoea, this was more severe than in the other groups, and may have reflected large intakes by individual greedy animals. Hampson and Smith (1986) observed that it was individual animals with the greatest intakes of a group that developed PWD (Figure 9), although this effect was not observed.
in later experiments using different diets (Hampson et al., 1988). Over eating may predispose to a temporary gastrointestinal stasis (Ruckebusch and Bueno, 1976), which in turn could favour proliferation of ETEC (Kenworthy and Crabb, 1963). However, although gastric stasis may precede scours in piglets (White et al., 1972), when opium tincture and spasmentral were given to weaned pigs to reduce gastro-intestinal motility, this was not followed by a higher incidence of PWD (Schulze, 1979).

Figure 9. Mean daily dietary intakes in weaned caged pigs ■■, developing diarrhoea (n=18); ●●●●, not developing diarrhoea (n=28) (data from Hampson and Smith, 1986).

Restriction of dietary intake after weaning has been beneficial in treatment or prevention of PWD (Palmer and Hulland, 1965; Smith and Halls, 1968; Svendsen, 1974; Bertshinger et al., 1979), but this procedure is mainly recommended for “cheap” diets of low digestibility; an alternative approach is to offer a better quality weaner diet (English, 1981; Etheridge et al., 1984a; Fowler, 1985). It is generally presumed that restriction of intake or improving the digestibility of the diet results in less unabsorbed substrate being present in the intestine to support the growth of ETEC. Paradoxically, good quality diets based on dried skim milk appear to induce less production of enterocyte brush-border maltase activities after weaning than do those based on uncooked cereals (McCracken, 1984), and restriction of postweaning intake also reduces the normal increases in these enzymes (McCracken and Kelly, 1984). Restriction of postweaning intake may reduce growth rate temporarily, but it does not appear to have any significant effect on growth to 90 kg weight (Ball and Aheme, 1982).

Influences of creep food on PWD

Potential benefits of creep food

Pigs weaned at six to eight weeks of age have traditionally been fed creep food to supplement declining milk production after three weeks of lactation. It is more difficult to justify the use of creep food for pigs weaned at three or four weeks of age, although English (1981) demonstrated that consumption of 610 g of high quality creep food before weaning at four weeks of age improved subsequent postweaning intake and performance. Judging from the weight gains of these pigs, the majority of the creep food was eaten in the last week before weaning; Friend et al. (1970), Okai et al. (1976) and Hampson and Kidder (1986) also found difficulty in getting pigs to eat much
creep food before three weeks of age. Madec and Josse (1983) found that consumption of less than 100 g of creep food before weaning was associated with increased variation in weaning weight, lower weaning weights, more preweaning diarrhoea, and an increased risk of developing PWD.

It therefore appears that even for pigs weaned at three weeks of age, offering creep feed may have some benefit, especially for litters where the sow is lactating poorly or unevenly (Fowler, 1985). Hampson and Kidder (1986) found that piglets fed creep food had a slight, but not significant increase in mean weight, with a smaller spread of weights at 20 days of age. However, brush-border lactase and sucrase activities were not influenced by consumption of creep food up to three weeks of age, and lactase activity declined in creep-fed piglets, but not those denied access to creep food, between 21 and 32 days of age. Friend et al. (1970) reported a 12% increase in pancreatic trypsin concentration in pigs fed creep food between one and three weeks of age, and Shield et al. (1980) found a higher pancreatic amylase but not protease activity in piglets fed creep food before weaning at four weeks of age. Aumaitre (1971) suggested that earlier development of sucrase, maltase, amylase and trypsin activities might be induced by encouraging earlier consumption of non-milk sugars, starch and protein. In general it remains debatable whether consumption of creep food before early weaning can significantly influence digestive development, and thus indirectly potentially reduce susceptibility to PWD.

Creep food, intestinal structure and hypersensitivity to diet

As previously indicated, piglets develop a series of changes in small intestinal structure and enterocyte brush-border enzyme activities immediately after weaning (Gay et al., 1976; Kenworthy, 1976; Smith, 1984; McCracken, 1984; Hampson and Kidder, 1986; Hampson, 1986a; Miller et al., 1986). Villus height can be reduced by 25% within 24 h of weaning, and continues to decline until around five days after weaning, after which time it stabilizes. This response could be initiated by a lack of a continuous supply of substrate in the intestine (McCracken and Kelly, 1984), and could possibly be mediated by a temporary reduction in crypt cell production rate as seen in gnotobiotic pigs weaned onto a dry diet (Byrne and Hall, 1984). However, this may not be the whole explanation. Hampson (1983) starved five pigs for two days after weaning, and compared these with other pigs of the same age which were either offered food after weaning or kept with the sow. The starved pigs did not show the usual postweaning crypt hyperplasia as occurred in the other weaned group, but both weaned groups showed similar significant reductions in height of villi and loss of lactase activity. Thus, consumption of food after weaning is necessary for crypt hyperplasia to occur, but lack of postweaning intake (and hence lack of crypt hyperplasia) may not be necessary for postweaning villus atrophy to occur. Crypt depth normally continues to increase steadily over an 11 day period after weaning, at a much greater rate than that seen in unweaned pigs. Continuous absence from the sow is required for these changes to occur, since pigs weaned for two days and then returned to their dam for three days show crypt elongation only equivalent to that of pigs weaned for two days (Hampson, 1983). The greatest relative elongation in crypts occurs in the distal half of the small intestine, and crypts remain long for five weeks after weaning, and probably remain so throughout adult life (Hampson, 1983). These changes almost certainly reflect an increased rate of enterocyte production and turnover (Al-Mukhtar et al., 1982a). Amongst the brush-border enzymes, the maltases (two and three) increase after weaning, alkaline phosphatase is largely unaffected, lactase declines, and sucrase declines to a minimum by five days after weaning and then increases. Sodium-dependent alanine transport into villus enterocytes is comparatively reduced after weaning at three but not at five weeks of age (Miller et al., 1986).

Such changes may temporarily reduce digestive and absorptive capacity, and may go a long way to explaining the increased susceptibility of the newly-weaned pig to diarrhoea. Changes in relative maturity of villus enterocytes in newly-weaned pigs may also explain their altered susceptibility to enterotoxins. Stevens et al (1972) demonstrated that 38-day-old pigs weaned for three days showed a greater secretory response to both LT and ST than did unweaned pigs of various ages. Furthermore, STa is active in the intestines of neonatal pigs, but not those of weaned piglets, whilst STb is active in both groups, but more active in weaned animals (Burgess et al., 1978;
Morris and Sojka, 1985). Alterations in surface properties of enterocytes after weaning may also influence the adhesion of ETEC; for example enterocytes of the newly-weaned rabbit show development of a receptor for the D-1-5 pili of \textit{E. coli} RDEC, which is an enteric pathogen of the weaned rabbit (Cheney and Boedeker, 1984).

Many mammals appear to undergo increases in crypt cell production rates, increases in sucrase activity and decreases in lactase activity around the third week of life (Moog, 1979). These changes are generally thought to be under the influence of hormones of the adrenal cortex and thyroid gland, and in the young pig successful attempts have been made to induce precocious maturation of certain digestive enzymes by oral treatment with tri-iodothyronine and prednisolone (Bainter and Nemeth, 1982). The more marked alterations which occur within the first five days of weaning pigs when they are three weeks of age are probably superimposed upon normal developmental changes. An understanding of the aetiology of these changes could help in the formulation of control measures for PWD. Suggestions as to all or some of their causes have included the toxic action of bacterial metabolites such as those produced by microbial decarboxylation and deamination of amino acids in the diet (Hill et al., 1970; Kenworthy, 1976), a poor and irregular supply of nutrient after weaning (Kelly et al., 1984), the action of rotaviruses (Lecce et al., 1982), or intestinal hypersensitivity to dietary antigens.

The latter intriguing possibility was raised after Stokes et al. (1981) elicited a delayed-type hypersensitivity response to ovalbumen in the intestines of unweaned pigs, and noted villus atrophy and crypt hyperplasia similar to that normally seen after weaning. Subsequently, these workers formulated an hypothesis that the previously unexplained intestinal changes which occur after weaning are the result of intestinal hypersensitivity to antigenic material in the weaner diet, and obtained evidence supporting this proposition (Miller et al., 1982, 1983, 1984; Newby et al., 1983, 1985; Hampson, 1986b; Bourne, 1986; Bourne et al., 1986; Stokes et al., 1986). They suggested that small intakes of creep food before weaning “primed” hypersensitivity, whilst large intake (> 600g) prevented it by establishing a state of immunological tolerance to the dietary antigen. Abrupt weaning without prior consumption of creep food was also protective, because the pigs were not primed to the dietary antigens. A brief summary of experiments and observations supporting this hypothesis are:

(i) Piglets which were exposed to creep food between seven and 10 days of age (“primed”) developed earlier and more prolonged shedding of ETEC, more diarrhoea, and more depression in their ability to absorb xylose after weaning than did piglets given creep food from seven days until weaning at 21 days of age (“tolerized”), or others given no creep food before weaning (“unprimed”).

(ii) Piglets were given cows’ milk as a supplement from seven days of age, and were then weaned onto diets which had either casein or hydrolyzed-casein as the protein source. The hydrolysed-casein was shown to be considerably less antigenic than the native casein. Only pigs weaned onto the casein diet developed diarrhoea (in the absence of ETEC). When killed 10 days after weaning the pigs on the hydrolyzed-casein diet also had lesser increases in crypt depth at a site 75% along the small intestine, and greater brush-border sucrase activities in the distal small intestine than did pigs fed the casein diet.

(iii) Pigs killed five days after having been abruptly weaned onto a diet in which the protein source was hydrolyzed-casein (hypoallergenic) had shorter crypts (differences significant at sites between 50 and 75 per cent along the small intestinal length) and greater brush-border lactase and sucrase activities than did animals receiving a diet containing casein as the protein source (Figure 10).

(iv) Pigs on a commercial farm where PWD was a problem performed better and had less diarrhoea after creep feed was no longer offered before weaning (abrupt weaning).

(v) Pigs weaned onto a diet containing a full-fat soya protein extract developed, by five days after weaning, a cutaneous delayed hypersensitivity response to soya extract injected intradermally into the ear; this coincided with maximal depression of ability to absorb xylose after weaning. Adequate feeding of the soya antigen before weaning apparently completely abrogated malabsorption, crypt hyperplasia and diarrhoea after weaning.
(vi) In weaned pigs, enterocyte brush-border alkaline phosphatase activity did not decline by five days after weaning, whereas disaccharidase activities did (Miller et al., 1986). This pattern of activity was similar to that seen in the intestine at sites where the gut immune system is activated (Smith, 1985).

(vii) The general observations that pigs weaned at three weeks of age are more susceptible to PWD than those weaned at five weeks. Young pigs may have insufficient time and opportunity to develop tolerance to dietary antigen, and also have limited proteolytic activity of the stomach and pancreas to cleave ingested proteins to small and less antigenic peptides and amino acids.

Figure 10. Crypt depths along the small intestine in 26-day-old piglets; △ - △, unweaned not receiving creep food; ● - ●, weaned for five days onto a diet containing casein as the protein source; ○ - ○, weaned for five days onto a diet containing hydrolyzed-casein as the protein source (data from Hampson, 1986b). Error bar = 1 sd.

These results and observations have generated considerable interest, but some experiments can be criticized because of a lack of information about relative food intakes on the various treatments, and as yet there have been no direct measurements of hypersensitivity in the small intestine. This latter information may be difficult to acquire since the only indicators of intestinal cell-mediated hypersensitivity reactions may be increases in intra-epithelial lymphocyte numbers and increased rates of cell production, which are mediated by lymphokines (Mowat and Ferguson, 1981). Another problem is the lability of the small intestine, which adapts readily and rapidly in only a limited number of ways to a variety of different stimuli, whether they be microbiological, hormonal or immunological.

As regards the experiments with hydrolyzed-casein diets, other possible explanations for their protective effect on the intestinal mucosa besides reduced antigenicity must be considered. For example, hydrolyzed-casein is known to be more readily digestible than is casein, and less is likely to reach the distal small intestine and colon (Morin et al., 1980). Hence if small peptides and amino acids in the intestinal lumen were able to stimulate crypt cell production directly (the luminal nutrition hypothesis; Diamond and Karasov, 1983), a similar outcome would result. Alternatively a hydrolyzed-casein diet could effect other aspects of intestinal physiology. For example, the partial hydrolysis of both milk and soya-proteins in the diet has resulted in reductions in trypsin and chymotrypsin activities in the duodenum of pigs to which the diets have been fed (Leibholz, 1981); diets containing unhydrolyzed soya-protein, which result in poor performance after weaning, may do the reverse and increase trypsin and chymotrypsin activities in the intestinal contents (Efird, 1982a). Since pancreato-biliary secretions are believed to have trophic effects on the gastro-
intestinal mucosa (Altmann, 1971), the extent of stimulation of their release could be another mechanism by which different diets affect the mucosa after weaning. Another possible explanation for the results could be that the hydrolyzed-casein diets stimulated less release of enteroglucagon, which is a peptide-hormone acting on the intestinal mucosa. Food reaching the lower ileum stimulates release of this peptide, which in turn increases crypt cell production rate (Al- Mukhtar et al., 1982b). A final possibility for the results obtained with the hydrolyzed-casein diets could involve insufficient substrate arriving in the distal small intestine for stimulation of "excess" production of bacterial products such as polyamines, which might normally stimulate enterocyte turnover (Hill et al., 1970). The latter explanation seems unlikely however, since oral te tracycline treatment to suppress the normal microbial flora did not prevent crypt hyperplasia in weaned pigs (Hampson, 1986b).

Other workers have cast doubt on the hypothesis of hypersensitivity to dietary antigens, at least as it stands at present. Fowler and Fraser (1985) used eight different preweaning feeding strategies in an attempt to either sensitize or tolerize piglets to their weaner diet, however the only treatment difference was a tendency for the abruptly weaned pigs to have diarrhoea for longer than any of those given creep food before weaning. Kelly et al. (1986) found that abrupt weaning did tend to lessen the severity of PWD, although a large creep intake before weaning at two weeks of age was not protective. Byrne and Hall (1984) "weaned" germ-free piglets onto a solid diet at 23 days of age, after having prefed one group with 45 g of this material at 10-12 days of age ("priming"). Both weaned groups then showed decreased crypt cell production rates compared with milk-fed controls. The primed piglets developed low serum passive haemagglutination titres to soyabean antigens, and had higher numbers of immunoglobulin-containing cells in their lamina propria, but there was no evidence of a hypersensitivity response in their small intestines. The lack of crypt hyperplasia in the "weaned" germ-free animals, whether or not they were primed, suggests that a normal intestinal flora is necessary for mucosal changes to occur. This flora could act simply by increasing overall immune reactivity in the gut. Exposure of germ-free pigs to the normal microbial flora of a piggery is itself sufficient to cause villus atrophy and increased rates of crypt cell production (Kenworthy and Allen, 1966b).

In studies of the small intestinal mucosa and its brush-border enzyme activities in pigs after weaning, no differences were seen between groups of pigs which were either abruptly weaned or offered creep food before weaning (Hampson and Kidder, 1986; Hampson, 1986a). In other experiments where piglets were fed various levels of creep food before weaning, no postweaning differences were seen between groups in growth rate, excretion of ETEC or rotavirus, or in ability to absorb xylose (Hampson and Smith, 1986). PWD did not occur in abruptly weaned pigs, but this was associated with small postweaning dietary intakes on this regimen. In a further experiment in which pigs were either not offered creep food, or forcibly "primed" with one of two diets before weaning, group effects were again not seen in any of the above parameters, nor in villus height or crypt depth in the small intestine (Hampson et al., 1988). These results suggest that feeding creep food does not influence intestinal changes occurring after exposure to the weaner diet, and therefore suggests that the hypersensitivity hypothesis is incorrect, or requires modification.

In summary, whilst changes in small intestinal mucosal structure and enzyme activities after weaning seem likely to be involved in predisposing to PWD, there still remains doubt as to their precise aetiology. Although diet is important, work with germ-free gnotobiotic piglets suggests that a normal flora is necessary before a change of diet will stimulate intestinal changes, even if there is "priming" with creep food. Evidence for the intestinal reactions being "primed" by creep food in conventional pigs is equivocal. These results do not exclude the possibility of an interaction between diet and normal flora causing changes directly (as suggested by Kenworthy, 1976), and such changes could be mediated by immunological reactions to both components. The decline in villus height to crypt depth ratio ceases around five days after weaning, stabilizes (Figure 11), and then is followed by a "normal" postweaning increase in sucrase activity (Hampson and Kidder, 1986; Hampson, 1986a). A lack of reversion to preweaning values suggests that there is a stable transition to an 'adult-type' intestine after a brief hiatus associated with and exaggerated by weaning at an early age. As pointed out by Bourne (1986), this stable adult pattern could result from
a continuous low-level exposure to new antigenic dietary material in the lumen of the intestine.

Figure 11. Villus height to crypt depth ratio at a site 25% along the small intestine in pigs killed between 21 and 32 days of age; 0-0, unweaned, offered creep food; □-□, unweaned, not offered creep food; ○-○, offered creep food before weaning at 21 days of age; ■-■, not offered creep food before weaning at 21 days of age (data from Hampson, 1986a).

CONCLUSIONS AND PRACTICAL IMPLICATIONS

PWD is a complex disease, essentially caused by the activity of certain strains of ETEC which proliferate in the small intestine after weaning. Very occasionally diarrhoea occurs in the absence of ETEC and other infectious agents, but it is usually mild and probably associated with malabsorption and/or changes in gastrointestinal motility. The ETEC which can cause PWD are certainly present in many piggeries, yet the occurrence of disease is very variable. There are many potential risk factors associated with the development of PWD, and where disease occurs all of these should be examined, and improvements made where possible.

Various aspects of the weaner diet can influence susceptibility to PWD. It should be borne in mind that young weaned pigs have a digestive system that is far from mature, with temporary reductions in this limited digestive and absorptive capacity occurring in the immediate post-weaning period. Under these circumstances it is not surprising that presenting large quantities of poorly digestible diet can predispose to diarrhoea. If the diet is not of high digestibility and there is reluctance to improve it, it appears that restriction of access to it is the most effective means of control of PWD. Improved growth rates can be obtained if the diet is highly digestible, milk-based and (ideally) fed in liquid form at regular intervals. Paradoxically, if PWD becomes a problem with such a diet because of overconsumption, then addition of some fibre may be beneficial. At present it would seem best to avoid high levels of non-milk sugars and of low quality protein in the weaner diet, and especially to be cautious with the use of soya protein in the first 10 days after weaning. If some form of hypersensitivity to diet (manifest as increased crypt cell production rates) does prove to be an inevitable manifestation of consuming an adult-type diet, it would seem sensible to avoid highly antigenic dietary material in the critical period immediately after weaning. As regards creep feeding, this procedure is probably not justified for pigs weaned at three weeks of age; creep food is expensive, largely wasted and appears to give little physiological response by three weeks of age. Its potential for priming intestinal hypersensitivity, although far from proven, must also be considered.

Further investigation is required into the relationship between form and type of diet at
weaning, proliferation of ETEC and occurrence of PWD. A better understanding of the condition will help when devising control measures, and the information may have wider relevance for reducing severity of other enteric conditions, improving utilization of the diet, and increasing growth rates. In particular more needs to be known about:

(i) The possible role of hypersensitivity to dietary antigens in predisposing to PWD.

(ii) Whether or not the small intestinal mucosal changes which occur after weaning can be modified by practical diets or dietary regimens.

(iii) If and how it is that intestinal structural changes occurring after weaning predispose to proliferation of ETEC (e.g. appearance of new receptors for the bacteria; malabsorption and selective use of unabsorbed substrate by ETEC etc).

(iv) Whether or not the normal intestinal bacterial flora has a role in mediating intestinal structural changes and/or regulating ETEC numbers after weaning.

(v) The mechanisms by which weaner diets which are based on cows’ milk fed in liquid form may act to inhibit proliferation and adhesion of ETEC.