THE HUMAN SECRETORY IMMUNOGLOBULIN SYSTEM: IMMUNOHISTOLOGICAL LOCALIZATION OF γA, SECRETORY "PIECE," AND LACTOFERRIN IN NORMAL HUMAN TISSUES*

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Studies from a number of laboratories have suggested that a local immunological system supplies immunoglobulins to the fluids that bathe certain mucous membrane surfaces. The predominance of γA producing plasma cells in the lamina propria of the rectum and colon (1–5), stomach (6–8), duodenum, and jejunum (1, 8, 9) has been reported. Furthermore, γA-producing plasma cells have been shown to predominate in the bronchial (10–11) and nasal (7) mucosa as well as in the parotid (12) salivary gland.

Immunofluorescent studies of human parotid salivary gland (12) showed that secretory "piece" was localized in acinar and ductular epithelial cells while γA was restricted to plasma cells of the interstitium, as well as the lumen of the glandular acini and ducts. On the basis of these results and other studies, a model was proposed for the synthesis of γA and secretory "piece" (*SP*) in separate cells and a possible mechanism was suggested by which these components might combine to form the 11S secretory γA molecule (12).

The present studies were undertaken to further investigate by means of immunofluorescent techniques, the cellular sites for γA and SP synthesis in a variety of normal human tissues. As a result of these investigations a revised model is presented involving at least in part, the intercellular (rather than intracellular) transport of γA and its complexing with SP in extracellular (intercellular and luminal surface) locations. Moreover, the cellular sites of synthesis of SP and its relationship to epithelial mucins have been more clearly delineated by studies in a variety of epithelial tissues.

Material and Methods

Tissue Specimens.—Immunohistochemical investigations were performed on various human tissue. 14 bronchial biopsies were obtained from two locations, eight from the carina and six

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from the middle lobe. In addition, tissues were obtained from three parotid and submaxillary glands, three small intestines, four colons, two gall bladders, six kidneys, two skin biopsies, and two pancreas specimens from organs removed surgically for various reasons. In each case samples of the tissue specimen were excised in areas remote from the lesion. Tissue specimens were judged normal or abnormal macroscopically, as well as on the basis of hematoxylin and eosin-stained sections examined under ordinary light microscopy.

Preparation of Tissues.—Specimens of the tissues were prepared for cryostat sectioning according to the method described by Eidelman and Davis (5). Specimens 1 or 2 mm thick were fixed in neutral isotonic buffered formaldehyde (10%) for 4 hr at 4–5°C followed by an overnight wash in 30% sucrose solution. The tissue was quick frozen in isopentane previously cooled to minimal temperature with liquid nitrogen. In addition, some tissues were frozen in isopentane without formaldehyde fixation. Consecutive sections approximately 4 μ thick were obtained from each tissue. The sections were air dried and stained immediately.

Fluoresceinated Antisera.—Fluorescein conjugated goat antiserum for human γG (Hyland) was made specific for the γchain by absorption with a γA myeloma protein. Antisera directed against human serum γA and γM were prepared in rabbits. An antiserum directed against human 11S colostral γA was produced in the goat. In addition, an antiserum directed against purified lactoferrin (*LF*) was produced in the rabbit. These antisera were conjugated with fluorescein isothiocyanate (FITC). The γA and γM antisera were made specific for the α and μ chain by absorption with γG. Colostral γA antisera was absorbed with γG plus LF to make it specific for secretory γA or concentrated normal human serum (NHS) plus LF to render it specific for SP. It must be emphasized here that this antiserum in addition to having SP activity also showed reactivity with one other component, LF, even though the 11S secretory γA used for immunization was shown to be homogeneous by all criteria applied including Ouchterlony analysis with potent antiwhole serum and antiwhole secretion antisera. For a more detailed discussion of the problem of the specificity of antisera against secretory components see Tomasi and Bienenstock (13). The LF was isolated by the method described by Tomasi and Bienenstock (13).

These five antisera were absorbed further with washed, mixed human ABO red blood cells, and rabbit brain and kidney powder to remove major blood group reactivity and to reduce background fluorescence.

Immunohistological Studies.—Consecutive sections of the various tissues were stained with the five fluoresceinated antisera for 1 hr at room temperature in a moist chamber. The sections were rinsed in phosphate buffered saline solution (PBS) pH 7.2 and then washed for 1 hr with five changes of PBS. Excess PBS surrounding the sections was blotted dry with bibulous paper. Two drops of glycerol/buffer (9/1) solution pH 8.0 were applied to the sections and then covered with a glass cover slip. The slides were examined immediately, under a Leitz ortholux microscope equipped with an osram HBO 200-watt high pressure mercury vapor lamp, UG 1 exciter filter, and a BG 38 suppression filter. Photographs were taken on a Kodak high speed Ektachrome (daylight) film. To facilitate localization of plasma cells, one of the consecutive sections from each tissue was stained by the May–Grünwald-Giemsa technique. Control experiments included incubation of sections of each tissue with fluoresceinated normal rabbit globulin and fluorescence blocking of γA, γG, and γM by prior incubation of the tissue with nonconjugated specific antisera. Blocking was also performed by absorption of the appropriate antiserum with purified preparations of γG, γM, and γA. Controls for SP staining included absorption of this antiserum with purified preparations of SP, secretory γA, and LF. The SP specific antiserum failed to stain lymph node and spleen tissue sections.

RESULTS

Bronchial Mucosa.—The staining patterns obtained with serum γA fluoresceinated antiserum were consistent in all the specimens tested. γA fluo-

rescence of bronchial mucosa from 14 biopsy specimens showed channels of staining in what were interpreted to be the intercellular spaces and also in the lumen of the glandular acini and ductules. No γA staining was seen in the cytoplasm of the epithelial cells with the exception of the apical surface of the glandular epithelial cells. Small numbers of interstitial "plasma" cells showed brilliant cytoplasmic staining.

Little or no epithelial cell or luminal staining occurred when γG or γM specific antisera were employed. Although no attempt was made to perform accurate quantitative fluorescent cell counts, it appeared by visual observation that the number of γG and γA staining cells were approximately equal, while γM cells were significantly fewer in number. The number of "plasma" cells varied considerably between biopsy specimens and appeared to be less than the number reported by Martinez-Tello and Blanc (10) for normal bronchial mucosa.

Staining with secretory γA fluoresceinated antiserum produced a similar pattern but in addition, epithelial cell cytoplasmic staining of both glandular acini and ductules was observed.

When SP antiserum was incubated with bronchial mucosal specimens no interstitial plasma cell staining occurred, however, brilliant fluorescence of the epithelium and the lumen of the glandular acini and ductules was observed.

It is noteworthy that SP staining was much more pronounced in the mucous type glandular epithelium with little staining in the serous demilunes of the mixed glands (see Fig. 2).

Salivary Glands.—γA staining of three submaxillary and parotid salivary glands revealed staining patterns quite similar to those obtained with bronchial biopsy tissues. γA staining was localized in what were probably the intercellular spaces between epithelial cells and in the lumen of the glandular acini and ductules. Interstitial "plasma" cell staining was, as in the case of the bronchial tissue, very few in number. Particularly brilliant, apical surface glandular epithelial cell staining was observed. However, no intracytoplasmic γA staining was seen other than the apical fluorescence mentioned above.

When tissue sections were incubated with γG or γM specific antisera, only slight staining of the glandular acinar and ductular epithelium of some specimens was observed. However, this was inconsistent and no intercellular or luminal staining was seen in any of the specimens. Scattered interstitial cells (few in number) stained for γA but not for SP. Occasional large focal collections of γA staining cells were seen, particularly in the submaxillary tissues. Very few and in most sections, no interstitial γG or γM staining plasma cells were observed.

SP staining was pronounced in the glandular acinar and ductular epithelium as well as in the lumen of these structures. Fig. 2 shows typical staining in the ductular epithelium of the parotid gland.

As in the case of the bronchial mucosae, the mucous type acinar epithelial

cells of the submaxillary gland stained brilliantly while no staining of the serous type cells was observed.

A point of particular emphasis is the fact that parotid salivary gland tissue is composed almost entirely of serous type glands (14). As pointed out above in the bronchial and submaxillary glands, SP staining was much more intense in the mucous secreting cells rather than the serous type. It is of interest therefore that the parotid epithelial cells showed positive staining for SP. However, in many parotid glandular acini only a few epithelial cells stained for SP, and in a number of acini no staining was observed.

Intestinal Tract.—Immunofluorescence studies of the GI mucosa were performed on human small intestine and colon. In all specimens examined a marked predominance of γA staining plasma cells was observed in the lamina propria. The majority of these γA staining cells were concentrated in the lamina propria basal to the secretory glands, while relatively fewer cells were present in the interglandular lamina propria. Heavy linear γA staining of the basement membrane area as well as intercellular spaces and the apical surface of the glandular epithelium was observed (see Fig. 3). γA staining was also observed in the lumen of the secretory glands.

γM and γG staining of consecutive sections failed to show staining in the areas described above for γA. The relative predominance of γA, γG, and γM staining "plasma" cells in the GI mucosa was markedly different from that observed in the bronchial mucosa. In the latter case the number of γA and γG staining plasma cells were approximately the same, γM being the least. However,in the GI mucosa γA staining plasma cells far outnumbered γG or γM. It is also interesting to note that γM plasma cells exceeded the number of γG plasma cells in the GI mucosa while the reverse was observed in the bronchial interstitium. These results were repeatedly observed when consecutive sections of the various specimens were stained in adjacent areas with the respective antisera and also by examining photographs taken of adjacent areas of consecutive sections stained with the different monospecific antisera.

SP staining was pronounced in the goblet cells, intercellular glandular epithelium, and lumen. In addition, linear surface fluorescence of the glandular epithelium was particularly striking. This linear surface staining was also observed in some tissue specimens when γA specific antiserum was employed. Photographs of representative sections are shown in Figs. 3 and 4.

Lactoferrin and Eosinophil Staining.—The results obtained with monospecific LF antiserum differed markedly from those obtained with SP antiserum. Consecutive sections of each tissue were stained with LF antiserum. In bronchial mucosa, parotid, and submaxillary glands LF staining appeared to be granular or speckled in contrast to the diffuse SP staining that was observed in the mucous acini of these tissues. Brilliant luminal fluorescence of the acini was observed. However, unlike SP no staining of the epithelium lining the duc-

tules was demonstrable. When small and large bowel sections were stained with LF antiserum, no staining of goblet cells or any other area was observed except for nonspecific eosinophil cellular fluorescence which was a constant observation throughout this investigation with all the various antisera. This type of fluorescent staining can be very misleading in the analysis of the distribution of staining and should be checked with appropriate histological staining methods. This was accomplished by May-Grünwald-Giemsa staining of the same tissue sections that had been previously stained with the fluorescent reagents.

Gall Bladder, Kidney, Pancreas, and Sweat Glands.—Fluorescent antibody studies of two human gall bladder specimens revealed γA staining in interstitial lymphoid cells and in the luminal surface mucous of the epithelium. Sections incubated with γG and γM antisera showed no epithelial cell staining; however, large numbers of γG staining interstitial cells and fewer γM staining cells were observed.

SP staining was striking in the gall bladder epithelium and surface mucous layer (see Fig. 2).

No staining of six normal human kidney specimens was observed with γA, γG, or γM antisera.

SP staining was localized in the cytoplasm of the tubular epithelium of two kidney specimens (fig. 2). In four specimens, SP staining appeared as a linear deposit on the surface of the tubular epithelium.

SP staining was very brilliant in the lumen of some tubules, however, no staining of the glomeruli was observed.

Incubation of pancreas tissue sections with γA, γG, and γM specific antisera failed to demonstrate immunofluorescent staining although SP staining was observed in many glandular epithelial cells and in the ductular epithelium as shown in Fig. 4.

Two skin biopsy specimens failed to stain with γA, γG, and γM antisera. However, when SP antiserum was incubated with skin tissue marked fluorescence of the secretory units of the sweat glands was observed. In addition, apical ductular epithelial and luminal staining was demonstrable. SP staining of pancreas and the secretory units of sweat glands are illustrated in fig. 4.

It is interesting to note that LF staining was not observed in gall bladder, pancreas, and sweat glands but was localized in the tubular epithelium of the kidney. The localization of SP and LF staining in the various tissues are summarized in Table I.

DISCUSSION

A major problem in interpreting fluorescent antibody studies of this type arises because of the difficulty in obtaining monospecific antisera. The fluorescent antibody technique is quite sensitive and will detect small amounts of

antibody which may be difficult to measure by other methods. Nearly all the SP antisera that we have examined to date reacted with LF in addition to SP and for this reason care must be exercised to avoid misinterpretation of the staining which resulted when these antisera were employed. The SP antiserum used in this work was absorbed with purified LF (as well as NHS) until no trace of this component was detectable by double diffusion in agar gel. The particular bleeding of antiserum used in these studies did not contain demonstrable antibodies against another secretion specific component called macromolecular component (MMC) (13) and so after absorption with LF and NHS was considered specific for SP. If the SP antiserum was absorbed with secretory γA virtually complete fluorescence blocking was obtained even though the

TABLE I

Localization of LF and SP in the Epithelium of Various Tissues with Fluoresceinated Antisera

Type of epithelium	Antiserum	
	LF	SP
GI rectum goblet cells	Neg.	Pos.
Bronchial goblet cells	Neg.	Pos.
Bronchial acinar cells	Pos.	Pos.
Submaxillary mucous acinar cells	Neg.	Pos.
Submaxillary serous acinar cells	Pos.	Neg.
Parotid acinar cells	Pos.	Pos.
Ductules of parotid, submaxillary, and bronchial mucosa	Neg.	Pos.
Gall bladder	Neg.	Pos.
Pancreas acini and ductules	Neg.	Pos.
Kidney-tubules	Pos.	Pos.

antiserum still reacted with LF on Ouchterlony analysis. Absorption of SP antiserum with free SP also resulted in fluorescence blocking.

If the SP antiserum, after the precautions that were taken, can be considered to react specifically with SP as the evidence indicates, then a model can be conceived in which γA and SP are synthesized in different cells. The γA produced by the majority of the plasma cell population in the lamina propria, is transported through the glandular or basement membrane into the intercellular spaces between the epithelial cells. The heavy concentration of γA along the basement membrane suggests that this structure may present a significant barrier to the diffusion of γA. The complete absence of SP staining in this location and in the interstitial regions is another indication of its formation on the luminal side of the basement membrane, e.g. in the epithelial cell.

The fluorescent photographs show a striking linear network of γA staining indicating that transport is occurring through defined channels. It seems probable on the basis of the morphological appearance of the staining patterns (see Fig. 3) that transport is occurring largely through intercellular channels although this remains to be proven. In this regard the only defined channels which are described in electron microscopic studies of intestinal epithelium are intercellular (15). Such studies on rat intestine show open intercellular channels of approximately 200–300 A except at the apical surface of the cell where

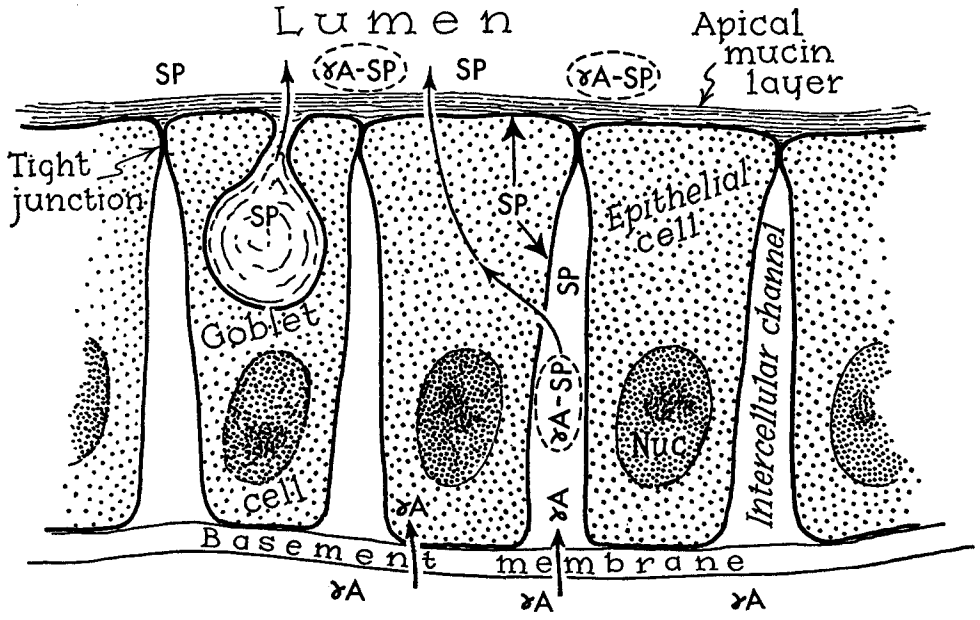

FIG. 1. A hypothetical model for the transport of γA and *SP* across mucosal membrane epithelium.

the cell membranes of adjacent columnar cells are actually in apposition, the so called tight junction or zona occludens. The tight junction at least in certain epithelium appears to prevent luminal macromolecules from directly reaching the intercellular space (15) although in other tissues, transport of macromolecules the size of horseradish peroxidase (MW 43,000) appears to take place from the lumen across the cell via intercellular channels (16). If the tight junction of the epithelium under discussion here, particularly the GI tract, does indeed effectively prevent macromolecules (the size of γA) present in the intercellular space from passing directly into the lumen then a mechanism such as that outlined in Fig. 1 might be operative. According to this hypothe-

sis the γA would pass through the basement membrane along intercellular channels until it reached the apical end. Being prevented by the tight junction from reaching the lumen via the intercellular route it would then pass into the cytoplasm of the epithelial cell and from there through the cell membrane into the lumen. Such a mechanism is consistent with the fluorescent data showing γA staining along the basement membrane, intercellular spaces, and in the apical surface of the epithelial cell but not elsewhere in the cytoplasm. The localization of γA in the GI mucosae described above is essentially identical with that described by Hugon and Borgers (17) for horseradish peroxidase in the mouse duodenum. It is of course possible that the apical staining is "nonspecific" in regard to the proposed mechanism perhaps resulting from surface absorption due to the high concentration of γA in the lumen. It is also recognized that a single mechanism may not apply to all epithelial membranes. As mentioned above some evidence is already available that capillary endothelium varies in its permeability to macromolecules in different anatomical locations (16), and in the kidney the "tightness" of the apical junctions of epithelial cells varies in different parts of the renal tubule (15). Thus in some tissues there could be direct access from the intercellular space to the lumen. Moreover the possibility that significant transport of γA may occur through the cytoplasm of the cell has not been excluded. It may be that the concentration of γA in the cytoplasm of the cell where it is relatively unconfined compared with that in the intercellular channels, may be insufficient to be detected using the present fluorescent technique.

Since both γA and SP staining occurs in what we have interpreted as intercellular spaces it is conceivable that combination of γA and SP takes place in these spaces during the transport process. Whether transport of γA occurs by simple diffusion or some more active mechanism is still a matter of conjecture. Likewise, the role of the SP in facilitating transport of γA as originally suggested (12, 18) remains completely unproven. It is apparent that in some secretions SP is produced in excess of γA and appears free in varying amounts in different secretions. On the other hand, evidence has been presented in our laboratory (unpublished) that nearly all of the γA in most secretions is complexed with SP. The small amount of free uncomplexed 7Sγ A that appears in some secretions may be derived directly from serum (especially if inflammation is present) and/or may be dissociated from the 11S secretory molecule during isolation procedures.

The occurrence of SP in the epithelial cells of the human sweat gland in the absence of observable staining for γA (or other immunoglobulins) is of considerable interest in regard to the function of SP. It seems that if SP has any

function in human sweat it is probably not that of transport or stabilization of the γA molecule as has been suggested for SP in other external secretions. This again points to the necessity for further characterization of SP before any far reaching conclusions are drawn concerning its function in the γA secretory system.

The intracellular staining of goblet cells in the GI mucosa giving a grape-like pattern to the fluorescent photographs indicates association of the SP with intracellular mucins. Likewise in the submaxillary gland where approximately 20% of the glandular acini are of the mucous type (14), SP staining occurred primarily in the mucous cells with little or no fluorescence occurring in the serous demilunes. However, SP is also found in cells which are not classically goblet or mucous cells morphologically such as those lining the ducts of the salivary glands, parotid acini, sweat glands, the pancreas, and the epithelium of the gall bladder. It appears therefore, that SP is a glycoprotein which is rather ubiquitous in epithelial tissues. The distribution of staining for SP compared to LF also suggests that differential epithelial function does exist. LF staining was not observed in the rectal mucosa, pancreas, sweat glands, and gall bladder where SP fluorescence was particularly prominent, whereas both were found in the parotid, and submaxillary salivary glands, and in bronchial glandular epithelial cells. However it has not been shown whether LF and SP are produced in the same epithelial cell in these tissues. Monkey ileum stained with anti-human SP (but not anti-human LF) and the staining pattern was very similar to that seen with human ileum. The distribution of both SP and LF determined by the fluorescent studies agrees well with the immunological determination of these components in the corresponding fluids. The studies reported here are not consistent with those reporting intra-epithelial cell γA staining in rabbit intestinal epithelium (19), parotid acini cells (20), and renal tubular epithelium (21), but are in agreement with those showing primarily an apical distribution in the human rectal mucosa and small intestine (3, 4, 22). Our findings are similar to those of Heremans and Crabbé (22) and we are in general agreement with the hypothesis developed by these workers concerning the transport of γA. In addition these workers showed apical and radial intercellular fluorescence due to γM in a patient who had γA deficient sprue and whose secretions contained predominantly γM. This suggests that a similar (intercellular) mechanism occurs for the transport of γM as for γA. In this regard Brandtzaeg et al. (23) recently demonstrated in a patient with hereditary telangiectasia and absent γA that the γM which occurs in these secretions appears to lack secretory "piece." If this finding is substantiated, it clearly shows the transport of an immunoglobulin macromolecule without SP and again raises a serious question regarding its transport function.

The results of the cellular localization of γA and SP reported here are in direct contrast of those reported by Rossen et al. (24). These workers reported γA and SP staining occurring together within individual cells of bronchial and nasal mucosa as well as submaxillary tissue. These cells were thought to be of the lymphoid-plasma cell line and the authors suggested that SP and γA are synthesized within the same cell. This type of staining was never observed in any of our previous or present work and has not been reported by other investigators (10, 22). The reason for this difference is not entirely clear. In addition to the problem of specificity of the antisera it should be emphasized that epithelial cells often break off from the acini particularly in diseased tissue and are difficult to identify as epithelial cells by fluorescent microscopy. Moreover nonspecific staining of eosinophil cells is observed with most antisera as well as normal rabbit globulins. Such reactions of the eosinophil cells are well known (25) and these cells were readily distinguished from lymphoid plasma cells by staining sections previously stained with fluoresceinated antiserum, or consecutive sections with the May-Grünwald-Giemsa stain.

One disturbing feature in regard to the thesis of local production of γA concerns the relative paucity of γA producing plasma-like cells in the interstitium of the bronchial mucosa and salivary glands. In both of these tissues, although heavy concentrations of γA can be seen localized in the lumen of the ducts and on the periphery of the gland as well as in intercellular locations, few interstitial plasma cells are seen either by fluorescent staining or light microscopy. It is interesting however, that in multiple sections of these tissues one often sees sizable focal collections of γA staining plasma cells. These are not definite lymph node structures but it is possible that these collections are the origin of a significant fraction of the γA in the salivary and respiratory tract secretions. Thus in the GI tract there is an extensive linear infiltration beneath the mucous membrane, whereas certain other tissues such as those mentioned above the immunoglobulin synthesizing cells are present in more scattered focal collections.

Although the majority of the studies to date suggest that the γA found in human external secretions arises from local synthesis, evidence is available suggesting that there may be differences in the mechanisms of secretion of immunoglobulins depending on both the organ and the species. For example, in the bovine mammary gland there appears to be selective transport from serum of a fast γG1 immunoglobulin and little evidence is available to support significant local synthesis (26). Similar but less complete studies suggest that the bovine salivary immunoglobulins are derived predominantly from serum (27). It will therefore be of considerable interest to study various organs in different species in order to determine whether the mechanism of secretions

of immunoglobulins in external secretions is species specific, organ specific, or both.

SUMMARY

The immunohistological localization of γA, secretory "piece" (*SP*), and lactoferrin (*LF*) in the mucosae of a variety of normal human tissues was investigated using specific fluoresceinated antisera. γA staining was localized in the apical portion of the mucosal epithelium, intercellular spaces, basement membrane area, and plasma cells of the interstitium or lamina propria of a number of normal human tissues.

SP was ubiquitous in the mucosal epithelium of all tissues studied which included parotid and submaxillary glands, bronchi, pancreas, GI tract, sweat glands, kidney, and gall bladder. In addition, SP staining was localized in the intercellular spaces and on the surface of the epithelial cells lining the lumen of the secretory glands. No SP staining was observed in the plasma cells of the interstitium or lamina propria surrounding the secretory glands in these tissues, and no SP staining was observed in sections of normal spleen or lymph node tissue. SP staining was observed in the sweat glands, pancreas, and kidney in the absence of γA staining.

LF was much less ubiquitous in the epithelial cells of the various tissues studied and appeared to be restricted primarily to the acinar epithelium of the bronchial mucosae, parotid, and submaxillary salivary glands, and was also found in renal tubular cells.

A hypothetical model for the transport of γA and SP across mucosal membrane epithelium is presented.

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FIG. 2. Immunofluorescence localization of SP in various normal human tissues with fluorescein labeled SP antiserum. (*a*) Bronchial biopsy specimen showing SP staining in the mucous acinar cells of mixed glands with no staining in the serous demilunes (arrows and dashed line). (*b*) Section of parotid salivary gland tissue. Note intense SP staining localized in the epithelium of a ductule. (*c*) Kidney biopsy specimen illustrating SP staining in the tubular epithelium. The fluorescence surrounding the tubules is due to bluish tissue autofluorescence (arrows). (*d*) Section of gall bladder tissue showing intense SP staining in the epithelial cells. × 675.

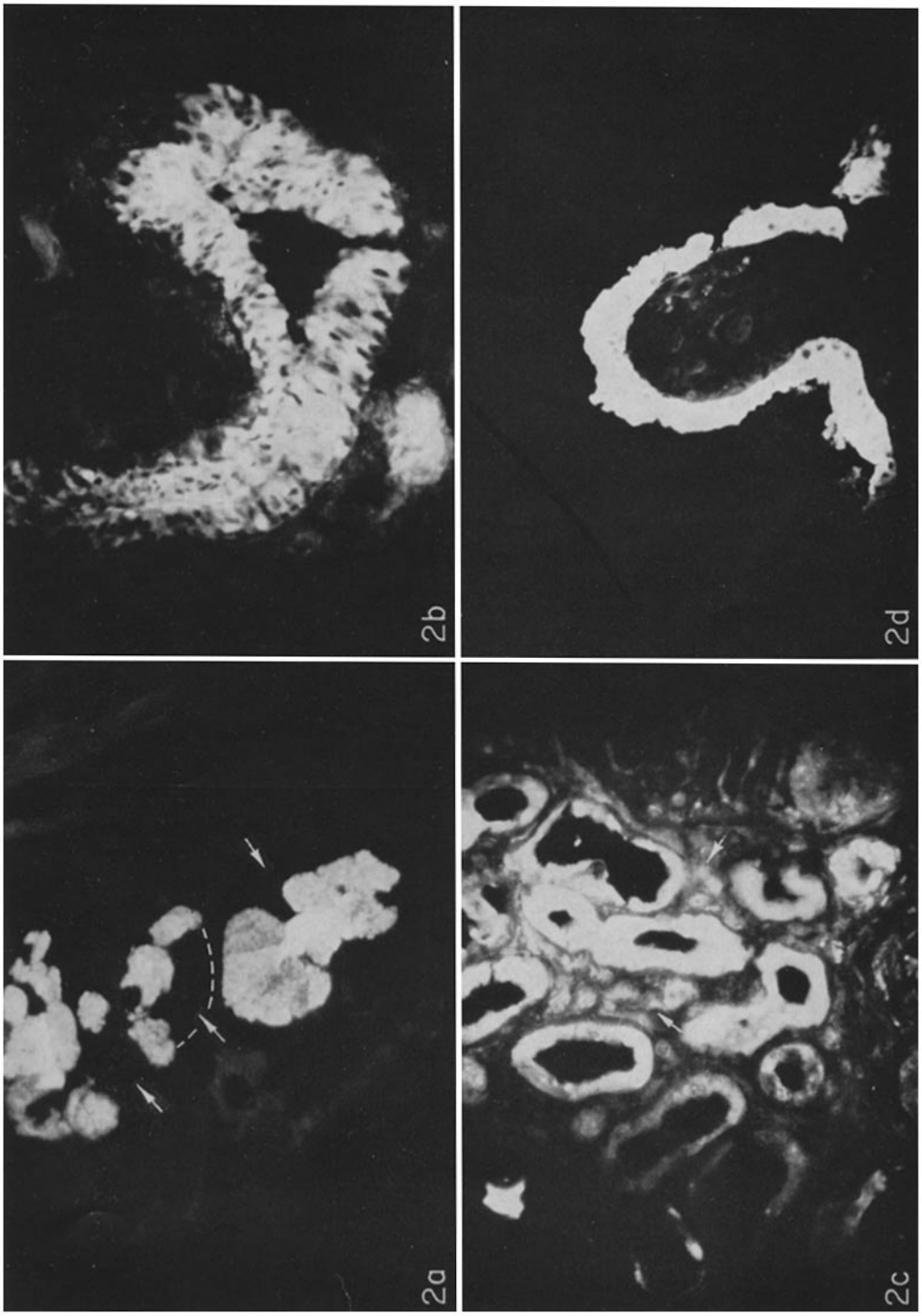
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FIG. 3. Immunofluorescence localization of γA and SP in human colonic mucosae. (*a*) Section of normal colon stained with 7SγA antiserum. Note γA fluorescence of plasma and lymphoid cells within the lamina propria (*LP*) along the basement membrane (*BM*) area and in the intercellular spaces (*IS*) between the glandular epithelial cells with no staining in the goblet cells (*GC*). × 675. (*b*) Section of same tissue as in (*a*) stained with 7SγA antiserum (low power magnification). Note the relatively weak γA staining along the luminal surface of the epithelial cells (arrow) that line the lumen (*L*) and the intense γA staining in the apical portion of the glandular epithelium with no staining in the goblet cells. Particularly heavy basement membrane (*BM*) staining is evident. × 270. (*c*) Section of colon showing glands cut in cross section stained with 7SγA antiserum. Note intense γA staining of the basement membrane area, and the intercellular spaces. × 675. (*d*) Section of colon stained with dilute secretory 11SγA antiserum. Linear SP staining on the surface of the epithelial cells (arrow) lining the lumen (*L*) and weak goblet cell (*GC*) staining is seen. γA fluorescence is localized along the basement membrane area (*BM*), intercellular spaces, and in plasma cells located in the lamina propria. × 675.

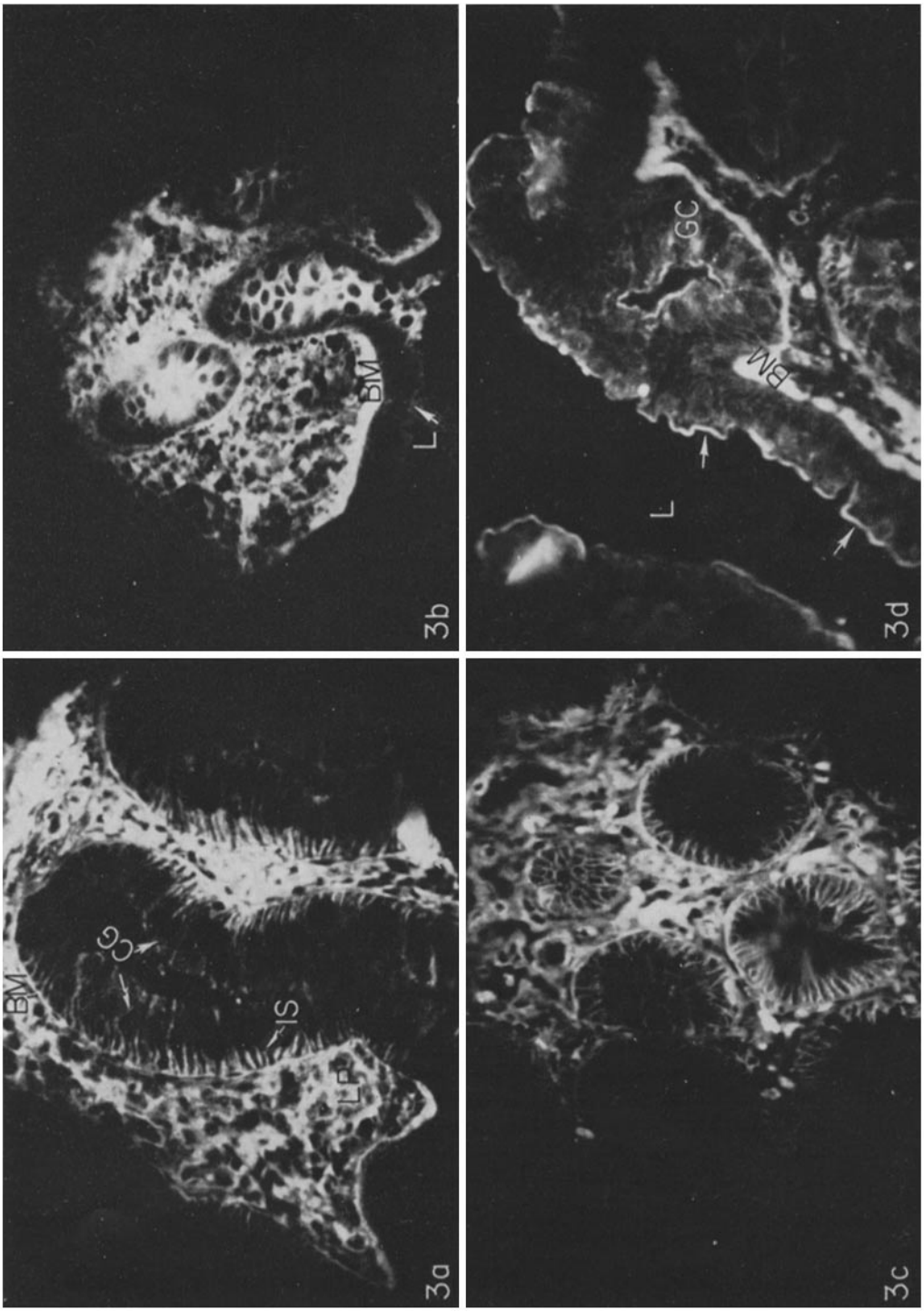
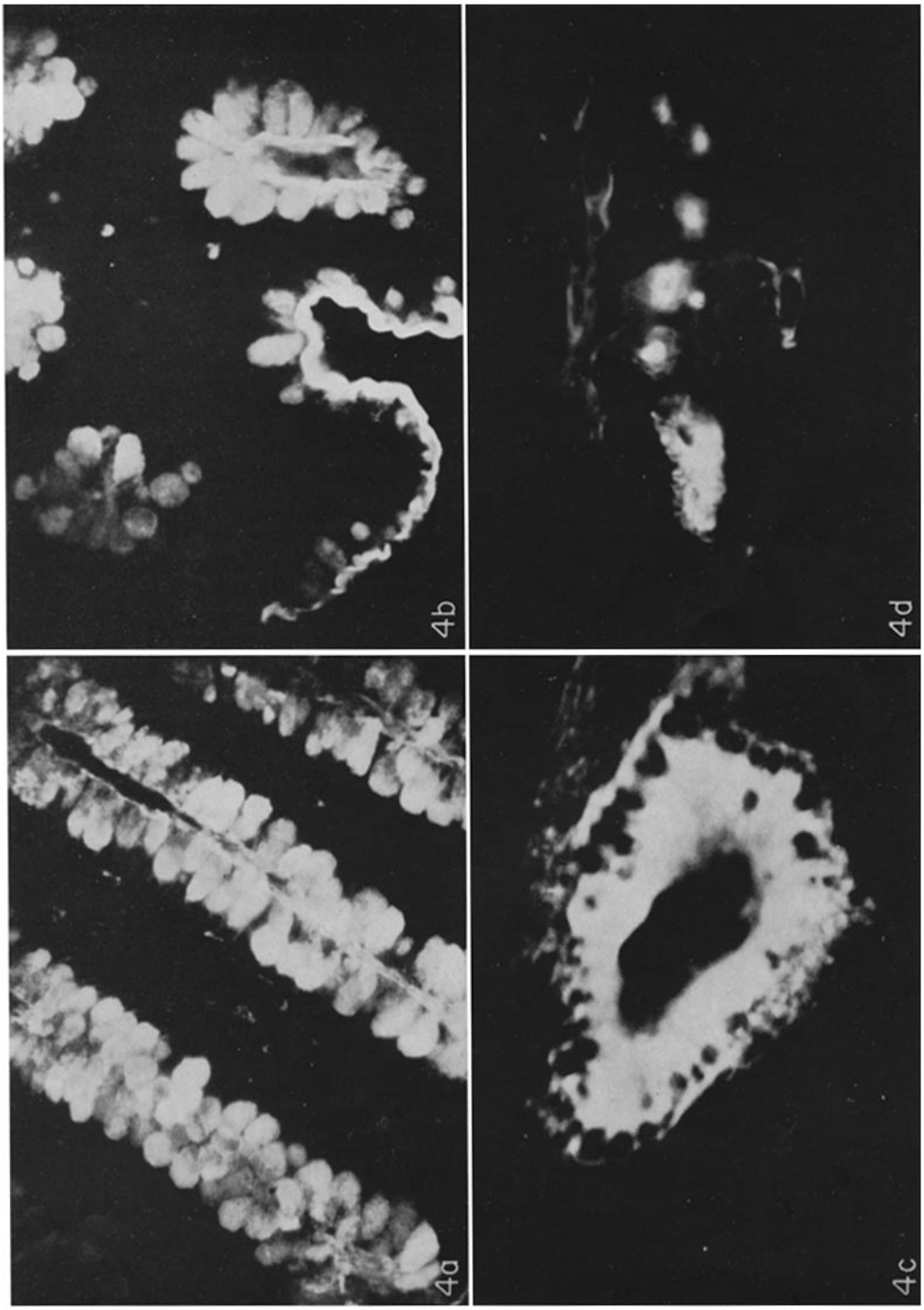

3b
BM
L
3d
GC
BM
L
3a
BM
G
IS
LP
3c

FIG. 4. SP staining in human colon, pancreas, and sweat glands. (*a*) Section of colon stained with undiluted SP antiserum showing SP staining localized in goblet cells and even more intense linear surface staining along the lumen of the glands. Note the absence of staining in the lamina propria except for occasional nonspecific eosinophil cell staining. × 675. (*b*) Section of colon showing cross section of glands stained with SP antiserum. SP staining is localized in the goblet cells and along the surface of the epithelium lining the lumen. × 675. (*c*) Section of pancreas tissue showing SP staining in the ductular epithelial cells. × 675. (*d*) Skin biopsy section showing SP staining localized in sweat glands (low power). × 270.


4b
4d
4a
4c