**Device**

# Rapid diagnosis of intracranial fungal infection : Technical note.

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*Because of the poor prognosis associated with intracranial fungal infection caused by molds, a technique for their rapid diagnosis could greatly aid therapeutic decision making. Combining the Gomori-methenamine silver stain with tissue touch preparations can yield a definitive diagnosis within a few hours.*

**Key words:** *Fungal infections, Touch preparations, Frozen sections.*

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The touch preparation technique was first used for the pathological examination of central nervous system (CNS) tissue in 1927.[1] The importance of this method for tissue examination has been described for a variety of CNS lesions, but it has not been specifically applied to fungal infection.[2,3] The advantages of this technique include the preservation of cellular detail, diagnostic accuracy and the rapidity of the procedure.[3] Paraffin-embedded sections have the disadvantage of requiring more than a day to process.

## Technique

A touch preparation is made by gently applying freshly cut tissue to a glass slide and then fixing it in 95 % ethanol for 15-30 seconds.[4] After oxidation in 5 % chromic acid solution for one hour, the tissue is rinsed under running water for a few seconds. Residual chromic acid is removed with 1 % sodium bisulfite over 1 minute. The slide is washed four times in distilled water and then placed in a working methenamine-silver nitrate solution at 60° C for 45 minutes until the section turns light brown. Silver impregnation is confirmed microscopically after the slide is dippped in distilled water.

Six changes of distilled water precede a three minute toning step in 0.1 % gold chloride solution. After a distilled water rinse, unreduced silver is removed over three minutes with 2 % sodium thiosulfate. The specimen is washed for five minutes under running tap water prior to being counterstained with working light green solution for 30-45 seconds. Dehydration is performed in 95 % ethanol (2 dips and absolute ethanol (10 dips) before clearing in xylene for 5 minutes.[4] Permount is used for mounting and the specimen is immediately ready for microscopic review (Fig.1).

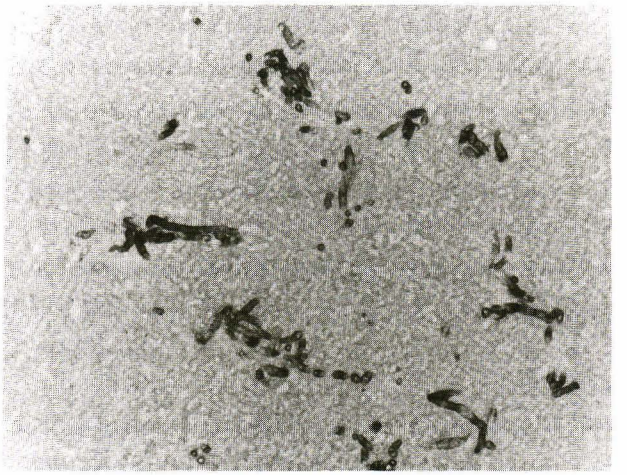

**Figure 1.**

## Discussion

As the number of immunocompromised patients who are at risk for developing CNS infectious or neoplastic processes continues to grow, the neurosurgeon is increasingly being called upon to provide a tissue diagnosis. This patient population includes those with the acquitted immunodeficiency syndrome, leukemia, lymphoma, and those receiving immunosuppressive medications after bone marrow or solid organ transplantation.[5] In these patients, infections of the CNS are often fatal and can be demonstrated on computed tomography scans of the head (Fig.2)[5-7] An aggressive approach to patients with treatable intracranial lesions may result in a prolongation of survival. In contrast, the diagnosis of a frequently fatal disease such

as an intracranial fungal infection caused by the *molds Aspergillus sp.* and the *Mucoraceae* may prompt the discontinuation of intensive medical therapy in a critically ill patient.

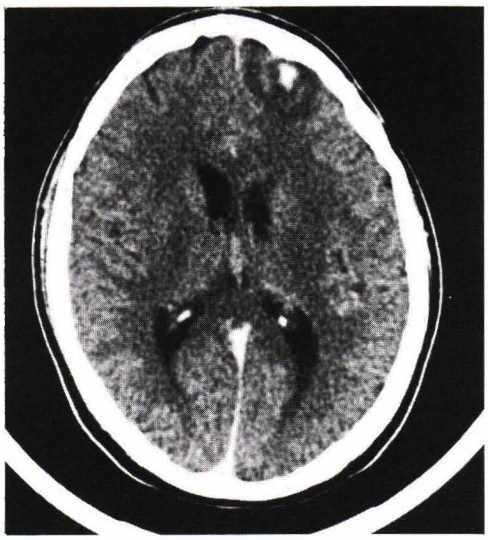

**Figure 2.**

In our experience, the demonstration of fungal hyphae on touch or frozen sections is difficult with hematoxylin and eosin stain because of the staining characteristics of the micro-organisms. Although the GMS stain is routinely used to diagnose fungal infections caused by molds, the exact characterization of the causative organism still requires culture. With the increasing use of stereotaxis to biopsy CNS lesion, the touch preparation technique is supposed to be an ideal way to examine small tissue samples by light microscopy, Touch preparation using the GMS stain will allow accurate identification of fungal hyphae and provide a rapid microscopic diag-nosis . The use of this procedure can result in a faster diagnosis of intracranial fungal infection that allows clinicians to choose an early , rational treatment plan.

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