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DIETARY REGULATION OF INTESTINAL GENE EXPRESSION

I. R. Sanderson and S. Naik
Department of Paediatric Gastroenterology, St Bartholomew's and the Royal London School of Medicine and Dentistry, London EC1A 7BE, United Kingdom; e-mail: I.R.Sanderson@mds.qmw.ac.uk; s.k.naik@mds.qmw.ac.uk

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■ **Abstract** We are becoming increasingly aware of inherited genetic abnormalities as causes of disease. However, alterations in gene expression can also contribute to other disease processes. Recently it has been suggested that our environment may alter such genes and thus be a direct influence on disease. Diet is a potent mechanism for altering the environment of cells of most organs, particularly the gastrointestinal tract. This review addresses the influence of nutritional factors on intestinal gene regulation. These influences include insulin, which is not a dietary component but responds to dietary changes, and butyrate, a short chain fatty acid produced by normal intestinal flora. Manipulation of diet may be a means of treating intestinal disorders. Nutritional treatment therefore is also discussed in the light of its effect on gene expression.

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INTRODUCTION

Part of the increasing advances in molecular biology includes the study of the regulation of gene expression in health and disease. The importance of gene expression is not restricted to inherited disorders but includes many other disease processes. New forms of treatment could develop if the relationship between disease processes and the ability to alter gene expression were more fully understood. Vectors to insert new genetic material into cells to alter gene expression are under active investigation. However, other means of altering gene expression by altering the molecular environment surrounding the cell remain to be fully explained. The greatest influence on the environment of the intestinal epithelium is diet. Understanding how dietary changes influence intestinal gene regulation may therefore result in the use of nutrition as a therapeutic tool of the future.

In bacteria, the *lac* operon (30) is a prime example of how nutritional changes alter gene expression. Adding lactose to bacterial culture media results in the induction of proteins that transport and hydrolyze lactose. Similarly, in *Escherichia coli*, lack of tryptophan was shown to lead to up-regulation of the genes that handle tryptophan, the *tryp* operon (60). Studies of nutrient-gene regulation in bacteria and yeasts are in abundance. In higher-functioning organisms, the situation is different. The mechanisms controlling gene expression are more complex, and the components of nutrition that may be responsible need to be identified. In addition, other factors may affect the luminal environment, thereby influencing such intestinal gene expression as hormones, growth factors, microbial flora, and antigens.

The luminal environment constantly changes with diet. The epithelial cell of the intestine is the first to encounter such changes, and so in this review, attention is focused on how nutrition may alter gene expression in the enterocyte. We also include the influence of insulin, which—although not a dietary nutrient—responds acutely to dietary changes, and butyrate, a short-chain fatty acid produced by normal intestinal flora.

PATHOPHYSIOLOGY

The regulation of gene expression by nutrition requires two mechanisms. First, enzymes or receptors within the enterocyte must recognize the nutrient. This results in a sequence of events that ultimately alters the transcription or translation of genes. Changes in the intestinal lumen may directly affect the apical aspect of the epithelial cell or act indirectly via hormones, growth factors, and cytokines (Figure 1). Pathways connecting dietary alterations to changes in gene expression are incompletely understood and are currently under investigation.

Dietary alterations may have effects on several genes mediated through many different pathways (Figure 2). Protein expression may be due not only to single

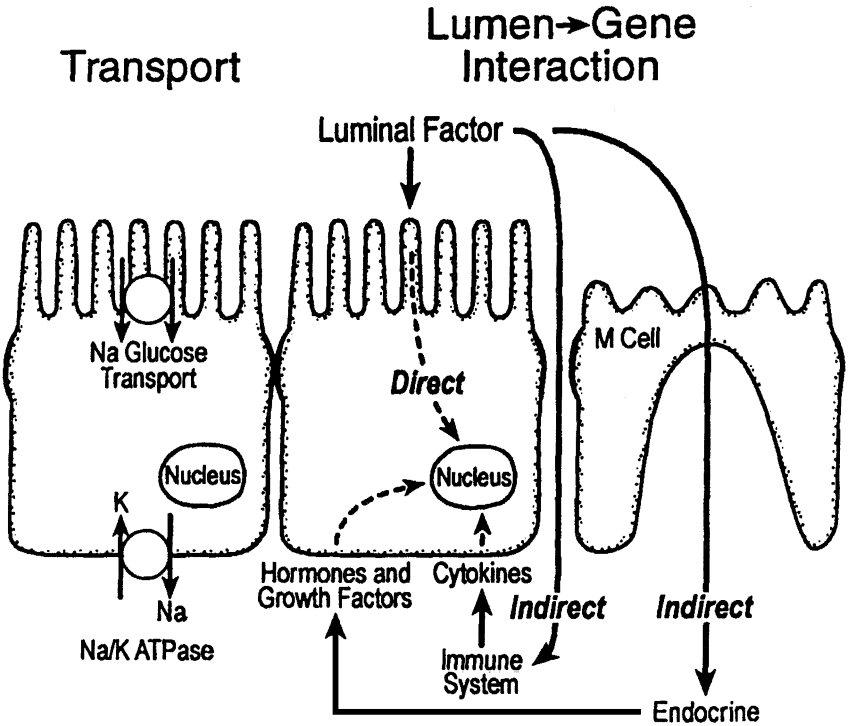

Figure 1 Dietary changes may affect gene expression directly (through the apical membrane) or indirectly (through the basolateral membrane).

gene induction in cells along the crypt-villus axis but also to alteration in the state of differentiation of epithelial cells, with gene expression being a part of these changes. Studies have shown that the rate of epithelial proliferation and differentiation are determined by luminal contents (49, 73). Also, alterations in luminal content may not affect the epithelium already formed but may induce a new lineage of cells to develop that express a gene arising from the crypt. In Crohn's disease, there is evidence that alongside ulcers, a cell lineage develops that secretes epidermal growth factor locally, which in turn enhances epithelial cell proliferation and promotes reconstitution of the epithelial barrier (142). Elucidating these changes and their mechanism of action may lead to therapeutic dietary manipulation.

Traditionally, it has been assumed that the expression of genes in the small intestine is preprogrammed and not influenced by events in the intestinal lumen. An alteration in epithelial cell phenotype secondary to nutritional factors would have several advantages. First, the intestine could adapt to absorb nutrients more effectively if specific digestive enzymes and transporters were up-regulated by the repeated intake of a particular nutrient (discussed in detail later). Second, as all mammals are fed from mother's milk, there is an opportunity for breast milk to influence the development of the epithelium through actions of its own constituents.

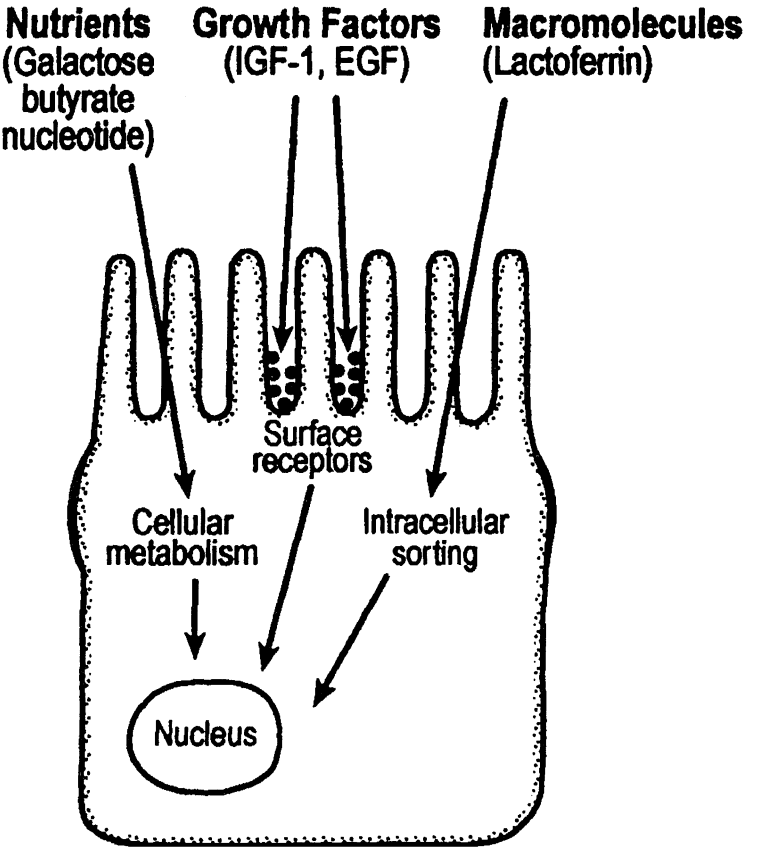

Figure 2 Cross talk through the apical membrane depends on the particular signal involved. IGF, Insulin-like growth factor; EGF, epidermal growth factor.

Third, if genes affected in the epithelium were immunologically important, the intestine could influence mucosal immune responses by signaling information to the mucosal immune system and beyond through changes in the expression of epithelial cell genes. Each of these areas is likely to represent important physiological mechanisms that have implications for child health.

BREASTFEEDING

Breast milk provides nutrition and is immunoprotective to the newborn. Because the intestinal epithelial cells are the first to encounter breast milk, it is reasonable to suppose that breast milk may influence the development of these cells. Several factors in milk can affect gene expression of the enterocyte. Growth factors such as epidermal growth factor have increased brush border enzyme activity and affect growth and development of the enterocyte. There is a greater effect in the fetus

and newborn than in the developed intestine (6, 17, 140). Other components of breast milk have also been shown to affect the same genes (55). Research into the exact mechanisms involved is currently under way.

One factor that has received much attention is lactoferrin. This is a glycoprotein found in high concentrations in breast milk, particularly colostrum. Similar in structure to transferrin, it is present in neutrophil granules, but in plasma its concentration is low in comparison to transferrin. The actions of lactoferrin are relevant to infant feeding practices because some artificial infant milk formulas now contain added bovine lactoferrin.

Specific lactoferrin binding sites have been identified in the intestinal brush border membrane of several species: a human fetus (65), rhesus monkeys (26), rabbits (81), and mice (59). Lactoferrin is resistant to the action of proteolytic enzymes (12, 25), and so a direct biological effect on the intestinal epithelium is a possibility. Cox et al (21) suggest that the major role of lactoferrin is iron delivery to the intestinal epithelium. In cultured epithelial cells depleted of iron, the lactoferrin binding sites actually increase in number, thereby specifically increasing the amount of iron transported into enterocytes (85).

In addition to increasing the bioavailability of iron in the intestine, studies in different cell lines have suggested that lactoferrin acts as a proliferative factor. These include human lymphocytes (53), mouse embryo fibroblasts, rat myoblasts (14), human colon adenocarcinoma cells (HT 29) (1), and adult rat crypt cells (90). Lactoferrin can also influence brush border enzyme activity. Oguchi et al (96) showed that the effect on sucrase and alkaline phosphatase activity depended on whether lactoferrin was saturated with iron.

Another suggested role for lactoferrin in vivo is to function as a scavenger for iron (and perhaps other substances) to prevent unbound iron from causing free radical–mediated damage to the tissue (110). This is an indirect effect on intestinal epithelial gene expression because if left unscavenged, these luminal factors could directly affect enterocyte gene expression. An example is the binding of lactoferrin to lipopolysaccharide, a major component of bacterial cell walls, which modifies its effect on enterocytes (86). However, a purely luminal role is not supported by the presence of specific receptors in the brush border membrane. Direct effects should be mediated through these receptors. The human lactoferrin receptor has now been cloned and sequenced (74). Research using transfected cell lines with lactoferrin DNA promoter response elements will help to determine whether the lactoferrin receptor does signal directly to the enterocyte.

Lactoferrin is secreted by neutrophils. Its effect on gene expression of cells of the immune system has been studied. This may give further insight into its mode of action in breast milk on enterocytes. Lactoferrin binds avidly to DNA in vitro (4, 5). It can also be taken up by leukemic cells and appear in the nuclei bound to DNA (43). He & Furmanski (54) determined which nucleotide sequences of DNA specifically bind to lactoferrin and quantified the binding of iron, which was greater in saturated than unsaturated lactoferrin. These sequences were added to an artificial gene promoter attached to a reporter gene and transfected into leukemic cells. The expression of the reporter gene was up-regulated by the addition of lactoferrin

to the culture media and to a greater degree with iron-saturated lactoferrin. The possibility exists, therefore, that lactoferrin from breast milk can cross into an infant's intestinal epithelium via its receptor, enter the nucleus, and program the cell. Lactoferrin does affect the proliferation and differentiation of intestinal epithelial cells (96), and its actions depend on the degree of iron saturation. These enterocyte gene expression changes may be mediated by mechanisms similar to those shown by He & Furmanski (54). Future research will help to resolve this issue.

Maternal diet influences the composition of breast milk. These changes could then lead to subsequent changes in the enterocyte. Several animal studies have elegantly demonstrated this effect. Jarocka-Cyrta et al (63) altered the lipid content of pregnant rats during gestation and suckling. At birth, the diet was continued or crossed over. Differences were seen in jejunal and ileal uptake of fatty acids, cholesterol, fructose, and glucose and in weight gain. Perin et al (100) altered maternal diet in rats during nursing. The same diet was given to weanlings and older rats. The three groups were compared by examining uptake of fatty acids, cholesterol, fructose, and glucose. Jejunal uptake of fatty acids was greater in sucklings. Different adaptive patterns between jejunum and ileum were noted in sucklings, and different patterns of adaptation between sucklings and weanlings in response to dietary lipid alterations were seen. These findings could not be explained by age- or diet-associated alterations in villus height. They emphasized the important effect dietary lipids had on development of the intestine. In this review, we first address the underlying changes in intestinal gene expression and return to intestinal development when discussing the effects of individual components of nutrition on enterocyte gene expression later.

SIGNALING FROM THE INTESTINAL LUMEN TO THE MUCOSAL IMMUNE SYSTEM

The intestine absorbs nutrients and at the same time functions as a barrier to the external environment. The mucosal immune system plays an important role in maintaining this balance. The epithelium has traditionally been thought to function merely as a passive barrier with areas of selective filtration. The notion that it could respond to luminal contents and signal their presence to the mucosal immune system is a relatively new concept. Nutrient transporters and disaccharidases are dependent on genes whose actions are restricted to the epithelial cell monolayer. Changes in their expression in response to an altered luminal environment are a form of cellular adaptation (Figure 3). However, when the epithelial cell secretes proteins whose actions go beyond the boundary of the cell, it becomes a mediator between luminal contents and target cells outside the epithelial barrier, particularly immunocytes in the mucosa. There is evidence that the intestinal epithelium can secrete cytokines (124), which directly alter immune responses. In addition, growth factors and their binding proteins can affect proliferation of immune cells and the enterocyte can act as an antigen-presenting cell itself.

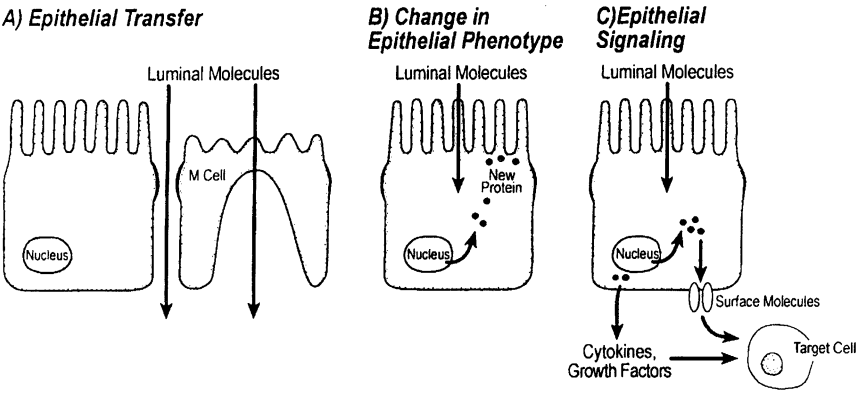

Figure 3 Possible interactions of molecules from the intestinal lumen with the epithelium. Traditionally, the epithelium has been seen as a seletive barrier to molecules, admitting those required for energy intake or immunosurveillance and excluding others (*A*). However, nutrients can alter the phenotype of epithelium to adapt to changing nutritional needs (*B*). When changes in the intestinal lumen induce new proteins that interact with mucosal immune cells, the epithelium can act as a membrane signaling information from the lumen to the immune system (*C*).

Regulation of epithelial cell genes that control the mucosal immune system enable luminal factors to influence immune responses while maintaining full epithelial barrier integrity. Although the absorption of luminal factors is an important method by which the mucosal immune system surveys the intestinal lumen, it requires the entry of molecules through the epithelial barrier. This provides a potential source for invasion by pathogens. The poliovirus, for example, enters by this means, exploiting the normal uptake of macromolecules. The ability of the epithelium to signal information to the mucosal immune system without the need for a physical pathway from the lumen to the serosal surface offers obvious advantages in terms of epithelial protection.

Mucosal immune responses play a major role in gastrointestinal diseases. Therefore, if the epithelium modifies the immune response according to the luminal environment, dietary alterations could be used therapeutically to manipulate the luminal environment, as in the use of enteral feeds for Crohn's disease (discussed later).

GENES OF IMMUNOLOGICAL IMPORTANCE IN THE INTESTINAL EPITHELIUM

Many studies now confirm that the intestinal epithelium can synthesize several immunologically active proteins. These include cytokines (33, 83, 106, 97, 118, 143), growth factors and binding proteins (22, 23, 120), and surface molecules. Human

diseases are associated with the increased production of some of these proteins, either in the circulation or in the gastrointestinal tract (77, 105). Dietary factors alter the expression of some of these proteins by the enterocyte.

Cytokines

Chemokines are a family of chemotactic cytokines that mediate migration and adhesion and activation of granulocytes and macrophages. They are low-molecular-weight, basic heparin-binding proteins divided into two large subgroups according to structural differences and chromosomal location. The α chemokines, clustered on chromosome 4, have an amino acid between the first two cysteines (CXC proteins) whereas the β family, located on chromosome 17, do not (CC proteins). Up-regulation of these chemokines is by stimulation of cytokines, such as interleukin (IL)-1, tumor necrosis factor, lipopolysaccharide, IL-6, or γ interferon (19, 139). Dexamethasone is a strong down-regulator (135).

In the intestine, many cell types can secrete chemokines; these include monocytes and macrophages, endothelial cells, fibroblasts, and epithelial cells (3, 128, 144). IL-8, an α chemokine, is a potent neutrophil attractant. In inflammatory bowel disease there is a marked neutrophil invasion, and increased levels of IL-8 have been documented (61, 78). Monocyte-chemoattractant protein 1, a β chemokine, attracts monocytes and macrophages and is constitutively expressed by intestinal epithelial cells. This is also up-regulated in inflammatory bowel disease (106). There is evidence emerging that these cytokines can be altered by dietary factors. Butyrate is a short-chain fatty acid produced by the normal gut flora. Levels vary according to the bacteria present and the type of diet. Formula-fed infants produce more butyrate in their stools compared with breast-fed infants, where the predominant fatty acid is acetic acid (70). Adult levels are reached by the age of 2 years (84).

IL-8 is not constitutively expressed by the enterocyte. Butyrate further enhances the production of IL-8 following stimulation with proinflammatory agents (IL-1, lipopolysaccharides) in intestinal epithelial cell lines. It is interesting to note that the same group found a simultaneous down-regulation of monocyte-chemoattractant protein 1. (42). The mechanism of action of butyrate appears to involve histone acetylation (discussed in further detail below). In rodents, the equivalent cytokine to IL-8 is macrophage inflammatory protein 2. Similar studies using cell lines derived from rat intestinal epithelium have also demonstrated an increase in macrophage inflammatory protein 2 in response to lipopolysaccharide and butyrate (97). It appears that during inflammation, butyrate alters both the number and type of cells in the gut mucosa, through changes in chemokine production of epithelial cells. If in vivo studies confirm this finding, dietary treatment to optimize butyrate levels and thus alter chemokine expression in the gut may become a feasible option for treatment. Enteral nutrition is an accepted treatment for pediatric inflammatory bowel disease (113). Breese et al showed a reduction

in γ interferon and IL-2 in the mucosa following enteral nutrition (9). Whether this effect was direct or mediated through changes in the enterocyte, possibly in relation to butyrate levels, needs further evaluation.

Class II Major Histocompatibility Complex and Invariant Chain Expression

To elicit an immune response, antigens require processing and presentation to T cells by antigen-presenting cells. Common antigen-presenting cells are macrophages, dendritic cells, B-lymphocytes, and Langerhans cells. The molecules that present the processed antigen to the T cell receptor are class II major histocompatibility complex (MHC) heterodimers (Ia in rodents). Other cells express class II MHC, including intestinal epithelial cells, which may function as antigen-presenting cells. Hershberg et al (56) used tetanus toxoid and HLA-DR–restricted T cells to show that cultured human enterocytes directed to express HLA-DR could initiate class II processing after internalization of antigen from the apical surface. CIITA, a class II transactivator, stimulates class II processing without altering barrier function of the enterocyte that is usually seen following stimulation with γ interferon. Here coexpression of CIITA facilitated antigen processing from the basolateral side.

Exogenous antigens require the presence of another protein for efficient processing and presentation, the invariant (Ii) chain. In mouse intestinal epithelium, diet strongly affects the expression of class II MHC and invariant chain. These molecules are expressed from before birth in the lamina propria but appear to be developmentally regulated in the epithelium (111). Their expression can be altered, by delaying weaning from mother's milk to normal chow. Three to four days after weaning onto normal chow, the mouse intestinal epithelium expresses both invariant chain and class II MHC. However, weaning onto an elemental diet similar to that used therapeutically in Crohn's disease did not induce expression of these molecules or their transcripts (112). Clearly, dietary manipulation influenced gene expression in the intestinal epithelium in vivo, again supporting the idea that alterations in gene expression may be a tool for treating disease.

Growth Factors

Growth in children is related to both nutritional intake and underlying disease. It is well documented that in developed countries, children in immigrant communities grow taller than their parents. In inflammatory diseases of childhood, growth is related not only to the degree of inflammation but also to poor nutritional intake. Growth hormone does not correlate directly with linear growth. Insulin-like growth factor (IGF)-1 is an important mediator of the effects of growth hormone and is thought to be mainly responsible for nutrient control of growth in health and disease (66, 75, 122).

In extracellular fluids, IGF-1 is bound to IGF-binding proteins (IGFBPs), six of which have been cloned (18). There is an inverse correlation between IGFBPs and IGF-1. Nutritional factors alter circulating levels of IGFBPs. Protein restriction increases IGFBP-1 levels, which fall on refeeding. IGFBPs are secreted mainly by Kupffer cells (138), whereas IGF-1 is synthesized by hepatocytes. Because IGFBP-1 is also sensitive to insulin, the direct effects of nutrition on its expression are unclear.

Immune responses depend on activation of T lymphocytes and on an ability to proliferate. Activated T and B cells have receptors for IGF-1. IGF-1 increases T and B cell proliferation and is chemotactic for activated T cells. Human intestinal epithelial cells secrete IGFBPs (94, 95). Through this mechanism, the enterocyte may influence proliferation of activated mucosal lymphocytes. In vitro studies have shown that nutritional factors can alter the production of IGFBPs. Butyrate increases IGFBP-2, which avidly binds to IGF-2, but down-regulates IGFBP-3, the molecule binding most strongly to IGF-1 (91). These changes correlate with IGFBP mRNA levels.

Staley et al (125) examined the effects of an IGF-1 analogue on intestinal growth. Newborn rat pups were fed either rat milk replacer or replacer with added IGF-1 analogue or they were maternally fed. After 3 days, differences were noted in villi height, cell count, IGF levels, and IGFBP levels, demonstrating an effect of nutrition on gene expression. Nutrition can, therefore, influence IGF/IGFBP secretion by the liver into the circulation, which results in growth changes of the whole individual (127), but it may also affect the IGF/IGFBP system in the epithelium and lead to proliferative changes of the local immune system.

Epidermal growth factor (EGF) enhances growth and differentiation of intestinal epithelial cells in vitro and in vivo (49, 69, 137). There is evidence of interaction between EGF and IGF in the intestine. EGF induces brush border enzyme activity in several species, including rabbits, rats, and piglets (40, 98). The increased enzyme activity could result from an actual increase in the number of enterocytes that can express these enzymes, i.e. cell proliferation or a more direct effect. EGF acts synergistically with carbohydrates and glucocorticoids to increase sucrase activity (103). Studies administering EGF to rats have shown an increase in DNA synthesis in the small intestine, particularly during suckling (20, 102). The underlying signaling pathways of EGF are not well understood but they appear to involve several protein kinases, in particular mitogen-activated protein kinase. EGF has a role to play in intestinal adaptation, signaling to the mucosal immune system and influencing enterocyte development (due to its presence in breast milk). Elucidating the pathways involved will contribute to the overall understanding of how EGF influences intestinal gene expression.

Vascular endothelial growth factor has recently been found in high concentrations in breast milk. It is interesting to note that a specific receptor has been identified on intestinal epithelium (119). Its role and influence by nutrients will further link breast milk actions to changes in the diet.

THE ROLE OF DIETARY NUTRIENTS

The effect of luminal contents on gene expression is multifactorial, as previously mentioned. A normal diet consists of varying proportions of carbohydrate, protein, fats, vitamins, and minerals. We therefore look at these components to examine their individual influence on intestinal gene expression. Several animal-based studies in particular have found that dietary modifications alter intestinal gene expression of various substances, but the exact underlying mechanisms in many cases have yet to be fully elucidated.

Carbohydrate

Glucose enters the epithelium by active transport via a sodium-coupled glucose transporter, SGLT1. Sodium-coupled glucose transporters represent part of a family of proteins that carries nutrients into a cell across concentration gradients. The activity of some transporters is increased by the presence of their substrate in the diet (28, 29, 36, 121). In bacteria, transporter activity (such as that of the lac operon) increases 100- to 1000-fold, whereas in mice, the figure for inducible transporters increases at most two- to threefold. In mice, an increase in dietary carbohydrate resulted in an enhanced expression of the glucose transporter, and changes were seen in the crypts before the villi (35, 37, 38). On stopping dietary carbohydrate, again the reduction in transporter was seen first in the crypts. This implies that the changes in gene expression were effected by new cell lineages from stem cells. It is the stem cell producing the new cell lineage that therefore must contain the machinery for detection of carbohydrate. The resulting cell population that ascends along the crypt-villus axis expresses this change.

Sucrose-isomaltase (SI) is a membrane-anchored hydrolase transported to the apical aspect of the epithelial cell, where it is split into two subunits by a pancreatic protease. It is essential for the digestion of sucrose and the terminal digestion of starch. In rats, no detectable SI activity appears in the intestine during the first 2 weeks postnatally, but it appears at weaning. Adult levels are reached by 4 weeks (72). Sebastio et al (115, 116) demonstrated a similar developmental profile for sucrase activity and sucrase-isomaltase mRNA in rabbits and human embryos, which suggests a primary site of control at the level of transcription. The expression of SI is affected not only by the ingestion of sucrose but also by EGF and glucocorticoids. Emvo et al (34) investigated the effects of hydrocortisone, EGF, and sucrose alone or in combination on the expression of sucrase activity and SI mRNA in normal and adrenalectomized rats. Each factor alone was able to increase sucrase activity and SI gene expression, with further enhancement on combining the factors. The difference between normal and adrenalectamized rats demonstrated an effect independent of endogenous glucocorticoids.

Lactase phlorizin hydrolase (LPH) is expressed in the small intestine of mammals. During suckling, levels of lactase are high; on weaning, levels fall. Goda

et al (47) fed 6-week-old rats either low-starch diets, containing long-chain or medium-chain triglycerides, or high-starch diets and found higher LPH mRNA levels and LPH protein synthesis in the high-starch group. They also found a lower lactase activity in the long-chain triglycerides group compared with the medium-chain triglyceride group. After, a 12-h force-feeding of sucrose but not α-methylglucoside, a nonmetabolizable sugar, enhanced LPH mRNA levels (47). Subsequently, Tanaka et al (132) have shown that the increase of LPH mRNA was due to alterations at the transcriptional level. They fed 7-week-old rats a low-carbohydrate diet (5.5% of energy as cornstarch) for 7 days. This diet was then continued for 12 h and compared with a diet containing varying amounts of glucose, fructose, sucrose, galactose, and α-methylglucoside. Lactase activity in the brush border membrane and in jejunal homogenates was measured. Transcription rates were measured using an intron probe and nuclear run on assays. Glucose, fructose, sucrose, galactose, and glycerol all increased lactase activity in the jejunal homogenates and brush border membranes and LPH mRNA, but only fructose increased the rate of LPH gene transcription.

Glucose-6-phosphate is an enzyme that plays an important role in glucose homeostasis. Chatelain et al (15) have shown that levels of glucose-6-phosphatase mRNA were present in liver, kidney, and small intestine of suckling rats but not in stomach, adipose tissue, heart, brain, or muscle. Jejunal glucose-6-phosphatase increased in the first 5 days after birth and then gradually fell. Weaning onto a low-fat, high-carbohydrate diet had no effect on jejunal levels but markedly increased liver glucose-6-phosphatase mRNA.

Other studies have examined the effect of glucose on glucose-dependent insulinotropic peptide in rats. Both glucose and fat increased intestinal mRNA expression and fasting decreased levels. The changes appear to be at a pretranslational level (57).

GLUT-5 is a transporter that enables fructose to be absorbed from the small intestine. Diets with a high fructose content up-regulate intestinal GLUT-5 mRNA levels and thus increase the rate of transport of fructose. This increase has been shown to be due to luminal fructose and not to circulating levels of fructose or its metabolites (24).

In diabetics, increasing fiber intake improves glycemic control. Interest has therefore arisen in the effect of additional fiber on glucose transport and glucose homeostasis. Proglucagon is a polypeptide synthesized by L cells in the distal ileum and processed in the intestine to glucagon-like peptides (GLP). GLP-1 stimulates insulin secretion, inhibits glucagon secretion, and delays gastric emptying. GLP-2 is involved with GLUT-2 secretion and epithelial cell proliferation. A high-fiber diet increases levels of GLP-1 and ileal proglucagon mRNA in rats and dogs (79, 104). These changes may be related to microbial fermentation of the fiber. Tappenden et al (133) have shown that supplementing total parenteral nutrition with short-chain fatty acids in rats with 80% bowel resection led to a significant rise in ileal proglucagon mRNA, GLUT-2 mRNA, mucosal mass, and ileal uptake of glucose.

Protein

Few studies have examined the direct effects of different dietary protein concentrations on intestinal gene expression. However, protein restriction strongly correlates with growth failure, so attention has been paid to the effects of dietary protein on levels of IGF and IGFBPs. We discussed the latter in relation to the intestine. If alterations in dietary protein affect IGF and IGFBP levels, an indirect effect on the intestine may result. In vitro studies using a human hepatic cell line showed that depletion of arginine, leucine, or cystine induced IGFBP-1 mRNA levels in a dose-dependent manner, to a degree similar to that found in patients with kwashiorkor (64). IGF-1 and IGF-2 levels were not affected. Chickens fed a low-protein diet had lower plasma levels of IGF-1 compared with control animals, which returned to normal on refeeding. IGFBP levels were induced by protein restriction, but there was no effect on IGF-2 levels (67).

Postnatal growth and development requires an adequate amount of dietary protein. In Wistar rats, restriction of maternal protein during gestation leads to decreased birth weight and liver weight of pups, with alterations in IGFBP expression and IGF-1 but not IGF-2 in serum and in liver tissue. A crossover study was then performed whereby the pups resumed a normal growth rate, but body weight remained below the control group. The reduction of serum IGF-1 and reduced somatic and organ growth supports a role for IGF-1 in the control of catch-up growth (87, 88).

The effect of protein intake is not restricted to the enteral route. Ma et al (76) demonstrated the positive effect of supplementation of parenteral nutrition with alanine and glutamine. Rats underwent an 80% small bowel resection. The control group was treated with total parenteral nutrition whereas the study group was given parenteral nutrition supplemented with alanine and glutamine. Mucosal thickness and villus height were increased in the study group, together with a reduction in bacterial translocation. Ileal mucosal IGF-1 mRNA was twice as high as that in the control group, and jejunal mucosal glutaminase was three times greater.

The breakdown of protein may be altered itself by restricting dietary protein intake. For example branched-chain aminotransferase is an enzyme that catalyzes the first step in the breakdown of leucine, valine, and isoleucine. The highest levels are in the stomach, but other tissues are also affected, including liver, muscle, and the intestine. In rats, baseline levels were determined. The rats were subjected to 5 weeks of protein malnutrition and then reintroduced to diets with varying amounts of protein. Enzyme levels were reduced during the malnourished state and recovered to above that of control levels on reintroduction of protein in a dose-dependent fashion. Whether this indicates a specific regulation of branched-chain aminotransferase activity requires further studies examining the exact amino acid profile of the diet and the total protein content (136).

Fat

Gastric lipase accelerates the digestion of triglycerides in human milk by releasing free fatty acids, which enhance pancreatic lipase and colipase activities. Its importance is seen in conditions where pancreatic lipase activity is low, as in cystic fibrosis and in premature and newborn infants (31, 41, 52, 71). In adult humans, changing from a low-fat to a high-fat diet doubles the amount of gastric lipase (2). In rabbits, a similar change in diet induced a 100% increase in gastric lipase. The triglyceride composition had no effect (8). The underlying mechanism, when elucidated, may allow manipulation of expression for clinical purposes, as in the nutrition of cystic fibrosis and oral intake by premature and newborn infants.

Cellular retinol-binding protein II (CRBP II) is a cytosolic protein found in abundance in the intestine. Increased levels of CRBP II mRNA and protein were found in the jejunum of rats fed a diet enriched with long-chain triglycerides compared with those fed low-fat diets or those fed diets with added medium-chain triglycerides. Retinoid X receptor (RXRα) interacts with the *cis* element of the CRBP II promoter. Levels of RXRα were similar in the long- and medium-chain triglyceride groups, which suggests that the expression of CRBP II was modulated by long-chain triglycerides or metabolites and not directly related to RXRα expression (48). This same group subsequently used nuclear run-on assays to show that the changes in CRBP II mRNA are due to changes at transcription level (129). This effect is greater with unsaturated fats (130).

Peroxisome proliferators are a diverse group of rodent hepatocarcinogens that include lipid-lowering agents, herbicides, and plasticizers. In rodents, administration increases the number and size of hepatocyte peroxisomes, and what is more interesting, it increases fatty acid metabolism by inducing peroxisomal β oxidative enzymes. Some members of the steroid hormone receptor superfamily of ligand-activated transcription factors have been identified that can be activated by peroxisome proliferators. They are termed peroxisome proliferator–activated receptors (PPAR), and four isoforms have been identified (α, β, γ, δ). PPARs are thought to play key roles both in mediating expression of fat-specific genes and in adipogenesis. In humans, the tissue expression and potential for regulation of PPAR expression in vivo is unknown. In the previously mentioned study in rats, the high-fat diet resulted in an increase of PPARα mRNA. Gel shift assays demonstrated enhanced binding activities of RXRα-PPARa to CRBP II with the addition of PPARα ligands.

A high-fat diet increases the risk of colonic, breast, and prostate cancer. In mice predisposed to intestinal neoplasia, treatment with a synthetic PPARγ ligand increased the number of polyps in the colon but not the small intestine. Activation of PPARγ may thus provide a link between a high-fat diet and an increased risk of colorectal neoplasia (107).

Fatty acid binding proteins (FABP) are cytosolic proteins that reversibly bind to long-chain fatty acids. Two main forms are intestinal FABP (I-FABP) and liver FABP (L-FABP). Both participate in the uptake and metabolism of long-chain

fats. Poirier et al (101) examined the effects of dietary oil and fat infusion on the expression of FABP in the small intestine of mice. Daily force-feeding of sunflower oil increased L-FABP mRNA and protein levels in the duodenum and proximal jejunum. Linoleic acid, the main fatty acid found in sunflower oil, caused an up-regulation in a time- and dose-dependent manner. Long-chain fats induced L-FABP in an intestinal cell line grown in serum-free media, thus excluding a hormonally mediated effect. This is in contrast to rats, in which exposure to a high-fat diet caused an increase in both L-FABP and I-FABP in the ileum (93, 117). The differences may be explained by the percentage of fat used in the diets. The study in rats used 20%–38% vegetable oil, nonphysiological, whereas the diet in mice was only slightly enriched in sunflower oil, 3%–6% in standard chow.

A further effect of dietary fat intake is on the expression of apoprotein B gene expression. Rabbits were fasted overnight, and there were no differences in apoprotein B mRNA levels on refeeding with saturated or unsaturated fats, although the latter had higher plasma cholesterol levels. However, following an overnight fast, the rabbits were given a gastric tube feed of their test diet and studied 2 h later. This resulted in a twofold increase in intestinal apoprotein B mRNA and a similar increase in transcriptional activity of the apoprotein B gene independent of the diet composition (39).

Vitamins and Minerals

Vitamin D Vitamin D is responsible for a wide range of biological responses due to the actions of its active metabolite 1α,25-dihydroxyvitamin D_3. Vitamin D_3 affects bone formation and mineral mobilization and modulates tubular resorption of calcium and phosphorus in the kidney. We concentrate on the effects on the intestine where calcium transport is stimulated. Calbindin D28K is a member of a superfamily of calcium binding proteins that share an affinity for calcium ions. Hall & Norman (51) examined the regulation of calbindin D28K gene expression in the intestine of chicks. Vitamin D–deficient chicks were fed either a calcium-supplemented diet (3%) or a low-calcium diet (0.4%). The levels of calbindin D28K mRNA and protein were unaffected, although serum calcium changed significantly. Administration of vitamin D_3 to both groups stimulated a 28-fold increase in duodenal calbindin 28K when measured 48 h later. The calcium ion alone cannot modulate calbindin gene expression but requires the presence of vitamin D_3. Similar results have been found in the rat intestine for calbindin 9K (10, 27).

Subtractive hybridization screening procedures have been used to isolate differentially expressed cDNA. Chou et al (16) used this technique based on vitamin D status to identify genes for novel proteins that are transcriptionally regulated by vitamin D metabolites in chick intestine and kidney. In the intestine, six mitochondrially encoded transcripts were up-regulated and seven nuclear encoded transcripts (including calbindin) were up- or down-regulated. In the kidney, mitochondrial and nuclear transcripts were affected differently by vitamin

D_3 status, demonstrating tissue-specific regulation by vitamin D_3 of nuclear and mitochondrial gene expression in the kidney and intestine.

Dietary Phosphorus Dietary phosphorus restriction up-regulates intestinal vitamin D receptor (VDR) through an as-yet-unknown mechanism. The effects of a low-phosphorus diet on VDR content in male Sprague Dawley rats in intestine, kidney, and splenic macrophages were examined (123). The low-phosphorus diet was given for 1–10 days. Serum phosphorus fell in the first 3 days, with a parallel increase in serum calcium. Serum vitamin D_3 rapidly increased in the first 5 days and then at a slower rate till day 10. In the control group, ligand binding assays showed that intestinal VDR increased 3.5-fold until day 3 and then fell to a plateau during days 5–10, whereas in the study group, Northern blot analyses showed intestinal VDR increased twofold by day 2 and approached control levels by day 5. A low-phosphorus diet did not up-regulate VDR mRNA levels in macrophages or the kidney. This indicates a tissue-specific up-regulation that is time dependent. Acute dietary restriction (day 1–5) up-regulates intestinal VDR mRNA through increased VDR gene expression, whereas chronic restriction (day 5–10) may be through mechanisms that are consistent with prolongation of the half-life of the receptor.

Copper Copper is an essential dietary trace element. Copper-deficient rats have hypertriglyceridemia and hypercholesterolemia and increased plasma levels of apoprotein B and E. However, intestinal apoprotein B and E mRNA are no different than normal rats, indicating a lack of effect of reduced dietary copper on intestinal gene expression at the mRNA level (80).

Zinc Zinc, another trace element, is involved in the regulation of gene expression especially through the metal response elements of the metallothionein genes. Whether there are other aspects of the function of zinc in gene expression remains unknown. Blanchard & Cousins (7) looked for intestinal mRNAs that are regulated by zinc. For 18 days, rats were fed either a zinc-deficient or a zinc-adequate diet or they were pair-fed a zinc-adequate diet. Total RNA was prepared from the intestine and analyzed using a reverse-transcriptase polymerase chain reaction method of mRNA differential display. Several cDNA bands were identified that differed in the zinc-deficient group. Of those confirmed to be regulated, several had homology to already known genes, such as cholecystokinin and uroguanylin. Novel sequences that did not match either the reported expressed sequence tags or the functionally identified genes were also identified. These need further characterization to determine whether they are primary or secondary effects.

We have discussed some of the important effects that individual dietary components have on intestinal gene expression. We now draw attention to two factors that, although not dietary constituents, are acutely affected by diet and that play a role in gene regulation of the intestine: insulin and short-chain fatty acids.

SHORT-CHAIN FATTY ACIDS

Undigested carbohydrates and some dietary fibers are fermented in the large intestine to form short-chain fatty acids. These include acetate, propionate, and butyrate. The latter is the preferred energy substrate for colonocytes, and as previously mentioned, levels vary according to bacterial flora present and type of diet. It has long been known that butyrate is involved in gene regulation. Studies with butyrate have shown an elevation of fetal hemoglobin by induction of gamma globin gene expression (82). Because butyrate is found in the lumen of normal intestine, one could suppose that it also plays a role in intestinal gene regulation.

Twenty years ago, evidence emerged that dietary fiber stimulated epithelial proliferation of the intestine and mucosal development, in the presence of gut microbes. Further studies showed that this effect was due to short-chain fatty acids (108). Subsequently, administering different types of fiber to rats has shown a correlation between cell proliferation indices and specific short-chain fatty acids, with butyrate having the most profound effect compared with acetate and propionate (62, 109). Depending on the substrate medium used and the dose of butyrate, inhibition of cell proliferation and differentiation can occur and indeed cell apoptosis can be induced (46, 68).

In addition to its effect on cell proliferation, butyrate influences the secretion of chemokines by the intestinal epithelium. This effect is through the ability of butyrate to inhibit the enzyme histone deacetylase (Figure 4). Core histone acetylation is increased, which alters chromatin structure and transcription rate. In intestinal epithelial cell line studies, where butyrate influenced chemokine secretion (42), similar results were obtained using trichostatin A. This compound is an antifungal agent and a potent inhibitor of histone deacetylase, thus confirming the mechanism of action of butyrate. The same mechanism of action is responsible for the previously mentioned induction of gamma globin expression by transcriptional activation (82).

INSULIN

Glucose concentrations in the blood tightly control the secretion of insulin from pancreatic islet cells. In the rapid response to glucose, alterations in gene expression play no part. Release of insulin from storage granules follows calcium influx, which in turn is probably induced by minor increases in ATP from the metabolism of glucose and other nutrients. Glucose, however, also exerts a more lasting effect on insulin production through increased transcription and translation of the insulin gene. The rate of translation of proinsulin mRNA to protein is enhanced by glucose by three methods. (*a*) The transfer of initiated insulin mRNA from free to membrane-bound ribosomes is increased. (*b*) The rate of pausing of the proinsulin as it passes along the ribosome is decreased. (*c*) The cellular machinery involved in the elongation of proteins in pancreatic islet cells is directly stimulated by glucose (50, 92, 141).

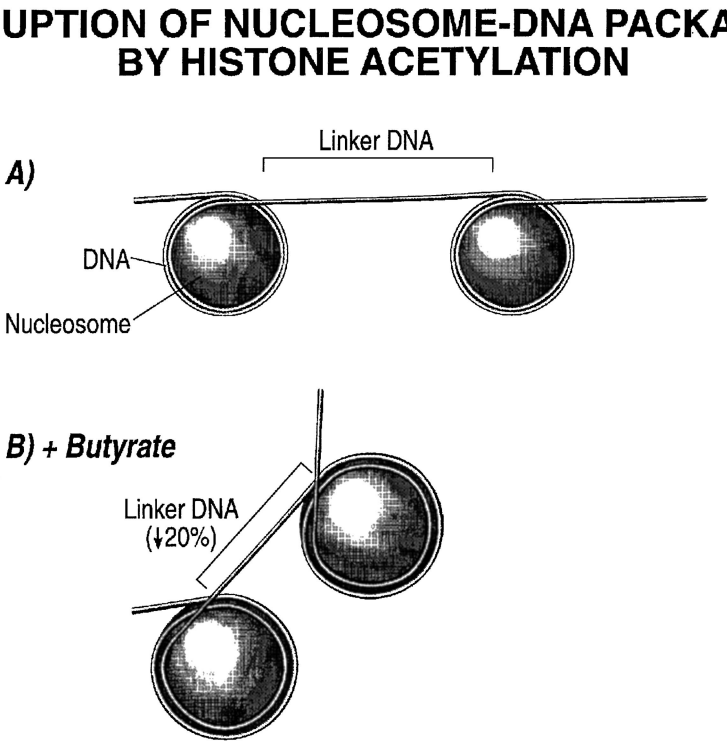

Figure 4 Relationship between nucleosomes and DNA. (*A*) DNA is wrapped two full turns around nucleosomes made up of unacetylated histones. (*B*) With butyrate-induced acetylation, the nucleosome expands, reducing the number of turns of DNA around the nucleosome to 1.8, with less linker DNA connecting each nucleosome. The result of this is that DNA cannot pass linearly from nucleosome to nucleosome but turns at an angle after every nucleosome, leading to disruption of nucleosome packaging.

Long-term changes in insulin production are also mediated by transcription. Rats fasted for 4 days have low insulin mRNA concentration, which returns to normal on refeeding (44, 45). These changes control the ability of the pancreas to secrete insulin during dietary manipulations and enable the islet cells to adapt to long-term dietary changes.

Several animal studies have looked at the effect of insulin on sucrase-isomaltase expression. An injection of streptozotocin renders animals diabetic, and increased intestinal levels of sucrase and isomaltase have been found (58, 99). Administering insulin at a dose sufficient to normalize blood glucose concentrations returns the enzyme activity to normal. Use of phloridzin to normalize blood glucose however, has no effect on sucrase or isomaltase activity. Insulin, rather than blood glucose, appears to regulate expression of sucrase-isomaltase (131). Buts et al (13) gave insulin to suckling rats between days 11 and 14. They found increased sucrase-isomaltase complex levels and increased mRNA, but IGF-1 appeared to

have no effect (13). The latter group had normal endogenous insulin production, with insulin being used to lower blood glucose and not to correct hyperglycemia. This difference may explain the conflicting results.

In diabetic rats following streptozotocin injection, proglucagon levels take 3 days to rise above control levels despite hyperglycemia. Both plasma proglucagon and intestinal proglucagon mRNA are increased at 3 days. Insulin and phloridzin both correct the hyperglycemia, but only insulin normalizes levels of plasma proglucagon and intestinal proglucagon mRNA (32).

Aldolase B is an enzyme of the glycolytic pathway. Enzyme activity and mRNA levels decline on fasting and increase on refeeding with a carbohydrate diet. To elucidate the mechanisms involved, Munnich et al (89) studied the role of hormones and substrate on the expression of aldolase B in intestine, liver, and kidney. The amount of cytoplasmic RNA was determined in the tissues of normal, adrenalectomized, thyroidectomized, diabetic, or glucagon- or cAMP-treated rats refed either a fructose- or a maltose-rich diet. In intestine, only diabetes and throidectomy affected gene expression. In normal fasted rats, no induction occurred with any of the hormones.

The intestinal oligopeptide transporter (Pept-1) plays a major role in protein nutrition and drug therapy. Thamotharan investigated the role of insulin in its expression (134). A human intestinal cell line (Caco-2) was used with glycylglutamine as the substrate. Physiological doses of insulin greatly stimulated the uptake of substrate by the cells within 30–60 min. Pept-1 gene expression was not affected, but Pept-1 protein in the apical membrane was increased. The effect of insulin persisted when Golgi apparatus was dismantled (involved in trafficking newly synthesized Pept-1) but was eliminated by disruption of apparatus involved in trafficking preformed Pept-1. Increased translocation from a preformed pool is the likely mechanism of action of insulin rather than an effect on gene expression.

CLINICAL IMPLICATIONS OF NUTRIENT-GENE INTERACTIONS

The benefits of trophic feeding compared with total parenteral nutrition in improving intestinal length, mucosal mass, and brush border enzyme activity have been established. Understanding these effects on intestinal gene expression will enable practicing clinicians to optimize their clinical management of patients, as in the use of nutrition in critically ill patients unable to eat, or in premature infants or patients following bowel resection where intestinal adaptation is important.

We have highlighted several factors in the intestinal lumen that affect gene expression in enterocytes. Dietary manipulation is an important aspect of management in gastrointestinal diseases. A classic example is the use of enteral feeding in the treatment of pediatric Crohn's disease. Initially it was assumed that improvements occurred because of a reduction in antigen load; however, whole protein polymeric diets are just as efficacious as elemental diets. It is likely that

the diets alter more than one factor in the local environment of the lumen. If we determine which factor(s) are more responsible for the subsequent reduction in inflammation, manipulation of the diet can occur to maximize response.

It has been suggested that short-chain fatty acid utilization by colonocytes in ulcerative colitis is impaired. The resulting changes in epithelial cells in turn leads to loss of mucosal barrier, resulting in inflammation. On this basis, butyrate enemas have been used in clinical studies. Several placebo-controlled studies showed no benefit (114, 126). Breuer et al (11), however, demonstrated a significant response in patients with ulcerative colitis of less than 6 months' duration. Epithelial loss is particularly evident in newly diagnosed patients, and the effects of butyrate on epithelial cell turnover may be important in these cases. Because in vitro studies suggest an up-regulation of proinflammatory chemokines with butyrate, it is not surprising that the results differ according to the patients studied. Further clarification of its role in intestinal gene expression and signaling to the mucosal immune system will result in better use of butyrate clinically.

It is interesting to note that the effect of butyrate on cell differentiation and apoptosis is causing interest in potential therapeutic use in colorectal carcinomas.

CONCLUSION

This review has attempted to lay down certain concepts of how dietary factors may affect the expression of genes in the intestine. The basic component is a system of recognizing changes in dietary factors acting on an epithelial cell (sensing) and a mechanism of converting these external molecule-cellular molecule interactions into changes in gene expression (signal transduction). The underlying mechanisms involved are beginning to be elucidated.

In the intestine, the epithelium can interact with other immune cells and may influence inflammatory reactions. Therefore, manipulation of the constituents of the lumen through dietary means could open up a new vista for treatment of human diseases. Genetic expression is regarded as a central feature of many human disorders. Understanding how nutrition can alter intestinal gene expression is an early step in the realization of its potential therapeutic implications for the future.

LITERATURE CITED

1. Amouric M, Marvaldi J, Pichon J, Bellot F, Figarella C. 1984. Effect of lactoferrin on the growth of a human colon adenocarcinoma cell line–comparison with transferrin. *In Vitro* 20:543–48
2. Armand M, Hamosh M, DiPalma JS, Gallagher J, Benjamin SB, et al. 1995. Dietary fat modulates gastric lipase activity in healthy humans. *Am. J. Clin. Nutr.* 62:74–80
3. Baggiolini M, Dewald B, Moser B. 1994. Interleukin-8 and related chemotactic

cytokines–CXC and CC chemokines. *Adv. Immunol.* 55:97–179
4. Bennett RM, Davis J. 1981. Lactoferrin binding to human peripheral blood cells: an interaction with a B-enriched population of lymphocytes and a subpopulation of adherent mononuclear cells. *J. Immunol.* 127:1211–16
5. Bennett RM, Davis J, Campbell S, Portnoff S. 1983. Lactoferrin binds to cell membrane DNA. Association of surface DNA with an enriched population of B cells and monocytes. *J. Clin. Invest.* 71:611–18
6. Berseth CL. 1987. Enhancement of intestinal growth in neonatal rats by epidermal growth factor in milk. *Am. J. Physiol.* 253:G662–65
7. Blanchard RK, Cousins RJ. 1996. Differential display of intestinal mRNAs regulated by dietary zinc. *Proc. Natl. Acad. Sci. USA* 93:6863–68
8. Borel P, Armand M, Senft M, Andre M, Lafont H, Lairon D. 1991. Gastric lipase: evidence of an adaptive response to dietary fat in the rabbit. *Gastroenterology* 100:1582–89
9. Breese EJ, Michie CA, Nicholls SW, Williams CB, Domizio P, et al. 1995. The effect of treatment on lymphokine-secreting cells in the intestinal mucosa of children with Crohn's disease. *Aliment. Pharmacol. Ther.* 9:547–52
10. Brehier A, Thomasset M. 1990. Stimulation of calbindin-D9K (CaBP9K) gene expression by calcium and 1,25-dihydroxycholecalciferol in fetal rat duodenal organ culture. *Endocrinology* 127:580–87
11. Breuer RI, Soergel KH, Lashner BA, Christ ML, Hanauer SB, et al. 1997. Short chain fatty acid rectal irrigation for left-sided ulcerative colitis: a randomised, placebo controlled trial. *Gut* 40:485–91
12. Britton JR, Koldovsky O. 1989. Gastric luminal digestion of lactoferrin and transferrin by preterm infants. *Early Hum. Dev.* 19:127–35
13. Buts JP, Duranton B, De Keyser N, Sokal EM, Maernhout AS, et al. 1998. Premature stimulation of rat sucrase-isomaltase (SI) by exogenous insulin and the analog B-Asp10 is regulated by a receptor-mediated signal triggering SI gene transcription. *Pediatr. Res.* 43:585–91
14. Byatt JC, Schmuke JJ, Comens PG, Johnson DA, Collier RJ. 1990. The effect of bovine lactoferrin on muscle growth in vivo and in vitro. *Biochem. Biophys. Res. Commun.* 173:548–53
15. Chatelain F, Pegorier JP, Minassian C, Bruni N, Tarpin S, et al. 1998. Development and regulation of glucose-6-phosphatase gene expression in rat liver, intestine, and kidney: in vivo and in vitro studies in cultured fetal hepatocytes. *Diabetes* 47: 882–89
16. Chou SY, Hannah SS, Lowe KE, Norman AW, Henry HL. 1995. Tissue-specific regulation by vitamin D status of nuclear and mitochondrial gene expression in kidney and intestine. *Endocrinology* 136:5520–26
17. Chu SH, Walker WA. 1991. Growth factor signal transduction in human intestinal cells. *Adv. Exp. Med. Biol.* 310:107–12
18. Cohick WS, Clemmons DR. 1993. The insulin-like growth factors. *Annu. Rev. Physiol.* 55:131–53
19. Colotta F, Borre A, Wang JM, Tatanelli M, Maddalena F, et al. 1992. Expression of a monocyte chemotactic cytokine by human mononuclear phagocytes. *J. Immunol.* 148:760–65
20. Conteas CN, DeMorrow JM, Majumdar AP. 1986. Effect of epidermal growth factor on growth and maturation of fetal and neonatal rat small intestine in organ culture. *Experientia* 42:950–52
21. Cox TM, Mazurier J, Spik G, Montreuil J, Peters TJ. 1979. Iron binding proteins and influx of iron across the duodenal brush border. Evidence for specific lactotransferrin receptors in the human intestine. *Biochim. Biophys. Acta* 588:120–28

22. Culouscou JM, Remacle-Bonnet M, Garrouste F, Fantini J, Marvaldi J, Pommier G. 1990. Production of insulin-like growth factor II (IGF-II) and different forms of IGF-binding proteins by HT-29 human colon carcinoma cell line. *J. Cell Physiol.* 143:405–15
23. Culouscou JM, Remacle-Bonnet M, Garrouste F, Marvaldi J, Pommier G. 1987. Simultaneous production of IGF-I and EGF competing growth factors by HT-29 human colon cancer line. *Int. J. Cancer* 40:646–52
24. David ES, Cingari DS, Ferraris RP. 1995. Dietary induction of intestinal fructose absorption in weaning rats. *Pediatr. Res.* 37:777–82
25. Davidson LA, Lonnerdal B. 1987. Persistence of human milk proteins in the breast-fed infant. *Acta Paediatr. Scand.* 76:733–40
26. Davidson LA, Lonnerdal B. 1988. Specific binding of lactoferrin to brush-border membrane: ontogeny and effect of glycan chain. *Am. J. Physiol.* 254:G580–85
27. Desplan C, Thomasset M, Moukhtar M. 1983. Synthesis, molecular cloning, and restriction analysis of DNA complementary to vitamin D-dependent calcium-binding protein mRNA from rat duodenum. *J. Biol. Chem.* 258:2762–65
28. Diamond JM, Karasov WH. 1987. Adaptive regulation of intestinal nutrient transporters. *Proc. Natl. Acad. Sci. USA* 84: 2242–45
29. Diamond JM, Karasov WH, Cary C, Enders D, Yung R. 1984. Effect of dietary carbohydrate on monosaccharide uptake by mouse small intestine in vitro. *J. Physiol.* 349:419–40
30. Dickson RC, Abelson J, Barnes WM, Reznikoff WS. 1975. Genetic regulation: the Lac control region. *Science* 187:27–35
31. DiPalma J, Kirk CL, Hamosh M, Colon AR, Benjamin SB, Hamosh P. 1991. Lipase and pepsin activity in the gastric mucosa of infants, children, and adults. *Gastroenterology* 101:116–21
32. Dumonteil E, Magnan C, Ritz-Laser B, Meda P, Dussoix P, et al. 1998. Insulin, but not glucose lowering corrects the hyperglucagonemia and increased proglucagon messenger ribonucleic acid levels observed in insulinopenic diabetes. *Endocrinology* 139:4540–46
33. Eckmann L, Jung HC, Schurer-Maly C, Panja A, Morzycka-Wroblewska E, Kagnoff MF. 1993. Differential cytokine expression by human intestinal epithelial cell lines: regulated expression of interleukin 8. *Gastroenterology* 105:1689–97
34. Emvo EN, Raul F, Koch B, Neuville P, Foltzer-Jourdainne C. 1996. Sucrase-isomaltase gene expression in suckling rat intestine: hormonal, dietary, and growth factor control. *J. Pediatr. Gastroenterol. Nutr.* 23:262–69
35. Ferraris RP, Diamond J. 1992. Crypt-villus site of glucose transporter induction by dietary carbohydrate in mouse intestine. *Am. J. Physiol.* 262:G1069–73
36. Ferraris RP, Diamond JM. 1989. Specific regulation of intestinal nutrient transporters by their dietary substrates. *Annu. Rev. Physiol.* 51:125–41
37. Ferraris RP, Diamond JM. 1993. Crypt/villus site of substrate-dependent regulation of mouse intestinal glucose transporters. *Proc. Natl. Acad. Sci. USA* 90: 5868–72
38. Ferraris RP, Villenas SA, Hirayama BA, Diamond J. 1992. Effect of diet on glucose transporter site density along the intestinal crypt-villus axis. *Am. J. Physiol.* 262:G1060–68
39. Fisher EA, Anbari A, Klurfeld DM, Kritchevsky D. 1988. Independent effects of diet and nutritional status on apoprotein B gene expression in rabbit. *Arteriosclerosis* 8:797–803
40. Foltzer-Jourdainne C, Garaud JC, Nsi-Emvo E, Raul F. 1993. Epidermal growth factor and the maturation of intestinal sucrase in suckling rats. *Am. J. Physiol.* 265: G459–66

41. Fredrikzon B, Olivecrona T. 1978. Decrease of lipase and esterase activities in intestinal contents of newborn infants during test meals. *Pediatr. Res.* 12:631–34
42. Fusunyan RD, Quinn JJ, Fujimoto M, MacDermott RP, Sanderson IR. 1999. Butyrate switches the pattern of chemokine secretion by intestinal epithelial cells. *Mol. Med.* 5:631–40
43. Garre C, Bianchi-Scarra G, Sirito M, Musso M, Ravazzolo R. 1992. Lactoferrin binding sites and nuclear localization in K562(S) cells. *J. Cell Physiol.* 153:477–82
44. Giddings SJ, Chirgwin J, Permutt MA. 1981. The effects of fasting and feeding on preproinsulin messenger RNA in rats. *J. Clin. Invest.* 67:952–60
45. Giddings SJ, Chirgwin J, Permutt MA. 1982. Effects of glucose on proinsulin messenger RNA in rats in vivo. *Diabetes* 31:624–29
46. Ginsburg E, Salomon D, Sreevalsan T, Freese E. 1973. Growth inhibition and morphological changes caused by lipophilic acids in mammalian cells. *Proc. Natl. Acad. Sci. USA* 70:2457–61
47. Goda T, Yasutake H, Suzuki Y, Takase S, Koldovsky O. 1995. Diet-induced changes in gene expression of lactase in rat jejunum. *Am. J. Physiol.* 268:G1066–73
48. Goda T, Yasutake H, Takase S. 1994. Dietary fat regulates cellular retinol-binding protein II gene expression in rat jejunum. *Biochim. Biophys. Acta* 1200:34–40
49. Goodlad RA, Wilson TJ, Lenton W, Gregory H, McCullough KG, Wright NA. 1985. Urogastrone-epidermal growth factor is trophic to the intestinal epithelium of parenterally fed rats. *Experientia* 41:1161–63
50. Guest PC, Bailyes EM, Rutherford NG, Hutton JC. 1991. Insulin secretory granule biogenesis. Co-ordinate regulation of the biosynthesis of the majority of constituent proteins. *Biochem. J.* 274:73–78
51. Hall AK, Norman AW. 1990. Regulation of calbindin-D28K gene expression in the chick intestine: effects of serum calcium status and 1,25-dihydroxyvitamin D3. *J. Bone Miner. Res.* 5:331–36
52. Hamosh M, Scanlon JW, Ganot D, Likel M, Scanlon KB, Hamosh P. 1981. Fat digestion in the newborn. Characterization of lipase in gastric aspirates of premature and term infants. *J. Clin. Invest.* 67:838–46
53. Hashizume S, Kuroda K, Murakami H. 1983. Identification of lactoferrin as an essential growth factor for human lymphocytic cell lines in serum-free medium. *Biochim. Biophys. Acta* 763:377–82
54. He J, Furmanski P. 1995. Sequence specificity and transcriptional activation in the binding of lactoferrin to DNA. *Nature* 373: 721–24
55. He Y, Chu SH, Walker WA. 1993. Nucleotide supplements alter proliferation and differentiation of cultured human (Caco-2) and rat (IEC-6) intestinal epithelial cells. *J. Nutr.* 123:1017–27
56. Hershberg RM, Cho DH, Youakim A, Bradley MB, Lee JS, et al. 1998. Highly polarized HLA class II antigen processing and presentation by human intestinal epithelial cells. *J. Clin. Invest.* 102:792–803
57. Higashimoto Y, Opara EC, Liddle RA. 1995. Dietary regulation of glucose-dependent insulinotropic peptide (GIP) gene expression in rat small intestine. *Comp. Biochem. Physiol. C* 110:207–14
58. Hoffman LR, Chang EB. 1992. Altered regulation of regional sucrase-isomaltase expression in diabetic rat intestine. *Am. J. Physiol.* 262:G983–89
59. Hu WL, Mazurier J, Sawatzki G, Montreuil J, Spik G. 1988. Lactotransferrin receptor of mouse small-intestinal brush border. Binding characteristics of membrane-bound and triton X-100-solubilized forms. *Biochem. J.* 249:435–41
60. Imamoto F, Yanofsky C. 1967. Transcription of the tryptophan operon in polarity mutants of *Escherichia coli*. I. Characterization of the tryptophan messenger RNA

of polar mutants. *J. Mol. Biol.* 28:1–23
61. Izzo RS, Witkon K, Chen AI, Hadjiyane C, Weinstein MI, Pellecchia C. 1993. Neutrophil-activating peptide (interleukin-8) in colonic mucosa from patients with Crohn's disease. *Scand. J. Gastroenterol.* 28:296–300
62. Jacobs LR, Lupton JR. 1984. Effect of dietary fibers on rat large bowel mucosal growth and cell proliferation. *Am. J. Physiol.* 246:G378–85
63. Jarocka-Cyrta E, Perin N, Keelan M, Wierzbicki T, Clandinin MT, et al. 1998. Early dietary experience influences ontogeny of intestine in response to dietary lipid changes in later life. *Am. J. Physiol.* 275:G250–58
64. Jousse C, Bruhat A, Ferrara M, Fafournoux P. 1998. Physiological concentration of amino acids regulates insulin-like-growth-factor-binding protein 1 expression. *Biochem. J.* 334:147–53
65. Kawakami H, Lonnerdal B. 1991. Isolation and function of a receptor for human lactoferrin in human fetal intestinal brush-border membranes. *Am. J. Physiol.* 261:G841–46
66. Kirschner BS. 1988. Nutritional consequences of inflammatory bowel disease on growth. *J. Am. Coll. Nutr.* 7:301–8
67. Kita K, Tomas FM, Owens PC, Knowles SE, Forbes BE, et al. 1996. Influence of nutrition on hepatic IGF-I mRNA levels and plasma concentrations of IGF-I and IGF-II in meat-type chickens. *J. Endocrinol.* 149:181–90
68. Kruh J. 1982. Effects of sodium butyrate, a new pharmacological agent, on cells in culture. *Mol. Cell Biochem.* 42:65–82
69. Kurokowa M, Lynch K, Podolsky DK. 1987. Effects of growth factors on an intestinal epithelial cell line: transforming growth factor beta inhibits proliferation and stimulates differentiation. *Biochem. Biophys. Res. Commun.* 142:775–82
70. Lee A, Gemmell E. 1972. Changes in the mouse intestinal microflora during weaning: role of volatile fatty acids. *Infect. Immun.* 5:1–7
71. Lee PC, Borysewicz R, Struve M, Raab K, Werlin SL. 1993. Development of lipolytic activity in gastric aspirates from premature infants. *J. Pediatr. Gastroenterol. Nutr.* 17:291–97
72. Leeper LL, Henning SJ. 1990. Development and tissue distribution of sucrase-isomaltase mRNA in rats. *Am. J. Physiol.* 258:G52–58
73. Levine GM, Deren JJ, Steiger E, Zinno R. 1974. Role of oral intake in maintenance of gut mass and disaccharide activity. *Gastroenterology* 67:975–82
74. Lonnerdal B. 1994. Lactoferrin receptors in intestinal brush border membranes. *Adv. Exp. Med. Biol.* 357:171–75
75. Lowe WLJ, Adamo M, Werner H, Roberts CTJ, LeRoith D. 1989. Regulation by fasting of rat insulin-like growth factor I and its receptor. Effects on gene expression and binding. *J. Clin. Invest.* 84:619–26
76. Ma Y, Jiang Z, Bai M. 1995. The change of gut barrier function and gene expression after surgical stress and parenteral nutrition. *Chung-Hua Wai K'o Tsa Chih* 33: 371–73(In Chinese)
77. MacDermott RP. 1996. Alterations of the mucosal immune system in inflammatory bowel disease. *J. Gastroenterol.* 31:907–16
78. MacDermott RP, Sanderson IR, Reinecker HC. 1998. The central role of chemokines (chemotactic cytokines) in the immunopathogenesis of ulcerative colitis and Crohn's disease. *Inflamm. Bowel Dis.* 4:54–67
79. Massimino SP, McBurney MI, Field CJ, Thomson AB, Keelan M, et al. 1998. Fermentable dietary fiber increases GLP-1 secretion and improves glucose homeostasis despite increased intestinal glucose transport capacity in healthy dogs. *J. Nutr.* 128: 1786–93
80. Mazur A, Nassir F, Gueux E, Cardot P, Bellanger J, et al. 1992. The effect of dietary copper on rat plasma apolipoprotein B, E

plasma levels, and apolipoprotein gene expression in liver and intestine. *Biol. Trace Elem. Res.* 34:107–13

81. Mazurier J, Montreuil J, Spik G. 1985. Visualization of lactotransferrin brush-border receptors by ligand-blotting. *Biochim. Biophys. Acta* 821:453–60
82. McCaffrey PG, Newsome DA, Fibach E, Yoshida M, Su MS. 1997. Induction of gamma-globin by histone deacetylase inhibitors. *Blood* 90:2075–83
83. McGee DW, Beagley KW, Aicher WK, McGhee JR. 1993. Transforming growth factor-beta and IL-1 beta act in synergy to enhance IL-6 secretion by the intestinal epithelial cell line, IEC-6. *J. Immunol.* 151:970–78
84. Midtvedt AC, Midtvedt T. 1992. Production of short chain fatty acids by the intestinal microflora during the first 2 years of human life. *J. Pediatr. Gastroenterol. Nutr.* 15:395–403
85. Mikogami T, Marianne T, Spik G. 1995. Effect of intracellular iron depletion by picolinic acid on expression of the lactoferrin receptor in the human colon carcinoma cell subclone HT29-18-C1. *Biochem. J.* 308:391–97
86. Miyazawa K, Mantel C, Lu L, Morrison DC, Broxmeyer HE. 1991. Lactoferrin-lipopolysaccharide interactions. Effect on lactoferrin binding to monocyte/macrophage-differentiated HL -60 cells. *J. Immunol.* 146:723–29
87. Muaku SM, Beauloye V, Thissen JP, Underwood LE, Ketelslegers JM, Maiter D. 1995. Effects of maternal protein malnutrition on fetal growth, plasma insulin-like growth factors, insulin-like growth factor binding proteins, and liver insulin-like growth factor gene expression in the rat. *Pediatr. Res.* 37:334–42
88. Muaku SM, Underwood LE, Selvais PL, Ketelslegers JM, Maiter D. 1995. Maternal protein restriction early or late in rat pregnancy has differential effects on fetal growth, plasma insulin-like growth factor-I (IGF-I) and liver IGF-I gene expression. *Growth Regul.* 5:125–32
89. Munnich A, Besmond C, Darquy S, Reach G, Vaulont S, et al. 1985. Dietary and hormonal regulation of aldolase B gene expression. *J. Clin. Invest.* 75:1045–52
90. Nichols BL, McKee KS, Huebers HA. 1990. Iron is not required in the lactoferrin stimulation of thymidine incorporation into the DNA of rat crypt enterocytes. *Pediatr. Res.* 27:525–28
91. Nishimura A, Fujimoto M, Oguchi S, Fusunyan RD, MacDermott RP, Sanderson IR. 1998. Short-chain fatty acids regulate IGF-binding protein secretion by intestinal epithelial cells. *Am. J. Physiol.* 275:E55–63
92. Nishio Y, Aiello LP, King GL. 1994. Glucose induced genes in bovine aortic smooth muscle cells identified by mRNA differential display. *FASEB J.* 8:103–6
93. Ockner RK, Manning JA. 1974. Fatty acid-binding protein in small intestine. Identification, isolation, and evidence for its role in cellular fatty acid transport. *J. Clin. Invest.* 54:326–38
94. Oguchi S, Walker WA, Sanderson IR. 1993. Insulin-like growth factor binding protein profile secreted by human intestinal epithelial cells varies with polarity. *Biochem. Biophys. Res. Commun.* 196: 789–93
95. Oguchi S, Walker WA, Sanderson IR. 1994. Profile of IGF-binding proteins secreted by intestinal epithelial cells changes with differentiation. *Am. J. Physiol.* 267: G843–50
96. Oguchi S, Walker WA, Sanderson IR. 1995. Iron saturation alters the effect of lactoferrin on the proliferation and differentiation of human enterocytes (Caco-2 cells). *Biol. Neonate* 67:330–39
97. Ohno Y, Lee J, Fusunyan RD, MacDermott RP, Sanderson IR. 1997. Macrophage inflammatory protein-2: chromosomal regulation in rat small intestinal epithelial cells. *Proc. Natl. Acad. Sci. USA* 94:10279–84

98. O'Loughlin EV, Chung M, Hollenberg M, Hayden J, Zahavi I, Gall DG. 1985. Effect of epidermal growth factor on ontogeny of the gastrointestinal tract. *Am. J. Physiol.* 249:G674–78
99. Olsen WA, Korsmo H. 1977. The intestinal brush border membrane in diabetes. Studies of sucrase-isomaltase metabolism in rats with streptozotocin diabetes. *J. Clin. Invest.* 60:181–88
100. Perin N, Keelan M, Jarocka-Cyrta E, Clandinin MT, Thomson AB. 1997. Ontogeny of intestinal adaptation in rats in response to isocaloric changes in dietary lipids. *Am. J. Physiol.* 273:G713–20
101. Poirier H, Niot I, Degrace P, Monnot MC, Bernard A, Besnard P. 1997. Fatty acid regulation of fatty acid-binding protein expression in the small intestine. *Am. J. Physiol.* 273:G289–95
102. Puccio F, Lehy T. 1988. Oral administration of epidermal growth factor in suckling rats stimulates cell DNA synthesis in fundic and antral gastric mucosae as well as in intestinal mucosa and pancreas. *Regul. Pept.* 20:53–64
103. Raul F, Kedinger M, Simon PM, Grenier JF, Haffen K. 1981. Comparative in vivo and in vitro effect of mono- and disaccharides on intestinal brush border enzyme activities in suckling rats. *Biol. Neonate* 39:200–7
104. Reimer RA, Thomson AB, Rajotte RV, Basu TK, Ooraikul B, McBurney MI. 1997. A physiological level of rhubarb fiber increases proglucagon gene expression and modulates intestinal glucose uptake in rats. *J. Nutr.* 127:1923–28
105. Reimund JM, Wittersheim C, Dumont S, Muller CD, Baumann R, et al. 1996. Mucosal inflammatory cytokine production by intestinal biopsies in patients with ulcerative colitis and Crohn's disease. *J. Clin. Immunol.* 16:144–50
106. Reinecker HC, Loh EY, Ringler DJ, Mehta A, Rombeau JL, MacDermott RP. 1995. Monocyte-chemoattractant protein 1 gene expression in intestinal epithelial cells and inflammatory bowel disease mucosa. *Gastroenterology* 108:40–50
107. Saez E, Tontonoz P, Nelson MC, Alvarez JG, Ming UT, et al. 1998. Activators of the nuclear receptor PPARγ enhance colon polyp formation. *Nat. Med.* 4:1058–61
108. Sakata T. 1987. Stimulatory effect of short-chain fatty acids on epithelial cell proliferation in the rat intestine: a possible explanation for trophic effects of fermentable fibre, gut microbes and luminal trophic factors. *Br. J. Nutr.* 58:95–103
109. Sakata T, Tamate H. 1978. Rumen epithelial cell proliferation accelerated by rapid increase in intraruminal butyrate. *J. Dairy Sci.* 61:1109–13
110. Sanchez L, Calvo M, Brock JH. 1992. Biological role of lactoferrin. *Arch. Dis. Child* 67:657–61
111. Sanderson IR, Ouellette AJ, Carter EA, Harmatz PR. 1992. Ontogeny of class II MHC mRNA in the mouse small intestinal epithelium. *Mol. Immunol.* 29:1257–63
112. Sanderson IR, Ouellette AJ, Carter EA, Harmatz PR. 1993. Ontogeny of Ia messenger RNA in the mouse small intestinal epithelium is modulated by age of weaning and diet. *Gastroenterology* 105:974–80
113. Sanderson IR, Udeen S, Davies PS, Savage MO, Walker-Smith JA. 1987. Remission induced by an elemental diet in small bowel Crohn's disease. *Arch. Dis. Child* 62:123–27
114. Scheppach W. 1996. Treatment of distal ulcerative colitis with short-chain fatty acid enemas. A placebo-controlled trial. German-Austrian SCFA study group. *Digest. Dis. Sci.* 41:2254–59
115. Sebastio G, Hunziker W, Ballabio A, Auricchio S, Semenza G. 1986. On the primary site of control in the spontaneous development of small-intestinal sucrase-isomaltase after birth. *FEBS Lett.*

208:460–64; Erratum. 1987. *FEBS Lett.* 223:423

116. Sebastio G, Hunziker W, O'Neill B, Malo C, Menard D, et al. 1987. The biosynthesis of intestinal sucrase-isomaltase in human embryo is most likely controlled at the level of transcription. *Biochem. Biophys. Res. Commun.* 149:830–39
117. Shields HM, Bates ML, Bass NM, Best CJ, Alpers DH, Ockner RK. 1986. Light microscopic immunocytochemical localization of hepatic and intestinal types of fatty acid-binding proteins in rat small intestine. *J. Lipid Res.* 27:549–57
118. Shirota K, LeDuy L, Yuan SY, Jothy S. 1990. Interleukin-6 and its receptor are expressed in human intestinal epithelial cells. *Virchows Arch. B* 58:303–8
119. Siafakas CG, Anatolitou F, Fusunyan RD, Walker WA, Sanderson IR. 1999. Vascular endothelial growth factor (VEGF) is present in human breast milk and its receptor is present on intestinal epithelial cells. *Pediatr. Res.* 45:652–57
120. Simmons JG, Hoyt EC, Westwick JK, Brenner DA, Pucilowska JB, Lund PK. 1995. Insulin-like growth factor-I and epidermal growth factor interact to regulate growth and gene expression in IEC-6 intestinal epithelial cells. *Mol. Endocrinol.* 9:1157–65
121. Solberg DH, Diamond JM. 1987. Comparison of different dietary sugars as inducers of intestinal sugar transporters. *Am. J. Physiol.* 252:G574–84
122. Soliman AT, Hassan AE, Aref MK, Hintz RL, Rosenfeld RG, Rogol AD. 1986. Serum insulin-like growth factors I and II concentrations and growth hormone and insulin responses to arginine infusion in children with protein-energy malnutrition before and after nutritional rehabilitation. *Pediatr. Res.* 20:1122–30
123. Sriussadaporn S, Wong MS, Pike JW, Favus MJ. 1995. Tissue specificity and mechanism of vitamin D receptor upregulation during dietary phosphorus restriction in the rat. *J. Bone Miner. Res.* 10:271–80
124. Stadnyk AW. 1994. Cytokine production by epithelial cells. *FASEB J.* 8:1041–47
125. Staley MD, Gibson CA, Herbein JF, Grosvenor CE, Baumrucker CR. 1998. Rat milk and dietary long arginine 3 insulin-like growth factor I promote intestinal growth of newborn rat pups. *Pediatr. Res.* 44:512–18
126. Steinhart AH, Hiruki T, Brzezinski A, Baker JP. 1996. Treatment of left-sided ulcerative colitis with butyrate enemas: a controlled trial. *Aliment. Pharmacol. Ther.* 10:729–36
127. Straus DS, Takemoto CD. 1990. Effect of dietary protein deprivation on insulin-like growth factor (IGF)-I and -II, IGF binding protein-2, and serum albumin gene expression in rat. *Endocrinology*127:1849–60
128. Strieter RM, Chensue SW, Standiford TJ, Basha MA, Showell HJ, et al. 1990. Disparate gene expression of chemotactic cytokines by human mononuclear phagocytes. *Biochem. Biophys. Res. Commun.* 166:886–91
129. Suruga K, Mochizuki K, Kitagawa M, Goda T, Horie N, et al. 1999. Transcriptional regulation of cellular retinol-binding protein, type II gene expression in small intestine by dietary fat. *Arch. Biochem. Biophys.* 362:159–66
130. Suruga K, Suzuki R, Goda T, Takase S. 1995. Unsaturated fatty acids regulate gene expression of cellular retinol-binding protein, type II in rat jejunum. *J. Nutr.* 125:2039–44
131. Takenoshita M, Yamaji R, Inui H, Miyatake K, Nakano Y. 1998. Suppressive effect of insulin on the synthesis of sucrase-isomaltase complex in small intestinal epithelial cells, and abnormal increase in the complex under diabetic conditions. *Biochem. J.* 329:597–600
132. Tanaka T, Kishi K, Igawa M, Takase S, Goda T. 1998. Dietary carbohydrates

enhance lactase/phlorizin hydrolase gene expression at a transcription level in rat jejunum. *Biochem. J.* 331:225–30

133. Tappenden KA, Drozdowski LA, Thomson AB, McBurney MI. 1998. Short-chain fatty acid-supplemented total parenteral nutrition alters intestinal structure, glucose transporter 2 (GLUT2) mRNA and protein, and proglucagon mRNA abundance in normal rats. *Am. J. Clin. Nutr.* 68:118–25
134. Thamotharan M, Bawani SZ, Zhou X, Adibi SA. 1999. Hormonal regulation of oligopeptide transporter pept-1 in a human intestinal cell line. *Am. J. Physiol.* 276:C821–26
135. Tobler A, Meier R, Seitz M, Dewald B, Baggiolini M, Fey MF. 1992. Glucocorticoids downregulate gene expression of GM-CSF, NAP-1/IL-8, and IL-6, but not of M-CSF in human fibroblasts. *Blood* 79:45–51
136. Torres N, Lopez G, De Santiago W, Hutson SM, Tovar AR. 1998. Dietary protein level regulates expression of the mitochondrial branched-chain aminotransferase in rats. *J. Nutr.* 128:1368–75
137. Ulshen MH, Lyn-Cook LE, Raasch RH. 1986. Effects of intraluminal epidermal growth factor on mucosal proliferation in the small intestine of adult rats. *Gastroenterology* 91:1134–40
138. Villafuerte BC, Koop BL, Pao CI, Gu L, Birdsong GG, Phillips LS. 1994. Coculture of primary rat hepatocytes and nonparenchymal cells permits expression of insulin-like growth factor binding protein-3 in vitro. *Endocrinology* 134:2044–50
139. Villiger PM, Terkeltaub R, Lotz M. 1992. Production of monocyte chemoattractant protein-1 by inflamed synovial tissue and cultured synoviocytes. *J. Immunol.* 149:722–27
140. Weaver LT, Walker WA. 1988. Epidermal growth factor and the developing human gut. *Gastroenterology* 94:845–47
141. Welsh M, Scherberg N, Gilmore R, Steiner DF. 1986. Translational control of insulin biosynthesis. Evidence for regulation of elongation, initiation and signal-recognition-particle-mediated translational arrest by glucose. *Biochem. J.* 235:459–67
142. Wright NA, Pike C, Elia G. 1990. Induction of a novel epidermal growth factor-secreting cell lineage by mucosal ulceration in human gastrointestinal stem cells. *Nature* 343:82–85
143. Yang SK, Eckmann L, Panja A, Kagnoff MF. 1997. Differential and regulated expression of C-X-C, C-C, and C-chemokines by human colon epithelial cells. *Gastroenterology* 113:1214–23
144. Yoshimura T, Leonard EJ. 1990. Secretion by human fibroblasts of monocyte chemoattractant protein-1, the product of gene JE. *J. Immunol.* 144:2377–83.