Biochimica et Biophysica Acta xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbadis

Review

Human NCL Neuropathology ☆

Josefine Radke [a,1], Werner Stenzel [a,2], Hans H. Goebel [a,b,*]

[a] Department of Neuropathology, Charité - Universitätsmedizin Berlin, Charitéplatz 1 | Virchowweg 15, D-10117 Berlin, Germany
[b] Department of Neuropathology, University Medicine of the Johannes Gutenberg University, Mainz, Germany

ARTICLE INFO

Article history:
Received 10 March 2015
Received in revised form 7 May 2015
Accepted 11 May 2015
Available online xxxx

Keywords:
Neuronal ceroid lipofuscinoses
CLN
Lipopigments
Fingerprint profiles
Lysosome
SCMAS
GROD
Ultrastructure

ABSTRACT

The neuronal ceroid lipofuscinoses (NCL) currently encompass fourteen genetically different forms, CLN1 to CLN14, but are all morphologically marked by loss of nerve cells, particularly in the cerebral and cerebellar cortices, and the cerebral and extracerebral formation of lipopigments. These lipopigments show distinct ultrastructural patterns, i.e., granular, curvilinear/rectilinear and fingerprint profiles. They contain — although to a different degree among the different CLN forms — subunit C of ATP synthase, saposins A and D, and beta-amyloid proteins. Extracerebral pathology, apart from lipopigment formation, which provides diagnostic information, is scant or non-existent. The retina undergoes atrophy in all childhood forms. While many new data and findings have been obtained by immunohistochemistry in mouse and other animal models, similar findings in human NCL are largely missing, thus recommending respective studies of archived brain tissues. The newly described NCL forms, i.e., CLN 10 to CLN 14, also require further studies to provide complete neuropathology. This article is part of a Special Issue entitled: "Current Research on the Neuronal Ceroid Lipofuscinoses (Batten Disease)," edited by Romina Kohan, Susan L. Cotman and Sara E. Mole.

1. Introduction

The neuronal ceroid lipofuscinoses (NCL) are diagnosed by mutations, but defined by morphological features — though diagnosed by morphology when genetics are not available. The morphological criteria entail loss of neurons, especially in the cerebral and cerebellar cortices and accumulation of lipopigments within cells. The former may, at least partially, give rise to clinical symptoms and the latter to diagnosis. The relationship between these two features, i.e., loss of neurons and accretion of lipopigments (Figs. 1 and 2), is still insufficiently known. Distribution and severity of cellular loss will best be appreciated by autopsy which, however, will hardly inform about the dynamics of the disease. As lipopigments accrue in almost every cell type and organ, i.e., the central and peripheral nervous systems and extracerebral tissues, the biopsy of easily accessible organs, e.g., blood lymphocytes, skin, conjunctiva, and rectum, have readily been employed in the diagnostic armamentarium. The main diagnostic tool for biopsies is the electron microscope. Complete description of an NCL and its full nosological ascertainment may be based on both biopsy and autopsy in an individual patient and individual form of NCL, of which so far 14 genetically identified forms exist, CLN1 to CLN14. As the morphologies of autopsy and biopsy are overlapping, the complete pathology is best illustrated by discussing the various levels of visual recognition, from the naked eye to the light and electron microscopes.

2. Gross pathology

Macroscopic pathology is confined to the central nervous system, largely to the cerebrum and the cerebellum, whereas brainstem and spinal cord appear rather unremarkable.

Long-term loss of nerve cells and their processes results in atrophy (Fig. 1A), especially of the cerebral cortex and the cerebellum, together with widening of the ventricles and the subarachnoid space. Looking at the mantle of the brain, the gyri are reduced in thickness, the sulci are gaping and the leptomeninges appear thickened, more severely in the occipital lobes than the frontal ones. The smooth glistening surface of the sectioned cerebral and cerebellar cortices may be replaced by a gritty, occasionally spongy appearance, due to excessive loss of nerve cells. The earlier neuronal death sets in, the more severe is atrophy, being farthest advanced in the infantile type and least in the adult type. Reduced brain weights in the individual CLN forms are given in Table 1.

However, a protracted course, for instance in CLN3, may further advance the atrophy. In addition, by longstanding good care, with the vital organs longer intact and the body in less severe decay, the brain continues to shrink to further atrophy. Massive loss of neurons in the

☆ This article is part of a Special Issue entitled: "Current Research on the Neuronal Ceroid Lipofuscinoses (Batten Disease)," edited by Romina Kohan, Susan L. Cotman and Sara E. Mole.

* Corresponding author at: Department of Neuropathology, Charité - Universitätsmedizin Berlin, Charitéplatz 1 | Virchowweg 15, D-10117 Berlin, Germany. Tel.: +49 30 450 536073; fax: +49 30 450 536940.
E-mail addresses: josefine.radke@charite.de (J. Radke), werner.stenzel@charite.de (W. Stenzel), hans-hilmar.goebel@charite.de (H.H. Goebel).
[1] Tel.: +49 30 450 536004; fax: +49 30 450 536940.
[2] Tel.: +49 30 450 536073; fax: +49 30 450 536940.

http://dx.doi.org/10.1016/j.bbadis.2015.05.007
0925-4439/

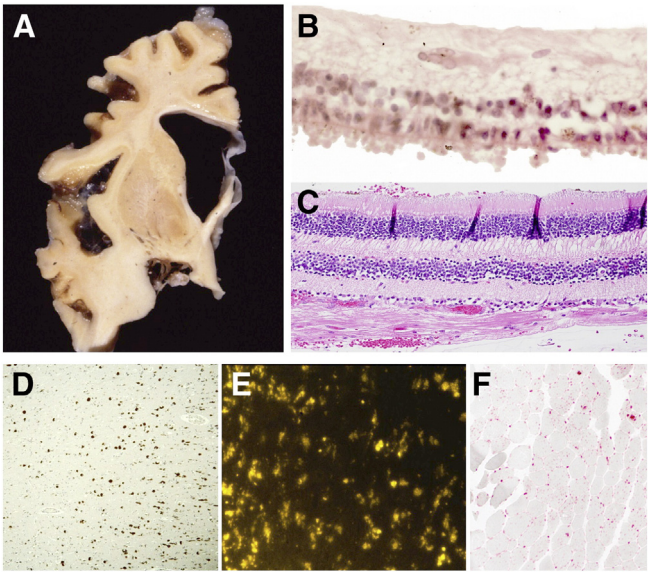

Fig. 1. A: Atrophic cerebral cortex in infantile NCL.
B: Atrophic retina with only remnants of photoreceptor inner segments and severe loss of neurons, CLN2, hematoxylin–eosin (original magnification: 16×).
C: Normal retina, hematoxylin-eosin (original magnification: 20×).
D: Numerous nerve cells, replete with SCMAS-containing lipopigements, CLN5 (original magnification: 16×).
E: Autofluorescence of lipopigments in the cerebral cortex, infantile NCL (original magnification: 40×).
F: Lipopigments in muscle fibers display enhanced histochemical activity of acid phosphatase (original magnification: 20×).

cerebral cortex and cerebellum also means reduced volume of hemispherical white matter, corpus callosum and long tracts, especially the pyramidal tracts which can be observed at the medullary and spinal cord levels. In CLN3, the substantia nigra may appear pale, perhaps owing to impaired formation or loss of neuromelanin in respective neuronal cell bodies. Juvenile and adult NCL brains may display a light brownish or deep yellow hue of grey matter because of excessive formation of lipopigments in nerve cells before their depletion.

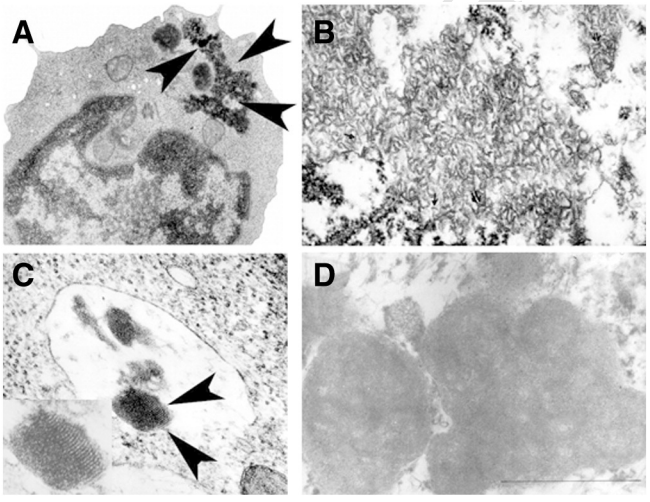

Fig. 2. A: Compact GROD in a lymphocyte, infantile NCL (original magnification: 11000×).
B: Curvilinear profiles, classic late infantile NCL (original magnification: 21600×).
C: Fingerprint profiles in a lysosomal vacuole of a lymphocyte, juvenile NCL (original magnification 21600×); Inset: Fingerprint profiles at high magnification.
D: GROD in vitamin E deficiency (original magnification, 21600×).

Table 1
Brain weight (grams) in CLN forms [34].

NCL type	Brain weight (g)
CLN1	240–450
CLN2	500–700
CLN3	800–1000
CLN4	900–1245
CLN5	450–880
CLN6	455–880
CLN7	590–880

3. Light microscopy

Histological abnormalities are variegated, sometimes contradicting and often incomplete. Three different aspects prevail in the tissues, of which the relationship to each other is not always clear: Firstly, the loss of nerve cells, largely identified by loss of nerve cell bodies rather than processes, is concomitant with subsequent reaction of microglia and astrocytes as well as, secondly, loss of myelin due to axonal degeneration and, thirdly, accumulation of lipopigments. Only the latter phenomenon is assessable in extracerebral organs while any additional convincing histopathology in visceral and other organs, such as skin or skeletal muscle, is largely absent. While with increasing age and survival, especially of patients with juvenile NCL, cardiac symptoms, frequently conduction defects, have become apparent, the few cardiopathological reports emphasize the often heavy formation of NCL lipopigments, with vacuolation and lipopigment formation of fingerprint and curvilinear profiles therein [1], particularly in the conduction system [2,3] and degenerative myocardial pathology, fatty substitution and fibrosis [3,4].

In view of the wealth of modern techniques applicable to the tissues in NCL, the knowledge of lesions within the central nervous system in NCL is still incomplete. As gross pathology and histopathology of the central nervous system are largely based on autopsies — brain biopsies were usually only studied for the diagnostic accumulation of lipopigments and their ultrastructure, rather than structural pathology by light microscopy — major and more complete autopsy reports date back from the premolecular era and even from times when NCL was part of the nosological group of "amaurotic familial idiocy," and such often comprehensive, long, old-time reports are still perfectly valid when retroactively associated with molecular data as provided for the original neuropathological report on classical CLN2, the late infantile form of Jansky–Bielschowsky [5–7]. Since that past history, NCL has been distinguished from other lysosomal diseases affecting the central nervous system by showing autofluorescence of enlarged nerve cells and has also been subclassified in various forms, earlier called infantile, late infantile, juvenile, and adult forms, but now enhanced by recent additional molecular results from CLN1 to CLN14. On the other hand, availability of autopsy tissues and the opportunity to study postmortem CNS by histopathology have been diminished and severely hampered by the dwindling numbers of autopsies. Hence, assessment of precise and complete pathological changes across the entire neuraxis is still patchy.

At autopsy, the cortices of the cerebrum and the cerebellum may variably be depleted of neurons. This emphasizes the fact that autopsy studies usually pertain to the end stage of the disease which is most advanced in the early-childhood forms, although in earlier times NCL patients often died of infectious diseases, e.g., tuberculosis in particular, thereby preventing progression of the NCL to reach its end stage. Loss of neurons is evaluated by recording density and layering of neuronal perikarya. Only a rare neuronal perikaryon may be present in the depleted cortex in CLN1 or infantile NCL. Loss of cortical layers and cytoarchitecture and its most severe form of tissue rarefaction, sponginess of cortical areas, may be encountered [7]. Preferential loss of neurons may only be recognized in NCL forms that, even at autopsy, do not show near-complete neuronal depletion, e.g., in juvenile and adult forms. Also, neuronal depletion and subsequent cortical atrophy

Please cite this article as: J. Radke, et al., Human NCL Neuropathology, Biochim. Biophys. Acta (2015), http://dx.doi.org/10.1016/j.bbadis.2015.05.007