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# The Sebaceous Gland Antigen Defined by the OM-1 Monoclonal Antibody is Expressed at High Density on the Surface of Ovarian Carcinoma Cells*

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**Abstract**—*A monoclonal antibody, designated OM-1, was raised against ovarian serous papillary cystadenocarcinoma (stage IV) cells. This antibody was found to react strongly with primary and metastatic ovarian serous cystadenocarcinomas and endometrioid carcinomas but the antigen detected was either absent or at very low levels in ovarian mucinous adenocarcinomas, clear cell carcinomas, benign serous and mucinous cystadenomas and Brenner tumours. The OM-1 antibody gave no detectable reaction with 93 other human tumours, including examples of breast and colon adenocarcinomas. In normal tissues the OM-1 antibody reacted with normal sebaceous gland cells, lung type II pneumocytes and placental syncytial trophoblasts. In the normal ovary OM-1 reactivity was confined to extremely weak staining of the surface epithelium. No reaction with any other ovarian cell type could be detected. No evidence of reaction with other normal cell populations present in 24 adult and seven foetal tissues was found. The antigen detected is compared with other ovarian tumour-associated antigens. The OM-1 antibody is likely to prove of value in the detection and diagnosis of ovarian carcinoma.*

## INTRODUCTION

OVARIAN cancer is the most lethal of gynaecological malignancies. Prognosis is generally poor as, due to the asymptomatic character of this disease, over 70% of affected women present with metastases [1]. Consequently, there is real interest in the development of ovarian tumour markers, particularly to aid in the early detection of disease and in the identification of metastatic tumour [2].

Many tumour markers, such as carcinoembryonic antigen (CEA), have been evaluated with respect to ovarian cancer. None of these has proved valuable [3, 4]. A number of antibodies to ovarian tumour-associated antigens have been described [5–7]. These antibodies vary in the specificity of their reaction with the different ovarian tumour types and with normal tissues. Of particular note are the monoclonal antibody described by Bhattacharya and colleagues [8], which is found on ovarian mucinous but not serous cystadenocarcinoma cells, and the OC125 monoclonal antibody developed by Bast and coworkers [9, 10], which reacts with ovarian serous but not mucinous cystadenocarcinomas.

In this paper we report the production and characterization of the murine monoclonal antibody OM-1, which reacts with a cell surface molecule expressed at high levels by ovarian serous adenocarcinomas and endometrioid carcinomas. This antigen was either not detected or present at very low levels in benign ovarian tumours and ovarian mucinous and clear cell carcinomas. Although not detected in foetal tissues, the OM-1-defined antigen was highly

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expressed by normal sebaceous gland cells and has consequently been designated sebaceous gland antigen (SGA).

## MATERIALS AND METHODS

*Cells and cell lines*

Normal blood and bone marrow samples were obtained from healthy donors. Mononuclear cells were separated by Ficoll density gradient centrifugation [11] and immediately tested. Cells from pleural and peritoneal effusions were collected by centrifugation, washed in buffered saline and immediately used for cytocentrifuge preparations (see below). Established cell lines used in this study were grown in suspension or monolayer culture in RPMI 1640 medium (Flow Laboratories, North Ryde, Australia), supplemented with 10% foetal calf serum (Flow Labs) and 40 $\mu$g/ml gentamycin (Roussel Pharmaceuticals, Castle Hill, Australia).

*Hybridoma production*

Human serous papillary cystadenocarcinoma cells were obtained from a pleural effusion in a 66-yr-old female patient. The sample comprised approximately 50% adenocarcinoma cells, the remaining cells being predominantly macrophages. Cells were stored in liquid nitrogen as a viable suspension in sterile aliquots. For production of hybridomas, adenocarcinoma cells were thawed and used as immunogen without further purification. Mice (BALB/c) were initially immunized intraperitoneally with $10^7$ viable cells and boosted intravenously 3 weeks later with a further $10^7$ viable cells. Subsequently, the spleen from an immunized mouse was removed after 4 days, fused with the P3-NS1/1-Ag 4.1 (NS1) mouse myeloma and hybrids propogated [12]. The hybrid OM-1 was selected on the basis of its reactivity with cell lines (see below) and was cloned by repeated successive limiting dilution [13].

*Purification of OM-1 antibody*

The cloned hybrid OM-1 was injected intraperitoneally into BALB/c mice ($10^6$ cells/mouse) and ascitic fluid collected 14–28 days later. The monoclonal antibody was purified from the ascitic fluid by ammonium sulphate precipitation [14] followed by protein A affinity chromatography [15]. The elution pattern of the antibody from the protein A column was consistent with it being an $IgG_{2b}$ antibody. However, immune fixation experiments [16] assessing the reaction of rabbit antisera specific for mouse immunoglobulin classes and subclasses (Miles Laboratories, Melbourne) with the OM-1 antibody have identified it as being of the IgM class. The protein concentration of the active fraction was determined [17] and the purified antibody stored at −70°C in aliquots at a concentration of 1 mg/ml. The maximum dilution of the 1 mg/ml stock solution of purified antibody which still gave maximal reaction by ELISA (see below) with the Colo 316 ovarian cell line was 1:1000. All results of all assays reported here were obtained using this purified antibody stock solution at a dilution of 1:500.

*Enzyme-linked immunoadsorbent assay (ELISA)*

The reactivity of the OM-1 antibody with solid tumour-derived cell lines was assessed by an indirect ELISA [18] incorporating horseradish peroxidase as enzyme on cells cultured in 96-well multi-titre trays (Costar, Disposable Products, Melbourne). Cells were fixed with 0.0025% glutaraldehyde prior to use. After a 2-hr incubation at 20°C with the test antibody, the cells were washed three times and incubated for 2 hr at 20°C with horseradish peroxidase-conjugated rabbit anti-mouse IgG antibodies. After washing, the enzyme substrate *o*-phenyldiamine (Sigma Chemical Company, St. Louis, MO) was added and reaction allowed to proceed at 20°C for 15 min. Reaction product was measured on a Titretek Multiscan (Flow Labs). The pan-human monoclonal antibody CI-panHu [de Kretser *et al.* submitted for publication], diluted 1:100, was used as positive control; normal mouse serum, diluted 1:100 in appropriate buffer, was used as negative control for all assays.

*Indirect immunofluorescence*

The reactivity of the OM-1 antibody with haematopoietic cell populations and cell lines was determined by indirect immunofluorescence [19]. Cell samples ($10^6$ cells/test) were incubated for 5 min at 20°C with 50 $\mu$l of test antibody, washed once in buffered saline and further incubated for 5 min at 20°C with 50 $\mu$l of fluorescein-conjugated goat anti-mouse IgG $(Fab)'_2$ fragment (Cappel Laboratories, West Chester, PA), diluted 1:20 in buffered saline. Cells were washed twice in buffered saline and fluorescence analysed on an Ortho System 50 cytofluorograph (Ortho Diagnostic Systems, North Ryde, Australia). Peripheral blood cell populations were separated using the 90° forward scatter and axial light loss parameters. The identity of separated populations was confirmed by indirect immunofluorescence using commercially available cell type-specific antibodies. The positive and negative control antibodies were as described for the ELISA.

*Immunohistochemistry*

The reactivity of the OM-1 antibody with cells in sections from various normal and malignant adult tissues and foetal tissues was assessed by a four-step immunoperoxidase staining technique [20]. Solid tissues were either poly-L-lysine-fixed for frozen sections [20] or Bouin's-fixed and paraffin-embedded as for routine pathology specimens [21]. Cytocentrifuge preparations (Shandon, London) of normal and leukaemic blood and bone marrow samples were tested after formol-saline fixation [22]. The OM-1 antibody was again used at a dilution of 1:500; positive and negative control antibodies were as described for the ELISA.

## RESULTS

*Reactivity with cell lines*

Of the 13 solid tumour-derived cell lines tested by ELISA, the OM-1 antibody only reacted with the ovarian carcinoma-derived cell lines Colo 316 and 80.13A (Table 1). No reactivity was detected by indirect immunofluorescence between the OM-1 antibody and any of the 15 leukaemia-derived cell lines tested (Table 1).

*Reactivity with solid tumours*

A total of 93 solid tumours were tested by immunoperoxidase staining for reactivity with the OM-1 antibody (Table 2). Of the 26 different tumour types represented, only ovarian tumours reacted with this antibody. The OM-1 antibody gave strong cell surface staining with both primary and metastatic serous cystadenocarcinomas. In primary tumours (Fig. 1A) heavy staining was confined to the cell surface, generally being more intense on the luminal surface, while staining of metastatic carcinoma cells was more general, involving both cell surface and cytoplasm (Fig. 2A, B). All 18 samples of serous cystadenocarcinoma tested were OM-1-positive. In endometrioid carcinomas OM-1 reactivity was apparent at the luminal cell surface as well as within the cytoplasm of tumour cells (Fig. 1B). In 2/3

*Table 1. Reactivity of antibody OM-1 with solid tumour and leukaemia-derived cell lines*

| Cell line | Cell type | Reactivity with: OM-1 | CI-panHu |
|---|---|---|---|
| | *Solid tumour-derived cell lines** | | |
| COLO 316 | ovarian+ | + | + |
| 80.13A | ovarian+ | + | + |
| MCF-7 | breast | - | + |
| HBL100 | breast | - | + |
| MB231 | breast | - | + |
| T47D | breast | - | + |
| A431 | epidermal | - | + |
| COLO 293 | kidney | - | + |
| MM96 | melanoma | - | + |
| COLO 357 | pancreas | - | + |
| COLO 16 | squamous | - | + |
| COLO 526 | uterus | - | + |
| PA-1 | teratocarcinoma | - | + |
| | *Leukaemia-derived cell lines†* | | |
| K562 | myeloid+ | - | + |
| THP-1 | monocytic | - | + |
| U937 | monocytic | - | + |
| RC2a | myelomonocytic | - | + |
| HL60 | promyeloid | - | + |
| NALM-1 | cALL | - | + |
| Reh | cALL | - | + |
| CEM | T-ALL | - | + |
| HSB-2 | T-ALL | - | + |
| RPMI 8402 | T-ALL | - | + |
| JURKATT | T-ALL | - | + |
| WIL-B | B-lymphoblastoid | - | + |
| CESS-B | B-lymphoblastoid | - | + |
| Raji | Burkitt's lymphoma | - | + |
| Namalva | Burkitt's lymphoma | - | + |

*Analysis by solid phase ELISA. Negative <3 times background; positive >3 times background.

†Indirect immunofluoresence analysed by cytofluorograph. Negative <twice background.

*Table 2. Reactivity of antibody OM-1 with tumour tissues**

| Tissue tumour | No. of samples | Reactivity with: OM-1 | CI-panHu |
|---|---|---|---|
| Ovarian | | | |
| Serous cystadenocarcinoma | 14 | + | + |
| Endometrioid adenocarcinoma | 3 | ++† | + |
| Mucinous adenocarcinoma | 4 | -/+‡ | + |
| Clear cell adenocarcinoma | 3 | -/+‡ | + |
| Benign serous cystadenoma | 7 | -/+‡ | + |
| Benign mucinous cystadenoma | 7 | -/+‡ | + |
| Benign Brenner tumour | 2 | - | + |
| Fibrosarcoma | 1 | - | + |
| Cervical | | | |
| Endometrial carcinoma | 1 | - | + |
| Undifferentiated carcinoma | 1 | - | + |
| Endocervical carcinoma | 1 | - | + |
| Vaginal | | | |
| Undifferentiated carcinoma | 1 | - | + |
| Breast | | | |
| Adenocarcinoma | 4 | - | + |
| Skin | | | |
| Melanoma | 4 | - | + |
| Squamous cell carcinoma | 5 | - | + |
| Colon | | | |
| Adenocarcinoma | 2 | - | + |
| Lymph node | | | |
| Lymphoma | 6 | - | + |
| Ovarian adenocarcinoma | 1 | + | + |
| Unknown primary? gynaecological | 1 | + | + |
| Brain | | | |
| Astrocytoma | 1 | - | + |
| Medulloblastoma | 1 | - | + |
| Lung | | | |
| Squamous cell carcinoma | 3 | - | + |
| Stomach | | | |
| Tumour | 1 | - | + |
| Parotid | | | |
| Tumour | 1 | - | + |
| Uterus | | | |
| Melanoma | 1 | - | + |
| Testis | | | |
| Teratoma | 1 | - | + |
| Seminoma | 1 | - | + |
| Pleural/peritoneal effussions | | | |
| Breast adenocarcinoma | 7 | - | + |
| Lung small cell carcinoma | 3 | - | + |
| Ovarian serous adenocarcinoma | 3 | + | + |
| Bone marrow | | | |
| Colon carcinoma | 1 | - | + |
| Prostate carcinoma | 1 | - | + |

*Reactivity detected by indirect immunoperoxidase staining of routinely processed paraffin embedded tissues.

†In two samples strong staining similar to serous adenocarcinomas was observed, but in one tumour only scattered cells stained.

‡In approximately 50% of samples (see text for details) patchy, weak staining similar in degree to that observed in normal ovarian surface epithelium was detected. This was confined to the epithelial component.

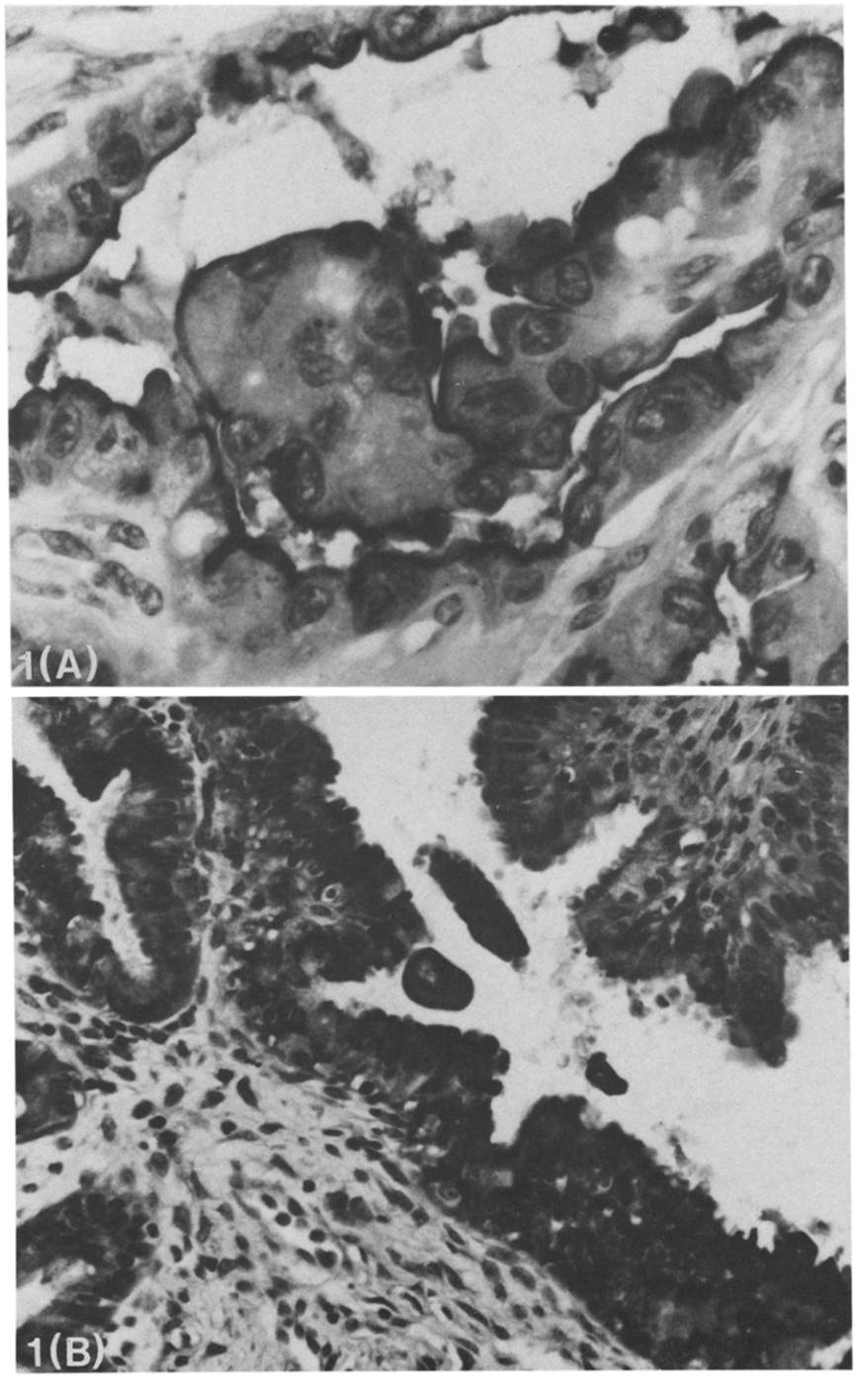


*Fig.* 1 (*A*, *B*).

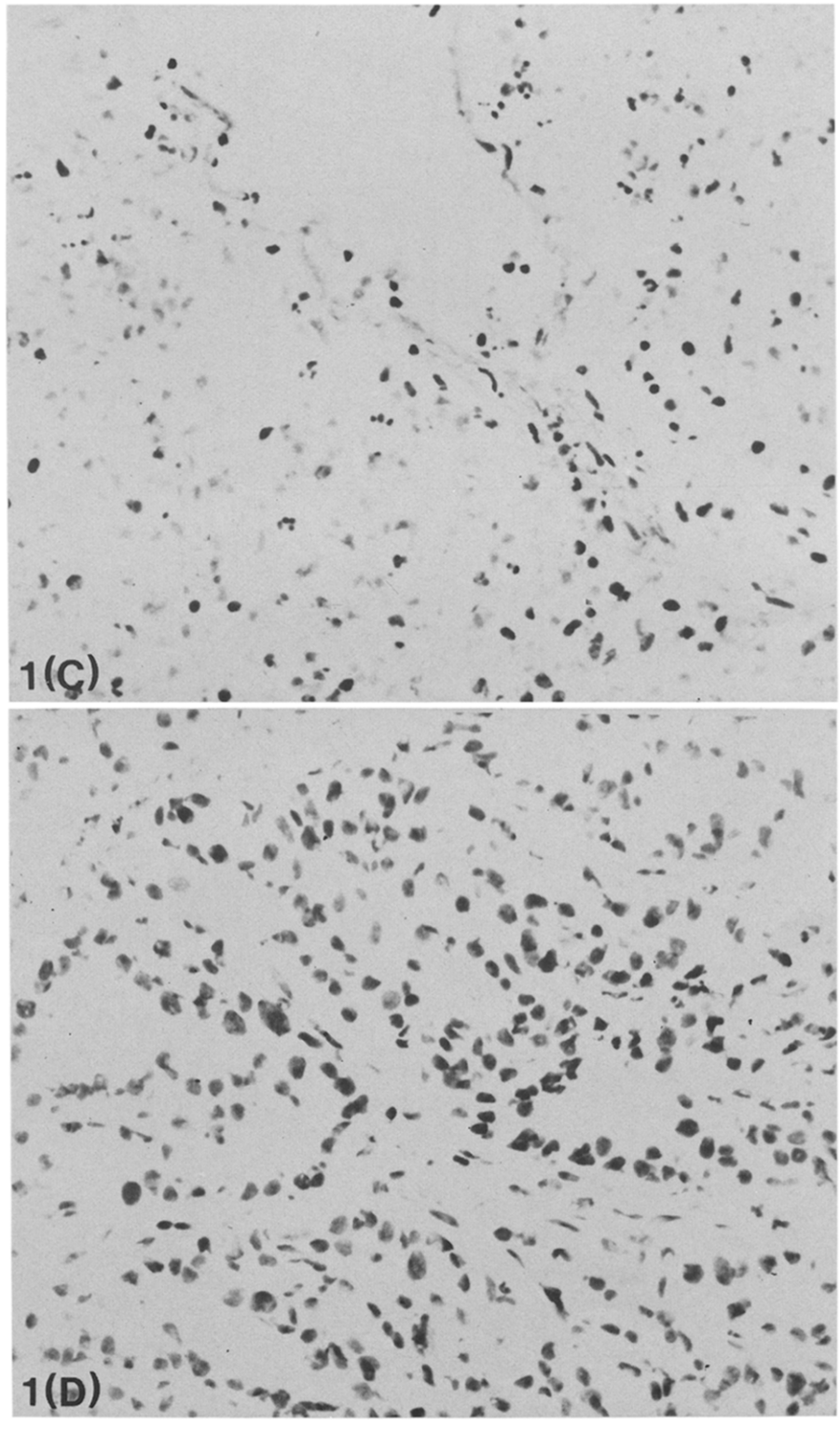


*Fig.* 1 (*C, D*).

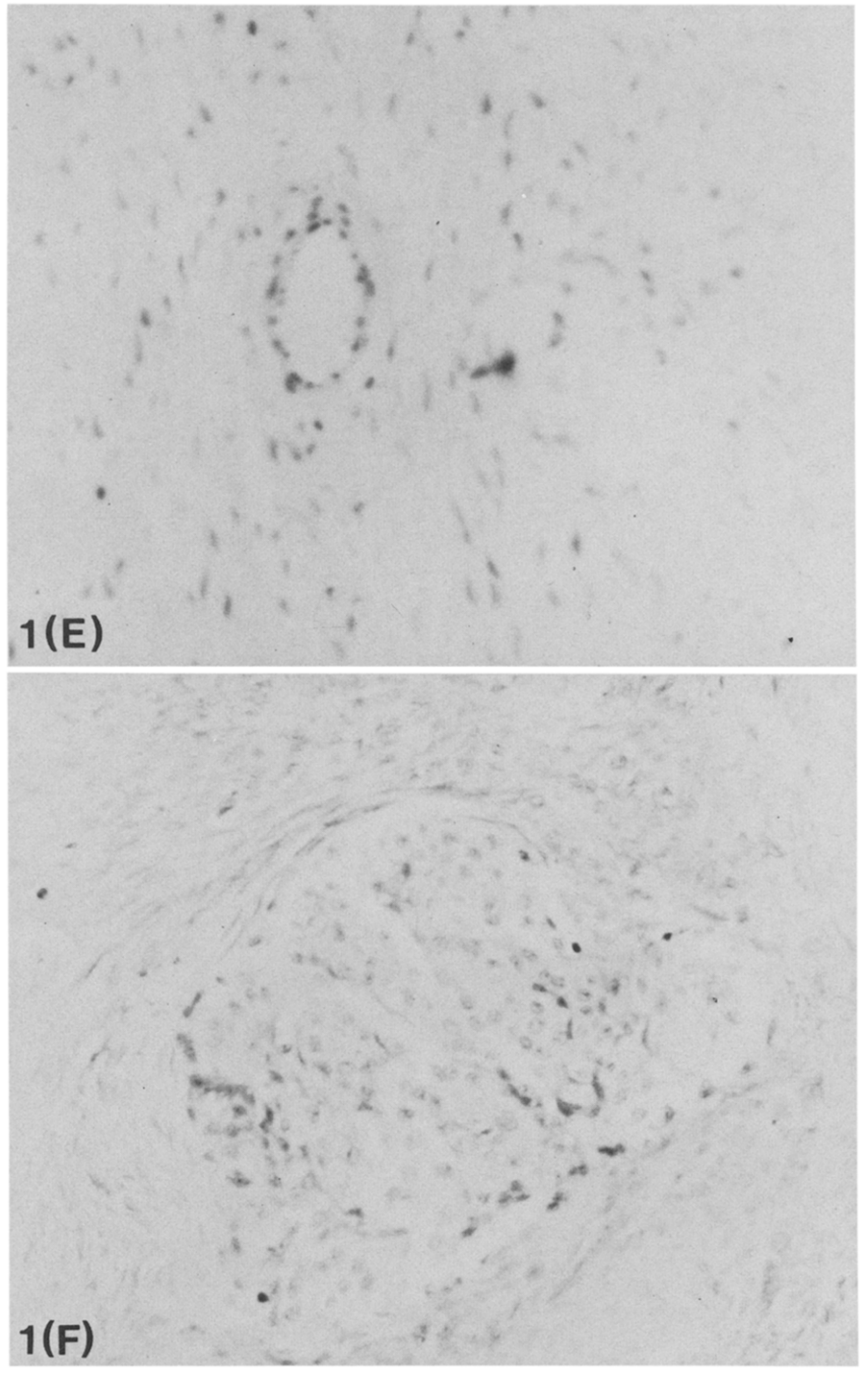


*Fig.* 1 (*E, F*).

*Fig.* 1. *Immunoperoxdase staining of primary ovarian tumours by antibody OM-*1. *Sections from Bouin's-fixed, paraffin-embedded tissues were stained by the immunoperoxidase technique described using antibody OM-*1. *Sections shown are: primary ovarian serous cystadenocarcinoma* (*A*), *ovarian endometrioid adenocarcinoma* (*B*), *ovarian mucinous cystadenocarcinoma* (*C*), *ovarian clear cell adenocarcinoma* (*D*), *ovarian serous cystadenoma* (*E*) *and Brenner's tumour* (*F*). *Gill's haematoxylin* (*A, B*) *and methyl green* (*C-F*) *were used as counterstains. Magnification:* × 460 (*A*), × 230 (*B-F*).

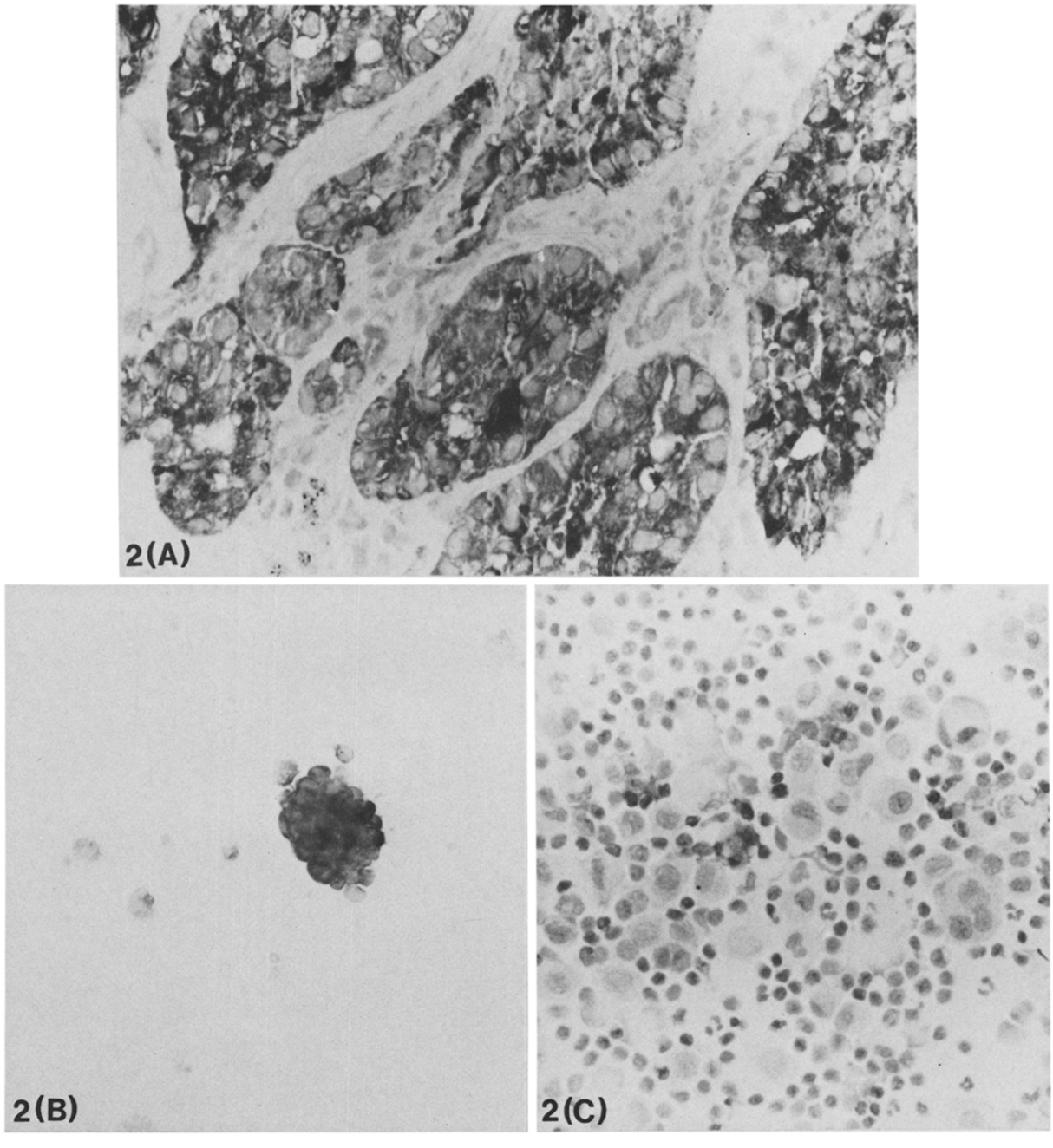


*Fig. 2. Immunoperoxidase staining of metastatic ovarian carcinoma cells by antibody OM-1. Sections from Bouin's-fixed, paraffin-embedded tissues and cytocentrifuge preparations were stained by the immunoperoxidase technique described using the OM-1 antibody. Shown are: section from neck lymph node containing metastatic ovarian adenocarcinoma (A), pleural effusion cells from a patient with ovarian serous cystadenocarcinoma (B) and pleural effusion cells from a patient with breast adenocarcinoma (C). Methyl green was used as counterstain. Magnification:* × 460 (*A*), × 230 (*B, C*).

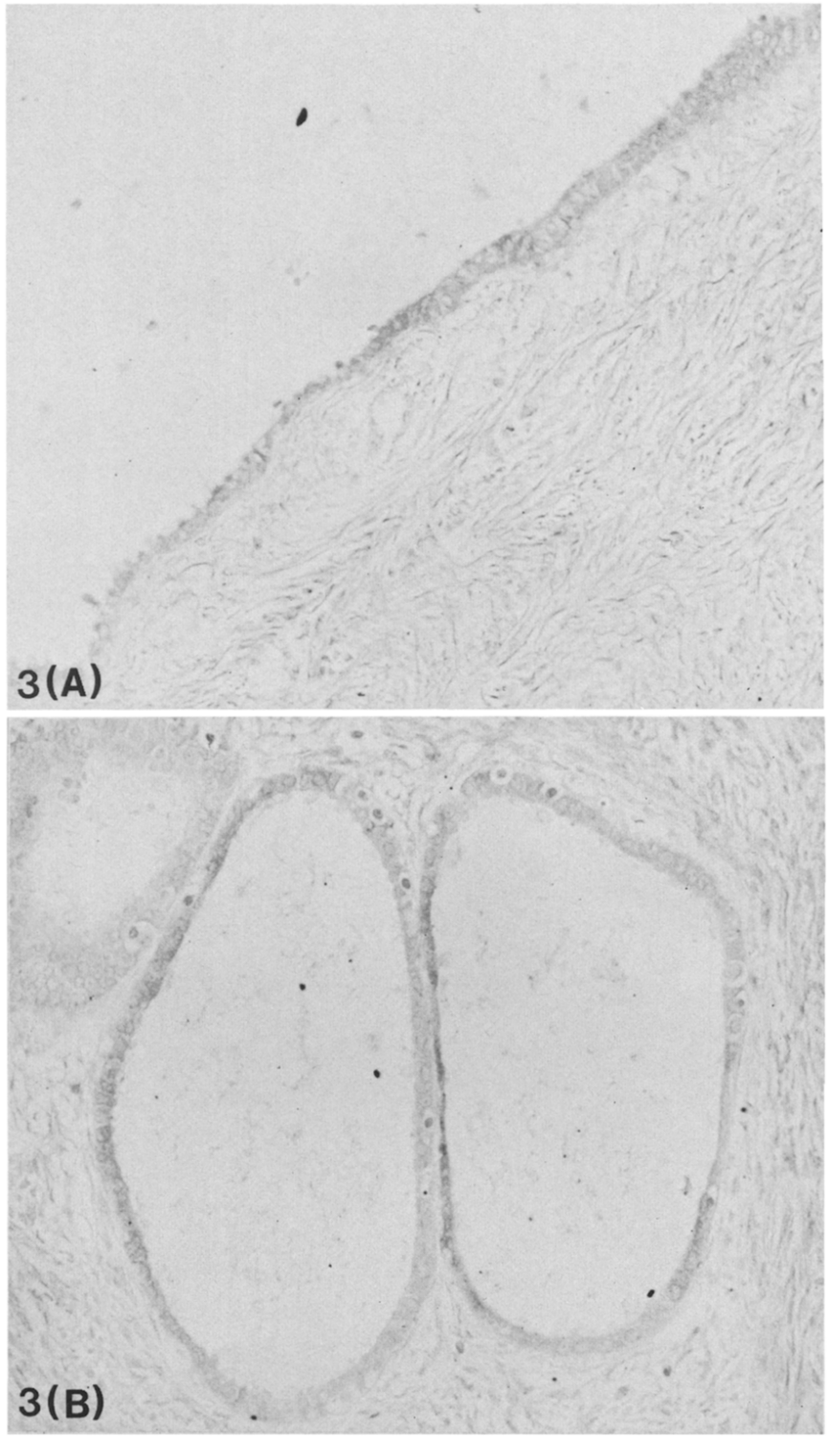


*Fig.* 3 (*A, B*).

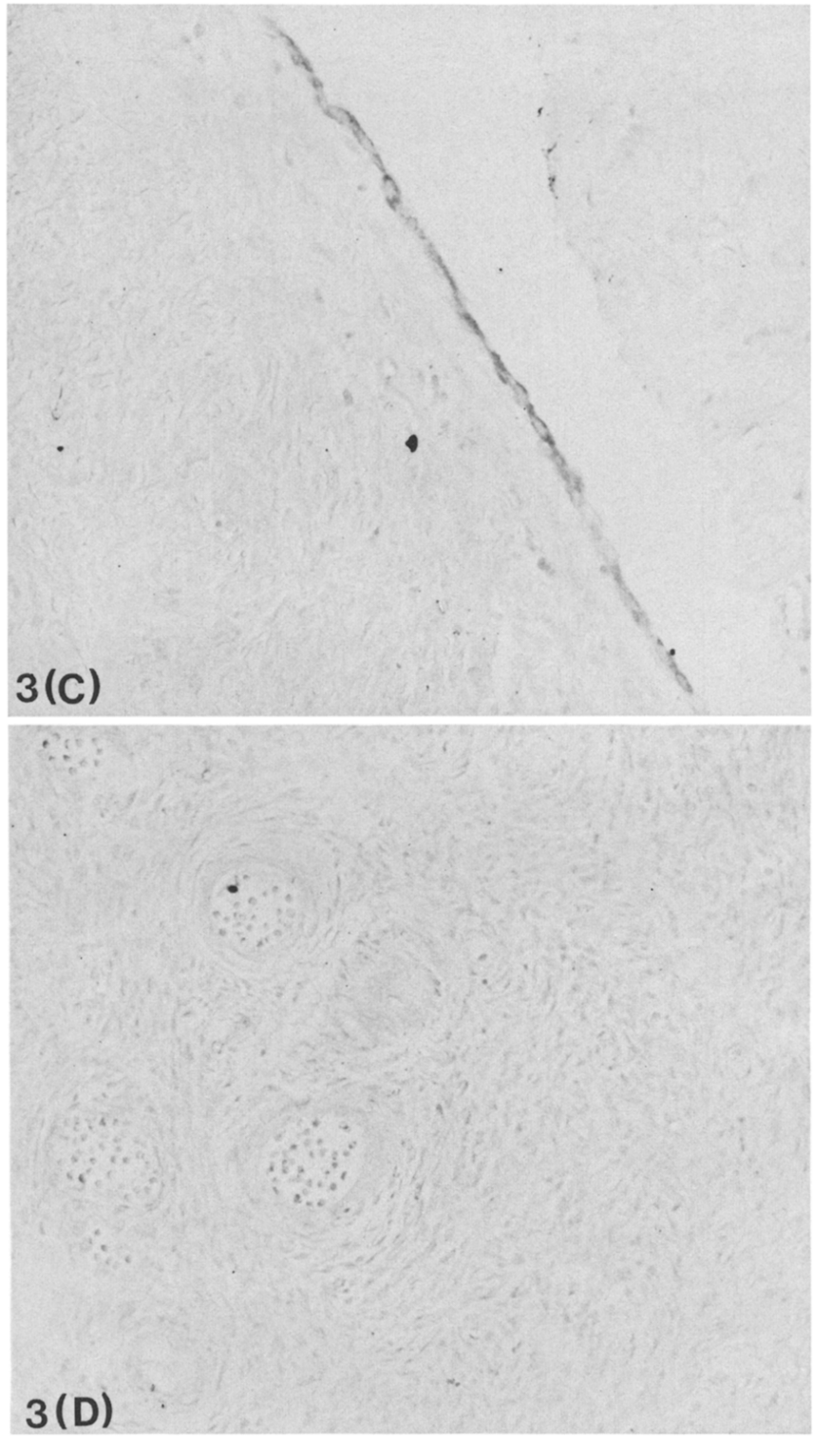


*Fig.* 3 (*C, D*).

*Fig.* 3. *Immunoperoxidase staining of normal ovary by antibody OM-*1. *Sections from Bouin's-fixed, paraffin-embedded tissues were stained by the immunoperoxidase technique described using the antibody OM-*1. *Areas shown are: surface epithelium* (*A*), *inclusion cysts* (*B*), *intermediate epithelium* (*C*) *and follicles and stroma* (*D*). *Methyl green was used as counterstain. Magnification:* × 230.

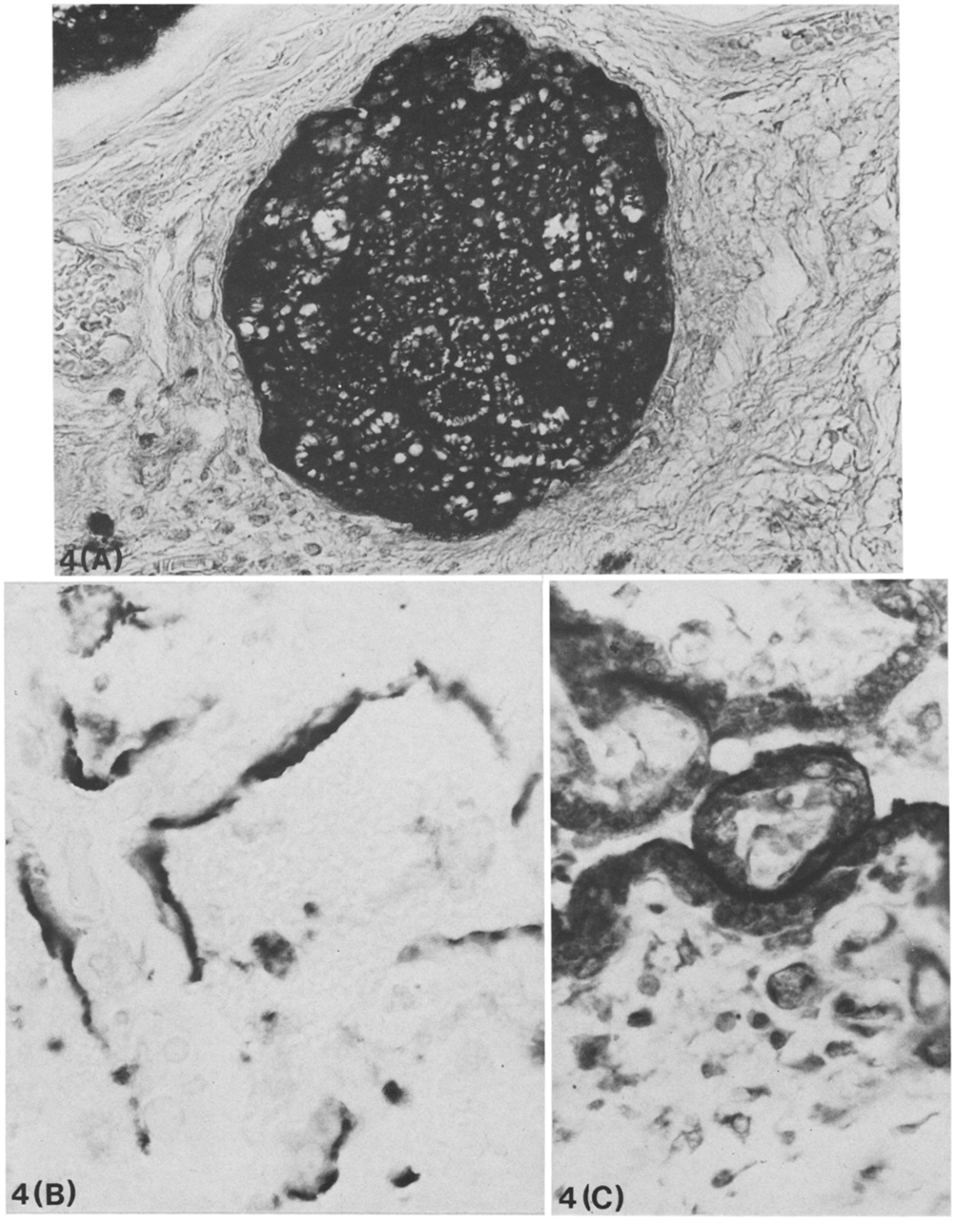


*Fig.* 4 (*A–C*).

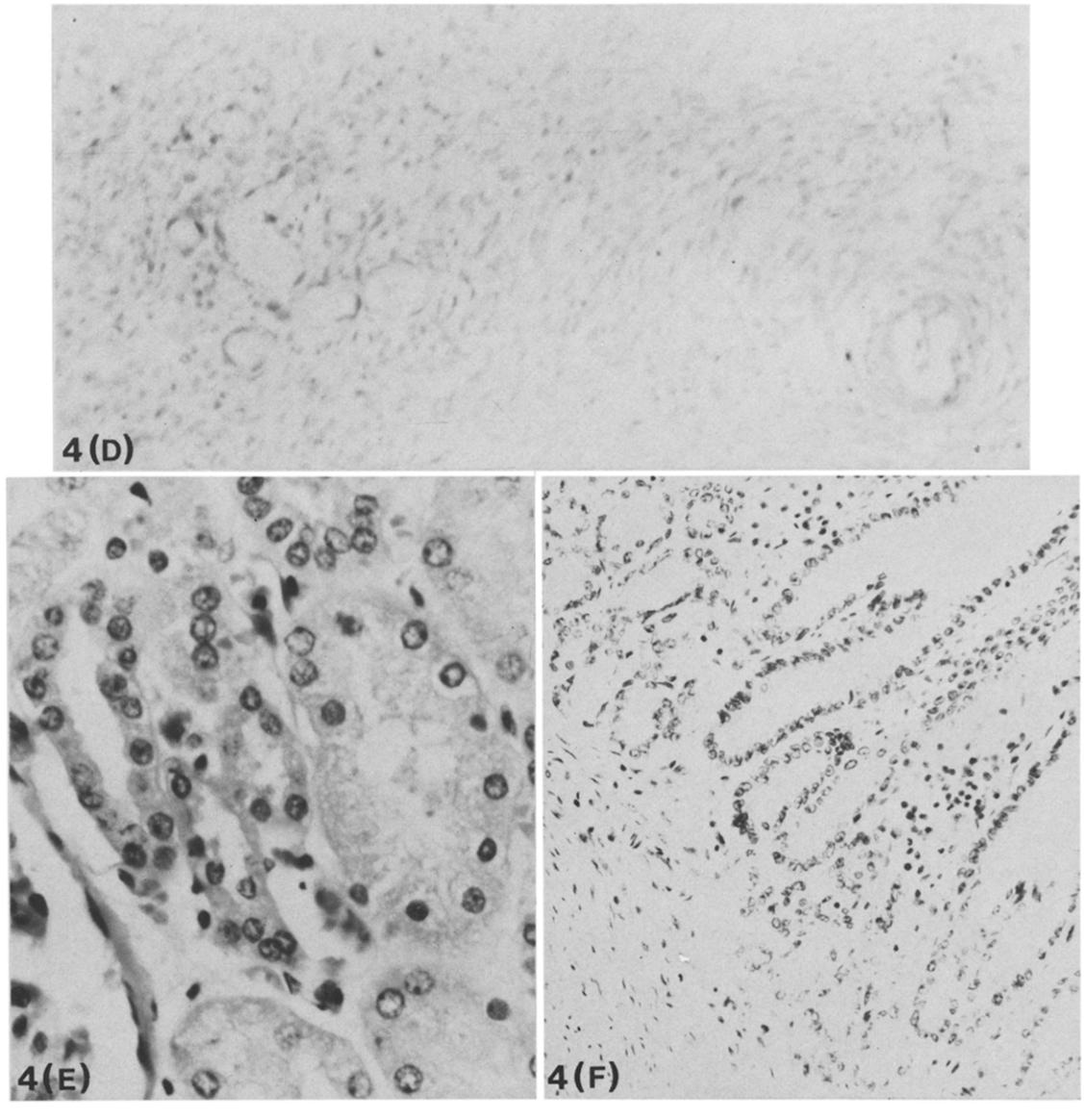


*Fig.* 4 (*D–F*).

*Fig.* 4. *Immunoperoxidase staining of normal tissues by antibody OM-1. Sections from Bouin's-fixed, paraffin-embedded tissues were stained by the immunoperoxidase technique described using antibody OM-1. Sections shown are: skin including sebaceous glands (A), lung (B), term placenta (C), kidney (D) and colon (E). Methyl green (A, B) and Gill's haematoxylin (C–E) were used as counterstains. Magnification:* ×230 (*A, B, E*), ×460 (*C, D*).

samples tested the pattern of staining was uniform throughout the section (i.e. all proliferating epithelium). However, in the third case tested only scattered cells within the proliferating epithelium were reactive, although staining was still strong. The only reactivity detected between the OM-1 antibody and ovarian mucinous cystadenocarcinomas, clear cell carcinomas, benign serous cystadenomas and benign mucinous tumours was an extremely weak and patchy staining involving less than 20% of the epithelial lining in 10/24 samples tested. In 2/4 malignant and 4/7 benign mucinous tumours this reactivity was confined to the basal cytoplasm of the involved epithelium (Fig. 1C). In 1/3 clear cell adenocarcinomas weak staining over the entire tumour cell surface was detected (Fig. 1D). In 3/7 benign serous cystadenomas weak staining of the epithelial cell surface and cytoplasm was observed (Fig. 1E). In all instances where weak reactivity was detected the intensity of staining was comparable to that seen in normal ovarian surface epithelium (see below). The only other ovarian tumours thus far tested were two benign Brenner tumours (Fig. 1F) and a fibrosarcoma, all of which were totally non-reactive with the OM-1 antibody. The OM-1 antibody gave no detectable reaction with breast or colon adenocarcinomas (Table 2, Fig. 2B). In light of the reactivity with normal cell types detailed below, it was of interest that squamous cell carcinomas of neither skin nor lung reacted with the OM-1 antibody.

*Reactivity with normal adult and foetal tissues*

In the 85 normal tissue samples tested by immunoperoxidase staining for reactivity with the OM-1 antibody the only normal cell types stained were sebaceous gland cells, lung type II pneumocytes, placental syncytial trophoblasts and ovarian surface epithelial cells (Table 3). A total of 24 adult and seven foetal tissues were represented in the panel.

In normal ovary extremely weak cell surface staining of the ovarian surface epithelium was detected (Table 3, Fig. 3A). Even weaker cytoplasmic staining was occassionally discernable in these cells. Inclusion cysts formed by involution of the surface epithelium also reacted extremely weakly with the OM-1 antibody (Fig. 3B). Interestingly, intermediate (?regenerating) surface epithelium was better stained by the OM-1 antibody, but the reaction was primarily cytoplasmic (Fig. 3C). None of the other cell types which comprise the ovarian stroma and follicles reacted with the OM-1 antibody (Fig. 3D). A single example of rete ovarii examined was non-reactive.

In sebaceous glands extremely strong reaction was observed over the entire surface of all cells contained within the glands while cells of the surrounding dermis were not stained (Fig. 4A). OM-1 reactivity on lung pneumocytes was principally cell surface (Fig. 4B), while on placental syncytial trophoblasts the staining was both cytoplasmic and cell surface (Fig. 4C). No reactivity was discernable with secretions of either sebaceous gland cells or pneumocytes. No reactive cells were found in normal kidney or colon, which are tissues where cross-reactivity with monoclonal antibodies is often found (Fig. 4D, E).

*Reactivity with haematopoietic cells*

The reactivity of the OM-1 antibody with cell populations in normal peripheral blood and bone marrow was assessed by indirect immunofluorescence and by immunoperoxidase staining (Table 4). No positive reaction with any cell population present could be detected by either technique.

## DISCUSSION

From the results above, it is clear that the molecule detected by the OM-1 antibody is a normal component of the surface membrane of sebaceous gland cells. Further, of all normal cell types thus far studied, sebaceous gland cells exhibit the highest concentration of this molecule. Therefore this molecule will henceforward be termed sebaceous gland antigen (SGA). SGA has also been detected on the cell surfaces of lung type II pneumocytes and placental syncytial trophoblasts. One function shared by these three cell types is the synthesis and secretion of protein-containing fluid. Although SGA has not been detected in any other secretory cell type represented in the tissue sections thus far analysed (see Table 3), the possibility that SGA may be expressed by other highly specialized secretory cells, such as the cells of Brunner's glands (duodenum), is being investigated.

Low levels of SGA have also been detected in normal ovarian surface epithelium. This is of particular interest as it is thought that epithelial tumours of the ovary, some of which we have found to be SGA-positive, arise through transformation of cells of the surface epithelium. Weak reactivity with the OM-1 antibody is retained by the epithelial cells lining inclusion cysts, which are derived from the normal surface epithelium. Of note was the finding that intermediate (?regenerating) surface epithelium, frequently present in tumour specimens, contained distinct amounts of intracellular SGA which were more readily detectable than the low levels present in the normal surface epithelium. This finding suggests that synthesis of SGA by ovarian

*Table 3. Reactivity of antibody OM-1 with normal adult and foetal tissues*

| Tissue | No. of samples | Reactivity with: OM-1 | CI-panHu |
|---|---|---|---|
| Sebaceous glands | 10 | +* | + |
| Skin | 10 | - | + |
| Vagina | 8 | - | + |
| Cervix | 7 | - | + |
| Ovary | | | |
| Surface epithelium | 5 | +† | + |
| Follicles, stroma | 8 | - | + |
| Rete ovarii | 1 | - | + |
| Breast | 1 | - | + |
| Testis | 2 | - | + |
| Prostate | 2 | - | + |
| Bladder | 1 | - | + |
| Oesophagus | 3 | - | + |
| Stomach | 2 | - | + |
| Small intestine | 2 | - | + |
| Colon | 4 | - | + |
| Gall bladder | 3 | - | + |
| Pancreas | 2 | - | + |
| Thymus | 1 | - | + |
| Spleen | 2 | - | + |
| Lymph node | 3 | - | + |
| Tonsil | 2 | - | + |
| Liver | 1 | - | + |
| Lung | 3 | +‡ | + |
| Heart | 1 | - | + |
| Kidney | 5 | - | + |
| Adrenal | 1 | - | + |
| Thyroid | 1 | - | + |
| Smooth muscle | 3 | - | + |
| Placenta | 3 | +§ | + |
| Foetal | | | |
| Skin (no sebaceous glands) | 1 | - | + |
| Thymus | 1 | - | + |
| Kidney | 1 | - | + |
| Small intestine | 1 | - | + |
| Large intestine | 1 | - | + |
| Blood vessels | 1 | - | + |
| Connective tissues | 1 | - | + |

Reactivity detected by indirect immunoperoxidase staining of routinely processed paraffin embedded tissues.
*All sebaceous gland cells staining.
†Very weak and patchy staining confined to surface epithelial cells.
‡Staining confined to type II pneumocytes.
§Staining confined to syncytial trophoblasts.

epithelium is allied to some form of proliferative activity. Consistent with this idea is the finding of higher levels of SGA in metastatic serous carcinomas compared to primary tumours of the same type.

The presence of SGA in normal ovarian epithelium raises the question of its presence in other Mullerian epithelia, namely the epithelial linings of the Fallopian tube, uterus and endocervix. Detailed studies of SGA expression in these tissues and in tumours deriving from these epithelia are currently underway. There is as yet no evidence with regard to the expression of SGA in foetal ovary, hence the possibility exists that SGA may be an ovarian oncofoetal antigen.

Regardless of the reason for the expression of SGA by certain epithelial ovarian neoplasms, its presence on the surface of these malignant cells may prove to be a valuable marker for the detection and diagnosis of ovarian cancer. In this respect the fact that benign ovarian tumours contain no more SGA than found in normal ovary is particularly important. Amongst ovarian malignancies the presence of SGA at levels higher than found in normal ovary is apparently restricted to ovarian epithelial tumours of the serous and endometrioid types. In all mucinous and clear cell carcinomas tested SGA was either not detectable or was present at the same low level as found in normal ovary. It would therefore

*Table 4. Reactivity of antibody OM-1 with normal haematopoietic cells*

| Population | Reactivity with: OM-1 | CI-panHu |
|---|---|---|
| Indirect immunofluoresecence* | | |
| Normal peripheral blood | | |
| Lymphocytes | - | + |
| Monocytes | - | + |
| Granulocytes | - | + |
| Platelets | - | + |
| Indirect immunoperoxidase staining† | | |
| Normal peripheral blood | | |
| Lymphocytes | - | + |
| Monocytes | - | + |
| Granulocytes | - | + |
| Platelets | - | + |
| Normal bone marrow | | |
| Lymphocytes | - | + |
| Metamyelocytes | - | + |
| Myelocytes | - | + |
| Platelets | - | + |
| Megakaryocytes | - | + |
| Megakaryoblasts | - | + |
| Granulocytes | - | + |

*Indirect immunofluoresecence analysed cytofluorographically. Negative <twice background.

†Populations verified by staining with other monoclonal antibody reagents or by examination by a haematologist.

appear that high levels of SGA in ovarian epithelial cells may arise as a function or a consequence of proliferative activity coincident with transformation of these cells leading to the development of serous or endometrioid carcinomas.

On the basis of its distribution in normal and tumour tissues, SGA is unlike any other antigen or marker previously described. In particular, SGA is distinct from CEA as SGA has not been detected in some CEA-positive mucinous cystadenocarcinomas [10]. The distribution of SGA amongst ovarian tumours is similar to that of the CA125 antigen [10] and is the converse of the pattern of reactivity seen with the monoclonal antibody described by Bhattacharya and coworkers [8], lending further weight to the hypothesis that ovarian mucinous cystadenocarcinomas are a group distinct from the other types of ovarian carcinoma [8]. However, SGA is distinct from CA125, which is reportedly absent from normal skin, presumably including sebaceous glands [9]. Interestingly, the extremely strong luminal cell surface pattern of staining observed with the OM-1 (anti-SGA) antibody in primary serous cystadenocarcinomas (see Fig. 1A) is markedly similar to the luminal cell surface staining seen with the OC125 (anti-CA125) antibody [9]. SGA is also distinct from two other ovarian tumour-associated antigens — OCA, which is found in all ovarian tumour types [23], and OCAA, which is reportedly expressed in both serous and mucinous cystadenocarcinomas [6].

The expression of SGA in normal tissues is remarkably similar although not coincident with that of placental alkaline phosphatase. The pattern of staining seen in placenta with the anti-placental alkaline phosphatase monoclonal antibody NDOG2 [24] is essentially the same as that seen with the OM-1 (anti-SGA) antibody. However, staining patterns of the two antibodies are dissimilar in cervix, lung and thymus, indicating that SGA and placental alkaline phosphatase are distinct molecules. Nevertheless, the preliminary data available on the distribution of these two molecules in ovarian epithelial malignancies is strikingly similar [24] in that both are strongly expressed by the epithelium of malignant serous tumours and are either absent or at low levels in mucinous and clear cell carcinomas. The significance of the high levels of expression of these two placenta-associated molecules in serous carcinomas is unclear.

Comparison of the distribution of SGA in normal ovary with that reported for other markers is difficult as the ovarian surface epithelium, which we have found to contain very low lvels of SGA, is particularly fragile and infrequently retained through routine processing procedures. Hence a reported negative result for reaction with

normal ovary may not exclude weak reactivity with the ovarian surface epithelium, which comprises less than 1% of the cell area present in a section of normal ovary. In the cases of the other markers discussed above, the presence of low levels in the ovarian surface epithelium has not been specifically excluded.

Among serous cystadenocarcinomas, high levels of SGA were detected in all 14 primary tumours tested, while SGA was either not detectable or at very low levels in all 16 benign ovarian tumour specimens. It has been reported that the CA125 antigen is not detectable in all serous cystadenocarcinomas, and that it is also present in some benign ovarian tumours [10]. Whether SGA expression is a consistent or variable feature of these ovarian tumours, and whether SGA is detectable in ovarian tumours other than serous cystadenocarcinomas and endometrioid carcinomas, are questions currently under investigation.

More extensive screening of OM-1 (anti-SGA) antibody reactivity in both ovarian tumours and other malignancies is necessary to comprehensively define the spectrum of SGA expression. Nevertheless, the results reported here indicate that SGA is likely to prove valuable in the differential diagnosis of ovarian malignancies. The finding that SGA retains its immunogenicity, as detected by the OM-1 antibody, through routine fixation and paraffin-embedding procedures is of particular importance as this obviates the necessity of having access to fresh tissue, and allows assessment of SGA expression in archival pathology specimens. On the evidence presented it is possible to discriminate on the basis of SGA expression between metastatic ovarian tumour cells and activated mesothelial cells found in peritoneal effusions. This should facilitate identification of metastatic spread and consequent staging of the disease.

It has not yet been established whether SGA is found at elevated levels in the serum of ovarian cancer patients. However, recent evidence on the use of serum levels of OCA and CA125 in the monitoring of tumour burden in ovarian cancer [23, 25, 26] indicates that ovarian tumour-associated antigens show definite potential as serum markers for monitoring and/or early detection of ovarian epithelial tumours.

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