What is this manual about?

This manual is designed to provide an overview of the services available in the Neuropathology Department and serve as a quick reference guide for all users. Laboratory Management is committed to:

- Ensure stringent adherence to quality in all laboratory procedures that meet requirements of internal and external quality assessment tests.
- Ensure appropriate collection, transport and processing of samples to ensure optimal performance of all the tests required.
- Ensure reports are accurate, timely and clinically useful.
- Ensure use of procedures and methods that are up to date with current practices.
- Ensure continued staff training at all levels to keep them updated with recent advances in the field.
- Periodically assess user satisfaction by feedback forms.

Copies of this manual are available in specimen collection centres, wards, OTs, casualty, short stay wards. Electronic version is available on the NIMHANS website.

Contact Us

Location: Department of Neuropathology, First floor, Administrative block, NIMHANS
Postal address: Department of Neuropathology, NIMHANS, Hosur Road, Bangalore 560 029
Phone: Off: 080-26995130 (for report enquiries, call between 9.30-4.30pm on working days only)
Email: neuropathology2008@gmail.com
Website: www.nimhans.kar.nic.in/neuropathology/.default.htm
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Our Services

Neurooncology (Brain tumor diagnosis)

- Intraoperative frozen section
- Brain tumor diagnosis: routine histology
- Immunohistochemistry

FISH: 1p19q codeletion
MGMT methylation (on research mode)

Muscle diseases

- Routine histology
- Enzyme histochemistry
- Immunohistochemistry
- Electron microscopy

Recently introduced: Western blot, Mitochondrial enzyme assays (currently on research mode)

Peripheral neuropathies

- Routine histology
- Semithin / electron microscopy (where indicated)
  Skin punch biopsy for small fibre neuropathy (on research mode only)
  Note: teased fibre preparation is NOT performed
- Routine histology for Lafora bodies, vasculitis
- Electron microscopy for NCL/storage disorders (where indicated)

Skin biopsies
Getting Started

Location of the Department & its sections

Main Histopathology Laboratory is located on the first floor of Administrative Block.

Electron Microscopy section is also located in the same building in the basement.

Transfusion Medicine Centre is housed in the Neurocentre Building, first floor, rear wing.

Clinical Pathology & hematology Section is located in the Casualty Block on the first floor.

Mortuary is housed in a separate building behind the Casualty and New Psychiatry buildings.

Human Brain Tissue Repository (Brain Bank) is housed in the Neurobiology Research Centre, ground floor, Room No. 004.
## Departmental working hours

<table>
<thead>
<tr>
<th>SECTION</th>
<th>Opening hours</th>
<th>Closing time</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Histopathology</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weekdays (Mon-Sat)</td>
<td>9.00 am</td>
<td>4.30 pm</td>
</tr>
<tr>
<td>Second Saturday, &amp; all closed Govt holidays declared by Institute (Sundays holidays)</td>
<td>9.30am</td>
<td>3.30pm</td>
</tr>
<tr>
<td><strong>Electron Microscopy</strong></td>
<td>9.00 am</td>
<td>4.30 pm</td>
</tr>
<tr>
<td>Weekdays (Mon-Sat) only</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Closed on second Saturdays, Sundays, &amp; Govt holidays declared by the Institute</td>
<td>---</td>
<td>---</td>
</tr>
<tr>
<td><strong>Clinical Pathology</strong></td>
<td>Open round the clock on all days</td>
<td>1st shift - 8.00am</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2nd shift - 3.00pm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3rd shift - 9.00pm</td>
</tr>
<tr>
<td><strong>Mortuary</strong></td>
<td>9.00am (all days including Sundays, &amp; holidays)</td>
<td>4.30pm</td>
</tr>
</tbody>
</table>

## Departmental faculty & contact numbers

<table>
<thead>
<tr>
<th>SECTION</th>
<th>Faculty</th>
<th>Conact No.s</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Histopathology</strong></td>
<td>Dr. SK Shankar (Emeritus Prof)</td>
<td>(2699)5786</td>
</tr>
<tr>
<td></td>
<td>Dr. Vani Santosh</td>
<td>(2699)5132</td>
</tr>
<tr>
<td></td>
<td>Dr. Yasha TC (HOD)</td>
<td>(2699)5133</td>
</tr>
<tr>
<td></td>
<td>Dr. N. Gayathri</td>
<td>(2699)5131</td>
</tr>
<tr>
<td></td>
<td>Dr. Anita Mahadevan</td>
<td>(2699)5137</td>
</tr>
<tr>
<td><strong>Electron Microscopy</strong></td>
<td>Dr. BK. Chandrashekar Sagar</td>
<td>(2699)5738</td>
</tr>
<tr>
<td></td>
<td>Dr. N. Gayathri</td>
<td></td>
</tr>
<tr>
<td><strong>Transfusion Medicine Centre</strong></td>
<td>Dr. Sundar Periyavan</td>
<td>(2699)5437</td>
</tr>
<tr>
<td><strong>Clinical Pathology</strong></td>
<td>Dr. Sangeetha Seshagiri</td>
<td>(2699)5750</td>
</tr>
<tr>
<td><strong>Mortuary</strong></td>
<td>Dr. Anita Mahadevan</td>
<td>(2699)5137</td>
</tr>
<tr>
<td></td>
<td>Dr. Yasha TC</td>
<td>(2699)5133</td>
</tr>
<tr>
<td></td>
<td>Dr. Vani Santosh</td>
<td>(2699)5132</td>
</tr>
<tr>
<td><strong>Human Brain Tissue Repository</strong></td>
<td>Dr. S.K. Shankar (Emeritus Prof)</td>
<td>(2699)5786</td>
</tr>
<tr>
<td></td>
<td>Dr. Anita Mahadevan</td>
<td>(2699)5137</td>
</tr>
</tbody>
</table>

### Section Phone number

<table>
<thead>
<tr>
<th>Section</th>
<th>Phone number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Office</td>
<td>(2699)5130</td>
</tr>
<tr>
<td>Residents Room (Post Doctoral Fellows)</td>
<td>(2699)5134</td>
</tr>
<tr>
<td>Discussion Room</td>
<td>(2699)5135</td>
</tr>
<tr>
<td>Laboratory</td>
<td>(2699)5136</td>
</tr>
<tr>
<td>Electron microscopy</td>
<td>(2699)5138</td>
</tr>
<tr>
<td>Clinical Pathology</td>
<td>(2699)5516</td>
</tr>
<tr>
<td>Mortuary</td>
<td>(2699)5213</td>
</tr>
<tr>
<td>Brain Bank (Neurobiology Research Centre)</td>
<td>(2699)5786</td>
</tr>
<tr>
<td>Museum (Neurobiology Research Centre)</td>
<td>(2699)5582</td>
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</table>
## List of Tests & Turn Around Time

<table>
<thead>
<tr>
<th>SI No</th>
<th>Tissue Type</th>
<th>Specific Test</th>
<th>Turn around Time*</th>
<th>#</th>
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<tbody>
<tr>
<td>1</td>
<td>Tumor tissue (Formalin fixed)</td>
<td>Histopathology</td>
<td>4 days</td>
<td>7 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Special stains or Immunohistochemistry</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Fresh tissue</td>
<td>Intra-operative smear/squash</td>
<td>30 mins after sample receipt</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Epilepsy surgery</td>
<td>Histopathology/Immuno-histochemistry</td>
<td>15 days</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Fresh Muscle</td>
<td>Histopathology &amp; enzyme histochemistry Immunohistochemistry</td>
<td>4-7 days 4 weeks</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Nerve biopsy (glutaraldehyde)</td>
<td>Histopathology examination</td>
<td>4-7 days</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>Skin (formalin/glutaraldehyde)</td>
<td>Routine histopathology examination Electron microscopy</td>
<td>4-7 days</td>
<td>6 weeks</td>
</tr>
<tr>
<td>7</td>
<td>Liver biopsy (formalin/alcohol)</td>
<td>Histopathology with special stains</td>
<td>4-7 days</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>Brain biopsy (Fresh)</td>
<td>Histopathology &amp; Immunohistochemistry</td>
<td>10 days</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>Paraffin block</td>
<td>Routine Immunohistochemistry</td>
<td>4 days 7 days</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Slides</td>
<td>Histopathology</td>
<td>2 days</td>
<td></td>
</tr>
</tbody>
</table>

* In working days
# General guidelines: Routine HPE - 4 days
Special stains/Immunohistochemistry - 7 days

## General Instructions for All Samples

All samples and requests are screened for the following:

1. All samples must be accompanied by a histopathology request form, completely filled with all relevant details. The request form for inpatient NIMHANS cases and outside Referral cases are different and non-exchangeable.
2. Request should be from treating physician. Requests from patient party are not entertained.
3. All samples should have at least two identifiers – patient name, and hospital number, hospital name (if from outside) with age and gender.
4. Personal identifiers of the sample and the request form should match.
5. Appropriate fixative/ container must be used; and transported/ stored in appropriate conditions to ensure stability of sample and optimum results.

### Sample receipt timings:

Mon-Sat: 9.00am – 4.30pm
Second Saturdays, Institute holidays: 9.00am-3.30pm
Sundays are holidays.
Neurosurgical biopsies

Intraoperative frozen – facility available ONLY for hospitals located close to NIMHANS

**Caution**

1. Inform pathologist prior to sending sample for frozen
2. Samples should be sent immersed in sufficient volumes of saline to prevent drying up in transit.
3. Samples SHOULD reach the Lab within 30mins-1hour during lab working hours.
4. Intraoperative frozen requests after lab working hours (4.30 pm) or holidays CANNOT be performed.
5. Any sample associated infection risk SHOULD BE CLEARLY STATED ON THE REQUEST FORM (eg., retroviral/HbsAg/suspected prion disease) and marked 'BIOHAZARD'

Neurosurgical biopsies

Surgical samples should be sent in 10% formalin

- Ensure sample is labelled correctly, placed in sufficient volumes of formalin and is accompanied by complete clinical details
- All sample transport requirements (see below for details) should be strictly adhered to

Small biopsies
(stereotactic/CT/MR guided/endooscopic)

**Samples to be sent in 10% formalin:**

1. If tissues are collected, in multiple bottles, the bottles to be labelled serially clearly INDICATING sites of biopsy
CAUTION

Diagnostic brain biopsy is a special test and requires extensive co-ordination between clinician and pathologist BEFORE biopsy is done for optimal yield. Hence strictly follow the instructions below:

1. Brief the pathologist as to complete clinical details prior to biopsy
2. Confirm date of biopsy previous day
3. Timings: Ensure samples reach within working hours
4. Indicate clearly if risk of infection (eg., retroviral status/HBsAg/prion etc) as it requires special precautions from our side. Label biopsy as ‘BIOHAZARD’

Tests performed:
- Routine and special stains;
- Immunohistochemistry

Neurosurgical biopsies

Diagnostic brain biopsies: In formalin

Biopsy site: based on clinical/imaging findings

Biopsy size: 1x1cm cube with overlying meninges (+ dura if pachymeningeal disease)

Fixative: 10% formalin

Note: Send in FRESH STATE for hospitals within Bangalore ONLY. Please discuss with pathologists PRIOR to sending

Epilepsy surgeries: In formalin

Label each sample clearly as to site

Fixative: 10% formalin
Aspirates

**Caution**

Any sample associated infection risk SHOULD BE CLEARLY STATED ON THE REQUEST FORM (eg., retroviral/HbsAg/suspected prion disease) and marked ‘BIOHAZARD’

Neurosurgical biopsies

**Cyst fluid aspirates:**

1. Fluids for pathological examination should be sent in 10% formalin
2. Additional sample for microbiological studies need to be submitted to your own lab in fresh state depending on diagnostic requirements (eg., for cytopsin/culture)

**INFECTIVE LESIONS (EG., ABSCESS WITH PUS)**

1. Send one sample in 10% formalin to Neuropathology
2. Send one sample (fresh) to your own Microbiology lab for additional studies like culture (esp if fungal infection is suspected)
**Muscle Biopsy**

**Biopsy site:** dependant on clinical/ENMG studies
- Obtain biopsy from affected muscle, with MRC Grade 3-4/5. Usual sites – vastus lateralis, biceps
- If involvement is purely distal, chose distal muscles (MRI/USG can help selection)

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**CAUTION: Collection & Transportation**

1. Fresh samples can be sent ONLY from hospitals within Bangalore or close enough so that biopsies reach WITHIN MAXIMUM of 12hrs from time of biopsy and should reach the lab the same day before 12 noon on any working day (Mon-Sat).
2. Biopsies are NOT accepted on Sundays, second Saturdays & Govt holidays
3. All other hospitals must send muscle in 10% formaldehyde

**Tests possible ONLY on fresh samples**
- a. Enzyme histochemistry
- b. Immunohistochemistry, Western blot for muscular dystrophies
- c. respiratory chain enzymes (for mitochondrial disorders)

**Formalin Fixed samples:**
Only Routine histology possible

**For electron microscopy:** muscle should be sent in 2.5% glutaraldehyde

**If paraffin blocks are sent:**
1. Only routine H&E and special stains are possible
2. Enzyme histochemistry/immunohistochemistry/electron microscopy not possible

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**Muscle Biopsy**

**Biopsy instructions (for fresh samples)**

- **Wrap muscle in saline soaked gauze**
- **DO NOT USE EXCESS SALINE**
- **Biopsy size – 2x2cms**

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**Biopsy specifications**
- Dissect with minimum trauma along the long axis of the muscle fibers
- Do not use electrocautery or a muscle clamp
- Additional bits if more tests are needed (eg, culture/enzyme studies for mitochondrial or rare genetic disorders)
- Provide site, date and time of biopsy
- State what tests are required

- **AVOID severely affected muscle with MRC grade <3**
- **AVOID muscle traumatized by IM injections or by EMG.**
Nerve Biopsies

**Biopsy site:** dependant on clinical/nerve conductions
- Clinically & electrophysiologically involved nerve
- Sural nerve/superficial peroneal nerve/dorsal cutaneous nerve/superficial radial (commonly biopsied)

**Fixative:**
- USE ONLY 2.5% gluteraldehyde
- DO NOT USE 10% buffered formalin
- DO NOT SEND FRESH (even for metachromatic stains)

**Tests performed**
a. **Routine Histopathology**
b. **Special stains:**
- Myelin stains (Kulchitsky pal)
- Masson trichrome for collagen
- Cresyl violet for metachromatic granules
- PAS-diastase stain & Lugol’s iodine (polyglucosan bodies)
- Fite Ferraco (for lepra bacilli)
- Congo red (for amyloid) etc
c. **Immunohistochemistry** (eg., ubiquitin for polyglucosan bodies, neurofilament for axons etc)
d. **Electron microscopy** (USE ONLY 2.5% glutaraldehyde)

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Nerve biopsies

**CAUTION: BIOPSY SPECIFICATIONS**
- Take a minimum length of 2 cms biopsy
- AVOID pinching/crushing nerve during procedure
- DO NOT tie nerve with thread
- AVOID COMMERCIALLY AVAILABLE CIDEX
- Use 2.5% gluteraldehyde
- Avoid 10% formalin as fixative
Skin Biopsies

**Biopsy site:** dependant on clinical differentials

- For PME diagnosis: axillary skin biopsy (biopsy should be DEEP enough to include Subcutaneous FAT)
- For vasculitis/Hansen’s: lesional site (margin)
- For amyloid: abdominal fat aspirate/biopsy
- For CADASIL/Neuroaxonal dystrophy/othersuspected storage diseases: axillary skin biopsy

**Fixative:**

- 2.5% gluteraldehyde for PME/CADASIL/Storage (wherein electron microscopy maybe required)
- 10% buffered formalin for others (vasculitis/amyloid/Hansen’s)
- SALINE/FRESH SPECIMEN NOT REQUIRED

**Tests performed**

a. **Routine Histopathology**

b. **Special stains:** PAS-diastase stain & Lugol’s iodine (for polyglucosan bodies), Fite Ferraco (for lepra bacilli), Congo red (for amyloid)

c. **Immunohistochemistry** (eg., ubiquitin for polyglucosan bodies, neurofilament/ubiquitin for neuroaxonal dystrophy etc)

d. **Electron microscopy:** Essential for: NCL, CADASIL, neuroaxonal dystrophy, storage disorders

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**Skin biopsies**

**CAUTION**

- Ensure biopsy is deep enough to include subcutaneous fat
- DO NOT USE COMMERCIAL AVAILABLE CIDEX

Note: Skin punch biopsy for small fibre neuropathy IS A DIFFERENT PROCEDURE with specific biopsy sites, fixative (paraformaldehyde-lysine-periodate), and cold chain. This test is not being routinely performed (available for NIMHANS patients ONLY on research mode)
SAMPLE ACCEPTANCE CRITERIA

Samples should be accompanied by Histopathology request form from TREATING PHYSICIAN (not patient party), completed in all respects with Priority Status of Request (Urgent, intraoperative frozen requirement or Routine) CLEARLY INDICATED ON THE REQUEST FORM

The following information must be documented in a LEGIBLE manner on the Request form.
1. *Patient’s Full Name (legible, preferably in CAPITALS).
2. *Patient’s Age/Sex
3. Consultant name / address / phone number
4. *Specimen Type
5. Date and time of collection
6. Complete Clinical Details with results of relevant investigations and Clinical Diagnoses considered
7. Test requested
8. Priority Status of Request (Urgent or Routine)
9. Details of any sample associated Infection Risk

Items marked with an * are minimum identifiers and failure to provide the data required may lead to delay in processing of the sample and release of reports.

ALL Samples should be labeled with the following
1. *Patients’ Full name (legible, preferably IN CAPITALS)
2. *Age/Sex
3. *Hospital name
4. *Specimen Type

Paraffin blocks/slides submitted for review:
Request form should contain block/slide identification number and number of blocks/slides submitted in addition to complete clinical details and diagnoses considered.

NON-COMPLIANCE WITH ACCEPTANCE CRITERIA

Compliance with acceptance criteria is mandatory for the referring clinicians to ensure accuracy of results.

Patient samples are precious and hence will not be rejected. However, non compliance with the above requirements will interfere with the correctness of results or lead to delay/withholding of results/ inability to perform certain tests.

The laboratory management does not bear responsibility for this.

Examples of non compliance:
1. Samples unaccompanied by completed Histopathology request form
2. Mismatch of information on the label and the request
3. Inappropriate transport temperature particularly for muscle biopsies
4. Excessive delay in transportation in case of fresh samples (muscle biopsies after 12.00pm)
5. Inappropriate transport medium
   a. specimen received in saline and autolysed at time of receipt
   b. dried specimen.
   c. leakage of fixative due to breakage of container

Safety precautions for suspected “CJD” / prion diseases

1. Operative procedure: Follow universal precautions
2. Send Biopsy Tissue fixed in 10% formalin to our lab, well sealed and double bagged to prevent leakage. DO NOT SEND FRESH
3. DECONTAMINATION PROCEDURES (after biopsy)
   a. Disposable clothing / instruments / gloves / cotton / gauze etc – double bag, immerse in conc hypochlorite and mark for incineration
   b. Disposable sharps – immerse in sharps can with concentrate hypochlorite (DO NOT DILUTE) for 30 mins, mark for incineration
   c. Non-disposable instruments - Steam autoclave at 134-136°C for 60mins (gravity displacement) or immerse in 2M NaOH
   d. Work surfaces, glassware – 2M NaOH

Note: 2M NaOH is prepared by dissolving 80gms NaOH in 1 litre H2O. reaction is exothermic(Ref: CJD Surveillance Unit, Edinburgh)
Release of laboratory reports

Telephonic reports

REPORTS WILL NOT BE GIVEN ON TELEPHONE TO PATIENTS/RELATIVES
- Reports over telephone is usually AVOIDED except for intraoperative frozen reports
- Reports maybe provided on telephone only to the treating physician by faculty/resident

- WORKING hours (9.00 am– 4.30pm) on working days (Mon-Sat)
- No enquiries possible on second Saturdays/holidays and Sundays as office is not manned

Release of laboratory reports

- All reports typed in by the office staff will be checked for typographic errors and patient demographic details by residents/faculty
- A printed copy of reports will be sent by post (NOT courier) and a copy by e-mail to the referring clinician ONLY (NOT patient party), if email id is provided
- For local cases, copies of reports will be handed over to patient party on producing payment receipt

Reports of test results considered “Critical Alert”

In consultation with clinicians, following have been considered as requiring “critical alert” for immediate notification to treating physician

- diagnosis of fungal infection, viral infections (particularly HSV) and protozoal infection (toxoplasmosis)
- CNS lymphoma, germinoma (radio/chemosensitive)
- CSF cytopsin positive for malignant cells/cryptococci

Reports are immediately informed (telephonically) to treating physician/resident in charge of the case. The clinician informed including date and time is documented on the request form. A final written report is sent subsequently.

Reports of intraoperative frozen sections

- Report will be informed telephonically to the clinician by the resident/faculty after verifying name of patient and operating surgeon
- Entry of the person (resident/surgeon/anesthetist/OT sister/OT technician) receiving the report, date and time will be recorded on the reverse side of the HP request form.
- Inability to reach anyone in OT to give reports will also be recorded.
Retention time of specimens/blocks/reports after reporting

Retention time of specimens/blocks/reports after reporting

1. Tissues remaining after grossing and generation of reports will be reviewed by reporting faculty. Those of research/teaching interest will be retained, and others will be discarded within 6-8 months from the date of reporting.

2. All paraffin blocks and slides will be preserved under appropriate conditions, chronologically numbered and will not be discarded.

3. Printed copies of all reports are bound and preserved protecting from any accident/loss due to fire etc.

4. Frozen muscle bits are retained for 10 years. Extra unstained cryosections for immunohistochemistry are retained for 1 year.

Additional tests that can be performed on paraffin blocks can be requested at any time. Additional tests on frozen muscle cryosections possible only if requested within the retention period.

Review of reports

Review of reports

Review of previously reported slides can be undertaken at any time as paraffin blocks are not discarded.

All requests should be accompanied by request form with clinical details and specifying reasons for requesting review.

Request for duplicate slides

Duplicate slides for referral to oncology centres/ to obtain second opinion can be provided

Requests should be from treating physician only.
### Summary of tests

<table>
<thead>
<tr>
<th>Sample</th>
<th>Tests possible (dependant on fixative)</th>
<th>Appropriate fixative, Special instructions, if any</th>
<th>Tests NOT possible due to sample type</th>
<th>Turn around time # (average)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor Fresh, unfixed</td>
<td>Frozen section/squash for intraoperative diagnosis (preliminary)</td>
<td>Saline Should reach lab within 30mins</td>
<td>Routine HPE, immunohistochemistry</td>
<td>15-20 mins</td>
</tr>
<tr>
<td>Tumor Formalin fixed</td>
<td>Routine histopathology (HPE) Immunohistochemistry</td>
<td>10% formalin</td>
<td>Frozen sections</td>
<td>4 days 7 days</td>
</tr>
<tr>
<td>Tumor Paraffin block</td>
<td>Review</td>
<td>NA</td>
<td>Frozen section</td>
<td>3 days (routine) 7 days (IHC)</td>
</tr>
<tr>
<td>Nerve biopsy Fixed</td>
<td>Routine histopathology, Myelin stain (Kpal) Immunohistochemistry*</td>
<td>Use ONLY 2% glutaraldehyde</td>
<td>-</td>
<td>4-7days* 7days</td>
</tr>
<tr>
<td>Semithin*</td>
<td>Use ONLY 2% glutaraldehyde NOT CIDEX</td>
<td></td>
<td></td>
<td>21days</td>
</tr>
<tr>
<td>Electron microscopy*</td>
<td>Use ONLY 2% glutaraldehyde</td>
<td></td>
<td></td>
<td>6 weeks</td>
</tr>
<tr>
<td>Nerve biopsy Paraffin block</td>
<td>Review</td>
<td>-</td>
<td>Kpal stain, semithin, EM Possible: Alternative myelin stains (Loyez)</td>
<td>4-7days* (routine) 7 days (IHC)</td>
</tr>
<tr>
<td>Skin Fixed</td>
<td>Routine HPE Electron microscopy</td>
<td>10% formalin/2% glutaraldehyde</td>
<td>-</td>
<td>4-7days* EM – 6 weeks</td>
</tr>
</tbody>
</table>

* in working days. Routine HPE - 4 days; Special stains/Immunohistochemistry - 7 days

### At a glance...

<table>
<thead>
<tr>
<th>Sample</th>
<th>Tests possible (dependant on fixative)</th>
<th>Appropriate fixative, Special instructions, if any</th>
<th>Tests NOT possible because of sample type fixative</th>
<th>Turn around time # (average)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Muscle Biopsy Fresh</td>
<td>Routine HPE, Enzyme Histochemistry, Immunohistochemistry Electron microscopy, Western Blot#, Enzyme studies#, genetics*</td>
<td>Saline soaked gauze Should reach lab within 30mins. <strong>Do not use too much saline</strong></td>
<td>-</td>
<td>4-7days* IHC - 4 weeks EM – 6 weeks</td>
</tr>
<tr>
<td>Muscle biopsy Fixed</td>
<td>Routine HPE Electron microscopy</td>
<td>10% formalin/2% glutaraldehyde</td>
<td>Enzyme Histochemistry Immunohistochemistry Western Blot Enzyme studies</td>
<td>4-7days* EM – 6 weeks</td>
</tr>
<tr>
<td>Muscle Paraffin block</td>
<td>Review</td>
<td>Nil</td>
<td>Enzyme histochemistry, semithin, EM not possible</td>
<td>3 days (routine)</td>
</tr>
<tr>
<td>Diagnostic Brain biopsy Fresh</td>
<td>Routine HPE, IHC, EM Frozen section for lipid, Culture (for bacterial/ TB/ fungal), PCR for viral / Genetics</td>
<td>Saline (for suspected infective/ neurodegenerative/metabolic etiology)</td>
<td>-</td>
<td>4-7days* (routine) 10 days (IHC) EM – 6 weeks</td>
</tr>
<tr>
<td>Brain biopsy fixed</td>
<td>Routine HPE Immunohistochemistry</td>
<td>10% formalin</td>
<td>Culture, special stains for lipid</td>
<td>4-7days* (routine) 10 days (IHC)</td>
</tr>
<tr>
<td>Liver biopsy Fixed</td>
<td>Storage disorder (lipid/glycogen)</td>
<td>10% formalin &amp;70% alcohol</td>
<td>Note: for lipid/glycogen demonstration, biopsy in 70% alcohol is MUST</td>
<td>4-7days* (routine)</td>
</tr>
</tbody>
</table>

* in working days. Routine HPE - 4 days; Special stains/Immunohistochemistry - 7 days
Sample transport requirements

TRIPLE PACKAGING SYSTEM FOR TRANSPORT OF PATHOLOGY SPECIMENS TO LAB

For purposes of transport of patient samples by road, all clinical samples, in accordance with UN guidelines are generally classified as Category B and assigned to UN3373 (Biological Substance, Category B) and should be packaged in accordance with UN packaging instructions PI650


The following procedures should be adopted for the transport of all specimens to the lab:

Step 1: Primary pack – should be water tight, leak proof, and labeled with patient detail and hospital name.

1. Place specimen with fixative in plastic container with tight screw cap. If there is more than one sample for the same patient, use separate plastic containers for each sample.
2. Label each container with patient name, details and site/biopsy

**Primary containers (screw cap, leak proof)**

- Not tight fitting / screw cap lid
- Inadequate volume of fixative
- Ideal: screw cap, leak proof, sufficient fixative
Step 2: Secondary pack – encloses primary pack(s), leak proof and water tight and protects the primary sample (see picture below)

1. Place the primary samples container in sealable plastic bag/ziplock cover/plastic container with screw cap.
2. Place large amounts of absorbent material (cotton/filter paper) between primary and secondary receptacle to absorb all spills, if any.

Step 3: Outer most pack: sealable & strong enough to protect contents from physical damage during transport.

1. Place secondary pack in a strong cardboard carton/ thermocol box (ideal)/plastic box. The smallest overall external dimension shall be 10x10 cm.
2. Place clinical datasheet/request form securely enclosed within a plastic cover to prevent it from getting soiled with leakage of sample contents.

LABELING OF OUTER PACKAGE
Biohazard Label for Category B as per UN recommendations should be affixed

TRIPLE PACKING SYSTEM MANDATORY FOR SAMPLE TRANSPORTATION TO NEUROPATHOLOGY LAB
PACKAGING FOR FRESH MUSCLE BIOPSIES (without fixative)

Step 1: Primary pack – should be water tight, leak proof, and labeled with patient detail and hospital name.

1. Place muscle biopsy wrapped in saline soaked gauze in zip lock cover and seal securely.
2. Label container with patient name, details and site/biopsy type.

Step 2: Secondary pack – encloses primary pack(s), leak proof and water tight

3. Place the zip lock cover in wide mouthed flask OR Thermocol box.
4. Place large quantities of ice (sprinkle cooking salt over ice) to maintain cold chain in transit.
5. Seal securely

Step 3: Outer most pack: sealable & strong enough to protect contents from physical damage during transport.

1. Place secondary pack in a strong cardboard carton.
2. Place clinical datasheet/request form enclosed in a separate plastic cover to prevent from getting wet or soiled.
3. Outer pack should contain name and address of consignee and sender’s name and address in CLEAR BOLD print.
4. Use arrows to indicate UP and DOWN on side of package.
5. Biohazard Label for Category B as per UN recommendations should be affixed.

ADDITIONAL INSTRUCTIONS FOR TRANSPORTATION OF FRESH MUSCLE

1. Samples SHOULD be hand delivered and MUST NOT be transported by courier as it should reach the lab within max of 12 hours of biopsy.
2. Samples should reach BEFORE 12 noon on working days (second Saturdays and Govt holidays, lab is open to received samples).
3. Samples should NOT be sent on Second Saturdays, Sundays or Govt holidays.
4. For muscle fixed in 10% formalin/2% glutaraldehyde, please follow the instructions as for “fixed” samples.
DEPARTMENT OF NEUROPATHOLOGY

CLINICIAN’S HANDBOOK

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