Neuromuscular Disorders 18 (2008) 90–96

www.elsevier.com/locate/nmd

Workshop report

147th ENMC International Workshop: Guideline on processing and evaluation of sural nerve biopsies, 15–17 December 2006, Naarden, The Netherlands

C. Sommer [a,*], S. Brandner [b], P.J. Dyck [c], L. Magy [d], S.I. Mellgren [e], M. Morbin [f], A. Schenone [g], E. Tan [h], J. Weis [i]

[a] *Department of Neurology, University of Würzburg, Würzburg, Josef-Schneider-Str. 11, 97080 Würzburg, Germany*
[b] *Department of Neuropathology, Institute of Neurology, London, United Kingdom*
[c] *Department of Neurology, Mayo Clinic and Mayo Foundation, Rochester, United States*
[d] *Department of Neurology, CHRU Dupuytren, Limoges, France*
[e] *Department of Neurology, University of Tromsö, Tromsö, Norway*
[f] *IRCCS Foundation - Neurological Institute "Carlo Besta", Milan, Italy*
[g] *Department of Neurosciences, Ophthalmology and Genetics, Universita degli Studi, Genova, Italy*
[h] *Department of Neurology, Hacettepe University, Ankara, Turkey*
[i] *Department of Neuropathology, University of Aachen, Aachen, Germany*

Received 3 August 2007

Keywords: Nerve biopsy; Peripheral neuropathy; Light microscopy; Electron microscopy; Diagnostic performance

1. Introduction

Fourteen clinicians and researchers (8 neurologists and 6 neuropathologists) from France, Germany, Italy, The Netherlands, Norway, Spain, Turkey, United Kingdom, and United States of America assembled in Naarden, The Netherlands, from 15 to 17 December, 2006, to participate in a workshop on the indications for nerve biopsy, the methods available for nerve workup, and on their diagnostic specificity and sensitivity. The results will lead to an evidence based guideline on processing and evaluation of nerve biopsies.

Peripheral neuropathies are a common and heterogeneous group of diseases. The differential diagnosis may be difficult because of the multitude of potential causes. Diagnostic algorithms for the investigation of peripheral neuropathies have been proposed, and the rate of successful identification of the underlying disease varies between investigators.

The evaluation of a nerve biopsy is often the final step in the diagnostic workup of neuropathies of unknown origin. While it is usually not considered necessary in neuropathies with causes which can be detected by other methods, like in diabetic neuropathies, or in Guillain-Barré Syndrome (GBS), it is the only method for the detection of some causes of neuropathies, like nonsystemic vasculitic neuropathy, and it can be very helpful in guiding further systemic diagnostic evaluation, like in amyloid neuropathies, and in some types of hereditary neuropathies, in which a straightforward genetic test is not yet available.

However, the value of nerve biopsy is still a matter of debate. While some authors have clearly shown a benefit for patient management from sural nerve biopsies, others have disagreed with this view. Obviously, the diagnostic value depends on the standards set for the evaluation of the biopsy, such that the questions of diagnostic value and standards for evaluation are interrelated.

If a diagnostic nerve biopsy is performed, most often the sural nerve is chosen. Sural nerve biopsy is an invasive procedure which leaves the patients with a sensory deficit,

* Corresponding author. Tel.: +49 931 201 23763; fax: +49 931 201 23697.
E-mail address: sommer@mail.uni-wuerzburg.de (C. Sommer).

0960-8966/$ - see front matter
doi:10.1016/j.nmd.2007.08.010

and, rarely, chronic pain. Therefore, the indication for a sural nerve biopsy should be considered carefully, and all measures should be taken to obtain optimal results for the benefit of the patients, if this procedure is performed. Since sural nerve biopsy cannot be easily repeated in the workup of peripheral neuropathies, failures due to lack of adequate standards should be avoided.

In clinical practice, one often encounters the situation that a sural nerve biopsy was performed in a patient, which in retrospect might not have been considered to be indicated because the cause of the neuropathy might have been detected by less invasive tests. It also happens that the indication was correct, but that the processing and evaluation are so poor that very little information can be gained from the biopsy. This dilemma was the motivation for performing research into the available evidence on the diagnostic value of nerve biopsies and the specific techniques used in their processing and evaluation.

There are no generally accepted guidelines on nerve biopsy processing and evaluation, and laboratories do not need to be certified to perform this procedure. Although there are some national guidelines, these are incomplete and not evidence based.

In preparation for the workshop, participants performed Medline researches on the subtopics discussed below and prepared evidence tables as a background for discussion in the workshop.

2. Indications for nerve biopsy

Angelo Schenone presented the evidence available on the indications for sural nerve biopsy, prepared in collaboration with Catherine Lacroix. The aim was to define the following patient groups:

1. Patients in whom sural nerve biopsy will be diagnostically helpful
2. Patients in whom sural nerve biopsy will have therapeutic implications
3. Patients who will definitely not benefit from sural nerve biopsy
4. Patients who are at risk for complications from sural nerve biopsy

Out of an extensive Medline research, 20 papers were identified to be of relevance to the subject. Out of the studies by Gabriel [1,2] with 50 and 355 patients, respectively, it could be concluded with grade IV evidence that sural nerve biopsy is helpful in inflammatory and dysimmune neuropathies, namely in vasculitis and chronic inflammatory demyelinating neuropathy (CIDP), also possibly in leprosy and in some forms of hereditary neuropathies. From the clinical perspective, sural nerve biopsy was more often helpful in patients with severe demyelinating, distal asymmetric, and multifocal types of neuropathy than in axonal and symmetric types. Also, nerve biopsy was more often diagnostic in acute and subacute than in chronic forms. Patient groups with therapeutic consequences were those with inflammatory/dysimmune neuropathies and with amyloid neuropathy. Toxic neuropathies were identified as a group where nerve biopsy did not contribute to finding the diagnosis, also metabolic neuropathies, with exceptions. In fact, focally swollen axons may be seen in patients with hexacarbon neuropathy.

Ten publications were identified dealing with complications from sural nerve biopsy, two of which were presented. The conclusions were that sural nerve biopsy is associated with prolonged sensory symptoms and sensory loss, that recovery occurs in all patients irrespective of diagnosis, and that residual sensory loss in diabetic and nondiabetic patients are comparable. In particular, the outcome was not worse in patients with vasculitis or diabetes. However, patients with diabetes and higher pre-biopsy sural nerve potentials and better glycemic control recovered better.

3. Methods of nerve biopsy and specimen processing and evaluation

On the subject of choice of nerve, Peter Dyck pointed out that the biopsied nerve is most likely to provide useful clinical information when the nerve to be biopsied is clinically affected and when an interstitial process is suspected. Usually a distal sensory nerve (i.e. the sural nerve) is biopsied. There are some rare indications for proximal biopsies, but the following rules should be observed: an expert MRI consultant, peripheral nerve surgeon and pathologist should be available. There should be unequivocal demonstration of a single MRI lesion (e.g. focal enlargement or enhancement) whose diagnosis will aid diagnosis and management. Benefits versus risks and side effects should be carefully assessed. The procedure should be carefully explained to the patient and agreement should be obtained.

Peter Dyck further pointed out that the procedures for nerve biopsies as well as for tissue processing and evaluation have been detailed in various textbook articles, e.g. [3]. However, to measure sensitivity, specificity, reproducibility, accuracy, meaningfulness, and monotonicity (measuring a consistent trend of change with time) of a method, a gold standard is needed. This gold standard is still lacking in the field of nerve biopsy evaluation. Therefore, a study was initiated by Caroline Klein, P. James B. Dyck, Christopher J. Klein, JaNean Engelstad, Peter C. O'Brien, and Peter J. Dyck, with the title "Masked and Independent Evaluations of Various Histologic Preparations". The aim of this now ongoing study is to compare different methods of workup of sural nerve biopsies from 100 patients. Comparisons will be performed between the diagnostic yield of teased fibers (100 strands systematically sampled), of paraffin sections stained with H&E, trichrome, and other stains, of semithin epoxy sections, including morphometry, of immunohistochemistry on paraffin sections (CD45, CD68, and others) and of transmission electron microscopy. An adequate number of nerves of healthy subjects will be prepared by similar methods.

4. Diagnostic usefulness of paraffin histology and plastic embedded sections

From an extensive literature search, Sebastian Brandner concluded that there are no data proving the superiority of one staining method over another in nerve biopsies. In the study of Deprez et al. [2], the contribution of nerve biopsy varied according to the neuropathological techniques used. Serial sections of frozen, paraffin-embedded, and resin-embedded material improved the sensitivity for interstitial pathology. A combined muscle and nerve biopsy increased sensitivity in the detection of vasculitis. Teasing of nerve fibers added critical information to other classical techniques in only 4/102 cases.

Dr. Brandner presented the algorithm of sural nerve evaluation as practiced in his laboratory at the Institute of Neurology, Queen Square. The value of paraffin histology and plastic embedded sections as presented by Dr. Brandner is summarized in Table 1.

Dr. Brandner suggested performing neurofilament immunohistochemistry, which labels axons of all sizes and gives a quick and relatively accurate estimate of axonal loss. He also recommended solochrome cyanine staining on longitudinal sections for better visualization of myelin, in particular when resin sections are not available. Congo red staining for amyloid and van Gieson Elastica staining for the evaluation of vessel walls were also considered useful. For the detection of inflammation, he advocated immunohistochemistry on paraffin sections using antibodies to UCLH1 (CD45RO, pan T cell, memory cells, and monocytes), CD3 (pan T cell), CD8 (T-suppressor/cytotoxic), CD20 (B cells), and optionally to CD4 (T-helper, inducer). CD68 immunohistochemistry for macrophages was also considered standard, to identify florid axonal or myelin degeneration.

Table 1
Value of paraffin histology and plastic embedded semithin sections

(*a*) *Value of paraffin histology with Haematoxylin & Eosin staining*
Very useful to evaluate
General appearance and quality of nerve biopsy Inflammation, in particular vasculitis
Digestion chambers, but not degeneration of axons per se
Others, such as tumor cells, sarcoidosis, giant cells
Moderately useful to evaluate
Axon density and myelination
Degeneration of axons
Endoneurial inflammation
Not useful to evaluate
Subtle axon loss
Patchy axon loss
Demyelination
Amyloid (unless there are big 'plaques')
(*b*)*Value of semithin plastic embedded sections*
Very useful to evaluate
Fiber (axon) density of myelinated fibers
Number and distribution of large and small myelinated fibers (semiquantitative assessment)
Onion bulbs
Regeneration clusters
Vessel pathology
Edema
Moderately useful to evaluate
Demyelination
Inflammation
Unmyelinated fibers
Macrophage density
Not useful to evaluate
Endoneurial inflammation
Amyloid (unless there are big 'plaques')
Diabetic changes (basement membranes)

5. Diagnostic usefulness of frozen sections and immunohistochemistry: inflammatory cells

Claudia Sommer performed a literature search trying to answer the following questions:

1. What is the diagnostic value of immunohistochemistry for cellular infiltrates?
2. Does immunohistochemistry for cellular infiltrates have a higher diagnostic yield than H&E stains?
3. What is the value of serial sections?
4. What is the value of macrophage or lymphocyte subtype markers?
5. Can the biopsy predict a treatment response?

There were no prospective studies available to answer these questions. Information was collected from retrospective analyses asking the question of diagnostic utility, from retrospective analyses asking a scientific question, and from individual clinical experience. In a retrospective study, Bosboom and colleagues [4] investigated the diagnostic value of sural nerve T cells in CIDP using biopsies from 23 patients with CIDP, 15 with other neuropathies, and 10 autopsy controls. They concluded that T cells were found in sural nerves of all CIDP patients as well as in all disease and normal controls. Only 6 CIDP patients had increased numbers and densities of T cells compared with patients with axonal neuropathy and controls.

Increased numbers and densities of sural nerve T cells in patients with CIDP were associated with female sex, a more severe disease course, worse outcome, highly elevated CSF protein level, and a larger sural nerve area. Eurelings et al. [5] investigated sural nerve biopsies from 25 patients with demyelinating neuropathy and monoclonal gammopathy. Increased sural nerve T cells were significantly associated with more progressive disease course and more pronounced weakness, IgG isotype, and malignancy. In a small study with 15 patients, Jann et al. [6] looked at the diagnostic value of sural nerve matrix metalloproteinase-9 (MMP-9) in diabetic patients with CIDP. MMP-9 immunohistochemistry was useful to detect CIDP in diabetic patients. Another study investigated the diagnostic value of macrophage distribution in sural nerve sections [7]. Whereas numbers of T cells and macrophages were not helpful in the distinction between CIDP and hereditary demyelinating neuropathies, clustering

of macrophages around endoneurial vessels could be found more often in CIDP and served as an easily detectable additional indication for an inflammatory neuropathy. A small number of further studies was identified, which asked a scientific question regarding particular markers (e.g. B7 costimulatory molecule, or specific chemokines), but these were not tested for their diagnostic performance. Dr. Sommer concluded that high numbers of inflammatory cells indicate inflammatory neuropathy, but that small numbers of inflammatory cells may be present in different kinds of neuropathy. The distribution of the inflammatory cells and special stains like for MMP-9 may aid in the diagnostic distinction. Immunohistochemistry is probably more sensitive in the detection of macrophages and T cells than H&E stains, but prospective studies are needed to clarify these issues.

6. Diagnostic usefulness of teased fiber studies

Svein Ivar Mellgren reported on the technique for teased fiber studies and showed examples of teased fiber pathology. The study of teased fibers allows evaluation of myelinated fibers and provides another window into the nerve biopsy than cross and length oriented sections in light and electron microscopy. For most purposes 100 fibers are analyzed, although it has not been formally shown which number is needed for reliable diagnostic evaluation. Pathological grades named A–H were classified by Dyck and colleagues [8] and control values were established. Pathologic findings using this grading system were described in patients with Sjögren's syndrome and with diabetes compared to controls. Dr. Mellgren offered the following arguments in favor of teased fiber studies: teased fiber studies (1) usually show abnormalities supportive of a neuropathy; (2) can be used for demonstration of fiber degeneration; (3) may show fibers in active axonal degeneration and their proportion; (4) may show evidence of regeneration; (5) visualize axonal atrophy and secondary segmental demyelination, axonal swellings, and tomacula; (6) can show changes suggestive of primary or secondary demyelination. The following arguments were offered against the use of teased fiber preparations: their usefulness is limited due to the age dependent amount of"pathology" especially in myelin in normal nerves; many centers consider nerve fiber teasing to be insufficiently informative to justify its cost in routine evaluation of sural nerve biopsies. A few authors studied the usefulness of teased fiber preparations formally. Among 24 patients who fulfilled the clinical criteria for CIDP, 14 met the AAN teased fiber criterion for demyelination, whereas 7 of these did not fulfill the electrodiagnostic criteria for demyelination. Three out of these 7 patients responded to treatment [9]. In 21 patients with CIDP, Bosboom and colleagues did not find teased fiber analysis useful to distinguish CIDP from chronic idiopathic axonal neuropathy [10]. In the 102 cases of Deprez et al. [2], in 4 cases fiber teasing provided contributive information altering the patient management.

7. Diagnostic usefulness of electron microscopy

Joachim Weis discussed the usefulness of electron microscopy (EM) in nerve biopsy evaluation. The relevant textbooks, including [11–15] extensively cover the issue of ultrastructural pathology of peripheral nerves. However, there are no class I–III studies using a blinded evaluation on the usefulness of electron microscopy in peripheral nerve biopsy evaluation thus far. Still, numerous case series and studies of single cases (class IV evidence) suggest that electron microscopy might be helpful in the diagnosis of several entities including

- CIDP, by virtue of demonstrating macrophage-associated demyelination.
- Neuropathy due to gammopathy, when associated with focal widening of myelin lamellae.
- Hereditary neuropathies displaying characteristic ultrastructural abnormalities including focally folded myelin in CMT1B caused by MPZ mutations and in CMT4B2 caused by mutation of the SBF2 gene. myelin outfoldings in CMT4B1, and a recently described autosomal recessive CMT characterized by exceedingly complex folding of myelin sheaths due to frabin/FGD4 [16], peculiar Schwann cell processes combined with basal lamina accumulation in CMT4C due to KIAA1985 gene mutation, as well as axonal neurofilament accumulation in CMT2E due to NEFL gene mutation.
- Hereditary diseases that affect both the PNS and the CNS and potentially other organs such as the leukodystrophies and CADASIL.
- Toxic neuropathies associated with characteristic inclusions exemplified by amiodarone neuropathy.

In addition, EM is the method of choice to visualize the unmyelinated nerve fibers in nerve biopsies. By all means, well-preserved and well-orientated tissue fixed in buffered glutaraldehyde or a similar solution is required to obtain meaningful results by EM analysis.

8. Diagnostic usefulness of special markers: immunoglobulin deposits

Michela Morbin presented the evidence for the value of demonstrating immunoglobulin deposits in sural nerve sections. No prospective studies addressing this point were identified. In most cases studies were designed to answer to other questions or to study pathogenetic mechanisms and the search for immunoglobulin was a side task.

Conditions in which immunoglobulin deposits in sural nerve have been found encompassed: paraproteinemic-dysglobulinemic neuropathy, diabetic neuropathy, paraneoplastic neuropathy, AIDP/GBS, CIDP, hereditary neuropathy, rheumatic disease, toxic neuropathy, HIV, hepatitis B, post-streptococcal infection.

Immunofluorescence or immunoperoxidase detection of immunoglobulin deposits is reported in various pathological processes. IgM perineurial deposits have been found even in normal nerves, and there were some problems with methodologies (background). The presence of IgM in a variety of neuropathological conditions suggests that such deposits may represent an unspecific process not only related to immune-mediate pathogenetic mechanisms. It has been suggested that IgM may be "entrapped" in the perineurium following an increase in permeability of the blood-nerve barrier, or as a consequence of abnormal function of thickened perineurial sheath.

Dr. Morbin reported on the usefulness of immunoglobulin stains in different etiological categories. Even if the detection of immunoglobulin deposits on nerve has no definite diagnostic value, its demonstration may be useful to characterize amyloid neuropathies in patients with monoclonal gammopathies. Moreover, most patients suffering from demyelinating neuropathy associated with monoclonal gammopathy of unknown significance (MGUS) with anti-MAG activity, deposition of IgM and the corresponding light chain were reported [17,18]. Detection of IgM deposits might even precede the detection of IgM gammopathy in serum [19]. The presence of immunoglobulin deposition in sural nerve seems to be significantly associated with severe outcome [20]. Thus, the demonstration of immunoglobulin deposits on sural nerve might endorse a more aggressive treatment.

9. Metalloproteinases (MMPs), RAGE, NFκB

Researching on the usefulness of immunohistochemistry, Ersin Tan identified 8 articles for MMPs, 4 for receptor for advanced glycation end products (RAGE) and nuclear factor κB (NFκB) combined, and one article for NFκB alone. In a study with 7 vasculitic and 6 noninflammatory neuropathies, immunostaining for MMP-1 distinguished between the groups [21]. MMP-9 and MMP-7 immunohistochemistry was positive in 5 GBS cases, but in none of 2 cases with hereditary neuropathy [22]. Immunoreactivity for MMP-2 and MMP-9 were present in 14 specimens with inflammatory neuropathies, but not in 4 with noninflammatory neuropathies [23]. A similar result was obtained in a follow-up study and for MMP-9 in a study with 12 patients with vasculitic neuropathy compared to 8 hereditary neuropathies. MMP-3 and -9 were present in 12 patients with systemic lupus erythematodes, but not in controls. MMP-9 immunostaining was also useful to diagnose CIDP in diabetic patients [6]. Immunoreactivity for TNF-α converting enzyme (TACE) a member of the A Disintegrin and Metalloproteinase (ADAM) family (ADAM 17) was seen in biopsies from 6 patients with GBS, but not from 2 patients with hereditary neuropathies [24]. Taking all studies together, Dr. Tan gave a strength B recommendation to use MMP-staining for the distinction of inflammatory and noninflammatory neuropathies.

Increased levels of RAGE were found in 16 cases of familial amyloidotic neuropathy compared to 4 controls [25]. RAGE immunoreactivity also distinguished vasculitic neuropathy ($n = 12$) from CMT ($n = 8$) and healthy controls ($n = 4$) [26] and diabetic neuropathy ($n = 10$) from controls ($n = 8$) [27]. NFκB immunoreactivity was higher in 12 cases of GBS and CIDP compared to 3 controls [28]. NFκB immunoreactivity was prominent in inflammatory neuropathies ($n = 8$) and FAP ($n = 4$), but not in hereditary neuropathies ($n = 4$) and controls ($n = 3$) [29]. Dr. Tan gave a strength B recommendation to use RAGE and NFκB immunostaining for the distinction of inflammatory and noninflammatory neuropathies.

10. Performance of sural nerve histology in comparison with peripheral blood and CSF

Laurent Magy reported on an extensive search in Medline, the Cochrane databases, and in personal files to determine the diagnostic performance of sural nerve histology in comparison with peripheral blood and CSF. He found class IV evidence for the usefulness of antiglycolipid antibodies in acute neuropathy syndromes. He also reported on class IV evidence for the use of sural nerve biopsy in selected cases of anti-MAG neuropathy with an IgM paraprotein. In contrast, for other paraproteinemic neuropathies, no recommendation could be made. In CANOMAD and POEMS syndrome, nerve histology may only be helpful in difficult cases. There was class VI evidence for the use of nerve biopsy in cryoglobulinemia. In the diagnosis of paraneoplastic neuropathies, in accordance with an EFNS task force [30], Dr. Magy concluded that nerve biopsy was usually not necessary, but might sometimes be helpful in distinguishing subacute sensory neuropathy from multiple mononeuropathy because of vasculitis. For borrelia-associated neuropathy, there was class IV evidence that nerve biopsy may be helpful in unusual situations. Whether nerve biopsy was useful in hepatitis-associated neuropathy remained unclear. Biopsy was considered of some value (class IV evidence) in asymmetric neuropathy in HIV infected patients. The question of whether nerve biopsy is of use in suspected CIDP with high or low CSF protein is discussed controversially. In lymphoma, nerve biopsy may help discover infiltrative neuropathy [31].

11. Conclusions

It was concluded that very little high quality evidence is available for the usefulness of specific methods in nerve biopsy workup and evaluation. This is in contrast to a large body of expert opinions and experience. An evidence based guideline on processing and evaluation of nerve biopsies will be compiled from the research done by the workshop participants. The following preliminary consensus was reached among the participants:

Indication for nerve biopsy:

- Nerve biopsies should be done for specific indications, for example to answer questions about diagnosis, classification, or prognosis.
- Nerve biopsy should not be done before adequate clinical, electrophysiological and laboratory characterization of the patients.
- The leading indication is the suspicion of an interstitial pathological process (e.g. vasculitis, inflammation, infection, amyloid deposition, lymphomatous infiltration, tumor).
- The patient should be properly instructed, and nerve biopsy should only be done with appropriate informed consent.

Requirements for the person evaluating a nerve biopsy:

- Biopsies should be read by professionals with adequate training and experience in reading and interpretation of nerve biopsies.
- Adequate clinical information should be provided and additional clinical information should be accessible on request.
- An interactive working relationship with the relevant disciplines involved is encouraged.
- The results should be discussed with clinicians taking care of the patient and regular nerve biopsy conferences are recommended.

Recommendation of specific procedures and stains:

Although consensus can be reached that certain procedures and stains should be mandatory (e.g. paraffin sections, semithin sections, immunohistochemistry for inflammatory cells), there is no formal evidence about the superiority of one method over another (e.g. whether demyelination can be better detected in teased fibers or in longitudinal semithin sections). A prospective study on these issues (Caroline Klein et al.) is underway. Details on the procedures recommended will be published in the final guideline. A preliminary recommendation is given in Table 2.

Table 2
Consensus on minimal requirements for processing of nerve biopsies

Paraffin or frozen sections	H&E stain, Congo red or thioflavin S, optionally Trichrome, myelin stain, EvG, iron stain, and others Immunohistochemistry for macrophages and T cells For optimal detection of inflammatory neuropathies, serial sections are recommended
Semithin sections	Toluidine blue, MBA-BF, or PPD
Immunohistochemistry	Although there is no formal proof (Class I–IV evidence) that IHC for cellular infiltrates is more efficient than H&E stain, experience shows that inflammatory cells are more easily detected and thus should be performed
Electron microscopy	Is useful under certain conditions Examples: detect decompacted myelin, focally folded myelin, detect axonal dystrophy, and other axonal changes, autonomic neuropathies, evaluation of unmyelinated axons, mitochondrial neuropathies

12. List of participants

S. Brandner (United Kingdom)
P.J. Dyck (USA)
Y. Harati (USA)
C. Lacroix (France)
M. Lammens (The Netherlands)
L. Magy (France)
S. I. Mellgren (Norway)
M. Morbin (Italy)
C. Navarro (Spain)
H. Powell (USA)
A. Schenone (Italy)
C. Sommer (Germany)
E. Tan (Turkey)
A. Urtizberea (ENMC)
J. Weis (Germany)

Acknowledgements

This workshop was made possible by the financial support of the European Neuromuscular Centre (ENMC) and its main sponsors and associated members:

– Association Française contre les Myopathies (France)
– Deutsche Gesellschaft für Muskelkranke (Germany)
– Telethon Foundation (Italy)
– Muscular Dystrophy Campaign (UK)
– Muskelsvindfonden (Denmark)
– Prinses Beatrix Fonds (The Netherlands)
– Schweizerische Stiftung für die Erforschung der Muskelkrankheiten (Switzerland)
– Österreichische Muskelforschung (Austria)
– Vereniging Spierziekten Nederland (The Netherlands) and ENMC associate member:
– Asociacion Española contra las Enfermedades Neuromusculares (Spain)

Additional support was provided by the Peripheral Nerve Society.

References

[1] Gabriel CM, Howard R, Kinsella N, Lucas S, McColl I, Saldanha G, et al. Prospective study of the usefulness of sural nerve biopsy. J Neurol Neurosurg Psychiatry 2000;69:442–6.
[2] Deprez M, de Groote CC, Gollogly L, Reznik M, Martin JJ. Clinical and neuropathological parameters affecting the diagnostic yield of nerve biopsy. Neuromuscul Disord 2000;10:92–8.
[3] Dyck PJ, Dyck PJB, Engelstad JN. Pathological alterations of nerves. In: Dyck PJ, Thomas PK, editors. Peripheral neuropathy. Philadelphia: Elsevier Saunders; 2005. p. 733–29.
[4] Bosboom WM, Van den Berg LH, De Boer L, Van Son MJ, Veldman H, Franssen H, et al. The diagnostic value of sural nerve T cells in

chronic inflammatory demyelinating polyneuropathy. Neurology 1999;53:837–45.
[5] Eurelings M, van den Berg LH, Wokke JH, Franssen H, Vrancken AF, Notermans NC. Increase of sural nerve T cells in progressive axonal polyneuropathy and monoclonal gammopathy. Neurology 2003;61:707–9.
[6] Jann S, Bramerio MA, Beretta S, Koch S, Defanti CA, Toyka KV, et al. Diagnostic value of sural nerve matrix metalloproteinase-9 in diabetic patients with CIDP. Neurology 2003;61:1607–10.
[7] Sommer C, Koch S, Lammens M, Gabreels-Festen A, Stoll G, Toyka KV. Macrophage clustering as a diagnostic marker in sural nerve biopsies of patients with CIDP. Neurology 2005;65:1924–9.
[8] Dyck PJ, Johnson WJ, Lambert EH, O'Brien PC. Segmental demyelination secondary to axonal degeneration in uremic neuropathy. Mayo Clin Proc 1971;46:400–31.
[9] Haq RU, Fries TJ, Pendlebury WW, Kenny MJ, Badger GJ, Tandan R. Chronic inflammatory demyelinating polyradiculoneuropathy: a study of proposed electrodiagnostic and histologic criteria. Arch Neurol 2000;57:1745–50.
[10] Bosboom WM, van den Berg LH, Franssen H, Giesbergen PC, Flach HZ, van Putten AM, et al. Diagnostic value of sural nerve demyelination in chronic inflammatory demyelinating polyneuropathy. Brain 2001;124:2427–38.
[11] Dyck PJ, Thomas PK. Peripheral neuropathy. 4th ed. Philadelphia: Elsevier; 2005.
[12] King R. Atlas of peripheral nerve pathology. London: Arnold; 1999.
[13] Schröder JM. Pathologie peripherer Nerven. Berlin: Springer-Verlag; 1999.
[14] Midroni G, Bilbao JM. Biopsy diagnosis of peripheral neuropathy. Boston: Butterworth-Heinemann; 1995.
[15] Vital C, Vallat JM. Ultrastructural study of the human diseased peripheral nerve. New York: Elsevier; 1987.
[16] Stendel C, Roos A, Deconinck T, Pereira J, Castagner F, Niemann A, et al. Peripheral nerve demyelination caused by a mutant rho GTPase guanine nucleotide exchange factor, Frabin/FGD4. Am J Hum Genet 2007;81:158–64.
[17] Yeung KB, Thomas PK, King RH, Waddy H, Will RG, Hughes RA, et al. The clinical spectrum of peripheral neuropathies associated with benign monoclonal IgM, IgG and IgA paraproteinaemia. Comparative clinical, immunological and nerve biopsy findings. J Neurol 1991;238:383–91.
[18] Ritz MF, Erne B, Ferracin F, Vital A, Vital C, Steck AJ. Anti-MAG IgM penetration into myelinated fibers correlates with the extent of myelin widening. Muscle Nerve 1999;22:1030–7.
[19] Gabriel JM, Erne B, Bernasconi L, Tosi C, Probst A, Landmann L, et al. Confocal microscopic localization of anti-myelin-associated glycoprotein autoantibodies in a patient with peripheral neuropathy initially lacking a detectable IgM gammopathy. Acta Neuropathol (Berl) 1998;95:540–6.
[20] Eurelings M, Moons KG, Notermans NC, Sasker LD, De Jager AE, Wintzen AR, et al. Neuropathy and IgM M-proteins: prognostic value of antibodies to MAG, SGPG, and sulfatide. Neurology 2001;56:228–33.
[21] Satoi H, Oka N, Kawasaki T, Miyamoto K, Akiguchi I, Kimura J. Mechanisms of tissue injury in vasculitic neuropathies. Neurology 1998;50:492–6.
[22] Kieseier BC, Clements JM, Pischel HB, Wells GM, Miller K, Gearing AJ, et al. Matrix metalloproteinases MMP-9 and MMP-7 are expressed in experimental autoimmune neuritis and the Guillain-Barre syndrome. Ann Neurol 1998;43:427–34.
[23] Leppert D, Hughes P, Huber S, Erne B, Grygar C, Said G, et al. Matrix metalloproteinase upregulation in chronic inflammatory demyelinating polyneuropathy and nonsystemic vasculitic neuropathy. Neurology 1999;53:62–70.
[24] Kurz M, Pischel H, Hartung HP, Kieseier BC. Tumor necrosis factor-alpha-converting enzyme is expressed in the inflamed peripheral nervous system. J Peripher Nerv Syst 2005;10:311–8.
[25] Sousa MM, Du Yan S, Fernandes R, Guimaraes A, Stern D, Saraiva MJ. Familial amyloid polyneuropathy: receptor for advanced glycation end products-dependent triggering of neuronal inflammatory and apoptotic pathways. J Neurosci 2001;21:7576–86.
[26] Haslbeck KM, Bierhaus A, Erwin S, Kirchner A, Nawroth P, Schlotzer U, et al. Receptor for advanced glycation endproduct (RAGE)-mediated nuclear factor-kappaB activation in vasculitic neuropathy. Muscle Nerve 2004;29:853–60.
[27] Bierhaus A, Haslbeck KM, Humpert PM, Liliensiek B, Dehmer T, Morcos M, et al. Loss of pain perception in diabetes is dependent on a receptor of the immunoglobulin superfamily. J Clin Invest 2004;114:1741–51.
[28] Andorfer B, Kieseier BC, Mathey E, Armati P, Pollard J, Oka N, et al. Expression and distribution of transcription factor NF-kappaB and inhibitor IkappaB in the inflamed peripheral nervous system. J Neuroimmunol 2001;116:226–32.
[29] Mazzeo A, Aguennouz M, Messina C, Vita G. Immunolocalization and activation of transcription factor nuclear factor kappa B in dysimmune neuropathies and familial amyloidotic polyneuropathy. Arch Neurol 2004;61:1097–102.
[30] Vedeler CA, Antoine JC, Giometto B, Graus F, Grisold W, Hart IK, et al. Management of paraneoplastic neurological syndromes: report of an EFNS task force. Eur J Neurol 2006;13:682–90.
[31] Vallat JM, De MH, Bordessoule D, Jauberteau MO, Tabaraud F, Gelot A, Vallat AV. Non-Hodgkin malignant lymphomas and peripheral neuropathies – 13 cases. Brain 1995;118:1233–45.