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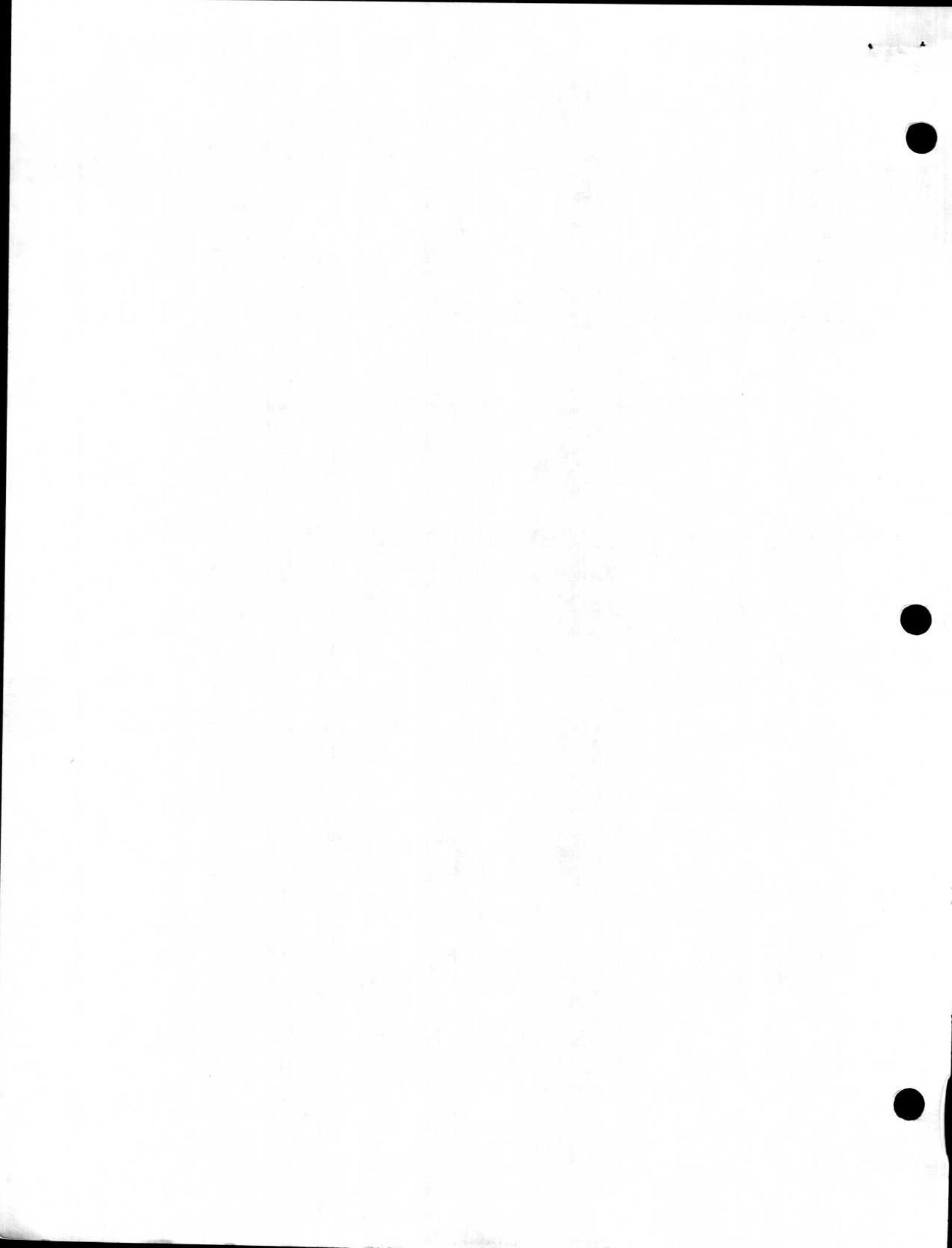
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## RAPID DIAGNOSIS OF BURKITT'S LYMPHOMA NEEDLE ASPIRATION

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### SUMMARY

The technique of needle aspiration, while giving somewhat less definitive results with Burkitt's lymphoma than biopsy and touch preparation, is extremely rapid and does not require a general anaesthetic. Aspiration may safely be performed in the orbit or under field conditions where biopsy would not be advisable. Good aspiration smears, when obtained, are as diagnostic as touch preparations. When the technique fails to yield adequate material, a biopsy may be performed as well.

### INTRODUCTION

Burkitt's lymphoma (BL) is the commonest form of cancer in many tropical areas (Stewart et al. 1973). It is rather remarkably responsive to chemotherapy, and long-term survival can be effected by treatment in up to 50% of cases, with temporary remission in virtually all (Morrow, Pike and Kisuule 1967; Clifford 1966; Ziegler et al. 1970; Williams 1971). Since central nervous system involvement, which considerably worsens the prognosis, becomes more likely with each passing day if treatment of this rapidly growing tumour is delayed, early diagnosis is of considerable importance. However, as treatment with cytotoxic agents is not without its dangers and as a number of infectious

and neoplastic processes can mimic BL, it is advisable to establish a microscopic diagnosis before beginning treatment.

One of us (E.H.W.) has diagnosed and treated about one hundred BL cases (Fig. 1) in West Nile District of Uganda over the past 12 years, using punch biopsies from which touch preparations (imprints) are made, and sending the specimen elsewhere for histological examination, usually waiting about 10 days for the pathologist's report to return. Recently, however, we have also performed needle aspiration of the tumour when the diagnosis has been suspected, often obtaining a diagnosis within a few minutes and without the use of a general anaesthetic. The technique and its

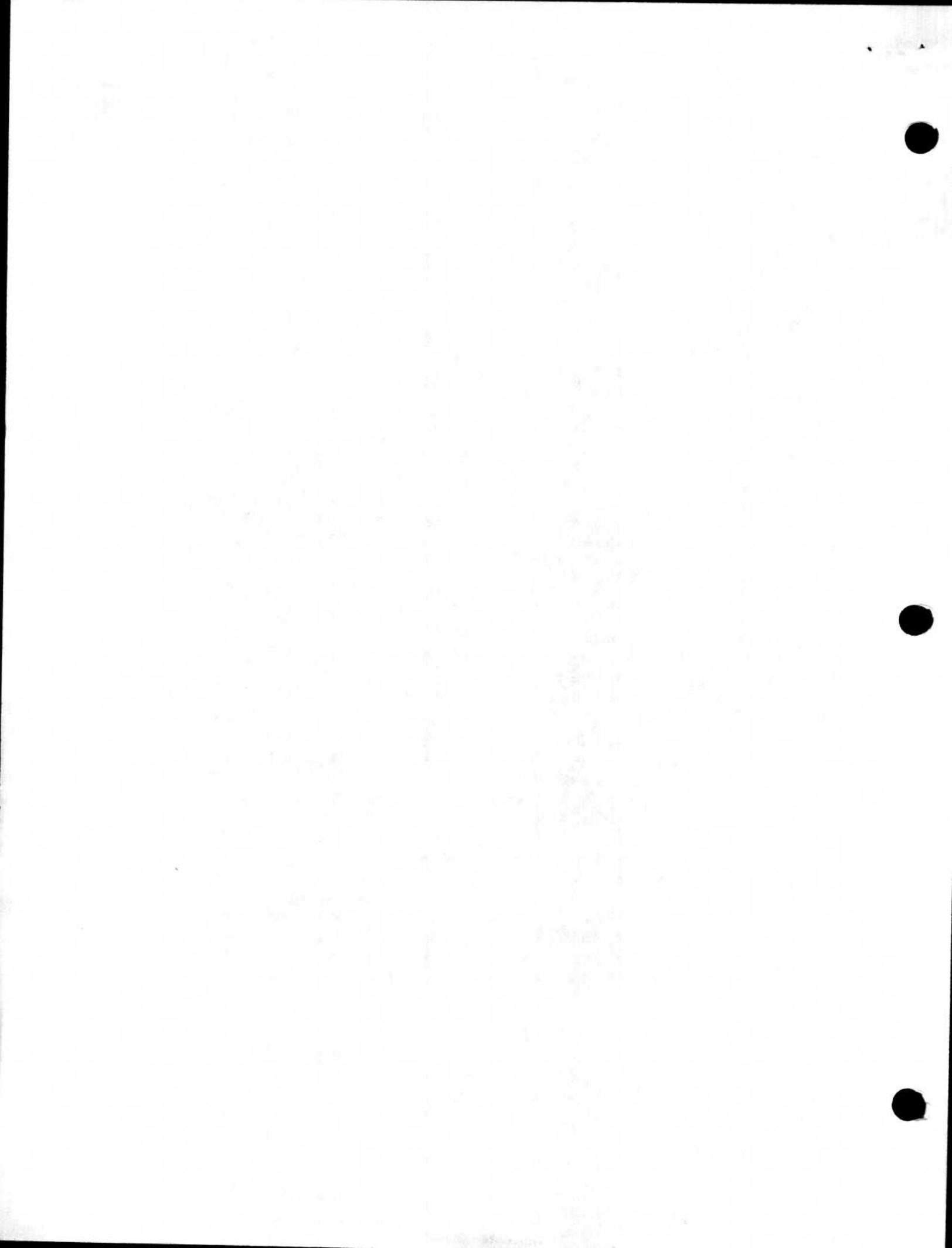




Fig. 1. The Burkitt's lymphoma patient from whom the specimens in Fig. 2 and 3 were taken. She had a small tumour of the right mandible (not visible), extensive ascites, and massive ovarian tumours.

results are described in the hope that others may find it useful.

#### METHODS

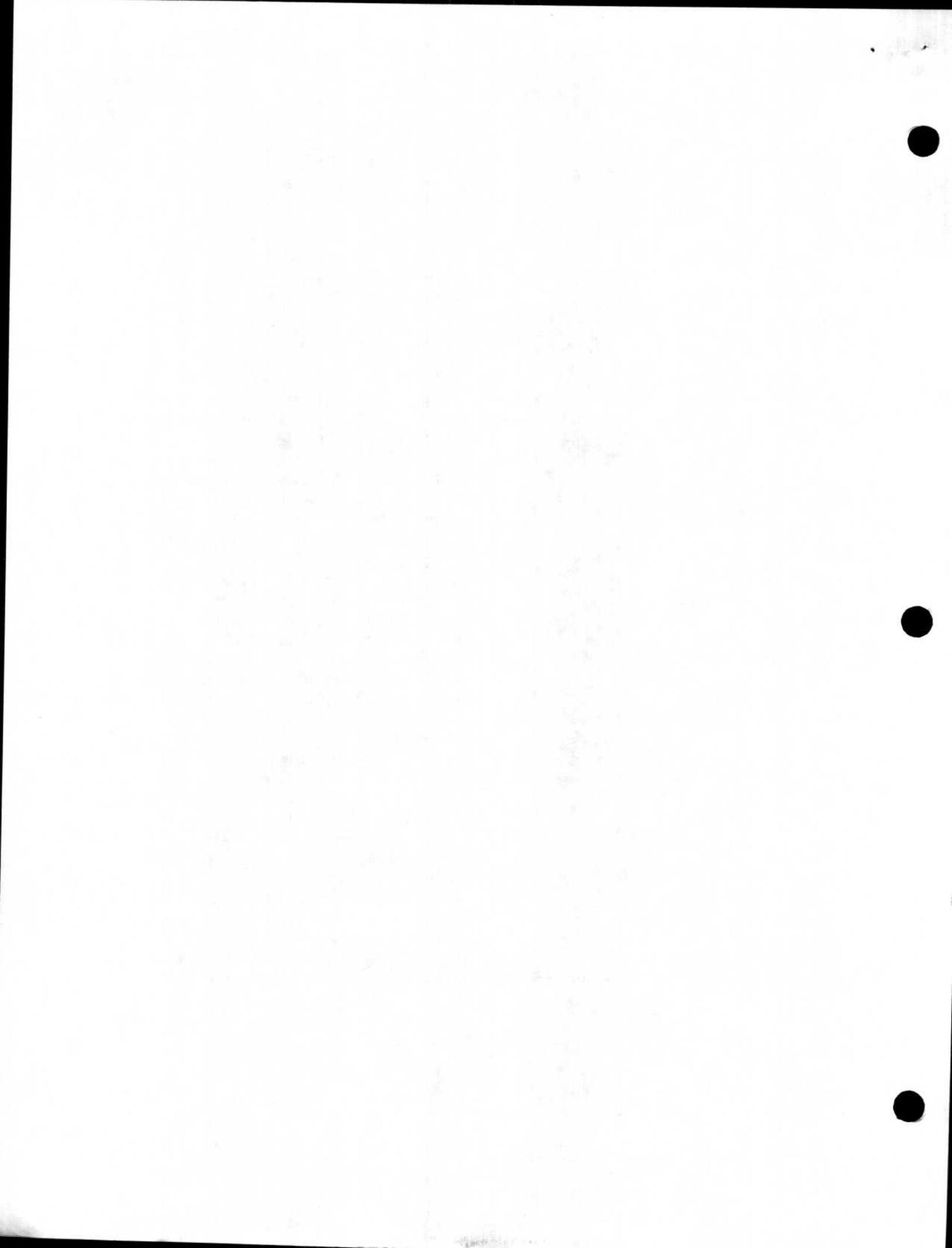
A dry 2 to 10 ml syringe and number 21 needle are used. Disposable equipment is

quite suitable. Where possible, jaw tumours are approached through the mouth, the needle being thrust straight into the tumour mass and usually causing very little pain. With subcutaneous or intra-abdominal tumours a small bleb of local anaesthetic under the skin may be advisable, although a quick stab as for an intramuscular injection is almost painless. Tumours of the liver and ovary and other intra-abdominal tumours may be aspirated safely if the patient is sufficiently reassured and/or sedated so that movement does not occur during the aspiration. With small retro-orbital tumours a general anaesthetic is necessary, and the needle must be inserted behind and around the eye in several places, as these tumours are often difficult to locate. Aspiration may be the only means of diagnosing an orbital tumour where biopsy might be unjustifiably destructive.

With the needle in the tumour, the plunger of the syringe is withdrawn quickly a few times to draw cells into the needle. The needle is then withdrawn from the tumour and its contents squirted onto microscope slides by repeated forcible expulsion of air from the syringe. If the tumour is BL, there is usually ample material for several slides. The slides are air dried, fixed and stained as for touch preparations.

It is important not to have local anaesthetic in the syringe or in the track of the needle, or the cells may be distorted. When preparing smears of cerebrospinal fluid for detection of BL cells, however, we have surprisingly enough obtained the best results by adding an equal amount of plain water before centrifuging lightly and preparing smears of the sediment.

Aspirates may be stained with any Romanowsky stain, but we have found Simeon's stain (Mackie and McCartney 1956), a modified Field's stain, very rapid



and reliable for aspirates and touch preparation.

The air-dried touch preparations or aspirates are fixed by dipping in 95% ethanol, then are allowed to dry or are rinsed in water. They are stained in Stevenel's Blue for a few seconds, rinsed in water, dipped in 0.5% Eosin momentarily, rinsed again and then dipped very briefly again in Stevenel's Blue before a final rinse and microscopic examination (Fig. 2 and 3). The staining can be adjusted by returning to either solution until the correct intensity of red and blue is obtained; the slide is then

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*Stevenel's Blue*

Solution A

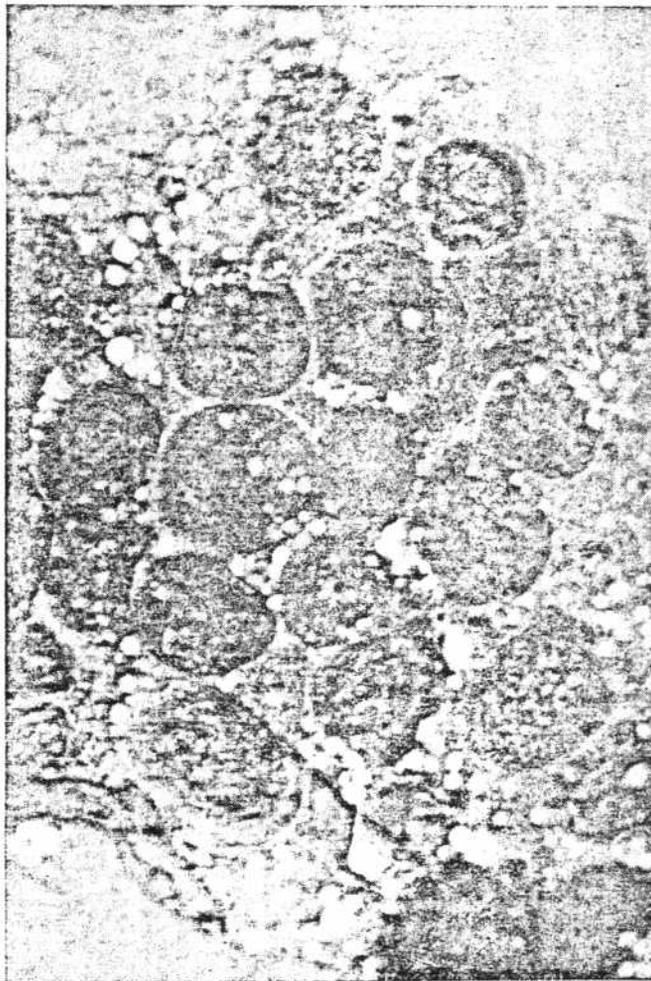
Methylene blue	1.0 g
H <sub>2</sub> O	75.0ml

Solution B

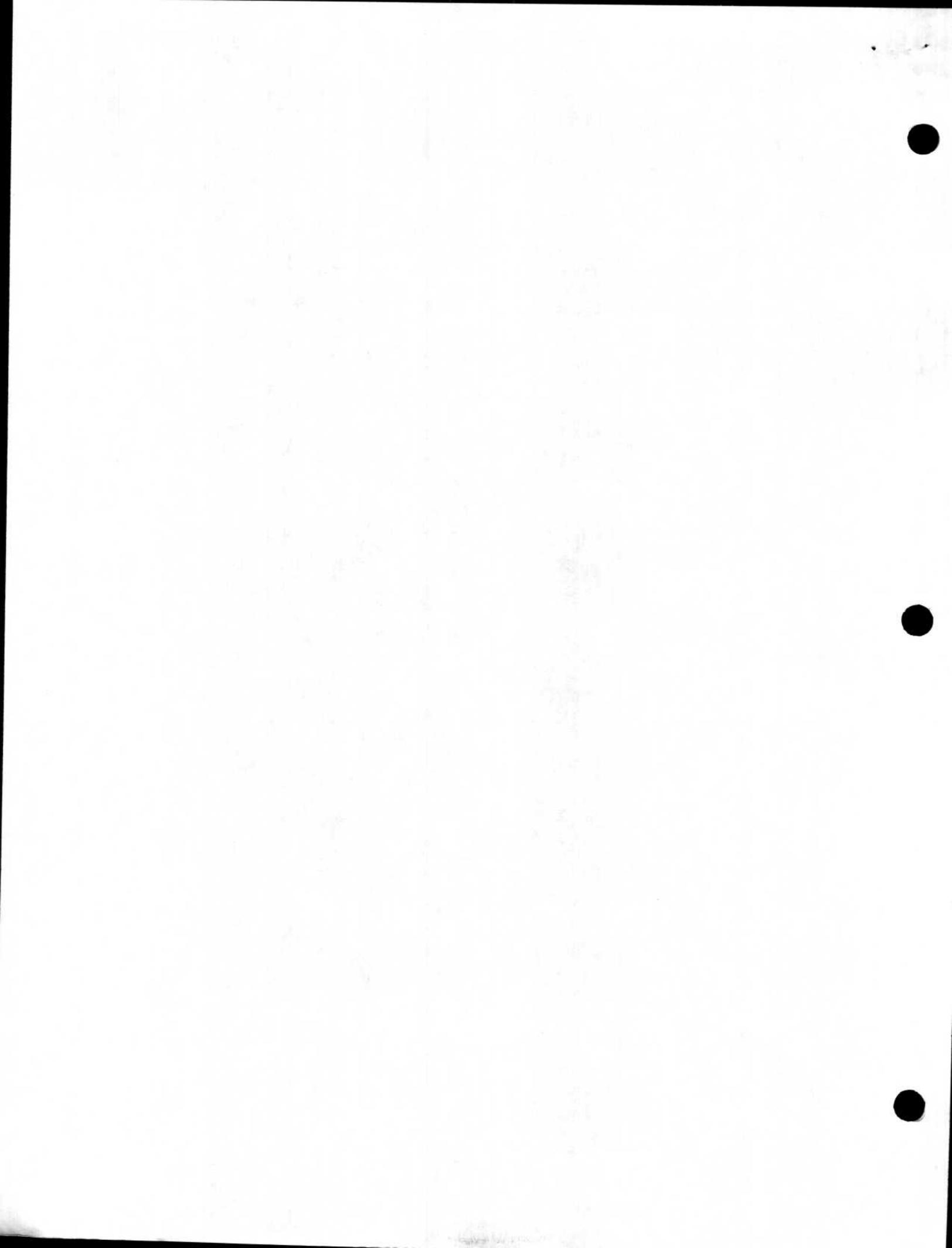
Potassium permanganate	1.5 g
H <sub>2</sub> O	75.0ml

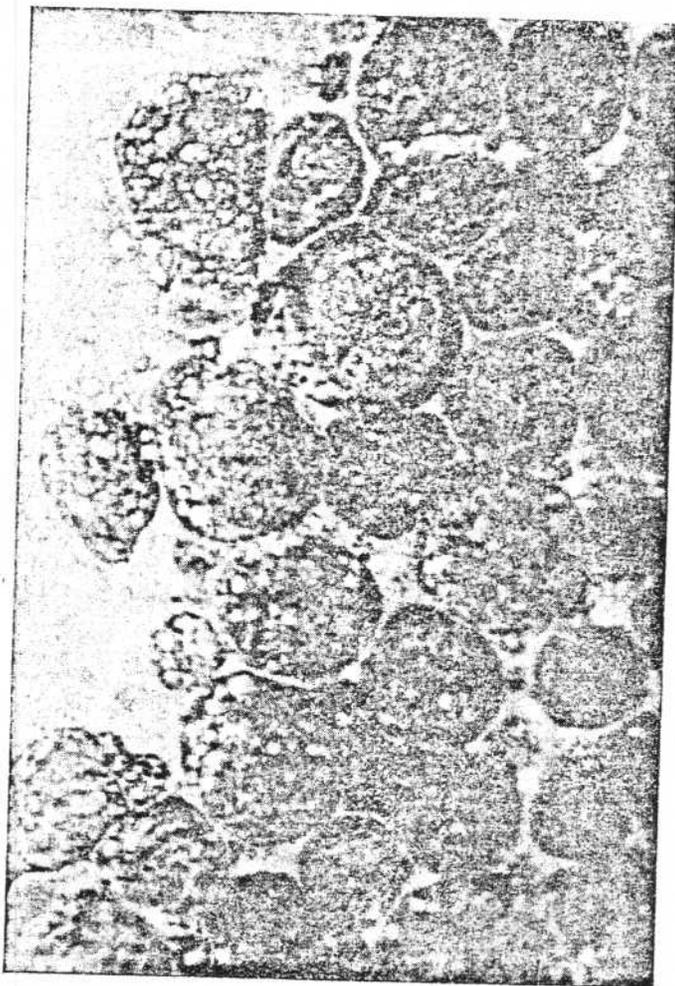
Dissolve completely and then mix A and B. A massive precipitate forms. Place in a boiling water bath for 1 hr, during which most of the precipitate redissolves. Filter before use.

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*Fig. 2.* Touch preparation from biopsy of mandibular tumour. Air dried, fixed in 95% ethanol, and stained by Simeon's method.





*Fig. 3.* Needle aspiration smear from the same site. Fixed and stained as in Fig. 2.

dried and a cover glass applied with a drop of a resinous mounting medium. The ability to adjust the depth of the staining is very advantageous in preparations of varying thickness.

In examining aspirates, the diagnosis is based on cytology of individual cells, since the histological relationships sometimes present in touch preparations of biopsies are not seen. The secret of accurate diagnosis appears to be in first locating those cells, often only a few, which are well fixed and stained, and then basing the diagnosis on their cytology. Cells which are distorted,

distended, or without cytoplasm should be disregarded. BL cells are of fairly uniform size, and have two to five nucleoli, evenly distributed chromatin, and occasional mitoses. The nucleus is often slightly indented. The scanty, non-granular basophilic cytoplasm forms a thin eccentric rim around the nucleus, and usually, but not invariably, contains small vacuoles. Lymphoblastic cells with abundant irregular cytoplasm and many vacuoles cast doubt on the diagnosis of BL.

To evaluate the usefulness of aspirates for tumour diagnosis, our aspirates and



touch preparations from 22 patients with BL and 9 with other neoplasms were re-examined. All patients had had biopsies examined histologically by the Pathology Department of Makerere Medical School, Kampala, Uganda. The aspirates and touch preparations were examined after covering the labels and assigning code numbers.

Each of us graded the slides as follows:

- 0—no evidence of malignancy
- 1—strong evidence for malignancy, probably BL
- 2—definite BL
- Other definite malignancy not BL

The sum of the scores by both observers was recorded, 0 or 1 being doubtful, 2 suggestive, 3 strongly suggestive (treatment indicated), and 4 diagnostic of BL.

## RESULTS

BL PATIENTS (biopsy read as BL)			
Aspirate		Touch preparation	
Score	No. of patients	Score	No. of patients
2	5 (23%)	2	1 (5%)
3	8 (36%)	3	2 (9%)
4	9 (41%)	4	18 (86%)
	22		21

OTHER MALIGNANCIES			
Aspirate		Touch preparation	
Score	No. of patients	Score	No. of patients
1/other	2	1/1	2
Other/other	4	Other/other	5
1/2	1		
	7		7

The total number of patients was 9, 2 lacking touch preparations and 2 lacking aspirates. The biopsy diagnoses were: histocytic lymphoma 2, Hodgkin's disease 1, retinoblastoma 1, hepatoma 1, Wilms' tumour 3, and reticulum cell lymphoma 1.

The case of Hodgkin's disease was diagnosed correctly by both observers by either aspirate or touch preparation. With the other tumours, however, the identification of the tumour was not accurate, the histocytic lymphomas being sometimes confused with BL and retinoblastomas with Wilms' tumour and/or BL. It should be noted that histologic sections of these tumours have also been difficult to interpret, one having been called retinoblastoma by one pathologist and BL by another.

## ACKNOWLEDGEMENT

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