Experimental Gerontology 48 (2013) 1085–1090

Contents lists available at ScienceDirect

Experimental Gerontology

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

Dietary restriction supports peripheral nerve health by enhancing endogenous protein quality control mechanisms

Sooyeon Lee, Lucia Notterpek *

Department of Neuroscience, College of Medicine, McKnight Brain Institute, University of Florida, Gainesville, FL 32610, United States
Department of Neurology, College of Medicine, McKnight Brain Institute, University of Florida, Gainesville, FL 32610, United States

ARTICLE INFO

Article history:
Received 10 September 2012
Received in revised form 7 December 2012
Accepted 17 December 2012
Available online 23 December 2012

Section Editor: Christiaan Leeuwenburgh

Keywords:
Myelin
Demyelination
Proteasome
Protein aggregation
Chaperones
Autophagy
Schwann cell
Glia
Neuropathy

ABSTRACT

The peripheral nervous system (PNS) comprises of an extensive network of connections that convey information between the central nervous system (CNS) and peripheral organs. Long myelinated nerve fibers are particularly susceptible to age-related changes, as maintenance of the insulating glial membrane requires extensive synthesis and processing of many proteins. In rodent models, peripheral demyelination caused by genetic risk factors or by normal aging are attenuated by intermittent fasting (IF) or calorie restriction (CR) supporting a role for dietary intervention in preserving neural function. This review will summarize recent studies examining mechanisms by which life-long CR or extended IF supports peripheral nerve health.

1. Introduction

Peripheral nerves are responsible for bringing information to and from the body and the central nervous system (CNS). The neuronal cell bodies of most peripheral nerves are located within the CNS or clustered in ganglia along the spinal cord, and extend long axons that innervate distant targets such as the skin and muscles. To protect these long projections the axons are wrapped in multiple layers of glial membranes called myelin. In the peripheral nervous system (PNS) Schwann cells provide this wrapping, with each glial cell forming a single internodal myelin segment. Non-myelinating Schwann cells are also present in the PNS and these cells ensheath presynaptic nerve terminals, which are the sites for communication between neurons and target cells.

As seen in other tissues, the cells of the PNS, including Schwann cells, are susceptible to age-associated insults such as DNA damage, oxidative stress, perturbed energy homeostasis, and accumulation of damaged proteins. Furthermore, since it is believed that neurons with long processes are particularly prone to aging-related impairment (Mattson and Magnus, 2006), the extensive lengths of peripheral axons make these cells exceptionally vulnerable. Indeed, aging-related peripheral neuropathy that is not associated with a specific underlying disease, such as diabetes or a known hereditary risk factor, is estimated to affect as much as 22% of people aged 60–74 years, and up to 58% of people aged 85 years and older (Mold et al., 2004). Neurological deficits caused by altered peripheral nerve function include slowed nerve conduction velocity, difficulties with walking and balance, decreased muscle strength and sensory discrimination (Ceballos et al., 1999). It is likely that the combined effects of environmental insults and genetic factors, as well as dietary and life style choices contribute to age-associated changes in peripheral nerve function.

Morphological changes in peripheral nerves with aging are detectable both within the cell bodies of neurons and Schwann cells, as well as along axons. Similar to other tissues, aging in the PNS is associated with increased generation of reactive oxygen species as a by-product of cellular respiration, which can impact DNA, membranes, lipids, and proteins. On light microscopic examination, peripheral nerves of aged rodents show segmental demyelination, myelin thinning, axonal degeneration and regeneration, Schwann cell proliferation and axonal swelling (Grover-Johnson and Spencer, 1981). Within the neuronal cell bodies, decreased efficiency in degradative mechanisms over time is manifested as an accumulation of oxidized proteins and lipids in granules known as lipofuscin (Gray and Woulfe, 2005). These

* Corresponding author at: Department of Neuroscience, McKnight Brain Institute of the University of Florida, 100 Newell Drive, Box 100244, Gainesville, FL 32610-0244, United States. Tel.: +1 352 294 5374; fax: +1 352 846 3854.
E-mail address: notterpek@ufl.edu (L. Notterpek).

0531-5565/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.exger.2012.12.008

structures are pathological signatures of aged cells, also called age pigment, and originate from cumulative inefficiency of multiple protective, repair and clearance mechanisms (Terman and Brunk, 2004). Oxidized lipids and proteins are also detected within Schwann cells of aged peripheral nerves (Opalach et al., 2010) along with other abnormalities such as infolded myelin and collagen pockets (Grover-Johnson and Spencer, 1981). Associated with the gross morphological alterations, subcellular compartments are also affected and include widening of the nodes of Ranvier (Rangaraju et al., 2009), which are critical for efficient signal propagation along axons. Alterations in subcellular axonal cytoskeletal elements and decreased efficiency in axonal transport with aging have also been described (Stokin and Goldstein, 2006), which can impair retrograde transport and deprive the neurons of target-derived trophic support.

The described morphological changes are most pronounced in distal nerve segments which are in close proximity to the contact sites with the target cells, such as skeletal muscle. The junction between a peripheral axon and a skeletal muscle cell is a specialized synapse, the neuromuscular junction (NMJ). In the neuromuscular research community it is well-recognized that aging impacts the morphological and functional aspects of NMJs. Abnormalities in the morphology of NMJs include axon terminal thinning and sprouting, widening of the synaptic cleft and fragmentation of the postsynaptic sites (Valdez et al., 2010). These changes are progressive and begin to occur around 18 months of age in normal mice (Valdez et al., 2010). In an attempt to reverse or slow these events, mice were subjected to one month of exercise (wheel running) beginning at 22 months, which led to the reversal of some synaptic abnormalities. In the same study, the authors examined NMJs from 24 month old life-long calorie restricted animals and found that the morphological alterations at the presynaptic terminals and postsynaptic motor end plate due to aging were significantly blunted (Valdez et al., 2010). The mechanism for these pronounced effects of reduced calorie diet on NMJ structure is unclear, but it could involve improvements in electrophysiological activity. In agreement with this possibility, a recent study demonstrated that maintaining drosophila on a low calorie diet is associated with an increase in synaptic vesicle release by motor neurons and improved vigor, and better motor functions (Rawson et al., 2012). These studies provide evidence for the concept that reducing calorie intake enhances neuronal processes, including synaptic activity (Martin et al., 2006).

While early signs of disease and degeneration in aging are often reported at distal sites of the PNS, Schwann cells along the entire length of the nerve are susceptible to aging-associated deterioration. Starting at the level of gene activation, recent studies have identified transcriptional alterations in aged Schwann cells, with the most significant changes occurring in genes encoding proteins that regulate lipid metabolism or the inflammatory response (Verdier et al., 2012). Indeed, damage to myelin is associated with an elevated immune response, and infiltration of circulating macrophages was observed in nerves of aged, as well as neuropathic rodents (Misko et al., 2002; Opalach et al., 2010). Subcellular pathways in Schwann cells that are affected with age include cell cycle-associated proteins, cytosolic chaperones and components of the autophagy-lysosomal pathway (Rangaraju et al., 2009). These deficiencies affect protein homeostasis and negatively impact the quality of myelin, as detected by morphological as well as biochemical analyses. Alterations in these pathways result in reduced ability of Schwann cells to process functionally important proteins (Fortun et al., 2005), while accumulating damaged or misfolded molecules (Opalach et al., 2010), a feature observed in multiple organisms (Grune et al., 2004).

2. Protein homeostatic mechanisms are critical in myelinating Schwann cells

To prevent the accumulation of misfolded/damaged proteins within the cytosol, mammalian cells utilize three mechanisms: 1) degradation by the ubiquitin-proteasome system (UPS); 2) suppression of aggregate formation by heat shock proteins (HSPs), also known as chaperones, and 3) clearance of abnormal cytosolic contents through autophagy. In agreement with findings in non-neural cells, our studies suggest that these three pathways are similarly critical for Schwann cell function, including myelin formation and maintenance, as well as for supporting axonal biology (Fortun et al., 2005; Opalach et al., 2010; Rangaraju et al., 2009). Moreover, these cellular processes are vulnerable to inefficiencies with aging and disease states. In nerves of aged rats, or mice with genetic defects linked to hereditary neuropathies, we found that impairment in these mechanisms lead to the accumulation of protein aggregates and nerve demyelination (Fortun et al., 2003; Fortun et al., 2006; Rangaraju et al., 2009).

The UPS is the primary mechanism for the removal and degradation of short-lived proteins, as well as for incomplete and/or otherwise damaged polypeptides. Decreased activity in UPS-mediated proteolysis with aging is observed in many cell types including neurons (Keller et al., 2000), where UPS function is necessary for maintaining activity-dependent plasticity through dendritic membrane remodeling (Hamilton et al., 2012). Schwann cells are similarly sensitive to changes in UPS function as this pathway is responsible for maintaining the levels of essential myelin proteins, including myelin basic protein (MBP) and peripheral myelin protein 22 (PMP22) (Akaishi et al., 1996; Notterpek et al., 1999). This is a particular challenge for Schwann cells, as dosage-sensitive, hydrophobic proteins such as PMP22 have high propensity to aggregate, which in turn can decrease the activity of the UPS (Fortun et al., 2005). In addition, the proteasome regulates cytosolic and receptor-protein kinases by terminating their activity through ubiquitination and degradation. Normal myelin formation and myelin repair involve a number of protein kinases including AKT, Erbs, and the Src-family of tyrosine kinases, whose activities are terminated through removal by the proteasome (Lu and Hunter, 2009). Thus, decreased UPS function may lead to irregular kinase activity and alter temporally regulated cell signaling events that are required for myelin maintenance and repair during aging.

During the course of our studies of peripheral nerve biology in normally aged rats and in neuropathic mice, we have detected an increase in protein aggregation and an associated decrease in UPS activity, as revealed by elevated levels of undegraded poly-ubiquitinated (pUb) substrates (Fortun et al., 2005; Fortun et al., 2006; Opalach et al., 2010). These findings are consistent with recent work that revealed a role for proteasome function in preventing age-related metabolic disorders, including obesity and hepatic steatosis (Tomaru et al., 2012). Abnormal cytosolic aggregates can absorb chaperones and other essential cellular proteins, and exert toxicity by the inability of affected cells to respond to subsequent stresses. Therefore, approaches to enhance proteasome function in aged cells and in protein misfolding diseases are of great interest, however are proving to be a challenge. Therefore, we and others have rather focused on preventing the accumulation of undegraded/damaged molecules either by dietary intervention (Opalach et al., 2010; Rangaraju et al., 2009) or by enhancing the function of chaperones and/or the autophagy-lysosomal pathway through pharmacological modulation (Fortun et al., 2007; Madorsky et al., 2009; Rangaraju et al., 2008; Rangaraju et al., 2010).

Because of the ability of HSPs to aid protein processing and prevent misfolding, the chaperone pathway provides a viable approach to slow age-associated changes within cells. Indeed, activation of chaperones has been shown to be protective in a variety of organisms and cell types, and multiple mechanisms by which increased levels of chaperones promote protein quality control have been identified (Hartl et al., 2011). In neurons, increasing HSPs through pharmacological activation is neuroprotective by reducing harmful protein aggregation in diseases such as Huntington's, Parkinson's and Amyotrophic Laterosclerosis (Neef et al., 2010). Chaperones are also