Experimental Neurology 216 (2009) 207–218

Contents lists available at ScienceDirect

Experimental Neurology

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

Neuritic dystrophy and neuronopathy in Akita (*Ins2*^{Akita}) diabetic mouse sympathetic ganglia☆

Robert E. Schmidt *, Karen G. Green, Lisa L. Snipes, Dongyan Feng

Division of Neuropathology, Department of Pathology and Immunology, Washington University School of Medicine, Saint Louis, Missouri, 63110, USA

ARTICLE INFO

Article history:
Received 1 November 2008
Revised 24 November 2008
Accepted 30 November 2008
Available online 10 December 2008

Keywords:
Diabetes
Akita mouse
Neuritic dystrophy
Neuronopathy
Degeneration
Sympathetic ganglia

ABSTRACT

Diabetic autonomic neuropathy is a debilitating, poorly studied complication of diabetes. Our previous studies of non-obese diabetic (NOD) and related mouse models identified rapidly developing, dramatic pathology in prevertebral sympathetic ganglia; however, once diabetic, the mice did not survive for extended periods needed to examine the ability of therapeutic agents to correct established neuropathy. In the current manuscript we show that the Akita (*Ins2*^{Akita}) mouse is a robust model of diabetic sympathetic autonomic neuropathy with unambiguous, spontaneous, rapidly-developing neuropathology which corresponds closely to the characteristic pathology of other rodent models and man. Akita mice diabetic for 2, 4 or 8 months of diabetes progressively developed markedly swollen axons and dendrites ("neuritic dystrophy") in the prevertebral superior mesenteric (SMG) and celiac ganglia (CG). Comparable changes failed to develop in the superior cervical ganglia (SCG) of the Akita mouse or in any ganglia of non-diabetic mice. Morphometric studies demonstrate an overall increase in presynaptic axon terminal cross sectional area, including those without any ultrastructural features of dystrophy. Neurons in Akita mouse prevertebral sympathetic ganglia show an unusual perikaryal alteration characterized by the accumulation of membranous aggregates and minute mitochondria and loss of rough endoplasmic reticulum. These changes result in the loss of a third of neurons in the CG over the course of 8 months of diabetes. The extended survival of diabetic mice and robust pathologic findings provide a clinically relevant paradigm that will facilitate the analysis of novel therapeutic agents on the reversal of autonomic neuropathy.

Introduction

Autonomic neuropathy is an increasingly recognized problem in human diabetes which may result in a variety of complaints involving cardiovascular, genitourinary, sudomotor and alimentary symptoms (Rundles, 1945) or result in subclinical disease. Studies of prevertebral sympathetic ganglia in autopsied diabetic human subjects demonstrate neuroaxonal dystrophy (NAD) (Duchen et al., 1980; Schmidt et al., 1993), an axonopathy represented by marked enlargement of distal axons containing a distinctive admixture of cytoskeletal, autophagic, vesicular and membranous elements. Immunohistochemical studies are consistent with the origin of NAD from other sympathetic neurons (Schmidt, 2002), possibly as the result of intraganglionic sprouting. These axonopathic changes are accompanied by a mild, poorly characterized decrease in neuronal density (Schmidt et al., 1993). Nerve terminal damage is likely to dis- or misconnect ganglionic neurons and, particularly for prevertebral ganglia serving the viscera, contribute to the loss of integrated reflexes. Rat models of diabetic sympathetic autonomic neuropathy show correspondence with human pathology, developing dystrophic axons in prevertebral ganglia (Schmidt, 2002) in the absence of significant neuronal loss (Schmidt, 2001). The fidelity of animal models to the neuropathology of aged and diabetic humans suggests that similar pathogenetic mechanisms may be involved with a comparable response to experimental therapeutic approaches.

Previously we have shown that non-obese diabetic (NOD) mice or streptozotocin-treated NOD/severe combined immune deficient (STZ-Rx NOD/SCID) mice develop dramatic axonal as well as dendritic pathology (thus designated "neuritic dystrophy") within a few weeks of onset of diabetes (Schmidt et al., 2003). Our recent studies with these models (Schmidt et al., 2008b) have shown that there is a continuum of ultrastructural changes in identified presynaptic axon terminals in diabetic mouse sympathetic ganglia which begin with early alterations in synaptic vesicle content and morphology and culminate in the development of anastomosing tubulovesicular membranous aggregates in swollen preterminal axons and formation of multivesicular autophagic bodies. Using STZ-treated NOD/SCID mice, we were able to show that erythropoietin and carbamylated erythropoietin prevented the development of experimental diabetic autonomic neuropathy (Schmidt et al., 2008a). Unfortunately, NOD and STZ-treated NOD/SCID mice, once diabetic, do not survive for

☆ Support: NIH awards R37 DK19645 and AG10299; Juvenile Diabetes Research Foundation Grants 1-2005-1085 and 1-2008-193

* Corresponding author. Fax: +314 362 4096.
E-mail address: reschmidt@wustl.edu (R.E. Schmidt).

0014-4886/$ – see front matter © 2008 Elsevier Inc. All rights reserved.
doi:10.1016/j.expneurol.2008.11.019

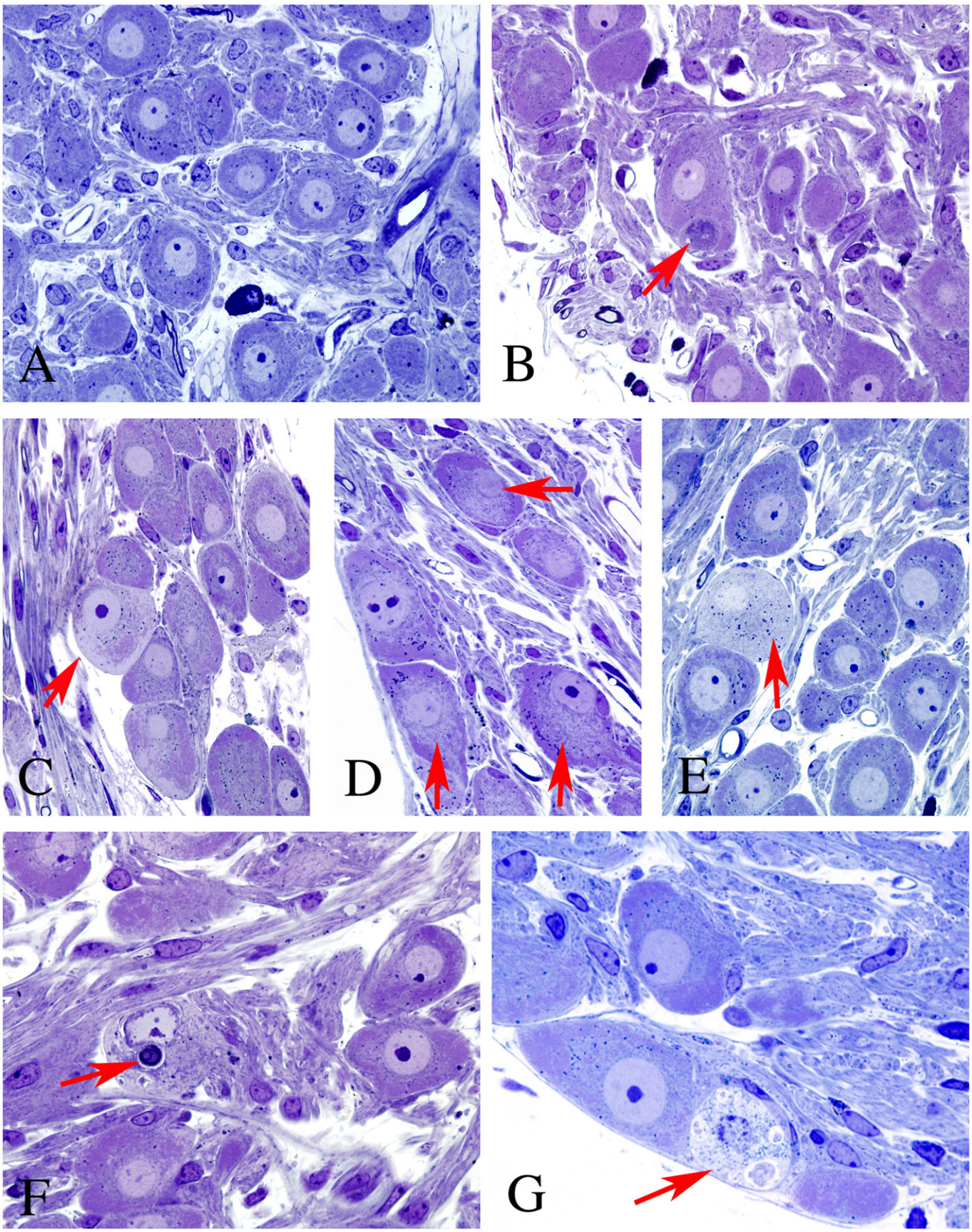

Fig. 1. Light microscopic appearance of the SMG and CG of control and diabetic Akita mice (A) Principal sympathetic neurons surrounded by neuropil with occasional mast cells (control SMG, 5 months of age). (B) A typical dystrophic neurite (arrow) is intimately associated with an adjacent cell body (2 month diabetic CG). (C–E) Neurons show patchy loss of Nissl substance involving the subplasmalemma (arrow, C), central perinuclear area (arrows, D) and entire perikaryon (arrow, E) [C, 4 month diabetic CG; D, 8 month diabetic SMG; E, 8 month diabetic CG]. (F,G) Degenerating neurons may contain large membranous aggregates (arrow, F) or disintegrating cytoplasm (arrow, G) [F, 2 month diabetic CG; G, 4 month diabetic SMG] (original magnification A–G, 500×).