Severe Neuropathy and Erectile Dysfunction After Androgen Deprivation and Cavernous Nerve Injury are Improved with Testosterone Administration

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INTRODUCTION

After radical prostatectomy (RP), many men develop erectile dysfunction (ED) due to injury to the major pelvic ganglia (MPG) and cavernous nerve (CN). The incidence of ED increases when androgen deprivation therapy (ADT) is used to shrink the prostate before RP.

Testosterone (T) is vital to maintenance of neuronal health and function. Without T nerves are vulnerable to injury. Following ADT, recovery of serum T is often slow or incomplete. Although T supplementation in survivors has been considered taboo, T has the potential to both repair nerves and improve erectile function in RP induced nerve injury.

OBJECTIVES

To determine if ADT before CN injury increased the severity of both neuronal damage and ED, and assess if supplementary T restored neuronal health and erectile function.

METHODS

Male Sprague-Dawley rats (12 weeks, n=9/grp)

Erectile Function Assessment

Cavernous nerve (CN) was stimulated at 2V, 4V and 6V at 16Hz, 0.5ms duration, and 30s pulse width for 1 min with 4 min rest. Intracavernosal pressure (ICP) and mean arterial pressure (MAP) were recorded.

Cavernosal Smooth Muscle Function – Myograph

- Contraction
  - Phenylephrine (PE; 10^{-8}M-3x10^{-5}M)
  - Endothelin-1 (ET-1; 10^{-9}M-10^{-7}M)
  - Electrical Field Stimulation (EFS; 16V, 0.5-32Hz)
- Relaxation
  - NO donor (DEA NONOate ; 10^{-9}M-10^{-4}M)
  - Non-adrenergic, non-cholinergic (NANC) relaxation (atropine 10^{-5}M; guanethidine 10^{-6}M; EFS)

MPG Culture and Neurite Outgrowth

Neuritogenesis and prevalence of nerve populations were measured in dissociated culture. MPGs were digested with collagenase and dispase, and plated on poly-l-ornithine and laminin coated coverslips (n=4/grp). After 72 hrs, neurons were fixed and immunofluorescently stained to measure neurite growth, neuron apoptosis and neuron type:

•β-tubulin (**TUJ1**, neuron identification)
•**DAPI** (nuclear marker)
•Terminal deoxynucleotidyl transferase dUTP nick end labeling assay (**TUNEL**, apoptosis)
•Tyrosine hydroxylase (**TH**, sympathetic neurons)
•Neuronal nitric oxide synthase (**nNOS**, nitrergic neurons)

RESULTS

Testosterone increases peak ICP after CAST+BCNI

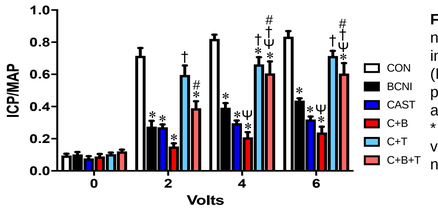


Figure 1. Cavernous nerve stimulated intracavernosal pressure (ICP) to mean arterial pressure (MAP). Data are mean±SEM. p<0.05, * vs. CON, Ψ vs. BCNI, † vs. CAST, # vs. C+B n=9/grp.

Testosterone normalizes penile contraction following CAST or CAST+BCNI

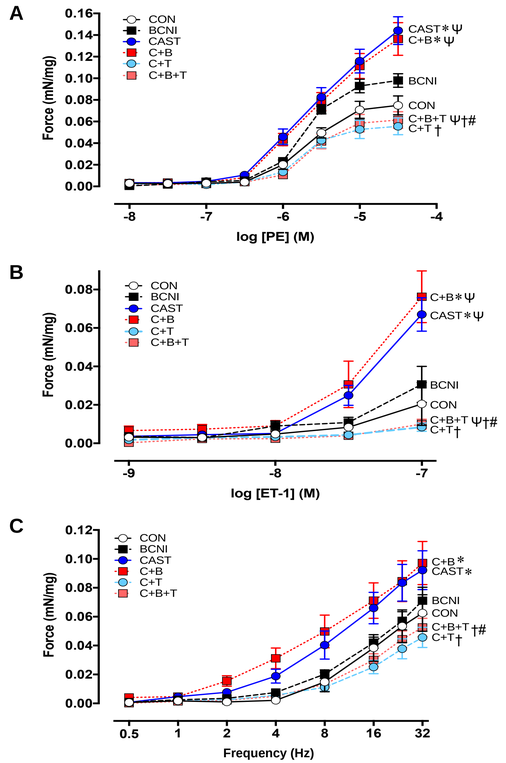


Figure 2. Contractile concentration response curves to **A)** phenylephrine (PE), **B)** endothelin-1 (ET-1) and **C)** electrical field stimulation (EFS) in cavernosal strips. Data are mean±SEM. p<0.05 * vs. CON, Ψ vs. BCNI, † vs. CAST, # vs. C+B, n=5-9/grp.

RESULTS

NO donor and NANC penile relaxation improves with testosterone supplementation

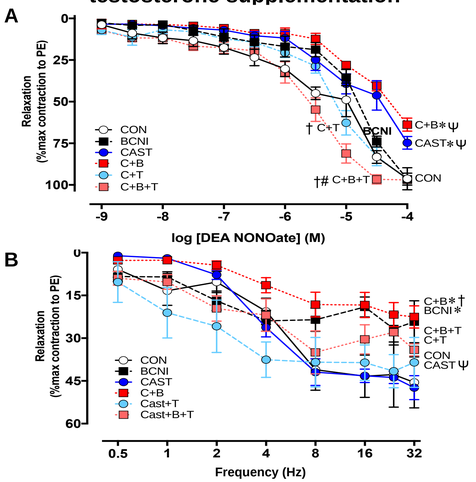


Figure 3. Relaxation concentration response curve to **A)** nitric oxide donor DEA NONOate and **B)** non-adrenergic, non-cholinergic (NANC) stimulation. Data are mean±SEM. p<0.05, * vs. CON, Ψ vs. BCNI, † vs. CAST, # vs. C+B, n=4-9/grp.

Testosterone restores neurite length, but not branching

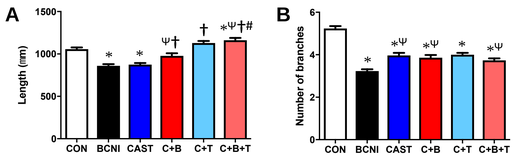


Figure 4. Average neurite **A)** length and **B)** branching after 72hrs in culture. Data are mean±SEM. p<0.05, * vs. CON, Ψ vs. BCNI, † vs. CAST, # vs. C+B n=4/grp.

Neuronal apoptosis is prevented by testosterone

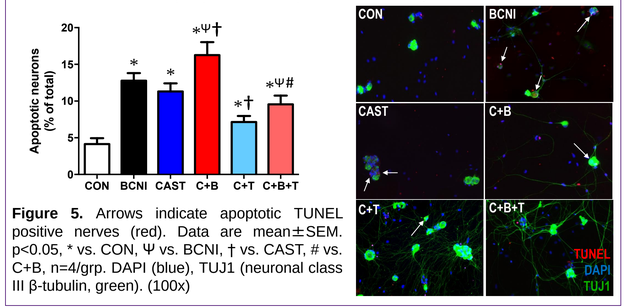
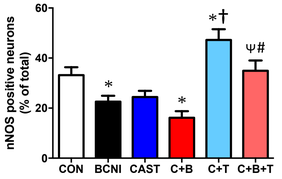


Figure 5. Arrows indicate apoptotic TUNEL positive nerves (red). Data are mean±SEM. p<0.05, * vs. CON, Ψ vs. BCNI, † vs. CAST, # vs. C+B, n=4/grp. DAPI (blue), TUJ1 (neuronal class III β-tubulin, green). (100x)

RESULTS

Nitrergic neurons are markedly reduced with CAST+BCNI and restored with testosterone



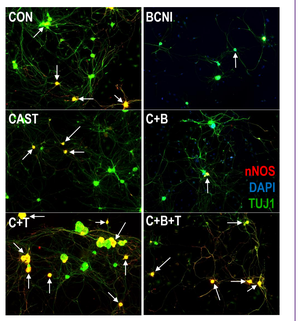
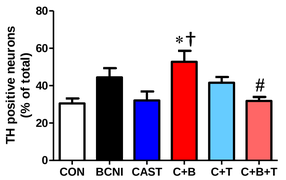


Figure 6. Arrows indicate nerves positive for neuronal nitric oxide synthase (nNOS, red). Data are mean±SEM. p<0.05, * vs. CON, Ψ vs. BCNI, † vs. CAST, # vs. C+B, n=4/grp. DAPI (blue), TUJ1 (neuronal class III β-tubulin, green). (100x)

CAST+BCNI significantly increases sympathetic neurons and normalizes with testosterone therapy



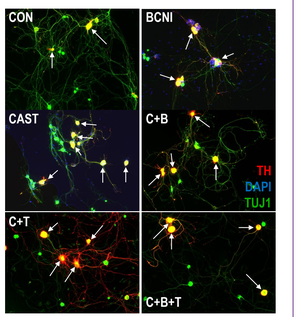


Figure 7. Arrows indicate sympathetic tyrosine hydroxylase (TH) positive nerves (red). Data are mean±SEM. p<0.05, * vs. CON, † vs. CAST, # vs. C+B, n=4/grp. DAPI (blue), TUJ1 (neuronal class III β-tubulin, green). (100x)

CONCLUSIONS

The combination of ADT and BCNI caused severe ED and markedly impaired neuronal health. Low T leaves post-injury nerves highly susceptible to increased apoptosis and Schwann cell activation. T supplementation rescued erections and neuron health after the increased impact of nerve injury during ADT. This work highlights the ability of T to improve erections and pelvic neuron health.

ACKNOWLEDGMENTS

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