Impaired Cavernous Reinnervation after Penile Nerve Injury in Rats with Features of the Metabolic Syndrome

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ABSTRACT

Introduction. The metabolic syndrome is a cluster of cardiovascular risk factors that predispose toward the development of diseases such as diabetes. Erectile dysfunction (ED) is common in men with metabolic syndrome, but its etiology is poorly understood. Pro-erectile nitrergic nerves innervating penile erectile tissue are also susceptible to mechanical injury during pelvic surgical procedures, which can lead to sexual dysfunction.

Aims. The aims of this article are: (i) to examine erectile function in an experimental model of metabolic syndrome, the phosphoenolpyruvate carboxykinase (PEPCK)-overexpressing rat; and (ii) to study function and cavernous reinnervation after penile nerve crush injury, which permits regeneration, in transgenic rats.

Methods. We analyzed the density of noradrenergic and nitrergic nerves and performed organ bath pharmacology to assess neurogenic activity.

Main Outcome Measures. By analyzing changes in neural structure, function, and pharmacologic responses of cavernous tissue after nerve crush injury, we were able to reveal neurologic deficits in rats with metabolic syndrome.

Results. Animals with features of metabolic syndrome did not develop notable changes in cavernous autonomic nerve density or nerve-evoked smooth muscle activity. However, regeneration of nitrergic nerves after crush injury in transgenic rats was impaired compared with injured controls. This was manifested as a deficit in axon regrowth and responses to axon activation. However, unlike injured controls, injured PEPCK-overexpressing rats did not develop a reduced maximal response to the nitric oxide (NO) donor, sodium nitroprusside. This suggests preserved NO responsiveness in tissues from rats with metabolic syndrome, despite impaired regeneration and return of function.

Conclusions. This study revealed that rats with features of metabolic syndrome display impaired cavernous nerve regeneration after penile nerve injury, but the degree of functional impairment may be attenuated due to reduced plasticity of NO signaling. This reinnervation deficit may be of clinical relevance for understanding why ED persists in some (particularly aged) men after pelvic surgery. **Nangle MR, Proietto J, and Keast JR. Impaired cavernous reinnervation after penile nerve injury in rats with features of the metabolic syndrome. J Sex Med 2009;6: 3032–3044.**

Key Words. Erectile Dysfunction; Nerve Regeneration; Diabetes; Metabolic Syndrome; Corpus Cavernosum

Introduction

Penile erection is a complex hemodynamic process driven by parasympathetic neurons located in the pelvic ganglia, which project to the corpora cavernosum via the penile (cavernous) nerves. Penis-projecting pelvic ganglion neurons are commonly termed "nitrergic," since they release the potent vasodilator, nitric oxide (NO), which is essential for the initiation of the erectile response [1]. By stimulating soluble guanylyl cyclase (sGC), NO increases cyclic guanosine monophosphate (cGMP) levels, which in turn promotes protein kinase G-dependent smooth muscle relaxation [2]. Arterial inflow increases and there is a concomitant reduction in venous outflow as veins are compressed against the tough fibrous sheath (tunica albuginea) encasing the erectile bodies.

Aging and erectile dysfunction (ED) are positively correlated, with half of men aged over 40

years reporting some form of ED [3]. There is a prevailing view that ED should be considered a part of the normal aging process, a manifestation of systemic vascular dysfunction in otherwise asymptomatic men [4,5]. For example, there is high concordance between the causes of ED and cardiovascular disease, together with an elevated prevalence of aging-related disorders such as the metabolic syndrome and insulin resistance, which often precede the development of noninsulin-dependent (type II) diabetes [4–6].

Diabetic men are approximately three times more likely to have ED than nondiabetic men, the relative risk increasing with age and lack of glycemic control [7]. The risk factors for diabetic ED are similar to those for vascular complications in general, as well as those of autonomic neuropathy [8]. In contrast to diabetes, the metabolic syndrome can be considered a "pathway" to disease, rather than an end disease by itself. It is characterized by a group of cardiovascular risk factors, including visceral obesity and insulin resistance, hyperinsulinemia, dysglycemia, dyslipidemia, and hypertension [9]. However, not all features need be present for diagnosis. The most salient risk factors are visceral obesity and insulin resistance [5].

Penis-projecting pelvic ganglion neurons and their axons are susceptible to iatrogenic injury during pelvic surgical procedures, such as prostatectomy or lower bowel resection for the removal of cancerous tumors. Despite the introduction of "nerve-sparing" surgical techniques [10], a common complication is prolonged or even permanent loss of penile erection [11,12]. Inadvertent damage to the neurovascular bundles can be caused by mechanical stretch or severance during prostate retraction and removal, cauterization of blood vessels, or local ischemia and inflammation, leading to axonal degeneration and a loss of neurogenic function [11,12]. Rates of ED vary greatly between clinical studies; however, only 40% of men reported penile erections rigid enough for satisfactory sexual intercourse in a large-scale follow-up study some 5 years after bilateral nerve-sparing operations [13]. Little is known about how aging disorders such as the metabolic syndrome affect postoperative sexual function outcomes after prostatectomy or other pelvic surgical procedures. Of interest, while diabetes is an independent risk factor for ED after prostate surgery, obesity alone is not [14–16].

Neurodegenerative processes undoubtedly contribute to ED in diabetes, but it is not clear whether the mechanism is nerve-specific, or whether it merely reflects the physiological challenge caused by vascular dysfunction [8]. In diabetic rodents, vasoactive therapies can prevent and reverse indices of ED, including reduced relaxation of cavernous tissue to nitrergic nerve stimulation [8,17]. Since the primary etiology of postprostatectomy ED is neurogenic, many studies have attempted to identify mechanisms that prevent neural degeneration or promote regeneration in animal models [18–20]. However, changes in cavernous tissue morphology and function also occur, and these may persist despite successful reinnervation [21,22].

ED has been previously reported in aged animals and experimental models of type II diabetes [23–25]. In the present study, we have asked whether the metabolic syndrome, a "pre-diabetic" state, is sufficient to cause changes in autonomic innervation and function in erectile tissue. Increased hepatic gluconeogenesis is associated with fasting hyperglycemia in type II diabetes [26]. Transgenic mice overexpressing the rate-limiting gluconeogenic enzyme, phosphoenolpyruvate carboxykinase (PEPCK), develop metabolic aberrations akin to those of type II diabetes [27,28]. We have examined cavernosum from rats overexpressing a noninsulin responsive PEPCK transgene, which develop many features of the metabolic syndrome, including visceral obesity and insulin resistance, but do not develop overt hyperglycemia [29,30]. This allowed us to determine the impact of the metabolic syndrome on erectile function without the confounding influence of excess blood glucose, a recognized independent risk factor for ED [8]. Further, by using an animal model with genetic predisposition toward obesity, we did not need to manipulate caloric intake or dietary composition.

Whether the metabolic syndrome affects penile nerve regeneration, or the ability of the penis to respond after nerve injury, is completely unknown. Moreover, understanding how pro-erectile neurons respond to physical trauma in these animals may reveal mechanisms that underlie functional problems associated with changes in metabolic status. Therefore, we have also performed surgical injuries to these nerves in rats with metabolic syndrome and asked whether the responses to neurotrauma differ from injured controls. In particular, we have investigated whether there are any differences in presynaptic (impaired axonal regrowth) or postsynaptic (changes in cavernous tissue responsiveness) mechanisms.