

Targeting bladder outlet obstruction from benign prostatic enlargement via the nitric oxide/cGMP pathway?

ANDRÉ REITZ*‡, MICHAEL MÜNTENER†, AXEL HAFERKAMP‡, MARKUS HOHENFELLNER‡ and BRIGITTE SCHURCH*

*Neuro-Urology, Swiss Paraplegic Center, Balgrist University Hospital, and †Department of Urology, University Hospital, Zurich, Switzerland, and

‡Department of Urology, University of Heidelberg, Germany

Accepted for publication 15 January 2005

KEYWORDS

nitric oxide, cyclic GMP, phosphodiesterase, BPH

INTRODUCTION

The high age-related incidence of benign prostatic enlargement (BPE) often associated with BOO and LUTS is of considerable significance for public health. Although rarely life-threatening, BPE has a severe impact on the quality of life of patients. Surgical therapy has been considered the reference standard for treating BPE, but pharmacological therapy is increasingly common, as reflected in the fewer surgical interventions for BPE over the last 20 years. Current medical therapies rely basically on three approaches: (i) α -adrenoceptor antagonists reduce prostatic smooth muscle tone by blocking the effects of noradrenaline released from sympathetic nerve terminals; (ii) 5α -reductase inhibitors reduce prostate volume by blocking the conversion of testosterone to dihydrotestosterone; and (iii) phytotherapeutic drugs seem to be able to improve LUTS by a more or less unknown mechanism.

During the last 25 years nitric oxide (NO) has been recognized as a unique biological signalling molecule involved in numerous physiological processes and body functions. NO was originally known as 'endothelium-derived relaxing factor', which if released by endothelium cells can induce the relaxation of vascular smooth muscle [1]. Furthermore, NO is also involved in immunological responses [2] and acts as an important noncholinergic, nonadrenergic neurotransmitter in both the central and peripheral nervous systems [3,4].

Also within the genitourinary system, NO appears to be an important messenger. Both NO-based mechanisms, the endothelium-derived and the neurogenically mediated

relaxation of cavernosal smooth muscle cells in the penis, have been reported [5,6]. Sildenafil, an inhibitor of phosphodiesterase (PDE) type 5, revolutionized the treatment of erectile dysfunction and highlighted the NO/cGMP pathway as a focus of attention for the urological community to further understand the role of NO in the genitourinary system. Thus, NO-based processes have also been implicated elsewhere in the genitourinary system, e.g. within the detrusor, the urethra and the striated external urethral sphincter. Currently there is growing evidence that the NO/cGMP pathway is involved in regulating the prostatic smooth muscle tone. This review considers the available knowledge about the significance of the NO/cGMP pathway for treating BOO and concomitant LUTS related to BPE. A hypothesis is developed and possible strategies to prove or disprove the suggested approach discussed.

NEUROANATOMY

The human prostate receives autonomic innervation from the pelvic plexus travelling through the cavernosal nerves. Basically, parasympathetic fibres innervate the glandular structures and control prostatic secretion via a cholinergic mechanism; sympathetic nerves innervate the capsule and stroma of the prostate, and control the smooth muscle tone via adrenergic receptors.

In both noradrenergic and non-noradrenergic nerves innervating the human prostate the enzyme NO synthase (NOS) has been found [7]. In a morphological study using immunohistochemistry, NOS activity was found in both the transition and the peripheral zone of the human prostate [8]. NO-containing neurones appeared to originate in the neurovascular bundles later penetrating the prostate capsule and dispersing in the glandular tissue. The level of NOS activity differs in both zones of the prostate, indicating that there is not a

uniform distribution within the gland; NOS activity was higher in the peripheral than in the transitional zone. In both zones NOS was localized to nerve fibres and ganglia within the smooth muscles of the prostatic stroma and the subepithelial plexus, as well as in the glandular epithelium [8].

Another morphological study on human prostatic tissue could not confirm these results and found no difference in nitrinergic innervation density among different parts of the prostate [9]. Using histochemical NADPH-diaphorase staining, NOS immunohistochemistry and ultrastructural NADPH examination, there was a dense nitrinergic innervation of the glandular epithelium, fibromuscular stroma and blood vessels [9].

In both studies NOS-containing nerves were found in close relationship to prostatic smooth muscle cells and it was speculated that NO promotes prostatic smooth muscle relaxation [8,9]. Furthermore, the rich nitrinergic innervation around the prostatic glands found in both morphological studies generated the assumption that NO is also involved in regulating prostatic secretion [8,9].

NO NEUROPHARMACOLOGY

Nerves containing NOS have the capacity to synthesise NO that can serve as a neurotransmitter. NOS catalyses the reaction of the amino acid L-arginine to NO and L-citrulline in the presence of oxygen and NADPH. Currently, three isoforms of the NOS are known, neuronal (first detected in neuronal tissues), endothelial (first found in vascular endothelium) and so-called inducible NOS (first detected in macrophages). After being released from the site of synthesis the NO diffuses freely to the target tissue where the molecule is thought to act as a neurotransmitter on nonadrenergic,

noncholinergic nerves. Throughout the lower urinary tract a nitrinergic innervation has been identified at different densities; almost all NO-induced effects are inhibitory. The NO-mediated responses are thought to act through an intracellular increase in the second messenger cGMP via stimulation of the enzyme guanylate cyclase [10–12]. NO-dependent relaxation of urethral smooth muscle was reported in various species, including rabbit [13] and men [14]. While the nitrinergic innervation and NOS enzyme activity is rich in the urethra, it seems to be sparse within the detrusor muscle. A NO-mediated detrusor smooth muscle relaxation is still controversial, although recent results suggest that detrusor relaxation and contractility might be modulated by NO levels and that NO released from the urothelium may be a mediator of detrusor relaxation during the storage phase of bladder function [15].

NO AND PROSTATIC PHYSIOLOGY

Prostatic specimens from various species including humans have been studied *in vitro*. Takeda *et al.* [16] were the first to find that NO is involved in the control of prostatic smooth muscle function. In their study, NO donors caused a relaxation of human and canine prostatic tissue, with the relaxing effect being significantly greater in the human than in the canine prostate. Heglund *et al.* [17] confirmed these observations in human prostatic tissue and reported a relaxing effect of NO on noradrenaline-contracted prostatic preparations, while the inhibition of NOS effectively counteracted the relaxing effects. Morphological and functional results in that study suggested that neuronally derived NO contributes to the inhibitory control of tension in the prostatic stroma.

Another important issue is the effect of age and prostatic volume on the nitrinergic innervation of the prostatic gland. Aikawa *et al.* [18] studied the effect of age on the endogenous NO-mediated prostatic smooth muscle relaxation and the nitrinergic innervation in the rabbit prostate, and found that both are reduced with ageing. In canine hyperplastic prostates the level of neuronal NO was reduced, suggesting that neuronal NOS expression is down-regulated in the prostate with benign cellular proliferation [19]. In human hyperplastic prostate tissue the nitrinergic innervation was lower than in

normal prostates [9]. Thus, a NO donor had an antiproliferative effect on human hyperplastic prostatic smooth muscle cells [20]. Gradini *et al.* [21] studied prostatic tissue from men with and without hyperplasia for different isoforms of NOS. While neuronal and endothelial NOS were expressed in both normal and hyperplastic glands, inducible NOS was expressed only in hyperplastic glands. The appearance of inducible NOS has been linked to the influence of sex hormones, which have been considered to be involved in the development of prostatic hyperplasia. From this study it was concluded that NO might have a potential role in the pathogenesis of BPE.

These findings may implicate a possible involvement of NO in the pathogenesis of BPE, because a reduced nitrinergic innervation or a relative NO deficiency may increase the tone of the prostatic smooth muscle, which potentially leads to the BOO associated with clinical BPE.

TREATING BPE VIA THE NO/cGMP PATHWAY

From currently available knowledge there is evidence that drugs acting on the NO/cGMP pathway might have a potential role in treating subvesical obstruction caused by BPE. The hypothesis relies on the relaxing effect of NO on prostatic smooth muscle cells that potentially decrease subvesical obstruction and improve both voiding and bothersome LUTS. Considering the pathophysiology of LUTS, the focus has shifted from the prostate to the bladder [22], and recent results suggest that detrusor relaxation and contractility may be modulated by NO levels [15]. NO augmented or released from the urothelium may be a mediator of detrusor relaxation during the storage phase of micturition and therefore may have favourable effects on LUTS.

Basically, two classes of drugs might be relevant for the suggested approach; first, oral NO donors, and second PDE-inhibiting drugs. As heavy bleeding is often reported during prostatic surgery, the gland is considered to have a rich blood supply. After oral intake of sildenafil there was a relevant increase in periurethral blood flow, using colour Doppler TRUS measurements [23], suggesting that oral administration is a feasible approach.

In vitro the NO donor sodium nitroprusside relaxed prostatic smooth muscle strips isolated from the transition zone of the prostate [24]. Currently, oral NO donors are widely used for treating coronary artery disease. Several advantages of NO donors make the further evaluation of their effect on infravesical resistance worthwhile. Many NO donors are well known drugs with good tolerability and a long established safety record, and their variable pharmacokinetic properties could be an advantage. Especially the fast-acting formulations with an onset of action within minutes could allow new treatment strategies with intermittent drug use alone or combined with a classical medical BPE therapy.

BPE and coronary artery disease occur in the same age groups and the coincidence of both problems in elderly people is supposedly high. Klotz *et al.* [25] studied 32 patients who had a urological evaluation before starting nitrate medication for cardiovascular disease. All patients underwent uroflowmetry with a determination of the postvoid residual urine volume, TRUS and PSA screening. According to prostatic symptom scores the authors found that 15 patients had obstructive voiding symptoms, while 17 reported no subjective voiding complaints. At 2 weeks and 3 months after starting nitrate medication the patients were re-evaluated; those who had reported obstructive symptoms before nitrate medication improved significantly as assessed by peak urinary flow rates, symptom scores and postvoid residual urine volume, while asymptomatic patients did not change. PSA values and prostate volumes remained unchanged in both groups. The authors concluded that NO medication influences voiding variables in patients with obstructive BPE and explained this by a potential smooth muscle relaxation within the prostate.

PDEs have been identified in different regions of the human prostate [24,26,27]. *In vitro*, the functional relevance is supported in that the adrenergically induced tension of prostatic smooth muscle strips could be relaxed by inhibitors of PDE-4 and -5 [24]. Furthermore, sildenafil inhibited the proliferation of prostatic hyperplastic tissue [28]. As sildenafil has revolutionized the treatment of erectile dysfunction, many men worldwide take PDE-inhibiting drugs regularly. The prevalence of both erectile dysfunction and LUTS increases with age, and a close relationship of sexual function and voiding function has been

recognized in several studies [29–31]. Medical treatment of lower urinary tract dysfunction is known to influence a patient's sexual function [32] but almost nothing is known of how treating erectile dysfunction with PDE-inhibiting drugs influences voiding dysfunction from BPE. Sairam *et al.* [33] assessed the effect of sildenafil on lower urinary tract function; the coincidence of erectile dysfunction and voiding difficulties associated with BPE in older men is well known, and indeed treatment with sildenafil appeared to improve urinary symptom scores in that study, suggesting a possible role of PDE-inhibiting drugs in treating BPE in the future.

Functional studies *in vivo* assessing the direct effect of NO on the human lower urinary tract are rare. However, after oral administration in healthy humans, a NO donor had a functionally relevant effect on the resting tone and contractile behaviour of the human external urethral sphincter *in vivo* [34]. In a functional study in humans with spinal cord injury, subvesical obstruction caused by detrusor-sphincter dyssynergia was successfully reduced by oral administration of a NO donor [35]. Recently, the immediate influence of systemic NO augmentation on bladder outlet resistance was investigated in healthy men using pressure-flow studies. Relative to the mean average flow rate, the average intravesical pressure during micturition, the ratio of mean average intravesical pressure to mean average flow rate, and the mean intravesical pressure at maximum flow rate, there was a significant reduction in bladder outlet resistance in healthy men within 20 min of sublingual administration of an NO-donor [36].

To confirm the suggested new approach, both urodynamic experiments and chronic clinical studies in men with BPH might be of value. As a first step, uroflowmetry is suggested in the absence and presence of NO. The mean and maximum flow rates, and the postvoid residual volume could be used as key variables. For further evaluating subvesical obstruction, pressure-flow studies are useful to determine the degree of obstruction at baseline and to study any potential improvement in the presence of NO. The combination of high bladder pressures and low flow rates suggests subvesical obstruction, which is in older men most likely associated with BPE. Thus, the degree of subvesical obstruction could be easily

compared in the absence and presence of NO using standardized nomograms.

The safety and efficacy of the suggested approach need to be assessed in clinical trials. For clinical long-term trials NO donors or PDE-inhibiting drugs with long half-lives are preferred, to maintain a sufficient drug level over several hours and to offer the opportunity to use a once-daily administration scheme. As BPE in older men causes obstruction and bothersome LUTS, clinical studies must address both changes in obstruction and LUTS, which can be assessed by frequency-volume charts, voiding diaries and the standardized IPSS questionnaire.

ACKNOWLEDGEMENTS

The authors are grateful to Prof Karl Erik Andersson, University of Lund, Sweden for his excellent comments on the manuscript which improved its quality and understanding.

CONFLICT OF INTEREST

None declared.

REFERENCES

- 1 Furchgott RF, Zawadzki JV. The obligatory role of endothelial cells in the relaxation of arterial smooth muscle by acetylcholine. *Nature* 1980; **288**: 373–6
- 2 Hibbs JB Jr, Vavrin Z, Taintor RR. L-arginine is required for expression of the activated macrophage effector mechanism causing selective metabolic inhibition in target cells. *J Immunol* 1987; **138**: 550–65
- 3 Brecht DS, Snyder SH. Nitric oxide, a novel neuronal messenger. *Neuron* 1992; **8**: 3–11
- 4 Rand MJ. Nitric oxide transmission. Nitric oxide as a mediator of non-adrenergic, non-cholinergic neuro-effector transmission. *Clin Exp Pharmacol Physiol* 1992; **19**: 147–69
- 5 Azadzi KM, Kim N, Brown ML, Goldstein I, Cohen RA, Saenz de Tejada I. Endothelium-derived nitric oxide and cyclooxygenase products modulate corpus cavernosum smooth muscle tone. *J Urol* 1992; **147**: 220–5
- 6 Burnett AL, Lowenstein CJ, Brecht DS, Chang TS, Snyder SH. Nitric oxide. A physiologic mediator of penile erection. *Science* 1992; **257**: 401–3
- 7 Jen PY, Dixon JS, Gearhart JP, Gosling JA. Nitric oxide synthase and tyrosine hydroxylase are colocalized in nerves supplying the postnatal human male genitourinary organs. *J Urol* 1996; **155**: 1117–21
- 8 Burnett AL, Maguire MP, Chamness SL *et al.* Characterization and localization of nitric oxide synthase in the human prostate. *Urology* 1995; **45**: 435–9
- 9 Bloch W, Klotz T, Loch C, Schmidt G, Engelmann U, Addicks K. Distribution of nitric oxide synthase implies a regulation of circulation, smooth muscle tone, and secretory function in the human prostate by nitric oxide. *Prostate* 1997; **33**: 1–8
- 10 Persson K, Alm P, Johansson K, Larsson B, Andersson KE. Co-existence of nitric, peptidergic and acetylcholine esterase-positive nerves in the pig lower urinary tract. *J Auton Nerv Syst* 1995; **52**: 225–36
- 11 Dokita S, Smith SD, Nishimoto T, Wheeler MA, Weiss RM. Involvement of nitric oxide and cyclic GMP in rabbit urethral relaxation. *Eur J Pharmacol* 1994; **266**: 269–75
- 12 Morita T, Tsujii T, Dokita S. Regional difference in functional roles of cAMP and cGMP in lower urinary tract smooth muscle contractility. *Urol Int* 1992; **49**: 191–5
- 13 Andersson KE, Garcia Pascual A, Persson K, Forman A, Tottrup A. Electrically-induced, nerve-mediated relaxation of rabbit urethra involves nitric oxide. *J Urol* 1992; **147**: 253–9
- 14 Ehren I, Iversen H, Jansson O, Adolffson J, Wiklund NP. Localization of nitric oxide synthase activity in the human lower urinary tract and its correlation with neuroeffector responses. *Urology* 1994; **44**: 683–7
- 15 Theobald RJ Jr. Differing effects of NG-monomethyl L-arginine and 7-nitroindazole on detrusor activity. *NeuroUrol Urodyn* 2003; **22**: 62–9
- 16 Takeda M, Tang R, Shapiro E, Burnett AL, Lepor H. Effects of nitric oxide on human and canine prostates. *Urology* 1995; **45**: 440–6
- 17 Hedlund P, Ekstrom P, Larsson B, Alm P, Andersson KE. Heme oxygenase and NO-synthase in the human prostate – relation to adrenergic, cholinergic and peptide-containing nerves. *J Auton Nerv Syst* 1997; **63**: 115–26

- 18 Aikawa K, Yokota T, Okamura H, Yamaguchi O. Endogenous nitric oxide-mediated relaxation and nitrinergic innervation in the rabbit prostate: the changes with aging. *Prostate* 2001; **48**: 40–6
- 19 Crone JK, Burnett AL, Chamness SL, Strandberg JD, Chang TS. Neuronal nitric oxide synthase in the canine prostate. aging, sex steroid, and pathology correlations. *J Androl* 1998; **19**: 358–64
- 20 Guh JH, Hwang TL, Ko FN, Chueh SC, Lai MK, Teng CM. Antiproliferative effect in human prostatic smooth muscle cells by nitric oxide donor. *Mol Pharmacol* 1998; **53**: 467–74
- 21 Gradini R, Realacci M, Ginepri A *et al.* Nitric oxide synthases in normal and benign hyperplastic human prostate: immunohistochemistry and molecular biology. *J Pathol* 1999; **189**: 224–9
- 22 Siroky MB. Lower urinary tract symptoms: shifting our focus from the prostate to the bladder. *J Urol* 2004; **172**: 1237–8
- 23 Pinggera A, Schuster F, Frauscher G, Bartsch H. Sildenafil citrate causes a 3 fold increase in periurethral prostatic blood flow. *J Urol* 2004; **171**: A1348
- 24 Uckert S, Kuthe A, Jonas U, Stief CG. Characterization and functional relevance of cyclic nucleotide phosphodiesterase isoenzymes of the human prostate. *J Urol* 2001; **166**: 2484–90
- 25 Klotz T, Mathers MJ, Bloch W, Noyal W, Engelmann U. Nitric oxide based influence of nitrates on micturition in patients with benign prostatic hyperplasia. *Int Urol Nephrol* 1999; **31**: 335–41
- 26 Stacey P, Rulten S, Dapling A, Phillips SC. Molecular cloning and expression of human cGMP-binding cGMP-specific phosphodiesterase (PDE5). *Biochem Biophys Res Commun* 1998; **247**: 249–54
- 27 Fawcett L, Baxendale R, Stacey P *et al.* Molecular cloning and characterization of a distinct human phosphodiesterase gene family PDE11a. *Proc Natl Acad Sci USA* 2000; **97**: 3702–7
- 28 Adolfsson PI, Ahlstrand C, Varenhorst E, Svensson SP. Lysophosphatidic acid stimulates proliferation of cultured smooth muscle cells from human BPH tissue: sildenafil and papaverin generate inhibition. *Prostate* 2002; **51**: 50–8
- 29 Rosen R, Altwein J, Boyle P *et al.* Lower urinary tract symptoms and male sexual dysfunction: the multinational survey of the aging male (MSAM-7). *Eur Urol* 2003; **44**: 637–49
- 30 Schou J, Holm NR, Meyhoff HH. Sexual function in patients with symptomatic benign prostatic hyperplasia. *Scand J Urol Nephrol Suppl* 1996; **179**: 119–22
- 31 Namasivayam S, Minhas S, Brooke J, Joyce AD, Prescott S, Eardley I. The evaluation of sexual function in men presenting with symptomatic benign prostatic hyperplasia. *Br J Urol* 1998; **82**: 842–6
- 32 Clifford GM, Farmer RD. Medical therapy for benign prostatic hyperplasia. A review of the literature. *Eur Urol* 2000; **38**: 2–19
- 33 Sairam K, Kulinskaya E, McNicholas TA, Boustead GB, Hanbury DC. Sildenafil influences lower urinary tract symptoms. *BJU Int* 2002; **90**: 836–9
- 34 Reitz A, Bretscher S, Knapp PA, Muntener M, Wefer B, Schurch B. The effect of nitric oxide on the resting tone and the contractile behaviour of the external urethral sphincter: a functional urodynamic study in healthy humans. *Eur Urol* 2004; **45**: 367–73
- 35 Reitz A, Knapp PA, Muntener M, Schurch B. Oral nitric oxide donors: a new pharmacological approach to detrusor-sphincter dyssynergia in spinal cord injured patients? *Eur Urol* 2004; **45**: 516–20
- 36 Muntener M, Schurch B, Wefer B, Hauri D, Reitz A. Systemic nitric oxide augmentation leads to a rapid decrease of the bladder outlet resistance in healthy men. *Eur Urol* 2005; **45** (Suppl): Abstract

Correspondence: André Reitz, Neuro-Urology, Swiss Paraplegic Center, Balgrist University Hospital, Forchstrasse 340, 8008 Zurich, Switzerland.
e-mail: areitz@balgrist.unizh.ch

Abbreviations: BPE, benign prostatic enlargement; NO(S), nitric oxide (synthase); PDE, phosphodiesterase.