Porphyric Neuropathy: An Ultrastructural and Quantitative Case Study

P.S. THORNER, J.M. BILBAO, A.A.F. SIMA and S. BRIGGS

SUMMARY: We report a case of acute neuropathy in a 46 year old female with porphyria variegata. Histologic, electron microscopic, and quantitative examinations of peripheral nerves were performed at onset of the neuropathy and at autopsy. The results revealed severe qualitative and quantitative changes in myelinated and unmyelinated fibers showing features indicative of an axonopathy with a distribution in keeping with a dying-back phenomenon.

RÉSUMÉ: Nous rapportons un cas fatal de neuropathie aigue chez une femme de 46 ans avec porphyrie variegata. Des examens histologiques, de microscopie électronique et quantitatifs des nerfs périphériques furent obtenus au début de la neuropathie et à l'autopsie. On montra ainsi des changements de structure importants et quantitatifs au niveau des fibres myélinisées et non myélinisées. Ces changements indiquent une axonopathie compatible avec le phénomène du "dying-back".

INTRODUCTION

The hepatic porphyrias are characterized by acute attacks of neurological dysfunction which often result in the patient's death (Goldberg, 1959; Eales & Linder, 1962; Eales, 1963; Bloomer, 1976). The clinical presentation includes colicky abdominal pain, neuropsychiatric disorders, and peripheral neuropathy. After a single attack the neuropathy may be reversible whereas repeated or prolonged attacks cause irreversible peripheral nerve damage (Berg, 1945; Flügel and Kruschky, 1977; Wochnik-Dyjas, et al, 1978).

The time course of the neuropathy may be rapidly progressive, reaching flaccid paralysis in days (Goldberg, 1959), or proceed at a slower pace evolving over months. There are conflicting reports regarding the basic pathology of porphyric neuropathy. Early authors (Denny-Brown & Sciarra, 1945; Gibson & Goldberg, 1956; Campbell, 1963), supported segmental demyelination as the primary process whereas recent investigators like Cavanagh and co-workers (Cavanagh & Mellick, 1965; Cavanagh & Ridley, 1967; Schoental & Cavanagh, 1977), and others (Sweeney, et al, 1970; Tschudy, et al, 1975), suggest that axonal degeneration is the basic structural change in porphyric neuropathy.

This report describes the qualitative and quantitative changes in a sural nerve biopsy and in post mortem material obtained from the tibial nerve, ventral and dorsal sacral roots.

CASE HISTORY

The patient was a 46 year old Caucasian woman with a past history of depression (1973, 1977) and abdominal pain with diarrhea in 1978. She was on no medications and denied any photosensitivity. In August 1979, she developed protracted vomiting, which was attributed to cholecystitis, and a cholecystectomy was performed. Post-operatively the vomiting continued and over the next three weeks, the patient's neurological status deteriorated. She remained alert and oriented but developed a flat affect and recent memory loss. She suffered overflow incontinence and constipation. Cranial nerves were normal. She developed an ascending motor weakness with 4/5 power in all muscle groups of the upper extremities and 3/5 power in all groups of the lower extremities. Tendon reflexes were absent at the knees, ankles and wrists. The biceps tendon reflexes were 1+. Decreased sensation to light touch only was noted in both hands and both feet.

A diagnosis of Guillain-Barré syndrome was considered and electrophysiologic examinations were performed by Dr. Henry Berry on September 20, 1979. The conduction velocity in the left lateral popliteal nerve was 38.0 m/sec, the lower limit of normal being 40 m/sec. The conduction velocities of the left superficial peroneal nerve and the left sural nerve were absent. Testing of the right frontalis muscle revealed fibrillation potentials and no motor units under voluntary control. The left tibialis anterior muscle showed no fibrillation or fasciculation and a few motor units under voluntary control. The right peroneal nerve and the left sural nerve were absent. Testing of the right frontalis muscle revealed fibrillation potentials and no motor units under voluntary control. The left tibialis anterior muscle showed no fibrillation or fasciculation and a few motor units under voluntary control. The upper limbs could not be examined.

A sural nerve biopsy was performed on September 26, 1979.

Four weeks later further neurological deterioration had occurred. The patient was disoriented to time and place, unable to perform calculations, and demonstrated loss of remote memory, judgement and insight. Cranial nerves were normal. Her muscle bulk had greatly decreased and power testing revealed 1/5 power in all muscle groups of the legs and 2/5 power in the arms.

A stocking type loss to pin-prick and touch was noted extending to above the knees.

From St. Michael's Hospital, Toronto General Hospital and The University of Toronto, Canada.

Requests for reprints to Dr. A.A.F. Sima, Division of Neuropathology, Department of Pathology, Banting Institute, 100 College Street, Toronto, Ontario, Canada MSG 1A5.

Vol. 8 No. 4

NOVEMBER 1981 — 281
CSF examination, CT scan of the head and a myelogram were all normal. A
diagnosis of porphyria variegata was considered and confirmed by laboratory
studies (Table 1).

Three distant female relatives had experi­
enced symptoms suggestive of porphyria
but there was no biochemical confirmation
of the diagnosis.

During her hospital course the patient
suffered three bouts of aspiration pneu­
monia which were treated with broad
spectrum antibiotics. In late November she
suffered three bouts of aspiration pneu­
monia which were treated with broad
spectrum antibiotics. In late November she
developed increasing agitation and sys­
temic candidiasis. She died on November

At autopsy systemic candidiasis involv­
ing the kidneys, myocardium, brain and
the spinal cord was noted. Bilateral
aspiration pneumonia was seen. The rele­
vant neuropathology is described in
detail.

MATERIAL AND METHODS

The sural nerve obtained by biopsy, and
ventral and dorsal S1 roots and
tibial nerve dissected at post-mortem
examination, 7 hours after death, were
fixed in cacaodylate buffered 2% glu­
taraldehyde, post-fixed in Millonig­
buffered 1% osmium tetroxide, dehy­
drated and embedded in Epon-Araldit.
Cross and longitudinal 1 micron thick
sections were stained with toluidine
blue for light microscopy. Ultra-thin
sections were stained with uranyl
acetate and lead citrate and examined
under the light microscope. In addition, spinal root ganglia, spinal
cord and skeletal muscle were fixed in
neutral formaldehyde and examined
under the light microscope.

Caliber-spectra and fiber densities
of myelinated fibers were calculated
from photomicrographs at a final
magnification of 1000. In each nerve
all myelinated fibers of one randomly
chosen fascicle were counted. Caliber­
spectra and fiber densities of un­
myelinated fibers were calculated from
unselected electron micrographs with
a final magnification of 15,000. The
axon-myelin ratios were examined in
randomly chosen fibers, which showed
no axon or myelin destruction, at a
final magnification of 15,000, using the
methods of Robertson and Sima
(1980). Linear regressions for the
relationship between the natural log.
of axis cylinder area and myelin
thickness as measured by the number
of lamellae were calculated (cf. Dyck et

Control nerves were obtained 8
hours post-mortem from a 52 year old
woman who died without neurological
disease. The morphometric measure­
ments were performed on a Hewlett­
Packard 9874A Digitizer interfaced
with a Hewlett-Packard 9825A Com­
puter.

RESULTS

Structural Changes:

Sural Nerve Biopsy: Light micro­
scopic examination of thick sections
revealed a marked reduction in the
number of large myelinated fibers
(Fig. 1). Electron microscopy showed
extensive axonal degenerative changes
consisting of dissolution of the axo­
plasmic organelles, granular degenera­
tion of the axoplasm and ingrowth of
the inner Schwann cell lip (Fig. 2),
sometimes forming honeycomb struc­
tures at the axon-Schwann cell inter­
face. Many of the affected fibers
showed well preserved myelin sheaths.
Other fibers revealed a later stage of
axonal degeneration, namely complete
disintegration of the axon (Fig. 3),
collapse and fragmentation of the
myelin and the development of de­
nervated Schwann cells (Fig. 3). Some
of the smaller myelinated fibers were
uninvolved. Unmyelinated fibers show­
ed axonal fragmentation and retrac­
tion of Schwann cell cytoplasm leaving
axons directly apposed to basement
lamina (Fig. 4). Large unmyelinated
fibers frequently demonstrated a se­
paration of neurofilaments and tubules
with the neurofilaments in a central
position (Fig. 5).

Tibial Nerve: Light microscopic ex­
amination of thick sections showed
depletion of large myelinated fibers
and a large number of free foamy
macrophages in the endoneurium (Fig.
6). In the electron microscope small­
sized myelinated fibers showed mark­
ed axonal atrophy. Others contained
accumulation of membranous lipid
structures in the axons. Clusters of
regenerating units consisting of axons
with thin myelin sheaths and duplic­
tated basement membranes were oc­
casionally found. The changes in the
unmyelinated fibers were similar to
those described in the sural nerve. In
particular, there was a large number of
single axon-Schwann cell units with
pronounced Schwann cell banding.
Occasionally unmyelinated fibers con­
tained dense accumulations of misdi­
rected 10nm neurofilaments which
caused axonal distention (Fig. 7).

Ventral and Dorsal roots: Changes
in the spinal roots were less severe than
in the tibial and sural nerves. Many
myelinated fibers appeared normal,
whereas others showed complex mem­

TABLE 1

<table>
<thead>
<tr>
<th>BLOOD</th>
<th>URINE</th>
<th>STOOL</th>
</tr>
</thead>
<tbody>
<tr>
<td>δ-ALA</td>
<td>0.69 mg% (N &lt; 0.30)</td>
<td>-</td>
</tr>
<tr>
<td>PORPHOBILINOGEN</td>
<td>POSITIVE (N = negative)</td>
<td>POSTIVE (N = negative)</td>
</tr>
<tr>
<td>UROPORPHYRINS</td>
<td>250 mg/d (N &lt; 30)</td>
<td>24mg/ day (N = 0)</td>
</tr>
<tr>
<td>COPROPORPHYRINS</td>
<td>803 μg/d (N=75-275)</td>
<td>265mg/ day (N=50-250μg/day)</td>
</tr>
<tr>
<td>PROTOPORPHYRINS</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
Figure 1 — Toluidine-blue stained plastic section of the sural nerve showing loss of myelinated fibers and denervated Schwann cell units some of which contain myelin debris. Mag. 750 x.

Figure 2 — Myelinated fiber from the sural nerve showing ingrowth of the adaxonal Schwann cell cytoplasm into the axon (arrow). Note granular degeneration of the axonplasm. Mag. 7480.

Figure 3 — Sural nerve showing myelinated fiber with complete disintegration of its axon (arrow). Another fiber (arrow-head) shows denervation and delamination of the myelin-sheath. Mag. 3630 x.

Figure 4 — Unmyelinated fibers of the sural nerve. One fiber shows fragmentation of the axoplasm (arrow). Other fibers are partly denuded from the ensheathing Schwann cell (arrow-heads). Mag. 33150 x.

branous Schwann cell processes invading the axons, and accumulation of organelle debris in the axons. Axonal atrophy was marked, particularly in large sized fibers. In addition, the ventral root contained numerous regenerating units. (Fig. 8).

QUANTITATIVE CHANGES

Caliber Spectra:

Myelinated fibers: The tibial and the sural nerves and the unifascicular spinal roots showed marked shifts of their caliber-spectra toward thinner diameters (Fig. 9). These shifts were more marked in the tibial and sural nerves and less marked in the ventral and dorsal roots. The fiber densities were decreased in the sural and tibial nerves, whereas they were increased in the spinal roots, particularly in the...
ventral root. The total fiber number showed an absolute decrease in the dorsal root and a slight increase in the ventral root (Table 2).

*Unmyelinated fibers:* Unmyelinated fiber sizes were shifted toward thinner diameters in both the tibial and the sural nerves. The tibial nerve showed a small population (approximately 2.5%) of unmyelinated fibers with larger diameters than were found in the control nerve (Fig. 9). These fibers represent in part those exhibiting axonal swelling due to dense accumulations of neurofilaments. The densities of unmyelinated fibers per area unit was markedly increased in the tibial nerve and marginally increased in the sural nerve (Table 2).

*Axon-myelin ratio:* Both the ventral root and the tibial nerve showed significantly decreased ratios between the axonal area and the number of myelin lamellae (Fig. 10), indicating severe axonal atrophy.

*Light microscopic examination:* Dorsal root ganglia showed mild loss of neurons. The ventral horns of the

---

**Figure 5** — Unmyelinated fiber of the sural nerve showing central displacement of neurofilaments and peripheral localization of neurotubules. Mag. 27950 x.

**Figure 6** — Toluidine-blue stained plastic section of the tibial nerve. There is depletion of large myelinated fibers and several free foamy macrophages. Mag. 750 x.

**Figure 7** — Tibial nerve showing enlarged unmyelinated axon packed with neurofilaments (arrow-heads). The diameter measured 3.5 microns. The parent Schwann cell contains in addition normal-sized axons (arrows). Mag. 14300 x.

**Figure 8** — Ventral root showing a cluster of small relatively thinly myelinated fibers. Each individual Schwann cell is surrounded by a basement membrane. In addition a redundant basement membrane can be seen surrounding the whole group of fibers (arrows). Mag. 5325.
spinal cord showed moderate neuronal loss and chromatolysis of preserved neurons. In skeletal muscle, grouped fiber atrophy was seen.

**DISCUSSION**

The present study has demonstrated severe qualitative and quantitative changes in myelinated and unmyelinated nerve fibers in a patient with porphyric neuropathy. The sural nerve biopsy obtained during the early course of the disease revealed acute degenerative changes such as axonal sequestration by the inner Schwann cell lip and granular degeneration of the axoplasm in myelinated axons. These changes were accompanied by only minor structural changes of the Schwann cell and myelin sheath and are therefore indicative of a primary axonal involvement. More severe changes consisted of complete axonal disintegration and Wallerian degeneration. The caliber-spectrum analysis demonstrated that large myelinated fibers were especially involved in the disease process. In contrast, Anzil and Dozic (1978), found small-sized fibers to be mainly involved in porphyric neuropathy.

The examination of the tibial nerve 9 weeks later revealed less acute degenerative changes. There was marked axonal atrophy, (objectively demonstrated by the axon-myelin ratio), numerous free foamy macrophages and a large number of regenerating units, indicating a more long-standing process. In the spinal roots, the axonal degenerative and atrophic changes were less obvious, although the ventral root revealed marked regenerative activity. This was also reflected by 1) a slight absolute increase in the number of myelinated fibers, presumably representing re-myelination of sprouting fibers, and 2) a significantly increased fiber density of the ventral root, since more small remyelinating fibers can be harbored per area unit. The presence of oc-casional degenerating neurons in the spinal ganglia and the ventral horns suggests that the disease process had progressed to the cell somata.

The qualitative changes and the topographic difference in severity of quantitative changes in the peripheral nerves is suggestive of a primary axonopathy of dying-back type. (Schoental & Cavanagh, 1977; Spencer & Schaumburg, 1977; Cavanagh, 1979; Sima, et al, 1981). Early investigators (Denny-Brown & Scirra, 1945; Gibson & Goldberg, 1956; Campbell, 1963) have suggested that demyelination is the primary disease process in porphyric neuropathy. In the present study demyelination was certainly a prominent pathologic feature, although this must be interpreted as being secondary to axonal degeneration, based on the results of more recent investigative techniques used here such as quantitative morphology and ultra-structure.

One might however argue that the axonal atrophy could possibly be due to disuse during the patient's hospitalization (Eisen et al, 1973). However this seems unlikely because of the sudden onset of the patient's neuromuscular complaints and the rapid progression to a flaccid areflexic quadriplegia.

The structural changes reported in the present case show similarities with those recently reported in mice after the injection of a synthetic porphyrin, tetraphenylporphinesulfonate (Sima, et al, 1981). In this experimental model, peripheral motor nerves exhibited progressive axonal degenerative changes with secondary demyelination, evolving from the motor nerve terminals (Felix & Sima, 1981), to the ventral roots (Sima, et al, 1981).

Qualitative and quantitative changes of unmyelinated fibers in porphyric neuropathy have not previously been reported, although the clinical picture of acute porphyria suggests that this fiber population is most likely involved in the disease process (Tschudy, et al, 1975; Becker & Kramer, 1977). In the present case, unmyelinated fibers showed axonal degeneration with sequestration of axonal material and marked atrophy. Similar structural changes of unmyelinated fibers have

**TABLE 2**

<table>
<thead>
<tr>
<th></th>
<th>MYELINATED FIBERS</th>
<th>UNMYELINATED FIBERS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>FIBER NUMBER/FASCICLE</td>
<td>FIBER DENSITY (FIBERS/MM²)</td>
</tr>
<tr>
<td>VENTRAL ROOT</td>
<td>P 4436</td>
<td>14,786</td>
</tr>
<tr>
<td>C 3655</td>
<td>4,641</td>
<td></td>
</tr>
<tr>
<td>DORSAL ROOT</td>
<td>P 4502</td>
<td>9,829</td>
</tr>
<tr>
<td>C 9741</td>
<td>8,776</td>
<td></td>
</tr>
<tr>
<td>TIBIAL NERVE</td>
<td>P 885</td>
<td>5,205</td>
</tr>
<tr>
<td>C 2384</td>
<td>6,425</td>
<td></td>
</tr>
<tr>
<td>SURAL NERVE</td>
<td>P 409</td>
<td>2,133</td>
</tr>
<tr>
<td>C 1807</td>
<td>6,146</td>
<td></td>
</tr>
</tbody>
</table>
previously been reported in experi­mental diabetes (Sima & Robertson, 1979; Sima, 1980). Many unmyelin­ated fibers were found to be denuded from the ensheathing Schwann cells, a finding that has been reported in painful diabetic neuropathy (Brown, et al, 1976), and in different models of experimental diabetic neuropathy (Sima & Robertson, 1979; Sima, 1980). An interesting observation was a small population of unmyelinated fibers that exhibited axonal swelling due to dense accumulations of neuro­filaments. This finding is similar to what is encountered in myelinated fibers in the experimental model referred to previously (Sima, et al, 1981). This observation may indicate that disturbed axonal transport could be of pathogenetic significance in the development of porphyric neuropathy (Schoental & Cavanagh, 1977; Cava­nagh, 1979; Felix & Sima, 1981; Sima, et al, 1981).

Although the mechanism(s) respon­sible for porphyric neuropathy is (are) poorly understood, it is generally felt that the precursors to porphyrins, delta-ALA and/or PBG are respon­sible for the neuropathy (Kramer, et al, 1973; Shanley, et al, 1975; Brodie, et al, 1976; Sima, et al, 1981), since these substances are elevated only in those porphyrias which show neurological manifestations. It has been suggested that these substances may inhibit enzymes and cofactors (Cavanagh &

![Porphyric Neuropathy](https://www.cambridge.org/core/terms).

In summary, the present study has demonstrated an axonal neuropathy affecting distal nerves more severely than proximal ones in a patient with porphyria variegata. Furthermore, severe structural and quantitative changes of unmyelinated fibers have been demonstrated for the first time in porphyric neuropathy.

ACKNOWLEDGEMENTS
The authors want to thank Ms Ann Catherine Lorusso and Mrs. Kazuko Hay for skillful technical assistance. This investigation was supported in part by grants from the Canadian Medical Research Council (MA-7117) and the Canadian Diabetic Association.

REFERENCES


