

Kiel classification is referred to as the small cleaved cell in the Lukes and Collins classification. Nuclear cleavage is a feature of centrocytes in some preparations but it is a rather more prominent feature of many T-cell lymphomas. The same criticisms could be applied to the use of the non-specific term 'large non-cleaved cell' for the centroblast.

It is apparent that in the Kiel classification many diagnoses imply an immunological, as well as a morphological, diagnosis and it might be supposed that a pathologist without access to immunological techniques could not use this classification. This is untrue for some tumours in which morphology correlates well with the immunological phenotype.⁹ Thus, a follicular growth pattern is probably the best B-cell marker available for malignant lymphomas. However, with diffuse large cell lymphomas, prediction of immunological phenotype on morphology alone would be less accurate. It is my view that immunological

techniques should be used for the diagnosis of malignant lymphomas and that diagnosis without access to these techniques will necessarily be limited.

THE LYMPH NODE BIOPSY

It is apparent from the foregoing that the classification of lymphomas is dependent on the accurate diagnosis of these tumours. This, in turn, depends upon the material and techniques available to the pathologist. The very least he should demand is a well taken, adequately fixed biopsy. Fragmented, traumatised lymph node biopsies are not satisfactory nor yet are large biopsies placed unsliced in inadequate amounts of fixative. The purpose of the lymph node biopsy is to obtain a diagnosis and if the quality of the biopsy precludes this, or worse still leads to an erroneous diagnosis, the exercise has failed. In this exercise the pathologist is paramount and should have no reservations in demanding that the surgeon, who is acting as a technician, should produce the best possible biopsy or re-biopsy if necessary.

Ideally the biopsy should be received fresh in the laboratory and carefully sliced. One slice can be used to make imprint cytological preparations which can be stained with Romanowski dyes as well as being used for cytochemistry. Immunological studies can be performed on dispersed cells and tissue may be frozen for immunohistochemistry.

TABLE 2

MODIFIED KIEL CLASSIFICATION (LENNERT, 1978)	
I	Low grade malignancy
	ML Lymphocytic
	B-CLL
	T-CLL
	Hairy cell leukaemia
	Mycosis fungoides and Sezary's syndrome
	T-zone lymphoma
	ML Lymphoplasmacytic/lymphoplasmacytoid (LP immunocytoma)
	ML Plasmacytic
	ML Centrocytic
	ML Centroblastic/centrocytic
	Follicular
	Follicular and diffuse
	Diffuse
	With or without sclerosis
II	High grade malignancy
	ML Centroblastic
	Primary
	Secondary
	ML Lymphoblastic
	B-lymphoblastic, Burkitt type and others
	T-lymphoblastic, convoluted cell type and others
	Unclassified
	ML Immunoblastic
	With plasmablastic/plasmacytic differentiation (B)
	Without plasmablastic/plasmacytic differentiation (B or T)

TABLE 3

LUKES AND COLLINS FUNCTIONAL CLASSIFICATION OF MALIGNANT LYMPHOMAS (LUKES AND COLLINS, 1977)

U-cell (Undefined)

T-cell

- Small lymphocyte
- Convoluted lymphocyte
- Immunoblastic sarcoma
- Lennert's lymphoma

B-cell

- Small lymphocyte
- Plasmacytoid lymphocyte
- Follicle centre cell lymphoma
 - Follicular or diffuse with or without sclerosis
 - Small cleaved
 - Large non-cleaved
- Immunoblastic sarcoma
- Hairy cell leukaemia

Histiocytic

Well-fixed tissue will provide good histology and immunohistochemistry of stable antigens, such as cytoplasmic immunoglobulin and cytokeratins. The increased availability of high quality **antisera** should make **immunohistochemistry** support available to all pathologists, either through their own laboratories or specialised central laboratories. The use of a relatively simple panel of markers on **fixed** tissue can take the guesswork out of the diagnosis of anaplastic tumours which may have great importance in the management of the patient.'

Both B-cells and T-cells undergo gene rearrangement early in their development. **Complimentary** DNA probes have been developed for the identification of these rearrangements and provide a sophisticated means for determining the lineage and clonality of lymphoid **proliferations**.^{11,12} Unfixed frozen tissue is required for the extraction of DNA providing a further argument in favour of receiving all lymph node biopsies in the laboratory fresh. Tissue may be **fixed** for electron microscopy

although the value of this technique in the diagnosis of malignant lymphoma is limited.

Some lymphomas show a relatively **mono-**morphous proliferation of a single clone of lymphoma cells. Lymphocytic and **lymphoblastic** lymphomas fall into this category. Others, however, show complex mixtures of cells in which the neoplastic clone may constitute only a minority population. Thus, many **follicular** lymphomas contain large numbers of helper and suppressor T-cells, polyclonal reactive **B-cells** and dendritic reticulum cells (Figs 1 and 2). These non-neoplastic elements are presumably attracted into the malignant lymphoma as a result of physiological influences exerted by the tumour cells. This cellular heterogeneity should be borne in mind when interpreting immunological or **immunohisto-**chemical data. Lymphokine production by lymphoma cells may be responsible for the prevalence of **eosinophils**, macrophages and high endothelial **venules** in many T-cell tumours. These non-neoplastic elements provide a valuable histopathological aid to the diagnosis of T-zone lymphoma.

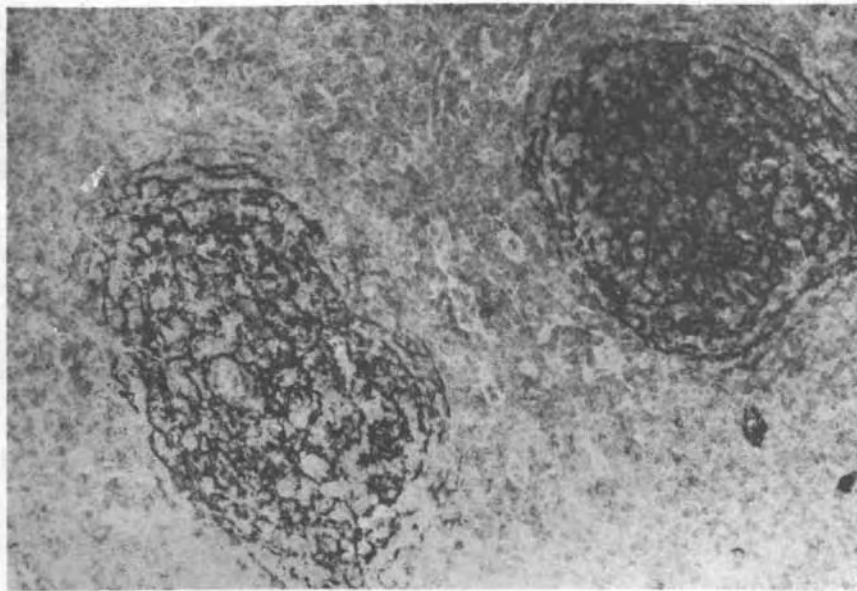


FIG. 1 : Frozen section of follicle centre cell lymphoma stained with an antibody to C3b receptors. **Dendritic** reticulum cells within the neoplastic follicles are clearly defined. PAP X 120.

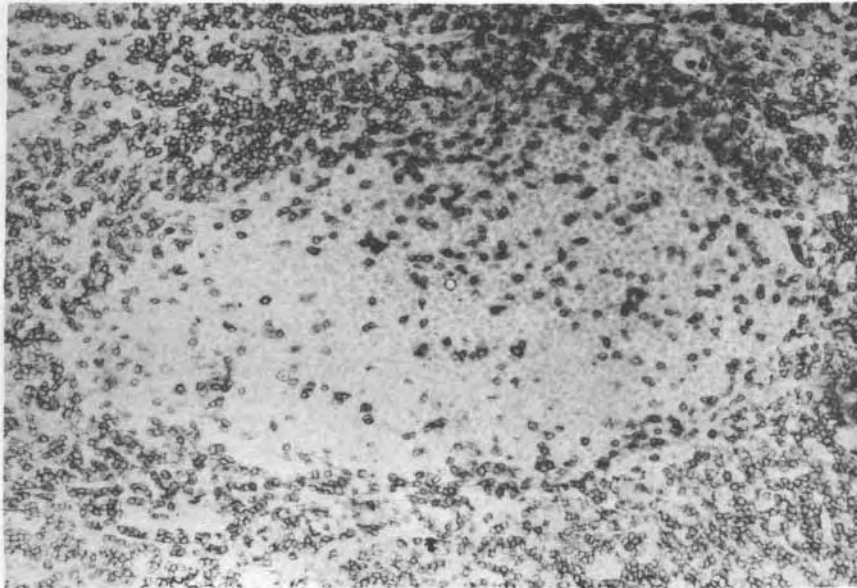


FIG. 2 : Frozen section of a follicle centre cell lymphoma stained with a monoclonal antibody to T-cells. Note the large number of reactive **T-cells** around, and within, the neoplastic follicle. PAP X 120.

Although it has been recognised for many years that extranodal lymphomas often have a better prognosis than nodal lymphomas, this fact does not appear to have been taken into account in any of the classifications proposed. It is our belief that many extranodal lymphomas arise from mucosa-associated lymphoid tissue and that they follow a physiological circulation pathway that causes them to remain localised for long periods of time.^{1 3} Thus, most cases of follicle centre cell lymphoma, centroblastic/centrocytic follicular arising in a peripheral lymph node are stage **III** or stage **IV** disease at presentation, whereas centroblastic/centrocytic lymphomas of the intestine are frequently localised to the bowel and regional lymph nodes.

B-CELL LYMPHOMAS

Lymphocytic/Plasmacytic Lymphomas

1. Malignant Lymphoma Lymphocytic

This tumour may be regarded as the tissue phase of chronic lymphocytic leukaemia although Pangalis *et al*¹⁴ reported patients with lymphocytic lymphoma followed up

for many years who did not develop leukaemia. Approximately **10 - 15%** of patients with chronic lymphocytic leukaemia will develop gross lymphadenopathy or tumour formation. Biopsies of lymph nodes from patients with chronic lymphocytic leukaemia, with minor degrees of lymphadenopathy, show a diffuse infiltrate with small lymphocytes and reticulin stains will usually show preservation of the underlying lymph node architecture. Lymph nodes from patients with gross lymphadenopathy (tumour forming group) often have an ill-defined, nodular appearance due to the presence of collections of lymphoblasts and pro-lymphocytes that stain less darkly than the small lymphocytes (Fig 3). These collections of proliferating blast cells are referred to as proliferation centres. Lymphocytic lymphomas occasionally undergo transformation to high-grade lymphomas (immunoblastic lymphoma) composed of blast cells, often showing marked pleomorphism. This transformation is sometimes referred to as Richter's syndrome¹⁵ and carries a very poor prognosis. Blast cells can be shown to express the same immunoglobulin class and light chain restriction as the original lymphocytic lymphoma and are presumably derived from the same clone.^{16,}

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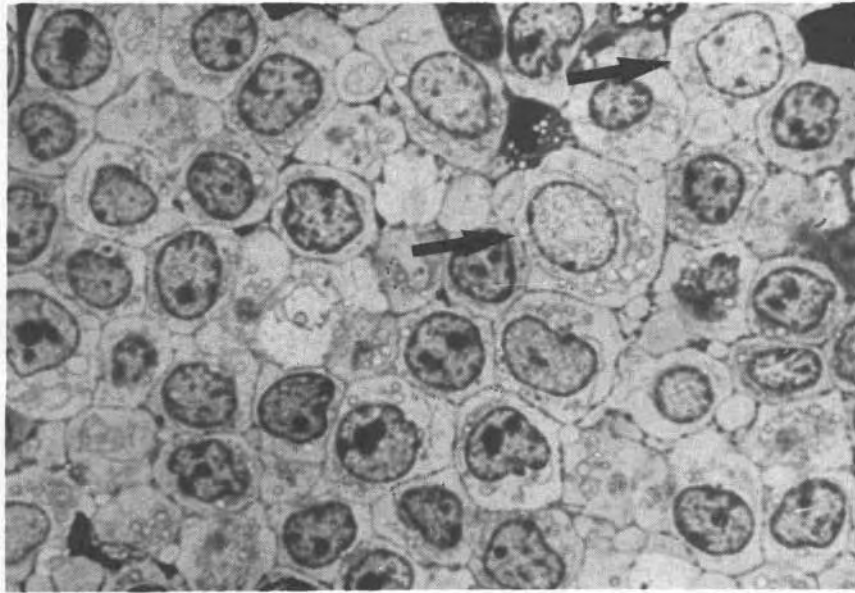


FIG. 3 : Plastic embedded section of malignant lymphoma, lymphocytic. Note the regular, rounded nuclei and the occasional lymphoblasts and pro-lymphocytes (arrows). Toluidine blue X 1200.

2. Malignant Lymphoma Pro-Lymphocytic

This is essentially the tissue phase of **pro-lymphocytic leukaemia** and is rarely seen by pathologists since although it causes **splenomegaly** it does not usually result in significant peripheral lymphadenopathy. The tissues are infiltrated by pro-lymphocytes with prominent central nucleoli. As with other leukaemic infiltrates, there is often a substantial preservation of the underlying tissue architecture. This may give the tumour an overall nodular appearance. However, the cytology of the cells and their monomorphism clearly distinguishes this tumour from follicle centre cell lymphoma.

3. Malignant Lymphoma Lymphoplasmacytic/Lymphoplasmacytoid

Lennert¹⁸ divides lymphoplasmacytic lymphoma into 3 histological types: a) **Lymphoplasmacytic**, composed of lymphocytes and typical plasma cells; b) **Lymphoplasmacytoid**, composed of small lymphocytes and cells intermediate between lymphocytes and plasma cells; c) **Polymorphic**, composed of a mixture of lymphocytes, plasma cells and follicle centre cells. I regard the latter group as representing follicle centre lymphomas with plasmacytic differentiation and prefer to categorise them with the follicle centre cell lymphomas. In the Kiel study, 3 clinical groupings of lymphoplasmacytic lymphoma were recognised:

a) lymph node type; b) splenomegalic type; c) ocular-cutaneous type. Approximately one-third of the cases have overt leukaemia. About one-quarter of the patients exhibit a **paraproteinaemia**, usually IgM, and are categorised as Waldenstrom's macroglobulinaemia.

Lymphoplasmacytic lymphomas are often associated with dilatation of the sinuses of lymph nodes which are **filled** with proteinaceous material. Amyloid occurs in a small proportion of cases, although frequently hyaline material resembling amyloid in H&E preparations, but not reacting as amyloid with special stains, is seen. Lymphoplasmacytoid lymphomas may, at **first** sight, appear very similar to lymphocytic lymphomas. The **Giemsa** stain usually shows more marked basophilia of a proportion of the tumour cells. The most characteristic feature of these tumours is the presence of PAS-positive **immunoglobulin** inclusions in the tumour cells. These inclusions (**Ducher-Fahey** bodies) indent the nucleus and frequently appear to be intranuclear in position.

FOLLICLE CENTRE CELL LYMPHOMAS

Malignant Lymphoma **Centrocytic**

Workers in Kiel and elsewhere have clearly established that malignant lymphoma **centrocytic** is a distinct **lymphoma**.^{19, 20, 21} The tumour is frequently widely disseminated at presentation with bone marrow and peri-

peripheral blood involvement. The cells in the peripheral blood characteristically have a cleaved nucleus. In tissue sections they form a monomorphic infiltration that is more destructive in its growth pattern than CLL. The tumour cells vary in appearance from almost rounded to more elongated cleaved cells depending, to some extent, on fixation (Fig 4). They may, therefore, be confused with lymphocytic lymphomas. However, the chromatin of the cells is less coarsely clumped and lymphoblasts and pro-lymphocytes are not seen in this tumour.

Malignant lymphoma centrocytic appears to be the same tumour that has been described in America as intermediate cell lymphoma or as mantle cell lymphoma.^{22, 23} With immunohistochemical techniques, in the early stages of the disease, it can be shown that the tumour cells surround and eventually overrun reactive lymphoid follicles. They do not expand from the follicles as do other follicle centre cell lymphomas. Immunological studies have shown that the cells of malignant lymphoma centrocytic have a different phenotype from the centrocytes found in follicle centre cell lymphomas. It is unfortunate, therefore, that they have been given the name centrocyte.

The evidence suggests that they are derived from an extra-follicular lymphoid cell. It would be less confusing if the term 'intermediate cell lymphoma' was adopted for this lymphoma to distinguish it from the follicle centre cell lymphomas that show centrocyte predominance and which have a much better prognosis. The term 'intermediate lymphoma' was coined because the tumour cells have a morphology intermediate between small lymphocytes and centrocytes.

Malignant Lymphoma – **Centroblastic/** Centrocytic

These follicle centre cell derived tumours are composed of varying mixtures of centroblasts and centrocytes (Fig 5). They may have a follicular or a diffuse growth pattern or show a mixture of both patterns. **Centroblastic/**centrocytic lymphoma is a disease of middle- and old-age and occurs rarely in childhood. Two-thirds of the patients present in stage **III** or **IV** at the time of diagnosis. Lymphoma cells can be detected in the bone marrow and peripheral blood in a high proportion of cases, both morphologically and by using immunological techniques.

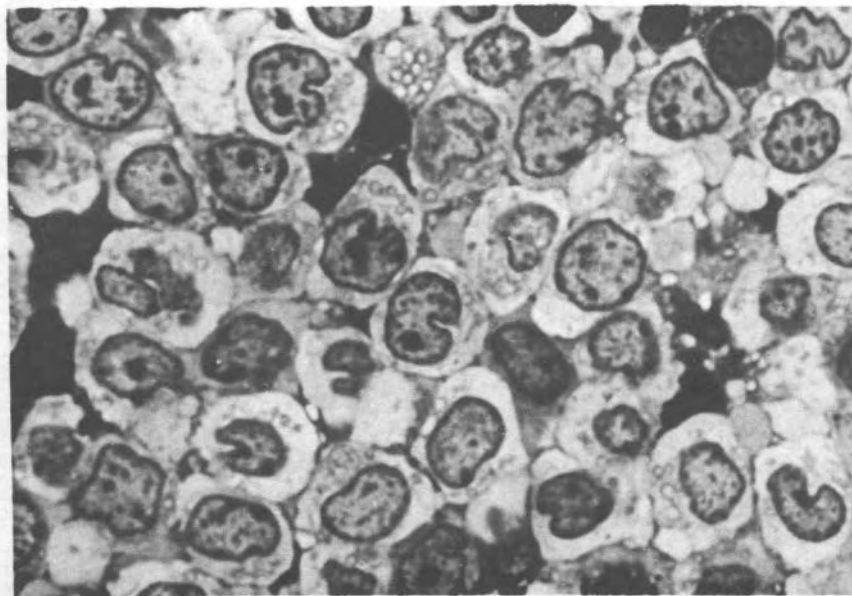


FIG. 4 : Plastic embedded section of a malignant lymphoma centrocytic. Note the irregularity of the nuclei compared with those shown in Figure 3. Toluidine blue X 1200.

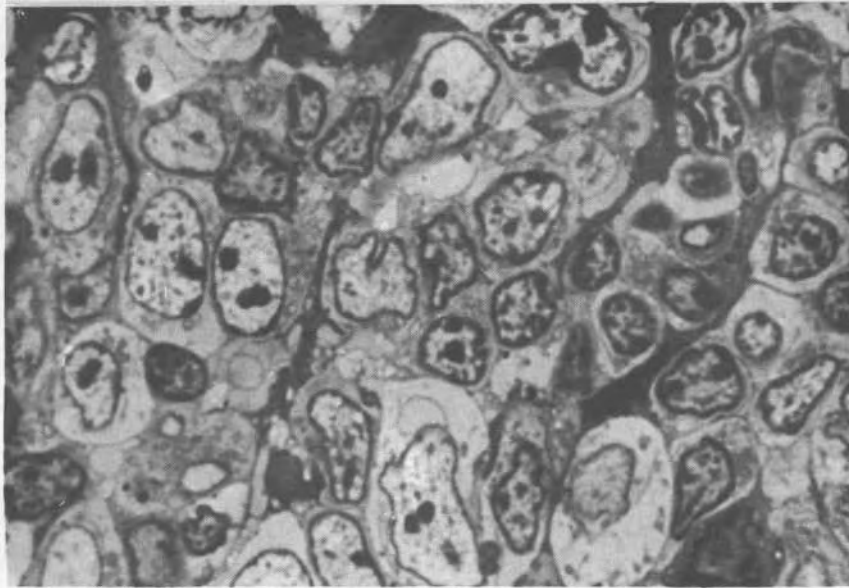


FIG. 5 : Malignant lymphoma centroblastic/centrocytic. Note that the centrocytes in this tumour are more pleomorphic than those shown in malignant lymphoma centrocytic (Figure 4). Toluidine blue X 1200.

Proposals have been made to sub-classify follicle centre cell lymphomas according to the proportion of centroblasts and centrocytes within the neoplastic proliferation.²⁴ It is not yet clear whether this has any prognostic significance but it does influence the appearance of the tumours and pathologists should be aware of this. The follicles in **centroblastic/centrocytic** lymphoma may abut one another or be widely separated by sheets of **interfollicular** T-cells amongst which there are often prominent high endothelial venules. With the passage of time, centroblastic/centrocytic lymphomas frequently progress from a **follicular** to a diffuse growth pattern, often associated with a progressive increase in the proportion of centroblasts to **centrocytes**.²⁵ Approximately 40% of the patients coming to autopsy are found to have centroblastic lymphoma. Lennert categorises these as secondary centroblastic lymphomas. They frequently show considerable pleomorphism.

Malignant Lymphoma – Centroblastic

Malignant lymphoma centroblastic usually has a diffuse growth pattern but is occasionally

follicular or follicular and diffuse. In the Western world it is the commonest of the large cell lymphomas. It may present *de novo* or following a pre-existing **centroblastic/centrocytic** lymphoma when it may be categorised as secondary centroblastic lymphoma.

The term 'centroblastic lymphoma' is usually used when greater than 50% of the tumour cells have the morphology of centroblasts. There are usually admixed centrocytes and if these predominate the tumour would be categorised as **centroblastic/centrocytic** diffuse. Typical centroblasts can be recognised by their rounded vesicular nuclei, usually with 3 – 4 nucleoli often attached to the nuclear membrane (Fig 6). The cytoplasm forms a narrow well-defined deeply basophilic ring around the nucleus. Scattered cells with prominent central nucleoli, resembling **immunoblasts**, are frequently seen in variable numbers within centroblastic lymphomas. In some tumours the cells can exhibit considerable degrees of nuclear pleomorphism with multinucleated cells showing some resemblance to Reed-Sternberg cells. Pleomorphism is particularly common in secondary centroblastic lymphomas.

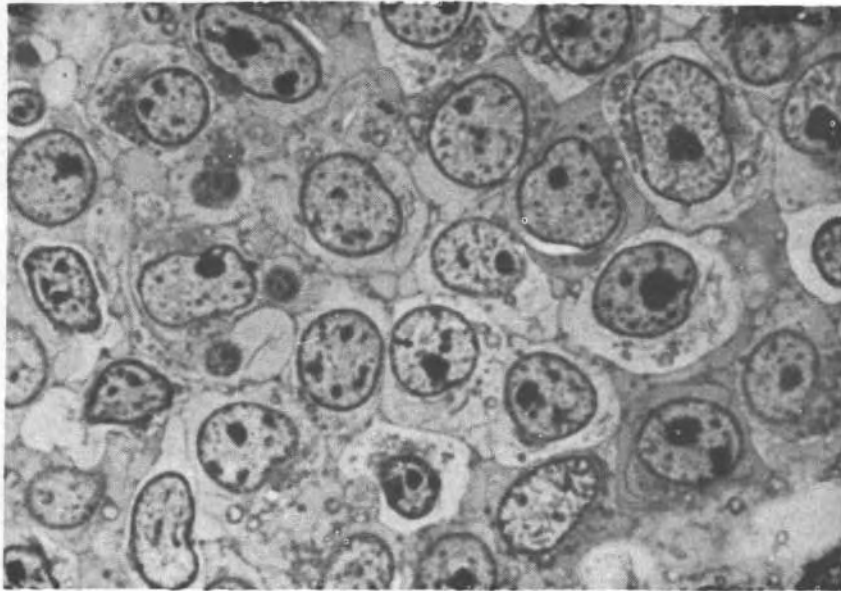


FIG. 6 : Plastic embedded section of malignant lymphoma centroblastic. The majority of cells have multiple nucleoli, sometimes attached to the nuclear membrane. Some cells have prominent central nucleoli and might be designated as immunoblasts. Toluidine blue X 1200.

Malignant Lymphoma - Immunoblastic

The term 'immunoblastic lymphoma' has been used rather indiscriminately by many authors and the distinction from centroblastic lymphoma is not always apparent. Immunoblasts characteristically have rounded nuclei with a prominent central nucleolus. Their cytoplasm is basophilic and they may show plasmacytic differentiation. Cells of this morphology are frequently found in varying numbers in centroblastic lymphomas. We use the term 'immunoblastic lymphoma' only if the tumour is composed predominantly of cells of this type.

Burkitt's Lymphoma

In the Kiel classification Burkitt's lymphoma is wrongly categorised as a lymphoblastic lymphoma. The cells of Burkitt's lymphoma have membrane immunoglobulin and represent a later stage in the B-cell maturation sequence than lymphoblastic lymphoma. Lukes and Collins²⁶ categorised Burkitt's lymphoma as a tumour arising from small non-cleaved follicle centre cells (Fig 7). We believe that the characteristic clinical and anatomical features of

Burkitt's lymphoma, with its predilection for the jaws and abdominal viscera without involvement of the peripheral lymph nodes and spleen, is due to the fact that this is a tumour of mucosa-associated lymphoid tissue. Although the tumour was originally described in Africa, morphologically similar tumours occur sporadically throughout the world. Many of these, however, have a different anatomic distribution from Burkitt's lymphoma. In particular, they involve the nasopharynx or the terminal ileum; sites at which Burkitt's lymphoma rarely occurs. These would also appear to be tumours of the mucosa-associated lymphoid tissue. However, they may be further along the B-cell differentiation pathway than Burkitt's lymphoma. This is illustrated by the fact that they sometimes show features of centrocytic differentiation and may also contain cytoplasmic immunoglobulin. It may be that the characteristic features of Burkitt's lymphoma are due to a combination of unique aetiological factors. The role of the EB virus may be to freeze the tumour cells in a state in which further differentiation is no longer possible.

