IMMUNOLOGY of the GASTROINTESTINAL TRACT

Edited by
Peter Asquith

Foreword by P.G.H. Gell

Churchill Livingstone

Immunology of the Gastrointestinal Tract

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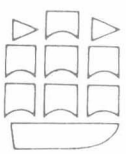

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Immunology of the Gastrointestinal Tract

Foreword

Any person (such as a scientifically inclined archangel) who was around 200 million or so years ago would have been able to observe the development of an interesting mutation among our distant progenitors in the maritime muds – the divergence of the alpha heavy Ig chain from the gamma heavy chain. This is where the subject of this book really starts; the point at which it became clear that something must and could be done about defence at surfaces as well as in fluids, by the then evolving chordates and vertebrates. It was evidently a successful effort, in that it persisted as a major means of defence; but just how the rather elaborate architecture of J chain and secretory piece, as well as the considerable structural differences in the alpha Fc itself, had to be the acquisition of these special powers, is still a bit obscure, at least to me.

Narrowing the time gap a little, the years between about 1920 when oral immunization and coproantibodies were being actively discussed and the early 1960s when Heremans described IgA, and Tomasi, Bienenstock and their colleagues indicated a function for it, marked an era when what went on in the guts was hardly respectable, at least in the eyes of the professional immunochemists who then dominated the field. The great mass of lymphoid tissue in the intestine, and even more that in the appendix (removed with the gayest abandon in that era), remained *terra incognita* except to morbid pathologists. Nevertheless the very first example of immunological tolerance, by the feeding of picryl chloride, was described by Chase about 1943, to sow seeds which have hardly all germinated even now (any gardener knows that seldom do all seeds in a batch germinate simultaneously: so in science). Indeed it would seem that tolerance *per os* is a subject of maximal practical and theoretical interest yet to be exploited. Moreover the complex dance of cells which goes on in the *lamina propria* of the intestine has only recently come under study, under stimuli from two directions – from academic immunologists interested in (and surprised by) the homing thither of labelled lymphocytes, and, to rather greater effect, from clinicians interested in what on earth could be going on in the tissues of coeliacs, in Crohn's disease, in ulcerative colitis and so on. These carry the (almost) unmistakable stamp of immunologically conditioned diseases: but just what precise mechanisms are at work? In particular what role is played by 'ordinary' and what by IgA and IgE antibody and what by the possibly more primitive mechanisms of cellular hypersensitivity? It is to questions of this sort that this book gives an informed, illuminating though inevitably still incomplete answer.

P. G. H. G.

Preface

My purpose in compiling this book has been to provide a comprehensive and up-to-date account of the immunology of the gastrointestinal tract. To this end the book has two parts: the first consists of three chapters on organisational and functional aspects which form an introduction and basis for Part 2. In this second part disorders of the gut from the mouth to the colon and rectum are considered, mainly as they affect man but also some attention is paid to animal diseases. In planning this book I invited a number of internationally known experts to write on their respective fields and I am very grateful to them for providing a series of extensive and authoritative reviews. The current marked and widespread interest in gut immunity is stimulated by attempts at unravelling the complexities of the immunological apparatus of the gut and also by the belief that some, perhaps many, intestinal diseases may have an immunological basis. I hope the book will be of value to clinicians, immunologists and basic scientists.

I would like to acknowledge my indebtedness to Professor Philip Gell FRS and members of his department who stimulated my interest in gut immunology, and to Dr W. T. Cooke who permitted me to study patients under his care. I am also grateful to Mr S. Jenkins and his staff at the Barnes Medical Library, University of Birmingham, who checked the many references, to Mr Roy Steel for checking the manuscripts and to the staff of Churchill Livingstone for their advice and help during the various stages of this book. Finally my thanks are also due to my wife Janet and our children John, Karen and Helen for their indulgence, encouragement and support.

Birmingham, 1978 P. A.

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PART I Organizational and functional aspects

1. The physiology of the local immune response

J. Bienenstock

INTRODUCTION

In 1919 Besredka published his findings showing that oral immunization of rabbits provided protection against otherwise fatal Shiga bacillus infection, regardless of the serum antibody level. In fact, his subsequent monograph entitled 'Local immunization', published in 1927, showed that he had developed theories of immunity, based on such experimental observations, before and during the First World War which he subsequently put into practice in the immunization of thousands of army personnel. The data recorded in his book clearly showed the efficacy of oral immunization in prevention of dysentery. Also at that time Davies (1922) showed that faecal antibody was present in the stools of patients with bacillary dysentery before serum antibody was present. It was some 40 years later that Heremans (1960) described a new immunoglobulin in serum different from those already known, gamma G and gamma M, and termed it gamma A or β_2A. Tomasi and his co-workers (1965), soon afterwards, showed that the IgA class of immunoglobulins predominated in several external secretions, that IgA-containing plasma cells were to be found in large numbers in the salivary gland, and that the secretory IgA in saliva and colostrum differed immunochemically from that found in serum, in being larger, and in having an extra polypeptide chain known at that time as secretory piece. From these observations came a renewed interest in mucosal immunity, a topic which has been the subject of several extensive reviews (Tomasi and Bienenstock, 1968; Heremans, 1968, Dayton, Small, Chanock, Kaufman and Tomasi, 1970; Tomasi and Grey, 1972; Mestecky and Lawton, 1974).

Evidence exists for the presence of IgG, IgM, IgA and IgE in external secretions, particularly if the mucosa is involved by an inflammatory process allowing the egress of serum proteins. The IgA immunoglobulin molecule predominates in external secretions, but it should also be recognized that IgG and IgM may play important roles in terms of local antibody production (*vide infra*). The IgE class of immunoglobulin has been shown to be synthesized locally, particularly in mucosal surfaces (Tada and Ishizaka, 1970). However, the IgE found in secretions does not differ from that found in serum, as it appears not to contain either a J chain or secretory component (Brandtzaeg, 1973).

IgA

STRUCTURE

Tomasi *et al.* (1965) first showed that IgA predominated in certain external secretions and that the IgA molecule differed from its serum counterpart. It is now known that the secretory IgA molecule in its classical 11S form consists of four light chains, four heavy chains, one secretory component and one J chain. The whole molecule has a molecular weight of approximately 385 000. In humans, the secretory component (SC) has a molecular weight of about 60 000 and the J chain about 15 000. The J chain has an enormously increased axial ratio, much higher than other molecules of similar molecular size (Koshland and Wilde, 1974). The J chain appears to be linked to the penultimate cysteine residue of the C terminal octapeptide of the alpha chain and potentially also the same octapeptide of the μ chain (Mestecky, Schrohenloher, Kulhavy, Wright and Tomana, 1974). The SC and J chain do not appear to be interconnected by disulphide bonds; however, the J chain may be necessary for the attachment of the SC (Eskeland and Brandtzaeg, 1974) rather than for the polymerization of the molecule (Halpern and Koshland, 1970). The SC is an unusual protein in that it appears to be totally deficient in methionine (Kobayashi, 1971). The J chain is clearly also found in other polymeric immunoglobulins such as IgM as shown by Metstecky, Ziram and Butler (1971), and Brandtzaeg (1973), and is thus not peculiar to secretory IgA or polymeric IgA molecules.

The SC in human secretory IgA is linked in the main to the IgA polymer by disulphide bridges (Tomasi and Bienenstock, 1968) whereas in the rabbit much of the secretory component is bound by non-covalent bonds. The SC is found in most secretions both in the free and bound forms. Antisera to the free form show antigenic (I) determinants (Brandtzaeg, 1971) which are inaccessible in the bound form and therefore such antisera can be used to identify free versus bound SC.

One version of the structure of the secretory IgA molecule is shown diagrammatically, in Figure 1.1 which is a composite drawing of data presented by a large number of workers in this field. This double Y configuration has been observed using electron-microscopy by Bloth and Svehag (1971) as the predominant form of secretory IgA in human colostrum. In gastrointestinal

secretions such as small intestinal fluid, approximately 33 per cent of the IgA is in the 7S IgA form (Bull and Tomasi, 1968) whereas parotid saliva usually contains less than 10 per cent of 7S IgA. It is important to understand that any attempt to estimate the amount of IgA and other immunoglobulins present in such secretions is fraught with a number of technical difficulties. Thus it is difficult to find a standard of satisfactory size; it is hard to retard proteolytic digestion or prevent losses in storage and during concentration; the external secretions particularly contain substances giving rise to non-immunologic precipitin lines and, if inflammation supervenes in the local mucosal surfaces, leakage of serum proteins may cause results which may be wrongly interpreted (Tomasi and Bienenstock, 1968, and Tomasi and Grey, 1972).

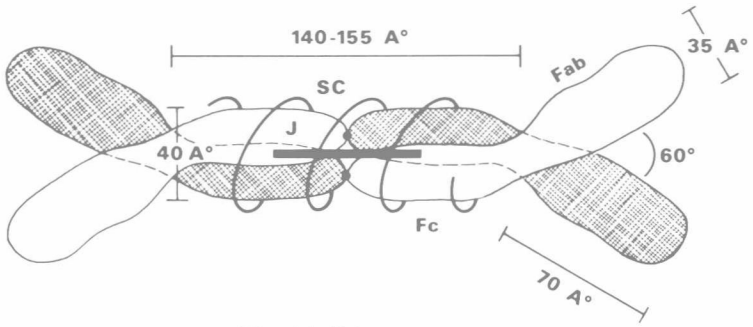

Fig. 1.1 Colostral 11S sIgA.

SYNTHESIS, ASSEMBLY AND TRANSPORT

IgA-containing cells predominate in the gastrointestinal tract as first shown by Crabbé and Heremans (1966). Heremans (1975) has estimated that the human gastrointestinal tract may contain 50 g of immunoglobulin containing lymphoid tissue, equivalent to the total cellular immunoglobulin content of the spleen. This lymphoid mass produces some 3 g of IgA per day, 50 per cent of which is secreted. The majority of the IgA-containing cells are found towards the bases of the villi, amongst the glands as well as near the Peyer's patches. The average half life of an IgA cell has been calculated at 4.7 days in the mouse (Mattioli and Tomasi, 1973).

The IgA appears to be synthesized as a dimer in most animals and in man, and to be locally synthesized in the adjacent mucosa in most tissues so far examined. The secretory component is thought to be synthesized by glandular epithelial cells but not goblet cells (Poger and Lamm, 1974; Brandtzaeg, 1974) and is probably concentrated by the Golgi apparatus (Van Munster, 1971). The J chain seems to be synthesized by the same cells as those making IgA (O'Daly and Cebra, 1971). The J chain may act as a clasp rather than a bracelet in joining two μ chains together, and a similar mechanism is suggested for the IgA polymer since only one mole of J chain appears to be present per mole of polymer (Koshland and Wilde, 1974).

The route of transport of the secretory IgA molecule

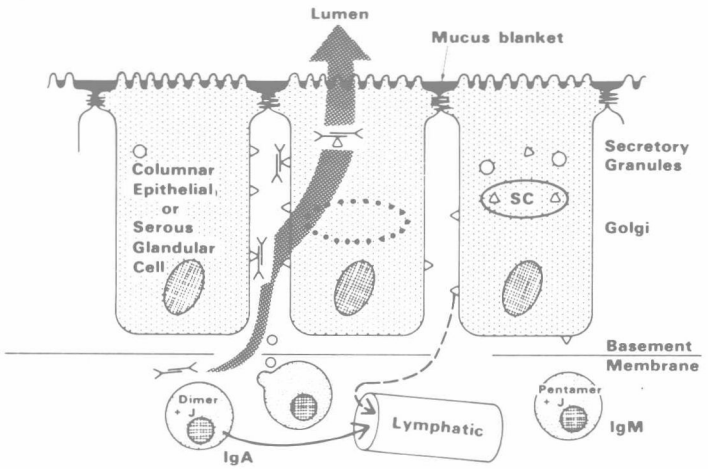

Fig. 1.2 Transport of immunoglobulins into external secretions.

is shown schematically in Figure 1.2. The IgA appears to pass between gaps found in the basement membrane (Tourville, Adler, Bienenstock and Tomasi, 1969) and is probably transported via a receptor mechanism consisting of secretory component on the outside of the cell membrane (Brandtzaeg, 1973) and is then transmitted to the luminal surface across the apical portion of the cell by exocytosis. Alternatively, membrane-bound packages of IgA follow the same route as has been suggested in the pig (Allen, Smith and Porter, 1973). There is some evidence that J chain may determine the selective combination not only of IgA but also IgM with secretory component (Eskeland and Brandtzaeg, 1974). Indeed further support that IgM may share a pathway with IgA comes from the observation that IgM purified from several secretions is 70 per cent saturated with secretory component (Brandtzaeg, 1973). In IgA deficiency, IgM often replaces IgA both in the experimental models of this condition and in man. The mode of transport of IgE into secretions is unknown. However, IgE does not bear secretory component when isolated from secretions. (Newcomb and Ishizaka, 1970).

In-vitro experiments have suggested a feedback control mechanism of the synthesis of the secretory IgA molecule (Lawton, Asofsky, Mage, 1970) since dimeric IgA stimulates synthesis of secretory component by rabbit mammary tissue in culture. Much of the locally synthesized IgA in most mammals, and possibly also in humans, may enter the lymphatics and be found in the circulation (Heremans and Vaerman, 1971). It is not clear whether the dimer in man or in animals has a special transport advantage especially since the predominant form of serum IgA in man is monomeric. However, Thompson and Asquith (1970) have shown secretory IgA in human serum, and an increased level of this in diseases associated with inflammation of mucous membranes. In most animals the majority of serum IgA is in the dimeric form and Dive and coworkers have shown that 50 per cent of biliary IgA in the dog emanates from serum (Heremans, 1975). Strober, Blaese and Waldmann (1970) have clearly shown that 96 per cent of salivary IgA is synthesized at local sites. Although small quantities of 7S serum IgA could pass into saliva, transport from serum to this secretion appeared to account for only a very small amount of the total. Thus most published evidence supports the view of local synthesis although some evidence exists to contradict it. The major IgA class exists in the form of two subclasses in the human: IgA1 and IgA2. In serum, IgA1 is the predominant subclass occupying approximately 80 per cent to 90 per cent of the total IgA, whereas in colostrum Grey, Able, Yount and Kunkel (1968) have shown that there was a relatively high percentage of IgA2. Whether this observation reflects a selective increase in IgA2 synthesizing cells in the breast, or some form of selective transport advantage of the IgA2 subclass, is not known.

ONTOGENY AND PHYLOGENY

Human newborn peripheral lymphoid intestinal tissue appears to lack IgA producing capacity (Van Furth, Schuit and Hijmans, 1965) and the same has been found to apply to rodents and the bovine species. The amount of IgA in cord blood is only 1 per cent of the normal adult serum level while that of IgM is about 20 per cent. Indeed, elevations of either IgM or IgA in cord blood have been considered as useful screening tests for the possibility of intra-uterine infections. Synthesis of IgG occurs as early as 10 weeks of gestation in the human foetus; synthesis of IgM by 12 weeks but no detectable IgA synthesis was found by Gitlin and Biasucci (1969) up to 32 weeks. The role of the intestinal contents in establishing the normal complement of immunoglobulin-containing cells in the intestine has clearly been shown by Crabbé, Nash, Bazin, Eyssen and Heremans (1970) in a study of conventionalization of germ-free mice. Even germ-free mice possess IgA-containing cells since dietary antigenic stimulation still occurs in such animals. There is no similar information available on ontogeny of development of the IgE class of immunoglobulin in terms of the aspects just discussed.

It is thought that IgA may be important in determining resistance to infection of mucosal surfaces. It has been recently suggested that the alpha chain may have diverged from the μ chain in evolution some 200 million years ago. This suggestion has been made on the basis of a comparison of amino acid sequences of human alpha, gamma and mu C terminal regions by Chuang, Capra and Kehoe (1973). Since mammals are considered to have appeared about 175 million years ago and birds diverged from reptiles about the same time this would place the IgA system further back than has been thought heretofore. The demonstration that chickens possess an IgA system supports this view (Lebacq-Verheyden, Vaerman and Heremans, 1972).

It is of further interest that the J chain appears to be a polypeptide of great evolutionary stability since Vaerman, Kobayashi and Heremans (1974) have shown extensive cross-reactivity of J chain across many species, including fish.

FUNCTIONS OF SECRETORY IgA

Antibody activity in this class has been shown to exist with specificity to bacteria, viruses, autoantigens, toxins and a variety of other antigens used to immunize experimental animals. The molecule has four combining sites and is more efficient in agglutination than IgG or the 7S IgA (Taubman and Genco, 1971). Human and rabbit secretory IgA surprisingly does not precipitate multivalent antigen, and although there are some differences in

this ability between these two species no really satisfactory explanation has yet been brought forward to account for these findings (Newcomb and Sutoris, 1974).

There is controversy over the ability of secretory IgA to opsonize (Zipursky, Brown and Bienenstock, 1973) a variety of antigens for phagocytosis (Kaplan, Dalmasso and Woodson, 1972). Most evidence currently weighs against opsonization either by peripheral blood phagocytes or by alveolar macrophages (Reynolds and Thompson, 1973). Secretory IgA will not activate complement by the classical pathway (Colten and Bienenstock, 1974), but the only study of 11S IgA antibodies so far reported in which human antiblood group A substance antibody was used has shown no activation of the alternate pathway (Colton and Bienenstock, 1974). Secretory IgA antibody blocks bacterial adherence to mucosal surfaces and thereby prevents colonization (Williams and Gibbons, 1972). It also may inhibit bacterial growth and change growth characteristics of bacteria (Brandtzaeg, Fjellanger and Gjervldsen, 1968). The molecule is markedly resistant to proteolysis (Tomasi and Bienenstock, 1968). This property in probably endowed by combination with SC, and is clearly an advantage in a proteolytic enzyme environment such as the gastrointestinal tract. The secretory IgA molecule binds to a variety of serum proteins, and the significance in this regard of the frequent combination with α-1-antitrypsin (Tomasi and Hauptman, 1974) is not known. It has been shown that IgA may act as a blocking antibody in terms of allergic reactions in nasal secretions (Turk, Lichtenstein and Norman, 1970) and that blocking of bacteriolysis may also be mediated in the same way (Hall, Manion and Zinneman, 1971). Adinolfi, Glynn, Lindsay and Milne (1966) have shown that together with a source of complement and lysozyme the antibody was effective in enhancement of bacteriolysis and recently this has been confirmed (Burdon, 1973; Hill and Porter, 1974). The secretory IgA antibody prevents absorption of albumin by rats after intragastric immunization (Heremans, 1975) and extensive studies by Walker, Isselbacher and Bloch (1973) have now elegantly shown that rat intestinal IgG1 is also effective in prevention of absorption of luminal antigen. In IgA-deficient humans, increased levels of serum antibody against dietary antigens have been recorded (Buckley and Dees, 1969) and infantile deficiency has been associated with subsequent atopy (Taylor *et al.*, 1973). It is quite possible that the function of secretory IgA to block or partially control antigen access to the organism may be an important modulating influence in human health and disease.

EPITHELIUM

Burnett (1959) first proposed the concept, later expanded by Heremans and Crabbé (1967), that antibodies lining the gastrointestinal tract may be regarded as an 'antiseptic paint'. It has recently been shown (Heremans, 1975) that mucin possesses cystein residues which may be available for protein interactions and that through this mechanism antibodies especially of the IgA class may be bound to the luminal surface of the epithelial cells of the intestine. In immunofluorescent studies of mucosal surfaces this layer of secretory IgA has clearly been demonstrated adherent to the apical surface of the columnar epithelial cells. The importance of this layer has been emphasized by experiments in which the action of a reducing agent on the mucous coat of immunized rat intestine allowed increased access of antigen across the epithelial layer (Walker and Bloch, personal communication).

The columnar epithelial layer consists of cells renewed in the crypts. The cells have microvilli on the luminal surface and synthesize and excrete into the lumen a number of proteins, one of which is the secretory component. The cells are capable of transporting luminal substances including carbohydrate, protein, fat, viruses and bacteria across the cell and basement membrane. It has long been known that in this cell layer are found cells variously referred to as lymphocytes and termed by Fichtelius (1968) thelio-lymphocytes. In the adult animal the ratio of these cells to epithelial cells in the small intestine is about 1:10. These cells are found in the intercellular spaces and can be recognized by their nuclei which are below the basal epithelial nuclei. The cell population is present in germ-free animals and in foetal intestine transplanted under the kidney capsule of syngeneic recipients (Ferguson and Parrott, 1972). In the chicken it is diminished in number by thymectomy or bursectomy (Bäck, 1970). Many of these cells possess granular inclusions (Collan, 1972) although they structurally resemble lymphocytes (Rudzik and Bienenstock, 1974). It has been suggested that some of these cells may be derived from mast cells on the basis of ultrastructural characteristics (Murray, Miller and Jarrett, 1968). These cells in the rabbit can be degranulated by PHA or Con A with the consequent release of histamine (Day and Bienenstock, unpublished). They may have a half life which differentiates them from the epithelial cells between which they reside (Darlington and Rogers, 1966) and from the lymphocytes in the lamina propria, and they therefore have been thought to represent a closed population. Whatever their derivation they have a resemblance both to lymphocytes and to mast cells and may act as specific passively sensitized sentinel cells at the mucosal surface. In hyperimmune pigs when antigen is placed into the lumen of a ligated segment of intestine a massive emigration of granulocytes appears within a few hours (Bellamy and Nielson, 1974) which does not leave any subsequent morphological evidence of per-

manent damage. The initiation of this stimulus is unknown but could conceivably be through the granular lymphocytes. This may also be the mechanism whereby pathotopic potentiation of mucosal surfaces occurs. This phenomenon originally observed and documented by Fazekas de St Groth and Donnelley (1950a and b) describes the influx of serum-borne immunity to a non-specifically stimulated site. Whether such cells indeed may be sensitized by IgE locally synthesized in the mucosa is not known. The older literature contains references to the importance of the intestinal tract in elimination of a variety of white cells (Ambrus and Ambrus, 1959; Teir, Rytömaa, Cederberg and Kiviniemi, 1963) and this aspect of immunity and defence has received little attention.

ORAL IMMUNIZATION

Oral immunization with a variety of antigens leads to local IgA antibody. Continued local feeding of protein antigens such as BSA in man and in animals eventually leads to a serum antibody response which in amount, class and affinity is indistinguishable from that seen following parenteral immunization (Rothberg, Kraft and Farr, 1967). The dissociation between serum and local antibody following oral immunization has been noted for more than 50 years (Davies, 1922). Oral immunization in man with Sabin vaccine led to a predominant IgA response in the intestinal secretions whereas parenteral immunization (Salk) did not lead to intestinal antibody (Ogra, Karzon, Righthand and MacGillivray, 1968). Both routes led to serum antibody. Similar results have been obtained with *Shigella* in monkeys and man, and with typhoid and cholera vaccines in man and animals (Shearman, Parkin and McClelland, 1972). The weight of this evidence suggests that the local response depends on the amount of antigen given orally and that attenuated live vaccines are effective in promoting resistance to infection, whereas killed local vaccines may be ineffective unless given repeatedly (Freter, 1962). Depending on the dose, parenteral immunization may lead to the appearance of local antibody in the intestine presumably due to dissemination of sufficient amounts of antigen to reach the intestinal lymphoid system (Shearman *et al.*, 1972).

The nature of the antigen used may also be highly significant. Hunter (1972) has shown that flagellin but not ovalbumin or bovine gammaglobulin will localize in the lamina propria of the unimmunized intestine and bronchus. Surprisingly, feeding of hapten (such as DNCB or picryl chloride) leads to tolerance and not immunity (Chase, 1946). However, feeding of ferritin to germ-free mice (Crabbé, Nash, Bazin, Eyssen and Heremans, 1969) led to a predominant IgA response in the serum, peripheral lymph nodes and intestine. Similar observations have been made following sheep red blood cell administration to germ-free (Bazin, Levi and Doria, 1970) as well as conventional animals (Andre, Bazin and Heremans, 1973). However, feeding of conventional animals with ferritin or BSA does not result in a predominant IgA response (Dolezel and Bienenstock, 1971a). The existence of prior immunity may well result in blocking of antigen uptake by the bowel and may influence the distribution and amount of antigen which is taken up into the portal circulation rather than the lymph.

ANTIGEN HANDLING

Oral feeding of BSA to hamsters (Dolezel and Bienenstock, 1971b) results in early appearance of antigen inside presumed intestinal macrophages. The observation that antigens will cross the intestinal wall and can be recovered intact in the circulation was first documented by Uhlenhuth (1900). Since then a wide variety of substances, which include bacteriophage, botulinus toxin, starch granules, ferritin, horse-radish peroxidase and BSA have been recorded in either portal or systemic circulation or lymph after oral feeding. The route for such absorption appears to be anatomically the reverse of that taken in the secretion of IgA (Warshaw, Walker, Cornell and Isselbacher, 1971). It is possible that secretion of proteins including antibody may share a common pathway (Kagnoff, Serfilippi and Donaldson, 1973) with absorption of proteins. Thus the antigen load could theoretically regulate antibody secretion.

S. typhi obtains entrance into the lamina propria and then passes to the draining lymph nodes via the regional Peyer's patches (Carter and Collins, 1974). Extensive parenteral immunization or oral immunization in rats has shown the formation of luminal immune complexes in the blockade of intestinal uptake (Walker, Wu, Isselbacher and Bloch, 1975) and luminal degradation. IgA (Heremans) and IgG1 (Walker, Isselbacher and Bloch, 1973) have similarly been incriminated in the rate of this process by two independent groups of observers. Removal of the mucous blanket by reducing agents led to increased uptake in preimmunized animals (Walker and Bloch, personal communication).

The mechanism of antigen handling may be crucial to the development or non-development of immunity. Oral administration of DNCB produces systemic tolerance in guinea-pigs. Such animals also have local mucosal tolerance and do not react to antigen introduced into the mucosal wall whereas following systemic sensitization a delayed type of hypersensitivity response occurs (Strauss and Bienenstock, unpublished). In this case the antigen appears mainly confined to the small

intestine (Silverman and Pomeranz, 1972). Similarly, tolerance has been achieved by the introduction of minute amounts of hapten into the portal system (Battisto and Miller, 1962). In dogs with porta-caval shunts the same experiment results in a systemic immune response (Cantor and Dumont, 1967). Similar experiments with analogous results have been performed with sheep red blood cells and hepatotoxins (Triger, Cynamon and Wright, 1973). The partitioning of antigen at the epithelial level either to the portal system or lymph, which may depend on the chemical nature of the antigen as well as other unknown factors, thus appears crucial in determining the nature of the immune response after oral immunization. Soluble antigen–antibody ratios favouring antigen cause increasingly inefficient removal of such complexes by the liver (Thomas and Vaez-Zadeh, 1974). Such compartmentalization in lymph or portal blood is well known to occur for lipids with more or less than 10–12 carbon atoms. Intragastric administration of sheep red cells to BALB/c mice for 4 days led to splenic tolerance for 12 weeks (Crabbé, Nash, Bazin, Eyssen and Heremans, 1969), and a primary type of response on secondary gastric immunization. The role of the Sulzberger–Chase phenomenon, effect of intestinal antibody, and partitioning of antigen between lymph and portal blood, result in a balance between immunity and tolerance. A better knowledge of this normal balance and maintenance of immunity is essential to understand how immune defences and control of bacterial flora of the intestine are maintained.

CELLULAR BASIS OF THE INTESTINAL IMMUNE RESPONSE

One of the most likely mechanisms whereby a local immune response occurs is that oral immunization results in the generation of immune competent cells in the intestine, and that these return after circulation. However, the oral immunization of rabbits by daily administration of 0.1 per cent to 2.0 per cent BSA in the drinking water produced no antigen reactive cells in the blood (Goldberg, Kraft, Peterson and Rothberg, 1971). These cells were found in the spleen whence they could be mobilized (Rothberg, Kraft, Asquith and Michalek, 1973). Such a circulation of cells following local immunization has also been suggested following BSA feeding of hamsters (Dolezel and Bienenstock, 1971a), sheep red blood cell administration to rats (André, Bazin and Heremans, 1973) and ferritin to germ-free mice (Crabbé, Nash, Bazin, Eyssen and Heremans, 1969). Following oral immunization sequential examination showed antibody-containing cells appeared earliest in the intestinal lamina propria followed by the mesenteric lymph node and spleen (André, Bazin and Heremans, 1973; Rothberg, Kraft and Michalek, 1973; Robertson and Cooper, 1973).

Blast cells in the thoracic duct (TD) consist mainly of non-recirculating cells (Moore and Hall, 1972) which home to the lamina propria of the intestine. Recent evidence suggests that the majority of these cells contain IgA, possess cell surface IgA and/or IgM, and synthesize almost exclusively dimeric IgA (Jensenius and Williams, 1974; Bienenstock, Rudzik, Clancy and Perey, 1974; Uhr and Vitetta, 1974). The majority of these cells may go on to IgA production. T-cell TD blasts also mainly home to the intestinal epithelium (Sprent and Miller, 1972; Guy-Grand, Griscelli and Vasalli, 1974). Although some homing occurs on the basis of antigen, TD blast cells home to neonatal unsuckled intestine (Halstead and Hall, 1972), as well as to subcutaneous transplants of isogeneic foetal intestine (Moore and Hall, 1972) in the same order as to normal intestine. Vitetta, Grundke-Iqbal, Holmes and Uhr (1974) have suggested that activated T-cells may be necessary for the switch to occur from IgM to other classes of immunoglobulins. Circumstantial evidence suggests that the thymus is implicated in IgA production since nude mice have little IgA, neonatal thymectomy of rabbits leads to depressed IgA and serum IgA antibody responses, and thymectomy added to bursectomy of neonatal chickens leads to total depletion of IgA (Perey and Bienenstock, 1973). Uhr and Vitetta (1974) found that in axenic mice most TD cells synthesized only IgM. Since axenic guinea-pigs are incapable of expressing delayed hypersensitivity (Lev and Battisto, 1970) they may lack the appropriate subpopulation of T-cells which when activated allow the switch from IgM to IgA to occur. Although there is still controversy as to whether or not IgM can switch directly to IgA, much circumstantial evidence supports this as one possibility (Martin and Leslie, 1974). However, the alternative suggestion of a switch from IgM to IgG to IgA (Cooper, Lawton and Kincade, 1972) has certainly not been excluded. The above synthesis would account for the repopulation of the intestinal lamina propria with IgM-containing cells in the chicken and human models of IgA deficiency and is supported by the frequent clinical observation of T-cell deficits in IgA deficiency. The discrepancy between two chicken models of IgA deficiency (bursectomy alone [Kincade and Cooper, 1973] as opposed to bursectomy and thymectomy [Perey and Bienenstock, 1973] at hatching) may be due to maturation differences in separate strains.

ROLE OF MUCOSAL LYMPHOID AGGREGATES

The Peyer's patches consist of multiple lymphoid aggregates covered by a specialized lymphoepithelium in-